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Review Article

NASAL SCHISTOSOMOSIS IN INDIAN LIVESTOCK – RETROSPECTIVE FOR FIVE DECADES

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ABSTRACT
Nasal schistosomosis is a snail-borne disease caused by the blood fluke, *Schistosoma nasale* which is widespread among bovine populations of many Indian states. Disease in cattle is characterized by the development of granuloma which blocks the nasal meatus resulting in severe respiratory distress. Though buffaloes are equally susceptible to infection, they do not exhibit the symptoms and lesions of the disease and they are often considered as carriers or reservoir hosts. Sheep and goats are uncommon hosts. Continued studies on morphology of the parasite, its prevalence, diagnosis and chemotherapy during the past five decades widened our knowledge on these fronts. Studies on experimental infections on intermediate host, and in domestic and laboratory animals contributed more towards understanding of life-cycle of the parasite and its biology, as well as pathogenic effects on the affected organs. Inter-transmissibility experiments of *S. nasale* between cattle and buffaloes and *vice versa* employing cercariae of cattle- and buffalo-origin were proved that the same species of parasite occurring in them. New egg counting techniques and different immuno-molecular methods have aided the assessment of infection. Oxyclozanide has been recognized as cost-effective, safe and orally administrable drug for the effective treatment of this disease.

Keywords: *Schistosoma nasale, Hosts, Prevalence, Pathogenesis, Molecular diagnosis, Chemotherapy*

INTRODUCTION
Nasal schistosomosis (NS) or snoring disease is a snail-borne trematode infection caused by blood fluke, *Schistosoma nasale* Rao, 1933 characterized by rhinitis, granuloma in nostrils, mostly confined to cattle causing breathing distress. The resultant morbidity, impaired working ability, reduced milk yield and stunted growth incurred heavy financial losses to farmers. Though buffaloes are equally susceptible to infection, the symptoms and lesions of the disease as observed in cattle are not usually expressed in them. Sheep and goats are rarely found infected. Excluding a few north-western areas, the disease in bovines is widespread in almost all parts of India. Updates on nasal schistosomosis were provided by Agrawal and Alwar (1992) and Agrawal (2012). Muraleedharan *et al.* (2014) catalogued the intermediate and final hosts of schistosomes and animal parasites reported from India. In the present review, contributions of scientists on this disease for the last five decades (1967-2018) have been further highlighted.

Morphology
Dutt (1967) conducted a detailed study on the morphology of adults *S. nasale* of cattle and buffaloes and no differences in morphology was found. The studies of Rajamohanan (1972), Biswas (1975) and Anandan (1985) had confirmed the description of Dutt (1967). However, Rajamohanan (1972) noticed that *S. nasale* of buffalo was slightly larger; male cuticle has tubercles without spines and constant number of 3-4 testes unlike those of cattle which varied from 2 to 6. Scanning electron microscopy of *S. nasale* of cattle revealed that the tegument is having dome-shaped tubercles with very fine spines and the presence of transverse grooves. Gynaecophoric canal is smooth having perforations and papillae (Gupta and Agrawal, 2002).

The shape egg of *S. nasale* is of modified spindle or boomerang, having a central concavo-convex body with attenuated ends, one end bearing a spine. Some variation in shape noticed in cattle mostly in terms of the depth of the concavity or difference of the spinal end, was well-illustrated whereas the eggs of parasites of buffaloes appeared uniform in shape (Dutt, 1967; Rajamohanan, 1972; Biswas, 1975; Muraleedharan *et al*., 1977a).

Larval forms and development in intermediate host

The egg hatches to miracidium which is pyriform having anterior conical projection. Its body is covered with 22 ciliated plates arranged in four tiers of 6, 9, 4 and 3 cells in order and the third set of bristles of sensory papillae contains 23-25 bristles (Dutt, 1967). The miracidia swim in water and their survival time was about 10hr in summer and more than 15hr in winter. Within that time they were attracted towards the snail intermediate host (IH), *Indoplanorbis exustus*. They penetrated into the snail's head region and foot-pad after shedding their ciliary coat within 15-20 min (Rajamohanan, 1972). The seventh day mother sporocyst was oval in shape, later became almond-shaped and matured by 15th day in the hepato-pancreas of the snails assuming an irregular-shape. It contained about a dozen daughter sporocysts in different stages of development which are cylindrical with rounded ends. Daughter sporocysts that emerged from the mother sporocysts were elongated or curved in shape. Their germ cells differentiated into numerous cercariae which released into water where they distributed with their head downwards and tail upwards and moved often by lashing of tail (Dutt, 1967). The cercariae produced are furcocercus, brevifurcate, non-ocellate and spinosed. Their excretory system has four pairs of flame cells - two anterior and one posterior pairs in the body and one pair in proximal part of tail stem. The terminal tip of furcal rami is tubular and protruded which is characteristic of *S. nasale* cercariae (Dutt, 1967).

Muraleedharan *et al*.(1975a) and Koshy *et al*. (1975) exposed miracidia of *S. nasale* of cattle and buffalo origin, and additionally of goat origin by Anandan and Raja (1988) and found that the developmental period for cercariae in snails varied from 23-84 days post-infection (DPI). Due to better infectivity of miracidia of buffalo origin to snails followed by release of more cercariae, buffaloes appeared to have major role in transmission of NS (Anandan and Raja, 1988). Released cercariae, being short-lived, swam swiftly in search of final host (FH). Muraleedharan *et al*. (1975b) noted that cercarial-survival period was up to 55hr at 28^{0}C and 124hr at 10^{0}C. Snails preferred certain months for cercarial shedding as observed from experimental and field studies. Rajamohanan (1972) experienced that *Indoplanorbis* species prevalent in Kerala was refractory to *S. nasale* infection, but those collected from Tamil Nadu harboured the infection. Older snails were not found infected. Dutt and Srivastava (1968) and Biswas (1975) recorded about 7% and 2% *I. exustus* harboured natural infection in Sitapur and Bareilly (Uttar Pradesh), respectively. Muraleedharan *et al*. (1976d) recorded higher cercarial infection from June to October in Karnataka whereas Thakre and Bhilegaonkar (1998) and Kolte *et al*. (2012) observed that infection was more common in September to January in Maharashtra. Most of the findings indicated that *I. exustus* was the main IH of *S. nasale*, but infection was also seen in *Lymnaea* species in Maharashtra and Tamil Nadu by Bhilgoankar *et al*. (1978) and Sivaseelan *et al*. (2004) respectively.

Development in final host

Upon contacting the final hosts (FH), the cercariae detach tail and enter the body of FH by skin penetration. They reach the blood circulation, develop to schistosomulae and migrate via heart, liver, lungs and finally to nasal veins where they attain maturity. The females start to lay eggs whose spiny ends assist them to bore their way through capillaries to reach nasal mucosa, cause irritation and inflammation, producing excessive nasal discharge which contains the fully-formed eggs. The prepatent period (PPP) in FH had shown wide variation from 92 to 254 DPI. So the total time for the completion of life-cycle partially in IH and FH even extended to 115-338 DPI (4-11months). Cross-transmission experiments conducted employing *S. nasale* cercariae of buffalo and cattle origin and *vice versa* cleared the suspicion of existence of species difference of parasites found in cattle and buffaloes (Dutt and Srivastava, 1968;

Gowrishankar, 1968; Rajamohanan, 1972; Biswas, 1975; Koshy *et al.,* 1975; Muraleedharan *et al*., 1976e; Anandan, 1985).

Pathology in experimental animals

Dutt and Srivastava (1968) showed that one bull calf infected with cercariae of buffalo origin and other three calves with large number of cercariae of cattle origin produced numerous nodules, ulcers and muco-purulent nasal discharge followed by respiratory distress for observation periods that varied from two to six years, One of the bull calves autopsied, yielded large number of male and female worms from nasal blood vessels and a few male worms from lungs. Buffalo calves infected with cercariae of cattle origin was maintained for two years without showing any lesions. The parasites of buffalo origin were found to produce appreciable amount of lesion in cattle while those of cattle origin produce only sub-clinical infection in buffaloes. The result of worm-recovery on autopsy was similar to that observed in bull calf.

