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Cereals And Pulse Based Ready-To-Cook Formulations Optimization And Microbial Profiling During Shelf-Life For Sustainable Food Innovations

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ABSTRACT

Cereals and pulses are inexpensive and sustainable sources of nutrients, minerals, and nutraceutical compounds. The deprived consumption of these cereals and pulses in native form is not successful. Hence development of ready-to-cook (RTC) blended formulations based on cereals and pulses will impede the use of these ingredients for balanced nutrition in our day-to-day foods. Accordingly in the present study, we selected five (5) different cereals and pulses, i.e., barley, finger millet, chickpeas, green gram, and horse gram, along with unripe banana, and arrowroot powder, for the development of RTC food formulations. Initially nine (9) combinations of RTC formulations were prepared and evaluated for its sensorial acceptance. Further, the optimized formulation was packed and stored at three different storage conditions (5°C, ambient room temperature, and accelerated (45°C)) for the evaluation of microbial load. Among the optimized formulations i.e., 80% base with 20% unripe banana (RTC-1) and 70% base with 15% each of unripe banana and arrow root (RTC-2) RTC formulations were highly accepted by the sensory panel members. The products stored in different conditions did not detect any microbial contamination even after 90 days storage. Hence, the optimized RTC formulations could be a low cost and shelf stable nutritional rich product for growing young and older populations in developing countries.

Key words- Cereals and Pulses, Unripe banana powder, Arrowroot, Ready-to-cook (RTC), Microbial analysis.

1. INTRODUCTION

Nowadays, the demand and desire for a nutritionally balanced cereal and pulse based blend combinations that are palatable, convenient and free from additives is on demanded (Banu et al., 2012). The whole grain based food products are healthy diet rich in proteins, dietary fibre, vitamins and minerals (Fardet, 2010). Ready-to-Cook (RTC) formulations, with long shelf life, especially devoid of microbial spoilage will help to maximum utilization cereals and pulses, in order to meet the nutritional needs of growing world population. (Food and Agriculture Organization [FAO], 2021). This paved many food industries to develop these convenience foods. RTC foods are those foods that are partially cooked and preserved long as a powder formulation or semi-solid material for convenience and to extend shelf-life. The development of such RTC foods using cereals and pulses is more demand to manage the modern lifetimes and nutrient requirements (Takhellambam et al., 2016).

Cereals and pulses are grains belonging to *Graminaceae* and *Fabaceae* or *Leguminosae*, respectively. They are excellent sources of carbohydrates, proteins, fiber, micronutrients, and bioactive compounds (Chaya et al., 2022). These cereal and pulses-based formulations could be a good alternative as nutrient sources for children below the age of 5 years (Bahwere et al., 2016). The regular consumption of cereals and pulses in specified quantities has many health beneficial potentials, reduces body weight, blood pressure, blood cholesterol, type-2 diabetes, obesity, and prevents colon cancer (Rebello et al., 2014; Bahwere et al., 2016).

The contamination of food products in various conditions along the production chain always is the point of concern (Rebello et al., 2014). The survival and growth of microorganisms in foods will be affected by the composition, processing conditions, packaging, and the storage of the finished products (temperature, time, light interference and stabilizers). Previous reports suggests that the food structure has been considered as the key factor for microbial contamination which determined the shelf-life of the products (Wilson et al., 2002).

Accordingly, in the present study, we selected five (5) different cereals and pulses such as barley, finger millet, chickpeas, green gram, horse gram and locally available unripe banana, and arrowroot powder for the development of RTC formulations (Chaya et al., 2022). Initially, different RTC blends were prepared with the above ingredients. Based on the sensory acceptability by the trained and untrained panel members, the optimized formulations were analyzed for their microbial analysis using standard methods in freshly prepared. Further, the optimized formulations were packed in aluminium silver foil pouches and RTC formulations were stored at three different storage conditions (5°C, ambient room temperature, and accelerated (45°C)) for 30 days, 60 days, and 90 days for quality evaluation of microbial load.

2. MATERIALS AND METHODS

2.1. Sources of the raw material:

The selected five (5) different cereals and pulses such as barley, finger millet, chickpeas, green gram, horse gram and locally available unripe banana, and arrowroot powder were procured from the local markets of Davangere, Karnataka, India. These selected cereals and pulses were cleaned and roasted on an ordinary household kitchen stove on a low flame until before turning light brown. Then they were allowed to come to room temperature and made into powder separately using a household mixer (Maharaja Whiteline Perfect W&R 500 Mixer grinder). The obtained powders were packed in an air-tight container in the dark until further use.

2.2. Chemicals:

The rose Bengal chloramphenicol agar, plate count agar, violet red bile broth, Baird Parker agar base, egg yolk tellurite emulsion and various other chemicals of analytical grade were obtained from Hi-Media (Bangalore, India).

