

Improvement of Bread Characteristics Using By-products from Indian Tropical Fruits

Thesis submitted

By

Debasmita Pathak

Doctor of Philosophy (Engineering)

Department of Food Technology & Bio-Chemical Engineering

Faculty Council of Engineering & Technology

Jadavpur University

Kolkata, India

2020

Dedicated

To

My Mother

- Title of the thesis: **Improvement of Bread Characteristics Using By-products from Indian Tropical Fruits**
- Submitted by: **DEBASMITA PATHAK**
- Date of Registration: **06.02.2013** (Index no. of Ph.D. registration: 130/13/E)
- Name, Designation and Institution of the supervisors:

1. RUNU CHAKRABORTY

Professor

Dept. of Food Technology & Bio-Chemical Engineering, Jadavpur University, Kol-700 032, India

2. UTPAL RAYCHAUDHURI

Professor (Retired)

Dept. of Food Technology & Bio-Chemical Engineering, Jadavpur University, Kol-700 032, India

- List of Publication:

❖ JOURNAL PUBLICATION:-

1. **Debasmita Pathak**, Jayshree Majumdar, Utpal Raychaudhuri, Runu Chakraborty, Study on enrichment of whole wheat bread quality with the incorporation of tropical fruit by-product. **International Food Research Journal**. 24(1): 238-246 (2017).
2. **Debasmita Pathak**, Jayshree Majumdar, Utpal Raychaudhuri, Runu Chakraborty, Characterization of physicochemical properties in whole wheat bread after incorporation of ripe mango peel. **Journal of Food Measurement and Characterization**. 10: 554-561 (2016).
3. **Debasmita Pathak**, Jayshree Majumdar, Utpal Raychaudhuri, Runu Chakraborty, Dynamic role of natural antioxidant sources on different parameters of bread quality - a review. **Nutrafoods**. 15: 163-178 (2016).

❖ CONFERENCE PROCEEDING:-

1. **Debasmita Pathak**, Utpal Raychaudhuri, Runu Chakraborty, A Study to Create Awareness towards Tropical Fruit Waste as a Source of Natural Antioxidant. Nutritional Deprivation in the Midst of Plenty (ISBN 978-93-83010-27-1). 109-111 (2016).

❖ ORAL PRESENTATION:-

1. Paper presented on 'Utilization of Raw and Ripe Mango Peel as a quality improver in Gluten Free Bread making', in International Conference on "5th

India Biodiversity Meet 2018” organized by Agricultural and Ecological Research Unit of Indian Statistical Institute, Kolkata, during 15th – 17th March, 2018.

2. Paper presented on ‘**Study on functional and rheological characteristics of gluten free flour and dough system using raw and ripe mango peel in combination**’ in **International Conference on Emerging Technologies in Agricultural and Food Engineering** organized by Dept. of Agricultural and Food Engineering, Indian Institute of Science Kharagpur, during 27th – 30th December, 2016.
3. Paper presented on ‘**A comparative study between raw and ripe mango seed kernel as a source of natural antioxidant**’, in **All India Seminar on “Post Harvest Management of Fruits and Vegetables”** organized by Agricultural Engineering Division, West Bengal State Centre, The Institution of Engineers, during 1st & 2nd June, 2016.
4. Paper presented on ‘**A study to create awareness towards tropical fruit waste as a source of natural antioxidant**’, in **National Seminar on Nutritional Deprivation in the Midst of Plenty – An Attempt to Create Awareness**, organized by Dept. of Food & Nutrition, Raidighi College in collaboration with Dept. of Botany, Raidighi College and Dept. of Microbiology, Sarsuna College, during 22nd & 23rd September, 2015.

❖ **POSTER PRESENTATION:-**

1. Poster presented on ‘**Optimization of ripe mango peel fortification level in bread using response surface methodology**’, in **International Conference on Recent Advances in Food Processing and Biotechnology**, organized by Centre of Food Science and Technology, Institute of Agricultural Sciences, Banaras Hindu University, during 5th & 6th April, 2016.
2. Poster presented on ‘**Characterization of ripe mango peel as a source of natural antioxidant**’, in **National Science Meet – 2016**, organized by Dept. of Food & Nutrition, Hiralal Mazumdar Memorial College for Women in collaboration with West Bengal State University, during 2nd & 3rd March, 2016.

❖ **WORKSHOP:-**

1. Participated in Workshop on ‘**Industrial Experiments for Engineering Students: WIEEST-2015/1**’ organized by Indian Statistical Institute, Kolkata during 17-19th February, 2016.

PROFORMA – 1

“Statement of Originality”

I, Debasmita Pathak, registered on 06/02/2013 do hereby declare that this thesis entitled “Improvement of Bread Characteristics Using By-products from Indian Tropical Fruits” contains literature survey and original research work done by the undersigned candidate as part of Doctoral studies.

All information in this thesis have been obtained and presented in accordance with existing academic rules and ethical conduct. I declare that, as required by these rules and conduct, I have fully cited and referred all materials and results that are not original to this work.

I also declare that I have checked this thesis as per the “Policy on Anti Plagiarism, Jadavpur University, 2019”, and the level of similarity as checked by iThenticate software is __10____%.

Signature of Candidate:

Debasmita Pathak
Date: 05/08/2020

Certified by Supervisor(s):

RUNU CHAKRABORTY

Professor

Dept. of Food Technology & Bio-Chemical

Engineering,

Jadavpur University, Kolkata, India

UTPAL RAYCHAUDHURI

Professor (Retired)

Dept. of Food Technology & Bio-Chemical

Engineering,

Jadavpur University, Kolkata, India

PROFORMA – 2

CERTIFICATE FROM THE SUPERVISOR/S

This is to certify that the thesis entitled “Improvement of Bread Characteristics Using By-products from Indian Tropical Fruits” submitted by Smt. Debasmita Pathak, who got her name registered on 06/02/2013 for the award of Ph. D. (Engg.) degree of Jadavpur University is absolutely based upon her own work under the supervision of Prof. Runu Chakraborty and Prof. Utpal Raychaudhuri (now retired) and that neither her thesis nor any part of the thesis has been submitted for any degree/diploma or any other academic award anywhere before.

RUNU CHAKRABORTY

Professor

Dept. of Food Technology & Bio-Chemical Engineering,

Jadavpur University, Kolkata, India

UTPAL RAYCHAUDHURI

Professor (Retired)

Dept. of Food Technology & Bio-Chemical Engineering,

Jadavpur University, Kolkata, India

ACKNOWLEDGEMENT

It is my proud privilege to express my deep sense of gratitude to my research supervisors, Prof. Runu Chakraborty and Prof. Utpal Raychaudhuri (now retired), Department of Food Technology and Bio-Chemical Engineering, Jadavpur University, for their valuable advice, guidance, unconditional love, unending zeal and continuous support during the course of investigation without which it would not have been possible for me to complete this work. I have learnt my scientific aspects and honed my research skills through their continuous encouragement and support.

I express my sincere respect and thanks to all other teachers Prof. Late Lalita Gouri Ray, Prof. Uma Ghosh, Dr. Dipankar Halder, Dr. Paramita Bhattacharjee, Dr. Debabrata Bera, Dr. Sunita Adhikari (Nee Pramanik), and Dr. Prasanta Kumar Biswas for providing encouragement during the project.

I am very much thankful to my seniors and labmates Dr. Jayeeta Bardhan, Dr. Arpita Das, Dr. Koustav Chakraborty, Dr. Sohini Ray, Mrs. Runni Mukherjee, Mr. Suman Kumar Saha, and Ms. Najmun Nahar, for maintaining a lively atmosphere and providing valuable support during the total research period. Furthermore my heartfelt thanks to M. Tech students Jayshree Majumdar and Payel Dasgupta who played a great role in my research project.

I also wish to thank all staff members and research scholars of the Department of Food Technology and Bio-Chemical Engineering, Jadavpur University for their endless help and encouragement during the work.

I would also like to acknowledge the moral support, blessings, affection provided by my parents and my family all through my research work.

Above all I express my gratefulness to the Almighty for making me able to achieve whatever I have.

Date: 05/08/2020

Debasmita Pathak

INDEX

<u>Content</u>	<u>Page no</u>
• Abstract	1 - 2
• <u>Chapter 1</u>	
❖ Background of the work: Dynamic role of natural antioxidant sources on different quality characteristics of bread	3 - 21
❖ Aims and Objectives	22
• <u>Chapter 2</u>	
Incorporation of tropical fruit by-product in whole wheat bread as a quality improver	23 – 35
• <u>Chapter 3</u>	
Optimisation of ripe mango peel enriched white bread preparation and its characterisation	36 – 49
• <u>Chapter 4</u>	
Combined effect of Raw and Ripe Mango Peel Powder on the quality characteristics of Gluten Free Bread	50 – 62
• Future Scope	63 – 64
• References	65 - 76

LIST OF TABLES

No. of Tables	Name of Tables	Page no.
1.1	Effect of natural antioxidant sources on different quality parameters of bread	10-12
2.1	Descriptors and definitions used for sensory profiling of RMPP added bread samples	28
2.2	Proximate composition of RMPP (g/100g dry sample, except for moisture)	30
2.3	Antioxidant properties of bread samples substituted with RMPP at different levels	32
2.4	Sensory evaluation of the bread samples substituted with RMPP at different levels	33
2.5	Physical parameters of bread samples substituted with RMPP at different levels	34
2.6	Colour parameters of bread samples substituted with RMPP at different levels	34
3.1	Face-centered central composite design with results of specific volume and DPPH radical scavenging activity of bread samples	41
3.2.(a)	Analysis of variance table for specific volume	42
3.2.(b)	Analysis of variance table for DPPH radical scavenging activity	43
3.3.	Experimental and predicted values of the responses at optimised conditions	46
3.4 (a)	Comparison between control and optimised bread	48-49
3.4 (b)		
4.1.	Physicochemical characteristics of different flour samples	56
4.2.	Functional characteristics of different flour samples	57
4.3	Physical parameters of MPP added Gluten free bread	59
4.4.	Antioxidant activity of MPP added gluten free bread	60
4.5	Sensory parameters of MPP added gluten free bread	61

LIST OF FIGURES

No. of Figures	Name of Figures	Page no.
2.1.	Image of Ripe Mango Peel Powder (RMPP)	25
2.2.	HPLC chromatogram of RMPP	31
3.1(a).	Response surface plots showing the effect of process parameters on specific volume	44
3.1(b).	Response surface plots showing the effect of process parameters on DPPH radical scavenging activity	45
3.2.	Desirability ramp for optimization	46
3.3.	Cross sectional images of bread loaves	47
4.1.	Viscoelastic properties of different types of GFB samples	57
4.2.	Images of Gluten Free Bread (GFB)	61

Abstract

Improved understanding of the relationship between the consumption of antioxidant-rich food and human health has resulted in the supplementation of food products with natural antioxidants. Enrichment of a staple food such as bread with natural antioxidants can provide additional health benefits. Sources of natural antioxidants include cereals, seeds, spices, herbs, green plants, fruit, vegetables and the less expensive and easily available waste products from the food industry. However, there are technological constraints to the fortification of bread, while sensory and physical properties are important for overall consumer acceptability. Synthetic antioxidants can have serious adverse effects on health, and their substitution with natural antioxidants enhances the therapeutic and functional properties of bread. This review (**Chapter: 1**) discusses the antioxidant activity of various types of breads enriched with natural antioxidants from natural sources and the associated physical and sensory attributes of these breads.

In the present study (**Chapter: 2**), the effect of ripe mango peel powder (RMPP) at 1, 3, and 5% on physical, antioxidant and sensory properties of whole wheat bread were evaluated. Breads incorporated with RMPP exhibited an improvement in antioxidant properties as total phenolics content increased from $(220.33 \pm 8.5$ to $757.8 \pm 13.5)$ mg GAE/100 gm along with the highest value of 2,2-diphenyl-1-picrylhydrazyl (DPPH) inhibition was 65.74 ± 0.51 %. The RMPP at 5% level also improve the fruity aroma and fruity taste in descriptive analysis. Both antioxidant activity and flavour were improved gradually with the addition of RMPP, so 5% can be considered as best addition level. Again at 5% RMPP showed maximum negative effect on bread texture. Loaf height, weight loss percentage and specific volume were decreased with the amount of RMPP incorporation. Bread density along with crumb moisture was increased after addition of RMPP. The brownness index of bread samples was significantly different and whiteness index were also decremented. For textural analysis hardness were also increased with level of addition. Though, in comparison with 1%, 3% showed lower effect for selected parameters. Thus, 3 % RMPP addition in whole wheat bread was regarded as best.

Ripe mango peel powder enriched white bread was prepared (**Chapter: 3**) by using a three-level three-factor face-centered central composite design (FCCD). Levels of independent variables, baking temperature (X_1), baking time (X_2) and RMPP addition level (X_3), were selected using single factor analysis to study their effect on specific

volume (Y_1 ; cm^3/g) and DPPH radical scavenging activity (Y_2 ; $\mu\text{mol AAE}/\text{g}$) of bread samples. The -1, 0 and +1 levels were fixed at 190, 210 and 230 for baking temperature ($^{\circ}\text{C}$), 15, 30 and 45 for baking time (min) and 3, 5 and 7 for RMPP addition level (%). For specific volume significantly negative linear effect by RMPP addition level and negative interaction effect by baking temperature and time was observed. DPPH radical scavenging activity was significantly affected by RMPP as linear factor (positive term) and baking temperature with time (negative term) and RMPP with baking time (positive term) as interaction factor. ANOVA results for both models indicate the goodness of fit and suitability of the regression model. Baking at 226°C for 19 min with 7% RMPP was selected as optimal condition with $2.500 \text{ cm}^3/\text{g}$ specific volume and $25.868 \mu\text{mol AAE}/\text{g}$ DPPH radical scavenging activity. Predicted value was confirmed through validation process. In comparison with control bread similar overall acceptability was observed for optimised bread. Also optimised bread showed delayed staling than control bread.

Both raw and ripe mango peel is the major by-product of mango processing industry and is a rich source of natural bioactive compounds that is very good for health. In the study (**Chapter: 4**) main aim was to evaluate flour quality along with the rheology of formulated gluten free dough also good texture with antioxidant activity and good consumer acceptability of the mango peel powder (MPP) added bread samples where both raw MPP and ripe MPP were mixed equally at different levels (2%, 4% and 6%). Flour samples with 4% MPP showed acceptable physical and functional characteristics. In rheology small change was observed for 4% level in comparison with 2% level but a marked increase in visco-elasticity was reported when 4% formulation was compared with control and 6% formulated samples. The phenolic and flavonoid content ranges from (0.18-1.85) mgGAE/g and (0.08-0.14) mgCE/g respectively. The antioxidant activity also increased on increasing level of MPP incorporation. The physical parameters (moisture, height, specific volume, density, crumb hardness, crust and crumb color) showed the best acceptable results on incorporation of 4% level MPP which also satisfies all the sensory attributes and was acceptable by the panelists. Thus, the overall result illustrates the importance of MPP incorporation in the sorghum-rice formulation yielding high antioxidant properties with desired physical and sensory attributes.

CHAPTER: 1

Background of the work

1. Dynamic role of natural antioxidant sources on different quality characteristics of bread

1.1. Introduction:

In biological systems, oxidative stress is produced by an imbalance between chemical stresses, caused by abnormal quantities of reactive oxygen species (ROS), and physical ability to detoxify the reactive intermediates. Oxidative stress if prolonged can damage the entire system, and has been implicated in over 100 common diseases such as cancer, diabetes, heart conditions, and neurological disease [1]. However, antioxidants can scavenge ROS and free radicals, thus reducing their numbers. The growing interest in foods rich in antioxidants has led to the development of a large market for antioxidant-rich products and ingredients.

The importance of natural antioxidants for food applications has been highlighted in numerous studies. Natural antioxidants are found in plants which can be consumed. For example, spices, herbs and essential oils are rich in antioxidant properties [2]. The consumption of natural antioxidants, including vitamins, enzymes, carotenoids, chelators, and phenolic compounds, have received considerable interest because of their presumed safety and potential nutritional and therapeutic effects, including anti-inflammatory, anti-carcinogenic and anti-atherosclerotic properties. Artificial antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) are synthetically produced chemical antioxidants, which are widely used for their antioxidant properties in the food industry. However, the use of these substances is restricted due to toxicity problems and the fear of liver damage and carcinogenesis [3]. Consequently, there is ongoing research on natural sources of antioxidants.

Bread, a low cost easily available energy source, has been a staple food for centuries. Main ingredients of bread preparations are wheat flour, yeast, sugar, salt, and water. The wheat grain contains many bioactive compounds, including antioxidant activity rich phenolic compounds mainly present in the bran and aleurone layer, although the processing of wheat bread also confers some antioxidant activity [4]. Many different synthetic antioxidants have been used in bread for years to increase storage stability. The

rapidly growing functional food and nutraceutical market in recent times has stimulated research on the use of natural antioxidants in bread which do not affect its textural and organoleptic properties. Functional and therapeutic properties, basic nutritional quality, sensory and textural properties must be carefully considered when incorporating various sources of antioxidants into bread. Recent studies have shown that the addition of small amounts of natural antioxidant sources does not significantly affect the textural and sensory properties of bread.

1.2. Literature review:

Many consumers prefer to eat healthier food to prevent non-communicable diseases. Consequently, researchers are seeking to work on improvement of bread quality through the optimisation process. Accordingly, cereals and their fractions, fruits and vegetables, gums and starches and agricultural wastes have been used to produce antioxidant-rich bread. Thus, this study aimed to review the antioxidant-rich bread, as well as the impact of fortification on both physical and sensory parameters are summarised in Table 1.1.

Black tea: Chinese steamed bread (CSB) was fortified with black tea with various teas to water (v/v) ratios [5]. At higher concentration of black tea, lightness (L) of CSB was decreased while redness (a) and yellowness (b) were increased, due to the presence of black tea polyphenols. Along with antioxidant activity, phenolic compounds of black tea improved the antimicrobial properties of CSB, with the best results seen at 5:0 ratio of tea to water. There were insignificant changes in the water activity, water content, texture, and volumetric properties of fortified CSB. In sensory evaluation black tea showed the addition of black tea had little effect on brightness, crumb structure, elasticity and stickiness but resulted in decreased smoothness and whiteness.

Rice bran: Stabilised rice bran (RB) was added at different percentages to wheat bread [6]. All substituted bread showed higher protein, ash, fat and total dietary fibre contents than control bread. Inclusion of RB negatively affected crumb colour (L*), bread volume and sensory parameters such as colour, texture and overall acceptability, but increased hardness, aroma, taste, a* and b* values. The highest and lowest values were seen in 30% RB substituted bread. Vitamin E content as indicated by tocopherol and tocotrienol content increased in proportion to the amount of RB. The addition of RB also increased the free and bound total phenolic content (TPC), total flavonoid content (TFC), ferulic acid, p-coumaric acid, sinapic acid, and ferric reducing antioxidant power (FRAP) and

2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging values compared to control bread, with the highest values seen in 30% RB substituted bread.

Quinoa leaves: Bread was fortified with different amounts of quinoa leaves (QL) as a source of antioxidants [7]. An increase in the percentage of QL resulted in bread with decreased loaf volume and higher bread crumb hardness, cohesiveness and chewiness/gumminess, with an insignificant effect on elasticity. Bread fortification with QL showed a linear increase in antioxidant properties and TPC and had a positive influence on FRAP, inhibition of linoleic acid peroxidation and metal chelating power. The maximum antiradical potential of bread was obtained with 5% QL supplementation.

Nejayote solids: Nejayote, maize-processing wastewater, in the form of solids was used to enhance antioxidant activity in bread [8]. Application of nejayote solids (NS) at different addition level significantly enhanced water absorption capacity, loaf weight and apparent density with slight decreased bread volume. On the other hand initial mixing time, both proof heights and bread heights also the texture of the bread loaf did not show any significant changes. Bread supplemented with NS showed higher dietary fibre, crude fibre and bioavailable calcium: from two slices (64 g) of bread with 9% NS provided calcium which is 29% of the recommended daily intake. Also with increased NS levels ferulic acid was increased proportionally and significantly improved antioxidant activity. Flavour and odour received low sensory scores, while overall acceptability, texture and colour were not affected by 9% NS substitution as compared to control.

Broccoli sprout powder: Bread was enriched with various proportions of broccoli sprout (BS) powder for its antioxidant and anti-carcinogenic activity [9]. Partial replacement of flour with BS powder had a strong negative effect on aroma, texture, taste, and overall acceptability and a slightly negative effect on colour. However, BS significantly enhanced the phenolic content of the bread, with the highest values seen with 2% substitution, although a linear relationship between increased phenolic content and the percentage of added BS was not seen. Fortification increased the level of free amino groups and also influenced the antiradical potential, with highest values seen at 4% BS. Reducing power was increased than control, where it was substantially highest at the lower level (2% BS) compared to the higher level of incorporation. The highest chelating activity and inhibition of lipid peroxidation were seen in bread with 4% and 3% BS respectively. Bread with 2% BS supplementation was determined to be best

regarding consumer acceptability, antioxidant activity and ability to inhibit prooxidative enzymes.

Onion skin powder: Bread supplemented with dry onion skin (OS) powder as a source of antioxidants showed an increase in quercetin concentration, scavenging activity, chelating power, lipid peroxidation, and reducing power compared to control bread [10]. As expected, the antioxidant properties (except for lipid peroxidation) of OS-fortified bread and overall acceptability were positively correlated with the addition of up to 2–3% OS. However, bread supplemented with 4–5% OS was less acceptable due to no further increase in antioxidant activity and the presence of excessive amounts of volatile compounds.

Chestnut flour: Chestnut flour (CF) a good source of phenolic compounds and fatty acids were incorporated into bread at different amounts [11]. Bread with up to 50% CF showed a significant linear increase in antioxidant activity and level of volatiles, including alcohols followed by aldehydes, ketones and furans. The levels of compounds present in CF, such as furans including furfural, 5-methyl furfural and 2-furfuryl alcohol, increased strongly as the amount of substitution increased. CF imparted nutty and smoky flavours due to the presence of guaiacol and 4-ethyl-guaiacol, demonstrating a positive effect on organoleptic properties. Bread made with 50% CF showed a homogeneous crumb structure, similar crumb hardness, cohesiveness and a darker colour compared to control bread.

Banana pseudo-stem flour: A fixed amount of banana pseudo-stem flour (BPF) was used with xanthan gum (XG) or carboxymethylcellulose (CMC) as a source of antioxidants in bread [12]. Proximate analyses showed that BPF bread had a high content of total dietary fibre, with a balanced soluble dietary fibre to insoluble dietary fibre ratio. The addition of BPF increased the phenolic content and free radical scavenging activity compared to control. However, supplementation with XG or CMC in bread containing BPF resulted in a significant decrease in phenolic content and free radical scavenging activity, whereas physical analyses indicated that supplementation with 10% BPF with XG reduced loaf volume, weight loss, height and higher density than control and CMC added sample. While all bread containing BPF had a darker crumb and lighter crust colour, mouthfeel, aroma, gumminess and taste perceptions were not affected. All bread received better acceptability scores than control bread, with the highest overall acceptability score seen for bread with added CMC.

Fennel seed powder: Various amounts of fennel seed (FS) powder were incorporated into bread as a source of antioxidants [13]. Supplementation significantly affected physical properties, with a negative effect on texture and decreased loaf volume and specific loaf volume, but with an insignificant effect on loaf mass. With increasing addition of FS, the redness (a^*) and yellowness (b^*) of the bread crumb significantly increased, while the lightness (L^*) significantly decreased. The appearance of the bread declined slightly, with the optimum taste and aroma scores obtained at 7–10% FS supplementation, while the best colour was achieved with 7% FS supplementation. A linear increase in TPC and FRAP values (but not DPPH radical scavenging activity which showed a reduction at 5% FS incorporation) was seen with increasing supplementation up to 7.0% FS, with further additions having little effect on antioxidant properties. Thus, supplementation with 5.0% and 7.0% FS powder was found to be optimum.

Cinnamon powder: Different amounts of cinnamon powder (CP), rich in fibre, fats, ash, with a high phenol content and antioxidant activity, were added to the bread [14]. Baking absorption and loaf weight increased linearly with the amount of CP added. Loaf volume and height increased up to 2% CP supplementation and decreased thereafter, while specific volume remained either equal to or less than that of control bread. Appearance and colour scores were high at 2% and 3% CP addition, with flavour, texture and overall acceptability scores highest at 2% CP incorporation. The hardness, gumminess and chewiness of the bread significantly increased above 2% CP incorporation. Besides, an adequate microbial inhibitory effect was observed due to the presence of cinnamaldehyde and eugenol at 2% CP incorporation. Fibre, ash, protein and fat content, phenolic content and antioxidant activity showed a significant linear increase as the level of supplementation increased with the highest values obtained at 4% CP incorporation.

Intermediated pearled wheat fraction: Bread was enriched with different amounts of pearled fractions from intermediate layers of wheat kernel as a source of antioxidants [15]. The L^* , a^* and hue values (h) of the crust of supplemented bread significantly increased with a low difference in chroma (C^*) values compared to control. Crust crunchiness, total break energy and acoustic emissions were highest in the bread containing 0% and 5% pearled fractions with lowest values seen at 20–25% incorporation. There was a decrease in bread volume with a proportional increase in gumminess, chewiness and hardness accompanied by an increase in the percentage of

pearled wheat fractions, while cohesiveness and resilience remained unaffected. The protein, dietary fibre, β -glucan and ash content along with the total antioxidant activity (measured as DPPH scavenging capacity) and TPC of bread increased linearly and significantly as supplementation increased, with maximum values seen at 25% substitution. However, the alkylresorcinol content was lower than expected, while the deoxynivalenol content increased linearly, indicating 10% substitution was the best level of supplementation.

Micronized by-products from debranned durum wheat: Different amounts of coarse and fine micronized bran fractions were incorporated both into sourdough and baker's yeast dough to make antioxidant-rich bread [16]. The intermediate and internal layers of durum wheat contain high amounts of protein, fat, starch and free amino acids and have a high peroxide value, while the external fraction has a high total dietary fibre content. However, the concentration of total free amino acids gradually increased with the addition of bran fraction and fermentation of the dough. Supplementation and dough fermentation increased total phenolic content, with sourdough wheat flour bread supplemented with 5% coarse fraction having the highest values. The hardness and fracturability of bread baked at 220°C and enriched with bran fractions were lower than those of control. Bran fractions intensified crust colour, especially with sourdough fermentation, with increased sweetness and overall perception of taste, while elasticity and dryness were unchanged. Sourdough bread showed more intense sensory characteristics, with higher acid flavour, acid taste and salty attributes.

Sorghum flour: Varying amounts of whole grain white sorghum flour (WSF) and red sorghum flour (RSF), which both provide total dietary fibre, were incorporated into flatbread as a source of antioxidants [17]. The total phenolic content and antioxidant capacity of red sorghum flatbread were significantly higher than that of white sorghum flatbread, which in turn was significantly higher than that of control bread. RSF bread had a higher content of bound phenolics and free phenolics than WSF bread or control bread, resulting in increased TPC. RSF flatbreads were darker in colour than controls, possibly due to the presence of higher levels of coloured polyphenolic compounds such as anthocyanins. Bread supplemented with 30% WSF and 40% RSF had the highest scores for sensory parameters like flavour and texture and the best overall acceptability scores.

Table1.1: Effect of natural antioxidant sources on different quality parameters of bread

Source of fortification	Antioxidant activity (assay value (highest shown))						Effect on physical properties	Effect on sensory evaluation	
	TPC	DPPH	FRAP	Others					
Black tea [5]	24.9±0.2 (GAE mg/g sample)		18.8±1.2 (Fe(II) mM/g sample)	ABTS 58.4±0.2 (percentage of inhibition)		Total flavonoid content 640.1±6.0 (QE µg/g sample)	Less effect on textural and other physical properties with darker colour	Overall good acceptance	
Rice bran [6]	183.9±2.9 free phenolics 110.4±8.3 bound phenolics (µg/g dw)	400 (mg TE/100 g dw) (total phenolics) (approx.)	400 (mg TE/100 g dw) (total phenolics) (approx.)	Total flavonoid content 76.5±0.6 (free) 34.4±3.4 (bound) (mg CATE/100 g dw)	Ferulic acid 4.2±0.6 (free) 285.1±8.3 (bound) (mg/g dw)	Sinapic acid 0.5±0.1 (free) 14.0±2.1 (bound) (mg/g dw)	<i>p</i> -Coumaric acid 2.1 ± 0.5 (free) 76.8 ± 4.0 (bound) (mg/g dw)	Lower volume with increased firmness	Low overall acceptability
Quinoa leaves [7]	2.90±0.05		6.07±0.15	ABTS 5.14±0.14 (scavenging %)	LPO 6.04±0.20 (scavenging %)	CHP 2.52±0.20 (% inhibition)	Linear increase in crumb hardness, cohesiveness and gumminess		
Nejayote solids [8]				Ferulic acid 14.16 (µg/100 g)		ORAC 2.32 (µM Trolox/100 g)	Very low alteration	Up to 9% supplementation without affecting sensory parameters	
Broccoli sprout powder [9]	2.0 (mg GAE/g dw) (approx.)		24.43±0.94 (EC ₅₀ mg dw/ml)	ABTS 65.64±2.63 (EC ₅₀ mg dw/ml)	LPO 11.91±0.42 (EC ₅₀ mg dw/ml)	CHP 25.86 ± 0.75 (EC ₅₀ mg dw/ml)		Satisfactory overall consumer acceptability up to 2% supplementation	
Onion skin powder [10]		17.03±2.01 (EC ₅₀ mg dw/ml)		ABTS 57.71±1.98 (EC ₅₀ mg dw/ml)	LPO 10.95±0.58 (EC ₅₀ mg dw/ml)	CHP 16.29±0.33 (EC ₅₀ mg dw/ml)		Satisfactory acceptability up to 3% supplementation	
Chestnut flour [11]		1.04 µmol (Trolox eq./g of dw)					Homogeneous crumb structure		
Banana pseudo-stem flour [12]	204.16±3.04 (mg GAE/100 g of sample)	241.39±23.06 (µmol TEAC/100 g of sample)	849.01±32.46 (µmol Fe(II)/100 g of sample)				Better physical characteristics with CMC	Higher acceptability with CMC	

Source of fortification	Antioxidant activity (assay value (highest shown))				Effect on physical properties	Effect on sensory evaluation
	TPC	DPPH	FRAP	Others		
Fennel seed powder [13]	25-30 (mg GAE/100 g)	80-100 (DPPH % inhibition)	2 (mg FeSO ₄ /100 g)		Crumb moisture and firmness increased	Good consumer acceptability up to 7% supplementation
Cinnamon powder [14]	0.94 (mg GAE/g dw)	27.67 (DPPH % inhibition)			Similar / better baking quality up to 2% addition	Maximum overall acceptability at 2% supplementation
Intermediated pearled wheat fraction [15]	3066 (mg/kg)	0.73 (mmol TE/kg)			Acceptable rheological and technological properties	
Micronized by-products from debranned durum wheat [16]	0.95±0.03 (mM GAE/g)	78.2±0.2 (inhibition %)			A better bread volume & hardness	Enhanced sensory properties of breads
Sorghum flour [17]	2.02±0.09 (mg GAE/g sample)	9.45±0.45 (µmol TE/g)			Observed differences in dough farinograph properties	No reduction in sensory acceptability
Coriander leaf powder [18]	30-35 (mg GAE/100 g of sample)	8-9 (DPPH % inhibition)	2.4-2.8 (mg FeSO ₄ /100 g)		Affected physical parameters, increase in crumb firmness and loaf weight	Highest overall acceptability at high percentage of supplementation
Ginger powder [19]	0.485±0.035 (mg GAE/g dw)	0.151±0.006 (µmol DPPH /mg dw ml ⁻¹)			Rheology of dough and texture of bread affected	Negative effect on bread acceptability
Turmeric powder [20]	150.5 (mg GAE/100 g)			β-Carotene-linoleate bleaching assay 79.8±0.6 % antioxidant activity	Hardness and crumb colour a* and b* values increased	Satisfactory overall consumer acceptability up to 4% supplementation
Wholegrain flour and fibres [21]	566 (µg/g) (bound) 168 (µg/g) (free)	80-90 (% inhibition) (bound) 30-40 (% inhibition) (free)			Reduced bread volume	Overall quality was in acceptable range
Anka (fermented rice) [22]	7.78±0.03 (EC ₅₀ mg extract /ml)	4.29±0.04 (EC ₅₀ mg extract/ml)			Decrease in specific volume	No significant differences with better colour and mouth feel
Grape by-product [23]	613.77±1.57 (mg GAE/10 g sample)	15.02 (mM TRE/g dm)	48.1 (mM Fe ^(II) /g dm)		Affected hardness, gumminess	Acceptable products up to 6% addition level

Source of fortification	Antioxidant activity (assay) value (highest shown)				Effect on physical properties	Effect on sensory evaluation	
	TPC	DPPH	FRAP	Others			
Pseudocereal flour [24]	2.65±0.1 (mg GAE/g dw)	7.53±0.18 (mmol Trolox/kg dw)	150.8±2.2 (mg Trolox/100g dw)			Buckwheat bread was more acceptable	
Grape seed extract [25]				ABTS (4-5) (nmol trolox/mg bread sample)	55% inhibition of the formation of CML in bread crust	No significant difference in hardness	Little effect on quality attributes
Sugar beet molasses based ingredients [26]		1677.3 (µmol TE/100 g)				Significant effect on specific volume	No interference at lower levels
Barley flour [27]	280 (mg GAE/100 g dm)		2.5 (mmol Fe ³⁺ /100 g dm)				Good consistency between sensory evaluation and amount of phenolics
<i>Auricularia auricula</i> flour [28]		(40-60) (DPPH % inhibition)				Little effect on loaf weight, loaf height and loaf volume up to 9% supplementation	No significant effect except at very high supplementation levels
Yam flour [29]		40 (DPPH % inhibition) (approx.)		ABTS 40 (total antioxidant capacity %) (approx.)		No significant effect at low levels of supplementation	No interference with bread acceptability

ABTS-2,20-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) radical scavenging activity, CHP-metal chelating activity, CMC- carboxymethylcellulose, CML-carboxymethyl-lysine, dm-dry matter, 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity, dw-dry weight, FRAP-feric reducing antioxidant power, LPO-inhibition of lipid peroxidation, ORAC-oxygen radical absorbance capacity, TEAC-Troloxequivalent antioxidant capacity, TPC-total phenolic content

Coriander leaf powder: Varying amounts of coriander leaf powder (CLP) were used in bread as an antioxidant source [18]. Incorporation of dried CLP enhanced moisture retention capacity and baking characteristics, showing higher crumb moisture up to 5% CLP supplementation and a slower staling rate. Physical parameters like crumb firmness and loaf weight increased, while specific volume and loaf volume values decreased linearly with increased supplementation. Similarly, as supplementation increased, the L*, a* and b* values of the crust decreased, while crumb L* values decreased and a* values increased, indicating a darker colour. With the level of supplementation TPC, DPPH radical scavenging activity and FRAP assay results showed a linear improvement. The

aroma acceptance scores gradually increased, while colour and taste were less acceptable with high CLP supplementation. Consequently, the overall acceptability of 3% and 5% CLP-substituted bread samples were much better than those with higher levels of supplementation.

Ginger powder: Different amounts of ginger powder (GP) were used to obtain antioxidant- rich bread [19]. The addition of GP enhanced the elasticity of the dough. Hardness and gumminess were significantly higher with 6% GP supplementation although changes in dough rheological properties were insignificant up to 4.5% GP supplementation. The bread had a finer and uniform crumb. A higher percentage of ginger lowered the L* and h values and elevated the C* value of crumb. The number of phenolics and radical scavenging activity increased in proportion with increased GP content of both crumb and crust, but to a great extent in the crust. Addition of the lowest percentage of GP (3%) in bread obtained maximum overall acceptability because shogaol was present at least amount.

Turmeric powder: Varying amounts of turmeric powder (TP) were used as a source of antioxidants in wheat bread [20]. Despite a loss of 32–54% of TPC after baking, the bread had a significantly higher phenolic content due to the presence of curcumin. Scavenging activity, thermal stability and antioxidant properties as measured by β -carotene-linoleate bleaching assays improved as supplementation was increased to 2–8% TP compared with control bread. The taste and overall acceptability of bread with turmeric powder were best at 0–4% TP supplementation, while aroma and texture did not vary. The best crumb colour was seen at 2% TP supplementation.

Wholegrain flour and fibres: Wheat, rye, barley and oat whole grain flours and cellulose (insoluble fibre) and xanthan gum (soluble fibre) in different proportions were added to wheat bread [21]. Bread containing oats had the highest levels of free phenolics, while rye bread had the highest level of bound phenolics and the best scavenging capacity. The antioxidant properties of bread containing cellulose and xanthan gum were very similar to those of control bread, indicating that inclusion of fibre does not affect the antioxidant properties of high fibre bread. Incorporation of wholegrain and fibre increased total dietary fibre, while the addition of whole wheat flour increased protein content. In general, there was a marked reduction in the average specific volume. The overall quality of wholegrain rich bread was acceptable except for that with added cellulose.

Anka (fermented rice): Monascus-fermented rice (Anka) was incorporated into bread to enhance its antioxidant properties [22]. Bread enriched with Anka flour had a slightly lower specific volume and fewer carbohydrates but higher amounts of reducing sugars, fat and fibre than rice and wheat bread. Anka-enriched bread had an attractive red colour with lower L* and whiteness index (WI) values and higher b* and a* values than wheat and rice bread. Antioxidant activity of both white bread and anka-enriched bread showed a comparable value with more effectiveness in scavenging activity and reducing power as measured by the inhibition of DPPH radicals as EC₅₀ values. Functional compounds like GABA and monacolin K were found in substantial amounts in Anka-enriched bread, which slightly decreased during baking. Anka-enriched bread obtained higher value for colour and mouth feel, but there was no difference in its appearance, flavour and overall sensory attributes compared to white bread.

Grape by-product: Grape by-products (GP) containing very large amounts of ash and dietary fibre were incorporated in different amounts to enhance the antioxidant properties of rye bread [23]. As GP level increased, phenolic compound together with free radical scavenging activity (DPPH) and reducing activity (FRAP) significantly increased. The phenolic compounds responsible for the improved antioxidant properties of GP-enriched bread were catechins, procyanidins, quercetin and also quercetin-3-β-D-glucoside. Significant increase for both hardness and gumminess of bread were observed with increasing level of GP, while insignificant changes were seen in cohesiveness, resilience, chewiness and springiness up to 8% GP addition. In sensory evaluation bread showed lower value for volume, porosity and overall acceptability with higher level of GP. Differences in aroma attributes were due to dominant fruity, alcoholic and sharp notes. However, different types of volatile esters and carbonyl compounds increased gradually.

Pseudocereal flour: Varying amounts of different pseudocereal (buckwheat, amaranth and quinoa) flours were used in bread to improve antioxidant activity [24]. The addition of buckwheat flour improved antioxidant activity more effectively than amaranth or quinoa flour. Bread supplemented with 30% buckwheat flour had the highest phenolic content, Trolox equivalent antioxidant capacity (TEAC) values as evaluated by FRAP and DPPH, while bread supplemented with 30% amaranth flour showed the highest flavonoid content. Organoleptic evaluation of pseudocereal-substituted bread indicated they were moderately acceptable. Buckwheat bread had better sensory profile than amaranth and quinoa bread with higher scores for colour and odour.

