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Research article

Indigenous knowledge on the practice of milk container fumigation and its effect on microbial safety of milk among pastoral communities in west Guji zone, southern Ethiopia

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ABSTRACT

Introduction: Milk is a high-risk food and has been implicated in many foodborne illnesses. Thus, the pastoral communities in rural Ethiopia used a traditional practice of milk container fumigation to maintain the quality and safety of milk.
Objective: to assess the indigenous knowledge on milk container fumigation practice and its effect on the microbial safety of milk among pastoral communities in the west Guji zone, southern Ethiopia.
Methods: A cross-sectional study design was conducted in six randomly selected kebeles of the pastoral districts in the west Guji zone, Southern Ethiopia from December to June/2022. The preservative plants and raw milk samples were collected and transported to the laboratory, to analyze the efficacy of plants on the microbial safety throughout milk storage. A variance analysis was used to compare the means of microbiological growth and pH measure among the treatments and control; while thematic analysis was for qualitative data.
Result: Four species of plants, namely: *Olea africana*, *Clerodendrum myricoides (Hochst) vatke*, *Rhamnus staddo*, and *Rhus natalensis* were identified from the study area; as they were used for fumigating milk storage containers to prevent a contamination of milk. According to respondents, the fumigation of milk containers was practiced by holding the container upside down over the smoke from a burning chip of each plant species. Accordingly, it was demonstrated that the *R. staddo* has relatively better efficacy in inhibiting microbial growth in milk than *O. africana* and *C. myricoides (Hochst) vatke*; while *R. natalensis* has no significant impact on microbial growth in milk over the storage period.
Conclusion: Pastoralists in the West Guji zone were fumigated the milk storage container by using smoke of *O. africana*, *R. staddo*, *C. myricoides (Hochst) vatke*, and *R. natalensis* plants. As such, it was identified that *R. staddo* has relatively better efficacy in inhibiting microbial growth in milk than *O. africana* and *C. myricoides (Hochst) vatke*; and it is a better plant to be recommended for the preservation of cow milk.

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1. Introduction

Dairy products have been utilized for human nutrition since some 11,000 years ago when farming in the Middle East began to replace hunting and gathering. Cattle herders initially began to ferment milk into yogurt and cheese to break down the milk sugar lactose, which was indigestible for adults [1]. As such, Fresh or raw cow milk as a diet contributes a great role in the survival of infants in humans all over the world [2].

Milk and milk products have been significant components in the diets of Africans and continue to play an important and increasing role in the diets of the growing population of both rural and urban communities [3]. In Ethiopia the dairy production indicated significant growth between 2000 and 2010 and the annual milk production is estimated to be 3.1 billion liters from cattle and the average milk yield per cow per day at country level is about 1.4 L. About 98% of this milk product is comes from rural areas [4].

Moreover, in pastoral communities raw milk is widely used for consumption [5]. Particularly, in Ethiopia the consumption of raw milk and its derivatives is common, which is not safe from consumer health point of view as it may lead to the transmission of various diseases [6]. Contamination of a raw milk occurs during milking, storage, transportation, and at market places [7]. These pathways for contamination have a potential to reduce a safety of milk that significantly contribute to disease transmission [8].

The world still faces great problems of food-borne diseases associated with contamination of the dairy products. Globally, it is estimated about 1.8 million childhood deaths annually, are due to dairy-related diarrheal illness [1](2). Milk-borne pathogens are of public health concern in many developing countries. This was evidenced by the study conducted in Mali, which found an increased risk of food-related diarrhea or vomiting in children consuming milk products [9].

Similarly, an observational epidemiological study conducted in Kenya reported that camel milk market chains pose the greatest risk for foodborne gastrointestinal illnesses (diarrhea and/or vomiting) [10]. Thus, there is a need for a sustainable system of preventive measures that minimize the safety risks associated with a raw milk consumption [11].

