Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2018; 7(3): 1336-1340
Received: 07-03-2018
Accepted: 11-04-2018

Sukirti Tiwari
Food Processing Laboratory, University School of Biotechnology, GGS Indraprastha University, Sector 16C, Dwarka, Delhi, India

Abha Sharma
Food Processing Laboratory, University School of Biotechnology, GGS Indraprastha University, Sector 16C, Dwarka, Delhi, India

Rajinder Kumar Gupta
Department of Applied Chemistry, Delhi Technological University, Shahbad Daulatpur, Bawana Road, Delhi, India

Correspondence
Rajinder Kumar Gupta
Department of Applied Chemistry, Delhi Technological University, Shahbad Daulatpur, Bawana Road, Delhi, India
Email: rkg67ap@yahoo.com

Formulation of cereal based spread using oats fermented with Baker's Yeast

Sukirti Tiwari, Abha Sharma and Rajinder Kumar Gupta

Abstract
Whole grain oats were fermented with Baker's yeast to obtain tastier, cholesterol free, low fat, cereal based spread. The phytochemical analysis showed an increase in phenolic content and antioxidant potential by 61.6% and 29.82%, respectively in the fermented oats as compared to the unfermented ones. The sensory evaluation of spread made from fermented oats gave a higher acceptance (8.67) as compared to the control spread made from unfermented oats (4.55). Furthermore, it was also observed that oats can act as fat replacers in products like spreads and marmalades as oat based spread was formulated by using only 10% fat, while on the other hand, the conventional fat based spreads are made by using 30% fat. The shelf life of fermented oat spread was found to be 21 days at refrigeration temperature with no phase separation.

Keywords: oats, fermentation, yeast, cereal spread, fat replacement

1. Introduction
Whole grain constitutes the major source of dietary protein, vitamins, minerals, fibers, carbohydrates (Blandino *et al*. 2002) [5]. The observational and clinical studies indicate that the regular consumption of whole grains can reduce the risk of various chronic diseases (Blandino and Klinken, 2014).

Among the various food grains, oats has been emerging out as excellent food grains with high health benefits and nutritional qualities. Oats (*Avena sativa*) are particularly high in soluble fiber, beta-glucan, lipids, proteins, and specific micronutrients as well as a unique source of polyphenols (Clemens and Klinken, 2014). Beta-glucan is the main functional component of cereal fibers mainly oats and barley and observed to have beneficial effects on LDL cholesterol, reduces it to 20-30% and reduces cardiovascular diseases. Beta-glucan fermentation in colon releases low molecular fatty acid, which shows its anti-carcinogenic effect (Oku, 1994; Salminen *et al*. 1998; Gallaher, 2000). Furthermore, beta-glucan also act as a prebiotic (Jaskari et al, 1998; Wood and Beer, 1998) and has high viscosity as well as water absorption capacities (Wood, 2007).

However, despite of all these health promoting benefits of oats, their consumption is not at par with other cereal grains. This is because of their bland, and unacceptable physical and organoleptic properties. Nevertheless, the physical and sensory parameters of oats can be enhanced by the process of fermentation (Martensson *et al*. 2001) [1]. Fermentation of oats with suitable microorganisms will lead to formation of flavour, acid development and consumption of carbohydrates. It was observed that the fermented oat based products have high suitability in terms of acidity, texture and overall appearance (Martensson *et al*. 2001) [1].

Furthermore, replacement of fat with dietary fiber is also drawing considerable attention these days. It has been observed that fiber can replace fat in high fat, ready to eat product, for example, bakery products (Knuckles *et al*. 1997, Lee *et al*. 2004; Lee *et al*. 2005; Brenan and Cleary, 2007; Kalingaand Mishra, 2009). Fat Replacers like fiber can replace most of the sensory and functional properties of fat, but since these are not chemically categorized as fat and therefore contribute lesser energy than fat. Cereal beta-glucan is considered as carbohydrate based fat replacer due to its water binding ability, high viscosity, and frothing and emulsion stability capacities (Barkus and Tamelli, 2000). Therefore, further research can be conducted where oats can be used as an alternative to fats in various spreads, margarines and mayonnaise.

Keeping all above in mind, the purpose of our present work is to develop a fermented oat based spread which is cholesterol free and low fat also determine its sensory, phytochemical and nutritional characteristics.

2. Materials and Methods

2.1 Starter culture

The microbial culture used in this study is *Saccharomyces cerevisae*, which is a yeast strain and was procured in the form of active dry yeast from the market. Starter culture was obtained by dissolving active dry yeast in distilled water (1:10) with a pinch of sucrose.

