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DETECTION OF MILK ADULTERANTS AND ASSESSMENT OF KAP IN TIRUPATI URBAN AREA

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ABSTRACT:

Milk is adulterated by different ways like unhygienic milk processing conditions and accidental chemical contaminations. These are mainly categorized into two groups i.e. (a) Edible Adulterants (EA)- water, starch, glucose, sugar, Sodium Chloride (NaCl) and Skimmed Milk Powder (SMP), (b) Chemical and preservative Adulterants (CPA)-formalin, urea, coloring agents, detergents and neutralizers. Present investigation was carried out in Tirupati Urban area, to detect the milk quality and adulteration in the different samples of milk were randomly selected from Local Milk Vendors (LMV) (10 Number), Cooperative Milk Societies (CMS) (8 Number) and Private Milk Companies (PMC) (10 Number). The Physical and chemical parameters of percent water content, pH and Specific Gravity(SG) assessed in all milk samples; high water content, high acidic pH and Low SG was observed in LMS and CMS samples; whereas, all PMC samples in permitted ranges. Edible adulterants mainly detected from the milk samples from LMV and CMS with water, glucose, sugar, NaCl and SMP; starch was identified only in the LMV milk samples. The milk samples from PMC shows negative results in both EA and CPA adulteration tests. Chemical and preservative adulterants such as detergent and neutralizers were detected from both LMV and CMS milk samples; whereas formalin, urea and coloring agents were not found in any milk samples. The KAP (Knowledge, Attitude and Practices) was assessed initially and after the awareness on milk adulteration. The KAP scores were increased after the awareness session than before. The study findings showed that the majority of the milk samples were identified the presence of different adulterants. These milk adulterations may lead to various health impacts on humans.

Key words: Local Milk Vendors, Cooperative Milk Societies, Edible Adulterants, Skimmed Milk Powder and Private Milk Companies.

INTRODUCTION:

Milk is a natural and high source of nutritious food and it is consumed by a majority of people as drink and various dairy products. However, the milk adulteration becomes a great issue among nations in Worldwide. Milk and milk derivative products are important in the diets of humans' and it is a good source of different nutrients, it contains various important nutrients like protein, fat, lactose (milk sugar), vitamins and mineral salts, whey, casein (Arun et al., 2015). Ancient Indians to now a day's milk and milk products preserving for future use and they plays a significant role in the Indian diets, milk is also used as infant food and ready to digest and absorb in the humans' body. Milk is obtained from different milk giving animals such as buffalo, cow, goat and camel.

Milk or dairy milk produces by different animal sources, mainly from cow and buffalos. The chores of milk production, transportation and processing done by middle man; if they abounded to keep maintain appropriate safe and hygienic condition of milk, it may lead to adulterated by harmful substances or bacteria which may reduce the quality of the milk.

The quality of milk is also effected by addition of water, is a very common adultant practice of drinking milk diluted with water or with any other substance is a major socio economic issue in the countries like India and other developing countries, it may increase the quantity but decrease the quality and makes it unfit for human consumption. (JS and DC, 2011). Sometimes milk adulterated with contaminated water, it may cause serious health issues in humans' especially in infants and children (Singuluri, 2014, Azad and Ahmed, 2016).

According to a survey conducted to comprehend the quality of milk by determination of adulteration was conducted by the FSSAI (2018); results indicate that the regular adulterant present in milk was water. Milk adulteration happened in different ways like dilution by water, extraction of milk fat from the whole milk and addition of other cheap solid substances like starch, glucose, sucrose and NaCl to increase the total solid content to meet the consumer acceptance. A report given by the FDA (2009) and FSSAI (2011), the permeable urea limit in milk is 70mg/ 100ml. Urea adulterated by milk in two ways- addition of urea intentionally and incorporation of synthetic milk to natural milk(Sharma et al., 2012).

Adulterated milk consumption has severe impact on human health, sometimes it short term and at times in the long way. The consumption of detergents adulterated in milk able to cause intestinal disorders like inflammation in the intestine and gastritis (Domingo et al., 2014). Urea presented milk can cause kidney disorder, because to excrete excess urea in human body kidneys need to work more (Kandapal et al., 2012). High amounts of starch obstruct digestion and it results diarrhea and excess consumption of starch may increase the glucose levels in the diabetic patients (Singuluri and Sukumaran, 2014).