Ideally, to prevent raw milk from contamination and ensure high-quality of dairy products, milk should be stored immediately in refrigerator below 4^{O}C. In practice, this is not possible for most small-scale producers in Africa, where modern cooling facilities including mechanical refrigeration or cooling tanks are not available; mainly due to high initial investment and running costs and technical problems, including the lack or unreliable supply of electricity [12].

In most rural areas of Ethiopia pastoral communities used an indigenous knowledge and practices to prevent milk from contamination [3]. As such, smoking of milk vessels with splinters of different tree species is commonly used as a milk preservation method in different parts of Ethiopia [11,13]. This traditional practice is locally known as 'korasuma' among the pastoralists of the west Guji zone, southern Ethiopia.

However, the type of plants used for smoking milk utensils and their method of application differs from place to place; even from household to household [11]. Besides, the pastoralists in the study area are believed that the smoke of all wood plants is considered equally effective in improving the quality and microbial safety of milk. Therefore, this study is expected to fill the gap by identifying the types of plants used for smoking milk storage containers, their application techniques and evaluate the efficacy of each preservative plant on the microbial growth in milk.

2. Methods and materials

2.1. Description of study area and period

This study was conducted in pastoral communities of west Guji Zone, Southern Ethiopia from December 2021–June/2022. Bule Hora town is the capital of West Guji Zone which is located 467 KM south of Addis Ababa, Ethiopia. The annual average temperature is 15 °C and rainfall ranges from 500 to 1250 mm. The Zone consists of one town administration and 9 districts (i.e., six agro-pastoralists and three of them are pastoralists). The study was particularly conducted in the three pastoral districts (Suro Magada, Dugda Dawa, and Melka Soda) [14,15].

2.2. Study design

➢ A cross-sectional study design was used.

2.3. Sampling technique and sample size

Six peasant associations (PAs) or kebeles were selected randomly (i.e., two from each of the pastoral districts found in the West Guji zone). From each of the selected Kebeles, twenty households were randomly selected and thus a total of 120 households were interviewed using a semi-structured questionnaire. Besides, twenty-four willing households (eight from each district) who had two or more lactating cows were randomly selected to provide the milk samples. A direct observation of the preservative plants and milk storage containers to be used by pastoralists was also made for cross-checking.

2.4. Field data collection

Data was collected by using a semi-structured questionnaire. The interview was conducted orally with female farmers in a face-to-face manner by trained enumerators who were conversant in the local dialect. The questionnaire was initially drafted in English language and after reviewing different literature it was translated into the Afan Oromo language following a local dialect. Generally, six enumerators was recruited to manage the formal survey.

2.5. Milk sample collection

About 450–500 ml of a raw milk sample was taken from 24 households that own more than three lactating cows proving that they did not fumigate their milk containers following the ISO 707 standard [16]. Subsequently, the samples were put into an icebox containing ice packs and then transported to the laboratory.

2.6. Plant sample collection

The preservative plant species and the woods that the community used for milk container fumigation were identified and collected from the different pastoral villages of the West Guji zone with the help of local pastoralists. The collected plant specimens were then coded by their local names, and transported to the Biology laboratory, at Bule Hora University for identification of their scientific name.

2.7. Container collection and fumigation method

The calabash containers that the community used for storing milk were collected from pastoral households, while the plastic containers were brought to the laboratory by buying them from the market. In the laboratory, the containers were fumigated following similar techniques of applications and procedures that were observed at households during the survey. Accordingly, the milk storage container was prepared by washing it thoroughly with clean water and dried. Then, it was fumigated by holding the container upside down over the smoke from burning dry chips of the plant for a minimum of 20 min.

2.8. Laboratory procedure

In the laboratory, a milk sample was thoroughly mixed and a liter of the milk was distributed into a control and experimental container for each plant and allowed to store by keeping at room temperature for about 24 h. The control containers were prepared in the laboratory by cleaning thoroughly only with tap water and dried; while the treatment container was prepared by cleansing and fumigating.

Microbial analysis: The milk samples were analyzed for total bacterial count (TBC) and pH before storage, and two times at 12-h intervals for two days.