Grape seed extract: Grape seed extract (GSE) added bread linearly enhanced antioxidant activity with increased GSE [25]. However, baking degraded GSE proanthocyanidins, thus lowering GSE antioxidant capacity. Hardness of the bread samples was insignificantly different. Bread colour was affected by GSE as lightness and colour intensity was decreased whereas redness and yellowness values increased. Sweetness, porosity, astringency, and stickiness were not affected by GSE but in comparison with control bread colour was improved. The catechins and proanthocyanidins in GSE greatly enhanced the antioxidant activity of bread. The abundant phenolics in GSE may help to reduce the health risks associated with Nε-(carboxymethyl) lysine (CML).

Sugar beet molasses-based ingredients: Fruit and vegetable powders obtained with osmotic dehydration (OD) in beet molasses and beet molasses without any processing were used in white bread to increase its antioxidant properties [26]. In comparison with OD fruit powders, vegetable powders contained significantly more protein, crude fibre, reducing sugars, and saccharose with minerals like Ca, Mg, K and Na. Bread supplemented with 10% OD red cabbage powder had the highest total moisture, ash, protein, crude fibre and mineral content, while bread supplemented with 10% OD plum powder had the highest crumb moisture, and bread supplemented with 10% OD apple powder had the highest reducing sugar content. Molasses and molasses-based ingredients significantly improve breads antioxidant activity. Highest value was observed for 10% OD plum powder added bread. Specific volume greatly decreased and colour darkened with increased supplementation. The crumb and crust of bread supplemented with 10% OD powders and molasses showed a decrease in yellowness (b^*) compared to bread made with 5% OD powders. A significant increase in crumb pore roughness and decrease in crumb elasticity were observed with increasing supplementation level. The taste attributes of bread supplemented with 5% OD was acceptable.

Barley flour: Varying amounts of barley flour (BF) from different varieties (Tyra, Cindy and STS 2-11) were added to wheat flour to make bread rich in antioxidants [27]. There were significantly more free phenolics consisting of flavonols and tocopherol than bound phenolics, including phenolic acids, in supplemented bread. The bread supplemented with the Tyra variety had significantly higher levels of free phenolics and total antioxidant activity than control bread. The total amount of phenolics and total antioxidant activity were highest in bread supplemented with the Tyra and Cindy

varieties. During baking, free phenolics decreased, while bound phenolics greatly increased and total antioxidant activity only changed slightly, with highest values seen in the Cindy and Tyra supplemented bread. The antioxidant properties of bread depended on the barley variety and the flour extraction rate. Other factors such as storage and baking procedure were less significant but did affect sensory attributes. Tyra supplemented bread was the least acceptable with the highest scores for bitterness, off-odour and off-flavour, followed by the Cindy variety.

Auricularia auricula polysaccharide flour: Various amount of *Auricularia auricula* (commonly called black woody ear or tree ear fungus) polysaccharide flour (AAPF) were added to wheat bread [28]. AAPF has a high content of crude fibre, carbohydrates and minerals such as Fe, Ca, P and Mg compared to wheat flour. AAPF-supplemented bread showed a marked increase in concentration-dependent scavenging activity, which increased as supplementation increased. Up to 9%, AAPF could be added without affecting the sensory qualities of the bread. Loaf weight, loaf volume and loaf height were slightly affected by 9% supplementation, but significantly negatively affected by 12% supplementation. Sensory characteristics like aroma, texture, taste and mouthfeel were not affected at supplementation levels of 3–12%, except for the colour which was affected by 12% supplementation. Bread made with 9% AAPF was considered the most suitable.

Yam flour: Different amounts of yam flour (YF) were added to wheat bread. YF contains a lot of ash, crude fibre and N₂ free extract (NFE), latter mainly as starch [29]. Loaf weight, loaf volume and loaf height showed no significant changes at 5% supplementation, while maximum changes were seen at 25% substitution. Free radical scavenging activity and total antioxidant activity showed a marked increase as the proportion of YF in the bread increased. The sensory score for colour increased as substitution increased. No statistically significant difference was observed in other sensory characters for bread made with up to 20% YF compared to wheat bread.

1.3. Factors affecting the antioxidant properties of bread:

This literature review shows that supplementation of wheat flour with sources of natural antioxidant can produce bread suitable for the functional food market. The quantity and quality of the final product are determined by different factors, as shown in Table 1.1. Enhancement of antioxidant properties mainly depends on the phenolic content of the

natural sources and the amount of supplementation. Total phenolic and flavonoid content shows a positive correlation with antioxidant activity as seen in the case of *Lens culinaris Medikus* seeds [30] and hence is used to determine product quality. A similar trend was observed when fennel seed powder, black tea, chestnut flour, banana pseudo-stem flour and yam flour were added to bread. However, antioxidant activity did not increase with higher levels of supplementation when broccoli sprout powder was used.

Free radical scavenging properties are generally evaluated using the DPPH (lipophilic stable free radicals), ABTS (hydrophilic radicals) and hydroxyl radical methods, as well as other experimental methods such as inhibition of lipid peroxidation and metal chelating activity evaluation. Increased FRAP, inhibition of lipid peroxidation and improvement of antiradical potential were shown when flour was supplemented with 4% and 5% quinoa leaves. Chelating power was also improved but only with 2% supplementation. This type of non-linear relationship may be partially explained by interactions between food components (especially phenolics, protein and starch) and gastrointestinal fluid [7]. A positive correlation was observed between increased levels of broccoli sprout powder and onion skin powder and protein–phenolic interactions. In a continuation of the chemical reaction, free amino acids were reduced and resistant starches increased, with a negative effect on protein and starch digestibility, highlighting the importance of adding the correct amount of functional ingredients. This finding may be because flour samples with low phenolic content are used at up to 20–40% supplementation levels, while spices, herbs and other extracts, such as green tea and grape seed with high phenolic content, are added at much lower supplementation levels of 2–10%.

Further in vivo studies on the production and marketing of naturally fortified antioxidant-rich bread are required. In previous research, bread enriched with a 0.5% nutraceutical compound (capsaicin extracted from red peppers, magnesium from barley germ and minerals) and eaten with salad when included in the diet for 3 months showed a significant increase in hydrosoluble antioxidants and decrease in oxidative stress in subjects compared to control [31]. This research also demonstrated the benefits of multicomponent functional food enriched with antioxidant compounds obtained from plant extracts.

Individual antioxidant compounds have been analysed in previous studies. Like vitamin E, ferulic acid, p-coumaric acid, sinapic acid from rice bran, curcumin from turmeric,

and quercetin from onion skin improve the functional properties of bread. Antioxidant profiles can be studied using a method combining spectrophotometry and HPLC [32]. Antioxidant activity is determined as the total effect of both bound and free phenolic compounds. Food products with wholegrain like bread, baking resulted in an increase in free phenolic acids but a decrease in bound phenolic acids with ferulic acid as the principal phenolic [33]. In bread supplemented with rice bran, both free and bound phenolics increased, but barley free phenolics decreased due to the interaction of phenolic compounds with proanthocyanidins and minerals such as iron. In contrast, fermentation and baking showed a positive effect by liberating bound ferulic acid in bread supplemented with nejayote. The loss of antioxidant properties during cooking can be avoided by the use of the microencapsulation technique as shown when supplementation with green tea extract is compared to supplementation with microencapsulated green tea extract. Thus, although baking reduces antioxidant activity, fortification results in increased antioxidant activity compared to unfortified bread.

The types of agents used in fermentation, an essential step in bread making, also influence antioxidant activity by altering the fermenting agent. In brewer's spent grain bread, sourdough and enzymes did not change the phenolic profile but did increase the in vitro antioxidant activity of bread [34]. The addition of legume (chickpea, lentil and bean) flours to wheat flour bread with a sourdough fermentation agent containing lactic acid bacteria showed a marked increase in DPPH radical quenching ability and phytase activity [35]. Similarly, when a micronized by-product of durum wheat was added to bread, the better nutritional quality was observed in sourdough bread compared with yeast-fermented dough. In contrast, *Monascus* mould improved bread enriched with Anka (fermented rice) by supplying monacolin K, GABA and dietary fibre with beneficial health effects. However, fermented citrus peel showed lower antioxidant activity than unfermented citrus peel when added to bread. Newer stress-resistant yeast strains producing antioxidant enzymes are being developed to enhance the fermentation ability of bread dough [36].

Finally, baking of bread in an oven or on a hearth significantly affects its phenolic profile and scavenging activity due to the formation of new products from sensitive compounds like polyphenols, reducing sugars and protein. The Maillard reaction and caramelization are the main chemical reactions during baking. The Maillard reaction is a chemical reaction between amino acids and reducing sugars that gives browned food its desirable

flavour and in baking causes the formation of a brown crust. Maillard reaction products also contribute to antioxidant activity. In bread fortified with intermediated pearled wheat fraction, more TPC content was found than supplied by the raw materials due to the formation of new intermediate phenolic products through the Maillard reaction. The antioxidant activity of rye bread, especially in the crust, was also affected by the Maillard reaction occurring during baking. Advanced Maillard reaction products resulted in good scavenging of peroxy and ABTS radicals and increased TPC, enhancing the formation of antioxidants during rye bread baking [37]. Typical chestnut flour volatiles such as furans and pyrroles were formed due to the same type of reaction, producing toasty and nutty notes which improved consumer acceptability. Also, natural antioxidant sources like grape seed extract showed inhibitory effects on the formation of CML the detrimental compound formed in bread crust during baking. The interaction between phenolics (flavonoids) and protein affects the antioxidant efficacy of flavonoids, as observed following onion skin fortification of bread [38].

Apart from their antioxidant activity, natural sources of antioxidants affected the digestive system. Negative effects resulting from the binding of phenolic compounds to protein and starch have been mentioned above. Similarly, inhibition of enzymes (protease and amylase) also reduces protein and starch digestibility, resulting in increased levels of free amino groups and resistant starch [7]. The anti-inflammatory activity associated with maca-enriched bread, in vitro release of catalase activator and the anticancer activity of broccoli sprout-fortified bread, demonstrated the dynamic role of natural antioxidant sources. Supplementation of wheat flour usually increased dietary fibre, which consequently affected glucose absorption. *S. rebaudiana* extracts added to wholegrain flours decreased glucose absorption by increasing dietary fibre, while the addition of maca flour had an inhibiting effect on α -amylase and α -glucosidase, thus also hindering glucose absorption. Different types of fibre have different impacts. When whole wheat flour, which has a high content of insoluble dietary fibre, was supplemented with cellulose, the amount of resistant starch increased while the amount of digestible starch decreased, thus delaying starch digestion. In contrast, barley, oat and rye contain higher levels of soluble dietary fibre and so have less effect on lowering glucose absorption [21].

The type of wheat flour used was also a major factor affecting antioxidant activity. The effect of the coat colour of wheat was studied. Investigations showed that red and white

wheat grainflour showed different phenolic acid profiles despite having comparable total phenolic acid content (TPAC) [39]. A similar effect was observed when red and white sorghum flours were added to wheat bread. Kisra, traditional Sudanese flatbread, was prepared using two different sorghum cultivars. The two types of bread showed significantly different FRAP and DPPH values, which were enhanced by baking, with increased fermentation time also resulting in higher values and hence increased antioxidant activity [40]. Therefore, a change in the type of dough flour can lead to a remarkable change in antioxidant activity. Another investigation found that, despite the loss of TPC and antioxidant activity following baking, bread containing pseudocereals showed significantly higher antioxidant capacity than bread made only with wheat flour [41].

The physical properties of a fortifying agent have a combined effect on overall acceptability. Gluten quality and quantity are important characteristics of bakery flour. Gluten quantity decreases when natural sources of antioxidants such as turmeric, ginger and coriander powder are added to wheat flour. High fibre and gluten-free flours such as rice bran, banana pseudostem, chestnut and sorghum, and spices such as fennel and cinnamon affect the gluten chain by diluting it with fibre and fibre induce increased water absorption capacity. With a decreased bread volume, high moisture content also affects bread quality negatively. Consequently, a higher level of coriander supplementation resulted in improved bread due to the lower level of fibre. The addition of micronized by-products of debranned durum wheat to sourdough also increased loaf volume compared to the same supplementation of yeast dough.

Hydrocolloids like CMC when added to banana pseudo-stem-fortified bread improved physical properties by balancing soluble and insoluble dietary fibre content with decreased antioxidant activity. The hydrocolloid-like properties of naturally formed alkali-treated fibre in nejayote solids also provide another method of improving bread. The darkness of crumb and crust is directly proportional to phenolic content and Maillard reaction. A higher level of reducing sugar in chestnut flour and higher levels of protein in rice bran increased the darkness of bread crust, while the crumb colour was affected by turmeric and coriander.

Another important parameter is the taste which is perceived as flavour combined with aroma. The acceptability of the sweet aroma of fennel seed, the pungency of ginger and the woody notes of chestnut is related to the amount of fortification. Differences in

processing like employing sourdough instead of yeast fermentation masked the inferior flavour of grape by- product and improved the sensory profile of bread containing micronized by-products from debranned durum wheat. At the increased level of Stevia rebaudiana, the product becomes less sweet than control due to the presence of chlorophyll pigment and glycosides. In general, higher levels of fortification with natural sources of antioxidants negatively affect bread quality, so it is important to identify optimum levels of supplementation.

In addition to the various processing parameters, storage conditions also affect antioxidants in bread. One study showed that the TPC and oxidative stability of bread samples decreased during storage depending on the manufacturing process. However, frozen dough bread lost fewer phenolics and had higher oxidative stability compared to frozen baked bread [42]. This indicates that the fortification ingredients, as well as various environmental, physical and chemical factors, affect the antioxidant capacity of bread enriched with natural sources of antioxidants.

Aim and Objectives

Aim of the study was to improve the quality of bread, which is a low cost, easily available, energy dense food and preferred by all age groups. For most of the people it is a common food in regular diet. So, if it is possible to improve the existing bread quality then all the people, despite of their socioeconomic status, will be benefited.

Below objectives were set to achieve the goal:

1. Enhancement of bread quality by improving antioxidant activity.
2. Utilisation of food processing waste as a low cost, easily available natural antioxidant source.
3. Also it has to be studied, whether this kind of enrichment process is able to improve any physical or sensory characteristics of bread samples along with antioxidant activity.

CHAPTER: 2

2. Incorporation of tropical fruit by-product in whole wheat bread as a quality improver

2.1. Introduction:

Most of the bakery products which we consume in our daily diet are processed from refined wheat flour. During the refining process, bran and germ, the most nutritious parts are removed from the whole grain. Due to the presence of fibre, whole grains are slowly absorbed than refined grains. So, the consumption of whole grain foods, as a part of a healthy diet, reduces the risk of some chronic diseases like obesity, coronary heart disease, type II diabetes, etc. [43].

There are very limited research works on consumer preference tests between refined and whole wheat bread. Most of the studies support the general hypothesis that, people like refined bread more than whole wheat bread but there are some cases also that contradict this fact. A food preference test among men of United States armed forces showed more preference for refined bread, on the other hand, a study among old cancer patients showed more preference for whole wheat bread than the refined one [44]. Also according to a taste test data [45], many children showed an equal liking for two products (refined and whole wheat bread). So, the preference of whole wheat bread to a subject depends on several factors. Some like it due to its health benefits whereas some dislike it for its sensory parameters.

Mango (*Mangifera indica* L.) is one of the most popular tropical fruits that are grown abundantly in more than 90 countries across the world [46]. In this array, with 42.2% of the world's total production, India is the largest mango producing country [47]. In West Bengal more than 41% of total fruit farming land is occupied by mango cultivation [48], producing commercial varieties like Fazli, Gulabkhas, Himsagar, Kishenbhog, Langra, Bombay Green, etc. Besides taking ripe mango as a table fruit, it is also used for preparing various traditional and commercial products like *murabba*, squash, jams, jelly, nectar, leather, puree etc. While processing these traditional and commercial products most of the time peel is considered as a waste, which is around (15-20) % of whole fruit [49]. This amount may vary for different varieties of mango. Therefore, sustainable utilisation of mango peel in the food industry could be an environment-friendly waste

management technique. Ripe mango peel in powder form already used as a natural antioxidant source in various wheat based products [50] [51] [52] and it was observed that up to a certain addition level it did not hamper the consumer acceptability.

The aim of our study is the incorporation of ripe mango peel powder (RMPP) at different levels (1%, 3%, and 5%) in whole wheat bread (WWB) as a source of antioxidant, to make it healthier. In this research work, antioxidant properties of bread samples along with physical and sensory properties are evaluated and compared with the control sample (without RMPP). It is assumed that with improved functional properties the whole wheat bread would be able to attract more consumers. Also, the mango peel may help to reduce the sensory barriers of whole wheat bread by its own aroma and taste.

2.2. Materials and methods:

2.2.1. Raw materials: Ripe mangoes (*Mangifera indica* L., cv. *Himsagar*), whole wheat flour, sugar, salt, and vegetable oil were purchased from a local market in Jadavpur, Kolkata. Compressed Baker's yeast (Saf Yeast Company Pvt. Ltd., Mumbai, India), Glycerol monostearate (GMS) (Loba Chemie Pvt. Ltd., Mumbai, India) and Potassium bromate ($KBrO_3$) (Merck Specialities Pvt. Ltd., Mumbai, India) were used in bread preparation.

2.2.2. Chemicals: Gallic acid (SD Fine-Chem Ltd., Mumbai, India), DPPH (Sigma-Aldrich, St. Louis, MO, USA), Folin-Ciocalteu reagent, sodium carbonate, ethanol (Merck Specialities Pvt., Ltd., Mumbai, India) were used in the investigation.

2.2.3. Preparation of Ripe Mango peel powder (RMPP): Ripe mango peels were collected by peeling freshly bought mangoes, followed by washing with tap water. Then the peels were cut into small strips (2×3) cm and blanched for (3-5) min first in boiling water and then in ice water.

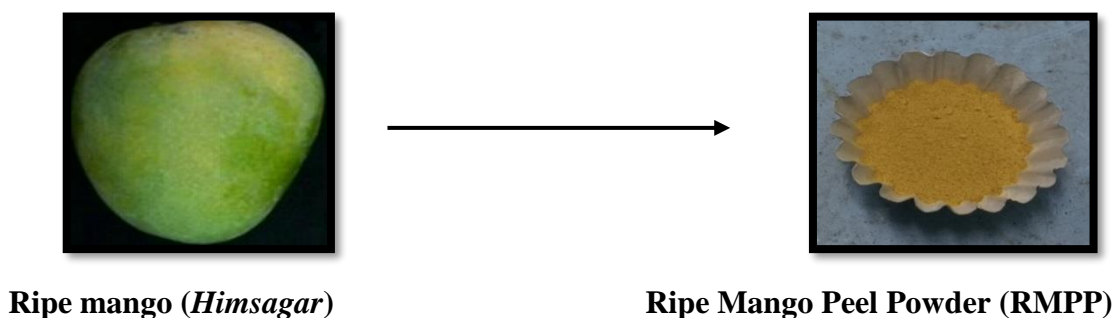


Figure 2.1: Image of Ripe Mango Peel Powder (RMPP)

After this, the peel was dried at $60\pm 2^{\circ}\text{C}$ using a Tray drier (Reliance Enterprise, Kolkata, India) until it attained constant weight. The dried peel was then powdered using a grinder (GX7, Bajaj Electricals Ltd, India) and was sieved through a 60 mesh (0.25mm) and stored at -20°C in an air-tight plastic container, resulting in preparation of Ripe mango peel powder (RMPP).

2.2.4. Bread preparation: The ingredients and their proportions required for the preparation of bread samples were whole wheat flour (100g), sugar (6.0%), salt (1.5%), vegetable oil (2.0%), GMS (1.5%), KBrO_3 (1ppm) and water at optimum amount along with pre-activated yeast (3g), in 20ml lukewarm water with 2g sugar in a bowl and kept at $35\pm 2^{\circ}\text{C}$ for 15min. Here, mixing was carried out by adding dry ingredients, shortening and the activated yeast in a bowl along with water and then kneaded until the dough obtained the required elasticity. After this, the dough was rounded and kept in a bowl for the first proofing at the room temperature ($30\pm 2^{\circ}\text{C}$) for about 30min with wet cloth cover. After the first proofing, the dough was punched and worked lightly for the excess gas to escape and the redistribution of the gas cells. The dough was then shaped to fit lightly in a greased baking mould. The dough was again kept for the final proofing for about 1hr at $40\pm 2^{\circ}\text{C}$. Finally, after the second proofing, the dough in the mould was baked in a Rotary oven (CM HS108, Chanmag Bakery Machine Co. Ltd., Taiwan) at $215\pm 2^{\circ}\text{C}$ for 18-20min. After baking, the prepared bread sample was cooled for about 1hr at room temperature and then analysis was carried out. Fortified samples were prepared in the same procedure, except RMPP was added individually with dry ingredients at 1%, 3%, and 5% level.

2.2.5. Analysis of RMPP: Weight of different fruit parts was taken on a citizen (model MP-300) electronic balance to calculate the Peel to fruit weight ratio (PFR).

Proximate analysis: The moisture, total protein, crude fat, ash, total dietary fibre, soluble and insoluble dietary fibre contents of RMPP were determined by AOAC standard methods (1999) [53].

HPLC assay: The phenolic compounds of RMPP were identified using HPLC analysis method [54]. Eighteen phenolic compounds were used as standards and these compounds were gallic acid, protocatechuic acid, (+)-catechin, p-coumaric acid, ascorbic acid, chlorogenic acid, caffeic acid, epicatechin, syringic acid, vanillic acid, ferulic acid,

sinapinic acid, rutin, rosmarinic acid, trans-cinnamic acid, quercetin, kaempferol, and apigenin.

2.2.6. Determination of antioxidant activity: Extract preparation: Bread crumb (1g) in 80% aqueous ethanol (10ml) was sonicated in a Sonication water bath (D150 IH, ultrasonic power 100 W and frequency 50 KHz, heater power 200 W, AC 220-240 V 50 Hz; Trans-o-sonic, Mumbai, India) until equilibrium. Then it was centrifuged at 8944×g for 15min and the supernatant was collected and subjected to further analysis.

Total phenolics content: The Folin–Ciocalteu reagent assay was used to determine the total phenolics content (TPC) [55] of the bread extract. After incubation at room temperature (30±2°C) for 90min, the absorbance of the mixture was observed at 760 nm. The result was expressed as gallic acid equivalent (GAE) in mg/100g of sample.

DPPH radical scavenging assay: The free radical inhibition activity of the bread extract was determined using the stable 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay [56]. The decrease in absorbance of the experimental mixture was measured at 515 nm. The percentage of inhibition was calculated as described in Eq. (1);

$$\% \text{ Inhibition} = (\text{Abs}_{\text{blank}} - \text{Abs}_{\text{sample}}) \times 100 / \text{Abs}_{\text{blank}} \quad \dots\dots\dots (1)$$

2.2.7. Sensory evaluation: The evaluation of the sensory attributes of the RMPP added breads were done by the method of descriptive analysis. The first step involving a section of judges was done by selecting members from faculties and students of Department of Food Technology & Bio-Chemical Engineering, Jadavpur University, Kolkata. The panel constituted 50 judges including equal number of females and males aged between 20 to 55 years. To distinguish the descriptive features of the bread samples, the panellists devised 8 descriptors for the bread and defined each of the features (Table 2.1) which covered the areas of appearance, aroma, taste, colour and oral texture. The panel members were trained in three training sessions of 1 h each to inculcate the ability to discriminate between the samples.

Table 2.1: Descriptors and definitions used for sensory profiling of RMPP added bread samples

Attribute	Definition
Appearance	
Porosity	The extent of perforation of the bread surface, this encompasses the holes and cracks allowing the permeation of air
Aroma	
Traditional bread aroma	Characteristic aroma of traditional whole wheat bread due to yeast fermentation
Fruity aroma	Primary aroma of ripe mango peel powder
Taste	
Fruity taste	Typical taste of fruit bread
After taste	Typical taste of caffeine as a residual taste sensation following consumption
Colour	
Crumb colour	Degree of darkness in crumb colour ranging from brown to dark brown
Oral-texture	
Hardness	The amount of force produced by molar teeth to bite completely through the sample
Stickiness	During food consumption the amount of force applied by tongue to remove it completely from the palate.

Finally, the assessment was conducted in individual booths under white light at room temperature at Dept. of Food Technology & Bio-Chemical Engineering, Jadavpur University, Kolkata. For evaluation, approximately 1 cm thick slice including the crust and crumb was presented to assessors. The samples were kept in a 3-digit coded glass covered with a glass lid. Water was given for mouth cleansing between two samples. The assessors evaluated the samples on an unstructured 9 cm line scale labelled on the left by 'none' and on the right with the term 'strong'. Ratings were registered on an evaluation form. Samples were assessed in triplicates in four sets [57].

2.2.8. Determination of physical parameters: Weight loss: Initial weight of dough and final weight of baked loaf was taken to calculate the percent weight loss as described in Eq. (2); [58]

$$\text{Weight loss (\%)} = \frac{\text{weight of dough} - \text{weight of baked loaf}}{\text{weight of dough}} \times 100 \quad \dots\dots\dots (2)$$

Specific volume and Density: The bread sample was weighed after cooling for 1hr at room temperature and the volume of the loaf was measured by the seed displacement method. From these, specific volume and density were calculated as described in Eq. (3) & (4); [59]

$$\text{Specific volume (cm}^3\text{/gm)} = \frac{\text{Loaf volume}}{\text{Weight of bread}} \quad \dots\dots\dots (3)$$

$$\text{Density (gm/cm}^3\text{)} = \frac{\text{Weight of bread}}{\text{Loaf volume}} \quad \dots\dots\dots (4)$$

Loaf height: Loaf height was measured by making an average of three data, which was taken at the left, right and middle position of baked loaf from top to bottom.

Moisture: The moisture content of the bread crumb was analyzed by taking the 5g samples and placed in a Hot air oven (Reliance Enterprise, Kolkata, India) at 105±2°C for 4 hrs. After drying, the sample was cooled in desiccator and weight was taken. The repetition of this process was done until a constant weight was achieved. Moisture content was determined according to the Eq. (5); [60]

$$\text{Moisture content (\%)} = \frac{\text{Initial wt. of the sample} - \text{Final wt. of the sample}}{\text{Initial wt. of the sample}} \times 100 \quad \dots\dots\dots (5)$$

Hardness: Hardness of the bread crumb was determined by a Texture analyser (TA.HD Plus Texture Analyser, USA). The analysis was performed with a 35mm diameter probe, compressing the sliced bread (40mm×40mm×30mm) at a cross-head speed of 0.1-2 mm/s and with 30-60 % deformation.

Colour: The whiteness index (WI) of the bread crumb and brownness index (BI) of the bread crust were measured by the Hunter Lab colour measurement system, ColorFlex 45/0, D65, 10° observer (Hunter Associates Laboratory Inc., Reston, VA, USA) by standardizing with white and black plate. The results were expressed according to the CIELAB colour measurement system as L* lightness (0; black to 100; white), a* (-; green to +; red), and b* (-; blue to +; yellow). The whiteness index and brownness index of bread sample were calculated based on the following Eq. (6); [22]

$$\text{Whiteness index (WI)} = 100 - ([100 - L^*]^2 + a^{*2} + b^{*2})^{1/2} \quad \dots\dots\dots (6)$$

and Eq. (7); [61]

$$\text{Brownness index (BI)} = \frac{[100(x-0.31)]}{0.17} \quad \dots\dots\dots (7)$$

$$\text{Where, } x = \frac{(a+1.75L)}{(5.645L+a-3.012b)}$$

2.2.9. Statistical analysis: The experimental design was completely randomized, replicated for three times and expressed as mean value \pm SD. All experimental data was statistically analyzed for analysis of variance (ANOVA) in Microsoft Office Excel 2007 using XLSTAT statistical software. The values for descriptive analysis were also analysed by two-way ANOVA. The comparisons between the mean values were tested using Duncan's new multiple range test at a level of $p \leq 0.05$.

2.3. Results and discussion:

2.3.1. Analysis of RMPP: In this research work ripe mango peel (RMP) of *Himsagar* variety, which is $12.86 \pm 1.37\%$ of total fruit weight, was used as a bread quality improver. RMP was converted in RMPP by tray drying up to 10.17% moisture level. Other components like total protein, crude fat and ash content were 4.35%, 2.16% and 3.87% respectively, as mentioned in Table 2.2. Also it contained 56.64% total dietary fiber, in which 20.21% was soluble dietary fiber and 36.43% was insoluble dietary fiber. Like PFR, proximate composition varies with the variety of mango, also ripening stage and analysis method may responsible for this variation [62] [63].

Table 2.2: Proximate composition of RMPP (g/100g dry sample, except for moisture)

Component	Content (%)
Moisture	10.17 ± 1.08
Total protein	4.35 ± 0.3
Crude fat	2.16 ± 0.1
Ash	3.87 ± 0.35
Total dietary fibre	56.64 ± 0.5
Soluble dietary fibre	20.21 ± 0.74
Insoluble dietary fibre	36.43 ± 0.49

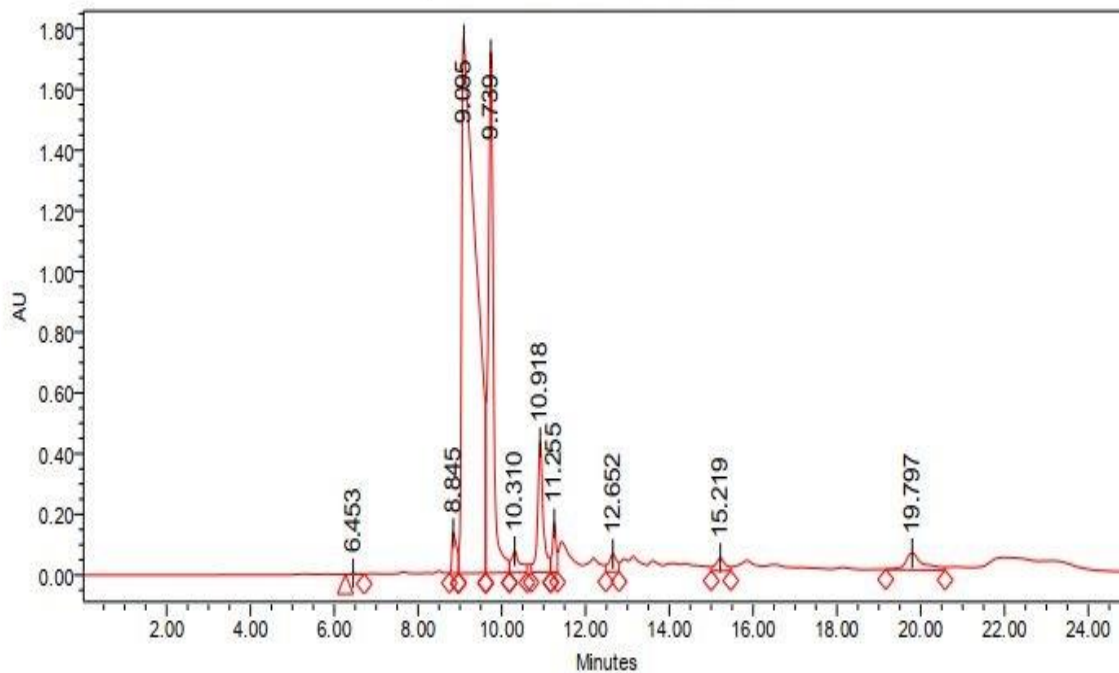


Figure 2.2: HPLC chromatogram of RMPP: ascorbic acid (6.453min), gallic acid (8.845min), dihydroxybenzoic acid (9.095min), catechin (9.739min), vanillic acid (10.310min), chlorogenic acid (10.918min), p-Coumaric acid (11.255min), ferulic acid (12.652min), apigenin (15.219min), kaempferol (19.797min)

The composition of phenolic compounds in RMPP is shown in Figure 2.2. Ten phenolic compounds, identified in *Himsagar* variety RMPP, were ascorbic acid, gallic acid, dihydroxybenzoic acid, catechin, vanillic acid, chlorogenic acid, p-Coumaric acid, ferulic acid, apigenin and kaempferol. According to the experimental data, catechin, dihydroxybenzoic acid, p-Coumaric acid and chlorogenic acid can be considered as predominant phenolic compounds. Like other varieties [64] [62] RMPP of *Himsagar* also contains both dietary fiber (DF) and phenolic compounds in a good amount. As a recent concept, this type of food ingredients is known as ‘antioxidant dietary fiber’ [65]. Though in this study RMPP is used as an antioxidant source, still analysis of DF content is important to explain the effect of RMPP on physical parameters.

2.3.2. Antioxidant activity: Total phenolics content and DPPH inhibition activity of control bread and other bread samples with different percentages of RMPP were observed and values were given in Table 2.3. There were significant differences in respect to TPC between all breads prepared with RMPP and control. 5 % incorporation of RMPP showed the highest value of TPC 757.8 ± 13.5 mg GAE/100gm compared to control. So, like other wheat based products [50] [51] [52] in this case inclusion of

RMPP helps to increase TPC in bread. A similar result was observed for DPPH inhibition activity. Though, phenolic compounds present in whole wheat flour showed 21.69% DPPH inhibition, still this value increased more than three times at 5% RMPP addition. So, looking forward, the improved antioxidant activity may help to increase the acceptability of whole wheat bread in the presence of popular wheat bread [44].

Table 2.3: Antioxidant properties of bread samples substituted with RMPP at different levels

Parameters	Control	1%	3%	5%
TPC (mg GAE/100gm)	220.33±8.5 ^a	404.67±16.04 ^b	507±11.5 ^c	757.8±13.5 ^d
DPPH (% inhibition)	21.69±0.72 ^a	30.27±1.03 ^b	50.29±1.13 ^c	65.74±0.51 ^d

Mean with different letters within a row is significantly different ($p \leq 0.05$)

2.3.3. Sensory evaluation: Scores for the various descriptors for RMPP added bread is shown in Table 2.4. Considering the crumb appearance depicted by porosity, the intensity decreased when the level of addition was increased, still an insignificant difference was found between 1% and 3% addition level. Though for specific volume and density analysis, these two samples showed a significant difference and a lower value for 3% than 1%. However, the traditional bread aroma decreased with the amount of RMPP addition; reason behind was increased fruity aroma. RMPP also imparted a fruity taste in the bread samples. Phenolic compounds present in the bran of whole grain imparted a bitter after taste in whole cereal based products [57]. Here this bitterness increased by the phenolic content of RMPP but at 3% and 5% addition level this value was lower than fruity taste. Crumb colour showed a similar trend like WI value (Table 2.6); where with respect to control significant difference (lower value) was found for 3% and 5%. For oral texture both hardness and stickiness values were increased gradually. So, as a positive outcome, flavour of whole wheat bread, one of the sensory barriers for consumer acceptability [66], was improved by RMPP addition. But textural betterment needs to be considered for its industrial implementation.

Table 2.4: Sensory evaluation of the bread samples substituted with RMPP at different levels

Attributes	Samples				SD
	Control	1%	3%	5%	
Porosity	7.8 ^c	7.44 ^b	7.21 ^b	6.81 ^a	0.27
Traditional bread aroma	7.7 ^c	7.54 ^{b,c}	7.21 ^{a,b}	6.86 ^a	0.58
Fruity aroma	2.19 ^a	3.5 ^b	4.16 ^c	4.67 ^d	1.07
Fruity taste	1.5 ^a	2.25 ^b	3.38 ^c	4.8 ^d	1.45
After taste	1.59 ^a	2.4 ^b	3.01 ^c	3.92 ^d	0.98
Crumb colour	6.4 ^a	7.66 ^{a,b}	8.19 ^b	8.53 ^c	0.94
Hardness	4.2 ^a	4.7 ^b	5.5 ^c	6.16 ^d	0.84
Stickiness	4.49 ^a	6.24 ^b	6.62 ^c	6.97 ^c	1.1

Mean with different letters within a row is significantly different ($p \leq 0.05$)

2.3.4. Physical parameters: The effect of RMPP addition on physical parameters (except colour) of bread loaves are shown in Table 2.5. Addition of mango peel in increasing order resulted in the continuous decrease of weight loss value than control sample. The reason behind the change could be because of the fibre present in mango peel that restricts the yeast activity during fermentation by creating a barrier around fermentable sugar. In the same way, when the result of the specific volume was compared, a lower value with significant change was observed for RMPP added bread than control sample. Gluten protein presents in dough got diluted due to the interaction with mango peel fibre, which might have reduced the CO₂ retention capacity resulting in a lower loaf height causing lower specific volume and higher density [6]. On the other side, crumb moisture content increased with the addition level of RMPP. From previous studies it can be assumed that the addition of a fibre containing substance can elevate the moisture content of a baked product [67]. The fruit waste, mango peel, is rich in hydrolysed dietary fibre especially in its ripe stage and absorbs more water during kneading. As a negative effect, it increases the loaf density and decreases the loaf volume with compact crumb structure. One of the important textural attribute that interfere the consumer acceptability of bread is hardness. According to Table 2.5 up to 1% RMPP addition had no significant difference for hardness value with control sample. But at higher level (3% and 5%) nutrient composition of mango peel affected the uniform structure that changed the textural characteristics of bread. This is in accordance

with the addition of other non-wheat based gluten free products where addition of quinoa leaves increased the hardness of the bread crumb [7].

Table 2.5: Physical parameters of bread samples substituted with RMPP at different levels

Parameters	Control	1%	3%	5%
Weight loss (%)	15.71±0.70 ^d	13.01±0.12 ^c	10.31±0.26 ^b	7.16±0.18 ^a
Loaf height (cm)	5.55±0.21 ^c	4.7±0.23 ^{bc}	4.43±0.14 ^b	4.0±0.43 ^a
Specific volume (cm ³ /g)	2.33±0.06 ^d	2.20±0.07 ^c	2.07±0.02 ^b	1.87±0.15 ^a
Density (g/cm ³)	0.41±0.01 ^a	0.45±0.03 ^b	0.48±0.02 ^c	0.52±0.02 ^d
Moisture (%)	33.97±0.45 ^a	37.51±0.8 ^b	40.82±1.14 ^c	44.41±1.22 ^d
Hardness (N)	2.47±0.32 ^a	3.0±0.4 ^a	3.64±0.26 ^b	3.93±0.15 ^b

Mean with different letters within a row is significantly different ($p \leq 0.05$)

Colour is another important parameter for bakery product which determines the consumer acceptability. WI for crumb and BI for crust were selected as colour indicators to study the changes. In Table 2.6 WI value for RMPP added samples were significantly decreased than control, except at 1% addition level. This could be due to the effect of RMPP imparting its own reddish yellow colour ($L^*-53.06$, $a^*-8.81$, $b^*-31.12$) in the bread crumb, which became dark at higher percentage of addition. Consumer may like this change, for example anka enriched bread obtained lower WI value than control bread but an opposite result was observed in sensory evaluation [22]. BI of the RMPP incorporated samples was higher as compared to the control, but an insignificant difference was also observed between 3% and 5%. Maillard reaction, the main reason for the darkening of crust, which might have been influenced by the colour of composite flour and the excess water retained by RMPP in bread dough [18].

