

EVALUATION OF HAEMATO BIOCHEMICAL CHANGES IN CONTINUOUS RATE INFUSION OF 2% LIGNOCAINE DURING KETOFOL ANAESTHESIA IN DOGS

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ABSTRACT

The effect of 2% lignocaine CRI during ketofol anaesthesia was studied in twelve adults dogs divided into two groups consisting of 6 animals each viz. Group-I (ketofol), Group-II (ketofol and 2% Lignocaine CRI) presented for hysterectomy procedure. Haematological and biochemical parameters were found to be within the physiological range. Ketofol along with 2% lignocaine as CRI showed no major advantage over ketofol alone.

Keywords: Atropine sulphate, Continuous rate infusion (CRI), Dog, Ketofol, Lignocaine, Xylazine HCl

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INTRODUCTION

An ideal general anaesthetic should bear property of quick and smooth induction, muscle relaxation, sedation, analgesia minimal adverse effects, and rapid and smooth recovery of protective reflexes. Since these characteristics cannot be provided by a single anaesthetic agent. Therefore the combination of anaesthetic agents is used to

achieve balanced anaesthesia (Thurmon and Short, 2007). Parenteral balanced anaesthesia can be achieved by pre-medication with sedative and analgesic agents and continuous rate infusion during the maintenance of anaesthesia by an anaesthetic agent.

Total intravenous anaesthesia is a good alternative to inhalation anaesthesia to carry out even major long-duration surgical procedures and selection of anaesthetic agent significantly improves intraoperative cardiovascular stability, perioperative analgesia, and the quality of recovery. An analgesic agent is always selected for peri- and postoperative analgesia (Bednarski, 2007).

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MATERIALS AND METHODS

The study was carried out on twelve adult dogs brought to the Teaching Veterinary Clinical Complex and Department of Veterinary Surgery and Radiology, Bihar Veterinary College, Patna for removal of infected uterus. Dogs were kept off feed for 12 hrs before surgery.

Blood samples were taken at 0 (base value), 15, 30, 60 min, and 120 min after administration of drugs. Biochemical and haematological parameters were estimated by Vet Chemistry analyzer SS-168 Vet using the commercial standard kit of Coral clinical Systems.

RESULTS AND DISCUSSION

Haematological observations

In group-I, haemoglobin level showed a significantly ($P<0.05$) decreasing trend till 60 min and then increased (Table I). Similar findings were also reported by Kumar *et al.* (2014) during continuous infusion of ketofol in dogs. In group-II, haemoglobin level showed insignificantly ($P>0.05$) decreasing trend till 30 min and then increased at 60 min and then decrease at the end. The decrease in haemoglobin level might be due to the splenic pooling of erythrocytes that occur with most of the anaesthetics. There was no significant difference in hemoglobin values among the groups at any time interval till the end of the anaesthetic sessions except at 60 min.

In group-I a significant decrease in PCV was observed after the administration

of ketofol in dogs. The present findings in group-II animals showed a significant ($P<0.05$) decrease up to 30 min and then increase at 60 min. Thereafter, it resumed gradually to the normal value. The decrease in PCV in groups might be due to pooling of circulating erythrocytes in the spleen of other reservoirs secondary to decreased sympathetic stimulation (Amarpal *et al.*, 1998) or due to inter-compartmental fluid shift to maintain normal cardiac output (Das, 2013). There was no significant difference in PCV values among groups.

In both group-I and group-II, total erythrocyte count showed an insignificant decreasing trend up to the 30th minute and a significant decrease at the 60th minute. In both groups it returned gradually to the normal value. Kumar *et al.* (2014) reported an insignificant decreasing trend of TEC remained within normal physiological limits indicating the safe and efficient role of ketofol in general anaesthesia in dogs. The decrease in total erythrocyte count in both groups might be due to pooling of circulating erythrocytes in the spleen or other reservoirs secondary to decreased sympathetic stimulation. The values of TLC did not register any significant change at the different time intervals of TIVA in groups.

