

# Effect of egg yolk substitution by sweet Whey Protein Isolate on texture, stability and colour of Gelato-style vanilla ice cream

WPI substitution of egg yolk in Gelato

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## INTRODUCTION

Gelato, an Italian style of ice cream, differs from other ice creams in having more powerful flavour and denser texture because it has less incorporated air (Damerow 1995; Marshall *et al.* 2003). Although they are usually made with milk and cream (less than 10% milk fat), they often contain more egg yolk than other ice creams and thus taste equally rich, if not richer (IDFA 2005; USDEC, 2001). Whey protein isolate (WPI) is obtained by the liquid resulting from the coagulation of milk and is generated from cheese manufacture. Sweet WPI, with a pH of at least 5.6, originates from rennet-coagulated cheese production

such as cheddar. WPI contains at least 90% proteins, with virtually all the lactose removed. An ion-exchange step is often used in a conjunction with membrane filtration (Goff 2003; Tunick 2008). Furthermore, the functional attributes of WPI that contribute to ice cream processing include foaming, gelation, emulsification and flavour binding, which give it an advantage from a formulation standpoint (Eissa *et al.* 2004; Morr 1985; Morr 1989). WPI most important functional property is its foaming ability. According to the study on the foaming properties of proteins by Zhu & Damodaran (1994), it was discovered that the WPI sample once heated for one minute at 70°C achieved better foaming ability and improved foaming stability than the rest of the protein samples. This progress in foaming properties resulted from not only conformational transformation but also by the proportion between monomer and polymeric protein species found in WPI. The outcomes established that while monomeric proteins facilitated foamability, polymeric proteins supported foam stability (Zhu & Damodaran 1994). In recent years, whey proteins, especially WPI, have become the most employed functional food proteins in food formulations.  $\beta$ -lactoglobulin ( $\beta$ -Lg) and  $\alpha$ -lactalbumin ( $\alpha$ -La) account for 70% of the total protein content of the whey and are responsible for emulsifying, gelation, hydration and foaming properties of WPI (Tosi *et al.* 2007). In typical food foams, for example, the presence of adsorbed proteins at the fluid interface between the aqueous and the gas phases is responsible for the stabilisation of the microstructure. For food manufacturers a method to improve the stability of food foams consists in enhancing the functional properties of proteins (Tunick 2008).

Egg yolks have long been used in the making of ice cream, and recently the use of dehydrated eggs by many ice cream manufacturers has become a common practice. Hence, egg yolk is a very widely used food emulsifier for the manufacturing of ice cream and many frozen desserts, because of its excellent emulsifying properties (Guilmineau & Kulozik 2006a; b). Besides making smoother textured ice cream with firmer body, egg yolks increase

nutritional value and help blend the flavours of other ingredients. They help in whipping up low-solid mixes and those containing fat from sources other than cream. On the other hand, increasing the amount of egg yolk in ice cream mixes will cause many changes such as foam melting, eggy flavour and higher level of cholesterol (Damerow 1995; Hatta *et al.* 2008). Egg yolks, however, can be a costly ingredient, while whey protein isolate is much more economical and is widely available. Therefore, the objective of this study was to determine the effects on physical properties (including texture, stability and colour) of frozen Gelato ice cream with whey protein isolate as a substitute of egg yolk over four weeks of storage.

## MATERIALS AND METHODS

### **Materials**

Spray dried egg yolk powder (32.9 g/100 g protein, 5.5% maximum moisture) was obtained from PACE Farms Pty. Ltd. Egg Production (Warabrook, NSW, Australia) and stored in a dry place at  $17\pm 1^{\circ}\text{C}$ . A commercial sweet whey protein isolate (100% WPI - Hydrolysed Mega Ripped Protein) was supplied by Top Nutrition Company, (Newcastle, NSW, Australia) and stored at  $17\pm 1^{\circ}\text{C}$ . Sucrose, vanilla extract (natural), skim milk powder (0.1%fat), commercial whole full cream milk (3.4% fat) and 35% fat cream were purchased from local supermarket. Milk and cream were stored at  $4\pm 1^{\circ}\text{C}$ .

