International Journal of Food Science and Nutrition
ISSN: 2455-4898; Impact Factor: RJIF 5.14
Received: 01-04-2020; Accepted: 19-04-2020; Published: 03-05-2020
www.foodsciencejournal.com
Volume 5; Issue 3; 2020; Page No. 08-16

Development of novel commercially viable stirred yogurt with prolonged shelf life incorporating osmotically dehydrated *Aloe vera* (*Aloe barbadensis Miller*) gel

DDTT Darshana Senarathna[1*], Vithushana Thangarajah[2], KHD Namal Abeysooriya[3]
[1]Department of Aquaculture and Fisheries, Faculty of Livestock Fisheries and Nutrition, Wayamba University of Sri Lanka, Makandura, Gonawila, Sri Lanka
[2]Department of Livestock and Avian Science, Faculty of Livestock Fisheries and Nutrition, Wayamba University of Sri Lanka, Makandura, Gonawila, Sri Lanka
[3] Department of Chemistry, Faculty of Science, University of Peradeniya, Peradeniya, Sri Lanka

Abstract
The present study investigated the possibility of using osmotically dehydrated *Aloe vera* gel as an additive to increase the nutritional quality and shelf life of stirred yoghurt. First, the extracted *Aloe vera* gel was osmotically dehydrated (OD) and pasteurized. Then it was added into yoghurt in three levels (5%, 7.5%, and 10%) and stirred. The best level of OD gel was selected through sensory evaluation and curd strength, and physiochemical properties, proximate composition, and microbial analysis were done to evaluate the quality of the final product for 35 days under 4°C. According to the results, the best OD Aloe gel percentage was 5% and all the physiochemical parameters were significantly ($P<0.05$) higher than the controlled one at an acceptable level. Microbial analysis proved that the product has a strong antimicrobial activity towards yeast and mold growth and coliform increasing the shelf life of the product.

Keywords: aloe vera, stirred yoghurt, osmotic dehydration

1. Introduction
Cow milk is considered to be the most popular type of milk consumed by the human due to its favorable nutrient composition than the other type of mammalian milk. Although there are some differences in the level of protein, mineral and fatty acids from the human milk, still it is found to be that cow milk is suitable not only for adults but also for infants [1]. However, there is an increasing demand for value-added cow milk products due to some reasons involving problems associated with gastrointestinal disorders, allergenicity for normal milk and sometimes as a means of treating gastrointestinal ulcers [2, 3]. The traditional type of Yoghurt has been known to mankind for over 6,000 years and different types of fermented milk products became popular with the dawn of civilization due to its countless health benefits [4, 5].
Yoghurt is considered to be a coagulated milk product that has been obtained from the fermentation of lactose into lactic acid in milk through the action of a series of bacteria such as *Lactobacillus acidophilus* (AL-5), *Bifidobacterium lactis* (Bb-12) and *Streptococcus thermophilus* with or without additions [6]. Recently, there is an increasing demand for dairy products enriched with such selective viable microorganisms or probiotic microorganisms as they strengthen and balance the beneficial bacteria in the gut providing health benefits, once administered into the body [7, 8]. According to the recent studies, consumption of yoghurt may cure some non-communicable diseases associated with the gastrointestinal tract such as enhancing lactose digestion by mal-digesters, lowering blood cholesterol, decreasing the risk of cancer improving immune response and helping the body to assimilate protein, calcium and iron, and diarrhea protection [9, 10, 11]. Therefore, yoghurt is categorized under "functional foods" class [8].
Currently, there are different types of yoghurt available in the market which were changed controlling the physical properties such as the heat treatment applied to the milk, amount and the type of the culture, additives, way of homogenization, coagulum and the type of stabilizers. [12]. Set yoghurt, stirred yoghurt and drinking yoghurt are the most common commercially available yoghurt products in the world while stirred yoghurt is the most popular [13]. To enhance the nutritional value, different types of flavorings, sweeteners, and additives can be added into the yoghurt while increasing the consumer acceptance rate. There is a high potential of using traditional medicinal herbs such as Aloe Vera as a resource of functional food and stabilizing agent with yoghurt industry. Undeniably, these herbs can play an important role in the treatment and prevention of many diseases while enhancing the value of the product.
Aloe Vera is considered to be a traditional medicinal plant that is currently used in food, pharmaceutical and cosmetic industries [14]. Aloe Vera has both external and internal benefits such as wound healing, skin moisturizing, healing skin burns, anti-inflammatory properties, protection from gastric ulcers, anti-cancer activity, anti-oxidant and anti-diabetic effects [15, 16, 17, 18]. Therefore, Aloe Vera is a potential candidate to use in yoghurt to enhance its nutritional and medicinal value.
In the yoghurt industry, gelatin is the most common hydrocolloid used as a thickening or stabilizing agent [19]. Gelatin is an ingredient compatible with the milk proteins and contributes good palatability and textural properties to the final product [20]. However, the requirement of a substitute for the gelatin was needed for many years for the vegetarians, halal and kosher markets due to some religious

and scientific reasons such as the spreading of bovine spongiform encephalopathy or "Mad Cow Disease" (MCD) [21]. Therefore, Aloe Vera gel with agar can be used as a potential alternative for gelatin in order to develop a Lacto – vegetarian yoghurt without significant changes of the organoleptic properties of gelatin added yoghurt [21, 22]. At the same time, Aloe Vera can be used as a potential natural preservative that can be used in dairy products due to its anti-fungal and anti-oxidant properties.

