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Physicochemical Characteristics of Chicken Eggshell Flour Produced by Hydrochloric Acid and Acetic Acid Extraction

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Abstract

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BACKGROUND: Several previous researchers have prepared chicken eggshell flour using hydrochloric acid (HCl) and acetic acid (CH₃COOH) solvents. However, the results of the physicochemical characteristics still very diverse.

AIM: This study aims to produce chicken eggshell flour using two different types of solvents (4% HCl and CH₃COOH 2N) and determine the proper solvent to obtain better physicochemical characteristics.

METHODS: This type of research was descriptive quantitative. The research stages initiated by preparing chicken eggshell samples by maceration using 4% HCl and CH₃COOH 2N. Afterward, chemical characteristics (moisture content, ash, calcium carbonate, calcium, and phosphorus) and physical characteristics (yield) of eggshell flour were determined. Processing and data conducted using the IBM SPSS statistic program.

RESULTS: The higher levels of calcium, calcium carbonate, phosphorus, ash, yield, and lower water content of the eggshell powder were generated by extraction using 4% HCl than 2N acetic acid (CH₃COOH).

CONCLUSION: The physicochemical characteristics of the eggshell powder extracted from the 4% HCl were better than the 2N acetic acid (CH₃COOH).

Introduction

Chicken eggshells as waste are mainly generated from the production of food processing industries such as cakes, bread, egg flour, frozen eggs, restaurants, households, and large-scale poultry hatcheries. In recent years, a small portion of chicken eggshells has been used as animal feed supplements, acid soil remediation, wastewater treatment, building block manufacturing, heterogeneous catalysts in biodiesel production, food supplements, cosmetics, and the pharmaceutical sector [1], [2]. However, most of the chicken eggshells worldwide are dumped in landfills which are likely to cause environmental problems [3]. The process of eggshell decomposition produces ammonia, hydrogen sulfide, and amines. Those components generate smell odors that attract mice, flies, insects, and pathogens such as *Escherichia coli* and *Salmonella* [4].

The chemical composition of eggshells consists of 1.0% protein (w/w), 95% calcium carbonate (w/w) [5]; in eggshells weighing 60 g contains about 2.3 g calcium and 97% calcium carbonate [6]; contains high calcium (Ca), which is between 385 and 401 mg/g, magnesium (Mg) 3.5 and 4.5 mg/g and total phosphorus

(P) 0.2 and 1.9 mg/g [1]; white chicken eggshell flour contains 113 mg/g calcium and 108 mg/g brown color [7]; calcium 2300.33 mg/L, magnesium 850.00 mg/L [2]; and 99% calcium carbonate content [8]. Calcium from eggshell flour has many advantages over synthetic calcium carbonate. It is more easily absorbed in the small intestine of mice [9]. The solubility of calcium carbonate from chicken eggshells is higher than from oyster shells. Besides, the presence of valuable mineral components (strontium, barium) makes it an excellent biomaterial for the production of dietary supplements [10].

Broiler chicken eggshells can be used very effectively in a variety of applications [11]. Tablets from broiler chicken eggshell powder were tested for equality with calcium supplement CIPCAL-500 tablets from oyster shell. The source, availability, and manufacture of calcium carbonate tablets using chicken eggshells are much easier than oyster shells which are the primary source of CIPCAL-500 tablets.

The research on the kinetics of calcium release in artificial stomachs shows that calcium is released more quickly from tablets containing calcium citrate from eggshells than synthetic calcium carbonate [12]. With the many advantages and benefits of eggshells, it can be an

alternative biomaterial that is renewable, cost-effective, and available. Poultry eggshells have a physical structure that is hard, rough, smells fishy, and has a less attractive color so that it is less desirable when used as food. Soaking using chemical solvents can improve the physical structure of the shell and reduce harmful components and remove organic compounds from the shell. One way to optimize the calcium content of eggshells is to use acetic acid [13].