### **Gelato ice cream manufacturing**

Nine batches of Gelato ice cream mix were made and each batch represented one treatment. Three replications were performed and a total of 108 Gelato ice creams manufactured. All treatments contained 7-8% milk fat, 10-14% milk solid non-fat (MSNF) and 20% sucrose. Stabilisers and emulsifiers were not added to any treatments. A summary of the formulations used is presented in Table 1. WPI levels (0, 20, 50, 80, 100%) throughout this paper refer to

the level of substitution of egg yolk. Mixes were made by mixing skim milk powder and whey protein isolate with milk and heated, at 45°C for 3 min. Egg yolk powder was reconstituted according to the manufacturers instructions, and stored at 4°C overnight. The reconstituted egg yolk and the sucrose were mixed together prior to adding to the milk to ensure proper mixing. Mixes were cook over low heat, stirring constantly, until a temperature of 85°C was reached and maintained for one minute. The mixture was removed from the heat and stirred with the cream, then passed through a fine strainer and placed over an ice bath to chill (to achieve 20°C). Vanilla extract was added and the mixture was stirred for one minute. The mixture was processed using an ice cream maker (II Gelataio, GC5000 model, Treviso, Italy). All samples were packed in 400mL Polypropylene containers (Julzar PTY. TD., Queensland, Australia). Samples were hardened by placing in a freezer (NUAIRE -85±1°C Ultralow Freezer) at -85±1°C for 30 minutes, and then stored in a commercial freezer (Fisher & Paykel freezer, model# H701) at approximately -18±1°C until the testing day.

### **Texture**

Texture Analysis was conducted using a microprocessor controlled texture analysis system in conjunction with data collection and analysis software (TMS-Pro, S.I. Instruments, S.A, Australia). The method of Lim *et al.* (2008) and Szczesniak (2002) was used to evaluate texture characteristics. The conditions for analysis in this study were as follows: a 2 mm diameter probe penetrated the ice cream to a depth of 10 mm. The analysis used a 250 N load cell while the probe speeds during and after penetration were 25 and 400 mm/min, respectively. The maximum force was recorded. Analysis was performed in triplicate.

### **Melting Rate**

The method of Ohmes *et al.* (1998) and Specter & Setser (1994) was used to evaluate melting characteristics. The dimensions of the sample varied slightly from one Gelato ice cream to another (4.5 to 5cm × 4 to 4.5 cm × 3 to 3.5 cm) due to the differences in physical characteristics (e.g., overrun). The sample was placed (at room temperature, 17±1°C) on a wire screen (56 holes/cm<sup>2</sup>) on top of a funnel that was attached to a 100 mL graduated cylinder underneath. The volume of the melted ice cream was recorded after 40 minutes. The measurements were taken in triplicate.

### **Colour**

A Chroma Meter (model CR-131 Minolta®, Minolta Camera Co. LTD., Osaka, Japan) was used to measure Gelato ice cream colour whiteness (*L*) and yellowness (*b*). After melting, the sample (10±1°C) was collected and its *L* and *b* values were measured at room temperature (17±1°C) using the Chroma Meter. Three measurements were taken per single sample and the average was calculated.

### **Statistical Analysis**

Analysis of Variance (ANOVA) (using SPSS for Windows) was used to evaluate the effect of egg yolk substitution by WPI, on texture, stability and colour properties of Gelato vanilla ice cream over the storage period. Significant differences were determined at ( $P < 0.05$ ).

## **RESULTS AND DISCUSSION**

### **Texture**

WPI additions to 4.5% egg yolk samples significantly ( $P < 0.05$ ) affected values for hardness (N) which ranged between 3.27N and 18.40N whereas for the 9% egg yolk samples values ranged between 3.90N and 35.53N (Figure 1). Increasing WPI concentration up to

100% along with storage time (in 4.5% egg yolk) increased the value of hardness. In addition, adding WPI at 100% level in 9% egg yolk samples caused a sudden and dramatic decrease in the hardness (N); this result was not expected and the explanation for it is unclear. Denatured whey proteins interact with  $\kappa$ -casein on the surface of casein micelles, cross-linked casein and whey proteins. There is a casein-casein attraction when the heat of milk increased above 85°C during ice cream manufacturing, which results in the gelation of milk as caseins approach their isoelectric point. Above 85°C, those denatured proteins aggregated and gave rise to increased viscosity and hardness (DeWit 1990; Li-Chan, 2004). Damodaran (2007) noted that the protein gel is responsible for formation of a three-dimensional network of aggregated protein molecules.