The objective of this study to develop a novel commercially viable Lacto - vegetarian stirred yogurt with prolonged shelf life incorporating osmotically dehydrated Aloe Vera gel and checking its viability to introduce to the global market.

2. Materials and Methods

2.1. Materials

The product development and the laboratory analysis of the milk, Aloe gel and yoghurt samples were carried out at the Food Preservation laboratory, Microbiology laboratory, Livestock laboratory, and Aquaculture laboratory in the Faculty of Livestock Fisheries and Nutrition at the Wayamba University of Sri Lanka. A. vera leaves were bought from a commercially cultivated Aloe vera farm in Jaffna, Sri Lanka. The starter culture of YoFlex Express 1.0 (CHR Honson, Denmark) was obtained from JL. Morison Son & Jones (Ceylon) PLC, Colombo, Sri Lanka. Agar-agar, vanilla, flavoring, coloring agents, plastic yoghurt cups and raw materials for preliminary trials were purchased from the Central Essence Pvt Lad, Kandy, Sri Lanka. Sugar and Skim milk powder were purchase from a supermarket in Kandy.

2.2. Preparation of the Aloe Vera gel

First, the A. vera leaves were washed to remove adhering extraneous materials. Then, lower one inch of the leaf base and spikes along leaf margins were removed. Leaves were washed using potable water after allowing for draining of yellowish latex for a few minutes. Outer green rinds were removed and then the resulting fleshy filet was cut into 1 cm2 cube using a stainless-steel knife. Aloe gel cubes were washed thoroughly in a beaker containing potable water for 5 min at gel to water 1: 6 at room temperature (28±1°C) to remove Aloin. This act was carried out two more times to removing the slime nature. Resulted A. vera gel cubes were subjected to osmosis. They were placed in an osmotic solution of 50°Brix at gel cubes to syrup ratio of l: 5 for 4 hours following the data of Pisalkar *et al*. (2014) [23]. Then the osmotic solutions were drained off and aloe gel cubes were recovered after osmosis. Resulted gel cubes were immediately rinsed with potable water and placed on dried muslin cloths to remove surface moisture. Finally, the gel cubes were well macerated until becoming homogeneous.

2.3. Preparation of Aloe Vera incorporated stirred yoghurt

Fresh cow milk (milk fat 3.04%, protein 3.60%, total solids 10.1% and pH 6.89) was used for yoghurt production. At first, yoghurt sample was homogenized at 200 bars for 30 minutes and pasteurized at 93°C for 3 minutes. Then sugar (9% w/v), coloring, agar-agar (0.3%) and vanilla flavor (0.1% w/v) were added and skim milk was added to adjusted the yoghurt percentage (75%) in the final product following the Sri Lankan standard for yoghurt (1989). Then the mixture was cooled until reaching to the temperature of 42°C and YoFlex Express 1.0 culture granule (CHR Honson, Denmark) containing lactic acid bacteria (0.004% w/v) were inoculated as a freeze-dried direct vat set (FD-DVS) culture into the yoghurt mix following the Incubation for 3 – 4 hours till the pH reach to 4.5. At this point, the yoghurt was stored in a refrigerator (4±1°C) overnight.

The prepared Aloe vera gel was pasteurized at 82°C for 2 minutes and cooled to 4°C, then the osmotically dehydrated liquid gel was added into the prepared yoghurt. The yoghurt samples were stirred well till get a homogeneous mixture and filled in 500 mL plastic cups along with the mixed Alo vera gel. Finally, the yoghurt samples were stored in the refrigerator for further quality evaluation. The yoghurt samples were analyzed at 1, 3, 5, 7, 10, 14, 18, 21, 28, 30- and 35-days interval. Three replicate yoghurts were made.

2.4. Determination of the aloe gel treatment levels (w/v)

Yoghurts were prepared incorporating 5%, 7.5%, 10%. 15%, 20% and 25% of aloe gel levels and 5%, 7.5% and 10%% aloe gel levels were selected as treatments through trial & error method.

2.5. Sensory evaluation

Sensory evaluation was done with 35 semi-trained panelists along with a Hedonic scale test. Sensory qualities such as color, aroma, appearance, consistency texture, taste, and overall acceptability were tested on a 9 point hedonic scale. Each sample was given a three-digit number and put in plastic cartons in a random order.

2.6. Curd strength

The curd strength was measured using an Instron Universal testing machine (Instron Corp., Canton, MA) to select the best percentage of Aloe vera gel to be added.

2.7. Proximate composition of Aloe Vera incorporated stirred yoghurt

The samples were mixed thoroughly and analyzed in triplicate for measuring the proximate composition such as crude fat, crude protein, crude fiber, ash, and dry matter content. The micro Kjeldahl method was used to determine the crude protein content of stirred yoghurt [24]. Crude Fat content was measured using the Gerber method [25], ash content by heating a 5g sample in a muffle furnace at 550°C for overnight [26], dry matter content was determined using a drying oven [26].

2.8. Physicochemical analysis of Aloe Vera incorporated stirred yoghurt

Titratable acidity, pH, viscosity and syneresis were measured as physiochemical parameters for the final product. Titratable acidity was measured according to the procedure of Yang and Li (2010) and pH was measured using the Hanna microprocessor pH meter. Syneresis was measured according to the method described by Wu *et al*. (2001) and viscosity was measured using the viscometer (BL model Tokimec, Japan).

