

## Optimizing the tray dryer temperature and time of white corn flour culture

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### Article history:

Received: 8 December 2020

Received in revised form: 24 January 2021

Accepted: 13 April 2021

Available Online: 19 September 2021

### Keywords:

AC culture,  
CC culture,  
RSM,  
Tray dryer,  
White corn flour

### DOI:

[https://doi.org/10.26656/fr.2017.5\(5\).718](https://doi.org/10.26656/fr.2017.5(5).718)

### Abstract

This research aimed to optimize the tray dryer temperature and time of white corn flour culture by Response Surface Methodology (RSM). There were two cultures used in this research, namely Amylolytic Culture (AC) and Complete Culture (CC). AC consisted of *Penicillium citrinum*, *Aspergillus niger*, *Acremonium strictum*, and *Candida famata*, while CC consisted of *Penicillium chrysogenum*, *Penicillium citrinum*, *Aspergillus niger*, *Rhizopus stolonifer*, *Rhizopus oryzae*, *Fusarium oxysporum*, *Acremonium strictum*, *Candida famata*, *Kodamaea ohmeri* and *Candida krusei/incospicua*. The independent variables in this study were drying temperature and time, where the quality indicators used were total viability of mold and yeast, water content, water activity, and pH. This research used a factor response surface methodology. Data were analyzed by ANOVA with an  $\alpha$  level of 95%. The result of this research showed that the optimum drying process for AC starter was 40°C for 10 hrs, with characteristic response viability  $8.8 \times 10^7$  CFU/g, water activity 0.43, water content 8.90%, and pH 4.05. CC starter showed an optimum drying process at 49°C for 4.5 hrs, with characteristic response viability  $4.9 \times 10^7$  CFU/g, water activity 0.49, water content 7.02%, and pH 3.95. The optimum tray dryer temperatures and times were achieved for AC and CC starters.

## 1. Introduction

White corn flour is a food commodity with limited use. This flour has a weakness, such as high viscosity, with high retrograde, the paste undergoes syneresis during storage, and low paste stability at high temperature and low pH (Aini *et al.*, 2010). Farasara *et al.* (2014) showed that fermentation with the addition of indigenous mould and yeast culture could change the characteristic of white corn flour paste of Anoman 1 after 36 hrs fermentation. The indigenous mould and yeast resulted from the isolation and identification of microorganisms in the spontaneous fermentation of white corn varieties of Anoman 1 and was grouped into AC and CC starters (Rahmawati *et al.*, 2013). To simplify the fermentation process and quality control, indigenous mould and yeast cultures were made the dried starter. Dried starters have been produced using sun

drying and oven drying methods (Rahmawati *et al.*, 2017). The sun-drying method was carried out by drying for 7 days, between 8.00 and 15.00 WIB, with a total drying time of 48 hrs. AC starters produced the best characteristics with viability of  $2.7 \times 10^8$  CFU/g and a moisture content of 13.34%. The sun-drying method has the disadvantage of uncontrolled temperatures and long drying times.

The weakness of this sun drying method may cause the growth of microorganisms to be less optimal. The oven-drying method was carried out at 40°C for 24, 48 and 72 hrs. The starter CC, which was dried for 48 hrs, had the best characteristics with a viability of  $5.8 \times 10^8$  CFU/g and a moisture content of 12.57%. This method has the advantage of being temperature controlled but still takes a long time to dry. In addition, the starter was still wet in the oven when drying for 24 hrs (Rahmawati

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et al., 2017). A more efficient and faster drying method is therefore needed, namely by using a tray dryer.

Rahmawati et al. (2019) have carried out the drying method for white corn starter using a tray dryer, where drying was conducted at 40 and 50°C for 1.5-6 hrs. This method has a more controlled temperature than the sunshine method and with a shorter time than the oven method. The tray dryer method can reduce the drying time and increase the efficiency of hot air contact with the material (Sari et al., 2017). The dried starter produced by Rahmawati et al. (2019) did not have optimal characteristics where the viability of the starters was  $<10^6$  CFU/g and the water content were  $> 10\%$ . Therefore, it is necessary to optimize the starter drying process which includes temperature and drying time using the tray dryer method. The optimization of the drying process for starters was carried out by using the D-optimal design of the Response Surface Methodology (RSM) method. RSM is a statistical and mathematical technique used primarily for the development, improvement and optimization of the production process (Myers et al., 2009). Based on the description above, this research aimed to optimize the tray dryer temperature and time of white corn flour culture by RSM.

## 2. Materials and methods

### 2.1 AC and CC starter preparation

AC consists of *Penicillium citrinum*, *Aspergillus niger*, *Acremonium strictum*, and *Candida famata*, while CC consists of *Penicillium chrysogenum*, *Penicillium citrinum*, *Aspergillus niger*, *Rhizopus stolonifer*, *Rhizopus oryzae*, *Fusarium oxysporum*, *Acremonium strictum*, *Candida famata*, *Kodamaea ohmeri* and *Candida krusei/incospicua*. These microorganisms used were previously isolated and identified from spontaneous fermentation of corn grits (Rahmawati et al., 2013)

A loop of each mould was streaked onto fresh Potato Dextrose Agar (PDA) slant and incubated for 5 days at 30°C. After five days, moulds were harvested by scraping, then suspended in 10 mL sterile water and appropriately dissolved to count using Mariendfeld haemocytometer. Yeast culture was prepared as above, but incubation was carried out for two days at 30°C. Yeast was also calculated using Mariendfeld haemocytometer (Rahmawati et al., 2017).

