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PREFACE

By the Grace and Blessings of Allah the Almighty, we would like to present, with great pleasure, the Volume 04 number 02 of *Food ScienTech Journal* (FSJ). This journal is part of the Universitas Sultan Ageng Tirtaya series of journal.

This journal was envisioned and founded to represent the growing needs of food technology as an emerging and increasingly vital field, now widely recognized as an integral part of agriculture and human living. Its mission is to become a voice of the food technology and science community, addressing researchers and practitioners in areas ranging from chemistry to management, from microbiology to industry, presenting verifiable methods, findings, and solutions.

The journal is intended as a forum for practitioners and researchers to share their research, idea, and solutions in the area of food science and technology. We would like to request for the reader to participate on writing the articles in this journal.

Thank you for your kind attention and support, hopefully this journal will provide lots of benefits for you and society.

Serang, December 2022

Editorial Team

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Differences in Eating Habits, Lifestyle, and Weight Changes of Indonesian College Students Based on Their Residence During The COVID-19 Pandemic

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ABSTRACT

The COVID-19 pandemic has indirectly forced people to adapt to the new normal, doing activities outside the home with proper health protocols or not leaving the house to reduce the spread of the virus; this also affects food consumption and sedentary behavior. For some people, the pandemic causes anxiety and even stress. The design of this study was cross cross-sectional, using a survey method online with Google Form carried out from April to September 2021. Descriptive analysis was conducted on 1185 respondents who are currently studying at universities in Indonesia., The results of the study stated that the pandemic had an impact on Indonesian students. This survey researched the differences in eating habits, changes in body weight, and lifestyle in Indonesian students based on where they lived during the Covid-19 pandemic. From the results of data analysis, it is known that most of the respondents experienced changes in body weight during the Covid-19 pandemic, whether they lived together or alone. However, more than half of the respondents have a habit of weighing their body weight irregularly. The results showed that the COVID-19 pandemic influenced eating habits, lifestyle, and changes in body weight. The community, especially students, both living alone (dormitory/boarding/apartments) or living with family or relatives should have sufficient knowledge and attitudes about nutrition to maintain unwanted weight changes during the pandemic.

Keywords: Indonesian students, pandemic impact, lifestyle, weight

INTRODUCTION

In March 2020, the World Health Organization (WHO) officially declared COVID-19 a pandemic. COVID-19, which stands for Corona Virus Disease, is a new infectious disease that spreads easily and quickly, thus requiring people to reduce activities outside the home, including students (Mattioli A V., Sciomer S, Cocchi C, Maffei S, 2020). Restrictions on physical activity outside the home implemented by the government and the existence of Distance Learning (PJJ) have led to changes in lifestyle, eating patterns, and people's weight, one of which is students (Pecanha T, Goessler KF, Roschel H, 2020). Due to the existence of PJJ, students return to their homes or hometowns, although not a few students remain in their dormitories or boarding houses.

The study's results stated that due to restrictions on activities outside the home, people tend to experience decreased physical activity and increased sedentary behavior. A sedentary lifestyle can increase appetite and ultimately change one's eating habits (Abidah *et al*., 2020). Changes in eating habits tend to lead to overeating with choices of high-calorie foods, fried foods, and sweet foods (Ruiz-Roso *et al*., 2020). A study on university students in Texas, United States showed that 34.5% (n=502) students experienced food insecurity during the pandemic, where the main cause was changes in current residential arrangements (OR = 2.70, 95% CI: 2.47, 2.95), given leave (OR = 3.22, 95% CI: 2.86, 3.64), was dismissed from work (OR = 4.07, 95% CI: 3.55, 4.66), or lost part-time job (OR = 5.73, 95% CI: 5.09 , 6.46) (Owens *et al*., 2020).

Not only in terms of food consumption and sedentary behavior, but the COVID-19 pandemic has also indirectly forced people to adapt to the new normal, doing activities outside the home with proper health protocols or not leaving the house to reduce the spread of the virus. For some people, the pandemic causes anxiety and stress (Ros Maria and Raharjo, 2020). The research results on students stated that out of 160 respondents, 84% felt stressed during the COVID-19 pandemic. Students feel cooped up at home and can't do anything other than lie down, watch, and do the online learning process (Gimon, 2020). It triggers a person to eat more and have sedentary behavior, affecting body weight.

Based on this background, researchers are interested in conducting survey research on college students studying in Indonesia regarding differences in eating habits, changes in body weight, and lifestyle in Indonesian students based on where they lived during the Covid-19 pandemic.

MATERIALS AND METHODS

This research was conducted on college students in Indonesian universities as the population by accomplishing an online survey using Google Form from April to September 2021. This study has passed ethical approval No:018/S.Ket/ KEPK/LPPM/III/2021 by Research Ethics Commission of Universitas Mohammad Husni Thamrin.

Based on sample calculations using the proportion estimation formula, this study's minimum number of samples was 382 respondents. Using the technique of sampling Snowball, we obtained a sample of 1185 respondents. There were 1186 responses recorded in the online form. Data cleaning was conducted to omit one respondent who did not complete the form. The instrument used in this survey consisted of 60 questions covering respondent characteristics, consumption aspects (changes in eating behavior, snack habits, breakfast habits, consumption of vitamins & supplements), then lifestyle aspects (exercise habits, sender behavior, stress), and changes in body weight (weight gain, routine weighing habit). Data analysis was conducted using the cross-tabulation method using SPSS.

RESULTS AND DISCUSSION

The research was conducted on all Indonesian students online. From the data collection results, which took about 35 days, there were 1185 respondents. The following presents the results of univariate data analysis based on the study's results (Table 1). It is delivered using columnar percentages to compare the proportion of characteristics and variables based on their place of living.

Respondents involved in this study were dominated by women, namely 71.1%, with the age of being in the late adolescence group being 91.2%. Research conducted by

Bolang *et al*. (2021) stated that the COVID-19 pandemic caused a change in the nutritional status of students. From the results of his research, it was found that male students had a higher average body weight during the Covid-19 pandemic than women, as well as their nutritional status. Many late teens and young adults experienced increased body weight and nutritional status during the pandemic compared to before the pandemic (Huber *et al*., 2021). However, in this study, female students experienced weight changes, while Huber (2021) found the opposite. As for the reason of women experience more weight changes than men, when they are bored, women tend to consume more staple foods and snacks (Mustofa, Husna, Hermawan, Langki, 2021).

Most Indonesian students in this study were in the western part of Indonesia, namely 88.2%. This is because we include college students, and the universities are scattered mainly in the Western Part of Indonesia. Still, there were also respondents in the central and eastern parts of Indonesia, 10.3% and 1.5%, respectively. With the spread of respondents throughout Indonesia, it is hoped that they can represent characteristics related to changes in body weight in students in Indonesia. One of the impacts of the Covid-19 pandemic on the world of education is changes in learning methods. The learning system throughout Indonesia has become distance learning with an online form that limits direct contact, as well as crowds in learning. Students from any region in Indonesia are expected to be able to access and obtain maximum education with freedom of knowledge (Abidah *et al*., 2020).

Viewed from the education level, most respondents have undergraduate/ professional education, namely 83%. However, some respondents are currently pursuing doctoral education, namely 0.9%. The education sector is almost the same as the health sector, 54.3%, and non-health, 45.7%.

This study has three variables: changes in body weight, consumption, and lifestyle factors. From the results of data analysis, it is known that most of the respondents experienced changes in body weight during the Covid-19 pandemic, namely 67.2% for students who live together and 66.8% for students who live alone. However, more than half of the respondents have a habit of weighing their body weight irregularly, whether they live together (57%) or alone (58.7%). During the pandemic, many changes in body weight occurred in the community, including among students. Changes in body weight during the pandemic due to increased consumption of snacks and diet can increase degenerative diseases (Lin *et al*., 2021).

Based on the results of the analysis of consumption factors (Table 2), for breakfast habits, some respondents who did not usually eat breakfast, namely 52%, experienced changes in eating patterns with almost the same proportions as those who were “not good” (47.4%) and “good” (52.6%). In contrast, the supplement consumption indicators did not differ. The proportion of changes in diet, where most of the respondents had taken supplements well during the pandemic, was 52.6%. Changes in consumption patterns during the Covid-19 pandemic for students consisted of increasing meal portions, frequency of cooking at home, frequency of snacks, and frequency of vegetables and fruit (Noviasty, Reny; Susanti, 2020).

The lifestyle factors of the respondents mainly experienced moderate stress, which was 77.1%, with bad sedentary behavior at 63.4%, good exercise habits at 63.7%, and prevalent sports such as bicycles. Activity restrictions during the Covid-19 pandemic can interfere with daily activities where there are several changes, including increased sitting, lying down, playing games, watching television, and using mobile phones (Chen *et al*., 2020). The pandemic causes an increase in non-communicable diseases due to decreased physical activity, increased stress, and high consumption of snacks (Ardella, 2020).

CONCLUSION

Most of the college students as the respondents are female in their late adolescents. They are currently studying to achieve their bachelor's degree/professional education in health education. The area where most of the respondents live is in the Indonesia Western Standard Time (WIB). Most respondents experienced changes in body weight (67.1%) during the Covid-19 pandemic, and half had a habit of weighing their weight irregularly (57.4%). Some respondents do not usually have breakfast and experience changes in eating patterns, with almost the same proportion between bad and good.

In contrast, the supplement consumption indicator does not differ from the balance of diet changes, where most respondents have taken supplements well during the pandemic. Most respondents experienced moderate stress, with bad sedentary behavior and good exercise habits. These results have shown a similar proportion whether the respondents live together with family and/or relatives or live alone in dormitory/boarding/apartments during their study.

The results showed that the COVID-19 pandemic influenced eating habits, lifestyle, and changes in body weight. The community, especially students, living alone or with family and/or relatives, should have sufficient knowledge and attitudes about nutrition to maintain unwanted weight changes during the pandemic due to unhealthy eating habits and healthy lifestyles. Institutions of education are essential to pay attention to this because changes in body weight in the long term can increase the risk of degenerative diseases

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Table 1. Characteristics of Respondents

Variables	**Live together**	**Live alone**	**Total n (%)**
Gender			
Male	214 (23.7)	129 (45.6)	343 (28.9)
Female	688 (76.3)	154 (54.4)	842 (71.1)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Age			
Late Adolescent	826 (91.6)	255 (90.1)	1081 (91.2)
Young Adults	53 (5.9)	26 (9.2)	79 (6.7)
Late Adult	19 (2.1)	2 (0.7)	21 (1.8)
Elderly	4 (0.4)	0 (0)	4 (0.3)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Current Level of Study			
Diploma	773 (85.7)	210 (74.2)	126 (10.6)
Bachelor/Professional	85 (9.4)	41 (14.5)	983 (83)
Master	38 (4.2)	27 (9.5)	65 (5.5)
Doctor	6 (0.7)	5 (1.8)	11 (0.9)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Field of education			
Health	516 (57.2)	127 (44.9)	643 (54.3)
Non-Health	386 (42.8)	156 (55.1)	185 (15.6)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Domicile			
WIB (Indonesia Western Standard Time)	817 (90.6)	228 (80.6)	1045 (88.2)
WITA (Indonesia Central Standard Time)	73 (8.1)	49 (17.3)	122 (10.3)
WIT (Indonesia Eastern Standard Time)	12 (1.3)	6 (2.1)	18 (1.5)
Total	902 (100,0)	283 (100,0)	1185 (100,0)

Table 2. Distribution of Research Variables

Variables	Live together n (%)	Live alone n (%)	Total n (%)
Body Weight Changes			
1. Body Weight			
Changing	606 (67,2)	189 (66,8)	795 (67,1)
Not changing	296 (32,8)	94 (33,2)	390 (32,9)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
2. Habit of Weighing			
Not Routine	514 (57,0)	166 (58,7)	680 (57,4)
Routine	388 (43,0)	117 (41,3)	505 (42,6)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Consumption			
1. Breakfast Habit			
Not Routine	468 (51,9)	148 (52,3)	616 (52,0)
Routine	434 (48,1)	135 (47,7)	569 (48,0)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
2. Dietary Changes			
Not Good	447 (49,6)	115 (40,6)	562 (47,4)
Good	455 (50,4)	168 (59,4)	623 (52,6)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
3. Supplements			
Not Good	408 (45,2)	124 (43,8)	532 (44,9)
Good	494 (54,8)	159 (56,2)	653 (55,1)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
Lifestyle			
1. Stress Level			
Severe	89 (9,9)	21 (7,4)	110 (9,3)
Moderate	684 (75,8)	230 (81,3)	914 (77,1)
Mild	129 (14,3)	32 (11,3)	161 (13,6)
Total	902 (100,0)	283 (100,0)	1185(100,0)
2. Sedentary lifestyle			
Not Good	574 (63,6)	177 (62,5)	751 (63,4)
Good	328 (36,4)	106 (37,5)	434 (36,6)
Total	902 (100,0)	283 (100,0)	1185 (100,0)
3. Exercise			
Not Good	335 (37,1)	95 (33,6)	430 (36,3)
Good	567 (62,9)	188 (66,4)	755 (63,7)
Total	902 (100,0)	283 (100,0)	1185 (100,0)

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Utilization of Edible-Insects as Protein Substitute in Food and Effects of Processing on Their Nutrient Contents and Protein Functionalities

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ABSTRACT

Population growth, poverty and climate change dictate the need for additional protein sources. Edible insects are potential protein substitutes and can provide both humans and animals with the required amount of protein, essential amino acids, and other indispensable nutrients. Incorporating edible-insects into familiar products and subjecting them to adequate processing that masks their natural appearance will improve their consumption. This article provides insights on the potentials of edible-insects as novel ingredients in food processing and its various benefits. Effects of processing on their nutritional and functional properties were also discussed. Literature was gathered through an online search on the Science Direct database and Google Scholar. Edible-insect powders, protein isolates and concentrates and oils were reported to be incorporated into bugger, chips, chocolate, bread, cookies, and other baked snacks. The addition of insect products improves protein, fat, fiber, and minerals contents. Insects also contain healthier lipids when compared with conventional proteins. Edible-insects will provide essential nutrients to the rapidly growing world population when more attention is given to their production, processing, safety, and marketing. Efforts need to be strengthened to secure global acceptance of insect protein since the conventional sources are not reliable and cannot satisfy the world population in decades to come.

Keywords: entomophagy, food processing, meat alternative, protein substitute, unconventional protein.

INTRODUCTION

Insects eating (entomophagy) was dated back to antiquity (Cartay *et al*., 2020; Liu and Zhao, 2019; Sun-Waterhouse *et al*., 2016) and the use of insects as food is as old as the history of mankind (Das *et al*., 2019). Insects can be processed into food that can provide essential nutrients (Cartay *et al*., 2020; Dube *et al*., 2013).

They are also fundamental in the pollination process (Cartay *et al*., 2020), production of honey, silk, natural colouring agents, (Testa *et al*., 2016), nutrients cycling and biological control (Dzerefos and

Witkowski, 2014), and improvement of soil fertility through waste bioconversion (van Huis *et al*., 2013).

There is an increasing demand for protein-rich foods, more attention is given to the novel sources of protein, researchers and marketers are putting more efforts into promoting edible-insects among new consumers to meet the obligatory need to diversify protein sources (Pippinato *et al*., 2020). The desire to reconsider discarded eating habits and food sources into modern recipes has fascinated the interest of researchers, chefs, and businessmen on edible-insect potentials as an alternative protein source (Testa *et al*., 2016).

Reports from many researchers shown that insect proteins can be used as a substitute for conventional protein in many products without adverse effects on their sensory qualities. Their inclusion in food has proven to improve the nutritional values of many processed foods (Imathiu, 2020).

Research in entomophagy covered many areas of study including food science, human and animal nutrition, health, entomology, ecology, agriculture, biochemistry, anthropology, sociology, and history (Yen, 2015).

The most studied insect in recent years were cricket, silkworm pupae, housefly (Testa *et al*., 2016), lepidopteran larvae, coleopteran larvae, different species of grasshoppers, locusts, and termites (Kelemu *et al*., 2015).

This article intended to provides insights on the potentials of edible-insects as novel ingredients in food processing and its various benefits. Effects of processing on their nutritional and functional properties were also discussed.

EDIBLE-INSECTS AS CONVENTIONAL PROTEINS SUBSTITUTE

Edible-insects and their products are used as ingredients in food processing, insect powders, protein isolates and concentrates, and oils were reported to be incorporated into burger, chips, chocolate, bread, cookies, and other baked snacks (Table 1).

The addition of insect products improved protein, fat, fibre, and minerals contents without affecting functional properties and sensory qualities. It also improved dough characteristics and baking properties (Table 1).

Gravel and Doyen (2020) reported that insect proteins are potential functional ingredients in foods and can play important roles in large scale food production. Food scientists are working hard on the possibilities of incorporating insects into some staple foods. At present, more attention is given to bakery goods. Substitution of wheat flour with 5 % insect flour improves the protein and fibre content of bread without affecting the dough properties and acceptability of the bread (González *et al*., 2019).

Insect flour at a 7.5 % level of substitution improves the nutritional content of corn flour extrudates snack mixture without affecting extrusion parameters (Igual *et al*., 2020). Extrusion cooking of soy concentrate with honeybee drone brood produced protein-rich snacks with 66 % protein content (Ulmer *et al*., 2020). Inclusion of 25 % crude black soldier fly larvae fat as a butter substitute in the production of cakes and cookies and 50 % in the production of waffles have no effects on the texture, colour, and overall liking of these products (Delicato *et al*., 2020).

Substitution of wheat flour with grasshopper powder at 100 g/kg grasshopper powder has no effects on the sensory qualities of the grasshopper-based bread, the substitution increased the protein content by 60 % and softness of the bread (Haber *et al*., 2019). Cricket powder at 15 % inclusion enhanced bread dough stability and reduced the degree of softening (Cappelli *et al*., 2020).

Leavened crunchy snacks produced with 30 % lesser mealworm powder exhibit low water activity (aw) and have pleasant sensory attributes and better mineral, protein, and amino acid contents than that produced from 100 % wheat flour (Roncolini *et al*., 2020). The low aw exhibited by lesser mealworm powder reveal the potential of using insect protein as a preservative in food.

Mulberry silkworm pupae and African palm weevil larvae can be used as a beef substitute in the production of filling for snacks and cuisines (Akande *et al*., 2020a). Skim milk can be replaced with locust and mulberry silkworm pupae powder in the production of high-energy biscuits, at 15 % inclusion, the nutritional value is above the minimum level prescribed by USAID (Akande *et al*., 2020b).

Another potential area of using insects is in improving the sensory properties of healthy foods with acceptance problems, silkworm powder was used by Biró *et al*. (2019) to mask the unpleasant sensory characteristic of buckwheat. Insects proteins are potential functional ingredients in food processing, the findings of Gravel and Doyen (Gravel and Doyen, 2020) revealed that insects protein concentrate and isolate are suitable ingredients with excellent functionality. The textural properties of insect proteins are amazing in 3D printing, their consumption can be improved through the production of foods with attractive, shapes, structure, and texture (Caporizzi *et al*., 2019).

PROCESSING OF EDIBLE INSECTS

Insects are consumed either raw, fried, or roasted (Das *et al*., 2019). The most common processing methods for insects are trimming, boiling, sun-drying, roasting, and pan-frying (Akullo *et al*., 2017; Ebenebe *et al*., 2017). In India, insect meals are prepared by boiling and roasting, sometimes with spices (Chakravorty, 2014), other processing methods are sand-roasting and osmotic drying using rock salt (Bose, 2020). Surplus catches during the season are preserved by sun-drying and stored for future use (Akullo *et al*., 2017), this ensures the continuous supply of insect protein year-round (Dube *et al*., 2013). The crude techniques used in the processing and preparation of insects improve their safety, shelf-life, nutritional value, and palatability (Akullo *et al*., 2017; Dube *et al*., 2013; Mézes and Erdélyi, 2020). Köhler *et al*. (2019) reported that insects purchased from the supermarket have better nutrient content than those gathered from the street, therefore, adopting modern techniques in insect processing will further improve their safety, commercial value, and versatility (Akullo *et al*., 2017). Edible insects were reported to contain many important nutrients but there is little information regarding the effects of different processing on their nutritional qualities, nutrients bioavailability, and toxicity (Ayensu *et al*., 2019).

Effects of Processing on the Qualities and Functionalities of Insect Products

The findings in Table 2 revealed that cooking affects nutrient contents and their bioavailability, digestibility, pH, and functional properties. Thermal processing of insects generally lower microbial counts and affects their distribution and activities during storage. The higher temperatures involved in cooking increase acidity due to the breaking down of glycogen into lactic acid. Higher temperature and low pH affect gelling and foaming properties, trigger coagulation, and increase surface hydrophobicity. Hydrolyses of both protein and lipid were observed during prolonged heating. Cooking has no effects on fatty acids composition and protein secondary structure, but heating with

NaCl decreases α-helix structure and surface hydrophobicity. Drying affects macronutrient contents, protein solubility, and oxidation (Table 2). Thermal processing of insects improved their safety, acceptability, digestibility, palatability (Garofalo *et al*., 2019; Megido *et al*., 2018; Murefu *et al*., 2019) nutritional quality, and bioavailability of sulfur-containing amino acids (Poelaert *et al*., 2018). Higher temperatures during cooking can have damaging effects on lipid and protein (Megido *et al*., 2018). Cooking methods can affect nutrients content and their bioavailability in insect, boiling, and vacuum cooking maintains good levels of polyunsaturated fatty acids and proteins in mealworms (Megido *et al*., 2018).

Reports in Table 2 show that extraction conditions can affect the properties of edible-insect protein extract. The solubility of the protein extract depends on the extraction pH. Increased protein content was recorded at pH of 10 in combination with Ultrasound treatment. Pressurised-liquid assisted extraction lower cholesterol level and increases PUFAs contents. Alkaline hydrolysis lower foam stability and emulsification properties of the protein extract. Exposure to ultrasound improved antioxidant activity, solubility, and foam expansion of the protein extract over a wide range of pH (2-12) (Table 2). Liceaga (2019) reported that the functional properties of insect flour can be improved by controlled enzymatic hydrolysis. Physical properties such as turbidity, dispersibility, and particle size of insect proteins can be modified by ultra-sonication. An increase in the sulfur hydride value, dispersibility, and zeta potential, and a decrease in turbidity and particle size in protein preparations and hydrolysates were observed in soldier fly larvae subjected to ultrasonic treatment (Mintah *et al*., 2020). Increased foaming and emulsifying abilities, and protein value were observed in grasshopper and honey bee brood protein extracts obtained using alkaline and sonication-assisted extraction (Mishyna *et al*., 2019a). Mishyna *et al*. (2019b) demonstrated the mechanism of gel aggregate formation by both covalent and non-covalent intermolecular interaction occurs during heat denaturation of freeze-dried honey bee brood protein, this unveils the potential of using insect proteins as a gelling agent. Fermentation is a promising technique for producing a wide range of edible insect products including extracts, sauces and paste (Castro-López *et al*., 2020).

The addition of insect proteins into wheat flour affects the dough and baking properties of baked foods. At 15 % incorporation, cricket flour increase dough tenacity and reduces extensibility, this led to poor volume in the baked product. The addition of insect flour also lowers expansion and crunchiness in extruded products. Defatting improved overall liking, digestibility, and lower hydrophobicity (Table 2)

The killing method of insects can affect the appearance and qualities of insect products. Killing generally increases the browning index and lowers radical scavenging. Killing in the absence of oxygen minimizes lipid oxidation and improves its nutritional qualities (Table 2). Killing methods affects the acceptability of insect by influencing saltiness and umami taste (Table 2). Leni *et al*. (2019) compare freezing (the most common killing method) with balancing and found out that the slow killing process involves in freezing gave room for many enzymic reactions including melanization reaction, killing by balancing inhibit browning, increase extractability and digestibility of protein and maintain essential amino acids. The killing method also affects the lipid composition of the insect products during storage, Caligiani *et al*. (2019) reported that killing by balancing provides more stable lipid while killing by freezing causes the formation of free fatty acids during storage probably due to the activation of lipases.

OTHER INDUSTRIAL APPLICATIONS

Scientists strongly believe that insects will play a vital role when the insect industry becomes well established and it will have positive impacts in many areas including industries, government, business, and research (Dossey *et al*., 2016) and its financial repercussions will impact the economy in general (Costa-Neto and Dunkel, 2016). In addition to the provision of food and feed insects can convert organic waste into important industrial raw materials. Insects have been foreseen as potential raw materials for many industrial processes (Tang *et al*., 2019) including food fortification during processing. Insects have higher anti-microbial peptides than any other animal, therefore, can be used as a novel source of antibiotic (van Huis, 2020a). Black soldier fly is a potential candidate for the production of antibiotics, textured protein, and bioplastic (van Huis, 2020b). Insect when used as feed have positive benefits on livestock health and welfare and their use as feed can reduce the use of antibiotics in livestock production (Dicke, 2018; Veldkamp and Eilenberg, 2018). Insect post-production waste is used as plant manure, and in an advanced way, can be used in the production of bio-methane gas using an anaerobic digestion process (Bulak *et al*., 2020).

RECOMMENDATIONS AND CONCLUSION

Authors recommends several points such as: at present, only few species are produced commercially, for effective promotion of entomophagy, the idea of insects gathering must be discarded, edible insects should be produced in large quantities at an industrial scale; People involve in traditional insect gathering should be transformed into insect farmers by providing them with the required skills and capital; Understanding the interaction between insect products and other food ingredients is critical in using insects as a novel ingredient in foods; Observing good personal hygiene throughout the production chain and the use of an appropriate HACCP system can improve the safety of edible insects; Processing into delicious, attractive, and irresistible meals with no visible insects, or their parts, and the use of an appropriate and eye-catching packaging system will promote edible-insects' acceptance.

Authors concluded that processing improves safety, stability, nutritional value, palatability, digestibility, nutrients bioavailability, acceptability, commercial value, functional properties, and versatility of edible-insects. The addition of processed insects into familiar foods improves their nutritional qualities and ingredients' functionality. Killing and processing methods affect protein functional properties, nutrient contents, and their bioavailability. Edible-insects and their products are used as ingredients in food processing. Insect powders, protein isolates, and concentrates, and oils were reported to be incorporated into burger, chips, chocolate, bread, cookies, and other baked snacks.

