

# PROCEEDINGS BOOK THE 7<sup>TH</sup> ANNUAL BASIC SCIENCE INTERNATIONAL CONFERENCE

**7-8 March 2017**

Ijen Suites Resort and Convention  
Malang, Indonesia

**Basic Science for Improving  
Survival & Quality of Life**

**Sub Topics:**

**Botany**

**Environmental Science and Technology**

**Instrumentation and Measurement**



**Faculty of Science  
Brawijaya University**



## **The 7<sup>th</sup> Basic Science International Conference**

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**Basics Science for Improving Survival and Quality of Life**

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# **Proceedings Book**

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## ABOUT BASIC

The Annual Basic Science International Conference is a scientific meeting aimed to promote mutual exchange between scientists and also experts, to discuss innovative ideas in scientific research, and to tackle contemporary problems through the application of knowledge that rise from sciences. The scope of this conference is fundamental and applied research in chemistry, biology, physics, and mathematics. The origin of this conference was initiated in year 2000, by the Faculty of Mathematics and Natural Sciences of Brawijaya University, under the name of Seminar Nasional Kemipaan (National Sciences Conference). Since then, the conference has been organized regularly on annual basis. In 2004, the conference changed its name to Basic Sciences Seminar (BSS) and started to invite international speakers and participants. The conference then expands its scope to international in 2011 and formally adopting the current name. The previous Basic Sciences International Conference was held at Atria Hotel Malang in 2016 with participants from many countries including Australia, Malaysia, Thailand, Japan, UK and Germany.



## WELCOME MESSAGE

On behalf of the organizing committee, I would like to welcome you to the 7<sup>th</sup> Annual Basic Science International Conference.

Firstly, I would like to thank all participants who have spent their time to come and join us for the conference. I believe that we will not be able to hold this conference successfully without participation from all of you. Secondly, I would like to thank the dean of faculty of Mathematics and Natural Sciences, Brawijaya University, because the faculty has provided us supports and facilities. I am thankful to our great keynote and invited speakers for their willingness to join the conference and share their scientific knowledge to all of us. Thanks to our reviewers who have made assessments and suggestions related to the abstracts. I also want to thank the sponsors which have made their contributions to this conference. Finally, I want to thank all members of the committee for their hard work to make this conference successful.

The Basic Science International Conference is held every year since 2010, and always organized by the Faculty of Mathematics and Natural Sciences, Brawijaya University. This conference is a forum that enables us to share our ideas among us. The participants are expected also to take their time and opportunities to know each other during the conference, in order to strengthen their networks and collaborations. In this conference, we have more than 300 participants from counties such as Indonesia, Japan, Australia, Germany, Switzerland, and Thailand. In the conference, we have plenary lectures and sessions for parallel oral presentations as well as poster presentations.

We hope that all participants enjoy all activities during the conference and this proceedings book will be useful for all of us.

Thank you very much.

Best regards,

Hari Arief Dharmawan, Ph.D.

Chairman of BaSIC 2017

## WELCOME MESSAGE

On behalf of the Dean of Faculty of Mathematics and Natural Sciences, Brawijaya University, I would like to extend my warmest welcome to all delegates from all over the world. Welcome to Malang, where Malang is one of the educational city in Indonesia. Malang, which is about more than 400 meters above sea level, has many tourist destinations. Malang is like a bowl, surrounded by some volcanoes in the east (Semeru and Bromo), west (Kawi and Kelud) and north (Arjuna and Welirang Complex), and in the south are coastal areas, where we have many beautiful new opening beaches.

We are very pleased to welcome you in the proceedings book of the seventh Annual Basic Science International Conference 2017. I would like to express my gratitude to all of the participants, keynote and invited speakers as well. Many thanks also go to the reviewers and the editorial team for their big effort in supporting this book of abstracts. Last but not least my big appreciation to the steering and organizing committees, in realizing this proceedings book.

