

# Preparation, properties of pullulan and sodium alginate membranes under different temperature

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## Reaction scheme

1) Though drying

Sodium alginate + pullulan + 4%w/v glycerol solution → oven

2) Through cross-linking

Sodium alginate + pullulan + 4%w/v glycerol solution → copper sulfate (10%w/v) water bath

## Experiment

### 1) Equipment

Beakers (100ml x26 )

Beakers (400ml x 2)

Glass rod

Grinder

Magnetic stirrer

Measuring cylinder

Oven

Watch glass x 6

Spatula

Water tank x 1

Water bath incubator

Watch glass x 6

Bunsen burner

Asbestosed wire gauze

Crucible tongs

### 2) Chemicals

Copper sulfate

Absolute ethyl alcohol

Glycerol (4%w/v)

Pullulan

Sodium alginate



Fig. 1 Oven

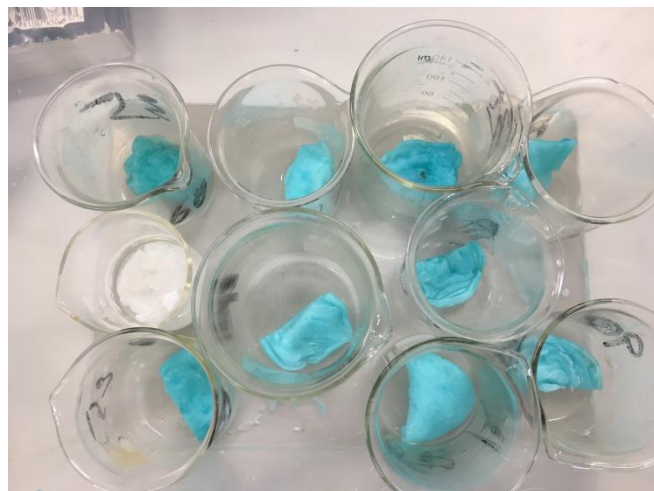
Fig. 2 Magnetic stirrer

Fig. 3 Water tank

### 3) Instructions

#### A) Make the membrane

- Grind sodium alginate (takes time to dissolve as granular shape, might form gel-like structure)
- Stir the mixture of chemicals (in proportion of 1.2g pullulan, 0.8g sodium alginate, 40ml 4%w/v glycerol solution)
- Wait for all solid components dissolved (usually take 15-20 minutes)
- Pour 20g of the mixture on same size of Petri dish evenly
- **For copper cross-linking:** water bath the Petri dish into water with proper temperature, use a dropper to add 10% w/v copper sulfate chloride solution drop by drop, until the copper sulfate solution submerge the membrane entirely. Immerse in for 20 minutes
- **For drying:** place the Petri dish into an oven, bake for 2h under controlled temperature
- Collect the membrane by scratching or peeling (aid with knife)



#### B) Immerse in water and alcohol

##### **For copper cross-linking membrane, do the following to all membranes made:**

- Use a pair of scissors to cut the membrane in halves
- Measure their weight respectively
- One of the pieces is placed in a 100ml beaker almost full of water
- The other pieces is placed in a 100ml beaker almost full of alcohol
- Wait until 4 days pass
- Wipe the samples immersed in water with filter paper to dry the surface
- Put the samples immersed in alcohol on filter paper and let it dry overnight (alcohol evaporates)
- Measure the weight of the samples

##### **For solvent-evaporation membrane:**

- Place the membrane on watch glass carefully to avoid folding
- Cut the membrane into regular shape, 5cm x 6cm
- Cut the rectangular shape into strips, 5cm x 2cm
- Separate the remaining membrane into 2 halves, measure the mass of 2 halves and record mass of them, place them into 100ml beakers which is full of alcohol or water respectively, immerse it for half a day
- For those membrane in water, measure the remaining gel-like pieces that can be drawn out of water
- For those membrane in alcohol, measure the mass after this dehydration

Fig. 4 cross-linking samples immersed in alcohol

#### 4) Temperature of membrane

Through cross-linking	Temp /°C	Room temp	40	50	60	70	80	90	
	Temp of water bath/°C	27.9	41.6	51.9	59.7	70.2	80.9	90.0	
	Temp of agent/°C	28.5	30.4	29.8	34.1	29.2	63.9	29.3	
	Temperature differences Water-agent/°C	-0.6	11.2	22.1	25.6	41.0	17.0	60.7	
	Mass used /g	20.93	18.44	19.89	18.603	19.06	13.78	14.77	
Through evaporation of solvent	Temp /°C			50	60	70	80	90	100

#### 5) Products

##### Cross-linking products

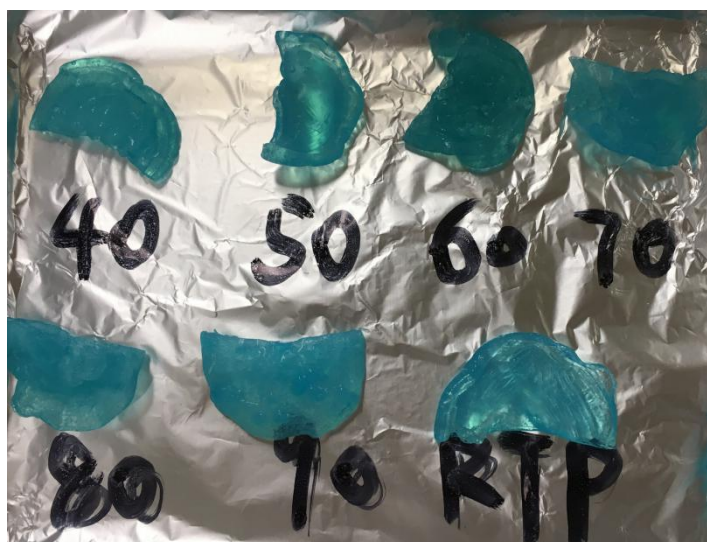


Fig.5 (left) cross-linking membrane immersed in water

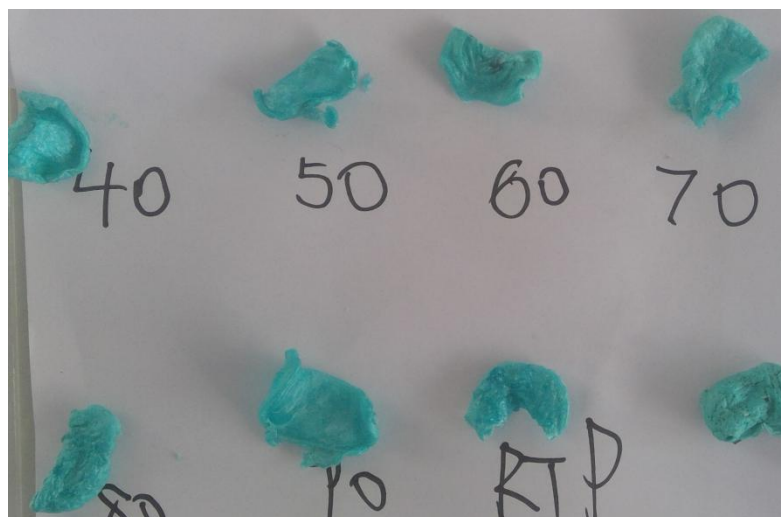


Fig.6 (right) cross-linking membrane immersed in alcohol

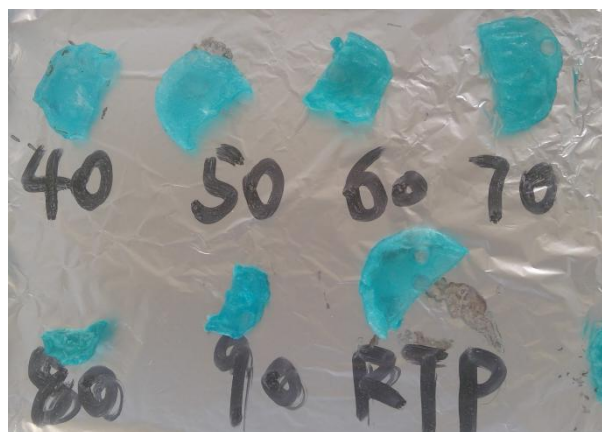


Fig.7 (left) cross-linking membrane immersed in water and left in air overnight

Fig. 8 (right) collection of some samples





# Solvent-evaporation membrane

Fig. 9&10 membrane

formed when temperature at 50°C  
The membrane under this temperature failed to form in 2 hours, so I added another hour for membrane formation.



Fig. 11&12 membrane formed when temperature at 60°C



Fig. 13&14 membrane formed when temperature at 70°C



Fig. 15&16 membrane formed when temperature at 90°C



Fig. 17&18 membrane formed when temperature at 100°C

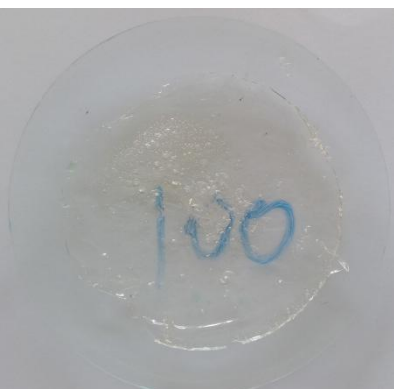
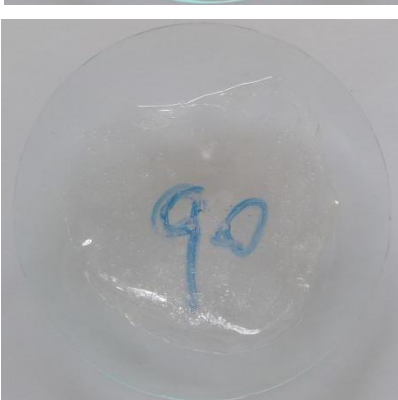
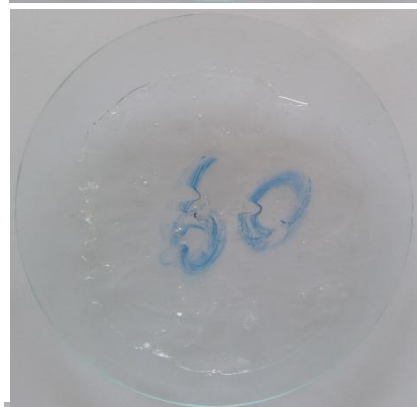




Fig 19&20 colour comparison of solvent-evaporation membrane (real colour & false colour)