2.9. Microbiological analysis of developed stirred yoghurt

Randomly selected few prepared Aloe vera yoghurt samples were diluted in a ratio of 1:10 (v/v) and used to analyze the total viable bacteria (TVB), coliform bacteria and yeasts and molds counts. TVB count was done using the plate count

agar medium and plates were incubated at 32ºC for 48 hr. Coliform bacteria count was conducted using the MacConkey agar medium and plates were incubated at 32ºC for 48 hr. Yeasts and molds counts were performed using the potato dextrose agar medium and the plates were incubated at 25ºC for 5 days. All the identification tests were done following the methods used by Barrow and Feltham (1993).

2.10. Data analysis

All the data collected from sensory evaluation and physicochemical evaluations were statistically analyzed. The sensory data were analyzed using the nonparametric Friedman procedure using the MINITAB software and all the measurements of physicochemical (keeping quality) parameters were made using three replications. The statistical design which was used to analyze the data is Completely Randomized Design (CRD). Data were analyzed using the SAS software package and means were compared using the Duncan's Multiple Range Test (DMRT).

3. Results and Discussion

Analysis of the Aloe vera gel and determination of the appropriate treatment levels

3.1. Compare the gel before and after dehydration

According to the data given in Table 1. Fresh Aloe vera gel contains a high amount of moisture almost 99% that may take responsibility for stimulating microbial growth leading to spoilage. Furthermore, after adding such a high amount of moisture into a dairy product its textural properties and shelf life also may reduce due to the less stability and the favorable nature for fungal growth. Therefore, it was a necessity to reduce the moisture level in the gel. Although there were a number of drying methods, osmotic dehydration (OD) [23] method was selected as there is not any physical damage to the gel which may cause to destroy some bioactive compounds in the gel. This caused to minimize the moisture content by 27.2%. The percent total soluble solid TSS has been increased after the osmotic dehydration process due to the increasing concentration of TSS and the gaining of sugar into gel cubes. There was no significant difference between the pH values before and after the dehydration process.

Table 1: Properties of fresh and osmotically dehydrated Aloe gel

Property	Fresh gel	Osmotically dehydrated gel
Moisture Content (%)	99.09±0.03	71.89±0.08
TSS (%)	01.08±0.02	14.47±0.06
pH	4.88±0.26	4.55±0.03

Data are presented as mean ± SD of triplicates

3.2. Sensory evaluation

In order to determine the best level of Aloe gel to be incorporated into the stirred yoghurt, 5%, 7.5%, and 10% were selected as suitable levels compared to the higher levels 15%, 20%, and 25%. This was determined by the trial and error method with preliminary studies. Furthermore, it is assumed that a high amount of Aloe gel may cause uncontrollable thickening which is suitable only for set yoghurt. Then, to select the most suitable and best Aloe gel percentage, a sensory evaluation was done to check the preference for color, aroma, appearance, consistency, taste, texture and overall acceptability of the samples. Panelists were able to distinguish a significant difference ($P<0.05$) in all attributes among the three stirred yoghurt samples.

Table 2: Median rank values of sensory evaluation

Treatment (Aloe gel% w/v)	Color	Aroma	Consistency	Texture	Taste	Overall Acceptability
5 (T1)	7.97±0.79	7.43±0.50	7.14±1.12	7.03±0.71	7.80±0.80	6.97±0.71
7.5 (T2)	7.29±0.46	5.23±0.73	5.11±0.96	4.66±0.94	5.46±0.66	4.37±0.73
10 (T3)	5.37±1.14	3.57±1.20	3.63±1.19	2.80±0.96	4.54±0.51	2.63±0.81

Data are presented as mean ± SD of triplicates

Table 2 and figure 1 show that the overall acceptability is ranged from 2.63 to 6.97 among the three yoghurt samples. Moreover, results showed that the overall acceptability of Aloe yoghurt was significantly different ($P<0.05$). All in all higher average values for color (279), aroma (260), consistency (250) taste (273), texture (246) and overall acceptability (244) were observed in stirred yoghurt developed by incorporating 5% OD Aloe gel. Therefore, it is apparent that the level of incorporation of Aloe Vera gel had a significant impact on sensory properties of developed stirred yoghurt samples. Stirred yoghurt developed from adding 5% Aloe Vera gel showed the best sensory qualities with a significant difference ($P<0.05$) compared to 7.5% and 10%.

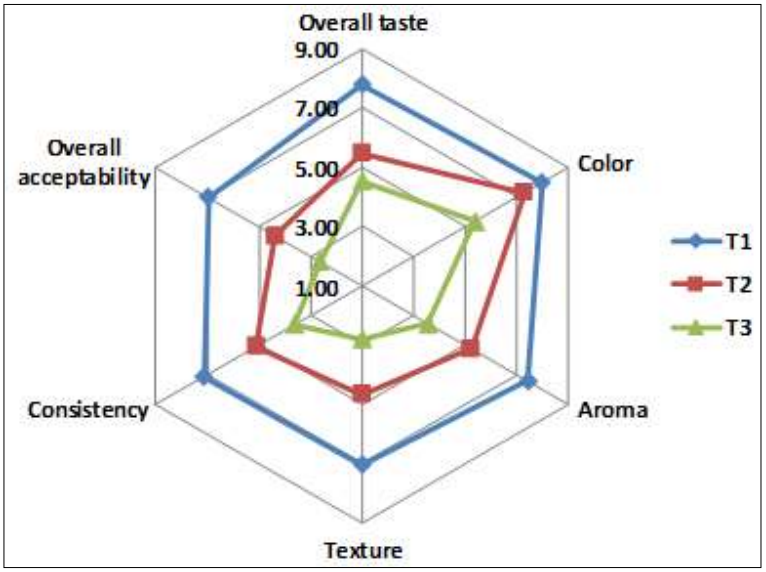

Fig 1: Ranking of sensory properties of yoghurt with different Aloe gel percentage levels