Biswas (1975) could infect only male calves of cattle and buffaloes while females acted refractory. Infections were established in them in two phases - primary and secondary. In primary exposure, the parasites developed in longer PPP of 131-254 DPI, and discharged viable eggs with gradual development of lesions from dot-like eruptions to cauliflower-like growths and increasing severity of symptoms with thick nasal discharge which later become thinner showing no viable eggs. When the worms and cauliflower-like growths developed in primary infection disappeared, the secondary exposure to cercariae was done. In secondary phase, infection was established in comparatively shorter PPP of 50-99 days and infected animals continued to discharge eggs for 80 to 100 days in 75% cases and up to 1000 days in the rest. On autopsy, worms were recovered from the nasal veins and lungs.

Rajamohanan (1972) and Biswas (1975) did not observe clinical symptoms or lesions in experimental buffalo calves. Biswas (1975) found that the fewer worms developed in them produced higher number of eggs. The infection developed within 168-190 days, and longer patency period was also noticed. Muraleedharan *et al.* (1976e) infected two buffalo calves with cercariae of cattle origin, one of which succumbed to infection on 17 DPI after brief illness and post-mortem revealed pneumonic changes and was recovered immature worms from lungs. The second calf, positive on 94 DPI showed mucus discharge and focal hyperaemia. On autopsy after 270 DPI, the nasal mucosa of the calf showed numerous raised nodules in the anterior two-thirds of nasal passage, and many male and female worms were recovered from nasal veins. Histopathology (HP) revealed hyperplastic changes of the mucosa and circumscribed areas of granulomatous reaction in the submucosal connective tissue and partial thrombosis in blood vessels which contained worms. Anandan *et al*. (1995) reported that the nasal mucosa of infected buffalo calves showed haemorrhage, fibrovascular proliferation and pronounced hyperplasia of lining epithelium, while cattle exhibited hyperplasia of squamous cell lining and many eggs with radiating eosinophilic sleeve giving actinobody-like appearance.

Experimental infection in sheep and goats

Dutt and Srivastava (1968) and Biswas (1975) reported that nasal discharge of goats became positive between 138-178 DPI, but they did not show symptoms and lesions and both male and female worms were recovered on autopsy. Sahay and Sahai (1976) noticed proliferation of connective tissues and necrosis of cells of bile ducts of infected kids and lambs. Anandan (1985) and Anandan *et al*. (1995) reported that the exposed lambs and goats to comparatively higher number of cercariae showed congestion of nasal mucosa and small eruptions, and on autopsy, male and female worms were recovered on 126 and 120 DPI from lambs and goat respectively. The infected lambs revealed thrombosed blood vessels, and eggs with miracidia surrounded by haemorrhage in the submucosa whereas infected goats showed clusters of eggs, pseudotubercles and cellular infiltration without actinobody formation. Agrawal (1996) reported that an infected lamb died on 105 DPI and no *S. nasale* was recovered by routine method, but yielded male worms from liver after perfusion. Thakre (1996) noticed congestion and emphysema in lungs and fatty changes in the liver of infected goats.

