

Acknowledgements

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Abbreviations

| ACIAR | Australian Centre for International Agricultural Research |
|---------|---|
| CLW | Community Livestock Worker |
| FAO | Food and Agriculture Organisation of the United Nations |
| G | Gauge |
| HI | Haemagglutination Inhibition |
| I-2 | Thermostable, live, avirulent ND vaccine available for local production |
| INIVE | National Veterinary Research Institute, Mozambique |
| mL | Millilitre |
| ND | Newcastle disease |
| NDV4-HR | Thermostable, live, avirulent, commercial ND vaccine |
| NGO | Non-governmental organisation |
| OIE | Office International des Epizooties |
| PRA | Participatory Rural Appraisal |
| PTD | Participatory Technology Development |
| TOT | Transfer of Technology |
| VETAID | British NGO specialising in community-based livestock development |
| μL | Microlitre |

1

Introduction

Rural poultry production is recognised as an important activity in all developing countries. However, over the past few decades, the focus has been on the production of commercial poultry in rural areas, while traditional village poultry systems have been largely ignored. Chickens in traditional village poultry systems provide scarce animal protein in the form of meat and eggs, and are available for sale or barter in societies where cash is not abundant. They are generally owned and managed by women and children (Guèye 2000; Spradbrow 1993–94). Village chickens also fulfill a range of other functions for which it is difficult to assign a monetary value. They are active in pest control, provide manure, are required for special festivals and to meet social obligations, they are essential for many traditional ceremonies and traditional treatment of illness (Alders 1996).

Although the output of traditional village chickens in terms of weight gain and number of eggs per hen per year is low, it is obtained with minimum input in terms of housing, disease control, management and supplementary feeding (Tables 1 and 2). Any cost-effective strategy that increases the productivity of these birds will assist in poverty alleviation and the improvement of food security. The increased availability of village chickens and eggs should result in an improved intake of protein by the population and increased access to cash and other resources. Chickens are often essential elements of female-headed and poor households. This is a particularly important contribution in areas where child malnutrition is common. Malnutrition has wider implications for development because protein-energy malnutrition in children inhibits their growth, increases their risk of morbidity, affects their mental development, and reduces their subsequent school performance and labour productivity (Pinstrup-Andersen et al. 1993).



2

The importance of Newcastle disease in village chickens in developing countries

The major constraint to production of village chickens in many developing countries is Newcastle disease (ND) (Alexander 1991, Spradbrow 1988). In these countries, circulating strains of ND virus are capable of causing 100% mortality in unprotected flocks. Outbreaks of ND are unpredictable and discourage villagers from paying proper attention to the husbandry and welfare of their chickens. The importance of ND is indicated by the fact that ND has a local name in many countries, for example, in the Western Region of Ghana it is known as *Konoku, Twase Obgo* in the Greater Accra Region, *Adza* in Volta Region and *Nkoko Yare* in the Twi language. In Mozambique, ND is known as *Muzungo* in the Shangaan language, *Mbendeni* in Xitswa, *Ete-éma* in Macua and *Chigubo-gubo* in Shona. In much of Asia, ND is known as Ranikhet disease.

There are many constraints to village chicken production (Sonaiya et al. 1999) including a range of bacterial and other viral diseases, internal and external parasites (Permin and Hansen 1998), poor nutrition and predation. However, in areas where ND is endemic, ND control through vaccination is generally a very cost-effective intervention and given a high priority by farmers. Village chicken farmers are disheartened by the loss of large numbers of their birds to ND outbreaks that often occur on an annual basis. Once the dramatic losses caused by ND can be controlled, farmers will be more receptive to other messages concerning improved poultry husbandry.



This manual aims to present information that will enable veterinary departments and development agencies to implement a sustainable ND control program. Topics discussed include the characteristics of ND, collection and submission of samples for the diagnosis of ND, ND control measures emphasising vaccination with thermostable vaccines, gender and ethnoveterinary aspects of ND control and the development of an extension program for ND control. It is hoped that the approaches outlined may serve as a guide for the development of control packages for other major constraints to village chicken production. NEWCASTLE DISEASE

| Feature | Village Chickens | Commercial Chickens |
|----------------------|--|--|
| Labour inputs | Minimal | Considerable |
| Housing | Trees; chicken houses of local material; inexpensive | Chicken unit using conventional materials; expensive |
| Nutrition | Scavenging feed resource base, leftover food, cereals, no supplements; inexpensive | Balanced commercial ration; expensive |
| Water | Well water, used water, natural sources | Clean water supply essential |
| Production | Low; could improve with better nutrition, disease control and shelter from predators | High; but require a high level of inputs |
| Meat quality | Little fat; pleasant flavour; preferred texture | More fat; less flavour; poorer texture |
| Adaptability | Good: good flight skills, more likely to escape predators, can scavenge for own food | Limited: poor flight skills, easily caught by predators, less skilled at scavenging |
| Veterinary inputs | None, occasional vaccinations | Control of many viral, bacterial and parasitic diseases essential for efficient production |
| Environmental impact | Minimal: can be positive through provision of organic fertiliser and pest control | Negative: intensive production of cereals for rations; occasional improper use of antibiotics, excess ammonia production. |
| Farming system | Complex: integrated farming system involving extensive crop and livestock production | Usually single enterprise, intensive |
| Genetic diversity | Extensive | Limited |

 Table 1: Comparison of village and commercial chickens.

Table 2: Comparison of village and commercial chicken flocks.

| Criteria | Village flocks | Commercial flocks |
|------------|------------------------------|-----------------------|
| Flock size | Small | Large |
| Age | Mixed age | Single age |
| Housing | Trees, simple chicken houses | Large chicken units |
| Source | Natural incubation | Artificial incubation |



3

Characteristics of Newcastle disease

Newcastle disease (ND) is caused by a paramyxovirus which mainly affects poultry. Chickens are the most susceptible host. The incubation period varies with the strain of virus, and is generally 4 to 5 days (range 2 to 15 days). The virus is readily inactivated by formalin, alcohol, merthiolate, lipid solvents, lysol and ultraviolet light (Bratt and Clavell 1972). ND virus may persist in undispersed chicken faeces for more than six months (Alexander et al. 1985) but under village conditions the virus is unlikely to survive outside a host for more than one month. Vaccination is a routine practice for the prevention and control of the disease. However, it is difficult to transport and maintain conventional thermolabile vaccines in ambient temperatures ranging from 24°C to 36°C.

3.1 The clinical signs of ND

The clinical signs of ND vary considerably according to the virulence and tropism of the ND virus involved, the species of bird, the age of host, the immune status of the host and environmental conditions. As a result, none may be regarded as a specific sign of ND.

- Chickens infected with virulent ND virus strains may die without showing any signs of illness.
- The chicken fluffs its feathers and appears to 'have its coat dragging on the ground' (Figure 1).
- Lethargy and inappetance.
- Respiratory signs such as mild rales and snick can be detected by careful observation.
- Severe respiratory distress and gasping.
- Swelling of the head and neck.
- Greenish diarrhoea.
- Marked decrease in egg production. Sometimes deformed eggs may be produced.



Figure 1: Farmers in many parts of the world observe that a chicken with ND 'has its coat dragging on the ground.'



Figure 2: Torticollis is generally seen in chickens only when ND is at an advanced stage.

- Nervous signs of tremor, torticollis, convulsions and paralysis of wings and legs will not be seen until the disease is advanced (Figure 2).
- Mortality may be very high, often reaching 50% to 100% (Figure 3).



Figure 3: When mortality of 50% to 100% is observed in a flock of chickens, ND virus is almost always the causative agent.

• Other domestic poultry such as turkeys and pigeons may also be affected. Normally ducks are resistant to the disease but on occasions, ducklings may be affected.

3.2 Post-mortem findings

Post-mortem findings are characteristic but not definitive. ND can be suspected if the following lesions are encountered, particularly in combination (and when the flock history is also consistent with an ND outbreak):

- · congestion and mucous exudate in the trachea;
- congestion of the lungs (heavier than normal; lungs sink in water/formalin);
- · haemorrhages of the mucosa of the proventriculus;

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- haemorrhagic and necrotic ulceration of lymphoid patches of the intestine, caecal tonsils and bursa of Fabricius;
- congested ovarian follicles in chickens in lay.

3.3 ND virus classification

The ND virus can be classified into five pathotypes based on the clinical signs induced in infected chickens (Beard and Hanson 1981):

- i) viscerotropic velogenic (VV) high mortality with intestinal lesions,
- ii) neurotropic velogenic (NV) high mortality following nervous signs,
- iii) mesogenic low mortality, respiratory and nervous signs, reduced egg production,
- iv) lentogenic mild or inapparent respiratory infections, deaths confined to young chickens,
- v) asymptomatic enteric inapparent intestinal infection. (apathogenic)

3.4 Epidemiology of ND

3.4.1 Route of infection

ND virus can infect through the respiratory tract, the ocular mucous membranes, and the digestive tract, although this usually requires very high doses of virus depending on the virulence of the strain. The virus is shed from the respiratory tract and in the faeces. Most strains of ND virus are heat-labile and do not persist for long periods in the environment (or in diagnostic samples). A few strains are heat-tolerant, and these are mainly the avirulent strains that seem to favour oral-faecal spread.

3.4.2 ND in commercial flocks

In large commercial poultry units, the virus enters flocks through some break in biological security (on food, people, eggs, vehicles), by the introduction of infected birds in multi-age farms, or by aerosol (in the air) from an adjoining property. Once a few birds are infected, spread within the flock will be mainly by aerosol. Large flocks will produce copious quantities of aerosol virus, which can spread with movements of air to other flocks. Vaccines contaminated with virulent ND virus have also initiated outbreaks within flocks. It is generally accepted that the virus is not transmitted through eggs (vertical transmission); the exception may be the apathogenic strains as they do not cause the death of embryos.

3.4.3 ND in village flocks

Epizootic ND. Few studies have been done in village flocks. Outbreaks of epizootic disease come readily to notice and are described in the literature. The usual source of virus is an infected chicken, and spread is usually attributed to the movement of chickens through chicken markets and traders. A chicken incubating ND can introduce the virus to an isolated, fully susceptible flock, resulting in up to 100% mortality.

Endemic ND. An endemic form of ND which causes only occasional deaths is recognised in village chickens. The number of deaths is relatively low and does not attract official attention. The affected flocks usually result from breeding birds that have survived an outbreak. Many birds are immune and the virus passes from susceptible bird to susceptible bird. This endemic form will often contribute to mortalities among young birds. Eventually there are enough susceptible birds to sustain an explosive spread of virus with numerous deaths. Studies with computer models indicate that a population of 1,000 birds is sufficient to maintain endemic virus. Such a population could be a large village, or several adjoining small villages.

Seasonality of ND outbreaks. Human activity influences the occurrence of ND. In Asia, when seed rice is required for the seed beds in paddy rice fields, chickens are sold to raise the funds to purchase seed. Increased turnover in the chicken markets leads to outbreaks of ND that have in the past been attributed to seasonal weather conditions. In Uganda, ND is reported during the dry season. This is probably not related to weather, but to the fact that farmers with no immediate tasks visit relatives and take chickens as gifts. In many areas the villagers recognise the season when ND will occur, or they recognise the early cases, and they dispose of their chickens by sale, thus initiating or sustaining outbreaks. For each rural area it will be necessary to establish the seasonal pattern of ND, and if possible to deduce the reasons for these patterns.



3.4.4 Impact of vaccination

Vaccines will alter the epidemiology of ND to some extent since they will prevent disease, but not infection. Vaccinated birds exposed to virulent virus will develop no clinical signs. However, some replication of the infecting virus will occur and birds will excrete virulent ND virus. This will probably not be excreted in quantities as large as those excreted by susceptible birds, but there will be sufficient virus to infect other chickens.



Figure 4: Newcastle disease can be transmitted from one village to another via people, vehicles, animals, baskets, hoes, cages and infected produce (egg shells, feathers, bones, intestines, etc.).

4

Collection and submission of samples for the diagnosis of Newcastle disease

The diagnosis of ND is important for several reasons. ND is a notifiable transboundary animal disease and countries are to inform the *Office International des Epizooties* (OIE) when an outbreak occurs. Confirmed outbreaks help national authorities to better understand the epidemiology of ND in their countries and to develop appropriate control strategies. Once ND control activities are underway, it is useful if the cause(s) of mortality among vaccinated birds can be diagnosed. Vaccination against ND cannot provide protection in 100% of birds and this message must be clearly understood by all involved. Also, it is important to diagnose other diseases that will become more apparent (and consequently, more important) once chicken numbers increase as a result of the control of ND.

4.1 Tissue samples

Since virulent ND virus strains are normally thermolabile, it is important to send samples properly packaged with icepacks. Wherever possible, please try to observe the following conditions:

- Fresh samples. Samples of spleen, lung and the entire head should be wrapped in plastic and placed into a coolbox with ice or icepacks.
- Where it is not possible to keep the samples cold or when it is not certain that samples will arrive at the laboratory within 24 hours. Samples of spleen, lung, entire head (or brain) and long bones should be conserved in 50% glycerine (glycerol) in saline and kept as cold as possible during dispatch.

The coolbox containing the samples should be clearly identified and accompanied by the following information:

- the name and address of the person sending the samples;
- the date and location where the samples were collected;



- case details age, sex, breed, vaccination and treatment history, clinical signs, mortality and description of the outbreak; and
- differential diagnosis.

Central laboratories will usually have submission forms to record this information.

A general guide to the post-mortem examination of domestic fowl is given in Appendix 1.

4.2 Serum samples

The reliability of any serological test depends to a large extent on the quality of the samples submitted. Haemolysed or contaminated samples will often give unreliable results. Poor quality samples will give poor quality results, and the birds will need to be re-tested.

4.2.1 Blood collection technique

Blood from domestic chickens is usually collected from a wing vein. Some workers prefer to use a scalpel blade to nick the wing vein and then collect the blood into a tube. This method is quick but blood collected in this manner is more likely to become contaminated. In addition, farmers often object to seeing their birds stained with blood and may not allow them to be bled again. This will cause problems in situations where repeat bleeding of birds is necessary. A full description of a wing vein collection technique using a syringe and needle is given in Appendix 2. This technique, once mastered, causes minimal difficulties in the field.

A separate needle should be used for each animal to avoid the risk of mechanically transmitting infectious agents from one animal to another, and/or the transfer of antibodies from one sample to the next.

Paired samples must be collected from the same bird 2 or 3 weeks apart in order to monitor the response to vaccination. Therefore, a means of identifying individual animals is required. Conventional methods such as numbered wing tags should be used when available. If not available, then individual tattoos or physical markings need to be recorded to permit the identification of specific animals.

Contamination of the container and stopper should be avoided. Blood and faecal material should be removed prior to dispatch to reduce the risk of contamination of laboratory staff handling the specimens.

4.2.2 Labelling of samples

Samples must be labelled serially (e.g., from 1 to 30) with a waterproof pen, preferably on an adhesive label. Do not write on the cap of the tube as it may be removed during testing. Do not label containers with water-soluble ink. It smudges when wet and may rub off if samples are chilled or frozen. Draw a line under numbers that can be misread if inverted, for example <u>18</u> and <u>81</u>. If samples are to be stored, record the date of collection including the year.

4.2.3 Avoiding haemolysis of samples

Haemolysis occurs as a result of poor collection technique, contaminated equipment or poor handling of the sample once it is collected.