- **Total bacterial count (TBC):** The Total Bacterial Count (TBC) involves growing bacteria into colonies on an agar gel, which contains nutrients to support microbial growth. A standard protocol was followed for the determination of the TBC of the milk sample by using the pour plate method on plate count agar (PCA) (Oxoid, UK). The milk sample was serially diluted by transferring 1 ml of milk to a universal tube containing 9 ml of buffered peptone water and added to the agar in a sterile container. Then the Petri dish was incubated at 37 °C for two days.
- **PH:** The PH value in the milk sample was measured before storage and two times at 12-h intervals after storage for two days by using a pH meter (PH-870 Model).

2.9. Study variables

2.9.1. Dependent variables

- Total bacterial count
- pH

2.9.2. Independent variables

- Type of plant
- Type of container

2.10. Data analysis

The qualitative data from the interview was analyzed by theme. Thematic analysis of the data was conducted by sorting and organizing information according to their thematic similarities and differences. Then, the information was categorized and studied to understand their relationships in the overall context of the study. The quantitative data was entered into Epi-Data version 3.1 and exported to SPSS version 25 for analysis. The significant variation in mean TBC among each treatments and control was tested by two independent samples T-test; while two-way ANOVA is used to analyze the mean variation in the total bacterial count and PH measure among treatment groups and control group in different containers.

2.11. Data quality assurance

For data quality assurance, the data collection tool was pre-tested before the actual survey. The administration of the interview was carried out by enumerators who spoke the local dialect (Afan Oromo). They were also trained and closely supervised by the researchers in the field. Moreover, Laboratory equipment was sterilized and a well-trained laboratory technician was used to conduct laboratory tests under aseptic conditions.

3. Result

3.1. The plants used for milk container fumigation

The most commonly used plants for fumigation milk containers among the pastoralists living in the west Guji zone, southern part of Ethiopia were: Ejersa (*Olea africana*), Tulange (*Clerodendrum myricoides (Hochst) vatke*), Kadida (*Rhamnus staddo*), and Xaaxessaa (*Rhus natalensis*) as reported by respondents (Table 1). Among the identified plants, *O. africana* was reported by all of the respondents 120 (100%); while *C. myricoides (Hochst) vatke*, *R. staddo*, and *R. natalensis* were reported only by 66 (55%), 15 (12.5%) and 42 (35%) of women pastoralists living in the three pastoral districts of this zone respectively.

3.2. Indigenous knowledge of milk fumigation practice

Pastoralists in the west Guji zone used different containers to store cow milk. It was observed when the women pastoralists used traditional containers made from calabash trees and plastic containers to store raw cow milk (Fig. 1A and B). According to the respondents, the containers were fumigated either by holding them upside down on the smoke of burning dried chips of the plants or by putting the burning chips inside the container until the smoke died. In addition, the burning end of the plant can be continuously rubbed against the internal wall of containers. After such vigorous rubbing for minutes, the finely powdered wood charcoal was removed from the container and the milk was stored in it.

3.3. Effect of plants on microbial growth in cow milk

Fig. 2 below shows the average load of total bacterial count in a milk sample stored in a plastic and calabash container throughout milk storage. The average TBC in raw milk samples was determined to be 7.6 $\pm$ 1.03 log CFU/ml before being stored in different containers. However, after storage; the value of TBC tended to increase over time up to the 36 h of storage in all containers and slightly reduced thereafter. In both plastic and calabash containers, the lowest total bacterial count throughout storage was obtained from a milk sample stored in a container fumigated with *R. staddo* smoke, while the highest bacterial count was recorded in a milk sample stored in an untreated (control) container both plastic and calabash containers. The mean TBC measured at 12, 24, 36, and 48hrs of storage was significantly varied among the fumigated containers and un-fumigated (control) containers in both plastic ($F = 99.403$; $p = 0.000$) and calabash containers ($F = 101.138$; $p = 0.000$).

