

Assessment of the Impact of Sheep and Goat Pox Lesions on Skin Quality in Selected Ware Houses of Central Ethiopia

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Background: The study was performed from January 2016 to May 2017 with the aim of identifying the impact of sheep and goat pox in skin quality in selected localities of central Ethiopia.

Materials and Methods: A total of 1360 fresh, salted and dried skins were examined and graded. Pox lesion positive skins were examined by PCR to identify the agent.

Results: The prevalence of sheep and goat in the study areas was 4.022%. From a total of 86 rejected skins, a large number of skins were in the rejected category (2.78%). There were highly statistically significant associations ($p < 0.05$) between sheep and goat pox and species and study areas. However, there were no statically significant associations ($p > 0.05$) between skin size and sheep and pox. Molecular diagnosis of the case revealed that goat pox virus was responsible for the lesion.

Conclusion: Goat pox virus is the main cause of skin rejection and the factor that causes downgrading of skin.

Keywords: central Ethiopia, sheep and goat pox, skin quality

Introduction

Ethiopia has a huge livestock population in Africa, possessing more than 56.7 million cattle, 29.3 million sheep, 29.1 million goats, 1.5 million camels, 7 million equines and 52 million chickens.¹ This makes Ethiopia stand second next to Nigeria, by the number of small ruminants in Africa.² Annually, 16.6 million small ruminant skins are produced in the country, among these 33% and 32.5% were obtained from sheep and goats, respectively.³ Despite a huge potential of off take rates; the production of high-quality skins was curtailed.^{4,5}

Fish, birds, and reptiles, as well as wild and domesticated animals, can provide hides and skins. Cattle, sheep, and goats are the most important sources of hide and skin in Ethiopia. In 1998/99, the potential production is projected to 2.38 million cow hides, 10.07 million sheep skins, and 7.38 million goatskins based on annual off-take rates of 7% for cattle, 33% for sheep, and 35% for goats. This leather industry's raw material is mostly obtained from rural parts of the country where basic facilities for slaughtering and subsequent marketing are either non-existent or non-existent. Hide obtained from cattle, as well as skins obtained from goats and sheep, are country's major items that account for the vast majority of agricultural export commodities followed by live animals.⁶

Sheep and goats play a substantial role in the gross domestic product of the country to date, the benefits gained from these small ruminants are fraught by

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different constraints. Livestock diseases are amongst the significant practical precincts that have slowed down the progress of the livestock sector by decreasing production and impeding the trade of live animal and animal products.^{7,8} Among these infectious diseases, sheep and goat pox are the main problem and widely spread diseases in all regions of Ethiopia.²

More than 35% of sheep and 56% of goat skins have downgraded and rejected due to pre- and post-slaughter defects. The majority of defects are caused by pre- and post-slaughter defects due to skin diseases and poor because of various antemortem and postmortem defects caused by poor animal husbandry and nutrition, skin diseases and parasites, improper slaughter and flaying operations and improper practices of curing, collection, transportation and storage.⁹ Tanneries state that only 10% to 15% of harvested skins qualify for top grades, with the rest downgraded and rejected mainly due to deterioration of skin quality owing to skin diseases and various defects.¹⁰ Sheep and goat pox is among the disease which is responsible for deterioration of the quality of skin.

Sheep and goat pox virus (SGPV) is highly contagious viral disease of sheep and goats, in the genus Capripox virus (CaPV), subfamily Chordopoxvirinae, and family Poxviridae.¹¹ The sheep pox virus (SPPV), goat pox virus (GTPV), and lumpy skin disease virus (LSDV) showed 96% nucleotide and amino acid similarity over their entire length.¹² The central genomic region surrounded by two identical inverted terminal repeats (ITR) at the ends of SGPV (ORFs 024 to 123) comprises homologues of conserved genes involved in basic replication systems as well as in viral DNA replication, transcription, RNA modification, and structure and assembly of intracellular mature and extracellular enveloped virions.^{12–14} While the terminal open reading frame (ORF) (001 to 023 and 124 to 156) contain genes involving with virulence, host immune evasion and host range functions.¹² Mature virion cell attachment (P32) comprises the main antigenic determinants which are important for the pathogenicity and diagnosis of the virus.¹⁵

In endemic areas losses due to SGP include the direct loss; mortality, and the indirect losses include reduced milk yield, weight loss, increased abortion rates, damage to skin, and increased susceptibility to pneumonia and fly strike.¹⁶

