DATE: 2017.03.23

# Aim:

Improve the quality of reprocessed paper made in laboratory (thinner and more smooth)

# Materials and Methods:

printed paper, water

- 1. Use blender to break and mix paper and water for 5 min  $\rightarrow$  4%, 2%, 1%, 0.5% pulp
- 2. Respectively take 30 g 4% pulp / 25 g 2% pulp / 50 g 1% pulp / 100 g 0.5% pulp, put into a Büchner funnel which connected to a side-arm flask with a tube leading to a vacuum pump
- 3. Pumping for 1 min
- 4. Take out the intertwined paper fibers and put into oven to bake for 20 min

# Result:

pulp	4%	2%	1%	0.5%
dry paper (g)	4	4	2	1
water (mL)	96	196	198	199
pulp	uneven, big	big	even, small	even, no fiber
appearance	fiber particles		fiber particles	particles
pulp amount	30	25	50	100
(g)				
Expected fiber	1.2	0.5	0.5	0.5
amount				
result	4.65 g, rough	1.31 g, weak	0.76 g	0.46 g, dense
Recovery rate	1.163	0.328	0.382	0.46

# **Conclusion**:

Reprocess paper made from diluted pulp has higher recovery rate and quality. All of the paper were wrinkle, we are going to try to exclude the problem by change the baking temperature. <u>DATE</u>: 2017.04.11

# Aim:

Improve the quality of reprocessed paper made in laboratory (thinner and more smooth)

# Materials and Methods:

printed paper, water

- 1. Use blender to break and mix paper and water for 3 min  $\rightarrow$  0.3%, 0.1% pulp
- 2. Respectively take 30 g 4% pulp / 25 g 2% pulp / 50 g 1% pulp / 100 g 0.5% pulp, put into a Büchner funnel which connected to a side-arm flask with a tube leading to a vacuum pump
- 3. Pumping for 1 min
- 4. Take out the intertwined paper fibers and put into oven to bake for 20 min

# **Conclusion:**

Try dilute pulp next time and fix the amount of the fiber

Test the effects of different condition and establish a stable flotation process

# Introduction:

We add detergent and Sodium hydroxide in pulp and use a pump to make bubbles. Because the ink can attach on the lipophilic side of bubbles, the ink will be dispersed from fiber and float to the surface with bubbles. The function of Sodium hydroxide is to make an alkaline environment to enhance the surface tension of the bubbles. Flotation is the one of important process to take out of the ink from fibers, so we test the amount of times adding detergent and the different doses of detergent during flotation process. Then we compare the amount of ink remaining on the paper and the weight of paper (fiber loss) in different condition.

DATE: 2017.05.16

Purpose: Test the flotation condition.

# Materials:

0.3% pulp, detergent, sodium hydroxide, bubble pump, Büchner funnel, side-arm flask, filter paper

# Methods:

- 1. Add NaOH 0.1ml (1M), detergent 3ml in 600ml paper pulp (0.3%)
- 2. Float for 20 minutes.
- 3. Use Büchner funnel and vacuum suction, filter paper to make paper.

## Result:

The first time we test flotation condition, we found that too many bubbles were produced, and so as to cause lots of fibers loss. In addition, it was difficult to separate the filter paper from the filter paper after drying.

DATE: 2017.06.26

Purpose: Float the pulp in different time and compare the paper loss.

# Materials:

0.3% pulp, detergent, sodium hydroxide, bubble pump, Büchner funnel, side-arm flask, filter paper, wire-netting

#### Methods:

1. Add NaOH 2.7 ml (0.1M), detergent 3 ml in 900 ml paper pulp

- 2. Float for 10 minutes and 15 minutes.
- 3. Make a control group that added the same doses of detergent and NaOH but didn't go through flotation.
- 4. Use Büchner funnel, vacuum suction, filter paper and wire-netting to make paper.
- 5. Observe the loss rate of the fiber after flotation

Result: In order to avoid the paper tear from the filter paper inconvenience, we used a kind of wire-netting as filter. Then we referred to Dr. Su's paper and modified the doses of NaOH, compared flotation which lasted at different time. Though we could distinguish the different from pulp going through flotation or not, we could not observe the significant difference from pulp floating for the different time.



