EFFECT OF FAT REPLACERS ON THE QUALITY CHARACTERISTICS OF NON-FAT YOGHURT

Thushara, V[1]. K. Radha[2*] and C. T. Sathian [3]

[1] M.Sc. Scholar, [2] Assistant professor, [3] Professor and Head
Department of Dairy Science,
College of Veterinary and Animal sciences, Mannuthy-680651, Kerala, India

ABSTRACT

Non-fat yoghurt was prepared by using inulin and whey protein concentrate (WPC) as fat replacers at one and two per cent level. The experimental yoghurts were compared with control yoghurt samples prepared from skim milk and whole milk. pH, titratable acidity, fat, total solids, syneresis, water holding capacity (WHC) and protein contents were studied. Enumeration of viable *L. delbrueckii* ssp. *bulgaricus* and *S. thermophilus* was also carried out. During storage, pH of all the yoghurt samples decreased whereas total solids increased. Addition of inulin and WPC has controlled the development of acidity during storage. Addition of inulin caused increased syneresis whereas WPC decreased the syneresis percentage. Addition of WPC (2%) has significantly increased the protein percentage and improved the WHC. Both inulin and WPC have increased the *Lactobacillus* as well as *Streptococcus thermophilus* count. Non-fat yoghurt incorporated with WPC (2%) had obtained maximum sensory scores.

Keywords: Fat replacers, Inulin, Whey protein concentrate, non-fat yoghurt, Functional yoghurt.

INTRODUCTION

Yoghurt is a popular and well accepted fermented milk product widely consumed all over the world. It is obtained by the controlled fermentation of milk by *Lactobacillus delbrueckii ssp. bulgaricus and Streptococcus thermophilus*. Yoghurt has achieved considerable economic importance worldwide owing to the high nutritional and therapeutic value. It has been attributed with nutraceutical, therapeutic and probiotic effects such as improvement in digestion, immune modulation, cholesterol lowering and anti carcinogenic activity. However, traditional yoghurt contains considerable amount of fat. As the relationship between fat consumption and heart diseases has been well established, the reduction of dietary animal fat has been recommended by nutritionists. As milk fat plays an important role in the texture, flavour and colour development of dairy products, lowering the fat content in dairy products will severely affect the sensory qualities of low fat/non fat dairy products. Hence, fat replacers were developed to overcome the textural defects and organoleptic problems in low-fat/no-fat products. The term fat replacer is used to describe any ingredient used to replace fat.

The major categories of fat replacers include carbohydrate based, protein based and fat based replacers. Among these, inulin has gained popularity as it provides physiological benefits and lower calorific value and can thus be used in foods designed for weight management. Whey protein concentrates can also be used as fat replacers because of their functional properties and low cost (Sodini *et al,* 2006). Whey protein concentrates are

***Corresponding Author:** Dr. K. Radha, Email: radhavet@gmail.com, Mobile: +91 9744481598

produced by ultrafiltration of whey to enrich the protein fraction by removal of lactose, minerals and other low molecular weight components. Reduced-fat yoghurts can be produced by replacing the fat partially with these fat replacers. The effect of different fat replacers on textural and physical properties of low fat yoghurts has been studied by various researchers but the results are inconsistent. Hence, the work is designed to study the effect of inulin and whey protein concentrate on the quality characteristics of non fat yoghurt.

MATERIALS & METHODS

Skim milk and whole milk required for the study were collected from the University Dairy plant, College Of Veterinary and Animal Sciences, Mannuthy, Kerala. To enhance the solids content in the milk, skim milk powder was used. Inulin, a prebiotic powder (Orafti ® HP inulin) and Whey protein concentrate (70%) were received as free samples from Brenntag Ingredients India Pvt Ltd, Gurgaon. Mixed starter culture containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus* (NCDC- 145) procured from National Collection of Dairy Cultures, National Dairy Research Institute, Karnal was used as starter culture for the preparation of yoghurt.

