SPECIAL ARTICLE

Rabies in Wildlife - A Threat!

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Abstract

Dogs are the principal reservoirs of rabies virus in developing countries and are responsible for majority (97%) of human infections. Other reservoirs and important vectors of rabies virus include wild and domestic Canidae, including wolves, foxes, coyotes, jackals, bats, cats, monkeys, skunks, raccoons and mongooses in different geographic locations. India is a habitat for various species of wildlife. In the recent years, episodes of rabies in wildlife in India have been reported and could be a potential source of rabies for adjoining domesticated animals and human population.

The approaches for controlling rabies in wildlife can be elimination of the reservoir species, elimination of rabies in the reservoir species, or protection of victim species from rabies infection via a reservoir (Rupprecht *et al.*, 2001). Another potential control method involves vaccination of wildlife reservoir species. In trap-vaccinate-release (TVR) programs target reservoir species with live-trapping and manually injecting liquid vaccine. For a large scale usage, ORV may be more economically and technically feasible alternative.

Although scanty, the available literature and documented evidences based on laboratory investigations prove the existence of its sylvatic cycle. Undoubtedly, this situation could be resulting in spillover of infection to domesticated animals and human population in the geographical locations in the vicinity of forest especially in the backdrop of deforestation.

Rabies control programs must be well designed to be efficacious, cost effective, be publicly supported. It should have negligible negative impact on wildlife, livestock, humans and landscapes. Environment and Forest department, Animal Husbandry and Animal Welfare Organizations (AWOs) should be the stake holders in planning, implementation and evaluation of the Rabies control program in wildlife.

Introduction

Rabies is primarily a disease of mammals, both terrestrial and airborne. Based on the extent of prevalence and species affected, three principal global areas of rabies have been defined. These areas are (1) countries with enzootic canine rabies (all of Asia, Latin America, and Africa); (2) countries in which canine rabies has been brought under control and wildlife rabies predominates (Western Europe, Canada, and the United States) and (3) rabies-free countries (mostly islands, including England, Australia, and Japan) (De Serres *et al.*, 2008). Taxonomically, the

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rabies virus belongs to the genus Lyssavirus ("lyssa" means madness) and family Rhabdoviridae. Furthermore, rabies virus geographic and host range with genotype varies at the global level with region specific reservoir species genotypes. In Europe, fox , bats with European bat lyssavirus 1& 2; Asia: Dogs with Classical / Dog rabies virus; Africa: dog, mongoose, antelope with Lagos bat virus, Mokola virus, Duvenhage virus; North America: foxes, skunks, raccoons, insectivorous bats with Classical rabies virus; South America: dog, vampire bats with Classical rabies virus ; Australia: Insectivorous bats with Australian Bat Lyssavius and in Middle East: Wolf, dog with Classical / Dog rabies virus (Yale *et al.*, 2014)

The disease circulates in two ecological cycles viz., the urban cycle wherein dogs are the major reservoirs and the sylvatic cycle involving varied wildlife species. Rabies that is transmitted sporadically from reservoir to non-reservoir species is referred as "spillover". The disease is maintained by spillover of infection from the wild life to dogs and vice versa. Humans and livestock are usually victims of this spill over infection from these primary reservoirs / hosts.

Dogs are the principal reservoirs of rabies virus in developing countries and are responsible for majority (97%) of human infections. Other reservoirs and important vectors of rabies virus include wild and domestic Canidae, including wolves, foxes, coyotes, jackals, bats, cats, monkeys, skunks, raccoons and mongooses in different geographic locations. Other mammals may rarely be infected. Furious form of rabies is usually displayed by Skunks, raccoons, foxes and dogs. Bats show dumb form of rabies and often found on the ground, unable to fly. Furthermore, rabies has the capacity to occur in unexpected species such as marine animals, as demonstrated by a case report in a ringed seal from Norway. These cases support the observation that practically all mammals are susceptible to rabies.