An acid solution as an immersion medium initially causes complex minerals (bond with other components) to change into simple forms (ions), so they will dissolve more easily. In this case, CH_3COOH acts as an enhancer, namely a molecule or compound that affects the shape and level of solubility of minerals. HCl is a strong acid that is least dangerous to handle than other strong acids because it contains chloride ions that are non-reactive and non-toxic. Concentrated hydrochloric acid dissolves many types of metals and produces metal chloride and hydrogen gas. It can also increase the solubility of a compound that is difficult to dissolve in water.

Each solvent has the same principle, and it will cause the shell's pores to open. Hence, the spaces may easily be reached by the solvent, which results in the compounds that bind to minerals quickly released optimally [14]. Research on the preparation of eggshell extract by extracting calcium in eggshells as calcium chloride using 4% (w/v) HCl solution for 3 h shows that the ratio of eggshell to HCl was 1:15 (w/v). After hydrolysis, the residue was removed by centrifugation at $\times 1774 \text{ g}$ for 10 min, and the solution was heated at $110\text{--}115^\circ\text{C}$ until dry. The results obtained were calcium chloride powder containing 87.38% (w/w) calcium chloride, protein 0.3%, ash 94.37%, and pH 5.27 and showed high solubility [15].

Calcium from eggshells can be produced by extraction using HCl [16]. Calcium carbonate in eggshells will react with HCl to form soluble calcium chloride, CO_2 , and H_2O gases. The best treatment was obtained from the addition of 2.5% HCl concentration to produce a chicken eggshell extract with a water content of $1.19 \pm 0.06\%$, 91% ash, 28.17% calcium, 98.4% CaCO_3 , and 0.8% $\text{Ca}_3(\text{PO}_4)_2$ with low solubility.

Based on the background and several previous research, the physicochemical composition of chicken eggshell flour generated by hydrochloric acid (HCl) and acetic acid (CH_3COOH) is still diverse. It is necessary to find out the proper solvent that produces better chemical characteristics (moisture, ash, calcium, and phosphorus) and physical (yield) eggshell extracts.

Methods

Research type and stages

This type of research was descriptive quantitative. The research stages included preparing

chicken eggshell samples and determining the chemical characteristics (moisture content, ash, calcium carbonate, calcium, and phosphorus) and physical characteristics (yield) of eggshell flour. This research was conducted in May - June 2019 at the Makassar Health Laboratory Center.

Materials and tools

The materials used in this study were chicken eggshells obtained from restaurants around the Hasanuddin University Campus in Makassar, 4% hydrochloric acid (HCl), CH_3COOH 2N, aquadest, and standard calcium solution. Equipment used during this study was including digital scales, basins, knives, winnowing pan, gas stove, pan, water bath, 1 L cup, electric oven, blender, 80 mesh filter, glass jars, electric furnaces, desiccators, porcelain cups, tube clamp, beakers, measuring flasks, Erlenmeyer, atomic absorption spectrophotometer (AAS), and UV-vis spectrophotometer.

Making flour from extracts of boiler chicken eggshells

The working method of extracting eggshell flour using a modified 4% HCl solvent [11] and solvent extraction of CH_3COOH 2N from [13].

Eggshell flour extraction using 4% HCl solvent

Boiler chicken eggshells were washed using running water, then rinsed with aquadest. Then boiled in boiling distilled water for 15 min, drained, and spread on a stainless steel pan, dried overnight by aerating it.

In the morning, chicken eggshells were dried in an oven at 200°C for 10 min, cooled, then mashed using a blender to become a powder and then filtered with an 80 mesh sieve. Simplicia chicken eggshell powder then packed in airtight packaging, ready to be extracted.

Sample extraction was conducted by maceration method using 4% (w/v) hydrochloric acid (HCl) solvent to attract the chemical components for 3 h. The comparison of the simplicity of chicken eggshells with HCl was 1:15 (w/v). After the sample was macerated for 3 h, the macerate was then filtered with filter paper. The residue and HCl solution was removed by heating in an oven at $110\text{--}115^\circ\text{C}$ until dry (3 h). After cooled, then mashed using a blender, then filtered (80 mesh), packed in airtight packaging.

