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Canna indica L and *Spirulina platensis* for Food Security

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Abstract: Climate change threats to food security, because it decreases agricultural productivity. *C. indica L* can adapt to environment and produce white and red rhizomes. Dominant content both of rhizomes are carbohydrates which increase protein as nutrition by adding *S platensis*. The research aims to determine potential of composite flour of *C. indica L* and *S. platensis* as alternative healthy foodstuff for food security. The method consists of formulation composite flour, proximate tests, determining amino and fatty acids, in vitro growth tests of Lactobacillus, cookies and meatball formulations. The result showed composite flour from red rhizome (RRCS) contains 11% water, 21.745% protein, 0.5% lipid, 9.99% dietary fiber, 57.81% carbohydrates, and 8.94% ash. While white rhizome (WRCS) contains, 8.75% water, 17.41 protein, 0.29% lipid, 9.54% dietary fiber, 67.601% carbohydrates and 5.94% ash. Amino and fatty acid appears in both composite flours were similar, except proline and oleic acid higher in RRCS. Lignoceric founded only in WRCS. In vitro test showed Lactobacillus (BAL) grew on modified MRSA from RRCS and WRCS. Both of composite flour processed into marketable cookies and meatballs formulations. Based on the result, *C. indica* L and *S. platensis* composite flour can be used as alternative food for food security.

Keywords: Change; Climate; Healthy Food; Security

Introduction

Food security is defined in Law No. 18 of 2012 as the food availability for the state to persons in sufficient numbers and of good quality, that is safe and nutritious, but that should not violate religious rules, customs, and culture (Muliarta et al., 2023) . Climate change is one of the factors threatening food security. The impact of climate change included drought, pest and disease resilience crops (Bvenura & Kambizi, 2022). Those conditions are the cause of agricultural productivity deduction. The simulation results of the annual crop productivity model due to climate change occurs in the middle of the 21st century. It shows a deduction in food security productivity globally approximately 3-12%. By the end of the 21st century will reach 11-25% (Wing et al., 2021). One of these phenomena already occur in West Nusa Tenggara Indonesia by lack of rice production (Khairulbahri, 2021). Other result shows that harvest corn production in China at time duration between 1979 to 2016 less of corn harvest or production by 1.7%. As facts happened the corn production became 5.19 kg 667 m -2 every temperature increase 1^{o} C (WU et al., 2021) the increase in temperature due to climate change that occurred in 1998 to 2017 also decreased corn production in Malang in 2018.

Lack of food production can be a cause of hunger. The World Food program explains that in 2020 there were 768 million people experiencing hunger (Syakirotin et al., 2022). Data shows that 95% of hunger that occurs is caused by lack of inadequate energy (Amelia et al., 2023) . The main source of energy comes from plants that produce carbohydrates. This phenomenon shows indications of food insecurity that can affect nutritional status (Firna et al., 2018). . Based on existing conditions, we need to find food source alternative which are can adapt with current climate change. This is in line with one of the government's steps in maintaining food security by diversifying food (Indrayani et al., 2023). In the growth of children,

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diversity of food consumption is needed to avoid stunting (Suhaimi et al., 2023). One alternative food source is jackfruit seed and oyster mushroom (Nirmalawaty et al., 2023). Other alternative food sources that are easy to grow and able to adapt to environment also need to be studied.

C.indica L. is a plant can adapt well to environmental such as intensity of sunlight and recommended for agroforestry practice (Sasaerila et al., 2021). C. indica L is native to South America and has spread throughout the world. This plant is widely cultivated in the tropics because it produces rhizome which is used as alternative food (Riandini et al., 2023)

The results of a study showed *C. indica L* contain 28.5% starch, 5.75% lipid, 4.72% crude protein, 25.1% fiber and vitamin with total estimated caloric values of 423/100 g dry weight (Ayusman et al., 2020a). *C. indica L* has two colors variation of the resulting rhizome, named as red and white. *C. indica L* starch belongs to the international standard quality range and can be used as confectioneries, noodles, thickening soups and sauces (Algar et al., 2019).

Based on nutrition content *C. indica L* has potential to use as alternative foodstuff. To increase protein levels on rhizome, *C. indica* L is proven by adding *S. platensis* which is a microalgae. This species contains chlorophyll a and phycobiliprotein that can live in saline conditions (Widawati et al., 2022). *S. platensis* can live on freshwater, in tropical weather and arid regions (Jung et al., 2019a) and able to adapt to the extreme environments such as pH and saline (Ismaiel et al., 2018).