Table 2.6: Colour parameters of bread samples substituted with RMPP at different levels

Parameters	Control	1%	3%	5%
Whiteness index	56.26±1.11 ^b	50.49 ± 0.88 ^b	49.47±0.93 ^a	48.11±1.06 ^a
Brownness index	81.39±7.69 ^a	94.81±5.65 ^b	117.52±2.55 ^c	123.57±4.61 ^c

Mean with different letters within a row is significantly different ($p \leq 0.05$)

2.4. Conclusion:

According to the present research work ripe mango peel powder can be considered as a potential health promoting functional ingredient. Both quantitative and qualitative aspect of antioxidant activity in RMPP added whole wheat bread samples show an increasing trend with respect to addition level. This trend is also supported by sensory evaluation, where 5 % RMPP level gained maximum scores for flavour. On the other hand, RMPP at 5% imparts highest negative impact on physical and sensory parameters apart from flavour due to the interference of dietary fibre and phenolic compounds with bread dough. Though in comparison with 1%, 3% sample shows lower effect for selected physical and sensory parameters. Thus, based on the objective, 3% RMPP can be considered as the best addition level. Also as a future scope, modification in processing parameters may improve the product quality even at a higher addition level.

CHAPTER: 3

3. Optimisation of ripe mango peel enriched white bread preparation and its characterisation

3.1. Introduction:

In daily life, people consume white bread as a low cost and readily available energy dense food. Additionally, easy amenability of refined wheat flour towards processing makes it more appropriate for various food preparations. Now a day where functional foods are becoming increasingly popular as a part of healthy lifestyle, white bread contains limited functional compounds [68][69]. Because the bran and germ, which contain most of the bioactive compounds (mainly dietary fiber and phenolic compounds), are removed during refining of whole wheat. Based on this fact, many research works were conducted in last few years to incorporate food industry by-products as functional ingredients in white bread preparation to make it healthier [70]. In this field, application of fruit by-products is a relatively new approach and yet to be investigated fully [71][72]. For example, increasing addition of pomegranate [73] and grape [74] processing by-products in white bread improve both total phenolic content and radical scavenging activity. On the contrary, the same addition negatively affects the physical parameters and the overall acceptability of the bread samples as well. Therefore, incorporation of fruit by-products could be an easy and cost effective method to enhance the bread quality, but for the desired outcome addition level should be optimised first in respect of both antioxidant activity and physical parameters. Apart from addition level, antioxidant activity mostly affected by baking conditions [4] as it involves both destruction and formation of phenolic compounds. Also baking temperature and time are two important factors which influence the physical parameters and thus determine the final quality of bread sample [75]. For this reason, both baking temperature and time can be considered as independent variables in the above mentioned optimisation process.

Both phenolic compounds and dietary fiber were present in good amount in Mango peel, whereas this amount and composition vary according to its variety [76]. As per literature review, ripe mango peel when used in bakery product like biscuit [50] and whole wheat bread [77] a remarkable change in DPPH radical scavenging activity from 26.13% to 88.84% and 21.69% to 65.74% were observed respectively. Besides, mango peel

possesses high water holding capacity [76] which might improve storage stability of bakery products. Hence, addition of ripe mango peel in white bread with acceptable sensory scores could help in value added product development as well as an environment-friendly waste management technique.

As per literature review, no research work has been done regarding ripe mango peel addition in white bread till now. In this study, production of ripe mango peel enriched white bread was optimised and selected parameters were compared with the control sample.

3.2. Materials and methods:

3.2.1. Raw materials: Ripe mangoes (*Mangifera indica* L., cv. *Himsagar*), refined wheat flour, active dry yeast, salt, vegetable oil and sugar were collected from the market of Jadavpur, Kolkata.

3.2.2. Chemicals: All the chemicals were purchased from Sigma-Aldrich (St. Louis, MO, USA).

3.2.3. Preparation of Ripe mango peel powder (RMPP): Same as section: 2.2.3.

3.2.4. Water holding capacity (WHC): The water holding capacity (WHC) was determined by mixing 1g powder sample with 10ml distilled water and kept idle for 24hrs at room temperature (30°C). After that with precision supernatant was removed to calculate WHC as grams of water retained per gram of solid [78].

$$\text{WHC (g/g)} = \frac{\text{Weight of sediment after draining supernatant} - \text{Sample dry weight}}{\text{Sample weight}} \quad \dots\dots\dots (1)$$

3.2.5. Experimental design: To study and optimize the combined effect of baking temperature (X_1), baking time (X_2) and RMPP addition level (X_3) on the specific volume (Y_1 ; cm³/g) and DPPH radical scavenging activity (Y_2 ; μmol AAE/g) of bread samples a three- level three-factor face-centered central composite design (FCCD) was used. Using single factor analysis, levels of independent variables were selected. The -1, 0 and +1 levels were fixed at 190, 210 and 230 for baking temperature (°C), 15, 30 and 45 for baking time (min) and 3, 5 and 7 for RMPP addition level (%). 17 randomised experiments (8 factorial points, 6 axial points and 3 centre points) were preformed. The nonlinear quadratic model is given as:

$$y = b_0 + \sum_{i=1}^3 b_i x_i + \sum_{i=1}^3 b_{ii} x_i^2 + \sum_{i \neq j}^3 b_{ij} x_i x_j \quad \dots\dots\dots (2)$$

Where, y is the measured response, b_0 is the intercept, b_i , b_{ii} and b_{ij} are the regression coefficients associated with linear, quadratic and interaction terms of independent variables. For optimisation the goals were set as follows: specific volume- target ($2.500\text{cm}^3/\text{g}$) and DPPH radical scavenging activity- maximum. After optimisation, triplicate experiments were performed under optimised conditions and experimental results were compared to values predicted by the model.

3.2.6. Bread preparation: For bread preparation following ingredients like, refined wheat flour (100g), active dry yeast (2.5g), sugar (8.0%), salt (1.5%), vegetable oil (4.0%) and water (65ml) were used. RMPP was added at different levels according to the experimental design by replacing the equal amount of wheat flour. Initially, all dry ingredients and shortening were mixed with water in a bowl, followed by kneading until the dough attained the required elasticity. First proofing was done after this at room temperature (30°C) for about 30min with wet cloth cover. Then the dough was kneaded lightly to redistribute gas cells and purge the excess gas. The dough was then placed in a greased baking mould for the final proofing at 40°C for 45min. In a rotary oven (CM HS108, Chanmag Bakery Machine Co. Ltd., Taiwan) dough was baked at different baking time and temperature combinations. Control bread (without RMPP) was prepared following the same protocol by baking for 20min at 220°C . Before analysis samples were cooled at room temperature for 1 hour.

3.2.7. Specific volume: Same as section 2.2.8.

3.2.8. Antioxidant activity: Same as section 2.2.6.

3.2.9. Total dietary fibre: Same as section 2.2.5.

3.2.10. Colour: Same as section 2.2.8.

3.2.11. Sensory evaluation: Sensory parameters of optimised bread and control bread were evaluated by a panel of 15 members including student, staff, and faculty of the Dept. of. Food Technology & Bio-Chemical Engineering, Jadavpur University, Kolkata. Based on 9-point hedonic scale sensory evaluation of the bread loaves were evaluated as appearance, colour, aroma, taste, mouth feel and overall acceptability. Range of the scale was 9 to 1 which denoted as like extremely to dislike extremely. A basic training was given to the panelists to understand the scoring process and the sensory parameters. The panelists were seated in individual sensory booths at room temperature under white light with water for mouth rinsing after each set of evaluation at Dept. of. Food Technology &

Bio-Chemical Engineering, Jadavpur University, Kolkata. Sliced bread samples were given with random codes. Each panelist was given both control and optimised bread samples for the evaluation. The evaluation process was repeated thrice [18].

3.2.12. Crumb firmness: A storage study of crumb firmness at room temperature (30°C) for 5 days was carried out for both optimised bread and control bread. Bread samples were evaluated by TA.XT Express texture analyzer, Stable Micro Systems, Surrey, UK, is the manufacturer. Loaves were sliced by hand into 25mm thickness from the centre of bread. A 36 mm diameter cylinder probe SMS P/36R with 5kg load cell, 40% penetration depth were the optimal test conditions. Pre-test value was 1.0 mm/s, test value was 1.7 mm/s and finally post-test speed was 10 mm/s. The compression force value (CFV) (g) at 25% compression of 25mm sample was used to express firmness value. To assess the stalling kinetics Avrami's non-linear regression equation was used by fitting hardness value (3); [79]

$$\theta = \frac{T_{\infty} - T_t}{T_{\infty} - T_0} = e^{-k*t^n} \quad \dots\dots\dots (3)$$

Where, θ - fraction of recrystallization resting to take place, T_{∞} - firmness in final day, T_t - firmness in t day, T_0 - firmness in zero day, k – rate constant and n – Avrami exponent.

3.2.13. Statistical analysis: All the studies were replicated 3 times and the means were reported. All the experimental data were analyzed statistically for analysis of variance (ANOVA) with Minitab 17.0 (Minitab, Inc., Pennsylvania, PA). Means were compared by Tukey's test at a significance level of $p \leq 0.05$.

3.3. Results and discussions:

3.3.1. Analysis of the model: To examine and optimize the coalesced outcome of three independent variables baking temperature (X_1), baking time (X_2) and RMPP addition level (X_3), a set of 17 randomized experiments (8 factorial points, 6 axial points and 3 centre points) were performed and obtained results are provided in Table 3.1.

Table 3.1: Face-centered central composite design with results of specific volume and DPPH radical scavenging activity of bread samples

Run order	Baking temperature (°C)	Baking time (min)	RMPP addition level (%)	Specific volume (cm ³ /g)		DPPH radical scavenging activity (µmol AAE/g)	
				Experimental	Predicted	Experimental	Predicted
1	190	15	3	2.728	2.755	5.660	4.729
2	230	15	3	2.946	2.957	14.914	15.514
3	190	45	3	3.044	3.051	8.757	8.446
4	230	45	3	2.739	2.777	4.163	4.551
5	190	15	7	2.402	2.367	10.928	10.598
6	230	15	7	2.556	2.553	25.396	25.765
7	190	45	7	2.577	2.571	21.457	20.916
8	230	45	7	2.304	2.281	20.414	21.403
9	230	30	5	2.682	2.660	16.481	14.133
10	190	30	5	2.697	2.704	6.385	8.497
11	210	15	5	2.654	2.654	16.074	16.364
12	210	45	5	2.681	2.665	16.566	16.041
13	210	30	3	2.957	2.874	13.686	13.939
14	210	30	7	2.364	2.431	25.786	25.299
15	210	30	5	2.717	2.665	17.871	16.573
16	210	30	5	2.581	2.665	13.778	16.573
17	210	30	5	2.665	2.665	17.602	16.573

The analysis of variance (ANOVA) procedure was used to statistically determine the effect of the parameters and their interactions on the yield, and the results were summarized in Table 3.2(a) and 3.2(b). The p-value was used to check the significance of each coefficient, which indicated the influence of the corresponding term on the yield. It can be presumed that any coefficient with p-value less than 0.05 has a substantial influence on the specific volume and DPPH radical scavenging activity.

According to the ANOVA result for specific volume, X_3 and $X_1 \cdot X_2$ coefficients are significant and all other terms are insignificant. The polynomial equation was derived through fitting the individual response to the equation 2 and given below in the uncoded form:

$$\text{Specific volume (Y}_1\text{; cm}^3\text{/g)} = 2.6648 - 0.0221 X_1 + 0.0059 X_2 - 0.2212 X_3 + 0.0168 X_1 \cdot X_1 - 0.0054 X_2 \cdot X_2 - 0.0123 X_3 \cdot X_3 - 0.1188 X_1 \cdot X_2 - 0.0041 X_1 \cdot X_3 - 0.0231 X_2 \cdot X_3$$

Table 3.2(a): Analysis of variance table for specific volume

Term	Effect	Coef	SE Coef	Adj SS	F-Value	P-Value
Constant		2.665	0.026	0.613	18.18	0.000
X ₁	-0.044	-0.022	0.019	0.005	1.30	0.292
X ₂	0.012	0.006	0.019	0.000	0.09	0.768
X ₃	-0.442	-0.221	0.019	0.489	130.65	0.000
X ₁ * X ₁	0.034	0.017	0.037	0.000	0.20	0.667
X ₂ * X ₂	-0.011	-0.005	0.037	0.000	0.02	0.889
X ₃ * X ₃	-0.024	-0.012	0.037	0.000	0.11	0.752
X ₁ * X ₂	-0.238	-0.119	0.022	0.113	30.17	0.001
X ₁ * X ₃	-0.008	-0.004	0.022	0.000	0.04	0.856
X ₂ * X ₃	-0.046	-0.023	0.022	0.004	1.14	0.321
Error				0.026		
Lack-of-Fit				0.017	0.70	0.675
Pure Error				0.009		
Total				0.639		
R-sq	95.90%	R-sq (adj)	90.62%	R-sq (pred)	74.13%	

According to the ANOVA result for DPPH radical scavenging activity, X₁, X₃, X₁², X₃², X₁*X₂, X₂*X₃ coefficients are significant and all other terms are insignificant. The polynomial equation was derived through fitting the individual response to the equation 2 and given below in the uncoded form:

$$\text{DPPH radical scavenging activity (Y}_2\text{; } \mu\text{mol AAE/g)} = 16.573 + 2.818 X_1 - 0.161 X_2 + 5.680 X_3 - 5.26 X_1^2 - 0.37 X_2^2 + 3.05 X_3^2 - 3.670 X_1 X_2 + 1.096 X_1 X_3 + 1.650 X_2 X_3$$

Table 3.2(b): Analysis of variance table for DPPH radical scavenging activity

Term	Effect	Coef	SE Coef	Adj SS	F-Value	P-Value
Constant		16.573	0.795	629.710	20.26	0.000
X ₁	5.636	2.818	0.588	79.416	22.99	0.002
X ₂	-0.323	-0.161	0.588	0.260	0.08	0.792
X ₃	11.360	5.680	0.588	322.650	93.42	0.000
X ₁ * X ₁	-10.52	-5.26	1.14	74.072	21.45	0.002
X ₂ * X ₂	-0.74	-0.37	1.14	0.368	0.11	0.754
X ₃ * X ₃	6.09	3.05	1.14	24.849	7.19	0.031
X ₁ * X ₂	-7.340	-3.670	0.657	107.746	31.20	0.001
X ₁ * X ₃	2.191	1.096	0.657	9.605	2.78	0.139
X ₂ * X ₃	3.301	1.650	0.657	21.787	6.31	0.040
Error				24.176		
Lack-of-Fit				13.695	0.52	0.758
Pure Error				10.481		
Total				653.886		
R-sq	96.30%	R-sq (adj)	91.55%	R-sq (pred)	79.80%	

Various descriptive statistics, such as p value and R-squared, determined using Analysis of variance (ANOVA) procedure was used to characterize the derived model. Low model p value (<0.001) and high model F value (Specific volume – 18.18; DPPH radical scavenging activity – 20.26) implies statistical significance of models. The non significant lack of fit and a high R² (Specific volume – 0.959; DPPH radical scavenging activity – 0.963) and adjusted R² (Specific volume – 0.906; DPPH radical scavenging activity – 0.916) values suggest that the fitted regression equation adequately models the response variable and model explains most of the variation observed in it. The insignificant lacks of fit values for both models indicate the goodness of fit and suitability of the regression model.

3.3.2. Effect of factors on specific volume: Characteristics of bread which satisfies the consumers first are volume and weight of the loaf. Based on the fact specific volume which is the ratio of these two properties, has been chosen as a response. Proofing height directly related with loaf volume and also quantity and quality of gluten protein present

in bread dough. Whereas loaf weight depends on the amount of dough baked and during baking degree of moisture and carbon dioxide loss from the loaf [80]. Here with increasing addition level of RMPP specific volume was decreased (Figure 3.1(a)). May be the dietary fibre content of RMPP diluted the gluten protein present in dough by thinning and breakage of the protein fibrils. As a result development of gas cells were restricted and the loaf volume was decreased [6][14][81]. According to the experimental design RMPP was added at different levels by replacing the equal amount of wheat flour which is another reason of weak gluten structure. The phenolic content of RMPP may also responsible for the reduced specific volume. Phenolic compounds primarily decreased mixing time and dough strength by interchanging disulfide bond with thiol group [82].

Baking temperature and time show combined effect on specific volume. As per response surface plot (Figure 3.1(a)) either high temperature short time or low temperature high time is good to increase bread specific volume. Evaporation of moisture from the dough during baking depends on the baking conditions. High temperature with long time is the reason of excessive moisture loss during baking [83][80].

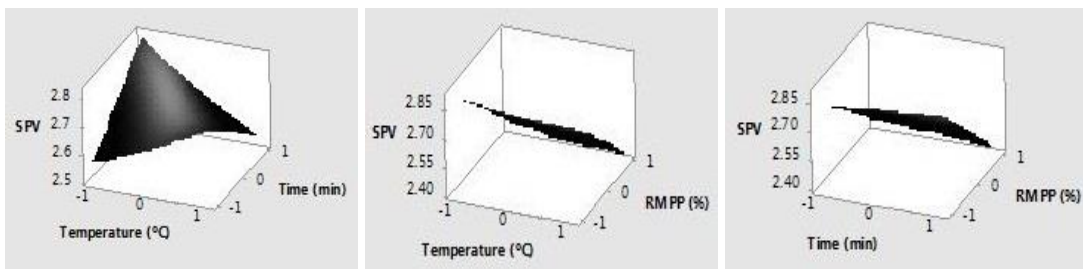


Figure 3.1(a): Response surface plots showing the effect of process parameters on specific volume; here hold values are Temperature (°C) 0, Time (min) 0 and RMPP (%) 0.

3.3.3. Effect of factors on DPPH radical scavenging activity:

Response surface plots show the positive linear effect of RMPP addition level on DPPH radical scavenging activity. In bakery products antioxidant potential is highly depends on recipe. Here phenolic content of RMPP (see chapter2) is responsible for this change in antioxidant activity.

According to the model, baking temperature had a positive linear effect on DPPH radical scavenging activity. Whereas in combination with baking time, for higher and lower baking temperature, time needs to be minimised and maximised respectively. Baking

involves both formation and destruction of antioxidant compounds [4]. Liberation of bound phenolics due to high baking temperature and formation of heat induced compounds from the Maillard reaction possibly best reason behind increased antioxidant activity during baking [84][85][86][87]. Also phenolic compounds are unstable at high heat which reduces the total antioxidant activity [13][20][7]. Again baking time and RMPP addition level had a positive interaction effect on antioxidant activity. At constant temperature when baking time is longer there to compensate the thermal and oxidative degradation of phenolic compounds, quantity of RMPP needs to increase. On the other hand, when baking time was low, due to minimum chances of phenolic degradation it is not required to increase the RMPP addition level.

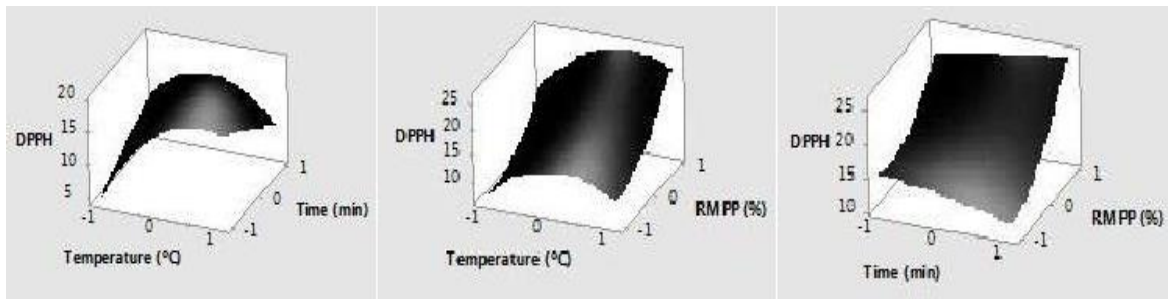


Figure 3.1(b): Response surface plots showing the effect of process parameters on DPPH radical scavenging activity; here hold values are Temperature (°C) 0, Time (min) 0 and RMPP (%) 0.

3.3.4. Both Optimization of processing parameters along with validation of the optimised conditions:

Derringer's desired function methodology was used to identify optimised processing conditions based on FCCD results, where DPPH radical scavenging activity was maximum and specific volume was 2.500 cm³/g [88]. From optimal points a desirability ramp was developed (Figure 3.2). Optimum processing conditions were, baking at 226°C for 19 min with 7% RMPP. Predicted DPPH radical scavenging activity at these conditions was 25.868 µmol AAE/g with composite desirability value of 0.999.

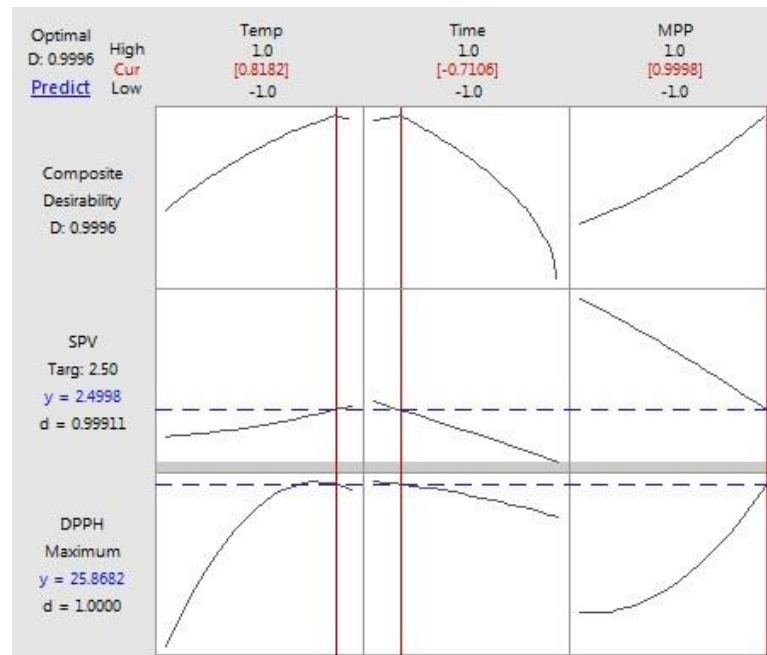


Figure 3.2: Desirability ramp for optimisation

To study the accuracy of this optimisation process, three bread samples were prepared under predicted optimal processing conditions. Both specific volume and DPPH radical scavenging activity of those three bread samples were measured. From Derringer's desired function method the experimental mean values along with predicted values obtained are shown in Table 3.3. Experimental values of both the responses were in agreement with the predicted values. The good correlation between experimental values and predicted values indicate the reliability of FCCD incorporate desirability function method and it could be used in this type of bread enrichment process to obtain maximum antioxidant activity with desired physical attribute.

Table 3.3: Experimental and predicted values of the responses at optimised conditions

Optimised conditions			Specific volume (cm ³ /g)		DPPH radical scavenging activity (µmol AAE/g)	
Baking temperature (°C)	Baking time (min)	RMPP addition level (%)	Predicted	Experimental	Predicted	Experimental
236	13	7	2.500	2.445±0.075	25.868	24.672±0.761

3.3.5. Comparison between control and optimised bread:

To study the role of RMPP as a functional ingredient, total dietary fiber (TDF) and total phenolics content (TPC) of both control and optimised bread samples were measured and compared (Table 3.4(a)). As a result 7% RMPP addition, in comparison with control

bread, TDF was increased by more than two times. Similar results were observed when 30% wheat flour was substituted with each rice bran [89] and whole wheat flour [21]. Also TPC value was increased from 0.209mg GAE/g to 1.959mg GAE/g, just like 7.5% RMPP added macaroni [51].

In sensory evaluation (Table 3.4(a)), fruity flavour of RMPP was able to improve the aroma of optimised bread, as observed for pomegranate whole fruit bagasse added bread [73] and RMPP added whole wheat bread [77]. According to the panellists' scores appearance, colour and taste of both control and optimised bread were insignificantly different to each other. Though in comparison with control bread specific volume value (Control- 2.62 cm³/g; Optimised-2.45 cm³/g) was lower and hardness value (Control-520.4g; Optimised-615.6g) was higher for optimised bread.

These changes in the value of specific volume and hardness do not show any negative effect on consumer acceptability. As per literature review [67][70][71][72] up to a certain level of changes in physical parameters due to the utilisation of natural antioxidant sources do not affect consumer preference. Based on these studies, 2.500 cm³/g was selected as target specific volume in optimisation process. Though in comparison with grape pomace powder added white bread [90], RMPP added whole wheat bread [77] and pomegranate whole fruit bagasse added bread [73] here specific volume and hardness were improved through the optimization process. Whiteness index of bread crumb also decreased from 67.86 to 56.01. Like anka enriched bread [22], panellists accept the darker bread crumb (Figure 3.3). Altogether this enrichment process showed an insignificant change on overall acceptability which indicates a good consumer acceptability of RMPP added white bread.



Control Bread



Optimised Bread

Figure 3.3: Cross sectional images of bread loaves

Table 3.4(a): Comparison between control and optimised bread

Parameters		Control	Optimised
Total dietary fiber (%)		2.613±0.103*	6.417±0.202*
Total phenolics content (mg GAE/g)		0.209±0.006*	1.959±0.025*
Sensory parameters	Appearance	7.247±0.100	7.177±0.075
	Colour	7.650±0.050	7.623±0.055
	Aroma	7.100±0.200*	7.580±0.061*
	Taste	7.413±0.103	7.250±0.050
	Overall acceptability	8.200±0.265	8.067±0.176

Mean values with superscripts (*) within the same row are significantly different (p≤0.05)

The recrystallization of starch molecules renders bread by increasing hardness and decreasing palatability during storage. The whole process known as staling, a great concern for bakery industry. Here, a comparative storage study between control and optimised bread was performed, by measuring hardness of bread loaves up to 5 days. Bread hardening kinetics was explained through Avrami model, based on starch retrogradation [79]. Kinetic parameters T_0 , T_∞ , k , and n , obtained from the fitted model, are shown in Table 3.4 (b). At 0 day crumb hardness was higher for optimised bread but at 5th day value was lower. As per Avrami equation, nature of bread hardening kinetics depends on both k and n values. In this study, the optimised sample had a lower value of k , which indicates a delayed retrogradation process in the sample. Whereas n value was higher, that indicates more sigmoidally shaped curve, with small increments in crumb firmness in first storage period and comparatively higher increments in intermediate periods [91]. Thus in comparison with control, optimised bread showed better storage stability. RMPP possesses high WHC (6.221±0.052 g/g) due to the presence of dietary fiber, which may help to maintain the crumb structure during storage by restricting the movement of moisture [78]. Also Siriamornpun et al. suggested phenolic compounds in RMPP of kaew variety were able to decrease the retrogradation in rice flour [92].

Table 3.4(b): Comparison between control and optimised bread

	Parameters	Control	Optimised
Storage study	T₀ (g)	520.4	615.6
	T_∞ (g)	2462.4	1954.3
	K	0.005	0.002
	n	4.171	4.953

3.4. Conclusion:

In this study three-level three-factor face-centered central composite design (FCCD) was successfully employed to optimise ripe mango peel enriched white bread preparation. According to the result, in this study, responses were significantly affected by all the selected factors. Specific volume was decreased with RMPP addition level and a negative interaction effect was observed by baking temperature and baking time. DPPH radical scavenging activity was improved with RMPP addition. Like specific volume also for antioxidant activity same type of effect was observed by baking temperature and baking time. Baking time with RMPP showed a positive interaction effect on antioxidant activity also. Through Derringer's desired function method optimised processing condition was obtained and as follows: baking at 226°C for 19 min with 7% RMPP. At this optimised condition specific volume and DPPH radical scavenging activity were 2.500 cm³/g and 25.868 μmol AAE/g respectively. Under this optimal condition experimental value was in agreement with predicted value. At the same time optimised bread was equally preferred like control bread. Along with antioxidant activity RMPP was able to improve storage stability of the bread.

CHAPTER: 4

4. Combined effect of raw and ripe mango peel powder on the quality attributes of gluten free bread

4.1. Introduction:

In the recent years gluten free products have received much of an attention due to the high rate of the celiac disease due to their persistent adherence on the gluten diets. It has been found that the celiac disease has increased in areas of the developing world, such as India, Middle East and North Africa, contributing to childhood morbidity and mortality [93]. Thus, the development of healthier and better quality gluten-free products would greatly improve the quality of life for celiac patients by treating their disorder.

Presence of two unique proteins – gliadin and glutenin make wheat flour an indispensable ingredient for bakery industry. Gluten – a combination of these two proteins, forms during dough formation and obtains its toughness from glutenin and elasticity from gliadin. Besides, other ingredients and mechanical agitation also help to form the film like structure of gluten that envelops the starch granules in the dough. That complex structure of gluten is the key feature of dough which retains gas and gives the desired volume of the final product. Among all bakery products, the consumer acceptability of bread mostly depends on its volume. So the absence of gluten in the bread formulations poses technological challenges and mainly affects the rheological and textural characteristics of the dough and final product [94]. Different types of cereals (rice, sorghum), pseudocereals (quinoa, amaranth), starch (potato, corn) and proteins (egg white, soy) along with hydrocolloids (xanthan, Hydroxypropyl methylcellulose), emulsifiers (Mono- and diglycerides of diacetyl tartaric acid ester), enzymes (transglutaminase) are successfully used in gluten free bread (GFB) making for a better quality product outcome [93].

Both raw and ripe mango processing industries incur a huge amount of peel as bio-waste. As per literature review [49][62][63][95] both raw and ripe mango peel are good source of polyphenols and dietary fiber. Ripe mango peel powder already used in gluten free cookies to enhance the antioxidant activity and total dietary fiber [96]. Lot of studies [97] showed utilisation of fiber rich flour in gluten free bread preparation was able to

improve the texture. So, use of mango peel powder in GFB preparation will improve texture of bread along with enhanced antioxidant activity. In comparison with ripe mango peel, utilisation of raw mango peel is still very limited. Fruity flavour of ripe mango peel powder was able to improve the flavour of whole wheat bread [77]. Whereas, higher value of polyphenols in raw mango peel give better antioxidant activity but bitter taste in the final product [98]. Thus the combined effect of the raw and ripe mango peel in equal proportion may compensate the negative impacts of the individual peel and brings forth a good quality GFB.

Objective of the present study was incorporation of both raw and ripe Mango peel powder in equal proportion at 2%, 4% and 6% level in the GFB formulation and to assess the influence of fibres on flour and dough characteristics along with the physical, chemical (antioxidant) and sensory properties of the final product.

4.2. Materials and methods:

4.2.1. Materials: Gluten free flours like sorghum and rice, sugar, salt, refined oil, egg and instant dry yeast (Best Products, Kolkata) were bought from the market at Jadavpur in Kolkata, India. Other ingredient was Guar gum (HiMedia Laboratories Pvt. Ltd., Mumbai, India) as a thickening agent.

4.2.2. Chemicals: Gallic acid (sd fine-chem Ltd., Mumbai, India), Folin-Ciocalteu reagent, ethanol, sodium carbonate from Merck Specialities Pvt. Ltd. at Mumbai, sodium nitrate, aluminium chloride, sodium hydroxide and 2, 2-diphenyl-1-picrylhydrazyl (DPPH) from Sigma-Aldrich, St. Louis, at MO, USA, were used for chemical analysis.

4.2.3. Preparation of Mango peel powder (MPP): Same as section: 2.2.3.

4.2.4. Preparation of composite flour: The blending of flour was obtained by mixture of sorghum flour (SF), rice flour (RF) in equal proportion (50:50) (SF+RF) with incorporation of 2%, 4%, 6% Mango peel powder (MPP) for bread preparation which was denoted as SF+RF+2% MPP, SF+RF+4% MPP, SF+RF+6% MPP respectively.

4.2.5. Bread preparation: The control bread was prepared by the combination of sorghum flour (SF) and rice flour (RF) in equal ratio (1:1). Straight dough method was employed for the dough preparation. The SF+RF along with the instant dry yeast and sugar were kneaded with hand mixer (Philips Daily Collection-HR1459) with 175ml of

water followed by the other ingredients like egg white, guar gum and oil. Salt was added at the last stage of kneading for better yeast activity. The prepared dough was then put in the mould panlubricated with oil and was then covered with a wet muslin cloth for proofing. The proofed dough was then put in rotatory oven (CM HS108) bought from Chanmag Bakery Machine Co. Ltd., Taipei, at Taiwan, and baked at $180\pm 2^{\circ}\text{C}$ around 60 min. After that baked loaf was further cooled for 1h prior to further physical and chemical analysis. The formulated flours with 2%, 4% and 6% MPP (raw and ripe mango peel powder in equal proportion) follow the same bread making procedure as that of control bread sample [99].

4.2.6. Moisture content: Same as 2.2.8.

4.2.7. pH: The pH of the different flour samples were measured using a pH meter (Thermo Orion Basic pH Meter, Model 420A pH/mV/ORP/temperature meter).

4.2.8. Color: Same as 2.2.8.

4.2.9. Water absorption capacity: The water absorption capacity of the flours was analyzed from Tsai's method (1998) [100] with slight modifications in it. The weight of the residue containing tube is recorded and water absorption capacity (WAC) is calculated by the following equation:

$$\text{WAC} = \frac{\text{weight of the sample after centrifugation} - \text{weight of the sample before centrifugation}}{\text{weight of the sample}} \quad (1)$$

4.2.10. Oil absorption capacity: The oil absorption capacity of flour samples were analyzed by the Beuchat method (1997) [101] with some modifications in it. The oil absorption capacity is calculated by the formula stated below:

$$\text{Oil absorption capacity} = \frac{\text{Weight of the sediment}}{\text{Weight of the sample}} \quad (2)$$

4.2.11. Swelling power: The swelling power of flour samples were analysed by the method of Leach et al. (1959) [102] with some modifications in it. The swelling power (SP) of the samples is calculated by the following equation:

$$\text{Swelling power} = \frac{\text{Weight of the wet mass of the sediment}}{\text{Weight of the dry sample}} \quad (3)$$

4.2.12. Rheology of dough: The dough samples were prepared as per formulation but without addition of yeast and the rheological behaviour were monitored in a rheometer

(Physica MCR 51; Anton Paar, Graz, Austria) with RheoPlus software package (version 2.65). 49.986mm parallel plates were used to maintain 3mm gap during sample analysis. A water circular device (Neslab RTE 7; Thermo Electron Corporation, Bremen, Germany) were used to maintain 25°C temperature. Before measuring samples were rested for 300 seconds and excess of samples were removed off. To identify the linear viscoelastic region under constant frequency 1 Hz strain sweep test was done keeping shear stress within 0.1-100 Pa. Within the linear viscoelastic region of all dough samples, one common stress value was taken to perform frequency sweep test within (0.1 Hz -100 Hz) [103]. Two graphs were prepared against the frequency (Hz), first with storage modulus (G') and second one with loss modulus (G'').

4.2.13. Specific volume: Same as 2.2.8.

4.2.14. Texture Analysis: Same as 3.2.11.

4.2.15. Preparation of the extract: Same as 2.2.6.

4.2.16. Total phenolic content: Same as 2.2.6.

4.2.17. Total flavonoid content: The total flavonoid content of the bread samples was determined with slight modifications of Xu and Chang 2007 [104]. Total flavonoid contents were expressed as milligrams of catechin equivalents (CAE) per gram of sample.

4.2.18. DPPH radical scavenging assay: Same as 2.2.6.

4.2.19. Sensory evaluation: Same as 3.2.10.

4.2.20. Statistical analysis: All the studies were replicated 3 times and the means were reported. All the experimental data were analyzed statistically for analysis of variance (ANOVA) with Minitab 17.0 (Minitab, Inc., Pennsylvania, PA). Means were compared by Tukey's test at a significance level of $p \leq 0.05$.

4.3. Results and discussions:

4.3.1. Characterization of flours and its formulations:

The physical properties of flours and its formulations are tabulated in Table 4.1. It was evident that the moisture content (%) of the SF was 6.63 ± 0.28 and in RF was 7.95 ± 0.36 both had a markedly significant difference ($p \leq 0.05$). The moisture content of the

combination of SF and RF in equal proportions was 7.63 ± 0.08 . There was an increasing trend of moisture content in the flours formulated with 2%, 4%, and 6% MPP (equal proportion of both raw and ripe) respectively. This increase was due to the presence of some amount of moisture in the MPP itself. Though, statistically this change was insignificant to each other. Generally the acceptable moisture content of flour for bakery products ranges from 14-15%. Here the increasing trend of moisture of the samples is around 10%. This allows the flour formulations to have a better shelf life and it also correlates with water absorption capacity of the flour as well.

However there was a slight increase in the pH of the flours formulated with MPP compared to that of the SF, RF, SF+RF respectively with no such significant difference. In spite of some intrinsic acidic character of raw mango peel the pH trend increased and was nearing to neutral pH. This may be due to presence of ripe mango peel along with the raw in equal proportions which masks the acidic nature of the raw mango peel.

Color of flour has an impact on the crumb color of the final product which is an important parameter to characterize the GFB from the wheat bread. The whiteness index (WI) of the rice flour was reported to be highest in the Table 4.1 whereas the combination of RF and SF flours had a whiteness index (WI) greater than that of SF alone. In the formulated flours of 2%, 4% and 6% there was a decreasing trend in the whiteness index (WI) due to the addition of the MPP. There was no such change in a^* value whereas the L^* and b^* values showed a marked difference from (85.95 ± 0.19 to 82.82 ± 0.06) and (14.37 ± 0.28 to 15.05 ± 0.14) respectively. This may be due to the reason that MPP has some enzymes like polyphenol oxidase and peroxidase which are responsible for enzymatic browning during processing that result in decreasing the lightness of the MPP [105]. These L^* , a^* and b^* values had an overall impact on the whiteness index of the flour formulations and thus a decreasing trend of whiteness index was reported in the Table 4.1.

Water absorption capacity of the flour is an important parameter for the dough development and quality of the final product. The water absorption capacity and the moisture content of the flour are interdependent to each other.

Table 4.1: Physical characteristics of different flour samples

Samples	Moisture (%)	pH	WI
SF	6.63±0.28 ^c	6.50±0.29 ^a	79.80±0.05 ^d
RF	7.95±0.36 ^b	6.58±0.29 ^a	88.71 ±0.06 ^a
SF+RF	7.63±0.08 ^b	6.47±0.28 ^a	82.51±0.06 ^b
SF+RF+2%MPP	8.67±0.37 ^a	6.87±0.33 ^a	81.18± 0.07 ^c
SF+RF+4%MPP	8.91±0.18 ^a	6.77±0.32 ^a	78.50±0.06 ^e
SF+RF+6%MPP	9.15±0.07 ^a	6.61±0.32 ^a	77.07±0.07 ^f

Mean with different letters within a column is significantly different ($p \leq 0.05$)

In Table 4.1 it was clearly reported that moisture content of SF was lower than RF and in combination of the SF+RF but at the same time the water absorption capacity was of the order SF>SF+RF and RF. This is because the lower the moisture content in the flour the higher is its water absorption capacity [106]. On addition of the MPP in flours there was an increasing trend in the water absorption quality. This is due to the hydrophilic nature of the fibre present in MPP and the interactions of water with the hydroxyl groups of those polysaccharides through hydrogen bonding [105].