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Table 1. Insect Products as Potential Protein Substitute

Food products	Insect/Insect product	Incorporation level	Major findings	Reference
Cake and cookies	black soldier fly larvae fat	Addition of 25 % as butter substitute	Does not affect consumer preference	(Delicato *et al*., 2020)
waffles	black soldier fly larvae fat	Addition of 50 % as butter substitute	Does not affect consumer preference	(Delicato *et al*., 2020)
Gluten-free bread	house cricket (*Acheta domesticus* L.)	Addition of 5.4 %	1. Does not affect fermentation process 2. Enrich the bread with unique flavor 3. Improved antioxidant content of the bread	(Nissen *et al*., 2020)
Crunchy snacks	lesser mealworm (*Alphitobius diaperinus*) powder	Inclusion of 30 %	1. Protein content increased by 99.3 %, also essential amino acids 2. Increased Fe, P, and Zn, with Zn content increased by 300 % 3. Possessed pleasant sensory attributes and low a_w	(Roncolini *et al*., 2020)
High-energy biscuit	Silkworm pupae (*Bombyx mori*) and locusts	Addition of 15 % as a protein substitute	1. Improve in protein, fat and, fiber contents and energy value were observed 2. The biscuits meet USAID standard 3. No effects on the sensory qualities	(Akande *et al*., 2020b)
3D printed cereal-based snacks	Yellow mealworms (*Tenebrio molitor*) larvae	Inclusion of 20 % larvae flour as a novel protein source	1. Increase dough softness and flowing efficiency, these subsequently improved the geometry of the snack 2. Microstructure was improved due to better water evaporation during baking 3. Improvement in essential amino acids contents was observed	(Severini *et al*., 2018)
Burger	Mealworm	Addition of 50 % as a meat substitute	1. Taste and appearance scores were above neutral. 2. Ranked above vegetable bugger	(Megido *et al*., 2016)

Buckwheat pasta	Silkworm	Inclusion of 10 g/100g pasta	1. Improve the nutritional value of the pasta without affecting its acceptability 2. Received higher sensory scores than the control 3. Reduced cooking time	(Biró *et al*., 2019)
Cookies	House cricket (*Acheta domesticus L.*)	Addition of 10 % as a wheat flour substitute	1. Has no effects on the flavor, taste, or texture. 2. Was accepted by more than 70 % of the panelists	(Sogari *et al*., 2017)
Bread	cricket (*Acheta domesticus*) powder	Addition of 10 % as a wheat flour substitute	1. Does not affect the sensory qualities 2. Improves protein and essential amino acids contents	(Osimani *et al*., 2018)
Bread	cinereous cockroach (*Nauphoeta cinerea*)	Addition of 10 % as a wheat flour substitute	1. Has no effects on the functional and sensory properties 2. Protein content increased by 49.16 %	(de Oliveira *et al*., 2017)
Tortilla chips and chocolate bars	crickets (*Acheta domesticus*),	Tortilla chips containing 15% cricket flour and chocolate bar with figs and 5.5% cricket flour	1. Chocolate bar with insect flour possess better sensory qualities, followed by whole cricket. 2. Snacks with visible whole insects were scored low	(Cicatiello *et al*., 2020)
Bread	*Hermetia illucens*, *Acheta domestica* and *Tenebrio molitor*	Addition of 5 % as a wheat flour substitute	1. *Hermetia illucens* and *Acheta domestica* increased dough stability and reduced water absorption 2. Improved protein and fibre contents	(González *et al*., 2019)
Extruded corn snacks	Cricket flour (*Acheta domesticus*)	Enrichment with 12.5 and 15 %	Improved the protein content to EC recommendation n. 1924/2006	(Igual *et al*., 2020)

Table 2. Effects of Processing on Nutritional Qualities, Functionality and Acceptability of Insect Products

Insects	Origin	Processing methods	Effects	Reference
Beetle & cricket	Wild	Boiling	Reduced Fe and Zn bio-accessibility by 50 % and lower protein content and digestibility	(Manditsera *et al*., 2019)
mealworms	Laboratory reared	Vacuum cooking Frying Boiling Oven cooking	Lower protein, lipid, and ash contents Lower protein and ash contents and increases lipid content Increases protein and ash contents and lower lipid content Increases protein and ash contents and lower lipid content	(Megido *et al*., 2018)
Cricket flour (*Acheta domesticus*)	Farmed	Addition of 15 % as a wheat substitute in bread	Increases dough tenacity (P) and curve configuration ratio (P/L), and reduces dough extensibility (L). these subsequently lead to a fall in volume	(Cappelli *et al*., 2020)
Cricket flour (*Acheta domesticus*)	Farmed	Addition of 15 % in extruded corn snacks	1. Lead to low crunchy and low expansion extrudates 2. Modified water solubility index and improve stability 3. 7.5 % inclusion was recommended for better extrudates parameters	(Igual *et al*., 2020)
Acheta domesticus and *Tenebrio molitor*	Farmed	ultrasound-assisted extraction and pressurized-liquid extraction	Causes a decrease in saturated fatty acids and monounsaturated fatty acids contents and increase PUFAs contents, also lower cholesterol level	(Otero *et al*., 2020)
Cricket-based snacks	Farmed	Effect of degree of processing and incorporation on the acceptability	Degree of processing affects acceptability, insect-based products should contain processed insects, not whole insects	(Gmuer *et al*., 2016)
Protaetia brevitarsis larvae	Wild	Defatting using hexane	1. decrease in hydrophobicity and tryptophan fluorescence intensity was observed 2. digestibility is higher than that of beef	(Lee *et al*., 2021)
Cricket	Farmed	Broth making by cooking in pouches at 85 °C for 1 h in a water bath	Lower pH due to the breakdown of glycogen and formation of lactic acid	(Farina, 2017)

Hermetia illucens larvae	Laboratory reared	Treatment of isolates and hydrolysates with fixed-frequency ultrasonic and sweep-frequency	Alkaline hydrolysis improved protein solubility, but lower forming stability and emulsifying properties. Ultrasound treatment improves antioxidant strength, solubility, and foam expansion properties over a wide pH range (2-12)	(Mintah *et al*., 2019)
Honey bee brood (*Apis mellifera*)		Effects of pH and temperature on aggregation and gelation	Highest coagulation achieved at 85 °C and pH 5-7 The least gelation concentrations vary between 5 to 11 % at pH of 7 and 3 respectively	(Mishyna *et al*., 2019c)
House cricket (*Acheta domesticus*)	Farmed	Effects of different killing method: blanching, steaming, freezing, carbon dioxide, vacuum, plastic bag, and carbon dioxide + blanching	1. all the killing methods were found to increase the browning index and reduce radical scavenging activity. 2. better proximate values were obtained by carbon dioxide, vacuum, and plastic bag killing 3. killing by freezing, carbon dioxide, vacuum, and plastic bag provide healthier lipids and minimize lipid oxidation	(Singh *et al*., 2020)
Mealworms (*Tenebrio molitor*)	Laboratory reared	Comparing the effects of different drying methods (fluidized bed drying, microwave drying, freeze-drying, vacuum drying, and rack drying)	1. all the drying methods affect the macronutrient contents 2. protein solubility is higher in freeze-drying and lower in microwave drying, fluidized bed drying, and vacuum drying 3. oxidative status is higher in freeze-dried samples	(Kröncke *et al*., 2018)
Yellow mealworm (*Tenebrio molitor*) and lesser mealworm (*Alphitobius diaperinus*) larvae	Farmed	Production of a minced meat-like product by addition of salt, binding agent, and spices to steamed/fried and pulverized larvae before frying	Processing methods lower microbial counts and affect their distribution in the finished products and during storage under different conditions	(Stoops *et al*., 2017)

Black cricket (*Gryllus assimilis*)	Farmed	Effects of thermal-aggregation on foaming and gelling properties of protein isolate	1. Heat treatment alone doesn't change the protein secondary structure but increase the surface hydrophobicity 2. critical gelling concentration achieved at 90 °C for 15 min exposure 3. the foamability at the treatment; 75 and 95 °C for 15 min, with or without NaCl is similar to that of whey protein isolate 4. heating with NaCl induced decrease in α-helix structure content and surface hydrophobicity	(Santiago *et al*., 2021)
Yellow mealworm (*Tenebrio molitor* L.)	Laboratory reared	Effects of freezing (−20 °C) and drying processes (90 °C for 1.5 h; 50 °C for 62 h) on molecular attributes	1. The treatments have no effects on fatty acid profiles 2. Hydrolysis of triacylglycerols was favoured by the low-temperature long time treatment 3. Low-temperature long time treatment also lead to protein hydrolysis which leads to an increase in free amino acid contents	(Melis *et al*., 2018)
Tenebrio molitor larvae	Farmed	Protein extraction using water at alkaline pH	1. At pH 10 and 45 °C protein content was increased by two folds 2. The solubility of the extracted protein depends greatly on pH	(Azagoh *et al*., 2016)
Crickets *(Acheta domesticus and Gryllodes sigillatus)*	Farmed	Effects of defatting and freeze-drying on nutritional value and sensory qualities of cereals bars enriched with insects	Defatting improved the overall liking and willingness to consume the products	(Ribeiro *et al*., 2019)
Crickets	Farmed	Effect of killing methods (freezing and heating) on the sensory and physiological properties of insect broth	There is significant difference between the samples on overall liking, saltiness, and umami scores	(Farina, 2017)

Red Guava (*Psidium guajava* L.) Chemical Properties Characterization Using Different Packaging Methods During Storage

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ABSTRACT

The limited shelf life of red guava fruit encourages efforts to maintain its shelf life by using the sealing and wrapping packaging method with storage at 9ºC for 12 days. The purpose of this study was to determine the effect of packaging method and storage time on the chemical properties of red guava (*Psidium guajava* L.). This study used a factorial design consisting of 2 factors. The first factor is the packaging method (without packaging, sealing and wrapping). The second factor is storage time (day 0, day 3, day 6, day 9 and day 12). The results showed that the highest vitamin C content was in the sealing packaging method, namely on the 3rd day of storage at 3.3%. the highest water content value is 90.45% on the packaging method of wrapping storage on day 0, the highest value of total titrated acid in fruit with packaging method of packaging storage day 0 is 0.08%, while the highest value of total dissolved solids was 7.33ºBrix on the 12th day storage sealing packaging method.

Keywords: chemical properties, red guava, storage time, and packaging method

INTRODUCTION

Indonesia is a country that produces abundant horticultural commodities. One of the most popular and abundant horticultural products in the community is guava fruit. Guava is a fruit that is easy to find and is a fruit that has a very high vitamin C content. Guava fruit has a vitamin C content of 87 mg/100 grams (Padang & Maliku, 2017) which is higher than tomatoes, which is 21 mg/100 grams (Sari *et al*., 2021). Guava is one of the horticultural products that is easily damaged so that without proper and proper treatment, guava will have a short shelf life and are easily damaged. The shelf life of guava stored at room temperature is only able to last a few days while at peak CO_2 and ethylene production the shelf life is only 3-6 days after harvest (S. E Widodo *et al*., 2012). The damage that occurs is usually caused by metabolic processes such as respiration and transpiration. The ongoing metabolic process will cause changes in food products. In addition to metabolic processes that damage commodities, there is physical damage due to various treatments carried out (Murtius & Hari, 2019).

One of the packaging that can maintain the quality of the product is plastic LDPE. The use of plastic LDPE is very effective and has advantages compared to other types of packaging. The advantages of plastic are that it is strong, transparent and has permeability to oxygen, water vapor and CO_2 (Asridaya, 2019). Packaging methods that can be used to package a horticultural product are sealing and wrapping. Treatment using this method can inhibit the decline in product quality and prolong the shelf life of the product (Hamdani *et al*., 2017). This limited shelf life encourages efforts to maintain the shelf life of red guava fruit by using sealing and wrapping packaging methods. The use of the sealing packaging method has the working principle of gluing plastic so as to minimize the possibility of air or water entering the material, so as to maintain product quality. While the use of wrapping packaging method is a form of modified atmosphere storage which has a principle, namely CO_2 in the packaging will be higher than O_2 so that respiration in fruit becomes low. The use of sealing and wrapping packaging methods is expected to be able to inhibit the high rate of transpiration and respiration. In addition, storage at a temperature of 9ºC is an effort to slow down the metabolic process of the fruit (Murtiwulandari *et al*., 2020). The purpose of this study was to determine the effect of packaging method and storage time on the chemical properties of red guava *(Psidium guajava* L.*)*

MATERIALS AND METHODS

Tools and Materials

The main ingredients needed in the research are red guava, a red brittle variety that have met the harvest requirements, namely the skin that is dark green turns light and yellowish (maturity rate 50% - 60%) (Mulato, 2015) obtained from the Juwana market, Pati Regency, Central Java, and LDPE plastic size of 30 cm x 15 cm with a thickness of 30 microns. While the analytical materials used in the study were 0.1 N NaOH, 1% phenolphthalein, aquades, amylum solution, 0.1 N iodine.

The main tool used in this research is a vacuum sealer machine brand Kris. While the tools used for analysis in the study were Erlenmeyer brand iwaki, funnel brand iwaki, measuring cup brand iwaki, dropper, beaker brand iwaki, burette brand iwaki, vacuum pump, oven brand mummert, iron tongs, aluminum cup, rag, storage rack, refrigerator brand LG-205L gross, blender brand cosmos CB-190, knife, scissors, spoon. , isolation.

Research Methods

The research method used is a factorial design consisting of 2 factors, namely the packaging method and storage time. The packaging for red guava fruit is without packaging (control), sealing, and wrapping with a storage time of 12 days stored at a refrigerator temperature (9ºC). While the analysis carried out in this study was the determination of vitamin C content, water content test, determination of total dissolved solids and total titrated acid.

Research Implementation

The material that has been obtained is sorted to get relatively homogeneous fruit and the same level of maturity with the condition that the fruit skin is light yellowish green (maturity level 50% - 60%). After that, the samples were washed to remove the adhering dirt and drained to dry. Then the guava fruit was weighed ± 125 grams and packed according to the packaging method (control, sealing and wrapping) and then stored in a refrigerator at a temperature of 9ºC for 12 days. Data collection on parameters is carried out with the following analysis:

1. Determination of Vitamin C Levels using the iodimetric method (Rahmawati & Hana, 2016).
2. Determination of Water Content (AOAC, 2005)
3. Determination of Total Dissolved Solids (Bayu *et al*., 2017)

4. Determination of Total Titrated Acid (Megama, 2016)

RESULTS AND DISCUSSION

Vitamin C levels

One of the vitamins found in fruits that is needed by the body is Vitamin C. Vitamin C has a role as an effective antioxidant in warding off free radicals that can damage tissues and cells (Putri & Setiawati, 2015). One fruit that contains vitamin C is red guava fruit. Vitamin C levels in red guava fruit can be seen in Figure 1.

Based on Figure 1, the content of vitamin C in red guava fruit during 12 days of storage decreased. The highest vitamin C content in guava fruit was on the sealing packaging method on the 3rd day of storage, namely 3.33% on the sealing packaging method and the lowest vitamin C content on the 12th day storage control packaging method was 2.91%.

Figure 1 shows the vitamin C content, the longer the storage time the less vitamin C content. The content of vitamin C at a temperature of 9ºC will experience a decrease in vitamin C faster, this can cause damage to the fruit cell structure by the freezing process (Dewi *et al*., 2007).

The results of the analysis of the vitamin C content in Figure 1 show that the storage on the 12th day had the lowest vitamin C content compared to the vitamin C content on the previous day's storage. This is influenced by the storage temperature and the length of storage time, besides that the activity of enzymes that play a role in the process of overhauling vitamin C is still ongoing during storage (Naibaho, 2014).

The decrease in vitamin C content during storage can be caused by the nature of vitamin C which is easily soluble in water and easily oxidized by air or exposed to heat (Putri & Setiawati, 2015). In addition, packaging with LDPE plastic is able to inhibit the oxidation rate of red guava fruit so that it will reduce the loss of vitamin C loss (Anggraini & Permatasari, 2017). So that the decrease in vitamin C content in red guava fruit is not significant.

Water content

Changes in water content in a product is one of the determining factors for the quality of the product. Moisture content is one of the physical properties of the material which indicates the amount of water contained in the material. The water content of red guava fruit can be seen in Figure 2.

Based on the results of the analysis of the water content of the guava fruit, it can be seen in Figure 2 that the highest water content of all treatments is in the packaging method of wrapping storage on day 0 (90.45%) and the lowest water content in the storage control packaging method on day 6 (83.40%). The occurrence of a decrease in the water content of guava fruit during storage is a frequent occurrence, this is caused by the loss of water content in the fruit after the picking process until the fruit ripens. Water loss in fruit is also caused by increased respiration rate during fruit ripening, low humidity and too high storage temperature. An increase in the rate of respiration and transpiration in fruit can occur if the storage temperature is too high in the fruit, so that it can trigger easy water loss in the fruit (Yulianti *et al*., 2016).

The water content of guava fruit in the sealing and wrapping packaging method seen in Figure 2 is more able to maintain the water content of the fruit when compared to the treatment without packaging (control), this is because both sealing and wrapping

packaging methods use LDPE plastic. Low permeability is able to suppress the rate of exit and entry of water vapor into the material. Low permeability can increase the moisture in the packaging so as to reduce the temperature in the packaging which will suppress the process of water loss in the material (Johansyah *et al*., 2014).

The existence of a hydrolysis process during ripening that requires a lot of water can cause an increase in water content so that the free water contained in the fruit tissue will come out along with enzyme activity (Djanis & Hanafi, 2009). Previous research on mustard greens packaged using LDPE plastic showed a decrease in water content on day 6. This is due to the transpiration process which can cause the water content to come out of the vegetables (Anggraini & Permatasari, 2017).

Total Dissolved Solids

The fruit maturity indicator is one of the quality parameters of the fruit. The characteristics of an indicator of fruit maturity are fruit hardness, water content and total dissolved solids. Total soluble solids include pectin, reducing sugars, non-reducing sugars, proteins and organic acids. The interpretation of sweetness can be seen from the total dissolved solids because the sugar contained in fruits is classified as a lot (Kusumiyati *et al*., 2019). The total dissolved solids content in red guava fruit can be seen in Figure 3.

Based on the results of the analysis in Figure 3 shows the total dissolved solids ranged from 6.00ºBrix to 10.00ºBrix. During storage, the total soluble solids in red guava fruit increased with increasing storage time.

The highest value of total dissolved solids was in the sample without packaging (control) on the 12th day of storage, which was 10.00ºBrix and the lowest total dissolved solids in the sample with sealing and wrapping packaging methods was 6.00ºBrix. This shows that the value of the total dissolved solids can accelerate the ripening of a fruit. Storage of fruit at low temperatures can reduce the rate of fruit ripeness. The delay in ripening time of fruit can extend its shelf life (Dhyan *et al*., 2014). The increase in sugar in fruit is caused by the hydrolysis of starch into glucose, fructose and sucrose compounds and the speed of this hydrolysis is greater than the speed of converting glucose into energy and water so that sugar accumulates in the tissues during storage (Naibaho, 2014). Total soluble solids and total sugar contained in guava fruit increased during ripening along with a decrease in fruit hardness. According to Yulianti *et al*. (2016), the starch content in guava fruit decreased significantly during the transition from mature green to overripe maturity. The use of wrapping packaging methods with LDPE plastic types is able to inhibit the rate of respiration so that it can inhibit the increase in total dissolved solids (Soesiladi E. Widodo *et al*., 2016).

Total Titrated Acid

The acid content in the fruit usually indicates the total titrated acid. The fruit will become sweet due to the ripening process which will undergo an overhaul of organic acids (Noorbaiti *et al*., 2012). The total titrated acid in red guava fruit can be seen in Figure 4.

Based on the results of the analysis of the total titrated acid shown in Figure 4, it decreased during the storage of red guava fruit for 12 days. The highest total dissolved acid value was 0.08% on the 3rd day storage sealing packaging method, the 6th and 9th day storage without packaging method, and wrapping packaging method on the 0th day of storage. While the lowest total value of titrated acid was 0.04% on the 12th day storage wrapping packaging method. Acid content decreased due to the conversion of acid to form sugar after ripening, while the increase in acid content in fruit was caused by changes in polysaccharides (pectin, starch and hemicellulose) into simple soluble sugars (Julianti, 2012).

The decrease in organic acids contained in fruit during storage is used in the respiration process. These organic acids are used as a source of fruit energy, so that the acidity value of the fruit can decrease (Umah, 2018). Ripe fruit will increase in sugar content and decrease in acid. An increase in sugar content and a decrease in acid content usually occurs in climacteric fruit. Guava fruit is a climacteric fruit so it will experience a decrease in acidity (Noorbaiti *et al*., 2012). The acidity of the fruit will decrease after the maximum acid increase occurs. The total titrated acid in red guava fruit decreased after the 12th day of storage. This is used as an indicator that the fruit enters the senescence phase after the peak of maturity.

CONCLUSION

The effect of packaging method and storage time of red guava fruit on chemical analysis was that the highest vitamin C content was the sealing method on the 3rd day of storage (3.33%) while the highest value in the control was 3.02 on day 0. The highest moisture content was in the packaging method of storage wrapping on day 0 (90.45%) while the control was 87,35%. The highest value of total dissolved solids was 7.33ºBrix on the 12th day of sealing method, while the control was 10.00ºBrix on the 12th day. The highest value of total titrated acid was in the packaging method of storage wrapping on the 0th day (0.08%) while for the control it was 0.08% on the 6th and 9th days.

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Table 1. Research Design

Packing Methods (P)	Storage Time (L)				
	0 Day (L_1)	3 Days (L_2)	6 Days (L_3)	9 Days (L_4)	12 Days (L_5)
Control (P_1)	P_1L_1	P_1L_2	P_1L_3	P_1L_4	P_1L_5
Sealing (P_2)	P_2L_1	P_2L_2	P_2L_3	P_2L_4	P_2L_5
Wrapping(P_3)	P_3L_1	P_3L_2	P_3L_3	P_3L_4	P_3L_5

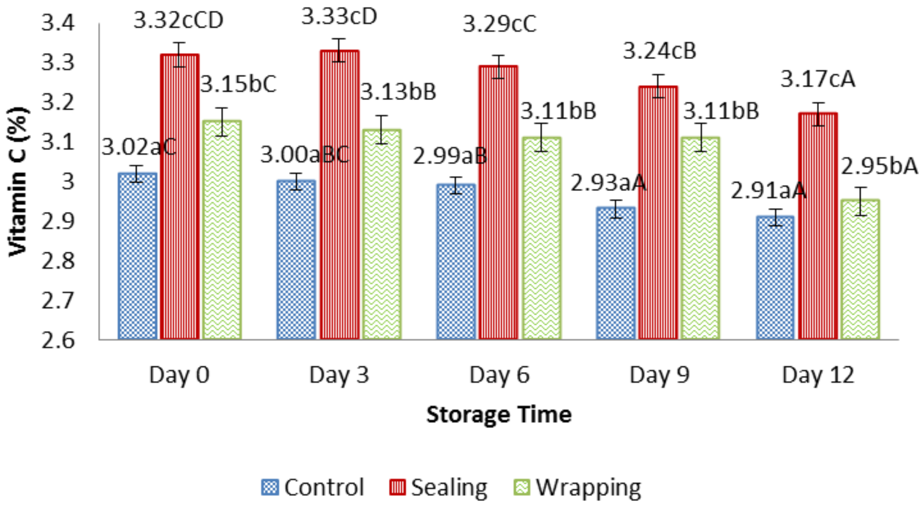

Figure 1. Vitamin C (%) of Red Guava

Information: The same notation on the same day (indicated in lowercase notation) showed no significant difference at = 0.05. The same notation on the same packaging method (shown in uppercase notation) showed no significant difference at = 0.05

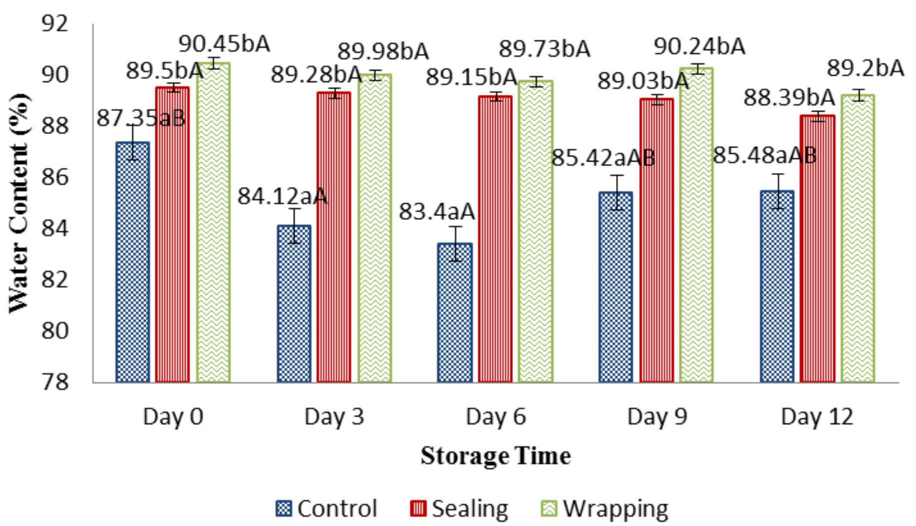

Figure 2. Water Content (%) of Red Guava

Information: The same notation on the same day (indicated in lowercase notation) showed no significant difference at = 0.05.The same notation on the same packaging method (shown in uppercase notation) showed no significant difference at = 0.05

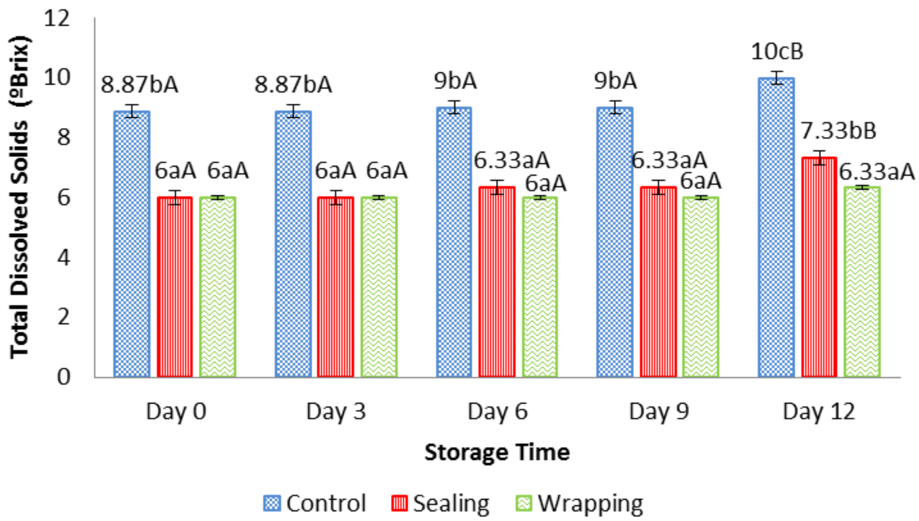

Figure 3. Total Dissolved Solids (ºBrix) of Red Guava

Information: The same notation on the same day (indicated in lowercase notation) showed no significant difference at = 0.05. The same notation on the same packaging method (shown in uppercase notation) showed no significant difference at = 0.05

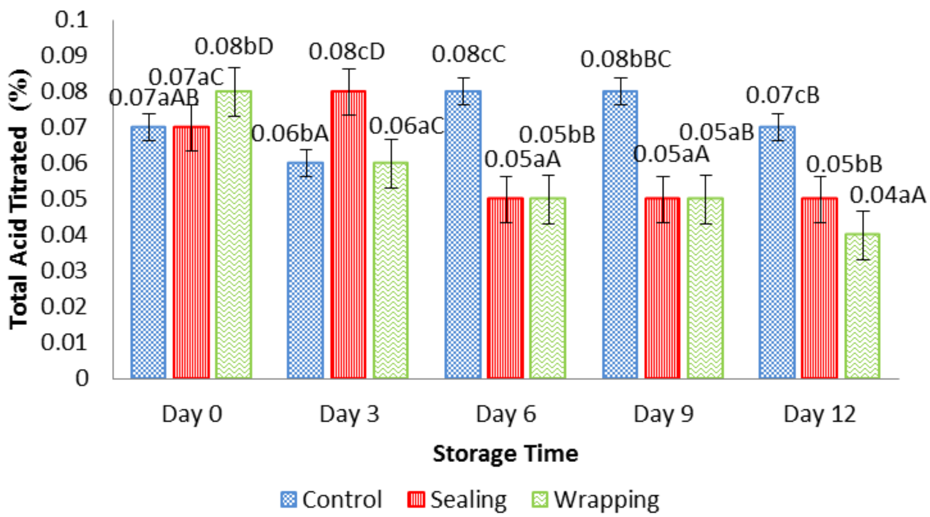

Figure 4. Total Titrated Acid (%) of Red Guava

Information: The same notation on the same day (indicated in lowercase notation) showed no significant difference at = 0.05. The same notation on the same packaging method (shown in uppercase notation) showed no significant difference at = 0.05

Effect of Sago Starch Concentration on Characteristic of Sago Glucose Syrup

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ABSTRACT

Opportunity to use sago as a basic ingredient for syrup glucose is very large because of the high carbohydrate content reached 75.88% - 85.08%. Sago starch contains 27% amylose and 73% amylopectin. This study aims to determine the effect of sago starch substrate concentration on reducing sugar, total dissolved solids, dextrose equivalent, and sweetness level from the glucose syrup produced. This research was conducted in 3 stages, namely gelatinization, liquefaction, and saccharification. The use of sago substrate concentrations were 25%, 30%, and 35%. The use of amylase enzyme is 0.1% dry weight and glucoamylase enzyme is 0.008 g/g dry weight. This study used a completely randomized design with a factorial pattern with two replications and data analysis using Duncan test. The use of α-amylase and glucoamylase enzymes in the manufacture of glucose syrup from sago starch affects the glucose syrup produced. The best result is obtained from 30% substrate concentration with reducing sugar value is 186.07 g/L, total dissolved solid is 36.13%, dextrose equivalent value is 62.02%, and sweetness level value is 33.92 ºbrix.
Keywords: glucose syrup, liquefaction, saccharification, sago

INTRODUCTION

The need of liquid sugar for industries tends to increase every year. Food, beverages, and pharmaceutical industries currently have a tendency to use glucose syrup (Rika et al., 2020). Glucose syrup is a clear and thick liquid containing D-glucose, maltose, and D-glucose polymer obtained from the hydrolysis of starches, such as tapioca, sago, corn starch, and tuber starch (Suripto et al., 2013). The food and beverage industry are starting to use glucose syrup a lot because has several advantages, including not crystallizing, being easier to process because it is more dissolved, more practical, and has a more attractive appearance when compared to granulated sugar in general (Permanasari & Yulistiani, 2017). In addition, glucose syrup has several advantages when applied to food products such as glucose ice cream products can suppress the freezing point and increase the smoothness of texture, in cake products it can keep cakes fresh and not easy to crack. Whereas in candy products, glucose can prevent microbiological damage and improve texture (Suripto et al., 2013).