2.2 Oat substrate

The oat mash used as a substrate was prepared from the whole grain oats and distilled water in a ratio of 1:4 (w/v). It was then heated in temperature controlled water bath at 95 °C for 10min with stirring. The samples were brought to room temperature before the addition of required amount of starter culture.

2.3 Fermentation and storage

The oat mash was then inoculated with 0.5%, 1%, 1.5%, 2% and 2.5% (v/v) of starter culture respectively and fermentation was carried out at 25 °C for 2 days. Fermented mass was further heated again at 95 °C for 10min to stop yeast fermentation.

2.4 Spread preparation

The products were made based on a total of six components consisting of three dry ingredients (oat powder, salt, and stabilizer) and three liquid ingredients (vegetable fat, water and lemon juice). Three formulations were prepared; following the three components coordinate mixture design (Table 1).

Table 1: Formulation for spread

Ingredient	% vegetable fat	% water	% oats	% lemon juice
Fat based spread	30	60	0	10
Formulation 1 (Unfermented oats)	10	60	20	10
Formulation 2 (fermented oats)	10	60	20	10

2.5 Antioxidant analysis of DPPH radical scavenging activity

The DPPH radical scavenging method is used to measure the free radical scavenging activity of different elements (Brand-Williams *et al*. 1995). A solution of 0.1mM concentration of DPPH (Sigma-Aldrich Chemie, Steinheim, Germany) in methanol was added to 0.5ml of well diluted phenolic extracts. The change in absorbance at 517nm was measured after 30 min of incubation. Following equation can calculate the DPPH radical-scavenging activity of phenolic extract
Radical scavenging activity
(%) = (OD Blank-OD Sample*100)/OD Blank

2.6 Estimation of crude fiber

The crude fiber content of unfermented and fermented oats was estimated by digesting defatted samples first in 1.25% of H_2SO4 for ½ h, then filtered it with muslin cloth and remaining residue again digested with 1.25% NAOH for 30 min. Remaining residue was kept in oven for overnight at 105 °C. Further, it was kept in a muffle furnace for 4hr at 600 °C.
% crude fiber was calculated by following
formula Crude fiber (%) = (W2-W1)-(W3-W1)
*100/W W = weight of sample
W1 = weight of empty crucible
W2 = weight of sample + weight of empty crucible
W3 = weight of sample after ignition + weight of empty crucible

2.7 Determination of fat content

5g of fermented and unfermented samples were taken in an Erlenmeyer flask and dissolved in 50ml petroleum ether. It was further kept in shaker at 140rpm and 60 °C for overnight. The solution was filtered and the residue was re-extracted with 50ml petroleum ether. Both the filtrate was combined and evaporated at 60 °C in oven. The recovered or extracted fat in the beaker was weighted and calculated.
Fat (%) = weight of extracting fat ×100/weight of sample

2.8 Determination of protein content

Samples were dissolved in distilled water (1:10) for making sample extract, 1ml sample extract were mixed with 9ml biuret reagent. Incubate it for 20m and then absorbance was taken at 550nm. The amount of total protein content was calculated with the standard calibration curve of albumin.
Protein standard curve equation, Y = 0.1422X + 0.0634

2.9 Analysis of carbohydrate content

The clear aqueous solution is taken in a test tube and mixed with sulfuric acid and phenol. Due to interaction of carbohydrate and phenol the color of the solution will change to yellow-orange. The initial carbohydrate concentration in a sample is proportional to the absorbance at 490nm. This method determines the total sugar present in a sample by converting all non-reducing sugar to reducing sugar by means of sulfuric acid. A calibration curve is prepared using known carbohydrate concentration.
Glucose standard curve equation, Y= 24.536X-0.2714

2.10 Sensory evaluation

Sensory test is conducted by selecting a group from large population for whom the product is proposed (Meilgaard and others 1999). A panel of 10 members was selected and intensity scales were defined using 9cm lines for acidity, sweetness, consistency, oat flavour, mouth-feel, appearance, and overall acceptability. Ranking for preference was shown by noting the most desirable product according to the panelists. The panelists gave a number of the products according to their preference from 1-9 using the same sensory qualifications as in the intensity evaluation, except for oat flavor which was excluded from the test (1= dislike extremely, 5 = neither like nor dislike, 9= like extremely). Fermented oats were used for making the test spread. Unfermented product was used as control.

2.11 Shelf life evaluation

The shelf life of spread from fermented oats at refrigerated temperature was analyzed by checking for bacterial contamination at regular interval of 5 days for 25 days. Phase separation of the product was also checked after 25 days after storage at refrigerated temperature.