### 2.2 Optimization using RSM

The Response Surface Methodology (RSM) method was used to maximize the drying process by using the Design Expert ® 7.0 (DX7) statistical application. The experimental design aims to achieve an optimum response by combining several components (Keshani et

al., 2010). The mixed design is D-optimal where it was necessary to have a lower limit (-1) and an upper limit (+1). The independent variables in this study were drying temperature and time. The experimental design was based on RSM (Table 1).

Table 1. Independent variables and the level used in the design process

| Independent variables | Limits     |            |
|-----------------------|------------|------------|
|                       | Lower (-1) | Upper (+1) |
| Temperatures (°C)     | 40         | 50         |
| Times (et al.)        | 0          | 10         |

The parameters of the experiment were drying time (hrs) and drying temperature (°C). Drying time between 0-10 hrs and drying temperature between 40-50°C (Rahmawati et al., 2017). The AC and CC starter qualities were determined based on total viability mould-yeast, moisture content (oven method), water activity (Rotronic Hygrolab water activity meter) and pH (Benchtop pH meter Mettler Toledo Seven Compact).

There were criteria for each variable and response when performing optimization. The observed response was viability with an importance level of 5 (+ + + + +), while the response to moisture, aw and pH had an importance level of 3 (+ + +). The importance value will determine the process conditions that were closest to the target response. The chosen optimal combination is the one having the highest desired value.

### 2.3 Making starters and drying with a tray dryer

AC and CC starter culture made by sterilizing corn flour, then put it into a sterile basin and adding sterile distilled water as much as 2/3 of the total weight of cornflour. Prepared culture suspensions (AC) containing  $10^6$  CFU/mL per microorganism, then piped as much as 10% of the amount of water used. After that, all stir until homogeneous and put 17 g in each petri dish. Petri dishes were then incubated at 30°C for 5 days. Furthermore, the dough is dried using a tray dryer with a range of 40-50°C for 0-10 hrs. The dried AC and CC yeast mould culture was made powder using a blender that has been sprayed with 70% alcohol (Rahmawati et al., 2019)

### 2.4 Response measurement

Response measurements were carried out on the dried powder sample that was inserted into a plastic clip containing silica gel. The responses measured included total mould and yeast viability, aw, moisture content, and pH (AOAC, 2006).

### 2.5 Verification of optimization results

The results were validated at the highest desirability

point for AC and CC starters respectively. The AC and CC starting process was repeated directly using the optimal drying process. In addition, the test process included direct measurement of the overall yeast mold viability, moisture contents, water activity and pH to generate the actual response variable.

### 2.6 Data analysis

The data analysis technique used in this study includes linear ( $y = ax + b$ ) models, quadratic ( $y = ax^2 + bx + c$ ) and 2FI models, using the Response Surface Methodology.

## 3. Results and discussion

### 3.1 Viability test

The viability of the total yeast mould starter could be seen in Table 2. Based on Table 2, we can see that the total value of yeast mould AC and CC starters ranged from  $10^7$ - $10^8$  CFU/g at drying temperatures < 50°C. In the meantime, the viability value of the yeast mould AC and CC starters at drying temperatures 50°C and drying time of 7.3 to 10 hrs were 0. According to Pitt and Hocking (2009), *Aspergillus Niger* may grow at a minimum temperature of 6 – 8°C, the maximum temperature of 45–47°C, and optimal growth of 35 to 37°C, while *Rhizopus oryzae* can grow at a temperature of 7 to 42°C with an optimal growth temperature of approximately 37°C. *Candida krusei* can grow optimally at a temperature of 37°C (Scorzoni, 2013). The mould and yeast, in the beginning, were suspected to die at 50°C with a drying time of more than 7.3 hrs. Passamani et al. (2014) stated that *A. niger* may grow optimal at temperatures between 24 to 37°C, aw greater than 0.95, and pH levels between 4 to 6.5. Ayinla et al. (2017) have

found that *Rhizopus oryzae* ZAC3 can grow in the pH of 4.0 to 8.0 and temperature of 25 to 55°C. Besides that, Samaranyake and Samaranyake (1994) stated that *C. krusei* is a mesophile that can only grow at temperatures up to 45 °C.

The heat resistance of microorganisms is different, which is represented by the D value. The D value is defined as the time in mins at a given temperature which is reducing 90% or a logarithm of the number of spores or certain vegetative cells. Rahmawati et al. (2019) reported that the D value of AC and CC at temperatures of 40°C was 271.86 mins (4.5 hrs) and 523.10 mins (8.7 hrs) respectively, while at 50°C was 147.06 mins (2.45 hrs) and 127.93 mins (2.13 hrs) respectively.