Viscosity is another factor having an effect on ice cream texture. As found in this study (results not shown), increasing samples viscosity was resulting firmer texture. The mass effect of the large amount of fat becomes evident and the viscosity increases (Damodaran 2007; Leighton *et al.* 1934). Li *et al.* (1997) and Prindiville *et al.* (1999) reported that a significant increase in viscosity with a higher fat content was noted in their studies. The fat network also affects hardness. In this study, a significant increase in hardness with a high fat content was obtained. According to Alfaifi & Stathopoulos (2009), a significant increase in hardness with higher fat content was observed and recorded in their study. Roland *et al.* (1999*a; b*) demonstrated that the textural attributes showed significant changes at each percentage of fat but no differences in the texture attributes between the samples containing 0.1 and 3% fat. However, Prindiville *et al.* (1999) reported that a significant difference in hardness among treatments and among batches within treatments. In particular, full fat ice cream (9% fat) was significantly harder than all other ice cream (0.5, 4 and 6% fat). Tharp *et al.* (1998) found that the hardness of ice cream increased as the level of destabilised fat increased.

Gelato ice cream with increased whey protein had ice crystals (results not shown) that were having smaller size and greater number in 100% WPI (within 9% egg yolk), compared with 50% WPI (9% egg yolk). This might be because whey proteins have good water-binding capacity, hence, less free water is available to form ice crystals, possibly resulting in the formation of smaller size and lesser number of ice crystals in ice cream. Recently, ice structuring proteins (from arctic fish or winter wheat) have been used to inhibit ice crystal growth and maintain uniformly small ice crystals in ice cream (Marshall *et al.*, 2003; Hartel 1996).

### **Melting rate**

The changes in the stability (i.e. increase or decrease) of Gelato ice cream during four weeks storage at  $-17\pm 1^{\circ}\text{C}$  are shown in Figure 2. In general, increasing WPI levels resulted in a significant effect ( $P < 0.05$ ) in the melt volume among 9% and 4.5% egg yolk samples over increasing storage time. The only exception is among the samples after one and three weeks of storage using 4.5% egg yolk. The control Gelato ice cream containing 9% egg yolk melted much more rapidly than other samples. On the other hand, the control samples containing 4.5% egg yolk melted at a similar rate as other Gelato ice cream samples among the same level. It appears that the action of the 9% egg yolk fat content in ice cream may be quite different from that of the 4.5% egg yolk fat content. The fat contributes greatly to the structure of ice cream during the freezing process by forming a partially coalesced, three-dimensional network of homogenised globules that, along with air bubbles and ice crystals, are responsible for stiffness and aridness in the Gelato ice cream (Muse & Hartel, 2004). This also contributes to melt-resistance and smoother texture in the frozen Gelato ice cream (Bolliger *et al.* 2000; Tharp *et al.* 1998). Our results showed that, in most cases, Gelato ice cream samples containing higher WPI levels, retained the shape of the “remaining portion”

(that which stayed above the screen) better than those having less WPI added. This finding is in agreement with other researchers (Tosi *et al.* 2007; Wit 1998) who found that whey proteins, especially WPI, have become the most employed, functional food proteins in food formulations because of their suitability to induce and stabilise aerated food products such as ice cream. This property is usually identified with the foaming ability of whey protein products in aqueous solution.

### **Colour**

As shown in Table 2, mean colour (whiteness-*L*) ratings for ice cream containing 0% (control), 20%, 50%, 80%, or 100% replacement of egg yolk with WPI did not significantly ( $P>0.05$ ) differ from the control sample during the storage time. As the fat content of ice cream samples increased, the amount of yellowness (*b*) also increased. In addition, ice cream samples *b* values decreased with increased storage period, especially among 9% egg yolk samples. However, Gelato ice cream with added WPI in 9% egg yolk samples had slightly higher *L* and *b* values than the 4.5% egg yolk samples. This could be because of the fat content and inherent yellow colour derived from egg yolk powder. This finding was in agreement with Ronald *et al.* (1990*a*; *b*) who noticed that the colour of the ice cream increased in whiteness (*L*) as the fat content increased. Moreover, the colour of Gelato ice cream increased in yellowness (*b*) as the fat content of the samples increased, which corresponded that the colour of ice cream being more yellow, especially with 9% egg yolk control sample.

