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Surabhi Kumari
M.Sc (food tech.) Student, Warner College of Dairy Technology, SHUATS, Naini, Allahabad, Uttar Pradesh, India

Shanker Suwan Singh
Assistant Professor, Food Technology, WCDT, SHUATS, Naini, Allahabad, Uttar, Pradesh, India

Komal Yadav
M.Sc (food tech, Student, Warner College of Dairy Technology, SHUATS, Naini, Allahabad, Uttar Pradesh, India

Bhavya
M.Sc food tech, Student, Warner College of Dairy Technology, SHUATS, Naini, Allahabad, Uttar Pradesh, India

Correspondence
Surabhi Kumari
M.Sc (food tech.) Student, Warner College of Dairy Technology, SHUATS, Naini, Allahabad, Uttar Pradesh, India

Development and quality assessment of Gluten-free Bread prepared by using Rice flour, Corn starch and Sago flour

Surabhi Kumari, Shanker Suwan Singh, Komal Yadav and Bhavya

Abstract
Baking industry is currently seeking to expand its products range, but also to costitute a way of maintain and improving general health of people. Bakery products are mainly prepared from wheat as its main ingredients. Bread is widely consumed and is an ideal vehicle for functional delivery. The objective of this work was to develop Gluten-free Bread. The use of rice flour, corn starch and sago flour blends as a source of high carbohydrate, protein and fat and absence of gluten in production of Gluten- free bread was studied. The flour blends of rice flour, corn starch and sago flour were composites at replacement levels of 40:35:25 (T_1), 50:30:20 (T_2), 60:25:15 (T_3) % while the wheat flour bread 100:00:00 (T_0) served as control bread. Various analysis parameters were analyzed by one way ANOVA to obtained a predicted optimum result prepared bread was subjected to chemical, microbial, and sensory analysis to evaluate the suitability of breads were T_1 protein (5.10 %), fat (4.58 %), ash (0.95 %), moisture (31.15 %) and carbohydrate (58.19 %) as comparable to control without adversely affecting the sensory parameters. Based on the result it was indicated that beneficial components of Rice flour, corn starch and sago flour made them more favorable choice for food technologist to develop gluten - free bread especially for celiac disease.

Keywords: Rice flour, corn starch, sago flour, celiac disease

Introduction
Bread is one of the oldest and largest consumed food stuff and is consumed across the globe by all age groups. Breads are defined as a fermented bakery product produced mainly from wheat flour, water, yeast, and salt by a series of process involving mixing, kneading, fermentation, proofing, shaping and baking (Dewettinck *et al*., 2008) [10]. A gluten-free bread is a bakery product that strictly excludes gluten, a mixture of proteins found in wheat and related grains, including barley, rye, oat, and all their species and hybrids (Such as spelt, kamut, and triticale). Gluten-free breads are made with rice flour, corn starch, sago flour and flours made by other cereals such as sorghum etc. (Biesiekierski, 2017) [3]. Gluten causes health problems for those with gluten-related disorders, including coeliac disease (CD), non- coeliac gluten sensitivity (NCGS) and wheat allergy.
Bread products made from wheat flour are consumed worldwide, some individuals are intolerant to prolamins of wheat and the related cereals oats, rye, and barley. This intolerance, celiac disease, seriously impairs intestinal absorption and can lead to severe malnutrition. (Davison *et al*., 1987; Ciclitira *et al*., 1987) [8].
The only certain remedy to this pathology is total omission of gluten from the diet. That is very difficult, however, since many foods contain gluten (Baldo *et al*., 1984) [4]. Many investigators have attempted to develop gluten-free yeast breads. Wheat starch has often been used as a wheat flour replacement.
The World Health Organization states that gluten-free foods should contain less than 1 mg gliadin per 100-g product. Non-wheat cereal products may also be used in bread making. Rice flour is useful since it lacks gluten and contains low levels of sodium and a high amount of easily digested carbohydrates, making it desirable in celiac diets.
Celiac disease (CD) is a chronic enteropathy characterized by an inadequate immune response to ingested gluten from wheat, rye, barley, triticale, and, in some cases, oats. This intolerance results in damage to the small-intestinal mucosa, leading to the malabsorption of nutrients. The only effective treatment for CD patients relies on a lifelong gluten-free diet.

