Table of Contents

<u>2023.11.05 Nucleic Acids - DNA TSP 70</u> 1
2023.11.07 Bacteriophage
2023.11.07 Precipitate Comparisons
2023.11.12 Decanted - Vinyl Production2
2023.11.14 TSP Culture Variants
2023.11.14 TSP culture variation
2023.11.15 TSP Culture Variations
2023.11.16 (Borax Polymer) & 100.6
2023.11.17 Solubility Methylation
2023.11.18 Borax polymer solubility
2023.11.19 Project Planning
2023.11.20 UV NIR TSP 70 Nucleic Acid25
<u>2023.11.21 Nucleic Acids (?) - DNA- PVA</u> 27
2023.11.22 Solubility-Health-Blood(Syn)31
2023.11.23 Blood(Syn)-Polymer Compare37
<u>2023.11.24 Abstracts AI - NIR Synth</u> 43
2023.11.25 Human Synth Blood Compare46
2023.11.26 Blood Synth Vinyl MID IR
2023.11.27 Research Paper:Blood Compare54
2023.11.28 Research Paper Video Audio57
2023.11.30 Another Mitigation Prospect60
2023.12.02 Abstracts 2022 Prepared66
2023.12.04 Media Summaries - Vinyl67
2023.12.05 Blood Clot Progession Study79
2023.12.06 Functional Group Worksheet81
2023.12.07 Functional Group Worksheet83
2023.12.08 Functional Group Worksheet95
2023.12.09 Functional Groups DNA99
2023.12.10 Functional Group Worksheet101
2023.12.11 Functional Group Worksheet104
2023.12.12 Research Paper - Regroup107
2023.12.13 Alkenes - Polymerization117
2023.12.14 Mitigation -Methylation

Table of Contents

2023.12.15 Paper Complete	126
2023.12.16 Bacteriophage Inquiry	128
2023.12.17 Enzyme - Bacteriophage - DNA	132
2023.12.18 DNA - Bacteriophage Inquiry	142
2023.12.19 DNA - Bacteriophage Inquiry	151
2023.12.23 DNA - Bacteriophage Inquiry	155
2023.12.24 Video Up - Regroup - Phage	158
<u>2023.12.25 Christmas - On We Go</u>	164
2023.12.26 Five TSP Separations	166
2023.12.27 Amines Amides VinyL	169
2023.12.28 TSP 070 Separations LC DNA	172
<u>2023.12.29 TSP 070 Acetone Study</u>	174
2023.12.30 TSP 070 Acetone-Phage-ImageDB	177
2023.12.31 Afflicted Skin	179

2023.11.05 Nucleic Acids - DNA TSP 70

Nucleotides Nucleic Acids DNA?

2023.11.07 Bacteriophage

Bacteriophage issue arises today.

2023.11.07 Precipitate Comparisons

Compared NIR plots Native Preciptate 20 Days (70.5) vs Titrated Precip (70.2)

70.5 > 70.2: 1195(Methyl) > 933(Methylene) 1506 Aromatic Amine, NH Amide Protein > 1515 Polyamide 1582 Aklyl Alcohol > 1611-1654 Vinyl, Polyamide, Acrylate

2023.11.12 Decanted - Vinyl Production?

From UV analysis: Tyrosine Protein

Strong surface polymer forming. Grid like structure under slide. CDB polymer grid matrix.

No definite VIS NIR. This does say that the alkyl alcohol is no longer in place here. However, what is occuring from both UV and NIR is:

Protein Tyrosine

From NIR comparison of 100.5 and 100.6. There is a shift from the amide/protein nature of 100.5 (titrate decant) to the vinyl region of 1643 in 100.6. Protein/tyrosine being identified via UV at 272 peak absorbance in 100.5.

Polymer microspheres 0.25 microns.

11/13/23 (100.5 started on 11/08/23) Therefore, 100.5 (decant, titration) is the culture showing the most visible change thus far in the vitals. Appearance of culture has changed from clear to cloudy. pH has steadily decreased. Strong signs of polymer formation under scope.

100.5 NIR major feature shows a shift of absorbance from the 1610 nm region to the 1510 nm regeion. It also looks like 100.5 shows a relative increase in the 1420 nm region.

1610 is the vinyl area 1510 is the amide protein area 1420 could be in the aromatic region, either ArCH or ArOH.

Nov 14 2023

It seems that the emphasis is 105.6 is the production either a protein or vinyl or both. It may be that they are one and the same thing.

Notes from Lab Log 11/14/23 on the observation of TSP 100.2 after 6 days incubation.

Too early to say the consequences of what is happening here. But we definitely have synth cell production here. This means the time to do so has apparently been shortened from 14-20 days to 6 days. 100.2 was established on 11/08/23. We can definitely say the L. enzyme is having an impact upon the culture, whether it will be for better or worse hopefully we shall learn to the better. What does seem the case is that the cells are of the small variety (~4 microns diameter) and that the color is apparently a grayish color.

I have added .08 gms of L.enzyme now to TSP 100.3 to attempt an acceleration of synth cell growth in that culture also. TSP 100.4 serving as the control shows no such surface growth at this time, so the enzyme definitely seems to be decreasing the synth cell production time.

Alkyl Alcohol Protein Shift from 275nm to 265 nm UV UV Structural Shifts TDS Shifts under observation

Notes from Lab Log 11/14/23 on the observation of TSP 100.2 after 6 days incubation.

TSP 100.2 Enzyme Addition TSP 100.2

Too early to say the consequences of what is happening here. But we definitely have synth cell production here. This means the time to do so has apparently been shortened from 14-20 days to 6 days. 100.2 was established on 11/08/23. We can definitely say the L. enzyme is having an impact upon the culture, whether it will be for better or worse hopefully we shall learn to the better. What does seem the case is that the cells are of the small variety (~4 microns diameter) and that the color is apparently a grayish color.

I have added .08 gms of L.enzyme now to TSP 100.3 to attempt an acceleration of that culture also. TSP 100.4 serving as the control shows no such surface growth at this time, so the enzyme definitely seems to be decreasing the production time.

TSP 100.6 Titration of TSP 100.0

Acidified the precipitate with H2SO4 for UV work. Worked very well. Alkyl alcohol identified. There does seem to be some activity at 1080. This corresponds to an alcohol also, of form RCOH.

915 Methyl 1643 Vinyl

From NIR comparison of 100.5 and 100.6. There is a shift from the amide/protein nature of 100.5 at 1523 nm (titrate decant) to this vinyl region of 1643 in 100.6.

2023.11.12 Titration of TSP 100.0 Acidified the precipitate with H2SO4 for UV work. Worked very well. Alkyl alcohol identified. There does seem to be some activity at 1080. This corresponds to an alcohol also, of form RCOH.

915 Methyl 1643 Vinyl

From NIR comparison of 100.5 and 100.6. There is a shift from the amide/protein nature of 100.5 at 1523 nm (titrate decant) to this vinyl region of 1643 in 100.6.

2023.11.14 TSP Culture Variants

I am in process of switching over to a digitial lab notebook as primary. Some paper notes will be carried in parallel but digital lab may easily become primary. Sortable by date, subject, notes, etc. Date entries and subject entries will both be made.

Let's mention some of the complexity that is taking place here. A single pH change introduced into the TSP culture allows for 7 different visual variants to develop, and this is only a start.

1. Mature native culture (2 weeks incubation) - top layer where polymer-synth cells (blood) develops 2. Mature native culture (2 weeks incubation) - middle layer comprising bulk of solution - to be systematically analyzed with controls. Preliminary work has already been done and is recorded prior to complexity of pH and temperature control became apparent over a couple of months of work.

3. Mature native culture (2 weeks incubation) - bottom layer, white precipitate. Also to be systematically studied with.

4. Native TSP culture, freshly developed over approx. 3 day period. ph \sim 3.8.

5. Titrated fresh TSP culture to raise pH from native ~3.8 to ~7.23 separates TSP into two components:

5-1: Transparent decantable solution of amber color. Forms bulk of solution

5-2: Precipitate: greyish in color. Upon centrifuge, it will be found that this grey precipitate actually separates into two separate layer: white and grey.

5-2-1 Grey precipitate (less dense)

5-2-2 White precipitate (more dense)

This brings us up to 7 visual variants with a single pH change introduced. No temperature control, room temperature on all.

Next, we have another variant that develops in the decanted transparent solution, this is number 8:

8. In the decanted titrate, after 2-3 days at room temperature, a polymer forms within the solution and turns the solution cloudy. All NIR signs are saying that this is a vinyl based polymer. Microscopic examination of this agrees, as "microspheres" of size approx. 250 nm are now visible under the scope with improved techniques. Vinyl microsphere production of this size is a standard commercial operation.

All of the above occurs as a result of a single pH modification with no temperature control. This lets us know what we are up against and why dozens of variants in form are presenting themselves under insufficiently controlled and monitored conditions.

Let's monitor culture vitals - in spreadsheet.

As a prelude to this, here is another observation developing involving 3 culture variations with enzyme study:

100.2 This culture applies .04 gm L. enzyme to 150 ml of DILUTE TSP. 100.3 This culture applies .04 gm L. enzyme to 150 ml of CONCENTRATED(Evaporated) TSP. 100.4 This culture is of CONCENTRATED TSP only.

The ratio between the dilute and concentrated TSP is approximately a factor of 3, i.e., the reduction of approx. 6000 ml to approx. 2000 ml. This means the impact of the enzyme in the dilute TSP is expected to be greater than that of the concentrate TSP. This is where an observation comes in that supports this. TSP 100.2 was to be discarded in favor of TSP 100.3, but it is found that TSP 100.2 becomes valuable within this observation. It will therefore be maintained to some degree.

The observation is that there is a color change between the three cultures. 100.2 is lightest in color with a whitish tint, 100.3 is intermediate with a slight white tint, and 100.4 is the darkest solution with an amber tint.

I also am seeing some surface development on 100.2. This will have to be examined under the scope. It will present a risk to the benefits surmised from the L. enzyme.

The significance of this is that this is one of the many ways it can be determined that the L. enzyme is having an impact upon the culture. Other methods of determination include:

"Vitals" monitoring (pH, TDS (total dissolved solids), and PPT (parts per thousand (actually specific gravity))
 UV and NIR monitoring.

It is consistently being established that the L. enzyme is having an impact upon the TSP culture from the onset. This is desirable, as interruption of protein development is a primary goal here.

Now to vitals in spreadsheet.

I have developed a model to establish the "interest level" in a culture. It is too complex to describe here but is based upon some rather sophisticated probability modeling. Maybe another time, but here are my scores:

TSP 100.8 95% Decant TSP 100.9 92% Decant TSP 100.5 62% Decant TSP 100.0 17% TSP 100.4 23% TSP 100.3 12% TSP 100.2 38% Enzyme study

It took a great deal of work to develop this model but it is worth it to me. All encoded into the culture spreadsheet.

This tells me interest level is in the list of

100.8 decant 100.9 decant 100.5 decant 100.2 decant

It is very demanding and time consuming to monitor these cultures so it helps tremendously to whittle down the playing field.

One of the first things learned from this is that it will be of higher interest to boost the enzyme concentration in 100.3 from \sim .04 gms to \sim .12 gms to anticipate similar results in the concentrated TSP enzyme trial.

Now bear in mind that 100.2 is now showing a significant surface layer development. This could be serious troubling, don't know, but let's see under the scope.

-

Too early to say the consequences of what is happening here. But we definitely have synth cell production here. This means the time to do so has apparently been shortened from 14-20 days to 6 days. 100.2 was established on 11/08/23. We can definitely say the L. enzyme is having an impact upon the culture, whether it will be for better or worse hopefully we shall learn to the better. What does seem the case is that the cells are of the small variety (~4 microns diameter) and that the color is apparently a grayish color.

I have added .08 gms of L.enzyme now to TSP 100.3 to attempt an acceleration of that culture also. TSP 100.4 serving as the control shows no such surface growth at this time, so the enzyme definitely seems to be decreasing the production time.

A question: Is development within TSP 100.5, 100.9 etc vinyl or protein, or BOTH? NIR shows both with some transitions taking place between the solution form of the titrate and the precipitate form of the titrate. Not sure yet which is dominant in which. Keep reviewing and collecting UV, VIS-NIR and NIR until you sort this out. Uniformity under scope indicated microspheres dominanting at 0.25 spherical diameter, but I think UV and NIR say both protein and vinyl are active.

There are definitely bio vinyl polymers. A hot topic is biodegradable vinyl polymers for use in medicine, i.e., the body. So we could easily have a combination here.

We have:

"The optical absorption measurements for poly(vinyl alcohol) evidenced bands in the UV region (see Figure 2) at 223, 282 and 327 nm assigned to n?p* transition, due to the presence of the C=O group and p?p* transition, respectively which arise from unsaturated bonds, C=O and/or C=C, which are present in the tail-head of the polymer."

https://www.researchgate.net/figure/UV-absorption-spectrum-of-the-polyvinyl-alcohol fig2 289203733

I have a spectrum that shares a great deal in the general sense of what is shown. I am out of range for the 223 but it certainly is following the profile. I have absorbance at ~338 and 282 general peak with derivative determination.

Also we have:

"Spectral examination of the solvolysis products of vinyl acetate and 1-methoxyvinyl ester copolymers shows unequivocally that the absorption bands in PVA at 225, 280, and 330 mµ are related to carbonyl-containing structures."

https://onlinelibrary.wiley.com/doi/abs/10.1002/pol.1963.100010412



2023.11.14 TSP culture variation

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2023.11.15 TSP Culture Variations

The cultures are very active and interesting. 100.7 has come to life, this is the petri dish series of the titration decant, set up on Nov 12, 72 hrs. The purpose of this series is to determine if surface area exposure is a factor on the polymer formation from the decant. It may note be as much a factor as first surmised as the polymer seems to uniformly distribute throughout the decant over time.

One thing interesting is that four cultures, supposedly identical were poured from the same container. But there is variability between the four, with one dish showing much higher surface layer development. This is an example of the high level of variation that can occur with cultures with unknown subtle factors at play.

The next observation concerns TSP 100.0, the primary active culture of main volume (\sim 1500 ml). This culture is now already producing the synth cell-polymer layer in large quantity on the surface. This was a breakthrough development and it has now been replicated on a large scale reliably. It was thought this process required a minimum of 2 weeks up to 3 weeks, but it has now taken place in 7 days, culture started on Nov 08 2023.

In addition, the three layer separation is also beginning to form. The three layers are an important feature of the mature culture. The three layers are:

- 1. Top: synth cell polymer layer, whitish in color.
- 2. Middle: bulk of volume, amber color solution.
- 3. Bottom: precipitate layer, whitish to light grey in color.

These layer are not yet well defined in TSP 100, but there separation is perceptible. The solution remains almost entirely grey. Oxidation will eventually turn the main solution amber.

I have my eye on a mid IR instrument, but unfortunately is just not likely to be financially feasible. I have a rather extensive library of work available from previous years. If I am unable to acquire (seems likely) there is another route that might be able to make headway. If I can make tentative identification of a material or functional group and can then acquire a sample of that material, then UV and NIR comparison may well be sufficient to establish the confirmation level desired.

I am already very close to substantiating the claim of polyvinyl alcohol. The factors are:

1. UV analysis shows a general match

2. MID IR - my plot of "CDB Protein Complex Sep 06 2015" looks to be one remarkable match to that from the SDBS database.

3. NIR looks like it is going to match also, but I do not have a reference yet.

So to push this confirmation level further:

1. Work hard to find a NIR plot of PVA - needs to be 900 - 1700 nm preferably.

2. ACQUIRE A SAMPLE of PVA and see if you can get a match. Then run what you have : UV, NIR and see what you can do.

SOLUBILITY COMMENTS:

This version of PVA has a solubility that depends upon pH it would seem. PVA is stated to be water soluble (pH implied here of \sim 7). Our proposed PVA is indeed soluble in water. But the synth cell-polymer is not, it is soluble in strong acid and possibly acetone, but not in water. These our important characteristics to keep in mind. Also our precipitate from the titration may well be a more pure form of PVA. Keep working on it...

I found a source for PVA and it should be on the way. Will help a lot. This is a good alternative to access to a MID IR instrument if I can make a good tentative identification. You have never had cultures at this level of specificity before.

Let's look for PVA reactions in the meantime.

There is an interaction between PVA, boric acid and iodine. Working on acquiring PVA and boric acid.

"Interaction of Poly(viny1 Alcohol), Boric Acid, and Iodine Though it has been known for some time that poly(viny1 alcohol)(PVAL), iodine, and boric acid form a c~mplex,'~ no quantitative estimation of the composition or the stability values of the complex has been reported. In this communication, we are giving for

the first time results regarding the composition, the molar extinction coefficient, and the stability constant of the complex. EXPERIMENTAL PVAL solution was prepared by weighing it exactly and dropping it into a measured quantity of doubly distilled water, raising the temperature to 85OC, and then allowing it to cool. Iodine and boric acid solutions were prepared by standard methods.6 The molecular weight of PVAL was determined by a viscosity method.7 The optical density (log Zo/Z) and the spectra were recorded by using a Specord-UV-VIS spectrophotometer. RESULTS AND DISCUSSION Absorption maxima of the PVAL-iodine complex formed in the presence and in the absence of boric acid has been reported'e2 in arbitrary units proportional to the absorbance. Zwick3 reported that the absorption maximum is shifted continuously from 580 to 700 nm by increasing the con- centration of boric acid at a constant concentration of iodine. But we observe that at a constant concentration of iodine, the intensity of color increases but the absorption maximum remains only at 14,500 cm-' (around 680 nm) and does not change with boric acid concentration (Fig. 1)"

https://www.researchgate.net/publication/250427602_Interaction_of_polyvinyl_alcohol_boric_acid_and_iodine

Good news, PVA and borax alone have a reaction. This is the "slime" affair.

I have produced a reaction with TSP 100.5 and borax. It is creating what appears to be a brittle polymer. Polymer formation by reaction has been achieved.

All signs are that this polymer shares some of the fundamental properties of PVA, but what version of polymer is formed is not certain. Brittle nature is important vs a flexible polymer. My take right now might be an acetate form. I have it under the scope photographed at 300x. It appears to have constructed a crystalline form. Application of heat indicates it may melt fairly easily. It appears quite pure.

Another important observation. The amount of enzyme added to TSP 100.2 and 100.3 completely changes how the culture responds. 100.2 has .04gms added to ~ 150ml of TSP 100.0. 100.3 has .12 grams added to ~ 150ml of TSP 100.0. The results ARE ENTIRELY DIFFERENT. TSP 100.2 now has a full fledged synth cell-polymer matrix that has formed on the top within 24 hrs. TSP 100.3 is now CLARIFIED & TRANSPARENT with no surface growth visible. This indicates the amount of enzyme has made a tremendous difference in the result. A greater concentration of enzyme appears to be more destructive to the culture growth process. The difficult factor to unwind is that TSP 100.3 was approx. 3 times more concentrated than 100.1 so I suspected they would balance each other out and act mostly the same. Not so, they have reacted completely differently. One case looks very unfavorable and the other quite favorable and promising. The control shows the usual amber solution color.

2023.11.16 (Borax Polymer) & 100.6

I am looking at 100.8. This is the larger volume decant sample prepared for additional pl testing. It will now also be used and given equal priority to polymerized protein production (presumed identity). It is following the same progression of 100.5 (also a titration decant) with:

1. Increasing cloudiness

2. Lowering of pH

3. Increased TDS

4. Increased PPT

as recorded on spreadsheet during vitals monitor today.

We notice that the "vitals" in no way capture the whole story. They are only a helpful indicator but they can miss very important events. An example is what is happening between 100.2, 100.3 and 100.4

100.4 is TSP control 100.2 is .04 gms enzyme in dilute TSP 100.3 is .12 gms enzyme in conc. TSP

The vitals indicate possible significance with TSP 100.2 which is true and of interest since it is forming a synth cell-polymer surface layer even though lighter concentration of enzyme has been added.

However, 100.3 by vitals shows nothing of particular interest. Visually, however, it is more than important, especially as it compares to 100.2 The only difference should be an increased concentration of enzyme to 0.12 gms. The culture is TRANSPARENT with NO synth cell - polymer surface layer. This is a huge difference, but the vitals miss it and only observation detects it. So stay alert is the lesson....

Now let's review the NIR spectrum of the borax-decant reaction from yesterday. It was presumed that it might have close ties with a polyvinyl compound. We did notice and are aware, however, that the borax reaction product was neither water soluble or flexible.

Note: (1) Weak peak (3) Strong peak

Tabular info comes from CRC NIR handbook

Peak info:

983 (2) OH from water (maybe but maybe not....) (979) Phenolic OH (990)

1170 (2) Alkene, polyenes (1170)

1347 (2) Methyl (1360) larger delta than normal

1436 (3) Aromatic amine (1432) Methylene (1440)

1480 (3) Polyamide (1480) Secondary Amine (1481) Amide/Protein (1483)

and in general, high absorbance in the range of 1436 - 1480; aromatic amine is strong here.

Now my initial thought was that polyvinyl alcohol related compounds might show up in this NIR spectrum. However, the OH is not dominant here if at all, and the compound is not soluble. The water question can be settled and separated out by sustained drying of the sample. Phenols are moderately soluble in water. Now we know that this compound is highly insoluble. This indicates that OH solubility is not a strong factor here. The polymer and protein signatures are what dominate here. For now, I will presume water over phenol in the 983 nm group but will keep an eye on this.

Most conductive polymers are polyenes. Polyenes are a main target of astrobiology. Polyenes share some chemical reaction properties with PVC vs PVA. PVC is C2H3Cl (notice no OH in any way). So if of a plastic

nature would seem to be more likely of PVC family than PVA since we are insoluble and somewhat crystalline nature under scope and brittle.

THE MATERIAL DOES NOT PASS A CONDUCTIVE TEST WITH THE OHMMETER AT THIS POINT.

so 983 peak remains undecided between OH water and alkene, polyene.

Now the aromatic amines, polyamides, secondary amine, and amide protein all speak of being a polymerized protein, especially in light of the borax reaction which has produced it.

This exists as the most current interpretation of this material that has been created, a polymerized protein. Unclear at this point if the polymerization is likely to come from aromatic or amide origin, or both for that matter. We know that in the end a "rubberized" polymer is developed, but this can come from the other TSP surface synth-cell polymer layer. But out of curiosity, is rubber aromatic or amide based?

Rubber is alkene based. OK, this gets very interesting. Synthetic rubber is a copolymer between styrene and butadiene. Styrene is a VINYL group attached to an aromatic ring. Butadiene is an alkene.

Guess what, all the signs are starting to point the same direction. Aromatic, vinyl, alkenes are the components of synthetic rubber, which is most characteristic of the mature clot gross properties. Aromatic, vinyl, alkenes appear to be dominant structures of our NIR plots. Do not keep trying to force a single functional group. The combination of these three functional groups along with synthetic blood go a long way toward explaining the culture findings in general.

Start looking at these three functional groups in a combined sense, understanding that separations in the culture will likely break these into parts. We will now keep an eye on the NIR plots more closely with these various combinations.

Let's create more of the proposed polymerized protein, now designated as TSP 100.10. Done. Drying.

I am trying to acidify TSP 100.10 material w/ moderately strong H2SO4. It is difficult. It may be slightly soluble in H2SO4 but certainly not easily. This means that both polymeric forms, TSP surface layer synth cell-polymer layer and this newly developed polymer from the decant and borax are BOTH very much insoluble in water and difficult to solubilize in reagents as well. Just like the clot was....

I need to conduct solubility tests on 100.10 just like I did on the synth surface polymer on TSP 070. More study there again as TSP 100.0 matures.

My goal is to attempt to dissolve (hydrolize) TSP 100.10 for a UV run to determine if protein is detected.

OK, that was straightforward. Definite absorbance at 275 nm. Protein confirmed. Polymeric protein remains confirmed. Now our goal is to try and determine the nature of the polymer. Our first NIR run says we have:

- 1. alkenes, polyenes
- 2. methyl possible
- 3. aromatic amines
- 4. methylene
- 5. polyamide
- 5. secondary amine
- 6. amide/protein
- 7. polyamide, amide

Therefore, the reduced and presumed polymer set is:

- 1. polyenes
- 2. aromatics
- 3. amines
- 4. polyamide

Seems like an ideal polymerized protein to me. Qualifies as a constituent of the final clotting process. Missing quality seems to be the rubber nature, but this seems to fit quite well for the TSP 070 surface synth layer - polymer.

The essence of the clot seems to be established at this point:

1. Synth erythrocytes w/hemoglobin

2. Two polymeric forms, one for structual density and the other for elasticity. Both highly insoluble, even in many strong reagents/acids/caustics.

This is a major achievement. It would certainly be of benefit to understand how the two polymers may join together.

Now the rubber property would be imparted by the vinyl group, which we now understand is a part of the copolymeric process of synthetic rubber. We may well have, and are expected to have a copolymer arrangement here.

This means that we would expect to see the vinyl component from one of the two polymers, and the strongest candidate for that is now the TSP synth cell - polymer surface layer. Let's look at NIR plots.

Guess what? (The database sure pays off). From notes of Nov 12, the vinyl component is in the precipitate! This is the other half of the TSP prior to titration!. We have all the parts we need. We have therefore separated between:

Three polymers, consistent of all needed parts:

One separated polymer contains:

- 1. polyenes
- 2. aromatics
- 3. amines
- 4. polyamide

(i.e., proteins and binding polymers)

The second polymer, i.e. the precipitate of the titration:

1. contains vinyl

(i.e., imparts elasticity)

and synthetic blood containing hemoglobin in a polymer matrix, also elastic in nature (thus protein & polymer here also).

This means you need to know if the precipitate contains protein also. I think NIR says yes. But let's work on that one.

This is all leading to the likely conclusion of three polymeric protein complexes:

1. Decant

- 2. Precipitate
- 3. Surface synth cell layer

Each one will have its own dominant chemical properties.

This would therefore certainly be a copolymeric operation. Expected that decant + precipititate = synth cell surface layer with synthetic blood.

The precipitate to the titration is 100.6. We have enough of this material to work with for now. It is important to realize that this layer, upon centrifuge, actually contains two layers, an upper grey layer higher in volume, and a lower white layer minimal in volume. This layer now becomes important in seeking out the vinyl contribution to the overall polymer structure. For now we will focus only on the grey upper layer as it comprises the bulk of the volume of centrifugation.

We have it. NIR evaluation of the borax polymer vs the precipitated tells us the distribution and make up of the synth surface layer.

TSP 100.6 (the precipitate from titration) has high absorbance in the 1640 region. This is the vinyl dominant region of NIR.

TSP 100.10 (the borax polymer) has high absorbance in the 1440 region. This is dominated by aromatic amine. The precipitate is not high in protein content. 293 nm and 232 peaks were identified in the 100.6 acidified precipitate.

So what this is saying is that the middle layer of the culture (most volume) contributes the bulk of protein. The precipitate contributes the elasticity.

So our three main components (everything is temperature and pH dependent for expression (~18 expressions thus far) are:

- 1. Synthetic blood (w/hemoglobin) (scores of proteins within blood) (surface layer of mature culture)
- 2. A set of proteins that can be polymerized with borax, structurally firm but not elastic. (within decant)
- 3. A vlnyl contribution for elasticity of the culture. (within precipitate)

This makes up the majority of the clot structure.

1,3-Butadiene (a synthetic rubber) from NIST data base UV spectral search shows a peak at 293. This is a vinyl compound and then a match for our spectrum of TSP 100.6 at this point (not the rest of the spectrum nor our 232 peak found.

NIST and Chemspider both have some UV searching capability if you can get a part of the name. VERY VALUABLE!

NOTE:

The styrene UV spectrum explains high absorbance towards the 232 peak [NOTE DEC 08 2023 I SEE NO BASIS FOR THIS PREVIOUS STATEMENT - IT IS NOT ACCEPTED AT THIS TIME].

Synthetic rubber is a copolymer between styrene and butadiene.

Structural formula of Styrene Butadiene Rubber:



Fig: Structural formula of Styrene Butadiene Rubber

2023.11.17 Solubility Methylation

The picture looks increasingly clear. It would seem that we have a genetically engineered co-polymerization process taking place here. The process involves synthetic blood, an aromatic protein polymer, and a synthetic rubber equivalent polymer. Their origin is the Cross Domain Bacteria (CDB). The result is increased coagulation or a clotting of the blood. The end result is definitely a lethal threat.

Need to outline for panel and conditions of disclosure.

Lab needs:

1.Look at TSP 070 on surface layer polymer. Expected now to be a combination of aromatic protein polymer and synthetic rubber (vinyl).

2. Solubility studies on aromatic protein polymer. Can iron be used to polymerize something like Boron (Borax) did?

Boron is a metalloid with 3 valence electrons.

"There are many forms of polymerization and different systems exist to categorize them."

"In more straightforward polymerizations, alkenes form polymers through relatively simple radical reactions;" "Step-growth and chain-growth are the main classes of polymerization reaction mechanisms. The former is often easier to implement but requires precise control of stoichiometry. The latter more reliably affords high molecular-weight polymers, but only applies to certain monomers."

"The manner in which polymerization is conducted is a highly evolved technology. Methods include emulsion polymerization, solution polymerization, suspension polymerization, and precipitation polymerization."

3. Methylation - put an end to a polymer chain reaction, it would seem? Can you induce methylation in the lab? Mitigation implications. Ultrasound now understood more clearly also. NAC, Vit C, Citrate influence on cultures again.

4. Consider setting up the next paper: (?) Cross Domain Bacteria (CDB) : Synthetic Blood, Bloplastics, and the Global Blood Clot

5. Head toward replication of TSP 070 activity, namely nucleic acids and bacteriophage.

6. Increased understanding of enzyme influence. Concentration appears dramatic. Work towards enzyme isolation.

7. Begin exploitation of NIST UV Spectra, can be very helpful. Suggest modeling of synthetic rubber by combining UV plots of styrene and butadiene.

UV Spectra on NIST site can be extremely helpful. Have not been aware of them. Some level of compensation for lack of MID IR instrument exists with this information. If you get to the point of prospective chemical group, e.g., vinyl, NIST can be used to bring up related UV spectra. This can add an important layer of confirmation to NIR work (and generalities) by bringing a chemical group into the picture of confirmation instead of only functional groups. No knowledge yet of affordable NIR data base for structure but functional group analysis is in good position with CRC handbook.

Looking at the comparison NIR plot between human blood and synthetic blood is most intriguing. There is much to be studied here. An acceptable reference spectrum for blood is now in question. However, the synth blood spectrum is not in doubt as to what it contains. Dramatic differences exist, and they are, in part at least, now anticipated and predictable. Notice how a major feature of the decant (e.g. 100.5 and 100.9) is that they gradually transform from clear amber toward white as a polymerization takes place. This dynamic change will be important to get a handle on. Vitals and pl (variable value?) are two important tools as well as UV monitoring. Is the protein polymer changing structure here and can you show this? It is almost certain to be the case.

The refractometer will hold its zero calibration value even after turning off. This is great, saves a lot of extra trouble. With respect to vitals, refraction is ending up being the most sensitive indicator with a variation of \sim 20% within cultures over time. TDS is next with a variation of \sim 8%. pH is the least sensitive, with a variation of \sim 3%. All are useful, especially when combined. The idea of adopting an RMS as a reference frame for my developed methods in probability analysis seems to be very convenient and logical and simple to implement.

For the record of years past, the final work from work years ago resulted in the probability model of:

 $\begin{array}{l} \mbox{Pr\%} = (200/pi) * atan(c * delta x) \\ \mbox{and} \\ \mbox{c} = 1/(\mbox{Pr range}) * tan(\mbox{Pr level\%} * (pi/200)) \end{array}$

VALUES IN RADIANS.

In this most recent implementation of my model, the RMS of the data set is used as the Pr range value. The Pr level adopted for that reference range is 71%. Peak Probability will be 1.414 * Pr%. This is a logical reference frame to adopt as an alternative to E90 or z=1 score, etc. This original work was done years ago and has shown itself to be a valuable and useful model to many applications. Much simpler to implement than the standard distribution curve and it is far more realistic to accomodate extreme values or handle the "long tail" problem.

Culture change can be modeled very well with this model. Changes in any parameter can be modeled in a difference from a reference culture value. Differences can then be converted to a daily slope. Daily slopes can then be modeled from a probabilistic sense using the model above and the interest level of each culture can be established for each sample. Overall interest level can be assessed with the average of the three different vitals monitored.

This model can be applied rather easily to any situation of interest, e.g, financial, data samples, etc, any time series data.

In terms of application of this model, TSP 100.5 and 100.9 are indeed high interest cultures. There is a lot of change going on with respect to the vitals as polymerization occurs. This change process can undoubtedly be studied (and learned from) in great detail. It is an area where interference of protein development can be anticpated with sufficient knowledge of internal biochemistry taking place.

TSP 100.5 is now pure white, compared to original amber color. Time span 7 days. TSP 100.9 is now a pale amber, time period 5 days.

The paper has been started.

Human Transformation : Synthetic Blood, Bioplastics, and the Global Blood Clot Clifford E Carnicom Nov 17 2023



2023.11.18 Borax polymer solubility

A few solubility tests have been run on the borax polymer today. Results are:

1. in strong acid (H2SO4), not soluble

2. in strong alkaline (NaOH-KOH), slowly soluble

3. in acetone, not soluble

4. in ethanol, not soluble

These are helpful results. It has not been published yet, however, the deceased blood clot was eventually dissolved in NaOH-KOH solution over a period of \sim 2 months. This was recorded on Aug 11 and Aug 13 2023 in Vol 30 of the laboratory notebooks.

This now becomes important as we have similar solubity behavior between the deceased blood clot (the most dense and developed, rubber polymer properties) and the borax polymer that has been developed. We know that KOH-NaOH has generally been our only successful reagent for dissolution of any of the filaments. We know they help to break down the disulfide bonds, as in hair.

A question is what other types of polymer bonds might they break down? In this case borate bonds are involved. So we have a good question coming up with the polyvinyl-borax polymer that can be made (the "slime" case). Will KOH-NOH work on that as well? If so, it shows it is effective on the polyvinyl bonds and borax, not just disulfide.

Yes, lye does break down slime. YT video demo: https://www.youtube.com/watch?v=abJ1ETlhZ-w He needed add some water instead of so much direct powder lye, but it still demonstrates the point. Water was used at the end and more clearly shows that breakdown has occurred.

This is valuable as we are not dealing with disulfide bonds here. Vinyl bonds are involved, as they are in our case also.

This was a valuable trial here. Another linkage in bond disruption between the clot polymer and one of the culture polymer forms.

Back to our questions of yesterday then:

Lab needs:

1.Look at TSP 070 on surface layer polymer. Expected now to be a combination of aromatic protein polymer and synthetic rubber (vinyl).

2. DONE Solubility studies on aromatic protein polymer. Can iron be used to polymerize something like Boron (Borax) did?

Boron is a metalloid with 3 valence electrons.

DONE. SEE LAB NOTES ABOVE.

3. DONE Methylation - put an end to a polymer chain reaction, it would seem? Can you induce methylation in the lab? Mitigation implications. Ultrasound now understood more clearly also. NAC, Vit C, Citrate influence on cultures again.

MUCH BETTER UNDERSTANDING IN PLACE. METHYLATION IS ACROSS THE BOARD IN THE HUMAN BODY. AFFECTS MOST EVERYTHING. ANY DISTURBANCE IN METHYLATION IS GOING TO CAUSE MAJOR PROBLEMS.

4. DONE Consider setting up the next paper: (?) Cross Domain Bacteria (CDB) : Synthetic Blood, Bloplastics, and the Global Blood Clot Title changed to: Human Transformation: Synthetic Blood, Bloplastics, and the Global Blood Clot PAPER HAS BEEN STARTED.

5. Head toward replication of TSP 070 activity, namely nucleic acids and bacteriophage.

6. Increased understanding of enzyme influence. Concentration appears dramatic. Work towards enzyme isolation.

7. Begin exploitation of NIST UV Spectra, can be very helpful. Suggest modeling of synthetic rubber by combining UV plots of styrene and butadiene.

8. DONE Blood Evaluation. Results highly favorable.

I want to start learning about methylation. Here we go, it is bigtime right away:

"DNA methylation is a biological process by which methyl groups are added to the DNA molecule. Methylation can change the activity of a DNA segment without changing the sequence. When located in a gene promoter, DNA methylation typically acts to repress gene transcription"

"As of 2016, two nucleobases have been found on which natural, enzymatic DNA methylation takes place: adenine and cytosine."

It is exactly what I envisioned. It does not apply only to an alkyl chain by any means. it is taking place here on an cyclic rings and aromatic rings as well. This says that it is quite feasible that methylation increase in the body may well assist in the disruption of an aromatic polymer protein. This is very favorable information. And by the way, DNA is a polymer as well. Yes, it absolutely is, it is a polynucleotide. Photos below.

Basically methylation helps to regulate DNA to stay healthy and thwart mutations.

"Epigenetics, the study of the chemical modification of specific genes or gene-associated proteins of an organism."

"The principal type of epigenetic modification that is understood is methylation (addition of a methyl group). Methylation can be transient and can change rapidly during the life span of a cell or organism"

A gene is a segment of DNA that contains sequences of many bases, varying in size from a few hundred to 2 million. Each gene affects a specific aspect of health. For example, some genes contain instructions on how to make specific proteins.

A gene is a specific segment of DNA that tells cells how to function

DNA is the genetic material or code that tells cells in the body how to replicate themselves. Almost every cell in an organism s body contains a complete copy of its genome, packaged into chromosomes

Methylation is a massively important topic in human health. There is a lot of ground to cover here:

"What is methylation?

At the most basic level, methylation and demethylation refer to the transfer of methyl groups simple structures of one carbon and three hydrogen molecules (CH3) to and from various bioactive compounds in the body. These compounds (e.g., proteins, enzymes, hormones, and more) must be methylated in order to function optimally or to create other substances required by the body.

For example, methylation is essential for the production of certain bioactive vitamins (e.g., folate and vitamin B12), amino acids, neurotransmitters, hormones, red blood cells, DNA, RNA, and antioxidants that directly impact cardiovascular, neurological, reproductive health, energy production, detoxification pathways, and more."

"Methylation influences just about every essential process in the body. It's needed for the production of amino acids, neurotransmitters, hormones, red blood cells, DNA, RNA, and antioxidants; for proper detoxification of hormones and toxins; and for determining whether certain genes will be expressed or not (the power of epigenetics). It's a big deal.

But when this vital process is compromised (which can happen as a result of a variety of dietary, lifestyle, and genetic factors) your physical and mental well-being, from a cellular to whole-body level, will pay the price.

The good news: A simple homocysteine test can help you get a glimpse of your own methylation cycle health and help determine whether you need to make any dietary or lifestyle changes "

https://www.mindbodygreen.com/articles/what-is-methylation

"What does methylation do to DNA, and why does it mater? As a metabolic process, it basically switches genes on and off and repairs DNA.

This is a big deal, because gene expression has the power to influence many aspects of health, including one s risk for certain diseases, such as neurological issues and some cancers.

Methylation and demethylation, which is the opposite process, basically work together to control different functions throughout the body."

https://draxe.com/health/methylation/

So quite easy to see how methylation is on par with enzymes in mitigation strategies.

Let's look at our blood to keep pulse on status.

Report: Blood status looks highly favorable. Methods make sense, pH regulation, methylation, protein interruption, electrolytes., salicylate

Working on research paper. Most dramatic blood culture images (the original appearance) is recorded on Sep 19 2023. Actually occurred in TSP 01. Phenomenal result.

I have a very significant plot now to look at. It is a NIR comparison of TSP 100.5 (Decant) over a 12 day incubation period. This is from the time that it is decanted (amber color) until it turns white approx 10 days later.

Main feature I am looking at is that appears there is a bulk of mass (absorbance) shift from the Nov 06 TSP 100.5 (amber) at a very broad peak of \sim 1620 +/- 20 nm to a more focused peak at Nov 18 TSP 100.5 (White) of 1516. They do however, continue to share a great deal, and in fact the majority of the turf from 1530 - 1650 nm.

Interpretation of this is: 1620 is alkene and vinyl 1520 is protein, 1515 is polyamide

this makes a lot of sense. It is saying it starts out stronger in the vinyl section but then shifts over to a protein nature. However, it still retains the vinyl nature. This means that a conglomerate is being formed, i.e. a polymerized protein as your surmise.

This is exactly what you envision happening. This answers a very important question on culture dynamics.

this makes a lot of sense. It is saying it starts out stronger in the vinyl section but then shifts over to a protein nature. However, it still retains the vinyl nature. This means that a conglomerate is being formed, i.e. a polymerized protein as your surmise.

This is exactly what you envision happening. This answers a very important question on culture dynamics.



2023.11.19 Project Planning

Lots going on, as usual.

The paper is posted, I regard it as a good mark on grounding. Steady she goes..

Adjust the list.

- 1. Done: solubility study on borax polymer. All perfectly consistent w/ KOH-NaOH impact.
- 2. Done: Methylation is at the heart of health. On solid ground
- 3. Done : Paper: Human Transformation: Synthetic Blood, Bloplastics, and the Global Blood Clot
- 10. DONE PDF of 2023 papers revised. Review lab notebook publishing schedule
- 4. UV Spectra on NIST are a new asset. Can we find NIR database?
- 5. Head toward replication of TSP 070 activity, namely nucleic acids and bacteriophage.
- 6. Enzyme concentration studies. Enzyme isolation studies.
- 7. MIxing the culture into blood in various ways. Impact?
- 8. Keep sorting out the chemistry of the culture dynamics and progression
- 1. Native ---pl series investigation?
- Decant
 pl series investigation?
- 2. Decant progression- transform pl series investigation
- 3. . Precipitate
- 4. Surface layer synth cells polymer
- 5. NIR of Human Blood vs Synth layer sort this out alcohol?

9. Duplicate TSP 070

- 1. Nucleic acids
- 2. Bacteriophage

The general sequence coming up is:

- 1. Nucleic acids, bacterophage
- 2. Health Lineage
- 3. Panel on:
- 1. Culture
- 2. Mitigation
- 4. Laboratory notes up phased

Quite the software battle today. Need a database that will kick out a report that autosizes for printing a

variable text field, such as this one. No dice, not an easy one. Jasper report may be a candidate but nobody up front doing this on regular basis. Very surprising. You have a problem on this. Database lab log is perfect for my use, but not for distribution. Access and other heavy duty dababase progams will do it, but not the simple software. Crystal reports, JasperReport. Astounding how difficutl that is.

OK fantastic. I have found it. Online converter. Amazing service, files look great. Use CS View if need to whittle file, and then the online converter, or maybe just straight with the whole file. Beautiful solutionl.

https://cloudconvert.com/

Cloudconvert.com is my ticket. I have created an account. I am free to write uninterrupted now.

2023.11.20 UV NIR TSP 70 Nucleic Acid

4. DONE UV Spectra on NIST are a new asset. Can we find NIR database? - NO. There simply are none that are available affordably. \$1500 min for 1000 spectra, always with cost. NIR is taking off in popularity due to its ease of use. But you are on your own with respect to spectral interpretation.

5. Head toward replication of TSP 070 activity, namely nucleic acids and bacteriophage.

6. Enzyme concentration studies. Enzyme isolation studies.

7. MIxing the culture into blood in various ways. Impact?

8. Keep sorting out the chemistry of the culture dynamics and progression

1. Native ----pl series investigation?

Decant
 pl series investigation?

2. Decant progression- transform pl series investigation

3. . Precipitate

4. Surface layer - synth cells - polymer

5. NIR of Human Blood vs Synth layer - sort this out - alcohol?

9. Duplicate TSP 070

1. Nucleic acids

2. Bacteriophage

.10. Construct a UV & NIR plot of polyvinyl alcohol?

The general sequence coming up is:

1. Nucleic acids, bacterophage

2. Health Lineage

3. Panel on:

1. Culture

2. Mitigation

This is the topic to work on today:

1. Native

---pl series investigation?

I will compare TSP 070 (Mature native culture Oct 19) against TSP 100 (Native Nov 08). Am interested in all 3 issues: pl behavior, UV and NIR.

First observation is that TSP 070 is transparent, TSP 100 is cloudy. This makes sense in that we know a mature culture eventually separates into 3 layers: top polymer, middle bulk of volume, bottom precipitate). Titration and decanting is an entirely separate process of separation. For now only time and gravity are involved in this analysis.

UV: On Nov 05 2023 we collected a UV spectrum on TSP 070, the mature culture. We had an absorbance of 259 nm. We looked up on this and we found a strong existence of the literature on nucleic acid absorbance at 259 nm. We have it again, as solid as can be withTSP 070, absorbance well defined at 259 nm.

OK, there are VERY important differences between the UV spectra of TSP 070 (Mature) and TSP 100 (under incubation, no complete separation into 3 layers yet). TSP 070 (Mature) has a very clearly defined peak at 259

nm. TSP 100 does not have this YET and instead has peaks at 273 and a very broad shallow peak at ~315. So quite different, and the absorption at 273 once again indicates likely tyrosine presence. This is therefore a very important difference to notice and to monitor. 315 nm is a little hard to pin down since it is no broad and shallow. Let's see if we can find a UV spectrum of DNA.

This information leads us to suggest that the direction of the mature culture will be the creation on nucleic acids. Next step is to monitor the second culture, i.e., TSP 100 and seed if the event repeats itself. Also, any other contenders for 259 nm absorption? What does NIR say? What is the UV profile of DNA? What is the NIR profile of DNA?

Ref UV DNA easy to come by:

Images attached.

https://www.researchgate.net/figure/UV-absorption-spectra-of-native-DNA-black-and-glycated-DNA-incubated-for-1-day

https://www.researchgate.net/figure/UV-absorption-spectra-of-native-DNA-DNA-lysine-photoadduct-and_fig1_40728359

I would say the case is pretty strong.

Also found NIR of DNA:

https://www.sshade.eu/data/SPECTRUM OP 20181101 05

should be looking for a NIR peak of about 1400 nm. possible a valley also near 1100.

if you can come up with a good idea of what something is, you can often find a reference spectra to determine if you are right or not. This method is acutally preferable to having a full IR spectrum and NOT having any idea what it is. This shows the value of being able to come up with a good estimate of what might be involved. Case for nucleic acids is developing because of this method, although originally you had no idea that DNA might be involved or even detectable with the culture. It may be.

2023.11.21 Nucleic Acids (?) - DNA- PVA

There is a good problem before us. Are we dealing with Nucleic Acids/DNA in TSP 70 or are there alternatives such as alcohols or aromatics involved? Why does the human blood NIR have such a broad strong peak in the apparent alcohol section? If you do have DNA/nucleic acids would you be able to isoate it with chilled alcohol? Is there anything else know to absorb so strongly at 259 nm? How do you reconcile the NIR plot of TSP 70 with UV plot and qualitative chemistry? How do you interpret the dynamics of culture change, as revealed by NIR comparison, between TSP 70 (mature) and TSP 100 (developing)? From analysis of the reference NIR plot, we expect to see a peak at ~1455. We have a broad shallow peak in that region but the peaks at 1500 and 1600 swamp 1455 out. Need to discern between all three.

A polyvinyl NIR andUV plot is now going to help you sort out the aromatic - alcohol problem.

We also have the NIR culture dynamics of TSP 070 vs TSP 100 now available. The shifts between 1600 and 1500 are very important now to identify as to what is actually changing. Alcohol - Aromatics - Vinyl?

Would TSP 100 mature more quickly if incubated? Room temps are pretty low now at night. Notice, however, that TSP 100 is indeed showing the changes of maturation. Color change and 3 layer separation is now readily visible. Also in the spare TSP 100 material. We definitely have solid replication of maturation; process takes ~ 2 weeks to become definite and identifiable.

Our previous list :

- 1. Head toward replication of TSP 070 activity, namely nucleic acids and bacteriophage.
- 2. Enzyme concentration studies. Enzyme isolation studies.
- 3. Mixing the culture into blood in various ways. Impact?
- 4. Keep sorting out the chemistry of the culture dynamics and progression
- 1. Native
- ---pl series investigation?
- 2. Decant
- 1. pl series investigation?
- 2. Decant progression- transform
- pl series investigation
- 5. . Precipitate
- 6. Surface layer synth cells polymer
- 7. NIR of Human Blood vs Synth layer sort this out alcohol?
- 8. Duplicate TSP 070
- 1. Nucleic acids
- 2. Bacteriophage
- 9. Construct a UV & NIR plot of polyvinyl alcohol?

The general sequence coming up is:

- 1. Nucleic acids, bacterophage
- 2. Health Lineage
- 3. Panel on:
- 1. Culture
- 2. Mitigation

A very smart move to acquire Polyvinyl Alcohol. Strong easy spectrum to acquire, 4% solution. 500ul in 3ml more than sufficient for strong UV spectrum. Peaks at 240 and 268. Distinctive spectrum, not like anything seen thus far. THe 240 peak shifted quite a bit with a strong concentration, the 268 held steady. The shape of the profile is another distinguishing feature to pay attention to in UV we see. If you were to just take the peak number of 268 you might want to say protein. VERY FAR FROM THE TRUTH, very far away from protein. You see one of your strong clues is the profile or slope of the spectrum, nothing like protein that we have seen. So now you pay close attention to this and it is all the more reason you must do your best to identify a reference candidate and FIND THE UV SPECTRUM for that candidate before you draw strong conclusion based upon peak info alone, and often singular peak info.

Now let's compare acquired PVA spectrum with what someone else said was a reference.

By direct measurement and acquistion of 99.9% PVA I have the following:

230 (220, 230)very strong 269 (270) weak 277 (280) weak (now revised with extended quartz cuvette use).

Now I notice I have another small peak at ~ 277 nm. With the slopes of my curve considered, this is also a massive departure even from the claimed 282. SO THIS IS A RADICAL DIFFERENCE. Let's look at NIST. NIST does not have UV. Another name for polyvinyl alcohol is Ethenol (not Ethanol!).

and guess what, it does not match at all with what is being claimed as a reference. This is important and damages confidence in "reference data".

This is the reference polyvinyl alcohol (PVA) information I have located before. This took place in Nov 14 notes (first day of digital lab notes):

We have:

"The optical absorption measurements for poly(vinyl alcohol) evidenced bands in the UV region (see Figure 2) at 223, 282 and 327 nm assigned to n?p* transition, due to the presence of the C=O group and p?p* transition, respectively which arise from unsaturated bonds, C=O and/or C=C, which are present in the tail-head of the polymer."

https://www.researchgate.net/figure/UV-absorption-spectrum-of-the-polyvinyl-alcohol fig2 289203733

I have a spectrum that shares a great deal in the general sense of what is shown. I am out of range for the 223 but it certainly is following the profile. I have absorbance at ~338 and 282 general peak with derivative determination.

Also we have:

"Spectral examination of the solvolysis products of vinyl acetate and 1-methoxyvinyl ester copolymers shows unequivocally that the absorption bands in PVA at 225, 280, and 330 m μ are related to carbonyl-containing structures."

https://onlinelibrary.wiley.com/doi/abs/10.1002/pol.1963.100010412

Neither NIST or Chemspider has a reference UV spectrum for polyvinyl alcohol. That is a surprise. NIST has Mid IR. Another thing we learn is that Chemspider requires the chemical name to find something, it does not like popular names. The chemical name for polyvinyl alcohol is ethenol which I had no idea of.