Dry of *S. platensis* content 55-70% protein, 15-25% carbohydrate, 6-9% lipid, 8-10% dietary fiber, and 3-7% humidity (Jung et al., 2019b). *S. platensis* also contains mineral, vitamin, fiber, phycocyanin, pigments, unsaturated fatty acids, beta carotene, thiamine, nicotinamide, pyridoxine, riboflavin, fatty acid, phenolic, phycobilin protein, catechin hydrate, epicatechin, enzyme Superoxide Dismutase (SOD). Antibacterial, anticancer, antidiabetic, antioxidant, antihyperglycemic, antiinflammation dan antihypertension (Kamaludin & Holik, 2022) .

In addition to nutritional content, it is necessary to know the potential source of these foodstuffs as prebiotics. The importance of prebiotic in food as for health promotor supporting fermentation of intestinal microbiota. Fermentation will produce of short Chain Fatty Acid (SCFA) that provides benefits for lipid, glucose, and immune homeostasis in conditions adequate intestinal microbiota (Silva et al., 2020). Existence of SCFA in intestine reduce the risk of colorectal cancer and inflammation in intestine(Campos-Perez & Martinez-Lopez, 2021). *S. platensis* from the Java Sea contains omega 6, gamma linoleic acid, carotenoid dan vitamin (Prijanti et al., 2022)

The obstacles to achieve food security is unacknowledgeable of health benefits alternative food sources in the community including process methods as various menus for daily consuming. However, composition of flour from *C. indica L* and *S. platensis* are not popular. As people do not know the benefits of consuming and processing into various menus which already common. Cookies and meatballs are favorable menus by market. The manufacturing processes from composite flour form *C. indica L* flour and *S. platensis* are unacknowledgeable. However, the biscuits manufacturing from wheat and Spirulina 2, 4, 6, and 5% was success (Saharan & Jood., 2017).

The aims of research are to determine potential of composite flour of *C. indica L* and *S. platensis* as alternative healthy foodstuff for food security in the face of climate change. The potency is based on nutritional considerations including the growth response of Lactobacillus in vitro as well as formulations for making cookies and meatballs

Method

Chemicals and instrumentation

Samples were prepared at chemistry laboratory of Al Azhar University, South Jakarta, Indonesia) and kept at room temperature. Afterwards proximate analysis was conducted. Fatty acids and amino acids composition was determined in the chemistry laboratory of the Center for Post-Harvest Research and Development, Bogor. In vitro growth of *L. casei* Shirota and *L. rhamnosus* was examined in the microbiology laboratory of Indonesia Al Azhar University.

Steps of the research

The stages of research consist of 5 which are carried out sequentially (Figure 1).

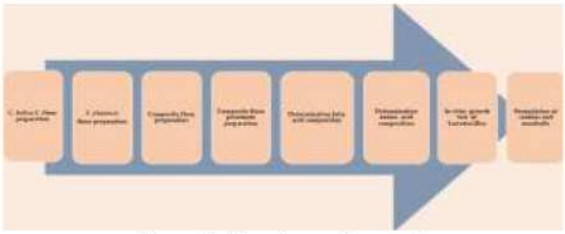

Figure 1. The stages of research

C. indica L Flour Preparation

Good-quality white and red *C. indica L* rhizomes were selected, peeled, washed, and sliced. These slices were immersed in sodium metabisulfite solution acted as an anti-browning agent. Then drained and sun dried for 3 days. The dried rhizomes were ground and sifted becoming flour.

S. platensis Flour Preparation

S. platensis with Optical Density (OD) 0.5-0.7 was sub-cultured in a medium containing Nitrogen (N), Phosphate (P), and Potassium (K) (16:16:16) up to 0.75 g/L, and 1% NaOH added to maintain the pH at 9.5 for 12 days. The samples reached OD 0.7-1 harvested for further processing into flour. The flour prepared through flocculation, filtering, drying, and milling. A bio flocculant solution containing a mixture of 20 mL of vinegar + 0.2 g of chitosan + 180 mL of distilled water added to *S. platensis* in a ratio of 1: 100 (v/v). Afterwards, the flour filtered by filter paper. The left residue dried in an oven at of 45-70°C for ± 1 day. Dried sample grounded and spirulina flour was obtained.

