Developing formulation of ice cream utilizing preserved whey protein concentrate and characterizing their physico-chemical traits

Abstract

Whey protein concentrate is a source for a number of high-quality proteins, carbohydrates, minerals and water-soluble vitamins and presence of all these components makes it a highly nutritious product. The composition of whey varies with the type of whey, pH and pre-heat treatment given to whey; Whey proteins are highly nutritious food supplements having therapeutic benefits too. The biological components of whey include Lactoferrin, β-Lactoglobulin, α-Lactalbumin, glycomacropeptides and Immunoglobulin. Ice cream is a frozen product obtained from milk along with required milk products, cane sugar, stabilizers, emulsifiers and edible flavours and colours. The present study pertains to characterizing the ice cream incorporating whey protein concentrate for their physico-chemical properties.

Keywords: Whey proteins, Physico-chemical properties, Ice cream, hurdle technology, preservation

Introduction:

Whey solids are well known for their nutritional properties and functional traits and these aspects are well documented (Muvihill, 1991; Mann, 1998; Jayaprakasha et al., 1999). Owing to their excellent nutritional and functional properties (viz., water binding, emulsifying, aeration, etc.) of whey solids they find numerous applications in food and dairy industry (Hufmann, 1996; Mann, 1998 Jayaprakasha *et al.*, 2005). The discharge of whey without treatment leads to environmental pollution in terms of public. Strict and stringent regulations imposed by pollution control board and also by the environmental agencies made scientists to look for the better ways of utilizing whey in human chain.

Whey represents about 80 to 90 per cent of original volume of milk used for preparation of cheese, paneer, chhana etc. In India, the major source of whey is from the production of chhana and paneer. In the absence of systematic survey/statistics the estimated

production of whey is about 5 million tons per annum (Gupta, 2008) is estimated to be more than 3 million tonnes which accounts to about 12 lakh tones of valuable milk solids in terms of nutrients (Khamrui and Rajorhia, 1998; Kim et al., 1987 Dwell more on whey concentrate, whey contains just 1-1.5% protein while Whey concentrate can contain proteins ranging from 5.0 to 6.5 % or so.

An interesting possibility for the utilization of whey for human nutrition is the hydrolysis of lactose into glucose and galactose (Renner and Abd El- salam, 1991). Hydrolysis generally improves desirability (Geilman, 1993), by increased osmotic pressure, easier digestibility, increased solubility, increased sweetness and enhances fermentability (Cotton, 1980, Arndt and Wehling, 1989; Timmermans, 1997).

Whey contains valuable lactose and proteins. Almost about 80 per cent of whey is wasted without being used in country, whey based drinks, Ricotta cheese etc. are produced to salvage whey). Whey carries low total solids 6.5% (specify TS) and has very low shelf life.

Hurdle in food is defined as the use of substances or processing parameters or various preservation factors, inhibiting the growth or causing death of various microorganisms Hurdle concept in food context was first introduced by Leistner (1994) and according to such concept each preservation parameter is termed as hurdles. Thus, the parameters like water activity (a_w), pH, redox potential, heat treatment, etc, which maybe bacteriostatic or bactericidal are hurdles. These hurdles may govern many preservation processes and more than one hurdle is often responsible for long shelf life of some specific food products (Leistner, 1994). Whey concentrate was preserved through combination of lowering the water activity (0.92-0.94%), decreasing the pH (i.e. to 5.2) of such media and incorporating potassium sorbate (0.2%), to achieve a storage life of about 3 months 30 ± 1 ^{o}C

The most delightful way to consume dairy solids and all the goodness of it is certainly through ice cream delicacies. It is not only a glamorous food but also a delicate, delicious, palatable and highly nutritious food. Although ice cream and frozen desserts

market in India is witnessing a respectable growth in recent years as a result of innovations in functional ingredients, technology, distribution and communication, yet the per capita consumption of ice cream in India is one of the lowest (i.e. 0.25 L) in the world (Ravindra, 2004).