Eggshell flour extraction using CH_3COOH 2N solvent

Chicken eggshells were washed clean, reduced in size, then immersed in aquadest at

100°C for 15 min, drained, then added 2N acetic acid (CH₃COOH) (shell: solvent = 1:2 (w/v)). After, it was heated in a water bath at 60°C for 3 h, removed, drained, washed with aquadest, and dried using a cabinet dryer at ± 50°C for 3 h. After chilling, mashed in a blender and filtered using an 80 mesh sieve, packed in airtight packaging.

Analysis of physicochemical characteristics

Methods for analyzing water content (AOAC, 925.09, 2005), ash content (AOAC, 941.12, 2005), calcium carbonate (AOAC, 925.09, 2005), calcium (AOAC, 968.08, 2005) using AAS at $\lambda = 422,7$ nm, phosphorus (AOAC, 948.09, 2005) using a UV-vis spectrophotometer at $\lambda = 660$ nm, and yield (AOAC, 2005). Measurement of the chemical characteristics of the eggshell extract was carried out in a triplo.

Data analysis

Processing and data analysis was conducted using the IBM SPSS statistics program.

Results and Discussion

The results of making eggshell extracts of broiler chickens using 2N acetic acid (CH₃COOH) and 4% hydrochloric acid (HCl) solvents can be seen in Figure 1.

The average physicochemical characteristics of chicken eggshell powder extracted by 2N acetic acid (CH₃COOH) and 4% hydrochloric acid (HCl) can be seen in Table 1.

Table 1: Physicochemical characteristics broiler eggs eggshell extract solvent extraction results HCl 4% and 2 N CH₃COOH

| Parameter (%) | Mean ± SD | |
|-------------------|--------------|-------------------------|
| | HCl 4% | CH ₃ COOH 2N |
| Calcium | 35.46 ± 2.04 | 27.41 ± 2.56 |
| CaCO ₃ | 95.54 ± 1.16 | 94.75 ± 2.28 |
| Phosphor | 0.61 ± 0.04 | 0.52 ± 0.08 |
| Water | 0.58 ± 0.06 | 0.75 ± 0.72 |
| Ash | 86.48 ± 0.62 | 79.18 ± 1.27 |
| Rendement | 81.24 | 75.33 |

Moisture content (AOAC, 925.09, 2005)

The water content of the eggshells extracted from the 4% HCl (0.58% ± 0.06) was lower than that of the CH₃COOH 2N (0.75% ± 0.72). The addition of 2.5% HCl concentration resulted in chicken eggshell flour with a water content of 1.19 ± 0.06% [16]. The water content of chicken shells was 0.95 ± 0.09% [2]. The shell characteristics caused the relatively low water content of the eggshells flour has a dense texture and is composed of lime. Low water content in the resulting chicken eggshell flour can extend shelf life with good quality. High moisture content tends to promote microbial contamination and chemical degradation in many substances [17].

Ash content (AOAC, 941.12, 2005)

The ash content of the eggshells from the 4% HCl extraction was higher (86.48% ± 0.62) than CH₃COOH 2N extraction (79.18% ± 1.27). Eggshell flour extracted by 4% HCl had an ash content of 94.37% [15]. The addition of 2.5% HCl concentration resulted in chicken eggshell flour with an ash content of 91% [16]. The results of this study are different from the results of other studies reveal the ash content of chicken eggshells of 45.29 ± 0.06% [2].

The ash content of the material shows the number of minerals in that material. The ash content is influenced by the type of shell used as the raw material.



Figure 1: Extract of chicken eggshell from maceration of 4% HCl and CH₃COOH 2N

Ash content measures the total amount of inorganic compounds such as minerals in eggshells [17]; [18]. The high ash content in the eggshell flour is thought to be caused by the high mineral content. In eggshells, there is calcium (CaCO_3) as the main constituent of eggshells.