Oil absorption capacity of flours is another important functional factor as it attributes to the flavor retention and mouthfeel of the final product. In general oil absorption capacity of the flours were less compared to the WAC. The OAC of the SF and RF were higher than its combination. The formulated sample showed an increase in oil absorption capacity when compared with SF+RF combination. This may be due to the low fat content in MPP as well as the hydrophobic part of protein entraps oil as a result increases the oil absorption capacity [107]. This ensures that the final formulated product to have a better shelf life with low chances of rancidity to occur.

The swelling power shows a significant decrease ($P \leq 0.05$) of formulated flours with 6%, 4% and 2% MPP compared to the SF, RF and SF+RF in combination. RF shows the maximum swelling power (7.83±0.25) due to the high quality of starch and its linear branching of amylopectin [108]. The particle size and fibre content of MPP has an impact on the decrease of the swelling power [109]. The protein and fat content of the blended flour with MPP at 2%, 4% and 6% formulation may also have an impact to reduce the swelling power due to inhibition of starch gelatinization [110].

Table 4.2: Functional characteristics of different flour samples

Samples	WAC	OAC	SP
SF	3.25±0.09 ^b	2.31±0.12 ^a	6.97±0.35 ^{bc}
RF	2.97±0.11 ^c	2.06±0.10 ^{ab}	7.83±0.25 ^a
SF+RF	2.97±0.13 ^c	1.87±0.09 ^b	7.82±0.22 ^{ab}
SF+RF+2%MPP	3.24±0.05 ^b	2.04±0.10 ^{ab}	6.76±0.36 ^{cd}
SF+RF+4%MPP	3.46±0.10 ^b	2.28±0.11 ^a	5.98±0.31 ^d
SF+RF+6%MPP	3.85±0.04 ^a	2.32±0.12 ^a	5.15±0.28 ^e

Mean with different letters within a column is significantly different ($p \leq 0.05$)

4.3.2. Dough Rheology: The consistency of the enriched dough samples with MPP was higher compared to the control sample due to the fibre content of the MPP. In presence of soluble fibre lower loss of tangent ($\tan \delta$) indicating the dough to exhibit higher elasticity compare to the control. The G' shows a decreasing trend than the G'' in Fig 4.1 only for 6% MPP formulation in spite of the insoluble fibres present in higher amounts [97]. This can be explained by correlating with WAC and SP values in Table 4.2. The fibres in 6% formulated dough was higher which allows large amount of water to be absorbed and also masks the swelling property of the flour thereby decreases the SP value. Large amount of water in dough makes the dough sticky and viscous rather than elastic in nature [51].

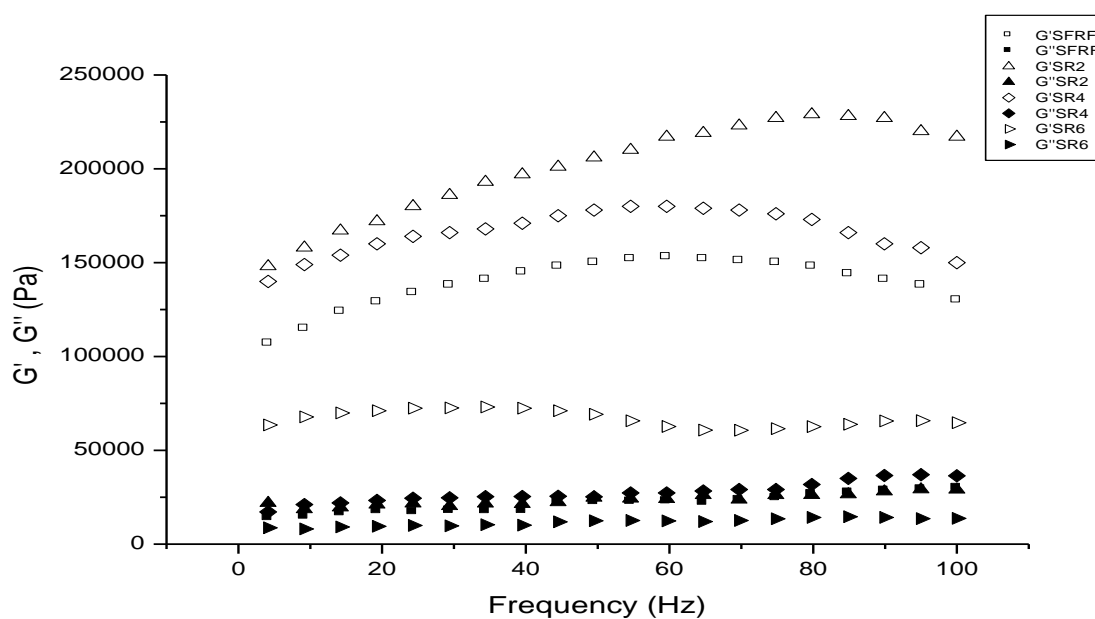


Figure 4.1: Viscoelastic properties of different types of GFB samples

It can be inferred that after a certain level of formulation the dough no longer remains viscoelastic and the final product quality is much inferior and unacceptable. Since the viscoelastic nature predominates the final structure of the bread thereby dough samples of 2% and 4% showing the highest viscoelastic nature.

4.3.3. Physical properties of Gluten free bread: The physical parameters of the bread samples are tabulated in table 4.3. The height and specific volume is an important quality parameter of the bread loaf. There exists a relationship between height and specific volume of the loaf to that of the water absorption capacity of the dough. In comparison with control, an insignificant change in the height and significant improvement in the specific volume at 2% and 4% MPP enriched bread samples were observed. This is due to the presence of fibre content of MPP which form a flim like structure with the starch matrix of rice flour and also guar gum to a certain extent [97]. After a significant increase, specific volume tends to decrease on incorporation of MPP at 6% formulation. This is due to the lower consistency of the dough resulting in unstable air vacuoles within the crumb structure owing to the failure of retention of carbon dioxide gas bubbles resulting in a decrease in the specific volume [97]. The dietary fibre in MPP has high water binding capacity and allows enhances water absorption capacity of flour as a result increases the moisture content of the formulated bread samples upto 6% MPP significantly ($P \leq 0.05$) to that of control sample. The explanation can be further supported with the values of WAC of the flour samples reported in Table 4.3. Color of the final bread is another important parameter for consumer acceptability. The color of the crust is measured in terms of brownness index (BI) and that of crumb is by whiteness index (WI). An insignificant change in BI value is observed. MPP is a rich source of polyphenols which acts as a substrate for enzymatic browning and leads to the decrease in WI of the formulated bread samples [101]. The whiteness index (WI) of the bread samples has a significant decrease from (64.4633 ± 0.015 to 53.17 ± 0.08). The L^* values in the formulated bread samples with that of the control bread (70.80 ± 0.21 to 53.90 ± 1.24) and the b^* values from control to 4% formulation (16.80 ± 0.48 to 19.38 ± 1.28) indicating the decrease in lightness and increase in yellowness respectively. Thus formulated bread samples have a darker crumb color compared to the control sample. The 6% formulated bread sample shows higher WI value than 4% may be due to improper baking in presence of high fiber content. With all other physical parameters as mentioned before hardness of bread crumb was reported in Table 4.3 as an indicator of bread texture. An

inverse correlation between the specific volume and hardness of the bread explains the changes of the texture profile of the bread samples. At 4% level of MPP the volume of the bread is maximum with a softer crumb texture. On addition of 6% MPP there is a significant increase in hardness of the bread compared to the control and other formulated samples were dense with more compact cells because fiber was present in high amount.

Table 4.3: Physical parameters of MPP added gluten free bread

Samples	SF+RF	SF+RF+2%MPP	SF+RF+4%MPP	SF+RF+6%MPP
Height (cm)	5.61±0.28 ^a	5.8±0.29 ^a	6.0±0.3 ^a	5.63±0.29 ^a
Specific volume (cm ³ /g)	1.25±0.062 ^c	1.41±0.055 ^b	1.57±0.061 ^a	1.15±0.053 ^c
Moisture content (%)	45.00±0.455 ^c	47.36±0.862 ^b	48.91±0.854 ^a	49.50±0.934 ^a
Colour	WI	64.46±0.02 ^a	54.59±0.58 ^b	53.17±0.08 ^c
	BI	587.68±0.15 ^a	587.70±0.26 ^a	587.85±0.04 ^a
Hardness	2231.99±45.093 ^a	1921.37±37.600 ^b	1775.37±28.612 ^c	2285.00±50.023 ^a

Mean with different letters within a row is significantly different ($p \leq 0.05$)

4.3.4. Antioxidant activity of Gluten free bread: Total phenolic content of the samples were estimated by Folin-Ciocalteu method. There was a linear significant increase ($P \leq 0.05$) in the phenolic content of the bread samples enriched with MPP. It ranges from 0.18mgGAE/g to 1.86mgGAE/g in control and 6% level respectively. Incorporation of MPP results in 9.15 fold increase of the phenolic content in the bread samples. This is due to the presence of the high phenolic content in the raw and ripe mango peel which ranges from 55 to 110 mg/g peel [63]. In spite of loss of polyphenols during baking there is still an increasing trend of the bread samples enriched with MPP.

At higher level of MPP in bread sample the flavonoid component was increased from 0.08mgCE/g-0.14mgCE/g in control bread and 6% MPP level respectively. Mango peel is rich in flavonoids [111] which results in the increase of the flavonoid content of the bread samples.

Antioxidant activity of the MPP added GFB samples were done through DPPH assay. At 517nm, DPPH a stable free radical has a characteristic absorption. The scavenging potential of the antioxidants is due to its hydrogen donating ability and the time required for discoloration of DPPH reflects free radical scavenging activity of the analysed sample. The increase in the scavenging potential of the bread samples formulated with MPP from 0.33% to 34.06% is due to the higher amount of the polyphenols and flavonoids present in MPP. MPP enhances both the nutritive as well as nutraceutical value of the bread samples due to its high phenolic content as well as its antioxidant activity.

Table 4.4: Antioxidant activity of MPP added gluten free bread

Samples	Total phenolic content (mg GAE/g)	Total flavonoid content (mg CE/g)	DPPH inhibition (%)
SF+RF	0.18±0.015 ^d	0.08±0.007 ^c	0.33±0.277 ^d
SF+RF+2%MPP	0.64±0.053 ^c	0.09±0.004 ^c	5.72±0.571 ^c
SF+RF+4%MPP	1.24±0.031 ^b	0.12±0.009 ^b	15.61±1.520 ^b
SF+RF+6%MPP	1.86±0.299 ^a	0.14±0.012 ^a	34.06±1.981 ^a

Mean with different letters within a column is significantly different ($p \leq 0.05$)

4.3.5. Sensory parameters of gluten free bread: Bread samples prepared with MPP at different levels were subjected for sensory evaluation for appearance, color, flavour, taste and overall acceptability. The appearance upto 4% MPP level have similar scores and are more acceptable to that of the control bread. On increasing level of MPP upto 4% the intensity of whiteness decreases which is supported by the WI values in Table 4.3. On increased level of 6% MPP poor crumb appearance is observed due to the fibrous structure making the network dense thereby decreasing the porosity of the loaf. The taste and flavour of the bread samples is enhanced on formulating it with MPP as it has a pleasant characteristic mango flavour. On increasing the level upto 6% there is a slight bitter after taste in the sample due to the high polyphenolic content. These data resembles to that of MPP formulated biscuit and whole wheat bread [96][98]. In comparison with raw mango peel added whole wheat bread here raw MPP addition level were improved from 1% to 2% by the utilization of Raw and Ripe MPP together. The overall acceptability of the bread samples shows highest score for 4% formulated sample as compared to control, 2% and 6% respectively. The lower score for 6% formulated bread

is due to the undesirable crumb appearance, color and taste. Thus it can be concluded that formulating the sample with 4% MPP gives the best sensory attributes.

Table 4.5: Sensory parameters of MPP added gluten free bread

Samples	SF+RF	SF+RF+ 2%MPP	SF+RF+ 4%MPP	SF+RF+ 6%MPP
Appearance	7.3±0.53 ^b	8.2±0.56 ^a	8.4±0.33 ^a	6.3±0.29 ^c
Color	8.03±0.05 ^a	7.00±0.50 ^b	6.50±0.50 ^b	4.50±0.50 ^c
Flavor	6.67±0.58 ^b	7.77±0.25 ^a	8.10±0.30 ^a	6.00±0.50 ^b
Taste	7.33±0.57 ^b	7.93±0.12 ^{ab}	8.13±0.32 ^a	5.83±0.29 ^c
Overall Acceptability	6.90±0.32 ^c	7.56±0.25 ^b	8.07±0.05 ^a	5.13±0.05 ^d

Mean with different letters within a row is significantly different ($p \leq 0.05$)

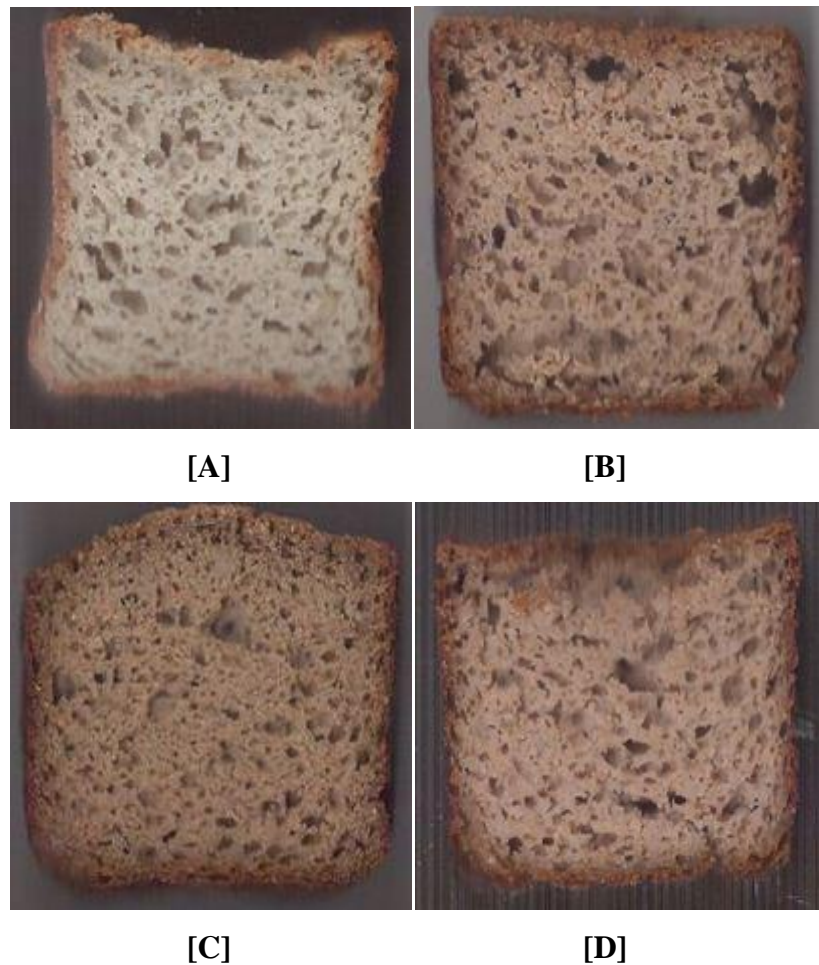


Figure 4.2: Images of Gluten Free Bread (GFB).

SF+RF (A), SF+RF+2% MPP (B), SF+RF+4% MPP (C), SF+RF+6% MPP (D)

4.3. Conclusion:

The physical, functional and rheological properties of the flour and dough samples fortified with raw and ripe MPP at 2%, 4%, and 6% level for gluten free bread preparations were studied to correlate with the physical, chemical and organoleptic characteristics of the final bread. The dough sample at 4% level MPP exhibits maximum viscoelasticity compared to that of the control, 2%, 6% samples. Generally incorporation of mango peel powder (MPP) at increased level improves the antioxidant properties but deteriorates the physical and sensory parameters simultaneously after reaching a certain level. Gluten free bread with 4% MPP shows maximum increase for specific volume, whereas lowest value for crumb hardness and thus sensory properties offering it to be the highest acceptable fortified bread. The novelty of this study is to incorporate both raw and ripe MPP in GFB formulations at different levels to enhance the physical and nutraceutical value of the final product.

FUTURE SCOPE

The study was conducted with three different types of bread-whole wheat bread, wheat bread and gluten free bread and the objective was to improve the existing antioxidant activity of the bread samples with natural antioxidant sources. Mango peel of “Himsagar” variety was selected as natural antioxidant source. This kind of fruit waste was easily available in summer. Mango peel was converted to powder using tray drying method and was used in different ratio in respective type of bread. In turn mango peel powder was successful to improve the antioxidant activity of each bread sample. Now a day the bread we consume has a limited use of natural antioxidant. So, in future, if it is possible to make such natural antioxidant rich bread, then it can easily capture the functional food market.

Along with that, parallel study was conducted to understand the additional effect of mango peel powder on the bread samples. By using ripe mango peel powder in whole wheat bread, it was possible to improve the taste and aroma of the bread as well as its antioxidant activity. Whole wheat bread is well known among the consumer due to its health benefit, on the other hand many dislike it due to its intense wheaty, yeasty flavour. With this study, the improvement of whole wheat bread may help to increase its popularity among the consumers though further study is required to improve the texture of whole wheat bread, which was degraded due to ripe mango peel powder addition.

As per literature review, it was observed that whenever natural antioxidant source was added to white bread, then its antioxidant activity started increasing gradually whereas the overall acceptability of the bread decreased due to the degradation in its physical property. To overcome this issue ripe mango peel powder added white bread preparation was optimized through Response surface methodology. Baking time, baking temperature and ripe mango peel powder addition level were selected as processing factor and specific volume and DPPH radical scavenging activity were selected as response. Baking time and baking temperature were considered as factor because both antioxidant activity and bread's physical property were dependent on that. In optimized processing condition the specific volume and DPPH radical scavenging activity of bread were 2.500 cm³/g and 25.868 μmol AAE/g respectively. Sensory evaluation of optimized bread and control bread showed similar overall acceptability. Along with that as an added advantage optimized bread showed better storage stability than control bread. It can be concluded

that, enrichment of bread with natural antioxidant sources using the optimization process would be helpful to get the desired output.

There is an increasing rate of gluten related disorder in last few years. It can be prevented only by excluding gluten from the regular diet. That's why gluten free bread is gaining popularity day by day. It is also a challenge for the food processors to produce gluten free bread. As per literature review, one of the best ways to overcome this limitation is incorporation of dietary fiber in gluten free bread formulation. That's why raw mango peel powder and ripe mango peel powder both were used in equal proportion as natural antioxidant and dietary fiber source. The ratio of soluble dietary fiber and insoluble dietary fiber was also very important along with the total dietary fiber content. Gluten free batter with 4% mango peel powder (Raw and Ripe in equal proportion) achieved maximum viscoelastic property. According to physical parameter, best quality bread *could be* obtained using the same additional level. Though antioxidant activity increased up to 6% gradually, yet 4% is considered as the best addition level as according to physical and sensory property it is the best. This dual effect of mango peel powder (Raw and Ripe in equal proportion) helped to make gluten free bread preparation easy and also improved the nutritional quality of that.

It is possible to improve the bread quality with the utilisation of mango peel powder. Also with that a huge fruit waste of mango processing can be recycled in the environment friendly way. Mango peel is considered as a waste and in this research work it has been processed in a very simple method (Peeling>Washing>Blanching>Drying>Grinding). For this reason, its utilization in bread processing can be considered as economical though detail cost analysis is required for the industry level implementation.

REFERENCES

1. R. Blomhoff, "Role of dietary phytochemicals in oxidative stress," in *Bioactive compounds in plants – benefits and risks for man and animals*, A. Bernhoft, Ed. The Norwegian Academy of Science and Letters, 2010, pp. 52–70.
2. A. Orphanides, V. Goulas, and V. Gekas, "Effect of Drying Method on the Phenolic Content and Antioxidant Capacity of Spearmint," *Czech J. Food Sci.*, vol. 31, no. 5, pp. 509–513, 2013.
3. M. Valko, D. Leibfritz, J. Moncol, M. T. D. Cronin, M. Mazur, and J. Telser, "Free radicals and antioxidants in normal physiological functions and human disease," *The International Journal of Biochemistry & Cell Biology*, vol. 39, pp. 44–84, 2007.
4. D. Diziki, R. Rozylo, U. Gawlik-Dziki, and M. Swieca, "Current trends in the enhancement of antioxidant activity of wheat bread by the addition of plant materials rich in phenolic compounds," *Food Sci. Technol.*, vol. 40, pp. 48–61, 2014.
5. F. Zhu, R. Sakulnak, and S. Wang, "Effect of black tea on antioxidant, textural, and sensory properties of Chinese steamed bread," *Food Chem.*, vol. 194, pp. 1217–1223, 2016.
6. M. Irakli, D. Katsantonis, and F. Kleisiaris, "Evaluation of quality attributes, nutraceutical components and antioxidant potential of wheat bread substituted with rice bran," *J. Cereal Sci.*, vol. 65, pp. 74–80, 2015.
7. M. Swieca, L. Seczyk, U. Gawlik-Dziki, and D. Dziki, "Bread enriched with quinoa leaves – The influence of protein – phenolics interactions on the nutritional and antioxidant quality," *Food Chem.*, vol. 162, pp. 54–62, 2014.
8. B. A. Acosta-Estrada, M. A. Lazo-Vélez, Y. Nava-Valdez, J. A. Gutiérrez-Uribe, and S. O. Serna-Saldívar, "Improvement of dietary fiber, ferulic acid and calcium contents in pan bread enriched with nejayote food additive from white maize (*Zea mays*)," *J. Cereal Sci.*, vol. 60, no. 1, pp. 264–269, 2014.
9. U. Gawlik-Dziki et al., "Anticancer and antioxidant activity of bread enriched with broccoli sprouts," *Biomed Res. Int.*, vol. 2014, 2014.

10. U. Gawlik-Dziki, M. Świeca, D. Dziki, B. Baraniak, J. Tomiło, and J. Czyz, "Quality and antioxidant properties of breads enriched with dry onion (*Allium cepa* L.) skin," *Food Chem.*, vol. 138, no. 2–3, pp. 1621–1628, 2013.
11. C. Dall'Asta, M. Cirlini, E. Morini, M. Rinaldi, T. Ganino, and E. Chiavaro, "Effect of chestnut flour supplementation on physico-chemical properties and volatiles in bread making," *LWT - Food Sci. Technol.*, vol. 53, no. 1, pp. 233–239, 2013.
12. L. Ho, N. Aziah, A. Aziz, and B. Azahari, "Physico-chemical characteristics and sensory evaluation of wheat bread partially substituted with banana (*Musa acuminata* X *balbisiana* cv . Awak) pseudo-stem flour," *Food Chem.*, vol. 139, no. 1–4, pp. 532– 539, 2013.
13. L. Das, U. Raychaudhuri, and R. Chakraborty, "Herbal fortification of common bread by fennel seeds," *Food Technol. Biotechnol.*, vol. 51, no. 4, pp. 434–440, 2013.
14. G. K. Dhillon and A. Kaur, "Quality Evaluation of Bread Incorporated With Different Levels of Cinnamon Powder," *Int. J. Food Sci. Nutr. Diet.*, vol. II, no. 7, pp. 2–7, 2013.
15. M. Blandino et al., "Nutritional and technological quality of bread enriched with an intermediated pearled wheat fraction," *Food Chem.*, vol. 141, no. 3, pp. 2549–2557, 2013.
16. C. G. Rizzello, R. Coda, F. Mazzacane, D. Minervini, and M. Gobbetti, "Micronized by-products from debranned durum wheat and sourdough fermentation enhanced the nutritional, textural and sensory features of bread," *Food Res. Int.*, vol. 46, no. 1, pp. 304–313, 2012.
17. A. Yousif, D. Nhepera, and S. Johnson, "Influence of sorghum flour addition on flat bread in vitro starch digestibility, antioxidant capacity and consumer acceptability," *Food Chem.*, vol. 134, no. 2, pp. 880–887, 2012.
18. L. Das, U. Raychaudhuri, and R. Chakraborty, "Supplementation of common white bread by coriander leaf powder," *Food Sci. Biotechnol.*, vol. 21, no. 2, pp. 425–433, 2012.

19. F. Balestra, E. Cocci, G. Pinnavaia, and S. Romani, "Evaluation of antioxidant, rheological and sensorial properties of wheat flour dough and bread containing ginger powder," *LWT - Food Sci. Technol.*, vol. 44, no. 3, pp. 700–705, 2011.
20. H. S. Lim, S. H. Park, K. Ghafoor, S. Y. Hwang, and J. Park, "Quality and antioxidant properties of bread containing turmeric (*Curcuma longa* L.) cultivated in South Korea," *Food Chem.*, vol. 124, no. 4, pp. 1577–1582, 2011.
21. S. Ragaee, I. Guzar, N. Dhull, and K. Seetharaman, "Effects of fiber addition on antioxidant capacity and nutritional quality of wheat bread," *LWT - Food Sci. Technol.*, vol. 44, no. 10, pp. 2147–2153, 2011.
22. Y. Tseng, J. Yang, C. Chen, and J. Mau, "Quality and antioxidant properties of anka- enriched bread," *Journal of Food Processing and Preservation*, vol. 35, pp. 518–523, 2011.
23. S. Mildner-Szkudlarz, R. Zawirska-Wojtasiak, A. Szwengiel, and M. Pacyński, "Use of grape by-product as a source of dietary fibre and phenolic compounds in sourdough mixed rye bread," *Int. J. Food Sci. Technol.*, vol. 46, no. 7, pp. 1485–1493, 2011.
24. J. Chlopicka, P. Pasko, S. Gorinstein, A. Jedryas, and P. Zagrodzki, "Total phenolic and total flavonoid content, antioxidant activity and sensory evaluation of pseudocereal breads," *LWT - Food Sci. Technol.*, vol. 46, no. 2, pp. 548–555, 2012.
25. X. Peng, J. Ma, K. W. Cheng, Y. Jiang, F. Chen, and M. Wang, "The effects of grape seed extract fortification on the antioxidant activity and quality attributes of bread," *Food Chem.*, vol. 119, no. 1, pp. 49–53, 2010.
26. B. Filipčev, L. Lević, M. Bodroža-Solarov, N. Mišljenovic, and G. Koprivica, "Quality characteristics and antioxidant properties of breads supplemented with sugar beet molasses-based ingredients," *Int. J. Food Prop.*, vol. 13, no. 5, pp. 1035–1053, 2010.
27. A. K. Holtekjølen, A. B. Bævre, M. Rødbotten, H. Berg, and S. H. Knutsen, "Antioxidant properties and sensory profiles of breads containing barley flour," *Food Chem.*, vol. 110, no. 2, pp. 414–421, 2008.

28. L. Fan, S. Zhang, L. Yu, and L. Ma, "Evaluation of antioxidant property and quality of breads containing *Auricularia auricula* polysaccharide flour," *Food Chem.*, vol. 101, no. 3, pp. 1158–1163, 2006.
29. C. Hsu, S. Hurang, W. Chen, Y. Weng, and C. Tseng, "Qualities and antioxidant properties of bread as affected by the incorporation of yam flour in the formulation," *International Journal of Food Science and Technology*, pp. 231–238, 2004.
30. M. Butu, S. Rodino, A. Butu, and M. Butnariu, "Screening of bioflavonoid and antioxidant activity of *Lens culinaris medikus*," *Dig. J. Nanomater. Biostructures*, vol. 9, no. 2, pp. 519–529, 2014.
31. M. Butnariu and A. Caunii, "Design management of functional foods for quality of lifeimprovement," *Ann. Agric. Environ. Med.*, vol. 20, no. 4, pp. 736–741, 2013.
32. M. Butnariu and C. Z. Coradini, "Evaluation of biologically active compounds from *Calendula officinalis* flowers using spectrophotometry," *Chem. Cent. J.*, vol. 6, no. 1, pp. 1–7, 2012.
33. E. S. M. Abdel-Aal and I. Rabalski, "Effect of baking on free and bound phenolic acids in wholegrain bakery products," *J. Cereal Sci.*, vol. 57, no. 3, pp. 312–318, 2013.
34. A. Ktenioudaki, L. Alvarez-Jubete, T. J. Smyth, K. Kilcawley, D. K. Rai, and E. Gallagher, "Application of bioprocessing techniques (sourdough fermentation and technological aids) for brewer's spent grain breads," *Food Res. Int.*, vol. 73, pp. 107–116, 2015.
35. C. G. Rizzello, M. Calasso, D. Campanella, M. De Angelis, and M. Gobbetti, "Use of sourdough fermentation and mixture of wheat, chickpea, lentil and bean flours for enhancing the nutritional, texture and sensory characteristics of white bread," *Int. J. Food Microbiol.*, vol. 180, pp. 78–87, 2014.
36. Y. Sasano, S. Takahashi, J. Shima, and H. Takagi, "Antioxidant N-acetyltransferase Mpr1/2 of industrial baker's yeast enhances fermentation ability after air-drying stress in bread dough," *Int. J. Food Microbiol.*, vol. 138, no. 1–2, pp. 181–185, 2010.

37. A. Michalska, M. Amigo-Benavent, H. Zielinski, and M. D. del Castillo, "Effect of bread making on formation of Maillard reaction products contributing to the overall antioxidant activity of rye bread," *J. Cereal Sci.*, vol. 48, no. 1, pp. 123–132, 2008.
38. M. Świeca, U. Gawlik-Dziki, D. Dziki, B. Baraniak, and J. Czyz, "The influence of protein-flavonoid interactions on protein digestibility in vitro and the antioxidant quality of breads enriched with onion skin," *Food Chem.*, vol. 141, no. 1, pp. 451–458, 2013.
39. C. A. Challacombe, E. S. M. Abdel-Aal, K. Seetharaman, and L. M. Duizer, "Influence of phenolic acid content on sensory perception of bread and crackers made from red or white wheat," *J. Cereal Sci.*, vol. 56, no. 2, pp. 181–188, 2012.
40. M. Zaroug, I. E. Orhan, F. S. Senol, and S. Yagi, "Comparative antioxidant activity appraisal of traditional Sudanese kisra prepared from two sorghum cultivars," *Food Chem.*, vol. 156, pp. 110–116, 2014.
41. L. Alvarez-Jubete, H. Wijngaard, E. K. Arendt, and E. Gallagher, "Polyphenol composition and in vitro antioxidant activity of amaranth, quinoa buckwheat and wheat as affected by sprouting and baking," *Food Chem.*, vol. 119, no. 2, pp. 770–778, 2010.
42. D. Novotni *et al.*, "Influence of frozen storage and packaging on oxidative stability and texture of bread produced by different processes," *LWT - Food Sci. Technol.*, vol. 44, no. 3, pp. 643–649, 2011.
43. J. Slavin, "Whole grains and human health," *Nutr. Res. Rev.*, vol. 17, pp. 99–110, 2004.
44. A. BAKKE AND Z. VICKERS N., "Consumer Liking of Refined and Whole Wheat Breads." *JOURNAL OF FOOD SCIENCE*, Vol. 72, pp. S473-S480, 2007.
45. W. B. Rolls, J. Delk, and Z. Vickers, "Determining a series of Whole Wheat Difference Thresholds for use in a Gradual Adjustment," *J. Sens. Stud.*, vol. 22, pp. 639–652, 2007.
46. A. S. Yadav and D. C. Pandey, "Geographical Perspectives of Mango Production in India," no. 4, pp. 257–265, 2016.
47. A. Jegede, "Top 10 Largest Mango Producing Countries In The World," *The Daily Records*, 2019.

48. S. K. Mitra, S. Mitra, B. Ghosh, and P. K. Pathak, "Mango cultivars and hybrids grown in West Bengal, INDIA," in *Acta Horticulturae*, 2013, no. 992, pp. 325–330.
49. C. M. Ajila, L. J. Rao, and U. J. S. P. Rao, "Characterization of bioactive compounds from raw and ripe *Mangifera indica* L. peel extracts," *Food Chem. Toxicol.*, vol. 48, no. 12, pp. 3406–3411, 2010.
50. C. M. Ajila, K. Leelavathi, and U. J. S. P. Rao, "Improvement of dietary fiber content and antioxidant properties in soft dough biscuits with the incorporation of mango peel powder," *Journal of Cereal Science*, vol. 48, pp. 319-326, 2008.
51. C. M. Ajila, M. Aalami, K. Leelavathi, and U. J. S. P. Rao, "Mango peel powder : A potential source of antioxidant and dietary fiber in macaroni preparations," *Innov. Food Sci. Emerg. Technol.*, vol. 11, no. 1, pp. 219–224, 2010.
52. K. Bandyopadhyay¹, C Chakraborty and S Bhattacharyya, "Fortification of Mango Peel and Kernel Powder in Cookies Formulation," *Journal of Academia and Industrial Research*, vol. 2, no. 12, pp. 661–664, 2014.
53. AOAC International, "AOAC: Official Methods of Analysis (Volume 1)," 9 CFR 318.19(b), 1999.
54. S. Kumar, S. Suhrita, and D. Runu, "Effect of microwave power on drying kinetics, structure, color, and antioxidant activities of corncob," *J Food Process Eng.* vol. 2022, no. July 2018, pp. 1–13, 2020.
55. R. M. Lamuela-ravents, "Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent," *METHODS IN ENZYMOLOGY*, vol. 299, no. 1974, pp. 152– 178, 1999.
56. M. E. Cuvelier and C. Berset, "Use of a Free Radical Method to Evaluate Antioxidant Activity," *Lebensm.-Wiss. u.-Technol.*, vol. 30, pp. 25–30, 1995.
57. S. P. Heenan, J. Dufour, N. Hamid, W. Harvey, and C. M. Delahunty, "The sensory quality of fresh bread : Descriptive attributes and consumer perceptions," *Food Res. Int.*, vol. 41, no. 10, pp. 989–997, 2008.
58. L. H. Ho, N. A. Abdul Aziz, and B. Azahari, "Physico-chemical characteristics and sensory evaluation of wheat bread partially substituted with banana (*Musa*

- acuminata X balbisiana cv. Awak) pseudo-stem flour,” *Food Chem.*, vol. 139, no. 1–4, pp. 532–539, 2013.
59. J. Tomi, N. Sad, N. Sad, N. Sad, and V. Crops, “Breadmaking potential and proteolytic activity of wheat varieties from two production years with different climate conditions,” *Food Chemistry*, no. 139, pp. 532–539, 2015.
 60. FSSAI, “MANUAL OF METHODS OF ANALYSIS OF FOODS-CEREAL AND CEREAL PRODUCTS,” NEW DELHI, 2015.
 61. L. Das, U. Raychaudhuri, and R. Chakraborty, “EFFECT OF BAKING CONDITIONS ON THE PHYSICAL PROPERTIES,” *International Journal of Food, Agriculture and Veterinary Sciences*, vol. 2, no. 2, pp. 106–114, 2012.
 62. C. M. Ajila and U. J. S. P. Rao, “Mango peel dietary fibre: Composition and associated bound phenolics,” *J. Funct. Foods*, vol. 5, no. 1, pp. 444–450, 2013.
 63. C. M. Ajila, S. G. Bhat, and U. J. S. P. Rao, “Valuable components of raw and ripe peels from two Indian mango varieties,” *Food Chemistry*, vol. 102, pp. 1006–1011, 2007.
 64. S. Siriamornpun, E. Tangkhawanit, and N. Kaewseejan, “Reducing retrogradation and lipid oxidation of normal and glutinous rice flours by adding mango peel powder,” *FOOD Chem.*, vol. 201, pp. 160–167, 2016.
 65. A. E. Quirós-Sauceda *et al.*, “Dietary fiber and phenolic compounds as functional ingredients: Interaction and possible effect after ingestion,” *Food Funct.*, vol. 5, no. 6, pp. 1063–1072, 2014.
 66. A. Bakke and Z. Vickers, “Effects of bitterness, roughness, PROP taster status, and fungi form papillae density on bread acceptance,” *Food Qual. Prefer.*, vol. 22, no. 4, pp. 317–325, 2011.
 67. D. Pathak, J. Mazumdar, U. Raychaudhuri and R. Chakraborty, “Dynamic role of natural antioxidant sources on different parameters of bread quality: a review,” *Nutrafoods*, Vol. 15, pp. 163–178, 2016.
 68. J. Ndife, J. Obiegbunna, and S. Ajayi, “Comparative evaluation of the nutritional and sensory quality of major commercial whole-wheat breads in Nigerian market,” *Adv. J. Food Sci. Technol.*, vol. 5, no. 12, pp. 1600–1605, 2013.

69. L. Yu, A. Nanguet, and T. Beta, "Comparison of Antioxidant Properties of Refined and Whole Wheat Flour and Bread," *Antioxidants* (Basel), Vol. 2(4), pp. 370–383, 2013.
70. Z. E. Martins, O. Pinho, and I. M. P. L. V. O. Ferreira, "Food industry by-products used as functional ingredients of bakery products," *Trends Food Sci. Technol.*, vol. 67, pp. 106–128, 2017.
71. M. Gómez and M. M. Martínez, "Fruit and vegetable by-products as novel ingredients to improve the nutritional quality of baked goods," *Critical Reviews in Food Science and Nutrition*, vol. 58 (13), pp. 2119-2135, 2018.
72. J. P. Trigo, E. M. C. Alexandre, J. A. Saraiva, and E. Manuela, "High value-added compounds from fruit and vegetable by-products – Characterization, bioactivities, and application in the development of novel food products," *Crit. Rev. Food Sci. Nutr.*, vol. 60, no. 8, pp. 1388-1416, 2019.
73. S. Bhol, D. Lanka, S. John, and D. Bosco, "Quality characteristics and antioxidant properties of breads incorporated with pomegranate whole fruit bagasse," *J. Food Sci. Technol.*, vol. 53, no. March, pp. 1717–1721, 2016.
74. R. Walker, A. Tseng, G. Cavender, A. Ross, and Y. Zhao, "Physicochemical, Nutritional, and Sensory Qualities of Wine Grape Pomace Fortified Baked Goods." *J Food Sci.*, Vol. 79, no. 9, pp. S1811-S1822, 2014.
75. Najafabadi, Leila & Hamdami, Nasser & Le-Bail, Alain & Monteau, Jean-Yves & Keramat, J., "Impact of Baking Bed and Baking Temperature on Staling of Sangak Bread," *Journal of Agricultural Science and Technology*, Vol. 17(2), pp. 375-386, 2015.
76. Serna, Liliana & García Gonzalez, Estefania & Torres-León, Cristian., "Agro-Industrial Potential of the Mango Peel Based on its Nutritional and Functional Properties.", *Food Reviews International.*, Vol. 32(4), pp. 364-376, 2015.
77. D. Pathak, J. Majumdar, U. Raychaudhuri and R. Chakraborty, "Characterization of physicochemical properties in whole wheat bread after incorporation of ripe mango peel," *J. Food Meas. Charact.*, vol. 10, no. 3, pp. 554–561, 2016.