3.3. Curd strength

The textural properties of yoghurt is considered to be crucial with regard to the quality of the products. It is directly related to the sensory perception of a particular product. Therefore, apart from the sensory evaluation, the curd strength was measured for three levels of Aloe vera gel to determine the best Aloe gel percentage. According to the data given in figure 2, the significantly highest ($P<0.05$) curd strength was observed in 10% Aloe gel added yoghurt while the lowest strength was observed in 5% Aloe gel added yoghurt. There was no significant difference observed between the control sample which was a stirred yoghurt without Aloe gel and the 5% Aloe gel added stirred yoghurt. But the 5% Aloe gel added yoghurt had a comparatively higher curd strength than the control one. However, it was assumed as almost similar to the controlled one and the 5% Aloe gel added stirred yoghurt was selected as the best one comparatively to 7.5% and 10% samples for further studies.

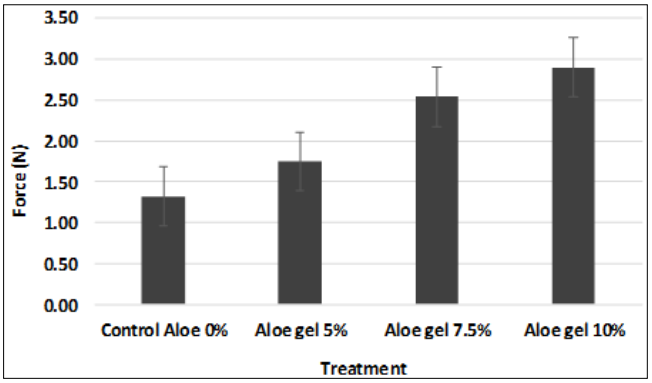

Fig 2: Change in curd strength of OD Aloe gel (5%, 7.5% and 10%) added stirred yoghurt and control stirred yoghurt

According to the sensory evaluation data and curd strength data, 5% OD Aloe gel was selected as the best and most appropriate level of Aloe vera gel. Therefore, to evaluate the physiochemical data, microbial analysis and overall keeping quality data, 5% Aloe vera gel added stirred yoghurt samples were selected.

Physiochemical properties of Aloe vera incorporated stirred yoghurt

3.4. pH

Table 3: Changes in pH of Aloe vera gel 5% incorporated yoghurt during the 5 weeks period under refrigeration

Yoghurt	pH					
	Day 1	Day 7	Day 14	Day 21	Day 28	Day 35
Control	4.68±0.01[a]	4.53±0.00[b]	4.43±0.01[b]	4.26±0.01[b]	4.02±0.01[b]	3.90±0.01[b]
Treated with OD Aloe gel 5%	4.82±0.00[a]	4.81±0.01[a]	4.80±0.01[a]	4.79±0.00[a]	4.78±0.01[a]	4.78±0.01[a]

[a, b] Means in the same column with different superscript are significantly different ($P<0.05$)

Data given in Table 3 and figure 3, clearly indicate that there is a considerable difference in pH between the control and Aloe gel 5% added yoghurt samples throughout the given time period. The pH in control samples was gradually decreased from 4.68 to 3.90 during the storage in refrigerator at 4°C within these 35 days. It is considered to be the reason for this pH reduction is utilization of residual lactose or carbohydrates by viable microorganisms and convert them into lactic acid, CO2 and formic acid. Furthermore, this phenomenon can be described as the post

acidification occurring due to the availability and action of the galactosidase on residual lactose which is still in the active state under very low temperatures (0-5°C) [7, 27]. In contrast, the yoghurt incorporated with Aloe gel shows that it keeps the pH in a certain level within the whole period with small fluctuations. Although the acid producing culture bacteria proportional to the concentration of available protein in the medium, pH was significantly higher (P<0.05) than the control one from beginning to the end even it contains high amount of protein compared to the control one [28]. It can be assumed that the presence of growth inhibitors and antimicrobial properties presence in Alo vera may restrict the activity of acid producing bacteria to some extent during the period [29]. But still the pH of the Aloe stirred yoghurt is in between 4.82 and 4.78 which proves that there is a controlled microbial action. Therefore, it can be assumed that the osmotically dehydrated Aloe vera gel has a preservative action on the developed stirred yoghurt without addition of any synthetic preservatives.

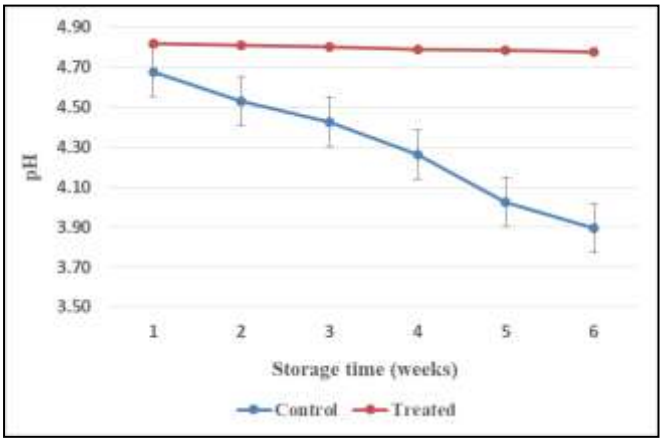

Fig 3: Changes in pH of yoghurt during storage at 4°C for 5 weeks

3.5. Titratable Acidity

Titratable acidity and pH are considered to be two interrelated concepts in food quality evaluation that deal with acidity. But the titratable acidity is a better predictor than pH of the way in which organic acids in a food impact flavor [30]. In the quality evaluation of yoghurt, the titratable acidity is expressed as the percentage of lactic acid in yoghurt. This is a very important factor to determine the shelf life and the acceptability of fermented dairy products [31]. The lactic acid production under pre-determined conditions during fermentation is essential for the formation of yoghurt gel network. On the other hand, continuous acid production after the fermentation is not desired as it leads to wheying-off, textural defects, and excess sourness, which masks the aroma compounds from the consumer [32].