The manufacture of glucose syrup consists of two hydrolysis methods, namely the enzymatic and non-enzymatic or its combination (Betiku et al., 2013). Enzymatic hydrolysis has fundamental differences with acid hydrolysis. Hydrolysis of starch is a breaking process of starch molecule to become constituent parts of the starch like dextrin, isomaltose, maltose, and glucose (Terahara et al., 2004). The enzymatic hydrolysis process is more effective than acid hydrolysis because the enzyme breaks the glycosidic bond specifically, leaving no residue and minimum color damage (Azmi et al., 2017). Glucose can be made from enzymatic hydrolysis by α-amylase and glucoamylase (Permanasari & Yulistiani, 2017).

The manufacture of glucose syrup by enzymatic hydrolysis consists of two stages, namely the liquefaction stage using the α-amylase enzyme and the saccharification stage using a mixture of glucoamylase and pullulanase enzymes. The α-amylase enzyme will cut the α-1,4-glycosidic bonds on the inside of the starch (amylose and amylopectin chains), while the glucoamylase and pullulanase enzymes will break the α-1,6 glycosidic bonds in the amylopectin polymer which is not able to be done by the α-amylase at liquefaction stage (Mardawati et al., 2019).

Sago stalks can be processed into sago starch. However, the use of natural starch (native) directly causes several problems, namely retrogradation, syneresis, low stability, and low resistance of pasta to pH and temperature. Therefore, it is necessary to modify starch physically, chemically, and enzymatically. The use of starch as a pharmaceutical product and fermentation medium is carried out through bioconversion, one of which is hydrolysis. Opportunity to use sago as a basic ingredient for syrup glucose is very large because the starch content is between 72% - 94% (Azmi et al., 2017). Sago starch contains 27% amylose and 73% amylopectin (Soraya et al., 2019). The purpose of this research is to determine the effect of sago concentration on the glucose syrup produced using α-amylase and glucoamylase enzymes.

MATERIALS AND METHODS

Tools and Materials

Sago flour obtained from North Luwu regency, South Sulawesi province, Indonesia. The other materials such as distilled water, α-amylase enzyme, glucoamylase enzyme, standard glucose DNSA (3,5-dinitrosalicylic acid) natrium sodium tartrate ($KNaC_4H_4O_6$-$4H_2O$), and NaOH were available from Hasanuddin University.

The tools in this research are spectrophotometer, oven, hotplate, autoclave, thermometer, pH meter, analytic measurer, desiccator, refrigerator, vortex, stopwatch, stirring rod, measuring glass, Erlenmeyer, reaction tube, micro pipette, and magnetic stirrer.

Methods

Raw Material Preparation

Sago flour dried by using blower then tested the water content. Sago concentration used in this research were 25%, 30%, and 35% w/v.

Gelatinization Process

Sago flour made in suspension by added distilled water. CaCl2 cofactor added, and the pH of suspension adjusted to 6.0-6.5 by added acid or base solution. The suspension heated until 121^0C and added α-amylase enzyme (0.1% dw). When the gelatinization process reached, the temperature maintained for 15 minutes (Megavitry, 2019).

Liquefaction Process

Suspension temperature lowered until 80^0C and the suspension added by the α-amylase enzyme (0.1% dw). The stirring process carried out for 90 minutes. At this stage, the result obtained was maltodextrin (Megavitry, 2019).

Saccharification Process

After 90 minutes of the liquefaction process, the suspension temperature lowered until 60^0C and pH set to 4.5 for the saccharification stage. Glucoamylase enzyme added into the suspension and stirred for 5 minutes. The suspension inserted into the Erlenmeyer 250 ml to incubated in a water bath shaker for 72 hours. Sampling was done in every 6 hours (Megavitry, 2019) (Figure 1).

Analysis

Reducing Sugar Analysis

The DNS method is a colorimetric technique that consists of a redox reaction between the 3,5-dinitro salicylic acid and the reducing sugars present in the sample. The reagent is a solution formed by the following compounds: 3,5-Dinitrosalicylic acid (2-hydroxy-3,5-dinitrobenzoic acid), which acts as an oxidant, Rochelle salt (sodium potassium tartrate), which prevents the dissolution of oxygen in the reagent and sodium hydroxide to provide the medium required for the redox reaction to occur (Garriga et al., 2017).

1 ml of clarified sample and 3 ml of DNS was pipetted into a test tube. The solution heated in boiling water for 5 minutes and immediately cooled in running water. Analysis of reducing sugar content was carried out using a spectrophotometer at wavelength of 550 nm.

Total Dissolved Solid Analysis

Determination of total dissolved solid was calculated using the water content method (Andarwulan et al., 2011). Around 5 g of sample dried using oven with temperature 105^0C for 6 hours. The weight was considered constant if the difference in weighing did not exceed 0.5 mg.

Dextrose Equivalent (DE) Analysis

According to (Yunianta et al., 2015), the value of DE can be determined by using the formula:

$$\mathrm{DE} = \frac{\sum \text{reducing sugar } (\frac{w}{v})}{\sum \text{total dissolved solid } (\frac{w}{v})} \text{ x } 100\%$$

Sweetness Level Analysis

The level of sweetness was tested using a hand refractometer (Apriyantono et al., 1989).

Data Analysis

Data processing was carried out using Completely Randomized Design (CRD) method with 2 replications. If the results are significantly different, Duncan's real distance difference test will be carried out as a further test.

RESULTS AND DISCUSSION

Reducing Sugar Level

Reducing sugars are sugars that have the ability to reduce electron-accepting compounds, this is due to the presence of free aldehyde and ketone groups. Glucose is a type of reducing sugar.

The results of the reducing sugars analysis showed that the use of 25% sago concentration resulted in a reducing sugar value of 143.93 g/L experienced a significant increase to the use of 30% sago concentration resulted in a reducing sugar value of 186.07 g/L, then the use of 35% sago concentration the reducing sugar value increased significantly of 211.94 g/L (Table 1).

Hydrolysis of starch by α-amylase enzyme produces glucose, maltose, maltotriose, and various types of α-limit dextrin, namely oligosaccharides consisting of 4 or more sugar residues containing many α-1,6 glycosidic bonds. Then the starch chain pieces that have been hydrolyzed by α-amylase enzyme will be further hydrolyzed into glucose by the glucoamylase enzyme, so that more glucose is produced. Each hydrolyzed sugar chain has one reducing sugar group so that the more starch hydrolyzed into simple chain sugars, the higher the amount of reducing sugar.

Increasing substrate concentration increases the rate of reaction. This is because more substrate molecules will be colliding with enzyme molecules, so more product will be formed. However, after a certain concentration, any increase will have no effect on the rate of reaction, since substrate concentration will no longer be the limiting factor. The enzymes will effectively become saturated, and will be working at their maximum possible rate (Istia'nah et al., 2020). The high concentration of sago starch allows the bond between the sago starch as substrate and enzymes to increase so that the resulting product in the form of simple chain sugars is also higher.

Increasing the substrate concentration can increase the enzyme reaction. The rate of reaction (V) catalyzed by the enzyme increases with increasing substrate concentration [S], until a state is reached where the addition of substrate concentration [S] no longer increases the initial rate of reaction and when all enzymes are saturated by substrate [ES], the reaction rate will reach maximum state (Mardawati et al., 2019). High concentration of the enzyme will affect the speed of the reaction. Furthermore, the substrate concentration is low, the rate of enzyme action is low. On the other hand, if the substrate concentration is high, the enzyme work will be fast and if the substrate is in excess, the enzyme will not decrease but remain constant (Budiyanto et al., 2019).

Several studies have been conducted and show that increasing the substrate concentration can increase the yield of reducing sugars, such as the study conducted by Ticoalu et al., (2016), regarding the utilization of purple sweet potatoes into anthocyanin drinks through enzyme hydrolysis, it was stated that the higher the concentration of purple sweet potatoes used, the higher the reducing sugar produced because the starch substrate that could be hydrolyzed by α-amylase and glucoamylase enzymes was also getting bigger.

Total Dissolved Solid Analysis

Total dissolved solid is the amount of solid contained in a material containing water. The results of total dissolved solids will be used to calculate the dextrose equivalent (DE) value of glucose syrup produced together with reducing sugar.

The results of total solid analysis showed that the use of 25% sago concentration resulted in total solids value of 31.09% and increased significantly to the use of 30% sago concentration with total solids value of 36.13%, then the use of 35% sago yielded total solids value of 38.83% (Table 2).

The significant increase in total solids with increasing concentration of sago was caused by the increase in the amount of starch in the suspension. Starch is composed of amylose and amylopectin, therefore the increasing concentration of substrate used means that the quantity of amylose and amylopectin also increases so that more water is bound to the substrate and causes an increase in total solids although at 35% sago starch concentration the increase in total dissolved solids was not very significant. The increase in total dissolved solids was caused by the breaking of long chains of carbohydrate compounds into soluble sugar compounds. The increase in total dissolved solids which is in line with the increase in temperature and cooking time is due to the higher the temperature causing the breaking of long chains of carbohydrate compounds into soluble sugar compounds to be faster, so that the sugar content in the suspension will dissolve more (Meikapasa & Seventilofa, 2016). Basically, the total dissolved solids of a material include reducing sugars, non-reducing sugars, organic acids, pectin, salts, and proteins which greatly affect the brix (Megavitry et al., 2019). The increase in reducing sugar resulted in the total soluble solids value of sago glucose syrup to increase.

Several studies have been conducted and show that increasing the substrate concentration can increase the total solids gain, such as the study conducted by Yunianta et al., (2015) regarding the production of glucose syrup made from canna starch, that the addition of canna starch substrate concentration causes a decrease in the water content in glucose syrup so that the total solids produced as a result of the saccharification process increase due to the binding of water by canna starch substrate. Similar results were obtained from a study conducted by (Mardawati et al., 2019), that the production of glucose syrup from corn starch, that the dissolved solids content of glucose syrup tends to increase with increasing substrate concentration.

Dextrose Equivalent Analysis

DE shows the amount of starch polymer that has been cut into simple sugar molecules, namely glucose, maltose, and dextrin. The DE value is the main parameter that describes how much starch is converted to glucose due to enzyme hydrolysis (Ni'maturohmah & Yunianta, 2015). Commercially the use of starch is influenced by the value of DE. The higher the DE solution, the higher the glucose level and the lower the dextrin level.

The results of dextrose equivalent analysis showed that the use of 25% sago concentration resulted in dextrose equivalent value of 57.57% and increased significantly to the use of 30% sago concentration with dextrose equivalent value of 62.02%. However, at 35% sago concentration, the dextrose equivalent value decreased to 60.55% (Table 3).

The higher the concentration of sago used the value of dextrose equivalent tends to decrease, because at higher starch concentration the time required for the enzyme to convert starch into dextrin takes longer so that increasing the concentration at the same time causes a decrease in product DE, while in this research the time used for each concentration was the same, namely 72 hours. Dextrose equivalent is a measure of the percentage of glycosidic bonds in starch that has been hydrolyzed, referring to the reducing sugar content, an indication of the large number of dextrose (glucose) molecules released during starch hydrolysis, on dry mass basis. So even though the value of reducing sugar produced increases, if the concentration of sago used also increases without the addition of enzyme and hydrolysis time, the dextrose equivalent produced can't decrease. Adrian et al., (2020) stated that DE decreased because the excess of the given substrate was not converted into reducing sugars because the enzymes used remained constant. Enzyme activity will decrease if the substrate concentration exceeds the optimum concentration, because excess substrate can be an inhibitor for enzyme activity.

The degree of hydrolysis is generally expressed as dextrose equivalent, the quantity indicates the dextrose (glucose) molecules released during hydrolysis of starch, on dry mass basis. The dextrose equivalent value is inversely proportional to the molecular weight, namely the degree of polymerization and as an indicator of the degree of hydrolysis, so glucose has dextrose equivalent of 100 while starch has a dextrose equivalent of zero (Sun et al., 2010).

Several studies have been conducted and show that the dextrose equivalent value can be influenced by the concentration of the substrate used, such as the research of Zadha & Raharjo (2013), regarding the isolation of dextrin from sorghum starch that the increase in substrate concentration is inversely proportional to the equivalent dextrose value obtained so that the addition of starch substrate concentration sorghum causes a decrease in the value of DE in the resulting dextrin. The smaller the starch concentration used and the longer the hydrolysis time used, the greater the DE value obtained until it reaches the optimal value, and inversely, the greater the starch concentration used and the shorter the hydrolysis time used, the smaller the DE value obtained.

Sweetness Level Analysis

The sweetness level is one parameter of how much simple sugars are formed in a product or food ingredient. The level of sweetness (°brix) can also determine the number of solids dissolved in a solution. The total value of dissolved solids was measured using a hand refractometer. The value measured on the °brix scale or hydrometer scale can indicate the percent by weight of sugar present in the solution.

The results of sweetness level analysis showed that the use of 25% sago concentration resulted in sweetness level value of 28.88°brix and increased significantly to the use of 30% sago concentration with sweetness level value of 33.92°brix, then the use of 35% sago yielded sweetness level value of 36.65°brix (Table 4).

The increase in the value of the sweetness level correlated with the increase in the total dissolved solids obtained. Basically, the total dissolved solids of a material include reducing sugars, non-reducing sugars, organic acids, pectin, salts, and proteins which greatly affect the brix. The higher the concentration of sago used, the more substrate that can bind to the enzyme so that more substrate can be converted by the enzyme. Sago contains amylose and amylopectin which can be converted into simple sugars by α-amylase and glucoamylase enzymes, so that with increasing concentrations of sago used, more amylose and amylopectin can react with enzymes and produce glucose which results in an increase in total dissolved solids and affect the sweetness level of glucose syrup. Hadiwijaya et al., (2020) stated that total dissolved solids are the content of water-soluble materials such as glucose, sucrose, fructose, and pectin. Total dissolved solids are often used as an indicator of sweetness. Kalsum & Surfiana (2013) stated DE value also affects the level of sweetness. The higher the DE syrup, the higher the sweetness level. The higher the DE, the easier the syrup to absorb and retain water, so syrup with a high DE is more hygroscopic, so it has a high level of sweetness

Measurement of total dissolved solids value using a refractometer aims to measure total sugar roughly. With the assumption that the higher the total dissolved solids value, the higher the sweetness of the glucose syrup produced. Basically, the total dissolved solids are sugars and various other compounds such as organic acids, soluble amino acids, fats, minerals, and others. The refractometer measures the total dissolved solids based on their refractive index. The refractive index value is obtained from the speed of light in a vacuum compared to when light penetrates the sample. When light penetrates the sample, its speed will decrease. This is due to the presence of dissolved solids in the sample. The higher the concentration of dissolved solids in the sample, the higher the refractive index. This also applies the other way around.

Several studies have been conducted and show that the sweetness level value can be influenced by the concentration of the substrate used, such as the research of

Adrian et al., (2020), regarding the saccharification of white sweet potato into sugar dextrose enzymatically the addition of white sweet potato substrate concentration causes increase in sweetness level. This is possible because the total solids other than reducing sugars are more due to the use of a larger substrate concentration than the others. The level of sweetness is the same as the number of monosaccharides formed, especially glucose which is the final product. The higher the glucose formed, the higher the level of sweetness.

CONCLUSION

The use of α-amylase and glucoamylase enzymes in the manufacture of glucose syrup from sago starch affects the glucose syrup produced. The treatment of sago starch concentration affects the value of reducing sugar and total dissolved solids. The best glucose syrup was obtained from the treatment of 30% sago starch concentration with reducing sugar value of 186.07 g/L, total dissolved solid value 36.13%, DE value of 62.02%, and sweetness level of 33.92ºbrix.

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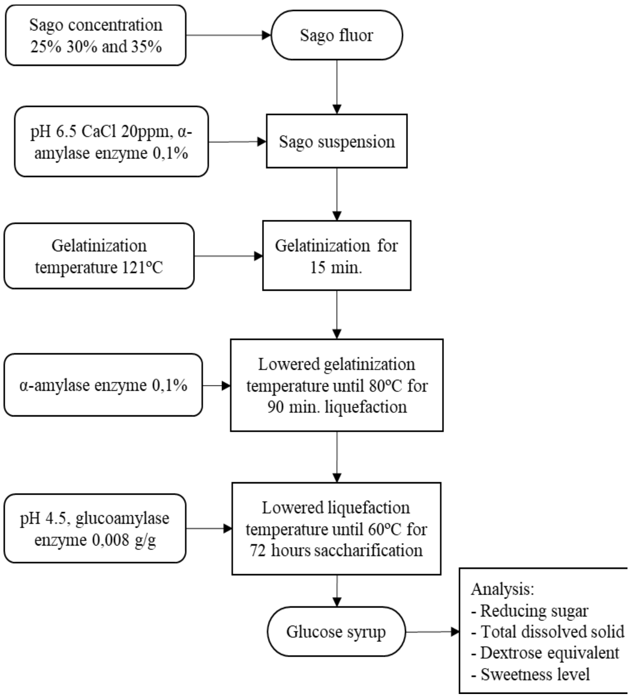

Figure 1. The research process of sago glucose syrup with variations in substrate concentration

Table 1. The Relationship of Variations in Substrate Concentration to Reducing Sugar (g/L)

Treatment	Average	Notation
25%	143.93	a
30%	186.07	b
35%	211.94	c

Table 2. The Relationship of Variations in Substrate Concentration to Total Dissolved Solid (%)

Treatment	Average	Notation
25%	31.09	a
30%	36.13	b
35%	38.83	c

Table 3. The Relationship of Variations in Substrate Concentration to Dextrose Equivalent (%)

Treatment	Average	Notation
25%	57.57	a
30%	62.02	b
35%	60.55	c

Table 4. The Relationship of Variations in Substrate Concentration to Sweetness Level (°brix)

Treatment	Average	Notation
25%	28.88	a
30%	33.92	b
35%	36.65	c

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Survival and Acidification Potential of *Lactobacillus Plantarum* MNC 21 Stored in Air-Dried Sorghum Flours

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ABSTRACT

Increased commercialization of indigenous fermented foods requires availability of affordable starter cultures. The starters should also maintain functionality when stored at ambient conditions, especially where erratic power supply makes constant refrigeration unachievable. This study evaluated the survival of *Lactobacillus plantarum* MNC 21 starter culture air-dried (at 25 or 30°C) in sorghum flour and stored at 25°C for 30 days. Two sorghum varieties (malted and un-malted) were used. To determine their fermentation efficiency during storage, sterile sorghum malt slurries were inoculated with the dried culture and fermented at 30°C for 24 h. Acidification potential was determined at 5 days intervals by measuring microbial counts, pH and titratable acidity. Microbial concentrations dropped from 8-9 log cfu/g on day 0 to 1 log cfu/g on day 30. Sorghum variety and whether it was malted or un-malted did not affect culture survival. Culture dried at 25°C had better survival during the first 10 days (8-9 log cfu/g) than that dried at 30°C (8 log cfu/g) but survival between days 20-30 was similar (1-4 log cfu/g). The acidification potential (ability to reduce pH to ≤4.5) decreased with storage time: 4 h (day 0), 24 h (day 15), > 24 h (day 20) to no acidification (days 25-30). Air drying of starter cultures in sorghum flours coupled with storage at ambient temperatures could be adopted as a short-term preservation method. This low-cost technology is suitable for processors in developing countries where maintenance of a cold chain is hampered by unreliable electricity supply.

Keywords: Air-drying, *Lactobacillus plantarum*, preservation, sorghum, starter cultures

INTRODUCTION

Traditionally, indigenous fermented foods are made by spontaneous fermentation. Wild cultures on the raw materials, utensils, processors and the environment are relied on to initiate the fermentation (Mukisa et al., 2012). Consequently, the sensory attributes and microbiological safety of the products are inconsistent. To address this, researchers have isolated starter cultures for some indigenous fermented foods (Mukisa et al., 2012). This is very important since these foods are becoming popular across ethnic boundaries and consumer preferences, something that has attracted the interest of many entrepreneurs. Therefore, preservation of the starter cultures is required to facilitate commercialization of indigenous fermented foods.

Among the different mechanisms available for starter culture preservation, drying is usually preferred because of easier transportation and better-quality control of the dried cultures (Tripathi and Giri, 2014). Common drying methods include hot-air drying, freeze drying, spray drying, and vacuum drying among others (Tripathi and Giri, 2014). However, some of these methods are associated with high heat, mechanical shearing, dehydration and osmotic pressure which lower cell viability (Tripathi and Giri, 2014). Consequently, protectants or carriers such as skim milk powder, groundnut shells, pea pericarp, safflower shells, cajanus pericarp and sunflower shells are employed to increase cell survival (Gosavi and Bagool, 2013).

Despite the effectiveness of the different drying methods mentioned earlier, the technologies for these methods are quite costly making it difficult for them to be fully adopted, especially in low-income countries. Additionally, the need for cold storage of the dried cultures is challenging, particularly in the rural areas where electricity supply is lacking or unreliable. Therefore, there is a need for a low-cost drying method for starter culture preservation that does not necessitate additional cold storage conditions. Therefore, this study examined the effect of low drying temperatures (25°C and 30°C) on the survival of *Lactobacillus* (*L.*) *plantarum* MNC 21 carried in sorghum flour and stored at 25°C. *L. plantarum* MNC 21 is a strong and fast acidifying culture that was isolated from *Obushera*, a fermented sorghum-millet beverage from Uganda (Mukisa, 2012). The *L. plantarum* MNC 21 preserved in the flour will be used as a starter culture for *Obushera* and related fermented cereal products.

MATERIALS AND METHODS

Microbial culture

L. plantarum MNC 21 isolated from *Obushera* by Mukisa (2012) was used. From the stock 0.1 mL was delivered into 100 mL of sterile MRS broth (CONDA, Madrid, Spain) and incubated at 30°C for 24 h. The LAB was sub-cultured thrice and recovered by centrifugation at 7,500 x g (5600 rpm in a Centrofriger BL-II centrifuge, JP Selecta SA, Barcelona, Spain) at 4°C for 10 min. The cell pellets were then suspended in 10 mL of sterile Ringer’s solution. Culture purity was verified using a microscope (020-518.500 DM/LS I/98, Leica, Germany).

Sorghum varieties

Sorghum was used in this study because it is one of the common staple crops in Uganda and several parts of Africa where it is used to produce non-fermented and fermented foods and beverages (Mukisa, 2012). Two sorghum varieties namely: *Epuripur* (white grained) and *Eyera* (brown grained) were used. The grains were obtained from the National Semi-arid Resources Research Institute in Serere, Uganda. The two sorghum varieties (*Epuripur* and *Eyera*) were chosen for this study because they are among the common varieties of sorghum grown in Uganda. White, brown, high and low tannin varieties were chosen to capture the effects of tannin content on the growth and viability of *L. plantarum* MNC 21.

Preparation of sorghum flours

To malt the sorghum grains, they were first sorted to eliminate foreign matter then washed using pressurized water. Ten kilograms of grain were soaked in 15 L of potable water containing 0.3% NaOH and steeped for 6 h. Thereafter, the water was drained and the steep vessel refilled with fresh water. The grain was further steeped for 10 h after which the water was drained and the grain placed on trays and germinated at 25°C. Germination was halted when the rootlets were about 1cm long; this took three days for *Epuripur* and two days for *Eyera*.

The grain was spread out into a 2 cm thick layer in a drying chamber at 65˚C. The grain was milled using a Wonder Mill (110 Volt model, California, USA) and held in moisture proof containers at ambient temperature (25°C).

Sterilization, inoculation and drying of the sorghum flours

Prior to sterilization, the sorghum flours were first conditioned by sprinkling them with 2 parts of sterile potable water for each part of flour, increasing their moisture content from 11% to 65%. The mixture was then microwaved (Samsung ME731K, Johor, Malaysia) at 600 W for 3 min. The microwaving power and time was arrived at after several preliminary experiments (results not shown). Three minutes was the shortest time required to sterilize the flours when using 600 W. The Total Plate Count, total coliforms and yeasts and molds tests were used to check sterility of the flour. A 10 log cfu/mL suspension of *L. plantarum* MNC 21 was aseptically sprayed into the sterile flour in a ratio of 1:1 (culture suspension: flour). The flour was divided into 2 equal parts and spread out in thin layers of about 5 mm thick. One part was dried at 25°C for 24 h in a sterile class II biology safety cabinet (SterilGARD 403A-HE-INT, Sanford, US) while the other was dried at 30°C for 24 h in a sterile incubator (Binder 240, Tuttlingen, Germany). The dried flour-culture mixtures were separately stored in sterile screw-capped glass bottles at ambient temperature (25°C) away from light. Cell survival was monitored for 30 days using plate counts.

Acidification potential of dried *L. plantarum* MNC 21

Acidification potential of the stored *L. plantarum* MNC 21 was determined at 5 days intervals for 30 days. For this test, sorghum malt flour (*Eyera* variety) was mixed with potable water to make a 1 L slurry of 6.3% total solids. The slurry was heated with continuous stirring to 90°C and held at this temperature for 15 min. The hot gelatinized mixture (100 mL) was aseptically transferred into sterile 250 mL glass bottles and cooled to ambient temperature. It was then inoculated in duplicate with 1 g of sorghum flour (containing about 6 log cfu/mL dried culture) and incubated at 30°C for 24 h. Samples were drawn at 0, 2, 4, 6, 8, 10, 12 and 24 h to determine pH and titratable acidity.

Sample analysis

Enumeration of *L. plantarum* MNC 21 counts, Total Plate Count (TPC) and total coliforms was done by pour plating selected serial dilutions of the sample in MRS agar, Plate Count Agar (PCA) and Violet Red Bile Lactose Agar (VRBLA), respectively. MRS agar was incubated at 30°C for 48 h, PCA at 37°C for 24 h and VRBLA at 37°C for 24 h. To determine yeast and mold counts, selected serial dilutions of the sample were surface spread on sterile preset Potato Dextrose Agar and incubated at 30°C for 5 days. All media was supplied by Laboratorios, CONDA, Madrid, Spain. The pH was determined using a pH meter (AG model, Mettler-Toledo Group, Switzerland). Titratable acidity (TA) was determined by titrating 10 mL of the extract against 0.1N NaOH using phenolphthalein indicator. Moisture content was determined using the hot air-oven method (AOAC, 2000).

Statistical analysis

Results were presented as means ± standard deviations (Mean ± SD) of two independent experiments. Data were subjected to one-way analysis of variance (ANOVA) to test for significant differences at $\alpha = 0.05$. Mean comparisons were made using the Least Significant Difference (LSD) test. Analyses were done using Statistix software (Statistix, 2021).

RESULTS AND DISCUSSION

Microwave sterilization and drying of sorghum flours

Table 1 shows the microbial counts of malted and un-malted sorghum flours before and after microwave sterilization. Microwaving the flours at 600W for 3 min was sufficient to reduce the total plate counts, total coliforms, yeasts and molds to undetectable levels.

Table 2 shows the moisture content of sorghum flours after microwaving, addition of wet culture and drying. Microwaving reduced ($p<0.05$) the moisture content of the conditioned flours from 65% to 3.9-4.2%. Spraying of the flours with wet culture increased their moisture contents to 48.8-49.3%. Drying of the flours containing the culture then lowered their moisture content to 10.8-12.9%.