78. F. Cornejo and C. M. Rosell, "Physicochemical properties of long rice grain varieties in relation to gluten free bread quality," *LWT - Food Sci. Technol.*, vol. 62, no. 2, pp. 1203–1210, 2015.
79. M. A. Kurek and A. Wierzbicka, "Optimization of beta-glucan and water content in fortified wheat bread using Response Surface Methodology according to staling kinetics," *LWT*, vol. 75, pp. 352–357, 2017.
80. T. A. Shittu, A. O. Raji, and L. O. Sanni, "Bread from composite cassava-wheat flour: I. Effect of baking time and temperature on some physical properties of bread loaf," *Food Res. Int.*, vol. 40, no. 2, pp. 280–290, 2007.
81. W. Han, S. Ma, L. Li, X. Zheng, and X. Wang, "Impact of wheat bran dietary fiber on gluten and gluten-starch microstructure formation in dough," *Food Hydrocoll.*, vol. 95, pp. 292–297, 2019.
82. J. Xu, W. Wang, and Y. Li, "Dough properties, bread quality, and associated interactions with added phenolic compounds: A review," *J. Funct. Foods*, vol. 52, no. November, pp. 629–639, 2019.
83. Peluola-Adeyemi, O.A & Obi, T.E & Ugbogu, D.I., "Effect of Temperature and Time on the Physical Properties of Bread Produced from Wheat – Cocoyam Flour Using Response Surface Methodology," *J. Food Technol. Res.*, vol. 3, no. 2, pp. 63–71, 2016.
84. R. C. Borrelli *et al.*, "Characterization of coloured compounds obtained by enzymatic extraction of bakery products," *Food Chem. Toxicol.*, vol. 41, no. 10, pp. 1367–1374, 2003.
85. P. Gélinas and C. M. McKinnon, "Effect of wheat variety, farming site, and bread-baking on total phenolics," *Int. J. Food Sci. Technol.*, vol. 41, no. 3, pp. 329–332, 2006.
86. F. Shahidi and J. D. Yeo, "Insoluble-bound phenolics in food," *Molecules*, vol. 21, no. 9, 2016.
87. K. Sharma *et al.*, "Temperature-dependent studies on the total phenolics, flavonoids, antioxidant activities, and sugar content in six onion varieties," *J. Food Drug Anal.*, vol. 23, no. 2, pp. 243–252, 2015.

88. M. Irakli, D. Katsantonis, and F. Kleisiaris, "Evaluation of quality attributes, nutraceutical components and antioxidant potential of wheat bread substituted with rice bran," *J. Cereal Sci.*, vol. 65, pp. 74–80, 2015.
89. H. Etgü and S. Kan, "EFFECT OF GRAPE (*VITIS VINIFERA L.*) POMACE ON THE QUALITY, TOTAL PHENOLIC CONTENT AND ANTI-RADICAL ACTIVITY OF BREAD," *Journal of Food Processing and Preservation*, vol. 38, pp. 980–986, 2014.
90. MEHMET HAYTA, GAMZE ÖZUG UR, HAKIYE ETGÜ and IBRAHIM TUGKAN SEKER, "EFFECT OF GRAPE (*VITIS VINIFERA L.*) POMACE ON THE QUALITY, TOTAL PHENOLIC CONTENT AND ANTI-RADICAL ACTIVITY OF BREAD", *Journal of Food Processing and Preservation*, Vol. 38, pp. 980–986, 2014.
91. E. Armero and C. Collar, "Crumb Firming Kinetics of Wheat Breads with Anti-staling Additives", *Journal of Cereal Science*, Vol. 28, pp. 165–174, 1998.
92. Sirithon Siriamornpun, Ekkarat Tangkhawanit, Niwat Kaewseejan, "Reducing retrogradation and lipid oxidation of normal and glutinous rice flours by adding mango peel powder", *Food Chemistry*, Vol. 201 pp. 160–167, 2016.
93. E. Gallagher, T. R. Gormley, and E. K. Arendt, "Recent advances in the formulation of gluten-free cereal-based products," *Trends Food Sci. Technol.*, vol. 15, no. 3–4, pp. 143–152, 2004.
94. K. Wang, F. Lu, Z. Li, L. Zhao, and C. Han, "Recent developments in gluten-free bread baking approaches: a review," *Food Science and Technology (Campinas)*, vol. 37, pp. 1–9, 2017.
95. C. M. Ajila, K. A. Naidu, S. G. Bhat, and U. J. S. P. Rao, "Bioactive compounds and antioxidant potential of mango peel extract," *Food Chem.*, vol. 105, no. 3, pp. 982–988, 2007.
96. B. Ng, "Fiber content and storage stability of antioxidant activity of nutraceutical mango products and gluten-free cookies," *2nd Int. Conf. Biotechnol. Food Sci.*, vol. 7, no. April 2011, pp. 36–39, 2011.

97. Kleopatra Tsatsaragkou, Styliani Protonotariou, Ioanna Mandala, "Structural role of fibre addition to increase knowledge of non-gluten bread", *Journal of Cereal Science*, Vol 67, pp. 58-67, 2016.
98. D. Pathak, J. Mazumdar, U. Raychaudhuri and R. Chakraborty, "Study on enrichment of whole wheat bread quality with the incorporation of tropical fruit by-product," *International Food Research Journal*, Vol. 24(1), pp. 238-246, 2017.
99. Calvin Onyango, Christopher Mutungi, Günter Unbehend, Meinolf G. Lindhauer, "Modification of gluten-free sorghum batter and bread using maize, potato, cassava or rice starch." *LWT - Food Science and Technology*, Vol. 44(3), pp. 681-686, 2011.
100. Tsai, S., Unklesbay , N., Unklesbay, K., and Clarke, "A. Water and absorptive properties of restructured beef products with five binders at four isothermal temperatures." *Lebensm. -Wiss. U.-Technol.*, Vol. 31, pp. 78-83, 1998.
101. Mbaeyi-Nwaoha, I.E., and Onweluzo, J.C.:Functional, "Properties of Sorghum (S. bicolor L.) - Pigeonpea (Cajanus cajan) Flour Blends and Storage Stability of a Flaked Breakfast Formulated from Blends. Pakistan." *Journal of Nutrition*, Vol. 12 (4), pp. 382-397, 2013.
102. Leach, H .W., Mc Cowen, L.D., and Schoch T.J., "Structure of the starch granule I. Swelling and solubility patterns of various starches." *Cereal Chem.*, pp. 36:534-543, 1959.
103. Mario, M.M., Alvaro, D., and Manuel, G. "Effect of different microstructural features of soluble and insoluble fibres on gluten-free dough rheology and bread-making." *J.Food Engg.* pp. 142:49-56, 2014.
104. Xu, B.J., and Chang, S.K.C. "A comparative study on phenolic profiles and antioxidant activities of legumes as affected by extraction solvents." *J. Food Sci.* Vol. 72, pp. S159-S166, 2007.
105. Ashoush, I.S., and Gadallah, M.G.E. "Utilization of mango peels and seed kernels powders as Sources of phytochemicals in biscuit." *World J. Dairy and Food Sciences.* Vol 6 (1), pp. 35-42, 2011.
106. Cauvain, S.P., and Young, L.S., *Bakery Food Manufacture and Quality: Water control and Effects.* Second edition. pp. 42-44. 2008.

107. Sankhon, A., Amadou, I., Yao, W.R., Wang, H., Qian, H., and Sangare, M. "Comparison of physicochemical and functional properties of flour and starch extract in different methods from Africa locust bean (*parkia biglobosa*) seeds." African journal of Traditional, complementary and alternative medicines. Vol. 11(2), pp. 264-272, 2014.
108. Singh, V., Okadome, H., Toyoshima, H., Isobe, S., and Ohtsubo, K. "Thermal and physicochemical properties of rice Grain, flour and starch." J. Agric. Food Chem. Vol. 48, pp. 2639-264, 2000.
109. Daou, C., and Zhang, H. "Study on Functional Properties of Physically Modified Dietary Fibres Derived from Defatted Rice Bran." J. Agri. Sci. Vol. 4, No. 9, 2012.
110. Menon , L., Dutta Majumdar , S., and Ravi , U. "Mango kernel flour as a potential ingredient in the composite flour of bread." Indian J. Natural Products and Resources. Vol. 5, pp. 75-82, 2014.
111. Marina, Z., and Noriham, "A. Quantification of total phenolic compound in vitro antioxidant potential of fruit peel extracts." International Food Research Journal. Vol. 21(5), pp. 1925-1929, 2014.

Dynamic role of natural antioxidant sources on different parameters of bread quality: a review

Debasmita Pathak, Jayshree Majumdar, Utpal Raychaudhuri, Runu Chakraborty*

Correspondence to:
Runu Chakraborty
crunu@hotmail.com

Keywords:

Bread, Natural antioxidants,
Sensory evaluation,
Physical properties,
Functional properties

Abstract

Improved understanding of the relationship between the consumption of antioxidant-rich food and human health has resulted in the supplementation of food products with natural antioxidants. Enrichment of a staple food such as bread with natural antioxidants can provide additional health benefits. Sources of natural antioxidants include cereals, seeds, spices, herbs, green plants, fruit, vegetables and the less expensive and easily available waste products from the food industry. However, there are technological constraints to the fortification of bread, while sensory and physical properties are important for overall consumer acceptability. Synthetic antioxidants can have serious adverse effects on health, and their substitution with natural antioxidants enhances the therapeutic and functional properties of bread and has the added advantage of increasing shelf-life. This review discusses the antioxidant activity and scavenging properties of various types of breads enriched with natural antioxidants from natural sources and the associated health benefits and sensory attributes of these breads.

Introduction

In biological systems, oxidative stress is produced by an imbalance between chemical stresses caused by abnormal quantities of reactive oxygen species (ROS) and physical ability to detoxify the reactive intermediates. Oxidative stress if prolonged can damage the entire system and has been implicated in over 100 common diseases such as cancer, diabetes, heart conditions and neurological disease [1]. However, antioxidants can scavenge ROS and free radicals, thus reducing their numbers. The growing interest in foods rich in antioxidants has led to the development of a large market for antioxidant-rich products and ingredients [2]. Antioxidants can also be used to prevent the degradation of food during processing and storage, thereby increasing its shelf-life.

The importance of natural antioxidants for food applications has been highlighted in numerous studies. Natural antioxidants are found in plants which can be consumed. For example, spices, herbs and essential oils are rich in antioxidant properties [3]. The consumption of natural antioxidants including phenolic and polyphenolic compounds, chelators, antioxidant vitamins and enzymes as well as carotenoids has received considerable interest because of their presumed safety and potential nutritional and therapeutic effects including anti-inflammatory, anti-carcinogenic and anti-athero-

*Department of Food Technology and Biochemical Engineering, Jadavpur University, Kolkata 700032, India
tel: ++91 983112 2626; fax: ++91 033 2414 6822

sclerotic properties. Artificial antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) are synthetically produced chemical antioxidants which are widely used for their antioxidant properties in the food industry. However, the use of these substances is restricted due to toxicity problems and fears of liver damage and carcinogenesis [4]. Consequently, there is ongoing research on natural sources of antioxidants. Bread is a good source of calories and other nutrients and has been a staple food for centuries. It is traditionally made from wheat flour, yeast, sugar, salt and water. Wheat grain contains many bioactive compounds, including antioxidants (phenolic compounds) particularly in the bran and aleurone layer, although the processing of wheat bread also confers some antioxidant activity [5]. Many different synthetic antioxidants have been used in bread for years to increase storage stability. The current rapidly growing functional food and nutraceutical market has stimulated research on the use of natural antioxidants in bread which do not affect its textural and organoleptic properties. Functional and therapeutic properties, basic nutritional quality and sensory and textural properties must be carefully considered when incorporating various sources of antioxidants into bread. Recent studies have shown that the addition of small

amounts of natural antioxidant sources does not significantly affect the textural and sensory properties of bread.

Dziki *et al* [5] discuss enhancement of the antioxidant activity of wheat bread by the addition of different plant materials along with changes in antioxidant activity during the bread-making process. Thus, the aim of this study was to review wheat and non-wheat bread and discuss the therapeutic and nutritional benefits achieved with the addition of sources of antioxidants as well as the impact of fortification on the sensory and textural properties of the bread.

Review of antioxidant rich breads

Many consumers prefer to eat healthier foods in order to prevent non-communicable diseases. Consequently, researchers are seeking to optimize bread-making technology in order to improve the quality, taste, functionality and bioavailability of this staple food. Accordingly, supplementation with cereals and their fractions, fruits and vegetables, gums and starches, and agricultural wastes has been used to produce antioxidant-rich bread. This research is discussed below and bread characteristics are summarised in Table 1.

Table 1 - Effect of natural antioxidant sources on different quality parameters of bread

Source of fortification	Antioxidant activity (assay) value (highest shown)							Effect on sensory evaluation	Effect on physical properties	Effect on functional properties
	TPC	DPPH	FRAP	Others						
Black tea [6]	24.9±0.2 (GAE mg/g sample)		18.8±1.2 Fe(II) (mM/g sample)	ABTS 58.4±0.2 (percentage of inhibition)		Total flavonoid content 640.1±6.0 (QE mg/g sample)		Overall good acceptance	Less effect on textural and physical properties with darker colour	Antimicrobial effect
Rice bran [7]	183.9±2.9 free phenolics 110.4±8.3 bound phenolics (mg GAE/100 g dw)	400 mg TE/100 g dw (total phenolics) (approx.)	400 mg TE/100 g dw (total phenolics) (approx.)	Total flavonoid content 76.5±0.6 (free) 34.4±3.4 (bound) (mg CATE/100 g dw)	Ferulic acid 4.2±0.6 (free) 285.1±8.3 (bound) (mg/g dw)	Sinapic acid 0.5±0.1 (free) 14.0±2.1 (bound) (mg/g dw)	<i>p</i> -Coumaric acid 2.1 ± 0.5 (free) 76.8 ± 4.0 (bound) (mg/g dw)	Lower volume with increased firmness and hardness	Low overall acceptability	
Green tea extract (GTE) and microencapsulated GTE [8]	216.48±1.01 (mg/loaf)							Lower overall scores due to astringency and colour	Insignificant effect with enhanced moisture content	
<i>Stevia rebaudiana</i> extracts (var. Morita) [9]		51.98 IC ₅₀ (mg/ml)						Lower sweetness and overall likeability	Insignificant effect up to 50% replacement	Antidiabetic and antimicrobial effect

Source of fortification	Antioxidant activity (assay value (highest shown))						Effect on sensory evaluation	Effect on physical properties	Effect on functional properties
	TPC	DPPH	FRAP	Others					
Quinoa leaves [10]	2.90±0.05		6.07±0.15	ABTS 5.14±0.14 (scavenging %)	LPO 6.04±0.20 (scavenging %)	CHP 2.52±0.20 (% inhibition)	No significant effect	Linear increase in crumb hardness, cohesiveness and gumminess	
Nejayote solids [11]				Ferulic acid 14.16 (mg/100 g)		ORAC 2.32 (µM Trolox/100 g)	Up to 9% supplementation without affecting sensory parameters	No alteration	Higher fibre and bioavailable calcium
Broccoli sprout powder [12]	2.0 (mg GAE/g dw)		56.98±1.94 (EC ₅₀ mg dw/ml)	ABTS 12.69±0.53 (EC ₅₀ mg dw/ml)	LPO 83.93±2.71 (EC ₅₀ mg dw/ml)	Rich in free amino acids	Satisfactory overall consumer acceptability up to 2% supplementation		Rich in free amino acids and chemopreventive potential
Onion skin powder [13]		59.29 ± 1.88 (mg dw/ml)		ABTS 164.52±5.65 (mg dw/ml)	LPO 14.67±0.87 (mg dw/ml)	Availability of quercetin	Satisfactory acceptability up to 3% supplementation	Insignificant difference	Availability of quercetin
Chestnut flour [14]		1.04 mmol (Trolox eq./g of dw)						Heterogeneous crumb structure	A richer volatile profile
Banana pseudo-stem flour [15]	204.16±3.04 (mg GAE/100 g of sample)	241.39 ± 23.06 (µmol TEAC/100 g of sample)	849.01 ± 32.46 (µmol Fe(II)/100 g of sample)				Higher acceptability with CMC	Increases volume with decreases in other physical characters	Rich in fibre
Fennel seed powder [16]	1.7 (mg GAE/100 g)	97±2% (DPPH % inhibition)	2±0.1 (mg FeSO ₄ /100 g)				Good consumer acceptability up to 7% supplementation	Crumb moisture and firmness increased	Abundant minerals and dietary fibre
Cinnamon powder [17]	0.94 (mg GAE/100 g dw)	27.67% (DPPH % inhibition)					Maximum overall acceptability at 2% supplementation	Increase in crumb moisture. 2% Cinnamon powder improved baking and textural properties	
Citrus peel [18]	132.3±3.9 (mg GAE/g)	192.3 ± 15.2 vitamin E equiv. (mg/g)		H ₂ O ₂ reduction method 75.9±1.3 Vitamin C equiv. (mg/g)					
Intermediated pearled wheat fraction [19]	3066 (mg/kg)	0.73 (mmol TE/kg)					Rheological and technological properties were acceptable	Acceptable rheological and technological properties	Rich in protein, dietary fibre, ash content and β-glucan
Maca flour [20]	0.39±0.07 (mg GAE/100 g dm)	21±1.7 Inhibition (%)		ABTS 40±1.7 (inhibition %)	Hydroxyl radical (•OH) 51±1.9 (inhibition %)	Peroxynitrite radicals 25±1.6 (inhibition %)	ORAC 1.621 ± 1.5 (inhibition %)		Anti-inflammatory effect and reducing glucose absorption
Micronized by-products from debraned durum wheat [21]	0.95±0.03 (mM GAE/g)	72.8±0.4 (DPPH % inhibition)					Enhanced sensory properties of breads containing bran	Weak structure, poor baking quality, decreased bread volume and elasticity, increased hardness	Free amino acids and insoluble fibre
Sorghum flour [22]	2.02±0.09 (mg GAE/g sample)	9.45±0.45 (µmol TE/g)					No reduction in sensory acceptability	Observed differences in dough farinograph properties	Remarkable amount of total dietary fibre

Source of fortification	Antioxidant activity (assay) value (highest shown)				Effect on sensory evaluation	Effect on physical properties	Effect on functional properties
	TPC	DPPH	FRAP	Others			
Barley hull and flaxseed hull [23]	4.05±0.31 (mg ferulic acid equivalent/g bread sample)	12.87 ± 2.02 (mg Trolox equivalent / g sample)		ORAC 36.44±14.01 (mg Trolox equivalent/g sample) (barley and flaxseed hulls)			
Barley flour and barley protein isolate [24]	369.60 (mg/100 g)	63.30% (DPPH % inhibition)		ACE inhibitory activity 75.30%	α-Amylase inhibitory activity 77.80		Increased the content of essential amino acids
Coriander leaf powder [25]	33 (mg GAE/100 g of sample)	28 (DPPH % inhibition)	37.5 (mg FeSO ₄ /100 g)			Highest overall acceptability at high percentage of supplementation	Affected physical parameters, increase in crumb firmness and loaf weight
Ginger powder [26]	0.710±0.009 (mg GAE/g dw)	0.242 ± 0.009 (mmol DPPH / mg dw/ml)				Negative effect on bread acceptability	Rheology of dough and texture of bread affected
Turmeric powder [27]	160 (mg GAE/100 g)			β-Carotene-linoleate bleaching assay 79.8%±0.6% antioxidant activity		Satisfactory overall consumer acceptability up to 4% supplementation	Hardness and crumb colour a* and b* values increased
Wholegrain flours and fibres [28]	580 (µg/g) (bound phenolics) 180 µg/g (free phenolics)	5% (bound phenolics) 38% (free phenolics) (% inhibition)					High total dietary fibre and protein content
Anka (fermented rice) [29]	3.86±0.05 (mg extract/ml)	7.78 ± 0.03 (mg extract/ml) (at 3% supplementation)				No significant differences with effect on colour and mouth feel	Decrease in specific volume
Grape by-product [30]	613.77±1.57 (mg GAE/100 g of sample)	15.02 (mmol TRE/g dm)	48 (mmol Fe(II)/g dm)			Acceptable products	Affected hardness, gumminess and sharpness
Pseudocereal flour [31]	7.25±0.23 (mg/g dw)					Improves acceptability	Higher presence of dietary fibre and β-glucans
Grape seed extract [32]	TEAC 4.8±0.1 (nmol Trolox/mg bread sample)			55% Inhibition of the formation of CML in bread crust		Little effect on the quality attributes	No significant difference in hardness
Sugar beet molasses-based ingredients [33]		1677 (µmol TE/100 g)				No interference at lower levels of supplementation	Significant effect on specific volume
Barley flour [34]	280 (mg GAE/100 g dm)		2.5 (mmol Fe/100 g dm)			Good consistency between sensory evaluation and the amount of phenolics	
Garlic and basil [35]	0.278 (mM GAE/g)		0.368 (mM Fe ²⁺ /100 g)				
<i>Auricularia auricula</i> flour [36]		57% (DPPH % inhibition)				No significant effect except at very high supplementation levels	Little effect on loaf weight, loaf height and loaf volume up to 9% supplementation
Yam flour [37]		79% (DPPH % inhibition)		ABTS 88% (total antioxidant property %)		No interference with bread acceptability	No significant effect at low levels of supplementation

ABTS 2,20-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) radical scavenging activity, *CHP* metal chelating activity, *CMC* carboxy methyl cellulose, *CML* ne-(carboxymethyl)lysine, *dm* dry matter, *DPPH* (di(phenyl)-(2,4,6-trinitrophenyl) iminoazanium radical scavenging activity, *dw* dry weight, *FRAP* ferric reducing antioxidant power, *LPO* inhibition of lipid peroxidation, *ORAC* oxygen radical absorbance capacity, *TEAC* trolox equivalent antioxidant capacity, *TPC* total phenolic content

Black tea

Chinese steamed bread (CSB) was fortified with black tea with various tea to water (v/v) ratios [6]. Increasing the black tea concentration decreased the lightness (L), while increasing the redness (a) and yellowness (b) of CSB, which was attributed to the presence of black tea polyphenols. The addition of black tea greatly increased phenolic content and antioxidant activity along with the antimicrobial properties of CSB, with the best results seen at a 5:1 ratio of tea to water. There were fewer changes in the water activity, water content, texture and volumetric properties of fortified CSB when a lower ratio of tea was employed. The addition of black tea had little effect on sensory scores for brightness, crumb structure, elasticity and stickiness but resulted in decreased whiteness and smoothness.

Rice bran

Stabilised rice bran (RB) was added at different percentages to wheat breads [7]. All substituted breads showed higher protein, ash, fat and total dietary fibre contents than control bread. Inclusion of RB negatively affected crumb colour (L^*), bread volume and sensory parameters such as colour, texture and overall acceptability, but improved hardness, aroma, taste, a^* and b^* . The highest and lowest values were seen in 30% RB substituted bread. Vitamin E content as indicated by tocopherol and tocotrienol content increased in proportion to the amount of RB. The addition of RB also increased the free and bound total phenolic content (TPC), total flavonoid content (TFC), ferulic acid, *p*-coumaric acid, sinapic acid, and ferric reducing antioxidant power (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging values compared to control bread, with the highest values seen in 30% RB substituted bread.

Green tea extract and microencapsulated green tea extract

Spray dried (SDMD) and freeze dried (FDMD) green tea polyphenols microencapsulated using maltodextrin were incorporated into bread to enhance its antioxidant properties [8]. Fortified bread had a significantly higher moisture content, a^* , b^* and ΔE as well as lower L^* values than

control bread. The reduced volume and increased crumb firmness of incorporated breads were statistically significant. These breads had lower overall sensory, taste, texture, crumb and crust colour scores than the control. Very similar amounts of polyphenols were present in the green tea extract (GTE) and in the encapsulates incorporated in the dough. However, after baking, the amount of polyphenols in incorporated breads was slightly reduced due to the degradation of polyphenols, but was still higher than that in control bread. The highest polyphenol content was seen in FDMD bread after baking and in non-encapsulated bread before baking. It was concluded that fortification of bread with green tea polyphenols can retain the functionality and quality characteristics of bread.

Stevia rebaudiana extracts (var. Morita)

Functional bread was prepared by partial replacement of sucrose with different amounts of *Stevia rebaudiana* extracts (var. Morita) [9]. Incorporated breads were rich in protein and crude fat. The substitution of up to 50% of sugar with stevia extracts did not adversely affect the softness of bread. The breads showed significantly higher antimicrobial activity with the highest value seen at 75% sugar substitution due to reduced sugar content. Multiple comparison tests revealed that sweetness decreased as the level of stevia extract increased. Bread with 50% replacement of sugar was well accepted. These breads had higher total dietary, insoluble and soluble fibre content than control bread, with a significant reduction in the caloric value. Bread containing stevia extract had good scavenging properties and efficiency in reducing polysaccharide digestion, which resulted in lower glucose absorption.

Quinoa leaves

Bread was fortified with different amounts of quinoa leaves (QL) as a source of antioxidants [10]. An increase in the percentage of QL resulted in a bread with decreased loaf volume and starch digestibility, and higher bread crumb hardness, cohesiveness and chewiness/gumminess, with an insignificant effect on elasticity. Bread fortification with QL showed a linear increase in antioxidant properties and TPC and had a positive influence

on FRAP, inhibition of linoleic acid peroxidation and chelating power. The maximum antiradical potential of bread was obtained with 5% QL supplementation.

Nejayote solids

Nejayote, a maize-processing wastewater, in the form of solids was used to enhance antioxidant activity in bread [11]. The addition of different amounts of nejayote solids (NS) to dough significantly enhanced water absorption, bread weight and apparent density with a slight decrease in bread volume, while the optimum mixing time, proof heights and bread heights were not significantly affected. Bread supplemented with NS showed higher dietary fibre, crude fibre and bioavailable calcium: two slices (64 g) of bread supplemented with 9% NS provided 29% of the recommended daily calcium intake. Ferulic acid increased proportionally with increased NS levels and significantly improved antioxidant activity. Flavour and odour received low sensory scores, while overall acceptability, texture and colour were not affected by 9% NS substitution as compared to control.

Broccoli sprout powder

Bread was enriched with various proportions of broccoli sprout (BS) powder for its antioxidant and anti-carcinogenic activity [12]. Partial replacement of flour with BS powder had a strong negative effect on aroma, texture, taste and overall acceptability and a slightly negative effect on colour. However, BS significantly enhanced the phenolic content and lipoxygenase enzyme (LOX) inhibition activity of the breads, with the highest values seen with 2% substitution, although a linear relationship between increased phenolic content and the percentage of added BS was not seen. Fortification resulted in an increase in the level of free amino groups but negatively influenced the antiradical potential, with highest values seen with 1% BS. Reducing power was decreased compared to control, whereas it substantially increased at higher levels compared to lower level of incorporation. The highest chelating activity was seen in bread with 3% BS, but was nevertheless still lower than in the control sample. Inhibition of lipid peroxidation was significantly lower in supplemented bread

compared to control. Bread with 2% BS supplementation was determined to be best regarding consumer acceptability, antioxidant activity and ability to inhibit LOX and xanthan oxidase (pro-oxidative enzymes).

Onion skin powder

Bread supplemented with dry onion skin (OS) powder used as a source of antioxidants [13] showed an increase in quercetin concentration, scavenging activity, chelating power, lipid peroxidation and reducing power compared to control bread. As expected, the antioxidant properties (except for lipid peroxidation) of OS-fortified bread and overall acceptability were positively correlated with the addition of up to 2–3% OS. However, bread supplemented with 4–5% OS was less acceptable due to no further increase in antioxidant activity and the presence of excessive amounts of volatile compounds.

Chestnut flour

Different amounts of chestnut flour (CF) were incorporated into bread as a source of antioxidants and fatty acids [14]. Bread with up to 50% CF showed a significant linear increase in antioxidant activity and level of volatiles, including alcohols, followed by aldehydes, ketones and furans. The levels of compounds present in CF, such as furans including furfural, 5-methylfurfural and 2-furfuryl alcohol, increased rose strongly as amount of substitution increased. CF imparted nutty and smoky flavours due to the presence of guaiacol and 4-ethyl-guaiacol, demonstrating a positive effect on organoleptic properties. Bread made with 50% CF showed a homogeneous crumb structure, greater crumb hardness, more cohesiveness and a darker colour compared to control bread.

Banana pseudo-stem flour

A fixed amount of banana pseudo-stem flour (BPF) was used with xanthan gum (XG) or carboxy methyl cellulose (CMC) as a source of antioxidants in bread [15]. Proximate analyses showed that BPF breads had a high content of ash, crude fibre and soluble, insoluble and total dietary fibre, with a balanced insoluble dietary fibre/soluble dietary fibre ratio. The addition of BPF resulted

in an increase in phenolic content and free radical scavenging activity compared to control. However, supplementation with XG or CMC in breads containing BPF resulted in a significant decrease in phenolic content and free radical scavenging activity, with 10% substitution having the strongest effect whereas physical analyses indicated that supplementation with 10% BPF with XG reduced loaf volume, weight loss, height and higher density. While all breads containing BPF had a darker crumb and lighter crust colour, mouth feel, aroma, gumminess and taste perceptions were not affected. All breads received better acceptability scores than control bread, with the highest overall acceptability score seen for bread with added CMC.

Fennel seed powder

Various amounts of fennel seed (FS) powder were incorporated into bread as a source of antioxidants [16]. Supplementation significantly affected physical properties, with a negative effect on texture and decreased loaf volume and specific loaf volume, but with an insignificant effect on loaf mass. With increasing addition of FS, the redness (a^*) and yellowness (b^*) of the bread significantly increased, while lightness (L^*) significantly decreased. The appearance of the bread declined slightly, with the optimum taste and aroma scores obtained at 7–10% FS supplementation, while the best colour was achieved with 5% FS supplementation. A linear increase in TPC and FRAP values (but not DPPH radical scavenging activity which showed a reduction at 5% FS incorporation) was seen with increasing supplementation up to 7.0% FS, with further additions having little effect on antioxidant properties. Thus, supplementation with 5.0% and 7.0% FS powder was found to be optimum.

Cinnamon powder

Different amounts of cinnamon powder (CP), which is rich in fibre, fats and ash, and has a high phenol content and antioxidant activity, were added to bread [17]. Baking absorption and loaf weight increased linearly with amount of CP added. Loaf volume and height increased up to 2% CP supplementation and decreased thereafter, while specific volume remained either equal to or less than that of control bread. Appearance

and colour scores were high at 2% and 3% CP addition, with flavour, texture and overall acceptability scores highest at 2% CP incorporation. The hardness, gumminess, cohesiveness and chewiness of the bread significantly increased above 2% CP incorporation. In addition, an adequate microbial inhibitory effect was observed due to the presence of cinnamaldehyde and eugenol at 2% CP incorporation. Fibre, ash, protein and fat content, phenolic content and antioxidant activity showed a significant linear increase as the level of supplementation increased with highest values obtained at 4% CP incorporation.

Citrus peel

Unfermented (UF) and fermented (F) citrus peel (CP) was processed under different dry hot-air temperatures to make four CP powders which were incorporated into toasting bread to enrich its antioxidant properties [18]. The water adsorption capacity of the dough increased with greater content of CP powder, resulting in faster toasting time and improved strength and elasticity. The crude fat and crude protein content of the unfermented citrus powder (UFCP) was higher than that of the fermented citrus powder (FCP), with crude fibre inversely proportional to moisture in CP. The UFCP extract showed better antioxidant activity (DPPH and H_2O_2 test) and higher total polyphenols, with the best values seen in UFCP dried at 150°C, indicating enhancement of phenolic compounds and antioxidant activity at higher drying temperatures. Sensory evaluation indicated greater acceptability of UFCP-supplemented bread compared to FCP-supplemented bread. Bread containing UFCP dried at 150°C achieved the highest score for flavour, while bread containing UFCP dried at 100°C had the best scores for hardness and surface texture. Both these breads together with bread containing FCP dried at 100°C achieved high overall acceptability scores, with bread containing FCP dried at 150°C scoring the lowest.

Intermediated pearled wheat fraction

Bread was enriched with different amounts of pearled fractions from intermediate layers of wheat kernel as a source of antioxidants [19]. The L^* , a^* and hue values (h) of the crust of supplemented

bread significantly increased with no difference in chroma (C^*) values compared to control. Crust crunchiness, total bread energy and acoustic emissions were highest in the bread containing 0% and 5% pearled fractions with lowest values seen at 20–25% incorporation. A decrease in bread volume with a proportional increase in gumminess, chewiness and hardness accompanied the increasing percentage of pearled wheat fractions, while cohesiveness and resilience remained unaffected. The protein, dietary fibre, β -glucan and ash content along with the total antioxidant activity (measured as DPPH scavenging capacity) and TPC of bread increased linearly and significantly as supplementation increased, with maximum values seen at 25% substitution. However, the alkylresorcinol content was lower than expected, while the deoxynivalenol content increased linearly, indicating 10% substitution was the best level of supplementation.

Maca flour

Maca flour (MF) as a powerful antioxidant was incorporated in different amounts into bread to increase its functional properties [20]. Bread containing MF had considerably increased antioxidant activity, with highest values seen at 20% MF incorporation. Antioxidant properties were determined according to TPC, scavenging effect on DPPH, ABTS and hydroxyl radicals as well as percentage of fluorescence decay inhibition and anti-inflammatory activity. The study also demonstrated that MF could reduce sugar intake as shown by an enzymatic assay where α -amylase and α -glucosidase were progressively inhibited as MF content increased, with maximum values found at 20% MF addition. Results showed that the biological properties of maca were retained after the bread-making process.

Micronized by-products from debranned durum wheat

Different amounts of coarse and fine micronized bran fractions were incorporated both into sourdough and baker's yeast dough to make antioxidant-rich bread [21]. The intermediate and internal layers of durum wheat contain high amounts of protein, fat, starch and free amino acids and have a

high peroxide value, while the external fraction has a high total dietary fibre content. Supplementation with up to 5% coarse fraction or bran fraction only changed water absorption capacity, development time and stability very slightly compared to control. However, the concentration of total free amino acids gradually increased with the addition of bran fraction and fermentation of dough. Supplementation and dough fermentation increased total phenol and scavenging activity, with wheat flour bread supplemented with 5% fine fraction having the highest values. The hardness, resilience and fracturability of breads baked at 220°C and enriched with bran fractions were lower than those of control. Bran fractions intensified crust colour, especially with sourdough fermentation, with increased sweetness, dryness and overall perception of taste, while elasticity decreased. Sourdough bread showed significantly higher acid flavour, acid taste and salty attributes for better overall consumer acceptability.

Sorghum flour

Varying amounts of wholegrain white sorghum flour (WSF) and red sorghum flour (RSF), which both provide total dietary fibre, were incorporated into flat bread as a source of antioxidants [22]. The total phenolic content and antioxidant capacity of red sorghum flat bread was significantly higher than that of white sorghum flat bread, which in turn was significantly higher than that of control bread. RSF bread had a higher content of bound phenolics and free phenolics than WSF bread or control bread, resulting in increased TPC. RSF flat breads were darker in colour than controls, possibly due to the presence of higher levels of coloured polyphenolic compounds such as anthocyanins. Breads supplemented with 30% WSF and 40% RSF had the highest scores for sensory parameters like flavour and texture and the best overall acceptability scores.

Barley hull and flaxseed hull

Barley hull extracts, flaxseed hull extracts and both combined were used as sources of antioxidants in Chinese steamed bread (CSB) [23]. The phytochemical profile of the bread improved significantly with the addition of barley hull extracts, flax-

seed hull extracts and both combined compared to control bread. Barley hull extract increased the content of ferulic acid and *p*-coumaric acid, and flaxseed hull extract introduced secoisolariciresinol diglucoside (SDG), ferulic acid glucoside (FeAG) and coumaric acid glucoside (CouAG) into CSB, while the combined extracts contributed their major phenolic compounds. The combined extracts significantly increased TPC, DPPH radical scavenging and oxygen radical absorbance capacity (ORAC) values compared to control.

Barley flour and barley protein isolate

Varying amounts of barley flour (BF) and barley protein isolate (BPI) were added as sources of antioxidants to pita bread [24]. The water holding capacity, solubility, emulsion and foaming properties of wheat flour fortified with BPI were superior compared to flour fortified with BF. Similarly, the content of essential and non-essential amino acids increased gradually as the amount of added BF and BPI increased rose from 0 to 15%. Bread made from 15% BPI had the highest levels of cystine, tyrosine, valine and essential amino acids. Protein, fat, ash, dry matter and fibre content increased with increasing supplementation, as did TPC, antioxidant activity, angiotensin converting enzyme (ACE) inhibitor activity and amylase inhibitory activity, with highest values seen at 15% BPI supplementation.

Coriander leaf powder

Varying amounts of coriander leaf powder (CLP) were used for antioxidant enrichment of bread [25]. Incorporation of dried CLP enhanced moisture retention capacity and baking characteristics, showing a higher crumb moisture up to 5% CLP supplementation and a slower staling rate. Physical parameters like crumb firmness and loaf weight increased, while specific volume and loaf volume values decreased linearly with increased supplementation. Similarly, as supplementation increased, the L^* , a^* and b^* values of the crust decreased, while crumb L^* values decreased and a^* values increased, indicating a darker colour. TPC, DPPH radical scavenging activity and FRAP assay results showed a sharp and almost linear increase with level of supplementation. The aroma accept-

ance scores gradually increased, while colour and taste were less acceptable with high CLP supplementation. Consequently, the overall acceptability of 3% and 5% CLP-substituted bread samples was much better than those with higher levels of supplementation.

Ginger powder

Different amounts of ginger powder (GP) were used to obtain antioxidant-rich bread [26]. The addition of GP enhanced the elasticity of dough. Hardness and gumminess were significantly higher with 6% GP supplementation, although changes in dough characteristics and bread rheological properties were insignificant up to 4.5% GP supplementation. Breads had a fine and uniform crumb. A higher percentage of ginger lowered the L^* and h values and elevated the C^* value of crumb. The amount of phenolics and radical scavenging activity increased in proportion with increased GP content of both crumb and crust, but to a great extent in crust. Addition of the lowest percentage of GP (3%) resulted in bread with the highest overall acceptability as it had the least amount of shogaol.

Turmeric powder

Varying amounts of turmeric powder (TP) were used as a source of antioxidants in wheat bread [27]. Despite a loss of 32–54% of TPC after baking, the breads had a significantly higher phenolic content due to the presence of curcumin. Scavenging activity, thermal stability and antioxidant properties as measured by β -carotene-linoleate bleaching assays improved as supplementation was increased to 2–8% TP compared with control breads. The taste and overall acceptability of breads with turmeric powder was best at 0–4% TP supplementation, while aroma and texture did not vary. The best crumb colour was seen at 2% TP supplementation.

Wholegrain flour and fibres

Wheat, rye, barley and oat wholegrain flours and cellulose (insoluble fibre) and xanthan gum (soluble fibre) in different proportions were added to wheat bread [28]. Bread containing oats had the highest levels of free phenolics, while rye bread

had the highest level of bound phenolics and the best scavenging capacity. The antioxidant properties of breads containing cellulose and xanthan gum were very similar to those of control bread, indicating that inclusion of fibre does not affect the antioxidant properties of high fibre bread. Incorporation of wholegrain and fibre increased total dietary fibre, while the addition whole wheat flour increased protein content. In general, there was a marked reduction in average specific and loaf volume. The overall quality of wholegrain-rich breads was acceptable except for that with added cellulose.

