#### **Review Article**

## Various Methods of Reducing Calcium Oxalate Levels in Tubers: A Review

# Ati Atul Quddus<sup>1\*</sup>, Erliza Hambali<sup>1,2</sup>, Mulyorini Rahayuningsih<sup>1</sup>, Ika Amalia Kartika<sup>1</sup>, Slamet Budijanto<sup>3</sup>

<sup>1</sup> Department of Agro-industrial Technology, IPB University, Bogor, Indonesia

<sup>2</sup> Surfactant and Bioenergy Research Center (SBRC), Bogor Agricultural University, Bogor, Indonesia

<sup>3</sup> Department of Food Science and Technology, IPB University, Bogor, Indonesia

\*E-mail: atisyaiful2020ati@apps.ipb.ac.id

Received: 02/12/2023	Revised: 26/12/2023	Accepted: 29/12/2023
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#### ABSTRACT

Tubers, as a highly promising agricultural commodity with distinctive flavors and nutritional content, pose health challenges due to the presence of calcium oxalate. Excessive consumption can lead to mechanical disturbances in the digestive and renal tubule systems. This complicates processing techniques to ensure that tubers, before being used as raw materials in the food and other industries, do not contain calcium oxalate. The research method involves a literature review, in-depth exploration of theories and concepts, and identification of variables related to the research method and context. From the literature study, it is evident that focusing on understanding fermentation parameters, such as time, temperature, pH, and the type and concentration of microorganisms, is crucial. This information is expected to provide insights into potentially effective methods for reducing calcium oxalate content in tubers, supporting the development of safer and healthier agricultural and food processing practices.

Keywords: Calcium oxalate; Various; Potential method; Reduction; Tubers.

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#### Introduction

Tubers can thrive in Indonesia [1], but the level of consumption among the population remains low. According to the Security Agency Food (BKP) of Agriculture, the tuber consumption value in 2019 was 15.9 kg/capita/year, which is still recommended below the 36.5 kg/capita/year. Tubers are known as a source of carbohydrates in daily food. The utilization of tubers is closely related to the recognition of their characteristics [1].

One type of tuber containing calcium oxalate crystals is the konjac tuber, which is almost evenly distributed throughout its central and peripheral parts [2]. Consumption of calcium oxalate can cause itching, burning sensations, irritation in the mouth, skin, and digestive tract [3]. Oxalic acid can bind calcium and is poorly absorbed by the body. Excessive consumption of calcium oxalate and oxalic acid can lead to kidney problems [4], [5], [6]. Additionally, in terms of chemical structure, calcium oxalate crystals absorb calcium, which is essential for nerve function and muscle fibers. In extreme cases, calcium absorption can lead to hypocalcemia and fatal paralysis [7]. To eliminate calcium oxalate content in tubers, pre-treatment is necessary before they are used as raw materials in the food and other industries. Therefore, in this paper, we discuss several potential methods to reduce calcium oxalate in tubers.

## Materials and methods

The data is sourced from a search through the electronic databases Web of Science (WoS) and Scopus, as leading scientific information platforms that access the most significant scientific databases and across various fields of publications knowledge. То develop a profound understanding of the topic, the authors conducted a systematic literature review of articles reviewed by peers over the last 10 years. Out of a total of 86 identified key studies, the authors categorized three method definitions for reducing calcium oxalate content in tubers as the focus of the study: mechanical, chemical, and biological methods. After reporting the findings of the systematic literature review, the authors discuss the best methods to reduce factors related to the reduction of calcium oxalate content in tubers.

## **Results and Discussion**

Calcium Oxalate Reduction Method in Tubers the results of a review of research articles that reduce calcium oxalate in tubers can be seen in Figure 1.

The figure 1 above explains three methods used to reduce the calcium oxalate content in tubers: mechanical, chemical, and biological. The processing processes of these three methods all begin with physical treatment in the form of stripping, washing, cutting, slicing, or crushing. This is adjusted to the advanced Method that will be used, whether mechanical, chemical, or biological.

## 1. Chemical Method

Calcium oxalate can be eliminated through chemical treatment, such as dissolving it in strong acid to decompose it into oxalic acid [8], [9]. Citric acid can penetrate the idioblast cell walls and dissolve calcium oxalate crystals [10]. Soaking in salt (NaCl) reduces calcium oxalate content through a reaction with Na+ and Cl- ions that attract calcium oxalate in the material [9]. Table 1 summarizes various mechanical and chemical methods for reducing calcium oxalate in tubers from research conducted in the last five years.

