

HISTOPATHOLOGICAL STUDY FOR USING OF POX INACTIVATED VACCINE IN GOATS

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ABSTRACT

For this study used goat pox inactivated vaccine (serial №57,58, year of 2014) produced at Biocombinat State Owned Enterprise (SOE).

On day 21 and at 1, 3, 6, 9, 12 months after vaccination, the vaccinated animals are challenged with a virulent "Ho" strain by subcutaneous inoculation with 0.1 ml ($10^{3.5}$).

We have evaluated that the goat pox inactivated vaccine is effective for up to 6 months.

KEY WORDS: virulent Ho strain, challenge, vaccination, goat pox

INTRODUCTION

Goat pox disease virus is member of the genus *Capripox* subfamily *Chordopoxvirinae*, family *Poxviridae*. Capripox viruses cause severe disease in their host species, characterized by fever and skin lesions[9]. In addition, with severe disease there are characteristic pox lesions in internal organs including the lungs and digestive tract. Disease can result in high morbidity and mortality depending on the virus isolation and host species[10]. This disease is great economic significance to farmers in regions in which they are endemic and are a major constraint to international trade in live stock and their products[8].

There were 52 million livestock, occupying about 41 percent or 22 million goats in our country[5]. Livestock industry is providing job for country side population and their products occupies not a little percent of gross domestic products (GDP) [5].

In Mongolia, the disease appeared in 1967s, then eradicated. However, it started to appear again in 2008-2009 in Dornod aimag. The latest in January to March 2015, sheep and goat pox diseases reported [4].

The purpose of our study was to determine the protective activity of the inactivated goat pox vaccine.

MATERIALS AND METHODS

This study was performed in the State Laboratory of Veterinary Medicine Evaluation and Validation

(SLVMEV) and Laboratories of Pathology and Virology, Institute of Veterinary Medicine. Purpose

of this study was to evaluate vaccine activity and investigate the patho-morphological changes of some parenchymal and immune organs of goats vaccinated with inactivated vaccine, and challenged with the virulent Ho strain of goat pox virus by commonly used methods [7]. In order to recover the goat pox virus, unvaccinated young goats were inoculated subcutaneously, in a shaved area of the abdomen in 20 spots (points) with 5 ml of the diluted virus strain.

RESULTS

Goats were inoculated subcutaneously under the tail with the virulent pox virus “Ho” strain, on day 7-8 after challenge with the virus strain animals were sacrificed for necropsy examination on day 21, at 1,3,6,9,12 months post vaccination.

Clinical signs in experimental goats

Body /rectal/ temperature changes.

The body /rectal/ temperature of experimental goats was measured twice a day in the morning and evening. On day 4 after inoculation with the virus strain “Ho” the body temperature became 41⁰C and later it was decreased gradually and on day 9 the body temperature was 37.1⁰C.

By clinical examination papules and nodules were spotted under the tail on day 9 virus challenge. The vaccine was effective for up to 6 months.

Pathological findings were the same, among macroscopic and microscopic changes of animals challenged with the virus on day 21, and at 1, 3 and 6 months after vaccination.

At 9 and 12 months after vaccination the vaccine protection activity was 50 percent. Therefore we

On day 8 after inoculation with the virus necropsy had performed and histopathology was determined according to methods [3,7]. Samples were fixed in 10% neutral buffered formalin and processed routinely. Sections were stained with Hematoxylin and Eosin (HE), some special stain (Methyl Green-Pyronin) was also used and examined by means of the light microscope [1].

We used 4 vaccinated and 3 unvaccinated /control/ goats for each experiment

have shown results on 21 day, at 6 and 9, 12 months of the experimental infection after vaccination.

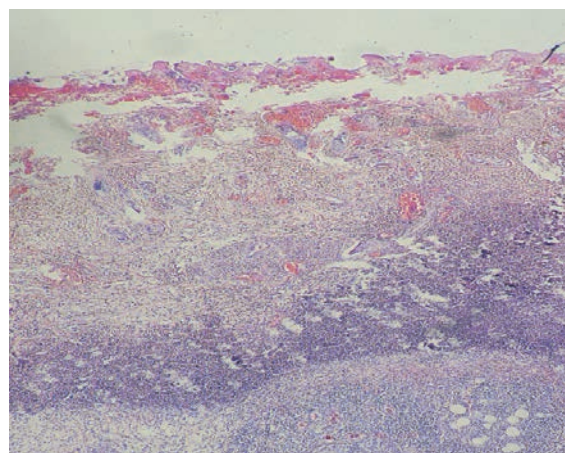
Gross and microscopic changes in experimentally infected animals.

We have used 3 unvaccinated, healthy goats in this purpose. Lesions were observed most tently in the skin, lungs and lymph nodes. Macules and papules were observed at the inoculation site of the skin of experimental goats (Pic.1). Infiltration of inflammatory cells was more severe and epithelial necrosis was present in the epidermis of the skin. Large proportion of epithelial cells were swollen and vacuolated, numerous spindle shaped multinucleated large cells are seen. The dermis of the skin was severely expanded with edema and hemorrhage, neutrophils and lymphocytes. (Pic.2) Enlargement (Pic.3) and inflammation in the lymph node (lymphadenitis) were observed (Pic.4).