## 6) Test on properties of membrane

### A) Thickness of cross-linking membrane

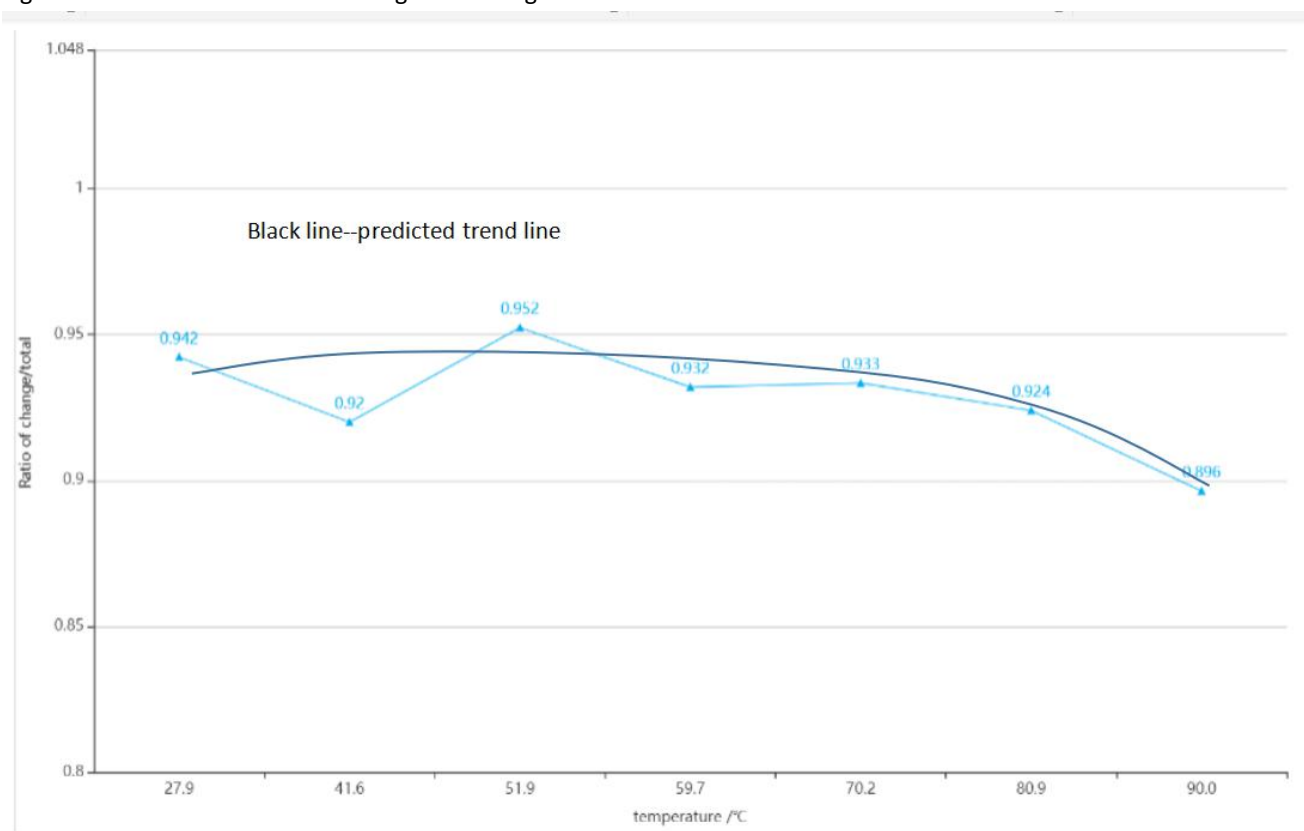
Immersed in alcohol	Temperature/°C	Room temperature	40	50	60	70	80	90
	Thickest thickness/mm	4.58	3.85	4.06	4.11	2.71	3.57	2.93
	Thinnest thickness/mm	2.06	2.69	3.26	2.74	2.54	2.17	1.69
	Difference between thickness /mm	2.52	1.16	0.80	1.37	0.17	1.40	1.24
Immersed in water	Thickest thickness/mm	3.55	4.58	7.58	2.53	4.77	1.24	2.56
	Thinnest thickness/mm	0.89	0.54	0.21	0.11	0.88	0.39	0.10
	Difference between thickness /mm	2.66	4.04	7.37	2.42	3.89	0.85	2.46

Because side chains in high temperature will interact quickly, so the surface might bent, and eventually the thickness is not even.

### B) Dehydration of membranes in alcohol

	temperature	Room temperature	40	50	60	70	80	90	100
Cross-linking	mass original data /g	9.71	7.84	10.30	8.29	9.01	7.24	6.47	
	After immersing mass /g	0.56	0.63	0.49	0.51	0.60	0.55	0.67	
	Cross-linking change of mass alcohol/g	-9.15	-7.16	-9.81	-7.73	-8.41	-6.69	-5.80	
	Ratio of change/total %	94.23	92.00	95.24	93.2	93.34	92.40	89.64	

Fig.21 ratio of membrane mass changed over original mass after immersed in alcohol





	temperature	Room temperature	40	50	60	70	80	90	100
Solvent evaporation	mass original data/g			0.47	0.60	0.57	0.71	0.26	0.54
	After immersing mass /g			0.41	0.44	0.45	0.60	0.20	0.38
	After immersing mass changed /g			0.06	0.16	0.12	0.11	0.06	0.16
	After immersing mass changed/per original mass			0.128	0.267	0.211	0.155	0.231	0.185
	After immersing mass remained/per original mass			0.872	0.733	0.789	0.845	0.769	0.815

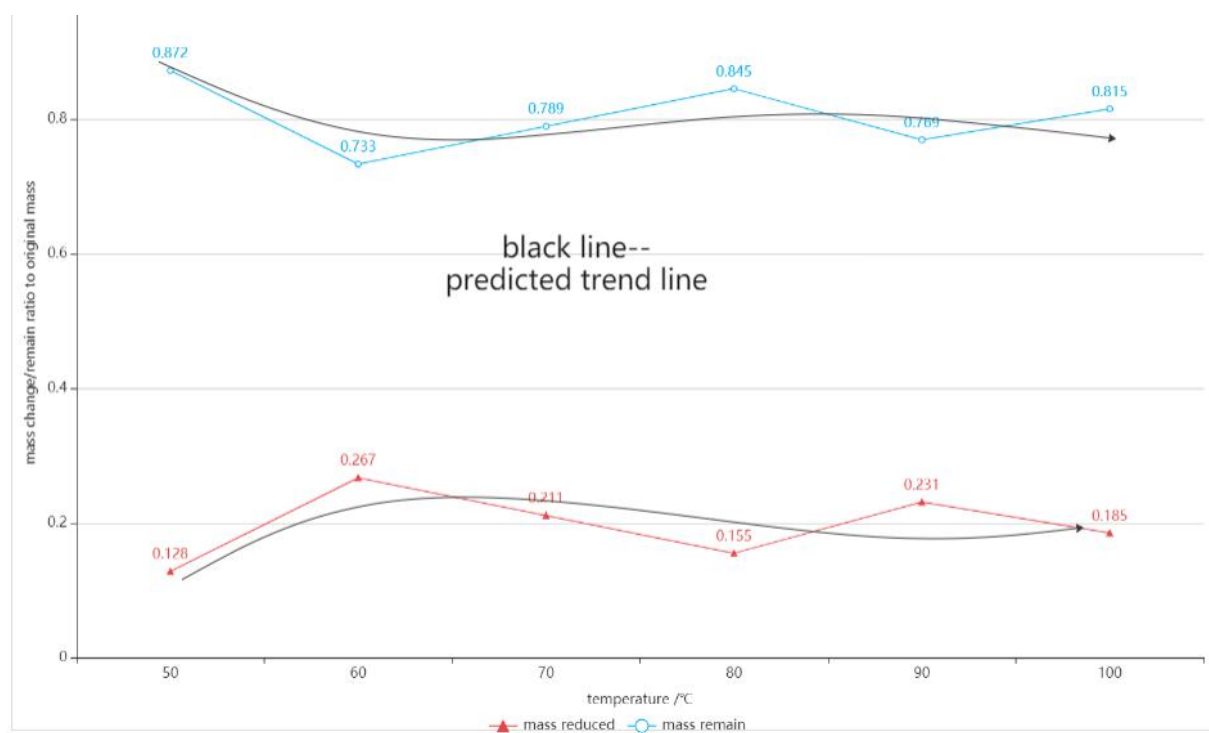


Fig 22. mass change/remain ratio to original mass

### C) Solubility and absorption of water by membranes

Temperature/°C	Room temp 27.9	40	50	60	70	80	90
Cross-linking mass original data in water/g	10.48	9.71	9.41	9.32	8.92 15.57	5.74	7.40
After immersing mass - water/g (varies by time )	6.91	5.47	7.28	6.10	6.06	4.07	4.83
	5.06		6.04		4.82 9.01		4.92 5.45
After immersing mass changed/g	-3.57	-4.24	-2.13	-3.22	-2.86	-1.67	-2.57
	-5.42		-3.37		-4.10		-2.48
					-6.56		-1.95

After immersing mass change water per gram of original (no unit)	-0.34	-0.44	-0.23	-0.35	-0.32	-0.29	-0.34
	-0.52		-0.36		-0.46		-0.34
					-0.42		-0.26
Ratio of mass change per gram/ water bath degree (10 <sup>-3</sup> )	12.19	10.58	4.43	5.86	4.56	3.58	3.74
	18.64		6.93		6.55		3.74
					5.98		2.86
Immersing mass ratio/ temperature differences 10 <sup>-3</sup>	--	-9.8	-9.95 -16.3	-13	-7.8	-17	-23 -4.2