# 2. Mechanical Method

The mechanical method reduces calcium oxalate content in products by separating oxalate crystals through size reduction using screening or filtration processes [22]. Although fast, this method requires significant energy, increases operational costs, and has the potential to pollute the environment. Research showed that optimizing mechanical milling of koniac flour resulted in a calcium oxalate content of 0.3% [23]. Handayani [24] study highlighted the impact of ball mill grinding time on yield, viscosity, and konjac flour glucomannan content, with the best results obtained after 4 hours of grinding. The ball mill method with chemical purification in 8 hours of grinding (L8) produced konjac flour with a calcium oxalate content of 0.89%, flour size of 180-322.7 µm, and whiteness degree of 69.65 [25].

## 3. Biological Methods

Cultivation process is an alternative method for reducing oxalate content in food materials by utilizing microbes that use oxalate as a source of nutrition. Several bacterial strains, such as Lactobacillus sp., have proven to degrade calcium oxalate, with examples like Lactobacillus plantarum and Lactobacillus acidophilus each having the ability to degrade oxalate by 42% and 38%, respectively. Bacteria like Bacillus sp. also have the capability through the enzyme oxalate decarboxylase (OxdC) [26], [27]. The fermentation process in kimchi, rich in oxalate, shows a reduction of up to 72.3% in swiss chard [28]. Lactic acid bacteria, especially Lactobacillus sp., in kimchi have the potential to degrade oxalate [29], [30]. Table 2 presents biological methods that, in the last five years, have assisted in reducing calcium oxalate in tubers with the aid of initial physical treatments.

## 4. Initial Treatment of Tubers

The initial treatment for reducing calcium oxalate in tubers involves three sub-processes: peeling and washing, size reduction (*slicing*), and grinding (*pulping*) (Figure 2). Combining these subprocesses produces clean and small tuber pulp, impacting the calcium oxalate reduction process during fermentation [30]. Peeling

and washing at the beginning affect the resulting tuber flour's calcium oxalate levels, with the peeling method showing a reduction [13]. Smaller pulp size facilitates microbial reduction, and wet milling produces lower levels than dry milling [30]. Future research need not optimize initial handling conditions.

Method and Results	Refrences
<b>Tubers Porang</b> Boiling the chips using $6 - 8\%$ NaCl solution at 80°C for $5 - 30$ minute.	
The higher the NaCl concentration and the longer the boiling time, the lower the calcium oxalate level in porang tuber flour. The best treatment was obtained by using 8% NaCl and boiling for 25 minutes with calcium oxalate levels 0.65%. Soak 2 g of porang flour in 200 mL of $3 - 7\%$ starfruit juice, $3 - 7\%$ lime juice,	[11]
and $10 - 20\%$ vinegar acid for 15 minutes.	
The higher the concentration of the starfruit juice solution, the higher the percent reduction of calcium oxalate in porang tuber flour, namely 62.68%. The same thing happened when using acetic acid solution with a reduction percentage of calcium oxalate in porang tuber flour of 90.27%. Meanwhile, using lime juice solution did not show a higher percent reduction in calcium oxalate in porang tuber flour as the concentration of lime juice solution increased. Percent reduction of calcium oxalate in porang tuber flour was 65.94% when using a 5% lime juice solution.	[12]
Soaking 50 g of chips in 250 mL of 3 $-$ 7% starfruit juice solution for 3 x 15	
minutes. The higher the concentration of starfruit juice solution, the higher the percent reduction of calcium oxalate in porang tuber flour, namely 37.38%.	[12]
Soak the chips in water for $0 - 180$ minutes without peeling and with peeling. The longer the soaking time, the levels of oxalate compounds in porang tuber flour decrease, both in the treatment without peeling and with peeling. In addition, in the treatment with peeling, the levels of oxalate compounds in porang tuber flour were smaller when compared to the treatment without peeling, where the levels of oxalate compounds were 197.57 ppm and 86.44 ppm respectively after soaking 180 minutes.	[13]
Soak 50 g of chips in 250 mL of 5% H2SO4 solution at room temperature, 40°C, 50°C and 60°C for 15 minutes. The higher the soaking temperature, the percent reduction of calcium oxalate in porang tuber flour increased, namely 53.91% at a soaking temperature of 60°C.	[14]
Boil the chips using 15% NaCl solution at 80°C for 25 minutes, then rinse with water and drain, and continue soaking in 0.02% sodium bisulfite solution for 10 minutes.	[15]
The method used can reduce 96.00% of calcium oxalate in porang tuber flour.	