Common causes of haemolysis include:

- slow flow from the needle, due to obstruction of the needle, or failure to insert it directly into the vein;
- heating of samples, usually in cars or after prolonged exposure to direct sunlight during collection;
- freezing;
- contamination of the sample by water;
- contamination by faecal and other material;
- forcible expulsion of blood through a needle;
- · bacterial contamination during collection; and
- use of non-sterile containers for collection or storage.

Haemolysis can be reduced by using clean, dry, sterile needles and avoiding contamination by water.

4.2.4 Storage of sera prior to dispatch

- Blood or serum samples should not be submitted in jars, non-sterile containers or syringes with needles attached.
- Samples should be allowed to clot before transporting them any distance. The samples should be held in a warm place until the clot retracts. Clots may not retract readily in cold weather or if samples are chilled too soon after collection.



- Once the clot has retracted, blood samples must be held chilled to reduce contamination, haemolysis and autolysis.
- If samples cannot be rapidly delivered to the laboratory and delays are likely to occur between collection and testing, it is preferable to separate the serum into a 5 mL or 1.8 mL sterile, screw-capped plastic container and submit the serum sample only. Transfer the original label or re-label.
- Blood samples for serology must not be frozen until the serum has been separated from the clot. Serum samples can be submitted frozen, provided that there are no blood cells present in the sample.

4.3 Dispatch of samples

N.B. Before sending samples, please ensure that:

- the samples are securely packaged;
- the label on the coolbox or container has the following:

URGENT

To: The name and address of the veterinarian responsible for your closest central veterinary laboratory.

From: The name and address of the person sending the samples;

- the samples are accompanied by an information sheet; and
- the relevant person in the central veterinary laboratory is informed when the package is expected to arrive and by what means of transport.

4.4 Communication of results

Always ensure that results are communicated to farmers in writing and that a verbal explanation of the results is also given.



5

Control of Newcastle disease

Vaccination is the only effective way of controlling ND (Figure 5). However, vaccines currently in use are mainly of benefit to commercial poultry producers whose chickens are kept in large, single-age, confined flocks. Manufacturers produce heat-labile ND vaccines in multi-dose vials, often containing 1,000 or 2,500 doses, which must be kept cold (within a 'cold chain') from manufacture until administration to the chickens. In contrast, village chickens are raised in small, multi-age, free-range flocks and large multi-dose vials of vaccine are inappropriate. The cold chain is difficult to maintain under village conditions and purchase of commercial vaccines is a drain on foreign exchange.

The Australian Centre for International Agricultural Research has supported projects leading to the production of vaccines suitable for use in village chickens. These have been selected for thermostability, so that a continuous cold chain is not necessary. If required, they can be given on some types of food (not all foods are suitable, see Section 5.1.3) to chickens that are not easily caught. The first of these vaccines, NDV4-HR, was successfully tested in Asia and Africa. It became a commercial vaccine, with the seed virus under commercial ownership. Although heat-resistant, it now comes in a large dose commercial format and costs foreign exchange.

The second thermostable ND vaccine is I-2, very similar to NDV4-HR but free of commercial ownership. Seed cultures can be made available without cost to countries that wish to test or produce their own vaccines (via Professor Peter Spradbrow, see Appendix 10). The simple techniques required for producing and testing the vaccine can be learnt at short workshops. In Asia it has been adopted as the official vaccine for village use in Vietnam, and it is being exported from that country. The I-2 ND vaccine is currently being tested in several African countries.



To date, all current ND vaccine strains will protect most birds with a haemagglutination inhibition serological antibody titre of log₂ 3 against all field strains. Polyvalent antisera do not detect antigenic differences between strains of ND virus.

5.1 Vaccination

ND vaccines currently in use in many countries include: La Sota (live vaccine, thermolabile); Hitchner B1 (live vaccine, thermolabile), ITA-NEW/NEW COVER (inactivated vaccine, thermostable); NDV4-HR (live vaccine, thermostable); and I-2 (live vaccine, thermostable) (see Table 3). The ND Clone LZ.58 (Nobilis ND Inkukhu) vaccine is a live, partially thermostable vaccine recently released by Intervet South Africa Pty. Ltd. (see Section 6.9). The first three vaccines must be kept in the refrigerator between 4 and 8°C and never frozen. Vaccines should not be used after the expiry date. Once a vial of thermolabile, live vaccine has been opened it should be used immediately and not stored for use the following day.



Figure 5: Vaccination against Newcastle disease is the only efficient way to control the disease in most countries.

During vaccination campaigns, vaccine should be stored in a coolbox or wrapped in a damp cloth, and not exposed to sunlight. The NDV4-HR and I-2 vaccines are thermostable (more details are given in a later section) but it is still important to keep them away from sunlight and as cool as possible ensuring that their activity outside the cold chain is as long as possible.

The vaccines HB1, La Sota, ND Clone LZ.58, NDV4-HR and I-2 can be administered via eye drop or drinking water. The NDV4-HR and I-2 vaccines can also be administered orally after mixing with certain foods (care must be taken to ensure that the chosen food does not contain agents that can inactivate the vaccine virus; see Section 5.1.3). The most efficient route of administration is via eye drop.

 Table 3: Comparison of Newcastle disease vaccines.

| | Live | Inactivated |
|----|--|--|
| 1. | Contain a small amount of living virus which replicates; cheaper | Must contain a large amount of inactivated virus; more expensive |
| 2. | Can be administered by many routes: eye drop, intranasal, spray, drinking water, oral, injection | Must be injected |
| 3. | Stimulate all forms of immunity | Stimulate only antibody-based immunity |
| 4. | Duration of immunity varies according to route of administration, usually not more than 4 months | Duration of immunity approximately 6 months |
| 5. | Difficult to store (except thermostable live vaccines, e.g. 1-2) | Less difficult to store |
| 6. | Not dangerous to vaccinator | Dangerous to vaccinator on accidental injection |

5.1.1 Eye drop administration

Correct dilution of the vaccine is critical. If eye-droppers are being used, they should be calibrated beforehand (see Appendix 3). In the absence of suitable eyedroppers, it is also possible to use the tip of a feather or a syringe to administer the drop. However, these two options should be seen as last resorts as they are inaccurate and cause considerable wastage of vaccine. Most live ND vaccines require re-vaccination at 3 to 4 monthly intervals.

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Eye drop administration provides good protection because after administration, the vaccine passes to the Harderian gland just behind the eye. The Harderian gland in chickens is a key organ in the development of the immune response.



Figure 6: Eye drop administration. When using an eye-dropper, hold it in a vertical position. Eye-droppers are calibrated according to the size of the drop that forms when the dropper is held in a vertical position.

5.1.2 Administration of the vaccine via drinking water is easier, but provokes a lower level of immunity than eye drop administration and requires more frequent application. The vaccine should be given twice, two to three weeks apart initially, with re-vaccination occurring at least every three months.

It is important to:

- remove drinking water from the chickens for one to two hours before the administration of the vaccine;
- mix the vaccine with a volume of water that the chickens will be able to drink during one hour, usually 5 to 7 mL of water per bird; and
- always use fresh and clean water.

In rural areas, it is best to give the drinking water in the morning just as the chickens are released from their chicken house. In areas with abundant surface water, chickens find their own source of drinking water and vaccination via water is not appropriate.

Do NOT:

- use metal water receptacles,
- use disinfectants to clean water receptacles as they will inactivate the vaccine virus,
- use treated tap water, (If you only have access to treated tap water, it is advisable to (i) let the treated tap water stand over night allowing the chlorine to dissipate, or (ii) add one teaspoon of powdered milk per 10 litres of water to neutralise the effects of the chlorine.)
- place water receptacles containing vaccine directly in sunlight or in hot areas,
- allow other animals access to the vaccine. It should be restricted to chickens.

5.1.3 Administration via feed

Oral vaccination of chickens with thermostable vaccines (i.e. NDV4-HR and I-2) has been successful in some developing countries. Good veterinary services, local availability of suitable grains and recovery of virus from the grain are important considerations for successful oral vaccination. One problem with food based ND vaccination is the low recovery of virus from some grains (especially maize), a consequence of either binding or inactivation. Therefore, the food used in any vaccination campaign should be recommended by the Veterinary Services Department. Seven to 10 grams of food per bird should be well mixed with the corresponding number of doses of appropriately diluted vaccine. With most grains, 1 mL of fluid will efficiently moisten 10 grams of grain. The treated food is best given in the morning as the birds are leaving the roost. The vaccine should be given twice, 2–3 weeks apart initially, with re-vaccination occurring at least every 2–3 months.

5.1.4 Administration via injection

Inactivated ND vaccines are administered only by intramuscular or subcutaneous injection (in the breast or the leg). The vaccine should be allowed to reach ambient temperature (approximately 28°C) and the contents well shaken prior to use. If stored in a cool, dark location, this vaccine may retain its activity for one to two weeks outside a refrigerator.

| Dose | Age |
|--------|--------------------|
| 0.2 ml | Day-old to 3 weeks |
| 0.3 ml | 3 to 5 weeks |
| 0.5 ml | 5 weeks and older |

Inactivated vaccines are more effective in chickens that have previously received a living vaccine. Re-vaccination is usually done every 6 months.

Accidental injection of this vaccine into the vaccinator can cause a serious localised reaction. Expert medical advice should be sought at once, and the doctor informed that the vaccine was an oil emulsion.

5.2 Timing of vaccinations

After administration of the vaccine, immunity does not develop immediately. One to two weeks is required for the full immune response to occur. Chickens should be vaccinated at least one month before an outbreak is likely to occur. Ask local village poultry farmers when ND outbreaks are most common and plan vaccination campaigns on a collaborative basis.

Immunity will diminish if chickens are not revaccinated. With the eye drop method of administration, chickens are best vaccinated at least three times a year. If oral routes of administration are used, chickens should be given a booster dose two to four weeks after the primary vaccination, with re-vaccination at three monthly intervals. Vaccinating village chicken poultry flocks at three to four monthly intervals will also provide protection for newly hatched chicks.

Inactivated and living ND vaccines contain a ND virus that is antigenically similar to the disease producing strains. Inactivated ND vaccine is usually administered every six months. In areas where outbreaks generally occur once a year, the vaccine may be strategically administered before the normal seasonal outbreaks are due to commence.

If the mode of administration requires the handling of individual birds, time vaccination campaigns to coincide with school holidays or weekends in order to enlist the services of children. The skill and energy of children can be invaluable, especially in areas where chickens roost in trees.

5.3 Benefit:cost considerations

When working with village chickens, it is essential that benefit:cost analyses of all interventions be done so that any ND control strategies are cost-effective. The main costs associated with the control of the disease are the purchase of the vaccine, transport and handling costs. The less frequently that chickens have to be vaccinated, the more cost-efficient the strategy. However, long revaccination intervals leave newly hatched chicks susceptible and where endemic ND is present, chick mortality will increase. There is still much to be studied in this area, but make sure that you have an idea of the pattern of ND outbreaks in each area so that you can start vaccinating before an outbreak occurs. Whenever possible, use eye drop administration.

Farmers must be informed of the different administration regimes and the frequency of application of the vaccine required to ensure adequate levels of protection with each administration route.

Eye drop administration of the NDV4-HR and I-2 vaccines promotes higher levels of immunity than oral administration. Consequently, with the eye drop method it is not necessary to administer the vaccine as frequently in order to maintain adequate levels of protection.

The main advantages of the thermostable, live vaccines are:

- thermostability they are able to reach sites beyond the cold chain in a viable state;
- ease of administration they can be applied by farmers at the village level; and
- they will spread from vaccinated to non-vaccinated chickens in close contact.

The cost of vaccine distribution and administration will be greatly reduced if Veterinary Services staff are not involved at the household level. The involvement of community vaccinators or community livestock workers (CLWs) in ND vaccination programs can greatly reduce costs and increase coverage (Appendix 4). In most cases the community vaccinator will receive payment from farmers to cover the cost of the vaccine and the labour of the vaccinator. As the community vaccinator lives locally, the labour costs paid by the farmers are most likely to stay within their community. If the vaccine is made within the country, then the majority of the costs associated with the production of the vaccine will stay within

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the country. This type of approach will encourage the development of sustainable livelihoods both within rural communities and supporting national services.

In situations where small-scale farmers are to pay for ND vaccine, there will usually be a direct relationship between the price of the vaccine and the number of farmers who can afford to pay for it. As shown in Figure 7, a small increase in the price of the vaccine (A) will result in a proportionally larger decrease in the number of farmers purchasing it (B). A risk assessment must be done to determine the most economically sustainable ND control strategy. The assessment will investigate the implications associated with the use of a range of ND vaccines to detail, for example

- the risks associated with the use of higher priced ND vaccines that will either increase the cost of ND control activities or reduce coverage of birds, thus leaving chickens not protected against ND,
- the risks associated with the use of lower priced ND vaccines that are not made using specific pathogen free eggs,
- the availability of foreign exchange for the purchase of imported vaccines.



Figure 7: The impact of vaccine price on demand by farmers for the vaccine and consequently vaccination coverage.

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5.4 Development of ND vaccination campaigns

In most cases, farmers will be expected to pay for the ND vaccine and so it is critical that the first vaccination campaign is a success. Most farmers will not grant you a second chance. The best way of ensuring good results is to prepare thoroughly before commencing with vaccinations in the field and to have the will and the resources to ensure that subsequent campaigns will be implemented at the recommended intervals.

5.4.1 Situation analysis

- Awareness of officials, veterinarians and extension workers. Is the control of ND in village chickens seen as a priority by decision makers? What information do they need to help them understand the importance of vaccinating regularly against ND? Will existing government policies (on cost recovery, for instance) facilitate the development of a sustainable ND control program?
- Farmer awareness (and priorities). Is ND a priority for farmers in the area where you plan to vaccinate? Do they know that a vaccine against ND exists?
- Village chicken population. Obtain an estimate of village chicken numbers and, if farmers are to pay for the vaccine, make an estimate of the percentage of farmers likely to do so. This will enable you to order an appropriate quantity of vaccine.
- **Training requirements**. Even if you plan to use a thermostable ND vaccine, it will not compensate for poorly trained personnel. For good results, make sure that all participants in the vaccination campaign have received appropriate training. Training will vary according to the function of the individual:
 - veterinary services staff
 - extension staff
 - community livestock workers or community vaccinators
- Seasonality of ND outbreaks. When are ND outbreaks most likely to occur? If there is thought to be a seasonal pattern to outbreaks, ensure that the campaign starts at least one month before the outbreaks are expected (see Participatory Epidemiology in Section 9.3.1).
- Agricultural and climatic calendar. Plan campaigns to coincide with times of the year when farmers are not very busy in their fields and access to the area is possible.

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- Gender analysis. The campaigns will meet with better success if arrangements are made with the person in the family who owns and cares for the chickens. Suggestions on how this information can be obtained may be found in Section 7.0.
- **Cost-recovery options**. The majority of farmers are willing to pay for a product if they believe they will get a good return on their investment. Discuss payment options with farmers and always give them advance notice so that they can arrange funds prior to the campaign.
- **Inputs**. Always make sure that you know where you can get the supplies necessary for the vaccination campaign and that the material is in stock:
 - vaccine, of appropriate quality and quantity;
 - eye-droppers (see Appendix 3); and
 - field allowances, etc. Even if you plan to work with CLWs, you will need to train and supervise them. These activities require funds and these funds must be confirmed before you begin your activities in the field.