The clinical signs in mongoose were described as variable. Interestingly, foxes known for their extremely cautious nature are said to lose their wild instincts when rabid especially in the prodromal stage. They move into settlements, reach for people and behave like a tamed animal. On the contrary, wild wolves are said to be generally timid around humans, when infected they become exceptionally aggressive and can bite many people and animals in a single attack. Any bat that is active by day, is found in a place where bats are not usually seen (for example in rooms in home or on the lawn), or is unable to fly, is more likely to be rabid. Therefore, it is best to avoid handling any bat. Monkeys can get rabies similar to human beings. However, they tend to die more quickly than humans. Weinmann *et al.* (1979) found that 9-10 monkeys which developed severe symptoms die within 20 days of infection. As for the rodents, the majority of rabid rodent reports were in woodchucks and some rodents as well as lagomorphs develop the furious form of rabies. Thus, various wild animal species act as the reservoirs of rabies but understanding the precise prevalence of rabies in these species at the global level is an herculean task!.

The extent of prevalence of rabies in wildlife determines the institution of suitable control strategies. However, unlike the dog rabies, the prevalence is a difficult attribute to estimate for wildlife disease such as rabies as the required denominator i.e. the population at a specific time or average population size during an interval of time, is almost never known. In view of this, there is paucity of information on rabies in wildlife. Neverthless, as per the CDC, Atlanta report of 2013, ninety-two percent of reported rabid animals in the United States were wildlife. During this period, 53 reporting jurisdictions reported 5,865 rabid animals and 3 human rabies cases to the CDC, representing a 4.8 percent decrease from the 6,162 rabid animals and 1 human case reported in 2012. Relative contributions by the animal groups were as follows: 89 dogs (1.5%), 86 cattle (1.5%), 247 cats (4.2%), 344 foxes (5.9%), 1,447 skunks (24.7%),1,598 bats (27.2%) and 1,898 raccoons (32.4%).

Indian Scenario

In India, rabies is primarily attributed to dogs. As for the rabies in wildlife is concerned, there are scanty reports with paucity of information. However, the details of animal bites comprising of various wild animal species is available but there is no clarity as for the confirmed cases of rabies among such cases is concerned. A national survey in 2003 reported that the biting animal mainly responsible for human rabies death was dog (96.2%) of which majority were strays (75.2%) followed by pet (11.1%), wild (3.5%) and cats accounted for 1.7% in India (Bhuyan, 2012). A study conducted on 250 animal bite victims in Pune showed that dog was the biting animal in 94.4% cases, followed by cat (2.4%), jackal (1.2%), mongoose (1.2%), monkey (0.4%) and horse (0.4%) (Shetty *et al.*, 2005). Recently, Ichhpujani *et al.* (2008) reported that dog bites caused maximum morbidity (92%), followed by monkey (3.2%), cat (1.8%) and fox (0.4%) in India. Of these, most bites were unprovoked (64.3%) by stray animals (64.7%). However, this report provides an overview of epidemiology of animal bites and retrospective information about rabies patients.

Episodes of wildlife rabies in India

India is a habitat for various species of wildlife. Although it is presumed that wildlife could be a potential source of rabies for adjoining domesticated animals and human population, only selected episodes or confirmed case of wild life rabies have been documented. The earliest report is by (Shah and Jaswal, 1976), where in the authors describe the episode of a rabid wolf attacking 12 humans and six animals in six villages in the course of February 3, 1973 before it was surrounded and killed. However, this report describes the complete episode but no laboratory tests have been carried out either on the wolf or human brain samples. Only lab test carried out was assessing neutralizing antibody level in 6 human survivors after 28 days of being bitten by wolf and treatment. These antibodies could be attributed to vaccination.

Singh *et al.* (1981) reported a case of rabies in lioness from Zoological Garden, Chatbir, Chandigarh.

Yet another incidence of lion rabies in 1987 reported by Singh *et al.* , 1991 where a lion at Renuka Safari in Himachal Pradesh had symptom of attacking other lions, biting & striking against wire fence where gums and teeth were injured in the process, pertaining to furious form of rabies seen. The lion was hyper aesthetic with roaring and hyper salivation. The animal succumbed to death on the fourth day after the onset of symptoms.

A dumb case of rabies in tigress from National Zoological Park, New Delhi confirmed from Laboratory investigation (Arora, 1990-91).