Preparation of yoghurts

Control skim milk yoghurt (C_1) and Control full fat yoghurt (C_2) were prepared with the addition of 3.5 per cent skim milk powder (SMP). Treatment groups of yoghurt were prepared by using inulin and whey protein concentrate (WPC) as fat replacers. Four experimental preparations of yoghurt incorporating one per cent inulin (T_1), two per cent inulin (T_2), one per cent WPC (T_3) and two per cent WPC (T_4) were prepared. The milk was pasteurized at 90ºC for five minutes, and cooled to 40 to 42ºC. Then it was inoculated with two per cent yoghurt starter culture comprising of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus* in 1:1 ratio and incubated at 42ºC. The yoghurt was then stored under refrigeration. Chemical and microbiological qualities of yoghurt samples were evaluated during storage.

The following determinations were carried out in control and treatment groups of yoghurt samples. pH (Cyber Scan 2500 digital pH meter), fat (IS: 1224, 1977), titratable acidity (IS: 11766, 1986), total solids content (IS: 12333, 1997), Syneresis (Rodarte *et al.,* 1993), Water Holding Capacity (Sodini, 2002) and protein (AOAC, 1995) were estimated on the first and seventh day of refrigerated storage. The counts of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus* were determined by plate count method (IS: 18, 1980). The fresh yoghurt samples were evaluated for their sensory characteristics such as colour & appearance, flavour, body & texture and overall acceptability on a 5-point hedonic scale as per the method recommended by IDF (1987). A panel consisting of seven members belonging to the Department of Dairy Science has evaluated the yoghurt samples. Statistical analysis of the data was done by using analysis of variance (ANOVA) of SPSS program, version 21 (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

The results obtained are presented in table 1 to 9. The pH values ranged from 4.32 to 4.47 on day one and 4.23 to 4.31 on day seven. The pH values of the yoghurt gradually decreased during storage, and this was found to be statistically significant ($p<0.01$). A similar trend was also observed by Almeida *et al*. (2009). The decrease in pH can be attributed to the post-fermentation acidification due to the metabolic activity of *S. thermophilus* and *L. bulgaricus* during storage.

The mean fat percentage of control full fat yoghurt was 3.50 on first and seventh days of storage. The mean fat percentage of control skim milk yoghurt was 0.30 on both first and seventh

days of storage. Yoghurt incorporated with 1, 2 per cent WPC and 1, 2 per cent inulin also contained 0.30 per cent fat on the first and seventh days of storage. No significant change in fat percentage was observed during storage in all the yoghurt samples. The results of the present study are in agreement with the findings of Kavas *et al.* (2003) who reported no change in fat percentage of yoghurt during storage.

Titratable acidity increased with the advancement of storage period in all the yoghurt samples. Statistical analysis revealed a significant increase in the titratable acidity on the 7th day from the initial value in all the samples. The highest acidity value was observed for the control full fat yoghurt sample. The sample containing 1% inulin had the lowest acidity percentage. The increase in titratable acidity observed during storage may be attributed to the growth of lactic acid bacteria and production of lactic acid. Addition of inulin and whey protein concentrate has controlled the development of acidity and the acidity values were lesser than the values in control samples. Akalin *et al.* (2007) and Sady *et al.* (2007) found that the addition of WPC to yogurt increased the buffering capacity which controlled the progress of acidification during storage.

Total solids content did not show any significant difference between control and treatment samples of yoghurt. However, significant increase was observed in the total solids content on the 7th day from the initial value in all the samples. The increase in total solids can be attributed to the evaporation of moisture during storage. Similar results were reported by Guven *et al*. (2005) and Patocka *et al*. (2006).

The syneresis percentage ranged from 2.37 to 2.93 on day 1, and 1.76 to 2.40 on day seven. Use of inulin and WPC as a fat replacer did not significantly affect the syneresis percentage. Addition of WPC at 2% level decreased syneresis percentage on 1st day when compared to control skim milk yoghurt. Significant decrease in syneresis percentage was observed on the 7th day from the initial value in all the yoghurt samples except for samples added with 2% WPC. Results of the present study are comparable with the results of Aziznia *et al.* (2008) who reported that the incorporation of WPC to non fat yoghurt caused a more compact structure consisting of robust casein particles and large aggregates. Firmness of yoghurt increased and susceptibility to syneresis decreased as the level of incorporation of WPC increased.

