Himachal Journal of Agricultural Research 46 (1): 109-115 (2020)

Capripoxvirus, an Emerging Threat to Sheep and Goat Husbandry in Himachal Pradesh

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Manuscript received:12.04.2020; Accepted: 23.06.2020

Abstract

***Capripoxvirus,* belong to *Poxviridiae* family, cause highly contagious disease of sheep, goat and cattle. Sheep and goat husbandry, mostly migratory type, is practised by many farmers, which contribute significantly to their income. Sheep and goat pox infections are economically important diseases leading to short- and long-term losses to the farmers. A steep rise in sheep and goat pox outbreaks has been recorded in Himachal Pradesh in last few years, therefore, this review summarises recent advances in the epidemiology, pathology and diagnostics of sheeppox and goatpox diseases. This will be helpful in implementing improved disease management strategies in Himachal Pradesh.**

Key words: *Capripoxvirus*, economic burden, goat, sheep.

Healthy livestock aids in improving the economic status and livelihood of poor rural communities and small/marginal farmers (Banerjee *et al.* 2015). Sheep and goats contribute significantly to the economy of the farmers in many mountainous states of India including Himachal Pradesh. Total sheep population in India is 74.26 million and goat population is 148.88 million (http://www.livestockcensus.gov.in/). Himachal Pradesh has 8,04,871 sheep and 11,19,491 goats population (http://hpagrisnet.gov.in/hpagris). Many infectious diseases cause heavy economical losses to the livestock owners. Various infectious diseases have been reported among the sheep and goats in Himachal Pradesh like capripox virus infection, bacterial infections of *Actinomyces pyogenes*, *Staphylococcus* spp., *Streptococcus pyogenes, Micrococcus* spp. etc. and *Chlamydia spp.* (Batta *et al.* 1996; Kumar *et al.* 2000; Chahota *et al.* 2015; Bhardwaj *et al.* 2017). *Capripoxvirus* infections cause significant economic losses and remain a major impediment to the genetic improvement of native sheep and goat germplasm with exotic breeds (OIE,1997). Both sheeppox and goatpox are endemic in Himachal Pradesh and frequent outbreaks have been reported in different parts of the state since 1999 (Batta *et al.* 1999; Verma *et al.* 2011; Sharma *et al.* 2013). Vaccination is the only practical control measure against *Capripoxvirus* infections in the country but it has not been included in the regular vaccination schedule in Himachal Pradesh. In this review, we summarized the recent advances in epidemiology, pathology, diagnostics and control measures of the capripox infection with special reference to the present scenario in Himachal Pradesh.

Capripoxvirus

Capripoxvirus namely sheeppoxvirus (SPPV) and goatpoxvirus (GTPV) cause OIE notifiable endemic pox diseases in India (Bhanuprakash *et al.* 2011). *Capripoxvirus* belongs to one of the genera of *Chordopoxvirinae* subfamily and family *Poxviridae* (OIE, 2010). Sheeppox virus, Goatpox virus and Lumpy skin Disease virus (LSDV) are the three members of capripoxvirus affecting sheep, goat and cattle, respectively (Babuik *et al.* 2009).

Sheeppox and Goatpox virus

Sheeppox and goatpox are the diseases caused by sheeppox virus and goatpox virus, respectively (OIE, 2010). Virions are enveloped, brick shaped with complex symmetry having size of 300 × 270 × 200 nm with double stranded DNA genome. The genomic size is approximately 150 kbps in SPPV and GTPV with terminal repeats at both ends (Tulman *et al.* 2002).

There genomes share 96% nucleotide identity among themselves. The coding region of *Capripoxvirus* contains 156 ORFs, of which central region (024-123) contain conserved genes involved in structure formation and replication while the terminal region both left (001-023) and right (125-156) is highly variable. It has been found that the genes located at the terminal regions (ankyrin repeat protein, kelch like protein, serpins) are mainly playing role in virulence and host interactions (Tulman *et al.* 2002).