Biochemical observation

In group-I and II, a significant increase in blood glucose level was observed during the period (Table II). In group-II, a significant increase in blood glucose level was observed at 15 min and thereafter insignificant increase

Table I. Haematological observations

Haemoglobin(gm/dl)					
Group	0 min	15 min	30 min	60 min	120 min
I	13.30±0.58	13.03±0.57	12.70±0.56	12.07±0.53 ^{bc}	12.78±0.56
II	14.30±0.54	13.95±0.53	13.85±0.52	14.13±0.53 ^a	13.39±0.51
PCV (%)					
I	35.51±1.56	34.78±1.53	33.92±1.49	32.23±1.42 ^{bc}	34.13±1.50
II	38.18±1.44	37.25±1.41	36.99±1.40	37.72±1.42 ^a	35.76±1.35
TEC (10⁶/cu. mm)					
I	6.20±0.27	6.07±0.27	5.92±0.26	5.62±0.25 ^{bc}	5.96±0.26
II	6.66±0.25	6.50±0.25	6.45±0.24	6.58±0.25 ^a	6.24±0.24
TLC (10³/cu.mm)					
I	9.14±0.64 ^b	8.39±0.58 ^b	8.01±0.56 ^b	8.27±0.57 ^b	8.62±0.60 ^b
II	12.76±0.63 ^a	11.75±0.58 ^a	11.26±0.55 ^a	11.61±0.57 ^a	12.08±0.60 ^a

The means with a different upper case superscript in a row differ significantly and the means with a different lower case superscript in a column differ significantly at (P < 0.05%)

Table II. Biochemical observations

Blood glucose (mg/dl)					
Group	0 min	15 min	30 min	60 min	120 min
I	71.63±0.44 ^{dE}	82.25±0.51 ^{bD}	85.20±0.52 ^{cC}	91.95±0.57 ^{cB}	94.88±0.58 ^{cA}
II	83.56±0.80 ^{bD}	96.10±0.92 ^{aC}	99.86±0.95 ^{aC}	107.92±1.03 ^{aB}	112.00±1.07 ^{aA}
BUN (mg/dl)					
I	12.27±0.43 ^{abB}	14.99±0.53 ^{abA}	13.98±0.49 ^{bcAB}	14.10±0.49 ^{bAB}	14.14±0.50 ^{abAB}
II	10.47±0.67 ^b	12.78±0.82 ^b	11.99±0.77 ^c	12.09±0.78 ^b	12.11±0.78 ^b
Creatinine (mg/dl)					
I	0.81±0.05 ^c	0.83±0.05 ^b	0.82±0.05 ^b	0.92±0.05 ^c	0.87±0.05 ^c
II	1.08±0.05 ^b	1.10±0.06 ^b	1.08±0.05 ^b	1.22±0.06 ^{bc}	1.15±0.06 ^{bc}
SGPT (ALT) (IU/L)					
I	40.59±0.21 ^{aC}	42.51±0.22 ^{abB}	45.65±0.24 ^{aA}	45.70±0.24 ^{aA}	42.31±0.22 ^{abB}
II	38.65±0.61 ^{abB}	40.51±0.64 ^{abB}	43.54±0.69 ^{abA}	43.71±0.69 ^{abA}	40.44±0.64 ^{abB}

The means with a different upper case superscript in a row differ significantly and the means with a different lower case superscript in a column differ significantly at (P < 0.05%).

at 30 min and again a significant increase was observed. A similar observation was also reported by Costa *et al.* (2013) and Ratnesh *et al.* (2014) during propofol anaesthesia, and Ayadi *et al.* (2008) during propofol or ketamine anaesthesia in xylazine premedicated dogs. An increase in serum glucose level might be due to decreased membrane transport of glucose, decreased glucose utilization; inhibition of insulin release mediated by alpha-2 receptors in pancreatic beta cells, and increased blood concentration of adrenocortical hormone (Das, 2013). Jena *et al.* (2014) observed that practically every anaesthetic stimulates the secretion from the adrenal cortex, responsible for gluconeogenesis during anaesthesia. The rise in glucose level might be due to an increase in circulatory catecholamines after premedication (Hall *et al.*, 1994) and also the effect of the anaesthetic agent on the subcortical pathway, which was responsible for the regulation of adrenocorticotrophic hormone (ACTH) and produce stress like conditions with increased release of glucocorticoids (Jena *et al.*, 2014). In group-I and II no definite trend of BUN value during Total Intravenous Anaesthesia (TIVA) at a different time interval, it fluctuated within normal physiological limits. It might be due to the opposite hemodynamic action of ketamine and propofol.