According to the data given in table 4 and figure 4, except on the 35th day, titratable acidity of the stirred aloe yoghurt and the control were not significantly different (P>0.05) throughout the Storage. It clearly shows that the titratable acidity in Aloe treated yoghurt is somewhat lower than the control one though it is not significant. The main reason for this phenomenon also might be the controllable restriction of fermentative bacteria by the Aloe vera gel as same as in the incident of pH. According to the Sri Lankan Standard Specifications, titratable acidity of yoghurt should be in the range of 0.8 to 1.25% lactic acid (SLS: part 2: 1989). Therefore, the Aloe treated stirred yoghurt can be considered to be acceptable in terms of titratable acidity even after 5 weeks of preparation as still, it is under the recommended value.

Table 4: Change in Titratable acidity of the developed stirred yoghurt during the storage at 4°C for 5 weeks period

Yoghurt	Titratable Acidity					
	Day 1	Day 7	Day 14	Day 21	Day 28	Day 35
Control	0.67±0.01^{a}	0.68±0.01^{a}	0.71±0.02^{a}	0.75±0.02^{a}	0.76±0.01^{a}	0.88±0.01^{b}
Treated with OD Aloe gel 5%	0.67±0.02^{a}	0.70±0.01^{a}	0.71±0.00^{a}	0.70±0.01^{a}	0.73±0.00^{a}	0.73±0.01^{a}

$^{a, b}$ Means in the same column with different superscript are significantly different (P<0.05)

3.6. Viscosity

Viscosity is considered to be the resistance that a liquid offers to an applied shearing force [33]. According to the data (Table 5), and the graph (Figure 5), they clearly present that the viscosity of both the control and Aloe gel treated sample have been gradually increased under the storage conditions (4°C) from the first week to 5th week. The viscosity of the Aloe 5% added stirred yoghurt was significantly higher (P>0.05) than the controlled one throughout the period from the very beginning. The reason for this phenomenon might be the addition of skim milk to increase the yoghurt amount in the final product (75%) following the SLS standards.

Table 5: Change in viscosity of 5% Aloe stirred yoghurt during the storage at 4°C for 5 weeks period

Yoghurt	**Viscosity**					
	Day 1	**Day 7**	**Day 14**	**Day 21**	**Day 28**	**Day 35**
Control	31.29±0.07^{a}	34.32±0.03^{a}	44.12±0.03^{a}	50.14±0.04^{a}	61.51±0.03^{a}	67.28±0.02^{a}
Treated with OD Aloe gel 5%	70.34±0.02^{b}	72.43±0.03^{b}	79.01±0.03^{b}	82.02±0.06^{b}	83.11±0.02^{b}	83.34±0.05^{b}

$^{a, b}$ Means in the same column with different superscript are significantly different (P<0.05)

Furthermore, agar-agar has been added as a hydrocolloid to develop the textural properties of the product where bind water reacting with milk proteins to stabilize the protein network preventing free movement of water [34]. Although stirred yoghurt has a poorly stabilized nature, still it is important to prevent being a liquid product. Therefore, the addition of such hydrocolloids in a lower amount can prevent this and it was able to replace gelatin which has an animal origin as this product was a vegetarian product. Moreover, after the stirring process, the Aloe gel pectin associated with the milk calcium may start the re-formation of the gel structure to some extent as there is no further physical force while increasing the viscosity. However, the viscosity of the final product did not have any negative impact on the sensory properties and the increasing viscosity also may take responsibility for remaining constant pH while limiting the action of acid-producing culture bacteria.

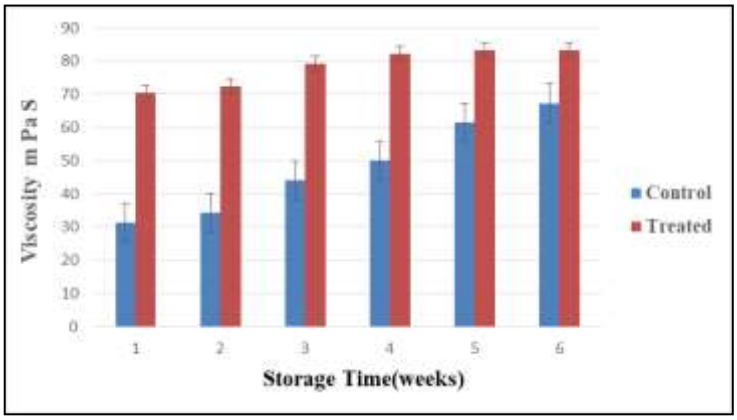

Fig 5: Change in viscosity of 5% Aloe stirred yoghurt during the storage

3.7. Syneresis

Syneresis or the whey separation is a process of expulsion of whey from a performed gel which will be eventually visible as surface whey. This phenomenon is considered to be a serious defect in the yoghurt industry as it affects consumer perception of yogurt. Because they may mistakenly understand that there is microbiological contamination of the product or spoiled.