Composite Flour Preparation

C. indica L flour from red and white rhizomes and *S. platensis* flour mixed at a ratio of 10:1. This mixture stirred until homogeneous and flour was obtained.

Composite Flour Proximate Composition

Composite Flour proximate composition was analyzed according to the Association of Official Methods of Analytical Chemists (AOAC, 2010) approved methods, including water, protein, lipid, ash, carbohydrate, and dietary fiber content.

Determination of Fatty Acids Composition

A total of 2 mL of the lipid-extracted sample put into a test tube and 4.5 mL of 0.5 N NaOH added in methanol. It stirred, heated for 5 minutes at 50°C, then cool it down. Afterwards, 3 ml of BF3 poured into the methanol and mixed using a vortex at 50°C for 5 minutes. Added Hexane and mixed it with vortex at 50°C for 5 minutes. Top layer of sample poured into a vial and injected into the GCMS machine.

Determination of Amino Acids Composition

A sample of 0.05 g put into an Erlenmeyer flask mixed with 5 mL of 6 N HCl. It was hydrolysed and put in an oven at 115°C for 12 hours, filtered with filter paper. A 0.5 mL of it was transferred into a test tube. The sample dried using nitrogen gas mixed with 3 ml of 0.02 N HCl. Afterward, filtration carried out with Millipore 0.45 PTEE. A total of 20 µL sample extract poured into a vial tube followed by of 140 µL buffer isolate and 40 L reinforcing reagent. The sample put into the oven and injected into the HPLC for 45 minutes.

In vitro growth of L. casei Shirota and L. rhamnosus

The MRSA medium used for vitro assays to grow *L. casei* Shirota and *L. rhamnosus*. Both composite flours applied as an additive to modify the medium. Glucose and peptone in the De Mann Rogosa and Shape Agar (MRSA) as substitute mixed with 20 g of *C. indica L* and 10 g of *S. platensis*. Based on the formulation, there were combinations of modified bacterial growth media.

L. casei Shirota grown in unmodified MRSA (UMW). Modified MRSA with the substitution of white rhizome flour (M-GCW), modified MRSA with the addition of *S. platensis* flour (M-PSW), as well as modified MRSA with the addition of white rhizome flour and *S. platensis* flour (M-GCW-PS).

L. rhamnosus grown in unmodified MRSA (UMR). Modified MRSA with substitution of red rhizome flour (M-GCR) then modified MRSA with the addition of *S. platensis* flour (M-PSR), as well as modified MRSA with substitution of *C. indica L* red rhizome flour and *S. platensis* flour (M-GCR-PS).

The growth medium set at a pH ranging from 6.4 to 6.6, while all media sterilized by autoclave and placed in 10 mL petri dishes. Stock cultures of *L. casei* Shirota and *L. rhamnosus* grown on the MRSA medium, then put into 10 mL of 0.85% NaCl and diluted. Both bacteria inoculated by spread plate method into each test medium with different formulations, and then incubated at 37°C aerobically for 48 hours. The total population in all media formulated as follows (Chouhan, 2015; Wulandari, 2023).

$$Total\ population\ \left(\frac{CFU}{mL}\right) = Number\ of\ colonies \times \frac{1}{Dilution\ factor} \times Total\ plating\ volume \quad (1)$$

Formulation cookies and meatball from C. indica L flour and S. platensis

At this stage, conducted testing of cookies and meatballs formulations acceptable by the community.

Result and Discussion

Proximate Analysis

The data analysis showed that the WRCS flour contained higher carbohydrates than the RRCS counterpart as 67.607% and 57.81% respectively. The WRCS flour contained 8.75% water, 0.29% lipid, 17.41% protein, and 5.94% ash. This content as presented in Table 1.

Table 1. Proximate Analysis of white and red rhizome composite flour

Nutrition	RRCS (%)	WRCS (%)
Protein	21.75	17.41
Total Lipid	0.5	0.29
Ash	8.94	5.94
Water	11	8.75
Carbohydrate	57.81	67.61
Fiber	9.99	9.54

The specification of WRCS and RRCS had lowest water content compared to wheat flour Codex 199-1995 which was 14.5% m/m. The results showed the potential

expired date both of composite flour were slower than wheat flour. It is possible to distribute both of composite flour to consumers.