The results of microbial inactivation by microwave treatment observed in this study agree with those of Najdovski et al. (1991) who achieved vegetative cell destruction at 650W for ≤ 5 min. Microwave energy has the ability to rapidly heat, disinfect and dry materials efficiently (Silva et al., 2021). The principle of operation of microwaves is based on rapid oscillation of water and polar molecules in an alternating electric field leading to friction and thus dissipation of heat energy which is in turn responsible for microbial inactivation (Cebrián et al., 2017; Najdovski et al., 1991). Kormin *et al.* (2013) reported that the heating efficiency of microwaves is determined by the dielectric constant and dielectric loss factor of the heated material.

Dielectric constant is a measure of the ability of the material to store electromagnetic energy while dielectric loss factor relates to the ability of a material to dissipate electromagnetic energy into heat (Budnikov et al., 2020). High dielectric constant and dielectric loss factor result in high and rapid heating of a material by microwave energy. The dielectric constant of a material increases almost linearly with increase in water content due to the strong dipolar character of water molecules (Bhargava et al., 2013). Therefore, conditioning (up to 65% moisture content) increased the dielectric properties of the sorghum flour resulting in effective heating and sterilization.

Safe storage of flours, particularly at ambient temperatures, requires that the flours have moisture content not more than 14% (UNBS, 2017). Otherwise, high moisture content favors growth of unwanted microorganisms such as spoilage fungi and it also accelerates bacterial cell degradation reactions leading to death (Fu et al., 2011). Drying the flours at 25°C and 30°C reduced the moisture content from 48.8-49.3% to 10.8-12.9% (Table 2). This implies that flour tempered to about 50% moisture content can be effectively dried at 25°C within 24 h thus eliminating the unnecessary energy cost associated with air-drying at 30°C. More so since drying at 30°C rather than at 25°C is more likely to lower cell survival because of heat and dehydration stress.

The choice of drying and storage temperatures used was based on the optimal growth temperature (about 28 - 30°C) of *L. plantarum* to minimize cell death due to heat damage. Additionally, these storage temperatures were investigated because they are the common prevailing temperatures in most parts of Uganda. On average, temperatures typically fluctuate between 25 – 30°C. So minimal energy is needed to achieve and maintain them. This is contrast to refrigeration temperatures which require electricity. In developing countries like Uganda, electricity supply is limited and unreliable which is challenge for many of many *Obushera* processors for who the preserved culture is intended. The study aimed at producing a preserved culture that could be safely stored at room temperature by the intended users.

Survival of dried *L. plantarum* MNC 21 during storage

Figure 1 shows the changes in *L. plantarum* MNC 21 counts during storage at 25°C in different sorghum flours. There was a general decrease in the counts throughout storage. *L. plantarum* MNC 21 cell concentrations dropped from 8-9 log cfu/g at day 0 to 1 log cfu/g at day 30. Sorghum flour type (malted vs non-malted) and variety did not affect the survival of *L. plantarum* MNC 21 (p>0.05). *L. plantarum* MNC 21 dried at 25°C had better survival during the first 10 days (8-9 log cfu/g) than that dried at 30°C (8 log cfu/g) but survival between days 20-30 was similar (1-4 log cfu/g). Between days 10-20, survival of cultures dried at 30°C was higher (p<0.05) than for those dried at 25°C. Cell levels dropped to <6 log cfu/g after 15-20 days.

The higher survival of *L. plantarum* MNC 21 dried at 30°C than at 25°C between day 10 and 20 of storage could be due to adaptation to low moisture content. *L. plantarum* MNC 21 dried at 30°C had a moisture content of 11% while that dried at 25°C had 13%. Low moisture content triggers specific transport systems in *L. plantarum* resulting in production of solutes such as carnitine which maintain cell membrane integrity and enhance survival (Kets and De Bont, 1997). High moisture content, as observed for *L. plantarum* MNC 21 stored at 25°C, lowered cell survival due to its acceleration of degradation reactions (Fu et al., 2011).

The survival of dried cultures is also affected by storage temperature. Cell viability greatly reduces with elevated storage temperature as a result of accelerated lipid oxidation and protein denaturation which lead to degradation of vital cell macromolecules (Fu et al., 2011). Nuylert *et al*., (2022) observed that dried cultures retained higher viability when stored at 4°C than at 30°C. Jofré *et al*. (2015) also observed higher survival of starter cultures stored at 4°C than 22°C. Therefore, the storage temperature used in this study (25°C) was also responsible for the decrease in culture survival over time. Nevertheless, storage at ambient temperatures such as 25°C can be adopted for short term storage of the starter culture considering that refrigerated storage cannot be guaranteed due to lack of power or its erratic supply.

The presence of oxygen in storage containers also lowers culture survival over time (Tripathi and Giri, 2014). Although cultures were kept in closed containers, they were not under complete oxygen-free conditions. Stobińska *et al*. (2017) observed improved viability for *Lactobacillus gasseri* stored in an anaerobic chamber than those stored in the presence of atmospheric oxygen. Otero *et al*. (2007) also observed higher cell viability of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* stored under nitrogen and vacuum compared to those exposed to oxygen. Oxygen favors oxidative reactions and accumulation of free radicals within the cell. Failure to metabolize these radicals or transport them out of the cell results in irreversible cell damage (Otero *et al*. 2007). Therefore, viability of the *L. plantarum* MNC 21 starters could be improved by using vacuum packaging.

Acidification potential of the dried *L. plantarum* MNC 21

Figures 2 and 3 show changes in pH and TA of slurries fermented by dried *L. plantarum* MNC 21 stored in different sorghum flours. The acidification potential (ability to reduce pH ≤4.5) of the cultures reduced with storage time. The ability to reduce pH was measured as the time taken to reduce pH to ≤4.5. This time to reduce pH to ≤4.5 increased with increasing storage time:

4 h (day 0), 24 h (day 15), > 24 h (day 20) to no acidification (days 25-30); meaning that the potential to acidify or reduce pH by *L. plantarum* MNC 21 decreased with storage time. Sorghum variety and malting did not (p>0.05) have an effect on the acidification potential of the dried cultures. The amount of acid produced by the stored cultures decreased with time of storage (Figure 3). The final average TA on day 0 day was 0.3% but dropped to 0.1% on day 20.

The activity of lactic acid bacteria starter cultures can be expressed by its rate of acid production, which is a useful parameter in defining effectiveness of the culture in technological processes. As such, acid production determined by pH and %TA is a highly reliable and reproducible method. The drop in pH of the sorghum malt slurries on day 0 of storage (Figure 2) is in agreement with Byakika et al. (2020) and Mukisa *et al*. (2017). *L. plantarum* metabolizes sugars to lactic acid resulting in a pH decline and a corresponding increase in %TA (Byakika et al. 2020). On day 0 of storage the rapid drop in pH was due to the higher cell numbers in the flours than those present in the subsequent days. The number of microbial cells positively corresponds with amount of sugar metabolized and eventually the amount of acid produced. Therefore, the decline in cell counts (Figure 1), explains the decline in the rate of acid production (Figures 2a and 2b). Failure to have any pH drop and increase in %TA on days 25 and 30 was probably because cell numbers were so low (1 log cfu/g) that 1 g of flour was insufficient to initiate fermentation within 24 h. Although cells dried at 30°C had higher survival than those dried at 25°C, between 10-20 days, their fermentation efficiencies were similar (p>0.05) suggesting that high survival does not always translate into high cellular activity.

It is vital that starter cultures rapidly drop pH to ≤4.5 to consequently inhibit pathogens. In this study, it took approximately 2 weeks for the cell concentration of dried *L. plantarum* MNC 21 cultures to drop below 6 log cfu/g (Figure 1) and for them for be able to reduce the pH to ≤4.5 in less than 24 h (Figure 2a). These observations suggest that *L. plantarum* MNC 21 in air-dried sorghum flours can be stored for 2 weeks at ambient temperature before losing its ability to rapidly acidify products.

CONCLUSION

L. plantarum MNC 21 starter culture can be preserved by convective air-drying at 25°C or 30°C in either malted or un-malted sorghum flour. The dried culture can be effective within two weeks of storage at ambient temperature. Therefore, LAB starters may be preserved by air drying in cereal flours and remain effective for at least two weeks without the need for cold storage. This approach is potentially useful for short term storage and distribution of starter cultures, especially in places where refrigeration might be difficult to achieve due to erratic supply or absence of electricity. Further studies should evaluate the possibilities of using vacuum packaging and other protective agents to extend the shelf stability of the stored cultures.

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Table 1. Effect of microwaving at 600 W for 3 minutes on microbial counts of sorghum flours

Sorghum flour	Counts before microwaving (log cfu/g)			Counts after microwaving (log cfu/g)		
	TPC	TC	YM	TPC	TC	YM
Malted *Eyera*	5.4±0.0	5.0±0.1	1.3±0.1	NG	NG	NG
Un-malted *Eyera*	5.6±0.0	5.1±0.1	1.2±0.1	NG	NG	NG
Malted *Epuripur*	5.6±0.0	5.4±0.0	1.0±0.0	NG	NG	NG
Un-malted *Epuripur*	5.4±0.0	5.3±0.0	1.4±0.0	NG	NG	NG

Values are means ± standard deviations, (n = 2). TPC=Total Plate Count, TC=Total Coliforms, YM=Yeasts and Molds, NG=No growth on the 10^{-1} dilution.

Table 2. Moisture content of sorghum flours

Sorghum flour	% Moisture content of flour after microwaving	% Moisture content of flour + LAB before drying	% Moisture content of flour + LAB dried at 25°C	% Moisture content of flour + LAB dried at 30°C
Malted *Eyera*	$(4.2±0.1)^a$	$(48.8 ±0.2)^a$	$(12.9±0.3)^a$	$(11.3±0.2)^a$
Un-malted *Eyera*	$(4.0±0.0)^a$	$(49.1±0.5)^a$	$(13.2±0.4)^a$	$(11.1±0.3)^a$
Malted *Epuripur*	$(3.9±0.0)^a$	$(49.3±0.3)^a$	$(13.1±0.1)^a$	$(11.4±0.1)^a$
Un-malted *Epuripur*	$(4.1±0.1)^a$	$(48.9 ±0.1)^a$	$(12.7±0.1)^a$	$(10.8±0.1)^a$

Values are means ± standard deviations. (n = 2 for MC). Values in the same column with same superscripts are not significantly different (p>0.05). Flour moisture before microwaving was 65%

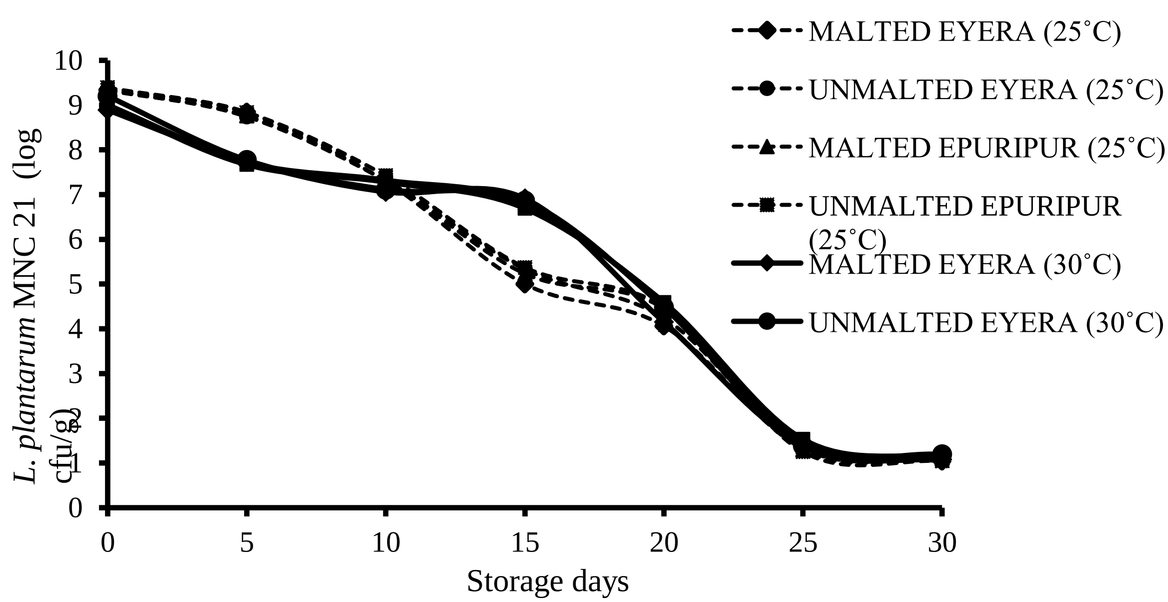

Figure 1. Counts of *L. plantarum* MNC 21dried at 25°C (dotted lines) and 30°C (solid lines) in malted and un-malted sorghum flours stored at 25°C.

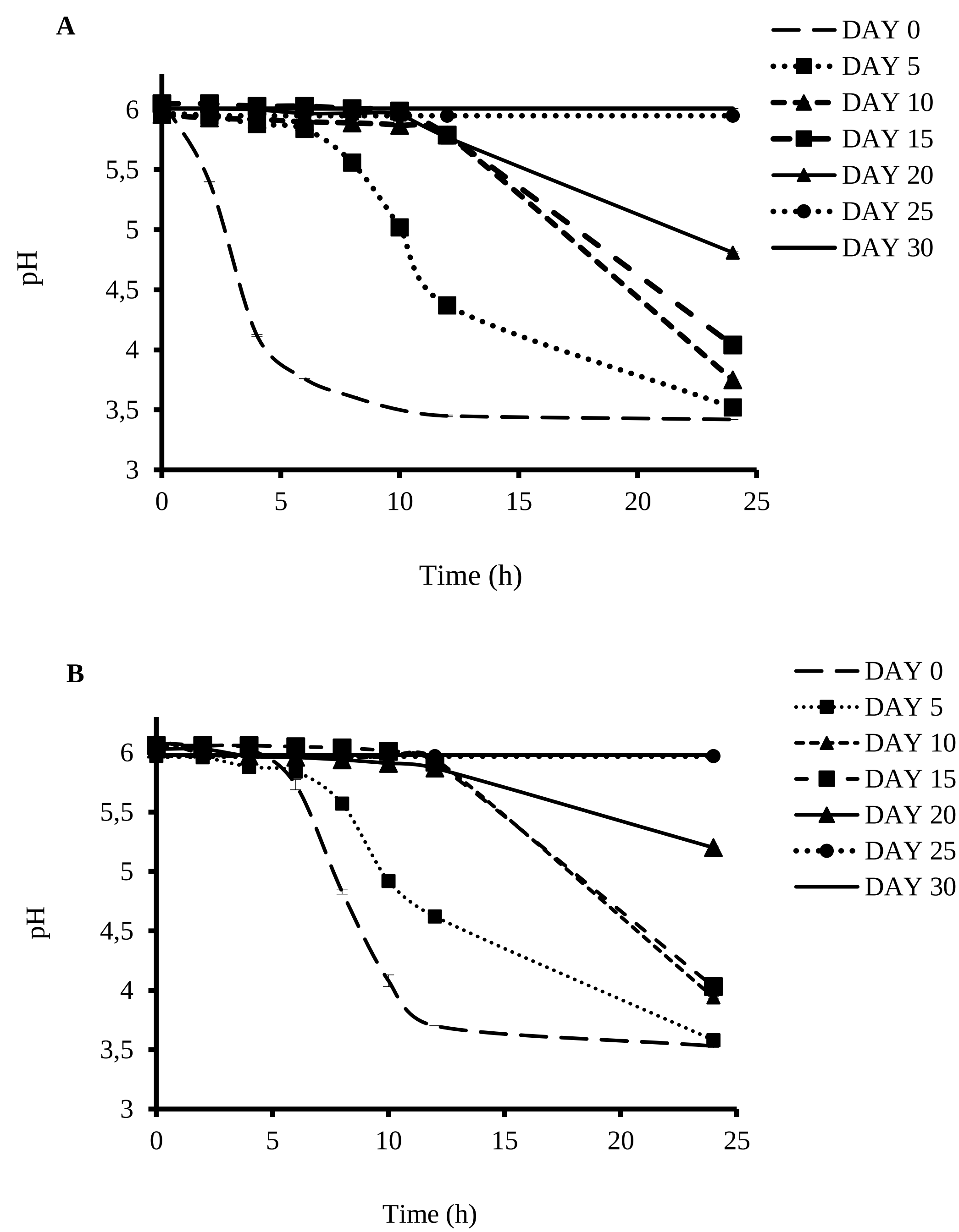

Figure 2. Changes in pH of *Obushera* fermented by *L. plantarum* MNC 21 dried in malted *Eyera* flour at 25°C (A) and 30°C (B), respectively.

The Effect of Beneng Taro (*Xanthosoma undipes* K.Koch) Flour Substitution on Physical and Sensory Characteristics of Muffins

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ABSTRACT

Beneng taro is an indigenous tuber that grows in Pandeglang, Banten. People of Pandeglang only use it in form of fried and steamed. Another effort to utilize beneng taro that has been done is to process it into flour. Beneng taro contains functional compounds including carotenoid pigments and dietary fiber. The use of beneng taro in food can increase product functionality and as food diversification . Muffins are one of the food that people like, but the main composition is wheat flour. The use of local flour in muffin can help the reduce of wheat flour, but will affect the characteristics of the resulting muffin. The purpose of this study was to evaluate the effect of adding beneng taro flour to muffins on the physical and sensory characteristics. This study used a completely randomized design (CRD) with concentration of substituted beneng taro flour (0%, 15%, 20%, 25%, and 30% w/w). The result of this research is the increase of beneng taro flour substitution in muffins decrease the expansion ratio, water content, lightness (L*) of the crust and crumb, a* chromaticity of the crust, b* chromaticity of the crust and crumb, hue (oh) of the crust and crumb, and sensory acceptance. However, The increase of beneng taro flour substitution increase the density of dough, density of muffin, and a* chromaticity of the crumb. The result of sensory evaluation showed that the increase of beneng taro flour substitution decrease the acceptance of aroma, taste, texture, and color. Based on the results of physical and sensory analysis, the muffin formula substituted with 15% beneng taro flour was the best formula.

Keywords: Beneng taro flour addition, food diversification, muffin

INTRODUCTION

Beneng Taro is an indigenous tuber of Banten Province which is commonly found in Pandeglang Regency. "Beneng" is an abbreviation of large (*besar*) and “koneng” (*kuning*) which comes from the Sundanese language which means big and yellow because it has a length of up to 120 cm, a weight of 42 kg, and a diameter of 50 cm (Haliza *et al*., 2012; Yursak *et al*., 2021). Beneng taro contains protein (6.25% wb), ash (3.43% wb), carbohydrate (84.88% wb), amylopectin (70.24% wb), crude fiber (2.29% wb), and dietary fiber (7.19% wb). This nutritional content of beneng taro is the highest among other types of taro (Apriani *et* *al*. 2011). The content of resistant starch, whose functional characteristics such as fiber in taro is greater than wheat and rice (Liu *et al*. 2006). The high content of dietary fiber and resistant starch can make beneng taro as food that has the effect of reducing the risk of coronary heart disease and cancer (Lattimer and Haub 2010). Beneng taro is also a good source of minerals, easy to digest, and absorbed by the body because of its small starch granule size (Yuniarsih *et al*. 2019). Minerals contained in taro include potassium, phosphorus, manganese, and copper (Temesgen and Retta 2015, Soudy *et al*. 2010). Beside that, beneng taro contains beta carotene pigment of 6.92 ppm (Budiarto and Rahayuningsih, 2017).

So far, the people of Pandeglang only use beneng taro by steaming and frying. Another effort to utilize beneng taro that has been done is to process it into flour. The advantages of processing beneng taro into flour is easier packaging and distribution, longer shelf life, and more practical processing. The addition of beneng taro flour in various food products has several objectives: 1) reducing the use of wheat flour and increasing the utilization of beneng taro, 2) increasing the nutritional value of the product, 3) increasing the functional value of a product such as carotene and dietary fiber (Rismaya *et al*. 2018).

One of bakery products that are consumed by many people today is muffin. Muffin is popular because it is practical and taste quite good. However, muffin has a weakness, it has low dietary fiber content. According to Rupasinghe *et al* (2008), the dietary fiber content of muffin made from 100% flour is only 1.30%, because wheat only has a small amount of dietary fiber, only 1.9% (Widaningrum *et al*. 2005).

The addition of beneng taro flour will affect the physical characteristics of the resulting muffins such as color, density, water activity, and expansion ratio. The addition of beneng taro flour is expected to increase the color of the muffins because it contains carotenoid pigments. Dietary fiber in beneng taro flour can affect the physical characteristics of muffin, including reducing expansion ratio, increasing density, reducing water activity, and decreasing organoleptic acceptance (Rismaya *et al*. 2016).

Based on the research of Rismaya *et al*. (2018), the addition of 25% pumpkin flour into muffin became the selected formula based on sensory quality and increased the value of dietary fiber. However, other quality parameters decreased compared to the control, including the volume of expansion decreased by 4%, the color parameter decreased by 31%, and sensory acceptance decreased by 16%. Based on research of Lestari and Maharani (2017), adding 20% of Belitung taro flour to white bread became the selected formula based on sensory quality compared to other formulas. However, other quality parameters decreased compared to the control, including the volume of expansion decreased by 30% and sensory acceptance decreased by 10%. Thus, the addition of beneng taro flour into the muffin in this study is to affect the quality of the muffins produced, so it is necessary to conduct research to determine its effect on the physical and organoleptic characteristics of the muffin.

MATERIALS AND METHODS

Tools and Materials

The material used in making muffin is beneng taro flour obtained from Talaga Warna Village, Pabuaran District, Serang Regency. Muffin raw materials obtained from Pasar Lama, Serang consist of wheat flour with medium protein (Segitiga Biru), margarine (Forvita), chicken eggs, salt (Dolpin), refined sugar (Gulus), mineral water (aqua), and Baking powder (koepoe-koepoe). The tools used are convection oven (Memmert, model UN55), aw meter

(Aqualab Pawkit), Chromameter (Hunterlab Colorflex EZ), desiccator, analytical balance (Excellent, model HZK), crucibles, sensory booth, mixer (Philips, model HR 1552), oven (Cosmos, model CO-9919R), microwave (Samsung, model MS23K3515AS), muffin tin mold, glassware (Pyrex), stationery, paper, millet, muffin cups and kitchenware.

Method

This study was conducted using five treatments, namely control muffins (100% wheat flour), muffins substituted with beneng taro flour with concentration of 15%, 20%, 25%, and 30% and carried out two replications. The research had been carried out in two stages, namely muffin production, and physical and organoleptic properties testing each of which is tested in duplicate. The physical tests carried out were water activity, water content, batter density, muffin density, expansion ratio, and color. The sensory tests carried out were aroma, taste, texture, and color of muffin.

Muffin Production

The formula for the beneng taro muffin refers to the research of Purnomo *et al*. (2014) with modification (Table 1). The flour is sifted to 60 mesh and weighed then put into the mixer bowl. Margarine, water, and salt that have been melted in the microwave are then put together in a mixer bowl and mixed with flour. Chicken eggs are added gradually into the batter and then stirred, then powdered sugar and baking powder are added to the batter and stirred. Batter pour into the muffin cup until ¾ height. The size of the muffin cup used is 4 cm in diameter and 3 cm in height. Then the batter is baked in an electric oven at 200°C for 25 minutes.

Water Content Analysis

Water content analysis refers to the AOAC (2006) method. Empty aluminium crucibles were dried in a convection oven at a temperature of 105°C to constant and weighed (W2). Furthermore, the muffin sample as much as 2 grams (W) was put into a cup and dried in an oven at 105°C until the sample was constant. Then the crucibles containing the sample was weighed its final weight (W1).

$$\text{Water Content (\%wb)} = \frac{W-(W2-W1)}{W} \times 100\%$$

Water Activity (aw) Analysis

The water activity (aw) of muffins was measured using an aw meter according to Ulfah *et al*. (2018), the sample is put into a tube and then put in an aw meter. The screen will show the measurement progress. After the value is stable, the instrument will sound which indicates the water activity measurement process has been completed.

Batter and Muffin Density Analysis

Batter and muffin density analysis are refers to Hasmadi (2010). Density is measured by dividing the weight of the material by the volume of space it occupies. The weight of the batter and muffin products was measured by weighing the muffins using an analytical balance. The volume of muffin batter is measured using a measuring cup by making the weight of the batter as a reference in determining the volume of batter that is put into the muffin cup. Meanwhile, the volume of muffin products was measured using the seed displacement method, using millet as the grain. The millet seeds are put into the measuring cup until the surface is flat, the container is knocked ten times so that all the space in the cup is filled. Furthermore, some of the seeds in the cup are temporarily transferred to another cup. Next, the muffin is put into a measuring container that contains some of the millet seeds and the container is filled with the millet seeds until it is completely flat. The remaining millet seeds were measured in volume with a measuring glass to determine the volume of the muffin (mL). Furthermore, the muffin density was calculated based on the result of dividing the muffin weight by the muffin volume (g/mL).

Expansion Ratio Analysis

The expansion ratio analysis refers to Rismaya *et al.* (2018). Method using seed displacement. The expansion ratio is the ratio between the volume of muffins after baking divided by the volume before baking multiplied by one hundred percent. Muffin volume was measured using the seed displacement method with millet seeds. The millet seeds are put into the measuring container until it is completely flat. After the container is full, some of the seeds are temporarily transferred to another container, then the muffins are put into the container and filled again with millet seeds from another container until they are completely flat. The remaining millet seeds were measured with a measuring cup as the volume of the muffin. The volume of the batter is determined using a measuring cup, muffin batter is first put into the measuring cup and the volume is recorded.

Color Analysis

Color analysis refers to Matos *et al*. (2014) by modifying the type of instrument used. Matos *et al*. (2014) using Konica Minolta CM-3500 chromameter. While the instrument used is the Hunterlab ColorFlex EZ. Color analysis was carried out on the crumb and crust of muffin. Measurement of the crust by placing the sample in the container directly, while the measurement of crumb by cutting the whole muffin at a ¾ height of the top and placing it in the container. Color measurement with the CIELAB system produces values of L*, a*, and b*. the value of L* indicates brightness with a value of 0 (black) to 100 (white), the value of a* indicates green (-a*) to red (+a*) and the value of b* indicates blue (-b*) to yellow (+b*). The degree of hue (^{o}h) is calculated as $\tan^{-1}$ (b*/a*).

Hue 342-18 : Red purple
Hue 18-54 : Red
Hue 54-90 : Yellow red
Hue 90-126 : Yellow
Hue 126-162 : Yellow green
Hue 162-198 : Green
Hue 306-342 : Purple
Hue 270-306 : Blue purple
Hue 198-234 : Blue green
Hue 234-270 : Blue

Sensory Analysis

Sensory hedonic analysis was carried out using the BSN (2006) method based on SNI 01-2346-2006 regarding sensory testing standards. This test aims to determine the level of consumer preference and opinion regarding the beneng taro muffin product. This test was carried out by 35 panelists of Food Technology students, University of Sultan Ageng Tirtayasa with an age range of 19-21 years with female and male gender. The panelists have been given technical guidance on how to give scores on a hedonic scale of 1-7, namely 1: dislike very much, 2: dislike, 3: rather dislike, 4: neutral, 5: rather like, 6: like, 7: really like. This test is carried out with 4 parameters, namely taste, color, aroma, and texture.

Statistical Analysis

The research design used was a completely randomized design (CRD) with one factor, namely flour substitution with beneng taro flour consisting of 2 replications and 5 levels, namely 0%, 15%, 20%, 25%, and 30% w/w. The data obtained then analyzed with ANOVA (Analysis of Variance) at =5% to determine the significant effect on each test parameter. If it shows a significant difference, then proceed with Duncan's Multiple Range Test (DMRT) at α = 5%.

RESULTS AND DISCUSSION

Expansion Ratio

The expansion ratio is one of the important parameters in muffin acceptance. The large volume expansion reflects a more hollow and porous structure, so it has a good acceptability value (Rismaya *et al*. 2018). Based on the results of Duncan's further test, Table 2 shows the average of muffin expansion ratio is decreased with the increase of concentration of beneng taro flour, the results showed a significant difference from muffins which were substituted for beneng taro flour by 15%.

