Abstract: The nutritional components of milk are used by the human body to function properly as well as for providing healthy growth and development. The more intake of milk and dairy products in Pakistan makes these eatables a critical target for adulteration with financial gains for selfish producers. To perform the qualitative analysis of milk adulterants in various samples of milk, available in local market of Lahore, Pakistan. An Experimental study conducted in FAHS-Lab 101, University Institute of Diet and Nutritional Sciences, Faculty of Allied Health Sciences, The University of Lahore in duration of 04 months from February till May 2019. Qualitative analysis of milk samples was performed at FAHS lab101, total 09 samples were taken from three different varieties; fresh milk, pasteurized milk and tetra pack milk. Random sampling was used to select the milk samples, and commonly used milk varieties were chosen for the analysis. All the milk samples contained urea by giving distinct yellow color on analysis. Sorbitol was found in 6 milk samples (3 local, 1 pasteurized, and 2 tetra pack) by depiction of distinct yellow green color. Hypochlorite was found in 8 milk samples by giving distinct yellowish brown color upon analysis. Starch, water, sodium chloride, formalin and boric acid were found in none of milk samples. It was concluded from the study that various samples of milk contain adulterants as urea, sorbitol, hypochlorite, cane sugar, and carbonate by giving distinct colors. Among the milk samples pasteurized samples were having comparatively better results in context to qualitative analysis.

Keywords: Qualitative analysis, milk, adulterants, local market, sorbitol, Lahore.

INTRODUCTION: Milk is used for good health and it is the form of best staple food for the customers to consume it for maintaining health. (Hospido et al., 2016)(Iribarren et al., 2010) (de Varies et al., 2010) Milk is being adulterated for the economic point of view. Gaining profit through the adulteration of milk is a major issue for the developing countries and it has adverse effects on the health of population (Lee C et al., 2018). Now the adulteration of milk has become a serious health and safety issue for the developing countries and different tests are used to detect the presence of harmful chemicals, ingredients and adulterants in these countries. Less information is present for performing these tests for the detection and evaluation of these harmful chemicals in these developing countries. Chemical qualitative tests are the most cheaper, convenient and popular test for milk adulteration. These tests are performed with special skills (Aparnathi et al., 2019). Different tests have been used for most of the contamination and harmful chemicals and the dealers also have innovative ideas and market strategies to add the contaminants in milk (Rameshbbhai et al., 2017). The most simple and reliable test is the detection of changes in color due to the presence of contaminants in milk by adding different chemicals. These tests are carried out under precautionary measures and certain protocols for the safety and quality of milk in the laboratory of Biosafety Level 1 and the chemical reagents are used to detect the contamination. Serious issue in the use of this harmful chemicals in these developing countries.
method is that it requires specific amount of chemical which require special skills and safely. Other than this issue, it is the most reliable and simple method among all qualitative technique through which the presence of chemicals can be detected in a very short time. The traders also use some hazardous chemicals to improve its physical appearance and to increase its shelf life. These chemical have many adverse effects on human health. Different colors, soaps and detergents are also added to enhance its physical appearance (Azad, T and Ahmed, S, 2016)(Montowska and Fornal, 2018)(Valdes A et al., 2018).

Qualitative tests are done to check the quality of milk whether adulterants are present in it or not. Current study was intended to detect milk quality in context of adulterants by depiction of specific colors signifies presence and absence of specific components as chemicals etc. So that proper action can be taken by regularity authority in maintaining milk quality and to make it safe for consumption.

METHODOLOGY:

Study Sample:
Total nine samples of milk were procured from the local market of Lahore. Samples were categorized on the basis of their shelf lives, three samples were of fresh milk procured from local market (having shelf life of 6-7 hrs), three were of pasteurized milk (having shelf life of 3-4 days) and three were of tetra pack milk (having shelf life of 4-5 months). Analysis was conducted at Faculty of Allied Health Sciences lab (FAHS lab) 101, University Institute of Diet and Nutritional Sciences (UIDNS), faculty of allied health sciences (FAHS), The University of Lahore, Lahore, after taking written informed consent from the concerned authority.