Ice cream is one of the fastest growing dairy products with a growth rate of 25-35 per cent Now a days there is great demand for low-cost ingredients which provide desired functional properties. Whey concentrate is one of the best low cost ingredient to meet better functional properties alternative and can certainly meet (functional properties) requirements Replacement of milk solids with such byproduct, in concentrated form, in ice cream production not only can reduce the cost of production but also improve its functional properties such as viscosity, good emulsifying, gelation abilities, whipping ability, melting resistance, and nutritional value.

Whey solids have been used to replace part of the conventional MSNF sources in several varieties of ice cream formulations recommended the use of dried whey at usage rate of 30-40 per cent of MSNF, without adversely affecting the organoleptic quality of ice cream. Steinsholt (1974) successfully replaced 20 per cent of usual MSNF source skim milk powder in ice cream mix with whey solids without any adverse effect on its sensory quality. Lactose, if added in excess concentration causes sandiness in ice cream; however this defect can be tackled With the increasing duration of hydrolysis process, the degree of hydrolysis increases Mjalo et al., 2005; Hnitsevych et al., 2017. Hydrolyzing the lactose present in whey (Arbuckle, 1986 – use some recent Ref.; see Ref. of Osmak et al. 2021- there are more too!).

Therefore, it was envisaged to develop formulation of ice cream mix utilizing whey concentrate in which lactose was pre-hydrolyzed and the concentrate preserved through use of hurdle technique. The process of preparing such ice cream was standardized and the products keeping quality was monitored during storage.

Materials and methods:

Materials:

The following materials were used in the study for the preparation of ice cream utilizing lactose hydrolysed whey concentrate.

Whey

Fresh cheese whey after rennet curd cutting and paneer whey used Paneer whey was procured from Students experimental dairy plant, Dairy Science College, Hebbal, Bangalore.

Enzyme

Lactozyme (β-galactosidase) was supplied by Novozyemes south Asia Pvt Ltd, Bangalore.

Citric acid

Food grade quality citric acid procured from the local market was used for preparation of paneer and for pH adjustment of paneer whey.

Whole milk

Fresh cow milk procured from dairy farm of Sri Venkateswara Veterinary University. Student Experimental Dairy Plant, Dairy Technology Dept at Dairy Science College, KVAFSU was used in preparing ice cream.

Cream

Fresh cream obtained by separating the cow milk in cream separator was used for ice cream mix preparation.

Skim milk powder

“Sangam Dairy” brand skim milk powder manufactured by Sangam Milk Producer Company Limited, Guntur District, Andhra Pradesh - 522213. Karnataka Milk Federation was procured from the local market (deleted last part).

Stabilizer and Emulsifier

Stamulcol ULV-1000 procured from Lucid Colloids Ltd., Jodhpur was a blend of stabilizer and emulsifier.

Flavour

Bush brand synthetic vanilla flavour was procured from local market

Sugar

Good quality cane sugar was purchased from the local market

Utensils and Glassware

Stainless steel grade of vessels of varying capacities and stirrers were used at various stages of the investigation. Borosil make of conical flasks, beakers, volumetric flasks, measuring jars were used for chemical and microbiological analysis.

Evaporator

Single effect, rising film vacuum operated evaporator (Anhydro, Copenhagen, Denmark) of 35 kg water evaporation/h capacity was used.

Analytical methods

Fat

Fat content of milk and Ice cream mix was determined by Gerber method as per BIS (1981).

Milk Solids-Not-Fat (MSNF)

MSNF content of milk was measured using ISI certified Zeal Lactometer by using the following formula

$$\textbf{SNF (\%) = 0.25 CLR+0.2 F +0.48}$$

Where, CLR-Corrected Lactometer Reading

F-Fat content in per cent

Total Solids (TS)

Total solids of milk and Ice cream were determined by gravimetric method as per BIS (1981).

Protein

Nitrogen content of Ice cream was determined by Micro-Kjeldahl method and protein content was computed by multiplying nitrogen value with 6.38 (BIS, 1981).

Moisture

Moisture content of dried products was determined according BIS (1981). Why only dried products – you should find TS of unconcentrated paneer whey, cheese whey, etc.

Ash

The ash content was determined following the standard procedure (BIS, 1981).

Lactose

Lactose content was determined by Lane-Eynon method.