In the edge of lungs some hemorrhagic changes can be seen. Microscopically, alveolar septa edema and thickening of alveoli with infiltration by inflammatory cells and interstitial pneumonia observed in the lungs (Pic. 5, 6).



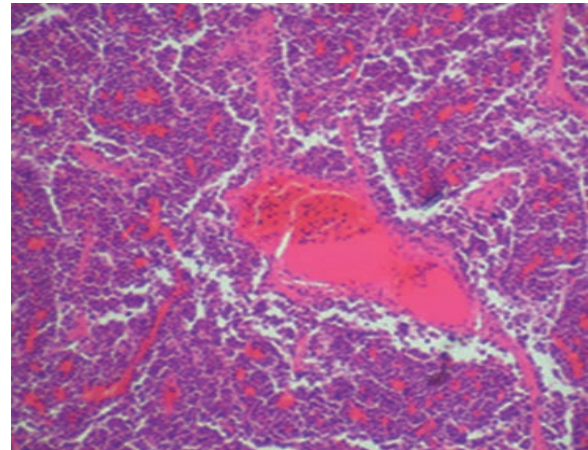
Picture 1. Papules are in the skin under the tail



Picture 2. Inflammatory cell infiltration and necrosis of the epithelial cells of epidermis HE x 100



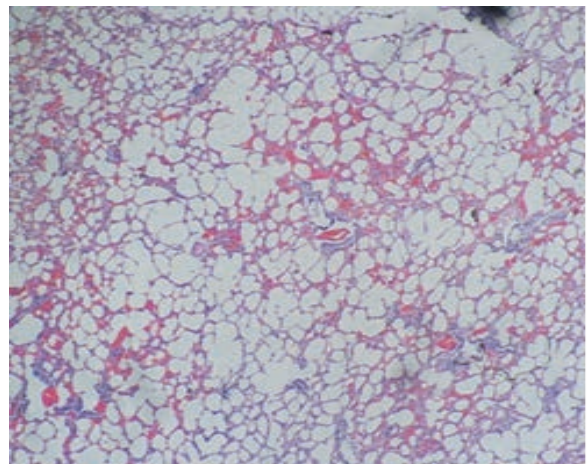
Picture 3. Enlargement of the lymph node (arrow)



Picture 4. Inflammation in the lymph node (lymphadenitis). HE x 100



Picture 5. Hemorrhagic changes are in the lung edge



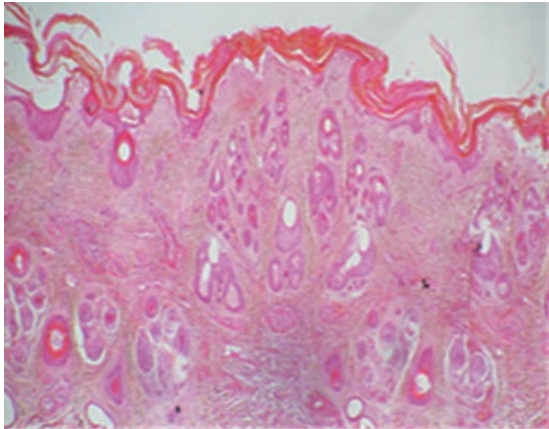
Picture 6. Hemorrhage and thickening of alveoli. HE x100

Gross and microscopic changes on day 21 post vaccination

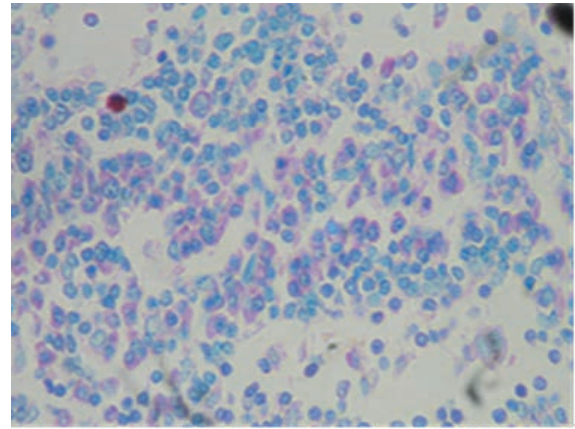
We have sacrificed animals for the necropsy examination on day 7 post challenge. Vaccinated goats had no any macroscopic changes in the skin and internal organs. Microscopically, the skin was normal (Fig. 7), plasma cells and germinal centers in lymph nodes have increased (Fig. 8). The skin and other internal organs were normal whereas unvaccinated control goats were infected with the virus. Unvaccinated goats developed signs of the clinical disease, including fever, loss of appetite, depression and papules were developed under the

tail. On 7 days post infection 3 goats were sacrificed for necropsy and further histopathology examination. Gross pathological changes in animals consisted of typical pox lesions involving the skin part of the tail, mucosal surfaces, lungs and spleen. Lymph nodes were enlarged compared with vaccinated animals.