The WRCS and RRCS flour had a total ash content of 5.94% and 8.94%. Ash related to in organic residue after complete oxidation in food sample in proximate analysis (Ismail, 2017). The mayor minerals present food such as Calcium (Ca), Natrium (Na), Kalium (K), Chloride (Cl), Phosphorus (P) and Sulfur (S), while trace mineral is Iodine (I), Zinc (Zn) , Selenium (Se), Iron (Fe), Manganese (Mn), Copper (Cu), Cobalt (Co) Molibdenum(Mo), Floride (F), Cromium (Cr) and Boron (B). Calcium influences variety of physiological processes to build and maintain bone, as well as normal nerve and muscle function. Phosphorus is a component of adenosine triphosphate (ATP) and nucleic acids. Mg, Co, Se, Zn, Fe, Mn, and Mo are important cofactor enzymes (Gharibzahedi & Jafari, 2017). The ash content was detected at 0.41% in the red rhizome and 0.47% in the white rhizome of *C. indica L.* The *S. platensis's* ash increasement content because it has minerals such as Ca, Fe, P, I, Mg, Zn, Se, Co, Mn, K, and Na *S. platensis* contains Ca, P, Mg, Fe and Zn respectively 620.8, 790.42, 285.90, 49.5, and 3.95 mg/100 g. (Gutiérrez-Salmeán et al. 2015).

Protein and amino acids

Protein is a macronutrient that forms important building blocks in the formation of biomolecules. Increasing protein, dietary fiber, and ash content in composite flour influenced by the addition of *S. platensis*. It was indicated by the elevation in protein from 4.72% to 21.75% and 17.41% in RRCS and WRCS. Protein content in RRCS is the highest from *Abalistes stellaris* (ikan etong) 18,89 and *Euthynnus affinis* (ikan tongkol) 20,27% (Januarita et al., 2021). The protein content found in RRCS composite flour is higher than that found in salted egg yolks, which is 16.29%. The protein content found in RRCS composite flour is higher than that found in salted egg yolks, which is 16.29% (Setyaji & Monica, 2023). This condition is also in the protein content of WRCS composite flour, which is 17,42%. Composite of 10% S platensis on artificial meat with other additions of wheat gluten, 10% soybeans, mushrooms, tofu showed a protein content of 29.32% (Sari et al., 2022).

The changes in dietary fiber content from 0.04% to 9.99% and 9.54%, as well as ash from 1.37% to 8.94% and 5.40%. Plant proteins have important role in regulation of metabolism and modulation to reduce cholesterol, and triglycerides through inhibition of synthase fatty acids which has impact as anti-obesity. Target regulation of cholesterol and triglyceride metabolism through genes that function to reduce cholesterol and triglycerides metabolism as well as dipeptidyl peptidase IV (DPP-IV). As an antidiabetic effect plant protein works on alpha amylase, alpha glucosidase metabolism gene cause to lower blood sugar (Nagaoka et al. 2021).

Amino acids as a part of protein contain in RRCS and WRCS flour are amino acid essential and non-essential. Essential amino acid contain in both of flour are phenylalanine, valine, threonine, methionine, histidine, leucine, and lysine. Actually, essential amino acids are main role in maintaining tissue and organ functions.

The non-essential amino acids needed for metabolism. Our body can produce or copy it but sometimes the production is insufficient. The types of non-essential found amino acids found in two composite flours are aspartate, serine, glycine, arginine, alanine, proline, and cysteine. Glycine can be synthesized by our body but the production is insufficient (Alves et al., 2019). RRCS and WRCS flour can be as food source containing glycine. Following are the amino acids contained in both type of composite flour compared to *S. platensis* (table 3). Amino acids content of *S. platensis* in both types of composite flour show similarities. Different types of amino acids are tryptophan which only found in *S. platensis* (Bortolini et al., 2022). The absence of tryptophan caused by different types of *S. platensis* culture medium used. RRCS flour also shown to contain higher proline compared to WRCS flour and *S. platensis*. Based on this, it is suspected that proline is not only derived from *S. platensis* but also from red rhizome *C. indica L.* Unlike the case with aspartate which has highest in WRCS, so it is predicted come from white rhizome *C indica L.* The highest Cysteine found in *S. platensis* and followed by WRCS flour.