Physico-chemical properties

The methods employed to analyze the physico-chemical attributes of milk, cream, and ice cream (why whey concentrate was left out?) are described below.

pH

pH of the whey sample (which sample – whey, milk, ice cream?) was measured using a digital pH meter. (Elico Pvt. Ltd.) at 25°C. About 20 ml of representative samples were used for pH determination.

Titratable acidity

Acidity was measured as per standard method (BIS, 1981).

Specific gravity

Specific gravity of **lactose hydrolyzed whey, lactose hydrolyzed sweet concentrate whey and ice cream** samples was measured at 20°C by using a Standard specific gravity bottle of 25 ml capacity by taking distilled water as a standard liquid.

Viscosity

Viscosity of cheese whey, whey concentrate, ice-cream mix and ice cream samples was measured using Falling Ball Viscometer at 20°C (Gilmont Instruments, USA). The temperature of the product was maintained at 20°C in water bath using cold water. The measuring tube was filled with the samples and the ball was inserted through the open end of the tube. Depending upon the falling time of the ball and the expected viscosity, different balls were used. The time taken by the ball to fall through a distance of 100 mm was measured accurately using $^{1}/_{10}$ sec stopwatch. The viscosity was calculated by using the following formula.

$$\mu = K (\rho t - \rho) t$$

Where, **μ**= Viscosity in (cP)

t= falling time of ball (min)

ρt = Density of the ball (g/ml)

ρ= Density of liquid at measuring temperature (g/ml)

K= Ball constant (0.3)

Melting resistance

Melting time of ice cream was observed by weighing 100 ± 1 g of sample (100 g of sample is too small quantity for such testing – wrongly followed procedure!) onto a wire gauge placed on a funnel over a beaker. The time taken by the sample to completely meltdown and fall into a controlled temperature chamber at 20°C was recorded and expressed as melting time in min (Arbukle, 1986).

Melting time: The heat shock resistance of the ice cream was determined by using a self-manufactured system. The melting time of one hundred grams of ice cream kept at 20ºC to melt completely in the chamber was measured. The initial temperature of the ice cream was set to be -70ºC. The onset melting time of ice cream was initiated at the point of weight change. The weight of the ice cream was determined every 1 min.

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Whipping rate

During freezing of ice cream at a regular interval of 2 min a standard volume of ice cream was drawn and the overrun was calculated. The time taken by the mix to attain 90 per cent over run was computed to find out the rate of whipping.

Overrun

The overrun of ice cream was calculated on weight basis using a standard 50 ml, cup by using sensitive top pan balance. Calculations were made using the following formula.

$$\% \text{ Overrun} = \frac{A-B}{B} \times 100$$

Where A-weight/unit volume of Ice cream mix

B-weight/unit volume of Ice cream

Penetration value

Using a cone penetrometer (deleted 'Made by') Associate Instruments Manufactures India Ltd.) penetration value was determined after hardening (should have been at deep freeze temp -18^{o}C. of ice cream. The distance in millimetre by which the cone travels in 5 s (print 'second' as 's') of the sample was noted. For each sample reading were recorded at 3 different spots and the mean value was noted.

Freezing point

The freezing point of the ice cream mix was determined using a Cryoscope (Cryostar I 'Advanced Milk Instruments Manufacturer', USA).

Statistical analysis

Completely randomized design (CRD) with one way analysis of variance (ANOVA) and simple t-test were used for analysing the data generated in the experiment. The tests of significance were carried out as per the SPSS -10 statistical packages.

Results and Discussion:

Physico-chemical properties of cheese whey

The physico-chemical characteristics of the cheese whey used in the experiment are presented in Table 1 and 2.

Physical properties

Cheese whey had an average pH of 5.42 with a titratable acidity of 0.21 per cent lactic acid. The average values of specific gravity, viscosity (cP) and water activity (a_w) of cheese whey were 1.024, 1.069 centi poise and 0.995 respectively. The pH of cheese whey varied between 5.39 to 5.45.

Chemical composition of cheese whey

The cheese whey had 6.68 per cent TS (Table 2).