Study Design:
Random sampling was used to select the milk samples, and commonly used milk varieties were choosing for the analysis. Substandard or rotten milk samples were not included; milk samples above expiry dates were also not selected. And procurement, milk samples were stored under safe and hygienic conditions in the refrigerator, and then the milk samples are thawed in the room temperature before performing the test on milk samples. The protocols of Standard Operating Procedures (SOP) were followed to use the test kit for analysis of milk adulteration.

For urea detection 1mL of milk was poured in test tube then 1mL of reagent #1 was added in a milk sample to defect urea. If urea was present then distinct and clear yellow colour appeared in the test tube, while no colour developed in negative milk. Light yellow colour was not the indication of urea because light yellow colour can also appear due to the presence of natural amount of urea in it.

The presence of hydrogen peroxide was detected by adding 2-3 drops of reagent # 3 in 1mL of milk sample in a test tube. Immediate difference in the colour was documented and the tube was not shacked during the record of results. Distinct red colour patches develop in the positive milk sample while no colour develops in negative milk sample.

For sorbitol detection 1mL of reagent #5A and 1mL of reagent #5B were added in 1mL of milk sample in test tube. Distinct yellow green colour appeared which showed the presence of sorbitol, while the appearance of reddish orange colour was the indicator of no sorbitol in milk sample.

For detection of Quaternary Ammonium Compounds (QAC) Reagent #6 with the quantity of 1mL was added was added in 1 mL of milk sample in a test tube. The appearance of distinct pink colour showed the presence of QAC in milk sample and no colour was the indication of negative milk sample. Development of mild ring of pink colour at the joints of two layers was observed as negative for quaternary ammonium compounds and emergence of pink colour in the upper layer was also observed that the sample was negative.

For detection of boric acid, 1mL milk sample was taken in the test tube and 1mL regent #7 #A along with 1mL regent #7 #B was taken. After waiting for a while, distinct orange red colour developed in positive milk sample and distinct orange yellow colour appeared in negative milk sample.

For detection of sodium chloride, 1mL milk sample was taken in test tube and 0.5 mL regent # 9A with 1mL regent # 9B were added. Distinct yellow coloured showed presence of sodium chloride and chocolate red colour appeared in negative milk sample.

For detection of carbonate 1mL of milk sample was taken in test tube and 1mL regent # 10 was added. Distinct red colour developed in positive milk sample and no colour developed in negative milk sample.

For presence of formalin 1mL milk sample was taken in test tube and 1mL regent #11 was added along the side, without shaking the test tube. Distinct purple colour ring developed at the junction of two layers in showed presence of formalin whereas yellow or brown ring developed in between the two layers in negative milk sample.

For detection of hypochlorite in a test tube 1mL of milk sample was added and 1mL regent 12#A with 1mL regent 12#B were poured. Distinct yellowish brown colour developed showed the presences of hypochlorite, no colour change showed absence of hypochlorite.
For detection of starch, 1mL milk sample was cooled down after boiling and then 2-3 drops of reagent #02 were poured in a test tube. The presence of distinct blue colour showed the contamination of milk with starch of starch in milk sample. No colour showed absence of starch in milk sample.

For detection of detergent/ soap, 1-2 drops of reagent# 4 were added in 1mL milk sample in test tube after boiling. The presence of distinct pink colour showed positive milk sample and negative milk sample showed no colour.

For detection of cane sugar, 1mL. regent #8 was boiled for 5 minutes in a 1mL of milk sample in a test tube. The appearance of distinct deep red colour showed presence of cane sugar and no colour in a test tube showed the negative milk sample.

RESULTS:
As shown in Table 1, Urea was present in almost all samples, whereas NaCl, H2O2, boric acid and formaline were not present in any of the samples. Whereas rest ones had mixed results. Among the milk samples pasteurized samples were having comparatively better results.

