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A Case Study of Rabies in a Buffalo in Chennai, Tamil Nadu, India.

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ABSTRACT

As per the WHO estimate roughly 36% of the world's rabies deaths occur in India each year, most of those when children come into contact with infected dogs. The occurrence of rabies in cattle's like buffalo is usually overlooked or un-noticed and hence underreported. On 22rd June 2014 the residents of Mylapore (Chennai) launched a complaint against a wild behaviour of a buffalo to Blue cross of India. Responding promptly, the doctors from Blue Cross immediately shifted the buffalo to its campus. On the next day 23th June 2014, the buffalo died within the campus of Blue Cross. The decapitated head of Buffalo was sent to Madras Veterinary College, Veppery in Chennai for histopathological examination. The histopathological examination of brain (Hippocampus), turned out to be positive for Negri bodies and Fluorescence antibody test (FAT) established the diagnosis of rabies.

Keywords: Rabies, Buffalo, Blue Cross, Fluorescence antibody test, Negri bodies

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INTRODUCTION

Rabies has terrified man since antiquity. The fear is by no means unfounded since the disease is invariably fatal and perhaps the most painful and horrible of all communicable diseases in which the sick person is tormented at the same time with thirst and fear of water (hydrophobia). Fortunately, development of rabies can be prevented to a large extent if animal bites are managed appropriately and in time.

Rabies is a zoonotic disease (a disease that is transmitted to Humans from animals) that is caused by a virus. The disease affects domestic and wild animals, and is spread to humans through close contact with infectious material, usually saliva, via bites or scratches of these infected animals.

The virus is a single stranded RNA virus belonging to the genus Lyssavirus of the family Rhabdoviridae [1]. The incubation period varies inversely with the proximity of bite to the central nervous system, but has been reported to vary from 21-80 days in dogs to upto 209 days in horses and cattle [2]. The clinical disease in cattle manifests as change in natural behaviour, excessive salivation, excitability mania, which ends in motor paralysis and death. In man and cattle mortality is 100% [2]

The incubation period in buffalo is said to be 2-12 weeks or longer. Early signs seen may include separation from the rest of the herd, anorexia, and docility. Milk production drops and bulls become more sexually active. After a few days, the aggressive phase may begin with the animal attacking. A fixed stare may be seen and the animal grinds its teeth. It sometimes develops pica. There is frequently inability to swallow with copious salivation and often develop a characteristic hoarse bellow. Tail paralysis is seen; posterior paresis and a swaying gait are noted. After one to five days, animals may assume ventral or lateral recumbency and die in convulsion, or become progressively paralyzed, comatose and die.

Case Report

On 22 June 2014, the residents of Mylapore in Chennai witnessed an unusual behaviour of buffalo, butting the innocent pedestrians of the street. The timid creature, not so aggressive like bull, usually hauling in a slow pace, behaved so wildly, creating utter nuisance to the public. This irritable nature & unprovoked wild behaviour alarmed the residents of the area. As nobody claimed to be the owner of the buffalo, the residents complained to the Blue Cross of India.

Acting swiftly, the doctors from Blue Cross promptly made a presumptive diagnosis of rabies, due to unusual bellowing of the buffalo & excessive salivation. The buffalo was sedated with xylazine and transported to Blue Cross and leashed (Figure 1). The next day buffalo died within the campus of Blue Cross, strengthening the presumptive diagnosis of rabies.

Figure 1: The tied rabid buffalo of this case report.

The decapitated head was sent to Pathology Department of Madras Veterinary College in Veppery, India. The demonstration of Negri bodies in hippocampus of brain and Fluorescent antibody (FAT) test finally confirmed the diagnosis of rabies infection.

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MATERIALS AND METHODS

Since the buffalo died had after rescuing, the decapitated head was sent to Madras veterinary College in Chennai for histopathological examination.