Geographical distribution of capripox infection

Sheep and goat pox infection are endemic in Africa, Middle Eastern countries including Egypt, Turkey, Iran, Iraq and Afghanistan and in central Asia and Indian subcontinent (Santhamani *et al*. 2013). The origin of the sheeppox disease was reported to be in central Asia and later on, the disease spread to many western countries. Goatpox was initially reported in Norway (Rafi and Ramyar, 1959). In India, first case of goatpox was reported in 1936, from the Imperial Institute of Veterinary research (IVRI) (Madhavan *et al.* 2016). In India, both the diseases have been reported from almost all states except from the north-eastern regions (Murthy *et al.* 1971; Sharma *et al.* 1986; Mondal *et al.* 2004; Bhanuprakash *et al.* 2005; Govindaranjan *et al.* 2005; Sivaseelan *et al.* 2005; Roy *et al.* 2008). In Himachal Pradesh, outbreaks among migratory and stationary flocks have been reported (Batta *et al.* 1999; Verma *et al.* 2011). The disease has become endemic in Himachal Pradesh and, many outbreaks have been investigated among sheep and goats between 2013 to 2018 in different regions of the state including Chamba, Mandi, Kangra, Hamirpur, Kinnaur, Shimla districts (Chahota *et al.* unpublished).

Host range

Capripoxviruses were considered to be host-specific and SPPV and GTPV infect sheep and goats of various breeds, respectively. However, some viral strains are reported to infect both sheep and goats with varying severity. Davies (1976) characterized a virus causing pox disease in a mixed flock of sheep and goats in Kenya. Under normal conditions GTPV and SPPV are highly host specific (Tulman *et al.* 2002), but the host specificity varies from isolate to isolate because of the gene adaptation to either sheep or goats in a restricted geographical area (Kitching and Taylor, 1985). Kenyan and Yemen isolates infect sheep and goats readily, whereas isolates from the Middle East and India are host specific (Kitching, 1983; Soman *et al.* 1985). Recently, infection of goats with SPPV was reported in India by Bhanuprakash *et al.* (2010). In Himachal Pradesh, the isolated capripox virus strains are host specific (our unpublished data). The incidence of disease is high in exotic breeds of sheep and goat as compared to indigenous species (Mullick, 1988). Spread of capripox infection to new geographical niches may also poses threat to the native wildlife as indicated by a recent report of GTVP infection in Wild Red Serow (*Capricornis rubidus*) (Dutta *et al.* 2019). Though this disease is now considered endemic in Himachal Pradesh, such incidences have not been recorded yet from wildlife. Nandi and Rao (1997) reported infection of *Capripoxvirus* other than its natural hosts in rabbit and reindeer. Few incidences where vaccine strains of SPPV with close similarity to LSDV were used for vaccination, produces mild to severe lesions at the site of inoculation in cattle host (https://www.cabi.org).

Transmission

Contact transmission: Transmission by close contact is most common method but other modes of transmission have also been reported. After infection, virus is secreted in nasal, ocular, oral secretions, urine, milk and due to stable nature of capripox virus, it can persist for extended time on inanimate objects (https://www.cabi.org). Indirect transmission is by the contact with infected objects such as shearing clippers, infected pens, yards used by infected animals (Mirzae *et al.* 2015). The virus persists for up to 6 months in animal pens and for at least 3 months in skin, dry scabs, and hair of infected animals (Kitching and Taylor, 1985a). Once the papule appears, the transmission of virus occurs *i.e.* around six days post-infection and reaches a maximum between 10 to 14 days following infection (Baibuk *et al.* 2009; Bowden *et al.* 2008). There is no reservoir reported for this virus (https://www.aphis.usda.gov). *in-utero* transmission from ewe to lamb and dam to kid has also been reported (Ethiopia, ESGPIP, 2009). Besides this, immune status of the animals has high influence on the disease outcome (Bhanuprakash *et al.* 2004).

Vector borne transmission: The virus survives in the body of *Stomoxy calcitrans* in laboratory setting for up to 4 days and in natural conditions, mechanical transmission of the virus by insects has been seen in Nigerian and Oman isolates of SPPV (Kitching and Meller, 1986).

Role of seasonal migration on disease transmission: In higher altitude of Himalayas, during extreme winters with heavy snowfall and scarcity of fodder, migration of sheep and goat is widely practised where flock moves to plane regions of neighbouring state for grazing pastures. The flock migrates back to high altitude during summer, when lush green pastures re-appears for grazing. This seasonal migration leads to

intermixing of infected flock with uninfected flock. Stress associated with migration predisposes animals to various infections propagating the disease to new niches of Himalayan region (Verma *et al*. 2011)