### 3.2 Moisture content

Water content and water activity (aw) are closely linked with starters' shelf-life. These two parameters are indicators of the availability of water in food for the survival of microorganisms. In addition to affecting chemical changes, the water content in food also determines the microbial content of foods (Herawati, 2008). Products that have higher water content will relatively have a shorter shelf life (Amanto et al., 2015). Table 2 tabulates the water content for AC and CC starters.

The initial moisture content of the AC starter was 56.00% and CC starter was 57.30%. In this study, the desired water content was <10%. With this water content value, it was hoped that the microorganisms will remain alive but did not carry out the metabolic activity. The dried starter that drying at 50°C tended to have a lower moisture content than a lower temperature (40°C).

Table 2. The viability and water content responses for AC and CC starter as the results of RSM optimization

| Treatment | Drying Temperature (°C) | Drying time (Hr) | Viability (CFU/g)  |                    | Water content (%) |       |
|-----------|-------------------------|------------------|--------------------|--------------------|-------------------|-------|
|           |                         |                  | AC                 | CC                 | AC                | CC    |
| 1         | 40                      | 0                | $6.75 \times 10^8$ | $2.40 \times 10^8$ | 56.00             | 57.30 |
| 2         | 40                      | 4.5              | $3.05 \times 10^8$ | $3.00 \times 10^8$ | 11.67             | 11.01 |
| 3         | 40                      | 4.5              | $5.50 \times 10^8$ | $2.70 \times 10^8$ | 12.52             | 11.29 |
| 4         | 40                      | 7.3              | $7.50 \times 10^8$ | $1.85 \times 10^8$ | 8.66              | 10.98 |
| 5         | 40                      | 10               | $5.00 \times 10^8$ | $1.70 \times 10^8$ | 8.59              | 10.26 |
| 6         | 43                      | 0                | $5.00 \times 10^8$ | $3.00 \times 10^8$ | 56.00             | 57.30 |
| 7         | 43                      | 8.6              | $1.00 \times 10^8$ | $2.40 \times 10^7$ | 9.16              | 11.53 |
| 8         | 45                      | 0                | $5.00 \times 10^8$ | $3.00 \times 10^8$ | 55.90             | 57.30 |
| 9         | 45                      | 5.9              | $1.40 \times 10^8$ | $5.20 \times 10^7$ | 8.91              | 9.11  |
| 10        | 45                      | 10               | $1.40 \times 10^7$ | $4.50 \times 10^7$ | 10.61             | 10.65 |
| 11        | 50                      | 0                | $3.80 \times 10^8$ | $7.80 \times 10^8$ | 55.90             | 57.30 |
| 12        | 50                      | 4.5              | $7.50 \times 10^7$ | $3.00 \times 10^7$ | 5.65              | 5.44  |
| 13        | 50                      | 4.5              | $2.00 \times 10^7$ | $1.10 \times 10^7$ | 5.79              | 5.22  |
| 14        | 50                      | 7.3              | 0                  | 0                  | 8.66              | 10.98 |
| 15        | 50                      | 10               | 0                  | 0                  | 8.91              | 10.28 |
| 16        | 50                      | 10               | 0                  | 0                  | 8.80              | 10.34 |

According to Rahmawati *et al.* (2019), it caused the drying rate at 50°C was faster than at 40°C. As we know that a higher drying rate resulted in a faster drying time.

Drying time can also reduce the material's moisture content. At a temperature of 40°C with different drying times, the moisture content of AC starters decreased from 56.00% to 11.67% after 4.5 hrs of drying and continued to dry to 8.59% after 10 hrs of drying. Likewise, with CC starters, the moisture content of CC starters decreased from 57.30% to 11.01% after 4.5 hrs of drying and continued to dry to 10.26% after 10 hrs of drying at 40°C. In line with Fitriani (2008) statement, that drying temperature and time higher will evaporate water molecules more than lower temperature and time. Besides that, this condition will make the products ability to release water from its surface to be greater and it will cause lowered water content. Abasi *et al.* (2009) reported that the temperature affected moisture content. The moisture content of the samples decreased with increasing temperature, from 2.72 to 1.15% db, for drying temperature of 60°C to 90°C, respectively.

### 3.3 Water activities value

Overall, mould can live at a minimum water activity value. *Aspergillus* lives at a minimum water activity of 0.98, *Rhizopus* 0.93, and *Penicillium* 0.99, where yeast can usually live around 0.88-0.94 (Muchtadi and Sugiono, 2013). A low aw value can make starters' microorganisms dormant. AC and CC starter were expected to have a longer shelf life. Table 3 showed the water activity of the AC and CC starter.

When starters are dried for a long period of time, the water content will decrease, and the water activity

starters will decrease as well. This is in line with Leviana and Paramita's research (2017), where the increase in the drying temperature will cause more water to evaporate. This will decrease the water content. Likewise, with the water activity value, the increase of drying time caused the water activity value in the material to decrease (Chen, 2019).

### 3.4 pH value

During the incubation process, the pH value of both AC and CC starters decreased, which becomes slightly sour (3.9-4.5) and was accompanied by a distinctive fermentation aroma. Table 3 shows the pH value for AC and CC starters. This indicated that the filler substrate (carbohydrates) has been metabolized by the microorganisms added to simpler compounds such as ethanol, carbon dioxide, and organic acids that can lower the pH value. In the fermentation process, metabolism occurs from the activity of organic acid-producing microorganisms, thereby reducing the pH (Anggraeni and Yuwono, 2014). According to Rahmawati *et al.* (2019), the initial pH value of AC and CC was around 4.00, where this value was appropriate for microorganism growth. The decrease in pH value was due to the activity of microorganisms that converted carbohydrates into acids during the fermentation process.