2.12 Statistical analysis

The experiments were done in triplicate and replicated twice. Results are expressed as average ± standard deviation (SD).

3. Result and Discussion

3.1 Starter culture concentration for fermentation of oats

The oat mash was inoculated with 0.5, 1.0, 1.5, 2.0 and 2.5% (v/v) of starter culture suspension of *S. cerevisae* separately aiming to achieve the appropriate taste, texture and flavor of the fermented substrate in minimum time. Short fermentation time is always preferable in order to minimize the risk of contamination (Blandino *et al*. 2002) [5]. Banigo *et al*. (1974), studied the manufacturing process of Ogi (traditional Nigerian fermented food) which involves the fermentation of maize, millet and sorghum with *S.cerevasie* for 24-72hrs to achieve desired texture, aroma and taste. Similarly, in the present study it was observed that 2% inoculum gave better taste, texture and aroma to the oats in 48h.

3.2 Phytochemical analysis of fermented and unfermented oats

The total increment of phenolic content in fermented oats was 61.6% as compared to unfermented oats (Table 2). This is because of yeast fermentation of oats, which released the bound phenolic compounds and free phenolic acids (Katina *et al*. 2007).

In grains, antioxidants are concentrated in the bran fraction, mainly as phenolic compounds in the form of insoluble bound ferulic acid (Hatcher and Krunger, 1997). With the aid of microbial fermentation, these bound phenolics in cereal grains can be released (Bhanja *et al*. 2007). As a consequence, the higher DPPH scavenging activity was observed in fermented oats when compared with unfermented ones (Table 3). Adom & Liu, (2002), provided strong evidence that the predominant source of antioxidant activity is derived from the phenolic compounds. The phenolic compounds in oats may be therefore able to, capture the free radicals formed in the human body, if their consumption is supported (Bryngelsson *et al*. 2002).

Table 2: Total phenolic content of fermented and unfermented oats

Samples	TPC (mg GAE g^{-1} grain)
Unfermented	195.2±0.31
Fermented	508.7±0.34

Table 3: Antioxidant activity of fermented and unfermented oats

Samples	% DPPH scavenging
Unfermented	43.02±2.44
Fermented	72.84±1.87

3.3 Nutritional analysis of fermented and unfermented oats

The nutritional profile of fermented and unfermented oats is given in Table 4. Generally, fermentation can decrease the level of carbohydrate along with some non-digestible poly or oligosaccharides (Blandino *et al*. 2002) [5]. In our study it can be seen that there is a decrease in carbohydrate content after fermentation (Table. 4). Unfermented oats have 63.6% carbohydrate while, after fermentation, it is reduced to 55.7%. Fermentation significantly improves the protein quality as well as the level of lysine in maize, millet, sorghum, and other cereals (Hamad and Fields, 1979). In our study it can be seen that there is slightly improvement in protein content of oats after fermentation. The protein content observed in unfermented oats was 7.81%, though fermented oats have 8.54% (Table. 4).

The average fat content of unfermented oats was observed to be 8.66%, which decreased to 6.46% after fermentation (Table 4). This decrease in fat is due to reduction of oleic acid content (Brindzova *et al*. 2008) [37]. Similar varietal composition of oat lipids was also reported previously (De La Roche *et al*. 1977; Saastamoinen *et al*. 1989). Oats contain a considerable amount of oleic acid. The polar lipids in the cell membranes are dominated by phospholipids and glyco-or galactolipids (Sugawara and Miyazawa, 2001).

The moisture and ash content of fermented and unfermented samples was found to be approximately same (Table 4). So, it can be said that there were no significant changes in moisture and ash content after fermentation.

Similarly, there was no significant change in crude fiber percentage after fermentation (Table 4). This indicates that beta-glucan in oats is not utilized by the yeast.

Gargi *et al*. 2017, also did nutritional analysis of fermented oats and our results were similar to them. However, we have also incorporated fermented oats into a product i.e. spread.

Table 4: Nutritional analysis of fermented and unfermented oats

Composition	Unfermented	Fermented
Fat (%)	8.66±0.115	6.46±0.305
Protein (%)	7.81±0.291	8.54±0.234
Crude fibre (%)	10.96±0.231	10.26±0.276
Carbohydrate (%)	63.6±0.256	55.7±0.249
Moisture (%)	10.77±0.189	10.55±0.129
Ash (%)	1.355±0.022	1.4±0.023
Energy (kcal)	330.02	285.84

Data is denoted as Average ± standard deviation

3.4 Formulation of spread

Oats based spread (Fig.1.) was formulated from both fermented and unfermented oats using the composition given above in Table.1. Butter based spread (Table.1.) was also made and it was observed that both fermented and unfermented oats can replace almost 60% fat in the formulated product. This is because the beta-glucan present in oats act as fat Replacers.