Clinical signs

Capripox is an acute viral infection with incubation period ranging from 1 to 2 weeks depending upon the strain of the virus and host susceptibility. Mortality and morbidity depend upon factors such as breed, immune status, previous history of exposure, age of animal and strain of virus. In newly introduced exotic breeds, young lambs and kids, mortality may reach up to 100%. In flocks, having reoccurrence of capripox virus infection cases, have morbidity and mortality rate ranging from 1 to 75% and less than 10%, respectively (Bhanuprakash *et al.* 2006). The infection begins with the onset of fever, anorexia, emaciation followed by the development of macules, papules, pustules and scab on the skin covering the full body or restricted only to a hairless part of the body (Fig. 1) (Baibuk *et al.* 2009). The dermis of infected animals become hyper keratinized and histologically develops oedema and have the presence of characteristic intracytoplasmic inclusion bodies (Murray *et al.* 1973). Respiratory, digestive and urinary systems involvement, usually contribute to the clinical outcome of the disease. Systemic signs include conjunctivitis, rhinitis, anorexia, lymph node enlargement and laboured breathing. Death commonly occurs due to secondary bacterial infection. Exotic breeds die before the lesions appear (USDA APHIS). Among native breeds of sheep in Himachal Pradesh, 18.75% mortality was observed (our unpublished data).

Disease diagnosis

The preliminary diagnosis of the capripox infection is done based on clinical signs and lesions observed macroscopically and microscopically. Such lesions can be confused with similar diseases like urticaria, contagious ecthyma, foot and mouth disease and PPR virus (Madhvan *et al.* 2016). Therefore, laboratory diagnosis using isolation, serological and molecular tests becomes necessary to confirm the disease (Madhavan *et al.* 2016). Virion can be detected in skin biopsies, dried skin scabs or nasal swabs in live animals and from cutaneous and visceral lesions in dead animals by electron microscopy without differentiating the *Capripoxvirus* genus. Serological tests using infectious *Capripoxvirus* suspensions and soluble antigen fraction which is non-infectious was commonly used. Various tests like Serum Neutralization Test (SNT), Fluorescent Antibody Test (FAT), Agar Gel Immunodiffusion tests are convenient to perform but are not able to distinguishing three members of *Capripoxvirus* (EFSA, 2015). Isolation of virus using skin scabs, nasal/ocular swabs, and lung tissues, are the suitable material for the identification of virus but is it time consuming. For isolation of virus, both primary and cancerous cell lines are in use. Lamb testicular (LT) cell line, kidney (LK) cell lines form the primary cell lines for the isolation of the virus, however other cell lines like ovine testicular cell, Vero cell are also suitable after adaption of virus to these cell lines for virus propagation (Plowright, 1958; Kalra and Sharma, 1981; Baibuk *et al.* 2007). The virus produces cytopathic effects like rounding, intracytoplasmic inclusion bodies, intense refractivity, chromatin fragmentation, detachment and plaque formation after 7 to 8 days of infection. (Soman and Singh, 1980). Detection of antigen by ELISA, Counter-immuno Electrophoresis Test is also used but all these methods are time-consuming. The severe contagious nature of the disease requires urgent and fast detection of the virus. Thus, molecular detection of virus by conventional PCR and Real time PCR, followed by nucleotide sequencing of conserved genes like *P32* are advantageous and time-saving tests. Multiplex PCR tests have been developed to detect virus based on *P32*, *RPO30*, *GPCR* genes (Santhamani *et al.* 2013; Orlova *et al.* 2006). Multiplex PCR to differentiate *Capripoxvirus* from Orf virus was developed using *I3L* and *E9L* genes of CaPV and Orf virus, respectively (Venkatesan *et al.* 2014). An automated mobile PCR platform for sequential steps of isolation of DNA followed by Real-time PCR has been evaluated for easy access in field condition (Armson *et al.* 2015). Real-time PCR was found to be more sensitive than conventional PCR for rapid detection of virus. The TaqMan based Real-time PCR was developed for early quantification of virus, and differentiation of *Capripoxvirus* and orf virus (Venkatesan *et al.* 2014). Loop-Mediated Isothermal Amplification (LAMP) assay is another method, which is simple and inexpensive and can be performed in rural settings for detection of virus (Notomi *et al.* 2000). LAMP assay with various structural gene like *P32* and non-structural genes like DNA polymerase, were optimized and evaluated (Das *et al.* 2012; Venkatesan *et al.* 2016). Partial or complete genome sequencing using gene like *P32, RPO30, GPCR, A92L* and analysis of *Capripoxviruses*, has been used for comparing the native strains *vis-à-vis* exotic strains reported in different countries for effective surveillance.