Anka (fermented rice)

Monascus-fermented rice (anka, red koji) flour was incorporated into bread to enhance its antioxidant properties [29]. Bread enriched with anka flour had a slightly lower specific volume and fewer carbohydrates but higher amounts of reducing sugars, fat and fibre than rice and wheat breads. Anka-enriched bread had an attractive red colour with lower L^* and whiteness index (WI) values and higher a^* and b^* values than rice and wheat breads. White bread and anka-enriched bread showed comparable antioxidant activity but were slightly less effective than rice-enriched bread regarding reducing power and scavenging ability as measured by the elimination of DPPH radicals and EC_{50} values. Substantial amounts of the functional components monacolin K and GABA were found in anka-enriched bread, which slightly decreased during baking. The colour and mouth feel of anka-enriched bread were rated higher, but there were no differences in its appearance, flavour and overall sensory attributes compared to white bread.

Grape by-product

Grape by-products (GP) containing very large amounts of ash and total dietary fibre were incorporated in different amounts to enhance the antioxidant properties of rye bread [30]. The levels of some phenolic compounds together with free radical scavenging activity (DPPH) and total antioxidant activity (FRAP) values increased gradually and significantly with increasing amounts of GP. The improved antioxidant properties of GP-enriched breads were attributable to the incorpora-

tion of phenolic compounds, mainly catechins and procyanidins. Considerable amounts of quercetin and quercetin-3- β -D-glucoside were also found. The hardness and gumminess of breads increased significantly with increasing amounts of GP, while insignificant changes were seen in cohesiveness, resilience, chewiness and springiness up to 8% GP supplementation. Sensory evaluation showed that as GP level increased, bread volume, porosity and overall acceptance decreased. Differences in aroma attributes were due to dominant fruity, alcoholic and sharp notes. However, different types of volatile esters and carbonyl compounds increased gradually.

Pseudocereal flour

Varying amounts of different pseudocereal (buckwheat, amaranth and quinoa) flours were used as a source of antioxidants for bread [31]. The incorporation of pseudocereal flour improved the antioxidant and scavenging properties of bread compared to control wheat bread. The addition of buckwheat flour improved antioxidant activity more effectively than amaranth or quinoa flour. Bread supplemented with 30% buckwheat flour had the highest phenolic content, TFC and Trolox equivalent antioxidant capacity (TEAC) values as evaluated by FRAP and DPPH, while bread supplemented with 15% buckwheat flour showed the highest flavonoid content. Organoleptic evaluation of pseudocereal-substituted breads indicated they were moderately acceptable. Buckwheat bread had a better sensory profile than amaranth and quinoa breads with higher scores for colour and odour.

Grape seed extract

Grape seed extract (GSE) added to bread linearly enhanced antioxidant activity with increased GSE [32]. However, baking degraded GSE proanthocyanidins, thus lowering GSE antioxidant capacity. No significant differences in hardness were observed between bread samples, but with increased addition of GSE, L^* and the E index decreased while a^* and b^* values increased, indicating that GSE affected bread colour. Addition of GSE did not affect quality attributes such as sweetness, porosity, astringency and stickiness but did im-

prove bread colour compared to the control. The catechins and proanthocyanidins in GSE greatly enhanced the antioxidant activity of bread. The abundant phenolics in GSE may help reduce the health risks associated with Ne-(carboxymethyl) lysine (CML).

Sugar beet molasses-based ingredients

Fruit and vegetable powders obtained with osmotic dehydration (OD) in beet molasses and pure beet molasses were incorporated into white bread to increase its antioxidant properties [33]. The OD vegetable powders contained significantly more protein, crude fibre, reducing sugars, saccharose, K, Na, Mg and Ca than the OD fruit powders. Bread supplemented with 10% OD red cabbage powder had the highest total moisture, ash, protein, crude fibre and mineral content, while bread supplemented with 10% OD plum powder had the highest crumb moisture, and bread supplemented with 10% OD apple powder had the highest reducing sugar content. The addition of molasses and molasses-based ingredients significantly increased the antioxidant activity of bread, with the highest values observed in bread formulated with 10% OD plum powder. Specific volume greatly decreased and colour darkened with increased supplementation. The crumb and crust of bread supplemented with 10% OD powders and molasses showed a decrease in yellowness (b^*) compared to bread made with 5% OD powders. Increased supplementation resulted in a gradual but significant increase in crumb pore roughness and a decrease in crumb elasticity. The taste attributes of bread supplemented with 5% OD were acceptable.

Barley flour

Varying amounts of barley flour (BF) from different varieties (Tyra, Cindy and STS 2-11) were added to wheat flour to make breads rich in antioxidants [34]. There were significantly more free phenolics consisting of flavonols and tocopherol than bound phenolics, including phenolic acids, in supplemented bread. The bread supplemented with the Tyra variety had significantly higher levels of free phenolics and total antioxidant activity than control bread. The total amount of phe-

nolics and total antioxidant activity were highest in breads supplemented with the Tyra and Cindy varieties. During baking, free phenolics decreased, while bound phenolics greatly increased and total antioxidant activity only changed slightly, with highest values seen in the Cindy and Tyra supplemented breads. The antioxidant properties of breads depended on the barley variety and the flour extraction rate. Other factors such as storage and baking procedure were less significant but did affect sensory attributes. Tyra-supplemented bread was the least acceptable with the highest scores for bitterness, off-odour and off-flavour, followed by the Cindy variety.

Garlic and basil

Garlic and sweet basil were added in different amounts to bread [35]. Total antioxidant activity (TAC) and polyphenol content were higher in the garlic and basil-flavoured breads than in white bread, and increased with increased levels of supplementation. Basil-flavoured bread showed higher TAC and polyphenol content than garlic-flavoured bread, with the maximum antioxidant activity seen in bread supplemented with 1.5% basil. Bread made with garlic and basil could be a source of antioxidants in the diet.

***Auricularia auricula* polysaccharide flour**

Various amounts of *Auricularia auricula* (commonly called black woody ear or tree ear fungus) polysaccharide flour (AAPF) were added to wheat bread [36]. AAPF has a high content of crude fibre, carbohydrates and minerals such as Fe, Ca, P and Mg compared to wheat flour. AAPF-supplemented bread showed a marked increase in concentration-dependent scavenging activity, which increased as supplementation increased. Up to 9% AAPF could be added without affecting the sensory qualities of the bread. Loaf weight, loaf volume and loaf height were slightly affected by 9% supplementation, but significantly negatively affected by 12% supplementation. Sensory characteristics like aroma, texture, taste and mouth feel were not affected at supplementation levels of 3–12%, except for colour which was affected by 12% supplementation. Bread made with 9% AAPF was considered the most suitable.

Yam flour

Different amounts of yam flour (YF) were added to wheat bread. YF contains a lot of ash, crude fibre and nitrogen free extract (NFE), the latter mainly as starch [37]. Loaf weight, loaf volume and loaf height showed no significant changes at 5% supplementation, while maximum changes were seen at 25% substitution. Free radical scavenging activity and total antioxidant activity showed a marked increase as the proportion of YF in the breads increased. The sensory score for colour increased as substitution increased. No statistically significant difference was observed in other sensory characteristics for bread made with up to 20% YF compared to wheat bread.

Factors affecting the antioxidant properties of bread

This literature review shows that supplementation of wheat flour with sources of natural antioxidant can produce a bread suitable for the functional food market. The quantity and quality of the final product are determined by different factors, as shown in Table 1. Enhancement of antioxidant properties mainly depends on the phenolic content of the natural sources and the amount of supplementation. Total phenolic and flavonoid content shows a positive correlation with antioxidant activity as seen in the case of *Lens culinaris* Medikus seeds [38], and so is used to determine product quality. A similar trend was observed when fennel seed powder, black tea, chestnut flour, banana pseudo-stem flour and yam flour were added to bread. However, antioxidant activity did not increase with higher levels of supplementation when broccoli sprout powder was used.

Free radical scavenging properties are generally evaluated using the DPPH (lipophilic stable free radicals), ABTS (hydrophilic radicals) and hydroxyl radical methods, as well as other experimental methods such as inhibition of lipid peroxidation and metal chelating activity evaluation. Increased FRAP, inhibition of lipid peroxidation and improvement of antiradical potential were shown when flour was supplemented with 4% and 5% quinoa leaves. Chelating power was also improved but only with 2% supplementa-

tion. This type of non-linear relationship may be partially explained by interactions between food components (especially phenolics, protein and starch) and gastrointestinal fluid [10]. A positive correlation was observed between increased levels of broccoli sprout powder and onion skin powder and protein–phenolic interactions. In a continuation of the chemical reaction, free amino acids were reduced and resistant starches increased, with a negative effect on protein and starch digestibility, highlighting the importance of adding the correct amount of functional ingredients. This finding may be due to the fact that flour samples with a low phenolic content are used at up to 20–40% supplementation levels, while spices, herbs and other extracts such as green tea and grape seed with a high phenolic content, are added at much lower supplementation levels of 2–10%.

Further *in vivo* studies on the production and marketing of naturally fortified antioxidant-rich breads are required. In previous research, bread enriched with a 0.5% nutraceutical compound (capsaicin extracted from red peppers, magnesium from barley germ and minerals) and eaten with salad when included in the diet for 3 months showed a significant increase in hydrosoluble antioxidants and decrease in oxidative stress in subjects compared to control [39]. This research also demonstrated the benefits of multicomponent functional food enriched with antioxidant compounds obtained from plant extracts.

Individual antioxidant compounds have been analysed in previous studies. Like vitamin E, ferulic acid, *p*-coumaric acid, sinapic acid from rice bran, curcumin from turmeric, and quercetin from onion skin improve the functional properties of bread. Antioxidant profiles can be studied using a method combining spectrophotometry and HPLC [40]. Antioxidant activity is determined as the total effect of free and bound phenolic compounds. In wholegrain products like bread, baking resulted in an increase in free phenolic acids but a decrease in bound phenolic acids with ferulic acid as the principal phenolic [41]. In bread supplemented with rice bran, both free and bound phenolics increased, but barley free phenolics decreased due to the interaction

of phenolic compounds with proanthocyanidins and minerals such as iron. In contrast, fermentation and baking showed a positive effect by liberating bound ferulic acid in bread supplemented with nejayote. The loss of antioxidant properties during cooking can be avoided by the use of the microencapsulation technique as shown when supplementation with green tea extract is compared to supplementation with microencapsulated green tea extract. Thus, although baking reduces antioxidant activity, fortification results in increased antioxidant activity compared to unfortified breads.

The types of agents used in fermentation, an essential step in bread making, also influence antioxidant activity by altering the fermenting agent. In brewer's spent grain breads, sourdough and enzymes did not change the phenolic profile but did increase the *in vitro* antioxidant activity of breads [42]. The addition of legume (chickpea, lentil and bean) flours to wheat flour bread with a sourdough fermentation agent containing lactic acid bacteria showed a marked increase in DPPH radical quenching ability and phytase activity [43]. Similarly, when a micronized by-product of durum wheat was added to bread, better nutritional quality was observed in sourdough bread compared with yeast-fermented dough. In contrast, *Monascus* mould improved bread enriched with anka (fermented rice) by supplying monacolin K, GABA and dietary fibre with beneficial health effects. However, fermented citrus peel showed lower antioxidant activity than unfermented citrus peel when added to bread. Newer stress-resistant yeast strains producing antioxidant enzymes are being developed to enhance the fermentation ability of bread dough [44].

Finally, baking of bread in an oven or on a hearth significantly affects its phenolic profile and scavenging activity due to the formation of new products from sensitive compounds like polyphenols, reducing sugars and protein. The Maillard reaction and caramelization are the main chemical reactions during baking. The Maillard reaction is a chemical reaction between amino acids and reducing sugars that gives browned food its desirable flavour and in baking causes the formation of a brown crust. Maillard reaction products also con-

tribute to antioxidant activity. In bread fortified with intermediated pearled wheat fraction, more TPC content was found than supplied by the raw materials due to formation of new intermediate phenolic products through the Maillard reaction. The antioxidant activity of rye bread, especially in the crust, was also affected by the Maillard reaction occurring during baking. Advanced Maillard reaction products resulted in good scavenging of peroxy and ABTS radicals and increased TPC, enhancing the formation of antioxidants during rye bread baking [45]. Typical chestnut flour volatiles such as furans and pyrroles were formed due to same type of reaction, producing toasty and nutty notes which improved consumer acceptability. Also, natural antioxidant sources like grape seed extract showed inhibitory effects on the formation of the detrimental compound CML in crust during baking. The interaction between phenolics (flavonoids) and protein affects the antioxidant efficacy of flavonoids, as observed following onion skin fortification of bread [46]. Apart from their antioxidant activity, natural sources of antioxidants affected the digestive system. Negative effects resulting from the binding of phenolic compounds to protein and starch have been mentioned above. Similarly, inhibition of enzymes (protease and amylase) also reduces protein and starch digestibility, resulting in increased levels of free amino groups and resistant starch [10]. The anti-inflammatory activity associated with maca-enriched bread, *in vitro* release of catalase activator and the anticancer activity of broccoli sprout-fortified bread demonstrate the dynamic role of natural antioxidant sources. Supplementation of wheat flour usually increased dietary fibre, which consequently affected glucose absorption. *S. rebaudiana* extracts added to wholegrain flours decreased glucose absorption by increasing dietary fibre, while the addition of maca flour had an inhibiting effect on α -amylase and α -glucosidase, thus also hindering glucose absorption. Different types of fibre have different impacts. When whole wheat flour, which has a high content of insoluble dietary fibre, was supplemented with cellulose, the amount of resistant starch increased while the amount of digestible starch decreased, thus delaying starch digestion.

In contrast, barley, oat and rye contain higher levels of soluble dietary fibre and so have less effect on lowering glucose absorption [28].

The type of wheat flour used was also a major factor affecting antioxidant activity. The effect of the coat colour of wheat was studied. Investigations showed that red and white wheat grain flour showed different phenolic acid profiles despite having comparable total phenolic acid content (TPAC) [47]. A similar effect was observed when red and white sorghum flours were added to wheat bread. Kisra, a traditional flat Sudanese bread, was prepared using two different sorghum cultivars. The two types of bread showed significantly different FRAP and DPPH values, which were both enhanced by baking, with increased fermentation time also resulting in higher values and hence increased antioxidant activity [48]. Therefore, a change in type of dough flour can lead to a remarkable change in antioxidant activity. Another investigation found that, despite loss of TPC and antioxidant activity following baking, breads containing pseudocereals showed significantly higher antioxidant capacity than bread made only with wheat flour [49].

The physical properties of a fortifying agent have a combined effect on overall acceptability. Gluten quality and quantity are important characteristics of bakery flour. Gluten quantity decreases when natural sources of antioxidants such as turmeric, ginger and coriander powder are added to wheat flour. High fibre and gluten-free flours such as rice bran, banana pseudo-stem, chestnut and sorghum, and spices such as fennel and cinnamon affect the gluten chain by diluting it with fibre and fibre-induced increased water absorption capacity. With a decreased bread volume, high moisture content also affects bread quality negatively. Consequently, a higher level of coriander supplementation resulted in improved bread due to the lower level of fibre. The addition of microionized by-products of debranned durum wheat to sourdough also increased loaf volume compared to the same supplementation of yeast dough.

Hydrocolloids like CMC when added to banana pseudo-stem-fortified bread improved physical properties by balancing soluble and insoluble dietary fibre content with decreased antioxidant activity. The hydrocolloid-like properties of natu-

rally formed alkali-treated fibre in nejayote solids also provide another method of improving bread. The darkness of crumb and crust is directly proportional to phenolic content and Maillard reaction. A higher level of reducing sugar in chestnut flour and higher levels of protein in rice bran increased the darkness of bread crust, while the crumb colour was affected by turmeric and coriander.

Another important parameter is taste which is perceived as flavour combined with aroma. The acceptability of the sweet aroma of fennel seed, the pungency of ginger and the woody notes of chestnut is related to the amount of fortification. Differences in processing like employing sourdough instead of yeast fermentation, masked the inferior flavour of grape by-product and improved the sensory profile of bread containing microionized by-products from debranned durum wheat. At increased level of *Stevia rebaudiana* the product becomes less sweet than control due to the presence of chlorophyll pigment and glycosides. In general, higher levels of fortification with natural sources of antioxidants negatively affect bread quality, so it is important to identify of optimum levels of supplementation.

In addition to the various processing parameters, storage conditions also affect antioxidants in bread. One study showed that the TPC and oxidative stability of bread samples decreased during storage depending on the manufacturing process. However, frozen dough bread lost less phenolics and had higher oxidative stability compared to frozen baked bread [50]. This indicates the fortification ingredient as well as various environmental, physical and chemical factors affect the antioxidant capacity of breads enriched with natural sources of antioxidants.

Acknowledgements

This research was financially supported by the Basic Scientific Research (BSR) Fellowship for Meritorious Students Programme under the University Grants Commission (UGC), India.

Conflicts of Interest

The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

REFERENCES

- Blomhoff R (2010) Role of dietary phytochemicals in oxidative stress. In: Bernhoft A (ed) Bioactive compounds in plants – benefits and risks for man and animals. Proceedings from a symposium held at The Norwegian Academy of Science and Letters and the publisher area is Oslo. The Norwegian Academy of Science and Letters, Oslo, pp 52–70
- Block G, Langseth L (1994) Antioxidant vitamins and disease prevention. *Food Technol* 48:80–84
- Orphanides A, Goulas V, Gekas V (2013) Effect of drying method on the phenolic content and antioxidant capacity of spearmint. *Czech J Food Sci* 31:509–513
- Valko M, Leibfritz D, Moncol J, Cronin MTD, Mazur M, Telser J (2007) Free radicals and antioxidants in normal physiological functions and human disease. *Int J Biochem Cell Biol* 39:44–84
- Dziki D, Różyło R, Dziki UG, Świeca M (2014) Current trends in the enhancement of antioxidant activity of wheat bread by the addition of plant materials rich in phenolic compounds. *Trends Food Sci Tech* 40:48–61
- Zhu F, Sakulnak R, Wang S (2016) Effect of black tea on antioxidant, textural, and sensory properties of Chinese steamed bread. *Food Chem* 194:1217–1223
- Irakli M, Katsantonis D, Kleisiaris F (2015) Evaluation of quality attributes, nutraceutical components and antioxidant potential of wheat bread substituted with rice bran. *J Cereal Sci* 65:74–80
- Pasirja D, Ezhilarasi PN, Indrani D, Anandharamakrishnan C (2015) Microencapsulation of green tea polyphenols and its effect on incorporated bread quality. *LWT Food Sci Technol* 64:289–296
- Ruiz-Ruiz JC, Moguel-Ordoñez YB, Matus-Basto AJ, Segura-Campos MR (2015) Antidiabetic and antioxidant activity of *Stevia rebaudiana* extracts (var. Morita) and their incorporation into a potential functional bread. *J Food Sci Technol* 52:7894–7903
- Świeca M, Sęczyk L, Dziki UG, Dziki D (2014) Bread enriched with quinoa leaves – the influence of protein-phenolics interactions on the nutritional and antioxidant quality. *J Food Chem* 162:54–62
- Acosta-Estrada BA, Lazo-Vélez MA, Nava-Valdez Y, Gutiérrez-Urbe JA, Serna-Saldívar SO (2014) Improvement of dietary fiber, ferulic acid and calcium contents in pan bread enriched with nejayote food additive from white maize (*Zea mays*). *J Cereal Sci* 60:264–269
- Gawlik-Dziki U, Świeca M, Dziki D, Swczyk Ł, Złotek U, Różyło R, Kaszuba K, Ryszawy D, Czyż J (2014) Anticancer and antioxidant activity of bread enriched with broccoli sprouts. *BioMed Res Int* 2014:608053
- Gawlik-Dziki U, Świeca M, Dziki D, Baraniak B, Tomiło J, Czyż J (2013) Quality and antioxidant properties of breads enriched with dry onion (*Allium cepa* L.) skin. *J Food Chem* 138:1621–1628
- Dall’Asta C, Cirlini M, Morini E, Rinaldi M, Ganino T, Chiavaro E (2013) Effect of chestnut flour supplementation on physico-chemical properties and volatiles in bread making. *LWT Food Sci Technol* 53:233–239
- Ho LH, Aziz NAA, Azahari B (2013) Physico-chemical characteristics and sensory evaluation of wheat bread partially substituted with banana (*Musa acuminata* X *balbisiana* cv. Awak) pseudo-stem flour. *J Food Chem* 139:532–539
- Das L, Raychaudhuri U, Chakraborty R (2013) Herbal fortification of common bread with fennel seeds. *Food Technol Biotechnol* 51:434–440
- Dhillon GK, Amarjeet K (2013) Quality evaluation of bread incorporated with different levels of cinnamon powder. *Int J Food Sci Nutr Diet* 2:70–74
- Shyu YS, Lu TC, Lin CC (2013) Functional analysis of unfermented and fermented citrus peels and physical properties of citrus peel-added doughs for bread making. *J Food Sci Technol* 51:3803–3811
- Blandino M, Sovrani V, Marinaccio F, Reyner A, Rolle L, Giacosa S, Locatelli M, Bordiga M, Travaglia F, Coisson JD, Arlorio M (2013) Nutritional and technological quality of bread enriched with an intermediated pearled wheat fraction. *J Food Chem* 141:2549–2557
- Puoci F, Malanchin R, Piangiolino C, Restuccia D, Curcio M, Parisi OI, Cirillo G, Picci N (2013) Maca flour: a powerful ingredient for functionally enhanced bread. *Int Food Res J* 20:1293–1300
- Rizzello CG, Coda R, Mazzacane F, Minervini D, Gobetti M (2012) Micronized by-products from debranned durum wheat and sourdough fermentation enhanced the nutritional, textural and sensory features of bread. *Food Res Int* 46:304–313
- Yousif A, Nhepera D, Johnson S (2012) Influence of sorghum flour addition on flat bread in vitro starch digestibility, antioxidant capacity and consumer acceptability. *J Food Chem* 134:880–887
- Hao M, Beta T (2012) Development of Chinese steamed bread enriched in bioactive compounds from barley hull and flaxseed hull extracts. *J Food Chem* 133:1320–1325
- Alu’datt MH, Rababah T, Ereifej K, Alli I, Alrababah MA, Almajwal A, Masadeh N, Alhamad MN (2012) Effects of barley flour and barley protein isolate on chemical, functional, nutri-

- tional and biological properties of pita bread. *J Food Hydrocolloids* 26:135–143
25. Das L, Raychaudhuri U, Chakraborty R (2012) Supplementation of common white bread by coriander leaf powder. *Food Sci Biotechnol* 21:425–433
 26. Balestra F, Cocci E, Pinnavaia G, Romani S (2011) Evaluation of antioxidant, rheological and sensorial properties of wheat flour dough and bread containing ginger powder. *LWT Food Sci Technol* 44:700–705
 27. Lim HS, Park SH, Ghafoor K, Hwang SY, Park J (2011) Quality and antioxidant properties of bread containing turmeric (*Curcuma longa* L.) cultivated in South Korea. *J Food Chem* 124:1577–1582
 28. Ragaee S, Guzar I, Dhull N, Seetharaman K (2011) Effects of fiber addition on antioxidant capacity and nutritional quality of wheat bread. *LWT Food Sci Technol* 44:2147–2153
 29. Tseng YH, Yang JH, Chen CH, Mau JL (2011) Quality and antioxidant properties of anka enriched bread. *J Food Process Preserv* 35:518–523
 30. Mildner-Szkudlarz S, Zawirska-Wojtasiak R, Szwengiel A, Pacynski M (2010) Use of grape by-product as a source of dietary fibre and phenolic compounds in sourdough mixed rye bread. *Int J Food Sci Tech* 46:1485–1493
 31. Chlopicka J, Pasko P, Gorinstein S, Jedryasa A, Zagrodzki P (2010) Total phenolic and total flavonoid content, antioxidant activity and sensory evaluation of pseudocereal breads. *LWT Food Sci Technol* 46:548–555
 32. Peng X, Maa J, Cheng KW, Jiang Y, Chen F, Wang M (2010) The effects of grape seed extract fortification on the antioxidant activity and quality attributes of bread. *J Food Chem* 119:49–53
 33. Filipčev B, Lević L, Bodroža-Solarov M, Mišljenović N, Koprivica G (2010) Quality characteristics and antioxidant properties of breads supplemented with sugar beet molasses-based ingredients. *Int J Food Prop* 13:1035–1053
 34. Holtekjølen AK, Bævre AB, Rødbotten M, Berg H, Knutson SH (2008) Antioxidant properties and sensory profiles of breads containing barley flour. *J Food Chem* 110:414–421
 35. Raba DN, Moigradean D, Poiana MA, Popa M, Jianu I (2007) Antioxidant capacity and polyphenols content for garlic and basil flavored bread. *J Agroalimnt Proc Technol* 13:163–168
 36. Fan L, Zhang S, Yu L, Ma L (2006) Evaluation of antioxidant property and quality of breads containing *Auricularia auricula* polysaccharide flour. *Food Chem* 101:1158–1163
 37. Hsu CL, Hurang SL, Chen W, Weng WM, Tseng CY (2004) Qualities and antioxidant properties of bread as affected by the incorporation of yam flour in the formulation. *Int J Food Sci Tech* 39:231–238
 38. Butu M, Rodino S, Butu A, Butnariu M (2014) Screening of bioflavonoid and antioxidant activity of *Lens culinaris Medikus*. *Dig J Nanomater Bios* 9:519–529
 39. Butnariu M, Council A (2013) Design and management of functional foods for quality of life improvement. *Ann Agric Environ Med* 20:736–741
 40. Butnariu M, Coradini C Z (2012) Evaluation of biologically active compounds from *Calendula officinalis* flowers using spectrophotometry. *Chem Cent J* 6:35
 41. Abdel-Aal ESM, Rabalski I (2013) Effect of baking on free and bound phenolic acids in wholegrain bakery products. *J Cereal Sci* 57:312–318
 42. Ktenioudaki A, Alvarez-Jubete L, Smyth TJ, Kilcawley K, Rai DK, Gallagher E (2015) Application of bioprocessing techniques (sourdough fermentation and technological aids) for brewer's spent grain breads. *Food Res Int* 73:107–116
 43. Rizzello CG, Calasso M, Campanella D, Angelis MD, Gobetti M (2014) Use of sourdough fermentation and mixture of wheat, chickpea, lentil and bean flours for enhancing the nutritional, texture and sensory characteristics of white bread. *Int J Food Microbiol* 180:78–87
 44. Sasano Y, Takahashi S, Shima J, Takagi H (2010) Antioxidant N-acetyltransferase Mpr1/2 of industrial baker's yeast enhances fermentation ability after air-drying stress in bread dough. *Int J Food Microbiol* 138:181–185
 45. Michalska A, Amigo-Benavent M, Zielinski H, del Castillo MD (2008) Effect of bread making on formation of Maillard reaction products contributing to the overall antioxidant activity of rye bread. *J Cereal Sci* 48:123–132
 46. Świeca M, Gawlik-Dziki U, Dziki D, Baraniak B, Czyz J (2013) The influence of protein–flavonoid interactions on protein digestibility in vitro and the antioxidant quality of breads enriched with onion skin. *J Food Chem* 141:451–458
 47. Challacombe CA, Abdel-Aal ESM, Seetharamana K, Duizer LM (2012) Influence of phenolic acid content on sensory perception of bread and crackers made from red or white wheat. *J Cereal Sci* 56:181–188
 48. Zaroug M, Orhan IE, Senol FS, Yagi S (2014) Comparative antioxidant activity appraisal of traditional Sudanese kiswa prepared from two sorghum cultivars. *J Food Chem* 156:110–116
 49. Alvarez-Jubete L, Wijngaard H, Arendt EK, Gallagher E (2010) Polyphenol composition and in vitro antioxidant activity of amaranth, quinoa, buckwheat and wheat as affected by sprouting and baking. *Food Chem* 119:770–778
 50. Novotni D, Curi D, Galic K, Skevin D, Nederal S, Kraljic K, Gabric D, Jezek D (2011) Influence of frozen storage and packaging on oxidative stability and texture of bread produced by different processes. *LWT Food Sci Technol* 44:643–649

Characterization of physicochemical properties in whole wheat bread after incorporation of ripe mango peel

Debasmita Pathak¹ · Jayshree Majumdar¹ · Utpal Raychaudhuri¹ · Runu Chakraborty¹

Received: 27 June 2015 / Accepted: 18 April 2016 / Published online: 26 April 2016
© Springer Science+Business Media New York 2016

Abstract In the present study, the effect of ripe mango peel powder (MPP) at 0, 1, 3, and 5 % on rheological, physical, textural, color, antioxidant and sensory properties of whole wheat bread were evaluated. Rheological data of wheat flour incorporated with MPP showed an increase in viscoelastic property. However, the loaf height, weight loss percentage and specific volume were decreased with amount of incorporation. Bread density along with crumb moisture was increased after addition of MPP. The brownness index of bread samples were significantly different and whiteness index were also decremented. For textural analysis hardness, cohesiveness and springiness were also increased with level of addition of MPP. Breads incorporated with MPP exhibited an improvement in antioxidant properties as total phenolic content increased from $(220.33 \pm 8.5$ to $757.8 \pm 13.5)$ mg GAE/100 gm along with the highest value of 2,2-diphenyl-1-picrylhydrazyl (DPPH) inhibition and ferric reducing antioxidant power of 65.74 ± 0.51 % DPPH inhibition and 1436.25 ± 88 μM $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}/100$ g respectively. The MPP levels elevated the fruity aroma, crumb colour, fruity taste, after taste and score for oral texture descriptors in quantitative descriptive analysis. However, at 5 % level porosity and traditional bread aroma decreased significantly. Thus, at optimised level without affecting the sensory and physical properties 3 % MPP addition in whole wheat bread was regarded as best.

Keywords Ripe mango peel · Whole wheat bread · Rheology · Baking · Texture · Antioxidant · Sensory properties

Introduction

Mango (*Mangifera indica* L.) is one of the most cultivated fruit which is consumed due to its delicious sweet taste. The world production of mango is more than 43 millions tons [1]. India is the largest producer of mangoes with 44.14 % of the total world production [2]. Mango consist of 33–85 % edible pulp, with 9–40 % inedible kernel and 7–24 % inedible peel [3]. Having high nutritional value with high range of polyphenols and specific enzyme contents, makes it a high potential candidate for nutraceutical use.

As a seasonal fruit, mango is highly perishable. Mango is processed and preserved in different forms like candy, leather, puree, nectar, jam, jelly, pickles and canned slices [4]. Because of this industrial processing, a huge amount of waste is being generated during production. Such by-products, which mainly consist of mango peel and kernels are a serious disposal problem. So, one should take actions towards either treatment or for sustainable techniques as a method for by-product utilisation in mango processing industry. This major step can lead to minimize the burden for appropriate waste treatment on local administrative bodies as well as industries.

It is known from literature that the peel contains different phytochemicals like polyphenols, carotenoids [5], vitamin E, dietary fibres [6] and vitamin C. It was proven that mango peel consists of good amount of antioxidant [7], anti proliferative [8] and antimicrobial properties [9]. Researchers have shown that the peel can be utilized as

✉ Runu Chakraborty
crunu@hotmail.com

¹ Department of Food Technology and Biochemical Engineering, Jadavpur University, Kolkata 700032, India

mango peel powder in biscuit [10], cookies [11], yoghurt [12], macaroni [13], muffin [14] and wine preparation [15] etc.

Thus, mango peel utilisation alone can lead to an effective way to not only for prevention of health and human hazards, but also have a commercial viability by the justified incorporation. As well as consumption of fruit polyphenols have health beneficial effects like preventing cardiovascular diseases, cancers, neurodegenerative diseases, Alzheimer's disease and ageing etc. [16]. From ancient age due to its convenience of making, affordability and ease in consumption, bread becomes a staple processed food. For this aspect ripe mango peel can act as potential source of antioxidant in bread.

In our study, oven dried ripe mango peel powder (MPP), which was been incorporated at different levels (1, 3 and 5 %) in whole wheat bread to enhance its antioxidant properties. Thus, aim of our investigation is to study the improvement in antioxidant properties of whole wheat bread along with the sensory properties of the bread as well as the physical, rheological properties of dough and baking properties by addition of MPP powder at different level of enrichment.

Materials and methods

Materials

Ripe mangoes were procured from local market of Jadavpur, Kolkata, India. After that they were washed with water to remove dirt. Further the cleaned peels were blanched and spread in thin layers in trays and dried at 60 ± 2 °C using a Tray dryer (Reliance Enterprise, Kolkata, India) for 18 h till the weight became constant. The dried peel was powdered using a Grinder (GX7, Bajaj Electricals Ltd, India) and was stored in airtight packets. Whole wheat flour, granulated sugar, salt, refined oil (as shortening agent) were purchased from the local grocery stores of Jadavpur, Kolkata. Compressed Baker's yeast (Saf Yeast Company Pvt. Ltd, Mumbai, India) was used as the leavening agent in bread preparation. Glycerol monostearate (GMS) (Loba Chemie Pvt. Ltd, Mumbai, India) and Potassium bromate (KBrO_3) (Merck Specialities Pvt. Ltd, Mumbai, India) were used in bread formulation. 2,4,6-tripyridyl-s-triazine (TPTZ) (Himedia Laboratories Pvt., Ltd, Mumbai, India), anhydrous ferric chloride (Rankem, New Delhi, India), ferrous sulphate heptahydrate, ethanol, Folin-Ciocalteu reagent, sodium carbonate, sodium acetate, acetic acid (Merck Specialities Pvt., Ltd, Mumbai, India), gallic acid (SD Fine-Chem Ltd, Mumbai, India) and DPPH (Sigma-Aldrich, St. Louis, MO, USA) were used.

Dough and bread preparation

The different dough samples of wheat flour (g) were added with MPP at the 0, 1, 3 and 5 % level. Whole wheat bread formulated without MPP (0 %) was used as control.

Straight dough method was applied for preparation of the dough. The dough was prepared by mixing flour-100 g, sugar-6 g, shortening-2.0 g, salt/NaCl-1.5 g, KBrO_3 -1 ppm, GMS 1.5 g and water at optimum amount along with pre activated compressed yeast-3.0 g in 20 ml luke-warm water and 2 g sugar in a bowl. The prepared dough was firstly proofed for 30 min at 30 ± 2 °C and consecutive second proofing was done in greased moulds at 40 ± 2 °C for 1 h with a cover of wet cloth in both the fermentation steps to maintain the moisture content. In between the proofing, the dough was punched along with slight kneading for removal of excess gas CO_2 . Fermented dough was baked in a rotary oven (CM HS108; Chanmag Bakery Machine Co., Ltd, Taipei, Taiwan) at 215 ± 2 °C for 18–20 min. After baking, bread samples were cooled down for about 1 h at room temperature (22 ± 1 °C) and stored in airtight packages.

Rheological measurements of dough

Each sample represents a dough prepared with different levels of MPP. Small deformation rheological measurements were performed by using a controlled stress strain rheometer (MCR 300, manufactured by Physica/Anton Paar; Ostfildern, Germany), with parallel plate geometry of 49.986 mm with RheoPlus software package (version 2.65). The upper plate was lowered until the thickness of sample was adjusted to 2 mm where the dough was placed in between them and the excess dough was trimmed off carefully. The exposed surface was covered with a wet layer of filter paper to prevent moisture loss during the test. All samples were allowed to rest for 5 min before measurements to allow dough relaxation. A frequency sweep test, angular frequency ranging from 0.1 to 200 ω , at a constant strain of 1 % was used to study the dough rheological changes. All experiments were carried out at a constant temperature of 25 °C. The dynamic rheological properties of samples were measured by the storage modulus G' , loss modulus G'' . Damping factor $\tan \delta$ and complex viscosity η^* [17].

Analysis of bread

Physical properties

The moisture content (dry basis) of samples was determined by AACC standard method where the initial weight (3 g) and final weight difference is measured by earlier

drying the crumb of samples in a hot air oven (Reliance Enterprise, Kolkata, India) at 105 °C, shown in Eq. (1) [18].

$$\begin{aligned} & \% \text{ moisture content} \\ & = \frac{\text{Initial wt. of the sample} - \text{final wt. of the sample}}{\text{Initial wt. of the sample}} \times 100 \end{aligned} \quad (1)$$

Loaf height was measured using a scale. The average of the three readings was recorded.

Volumes of the loaves were measured by the seed displacement method. From these, the specific volume and loaf density was calculated as described in Eqs. (2) and (3) respectively [19, 20]

$$\text{Specific volume (cm}^3\text{/gm)} = \frac{\text{(Loaf volume of bread)}}{\text{(Weight of bread)}} \quad (2)$$

$$\text{Density (gm/cm}^3\text{)} = \frac{\text{(Weight of bread)}}{\text{(Loaf volume of bread)}} \quad (3)$$

The dough and the baked loaf were weighed, and the percent weight loss was calculated as described in Eq. (4) [21]

$$\begin{aligned} \% \text{ Weight loss} & = \frac{\text{Weight of dough} - \text{Weight of baked bread}}{\text{Weight of dough}} \\ & \times 100 \end{aligned} \quad (4)$$

Texture

Bread textural characteristics were evaluated with a Texture Analyser (TA.HD Plus Texture Analyzer, USA) equipped with a 35 mm diameter aluminium cylinder probe. Before performing the analysis, for each sample, slices were cut from the central portion of four different bread loaves of dimensions of 40 mm × 40 mm × 30 mm. Texture profile analysis (TPA) was performed with in 30–60 % deformation and 0.1–2 mm/s cross-head speed. The considered textural parameters were hardness, cohesiveness and springiness [22].

Colour

The colour parameters L* (lightness), a* (redness to bluishness), b* (yellowness to greenness) of the baked loaves crumbs were determined using a digital colorimeter (ColourTec PCM, Accuracy Micro Sensor Inc., USA).

To determine brownness index (BI) given in Eqs. (5) and (6), colour parameters L*, a*, b* were determined at each point of the top crust of bread. [23]

$$\text{BI} = \frac{[100(X - 0.31)]}{0.17} \quad (5)$$

$$\text{where, } X = \frac{(a+1.75L)}{(5.645 \times L + a - 3.012b)}$$

Whiteness index of breads given in Eq. (4) was measured on crumb [24].

$$\text{WI} = 100 - \left([100 - L^*]^2 + a^{*2} + b^{*2} \right)^{1/2} \quad (6)$$

Antioxidant activity

Sample extraction

The sample extraction procedure was used to determine the total phenolics content and the radical scavenging activity. The breads were sliced; crust and crumb were separated manually. 1 g of sample was weighed and mixed with 80 % ethanol. Then the mixture was sonicated to agitate the particle in sample. Then it was centrifuged at 8944 × g for 15 min. Finally the supernatant was filtered and the extraction was completed by storing them in tubes at very low temperature or in refrigerator.

Total phenolics content

The extract amounting 0.5 ml was dissolved in 1.8 ml of distilled water and 0.2 ml of the Folin–Ciocalteu reagent was added. After 5 min, 2 ml of 7 % sodium carbonate was added and the contents mixed thoroughly by further addition of 0.8 ml distilled water again. Incubated at room temperature for 90 min, colour developed and the absorbance was observed at 750 nm by UV–Vis spectrophotometer (U 2800; Hitachi, Tokyo, Japan) using gallic acid as a standard. The results were expressed as mg GAE/100 g fresh material [25].