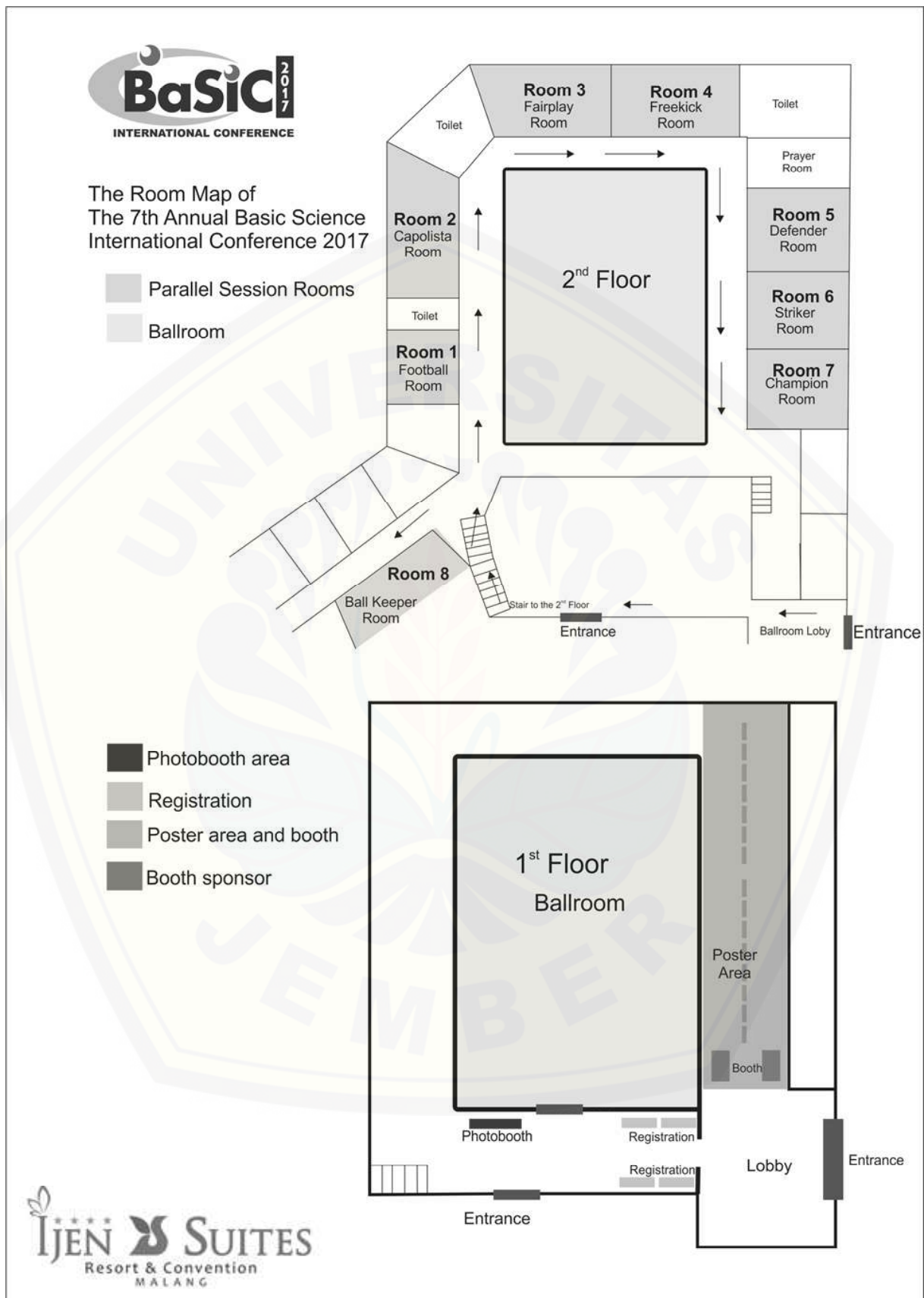
Thank you.

Faculty of Mathematics and Natural Sciences,

Dean,

Adi Susilo, Ph.D.

## CONFERENCE VENUE



## CONFERENCE PROGRAM

### Day One: March 7<sup>th</sup>, 2017

07.30 – 08.30	Registration
08.30 – 09.00	Opening Ceremony
09.00 – 09.45	<b>Plenary Lecture 1:</b> <i>CRISPR/Cas9: Basics and Applications in "gene surgery".</i> Prof. Dr. Wolfgang Nellen, Institut fur biology, Germany
09.45 – 10.00	Coffee Break
10.00 – 10.45	<b>Plenary Lecture 2:</b> <i>Use of Wavelet Analyses with Potential Field Data in Exploration and Monitoring Studies</i> Dr. Guillaume Mauri, Neuchatel University, Switzerland
10.50 – 11.35	<b>Plenary Lecture 3:</b> <i>Mathematics for Solving 5G Massive Wireless IoT Networks Problems</i> Dr. Eng. Khoirul Anwar, S. T., M. Eng., Telkom University
11.35 – 12.30	Lunch
12.30 – 15.00	<b>Parallel Session 1</b>
15.00 – 15.30	Poster Session & Coffee Break
15.30 – 17.30	<b>Parallel Session 2</b>
17.30 – 19.00	Breaks
19.00 – 21.00	Gala Dinner