So we have a little bit of an issue. I have UV by direct measurement. Reference claims of absorption are not working well at all. Right now I trust my direct measurement the most.

Another source is giving of PVA dissolved in water (just like I am doing) with peaks of:

231, 281, and 324.

https://link.springer.com/article/10.1007/s40089-015-0150-y

another source gave (see above);

223, 282 and 327 nm

another source gave:

225, 280, and 330 mµ

The average reference from all three of these sources is: 226, 281, 327. Now I can not measure 226 easily so that one is out. Let's redo my spectrum from 250 to 350. The 324 peak is shown to be weak. The 281 is much stronger. Let's see if we can isolate these. Is ther any possibility I have contamination? It would not seem likely to me. I have run it again. I have well defined peaks at: 268.5 276.5.

These simply do not match the reference claimed. The 276.5 is distinctly separate, with respect to slope, of the claimed 281 nm. The 327 does not even show up. This is entirely perplexing. I have a purchased commercial product that is stated to be, acts like, looks like, makes a film like PVA. But my own spectrum acquired directly with no apparent difficulties does not match stated reference spectra. I am glad that I am doing this but find it to be MOST puzzling.

OK, we are going to run another reference spectrum of some sort with a different material. There is simply no reason to doubt my instrument at this point.

Isopropyl alcohol should be an example, but NIST and Chemspider does not have this either. I found NIR of isopropyl alcohol but not UV at: https://www.researchgate.net/figure/Isopropyl-alcohol-spectrum-generated-in-10-milliseconds-and-are-compared-to-Pac

OK, I am going to use acetic acid, vinegar, as a reference control. In addition I actually have 2 quartz cuvettes that wil extend my range down to 190 nm. This is a crucial advantage. I will buy two more, take advantage of them, and I will extend my UV range considerably.

For special and important cases, I will now be able to use quartz cuvettes to get down to 190 nm.

This is great. A 25% increase in capability of the instrument for \$25.

Acetic acid comes out somewhat OK. I get a strong peak at 200 nm, NIST shows it at 210, another shows it at 212, but at least we are in the neighborhood and can get significantly extended range now. Let's go back to PVA with the quartz cuvette. If you are willing to use 1 cuvette at a time your 4 cuvettes should last indefinitely if you take care of them.

OK, this change was incredibly valuable to make. I have not matched PVA where it counts. I have the peak now at 230 nm. Close enough and shows me the kind of error that can exist even under very good conditions. What happens here is the peak at 327 simply does not exist for me. The peak at 281 is very weak, exists but poor to bank on. I have an additional peak at 269 that no one is talking about, also weak but more more defined. However, at 230, it is off the chart, and absorbance is of 2 magnitude. Extremely strong with 1 drop PVA in 3 ml cuvette. You would simply never see this without the extended range, and the magnitude of this peak is so much greater than the others that this peak alone is what serves as the reference match point. And now we have matched PVA.

This brings in a new lease to UV analysis. I may discover quite a bit more now with UV analysis of most any sample. Lot's that can go on below that 235 nm point.... This really does change things.

Now we can move on. We now have uniqueness of PVA available with combination of UV and NIR card has been made (nice film appearance).

I think we need to remeasure UV on TSP 70 and TSP 100 with extended cuvette range and then combine with the NIR data to sort out whether we have alcohols or aromatics involved.

A DNA spectrum:

https://www.degruyter.com/document/doi/10.1515/nanoph-2020-0225/html

Measured UV:

PVA : 230 (220, 230)very strong 269 (270) weak 277 (280) weak

Now for the chilled alcohol DNA test:

I see absolutely no sign of DNA strands being formed and rising within chilled alcohol being placed on the top of TSP 070.

There is, however, a definite precipitate being formed so an important reaction of some type is taking place. Recall our early test with alcohol that also produced a precipitate. Never investigated further. The nature of this precipitate will need to be determined. The precipitate forms in the TSP 070 portion of the culture-alcohol mixture, not in the alcohol. Further indication that it does not involve DNA, but a protein or polymer is very likely. The ethanol may therefore not need to be chilled at all but it remains so for storage right now until test is made with room temperature ethanol.

The precipitate does centrifuge to the bottom of the tube within the TSP 070 solution. Alcohol stays clear above. Let' start by looking under the scope.

There is very little material to work with, just enough to get under the scope.

And yet another form. My vote is protein. It is composed of very small needle crystals, about .4 um in thickness and 6-10 um long. Interspersed with occasional groups of undersized synth cells.

I would say the Nucleic acid - DNA hypothesis is going out the door and that we likely have a protein in solution. Not sure what the ~210 nm UV absorbance is referring to yet. But combined with NIR work, this is going the direction of an aromatic protein within TSP 070, not an alcohol.

[NOTE FOR DEC 09 2023: A THOROUGHLY CHILLED ALCOHOL DNA TEST GIVES A POSTIVE RESULT. NO SETTLING PRECIPITATE FORMED].

[ALL 3 TESTS FOR DNA ARE POSTIVE: UV, NIR (HYDROXYL GROUP), CHILLED ALCOHOL)



2023.11.22 Solubility-Health-Blood(Syn)

I am seeing that the CRC NIR reference is not as specific as it appears to be. Acquiring a direct measurement control NIR plot of Polyvinyl Alcohol (PVA) is turning out to be very insightful. CRC Handbook cannot be counted on as the almighty NIR reference that I had presumed it was. This introduces a major level of caution as well as additional verification required on any NIR claims that are made. I will demonstrate this point with an analysis of the pure, directly acquired and measured PVA. CRC will be a very useful reference, but not an absolute reference, at it will now be combined more readily with the generalized charts such as from Galaxy Scientific.

(The image icons within this database serve as major prompts; they must be opened jointly and separately within an image editor or viewer)

TO DEMONSTRATE THE WEAKNESSES IN THE PROCESS: Now let's make a standard analysis with the info I have available from below and measured directly:

UV : PVA : 230 (220, 230)very strong 269 (270) weak 277 (280) weak TSP 070 213 261

269 280 peaks indicate protein 230 somewhat unknown but as we recall it is a good indicator of protein folding changes so these are both in support of protein.

NIR:

Peak 1201 (1200) 1489 (1490) 1574 (1570) 1669 (1670) Broad Absorption Peak 1450-1600 Valley: 1005 (1000, 10010) 1284 (1280)

Valley at 1005 would indicate LACK of secondary alcohol (-CH-OH) (1004) LACK of primary aromatic amine (1003) LACK of OH from tertiary alcohol (-C-OH) (1006)

Peak of 1201 indicates OH from water (1200) (sample is dried thoroughly) Methyl (1195, 1194)

Valley at 1284 would indicate LACK of [NO CRC SIGNAL LISTED 2012]

Peak at 1489 indicates primary aromatic amine (1489.5) primary aromatic amine (1487) secondary amine (1486) amide/protein (1490) OH polymeric (1490) amide/protein 1492 aromatic amine (1493) (1493.5)

Peak at 1574 indicates polyamide (1570) amide/protein (1570) alcohol or water (1580) alkyl alcohol or water (1583)

Now what we see here is the preponderance of information points VERY strongly toward the existence of protein.

However, here is the chemical formula for polyvinyl alcohol: $\ensuremath{\mathsf{C2H4O}}$

or

[-CH2CHOH-]n

and the Lewis diagram is insightful as it shows how the chain forms with alternating H, OH bonds.

NOW THE BIG LESSON HERE IS THAT THERE IS NO PROTEIN WHATSOEVER INVOLVED. A PROTEIN ASSESSMENT WOULD BE ENTIRELY ERRONEOUS EVEN THOUGH THERE IS SUBSTANTIAL INFORMATION TO ACCEPT THAT INTERPRETATION.

So our job is to study this situation more thoroughly to prevent this from ever happening.

1. First lesson is no matter how many entries there are for a certain category that does not necessarily mean

that is what it is at all. A single alternate entry can (and is here) the proper choice (in this case "polymeric alcohol" at 1490. Truth is the correct choice was spot on, you just needed to be observant to catch it. Therefore everything you know, see and can learn is important in the process, and in this case, physical observation and behavior and solubility of the material is key information in the process.

2. Second lesson is that UV was a total loss and detour on both accounts. Protein inference was incorrect on BOTH accounts, folding and blip in the 270-280 region. I would say the shape of the UV profile is now as important as the peak absorbance. Certain classes of materials will likely exhibit similarity in slope profiles. NIST data base might be useful here but you can see how much even reference spectra can vary depending upon scales used. This also shows the value of the colorimetric route on proteins - this should settle the question.

What this means is that you need to be very CAREFUL at all times about being duped into something. You must check, check, cross check and put all your wits together, and independent methods and controls are ALWAYS essential. No guessing or surmising is allowed when you draw the final conclusions.

Now if you play your cards right, NIR actually gives us a perfect answer:

- 1. Methyl group (does seem likely or necessary at the end of the chain.
- 2. polymeric alcohol
- 3. alcohol

If you play it wrong, you call it a protein because of redundancy and inference from reference sources.

and

MID IR WOULD HAVE LIKELY EASILY SETTLED THE AFFAIR BUT YOU DO NOT HAVE IT NOW.

Now the lesson from all this is to back up and review what you know and what might be assumptions or bias influence.

Back to our list.

9. DONE Construct a UV & NIR plot of polyvinyl alcohol? (and we learned a great deal from it, didn't we...?)

1. Head toward replication of TSP 070 activity, namely nucleic acids (now very questionable) and bacteriophage (still seems likely).

- 2. Enzyme concentration studies. Enzyme isolation studies.
- 3. Mixing the culture into blood in various ways. Impact?
- 4. Keep sorting out the chemistry of the culture dynamics and progression
- 1. Native
- ---pl series investigation?
- 2. Decant
- 1. pl series investigation?
- 2. Decant progression- transform
- pl series investigation
- 5. . Precipitate
- 6. Surface layer synth cells polymer
- 7. NIR of Human Blood vs Synth layer sort this out alcohol?
- 8. Duplicate TSP 070
- 1. Nucleic acids
- 2. Bacteriophage

The general sequence coming up is:

- 1. Nucleic acids, bacterophage
- 2. Health Lineage
- 3. Panel on:
- 1. Culture
- 2. Mitigation

Three good problems:

1. Try now to settle out the nucleic acid -protein issue. 190nm should also help.

2023.11.22 Solubility-Health-Blood(Syn)

- 2. NIR human blood -synth later comparison (sort out confusion there)
- 3. Bacteriophage proposition

Let's revisit this human blood - synthetic blood issue.

Microscope settings that seem improved or best at 3200x.

- 1. Only refers to screen center
- 2. Condenser set at about 1/2 of eyepiece, eg. 40x = .65 set at .32
- 3. Light diaphram set in lower range eg 2-7
- 4. Exposure about 1/4
- 5. Gain looks to be close to 0.
- 6. White balance about 1/6
- 7. Tint about 40%
- 8. Hue 45
- 9. Saturation 30
- 10. Brightness 90
- 11. Contrast 95
- 12 Gamma 100

Fine adjustment of exposure with diaphragm.

I did try, for the first time, to withdraw fluid from the sac of fluid that builds up in my lower feet. Very shallow needle entry, at most 1/8 inch. Was not able to withdraw any fluid. Small drop of blood appeared on surface after withdrawal. Looked under scope at blood, highly variable results, slide preparation not standard so will hold off judgement. Definite CDB presence, however.

In weeks past, I was able to evaluate a MID IR blood sample of mine I had taken in Nov of 2015. As I recall, I was not able to identify any significant abberation in the functional groups expected between 3826 and 1536 cm-1 (disregarding 1536-600 fingerprint region). It would certainly be of value to have MID IR again but that is also quite a luxury financially. A true reference for blood has been lost by now. The fingerprint region would be a very important part of the picture to analyze. This comparison with human blood reference spectra was made on Oct 31 2023 in handwritten notebook Vol 33. I actually have close to 100 pages of handwritten notes in addition to these digital notes that were started on Oct 30 2023. So there is much lab work that is not written until the major switchover on Nov 16 2023. Also a comparison was made in hand notes on Nov 03 2023.

I have been trying to relocate my notes about the solubility tests on the polymer-synth layer. They took place on Nov 04 2023 hand notes. OK, result was that they polymer-synth layer is soluble in strong H2SO4, NOT KOH-NaOH. Also in acetone to some degree. Projection was made that this will be useful for LC work later on.

Now here is where the conflict is arising. You are saying that you identified that the blood clot dissolves over time in KOH-NaOH. You are also saying apparently that the blood clot did not dissolve in H2SO4.

But now on Nov 04 2023 you are saying explicitly that the polymer-synth layer only dissolved in strong H2SO4 and possible acetone, but definitely did not and does not dissolve in KOH-NaOH.

THIS IS A HUGE DIFFERENCE AND A BREAK ON LOGIC. The presumption is that the polymer-synth layer and the blood clot would be subject to a similar breakdown mechanism, i.e., both dissolve in lye or both dissolve in acid. These notes say this is not the case.

THIS IS A BIG FLAG AND THIS NEEDS TO BE EXAMINED PRONTO ...

Here are some other notes from Nov 18 (digital notebook) that are relevant:

"These are helpful results. It has not been published yet, however, the deceased blood clot was eventually dissolved in NaOH-KOH solution over a period of ~ 2 months. This was recorded on Aug 11 and Aug 13 2023 in Vol 30 of the laboratory notebooks.
This now becomes important as we have similar solubity behavior between the deceased blood clot (the most dense and developed, rubber polymer properties) and the borax polymer that has been developed. We know that KOH-NaOH has generally been our only successful reagent for dissolution of any of the filaments. We know they help to break down the disulfide bonds, as in hair."

So look at the above and what it is saying. It is saying the polymer-synth layer dissolves only in H2SO4 directly (and maybe acetone) and NOT base, but then when the borax polymer is formed IT dissolves in base. This is getting very curious. The borax polymer is formed by combination of borax and TSP 100.5 (hand notes on Nov 15 2023). The polymer formed is called 100.10. 100.5 is the decant, NOT the polyjmer-synth layer.

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OK we have 2-3 different compounds going on here reacting with acid and base. Let' sort these out:

100.5 is the decant.

100.10 is the borax polymer (made with borax combined with 100.5)

The clot dissolves in KOH-NaOH slowly.

The borax polymer (using 100.5 and borax) disslolves in KOH-NaOH.

The polymer-synth layer dissolves in Strong H2SO4 (and maybe acetone) but NOT in KOH-NaOH.

Also I am recalling the decant forms a precipitate when combined with a carbonate. See those questioning notes also - what if they do not dissolve in the body as well?

This says that the polymer-synth layer and the borax polymer are two very entirely different things. The borax polymer is apparently behaving closer to the clot in terms of acid-base behavior vs the borax polymer and the polymer-synth layer.

Think about this: We know that the decant mixed with borax creates a polymer. This polymer is very hard to dissolve, requiring lye.

Now you also tried an experiment with carbonate and the decant. You used washing soda (sodium carbonate). Apparently you wanted to see what would happen if it was placed in a base solution? It would be great to recall

what you motive was that started that process. The point here is that it created a precipitate. So the question is how soluble is that precipitate? How do you know it does not build up in the body as well? What is that precipitate made of, other than to presume it is a carbonate form. Well, what are kidney stones? I suspect calcium carbonate....

There are potentially some serious issues with any use of carbonates, such as baking soda, washing soda. These are both carbonates, sodium as well. Could easily bring too much sodium into the body. It would only be able to be used sparingly. Think about this:

Decant + carbonates are producing a precipitate. (baking soda) Decant + borax is producing a polymer.

Neither one of these is desired in the body. Watch out - health risks here. Presents a dilemma when trying to increase alkalinity in the body.

Excess of baking soda can also cause edema, salt imbalance here.

"In many species the pancreatic duct epithelium secretes HCO3- ions at a concentration of around 140 mM by a mechanism that is only partially understood."

You can work with this concentration amount and equate it to baking soda ingestion.

Let's look at our NIR plot we made comparing synth layer to human blood:

Human blood has (all statements will be relative in this study) high absorbance in the 900-950 nm region relative to polymer-synth layer. Synth refers to synthetic blood detected within the polymer layer. Synth layer is white and insoluble except in lye and possible partial in acetone.

CRC 900-950 nm is dominated by the methy groups and methylene. I presume this is the fatty acids in blood.

On Oct 31 and on Nov 03 2023 I compared human blood Mid IR data with reference data. I had a match with all groups on my Mid IR spectra with the reference blood on the upper portion of the spectrum. Lipids, or alkanes are a part of that. So I do not see a problem here with normal blood absorbing in this region. The NIR spectra of the polymer-synth layer, however, shows very low relative absorbance here. This is our first major difference.

The next place we show NIR absorbance in blood is at 1524 nm, and it is relatively low. The 70% level of the peak spans from about 1500-1550nm. Our closest match here is 1520 which has amide by CRC and this also would be a match to human blood.

This is all we get out of the human blood NIR spectrum and it remains a match with expectations, as the MID IR plot of 2015 does. So no real surprises here.

However, when we look at out polymer-synth layer we have an entirely different story. It has a very strong peak at 1520 which matches both amide and POLY amide. But very high relative to human blood. In addition we have a very strong broad peak centered about 1604. This is in the polyamide vinyl region (1598-1613) CRC.

Also at 1700 we have higher methyl absorbance (even though we cannot see peak) than the polymer synth layer. This is also consistent with the 900-950 nm region findings.

Conclusion: for polymer-synth layer relative to human blood Likely to have methyl disruption (i.e., decrease in fatty acids?) Likely to polymerization of amides taking place Likely to have vinyl groups active

Fatty acids have many health benefits, especially w.r.t. cardiovascular disease, cell membrane health, etc. Excess baking soda probably not a good idea, fish oils may be. VIt B complex, enzyme research, etc. Acidity problems remain.

Would be helpful to look for methyl disruption in human samples.



2023.11.23 Blood(Syn)-Polymer Compare

Forefront topics are:

- 1. Clot solubility conflict : polymer synth layer vs clot solubility
- 2. Comparison between human blood and synth layer
- 3. Bacteriophage prospect

These are major topics.

Lots of issues coming up as usual.

Carbonate precipitation with decant is now raising some important questions. Carbonate ions may be a problem, e.g, baking soda. Also excess sodium with baking soda another problem. Also the false borax promotion issue. Look like significant health issues with any precipitate or polymer that forms. How would you know they would dissolve - polymers not, and carbonates only in acids in general. You need to find out the nature of the precipitate with sodium carbonate added to the decant.

Is there a magnesium carbonate? Yes, there most certainly is. Can cause diarreaha but could be a candidate. Mg citrate would also double up on the magnesium ion, so need to watch that on intestinal fortitude... Magnesium carbonate has poor absorption, it is stated. Too much magnesium in the blood dangerous.

Need to run a culture control with magnesium citrate alone.

Right dosage is likely important. Edema taking place in right foot on afflicted ankle/leg at night. Could be salt imbalance in connection with affliction.

Alka selter tablet has about 1 gm citric acid, 2 gms baking soda [~2.5 mm in vial) (and aspirin) Mg citrate has 100 mg Mg pink salt

AM urine pH 5.3- 5.6. Too low. Want to get this up to 6.5 or so with minimum addition. Also increase water intake. Body temp running avg. 97.5, not too bad. 96.5 is. 98 better.

Enzymes Vit B, C

Human blood-synth layer comparison to continue.

Mid IR exploration of vinyl group with historical "CDB Protein Complex" IR data (cerca 2015) looks to be most important. Initial peak match search brings up following candidates as:

1. Butyl vinyl ether C6H12O

2. Cyclohexyl vinyl ether C6H14O

These candidates have essentially exact matches with IR collected Sep 2015 of "CDB Protein Complex". Out of about 10 candidates from entire SDBS database. These are lowest molecular weight. I am guessing we have something here.

Look at physical properties, synthetic rubber, etc.

Combination of human-synth blood comparison with polymerization probable is a worth topic.

Let's get back to the blood comparison.

Recalling from yesterday:

Conclusion: for polymer-synth layer relative to human blood:

1. Likely to have methyl disruption (i.e., decrease in fatty acids?)

2. Likely to polymerization of amides taking place

3. Likely to have vinyl groups active

2023.11.23 Blood(Syn)-Polymer Compare

and

Would be helpful to look for methyl disruption in human samples. and always allow for error and mistakes!

Main mitigation strategies are:

- 1. Interfere with protein development
- 2. Interfere with polymerization
- 3. Interfere with acidity
- 3. Support methylation
- 4. Disrupt polymer structure within and below skin

4. and the usual of everything before, antioxidation, etc.

5. and now added on Dec 05 2023 the new information coming to light on acidic amino acids and pl resarch on blood proteins.

We also learned some very important acid-base properties and that things are behaving quite differently:

OK we have 2-3 different compounds going on here reacting with acid and base. Let' sort these out:

100.5 is the decant.

100.10 is the borax polymer (made with borax combined with 100.5)

The clot dissolves in KOH-NaOH slowly.

The borax polymer (using 100.5 and borax) dissolves in KOH-NaOH.

The polymer-synth layer dissolves in Strong H2SO4 (and maybe acetone) but NOT in KOH-NaOH.

This says that the polymer-synth layer and the borax polymer are two very entirely different things. The borax polymer is apparently behaving closer to the clot in terms of acid-base behavior vs the borax polymer and the polymer-synth layer.

Now let's look at UV for both human blood and the synth layer. Also look at the synth layer again under the scope.

The UV spectrum of blood is far more interesting than I would ever have imagined. It is the most active UV spectrum I have encountered thus far. Busy all the way across from 190 nm to 400 nm. Here are peaks:

Human Blood : 232 (230) 275 (270, 280) 346 (350) ~400+(another peak developing)

We know 230 is commonly associated with protein peak folding. 275 would be classic protein area. 346 unknown now.

And here is interesting info, but it does not match our finding. We have a strong peak at 345, 346, not 365.

"Journal of Forensic Sciences J Forensic Sci. 2020 Sep; 65(5): 1716 1721. Published online 2020 Apr 28. doi: 10.1111/1556-4029.14439

Ultraviolet 365 as an Alternative Light Source for Detection of Blood Serum

These results show that ultraviolet (UV) at 365 nm (UV365) was effective in the detection of even small amounts of blood plasma and serum, compared with UV at 395 nm, which was not. UV365 was also found to be useful in distinguishing blood imprints from clotting blood which had been transferred to material versus blood that had been added directly. Taken together, these results demonstrate that UV365 may be utilized as a simple, nondestructive method for blood serum detection."

Now the difference alone is what of greatest interest to me. I have a very clear strong peak at 345 nm, not 365. 365 is closer to the bottom of a valley for me. So why this difference? What is 345 measuring and what is 365 measuring? I could not imagine using 365 right now compared to what I am seeing at 345.

I have found a good lead on interpretation of UV of blood (in this case it is mouse blood):

"UV-Visible absorption spectra of blood and EB diluted in water. A. Five microliters of a BALB/c mouse blood

were drawn and diluted to 250 m L (1:50 dilution) with distilled water, centrifuged and the supernatant collected for analysis. Indicated peaks are: 1) protein amide backbone and nucleic acids (220 nm); 2) proteins with chromophoric amino acids and other small chromophoric molecules (280 nm); 3) globin-heme interaction (340 nm); 4) soret band (420 nm); 5) oxyhemoglobin b -band (, 540 nm); and 6) oxyhemoglobin a -band (, 575 nm). Arrows show the common wavelength to measure hemoglobin (540 nm) and EB (620 nm). doi:10.1371/journal.pone.0110551.g001

https://www.researchgate.net/figure/UV-Visible-absorption-spectra-of-blood-and-EB-diluted-in-water-A-Five-microliters-o

So we have the following:

peaks are:

1) protein amide backbone and nucleic acids (220 nm);

- 2) proteins with chromophoric amino acids and other small chromophoric molecules (280 nm);
- 3) globin-heme interaction (340 nm);
- 4) soret band (420 nm)

and ours measured:

Human Blood : 232 (230) 275 (270, 280) 346 (350) ~400+(another peak developing)

So we have:

1. A very general match at 220-230 nm region (also note nucleic acid in addition to protein reference). (remember mouse blood)

2. A decent acceptable match at 275 - 280 nm for proteins, although some shift (remember reference here is mouse blood)

3. A decent acceptable match at 340 - 345 nm. (remember mouse)

4. Notice our external match quite likely developing at ~400 nm.

So this is quite decent and gives us some acceptable UV references for blood.

Now let's start looking at the synth layer.

I have some very good photos of the synth cells under the scope. It takes some doing. First step is that slide needs to be mildly flushed with water to clean up the surrounding polymer solution. Then the best results will be obtained as the slide dries from the water under the cover slip. Most cells will congregate and not make good viewing however some stray cells will provide a good view.

The microscope at 8000x is working best at:

1. Height of stage q tip height with one end cut off. This step is also critical. 1/8 inch change seems to affect greatly.

- 2. Brightness of lower dial lamp will have moderate impact, usually between 5-10.
- 3. The diaphragm is set at 0.3 (1/4 of what the objective says)
- 4. Exposure Timed 40%
- 5. Gain 20%
- 6. Color temp 20%
- 7. Tint 45%
- 8. Hue 45%
- 9. Saturation 30%
- 10. Brightness 95%
- 11. Contrast 100%
- 12. Gamma 100%

I also have a good macro picture of the polymer synth layer on a glass slide. So good, we have

1. UV of human blood, very distinctive.

2. Microphotos of synth blood. Easy to compare to human blood photos. 8000x.

3. Macro photo of synth polymer layer.

4. We have a NIR comparison that demonstrates the polyamide/vinyl nature of the synth blood. Likely methylation disturbance at play.

- 5. Mid IR of vinyl groups is coming into play
- 6. Macro photo or culture dish surface layer.
- 7. UV comparison spectra between human blood and synthetic blood, high correspondence.

8. Photo of "blood red" color change upon mixing polymer surface layer and strong H2SO4

also recall kastle meyer, hemoglobin, and pl determination.

A couple more phenomenall things have happened.

1. When you take the dried synth material and put in in strong H2S04, it turns immediately deep red. Slight purple tint. Hard to avoid the situation with blood. I have photo.

2. In addition, there is significant overlap in the UV spectrum with human blood, especially from 190 nm to 300 nm. Peaks, valleys and general profile coincides as well.







CI Laboratory Notebook Vol XXXIII : Appendix (Digital Format)



NIR TSP 070 Polymer-Synth Cells vs Polymer-Synth Cells in H2SO4 Carnicom Institute Nov 2023 Normalized



2023.11.24 Abstracts AI - NIR Synth

I have done a good job today of learning to prepare abstracts for all papers of 2023. ChatGBT is integrated withn a newer version of my word processor. I have given it a run, and it works quite well. BIggest difficulty was getting a suitable format for intergration within Wordpress and Substack, not the generation of the abstracts.

Process is:

1. An Abstract database has been made. Can now be organized by title, date, link, abstract. Very helpful.

2. Output will come from the database itself. Convert file to HTML (great option available) and make sure it will present on a portrait view ok.

3. This html can be loaded with Firefox from the hard drive. Firefox seems to be superior to Brave in several respects.

- 4. It can then be uploaded into both Wordpress and Substack and works quite well on both.
- 5. Last step is to attach the links recorded in the database to the titles of the papers.

Steps of generating abstract are generally:

- 1. Copy the paper into the Starmaker wordprocess (new and latest greatest version with ChatGBT integrated.
- 2. Choose the summarize text option, mid length.
- 3. Copy the summary into the database, with any minor changes required.

A really helpful and valuable addition. Still took lots of work but should be even a little smoother next time. Suggest we work on one year at a time.

OK, we are continuing to look at a comparison between the human blood and synth blood.

1. I am curious, how much H2SO4 must be used to generate the red color?

2. Why did this material not evaporate or dry on the ceramic plate, even after overnight? It has unusual volatility properties.

3. We have NIR plots of the polymer vs the acidified red version of the polymer - synth layer.

It is very difficult to produce the purple - red color from the polymer. The color is actually a rich deep purple. It requires extremely concentrated sulfuric acid to produce it. Not sure yet if the heat generated in the acid-water reaction is a part of or requirement in the process. It requires about 1 ml of full strength H2SO4 with approx. 1 ml of water. I am not sure if drying the polymer-synth layer assists the process or not, I have added both dried and fresh polymer. It is a remarkable color.

So two properties are now unusual:

1. Deep rich purple, or purple red color in concentrated sulfuric acid.

2. The material seems very difficult to dry and/or vaporize.

The polymer does seem to have a very stout vinyl type quality as it is dried.

"In most cases, purple blood is caused by a rare condition called sulfhemoglobinemia, where sulfur atoms bind to the iron in hemoglobin. This prevents hemoglobin from carrying oxygen effectively, causing a purplish hue to the blood."

"Bacterial Infections

Certain bacterial infections, like endocarditis or sepsis, can also turn blood purple. Bacteria produce sulfur-containing compounds that bind to hemoglobin, inducing sulfhemoglobinemia."

"Sulfhemoglobinemia is a condition in which sulfhemoglobin, a molecule formed when sulfur binds to hemoglobin, is present in the blood. This molecule gives the blood a purple color. Normally, hemoglobin carries oxygen throughout the body, and it has a red color when oxygenated and a darker red color when deoxygenated.

However, when sulfhemoglobin forms, it changes the conformation of hemoglobin, altering its ability to bind and release oxygen. This leads to a decrease in oxygen-carrying capacity and a change in the color of the blood. The purple color is a result of the interaction between the sulfur and the iron in the hemoglobin molecule."

H2SO4 added to human blood turns it brown, which is expected. It is oxidizing the iron.

However, H2SO4 added to the polymer turns it purple to purple red.

"When sulfuric acid is added to blood, the resulting color change can vary depending on several factors. The color change is primarily due to the reaction between the acid and the components present in the blood.

If the blood contains a high concentration of hemoglobin, which is responsible for carrying oxygen in red blood cells, the reaction with sulfuric acid can result in a purple color. This occurs because the acid causes the oxidation of hemoglobin, leading to the formation of a purple-colored compound called hematin.

On the other hand, if the blood has a lower concentration of hemoglobin or if other substances are present, such as bilirubin or other breakdown products of red blood cells, the reaction with sulfuric acid can result in a brown color. This is because the acid can react with these substances, leading to the formation of brown-colored compounds.

It's important to note that the color change observed when sulfuric acid is added to blood may also be influenced by the concentration and purity of the acid used, as well as the specific characteristics of the blood sample. Therefore, variations in the color change can occur in different situations."

"When blood is exposed to sulfuric acid, it undergoes a chemical reaction known as a "deoxyhemoglobin-sulfuric acid reaction." This reaction causes the blood to turn purple.

Sulfuric acid is a strong acid that can denature proteins, including hemoglobin, which is responsible for carrying oxygen in our blood. Hemoglobin normally carries oxygen in its oxygenated form, which gives blood its bright red color. However, when sulfuric acid comes into contact with hemoglobin, it breaks down the structure of hemoglobin and converts it into a different form called deoxyhemoglobin. Deoxyhemoglobin has a darker color, appearing more purple or maroon."

"Artificial blood, also known as synthetic blood or blood substitute, refers to a substance that can temporarily fulfill the functions of natural blood in the human body. It is primarily developed to address the shortage of donated blood and to provide an alternative in emergency situations.

There are two main types of artificial blood: oxygen-carrying solutions and oxygen-carrying particles.

1. Oxygen-carrying solutions: These solutions are designed to carry and deliver oxygen to tissues, similar to the red blood cells in natural blood. They are typically composed of hemoglobin-based molecules or perfluorocarbons. Hemoglobin-based solutions use modified hemoglobin molecules derived from human or animal sources, while perfluorocarbons are synthetic compounds that can dissolve and carry oxygen.

2. Oxygen-carrying particles: These particles are microscopic in size and are engineered to transport oxygen. They can be made from various materials, such as lipids, polymers, or nanoparticles. Oxygen-carrying particles are designed to mimic the oxygen-carrying capacity of red blood cells.

Artificial blood has several potential advantages over donated blood. It can be produced in large quantities, ensuring a consistent supply. It can also be stored for longer periods and does not require blood typing or cross-matching, reducing the risk of transfusion reactions. Additionally, artificial blood is free from infectious agents that can be transmitted through donated blood.

However, the development and approval of artificial blood have faced numerous challenges. One major hurdle is replicating the complex functions of natural blood, such as regulating blood pressure, clotting, and immune responses. Ensuring the safety and efficacy of artificial blood has also been a significant concern.

While there have been advancements in the field of artificial blood, it is important to note that it is not yet widely available for clinical use. Extensive research and rigorous testing are still ongoing to overcome the existing limitations and bring artificial blood to the forefront of medical practice."

"When it comes to blood, bacteria and synthetic biology can also play a role. For instance, bacteria can be engineered to produce specific proteins or enzymes that could be used in blood-related applications such as clotting factors or blood substitutes. Synthetic biology techniques can also be employed to modify blood cells or create artificial blood components for various medical purposes."

An interesting one:

"The Carnicom Institute is a non-profit research organization that focuses on the study of geoengineering, aerosol operations, and related health issues. It was founded by Clifford E. Carnicom in 2008 and is based in New Mexico, United States. The institute conducts independent scientific research and provides educational resources to raise awareness about the potential risks associated with geoengineering activities.

Cross-domain bacteria, also known as "Morgellons," is a term used to describe a condition where individuals experience unusual skin sensations and the presence of fibers or filaments embedded in their skin. Some individuals believe that these fibers are a result of a new type of bacteria that can infect both humans and the

environment. However, it is important to note that the existence and nature of cross-domain bacteria are still a subject of debate and controversy within the scientific community. Further research is needed to fully understand this phenomenon."

NIR of synth polymer layer: 1196 Methyl (1194, 1195) weaker peak "Methyl groups typically exhibit absorption peaks in the near infrared (NIR) region around 1196 nm."

1521 Amide/Protein (1520) Secondary Amine (1520) Polyamide (1515) "In the near infrared region, the amide bonds exhibit specific absorption bands around 1521 nm. By measuring the intensity of the absorption at this wavelength, NIRS can provide information about the presence and concentration of proteins in a sample."

1581 Alcohol (1580) alkyl alcohol (1583) "The wavelength you mentioned, 1581 nm, falls within the near-infrared range and is commonly used in NIRS for alcohol analysis. At this wavelength, alcohol molecules absorb light in a characteristic manner, allowing for the quantification of alcohol concentration.

1609 Vinyl (1613) Polyamide (1618) "In the case of vinyl, NIRS can be used to study its absorption properties in the near infrared range, specifically at a wavelength of 1609 nm"

Overall, the use of NIRS at 1609 nm can help in characterizing vinyl and understanding its absorption behavior in the near infrared range."

1580-1609 Broad Peak

This is saying that we have a protein/polyamide vinyl, alkyl alcohol dominanting the composition. This would all be consistent with solubility and physical properties. We seem to be on track quite well here.

Now for the polymer synth layer in STRONG H2SO4 (Red-purple color) non volatile -sulfhemoglobinemia (hemoglobin is a part of the reaction).

1440 Methylene (1440) aliphatic hydrocarbons, OH Aromatic (1420), OH Phenolic (1420)

Work on this with physical propeties, UV

2023.11.25 Human Synth Blood Compare

I am working on developing a comparison between human blood and the synth blood-polymer layer. Let's recap the factors that have surfaced:

FROM PREVIOUS DAYS:

From NIR Analysis: Conclusion: for polymer-synth layer relative to human blood:

- 1. Likely to have methyl disruption (i.e., decrease in fatty acids?)
- 2. Likely to polymerization of amides taking place
- 3. Likely to have vinyl groups active

"UV-Visible absorption spectra of blood and EB diluted in water. A. Five microliters of a BALB/c mouse blood were drawn and diluted to 250 m L (1:50 dilution) with distilled water, centrifuged and the supernatant collected for analysis. Indicated peaks are: 1) protein amide backbone and nucleic acids (220 nm); 2) proteins with chromophoric amino acids and other small chromophoric molecules (280 nm); 3) globin-heme interaction (340 nm); 4) soret band (420 nm); 5) oxyhemoglobin b -band (, 540 nm); and 6) oxyhemoglobin a -band (, 575 nm). Arrows show the common wavelength to measure hemoglobin (540 nm) and EB (620 nm). doi:10.1371/journal.pone.0110551.g001

https://www.researchgate.net/figure/UV-Visible-absorption-spectra-of-blood-and-EB-diluted-in-water-A-Five-microliters-of-blo

So we have the following:

peaks are:

protein amide backbone and nucleic acids (220 nm);
 proteins with chromophoric amino acids and other small chromophoric molecules (280 nm);
 globin-heme interaction (340 nm);

4) soret band (420 nm)

and ours measured:

Human Blood : 232 (230) 275 (270, 280) 346 (350) ~400+(another peak developing)

So we have:

1. A very general match at 220-230 nm region (also note nucleic acid in addition to protein reference).

(remember mouse blood)

2. A decent acceptable match at 275 - 280 nm for proteins, although some shift (remember reference here is mouse blood)

3. A decent acceptable match at 340 - 345 nm. (remember mouse)

So this is quite decent and gives us some acceptable UV references for blood.

Now let's start looking at the synth layer.

I have some very good photos of the synth cells under the scope.

I also have a good macro picture of the polymer synth layer on a glass slide. So good, we have

1. UV of human blood, very distinctive.

2. Microphotos of synth blood. Easy to compare to human blood photos. 8000x.

3. Macro photo of synth polymer layer.

4. We have a NIR comparison that demonstrates the polyamide/vinyl nature of the synth blood. Likely methylation disturbance at play.

5. Mid IR of vinyl groups is coming into play

6. Macro photo or culture dish surface layer.

7. UV comparison spectra between human blood and synthetic blood, high correspondence.

8. Photo of "blood red" color change upon mixing polymer surface layer and strong H2SO4

also recall kastly meyer, hemoglobin, and pl determination.

9. MID IR correlation with non fitting albumin peaks.

A couple more phenomenall things have happened.

1. When you take the dried synth material and put in in strong H2S04, it turns immediately deep red. Slight purple tint. Hard to avoid the situation with blood. I have photo.

2. In addition, there is significant overlap in the UV spectrum with human blood, especially from 190 nm to 300 nm. Peaks, valleys and general profile coincides as well.

It is very difficult to produce the purple - red color from the polymer. The color is actually a rich deep purple. It requires extremely concentrated sulfuric acid to produce it.

So two properties are now unusual:

1. Deep rich purple, or purple red color in concentrated sulfuric acid.

2. The material seems very difficult to dry and/or vaporize.

The polymer does seem to have a very stout vinyl type quality as it is dried.

"In most cases, purple blood is caused by a rare condition called sulfhemoglobinemia, where sulfur atoms bind to the iron in hemoglobin. This prevents hemoglobin from carrying oxygen effectively, causing a purplish hue to the blood."

"Bacterial Infections

Certain bacterial infections, like endocarditis or sepsis, can also turn blood purple. Bacteria produce sulfur-containing compounds that bind to hemoglobin, inducing sulfhemoglobinemia."

"When blood is exposed to sulfuric acid, it undergoes a chemical reaction known as a "deoxyhemoglobin-sulfuric acid reaction." This reaction causes the blood to turn purple.

Sulfuric acid is a strong acid that can denature proteins, including hemoglobin, which is responsible for carrying oxygen in our blood. Hemoglobin normally carries oxygen in its oxygenated form, which gives blood its bright red color. However, when sulfuric acid comes into contact with hemoglobin, it breaks down the structure of hemoglobin and converts it into a different form called deoxyhemoglobin. Deoxyhemoglobin has a darker color, appearing more purple or maroon."

NIR of synth polymer layer: 1196 Methyl (1194, 1195) weaker peak "Methyl groups typically exhibit absorption peaks in the near infrared (NIR) region around 1196 nm."

1521 Amide/Protein (1520) Secondary Amine (1520) Polyamide (1515) "In the near infrared region, the amide bonds exhibit specific absorption bands around 1521 nm. By measuring the intensity of the absorption at this wavelength, NIRS can provide information about the presence and concentration of proteins in a sample."

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Overall, the use of NIRS at 1609 nm can help in characterizing vinyl and understanding its absorption behavior in the near infrared range."

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This is saying that we have a protein/polyamide vinyl, alkyl alcohol dominanting the composition. This would all be consistent with solubility and physical properties. We seem to be on track quite well here.

Now for the polymer synth layer in STRONG H2SO4 (Red-purple color) non volatile

1440 Methylene (1440) aliphatic hydrocarbons, OH Aromatic (1420), OH Phenolic (1420)

Work on this with physical propeties, UV

Human blood-synth layer comparison to continue.

Mid IR exploration of vinyl group with historical IR data (cerca 2015) looks to be most important. Initial peak match search brings up following candidates as:

1. Butyl vinyl ether C6H12O

2. Cyclohexyl vinyl ether C6H14O

These candidates have essentially exact matches with IR collected Sep 2015 of "CDB Protein Complex". Out of about 10 candidates from entire SDBS database. These are lowest molecular weight. I am guessing we have something here.

Look at physical properties, synthetic rubber, etc.

Combination of human-synth blood comparison with polymerization probable is a worth topic.

Therefore we have quite a bit of information to work with. Our main topics covered are:

- 1. UV Spectrums compared
- 2. NIR spectrums compared
- 3. Expected impact upon blood
- 4. Nature of vinyl
- 5. Red color of polymer in H2SO4, Photos
- 6. Photographs under scope synth cells
- 7. Polymer macro photographs
- 8. We have reference MID IRs for both human blood and the "protein complex" from cerca 2015

Major removal information related to this reaction (sulfuric acid + blood) has been scrubbed from AI Chat and YouTube after original inquiries 24 hours earlier. Any relationships to use of sulfuric acid in the reaction have been removed.

So the conclusion is because this reaction is occurring with the synth polymer layer it is another level of confirmation of the existence of hemoglobin within the synth polymer layer. Scrubbing of information looks to be pretty clear on this research.

Now, is there anything more that we can learn with the equipment and means that we have?

1. What are the vinyl groups in MID IR and is ther any sign of it within our historical MID IR blood spectra? 2. Can you find any signs of the problem in historical MID IR?

What should be the title of this paper?

Human Blood vs. Synthetic Blood : A Comparison & a VERY Strong Lead

In addition, the more detailed vinyl identification candidate is an important topic. Look into the type of polymerization with the specific vinyl under investigation.

"Cyclohexyl vinyl ether is used in the production vinyl chloride polymers. In fluoropolymer resins, it contributes solubility, adhesion and crosslinking making a highly durable coating."

"Cyclohexyl vinyl ether is an organic compound with the chemical formula C8H14O. It is a colorless liquid that is used as a monomer in the production of various polymers and copolymers. It can also be used as a solvent or as a reagent in organic synthesis. Cyclohexyl vinyl ether is known for its ability to undergo polymerization reactions via radical or cationic mechanisms."

"The vinyl group, which consists of a double bond between two carbon atoms, typically absorbs infrared (IR) radiation in the range of 1640-1680 cm⁻¹. This absorption is due to the stretching vibrations of the carbon-carbon double bond."

The bacteriophage is an important topic.

Electrochemical measurements of organic redox reactions? That would open up a whole new ballgame.

An important question arises. We now have a significant MID IR match level occurring between the "CDB Protein Complex" MID IR spectrum collected on Sep 05 2015 and the compound Cyclohexyl VInyl Ether involving 8 peaks. Quite strong.

Now we also have a strong match between CEC collected blood spectrum of Nov 07 2015 and the spectrum of albumin, the primary protein in blood plasma, recorded on the CDBS.

Now the question is, are there any peaks that show up on the CVE spectrum that also show up on the CEC blood spectrum that do not show up on the reference MID IR listing for human blood?

So the peaks on the Cyclohexyl vinyl ether spectrum that match the CDB Protein Complex of 09.05.15 are: 2927, 2858, 1739, 1641, 1456, 1377, 1177, 1040.

Now which of these peaks match CEC Blood Spectrum of 11.07.15? 2919, 2857, 1646, 1452 and 1389.

Now which of these peaks do not match my reference MID IR blood data of the notes of Nov 03 2023 (hand notes, Vol 33)? Also human blood reference work done on Oct 31 2023 (hand notes).

2919 cm-1 does not show a match yet.
2858 does show a match (lipids claimed).
1646 does show a match (amide I C=O claimed).
1452 does not show a match.
1389 does show a match (lipids and proteins claimed).
1042 does not show a match.

Now in human blood we are apparently seeing a NIR entry of 1628 nm. This corresponds to vinyl and vinylidene (1630, 1631) and this is a huge red flag that should now be there.

In addition, I am now to trace MID IR peaks of: 2919 cm-1, 1452 cm-1 and 1042 cm-1. and look for any pattern of ill fit shared between MID IR and NIR.

2023.11.26 Blood Synth Vinyl MID IR

I have an interesting observation regarding the purple color on the synth-polymer layer, and I need to recall what I did. I believe it was the following:

1.A small piece of the dried synth polymer layer (a tough vinyl sure fits the bill) in about a ml of H2O. Then I think a drop of conc. H2SO4, maybe two or three. No purple color change takes place under these conditions of a weaker H2SO4 solution. Put it in the frig because of something I remember reading regarding chilling inducing a purple reaction with blood and sulfur.

Remembered two days later to look at it. It is indeed purple, just as I have been able to achieve with very conc. H2SO4 at rooom temperature with the synth layer.

So I will be:

1. Looking at this more and for replication.

2. Try to find what I read regarding H2SO4, blood and chilling purple reaction.

3. Ask the question, this means that the vinyl compound/product would have a reaction similar to with blood and therefore share another characteristic of blood and hemoglobin interaction....It also means that sulfur interacts with the synth polymer without doubt, in a way that is similar to how sulfur can react with blood.

"Sulfhemoglobinemia is a rare condition characterized by the presence of sulfhemoglobin in the blood. Sulfhemoglobin is a form of hemoglobin that contains sulfur instead of iron in its heme group. This alteration in the structure of hemoglobin affects its ability to bind and transport oxygen, leading to a decrease in oxygen-carrying capacity in the blood.

The exact cause of sulfhemoglobinemia is not fully understood, but it is thought to be a result of exposure to certain chemicals or medications. Some known triggers include certain drugs like phenacetin, sulfonamides, and aniline dyes, as well as exposure to industrial chemicals and toxins.

Symptoms of sulfhemoglobinemia can vary but may include cyanosis (bluish discoloration of the skin), shortness of breath, fatigue, dizziness, and confusion. In severe cases, it can lead to tissue damage and organ dysfunction.

Diagnosis of sulfhemoglobinemia is typically made through a blood test that measures the levels of sulfhemoglobin in the blood. Treatment options are limited, and there is no specific cure for the condition. In most cases, the condition resolves on its own once the exposure to the triggering agent is removed. Supportive care, such as oxygen therapy, may be provided to manage symptoms and improve oxygenation.

It is important to consult with a healthcare professional for a proper diagnosis and management of sulfhemoglobinemia. They can provide personalized advice and guidance based on individual circumstances."

"Sulfhemoglobinemia is a rare condition in which there is excess sulfhemoglobin (SulfHb) in the blood. The pigment is a greenish derivative of hemoglobin which cannot be converted back to normal, functional hemoglobin. It causes cyanosis even at low blood levels.

It is a rare blood condition in which the ß-pyrrole ring of the hemoglobin molecule has the ability to bind irreversibly to any substance containing a sulfur atom.[1][2] When hydrogen sulfide (H2S) (or sulfide ions) and ferrous ions combine in the heme of hemoglobin, the blood is thus incapable of transporting oxygen to the tissues.

Presentation

Symptoms include a blueish or greenish coloration of the blood (cyanosis), skin, and mucous membranes, even though a blood count test may not show any abnormalities in the blood. This discoloration is caused by greater than 5 grams per cent of deoxyhemoglobin, or 1.5 grams per cent of methemoglobin, or 0.5 grams per cent of sulfhemoglobin, all serious medical abnormalities.[citation needed."

"Occurs when a sulfer atom binds to porphyrin ring of hemoglobin, resulting in permanent oxidation of iron to the ferric state, incapable of oxygen transport

Most often associated with phenazopyridine, dapsone, metoclopramide, sumatriptan

Also associated with industrial chemicals, including trinitrotoluene, hydroxyl amine sulfate, dimethyl sulfoxide, Hydrogen Sulfide"

"Sulfhemoglobinemia is a rare condition where the blood takes on a purple color due to the presence of sulfhemoglobin. Sulfhemoglobin is a compound formed when sulfur combines with hemoglobin, the protein responsible for carrying oxygen in red blood cells. This condition can occur as a result of certain medications, chemicals, or toxins, and it can lead to symptoms such as shortness of breath, fatigue, and cyanosis (bluish-purple discoloration of the skin). Treatment for sulfhemoglobinemia typically involves identifying and discontinuing the causative agent, and in severe cases, blood transfusion may be necessary. It is important to consult a healthcare professional for proper diagnosis and management of this condition."

It is a morbid condition.

Major removal information related to this reaction has been scrubbed from AI Chat and YouTube after original inquiries 24 hours earlier. Any relationships to use of sulfuric acid in the reaction have been removed.

So the conclusion is because this reaction is occurring with the synth polymer layer it is another level of confirmation of the existence of hemoglobin within the synth polymer layer. Scrubbing of information looks to be pretty clear on this research.

OK, there is some very clear evidence available of vinyl in the blood. RIght now it is from NIR, we will see if it exists in MID IR afterwards. If we look at our notes of Oct 31 2023 (handwritten) we have a clear entry of absorbance at 1627 nm within a human blood analysis. This is clearly indicated within the CRC reference as being of vinyl nature from 1621 - 1637. Five consecutive entries with no real competition except for acrylate at 1621. This is a sufficient entry to show crossover between synth polymer analysis and human blood.

OK, we are at the point now where we can provide direct evidence of vinyl influence on human blood via NIR. The method is to focus on human blood alone, not comparing it to anything, and look at with NIR. We also focus in on the vinyl region of 1627 nm and remove extraneous examinations. The evidence is quite strong now of the existence of vinyl groups, polymerization, synthetic blood, within human blood. This is from a human blood sample 2023, unvaccinated individual.

It now becomes less important, but the question arises, can you show evidence of vinyl presence in a human blood MID IR spectrum available in 2015? We know we can show evidence from the MID IR of a "CDB Protein Complex" MID IR spectrum of 2015 in combination with use of SDBS database.

"The typical wavelength range for infrared absorption of vinyl groups is around 1600-1650 cm^-1."

Caution: Amides also absorb in ths region!

"The amide absorption in the infrared region of human blood typically occurs at wavelengths between 1600 and 1700 cm-1. This absorption is mainly attributed to the stretching vibrations of the amide I and amide II bonds in proteins present in the blood." The

Now we definitely have absorption of the protein complex MID IR of Sep 05 2015 at 1638 cm-1. This confirms the likely existence of the vinly group detected within the cultures of 2015. Now what about in blood? We have absorption at 1646 but so does amide.