The Negri bodies were detected by impression smears of the brain and stained by Seller's technique. The slide was made from Ammon's horn, from the cerebral cortex and finally from cerebellum. Samples (at least 6) from these 3 areas on each side of the brain were examined microscopically. With a pair of scissors a small transverse section of brain tissue (Ammon's horn, cerebrum or cerebellum) were cut and placed in a blotting paper with the cut surface facing upward. A clean slide is touched against the cut surface of section and pressed downwards gently downwards with just enough pressure exerted to create a slight spread of the exposed surface against the tissue. while still moist, the slide is flooded with Seller's stain (basic fuchsin and methyelene blue in methanol) was allowed to remain for few seconds, rinsed under the tap, and dried at room temperature without blotting.

The Fluorescent antibody (FAT) test was performed as per kit manufacturer's guidelines: A small fraction of the brain sample was smeared using wire loop on one part of a slide and then was air dried and fixed in cold acetone for one hour at -20^{o}C.The slides were air dried and then the rabies conjugate was applied at 1:40 and incubated for 30 minutes at 37^{o}C in a humid chamber after which excess conjugate was removed from the slides by rinsing it with 7.4 pH PBS solution about 3-5 minutes and was allowed to air dry. The cover slips were mounted with buffered glycerol mounting medium and the slides were examined using a fluorescence microscope within 2 hours after staining. When brilliant apple-green fluorescence colored or greenish yellow objects is exhibited against a black background then the test slide is positive

RESULTS

The death of buffalo followed by demonstration of Negri bodies (Figure 2) in hippocampus of brain and Fluorescent antibody test (FAT) (Figure 3) confirmed the case of rabies in buffalo.

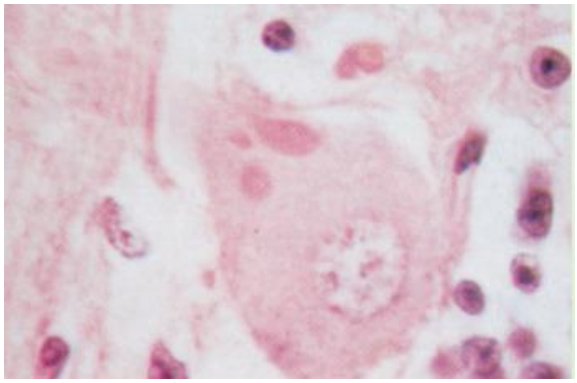

Figure 2: Demonstration of Negri Bodies, inclusions within the cytoplasm of nerve cells of the brain tissue.

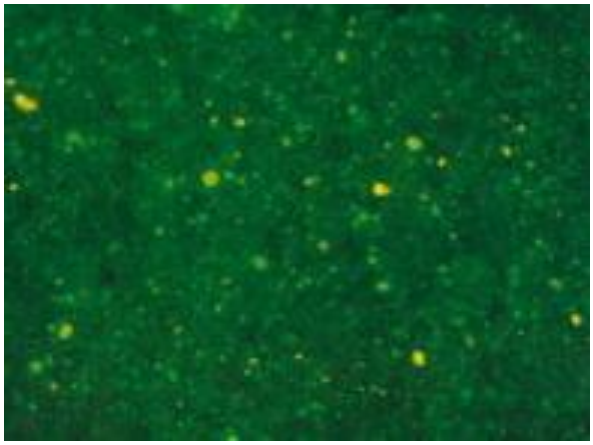

Figure 3: Demonstration of fluorescent-antibody test (FAT) on brain tissue.

DISCUSSION

The diagnosis is by demonstration of Negri bodies in brain tissue. Negri bodies as round or oval inclusions within the cytoplasm of nerve cells of animals infected with rabies. Negri bodies may vary in size from 0.25 to 27 µm. They are found most frequently in the pyramidal cells of Ammon's horn, and the Purkinje cells of the cerebellum. They are also found in the cells of the medulla and various other ganglia. Negri bodies can also be found in the neurons of the salivary glands, tongue, or other organs. Staining with Mann's, Giemsa, or Sellers stains can permit differentiation of rabies inclusions from other intracellular inclusions. With these stains, Negri bodies appear magenta in colour and have small (0.2 µm to 0.5 µm), dark-blue interior basophilic granules.