## Table 1. Continue

Method and Results	Refrences
Combination of soaking in 5% salt solution for 4 hours and blanching at 90°C for 15 minutes. The combination of the method of soaking in a 5% salt solution for 4 hours and blanching at a temperature of 90°C for 15 minutes can reduce the calcium oxalate content in porang tuber flour by 51.85%, which is greater than using just one method (soaking in the solution). 5% salt for 4 hours can reduce 14.81% calcium oxalate in porang tuber flour, while Blanching at 90°C for 15 minutes can reduce 18.52% calcium oxalate in porang tuber flour.	[16]
Soak 50 g chips in 250 mL of 5% lime solution at room temperature for 15, 30, 45, and 60 minutes. The longer the soaking time in 250 mL of 5% lime solution at room temperature, the percent reduction of calcium oxalate in porang tuber flour increased, namely 31.79% at a soaking time of 60 minutes.	[12]
Purple Taro Tubers	
Soak 100 g chips in 250 mL NaCl 5 $-$ 15% at room temperature for 60 minutes.	
Soaking 100 g of chips in 250 mL of 10% NaCl at room temperature for 60 minutes can reduce 22.89% of calcium oxalate in purple taro tuber flour and is the best treatment.	[25]
Boiling or steaming the chips for 20 minutes (500 g chips in 1 L of water at 100 °C) and soaking in a chemical solution (6% NaHCO3, 20% CH3COOH, and 10% NaCl). Boiling and soaking in 10% NaCl for 60 minutes can reduce 75.03% of calcium oxalate in purple taro tubers and is the best treatment.	[26]
Soak 500 g chips in 1 L CH3COOH 20% for 30 minutes, 1 L NaCl 10% for 15 minutes, and 1 L NaHCO3 6% for 60 minutes, each at room temperature.	
Soaking 500 g chips in 1 L of 6% NaHCO3 for 60 minutes at room temperature can reduce 45.58% of calcium oxalate in purple taro tubers and is the best treatment.	[27]
Tubers Iles-iles	
Soak 1.5 kg chips in $0 - 15\%$ NaCl solution at room temperature for 60 minutes after soaking in warm water at 40°C for 3 hours.	
The higher the NaCl concentration, the percent reduction of calcium oxalate in iles-iles tuber flour increased, namely 91.60% at a NaCl concentration of 15%.	[28]
Soaking chips at room temperature using 7% lime solution or 7% CaCO3. The method used can reduce $60 - 70\%$ of calcium oxalate in iles-iles tuber flour.	[29]
Tubers Kimpul	
Boiling or steaming the chips for 20 minutes (500 g chips in 1 L of water at 100°C) and soaking in a chemical solution (6% NaHCO3, 20% CH3COOH, and 10% NaCl).	
Boiling 500 g of chips in 1 L of water at a temperature of 100 °C and soaking in 20% CH3COOH for 30 minutes can reduce 60.12% of calcium oxalate in kimpul tubers and is the best treatment.	[26]

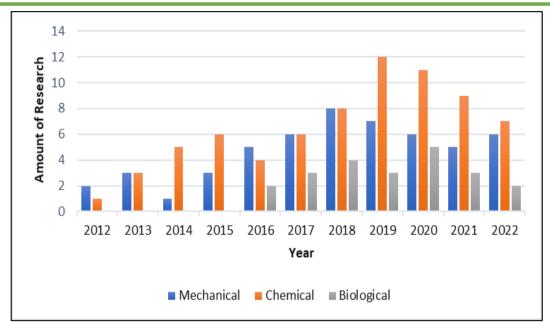
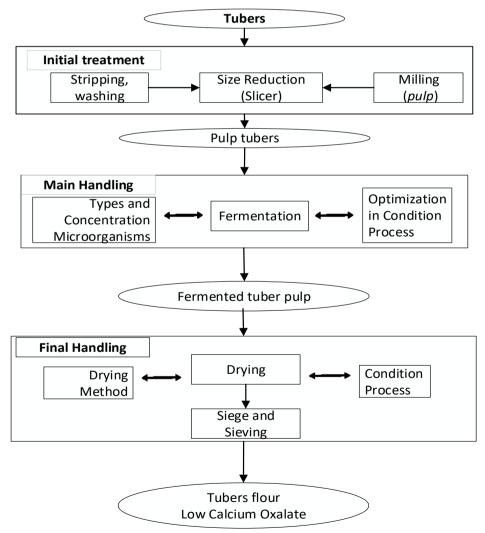
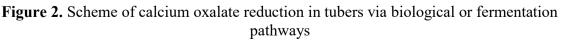