Table 1: Adulterants present in different milk samples

<table>
<thead>
<tr>
<th>Adulterants</th>
<th>Local milk</th>
<th>Pasteurized milk</th>
<th>Tetra pack milk</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sample 1</td>
<td>Sample 2</td>
<td>Sample 3</td>
</tr>
<tr>
<td>Urea</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Sorbitol</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>NaCl</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Hypochlorite</td>
<td>Yes</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>H2O2</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>QAC</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Boric acid</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Carbonate</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Formalin</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
</tbody>
</table>

Among the heat based adulterants (starch, detergent (saop) and cane sugar), no milk sample had starch in it. Whereas, only one milk sample of pasteurized milk had detergent in it and most of the milk samples were having cane sugar in it, Table 2.

Table 2: Heat based adulterants in milk samples

<table>
<thead>
<tr>
<th>Adulterants</th>
<th>Local milk</th>
<th>Pasteurized milk</th>
<th>Tetra pack milk</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Sample 1</td>
<td>Sample 2</td>
<td>Sample 3</td>
</tr>
<tr>
<td>Starch</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Detergent</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>Cane sugar</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
</tbody>
</table>

DISCUSSION:
Milk is rich in nutrients and it considered as a complete nutrition globally due to the presence of nutritionally rich composition so, milk and milk products are the target for adulteration (Conceicao et al., 2019). One of the most untrustworthy food processing methods is the use of milk adulterants, which have different adverse effects on human health and also causing serious diseases and also reduces the nutritional value of milk (Chakraborty and Biswas, 2018). Adulterants in milk are detected by the changes in colour due to the chemical reactions of different reagents. These reactions are valid only for the small amount of concentrations and are not precise (Azad, T and Ahmed, S, 2016). 11 potential chemicals are used as adulterants for the milk and other liquid foods which are hydrogen peroxide, sucrose, carbonate, bicarbonate, formaldehyde, chloride, starch, citrate, hydroxide, hypochloritie and water (Hansen and Holroyd, 2019). In current study, qualitative analysis was used for the detection of these adulterants present in milk. Aparnathi et al., stated that processing treatments like sterilization affect these qualitative tests significantly (Aparnathi et al., 2019). Analytical methods have been developed for most of the adulterants but some traders find more innovative ways to adulterate the milk with cheaper ingredients (Rameshbhai et al., 2017). In current study, urea, sorbitol, hypochlorite, carbonate and cane sugar were the common adulterants found in milk, most of which are edible. Recent reports have proved the presence of contaminants like urea, sucrose, maltodextrin, water, glucose, salt, gelatin, starch, and detergent for the intensions of milk adulteration (Rameshbhai et al., 2017). Azad and Ahmed, also stated that some consumable compounds are also added as adulterants which enhance the taste of the milk (Azad and Ahmed, 2016). In current study every sample urea was present as Khan et al., stated that urea is added with the water for the thick, creamy and concentrated appearance (Khan et al., 2015). Urea is added in milk which causes severe stomach ailments and may cause diseases of metabolic syndrome (Jha S.N et al., 2015; Nascimento et al., 2017; Xie et al., 2018). To expand the shelf life of milk, different chemicals are used like formaldehyde, sodium hypochlorite, sodium hydroxide and hydrogen peroxide (Guerreiro et al., 2018; Guerreiro and T.M., 2018). The appearance of milk is improved by mixing different compounds like colours,
detergents and soaps (Azad and Ahmed, 2016), which were also present in some samples.

CONCLUSION:
Adulterants are added so forth in the products to maintain demand and supply ratio and make it available for consumption. Milk is most adulterated product with different salts and chemicals as formalin, sorbitol, hypochlorite, sodium chloride, boric acid and water. Milk adulterants compromised the quality and make it hazardous for human consumption. It is concluded from qualitative analysis that milk from local markets of Lahore contains adulterants and by law action should be taken for its regulation.

Recommendations:
The current study is only qualitative analysis of liquid milk samples. Research can be carried out on qualitative analysis of dry milk powder. For more accurate results, further research can be carried out for quantitative analysis of milk adulterants.

REFERENCES: