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1 Vitamin -Medication and Post Vaccination Rabies Antibody
2 Titres in Nigerian Dogs

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6 **Abstract**

8 **Index terms—**

9 **1 I. INTRODUCTION**

10 Rabies is fatal encephalitis, caused by Rabies virus ??Nandi and Kumar, 2011). It is a zoonosis that occurs in
11 all parts of the world, accounting for more than 59,000 deaths per year ??WHO, 2013). Of human deaths caused
12 by rabies, 56 % occur in Asia while 44% is in Africa (Bourhy et al., 2010, WHO, 2013; OIE, 2016). The disease
13 affects all warm blooded animals ??Ogunkoya et al., 2003), both wild and domestic including man. This wide
14 host range makes its control difficult.

15 Main method of rabies control in most countries is annual vaccination of dogs and cats. Such vaccinated
16 animals need to be tested to ensure they have high levels of rabies antibodies but presently, the method of
17 assessing effectiveness of anti-rabies vaccination processes is by challenging vaccinated laboratory animals with
18 the virus (WHO, 2013).

19 Virus-neutralization tests to assess immune response in vaccinated animals also involves challenging laboratory
20 animals with the live virus. By such methods antibody titre of 0.5 IU/ml has been suggested as protective (WHO,
21 2013) but it has also been reported that vaccinated animals with lower antibody levels survived RABV challenge.
22 Other reports also have it that some animals with neutralizing antibody titres at time of challenge came down
23 with the disease (Aubert, 1992) though such animals were better protected against RABV than those without
24 detectable virus neutralizing antibodies (Aubert, 1992).

25 The type of vaccine used, number of vaccinations, intervals between vaccinations and blood sampling, age at
26 vaccination, sex, reproductive status, size and breed can influence antibody response of animals to anti-rabies
27 vaccines ??Mansfield et al., 2004). Also, it has been reported that in post-vaccination serological studies, the
28 percentage of animals with inadequate titres range between 3.10 to 8.

29 **2 II. MATERIALS AND METHODS**

30 Twenty Nigerian mongrel dogs, aged 3 months and above were randomly assigned to 4 groups (A, B, C and D)
31 of 5 dogs each. They were vaccinated with a foreign anti-rabies vaccine (Provac ®). Group A was given post
32 vaccination vitamins‘treatment (Vetzyme®) for one day.

33 Group B was given the post vaccination vitamin-treatment for 2 days, group C was treated for 3 days while
34 group D served as control (no post vaccination vitamin-medication). Two weeks post vaccination, blood samples
35 were collected from each dog for passive haemagglutination (PHA) test, to determine titres of antibodies against
36 rabies.

37 Chicken RBCs were sensitized with Rabies virus (Gough and Dierks, 1971). To 30 ul of 0.25 % chicken RBC,
38 equal volumes (30 ul) of Rabies virus and of 0.1% chromium chloride in 0.86 % NaCl were added. The mixture
39 was kept at room temperature for 5 minutes. Then the RBC was washed again, 3 times with 0.86 % NaCl. To
40 obtain a highly concentrated RBC, the entire For the PHA test, to a clean "V" bottomed microtiter plate, 0.05ml
41 of 0.86 % NaCl was put in each well in a row. Equal volume of the dog-serum (after inactivation) was added to
42 the first well in the row and serially double-diluted till the last well. Then, equal volume of the sensitized chicken
43 RBCs was added to each well and the setup was incubated at 4 0 C for 1 hour. Reciprocal of highest dilution of
44 each dog-serum sample which gave complete agglutination of the Rabies virus-sensitized chicken RBCs was read

4 V. CONCLUSION

45 as titre of Rabies antibody in it. Means of antibody titres of the 4 groups of dog sera were compared by one way
46 analysis of variance (ANOVA).

47 3 III. RESULTS

48 Rabies antibody titres in sera of Nigerian dogs vaccinated with anti-rabies vaccine and given post vaccination
49 Vitamin-B treatment increased from 24.74 ± 0.08 in the controls to 64.08 ± 0.03 in those treated with the vitamins
50 for 1 day, 89.29 ± 0.17 in those treated with the vitamins for 2 days and 140.73 ± 0.14 in those treated with
51 the vitamins for 3 days (Table 1) washing fluid was decanted, leaving just sufficient for homogenization of the
52 RBCs. Haematocrit concentration of the RBC was determined by inserting a capillary tube into the container
53 and allowing the tube to fill up to one quarter. Then its other end was sealed before it was centrifuged in a
54 microcentrifuge at $1176 \times g$ (3000rpm) for 5 minutes. The haematocrit value was read on a PCV reader. amin
55 There is therefore a need to adopt a simpler, cheaper and more rapid test so that anti-rabies vaccinations can
56 be verified to ensure that vaccinated animals have protective antibody titres. Also, post vaccination Vitamins-
57 treatment is known to enhance immune response in poultry.

58 It may also enhance rabies antibody titres in vaccinated dogs and so prolong the time vaccinated dogs would
59 remain protected.