In group-I and II, creatinine values were recorded a little higher than their base value during TIVA at different time intervals. It fluctuated within physiological limits. It might be due to the opposite hemodynamic action of

ketamine and propofol. Using 2% lignocaine as CRI did not cause any significant change. It might be due to the temporary inhibitory effects of individual anaesthetic drugs on the renal blood flow leading to a decrease in glomerular filtration rate. In group I and II, alanine aminotransferase values significantly increased up to 6 min during TIVA and thereafter gradually decreased towards the normal. An increase of ALT enzyme in the serum might be due to cellular destruction of heart, liver, kidney, pancreas brain tissue, or skeletal muscle.

No significant differences in ALT values within the groups were recorded. The transient variation in ALT values was within physiological limits during the anaesthetic period might be indicative of the non-toxic effect of all the anaesthetic drugs on heart, liver, kidney, pancreas brain tissue, or skeletal muscle.

Conclusions

In group-I and group-II induction with ketofol was rapid and smooth, no apnoea was observed, recovery was very smooth. Based on the biochemical and haematological parameters, the ketofol and the ketofol-lignocaine 2% was more suitable. Analgesia and muscle relaxation were excellent in ketofol and ketofol-lignocaine 2%. The duration of surgical anaesthesia of ketofol-lignocaine 2% was longer as compared to Ketofol alone. Maintenance of anaesthesia was smooth and stable with good muscle relaxation and better analgesia than group I. Initial significant decline was observed in Hb, PCV, and TEC which steadily returned to the pre-anaesthetic

level at end of observation. Significant increase in glucose, BUN, creatinine, ALT values were recorded in these groups in early phase then shows declining trend towards the pre-anaesthetic level.

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DEVELOPMENT OF FUNCTIONAL MILK LOLLIES UTILIZING LYOPHILIZED TAMARIND SEED KERNEL POWDER

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ABSTRACT

A research was undertaken to enrich the fibre content in milk lollies by incorporating defatted tamarind seed kernel powder (TSKP) at 0.25, 0.5 and 0.75 per cent levels in suitable form. The developed products were subjected to sensory, physicochemical, proximate, microbiological and cost analysis. Sensory analysis revealed good overall acceptability. The pH values increased in milk lollies with consequent decrease in titratable acidity. As the inclusion level of TSKP increased both the specific gravity and viscosity increased from 1.10±0.00 to 1.13±0.00 and 48.35±1.63 to 57.52±1.38 respectively with a highly significant difference ($p \leq 0.01$). The melting resistance showed a marked improvement with inclusion of TSKP and the time of first drip was the lowest (2 minutes) for control and the highest (5.5 minutes) for milk lollies with 0.75 per cent TSKP. The proximate analysis of milk lollies showed no significant difference among control and treatment milk lollies. Coliforms were absent in all the samples and total viable count for milk lollies were within the acceptable level (5.10 to 5.20 log cfu/ml). Though the cost analysis of milk lollies revealed a slight increase in the rates of treatment groups compared to that of control, the rates were found to be acceptable with added health benefits.

Key Words: Milk lollies, Fibre enrichment, Tamarind Seed Kernel Powder (TSKP), Analysis

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INTRODUCTION

Fortified functional foods are those food products which exhibit added health benefits on addition of one or more functional ingredients. The Institute of Medicine's

Food and Nutrition Board (IOM/FNB, 1994) defined functional foods as “any food or food ingredient that may provide a health benefit beyond the traditional nutrients”. The food markets are in the verge of production of functional foods to satisfy the consumers with increased demands for convenient, healthy and tasty food products. Development of functional foods without affecting their sensory qualities is the utmost idea to explore the market potential of functional foods.

Dairy products by virtue are natural functional foods but, further incorporation of functional ingredient makes them still a wholesome food. Milk and milk products are the best source of calcium. Dairy products act as an excellent vehicle to carry several of external nutrients, probiotics as well as prebiotics. Milk lollies have already been accepted by the people of all ages. The soluble form of fibre can easily be incorporated into the milk lollies. The rich creamy taste of milk lollies will mask the flavour and taste reflected due to the incorporation of dietary fibre from plant source.

The benefits of dietary fibre include preventing constipation, lowering the risk of coronary heart disease and colon cancer, attenuating blood glucose levels, reducing serum cholesterol levels, providing weight management, and functioning as a prebiotic (Kritchevsky, 1998; Joanne *et al.*, 2003; Ohr, 2004). Dietary fibres can be either soluble or insoluble as per their ability to form dispersion with water or not respectively. Soluble dietary fibres are being used in the preparation of various dairy products especially due to their property to form viscous gels with water. As soluble fiber dissolves forming a gel which

improves digestion, reduces blood cholesterol and sugar. It helps the body to improve the blood glucose control thus reducing the risk of diabetes. The increasing demand for soluble fibre has led to the search of non-conventional sources of soluble fibre thus reducing the production cost of the functional foods.