### Day Two: March 8<sup>th</sup>, 2017

07.30 – 08.10	Registration
08.10 – 08.55	<b>Plenary Lecture 4:</b> <i>The Roles of Metal Ions in Diabetes – Metal Drugs and Supplements</i> Prof. Peter Andrew Lay, Sydney University, Australia
09.00 – 09.45	<b>Plenary Lecture 5:</b> <i>Functionalization of Stainless Steels Via Low Temperature Plasma Nitriding</i> Prof. Tatsuhiko Aizawa, Shibaura Institute of Technology (SIT), Japan
09.45 – 10.00	Coffee Break
10.00 – 12.00	<b>Parallel Session 3</b>
12.00 – 13.00	Lunch
13.00 – 14.30	<b>Parallel Session 4</b>
14.30 – 15.00	Coffee Break
15:00 – 16.00	<b>Parallel Session 5</b>
16.00 – 16.30	Closing Ceremony & Award Announcement

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**Basics Science for Improving Survival and Quality of Life**



**Plenary Lectures**

# Examination of Coffee Pulp Waste for Medium in Cellulase Production by *Aspergillus Species*

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**Abstract** – An isolate, identified as *Aspergillus* species can grow well by introducing coffee pulp medium as carbon and nitrogen source in solid state fermentation without any nutrient added. This genus optimum produced cellulase in five days of fermentation at 37°C. Purification, gave the yield 0.13% and 477 fold. The enzyme stable at pH 3.5 to 6.5 and below 55°C, while the optimum activity at pH 5 and 50°C respectively. It is suggested that coffee pulp waste can be used as a cheap medium for cellulase production.

## 1. INTRODUCTION

Indonesia as the world's fourth largest exporter of coffee, so necessary to adopt a careful control and integrated waste either in the process of coffee production. In the coffee processing, 45% of the polysaccharides-rich coffee pulp were produced. In Indonesia, a huge amount of coffee pulp (CP) and estimated at least 81 million tons were released annually. Research proved that CP waste can be utilized as a material for bioethanol [1] [2]. It was reported that CP can be utilized as substrate in production of protease, xylanase, and endoglucanase production [3] [4] [5]. Other investigations, CP can be modified as absorbent, activated carbon [6] [7], and another industrial purpose as well [8] [9]. However, microbial research utilization of these potential wastes is still limited and less attention. A promising strategy through microbial utilization of CP to produce enzyme cellulase is one of environmentally friendly and possibly to increase the economic value of this biomass.

## 2. METHODS

### 1.1 Optimizing and Harvesting of Crude Cellulase Production of *Aspergillus* sp. through Solid State Fermentation of Coffee Pulp

Ten gram of water saturated CP in 500 mL flask was sterilized, inoculated with *Aspergillus* sp., and then incubated at 30°C. To optimize cellulase production, the activity crude cellulase was daily examined. To harvest crude cellulase was done by 1% NaCl extraction on 200 ml water, containing 0.01% natrium azide (v/v), followed by shaking 120rpm at room temperature for 12 hours. To recover crude cellulase from suspension, filtration using 40µm glass filter was done. Then to remove remaining cells or debris from crude cellulase filtrate, the centrifugation at 8000rpm for 10 minutes. The supernatant was dialyzed using PS MidiKros Filter Modules, 10 kD against 20 mM acetate buffer pH 5. Solution as a crude cellulase was then stored at 4°C till used for examination of cellulase activity. To produce large scale crude cellulase, 100gr of CP was prepared in 5L flask. And for recovery of crude cellulase, the same previous procedure steps were used.

### 1.2 Examination of Crude Cellulase Activity

The activity crude cellulase was examined by measuring of reducing sugar against 1% carboxy methyl cellulose (CMC) during hydrolysis. The CMC substrate solution was prepared in 20 mM acetate buffer pH 5. The reaction mixtures of 50µL crude cellulase and 100µL substrate were homogenized and incubated at 37°C for an hour. Quantification of reducing sugar released, the method of Somogy-Nelson was employed [10] [11].

### 1.3 Cellulase Purification

Purification was done in 25°C room temperature 20mM acetate buffer pH 5. All purification steps were described in results and discussion.