Water holding capacity (WHC) between control and treatment groups showed significantly higher values for control full fat yoghurt and yoghurt added with two per cent WPC than the other four groups. An increasing trend in water holding capacity of yoghurt was noticed during storage in all the six groups of yoghurt. The higher water holding capacity of yoghurt fortified with whey protein concentrates when compared to other treatment groups can be attributed to the higher protein content in that group which might have improved the gel strength and restricted the water mobility.

Cheng *et al.* (2003) found that when skim milk powder was replaced by whey protein concentrates, yogurt showed an increase in water-holding capacity. Berber (2011) reported that water holding capacity of yoghurt incorporated with 1% WPC, control full fat yoghurt and control skim milk yoghurt were 682.43, 725.54 and 582.06 respectively. Berber *et al.* (2015) had also reported that yoghurt incorporated with whey protein concentrate showed better water holding capacities than controls.

Significantly higher protein values were observed for samples containing 1% and 2% WPC when compared to the yoghurt containing 1 percent and 2 percent Inulin and control samples. The results of the present study are in agreement with the findings of Guven *et al*. (2005) and Berber (2011).

Lactobacillus and *Streptococcus* count of control and experimental yoghurt samples are shown in Table 8. Yoghurt incorporated with inulin and whey protein concentrate showed significantly higher counts for both *Lactobacillus* and *Streptococcus* when compared to control skim milk yoghurt. A similar trend was observed by Sady *et al.* (2007) and Marafon *et al*. (2011). In their study addition of whey protein concentrate (WPC) to replace non-fat dry milk resulted in an increase in *Streptococcus* and *Lactobacillus delbrueckii* ssp. *bulgaricus* counts during storage. However, Mazloomi *et al.* (2011) had reported that the addition of inulin to milk decreased the viability of *L. delbrueckii* ssp. *bulgaricus* during the storage of yoghurt.

Sensory scores between control and treatment groups of yoghurt showed no significant difference. However, yoghurt incorporated with whey protein concentrate had obtained better flavour and overall acceptability scores than yoghurt incorporated with inulin. Yoghurt incorporated with 2 per cent WPC had obtained maximum sensory scores among the treatment groups of yoghurt. Similar results were reported by Berber *et al.* (2015). In their study, sensory results revealed that yoghurt incorporated with whey protein concentrate (80%) had better flavour and overall liking scores than controls. However, Crispin-Isidro *et al.* (2015) reported that reduced fat yogurt incorporated with 40g L-1 inulin or 60 g L-1 agave fructans had superior sensory characteristics than that of the control. The lower levels of inulin used in the present study might be the reason for comparatively lower sensory scores in inulin added yoghurts.

CONCLUSION

From the above findings, it can be concluded that non-fat yoghurt with acceptable quality can be prepared by using fat replacers such as whey protein concentrate and inulin at both one and two per cent levels. Addition of inulin and whey protein concentrate has controlled the development of acidity and the acidity values were lesser than the values in control samples. Addition of inulin caused increased syneresis in non-fat yoghurt whereas whey protein concentrate decreased the syneresis percentage. Control full fat yoghurt and yoghurt added with two per cent WPC showed better water holding capacities than the other yoghurt samples. Both inulin and WPC have increased the *Lactobacillus* as well as *Streptococcus thermophilus* count. In the present study no significant difference in the sensory scores were observed between control and treatment groups of yoghurt. Slightly lower sensory scores were obtained for control skim milk yoghurt and all treatment groups when compared to control full fat yoghurt. However, yoghurt incorporated with two per cent WPC had obtained almost similar sensory scores as that of full fat yoghurt.

ACKNOWLEDGEMENT

The authors acknowledge Kerala Veterinary and Animal Sciences University for providing financial support and infrastructure facilities required for the conduct of research work.