In Andhra Pradesh, during 2010, a wolf sneaked into the human habitation and had bitten several persons. The animal was trapped and killed. The brain tissue was found positive for rabies virus based on laboratory tests. All the bitten individuals were immediately provided post exposure prophylaxis and survived (Personal communication Dr Sampath, 2010).

Among wildlife, rabid wolf attacks have been recorded and published based on laboratory tests elsewhere. However, as per our knowledge, until 2013, such evidences of rabies based on thorough laboratory investigation in the Indian wolf (Canis lupus pallipes) have not been recorded in the country. We have reported one such thoroughly investigated case of rabies in wolf (Isloor *et al.*, 2014). As per this report, on 29 June 2013, a wolf (Canis lupus pallipes) entered two villages (2 km away from each other). The wolf bit 15 cattle in the first village, and then ran into a nearby village and bit 10 other cattle, 2 calves, one woman, and two men. Villagers later trapped and killed the wolf. The head was transported to the OIE Reference Rabies Diagnostic Laboratory, Dept. of Veterinary Microbiology, Veterinary College, Bengaluru for laboratory diagnostic confirmation and RABV characterization.

Direct fluorescent antibody (DFA) test and a direct rapid immunohistochemistry test (dRIT) were used for the laboratory confirmation of rabies in wolf from the southern Indian state of Karnataka. The polymerase chain reaction (PCR) amplified products of complete N and G genes of this wolf RABV were sequenced and processed. High homology and genetic relatedness of this rabies virus isolate were revealed from BLAST results and phylogenetic analysis with those from nearby geographic locations such as in India (human, buffalo), Pakistan (wolf, cattle) and Nepal (goat). Further analysis of the deduced amino acid sequences of the complete N and G proteins revealed that wolf rabies virus isolate recovered in this study belonged to the Arctic-like lineage in Indian subcontinent, which is wide spread throughout the region. To ascertain if wolves (and other wild species) serve as a significant population for RABV enzootic maintenance and to characterize regional lyssa viruses in India these findings should be emphasized for the necessity of epidemiological surveillance. Recently, another case report of rabies in wolf (Canis lupus) from Karnataka was described by Sumana *et al.* (2014). In this episode, brain sample from suspected rabid wolf from northern part of Karnataka was subjected for DFA and RT-PCR using partial N gene (607 bp) as the target. Further sequencing and phylogenetic analysis revealed that this wolf isolate was closely related to canine rabies virus suggesting the mechanism of species spillover. This study also provides scientific evidence of same classical rabies virus circulating in both sylvatic and urban forms of rabies in India. Such more studies are warranted in wild rabies epidemiology and prevention of rabies in associated human population and domestic animals.

In yet another south Indian state of Kerala, a boy bitten by a squirrel during 2013 in Kerala died of clinical manifestation of rabies. However, no laboratory based confirmation was made in this episode. Furthermore, a wild pig from Nilambur in Kerala during 2014 suspected for rabies with biting tendency was killed and confirmed to be rabid based on DFA positivity of the brain tissue (Personal communication Dr. Vijayan).

Hyenas are the animals of the family Hyaenidae of the Carnivora. These animals are behaviorally and morphologically similar to canines in several aspects. A brain sample of Hyena from south Gujrat was found positive by DFA and RT-PCR (Personal communication Dr. Jhala, 2015).

Omesh,, 2016 reported a rabies in monkey (M. mulatta) from Shimla municipality. The monkey was found to be positive for Rabies virus by biological test (BT) done by the Central Research Institute (CRI) Laboratory, Kasauli, Himachal Pradesh.

Prevention and Control of disease in wild animals

Prevention of disease from infecting humans and domestic animals and reduction in the economic and personal costs associated with a rabies outbreak should be the overall goal of rabies management (Uhaa *et al.*, 1992). The differences in population dynamics, structure and behavior among the major wildlife reservoirs described by passive disease surveillance system is an important component of any management program and an approach that tracks rabies variants in populations providing temporal and spatial distribution information (Hanlon *et al.*,1999). When appropriate control programs applied to wildlife populations, the spread of a rabies epizootic and introduction of the disease can be prevented or even disease in enzootic or epidemic areas can be eliminated (Wandeler, 1991).