The presence of Negri bodies is variable. Histological staining for Negri bodies is neither as sensitive nor as specific as other tests. Some experimentally-infected cases of rabies display Negri bodies in brain tissue; others do not. Histological examination of tissues from clinically rabid animals show Negri bodies in about 50% of the samples; in contrast, the dFA test shows rabies antigen in nearly 100% of the samples. In other cases, non-rabid tissues have shown inclusions indistinguishable from Negri bodies. Because of these problems, the presence of Negri bodies should not be considered diagnostic for rabies [3].

However the gold standard for diagnosis of rabies is direct fluorescent antibody test. Combined accuracy and speed of this technique was introduced by Goldwasser and Kissling in 1958. The FAT is accurate, sensitive and rapid. Results can often be obtained within 1 to 2 hours of receipt of the specimen.

As per the National Centre for Communicable Disease, India, rabies is an acute viral disease that causes fatal encephalitis in virtually all warm blooded animals including human beings. The virus is found in some wild and domestic animals, and is transmitted to other animals and to humans through their saliva following bites, scratches, licks on the broken skin and mucous membrane. In India dogs are responsible for 97% of human rabies; followed by cats (2%); jackals, mongoose & others (1%).The disease is mainly transmitted by the bite of rabid dog [4].

From the above data, it is obvious that rabies in cattle's like buffalo is a rare or uncommon incidence. This uncommon incidence of rabies in buffalo should never be ignored. But how did the Buffalo acquire the disease, either through the bite of rabid dog or a rodent is not known. The transmission of disease is mainly through saliva of the infected animal. The chances of acquiring the infection through the consumption of buffalo milk is rare or nil, but can't be ruled out either. On November 12, 1998, the Virology Laboratory of the Massachusetts Department of Public Health (VLMDPH) diagnosed rabies in a 6-year-old Holstein dairy cow from a farm in Worcester County [5]. Further analysis of the cow's brain tissue with monoclonal antibodies revealed the cow was infected with a variant of the rabies virus associated with raccoons in the eastern United States. The cow had loss of appetite beginning November 4 and hyper-salivation beginning November 6. An intestinal obstruction was suspected initially as the cause of illness. However, the cow became ataxic and aggressive and died on November 8. The cow had been milked 12 times during the week before death. Milk from the cow had been pooled with milk collected from other cows, and an unpasteurized portion was distributed for human consumption. Public health investigations identified 66 persons who drank unpasteurized milk collected from this dairy during October 23-November 8. All 66 received rabies post-exposure prophylaxis (PEP). Although transmission of rabies virus from consuming unpasteurized milk from an infected animal is theoretically possible, no human has ever been reported to develop rabies via this route. Milk that has been heat pasteurized presents no risk for rabies virus transmission.

In clinical practise post exposure prophylaxis is strongly recommended for infected stray dog bite. The clinician should also, consider an exposure to cattle (especially wild behaving animal) before excluding a case of Rabies. The highly susceptible population like herdsman, cattleman, and shepherd should undergo periodic pre exposure vaccination.

REFERENCES

[1] George M,James N and George STB. 1996, Rabbis and Rabies:A pictoral History of Rabies through the ages.Laboratorios Baer.S.A DE C.V Mexico,D.F

[2] http://seard.info/fichiers/articles1999086087L.PDF

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[3] http://www.cdc.gov/rabies/diagnosis/histologic.html
[4] National Guidelines on Rabies Prophylaxis/Govt of India/2013
[5] CDC.gov/MMWR/Weekly/March 26,1999/48(11);228-229