5.4.2 Preparatory phase

- Appropriate extension materials. Prepare, pretest and duplicate the necessary extension material.
- **Training of personnel**. Train personnel well in advance of the campaign. They need time to go back to their respective areas to raise farmer awareness, collect information and make their own preparations.
- Timing of campaign. Decide in consultation with staff, CLWs and farmers. Consider weather conditions, the farmers' annual work plan and the pattern of ND outbreaks.
- Extension activities. Start at least one month prior to the campaign.
- Vaccine administration options. Use eye drop administration whenever possible. However, in certain circumstances farmers may opt for oral administration (see Section 5.1). Consider whether the vaccinator is to travel to individual houses or if farmers bring their birds to pre-arranged points.
- **Inputs**. Vaccine, eye-droppers and syringes, per diems, transport, registration books, cool boxes or baskets and cloth must be procured.

5.4.3 Recommendations

- Commence campaigns at least one month prior to the season when ND outbreaks are more common.
- Postpone the vaccination campaign if is suspected that an outbreak of ND is in progress.
- Vaccinate healthy chickens only.
- Always inform farmers of the need to revaccinate their birds.
- Campaigns are best held during the weekends or school holidays.
- Cost-recovery, at least partial, is essential.
- Never promise protection of 100% of chickens.
- · Emphasise that the vaccine protects against ND only.

5.4.4 Implementation

On the first day of the vaccination campaign, you will have:

- trained teams;
- vaccine and other inputs available;
- decided, in coordination with farmers, on the site of vaccination:
 - house-to-house visits; or
 - central vaccination points;
- participating farmers registered;
- a way of identifying vaccinated chickens;
- a system in place for the vaccinator to register the number of birds vaccinated and payment received.

5.4.5 Monitoring and Evaluation

This is an essential part of a ND control program.

• **Timing and frequency.** The timing and frequency of monitoring visits will vary according to the position of the person(s) involved (e.g. CLW or Livestock Officer) and the type of monitoring being undertaken. Monitoring of activities should occur at regular intervals to enable timely adjustments to be made.

- One week to one month after vaccination, CLW confirms that birds are healthy following vaccination; and
- Three months after vaccination is an ideal time to monitor chicken numbers, farmer attitudes and to prepare for the following campaign if vaccination is being done every four months via eye drop.
- **Participatory process**. In theory all stakeholders should participate in the monitoring process. Stakeholders may include community representatives (male and female), government officials, project staff and, where relevant, consultants.
- Indicators. All stakeholders should have a say in defining the indicators of success. Possible indicators may be:

Short-term changes in:

- household chicken numbers;
- the number and types of people participating in vaccination campaigns;
- the level of community involvement in campaigns;
- the economics of households;
- the number of chickens sold or traded; and
- home consumption of chickens and eggs.

Long term changes in:

- the number and diversity of livestock species raised;
- the demography of households; and
- primary school enrolment statistics

Ultimately, the question which needs to be answered is whether the control of ND has assisted in poverty alleviation and improved food security.

• Identification of other constraints. Front line extension staff should be encouraged to identify other constraints that limit poultry production in order to work with farmers in a process of continuous improvement.

5.5 Other control strategies

- Avoid the introduction of new birds to flocks during the periods of the year when ND occurs more frequently.
- Do not return from market with chickens that have failed to sell. Instead, arrange to keep them in another place.

- Avoid contact with people, cars and animals that have been in contact with the virus and other parts of infected chickens (e.g. eggs, feathers, etc.). Dogs and cats can also transmit the virus if they have access to chickens killed by ND.
- Minimise contact between chickens and other poultry, such as ducks, pigeons, turkeys and guinea fowl.
- Good housing can reduce disease transmission. An elevated chicken house that
 is well-ventilated allows faeces to fall through to the ground and so minimises
 contact with various infectious agents (Figure 8). Keep chickens and chicks
 away from the base of the chicken house where the faeces have accumulated or
 clean the area regularly. Encourage the use of local remedies to control
 ectoparasites (e.g. fleas and mites) in the houses when commercial insecticides
 are not available.



Figure 8: Elevated chicken houses with a slat floor and metal guards to prevent the entry of rodents and snakes will improve chicken health and survival.

- House hens with young chicks in a clean, safe chicken house.
- Provide some supplementary feed, such as maize bran, ground grains, green leaves, ground sea shells, insects, insect larvae and worms. Good nutrition will give chickens a better chance of combating infections. Supplementary feeding is especially important for chicks, and a creep feeder can be made from local materials to ensure that chicks are able to receive food without greatly increasing the amount of food given to the household poultry flock (Figure 9). A creep feeder also provides chicks with shelter from airborne predators.
- Always provide water; fresh, clean water is best when available.



Figure 9: A creep feeder that will improve chick nutrition can be made from locally available materials.

5.6 Control measures during an outbreak

- Isolate all sick chickens.
- Slaughter chickens that are very ill. Do not transport chickens that are ill or dead to other areas that are free of the disease.
- Bury or burn all dead chickens. If, for any reason, it is not possible to do this, any part of the chicken that has not been used should be buried or burned.
- Do not vaccinate chickens that are showing signs of illness.
- Once an ND outbreak has commenced in a village, it is best not to vaccinate as it is impossible to identify birds that are incubating the disease but not yet showing signs of illness. Farmers will often associate the vaccine with the death of chickens that are vaccinated in the face of an outbreak.
- Advise farmers to wait for at least one month after the last mortality before re-stocking.
- Advise farmers to contact the Veterinary Services Officer, Extension Worker or Community Livestock Worker in their area when they notice any signs of illness.



6

Introduction to live, thermostable Newcastle disease vaccines

A thermostable vaccine enables distributors and users to reduce the problems associated with inadequate cold chains in the field. It is essential that users understand that a thermostable vaccine must still be treated with some of the respect due to a conventional biological product, that is, you cannot expose the vaccine to sunlight and frequent shifts in temperature and still expect it to remain active.

6.1 The NDV4-HR vaccine

The heat resistant V4 (NDV4-HR) vaccine against ND has yielded encouraging results in Cameroon (Bell et al. 1995), Ghana (Amakye-Anim et al. 1998), South Africa (Magalo, pers. comm.), Tanzania (Spradbrow and Foster 1997), Zambia (Alders et al. 1994) and many countries in Southeast Asia (Spradbrow 1993–94).

The NDV4-HR vaccine is a living vaccine with the following characteristics:

- it is thermostable, retaining its activity for 12 weeks at a temperature of 28°C in freeze-dried form (Ideris et al. 1987);
- it can be administered via eye drop (intraocular), nose drop (intranasal), oral drench, or drinking water; mixed with certain feeds or by injection (Spradbrow 1993–94, Anon. 1991);
- its ease of administration makes it suitable for use by village farmers;
- the vaccine strain can be transmitted by contact from vaccinated to non-vaccinated birds (Alders et al. 1994, Spradbrow 1993–94);
- it is avirulent and can be safely administered to chickens of any age from day-old to adult (Spradbrow 1993–94, Anon. 1991);
- its biological safety is superior to that of other living ND vaccine strains such as B1 or La Sota (Anon. 1991).


FAO recommends this vaccine for the control of Newcastle disease in village chickens in tropical countries and developing countries as a means of improving the food security of rural communities (FAO 1997).

This vaccine is available from:

- Fort Dodge Australia Pty. Ltd.
 23 Victoria Avenue
 Castle Hill, NSW 2154, Australia
 Contact: John Reeves, Sales and Market Manager
 Intensive Animal Industries
 Tel: +61-2-98992111
 Fax: +61-2-98992151
 Email: ReevesJ@fortdodge.com.au
- Malaysian Vaccines and Pharmaceuticals Sdn. Bhd. Malaysian Technology Development Corporation Sdn Berhad Lot 11182, Batu 20 Jalan Puchong Kajang Pulau Meranti, 47100 Puchong Selangor Darul Ehsan, Malaysia Tel: +60-3-5715701/2 Fax: +60-3-5712557/5717623 E-mail: <u>halim@mtdc.com.my, drmazlan@mtdc.com.my, mvp@tm.net.my</u> Website: http://www.mvp.com.my/

6.2 The ND I-2 vaccine

The Australian Centre for International Agricultural Research (ACIAR) commissioned workers at the Virus Laboratory in the University of Queensland to produce a seed virus similar to NDV4-HR that could be made available without cost to laboratories in developing countries (Bensink and Spradbrow 1999). Forty-five isolates of avirulent ND virus were examined for antigenicity, safety and ability to spread. The most promising of these isolates were checked for their thermostability and the more resistant isolates selected for enhanced heat resistance. The result was strain I-2, which was amplified in eggs from a disease-free flock to form a master seed. The seed was tested for safety and for freedom from bacterial contamination.

Strain I-2 has undergone laboratory tests in several countries and has proved to be protective against local virulent strains of ND virus. In Vietnam it has been officially recognised as the ND vaccine for village chickens, after extensive laboratory and village trials (Tu et al. 1998). In Tanzania it has given protection for at least two months after vaccination (Wambura et al. 2000). Field records in Mozambique indicate that I-2 ND vaccine provides approximately 80% protection in the field in the face of an outbreak, when given every 4 months via eye drop. In one area where vaccination with I-2 vaccine was performed at 4-monthly intervals with assistance from VETAID (a British NGO), the chicken population in 134 households increased from an average of seven birds per family to 20 birds per family in a six-month period (Pagani 1999). During a 5 month field trial, where bird numbers where monitored on a 2-weekly basis, a 50% increase in bird numbers and increased home consumption of chickens was noted (Dias et al. 2001).

ND vaccine of acceptable standard can be produced from strain I-2 in central laboratories or in some cases, in regional laboratories in developing countries. The vaccine can be produced in eggs that are not specific-pathogen-free, but which come from a flock that is regularly screened for key poultry diseases (such as pullorum disease). It can be produced and stored in liquid form, and suitably diluted in a protective solution such as 1% gelatin (in which the vaccine will maintain its activity for at least 12 weeks at 22°C; Bensink and Spradbrow 1999) before use. The thermostable vaccine is then best administered via eye drop. The I-2 vaccine produced in Mozambique will retain its activity for 8 weeks at 28°C when in freeze-dried form and stored in the dark (Alders et al. 2001).

6.3 Storage and transport conditions for thermostable ND vaccines

If users have access to normal cold chain facilities, then by all means these should be used, even when dealing with a thermostable vaccine. Freeze-dried vaccine stored at $4-8^{\circ}$ C will retain high titre for a longer period than that stored at ambient temperature. At $4-8^{\circ}$ C, the vaccine should maintain an adequate titre for at least one year.

When taking the vaccine to the field, place it in a cool box with ice or an ice pack. DO NOT FREEZE the vaccine (unless the instructions specifically indicate that the vaccine may be frozen). Freeze-dried vaccine packaged under vacuum rather than with nitrogen will lose the vacuum and gain moisture if the vial is frozen. The rubber cap on the vial contracts when frozen enabling moist air to enter the vial. When this occurs, the shelf-life of the vaccine is reduced.

These vaccines are thermostable, but attention to the conservation of the vaccine once removed from refrigeration will ensure optimal results:

- Always keep the vaccine away from sunlight.
- When transporting the vaccine in the field, wrap it in a damp cloth and carry it in a covered open-weave basket (Figure 10). This allows evaporative cooling which helps to keep the vaccine cool and the cover prevents contact with sunlight.



Figure 10: Thermometers can be used to check internal temperatures of possible vaccine transport containers to assist with selection in areas where cool boxes and ice packs are not an option. Covered open-weave baskets (A) with the vaccine wrapped in a damp cloth inside usually provide the coolest environment. Closed synthetic backpacks (B) and plastic bags (C) are not suitable. The temperatures shown were recorded after carrying the containers in the field for 20 minutes on a sunny 35°C day.

• Record the date the vaccine leaves the cold chain, it will remain effective for 2–3 months only.

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• Store the vaccine in a cool, dark location, for example, near the base of a clay water pot (Figure 11).



Figure 11: Once thermostable vaccines leave the cold chain, they are best stored in a cool and dark location. Near the base of clay drinking pots kept in a dark place by many rural families is ideal.

6.4 Administration of thermostable ND vaccines

Standard dose. As with other live ND vaccines such as La Sota, a minimum of 10^{6} EID₅₀/bird is required to produce an adequate level of protection. EID₅₀ (50% embryo infectious dose) is a laboratory measure of the content of living infectious virus in a vaccine. It has been demonstrated that birds that received a higher oral dose of the NDV4-HR vaccine generated a higher immune response when confined in cages with wire floors (Spradbrow et al. 1988). [The same report indicated that the dose responsiveness to oral vaccination was no longer apparent when groups of vaccinated chickens were housed together on litter. The explanation for this result was that the vaccine virus replicated and was excreted in the faeces and the birds were then re-infected by the virus in the environment.] This means that even though the thermostable vaccine can survive at ambient temperatures, attempts to improve its conservation will result in a slightly higher vaccine titre at the time of vaccination and consequently a higher and longer lasting immunity. This is particularly important when birds are not housed together at night.

Administration route. These vaccines can be administered via eye drop, drinking water, certain feeds and injection. Field trials in Mozambique indicated that almost all farmers preferred eye drop administration even though it required the capture of birds. In their opinion, eye drop administration of the vaccine produced a greater survival rate, had a lower frequency of administration and was easy. It is important to confirm that the eye-dropper to be used is made of virus-friendly plastic and that it is calibrated to ensure that one drop contains one dose (see Appendix 3). Calibration of the eye-dropper and administration of the eye drop to the bird is done with the dropper in a vertical position to make sure that drops of a uniform size are produced (see Figure 6).

Age of bird — the same dose is given to birds of all ages, from day-old chicks to adults.

Vaccination schedule. For eye drop administration, the vaccine should be administered once, with re-vaccination every 3–4 months. Via drinking water, the vaccine should initially be given on two occasions, two to three weeks apart, with re-vaccination occurring at least every three months.

6.5 Dilution and use of thermostable ND vaccines

These vaccines may be diluted using locally available potable water. It is recommended that the water is boiled and left to cool overnight in a non-metallic container before use.

Chlorinated tap water is unsuitable. If, however, this is the only water available, let the treated tap water stand overnight to allow the chlorine to dissipate.

Once the freeze-dried vaccine has been diluted, it is advisable to follow this simple rule for eye drop administration:

- Day $1 \Rightarrow 1$ drop per bird (i.e. first day of vaccination campaign)
- Day 2 ⇒ 2 drops per bird
- Day 3 \Rightarrow discard

6.6 Horizontal spread of thermostable ND vaccine virus

The thermostable live ND vaccines spread from vaccinated to unvaccinated birds when housed together (Alders et al. 1994, Bensink and Spradbrow 1999, Tu et al. 1998, Spradbrow 1993–94). The degree of spread under field conditions is less when birds roost in trees and horizontal transmission should not be seen as a reliable substitute for vaccinating village birds.

6.7 Safety issues

The avirulent live ND vaccines such as I-2 and NDV4-HR are unusual in that it is not possible to overdose with them. They are harmless to both bird and handler. Both the I-2 and NDV4-HR vaccines produce no evidence of clinical respiratory signs, weight loss, mortality in young chickens or drop in egg production after vaccination (Bensink and Spradbrow 1999, Heath et al. 1992). The safety performance of original V4 (avirulent) vaccine is superior to both the HB1 (lentogenic) and La Sota (mesogenic) vaccine strains (Table 4).

6.8 Genetic sequencing of thermostable ND vaccines

Genetic analysis indicates a relationship between the chemical structure of limited areas of the genome of strains of ND virus and the virulence of these strains. An area of apparent importance is the cleavage site of the fusion protein on the surface of the virus particle. Particular amino acid patterns around the cleavage site in virulent strains have become known as the virulence sequence. V4 and I-2 and other vaccines such as La Sota and HB1 lack the virulence sequence (see Appendix 5).