60 According to WHO recommendations, titre of 0.5 IU/ml or its equivalent is adequate vaccination response and
61 a booster vaccination is recommended once the level goes lower. Despite annual vaccination of Nigerian dogs,
62 records still show that dog bites are still responsible for a very high percentage of cases of human rabies in Nigeria
63 ??Ogunkoya, 2008). Yet the immune status of Nigerian dogs (even when vaccinated) is not known. Existence
64 of these prophylactic measures against rabies does not diminish the fact that rabies is still a significant cause
65 of human and animal mortality (WHO, 2018). Success of post-exposure prophylaxis against rabies infection in
66 endemic countries is usually hindered by obstacles such as cost of vaccine, availability of post infection care and
67 lack of awareness about the disease in rural areas. Critical prophylactic regulations such as massive vaccination
68 of dogs often fail to achieve their aims in rabies-endemic countries due to lack of awareness and vaccine-related
69 issues such as inadequate transportation and storage facilities for vaccines (Ullas et al., 2012).

70 Rabies and canine distemper are the most endemic viral diseases of dogs in Nigeria to which vaccination is
71 usually applied as a control measure (Ezeibe et al., 2008; ??woha, 2015). In Nigeria, though rabies has been
72 responsible for many human deaths and suffering, vaccination has been poorly implemented. Shortcomings in
73 anti-rabies vaccination in Nigeria include low coverage and poor compliance rates as in most other developing
74 countries (Fagbami et al., 1981).

75 Challenges to use of vaccine in rabies control include, high cost of materials, lack of reagents, lack of biologics,
76 lack of chemicals, lack of consumables and other supplies, lack of constant electric power and failure of governments
77 to enforce legislations on annual vaccination of dogs and cats (Foggin and Swanepoel, 1985). Also, there are
78 problems of lowered vaccine potency due to improper cold chain, failure to achieve sufficient herd immunity after
79 vaccination and antigenic variations (Foggin and Swanepoel, 1985).

80 In this study, post vaccination treatment of the Nigerian dogs with Vitamins improved their antibody responses
81 to the vaccine. Prolonging the post vaccination Vitamins' treatment for three days increased the antibody titres
82 from 24.80 ± 10.91 to 140.80 ± 31.35 . Stantic-Pavlinic et al. ??2004) reported that vitamin C enhances interferon
83 production in humans and could therefore be used for stimulation of interferon response to rabies vaccines. So,
84 these results agree with their suggestion. It is also in agreement with Igado et al (2010) who reported that
85 vitamin C has immune-potentiating effects.

86 Vetzyme is a tablet dosage form of Vitamins and antioxidants. Its use in this study was for convenience.
87 With the tablet-vitamin medication, dog owners do not need to come back to the clinic for days after anti-rabies
88 vaccination. This makes adoption of post vaccination Vitamins-treatment for control of rabies, convenient.

89 Currently, most veterinarians in Nigeria do not practice post vaccination Vitamins medication for rabies
90 control. Though dogs in the control group in this study had titres up to the protective titre, such titres may not
91 remain at protective level for a long time. So, it would be good to ensure higher antibody levels in vaccinated
92 dogs so that the titres may not fall below the protective level before the dogs are revaccinated. Administering
93 multivitamins to dogs for three days post anti-rabies vaccination would ensure high levels of antibodies that
94 would remain at protective level till next annual revaccination.

95 4 V. CONCLUSION

96 Post vaccination Vitamins treatment of dogs under the Nigerian environmental conditions is necessary to improve
97 their antibody responses to control rabies in Nigeria. For Nigerian veterinarians to start post vaccination
98 assessment of dogs for rabies antibodies, there is a need to adopt a simple, inexpensive and yet reliable diagnostic
99 test. against rabies. Also, post vaccination assessment of dogs for rabies antibody titres can be adopted if
100 veterinarians are taught to use the rapid and inexpensive passive haemagglutination test which does not require
101 sophisticated equipment and results are got within 3 hours. These two strategies could enhance the success of
102 rabies control efforts in Nigeria.



Figure 1:

1

IV. DISCUSSION

Rabies is the most fatal infectious disease that is known worldwide (WHO, 2018). Although it has been neglected for some time, international health organizations (WHO, OIE and FAO) are now working together to eradicate the disease by the year 2030. They have adopted different strategies to achieve this goal, mainly through intensive vaccination of domestic and wild carnivores. New generation anti-rabies vaccines that are more economical and more efficient than conventional vaccines are now being used. There are even oral vaccines against rabies (plant based) that are showing promising results (Lucka et al., 2015; Laere et al., 2016).

Figure 2: Table 1 :

Previous clinical studies have found that Rabies virus neutralizing antibody titres of between 8-16 IU/ml is protective (CDC, 2016) but the neutralization test is complex, such that most field veterinarians cannot run it. Results of Rabies passive correlation coefficient of 0.81 with results of Mouse Serum Neutralization test (Gough and Dierks, 1971) and sophisticated and less Its adoption would veterinarians in most

haemagglutination (RPHA)ye
RPHA test requires less
reduce
limitations

Figure 3:

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