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Development of Pastilles containing Fermented Garlic to Improve Acceptability and Health Benefits for the Elderly in a Post-Vaccination Program

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Abstract

Wild foods are a source of nutrients. Garlic is a plant that contains some bioactive components. Fermented garlic can be used as a raw food material to formulate healthy food. Eating functional foods, such as Pastilles which include fermented garlic, can boost the body's immune. The purpose of this study was to analyze preference acceptance of and nutrition values in pastilles. This work applied a randomized experimental study design. Organoleptic properties with the hedonic tests were conducted using Friedman and Wilcoxon signed-rank tests. Results showed that formula F1 (pastilles with sago flour and mint essence) was the most preferred level ingredient for the pastilles with the highest average score is 3.57. The content of sago powder and mint essence significantly affected the smell and taste of the pastilles ($P<0.05$). The addition of mint essence has been shown to increase the acceptance of fermented garlic pastilles with an above 40% score. Pastilles made from fermented garlic also offered enough nutrients and bioactive compounds. The conclusion of this study shows that pastilles with FG can be well received by respondents and have been tested to have bioactive content.

Keywords

Bioactive
Fermented garlic
Organoleptic
Pastilles

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Introduction

We were currently in a state of crisis due to a global pandemic caused by the coronavirus infection. Coronavirus, also known as COVID-19, was a specified virulent disease dependent on the human immune system. The strong antioxidant content of native foods like garlic is highly beneficial for boosting immunity. Garlic (*Allium sativum L*) has been used for centuries in many societies to combat parasitic, fungal, bacterial, and viral infections (Citarasu *et al.*, 1999). Garlic is effective as a blood acid-lowering agent (Sumiyoshi, 1997), antibacterial (Kumar & Berwal, 1998), antihypertensive (Suetsuna, 1998), hepatoprotective, and insecticide (Wang *et al.*, 1998) in various human and animal therapies. It has been reported that the use of garlic extracts reduced serum cholesterol levels (Augusti, 1977) and increased clotting blood time (Bordia *et al.*, 1975). These effects of garlic were due to the presence of various organosulfur compounds, such as allicin (Augusti & Mathew, 1974). Allicin is the most potent component present in garlic with active and direct antiparasitic effects (Adler & Holub, 1997). The majority of garlic-containing foods are capable of enhancing nutritional value. Some consider garlic as an immune system booster and compare it to vitamin C in humans (Adetumbi *et al.*, 1986; Shakya & Labh, 2014). Many scientific studies have shown that allicin can actively destroy a wide range of pathogens such as fungi, bacteria, and even viruses (Nya & Austin, 2009). It was a proven immunostimulant and anti-infective (Reuter, 1996). *Allium sativum* L species have immune-enhancing activities such as promoting lymphocyte synthesis, cytokine release, phagocytosis, and natural killer cell activity (Kyo *et al.*, 1998).