"The simplest vinyl compound is vinyl chloride (C2H3Cl)."

"An ether functional group is a functional group in organic chemistry that consists of an oxygen atom bonded to two alkyl or aryl groups. It is represented by the formula R-O-R', where R and R' are alkyl or aryl groups. Ethers are characterized by the presence of the oxygen atom in the molecule, which gives them unique chemical and physical properties"

"Ether absorption in the infrared region typically occurs in the range of 1000-1300 cm-1. This absorption is due to the C-O stretching vibration of the ether functional group. The exact position of the absorption peak may vary depending on the specific structure of the ether molecule."

Now, here is what we are seeing. The 2015 CDB MID IR shows strong absorption at 1038 cm-1. THis looks like it MAY correspond to ether.

Molecules that contain a C-O bond typically absorb infrared radiation at around 1038 cm-1. Ether, which has the chemical formula R-O-R', where R and R' can be any alkyl or aryl group, contains a C-O bond and can absorb infrared radiation at this frequency.

I have found the MID IR correlation that I am seeking. My spectrum with albumin matches EXTREMELY WELL except for 2 peaks that exist in the range of ethers and vinyls. 1298 and 1160 cm-1. SDBS will find that the closest match to these two peaks is ethyl vinyl ether, exactly the top candiate from the CDB Protein complex MID IR of 2015.

Poly(vinyl cyclohexyl ether) (PVChE) is a type of polymer that is derived from vinyl cyclohexyl ether monomers. It is often used in the field of biological materials due to its unique properties and potential applications.

PVChE is biocompatible, meaning it is non-toxic and does not cause harm to living organisms. This makes it suitable for use in medical and biological applications, such as drug delivery systems and tissue engineering.

PVChE can be easily synthesized and modified to achieve desired properties, such as controlled release of drugs or enhanced mechanical strength. It can also be combined with other polymers or materials to create composite materials with specific functionalities.

In addition, PVChE has good solubility in various solvents, allowing for easy processing and fabrication into different forms, such as films, fibers, and hydrogels. This versatility makes it a valuable material for a wide range of biological applications.

Overall, PVChE is a promising material in the field of biological materials due to its biocompatibility, tunable properties, and processability. Ongoing research and development in this area aim to further explore its potential applications and optimize its performance in various biological systems.

Health observation: Besides severe skin damage (best analogy is a year + long internal burn and bee sting (eg, formic acid)) that covers the entire lower leg, angle and foot) significant edema is in place. Swelling is aggravated further with any increase in salts such as sodium (sodium bicarbonate used in pH control is counterproductive it seems). The observation is that ultrasound applied diligently and with requisite pain at point of core pain reduces the swelling temporarily in that area so that leg is normally sized. US drives the fluid away, albeit only locally. It is already known from under the scope that US separates the blood from the polymer.



2023.11.27 Research Paper:Blood Compare

Keep refining the flow of the paper in progress: Human Blood vs. Synthetic Blood : The Path to the Blood Clot (revised title)

After intro, we are on the opening subject of microscopic comparison first.

This is the general flow outline of the research paper in progress:

"I also have a good macro picture of the polymer synth layer on a glass slide. So good, we have

- 1. UV of human blood, very distinctive.
- 2. Microphotos of synth blood. Easy to compare to human blood photos. 8000x.
- 3. Macro photo of synth polymer layer.
- 4. We have a NIR comparison that demonstrates the polyamide/vinyl nature of the synth blood. Likely methylation disturbance at play.
- 5. Mid IR of vinyl groups is coming into play
- 6. Macro photo or culture dish surface layer.
- 7. UV comparison spectra between human blood and synthetic blood, high correspondence.
- 8. Photo of "blood red" color change upon mixing polymer surface layer and strong H2SO4
- also recall kastly meyer, hemoglobin, and pl determination.
- 9. MID IR correlation with non fitting albumin peaks.

A couple more phenomenall things have happened.

1. When you take the dried synth material and put in in strong H2S04, it turns immediately deep red. Slight purple tint. Hard to avoid the situation with blood. I have photo."

In terms of lab work, running questions are:

1. How does the blood clot NIR compare to the poly-synth layer NIR? This would give important information about changes that could explain the solublility behavior (i.e., or the lack of it) This would lead to an increased understanding of the polymerization process.

2. I wonder what produced the purple color in the synth layer + H2SO4 in the refrigerator? I can't duplicate that yet.

3. The bacteriophage is a huge issue.

4. Might also want to revisit the nucleic acid prospect.

5. Additional enzyme and polymerization study. Methylation also continues. Mitigation pursuits.

Observation: The TSP 100.7D series in petri dishes does not seem to produce anything particularly unuusual. I think the motive for this series was an attempt to produce the polymer-synth layer assuming that increased surface area of petri dishes might be more beneficial. This did not show to be the case. Thus far, large VOLUME of culture COMBINED with large surface area seems to be the most favorable condition for production of the polymer synth layer. I now have two very productive cultures. Both were created with 6500 ml cultures that were concentrated down to about 1500 ml. The containers have approx a 7-8 inch diameter. There are the series TSP 070 and TSP 100 which are very productive with strong separation into 3 distinct layers. I will discard the TSP 100.7D petri dish series. This series came from Nov 12. I did decide to look under the scope. Synth cell production is most certainly there; just not anything of the magnitude of the larger culture volumes.

--

This is our most current status with scope work at 8000x: The microscope at 8000x is working best at:

^{1.} Height of stage q tip height with one end cut off. This step is also critical. 1/8 inch change seems to affect greatly.

^{2.} Brightness of lower dial lamp will have moderate impact, usually between 5-10.

^{3.} The diaphragm is set at 0.3 (1/4 of what the objective says)

4. Exposure Timed 40%
 5. Gain 20%
 6. Color temp 20%
 7. Tint 45%
 8. Hue 45%
 9. Saturation 30%
 10. Brightness 95%
 11. Contrast 100%
 12. Gamma 100%

Today, at 3200x, it seems as though these settings can hold. It does seem that exposure control is most precise howeve

Observation: US is positively decreasing the level of edema when it is applied at the core afflicted region of the lower leg. It drops the skin level down in swelling approx 1/4 or so, very noticeable. All consistent with known properties and behavior of the polymer - blood interaction under US energy. A level of pain accompanies the procedure, approx 15 min duration.

I am now in a position to start comparing the NIR of a blood clot to the NIR of the synth polymer layer. The main uncertainties of comparison are:

1. There is some ambiguity whether the clot comes from the deceased individual or a live individual (externally clotted). The bias is strongly towards the deceased clot which is favorable, however this will likely be repeated. 2. The trend is removed from the blood clot but not from the synth-polymer layer so no magnitudes of any kind can be considered.

With that being said, here is what we start to see. The main issue here involves similarity or difference in basic structure and solubility. One thing we do know is that the clot dissolves in NaOH and the synth polymer layer dissolves in strong H2SO4. So we know already they are very different from one another and yet the presumption is that they originate generally from the same source material. This means, since we knowe they have important differences, that comparison of the clot with other forms observed will be made.

NIR Comparison:

Start with the blood clot: Clot: 1130 (1130) 1374 (1370) 1443 (1440) `1575 (1570, 1580)

and then the synth-polymer layer: Synth-Polymer: 1195 (1190, 1200) 1520 (1500) 1594 (1590) Broad 1582-1609 (1580-1610) 1700 (1700)

NIR Comparison: Deceased Blood Clot vs Synth Polymer Carnicom Institute Nov 2023 Clot Trend Removed - Normalized



2023.11.28 Research Paper Video Audio

Carry forward:

Keep refining the flow of the paper in progress: Human Blood vs. Synthetic Blood : The Path to the Blood Clot (revised title)

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A couple more phenomenall things have happened.

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3. The bacteriophage is a huge issue.

4. Might also want to revisit the nucleic acid prospect.

5. Additional enzyme and polymerization study. Methylation also continues. Mitigation pursuits.

I want to start revisiting the bacteriophage prospect. This event was recorded entriely in handwritten notes of Nov 07 2023. It involved the titration of TSP 70 (original large volume culture initiated on Oct 19) with a 20 day old culture. It was apparently titrated from a level of 3.75 to 6.2. Along the way, it appears a pl of 4.31 was determined. The analyte was apparently the mid level (main volume) solution of the TSP culture, understanding that it separates over time into 3 separate layers:

- 1. Surface synth-polymer layer (white)
- 2. Main volume of culture solution (amber)
- 3. Bottom precipitate layer (not examined in depth to my recollection thus far)

Let's try to find this titration record to determine what the equilibrium point was. The equilibrium titration point was recorded at a pH of 5.26. I used 50:1 NaOH-KOH titrant and was recorded in the notes of Nov 06 2023. I carried the pH up to 6.2 therefore.

It is important to realize that this was a 20 day old culture. Notice the difference in pl from the determination of pl of a fresh culture. Fresh culture was 5.31 as I recall (human blood proteome match) whereas the 20 day culture main solution (i.e., some separation has taken place by then) had the pl of 4.31. Variation of pl w.r.t. time was discussed within the notes - this remains uncertain at this time.

Now the culture is approx. 39 days old so conditions are somewhat different. Interestingly enough, TSP 100 series is about 20 days old (started on Nov 08 2023) so we actually do have a secondary reference in the

matter.

What happens next is that you take a UV spectrum on the 6.2 remainder. At thiws point you see a strong peak showing up at 269 nm, apparently quite definite. It is research of this peak that leads to the bacteriophage issue. I think the issue is that proteins are undersood to to center on 280 nm. DNA understood to center on 260. Now tyrosine is know to be at 274 nm and this seems to be the lowest I can find for protein issues. It just seems too far out of range at 269 to place this in the protein category. I believe the peak was also pronounced and has a different profile than protein.

This also causes the nucleic acid issue to reemerge as that UV spectrum had an apparent peak at 259 nm. Problem there was that the peak was so broad that it was more difficult to isolate with complete confidence that it had no protein nature. But this too will eventually need to be revisited. But for now, we will continue to address the bacteriophage issue.

Do I still have the 269 nm sample available to study? Let's see. I have:

TSP 70.1D YES I HAVE IT HERE. see below. TSP 70.2 see below TSP 70.3 see below TSP 70.4 see below. TSP 70.5 see below. and a H2SO4 control.

and this is the information that I have recorded in the spreadsheet log. Good work here, I need this.

Nov 07/ 2023 TSP 70.5C (B) X.XX-1107 TSP 70.5 Centrifuged TSP 70.0 Bottom Layer TSP 70.5 Centrifuged TSP 70.0 Bottom Layer Have an interest in comparing this to TSP 70.2C (B) ?-1107. NIR comparison has been made, in notes for Nov 07.

Nov 07/ 2023 TSP 70.2C (B) X.XX-1107 TSP 70.2 Centrifuged Titrate Precipitate TSP 70 was titrated. 70.1 is decant, 70.2 is precipitate, then centrifuged This shows itself to actually be two layers. Presumed acidic as decant has ph delta from 3.75 to 6.20.

Nov 06/ 2023 TSP 70.4S (L) X.XX-1106 TSP 70.4 Solubilized Polymer-Synth Layer in Acetone Solubility tests on Polymer-Synth Layer of TSP 70.0 Acetone Success. No testing done whatsoever. Acetone control required. Presumption is that UV should also show peak at 260. Unproven - needs to be done.

Nov 06/ 2023 TSP 70.3A (S) Nov 07/ 2023 TSP 70.1D (M) 6.20-1107 TSP 70.1 Titrate Decant Titrated 20 day TSP 70 Culture Decanted This

is what shows to apparently contain bacteriophages. Big issue here. Absorbance is at 269nm. Matches bacteriophage UV profile extremely well. 700-1100 NIR of same sample shows the alkyl alchol w/no hydrogen bonding at 962nm.

I DO HAVE IT then. 70.3A is of interest for later as it is where the 259-260 peak shows up. But the 269 nm question looks like it requires replication.

Now this culture is dated Nov 07 2023. So this is when titration was done. THe culture was approx 20 days old. The culture is now approx 40 days old but we have it which is great. Now the difference in appearance of this culture from the original clear decant is that it has a synth polymer layer on top now and the solution is cloudy. So it has changed as we have seen before in the decant versions. It serve as a case of interest to see how the UV spectrum may have changed.

This sample size is the largest of my set and is approx 40 ml. The other residual samples are on the order of a few ml. I will extract from the mid level solution area. I may also be able to get pH information at some point, however, I do not wish to disturb or mix the synth-polymer layer.

Good records were helpful here. Impossible to retrace or recollect from memory.

We have another important advantage now in UV. We can record with confidence from 190 nm to 400 instead of 235 nm to 400 using the quartz cuvettes I now have in place. A much broader and better UV picture.

I have replicated the spectrum. As the plot is difficult to interpret because of the broad flat peak, I have used a differential approach. I have two peaks that at at the zero point (first once crosses, 2nd one asymptotic:

210.3 nm

257 nm

The center of the broad flat peak is approx 252 nm so thus far there is no way that it approaches the protein section. If anything we have to wonder again if we are in the 260 region for nucleic acids again.

Our notes of Nov 21 2023 are in some detail on this issue. It is somewhat problematic no matter how you go about it. You are considering:

- 1. protein
- 2. nucleic acids or DNA
- 3. bacteriophage

4. or none of these and something new.

Remember you did just LOOK at it under the scope and saw small crystals and apparently what seems to be protein.

molecule wavelength of maximum absorption (nm)

some more information to consider. Notice all alkenes, 1, 2, 3 bonds causess displacement like we are seeing. ethene 171

buta-1,3-diene 217 (UV NIST Spectrum does not seem to match this at all, they show a peak at ~291 nm. hexa-1,3,5-triene 258 (No UV spectrum available on NIST)

We have some problems here. Best we may have is NIR.

2023.11.30 Another Mitigation Prospect

I am recognizing the similarity between nylon and solubility of the synth-polyamide layer. They are both soluble in strong sulfuric acid and maybe not much else. Acetone maybe some? That suggests that they share the same characteristic of both being polyamides.

Strong research topic here....

Note this comment:

(Note: "Red cell membranes have a negative charge (zeta potential) that causes red cells to repel each other. In the presence of increased positively charged plasma proteins such as fibrinogen or immunoglobulins, the negative charge on the red cell surface is diminished, allowing red cells to stick together." Ref. hematology.org)

It would not be surprising if this ends up being quite important. Now start thinking about this. You have an isoelectric point of 5.33. This is when the charges are zero, i.e., electrically neutral. What charge would the protein have if it were at pH 7.4?

"The isoelectric point (pl) of a protein is defined as the pH at which the net charge of a protein molecule is zero. Accordingly, proteins are positively charged at a pH below their pI and negatively charged at a pH above their pI. The protein pI varies greatly from extremely acidic to highly alkaline values ranging from about 4.0 to 12.0. Hence, pI values have long been used to distinguish between proteins in methods for protein isolation, separation, purification, crystallization, etc. Amino acid composition of a protein sequence primarily defines its pI, based on the combination of dissociation constant (pKa) values of the constituent amino acids. Out of twenty common amino acids, two amino acids, aspartic acid, and glutamic acid, are negatively charged and three amino acids, lysine, arginine, and histidine, are positively charged at the neutral pH, as defined by their pKa values."

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8667598/

This looks like it has our answer. The pH of the culture is on the order of 3.8. The dominant pH of the human blood proteome is 5.3. The pH of the culture is lower than the dominant pI of the blood proteome. This means that the blood proteins are becoming more positively charged with the introduction of the CDB.

OK I have said it in the paper in progress:

"((Note: Red cell membranes have a negative charge (zeta potential) that causes red cells to repel each other. In the presence of increased positively charged plasma proteins such as fibrinogen or immunoglobulins, the negative charge on the red cell surface is diminished, allowing red cells to stick together. Ref. hematology.org)

The isoelectric point (pl) of a protein is defined as the pH at which the net charge of a protein molecule is zero. Accordingly, proteins are positively charged at a pH below their pI and negatively charged at a pH above their pI. The protein pI varies greatly from extremely acidic to highly alkaline values ranging from about 4.0 to 12.0. Hence, pI values have long been used to distinguish between proteins in methods for protein isolation, separation, purification, crystallization, etc. Amino acid composition of a protein sequence primarily defines its pI, based on the combination of dissociation constant (pKa) values of the constituent amino acids. Out of twenty common amino acids, two amino acids, aspartic acid, and glutamic acid, are negatively charged and three amino acids, lysine, arginine, and histidine, are positively charged at the neutral pH, as defined by their pKa values.

Ref. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8667598/

This looks like it has our answer. The pH of the culture is on the order of 3.8 (strongly acidic). The dominant pH

of the human blood proteome is 5.3. The pl of the CDB culture is determined at 5.33. The pH of the CDB culture is lower than the dominant pl of the blood proteome as well as lower than the CDB culture pl. This means that the blood proteins are being exposed to positively charged proteins with the introduction of the CDB.0. This would explain increased coagulation. CEC)"

This therfore provides an additional potential mitigation strategy. Increase intake of negatively charged proteins.

Mitigation prospects:

1. Protein disruption : enzymes

- 2. Methyl Disruption : Vit B, Fatty acids?
- 3. pH regulation (risks as well, e.g, sodium increase, polymerization, misinformation, blood pH vs urine ph)
- 4. Ultrasound
- 5. Negatively charge protein intake?
- 6. Antoxidants and the usual two dozen suspects
- 7. Balm, e.g., methyl salicylate

The blood proteome distribution (dominant pl of 5.3) is at:

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7159840/

"Blood plasma is a major fluid compartment in human body, with a narrow normal range of pH $(7.40\pm0.05)12$. Thus the average pH of human blood is in the middle of the trough observed for the theoretical pl distribution of blood proteome (Figure 1[A]). Whether such a coincidence is by chance or by design, the density of normal proteoforms in the pl region 7.4 ± 0.1 is very low (ca. 20 times lower than in the region around pl 5.3 ± 0.1)."

The mitigation idea would be to increase intake of proteins with a pH greater than the dominant pl of blood, which is 5.3.

"Accordingly, proteins are positively charged at a pH below their pl and negatively charged at a pH above their pl."

We need the latter part of this sentence. Negatively chared amino acids are:

Aspartic acid pKa = 3.7 pl = 2.87Glutamic Acid pKa = 4.1 pi = 3.08

This means that these proteins are negatively charged anytime that they are in a pH environment that is above approximately 3.0. Blood is 7.0 so it is significantly above these pl values. So we ask what are nutritional sources of aspartic and glutamic acid?

"What is Aspartic Acid (Aspartate)?

Definition- Aspartic acid or aspartate, also known as amino succinic acid is a non-essential amino acid that is synthesized itself in the human body through different sources of foods. It is mainly responsible for synthesizing proteins and regulating hormones so also known as building blocks.

Aspartic acid is involved in synthesizing four different amino acids as it plays a vital role in Kreb s cycle; methionine, isoleucine, lysine, and threonine. It is an aspartate family and a proteinogenic amino acid. It is also a neurotransmitter.

Aspartic acid is the product formed by the hydrolysis of proteins. Aspartic acid was first identified in 1868 from legumin in plant seeds. As aspartic acid are non-essential amino they are synthesized in the body from oxalo acetic acid that is produced during the metabolism of carbohydrates.

Structure of Aspartic Acid (Aspartate)

Structure of Aspartic Acid (Aspartate)

Aspartic acid consists of two functional groups, one amino group is basic in nature and the other is the acidic carboxyl group. Therefore, amino acids molecule exists as a zwitterion. Aspartic acid is alanine with one of the ß hydrogens replaced by a carboxylic acid group. The pKa of the ß carboxyl group of aspartic acid in a polypeptide is about 4.0. It is a dibasic amino acid having two carboxyl groups; one on alpha carbon atom and another on the side chain. Aspartic acid has an alpha-keto homolog. Aspartic acid is divided into two forms; L-aspartic acid and D-aspartic acid. L configuration is a more common and dominant form. L-aspartic acid is

typically involved in the production of antibodies and is part of protein synthesis in the body which is responsible for increasing the immune system. D-aspartic acid is not involved in protein synthesis and is mainly found in the pituitary gland and testes which is used in the regulation, synthesis, and release of testosterone and luteinizing hormone.

Sources of Aspartic Acid (Aspartate) It is found in sugar cane and sugar beets molasses, asparagus, avocado, sprout seeds, and oat flakes. Animal Sources includes: Sausage meat, Luncheon meat and wild game. Other sources includes magnesium aspartate that is salt of aspartic acid and sweeter aspartame. Physical Properties of Aspartic Acid (Aspartate) Molecular weight: 133.10 White, crystalline solids Polar Acidic Hydrophilic Orthorhombic, bisphenoidal leaflets or rods Sour in taste Chemical Properties of Aspartic Acid (Aspartate) Melting point: 270°C Solubility: 5390 mg/L at 25 °C Density: 1.6603 at 13 °C LogP: -3.89 pKa: 2.77 because of two carboxyl molecule Biosynthesis of Aspartic Acid (Aspartate) Protein extraction, chemical synthesis, and enzymatic conversion are three main methods to produce aspartic acid. A large number of amino acids are produced in the extraction method from the hydrolysis of protein. In this method, L-aspartic acid should be separated. Chemical synthesis requires high temperature and pressure in a racemic mixture producing both isomers of aspartic acid. So, enzymatic conversion is the best method for the production of aspartic acid. Bacterial fermentation is the best for the highest yield of amino acids. Pseudomonas, Bacillus, and Proteus are considered as the main producers but E. coli and Corynebacterium

Biosynthesis of Aspartic Acid (Aspartate)

glutamacium are mostly preferred by industries.

Figure: Biosynthesis of Aspartic Acid (Aspartate) from Fumaric Acid. Image Source: Kurt Rosentrater (MDPI). In the 1960s, the fermentation process is developed and patented that utilizes sugar-free medium and uses fumaric acid as a sole source of carbon. Ammonia serves as a nitrogen source that is used in catalysis. Ammonia and fumaric acid are used in combination as 1:1 or 1:2 ratio. the pH of the broth is initialized to 7 and that naturally increases from 8.4 to 9.6 in the initial stage that allows for the production of acid. Fermentation can be done with or without agitation for 2 to 10 days at 27-40 °C. L-aspartic acid will be secreted and accumulate in the culture broth. Different downstream processes are available to separate L-aspartic acid from the culture broth. But in the case of batch fermentation, ion exchange resins can be used to separate and purify L-aspartic acid followed by crystallization. L-aspartic acid can be separated by adjusting the broth to 90 ?C and a pH of 2.8 with sulfuric acid in continuous fermentation. After the pH is adjusted to 2.8, the isoelectric point will cause L-aspartic acid to precipitate out of the solution. It is then subjected to a two-hour incubation period at 15 °C to induce protein crystallization. Under these conditions, L-aspartic acids yield 95%.

Production of Aspartic Acid (Aspartate)

Figure: Production of L-Aspartic Acid. Image Source: Kurt Rosentrater (MDPI).

Functions and Uses of Aspartic Acid (Aspartate)

It is easily available multivitamins that are found in different forms as tablets, powders, and fluids. Because of its role in regulating testosterone levels, D-aspartic acid is used as increasing muscle mass. It also helps in keeping the concentrations of NADH (Nicotinamide adenine dinucleotide) high in brain cells and also increase the mind sharpness leading to further production of neurotransmitters as well as chemicals needed for normal mental functioning.

Can also be used for increasing fertility.

It is also used to produce poly aspartic acid that is used as a fertilizer synergist.

It also aids in energy production, RNA and DNA synthesis, and liver detoxification.

It also helps in removing excessive toxins from the cells like ammonia.

It is also used as building blocks molecules for active pharmaceutical agents.

It is useful in making culture medias, detergents, fungisides and germicides."

https://thechemistrynotes.com/aspartic-acid/



Name (abbreviations)	Side chain	pl	Name (abbreviations)	Side chain	pl
alanine (ala, A)	—-СН3	6.11	methionine (met, M)		5.74
arginine (arg, R)	$\begin{array}{c c} & & & & \\ & & H_2 & H_2 & H_2 & H \\ & & & C & C & C & N \\ \end{array} \begin{array}{c} & & & \\ & & & & \\ & & & \\ & & & & \\ & & & & \\ & & & & \\ & & & \\ & & & & \\ & & & \\ & & & & \\ & & & & \\ & & & & \\ & & $	10.76	phenylalanine (phe, F)		5.91
asparagine (asn, N)	H ₂ C NH ₂	5.41	proline (pro, P)	CH ₂ CH ₂ CH ₂	6.30
aspartic acid, aspartate (asp, D)		2.87	serine (ser, S)	Н ₂ ————————————————————————————————————	5.68
cysteine (cys, C)	<u>— с — с — он</u> — с — sн	5.02	threonine (thr, T)	CH CH3	
glutamine (gln, Q)		5.65			5.60
glutamic acid, glutamate (glu, E)	н ₂ н ₂ н ₂ 	3.08	tryptophan (trp, W)		
glycine (gly, G)	— Н	6.06			5.88
histidine (his, H)		7.64			
isoleucine (ile, I)	СН ₃ — Ц Н2 С СН3	6.04	tyrosine (tyr, Y)	Нг ОН	5.63
leucine (leu, L)	СH ₃ H ² — СH—СH ₃	6.04	valine (val, V)	СH ₃ СH—СH ₃	6.02
lysine (lys, K)	H2 H2 H2 H2 H2 H2 H2	9.47			

Figure 13.1.4: Amino acid side chains and pI values.



2023.12.02 Abstracts 2022 Prepared

Prepared abstracts for 2022 on both carnicominstitute.org and substack sites.

2023.12.04 Media Summaries - Vinyl

I have completed several important summaries of the work that has been done in 2022-2023. I have made a video as well as two audio summaries of 2022 and 2023 abstracts. All work has been prepared with Al assistance; tools are very helpful and allow work to be done which would never be done otherwise.

I suspect work on the order of 3 weeks full time has been completed within about 3 days of work. That is a leverage of about 20:1. I easily could have hired a team of 3 people working for those same 3 weeks. Estimated cost: 150 per day x 3 folks = \$450/day. 450 * 21 days = \$94,500. Not exactly an exaggeration and it show the power of the tools and shows how work that never would have been done has been completed for about \$100 of tools (at most) and 3 days of my time. It will never be quite the same again for this particular type of work that helps others to access and understand better.

Let's get back to the paper:

tell me about polyamides.

Polyamides are a class of polymers that are known for their toughness, strength, and resistance to wear and tear. They are commonly used in various applications, including textiles, engineering plastics, and automotive parts.

One of the most well-known polyamides is nylon, which was first developed in the 1930s. Nylon is a synthetic polyamide that is made from a combination of chemicals, typically derived from petroleum. It is known for its high tensile strength, good elasticity, and excellent resistance to abrasion and chemicals.

Polyamides have a unique molecular structure that consists of repeating units linked by amide bonds. This structure gives them their characteristic properties. The amide bonds create a strong and rigid polymer chain, which contributes to the strength and durability of polyamides.

In addition to nylon, there are several other types of polyamides, each with its own set of properties and applications. Some common examples include:

1. Polyamide 6 (PA 6): This type of polyamide is derived from caprolactam and is widely used in the production of fibers, such as nylon 6. It is known for its excellent mechanical properties, high melting point, and good resistance to chemicals.

2. Polyamide 66 (PA 66): PA 66 is made from a combination of adipic acid and hexamethylenediamine. It is similar to nylon 6 but has a higher melting point, better resistance to heat, and improved mechanical properties.

3. Polyamide 11 (PA 11): PA 11 is a bio-based polyamide derived from castor oil. It is known for its excellent resistance to chemicals, UV radiation, and hydrolysis. It is often used in applications where high-performance and sustainability are required, such as automotive components and sports equipment.

4. Polyamide 12 (PA 12): PA 12 is a flexible and tough polyamide that is commonly used in the production of tubing, pipes, and hoses. It has good resistance to chemicals, impact, and abrasion.

Polyamides are versatile materials that can be modified to meet specific requirements. They can be reinforced with fillers, such as glass fibers, to enhance their strength and stiffness. They can also be blended with other polymers to create hybrid materials with unique properties.

Overall, polyamides are widely used in various industries due to their excellent mechanical properties, chemical resistance, and durability. Their versatility and ability to be tailored to specific applications make them an important class of polymers.

what is the difference between amines and amides?

Amines and amides are both organic compounds that contain nitrogen, but they differ in their functional groups and chemical properties.

Amines are compounds that have a nitrogen atom bonded to one or more alkyl or aryl groups. They can be classified as primary, secondary, or tertiary amines, depending on the number of alkyl or aryl groups attached

to the nitrogen atom. Amines are basic in nature and can act as nucleophiles in chemical reactions. They often have a strong ammonia-like odor.

Amides, on the other hand, are compounds that have a nitrogen atom bonded to a carbonyl group (C=O). The carbonyl group is typically attached to an alkyl or aryl group. Amides can be classified as primary, secondary, or tertiary, similar to amines. Unlike amines, amides are not basic and do not have a strong odor. They have higher boiling points and are less volatile than amines.

In terms of their chemical properties, amines can undergo reactions such as nucleophilic substitution, alkylation, and oxidation. Amides, on the other hand, are less reactive and undergo reactions such as hydrolysis, reduction, and dehydration.

In summary, the main difference between amines and amides lies in their functional groups and reactivity. Amines have a nitrogen atom bonded to alkyl or aryl groups and are basic in nature, while amides have a nitrogen atom bonded to a carbonyl group and are less reactive.

can amines form a polymer?

Yes, amines can form polymers. A polymer is a large molecule composed of repeating subunits called monomers. Amines, which are compounds that contain a nitrogen atom bonded to one or more carbon atoms, can undergo polymerization reactions to form polyamines.

Polyamines are polymers that consist of repeating amine units. They can be formed by various polymerization methods, such as step-growth polymerization or chain-growth polymerization. In step-growth polymerization, two or more monomers react together to form a polymer, while in chain-growth polymerization, monomers add one by one to the growing polymer chain.

Polyamines have a wide range of applications, including as adhesives, coatings, and flocculants. They can also be used in biomedical and pharmaceutical fields, as well as in the production of certain plastics and fibers.

what are the chemical and physical properties differences between polyamines and polyamides?

Polyamines and polyamides are two different types of polymers with distinct chemical and physical properties.

Chemical Properties:

1. Composition: Polyamines are polymers composed of repeating amine (-NH2) functional groups, while polyamides are polymers composed of repeating amide (-CONH-) functional groups.

2. Reactivity: Polyamines are generally more reactive than polyamides due to the presence of primary and secondary amine groups. They can undergo various chemical reactions such as condensation, cross-linking, and complex formation. Polyamides, on the other hand, have lower reactivity but can still undergo certain reactions like hydrolysis and esterification.

3. Solubility: Polyamines are typically soluble in polar solvents like water and alcohols due to the presence of amine groups that can form hydrogen bonds. Polyamides, however, are generally insoluble in water but can dissolve in certain organic solvents like formic acid, sulfuric acid, and phenol.

Physical Properties:

1. Melting and Boiling Points: Polyamines generally have lower melting and boiling points compared to polyamides. This is because polyamines have weaker intermolecular forces (such as hydrogen bonding) between the polymer chains, leading to lower energy requirements for breaking these forces. Polyamides have stronger intermolecular forces, resulting in higher melting and boiling points.

2. Strength and Rigidity: Polyamides are known for their high strength and rigidity, making them suitable for applications requiring structural integrity. Polyamines, on the other hand, are generally more flexible and have lower strength due to the presence of amine groups that disrupt the polymer chain packing.

3. Thermal Stability: Polyamides exhibit higher thermal stability compared to polyamines. Polyamides can withstand higher temperatures without significant degradation due to the strong amide linkages, whereas polyamines may undergo thermal decomposition at lower temperatures.

4. Electrical Conductivity: Polyamides are generally insulators, while some polyamines can exhibit conductivity due to the presence of amine groups that can act as charge carriers.

Overall, the chemical and physical properties of polyamines and polyamides differ significantly due to their

chemical compositions and structural arrangements. These differences make them suitable for various applications in industries such as textiles, coatings, adhesives, and biomedical materials.

give me some examples of both polyamines and polyamides. give some commercial product examples also.

Polyamines are organic compounds that contain multiple amino groups (-NH2). Here are some examples of polyamines:

1. Ethylenediamine: It is a common polyamine with the chemical formula C2H8N2. Ethylenediamine is used as a building block in the production of various chemicals, such as nylon, pesticides, and pharmaceuticals.

2. Putrescine: It is a diamine with the chemical formula C4H12N2. Putrescine is naturally found in living organisms and plays a role in biological processes. It is also used in the production of polyamides, pharmaceuticals, and cosmetics.

3. Spermidine: It is a triamine with the chemical formula C7H19N3. Spermidine is found in all living cells and is involved in cell growth and differentiation. It is used in various research applications, such as cell culture and molecular biology experiments.

Polyamides, on the other hand, are polymers that contain amide (-CONH-) linkages in their backbone. Here are some examples of polyamides:

1. Nylon: Nylon is a synthetic polyamide commonly used in textiles, such as clothing, stockings, and carpets. It is also used in the production of various other products, including ropes, fishing lines, and automotive parts.

2. Kevlar: Kevlar is a high-strength polyamide known for its exceptional toughness and resistance to impact. It is used in the production of bulletproof vests, helmets, and other protective gear. Kevlar is also used in the aerospace industry for its lightweight and high strength properties.

3. Polyamide-imide (PAI): PAI is a high-performance polyamide used in applications that require excellent mechanical properties, high temperature resistance, and chemical resistance. It is used in industries such as automotive, electronics, and aerospace for components like bearings, seals, and electrical connectors.

These are just a few examples of polyamines and polyamides, and there are many other compounds and commercial products associated with these polymer families.

now tell me three common ways that synthetic rubber is made.

Sure! Here are three common ways that synthetic rubber is made:

1. Polymerization: This method involves the polymerization of monomers to form long chains of rubber molecules. The most common monomers used in synthetic rubber production are butadiene and styrene. Polymerization can be achieved through various techniques such as emulsion polymerization, solution polymerization, or bulk polymerization.

2. Copolymerization: In this approach, two or more different monomers are combined to create copolymers. By blending different monomers, the resulting synthetic rubber can have enhanced properties compared to rubber produced from a single monomer. For example, the copolymerization of butadiene and styrene produces a type of synthetic rubber called Styrene-Butadiene Rubber (SBR), which is widely used in tire manufacturing.

3. Vulcanization: Vulcanization is a process used to improve the properties of synthetic rubber. It involves the addition of various chemicals, such as sulfur, accelerators, and activators, to the rubber compound. The rubber is then heated, which causes the chemical additives to react and cross-link the rubber molecules. This cross-linking process enhances the rubber's strength, elasticity, and resistance to heat, aging, and abrasion.

These are just a few common methods used in the production of synthetic rubber. The specific technique chosen depends on the desired properties and applications of the final rubber product.

is butadience a vinyl product?

No, butadiene is not a vinyl product. Butadiene is a colorless gas that is used as a raw material in the production of synthetic rubbers, such as styrene-butadiene rubber (SBR) and polybutadiene rubber (PBR). Vinyl products, on the other hand, are made from vinyl chloride monomer and include materials like polyvinyl chloride (PVC).

what are examples of polymer vinyls?
Some examples of polymer vinyls include:

1. Polyvinyl chloride (PVC): PVC is a widely used polymer vinyl that is known for its versatility and durability. It is commonly used in construction materials, pipes, electrical cables, and vinyl flooring.

2. Polyvinyl acetate (PVAc): PVAc is a polymer vinyl often used in adhesives, paints, and coatings. It is also the main component in white glue or wood glue.

3. Polyvinyl alcohol (PVA): PVA is a water-soluble polymer vinyl that is used in various applications such as adhesives, coatings, and textiles. It is also used in the production of PVA films and fibers.

4. Polyvinylidene chloride (PVDC): PVDC is a polymer vinyl that is known for its excellent barrier properties against gases and moisture. It is commonly used in food packaging films, shrink wrap, and protective coatings.

5. Polyvinylpyrrolidone (PVP): PVP is a polymer vinyl that is used in pharmaceuticals, cosmetics, and personal care products. It is often used as a binder, film-former, or stabilizer in various formulations.

These are just a few examples of polymer vinyls, and there are many more variations and derivatives available in the market.

what are the chemical and physical property differences between dienes and trienes?

Dienes and trienes are both types of hydrocarbons that contain double bonds. However, there are some differences in their chemical and physical properties:

1. Chemical Properties:

Dienes have two carbon-carbon double bonds, while trienes have three carbon-carbon double bonds.
 Dienes can undergo addition reactions with electrophiles, such as hydrogenation or halogenation, at each double bond independently. Trienes can also undergo similar addition reactions, but with the presence of three double bonds, the reactions can be more complex.

 Dienes can undergo polymerization reactions to form polymers, such as synthetic rubbers. Trienes can also undergo polymerization, but the presence of more double bonds can lead to more complex polymer structures.
 Dienes can undergo Diels-Alder reactions, which involve cycloaddition reactions with dienophiles. Trienes can also participate in Diels-Alder reactions, but with the presence of more double bonds, the reactions can have more diverse outcomes.

2. Physical Properties:

- Generally, as the number of double bonds increases from dienes to trienes, the boiling points and melting points tend to increase. This is due to the increased intermolecular forces (Van der Waals forces) between the molecules.

- The presence of double bonds in both dienes and trienes can lead to increased reactivity, making them more prone to oxidation and other chemical reactions compared to saturated hydrocarbons.

- Both dienes and trienes are typically liquids or low-melting solids at room temperature, depending on the specific structure and molecular weight.

It's important to note that these properties can vary depending on the specific diene or triene compound, as different substituents and structural arrangements can influence their behavior.

so we know that dienes and trienes are alkanes.

what it is called when you have two vinyl functional groups combined

When two vinyl functional groups are combined, it is called a vinyl group dimerization or vinyl dimerization.

Look at our notes of Nov 16 2023:

I am looking at 100.8. This is the larger volume decant sample prepared for additional pl testing. It will now also be used and given equal priority to polymerized protein production (presumed identity). It is following the same progression of 100.5 (also a titration decant) with:

```
1. Increasing cloudiness
```

Lowering of pH
 Increased TDS

4. Increased PPT

as recorded on spreadsheet during vitals monitor today.

We notice that the "vitals" in no way capture the whole story. They are only a helpful indicator but they can miss very important events. An example is what is happening between 100.2, 100.3 and 100.4

100.4 is TSP control 100.2 is .04 gms enzyme in dilute TSP 100.3 is .12 gms enzyme in conc. TSP

The vitals indicate possible significance with TSP 100.2 which is true and of interest since it is forming a synth cell-polymer surface layer even though lighter concentration of enzyme has been added.

However, 100.3 by vitals shows nothing of particular interest. Visually, however, it is more than important, especially as it compares to 100.2 The only difference should be an increased concentration of enzyme to 0.12 gms. The culture is TRANSPARENT with NO synth cell - polymer surface layer. This is a huge difference, but the vitals miss it and only observation detects it. So stay alert is the lesson....

Now let's review the NIR spectrum of the borax-decant reaction from yesterday. It was presumed that it might have close ties with a polyvinyl compound. We did notice and are aware, however, that the borax reaction product was neither water soluble or flexible.

Note: (1) Weak peak (3) Strong peak

Tabular info comes from CRC NIR handbook

Peak info:

983 (2) OH from water (maybe but maybe not....) (979) Phenolic OH (990)

1170 (2) Alkene, polyenes (1170)

1347 (2) Methyl (1360) larger delta than normal

1436 (3) Aromatic amine (1432) Methylene (1440)

1480 (3) Polyamide (1480) Secondary Amine (1481) Amide/Protein (1483)

and in general, high absorbance in the range of 1436 - 1480; aromatic amine is strong here.

Now my initial thought was that polyvinyl alcohol related compounds might show up in this NIR spectrum. However, the OH is not dominant here if at all, and the compound is not soluble. The water question can be settled and separated out by sustained drying of the sample. Phenols are moderately soluble in water. Now we know that this compound is highly insoluble. This indicates that OH solubility is not a strong factor here. The polymer and protein signatures are what dominate here. For now, I will presume water over phenol in the 983 nm group but will keep an eye on this.

Most conductive polymers are polyenes. Polyenes are a main target of astrobiology. Polyenes share some chemical reaction properties with PVC vs PVA. PVC is C2H3Cl (notice no OH in any way). So if of a plastic nature would seem to be more likely of PVC family than PVA since we are insoluble and somewhat crystalline nature under scope and brittle.

THE MATERIAL DOES NOT PASS A CONDUCTIVE TEST WITH THE OHMMETER AT THIS POINT.

so 983 peak remains undecided between OH water and alkene, polyene.

Now the aromatic amines, polyamides, secondary amine, and amide protein all speak of being a polymerized

protein, especially in light of the borax reaction which has produced it.

This exists as the most current interpretation of this material that has been created, a polymerized protein. Unclear at this point if the polymerization is likely to come from aromatic or amide origin, or both for that matter. We know that in the end a "rubberized" polymer is developed, but this can come from the other TSP surface synth-cell polymer layer. But out of curiosity, is rubber aromatic or amide based?

Rubber is alkene based. OK, this gets very interesting. Synthetic rubber is a copolymer between styrene and butadiene. Styrene is a VINYL group attached to an aromatic ring. Butadiene is an alkene.

Guess what, all the signs are starting to point the same direction. Aromatic, vinyl, alkenes are the components of synthetic rubber, which is most characteristic of the mature clot gross properties. Aromatic, vinyl, alkenes appear to be dominant structures of our NIR plots. Do not keep trying to force a single functional group. The combination of these three functional groups along with synthetic blood go a long way toward explaining the culture findings in general.

Start looking at these three functional groups in a combined sense, understanding that separations in the culture will likely break these into parts. We will now keep an eye on the NIR plots more closely with these various combinations.

Let's create more of the proposed polymerized protein, now designated as TSP 100.10. Done. Drying.

I am trying to acidify TSP 100.10 material w/ moderately strong H2SO4. It is difficult. It may be slightly soluble in H2SO4 but certainly not easily. This means that both polymeric forms, TSP surface layer synth cell-polymer layer and this newly developed polymer from the decant and borax are BOTH very much insoluble in water and difficult to solubilize in reagents as well. Just like the clot was....

I need to conduct solubility tests on 100.10 just like I did on the synth surface polymer on TSP 070. More study there again as TSP 100.0 matures.

My goal is to attempt to dissolve (hydrolize) TSP 100.10 for a UV run to determine if protein is detected.

OK, that was straightforward. Definite absorbance at 275 nm. Protein confirmed. Polymeric protein remains confirmed. Now our goal is to try and determine the nature of the polymer. Our first NIR run says we have:

- 1. alkenes, polyenes
- 2. methyl possible
- 3. aromatic amines
- 4. methylene
- 5. polyamide
- 5. secondary amine
- 6. amide/protein
- 7. polyamide, amide

Therefore, the reduced and presumed polymer set is:

- 1. polyenes
- 2. aromatics
- 3. amines
- 4. polyamide

Seems like an ideal polymerized protein to me. Qualifies as a constituent of the final clotting process. Missing quality seems to be the rubber nature, but this seems to fit quite well for the TSP 070 surface synth layer - polymer.

The essence of the clot seems to be established at this point:

1. Synth erythrocytes w/hemoglobin

2. Two polymeric forms, one for structual density and the other for elasticity. Both highly insoluble, even in many strong reagents/acids/caustics.

This is a major achievement. It would certainly be of benefit to understand how the two polymers may join together.

Now the rubber property would be imparted by the vinyl group, which we now understand is a part of the copolymeric process of synthetic rubber. We may well have, and are expected to have a copolymer arrangement here.

This means that we would expect to see the vinyl component from one of the two polymers, and the strongest candidate for that is now the TSP synth cell - polymer surface layer. Let's look at NIR plots.

Guess what? (The database sure pays off). From notes of Nov 12, the vinyl component is in the precipitate! This is the other half of the TSP prior to titration!. We have all the parts we need. We have therefore separated between:

Three polymers, consistent of all needed parts:

One separated polymer contains:

1. polyenes

2. aromatics

3. amines

4. polyamide

(i.e., proteins and binding polymers)

The second polymer, i.e. the precipitate of the titration:

1. contains vinyl

(i.e., imparts elasticity)

and synthetic blood containing hemoglobin in a polymer matrix, also elastic in nature (thus protein & polymer here also).

This means you need to know if the precipitate contains protein also. I think NIR says yes. But let's work on that one.

This is all leading to the likely conclusion of three polymeric protein complexes:

- 1. Decant
- 2. Precipitate

3. Surface synth cell layer

Each one will have its own dominant chemical properties.

This would therefore certainly be a copolymeric operation. Expected that decant + precipititate = synth cell surface layer with synthetic blood.

The precipitate to the titration is 100.6. We have enough of this material to work with for now. It is important to realize that this layer, upon centrifuge, actually contains two layers, an upper grey layer higher in volume, and a lower white layer minimal in volume. This layer now becomes important in seeking out the vinyl contribution to the overall polymer structure. For now we will focus only on the grey upper layer as it comprises the bulk of the volume of centrifugation.

We have it. NIR evaluation of the borax polymer vs the precipitated tells us the distribution and make up of the synth surface layer.

TSP 100.6 (the precipitate from titration) has high absorbance in the 1640 region. This is the vinyl dominant region of NIR.

TSP 100.10 (the borax polymer) has high absorbance in the 1440 region. This is dominated by aromatic amine. The precipitate is not high in protein content. 293 nm and 232 peaks were identified in the 100.6 acidified precipitate.

So what this is saying is that the middle layer of the culture (most volume) contributes the bulk of protein. The precipitate contributes the elasticity.

So our three main components (everything is temperature and pH dependent for expression (\sim 18 expressions thus far) are:

- 1. Synthetic blood (w/hemoglobin) (scores of proteins within blood) (surface layer of mature culture)
- 2. A set of proteins that can be polymerized with borax, structurally firm but not elastic. (within decant)
- 3. A vlnyl contribution for elasticity of the culture. (within precipitate)

This makes up the majority of the clot structure.

1,3-Butadiene (a synthetic rubber) from NIST data base UV spectral search shows a peak at 293. This is a vinyl compound and then a match for our spectrum of TSP 100.6 at this point (not the rest of the spectrum nor our 232 peak found.

NIST and Chemspider both have some UV searching capability if you can get a part of the name. VERY VALUABLE!

The styrene UV spectrum explains high absorbance towards the 232 peak. Synthetic rubber is a copolymer between styrene and butadiene.

Also our notes of Nov 26 2023 look to be of interest:

I have an interesting observation regarding the purple color on the synth-polymer layer, and I need to recall what I did. I believe it was the following:

1.A small piece of the dried synth polymer layer (a tough vinyl sure fits the bill) in about a ml of H2O. Then I think a drop of conc. H2SO4, maybe two or three. No purple color change takes place under these conditions of a weaker H2SO4 solution. Put it in the frig because of something I remember reading regarding chilling inducing a purple reaction with blood and sulfur.

Remembered two days later to look at it. It is indeed purple, just as I have been able to achieve with very conc. H2SO4 at rooom temperature with the synth layer.

So I will be:

1. Looking at this more and for replication.

2. Try to find what I read regarding H2SO4, blood and chilling purple reaction.

3. Ask the question, this means that the vinyl compound/product would have a reaction similar to with blood and therefore share another characteristic of blood and hemoglobin interaction....It also means that sulfur interacts with the synth polymer without doubt, in a way that is similar to how sulfur can react with blood.

"Sulfhemoglobinemia is a rare condition characterized by the presence of sulfhemoglobin in the blood. Sulfhemoglobin is a form of hemoglobin that contains sulfur instead of iron in its heme group. This alteration in the structure of hemoglobin affects its ability to bind and transport oxygen, leading to a decrease in oxygen-carrying capacity in the blood.

The exact cause of sulfhemoglobinemia is not fully understood, but it is thought to be a result of exposure to certain chemicals or medications. Some known triggers include certain drugs like phenacetin, sulfonamides, and aniline dyes, as well as exposure to industrial chemicals and toxins.

Symptoms of sulfhemoglobinemia can vary but may include cyanosis (bluish discoloration of the skin), shortness of breath, fatigue, dizziness, and confusion. In severe cases, it can lead to tissue damage and organ dysfunction.

Diagnosis of sulfhemoglobinemia is typically made through a blood test that measures the levels of sulfhemoglobin in the blood. Treatment options are limited, and there is no specific cure for the condition. In most cases, the condition resolves on its own once the exposure to the triggering agent is removed. Supportive care, such as oxygen therapy, may be provided to manage symptoms and improve oxygenation.

It is important to consult with a healthcare professional for a proper diagnosis and management of sulfhemoglobinemia. They can provide personalized advice and guidance based on individual circumstances."

"Sulfhemoglobinemia is a rare condition in which there is excess sulfhemoglobin (SulfHb) in the blood. The pigment is a greenish derivative of hemoglobin which cannot be converted back to normal, functional hemoglobin. It causes cyanosis even at low blood levels.

It is a rare blood condition in which the ß-pyrrole ring of the hemoglobin molecule has the ability to bind irreversibly to any substance containing a sulfur atom.[1][2] When hydrogen sulfide (H2S) (or sulfide ions) and ferrous ions combine in the heme of hemoglobin, the blood is thus incapable of transporting oxygen to the tissues.

Presentation

Symptoms include a blueish or greenish coloration of the blood (cyanosis), skin, and mucous membranes, even though a blood count test may not show any abnormalities in the blood. This discoloration is caused by greater than 5 grams per cent of deoxyhemoglobin, or 1.5 grams per cent of methemoglobin, or 0.5 grams per cent of sulfhemoglobin, all serious medical abnormalities.[citation needed."

"Occurs when a sulfer atom binds to porphyrin ring of hemoglobin, resulting in permanent oxidation of iron to the ferric state, incapable of oxygen transport

Most often associated with phenazopyridine, dapsone, metoclopramide, sumatriptan Also associated with industrial chemicals, including trinitrotoluene, hydroxyl amine sulfate, dimethyl sulfoxide, Hydrogen Sulfide"

"Sulfhemoglobinemia is a rare condition where the blood takes on a purple color due to the presence of sulfhemoglobin. Sulfhemoglobin is a compound formed when sulfur combines with hemoglobin, the protein responsible for carrying oxygen in red blood cells. This condition can occur as a result of certain medications, chemicals, or toxins, and it can lead to symptoms such as shortness of breath, fatigue, and cyanosis (bluish-purple discoloration of the skin). Treatment for sulfhemoglobinemia typically involves identifying and discontinuing the causative agent, and in severe cases, blood transfusion may be necessary. It is important to consult a healthcare professional for proper diagnosis and management of this condition."

It is a morbid condition.

Major removal information related to this reaction has been scrubbed from AI Chat and YouTube after original inquiries 24 hours earlier. Any relationships to use of sulfuric acid in the reaction have been removed.

So the conclusion is because this reaction is occurring with the synth polymer layer it is another level of confirmation of the existence of hemoglobin within the synth polymer layer. Scrubbing of information looks to be pretty clear on this research.