### 1.4 Effect of Temperature and pH on Cellulase Optimum Activity

Optimum Temperature (°C) and pH on purified cellulase activity were examined in a range temperature 30 to 70 °C and pH 3 to 7.5. The pH 3 to 5 and pH 5 to 7.5 acetate and phosphate buffers 20 mM were used. Optimum activity of purified enzyme was measured using Somogy-Nelson method.

### 1.5 Temperature and pH Effect on Cellulase Stability

Stability of purified cellulase activity on temperature and pH were examined in a range temperature 30 to 70°C and pH 3 to 7.5. The pH 3 to 5 and pH 5 to 7.5 acetate and phosphate buffers 20 mM were used. Stability of cellulase was measured using the same method above.

## 3. RESULTS AND DISCUSSION

Optimum production of cellulase activity was obtained after 5 days solid state fermentation at room temperature. At that time, observation showed that *Aspergillus* sp. grow rapidly even though no any nutrient added during fermentation, and some liquefy form appeared in CP medium. As the cultivation time increased, the liquefied forms also increased. Mean, *Aspergillus* sp. released some extracellular enzymes which possible hydrolysed CP actively. Some research reports that *Aspergillus* sp. secreted extracellular enzymes in board spectrum [12] [13] which capable utilized biomass as carbon and nitrogen source [8] [14] [15].

Cellulase activity by quantifying on reducing sugar production revealed that gave optimum in 5 days fermentation and no significant increasing on enzyme activity in 6-7 days fermentation as shown in Figur 1. The optimum reducing sugar production was achieved 1.4µg/ml. Base on this results, large scale fermentation was done for crude enzyme source in cellulose purification.

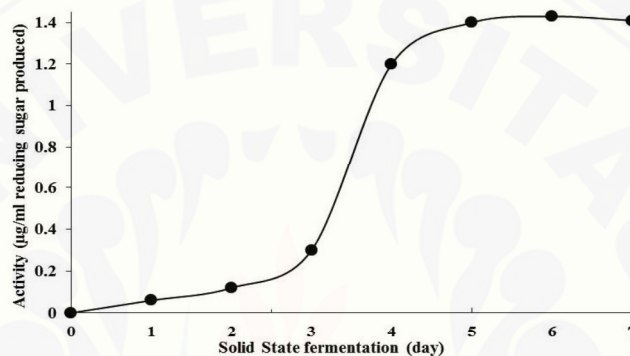


Figure 1. Optimization of Cellulase Production of *Aspergillus* sp. in Solid State Fermentation of Coffee Pulp

Purification of cellulase was started by precipitated of dialyzed crude cellulase in 65% saturated ammonium sulfate and centrifuged at 12000 rpm for 20 minutes. The precipitates were dissolved in buffer. Remaining ammonium sulfate was removed by overnight dialysis against 20mM acetate buffer at 4°C. This solution was loaded onto DEAE Toyopearl 650M anion exchanger open column pre-equilibrated with buffer and eluted with 0–0.5M NaCl linear gradient. Active fractions (Figure 2) were pooled and dialyzed against buffer to remove NaCl. As shown in Figure 2, the active peak which had cellulase activity was subsequently loaded on Pharmacia FPLC (Fast Protein Liquid Chromatography) using DEAE Cellulofine A100 column (anion exchanger). The linear gradient of NaCl from 0.1 M to 0.3 M used. Further step, the active fraction was purified on super dex 75 pg (gel chromatography).

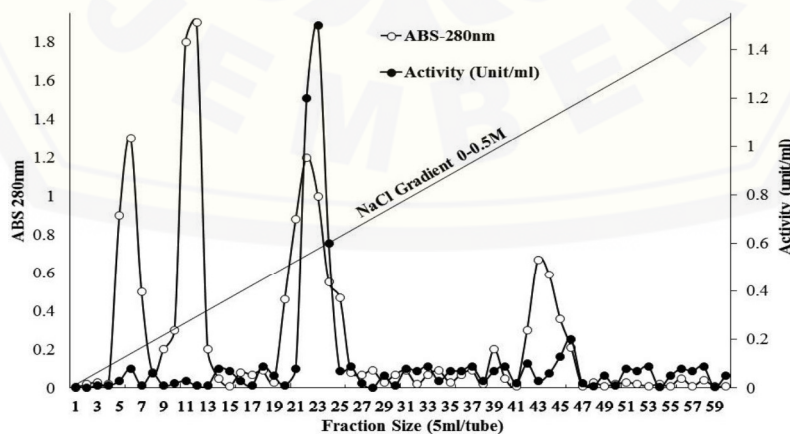


Figure 2. Purification on DEAE Toyopearl 650M column pre-equilibrated with acetate 20mM pH 5 buffer and eluted with linear gradient 0-0.5 M NaCl