• <u>DATE</u>: 2017.07.01

<u>Purpose</u>: Observe whether enzyme reaction affects flotation.

# Materials:

0.3% pulp, detergent, sodium hydroxide, bubble pump, Büchner funnel, side-arm flask, filter paper, wire-netting

# Methods:

- 1. Heat the pulp to 60°C
- 2. Add Xylanase 1.44ml(3.6U) to react for 3 minutes

- 3. Heat the other pulp without enzyme
- 4. Added NaOH 1.8 ml (0.1M), detergent 15ml (10X dilute)
- 5. Float for 20 minutes.
- 6. Use Büchner funnel, vacuum suction, filter paper and wire-netting to make paper.
- 7. Analyze the black level of paper to observe whether enzyme reaction affects flotation.

# Result:

We bought commercial enzyme, Xylanase and Lipase. Based on Dr. Su's paper, we heated the pulp to 60°C to make enzyme react at high efficiency. But we found that was difficult to make bubbles if we added detergent when we had not cooled down yet.

# • DATE: 2017.07.02

<u>Purpose:</u> Observe whether enzyme reaction affects flotation.

# Materials:

0.3% pulp, detergent, sodium hydroxide, bubble pump, Büchner funnel, side-arm flask, filter paper, wire-netting

### Methods:

- 1. Heated the pulp to 60°C
- 2. Added Xylanase 1.44ml(3.6U) to react for 3 minutes
- 3. Heat the other pulp without enzyme.
- 4. Add NaOH 1.8 ml (0.1M) and cool down the pulp.
- 5. Add detergent 15ml (10X dilute) and floated for 20 minutes.
- 6. Use Büchner funnel, vacuum suction, filter paper and wire-netting to make paper.
- 7. Analyze the black level of paper to compare the effect of enzyme.

# Result:

We found that was better to make bubbles if we added detergent when we cooled down pulp first.

#### DATE: 2017.08.28

Purpose :Observe the effect that we disperse detergent adding in 4 times.

#### Materials:

0.3% pulp, detergent, sodium hydroxide, bubble pump, Büchner funnel, side-arm flask, filter paper, wire-netting

# Methods:

- 1. Heat the pulp to 60°C
- 2. Added Xylanase 1.44ml(3.6U), Lipase 1.44ml(3.6U) to react for 60 minutes
- 3. Heat to boiling when the time came up
- 4. Add NaOH 1.8 ml (0.1M) and cool down the pulp.
- 5. Add the detergent 0.3ml (no dilute) four times (for 5 minutes one time).
- 6. Blend the pulp during flotation.
- 7. Use Büchner funnel, vacuum suction, filter paper and wire-netting to make paper.
- 8. Analyze the black level of paper

### Result:

In order to improve the accuracy of the experiment, we boiled pulp to stop the reaction of the enzyme. It was found that the fiber gathered easily after boiling, so after that we added NaOH 1.8 ml in one cup of 600ml pulp when the time came up and waited for cooling down. We could observe the significant difference to distinguish whether added the enzyme. So we used this protocol to continue the experiment

## Conclusion:

At the beginning, the data of retaining ink analysis had been a great variable. So we had to improve the flotation process before analyze amount of retaining ink to establish a data base.

During flotation condition testing, we thought that was not a stable way to make bubbles if we added all detergent at the beginning. Because at the beginning flotation strength was strong enough, but it would be weaker and weaker, even so weak that there was no bubble to float out of the cup. So we dispersed adding detergent into four times in flotation process, and blended the pulp slightly to prevent the fibers from accumulating at the bottom.