The milk deliver chains such as milk vendors, milk societies or milk processing companies should maintain on accurate sanitation and good manufacture practices. Studies indicated that the attainment of education on improving knowledge, attitude, and practices of the community in the direction of the safety and quality of milk and other milk products are inadequate in most of the developing countries (Alonso et al., 2018; Lindahl et al., 2018b).

A study conducted by Albert (1995) was described the Knowledge on food safety among urban and rural adults respondents; the results indicated the knowledge scores on food safety was very less among urban respondents when compared with rural respondents (Niharika et al., 2022).

Adulterated milk has susceptible to individual wellbeing and knowledge on improper food handling practices is to consider it an emerging situation both nationally and globally. It is important to take an action on mitigation strategies were needed for proper behaviors on milk handling practices of milk consumers.

The intentions of this study was to detection of various adulterants in different milk samples and assess the pre and post awareness on Knowledge, Attitude, and Practices **(KAP)** of women in Tirupati urban with regard to milk consumption and milk adulterants and evaluate the results.

METHODOLOGY:

The milk samples were collected from the different sources such as Local Milk Vendors (LMV, NO: 10); Cooperative Milk Society (CMS, NO: 8) and Private Milk Companies (PMC, NO: 10) packed milks were obtained by random sampling method in Tirupati urban area, Andhra Pradesh, India. Samples were collected in a sterilized bottles, each sample volume is 100ml; these sampled milk bottles were kept in refrigerator for further laboratory tests to find out the presence of adulterants in the collected milk samples. The sampled milk was analyzed for testing quality of milk and presence of various adulterants. The quality of milk was assessed through percent of water (moisture) content, Specific Gravity (SG) and pH (acidity).

Water: Presence of extraneous water in market milk was determined by the method of Association of Official Analytical Chemists (AOAC, 2000).

pH: It was assessed by using pH meter. The pH ranges indicates the acidity of milk. The standard pH range is 6.6-6.8 is used for raw milk (Gamechu et al., 2015).

Specific Gravity (SG): Normal milk has a density of 1.026 to 1.032 gm/ ml (or 26-32 on the lacto meter reading) (Robinson, 2002).

The adulterants were categorized into 2 groups. (a) Edible adulterants – Starch, Glucose, Sugar, NaCl and SMP. (b) Chemical and Preservative adulterants – Formalin, Urea, Coloring agents, Detergent, Sodium carbonate and Sodium bicarbonate.

a. Edible adulterants:

1. Starch: Take 3ml sample in a test tube. After boiling it thoroughly, keep it to room temperature. Add 1 drop of 1% Iodine solution. Appearance of blue color indicates the presence of starch. (Sharma et al., 2012).
2. Glucose: Take 1ml of milk sample in test tube. Add 1ml of modified barfoeds reagent. Heat the mixture for exact 3min in a boiling water bath. Rapidly cool under tap water. Add 1ml of Phospomolbidic acid reagent to the turbid solution. Immediate appearance of deep blue color indicates the presence of glucose (Sharma et al., 2011).
3. Sugar: Take 5ml milk sample in a test tube. Add 1ml concentrated HCl and 0.1gm Resorcinol solution. Place the test tube in water bath for 5min. Appearance of red colour indicates the presence of added sugar (Sharma et al, 2012).
4. Nacl: Take 5ml of milk sample into a test tube. Add 1ml of 0.1N silver nitrate solution. Mix the contents thoroughly and add 0.5ml of 10% Potassium chromate solution. Appearance of yellow color indicates the presence of added salts, whereas, brick red color indicates the milk free from added salt (Sharma et al., 2012).
5. Skimmed Milk Powder (SMP): Take some milk in test tubes, add few drops of Nitric acid drop wise, if the milk turns yellow color – it indicates the absence of SMP (Sharma et al., 2012).

b. Chemical/ Preservative adulterants

1. Formalin: Take 10ml of milk sample in a test tube. Add 5ml concentrated sulfuric acid with a little amount of ferric chloride without shaking. Appearance of violet or blue color at the junction of 2 liquid layers indicates the presence of formalin. (Sharma et al, 2012)
2. Urea: Take 5ml of milk sample in test tube. Add 5ml P-Dimethyl amino benzaldehyde reagent. Appearance of distinct yellow color indicates presence of added, formation of slight yellow color indicates natural urea in milk. (Sharma et al., 2012).
3. Coloring agent: Add a few drops of HCl to milk sample- appearance of pink color indicates the Azodyes (De Soja et al., 2000)
4. Detergent: Take 5ml of milk sample into a 15ml test tube. Add 1ml of Methylene blue dye solution and 2ml of Chloroform. Vortex the contents for about 15sec and centrifuge at about 1100rpm/ 3min. Relatively, more intense blue color in lower layer indicates presence of detergent in milk. Relatively more intense blue color in upper layer indicates absence of detergent in milk. (Sharma et al., 2011).
5. Neutralizers: Take 5ml milk in a test tube of 20ml capacity. Add 5ml ethanol (95%) and Rosalic acid solution (0.05% in ethanol) 4drops, mix the contents by manual shaking. Formation of pink color indicates presence of neutralizers in milk (Sharma et al., 2011).