Among the several sources of soluble fibres, the seeds of *Tamarindus indica L.*, a highly under-utilized by-product from the agricultural industry, which is about 34 per cent of entire fruit, serves as a good source of dietary fibre especially soluble fibre. The specific soluble fibre of tamarind seed kernel powder (TSKP) is the polysaccharide called xyloglucan, which are utilized as an alternative for food grade polysaccharides owing to their higher molecular weight. Tamarind xyloglucan is beneficial to human health as this compound possesses several physiological functions, such as hypoglycemic effects (Sone *et al.*, 1992), antihyperlipidemic effects (Yamatoya *et al.*, 2011), and as a mucosal protector with barrier protective properties (Pique *et al.*, 2018).

The present study was carried out to analyze the sensory, physicochemical, proximate, microbiological attributes and cost of production after inclusion of defatted tamarind seed kernel powder into milk lollies at different levels.

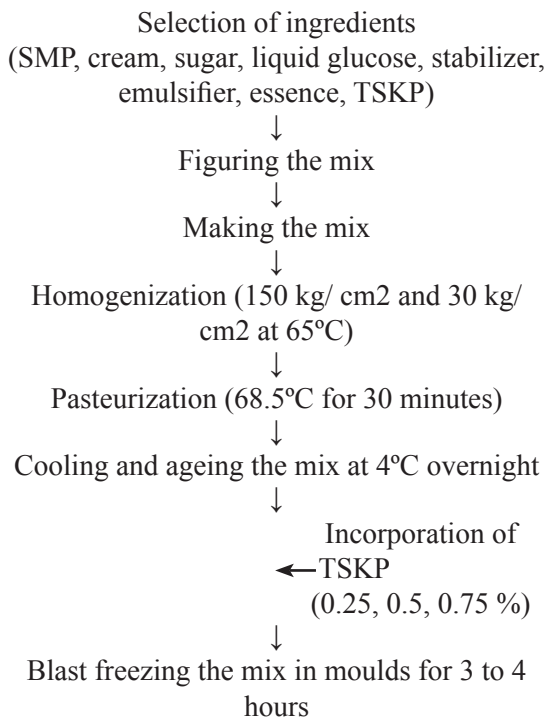
MATERIALS AND METHODS

This study compared the properties of control milk lollies with the milk lollies incorporated with different levels viz. 0.25, 0.5 and 0.75 per cent levels of defatted tamarind seed kernel powder (TSKP). Milk

for preparation of milk lollies was obtained from the Model Dairy Plant, Department of Livestock Products Technology (Dairy Science), Madras Veterinary College, Chennai. Other raw materials were purchased from local markets, Chennai. Tamarind seeds obtained were washed, dried in hot air oven at 60° C for 6 hours, roasted at 150° C for 10 minutes, mechanically crushed and decorticated, then ground to a fine powder and sieved. The TSK powder was subjected to ether extraction to remove the fat. The TSKP was incorporated into the milk lollies prior to blast freezing.

The procedure for the preparation of milk lollies is represented as a flow chart 1.

Flow chart 1: Process for the preparation of milk lollies



Milk lolly

↓
Packaging

↓
Hardening and storage (– 23°C to – 29°C)

Physicochemical analysis

Fat content was estimated by Gerber's method (Sukumar De, 1991). The pH of the mix after ageing was estimated using digital pH meter (Systronics micro pH systems, 361). The titratable acidity of the milk lollies was measured by titration of the prepared mix against 0.1N sodium hydroxide with phenolphthalein indicator as per AOAC, (2000) method. The specific gravity of the milk lolly mix was determined as per the method described by Sommer, (1951). The viscosity was estimated according to the protocol mentioned in BIS procedure IS: 1479 (Part I) 1960. Meltdown was estimated as per the protocols of Khillari *et al.* (2007). Estimation of moisture, crude protein, crude fibre, ether extract ash content and nitrogen free extracts were done as per AOAC, 2000 method. The sensory evaluation was carried out by a panel of judges comprising of 6 members using of 9 – point hedonic scale (Adhikari *et al.*, 2011). Standard plate count and coliform counts were estimated as per IS, SP: 18 (Part XI), (1981). The data obtained were subjected to one way analysis of variance (ANOVA). The test of significance was determined with Duncan's test at $p < 0.05$ probability using the SPSS statistics® 20 software Snedecor and Cochran, 1994.