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Efforts to increase garlic consumption encourage the creation of ways that can increase the convenience of consuming garlic directly. The physicochemical properties of garlic occur during the fermentation process, where its appearance eventually turns black. Compared with fresh garlic, fermented garlic does not have a strong smell due to the reduced allicin content (Zhang *et al.*, 2015). Fermented garlic/black garlic has better known for its preferred organoleptic properties and benefits (Azizah *et al.*, 2020; Wang *et al.*, 2010). The transition from garlic to black garlic leads to an increase or decrease in the components present in black garlic (Zhang *et al.*, 2016). Black garlic contains several some many nutrients that provide therapeutic potential. The benefits of black garlic include antioxidants (Azizah *et al.*, 2020) and immunomodulatory (Wang *et al.*, 2010) can protect and maintain the body. The immunomodulatory properties of black garlic were shown by regulating cells involved in the immune system. A properly functioning immune system optimizes its function (Wang *et al.*, 2010). The immunomodulatory effects of black garlic were related to the components it contains. Compounds in black garlic can inhibit and prevent the attack of pathogens (Wang *et al.*, 2010). Black garlic can have a beneficial effect on the body's health, especially in protecting the body from threats that interfere with the body's immune system. However, the acceptance of black garlic is still quite low. One of the antioxidants contained in black garlic is flavonoids. Flavonoids have many funcitons as an antioxidant, anti-inflammatory, and neuroprotective effects (Fan *et al.*, 2022). The world means total flavonoid intake is reported to be about 400 mg/day and ranges from 150 to 600 mg/day (Escobar-Cévoli *et al.*, 2017). In addition, other bioactive ingredients such as polyphenols also have benefits. As an antioxidant, polyphenols protect cellular components from oxidative damage and therefore Limit the risk of various degenerative diseases associated with oxidative stress (D'Archivio *et al.*, 2007). Innovation in black garlic requires innovative, healthy, and delicious products. One of the product development strategies is to produce pastilles containing fermented garlic. Pastilles were soft candy but sweeter than gum, and has a rubber effect using a piece of gum and gelatin (Zainol *et al.*, 2020). They were produced for medicine applications to relieve sore throat, cough, and oral thrush while pharmaceuticals were extremely uncomfortable to consume into the eighteenth century (Lubbers & Guichard, 2003). The advantages of pastilles were their long shelf life and reduced susceptibility to microbial decay because of their high sugar content. (Ramlan *et al.*, 2021; Subramaniam, 2016). Pastilles have a long life because of low water activity (Basiri, 2020). Pastilles were known to be mainly consumed by kids but also can be used by the elderly, because of their relaxed form, their structure, and their strong flavors, with the feeling of the elderly. The growing awareness of the importance of maintaining the body's immune system is pushed by increasing the finding of substances that can maintain the body's immune system (Wang *et al.*, 2010). Natural ingredients have long been used to maintain the immune system of the body, especially those with immune antioxidant and immunomodulatory properties (Adi *et al.*, 2019). Garlic has been reported as a plant with antioxidant and immunomodulatory properties (Singh & Singh, 2019). In the produce of pastilles, sago flour is also needed for base material. Sago mostly contains starch and complex carbohydrate (ANJ internal research, 2017). Sago flour is an alternative food raw material that can be developed into other types of food products such as pastilles. Sago starch has been analyzed with gluten-free contents. Gluten has been suggested for people with an auto-immune conditions such as elderly people (ANJ internal research, 2017). In addition, the addition of porang flour substitution was used to improve the texture of the candy. The use of porang flour is based on the glucomannan content in it which is useful for increasing the elasticity of the texture of a product including pastilles. Porang contains glucomannan with 15-64% (dry base) (Supriati, 2016). It also contains other carbohydrates, such as starch, polyose, and crude fiber which are approximately 2, 14, and 8.0%, respectively (Ohtsuki, 1968). The high content of glucomannan or other polysaccharides in porang is become potential to be developed in the food industry and health science (Astuti *et al.*, 2017; Zhang *et al.*, 2005). The purpose of this research is to see the acceptability of the formulated pastilles products along with the health benefits the elderly people will get.

Materials and methods

Research design

This study applied a randomized experimental research design with six redundancies. Distinctive pastilles equations were created utilizing different substances of porang flour and mint essence. The variable parameters in this work were a color, smell, taste, and texture properties. Panelists were asked to fill in informed consent before carrying out the organoleptic test. The relevant procedures were also approved by Universitas Airlangga Faculty of Dental Medicine Health Research Ethical Clearance Commission (number: 295/HRECC.FODM/VI/2021).

Development of pastilles containing fermented garlic

All cooking processes, including the preparation of fermented garlic, porang flour, and all the pastilles formulas were conducted in the Nutrition Laboratory, Nutrition Department, Faculty of Public Health, Universitas Airlangga, Surabaya, Indonesia. Four formulas were developed in this study: F0, F1, F2, and F3 (Table 1).