REFERENCES

AOAC. 1995. Official Methods of Analysis. (15th Ed.) Horwith, Washington.

Akalin, A.S., Gonç, S., Unal, G. and Fenderya, S. 2007. Effects of fructooligosaccharide and whey protein concentrate on the viability of starter culture in reduced fat probiotic yogurt during storage. J. Food Sci. **72**: 222-227.

Almeida, K.E., Tamime, A.Y. and Oliveira, M.N. 2009. Influence of total solids contents of milk whey on the acidifying profile and viability of various lactic acid bacteria. LWT - Food Sci. and Technol. **42**: 672-678.

Aziznia, S., Khosrowshahi, A ., Madadlou, A ., Rahimi, J . 2008 Whey protein concentrate and gum tragacanth as fat replacers in non fat

yogurt: chemical, physical, and micro structural properties. J. Dairy Sci. 91(7):2545-52.

Berber, M. 2011. Whey Protein Concentrate as a Substitute for Non-Fat Dry Milk in Yogurt. *B.Sc. thesis*, The Ohio State University.

Berber, M., González-Quijano, G.K., Alvarez, V.B. 2015. Whey Protein Concentrate as a Substitute for Non-fat Dry Milk in Yogurt. J. Food Process Technol. 6: 530. doi: 0.4172/2157-7110.1000530

Cheng, L.J., Augustin, M.A. and Clarke, P.T. 2003. Yogurts from skim milk – whey protein concentrate blends. Aust. J. Dairy Technol. **55**: 110.

Crispin-Isidro, G., Lobato-Calleros, C., Espinosa-Andrews, H., Alvarez-Ramirez, J. (2015). Effect of inulin and agave fructans addition on the rheological, microstructural and sensory properties of reduced-fat stirred yogurt. Food Sci. Technol. **62**: 438-444.

Guven, M., K. Yasar, O.B. Karaca and A.A. Hayaloglu. 2005. The effect of inulin as a fat replacer on the quality of set-type low-fat yogurt manufacture. Int. J. Dairy Technol. **58**:180-184.

IDF. 1987. *Sensory Evaluation of Dairy Products, Standard 99A*. International Dairy Federation, Brussels, Belgium.

IS: 11766, 1986. Method for determination of titratable acidity in milk powder and similar products. Bureau of Indian Standards, New Delhi.

IS: 1224, 1977. Part II, Determination of Fat by Gerber Method. Bureau of Indian Standards, New Delhi.

IS: 12333, 1997. Milk, Cream and evaporated milk: determination of total solids. Bureau of Indian Standards, New Delhi.

IS: 18, 1980. Part I, ISI Handbook of food analysis. Bureau of Indian Standards, New Delhi.

Kavas, G., H. Uysal, S. Kilic, N. Akbulut and H. Kesenkas. 2003. Some properties of yoghurt produced from goat milk and cow-goat milk mixtures by different fortification methods. Pak. J. Biol. Sci. **6:** 1936-1939.

Marafon, A.P., Sumi, A., Granato, D., Alcantara, M.R., Tamime, A.Y. and Nogueira de Oliveira, M. 2011. Effects of partially replacing skimmed milk powder with dairy ingredients on rheology, sensory profiling, and microstructure of probiotic stirred-type yogurt during cold storage. J. Dairy Sci. **94**: 5330-5340.

Mazloomi, S.M., Shekarforoush, S.S., Ebrahimnejad, H. and Sajedianfard, J. 2011. Effect of adding inulin on microbial and physicochemical properties of low fat probiotic yoghurt. Iranian Vet. J. Res. **12:** 93-98.

Patocka, G., R. Cervenkova, S. Narine and P. Jelen. 2006. Rheological behavior of dairy products as affected by soluble whey protein isolate. Int. Dairy J. **16**(5): 399-405.

Rodarte, C.W., M.V. Galvan, A. Farres, F. Gallardo, V.E. Marshall and M.G. Garibay. 1993. Yogurt production from reconstituted skim milk powders using different polymer and non-polymer forming starter cultures. J. Dairy Res. **60:**247-254.