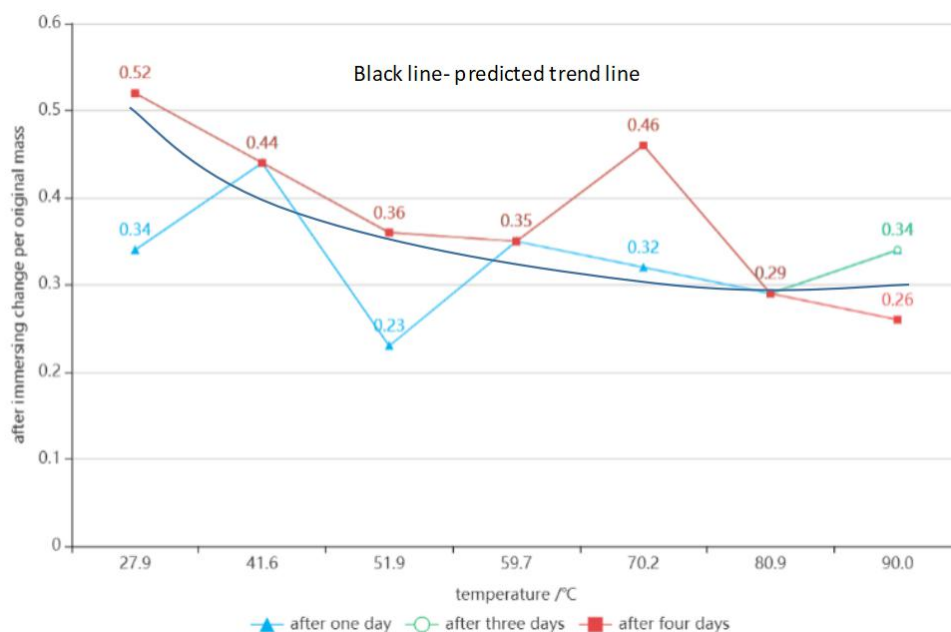


Fig 23. mass change per original mass after immersed in water

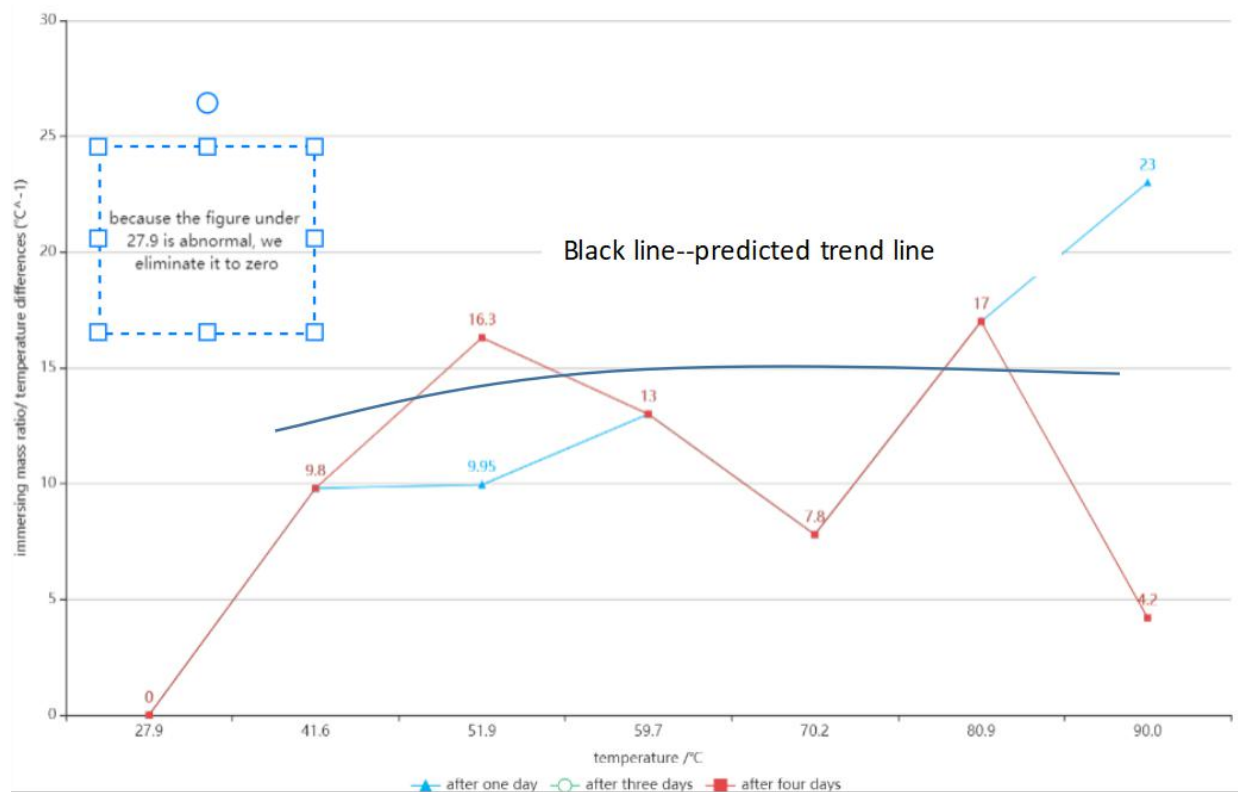
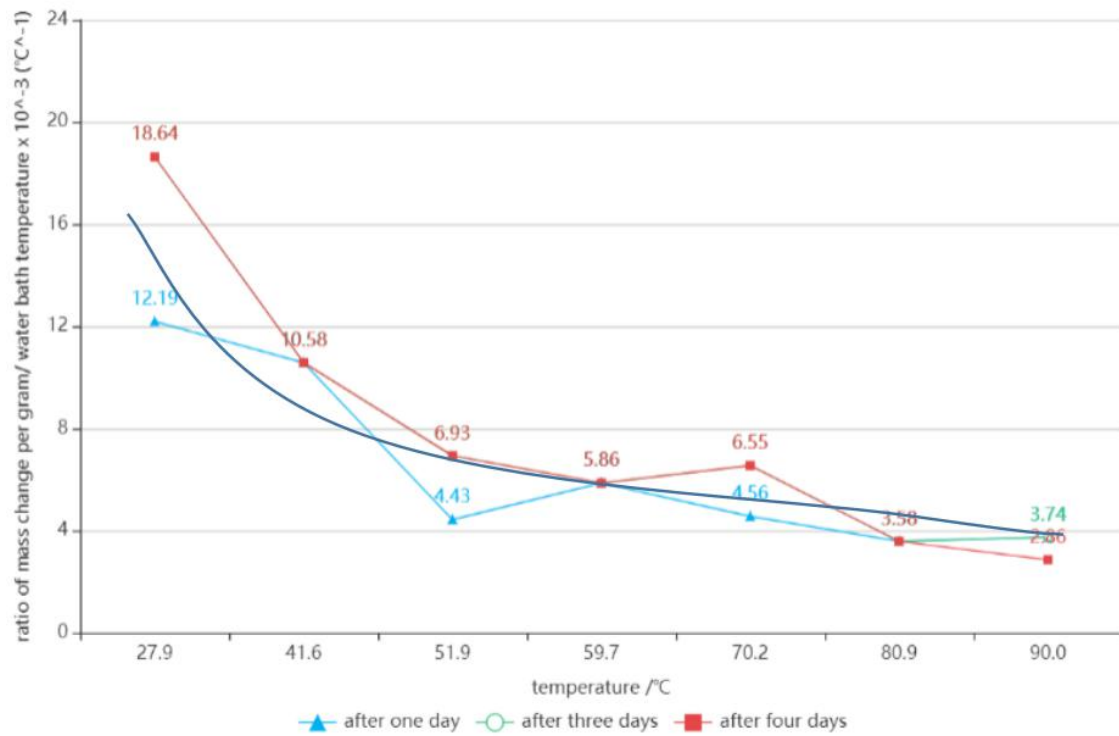


Fig 24.proportion of reduced mass changed when immersed in water per water bath degree

Fig 25. proportion of reduced mass changed when immersed in water per difference in temperature of water bath difference

Mass after long time /g	0.308	0.428	0.344	0.305	0.291	0.288	0.282
Change of mass after long time/g	-10.172	-9.282	-9.066	-9.015	-8.629	-5.452	-7.118
Immersing ratio/mass long time changed	0.0294	0.044	0.036	0.0327	0.0326	0.05017	0.0381

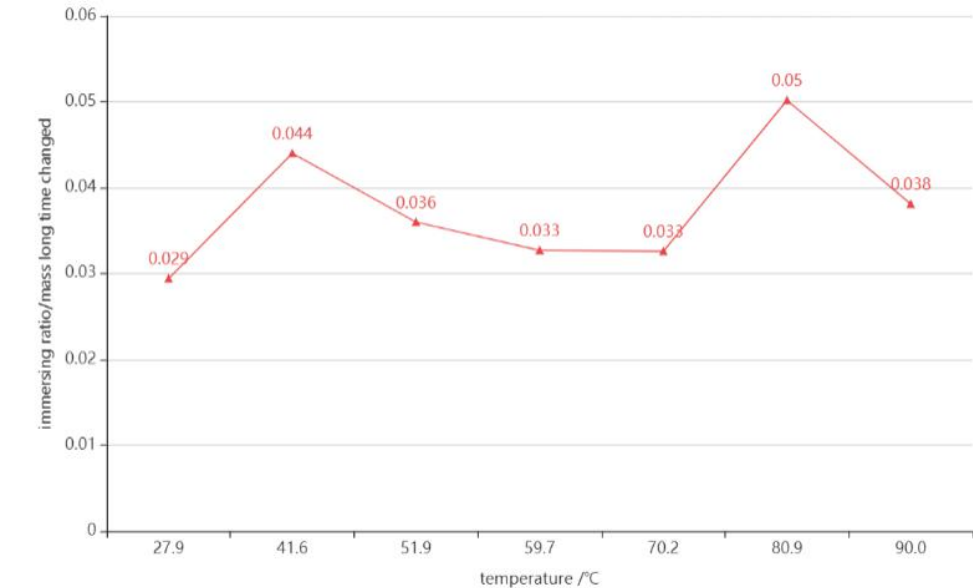


Fig 26. mass remain after a long time over original mass

Temperature/°C	50	60	70	80	90	100
Evaporation mass original data in water/g	0.42	0.57	0.59	0.41	0.69	0.77
After immersing mass - water/g	7.76	6.01	4.42	4.80	4.94	0 (all dissolved)
After immersing mass changed /g	+7.34	+5.44	+3.83	+4.39	+4.25	
After immersing mass changed/per original mass	17.48	9.54	6.49	10.71	6.16	

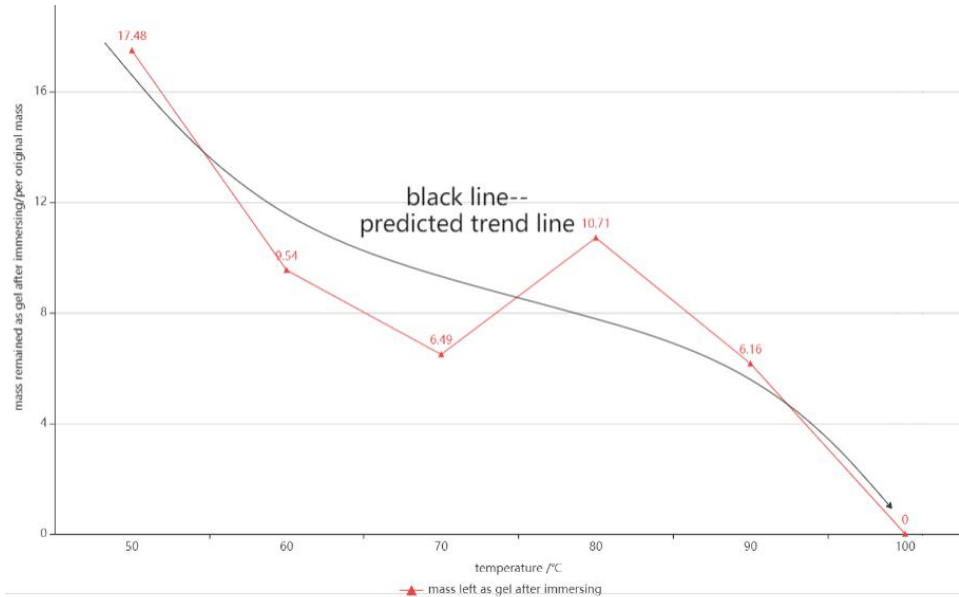


Fig 27. mass of gel left in water after immersed per original mass

#### D) Mechanical strength of solvent-evaporation membrane

Temperature/°C	50	60	70	80	90	100
Maximum tension required to break the membrane string / N	5.0	5.8	5.8	9.0	5.5	Exceeding maximum range (10N)