Experimental infection in laboratory animals

Dutt and Srivastava (1968) infected three albino mice with cercariae of *S. nasale* and recovered immature worms from their lungs, and from the heart of one mouse. Biswas (1975) and Biswas and Subramanian (1990) exposed rabbits, guinea pigs, rats and albino mice of both sexes with cercariae obtained from laboratory-bred *I. exustus*. None of them got infected except male mice in which very few schistosomulae developed and migrated through circulation towards lungs where the further development of immature worms was arrested. Comparatively fewer immature parasites were recovered on 100 DPI; males were differentiated with indistinct testes, but no ovaries were recognized in females. The lungs of infected male mice showed signs of congestion, lymphoid hyperplasia, arteriolar and perivascular hypertrophy. Anandan (1985) and Sahay and Sahai (1976) infected rabbits, guinea pigs and mice and recovered *S. nasale* from liver, lungs and heart of some of these animals. Sahay and Sahai (1976) revealed degenerative changes and coagulative necrosis of the liver. Hepatic cells of mice showed increased deposition of haemosidrin pigments. Lungs showed focal areas of congestion and consolidation. Sahay *et al.* (1977) revealed marked depletion of PAS-substances from hepatic cells around the affected bile ducts and depletion of protein. Von Kupffer's cells were positive for calcium deposits. Agrawal (1996) infected rabbits and recovered male and female worms with typical eggs, inhabited in hepato-portal system, but no worms were recovered from nasal veins. Sapate *et al.* (2001) found immature worms developed in liver and lungs of golden hamsters and mastomyes on 75 DPI, with necrotic foci in liver, extremely dilated hepatic veins and congestion in lungs.

Prevalence of natural infection in bovines

The state-wise prevalence of *S. nasale* infection in cattle and buffaloes reported for the period 1968-2018 has been furnished in Table 1. Differences noted in the prevalence depended on many epizootiological factors as envisaged by Anantaraman (1981). The wide range might be one of reasons for variations observed prevalence rate of parasites in particular month or season of the year and also migrating animals from one area to another, sales or slaughter. The observation of Dutt and Srivastava (1968) that all the cattle and buffaloes of a farm at Sitapur (Uttar Pradesh) were positive for *S. nasale* indicated both these species were equally susceptible to infection. However, Bhatia and Rai (1983) recorded higher infection in cattle than in buffaloes of Uttar Pradesh. An outbreak form of NS among cattle and buffaloes in Tamil Nadu was reported by Sundar *et al.* (2004). A four-month old bull-calf found positive for ova of *S. nasale* (Sreeramulu, 1994) which appeared to be the minimum age recorded in household surveys whereas previous report of minimal age was 9 month (Muraleedharan *et al*. 1976c). Higher incidence of NS was recorded during rainy seasons by majority of investigators. On the contrary, greater prevalence of infection was recorded during summer followed by winter and rainy season by Jayalakshmi *et al.* (2016). Sreeramulu (1994) reported that Hallikar bullocks were more prone to infection than the non-descript bullocks while Muraleedharan *et al*. (1976c) reported that the similar difference noted in their studies was not significant. Ottalwar *et al* (2004), Jayalakshmi *et al.* (2016) and Yogeshpriya *et al*. (2018) noticed higher prevalence of infection in crossbred than indigenous cattle. In a locality near Jabalpur (Madhya Pradesh), the local cattle were found negative for NS while infection was detected in cross-bred cattle and buffaloes (Banerjee and Agrawal, 1991). Ottalwar *et al*. (2004) and Bulbul *et al*. (2017) observed higher prevalence of infection in male than in female cattle. Jayalakshmi *et al.* (2016) and Yogeshpriya *et al*. (2018) reported that the percentage of infection exceeded in females. Biswas and Subramanian (1978) found this infection only in a few buffaloes of Bareilly district, but not in cattle. A higher trend of infection in buffaloes compared to cattle, was exhibited in certain states like Kerala (Rajamohanan, 1972; Ravindran and Kumar, 2012), Assam (Rajkhowa *et al*., 1992), Tamil Nadu (Sivaseelan *et al*., 2004) and Andhra Pradesh (Jayalakshmi *et al.*, 2016). Higher percentage of infection was recorded in graded Murrah than non-descript buffaloes (Jayalakshmi *et al.*, 2016).

Sheep and goats

Sen and Ray (1969) detected *S. nasale* infection in three goats in West Bengal. Achuthan and Alwar (1973) reported 1.8% infection in sheep and 41.4% in goats in Tamil Nadu while Muraleedharan *et al*.

(1973) reported 2.3-3.2% infection in sheep in Karnataka. Rajkhowa *et al*. (1992) recorded 1.8% goats having infection in Assam.