3.4. Comparison of the microbial load in fumigated and un-fumigated containers

The microbial growth of total mesophilic bacteria, in milk stored in a container treated with *O. africana*, *R. staddo*, and *C. myricoides (Hochst) Vatke*, showed a significant change over 12, 24, 36, and 48 h periods of storage relative to the un-fumigated container as seen in Table 2 below ($p < 0.05$). As compared to the total bacterial count in an un-fumigated container, the highest significant difference in mean TBC was observed from the milk sample treated with *R. staddo*; while the lowest was seen in the milk sample stored in a container treated with *C. myricoides (Hochst) vatke* in both plastic and calabash container. On the contrary, there was no observed significant difference in mean TBC measured at 12, 24, 36, and 48 h in a milk sample stored in a container fumigated with *R. natalensis* relative to the un-fumigated container in both plastic and calabash containers ($P > 0.05$).

Table 1
The plants used for fumigating milk container that identified from pastoral communities of west Guji zone, southern Ethiopia, 2022.

Name of the plants			Parts used	Forms used
Local language (Afaan Oromo)	Scientific	Family		
Ejersa	*Olea africana*	*Oleaceae*	Stem and Root	Dry and fresh
Tulange	*Clerodendrum myricoides (Hochst) Vatke*	*Lamiaceae*	Stem	Dry
Xaaxessaa	*Rhus natalensis*	*Verbenaceae*	Stem	Dry
Qadiidaa	*Rhamnus staddo*	*Rhamnaceae*	Stem	Dry

Fig. 1. The photo shows the milk storage container used by the women of the Guji pastoral communities in West Guji zone, southern Ethiopia.

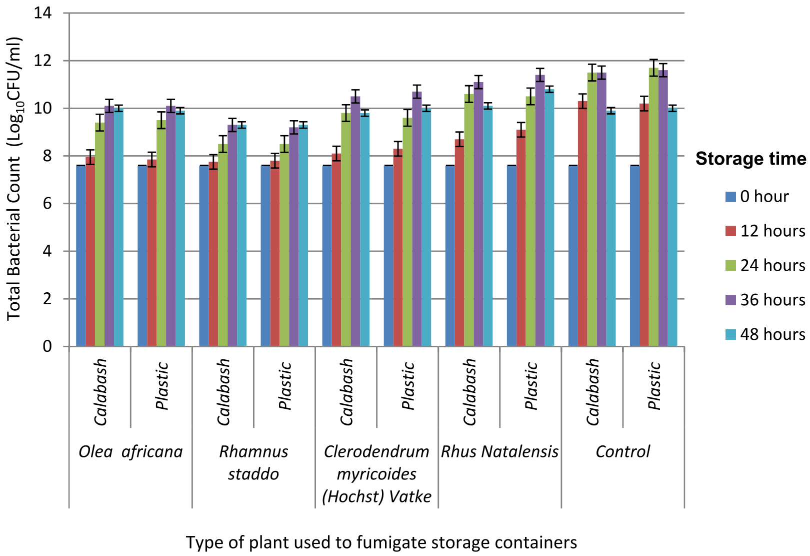

Fig. 2. The mean total bacterial count in a plastic and calabash container fumigated with different plant at different period of storage.

Table 2
The mean total bacterial count in containers fumigated with different wood smoke at different period of storage.

Type of plant	Type of container	Storage Time					Sig. level	
		0 h	12 h	24 h	36 h	48 h	t-value 95% CI	*P*-value
Olea africana	Calabash	7.6	7.95	9.4	10.1	10	−2.426	0.023
	Plastic	7.6	7.85	9.5	10.1	9.9	−2.524	0.018
Rhamnus staddo	Calabash	7.6	7.75	8.5	9.3	9.3	−3.884	0.001
	Plastic	7.6	7.8	8.5	9.2	9.3	−3.962	0.001
Clerodendrum myricoides (Hochst) Vatke	Calabash	7.6	8.1	9.8	10.5	9.8	−2.062	0.049
	Plastic	7.6	8.3	9.6	10.7	9.7	−2.104	0.045
Rhus natalensis	Calabash	7.6	8.7	10.6	11.1	10.1	−1.047	0.304
	Plastic	7.6	9.1	10.5	11.4	10.8	0.629	0.534
Control	Calabash	7.6	10.3	11.5	11.5	9.9		
	Plastic	7.6	10.2	11.7	11.6	10		