OK, there is some very cliear evidence available of vinyl in the blood. RIght now it is from NIR, we will see if it exists in MID IR afterwards. If we look at our notes of Oct 31 2023 (handwritten) we have a clear entry of aborbance at 1627 nm within a human blood analysis. This is clearly indicated within the CRC reference as being of vinyl nature from 1621 - 1637. Five consecutive entries with no real competition except for acrylate at 1621. This is a sufficient entry to show crossover between synth polymer analysis and human blood.

OK, we are at the point now where we can provide direct evidence of vinyl influence on human blood via NIR. The method is to focus on human blood alone, not comparing it to anything, and look at with NIR. We also focus in on the vinyl region of 1627 nm and remove extraneous examinations. The evidence is quite strong now of the existence of vinyl groups, polymerization, synthetic blood, within human blood. This is from a human blood sample 2023, unvaccinated individual.

It now becomes less important, but the question arises, can you show evidence of vinyl presence in a human blood MID IR spectrum available in 2015? We know we can show evidence from the MID IR of a "CDB Protein Complex" MID IR spectrum of 2015 in combination with use of SDBS database.

"The typical wavelength range for infrared absorption of vinyl groups is around 1600-1650 cm^-1."

Caution: Amides also absorb in ths region!

"The amide absorption in the infrared region of human blood typically occurs at wavelengths between 1600 and 1700 cm-1. This absorption is mainly attributed to the stretching vibrations of the amide I and amide II bonds in proteins present in the blood." The

Now we definitely have absorption of the protein complex MID IR of Sep 05 2015 at 1638 cm-1. This confirms the likely existence of the vinly group detected within the cultures of 2015. Now what about in blood? We have absorption at 1646 but so does amide.

"The simplest vinyl compound is vinyl chloride (C2H3Cl)."

"An ether functional group is a functional group in organic chemistry that consists of an oxygen atom bonded to two alkyl or aryl groups. It is represented by the formula R-O-R', where R and R' are alkyl or aryl groups. Ethers are characterized by the presence of the oxygen atom in the molecule, which gives them unique chemical and physical properties"

"Ether absorption in the infrared region typically occurs in the range of 1000-1300 cm-1. This absorption is due to the C-O stretching vibration of the ether functional group. The exact position of the absorption peak may vary depending on the specific structure of the ether molecule."

Now, here is what we are seeing. The 2015 CDB MID IR shows strong absorption at 1038 cm-1. THis looks like it MAY correspond to ether.

Molecules that contain a C-O bond typically absorb infrared radiation at around 1038 cm-1. Ether, which has the chemical formula R-O-R', where R and R' can be any alkyl or aryl group, contains a C-O bond and can absorb infrared radiation at this frequency.

I have found the MID IR correlation that I am seeking. My spectrum with albumin matches EXTREMELY WELL except for 2 peaks that exist in the range of ethers and vinys. 1298 and 1160 cm-1. SDBS will find that the closest match to these two peaks is ethyl vinyl ether, exactly the top candiate from the CDB Protein complex MID IR of 2015.

Poly(vinyl cyclohexyl ether) (PVChE) is a type of polymer that is derived from vinyl cyclohexyl ether monomers. It is often used in the field of biological materials due to its unique properties and potential applications.

PVChE is biocompatible, meaning it is non-toxic and does not cause harm to living organisms. This makes it suitable for use in medical and biological applications, such as drug delivery systems and tissue engineering.

PVChE can be easily synthesized and modified to achieve desired properties, such as controlled release of drugs or enhanced mechanical strength. It can also be combined with other polymers or materials to create composite materials with specific functionalities.

In addition, PVChE has good solubility in various solvents, allowing for easy processing and fabrication into different forms, such as films, fibers, and hydrogels. This versatility makes it a valuable material for a wide range of biological applications.

Overall, PVChE is a promising material in the field of biological materials due to its biocompatibility, tunable properties, and processability. Ongoing research and development in this area aim to further explore its potential applications and optimize its performance in various biological systems.

Health observation: Besides severe skin damage (best analogy is a year + long internal burn and bee sting (eg, formic acid)) that covers the entire lower leg, angle and foot) significant edema is in place. Swelling is aggravated further with any increase in salts such as sodium (sodium bicarbonate used in pH control is counterproductive it seems). The observation is that ultrasound applied diligently and with requisite pain at point of core pain reduces the swelling temporarily in that area so that leg is normally sized. US drives the fluid away, albeit only locally. It is already known from under the scope that US separates the blood from the polymer.

"Polystyrene with DVB: Cross-linking between polymer chains can be introduced into polystyrene by copolymerizing with p-divinylbenzene (DVB). DVB has vinyl groups (-CH=CH2) at each end of its molecule, each of which can be polymerized into a polymer chain like any other vinyl group on a styrene monomer."

"Addition polymers from conjugated dienes Polymers from conjugated dienes usually give elastomer polymers having rubber-like properties.

Table 2. Addition homopolymers from conjugated dienes

Monomer Polymer name Trade name Uses

H2C=CH-C(CH3)=CH2 polyisoprene natural or some synthetic rubber applications similar to natural rubber H2C=CH-CH=CH2 polybutadiene polybutadiene synthetic rubber select synthetic rubber applications H2C=CH-CCI=CH2 polychloroprene Neoprene chemically-resistant rubber

All the monomers from which addition polymers are made are alkenes or functionally substituted alkenes."

Recall the outline of our paper: (life is quite complex here):

Outline of our paper:

I also have a good macro picture of the polymer synth layer on a glass slide. So good, we have

1. UV of human blood, very distinctive.

2. Microphotos of synth blood. Easy to compare to human blood photos. 8000x.

3. Macro photo of synth polymer layer.

4. We have a NIR comparison that demonstrates the polyamide/vinyl nature of the synth blood. Likely methylation disturbance at play.

- 5. Mid IR of vinyl groups is coming into play
- 6. Macro photo or culture dish surface layer.
- 7. UV comparison spectra between human blood and synthetic blood, high correspondence.

8. Photo of "blood red" color change upon mixing polymer surface layer and strong H2SO4

also recall kastly meyer, hemoglobin, and pl determination.

9. MID IR correlation with non fitting albumin peaks.

A couple more phenomenall things have happened.

1. When you take the dried synth material and put in in strong H2S04, it turns immediately deep red. Slight purple tint. Hard to avoid the situation with blood. I have photo.

2. In addition, there is significant overlap in the UV spectrum with human blood, especially from 190 nm to 300 nm. Peaks, valleys and general profile coincides as well.

Also you have a recollection of a compound which is "essentially two vinyl groups joined together" but you cannot find it. what was it?

I FOUND IT! GOOD WORK. It is butadiene.

Now what we need to to do is establish a pattern of likely behavior:

vinly - polymer - butadiene rubber components polyamide - sythetic blood styrene

combinations. polyamindes dissolving in surfuric acids polyamides, nylon, protein differences, sulfuric acid, formic acid

is butadiene a vinyl?

No, butadiene is not a vinyl. Butadiene is a colorless gas that is used in the production of synthetic rubber, while vinyl refers to a type of plastic that is made from vinyl chloride monomers.

So no wonder it was and is confusing. Butadiene is not technically a vinyl since "vinyl" is a particular form of plastic. But VINYL FUNCTIONAL GROUPS make up butadiene, and even though it is a gas, it is considered essentially as two vinyl groups joined by a single carbon bond. So from my perspective, it is definitely a vinly compound. Semantics going on here...

"1,3-Butadiene is a simple conjugated diene with the formula C4H6 and can be viewed structurally as two vinyl groups (CH2=CH2) joined together with a single bond."

is butadiene a polymer?

No, butadiene is not a polymer. It is a small molecule that is commonly used as a monomer to produce synthetic rubbers such as polybutadiene, which is a polymer.

is triene a polymer?

look into all spectroscopy matches involving butadiene, ethers vinyl... look at the mid ir matches again. separate between amines and amides, polyamides, etc. No, triene is not a polymer. Triene refers to a molecule that contains three double bonds. It is a type of unsaturated hydrocarbon.

You have a lot of data now, you need to synthesize it into a composite picture that demonstrates a polymeric progression.

I have mapped out a 7 way grid to collect data from different culture states. WIII within that grid to try and synthesize information and transfer here as it develops.

2023.12.05 Blood Clot Progession Study

I have a ton of data here to try and assimilate and synthesize. I have to get a handle on to approach it. We have 4 main steps:

- 1. The native culture characteristics, as best we can determine.
- 2. The incubated culture where it separates into 3 layers. Top, middle, bottom.
- 3. More detailed analysis of the three layers with an eye as to how the both separate and have combined.
- 4. A projection into the blood clot.

Let's start by trying to get a handle on the native culture. The first standard culture was TSP 070, created on Oct 11 2023. It still exists and is quite mature now, almost two months old.

NIR study will be a mainstay of course, maybe some UV. Some solubility. We know highly affected by pH in addition to time separation.

Let's collect what we have on relevant NIR data.

Looks like our 1st NIR plot involving TSP 70 series was on Nov 07. So it was a month old by the time we started looking at it that way, apparently.

This project of separation is looking very difficult between discerning the original native culture vs the time separated culture. I think we are going to have to focus on everything that is collected and look primarily for repetion, either in TSP 070 or TSP 100.

TSP 070 came into existence on Oct 08. Our studies immediately afterwards primarily involved titration and pl studies, trying to learn about the acidic nature of the culture.

But it looks like our first data came in on Oct 28 regarding functional groups. So it was about 20 days old. Now what happened here is I was exploring the use of chromatography. Surprisingly it may have been more insightful than I knew at the time. I was working with the decant so this means the main volume AFTER precipitation by pH.

UV:

274 nm Protein (best estimate is tyrosine as a dominant amino acid). Highly soluble so the OH group makes sense here.

315 nm UNKNOWN - repeated later?

NIR:

1033 Polyfunctional alkyl alcohols (1029) Ethers and Esters also containing alcohols (we know everything is still soluble her4e).

963 Alkyl alcohols

Now what we see is that on Nov 12 BOTH TSP 100 and TSP 070, regardless of incubation time both displayed absorbance at ~315 nm. So yes, we do have a repeat. But we do not know what 315 refers to.

Ideas are:

1. Synthesized metal oxide nanoparticles

2. Carbohydrates in H2SO4 but we are not in H2SO4 as far as we know.

Oct 28 2023

So what comes out of the first functional group analysis of TSP middle layer is:

- 1. Protein (with increased interest upon tyrosine).
- 2. Alkyl alcohols
- 3. Ether and Ester Alcohols
- 4. Calcium ions (from a water test).
- 5. Interest in metallic nanoparticles (315 nm).
- 6. Acidic (pH is ~3.8)

Two references do indicate nanoparticles at 315nm.

On Oct 29 we make a NIR run with the carbonate precipitate from the decant.

There certainly is no purported "AI" that can handle this problem that lies ahead. Synthesizing information from widely disparate sources and methods is required. Will need to plug away one day at a time to try to extract the relevant info. Ranking is going to end up being very important so we need to identify the factors ahead of time as much as possible. Considerations are:

- age of sample
 type of sample
 solubility of sample
 pH of sample if possible, or direction towards biological pH of 7.4

Let's see if I can enter the incoming information into a spreadsheet so I do not have to enter it twice.

2023.12.06 Functional Group Worksheet

I am getting a spreadsheet-worksheet set up to start approaching this problem. Lots of info to synthesize. I have one advantage already, I can tally up in real time as I go the frequencies of functional groups appearing. Then I will work in a system for ranking including the variables of age, pH, solubility, etc. I have to get it right to begin with so I do not have to repeat myself. I will need to go through each daily lab notes starting from Oct 06 (TSP 070 creation) to the present day. This is going to take some work, but it give me the info I need.

The goal is to see what functinal group and chemical progression exists from the native culture > synthetic blood > clot exists based upon the lab data.

Retracing the collected information day by day has brought up an interesting observation for the notes of Nov 03 2023 where human blood was compared again refernce values in the MID IR literature and synth blood and human blood were contemporaneously compared vi NIR.

I had made a note of 1296 cm-1 being measured and not fitting anywhere in the reference values. I then dismissed that as a fluke. I see not that the measurement was not made accurately and that the actual value for the Aug 16 2015 human blood spectrum was at 1310 cm-1. In addition, I see another absorbance peak at 1167 cm-1.

I consulted 3 different reference sources. The 1310 is now actually explained by reference source no. 2 as protein so this is now accepted. 1167 cm-1 still exists as a valid measurement and I have no curent match.

It looks like 1167 does match with a potential vinyl group.

My notes of Nov 26 2023 are rather of interest. This takes some sorting out. Here are the notes, which are subsequently to be adjusted:

"It now becomes less important, but the question arises, can you show evidence of vinyl presence in a human blood MID IR spectrum available in 2015? We know we can show evidence from the MID IR of a "CDB Protein Complex" MID IR spectrum of 2015 in combination with use of SDBS database.

"The typical wavelength range for infrared absorption of vinyl groups is around 1600-1650 cm^-1."

Caution: Amides also absorb in ths region!

"The amide absorption in the infrared region of human blood typically occurs at wavelengths between 1600 and 1700 cm-1. This absorption is mainly attributed to the stretching vibrations of the amide I and amide II bonds in proteins present in the blood." The

Now we definitely have absorption of the protein complex MID IR of Sep 05 2015 at 1638 cm-1. This confirms the likely existence of the vinly group detected within the cultures of 2015. Now what about in blood? We have absorption at 1646 but so does amide.

"The simplest vinyl compound is vinyl chloride (C2H3Cl)."

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"Ether absorption in the infrared region typically occurs in the range of 1000-1300 cm-1. This absorption is due to the C-O stretching vibration of the ether functional group. The exact position of the absorption peak may vary depending on the specific structure of the ether molecule."

Now, here is what we are seeing. The 2015 CDB MID IR shows strong absorption at 1038 cm-1. THis looks like it MAY correspond to ether.

Molecules that contain a C-O bond typically absorb infrared radiation at around 1038 cm-1. Ether, which has the chemical formula R-O-R', where R and R' can be any alkyl or aryl group, contains a C-O bond and can absorb infrared radiation at this frequency.

I have found the MID IR correlation that I am seeking. My spectrum with albumin matches EXTREMELY WELL except for 2 peaks that exist in the range of ethers and vinys. 1298 and 1160 cm-1 [NOTE BELOW, THESE ARE NOW SUPERCEDED WITH 1310 CM-1 AND 1167 CM-1]. SDBS will find that the closest match to these two peaks is ethyl vinyl ether, exactly the top candiate from the CDB Protein complex MID IR of 2015."

First off, my MID IR sample here (Aug 16 2015) is actually blood serum, not whole blood. So Albumin is actually our best reference. Two peaks remain of interest. 1310 cm-1 and 1167 cm-1. Any measurement in notebooks on Nov 03 2023 about a peak of 1296 cm-1 are now superceded with 1310 cm-1 measurement; the pick was not identified accurately in our spectrum. So two peaks remain in interest, 1310 cm-1 and 1167 cm-1.

Next, I have MID IR reference spectrum for Albumin. These two peaks simply do not fit the spectrum. My additional peaks did match and they indeed are what caused albumin to show up in the list.

However, a more focused search on those particular peaks led to the strongest candidate to be ethyl vinyl ether as mentioned above. I now remain curious about this because of the correlation to the CBB Protein Complex MID IR Spectrum. It is therefore now time to revisit the SDBS search using these two peak values and see if ethyl vinyl ether remains a viable candidate.

Now 1 of 3 reference sources did assign a 1308 cm-1 absorption to a protein, but the other two did not. Let's look at SDBS. Blood and serum have minor differences here. We will use blood as the question is whether a vinyl group unexpectedly shows up in the blood, even years ago.

OK, we have an important level of confirmation going on here on vinyl groups in the blood, back as far as 2015 and 2017, two different samples. The average two peaks of interest are 1298 and 1160 cm-1. With search specs of 1 (that's one!) cm-1 and threshold of 71%, the closest match in the entire database is indeed again ethyl vinyl ether. There is no requirement that vinyl be included as a condition in the search, the peaks alone are justification for this compound. This is a very high level of confirmation of vinyl groups in blood.

Strong acids dissolve nylon (polyamides).

Synth-polymer of TSP dissolves in strong H2SO4. Clot dissolves in KOH-NaOH as I recall. Solubiiity tests took place on Nov 04 2023. Also alcohol detection methods took place on Nov 04 2023.

Health observation. With sustained use of ultrasound am drawing polymer materials from my lower leg/ankle. Sustained project over a year long. Painful with much patience and perseverance required. As the ankle/lower leg improves slowly, the material is now moving lower into the foot. Noticeable "edema" is more frequent, but this is not just water. It will be the water soluble protein/polymer complex. Theory is that whatever can come out of the skin represents an improvement. Be prepared for the long haul.

On Nov 05 2023 the work took a strong turn toward the prospect of nucleic acids within the polmer synth material. I have strong well defined absorbance at 260 nm. Will focus on the profile of the curve here also. Chilled alcohol test failed, but do nucleic acids behave the same. You also had hydrolyzed the polymer synth material with strong H2SO4.

Notes of Nov 12 show influence of L. enzyme upon functional groups of culture. All influences appear positive; polyamide, vinyl, aliphatic hydrocarbons, and halogenation.

2023.12.07 Functional Group Worksheet

Continuing to collect the data, one page, one day at a time. Now on Nov 13, 2023 of handwritten notes. Am accumulating a detailed analysis of acquired chemical knowledge from the culturing process. The starting point is when the culture process stabilized from the development of TSP 070 onwards. Two large cultures are in active monitoring and use. I will attempt to provide a summary view of the worksheet results as I complete the process.

Switch to the digital log is starting to take place around Nov 14 2023. Complexity of data management is acknowledged.

On Nov 14 2023 the probability model developed was specified again:

c = (1/range) * tan ((Problevel * pi) / 200.)

Problevel = (200 / pi) * arctan(c * deltax)

Very elegant to use.

This was developed in radians, there is no need to convert anything. Problevel is 0-100 (i.e, percent, e.g, 70.1). Range is (e.g, RMS). Delta x is measured sample value.

Have been adopting the RMS value as the reference range now at 70.% (.707). This takes care of sign and centering issues. Very easy to implement. Peak value = 1.414 * Problevel.

"The form factor of an AC waveform is the ratio of its RMS value divided by its average value. Square-shaped waveforms always have crest and form factors equal to 1, since the peak is the same as the RMS and average values. Sinusoidal waveforms have an RMS value of 0.707 (the reciprocal of the square root of 2) and a form factor of 1.11 (0.707/0.636)."

Factor of 2 comes from peak to peak.

In our case here, however, it does look like using the range at a 95% level of confidence is very suitable. We can then cut off values at >= 80% (or whatever level is desired, for example).

Nov 16 2023 has a full switchover to the digital log. Doubtful that I see a return to handwritten notes except for computational or diagram work, etc. required.

Nov 16 Notes have the following:

We have it. NIR evaluation of the borax polymer vs the precipitated tells us the distribution and make up of the synth surface layer.

TSP 100.6 (the precipitate from titration) has high absorbance in the 1640 region. This is the vinyl dominant region of NIR.

TSP 100.10 (the borax polymer) has high absorbance in the 1440 region. This is dominated by aromatic amine. The precipitate is not high in protein content. 293 nm and 232 peaks were identified in the 100.6 acidified precipitate.

Recall that TSP 100.6 is:

TSP 100.6C (B) X.XX-1110 Centrifuged Precipitate TSP 100.0 post Titration Two layers grey/white centrifuge precipitate, grey dominates

What interests me here is the UV 293 nm and 232 peaks were not expounded upon. What might these tell us?

I actually did make some comments, very interesting comments:

1,3-Butadiene (a synthetic rubber) from NIST data base UV spectral search shows a peak at 293. This is a vinyl compound and then a match for our spectrum of TSP 100.6 at this point (not the rest of the spectrum nor our 232 peak found.

NIST and Chemspider both have some UV searching capability if you can get a part of the name. VERY VALUABLE!

The styrene UV spectrum explains high absorbance towards the 232 peak. Synthetic rubber is a copolymer between styrene and butadiene.

We also know that we have this information regarding the 232 region and protein folding: "Revisiting absorbance at 230 nm as a protein unfolding probe" https://www.sciencedirect.com/science/article/pii/S0003269709001894

Now this is important. poly Butadiene ALONE is a synthetic rubber, it does not need styrene.

"Polybutadiene - Wikipedia

Polybutadiene rubber is a polymer formed from the polymerization of the monomer 1,3-butadiene. Polybutadiene has a high resistance to wear and is used especially in the manufacture of tires, which consumes about 70% of the production."

Styrene butadiene is just another format of synthetic rubber that brings in the aromatic:

"Styrene-butadiene rubber (SBR) | Britannica

styrene-butadiene rubber (SBR), a general-purpose synthetic rubber, produced from a copolymer of styrene and butadiene. Exceeding all other synthetic rubbers in consumption, SBR is used in great quantities in automobile and truck tires, generally as an abrasion-resistant replacement for natural"

Yes, indeed butadiene has what is needed. It is regarded as two vinyl groups joined together. That is everything we need to form synthetic rubber. It is all falling into place. The styrene is now just a variation to be considered, it makes a stronger rubber.

"Butadiene

1,3-Butadiene is the organic compound with the formula CH2=CH-CH=CH2. It is a colorless gas that is easily condensed to a liquid. It is important industrially as a precursor to synthetic rubber. The molecule can be viewed as the union of two vinyl groups. It is the simplest conjugated diene. Wikipedia"

"What have golf balls, tyres and Lego got in common? They are all made from polymers, from which the main starting material is butadiene."

"1,3-Butadiene (/?bju?t?'da?i?n/)[8] is the organic compound with the formula CH2=CH-CH=CH2. It is a colorless gas that is easily condensed to a liquid. It is important industrially as a precursor to synthetic rubber. The molecule can be viewed as the union of two vinyl groups. It is the simplest conjugated diene.

Although butadiene breaks down quickly in the atmosphere, it is nevertheless found in ambient air in urban and suburban areas as a consequence of its constant emission from motor vehicles.[9]

The name butadiene can also refer to the isomer, 1,2-butadiene, which is a cumulated diene with structure H2C=C=CH-CH3. This allene has no industrial significance."

and the answer is yes, both sulfuric acid and HCL can dissolve tires. So solubility tests are therefore quite important, including a revisit on the clot situation.

"What Will Dissolve Tire Rubber?

There are many chemicals that can dissolve tire rubber, but the most common and effective is sulfuric acid. When mixed with water, sulfuric acid creates a highly corrosive solution that can quickly break down rubber. Other strong acids like hydrochloric acid and nitric acid can also dissolve tire rubber, but they are not as commonly used because they are more dangerous to handle."

The chain is all falling into place, isn't it?...

This was our solubility information obtained on Nov 18 2023 and corresponding comments:

A few solubility tests have been run on the borax polymer today. Results are:

- 1. in strong acid (H2SO4), not soluble
- 2. in strong alkaline (NaOH-KOH), slowly soluble
- 3. in acetone, not soluble
- 4. in ethanol, not soluble

These are helpful results. It has not been published yet, however, the deceased blood clot was eventually dissolved in NaOH-KOH solution over a period of \sim 2 months. This was recorded on Aug 11 and Aug 13 2023 in Vol 30 of the laboratory notebooks.

This now becomes important as we have similar solubity behavior between the deceased blood clot (the most dense and developed, rubber polymer properties) and the borax polymer that has been developed. We know that KOH-NaOH has generally been our only successful reagent for dissolution of any of the filaments. We know they help to break down the disulfide bonds, as in hair.

A question is what other types of polymer bonds might they break down? In this case borate bonds are involved. So we have a good question coming up with the polyvinyl-borax polymer that can be made (the "slime" case). Will KOH-NOH work on that as well? If so, it shows it is effective on the polyvinyl bonds and borax, not just disulfide.

Yes, lye does break down slime. YT video demo: https://www.youtube.com/watch?v=abl1ETlhZ-w

He needed add some water instead of so much direct powder lye, but it still demonstrates the point. Water was used at the end and more clearly shows that breakdown has occurred.

This is valuable as we are not dealing with disulfide bonds here. Vinyl bonds are involved, as they are in our case also.

This was a valuable trial here. Another linkage in bond disruption between the clot polymer and one of the culture polymer forms.

Here is some progression information that I obtained, also from Nov 18 2023

I have a very significant plot now to look at. It is a NIR comparison of TSP 100.5 (Decant) over a 12 day incubation period. This is from the time that it is decanted (amber color) until it turns white approx 10 days later.

Main feature I am looking at is that appears there is a bulk of mass (absorbance) shift from the Nov 06 TSP 100.5 (amber) at a very broad peak of \sim 1620 +/- 20 nm to a more focused peak at Nov 18 TSP 100.5 (White) of 1516. They do however, continue to share a great deal, and in fact the majority of the turf from 1530 - 1650 nm.

Interpretation of this is: 1620 is alkene and vinyl 1520 is protein, 1515 is polyamide

this makes a lot of sense. It is saying it starts out stronger in the vinyl section but then shifts over to a protein nature. However, it still retains the vinyl nature. This means that a conglomerate is being formed, i.e. a polymerized protein as your surmise.

This is exactly what you envision happening. This answers a very important question on culture dynamics.

Let' see if this plot has been addressed yet. No, it has not, another important addition.

Am looking at the reaction between baking soda and citric acid and wish to also contrast it with baking soda + (either calcium citrate, magnesium citrate, or sodium citrate, for example)

well, baking soca + citric acid creates sodium citrate + co2 gas.

2023.12.07 Functional Group Worksheet

The reaction is: H3C6H5O7(aq) + 3 NaHCO3(s) ? 3 CO2(g) + 3 H2O(l) + Na3C6H5O7(aq)Notice that 3 moles of sodium bicarbonate are used to produce 1 mole of sodium citrate.

(Think about mixing magnesium sulfate + citric acid...)

"Sodium citrate is the trisodium salt of citric acid. It has a role as a flavouring agent and an anticoagulant. It contains a citrate(3-).

ChEBI

Sodium citrate is the sodium salt of citric acid. It is white, crystalline powder or white, granular crystals, slightly deliquescent in moist air, freely soluble in water, practically insoluble in alcohol. Like citric acid, it has a sour taste. From the medical point of view, it is used as alkalinizing agent. It works by neutralizing excess acid in the blood and urine. It has been indicated for the treatment of metabolic acidosis. DrugBank

Sodium Citrate is the sodium salt of citrate with alkalinizing activity. Upon absorption, sodium citrate dissociates into sodium cations and citrate anions; organic citrate ions are metabolized to bicarbonate ions, resulting in an increase in the plasma bicarbonate concentration, the buffering of excess hydrogen ion, the raising of blood pH, and potentially the reversal of acidosis. In addition, increases in free sodium load due to sodium citrate administration may increase intravascular blood volume, facilitating the excretion of bicarbonate compounds and an anti-urolithic effect."

This is the important aspect of what we are after:

organic citrate ions are metabolized to bicarbonate ions, resulting in an increase in the plasma bicarbonate concentration, the buffering of excess hydrogen ion, the raising of blood pH, and potentially the reversal of acidosis.

The strategy is to create citrate ions without increasing the sodium intake in a significant fashion.

the creation of sodium citrate, somewhat acts as the precursor to the reaction: baking soda + magnesium citrate >

here is a response (search was on carbonate citrate reaction, since nothing came up strong with the above, as if there is no significant reaction involved. But once calcium is part of the equation, this is of interest. It says to me the process is counteproductive, and that baking soda + citric acid is where the interest lies, not baking soda + a citrate form:

Why does Calcium (specifically Calcium Carbonate) inhibit the Citrate buffer system?

Is it because citrate chelates calcium and then is unable to react with other chemicals being added? If so can someone explain this reaction to me, or if not provide what the reason is? If anyone has any studies they've found on it that would be incredibly helpful! Thank you!

"Inhibited" is not probably the right term, as there is no catalyst inhibition, affecting the reaction kinetics.

If we consider about neutral pH range, the citrate buffer keeps pH by equilibrium reactions

HA2-?--?H++A3-HA2-+OH-?--?H2O+A3-

with pKa3=6.4

Carbonate as a base spends the buffer capacity

CO32-+2HA2-?--?CO2(g)+H2O+2A3-

If it was sodium carbonate, sodium ions would be just bystander ions.

But calcium ions form chelate with citrate 3:2, decreasing concentration of free hydrogen citrate + citrate ions.

So the overall calcium carbonate effect is being base and chelating citrate.

I.E., CALCIUM CHELATES WITH CITRATE, SO THIS WOULD TIE UP THE CITRATE. NOT WHAT IS DESIRED.

Now back to this idea, trying to get rid of sodium in the picture:

(Think about mixing magnesium sulfate + citric acid...)

Health Benefits of Magnesium Citrate - WebMD Magnesium citrate is formed by combining magnesium oxide with citric acid, a reaction that creates magnesium citrate and water. Magnesium citrate dissolves readily in water, so it may...

Now, at this point, why not just take magnesium citrate straight and forget about making it with something else.

Look at this statement again:

In addition, increases in free sodium load due to sodium citrate administration MAY increase intravascular blood volume, facilitating the excretion of bicarbonate compounds and an anti-urolithic effect."

so urolithic is the production of stones, anti-urollthic will be the reduction of stone formation.

NOTICE IT SAYS MAY. ALSO DO WE WISH TO EXCRETE BICARBONATE COMPOUNDS? Well, we know the borax polymer is certainly a carbonate compound, and we know that this is very much like a stone.

Borax again is what?

Borax is a sodium salt with boron. It is not a carbonate like baking soda or washing soda, which are. You formed a carbonate precipitate by adding sodium carbonate to the decant. That definitely formed a carbonate compound. You need to be careful here that you do enhance the formation of precipitates within the body.

What are the risks of: too many sodium ions in the body? too many sulfate ions in the body? too many carbonates in the body? too much magnesium in the body?

Lots of serious questions here.

How much salt is too much?

The U.S. Food and Drug Administration (FDA) recommends that adults eat less than 2,300 milligrams of sodium a day. That s about one teaspoon of table salt. If you eat more than that in one day, it s not going to hurt you, says Zumpano. But if you consistently exceed the recommended amount, you re eating too much, and it can impact your health.

Most Americans consume about 3,400 milligrams of sodium a day. Only elite athletes or people who profusely sweat could possibly need this much sodium.

S0

Effects of eating too much salt

A sodium-heavy diet makes you retain fluid, which leads to swelling in the short term. But far more concerning is its impact on your body over time. Excess sodium intake can negatively affect your kidney function. It also leads to high blood pressure, the top risk factor for stroke.

If you only check your blood pressure once a year at your checkup, it could be normal that day, explains Zumpano. Perhaps you ve fasted, had a cup of coffee, peed a lot, and so your blood pressure seems fine. But it might not be so normal if you were tested after eating out at a restaurant.

Signs your sodium intake is too high

It is important to know how sodium affects you, as everyone is different. After eating a salty meal, look for the following signs to see if you re more sensitive to sodium is effects.

Bloating

Sodium attracts water. If you eat a lot of salty foods, you II experience fluid retention (when sodium holds water in your body). The result? You feel swollen and look puffier, especially around the abdomen and eyes. You may also notice swelling in your hands and feet.

Increased thirst

Recent research shows higher levels of sodium don t cause people to be thirstier. But despite that, some people can t help but chug down more liquids after eating salty foods.

Rise in blood pressure

Sodium is primarily located in our blood, and it pulls water to it. So, if you eat too much sodium, more water enters your bloodstream. The greater volume of blood pushes against blood vessel walls, causing high blood pressure (hypertension).

Poor sleep quality

Eating too much sodium can mess with your sleep, especially if you eat it in the evening. A spike in blood pressure, along with urges to drink and pee, makes staying asleep difficult.

How to flush sodium out of your system

If you re feeling the effects of too much salt, there are a few ways to rid yourself of excess sodium. The very best thing to do is sweat, says Zumpano. The body naturally removes sodium through sweat, tears and urine.

To even out your sodium level, get sweaty by exercising or sitting in a sauna. Drink plenty of fluids and eat a low-sodium diet the next day, too.

Eating too much salt is bad for your health. Making a conscious effort to manage the sodium in your diet will pay off you II avoid the bloating and thirst that follows a salty meal and the risk of high blood pressure down the road. That s definitely a win-win.

So we have a case where ONE foot is swelling at night. ONE FOOT. THE AFFLICTED LOWER, LEG AND ANKLE. So the indication is that we have a salt imbalance within THAT foot, but not both feet, so not general in any sense of the word. So the question is, what is actually producing the imbalance. Sodium attracts water. It says that WITHIN that foot, there may be an excess of sodium. How and why is the question there?

One thing we do know, the vascular system in the afflicted foot is totally screwed up. We also know from NIR that the polymer withdraws the water from the skin. There is lots to think of here.

This is really getting involved, difficult and interesting to sort out. Not all today.... Look at my notes of Oct 23 2032 (handwritten notes) and we can see we did a series of ion tests on the culture. The most striking finding was the presence of calcium ions. We got a reading of 120 mg/L. This is high.

Now we will study this article in the future closely, and watch closely what it says about calcium effects and levels, and 120 mg/L is nothing to be dismissed.

In the past five decades or so evidence has been accumulating about an environmental factor, which appears to be influencing mortality, in particular, cardiovascular mortality, and this is the hardness of the drinking water. In addition, several epidemiological investigations have demonstrated the relation between risk for cardiovascular disease, growth retardation, reproductive failure, and other health problems and hardness of drinking water or its content of magnesium and calcium. In addition, the acidity of the water influences the reabsorption of calcium and magnesium in the renal tubule. Not only, calcium and magnesium, but other constituents also affect different health aspects. Thus, the present review attempts to explore the health effects of hard water and its constituents.

Keywords: Alzheimer's disease, calcium, cancer, cardiovascular disease, diabetes, hard water, magnesium, reproductive health

Go to:

INTRODUCTION

Water is essential for hydration and therefore, for life. It is also very important in food preparation and cooking, sanitation and hygiene, and a wide range of other uses. The drinking water supply has a primary objective of protecting human health, including ensuring access to adequate quantities of safe water. It is estimated that approximately 17% of the world's population uses water from the unprotected and remote sources, 32% from some form of protected sources and 51% from some sort of centralized (piped) system to the dwelling or a plot. Of the latter, a small but increasing proportion applies some form of treatment within the home. Individual water consumption occurs both at home and elsewhere, such as at schools and workplaces. Drinking-water is consumed not only as water per se but also in beverages and incorporated in food-stuffs. In response to increasing global and local water scarcity, there is an increasing use of sources such as recovered/recycled water, harvested rainwater, and desalinated water. 884 million people lack access to safe

water supplies; approximately one in eight people.[1] Among them a good percentage consumes hard water, which is considered to be a significant etiological factor around the globe causing many diseases such as cardiovascular problems, diabetes, reproductive failure, neural diseases, and renal dysfunction and so on.

Hard water is usually defined as water, which contains a high concentration of calcium and magnesium ions. However, hardness can be caused by several other dissolved metals; those forms divalent or multivalent cations, including aluminum, barium, strontium, iron, zinc, and manganese. Normally, monovalent ions such as sodium and potassium do not cause hardness. These divalent cations have a propensity to come together with anions in the water to form stable salts. The type of anion found in these salts distinguishes between the two types of hardness-carbonate and non-carbonate hardness [Table 1].

Table 1

Carbonate and non-carbonate hardness compounds

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Carbonate hardness is caused by the metals combined with a form of alkalinity. Alkalinity is the capacity of water to neutralize acids and is attributed to compounds such as carbonate, bicarbonate, hydroxide, and sometimes borate, silicate, and phosphate. In contrast, non-carbonate hardness forms when metals combine with anything other than alkalinity. Carbonate hardness is sometimes called temporary hardness because it can be removed by boiling water. Non-carbonate hardness cannot be broken down by boiling the water, so it is also known as permanent hardness. In general, it is necessary to distinguish between the two types of hardness because the removal method differs for the two. Total hardness includes both temporary and permanent hardness caused by the calcium and magnesium, on the basis of which water is categorized as soft or hard and very hard [Table 2]. The ratio of calcium and magnesium in water is also a crucial factor indicating the hardness and in the causation of several hard water health problems. Hardness generally enters groundwater as the water percolates through minerals containing calcium or magnesium. The most common sources of hardness are limestone (which introduces calcium into the water) and dolomite (which introduces magnesium.) Since, hardness enters the water in this manner groundwater generally has a greater hardness than surface water.[2]

Table 2

Concentrations of dissolved calcium and magnesium in soft and hard water

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POTENTIAL HEALTH EFFECTS

Hard water has no known adverse health effect, WHO says at its Geneva Conference. In addition, hard water, particularly very hard water, could provide an important supplementary contribution to total calcium and magnesium intake.[3] The health effects of hard water are mainly due to the effects of the salts dissolved in it, primarily calcium and magnesium. To a large extent, individuals are protected from excess intakes of calcium by a tightly regulated intestinal absorption mechanism through the action of 1, 25-dihydroxy-vitamin D, the hormonally active form of vitamin D. Although, calcium can interact with iron, zinc, magnesium, and phosphorus within the intestine, thereby reducing the absorption of these minerals. On the other hand, the major cause of hypermagnesemia is renal insufficiency associated with a significantly decreased ability to excrete magnesium.[4] Increased intake of magnesium salts may cause a change in bowel habits (diarrhea). Drinking-water in which both magnesium and sulfate are present in high concentrations (~250 mg/l each) can have a laxative effect. Laxative effects have also been associated with excess intake of magnesium taken in the form of supplements, but not with magnesium in the diet.

Cardiovascular disease

In most large-scale studies, an inverse relationship between the hardness of drinking-water and cardiovascular disease has been reported [5,6,7,8,9,10,11] However, no such association has been found in some other studies, [12,13] particularly in those involving small geographical areas a clear association is often not found.[14] The extent to which confounding variables, such as climatic, socioeconomic, or major risk factors, may account for the inverse relationship is unclear. Nevertheless, in a number of studies, a weak inverse relationship was reported after allowance was made for climatic and socioeconomic factors[15] and after major risk factors such as hypertension, smoking habits, and elevated serum lipids were taken into account.[16,17] An inverse relationship between hardness and cardiovascular disease has been reported in men after allowing for climatic and certain social factors, but only up to about 170 mg of calcium carbonate/l [18] A variety of hypotheses have been proposed to explain the possible inverse association.[19,20,21,22,23,24] However, none has been fully substantiated, nor has a particular element been found to be conclusively associated with the cardiovascular disease. It may be correlated with a high level of magnesium in hard water, which has some anti-stress actions against coronary heart disease. In a study of regional differences in mortality in cardiovascular diseases in 76 municipalities in mid-Sweden a considerable gradient was found between the western areas with a high mortality and soft water and the eastern areas with a low mortality and hard water. The hardness of water defined as the sum of the content of calcium and magnesium, was shown to be of

considerable influence of the differences in mortality compared to major risk factors.[16] The incidence of coronary heart disease varies widely in different geographical regions over the world and serious epidemiological studies have been carried out to identify variables that could explain this fact. The role of water hardness has been widely investigated and evaluated for many years in several studies have found differences in cardiovascular disease have been discussed.[16,25,26,27,28,29] Earlier studies have found positive correlations between water and dietary magnesium and calcium and blood pressure.[30,31,32,33] In Finland and South Africa it was found that the incidence of death ascribed to ischemic heart disease is inversely correlated with the concentration of magnesium in drinking water[29] and also in a Swedish case control study magnesium, and calcium in drinking water were associated with a lower mortality from acute myocardial infarction in women but not with the total incidence.[26,34] However, other studies could not confirm these findings,[31,32] conversely, in a study of magnesium in drinking water supplies and mortality from acute myocardial infarction in North-West England, there was likewise evidence of an association between magnesium and cardiovascular mortality.[25] In a Swedish study, the skeletal muscle magnesium levels were a significantly higher in persons living in an area with a higher water magnesium.[35] The concentration of magnesium in striated muscle has been used as a marker to evaluate the ion content in the soft tissue.

Cancer

Important findings in this field were provided recently by Taiwanese scientists. In most of their studies, the authors indicated a negative statistical association of various types of cancer morbidity/mortality with the hardness of water and calcium. In a review of these publications, it is worth noting the results concerning the possible association between the risk of gastric cancer and the levels of calcium and magnesium.[36] Some studies suggest there was a significant protective effect of calcium intake from drinking water on the risk of gastric cancer. Magnesium also exerted a protective effect against gastric cancer, but only for the group with the highest levels of magnesium exposure.[37] In another matched case-control study, the authors found a possible association between the risk of colon cancer and hardness levels in drinking water from municipal supplies in Japan (obtained trend analyses showed an increasing odd ratio for the cancer with decreasing hardness in drinking water). [38,39] Similar epidemiological trends were also achieved for the relations between hardness levels in drinking water, and the risk of rectal cancer and pancreatic cancer mortality, however, the researchers did not find any association with magnesium levels (the adjusted odds ratios were not statistically significant for the relationship between magnesium concentrations in drinking water and rectal cancer).[40] One of the strongest epidemiological evidences of significant protective effect of magnesium intake from the drinking water was that gave for the risk for esophageal cancer and ovarian cancer [41,42] Unfortunately, these authors did not find any results pertaining to the similar trend between drinking-water magnesium and liver cancer. The first strong evidence concerning the possible ecological relation between exposure to water magnesium and hepatic cancer was reported in Eastern Europe.[43]

Cerebrovascular mortality

Some reports suggest there is a significant protective effect of magnesium intake from the drinking water on the risk of cerebrovascular disease.[44] Despite their inherent limitations, studies on the ecological correlation between mortality and environmental exposures have been used widely to generate or discredit epidemiological hypotheses. Dietary calcium is the main source of calcium intake. Epidemiological studies have shown that dietary calcium is inversely associated with the blood pressure. With much of the epidemiological literature suggesting a relationship between the dietary calcium and blood pressure, it would seem reasonable to expect that intake of dietary calcium could reduce the risk of cardiovascular events, such as stroke that are commonly associated with hypertension.[45] However, controlling for magnesium levels eliminates the perceived effect of calcium levels on cerebrovascular mortality. In the general population, the major proportion of magnesium intake is through food, and a smaller proportion is through drinking water. For individuals with the borderline magnesium deficiency, waterborne magnesium can make an important contribution to their total intake. In addition, the loss of magnesium from food is lower when the food is cooked in magnesium-rich water. Magnesium in water can also play a critical role because of its high bioavailability. Magnesium in water appears as hydrated ions, which are more easily absorbed than magnesium in food. The contribution of water magnesium among persons who drink water with a high magnesium levels could be crucial in the prevention of magnesium deficiency. The significant association between mortality from the cerebrovascular disease and the levels of magnesium in drinking water is supported by knowledge of the functions of magnesium. Magnesium is an enzyme (Na+/K+ ATPase) activator and regulates cellular energy metabolism, vascular tone, and the cell membrane ion transport. A lack of magnesium leads to a decrease in the concentration of intracellular potassium and an increase in calcium levels. Magnesium deficiency may increase the contractility of blood vessels. Magnesium causes vasodilation by stimulation of endothelial prostacyclin release and in vivo, prevents vasoconstriction of the intracranial vessels after experimental subarachnoid hemorrhage. In addition, fear of cerebrovascular disease should not deter anyone from drinking water with a low magnesium levels. In conclusion, the results of the present study show that there is a significant protective effect of magnesium intake from drinking water on the risk of cerebrovascular disease. This is an important finding for the Taiwan water industry and human health risk assessment.[46]

Malformations of central nervous system

There is good evidence that environmental influences must play some part, possibly a major part, in the etiology of neural tube malformations in the human embryo. Almost all that evidence, however, relates to

non-specific and uncertain markers of as yet unidentified specific teratogens. For example, the frequency of malformations of the central nervous system varies greatly from country to country.[47,48] It also varies from area to area within countries: In the United States for the period 1950-59, mortality from spina bifida (myelomeningocele) was 2-3 times greater on the Atlantic coast than on the Pacific coast;[49] in South Wales the frequency of central nervous system malformations in the coal-mining valleys is almost twice as high as in the coastal plain; [50] in England and Wales as a whole the frequency is highest in the north, the north-west, and Wales and lowest in the East, Southeast, and South.[51,52] It is higher in first-born than in later born infants and in infants born to younger and older mothers than to mothers in mid-reproductive life.[51,53] It is higher among infants born in the poorer than in the well-to-do strata of society [54,55,56] It tends to be higher among winter than among summer births. [51,53,55,57,58,59] Striking secular swings in frequency have been reported in the Birmingham, Scotland, Dublin, and Boston. 70-71 Penrose[48] seems to have been the first to speculate the geographical variations observed in the incidence (of anencephalus) might suggest a causal agent, such as the presence or absence of trace elements in the water supply. This suggestion has been taken up by Fedrick.[60] She related data on anencephalus for 10 different areas in the United Kingdom (from 10 different studies and relating to 10 different time-periods) to information about the water supplies of those areas obtained from various sources. Despite such manifestly unsatisfactory data, she found that the frequency of anencephalus was a significantly related to measurements of the total hardness, calcium content, and pH of the local water supplies. Stocks[52] examined the mean annual death rates (still-births plus infant deaths) for congenital malformations in the 15 hospital regions of England and Wales. Mortality rates were highest in the north and the west and lowest in the Southeast. Having observed that mortality from cardiovascular disease followed much the same regional pattern, he proceeded to correlate death rates from congenital malformations in the 15 regions with death rates of women aged 25-54 from the certain causes in the corresponding regions. He found that mortality from malformations of the neural tube correlated very closely with mortality from the cardiovascular diseases, whereas other malformations produced insignificant negative correlations. He concluded that because mortality from cardiovascular disease in the county boroughs of England and Wales has been shown to be strongly an associated with softness of their water supplies, [61] a water factor might be responsible for the regional variations of mortality from the central nervous system malformations. In this paper, and against this background, we relate area differences in mortality from malformations of the central nervous system in South Wales to estimates of the hardness of the water supplies in these areas. We also present new data on perinatal mortality from anencephalus in the county boroughs of England and Wales and relate them to estimates of the hardness of their water supplies.

Alzheimer's disease

The issue of aluminum as a cause for Alzheimer's disease has been contentious. In special circumstances such as renal failure and massive exposure to aluminum in certain occupations aluminum may cause brain pathology similar to Alzheimer Disease. However, there is no definite evidence of the role of this metal in the causation or development of Alzheimer disease. In a survey of 88 county districts within England and Wales, rates of Alzheimer's disease in people under the age of 70 years were estimated from the records of the computerized tomographic (CT) scanning units that served these districts. Rates were adjusted to compensate for differences in the distance from the nearest CT scanning unit and for differences in the size of the population served by the units. Aluminum concentrations in water over the past 10 years were obtained from water authorities and water companies. The risk of Alzheimer's disease was 1.5 times higher in districts where the mean aluminum concentration exceeded 0.11 mg/l than in districts where concentrations were less than 0.01 mg/l.[62]

Diabetes

Hard water is indicative of the presence of higher levels of magnesium. In certain areas, drinking water actually contains 100% or more of the recommended daily allowance about magnesium, which is around 300-400 mg daily with levels varying according to gender and age. Because, all kinases and other ATP-related enzymes and channels regulating insulin action are dependent on magnesium, it is not surprising that serum magnesium concentrations have been found to be decreased in non-diabetic subjects with metabolic syndrome and that hypomagnesaemia is a common feature in subjects with type-2 diabetes. Whether the low intracellular magnesium content is secondary to or precedes insulin resistance is unclear; however, recent evidence suggests that sub-clinical magnesium deficiency may precipitate a diabetic state. Studies are needed to determine the role of sub-clinical magnesium status in diabetes risk. This should include measures of glycosylated hemoglobin, an indicator of glycemic control that has been found to respond to oral magnesium supplementation and to correlate negatively with serum ionized magnesium or serum total magnesium in type 2 diabetics.[63]

Childhood atopic dermatitis

Atopic dermatitis (or eczema) is an inflammatory, chronically relapsing, non-contagious and pruritic skin disorder. The environment plays an important part in the etiology of atopic eczema, but the specific causes are unknown. Exposure to hard water is thought to be a risk factor for eczema. The prevalence of symptoms of atopic eczema among Japanese, Nottinghamshire and Spanish children is the most. The reasons for such a high prevalence are unknown. The study used data on water hardness and chlorine content of the water supply; prevalence of atopic dermatitis diagnosed by physicians and episodes of wheezing reported by the parents; and potential confounding factors by socioeconomic and health-care status per municipality. The prevalence of atopic eczema was significantly higher in the highest water hardness category than that in the

lowest respectively. A significant relationship between the chlorine content of the water supply and the prevalence of atopic dermatitis was observed after adjustment for confounding factors. Water hardness may increase the risk of atopic dermatitis among elementary-school children in Japan, as well as in the United Kingdom.[64]

Kidney stones

The hardness of water is due to the presence of carbonate and sulfate salts of calcium and magnesium. More than 3/4th of kidney stones are generally composed of calcium salt and usually occur as calcium oxalate and less commonly as calcium phosphate. The remaining 20% of stones are composed of uric acid, struvite and cystine stone. Stones form in urine that is supersaturated and this saturation is dependent on chemical free ion activity, which makes the urine under-saturated. In this situation, the stone will not grow and may even dissolve. Increased urinary ion excretion and decreased urine volume will both an increase free ion activity and favor stone formation and growth. Formation of kidney stones (nephrolithiasis) is based on genetic, metabolic, nutritional and environmental factors. Metabolic factors involved in stone formation include hypercalciuria, hypocitraturia (due to renal disease), hyperuricosuria, hyperoxalaturia, cystinuria and infections. Environmental and nutritional factors include dehydration, high salt intake, a diet rich in animal proteins and calcium rich diet when oxalate intake is restricted. The impact of water hardness of urinary stone formation remains unclear, despite a weak correlation between water hardness and urinary calcium, magnesium, and citrate excretion. Several studies have shown no association between water hardness and the incidence of urinary stone formation. A correlation between water hardness and urinary calcium, citrate, and magnesium levels has been observed although the significance of this is not known. Some studies suggest that in the preventive approach to calcium nephrolithiasis, intake of soft water has been preferable to hard water since it is associated with a lower risk for recurrence of calcium stones.[65]

Reproductive health

There are few reports of the effect of water hardness over reproductive health of men, most of them emphasized on the effect of its constituents, calcium, and magnesium,[66] while others on some other constituents like fluoride.[67] However, some reports show the occurrence of reproductive failure and stillbirth in India in hard water regions of India.[68,69] Some of these are showing the effect of excess calcium on the reproductive system and its negative influence on fertility.[68,70,71] These reports demonstrated the oxidative stress induced infertility in men by calcium,[72] but showed beneficial effects of magnesium.[4] There are also some reports of effects of waterborne fluoride on growth, reproduction and survival which showed long-term exposure of fluoride causes a progressive decline in reproduction.[67] On the contrary, in female, magnesium sulfate present in hard water is indicated to prevent eclampsia in patients with pre-eclampsia. Magnesium sulfate decreases the risk of developing eclampsia around 50% and also decreases maternal mortality.[73] The WHO considers that magnesium sulfate is the elective drug for the prevention of eclampsia in patients suffering from pre-eclampsia. Magnesium sulfate has also been demonstrated to prevent preterm labor.[74]

Digestive health and constipation

Even GI health is also reported being benefited from hard water since it provides potentially alleviating effects on the onset of constipation in the 85% cases. A rich union of calcium and magnesium in hard water, in a right combination, helps to combat constipation. The calcium in hard water results in teaming up with excess bile and its resident fats to lather up the soap like insoluble substance, which is emitted from the body during bowel movements. Indeed, many renowned scientists have considered hard water as a boon as it has some fantastic health benefits that seem to encourage longer life expectancy and improved health. Magnesium salt represents with a laxative effect. This provides a rapid evacuation of intestine. Magnesium citrate, magnesium phosphate, and magnesium hydroxide are also used. The American Gastroenterology Association recommends milk of magnesia for the management of constipation as one of the therapeutic options; however, the Rehabilitation Nursing Foundation discourages the routine use of saline magnesium laxatives due to possible side effects such as abdominal cramping, watery stools, and potential for dehydration and hypermagnesium. They only indicate the use of these laxatives in end-stage patients when other options have failed, and with and adequate prospective evaluation of magnesium levels.