### **CONCLUSION**

The results obtained in this substitution study showed that formulations based on using 4.5% and 9% egg yolk with four levels of WPI are suitable for manufacturing Gelato

ice cream. In general, a significant improvement was observed in the texture and stability (melting rate) characteristics among 9% and 4.5% egg yolk samples with increased storage time. Moreover, added WPI in 9% egg yolk samples had slightly higher *L* and *b* values than 4.5% egg yolk samples. A strong fat content effect was observed in all physical properties measured. The study of the egg yolk substitution has demonstrated that replacement by WPI can prove advantageous in commercial applications since this widely available and cheap ingredient can be used in Gelato ice cream manufacture without a compromise to the functional properties of the product.

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#### REFERENCES

- Alfaihi M and Stathopoulos C (2009) Effect of egg yolk substitution by sweet whey protein isolate (WPI), on physical properties of Gelato vanilla ice cream. *Milchwissenschaft* (In Press)
- Bolliger S, Goff H and Tharp B (2000) Correlation between colloidal properties of ice cream mix and ice cream. *International Dairy Journal* **10** 303-309
- Damerow G (1995) Frozen custard. In *Ice cream! The whole scoop*. (Eds Damerow G) pp. 90-113. Aurora, USA: Glenbridge Publishing Ltd
- Damodaran S (2007) Inhibition of ice crystal growth in ice cream mix by gelatin Hydrolysate. *Journal of Agriculture and Food Chemistry* **55** 10918-10923
- DeWit J (1990) Thermal stability and functionality of whey proteins. *Journal of Dairy Science* **73** 3602-3612
- Eissa A, Bisram S and Khan S (2004) Polymerization and gelation of whey protein isolates at

- low pH using transglutaminase enzyme. *Journal of Agriculture and Food Chemistry* **52** 4456-4464
- Goff H (2003) Ice cream. In *Advanced dairy chemistry*, Vol.1, *Proteins* (Eds Fox P & McSweeney P) pp. 1064-1067. New York, USA: Aspen Publishers
- Guilmineau F and Kulozik U (2006a) Impact of a thermal treatment on the emulsifying properties of egg yolk. I: Effect of the heating time. *Journal of Food Hydrocolloids* **20** 1105-1113
- Guilmineau F and Kulozik U (2006b) Impact of a thermal treatment on the emulsifying properties of egg yolk. II: Effect of the environmental conditions. *Journal of Food Hydrocolloids* **20** 1114-1123
- Hartel R (1996) Ice crystallization during the manufacture of ice cream. *Trends in Food Science Technology* **7** 315-321
- Hatta H, Kapoor M and Juneja L (2008) Bioactive components in egg yolk. In *Egg bioscience and biotechnology*, (Eds Mine Y) pp. 185-239. New York, USA: J Wiley & Sons, Inc
- International Dairy Foods Association (2005) What's in the ice cream aisle? Definitions of frozen dessert products, 1 p. <http://www.idfa.org/facts/icmonth/page5>. Accessed 19 March 2009
- Leighton A, Leviton A and Williams O (1934) The apparent viscosity of ice cream: I. The sagging beam method of measurement. II. Factors to be controlled. III. The effects of milk fat, gelatin and homogenization temperature. *Journal of Dairy Science* **17** 639-650
- Li Z, Marshall R, Heymann H and Fernando L (1997) Effect of milk fat content on flavor perception of vanilla ice cream. *Journal of Dairy Science* **80** 3133-3141
- Li-Chan E (2004) Properties of proteins in food systems: an introduction. In *Proteins in food processing*, (Eds Yada R) pp. 4-12. Cambridge, UK: Woodhead Publishing Limited and CRC Press LLC
- Lim S, Swanson B, Ross C and Clark S (2008) High hydrostatic pressure modification of whey protein concentrate for improved functional properties. *Journal of Dairy Science* **91** 1308-1316
- Marshall R, Goff H and Hartel R (2003) Sherbets, sorbets and ices. In *Ice Cream*. Vol. 6 (Eds Marshall R, Goff H & Hartel R) pp. 265-273. New York, USA: Plenum Publishers.
- Morr C (1985) Functionality of heated milk proteins in dairy and related foods. *Journal of Dairy Science* **68** 2773-2781