The decrease in the expansion ratio due to an increase in the concentration of beneng taro flour was associated with a decrease in the gluten tissue formed due to the increase of dietary fiber content of beneng taro flour in the batter. The gluten tissue of gliadin and glutenin proteins plays a role in the formation of an elastic film layer that can hold carbon dioxide, so that uniform pores are formed (Wulandari and Lembong, 2016). According to Lestari and Susilawati (2015), beneng taro flour does not have a protein that forms the structure of the gluten tissue which causes a decrease in gluten in the batter. The decrease in the formation of gluten tissues in the batter causes the batter's ability to retain gas during baking to be reduced, so the resulting expansion ratio is low.

The presence of water in the batter is also thought to affect the rheological characteristics of the batter and the quality of muffin products. Water in the batter besides helping the formation of the gluten tissue structure is also needed for the starch gelatinization process, this process is important in the formation of the cake structure (Rismaya *et al*. 2018). According to Muhandri and Subarna (2009), an increase in the water content of the batter causes more water to diffuse into the starch granules which can cause the starch granules to swell and become irreversible, thereby increasing starch gelatinization. The increase in the degree of gelatinization causes more amylose to be released from the starch granules, which functions as a binder which together with the gluten tissue can produce an elastic and cohesive batter mass. The elastic batter plays a role in the formation of the muffin form tissue, so it can be pushed by gas during the baking process.

Dietary fiber has high absorption and water holding capacity which can affect the elasticity of the batter (Struck *et al.* 2016). The dietary fiber content of beneng taro flour reaches 9.52% (Putri *et al.* 2021), while the dietary fiber content of wheat flour is only 1% (USDA, 2022). The higher dietary fiber, the more water will be absorbed, thus lowering the expansion ratio, therefore the more addition of beneng taro flour, the higher the food fiber content will be, and the more water will be absorbed, so the smaller the muffin expansion ratio (Rismaya *et al*. 2018). Water in the batter which is bound by beneng taro dietary fiber will cause the hydration process of starch in the batter to be disturbed so that the starch gelatinization process is hampered and results in decreased elasticity of the batter, less elastic batter causes the formation of muffin structures to be disturbed, as a result of the batter's ability to hold gas during baking decreases.

These results are in similiar with research by Rismaya *et al.* (2016) which substituted pumpkin flour in muffins and Permatanisa and Murtini (2021) research which added soursop puree to muffins. Pumpkin flour has dietary fiber of 14.81% (Trisnawati *et al.*, 2014). The research results of Rismaya *et al.* (2016) showed that the expansion ratio of muffins added with pumpkin flour by 25% decreased to 3% compared to the control. Soursop puree has as much as 3.41% fiber (USDA, 2022). The results of the research by Permatanisa and Murtini (2021) showed that the expansion ratio for muffins that were added 15%

soursop puree decreased to 24% compared to the control.

Density

Density is calculated based on the ratio between the mass of the material and the volume it occupies. A low density value indicates a small mass can fill a large space. The higher the density value, the more compact or dense the product is (Rismaya *et al*. 2018).

Based on the results of Duncan's further test, Table 2 shows the average density of muffin batter, which increases with the concentration of added beneng taro flour, the results show a significant difference starting from the muffin batter substituted with 15% beneng taro flour. A large batter density is thought to reduce the muffin expansion ratio (Rismaya *et al*. 2018). The substitution of beneng taro flour increased the density of the batter and decreased the batter expansion during baking.

The same thing happened to the average density of muffins which increased with the increase of the addition concentration of beneng taro flour. The results showed a significant difference ($p<0.05$) starting from the muffin dough which was substituted for beneng taro by 15%. The increase in the density of the substitution muffin was influenced by the high dietary fiber contained in the beneng taro flour. Similar research conducted by Struck *et al*. (2016) showed that the density of muffins can be increased by substituting several sources of dietary fiber (wheat fiber, pea fiber, apple fiber). In addition, research by Rismaya *et al*., (2018) also showed that the addition of 25% pumpkin flour would increase the muffin density by 31%. Muchtadi (2001) stated that the total dietary fiber component can increase the density of a material or product. Resistant starch can cause crumb muffin matrices to become compact (Baixauli *et al*. 2008). The compact and dense crumb structure causes the muffin volume to be low, so the density of the resulting becomes high.

The higher product expansion ratio value, the lower the density produced. High expansion causes the product have a low mass but high volume, so the density value is low. The high expansion ratio can be described by the shape of the particles which are hollow and porous. The smooth and uniform hollow structure can be used for by the batter's ability to hold gas. Pumpkin flour substitution to muffin decreased the batter's ability to hold gas, resulting in a less hollow structure. Less hollow structures have a low volume with a large mass, resulting in a high density (Rismaya, 2016).

Water Content and Water Activity

Water content is an important component that can determine the stability and durability of food products. In table 2 can be seen that the highest average water content of muffin products is in control muffins (10.53%$\pm$0.44), and the lowest is muffins substituted with beneng taro flour by 30% (8.95%$\pm$0.14). Based on further tests with Duncan, there were significant differences ($p<0.05$) in the water content of control muffins and those substituted with beneng taro flour, while between muffins substituted with beneng taro flour with various concentrations, there was no significant difference ($p>0.05$). In general, the more the concentration of beneng taro flour is added, the water content will decrease. The results are similar with the research of Permatanisa and Murtini (2021) that muffins substituted with 15% soursop puree has a 2% decrease in water content compared to controls and Rismaya et *al.,* (2018) who added 80% pumpkin flour to muffins resulted in a decrease water content as much as 2% compared to control.

Rismaya *et al*. (2018) stated that the addition of pumpkin flour which is high in fiber in the muffins causes a decrease in the value of water content compared to muffins made from wheat flour. The decrease in the water content of the muffins with the increasing concentration of added taro beneng flour was related to the ability of dietary fiber to bind water. The ability of dietary fiber to bind water is related to the role of hydrophilic groups (Winaktu and Gracia, 2011). According to Kusnandar (2010), water chemically bound to hydrophilic groups is hard to remove during the drying process, so that less free water molecules are evaporated. In the beneng taro muffin batter, some of the water is bound by dietary fiber which results in a decrease in free water that can be evaporated, thereby lowering the water content of the beneng taro muffin.

While water activity (aw) is the amount of available water that can be used for enzymatic reactions and microbial growth. Water that can be used for reactions and microbial growth is free water, while the type of water that is evaporated is free water and weakly bound water (Kusnandar, 2010). In table 2, based on Duncan's further test results on aw muffins, there were no significant differences in all muffin formulas ($p>0.05$).

Lightness (L*)

Color is an important factor that is first seen by consumers in determining the acceptance of a product. Color measurement in this research is important to determine the effect of beneng taro flour substitution treatment on the color quality of the resulting muffins. Color testing is carried out on the crust and crumb of muffin.

The L* value represents the lightness parameter with a value of L*=0 meaning black and L*=100 meaning white (Andarwulan *et al*. 2011). Table 3 shows that the value of L* crumb muffins substituted with beneng taro, from the results of further tests was significantly different from the control ($p<0.05$). In general, the higher the substitution of beneng taro, the lower the lightness value of muffin crumb. These results are in similar with research by Rismaya *et al*. (2018) which showed that adding 25% of pumpkin flour to muffins would reduce the L* value by 30% compared to the control. Research by Budoyo *et.al* (2014) also showed that adding 15% of pumpkin flour would reduce the L* value by 13%.

Table 3 shows that the L* value of crust muffins substituted with beneng taro flour at concentration of 25% and 30%, from the further test results were significantly different from the control, but the muffins substituted with beneng taro flour with 15% and 20% were not significantly different from the control. In general, the higher the substitution of beneng taro flour, the lower the lightness value of the muffin crust.

The value of L* in crust ranged from 29.07-39.79 while the value of L* in crumb ranged from 50.21-67.14. This data shows that the lightness of the crust is lower than the crumb. Pictures of muffins with varying concentrations of beneng taro flour can be seen in Figure 1. As beneng taro flour along with wheat flour is a contributor of protein, so intense Maillard browning reaction occurred in the crust, the crumb does not undergo Maillard reaction, but is affected by the ingredients in the batter. Intense maillard browning makes the color of the crust is darker than crumb (Conforti and Davis, 2006).

The decrease in the lightness value of L* in substitution muffins was influenced by the difference in the color of the flour used as raw material of muffins. Beneng taro flour has a darker color (low L*) than wheat flour (Wongsagonsup *et al*. 2015; Rostianti *et al*. 2018). Beneng taro flour has a yellow color due to the presence of carotenoid pigments of 6.92 ppm (Budiarto and Rahayuningsih 2017). The decrease in lightness value (L*) is also influenced by the formation of brown color due to non-enzymatic browning reactions (Maillard reaction). The formation of brown color is more dominant in the substitution muffins due to the high content of reducing sugar in the beneng taro flour. In addition to the presence of amino acids and reducing sugars, the Maillard reaction is also influenced by water content (Laguna *et al* 2011; Kusnandar 2010). The high water content in the batter tends to shift the reaction towards the formation of N-substituted glycosylamine as a precursor for the formation of melanoidin compounds to be inhibited (Kusnandar, 2010). This melanoidin compound plays a role in the formation of the brown color in muffins. In muffins substituted with beneng taro flour, the presence of water is bound to dietary fibers so that the presence of water in the batter is reduced. The presence of low water in the beneng taro batter muffin causes the reaction to shift towards the formation of more melanoidin compounds, until the brown color becomes more dominant. These results are in accordance with the research of Rismaya *et al*. (2018) which stated that the lower water content of the batter causes the color of the pumpkin muffins to be darker.

a* Chromaticity

The a* chromaticity value indicates green (negative) to red (positive) color (Santoso *et al*. 2013). The average value of a* chromaticity of crust muffins produced ranged from 14.24-19.26, and the average value of a* chromaticity of crumb muffins ranged from 5.93-8.36, with the highest average of a* chromaticity obtained in muffins substituted with 30% beneng taro flour ,namely 8.36 for crumb and 19.26 for crust, and the lowest average value of chromaticity a* was found in muffin control, namely 5.93 for crumb and 14.24 for crust. The average value of a* chromaticity of muffins shows that the average value of a* chromaticity is positive which indicates a tendency towards the red color.

Table 3 shows that the values of a* crust muffins in substitution of beneng taro flour from the further test results were significantly different from the control ($p<0.05$), Significantly different results were shown starting with substituted muffins of 20%. In general, the higher the substitution of beneng taro flour, the lower a* chromaticity value of the muffin crust. Table 3 shows that the a* chromaticity value of the beneng taro crumb muffin of substitution was 30%, the results of the further test were significantly different from the control but not significantly different from the other formulas. In general, the higher the substitution of beneng taro flour, the higher the value of a* chromaticity crumb muffin.

The results of a* chromaticity in crumbs indicate a tendency that the higher concentration of substituted beneng taro flour added, the higher a* chromaticity value. A high a* chromaticity value was found in muffins with a dark surface (L* low) (Rismaya 2016). These results are in similiar with the study of Budoyo *et al*. (2014) which showed an increase of 66% in the a* chromaticity muffin substituted with pumpkin flour by 15% compared to the control. The increase in a* chromaticity value was influenced by the formation of a more dominant brown color resulting from the Maillard reaction that occurred in the batter. While the chromaticity value of a* crust muffins shows the opposite, this is presumably due to the effect of oven baking which produces a slightly uneven heat on the surface of the muffin crust so that it accelerates the formation of color tends towards redness on the surface of the muffin which is more exposed to heat than crumb.

b* Chromaticity

The value of b* chromaticity shows a blue (negative) to yellow (positive) color

(Santoso *et al*. 2013). The average value of b* chromaticity crust muffins produced ranged from 17.33-31.94, and the average value of b* chromaticity crumb muffins ranged from 25.81-37.09, with the highest average of b* chromaticity obtained at 30% control muffins, namely 37.09 for crumb and 31.94 for the crust. The average value of b* chromaticity of muffins shows that the average value of b* chromaticity is positive which indicates a tendency towards yellow.

Table 3 shows that the value of b* chromaticity crust muffins with substitution of beneng taro flour of 20%, 25%, and 30%, from the further test results were significantly different from the control ($p<0.05$), Significantly different results were shown starting with muffins which were substituted by 20%. In general, the higher the substitution of beneng taro flour, the lower the of b* chromaticity crust muffins. Table 3 shows that the b* chromaticity values of crumb muffin at concentrations of 15%, 20%, 25%, and 30%, from the further test results were significantly different from the control ($p<0.05$).

These results are similiar with the study of Budoyo *et al.,* (2014) which showed a 20% decrease in the b* chromaticity value muffin substituted with pumpkin flour by 15% compared to control. The more concentration of substituted beneng taro flour in muffins causes a decrease in the value of b* chromaticity which is thought to be due to the yellow-orange color due to the natural pigment content of beneng taro (carotenoids) and the formation of dark brown color resulting from the Maillard reaction is more dominant in substitution muffins (Rismaya, 2016). The decrease of b* chromaticity because of the forming of melanoidin pigment from Maillard reactions from baking process. The presence of carotenoid pigments in muffin is covered by melanoidin pigments (Rismaya, 2016).

Hue (°h)

Chromatic color or Hue is a real color that can be observed by the eye, such as red yellow blue, green and so on (Andarwulan *et al*. 2011). The measurement values of L*, a* chromaticity and b* chromaticity can be converted to Hue (°h) values (Santoso *et al*. 2013). In general, the Hue of crust muffin values obtained ranged from 87.93-102.40 which indicated the color range of yellow red (Hue 54-90) and yellow (Hue 90-126). Meanwhile, the value of Hue crumb muffins obtained ranged from 125.78-141.29 which indicated the yellow color range (Hue 90-126) and yellow green (Hue 126-162). Table 3 shows that further test results showed that all muffins with various concentrations of beneng taro flour showed a Hue crust value that was not significantly different with control muffins ($p>0.05$). However, based on the results of further tests showed that all muffins with various concentrations of beneng taro flour showed significantly different hue crumb values with the control ($p<0.05$), the hue value showed a significant difference in muffins which were substituted for beneng taro flour by 30%. In general, the higher the concentration of beneng taro flour substitution, the lower hue value in the crust and crumb muffins.

These results are consistent with research by Rismaya (2016) which showed a 12% decrease in hue values in muffins substituted with pumpkin flour by 25% compared to controls. This decrease of hue value indicates the color of the muffins substituted with beneng taro flour tends to yellow for the crust and greenish yellow for the crumb which is more dominant. Meanwhile, wheat flour did not have a yellow and greenish yellow color, so the control muffins did not have a greenish crumb color and a yellowish crust. According to Muchtadi (2010), the difference in the color of the crumb in the product is strongly influenced by the color of the flour used as raw material. Beneng taro flour has a yellow color that comes from carotenoid pigments. Carotenoid pigments in beneng taro flour about 6.92 ppm (Budiarto and Rahayuningsih, 2017). In addition, the color difference is also influenced by the formation of a brown color due to the Maillard reaction that occurs between reducing sugars and free amine groups (Man *et al*. 2014; Struck *et al*. 2016). The higher sugar and starch content of beneng taro, which is 81.81% (Kusumasari *et al*., 2019) causes the formation of brown color due to the Maillard reaction to occur more in substitution muffins.

Sensory Characteristics

Aroma is one of the sensory properties received by the sense of smell that can affect the level of sensory acceptance. Based on table 4, Duncan's further test results showed that there were no significant differences aroma in all types of muffin formulations ($p>0.05$), the value shows that panelists give neutral acceptance. These results are in similar with the research of Lestari and Susilawati (2015) who substituted beneng taro flour for noodles, giving the result that the higher the concentration of beneng taro flour did not give significantly different results to the aroma. This result is presumably because the aroma of beneng taro flour tends to be neutral so that it does not have a significant effect on the panelists' sense of smell even though there is a difference in the concentration of substituted beneng taro in muffins.

Color is a factor that must be considered in product development, because panelists will evaluate the product first through its visual appearance. Based on table 4, Duncan's further test results showed that there was a significant difference in panelists' acceptance of the color of the control muffins with muffins substituted with beneng taro flour in various concentrations ($p<0.05$). Panelists give neutral for color for the muffin. The more beneng taro flour added to the muffins will cause a decrease in color acceptance because the resulting color impression is dark due to the presence of amino acid, carotenoid pigments, and the Maillard reaction that causes brown color due to the higher sugar and starch content of beneng taro than wheat flour. Carotenoid pigments in beneng taro flour about 6.92 ppm (Budiarto and Rahayuningsih, 2017). These results are in similair with the research of Lestrai and Susilawati (2015), the more taro beneng flour added to the noodles, the panelist's preferred color will decrease due to browning. In addition, because the brown color of flour is caused by the content of the amino acid lysine in taro tubers which will increase the brown color, lysine contains two amino groups so that it is more reactive to reducing sugars and produces a more concentrated brown color (Kurniawati and Ayustaningwarno, 2012).

The taste attributes depend on the composition of the ingredients used in the production of the product. Table 4 shows the results of Duncan's further test showing that the level of panelists' acceptance of the control muffin taste was significantly different ($p<0.05$). Significantly different results began to be shown in muffins which were substituted for beneng taro flour by 15%. The acceptance of the muffin control taste was rather like while the other four tended to be neutral. The higher the concentration of beneng taro flour, it tends to decrease the panelists' acceptance of taste. This is due to the high content of oxalate compounds in beneng taro flour, which is 648.87 ppm (Budiarto and Rahayuningsih, 2017). The more addition of beneng taro flour, the higher the oxalate compounds it contains. Oxalate compounds can stimulate the oral cavity and skin, causing itching or irritation. This itching effect can occur when calcium oxalate crystals are released and can irritate the skin causing small holes on contact (Aviana and Loebis, 2017).

Texture is one of the sensory properties of food products that are important in consumer acceptance. One of the parameters for good muffin quality is the crumb texture which is soft and not hard (Rismaya *et al.,* 2018). Table 4 shows that the results of Duncan's advanced test showed no significant difference between the control and muffins substituted with 15-25% taro flour ($p>0.05$), and began to show significant differences with muffins substituted with 30% ($p<0.05$). The panelist's assessment of the muffins substituted for beneng taro flour was somewhat disliking to neutral. The results of this study are in accordance with Rismaya *et al*. (2018) and Budoyo *et al.* (2014) which showed that the more pumpkin flour added to the muffins, the lower the panelist acceptance. Gluten is able to form films that are not easily torn, elastic, and extensible so that they can trap CO2. Substitution of flour with a higher dietary fiber content than wheat will reduce the availability of gluten so that the gas trapping ability also decreases causing the texture of the muffins to become tougher (Budoyo *et al.,* 2014).

CONCLUSION

The increase of beneng taro flour substitution in muffins decrease the expansion ratio, water content, lightness (L*) of the crust and crumb, a* chromaticity of the crust, b* chromaticity of the crust and crumb, hue (^{o}h) of the crust and crumb, and sensory acceptance. However, The increase of beneng taro flour substitution increased density of batter, density of muffin, and a* chromaticity of the crumb. Changes in the physical and sensory properties of muffins are related to the presence of dietary fiber in beneng taro flour. Based on the results of physical and sensory analysis, the muffin formula substituted with 15% beneng taro flour was the best formula. Although the control muffins had a higher acceptance score by panelists, expansion ratio, lightness, and hue, but the muffins substituted with beneng taro flour presence of functional components of dietary fiber and can help reduce the using of wheat flour.

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Table 1. Muffin basic formula design

Ingredient (g)	F1	F2	F3	F4	F5
Wheat Flour	525	446.25	420	393.75	367.5
Beneng Taro Flour	-	78.75	105	131.25	157.5
Margarine	345	345	345	345	345
Water	163	163	163	163	163
Salt	3	3	3	3	3
Chicken Egg	300	300	300	300	300
Refined Sugar	380	380	380	380	380
Baking Powder	10	10	10	10	10

Note: F1 = Control / Concentration of Wheat Flour 100%, F2 = Concentration of Beneng Taro Flour 15%, F3 = Concentration of Beneng Taro Flour 20%, F4 = Concentration of Beneng Taro Flour 25%, F5 = Concentration of Beneng Taro Flour 30%

Table 2. Average Value of several physical properties of muffins

Formula	Expansion Ratio (%)	Batter Density (g/mL)	Muffin Density (g/mL)	Water Content (%)	Water Activity
F1	276,56±2,21^{a}	1,15±0,01^{a}	0,37±0,02^{a}	10,53±0,44^{a}	0,67±0,03^{a}
F2	245,83±3,54^{b}	1,21±0,00^{b}	0,44±0,01^{b}	9,31±0,12^{b}	0,70±0,01^{a}
F3	238,39±3,79^{bc}	1,30±0,00^{c}	0,47±0,00^{b}	9,02±0,12^{b}	0,71±0,00^{a}
F4	235,04±4,09^{c}	1,31±0,00^{c}	0,47±0,02^{b}	8,96±0,14^{b}	0,71±0,05^{a}
F5	225,89±1,26^{d}	1,35±0,00^{d}	0,52±0,03^{c}	8,95±0,14^{b}	0,71±0,02^{a}

Note: F1 = Control / Concentration of Wheat Flour 100%, F2 = Concentration of Beneng Taro Flour 15%, F3 = Concentration of Beneng Taro Flour 20%, F4 = Concentration of Beneng Taro Flour 25%, F5 = Concentration of Beneng Taro Flour 30%, Value with different notation in the same column has a significant difference at 5% (Duncan test)

Table 3. Average Value of muffin color attributes

Part of Muffin	Formula	Color Attributes			
		L*	a*	b*	Hue (°h)
Crust	F1	39,79±3,64^{a}	19,26±0,63^{a}	31,94±3,93^{a}	102,40±6,87^{a}
	F2	38,53±2,97^{a}	18,11±0,58^{ab}	28,33±3,11^{ab}	99,90±6,12^{a}
	F3	34,30±2,54^{ab}	17,03±0,39^{bc}	23,58±2,39^{bc}	94,30±4,20^{a}
	F4	30,77±0,59^{b}	16,18±0,42^{c}	19,85±1,52^{c}	88,60±2,47^{a}
	F5	29,07±1,04^{b}	14,24±1,30^{d}	17,33±2,90^{c}	87,93±4,41^{a}
Crumb	F1	67,14±1,06^{a}	5,93±0,84^{a}	37,09±2,53^{a}	141,29±1,14^{a}
	F2	54,64±0,07^{b}	7,30±0,54^{ab}	26,34±3,95^{b}	129,85±1,97^{b}
	F3	52,10±1,92^{bc}	7,48±0,14^{ab}	23,95±1,39^{b}	126,75±1,11^{bc}
	F4	50,32±0,63^{c}	7,52±0,20^{ab}	23,26±0,32^{b}	125,93±0,37^{bc}
	F5	50,21±0,54^{c}	8,36±0,26^{b}	25,81±0,06^{b}	125,78±0,85^{c}

Note: F1 = Control / Concentration of Wheat Flour 100%, F2 = Concentration of Beneng Taro Flour 15%, F3 = Concentration of Beneng Taro Flour 20%, F4 = Concentration of Beneng Taro Flour 25%, F5 = Concentration of Beneng Taro Flour 30%, Value with different notation in the same column has a significant difference at 5% (Duncan test)

Table 4. Average panelist acceptance level of muffin sensory attributes

Formula	Panelist Acceptance Level			
	Aroma	Color	Taste	Texture
F1	4,43±1,20^{a}	6,14±1,09^{a}	5,46±1,24^{a}	4,63±1,40^{a}
F2	4,77±1,24^{a}	5,43±1,09^{b}	4,86±1,22^{ab}	4,43±1,29^{a}
F3	4,60±1,54^{a}	3,57±1,31^{c}	5,11±1,08^{ab}	4,31±1,32^{a}
F4	4,14±1,38^{a}	4,23±1,42^{c}	4,80±1,45^{c}	4,46±1,52^{a}
F5	4,26±1,36^{a}	3,60±1,63^{c}	4,54±1,27^{c}	3,34±1,35^{b}

Note: F1 = Control / Concentration of Wheat Flour 100%, F2 = Concentration of Beneng Taro Flour 15%, F3 = Concentration of Beneng Taro Flour 20%, F4 = Concentration of Beneng Taro Flour 25%, F5 = Concentration of Beneng Taro Flour 30%, Value with different notation in the same column has a significant difference at 5% (Duncan test)

F1 F2 F3 F4 F5

Figure 1. Beneng Taro Muffins with Different Formulations
Note: F1 = Control / Concentration of Wheat Flour 100%, F2 = Concentration of Beneng Taro Flour 15%, F3 = Concentration of Beneng Taro Flour 20%, F4 = Concentration of Beneng Taro Flour 25%, F5 = Concentration of Beneng Taro Flour 30%

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Oxide-Based Nanocomposites for Food Packaging Application: A Review

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ABSTRACT

Silver nanoparticles and/or nanoclay [particularly montmorillonite] are used in the majority of nanotechnology applications for food packaging. Other nanomaterials, on the other hand, can also be integrated into packaging. Metal oxide nanoparticles have been added to petroleum-based and biopolymers to produce nanocomposites with improved mechanical, barrier, antioxidants and antimicrobial properties. Nanoparticles migration from packaging, on the other hand, is a source of concern due to their potential toxicity in the human body and the environment. The purpose of this article therefore, was to review the available literature on the utilization of metal oxide-based nanoparticles to produce nanocomposites for food packaging application. Advantages of incorporating metal oxide-based nanoparticles into polymers, as well as migration of these nanomaterials from packaging into foods are discussed. Incorporation of metal oxide nanoparticles into polymers allows for the production of nanocomposites with increased mechanical strength, water and oxygen barrier properties, and can also confer other additional functional properties, such as antioxidant, antimicrobial activity and light-blocking properties. According to migration studies, only a small quantity of nanomaterial migrates from packaging into food simulants or foods, implying that consumer exposure to these nanomaterials and the health concerns associated with them are low. Nonetheless, there is a scarcity of information on the migration of nanomaterials from packaging into actual foods, and more research is desperately needed in this area. This manuscript is useful in the food industries as it indicates the applicability and potential of the oxide-based nanocomposites as a promising approach for use in food packaging applications.
Keywords: Beneng taro flour addition, food diversification, muffin

INTRODUCTION

Food packaging plays a vital role in food safety, quality, and shelf-life extension by protecting food products from physical, environmental, chemical, and microbial hazards during storage and transportation (Youssef & El-Sayed, 2018; Hoseinnejad et al., 2018). In the international market, packaging also simplifies product end-use suitability and communication at the consumer level. New packaging materials have been developed because of advances in materials science and a need for nutritious food with few additives that stay fresh for longer. Multilayered packaging has improved "barrier properties" and allowed new product lines to be developed like retorted foods; yet this packaging is expensive to manufacture and difficult in recycling (Bumbudsanpharoke & Ko, 2015; Duncan, 2011). As a result, developing monolayer packaging having improved properties, transparency, lightweight, biodegradability or ease of recycling, is required, and nanotechnology can assist in this endeavour (Chaudhry et al., 2008; Garcia et al., 2018).

Nanotechnology is a multi-disciplinary discipline in which materials with at least one dimension between 1–100 nanometers are used (Bumbudsanpharoke & Ko, 2015; Mihindukulasuriya & Lim, 2014). Nanomaterials have a larger surface area than their bulk equivalents due to their small size, resulting in increased reactivity and properties that differ from macroscale level materials (Uskokovi, 2007). Nanomaterials and polymers can thus be mixed to produce nanocomposites having improved barrier, mechanical and thermal properties. As nanoparticles are incorporated into the polymer matrix, they act as reinforcement, and their lesser water and gases permeability causes a diffusion path that is longer and more convoluted, resulting in improved barrier properties in comparison to neat polymer. In addition, bonds formation between the nanoparticles and polymer reduces the sites number that may interact with water molecules in the polymer chain (Duncan, 2011; Mihindukulasuriya & Lim, 2014). A nanomaterial's high aspect ratio and homogenous dispersion in a polymer matrix alters the molecular mobility and relaxation of polymer chains, therefore enhancing mechanical and thermal resistance (Bumbudsanpharoke et al., 2015). Furthermore, the increased strength of nanocomposites may be due to the bond between polymer chains and nanoparticles (Zhou et al., 2009; Garcia et al., 2018).

Using nanotechnology involving metal oxide nanoparticles and either petroleum–based or biobased biopolymers, "active packaging" & "intelligent packaging" can be developed. Food is protected by active packaging, via mechanisms activated by internal or external factors, whereas "intelligent packaging" offers info on the food's real status (Chaudhry et al., 2008; Ding et al., 2020; Mihindukulasuriya & Lim, 2014). Direct contact between metal oxide nanoparticles and microbial cells and/or the release of antimicrobials like metal ions (Zn^{2+}, Ag^{+}, Cu^{2+}, Ti^{4+}, etc.) and reactive oxygen species are required for active packaging to demonstrate antimicrobial activity (Garcia et al., 2018; Jafarzadeh et al., 2020); nevertheless, the release of antimicrobials must be regulated, and nanoparticles migration is undesired owing to the unknown outcome on the human body (Llorens et al., 2012; Duncan, 2011).

Some of the nanomaterials that can be included into packaging are nanosilver and other metals plus their oxides, "nano-cellulose", "essential oil nano-emulsions" and "nanoclays" (Bumbud-sanpharoke & Ko, 2015; Eleftheriadou et al., 2017; Llorens et al., 2012; Mihindukulasuriya & Lim, 2014). Despite the fact that the number of articles on novel forms of nanocomposites is growing, the scientific literature is still predominated by nanosilver and nanoclay applications (Bumbudsanpharoke & Ko, 2015; Mihindukulasuriya & Lim, 2014).

Metal oxide nanoparticles that are also utilized in food packaging include Aluminum oxide, Silica, Titanium dioxide and Zinc oxide. These nanomaterials are commonly utilized as photocatalysts having antimicrobial plus ethylene-scavenging properties, and they can as well improve the nanocomposites' "tensile strength", barrier properties to UV & gas (Bumbudsanpharoke & Ko, 2015; Llorens et al., 2012). These properties would be beneficial for packaging fresh foods that lose water readily and are susceptible to spoilage by microbes and enzymes (Llorens et al., 2012; Garcia et al., 2018). However, the various features of nanomaterials that make them superior to their bulk equivalents may also lead to a variety of toxicological properties, raising consumer concerns. Thus, to assure the safety of nanocomposite packaging, nanomaterials migration from packaging into foods must be identified (Störmer et al., 2017; Garcia et al., 2018).

This review article therefore focuses on metal oxides as nanomaterials for the purpose of food packaging. Although there are some reviews on uses of nanomaterials in food packaging, their scopes is different from the current review. The focus of previous studies was on the general perspective for nanomaterials in packaging (Bumbud-sanpharoke et al., 2015; Mihin-dukulasuriya & Lim, 2014), market situation (Bumbudsanpharoke & Ko, 2015), and regulations (Bumbudsanpharoke & Ko, 2015; Wyser et al., 2016). To the best of our knowledge, there are lack of reviews focusing on nano-metal oxides in packaging. Therefore, the purpose of this article was to summarize the advantages of integrating nano-metal oxides into polymer-based food packaging, and to provide information on how such nanomaterials may migrate into packaged foods. Metal oxides like Aluminum oxide (AlO_x), Copper oxide (CuO), Magnesium oxide (MgO), Silicon dioxide (or Silica, SiO_2), Titanium dioxide (TiO_2) and Zinc oxide (ZnO) are discussed. The information presented here is expected to complement earlier publications and aid in drawing conclusions about the safety of nanocomposites used in food packaging. The current search was conducted among PubMed, ScienceDirect, Embase and Scopus databases up to 2021 to retrieve the related articles. The following keywords were included "nanocomposite" OR "nanomaterial" OR "nano" OR "nano-food packaging" OR "nano-packaging" OR "Metal oxide" "AND "Packaging" OR "food packaging" OR "food AND "migration". The reference of retrieved articles was checked for additional articles. Full text articles in the English language were included. Review articles, correspondence, thesis, letter to the editor, and conference were excluded.

NANOSCALE METAL OXIDES IN FOOD PACKAGING

The most recently explored materials in food packaging are metal oxides (MOs) & mixed metal oxides (MMOs). The use of silver-NPs and/or nanoclay (particularly montmorillonite (MMT)) in Nanotechnology has received a lot of attention. MOs and MMOs, on the other hand, have structural and morphological features that make them promising nanoparticles in a typical composite profile. It's still challenging to synthesize suitable MMOs with the right physicochemical properties that account for the strong interaction with the polymer matrix (Atkins et al., 2006 Jafarzadeh et al., 2020).

When the metal has an oxidation number of I, II, or III, M^+ and M^{2+} ions (MxO: x = 1, 2, 3, etc.) form oxides, and are categorized as: i) MO having a rock salt structure. ii) M_2O having an antifluorite or rutile structure. (iii) M_2O_3 oxides (Atkins et al., 2006; Jafarzadeh et al., 2020).

The metal oxides are formed by defects that differ from structure to structure. The size of the defects is related to the metal oxides type, as well as the changes in partial pressure of oxygen above a metal oxide, both of which cause the lattice parameter and equilibrium composition to fluctuate continuously. Antimicrobial activity is found in a variety of d-block MOs, many of which are n-type semi- conducting MOs, such as CuO, ZnO, TiO_2, MgO, and others. Their activity may be due to minute differences in stoichiometry and O-atom defects (Jafarzadeh et al., 2020).

Metal oxide nanoparticles have been mixed into either biobased or petroleum-based polymers, to make nanocomposites with improved properties and inherent antimicrobial properties. On the other hand, nanoparticles migration from packaging films is a cause for concern due to their possible toxicity in the environment and the human body as well. In packaging, several nanoscale metal oxides and structural-based nanoparticles (such as clay) are utilized to enhance their antimicrobial activities, barrier plus mechanical properties and thermal stability. This section will cover recent breakthroughs in metal oxides-reinforced polymeric nanocomposites for food packaging, such as Aluminium (Al), Copper (Cu), Magnesium (Mg), Silicon (Si), Titanium (Ti) & Zinc (Zn) oxides (Jafarzadeh et al., 2020).

OXIDES USED IN NANOCOMPOSITES FOR FOOD PACKAGING

Aluminum Oxide (AlOx)

For long, Aluminum has been utilized in metallized films to safeguard the packaged foods from ultraviolet irradiation and oxygen. These films, nonetheless, are opaque, thick, non-microwavable and recycling is arduous. Aluminum oxide-based coatings, which are light, transparent, and microwavable, are a viable alternative to such films (Struller et al., 2014). Physical vapour deposition or chemical vapour deposition processes are used to produce aluminum oxide thin films, which are utilized as microelectronics coatings (Cibert et al., 2008). Furthermore, atomic layer deposition has been used to produce Al_2O_3 thin films, which have superb barrier properties and have been advocated for use in food packaging, specifically biodegradable packaging (Hirvikorpi et al., 2010).

Störmer et al., (2017) describe silicon oxide and aluminium oxide coatings as "nanocoatings" or "ultrathin films" because they may produce stable layers that are uniformly scattered and have a thickness of 10–100 nanometers. Some researchers suggest that such films should be classified as "nanostructures" rather than "nanomaterials" under European regulations because they are different from principal nanoparticles and form strong bonds with polymers (Bott et al., 2014; Vähä-Nissi et al., 2015).

Coating board paper with PLA-Al2O3 and PLA film double-coated with alginate-chitosan layer and Al_2O_3 layer significantly improved their aroma, oxygen, and water barrier properties (Hirvikorpi et al., 2011; Hirvikorpi et al., 2010). The water vapour transmission rate of PLA double-coated was 25 g/m^2/day, which was significantly better than pure PLA (53 g/m^2/day) and much nearer to the permissible value for dry food packaging (0–10 g/m^2/day) (Hirvikorpi et al., 2011). It has also been reported that coating polyethylene terephthalate with aluminium oxide improves the composite films barrier properties, suggesting that it could be a viable substitute to the currently used metallized retortable packaging; results of biaxially oriented polypropylene coating, on the other hand, were inconsistent (Struller et al., 2014), indicating that further study into AlOx nanocomposites is required (Garcia et al., 2018).

COPPER OXIDE-BASED NANO MATERIALS

The two primary categories of materials in food packaging are copper nanoparticles (CuNPs) and copper oxide nanoparticles (CuONPs). CuONP, on the other hand, is one of the most investigated metal oxides in industrial food packaging, because of its antimicrobial activity, which inhibits viruses, fungi and bacterial growth (Kuswandi & Moradi, 2019). Because of their large surface area, CuONPs interact with the cell membrane. Sonochemical (Ávila-López et al., 2019), microwave (Karunakaran et al., 2013), autocombustion (Kamble & Mote, 2019), electrochemical (Jadhav et al., 2011), thermal decomposition (Ibrahim et al., 2018), and other methods can all be used to produce nanosized CuO.

In bioorganisms, copper is a key element for metabolism and transport of electron. Copper levels above a certain threshold can inhibit bacterial cell development. CuO's mechanisms for limiting cell growth can be split into 4 categories:

I. Enzymes inactivation II. Essential ions exchange III. Generation of H_2O_2 free radicals, attacking protein functional groups IV. Breaking the plasma membrane's integrity (Jafarzadeh et al., 2020).

Cu2+-incorporated poly(3-hydroxybutyrate- co-3-hydroxyvalerate) (PHB-co-3HV) (Castro et al., 2018), agar (Roy & Rhim, 2019), cellulose (Muthulakshmi et al., 2019) and chitosan (Almasi et al., 2018), have all been investigated for antimicrobial activity in packaging systems. CuONPs have been shown to have antibacterial activity against Gram-positive (G +) & Gram-negative (G –) bacteria in practically all biopackaging systems. However, in some polymeric systems, such as CuO & Chitosan nanofibers, a synergistic effect is observed (Almasi et al., 2018).

It is well known that the antimicrobial activity of MOs such as CuO is governed by their structure, size, morphology, surface area and oxidation states variation. Furthermore, doping/ coupling CuO with other active materials (such as MOs and M) may have an impact on the ultimate properties of packaging films. Cu/CuONPs, CuONPs/AgNPs, and other nanohybrid systems can provide improved optical, Oxygen/water barrier properties and antimicrobial, with biocidal activity against G+ve /–ve bacteria (Jafarzadeh et al., 2020).

Copper is attractive for active packaging because it shows antimicrobial properties that are broad-spectrum. Copper ions work by readily receiving and giving electrons, demonstrating a strong redox potential and the ability to destroy microbial cells components, killing them (Nan et al., 2008: Garcia et al., 2018). The incorporation of nano–CuO into LDPE produced an antibacterial nanocomposite, which was evaluated for cheese packaging, and resulted in 4.21 log CFU/g of coliforms reduction after 30 days of refrigerated storage (Beigmohammadi et al., 2016). Nano-CuO/ZnO was added to chitosan, which resulted in nanocomposite films with improved antimicrobial activity, decrease in moisture permeability, improved quality attributes of guava fruits, extending its shelf life by 1 week (Kalia et al., 2021). In addition, CuO incorporated into Chitosan killed more than 99% bacteria (Raghavendra et al., 2017). Nanocomposite was prepared by incorporating nano-CuO into Agar resulting in increased UV-light barrier property and potent antibacterial activity against E. coli and L. monocytogenes (Roy and Rhim 2019).

Magnesium Oxide (MgO)

Magnesium oxide (MgO) is a colorless naturally occurring, crystalline mineral having a high melting point and because of its large-scale production, it is used in a variety of industries. MgO is a material with a poor electrical conductivity and a high thermal conductivity. In recent years, research has focused on MgO's excellent antimicrobial activity and high stability when compared to organic antimicrobial agents. Its application as a multifunctional solid material is due to this combination of properties. Numerous construction materials in food packaging has been replaced with MgONP-based packaging in the United States and Europe [Food additive approved by the European Union (E number 530)], because it is thermally stable, recyclable, gas impermeable and flexible, and possessing antimicrobial activity as well as Hydrothermal, laser ablation, microemulsion, microwave-assisted, sol–gel, ultrasound-assisted and wet chemical reaction processes can be used to synthesize MgONPs (Mirtalebi et al., 2019).

MgONPs are incorporated into biodegradable polymers including polylactic acid (PLA), Polycaprolactone (PCL), and polyhydroxybutyrate (PHB) to produce nanocomposite with enhanced characteristics and antimicrobial activities. Pure PLA degrades gradually in the environment to CO2, CH_4 and H_2O. Industrial-scale nanocomposites with enhanced barrier, mechanical, thermal, optical and antibacterial/microbial properties can be formed by incorporating MgONPs into the aforementioned polymers. For instance, Swaroop and Shukla (2019) found that nanocomposite films of PLA/MgONP comprising 2 wt.% MgO enhanced plasticity and tensile strength by more than 146 % and 22 %, respectively. When compared with the sample containing 1wt.% MgONPs, the oxygen and water vapor barrier properties of this nanocomposite was also improved by about 65 % and 57 %, respectively. They also mentioned the film's antimicrobial properties, stating that after 24 hours of treatment in the presence of 1wt.% MgONPs, E. coli bacteria (about 44 %) were killed. Zhang et al., (2020) demonstrated that ternary nanocomposites prepared with the combination of Poly(butylene adipate-co-terephthalate) (PBAT) and MgO/Ag showed improved mechanical and barrier properties, with composite films having 3 wt% NPs content exhibiting the optimal properties. In addition, the ternary composite film inhibition rates against E. coli and S. aureus exceed 90 %. Nanocomposites prepared by combining Starch-albumin with MgO increases film thickness, decreased water vapor permeability, increased antioxidant activity, antimicrobial properties (Hosseini et al., 2021). Silva et al., (2017) observed that packaging made of chitosan and nano-MgO exhibited remarkable improvement in thermal stability, UV shielding and moisture barrier properties. Wang et al., (2020a) reported improved thermal stability, better UV shielding performance, as well as water-insolubility compared to pure CMCS, exhibited excellent antimicrobial activity against L. monocytogenes and S. baltica by combining Chitosan-CMCS with nano-MgO.

Silica (SiO_2)

Silicon is a metalloid that is found in the forms of silica (SiO_2) and silicates and is the most plentiful solid element on Earth. Nonstick coatings for jars, bottles and bags are made with SiO_2 as the main raw ingredient in numerous food industries (Bumbudsanpharoke et al., 2015; Liu et al., 2018). SiO_2 is a promoter (a compound that boosts catalytic activity), like other MO. SiO_2–NPs' activities are influenced by factors such as high surface area, low thermal conductivity, average particle size, low level of toxicity, supreme insulation, biocompatibility and stability. Silica can be used in food packaging films as an antimicrobial agent or as a strengthening agent once it has been modified with organic matter. Examples of modified SiO_2 are Tetraethoxysilane, 3-isocyanato-propyltriethoxysilane, and aminopropyl-trietoxysilane. When compared to pure polymers, polymer/SiO_2 nanocomposites have been demonstrated to have improved water and gas barrier properties, in addition to better mechanical strength. Incorporating nano–SiO_2 into plastics increased the mechanical strength and thermal stability of the plastics (Hasan et al., 2006). In comparison with the pure polymer, EVOH-SiO_2 nanocomposites containing 5 wt.% nano-SiO_2 had significantly improved water plus gas barrier properties in addition to improved mechanical strength (Liu et al., 2010). Modifying nano-SiO_2 (0.09 wt.%) with ethylene/vinyl acetate (EVA) and blending with polypropylene, produced nanocomposites with an increase in tensile strength and a decrease in gas permeability, as well as adsorbed a smaller amount of ink solvent as compared to the pure polymer, which could be beneficial for laminated food packaging because for printing, the layer of polypropylene is used (Li et al., 2016).

Another way to make SiO_2-based nanocomposites is to coat the polymers. The sol–gel procedure was used to make a polylactic acid–based coating with nano–SiO_2 utilizing tetraethoxysilane as a precursor and 3-isocyanato-propyl-triethoxysilane as a coupling agent, respectively, the coating can be applied to PLA films to produce biodegradable packaging with gas barrier properties that are up to 70% better than pure PLA films while yet being transparent (Bang & Kim, 2012). Zhao et al., (2020) revealed that packaging made of nano-SiOx-coated PLA decreased oxygen and water vapor transmission rates by 48.45% and 28.53%, respectively, approximately 100 nm SiOx layer could prevent diethylene glycol dibenzoate from migrating and diffusing into food simulants.

There are only a few investigations on SiO_2 nanocomposite applications. None-theless, Luo et al., (2015) reported on the use of LDPE–SiO2 packaging for chilled white shrimp, which increased shelf life and kept the shrimp fresh for eight days, implying that this nanomaterial could be combined with polymers to manufacture “active packaging” with antimicrobial as well as enzyme-inhibitory properties.

According to Donglu et al., (2016), combining SiO_2/TiO_2/Ag nanoparticles with LDPE produced a multifunctional nano-composite capable of regulating the levels carbon dioxide and oxygen, scavenging ethylene and inhibiting microbial growth, thereby prolonging the packaged mushroom’s shelf life, implying that using a combination of nanomaterials to develop nanopackaging can be a good strategy (Garcia et al., 2018).

SiO_2–nanoparticles are utilized separately as a reinforcing agent in the most widely published studies in food packaging, and they have antimicrobial properties when combined with organic and inorganic materials. For instance, ethylene scavengers, carbon dioxide and oxygen regulators plus microbial growth inhibitors have been described using a mixture of SiO_2/Ag, SiO_2/TiO_2/Ag, SiO_2–Al_2O_3, $CoFe_2O_4$ /SiO_2/Ag, and silica–carbon–silver (SiO_2 /C/Ag) nanosystems mixed into polymer matrices (Jafarzadeh et al., 2020). Thus combining nanomaterials with SiO2 can be an effective technique for developing nanopackaging. The oxygen/water barrier effect of SiO_2 nanoparticles incorporated into polymer matrix is depicted in Figure 1.

Regarding the oxygen barrier performance, the addition of SiO_2-NPs reduces the oxygen permeability for two main reasons: i) according to the “tortuosity path” concept, the impermeable filler represents a physical barrier to the permeant transfer across the material's thickness, thus reducing the diffusion coefficient (Silvestre et al., 2011); ii) according to the “interface effect” concept, fillers with high surface area promote local changes in the polymer matrix properties at the interfacial regions (Duncan, 2011). In particular, the higher the extent of the cooperative forces at the filler/polymer interface, the lower the segmental mobility of the molecules (Rovera et al., 2020).

Titanium Dioxide (TiO_2)

Owing to its bright white coloration, titanium dioxide (TiO_2) is extensively used as a pigment in a variety of products, such as Foods, Nutraceuticals, Supplements, as well as Cosmetics (Weir et al., 2012; Guseva et al., 2020). TiO2 nanoparticles are promising candidates for reinforcing polymer matrixes in food packaging applications because of their low cost, nontoxicity, and biocompatibility (Lotfi et al., 2019; Mohammadi et al., 2015; Segura et al., 2018; Venkatesan & Rajeswari, 2017; Xie & Hung, 2018; Cao et al., 2020). The synthesis of TiO2 nanoparticles can be done in three different forms: anatase, brookite or rutile, each with distinct characteristics and reactivities based on their band gap (Llorens et al., 2012). To synthesize these nanoparticles, the sol–gel methodology is commonly utilized (Ibrahim & Sreekantan, 2011; Bodaghi et al., 2013). Carré et al., (2014) and Dalrymple et al., (2010) revealed that TiO2 nanoparticles had photocatalytic antibacterial characteristics, generating reactive oxygen species (ROS) and killing bacteria by peroxiding lipids in the cell membrane and oxidizing proteins and DNA after being exposed to UV light. The anatase form, in particular, has low toxicity, high photocatalytic efficiency and can become active after exposure to near-UV light; it is inexpensive as well (Carré et al., 2014). The highest antimicrobial effect was achieved when exposed to UV-A light (Chorianopoulos et al., 2011).

Because of its photocatalytic activity, TiO_2 could be used to disinfect food contact surfaces and decontaminate water (Llorens et al., 2012). Furthermore, this property could be beneficial in the development of antimicrobial active packaging (Huang et al., 2018). Various polymeric matrices, including conventional plastics and biopolymers, can be used to produce such packaging. Xie and Hung (2018) prepared a UV-A activated TiO_2 embedded biodegradable polymer films. They found that 2 hours of UV-A light illumination at a light intensity of 1.30 ± 0.15 mW/cm2 reduced bactericidal activity by 1.69 log CFU/mL. Nanocomposites produced from Guar gum-TiO_2 showed UV-light and oxygen barrier properties, antimicrobial activity against *L. monocytogenes, Salmonella, enterica sv, typhimurium* (Arfat et al., 2017). Salarbashi et al., (2018a) also confirmed that soybean polysaccharides-TiO_2 film displayed antimicrobial activity against S. aureus, Pseudomonas, aeruginosa, E. coli. Wheat gluten/nanocellulose-TiO_2 film had antimicrobial activity against Saccharomyces cerevisiae, E. coli, and S. aureus (El-Wakil et al., 2015). Pectin-TiO_2 nanocomposites exhibited antimicrobial activity against E. coli (Nešić et al., 2018). In addition, soybean polysaccharides incorporated with TiO2 nanoparticles showed decrease in moisture content and water-vapor permeability, increased tensile strength and antimicrobial activity (Salarbashi et al., 2018b). TiO_2 nanoparticles incorporated into edible films of whey protein had antimicrobial effects (AlizadehSani et al., 2017), and good moisture barrier and mechanical properties (Zhou et al., 2009). 2–5 wt% TiO_2 nanoparticles mixed with ethylene-vinyl alcohol (EVOH) produced self-sterilizing nanocomposites that were effective against both Gram-positive as well as Gram-negative bacteria (Cerrada et al., 2008).

Some of the developed TiO_2 nanocomposites have been tested as food packaging. In in vitro trials and as real packaging for chopped lettuce, E. coli growth was inhibited using oriented–polypropylene (PP) films coated with TiO_2 nanoparticles, following exposure to black light (Chawengkijwanich & Hayata, 2008). LDPE films containing TiO_2 nanoparticles inactivated yeasts and Pseudomonas spp., in vitro, as well as when used as packaging for pears, and the impact was increased by UV-A light exposure (Bodaghi et al., 2013). A similar result was demonstrated for chitosan-TiO_2 films, which exhibited antimicrobial activity against *Escherichia coli, Staphylococcus aureus, Candida albicans,* and *Aspergillus niger* with 100% sterilization in 12 h, it also possessed UV-blocking properties, decreasing light transmittance in visible light region of the composite film and extended the shelf life of red grapes (Zhang et al., 2017). Nanocomposites with improved oxygen barrier properties were produced when TiO_2 (4 wt%) was integrated into LDPE films, and its usage as packaging reduced the rancidity incidence in almonds by 78 percent in comparison to the control (Nasiri et al., 2012). According to Li et al., (2017a) PLA-TiO2 nanocomposites showed antimicrobial activity, cottage cheese freshness preservation, and a 25-day extension of the cheese shelf life. Other foods like short-ripened cheese, strawberries, shrimp and mangoes have been demonstrated to minimize deterioration and retain their quality using similar nanocomposite films (Gumiero et al., 2013; Li et al., 2017; Luo et al., 2015; Chi et al., 2019). Furthermore, TiO2's photocatalytic activity exhibits ethylene-removing properties (Bodaghi et al., 2015; Wang et al., 2010; Bohmer-Maas et al., 2020), Li et al., (2017b), which could be effective in delaying fruit ripening. Incorporating TiO_2 into polymers also produces nanocomposites that blocks oxygen and UV-light (Guo et al., 2014; Krehula et al., 2017; Achachlouei and Zahedi, 2018; Ahmadi et al., 2019; Goudarzi et al., 2017; Yousefi et al., 2019). UV light blocking is desirable because it protects packaged foods while also delaying the ageing and degradation of nanocomposites. Díaz-Visurraga et al., (2010) combined TiO_2 nanotubes (0.05 % to 0.1 % w/v) into chitosan to develop UV-blocking nanocomposite films that were effective against *Salmonella enterica, Escherichia coli*. and *Staphylococcus aureus*.

Active packaging with enhanced properties can be developed, when TiO_2 is used in a synergistic way, for instance, HDPE-TiO2-marigold extract nano-composite was reported to maintain soybean oil stability via the antioxidant effect of the carotenoid component; as carotenoids degrade easily when exposed to light, their effectiveness was preserved by the UV-blocking effect of TiO_2 (Colín-Chávez et al., 2014). A similar finding was reported by Vejdan et al., (2017) for gelatin-agar-TiO_2 films, which delayed fish oil oxidation. According to a study, TiO_2-based nanocomposites could prevent plastic additives such as ethylene glycol from migrating into food from packaging, besides improved mechanical as well as antibacterial properties (Farhoodi et al., 2017).

Zinc Oxide (ZnO)

ZnO is a white powder that is extensively used in sunscreens as well as in the Food industry (Espitia et al., 2012). Due to their inexpensive cost, low toxicity, in addition to UV-blocking properties, ZnO nanoparticles have an advantage over Ag nanoparticles and they can also be utilized as an antimicrobial material (Chaudhry et al., 2008; Llorens et al., 2012). Wurtzite, zinc blende, and rock salt are the three crystal structures of ZnO. In comparison to the other structures, the wurtzite structure is more thermodynamically stable. Various physicochemical and biological methods are used to synthesize ZnONPs (Kumar et al., 2019). Mechanochemical processing (MCP) and physical vapour synthesis (PVS) are used in industrial production of ZnONPs, while wet chemistry methods such as coprecipitation (Ubani & Ibrahim, 2019), microwave (Salah et al., 2019), thermal decomposition (Alp et al., 2018), sol–gel (Delice et al., 2019; Khan et al., 2016), and hydrothermal (Kumaresan et al., 2017) methods can also be used. Nano-ZnO, like TiO_2, has an antimicrobial effect due to a photocatalytic reaction, but unlike TiO_2, it also produces Zn^{2+} ions, which damage and kill bacterial cells (Bumbudsanpharoke & Ko, 2015), and UV exposure does not trigger the antimicrobial activity (Espitia et al., 2012).

ZnO, like TiO_2, has been integrated into or coated onto a variety of synthetic and biological polymers to produce antimicrobial nanocomposites. The growth of both *Staphylococcus aureus* and *Escherichia coli* were inhibited using polyvinyl chloride (PVC) films coated with ZnO; however they were more effective against S. aureus (Li et al., 2009). Furthermore, nanocomposites containing nano-ZnO have better mechanical and oxygen barrier properties (Lepot et al., 2011). With the incorporation of nano-ZnO (0.5 wt%) into HDPE and modification by the coupling agent KH550, nanocomposite films with increased mechanical, barrier and UV-blocking properties, in addition to antimicrobial activity, especially against Staphylococcus aureus was produced (Li & Li, 2010). According to Ghozali et al., (2020) LDPE-ZnO nanocomposites was effective by showing a widest inhibition zone against E. coli. In addition, a study (Applerot et al., 2009) reported the production of antimicrobial glass via coating it with ZnO nanoparticles, which resulted in 89 % decrease in E. coli counts.

Biodegradable packaging development is appealing since it would result in generation of less waste as well as environmental pollution while lowering reliance on oil; nevertheless, bioplastics have high water permeability, which is a disadvantage. As a result, nanocomposites with enhanced properties have been produced with the incorporation of nano-ZnO into bioplastics. By incorporation of ZnO up to 3% into the polymer, multifunctional PLA-ZnO nanocomposites were produced, yielding a biodegradable packaging film with antibacterial effects against both G +/– strains and improved water barrier properties, UV light blocking ability (Pantani et al., 2013). Coating of ZnO (0.5 wt%) to PLA coating layer was effective in inactivating *E. coli* and *S. aureus*, with *E. coli* been more susceptible, showing 3.14 log reduction (Zhang et al., 2017). The effectiveness of PLA/ZnO nanocomposites showed increase in the hydrophobicity, good UV-light barrier properties and excellent antibacterial activity against *S. aureus* and *E. coli* (Kim et al., 2019). Poly (butylene adipate-co-terephthalate) (PBAT) combined with nano-ZnO yielded another biodegradable nano-composites with improved mechanical, thermal as well as antimicrobial properties (Venkatesan & Rajeswari, 2017). Nanocomposites prepared by combining poly(ε-caprolactone) (PCL) with nano-ZnO, (PCL/ZnO5%) had optimal antibacterial activity (Pina et al., 2020). Qiu et al., (2019) in their work, documented that nanocomposites developed by incorporating nano-ZnO into chitosan significantly improve mechanical and antibacterial properties. Nanocomposites produced with the combination of nano-ZnO with PBAT, showed high antibacterial activity against S. aureus, P. aeruginosa and B. subtilis strains (Seray et al., 2020). Starch-kefiran combined with ZnO yielded nanocomposites that exhibited increased Tensile strength; decreased in WVP by about 16%, decrease in moisture absorption of the films (Shahabi-Ghahfarrokhi and Babaei-Ghazvini 2019). Nanocomposites of Zein–ZnO increased the hydrophobicity and reduced up to 3 times, the amount of water uptake of the composite films compared to pure zein, UV-blocking properties, and antibacterial activity (Schmitz et al., 2020).