### 3.5 Mathematical model relationship between process parameters and response

Table 4 shows the math model of the drying process parameter as a response of the AC starter and Table 5 for CC starter model. The linear model indicated that only temperature and drying time influence the response, but not the interactions between them. The quadratic model

Table 3. The water activity and pH value responses for AC and CC starter as the results of RSM optimization

| Treatment | Drying Temperature (°C) | Drying time (Hr) | Water Activity |       | pH value |      |
|-----------|-------------------------|------------------|----------------|-------|----------|------|
|           |                         |                  | AC             | CC    | AC       | CC   |
| 1         | 40                      | 0.0              | 0.929          | 0.945 | 4.20     | 4.15 |
| 2         | 40                      | 4.5              | 0.460          | 0.474 | 4.22     | 4.50 |
| 3         | 40                      | 4.5              | 0.454          | 0.465 | 4.26     | 4.50 |
| 4         | 40                      | 7.3              | 0.430          | 0.426 | 4.00     | 4.65 |
| 5         | 40                      | 10.0             | 0.433          | 0.421 | 3.95     | 4.60 |
| 6         | 43                      | 0.0              | 0.929          | 0.945 | 4.28     | 4.20 |
| 7         | 43                      | 8.6              | 0.558          | 0.645 | 4.05     | 4.60 |
| 8         | 45                      | 0.0              | 0.929          | 0.959 | 4.28     | 4.20 |
| 9         | 45                      | 5.9              | 0.518          | 0.570 | 4.20     | 4.10 |
| 10        | 45                      | 10.0             | 0.449          | 0.478 | 4.26     | 4.23 |
| 11        | 50                      | 0.0              | 0.942          | 0.959 | 4.20     | 4.12 |
| 12        | 50                      | 4.5              | 0.460          | 0.474 | 4.33     | 4.22 |
| 13        | 50                      | 4.5              | 0.454          | 0.438 | 4.50     | 4.20 |
| 14        | 50                      | 7.3              | 0.438          | 0.426 | 4.40     | 4.31 |
| 15        | 50                      | 10.0             | 0.372          | 0.356 | 4.44     | 4.28 |
| 16        | 50                      | 10.0             | 0.380          | 0.367 | 4.50     | 4.40 |

Table 4. The math model of drying proses parameter as a response of the AC starter

| Parameter                        | Math model | Significance model ( <i>p</i> ) | Lack of fit              | R squared | Adj R <sup>2</sup> model | Pred R <sup>2</sup> model | Adeq precision |
|----------------------------------|------------|---------------------------------|--------------------------|-----------|--------------------------|---------------------------|----------------|
| Viability (CFU/g)                | Linear     | 0.0123 (significant)            | 0.3702 (not significant) | 0.6237    | 0.5401                   | 0.3511                    | 6.248          |
| Water content (%)                | 2FI        | 0.0016 (significant)            | 0.0404 (significant)     | 0.8382    | 0.7775                   | 0.6959                    | 10.565         |
| Water activity (a <sub>w</sub> ) | Quadratic  | 0.0440 (significant)            | 0.0021 (significant)     | 0.7950    | 0.6242                   | 0.3560                    | 7.238          |
| pH                               | 2FI        | 0.0002 (significant)            | 0.6288 (not significant) | 0.8989    | 0.861                    | 0.7554                    | 13.784         |

Adj: Adjusted, Pred: Predicted, Adeq: Adequate

Table 5. The math model of drying proses parameter as a response of the CC starter

| Parameter                        | Math model | Significance model ( <i>p</i> ) | Lack of fit              | R squared | Adj R <sup>2</sup> model | Pred R <sup>2</sup> model | Adeq precision |
|----------------------------------|------------|---------------------------------|--------------------------|-----------|--------------------------|---------------------------|----------------|
| Viability (CFU/g)                | Quadratic  | 0.0474 (significant)            | 0.9033 (not significant) | 0.7894    | 0.6139                   | 0.2087                    | 5.656          |
| Water content (%)                | 2FI        | 0.0023 (significant)            | 0.0019 (significant)     | 0.8209    | 0.7537                   | 0.6636                    | 8.499          |
| Water activity (a <sub>w</sub> ) |            | 0.0616 (not significant)        | 0.0163 (significant)     | 0.7679    | 0.5746                   | 0.2841                    | 6.541          |
| pH                               | Quadratic  | 0.0193 (significant)            | 0.0769 (not significant) | 0.8479    | 0.7211                   | 0.3705                    | 6.974          |

Adj: Adjusted, Pred: Predicted, Adeq: Adequate

showed that each factor influences the response and interaction between temperature and drying time. The 2FI model means the response is influenced by the temperature-drying interaction.