Table 1: State-wise prevalence of *Schistosoma nasale* infection in bovines

State/region	% of prevalence/ No. of cases		Reference
	Cattle	Buffaloes	
Assam (including Guwahati, Kamrup)	9.92-45.00%	27.50%	Rajkhowa *et al*. (1992); Agrawal *et al*. (1998); Deka (1998); Phookan (2012); Bulbul *et al*. (2017)
Andhra Pradesh (different parts)	8.82-80.00%	22.04-60.0%	Christopher and Rao (1975); Sreeramalu (1994); Rao and Hafeez (2005); Jayalakshmi *et al.* (2016); Didugu and Reddy (2017)
Bihar	18.75%	-	Kishor (2008)
Chhattisgarh (Durg)	36.77%	8.33%	Ottalwar *et al*. (2004)
Haryana (Panchkula),1st time	CB cow	-	Sangwan *et al*. (2015)
Karnataka (Bangalore, Mandya and Mysore)	29.90-65.6%	2.60-35.70%	Muraleedharan *et al*. (1976c); Sumanth *et al*. (2004a)
Kerala (northern and middle)	11.10-25.0%	23.4-46.83%	Rajamohanan (1972); Ravindran and Kumar (2012); Nimisha *et al*. (2017)
Madhya Pradesh	13.30%	48.15%	Banerjee and Agrawal (1991)
Maharashtra (including Gadchiroli, Gondia, Chandrapur and Bhandara)	17.14-42.50%	-	Bhilegaonkar *et al.* (1977); Thakre and Bhilegaonkar (1998); Ravindra (2007); Kolte *et al*. (2012); Mukund (2012)
Orissa (Cuttack and Balasore)	72.40%	59.16%	Sahoo (1994); Mahapatra (1997)
Puducherry	Bullocks	-	Latchumikanthan *et al*. (2014)
Rajasthan (Bikaner)	CB cow	-	Qadri and Ganguli (2016)
Tamil Nadu (Madurai, North Arcot, Salem, Tanjavur and Tirunelveli)	15.50-95.65%	24.4-100%	Achuthan and Alwar (1973); Sivaseelan *et al*. (2004); Sundar (2004); Yogeshpriya *et al*. (2018)
Uttar Pradesh			
Sitapur farm	100%	100%	Dutt and Srivastava (1968)
Bareilly	-	3.46-8.79%	Biswas &Subramanian (1978)
Different districts	575 cases	24 cases	Bhatia and Rai (1983)
West Bengal (24 Parganas, Midnapur and Burdwan)	0.82-2.89%	-	Biswas (1975)

Symptoms

Dutt and Srivastava (1968) reported 100% infection in a herd consisting of cattle and buffaloes of which 70% of cattle exhibited clinical symptoms, but none of the buffaloes did. Rao and Devi (1971) observed that positive cattle were excreting 2-3 times more ova than buffaoles. Contrary to this observation, Gowrishankar (1968), Rajamohanan (1972) and Muraleedharan *et al.* (1976b) confirmed that nasal discharge of buffaloes contained more number of fully formed eggs and miracidia. Muraleedharan *et al*. (1976b) observed that the progress of disease could be related to nature of nasal discharge and egg output. Serous discharge appeared in the early stage and mucous/mucopurulent discharge in the advanced stage of the disease. According to the development of lesions, reduction in egg scores was found in most of the cases whereas only very few of them showed higher egg score. Positive cattle showed typical clinical symptoms with seromucoid nasal discharge (Ottalwar *et al*., 2004) while Sivaseelan *et al*. (2004) noted blood-tinged mucopurulent nasal discharge. Buffaloes produced larger quantity of slightly opalescent or clear serous nasal discharge (Gowrishankar, 1968; Rajamohanan, 1972; Muraleedharan *et al*., 1976b; Ottalwar *et al*., 2004), Many workers including Rajkhowa *et al*. (1992) and Sivaseelan *et al*. (2004) reported infection in buffaloes was often unnoticed being subclinical and symptomless. Mouli and Christopher (1993) observed some unusual clinical manifestations in buffaloes like paresis, lameness, oedema of limbs and lower jaw, recurrent tympany, purulent bilateral conjunctivitis and respiratory distress associated with this infection. NS could be suspected in buffaloes showing signs of reduced water intake and sudden drop in milk yield (Didugu and Reddy, 2017). A she-buffalo brought for treatment of severe epistaxis, was found to have typical nasal lesions of NS on endoscopic examination, confirmed by characteristic eggs in nasal washings by microscopy (Satbige *et al.*, 2018).