Calcium carbonate levels

Calcium carbonate in the eggshell of broilers from the extraction of 4% HCl was higher ($95.54\% \pm 1.16$) than the extract from CH_3COOH 2N ($94.75\% \pm 2.28$). Eggshell flour extracted from HCl 4% had a calcium chloride content of 87.38% (w/w) [15]. The addition of 2.5% HCl concentration resulted in chicken eggshell flour with 98.4% calcium carbonate CaCO_3 content [16]. Eggshell flour extracted by 4% HCl had a calcium carbonate content of 85% [11].

Calcium levels (AOAC, 2005)

The calcium content of the eggshell from the extraction of the 4% HCl was higher ($35.46\% \pm 2.04$) than CH_3COOH 2N ($27.41\% \pm 2.56$). The addition of 2.5% HCl concentration resulted in chicken eggshell flour with a calcium content of 28.17% [16]. Eggshell flour extracted by 4% HCl had a calcium content of 38% [11]. Chicken eggshell flour extracted from CH_3COOH 2N had different calcium levels of 20.67% and 6.41% [13].

Research on the comparison of calcium levels from various poultry eggshells shows that the average calcium content of poultry eggs extracted with acetic acid ranges from 20.67 to 25.73% [19]. The research results on calcium testing using the AAS method showed that in 100 g of eggshell flour, there was 7.2 g of calcium (7.2%) [20], [21]. Calcium from eggshells consists of 98.4% CaCO_3 and 0.8% $\text{Ca}_3(\text{PO}_4)_2$ with low solubility [16]. The high content of calcium in chicken eggshells is expected to meet the calcium needed by the living body through fortification and supplementation [22].

Phosphorus (P) content

The phosphorus content of chicken eggshells extracted from 4% HCl ($0.61\% \pm 0.04$) was higher than that of CH_3COOH 2N ($0.52\% \pm 0.08$). The addition of a 2.5% HCl resulted in chicken eggshell flour with a $\text{Ca}_3(\text{PO}_4)_2$ content of 0.8% [16]. Chicken eggshells consist primarily of calcium carbonate and a small portion of phosphate. Phosphorus is not formed freely in nature and is available in the form of phosphates and orthophosphates. Phosphorus is a mineral that is relatively abundant in the animal body.

Phosphorus is the second most abundant mineral after calcium in the body and, together with calcium, is bound in the bone framework. The relationship between calcium and phosphorus in plasma

has a very significant correlation, which is 0.998. The ratio of calcium and phosphorus to plasma for bone formation ranges from 1.5: 1. The results of this study indicated the ratio of Ca: P for the extraction of HCl 4% = $35.46\% \pm 2.04$: $0.61\% \pm 0.04$, while the extraction of CH_3COOH 2N yields a ratio of Ca: P = $27.41\% \pm 2.56$: $0.52\% \pm 0.08$.

Yield (AOAC 2005)

The yield is the percentage of eggshell flour calculated based on the ratio between the eggshell flour produced and the weight of raw materials (chicken eggshells) that have been cleaned. The more yields are produced, the more efficient the treatment is applied. This study obtained the yield value of chicken eggshell powder extracted from 4% HCl was higher (81.24%) than CH_3COOH 2N (75.33%).

The results of other studies showed that the average yield of eggshell flour extracted with acetic acid (CH_3COOH 2N) ranged from 68.97% to 88.83% [13]. Processing of poultry eggshells into shell flour by immersion method (distilled water, HCl, CH_3COOH , and NaOH) had a high yield, ranging from 78.37% to 98.62% [19].

Conclusion

The chemical and physical characteristics of chicken eggshell powder extracted by 4% HCl were better than 2N acetic acid (CH_3COOH). It can be seen from the higher levels of calcium, calcium carbonate, phosphorus, ash, yield, and lower water content than the eggshell flour extracted by 4% HCl compared to the 2N of acetic acid (CH_3COOH).

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