Figure 1. Research that has been carried out to reduce calcium oxalate in tubers





The primary treatment for reducing calcium oxalate in tubers involves one subprocess: fermentation (Figure 2). Identifying process conditions (time. temperature, pH, microorganism type, and concentration) is crucial for optimum conditions in reducing calcium oxalate levels in tuber flour. Fermentation process optimization is a key agenda for future research. The final treatment for reducing calcium oxalate in tubers includes two subprocesses: drying, flouring, and sieving (Figure 2). Combining these subprocesses is applicable to producing low water and calcium oxalate content in tuber flour. Understanding drying method and conditions is essential for maintaining flour post-fermentation. Previous quality research indicates using an oven or cabinet dryer at 60°C for 6 - 24 hours for drying tuber flour after the initial handling process. Sunlight drying has also been explored [30]. Flouring and sieving depend on sieve type and size, with the hypothesis that using a sieve above 135 µm or a 100 mesh sieve allows calcium oxalate crystals to pass through, resulting in low calcium oxalate content in tuber flour [31]. Future research need not optimize conditions for drying, flouring, and sieving subprocesses in the final handling of fermented tuber flour.

 Table 2. Several biological methods used to reduce calcium oxalate in tubers with the help of physical treatment in the initial process

Method and Results	Reference	
Tubers Porang		
Ferment chips for 21 days at room temperature using Bacillus subtilis which has been adapted to media containing 10% porang tubers for 4 days ( $pH = 5.5$ and temperature = $37^{\circ}C$ )	[22]	
Fermentation with the addition of $10 - 30\%$ Bacillus subtilis (dbd) can reduce calcium oxalate levels in porang tuber flour by around $58 - 65\%$ without changing the nutritional composition.	[32]	
Wet milling and fermentation using Lactic Acid Bacteria (LAB) with a LAB:water ratio = 1:1000 for 6–24 hours.		
The longer the fermentation time, the calcium oxalate levels in porang tuber flour decrease, except for the 24 hours fermentation time which is higher compared to other fermentation times. In this method, the best treatment was produced during fermentation for 18 hours with a calcium oxalate content in tuber flour of 2.64%. Dry grinding and long fermentation time using Lactic Acid Bacteria (LAB) with a LAB:water ratio = 1:1000 for 6-24 hours.	[30]	
The longer the fermentation time, the calcium oxalate levels in porang tuber flour increase, except for the 12 hours fermentation time which is lower compared to other fermentation times. In this method, the best treatment was produced during fermentation for 12 hours with a calcium oxalate content in tuber flour of 4.08%. Ferment chips (thickness $\pm$ 0.5 cm) at room temperature for 0 – 72 hours.	[30]	
The longer the fermentation time, the calcium oxalate levels in porang tuber flour increase, except for the 24 hour fermentation time which is lower compared to other fermentation times.	[33]	
Ferment chips (thickness $\pm$ 0.5 cm) at room temperature using kimchi bacteria (Lactobacillus sp. and Bacillus sp.) at pH 5-7 and fermentation time for 24-72 hours.	[22]	
The fermentation method at different pH conditions and fermentation times produces calcium oxalate levels in porang flour ranging from $0.050-0.069 \text{ mg/g}$ , where fermentation for 24 hours at pH 6 is the best treatment.	[32]	

#### Table 2. Continue

Method and Results	Reference
Taro Tubers	
Ferment chips naturally (Chips:Air = 1:2) for $0-72$ hours.	
The longer the fermentation time, the calcium oxalate levels in taro tuber flour	[24]
decrease, except for the fermentation time of 72 hours which is higher compared	[34]
to the fermentation time of 48 hours.	
Tubers Kimpul	
A total of 500 g of chips was fermented in a closed bottle (bottle capacity 1.5 L)	
using Saccharomyces cerevisiae and Rhizopus inoculum for 48 and 72 hours.	
Fermentation for 72 hours produced calcium oxalate levels in kimpul flour of	[27]
0.334 mg/g, which was lower compared to fermentation for 48 hours which	
produced calcium oxalate levels in kimpul flour of 0.472 mg/g.	

#### Conclusions

Reducing calcium oxalate levels in tubers can be achieved through three methods: mechanical. chemical. and biological. Biological methods are considered potential because they are safer, utilizing microbes beneficial to the body, and can reduce production costs compared to other methods. Chemical methods involve high costs, limited availability, and have the potential to impact health if used over a prolonged period. Mechanical methods, although fast, require high costs and energy and are challenging to apply on a small scale. In biological methods, fermentation parameters such as time, temperature, pH, and the type of microorganism need to be identified for optimal results.

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