Table 4. Comparative safety of Newcastle disease vaccine strains(Heath et al.1992).

| Signs in vaccinated birds | Vaccine strain | | | |
|-----------------------------|----------------|----------------------------|--------------------------------|--|
| | V4 | HB1 | La Sota | |
| Sneeze test | Nil | Definite signs | Pronounced signs | |
| Respiratory disease | Nil | Clinical respiratory signs | Clinical respiratory signs | |
| Weight gain | No effect | Significant reduction | Highly significant suppression | |
| Mortality in young chickens | Nil | Yes | Yes | |
| Egg production drop | Nil | 5–10% | >10% | |

6.9 Partially thermostable Newcastle disease vaccine — Nobilis ND Inkukhu

Nobilis ND Inkukhu is not a thermostable vaccine in the true sense, it is a freeze-dried vaccine that in the freeze-dried form is stable for up to 7 days in temperatures not exceeding 30°C. At such temperatures, the infectivity titre remains stable for seven days.

Once reconstituted with a diluent, it should be treated as any standard freezedried ND vaccine. Furthermore, once removed from refrigeration for an extended period, it must be used within the seven day period and not returned to refrigeration for further storage. The expiry date printed on each vial is valid only when the vaccine is held constantly under refrigeration.

This product is distributed in South Africa by Intervet and is aimed primarily at the smallscale commercial farmer in outlying areas. It offers this type of farmer the ease of transporting the vaccine from the supplier (with refrigeration) to his/her farm (often without refrigeration) without the cold chain required for conventional freeze-dried vaccines.

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The vaccine strain is ND Clone LZ.58 originally marketed by Mycopharm in the Delvax range. The vaccine Delvax ND Clone LZ.58 has proven its efficacy in numerous poultry producing countries as a primer and booster vaccine against ND. It is a clone of the La Sota strain and will induce some post-vaccinal reactions in chickens. Post-vaccinal reactions occur during the first week following vaccination and include a mild snick and a very slight rise in mortality (less than 0.2%).



7

Gender aspects of village chicken production and the control of Newcastle disease

Gender is defined as the socially determined differences between women and men, as opposed to the word 'sex' which denotes physical differences. Gender differences are historically determined, culturally specific and dynamic. They define how women and men interact in a specific context, and what is considered appropriate for women and men to do, thus determining their respective development options and constraints (Gujit 1994).

In traditional village poultry production systems, we need to learn who does what and then help them do it better. Collecting gender disaggregated data helps us to determine how the tasks associated with village chicken production are divided within families. Extension messages can then be tailored for the target audience. It is well-known that direct communication with the person who actually does the work is more effective.

In development activities, it is important not to focus on women in isolation, but to understand women's roles and positions within the family and the wider community. In some cases, providing training exclusively for women is not necessarily the most efficient approach. For instance, the design of chicken houses can have a major influence on flock health, and in many countries it is the men who are responsible for building the houses. Consequently, if men are included in the relevant training sessions, the transfer of appropriate technologies may proceed with fewer difficulties. Nonetheless, it is important to stress that, in general, women should be targeted for training and that, where possible, the extension workers involved in rural poultry production should be women.



Outlining tasks associated with the production of village chickens according to age and gender helps to determine who in the family should be targeted when developing extension material associated with various aspects of poultry production. Table 5 shows the disaggregation of tasks associated with village poultry production formulated at a workshop in Cambodia.

 Table 5: Disaggregation by gender and age of tasks associated with village

 chicken production in Cambodia.

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| Task | Man | Woman | Воу | Girl |
|---|-----|------------|------------|------------|
| Feeding chickens | | × | × | × |
| Construction of chicken house | X | | | |
| Catching chickens | X | | × | |
| Who should be informed about the need to catch chickens | | X | | |
| Selling chickens | | × | | |
| Deciding when to sell chickens | X | X ? | | |
| Deciding whether to vaccinate chickens | X | × | | |
| Deciding when to eat chickens | X | × | | |
| Eating chicken | X | × | | |
| Eating eggs | | | 🗶 (rarely) | 🗶 (rarely) |

It is often a problem for women farmers in rural areas to attend extension meetings conducted by a male extension worker. In the event that female extension workers are not available, male extension workers can work with a female assistant. The assistant should be fluent in the local language and be able to explain the messages that the extension worker wishes to convey as well as facilitating discussion among the women farmers. Formal training in agriculture is not essential for the female assistant.

The first visit can be critical. Always talk with the village headman first and determine whether or not senior leaders (including senior women) would be interested in the project activities. For visits to individual households, it is advisable to time the visit so that discussions can be held with both husband and wife. Once the objectives of the project have been discussed, the couple can decide whether or not they wish to participate. If they wish to participate, they must then decide whether one or both will join in the project activities. Women's associations within religious groups may also wish to participate in project activities.

It is crucial to remember that the care of poultry is just one of the tasks that rural women undertake. Project activities need to fit into their already busy work schedule. Wherever possible, project meetings should take place within or near to women's work places to reduce the time taken travelling to meetings. Childcare will also be less difficult. Attendance at meetings is likely to be higher if they are held at a time that most women agree is preferable. Shorter, more frequent meetings in areas closer to their homes may be the best approach. Project staff should also take into account the fluctuations in workload throughout the year; for example, activities during the planting and harvesting seasons, when women have an enormous amount of work to complete, should be avoided where possible (Alders 1996).

In many rural areas, women have not received formal education and so speak only local languages. Consequently, it is important that project meetings, training sessions, etc. be conducted in the local language and that non-formal methods of training be employed, especially where illiteracy is common (Alders 1996).

Community Health Nurses can also assist with the dissemination of information. These nurses frequently visit households to discuss health issues relating to mothers and infants. When discussing the vaccination of infants, they can also inform families of the availability of vaccines for their chickens and could suggest that interested women contact the local Community Livestock Worker or staff from the Veterinary Services Department for more information. Information on the nutritional benefits associated with the consumption of chicken meat and eggs should also be made available. This is very important in areas where there are taboos associated with the consumption of eggs.



8

Ethnoveterinary knowledge and Newcastle disease

Ethnoveterinary medicine (sometimes also called veterinary anthropology) deals with folk beliefs, knowledge, skills, methods and practices pertaining to the health care of animals (Guèye 1999; Mathias-Mundy and McCorkle 1989). Collecting information on ethnoveterinary knowledge in particular regions enables veterinarians to understand farmers' knowledge of the disease transmission process, local remedies that may be worthy of further study and the type of animal husbandry currently being practised. Examples of information collected in Ghana and Mozambique are given below.

In Ghana, detailed veterinary studies of the epidemiology of ND are yet to be done. However, compiling the knowledge of farmers assisted in the development of a ND control strategy. There are six agroecological zones within Ghana and this probably contributes to the variation in ethnoveterinary knowledge found within the country.

Farmers are generally aware of the seasonality of ND and this can assist in defining the timing of strategic ND vaccination campaigns. In addition, a vaccine which can be administered effectively in the same way as local remedies (e.g. via drinking water) is likely to be adopted readily by farmers.

- **Transmission of ND**. Many farmers know that ND is caused by the introduction of diseased birds. However, in some areas farmers believe that ND may have a spiritual origin. For instance, some believe that more birds die at Christmas because God wants more chickens at this time.
- Local treatments. All treatments described here are given by farmers when signs of ND appear in their birds or their neighbour's, but they acknowledge that these treatments have little impact on the course of the disease. Traditional medicine is generally not used to prevent the birds from getting ND. Some of the remedies administered include:
 - ground chillies in drinking water;
 - bark of the mango tree in drinking water; and
 - white vinegar given orally.



• Timing of outbreaks. Farmers generally associate ND outbreaks with the coming of the 'Harmatan winds' although some farmers recognise that an outbreak could occur at any time of the year.

In Mozambique there are ten agroecological zones and in the different zones, the name given to the disease varies as do the treatments used, for example:

- Local names for ND. *Chigubo-gubo* (Manica Province); *muzungo* (Maputo and Gaza Provinces); *mbendeni* and *quitjuku* (Inhambane Province). In some cases, the local name simply translates as "chicken epidemic" and so may be applied to a range of diseases. However, in general, when farmers mention these names, they are referring to high mortalities suffered by their flocks at specific periods and the signs described are those of ND.
- **Transmission of ND**. Many farmers know that ND is caused by the introduction of new birds which already have the disease.
- Local treatments. The remedies administered include:
 - OMO washing detergent mixed with food or water (Figure 12);
 - 3 or 4 drops of potassium permanganate in drinking water;
 - ground garlic mixed with maize bran or water;
 - sap of the *calveiro* cactus mixed with maize bran and water;
 - car battery water;
 - 30 to 40 chillies ground with salt and diluted in 2 litres of water and placed in a clay drinking pot for chickens of all ages;
 - chillies and garlic in water (Figure 12);
 - the bark of the Uepa (Tamarind) tree is placed in a drinking water receptacle and water added;
 - ground mango tree leaves in drinking water;
 - chopped roots of intxikile in drinking water; and
 - chopped fruit of the kulikwa in drinking water.
- Local methods of control. Diseased birds, and in some cases dead birds, are eaten and some farmers will bury the parts not eaten or put them in a pit latrine. Many farmers sell their birds as quickly as possible when the signs of ND appear.
- **Timing of outbreaks**. Farmers in Bilene District in Gaza Province state that ND occurs most commonly from August to October when mango trees are flowering. They also recognise that the disease may appear any time following the

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introduction of a diseased bird. In the districts of Inhambane, Jangamo and Homoine in Inhambane Province, outbreaks usually occur in September–October and again in January.



Figure 12: There are no known cures for ND. Although many traditional remedies exist, their efficacy is yet to be confirmed.



9

Development of an extension program for Newcastle disease vaccination campaigns

There is a growing understanding that rural people themselves are knowledgeable on the many subjects that touch their lives and that they possess a creativity and analytical capacity (Chambers 1991) which can greatly assist in the development of improved agricultural practices. Sriskandarajah et al. (1989) suggested that knowledge is not a commodity, for transfer from the informed to the uninformed, but the outcome of a dynamic, collaborative process between co-learners. Consequently, ideas about agricultural extension are changing to incorporate new ideas on participatory technology development (PTD).

Changing extension services which have been based on the transfer-of-technology (TOT) to PTD cannot be achieved rapidly. There are compelling reasons why participatory approaches are now recommended, especially with village poultry farmers since

- very little systematic work has been done on farming systems involving village poultry;
- extension messages are often ill-adapted to the management objectives of small-scale farmers (Adams 1982);
- farmers whose only livestock is village poultry generally belong to the poorer sections of rural communities, and may not have had regular contact with livestock extension programs which have frequently focused on ruminants;
- extension workers frequently find it easier to work with the better-off farmers who are usually more educated (Adams 1982) as they have more in common;
- participatory approaches allow extension workers to learn from and with rural people, eliciting and using their criteria and categories, and finding, understanding and appreciating indigenous technical knowledge (Chambers 1991).



9.1 Features of extension for village poultry production

What's different?

- Livestock extension networks have tended to focus on male farmers with cattle and small ruminants.
- Since the care of village poultry is often the responsibility of women and children, extension messages need to be specially designed and transmitted to reach their target. Men may or may not be targeted depending on their level of involvement in village chicken production.
- Village poultry extension is a relatively new field; we still have much to learn.
- Farmers are often reluctant to invest (time, money, materials) in chickens due to bad experiences in the past (e.g. frequent high mortality due to ND).
- Many farmers are unaware that it is possible to vaccinate chickens.

What's the same?

- Group work is generally the preferred way of interacting with farmers.
- Strategic planning of extension activities is critical, for example, the timing of ND vaccination campaigns is of vital importance to ensure that chickens are vaccinated before an outbreak occurs.
- The range of communication methods available to the extension worker is the same; although in many cases, non-formal methods of communication will be emphasised.
- Extension activities should be linked to ongoing collaborative research involving both specialists and farmers.

9.2 Extension methods

9.2.1 Group methods

There are a number of ways the extension agent can bring farmers together to undertake his/her extension work, for example:

• **Group meetings**. Working with groups of village poultry farmers is often one of the best ways of carrying out extension activities. When planning activities care must be taken to facilitate the participation of poultry farmers. Section 7.0

deals with issues to be considered in situations where the majority of village poultry farmers are women. In many rural areas, it is important that meetings, training sessions, etc. be conducted in the local language and that non-formal methods of training be employed, especially where illiteracy is common (Alders 1996). Drama and song can be extremely useful methods of conveying information. Teaching aids such as flip charts that use visual images to accompany an oral presentation can be very effective. Presenting appropriate extension material at local primary and secondary schools can also be a very rewarding exercise.

- **Demonstrations**. Farmers like to see how a new idea works (Oakley and Garforth 1985). Demonstrations are essential when training farmers in vaccination techniques and can also be useful when demonstrating designs for poultry houses, and food and water receptacles.
- Field days. Field days are an excellent way for farmers to share their ideas and to learn from the experiences of other farmers. These days also provide an opportunity for farmers to meet representatives of the government livestock and extension departments and learn about available services. Topics covered may include: ND vaccination techniques and vaccine conservation under village conditions; construction of poultry houses; design of feed and water receptacles; improved nutrition using locally available food (e.g. earthworms, termites, insects, ground shells, nutritious green leaves); the performance of different types of local birds and local poultry husbandry practices; and demonstrations on different diseases and how they may be controlled.
- **Tours/exchange visits**. These are generally extremely useful and can be great fun. However, it is often difficult for women to find the time and resources needed to leave their farms for any significant period of time.

9.2.2 Individual methods

In many cases, individual farm visits are not suited to situations where the extension worker is male and most poultry farmers are women. Poultry farmers can be encouraged to visit the local government or extension offices where possible. This method is more likely to be employed by Community Livestock Workers than extension workers.

9.2.3 Mass methods

Mass media include media that convey information by sound (radio, audio cassettes); moving pictures (television, film, video); and print (posters, newspapers, leaflets). The preparation of printed material in black and white enables the material to be photocopied long after the project has finished and in areas remote from the area where it was first produced. These channels of communication expose large numbers of people to the same information at the same time but do not readily allow an exchange of information between the farmers and the producers of the extension material. The attraction of mass media to extension services is the high speed and low cost with which information can be communicated to people over a wide area (Oakley and Garforth 1985).

Radio can be a very useful medium. Local radio stations frequently broadcast in local languages, and most villages will have at least one radio. Information can be received where literacy rates are low, and prepared statements, interviews (suggestions for a question and answer session may be found in appendix 6), drama and song can be used (Figure 13). Radio can assist in the coordination of vaccination campaigns. Interviews with local farmers who are skilled village poultry producers and who are willing to share their ideas will increase greatly the size of the listening audience. Farmers who are supportive of interventions such as ND vaccination can make a big impression on their fellow farmers. Conduct some audience surveys to ensure that your messages are being received clearly and that your target group is listening to the radio at the time your messages are being broadcast.

Extension material may be provided to the Ministry of Education for inclusion in the agriculture syllabus in schools.

Before selecting the extension method(s) to be employed, remember:

- No one method is better than any other the choice of methods depends on the situation;
- Use a range of methods experience has shown that new information reaches farmers more quickly when a range of communication methods are used;
- Different methods can be used together for example, the demonstration of a new technique may lead to a group discussion. The use of these two methods will reinforce the message.

• Use visual and written material where possible — this will reinforce the messaged being communicated. (Kang and Song 1984)



Figure 13: Song and drama performed in local languages are excellent ways of communicating ideas to farmers.

9.3 Participatory rural appraisal, participatory learning methods and participatory technology development

Participatory rural appraisal (PRA) was developed to gain information directly from rural communities and to enable the communities to do the analysis and planning using the information obtained. PRA has three foundations: methods; behaviour and attitudes; and sharing (Chambers 1991).