### 3.6 Effect of drying process on starters viability

Rahmawati *et al.* (2017) have dried the starter culture using an oven with a temperature below 40°C resulted in the wet starters (not yet dry). It showed that the starter could be dried at the lowest 40°C temperature. However, the drying process using a tray drier cannot generate heat if the temperature is less than 40°C were used. Based on this, the drying temperature used was up to 50°C (Rahmawati *et al.*, 2019). The results showed that drying at 50°C for 6 hrs still produced the number of microorganisms that met the minimum requirements for the viability of starter microorganisms (10<sup>6</sup> CFU/g). These results were in line with Oliveira *et al.* (2002) where a good fermented drink produced by the number

of bacteria was at least 5.3×10<sup>6</sup> CFU/mL.

Thus, drying starters at a temperature of 40-50°C were expected to maintain a significant total viability mold and yeast. Figure 1 and Figure 2 show the graph of the relationship between temperature and drying time on the viability response of AC and CC starters respectively.

Based on the graph in Figure 1, an increase in temperature and drying time with a tray dryer caused a decrease in yeast viability on the starters. At a temperature of 50°C. with a drying time of about 7.3 hrs, a value of 0 was produced. It indicates the absence of mould-yeast during the analysed. AC starters are mostly yeast. At this temperature and drying time, the yeast on AC starters was suspected to die. Vegetative yeast cells are killed with humid heat at 50-60°C in 10-15 mins (Pelczar, 2012).

Figure 1 provides an overview of the AC-starters

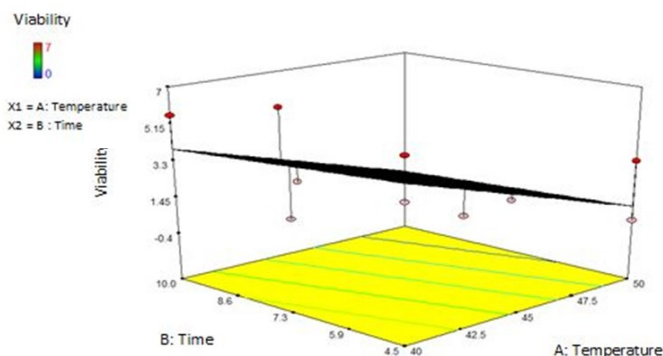


Figure 1. A 3D graphical combination between temperature and drying time to the viability response of AC starters.

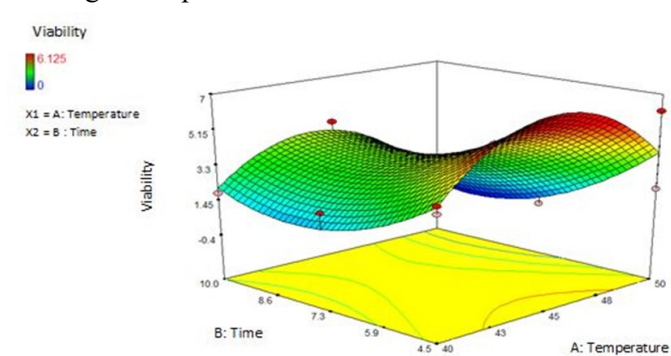


Figure 2. A 3D graphical combination between temperature and drying time to the viability response of CC starters

viability response model. The red colour in the figure showed a high viability value, while the blue colour showed a low viability value. Microbes have different heat resistance as expressed by Rahmawati *et al.* (2019), the calculation results showed that the values of D starters AC and CC at 40°C were 271.86 mins (4.5 hrs) and 523.10 mins (8.7 hrs) while at 50°C were 147.06 mins (2.45 hrs) and 127.93 mins (2.13 hrs) respectively. AC starters were more heat-resistant. It caused AC starter to contain fewer types of microbes, so, when making starter cultures the competition between microbes was lower. This resulted in more available microbes. It was indicated by the higher initial microbial viability than CC starter.

The program selected model for appropriate viability response is a linear model with an  $R^2$  value of 0.6237. AC mould-yeast viability response model has a 0.0123 p-value ( $\text{Prob} > F$ ). This showed that the model can still describe the viability response (AC), as it has a p-value  $< 0.05$ . The results of ANOVA also showed that temperature and drying time had a significant impact on viability response. This is evidenced by the insignificant fit shortage,  $> 0.05$  (0.3702). Therefore, in this study, the viability modelling shows that the temperature factor (40-50°C) and drying time (0-10 hrs) have a significant effect on the AC viability response.

Figure 2 provides a surface overview of the CC dissolved viability response model. The model chosen by the program for the appropriate viability response was a quadratic model with an  $R^2$  value of 0.7894. The CC mould-yeast viability response model has a p-value ( $\text{Prob} > F$ ) of 0.0474. This showed that the viability response (AC) can still be described well by the model because it has a p-value  $< 0.05$ . ANOVA results also showed that the temperature and drying time had a significant effect on the viability response. This is evidenced by the insignificant Lack of fit, which is  $> 0.05$  (0.9033). Therefore, the viability modelling in this study showed that the temperature factor (40-50°C) and drying time (0-10 hrs) significantly influenced the CC viability response.