Lesions in cattle

Analysis of lesions of 174 NS-positive cattle of two enzootic areas of Karnataka revealed that 47.7% had nodular type, 36.8% cauliflower-like growths, 10.9% nasal hyperaemia and 4.6% without visible lesions (Muraleedharan *et al*., 1976b). According to Sumanth e*t al*. (2004a), among 300 male cattle slaughtered at the abattoir of Bangalore, 65.6% was found positive by microscopical examination of nasal scrapings for eggs while *S. nasale* worm was recovered from 72.6% of them on examination of nasal cuttings. The infection was found mild in 59.2% cattle, moderate in 35.3% and heavy in 5.5%. Biswas (1975) noticed that by the advancing stage of disease, gradual reduction was noticed in the number of eggs discharged which might be due to trapping of eggs in the tissues avoiding their free release. Unilateral or bilateral miliary/cauliflower-like nasal granuloma in cattle was observed by Ottalwar *et al*. (2004), Kishor (2008) and Qadri and Ganguly (2016). An outbreak of NS in cattle with prominent growths was reported by Sundar *et al.* (2004) and HP of nasal scrapings showed the presence of numerous inflammatory cells, squamous cell metaplasia and actinobody formation around ova. Mukund (2012) observed that the nasal mucosal layers showed severe granulomatous reaction consisted of cellular infiltration, congestion of veins and presence of worms with inflammation in the surrounding area. The rhinoscopic diagnostic procedure displayed the extent of closely packed lesions obstructing nostrils and the rhinoscopy-guided mucosal biopsy enabled routine HP (Swamy *et al.,* 2016).

Lesions in buffaloes

The type of lesions reported in buffaloes varied from pin-head or pea-sized nodulations, miliary eruptions, minute ulcers or patchy congestion of mucosa (Rao and Devi, 1971; Rajamohanan and Peter, 1975; Muraleedharan *et al*., 1976b). HP studies of Gowrishankar (1968) noted actinobody-like coating around ova and thickening of blood vessels. More number of eosinophils were noted by Gowrishankar (1968) and Rao and Devi (1971). Rajamohanan and Peter (1975) and Muraleedharan *et al*. (1976e) reported the absence of actinobody formation, but marked pronounced cellular reaction consisted of mononuclear cells, polymorphs and eosinophils. Rajkhowa *et al*. (1992) did not find lesions in positive buffaloes. Mouli and Christopher (1993) reported growths over mucosa with respiratory distress in 3.8% buffaloes while 58.6% had only pin-head sized elevations. Non-development of lesions in buffaloes was not due to any histological difference of the nasal mucus membrane, but assigned to better host-parasite relationship

(Rajamohanan, 1972; Biswas, 1975). In cattle the development of lesions it might be sequel to non-specific cellular response on the part of the host (Biswas, 1975). Smaller nodulations noticed in few buffaloes might be attributable to break down of body resistance (Muraleedharan *et al.* (1976b). Anantaraman (1981) doubted that buffaloes possess an intrinsic immunological factor capable of producing minimal lesions without impairing the maturation and reproductive potential of *S.nasale*.

Haematological studies

Haematological studies of infected buffaloes recorded higher ESR and the electrophorectic analysis of the serum protein revealed a rise in gamma globulin fraction (Rajamohanan, 1972). Studies of Koshy and Alwar (1974) indicated that bovines manifested hyper-gammaglobilinaemia and hypo-albuminaemia. Cattle showed a reduction in the alpha globulin fraction while buffaloes showed an increase. Sundar *et al* (2004) reported haematological values of infected cattle: Hb 7.05g%, PCV 34.5%, RBC 5.5/million, WLC 3,748cmm, total protein 7.11g% and albumin 3.43g%.