The general prevalence of CD is estimated to be one in 300 (Collin *et. al.*, 1997), although recent population-based screening studies suggest that the prevalence may be even higher (one in 100). At previous trials, rice flour, cornstarch, and cassava starch were used separately in preparing bread because these raw materials are normally used by celiac people in food preparation. Nevertheless, better technological results were obtained when these raw materials were used together in the mixture. The aim of the present study was to use rice flour, cornstarch, and cassava starch in gluten-free bread making and to statistically establish optimal amounts of each ingredient.

Rice flour

Rice (*Oryza sativa*) flour is a popular and widely used gluten-free flour. Rice flour (Also rice powder) is a form of flour made from finely milled rice. Rice flour may be made from either white rice or brown rice. To make the flour, the husk of rice or paddy is removed and raw rice is obtained, which is then ground to flour. Rice flour contains fat, carbohydrates, protein, calcium, iron. Rice flour is very common in parts of the world where rice is more of a staple ingredient than is wheat or wheat flour. In the 1980's, it was introduced to the western part of the world, where it became a primary ingredient in many processed foods in the baking industry. Of the world's total rice production, 90% is grown and consumed in Asia. White rice flour is enriched with vitamins and minerals to meet the nutrient requirements. Rice flour is gluten-free; therefore, individuals with celiac disease can safely consume it. It is often used as a healthier alternative to wheat flour because it is easier to digest and richer in fiber. Because rice posses unique nutritional, hypoallergenic, colorless, and bland taste properties, it is used in baby foods and puddings. Its role in development of foods for gluten-intolerant patients especially has been increasing.

Table 1: Nutrient Composition of rice Flour

Moisture %	13.3
Protein %	8
Fat %	0.4 - 1
Carbohydrate %	79.1
Fiber %	0.6
Ash %	0.4 - 0.7
Calcium (mg/100g)	10
Phosphorous (mg/100g)	150
Iron (mg/100g)	2.2
Thiamine (mg/100g)	210
Nicotinic acid (mg/100g)	3.8

Corn starch

Corn starch or maize starch is the starch derived from the corn (Maize) grain. The starch is obtained from the endosperm of the kernel. Corn starch is a common food ingredient. It is versatile, easily modified, and finds many uses in industry as adhesives, in paper products, as an anti-sticking agent. Corn starch contains carbohydrates, dietary fiber, iron. It has medical uses, such as to supply glucose for people with glycogen storage disease. The corn is steeped for 30 to 48 hours, which ferments it slightly. The germ is separated from the endosperm and those two components are ground separately (still soaked). Next the starch is removed from each by washing. The starch is separated from the corn steep liquor, the cereal germ, the fibers and the corn gluten mostly in hydro cyclones and centrifuges, and then dried. (The residue from every stage is used in animal feed and to make corn oil or other applications). This process is called wet milling. Finally, the starch may be modified for specific purposes.

Table 2: Nutrient composition of corn starch.

Moisture (%)	10
Protein (%)	0.1
Ash (%)	0.2
Fiber (%)	0.1
Fat (%)	0.2
Carbohydrates (%)	87.5

