# Chapter 4

# Testing Newcastle Disease Virus Vaccines for Efficacy

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### **P.B. Spradbrow \***

THE standard tests for Newcastle Disease Virus (NDV) vaccines include tests for extraneous microorganisms that indicate the microbiological purity of the vaccine, tests for safety that show the vaccine is innocuous, potency tests that measure the antigen content of the vaccine and efficacy tests that demonstrate the immunisation potential of the vaccine. Efficacy tests should be conducted under conditions similar to those that will prevail when the vaccine is used in the field. Organisations that are contemplating pilot trials on village chickens may first wish to conduct their own efficacy tests, as an indication that the pilot trial should proceed. The efficacy test should indicate that, at least under optimal conditions, the test vaccine will protect against local strains of Newcastle Disease virus. Field staff can then proceed, confident that a successful field vaccination program is within reach.

A basic efficacy test compares the response of vaccinated and control chickens to challenge with a virulent strain of microorganism (see summary Table I). The Newcastle Disease efficacy test should use local pathogenic field isolates of Newcastle Disease virus.

The first requirement is susceptible chickens, that is chickens that are free from Newcastle Disease antibodies and that will be able to respond to vaccination and challenge. Test and control chickens should be from the same source and separate isolation facilities will be required for test and control chickens. Ideally the chicks should be hatched from specific-pathogen-free eggs or from eggs from a farm that has no Newcastle Disease problem and that uses no Newcastle Disease vaccine. If there is no option to the use of local, commercial chicks, they will need to be held in isolation until maternally-derived antibodies have waned.

The following is an outline for a suggested efficacy test:

Day 1. Select one-day-old chicks (antibody-free if possible) and place them in isolation. The group should be large enough to supply, after brooder losses, at least 20 test chickens, at least 10 control chickens and at least 5 chickens to serve as donors and source of infection and challenge with the pathogenic virus.

Day 21. Bleed all chickens and perform Newcastle Disease haemagglutination-inhibition tests. The chickens should be free of antibodies. Separate and isolate the test and combined control/donor groups. They should have no contact with other chickens to avoid any chance of spread of vaccine virus between groups. Vaccinate each test chicken by any appropriate route by eye drop, intramuscular injection, etc., with one field dose of vaccine.

Day 35. Bleed all chickens and perform haemagglutination-inhibition tests. Revaccinate all test chickens, using the same dose of vaccine and route of inoculation.

Day 49. Bleed all chickens and perform NDV haemagglutination-inhibition tests. Inoculate the donor chickens intranasally with at least one 100% lethal dose of a local isolate of virulent Newcastle Disease virus. Place all the chickens together, so that the test birds and control birds are challenged by a natural route by contact with the donor birds.

Days 50-63. Observe the chickens closely. Autopsy all dead birds and examine for signs of Newcastle Disease. The donor birds could be expected to die within 4 or 5 days, and the control birds within the following week. The degree of protection afforded the vaccinees will need to be set by the experimenter and will depend on the degree of protection that is being sought in the field. The protection rate required will depend on the control policies of the livestock authorities, and could range from 60 to 100%.

<sup>\*</sup> Department of Veterinary Pathology and Public Health, University of Queensland, St. Lucia, Brisbane, Australia.

Day 1	50 day-old chickens collected and isolated from any infectious diseases.	
Day 21	HI-test for NDV antibody all chickens. Only chicke two separate and isolated groups: Group 1 Vaccination Group	ns free from HI-antibody to be used. Divide into Group 2 Control and Donor Group
	20 chickens Vaccinate all chickens with $10^6 \text{ EID}_{50}$ /chicken Route: any	10 control and 5 donor No vaccination
Day 35	HI-Test Revaccinate chickens using same method as day $21$ with $10^6$ EID <sub>50</sub> /chicken	HI-Test No vaccination
Day 49	HI-Test	HI-Test Inoculate 5 donor chickens with lethal dose of local NDV
	Combine Groups 1 and 2, Vaccinated, Control and Donor in one pen. Donor group act as source of challenge.	
Days 50-63	Observe all chickens. Autopsy all dead chickens for presence of NDV.	
Expected Po	sitive Result: Donor birds die 4-5 days after infectior Vaccinated chickens survive.	n. Control birds die within the next 7 days.

### TABLE 1.