Table 6: Change in syneresis of 5% Aloe stirred yoghurt during the storage at 4°C for 5 weeks period

Yoghurt	Syneresis					
	Day 1	Day 7	Day 14	Day 21	Day 28	Day 35
Control	12.81±0.07[a]	13.44±0.02[a]	13.80±0.01[b]	15.13±0.05[b]	15.05±0.04[a]	16.25±0.04[b]
Treated with OD Aloe gel 5%	13.23±0.05[a]	14.92±0.02[a]	16.56±0.05[a]	17.12±0.01[a]	17.65±0.06[a]	18.18±0.03[a]

[a, b] Means in the same column with different superscript are significantly different (P<0.05)

Table 6 and figure 6 show the Aloe gel added stirred yoghurt has significantly higher (P>0.05) syneresis than the controlled one under storage conditions after 2nd week. This might be due to four main reasons, first, there should be a direct relationship between acidity and syneresis in yoghurt as total titratable acidity produced by the starter culture and the presence of OD *Aloe vera* gel decrease colloidal stability of casein micelles while increasing the syneresis [35, 36]. Second, the longer storage time could lead to greater proteolysis following continuous fermentation, increasing the whey separation of the final product [36]. Third, the high inherent moisture content and poor water binding capacity of the *Aloe vera* gel and the fourth reason is the destruction of the gel structure during the stirring process. However, this is common in low-fat yoghurt as it has a low solid content. Although osmotically dehydrated Aloe gel has higher protein content and total solid content, it shows comparatively higher syneresis than the control one.

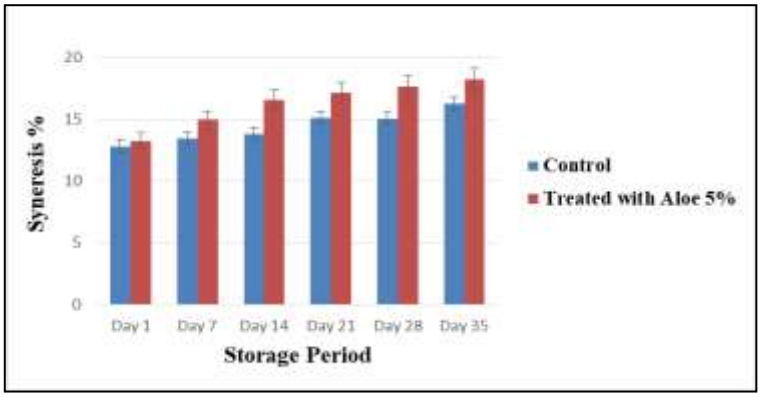

Fig 6: Change in syneresis of 5% Aloe stirred yoghurt during the storage at 4°C for 5 weeks period

3.8. Proximate composition of *Aloe vera* incorporated stirred yoghurt

Table 7: Proximate composition of Aloe yoghurt

Component	Value ± SD
Dry matter%	93.32±0.01
Ash%	5.71±0.02
Crude Fat%	3.43±0.03
Crude Fiber%	0.02±0.00
Crude Protein%	5.23±0.01

The results of the proximate composition analysis of the selected sample are shown in Table 7. According to the SLS standards (1989) for yoghurt, minimum milk fat percentage by mass is 3.0, when determined by Gerber method. The product was in acceptable level in compliance with SLS standards. The major fat source for the product was cow's milk since "Aloe gel" is a poor source of fat. Aloe yoghurt is low in crude fiber since aloe gel is a very poor source of fiber. The average protein content of commercially available yoghurt is 3.501%. The addition of skim milk powder and *Aloe vera* gel causes to increasing the protein level in yoghurt since both acts as a good source of protein [37]. Therefore, the final product which was developed with 5% OD Aloe gel can be considered as a nutritionally enriched product comparatively to commercially available normal stirred yoghurt.

3.9. Microbial Analysis

Table 8: Change in the total viable microbial count during the storage at 4°C for 5 weeks

Yoghurt	Total viable count					
	Day 1	Day 7	Day 14	Day 21	Day 28	Day 35
Control	$1.98\times10^{6\,b}$	$1.88\times10^{6\,b}$	$1.80\times10^{6\,b}$	$1.72\times10^{6\,b}$	$1.64\times10^{6\,b}$	$1.34\times10^{6\,b}$
Treated with 5% of Aloe vera	$1.82\times10^{6\,a}$	$1.74\times10^{6\,a}$	$1.66\times10^{6\,a}$	$1.54\times10^{6\,a}$	$1.24\times10^{6\,a}$	$0.94\times10^{6\,a}$

[a, b] Means in the same column with different superscript are significantly different (P<0.05)

It is evident from results (shown in Table. 8) that there was a significant (p<0.05) decrease in the total viable count (CFU/mg) in both control and Aloe treated stirred yoghurt samples during the storage at 4°C for 35 days. Furthermore, there is a significant (p<0.05) difference between the control sample and the treated sample. The increasing acidity during the storage is considered to be the main reason for this reduction of total viable count during the given period. These findings were consistent with some previous research findings that clearly state the decreasing of viable colony counts during the storage [38].