Participatory learning methods enable the extension worker to gain an understanding of the communities with whom s/he is working. Changes at the village and household level over time can be noted and s/he is able to constantly update material and provide opportunities for farmers to comment on all material produced.

Participatory technology development (PTD) often involves PRA techniques and takes participatory learning beyond situation analysis alone. PTD is essentially a process of purposeful and creative interaction between rural people and outside facilitators (Van Veldhuizen et al. 1997). Through this interaction, the partners

try to increase their understanding of the main traits and dynamics of the local farming systems, to define priority problems and opportunities, and to experiment with a selection of options for improvement that are most likely to succeed. The options are based on ideas and experiences derived from indigenous knowledge (both local and from farmers elsewhere) and formal science.

9.3.1 Participatory rural appraisal methods and Newcastle disease

It is vital that potential users of participatory technologies understand that the mere application of these techniques is not sufficient. The key to success is the belief that farmers can make a worthwhile contribution to the development process. If field workers have this attitude, then they will understand that the function of participatory methodologies is to facilitate understanding between groups of people (the outside field worker and the local farmer). Our objective is not to do participatory exercises and write reports; our objective is to understand farmers' realities with regard to village poultry production (Alders 2000).

Within the agricultural arena, most participatory technologies have been developed for use with crop or ruminant production. The range of methodologies discussed below is not sufficient to enable us to gain an understanding of all aspects of village poultry production. When we come upon new ground, we must keep communicating with farmers until we come to a common understanding.

- Secondary data review. Gather as much written information as you can on the history of ND in the country or area of interest. Patterns or seasonality in ND outbreaks may be apparent in reports. What has been the approach to ND control in the past: what vaccines were used, what personnel were involved in control activities and what lessons were learnt?
- **Direct conversation**, **including wandering around**. As simple as this sounds, it is a very important activity to undertake with both farmers and technical staff. Village poultry have often received very little attention and, consequently, there is little written about them. In many cases, the only way to learn what has happened is through open and friendly discussions. In addition, it is human not to report failures in writing.
- DIY (do-it-yourself), taking part in activities. Take part in training sessions, community discussions, vaccination campaigns and monitoring exercises. It is a useful way to find out what is and is not really happening.

- Key informants. Seek out people in the community recognised for their chicken raising skills and technical staff with experience and interest in village poultry.
- Key indicators. Discuss with community and technical groups how they measure the success of ND control activities. Possible responses may be:
 - increase in the number of chickens per household;
 - increase in the number of chickens vaccinated;
 - decrease in the number of chickens dying;
 - increase in the number of farmers presenting chickens for vaccination;
 - improve child nutrition; and
 - increase in the number of children attending school.
- Workshops and brainstorming. Bring key players together when developing a new ND control program. New interventions, no matter how well thought out or how successful they have been elsewhere, still need to be adapted to local conditions and local staff must be adequately trained.
- **Transects and group walks**. These activities can provide an enormous amount of information in a short time. While walking:
 - look at the chickens. How many chickens are running around? Are they healthy?
 - note the presence or absence of chicken houses. How easy will it be to catch chickens? Are the chicken houses well-designed?
 - note the distances between households. How many birds could be vaccinated in a day if the vaccinator moves on foot or by bicycle?
 - assess the demographics of the area. Who lives in the area? What are their living conditions?
 - note times of day when people are at home. What are the best times of day to hold meetings with farmers or to ask them to present their birds for vaccination?
- **Priority ranking and scoring**. Before starting ND control activities, meet with communities to ensure that such an activity is a priority from their point of view. If possible, discuss priorities separately with different groups in the community (e.g. male and female, young and old) as priorities may vary. One easy way of ranking priorities involves the use of index cards. Participants are asked to identify their priorities and a symbol (or word) is drawn onto a card. When all priorities have been identified, the cards are presented individually to

ensure that people recognise each symbol. Participants are then asked to place the cards in a line, in order of priority. This exercise can take a long time and the facilitator must be careful that s/he does not try to influence the outcome.

- Ethnoveterinary knowledge. To learn about the knowledge possessed by farmers in relation to animal production, a range of techniques is available. These techniques include informal interviews, group discussions, ranking and using a question list about specific diseases (Young 1992).
- **Participatory epidemiology**. This is an emerging field that is based on the use of participatory techniques for the harvesting of qualitative epidemiological intelligence contained within community observations, existing veterinary knowledge and traditional oral history (Mariner 1999).

In situations where the epidemiology of ND is not well understood, participatory epidemiology provides a cost-efficient way of obtaining qualitative data about whether or not there is a seasonal nature to ND outbreaks. This information can be very useful when developing a ND control program. It is advisable to undertake exercises in participatory epidemiology with representative samples of village chicken farmers in different agroecological zones.

Ask participating farmers or technical staff to complete a table (see Figure 14) indicating the months in which ND outbreaks are more likely to occur according to their own experience.

• **Question:** In which month(s) of the year is Newcastle disease more likely to occur? Place an X in the box next to the appropriate month(s).

| January | | July | X |
|----------|---|-----------|---|
| February | | August | |
| March | | September | |
| April | | October | |
| Мау | X | November | |
| June | × | December | X |

Figure 14: The response to a question used to determine whether there is a seasonal pattern to ND outbreaks.

To compile the data, the number of Xs per month are counted and these figures are used to construct a bar graph showing the number of Xs recorded for each month of the year. From a preliminary exercise conducted with workshop participants in Bhutan (see Figure 15), it was found that ND outbreaks were more likely to occur in the months of June and July.



Figure 15: Seasonality of ND outbreaks in Bhutan based on the experience of workshop participants.

When working with time periods, be sure to use time intervals familiar to farmers (e.g. months, specific festivals, seasonal events such as the flowering of particular plants or trees). In some circumstances, a farmer's calendar may not coincide with European calendar months. For instance, in Bhutan the New Year commences in the month of February. If farmers are not literate, then they may indicate their choices via ballot boxes that are appropriately marked with symbols for each period.

9.4 The extension worker

The extension worker is the critical element in all extension activities (Oakley and Garforth 1985). S/he has to work with people in many different ways and requires much tact and resourcefulness. No model of an extension worker's role is applicable to all situations. Every agent must consider each situation individually and adopt a position or role suitable to that situation.

There are four main areas to be considered when selecting extension workers (Adams 1982, Oakley and Garforth 1985).

- Educational qualifications. The level of education of the majority of farmers should determine the educational level of the extension agent. If farmers and extension workers have nothing in common, it is very difficult to identify a starting point for a program of improvement. In such cases, an alternative is to recruit the extension agents from the community and give them good training plus regular in-service training.
- Knowledge. The extension worker must be adequately trained in the technical aspects of his/her work and have a good working knowledge of the main elements of the agricultural system in which s/he is working. This includes anthropological and sociological studies of the rural area, local traditions, practices, culture and values. An understanding of the main approaches to adult education and group dynamics along with other techniques of developing farmer participation in extension activities is important.
- **Personal qualities**. The extension worker must have an ability to respect and to communicate with farmers; an ability to get on with people; enthusiasm for the job; and common sense and initiative.
- **Professional qualities**. The extension worker must have the ability to see problems through the eyes of the farmer and to be able to put him/herself in the farmer's shoes (*empathy*). The degree to which his/her advice or opinion is accepted by the farmers indicates his/her *credibility*. This will depend on the differences in background and also on the technical competence shown by the extension worker. *Humility* in an extension worker is shown by willingness to listen and learn before offering advice. *Professional commitment* to his/her work is essential if the extension agent is to succeed, as much of the time s/he is without direct supervision and under-resourced.

The selection of extension workers who are culturally acceptable is of utmost importance. In most cultures there are strong unwritten rules about what types of outsiders can be trusted (Zivetz 1990). This can relate to an individual's age, gender, social status, ethnicity or political affiliation. In areas where the majority of poultry farmers are women, efforts should be made to increase the number of women working as extension agents.

9.5 The development of extension programs for village poultry production

According to Oakley and Garforth (1985), an extension program is a written statement which contains the following four elements:

- objectives which the agent expects to be achieved in the area, within a specified period of time;
- means of achieving these objectives;
- · resources that are needed to fulfill the program; and
- a work plan indicating the schedule of extension activities that will lead to the fulfillment of the program objectives.

All of the above must be based on a solid understanding of the local situation. It is crucial that adequate time be given to collecting baseline data (see Appendix 7 for a suggested village poultry questionnaire) and to gaining an understanding of local priorities, resources and capabilities. Local farming systems and ethnoveterinary knowledge (husbandry practices, diseases, local remedies) relating to village poultry are just two of the areas which need to be investigated. Accessing farmers whose only livestock is poultry may not be an easy process. Much time and attention should be given to enabling these farmers to have an input into the content of the extension program. If correctly used, PRA and participatory learning methods will greatly assist the extension program (Chambers 1991, Mascarenhas 1991).

In areas where ND is endemic, it is suggested that village chicken extension programs concentrate initially on the control of ND. Until mortalities due to ND are reduced, farmers are unlikely to take an interest in learning about other ways of improving production in their poultry (Spradbrow 1996).

The first few ND vaccination campaigns in an area will be crucial for the success of ongoing extension activities. It will be easier to develop a suitable extension program for village chickens if:

- the vaccination campaigns are strategically implemented prior to seasonal outbreaks;
- the pre-campaign awareness-raising program is appropriately designed and enthusiastically implemented;

- the people doing the vaccinations have a good rapport with the farmers; and
- follow up work is done once it is obvious that farmers who vaccinated their flocks have lost fewer chickens due to ND.

Commence with a single, clear, consistent message presented in ways easily understood by farmers. Train your frontline extension staff well and equip them to carry the message to farmers (perhaps using illustrated flip charts, pamphlets, posters, audiocassettes, drama and song). Ensure that the message also reaches the farmers via a second source (e.g. radio, newspapers) to support the work being done by the extension workers.

Always stress that vaccination is never 100% effective (see Appendix 8). Birds may also die from causes other than ND. Vaccination campaigns should not be initiated in the face of an outbreak of ND, as farmers may associate mortalities in their chickens with the vaccination process.

If farmers are to be asked to purchase the ND vaccine, start the extension campaign early so that they have time to save enough to have all of their chickens vaccinated. Once ND control activities are in place, attention should be given to the identification of other constraints that farmers are willing to address. Remedies for these constraints might include improved housing, control of endoand ectoparasites, and improved nutrition through supplementary feeding.

9.6 Key topics for inclusion in field trial extension activities

It is essential that certain issues are discussed prior to initiating a field trial.

- Emphasise that it is a *trial* that is being undertaken and not a vaccination *campaign*. The outcome of the trial cannot be predicted and not all groups will necessarily demonstrate adequate levels of protection to the disease.
- A form of compensation should be discussed prior to starting, e.g. offer to vaccinate birds free of charge using the route of administration found to be most effective for a certain period after the trial finishes.
- In order to remove the possibility of bias, treatment groups can be allocated to different farmers or different communities using a lottery system conducted during a community meeting where representatives of all groups are present.

An ND vaccine field trial protocol is given in Appendix 9.

9.7 Field pre-testing of new extension material

It is vital that new extension material be pre-tested in the field prior to widespread diffusion to ensure that it will effectively communicate the desired message(s) to farmers (Bertrand 1978, Dudley and Haaland 1993; Haaland 1984; Zimmerman et al. 1996). A representative sample of the target group(s) should be selected and the material presented to them for discussion before it is finalised and reproduced for distribution. The type of language employed should reflect the everyday language of the target group(s). Technical words and expressions should be avoided as much as possible. When material is translated into local languages, the translation should also be checked by local farmers. Likewise, visual images should be checked to confirm that farmers will interpret the images in the intended manner. In rural areas, women often receive less formal education and may have difficulty in interpreting visual material presented in western ways.

Pre-testing does cost money but it can be done in relatively simple and cheap ways. The amount is insignificant compared to actual production costs and it can save money by avoiding the production of materials that are not understood or accepted (Bertrand 1978).



10

Conclusion

The control of ND in village chickens is much more than the control of an animal disease. It can make a vital contribution to the improvement of household food security and poverty alleviation in many developing countries. In some circumstances, it will provide the first contact between small-scale farmers and national veterinary services. As farmers increase their chicken numbers, some will use surplus birds to invest in small ruminants and eventually large animals.

The control of ND will contribute to improved village poultry production as a whole by assisting the process of data collection and reinforcing cooperative links with farmers. These links will greatly facilitate the ongoing work required to enable village chickens to demonstrate their true genetic potential.



11

References

- Adams, M.E. 1982. Agricultural Extension in Developing Countries, Intermediate Tropical Agriculture Series, Harlow, Longman.
- Alders, R. 1996. Facilitating women's participation in village poultry projects: experiences in Mozambique and Zambia. Proceedings of the 20th World's Poultry Congress, New Delhi, India, Vol. III, 441–447.
- Alders, R.G. 2000. Participatory methodologies and village poultry production. Proceedings of the Workshop on the Possibilities for Smallholder Poultry Projects in Eastern and Southern Africa, Morogoro, Tanzania, 22–25 May 2000, 103–109.
- Alders, R.G., Fringe, R. and Mata, B.V. 2001. Characteristics of the I-2 Live Thermostable Newcastle Disease Vaccine Produced at INIVE. ACIAR Proceedings of the SADC Planning Workshop Newcastle Disease Control in Village Chickens, Maputo, 6–9 March, 2000.
- Alders, R.G., Inoue, S. and Katongo, J.C. 1994. Prevalence and evaluation of Hitchner B1 and V4 vaccines for the control of Newcastle disease in village chickens in Zambia. Preventive Veterinary Medicine, 21: 125–132.
- Alexander, D.J. 1991. Newcastle disease. In: Rweyemamu, M.M., Palya, V., Win, T. and Sylla, D., ed. Newcastle Disease Vaccines for Rural Africa. Debre Zeit, Ethiopia, Pan African Veterinary Vaccine Centre, 7–45.
- Alexander, D.J., Wilson, G.W.C., Russell, P.H., Lister, S.A. and Parsons, G. 1985. Newcastle disease outbreaks in fowl in Great Britain during 1984. The Veterinary Record 117: 429–434.
- Anon. 1991. Websters Newcastle Disease Vaccine for Village Chickens. Castle Hill, Australia, Websters Pty Ltd, Information Dossier.
- Amakye-Anim, J., Alders, R.G., and Spradbrow, P.B. 1998. Trials with V4 Newcastle Disease Vaccine in Ghana. Fourth Asia Pacific Poultry Health Conference, Melbourne, Australia, November 22–26, 1998, Scientific Proceedings, 124.