Table 2. Amino acids in the Composite Flour and Bartolini *et al.*, 2022

Amino acids	RRCS (%)	WRCS (%)	Bartolini (et al., 2022}
Aspartate	0.62	0.75	0.63
Serine	0.31	0.32	0.30
Glutamate	0.82	0.87	0.84
Glycine	0.35	0.37	0.34
Histidine	0.12	0.12	0.11
Arginine	0.49	0.46	0.44
Threonine	0.34	0.37	0.33
Alanine	1.99	0.29	0.50
Proline	1.99	0.29	0.25
Cysteine	0.002	0.03	0.06
Tyrosine	0.18	0.29	0.30
Valine	0.42	0.45	0.42
Methionine	0.08	0.01	0.17
Lysine	0.3	0.33	0.34
Isoleusine	0.34	0.37	0.36
Leucine	0.52	0.59	0.61
Phenylalanine	0.3	0.34	0.33

The amino acid contained in WRCS and RRCS flour were aspartate, serine, alanine, glutamate, glycine, histidine, cysteine, arginine, methionine, glutamate, valine, and isoleucine will be explained as follows. Aspartate in the form of L-aspartate has function as protein biosynthesis and a neurotransmitter (Johnson, 2017). By adding D-aspartate to a neurotransmitter also functions as a regulation in reproduction, hormones, and nerve protection (Li et al., 2018). Serine as a non-essential is a precursor for the biosynthesis of glycine and cysteine, thus the availability of serine is needed for the formation of both types of amino acids.

Alanine serves to avoid the symptoms of paresthesia and reduces urine overflow, while the addition of beta-alanine increases exercises intensity in athletes (Harris & Stellingwerff, 2013). Glutamate is a neurotransmitter to the nerve system, name as presynaptic an postsynaptic (Hao & Plested, 2022). Glutamate can be synthesized from glucose a various nitrogen containing nutrients, as well as glutamine, aspartate, asparagine, serine, glycine, proline, and alanine. On certain circumstances, glutamine, glutamate, glycine, proline, and arginine could not be synthesized in quantity or quality because the influence of pathophysiology.

Imbalance and indispensable amino acids will reduce growth and trigger various diseases. For the example glycine, very effective for promoting owth, improving health, and preventing various diseases including cancer, cardiovascular disease, some inflammatory diseases, obesity, and diabetes. Also influencing sleep quality and neurological function. Also serves as a precursor for several molecular metabolites, such as creatine, glutathione, purines, and porphyrins (Razak et al., 2022).

Histidine in de essential amino acids that affect proton buffers, metal ion chelation, scavenging of reactive oxygen and nitrogen species, erythropoiesis, and histaminergic systems. Histidine contain in proteins ch as haemoprotein, HIS glycoprotein, hystatins, HIS calcium binding protein and filaggrin, HIS containing dipeptides especially carnosine, methyl and Sulphur. Thus, it contains of HIS (3 methylhistamine, 1 ethylhistidine and ergothioneine. HIS as component preservation and myocardial protect in cardiac surgery. HIS supplementation is needed on neurological disorders, atopic dermatitis, metabolic syndrome, diabetes, uremic anemia, ulcers, inflammatory bowel syndrome, malignancies, muscle performance enduring strenuous exercise (Holeček, 2020). Diet methionine 46 mg/ kg/ d, lysine 6.0 g/ d, histidine 8.0 g/ d can reduce fat level and skin condition so can be used for herpes treatment (Cynober et al., 2020). Cysteine functions as an antioxidant has potential to capture Reactive Oxygen Species (ROS). Cysteine function as whitening by forming pheomelanin which produces yellowish or reddish colors and blocking the formation of eumelanin which produces dark colors (Oshimura & Sakamoto, 2017).

Arginine supplements may increase Growth Hormone (GH) and Thyroid Stimulating Hormone (TSH) (Apolzan et al., 2022). Research shows by giving 10 g to men aged 18 to 39 years increases muscle mass by 18.5 kg / up to 25 kg / m2 (Liang et al., 2021a). Arginine is required in spermatogenesis of pregnancy and neonates to maintain nitrogen balance (Watford & Wu, 2018). Methionine is a non-essential amino acid precursor cysteine, as well as phenylalanine as a tyrosine precursor. The intake is needed of methionine and phenylalanine in nutrient. A single intake of good leucine supplements with protein supplements can improve of sarcopenia, which is loss of muscle mass because of decreased mobility (Tezze et al., 2023).