Bone mineral density

The correlation between calcium and magnesium in drinking water and its impact on bone health are unidentified. There is some evidence that high-calcium water is beneficial to bone.[75] It has been reported in a study that spine mineral density was significantly higher in women aged 30-70 years living in Sangemini, a region of central Italy, who drank the local high-calcium water (318 mg/l), compared with women in the same region who drank low calcium water (

Other health effects

The results of several studies have suggested that a variety of other diseases are also inversely correlated with the hardness of water, including anencephaly[78,79] and various types of cancer.[80,81,82,83,84,85,86] However, the significance of these results is unclear, and it has been suggested that the associations may reflect disease patterns that can be explained by social, climatological, and environmental factors, rather than by the hardness of the water.

Go to:

CONCLUSIONS

Hardness is important for drinking-water from the point of view of both aesthetic acceptability and operational considerations. Although, there is some evidence from epidemiological studies for a protective effect of magnesium or hardness on cardiovascular mortality, the evidence is being debated and does not prove causality. Further studies are being conducted. In spite of this, drinking-water may be a source of calcium and magnesium in the diet and could be important for those who are marginal for calcium and magnesium intake. Where drinking-water supplies are supplemented with or replaced by dematerialized water that requires conditioning, consideration should be given to adding calcium and magnesium salts to achieve concentrations similar to those that the population received from the original supply. Consumers should be informed of the mineral composition of their water when it has been altered by piped suppliers or treatment device manufacturers and by means for supplementing if desired. The contribution of drinking-water minerals for mineral nutrition should be considered where changes in supply are proposed or where novel sources, such as seawater or brackish water, are exploited for drinking-water. There are insufficient data to suggest either minimum or maximum concentrations of minerals at this time, and so no guideline values are proposed.

MOST DEFINITELY TO BE CONTINUED...

Let's start looking at making probability estimates of the various functional groups now. Probability estimates are looking very reasonable and conservative at this point. 70% probablility level is probably quite low for such an extensive data set. RMS looks to capture the data set at a much higher level, eg 80, 85%.





2023.12.08 Functional Group Worksheet

Continuing:

From Nov 16 2023 Notes:

This exists as the most current interpretation of this material that has been created, a polymerized protein. Unclear at this point if the polymerization is likely to come from aromatic or amide origin, or both for that matter. We know that in the end a "rubberized" polymer is developed, but this can come from the other TSP surface synth-cell polymer layer. But out of curiosity, is rubber aromatic or amide based?

Rubber is alkene based. OK, this gets very interesting. Synthetic rubber is a copolymer between styrene and butadiene. Styrene is a VINYL group attached to an aromatic ring. Butadiene is an alkene.

Guess what, all the signs are starting to point the same direction. Aromatic, vinyl, alkenes are the components of synthetic rubber, which is most characteristic of the mature clot gross properties. Aromatic, vinyl, alkenes appear to be dominant structures of our NIR plots. Do not keep trying to force a single functional group. The combination of these three functional groups along with synthetic blood go a long way toward explaining the culture findings in general.

So our three main components (everything is temperature and pH dependent for expression (~18 expressions thus far) are:

- 1. Synthetic blood (w/hemoglobin) (scores of proteins within blood) (surface layer of mature culture)
- 2. A set of proteins that can be polymerized with borax, structurally firm but not elastic. (within decant)
- 3. A vlnyl contribution for elasticity of the culture. (within precipitate)

This makes up the majority of the clot structure.

1,3-Butadiene (a synthetic rubber) from NIST data base UV spectral search shows a peak at 293. This is a vinyl compound and then a match for our spectrum of TSP 100.6 at this point (not the rest of the spectrum nor our 232 peak found.

NIST and Chemspider both have some UV searching capability if you can get a part of the name. VERY VALUABLE!

The styrene UV spectrum explains high absorbance towards the 232 peak. Synthetic rubber is a copolymer between styrene and butadiene.

I must have looked up the UV on NIST for styrene based upon leads. Let's do this again.

What are the physical properties of styrene? (AI)

The physical properties of styrene include:

- 1. Appearance: Styrene is a clear, colorless liquid.
- 2. Odor: It has a sweet, floral, and somewhat fruity odor.
- 3. Melting Point: The melting point of styrene is around -30.6 degrees Celsius (-23.1 degrees Fahrenheit).
- 4. Boiling Point: Styrene has a boiling point of around 145 degrees Celsius (293 degrees Fahrenheit).
- 5. Density: The density of styrene is approximately 0.91 grams per cubic centimeter.
- 6. Solubility: Styrene is soluble in a wide range of organic solvents, such as ethanol, acetone, and benzene.
- 7. Vapor Pressure: It has a relatively low vapor pressure at room temperature.
- 8. Flammability: Styrene is highly flammable and can form explosive mixtures with air.
- 9. Toxicity: Exposure to styrene vapor or liquid can cause irritation to the eyes, skin, and respiratory system.

Prolonged or high-level exposure may have more severe health effects.

It is important to note that these properties may vary slightly depending on the specific grade or purity of the styrene.

what is the UV absorbance of styrene? (AI)

The UV absorbance of styrene can vary depending on factors such as concentration, solvent, and wavelength of light used for measurement. Generally, styrene exhibits weak absorption in the UV region. Some studies have reported a maximum absorption peak around 250-260 nm. However, it is recommended to consult specific literature or experimental data for accurate and detailed information on the UV absorbance of styrene under specific conditions.

This shows you that there is a lot of variability in the assessment of UV absorption of sytrene. Anywhere from 232 (apparently? up to 260). That is a wide range. Profile of absorption is therefore important also.

So where and how and why did I make the comment about styrene at 232 nm?

Guess what, I can create my own spectrum to get me in the ball park with styrofoam. It is a form of polystyrene. Great.

But we have this problem:

"HCL will not melt the styrofoam. Needs to be petroleum based. For that matter, you could pour battery acid (sulfuric acid - H2SO4) in a styrofoam cup and it would not melt the cup. Both Hydrochloric and Sulfuric acids are inorganic (they do not contain Carbon) Styrofoam does. . First rule in Chemistry-101, like dissolves like."

This is important as it also affects interpretation of clot, synth-polymer layer, and the borax polymer. Need to now revisit all of that.

So acetone and gasoline work, but acetone may damage the quartz cuvettes? Let' see. At \$20 a pop, I cannot afford that.

So yes, acetone will damage quartz cuvettes. However, I can afford plastic cuvettes and this will get me down to 230 nm which should be adequate. So let's try that.

Behavior of sample was strange so I reconstructed the control series, in two stages.

1. 1st control is H2O.

2. 2nd control is H2O + 100 ul acetone.

3. Sample is H2O + 100 ul (acetone + sytrofoam mix) and then sample with H2O + 300 ul (acetone + styrofoam mix).

Acetone has its own distinctive UV profile but that is to be dismissed and disregarded here. The only impact of the styrofoam was to shift the strong acetone peak at \sim 288 nm to:

1. 290 nm with the 100 ul addition of the styrofoam.

2. 298 nm with the 300 ul addition of the styrofoam.

So there is nothing real dramatic here with, either way, no noticeably different peak behavior in the 230 - 260 nm range. So these results are somewhat ambiguous.

I find this information:

Polystyrene (PSt) exhibits a new UV absorption band at around 290nm. https://link.springer.com/article/10.1007/BF00310794

which is what we get for the lower concentration sample (slight shift from acetone). However, as we notice, a stronger concentration shifted the peak further to the right. Nevertheless, from what we find and measure directly

The UV peak of styrene looks to be about 244. Not exactly 232. Remember though that this is a borax polymer, it is solid and inflexible, so you do not expect it to be of rubber nature.

So this says styrene is some where in the neighborhood of 240-260, depending on who you are listening to, and that polystyrene is somewhere on the order of 290 - 300 depending upon concentration, at least as dissolved in acetone.

This is far too much variation for me and therefore attributing the peak at 232 to styrene seems useless and fruitless to me. You might try an NIR plot but I see no value in UV here.

NOTE:

The styrene UV spectrum explains high absorbance towards the 232 peak [NOTE DEC 08 2023 I SEE NO BASIS FOR THIS PREVIOUS STATEMENT - IT IS NOT ACCEPTED AT THIS TIME].

Let's carry on keeping the styrene proposition in mind but not seeing any data collection for that compound yet. I think we need a NIR spectrum.

OK, NIR looks like a great way to approach this problem. Dissolving styrofoam in acetone created a residual polymer material which did not remain in solution, It truly does form a visible plastic. Making an NIR card of this looks to be the best approach without any doubt. It is a good solid sample to work with, but in a putty-plastic form. The solution approach seems to be much harder to extract from. The NIR card should be a good test.

I have an excellent NIR collection on styrofoam (polystyrene). Let's see what it shows.

Polystyrene (Styrofoam) CI

1146 (moderate) ArCH (Aromatic CH) (1143)(1142)
1192 (weak) Methyl (1194) (1195)
1405 (moderate) OH Methanol - OH with Hydrogen Bonding (1408) Methylene (1395)
1638 (weak) CH from vinyl group (CH2=CH-) CH from vinyl associated group (1637) CH Vinyl - Aliphatic Hydrocarbons (1635)
1682 (strong) Aromatic CH (1680) (1685) CH Methyl (1682)

Certainly looks to be on track.

Now look at notes of Nov 18. Instead of thinking only aromatic protein, with what you know about polystyrene and the dominant compoent of aromatic CH, you see that a progression of the following may well be in order:

1. water soluble protein and vinyl groups (are there aromatics here as well?)

2. non water soluble polyamide (like nylon, etc), not just consideration of an aromatic protein

3. rubber clot (vinyl and styrene).

"I have a very significant plot now to look at. It is a NIR comparison of TSP 100.5 (Decant) over a 12 day incubation period. This is from the time that it is decanted (amber color) until it turns white approx 10 days later.

Main feature I am looking at is that appears there is a bulk of mass (absorbance) shift from the Nov 06 TSP 100.5 (amber) at a very broad peak of \sim 1620 +/- 20 nm to a more focused peak at Nov 18 TSP 100.5 (White) of 1516. They do however, continue to share a great deal, and in fact the majority of the turf from 1530 - 1650 nm.

Interpretation of this is: 1620 is alkene and vinyl 1520 is protein, 1515 is polyamide

this makes a lot of sense. It is saying it starts out stronger in the vinyl section but then shifts over to a protein nature. However, it still retains the vinyl nature. This means that a conglomerate is being formed, i.e. a polymerized protein as your surmise.

This is exactly what you envision happening. This answers a very important question on culture dynamics."

What is really important here is the demonstrated transition or ships from alkene and vinyl (alcohol also? aromatic also?, tyrosine, etc) all water soluble towards a protein and polyamide, which we know to be insoluble. This is a critical feature to demonstrate.

IT IS POLYMERIZING.

Now revisting the nucleic acid - DNA issue. This still remains as a distinct possibility. You now have two mature cultures to compare to one another TSP 070 and TSP 100. We see that as the culture matures, over 1-2 months, the middle layer by UV presents a spectrum that seems to coincide quite strongly with nucleic acid - DNA properties. Even if we restrict ourselves to the 260/280 ratio we find:

1. TSP 100: Nov 08 2023 (1 month age)

.77/.57 = 1.35

2. TSP 070 (Oct 08 2023 (2 months age)

1.62 / 1.23 = 1.32

Both samples were restricted to 300 ul in 3ml H2O.

This presents an estimate of DNA (Nucleic acid) concentration of 15 % DNA, 85% Protein.

Corrleation of profile with Nucleic Acids can also not be dismissed.

This situation simply does not correspond with protein only. The profile as is cannot occur in a protein only environment and the peak for TSP 070 as a 60 day old culture is extremely well defined and far away from 280 region. 280 region also is not peaking in any way as 260 area is. I say you have a strong case of nucleic acids or DNA being present.

Concentration graph (now modified) image source is: https://www.agilent.com/cs/library/applications/A260A280-spectral-scanning-5994-2538EN-agilent.pdf

Alcohol test. NIR test.

2023.12.09 Functional Groups DNA

I now have the NIR of the middle layer of TSP 070 and TSP 100 for the nucleic acid inquiry. This will be itemized here but preliminary review shows that it supports the claim for nucleic acid/DNA existence.

Also the following links are going to be helpful:

https://bio.libretexts.org/Courses/University_of_California_Davis/BIS_2A%3A_Introductory_Biology_(Britt)/01%3A_Readin

https://socratic.org/questions/what-are-the-functional-groups-of-dna

https://biologydictionary.net/hydroxyl-group/

NIR TSP 100 (30 day culture) & TSP 070 (60 day culture). Both spectra are essentially identical, but TSP 070 (older culture) is more clearly defined in general.

1200 nm (moderate) OH (1200) hydroxyl

1505 nm (strong) aromatic amine (1502.5) alkyl alcohol, containing one OH, R-C-OH (1500) NH combination band (NH2-C=O - NH2 - urea)

1597 nm (strong) polyamide (C=O/N-H combination) (1598)

1642 nm (weak) vinyl group (CH2=CH-) (1637)

1692 nm (moderate) Protein (CONH2) (1690) Aromatic CH (1689) Methyl (1693-1695) 8 instances

This is an important discerning NIR plot. The first stage of analysis is to acquire the functional groups of both proteins and DNA. I will discern from the NIR plot which of those exist and what can be associated with protein or DNA. All functional groups will enter into the worksheet, however. The question confined here is whether or not the DNA/nucleic acid prospect is supported above and beyond the UV 260/280 finding.

The links now become important.

The functional groups of DNA are:

The functional groups are amine, amide, hydroxyl, glycoside linkage, and phosphodiester

The functional groups of protein are:

Amino acids are the monomers that make up proteins. Each amino acid has the same core structure, which consists of a central carbon atom, also known as the alpha (a) carbon, bonded to an amino group (NH2), a carboxyl group (COOH), and a hydrogen atom.

Peptide bond formation is a condensation reaction. The carboxyl group of the first amino acid is linked to the amino group of the second incoming amino acid. In the process, a molecule of water is released and a peptide bond is formed. Try finding the backbone in the dipeptide formed from this reaction. The pattern you are looking for is: N-C-C-N-C-C

The sequence and the number of amino acids ultimately determine the protein's shape, size, and function. Each amino acid is attached to another amino acid by a covalent bond, known as a peptide bond, which is formed by a dehydration synthesis (= condensation) reaction. The carboxyl group of one amino acid and the amino group of the incoming amino acid combine, releasing a molecule of water and creating the peptide bond.

and this is a crucial statement:

Amino acids are the function units of protein. Each amino acid molecule contains a carboxyl group at one end, and an amino group at the other end. Amino groups consist of a nitrogen bonded to two hydrogens, which is attached to the carbon backbone of the amino acid. When the protein is formed, the carboxyl group loses the hydroxyl group attached to it, while the amino group loses a hydrogen. With the loss of these molecules, the amino group binds to the carbonyl group, forming a peptide bond. What else is produced during this reaction?

So after the protein is formed, the hydroxyl group no longer exists, and water is formed in the process. Our sample has been dried, however, and all water removed. Therefore there should be no hydroxyl group present in our NIR sample.

However, DNA maintains a hydroxyl group. Our NIR plot is therefore further supportive of the DNA prospect within the TSP cultures, above and beyond the UV which shows a strong 260/280 ratio presence. This is a critical assessment.

The fully chilled ethanol DNA tests (both sample and ethanol are chilled prior to mixing) are fully successful in indicating the presence of DNA. There is a white layer formed at the alcohol-sample interface. Material produced is minimal but the appearance is fully in keeping with all previous DNA control tests over the years.

All three DNA inquiry tests are with positive results:

- 1. UV 260/280 absorbance ratio.
- 2. NIR Hydroxyl group detection
- 3. Chilled alcohol tests.

This is an important achievement. Bear in mine that DNA extraction was succussful on 3 prior events in past years, so this is once again fully consistent. What is different here is that the method derives from a water soluble portion of a mature culture. No solid extraction processes were required.

Now we return to the worksheet.

These are the NIR peaks of TSP 100.5 identified on Nov 18 2023: (CONCENTRATED Decanted TSP 100.0 post Titration)

TSP 100.5 NOV 18: 1397 1520 1598 1646

1397 Methyl (1396) Methylene (1395) 1520 Amide/protein (1520) Amine (1520) 1598 Polyamide (C=0/N-H) (1598) 1646 Vinyl (1637) Methyl (1654)

The acid-base behaviors of the:

- 1. synth polymer
- 2. carbonate precipitate

3. rubberized blood clot

need to be sorted out.

2023.12.10 Functional Group Worksheet

Continuing, now with Nov 23 2023.

The acid-base behaviors of the:

1. synth polymer

2. carbonate precipitate

3. rubberized blood clot

need to be sorted out.

Human blood-synth layer comparison to continue.

Bacteriophage issue has not been addressed properly.

Mid IR exploration of vinyl group with historical "CDB Protein Complex" IR data (cerca 2015) looks to be most important. Initial peak match search brings up following candidates as:

1. Butyl vinyl ether C6H12O

2. Cyclohexyl vinyl ether C6H14O

These candidates have essentially exact matches with IR collected Sep 2015 of "CDB Protein Complex". Out of about 10 candidates from entire SDBS database. These are lowest molecular weight. I am guessing we have something here.

Look at physical properties, synthetic rubber, etc.

Combination of human-synth blood comparison with polymerization probable is a worth topic.

A new variation on the growth forms. The fungal filament form is well established. I do have a major culture of only that form that has been developed over the last 2-3 months. It creates a large mass of mold--fungal-filament appearing material.

What is different today is that a spare portion of TSP 100 set aside is producing a pure large growth form on its surface, approx 2" x 1". A large central mass with appendages extending out on the surface. Colors are grey, white, pink and blue grey. It is a very clean form of this variation that comes fom a purified TSP 100 culture of \sim 30 days age. This was overflow material from the culture that has been sitting idle. This culture produces the synth-polymer layer also. It will be of interest if the synth cells can be identified within such a mass, as if so, it represents a clotting form in itself. It is solid. Database image included.

This is a major topic that looks like it was never followed up with, notes are from Nov 27 2023:

I am now in a position to start comparing the NIR of a blood clot to the NIR of the synth polymer layer. The main uncertainties of comparison are:

1. There is some ambiguity whether the clot comes from the deceased individual or a live individual (externally clotted). The bias is strongly towards the deceased clot which is favorable, however this will likely be repeated. 2. The trend is removed from the blood clot but not from the synth-polymer layer so no magnitudes of any kind can be considered.

With that being said, here is what we start to see. The main issue here involves similarity or difference in basic structure and solubility. One thing we do know is that the clot dissolves in NaOH and the synth polymer layer dissolves in strong H2SO4. So we know already they are very different from one another and yet the presumption is that they originate generally from the same source material. This means, since we knowe they have important differences, that comparison of the clot with other forms observed will be made.

NIR Comparison:

Start with the blood clot:

2023.12.10 Functional Group Worksheet

Clot: 1130 (1130) 1374 (1370) 1443 (1440) `1575 (1570, 1580)

and then the synth-polymer layer: Synth-Polymer: 1195 (1190, 1200) 1520 (1500) 1594 (1590) Broad 1582-1609 (1580-1610) 1700 (1700)

Clot:

1130 (1130) ArCH (1142, 1143)

1374 (1370) Methyl (1370)

1443 `Aromatic Amine (1443) (1445) (1441) (1446) OH from sugar (1441) Methylene (1440) ArCH (1446)

1575 (1570, 1580) Amide (1570) Polyamide (1570) Alcohol (1580) OH (1580)

We are now at a very important juncture point in the work, and this is the NIR analysis of the blood clots that took place approximately in Jun of 2023. First step is to recall the notebook volume(s). It appears to be Vol 30. Jun 22 2023 mentions receipt of blood clots.

The first plot visited is that of the Live Individual, Vaxxed. Plot is detrended, normalized. All of the above matches fine, but two more peaks are to be added:

1504 Aromatic Amine (1502.5)

1626 Vinyl, Vinyllidene (1630) (1621) (1631)

I see now that the NIR of the blood clots must be revisited. I do not have all data that could be available. First issue is that there is some confusion on which sample is being used above. All direct statements state that it is Sample C, which is from a living individual whose blood was allowed to settle outside the body and formed a clot. My notes (i.e., NIR plot) sometimes refer to it as from the deceased individual and this is where it becomes confusing. I do think it is from Sample C notes of Jun 22 2023.

We have enough information to establish the overlap of the culture with the blood clot. But let's look at the clots again and attempt to prepare NIR cards for each sample that we have.

I have now prepared NIR cards of all three clot samples, L, C, D.





2023.12.11 Functional Group Worksheet

Lots of data now available to interpret. chi-square application? I have vinyl gloves here - solubility tests. Are DNA results included in worksheet? Variability of clot NIR, additional clots. Protein, polymer, synth blood, filaments are all in place.

I have a strong NIR plot of the deceased clot. One question I have is variability between other clots however the deceased clot seems to be the epitome of the end of the progression under study. Rubber nature is strong with this clot.

Blood Clot Deceased:

935 Methylene (930) (930) weak

984 ArOH (990) Water (979) Not Likely, sample is dried. strong

1179 Alkenes, polyenes (1170) weak

1422 ArOH (1420) Phenolic OH (1420) Aromatic CH (1417) strong

1454 Aromatic Amine (1452.5) Water (1453) Not Llkely, sample is dried Alcohol (1452) weak

1518 Amide/Protein (1520) Amine (1520) Polyamide (1515) strong

1648 Methyl (1654) Vinyl (1637) strong

1682 Methyl (1682) Aromatic CH (1685) moderate

This certainly looks like a very representative sampling of the functional groups encountered.

OK, the analyis of clot vs., culture progression is complete.

ON TAP:

1. Finish paper.

- 2. Bacteriophage inquiry.
- 3. Chi square investigations.
- 4. Binomial test?

The paper is proceeding well and falling into place at the level required. This will give me some breathing room.

CI BLOOD CLOT PROGRESSION WORKSHEET (not original formatting)

Native Form Native Form Native Form Date of Methods of Date Origin Age In H2O H2O pH HYDROCARBONS ALCOHOLS SDBS PROTEIN POLYMER Study Sample Name Study of Sample Days Soluble Insoluble (0 if Unknown) Compound 1 Compound 2 Compound 3 Compound 4 Compound 5 Compound 6 Compound 7 Compound 8 Compound 9 Compound 10 Compound 11 Compound 12 Compound 13 Compound 14 Compound 15 Compound 16 Compound 17 Compound 18 Compound 19 Compound 20 Compound 21 Compound 22 Compound 23 Compound 24 Compound 25 Compound 26 Compound 27 Compound 28 Compound 29 Compound 30 Compound 31 Compound 32 COMMENTS

10/28/23 TSP 070 Decant LC, UV, NIR 10/08/23 20 1 acidic Alkyl Alcohol Ether Alcohol Ester Alcohol Protein Tyrosine Calcium Ions Nanoparticles 10/29/23 TSP 070 Decant LC, UV, NIR 10/08/23 21 1 acidic Alcohol Protein Tyrosine 10/29/23 TSP Carbonate Decant LC, UV, NIR 10/08/23 21 1 acidic Methyl Alcohol Alkyl Alcohol Protein Amide Aromatic CH Aromatic Amine Methyl Alcohol Aromatic Amine 10/30/23 2nd Level Decant from Carbonate Decant UV 10/08/23 22 1 12.0 Protein 10/30/23 2nd Level Precipitate from Carbonate Decant - H2SO4 Hydrolyzed UV 10/08/23 22 1 acidic Protein 10/31/23 Human Blood NIR 10/31/23 0 1 7.4 est Vinyl Vinylidene Vinyl Vinylidene 10/31/23 Synthetic Blood - Polymer NIR 10/08/23 23 1 acidic Methyl Alcohol Amine Polyamide Aromatic Amine Polyamide 11/02/23 Synthetic Blood - Polymer Meter 10/08/23 25 1 acidic Hemoglobin Meter Kastle Meyer Hemoglobin Kastle Meyer 11/03/23 Human Blood - REFERENCE MID IR 08/16/15 3001 1 7.4 est Ethyl Ether Vinyl High level confirmation of ethyl vinyl ether (or related compounds) in the blood of 2015 and 2017 with MID IR 11/04/23 Synthetic Blood - Polymer - (H2SO4 Hydrolyzed) VIS-NIR 10/08/23 27 1 solid Methyl Alcohol Alkyl Alcohol Aromatic CH Methyl Aromatic CH 11/05/23 Synthetic Blood - Polymer - (H2SO4 Hydrolyzed) DNA PROSPECT UV 10/08/23 28 1 solid Nucleic Acids - DNA Nucleic Acids - DNA 11/07/23 TSP 100.5D (M) 7.20-1110 CONCENTRATED Decanted TSP 100.0 post Titration NIR 11/06/23 1 1 acidic Alkenes Vinyl 11/07/23 TSP 70.1 Titrate Decant UV 10/08/23 30 1 acidic Alkyl Alcohol Bacteriophage? 11/07/23 TSP 70.5 Centrifuged TSP 70.0 Bottom Layer NIR 10/08/23 30 1 solid Methyl Alkyl Alcohol Protein Amide Aromatic Amine 11/07/23 TSP 70.2 Centrifuged Titrate Precipitate NIR 10/08/23 30 1 acidic Methylene Polyamide Vinyl Vinylidene Acrylate Polyamide Vinyl Vinyl Vinyl Vinyl 11/13/23 TSP 100.6C (B) X.XX-1110 Centrifuged Precipitate TSP 100.0 post Titration NIR 11/10/23 3 1 acidic Methyl Alkyl Alcohol Vinyl Two layers grey/white centrifuge precipitate, grey dominates 11/13/23 TSP 100.5D (M) 7.20-1110 CONCENTRATED Decanted TSP 100.0 post Titration UV, VIS-NIR 11/10/23 3 1 acidic Protein Tyrosine Amber solution 95% of volume 11/13/23 TSP 100.0N (M) 3.81-1108 VIS-NIR 11/08/23 5 1 acidic Alkyl Alcohol Protein 11/13/23 Urine - Polymer (Blood) Matrix Vinyl (Polymer Deduction) 11/13/23 TSP 100.5D (M) 7.20-1110 CONCENTRATED Decanted Polymer Suface Layer Microscope 11/10/23 3 1 solid Protein Tyrosine Vinyl (Observation 250nm) Amber solution 95% of volume 11/15/23 Borax Polymer (developed in lab from TSP 100.5 & Borax) NIR 11/10/23 5 1 solid Methyl Methylene Alkenes Phenolic OH Protein Amine Amide Polyamide Aromatic CH Butadiene (293 nm) Polyenes 11/18/23 TSP 100.5D (M) 7.20-1110 CONCENTRATED Decanted TSP 100.0 post Titration 11/10/23 8 1 acidic Methyl Protein Amine Amide Polyamide Vinyl 11/20/23 TSP 070 UV 10/08/23 43 1 acidic Nucleic Acids - DNA 11/20/23 TSP 100 UV 11/08/23 12 1 acidic Protein Tyrosine 11/21/23 TSP 070 UV 10/08/23 44 1 acidic Nucleic Acids - DNA 11/22/23 Synthetic Blood - Polymer NIR 10/08/23 45 1 solid Amine Amide Polyamide Vinyl 11/23/23 CDB Protein Complex 2015 MID IR 06/01/15 3097 1 film Butyl Ethyl Ether (SDBS) Ethyl Vinyl Ether (SDBS) Cyclohexyl vinyl ether C6H14O (SDBS) 11/23/23 Synthetic Blood - Polymer - (H2SO4 Hydrolyzed) TSP 070 UV 10/08/23 46 1 solid Protein Amide Nucleic Acids - DNA 11/23/23 Synthetic Blood - Polymer - TSP 070 - Dried. Combined with H2SO4 Qualitative 10/08/23 46 1 solid Hemoglobin 11/09/23 TSP 070 Native pl Determination - Match Human Blood Proteome Titration 10/08/23 32 1 acidic Hemoglobin 11/24/23 Synthetic Blood - Polymer - (H2SO4 Hydrolyzed) - Purple - Sulfhemoglobinemia NIR 10/08/23 47 1 solid Methylene Phenolic OH Hemoglobin Aromatic OH 11/26/23 Human Blood - Vinyl Inquiry NIR 11/07/15 2941 1 7.4 est Vinyl Vinyl Vinyl Vinyl Vinylidene Vinylidene Acrylate 11/26/2023 Human Blood - Vinyl Inquiry MID IR 11/07/15 2941 1 7.4 est Ethyl Vinyl Ether (SDBS) Ether Vinyl 12/08/2023 TSP 070 DNA UV 10/08/23 61 1 Nucleic Acids - DNA NIR 10/08/23 Nucleic Acids - DNA
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Count 36 20 16 RMS 7.1 12 1 5 3 2 9 7 2 1 1 4 2 14 5 5 5 8 9 8 3 10 9 1 23 8 2 1 0 0 0 1 1 Range 23.0 cFREQUENCY 0.5524 90 32 78 65 53 87 84 53 32 32 73 53 92 78 78 78 86 87 86 65 89 87 32 95 86 53 32 0 0 0 32 32

Combined Clot All Groups Frequency Pr% Groups Method Methods

Vinyl 95 X NIR UV Methyl 90 X NIR NIR Protein 90 X NIR LC Aromatic Amine 90 X NIR MID IR Alcohol 85 X NIR VIS Alkyl Alcohol 85 NIR Microscopy Amide 85 X NIR Titration Polyamide 85 X NIR Qualitative Aromatic CH 85 X NIR Meter Nucleic Acids - DNA 85 Vinylidene 85 NIR Methylene 80 X NIR Hemoglobin 80 Tyrosine 80 Amine 80 X NIR Alknes N=15/30 Polyenes 10/12 36 samples Aromatic OH

2023.12.12 Research Paper - Regroup

Regrouping.

1. Research paper needs to pull itself in toward close. Look at bring Prob level to 75% vs 80%. How does it affect inclusion/overlap list?

It does not change anything. Leave at 80% Probability.

2. Does vinyl dissolve in acid/base? Compare to clot behavior.

Vinyl does not dissolve quickly or easily in either strong H2SO4 or NaOH-KOH. Same as clot behavior. This is therefore, an important observation.

3. Outline the chemistry/functional groups towards synthetic rubber.

From previous notes:

"Rubber is alkene based. OK, this gets very interesting. Synthetic rubber is a copolymer between styrene and butadiene. Styrene is a VINYL group attached to an aromatic ring. Butadiene is an alkene.

Guess what, all the signs are starting to point the same direction. Aromatic, vinyl, alkenes are the components of synthetic rubber, which is most characteristic of the mature clot gross properties. Aromatic, vinyl, alkenes appear to be dominant structures of our NIR plots. Do not keep trying to force a single functional group. The combination of these three functional groups along with synthetic blood go a long way toward explaining the culture findings in general.

2. Two polymeric forms, one for structual density and the other for elasticity. Both highly insoluble, even in many strong reagents/acids/caustics.

This is a major achievement. It would certainly be of benefit to understand how the two polymers may join together.

Now the rubber property would be imparted by the vinyl group, which we now understand is a part of the copolymeric process of synthetic rubber. We may well have, and are expected to have a copolymer arrangement here.

The picture looks increasingly clear. It would seem that we have a genetically engineered co-polymerization process taking place here. The process involves synthetic blood, an aromatic protein polymer, and a synthetic rubber equivalent polymer. Their origin is the Cross Domain Bacteria (CDB). The result is increased coagulation or a clotting of the blood. The end result is definitely a lethal threat.

Mid IR exploration of vinyl group with historical "CDB Protein Complex" IR data (cerca 2015) looks to be most important. Initial peak match search brings up following candidates as:

1. Butyl vinyl ether C6H12O

2. Cyclohexyl vinyl ether C6H14O

These candidates have essentially exact matches with IR collected Sep 2015 of "CDB Protein Complex". Out of about 10 candidates from entire SDBS database. These are lowest molecular weight. I am guessing we have something here.

Look at physical properties, synthetic rubber, etc.

now tell me three common ways that synthetic rubber is made.

Sure! Here are three common ways that synthetic rubber is made:

1. Polymerization: This method involves the polymerization of monomers to form long chains of rubber molecules. The most common monomers used in synthetic rubber production are butadiene and styrene. Polymerization can be achieved through various techniques such as emulsion polymerization, solution polymerization, or bulk polymerization.

2. Copolymerization: In this approach, two or more different monomers are combined to create copolymers. By blending different monomers, the resulting synthetic rubber can have enhanced properties compared to rubber produced from a single monomer. For example, the copolymerization of butadiene and styrene produces a type of synthetic rubber called Styrene-Butadiene Rubber (SBR), which is widely used in tire manufacturing.

3. Vulcanization: Vulcanization is a process used to improve the properties of synthetic rubber. It involves the addition of various chemicals, such as sulfur, accelerators, and activators, to the rubber compound. The rubber is then heated, which causes the chemical additives to react and cross-link the rubber molecules. This cross-linking process enhances the rubber's strength, elasticity, and resistance to heat, aging, and abrasion.

These are just a few common methods used in the production of synthetic rubber. The specific technique chosen depends on the desired properties and applications of the final rubber product.

is butadience a vinyl product?

No, butadiene is not a vinyl product. Butadiene is a colorless gas that is used as a raw material in the production of synthetic rubbers, such as styrene-butadiene rubber (SBR) and polybutadiene rubber (PBR). Vinyl products, on the other hand, are made from vinyl chloride monomer and include materials like polyvinyl chloride (PVC).

what are examples of polymer vinyls?

Some examples of polymer vinyls include:

1. Polyvinyl chloride (PVC): PVC is a widely used polymer vinyl that is known for its versatility and durability. It is commonly used in construction materials, pipes, electrical cables, and vinyl flooring.

2. Polyvinyl acetate (PVAc): PVAc is a polymer vinyl often used in adhesives, paints, and coatings. It is also the main component in white glue or wood glue.

3. Polyvinyl alcohol (PVA): PVA is a water-soluble polymer vinyl that is used in various applications such as adhesives, coatings, and textiles. It is also used in the production of PVA films and fibers.

4. Polyvinylidene chloride (PVDC): PVDC is a polymer vinyl that is known for its excellent barrier properties against gases and moisture. It is commonly used in food packaging films, shrink wrap, and protective coatings.

5. Polyvinylpyrrolidone (PVP): PVP is a polymer vinyl that is used in pharmaceuticals, cosmetics, and personal care products. It is often used as a binder, film-former, or stabilizer in various formulations.

These are just a few examples of polymer vinyls, and there are many more variations and derivatives available in the market.

what are the chemical and physical property differences between dienes and trienes?

Dienes and trienes are both types of hydrocarbons that contain double bonds. However, there are some differences in their chemical and physical properties:

1. Chemical Properties:

- Dienes have two carbon-carbon double bonds, while trienes have three carbon-carbon double bonds.

- Dienes can undergo addition reactions with electrophiles, such as hydrogenation or halogenation, at each double bond independently. Trienes can also undergo similar addition reactions, but with the presence of three double bonds, the reactions can be more complex.

 Dienes can undergo polymerization reactions to form polymers, such as synthetic rubbers. Trienes can also undergo polymerization, but the presence of more double bonds can lead to more complex polymer structures.
Dienes can undergo Diels-Alder reactions, which involve cycloaddition reactions with dienophiles. Trienes can also participate in Diels-Alder reactions, but with the presence of more double bonds, the reactions can have more diverse outcomes.

2. Physical Properties:

- Generally, as the number of double bonds increases from dienes to trienes, the boiling points and melting points tend to increase. This is due to the increased intermolecular forces (Van der Waals forces) between the molecules.

- The presence of double bonds in both dienes and trienes can lead to increased reactivity, making them more prone to oxidation and other chemical reactions compared to saturated hydrocarbons.

- Both dienes and trienes are typically liquids or low-melting solids at room temperature, depending on the specific structure and molecular weight.

It's important to note that these properties can vary depending on the specific diene or triene compound, as different substituents and structural arrangements can influence their behavior.

so we know that dienes and trienes are alkanes.

what it is called when you have two vinyl functional groups combined

When two vinyl functional groups are combined, it is called a vinyl group dimerization or vinyl dimerization.

This makes up the majority of the clot structure.

1,3-Butadiene (a synthetic rubber) from NIST data base UV spectral search shows a peak at 293. This is a vinyl compound and then a match for our spectrum of TSP 100.6 at this point (not the rest of the spectrum nor our 232 peak found.

NIST and Chemspider both have some UV searching capability if you can get a part of the name. VERY VALUABLE!

The styrene UV spectrum explains high absorbance towards the 232 peak. Synthetic rubber is a copolymer between styrene and butadiene.

Chemicals that dissolve rubber

To understand what dissolves rubber, we need to examine the chemical composition of rubber itself. Rubber is primarily composed of polymers, which are long chains of repeating units called monomers. While rubber is resistant to many chemicals, there are specific substances that can break down these polymer chains and dissolve rubber. Some of these chemicals include:

Organic solvents: Certain organic solvents, such as acetone and benzene, have the ability to dissolve rubber. These solvents can penetrate the polymer chains and weaken the structure of rubber, causing it to break down.

Strong acids and bases: Strong acids, like sulfuric acid, and strong bases, like sodium hydroxide, can also dissolve rubber. These chemicals react with the rubber s polymer chains, causing them to break apart and dissolve.

Certain oils and fuels: Some oils and fuels, particularly those with high aromatic content, can dissolve rubber. These substances can penetrate the rubber and cause it to swell, leading to its eventual dissolution.

Factors affecting rubber dissolution

Several factors can influence the dissolution of rubber. These include:

Temperature: Higher temperatures can increase the solubility of rubber in certain chemicals. Therefore, exposure to elevated temperatures can accelerate the dissolution process.

Duration of exposure: The longer rubber is exposed to dissolving agents, the greater the chances of it being dissolved. Prolonged exposure allows the chemicals to penetrate deeper into the rubber, leading to its breakdown.

Rubber composition: Different types of rubber have varying resistance to specific chemicals. Factors such as the presence of additives or the type of polymer used in the rubber s composition can affect its susceptibility to dissolution.

Tell me about synthetic rubber.

Synthetic rubber is a type of rubber that is artificially produced rather than being derived from natural sources such as latex from rubber trees. It is created through a chemical process known as polymerization, where various monomers are combined to form long chains called polymers.

There are several types of synthetic rubber, each with its own unique properties and uses. Some common types include:

1. Styrene-butadiene rubber (SBR): SBR is one of the most widely used synthetic rubbers. It is highly resistant

to abrasion and has good flexibility, making it suitable for applications such as tires, conveyor belts, and footwear.

2. Polybutadiene rubber (BR): BR is known for its exceptional elasticity and low-temperature flexibility. It is commonly used in the production of tires, as well as in various industrial applications where high resilience is required.

3. Neoprene: Neoprene is a versatile synthetic rubber that is resistant to oil, heat, and weathering. It is commonly used in the manufacturing of wetsuits, gaskets, hoses, and electrical insulation.

4. Nitrile rubber (NBR): NBR is known for its excellent resistance to oil, fuel, and chemicals. It is widely used in the automotive and aerospace industries for applications such as seals, gaskets, and hoses.

5. Ethylene-propylene-diene rubber (EPDM): EPDM is a synthetic rubber with excellent weather resistance and electrical insulation properties. It is commonly used in roofing materials, automotive parts, and electrical insulation.

Synthetic rubber offers several advantages over natural rubber, including better resistance to wear, aging, and chemicals. It can be formulated to have specific properties, making it suitable for a wide range of applications. Additionally, synthetic rubber production helps reduce dependence on natural rubber sources and provides more consistent supply and quality control.

However, synthetic rubber production also has environmental implications. The process of manufacturing synthetic rubber involves the use of petrochemicals, which are derived from non-renewable fossil fuels. Efforts are being made to develop more sustainable and eco-friendly alternatives to synthetic rubber production.

Is vinyl used in synthetic rubber

No, vinyl is not used in the production of synthetic rubber. Synthetic rubber is typically made from petroleum-based materials such as styrene and butadiene. Vinyl, on the other hand, is a type of plastic made from vinyl chloride monomers.

what is butadiene

Butadiene is a colorless gas with a mild aromatic odor. It is a highly flammable compound that is commonly used in the production of synthetic rubber. Butadiene is also used as a chemical intermediate in the manufacturing of various products, including plastics, resins, and coatings. It can be produced from petroleum or by the thermal cracking of natural gas liquids.

Does butadiene have any vinyl groups within it

No, butadiene does not have any vinyl groups within it. Butadiene is a hydrocarbon compound with the chemical formula C4H6, consisting of four carbon atoms and six hydrogen atoms. It is a conjugated diene, meaning it has two double bonds between carbon atoms.

What is a vinyl functional group

A vinyl functional group is a chemical group consisting of two carbon atoms bonded together with a double bond. It is represented by the symbol "-CH=CH2". The vinyl group is commonly found in organic compounds and is known for its reactivity in various chemical reactions. It is often used in the synthesis of polymers, such as polyvinyl chloride (PVC), and in the production of pharmaceuticals and other organic compounds.

What is styrene

Styrene is a colorless liquid hydrocarbon that is derived from petroleum. It is primarily used in the production of polystyrene, a versatile plastic material commonly found in packaging, insulation, and disposable products. Styrene is also used in the manufacturing of other plastics, rubber, resins, and synthetic fibers. It is important to note that prolonged exposure to styrene can have health effects, and it is classified as a possible human carcinogen by the International Agency for Research on Cancer.

Is there a relationship between a diene and a vinyl group

Yes, there is a relationship between a diene and a vinyl group. A diene is a molecule that contains two double bonds, while a vinyl group is a functional group consisting of a carbon-carbon double bond. In a diene, one of the double bonds can be considered as a vinyl group.

THIS IS CRUCIAL INFO. YOU HAD TO KNOW WHAT TO ASK TO GET THS INFO. OTHERWISE VINYL RELATIONSHIP WAS NOT DISCLOSED.

How do you make synthetic rubber from butadiene and styrene

To make synthetic rubber from butadiene and styrene, you can follow these steps:

1. Obtain butadiene and styrene: Butadiene and styrene are both commercially available chemicals. You can purchase them from chemical suppliers.

2. Polymerization: Mix the butadiene and styrene in the desired ratio. Typically, a ratio of 75-80% butadiene and 20-25% styrene is used for making a synthetic rubber called styrene-butadiene rubber (SBR).

3. Initiation: Add a suitable initiator, such as a peroxide or an organic metal compound, to the mixture. This initiates the polymerization reaction.

4. Polymerization reaction: Heat the mixture to a specific temperature, typically around 50-70 degrees Celsius, to initiate the polymerization reaction. The reaction can be carried out in a reactor vessel under controlled conditions.

5. Stirring and heating: Stir the mixture continuously to ensure uniform mixing and heat it for a specific duration to complete the polymerization process. The reaction time can vary depending on the desired properties of the synthetic rubber.

6. Cooling and solidification: Once the polymerization is complete, cool the mixture to room temperature to solidify the synthetic rubber.

7. Post-treatment: The solidified synthetic rubber may require further processing steps like washing, drying, and cutting into desired shapes or sizes.

8. Quality control: Test the synthetic rubber for its physical and chemical properties to ensure it meets the required specifications. This may include testing for tensile strength, elongation, hardness, and other relevant parameters.

Note: The exact process conditions and additives used may vary depending on the specific requirements and intended applications of the synthetic rubber being produced. It is always recommended to follow established industrial protocols and safety guidelines when working with chemicals and conducting polymerization reactions.

NOTE: LEWIS DIAGRAMS DO NOT TRANSCRIBE CORRECTLY. BUT THE IDEA IS HELPFUL.

What is the Lewis diagram for styrene

To draw the Lewis diagram for styrene (C8H8), we first need to determine the number of valence electrons for each atom. Carbon (C) has 4 valence electrons, and hydrogen (H) has 1 valence electron.

Styrene consists of a benzene ring (C6H6) with an attached ethenyl group (C2H3). The benzene ring has alternating double and single bonds, while the ethenyl group has a double bond.

Here's the Lewis diagram for styrene:

In this diagram, the double bonds are represented by two lines, while the single bonds are represented by a

single line. The carbon atoms in the benzene ring are connected by single bonds, while the carbon atoms in the ethenyl group are connected by a double bond. The hydrogen atoms are attached to the carbon atoms.

Note: The Lewis diagram is a simplified representation of the molecule's structure, showing only the valence electrons and the connections between atoms. It does not show the three-dimensional shape of the molecule.

What is the Lewis diagram for butadiene

The Lewis diagram for butadiene (C4H6) can be represented as follows:

нннн \/\/ C=C=C=C НH

What is the difference between a dieneand a butadiene

A diene is a hydrocarbon compound that contains two double bonds in its structure. It is a general term used to describe any molecule with two double bonds, regardless of the number of carbon atoms present. On the other hand, butadiene is a specific type of diene that contains four carbon atoms and two double bonds. In other words, all butadienes are dienes, but not all dienes are butadienes.

Describe the chemical reaction between butadiene and styrene to form synthetic rubber

The chemical reaction between butadiene and styrene to form synthetic rubber is known as copolymerization. It involves the combination of monomer molecules of butadiene and styrene to create a polymer chain.

The reaction is initiated by a catalyst, typically an organometallic compound such as a lithium or nickel catalyst. The catalyst helps in breaking the double bonds present in the monomer molecules and facilitates the formation of new bonds.

During the reaction, the double bond in butadiene undergoes a process called addition polymerization. The double bond is opened up, and the carbon atoms on either side of the double bond attach to the monomer units of styrene. This results in the formation of a long polymer chain with repeating units of butadiene and styrene.

The copolymerization of butadiene and styrene allows for the incorporation of different properties from both monomers. Butadiene contributes flexibility and resilience to the synthetic rubber, while styrene adds strength and rigidity. This combination of properties makes the resulting synthetic rubber suitable for various applications, such as tire manufacturing, automotive parts, and industrial products.

Overall, the chemical reaction between butadiene and styrene to form synthetic rubber involves the addition of monomer units through copolymerization, resulting in the creation of a polymer chain with desirable properties for rubber production.

Does styrene have a vinyl group in it

Yes, styrene does have a vinyl group in its chemical structure. The vinyl group is a functional group consisting of a carbon-carbon double bond (C=C). In the case of styrene, the vinyl group is attached to a benzene ring.

Does the chemical compound butadiene Contain a vinyl group

Yes, the chemical compound butadiene contains a vinyl group. The vinyl group is a functional group consisting of a carbon-carbon double bond and a single bond to a hydrogen atom. In butadiene, there are two vinyl

groups present, as it contains two carbon-carbon double bonds.

Is Ethyl vinyl ether related to synthetic rubber

Yes, ethyl vinyl ether is related to synthetic rubber. It is a monomer that is used in the production of certain types of synthetic rubber, such as ethylene-vinyl acetate (EVA) copolymers. These copolymers are commonly used in the production of various rubber products, including shoe soles, gaskets, and seals.

Describe ethylene-vinyl acetate (EVA) copolymers.

Ethylene-vinyl acetate (EVA) copolymers are a type of thermoplastic elastomer that is made from the copolymerization of ethylene and vinyl acetate monomers. This copolymerization process results in a material that combines the properties of both ethylene and vinyl acetate.

EVA copolymers have a wide range of applications due to their versatility and desirable characteristics. They are known for their excellent flexibility, low-temperature toughness, and good impact resistance. EVA copolymers also have a low melting point, which makes them easy to process and mold into various shapes.

One of the key features of EVA copolymers is their ability to be easily modified. The vinyl acetate content in the copolymer can be adjusted to achieve different levels of flexibility and hardness. This allows for a wide range of applications, from soft and flexible products such as foam padding and shoe soles, to harder and more rigid products like automotive parts and electrical cable insulation.

EVA copolymers also have good chemical resistance, which makes them suitable for use in applications where they may come into contact with oils, solvents, or other chemicals. They have good adhesion properties, allowing them to bond well with other materials, and they can be easily processed using various techniques such as injection molding, extrusion, and blow molding.

Overall, EVA copolymers are a versatile and widely used material in industries such as footwear, packaging, automotive, electronics, and sports equipment, among others. Their combination of flexibility, toughness, and processability makes them an ideal choice for many different applications.

Give me some examples of some commercial products that contain ethyl vinyl acetate

Sure! Here are some examples of commercial products that may contain ethyl vinyl acetate (EVA):

1. Athletic shoes: EVA is often used in the midsole of athletic shoes to provide cushioning and support.

2. Flip-flops: Many flip-flops have EVA foam footbeds for comfort and shock absorption.

3. Yoga mats: EVA foam is commonly used in yoga mats due to its softness and ability to provide a non-slip surface.

4. Packaging materials: EVA can be found in various packaging materials, such as foam inserts or protective padding.

5. Children's toys: Some toys, like building blocks or foam puzzles, may use EVA foam for safety and durability.

6. Orthotic insoles: EVA is frequently used in orthotic insoles to provide cushioning and support for the feet.

7. Safety helmets: EVA foam liners are often used in safety helmets to absorb impact and provide comfort.

8. Luggage and bags: Some luggage and bags may have EVA foam padding or inserts to protect delicate items.

9. Exercise equipment: EVA foam may be used as padding on exercise equipment, such as weight benches or yoga blocks.

10. Insulation materials: EVA foam can be used as insulation in various applications, such as HVAC systems or construction projects.

Please note that the specific use of EVA in these products may vary, and it is always recommended to check the product specifications or labels for accurate information.

How do you make ethyl vinyl acetate from ethyl vinyl ether

To make ethyl vinyl acetate from ethyl vinyl ether, you will need to perform an esterification reaction. Here are the steps you can follow:

1. Gather the necessary materials and equipment: ethyl vinyl ether, acetic acid, a catalyst (such as sulfuric acid), a round-bottom flask, a condenser, a heating source (such as a hot plate), and a collection vessel.

2. Set up a reflux system: Attach the round-bottom flask to the condenser and connect the condenser to the collection vessel. This will allow for the reaction mixture to be heated and the volatile components to condense and collect.

3. Add the reactants: In the round-bottom flask, combine ethyl vinyl ether and acetic acid in the desired stoichiometric ratio. The molar ratio should be 1:1 for the reaction to proceed efficiently.