Table 1: Physical properties of cheese whey

Properties	Cheese whey	Range
pH	5.42	5.39-5.45
Acidity (% LA)	0.21	0.20-0.22
Specific gravity	1.024	1.021-1.027
Viscosity(cP)	1.069	1.066-1.072
Water activity(a_w)	0.995	0.992-0.998

Keep 1st column matter left aligned

Table 2: Chemical composition of cheese whey

Constituents	Average %	Range %
Total solids	6.68	6.58-6.78
Protein	0.83	0.80-0.86
Fat	0.35	0.33-0.37
Lactose	4.90	4.7-5.10
Ash	0.60	0.5-0.70

The TS was made up of 0.83 per cent protein, 0.35 per cent fat, 4.9 per cent lactose and 0.60 per cent ash.

Process optimization for the preparation of plain whey concentrate

Plain whey concentrate was prepared by condensing the fresh whey to different TS levels (40-55 per cent) in a falling film evaporator.

Effect of condensing whey on the physico-chemical properties of whey concentrate

Fresh cheese whey was condensed to TS levels (40, 45, 50 and 55 per cent). The physical properties of condensed whey prepared are depicted in Table-3. With the increase in concentration, the pH decreased from 4.98 to 3.84. The pH of whey was 4.98 at 16 per cent TS, whereas it was 3.97, 3.96, 3.88 and 3.84 at 40, 45, 50 and 55 per cent TS. The acidity tended to increase as the TS of whey concentrate was raised. At total solids 16 (control) per cent; the acidity was 0.613 per cent LA whereas at 40 per cent total solids it was 1.078 per cent LA.

The specific gravity of 16 (control) per cent TS whey concentrate was 1.057 whereas at 40 per cent TS it was 1.297. With an increase in the TS level there was a proportionate increase in the viscosity. The viscosity of whey concentrate 16 (control), and those condensed to 40, 45, 50 and 55 per cent TS was 1.24, 17.01, 19.08, 22.94 and 26.81 cP, respectively. Similarly, with an increase in the TS level there was a progressive decrease in the water activity of the whey concentrate. The water activity (a_w) of whey concentrates was 0.983, 0.942, 0.923, 0.901 and 0.886 at 16 (control), 40, 45, 50 and 55 per cent TS levels respectively.

The increase in the TS level as a result of further condensing was found to be statistically ($p<0.05$). The protein content at 16, 40, 45, 50 and 55 per cent TS of whey concentrate was 1.99, 4.98, 5.58, 6.21, 6.68 respectively and the result was found to be significant ($p<0.05$). The fat content of whey concentrate at 16, 40, 45, 50 and 55 per cent TS was (deleted 'found to be') 0.84, 2.10, 2.36, 2.62 and 2.88 respectively and the result was found to be significant ($p<0.05$).

The lactose at 16 per cent (control) TS of whey concentrate was 11.74 per cent whereas at 40 per cent it was 29.92 per cent. With an increase in the TS level of whey concentrate, there was a proportionate increase in the lactose content. The lactose content at whey concentrate TS of 16 (control), 40, 45, 50 and 55 per cent was 11.74, 29.92, 33.03, 36.68 and 40.34 per cent respectively. Similarly, with an increase in the TS level, there was a progressive increase in the ash content of whey concentrate; the values were 1.43, 3.58, 4.04, 4.48 and 4.94 per cent as noted at 16 (control) 40, 45, 50 and 55 per cent TS of whey concentrate respectively (Table no.3).

Table 3: Effect of degree of condensing on the physico-chemical properties of whey concentrate

Level of TS (%)	Physical attributes					Chemical Composition			
	pH	**Acidity (%LA)**	**Specific gravity**	**Viscosity (cP)**	**Water activity (a_w)**	**Protein (%)**	**Fat (%)**	**Lactose (%)**	**Ash (%)**
Control (16)	4.99	0.61	1.057	1.24	0.983	1.99	0.84	11.74	1.43
40	3.97	1.08	1.297	17.01	0.942	4.98	2.10	29.92	3.58
45	3.96	1.15	1.310	19.08	0.923	5.59	2.34	33.03	4.04
50	3.88	1.18	1.317	22.94	0.901	6.21	2.62	36.68	4.48
55	3.84	1.24	1.322	26.81	0.886	6.68	2.88	40.34	4.94
CD	0.036	0.032	0.006	0.17	0.004	0.017	0.064	0.058	0.018