3.5. Comparison of the type storage container on bacterial load

As shown in Table 3 below, the mean comparisons of total bacterial counts measured at 12, 24, 36, and 48 h were made considering the type of storage containers. It was found that there is no statistically significant difference in average total bacterial count measured throughout storage, due to the container difference which is in plastic and calabash containers ($p > 0.05$).

3.6. Effect of fumigation on the PH of milk

As shown on Fig. 3 above the pH of the raw milk sample was determined to be 6.14. However, after storage the pH of milk was decreased gradually along the storage period and reached 4.13–4.96 in all containers. Relatively, the rate of pH decrease was high in milk samples from un-fumigated containers in both plastic and calabash containers, while the lowest decrease in pH value was observed in a milk sample stored in a container fumigated with the smoke of *R. staddo*, and reached about 4.95 and 4.96 in plastic and calabash container respectively. The pH value measured at different storage periods was significantly varied among the containers in both calabash ($F = 190.155$; $p = 0.000$) and plastic container ($F = 916.054$; $p = 0.000$). However, there is no observed statistically significant pH value due to the container difference as indicated in the above figure ($P > 0.05$).

4. Discussion

The fumigation of milk storage containers by smoke from smoldering wood of different plant species is one of the cultural practices used to preserve milk for a long period; among the pastoral communities of the west Guji zone. The result of this study identified that plants such as *O. africana*, *R. staddo*, *C. myricoides (Hochst) vatke***,** and *R. natalensis* were used in the study area for fumigation of the milk storage container. Among the identified plants, *O. africana* is the most frequently used plant for fumigating milk storage containers; as it was reported by the respondents of the three districts. The finding of this study was in line with the studies conducted in different zones of Ethiopia; Borena zone [17], west Wolega [18], west and east Shewa [19](20), central highlands of Ethiopia [21], and also neighboring country, Kenya [7](22) that reported *O. africana* as a most commonly used plant for fumigating milk handling and storage equipment. This might be associated with the abundance or/and distribution of *O. africana* plant species in wide agroecological zones.

But this finding is different from the study conducted in northern Ethiopia [13], which reported burning chips of plants like *Acacia etabaica*, *Olea europaea* ssp. *cuspidata*, and *Cadia purpura* were used for fumigating milk storage containers. In addition Except for *O. africana*, different findings were reported from other studies conducted in northern Ethiopia [13,21,23,24], that identified plants like *Clerodendrum myricoides*, *Terminalia brownie*, *Juniperous procera*; *Acacia etbaica*, *Olea europaea* ssp. *cuspidata* and *Cadia purpurea*, *Vernonia amygdalina*, *Aoe elegans*, *Solanum schimperianum hochst*, as they are used for smoking milk storage containers in the study area. This variation in the type of preservative plants among the communities in different parts of Ethiopia might be associated with possible variability in the abundance of the plant species and specific indigenous knowledge of the communities. Likewise, other researchers have stated that local topographic attributes like altitude also bring variance in the relative abundance of plants, as stated in the previous research [20].

According to the respondents, the fumigation of the milk storage container was practiced either by holding the milk storage container upside down on the smoke of burning chips from the plants or by putting the burning chips inside the utensils and continuously spinning the container until the smoke dies. In addition, the burning end of the plants was rubbed against the internal wall of containers and removed from the inside of the container before milk was poured into it. This technique of application was similar to that practiced in communities living in the northern [23]**,** eastern [25]**,** and central [20] parts of Ethiopia and the neighboring Country, Kenya [22].