Glutamate undertake decarboxylation will transform into Gamma Aminobutyric Acid (GABA) as a neurotransmitter with microbiota's help. Microbiota-gut-brain axis disorder in cases of depression corrected via human microbiota function through transplantation to sti late GABA (Mayneris-Perxachs et al., 2022).

L-Valine, L-isoleucine, and L-leucine are essential amino acids or synthesize nsufficiency in humans and animals. It is referred to as Branched Chain Amino Acids (BCAAs) because they contain branched side chains in scaffolds necessary for growth, especially physiology and biology. Increa ment levels of BCAAs in humans will reduce accuracy and development of cardiovascular disease, T2DM metabolic disorders, obesity, and neurodegenerative (Liang et al., 2021b).

Lipid and Fatty Acids

The lipid content in RRCS and WRCS flour relatively low as 0.29% and 0.50% compared to standard dietary lipids. People's habits in consuming foods with low lipid levels can minimize the prevalence of obesity (Muriyati et al., 2023) . Both composite flours contain same fatty acid presented in Table 3.

Table 3. Fatty acids in the Composite flour of *C. indica L* and *S. platensis*

Fatty Ac	RRCS (%)	WRCS (%)
Myristic Acid	1.29	1.76
Palmitoleic acid	1.60	2.55
Palmitic Acid	38.25	39.79
Linoleic Acid	24.29	25.94
Oleic Acid	21.57	6.13
Stearic Acid	6.13	1.93
Behenic Acid	1.73	0.8
Lignoceric Acid	0	0.8

The highest saturated fatty acids were palmitic as in 39.79% in WRCS and 38.25% in RRCS. Palmitic acid required for average dietary fat intake of 20 g/day for 20-30% of total fatty acids has functions in fetal brain development (Murru et al., 2022). The second largest fatty acid in both flours is linoleic acid (LA) with number in RRCS and WRCS 24,29% - 25,94%. The LA content found in *Terminalia catapa* L seeds shows lower levels compared to those found in RRCS and WRCS, which is 13.09% (Nirwana et al., 2022). In addition to LA also needed intake of Linolene acid (omega 3) which can be found in Portulaca oleracea (Dewanti et al., 2023; Muhammad et al., 2023).

LA (omega-6) known as Ω-6 polyunsaturated is one of the 6 Polyunsaturated Fatty Acids (PUFA) has essential fatty acids. It affects insulin sensitivity and improves pancreatic ß-cell function (Marangoni et al., 2020). LA fatty acid (PUFA) plays a role in glucose homeostasis in regulating blood sugar homeostasis by increasing insulin sensitivity, increasing blood sugar binding due to improved pancreatic beta cell function in sex, genetics and different environmental factors, besides reduce cardiovascular disease (Hamilton & Klett, 2021).

Palmitoleic found in WRCS at 2.55% and 1.6% RRCS flour. Palmitoleic 9-hexadecenoic acid; 16:1 -7 is a lipokine that functions as a nutraceutical in the treatment of non-alcoholic liver disease. Murinae is inflammation by induction of fat intake of 59% for 12 days, showed increasing insulin tolerance and decreasing serum insulin on day 15 after administration of palmitoleic supplementation 300 mg / kg / day(Souza et al., 2020). Non-essential oleic acid in RRCS flour at 21.57% while WRCS flour is 6.13%. Intake oleic acid can maintain weight via metabolism stimulating AMP activated protein kinase.

In addition, through the prevention of nucleotide binding oligomerization domain like receptor 3/ caspase – 1 inflammasome pathway, induction of oleoylethonalamide synthetase and the down regulation stearoyl-Co A desaturated activity(Tutunchi et al., 2020)

Stearic content in RRCS higher than WRCS flour at 6.13% and 1.93%. Stearic acid saturated with 18 carbon long chain fatty acids, also induces apoptosis and autophagy (Yang et al., 2019). Stearic acid can suppress tumor cell growth. Lignoceric acid specifically found in WRCS flour only and has saturated fatty acids. Behenic acid higher in RRCS flour and stable during binding to proteins as ligand complex in tackling T2DM (Nath et al., 2022). Myristic levels almost same in the two composite flours. Myristic is a saturated fatty acid (14:0) that is essential in lipid milks. These fatty acids are beneficial for cardiovascular health. Research shows the presence of myristic acid is associated with increased concentrations of ApoCIII in cardiovascular patients. Invitro experiments showed myristic acid stimulates ApoCIII in the expression of HepG2 cells and slows down HDL catabolism (Olivieri et al. 2020)

Fiber

The composite flour specification shows highest percentage of dietary fiber as 9.99% for WRCS and 9.54% for RRCS flour. Dietary fiber increases health quality by protecting against constipation problems and preventing digestive cancers, regulating blood glucose and decreasing the risk of diabetes, and reducing cardiovascular disease risk by weight loss through lipid regulation. This also suppresses lipid levels by about 5-10% cholesterol, controls blood glucose, improves immune function, and reduces inflammation (Surampudi et al. 2016). Blood glucose reduction influenced by the dietary fiber effect in composite flour.

Fiber consumption 10 g/day for a median intervention duration of 8 weeks in diabetic type 2 patients significantly reduced HbA1c, fasting blood glucose, fasting insulin, and Homeostatic Model Assessment for Insulin Resistance. Fiber foods are effective in improving glycemic control and insulin sensitivity in diabetic type 2 patients (Mao et al., 2021).

In vitro growth of Lactobacillus

The total population of *L. casei* Shirota cultured in UMW, M-GCW, and M-GCW-PS showed no significant difference ($P > 0.05$), bacterium was 8.23± 0.04, 8.17 ± 0.02, and 8.17± 0.00 CFU/mL in the three media. Table 4 shows a significant difference in the total population in M-PSW at 8.11 ± 0.03 CFU/mL. The higher growth in UMW due to immediate fermentation of glucose, while in M-GCW still through the process of starch decomposition into monosaccharides. *L. casei* Shirota cultured on M-PSW requires time to break down proteins to grow into amino acids.

Table 4. Total population of *L. casei* Shirota growth in various media

Media	Total Population (CFU/mL)
UMW	8.23 ± 0.04
M-GCW	8.17 ± 0.02
M-PSW	8.11 ± 0.03
M-GCW-PS	8.17 ± 0.00

S. platensis contained in the culture media as source of cysteine, proline, glycine and glucose (Zhang et al. 2020) .The result of *L. rhamnosus* cultured in UMR and M-GCR-PS showed no significant difference ($P > 0.05$). According to Table 5, the total population in UMR was 7.79 ± 0.10 CFU/mL and M-GCR-PS contained 7.85 ± 0.00 CFU/mL. However, the total 7.77 ± 0.08 CFU/mL and 7 .55 ± 0.11 CFU/mL found on the M-PSR and modified M-GCR had a significant difference ($P < 0.05$).

The population of *L. rhamnosus* cultured on media M-GCR lower in numbers. Maybe it is due to higher flavonoid content in the red *C. indica L* rhizome compared to white one. Flavonoids as antibacterials can inhibit nucleic acid synthesis, cytoplasm, membrane permeability, and porin functions, including energy metabolism systems, inhibiting binding and formation of biofilms (Xie et al., 2015). The red rhizome of *C. indica L* contains flavonoids, tannins, and sterols are antimicrobial and anthelmintic (Kumbhar et al., 2018). Rhizome in *C. indica* L also contains alkaloids, saponins, polyphenols and terpenoids (Ifandari et al., 2020). *C. indica L* juga while tannins reduce Lactobacillus growth (Peng et al., 2018). Anthocyanins are also mainly antimicrobial *Escherichia coli* dan *Staphylococcus aures* (Khairi et al., 2023) (Khairi et al., 2023).

The *Lactobacillus* in vitro growth is inhibited by flavonoids and tannins of *C. indica L*, but it can support sterol content (Miedes et al., 2023). Reason for higher growth of *L. rhamnosus* on MRSA media because glucose content can used directly. In modified MRSA, the glucose dominantly source comes from the rhizome of *C. indica* L. Research shows the *C. indica L* rhizome has eccentric hila type of glucose (Huang et al. 2015), while the starch content estimated resistant and has higher concentration than glucose.

Table 5. Total Population of *L. rhamnosus* growth in various media

Media	Total Population (CFU/mL)
UMR	7.79 ± 0.10
M-GCR	7.55 ± 0.11
M-PSR	7.77 ± 0.08
M-GCR-PS	7.85 ± 0.00

The content of amino acids, such as methionine, cystine, lysine, and tryptophan, and fatty acids, including palmitic acid, oleic acid, and linoleic acid contained in *S. platensis* supports the growth of *L. rhamnosus. S. platensis.* Beta-carotene can be converted into vitamins A and B, as well as minerals by 3-7%.