*All values are average of three trials

(Made 2 digits after full-point for pH, Acidity, Viscosity values, protein and fat content, as applicable)

Effect of varying sugar levels and vacuum condensing on the physico-chemical properties of whey concentrate

Fresh Cheese whey added with an optimum level of sugar was subjected for condensing to best accepted level. The effect of level of condensing on various physical properties of condensed whey is depicted in Table-4. With the increase in concentration, the pH decreased from 4.987 to 3.77. The pH of whey was 4.987 at 0 (control) per cent sugar, whereas it was 3.99, 3.95, 3.89 and 3.77 at 10, 12, 14 and 16 per cent sugar levels respectively. In contrast to the pH, acidity increased as the sugar addition with condensing level increased. At sugar 0 per cent (control), the acidity was 0.613 per cent whereas at 16 per cent sugar it was 1.30 per cent.

The specific gravity at 0 (control) per cent TS was 1.057 whereas specific gravity of whey concentrate of 16 per cent TS was 1.325. With the increase in the total solids level there was proportionate increase in the viscosity. The viscosity of whey concentrate at 0 (control), 10, 12, 14 and 16 per cent sugar levels was 1.24, 17.02, 19.09, 22.33 and 27.52 cP respectively. Similarly, with the increase in the sugar level there was progressive decrease in the water activity of whey concentrate. The water activity (a_w) was 0.983, 0.925, 0.908, 0.882 and 0.879 at 0 (control), 10, 12, 14, and 16 per cent sugar levels.

Table 4: Effect of varying sugar levels and vacuum condensing on the physico-chemical properties of whey concentrate

Sugar level (%)	Physical attributes				
	pH	Acidity (% LA)	Specific gravity	Viscosity (cP)	Water activity (a_w)
Control	4.99	0.613	1.057	1.24	0.983
10	3.99	1.076	1.312	17.02	0.925
12	3.95	1.163	1.315	19.09	0.908
14	3.89	1.191	1.321	22.33	0.882
16	3.77	1.30	1.325	27.52	0.879
CD	0.089	0.084	0.007	0.0178	0.009

*All values are average of three trials

Effect of varying sugar levels and vacuum condensing on chemical composition of whey concentrate

optimized TS level of whey concentrate. The increase in the TS content of condensed whey was commensurate with the level of addition of sugar; such effect was found to be significant ($p<0.05$) (Table 5). With an increase in the level of sugar in whey concentrate, there was a progressive decrease in the protein content. The protein content of control (and whey concentrate having 10, 12, 14 and 16 per cent sugar was 1.99, 4.98, 4.72, 4.33 and 4.15 respectively; such effect was significant ($p<0.05$). The fat content of control and those containing sugar at levels of 10, 12, 14 and 16 per cent was 0.84, 2.08, 2.06, 2.03 and 2.01 per cent respectively; such effect was also found to be significant ($p<0.05$).

The lactose content of control ice cream was 11.74 per cent, whereas at 16 per cent sugar level the lactose content was 35.04 per cent. With an increase in the sugar level, there was a proportionate decrease in the lactose content of whey concentrate. The total carbohydrates content for control and those containing added sugar at levels of 10, 12, 14, and 16 per cent was 11.74, 29.92, 32.33, 33.56 and 35.04 per cent respectively. With increasing level of sugar addition, there was a progressive decrease in the ash content of whey concentrate (rather the ash content should rise, since sugar also has ash in it!). The ash content was 1.43, 3.58, 3.45, 3.37 and 3.22 of control as well as of experimental samples containing sugar levels of 10, 12, 14 and 16 per cent respectively.

Preparation of lactose hydrolysed sweetened concentrated whey

Whey concentrated (in concentrated or unconcentrated form (be specific) – why sugar level was not reduced, since lactose hydrolysis makes such lactose-hydrolyzed whey sweeter than its untreated counterpart!!) was subjected to lactose hydrolysis. The enzyme concentration attempted was such that about 80 per cent lactose hydrolysis was feasible, maintaining conditions such as pH 6.5 and 4hrs of incubation at 37 °C as recommended by the supplier of the enzyme.