Table 1. The formulas of pastilles containing fermented garlic

Ingredient	F0		F1		F2		F3	
	g	%	g	%	g	%	g	%
Fermented garlic	91.20	92.30	91.20	91.20	91.20	91.20	91.20	92.30
Sago flour	5.70	5.80	5.70	5.70	0.00	0.00	0.00	0.00
Porang flour	0.00	0.00	0.00	0.00	5.70	5.70	5.70	5.80
Citric acid	0.00	0.00	0.60	0.60	0.60	0.60	0.00	0.00
Mint Essence	0.00	0.00	0.60	0.60	0.60	0.60	0.00	0.00
Honey	1.90	1.90	1.90	1.90	1.90	1.90	1.90	1.90
Sucralose	0.00019	0.0001	0.00019	0.00019	0.00019	0.00019	0.00019	0.0001

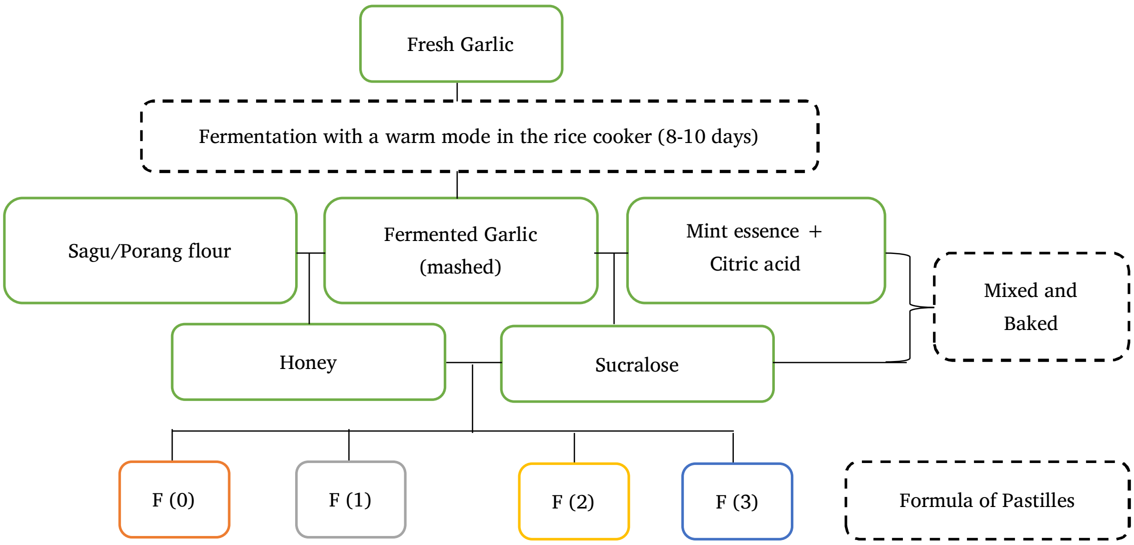

Fig. 1. Production Process of Pastilles.

The percentages of fermented garlic added to pastilles were based on the quantity to show the target health benefit, especially the bioactive compound. Organoleptic tests with untrained panelists were conducted on an elderly communities living in Mulyosari, Surabaya, East Java, Indonesia. Inclusion criteria for organoleptic assessments included elderly people aged >50 years, ability to write and read, and exclusion criteria included color-blindness.

To prepare fermented garlic (Fig. 1), garlic (*Allium sativum)* was fermented for 8 days using a rice cooker in heat mode (60-70 ^{0}C). The fermented garlic was then crushed using a blender. Table (1) shows the composition of each formulation of pastilles containing fermented garlic. All ingredients shown in Table (1) were mixed in a plastic bowl. Further, each part of the mixture was pressed using a rolling pin, and molded manually using a cake mold. The pastilles were baked (60 ^{0}C) for 40 min to reduce the water content. Finally, the pastilles were sprinkled with sugar.

Study procedure description

Organoleptic evaluations, including the color, smell, taste, and texture properties, was conducted using a questionnaire form with a 5-point hedonic scale, where the samples were scored as follows; 5 = very like; 4 = like; 3 = fair; 2 = dislike, and 1 = worst. The organoleptic evaluation was done by a panel including (or composed of) 30 untrained elderly (age >50 years). Panelists were instructed to drink water before consuming each pastilles formula. Analysis of organoleptic results was carried out for the best formula chosen by the panelists, at the Nutrition Laboratory, Department of Nutrition, Faculty of Public Health, Universitas Airlangga, Surabaya, Indonesia.

Nutrition and Bioactive Analysis

Proximate compositions of energy, protein, fat, and carbohydrate were determined using the Association of Official Analytical Chemistry (AOAC) standardized methods. Polyphenolic compounds were quantified with Folin Ciocalteau 2N reagent; the absorbance was estimated at 765 nm, and the outcomes were described in mg gallic corrosive/100 g dry example (Waterhouse, 2002). With an Evolution 201 spectrophotometer and the absorbance at 515 nm was estimated to determine the amount of eliminated anthocyanins (Thermo Fisher Scientific, Waltham, MA, USA). The total anthocyanin content was determined with an annihilation coefficient of 27300 (Jansen & Flamme, 2006).

Statistical analysis

The organoleptic data were tested for normality and homogeneity of variance before submitting them to Analysis of Variance (ANOVA). If a difference was identified, analysis was continued by Fisher's least significant difference at a significance level of $P \leq 0.05$. However, when the assumptions of normality and homogeneity did not meet, the data were analyzed using the Friedman test. When significant differences occurred, means were separated using Wilcoxon signed-rank tests ($\alpha = 0.05$). Statistical analysis was performed using IBM Statistics SPSS 22 (IBM Corp., New York, NY, USA).

Results and discussion

The hedonic test was used to measure the organoleptic properties of the 4 formulas. The average score of each formula is shown in Table (2). Each formula is calculated by the average general acceptability, which includes color, aroma, taste, and texture to determine the best formula. It seems that the higher level score of the formula was F1 score (3.57) which contained sago flour and mint essence improves the taste and texture properties, while the lowest average was for F2 with a value of 3.37.

Table 2. The hedonic test result of pastilles containing fermented garlic

Indicator	Formula			
	F0	F1	F2	F3
Color	3.27	3.30	3.17	3.23
Smell	3.60	3.60	3.40	3.57
Taste	3.73	3.77	3.47	3.50
Texture	3.57	3.60	3.43	3.33
Average	3.54	3.57	3.37	3.41
St.deviation	0.194	0.196	0.135	0.155

Color

Most of the panelists gave a score of 3 on each formula. While the most widely accepted formula based on the color aspect was F1 with an average score of 3.30, with 20% of the panelists giving a score of 5 or very like (Fig. 2).

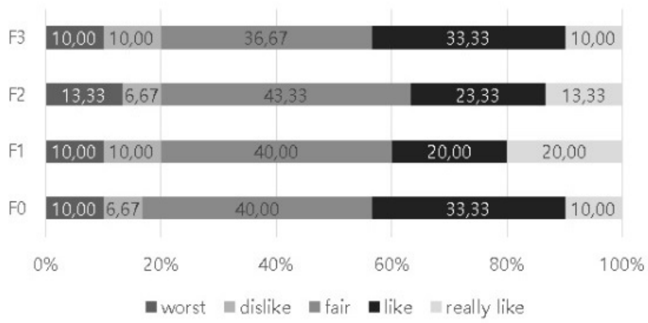

Fig. 2. The level of color preference of pastilles containing fermented garlic.

Smell

Formulas F0 and F1 were most accepted by most panelists with the same hedonic test average value (3.60). The category of really like from the hedonic test between F0 and F1 have the same percentage too (10% panelists), while the most chosen by the panelists was F3, which was chosen by as many as 16.67% of the panelists (Fig. 3).

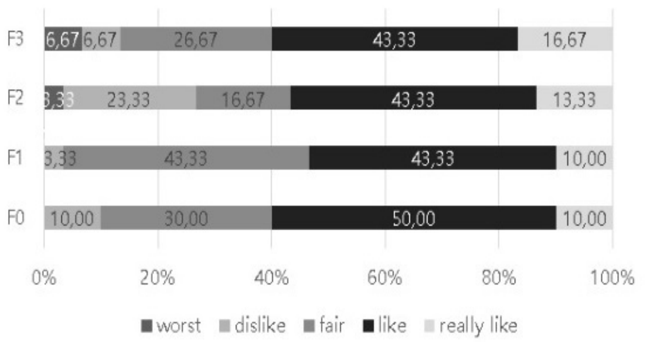

Fig. 3. The level of smell preference of pastilles containing fermented garlic.

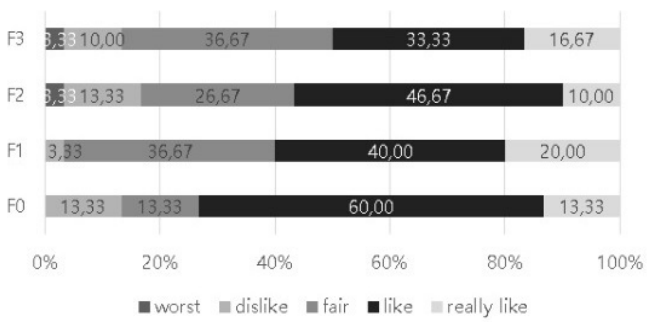

Fig. 4. The level of taste preference of pastilles containing fermented garlic.

Taste

Pastilles fermented garlic with mint essence (F1) showed the highest score for taste (3.77; Table 2) and gained the most positive response (20% likes). This response was followed by F3 (16.67% like), whose formula was composed of porang flour and without the addition of mint essence. No one voted worst for F0 and F1 (Fig. 4).

Texture

Formula with sago flour and mint essence (F1) was the most accepted texture voted by the panelists, with a score of 3.60 (Table 2). F0 ranked 2 for the texture, with a value that is not much different from F1, which is 3.57. The difference was seen in the number of panelists who voted dislike, which is 3.33% for F1 and 13.33% for F0 (Fig. 5).

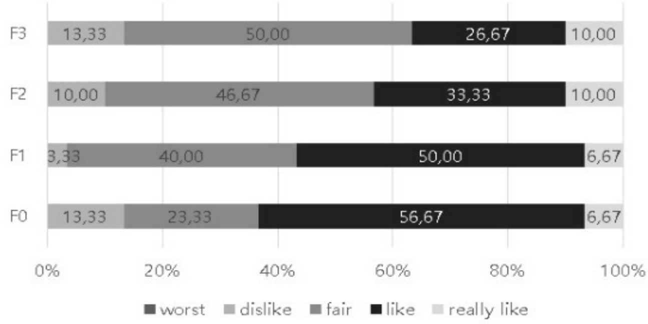

Fig. 5. The level of texture preference of pastilles containing fermented garlic.

Table 3. The Statistical analysis test result of pastilles containing fermented garlic (*p-value*)

Indicator	Formula		
	F1	**F2**	**F3**
Color	0.981	0.697	0.913
Smell	0.947	0.587	0.887
Taste	0.973	0.244	0.283
Texture	0.902	0.365	0.172

Statistical analysis using the friedman test was performed to assess the difference in responses between color, smell, taste, and texture responses (Table 3). The results show no data were significantly different for all category. This result also demonstrates the panelists' formula preferences. Based on the mapping shown in Fig. (2) to (5), F1 was the most accepted formula with an average score of 3.57 and had good acceptance in terms of every category (color, smell, taste, and texture).

As described above, pastilles containing fermented garlic with different filler quantity of sago flour and porang flour has acceptable organoleptic properties, including color, smell, taste, and texture.

Color is undoubtedly a most important product-intrinsic sensory cue that affects people's food and beverage consumption (Spence, 2015). In all formulas, the colors displayed no difference, they all have a homogeneous black color because of the original color of fermented garlic. The transformation of fresh garlic to fermented garlic produces black garlic with dominant colors of all formulas (Fig. 3)

The texture of pastilles was influenced by the addition of sago and porang flour. According to the textural comparison, the formula added with porang flour looks better than sago flour. This can be seen through the analysis of the texture data obtained showing that the pastilles with porang flour look more elastic. The fermentation of black garlic undergoes polysaccharide cell wall degradation causing tissue softening so that the texture of black garlic resembles gum. Formula with porang flour resulted in product more elasticity than sago flour. This is for improving physical performance but not for influencing the nutritional content. Porang tubers contain calcium oxalate crystals which in the extraction process produce a compound called glucomannan. The content of glucomannan in porang tubers is known as Konjac Glucomannan (KGM). KGM is a soluble polysaccharide dietary fiber that is low in calories. The addition of konjac functions as a gelling agent, thickener, emulsifier, and product stabilizer (Karo *et al.*, 2021). Based on these advantages, it is expected that konjac can be used as a substitute for sago flour in increasing elasticity and maintaining the texture of pastilles. However, the results of the organoleptic test, the formula with the addition of porang flour (F2 and F3) did not show a better acceptance of texture parameters than the formula with sago flour.

In terms of taste and smell, pastilles fermented garlic supplemented with sago flour F1 was given the highest score. This indicates that the addition of mint essence improves the acceptability of both organoleptic properties. Fermented garlic has a distinctive sharp taste and aroma, along with this, mint is also an ingredient that has a distinctive and strong character. The addition of mint essence has been shown to increase the acceptance of fermented garlic pastilles in terms of taste and aroma. This is due to the ability of mint essence to improve the sharp taste and aroma. So that the panelists will more easily accept the taste and aroma of mint which is already familiar to the panelists. This is also reinforced through research conducted by Bajaj and Urooj which explains the good reception with the addition of mint in food products (Bajaj *et al.*, 2006).

In the sum of organoleptic characteristics (color, smell, taste, and texture), all formulas (F0-F3) have good acceptability scores (score 4: like). Meanwhile, the most preferred pastilles of all the parameters by the panelists were pastilles containing fermented garlic with sago as a filler and mint essence added (formula 1). In addition, F1 also has a better reception when compared to F0. This is in line with review conducted by Kimura et al. which explains that black garlic or fermented garlic has quite a lot of advantages in terms of acceptance because it has been proven to be better in taste when compared to fresh garlic. Black garlic tends to have a sweeter taste than fresh garlic (Kimura *et al.*, 2017).

Nutrition and Bioactive Value

Pastilles containing fermented garlic contained macronutrients and micronutrients in addition to other bioactive components (Table 4). As shown in Table (4), each formula shows a similar bioactive compound. The only difference in nutritional content was carbohydrates and fiber. The difference value in the fiber content, is 1.8 to 1.9 g per 10 g of pastilles. The fiber content has been fulfilled by 7.6% of the total RDA of fiber for the elderly per day. The difference in the amount of fiber content is influenced by the use of porang flour. The flavonoid content in pastilles fulfills about 8.2% of the average minimum daily intake requirement. There is no reference to the recommended daily intake of polyphenols that mentions the minimum amount of intake, but there are several studies that explain the average daily intake of polyphenols. The average intake of polyphenols based on several studies shows levels ranging from 900-3000 mg/day (Saura-Calixto *et al.*, 2007; Taguchi *et al.*, 2017). When compared to pastilles products, the polyphenol was fulfilled only about 5.3% of the average daily intake.

Table 4. The nutrition and bioactive content of pastilles containing fermented garlic per portion (10 g)

Nutrition/bioactive component	Formula			
	F0	F1	F2	F3
Energy (kcal)	29.70	29.70	29.10	29.10
Protein (g)	1.00	1.00	1.10	1.10
Fat (g)	0.10	0.10	0.10	0.10
Carbohydrate (g)	6.00	6.00	5.80	5.80
Fiber (g)	1.80	1.80	1.90	1.90
Polifenol (μgGAE/g)	48.10	48.10	48.10	48.10
Flavonoid (μgQE/g)	12.40	12.40	12.40	12.40

Conclusion

The acceptance through the organoleptic test of pastilles containing fermented garlic showed that the formula is liked by all panelists. The addition of sago powder and mint essence to the formula can increase the acceptance of the formula from all parameters (colors, smell, taste, and texture), so that pastilles containing fermented garlic with sago powder as a filler and mint essence (F1) was the most preferred formula. Pastilles containing fermented garlic provided enough nutrients such as protein, vitamins A and C, calcium, and zinc, with a bioactive components such as polyphenols, and flavonoids. Pastilles serve as one of the promising functional snacks for the elderly during the pandemic COVID-19, with the recommended daily consumption portion of 10 g or 4 grains of pastilles in a day.

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