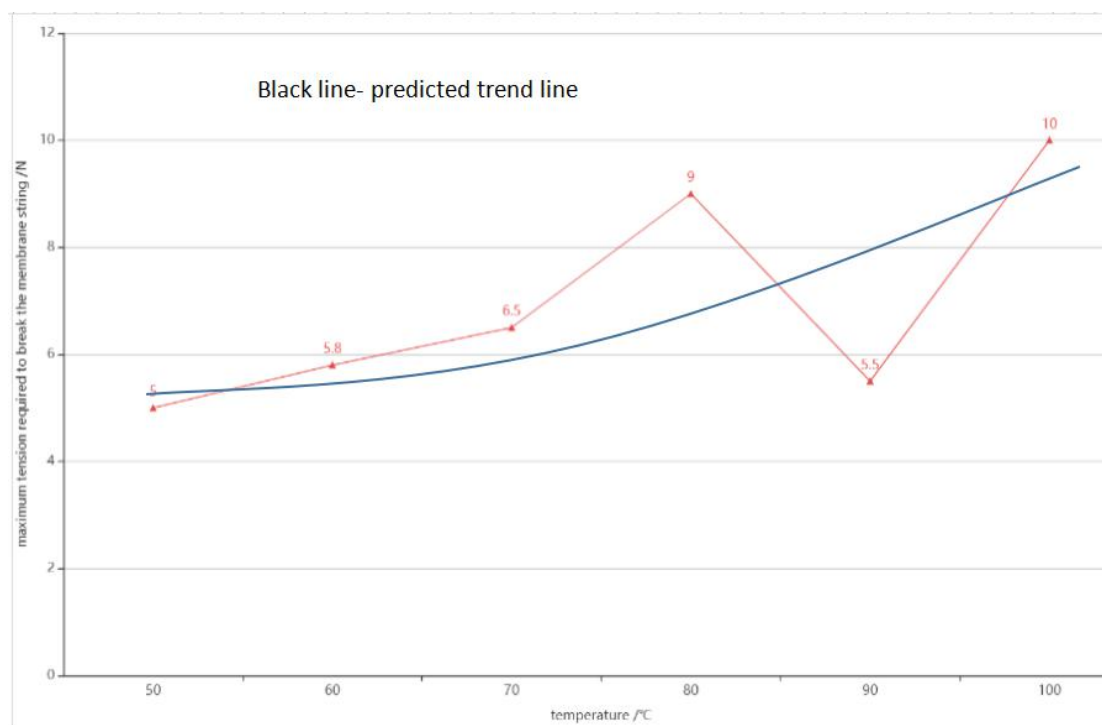


Fig 28. maximum tension required to break the membrane string /N



# Experiment analysis

## From the appearance of product

Under different temperature, membrane component tend to interact differently. This is probably related to how the temperature influence the tangling of side chains in polysaccharide.

Generally through cross-linking the difference between thickness of membrane under different temperature is more obvious, and for solvent-evaporation is less obvious considering its compact structure.

The membrane formed by both mechanisms are previous to light, but the colour of cross-linking membrane is blue because i used copper ions as cross-linking agent. The color of solvent-evaporation membrane is totally transparent, but with the temperature of formation increase, the membrane tend to be more yellowish (with some caramel colour). this is attribute to the Caramelization effect.

For solvent-evaporation membrane, under higher temperature there are likely to me more bubbles in the membrane, and the membrane is harder. This can also use Caramelization effect to explain.

## From dehydration, dissolving and absorption of water

### A) Cross-linking

Cross-linking membrane will have significant change in the thickness of membrane after immersed in water and alcohol respectively. Form the data, we can find out that the greatest difference between thickness of membrane which immersed in alcohol is the group made under room temperature, and such measure of membrane in water is the group made under 50°C.

Cross-linking membrane immersed in alcohol are dehydrated because water diffuse from high concentration to low concentration. Therefore, by this process we remove the free water combined in the membrane structure and left the water combined in the structure precisely. For the case of immersing in water, the mechanism is more complicated. This is because the membrane will partially dissolve in water (probably because not every position allows cross-linking to form have already formed cross-linking, so the vacant places may offer water to dissolve the structure) and partially absorb water (the water are mainly adsorbed after immersed in water, but before that during the membrane formation there are lots of water absorbed in the membrane structure). therefore, we need special measurement to determine how many water are freely absorbed in the membrane structure and the extend of dissolving.

So we now have the data of membrane being immersed in water and the dried in air for a long time. By calculating this mass remain/total and compare to the mass remain/total in membrane immersed in alcohol, we can determine the proportion of membrane dissolved.

Temperature/°C	Room temp	40	50	60	70	80	90
Remaining mass (immersed in water and dried)/mass original (M4)	0.0294	0.044	0.036	0.0327	0.0326	0.05017	0.0381
Remaining mass (immersed in alcohol) /original mass (M2)	0.0577	0.08	0.0476	0.068	0.0666	0.076	0.1036
Ratio of dissolved mass in water (m) g	0.0283	0.036	0.0116	0.0353	0.034	0.02583	0.0655
After immersing in water mass change water per gram of original (no unit) (M3) Influence both on dissolve polysaccharide and other things	-0.34	-0.44	-0.23	-0.35	-0.32	-0.29	-0.34
	-0.52		-0.36		-0.46		
							-0.26
Removing the influence of dissolved mass and find the changed proportion (water) (M3+m)	0.3117	0.404	0.2184	0.3147	0.286	0.26417	0.2745
	0.4917		0.3484		0.426		
							0.1945
Ability of dissolved mass to combined water (m/M3+m)	0.09	0.089	0.053	0.112	0.118	0.0978	0.2386
	0.057		0.033		0.0798		
							0.3367
The best fitting trend (-0.05)	0.007	0.039	0.003	0.062	0.068	0.0478	0.1886

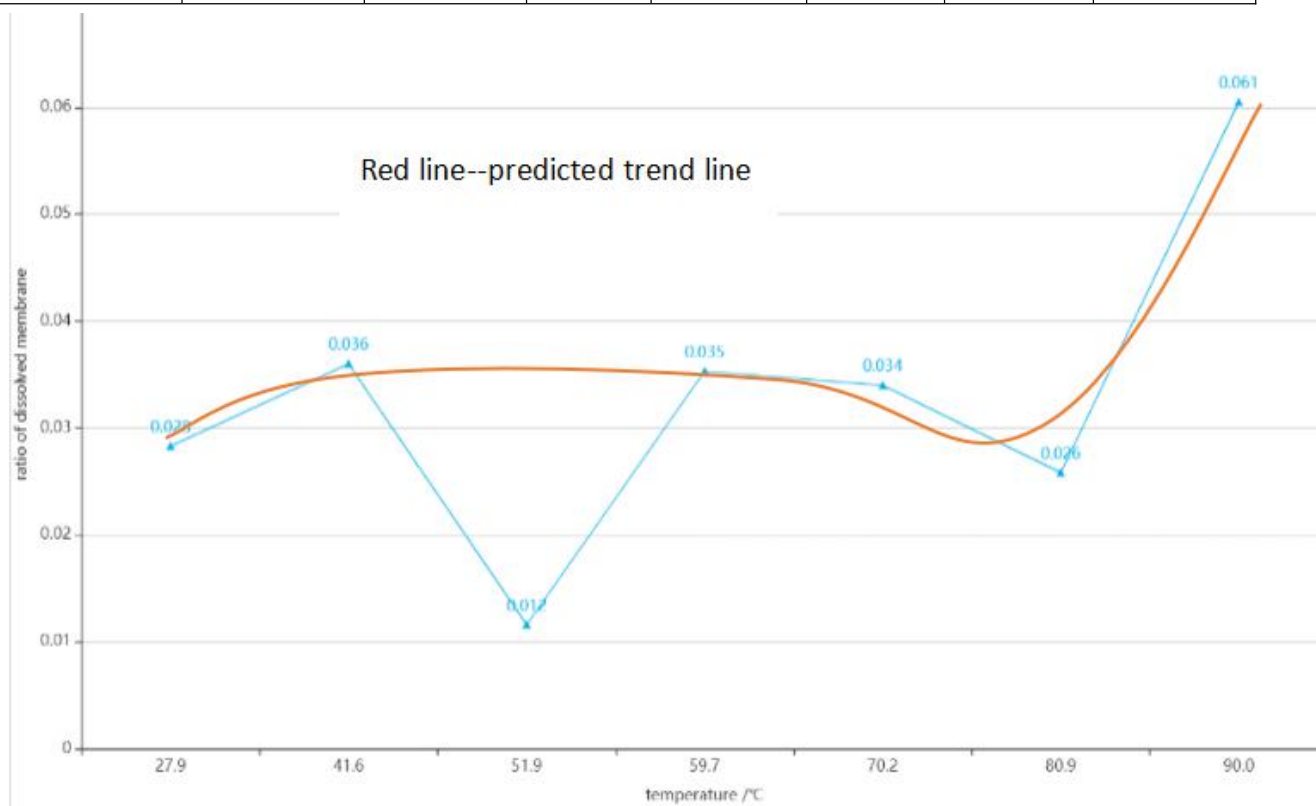


Fig 29. proportion of mass loss in water (m)

This graph shows the trend of mass dissolved in water under different temperature. Generally, the amount of dissolved mass in high temperature is more, probably because the molecules have

more kinetic energy when forming the membrane, so the basic membrane structure formed under a short period of time. In result, the polysaccharide which is the central part of membrane may not directly combine with copper ion to form chemical bonds and therefore remain more features of polysaccharide mixture which can completely dissociate in water. Under lower temperature, when the cross-linking agent is reacting on the surface of membrane, it can also penetrate and diffuse into the inner part of membrane because the slower motion of particles allows longer time to form a firm barrier outside the membrane. Therefore, there are more time available for the copper cross-linking to form, and the product is more compact and less soluble.

Worth mentioning, the membrane at 50°C have the least mass dissolved. Referring to the former data, the thickness of membrane under such temperature is the thickest, so more reagents are accumulated in the center of the membrane structure. This suggested that the surface area for membrane under 50 °C is smallest and thus the rate of diffusion in water is lower than predicted trend. It is also reasonable to attribute the concave-convex structure to the interaction within polysaccharide chains.