RESULTS AND DISCUSSION

The sensory analysis scores of the milk lollies were presented in the Figure 1 and

it was evident from the results that there were no significant difference ($p>0.05$) among the sensory attributes viz. appearance, flavour, body, texture and overall acceptability. Similar results were explained by Isik *et al.* (2011), who prepared ice cream with inulin and isomalt, where sensory analysis revealed that ice cream with inulin and isomalt up to 6.5 per cent was similar to the control. Homogenization of milk lolly after addition of lyophilized TSKP and use of minimal levels did not affect the appearance of milk lollies, which was supported by Kurt and Atalar, (2018) in ice cream incorporated with quince seed powder at 0.25, 0.5 and 0.75 per cent levels.

The flavour ranged from 8.67 to 8.83. There was a mild decrease in flavour scores of treatment than that of control milk lollies and was accepted in the process of developing a healthy product. The body and texture scores were the same (8.83 ± 0.17) in both the control and treatment milk lollies. Jellose present in tamarind seed kernel powder is highly preferred in the preparation of frozen desserts like ice cream as described by Vinod, (1997). The jellose supported the matrix of milk lollies thus maintaining the body and texture. The results obtained showed that milk lollies could be produced using functional ingredients without affecting the overall acceptability of the final product, which was in accordance with the observations made by Kurt and Atalar, (2018) in the preparation of ice cream admixed with quince seed powder at 0.25, 0.5 and 0.75 per cent levels.

The physicochemical properties like pH, titratable acidity, specific gravity, viscosity and melt down for the control and treatments showed a highly significant

difference ($p\leq 0.01$) while fat showed no significant difference ($p>0.05$), which was obvious from the Table I. Analysis of fat per cent of the control, MLT_1 , MLT_2 and MLT_3 milk lollies by Gerber's method showed no significant difference (10.00 to 10.17 per cent) between the groups which was due to the use of defatted TSKP.

The pH values of the control as well as treatment (MLT_1 , MLT_2 and MLT_3) milk lollies displayed a highly significant ($p\leq 0.01$) difference. The values were increasing mildly with increase in the level of incorporation of TSKP, which might be due to the influence of pH (5.2 ± 0.01) of TSKP as reported by Oluseyi and Temitayo, (2015). There was a decrease in titratable acidity with increase in the level of incorporation of lyophilized TSKP, which might be due to an increase in pH with increasing concentration of the added purified TSKP. Similar trend was observed by Natukunda *et al.* (2016), in mango juice enriched with tamarind kernel powder.

Specific gravity between the control and treatments (MLT_1 , MLT_2 and MLT_3) milk lollies showed a highly significant ($p\leq 0.01$) difference. The specific gravity increased with the addition of lyophilized TSKP proportionately which might be due to the increase in the total solids content of the milk lollies, where Kebary *et al.* (2020) also observed the same trend in ice milk added with 0.5, 1, 1.5 and 2.0 per cent mango peel powder.

The viscosity values were highly significant ($p\leq 0.01$) between the control and treatment milk lollies. The increase in viscosity could have been due to the

xyloglucan, a polysaccharide present in TSKP which has higher viscosity inherently, which coincided with study of Sultana *et al.* (2020), who observed that the viscosity of the control ice cream (48.35 centipoises) was lesser than that of tamarind kernel powder incorporated ice cream (86.4 centipoises).

From the Figure 2, the meltdown time had highly significant difference ($p \leq 0.01$) between treatments (MLT_1 , MLT_2 and MLT_3) and the control milk lollies, as the treatments took longer time to melt compared to the control. The time of first drip of milk lolly increased with increasing ratio of TSKP which coincided with the study of Kurt and Atalar, (2018), in ice cream with quince seed powder. This increased melting resistance was needed for consumer acceptance in countries with tropical climate. These melt down values indicated that TSKP produced a higher stabilization effect on the milk lollies network. Similar to this study, the addition of high fiber persimmon puree (Karaman *et al.*, 2014) and cape gooseberries (Erkaya *et al.*, 2012) to ice cream resulted in better melting resistance due to the effect of dietary fiber on the water-holding capacity.

The proximate composition of the control milk lollies and those incorporated with lyophilized TSKP showed no significant difference ($p > 0.05$) in the parameters like moisture, crude protein, ether extract, total ash and nitrogen free extract between the control and treatment milk lollies and were presented in Table II.