N E W C A S T L E D I S E A S E

- Beard, C.W. and Hanson, R.P. 1981. Newcastle disease, In: Hofstad, M.S., ed. Diseases of Poultry, 7th ed., Ames, Iowa, Iowa State University Press, 452–470.
- Bell, J.G., Fotzo, T.M., Amara, A. and Agbebe, G. 1995. A field trial of the heat resistant V4 vaccine against Newcastle disease by eye drop inoculation in village poultry in Cameroon. Preventive Veterinary Medicine, 25: 19–25.
- Bensink, Z. and Spradbrow, P. 1999. Newcastle disease virus strain I-2- a prospective thermostable vaccine for use in developing countries. Veterinary Microbiology, 68: 131–139.
- Bertrand, J.T. 1978. Communications pretesting. Media Monograph 6 of the Communication Laboratory, Community and Family Study Centre, University of Chicago.
- Bratt, M.A. and Clavell, L.A. 1972. Haemolytic interaction of Newcastle disease virus and chicken erythrocytes. I. Quantitative comparison procedure. Applied Microbiology, 23: 454–460.
- Chambers, R. 1991. Participatory rural appraisals; past, present and future. Forests, Trees and People Newsletter No. 15/16, 4–9.
- Dias, P.T., Fringe, R., Mata, B.V. and Alders, R.G. 2001. Laboratory and Field Trials with Thermostable Live Newcastle Disease Vaccines in Mozambique. ACIAR Proceedings of the SADC Planning Workshop Newcastle Disease Control in Village Chickens, Maputo, 6–9 March, 2000.
- Dudley, E. and Haaland, A. 1993. Communicating Building for Safety. London, Intermediate Technology Publications.
- FAO 1994. A manual for the primary animal health care worker: guidelines for training. Rome, FAO.
- FAO 1997. Guidelines for the inclusion of improved household poultry production. Diversification component of the Special Programme for Food Security. Rome, FAO.
- Guèye, E.F. 1999. Ethnoveterinary medicine against poultry diseases in African villages. World's Poultry Science Journal, 55: 187–198.

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- Guèye, E.F. 2000. The role of family poultry in poverty alleviation, food security and the promotion of gender equality in rural Africa. Outlook on Agriculture, 29 (2): 129–136.
- Gujit, I. 1994. Making a difference: integrating gender analysis into PRA Training. Rapid Rural Appraisal Notes No. 19, Special Issue on Training, Sustainable Agriculture Programme, International Institute for Environment and Development. February 1994, 49–55.
- Haaland, A. 1984. Pretesting Communication Materials with Special Reference on Child Health and Nutrition Education. Rangoon, UNICEF.
- Heath, B.C., Lindsey, M.J., McManus, K.P. and Claxton, P.D. 1992. Webster's Newcastle disease vaccine for village chickens. In: Spradbrow, P.B. ed., Newcastle disease in village chickens. Canberra, ACIAR Proceedings No. 39, 104–109.
- Ideris, A., Ibrahim, A.L., Spradbrow, P.B. and Hung Seng, C. 1987. Development of Food Pellet Newcastle Disease Vaccine. In: Copland, J.W., ed. Newcastle Disease in Poultry: A New Food Pellet Vaccine. Canberra, ACIAR, 20–23.
- Kang, J.T. and Song, H.K. 1984. Individual and group methods in extension training. In: Rural Extension: Reference Manual, 2nd Edition, B.E. Swanson (ed.), Rome, FAO, 129–140.
- Mariner, J.C. 1999. Participatory Epidemiology: Methods for the Collection of Action-Oriented Epidemiological Intelligence. Vetwork UK, RDP Livestock Services B.V. and FAO.
- Mascarenhas, J. 1991. Participatory rural appraisal and participatory learning methods: recent experiences from MYRADA and South India. Forests, Trees and People Newsletter No. 15/16, 10–17.
- Mathias-Mundy, E. and McCorkle, C.M. 1989. Ethnoveterinary medicine: an annotated bibliography. Bibliographies in Technology and Social Change Series No. 6, Iowa State University Research Foundation.
- Oakley, P. and Garforth, C. 1985. Guide to extension training. Rome, Food and Agriculture Organisation of the United Nations.

N E W C A S T L E D I S E A S E

- Pangani, P. 1999. Stock Breeding Support Programme in Gaza and Inhambane Provinces. Draft Activity Report by the Animal Health Advisor. VETAID, Xai Xai, October 1999.
- Permin, A. and Hansen, J.W. 1998. Epidemiology, Diagnosis and Control of Poultry Parasites. FAO Animal Health Manual No. 4. Rome, FAO.
- Pinstrup-Andersen, P., Burger, S., Habicht, J. and Petersen, K. 1993. Protein Energy Malnutrition. In: Jamison, D.T., Mosley, W.H., Measham, A.R. and Bobadilla, J.L., ed. Disease Control Priorities in Developing Countries, Oxford University Press, 391–420.
- Sonaiya, E.B., Branckaert, R.D.S. and Guèye, E.F. 1999. Research and development options for family poultry. First INFPD/FAO Electronic Conference on Family Poultry, Guèye, E.F., (Ed). (http://www.fao.org/ag/aga/agap/lpa/fampo1/ intropap.htm)
- Sriskandarajah, N., Bawden, R.J. and Packham, R.G. 1989. Systems Agriculture: A Paradigm for Sustainability. Paper presented at the Ninth Annual Farming Systems Research/Extension Symposium, University of Arkansas, Fayetteville, Arkansas, USA, October 9–11, 1989.
- Spradbrow, P.B. 1988. Geographical distribution. In: Alexander, D.J., ed. Newcastle disease. Boston, MA, Kluwer Academic Publishers, 247.
- Spradbrow, P.B. 1993–94. Newcastle Disease in Village Chickens. Poultry Science Review, 5, 57–96.
- Spradbrow, P.B. 1996. Protection against important diseases including Newcastle disease. Proceedings of the 20th World's Poultry Congress, New Delhi, India. Vol. I, 31–34.
- Spradbrow, P. and Foster, A. 1997. Counting your Chickens. In: Lawrence, J., ed. Partners in the Harvest. Canberra, Australian Centre for International Agricultural Research, 51–57.
- Spradbrow, P.B., Samuel, J.L. and Ibrahim, L. 1988. Serological response of chickens to oral vaccination with Newcastle disease virus. Veterinary Microbiology, 16: 255–262.

- Tu, T.D., Phuc, K.V., Dinh, N.T.K., Quoc, D.N. and Spradbrow, P.B. 1998.
 Vietnamese trials with a thermostable Newcastle disease vaccine (strain I-2) in experimental and village chickens. Preventive Veterinary Medicine, 34: 205–214.
- Van Veldhuizen, L., Waters-Bayer, A. and De Zeeuw, H. 1997. Developing Technology with Farmers: A Trainer's Guide for Participatory Learning. London, Zed Books Ltd.
- Wambura, P.N., Kapaga, A.M. and Hyera, J.M.K. 2000. Experimental trials with a thermostable Newcastle disease virus (strain I-2) in commercial and village chickens in Tanzania. Preventive Veterinary Medicine, 43: 75–83.
- Young, J. 1992. How to collect ethnoveterinary information. Report on the ITDG Village Animal Healthcare Workshop, Kenya, February 1992. Appendix 5, 12–15.
- Zimmerman, M., Newton, N., Frumin, L. and Wittett, S. 1996. Developing Health and Family Planning Materials for Low-Literate Audiences: A Guide. Washington DC, Program for Appropriate Technology in Health.
- Zivetz, L. 1990. Project Identification, Design and Appraisal: A Manual for NGOs. Canberra, The Australian Council for Overseas Aid.



Appendix 1: Post-mortem technique for domestic fowl

The use of a consistent routine of post-mortem examination will greatly enhance your ability to recognise abnormalities in organs and tissues. This sequence of dissection for the domestic fowl allows observation of all body systems, and outlines suitable methods for collection of specimens for laboratory examination.

When selecting birds for autopsy, take live birds that are showing typical signs, rather than those that are *in extremis* or dead. In these, the primary disease may be obscured by secondary diseases or by post-mortem decomposition.

Examine the bird for clinical abnormalities before it is killed. This may indicate a particular system or organ that needs special attention during the post-mortem examination.

Humane killing of birds

It is important to kill birds in a humane, efficient manner which does not itself cause changes that might confuse the diagnosis.

Cervical dislocation

- Grasp the legs and primary wing feathers with one hand, so the bird cannot flutter.
- With the other hand, grasp the bird's head from above, holding the head between the first (index) and second fingers. Curve the fingers along the bottoms of the jaw. This avoids pressure on the larynx and tongue when the neck is broken.
- Hold the bird across your body, with its head downwards. No undue force is used at any time up to this point. Bend the bird's head backwards.
- Break the bird's neck using a fairly strong rapid stretching action, keeping the head bent backwards. The bird will lose consciousness immediately, but will make strong reflex movements for about 2 minutes after neck dislocation. While struggling continues, keep the bird immobilised by maintaining your grip on the wing bases. Elevate the head to lessen the likelihood of inhalation of crop content which may be regurgitated.



Intravenous injection of air

- Grasp the bird by the wing bases with the left hand and immobilise it over the edge of a table.
- Pluck a few feathers over the brachial vein.
- Compress the base of the wing with the left index finger to distend the brachial vein.
- Place the needle into the vein and rapidly inject 6–7 mL of air. Reflex struggling is brief and there is no trauma to neck structures. The bird does not regurgitate, as it may after cervical dislocation. Again, the wing bases should be held firmly to prevent the carcass escaping.

Materials:

A sharp knife A pair of scissors A pair of shears (or garden secateurs) A pair of forceps Bottles containing 50% glycerine in saline Bottles containing 10% formalin (Remember that the volume of 10% formalin used should be at least 10 times that of the tissue to be fixed.) A coolbox with ice or ice-packs (if possible)

Post-mortem technique for domestic fowl

- Examine the exterior of the carcass:
 - the vent for discharges;
 - the skin by reflecting the feathers, which may hide skin lesions or external parasites (see Permin and Hansen 1998 for more detail on the diagnosis of poultry parasites); and
 - the orifices of the head for discharges and the colour of the mucous membranes.
- Position the carcass on its back with legs towards you.
- Push a sharp knife through the fold produced by lifting the skin on the posterior part of the sternum. Draw the knife back through the fold, producing a v-shaped incision. Enlarge this until the flap of skin that has been freed can be grasped firmly and drawn forward with the left hand, while holding the legs in the right hand. Continue the skin incision along the neck to the head using the knife.

- Reflect the skin from the upper part of the legs, and dislocate the hip joints. The carcass can now lie flat. The general colour and condition of the musculature is noted at this point.
- Rotate the carcass so that the head lies towards you. Reflect the skin from the structures of the neck.
- Open the mouth and cut through the left temporo-mandibular joint with shears. Cut open the mouth with scissors, continuing the incision through the wall of the pharynx and oesophagus as far as the crop. Open and examine for changes.
- Cut horizontally through the beak just above the nostrils with shears to open the nasal cavities and infra-orbital sinuses. Open and examine for changes.
- Remove the small cartilagenous nasal turbinates using small scissors or a scalpel blade.
- Using small or sharp-pointed scissors, enter the larynx and open the trachea using only the tips of the scissors. Take care not to damage the mucosa. Spread open the trachea and examine the mucosa for exudate and inflammation.
- Rotate the carcass back to its original position with the tail towards you.
- Open the abdomen with scissors, starting just above the pubis. Continue the abdominal incision through the costochondral junctions of the last few ribs on the right-hand side of the bird. Use the scissors to cut the pectoral muscles of the left side of the thorax.
- Cut the remaining ribs, the coracoid bone and the clavicle on the left side using shears. Cut the remaining ligaments and muscles and reflect the sternum to your left until the coracoid bone on the other side breaks. This allows the contents of the thorax and abdomen to be displayed.
- Cut the omentum and reflect. The first structures to be displayed are the abdominal airsacs; these are easily destroyed and should be examined at this early stage of the abdominal dissection. They should be very thin and transparent; a pale mucoid thickened appearance suggests chronic respiratory disease.
- Draw the gizzard out to your left. Hold the small intestine by its mesentery and draw out the intestine to the left, freeing mesenteric attachments and airsacs.
Most of the abdominal viscera and organs are now exposed (Figure 16), and the dissection has reached what can be called the Display Stage. This is a most important stage of dissection in any species and is the time for recording the general state of nutrition, presence or absence of anaemia, exudates in serous cavities, malposition of viscera, etc.



Figure 16: The display stage of the necropsy showing the major organs of the chicken. The trachea, proventriculus and caeca should be examined for lesions.

• Free the intestine by cutting the colon just before it enters the cloaca. Cut the colonic, caecal and small intestinal mesenteries close to the gut and draw the bowel out to the left. Free the paired caeca.

It is best not to separate the two arms of the u-shaped duodenum, for the pancreas lies between and is easily damaged. The two arms of the duodenum can be opened and examined and may then be fixed as a whole for histological examination.

- Open the remaining small intestine with the scissors, taking care not to damage the underlying mucosa. Any part of the mucosa that shows abnormality can be examined microscopically by taking a scraping of the full thickness of the mucosa, using a blade of the scissors or a scalpel blade. Transfer the mucosal sample to a microscope slide. Apply a coverslip and carefully press onto the sample using a pointed instrument, not your dirty glove. Microscopic examination of this wet preparation will reveal the presence of various stages of coccidiosis.
- Open the intestine along its entire length. Examine the junctions of the small intestine and the colon at the base of the caeca. Note the caecal tonsils (small nodules of lymphoid tissue) at this junction. These may contain small red spots which suggest local haemorrhage, but are in fact normal.
- Open the paired caeca. Scrapings of the mucosa may be examined for caecal coccidiosis. Always examine the very tips of the caeca, where the small nematode *Heterakis gallinae* may be found.
- Cut the heavy muscular wall of the gizzard with a knife and extend the incision up through the proventriculus and lower oesophagus to the crop. The lining of the gizzard should be tough and tightly adherent to the mucosa. If it strips easily to reveal oedema or underlying haemorrhage in a freshly dead bird, toxic damage may be suspected.
- Examine the crop for presence and type of feed and any degeneration of the mucosa.
- Inspect the spleen by reflecting the gizzard to the left. Reflect the opened gizzard and proventriculus to the left.
- Examine the heart. Note the size and contents, if any, of the pericardial sac. An increase in size of the heart in relation to the carcass gives the best indication

of the presence of heart failure. Remove the heart by cutting through the vessels and atria at its base. There will be froth in the right atrium if the bird was killed by an intravenous injection of air.

- Cut the myocardium of the right and left ventricle from base to apex, and examine the interior of the heart. For histological examination, the whole open heart should be fixed in formalin. In cases of suspected septicaemia, blood can be aseptically aspirated before removing the heart.
- Inspect the liver *in situ* and note its colour, shape and size relative to the carcass. Normal liver is very fragile and should be handled gently. The texture of the organ is noted during handling, and the consistency of the liver is noted while cutting a block for histological examination. Cut the liver from its attachments. The surface may be cleaned by wiping with the knife blade. This is preferable to washing with water which causes osmotic damage to the tissue.
- Note the size of the gall bladder. Remember that the gall bladder has not been stimulated to empty in birds that have not been eating, and may appear overfull.
- Dissect the lungs away from the ribs and roof of the thorax. Normal aerated lung can be fixed by floating the organ unsliced in formalin, as the fixative diffuses readily through air-filled tissue.
- Examine the ovary which often contains wrinkled, discoloured, involuting ova in birds or normal hens that have recently gone off lay. Reflect the ovary to reveal the adrenal glands.
- Remove the ovary and remains of the abdominal airsacs to reveal the kidneys. Note their size, shape and colour. Check the ureters for the presence of excessive amounts of white urates, which might indicate nephrosis. This should be done before the kidneys are removed. Normal kidney is very friable.
- Examine the sciatic nerve plexus which can be observed on each side of the spinal column. Check the spinal column for deformity at this stage.
- Display the continuation of the sciatic nerve in each leg by reflecting the adductor muscle of the thigh. The normal unstretched nerve should be glistening white, with fine cross striations.
- Examine the fine intercostal nerves, and the brachial plexus on each side, dorsal to the thoracic airsacs. Particular attention to nerves is necessary in the

diagnosis of Marek's disease, a viral infection which causes enlargement and greyish discolouration of affected nerves.