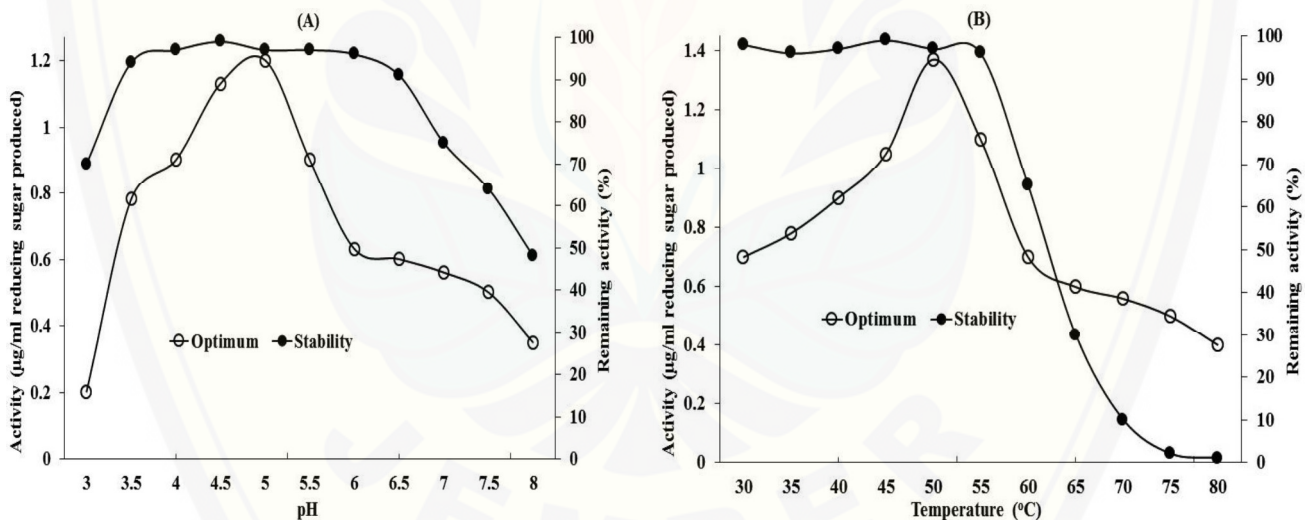
The purification procedure of cellulase summarized in Table 1 resulted in 0.13% yield, 477 fold purification. The enzyme production through solid state fermentation usually gave a good yield. However, in this study resulted in

lower yields which probably caused by optimal conditions for solid state fermentation has not fulfilled [16]. Some reports described that the production of enzymes under solid state fermentation depends on several factors. Significant increase in cellulase production influenced by increasing in inoculum concentration. Physical factors such as moisture, temperature, and pH are critical factors in solid state fermentation which influence growth, metabolism, biosynthesis and enzyme secretion as well [17] [18] [19].

**Table 1. Cellulase Purification Step**

Purification Step	Total ABS-280	Total Activity (unit)	Spec. act. (unit)/ABS280	Yield (%)	Fold
Crude enzyme	995000	16267	0.016	100	1
Ammonium sulfate Precipitation	218000	14587	0.067	21.91	4
DEAE Toyopearl 650M	94035	12997	0.138	9.45	8
DEAE Cellulofine A100	8976	11980	1.335	0.90	82
Superdex 75G	1256	9789	7.794	0.13	477

The effect of pH on the cellulase activity was measured after 1-hour incubation at 37°C of each enzyme in 1 ml 1% CMC substrate at various pH values. As shown in Figure 3 (A), purified cellulase exhibited maximum activity at pH 5 and retained nearly 100% activity in a pH range of 3-6.5 after 60 minutes exposure to corresponding pH values. The enzyme had optimum activity at 50°C and is nearly 100% stable below 55°C after 30 minutes exposure to respective temperatures shown in Figure 3 (B).



**Figure 3. Effect of pH (A) and Temperature (B) on Activity and Stability of Cellulase**

#### 4. CONCLUSIONS

Coffee pulp waste as a medium for the production of cellulase has been proven. This research despite getting a low yield, but the advantage was noted that during cellulase production via solid state fermentation no requirement the addition of any nutrient. Therefore it is necessary to do further research on the optimization of the cellulose based CP production so that the production efficiency will be increased and feasible in industrial scale.

#### 5. ACKNOWLEDGEMENTS

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