The approaches to control rabies in wildlife are: elimination of the reservoir species, elimination of rabies in the reservoir species, or protection of victim species from rabies infection via a reservoir (Rupprecht *et al.*, 2001). These methods may be applied in combination. Control Elimination of a reservoir species is impractical, expensive, ecologically unacceptable (unless an introduced species), and ethically unacceptable (Rupprecht *et al.*, 2001).

Identification of objectives is the first step with Rabies management program. Later, successful management programs will identify and execute a control method that affects mainly the target (i.e., reservoir) species (Winkler and Jenkins, 1991). However, Hanlon *et al.* (1999) states that the management of rabies in wildlife is complicated by the ecologic and biologic factors associated with wildlife reservoirs. An important public health problem originating in wildlife can be managed by the multiagency approach but the limitations of available control methods, and the broad range of public attitudes toward wildlife makes management difficult". Vaccination of wildlife reservoir species is the potential method for control of rabies. In trap-vaccinate-release (TVR) programs targeting reservoir species live-trap and manually inject liquid vaccine (i.e., parenteral vaccination; Wandeler 1991 and Hanlon *et al.*, 1999). However, parenteral rabies vaccination efficacy in wildlife has not been established. Whereas in the United States, an oral rabies vaccine is currently licensed for use in raccoons (Jenkins *et al.*, 2001). For a large scale use oral rabies vaccination (ORV) may be more economically and technically feasible alternative. ORV may be more intrusive on the landscape since it is less invasive to individual animals than TVR. Immune response is elicited once ORV baits attractive to targeted reservoir species is taken (bitten) into the mouth or pharyngeal tissues releasing an encapsulated, attenuated rabies virus vaccine (Wandeler, 1991 and Hanlon *et al.*, 1998). ORV has been successfully applied to control rabies in raccoons, fox, and coyotes in North America; however, prior implementation of ORV programs should consider numerous questions.

Recombinant vaccines have been proved as safe and efficacious alternative to modified-live vaccines (Rupprecht *et al.*, 1995). The only ORV currently licensed for State and Federal rabies control programs in the United States is the vaccinia recombinant Raboral V-RG vaccine manufactured by Merial (Jenkins *et al.*, 2001).

Vaccine is delivered via baits. Baits can be made of many different materials (e.g., fishmeal, dogfood, meat, cheese, fermented egg products, cornmeal (Rosatte *et al.*, 1998). Scented baits (e.g., species-specific scent lures or urines) are used to attract target species while attempting to be less attractive to not-target species (Rosatte *et al.*, 1998). The distribution of baits can be done manually (while walking or from a boat or vehicle) in a random or uniform manner or by direct placement in preferred habitats (Anthony *et al.*, 1990). Baits can be broadcast by aircraft or by using bait-delivery devices (Andelt and Woolley, 1996). Bait uptake by all age classes (rabies reservoir) can be greatly enhanced by placement of baits in habitats preferred by targeted species, either as a single approach or in combination with other distribution methods, thus increasing the proportion of the host population that is vaccinated (Robbins *et al.*, 1998).

Europe and North America has tested the safety of baits ingested or contacted by target and non-target species. According to Rupprecht *et al.*, 2001 baits and vaccines have been found to be safe in >50 vertebrate species including non-human primates (Rupprecht *et al.*, 1992) and immune compromised animals (Hanlon *et al.*, 1997). Resultant level of protection by vaccine exposure is unknown in non-target species but it may also result in an immune response. Danger of a public health risk from ORV remain unfounded (McGuill *et al.*, 1998). Bait exposure to human in the field has been extremely low, especially when information explaining the program has been made available to the public in advance of and during the baiting (Rosatte *et al.*, 1990; McGuill *et al.*, 1997 and Robbins *et al.*, 1998). To identify the purpose and to provide contact information, baits are commonly labeled. Other outreach programs include the mailing of informational flyers to treatment-area residents, announcements through local media, presentations to local groups and schools, and posting warning information notices in treatment areas.

In areas with reoccurring rabies epizootics ORV has proven to be cost-effective, attractive and acceptable by the public (Robbins *et al.*, 1998), and efficacious (Robbins *et al.*, 1998 and Hanlon *et al.*, 1998). MacInnes *et al.*, 2001 demonstrated that ORV has been used to eradicate rabies from enzootic area such as eastern Ontario. Control of rabies has been achieved in programs where bait consumption by the target species has attained or exceeded 60% (Wandeler, 1988, Rosatte *et al.*, 1992 and Robbins *et al.*, 1998), although projects with uptake rates above and below this threshold also have reported being successful (Fearneyhough, 1996). Apparently it may not eradicate the disease in the population in all cases but these programs are successful at stopping or slowing the rate of rabies progression. However, rabies has been eliminated from red fox populations in France (Aubert *et al.*, 1994) and in eastern Ontario (MacInnes *et al.*, 2001). A well-coordinated aggressive effort eliminated rabies from a 30,000 km study area in 7 years in Ontario. In United States, Fox rabies was absent from the area for 3 years after ending the ORV program but raccoon rabies was then introduced from adjacent areas (MacInnes *et al.*,2001). However, Germany has noted the persistence of rabies in treatment areas and have required continuation of oral vaccination efforts. Additional ORV applications were required to boost immunity of the population previously treated and to treat individuals immigrating into the area and to the portion of the population added through recruitment (Mitchell *et al.*, 1997). The ability to eliminate fox rabies doesn't appear to be related fox density but to fox ecology (MacInnes *et al.*, 2001); however, the influence of gaps in ORV application must also be considered.

The control programs strategies are expected to meet the conditions of the treatment area, nature of the rabies event, location in proximity to human populations, density and dynamics of the host population, and other factors.

Population declines are commonly seen in all age classes during a rabies epizootic. This mortality differs from other mortality factors that routinely affect the juvenile and first year age classes (Blancou *et al.*, 1991). Density within a given area is controlled more by social regulation, habitat suitability, food availability (carrying capacity) and to a lesser degree mortality factors. Predicting post-epidemic population densities may be difficult without the knowledge of specific factors controlling population densities (i.e.,carrying capacity, habitat, reproduction-recruitment, mortality factors, and the potential for emigration), (Schubert *et al.*, 1998). Therefore, rabies control programs with effect of vaccination programs on wildlife in different species and under different conditions remains controversial reflecting the variety of outcomes (i.e., decreases, increases, no change).

Oral Rabies Vaccine programs have been conducted in Europe (Wandeler *et al.*, 1988 and Wandeler, 1991), Canada (Rosatte *et al.*, 1992 and MacInnes *et al.*, 2001). In the United States ORV programs have been conducted for raccoon rabies control in New Jersey, Massachusetts, New York, Florida, Vermont, and Ohio (Hanlon *et al.*, 1993, Hanlon *et al.*, 1996, Mitchell and Heilman 1996, Robbins *et al.*, 1996, Roscoe *et al.*, 1996 and Rupprecht *et al.*, 2001) and for coyote and gray fox in Texas (Fearneyhough, 1996). No oral rabies vaccination program has been initiated for skunks because they are largely resistant to the recombinant oral vaccine licensed in the United States (Rupprecht *et al.*, 2001).

Conclusion

Wildlife rabies is lesser known in India and is the tip of the iceberg. Although scanty, the available literature and documented evidences based on laboratory investigations prove the existence of it's sylvatic cycle. Undoubtedly this situation could be resulting in spillover of infection to domesticated animals and human population in the geographical locations in the vicinity of forest especially in the backdrop of deforestation. However, most of such episodes could be going unnoticed due to various reasons and thereby indirectly underestimating the impact of sylvatic rabies in India. In view of this scenario, the little information available on wildlife rabies in India emphasizes the necessity of epidemiological surveillance to characterize regional lyssaviruses to ascertain if wild species serve as a significant population for enzootic maintenance of rabies virus in India. Rabies control programs must be justified and must be well designed to be efficacious, cost effective, be publicly supported. It should have negligible negative impact (on wildlife, humans, and landscapes). Rabies in wildlife is a threat to domestic animals and humans population in rural areas. Dept. of Environment and Forest, Animal Husbandry and AWOs need to be coordinated to initiate control of rabies in wildlife. Submission of brain samples from suspected wild animals be encouraged for lab confirmation. Use of ORV having negligible negative impact on wildlife, livestock, humans and landscapes can be explored in India.

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