- Examine the spinal and vagus nerves in the neck.
- In young birds examine the thymus, the thyroid and parathyroid glands. The thyroids and parathyroids lie together on each side of the neck close to the origin of the common carotid arteries.
- Examine the leg joints. Open the tibio-tarsal joint by cutting the medial collateral, and dorsal tibio-tarsal ligaments and dislocating the joint. Pass the blade between the posterior end of the tibia and the sesamoid bone in the Achilles tendon. The stifle joint is opened in a similar manner.
- Examine the growth plates of long limb bones especially in young lame meat birds that have grown rapidly. Expose the proximal end of the tibia, and shave off part of the bone to display the growth plate.
- Examine the bone marrow by splitting the femur. The marrow in the normal bird is red.
- Remove the head by cutting through the atlanto-occipital joint.
- Remove the skin from the head and open the cranium using pointed shears. Begin just above the occipital condyles lateral to the foramen magnum. Then extend the incisions on each side to meet above the eyes. Care must be taken to avoid damage to the underlying brain. Remove the cranial cap carefully. Cut away the remnants of the dura matter and observe the brain.
- Remove the brain by cutting the cranial nerves beneath it. Gently displace the brain back until it can be extracted and all surfaces examined.
- In young birds, open the bursa of Fabricius through its opening to the cloaca.

Based on the technique used by Professor Roger Kelly. Adapted by Dr Mary Young. The Division of Veterinary Pathology and Anatomy, The University of Queensland, Australia



Appendix 2: Collection of blood from the wing vein of chickens

This technique uses a needle and syringe. A 25 G ($0.50 \times 16 \text{ mm}$) needle is used for chicks under 4 weeks of age, and a 23 G ($0.65 \times 32 \text{ mm}$) needle for older chickens. Plastic syringes of 1.0 or 2.5 mL capacity are convenient. Both needles and syringes can be washed for reuse.

If you have an assistant

- Ask the assistant to hold the chicken horizontally against her or him with its head to her right.
- Pull the right wing out towards you, if necessary pluck away the small feathers from the underside overlying the humerus, and swab with 70% alcohol. The wing vein, named in various text-books as the brachial, ulnar or cutaneous ulnar, is clearly visible running between the biceps and triceps muscles.
- Insert the needle under the tendon of the pronator muscle, in the triangle formed where the wing vein bifurcates (see Figure 15), pointing the needle proximally i.e. in the direction of the blood flow. Do not go too deep or the needle will scrape the humerus and the chicken will struggle. Likewise keep clear of the ulnar nerve. With a little gentle probing you should enter the vein easily. This approach from under the tendon makes it easier to enter the vein than does aiming directly for it, and also tends to steady the needle if the bird moves.

If working alone

- Sit with the chicken horizontal between your thighs, head away from you, lying half on its back and half turned on its right side.
- Clamp down its legs with your left elbow (if you are right-handed) and its neck with your left forearm, and with your left hand spread out its left wing.
- With your right hand, proceed as above.

This works with all chickens except the dedicated strugglers. Some people prefer to hold the birds with the head towards them; if you can perfect both techniques you will have two veins to choose from.

Withdraw blood by gentle suction since the veins on chickens collapse readily. After the needle is removed, apply pressure to the vein for a few seconds to discourage further bleeding. Immediately label the syringe with the number of the chicken.



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Figure 17: Illustration of a convenient anatomical site for bleeding chickens (adapted from J. Samuel).

If the blood is for serum collection, leave it in the syringe and store the syringe in a slanting position, with the needle end pointing upwards. Leave an air space between the blood and the end of the syringe. After collection, if possible, leave the syringes in a warm room at 37°C for one hour to assist coagulation. If the

blood is for the preparation of red blood cells, the collection will have been made into an anticoagulant. Mix the blood gently while it is in the syringe, remove the needle and transfer the blood to a vessel with a screw cap. If the blood is discharged through the needle, there is a chance that some blood cells will haemolyse.

It is possible to pierce a wing vein with a needle and then to collect the freely flowing blood into a small vial. This delivers a less satisfactory sample that will invariably be contaminated with bacteria. Also the chicken is likely to be discoloured with blood, and some owners will object to this.

Based on the technique used by Dr Janeen Samuel (Australia) and Dr Rini Dharsana (Indonesia).



Appendix 3: Calibration and care of Eye-droppers

Eye-droppers

Eye-droppers are made of flexible plastic, preferably low density polyethylene. The ideal eye-dropper has a removable tip, protected by a screw top cap. A suitable eye-dropper should:

- hold a suitable volume of vaccine;
- not inactivate (destroy) the vaccine virus; and
- deliver drops of an appropriate size.

Volume of droppers

Eye-droppers of up to 30 mL capacity can be used. It is not necessary to place a full 30 mL of vaccine in these droppers. The volume of vaccine that is used will depend on the number of chickens to be vaccinated, and the size of the drop delivered by the dropper.

Testing for antiviral activity

Vaccine virus will be killed on exposure to some plastics. A sample of a new batch of eye-droppers should be tested before use in the field. This test must be done in a laboratory that is able to measure the infectivity of the vaccine virus. Vaccine virus should be diluted for use and then divided into two parts. The diluent should contain antibiotics as the laboratory will require vaccine free of contamination when the virus content is measured in eggs. Place half the vaccine in the eye-dropper and half in a stoppered, sterile glass test tube (or leave it in the vaccine vial). Store both overnight in a cool, dark location. The two preparations are then tested to confirm that there is little or no difference in virus content between the vaccine stored in the eye-dropper and that stored in the test tube.

Size of drops

The volume of diluent used to reconstitute freeze-dried vaccine, or to dilute liquid vaccine, will depend on the size of the drop that is formed by the eye-dropper. It



is the outside diameter of the tip, not the inside diameter, that determines drop size. It is best to use an eye-dropper that produces more than 40 drops per mL. If the eye-dropper produces 66 drops per mL (an ideal number) it means that each drop is approximately 15 μ L. This volume is ideal for the small eye of a chicken.

Human eye-droppers are not as convenient for use in chickens. These often produce drops of 25 μL to 35 μL . Such drops are large compared to the size of a chicken's eye and splashing of the drop and wastage of the vaccine can occur.

Each new batch of eye-droppers should be calibrated to ensure that chickens receive the correct dose of vaccine.

Calibration Method Number 1 — for use with freeze-dried vaccine

- 1. Remove the tip of the eye-dropper (Figure 18, step 1), add 1 mL of water to the dropper (steps 2 to 5) and then replace the tip securely (step 6).
- 2. Hold the eye-dropper upside down, squeeze the dropper very gently and count the number of drops that fall from the tip (step 7). Remember that the eye-dropper should be held in the vertical position (see Figure 6 in Section 5.1.1). It is generally advisable to repeat this process three times and to use the average number of drops in the calculation below.
- 3. Use the following formula to calculate the volume of diluent required to dilute the number of doses of the vaccine per vial and the eye-dropper in use:

Volume of diluent (mL) = $\frac{\text{No. of doses of vaccine per vial}}{\text{No. of drops formed per mL}}$

Example 1: How much diluent should be added to a vial containing 250 doses of ND vaccine given that 1 mL of water in the eye-dropper yielded 50 drops?

Volume of diluent (mL) = $\frac{250 \text{ doses per vial}}{50 \text{ drops per mL}}$ = 5 mL per vial

Example 2: How much diluent should be added to a vial containing 100 doses of ND vaccine given that 1 mL of water in the eye-dropper yielded 37 drops?

Volume of diluent (mL) = $\frac{100 \text{ doses per vial}}{37 \text{ drops per mL}}$ = 2.7 mL per vial

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Calibration Method Number 2 — for use with freeze-dried vaccine

This method is easier for people less familiar with syringes and mathematical calculations. It is better if two people to work together.

- 1. Check the vaccine label to determine the number of doses per vial.
- 2. Remove the tip of the eye-dropper (Figure 19, step 1), fill the eye-dropper with water (step 2) and replace the tip (step 3).
- 3. Remove the plunger from a 10 mL or 20 mL syringe (step 4) and hold the syringe vertically with the tip down. The tip should be closed with a finger or a thumb (step 5).
- 4. Hold the eye-dropper vertically, squeeze the eye-dropper very gently and commence counting drops into the syringe (step 6). Continue counting until the number of drops equals the number of doses contained in the vaccine vial. Many people find it easier to count the drops in groups of ten and record the number of groups. For instance, for a 250 dose vial, count 25 groups of 10 drops to give a total of 250 drops. Working in pairs, people count to 10 and then make a mark on the ground.
- 5. Hold the syringe vertically and check the level of the water against the marks on the syringe. This is the volume required to dilute the vaccine. Repeat three times.

If it is necessary to use glass eye-droppers with a rubber bulb, this method of calibration can be used.

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Figure 19: Calibration method number 2.

Calibration Method Number 3 — for use with "wet" vaccine

The method described below should be considered a guide rather than a formal "calibration" method and may be used when working with locally-produced "wet" I-2 ND vaccine. It will be useful in circumstances where those involved with fieldwork would prefer not to perform complex calculations. A more accurate method for use with "wet" vaccines may be found in the companion ACIAR ND laboratory manual.

- 1. Remove the tip of the eye-dropper (Figure 19, step 1), fill the eye-dropper with water (step 2) and replace the tip (step 3).
- 2. Remove the plunger from a 5 mL or 10 mL syringe (step 4) and hold the syringe vertically with the tip down. The tip should be closed with a finger or a thumb (step 5).
- 3. Hold the eye-dropper vertically, squeeze the eye-dropper very gently and commence counting drops into the syringe (step 6).
- 4. Count the drops until the water in the syringe has reached the 1 mL mark.
- 5. Record the number of drops. Repeat this process twice to confirm that the number of drops counted is correct.

If 50 or less drops were required to fill the syringe to the 1 mL mark, then the eye-dropper is suitable for administration of the "wet" I-2 ND vaccine by single eye-drop application. Eye-droppers that require considerably less than 50 drops to yield 1 mL, will deliver a higher dose of the vaccine. This will result in a greater immune response by the bird and will not cause any harmful reactions as the I-2 vaccine remains innocuous even in large doses.

This rule may be used providing that the following conditions are met:

- The eye-dropper is made of a suitable plastic and has been approved by the laboratory producing the I-2 vaccine.
- The I-2 vaccine is prepared by mixing equal volumes of allantoic fluid and a stabiliser (such as 2% gelatin).
- The average titre of allantoic fluid is 10⁹ EID₅₀/mL of I-2 virus. The laboratory distributes the vaccine such that it contains 10⁷ EID₅₀ of I-2 virus per dose to ensure that after transport each bird will receive a dose of 10⁶ EID₅₀ that is required to provoke an adequate immune response.

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This method is based on the fact that 10 μ L of allantoic fluid with a titre of 10⁹ EID₅₀/mL will contain 10⁷ EID₅₀ of I-2 virus. When this allantoic fluid is mixed with an equal volume of a stabiliser, 10⁷ EID₅₀ of I-2 virus will be contained in 20 μ L. Fifty drops containing 20 μ L each are required to make 1 mL.

How to care for plastic eye-droppers

To ensure a long life for eye-droppers, they must be cleaned and stored correctly after use.

- 1. Wash in cool, clean water only. Do not use HOT water.
- 2. Do not use treated tap water. If you only have access to treated tap water, it is advisable to let it stand overnight to allow the chlorine to evaporate.
- 3. Do not use disinfectants as they will inactivate the vaccine virus.
- 4. Do not clean the tip of the eye-dropper with anything abrasive.
- 5. Do not force anything into the tip of the eye-dropper that will enlarge the opening.
- 6. Allow the eye-dropper to dry thoroughly and then wrap in a dry clean cloth.
- 7. Store away from direct sunlight, sources of heat, rats and mice!



Appendix 4:

The role of community livestock workers in the control of Newcastle disease

It is now well recognised that any non-compulsory program that does not have community participation is unlikely to be sustainable in the long term. In addition, government veterinary and extension services in most countries are unable to provide cost-effective services such as routine vaccination to all local communities, especially those in remote areas. Therefore, community participation is crucial to the development of a sustainable program to control ND in village chickens.

In many developing countries, the work of government veterinary and extension staff has been complemented at the village level by persons from the local community, the Community Livestock Workers (CLWs). A CLW is a man or a woman selected by the local community or appointed with their agreement to deal with animal health and production in the community (FAO 1994). S/he must be able to communicate effectively in the language used by farmers, read labels and instructions, and keep appropriate records.

In situations where most farmers raise only poultry, it may be best to commence with the training of community vaccinators. These vaccinators may receive further training to become CLWs should the community demand such a service.

Many factors affect the success of CLW programs. These include: Social factors

- The program must be based on community needs and use existing structures or organisations.
- The community must be involved in all parts and stages of the program.
- The CLW must be chosen and respected by the community.
- There should be an ongoing exchange of information between all stakeholders.

Technical factors

- CLWs and their trainers must identify and respect local knowledge.
- They must be able to fulfill their technical responsibilities.



• There must be collaboration with beneficiaries and sources of technical support to ensure that CLWs offer appropriate technical advice.

Institutional factors

• The CLW program must cooperate with all existing institutional structures (government, traditional, local project, government veterinary and livestock extension services, NGOs) in the planning and implementation of the program.

Economic factors

- To ensure economic sustainability, the local community must be involved in the development of cost-recovery mechanisms for the program.
- Charging for services (financial contribution by the beneficiaries to cover the cost of the vaccine and its administration by a CLW) is essential to the success of the program. To start with, charges may represent only partial cost-recovery.
- CLWs will obtain most benefit from ND control activities if they also raise village chickens. Protecting their own birds from ND will provide a greater economic return than the money earned vaccinating the birds of others.

Environmental factors

- The program should not contribute to environmental degradation.
- Care with the use of chemicals and antibiotics should be emphasised.

Compared to government veterinary and extension staff, CLWs

- provide more cost-efficient services, and transport and labour costs are reduced;
- are more flexible in their working hours, e.g. they are available for weekend and after-hours work; and
- are more accessible to village poultry owners.

To ensure sustainability of the program, CLWs must:

- be assured of a reliable supply of vaccine (and other necessary inputs);
- receive appropriate training;
- be answerable to their community;

- be able to monitor their own work;
- · be provided with incentive in cash or kind; and
- receive good technical follow-up and support.

The training of CLWs is an important component of the ND control program. Factors to be considered include:

- who does the training?
- where should the training be conducted?
- who is responsible for post-training supervision?
- who is responsible for monitoring?
- who is responsible for evaluation?

The training program for CLWs involved in control of ND should include:

| Features of a chicken | — simple anatomy — recognition of healthy and sick chickens |
|---|--|
| Handling of a chicken | |
| Husbandry | housing: ventilation, cleaning, predators nutrition: young chicks, use of supplements |
| Diseases of chickens | clinical signs, field diagnosis, treatment and control of: Newcastle disease External parasites Internal parasites (coccidiosis, helminths) Fowl cholera, Fowl pox |
| Vaccination techniquesRecord keeping | eye drop and drinking water for ND vaccines number of cases diagnosis and treatment of cases outcome of treatment |

- inventory of stock (pharmaceuticals, etc.)
- vaccinations performed
- payment received

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In order to perform eye drop vaccination, CLWs must be able to:

- read numbers on a syringe;
- understand the meaning of the lines and intervals between the numbered lines on a syringe;
- read and check the number of doses of ND vaccine per vial and the expiry date of the vaccine;
- use the syringe to put the appropriate volume of water into a vial and draw vaccine out (if using vaccine that requires dilution);
- · check that the vaccine is properly diluted;
- · shake vial completely to dissolve all vaccine;
- assemble an eye-dropper;
- hold the eye-dropper vertically to form a drop of the correct size;
- · check that the correct number of drops leave the eye-dropper;
- · hold a chicken gently and calmly; and
- clean an eye-dropper and syringe correctly.