2.3. Preparation of RTC food formulations:

Initially, nine (9) combinations of the RTC food formulations were prepared with different combinations of cereals, pulses, unripe banana, and arrow root powders based on their nutritional composition (Chaya et al., 2022). The RTC formulations were prepared based on the assumption that the formulations should serve from 6 to 70 year of age consumers with 25-50 g per day. During RTC preparation, a base was prepared using barley, finger millet, chickpeas, green gram, and horse gram, thoroughly mixing these flours. After the base preparation, the unripe banana powder and arrowroot powder were supplemented alone or in combinations with eight RTC food formulations with different percentages, and base preparation alone was kept as control (F-9).

2.4. Sensory analysis:

The trained and untrained sensory panel members screened the sensory profile of the prepared nine RTC formulations for their sensory acceptability (Kumar et al., 2020a). A quantitative descriptive analysis (QDA) comprised of a 15-point scale with different sensory parameters was screened to determine the best combination of RTC formulation using selected cereals, pulses, unripe banana, and arrowroot powder. Sensory evaluation was performed under ambient conditions by providing a glass of lukewarm water between the samples to cleanse the palate. The mean score of 30 members was considered to select the optimized combination of ingredients for RTC formulation preparations.

2.5. Packaging:

For shelf-life analysis, the above optimized RTC formulations were packed in leak-proof, waterproof, aluminium silver foil bags of 100 g each that could be used for both cold and hot food storage purpose. The packed RTC formulations were stored at three different storage conditions (5°C,

ambient room temperature, and accelerated (45°C)). The triplicate samples were withdrawn 30 days, 60 days, and 90 days for quality evaluation of microbial load.

2.6. Microbial analysis:

The microbial analysis of the prepared RTC formulations was determined using standard methods (Kumar et al., 2020a). A 10 g powder sample was dissolved in 100 mL of 0.9% saline. The aseptically serially diluted samples up to 10^{-4} (g/ml) were analyzed for total aerobic bacteria, coliforms, yeast, and moulds, respectively. For total aerobic bacterial count 100 µl of the sample was spread aseptically on plate containing plate count agar. Similarly, 100 µl of sample was spread on rose Bengal chloramphenicol agar medium for the yeast and mould counts. The *Staphylococcus* count was recorded in Baird parker agar medium supplemented with 5 ml of egg yolk and tellurite emulsion per 100 ml respectively. The *E. coli* count of the RTC formulations was determined using violet red bile agar medium. Triplicate plates were used for each dilution (10^{-1}, 10^{-2}, 10^{-3} and 10^{-4}). The colonies developed for bacterial counts were incubated at 37 °C for 24 and 48 h and the fungal count was recorded after 5 days of incubation at 28 °C. The results of microbial growth were expressed as log/g of sample.

2.7. Statistical analysis:

All the sensory screening data were presented as mean ± S.D of 25 trained and untrained panel members.

3. RESULTS AND DISCUSSION

3.1. Preparation of RTC food formulations:

The screening of different cereals, pulses, unripe banana, and arrowroot recorded that the barley (20%), finger millet (30%), chickpea (25%), green gram (12.5%), horse gram (12.5%) was prepared as base material and then different percentages of unripe banana (10-30%) and arrowroot (10-30%) were fortified. The prepared combinations were properly blended to even distribution of ingredients and to avoid formation of clumps. All the preparations were done in controlled and hygienic conditions to avoid microbial contaminations.

3.2. Sensory analysis:

The prepared nine combinations of RTC formulations with a base composition were prepared using cereals and pulses. To the base composition, different percentages of unripe banana and arrowroot powders were supplemented either single or in combination of both. Similarly, a control sample without unripe banana and arrowroot powders was maintained. Once prepared, the nine (9) RTC formulations were initially screened for sensory acceptability. Figure 1 shows the sensory profile of the nine combinations of RTC formulations for selecting the best combination of ingredients. Based on the highest sensory score obtained, the panel members suggested three varieties (RTC-2, RTC-4, and RTC-8) and to compare one control sample (RTC-9) selected for shelf-life analysis. Accordingly, a large batch of optimized RTC formulation was prepared such as RTC-2, RTC-4, and RTC-8

formulations which were labeled as RTC-1, RTC-2, RTC-3, and the control sample as RTC-4, respectively.