Microscopically, skin lesions, including focal inflammation in the epidermis, epithelial cells are swollen and necrosis in the epidermis were observed. The dermis of the skin was severely expanded with edema, hemorrhage and cell infiltration.



Picture 7. Histology of the skin of the vaccinated goat, HE x 200



Picture 8. Plasma cells are in the lymph node, Methyl green-Pyronin, x 400

Gross and microscopic changes at 9 and 12 months post vaccination

Goats were inoculated subcutaneously under the tail by virulent “Ho” strain at the 9 and 12 months post vaccination. Vaccinated two goats did not have any characteristic signs of the disease. Whereas remaining 2 vaccinated goats had clinical signs. These infected goats were sacrificed for necropsy. Macule and papules were developed under the tail (Pic.9). Also the epidermis was necrotic, vesicle formation, easy to desquamate and lesions consisted

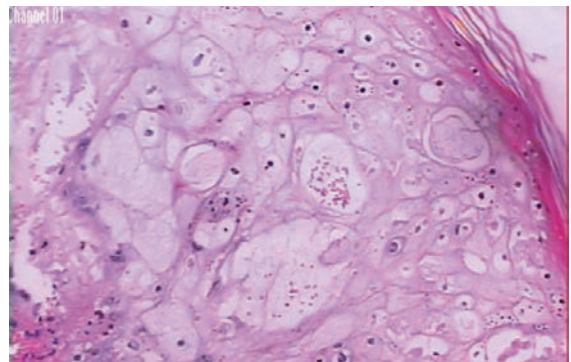
of widespread dermal accumulation of the large area (Pic.10,11).

Microscopically, the affected lung tissue has hemorrhage, thickening of the interlobular septa, interstitial pneumonia (Pic.12). Depletion of the lymphocyte population and absence of germinal centers in lymph nodes are observed.

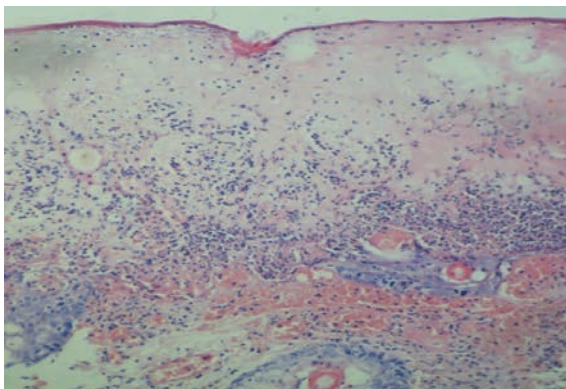
From these results, it is showing that the vaccine activity has decreased at 9-12 months post vaccination.



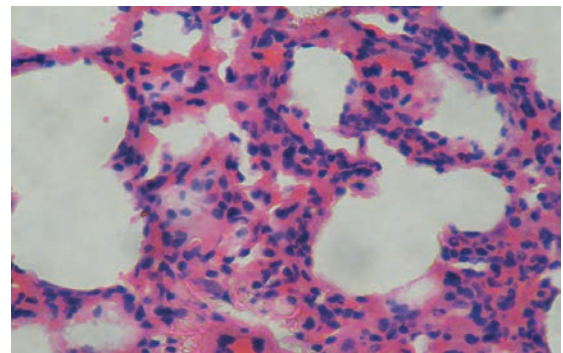
Picture 9. Papules under the tail of the infected goat



Picture 11. Vacuolated degeneration and necrosis in the epidermis of the skin, HE x200



Picture 10. Necrosis and infiltration of inflammatory cells in the skin, HE x 100



Picture 12. Thickening of the wall of alveoli HE x 200

DISCUSSION

The vaccine currently used against goat and sheep pox in endemic most countries are live field isolates, which have been attenuated by multiple passages in cell culture and in chorioallantoic membranes of embryonated hen's egg [8].

In Mongolia, used sheep pox and goat pox vaccines were produced at Biocombinat SOE. The sheep pox vaccine was an attenuated live Piergo strain and in primary

lamb testis cell cultures. The goat pox vaccine was isolated from an infected goat during outbreak of the disease in Hovdaimag, in 1967s. This virus was passaged in naive goats, scabs were collected from resultant lesions and ground up manually, treated

with formalin to kill the virus and finally combined with an adjuvant (aluminium hydroxide) before been injected into animals as a vaccine [4,6].

These live attenuated vaccines work well than the inactivated vaccine. The inactivated vaccine is short duration protection, low level of antibody inducement [8].

The vaccine currently used against goat pox virus considered about 12 months protection by the old standard. However our study result determined the duration of the vaccine protection is 6 months. Therefore, we need to improve the technology of the goat pox vaccine.

CONCLUSION

"Ho" strain, which we used for challenge is still effective enough to show symptoms and morphological changes of pox in goats. By histopathological results, active immunity of

inactivated vaccine against goat pox infection is up to 6 months.

Thereafter vaccine protection was decreased (50 percent) at 9 and 12 months post vaccination.

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