Table 5: Effect of addition of varying sugar levels and vacuum condensing on the chemical composition of whey concentrate

Level of sugar addition (%)	Chemical Composition (%)			
	Protein	Fat	Lactose	Ash
Control (0)	1.99	0.84	11.74	1.43
10	4.976	2.08	29.92	3.58
12	4.72	2.06	32.33	3.45
14	4.33	2.03	33.56	3.37
16	4.15	2.01	35.04	3.22

CD	0.01	0.18	0.02	0.017

*All values are average of three trials

The physical properties of whey used in this investigation are represented in Table-l. The physical properties of cheese whey such as pH, acidity, specific gravity, Viscosity (cP) and water activity (a_w) were within the range of results reported by Jayaprakasha. The cheese whey used in this investigation had an average pH of 5.42 and its titratable acidity was 0.21 % LA. The specific gravity, viscosity and water activity ($a_{w)}$ of whey was found to be 1.024, 1.069 and 0995 cP, respectively which were quite similar to that reported by Jayaprakasha (1992).

Table-2 represents the chemical composition of cheese whey used in this investigation. Whey had an average TS content of 6.68 per cent which comprised of 0.83 per cent protein, 0.35 per cent fat, 4.9 per cent lactose and 0.60 per cent ash.

Whey concentrate was prepared by condensing whey varying TS levels in combination with use of potassium sorbate at 0.2 per cent followed by sweetening the whey concentrate with sugar. The results pertaining to these are presented in Table 3, 4 and 5 respectively.

From among the various levels of sugar tried, addition of 10.0 per cent sugar to whey was found to be optimum in combination with use of 0.10 per cent potassium sorbate. The level of optimum sugar led to resultant ice cream having desired sweetness intensity.

Fresh whey was subjected for condensing to various levels of total solids. As the level of condensing increased from the initial 16 per cent to 55 per cent, there was proportionate decrease in pH and increase in titratable acidity which are mainly due to the concentration of the components which are responsible for pH and acidity. The increase in concentration also resulted in increase in specific gravity and viscosity of the whey concentrate. Similarly, as the level of TS of whey concentrate increased there was significant ($p<0.05$) decrease in the water activity (Table-3) which can be mainly attributed to the increased soluble solutes such as added sucrose along with lactose and minerals. Water activity (a_w) of the whey concentrate was 0.886. The decrease in water activity (a_w) from 0.983 at 50 per cent total solids. The decrease in the water activity (a_w) of the whey concentrate as compared to initial whey was

ascribed to the effect of vacuum condensing. As a result of concentration have been reported by several workers and our results are in agreement with their reports (Jayaprakasha *et al.*, 1997). Lactose crystallization was observed when whey was condensed to 55 per cent TS. Considering such finding, 50 per cent TS of whey concentrate in the final product was found to be optimum in this investigation.

While increasing the level of concentration of whey from 16 to 55 per cent TS, there was a significant ($p<0.05$) increase in protein, lactose, fat and ash content of whey concentrate.

The increase in concentration also resulted in increase in specific gravity and viscosity of the whey concentrate. Similarly, as the level of concentration increased there was a significant ($p<0.05$) decrease in the water activity of the whey concentrate (Table-4) (The decrease in water activity (a_w) from 0.983 to 0.879 at 16 per cent added sugar led to increased storage stability of the condensed whey concentrate Lactose crystallization was observed when whey concentrate contained 16 per cent sugar. Therefore, 10 per cent level of sugar in preparing whey concentrate was found optimum.

Similarly, as the level of addition of sugar increased there was significant increase in the total carbohydrate and there was slight decrease in ash content of the whey concentrate (Table-5).

Conclusion

In this investigation a technology comprised of what – lactose hydrolysis, vacuum concentration of whey to specific TS level, addition of sugar, use of preservative, etc was developed for the efficient utilization of whey for the preparation of ice cream. The formulated ice cream not only helps in increasing the shelf life of whey but also helps in preserving whey concentrate in their natural form. The ice cream prepared utilizing such sweetened whey concentrate increased the mix viscosity resulting in smoother and refreshing nature they can provide potential profit margins which in turn add to the growth and development of dairy industry.

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