### 3.7 Drying process on starters moisture content

Apart from the drying process, the fermentation process in making starters also plays a role in reducing levels. Pusparani and Yuwono (2014) stated that during the fermentation process, the breakdown of starch by enzymes produced by microorganisms will produce simple sugars such as glucose and accompanied by the release of water. This is known as starch degradation. Starch degradation is characterized by a decrease in the ability of the material to retain water due to the loss of hydroxyl groups. The graph of the relationship between

the combination of temperature and drying time to the water content response of AC starters can be seen in Figure 3.

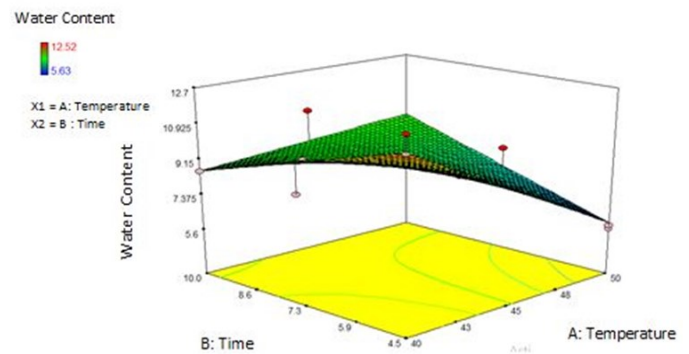


Figure 3. A 3D graphical combination between temperature and drying time to the water content response of AC starters

The water content response value from AC starters ranged from 5.63 to 12.52%. Figure 3 provides an overview of the AC starters moisture response model. The image's red colour indicated high water content, while the blue colour indicated low water content.

The model chosen by the water content response program was the 2FI model with an  $R^2$  value of 0.8382. The AC moisture response model has a 0.0016 p-value ( $\text{Prob} > F$ ). This shows that the model can still describe the viability response (AC), as it has a p-value  $< 0.05$ . However, the results of ANOVA did not show that the temperature and drying time had a significant effect on the water content response with a significant fit value shortage,  $< 0.05$  (0.0404). The significant lack of fit value indicates that the temperature (40-50°C) and drying time parameters (4.5-10 hrs) have no significant effect on the water content response in AC starters.

Figure 4 shows the graph of the relationship between the combination of temperature and drying time to CC starter of water content response. The response value to the moisture content generated from CC starters ranged from 5.22-11.53%. The model chosen by the water content response program was the 2FI model with a 0.8209  $R^2$  value. The CC moisture response model has a 0.0023 p-value ( $\text{Prob} > F$ ). This shows that the model can still describe the viability response (AC), as it has a p-value  $< 0.05$ . However, the results of ANOVA did not show that the temperature and drying time had a significant effect on the water content response with a significant fit value shortage,  $< 0.05$  (0.0019). The significant lack of fit value indicates that temperature parameters (40-50°C) and drying time (4.5-10.0 hrs) do not significantly affect the response of moisture content in CC starters.

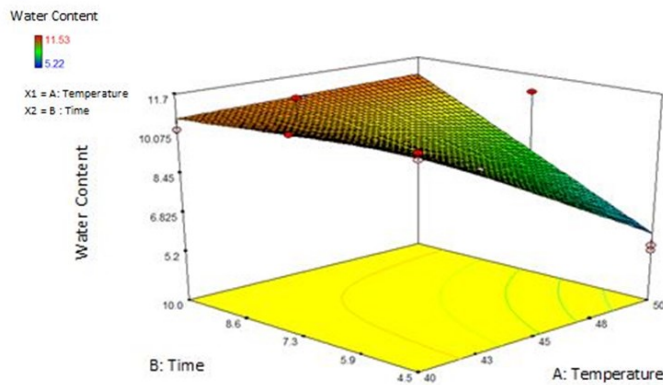


Figure 4. A 3D graphical combination between temperature and drying time to the water content response of CC starters

### 3.8 Effect of drying process on water activity value

Apart from the drying process, the fermentation process in making starters also plays a role in reducing the levels. Pusparani and Yuwono (2014) state that during the fermentation process, the breakdown of starch by enzymes produced by microorganisms will produce simple sugars such as glucose and accompanied by the release of water. This is known as starch degradation. Starch degradation is characterized by a decrease in the ability of the material to retain water due to the loss of hydroxyl groups. The graph of the relationship between the combination of temperature and drying time to the water content response of AC starters can be seen in Figure 5.

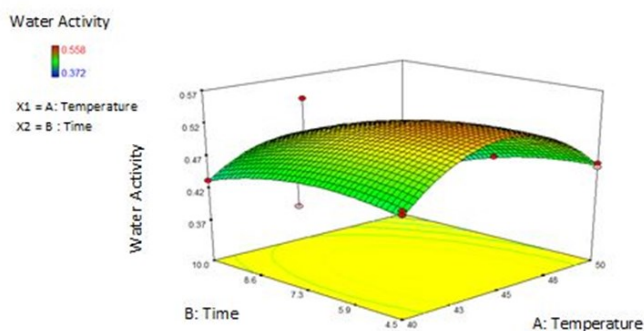


Figure 5. A 3D graphical combination between temperature and drying time to the water activity response of AC starters

Water activity (aw) indicated the amount of free water in a product. Free water in food was needed by growing microorganisms for nutritional processes, a medium for enzymatic reactions, and cellular component synthesis (Chen, 2019). The lower a product's aw value, the lower the risk of chemically or microbiologically damaging the food product. The smaller a product's aw value, the longer the product's shelf life since bacteria, moulds, and yeasts require high aw to grow. Overall, the minimum water activity for bacterial growth is 0.75, mould is 0.60, while the minimum yeast growth is 0.80 (Susilo *et al.*, 2019).