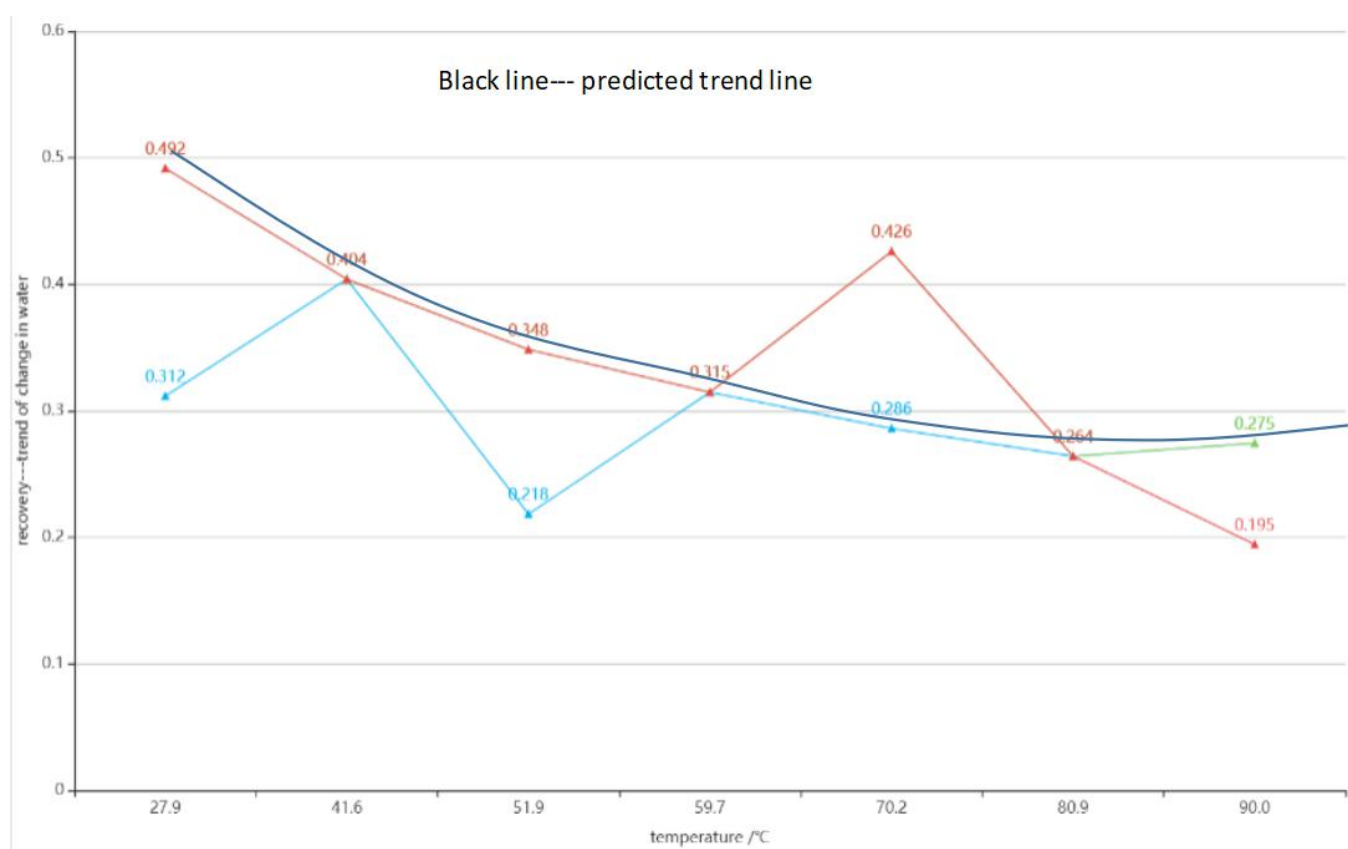


Fig 30. Removing influence of dissolved polysaccharide mixture and calculate how many proportion of mass reduced due to various reasons

This figure study on the change in water amount under various temperature, and it required mathematical processing to transform the data plotted here.

Deduction on the component of the membrane:

Normal cross-linking membrane (M1):  $M1=A+B+C+D$

Polysaccharide mixture (A) + combined water (B) + free water(C) + water trapped in center of membrane (D)

After immersed in alcohol (M2):  $M2=A+B$

Polysaccharide mixture (A) +combined water (B)

After immersed in water (M3):  $M3=A'+B'+C'+D'=A+B+C+D-m-n+p$

Dissolved Polysaccharide mixture (A') + changed combined water(B')+ more free water (C')+ water trapped in center of membrane (D')

And be more specific, the dissolved polysaccharide will take away some water combined to membrane component and water originally trapped in center because they dissolved in water as well.  $A'+B'+D'=A+B+D-m-n$  (m for water loss due to dissolved structure, n for water loss due to its position in center)

For free water, the material tend to absorb water or any other small molecules. Here I used water to test the absorption of water. Therefore,  $C=C'+p$ . Base on the result, I can see that though the membrane absorbed water, the effect on dissolved amount is more significant. Therefore, we can conclude that  $m+n>p$

After immersed in water and dried for a long time (M4):  $M4=A'+B'$

Dissolved Polysaccharide mixture (A') + changed combined water (B')

There is no free water left, so  $C'=0$  in this case; there might be slightly water still trapped in membrane center, so we use approximation to say this amount is zero.  $D'\rightarrow 0$ .

By subtracting M1 by M2, we can get C and D (mass of free water and water trapped in center of membrane) :  $M1-M2=A+B+C+D-(A+B)=C+D$

$M3-M4=A'+B'+C'+D'-(A'+B')=C'+D'=C+D-n+p$

$M2-M4=A+B-(A'+B')=m$

$M3+m=A'+B'+C'+D'-m=(A+B+C+D-m-n+p)+m=A+B+C+D-n+p$

We can have the data of M1 as original mass

$M1-(M3+m)=p-n$

$M1-M2=A+B$

Firstly we can compare whether the p-n is calculated to be the same to verify our result

Temperature/°C	Room temp	40	50	60	70	80	90
$\Delta(p-n) /g$	7.21/5.327	5.787	7.3548/6.13	6.387	6.369/5.12	4.2236	5.3867/5.9607
$\Delta(p-n)/M1$ (no unit)	0.688/0.508	0.59598	0.7816/0.6514	0.6853	0.714/0.574	0.7358	0.7279/0.8055
A+B	9.8	8.9332	8.9621	8.6862	8.3259	5.3037	6.63336
A+B/M1	0.935	0.92	0.952	0.9739	0.93339	0.924	0.8964

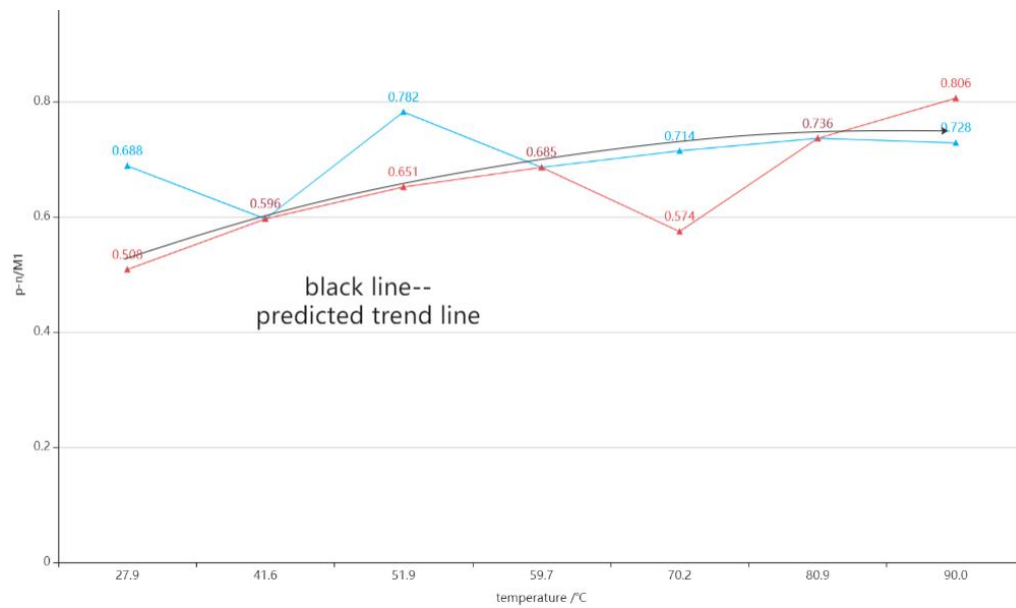


Fig 31. p-n/M1 and its predicted trend line (talk about free water combined in the membrane)

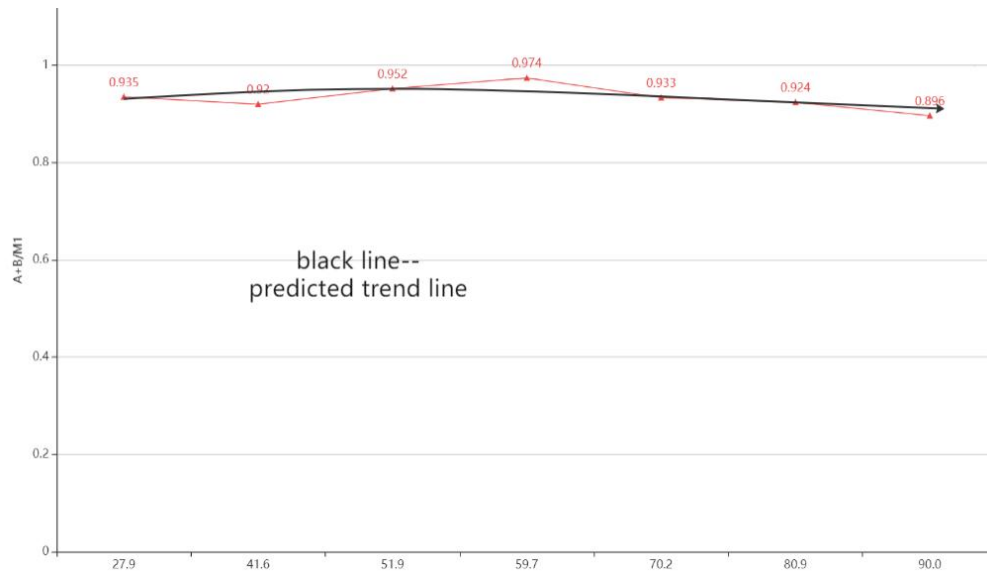


Fig 32. the proportion of solid structure in the original mass

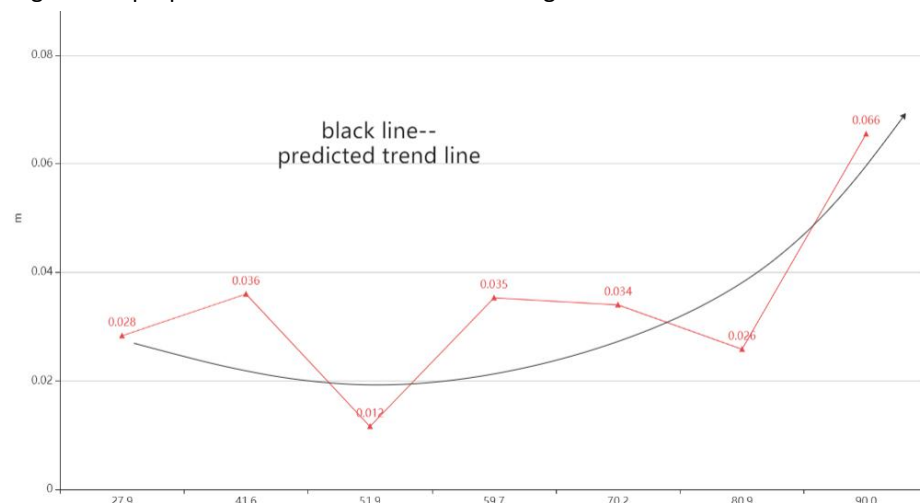
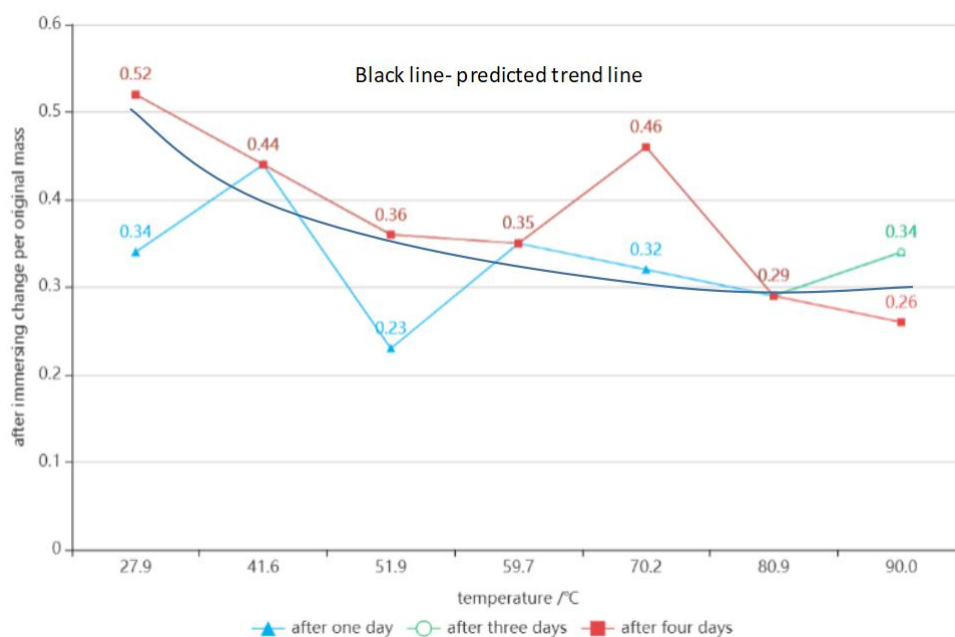


Fig 33. Mass ratio of dissolved membrane



Fig 34. The reduced of mass/total mass with all the factors in.