The result of this study tested that the average TBC in milk samples collected from households before being stored in different containers was 7.6 ± 1.03 log CFU/ml. However, after milk was stored in the fumigated and un-fumigated containers; the value of TBC measured at different times tended to increase up to 36 h of storage in different containers and very slightly reduced thereafter. It observed that mean TBC measured at 12, 24, 36, and 48hrs periods were significantly varied among the containers in both plastic ($p = 0.000$) and calabash containers **(**$p = 0.000$**).** This indicates the milk container fumigation had an inhibitory effect on microbial growth in milk, as was reported in different previous studies [21](18) [26](17) [24](20) [25](13) [6].

Relatively, as compared to the TBC in the milk sample from the un-fumigated container, the highest significance difference in the mean total bacterial count was obtained in the milk sample stored in a container fumigated with *R. staddo* at 12 h, 24 h, and 36 h and 48 h periods while the lowest significant mean difference means total bacterial count was observed in a milk sample that fumigated with *C. myricoides (Hochst) Vatke* in both plastic and calabash container. However, fumigation of the milk container with Rhus natalensis has no significant contribution to microbial growth change throughout milk storage. This indicated that the fumigation had

Table 3
The load of total bacteria count in plastic and calabash container in pastoral communities of west Guji zone, southern Ethiopia, 2022.

Type of plant	Type of container	TBC					Sig.level	
		0 h	12 h	24 h	36 h	48 h	t-value 95% CI	*P*-value
Olea africana	Calabash	7.6	7.95	9.4	10.1	10	0.05	0.96
	Plastic	7.6	7.85	9.5	10.1	9.9		
Rhamnus staddo	Calabash	7.6	7.75	8.5	9.3	9.3	0.036	0.971
	Plastic	7.6	7.8	8.5	9.2	9.3		
Clerodendrum myricoides (Hochst) Vatke	Calabash	7.6	8.1	9.8	10.5	9.8	−0.188	0.852
	Plastic	7.6	8.3	9.6	10.7	10		
Rhus natalensis	Calabash	7.6	8.7	10.6	11.1	10.1	−0.517	0.609
	Plastic	7.6	9.1	10.5	11.4	10.8		
Control	Calabash	7.6	10.3	11.5	11.5	9.9	−0.109	0.914
	Plastic	7.6	10.2	11.7	11.6	10		

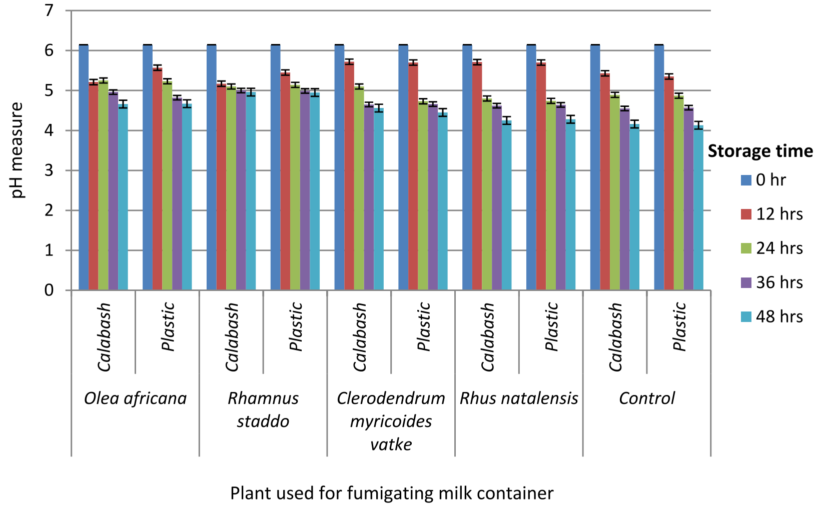

Fig. 3. Change in pH measure in milk samples during storage in plastic and calabash container over storage period.

a significant contribution to microbial growth change in the container fumigated with *R. staddo*, *O. africana*, and *C. myricoides (Hochst) Vatke* plant relative to the un-fumigated container of their counterpart.