C. indica L contains bioactive hydroxytrimethoprim, 3-7 epoxycarophyllan 6 one, swietenine, typhasterol, hexacosanedioic acid a 3-beta, 6-alfa, 7-alpha trihydroxy 5 beta choline 24 oleic acid function as antimicrobe, anthelmintic potential and HIV reverse transcriptase inhibition (Kumbhar et al. 2018b). *C. indica* is rich in 67.99% carbohydrate and 22.50% fiber, 319.15 calory/100 grams. Rhyzome contains 24405 ppm potassium, sodium, calcium, and magnesium (7500, 1345 and 1610 ppm) also Cu, Ni, Mn, Fe, Zn (Gaur 2014). *C. Indica* L has potential as functional food, source of vitamins, nutritional and nutraceutical. Biomass and nutrient characteristics are fiber 25.1%, starch 28.5%, crude oil 4.72% and lipid 5.75% total calories 423 Kcal/100 g wet weight. Rhizome contains minerals and vitamins. Bioactive compounds such as rosmarinic acid, psoromic acid, usnic acid, isoeugenitol, ellagic acid, coumarin acid and switenine able to protect free radical cells including DNA and protein degradation. Acetone extract in rhizome shows antioxidant activity. Ic 50 values with DPPH, ABTS+, O2_, showing 21.23 and 170 ug/ml. Decreasing of Ferric Reducing Antioxidant Power (FRAP), Cupric Ion Reducing Antioxidant Capasity (CUPRAC) and DNA protection correlated with total phenolids and flavonoids. Beta carotene and linoleic acid show significant acetone extract, approxiamtel IC50 is 2.35 ug/ml and acetone extract IC 50 27.1 ug/ml. *C indica* become healthy potential source of nutrients and metabolism (Ayusman et al., 2020b).

Glucose source of growing culture media of *L. casei* Shirota can be substituted with white rhizome *C. indica L.* Red rhizome has greater potential as glucose substitute of growing of *L. rhamnosus. S. platensis* also has potential as a prebiotic and protein source in modified MRSA. As for the two Lactobacillus. Based on these facts, white rhizome flour of *C. indica L.* and *S. platensis* has potential of prebiotic system.

Cookies and meatballs from composite flour

To make cookies and meatball formulation which are healthy and acceptable by market was going through various tests. The acceptable formulation is using *C. indica L* flour 80 gr, *C. indica L* starch 20 gr, coconut oil 60 gr, coconut sugar 50 gr, milk skim powder 40 gr, egg 40 gr and *S. platensis* powder 1 g. Cookies product result is fibrous, soft, and crunchy. Also has density to ensure it do not crumble easily. Taste close to sweet and has smell of coconut aroma to satisfy consumers. The following is presented a figure describing the product of the research process (Figure 2).

Figure 2. Product of research process

Rhizome produced from planting *C. indica* L after being processed into flour is mixed with S. platensis which is the result of cultivation. A mixture of the two

types of flour is called composite flour. Lactobacillus invitro growth tests showed positive results for two types of composite flour. Furthermore, the composite flour was successfully made into cookies and meatballs that were favored.

The composition flour of *C. indica* and *S. platensis* also using for making meatballs and successfully has savory taste, with chewy texture and delicate meat aroma. Formulation of was diced meat 2000 g, *C. indica L* flour 250 g, sago 250 g, *S. platensis* 1 gram, gelatinous 14 gr, egg 80 g, garlic 100 g, pepper 2 tea spoon, onion 100 g. It acceptable by consumers. Cookies and meatballs are safe to consumer who has gluten allergic because not using wheat flour.

Conclusion

Based on the consideration of its nutritional content, potential as a prebiotic and can be processed into food variations such as cookies and meatballs, rhizomes, C. indica L and S. platensis can be used as alternative foods to maintain food security. WRCS and RRCS flour contain carbohydrates, proteins, and lipids that can meet daily nutritional needs and contain prebiotics.

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Author Contribution

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Conflicts of Interest

The results of the research are objective and do not cause problems and conflicts of interest from various parties

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