Note: p-n is actually a negative number because in total the water is lost after immersing. Fig (31) suggest that group with higher temperature tend to have higher amount of water loss. The mechanism underlying is the same as Fig (29). revealed. Fig (32) shows that in all membrane the proportion of polysaccharide and combined water by mass is mainly the same and fluctuate around 93% . Fig(33). shows the proportion dissolved in water and that with the temperature of membrane formation increase the extend of dissolve increased. However in Fig (34) shows that the total mass decreased under higher temperature is lesser in general. This suggested that other than free water, combined water, dissolved mass and loss of ingredients in the center of membrane, there are also other factors influencing the water amount attached to the membrane structure. My hypothesis is that this unconcerned factor is related to the adsorption of water and determined by the surface area of the membrane. And why under this temperature the appearance of membrane is so flat also is related to temperature.

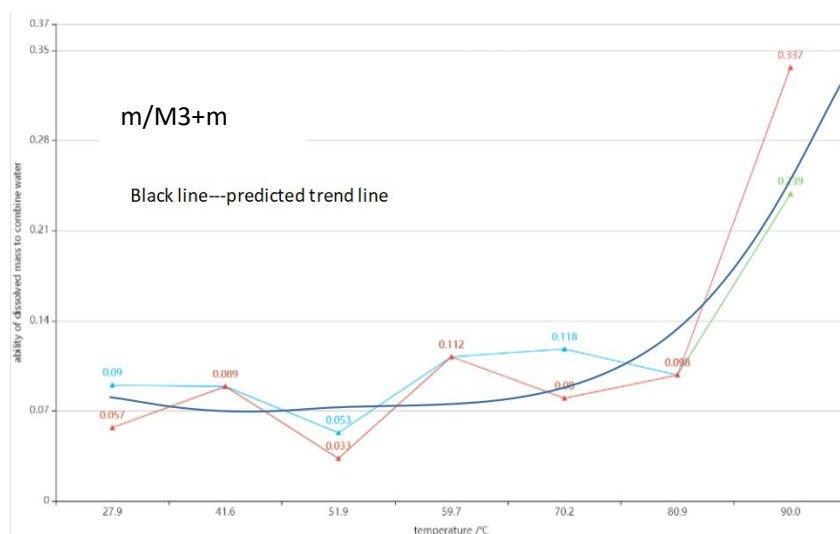
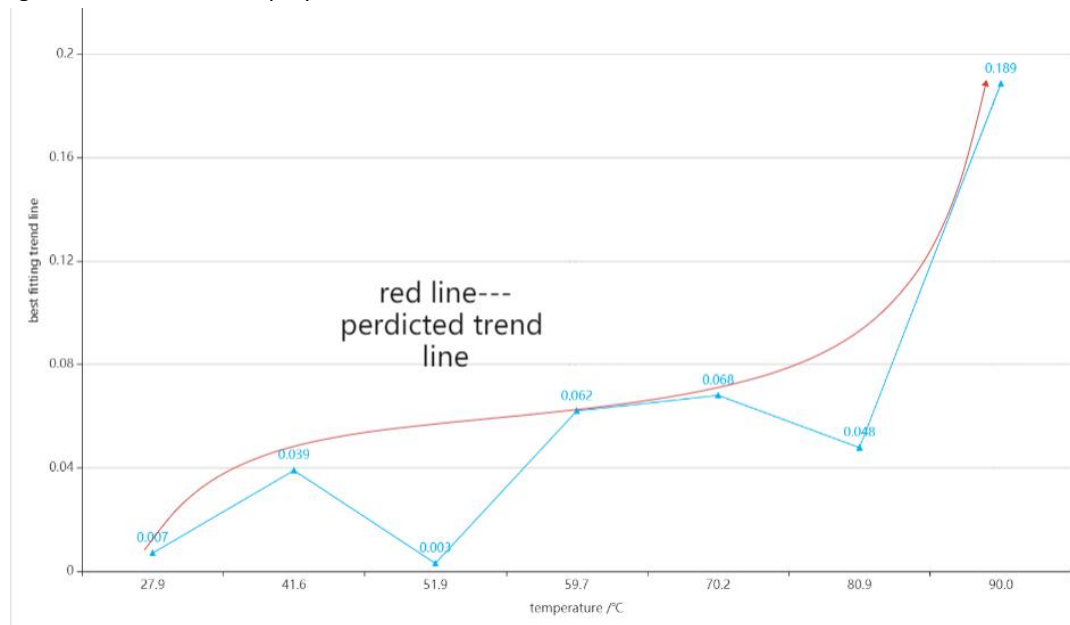


Fig 35. The ability of polysaccharide to combined water

Fig 36. Best trend line of polysaccharide to combined water



When the polysaccharide mixture dissolves in water, some water that is fixed in the membrane will also dissolve. With this loosen structure, the water trapped in the center of membrane and free water are more likely to escape. Therefore, by comparing the ratio of mass dissolved over water loss, we know that how many of mass correspond to per unit of water loss. From data displayed in Fig 33. and Fig 34. we can see that more mass are required to dissolve certain amount of water under high temperature.

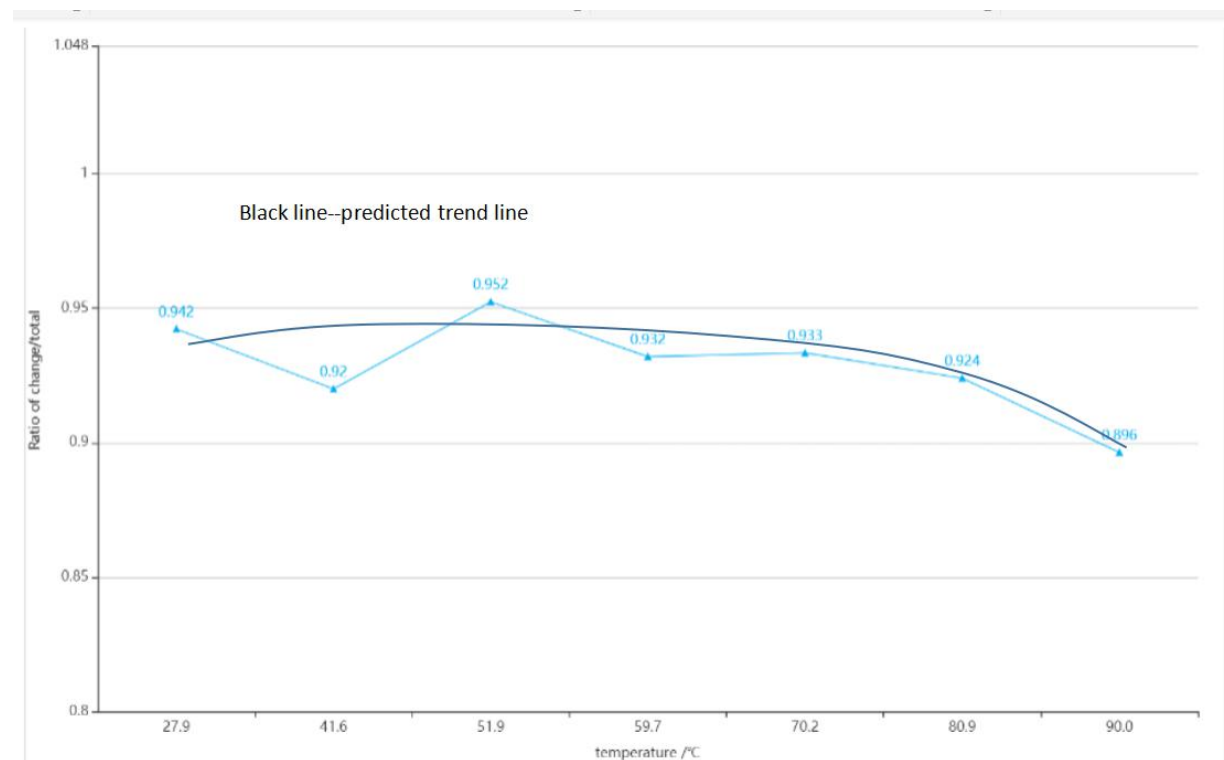


Fig 37. ratio of water loss per original mass after immersed in alcohol and dried

This show that under the influence of ethanol the mass reduce significantly, and such reduction became more significant higher temperature.

However, the conclusion drawn by the deduction model might not be exactly right, because alcohol can have the same function as glycerol to act like stabilizer in the membrane structure. Therefore, some ethanol molecules are attached to the inner part of membrane, which might stiffen the membrane to forbid some extend of disassembling of structure and cause some mass increase.

### B) Solvent-evaporation membrane

	temperature	50	60	70	80	90	100
In alcohol	After immersing mass changed/per original mass	0.128	0.267	0.211	0.155	0.231	0.185
	After immersing mass remained/per original mass	0.872	0.733	0.789	0.845	0.769	0.815
In water	After immersing mass changed/per original mass	17.48	9.54	6.49	10.71	6.16	---