# Introduction

In order to speculate the efficiency of enzymatic deinking, we established the recycled paper process without enzymes in lab, first. Then, we compare the process we made with or without enzymes to confirm that the process with enzyme is better than one without enzymes. Our process can be briefly described in six parts, paper pulping, deinking, flotation and paper making. The enzymes, Xylanase, Glucanase and Lipase are added in deinking part, which is the key part to help the ink dropped off from paper. To determine our efficiency of the process with enzymes, we use same amount of commercial enzymes to react on the paper pulp made by the paper printed with the black surface area in the following ratio, 100%, 75%, 50% and 25%. After the reprocessed paper is made, we analyzed the gray scale of reprocessed paper to check the efficiency of our method with commercial enzymes. As the results, the reprocessed paper made with enzymes is brighter than the one without enzymes. The reprocessed paper made from the higher black surface area had greater gray scale between control and treatment group

#### Date

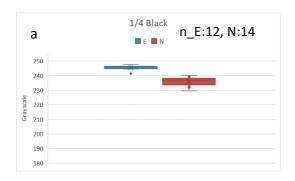
9/13~10/17

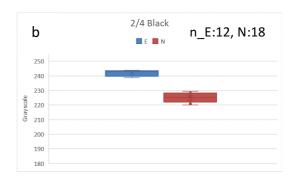
# Material and method

- 1. Make 600 mL 0.3% paper pulp made by the paper printed with the black surface area in the following ratio, 100%, 75%, 50% and 25% by blender for 3min and 30sec and pour in a beaker.
- 2. Heat the pulp to 60 °C and add Xylanase, Lipase, Glucanase(108U)
- 3. Reaction for 1 hr.
- 4. Cool down and add 1.8 mL NaOH and 0.3 mL detergent to the pulp.
- 5. Put a bubble stone with pump in the beaker and start flotation for 20 min, add 0.3 mL detergent each 5 min.
- 6. Discard the bubble flow through and fibers adhere to the beaker.
- 7. Use the deinked pulp to make reprocessed paper and scan it.
- 8. Measure the grayscale.

# Result

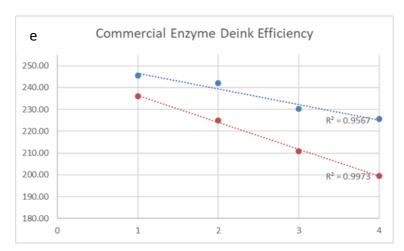
The reprocessed paper made with enzymes is brighter than the one without enzymes. The reprocessed paper made from the higher black surface area had greater grayscale between control and treatment group.











E	1/4 Black(12)	2/4 Black(10)	3/4 Black(12)	Black(12)
Mean	245.64	241.94	230.24	225.64
SD	1.59	1.85	4.05	2.22
ink	7.02	12.62	30.33	37.30
deink(%)	14.68	25.58	29.41	39.47
N	1/4 Black(14)	2/4 Black(18)	3/4 Black(21)	Black(18)
Mean	235.94	225.04	210.80	199.56
SD	2.90	3.17	1.48	7.66
ink	21.71	38.19	59.75	76.76

# Discussion

First, we added commercial enzymes to react on paper pulp with different black level to test enzyme effect. As the result, we can confirm that commercial enzymes have significant deinking effect. The linear relationship on e can be further explained that commercial enzyme can work on the paper printed with different amount of ink.

# Ink particles analysis

# Introduction

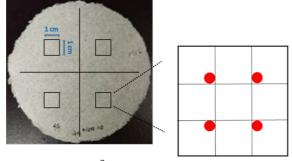
To distinguish whether the enzyme deinking method we used effective or not, we develop an Ink particles analysis assay. The Ink particles analysis assay was used on reprocess papers we made in laboratory. Analysis the amount of ink particles remain on reprocess papers and infer enzyme deinking efficiency.

#### The Ink

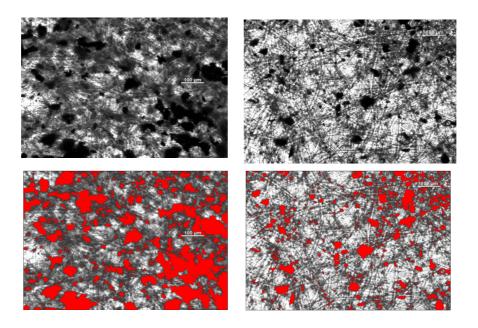
Date: 2017.05.15~2017.06
Materials and Methods:

reprocessed papers, slides, nail polish, microscope

- 1. Make reprocessed paper
- 2. Cut 4 pieces of 1 cm<sup>2</sup> paper from the reprocessed paper