4. Add a catalyst: Add a small amount of a catalyst, such as sulfuric acid, to the reaction mixture. The catalyst helps to facilitate the esterification reaction.

5. Heat the reaction mixture: Place the round-bottom flask on a heating source, such as a hot plate, and heat the mixture under reflux conditions. Refluxing means that the mixture is heated to boiling and the vapors are condensed and returned to the reaction flask.

6. Allow the reaction to proceed: Let the reaction mixture reflux for a certain period of time. The reaction time can vary but typically ranges from a few hours to overnight.

7. Monitor the progress of the reaction: You can periodically sample the reaction mixture and analyze it using techniques such as gas chromatography or infrared spectroscopy to determine the conversion of ethyl vinyl ether to ethyl vinyl acetate.

8. Collect the product: After the desired reaction time, cool the reaction mixture and collect the product, ethyl vinyl acetate, which will have condensed in the collection vessel.

9. Purify the product: The collected ethyl vinyl acetate may contain impurities. To purify the product, you can perform techniques such as distillation or extraction to separate and remove any impurities.

What are the chemical and physical properties of ethyl vinyl ether

Ethyl vinyl ether (C4H8O) is a colorless, flammable liquid with a sweet, ether-like odor. Here are some of its chemical and physical properties:

Chemical properties:

- 1. Molecular weight: 72.11 g/mol
- 2. Chemical formula: C4H80
- 3. Boiling point: 35-36 °C (95-97 °F)
- 4. Melting point: -115 °C (-175 °F)
- 5. Density: 0.736 g/cm3
- 6. Solubility: Ethyl vinyl ether is soluble in many organic solvents, such as ethanol, ether, and chloroform.

7. Reactivity: Ethyl vinyl ether can undergo various chemical reactions, including polymerization, oxidation, and hydrolysis.

Physical properties:

1. Appearance: Ethyl vinyl ether is a clear, colorless liquid.

2. Odor: It has a sweet, ether-like odor.

3. Flammability: Ethyl vinyl ether is highly flammable and can form explosive mixtures with air.

4. Vapor pressure: The vapor pressure of ethyl vinyl ether is relatively high, which means it can easily evaporate at room temperature.

5. Miscibility: It is miscible with many organic solvents but immiscible in water.

6. Stability: Ethyl vinyl ether is relatively stable under normal conditions but can react with oxygen in the air to form peroxides, which can be explosive.

What are the chemical and physical properties of ethyl vinyl acetate

Ethyl vinyl acetate (EVA) is a copolymer of ethylene and vinyl acetate. It has a number of chemical and physical properties, including:

Chemical properties:

- 1. Chemical formula: (C2H4)x(C4H6O2)y
- 2. Molecular weight: Variable, depending on the ratio of ethylene to vinyl acetate units.
- 3. Melting point: Typically ranges from 40 to 90 °C, depending on the vinyl acetate content.
- 4. Solubility: EVA is soluble in many organic solvents, such as acetone, toluene, and chloroform.

Physical properties:

- 1. Appearance: EVA is a soft, flexible, and transparent material, often available in the form of pellets or sheets.
- 2. Density: The density of EVA ranges from 0.92 to 0.96 g/cm³, depending on the vinyl acetate content.
- 3. Hardness: EVA has a relatively low hardness, typically ranging from 20 to 90 Shore A.
- 4. Flexibility: EVA is highly flexible and can be easily molded or shaped at room temperature.

5. Thermal stability: EVA has good thermal stability, with a maximum service temperature of around 70 to 80 °C.

6. Electrical properties: EVA is an electrical insulator and has good dielectric properties.

7. UV resistance: EVA has good resistance to UV radiation, making it suitable for outdoor applications.

These properties make EVA a versatile material used in various industries, including packaging, footwear, automotive, and construction.

What are some of the differences and similarities between ethel vinyl ether And Ethel vinyl acetate

Ethyl vinyl ether and ethyl vinyl acetate are both chemical compounds that belong to the class of vinyl ethers. However, they have some differences and similarities:

1. Structure: Ethyl vinyl ether (C4H8O) has a simple structure consisting of an ethyl group attached to a vinyl group. Ethyl vinyl acetate (C6H10O2) has a more complex structure, with an ethyl group attached to a vinyl group and an acetate group.

2. Physical Properties: Ethyl vinyl ether is a colorless liquid with a boiling point of 35-37°C. Ethyl vinyl acetate is also a colorless liquid but has a higher boiling point of 72-73°C.

3. Reactivity: Both compounds are highly reactive due to the presence of the vinyl group. They can undergo various chemical reactions such as polymerization, esterification, and oxidation.

4. Uses: Ethyl vinyl ether is primarily used as a monomer in the production of various polymers, resins, and coatings. Ethyl vinyl acetate is commonly used as a binder in adhesives, sealants, and coatings, as well as in the production of vinyl acetate-ethylene copolymers.

5. Toxicity: Ethyl vinyl ether is considered to be highly flammable and may cause irritation to the respiratory system and eyes. Ethyl vinyl acetate is also flammable but is generally considered to have low toxicity.

In summary, while both ethyl vinyl ether and ethyl vinyl acetate are vinyl ethers, they differ in terms of their chemical structure, physical properties, reactivity, uses, and toxicity.

^{4.} Stark visual evidence of clotting issue.

5. Bacteriophage prospect

2023.12.13 Alkenes - Polymerization

Are alkenes subject to Polymerization

Yes, alkenes are subject to polymerization. Polymerization is a process in which monomers (smaller molecules) join together to form polymers (larger molecules). Alkenes, which are unsaturated hydrocarbons with at least one carbon-carbon double bond, can undergo addition polymerization. In this process, the double bond is broken and the monomers add together to form a long chain polymer. Examples of polymers formed from alkenes include polyethylene and polypropylene.

What is ethylene

H H || C double bond C || H H

Ethylene is a colorless, flammable gas that is commonly used in various industries. It is a hydrocarbon compound with the chemical formula C2H4. Ethylene is produced naturally by plants as a hormone to regulate growth and development. It is also produced synthetically for commercial purposes, such as in the production of plastics, solvents, and various chemical intermediates. Ethylene is highly reactive and is used in many industrial processes, including the ripening of fruits, polymerization reactions, and as a fuel in welding and cutting operations.

Ethyl vinyl ether, also known as ethoxyethene or EVE, is an organic compound with the chemical formula C4H8O. It is a colorless, flammable liquid with a sweet, ether-like odor.

Ethyl vinyl ether is primarily used as a monomer in the production of various polymers and copolymers. It can undergo polymerization reactions to form polyvinyl ethers, which find applications in adhesives, coatings, and modified resins.

In addition to its role as a monomer, ethyl vinyl ether has some applications as a solvent. It can dissolve a wide range of organic compounds, making it useful in various industries such as pharmaceuticals, paints, and coatings.

It is important to note that ethyl vinyl ether is highly flammable and should be handled with care. It can form explosive mixtures with air and should be stored in well-ventilated areas away from ignition sources. Furthermore, ethyl vinyl ether can be harmful if inhaled, swallowed, or absorbed through the skin. It may cause irritation to the respiratory system, eyes, and skin. Therefore, proper safety precautions, such as wearing protective equipment, should be followed when handling this compound.

As with any chemical, it is always recommended to consult the safety data sheet (SDS) and follow the guidelines provided by the manufacturer for safe handling, storage, and disposal of ethyl vinyl ether.

Propene, also known as propylene, is an organic compound that belongs to the alkene group of hydrocarbons. It is a colorless gas with a slightly sweet and gasoline-like odor. Propene is highly flammable and can form explosive mixtures with air.

One of the most important uses of propene is as a monomer in the production of polypropylene, which is a versatile plastic used in a wide range of applications, including packaging, textiles, automotive parts, and more. Polypropylene is known for its strength, flexibility, and resistance to heat, chemicals, and moisture. Propene is also used as a fuel gas for heating and cooking, as a refrigerant, and as a raw material in the production of various chemicals. It is used to manufacture propylene oxide, which is further used in the production of polyurethane foams, coatings, and adhesives.

2023.12.14 Mitigation - Methylation

Ok, this was my developing hunch, and it is quite correct. This is highly important:

Can methylation be used to terminate polymerization?

Yes, methylation can be used to terminate polymerization reactions. Methylation refers to the addition of a methyl group (-CH3) to a molecule. In the context of polymerization, methylation can be used to terminate the growth of a polymer chain by adding a methyl group to the reactive end of the chain, preventing further monomer addition.

For example, in the synthesis of certain polymers, such as polyethylene or polypropylene, a small amount of a methylating agent can be added to the reaction mixture. This methylating agent reacts with the active chain end, adding a methyl group and effectively terminating the polymerization process.

Methylation can also be used as a strategy to control the molecular weight or chain length of a polymer. By adding a methylating agent at specific intervals during the polymerization reaction, the growth of the polymer chains can be controlled, resulting in polymers with desired properties.

Overall, methylation can be a useful tool in polymerization reactions to control chain length, terminate polymer growth, and manipulate the properties of the resulting polymer.

This is as strong a method as anything developed thus far. Add to the list.

This therfore provides an additional potential mitigation strategy. Increase intake of negatively charged proteins.

Mitigation strategies (laboratory and evidence based):

1. Protein disruption : enzymes

2. Methylation disruption or reduction (vit B) TERMINATION OF POLYMERIZATION WITH METHYLATION. Fatty acids and fats chemistry double bonds (fish oils) DNA regulation and repair Methylation is of major importance

3. Negatively charge protein intake?

Newer trial includes:

1. Glutamic acid supplement <= ~.5 gm (powder form now added to pH control solution trial)

2. Aspartic acid supplement <= ~.5 gm

4. pH regulation (risks as well, e.g, sodium increase, polymerization, borax, misinformation, blood pH vs urine ph)

pH regulation is now formed as

1. low concentration sodium bicarbonate (e.g. <= gm)

- 2. citric acid (3 to 1 ratio on baking soda)
- 3. pinch pink salt
- 4. single alka seltzer
- 5. full cup water

6. monitor pH during day, urine will rise. eg, 5.5 to 6.2 - 6.5

Keep the sodium intake as low as possible. Single asprin.

Calcium citrate, magnesium citrate supplements are optional additions. Yes, likely best to rotate between all 3 citrate forms.

Solution in final form is fairly neutral taste (glutamic acid note above) pH \sim 6.2 Too much sodium can complicate matters. Edema as well.

5. Ultrasound - very gradual polymer degradation, difficulties.

- 6. Antoxidants and the usual two dozen suspects (eg Vit C, NAC, alpha lipoic
- 7. Balm, e.g., methyl salicylate, menthol, tea tree, chappara(creosote bush)

We know now that the balm either causes: 1. pain reduction

2. relocation

or

3. forcing to polymer to the surface skin level. Once at the skin level severe itching and sores will likely break out.

Ultrasound will, VERY SLOWLY, with measurable pain and incredible patience, degrade the polymers and lead to eventual removal from the skin. Years may easily be involved in eht process.

8. Another major mitigation strategy has been identified. 265 nm UV. Broad distribution in the literature, even against SARS.

Dec 28 Notes. LC run with acetone isolates 265 nm absorbance peak. Literature replete wi/effectiveness of Deep UV against bacterial DNA.

9. We have another mitigation route to develop. Acetone + TSP = a plastic [MAYBE - MAYBE NOT?!]. The body produces acetone during fat metabolsim. This is difficult, by interference in this process could be helpful.

What we learn here (on Dec 28 2023) is that acetone INDUCES polymerization of the TSP. Acetone is produced by the human body.

From work of Jan 03 we see that we likely have a calcium - polyamide and/or vinyl complex being formed. This raises calcium interference issues as well.

[This conclusion further confirmed with the work of Jan 05 2023 notes:

"We therefore have a high confidence of our compound as being a

***CALCIUM CARBONATE POLYAMIDE OR POSSIBLE CALCIUM CARBONATE VINYL COMPOUND.

and here we go further as the constituents are elevated in pH of the body relative to that of the culture:

As the pH increases, is calcium carbonate more or less soluble in water?

As the pH increases, calcium carbonate becomes less soluble in water. This is because calcium carbonate is an alkaline compound and its solubility decreases as the solution becomes more alkaline.

This raises serious issues on detriment to health with iron and calcium disruption and theft."]

10. Advocation of borax is dangerous, it also produces an brittle insoluble polymer in conjunctioin with the TSP.

Notes of Jan 04 2024: - Directly related to mitigation progress and plans:

"Blood looks excellent. Free standing, circular geometry, minimal deformation, isolated cdb in surrounding plasma, no CDB invasion of cells of significance observed. Sustained favorable conditions now. Quite the turn around. All based upon laboratory evidence and discoveries. Slide prepares evenly and easily. Notice no pressure on slide is required to produce smear, a very gentle process. Even flow of blood upon slide now.

3 main phases are required:

- 1. Get the blood back to a normal state (stop the protein formation at the beginning of the chain).
- 2. Stop or interfere with the polymerization process.
- 3. Remove existing polymers."

So we see that there are important differences between fatty acids and fats (from ai chat below). Therefore, methylation disturbance are important to both functions of methyl influence: fats and polymerizaton. DNA repair is another. So methylation is extremely important and any interference to that process causes major damage and interruption. A very big deal here...

PREVIOUS NOTES:

I am recognizing the similarity between nylon and solubility of the synth-polyamide layer. They are both soluble in strong sulfuric acid and maybe not much else. Acetone maybe some? That suggests that they share the same characteristic of both being polyamides.

Strong research topic here....

Note this comment:

(Note: "Red cell membranes have a negative charge (zeta potential) that causes red cells to repel each other. In the presence of increased positively charged plasma proteins such as fibrinogen or immunoglobulins, the negative charge on the red cell surface is diminished, allowing red cells to stick together." Ref. hematology.org)

It would not be surprising if this ends up being quite important. Now start thinking about this. You have an isoelectric point of 5.33. This is when the charges are zero, i.e., electrically neutral. What charge would the protein have if it were at pH 7.4?

"The isoelectric point (pl) of a protein is defined as the pH at which the net charge of a protein molecule is zero. Accordingly, proteins are positively charged at a pH below their pI and negatively charged at a pH above their pI. The protein pI varies greatly from extremely acidic to highly alkaline values ranging from about 4.0 to 12.0. Hence, pI values have long been used to distinguish between proteins in methods for protein isolation, separation, purification, crystallization, etc. Amino acid composition of a protein sequence primarily defines its pI, based on the combination of dissociation constant (pKa) values of the constituent amino acids. Out of twenty common amino acids, two amino acids, aspartic acid, and glutamic acid, are negatively charged and three amino acids, lysine, arginine, and histidine, are positively charged at the neutral pH, as defined by their pKa values."

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8667598/

This looks like it has our answer. The pH of the culture is on the order of 3.8. The dominant pH of the human blood proteome is 5.3. The pH of the culture is lower than the dominant pI of the blood proteome. This means that the blood proteins are becoming more positively charged with the introduction of the CDB.

OK I have said it in the paper in progress:

"((Note: Red cell membranes have a negative charge (zeta potential) that causes red cells to repel each other. In the presence of increased positively charged plasma proteins such as fibrinogen or immunoglobulins, the negative charge on the red cell surface is diminished, allowing red cells to stick together. Ref. hematology.org)

The isoelectric point (pl) of a protein is defined as the pH at which the net charge of a protein molecule is zero. Accordingly, proteins are positively charged at a pH below their pI and negatively charged at a pH above their pI. The protein pI varies greatly from extremely acidic to highly alkaline values ranging from about 4.0 to 12.0. Hence, pI values have long been used to distinguish between proteins in methods for protein isolation, separation, purification, crystallization, etc. Amino acid composition of a protein sequence primarily defines its pI, based on the combination of dissociation constant (pKa) values of the constituent amino acids. Out of twenty common amino acids, two amino acids, aspartic acid, and glutamic acid, are negatively charged and three amino acids, lysine, arginine, and histidine, are positively charged at the neutral pH, as defined by their pKa values.

Ref. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8667598/

This looks like it has our answer. The pH of the culture is on the order of 3.8 (strongly acidic). The dominant pH of the human blood proteome is 5.3. The pI of the CDB culture is determined at 5.33. The pH of the CDB culture is lower than the dominant pI of the blood proteome as well as lower than the CDB culture pI. This means that the blood proteins are being exposed to positively charged proteins with the introduction of the CDB.0. This would explain increased coagulation. CEC)"

This therfore provides an additional potential mitigation strategy. Increase intake of negatively charged proteins.

Mitigation prospects:

- 1. Protein disruption : enzymes
- 2. Methyl Disruption : Vit B, Fatty acids? TERMINATION POLYMERIZATION WITH METHYLATION.
- 3. pH regulation (risks as well, e.g, sodium increase, polymerization, misinformation, blood pH vs urine ph)
- 4. Ultrasound
- 5. Negatively charge protein intake?
- 6. Antoxidants and the usual two dozen suspects
- 7. Balm, e.g., methyl salicylate

The blood proteome distribution (dominant pl of 5.3) is at:

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7159840/

"Blood plasma is a major fluid compartment in human body, with a narrow normal range of pH $(7.40\pm0.05)12$. Thus the average pH of human blood is in the middle of the trough observed for the theoretical pI distribution of blood proteome (Figure 1[A]). Whether such a coincidence is by chance or by design, the density of normal proteoforms in the pI region 7.4 ± 0.1 is very low (ca. 20 times lower than in the region around pI 5.3 ± 0.1)."

The mitigation idea would be to increase intake of proteins with a pH greater than the dominant pl of blood, which is 5.3.

"Accordingly, proteins are positively charged at a pH below their pl and negatively charged at a pH above their pl."

We need the latter part of this sentence. Negatively chared amino acids are:

Aspartic acid pKa = 3.7 pl = 2.87Glutamic Acid pKa = 4.1 pi = 3.08

This means that these proteins are negatively charged anytime that they are in a pH environment that is above approximately 3.0. Blood is 7.0 so it is significantly above these pl values. So we ask what are nutritional sources of aspartic and glutamic acid?

"What is Aspartic Acid (Aspartate)?

Definition- Aspartic acid or aspartate, also known as amino succinic acid is a non-essential amino acid that is synthesized itself in the human body through different sources of foods. It is mainly responsible for synthesizing proteins and regulating hormones so also known as building blocks.

Aspartic acid is involved in synthesizing four different amino acids as it plays a vital role in Kreb s cycle; methionine, isoleucine, lysine, and threonine. It is an aspartate family and a proteinogenic amino acid. It is also a neurotransmitter.

Aspartic acid is the product formed by the hydrolysis of proteins. Aspartic acid was first identified in 1868 from legumin in plant seeds. As aspartic acid are non-essential amino they are synthesized in the body from oxalo acetic acid that is produced during the metabolism of carbohydrates.

Structure of Aspartic Acid (Aspartate)

Structure of Aspartic Acid (Aspartate)

Aspartic acid consists of two functional groups, one amino group is basic in nature and the other is the acidic carboxyl group. Therefore, amino acids molecule exists as a zwitterion. Aspartic acid is alanine with one of the ß hydrogens replaced by a carboxylic acid group. The pKa of the ß carboxyl group of aspartic acid in a

polypeptide is about 4.0. It is a dibasic amino acid having two carboxyl groups; one on alpha carbon atom and another on the side chain. Aspartic acid has an alpha-keto homolog. Aspartic acid is divided into two forms; L-aspartic acid and D-aspartic acid. L configuration is a more common and dominant form. L-aspartic acid is typically involved in the production of antibodies and is part of protein synthesis in the body which is responsible for increasing the immune system. D-aspartic acid is not involved in protein synthesis and is mainly found in the pituitary gland and testes which is used in the regulation, synthesis, and release of testosterone and luteinizing hormone.

Sources of Aspartic Acid (Aspartate) It is found in sugar cane and sugar beets molasses, asparagus, avocado, sprout seeds, and oat flakes. Animal Sources includes: Sausage meat, Luncheon meat and wild game. Other sources includes magnesium aspartate that is salt of aspartic acid and sweeter aspartame. Physical Properties of Aspartic Acid (Aspartate) Molecular weight: 133.10 White, crystalline solids Polar Acidic Hvdrophilic Orthorhombic, bisphenoidal leaflets or rods Sour in taste Chemical Properties of Aspartic Acid (Aspartate) Melting point: 270°C Solubility: 5390 mg/L at 25 °C Density: 1.6603 at 13 °C LogP: -3.89 pKa: 2.77 because of two carboxyl molecule Biosynthesis of Aspartic Acid (Aspartate) Protein extraction, chemical synthesis, and enzymatic conversion are three main methods to produce aspartic acid. A large number of amino acids are produced in the extraction method from the hydrolysis of protein. In this method, L-aspartic acid should be separated. Chemical synthesis requires high temperature and pressure in a racemic mixture producing both isomers of aspartic acid. So, enzymatic conversion is the best method for the production of aspartic acid. Bacterial fermentation is the best for the highest yield of amino acids. Pseudomonas, Bacillus, and Proteus are considered as the main producers but E. coli and Corynebacterium

Biosynthesis of Aspartic Acid (Aspartate)

glutamacium are mostly preferred by industries.

Figure: Biosynthesis of Aspartic Acid (Aspartate) from Fumaric Acid. Image Source: Kurt Rosentrater (MDPI). In the 1960s, the fermentation process is developed and patented that utilizes sugar-free medium and uses fumaric acid as a sole source of carbon. Ammonia serves as a nitrogen source that is used in catalysis. Ammonia and fumaric acid are used in combination as 1:1 or 1:2 ratio. the pH of the broth is initialized to 7 and that naturally increases from 8.4 to 9.6 in the initial stage that allows for the production of acid. Fermentation can be done with or without agitation for 2 to 10 days at 27-40 °C. L-aspartic acid will be secreted and accumulate in the culture broth. Different downstream processes are available to separate L-aspartic acid from the culture broth. But in the case of batch fermentation, ion exchange resins can be used to separate and purify L-aspartic acid followed by crystallization. L-aspartic acid can be separated by adjusting the broth to 90 ?C and a pH of 2.8 with sulfuric acid in continuous fermentation. After the pH is adjusted to 2.8, the isoelectric point will cause L-aspartic acid to precipitate out of the solution. It is then subjected to a two-hour incubation period at 15 °C to induce protein crystallization. Under these conditions, L-aspartic acids yield 95%.

Production of Aspartic Acid (Aspartate)

Figure: Production of L-Aspartic Acid. Image Source: Kurt Rosentrater (MDPI).

Functions and Uses of Aspartic Acid (Aspartate)

It is easily available multivitamins that are found in different forms as tablets, powders, and fluids. Because of its role in regulating testosterone levels, D-aspartic acid is used as increasing muscle mass. It also helps in keeping the concentrations of NADH (Nicotinamide adenine dinucleotide) high in brain cells and also increase the mind sharpness leading to further production of neurotransmitters as well as chemicals needed for normal mental functioning.

Can also be used for increasing fertility.

It is also used to produce poly aspartic acid that is used as a fertilizer synergist.

It also aids in energy production, RNA and DNA synthesis, and liver detoxification.

It also helps in removing excessive toxins from the cells like ammonia.

It is also used as building blocks molecules for active pharmaceutical agents.

It is useful in making culture medias, detergents, fungisides and germicides."

Describe the differences between saturated and unsaturated fats

Saturated fats are typically solid at room temperature and are derived from animal sources such as meat and dairy products. They are called "saturated" because their chemical structure is saturated with hydrogen atoms. These fats have a higher melting point and are more stable, making them less prone to spoilage. However, consuming excessive amounts of saturated fats has been linked to an increased risk of heart disease.

On the other hand, unsaturated fats are usually liquid at room temperature and are derived from plant sources, such as oils obtained from nuts, seeds, and vegetables. They are called "unsaturated" because they have double bonds in their chemical structure, which results in fewer hydrogen atoms. Unsaturated fats can be further classified into two types: monounsaturated fats and polyunsaturated fats.

Monounsaturated fats have one double bond and can be found in foods like olive oil, avocados, and nuts. They are considered heart-healthy and can help lower bad cholesterol levels.

Polyunsaturated fats have multiple double bonds and are found in foods like fatty fish, flaxseeds, and soybean oil. They are also beneficial for heart health and can help reduce inflammation in the body. Polyunsaturated fats are further categorized into omega-3 and omega-6 fatty acids, both of which are essential for the body but need to be balanced in the diet.

In summary, saturated fats are solid at room temperature and primarily come from animal sources, while unsaturated fats are liquid and typically derived from plant sources. Unsaturated fats can be further divided into monounsaturated and polyunsaturated fats, both of which have different health benefits. It is generally recommended to limit saturated fat intake and replace it with healthier unsaturated fats for optimal health.

https://thechemistrynotes.com/aspartic-acid/

Is there a difference between fatty acids and saturated or unsaturated fats?

Yes, there is a difference between fatty acids and saturated or unsaturated fats.

Fatty acids are the building blocks of fats. They are organic molecules that consist of a carboxylic acid group (-COOH) attached to a hydrocarbon chain. Fatty acids can vary in length and can be saturated or unsaturated.

Saturated fats are fats that consist of saturated fatty acids. Saturated fatty acids have no double bonds between the carbon atoms in the hydrocarbon chain. These fats are usually solid at room temperature and are found in animal products like meat, dairy, and some tropical oils.

Unsaturated fats are fats that consist of unsaturated fatty acids. Unsaturated fatty acids have one or more double bonds between the carbon atoms in the hydrocarbon chain. These fats are usually liquid at room temperature and are found in plant-based oils, nuts, seeds, and fatty fish.

In summary, fatty acids are the individual molecules that make up fats, while saturated and unsaturated fats are different types of fats based on the presence or absence of double bonds in the fatty acid molecules.

Do fatty acids have any double carbon bonds?

Yes, fatty acids can have double carbon bonds. Fatty acids are organic compounds consisting of a long hydrocarbon chain with a carboxyl group (-COOH) at one end. They can be classified into two main types based on the presence or absence of double bonds: saturated fatty acids and unsaturated fatty acids.

Saturated fatty acids do not have any double carbon bonds in their hydrocarbon chain. They are fully saturated with hydrogen atoms and have a straight and rigid structure.

On the other hand, unsaturated fatty acids have one or more double carbon bonds in their hydrocarbon chain. These double bonds introduce kinks or bends in the structure, making them more flexible compared to saturated fatty acids.

Unsaturated fatty acids can be further categorized into monounsaturated fatty acids (MUFAs) and

polyunsaturated fatty acids (PUFAs) based on the number of double bonds they possess. MUFAs have one double bond, while PUFAs have two or more double bonds.

The presence of double bonds in unsaturated fatty acids affects their physical properties, such as melting point and fluidity. The more double bonds present, the lower the melting point and the more liquid the fatty acid at room temperature.

Give me some examples of fatty acids with double carbon Bonds Sure! Here are some examples of fatty acids with double carbon bonds:

1. Oleic acid: It is a monounsaturated fatty acid with one double bond, commonly found in olive oil and other vegetable oils.

2. Linoleic acid: It is a polyunsaturated fatty acid with two double bonds, commonly found in vegetable oils like sunflower oil and safflower oil.

3. Alpha-linolenic acid: It is a polyunsaturated fatty acid with three double bonds, commonly found in flaxseed oil, chia seeds, and walnuts.

4. Arachidonic acid: It is a polyunsaturated fatty acid with four double bonds, commonly found in animal fats like meat, eggs, and dairy products.

5. Docosahexaenoic acid (DHA): It is a polyunsaturated fatty acid with six double bonds, commonly found in fatty fish like salmon, mackerel, and sardines.

These are just a few examples of fatty acids with double carbon bonds. There are many more types of fatty acids with varying numbers and positions of double bonds.

Why do we need fatty acids? Why not just fat such as saturated or unsaturated fats?

Fatty acids are essential for our bodies because they play various important roles in our overall health. While fats, including saturated and unsaturated fats, provide energy and help absorb fat-soluble vitamins, fatty acids have specific functions that contribute to our well-being. Here are a few reasons why we need fatty acids:

1. Building blocks: Fatty acids are the building blocks of fats, which are essential for the structure and function of every cell in our bodies. They help form cell membranes, which control the movement of substances in and out of cells.

2. Energy storage: Fatty acids are stored as triglycerides in adipose tissue, serving as a long-term energy reserve. When the body needs energy, these stored fatty acids are broken down and released to provide fuel.

3. Hormone production: Fatty acids are crucial for the production of hormones, including steroid hormones like estrogen and testosterone. These hormones are involved in various bodily functions, such as regulating metabolism, growth, and reproduction.

4. Brain function: The brain is composed of approximately 60% fat, and fatty acids are essential for its proper functioning. Omega-3 fatty acids, in particular, play a vital role in brain development, cognitive function, and mood regulation.

5. Inflammation regulation: Some fatty acids, such as omega-3s, have anti-inflammatory properties. They help balance the body's inflammatory response, which is important for maintaining overall health and reducing the risk of chronic diseases.

While fats provide a concentrated source of energy, fatty acids are the specific components that carry out these essential functions in our bodies. It's important to consume a balanced diet that includes both fats and a variety of fatty acids to support optimal health.

So we see that there are important differences between fatty acids and fats. Therefore, methylation disturbance are important to both functions of methyl influence: fats and polymerizaton. DNA repair is another. So methylation is extremely important and any interference to that process causes major damage and interruption. A very big deal here...

Today:

- 1. Paper continuance
- 2. Bacteriophage prospect?

AI chat confirmation:

What is the electrical charge of a protein at a pH which is less than its isoelectric point?

At a pH that is less than the isoelectric point (pl) of a protein, the protein will have a net positive charge. This is because the pH is lower than the pI, meaning that the concentration of H+ ions is higher than the concentration of OH- ions in the solution.

Proteins are composed of amino acids, which have ionizable functional groups (such as amino and carboxyl groups) that can either gain or lose protons (H+ ions) depending on the pH of the solution. At a pH below the pl, the amino groups on the protein will be protonated (gain H+ ions), resulting in a net positive charge on the protein.

The magnitude of the positive charge will depend on the specific amino acid composition of the protein and the difference between the pH and the pl. The more basic amino acids (such as lysine and arginine) in the protein, the higher the positive charge will be at a given pH below the pl.

If we introduce a positively charged protein into blood will it cause increased clotting of the blood

Introducing a positively charged protein into the blood may or may not cause increased clotting, as it depends on various factors. Positively charged proteins, such as histones, can potentially interact with negatively charged components in the blood, such as platelets and red blood cells, leading to increased clotting. However, the extent of clotting will depend on the specific characteristics of the protein, its concentration, and the overall balance of clotting and anticoagulant factors in the blood. It is essential to consider that clotting is a complex process regulated by multiple factors, so the introduction of a positively charged protein alone may not necessarily cause significant clotting. It is always recommended to consult a medical professional or conduct further research for a detailed understanding of the specific protein and its effects on clotting.

2023.12.15 Paper Complete

Realy nice to have some breathing room. Paper is complete and it is especially important as it follows the progression through from the CDB source to blood clot development. It is a very logical resting point. It will raise ire among some.

Now can regroup on priorities.

The most pressing question to me is the bacteriophage prospect. This needs to be looked into further. There are, of course, infininte questions that can be asked, but what is of immediate interest and value? Bacteriophage question and panel plans are undoubtedly the highest priorities now.

Also an interest in: "Electrochemical measurements of organic redox reactions? That would open up a whole new ballgame."

Bacteriophage issue first arose on Nov 07 2023.

It looks like TSP 070 is involved (Nov 19 2023):

"9. Duplicate TSP 070

1. Nucleic acids

2. Bacteriophage"

It looks like the notes of Nov 07 2023 are going to be our best reference. Looking at notes, TSP 070 is the source of the finding. Still need to try to trace down what iteration or variation.

It is fortunate that I made notes on this in my spreadsheet on culture variations.

It is really interesting that I apparently found a bacteriophage UV profile. Now let's see if I can retract what was done for TSP 70.1D. It would appear that I took TSP 070, brought the pH up to 6.2 and decanted. At the time it was a 20 day old culture. A question is was it decanted on Nov 07 2023 and then studied, or was it decanted prior (maybe 20 days) and then studied after sitting for 20 days. Let's work out the date. TSP 070 was started on approx. Oct 20. A 20 day culture would bring it close to Nov 09. So

this matches quite well. This indicates that the titration took place on Nov 07, and then was examined by UV.

Now let's look at written notes to see if this appears to be consistent. I see we no longer have TSP 70.1D available, and this is OK since it would need to be recreated anyway.

Nov 07/ 2023 TSP 70.1D (M) 6.20-1107 TSP 70.1 Titrate Decant Titrated 20 day TSP 70 Culture Decanted This is what shows to apparently contain bacteriophages. Big issue here. Absorbance is at 269nm. Matches bacteriophage UV profile extremely well. 700-1100 NIR of same sample shows the alkyl alchol w/no hydrogen bonding at 962nm.

The pH of TSP 070 is now 3.49. This is getting very close to a 60 day old culture now. This means that a very gradual decline in the pH has continued since the base value of \sim 3.8 was established.

I have now taken a 35 ml sample of TSP 070 at pH 3.49 and have raised the pH to 6.2. Definite precipitation takes place as we recall before. We are now after decanting from the precipitate to settle on the bottom, either with time lapse or centrifugation, or both.

+

Notes of Nov 21:

TSP 070 213 261 Spreadsheet notes: Notes of today, Dec 15 2023:

and now you have on Dec 15 2023 with TSP 070.7 (new sample creation)

TSP 70.7 262.5 (260) 221 (220)

SO IT REALLY IS NOT THE SAME AS YOUR DESIGNATED DNA SAMPLE. THE SHIFT IS A LITTLE TOO MUCH.

DNA CANDIDATE? : TSP070 217 260 DNA ACTUAL : ADENINE AND TYHIME \sim 204 NM and $\sim\!260$ nm.

This is a fairly close match.

An even more interesting match with measurement at 213 nm and 260 nm vs my 213 nm and 261 nm. Can't get any closer than this.

https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-dna-absorbance-with-the-sts-uv-microspectrometer.https://www.chemeurope.com/en/whitepapers/126492/measuring-absorbance-with-the-sts-uv-microspectrometer.https://whitepapers/126492/measuring-absorbance-with-the-sts-uv-microspectrometer.https://whitepapers/126492/measuring-absorbance-with-the-sts-uv-microspectrometer.https://whitepapers/126492/whi

It shows peaks at 213 nm and 260 nm. Can't get much closer than this.

UV therefore makes a strong case for DNA. Next we study NIR and see how it correlates. Any confusion in spectra to be clarified. DNA solubility in alcohol test?

The UV differences between TSP 70 and TSP 100 are:

1. The 210 very strong peak of TSP 070 is shifted left to 205 in TSP 100.

2. The 260 peak of TSP 070 (which I now see might be interpreted to be as far to the right at 268 nm, is at 272 nm in TSP 100, and is more identifiable there than in TSP 070. THis means that in BOTH cases of TSP 70 and 100 this peak this peak could be interpreted as a protein vs headed over to 260 as Nucleic acid.

SO BE VERY CAUTIOUS HOW YOU GO ABOUT THIS. YOU NEED CLEAR SEPARATION BETWEEN THESE CHOICES.

TSP 70.7 262.5 (260) 221 (220)

DNA Assessment 260 (260) 213 (210)

These are different enough to know that something is going on.

also a sharper valley in TSP 070.7 near 243 than in DNA Assessment. also the drop from 275 -295 is also sharper in TSP 070.7 than in DNA Assessment.

There is a difference here. We need to get UV profiles side by side, and then NIR profiles side by side. Concentration is affecting the peaks as well. NIR is going to be what discerns.

2023.12.16 Bacteriophage Inquiry

I am starting to look into the bacteriophage question more systematically. Many questions coming up very quickly.

1. Need to pin down UV differences clearly between:

TSP 070 mid layer TSP 070.7 decant after pH change from 3.5 to 6.2 Settled precipitate from pH rise.

2. Then we go to NIR

Some preliminary info is available. Raises questions about vinyl in phage prospect. Genetic engineering of phages is most definitely taking place. Same samples as above to be analyzed.

3. Account for all differences between the three states.

4. Need to see if any alternate detection methods for phages exist.

Side note:

(One thing we are seeing is that the behavior of vinyl in strong H2SO4 and strong NaOH-KOH is on par with samples we have seen. Vlnyl is VERY slowly dissolving in strong H2SO4. Not dissolving in NaOH KOH. Now what we see with the clot is that it very slowly dissolved in NaOH KOH. As I recall, the synth polymer layer only dissolved in strong H2SO4, and possible to some degree in acetone. We will need to sort out these differences over time. We know we have plastics vs rubbers, but we don't know when or how much of each at the various stages between synth polymer layer and clot.)

Side note:

You have forgotten that you have a conductivity meter available. This is useful as another "vital" measurement, and especially good for LC work as well.

Refractometry pH TDS Conductivity

are all useful as vitals or general assessment tools, and assists to LC work.

We also see that concentration of the UV samples may make a big difference in the UV profile, so standardize and record this along the way to try and keep an even par of absorbance magnitude.

Our first sample is TSP 070 mid layer. Already the questions start developing. Peak at 267. Valley at 243. Peak at 217.5

or

TSP 070 Middle Layer 267.5 (270) [Magnitude here is \sim 1.6] Valley @ 243 (240) 217.5(220) [300 ul] Maximum magnitude occurs at 217.5 of Absorbance 2.8 (quite high).

Let's run a sample at 100 ul vs 300 ul in the 3 ml. cuvette.

One thing already interesting is this profile I think is already close to the bacteriophage prospect. We will need to run trials of TSP 100 also, to look for these differences as well.

Next thing that we learn is that concentration is significantly affecting both the valley depth and profile at 243 nm and also is DISPLACING the peak in the \sim 200 range. Our profile is now:

TSP 070 Middle Layer 267.5 (270) [Magnitude here is \sim 0.6] Valley @ 243 (240) 206(210) [100 ul] Maximum magnitude occurs at 217.5 of Absorbance 2.2 (still reasonably high).

Here the profile is quite different also between 100 ul and 300 ul. 100 ul is quite diminished, with gradual slopes, undefined peaks, undefined valleys, and a significantly shifted peak from 217.5 to 206 nm. These are big differences.

So there are important differences to learn from here. You need to seek to get absorbances at the peaks of interest at roughly the same magnitude, i.e, concentration FOR THAT PEAK, not necessarily the maximum absorbance.

Also maximum magnitude of peak on the profile does not necessarily help you out. It depends WHAT THE PEAK OF INTEREST actually is. In our case it is the 240-300 range that is most important right now. This range covers the valley and peaks of bacteriophage, DNA and protein. I think you need to adjust your work to zero in on this range for now.

So let's regroup already. Designate the range from 230 to 290, at 0.1 resolution. Try to get your absorbance values up near >= 1.5 for your trials in this region.

100 ul \sim 0.5 magnitude 300 ul \sim 1.5 magnitude 500 ul \sim 2.2 magnitude

Believe or not, the 300 ul profile is actually the most descriptive. 500 ul actually levels out the profile and 100 ul is not defined enough.

So a magnitude of ~ 1.5 seems to be the most desirable in this range of 230 to 290 so far.

So now we start from scratch again under these conditions.

TSP 070 Middle Layer 266.5 (270) [Magnitude here is ~1.7] Valley @ 244 (240) [300 ul] Spectrum saved.

It is not impossible that we are transitioning through two different pl's on the pH change form 3.5 to 6.2. A single drop of NaOH-KOH causes immediate coagulation of what appears to be a polymer (looks like vinyl). The solution, however, remains relatively clear. Upon further increase as the pH approaches 6.2 the solution turn cloudly. Indicates two transitions taking place. Focused titration will be required across this pH range. This could easily explain our two layer centrifugated layer, one white and one gray. NOTE.

THIS IS WHAT IS HAPPENING. YOU ARE GETTING TWO SEPARATE PRECIPITATES DURING THIS pH TRANSITION. THIS WILL EXPLAIN THE WHITE AND GREY DUAL LAYERS.

What this means therefore is that the nature of the decant will also go through two stages, so this process becomes more involved. You will have five products minimum just from the pH transition.

Original middle layer. Decant no. 1 (pH slightly above 3.5) Decant no. 2 (pH raised to 6.2) White precipitate corresponding to decant no. 1 Grey precipitate corresponding to decant no. 2.

We see that we are going to need to revamp again and study each stage of the process separately. Right now, you are using a "combined decant", and this is not the whole story. This combined decant, nevertheless, is now now providing the following:

TSP 070 Middle Layer 266.5 (270) [Magnitude here is \sim 1.7] Valley @ 244 (240) [300 ul] Spectrum saved. Native culture.

VOID

TSP 070 Combined Decant 262.4 (260) [Magnitude here is ~1.5] Valley @ 248.2 (250) [300 ul] Spectrum saved.. THIS IS NO LONGER USEFUL. YOU MUST SEPARATE EACH LEVEL INTO ITS OWN SPECTRUM. VOID

This will now be:

TSP 070 Native UV TSP 070 Middle Layer 266.5 (270) [Magnitude here is \sim 1.7] Valley @ 244 (240) [300 ul] Spectrum saved. Native culture.

TSP 070.10 Decant pH 4.0 UV TSP 070.10 264.5 (260) [Magnitude here is ~1.6] Valley @ 246.6 (240) [300 ul]

TSP 070.11 Decant pH 6.2 UV

TSP 070.12 Precip pH 4.0 NIR

TSP 070.13 Precip pH 6.2 NIR

Now can consider 3 divisions as a start, 280, 270, and 260.

280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)

Now obviously the precipitates are equally important here. You have two to analyze, as well as produce.

Now, if you look at what is going on here, the native TSP 070 is indicating a bacteriophage already in place. Then as you get to the combined decant, you are closer to DNA values.

Now as I recall when the culture was new, your readings were stronger in the protein range. This is indicating that the transition is toward:

polymer production bacteriophage DNA

as the culture ages.

Profile is also equally important to consider here.

You have a reasonably strong case for the bacteriophage existing in the native culture at 60 days old, and then DNA becoming stronger as you decant at a pH of 6.2.

We really have a lot of separation going on. Let's revamp again. The labeling of cultures will need to be

TSP 070 Native TSP 070.10 Decant 4.0 (for example) TSP 070.11 Decant 6.2 (for example) TSP 070.12 Precipitate pH 4.0 white (for example) TSP 070.13 Precipitate pH 6.2 grey (for example)

There is a lot of variable activity with the pH changes. Instead of taking the pH all the way to 4.0, it looks like you should just take it to about 3.7 to 3.8 and immediately collect the precipitate. It seems like it dissolves over time.

Also when you take the decant from this process, you get a more cloudy solution when you get to pH 6.2. I am in the process of separating that out now.

What this means is that TSP 070.10 and TSP 12 are to be replaced with the results at pH 3.8 instead of 4.0.

The stage which raises the pH from \sim 4.0 to 6.8 produces a high amount of precipitate, apparently much higher than that which results from the change from 3.5 to 4.0 (or now 3.8).

Note that the decant FROM 70.10 was used to create TSP 70.11. It was a result of two completely independent staged precipitation events. There is no "combined decant " anymore.

Concentrations are 300 ul. Now going back to the beginning in sequence with the decants:

Now can consider 3 divisions as a start, 280, 270, and 260.

280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)

TSP 070 Native UV TSP 070 Middle Layer 266.2 (270) [Magnitude here is ~1.6] Valley @ 244.7 (240) [300 ul] Spectrum saved. Native culture. This is closest in proximity to the bacteriophage range. Also nice steady drop off to protein region of 280. Delta = 1.8nm and 0.3 nm. Quite close.

TSP 070.10 Decant pH 4.0 UV TSP 070.10 261.0 (260) [Magnitude here is ~1.7] Valley @ 247.3 (250) [300 ul] This is closest to the DNA range. Nice steady drop off to the 280 range for protein measurement ratio as well. Delta = 1.0nm. Quite good with very decent 260/280 ratio.

TSP 070.11 Decant pH 6.2 UV TSP 070.10 262.5 (260) [Magnitude here is \sim 1.5] Valley @ 247.5 (250) [300 ul] Valley slope has flattened out here and peak at 262.5 also not quite as well defined.

This indicates to me that we have evidence for both bacteriophage and DNA. Bacteriophage is indicated most strongly by TSP 070 Native culture, middle layer with \sim 60 day old culture. DNA is indicated most strongly by TSP 070.10, first level decant with pH raised from 3.5 to 4.0. \sim 60 day culture.

I have significantly improved the NIR sample card process for liquid samples. Precipitates in liquid are optional. The improvement is to poke needle holes in the sample circle. This allows for greater containment and adhesion of the sample to the sample region. Looks to work well and avoid the runouts from the sample region. Dry moderately with mild heat for best results.



2023.12.17 Enzyme - Bacteriophage - DNA

Redox organic reactions remain of interest with electrochemistry.

Major observation today with the influence of the L. enzyme upon the culture. It shows a significant degrading effect upon TSP 100.2. This culture was almost discarded as it was thought to be no longer beneficial. Hardly the case. Photo will be taken. TSP 100.2 was started on Nov 08. TSP 100.2 was a low concentration enzyme addition to an non-concentrated culture trial. TSP 100.3 was a higher addition enzyme concentration to a more concentrated culture. TSP 100.4 was a control with no culture added. Cultures are approx. 150 ml. each. It is of interest that the lower concentration, almost to be discarded is by far the most impacted. TSP 100.3 does not show a significant degradation. Basically TSP 100.2 looks dead or dying, with a dark brown color. Will put under the scope. In addition, there is a foul odor presumably coming from TSP 100.2 since no other active cultures have such.

Also, the modified NIR cards are a major success. This method of a porous sample region (needle pokes over 1 inch diam circle on card) is a major improvement. Works exactly as anticipated. Confined, more concentrated, and more dispersed sample within the card sample region.

I now have five NIR plots to evaluate. They are:

TSP 070 Native TSP 070.10 Decant 4.0 (for example) TSP 070.11 Decant 6.2 (for example) TSP 070.12 Precipitate pH 4.0 white (for example) TSP 070.13 Precipitate pH 6.2 grey (for example)

We also already have the following UV information:

The stage which raises the pH from \sim 4.0 to 6.8 produces a high amount of precipitate, apparently much higher than that which results from the change from 3.5 to 4.0 (or now 3.8).

Note that the decant FROM 70.10 was used to create TSP 70.11. It was a result of two completely independent staged precipitation events. There is no "combined decant " anymore.

Concentrations are 300 ul. Now going back to the beginning in sequence with the decants: Now can consider 3 divisions as a start, 280, 270, and 260.

280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)

TSP 070 Native UV TSP 070 Middle Layer 266.2 (270) [Magnitude here is ~1.6] Valley @ 244.7 (240) [300 ul] Spectrum saved. Native culture. This is closest in proximity to the bacteriophage range. Also nice steady drop off to protein region of 280. Delta = 1.8nm and 0.3 nm. Quite close.

TSP 070.10 Decant pH 4.0 UV TSP 070.10 261.0 (260) [Magnitude here is ~1.7] Valley @ 247.3 (250) [300 ul] This is closest to the DNA range. Nice steady drop off to the 280 range for protein measurement ratio as well. Delta = 1.0nm. Quite good with very decent 260/280 ratio.

TSP 070.11 Decant pH 6.2 UV TSP 070.10 262.5 (260) [Magnitude here is \sim 1.5] Valley @ 247.5 (250) [300 ul] Valley slope has flattened out here and peak at 262.5 also not guite as well defined.

Now carrying forward.

This indicates to me that we have evidence for both bacteriophage and DNA. Bacteriophage is indicated most strongly by TSP 070 Native culture, middle layer with ~60 day old culture. DNA is indicated most strongly by TSP 070.10, first level decant with pH raised from 3.5 to 4.0. ~60 day culture.

Let ' start with TSP 070 and compare with previous analysis, use worksheet from research paper.

TSP 070 Native 60 Day Culture:

UV TSP 070 Middle Layer 266.2 (270) [Magnitude here is \sim 1.6] Valley @ 244.7 (240) [300 ul] Spectrum saved. Native culture.

This is closest in proximity to the bacteriophage range. Also nice steady drop off to protein region of 280. Delta = 1.8nm and 0.3 nm. Quite close.

NIR TSP 070 Native 933 (moderate peak) Methylene (930) 1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530)

1585 (stong broad absorbance) OH (1583)(1580)

1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl groups.

Therefore, amides, polyamides, proteins(? uncertain here see AI chat below) and vinyl groups seem to characterize the middle layer of TSP 070 60 day culture.

What is the difference between amides and proteins?

Amides and proteins are both organic compounds that contain nitrogen. However, there are some key differences between the two:

1. Structure: Amides are a type of functional group that consists of a carbonyl group (C=O) bonded to a nitrogen atom (N). Proteins, on the other hand, are complex macromolecules composed of amino acids linked together by peptide bonds.

2. Composition: Amides are relatively simple compounds, typically formed by the reaction between a carboxylic acid and an amine. Proteins, on the other hand, are large and complex molecules composed of long chains of amino acids.

3. Function: Amides have a variety of uses, such as being involved in the synthesis of pharmaceuticals, polymers, and solvents. Proteins, on the other hand, have diverse functions in living organisms, including structural support, enzymatic activity, transport, and signaling.

4. Biological significance: Amides are not typically found as major components in living organisms, although they can be found in small quantities. Proteins, on the other hand, are essential for the structure and function of cells and are one of the most abundant biomolecules in living organisms.

In summary, while both amides and proteins contain nitrogen, they differ in terms of structure, composition, function, and biological significance.

Do filamentous bacteriophages contain amides or proteins?

Filamentous bacteriophages contain proteins.

What are the characteristics of amides and how do they affect biology?

Amides are organic compounds that contain a carbonyl group (C=O) bonded to a nitrogen atom (N). They have the following characteristics:

1. Structure: Amides have a planar structure due to the presence of the double bond between carbon and oxygen. The nitrogen atom is sp2 hybridized and forms a sigma bond with the carbon atom.

2. Hydrogen bonding: The presence of a nitrogen atom with a lone pair of electrons allows amides to form hydrogen bonds. This property is important in biological systems as it contributes to the stability and structure of proteins and nucleic acids.

3. Polarity: The presence of the carbonyl group makes amides polar molecules. This polarity influences their solubility in water and other polar solvents.

4. Stability: Amides are relatively stable compounds due to the resonance effect of the carbonyl group. This stability is important for the structure and function of proteins, which are composed of amino acid amides.

In biology, amides play crucial roles in various biological processes:

1. Protein structure: Amides are found in the peptide bonds that link amino acids in proteins. These bonds provide the backbone structure of proteins and determine their folding and three-dimensional shape.

2. Enzymatic reactions: Amides can act as substrates or products in enzymatic reactions. For example, amide bonds are involved in peptide bond formation and hydrolysis, which are catalyzed by enzymes called peptidases.

3. Drug design: Many drugs and pharmaceuticals contain amide groups. The presence of amides in drug molecules can affect their stability, solubility, and interactions with biological targets.

4. Biological signaling: Certain amides, such as neurotransmitters and hormones, play crucial roles in cell signaling and communication within the body. Examples include acetylcholine, serotonin, and adrenaline.