Sady, M., J. Domagala, T. Grega and D. Najgebauer-Lelko. 2007. Quality properties of non-fat yoghurt with addition of whey protein concentrate. Biotech. Animal Husb. **23** (5-6): 291 – 299.

Snedecor, G.W. and W.G. Cochran. 1994. *Statistical Methods*. (8th Ed.). Oxford and IBH Publishing Company, Calcutta. 564p.

Sodini, I., A.Lucas, M.N. Oliveira., F. Remeuf and G. Corrieu. 2002. Effect of milk base and starter culture on acidification, texture, and probiotic cell counts in fermented milk processing. J. Dairy Sci. **85:** 2479–2488.

Sodini I., J. Mattas and P.S. Tong . 2006. Influence of pH and heat treatment of whey on the functional properties of whey protein concentrates in yoghurt. Int. Dairy J. **16**: 1464-1469.

Table.1 pH (Mean±S.E.) of yoghurt samples during storage

Sample	1st day	7th day	t value
C1(full fat)	4.44±0.02^{ab}	4.31±0.02^{a}	5.79**
C2(skim milk)	4.38±0.02^{bc}	4.28±0.02^{ab}	6.95**
T1(1% inulin)	4.45±0.01^{a}	4.27±0.02^{ab}	8.88**
T2(2% inulin)	4.36±0.01^{c}	4.24±0.01^{b}	10.40**
T3(1% WPC)	4.47±0.03^{a}	4.29±0.01^{ab}	7.06**
T4(2% WPC)	4.32±0.01^{c}	4.23±0.01^{b}	10.14**
F- value	6.84**	2.48*	

** Significant at 0.01 level; * Significant at 0.05 level

Table.2. Fat percentage of yoghurt samples during storage

Sample	1st day	7th day
C1(full fat)	3.50	3.50
C2(skim milk)	0.30	0.30
T1(1% inulin)	0.30	0.30
T2(2% inulin)	0.30	0.30
T3(1% WPC)	0.30	0.30
T4(2% WPC)	0.30	0.30

Table.3 Titratable acidity (Mean±S.E.) of yoghurt samples during storage

Sample	1st day	7th day	t value
C1(full fat)	0.91±0.007^{a}	1.18±0.006^{a}	30.71**
C2(skim milk)	0.83±0.010^{b}	1.17±0.012^{a}	35.80**
T1(1% inulin)	0.71±0.006^{e}	1.06±0.009^{d}	70.00**
T2(2% inulin)	0.73±0.004^{de}	1.07±0.007^{cd}	33.28**
T3(1% WPC)	0.73±0.004^{d}	1.09±0.010^{bc}	35.50**
T4(2% WPC)	0.75±0.006^{c}	1.11±0.010^{b}	41.97**
F- value	121.21**	28.75**	

** Significant at 0.01 level

/Table.4 Total solids percentage (Mean±S.E) of yoghurt samples during storage

Sample	1st day	7th day	t value
C1(full fat)	13.87±0.23	16.56±0.40	12.48**
C2(skim milk)	15.06±0.48	17.06±0.40	5.71**
T1(1% inulin)	14.22±0.31	16.21±0.41	10.85**
T2(2% inulin)	14.94±0.51	17.07±0.40	6.66**
T3(1% WPC)	14.57±0.42	16.76±0.38	12.55**
T4(2% WPC)	14.88±0.35	17.05±0.37	7.66**
F- value	1.36^{ns}	0.76^{ns}	

** Significant at 0.01 level (p≤0.01); ns- Non-significant at 0.05 level

Table.5 Syneresis percentage (Mean±S.E) of yoghurt samples during storage

Sample	1st day	7th day	t value
C1(full fat)	2.37±0.15	2.02±0.17	4.89**
C2(skim milk)	2.88±0.19	2.06±0.16	7.20**
T1(1% inulin)	2.93±0.18	2.26±0.16	5.59**
T2(2% inulin)	2.87±0.17	1.76±0.14	11.44**
T3(1% WPC)	2.72±0.17	2.40±0.16	1.92*
T4(2% WPC)	2.37±0.22	2.26±0.16	0.62^{ns}
F- value	1.20^{ns}	1.88^{ns}	