DPPH free radical scavenging activity

Radical scavenging activity was determined by using stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) as the free radical. The extraction of antioxidants was conducted as reported above. The decrease in absorbance at 515 nm was determined at 25 °C by spectrophotometrically. The percentage of inhibition or the percentage of discoloration was calculated [26].

Ferric reducing antioxidant power (FRAP) assay

FRAP assay was based on the reduction of Fe³⁺-TPTZ to a blue coloured Fe²⁺-TPTZ. The FRAP reagent was freshly prepared by mixing acetate buffer (pH 3.6), TPTZ solution

(10 mM TPTZ in 40 mM HCl) and 20 mM FeCl₃ solution in 10:1:1 v/v ratio. 100 µl of sample was taken and 3 ml of the FRAP reagent added into it. The absorbance was taken at the interval of 0 and 30 min (after incubating at 37 °C) at 593 nm. The antioxidant potential of the extracts was determined against a standard curve plotted using µmol FeSO₄·7H₂O/l [27].

Sensory evaluation

The evaluation of the sensory attributes of the MPP fortified bread was done by the method of quantitative descriptive analysis (QDA). The first step which consists of selection of judges was done by selecting members from faculty and students of Department of Food Technology and Biochemical Engineering, Jadavpur University, Kolkata. The panel constituted 50 judges aged between 20 and 55 years and 25 females and 25 males. To distinguish the descriptive feature for the bread, the panellists devised 8 descriptors for the bread and defined each of the feature of the sensory evaluation who covered the areas of appearance, taste, aroma and oral texture. Porosity, traditional bread aroma, fruity aroma, crumb colour, after taste, fruity flavour hardness and stickiness were the parameters on which the MPP incorporated bread were assessed. The panel members were trained in three training sessions of 1 h each to inculcate the ability to discriminate between the samples. Finally, the assessment was conducted in individual booths under white light at room temperature at Jadavpur University, Dept of Food Technology. For evaluation, approximately 1 cm thick slices including the crust and crumb, was presented to assessors. The assessors evaluated the samples on an unstructured 9 cm line scale labelled on the left by 'none' and on the right with the term 'strong'. Ratings were registered on an evaluation form. The samples were kept in a 3-digit coded glass covered with a glass lid. Samples were assessed in triplicates in four sets [28].

Statistical analysis

The experimental design was completely randomized with three replicates and expressed as mean value ± SD. All experimental data were analyzed statistically for analysis of variance (ANOVA) using Microsoft Excel 2007. The scores for QDA was also analysed by analysis of variance (two-way ANOVA as factors). The comparisons between the mean values were tested using Duncan's new multiple-range test at a level of $p \leq 0.05$.

Results and discussion

Rheological measurements of dough

Frequency sweep tests were performed to investigate the effects of MPP on the rheological parameters such as storage modulus (G'), loss modulus (G'') and damping factor ($\tan \delta$) of bread dough. In case of MPP substituted samples of 1, 3 and 5 % as well as control both G' and G'' showed frequency dependence behaviour i.e. both the modulus increased with the increase in angular frequency which is shown in Fig. 1. G' was greater than G'' in all the angular frequencies studied, indicating viscoelastic soft solid nature of the dough. The frequency sweep data showed that the dough from all the samples displayed higher G' and G'' at higher frequencies compared to those at low angular frequencies. Dough at lower frequencies showed lower $\tan \delta$ values that increased with the increase in the angular frequency indicated in Table 1. A similar change in G' , G'' and $\tan \delta$ of dough with the increasing frequency was observed due to incorporation of ginger powder, where active ginger components affected the dough structure due to cross-linking by the active sites [29]. The complex viscosity in frequency sweep profile was also showed as a linearly decreasing function with decreasing gradient of angular frequency which was earlier seen when hydrocolloids were added in dough [30]. However, as the percentage of incorporation increased η^* also increased and $\tan \delta$ showed a linear decrease with maximum and minimum at 5 % MPP bread respectively. This was likely due to interactions between the fibre structure and the gluten network leading to a denser dough structure.

Analysis of bread

Physical properties

For determining the moisture content (Table 2) of the bread samples, the crumb moisture of the samples was observed. The crumb moisture had a uniform linear relation with the amount of fortification which was highest at 5 %. The significant increase in moisture might be due to the presence of fibre in mango peel, which entrapped the moisture by increasing the water absorption activity of the dough [31].

There was a significant decrease in the specific volume of supplemented breads as compared to the control. In terms of loaf density, bread containing MPP at 1–5 % was found to be denser than control. This reflects that 5 % MPP incorporated bread had a tightly packed crumb structure.

Fig. 1 Storage modulus (G') and loss modulus (G'') for various levels of MPP substituted dough samples

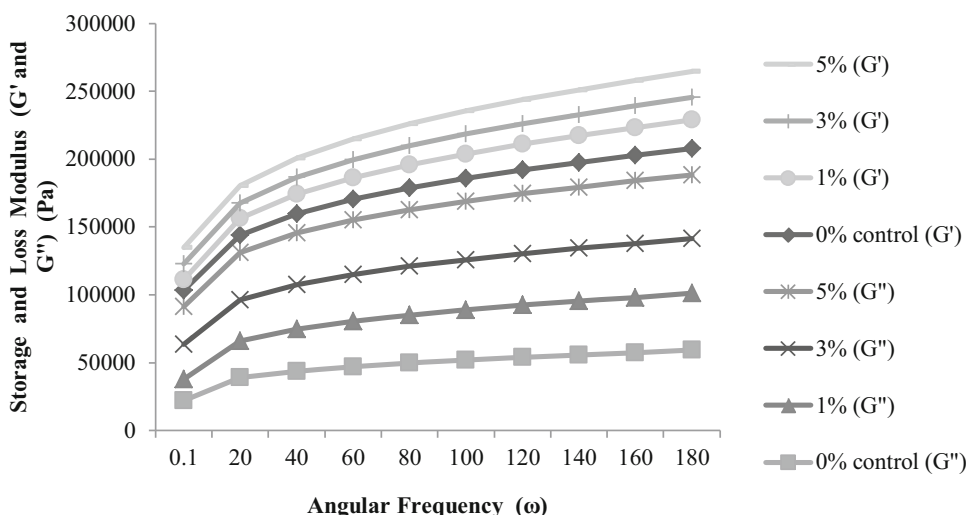


Table 1 Rheological properties of dough samples substituted with MPP at different levels

Parameters	0 % (control)	1 %	3 %	5 %
Complex viscosity (η^*) (Pa s)	3233 ± 331 ^a	3733 ± 135 ^{a,b}	4167 ± 292 ^b	4584 ± 207 ^c
Damping factor (tan δ)	0.554 ± 0.02 ^d	0.517 ± 0.01 ^c	0.484 ± 0.03 ^b	0.450 ± 0.01 ^a

Each value is expressed as mean ± SD ($n = 3$). Mean with different letters within a row are significantly different ($p \leq 0.05$)

Table 2 Physical properties of bread samples substituted with MPP at different levels

Parameters	0 % (control)	1 %	3 %	5 %
Moisture (%)	33.97 ± 0.45 ^a	37.51 ± 0.8 ^b	40.82 ± 1.14 ^c	44.41 ± 1.22 ^d
Specific volume (cm ³ /gm)	2.33 ± 0.06 ^d	2.20 ± 0.07 ^c	2.07 ± 0.02 ^b	1.87 ± 0.15 ^a
Density (g/cm ³)	0.41 ± 0.01 ^a	0.45 ± 0.03 ^b	0.48 ± 0.02 ^c	0.52 ± 0.02 ^d
Weight loss (%)	15.71 ± 0.70 ^d	13.01 ± 0.12 ^c	10.31 ± 0.26 ^b	7.16 ± 0.18 ^a
Loaf height (cm)	5.55 ± 0.21 ^c	4.7 ± 0.23 ^{b,c}	4.43 ± 0.14 ^b	4.0 ± 0.43 ^a

Each value is expressed as mean ± SD ($n = 3$). Mean with different letters within a row is significantly different ($p \leq 0.05$)

Possible explanation might be bread with less porous crumb structure representing increased density is inversely proportional to volume and subsequently low height [32]. The decrease in weight loss was significant at even 1 % addition and showed a range of 15.71 ± 0.70–7.16 ± 0.18 %. The same trend was followed in bread height also, where the decrease in height indicated a significant difference at ($p \leq 0.05$).

Texture

Hardness, cohesiveness and springiness of bread crumb samples with different percentages of MPP in whole wheat bread are indicated in Table 3. The incorporation of MPP led to linear increase in crumb hardness compared to control. This is comprehended by earlier studies where incorporation of chestnut flour increased hardness [33] and

may be due to the addition of fibre, which probably caused the thickening of the walls surrounding the air bubbles in the crumb. The higher amount of starch and sugars contained in MPP led the ability of a material to stick to itself. The cohesiveness of bread crumb ranged from 0.26 ± 0.02 to 0.82 ± 0.05 showed a linear increase, embarking that the 5 % MPP bread having the stickiest crumb. This increase was also studied earlier where subsequent addition elevated cohesiveness due to increase in stickiness [34]. In the very similar manner MPP addition, not only significantly affected the springiness but also showed a marked increment in compliance to the past studies.

Colour

Determination of bread crust's and crumb's colour is important to determine its acceptability. The whiteness

Table 3 Texture of the bread samples substituted with MPP at different levels

Parameters	0 % (control)	1 %	3 %	5 %
Hardness (N)	2.47 ± 0.32 ^a	3.0 ± 0.4 ^a	3.64 ± 0.26 ^b	3.93 ± 0.15 ^b
Cohesiveness	0.26 ± 0.02 ^a	0.34 ± 0.06 ^a	0.66 ± 0.07 ^b	0.82 ± 0.05 ^c
Springiness	0.29 ± 0.02 ^a	0.33 ± 0.06 ^b	0.41 ± 0.04 ^{b,c}	0.51 ± 0.05 ^c

Each value is expressed as mean ± SD ($n = 3$). Mean with different letters within a row is significantly different ($p \leq 0.05$)

index (WI) and the brownness index (BI) were evaluated as given in Table 4 as colour parameters for the sample prepared. The bread prepared by MPP showed a significant difference in WI compared to the supplemented samples. This may be due the effect of MPP imparting a brownish yellow colour in the bread crumb, which got dark at higher percentage of addition. This actually facilitates the acceptability by enhancing the brown colour of whole wheat bread, also observed in anka enriched bread [35].

However, the BI of the MPP incorporated sample increased as compared to the control. Nevertheless, the addition of mango peel gave a much desirable brown colour to the crust causing a darkening of brown colour of crust. This may be assumed due the sugar activity generated by Millard and caramelisation reaction, mentioned in earlier studies, the BI increase with the amount of incorporation in the bread formulation [36] [37]. This was obviously visible by linear decrease in L^* and increase in b^* values in case of BI and WI values. Fortification levels were inversely proportional with the L^* value of the crust and crumb and increase in b^* is due to the greenish colour intensification. This means that the crumb and crust stained towards darker colours, tending to brown and assuming a more vivid colour compared with the control bread.

Antioxidant activity

Total phenolics content (TPC) and total antioxidant activity of control bread and fortified bread samples with different percentages of MPP were observed and values were given in Table 5. There were significant differences in respect to total phenolic content between all breads prepared with MPP and control. 5 % incorporation of MPP showed the highest value of TPC 757.8 ± 13.5 mg GAE/100 gm compared to control. This led to the assumption, that

inclusion of MPP may elevate the amount of polyphenol amount in bread and other products like biscuit [10]. Mango peel is enriched with phenols because of its dietary fibres, various phenolic compound and vitamins [38]. The same relation can be found in mango peel fortified biscuits.

The similar trend was seen in antioxidant activity by measuring the FRAP (Ferric Reducing Antioxidant Power) and DPPH scavenging property of the incorporated bread. The DPPH and FRAP values showed a linear increase with the amount of incorporation of MPP. In case of FRAP up to 3 % addition resulted in twice the amount of FRAP value. However a significant greater increment was also seen in 5 % incorporation indicating a value of 1436.25 ± 88 μ M $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}/100$ g. The range for DPPH showed the value from 21.69 ± 0.72 % inhibition to 65.74 ± 0.51 % inhibition, resulting in a threefold increase till 5 % incorporation. Previous studies also revealed that mango peel incorporation in baked products increased antioxidant activity [39].

Sensory evaluation

The scores for the various descriptors for MPP fortified bread is shown in Table 6. Considering the crumb appearance depicted by porosity, the intensity decreased significantly when the level of addition was higher than 1 %. However, the traditional bread aroma also decreased the amount of fortification increased showing an inverse relationship with the fruity aroma. The fruity taste which should be somehow related to the sugar incorporation showed the highest value at 5 % MPP bread. These aromatic compounds found in peel might interfere and gave the characteristic bitter after taste, which was prevalent with the addition of MPP in bread. The crumb colour showed a linear increase and highest value at 5 % MPP in

Table 4 Colour of the bread samples substituted with MPP at different levels

Parameters	0 % (control)	1 %	3 %	5 %
WI	56.26 ± 1.11 ^b	50.49 ± 0.88 ^b	49.47 ± 0.93 ^a	48.11 ± 1.06 ^a
BI	81.39 ± 7.69 ^a	94.81 ± 5.65 ^b	117.52 ± 2.55 ^c	123.57 ± 4.61 ^c

Each value is expressed as mean ± SD ($n = 3$). Mean with different letters within a row are significantly different ($p \leq 0.05$)

WI whiteness index, BI brownness index

Table 5 Antioxidant activities of the bread samples substituted with MPP at different levels

Parameters	0 % (control)	1 %	3 %	5 %
TPC (mg GAE/100 gm)	220.33 ± 8.5 ^a	404.67 ± 16.04 ^b	507 ± 11.5 ^c	757.8 ± 13.5 ^d
FRAP (µM FeSO ₄ ·7H ₂ O/100 g)	158.77 ± 2.71 ^a	411.25 ± 32.19 ^b	1123.75 ± 11.58 ^c	1436.25 ± 88 ^d
DPPH (% inhibition)	21.69 ± 0.72 ^a	30.27 ± 1.03 ^b	50.29 ± 1.13 ^c	65.74 ± 0.51 ^d

Each value is expressed as mean ± SD ($n = 3$). Mean with different letters within a row are significantly different ($p \leq 0.05$)

Table 6 Sensory evaluation of the bread samples substituted with MPP at different levels

Attributes	Samples				SD
	0 % (control)	1 %	3 %	5 %	
Porosity	7.8 ^c	7.44 ^b	7.21 ^b	6.81 ^a	0.27
Traditional bread aroma	7.7 ^c	7.54 ^{b,c}	7.21 ^{a,b}	6.86 ^a	0.58
Fruity aroma	2.19 ^a	3.5 ^b	4.16 ^c	4.67 ^d	1.07
Fruity taste	1.5 ^a	2.25 ^b	3.38 ^c	4.8 ^d	1.45
After taste	1.59 ^a	2.4 ^b	3.01 ^c	3.92 ^d	0.98
Crumb colour	6.4 ^a	7.66 ^{a,b}	8.19 ^b	8.53 ^c	0.94
Hardness	4.2 ^a	4.7 ^b	5.5 ^c	6.16 ^d	0.84
Stickiness	4.49 ^a	6.24 ^b	6.62 ^c	6.97 ^c	1.1

Mean with different letters within a column is significantly different ($p \leq 0.05$)

bread due to the increase in Millard reaction products by addition of sugar from peels [40]. Following the same trend for oral texture descriptors the hardness and stickiness also increased significantly than control. The 5 % MPP substituted bread was significantly different from the control with highest hardness, stickiness, crumb colour as well as lower porosity. Thus, the addition of 3 % MPP in bread would be a diligent decision as up to this level the scores were well acceptable.

Conclusion

According to the present research work ripe mango peel powder could be regarded as a potential health-promoting functional ingredient. Both quantitative and qualitative aspect of antioxidant activity in MPP fortified bread sample showed a linear increase with respect to fortification level. Viscoelasticity of dough sample like antioxidant activity also showed a linear result with increased level of fortification. Interference of dietary fibre and phenolic content also imparted a negative impact on other physical parameters like hardness, WI, specific volume at maximum fortification level. But at 3 % level it showed a close difference with control sample in all physical parameters. This trend was also supported by sensory evaluation, where 3 % MPP level gained favourable scores. Thus, it is important to choose appropriate amount of mango peel powder and processing parameters to obtain healthy bread.

Acknowledgments This research work is financially supported by the Basic Scientific Research (BSR) fellowship for meritorious student programme under University Grants Commission (UGC), India.

References

- S.V. Jawale, J.N. Ghulghule, Constraints and suggestions of Kesar mango production in export zone of Marathwada region. *IJCBM* **4**(5), 713–721 (2015)
- FAOSTAT, Food and Agriculture Organization of the United Nations Statistics Division (2014), <http://faostat3.fao.org/browse/Q/QC/E>. Accessed 14 April 2016
- J. De La Cruz Medina, H.S. García, *MANGO: Post-harvest Operations* (FAO, Rome, 2002)
- H.T. Chan Jr, *Mango Processing and Products* (Tropical Fruit and Vegetable Research Laboratory, USDA-Agricultural Research Service, Hilo, Hawaii)
- H.E. Khoo, K.N. Prasad, A. Ismail, N. Mohd-Esa, Carotenoids from *Mangifera pajang* and their antioxidant capacity. *Molecules* **15**, 6699–6712 (2010)
- J.A. Larrauri, P. Rupérez, F. Saura-Calixto, Mango peel fibres with antioxidant activity. *Z. Lebensm. Unters. Forsch. A* **205**(1), 39–42 (1997)
- Z. Marina, A. Noriham, Quantification of total phenolic compound and in vitro antioxidant potential of fruit peel extracts. *IFRJ* **21**(5), 1925–1929 (2014)
- H.J.Y. Kim, H. Moon, D. Kim, M. Lee, H. Cho, Y.S. Choi, M.A. Kim, S.K. Cho, Antioxidant and antiproliferative activities of mango (*Mangifera indica* L.) flesh and peel. *Food Chem.* **121**(2), 429–436 (2010)
- S. Parashar, H. Sharma, M. Garg, Antimicrobial and antioxidant activities of fruits and vegetable peels: a review. *J. Pharmacogn. Phytochem.* **3**(1), 160–164 (2014)
- I.S. Ashoush, M.G.E. Gadallah, Utilization of mango peels and seed kernels powders as sources of phytochemicals in biscuit. *World J. Dairy Food Sci.* **6**(1), 35–42 (2011)

11. K. Bandyopadhyay, C. Chakraborty, S. Bhattacharyya, Fortification of mango peel and kernel powder in cookies formulation. *JAIR* **2**(12), 661–664 (2014)
12. O.I. El-Batawy, I.S. Ashoush, N.S. Mehanna, Impact of mango and pomegranate peels supplementation on quality characteristics of yoghurt with or without whey powder. *World J. Dairy Food Sci.* **9**(1), 57–65 (2014)
13. C.M. Ajila, M. Aalami, K. Leelavathi, U.J.S.P. Rao, Mango peel powder: a potential source of antioxidant and dietary fiber in macaroni preparations. *Innov. Food Sci. Emerg. Technol.* **11**(1), 219–224 (2010)
14. J. Ramirez-Maganda, F.J. Blancas-Benitez, V.M. Zamora-Gasga, M.L. García-Magaña, L.A. Bello-Pérez, J. Tovar, S.G. Sáyago-Ayerdi, Nutritional properties and phenolic content of a bakery product substituted with a mango (*Mangifera indica*) 'Ataulfo' processing by-product. *Food Res. Int.* **73**, 117–123 (2015)
15. S. Varakumar, K. Naresh, O.V.S. Reddy, Preparation of mango (*Mangifera indica* L.) wine using a new yeast-mango-peel immobilised biocatalyst system. *Czech J. Food Sci.* **30**(6), 557–566 (2012)
16. G. Pace, P. Lima, F. Vianello, C.R. Corrêa, R. Arnoux, M.G. Borguini, Polyphenols in fruits and vegetables and its effect on human health. *Food Nutr. Sci.* **5**, 1065–1082 (2014)
17. L. Das, U. Raychaudhuri, R. Chakraborty, Role of hydrocolloids in improving the physical and textural characteristics of fennel bread. *Int. Food Res. J.* **20**(5), 2253–2259 (2013)
18. Association of Official Agricultural Chemists (AOAC), *Official Method of Analysis*, vol. 14 (Association of Official Agricultural Chemists, Washington, DC, 1984)
19. C.L. Hsu, S.L. Hurang, W. Chen, Y.M. Weng, C.Y. Tseng, Qualities and antioxidant properties of bread as affected by the incorporation of yam flour in the formulation. *Int. J. Food Sci. Technol.* **39**(2), 231–238 (2004)
20. B.A. Acosta-Estrada, M.A. Lazo-Vélez, Y. Nava-Valdez, J.A. Gutiérrez-Urbe, S.O. Serna-Saldívar, Improvement of dietary fiber, ferulic acid and calcium contents in pan bread enriched with nejayote food additive from white maize (*Zea mays*). *J. Cereal Sci.* **60**, 264–269 (2014)
21. L.H. Ho, N.A.A. Aziz, B. Azahari, Physico-chemical characteristics and sensory evaluation of wheat bread partially substituted with banana (*Musa acuminata* X *balbisiana* cv. *Awak*) pseudo-stem flour. *Food Chem.* **139**, 532–539 (2013)
22. M.C. Bouane, Texture profile analysis. *Food Technol.* **32**(7), 62–66 (1978)
23. T.A. Shittu, A. Dixon, S.O. Awonorin, L.O. Sanni, B. Maziya-Dixon, Bread from composite cassava–wheat flour. II: effect of cassava genotype and nitrogen fertilizer on bread quality. *Food Res. Int.* **41**, 569–578 (2008)
24. C. Collar, A. Angioloni, Nutritional and functional performance of barleyflours in breadmaking: mixed breads vs wheat breads. *Eur. Food Res. Technol.* **238**, 459–469 (2014)
25. V.L. Singleton, R. Orthofer, R.M. Lamuela-Raventos, Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin–Ciocalteu reagent. *Methods Enzymol.* **299**, 152–178 (1999)
26. W. Brand-Williams, M.E. Cuveleir, C. Berset, Use of a free radical method to evaluate antioxidant activity. *LWT Food Sci. Technol.* **28**(1), 25–30 (1995)
27. I.F. Benzie, J.J. Strain, The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP, assay. *Anal. Biochem.* **239**, 70–76 (1996)
28. S.P. Heenan, J.P. Dufour, N. Hamid, W. Harvey, C.M. Delahunty, The sensory quality of fresh bread: descriptive attributes and consumer perceptions. *Food Res. Int.* **41**(10), 989–997 (2008)
29. F. Balestra, E. Cocci, G. Pinnavaia, S. Romani, Evaluation of antioxidant, rheological and sensorial properties of wheat flour dough and bread containing ginger powder. *LWT Food Sci. Technol.* **44**(3), 700–705 (2011)
30. L. Das, U. Raychaudhuri, R. Chakraborty, Effects of hydrocolloids as texture improver in coriander bread. *J. Food Sci. Technol.* **52**(6), 3671–3680 (2015)
31. H. Erkan, S. Celik, B. Bilgi, H. Koskel, A new approach for the utilization of barley in food products: barley tarhana. *Food Chem.* **97**, 12–18 (2006)
32. Y.S. Shyu, T.C. Lu, C.C. Lin, Functional analysis of unfermented and fermented citrus peels and physical properties of citrus peel-added doughs for bread making. *J. Food Sci. Technol.* **51**(12), 3803–3811 (2013)
33. C. Dall'Asta, M. Cirlini, E. Morini, M. Rinaldi, T. Ganino, E. Chiavaro, Effect of chestnut flour supplementation on physico-chemical properties and volatiles in bread making. *LWT Food Sci. Technol.* **53**, 233–239 (2013)
34. G.K. Dhillon, A. Kaur, Quality evaluation of bread incorporated with different levels of cinnamon powder. *IJFS* **II**(7), 1–6 (2013)
35. Y.H. Tseng, J.H.W.A. Yang, C.H. Chen, J.L. Mau, Quality and antioxidant properties of anka enriched bread. *J. Food Process. Pres.* **35**(4), 518–523 (2010)
36. C. Collar, T. Jiménez, P. Conte, C. Faddaba, Impact of ancient cereals, pseudocereals and legumes on starch hydrolysis and antiradical activity of technologically viable blended breads. *Carbohydr. Polym.* **113**, 149–158 (2014)
37. T.A. Shittu, A.O. Raji, L.O. Sanni, Bread from composite cassava–wheat flour: I. Effect of baking time and temperature on some physical properties of bread loaf. *Food Res. Int.* **40**, 280–290 (2007)
38. C.M. Ajila, S.G. Bhat, U.J.S.P. Rao, Valuable components of raw and ripe peels from two Indian mango varieties. *Food Chem.* **102**(4), 1006–1011 (2007)
39. C.M. Ajila, K. Leelavathi, U.J.S.P. Rao, Improvement of dietary fiber content and antioxidant properties in soft dough biscuits with the incorporation of mango peel powder. *J. Cereal Sci.* **48**(2), 319–326 (2008)
40. A. Michalskaa, M. Amigo-Benaventb, H. Zielinskia, M.D. del Castillo, Effect of bread making on formation of Maillard reaction products contributing to the overall antioxidant activity of rye bread. *J. Cereal Sci.* **48**, 123–132 (2008)

Study on enrichment of whole wheat bread quality with the incorporation of tropical fruit by-product

Pathak, D., Majumdar, J., Raychaudhuri, U. and *Chakraborty, R.

Department of Food Technology and Biochemical Engineering, Jadavpur University, Kolkata – 700032, India

Article history

Received: 23 August 2015

Received in revised form:

2 April 2016

Accepted: 7 April 2016

Abstract

In the present study Raw Mango Peel (RMP) powder was added to fortify whole wheat bread at three different levels (1%, 3% and 5 %) to increase its antioxidant properties than control (0% RMP powder) bread. This study also investigated the effects of RMP powder on the physical and sensory characteristics of bread along with the rheological effects on the dough. The content of polyphenols along with 2,2-diphenyl-1-picrylhydrazyl (DPPH) and Ferric reducing antioxidant powder (FRAP) value with a range of (21.51±0.45-68.54±2.15) % inhibition and (162.87±4.38-2232.31±47) $\mu\text{mol FeSO}_4 \cdot 7\text{H}_2\text{O} / 100\text{g}$ fresh sample respectively increased linearly, showing greatest level at 5% RMP powder. Increase in the level of RMP powder, percentage weight loss, loaf height, loaf volume and whiteness index (WI) decreased but crumb moisture and loaf density increased with a significant change in browning index (BI). Addition of peel also affected the rheological properties where addition leads to stiffer dough. Sensory evaluation through Quantitative Descriptive Analysis (QDA) showed that the incorporation of RMP powder increased the intensity of descriptors like hardness, rubbery texture, fruity aroma, after taste and decreased porosity and traditional bread aroma.

Keywords

Raw mango peel
Whole wheat bread
Antioxidant
Physical properties
Rheology
Sensory

© All Rights Reserved

Introduction

Tropical countries possess a wide diversity of fruits and cereals with many possibilities of commercial exploitation; some of them are considered rare and exotic. Mango (*Mangifera indica* L.) a drupe fruit, belongs to family Anacardiaceae order Rutales. Grown abundantly in 85 different countries in the world comprising both the tropical and sub tropical region of the globe with an overall production of 33.45 MMT is regarded as one of the most important tropical fruits (FAOSTAT, 2007). Interestingly, the total tropical fruit consumed, mango takes up 50% of it labelling it as one of the most popular edible fruits in the world. India is the supreme producer of the fruit contributing to about 60% of world production by producing 13.5MMT of mango fruits (Bhattacharjee *et al.*, 2011). As a seasonal fruit, raw mango is processed as some of the most popular food products such as puree, candy, amchoor (mango powder), nectar, leather, pickles, canned slices, marmalade, aam panna and chutney which are not only consumed nationwide but have also gained global market coverage and sales (Ravani and Joshi, 2013).

As some of the products are manufactured with the peel, many are formed by removal of the peel

to obtain the succulent flesh. In this technique of processing, mango, which consists of about 15-20% of the peel (Tunchaiyaphum *et al.*, 2013) is generated as the major by product from various sources. This peel is discarded as a waste, therefore, proper treatment or sustainable utilisation of peel in any form can minimise the cost of waste treatment for industry and local administration as well as could be a measure of value addition and environment friendly waste management technique. Most studies on the exploitation of mango peels have been dealing with their use as a source of pectin (Kermani *et al.*, 2015) which is considered as a high quality dietary fiber.

Mango peels were shown to be a good source of polyphenols, carotenoids, dietary fibre (pectin) and other bioactive compounds (Berardini *et al.*, 2005) as well as antiproliferative activities (Kim *et al.*, 2010) which possess various beneficial effects on human health including protection against cardiovascular diseases, cancer and other degenerative diseases. Therefore, due to its massive health promoting affects mango peels are incorporated in muffins (Ramirez-Maganda *et al.*, 2015), cookies (Bandyopadhyay *et al.*, 2014) and macaroni (Ajila *et al.*, 2010) as well as peels are fermented for wine (Varakumar *et al.*, 2012) preparation.

In our study, oven dried RMP powder, has been

*Corresponding author.

Email: crunu@hotmail.com

Tel: +91 983112 2626; Fax: +91 033 2414

incorporated at different levels (1%, 3% and 5%) in whole wheat bread to enhance its antioxidant properties. Thus, aim of our investigation was to study the improvement in antioxidant properties of whole wheat bread along with the physical, sensory and also rheological properties of the dough by addition of RMP powder at different levels.

Materials and Methods

Materials

Raw mangoes were procured from local market of Jadavpur, Kolkata, India. Whole wheat flour, granulated sugar, salt, refined oil (as shortening agent) was been purchased from the local grocery stores of Jadavpur, Kolkata. Compressed Baker's yeast (Saf Yeast Company Pvt. Ltd., Mumbai, India) was used as the leavening agent in bread preparation.

Chemicals

Chemicals, Glycerol monostearate (GMS) (Loba Chemie Pvt. Ltd., Mumbai, India) and Potassium bromate (KBrO_3) (Merck Specialities Pvt. Ltd., Mumbai, India) were also used in bread formulation. 2,4,6-tripyridyl-s-triazine (TPTZ) (Himedia Laboratories Pvt. Ltd., Mumbai, India), anhydrous ferric chloride (Rankem, New Delhi, India), ferrous sulphate heptahydrate, ethanol, Folin-Ciocalteu reagent, sodium carbonate, sodium acetate, acetic acid (Merck Specialities Pvt. Ltd., Mumbai, India), gallic acid (SD Fine-Chem Ltd., Mumbai, India) and DPPH (Sigma-Aldrich, St. Louis, MO, USA) were used in the investigation.

Preparation of the raw material

Raw mango peels were prepared by peeling freshly bought mangoes which were followed by washing with tap water to remove adhering dust and dirt particles. Then the peels were being blanched for enhancing its shelf life and improve the colour. The peel was spread in thin layers in trays and dried at $60 \pm 2^\circ\text{C}$ using a Tray dryer (Reliance Enterprise, Kolkata, India) for 18 h till the weight becomes constant. For ease in utilization the dried peel was powdered using a Grinder (GX7, Bajaj Electricals Ltd, India). The powdered peel for subsequent use was stored in air-tight packets in a refrigerator (-20°C). The RMP powder was mixed at 1, 3 and 5% level with 100 g whole wheat flour for the preparation of desired composite flour.

Bread preparation

For the preparation of control bread, the activation of yeast was done by dissolving yeast

(3 gm) in luke warm (at 37°C) water (20 ml) with previously dissolved sugar and kept for 15 min. In the meanwhile the dried material constituting whole-wheat flour 100 g, sugar 6 g, salt 1.5g and GMS 1.5 g with remaining components like KBrO_3 1ppm were mixed in a bowl. Shortening (4 g), required amount of water (50 ml) and activated yeast were added and final mixing was done manually. The kneading was done for about 10 min for proper formation of gluten chain with appropriate mixing of ingredients in accordance to straight dough method. After this the first proofing at $30 \pm 2^\circ\text{C}$ for 30 min was done by covering the bowl with wet cloth containing the rounded dough resulting in increase in volume. Further, at the juncture of second proofing the pre proofed dough was punched and kneaded slightly to remove the excess CO_2 from it. The worked up dough was transferred into greased moulds and kept for 1hr at $40 \pm 2^\circ\text{C}$ with wet cloth cover. Finally, the bread was baked in rotary oven (CM HS108; Chanmag Bakery Machine Co. Ltd., Taipei, Taiwan) at $215 \pm 2^\circ\text{C}$ for 20-24 min. The baked loaf was cooled at the ambient temperature for 1 h prior to packing in airtight bags. Similarly, the fortified bread samples were prepared by using composite flour (RMP powder at 1%, 3% and 5% level) in other ingredients (without KBrO_3).

Crumb moisture

The moisture content (dry basis) of bread crumb samples was measured by weight difference before and after drying of the samples (initial sample weight about 3 g) in a hot air oven (Reliance Enterprise, Kolkata, India) at 105°C (AACC, 2000).

Baking properties

Bread samples were weighed after cooling and loaf height was measured using a scale. The reading was taken from the top to the bottom of the loaf at three different sides (left, middle, and right). The average of the three readings was recorded. Volumes of the loaves were measured by the seed displacement method. From these, the specific volume (Steffolani *et al.*, 2015), bread density and weight loss (Ho *et al.*, 2013) of the loaf was calculated.

Texture

The textural profile analysis (TPA) of the crumb of bread samples was determined by the texture analyzer (TA.HD Plus Texture Analyzer, USA.). The analysis was done by a 35 mm diameter probe compressing the sliced bread ($40 \text{ mm} \times 40 \text{ mm} \times 30 \text{ mm}$) at a cross-head speed of 0.1–2 mm/s. Hardness, cohesiveness and springiness of the crumb were measured based on the force–time curve (Bouane,

1978).

Loaf colour

The colour was measured using a Hunter Colour Measuring System, ColourFlex 45/0, D65, 10° observer (Hunter Associates Laboratory Inc., Reston, VA, USA). While to analyze the effect of colour change in the crumb, whiteness index is being measured (Ulziijargal *et al.*, 2013) as described in Eq. (1)

$$WI = 100 - ([100 - L]^2 + a^2 + b^2)^{1/2} \quad \dots\dots\dots (1)$$

The brownness index (BI) was calculated (Shittu *et al.*, 2007) to measure colour change in the crust with the addition of RMP powder as described in: Eq. (2)

$$BI = \frac{[100(X-0.31)]}{0.17} \quad \dots\dots\dots (2)$$

$$\text{Where, } X = \frac{(a + 1.75L)}{(5.645 \times L + a - 3.012b)}$$

Dough rheology

The dough samples without yeast with the same formulation of bread were used for studies. Dynamic oscillatory tests were performed in a controlled stress rheometer (Physica MCR 51 Anton Paar, Germany) with parallel plate of 49.986 mm and the measurements monitored with RheoPlus software package (version 2.65). All experiments were carried out at a constant temperature of 25°C. The flour dough was carefully placed between the plates. The upper plate was lowered to a fixed gap of 2.0 mm during loading. The excess of the dough sample outside the top plate edge was trimmed. A water-saturated filter paper to surround the measuring plates was used to minimise drying of the dough sample during measurement. All samples were allowed to rest for 5 min before measurements to allow dough relaxation. Frequency-sweep tests of the dough samples were performed at a constant strain of 1% and angular frequency ranging between 0.1 and 200 ω . Dynamic moduli G' , G'' and $\tan \delta$ (G''/G') were obtained as a function of frequency. G' is the dynamic elastic or storage modulus, related to the material response as a solid, while G'' is the viscous dynamic or loss modulus, related to the material response as a fluid. $\tan \delta$ is related with the overall visco-elastic response: low values of this parameter indicate a more elastic nature (Das *et al.*, 2013).

Extract preparation

Samples (1 g) in 80% aqueous ethanol was

sonicated in a sonicator (Trans-o-sonic/D150-IM, Mumbai) at room temperature (approx. 25°C) for proper mixing of particle and centrifuged in cold (4°C) at 8944×g for 15 min and the supernatant extracted by using a filter paper. The extracts were transferred into plastic tubes, and kept in the refrigerator until analysis.

Total phenolic content

The extract amounting 0.5 ml was dissolved in 1.8 ml of distilled water and 0.2 ml of the Folin-Ciocalteu reagent was added. After 5 min, 2 ml of 7% sodium carbonate was added and the contents mixed thoroughly by further addition of 0.8ml distilled water again. After incubation at room temperature for 90 min, colour developed and the absorbance was observed at 750 nm by UV-Vis spectrophotometer (U 2800; Hitachi, Tokyo, Japan) using gallic acid as a standard. The results were expressed as mg GAE/100 g fresh material (Singleton *et al.*, 1997).

DPPH radical scavenging assay

The free radical scavenging capacity of sample extract was determined using the stable 2, 2-diphenyl-1-picrylhydrazyl radical (DPPH). The decrease in absorbance of the resulting solution was measured spectrophotometrically at 517 nm (Yu *et al.*, 2003). The percentage of inhibition or the percentage of discoloration was calculated as described in. Eq. (3)

$$\% \text{ Inhibition} = \frac{(\text{Abs}_{\text{blank}} - \text{Abs}_{\text{sample}}) \times 100}{\text{Abs}_{\text{blank}}} \quad \dots\dots\dots (3)$$

Ferric reducing antioxidant power (FRAP) assay

FRAP assay was based on the reduction of Fe^{3+} -TPTZ to a blue coloured Fe^{2+} -TPTZ. The FRAP reagent was freshly prepared by mixing acetate buffer (pH 3.6), TPTZ solution (10 mM TPTZ in 40 mM HCl) and 20 mM FeCl_3 solution in 10:1:1 v/v ratio. 100 μl of sample were taken and to it 3ml of the FRAP reagent added. The absorbance was taken at 0 and 30 min (after incubating at 37°C) at 593 nm. The antioxidant potential of the extracts was determined against a standard curve using $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ and expressed as $\mu\text{mol FeSO}_4 \cdot 7\text{H}_2\text{O}/100\text{g}$ fresh material (Benzie and Strain, 1996).

Sensory evaluation

Sensory analysis of raw mango peel fortified bread was done by quantitative descriptive method (QDA). Ten panellists (aged between 20 to 50 yrs), consisting students, research scholar and faculty members of the Dept. of Food Technology and Biochemical Engineering, Jadavpur University,

Kolkata were selected for descriptive profiling test. The panel members were trained to recognise the changes in the sensory quality between the control and the RMP bread. Each training session was of 1 hr and total 3 sessions were conducted. The judges in the panel discussed about the parameter to be considered as sensory attributes of bread. Total 6 sensory attributes consisting porosity, traditional bread aroma, fruity aroma, crumb colour, after taste, hardness and rubbery texture were selected for consideration which invaded the parameters of appearance, aroma, colour, taste and oral texture. For evaluation, approximately 1cm thick slices including the crust and crumb, was presented to assessors. The assessors evaluated the samples on an unstructured 9 cm line scale labelled on the left by 'none' and on the right with the term 'strong'. Ratings were registered on an evaluation form. The samples were kept in a 3-digit coded glass covered with a glass lid. Samples were assessed in triplicates in four sets (Heenan *et al.*, 2008).