Economical impact of *Capripox* infection

Capripoxvirus infection results in the disruption in trade of sheep and goat products like hide, wool, chevon/mutton owing to heavy production loss of

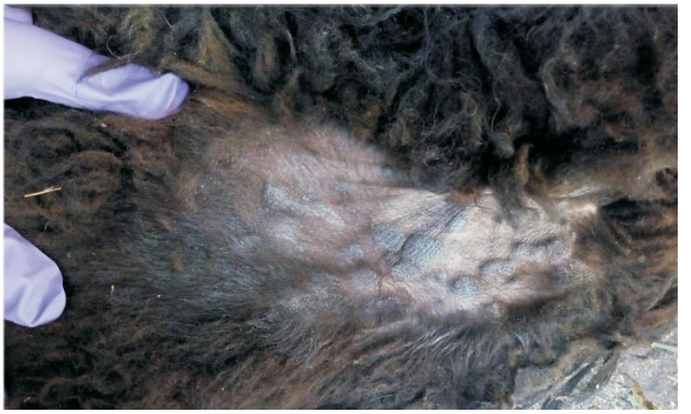

Fig 1. Pock lesions on skin

sheep and goat (Madhvan *et al.* 2016). In Himachal Pradesh, the morbidity rate in an outbreak was 5.18% and mortality was 2.45% (Verma *et al*. 2011). Due to pock lesions on skin of animals, the market value of wool and hide decreases (Green, 1959). Long course of the disease also lowers the quality of the chevon/mutton. The loss due to capripox infection was estimated over INR 105 million in Maharashtra state alone (Garner *et al*. 2000). The average mortality and morbidity rate were 49.5% and 63.5%, respectively with recovery rate of almost 6 years limiting the trade with other countries (Garner *et al.* 2000). Earlier there was no report of Lumpy skin disease in India but recently in August 2019, cases were reported from the state of Odisha with a morbidity rate of 7.1% (Sudhakar *et al*. 2020).

Control and prevention

Capripox disease is highly contagious in nature, where introduction of infected animals to new niches transmit the infection quickly. Those countries which are now disease free, maintains the status by strictly restricting the movement of diseased animals. To eliminate/control the disease, from enzootic areas, strict measures including ring vaccination (Carn, 1993) were followed. In countries like India, where slaughter policies can't be enforced because of socio-economic constrains and also it is difficult to restrict/monitor animal movement, vaccination of sheep and goat become the appropriate and efficient method to manage the disease. Presently, live attenuated SPPV-RF vaccine strain, attenuated by serial passages in primary lamb testicular cell, is used against sheeppox (Bhanuprakash *et al.* 2006).

Cross protection among sheep and goat due to sheep and goatpox vaccine or *vice-versa* is partial (Bhanuprakash *et al.* 2012). Another vaccine strain was used in Tamil Nadu as indigenous strain, passaged in ovine thyroid cells and lamb testicular cell as SPPV-Ranipet (Bhanuprakash *et al.* 2004). A new safe and potent candidate vaccine than other vaccine has also been developed using indigenous strain, Srinagar strain (Srinagar, 38/00) of SPPV (Yogishardaya *et al.* 2011). Among all these vaccine strains, RF strain is generally considered to be the safest of all, in terms of its longest usage track record (Bhanuprakash *et al.* 2003). An attenuated live, vero cell based, goatpox vaccine has also been developed by the Indian Veterinary Research Institute (IVRI) that provides protective immunity for 52 months (Bhanuprakash *et al.* 2012). Combined vaccine is available to combat capripox and PPR virus disease. SPPV-RF and PPRV- Sungri/96, GTPV-Uttarkashi and PPRV-Sungri/96 combined vaccines are available in India (Bhanuprakash *et al.* 2012). Recombinant vaccines are also on the way to combat the disease, using H or F- genes affective against both capripox and PPR (Berhe *et al.* 2003; Caufor *et al.* 2014). Currently no vaccine with DIVA competence is available all over the world (Tuppurainen *et al*. 2017). It is considered that infection or vaccination provides long term immunity to the animals.

Conclusion

Capripoxvirus, affects the livelihood of small and marginal farmers as sheep and goat are mainly reared by these groups. The devastating economic losses due to disease affects overall production and trade of small ruminants and their products. Himachal Pradesh has large number of sheep and goat population. The *Capripoxvirus* infection has been emerging in the state with increasing case fatality rate affecting the economic growth of the farmers. To overcome these losses by the disease, rapid control and preventive measures need to be adopted. Field level quick

diagnostic tests and regular vaccination of the flocks will help in controlling the disease. Studies need to be conducted to estimate actual prevalence associated with the disease for implementing control measures. Himachal Pradesh has rich diversity of wild life ruminant fauna. Surveillance to wild life fauna is also required to determine the extent of endemicity of the disease in Himachal Pradesh.

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