** Significant at 0.01 level; * Significant at 0.05 level; ns - Non-significant at 0.05 level

Table.6 WHC (g/ kg) of yoghurt (Mean±S.E) samples during storage

Sample	1st day	7th day	t value
C1(full fat)	$653.75±8.22^{a}$	731.25±10.59	11.48**
C2(skim milk)	$623.75±7.30^{b}$	716.25±13.87	6.55**
T1(1% inulin)	$621.25±6.10^{b}$	718.75±13.68	8.51**
T2(2% inulin)	$622.50±5.90^{b}$	720.00±11.95	11.60**
T3(1% WPC)	$626.25±5.65^{b}$	711.25±11.40	6.56**
T4(2% WPC)	$647.50±5.90^{a}$	708.75±14.20	3.30*
F- value	4.70**	0.39^{ns}	

** Significant at 0.01 level; * Significant at 0.05 level; ns- Non-significant at 0.05 level

Table.7 Protein percentage (Mean±S.E) of yoghurt samples during storage

Sample	1st day	7th day	t value
C1(full fat)	6.99±0.12[a]	7.01±0.12	0.89[ns]
C2(skim milk)	7.06±0.09[a]	7.08±0.04	0.92[ns]
T1(1% inulin)	6.76±0.08[b]	6.76±0.06	1.05[ns]
T2(2% inulin)	6.80±0.12[b]	6.86±0.15	1.03[ns]
T3(1% WPC)	7.40±0.06[c]	7.42±0.05	0.84[ns]
T4(2% WPC)	8.00±0.13[d]	8.05±0.09	1.43[ns]
F- value	20.51**	17.96**	

** Significant at 0.01 level; ns - Non-significant at 0.05 level

Table. 8 Microbiological analysis of yoghurt samples during storage

Samples	*L. bulgaricus* log (c.f.u/g)		*S. thermophilus* log (c.f.u/g)	
	1st day	7th day	1st day	7th day
C1(full fat)	22.44±0.06[a]	22.74±0.08[a]	22.29±0.06[a]	22.65±0.05[a]
C2(skim milk)	22.00±0.08[d]	22.14±0.07[b\]	21.60±0.14[b]	21.95±0.12[c]
T1(1% inulin)	22.35±0.03[ab]	22.60±0.02[a]	21.91±0.05[ab]	22.42±0.04[ab]
T2(2% inulin)	22.33±0.03[abc]	22.67±0.01[a]	21.93±0.11[a]	22.42±0.07[ab]
T3(1% WPC)	22.12±0.17[bcd]	22.39±0.31[b]	21.90±0.08[ab]	22.34±0.06[b]
T4(2% WPC)	22.05±0.06[cd]	22.40±0.07[ab]	21.81±0.11[ab]	22.31±0.08[b]
F value	3.83**	3.95**	4.85**	8.18**

** Significant at 0.01 level

Table. 9. Sensory evaluation (Mean ± S.E) of yoghurt samples

Sample	Appearance& colour	Body & Texture	Flavour	Overall Scores
C1(full fat)	4.66±0.16	4.50±0.18	8.67±0.21	17.83±0.42
C2(skim milk)	3.75±0.62	4.00±0.63	7.67±1.08	15.41±2.27
T1(1% inulin)	3.75±0.60	3.75±0.47	6.83±0.83	14.33±1.81
T2(2% inulin)	4.33±0.21	3.58±0.30	6.17±0.98	14.08±1.33
T3(1% WPC)	4.16±0.50	3.83±0.54	7.17±1.27	15.16±1.98
T4(2% WPC)	4.16±0.33	4.16±0.50	8.33±0.61	16.66±1.37
Chi-Square value	11.09	9.71	9.82	9.40
p-value	0.05[ns]	0.08[ns]	0.08[ns]	0.09[ns]

ns- Non-significant at 0.05 level