The effect of SGPV on the skin is very high. Lesions developed secondary to virus invasion of the epithelium,

ischemic necrosis produced by vascular damage and stimulates the host cell DNA replication which causes epidermal hyperplasia.¹⁷ The epidermis shows hydropic degeneration and ruptured vesicles at some places exposing the dermis.¹⁸ Edema, fibroblast proliferation, and accumulation of cellular exudates in the stained section of the dermis.¹⁹ Extravasations of erythrocytes and coagulative necrosis with effusion of inflammatory cells intermixed with tissue debris was noticed in the hypodermis.¹⁸ The impact of these pathogenesis mechanisms of the virus may also affect the tannery industry in countries like Ethiopia where leather is the largest source of foreign currency. Now days, the current foreign trade revenue of hide and skin has dropped by 9–10% on domestic and export markets.²⁰ In Ethiopia, it is projected that a higher proportion of skin defects are developed ante-mortem.^{5,21} Among infectious diseases, sheep and goat pox are the major trait for the small ruminant sector and the second largest cause of skin rejection next to parasitic causes.²² Skin grades on skin collected from different areas of Ethiopia showed that greater than 85% hide was rejected due to pox and less than 6% was graded 1–4.²² The main aim of this research is to assess the impact of SGP in warehouses and the quality impacts of SGP on skin in ware houses.

Materials and Methods

Study Area

The research was conducted in West Shewa, East and west Arsi. Arsi Negele is located in the west Arsi zone of Oromia regional state. Arsi Negele is located at a longitude of 7°21' N and latitude of 38°42'E and 2043 meters above sea level. Ziway is located at the longitude of 7°56' N and latitude of 38°43' E and 1636 meters above sea level. Dhera is located 30 Km away from Adama on the highway connecting Adama to Assela. Dhera is located at the longitude of 8°15' N and latitude of 39°20' E and 2430 meters above sea level¹ (CSA, 2015). These areas receive bimodal rainfall and local farmers practice rain feed agriculture and also practice irrigation.

Sample Size

A cluster sampling method was applied to identify skin defects from skins collected from three districts. Sample size was estimated according to Thrusfield,²³ the sample size was 384 where an expected prevalence of 50% is to be estimated with a desired absolute precision of ±5%. To

maximize the accuracy of the data produced by the survey, we sampled more than the average.

Study Design

Three (each with a potential of collecting >5000 skins per annum) private skin collection shades/stores were purposively selected based on proximity to the transport access. Twenty percent of the warehouses' sheep and goat skins (fresh, salted or air-dried skins) were selected using a simple random sampling method. Complete physical examination was performed on randomly selected skins and data were recorded to generate information related to sheep and goat pox like; distribution of the lesion, lesion type, size, and skin grade. The skin was categorized by size, preservation methods (fresh, salted, and air dried) and skin defects caused by sheep and goat pox.²⁴ Skin inspection was made by day light to check for any defects.

Laboratory Examination

Skin biopsies for virus genome detection were collected from skin lesions and placed in a sterile screw-capped test tubes and placed immediately on an icebox and then in -20°C and sent to the department of Molecular biology in National Veterinary Institute (NVI), Ethiopia, for molecular diagnosis.²⁵ Skin tissues were rinsed with phosphate buffered saline 3 times. Skin tissues were analyzed, minced with a sterile scissor and crushed with a sterile pestle and mortar as described by Mangana-Vougiouka et al.^{26,27} The viral genomic DNA extraction was done using the QIAamp DNA Mini Kit (QIAGEN, Hilden, Germany) using the manufacturer's instructions as a base and finally the DNA was eluted using 50 μL elution buffer. The PCR was conducted using RNA polymerase 30KDa (RPO30) primers: Forward primer 5' TCTATGTTCTTGA TATGTGGTGGTAG 3' and Reverse primer 5' AGTGATTAGGTGGTGTATTATTTTCC 3'. Polymerase chain reaction (PCR) was carried out in a 25 μL reaction volume in a 200 μL capacity PCR tube containing 12.5 μL Maxima Hot Start Green PCR Master Mix (QIAGEN, Hilden, Germany), 0.5 μL of each primer (10pmol/ μL), 1 μL of extracted DNA and 10.5 μL of nuclease free water. The amplification was performed according to Lamien et al²⁸ in a thermocycler (Eppendorf AG, Hamburg, Germany) adjusted as denaturation (95°C for 4 min), followed by 35 cycles of denaturation (95°C for 30 sec), annealing (55°C for 30 sec) and extension (72°C for 30 sec) and final extension (72°C for 5 min). Three percent ultrapure, electrophoresis grade agarose gel containing

1 $\mu\text{g}/\text{mL}$ Ethidium Bromide in TAE buffer, was prepared and casted in a mold. When the gel was completely solidified, the combs was removed carefully and the gel was placed in the electrophoresis tank containing 1X TAE running buffer before loading the samples. In the centre of the first well, 5 μL of 100bp DNA ladder was loaded, while in the remaining wells, 6 μL of sample DNA with 2 μL of DNA loading dye (50% glycerol, 6x TAE, 1% bromophenol blue) was loaded by using micropipettes. After electrophoresis on 3% agarose gel (1hr at 100V) the PCR product was visualized on a UV Transilluminator (UVtec, Cambridge, UK).