Sago flour

Sago (Tapioca Sago) (Sabudana) is a processed, ready to cook agricultural food product. The only raw material for manufacturing Sabudana is "tapioca root" internationally known as "cassava". Sabudana is considered an acceptable form of nutrition. It is used in a variety of dishes such as desserts like "kheer" (Boiled with sweetened milk) or Khichadi, vada, bonda etc. (Mixed with Potatoes, Ground nuts, rock-salt, and black-pepper or green chillies). Sabudana (Sago) is a produce, prepared purely from Tapioca Root ("Cassava") Botanical name is "Mani hot Esculenta Crantz Syn. Utilissima". In India, the names 'Sago', 'Tapioca Sago', 'Tapioca Globules', 'Sabudana', 'Javvarisi', 'Sabbakki', 'Saggubeeyam' are all used for the same commodity 'Sabudana. It provides only carbohydrate food value, and is low in protein, vitamins and minerals. Sago starch can be baked (Resulting in a product analogous to bread, pancake, or biscuit) or mixed with boiling water to form a paste. In many developing countries bread consumption is continually expanding and there is increasing dependence on imported wheat. Most of these countries, however, grow staples other than wheat that can be used for bread. Some grow various starchy tubers such as sago, yam or sweet potatoes and some others grow cereals such as maize, millet or sorghum. It would therefore be economically advantageous for those countries if imports of wheat could be reduced or even eliminated and the demand for bread could be met by the use of domestically grown products instead of wheat. Sago is a common ingredient used in Indian recipes. In gruel form, sago can function as a healthy alternative to carbonated drinks, providing energy without any artificial chemicals and sweetener.

Table 3: Nutrient composition of sago

Moisture (%)	13.5
Protein (%)	0.15
Ash (%)	0.73
Fiber (%)	0.4
Fat (%)	0.65
Starch (%)	70.25

Justification

The population everywhere desire to eat a healthier diet without changing their conventional dietry patterns. The term gluten-free bread is generally being used for foods that may provide a health and physiological benefits. People decide to go gluten free for a few different reasons – originally, gluten free foods were made available for those suffering from celiac disease, or for those who are sensitive to gluten. More recently a lot of people have decided to go gluten free as they enjoy the taste of well-made items that are free of gluten as they become more widely available. Celiac disease is a condition in which the mucous membrane of the small intestine of glutenin tolerant people is damaged by gluten. The reason gluten can be such an issue for some people is because wheat can be incredibly difficult to digest for some, leaving behind gas in the stomach that can make those with sensitivities feel bloated and uncomfortable. Refined wheat also has very little nutritional value and therefore taking this out of our diet is not usually a problem for us health-wise. Sticking to a gluten free diet can be tricky, and for most of us, cutting bread and cake can be a hard one. Gluten free products allow you to include a little bit of what you enjoy in your diet and also help you feel full throughout the day without the horrible bloated feeling that can come from a sensitivity.