Kalinga (2010), shows the replacement of fat with beta glucan in bakery products. i.e. cake. Beta-glucan extracted from oats and barley and added to a cake formulation at different levels as a fat replacer.

Fig 1: Oats spread

3.5 Sensory evaluation

Generally, the panelists gave higher scores to a product made from fermented substrate.

Formulation A received the highest overall acceptability (8.67), while the control, i.e. spread made from unfermented oats was unacceptable (4.55). This indicates the importance of

fermentation in making the product (spread) from the oats acceptable.

Therefore, it can be concluded that fermentation can be used to enhance the taste and aroma of bland oats, which can thus be incorporated into different novel healthier and tastier food products.

Table 5: Average score of sensory attributes

Formulations	Appearance	Taste	Spread-ability	Smoothness	Overall acceptability
A (Spread from fermented oats)	8.9±0.251	9±0.1	7±0.152	2±0.057	8.67±0.083
Control (spread from unfermented oats)	1±0.547	2±0.547	3±0.547	6±0.456	4.55±0.404

The data is given as Average ± standard deviation and the values are based on a nine-point hedonic scale. Where 1= dislike, 5 = neither like nor dislike, 9 = like.

3.6 Estimation of shelf life

Too few to count (TFTC) colonies were seen in oat based spreads on the 25th day of refrigerated storage. Phase separation was also not seen on the 25th day of storage. So, it can be said that the product can be stored in refrigerator for more than 21 days.

4. Conclusion

In order to utilize health benefits of oats in various innovative products, like spreads and margarines, fermentation is a necessary step to improve its bland taste and flavor and thus increase consumer acceptability. The sensory evaluation showed that the spread made from fermented oats is highly acceptable (8.67 score) compared to that with the unfermented one (4.55 score). In addition to increasing the sensory characteristics, fermentation also increased the TPC and antioxidant content of the substrate. Furthermore, the beta glucan present in oats which was not utilized during the fermentation process can also be exploited as a fat replacer in conventional fat-based products by 60%.

5. References

1. Martensson O, Andersson C. Formulation of an oat based fermented product and its comparison with yoghurt. J Sci Food Agric. 2001; 81:1314-1321.
2. Angelov A, Gotcheva V. Development of new oat- based probiotic drink. International Journal of Food Microbiology. 2006; 112:75-80.
3. Das G, Maria J. Nutrient profile of fermented oats. International Journal of Food Science and Nutrition, 2017, 69-71.
   ISSN: 2455-4898,
4. Prado F, Pandey A. Trend in non-dairy probiotic beverages. Food Research International, 2007, 41.
5. Blandino A, Cantero D. Cereal based fermented foods and beverages. Food Research International. 2002; 36:527-543.
6. Thomas K, Ingledew W. Practical and theoretical consideration in the production of high concentration of alcohol by fermentation. Process Biochemistry, 1995, 0032-9592, 00073-9.
7. Cai S, Wang O, Wang W, Zhu F, Ji B, Zang D. Comparative study of the effects of solid state fermentation with three filamentous fungi on the total phenolic content, flavonoids and antioxidant activities of subfractions from oats. J Agric food chem. 2012; 60:507-513.
8. Aeschelimann A, Vonstocar U. The effect of yeast extract supplementation on the production of lactic acid from whey permeates by *Lactobacillus helveticus*. Applied microbiology and Biotechnology. 1990; 32:398-440.
9. Gupta S, Cox S, Ghannam N. Process optimization for the development of a functional beverage based on lactic acid fermentation of oats. Biochemical engineering journal, 2010.
10. Casey GP, Ingledew WM. Ethanol tolerance in yeast. CRC Crit. Rev. Biotechnol. 1986; 13:219-80.
11. Arunachalam K, Gill HS, Chandra RK. Enhancement of natural immunity functions by dietary consumption of *Bifidobacteriumlactis* HN019. European journal of clinical Nutrition, 2000, 54:1-4.
12. Ingledew WM, Casey GP. Rapid production of high concentrations of ethanol using unmodified industrial yeast. In Biotechnology and Renewable Energy, Ed. M. Moo-Young, S. Hasnain & J. Lamptey. Elsevier, New York, 1986, 246-57.
13. Scott RW, Solubilization of beta-glucan during mashing. J Inst. Brew. 1972; 78:411-2.
14. Chae HJ, Joo H, In MJ. Utilization of brewer's yeast cells for the production of food grade yeast extract. Part I: Effects of different enzymatic treatments on solid and protein recovery and flavor characteristics. Bioresource Technology. 2001; 76:253-258.
15. Anderson JW, Deakins DA, Floore TL, Smith BM, Whitis SE. Dietary fiber and coronary heart disease. Critical Reviews in Food Science and Nutrition, 1990a, 29:95-147.
16. Anderson JW, Spencer DB, Hamilton CC, Smith SF, Tietyen J, Bryant CA *et al*. Oat-bran cereal lowers serum total and LDL cholesterol in hypercholesterolemic men. American Journal of Clinical Nutrition. 1990b; 52:495-499.
17. Andersson AAM, Armo E, Grangeon E, Fredriksson H, Anderson R, Aman P. Molecular weight and structure units of (1-3, 1-4) -β-glucan in dough and bread made from hull-less barley milling fractions. Journal of Cereal Science. 2004; 40:195-204.
18. Anon. Fat Substitute Update. Food Technology. 1990; 44:92-97.
19. Anon. Food labelling: soluble dietary fibre from certain foods and coronary heart disease. Federal Register: USFDA, 1997, 67.
20. Anon. Position of the American Dietetic Association: Fat replacers. Journal of the American Dietetic Association. 1998; 98:463-468.
21. Anon. The definition of dietary fiber. Report of the dietary fiber definition committee to the board of directors of the American Association of Cereal Chemists. USA: American Association of Cereal Chemists, 2001.
22. Anon. Food Labelling: health claims; oats and coronary heart disease. Federal Register: USFDA, 2002, 62.
23. Arozarena I, Bertholo H, Empis J, Bunger A, Sousa I. Study of the total replacement of egg by white lupine protein, emulsifiers and xanthan gum in yellow cakes. European Food Research and Technology. 2001; 213:312-316.