Data Analysis

The data was collected by a pretested questionnaire and filled in a spread sheet Excel analysis was by using SPSS version 24 (SPSS, Inc.) and a significant association between variables was said to exist if the computed P-value is ≤ 0.05 . Student's *t*-test was applied to see the difference between species, preservatives used and study area.

Results

A total of 2014 skins (998 sheep skin and 1016 goat skin) were examined during the study period (Table 1). The prevalence of SGP in warehouse was 4.02% (n= 81). The vast majority of the SPG lesions were scars (n=45), followed by nodules and papule (n=19) and (n=13), respectively. According to Ethiopian Standard Authority (ESA, 2012), only a small proportion of shoat skins drop in the extra small (1%), very small (11.56%), and extra-large category (0.145%). Large proportion of shoat skin were categorized under the small (21.6%), medium (31.1%), large (25.76%), and very large (8.6%) categories.

Despite a large number of SGP, positive skins fall in small, medium, and large categories; there was no substantial variance between different sizes and SGP (P.0.05) as shown in table. However, there was a significant difference between species ($\chi^2=8.314$; P=0.016) and study area ($\chi^2=53.647$; P=0.000).

As depicted in Table 2 SGP was responsible for an epic downgrading of skin (n= 27, 1.3% fall in grade 5) and rejection of skin (n=21, 1%), and usually a large proportion of the affected skin fall in grades 4, 5, 6 and reject category. Among rejected skins (n=79), 25% was due to SGP (Table 2). Furthermore, there is a strong correlation and association between skin down grading and SGP (P < 0.05).

All samples, whether it was collected from sheep or goat, were given a 172 bp sized DNA product during RPO30 gene

Table 1 The Distribution of SGP in Terms of Spp., Size, Study Area, Preservation and Lesion Types

| Variables | Classes | N | SGP | χ^2 | P value |
|--------------|-------------|------|-----|----------|---------|
| Species | Ovine | 1016 | 49 | 8.314 | 0.016 |
| | Caprine | 998 | 32 | | |
| Size | Extra-Small | 22 | 0 | 11.525 | 0.485 |
| | Very Small | 233 | 8 | | |
| | Small | 436 | 17 | | |
| | Medium | 627 | 26 | | |
| | Large | 519 | 23 | | |
| | Very Large | 174 | 4 | | |
| | Extra-Large | 3 | 0 | | |
| Study Area | A. Negele | 804 | 2 | 53.647 | 0.000 |
| | Ziway | 700 | 53 | | |
| | Dhera | 510 | 26 | | |
| Preservation | Fresh | 832 | 46 | 12.271 | 0.015 |
| | Dry | 110 | 1 | | |
| | Salted | 1072 | 34 | | |
| Viral Lesion | Macule | 12 | 12 | 2067.723 | 0.000 |
| | Nodule | 19 | 19 | | |
| | Papule | 13 | 5 | | |
| | Scar | 45 | 45 | | |

Note: n=2014.

amplification. All samples, whether it was collected from sheep or goat, were given a 172bp sized DNA product during RPO30 gene amplification. RPO30 gene of SPPV has 151bp size while the GTPV and LSDV have 172bp size.

Discussion

A lot of works done on skin quality fail to describe problems that can downgrade skin quality infectious diseases like sheep and goat pox (SGP). This study discloses the most significant aspects of SGP in the quality of skin in warehouses.²⁹ Epithelial regeneration from the underneath of the scab takes several weeks. The disease causes irreversible damage

to the skin and a star-shaped, hairless or wool less scar is formed. The scar affects the grain side of the skin.³⁰

The total prevalence of SGP in warehouses in this study was 4.02%; 2.28% and 1.69% of the pox lesion was observed on fresh skin and salted skin, respectively. Our finding is in line with other findings.^{21,31} Kahsay et al²¹ documented that the prevalence of SGP in salted skin and dry skin was 15% and 6.8%, respectively. While Tsigab et al³¹ reported that the prevalence of pox lesion was 3.6% and 4% in dry skin and wet skin, respectively. Pox lesions were observed more often on wet blue goat skins (10.8%) and wet blue hides (8.1%) than in pickled sheep skins (1.2%).²¹

In Tannery-based studies conducted in Bahirdar tannery and Modjo export tannery, the prevalence of SGP was 10%³² and 9.5%.³³ Usually, the prevalence of SGP lesions in tanneries is by far larger than that of warehouses. After the tanning process, fully recovered lesions become more protuberant in the form of white spots on the skin and usually confusing Cokkel scars.³⁰