Diagnosis

NS in cattle was routinely confirmed by microscopical examination of nasal discharge/scrapings. For evaluating the egg production potentials of worms, counting of eggs present in 1 cc of measured sample of nasal mucus in EPCC method of Rao and Devi (1971) or in 1 gm of weighed sample of nasal scrapings in EPG method of Muraleedharan *et al* (1976a) were adopted following the necessary procedures. The superiority of EPG technique over EPCC method was confirmed (Muraleedharan *et al*., 1976a; Sumanth *et al*., 2004a). Clinical cases in cattle showed higher EPG than those with symptomless infection (Banerjee and Agrawal, 1991). To detect sub-clinical carrier cases or early developing stage of the infection, certain immunological tests such as miracidial immobilization test, gel diffusion technique and passive agglutination test were tried by Biswas (1975), but results were inconclusive, though some antibody response was demonstrable. Other tests used were cercarian Hullen reaction (CHR), intradermal test (IDT), dot-ELISA, counter current immunoelectrophoresis (CIEP) and polymerase chain reaction (PCR). In CHR, the cessation of movements of cercariae by the formation of an envelope around them, 15min after mixing cercarial suspension to inactivated serum of *S. nasale*-infected animals was demonstrated (Rajamohanan and Peter, 1972). IDT using *S. nasale* homogenates and metabolites gave inconclusive results (Anandan, 1985). But Jagannath *et al.* (1988) and Sano *et al*. (1988) conducted IDT in cattle by intradermal injection of *S. japonicum*-whole worm antigen and could detect *S. nasale* infection. Sumanth *et al*. (2003) evaluated simple, rapid, sensitive dot-ELISA for serodiagnosis of NS in cattle by *S. nasale* whole-worm-antigen and the test detected 98.16% of known sera samples of cattle. CIEP was also evaluated using whole-worm antigens of *S. nasale* and the technique was sensitive and specific for the diagnosis of early and well-established stages of infections (Sumanth *et al.*, 2004b).

The PCR with 328 and 1088bp products of *S. nasale* amplified with mitochondrial genes of 125r RNA and cytochrome oxidase sub-unit I gene of isolates from Bangalore was performed (Murthy, 2012). The specificity was checked by NCBI nucleotide sequence library, and phylogenetic analysis of 125 ribosomal RNA gene nucletide sequence was done. These studies indicated that Bangalore isolates of *S. nasale* was distinct from Sri Lankan and Bangladesh isolates. NS in the subclinical stage could be diagnosed by PCR technique which offered greater sensitivity and specificity (Mukund, 2012). The designed primer pair was highly specific in amplifying the Cox-1 gene of *S. nasale*. The study indicated that the Indian *S. nasale* isolate matched 99.6% with *S. nasale* of Sri Lankan origin and formed a single clade with the other South Asian *Schistosoma* spp. Bulbul (2016) reported from Assam that phylogenetic tree analysis of both the cercaria and the adult worm of *S. nasale* based on COI gene sequence and 28S gene sequences that they were closely related to Nepal and Bangladesh forms.

Treatment

Chemotherapeutic trials with various drugs conducted in cattle have been summarized in Table 2. Among the drugs, oxyclozanide appeared to be more effective and cheaper having the advantage its oral administration. Relapses of infection had often happened mostly due to the repeated exposure of treated animals to snail-infested areas. A she-buffalo brought for treatment of severe epistaxis, was found

positive for ova of *S. nasale* and treated with schedule dose of praziquantel successfully (Satbige *et al.*, 2018). The drug was also found effective against *S. nasale* in experimentally infected sheep and goats at 60mg/kg bw (Anandan and Raja, 1987).