The results of microbiological analysis of the control and treatment milk lollies revealed that the total viable counts

were within the limits (5.10 to 5.20 log cfu/ml) while the coliform counts were absent which might be due to the hygienic conditions followed at each level of preparation.

The cost analysis of the control and treatment (MLT_1 , MLT_2 and MLT_3) milk lollies presented in Table III revealed that there was a slight increase in the cost of functional milk lollies incorporated with lyophilized TSKP compared to the control which was due to the cost involved in the process of dialyzing and lyophilization of tamarind seed kernel powder. Tamarind seed being an underutilized agro by-product, its availability is more, thus the cost of raw materials reduced. As the consumer perception of health is important and crucial, this increase of cost might be acceptable.

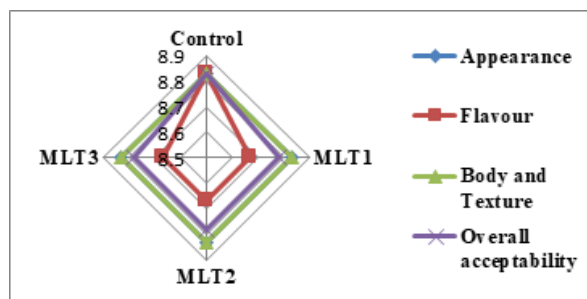


Figure I. Radar chart for the sensory analysis of milk lollies incorporated with lyophilized tamarind seed kernel powder

MLT_1 – Milk lollies with 0.25 per cent tamarind seed kernel powder

MLT_2 – Milk lollies with 0.5 per cent tamarind seed kernel powder

MLT_3 – Milk lollies with 0.75 per cent tamarind seed kernel powder

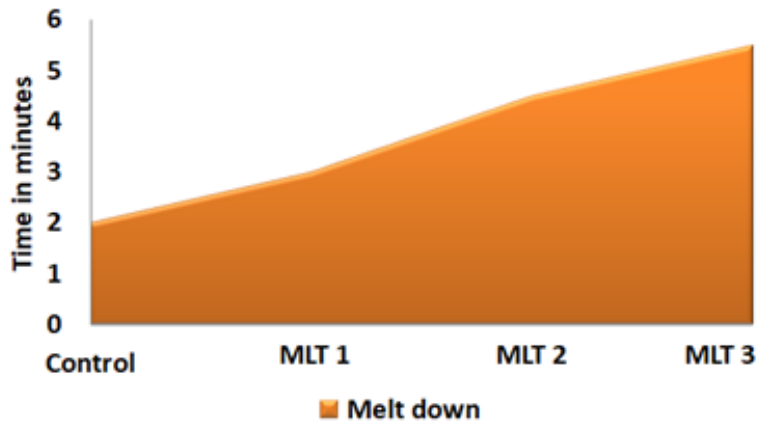


Figure II. Area chart for the melt down of milk lollies incorporated with lyophilized tamarind seed kernel powder

Table 1: Physicochemical properties of milk lollies incorporated with lyophilized tamarind seed kernel powder (mean±SE) #

Physicochemical Attributes	Types of milk lollies				F value
	Control	MLT ₁	MLT ₂	MLT ₃	
Fat (per cent)	10.00±0.11	10.17±0.06	10.00±0.09	10.14±0.12	1.77 ^{NS}
pH	6.38±0.01 ^a	6.40±0.01 ^{ab}	6.42±0.01 ^{bc}	6.44±0.01 ^b	8.48 ^{**}
Titratable acidity (% LA)	0.26±0.00 ^d	0.25±0.00 ^c	0.23±0.00 ^b	0.21±0.00 ^a	96.74 ^{**}
Specific gravity	1.10±0.00 ^a	1.12±0.00 ^b	1.13±0.00 ^c	1.13±0.00 ^d	432.82 ^{**}
Viscosity (centipoises)	48.35±1.63 ^a	51.53±1.05 ^{ab}	55.21±2.27 ^{bc}	57.52±1.38 ^c	6.05 ^{**}
Melt down (minutes)	2.00±0.00 ^a	3.00±0.00 ^a	4.50±0.50 ^b	5.50±0.50 ^b	19.33 ^{**}

- (n=6)

Control: Plain milk lollies

MLT₁, MLT₂ and MLT₃: Milk lollies with 0.25, 0.5% and 0.75% TSKP respectively

Means bearing different superscripts within rows differ significantly

^{NS} –Non significant (p>0.05), ^{**} –Highly significant (p≤0.01)