Figure 5 shows the graph of the relationship between temperature and drying time to the response of aw starters of AC. The AC-generated response ranged from 0.372 to 0.558. The red colour shows a high aw, while the blue colour shows a low aw.

The model selected for the appropriate aw response is the quadratic model. Figure 5 provides an overview of the aw response model. The image's red colour indicated a high aw value, while the blue colour indicated a low aw value. The aw (AC) response model has a value of 0.0440 p (Prob > F), indicating that the model was significant and can be described well at a 5% level (p-value < 0.05). However, the ANOVA results showed a significant fit shortage, < 0.05 (0.0021). This meant that the temperature and drying time does not affect the AC response.

Meanwhile, the CC-starter aw response ranged from 0.356 to 0.645. The CC-starters water activity parameter (aw) represented the mean mathematical model. This showed that CC starters' mould-yeast viability and moisture content due to treatment occurs randomly and cannot be explained by the model. Figure 6 shows the graph of the relationship between temperature and drying time to the aw of CC response.

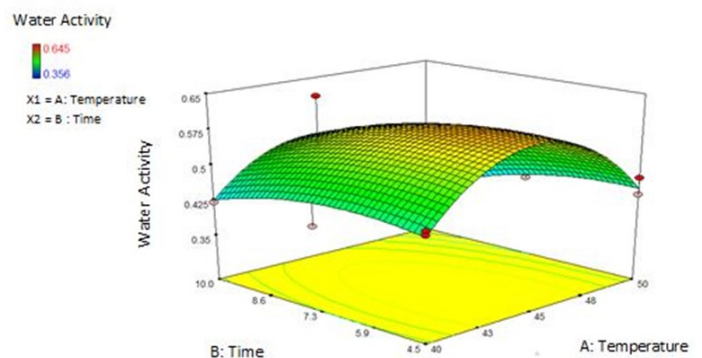


Figure 6. A 3D graphical combination between temperature and drying time to the water activity response of CC starters

### 3.9 Effect of drying process on pH value

Acidity or pH indicates the active concentration of hydrogen ions. The pH value is used to determine the variety of microorganisms that may grow on the product where each microorganism has a specific growth pH. Pratama *et al.* (2013) stated that the final results of the pH value for yeast bread, tempeh yeast, and *Lactobacillus plantarum* were 4.37; 3.43; and 3.93 at 96 hrs of fermentation respectively. For microorganisms, pH influenced growth and survival. Each type of microorganism has an optimum growth pH and pH range. In general, mould and yeast can grow more widely than bacteria (Rahayu and Nurwitri, 2012). Mould has a very wide growth pH ranged from 2.0-8.5, while yeast has a growth pH range from 4.0-4.5 and will

not grow well under alkaline conditions (Muchtadi and Sugiyono, 2013).

The group of microorganisms capable of fermenting food nutrients will convert some or all of the food components into fermented products, such as lactic acid, ethanol, CO<sub>2</sub>, or other organic acids. Organic acid accumulation causes pH to decrease during incubation. According to Kartohardjono *et al.* (2007), CO<sub>2</sub> gas is often called acid gas because CO<sub>2</sub> gas has acidic properties. CO<sub>2</sub> gas contributes to the pH value. Figure 7 shows the relationship between the combination of temperature and drying time to the pH response of AC starters. The pH response from AC starters ranges from 3.95 to 4.50. The image's red colour indicates high pH, while the blue colour indicated low pH.

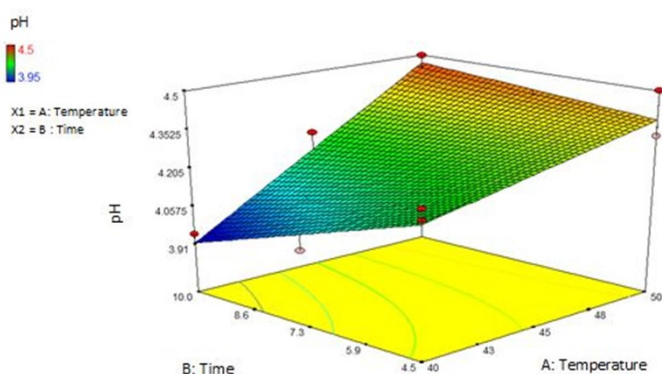


Figure 7. A 3D graphical combination between temperature and drying time to the pH response of AC starters

The model chosen by the program is the 2FI model with an R<sup>2</sup> value of 0.8989. The AC pH response model has 0.0002 p-value (Prob > F). This shows that the model can still describe the pH (AC) response as it has a p-value < 0.05. ANOVA results also showed that the temperature and drying time had a significant pH response effect. This is evidenced by the insignificant fit lack, > 0.05 (0.629). Temperature parameters (40-50°C) and drying time (4.5-10 hrs) have a significant impact on pH response on AC starters.