- Morr C (1989) Beneficial and adverse effects of water-protein interactions in selected dairy products. *Journal of Dairy Science* **72** 575-580
- Muse M and Hartel R (2004) Ice cream structural elements that affect melting rate and hardness. *Journal of Dairy Science* **87** 1-10
- Ohmes R, Marshall R and Heymann H (1998) Sensory and physical properties of ice cream containing milk fat or fat replacers. *Journal of Dairy Science* **81** 1222-1228
- Prindiville E, Marshall R and Heymann H (1999) Effect of milk fat on the sensory properties of chocolate ice cream. *Journal of Dairy Science* **82** 1425-1432
- Ronald A, Phillips L and Boor K (1999a) Effects of fat content on the sensory properties, melting, color and hardness of ice cream. *Journal of Dairy Science* **82** 32-38
- Ronald A, Phillips L and Boor K (1999b) Effects of fat replacers on the sensory properties, color, melting, and hardness of ice cream. *Journal of Dairy Science* **82** 2094-2100
- Specter S and Setser C (1994) Sensory and physical properties of a reduced-calorie frozen dessert system made with milk fat and sucrose substitutes. *Journal of Dairy Science* **77** 708-717
- Szczesniak A (2002) Texture is a sensory property. *Journal of Food Quality and Preference* **13** 215-225
- Tharp B, Forrest B, Swan C, Dunning L and Hilmoe M (1998) Basic factors affecting ice cream meltdown. In: International Symposium on Ice Cream. Athens, Greece, pp 54-64
- Tosi E, Canna L, Lucero H and Ré E (2007) Foaming properties of sweet whey solutions as modified by thermal treatment. *Journal of Food Chemistry* **100** 794-799
- Tunick M (2008) Whey protein production and utilization: A brief history. In *Whey processing, functionality and health benefits*, (Eds Onwulata C & Huth P) pp. 1-10. USA: Wiley-Blackwell
- United States Dairy Export Council (2001) Industry overview, 1p. <http://www.usdec.org>
- Wit J (1998) Nutritional and functional characteristics of whey proteins in food products. *Journal of Dairy Science* **81** 597-608
- Zhu H and Damodaran S (1994) Heat-induced conformational changes in whey protein isolate and its relation to foaming properties. *Journal of Agriculture and Food Chemistry* **42** 846-855