Overall, the characteristics of amides make them important molecules in biology, contributing to the structure, function, and regulation of biological systems.

Do proteins contain amides?

Yes, proteins contain amide bonds, also known as peptide bonds. These bonds are formed through a condensation reaction between the carboxyl group of one amino acid and the amino group of another amino acid. This bond is responsible for connecting amino acids together to form a polypeptide chain, which then folds into a specific three-dimensional structure to become a functional protein.

What are the general characteristics of the following compounds?

- 1. Acetamide (CH3CONH2)
- 2. Formamide (HCONH2)
- 3. N,N-Dimethylacetamide (CH3CON(CH3)2)
- 4. N-Methylformamide (HCONHCH3)
- 5. Benzamide (C6H5CONH2)
- 6. Acrylamide (CH2=CHCONH2)
- 7. Succinimide (C4H5NO2)
- 8. Phthalimide (C8H5NO2)
- 9. Glycine (aminoacetic acid, NH2CH2COOH)
- 10. Asparagine (amino acid, HO2CCH(NH2)CH2CONH2)
- 1. Acetamide (CH3CONH2):
- Organic compound
- Contains an amide functional group
- Colorless, crystalline solid
- Soluble in water
- Has a mild odor
- Used as a solvent and in the production of certain chemicals
- 2. Formamide (HCONH2):
- Organic compound
- Contains an amide functional group
- Colorless liquid
- Miscible with water
- Has a faint odor
- Used as a solvent, in the production of certain chemicals, and as a reagent in organic synthesis

3. N,N-Dimethylacetamide (CH3CON(CH3)2):

- Organic compound
- Contains an amide functional group
- Colorless liquid
- Miscible with water
- Has a mild odor
- Used as a solvent, particularly in the production of pharmaceuticals and polymers
- 4. N-Methylformamide (HCONHCH3):
- Organic compound
- Contains an amide functional group
- Colorless liquid

- Miscible with water

- Has a faint odor
- Used as a solvent and in the production of certain chemicals
- 5. Benzamide (C6H5CONH2):
- Organic compound
- Contains an amide functional group
- White crystalline solid
- Slightly soluble in water
- Has a weak odor
- Used as an intermediate in the production of various chemicals and pharmaceuticals

6. Acrylamide (CH2=CHCONH2):

- Organic compound
- Contains an amide functional group
- White crystalline solid
- Soluble in water
- Has a faint odor
- Used in the production of polymers, as a flocculant, and in certain laboratory applications
- 7. Succinimide (C4H5NO2):
- Organic compound
- Contains an imide functional group
- White crystalline solid
- Slightly soluble in water
- Has a faint odor
- Used as a precursor in the synthesis of various chemicals and pharmaceuticals
- 8. Phthalimide (C8H5NO2):
- Organic compound
- Contains an imide functional group
- White crystalline solid
- Slightly soluble in water
- Has a faint odor
- Used as a precursor in the synthesis of various chemicals and pharmaceuticals

9. Glycine (aminoacetic acid, NH2CH2COOH):

- Organic compound
- Amino acid
- White crystalline solid
- Highly soluble in water
- Has a sweet taste
- Used as a building block in protein synthesis, as a nutrient supplement, and in certain pharmaceuticals

10. Asparagine (amino acid, HO2CCH(NH2)CH2CONH2):

- Organic compound
- Amino acid
- White crystalline solid
- Highly soluble in water
- Has a sweet taste
- Used as a building block in protein synthesis and as a nutrient supplement

So one thing I see is that amides allow for solubility in water, but they also commonly give structural integrity to things such as proteins and polymers. That is a perfect fit.

Is a protein a polyamide?

Yes, a protein is a type of polyamide. Polyamides are polymers that contain amide bonds, which are formed when a carboxylic acid group reacts with an amino group. Proteins are composed of amino acids, which are monomers that contain both an amino group and a carboxylic acid group. These amino acids are linked together through amide bonds, forming a polyamide chain, which is known as a protein.

Is a polyamide a protein?

No, a polyamide is not a protein. Polyamides are a type of synthetic polymer that contain amide groups (-CONH-) in their chemical structure. Proteins, on the other hand, are complex organic macromolecules made

up of amino acids linked together by peptide bonds.

Do filamentous bacteriophages contain amides?

No, filamentous bacteriophages do not contain amides. They are composed primarily of proteins and nucleic acids, specifically single-stranded DNA.

So here is the problem that arose and I delivered...

If all proteins contain amides and if all bacteriophages contain protein then why don't bacteriophages contain amides?

Bacteriophages do contain proteins, but not all proteins necessarily contain amides. Amides are a specific type of chemical bond found in proteins, formed between the carbonyl group of one amino acid and the amino group of another amino acid. While most proteins do contain amide bonds, there are other types of chemical bonds present in proteins as well.

Bacteriophages are viruses that infect and replicate within bacteria. They have a protein coat, known as a capsid, which protects their genetic material. This capsid is made up of proteins, and these proteins can contain amide bonds. However, it is important to note that not all proteins in bacteriophages necessarily have amide bonds. The presence or absence of amide bonds in proteins depends on the specific amino acid sequence and the type of chemical bonds formed between the amino acids.

In summary, while bacteriophages do contain proteins, the presence or absence of amide bonds in these proteins depends on their specific amino acid sequence and the type of chemical bonds formed.

Can bacteriophages contain amides?

No, bacteriophages do not contain amides. Bacteriophages are viruses that specifically infect and replicate within bacteria. They are composed of genetic material (either DNA or RNA) enclosed in a protein coat called a capsid. They do not have the ability to synthesize or incorporate amides into their structure.

I am definitely trying to pin this fellow down, since he is weasling right and left:

Can the protein capsid of a bacteriophage contain amides?

No, the protein capsid of a bacteriophage does not typically contain amides. The protein capsid is composed of repeating protein subunits called capsomeres, which are usually made up of amino acids linked together by peptide bonds. Amides, on the other hand, are a specific type of organic compound that contain a carbonyl group bonded to a nitrogen atom. While amides can be found in other biological molecules such as proteins and peptides, they are not typically found in the protein capsid of a bacteriophage.

Describe the chemical composition of a capsomere of a bacteriophage

A capsomere is a structural unit of a bacteriophage, which is a virus that infects bacteria. It is composed of proteins and plays a vital role in forming the capsid, the protein coat that encloses the genetic material of the virus.

The main protein component of a capsomere is typically called a protomer. Multiple protomers come together to form the capsomere, and several capsomeres assemble to create the capsid.

The protomers are often arranged in a symmetrical pattern, giving the capsid its characteristic shape. The arrangement can vary depending on the specific bacteriophage, but common symmetrical patterns include icosahedral, helical, or complex shapes.

The proteins in a capsomere are usually highly stable and resistant to environmental conditions, as they need to protect the viral genetic material during transmission and infection. They also play a crucial role in recognizing and binding to specific receptors on the surface of bacterial cells, allowing the virus to attach and initiate the infection process.

Overall, the chemical composition of a capsomere primarily consists of proteins, which form the structural framework of the bacteriophage and contribute to its infectivity and stability.

Does the capsomere of a bacteriophage contain amides?

No, the capsomere of a bacteriophage does not contain amides. Capsomeres are protein subunits that make up the capsid, which is the outer protein coat of a bacteriophage. Capsomeres are typically composed of amino acids, which are the building blocks of proteins, but they do not contain amides specifically.

OK, you are finally getting there. A bacteriophage does not normally (and seems like seldom to never) amide functional groups, but does contain protein. This is a very important distinction.

Now if we look at our UV spectra for TSP 070, we can conceiveably make the case that it matches spectral characteristics of a filamentous bacteriophage.

But our NIR plot is not so clear, because it seems to present likely cases of :

NIR TSP 070 Native

933 (moderate peak) Methylene (930)

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry.

1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530)

1585 (stong broad absorbance) OH (1583)(1580) There is no real competition here.

1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl groups.

Now to work through this , we forgot to inspect the 1585 absorbance more closely. If we dismiss moderate or weak peaks, valleys, etc, we are left with:

amide OR protein, secondary amine, OH, and vinyl.

if we choose protein over amide, and amine, OH and vinyl exist, this indicates a candidate of a genetically engineered bacteriophage. This is not at all impossible, is it? Once you cross that line, you have a lot of things that can make sense.

and so we have:

Is it possible to genetically engineer A bacteriophage to contain vinyl functional groups?

Yes, it is theoretically possible to genetically engineer a bacteriophage to contain vinyl functional groups. Bacteriophages are viruses that infect bacteria, and they can be modified through genetic engineering techniques to introduce specific genetic material or functional groups.

To introduce vinyl functional groups, you would need to identify and modify the relevant genes responsible for producing the desired functional groups. This could involve inserting genes encoding enzymes or proteins involved in vinyl group synthesis, or modifying existing genes to produce vinyl groups.

The process of genetically engineering a bacteriophage involves several steps, including isolating and manipulating its genetic material, introducing the desired modifications using techniques like DNA recombination or gene editing, and then reintroducing the modified genetic material into the bacteriophage.

It is important to note that the feasibility and success of such genetic engineering experiments can vary depending on the specific bacteriophage and the desired modifications. Additionally, ethical considerations and regulatory guidelines may need to be taken into account when conducting genetic engineering experiments.

Now let's look at the generalized NIR chart:

Our target areas are restricted to 1530 nm, 1585 nm and 1640 nm. On the ASD generalized NIR chart:

1530 would be RNH2 (amine) 1585 is not a good match for anything 1640 is ArCH

and so we ask:

Can synthetic vinyl polymers contain an aromatic hydrocarbon?

Yes, synthetic vinyl polymers can contain aromatic hydrocarbons. Vinyl polymers are made from monomers containing a vinyl group, such as vinyl chloride or vinyl acetate. These monomers can be copolymerized with other monomers, including those containing aromatic hydrocarbons, to produce vinyl polymers with desired properties. For example, styrene (an aromatic hydrocarbon) can be copolymerized with vinyl chloride to produce a vinyl polymer called polyvinyl chloride (PVC).

Do bacteriophages contain amines?

No, bacteriophages do not contain amines. Bacteriophages are viruses that specifically infect and replicate within bacteria. They consist of genetic material, either DNA or RNA, enclosed in a protein coat called a capsid. Amines, on the other hand, are organic compounds that contain a nitrogen atom bonded to one or more carbon atoms and hydrogen atoms. They are commonly found in biological systems and play important roles in various biochemical processes.

I definitely know how to produce contradictions in the AI chat:

Does the bacteriophage capsid contain amines?

Yes, the bacteriophage capsid can contain amines. Amines are organic compounds that contain a nitrogen atom bonded to hydrogen atoms. They are often found in proteins, which make up the structure of the capsid. Amino acids, the building blocks of proteins, contain amine groups. Therefore, the capsid of a bacteriophage, which is composed of proteins, can contain amines.

This is certainly beginning to paint a portrait that is consisent with the presence of a bacteriophage.

But this now, is VERY important to reconcile:

What is the chemical composition of filamentous bacteriophage dNA? Does it contain a hydroxyl functional group?

The filamentous bacteriophage DNA is composed of deoxyribonucleotides, which consist of a deoxyribose sugar, a phosphate group, and one of four nitrogenous bases (adenine, guanine, cytosine, or thymine). It does not contain a hydroxyl functional group; instead, it has a hydrogen atom attached to the 2' carbon of the deoxyribose sugar.

But now look at this. This was and is a part of your case for the existence of DNA:

Does DNA contain a hydroxyl group

No, DNA does not contain a hydroxyl group. A hydroxyl group (-OH) is a functional group that consists of an oxygen atom bonded to a hydrogen atom. In DNA, the sugar component (deoxyribose) does not have a hydroxyl group attached to the 2' carbon atom, which distinguishes it from RNA.

It will be very important to reconcile this information.

We can see in the image below very clearly along with text description that a hydroxyl group does exist in the structure of DNA. Be very cautious of AI Chat responses, they are suprisingly variable when trying to pin them down to absolutes, just like I am...

https://socratic.org/questions/what-are-the-functional-groups-of-dna

An amide group is an amine with a carbonyl attached. An amide is basically a carbonyl extension of an amine, and the carbonyl bond is stabilizing as well as bringing polarity (solubility) into the picture.Boy that's interesting

DNA is dominated by amines, and then further by amides, phosphates and sugars.

Does DNA contain Amides or amines?

DNA contains both amides and amines. The backbone of DNA is made up of phosphate groups, which are connected to the sugar molecules through phosphodiester bonds. The sugar molecules in DNA contain amide functional groups. Additionally, the nitrogenous bases in DNA, adenine (A), cytosine (C), guanine (G), and thymine (T), contain amine functional groups. These amine groups are involved in hydrogen bonding and base pairing, which are important for the structure and function of DNA.

DNA does contain a hydroxyl group true or false?

False. DNA does not contain a hydroxyl group. It contains a deoxyribose sugar molecule, which lacks a hydroxyl group at the 2' position of the sugar ring. This is why it is called deoxyribonucleic acid (DNA) as opposed to ribonucleic acid (RNA), which does contain a hydroxyl group at the 2' position.

I will therefore keep working on this. Conflicts continue...

What is the difference between dNA and RNA? What is the difference in their purpose and function? What is the importance of the hydroxyl group with respect to those differences?

DNA (deoxyribonucleic acid) and RNA (ribonucleic acid) are both nucleic acids that play crucial roles in the storage and transmission of genetic information within living organisms. While they share some similarities, there are several key differences between DNA and RNA.

1. Structure: DNA is a double-stranded molecule that forms a double helix structure, whereas RNA is usually single-stranded. DNA consists of a sugar-phosphate backbone with four nitrogenous bases: adenine (A), thymine (T), cytosine (C), and guanine (G). RNA also has a sugar-phosphate backbone, but it contains the base uracil (U) instead of thymine.

2. Sugar Component: The sugar component in DNA is deoxyribose, which lacks a hydroxyl group (-OH) on the 2' carbon. In contrast, RNA contains ribose, which has a hydroxyl group on the 2' carbon.

3. Stability: DNA is more stable than RNA due to the absence of the hydroxyl group on the 2' carbon of deoxyribose. This hydroxyl group makes RNA more susceptible to degradation by enzymes and chemical reactions.

4. Function: DNA serves as the primary genetic material, storing the instructions needed for the development, functioning, and reproduction of living organisms. It is mainly found in the nucleus of cells. RNA, on the other hand, has various functions, including transferring genetic information from DNA to protein synthesis machinery (messenger RNA or mRNA), catalyzing biochemical reactions (ribozymes), and assisting in the assembly of proteins (transfer RNA or tRNA).

The hydroxyl group on the 2' carbon of ribose in RNA is important for its function and versatility. This hydroxyl group allows RNA to participate in various chemical reactions and form stable secondary structures. Additionally, the presence of the hydroxyl group in RNA enables it to be more reactive and flexible than DNA, allowing it to perform its diverse roles in cellular processes.

Do bacteriophages contain dNA or RNA?

Bacteriophages can contain either DNA (deoxyribonucleic acid) or RNA (ribonucleic acid) as their genetic material. However, the majority of bacteriophages have DNA genomes.

How is this for an answer that covers all bases? Looks like anything goes! Now she is saying that DNA DOES have a hydroxyl group.

What is single strand DNA? What is the difference between single strand DNA and RNA? Does single strand dNA contain a hydroxyl group? Do bacteriophages contain dNA or RNA? Do bacteriophages contain only single strand dNA? Does RNA have both single and double strand forms? Is there a hydroxyl group on bacteriophage single strand dNA?

Single strand DNA (ssDNA) refers to a DNA molecule that consists of a single strand of nucleotides. It is typically found as a result of DNA replication or during certain stages of DNA repair.

The main difference between single strand DNA and RNA (ribonucleic acid) is the sugar component in their nucleotides. DNA contains deoxyribose sugar, while RNA contains ribose sugar. Additionally, RNA uses uracil (U) as a base instead of thymine (T) found in DNA.

Regarding the hydroxyl group, single strand DNA does contain a hydroxyl group (-OH) on its sugar component, just like double strand DNA. This hydroxyl group is important for DNA replication and other enzymatic processes.

Bacteriophages can contain either DNA or RNA, depending on the type of bacteriophage. However, the majority of bacteriophages contain DNA as their genetic material.

Bacteriophages can have both single strand DNA (ssDNA) and double strand DNA (dsDNA) forms. Some bacteriophages have a single strand DNA genome, while others have a double strand DNA genome.

RNA can exist in both single strand and double strand forms. Single strand RNA (ssRNA) is more common and is involved in various cellular processes such as protein synthesis. Double strand RNA (dsRNA) is less common but can be found in certain viruses or during RNA interference (RNAi) processes.

As for the hydroxyl group on bacteriophage single strand DNA, it depends on the specific nucleotide sequence. Each nucleotide in DNA has a hydroxyl group on its sugar component. However, it's important to note that the hydroxyl group is not present on the phosphate backbone of the DNA molecule.

"Deoxyribose Sugar

The deoxyribose sugar of the DNA backbone has 5 carbons and 3 oxygens. The carbon atoms are numbered 1', 2', 3', 4', and 5' to distinguish from the numbering of the atoms of the purine and pyrmidine rings. The hydroxyl groups on the 5'- and 3'- carbons link to the phosphate groups to form the DNA backbone. Deoxyribose lacks an hydroxyl group at the 2'-position when compared to ribose, the sugar component of RNA."

http://www.biology.arizona.edu/biochemistry/activities/DNA/10t.html

Another major observation/discovery today. The TSP 070 culture (60 days) does appear to be generating an electromagnetic field. Numerous measurements with an EF meter. Human presence is a part of the circuit.



We see a glycoside linkage (O-C-N-) between the sugar and the base, and a hydroxyl group (-OH) and a phosphate ester $(R-O-PO_3^{2-})$ on the sugar.

Finally, we look at a DNA strand.


2023.12.18 DNA - Bacteriophage Inquiry

These notes carry on continuously from Nov 17 2023.

Three research fields of future:

- 1. EF field detected in TSP 070 culture. (volts/meter)
- 2. Organic redox reactions?
- 3. The demise of TSP 100.2 with enzymes
- 4. DNA, Protein, Bacteriophage separation
- 5. Health overlap between blood clots, culture products, and health symptoms

In essence we have 5 NIR plots, and we are attempting to distinguish between 3 different structures:

- 1. Proteins
- 2. DNA
- 3. Bacteriophage

In addition, however, we need to learn to distinguish between:

- 1. Amides and Amines (Amides stabilize amines to allow protein development)
- 2. Single strand and double strand DNA if it is possible
- 3. RNA (additional hydoroxyl group, higher solubility) and DNA

There is a fair amount of confusion and contradiction taking place in the AI chats regarding the presence of absence of hydroxyl groups in DNA. Also the presence or lack of amides within the capsin, capsomeres, protein of bacteriophages. You are working on sorting that out.

In addition, we see that the first division of spectroscopy is taking place at approximately the following areas:

260 DNA (260/280 ratio with sharp dropoff) 270 Bacteriophage (259-261 area and valley at 245) 280 Protein (274-280 - broader range of accomodation)

The five NIR plots we have are:

TSP 070 Native TSP 070.10 Decant pH 4.0 TSP 070.11 Decant pH 6.2 TSP 070.12 Precipitate pH 4.0 white TSP 070.13 Precipitate pH 6.2 grey

We combine the NIR with preceding UV one at a time in effort to assess between 3 categories above, protein, DNA, bacteriophage.

We have learned that UV can at times have shifts from concentration variance alone so be very careful on that.

I have interesting and often contradictory information coming from AI response and reference sources on amides, amines, proteins, capsids, capsomeres, DNA, hydroxyl groups, single strand, double strand, etc, - keep sorting it out.

I am taking them one at a time with an eye to discern the topics above.

-

Start by recalling our data for TSP 070 Native culture:

TSP 070 Native

UV TSP 070 Middle Layer 266.2 (270) [Magnitude here is \sim 1.6] Valley @ 244.7 (240) [300 ul] Spectrum saved. Native culture.

This is closest in proximity to the bacteriophage range. Also nice steady drop off to protein region of 280. Delta = 1.8nm and 0.3 nm. Quite close.

NIR TSP 070 Native 933 (moderate peak) Methylene (930)

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530)

1585 (stong broad absorbance) OH (1583)(1580)

1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl groups.

Therefore, amides, polyamides, proteins(? uncertain here see AI chat below) and vinyl groups seem to characterize the middle layer of TSP 070 60 day culture.

I see there is another aspect we can use to try and discern between the functional groups, and this is the valleys. Lack of absorbance can be valuable as well. So in this case let's also give notice to:

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

Notice that DNA does not actually have aromatic amines from what a see. Cyclic, yes, but not aromatic.

and here we go:

Does DNA have aromatic amines?

No, DNA does not contain aromatic amines. Aromatic amines are organic compounds that have an amine group (-NH2) attached to an aromatic ring. DNA, on the other hand, is a complex molecule made up of nucleotides, which consist of a sugar, a phosphate group, and a nitrogenous base. The nitrogenous bases in DNA are adenine (A), cytosine (C), guanine (G), and thymine (T), none of which are aromatic amines.

Assuming our question is directed toward DNA existence, this now reduces the 1428 valley set to:

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430)

Do recall that these valley signals are actually very weak overall in comparison to the strong peaks, so weight that accordingly. Now DNA absolutely has amides in it. It seems to me that it has amines in it, however, this is not stated to be the case, so be careful here also:

Now we see that we must be incredibly cautious with use of the so called AI chats. I was beginning to come to the same conclusion from yesterday's inquiries.

Let's get our terminology correct on amines. You had it quite wrong. Your staring point is the nitrogen atom. What matters is how many CARBON atoms are attached (NOT HYDROGEN!) to the nitrogen.

1 carbon attached = primary amine 2 carbons attached = secondary amine 3 carbons attached = tertiary amine

carbon form can be either akyl OR aryl. Aryl requires aromatic. You need also to know cyclic, heterocycic, alicyclic. also cycloalkane, cycloalkene, cycloalkene.

In organic chemistry, a cycloalkene is a type of alkene hydrocarbon which contains a closed ring of carbon atoms and either ONE OR MORE double bonds,

and for amides, it is: (so it is the same idea as with amines).

A primary (1°) amide has nitrogen attached to a single carbon; a secondary (2°) amide has the nitrogen attached to two carbons; a tertiary (3°) amide has the nitrogen attached to three carbons.

Amide - Wikipedia

Amides are qualified as primary, secondary, and tertiary according to whether the amine subgroup has the form -NH 2, -NHR, or -NRR', ... In primary and secondary amides, the presence of N-H dipoles allows amides to function as H-bond donors as well. Thus amides can participate in hydrogen bondin

24.1: Structural, Physical, and Spectral Characteristics of Amides A strong carbonyl absorption is evident in the spectra of all amides, although the frequency of absorption varies somewhat with the structure of the amide. Thus primary amides generally absorb near \(1680 \:

 $text{cm}^{-1})$, whereas secondary and tertiary amides absorb at slightly lower

A strong carbonyl absorption is evident in the spectra of all amides, although the frequency of absorption varies somewhat with the structure of the amide. Thus primary amides generally absorb near 1680cm-1 , whereas secondary and tertiary amides absorb at slightly lower frequencies. The N-H stretching frequencies of amides are closely similar to those of amines and show shifts of 100cm-1 to 200cm-1 to lower frequencies as the result of hydrogen bonding. Primary amides have two N-H bands of medium intensity near 3500cm-1 and 3400cm-1 , whereas secondary amides, to a first approximation, have only one N-H band near 3440cm-1

So the essence is you start with the nitrogen. Then you count the carbons attached to that nitrogen. The amide contains the carbonyl group.

you then need to be clear on

aliphatic aryl cycloalkane, cycloalkene, cycloalkyne aromatic heterocyclic primary, secondary, and tertiary amine primary, secondary, and tertiary amide

and to be so is very helpful.

LOOK CLOSELY AT THIS SECTION!

Does DNA have primary or secondary amines?

DNA does not have primary or secondary amines. It is composed of nucleotides, which consist of a nitrogenous base, a sugar molecule, and a phosphate group. The nitrogenous bases in DNA are adenine (A), cytosine (C), guanine (G), and thymine (T), and they do contain nitrogen atoms, but they are not classified as primary or secondary amines.

AND QUITE THE SURPRISE HERE:

Does DNA have Amides?

No, DNA does not contain amides. DNA is composed of nucleotides, which consist of a sugar molecule (deoxyribose), a phosphate group, and a nitrogenous base (adenine, thymine, cytosine, or guanine). Amides, on the other hand, are a type of organic functional group that contain a carbonyl group (C=O) bonded to a nitrogen atom.

NOW LOOK AT THIS RESPONSE FROM YESTERDAY:

Does DNA contain Amides or amines?

DNA contains both amides and amines. The backbone of DNA is made up of phosphate groups, which are connected to the sugar molecules through phosphodiester bonds. The sugar molecules in DNA contain amide functional groups. Additionally, the nitrogenous bases in DNA, adenine (A), cytosine (C), guanine (G), and thymine (T), contain amine functional groups. These amine groups are involved in hydrogen bonding and base pairing, which are important for the structure and function of DNA.

THIS IS INSANE. COMPLETELY CONTRADICTORY INFORMATION DEPENDING ON HOW THE SAME QUESTION. THIS TELLS YOU TO BE VERY CAREFUL AND DO NOT ASSUME AICHATS ARE VALID. VERY VERY DANGEROUS. CROSS CHECKING IS REQUIRED AT ALL TIMES.

You are going to have to analyze this by yourself with Chat acting only as an assist, and not exactly the most competent one.

So my conclusion is that DNA has both amines (primary and secondary) and amides and hydroxyl groups in it. So take that Mr Chat. But, at the same time, DNA does not have aromatic amines it (cyclic but not aromatic). I see two primary amides in DNA but no secondary amides.

So now we start studying the structure of DNA independently.

1. We see that the bases are clearly heterocylic.

- 2. They are all also cycloakenes.
- 3. They are all also secondary amines. (this is now a very important distinction to start making).

4. Three out of four of the bases are also secondary amides. One of the bases does not contain an amide.

5. None of them are aromatics.

This is now very important information to include and discern in our structural analysis we make. Go back to the beginning.

Start by recalling our data for TSP 070 Native culture:

TSP 070 Native

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I see there is another aspect we can use to try and discern between the functional groups, and this is the valleys. Lack of absorbance can be valuable as well. So in this case let's also give notice to:

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

The next requirement is to try and find out the chemical structure of bacteriophage single strand DNA. That is becoming quite demanding.

What is the chemical structure in detail for single strand bacteriophage dNA?

The chemical structure of single-strand bacteriophage DNA (ssDNA) is composed of nucleotides. A nucleotide is made up of three components: a phosphate group, a sugar molecule (deoxyribose in the case of DNA), and a nitrogenous base.

In ssDNA, the sugar-phosphate backbone forms the structural framework of the molecule. The phosphate group of one nucleotide is linked to the sugar molecule of the adjacent nucleotide through a phosphodiester

bond, creating a linear chain.

The nitrogenous bases, which include adenine (A), thymine (T), cytosine (C), and guanine (G), are attached to the sugar molecule. The bases are responsible for the genetic information encoded in the DNA sequence. In ssDNA, the bases can occur in any order along the sugar-phosphate backbone.

The nitrogenous bases pair with each other in a complementary manner. Adenine (A) pairs with thymine (T) through two hydrogen bonds, while cytosine (C) pairs with guanine (G) through three hydrogen bonds. This base pairing allows the ssDNA to fold back on itself and form secondary structures, such as hairpins or stem-loop structures.

Overall, the chemical structure of ssDNA consists of a linear chain of nucleotides, with the sugar-phosphate backbone providing stability and the nitrogenous bases encoding genetic information.

Does single strand DNA of a filamentous bacteriophage contain a hydroxyl group?

No, single-stranded DNA of a filamentous bacteriophage does not contain a hydroxyl group. The hydroxyl group (-OH) is typically found on the 3' end of the DNA strand, which is involved in DNA replication and synthesis. However, filamentous bacteriophages have a circular single-stranded DNA genome that lacks free ends and therefore does not have a 3' hydroxyl group.

Does single strand DNA of a filamentous bacteriophage contain a Alcohol functional group?

No, single-stranded DNA of a filamentous bacteriophage does not contain an Alcohol functional group. The Alcohol functional group is characterized by a hydroxyl (-OH) group attached to a carbon atom. DNA, whether single-stranded or double-stranded, does not have an Alcohol functional group.

TALK ABOUT CONTINUED CONFUSION:

Does the sugar within DNA contain an alcohol functional group?

No, the sugar within DNA does not contain an alcohol functional group. The sugar in DNA is called deoxyribose, which is a 5-carbon sugar molecule. It has five hydroxyl groups (-OH) attached to its carbon atoms, but these are not alcohol functional groups.

Why are the hydroxyl groups attached to the sugar of DNA not considered to be alcohol functional groups?

The hydroxyl groups attached to the sugar in DNA are not considered to be alcohol functional groups because they are involved in the formation of phosphodiester bonds between nucleotides. In DNA, each sugar molecule has three hydroxyl groups: one attached to the 3' carbon, one attached to the 5' carbon, and one attached to the 2' carbon.

The 3' hydroxyl group of one sugar molecule reacts with the phosphate group of another sugar molecule, forming a phosphodiester bond. This bond connects the sugar-phosphate backbone of the DNA molecule. The 5' hydroxyl group of the sugar is also involved in the formation of phosphodiester bonds.

Since these hydroxyl groups are involved in the structural integrity of DNA and play a crucial role in the formation of the DNA backbone, they are not considered to be alcohol functional groups in the traditional sense.

OK, I AM FINALLY STARTING TO FIGURE OUT WHERE THEY ARE COMING FROM. IT DOES NOT MEAN I COMPLETETLY AGREE. THIS IS SAYING IF YOU HAVE OH AND H AND GET WATER FROM IT THAT THE OH IS NOT CONSIDERED AN OH FUNCTIONAL GROUP BECAUSE IT IS NOT ATTACHED TO A CARBON. BUT IT STILL HAS ALL THE PROPERTIES OF AN OH GROUP.

What is the definition of a hydroxyl group? What is the definition of a alcohol functional group? What is the difference specifically between a hydroxyl group and an alcohol group? Under what conditions is a hydroxyl group not considered to be an alcohol functional group?

A hydroxyl group is a chemical functional group consisting of an oxygen atom bonded to a hydrogen atom. It is represented by the formula -OH. The hydroxyl group is commonly found in alcohols, phenols, and carboxylic acids.

An alcohol functional group, on the other hand, specifically refers to a hydroxyl group attached to a carbon atom. It is represented by the formula R-OH, where R represents an alkyl or aryl group. Alcohols are a class of

organic compounds that contain one or more hydroxyl groups attached to carbon atoms.

The main difference between a hydroxyl group and an alcohol functional group is that a hydroxyl group can be found in various compounds, including alcohols, phenols, and carboxylic acids, while the alcohol functional group specifically refers to the hydroxyl group attached to a carbon atom.

Under certain conditions, a hydroxyl group may not be considered an alcohol functional group. For example, in inorganic chemistry, hydroxyl groups attached to metal ions are not referred to as alcohol functional groups since they do not involve carbon atoms.

So the answer is that for it to be considered an alcohol it must have carbon attached. But it does remain a hydroxyl as it does not need to be attached to anything specifically. OK, this was painful but very necessary and important to understand. DNA has numerous hydroxyl groups but they are not considered to be alcohol functional groups. Very helpful now to be able to proceed, especially with NIR interpretation of dried samples.

Now we can agree that both primary amides and aromatic amines are not in DNA, so this is true and conforms to DNA existence.

1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

and now for the other valley:

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. So we will now go the the Galaxy NIR char since there is no CRC 2012 entry: On the Galaxy NIR chart, 1086 does correspond to NIR of ArCH. This is also valid for DNA, there are no aromatics in DNA.

Therefore the valley findings are coincident with DNA existence.

NIR TSP 070 Native 933 (moderate peak) Methylene (930)

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530) [alkyne is not consistent with DNA so to be dismissed]

1585 (stong broad absorbance) OH (1583)(1580)

1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl groups.

At 933 which is a weak(actually VERY LOW absorbance) peak, it we attach methylene, this does not exist in DNA, but the signal is so weak that strong priority cannot be given to it. In Galaxy NIR, 933, also correponds to CH and CH2 and ROH all would be possibilities.

We can see that the "hydroxyl group" in DNA is fraught with all kinds of issues. Let's delay on this for right now as it is a very involved topic.

This guy just keeps beating around the bush like nobody I have seen....

Does DNA contain oxygen hydrogen bonds Or hydrogen oxygen bonds? Does single strand DNA contain oxygen hydrogen bonds? Are there hydroxyl groups in DNA? Are there hydroxyl groups in RNA? Are there hydroxyl groups in single strand DNA of filamentous bacteriophages?

DNA contains hydrogen bonds between the oxygen atom in the phosphate group and the hydrogen atoms in the deoxyribose sugar. These are known as oxygen hydrogen bonds.

Single-stranded DNA also contains oxygen hydrogen bonds between the phosphate group and the deoxyribose sugar.

DNA does not contain hydroxyl groups. These groups are found in RNA, where the ribose sugar has a hydroxyl group attached to the 2' carbon.

Filamentous bacteriophages have single-stranded DNA, but they do not have hydroxyl groups in their DNA.

Filamentous bacteriophages, such as M13 phage, do not contain oxygen hydrogen bonds. These phages consist of a single-stranded DNA genome that is coated with proteins, and the DNA does not form hydrogen bonds with oxygen atoms.

Does a filamentous bacteriophage contain oxygen hydrogen bonds? Does DNA contain oxygen hydrogen

bonds? Does RNA contain oxygen hydrogen bonds? What are all of the chemical bond types in filamentous bacteriophages?

However, both DNA and RNA do contain oxygen hydrogen bonds. In DNA, oxygen atoms in the phosphate backbone form hydrogen bonds with hydrogen atoms in the sugar molecules. Similarly, in RNA, oxygen atoms in the ribose sugar also form hydrogen bonds with hydrogen atoms.

Regarding the chemical bond types in filamentous bacteriophages, the main types of bonds present are covalent bonds. Covalent bonds are formed between atoms within the DNA backbone and between amino acids in the proteins that coat the phage. Additionally, non-covalent interactions such as hydrogen bonds, electrostatic interactions, and hydrophobic interactions play a role in stabilizing the overall structure of the phage.

What specific molecular chemical bonds exist within filamentous bacteriophages?

Filamentous bacteriophages, such as M13 and fd phages, are composed of a single-stranded DNA genome enclosed within a protein coat. The specific molecular chemical bonds present in these phages include:

1. Phosphodiester bonds: These are the covalent bonds that connect the nucleotides within the DNA strand. Phosphodiester bonds are formed between the phosphate group of one nucleotide and the sugar molecule of the adjacent nucleotide.

2. Hydrogen bonds: These weak bonds form between complementary bases of the DNA strands. Adenine (A) forms two hydrogen bonds with thymine (T), while guanine (G) forms three hydrogen bonds with cytosine (C).

3. Disulfide bonds: Some filamentous phages, like M13, have cysteine residues on their coat proteins that can form disulfide bonds. These bonds are formed through the oxidation of two cysteine residues, resulting in the linkage of their sulfur atoms.

It is important to note that the specific chemical bonds may vary slightly depending on the specific type or strain of filamentous bacteriophage.

This one seems to give the answer I am after:

Tell me about the deoxyribose sugar. Does it have oxygen hydrogen bonds? Does a filamentous bacteriophage contain deoxyribose sugar?

Deoxyribose is a five-carbon sugar molecule that is a key component of DNA (deoxyribonucleic acid). It is similar to ribose, which is the sugar found in RNA (ribonucleic acid), but with one key difference - deoxyribose lacks an oxygen atom on the 2' carbon, hence the name "deoxy."

Deoxyribose sugar does have oxygen-hydrogen bonds, as it contains several hydroxyl (-OH) groups attached to the carbon atoms. These hydroxyl groups can form hydrogen bonds with other molecules, such as phosphate groups in DNA.

A filamentous bacteriophage is a type of virus that infects bacteria. Unlike DNA viruses, which have double-stranded DNA, filamentous bacteriophages have single-stranded DNA genomes. The sugar component in the DNA of filamentous bacteriophages is deoxyribose, making it consistent with the sugar found in other DNA molecules.

I can get a different answer each time I ask the same question. This answer satisfies the proposition that OH bonds exist in both DNA and filamentous bacteriophage DNA.

Now back to the Galaxy /CRC interpretation of the 933 peak in TSP 070:

At 933 which is a weak(actually VERY LOW absorbance) peak, it we attach methylene, this does not exist in DNA, but the signal is so weak that strong priority cannot be given to it. In Galaxy NIR, 933, also correponds to CH and CH2 and ROH all would be possibilities.

I verify the following by inspection of the DNA structure:

Does DNA have carbon hydrogen bonds? Does DNA contain a methyl functional group? Does DNA contain a methylene functional group?

Yes, DNA does have carbon-hydrogen (C-H) bonds. These bonds are present in the sugar-phosphate backbone

of the DNA molecule.

DNA does not contain a methyl functional group. However, DNA can undergo a process called methylation, where a methyl group (-CH3) is added to the DNA molecule. This methylation can affect gene expression and play a role in various cellular processes.

DNA does not contain a methylene functional group (-CH2-). The backbone of DNA is composed of a sugar-phosphate chain, with the sugar being deoxyribose in the case of DNA.

This now only leaves the ROH group as a candidate. We have already determined that DNA contains "several hydroxyl groups (OH) attached to the carbon atoms. This is therefore consistent with DNA presence.

What we are actually doing here is to see if we have a NIR spectrum that is consistent with the existence of proteins and DNA, as we see that it is already consistent with UV absorption of a filamentous bacteriophage. If it is consistent with proteins, amides, secondary amines, hydroxyl groups, then it is consistent with bacteriophage presence.

So next we go to the 1585 nm peak. In CRC, the detials are important. It actually states that the OH occurs in combination with the vinyl alcohol spectra of the literature. Well, how is that for a match with the last peak of 1640 nm being vinyl.

933 (moderate peak) Methylene (930) [ROH from Galaxy]

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530) [alkyne is not consistent with DNA so to be dismissed]

1585 (stong broad absorbance) OH (1583)(1580)

1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl groups.

We actually have everything consistent with the proposition of a filamentous bacteriophage within TSP 070. This is quite astounding. Not only that, it is actually consistent with a genetically engineered filamentous bacteriophage that contains vinyl polymeric production groups.4:00 earlier well I guess you don't want that going on okay that's pretty good actually



Heterocyclic compounds

2023.12.19 DNA - Bacteriophage Inquiry

Continuing on with the notes of Dec 17 2023.

We have now made a first pass on TSP 070 in its native form, and have good cause to suspect the existence of a bacteriophage. We may have a paper developing, "The Case for a Bacteriophage".

Now let's go on to the 2nd of 5 sample runs with NIR available on each.

In addition, we see that the first division of spectroscopy is taking place at approximately the following areas: 280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)

The five NIR plots we have are:

TSP 070 Native TSP 070.10 Decant pH 4.0 TSP 070.11 Decant pH 6.2 TSP 070.12 Precipitate pH 4.0 white TSP 070.13 Precipitate pH 6.2 grey

We combine the NIR with preceding UV one at a time in effort to assess between 3 categories above, protein, DNA, bacteriophage.

We have learned that UV can at times have shifts from concentration variance alone so be very careful on that.

I shall work on TSP 070.10.

We also now have a much better handle on the classification of major organic systems, such as aliphatic, aryl, cycloalkane, cycloalkene, cycloalkyne, aromatic, heterocyclic, primary, secondary and tertiary amines and amides.

This now becomes very important in discerning and interpreting the NIR when finer distinctions are required, such as in this sample series.

TSP 070 Native UV TSP 070 Middle Layer 266.2 (270) [Magnitude here is ~1.6] Valley @ 244.7 (240) [300 ul] Spectrum saved. Native culture. This is closest in proximity to the bacteriophage range. Also nice steady drop off to protein region of 280. Delta = 1.8nm and 0.3 nm. Quite close.

TSP 070.10 Decant pH 4.0 UV TSP 070.10 261.0 (260) [Magnitude here is ~1.7] Valley @ 247.3 (250) [300 ul] This is closest to the DNA range. Nice steady drop off to the 280 range for protein measurement ratio as well. Delta = 1.0nm. Quite good with very decent 260/280 ratio.

Now the first thing we notice is that the 261.0 UV absorbance speaks more closely to DNA vs Bacteriophage. Also the valley has shifted to the right. So the major peak moves left, the valley on the left moves to the right. Also the profile is in good form for the 260/280 ratio. This was the case use for DNA presentation.

Now we go to NIR.

930 (minor peak) Methylene (930):

"From Dec 18 2023 Notes:

At 933 which is a weak(actually VERY LOW absorbance) peak, it we attach methylene, this does not exist in DNA, but the signal is so weak that strong priority cannot be given to it. In Galaxy NIR, 933, also correponds to CH and CH2 and ROH all would be possibilities.

I verify the following by inspection of the DNA structure:

Does DNA have carbon hydrogen bonds? Does DNA contain a methylene functional group?

Yes, DNA does have carbon-hydrogen (C-H) bonds. These bonds are present in the sugar-phosphate backbone of the DNA molecule.

DNA does not contain a methylene functional group (-CH2-). The backbone of DNA is composed of a sugar-phosphate chain, with the sugar being deoxyribose in the case of DNA.

This also leaves the ROH group as a candidate. We have already determined that DNA contains "several hydroxyl groups (OH) attached to the carbon atoms. This is therefore consistent with DNA presence."

1429 (minor valley)"

"From TSP 070 study: 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

Now we can agree that both primary amides and aromatic amines are not in DNA, so this is true and conforms to DNA existence.

1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)"

This result is exactly the same as for TSP 070. We know now that DNA has secondary amides in it but not primary. We also know that DNA has no aromatics in it. So this LACK of absorbance remains consistent with DNA existence or proposal.

1533 (strong)

From Dec 18 notes on TSP 070 again: "1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530) 1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530) [alkyne is not consistent with DNA so to be dismissed]

AND QUITE THE SURPRISE HERE:

Does DNA have Amides?

No, DNA does not contain amides. DNA is composed of nucleotides, which consist of a sugar molecule (deoxyribose), a phosphate group, and a nitrogenous base (adenine, thymine, cytosine, or guanine). Amides, on the other hand, are a type of organic functional group that contain a carbonyl group (C=O) bonded to a nitrogen atom.

NOW LOOK AT THIS RESPONSE FROM YESTERDAY:

Does DNA contain Amides or amines?

DNA contains both amides and amines. The backbone of DNA is made up of phosphate groups, which are connected to the sugar molecules through phosphodiester bonds. The sugar molecules in DNA contain amide functional groups. Additionally, the nitrogenous bases in DNA, adenine (A), cytosine (C), guanine (G), and thymine (T), contain amine functional groups. These amine groups are involved in hydrogen bonding and base pairing, which are important for the structure and function of DNA.

THIS IS INSANE. COMPLETELY CONTRADICTORY INFORMATION DEPENDING ON HOW THE SAME QUESTION. THIS TELLS YOU TO BE VERY CAREFUL AND DO NOT ASSUME AICHATS ARE VALID. VERY VERY DANGEROUS. CROSS CHECKING IS REQUIRED AT ALL TIMES. You are going to have to analyze this by yourself with Chat acting only as an assist, and not exactly the most competent one.

So my conclusion is that DNA has both amines (primary and secondary) and amides and hydroxyl groups in it. So take that Mr Chat. But, at the same time, DNA does not have aromatic amines it (cyclic but not aromatic). I see two primary amides in DNA but no secondary amides.

So now we start studying the structure of DNA independently.

1. We see that the bases are clearly heterocylic.

- 2. They are all also cycloakenes.
- 3. They are all also secondary amines. (this is now a very important distinction to start making).
- 4. Three out of four of the bases are also secondary amides. One of the bases does not contain an amide.
- 5. None of them are aromatics.

This is now very important information to include and discern in our structural analysis we make. Go back to the beginning."

So yesterday was quite a round. Independent study of DNA shows that it contains both secondary amines and secondary amides.

This satisifies therefore:

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530)

The UV shift from 266 nm to 261 nm is one of the main distinctions present. THe fact that the UV spectrum has shifted over to 261, away from both protein and bacteriophage, further supports the DNA argument.

Now as far as the protein issue goes, in this case pure DNA would contain no protein. However, our solution is hardly pure. Our estimate on the DNA 260/280 ratio is 15% DNA. So we know we have protein in there also. So best we can tell this peak is fuilly satisified also as consistent with DNA presence.

1589 (strong)

From Dec 18 Notes: "1585 (stong broad absorbance) OH (1583)(1580)" Now in this case our strongest candidate is still at 1583, which is basically indicative of a vinyl - OH connection, also the same as Dec 17 notes. We cannot really do anyting more here.

1640 (strong) (sharp dropoff after) vinyl (1637) so same as before, vinyl presence. Sharp dropoff corresponds to lack of methy absorption on a large scale.

1535-1640 represents broad strong absorbance. This is dominant region. Both TSP 070 and TSP 070.10 are strong absorbers in this region relative to the remaining samples. These 2 of the 5 samples are actually fairly close to one another.

This is as far as we can currently go with this sample. Onward we go. Now lets look at the pH 4.0 decant. This is TSP 070.10.

In organic chemistry, a vinyl group is a functional group with the formula -CH=CH2.

OK, you have missed and now understand the significance of the methylene group. It is strongly indicative of the vinyl presence as it is at the heart of vinyl structure. Also at the heart of butadiene and styrene and therefore synthetic rubber structure. This is showing its importance when you study the transition of TSP 70 to decant and precipitate forms. It also ties in with the proposed genetically engineered bacteriophage and DNA studies.

Because of this understanding, we are going to back up again. Look at TSP 070 to start.

Here is what we are seeing. The ~930 NIR interpretation can be taken in two different ways for both TSP 070 Bacteriophage candidate) and TSP 070.10 (DNA candidate). CRC gives only methylene as an option. We now know that methylene -CH2 is quite fitting in both cases due to its being a primary component of the vinyl functional group. Both samples also show strong vinyl presence (in fact it dominates beyond amide/amines/protein) so this is a perfectly valid interpretation. Our proposition is a genetically engineered bacteriophage at this point that does contain vinyl functional groups.

Now, only from the Galaxy NIR chart, you have the possibility of a ROH, not mentioned in CRC. But we now know that there are hydroxyl groups within DNA, double or single strand, so this is also acceptable as well. The functional groups remain consistent here, considering the genetically engineered vinyl proposition.

Now we can continue with TSP 70.12. which is the pH 4.0 precipitate.

OK, the general interpretation is as follows. The 930 peak region has disappeared, and in general any activity up to approx 900-1100 nm has disappeared. This 930 region can indeed be accepted as methylene per CRC 2012 as it relates to vinyl existence. There is a minor valley at approx 1438 vs the 1427 region of TSP 070 and TSP 070.10. Keep in mind that TSP 70.12 is the precipitate from the first level decant where the pH rises from 3.5 to 4.0.

now 1427 nm was:

"Now we can agree that both primary amides and aromatic amines are not in DNA, so this is true and conforms to DNA existence.

1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)"

Recall 1428 nm was the LACK OF ABSORBANCE, NOT ABSORBANCE, i.e., negative existence.

Now 1438 nm, also expressing negative existence is:

is also methylene and primary aromatic amines, which certainly matches with the DNA situation.

The primary shift we see is from any strong absorbance in the range of 1525 nm - 1650 nm, as it existedn in both TSP 070 and TSP 070.10,

has now been shifted into 1650-1700.

1650-1700 is dominated by two things. Methyl groups and ArCH. This is therefore a significant change. This is of the styrene nature.

Now the bad news from this is that a very slight increase of pH produces this change. Secondly, it took close to two month before the pH dropped down to 3.5 to provide for this setup to even try raising the pH. This means that it could be a fairly well hidden method of producing a styrene type compound.

Now Ar -CH3 is actually methylbenzene, or tuolene. Not too much of a stretch to get to stryene, which is Ar -C=CH3, which adds a vinyl group onto Ar. Notice we actually do have high absorbance, just not peaking, in the vinyl region also, so both types of compound could easily be considered here.

This is therefore an important change. In essence, it appears that an aromatic polymerized compound is separated off by a small change in pH in a very mature culture where the pH has been allowed to drop even further from \sim 3.8 to \sim 3.5.

This gets us through the first level of decant with TSP 070 for a very mature culture, both first level decant and first level precipitate.

Now we go to TSP 070.11 which is the second level decant that raises the pH from \sim 4 to 6.2. Here is what we have:

TSP 070.11 Decant pH 6.2 UV TSP 070.10 262.5 (260) [Magnitude here is \sim 1.5] Valley @ 247.5 (250) [300 ul] Valley slope has flattened out here and peak at 262.5 also not quite as well defined.

2023.12.23 DNA - Bacteriophage Inquiry

Recalling and continuing.

Recall notes of TSP 70.12, the first level precipitate where pH is raised from 3.5 to 4.0:

"This is therefore an important change. In essence, it appears that an aromatic polymerized compound is separated off by a small change in pH in a very mature culture where the pH has been allowed to drop even further from \sim 3.8 to \sim 3.5.

This gets us through the first level of decant with TSP 070 for a very mature culture, both first level decant and first level precipitate.

Now we go to TSP 070.11 which is the second level decant that raises the pH from \sim 4 to 6.2. Here is what we have:

TSP 070.11 Decant pH 6.2 UV TSP 070.10 262.5 (260) [Magnitude here is \sim 1.5] Valley @ 247.5 (250) [300 ul] Valley slope has flattened out here and peak at 262.5 also not quite as well defined."

The idea of an aromatic polymer is therefore a significant change in the development of the first level precipitate TSP 70.12

Now let's go back to TSP 70.11 Decant pH 6.2:

"In addition, we see that the first division of spectroscopy is taking place at approximately the following areas:

280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)

As far as UV goes, 262.5 is closest to 260 over 270 and 280. 260 is anticipated DNA Range: Now this makes sense in that our previous decants went from \sim 3.8 - 4.5 up to \sim 6.2 so the first level decant was simply bypassed. What we are seeing is that the first level decant (ie. pH from 3.5 to 3.8-4.0) appears to separate a bacteriophage near the 270 range and the second level decant (pH 4.0 up to 6.2). appears to separate out DNA closer to the 260 range.

Now recall what TSP 070 native culture does.

TSP 070 has UV of 266.2. This is closest to 265 which is right in the middle. It is not a strong bias either way, and this suggests it could easily be both bacteriophage at 270 and DNA. Slight bias is toward bacteriophage.

This is where you really need MID IR to get another level of distinction but we shall go through notes carefully and see if this interpretation is correct:

TSP 070 Initial pH 3.5 Native Culture UV 266.2 (mid between DNA and bacteriphage, bias towards bacteriophage)

TSP 070.10 FIrst level decant pH 3.5 to 3.8 - 4.0 UV UV 261.0 (closest to DNA) TSP 070.12 FIrst level precipitate Aromatic polymer indicated ArCH3

TSP 070.11 Second level decant pH 4.,0 to 6.2 UV 262.5 (closest to DNA)

So now the big question is when and under what conditions did we see the 269 peak?