Table: 2 Chemotherapy against nasal schistosomosis in cattle

Drug / Dosage / Route	Duration of treatment	Efficacy	References
Anthiomaline (LAT), 10-20ml, I/M	3-7days	Less effective	Bhatia and Rai (1976)
	Four successive days	75% cured	Anandan and Lalitha (1979)
	Thrice at weekly intervals	40% cured, 60% reduced lesions	Muraleedharan and Rajasekhar (1996)
		Temporary effect	Agrawal *et al.*(1998)
		Cured	Sivaseelan *et al*. (2004); Qadri and Ganguly (2016); Yogeshpriya e*t al*. (2017)
	Three days	Effective	Ravindra (2007)
LAT+ Praziquantel 10mg /kg bw orally	Three day Once	More effective	Ravindra (2007)
LAT + Praziquantel 20mg/kg bw orally	Once for 6 weeks Single dose	Cured	Swamy *et al*. (2016)
Sodium antimony tartarate (SAT) @ 1.5 mg/kg bw as, 2% solution in 10% dextrose saline I/V	Schedule 1)Twice daily for 2 days Schedule 2) Once daily for 4 days Stall-fed	Fairly effective	Muraleedharan *et al*. (1977b)
SAT 2-3mg/kg bw I/V	Four days treatment	Cured, 25% relapse	Anandan and Lalitha (1979)
SAT, 2.5% aqueous solution , 20 ml, I/V	Three consecutive days	Good relief	Sreeramulu (1994)
Antimony tartrate mother tincture I/M+ Belladonna 10 M pills	2 daily for 4 days 10 pills/ day for 4 days	Cured	Rao and Sreemannarayana (1980)
Antimosan, maximum of 40ml/adult, I/M	3 injections at 4 days intervals	Effective	Bhatia and Rai (1976)

Neguvon a)6gm as aqueous paste b)as10% ointment	Applied over lesions for 10 days	Temporary recovery; paste acted better	Muraleedharan *et al*. (1977b)
Neguvon, oral a)30mg /kg bw b)40mg/kg bw	a) daily for 8 days b) 4 alternate days	Temporary effect, relapsed after 70 days	
Sodium antimonyl-dimethylcysteino tartar-ate 7.5mg/kg bw, I/M	5 days	71.5% curative	Anandan and Lalitha (1979)
Praziquantel 20mg/kg bw, oral	Single dose	80% cured by 4th week. Curative 100% effective	Rahaman *et al*. (1988) Sano *et al*. (1988) Dambarudhara *et al*. (2016)
Praziquantel 10mg/kg bw, oral	Single dose	Effective	Ravindra (2007)
a)Oxyclozanide 10mg/kg bw, oral	Three doses at weekly interval	50% cured, 50% reduced lesions Most effective	Muraleedharan and Rajasekhar (1996) Kinkar and Kanchan (2016); Kumar and Jayaprakash (2016)
b)Rafoxanide 75mg/kg bw, oral	Single dose	50% reduced lesions	Muraleedharan and Rajasekhar (1996)
c)Levamisole /30kg bw, S/C	Thrice at weekly intervals	14% cured 57% reduced lesions	
d)Ivermectin 1ml/50kg bw, I/M	Single injection	20% cured 20% reduced lesions	
Triclabendazole 20 mg/kg bw, orally	Single dose	Curative Snoring reduced	Agrawal (2012) Sangwan *et al.* (2015)

Conclusion

There is need for further epizootiological investigations to understand the reason for the existing variations in the distribution of *S. nasale* parasite in cattle and buffaloes of certain areas and the absence of infection in certain other areas despite the availability of IH. Different control measures advocated for snail eradication have to be sorted out and integrated for their effective implementation. The natural immunological resistance to *S. nasale* infections in buffaloes has to be fully exploited for inducting the similar condition in cattle by nano-biotechnology. The development of a prophylactic vaccine by the help of advanced immunology is highly stressed. Moreover, the methodology for an autogenous vaccine on the lines of papilloma virus can be evolved to alleviate the lesions of already suffering cattle. Cattle is the

only model for animal schistosomosis where development as well as changes in lesions can be visualized directly through their nostrils which is advantageous especially for assessing the progress of drug trials.

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