Figure 1

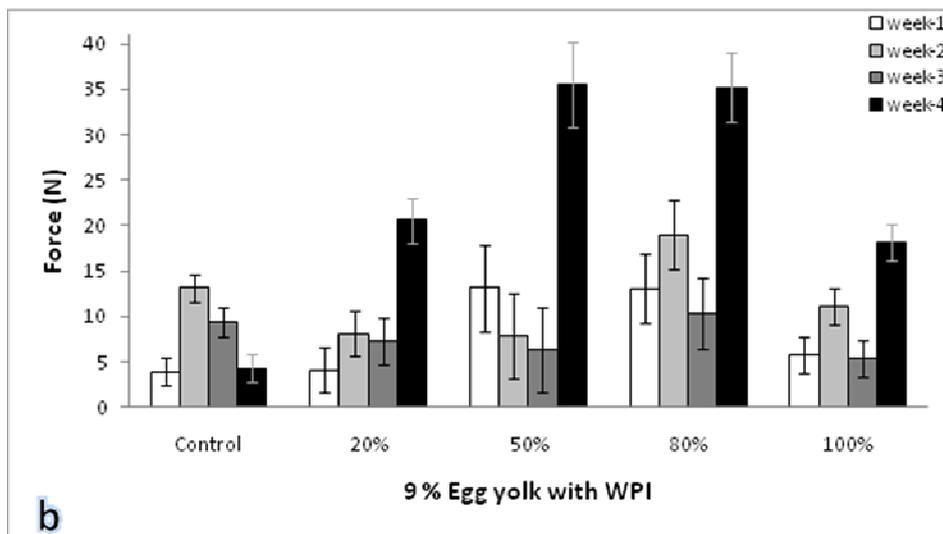
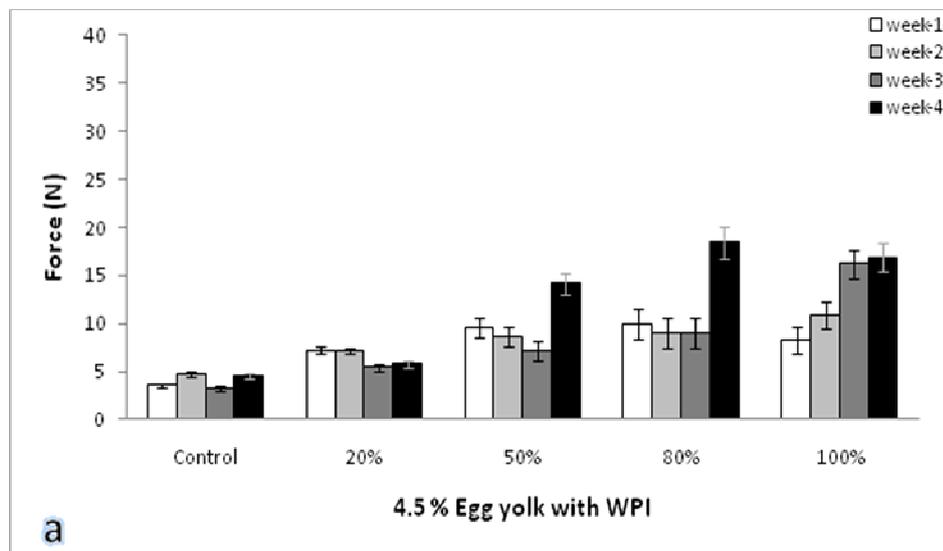
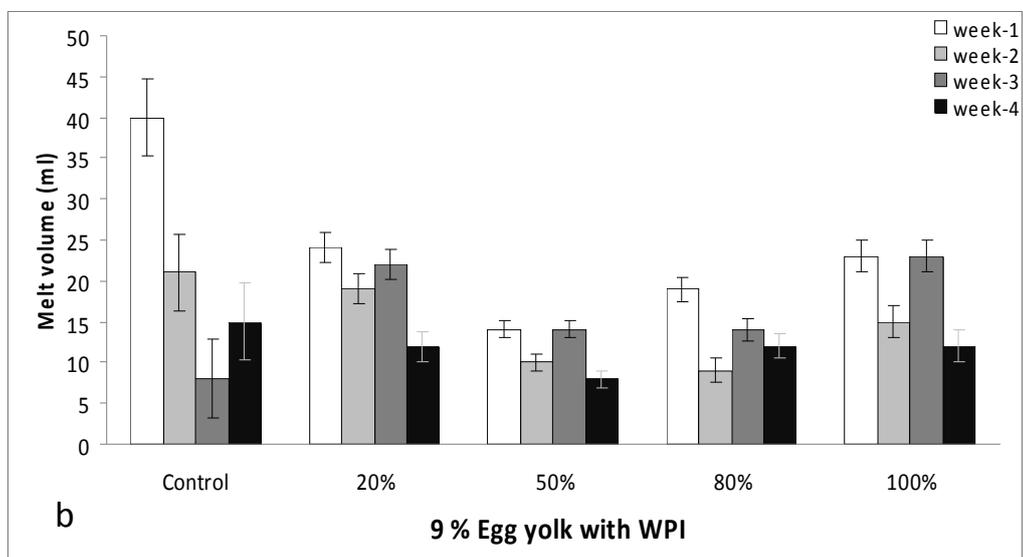
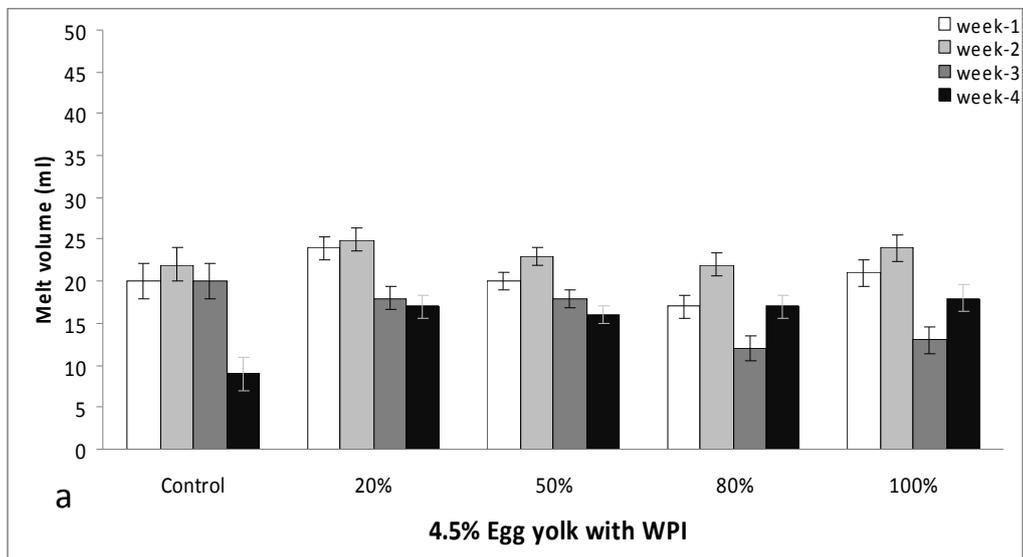