Knowledge, Attitude and Practices (KAP) on Milk Adulteration: To collect KAP data the participants, who was responsible for purchase of milk in house hold level were selected 100 members of women employees from the Sri Padmavati Mahila Visvavidyalayam (SPMVV), Tirupati. The selected women respondents KAP assessment was focused on milk consumption behavior, quality of milk, knowledge on milk adulterants and their impact on human health. The questionnaire represents consumer awareness and behavior towards milk adulteration. KAP data collected in two time interlude, that is initially and after giving the awareness session on milk adulteration. The questionnaire contains total 15 questions (Knowledge-5, Attitude -5 and Practice-5), each question scores 2 points. In the KAP awareness session to the respondents, general milk quality tests and explained about importance of safe and hygienic milk consumption, possibilities of milk adulteration through charts and demonstration. After KAP awareness session- all the respondents were assessed with the same questionnaire, which was used initial KAP assessment; to evaluate the variation of knowledge and behavior in two time points. The obtained data was tabulated and analysis was carried out according to the statistical methods

Results and Discussion:

Milk samples were obtained from various sources like Local Milk Vendors (LMV); Cooperative Milk Societies (CMS) and Private Milk Companies(PMC) packed milk in Tirupati Urban area were examined for adulteration with various substances present in the sampled milk.

The Physical and Chemical parameters such as percent of water (moisture) content, pH and Specific Gravity (SG) of different milk samples were assessed and results were tabulated in Table 1.

Table 1: Mean±SD of Physical and Chemical parameters of Different milk samples

Parameters	LMV N=10	CMS N= 8	PMC N= 10
%Water (Moisture) Content Mean±SD	88.30±0.88	88.05±0.53	87.61±0.12
pH Mean±SD	6.41±0.13	6.53±0.09	6.68±0.08
Specific Gravity Mean±SD	1.0208±0.0054	1.0234±0.0042	1.0277±0.0015

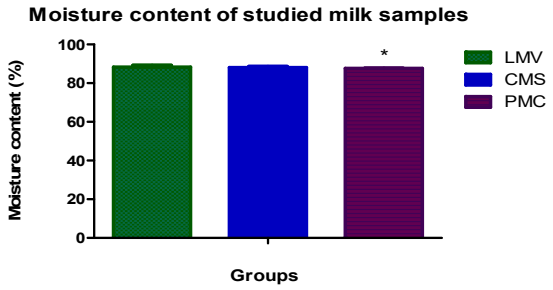

Fig. 1 Percent Moisture (Water) content in Different milk samples

Addition of water: The results of milk adulteration with water was depicted in fig.1; Among all the analyzed milk samples, the LMV samples 80% and 50% of CMS milk samples were found adulterated with water; while PMC milk samples found to be no addition of water. The LMV, CMS and PMC milk samples percent of water ranges 87.60- 90.50, 87.50- 88.95 and 87.40-87.75 respectively. All values were given as mean ± SD of observations. The statistical comparison between groups was done using one-way Anova followed by Tukey multiple comparison test.* = $p< 0.005$ on comparison between LMV Vs PMC. Unpacked LMV samples are highly adulterated with water, when the values compared with PMC samples of standardized milk. LMS and CMS milk samples moisture content was high; whereas the PMC milk samples were generally processed and standard milk and obtained values in acceptable limits. The addition of water with natural raw milk increases the percent of moisture content; in compliance with other studies showed that the raw milk samples (89.5%) contains high moisture compared with processed milk such as Pasteurized milk (88.1%) and Ultra high temperature treated milk (87.7%) (Hossain et al.,(2000) and Mounika and Sireesha (2018).

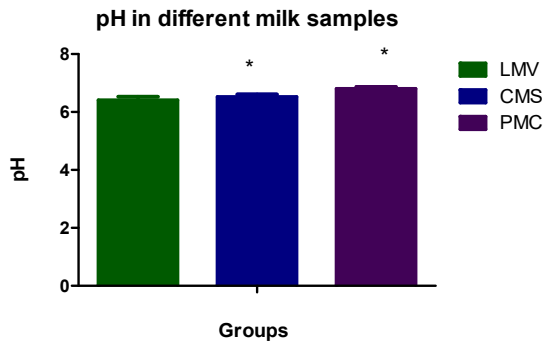

Fig. 2 pH values in Different milk samples

pH: The findings related to pH of milk presented in fig.2. The observations showed that the LMV milk samples showed more Acidic nature (6.3- 6.5) followed by CMS Milk Samples (6.4- 6.6). The PMC milk samples are in an acceptable range of pH (6.6-6.8). All values were given as mean ±SD for different milk samples. The statistical comparison between groups was done using one-way Anova followed by Tukey multiple comparison test. * = p< 0.005 on comparison between LMV Vs CMS; LMV Vs PMC. The higher acidic pH observed among the samples of LMV when compared with other sources of CMS and PMC milk samples. The higher acidity of milk due to the activity of microbes and enzymes (Faraz et al., 2013 and Rahman 1995). In this present experiment acidity of the milk from LMV samples were high which means that the milk was not freshly processed or chilled during transportation or preservation by the vendors.

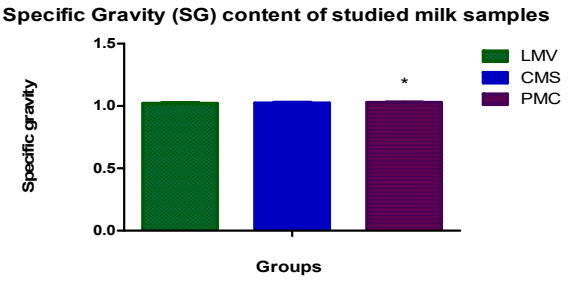

Fig. 3 Specific Gravity (SG) in Different milk samples

Specific Gravity (SG): The obtained SG values were portrayed in fig.3. The SG content from different milk samples ranges were 1.009 to 1.027 for LMV, 1.017- 1.029 for CMS and 1.027-1.030 for PMC respectively . All values were given as mean ± SD of observations. The statistical comparison between groups was done using one-way Anova followed by Tukey multiple comparison test.* = p< 0.005 on comparison between LMV Vs PMC. The obtained values of milk samples indicate that the SG content is very low in LMV followed by CMS; whereas the values of PMC were in normal range. The possibilities of low SG content were; if water has been added, the lactometer reading will be below 26 and if any solids such as flour have been added, the reading will be above 32. Mahedi (2012) stated that the SG indicates the percent of Total Solid (TS) content in milk, TS plays an important role in physico- chemical, sensory, textural characteristics and also the shelf life of any milk and milk products.

The data revealed that the edible adulterants in different sources of milk samples were presented in Table 2. The results showed that the PMC milk exhibited negative to all edible adulterants. The findings related to LMV and CMS of edible milk adulterants revealed that the starch is present only in LMV milk samples (20%); Starch is usually added to milk after addition of water as thickening agent in milk. The Obtained results were supported by Indumathi and Obulareddy (2015) and Ramya et al., (2015), they also identified starch in milk samples.

Table 2: Detection of Edible Adulterants in different milk samples

Adulterants	Local Milk Vendor's samples (10) (%)		Cooperative Milk Societies samples (8) (%)		Packed Milk Companies samples (10) (%)	
	Positive	Negative	Positive	Negative	Positive	Negative
Starch	20	80	0	100	0	100
Glucose	20	80	25	75	0	100
Sugar	30	70	25	75	0	100
NaCl	40	60	50	50	0	100
SMP	40	60	25	75	0	100

The glucose as adulterant in CMS (25%) followed by LMV milk (20%). Nirwal et al., (2013) were reported similar positive results for glucose adulteration in milk samples collected in Dehradun.
Adulterant sugar was found in some milk samples, those were LMV (30%) followed by CMS (25%). Results of current findings were similar positive sugar adulteration was observed in the studies of Singuluri and Sukumaran (2014) and Chanda et al., (2012). It indicates that the sugar is common adulterant in milk to build up the Lacto meter reading.
NaCl as an adulterant high in CMS (50%), followed by LMV (40%); It is usually used to hide the water content in milk; it gives false reading in lactometer. The present milk samples showed positive results for Nacl, which is supported by the similar results obtained by Ramya et al., (2015) and Indumathi and Obulareddy(2015). According to the ayub et al., (2007) reported that the high content of Nacl in milk can lead to health issues like kidney problems, disturbance in speech and sensory.