The reason for comparatively lower bacteria viable count in OD Aloe treated stirred yoghurt is the availability of antimicrobial agents that may inhibit the growth of probiotics strain during the storage period [39]. However, the data (Table 8) clearly show that there are antibacterial activities of OD *Aloe vera* foliar gel in yoghurt during the storage period. There are some research findings associated with the viable count of two main culture bacteria, *Lactobacillus acidophilus*, and *Bifidobacterium lactis.* Panesar and Shinde (2011) show the effect of Aloe vera gel on survival of the L. *acidophilus* in *Aloe vera* fortified probiotic yoghurt. It is assumed that Bifidobacteria strains are more susceptible to loss of cell count due to lower acidic pH and oxygen tolerance [40, 41]. Moreover, it is considered that acid sensitivity is occurred due to the influence of H+ - ATPase as well as occurring of toxic oxygen metabolites o^{-}_{2}, OH^{-} and H_2O_2 due to the loss of oxygen-scavenging system lead to cell death [42].

Table 9: Change in yeast and mold count during the storage time

Yoghurt	Yeast and mold count (CFU/g)					
	Day 1	**Day 7**	**Day 14**	**Day 21**	**Day 28**	**Day 35**
Control	Nil	Nil	Nil	0.17×10^{3}	0.86×10^{3}	1.56×10^{3}
Treated with 5% of *Aloe vera*	Nil	Nil	Nil	Nil	Nil	0.08×10^{3}

Table 10: Change in the total coliform count during the storage time

Yoghurt	Total Coliform Count during the storage period (CFU/g)					
	Day 1	**Day 7**	**Day 14**	**Day 21**	**Day 28**	**Day 35**
Control	Nil	Nil	Nil	Nil	Nil	Nil
Treated with 5% of Aloe vera	Nil	Nil	Nil	Nil	Nil	Nil

According to the yeast and mold count (Table 9), in the control sample, it has been started to increase from 21st day and exceeded the standard limit 10 CFU/g (SLS 824: Part 2:1989) by 35th day. In contrast, the OD Ale gel treated stirred yoghurt sample shows that there is no any yeast and mold growth till 35th day where still it is acceptable to consume since the yeast and mold growth is under the recommended value. Table 10 clearly present that there is no coliform count in both controlled and treated samples throughout the storage at 4°C. The main reason for this observation is the antimicrobial properties of different metabolic products including organic acids, fatty acids, hydrogen peroxide, and relatively small diacetyl, produced by lactic acid-producing bacteria. However, the reason for yeast and mold growth in later stages is the reduction of oxygen and increasing acidity while decreasing the viable culture bacterial cell count. Because it makes a favorable condition for growing yeast and mold. But in OD Aloe gel added stirred yoghurt had an extended shelf life as both the cultural bacteria and Aloe gel working as biological and natural preservatives. Total coliform counts were zero in both the treated and controlled samples probably due to

good hygienic practices adapted during the manufacturing process. All in all, microbial analysis proves that OD Aloe gel 5% added stirred yoghurt shows high resistance for microbial growth while keeping the growth of beneficial bacteria in a controlled level

4. Conclusion

In this study, we developed an osmotically dehydrated *Aloe vera* gel incorporated stirred yoghurt. We evaluated different features such as the best percentage of gel, pH, titratable acidity, viscosity, syneresis, proximate composition, and the microbial analysis for 35 days of storage period. Finally, 5% OD Aloe gel added stirred yoghurt was selected as the most preferred yoghurt and according to the observed microbial and physiochemical quality changes during the given period, it can be concluded that this product has a prolonged shelf life and it can be introduced into the commercial market as a nutritionally enriched profitable fermented dairy product. It is possible to reduce the wastage of surplus seasonal supply of products to the market due to its prolonged keeping quality.

5. Acknowledgement

We acknowledge Dr. W.M.H.K Wijenayake, Senior lecturer, Department of Aquaculture and Fisheries, Faculty of Livestock Fisheries and Nutrition, Wayamba University of Sri Lanka for providing required resources to conduct the research.