Figure 2



**Table 1:** Gelato-style Ice Cream formulations

Ingredients (g)	4.5% Egg yolk					9% Egg yolk				
	Control*	20%	50%	80%	100%	Control*	20%	50%	80%	100%
Whole Milk	57.13	57.13	57.13	59.13	60.00	51.60	53.97	56.86	54.35	54.71
Cream	14.29	14.29	14.29	14.29	14.29	14.84	14.47	14.14	16.65	16.29
Sugar	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00	20.00
Dried egg yolk	4.50	3.60	2.25	0.90	-	9.00	7.20	4.50	1.80	-
SMP**	4.08	4.08	4.08	2.08	1.21	4.56	2.56	-	-	-
WPI***	-	0.90	2.25	3.60	4.50	-	1.80	4.50	7.20	9.00

\*Control mix had no whey protein isolate, \*\* SMP (Skim Milk Powder), \*\*\*WPI (Whey Protein Isolate).

**Table 2:** Mean *L* and *b* values of Gelato ice cream with different levels of WPI substitution of egg yolk over storage†.

WPI (%)	Week 1		Week 2		Week 3		Week 4	
	<i>L</i>	<i>b</i>	<i>L</i>	<i>b</i>	<i>L</i>	<i>b</i>	<i>L</i>	<i>b</i>
<b>4.5% egg yolk</b>								
0% (control)	42.72	12.31	47.53	9.12	50.98	9.54	53.18	9.68
20%	48.01	10.22	49.92	12.72	52.88	9.35	56.29	9.83
50%	46.70	11.07	49.49	11.31	51.83	9.33	52.10	11.52
80%	45.19	9.90	52.42	10.32	52.23	9.14	51.91	10.90
100%	45.13	8.38	52.58	8.85	48.49	8.84	54.39	9.36
<b>9% egg yolk</b>								
0% (control)	51.72	10.82	51.37	10.18	54.15	11.92	50.91	16.87
20%	49.56	9.00	51.99	9.13	53.60	11.08	51.99	15.92
50%	38.87	10.15	48.36	12.98	52.67	14.45	52.03	14.49
80%	45.48	10.16	50.34	10.84	52.77	12.22	50.83	12.88
100%	46.29	8.84	49.51	9.68	48.84	10.11	49.96	10.38

† Values represent the mean of 3 replicate trials, **WPI-** *Whey protein isolate*,

**L-** *Measure of whiteness*, **b-** *measure of yellowness*.

# Effect of egg yolk substitution by sweet Whey Protein Isolate on texture, stability and colour of Gelato-style vanilla ice cream

## **Abstract**

Gelato samples were made with two levels of egg-yolk (4.5 and 9%). For each level, samples with and without Whey Protein Isolate (WPI) were prepared. Texture and melting rate values were significantly ( $P < 0.05$ ) higher in 9% egg-yolk samples over the storage period than in samples containing 4.5%. Increasing WPI led to a significant increase in hardness at both levels of egg-yolk. Gelato with added WPI in 9% samples had slightly higher whiteness ( $L$ ) and yellowness ( $b$ ) values than 4.5% samples. A significant effect of fat content was observed in all physical properties measured. Results suggest that this approach can deliver functionality at a lower cost and can produce a quality Gelato.

**Keywords:** Gelato, WPI, egg yolk, texture, ice-cream, colour

Fig.1. Comparison of hardness (N) values in Gelato-style ice creams, stored for four weeks, with different levels of WPI substitutions using (a) 4.5% and (b) 9% egg yolk. Values are the means of three replicates.

Fig. 2. Effect of WPI substitution on the melt volume (ml) of Gelato-style ice creams, stored for four weeks, with different levels of WPI substitutions using (a) 4.5% and (b) 9% egg yolk. Values are the means of three replicates.