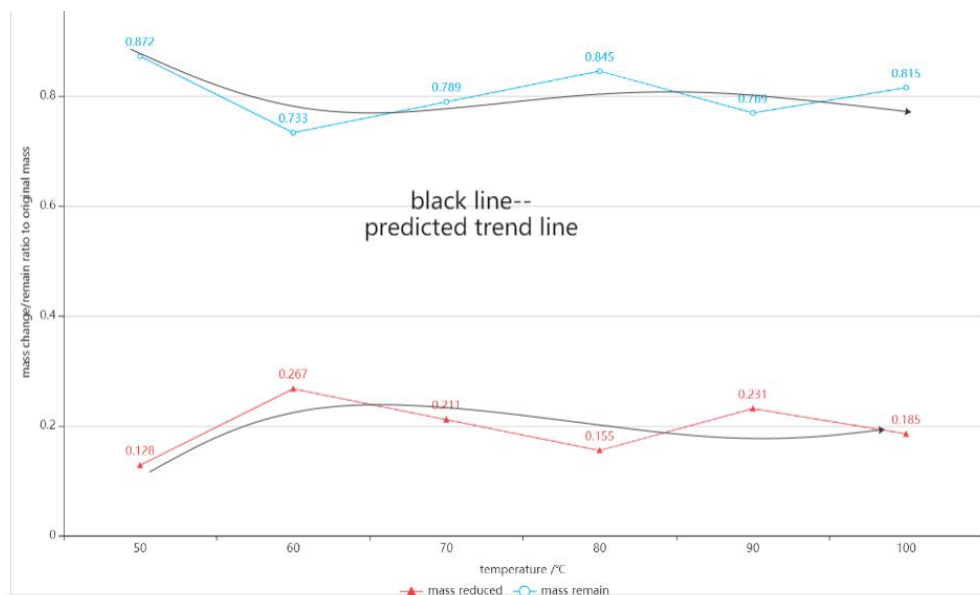


Fig 38. mass change/remain ration to original mass

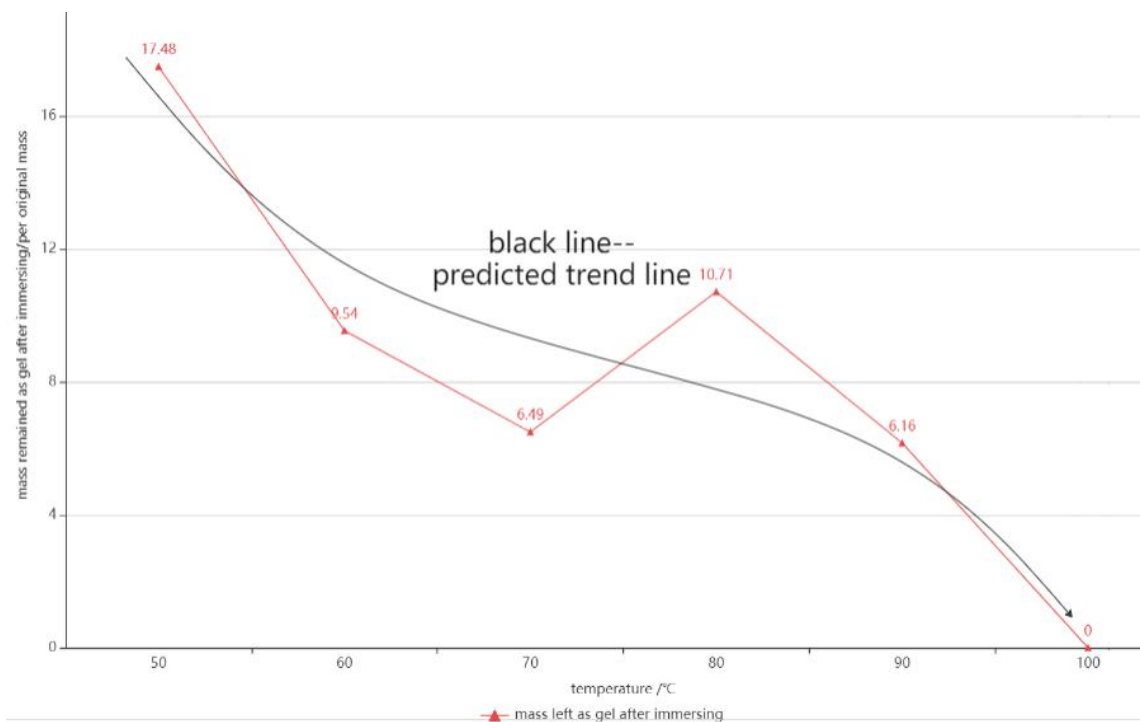


Fig 39. mass remained as gel after immersing/per original mass

Fig 38 illustrated that proportion of mass left in the membrane generally remains at the same level, but there are decreases in the remaining solid component when the sample is formed under higher temperature. Likewise, the proportion of water loss under higher temperature is larger. Fig 39 illustrated how many remains of membrane are left as gel in water because they are half dissolved (theoretically the membrane can dissolve entirely in water, but then there won't be comparison, therefore I choose a certain time before the membrane all dissolved. Unfortunately, the membrane under 100°C still completely dissolved, so the remaining mass are noted as 0 ). There is a significant trend that as the temperature to make the membrane is higher, the membrane can dissolved entirely after immersed in water overnight.

Base on these 2 conclusions, I deduce that the membrane formed under higher temperature have more combined water inside the structure of membrane, so the structure is more likely to break after water is used to dissolve the membrane structure. The reason to such structure may related to the altered function of plasticiser (glycerol), the increase kinetic energy of polysaccharide and water , the increasing Caramelization effect and etc.



### From extension of solvent-evaporation membrane

(the membrane formed by cross-linking is too rigid and show plasticity quite a lot)

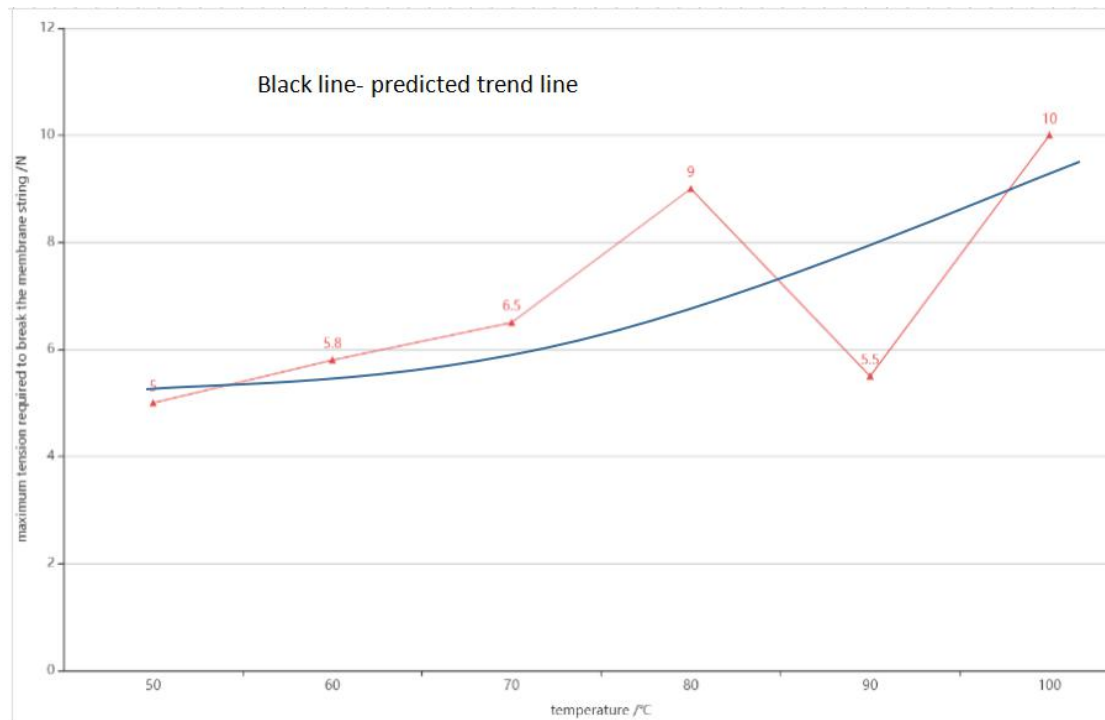


Fig 40. the maximum tension required to break the membrane string/N

For the sample made at higher temperature, generally the maximum tension required to break the membrane is larger. This shows that group with higher temperature of formation have greater stability and have a more compacted structure.

## Improvement on experiments

### 1) During making the membrane

I controlled the amount of ingredients used and size of Petri dish used. However, there are still many factors that I failed to control.

#### For cross-linking

I failed to control the temperature of reaction precisely. Initially I planned to heat copper sulphate solution up to the wanted temperature using the incubator. However, I sooner found that the incubator cannot heat the copper sulphate to the desired temperature precisely and require quite a lot of time (probably due to the feature of solution). Therefore, to ensure we can heat the polysaccharide mixture evenly, I decided to use water bath, which is I put water inside the incubator and let the Petri dish float on the water. Afterwards, I add copper sulphate solution drop by drop from the center of Petri dish until the membrane is entirely immersed in the copper sulphate solution. But this process is also not that precise because the temperature of water bath and cross-linking agent (copper sulphate) are not precisely controlled. Especially for the experiment conducted under 80°C when I just finished trying to heat the copper sulphate to have a solution bath, I directly used the cross-linking agent under 60°C, which is significantly higher than other groups with cross-linking agent under 30°C.

I failed to control the surface area and thickness of the membrane. As I the cross-linking membrane are immersed in copper sulphate, the membrane bend and traps some polysaccharide mixture inside. Therefore, not all reagents sufficiently reacted with cross-linking agent and the surface area of membrane is also distorted. Besides I set the time to immerse the membrane in cross-linking agent for 20 minutes, but for those reagents which already trapped in membrane such time is not sufficient for the membrane to be a solid structure. Instead, if we cut the 'membrane' in halves (which I did), we can find out that actually the central part of membrane is still liquid.

#### For evaporation of solvent

Basically the membrane were made quite precise because what I really need to do is just to put the polysaccharide mixture in a Petri dish and place it in the drier.

Except for group under 50°C I added one hour more because under 50°C there are no membrane formed, so in order to test the properties I choose to prolong time for heating.

### 2) During putting the membrane in water and alcohol

In this step, generally the uncontrollable variable are greater.

#### For cross-linking

When the 'membrane' is ready, I use scissors to cut it in halves to immerse in water and alcohol. However, the polysaccharide mixture in the center of membrane are still liquid. Therefore, I quickly immerse those membrane (more like a sac with liquid) into room temperature cross-linking agent and allow copper sulphate to seal the liquid quickly. This measure might influence quite a few property of our product, but generally the sealing process happen under similar conditions, so the variable is sort of controlled.

I failed to control the time of immersing in water and alcohol, which have quite severe influences.

The error occurs because it takes sometime to make the membrane, and generally I just immediately immerse the already-made membrane into water or ethanol right-away, so there will be certain time differences in immersing, especially for those groups made in days later than the first group. Although I used to measure the wrong data, after realizing the time issue I added an additional day to those samples which required more time immersing and got a nice trend line.

I also failed to control the pure absorption of water because after I finished the experiment I found out that ethanol will act as a stabilizer in the membrane and get in the structure of membrane to stiffen it. This actually explains why the colour of membrane after immersing in alcohol tends to be lighter and opaque. (At first I thought this is mainly because ethanol washes some copper ions in the membrane, but then I found out that transparent polysaccharide mixture placed in ethanol will become white solid. After some investigation through internet I changed my hypothesis.) Besides, I didn't control the concentration of ethanol used (changed from 75% to 99%). Therefore, this may be considered to be a change in how stabilizer influences the membrane structure. Since the amount of ethanol is far more than sufficient and the amount of ethanol that reacts with membrane is not very significant, I choose to neglect those 2 variables.

For solvent-evaporating membrane, I guess ethanol didn't manage to be an effective stabilizer because the colour of membrane didn't change much after immersing in alcohol.

### 3) During testing properties

I mainly weigh the products before and after immersing, so the main error on the figures are caused by the balance. However, there are some errors present, especially for the test on ability of dissolving in water because it is impossible to get all gel out, so there has to be some missing gel left. Apart from that, the measure of tension might not be precise because I can only observe the correct reading for one or two seconds, so the readings have parallax error.

## Conclusion & hypothesis

### **For cross-linking membrane:**

The amount of mass dissolved in water under high temperature is more.

Thicker membrane tend to dissolve less (not for sure).