- 3. Mounting the 1 cm² reprocessed paper on the slide with nail polish, air-dry the slide (補 slide 圖)
- 4. Observe Red areas of slide with 100X magnified by microscope and take photo
- 5. Analysis the area of ink particles on the photo by ImageJ Result :



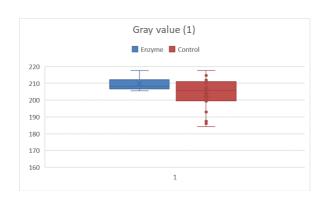
• Date: 2017.07,14~

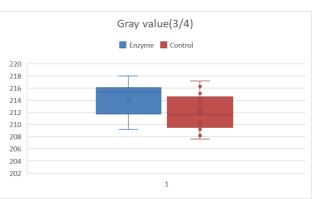
# Materials and Methods:

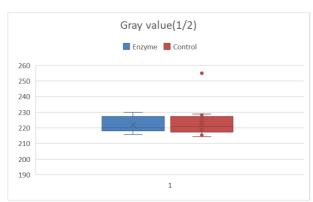
reprocessed papers, scanner

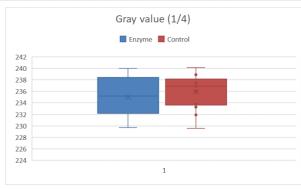
- 1. Make reprocessed paper
- 2. Scan the paper at 8-bit gray value, 4800 dpi
- 3. Analysis the average gray-scale value by ImageJ

# Result:









# Conclusion:

We first use microscope to observe the ink particles,

# Yeast secreted enzyme deink assay

Date: 2017.10.29

# Purpose:

To test whether the enzyme yeast produced has the ability to deink.

#### Material:

- paper pulp
- enzymes(xylanase/ glucanase/ lipase) produced by yeast

#### Procedure:

- 9. Make 600 mL 0.3% pulp by blender for 3min and 30sec and pour in a beaker.
- 10. Heat the pulp to 30 °C and add 1 mL enzymes that had concentrated by Amicon® Ultra-15 centrifugal filter(induced/ non-induced yeast medium)
- 11. Reaction for 1 hr.
- 12. Add 1.8 mL NaOH and 0.3 mL detergent to the pulp.
- 13. Put a bubble stone with pump in the beaker and start flotation for 20 min, add 0.3 mL detergent each 5 min.
- 14. Discard the bubble flow through and fibers adhere to the beaker.
- 15. Use the deinked pulp to make reprocessed paper and scan it.
- 16. Measure the gray value.

## Result:

	Induced	Non-induced	PBS
Mean	243.34	237.82	235.83
SD	3.04	0.42	1.05
ink	10.50	18.85	21.87
deink(%)	11.37	3.02	0.00
recycle rate	0.47	0.50	0.54

	PBS					
			dry weight (g)	pulp (g)	recycle rate	
1	20171029;allb;PBS_2ml026.jpg:07182-03534	234.32	0.29	170.00	0.57	
2	20171029;allb;PBS_2ml026.jpg:11566-03694	236.91	0.31	180.00	0.57	
3	20171029;allb;PBS_2ml026.jpg:11238-08006	236.17	0.31	180.00	0.57	
4	20171029;allb;PBS_2ml027.jpg:07182-03534	236.32	0.28	180.00	0.53	
5	20171029;allb;PBS_2ml027.jpg:11566-03694	234.71	0.26	170.00	0.51	
6	20171029;allb;PBS_2ml027.jpg:11238-08006	236.54	0.25	170.00	0.50	
	Mean	235.83			0.54	
	SD	1.05			0.03	