Figure 8 shows the relationship between the combination of temperature and drying time to the pH response of CC starters. The model selected by the program is the 2FI model with an R<sup>2</sup> value of 0.8479. The CC pH response model has a 0.0193 p-value (Prob > F). This showed that the model can still describe the pH (CC) response as it has a p-value < 0.05. ANOVA results

also showed that the temperature and drying time had a significant pH response effect. This is evidenced by the insignificant fit lack, > 0.05 (0.0769). Temperature parameters (40-50°C) and drying time (4.5-10.0 hrs) influenced the pH response in AC starters significantly.

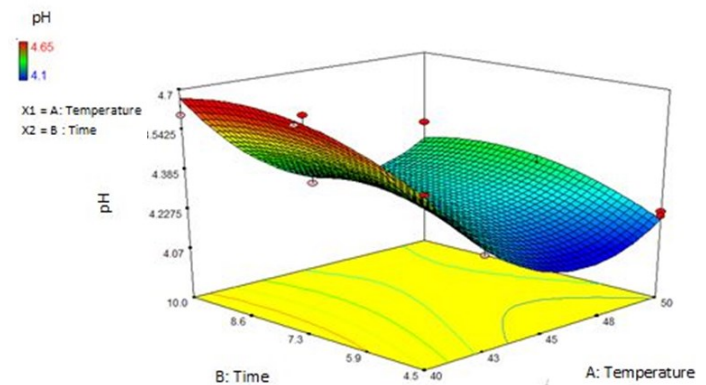


Figure 8. A 3D graphical combination between temperature and drying time on the pH response of CC starters

### 3.10 Process optimization with RSM

The process optimization stage aimed to obtain the drying process conditions for starting cornflour with an optimal response value based on the specified criteria. Based on the numerical optimization performed, a corn flour solution starts drying formula with the highest desirability value recommended by RSM for each starter, as presented in Table 6. The desirability value is a parameter showing the best optimization results with a range of 0–10. The closer to 1.0 the recommended solution can fulfil the desires according to the criteria of the stated objectives and interests (Myers *et al.*, 2009). The combination of drying formula for corn flour starters selected by the AC starter program was a temperature of 40°C for 10 hrs, while CC starters are 49°C for 4.5 hrs.

Based on the data in Table 6, the optimum formula for AC starters has a predictive response of 3.929 log CFU/g or  $8.5 \times 10^3$  CFU/g, 8.60% water content, 0.433 water activity, and pH 3.91. While the optimum CC starter formula (Table 6) has a predictive response of 4.958 log CFU/g or  $9.0 \times 10^4$  CFU/g, 6.48% water content, aw 0.499, and pH 4.13.

### 3.11 Results verification

Result verification was performed at the point with the highest desirability value, respectively, for AC and

Table 6. Comparison of response predictions with verification results of AC and CC starter

| Starter and response |            | Temperature (°C) | Time (Hr) | Viability (CFU/g) | Water activity (a <sub>w</sub> ) | Water content (%) | pH   |
|----------------------|------------|------------------|-----------|-------------------|----------------------------------|-------------------|------|
| AC                   | Prediction | 40               | 10        | 3.929             | 0.433                            | 8.60              | 3.91 |
|                      | Actual     | 40               | 10        | 7.944             | 0.425                            | 8.90              | 4.05 |
| CC                   | Prediction | 49               | 4.5       | 4.958             | 0.499                            | 6.48              | 4.13 |
|                      | Actual     | 49               | 4.5       | 7.698             | 0.487                            | 7.02              | 3.95 |



CC starters. The process of starting AC and CC was repeated directly using the optimum drying process formula. In addition, the testing process included measuring the total viability of yeast fungi, moisture content, water activity, and pH directly to generate the actual response variable. The predicted response can be compared with AC and CC starters verification results in Table 6.

Based on the verification, there is a significant difference in the value of starter viability between the formula solution suggested by RSM and the verification. According to Rahmawati *et al.* (2020), the optimization using RSM was unfit to describe the viability response model. This due to an AC-indigenous cocktail yeast mould culture consists of more than one microorganism. So, the activity of AC during the fermentation process varies, because the optimum conditions for growth during incubation for each microorganism vary and maybe there was competition for nutrients by microorganisms varies. On the other hand, the value of 0 in the viability result affected the design of RSM's optimum formula.

#### 4. Conclusion

Based on the research results, it can be concluded that the combination of temperature and drying time affects the characteristics of the white corn flour starters. The optimum drying process for AC starters is at a temperature of 40°C for 10.0 hrs with viability characteristics of 7.944 log CFU/g or  $8.79 \times 10^7$  CFU/g, 8.90% moisture content, aw 0.425 and pH 4.05. The optimal drying process for CC starters is at a temperature of 49°C for 4.5 hrs with viability characteristics of 7.698 log CFU/g or  $4.9 \times 10^7$  CFU/g, the water content of 7.02%, aw 0.487 and pH 3.95.

#### Conflict of interest

The authors declare no conflict of interest.

#### Acknowledgements

The authors would like to acknowledge the Indonesian Ministry of Research and Higher Education – Directorate of Research and Community Empowerment for grant research No. 28/AKM/PNT/2019.

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