24. Gibson GR, Roberfroid MB. Dietary modulation of the human colonic microbiota: Introducing the concept of prebiotics. Journal of Nutrition. 1995; 125:1401-1412.
25. Gill HS, Cross ML, Rutherfurd KJ, Gopal PK. Dietary probiotic supplementation to enhance cellular immunity in the elderly. British Journal of Biomedical Science. 2001; 57:94-96.
26. Bamforth CW. β-Glucan solubilizes. J Inst. Brew. 1989; 95:314-5.
27. Maisch WF. Fermentation processes and products. In Corn Chemistry and Technology, ed. S. A. Watson & E E. Ramsted. American Association of Cereal Chemists, St Paul, MN, 1987, 553-74.
28. Wood PJ. Oat β-glucan-physiochemical properties and physiological effects. Trends Food Sci Technol. 1991; 12:311-314.
29. Lambo A, Öste R, Nyman MEGL. Dietary fibre in fermented oat and barley-glucan rich concentrates. Food Chem. 2005; 89:283-293.
30. Chavan J, Kadam S. Critical reviews in food science and nutrition. Food Sci. 1989; 28:348-400.
31. Kalinga DN, Mishra VK. Pasting and sorption behavior of β-glucan and flour mixtures. In Panozzo, JF, & Black, CK. 58th Australian Cereal Chemistry Conference, 2008, 203-206.
32. Lazaridou A, Biliaderis C, Izydorczyk M. Molecular size effects on rheological properties of oat-glucans in solution and gels, Food Hydrocolloid. 2003; 17:693-712.
33. Kedia G, Wang R, Patel H, Pandiella S. Use of mixed cultures for the fermentation of cereal-based substrates with potential probiotic properties, Proc. Biochem. 2007; 42:65-70
34. Steinkraus KH. Lactic acid fermentation in the production of foods from vegetables, cereals and legumes. Antonie van Leeuwenhoek. 1983; 49:337-348.
35. Bhanja T, Kumari A, Banerjee R. Enrichment of phenolics and free radical scavenging property of wheat koji prepared with two filamentous fungi. Bioresource Technology. 2009; 100:2861-2866.
36. Hesseltine CW. The future of fermented foods. Nutrition Reviews. 1983; 41:293-301.
37. Brindzova L, Certik M, Rapta P, Zalibera M, Mikulajova A, Takacsova M. Antioxidant activity, β-glucan and lipid contents of oat varieties. Czech J Food Sci. 2008; 26:163-173.
38. Garcia K. Quality characterization of cholesterol free mayonnaise type spreads containing rice bran oil. B.S. chemical engineering, 2006, 14-22.