In Ethiopia, very limited work has been done on sheep and goat pox virus but some researches have been made on participatory disease surveillance (PDS) in selected districts of Afar region and Northeastern part of Ethiopia and central Ethiopia Gari et al³⁴ and seroprevalence and distribution of sheep and goat pox virus in Northwest Amhara region Ethiopia were reported by Fentie et al.² Furthermore, isolation and characterization of poxvirus was done by Demena³⁵ in west Shoa and central Ethiopia. A report on epidemiology and economic importance of sheep and goat pox is highly distributed in all regions of Ethiopia and economically important due to production loss and mortality.^{8,2}

According to Assefa et al,³² large and very large skins are highly affected and 90% of the affected skin classified in the reject category. However, in our study, a substantial number of skin fell in grade 2, 4, and rejected category (69.13%). A study conducted in Punjab, Pakistan, old

Table 2 The Impact of SGP on the Total Grade of the Skin

| Grand Grade | n | Percentage | SGP | Percentage of SGP (n=2014) | Proportion of SGP for Downgrade (%) |
|----------------------------|------|------------|-----|----------------------------|-------------------------------------|
| 1 | 402 | 19.95 | – | – | – |
| 2 | 512 | 25.53 | 15 | 0.7 | 2.93 |
| 3 | 652 | 32.518 | – | – | – |
| 4 | 268 | 13.5 | 10 | 0.5 | 3.7 |
| 5* | 95 | 4.7 | 27 | 1.3 | 28.4 |
| 6* | 17 | 0.8 | 8 | 0.4 | 47.05 |
| Rejected at the warehouse* | 79 | 3.9 | 21 | 1.0 | 26.58 |
| Total | 2014 | 100 | 81 | | |

Note: *Rejected skin classes.²⁴

lesions of pox are the second largest skin and hide problem followed by skin atrophy.¹⁹ However, in our study, the proportion of SGP was 4.02%. Pox accounted for 1.5%, 15.5%, 6.8% and 8.3% of pickled sheep skins, wet blue or salted goat skins, wet blue or dry goat skins and wet blue hides, respectively, being rejected.²¹

Sheep and goat pox (SGP) is a highly transmissible viral disease that results in an extensive loss in the production and productivity of small ruminants in Ethiopia. Regarding the status of the disease in Ethiopia, SGP was endemic in almost all the regions of Ethiopia.^{34,35} In Ethiopia, a total of 57,638 small ruminants contracted the disease and more than 4.8 million of them were at risk in areas where outbreaks occurred. Out of the 57,638 sick small ruminants, 6,401 animals died with a case fatality rate of 11.11%. Only about 35–40% the disease was reported in Ethiopia; the actual figures in terms of affected, vaccinated and dead animals are expected to be higher than the reported numbers. Although there were no detailed studies on the prevalence of SGP in Ethiopia, some reports indicate that it is one of the widely distributed and it is the common problem in small ruminant sector of Ethiopia.³⁷

The percentage of SGP was 10.34% and 12.88% in sheep and goats, respectively, in Adama town, Oromia Regional State.³⁴ According to Woldemeskel and Marsha,³⁸ the prevalence of pox was 22% in sheep and 18% in goats in Wollo, Northeast Ethiopia. The seroprevalence between sheep and goat pox was 17% and 15.5%, respectively, in Northwest Amhara Region.² According to Teshome,³⁹ the prevalence was 40% in sheep and 8.12% in goats in Gondar University veterinary clinic. According to Molla et al,⁴⁰ the prevalence was 31.96% in sheep and 35.28% in goats in Gamo Gofa zone of SNNRP. According to Kebede et al, the overall prevalence of small ruminant pox was 11.23%, out of which 12.9% were goats and 9.5% were sheep. The prevalence of sheep and goat pox in the country as well in the study area is pretty high; that is why the overall prevalence of pox lesions in the warehouse. Wounds and scars resulting from pox or tick infestations are the common pre-slaughter defects seen in pickled skins, and wet blue skins next to scratches, cackle, poor substance, and brandings.²¹

Conclusion

In this study, SGP was an important economical disease in the small ruminant sector as well as in the tanning sector. It causes a considerable loss due to decrement in skin grade. The percent loss in warehouses was very high.

Data Sharing Statement

All the data is contained in the manuscript.

Ethics

Our study population is inanimate object (skin already collected from slaughtered sheep and goat for food purpose). The research did not include any live animal or questionnaire survey. Because of this reason, the manuscript is exempt from presenting a review by an institutional review board or ethical committee.

Consent for Publication

All authors read and approved the final manuscript.

Disclosure

The authors report no conflicts of interest for this work and declare that there is no direct or indirect conflict of interest which may call into question the validity of this study.

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