Statistical analysis

The experimental design was completely randomized, with three replicates. All data were expressed as mean values \pm SD. All the experimental data were analyzed statistically for analysis of variance (ANOVA) with Microsoft Excel 2007. The scores for QDA was analysed by analysis of variance (two-way ANOVA as factors) applied to (panellist and sample) and their interaction to study the differences between the samples. The comparisons between the mean values were tested using Duncan's new multiple-range test at a level of $p \leq 0.05$.

Results and Discussion

Crumb moisture and baking properties

The effect of addition of RMP powder on crumb moisture and baking properties the bread samples are shown in Table 1. The moisture content increased linearly due to the addition of RMP highest at 5%. The fiber is water retaining, expandable, adsorptive hence the digestive enzyme fails to decompose it (Shyu *et al.*, 2013). By previous studies we can assume that the addition of a fibre containing substance can elevate the moisture content of a baked product (Lim *et al.*, 2011).

Significant variations were observed in various baking quality parameters of peel bread. RMP powder 5% had the lowest weight loss value (7.36%), which ranged from $15.64 \pm 0.60\%$ to $9.04 \pm 0.32\%$ decreasing significantly. This reduction may be attributed due to the addition of fibre which may not be easily

consumed by the yeast during fermentation causing a barrier in fermentable sugar. In coordination with it the loaf height also decreases significantly, with a steep decrement where the lowest loaf height of 4.02 cm was indicated by 5% substitution. This indicates that peel substitution decrease height of the final baked product.

The specific volumes of the bread ranged from $(2.69 \pm 0.05$ to $2.10 \pm 0.02)$ cm^3/g . The highest level addition of peel resulted in the lowest specific volume. In terms of loaf density, bread containing RMP powder, were significantly ($p \leq 0.05$) denser than the control. Possible explanations for the high density of bread may include factors such as water holding capacity of peel containing both soluble and insoluble fibre (Dhillon and Kaur, 2013). Bread with high density was also low in specific volume with less visible air pockets due to the compact crumb structure.

Texture and loaf colour

The textural and colour parameters are shown in Table 2. The hardness of the bread increased linearly as the amount of addition of RMP powder increased showing the maximum value of at 5% substitution. This is in accordance with the addition of other non-wheat based gluten free products where addition of quinoa leafs increased the hardness of the bread crumb (Świeca *et al.*, 2014). The partial substitution of whole wheat bread by a non-proteinous fibre rich powder may lead to change in yeast activity along with stiffening of the air cells leading to a more firm crumb texture. Following the same trend a significant increase in cohesiveness and springiness of the bread fortified with RMP powder was observed in comparison to the bread without peel powder. A greater amount of sugar in mango peels causing the adhesion of the particle might forms a sticky network contributes to the increased cohesiveness of the substituted bread. This observation was also concluded when chestnut flour was added in bread (Dall'Asta *et al.*, 2013). However, some studies also show that the springiness increased at higher percentage of incorporation (Mildner-Szkudlarz *et al.*, 2010).

Colour is an important quality indicator for baked food products which indicates various factors including raw material, baking temperature and time etc. The whiteness index (WI) of the incorporated samples differed significantly from the control. Furthermore the incorporation of 3% and 5% of RMP powder significantly affected the crumb colour imparting a slight green colour due to the addition of raw peel. This attribute was rather attractive from

Table 1. Crumb moisture and baking properties of the bread samples substituted with RMP powder at different levels

Parameters	0% (control)	1%	3%	5%
Moisture (%)	31.63±0.72 ^a	40.16±0.59 ^b	43.47±0.35 ^c	46.49±0.63 ^d
Weight loss (%)	15.64±0.60 ^d	12.01±0.22 ^c	9.04±0.32 ^b	7.36±0.22 ^a
Loaf height (cm)	5.54±0.4 ^c	5.03±0.62 ^c	4.6±0.33 ^b	4.0±0.2 ^a
Specific volume (cm ³ /gm)	2.69±0.05 ^d	2.43±0.36 ^c	2.23±0.15 ^b	2.10±0.02 ^a
Density (g/cm ³)	0.37±0.02 ^a	0.41±0.02 ^b	0.44±0.01 ^c	0.48±0.02 ^d

Each value is expressed as mean ± SD (n = 3). Mean with different letters within a row is significantly different (p<0.05).

Table 2. Textural and colour parameters of the bread samples substituted with RMP powder at different levels

Parameters	0% (control)	1%	3%	5%
Hardness (N)	2.6±0.11 ^a	3.2±0.14 ^b	3.7±0.16 ^c	4.0±0.19 ^c
Cohesiveness	0.26±0.01 ^a	0.30±0.01 ^b	0.60±0.03 ^c	0.72±0.02 ^d
Springiness	0.30±0.01 ^a	0.36±0.01 ^b	0.40±0.02 ^c	0.55±0.02 ^d
WI	57.57±1.18 ^d	54.67±0.93 ^c	52.47±1.3 ^b	50.67±0.96 ^a
BI	79.71±5.2 ^a	87.25±3.6 ^a	90.37±1.28 ^b	121.47±17.12 ^c

Each value is expressed as mean ± SD (n = 3). Mean with different letters within a row is significantly different (p<0.05). WI (whiteness index), BI (brownness index)

consumer viewpoint and has also been indicated in anka enriched bread (Tseng *et al.*, 2010). This result is also in compliance with the study (Wu and Shiau, 2013) of pineapple peel where the incorporation of peel also significantly lowered the WI due to addition, imparting its own colour.

The crust of the breads containing RMP powder changed from brown to greenish brown. Colour analysis of the crust indicated that samples with the addition of RMP powder had darker crust, this can be ascribed to the darkening effects of Maillard and caramelization browning due its high sugar content, as previously hypothesized (Shittu *et al.*, 2008) which could be attributed by the higher brownness index (BI) also studied earlier by (Das *et al.*, 2012). However, the increase in brownness was significant at (p≥0.05), even at higher substitution level as the green colour of peel was suppressed by the dominant brown colour.

Dough rheology

A correct description of the changes in dough behaviour is necessary to maintain handling and machinability in industrialized production. The results of the frequency sweep test carried out on dough samples with RMP powder substituted and on the control samples are shown in Table 3. In all dough samples the storage modulus (G') was higher than the loss modulus (G''), indicating that the dough had a solid, elastic-like behaviour. Dough made with 5% of RMP powder showed statistically significant differences in terms of damping factor (tan δ) and complex viscosity (η*) compared to the other samples. Generally higher G' and lower tan δ indicate a more elastic and solid-like material.

The addition of RMP powder caused an increase of the G' twice as compared to G'', indicates increased degree of elasticity of the dough. The decrease of tan δ implies that dough exhibits a different degree of

Table 3. Rheological parameters of the bread samples substituted with RMP powder at different levels

Parameters	0% (control)	1%	3%	5%
Storage Modulus (G') [Pa]	114540±3690 ^a	133397±8173 ^b	144949±2264 ^c	155686±5418 ^d
Loss Modulus (G'') [Pa]	58525±761 ^a	61138±1619 ^b	67270±2857 ^c	74217±1418 ^d
Complex Viscosity (η^*) [Pa.s]	4519.97±220 ^a	6182.727±108 ^b	7183.93±59 ^c	7549.96±377 ^d
Damping factor ($\tan \delta$)	0.507±0.02 ^c	0.479±0.01 ^b	0.463±0.02 ^a	0.455±0.01 ^a

Each value is expressed as mean \pm SD (n = 3). Mean with different letters within a row is significantly different ($p \leq 0.05$).

Table 4. Total phenols and scavenging properties of the bread samples substituted with RMP powder at different levels

Parameters	0% (control)	1%	3%	5%
TPC (mg GAE/100gm)	227.66±7.5 ^a	443±11.0 ^b	510±12.5 ^c	794±8.32 ^d
FRAP ($\mu\text{M FeSO}_4 \cdot 7\text{H}_2\text{O} / 100\text{g}$)	162.87±4.38 ^a	476.13±11.08 ^b	1360.75±39.55 ^c	2232.31±47 ^d
DPPH (% inhibition)	21.51±0.45 ^a	33.61±0.78 ^b	61.19±1.90 ^c	68.54±2.15 ^d

Each value is expressed as mean \pm SD (n = 3). Mean with different letters within a row is significantly different ($p \leq 0.05$).

cross-linking and that structural changes took place with changing RMP powder level. It is known that an increase in the cross-linking of a polymer system causes the G' to increase and the loss tangent to decrease (Watersa *et al.*, 2011). Peel's addition, at the microstructural level, gives rise to a network with a higher density of cross-links. The increase in η^* indicated increased resistance to deformation. This is likely due to interactions between the fibre structure and the wheat proteins.

Antioxidant activity

Total phenolic content is an important parameter to study as it determines the polyphenolic content of the sample which is interrelated with the antioxidant activity of the sample. As shown in Table 4, the amount of phenolics increased proportionately to the increase of RMP powder level in the fortified breads. This, significant increase is in accordance with many tropical fruit (Moo-Huchin, 2015).

Table 4 shows the scavenging properties including DPPH and FRAP of the bread samples. It was noted that the values of DPPH and FRAP increased significantly indicating the scavenging properties of the fortified products. The values of FRAP ($\mu\text{M FeSO}_4 \cdot 7\text{H}_2\text{O} / 100\text{g}$) increased compared to the bread with no RMP powder, giving the highest value of 2232.31±47 $\mu\text{M FeSO}_4 \cdot 7\text{H}_2\text{O} / 100\text{g}$ at 5% fortification. Whereas, the linear increment was also termed in DPPH (% inhibition) showing about three times leap from 0% to 5% RMP powder resulting a range of 21.51% to 68.54% inhibition.

The increase in total phenolic content and free radical scavenging activity may be attributed due to the presence of a rich variety of bioactive components such as polyphenols, dietary fibre and vitamin C etc (Ajila *et al.*, 2007) in mango peels. The incorporation of mango peel powder in biscuit (Ajila *et al.*, 2008) and macaroni (Ajila *et al.*, 2010) have also shown

Table 5. Sensorial attributes and scores of the bread samples substituted with RMP powder at different levels

Sensory Descriptors	Samples				Std dev
	0% (control)	1%	3%	5%	
Porosity	7.59 ^d	7.33 ^c	7.11 ^b	6.65 ^a	0.40
Traditional bread aroma	8.43 ^d	8.02 ^c	7.69 ^b	7.07 ^a	0.57
Fruity aroma	2.17 ^a	3.93 ^b	4.63 ^c	5.24 ^d	1.33
After taste	2.59 ^a	2.99 ^b	4.03 ^c	5.17 ^d	1.15
Crumb Colour	6.71 ^a	7.14 ^b	7.48 ^c	8.01 ^d	0.55
Hardness	4.27 ^a	5.03 ^b	5.47 ^c	6.46 ^d	0.91
Rubbery	5.68 ^a	6.15 ^b	6.78 ^c	7.07 ^d	0.62

Mean with different letters within a column is significantly different ($p \leq 0.05$).

increased phenolic and scavenging property with increased in amount of addition of peel.

Sensory evaluation

The sensory attributes, with their assessment scores by the evaluators for each sample are shown in Table 5. All the samples were statistically different ($p \leq 0.05$) on the mentioned sensory attributes of the RMP fortified breads.

Porosity which represents the appearance of bread decreased significantly as the amount of fortification increased. The lower scores for incorporated sample compared to the control may be due to the compact air cells in crumb caused by fibre addition. While observing the aroma descriptors, an inverse relationship between traditional bread aroma and fruity aroma was seen. As the amount of addition of level of peel increased the aromatic compounds masked the traditional yeasty aroma. However, the overall low scores of after taste which may include attribute of bitterness was low. As the amount of peel incorporation increased simultaneously the level of sugar causing a darker colour due to Millard reaction also increases leading to more intense crumb colour showing highest value at 5% RMP substitution. Considering the oral texture parameters both hardness and rubbery texture incremented significantly on addition of peel powder in bread. The values of both the descriptors for RMP bread were significantly larger when compared to bread with no level of peel also studied earlier, highest value at 5% RMP (Shin *et al.*, 2013).

Conclusion

Addition of RMP powder at different percentage (from 0% to 5%) changes dough viscoelasticity, physical, antioxidant and sensory properties of bread. The dough with the highest amount of

RMP powder (5%) showed the highest value of viscoelasticity evaluated by fundamental rheological measurements. This sample showed, after the lowest WI values regarding the crumb colour and the highest BI value for crust colour along with slight decrement in baking properties with greater retention of moisture. Incorporation of RMP powder in formulation markedly increased the total phenolics content and the radical scavenging activity of bread extracts. However, RMP incorporation at highest level increased the hardness, rubbery texture with significant decrease in crumb porosity and traditional bread aroma. Thus, optimum level of addition of RMP is an important factor to be considered. This leads to a conclusion that RMP powder at 3% can be added in bread to increase the antioxidant activity of whole wheat bread without much affecting the sensory attributes

Acknowledgement

This research work is financially supported by the Basic Scientific Research (BSR) fellowship for meritorious student programme under University Grants Commission (UGC), India. The authors report no conflict of interest. The authors alone are responsible for the content and writing of the paper.

References

- Ajila, C. M., Aalami, M., Leelavathi, K. and Prasada Rao, U. J. S. 2010. Mango peel powder: A potential source of antioxidant and dietary fiber in macaroni preparations. *Innovative Food Science and Emerging Technologies* 11(1): 219–224.
- Ajila, C. M., Leelavathi, K. and Prasada Rao, U. J. S. 2008. Improvement of dietary fiber content and antioxidant properties in soft dough biscuits with the incorporation of mango peel powder. *Journal of Cereal Science*

- 48(2): 319-326.
- Ajila, C. M., Bhat, S. G. and Prasada Rao, U. J. S. 2007. Valuable components of raw and ripe peels from two Indian mango varieties. *Food Chemistry* 102(4): 1006–1011.
- AACC. 2000. Approved Method of the AACC. 10th Ed. American Association of Cereal Chemists, St. Paul, MN, USA.
- Bandyopadhyay, K., Chakraborty, C. and Bhattacharyya, S. 2014. Fortification of Mango Peel and Kernel Powder in Cookies Formulation. *Journal of Academia and Industrial Research* 2(12): 661-664.
- Benzie, I. F. F. and Strain, J. J. 1996. The ferric reducing ability of plasma (FRAP) as a measure of “antioxidant power”: the FRAP assay. *Analytical biochemistry* 239(1): 70–76.
- Berardini, N., Knödler, M., Schieber, A. and Carle, R. 2005. Utilization of mango peels as a source of pectin and polyphenolics. *Innovative Food Science and Emerging Technologies* 6(4): 442-452.
- Bhattacharjee, A., Ghosh, S. K., Neogi, K., Aich, A., Willard, B., Kinter, M., Sen, S. K., Ghosh, D. and Ghosh, S. 2011. Deposition of stearate-oleate rich seed fat in *Mangifera indica* is mediated by a FatA type acyl-ACP thioesterase. *Phytochemistry* 72(2-3): 166–177.
- Bouane, M. C. 1978. Texture profile analysis. *Food Technology* 32(7): 62-66.
- Dall’Asta, C., Cirlini, M., Morini, E., Rinaldi, M., Ganino, T. and Chiavaro, E. 2013. Effect of chestnut flour supplementation on physico-chemical properties and volatiles in bread making. *LWT - Food Science and Technology* 53(1): 233-239.
- Das, L., Raychaudhuri, U. and Chakraborty, R. 2013. Role of hydrocolloids in improving the physical and textural characteristics of fennel bread. *International Food Research Journal* 20(5): 2253-2259.
- Das, L., Raychaudhuri, U. and Chakraborty, R. 2012. Effect of baking conditions on the physical properties of herbal bread using rsm. *International Journal of Food, Agriculture and Veterinary Sciences* 2(2): 106-114.
- Dhillon, G. K. and Kaur, A. 2013. Quality evaluation of bread incorporated with different levels of cinnamon powder. *International Journal of Food Science, Nutrition and Dietetics (IJFS)* II(7): 1-6.
- FAOSTAT. 2007. FAO statistics, Food and Agriculture Organization of the United Nations. Database. <http://faostat.fao.org/> [Accessed Date: September 9, 2014].
- Heenan, S. P., Dufour, J. P., Hamid, N., Harvey, W. and Delahunty, C. M. 2008. The sensory quality of fresh bread: Descriptive attributes and consumer perceptions. *Food Research International* 41(10): 989–997.
- Ho, L. H., Aziz, N. A. A. and Azahari, B. 2013. Physico-chemical characteristics and sensory evaluation of wheat bread partially substituted with banana (*Musa acuminata* X *balbisiana* cv. *Awak*) pseudo-stem flour. *Food Chemistry* 139(14): 532–539.
- Jamsazzadeh Kermani, Z., Shpigelman, A., Pham, H. T. T., Van Loey, A. M. and Hendrickx, M. E. 2015. Functional properties of citric acid extracted mango peel pectin as related to its chemical structure. *Food Hydrocolloids* 44: 424-434.
- Kermani ZJ, Shpigelman A, Pham HTT, Loey AMV, Hendrickx ME. (2015). Functional properties of citric acid extracted mango peel pectin as related to its chemical structure. *J. Food Hydrocolloids* 44: 424-434.
- Kim, H., Moon, J. Y., Kim, H., Lee, D. S., Cho, M., Choi, H. K., Kim, Y. S., Mosaddik, A. and Cho, S. K. 2010. Antioxidant and antiproliferative activities of mango (*Mangifera indica* L.) flesh and peel. *Journal of Food Chemistry* 121(2): 429–436.
- Lim, H. S., Park, S. H., Ghafoor, K., Hwang, S. Y., and Park, J. 2011. Quality and antioxidant properties of bread containing turmeric (*Curcuma longa* L.) cultivated in South Korea. *Food Chemistry* 124(4): 1577–1582.
- Mildner-Szkudlarz, S., Zawirska-Wojtasiak, R., Szwengiel, A. and Pacyński, M. 2010. Use of grape by-product as a source of dietary fibre and phenolic compounds in sourdough mixed rye bread. *International Journal of Food Science and Technology* 46(7): 1485–149.
- Moo-Huchin, V. M., Moo-Huchin, M. I., Estrada-León, R. J., Cuevas-Glory, L., Estrada-Mota, I. A., Ortiz-Vázquez, E., Betancur-Ancona, D. and Sauri-Duch, E. 2015. Antioxidant compounds, antioxidant activity and phenolic content in peel from three tropical fruits from Yucatan, Mexico. *Food Chemistry* 166: 17–22.
- Ramirez-Maganda, J., Blancas-Benitez, F. J., Zamora-Gasga, V. M, García-Magaña, M. D. L, Bello-Pérez, L. A., Tovar, J. and Sáyago-Ayerdi, S. G. 2015. Nutritional properties and phenolic content of a bakery product substituted with a mango (*Mangifera indica*) ‘Araulfo’ processing by-product. *Food Research International* 73: 117-123.
- Ravani, A. and Joshi, D. C. 2013. Mango and its by product utilization—a review. *Trends in Post Harvest Technology* 1(1): 55-67.
- Shin, D. J., Kim, W. and Kim, Y. 2013. Physicochemical and sensory properties of soy bread made with germinated, steamed, and roasted soy flour. *Food Chemistry* 141(1): 517–523.
- Shittu, T. A., Dixon, A., Awonorin, S. O., Sanni, L. O. and Maziya-Dixon, B. 2008. Bread from composite cassava–wheat flour. II: Effect of cassava genotype and nitrogen fertilizer on bread quality. *Food Research International* 41(6): 569–578.
- Shittu, T. A., Raji, A. O. and Sanni, L. O. 2007. Bread from composite cassava-wheat flour: I. Effect of baking time and temperature on some physical properties of bread loaf. *Food Research International* 40(2): 280–290.
- Shyu, Y. S., Lu, T. C. and Lin, C. C. 2013. Functional analysis of unfermented and fermented citrus peels and physical properties of citrus peel-added doughs for bread making. *Journal of Food Science and Technology* 51(12): 3803-3811.
- Singleton, V., Orthofer, R. and Lamuela-Raventos, R. 1997. Analysis of total phenols and other oxidation

- substrates and antioxidants by means of Folin–Ciocalteu reagent. *Methods in Enzymology* 299: 152–178.
- Steffolani, E., Martinez, M. M., León, A. E. and Gomez, M. 2015. Effect of pre-hydration of chia (*Salvia hispanica* L.), seeds and flour on the quality of wheat flour breads. *LWT - Food Science and Technology* 61(2): 401-406.
- Świeca, M., Sęczyk, Ł., Gawlik-Dziki, U. and Dziki, D. 2014. Bread enriched with quinoa leaves – The influence of protein–phenolics interactions on the nutritional and antioxidant quality. *Food Chemistry* 162: 54–62.
- Tseng, Y. H., Yang, J. H., Chen, C.H. and Mau, J. L. 2011. Quality and antioxidant properties of anka enriched bread. *Journal of Food Processing and Preservation* 35(4): 518–523.
- Tunchaiyaphum, S., Eshtiaghi, M. N. and Yoswathana, N. 2013. Extraction of bioactive compounds from mango peels using green technology. *International Journal of Chemical Engineering and Applications* 4(4): 194-198.
- Ulziijargal, E., Yang, J. H., Lin, L.Y., Chen, C. P. and Mau, J. L. 2013. Quality of bread supplemented with mushroom mycelia. *Food Chemistry* 138(1): 70–76.
- Varakumar, S., Naresh, K. and Reddy, O. V. S. 2012. Preparation of Mango (*Mangifera indica* L.) Wine Using a New Yeast-Mango-Peel Immobilised Biocatalyst System. *Czech Journal of Food Sciences* 30(6): 557–566.
- Wu, M. Y. and Shiau, S. Y. 2013. Effect of the amount and particle size of pineapple Peel fiber on dough rheology and steamed Bread quality. *Journal of Food Processing and Preservation* Doi:10.1111/jfpp.12260.
- Waters, D. M., Ryan, L. a M., Murray, P. G., Arendt, E. K. and Tuohy, M. G. 2011. Characterisation of a *Talaromyces emersonii* thermostable enzyme cocktail with applications in wheat dough rheology. *Enzyme and Microbial Tech* 49(2): 229– 236.
- Yu, L., Perret, J., Harris, M., Wilson, J. and Haley, S. 2003. Antioxidant properties of bran extracts from Akron wheat grown at different locations. *Journal of Agriculture and Food Chemistry* 51(6): 1566-1570.

A Study to Create Awareness towards Tropical Fruit Waste as a Source of Natural Antioxidant

Debasmita Pathak¹, Utpal Raychaudhuri² and Runu Chakraborty^{3*}
Department of Food Technology and Biochemical Engineering, Jadavpur University,
Kolkata 700032, India

¹debasmita.pathak@gmail.com, ²utpal31@hotmail.com, ^{3*}Corresponding author: crunu@hotmail.com

Abstract

Raw Mango Peel (RMP) is a rich source of many bioactive components including polyphenols, flavonoids, carotenoids, vitamin C and vitamin E etc. It is generated as waste during processing of mango pulp for various products both in household and industrial level.

In this technique of mango processing, 13-15% consists of the peel. In the present study, oven dried raw mango peel powder (RMPP) was prepared to investigate the physical, antioxidant and sensory properties. The effect of oven drying on RMPP showed better lightness and yellowness. The content of polyphenols, flavonoids was remarkably high. Free radical scavenging activity was also studied by DPPH radical scavenging assay to investigate the antioxidant quality. Moreover, by sensory evaluation this peel powder was found to have good overall acceptability with acceptable scores in colour and aroma parameters. Then RMPP is incorporated in traditional roti at 4% level. RMPP enriched roti showed high antioxidant property and an acceptable sensory profile than control sample. Thus results suggest that the use of RMPP is suitable for enhancing the nutritional quality when incorporated in food substances.

Keywords: Raw mango peel, colour, antioxidant properties, sensory properties, enriched roti.

Introduction

Mango (*Mangifera indica* L.) is one of the most important tropical fruit which grows abundantly in 83 different countries across the world with the annual production of 25 MMT and is being consumed about 50% of the total tropical fruit [1]. Raw mangoes are used for preparing various traditional and commercial products like raw slices in brine, amchur, pickle, murabba, chutney, panhe (sherbet) and squash etc. To prepare these above stated products, the peel of mango (which is about 15-20%) is generated as waste [2]. Mango peels are good sources of polyphenols, carotenoids, dietary fibre (pectin) and other bioactive compounds [3]. The polyphenol content is found significantly higher in raw peels compared to ripe peel while the carotenoids and vitamin E are found significantly higher in ripe peels [4]. Gallic acid, protocatechuic acid, gentisic acid and syringic acid are the phenolic acids identified in phenolic fractions of raw and ripe mango peel extracts. The addition of natural antioxidants in foods and biological systems has received considerable interest because of their presumed safety and potential nutritional and therapeutic effects.

Proximate composition of raw mango peel is around - moisture (66%), carbohydrate (22.8%), protein (1.76%), fat (2.16%), dietary fibre (3.28%) and ash (1.16%) [5]. Raw mango peel can be used as an ingredient in processed food products such as bakery products, breakfast cereals, pasta products and beverages. Also we can find ripe mango peels used in muffins, biscuits, macaroni and wine preparation.

Thus the main objective of this work is to improve antioxidant property of raw mango peel through the drying process and its utilization in daily food.

Materials and Methods:

Raw Materials: Raw mangoes, wheat flour, salt were purchased from local market of Jadavpur, Kolkata, India.

Chemicals: Gallic acid (SD Fine-Chem Ltd., Mumbai, India), DPPH (Sigma-Aldrich, St. Louis, MO, USA), Folin-Ciocalteu reagent, sodium carbonate, ethanol (Merck Specialities Pvt., Ltd., Mumbai, India), sodium hydroxide (Himedia Laboratories Pvt., Ltd., Mumbai, India), sodium nitrite and aluminium chloride (LOBA Chemie) were used in the experiment.

Raw mango peel powder preparation: Raw mango peel (RMP) was blanched by dipping in hot water followed by cold water. Mango peels were cut into small strips before drying. Samples were dried in a tray dryer (ICT, Kolkata, India) at 60°C upto constant weight. The dried samples were then ground in a common grinder (Prestige Stylo Mixer Grinder, Prestige, India) and packaged in freezer proof air tight container. Thus raw mango peel powder (RMPP) was obtained.

Traditional roti preparation: Traditional roti is prepared with wheat flour, salt and water. This is denoted as control sample. In raw mango peel powder (RMPP) enriched roti 4% RMPP is added during dough formation.

Percentage weight ratio: Weight of different fruit parts was taken on a citizen (model MP-300) electronic balance to calculate the Percentage weight ratio (PWC).

Color: The color of the samples were measured using the Hunter Lab color measurement system, ColorFlex 45/0, D65, 10° observer (Hunter Associates Laboratory Inc., Reston, VA, USA) according to the CIELAB system of color measurement as L*-whiteness/darkness (100-0), a*-redness/greenness (+ to -) and b*-yellowness/blueness (+ to -) [6].

Total phenolic content: The total phenolic content (TPC) was determined according to the Sharma and Gujral 2010 [7]. After incubation at room temperature for 90 mins, the absorbance of the mixture was observed at 750 nm. The results were expressed as mg of gallic acid equivalents (GAE) per 1 gram of sample.

Total flavonoid content: The total flavonoid content of sample was determined with slight modifications of Xu and Chang 2007 [8]. Total flavonoid contents were expressed as milligrams of catechin equivalents (CAE) per 1 gram of defatted sample.

Radical DPPH scavenging capacity: The free radical scavenging capacity of sample extracts was determined using the stable 2, 2-diphenyl-1-picrylhydrazyl radical (DPPH). The reaction was monitored by reading absorbance at 517 nm according to Yu et al. 2003 [9].

Sensory Analysis: Samples were coded and presented to 10 panel members for sensory scoring. The panel members, who were familiar with sensory analysis techniques, were postgraduate students and research scholars of the Department of Food Technology and Biochemical Engineering (Jadavpur University, India). Samples were scored for appearance, taste, color, aroma and overall acceptability according to numerical scoring system. The model used in this analysis was an acceptance test on the hedonic scale, with values ranging from "1" (extremely disliked) to "9" (extremely liked). Mean and standard deviation were individually calculated for scores obtained for all quality attributes of each product.

Results and Discussions:

Raw Mango Peel & Raw Mango Peel Powder: As a physical property peel to fruit weight ratio (table 1) was 13.37%. In color profile (table 1) whiteness and yellowness increased in RMPP from (30.67±0.83 to 62.83±0.23) and (16.45±0.29 to 26.56±0.41) respectively. But greenness decreased in RMPP from (-7.81±0.25 to -0.76±0.19) due to drying temperature has an effect on coloring pigment.

Table 1: PHYSICAL PROPERTIES OF RAW MANGO PEEL

Parameters	RMP	RMPP
Peel to fruit weight ratio (%)	13.37±1.04	-----
L*	30.67±0.83	62.83±0.23
a*	-7.81±0.25	-0.76±0.19
b*	16.45±0.29	26.56±0.41

Values are given as means of 3 samples (n=3) ± SD. L*-whiteness/darkness (100-0); a*-redness/greenness (+ to -); b*yellowness/blueness (+ to -).

Antioxidant profile (table 2) of RMPP was much better in qualitative and quantitative aspect. TPC and flavonoid value of RMPP is 8.26±0.41 mg GAE/g and 1.16±0.06

mg CE/g respectively where as in RMP it is 1.05±0.05 mg GAE/g and 0.53±0.03 mg CE/g respectively. DPPH inhibition percentage is more than double in RMPP than RMP.

Table 2: ANTIOXIDANT PROPERTIES OF RAW MANGO PEEL

Parameters	RMP	RMPP
TPC (mg GAE/g)	1.05±0.05	8.26±0.41
Flavonoid (mg CE/g)	0.53±0.03	1.157±0.06
DPPH (% inhibition)	13.97±0.7	50.87±0.79

Values are given as means of 3 samples (n=3) ± SD. TPC= Total phenolic content, DPPH= 2, 2-diphenyl-1-picrylhydrazyl radical

As a whole, organoleptic acceptability is an important parameter for all processed food. RMP scored higher in all aspect but RMPP also scored above 6 and close to RMP in all aspect. So from this point it could be established that RMPP was also acceptable like RMP. According to this comparative study we can use RMPP in food as a source of natural antioxidant.

Table 3: SENSORY PROPERTIES OF RAW MANGO PEEL

Parameters	RMP	RMPP
Color	8.5±0.5	7.76±0.25
Aroma	8.6±0.29	7.3±0.25
Overall acceptability	8.3±0.3	7.6±0.4

Values are given as means of 3 samples (n=3) ± SD.

Traditional Roti: As a color profile (table 4) whiteness and yellowness decreased and redness increased in 4% RMPP enriched roti than control. This is due to presence of RMPP. DPPH inhibition percentage (table 4) was almost absent in control sample but in 4% RMPP enriched roti it was 28.55%. During processing antioxidant property still present which means this natural antioxidant is acceptable for processed food.

Table 4: COLOR & ANTIOXIDANT PROPERTIES OF RMPP ENRICHED ROTI

Parameters	CONTROL	4% RMPP
L*	78.95±0.76	65.44±0.70
a*	1.80±0.31	4.48±0.15
b*	15.43±0.19	14.31±0.07
DPPH (%)	0.54±0.03	28.55±0.52

Values are given as means of 3 samples (n=3) ± SD.

In sensory properties (table 5) all values were higher in control sample but difference with 4 % RMPP enriched roti it was very close and also equal or higher than 7. If we consider overall acceptability then in control it was 7.96 and in 4% RMPP it was 7.5. So, 4% RMPP enriched roti was also acceptable.

Table 5: SENSORY PROPERTIES OF RMPP ENRICHED ROTI

Parameters	CONTROL	4%RMPP
Color	7.8±0.29	7±0.3
Aroma	8.1±0.29	7.1±0.17
Appearance	8.17±0.28	7.5±0.46
Taste	7.6±0.58	7.6±0.42
Overall acceptability	7.96±0.05	7.5±0.45

Values are given as means of 3 samples (n=3) ± SD.



Conclusion:

Through the drying process, we can prepare RMPP which shows better antioxidant property than RMP. Also 4% RMPP improves the antioxidant property of control sample (roti), with acceptable physical and sensory properties. So, we can conclude that by using drying process we can convert that raw mango peel into powder and can include it in our daily diet.

References

1. Mary Antoni Rosalin, A. Vinayagamoorthy (2014) Growth analysis of mangoes in India. International Journal of Business and Administration Research Review Vol.1, 5: 157-164.
2. Tunchaiyaphum S., Eshtiaghi M.N, Yoswathana N. (2013) Extraction of Bioactive Compounds from Mango Peels using Green Technology. Int. J. Chemical Eng. And Applications 4(4): 194-198.
3. Berardini N, Kno` dler M, Schieber A, Carle R. (2005) Utilization of mango peels as a source of pectin and polyphenolics. Innovative food Sc & Emerging Tech 6(4): 442-452.
4. C.M. Ajila, L. Jaganmohan Rao, U.J.S. Prasada Rao (2010) Characterization of bioactive compounds from raw and ripe *Mangifera indica* L. peel extracts. Food and Chemical Toxicology 48: 3406-3411.
5. C.M. Ajila, S.G. Bhat, U.J.S. Prasada Rao (2007) Valuable components of raw and ripe peels from two Indian mango varieties. Food Chemistry 102: 1006-1011.
6. Nisha P, Ananthanarayan Laxmi, Singhal Rekha S (2005) Effect of stabilizers on stabilization of idli (traditional south Indian food) batter during storage. Food Hydrocolloid 19: 179-186.
7. Sharma P, Gujral HS (2010) Antioxidant and polyphenol oxidase activity of germinated barley and its milling fractions. Food Chem 120: 673-678.
8. Xu BJ, Chang SKC (2007) A comparative study on phenolic profiles and antioxidant activities of legumes as affected by extraction solvents. J. Food Sci 72: S159-S166.
9. Yu L, Perret J, Harris M, Wilson J, & Haley S (2003) Antioxidant properties of bran extracts from Akron wheat grown at different locations. J. Agric. Food Chem 51: 1566-1570.



Certificate of Participation

This is to certify that **Debasmita Pathak**, Jadavpur University has participated and presented an oral entitled “*Utilization of raw and ripe mango peel as a quality improver in gluten free bread making*” in the “5th India Biodiversity Meet 2018 (International Conference)”, held at Indian Statistical Institute, Kolkata, from 15th -17th March, 2018.

Abhishek Mukherjee
Organising Secretary

Sabyasachi Bhattacharya
Head, AERU, ISI

5th
INDIA
BIODIVERSITY
MEET 2018
International Conference
15 -17 MARCH, 2018



International Conference
**Emerging Technologies in
Agricultural and Food Engineering**



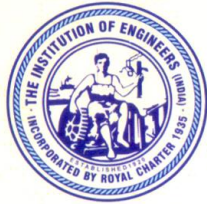
27 – 30 December 2016

Certificate of Presentation

This is to certify that Debasmita Pathak and Payel Dasgupta
of Jadavpur University, Kolkata
has given an ORAL presentation in the Technical Session AEE/NREM/PMS/FBPE at ETAEE 2016
organized by Agricultural and Food Engineering Department, Indian Institute of Technology Kharagpur,
India.

Professor V. K. Tewari
Chairman

Professor H. N. Mishra
Organizing Secretary



Agricultural Engineering Division West Bengal State Centre

The Institution of Engineers (India)

(Established 1920, Incorporated by Royal Charter 1935)

8, Gokhale Road, Kolkata - 700 020

This is to certify that

Shri/Ms./Dr.

Debasmita Pathak

Participated as a registered delegate and presented a paper in the All India Seminar on "Post Harvest Management of Fruits and Vegetables" held at Kolkata during 01-02 June, 2016, organised by the Agricultural Engineering Division, West Bengal State Centre, The Institution of Engineers (India).

Debari Nag
Dr. D. Nag

Organising Secretary
All India Seminar

Kashmir Lal Mallik
Er. Kashmir Lal Mallik

Honorary Secretary
WBSC, IE(I)

Sisir Kumar Banerjee
Er. Sisir Kumar Banerjee

Chairman
WBSC, IE(I)

**UGC Sponsored Two Days National Seminar
on**

“Nutritional Deprivation in the Midst of Plenty” - An Attempt to Create Awareness

Date : 22nd & 23rd September, 2015



Venue: Raidighi College, University of Calcutta, 24 PGS (South)-743383



Organized by:

Department of Food & Nutrition, Raidighi College

: In Collaboration With:

Department of Botany, Raidighi College

Department of Microbiology, Sarsuna College

Certificate of Participation

This is to certify that Shri/ Smt./ Dr./ Prof. *Debasmita Pathak*
of *Jadavpur* University..... has participated as Speaker/ Chairperson/
Paper presenter /Poster presenter/ Delegate/ Rapporteur in the UGC sponsored National Seminar 2015 held at Raidighi
College, University of Calcutta. He/ She Delivered the topic entitled *A Study to Create Awareness*
towards Tropical Fruit Waste as a Source of Natural Antioxidant.

Rukshana Irani
Convenor/Organizing Secretary
Raidighi College

Dr. Sasabindu Jana
Principal
Raidighi College



International Conference
on
Recent Advances in Food Processing and Biotechnology

Centre of Food Science and Technology, Institute of Agricultural Sciences
Banaras Hindu University, Varanasi

5-6 April, 2016

Certificate of Participation

This is to certify that Prof. / Dr. / Mr. / Ms. Aebasmita Pathak has
participated in International Conference on **Recent Advances in Food Processing and Biotechnology**
as a part of **Centenary Year Celebration** of Banaras Hindu University.

He / She has presented a paper on Optimization of Ripe Mango peel fortification level in Bread using Response Surface Methodology
in Poster / Oral Presentation / Young Scientist Award session.

Abhishek Dutt Tripathi

Organizing Secretary

Anil Kumar Chauhan

Convener

Ravi Pratap Singh

Director

NATIONAL SCIENCE MEET - 2016



Organised by

Hiralal Mazumdar Memorial College for Women,
Dakshineswar, Kolkata
in collaboration with
West Bengal State University,
Barasat, North 24 parganas



This is to certify that

Prof./Dr./Mr./Ms. *DEBASMITA PATHAK*

*has participated as Quiz Competitor/ Science Model Demonstrator/ Delegate/
Poster Presenter/ Resource Person/ Session Chairperson
in National Science Meet held on 2nd and 3rd March, 2016*

Dr. Soma Ghosh
President, National Science Meet
Principal
Hiralal Mazumdar Memorial College
For Women
Dakshineswar, Kolkata-700 035

Prof. Basab Chaudhuri
Vice Chancellor, West Bengal State University

Workshop on
Industrial Experiments for Engineering Students: WIEEST-2015/1
February 17-19, 2016

Certificate of Participation

This is to certify that Ms. Debasmita Pathak, Research Fellow, Food Technology & Biochemical Engineering, Jadavpur University, Kolkata has actively participated in the above workshop organized by SQC & OR Unit of Indian Statistical Institute, Kolkata.

Prasun Das

Prasun Das
Head, SQC & OR Unit, Kolkata

February 19, 2016

Sugata Adhikari

Sugata Adhikari
Organizing Secretary