The group with higher temperature tend to have higher amount of water loss.

In all membrane the proportion of polysaccharide and combined water by mass is mainly the same.

Total mass decreased under higher temperature is less in general.

For membrane made under higher temperature, water reduce more when immersed in alcohol.

### Hypothesis:

After we immerse the membrane in water for a long time, the membrane will eventually dissolve or exist with similar structures. The reason why membrane structure varies under different temperature because the distributions and interactions of polysaccharides, glycerol and water are different. However, this temporary structure is just a result of equilibrium under these reactants. With sufficient water molecules to shift the equilibrium, the membrane can be disassembled eventually. If it does not entirely disassembled, when the new equilibrium is reached the membrane will also show same structure and properties. Therefore, the temperature of cross-linking membrane during membrane formation mainly contributes to the temporary structure, and with proper conditions this structure can be altered.

### **For solvent-evaporation membrane:**

Membrane made at higher temperature tend to have remain less mass when immersed in alcohol.

Membrane made at higher temperature tend to dissolve more in water.

Membrane made at higher temperature tend to have better mechanical strength.

### Hypothesis:

There are more combined water in the structure of membrane formed at higher temperature, so its mass always change significantly higher than membrane in other temperature.

## Mechanism of membrane formation

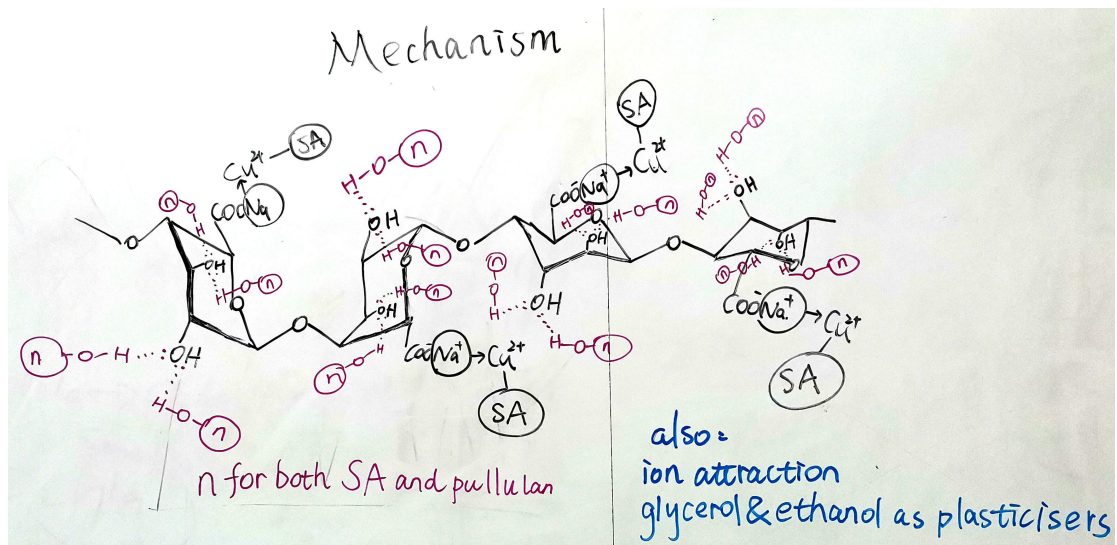


Fig 41. illustration of mechanism of SA and pullulan membrane

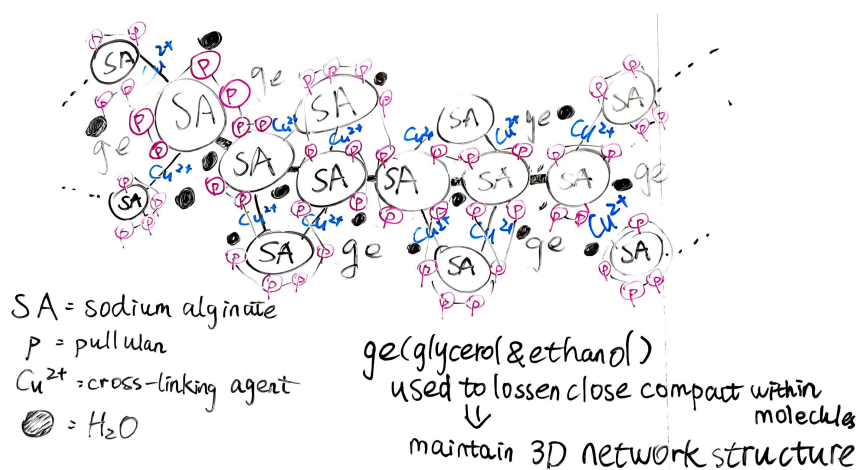


Fig 42. illustration of membrane structure with smaller amount of combine water (lower temp)

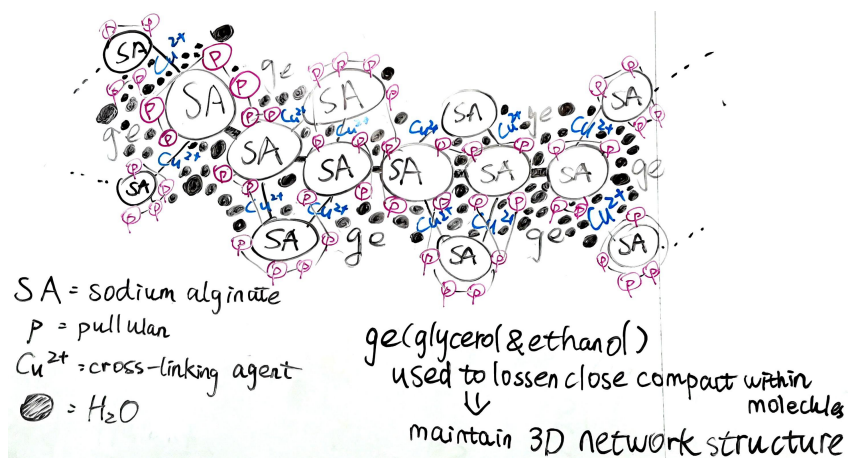


Fig 43. illustration of membrane structure with higher amount of combine water (higher temp)



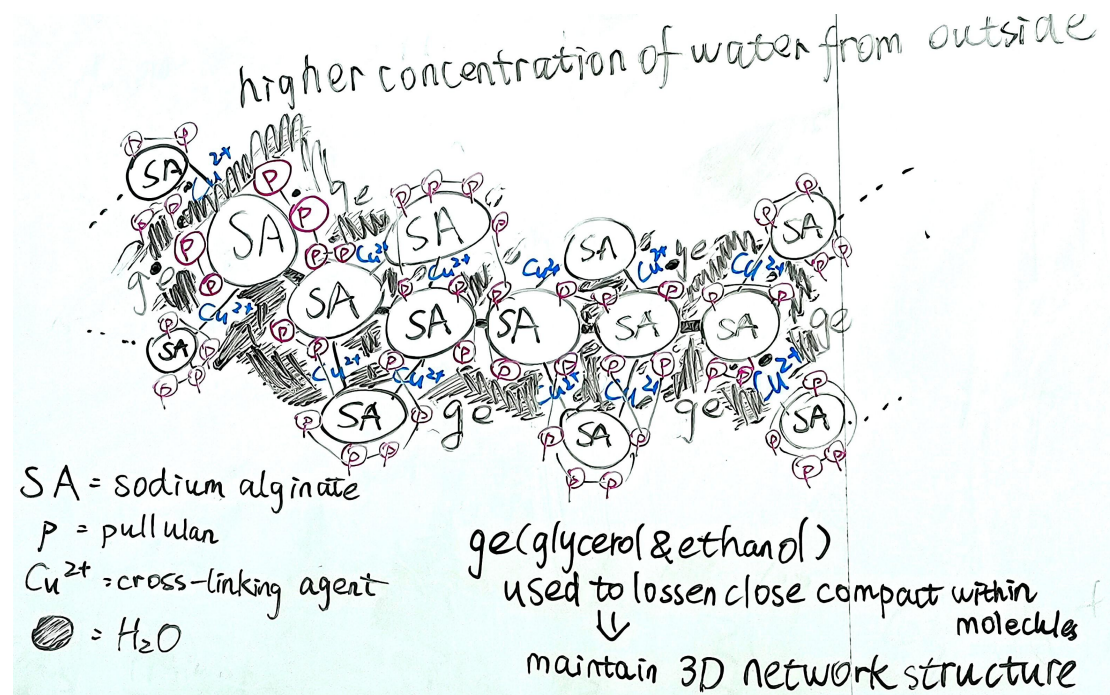


Fig 44. illustration of membrane structure when cross-linking membrane are immersed in water (black area suggest water in the membrane structure)

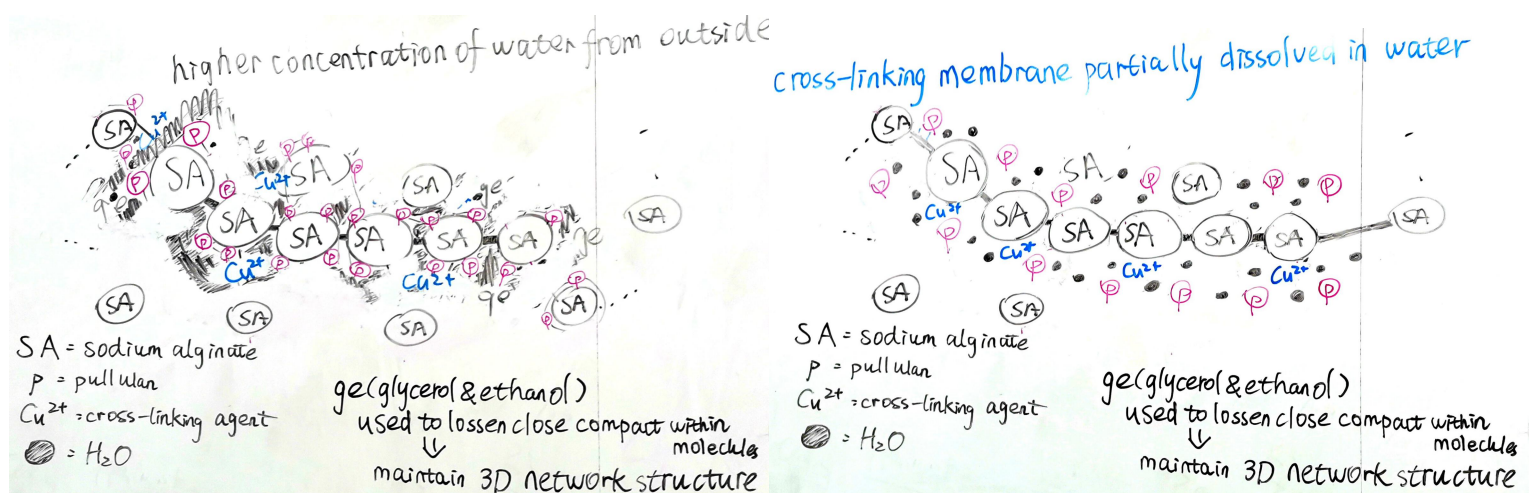


Fig 45&46. illustration of membrane structure when cross-linking membrane are immersed in water (mainly focus on how water break the structure and dissolve the packed membrane)