Review of literature

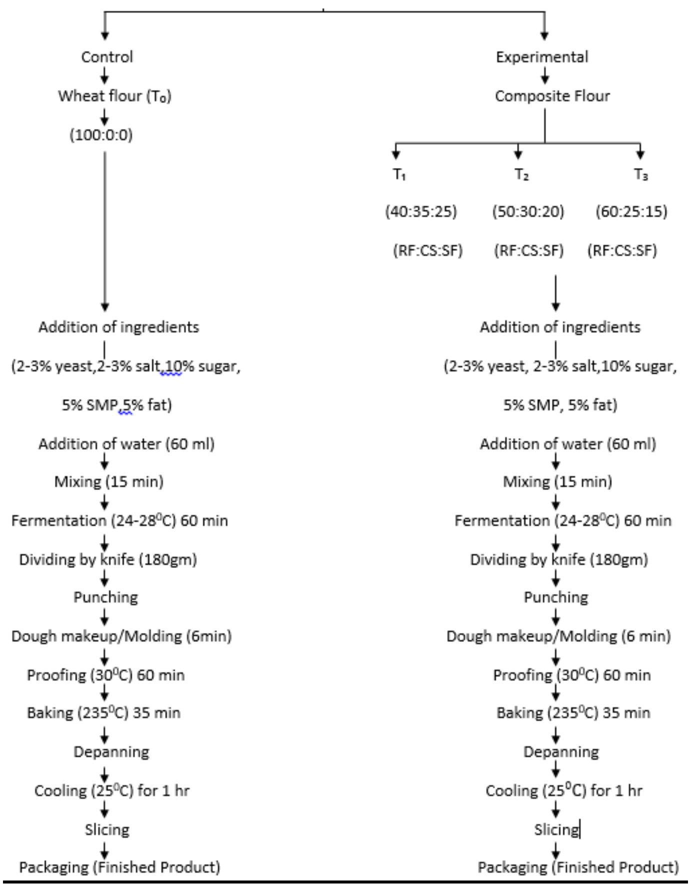
Reported that elimination of gluten increases the role and importance of starch in providing structure and texture to gluten-free products. Starch is the primary source of stored energy in many plants including cereals, legumes, roots and tubers. It provides 70%–80% of the calories consumed by humans worldwide. Reported that isolated wheat starch is often utilised in gluten-free products, but starch-based ingredients should ideally originate from raw materials that are naturally gluten-free. Reported that the incorporation of various hydrocolloids into dough formulations has made it possible to produce bread from rice flour. Studied the effect of rice and waxy rice on the French fry-like products using extrusion before frying. Fries from waxy rice flour had texture most comparable to commercial fast food fried potatoes while fries from non-waxy rice flours were too hard and gummy. Reported that similar treatment may also be applied to bakery products although too high content of high amylose starch in bread leads to deterioration of its quality. The similar work for fasting biscuits was conducted by using sago, peanut, banana, potato, foxtail millet, barnyard millet in different proportions. Reported that Corn ranks as the second most widely produced cereal crop worldwide. Because of the high productivity, corn is by far the most economical cereal to produce. Corn flour contains high levels of many important vitamins and minerals, including potassium, phosphorus, zinc, calcium, iron, thiamine, niacin, vitamin B-6, and folate. Reported that High amylose corn starch is commonly used to increase the level of resistant starch in foods. Gujral *et al.*, (2004) [14] reported that Rice flour exhibits properties such as the absence of gluten, low levels of sodium, protein, fat and fiber, and high amount of easily digested carbohydrates, which are desirable for certain special diets. It has also bland taste, white color, and hypoallergenic properties. Guha *et al.*, (2006) [15] reported that the glutinous rice was suitable material to produce the expanded extruded rice product such as ready-to-eat snacks, breakfast cereal with low bulk density, high expansion and low shear stress. Reported that Rice flour is increasingly popular as a substitute of wheat flour in the preparation of products consumed by wheat-intolerant or celiac patients. It is the most suitable cereal grain flour for the production of gluten-free products due to its bland taste, white colour, digestibility, and hypoallergenic properties.

Materials and Methods

Procurement and collection of ingredients

- Rice to be purchased from local market of Prayagraj.
- Corn starch to be purchased from local market of Prayagraj.
- Sago to be purchased from local market of Prayagraj.
- Other materials also to be purchased from local market of Prayagraj.

Flow diagram adopted for preparation of control and experimental bread



Result and Discussion

The analyzed data is presented in this chapter under the following headings:

1. Chemical characteristics
2. Microbial characteristics
3. Organoleptic characteristics
4. Statistical characteristics
5. Estimation of cost of production

Table 4: Average data for different parameters of control and experiments (in percent)

Parameters	Treatments			
	T_0	T_1	T_2	T_3
1. Chemical Analysis				
Moisture	31.72	31.15	31.23	31.36
Fat	4.21	4.58	4.75	4.80
Protein	9.64	5.10	5.60	6.42
Carbohydrate	53.41	58.19	57.50	56.71
Ash	1.00	0.95	0.90	0.68
2. Microbiological analysis				
SPC × 10^{-3} (colony forming unit /gm)	3.8	4.2	4	4.4
Yeast & Moulds count (cfu/gm)	1.4	1.6	1.6	2.2
Coliform count	Nil	Nil	Nil	Nil
3. Organoleptic Score (9- point hedonic scale)				
Color and Appearance	7.54	8.08	7.39	6.7
Body and Texture	7.85	8.04	7.44	6.76
Flavor and taste	7.73	8.16	7.4	7.12
Overall acceptability	7.70	8.09	7.40	6.85
4. Cost analysis				
Cost in Rs./100g	5.87	41.25	36.36	31.45