High percent of LMV milk samples i.e. 40 percent, followed by CMS milk samples 25 percent were adulterated by SMP. Indumathi and Obulareddy (2015) research shows 16% to 24% of samples were in positive towards adulteration with SMP. Common use of SMP was adulterated in Milk to increase accumulation of milk Total solid content.

Table 3 data revealed that the adulteration of milk by use of chemicals and preservative adulterants in different sources of milk samples. The results showed that the PMC milk exhibited negative to

all preservatives and chemical adulterants. The findings related to LMV milk samples and CMS milk samples shown some extent of adulteration by chemicals and preservatives.

Table2: Detection of Chemicals and Preservatives adulterants in different milk samples

Adulterants	Local milk vendor's samples (10) (%)		Cooperative milk societies samples (8) (%)		Packed milk (pvt companies samples (10) (%)	
	Positive	Negative	Positive	Negative	Positive	Negative
Formalin	0	100	0	100	0	100
Urea	0	100	0	100	0	100
Coloring agent	0	100	0	100	0	100
Detergents	50	50	25	75	0	100
Neutralizers	30	70	50	50	0	100

The presence of formalin, urea and coloring agent not appeared in LMV and CMS milk samples in the present study. The same findings were observed in other study conducted by Rashedul (2012).

LMV and CMS milk samples in the present study indicated some extend positive results for adulteration with detergent. The detergent adulterant was highly present in LMV milk samples (50%) followed by CMS milk samples (25%).The addition of detergent to the milk for increase the appearance of distinctive white color in the milk (Indumati and Obul Reddy, 2015). The addition of detergents in milk causes severe gastro intestinal disorders such as intestinal inflammation and gastritis problems.

Neutralizers such as sodium bicarbonate or sodium carbonate basically used to preserve milk from deterioration and adjust the milk pH. Neutralizers found high in CMS (50%), followed by LMV (30%) milk samples. The obtained results were similar to the findings of research conducted by Indumati and Obul Reddy (2015) and Ramya et al., (2015). The addition of neutralizers (sodium carbonate or sodium bicarbonate) might concern the hormonal signaling system; which is important for regulate human body development and reproduction (Manual of Methods of Analysis of Foods: Milk and Milk Products, 2005).

Table 3: KAP scores of respondents before and after awareness session on milk adulteration

Categorization of Scores	Before Awareness (100) %	After Awareness (100) %
Low < 1-10	43	10
Medium >11-20	46	52
High > 21-30	11	38

Present study was projected to found that the selected respondents KAP on milk adulteration. The obtained scored were presented in table 3. The selected respondents KAP scores before the awareness session on milk adulteration was between low to medium range, it represents the respondent's poor knowledge on milk adulteration. A majority of 46 percent of the study respondents fall under medium scores of KAP, i.e. between 11- 20 score; next followed by 43 percent of respondents in low KAP scores 1-10 and 11 percent of high KAP scores 21-30. After initial KAP, to the respondents an awareness session was given on milk adulteration. It was observed that the high percent of respondents before awareness in medium and low sores; whereas after awareness session on milk adulteration, KAP scores of the respondents were increased to medium (52%); followed by high (38) and low (10). The KAP scores of the respondents were increased after awareness session. This results indicates that the consumer awareness towards information help to create attentive about milk adulteration.

Conclusion:

The present study assessed 28 milk samples from different sources like LMV, CMS and PMC and assessed presence of various adulterants. In this different milk samples were observed that the amalgamation of edible foods (Starch, Glucose, sugar, NaCl and SMP), preservatives (Neutralizers) and detergents were as adulterants mainly in some milk samples of LMV and CMS; however, in PMC milk samples results showed negative results in all adulteration tests. KAP on milk adulteration was assessed on selected respondents to figure out the knowledge on milk adulteration. The KAP testing carried out apparently in two times; i.e. before and after awareness session on milk adulteration; outcome of the results reveal that the after awareness session on milk adulteration exhibited increase in scores of the respondents. The existing research information discovered and suggested to point towards the need of consumer awareness on milk adulteration and effect of adulterated milk consumption, its consequences on human health. Therefore, it is suggested with the intention of the consumers wellbeing; milk consumers as well as local milk vendors and Cooperative milk societies also need to aware of the good knowledge, attitude and practices on milk processing, preservation and possibilities of unintentional adulterants in milk. So the respective government organizations need to assess the ground level milk samples and also need to put into action of regulation to enhance the quality of milk.

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