Food packaging tests have been carried out on some of the produced ZnO nanocomposites. Emamifar et al., (2011) observed that LDPE-ZnO-Ag nano-composites were successful at inhibiting *Lactobacillus plantarum* growth in orange juice, while Li et al., (2011) discovered that nano-ZnO-coated PVC packaging reduced decay and prevented bacteria and fungi in cut apples. ZnO nanoparticles incorporated into a poly-lactic acid (PLA) matrix resulted in nanocomposite films that inhibits microbial growth and preserved the organoleptic qualities of fresh-cut apple at 4° C, extending the shelf life by 2 weeks (Li et al., 2017). El-Sayed et al., (2020) demonstrated that coating Ras cheese with chitosan/guar gum/roselle extract-ZnO protects their surface for around three months from yeasts, molds and other bacteria growth as compared to uncoated cheese. Heydari-Majd et al., (2019) in their work, documented the effectiveness of nanocomposites of PLA-ZnO-(*Zataria multiflora* Boiss. essential oil and *Menthe piperita* essential oil) for enhancing antibacterial and antioxidant activities and extends shelf life of fillets by 8 days. Chitosan– cellulose acetate phthalate film loaded with 5% (w/w) nano-ZnO exhibited optimal tensile strength, significantly lower the water vapour and oxygen transmission rates, excellent UV shielding ability, antimicrobial properties and extended the shelf life of black grape fruits up to 9 days (Indumathi et al., 2019). Furthermore, *Salmonella* in egg whites was inactivated after 3 weeks of storage in glass jars coated with a mixture of PLA, allyl isothiocyanate, ZnO nanoparticles and nisin (Jin & Gurtler, 2011). In addition, in a study, ZnO nanocomposites were found to be effective against yeasts and molds that cause bread spoilage (Noshirvani et al., 2017). Negahdari et al., (2021) revealed the effectiveness of PLA-(added Origanum majorana essential Oil)-ZnO nanocomposites, (1.5% O. majorana EO and 1% ZnO NPs) extended meat shelf life and lead to the most favorable sensory properties of ground meat. Chitosan-ZnO nanocomposites exhibited excellent antimicrobial activity in raw meat by complete inhibition of microbial growth on the sixth day of storage at 4 °C (Rahman et al., 2017). Coating of pomegranate with the combination of Nano-ZnO + CMC extended its storage life by decreasing total yeast + mold during 12 days of storage while total mesophilic bacteria decreased during 6 days of storage (Saba and Amini 2017). Polyurethane/chitosan with nano-ZnO composite films showed enhanced antibacterial properties, barrier properties, and extended the shelf life of carrot pieces up to 9 days (Sarojini et al., 2019). The chemical composition, surface functional chemical groups, crystalline structure, specific surface area and morphology of ZnONPs can all be used to their advantage. ZnONPs possess a broad spectrum of antimicrobial activity and a low proclivity to induce resistance (Reyes-Torres et al., 2019). Although the specific mechanism of most NP reactions/interactions is still not known, the antimicrobial activity of nanoparticles can be predicted based on electrostatic interaction, the release of antimicrobial ions, and reactive oxygen species (ROS) formation (Figure 2).

MECHANICAL PROPERTIES

Deformability, TS, elongation at break (EAB), and elastic modulus (EM) are important mechanical characteristics of food packaging since these materials must maintain their integrity during storage, transport, and handling. The maximum stress that the film can withstand while being stretched or pulled before failing or breaking is known as TS and EM, which indicates the flexibility and intrinsic stiffness of the films, respectively (Jafarzadeh, 2017). The mechanical properties of biopolymer films depend both on their composition and on the environmental conditions. For example, the addition of plasticizers causes a higher mobility of polymer chains, which leads to expanded elongation and diminished TS of the plasticized films. Embedding of different additives, such as cross-linking materials or lipids, can improve film strength and extensibility (Vieira *et al.,* 2011). Moreover, humidity and moisture of the environment influences the mechanical properties of polymer/biopolymer films. For example, hydrophilic films absorb humidity more promptly at higher moisture levels, consequently enhancing the plasticizing impact of water, which subsequently decreases the TS and increases the extendibility of the films. In addition, the contact between polymer/biopolymer packaging materials and packaged product can likewise influence the functioning of packaging films.

In recent years, the incorporation of NPs has turned into a well-known way to improve the properties of different films, since the utilization of NPs typically gives them improved mechanical properties (Cho & Rhee, 2002). This result is due to the increased surface interaction between the matrix and NPs with a high surface area, as well as the hydrogen bond formation between them. EAB has a reverse relation to TS in most cases, and YM is directly related to TS. Furthermore, the mechanical properties of films are closely associated with the density and distribution of the intra- and intermolecular interactions between polymer chains in the film matrix.

OPTICAL PROPERTIES

The optical properties (color, transparency, and UV transmission/absorbance) of packaging materials are extremely significant since maintaining food quality depends greatly on their protection against UV and light. Also, to prevent lipid oxidation, retain food nutrients, and prevent the discoloration of food, the packaging must protect the content against UV light. Furthermore, the consumers' acceptance of food packaging is influenced by the color of the packaging, and it is one of the most important properties of films which could affect their application (Jafarzadeh *et al.,* 2017; Jafarzadeh and Jafari, 2020).

The apparent color properties of neat semolina films showed that they are totally transparent and colorless; however, the addition of ZnO-nr significantly decreased the L* and a* values, meaning an increased number of NPs, resulting in reduced transmissions from films (Jafarzadeh *et al.,* 2017; Jafarzadeh and Jafari, 2020). Arfat *et al.,* (2017) reported that the addition of Ag-Cu NPs into fish gelatin films significantly influenced their color by reducing the lightness (L*), while the a* and b* values significantly increased. These results were consistent with those for agar nanocomposite films incorporated with nano-silver, as reported by Rhim *et al.,* (2013). Moreover, the control gelatin film remained highly transparent while the addition of nano-clay was found to decrease the transparency of films. It has been reported that nanocomposite films with nano-silver had a brown surface color with reduced transparency (Kanmani and Rhim 2014b). Zolfi *et al.,* (2014) also reported that the transparency of nanocomposite kefiran-WPI films significantly decreased with the addition of TiO_2, and this decrease was dependent on the level of NPs.

The light transmission and absorption spectra of nanocomposite films is determined by UV-vis spectroscopy. Three zones have been identified for the UV region, namely, UVA (320–400 nm), UVB (280–320 nm), and UVC (180–280 nm). The nanocomposite films incorporating ZnOnr showed a clear absorption peak, but with the addition of ZnO-nr displayed greater absorption peaks at UVA (Jafarzadeh *et al.*, 2017). Also, Kanmani and Rhim (2014a) and Rouhi *et al.*, (2013) reported a lower UV transmission by the incorporation of Ag-clay and ZnO-nr into gelatin films.

THERMAL PROPERTIES

It has been demonstrated that thermal analysis methods can be used to define suitable processing conditions, applications, and polymer chain structures. The thermal profiles of polymers can be investigated by thermogravimetric analysis (TGA), derivative thermogravimetry (DTG), differential thermal analysis (DTA), and differential scanning calorimetry (DSC) (Gabbott, 2008). TGA describes the relation between the weight change and temperature. The amount of mass decreased versus temperature, or time, in a controlled atmosphere can provide information about thermal and oxidative stabilities of materials. Based on TGA thermogram, the composition of materials can also be identified. Using TGA, the mass loss/mass gain due to decomposition, oxidation, or loss of volatiles can be examined. It is a useful technique for measuring the polymeric materials like thermoplastics, films, fibers, etc. In industries, TGA measurements can be used to select materials for end-use applications, either by product performance or/and product quality.

DSC is a thermal technique to obtain a wealth of information about materials, including polymers, and organic–inorganic composites/hybrids. The energy changes during continuous heating and cooling can be obtained from DSC measurements. This enables the scientists to find the transition temperatures, like glass transition temperature (Tg), melting temperature (Tm), and crystallization temperature (Tc). In addition, this quantitative thermal analyzer can provide detailed information regarding the degree of crystallinity. Melting is an endothermic process, that is, the sample absorbs energy. Integrating the peak area gives the heat of fusion (ΔH_f). Crystallization of the polymer, which is a process of partial alignment of molecular chains, occurs upon cooling, mechanical stretching, or solvent evaporation. Crystallization can affect optomechanical, thermomechanical, and chemical properties of the polymers (Billmeyer, 2007).

THE EFFECT OF OXIDE-BASED NANOPARTICLES ON DIFFERENT PROPERTIES OF FOOD PACKAGING MATERIALS

In order to protect the foods from moisture, oxygen, pathogenic micro-organisms, dust, light, and a variety of other harmful or dangerous materials, the packaging must be inert, cheap to produce, light weight, easy to dispose of or reuse, able to withstand extreme conditions all through processing or filling, impervious to a variety of environmental storage and shipping conditions as well as resistant to physical and mechanical abuse. A substantial number of commercial food packaging materials are made of non-biodegradable materials, which pollute the environment and utilize petroleum derivatives in their manufacture. Bio-nanocomposites for food packaging are not only safer and extend the shelf life of food, but they are also more environmentally friendly because they eliminate the need for plastics as packaging materials. However, present biodegradable films have weak mechanical and barrier properties, which must be enhanced before they can completely replace traditional packaging and, as a result, contribute to global waste reduction. As a result, the primary objective for nano-packaging is to extend the shelf life of food packaging by improving barrier properties to reduce transfer of gas and moisture, UV light exposure, and enhance the mechanical, thermal, and antimicrobial properties (Jafarzadeh *et al.,* 2020). Figure 3 depicts the most important requirements for food packaging materials.

OXIDE NANOPARTICLE MIGRATION FROM PACKAGING

When nanomaterials are utilized in foods as additives, they are characterized as free; when they are incorporated into food contact materials, they are classified as embedded. This distinction is important when nanomaterials' oral exposure and toxicological risk is been estimated, because in the latter instance, nanomaterials must migrate to the packaging’s surface, and then interact with the food (EASAC & JRC, 2011; Störmer *et al.,* 2017). Diffusion, dissolution, and abrasion of the packaging surface can promote nanomaterial migration into food, which is a matter for concern because it may have severe health repercussions (Wyser *et al.,* 2016). Nanoparticles have a large surface area-to-volume ratio due to their small size, and therefore may have different physicochemical properties compared to bulk-sized materials (Chaudhry *et al.,* 2008). Moreover, their charge, composition, and surface morphology, as well as the chemistry of the nanomaterial itself, may affect their toxicology (Wyser *et al.,* 2016). As a result, the toxicological characteristics of nanomaterials must be determined, which necessitates physicochemical characterization in addition to *in vitro* plus *in vivo* experiments (Oberdörster *et al.,* 2005). It's also crucial to figure out whether and how much nanoparticles in packaging migrate when they come into touch with food, as well as the impact of consuming them on the gastrointestinal system and other organs. Nonetheless, the existing data is inadequate, and further investigation is urgently required in this area.

Furthermore, nanomaterials release from packaging is a source of worry because these materials may contaminate the environment and be consumed by humans in turn. The migration of nanomaterial into non-food matrices is complicated, with some materials forming aggregates and others dissolving into ions or binding with organic matter. However, little research has been done on the environmental destiny of nanomaterials from packaging (Karimi *et al.,* 2018). A study (Part *et al.,* 2018) examined the likely destiny of nanomaterials (mostly silver & titanium dioxide) in garbage, indicating that while waste degrading mechanisms are unaffected, microbial communities may be altered. However, there is a significant knowledge deficit in this area.

The majority of research on nanomaterial migration from food packaging to foods has focused on silver nanoparticles (Bumbudsanpharoke & Ko, 2015; Störmer *et al.,* 2017), with only a few investigations available on oxide nanoparticles migration. The “European Commission (EC)” has enacted rules that describe the criteria under which migration tests have to be carried out (EC, 2007, 2011b). Depending on the type of food product in contact with the packaging, food simulants such as water, ethanol (10%–50% v/v), acetic acid (3% v/v), and vegetable oil can be used. As a result, such simulants have been used in most migration studies, and research on nanoparticles migration into real foods is scarce.

Even though the amounts of nanomaterial migrated have been reported to be small (Bumbudsanpharoke and Ko, 2015; Echegoyen and Nern, 2013; Farhoodi *et al.,* 2014), the migration rate of “nanosilver” and “nanoclay” components (Al and Si) increases with increasing temperature and time.

Migration studies dealing with oxide nanoparticles have been documented by some researchers. Lin *et al.,* (2014) investigated the influence of 30 and 100 nm particle size on Ti migration into food simulants (acetic acid and ethanol), from LDPE-TiO_2 nanocomposites. As a result, they noticed that raising the exposure temperature caused the nanomaterial to migrate from the packaging, into the acetic acid in particular; after exposure of 8 hours to 3 percent acetic acid at 100 °C, the maximum amount of migrated Ti was 12.1 g/ kg. In one more investigation (Lian *et al.,* 2016), high hydrostatic pressure was used to compact and stabilize polyvinyl alcohol-chitosan-TiO_2 nanocomposites; migration tests showed that only a small amount of Ti ($3.87\times10^{-3‰}$ for film treated at 200 MPa) migrated in olive oil after eleven hours, but none in acetic acid, water or ethanol. The toxicity of nano-TiO_2 has been found to be minimal; to demonstrate tissue damage and bioaccumulation in mice, a substantial dose of 5 g/kg administered orally at once was required (Wang *et al.,* 2007).

Alternate to petroleum-based plastics, bioplastics have been promoted as a better-for-the-environment; nevertheless, they have drawbacks like poor gas and water barrier properties. It is therefore advantageous to add nanoparticles to bioplastics to enhance mechanical as well as barrier properties; however nanomaterial migration into packaged foods is still a concern. The quantity of Ti migrated from PLA-based nanocomposites was examined (Li *et al.,* 2018; Li *et al.,* 2017), suggesting that the quantity of nanomaterial migrated was low and within the safe range, as in synthetic composites. Furthermore, Li *et al.,* (2018) compared the amount of Titanium that migrated from Titanium dioxide into a liquid simulant as well as the real product (cheese), and found that the amount of Titanium migrated to the food was significantly lower than that in the simulant, implying that food simulants migration testing may overestimate the real exposure, especially when dealing with solid foods.

The migration of silver and zinc from LDPE-Ag-ZnO nanocomposites into orange juice was investigated by Emamifar *et al.,* (2011), after 112 days of refrigerated storage, they found that both migrated in low amounts of 0.15 and 0.68 g/L, respectively, remaining within the range regarded safe for consumers. Panea *et al.,* (2014) used an aqueous simulant to test the migration of silver and zinc from an LDPE-Ag-ZnO nanocomposite for packaging chicken for 10 days; only a minuscule quantity of silver (0.01 mg/kg) as well as zinc (2.44 mg/kg) migrated, both of which were below the European legislation established limit for zinc (5 mg/kg). It is worth noting that ZnO is GRAS for food applications and has a low toxicity in bulk form (Espitia *et al.,* 2012). Furthermore, the "European Food Safety Authority (EFSA)" stated that ZnO nanoparticles don't migrate from polyolefins and unplasticized polymers; consequently, the release of soluble ionic Zn is the primary concern when evaluating the safety of nano-ZnO in food contact materials (EC, 2016).

According to the European Food Safety Authority (EC, 2016), when incorporated into polymers, silanated synthetic amorphous SiO_2 nanoparticles don't migrate. Similarly, a research found that only (0.23 mg/kg) of CuO migrated from an LDPE-based nanocomposite (Beigmohammadi *et al.,* 2016), which is also lower than the "European legislation's" limit for Cu release from plastics (5 mg/kg), though it is worth noting that the nanoform hasn't been ratified (EC, 2011b). At 0.5 mg/dm^2 concentrations, equivalent to 0.9 mg/dm^2 Al_2O_3, aluminium has been observed to migrate from an Al_2O_3-coated polymer to acetic acid, implying that to prevent migration, an extra polymer layer on top of such coatings may be required (Vähä-Nissi *et al.,* 2015). Only nanoparticles with diameters less than 3.5 nm can migrate, according to a migration model based on TiN-LDPE nanocomposites, whereas most nanoparticles in the form of composites clumps with sizes greater than 100 nm in practice; as a result, nanoparticles integrated into plastics are immobilized, and there's virtually no migration risk into foods (Bott *et al.,* 2014; Störmer *et al.,* 2017).

To sum up, nanoparticles integrated into polymers tend to agglomerate and remain firmly ingrained in the polymeric matrix, and hence don't migrate, according to studies on nanoparticle migration. This seems to be especially true for non-ionizing materials such as Ti. Materials such as Zn, conversely, can still migrate as soluble ions, and while ion migration is essential for antibacterial actions, the migrated levels from active packaging must be kept below the safe limits. When nanoparticles immobilized on the surface of packaging come into contact with the microorganisms, antimicrobial effects can be produced as well. However, as long as the polymer is intact and the nanoparticles are adequately incorporated, there appears to be little chance of nanoparticle migration.

CONCLUSION AND FURTHER REMARKS

Nanocomposites made of metal oxides are shown to have improved mechanical, barrier, as well as antimicrobial properties. TiO_2 and ZnO in particular have been demonstrated to be effective against a variety of bacterial strains and, due to the former's non-ionization and latter's lower toxicity, could be used instead of Ag nanoparticles. The incorporation of metal oxide nanoparticles into biodegradable polymers allows for the production of nanocomposites with improved mechanical strength, water and oxygen barrier properties, and other additional functional properties which include antimicrobial and antioxidants activities and light-blocking properties, making them viable petroleum-based plastics replacements.

According to migration studies, only a small quantity of nanomaterial migrates from packaging into food simulants or foods, implying that consumer exposure to these nanomaterials and the health concerns associated with them are low. Furthermore, if nanoparticles are merely utilized to enhance "mechanical and barrier properties", the migration risk can further be reduced by incorporating a functional barrier between the food and the nanocomposite. Nonetheless, a dearth of human research, and a thorough characterization of the nanomaterials properties and toxicology raises doubt, and questions regarding nanotechnology's application persist.

Even though they have exhibited beneficial properties, most oxide-based nanocomposites are still in the development stage. One of the challenges that need to be addressed right now is the lack of validated methods for determining nanomaterial migration from packaging. Because regulators are taking a careful approach, this signifies that nanoparticles migration values should be below the detection boundaries in practice. Nonetheless, safety concerns will persist till satisfactory answers are found to these questions: "how much nanomaterial migrates into real foods from packaging"? "In what form is it ions or nanoparticles"? As well as "how toxic it is" thus stopping the mass commercialization of a technology that has proven enormous potential in the laboratory. As a result, academics, industry, and governments are being urged to prioritize this effort.

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Table 1. The properties and activities of various SiO_2/polymer nanocomposites

Nano-material	**Method of preparation**	**Polymer Matrix**	**Properties/activity**	**Ref.**
SiO_x	Plasma enhanced chemical vapor deposition (PECVD)	PLA (SiO_x/PLA-coated films).	Decreased oxygen and water vapor transmission rates by 48.45% and 28.53%, respectively, approximately 100 nm SiO_x layer could prevent diethylene glycol dibenzoate from migrating and diffusing into food simulants.	Zhao *et al.*, (2020)
SiO_2	Casting method	Whey protein isolate/pullulan	Tensile strength increased, improved barrier properties of the films, water vapor permeability decreased.	Hassannia-Kolaee *et al.*, (2016)
SiO_2	Extruded film	Polypropylene (surface treated with ethylene vinyl acetate)	The permeability of O_2 as well as water vapour was deceased by 28 percent and 23.8 percent, respectively. Tensile strength increased by up to 30%.	Li *et al.*, (2016)
SiO_2	Extruded film	LDPE (titanate crosslinking)	Improved gas barrier properties, resulting in 33% increase in packaged shrimp shelf life.	Luo, Xu, *et al.*, (2015)
SiO_2	Cast film	PLA	The permeability of O_2 and water vapour was lowered by 50%.	Bang & Kim, (2012)
SiO_2	Blown film	EVOH	Water permeability coefficient decreased by 54%; mechanical strength increased by 50% and heat resistance increased.	Liu *et al.*, (2010)
Ag + TiO_2 + SiO_2	Blown film	LDPE	The packing kept the mushrooms' nutrient content and quality for 2 weeks by lowering the respiration rate and scavenging ethylene.	Donglu *et al.*, (2016)

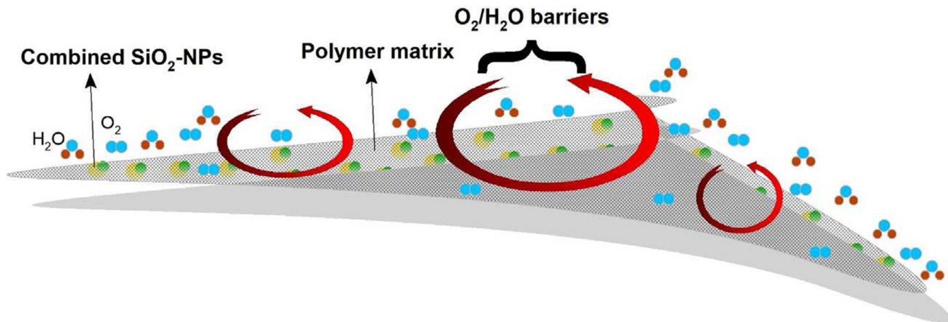

Figure 1. SiO_2 nanoparticles incorporated into polymer matrix provide an O_2/H_2O barrier effect. Adopted from Jafarzadeh *et al.,* (2020)

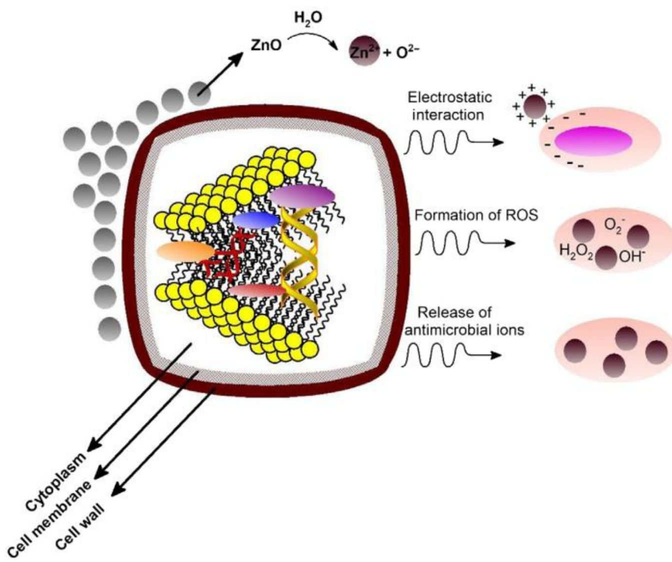

Figure 2. Mechanisms of ZnO nanoparticles antimicrobial activities (Jafarzadeh *et al.,* 2020)

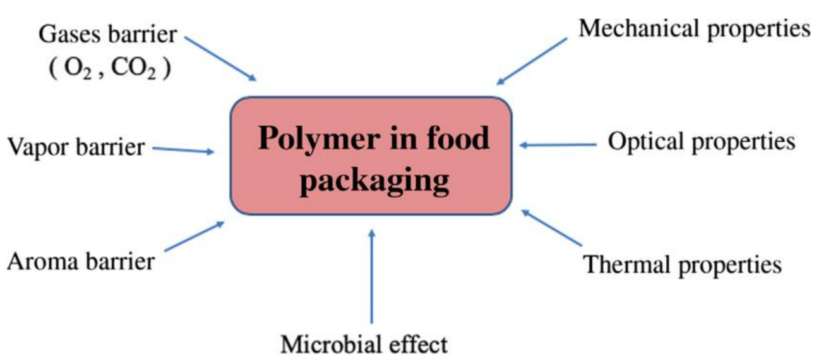

Figure 3. General requirements for polymers in food packaging (Jafarzadeh *et al.,* (2020).

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Physical and Sensory Characteristics of Food Bar Based on Beneng Taro (*Xanthosoma undipes* K. Koch) and Soy Protein Isolate

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ABSTRACT

Beneng taro, a local food plant in Banten Province, has enormous potential to be developed as an effort to diversify food, one of which is used as a raw material in the manufacture of food bars. Ingredients formulation and roasting temperature can affect the characteristics of the resulting food bar. Therefore, researchers consider it necessary to conduct research related to the physical and sensory characteristics of food bars based on beneng taro, mocaf and soy protein isolate as one of food diversification, as well as the best formulation and roasting temperature of the food bar. This study used a completely randomized split plot design with two factors, namely roasting temperature and ingredients formulation. The L* value, a* value, and texture were all significantly impacted by the roasting temperature, according to the results. Meanwhile, the L*, a*, and b* values were significantly impacted by the addition of beneng taro flour and soy protein isolate. The a* value, b* value, as well as the panelists' evaluation of the color, texture, and overall parameters are significantly impacted by the interaction between the two factors. The formulas for the chosen food bars contained 30% taro flour and 70% soy protein isolate, and they were baked at 140 °C (S_2R_2) with the following characteristics: texture 26,59 N; L* value 46,75; a* 14,99; b* 32,17; and the value of preference for color, taste, aroma, texture and overall is 2,23; 2,10; 2,23; 2,15; and 2,25.
Keywords: Beneng taro, food bar, soy protein isolate

INTRODUCTION

Beneng taro is a local food plant of Banten Province which is spread in the area around Juhut Village, Karang Tanjung District, Pandeglang Regency. Ningsih & Hermita (2016) states that beneng taro can generally grow on the edge of forests, river banks, swamps, and cliffs with humus. This taro grows in the tropical lowlands at an altitude of 250-700 m above sea level (asl) with sufficient rainfall ranging from 175-250 cm/year. Suhaendah et al. (2021) mentioned that previously taro plants grew wild and were considered a nuisance plant because their growth was very easy and fast, but now they have started to be cultivated because they have enormous potential.

According to Yuliani (2013) beneng taro tubers have a length of 1,2-1,5 m and a weight of 35-40 kg at the age of 2 years. Bulb circumference can reach about 45-55 cm. Beneng taro tubers are produced from trees that have a height of about 2-2,5 m with a leaf width of 1 meter. Therefore, Budiarto & Yunia (2017) stated that taro beneng has unique characteristics where the tubers are large and yellow (koneng) with stems which are the largest part that can be consumed. Based on the data obtained in the research of Muttakin et al. (2015) beneng taro has a protein content of 8,77%; carbohydrates 84,88%; starch 6.97%; ash 8.53%; fat 0.46% and 84.65% water.

Beneng taro has been used by local residents to make chips and as a food ingredient that is processed by steaming, boiling or frying. Considering its enormous potential, further processing is one of the alternatives needed to reduce the decline in the quality of beneng taro and as an effort to diversify products made from beneng taro. After the harvesting process is carried out, beneng taro can be processed into flour which is a semi-finished product. Later this flour can be used as raw material in the manufacture of products such as brownies, macaroni, dry noodles, cookies and food bars.

Food bar is a food product in the form of a rod and solid. According to Aini et al. (2018) food bar is one of the processed food products in solid, square shape and has a low a_w (water activity) value. Purwanti (2019) adds that food bars generally have an water activity value in the range of 0,65–0,85 and a water content of around 15%-30%. There are several ingredients commonly used in making food bars such as margarine, milk, eggs, sugar and so on. Food bars can be made using local raw materials such as purple sweet potato flour (Nurhayati et al., 2018; Fadhlan et al., 2021; Elisabet, 2018), red bean flour (Anandito et al., 2016) and soybean flour (Fanzurna and Mohamad, 2020).

According to Elisabet (2018) the process of making a food bar begins with the manufacture of various flours that will be used, followed by the process of mixing all the ingredients that will be used until the dough becomes smooth, then molding and baking in the oven. In making a food bar, there are several things that can affect the characteristics of the resulting food bar, including the formulation of raw materials and the roasting temperature at the time of making the food bar.