	Induced				
			dry weight (g)	pulp (g)	recycle rate
1	20171029;allb;Y_Xyl_lip_induced_1ml021.jpg:07	238.40	0.25	170.00	0.48
2	20171029;allb;Y_Xyl_lip_induced_1ml021.jpg:11	245.25	0.21	170.00	0.41
3	20171029;allb;Y_Xyl_lip_induced_1ml021.jpg:10	240.73	0.28	190.00	0.49
4	20171029;allb;Y_Xyl_lip_induced_1ml021.jpg:11	247.26	0.23	170.00	0.44
5	20171029;allb;Y_Xyl_lip_induced_1ml022.jpg:07	242.02	0.28	180.00	0.51
6	20171029;allb;Y_Xyl_lip_induced_1ml022.jpg:11	242.13	0.28	180.00	0.51
7	20171029;allb;Y_Xyl_lip_induced_1ml022.jpg:10	246.56	0.25	180.00	0.46
8	20171029;allb;Y_Xyl_lip_induced_1ml022.jpg:11	241.48	0.28	180.00	0.51
9	20171029;allb;Y_Xyl_lip_induced_1ml023.jpg:07	245.34	0.23	170.00	0.45
10	20171029;allb;Y_Xyl_lip_induced_1ml023.jpg:11	239.16	0.26	180.00	0.47
11	20171029;allb;Y_Xyl_lip_induced_1ml023.jpg:10	246.02	0.23	170.00	0.45
12	20171029;allb;Y_Xyl_lip_induced_1ml023.jpg:11	245.74	0.25	180.00	0.46
	Mean	243.34			0.47
	SD	3.04			0.03
	Non-induced				
			dry weight (g)	pulp (g)	recycle rate
1	20171029;allb;Y_Xyl_lip_noninduced_1ml024.jpg	238.38	0.27	180.00	0.49
2	20171029;allb;Y_Xyl_lip_noninduced_1ml024.jpg	237.92	0.28	180.00	0.51
3	20171029;allb;Y_Xyl_lip_noninduced_1ml024.jpg	237.19	0.28	180.00	0.52
4	20171029;allb;Y_Xyl_lip_noninduced_1ml025.jpg	238.11	0.27	180.00	0.49
5	20171029;allb;Y_Xyl_lip_noninduced_1ml025.jpg	237.74	0.27	180.00	0.50
6	20171029;allb;Y_Xyl_lip_noninduced_1ml025.jpg	237.59	0.27	180.00	0.51
	Mean	237.82			0.50
	SD	0.42			0.01

#### Introduction

It's necessary to confirm that the paper deinking process of device which we designed can work, so we deinked the paper first before paper pulping. After flotation, paper making, we analyzed the gray value of reprocessed paper to determent the efficiency of the enzyme that reacted on the paper fibers and inks.

#### Date

2017/10/20, 2017/10/23, 2017/10/24, 2017/10/25

#### Material and method

Beaker, iron plate, plastic wrap, pump, bubble stone, sodium hydroxide, Büchner funnel, iron net, filter paper, oven, enzyme (Xylanase, Glucanase, Lipase)

- 1. Mix the commercial enzyme into a solution (0.8U/ml)
- 2. Pass the papers (3.6g) through the solution and placed in a 60 degree oven for 4 minutes
- 3. Blend the paper into pulp for 3.5 minutes in pH6 water
- 4. Add sodium hydroxide (1.8ml, 1M) and detergent( four times, 0.3ml per 5 minutes)
- 5. Put the pump and make bubbles to float the paper pulp.
- 6. Use Büchner funnel, iron net, filter paper to make paper
- 7. Put into the oven.

	E	N
Mean	250.13	220.84
SD	0.49	2.147
ink	0.22	44.56
recycle rate	0.52	0.40
deink(%)	44.34	

# deinking before paper pulping and going through the others process

We mixed the commercial enzyme into a solution. The papers were soaked by the solution within three seconds and placed in a 60 degree oven for 4 minutes. Going through paper pulping, flotation and paper making, also, we analyzed the gray value of paper.

# Different concentration enzyme deink assay

# Purpose:

Test different concentration of commercial enzyme

#### Material:

- paper pulp
- enzymes(xylanase/ glucanase/ lipase) (品牌、貨號)

# Procedure:

- 17. Make 600 mL 0.3% pulp by blender for 3min and 30sec and pour in a beaker.
- 18. Heat the pulp to 60 °C and add enzymes as table1.
- 19. Reaction for 1 hr.
- 20. Add 1.8 mL NaOH and 0.3 mL detergent to the pulp.
- 21. Put a bubble stone with pump in the beaker and start flotation for 20 min.
- 22. Add 0.3 mL detergent each 5 min.
- 23. Discard the bubble flow through and fibers adhere to the beaker.
- 24. Use the deinked pulp to make reprocessed paper and scan it.
- 25. Measure the gray value.

Table 1