Summary and Conclusion

Physico- chemical analysis

Moisture percentage

There was significant difference in moisture content of different treatment combinations. Maximum moisture percent was recorded in the sample of T_0 (31.76) followed by T_3 (31.36), T_2 (31.24), T_1 (31.15). The difference in moisture was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Ash percentage

There was significant difference in ash content of different treatment combinations. Maximum ash percent was recorded in the sample of T_0 (1.01) followed by T_1 (0.96), T_2 (0.90), T_3 (0.68). The difference in ash was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Protein percentage

There was significant difference in protein content of different treatment combinations. Maximum protein percent was recorded in the sample of T_0 (9.64) followed by T_3 (6.43), T_2 (5.61), T_1 (5.11). The difference in protein was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Fat percentage

There was significant difference in fat content of different treatment combinations. Maximum fat percent was recorded in the sample T_3 (4.81) followed by T_2 (4.75), T_1 (4.59), T_0 (4.21). The difference in fat was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Carbohydrate percentage

There was significant difference in carbohydrate content of different treatment combinations. Maximum carbohyfrate percent was recorded in the sample of T_0 (31.76) followed by T_1 (58.20) followed by T_2 (57.50), T_3 (56.71), T_0 (53.42). The difference in carbohydrate was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Organoleptic analysis

Color and appearance score

There was significant difference in color and appearance score of different treatment combinations. Maximum color and appearance score was recorded in the sample of T_0 (31.76) followed by T_1 (8.08) followed by T_0 (7.54), T_2 (7.39), T_3 (6.70). The difference in colour and apperance was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Body and texture

There was significant difference in body and texture score of different treatment combinations. Maximum body and texture score was recorded in the sample of T_0 (31.76) followed by T_1 (8.04) followed by T_0 (7.84), T_2 (7.44), T_3 (6.76). The difference in body and texture was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Flavor and taste

There was significant difference in flavor and taste score of different treatment combinations. Maximum flavor and taste score was recorded in the sample of T_0 (31.76) followed by T_1 (8.16) followed by T_0 (7.73), T_2 (7.54), T_3 (7.24). The difference in flavor and taste was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Overall acceptability

There was significant difference in overall acceptability score of different treatment combinations. Maximum overall acceptability was recorded in the sample of T_0 (31.76) followed by T_1 (8.09) followed by T_0 (7.69), T_2 (7.41), T_3 (6.85). The difference in overall acceptability was due to the composition difference of wheat flour, rice flour, corn starch and sago flour which are used in different propositions in different treatments.

Microbiological parameters

Yeast and mould count (cfu/gm)

The highest mean value for yeast and mould count of bread for control and treatment samples was recorded and was significantly viable as T_3 (2.20) followed by T_1 (1.60), T_2 (1.60) and the lowest is T_0 (1.40).

Coliform count

It is evident from the experiment that the coliform test of control and experimental sample was 100 percent negative.

Standard plate count

The highest mean value for standard plate count of bread for control and treatment samples was recorded and was significantly viable as T_3 (4.40) followed by T_1 (4.20), T_2 (4.00) and the lowest is T_0 (3.80).

Conclusion

The present experiment entitled "Development and Quality Assessment of Gluten-Free Bread Prepared by Using Rice flour, Corn Starch and Sago flour" was conducted for the preparation of bread using rice flour, corn starch and sago flour in the proportions, (40:35:25), (50:30:20), (60:25:15). Thus final product was analyzed for their Physico-chemical, Microbiological, sensory and cost. Our results show, therefore, that high- quality, gluten- free bread with good taste and appearance, suitable for celiac patients, can be made from a mixture of 3 raw materials normally consumed in other foods: cornstarch, rice flour, and sago flour. The indicative optimal formulation for this bread was (T_1) rice flour 40%, corn starch 35% and sago flour 25% and its validation will be made with a larger panel.

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