6. References

1. Kumar S, Kumar K, Suman S, Kumar P. Cow Milk and Human Health-A Review. Res Rev J Dairy Sci Technol, 2014.
2. Farnworth ER, Mainville I, Desjardins MP, Gardner N, Fliss I, Champagne C, *et al*. Growth of probiotic bacteria and bifidobacteria in a soy yogurt formulation. Int J Food Microbiol, 2007.
3. Haenlein GFW. Goat milk in human nutrition. In: Small Ruminant Research, 2004.
4. McKinley MC. The nutrition and health benefits of yoghurt. Int. J Dairy Technol, 2005;
5. Rasic JJK. Yoghurt: scientific grounds, technology, manufacture and preparations. Staempfli +cie ag berne, 1978.
6. Lourens-Hattingh A, Viljoen BC. Yogurt as probiotic carrier food. Int. Dairy J, 2001.
7. Panesar PS, Shinde C. Effect of Storage on Syneresis, pH, Lactobacillus acidophilus Count, Bifidobacterium bifidum Count of Aloe vera Fortified Probiotic Yoghurt. Curr Res Dairy Sci, 2012.
8. Saad N, Delattre C, Urdaci M, Schmitter JM, Bressollier P. An overview of the last advances in probiotic and prebiotic field. LWT - Food Sci. Technol, 2013.
9. Salwa AA, Galal EA, Neimat AE. Carrot yoghurt: sensory, chemical, microbiological properties and consumer acceptance. Pakistan J Nutr, 2004.
10. Iwalokun BA, Shittu MO. Effect of Hibiscus sabdariffa (calyce) extract on biochemical and organoleptic properties of yogurt. Pakistan J Nutr, 2007.
11. Vahedi N, Tehrani MM, Shahidi F. Optimizing of Fruit Yoghurt Formulation and Evaluating Its Quality During Storage. J Agric Environ Sci, 2008.
12. Küçüköner E, Tarakçi Z. Influence of different fruit additives on some properties of stirred yoghurt during storage. Milchwissenschaft, 2004.
13. Yildiz F. Development and manufacture of yogurt and other functional dairy products, 2016.
14. Pisalkar PS, Jain NK, Jain SK. Osmo-air drying of aloe vera gel cubes. J Food Sci Technol, 2011.
15. Rajeswari RM, Umadevi CSR, Selvavenkadesh S, Sampath KP, Kumar DB. Aloe vera: The Miracle Plant Its Medicinal and Traditional Uses in India. J Pharmacogn Phytochem, 2012.
16. Hu C, Huang J, Fang N, Xie S, Henderson GM, Cai Y, *et al*. Adsorbed silica in stalagmite carbonate and its relationship to past rainfall. Geochim Cosmochim Acta, 2005.
17. Reynolds T, Dweck AC. Aloe vera leaf gel: A review update. J Ethnopharmacol, 1999.
18. Chithra P, Sajithlal GB, Chandrakasan G. Influence of aloe vera on the healing of dermal wounds in diabetic rats. J Ethnopharmacol, 1998.
19. Gomez-Guillen MC, Gimenez B, Lopez-Caballero ME, Montero MP. Functional and bioactive properties of collagen and gelatin from alternative sources: A review. Food Hydrocoll, 2011.
20. Salvador A, Fiszman SM. Textural Characteristics and Dynamic Oscillatory Rheology of Maturation of Milk Gelatin Gels with Low Acidity. J Dairy Sci, 1998.
21. Morrison NA, Sworn G, Clark RC, Chen YL, Talashek T. Gelatin alternatives for the food industry. Prog Colloid Polym Sci, 1999.
22. Rasmussen RS, Morrissey MT. Marine Biotechnology for Production of Food Ingredients. Adv. Food Nutr. Res, 2007.
23. Pisalkar PS, Jain NK, Pathare PB, Murumkar RP, Revaskar VA. Osmotic dehydration of aloe vera cubes and selection of suitable drying model. Int Food Res J, 2014.
24. Helrick K. Official methods of analysis (AOAC), 1990.
25. Kleyn DH, Lynch JM, Barbano DM, Bloom MJ, Mitchell MW. Determination of fat in raw and processed milks by the Gerber method: Collaborative study. J AOAC Int, 2001.
26. AOAC. Official Methods of Analysis of AOAC International, 2005.
27. Kailasapathy K. Survival of free and encapsulated probiotic bacteria and their effect on the sensory properties of yoghurt. LWT - Food Sci Technol, 2006.
28. Rawson HL, Marshall VM. Effect of “ropy” strains of Lactobacillus delbrueckii ssp. bulgaricus and Streptococcus thermophilus on rheology of stirred yogurt. Int J Food Sci Technol, 1997.
29. Jiang M, Deng K, Jiang C, Fu M, Guo C, Wang X, *et al*. Evaluation of the Antioxidative, Antibacterial, and Anti-Inflammatory Effects of the Aloe Fermentation Supernatant Containing Lactobacillus plantarum HM218749.1. Mediators Inflamm, 2016.
30. Tyl C, Sadler GD. pH and Titratable Acidity. 2017.
31. Mahmoudi R, Zare P, Hassanzadeh P, Nosratpour S. Effect of Teucrium polium essential oil on the physicochemical and sensory properties of probiotic yoghurt. J Food Process Preserv, 2014.
32. McFeeters RF. Fermentation Microorganisms and Flavor Changes in Fermented Foods. In: Journal of Food Science, 2004.
33. Bergthaller W. Kramer, A., und B. A. Twigg: Quality

Control for the Food Industry, Vol. 1. Fundamentals, 3rd Ed., The AVI Publishing Company, Inc., Westport, Connecticut, 1970. 19 Kapitel mit 556 Seiten, Preis in USA $ 16,50, im Ausland $ 17,50. Starch – Stärke, 1971.
34. Tamime AY, Robinson RK. Tamime and Robinson's Yoghurt: Science and Technology: Third Edition, 2007.
35. Fox PF, Guinee TP, Cogan TM, McSweeney PLH. Fundamentals of cheese science, second edition, 2016.
36. Michael M, Phebus RK, Schmidt KA. Impact of a plant extract on the viability of Lactobacillus delbrueckii ssp. bulgaricus and Streptococcus thermophilus in nonfat yogurt. Int Dairy J, 2010.
37. Damin MR, Alcântara MR, Nunes AP, Oliveira MN. Effects of milk supplementation with skim milk powder, whey protein concentrate and sodium caseinate on acidification kinetics, rheological properties and structure of nonfat stirred yogurt. LWT - Food Sci Technol, 2009.
38. Kailasapathy K, Harmstorf I, Phillips M. Survival of Lactobacillus acidophilus and Bifidobacterium animalis ssp. lactis in stirred fruit yogurts. LWT - Food Sci Technol, 2008.
39. Nejatzadeh-Barandozi F. Antibacterial activities and antioxidant capacity of Aloe vera. Org Med Chem Lett, 2013.
40. Godward G, Sultana K, Kailasapathy K, Peiris P, Arumugaswamy R, Reynolds N, *et al*. The importance of strain selection on the viability and survival of probiotic bacteria in dairy foods. Milchwissenschaft, 2000.
41. Vasiljevic T, Shah NP. Probiotics-From Metchnikoff to bioactives. Int. Dairy J, 2008.
42. Cruz AG, Antunes AEC, Sousa ALOP, Faria JAF, Saad SMI. Ice-cream as a probiotic food carrier. Food Res. Int, 2009.