The findings of this study that revealed *O. africana* treated milk has the lowest mean TBC relative to other plants, in the absence of *R. staddo* was in line with the findings of the different previous studies conducted in Kenya [27], and Ethiopia [24]. But this was different compared with the finding reported from the study conducted in the Tigray region, Ethiopia [13], which revealed the *Acacia etabaica* smoked containers resulted in lower microbial counts in fermented milk as compared to the other smoking chips *Olea europaea* ssp. *Cuspidate* smoked container. This difference in mean TBC in the milk sample that was fumigated with the smoke of different plants might have been attributed to the phytochemicals in the plant extracts that might have influenced the microbial growth in milk as mentioned in the previous study conducted in Kenya [27].

Furthermore, the result of this study showed the PH of raw milk as it was collected from the households before storage was 6.14. However, this result was significantly decreased over the storage period and reached 4.13–4.96 after 48hrs of storage in all fumigated and un-fumigated containers. A similar finding in the decrease in pH of milk over the storage period was reported by a national study [25], and another study conducted in Kenya [27]. This is obvious that during milk storage, the pH was decreased because of the fermentation of lactose to lactic acid by lactose-fermenting microorganisms as mentioned in the previous study [21]. However, the variation among plants may be attributed to the phytochemical contents of the plants as it was mentioned in a previous study [25]. Relatively, the rate of pH decrease was low in a milk sample stored in a container fumigated with the smoke of *R. staddo* as compared to other plants. At the end of the fermentation period, the pH recorded in milk samples treated with *R. staddo* was 4.95 and 4.96 in plastic and calabash containers respectively. Besides, the mean TBC measure in the milk sample treated with this plant was lower compared to other plants, as indicated above in the result part of this study. This follows the general pathway of milk fermentation, the production of lactic acid from lactose by lactobacillus bacteria in the milk lowers the pH of milk and the milk forms a curd. So, preventing or stabilizing the growth of lactose fermenting microbes in milk helps to prevent coagulation and improve the shelf life or the quality of raw milk.

5. Conclusion and recommendation

Pastoralists in the West Guji zone were practicing the fumigation of milk storage containers by using plant species such as *O. africana*, *R. staddo*, *C. myricoides (Hochst) Vatke* and *R. natalensis*; to prevent milk contamination. Moreover, it was demonstrated that fumigation of milk storage container with *R. staddo* have better efficacy in hindering microbial growth in milk followed by *O. africana*, and *C. myricoides (Hochst) Vatke*, and further studies are needed to investigate the phytochemicals contents of this tree species, and the effect of different fumigation methods on total bacterial count. In this study, even though the fumigation techniques could have a reduction effect on load of TBC, it is not per se an indication of milk safety as a pathogenic bacteria can persist in a milk following fumigation. Moreover, fumigation techniques can produce Polycyclic Aromatic Hydrocarbons, toxic compounds whose concentration must be further investigated in this type of milk storage.

Ethics statement

Ethical clearance letter (Ref. No. BHU/RPD1419/2021) was obtained from the Ethical Review Board of Bule Hora University. A formal letter of cooperation (Ref. No. BHU/RPD/1037/21) was obtained from the Research and publication directorate of the University to communicate with local administrative bodies in the west Guji zone. Verbal consent was also obtained from each participant before the interview to confirm their willingness to participate. Furthermore, before data collection, the participants were informed about the purpose of the study, and their right to refuse participation and continue the interview.

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Data availability statement

Data will be made available on request.

CRediT authorship contribution statement

Hailu Lemma: Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Software, Resources, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Lechisa Asefa:** Validation, Methodology, Formal analysis. **Tibesso Gemechu:** Writing – original draft, Supervision, Methodology. **Fitsum Demissie:** Supervision, Funding acquisition. **Boko Loka:** Supervision. **Degefa Dhengesu:** Investigation, Formal analysis. **Gudeta Kumela:** Supervision, Software. **Gedeno Karbana:** Investigation, Formal analysis. **Chala Daba:** Supervision, Investigation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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