The results of research by Nurhayati et al. (2018) using purple sweet potato flour and ripe banana (*Musa paradisiaca* Formatypica) showed that the food bar with the best characteristics was a food bar with 30% purple sweet potato flour added and 70% banana agung with a roasting temperature of 120℃ for 40 minutes and 5 minutes. The physical characteristics of the food bar are texture value of 75,22 g/mm, color of L, a*, and b* respectively of 43,47; 7,67; and 18,25. Based on the research by Fanzurna and Mohamad (2020) which utilizes kepok banana peel flour and soybean flour as the basic ingredients for making food bars, it shows that the best food bar formulation is the addition of 10% kepok banana peel flour and 90% soy flour. In this formulation, the favorite values for color, aroma, texture, taste and aftertaste are 5,10; 5,03; 4,43; 5,63; and 5,17.

Muchtadi & Sugiyono (2014) explain that the roasting temperature has a very large influence on the manufacture of food bars. The research results of Rahman et al. (2011) showed that the optimal roasting time and temperature in the process of making banana-based food bars is when the roasting temperature is 120℃ for 40 minutes and the temperature is 140℃ for 5 minutes. The characteristics of the resulting food bar are 18,02% water content; 2,75% ash content; fat content 4,86%, protein content 8,74%; and 63,27% carbohydrate content. Based on the research of Rahmawati et al. (2020) a temperature of 130℃ for 20 minutes is the best temperature and roasting time in making sorghum flour-based food bars. Based on the results of the formulation and the best roasting temperature, the sorghum flour-based food bar has a hardness value of 1597.56 g force.

The results of Budiarto and Yunia researchs (2017) show that beneng taro flour processed by local residents has a water content of 6,10%; 6,11% ash content; fat content 0,39%; protein content 6,70%; and 80,70% carbohydrate content. Based on these results in terms of protein content, the protein contained in beneng taro flour is almost equivalent to the protein found in low protein wheat flour, so that beneng taro flour can be used as raw material for making food bars.

This study aimed to determine the physical and sensory characteristics of a food bar made from taro beneng flour, soy protein isolate and mocaf as well as the best formulation and roasting temperature of the food bar.

MATERIALS AND METHODS

Tools and Materials

The tools used in the food bar manufacturing process are digital scales type PT-238 (Kova Fabio), containers, trays, brushes, basins, knives, cutting boards, analytical balance type H7K (excellent), electric cabinet dryer model AST105E, disk mill machine type AGR-MD24 (Maksindo), blender type HR2223/60 (Philips), sieve size 100 mesh, spoon, plastic spatula, mixer type HM-620 (Miyako), pan size 24 cm × 24 cm, electric oven type KBO-190LW (Kirin) and a ruler. Meanwhile, the tools used in the analysis process are texture analyzer type LLYOD TA1 (Ametek), chromameter model colorFlex EZ (Hunterlab), tasting booth for sensory assessment, stationery, label paper, HVS paper and camera.

The ingredients used in making the food bar are taro beneng from Juhut-Pandeglang Village, Banten, mocaf (Mocafine), margarine (Forvita), refined sugar (Rose Brand), egg yolks, skim milk (Indoprima), salt (Mama Suka), soy protein isolate (Para Agibusiness), NaCl solution and water. Procedures and formulations in making food bars refers to the research of Nurhayati et al. (2018).

Method

The design of this study used a quantitative method with a completely randomized split plot design consisting of two factors and three replications. The first factor is the roasting temperature (S) as the main plot consisting of three levels, namely:

S_1 = 120℃
S_2 = 140℃
S_3 = 160℃

The second factor is the ingredients formulation as subplot which consists of four levels, namely:

R_1 = Soy protein isolate : Beneng taro flour = 80%:20%
R_2 = Soy protein isolate : Beneng taro flour = 70%:30%
R_3 = Soy protein isolate : Beneng taro flour = 60%:40%
R_4 = Soy protein isolate : Beneng taro flour = 50%:50%

This research was carried out through three stages, namely the production of beneng taro flour, the production of food bars, as well as physical and sensory analysis. Physical analysis carried out is color and texture analysis. Meanwhile, the sensory analysis carried out was the level of panelists' acceptance of the parameters of color, texture, aroma, taste and overall.

Production of Beneng Taro Flour (Putri et al., 2021)

The making of beneng taro flour begins with the cleaning process of fresh taro beneng, then the process of peeling the skin using a knife is carried out. Then the washing process was carried out again in running water with a ratio of 2:1 (v/w). Beneng taro that has been cleaned and peeled is reduced in size, soaked in 10% NaCl solution for 2 hours. After the soaking process with NaCl solution, the taro beneng was then washed with water. After that, the drying process is carried out at a temperature of 50 - 60°C with a cabinet dryer for 6-12 hours. Beneng taro that has been dried is then milled using a disk mill machine until it becomes coarse flour. Furthermore, the flour is mashed using a blender and sieved manually using a 100 mesh sieve.

Production of Food Bars (Nurhayati et al., 2018)

The food bar production stage was completed by weighing and mixing the taro beneng flour and soy protein isolate according to the treatment ratio that had been set. The preparation of auxiliary components, which include 10% mocaf, 30% liquid margarine, 25% refined sugar, 10% egg yolk, 35% skim milk, and 0,25% salt of the total flour used, begins the process of producing a food bar. The egg yolks, liquid margarine, and powdered sugar are then combined with a mixer on low speed for 5 minutes. Furthermore, skim milk, salt, and composite flour containing mocaf were added at a rate of up to 10%, as were beneng taro flour and soy protein isolate, according to the treatment that was gradually determined. Then, gradually add water to make a smooth dough. Manually mix again with a spatula until the dough is uniformly spread. Then do the molding of the dough into the pan 24 cm × 24 cm. The molded dough is then cooked for 40 minutes at 120°C in a preheated oven. The resting or chilling phase is then carried out at room temperature for 10 minutes before cutting. The half-cooked dough was then cut with a size of 6 × 3 × 2 cm^3. The roasting procedure was then repeated in the oven for 5 minutes at the proper temperature for the stated treatment. After that, the roasting process was carried out again in the oven at the appropriate temperature for the specified treatment for 5 minutes. After baking, the food bar is cooled by allowing it to cool at room temperature for a few minutes after being taken from the oven.

Color Analysis (Fardiaz, 1984 <u>in</u> Lumba et al., 2017)

The color of the resulting food bar was measured using a chromameter brand Hunterlab colorFlex EZ spectrophotometer. The color notation system used is the hunter system where L* (brightness), a* (± red - green), b* (± yellow - blue). Before the measurement process, the chromameter is calibrated first with the white standard contained in the tool. The color measurement process is carried out by placing the sample in the cuvette that has been provided and then pressing the start button to start the measurement process. The light source is on and the reflectance is measured, so that the L*, a* and b* values will be obtained from the sample with a range of 0 (black) to ± 100 (white).

Texture Analysis (Kulthe et al., 2014)

Texture analysis of food bars in this study used a texture analyzer (LLOYD TA1). The analysis process begins by placing the sample in the sample storage area available in the tool, then the sample is pressed with a constant pressing speed of 50 mm/minute, the maximum load cell force is 1 kg and compression is 75%. The maximum force required to break a food bar is its hardness value (Newtons (N)).

Sensory Analysis (Garnida, 2020)

Sensory analysis in this study was conducted to measure the level of preference or the level of panelists' acceptance of food bar products. Sensory analysis in this study used a hedonic method (scoring test) which included color, texture, aroma, taste and overalls which were tested by 40 untrained panelists with 7 rating scales, namely 1 =

Very Disliked, 2 = Disliked, 3 = Somewhat Disliked Liked, 4 = Neutral, 5 = Slightly Liked, 6 = Liked and 7 = Very Liked. The results of the sensory data obtained are then processed in the form of a spider web.

Data analysis

The data obtained were analyzed by test of variance with a significance level of 5%. Then interpreted according to the observed parameters to see the tendency of each parameter. If the data obtained is significantly different, a DMRT (Duncan Multiple Range Test) further test will be carried out to determine the significant difference between treatments at a significance level of 5%. Data analysis using SPSS (Statistical Package for the Social Sciences) software.

RESULTS AND DISCUSSION

Physical Characteristics

Food Bar Color

The parameter used to indicate how light and dark a material is is called L* or brightness. The brightness level (L*) has a value range from 0 to 100. The absolute black material has a value of 0, while the white material has a value of 100. The brighter the material, the greater the L* value (Winarno, 2004).

The results of analysis of variance (Table 1) showed that the treatment of variations in roasting temperature had a significant effect (P<0,05) and the treatment of variations in ingredient formulations had a very significant effect (P<0,01), while the interaction between the two factors did not show a significant effect (P>0,05) to the value of L*. The L* value of the food bar as a result of this study ranged from 42,84 (sample S_3R_4) to 49,18 (sample S_1R_1). This result is higher than the research result of Nurhayati et al. (2018) which has an L* value ranging from 43 – 44. This shows that the food bar based on beneng taro, mocaf and soy protein isolate has a higher brightness value than the food bar made with purple sweet potato flour and ripe banana agung.

The more addition of beneng taro flour will decrease the brightness value (L*). This is presumably because the color of beneng taro flour is brownish yellow so that it makes the color of the food bar more brown and there is a degradation of carotenoid pigments contained in beneng taro flour. The higher the roasting temperature, the lower the brightness value (L*). This is thought to be due to the Maillard reaction during the food bar roasting process.

According to Tamanna and Mahmood, (2015) when a material contains protein components and reducing sugars that are processed at high temperatures, a Maillard reaction will form. Seftiono and Intan (2020) added that a change in the color of a product can occur due to a decrease in color stability, as well as the Maillard reaction which produces melanoidin pigments as a form of brown color during the roasting process. Proteins in skim milk and soy protein isolate were the source of amino groups, while reducing sugars were obtained from added refined sugar.

The parameter used to indicate the greenish-red color of a material is called the a* (redness) value. Meanwhile, the parameter used to indicate the bluish-yellow color of a material is called the b* (yellowness) value (Winarno, 2004). The results of analysis of variance (Table 1) showed that the treatment of variations in roasting temperature, formulation of ingredients, and the interaction between the two factors showed a very significant effect (P<0,01) on the a* value. The results of the analysis of variance (Table 1) showed that the variation of the roasting temperature did not show a significant effect (P>0,05), while the variation of the material formulation and the interaction between the two factors showed a very significant effect ($P<0,01$) on the value of b*.

The a* value of the food bar as a result of this study ranged from 11,80 (sample S_1R_1) to 16,80 (Sample S_3R_4). Meanwhile, the b* food bar value as a result of this study ranged from 28,73 (sample S_1R_3) to 32,77 (sample S_3R_1). This result is higher than the research result of Nurhayati et al. (2018) which has a* value ranging from 7 to 8 and b* from 16 to 18. This shows that the food bars based on beneng taro, mocaf and soy protein isolate have increased color and are similar to food bars made with purple sweet potato flour and ripe banana.

The more the addition of beneng taro flour and the higher the roasting temperature, the higher the a* and b* values produced. This can be caused by beneng taro containing carotenoid, namely a group of pigments that cause yellow, orange to red colors (Wahyuni et al., 2020). According to Budiarto and Yunia (2017), the carotenoid content in Taro Beneng flour is quite high, reaching 6,92 ppm or 0,692 mg/100 g.

Food Bar Texture

One of the factors that determine the quality of foodstuffs in the form of solid foods such as food bars is texture. The level of hardness of the food material determines how much pressure must be applied to crush it (Aulia, 2017). The results of analysis of variance (Table 2) showed that the treatment of variations in roasting temperature showed a very significant effect ($P<0,01$), while variations in the formulation of materials and interactions between the two factors did not show a significant effect ($P>0,05$) on the texture value. The texture of the food bar as a result of this study ranged from 25,96 (sample S_2R_4) – 33,06 (sample S_3R_2) N. Based on the results of the analysis, there was a decrease in the texture value in the ratio of 70:30 (R_2) and 50:50 (R_4) ingredient formulations.

This result is not in accordance with the literature described by Haliza et al. (2012) and Jariyah et al. (2017) which explains that the more taro flour is added, the food products produced tend to be harder. Haliza et al. (2012) added that the high content of dietary fiber in taro beneng flour is thought to be one of the components of the material that can affect the hardness value of the product. The hardness value of a food product increases along with the increase in the amount of dietary fiber contained in the material. According to Nurapriani (2010) the content of dietary fiber in beneng taro is high when compared to other taro cultivars such as taro butter which contains 6,08% dietary fiber. The result of Putri et al. (2018) research also revealed that the dietary fiber content of taro beneng was 9,52%.

Jariyah et al. (2017) explained that the presence of starch in taro flour will cause food products to be harder. This is because starch has a function to form texture, density, water binding and increase the volume of food products. According to Kusumasari et al. (2019) the starch content in taro beneng flour is 56,29%. Nindyarani et al. (2011) stated that the starch content in purple sweet potato flour was 74.57%, when compared to the starch content in beneng taro, the starch content in beneng taro was relatively low. Flour that has a high starch content will provide a strong and compact texture. This is in line with research conducted by Wulandari (2017) that the addition of more sweet potato flour compared to red bean flour has a higher texture value. Nindyarani et al. (2011) explained the main characteristics of starch as a determinant of texture, namely the nature of gelatinization and retrogradation.

In addition to the chemical content of the ingredients, the roasting temperature can also affect the hardness value of the food bar, an increase in the roasting temperature causes the texture of the food bar to get harder. This is supported by Muchtadi and Sugiyono

(2014) that the level of hardness of a food product is influenced by the roasting temperature. These results are in accordance with the research conducted by Azizaah et al. (2022) where a higher roasting temperature has a higher hardness value than a food bar with a lower roasting temperature. High roasting temperatures can cause the water content of the material to decrease due to evaporation of water in the heating process. The lower the water content in a food product, the harder the texture of the product will be.

Sensory Characteristics

Sensory analysis is a process of identifying, analyzing and interpreting product attributes through the five human senses such as the senses of sight, smell, taste, touch, and hearing. The purpose of sensory analysis is to determine the response or impression obtained by the human senses to a stimulus caused by a product. Based on the panelists' preference for color, taste, aroma, texture and overall parameters, a sensory assessment of the food bar was conducted (Figure 1). Color is a very important component in determining the quality and degree of acceptance of a material (Cicilia et al., 2021). Color is also the first impression obtained from a product to determine acceptance or rejection by consumers of a product.

Panelists' assessment of the color of the food bar has a real effect. The color of the food bar that was most favored by the panelists was the S_2R_3 sample (formulation 60:40; roasting temperature 140℃) while the S_2R_4 sample (formulation 50:50; roasting temperature 140℃) was the sample that the panelists did not like. This shows that on average the panelists evaluate that the food bar with more beneng taro flour added and baked at a higher temperature had a less favorable brown color than the addition of more soy protein isolate. Pradipta & Widya (2015) added that a change in the structure of starch granules will produce a brownish color when subjected to the heating or roasting process for a long time. This is in accordance with research conducted by Seftiono & Intan (2020) that the more addition of beneng taro flour will produce a darker color and are not favored by the panelists due to carotenoid degradation and the Maillard reaction.

Taste has an important role in determining how good a food product is. Consumers will not accept a food product even though it has a good and attractive color and aroma if it does not taste as good. The sense of taste is able to detect taste. A compound must be soluble in saliva so that the microvillus connection and the impulses that are formed are sent through the fibers to the condition center so that the compound can be recognized for its taste (Haliza et al., 2012).

Panelists' assessment of the taste of the food bar did not have a real effect. The taste of the food bar that was most favored by the panelists was sample S_2R_3 (formulation 60:40; roasting temperature 140℃), while sample S_2R_1 (formulation 80:20; roasting temperature 140℃) was a sample that the panelists did not like. This shows that on average the panelists assessed that the food bar with more beneng taro flour added and baked at a higher temperature had a more favorable taste than the addition of more soy protein isolate. This is presumably because the addition of soy protein isolate caused a Maillard reaction which caused an after taste that was not favored by the panelists. Fajri et al. (2013) explain that chemical compounds, temperature, consistency, interactions with other flavor components, the length of the cooking process, and many other factors can affect the taste of food products. Making food bars also uses skim milk, margarine and salt to add flavor. In addition, sugar is also added to give a sweet taste to the food bar.

Aroma is an odor caused by chemical stimuli detected by the olfactory nerves in the nasal cavity when a food product enters the mouth (Garnida, 2020). Consumer perceptions of the taste of food products are usually influenced by the aroma, so aroma is one of the important factors in organoleptic testing. Panelists' assessment of the aroma of the food bar did not give a real effect. The aroma of the food bar that was most favored by the panelists was sample S_2R_2 (formulation 70:30; roasting temperature 140℃), while sample S_2R_1 (formulation 80:20; roasting temperature 140℃) was a sample that the panelists did not like. This shows that on average the panelists assessed that the food bar added with beneng taro flour had a more favorable aroma than the addition of more soy protein isolate.

This is thought to be due to the unpleasant smell of soy protein isolate which is still felt from soybean raw materials. This is in accordance with research conducted by Juita et al. (2018) that red bean flour has a distinctive nutty aroma so that it is not liked by panelists because of the unpleasant aroma. The presence of an unpleasant odor in a food product is a consideration for consumers in accepting or rejecting the product. Based on the research of Cicilia et al. (2021) the presence of an unpleasant odor in modified jackfruit seed flour cookies caused the panelist's assessment to be low. This shows that unpleasant odors can reduce panelists' assessment of the aroma of a food product.

Pertiwi et al. (2017) explained that the presence of unpleasant odors was due to the presence of the lipoxygenase enzyme which naturally gives nuts a special aroma. Fanzurna & Mohamad (2020) added that a food product with a bad taste is caused by the presence of volatile components that make up the aroma, including aromatic compounds and esters. The volatile compounds in the material will evaporate when the roasting process occurs, resulting in a distinctive aroma in the material. Each ingredient used in the manufacture of food bars produces a different aroma. The addition of skim milk and vanilla can reduce the unpleasant odor found in food bars. According to Lestari (2015) taro flour has a savory aroma characteristic so that the amount of taro flour used will affect the aroma produced.

The sensation of pressure that can be observed with the mouth through biting or chewing, as well as touch with the fingers is called texture. The sensory attribute that is often used as a benchmark in texture assessment by consumers is hardness. The texture of a product is strongly influenced by the water content contained in a product (Ferdiansyah, 2015).

The texture that is not too hard and not too soft is preferred by the panelists. Panelists' assessment of the texture of the food bar has a real effect. The texture of the food bar that was most favored by the panelists was the S_3R_1 sample (80:20 formulation; 120℃ roasting temperature), while the S_2R_4 sample (50:50 formulation; 140℃ roasting temperature) was the sample that the panelists did not like the texture. This shows that on average the panelists evaluate that the food bar with more beneng taro flour added and baked at a higher temperature had an unfavorable texture compared to the addition of more soy protein isolate and a lower temperature.

This is presumably because the protein contained in soy protein isolate is higher than that of taro beng flour. This is in accordance with the research conducted by Cicilia et al. (2021) that the cookies density value of the research results is influenced by the higher protein content of modified jackfruit seed flour compared to wheat flour. Rauf (2015) explained that proteins will be easily denatured at high temperatures, causing the hydrogen bonds to break which will form a helical structure and proteins will interact with water. Water that is absorbed into the starch during gelatinization during baking can cause the water content of a material to decrease, causing the density of cookies to become harder.

The conclusion of the panelist's assessment of the food bar from several parameters carried out is the overall sensory parameter. Although there are several methods of objective analysis that can be used as a sign of a decrease in the quality of a food ingredient, the final determination is panelist satisfaction (Winarno, 2004). Overall, the most preferred food bar by the panelists was the S_2R_3 sample (formulation 60:40; roasting temperature 140℃), while the S_2R_1 sample (formulation 80:20; roasting temperature 140℃) was the least preferred sample by the panelists as a whole.

CONCLUSION

The selected food bar formulations were 30% beneng taro flour and 70% soy protein isolate with a roasting temperature of 140℃. The characteristics of the food bar formulation S_2R_2 are texture 26,59 N; value L* 46,75; value a* 14,99; value b* 32,17; and the value of color, taste, aroma, texture and overall preference respectively are 2,23; 2,10; 2,23; 2,15; and 2,25.

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Table 1. The response of L*, a* and b* food bar values to variations in roasting temperature and ingredient formulation

Variation of roasting temperature (S) (℃)	Variation of ingredient formulation (R) (%)				Average
	80:20 (R_1)	70:30 (R_2)	60:40 (R_3)	50:50 (R_4)	
L* value					
120℃ (S_1)	49,18±0,92 tn	46,74±0,39 tn	46,15±1,86 tn	45,47±1,54 tn	46,89^{B}
140℃ (S_2)	48,41±0,43 tn	46,75±0,38 tn	45,88±0,72 tn	45,40±0,06 tn	46,61^{B}
160℃ (S_3)	47,12±0,38 tn	46,80±1,34 tn	44,65±1,05 tn	42,84±0,65 tn	45,35^{A}
Average	48,24^{Z}	46,77^{Y}	45,56^{X}	44,57^{W}	
a* value					
120℃ (S_1)	11,80±0,49 a	14,36±0,21 c	13,04±0,83 b	15,56±0,43 d	13,69^{A}
140℃ (S_2)	12,20±0,95 a	14,99±0,05 cd	15,09±0,25 cd	15,53±0,18 d	14,45^{B}
160℃ (S_3)	14,67±0,34 cd	15,48±0,38 d	15,24±0,01 d	16,80±0,41 e	15,55^{C}
Average	12,90^{W}	14,95^{Y}	14,46^{X}	15,96^{Z}	
b* value					
120℃ (S_1)	28,77±0,38 a	32,27±0,39 de	28,73±0,16 a	31,73±0,92 cde	30,37 tn
140℃ (S_2)	29,23±1,24 a	32,17±0,61 de	29,62±0,64 ab	31,71±0,32 cde	30,68 tn
160℃ (S_3)	32,77±0,25 e	31,06±0,72 cd	29,12±0,82 a	30,71±1,10 bc	30,91 tn
Average	30,26^{X}	31,83^{Y}	29,16^{W}	31,38^{Y}	

Table 2. Texture response (N) of food bar to variations in baking temperature and ingredient formulation

Variation of roasting temperature (S) (℃)	Variation of ingredient formulation (R) (%)				Average
	80:20 (R_1)	70:30 (R_2)	60:40 (R_3)	50:50 (R_4)	
120℃ (S_1)	29,47±1,16 tn	26,46±2,18 tn	29,73±1,74 tn	29,50±1,86 tn	28,79^{A}
140℃ (S_2)	28,10±3,24 tn	26,59±1,91 tn	31,64±1,08 tn	25,96±1,72 tn	28,07^{A}
160℃ (S_3)	32,41±1,66 tn	33,06±3,24 tn	31,80±1,16 tn	29,68±2,10 tn	31,74^{B}
Average	30,00^{tn}	28,70^{tn}	31,06^{tn}	28,38^{tn}	

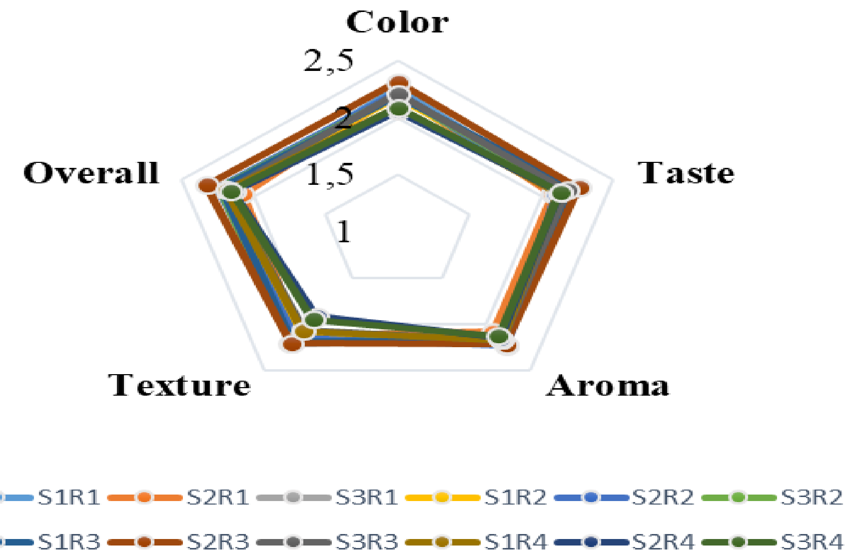

Figure 1. Sensory characteristics of food bars made of formulation S_1R_1, S_2R_1, S_3R_1, S_1R_2, S_2R_2, S_3R_2, S_1R_3, S_2R_3, S_3R_3, S_1R_4, S_2R_4 and S_3R_4

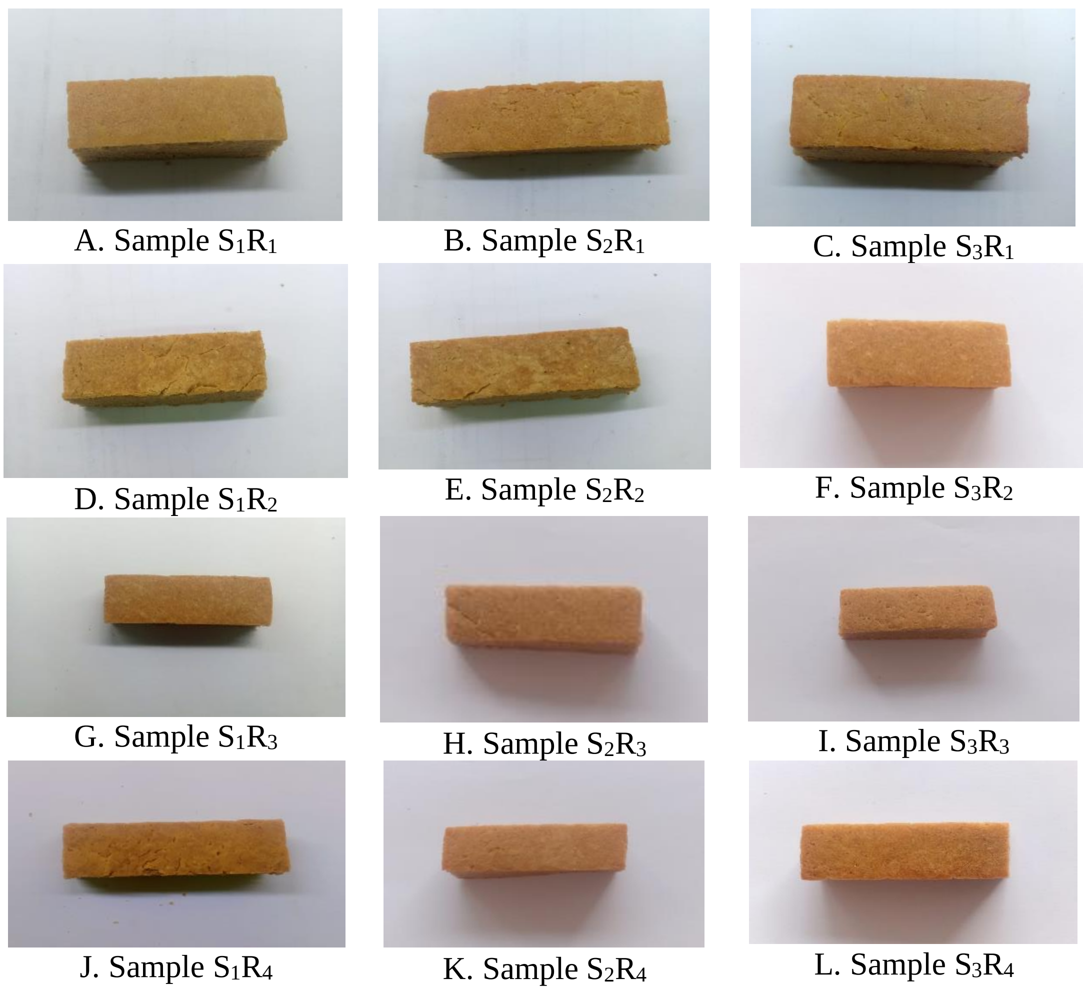

Figure 2. Food bar appearances

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