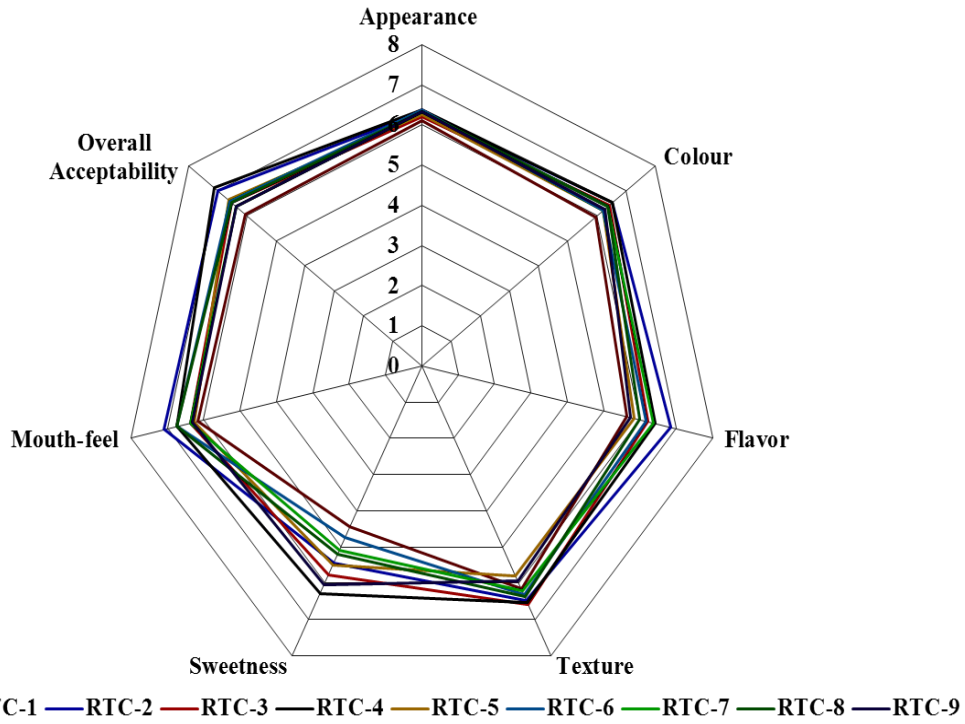

Figure 1- Sensory profile of the prepared different combinations of RTC food formulation

3.3. Packaging:

As the main concern of any food product is the microbial load, to avoid this prepared product was packed in hygiene condition. For shelf-life analysis, the above optimized RTC formulations were packed in leak-proof, waterproof, aluminium silver foil bags of 100 g each that could be used for both cold and hot food storage purpose. The packed RTC formulations were stored at three different storage conditions (5°C, ambient room temperature, and accelerated (45°C)). The triplicate samples were withdrawn 30 days, 60 days, and 90 days microbial safety.

3.4. Microbial analysis:

The microbial load of freshly prepared RTC formulation and shelf-life stored samples were evaluated to evaluate the safety of the prepared product for human consumption using standard methods (Kumar et al., 2020a). Table 1 and Table 2 show the viable microbial content of fresh and shelf-life stored samples. It is clearly recorded that there was no detectable microbial contamination of total aerobic bacteria, coliforms, yeast, and moulds in all the RTC samples analyzed. As the RTC formulations are dry with very less moisture content and processed hygienically, there was no observation of colonies. Kumar et al., (2020a) reported similar observations in betalains fortified food formulations such as spread, beverages, and jellies. In addition, similar observation of baby food flours with very less growth of microbial load after 90 days of storage in different packaging materials (paper, polyethene

and woven polypropylene bags) and protein rich cereal bar for 9 months of storage at RT and 37°C was reported (Forsido et al., 2021; Padmashree et al., 2012).

Table 1- Microbial analysis of the freshly prepared RTC food formulation

Sample	Total aerobic bacteria	Coliforms	Yeast and mould
RTC-1	Nil*	Nil*	Nil*
RTC-2	Nil*	Nil*	Nil*
RTC-3	Nil*	Nil*	Nil*
RTC-4	Nil*	Nil*	Nil*

All the values are mean ± SD of three replicates. *No colonies were observed in 10^{-1} dilution.

Table 2- Microbial analysis of the prepared RTC food formulation during shelf-life analysis at different temperature conditions

Sample	Storage Condition	30 days			60 days			90 days		
		Total aerobic bacteria	Coliforms	Yeast and mould	Total aerobic bacteria	Coliforms	Yeast and mould	Total aerobic bacteria	Coliforms	Yeast and mould
RTC-1	**5 °C**	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*
	RT	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*
	37 °C	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*
RTC-2	**5 °C**	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*
	RT	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*
	37 °C	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*	Nil*

All the values are mean ± SD of three replicates. *No colonies were observed in 10^{-1} dilution.

4. CONCLUSION

The present study concludes that the optimization of RTC formulations based on cereals and pulses that have good shelf-life stability that could meet the nutrient and loss of grains for the growing children and older people. Among the prepared combination with 20% unripe banana fortification, 10% fortified arrowroot formulation, and 15% each of unripe banana and arrowroot powder fortified

formulations were selected by the sensory panel members. The optimized formulation was prepared in large scale and packed in aluminium silver pouches which were recorded to be suitable for the prepared RTC food formulations. The shelf-life analysis proved that the products are safe up to 90 days under different storage conditions (5°C, ambient room temperature, and accelerated (45°C)) and required no additive or preservatives. Hence the RTC food formulations could be low cost sustainable foods for lower and middle income population to balance nutritional needs of the body to fight against malnutrition in developing countries.

5. CONFLICT OF INTEREST

The authors declare that they have no competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

6. ACKNOWLEDGMENTS

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