Polyvinyl alcohol had a weak peak there on Nov 21 2023. Meaningless right now. Same on Nov 21 2023. Meaningless with PVA.

Nov 28 2023 Notes referring to work done on Nov 07 2023:

"Do I still have the 269 nm sample available to study? Let's see. I have:

TSP 70.1D YES I HAVE IT HERE. see below.

Nov 07/ 2023 TSP 70.1D (M) 6.20-1107 TSP 70.1 Titrate Decant Titrated 20 day TSP 70 Culture Decanted This is what shows to apparently contain bacteriophages. Big issue here. Absorbance is at 269nm. Matches bacteriophage UV profile extremely well. 700-1100 NIR of same sample shows the alkyl alchol w/no hydrogen bonding at 962nm."

"I DO HAVE IT then. 70.3A is of interest for later as it is where the 259-260 peak shows up. But the 269 nm question looks like it requires replication.

Now this culture is dated Nov 07 2023. So this is when titration was done. THe culture was approx 20 days old. The culture is now approx 40 days old but we have it which is great. Now the difference in appearance of this culture from the original clear decant is that it has a synth polymer layer on top now and the solution is cloudy. So it has changed as we have seen before in the decant versions. It serve as a case of interest to see how the UV spectrum may have changed.

This sample size is the largest of my set and is approx 40 ml. The other residual samples are on the order of a few ml. I will extract from the mid level solution area. I may also be able to get pH information at some point, however, I do not wish to disturb or mix the synth-polymer layer.

Good records were helpful here. Impossible to retrace or recollect from memory.

We have another important advantage now in UV. We can record with confidence from 190 nm to 400 instead of 235 nm to 400 using the quartz cuvettes I now have in place. A much broader and better UV picture.

I have replicated the spectrum. As the plot is difficult to interpret because of the broad flat peak, I have used a differential approach. I have two peaks that at the zero point (first once crosses, 2nd one asymptotic: 210.3 nm

257 nm

The center of the broad flat peak is approx 252 nm so thus far there is no way that it approaches the protein section. If anything we have to wonder again if we are in the 260 region for nucleic acids again."

Therfore what we are seeing is that the case for DNA is stronger in both cases of decants, i.e, TSP 070.10 and TSP 070.12. The bias shifts slightly with the native culture to TSP 70 of 266.2. but still almost an even call. The case for bacteriophage remains rather weak at this point, HOWEVER, THE CASE OF NOV 07 COULD SHIFT THE TIDE AND MAKES THINGS INTERESTING. Let's look at this closer. We are looking for the 269-270 region of activity.

This was a 20 day old culture of TSP 070. That would place it around Nov 10 which matches Nov 07 2023 notes. This was a decant which means the middle layer of the culture was precipitated.

Here is another important note I see on Nov 07. The nucleic/DNA issue actually arose with the use of the SURFACE LAYER of TSP 070, i.e., the developing synth polymer layer! This means that I acidified it with strong H2SO4 to be able to dissolve it for use in the UV spectrum. Watch out for the cuvette when you do that, probably plastic cuvette is best here - take care of the quartz cuvettes!

This means that we have DNA evidence replicating in many different directions now.

Now we made a note on Nov 07 that pH was raised to 6.6. So we have a 20 day old culture, middle layer, TSP 070, raised to pH 6.6. Single stage pH increase presumably from 3.8 (right above the first level rise and decant) to 6.6. Incubation at 98.6 is mentioned as being potentially of value. Also what I am doing here is comparing two different pH increases on the TSP 070 culture, one on Nov 06 and one on Nov 07 2023. So I am comparing two samples that should be similar but one appears to be incubated and the other not. Not sure which is which at this point.

What happens here is that I get strong absorbance at 269-270 nm. I also NOW HAVE A VALLEY AT 258 nm which indicates that the DNA/nucleic acids DO NOT APPEAR TO BE PRESENT.

I also mention a pH of 6.2 in a sample, we may have a range of two samples, with pH from 6.2 - 6.6. This is where we establish a sample codinng system because the sample variation has become quite complex.

What I am seeing is that I have a 20 day old culture from TSP 070. I am using the middle layer, pH presumably

is stated at 3.8. I titrate it, and it looks like I titrate it to 6.2. I run both UV and NIR-VIS on it. Strong peak at 269-270. Valley at 258. I realize that it is quite different from others. NIR-VIS picks up an alkyl alcohol at 962 nm.

I then centrifuge the sample and THIS IS WHERE AND WHEN I see the two layer precipation showing up, one white one grey. This very likely corresponds to the two decants/two precipitates of the current study. I then end by creating a coding system for the samples.

I make comments regarding how variable the cultures are with respect to (wrt) time.

So we end up having a very strong observation set here with decent notes. Now a question is can you duplicate it? You have no 20 day cultures anymore. Our cultures are approx 30 and 60 days old. TSP 100 may be in range to use at approx 30 days vs 20.

Now what we see is that the second level decant, i.e, TSP 070.11 should show a general migration from the DNA range of 260 nm to the bacteriophage range of 270 compared to TSP 070, native culture. What is different is that the pH change is broken into two stages but the end result is much the same.. Let' see if this is true.

TSP 070 UV comes out at: 266.2 on Dec 16 2023 (close to 60 day culture, however). closer to bacteriophage. What was TSP 070 20 days into the culture?

TSP 070.11 comes out at 262.5. (closer to DNA)

Wrong direction, however, far too many variables are active here, especially with significant cuture aging.

We need to back up as much as we can. Start with TSP 100.0. Notice that TSP 100 has NOW FULLY CLARIFIED in the middle layer. We know that this is a significant stage in maturation. Let's look at the pH.

The pH is 3.5 so it is fully down to the lower level level now.

Centrifugation has only produced the grey layer. No white layer. Peak is approx. 258. This points toward DNA presence, not bacteriophage.

Everything says that we would need to start a new culture to determine if the bacteriophage prospect is genuine or not.

I see nothing more that I can do with this now.

2023.12.24 Video Up - Regroup - Phage

The video summary of Human Blood vs Synthetic Blood : The Path to the Blood Clot is complete. Five minute summary. The video is receiving fairly high traffic which appears to be coming independent of the web sites. There is no feedback system unless viewers are logged in which is not going to happen. This paper and accompanying video are a watershed in the research, as it clearly ties the blood clotting increase and "Morgellons" together under a common source.

I think what you need to do is itemize the differences between the various decants and precipitates. What you have to work with and what is unknown. You are seeking to get the IR spectrometer, overseas communication.

The bacteriophage may well require that you need to start another culture. You now have two completely separated cultures with 3 layers each now; the separation process takes about a month. However, before you start another culture, realize that you have:

1. Three distinct layers of culture to separate.

 The mid layer alone has two decants and two precipitates as a minimum, assuming the pH gets down to 3.5. Including the native culture, this means you have a minimum of five variations to analyze and size up.
 We notice that the pH 6.2 decant, EVEN AFTER CENTRIFUGE, is still cloudy and not transparent. This means that another separation is likely needed, meaning another PI. This might add another layer of two more variations.

4. Not difficult to see up to 10 separations just within our immediate culture.

5. LC Chromatography has many potential avenues for further separation here.

6. You are very interested in using electrochemistry for organic reactions; it looks to be a leading research topic.

7. One culture looks to be completely destroyed by the L. enzyme.

8. Remember that we have the detection of an electromagnetic field with the culture. A very big deal also.

The work in place thus far does represent a milestone in the research. The common ground between increased clotting apparently correlated with the "Covid Era", and the CDB and "Morgellons" is now established with clarity. This relationship is unknown to the vast majority of the population.

I have another "disclosure" project planned over the winter season. It is delayed through illness on part of the coordination. The project wil remain confidential until appropriate to release. The topics to be covered are the legal considerations in both culture work and mitigation prospects.

You hope to have a MID IR instrument, although time for its use will be fairly minimal.

Main objectives for this unique winter season with access to a minimal lab is:

1. Determination of origin and nature of the increased clotting taking place since the Covid Era transpired. (done)

2. Determine what, if anything, can be done to reduce or eliminate the damage in place (partial).

3. It would be beneficial to determine WHY increased clotting appears to result from the Covid Era (i.e., "vaccinations") but this seems difficult to approach. You need proper samples for this and you have none. You need access to information on the purported "vaccines" and you have little to none.

You have also shown that the CDB exist within dental anesthesia but the work was not distributed. It is not likely that it will ever be.

It does not seem reasonable that you can or should take on the vaccine cause/effect problem without having access to adequate samples.

Clearly it is always of benefit to have the chemistry of the culture products more clearly defined. This always take a great deal of work, time and effort. MID IR is the best tool if you are able to bring it into the methods. For now you have to work with what you have, and this happens to be rather minimal.

Strongest tools here now are likely LC and NIR, but all methods you have used are helpful and valuable, well over a dozen techniques.

It seems like a good reference point right now is to collect UV and VIS NIR data on all samples available from current cultures, SOLID and liquid. Dissolve if solid, consider plastic cuvettes for acidic samples.

"In addition, we see that the first division of spectroscopy is taking place at approximately the following areas:

280 > protein (270-280 region) 270 > bacteriophage (269 peak and 245 valley) 260 > DNA (260/280 ratio)"

There are actually several stages you can go through. UV NIR LC on each sample type/phase. Then NIR. Step through TSP 070 and TSP 100. I will use 300 ul as the default on each UV run.

TSP 070 Middle Layer:

Peak 267 (270) Strong Valley 244 (245, 240) Strong, well defined Peak 216 (220) Strong, Magnitude ~3.0

Now what is interesting here is that you are actually already in the bacteriophage range closer than anything else. This is the closest match of the three main groups. Notice also you have the valley correpondence. So are immediately in range within one of the major questions before you now with the very first sample tested. You also already have a decent NIR plot for this sample.

UV spectrum of T7 bacteriophage at:

https://www.researchgate.net/figure/Absorption-spectrum-of-bacteriophage-T7-dark-sample-before-full-line-and-after-dark-sample-before

"Phage T7 is a member of the Podoviridae family"

and is therefore NOT a match for the filamentous bacteriia. T7 bacteriophages kil the host, filamentous bacteriophages do not. So only regard this phage as a general reference; it is not intended to be a match for filamentous phage, which are in the family

"While most phages are pathogens

that kill their bacterial hosts, the filamentous phages of the subclass Inoviridae (INOVIRIDAE)! live in cooperative relationships with their bacterial hosts, akin to the principal behaviours found in the modernday sharing economy: peer-to-peer support, to offset any burden.

Filamentous phages impose very little burden on bacteria and offset this by providing service to help build better biofilms, or provision of toxins and other factors that increase virulence, or modified behaviours that provide novel motile activity to their bacterial hosts"

So ultimately we would need to find a UV spectrum for INOVIRIDAE but below is at least a general start, and the general UV profile is helpful.

Notice how similar our profile is. Notice however, that the spectrum image shows a continuous rise up through the range limit of 200 nm.

Bun notice in our plot we have a peak at 216 nm. This particular spectrum has the peak at approx. 262 nm (closer to DNA), the valley is at approx. 237 nm and then a rise up through 200 nm. General profile is quite similar.

We actually have a rather strong case for bacteriophage fairly early in the investigation. I have found a UV spectrum and numerous references to the 269 peak and one to the 245 valley, as well as the general profile match.

https://www.researchgate.net figure

Spectrum-of-a-10-fold-diluted-M13-phage-after-ultracentrifugation-The-virus-has-a-UV_fig2_319313582 Figure 2. Spectrum of a 10-fold diluted M13 phage after...

The virus has a UV absorbance spectrum with a maximum at 269 nm and a minimum at 245 nm. The purity is assessed by two ratios: max to min absorbance and baseline to max absorbance....

www.abdesignlabs.com technical-resources bacteriophage-spectrophotometry

Quantification of Bacteriophage by Spectrophotometry

Here is a UV absorption spectrum of filamentous phage purified by PEG-precipitation and dissolved in TBS; the spectrum typically exhibits a shallow maximum around 269 nm: The relationship between virion number and absorption is given by the following formula: This formula was established by

https://www.ncbi.nlm.nih.gov pmc articles PMC8588016

Modification of a Tumor-Targeting Bacteriophage for Potential ... The filamentous bacteriophage, ... The peak at 269 nm corresponds to the absorption of M13 bacteriophages . The spectra of the control samples, in contrast to the experimental ones, did not have an absorption peak at 494 nm, which indicates the modification of the bacteriophage shell with the

https://www.ncbi.nlm.nih.gov pmc articles PMC7596064 Improvements in the production of purified M13 bacteriophage bio ...

The UV-Vis quantification of DNA and proteins is calculated using the absorption at 260 and 280 nm, respectively. Therefore, the presence of a peak at 269 nm and a higher absorption in the region below 240 nm in the UV-Vis spectrum of M13 (Fig. 2) is in correspondence with the previous reports 49.

https://www.researchgate.net/figure/Spectrum-of-a-10-fold-diluted-M13-phage-after-ultracentrifugation-The-virus-has-a

and most importantly, M13 IS a filamentous bacteriophage. It does infect E coli so it is an archetype.

https://www.nature.com articles s41598-020-75205-3 Improvements in the production of purified M13 bacteriophage bio ... M13 is a filamentous virus which infects bacteria (bacteriophage) and in particular, the Escherichia coli (E. coli) strains showing the F- pilus 1. It measures approximately 1 µm in length and...

So this is a pretty big deal here, as it comes from the native mature culture. Absorbance is roughly a factor of 5 to 1 over absorbance at ~300 nm which is a model used for concentration estimate of virions.

Now let's pick up the NIR-VIS section on the desktop spectrometer.

We have 962 nm weak but definite and detectable. This, as we have seen before is attached to

962 (960) Alkyl alcohol with no hydrogen bonding in CC4 (962). R-C-OH. No competition. Galaxy PDF has candidates as:

H2O Eliminated by control test. ROH ArOH

Our point of overlap is ROH. Water has been established as the blank so it should not be water. Test this with a water sample.

I have run the water control test and it comes out fine. Water presence is completely subtracted properly from the spectrum, no absorbance at 962 nm.

This says that we conceivably have the choices of alkyl alcohol, ROH and ArOH. In all cases see that they are alcohols. Use NIR to try and discern further .

Now from Dec 18 Notes:

NIR TSP 070 Native 933 (moderate peak) Methylene (930)

1530 (strong peak) Amide/Protein (1530) Secondary Amine (1530) Alkyne (1530) 1585 (stong broad absorbance) OH (1583)(1580) 1640 (strong absorbance, drops sharply after this pont) VInyl (1637) After this point, signal is dominated by Methyl and ArCH.

Also broad strong absorbance between 1535-1640. This is dominated by amides, polyamides, and vinyl

groups.

Therefore, amides, polyamides, proteins(? uncertain here see AI chat below) and vinyl groups seem to characterize the middle layer of TSP 070 60 day culture.

I see there is another aspect we can use to try and discern between the functional groups, and this is the valleys. Lack of absorbance can be valuable as well. So in this case let's also give notice to:

1086 (moderate valley) LOWER TO NO ABSORBANCE No CRC 2012 entry. 1428 (moderate valley) LOWER TO NO ABSORBANCE Primary Amides (1430) Aromatic Amine (1432)

Notice that DNA does not actually have aromatic amines from what a see. Cyclic, yes, but not aromatic.

Now, one way you can approach the NIR data is to ask if ANY strong signal conflicts with a determination of bacteriophage.

The one exception we are willing to make is that this will require a genetically engineered bacteriophage to include the existence of vinyl. But this is not at all impossible as the AI chat expressed. Also keep; in mind the desire to discern between ROH and ArOH.

Now at 1530 nm we know that filam. phages have SS DNA and amides and amines are perfectly valid. Protein is also valid because of the capsin.

Now at 1585 we know that DNA does have hydroxyl groups. UV and VIS NIR also say alcohol, but they have alkyl alcohol or ArOH.

One thing that every one agrees upon is the existence of a hydroxyl group and this does not conflict with filam. phage DNA.

And our last group is a strong signal on vinyl with 1640 (1637).

This will require a genetically engineered phage or the presence of vinyl compounds in solution in conjunction with a phage. These are both realistic possibilities.

The NISt UV website is down. Inconvenient as always. We have a very useful answer from AI:

Where does the vinyl functional group absorb in the ultraviolet spectrum?

The vinyl functional group typically absorbs in the ultraviolet spectrum at wavelengths around 200-230 nm.

We therefore have a perfect match here with our spectrum with absorbance at 216 nm, dead center in the region specified.

All signs are therefore confirming of the existence of a filam phage accompanied with vinyl presence, either within a genetically engineered phage or in accompanying solution.

The case is now rather strong. This is the native culture before any pH separations are completed. Quite remarkable.

Are there any other ways that the existence of a filamentous phage might be determined?



Spectrum of a 10-fold diluted M13 phage after ultracentrifugation. The virus has a UV absorbance spectrum with a maximum at 269 nm and a minimum at 245 nm. The purity is assessed by two ratios: max to min absorbance and baseline to max absorbance. Here, the 269/245 ratio is 1.37 and the 350/269 ratio is 0.02, all indicating that the phages are viable and pure. Pure sample corresponds to 16 mg.



Absorption spectrum of bacteriophage T7 dark sample before (full line) and after (dashed line) 2 years long space fl ight.

2023.12.25 Christmas - On We Go

We are at the point of having a strong case for filam. phage existence using TSP 070 in its native form, 60 days age. Now to have this as observed, one or both of two cases would seem to be able to account for the existence of the vinyl groups:

1. The phage has been genetically engineered to contain or produce vinyl groups (quite feasible). 2. Vinyl groups exist in solution along with a phage. (also feasible).

This means that it is worthwhile to see if a vinyl component can be isolated more strongly from the solution. This could be either through a decant, a precipitate, or a centrifgue, we shall work on each.

One question is what is the UV absorbance of vinyl? We have a chat response of 200-230 nm so we have a perfect match there. Let's look at the two decants first since we have them at hand, pH 4.0 and pH 6.2.

Here is our original information from yesterday Dec 24 2023 on TSP 070 native culture:

TSP 070 Middle Layer:

Peak 267 (270) Strong Valley 244 (245, 240) Strong, well defined Peak 216 (220) Strong, Magnitude ~3.0

and here is what we get with the first level decant:

Starting with TSP 70.10 which is the decant of pH 4.0 and 300 ul.

216 (220) Strong. Magnitude 2.4. Valley 245 (240, 250) Less well defined but still a strong feature in the profile 265 (260, 270) Strong. We therefore see a very slight shift toward the left.

Therefore it is highly similar to the native culture but some differences are starting to show. It would obviously be of interest to see the precipitate that forms from the pH rise from 3.5 to 4.0. Difficult to collect enough material? We will see.

But while we have the sample, let's pick up the VIS-NIR segment. Extremely weak (mag. .008) but we stil have a defined peak at 962 nm. 962 nm (962) aklyl alcohol, ROH, and ROH are all possibilities under combined CRC or Galaxy Scientific. What is common to all is the hydroxyl group.

Therefore the decant shows some slight modifications but nothing extreme. I would like to know if the vinyl aspect can be separated. Ideas include:

1. Studying the precipitate that forms from the first level decant. (Use our existing comprehensive DPLOT NIR spectrums.

2. LC separation possible?

NIR is always first.

NIST is operating again. Both NIST and SDBS will allow for wildcard searches, very helpful. What is the lowest molecular weight vinyl compound? Where does it absorb UV? How much does this absorbance vary with respect to type of vinyl compound?

The NIR set you have of all 5 TSP samples simultaneous on one plot will help you to identify the differences you are looking for and any separation of the vinyl compounds between the decants or the precipitates.

divinyl ether on NIST has a broad UV peak at 202.5, C4H6O. Probably a good representative compound to use. Strong absorbance from 195 to 205 nm.

What we are looking for is whether or not the vinyl groups ever show separation from the TSP 070 mature culture and at what point if so. This means we are looking for differences from native, decants, precipitates, age of culture, etc..

Here are our notes from Dec 19 2023 which gives us the type of information that we are seeking: This is for TSP 70.12, the first precipitate.

"The primary shift we see is from any strong absorbance in the range of 1525 nm - 1650 nm, as it existed in both TSP 070 and TSP 070.10,

has now been shifted into 1650-1700.

1650-1700 is dominated by two things. Methyl groups and ArCH. This is therefore a significant change. This is of the styrene nature.

Now the bad news from this is that a very slight increase of pH produces this change. Secondly, it took close to two month before the pH dropped down to 3.5 to provide for this setup to even try raising the pH. This means that it could be a fairly well hidden method of producing a styrene type compound.

Now Ar -CH3 is actually methylbenzene, or tuolene. Not too much of a stretch to get to stryene, which is Ar -C=CH3, which adds a vinyl group onto Ar. Notice we actually do have high absorbance, just not peaking, in the vinyl region also, so both types of compound could easily be considered here.

This is therefore an important change. In essence, it appears that an aromatic polymerized compound is separated off by a small change in pH in a very mature culture where the pH has been allowed to drop even further from \sim 3.8 to \sim 3.5.

Notice the pH rise from 3.5 to 4.0 is directly responsible for inducing this polymerization change. 1650-1700 absorbance IS NOT high within TSP 070 native. In fact, it drops off very sharply toward zero. However, 70.12 and 70.14 are both with very strong absorbance in this range. This says the polymerization is taking place BECAUSE of the pH rise.

2023.12.26 Five TSP Separations

Now reviewing what we are learning yesterday from the UV and NIR analysis of the five TSP 070 variations, native, decant and precipitate.

What we see is that the rise of pH in general, through the stages of 3.5 to 4.0 as well as continuing from 4.0 to 6.2 appears to have the effect of creating an aromatic polymer which appears as a precipitate. This precipitate would seem to have a polymer styrene character, i.e., polystyrene is a target candidate.

Now the the decant, even after its final pH rise up to 6.2, still shows the vinyl content as actually a remaining primary component. The proteins and amides still seem to exist, possibly slightly reduced in magnitude from the native culture, but with a stronger shift into the vinyl region even after this final decant taking place.

Now as far as UV goes, in the native culture to begin with we have a rather strong case for bacteriophage,protein, amide, amine. hydroxyl, appropropriate absorbance, with a vinyl component that is uncertain whether in solution or within a proposed genetically engineered phage.

The first level decant of pH 4.0 changes this profile some, but not in a dramatic way.

At this point I am curious:

1. How decant at pH 6.2 is behaving with respect to UV (protein, dna,phage? - vinyl, aromatic?) NIR seems to say protein/amides in existence, and vinyl.

2. Can I make a determination of whether the vinyl group is within or only external to the phage prospect?

Here is what we have on UV with TSP 70.11 (pH 6.2):

First off, there does appear to be a layer of polymerization on the surface; this sample has been sitting idle for a while.

Second the solution is actually more cloudy than TSP 70.10 (pH 4.0). This tells me that there is still more going on within this solution, and that another decant is likely possible. We still would like to recover the two layer precipitate form. As far as UV goes, we have:

266 (peak) (strong but broad and not well defined) (270)254 valley (very broad and not well defined)219 peak (very strong, magnitude at 2.1, very well defined) vinyl indication

In terms of the vinyl component, this matches very well what the NIR plot shows. NIR shows a very strong vinyl aspect to this second level decant at pH 6.2. It actually appears to be the most dominant expression of the vinyl existence over the five samples. This also concurs with the visible behavior of a polymer layer forming on the surface as well as remaining (actually becoming more cloudy as we noticed before) cloudy in solution. This may be the most effective means of separation of the vinyl component. It also suggests that the vinyl component is actually a separate affair from the bacteriophage prospect since the bacteriophage UV spectrum is not as well coincident here.

There is another thing happening with NIR There is a shift from the native cultures of absorbance in the 1530 nm region (associated with amides/proteins (.NH or .NH2) (as we study this topic now, this should mean primary or secondary amides)

or secondary amnes to (within TSP 70.11) approx. 1498 +/-. 1498 (approx.) NH primary aromatic amine (1496.5) alcohol, water (1500) amide (.NH or .NH2)(1500) alkyl alcohol containing one OH(1500)

Because of the non-coincident repetion of the amide(.NH or .NH2) this group is LESS LIKELY. This means our choices our more prone to an alkyl alcohol or and aromatic amine. Let's now immediately look at VIS NIR at 962 nm to examine the alkyl alcohol prospect:

The 962 nm VIS-NIR peak is essentially non existent. A minor variation can be detected, but it is not defined well enough to establish as a group. This may now cast the vote stronger towards an amide or an aromatic amine. Let's look at the Galaxy PDF. Galaxy PDF give us either CONHR (an amide) or RNH2 (amine).

This cast our weight toward the amide side with both NIR and UV leading this direction.

"Amides are qualified as primary, secondary, and tertiary according to whether the amine subgroup has the form -NH2, -NHR, or -NRR', where R and R' are groups other than hydrogen.[5]"

This is leading to an estimation of TSP 70.11 as dominated by vinyl and amide groups.

I obviously need to learn to distinguish properly between primary, secondary, tertiary amides. I believe I have the amines under wrap now.

OK, the rationale for primary secondary tertiary amines and amides is actually just the same. It is all about how many carbons are attached to the nitrogen of the amine, it has nothing to do with the "number of" carbonyl groups attached. Only one carbonyl group is required to make a primary, secondary or tertiary amide. ONLY ONE IS NEEDED.

So an amine is either NH NH2 or NR' R'' R'''

Primary amines have 1 carbon attached, this means it is NH2. If 2 carbon atom atoms are attached to the nitrogen it is a seconary amine REMEMBER, IT REALLY HAS NOTHING TO DO WITH THE NUMBER OF HYDROGEN ATOME, THE NUMBER OF CARBON ATOMS ATTACHED TO THE NITROGEN IS WHAT MATTERS. If 3 carbon atoms are attached to the nitrogen it is a tertiary amine.

IF THERE ARE NO CARBON ATOMS ATTACHED IT IS NOT AN AMINE! IT IS A SPECIFIC COMPOUND, NAMELY AMMONIA!

Now to understand primary, secondary, tertiary amides, thing about what you start with. You have a nitrogen that has a carbonyl group attached, by definition, to make an amine become an amide. So one of the carbon bonds is already taken up as attached to the nitrogen, and that makes it a primary amide.

Now if you replace one of the two remaining hydrogens attached to the nitrogen with a second carbon based group, you have a secondary amide. And likewise the third hydrogen with a third carbon you have a tertiary amide. This is important, and one way you might be able to distinguish DNA from a protein, for example. Actually very cool to be able to distinguish structure at this level.

https://onlinelibrary.wiley.com doi abs 10.1002 9780470015902.a0029482 Filamentous Bacteriophages: Biology and Applications Sep 27, 2022Filamentous phage diversity and range were vastly underestimated until 2019, when bioinformatic analyses of prokaryotic genomes and shot-gun metagenomes discovered 10,295 novel filamentous phage-derived prophages in nearly all bacterial phyla and in some archaea. ... Filamentous

At least a half dozen references to filamentous phage absorbance at approx 269 (265 also mentioned) nm: https://duckduckgo.com/?hps=1&q=filamentous+bacteriophage+269+nm+uv&atb=v394-1&ia=web

I am after whether the vinyl contribution can be separated from the prospective bacteriophage. TSP 070 native is probably our best bet to start with and centrifuge.

I have run a control UV trial on a segment of vinyl glove in strong sulfuric acid. I have a extremely sharp strong peak at 196 nm.

I have a moderate peak at 230 nm and a broad fairly weak peak at 310 nm. This is helpful. I used 100 ul of the sample in 3 ml cuvette to try to keep the solution weak as to cause no damage to the quartz cuvette. No damage apparent; time of exposure kept to minimum. Signal is more than sufficiently strong (0.55 absorbance at the 196 nm peak).

When we centrifuge TSP 070 native culture, the solution remains completely transparent. There is NO precipitate that settles on the bottom. So the precipitate forms only as a function of pH increase.

When we make a UV run on the top portion of this transparent solution we get:

- 268.2 well defined peak (strong)(bacteriophage remains justified)
- 243.2 (valley) (very well defined)(bacteriophage justified)
- 222.2 well defined strong peak (vinyl absorption justified)

At this point bacteriophage and vinyl groups are justified. We know that any vinyl group is soluble here and cannot be centrifuged. Will LC separate anything here?





2023.12.27 Amines Amides Vinyl

Additional notes on primary, secondary... in the case of BOTH amines and amides, what matters is the number of carbon R groups attached to the nitrogen. It does not change anything in nomenclature regardless if it is amine or amide. What distinguishes the amide is that it has a carbonyl group attached. If this is all that is happening, then there would be two hydrogens left on the nitrogen. Because only one carbon is attached to the nitrogen it would then be a primary aminde. This would then be called an .NH2 amide. Take off another hydrogen and replace with another carbon R', and it would become a secondary amide with name .NH amide. If you replaced the last hydrogen with another carbon R'', then you would have a tertiary amide (I suppose it would need to be called a .N amide).

The same principles apply to amines also, same idea.1,2,3 carbons attached to the nitrogen atom means primary, secondary, tertiary amines respectively.

Does An amine functional group require a hydrogen atom?

No, an amine functional group does not require a hydrogen atom. An amine functional group consists of a nitrogen atom bonded to one or more carbon atoms. The nitrogen atom can have one or two additional hydrogen atoms bonded to it, but it is not necessary for the presence of a hydrogen atom in order for a compound to be classified as an amine.

Notice that if an amide exists, then an amine group exists within it by definition, since an amide is an amine with a carbonyl group attached.

Now if you start looking at DNA more closely, you can step through each base form and determine what form of amine and amide would exist within each base, and then they can be tallied up for a general assessment of DNA structure.

On the first base (C) we see that it has:

- 1. a primary amine (i.e, .NH2 (lots of hydrogens left, only one used, so it is primary)
- 2. a secondary amine
- 3. a teriary amine
- 4. a secondary amide (has two carbons attached in ring, the ring is both cycloalkene and heterocyclic)
- 5. a tertiary amide

On the second base (G) we see:

- 1. a secondary amine
- 2. a primary amine
- 3. a tertiary amine
- 4. a secondary amide

On the third base (A) we have:

- 1. primary amine
- 2. secondary amine
- 3. tertiary amine
- 4. NO AMIDE

On the fourth base (T) we have:

- 1. a secondary amine
- 2. a tertiary amine
- 3. a secondary amide
- 4. a tertiary amide

Therefore, in summary, being looked at for the first time in detail, DNA would appear to have:

- 1. primary, secondary, and tertiary amines
- 2. secondary and tertiary amides.

This says NO primary amides exist in DNA, which would require an amide group (N with carbonyl) with only one carbon attached, i.e., the carbonyl group). (This would be the .NH2 form, therefore) It would would require 0=C-NH2 to exist within DNA, and it does not.

This therefore might become a discerning factor in IR analysis in the future. Keep this in mind.

Н

A primary amine has the form: N-C OR 1 Carbon attached to Nitrogen OR .NH2

H C I A secondary amine has the form N-C OR 2 Carbons attached to Nitrogen OR .NH I H C I A tertiary amine has the form N-C OR 3 Carbons attached to Nitrogen OR .N

С

H

So a primary amide has the form : O=C-N-H OR 1 Carbon(THAT INCLUDES THE carbonyl group) attached to Nitrogen OR O=C-NH2

С

A secondary amide has form : 0=C-N-H OR 2 Carbons(THAT INCLUDES THE carbonyl group) attached to Nitrogen OR O=C-NCH

A tertiary aminde has form: O=C-N OR 3 Carbons(THAT INCLUDES THE carbonyl group) attached to Nitrogen OR O=C-NC2

c

С

It is actually quite a bit involved to sort this out and it is understandable why it is not immediately visualized. Notation in the form of .NH2 in relation to amides is not particularly descriptive but you can back it out now with the above study.

This was very necessary to work through and will be helpful as discerning factors in the future.

Now the actual question at hand right now is whether or not the vinyl group can be separated. So far it seems like it is carrying through no matter what form you seem to be in, decant, pH changes, precip, etc. You are trying to determine if the vinyl group is within the bacteriophage prospect or whether it can be separated.

I am going to make an LC run on the native culture TSP 070. I am changing the refractrometer indicator over to electrical conductivity. I think that it will be a more sensitive indicator and quicker and easier to use vs. drop preparation. My indicators are now, therefore, pH, EC (electrical conductivity) and TDS. It has been two months since I have done this so I wll need to reestablish my protocols. Volume is used over drop count.

What we see is electrical conductivity is so sensitive that if the elute reaches a steady state condition, EC monitoring alone is almost certainly sufficient to detect a change so we can use only EC for the steady state portion. This saves a lot of likely unfruitful effort.

From a protocol and technical standpoint I had a very clean LC run. Easily detected the analyte of TSP 070. Vlal #5 is the clear winner in concentration level. UV runs were also insightful and shows the effect of concentration. The vinyl groups are especially subject to variation in UV with a range of 195 nm (low concentration) to a high of 216 nm (high concentration).

You have a UV profile to previous runs with TSP 070, using vial #5, peaks at 216 nm, valley at 245 nm, peak at 265 nm. Continues to raise the bacteriophage prospect.

The next LC run is going to use acetone, a more polar solvent. Be careful of plugging the column by creating

an insoluble precipitate via pH. Continue use of native solution. I will save vial #5 solution approx. 10 ml. for now.

Acetone absorbs UV apparently at 329 nm so this is good as it is outside normal range of consideration thus far.

LC runs should have a trial test tube run prior to assess expected behavior in a column, including solubility, miscibility and precipitation.

For example, TSP 070 middle layer native + acetone may be creating three layers, if not even four.

2023.12.28 TSP 070 Separations LC DNA

I am in the middle of a LC run with TSP 070 native culture using acetone. The primary question is whether a vinyl component can be separated from the TSP 070 mature culture address the uncertainty of a genetically engineered bacteriophage (prospect).

In the process of doing so after tubes settling overnight, I see two interesting events.

1. Vial "No. 5", which was a result of a water eluent, has now been sitting for a couple of days. It unexpectedly has a precipitate (minor amount) at the bottom of the tube. This is somewhat unexpected because it was the decant from a centrifuged TSP 070 middle layer to begin with. Vial No. 5 was grand central station of that particular LC run. So either some type of precipitate was formed from unknown dynamics of the LC run or it collected material that may have been residue in the tube (i.e., even sand...?). Might be best to look at it under the scope.

2. Now even more interesting to me is a test tube trial mixing acetone with TSP 070 centrifuged decant. This was, and should be done on a regular basis, as preliminary to a LC run to assess potential behavior in the LC column. Now what happened here at the time of mixing is that 2-3 layers were visible as forming. Now that it has settled overnight we large amount of precipitate that has formed and settled on the bottom of the test tube. The remaining solution is quite transparent with a slight yellow tint. This looks to be quite significant and induced by the acetone specifically. I will most certainly come back to this - it is a vinyl candidate.

Remember that our electrical field detection on the TSP 070 culture is a high priority research topic.

In addition, there is indeed work going on with electrochemical detection of organic compounds. This could offer a much potential in the future, but it could be years before protocols are standardized. But very worthy of investigation in the interim, for example, does a compound like polyvinyl alchohol have an elecrochemical signature? Many papers on the subject now exist:

https://duckduckgo.com/?q=detection+of+organic+compounds+with+electrochemistry&atb=v394-1&ia=web

Reagents are now expensive even for LC work, so electrochemistry has many advantages. NIR also of course. Think about how effective EC is already showing itself to be with the detection of change in the LC column. It is very sensitive as a detector.

Now let's continue with the LC run, recorded on a spreadsheet. Indicators of change are electrical conductivity (EC), TDS, and pH. I am on vial no. 16 out of 25.

Looking at the LC acetone run results, it appears that I may have two components to investigate.

Vial EC TDS pH 8 >20.00 9270 4.60 Grand central station here. Amber tint.

and

15 0.40 175 6.59 17 0.41 174 6.96

Now looking at vial 8 is somewhat interesting as this is a definite very strong separation in place. What we have here is:

218 nm strong peak well defined (attributed to vinyl thus far).240 nm valley well defined (has shifted to here from 245)265 peak well defined

So notice that this is essentially a shift from the 269 listed commonly for the filam. phage. However, this has also been stated to be down to 265. The valley 245 peak has only been found in one paper thus far. These shifts are therefore not out of range. This solution, however, is thought to be relatively "pure" so it of interest that the vinyl group still appears quite strong.

You are getting a VERY strong readout in the literature on this topic:

"https://pubs.acs.org doi 10.1021 acsphotonics.2c00041 Bright Future of Deep-Ultraviolet Photonics: Emerging UVC Chip-Scale ... The germicidal effectiveness peaks at ~260-265 nm, which also corresponds to the peak of UV absorption for bacterial DNA/RNA (Figure 1a). (1,2) Thus, technically any UV source emitting in the 260-265 nm band in

principle will be more effective for disinfection."

Looks to me like you hit a real hotspot in the research here, right in conjunction with filam. bacterial phages.

Looking at our acetone solubility test tube results with the TSP 070 and the precipitate formed will be of much interest.

Now let's go to vials 15 and 17.

We have a rather phenomenal event here with vial 15. Vials 15-17 and even beyond are very stable in the vitals. What has happened is I now have very strong isolated absorbance at 265 nm. A strong well defined large peak. Acetone absorbs at 329 nm from literature and control test with acetone shows no conflict whatsoever. What the data shows is a strong isolation of bacterial DNA. The literature supports this strongly.

The bacteriophage is not a requirement at this time, but still remains a definitel possibility, if not likellihood.

The absorbance at 265 is a perfect overlap in frequency and magnitude with the TSP 070 LC sample in Vial No. 5. (500 ul of Vial 15 was used, 300 ul of Vial No. 5).

Another significant event has taken place. The broad strong absorption in the 215 nm region of vinyl is now removed. There is only a small narrow relative low magnitude peak at approx 196 nm. This is saying that the vinyl component has been separated out from Vial 15 and that VIal 15 is rather relatively purified bacterial DNA. The 260/280 ratio is actually close to 2 to 1, quite phenomenal for purity standards.

One proposal here is that if you were to chill a vial and alcohol, you would think that DNA might be visibly produced. I will chill the alcohol and vial 15 contents for later study.

This is saying that TSP 070 middle layer is surmised to have primary components as protein, DNA, vinyl. Need to check on the alkyl alcohol also at what level and stage it is showing up.

Now let's start looking at the precipitate settled on the tube with TSP 070 centrifuged clear + acetone.

The first thing done here is to look at the clear solution which remains after settling over night and forms the precipitate. The UV profile is completely different than anything seen before. It is changing the nature of TSP 070 in a very significant way. What we have is:

> 330 nm to 400 nm : low absorbance, no significant activity

An extremely sharp rise to approx 296 nm and then a sharp flattening of absorbance as we decrease to approx 275 nm.

Then we have a gradual rise in absorbance to a distinct peak that develops at approx. 240 nm.

Then a sharp decrease up to and including the 190 nm range limit.

Quite different than anything involving TSP 070 before.

The first things we observe with the precipitate is that:

1. It is white

2. Rather dense, as it settles quickly in solution

3. Vinyl is certainly a candidate here.

I am preparing a NIR card. This material should be easy to produce in relative volume.

What we learn here is that acetone INDUCES polymerization of the TSP. Acetone is produced by the human body.

2023.12.29 TSP 070 Acetone Study

The TSP 070 acetone trial is producing some surprising results.

1. First, we do have a significant precipitate being formed. NIR plot is uncertain at this time; I wish to concentrate the sample further on the NIR card.

2. The UV of an LC run that produces two separate components has an interesting result. The first elute has the classic signs of bacterial DNA abosorbance at approx 265 nm, the strong valley approx 244 nm, and the strong vinyl component at approx 218.

3. However, the second elute detected in the LC run shows very strong isolated DNA absorbance also at 265 nm but eliminates 90% of the vinyl signal area.

4. In addition crystal growth takes place on the surface of the decant within 24 hrs of TSP 070 -acetone mix. This is in a dilute ACETONE environment. 75 ml acetone and then water added to bring to 250 ml was then added to approximately equal volume of TSP 070. This crystal growth is variable in size and growth patterns. Veinous structure is also detected within these crystals, however.

So what would normally be considered very toxic to biological growth allows it to proceed. Rather remarkable.

Recall your alcohol test also planned. Electromagnetic field detection also important on horizon. Want to look at settled precipitate of full size mature TSP 070 culture.

What I wish to do now is concentrate and dry the precipitate to produce a more dense NIR card sample.

I have looked at the TSP 070 - acetone settled precipitate under the scope. Not anything dramatic as the preliminary NIR card also indicated. Dominated by small needle like crystals. 3200x. Both NIR and microscopy and visual texture are not indicating a polymer or plastic compound as of yet.

Microscope, UV, NIR give you a decent first handle on the the nature of a material. Mid IR would be a powerhouse of course.

I am looking at UV of this precipitate. It shows evidence of the 268 nm absorbance which we once again find associated with filam. phages.

You are limiting the range from 300 to 190 nm. Extending the range from 190 nm to 400 nm is introducing reference errors and cuvette problems I believe due to the variation in acetone levels, reference water, and the precipitate which was created in conjunction with acetone. 300 nm limit seems to avoid any problems and spectrum makes sense.

Valley is at 247 nm.

Very sharp strong peak magnitude 1.5 at 197 nm which we have seen with vinyl.

Here is a big surprise for me:

What chemical functional group absorbs ultraviolet at 196 nanometers?

The chemical functional group that absorbs ultraviolet light at 196 nanometers is the benzene ring.

What chemical functional group absorbs ultraviolet between 196 and 198 nanometers?

The chemical functional group that absorbs ultraviolet light between 196 and 198 nanometers is the conjugated double bonds.

(This is consistent with a benzene ring).

At what wavelength in nanometers does a benzene ring absorb ultraviolet?

Benzene typically absorbs ultraviolet (UV) light in the range of 200-280 nanometers (nm).

What chemical structural change in a benzene ring causes absorption to occur at a higher ultraviolet wavelength?

The absorption of ultraviolet (UV) light by a molecule is primarily determined by the presence of certain functional groups or chromophores, which are responsible for the absorption of specific wavelengths of light. In the case of benzene, the absorption of UV light occurs in the range of 200-300 nm, which is known as the aromatic region.

The absorption of UV light by benzene is mainly attributed to the presence of alternating double bonds within the benzene ring, which forms a conjugated system. This conjugated system allows for the delocalization of p electrons over the entire benzene ring, resulting in the absorption of UV light at lower wavelengths.

To cause absorption to occur at a higher UV wavelength, a chemical structural change in the benzene ring would need to disrupt this conjugation. One way to achieve this is by introducing electron-withdrawing groups (EWGs) onto the benzene ring. EWGs are functional groups that withdraw electron density from the ring, thereby breaking the conjugation and shifting the absorption wavelength to higher energies (lower wavelengths).

Examples of EWGs that can be introduced onto the benzene ring include nitro groups (-NO2), carbonyl groups (such as -C=O), or halogens (such as -Cl or -Br). These groups can alter the electronic structure of the benzene ring, resulting in a decrease in the extent of conjugation and a redshift in the absorption wavelength to higher UV energies.

What is an example of a chemical compound that has a carbonyl group attached to a benzene ring?

One example of a chemical compound that has a carbonyl group attached to a benzene ring is acetophenone. Its chemical formula is C8H8O and its structure consists of a benzene ring with a carbonyl group (C=O) attached to it.

At what wavelength in nanometers does acetophenone Absorb ultraviolet?

Acetophenone absorbs ultraviolet light at a wavelength of around 260-270 nanometers.

Comment: That is certainly a big shift from 196 nm? Now the fact that such a "minor" change in benzene causes a shift all the way to 260 to 270 nm is a big red flag for you. Right in the primary target range of DNA, bacterial phages, etc.

On the first level, the way to not as likely err here is to look at the UV profile of a control or reference. If you can't get it you have a problem. Look at the full spectrum such as the valleys. Utlimately you may also simply require some alternate techniques, and cross checking like the alcohol test. These could all be crucial. Multiple test methods are actually required in essentially all circumstances. MID IR please!

Now an important finding, and especially relevant with respect to the comments immediately above. The results with the chilled alcohol test for DNA presence using vial # 15 from the LC run of TSP 070 - acetone is coming up completely negative. This seriously calls into question any claim of bacterial DNA obtained from this particular LC run. It does not negate the previous DNA culture tests analyzed, but it certainly introduces caution into overstating any claims while additional verification remains sought after.

Now we know the UV spectrum was different than anything seen before. Let' review what we saw.

Here are our notes from yesterday, Dec 28 2023:

"Now let's start looking at the precipitate settled on the tube with TSP 070 centrifuged clear + acetone.

The first thing done here is to look at the clear solution which remains after settling over night and forms the precipitate. The UV profile is completely different than anything seen before. It is changing the nature of TSP 070 in a very significant way. What we have is:

> 330 nm to 400 nm : low absorbance, no significant activity

An extremely sharp rise to approx 296 nm and then a sharp flattening of absorbance as we decrease to approx 275 nm.

Then we have a gradual rise in absorbance to a distinct peak that develops at approx. 240 nm.

Then a sharp decrease up to and including the 190 nm range limit.

Quite different than anything involving TSP 070 before.

The first things we observe with the precipitate is that:

- 1. It is white
- 2. Rather dense, as it settles quickly in solution"

This is a new type of sample produced. It is not to be associated with all the bactereral DNA/phage DNA investigations. This is a separate chemical reaction between TSP 070 and acetone.

- With the acetone precipitate, you may well be dealing with something entirely different than before:
- 1. We have made a microscopic examination.
- 2. You have a UV profile/spectrum.

You realize what you sorely need but do not have. An image database. You need to see your images to sort out the data complexity at times. I cannot open two instances of this database, need to think on this for a bit.

2023.12.30 TSP 070 Acetone-Phage-ImageDB

I have now made a very useful Lab Log Image Database in conjuction with the text log. Output is a PDF file. looks good. Landscape format. Date sort or title sort both can be available. PDF format allows to be used actively in conjunction with the Lab Log database. PDF format can be text searched. Image database will need to be kept updated to be useful, photo editor can work for the current daily needs.

There is a very fine line between bacterial DNA absorption and filamentous Bacteriophage DNA absorption; I am not sure that I am capable of resolving that difference or not. Range seems to be between 265 - 269 nm, that is a tight line.

Immediate topics are:

1. Nature of compound formed with TSP 070 + acetone. Use both UV, NIR and scope.

2. Reference surprise difference between ceramic and NIR card for TSP 070 + acetone compound. Controls will need to be run and explanation for differnce sought.

3. Electrical field detection on TSP 070. Repeat? Same on TSP 100?

It took quite a long time for the Chat to come up with "AN" answer to this question. I have learned not to trust the results on any difficult question with multiple crosschecking the inquiry from several different angles as there are almost always significant conflicts and contradictions that result with the answers.

Does bacterial dNA absorb ultraviolet energy at exactly the same wavelength that a filamentous bacteriophage absorbs ultraviolet energy?

No, bacterial DNA and filamentous bacteriophage do not absorb ultraviolet (UV) energy at exactly the same wavelength. The absorption of UV energy by molecules depends on their chemical structure and composition. While both bacterial DNA and filamentous bacteriophage contain nucleic acids, their specific sequences and arrangements can vary, resulting in different absorption properties.

Bacterial DNA typically absorbs UV light most strongly at a wavelength of around 260 nm. This is because nucleic acids, such as DNA, contain aromatic rings that absorb UV light in the range of 250-270 nm.

On the other hand, filamentous bacteriophages, like other proteins, absorb UV light primarily in the range of 280-290 nm. This is because proteins contain amino acids with aromatic side chains, such as tryptophan and tyrosine, which absorb UV light in this range.

Therefore, while both bacterial DNA and filamentous bacteriophage absorb UV energy, they do so at different wavelengths due to their different molecular compositions.

at what wavelengths in nanometers does Bacterial dNA absorb ultraviolet?

Bacterial DNA typically absorbs ultraviolet (UV) light in the range of 260-280 nanometers (nm). The maximum absorption occurs around 260 nm.

At what wavelengths in nanometers do filamentous bacteriophages absorb ultraviolet?

Filamentous bacteriophages typically absorb ultraviolet (UV) light in the range of 260-280 nanometers (nm).

At what wavelengths in nanometers is filamentous bacteriophage DNA ultraviolet energy absorbed?

Filamentous bacteriophage DNA typically absorbs ultraviolet (UV) energy at wavelengths below 300 nanometers (nm). Specifically, the absorption peak is often observed around 260 nm.

"https://link.springer.com protocol 10.1007 978-1-4939-7386-6 4

Filamentous Bacteriophage Viruses: Preparation, Magic-Angle ... - Springer Pure phages exhibit a typical UV absorption spectrum having a broad plateau at 260-280 nm, with a shallow maximum at 269 nm and a minimum at 245 nm resulting from both proteins and DNA [19, 27]. Phage purity is
CI Laboratory Notebook Vol XXXIII : Appendix (Digital Format)

determined by the ratio between maximum and minimum absorbance, and by the ratio"

On Dec 24 +/- we have two separate literature UV spectra of bacteriophages. One is of T7 and the other is of M13.

T7 is not of filamentous nature: https://www.jbc.org article S0021-9258(20)53625-9 fulltext Structural Characterization of the Bacteriophage T7 Tail Machinery*? Most bacterial viruses need a specialized machinery, called "tail," to inject their genomes inside the bacterial cytoplasm without disrupting the cellular integrity. Bacteriophage T7 is a well characterized member of the Podoviridae family infecting Escherichia coli, and it has a short noncontractile tail that assembles sequentially on the viral head after DNA packaging.

M13 is.

PURIFIED M13 absorbs at 269 nm and valley at 245 nm according to literature

According to T7 spectrum shown, max absorbance seems to be at approx 261.5 and valley seems to be at approx. 236.5. This most likely assumes again it is a pure sample since this was done on a space mission.

So all phages are definitely not the same.

OK we see above why absorption in phage is at approx 270. It is as I suspected. Because it has BOTH proteins and DNA, it is absorbing generally midway between 260 (DNA) and 270(proteins - to even 280). So your three tier divison layer is a very reasonable argument to make.

What we would really like to do is make the distinction between phage prospect and DNA the best we can, again and again.

Then we would like to go on with the TSP 070 + acetone compound nature as a separate problem.

(Note recall: the bacterial mitigation protocol is at 265 nm).

I think what I need to do is tally up all my observations in a frequency count just like I did with the functional groups tally to get a handle on where the observations lie, 260, 270, 280 nm etc.. As it is, they are all scattered. But now, between image and textual database I can tally these up.

2023.12.31 Afflicted Skin

Need to:

- 1. Make a frequency tally on the UV absorbance 260-280 nm 2. Determine the nature of the TSP 070 acetone precipitate
- 3. Electromagnetic field detection from the culture

But for now a quick important paper is in the works. The influence of the polymers on the skin is now clearly documented.

Cross Domain Bacteria and Synthetic Biology Equivalence: Blood clots, Skin Affliction & Polymers

The paper is written.