The ND vaccination kit for CLWs should contain:

- syringe (10 mL or smaller if appropriate), needle optional;
- calibrated eye-dropper;
- ND vaccine
- coolbox and ice pack or damp cloth and basket;
- record book and pencil; and
- chicken marker-leg band, wing tag, coloured thread or cord etc.

Indicators of success to be used by CLWs to evaluate their work:

- an increase in the number of chickens per family/household;
- · farmers continue to participate in subsequent vaccination campaigns;
- new farmers present their chickens for vaccination at each campaign; and
- payment received from farmers for the vaccination of their chickens is sufficient to buy vaccine for the following campaign and to cover any transport or labour costs involved.

Appendix 5: Comparison of some strains of Newcastle disease virus

| Strain designation | Conventional classification ^a | Use | Mean death time ^b | ICPI¢ | IVPId | к ^е | Virulence sequence ^f |
|-----------------------|--|--------------------------------------|---------------------------------|-------|-------|-----------------------|------------------------------------|
| I-2 | Avirulent | Village vaccine | >150 | 0 | 0 | NR ^g | No |
| V4 | Avirulent | Village and commercial vaccine | >150 | 0.16 | 0 | 0.23 | No |
| Hitchner B1 | Lentogenic | Commercial vaccine | 120 | 0.20 | 0 | NR | No |
| La Sota | Lentogenic | Commercial vaccine | 103 | 0.40 | 0 | 2.08 | No |
| Komarov | Mesogenic | Commercial vaccine | 69 | 1.41 | 0 | NR | Yes |
| Mukteswar | Mesogenic | Commercial vaccine | 46 | 1.40 | 0 | NR | Yes |
| Herts 33/56 | Velogenic | Challenge strain | 48 | 1.88 | 2.7 | 0.86 | Yes |

^a These useful terms are no longer used by OIE

^b Time in hours for a minimal lethal dose to kill chicken embryos

^c Intracerebral pathogenicity index, ranging from 0 (least pathogenic) to 2 (most pathogenic)

^d Intravenous pathogenicity index, ranging from 0 (least pathogenic) to 3 (most pathogenic)

^e Rate constant of thermostability of infectivity at 56°C. Lower figures indicate greater heat stability

^f The amino acid sequence ¹¹²RRQR(orK)RF¹¹⁷ at the cleavage site of the F protein

^g Not recorded

Data derived from various sources:

- Alexander, D.J. 1997. Newcastle disease and other avian paramyxoviridae infections. In: Diseases of Poultry, 10th Ed., B.W. Calnek (ed.) London, Mosby-Wolfe, 541–569.
- Alexander, D.J. and Allan, W.H. 1973. Newcastle disease. The nature of the virus strains. Bulletin of the Office International des Epizooties, 79: 15–26.
- Collins, M.S., Bashiruddin, J.B. and Alexander, D.J. 1993. Deduced amino acid sequences at the fusion protein cleavage site of Newcastle disease viruses showing variations in antigenicity and pathogenicity. Archives of Virology, 128: 363–370.
- Ideris, I. 1989. Vaccination of Village Chickens Against Newcastle Disease. Ph.D. Thesis. Universiti Pertanian Malaysia. Serdang, Malaysia. 281 p.
- Lomniczi, B. 1975. Thermostability of Newcastle disease virus strains of different virulence. Archives of Virology, 47: 249–255.
- Ru, M. (pers. comm. 1999)
- Wambura, P. (pers. comm. 2000)

Appendix 6: Questions and answers

Q: Why village chickens?

A: Village chickens are important because of their high numbers and their wide ownership by rural folk. They provide an important source of protein for families and they can be sold to generate funds.

Q: Why Newcastle disease?

A: It is the major problem identified by village chicken farmers and it can kill almost all susceptible chickens during an outbreak.

Q: Why the need to vaccinate?

A: The only way to control the disease is to vaccinate because there is no known treatment.

Q: Is vaccination free?

A: No, there will be a small charge per chicken.

Q: Can chickens be eaten or sold after vaccination?

A: Yes.

Q: How many times must chickens be vaccinated?

- A: It depends on the route of administration:
 - eye drop: every 3 to 4 months;
 - drinking water or suitable food: two initial vaccinations, then every 3 months.



Q: Will the vaccination harm chickens?

A: No, the NDV4-HR and I-2 vaccines will not harm the chickens but the birds need to be handled gently to avoid stress.

Q: Will the vaccine protect all chickens against all diseases?

A: No, the vaccine is only to prevent ND.

Q: Will all the chickens survive ND after vaccination?

A: It is not possible to guarantee 100% protection because small numbers of chickens may not respond to the vaccine especially if they are malnourished.

Q: Will the NDV4-HR and I-2 vaccines harm humans?

A: No, there are no reported cases of the vaccine harming humans.

Q: Can farmers use the vaccine by themselves?

A: Yes, after receiving basic training.



Appendix 7: Suggestions for a village poultry questionnaire

Date:

Name of interviewer:

Number of questionnaire:

Introduction: Village poultry are an important part of rural life in [name of country or region]. In order to assist village poultry farmers increase their production, we would like to know more about the major problems associated with raising village poultry. The responses given by farmers to this questionnaire will assist the Livestock Services Division to prepare an assistance package.

1. Livestock kept:

| Species | Number | Where did you get them? What breed? | Who takes care of them? |
|---------------|--------|--|-------------------------|
| Chickens | | | |
| Ducks | | | |
| Pigs | | | |
| Goats | | | |
| Sheep | | | |
| Rabbits | | | |
| Guinea pigs | | | |
| Cattle | | | |
| Other — what? | | | |



2. How many chickens do you have?

| Category | Number |
|---------------|--------|
| Adult females | |
| Adult males | |
| Growers | |
| Chicks | |

3. When was the last time that your animals received treatment?

What was the treatment?

Who gave the treatment?

4. What are the tasks associated with keeping village chickens and who is responsible for doing them?

| Person | Tasks (give water/food, build the chicken house, etc) |
|--------------|---|
| Husband | |
| Wife | |
| Son(s) | |
| Daughter(s) | |
| Other — who? | |

5. Who owns the chickens? (Mark the correct response with an "X")

| Husband | |
|--------------|--|
| Wife | |
| Son(s) | |
| Daughter(s) | |
| Other — who? | |

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6. Daily routine for village chickens:

| Activity(ies) | Time/Frequency | Who is responsible? |
|-------------------------------|----------------|---------------------|
| Shut the chickens in at night | | |
| Let chickens out in morning | | |
| Cleaning the chicken house | | |
| Give water | | |
| Give food | | |
| Other — what? | | |

7. Where do your chickens sleep?

| Location | Who made it? With what materials |
|-----------------------------|----------------------------------|
| Tree | |
| Chicken house on the ground | |
| Elevated chicken house | |
| Other — where? | |

| Parameter | Number |
|--|--------|
| 8. How many eggs on average does a hen lay per clutch? | |
| 9. How many eggs on average hatch per clutch? | |
| 10. How many chicks on average survive the first two months? | |
| 11. At what age do chickens first lay eggs? | |

12. Are you satisfied with the production of your village chickens?

Yes/No (circle the correct response)

| Why? Do you | | |
|--------------------|---------|------|
| | Chicken | Eggs |
| Sell | | |
| Eat regularly | | |
| Use for ceremonies | | |
| Other, what? | | |

13. What do you do with the chickens and eggs?

| | Chicken | Egg |
|-------------------------------------|---------|-----|
| Never sell | | |
| Sell, for how much? | | |
| Exchange for other products — what? | | |
| Where do you sell them? | | |

14. How many chickens and eggs has your family eaten over the last month? Who ate them?

| | No. eaten | Eaten by whom? |
|----------|-----------|----------------|
| Chickens | | |
| Eggs | | |

15. When and why do you sell your chickens and eggs?

16. How many chickens and eggs have you sold in the last six months?

N E W C A S T L E D I S E A S E

17. In your opinion, what are the main causes of chicken mortality?

| Birds of prey | |
|---------------|--|
| Cats and dogs | |
| Wild mammals | |
| Theft | |
| Accidents | |
| Lack of feed | |
| Diseases | |

18. How many of your birds have died in the last six months?

| | From disease | Slaughter | Other causes |
|---------------|---------------|---------------|---------------|
| | Chicks Adults | Chicks Adults | Chicks Adults |
| Chickens | | | |
| Ducks | | | |
| Other — what? | | | |

19. What do you do with your chickens when they are sick?

| Eat them | |
|---------------|--|
| Sell them | |
| Treat them | |
| Other — what? | |

| | Conventional | Traditional |
|----------------|--------------|-------------|
| Treatment | | |
| | | |
| | | |
| How to proparo | | |
| and administer | | |
| | | |
| | | |

21. Where do you get this treatment from?

| Veterinary Services | Traditional healer | |
|---------------------|--------------------|--|
| Pharmacy | NGO/Project | |
| Shop/market | Other — where? | |

22. What type of food do you give your chickens?

| Type of food | Frequency | Time of year |
|---------------------|-----------|--------------|
| Nothing | | |
| Maize | | |
| Rice | | |
| Food scraps — what? | | |
| Wheat | | |
| Maize bran | | |
| Other — what? | | |



23. Do you give water to your birds? Yes/No. If yes, where does the water come from? What type of container do you put the water in?

| Water source | Container | |
|--------------|---------------|--|
| Borehole | Plastic bowl | |
| Well | Metal bowl | |
| River/stream | Ceramic bowl | |
| Used | Tin | |
| Rainwater | Bamboo trough | |
| Other | Other | |

24. What is the local name for Newcastle disease?

25. Can you describe the symptoms of Newcastle disease?

26. In which month(s) of the year is Newcastle disease more likely to occur? Place an X in the box next to the appropriate month(s).

| January | July | |
|----------|-----------|--|
| February | August | |
| March | September | |
| April | October | |
| Мау | November | |
| June | December | |

27. Did you know that there are vaccines that can prevent Newcastle disease? Yes/No $% \left(\frac{1}{2}\right) =0$

28. Have you already participated in a Newcastle disease vaccination campaign? Yes/No

29. If yes, what were the results?

- 30. Other comments?
- 31. Personal details:

| Name: | Village: |
|---------------------------------|------------------|
| District: | Province: |
| Male/Female: | Age: |
| Ethnic group: | Local languages: |
| Who is the head of your family: | |

Thank you for answering this questionnaire. Your comments will be of great assistance to the Veterinary Services Department in its preparation of a program to improve village chicken production.





Adapted from Tizard, I. 1987. Veterinary Immunology: An Introduction. London, W.B. Saunders.

For ND vaccines:

- 1. This is unlikely with ND vaccines due to the lack of antigenic variation between strains. Mislabelling of the vaccine is also unlikely.
- 2. Counterfeit vaccine.
- 3. Vaccinating in the face of an outbreak.
- 4. Eye-dropper not correctly calibrated; bird did not drink sufficient water, reduction of vaccine titre because of inadequate storage.
- 5. Improper vaccine conservation exposed to sunlight; exposed to extremely high temperatures during transport or storage; held outside the cold chain beyond the recommended period. Vaccine mixed with an inappropriate food carrier, e.g. maize.
- 6. A very small percentage of birds will not mount an adequate immune response post vaccination.
- 7. Chicks up to the age of three weeks may have passive immunity to ND that will interfere with vaccination.
- 8. Malnourished bird; infection with immunosuppressive diseases such as Infectious Bursal Disease; certain parasitic infestations.



Appendix 9: The design and implementation of vaccine trials

Although the efficacy of thermostable ND vaccines is now proven, some occasions may still arise when vaccine trials need to be done. The objectives of the trials should be determined and the necessary preparations undertaken to ensure that the funds invested in the trials result in a successful outcome. The trial must be designed in accordance with the objectives. More information concerning laboratory techniques may be found in the companion ACIAR manual on the small-scale production of thermostable ND vaccine or via the website listed on the last page of this manual.

Pre-check

Materials:

Laboratory equipment;

Laboratory reagents, e.g. phosphate buffered saline, anticoagulant, etc.; Reliable electricity supply (working back-up generator);

Funds for:

- chickens, brooder, chicken house, feeder;
 - waterer and rations
 - personnel (including overtime for weekend work)
- additional equipment
- office supplies
- vaccine

Trained personnel — veterinary and support staff.

Designing the experiment

Routes of administration of the vaccine should correspond with what is feasible in the field:

- · chicken housing-ease of access to vaccinate chickens individually;
- · possible food carriers-not recommended but some may want to try;
- water sources-options for administering vaccine via drinking water, availability of surface water, and for dilution of the vaccine;
- economic issues-how many times a year are farmers willing to pay for the vaccine.



Vaccine Laboratory Trials

Possible objectives

- To compare the levels of protection afforded by different routes of administration and different administration regimes.
- To determine the potency of a new batch of vaccine.
- To train laboratory staff.
- To investigate possible foods suitable for use as carriers for the vaccine.
- To confirm that the vaccine strain of the ND virus provides protection against local strains (this is not a priority, as to date all vaccine ND virus strains protect against all field strains).

Materials:

- day old chicks (minimum of 10 per group, best to order in excess of final needs);
- balanced ration;
- chicken house, with brooder, feeders and waterers
 - capable of separating treatment and control groups (best kept in separate houses with a different animal attendant looking after the control group to decrease the risk of the vaccine virus spreading to the control birds)
 - chickens kept on litter not in cages;
- wing tags;
- vaccine;
- ND virus challenge strain (if challenge is to be done; not necessary if antibody titres only are being used to monitor response to vaccination);
- standard positive and negative sera;
- blood collection equipment: syringes, needles, Eppendorf tubes and cryotubes with screw tops;

- laboratory equipment: pipettors, pipette tips, reagents, reagent troughs, egg incubators, centrifuge, centrifuge tubes, microtitre plates (96 well, V bottom), antibiotics, gelatin, skim milk powder, fertile eggs, candling lamps, 70% alcohol, etc; and
- record books and pens.

Experimental protocol

One possible design for a vaccine laboratory trial where only one vaccine and one route of administration is involved is shown in Figure 20.



Figure 20: Design of laboratory trials with live ND vaccine. The double lines indicate the housing of the groups in two different units.

- Day 1 (i) place chicks in brooder
- (i) allocate chicks randomly to experimental groups (equal numbers (3 wks)(ii) per group) with surplus chicks going to control group;
 - (ii) place groups into experimental chicken house with control group in a separate building if possible;
 - (iii) tag chickens and collect serum sample from each;

- (iv) vaccinate test groups, record vaccine batch number and expiry date
 - food carrier
 - drinking water
 - eye drop
- (v) the inclusion of 'in-contact' chickens with vaccinated chickens is optional (i.e. unvaccinated chickens housed together with vaccinated chickens to check for horizontal spread of vaccine virus).
- Day 35 (i) collect serum samples from all chickens;
- (5 wks) (ii) repeat vaccinations.
- Day 49 (i) collect serum samples from all chickens (experiment may end here
- (7 wks) if no challenge is to be performed or serum antibody levels may be monitored over time to determine rate of decline of antibodies)
 - (ii) introduce challenge strain by contact with inoculated control chickens (if challenge is to be performed)
- Day 63 (i) collect serum samples from all surviving chickens
- (9 wks)
- **Post trial** (i) haemagglutination inhibition (HI) tests performed on all sera [using 4 HA units];
 - (ii) calculate geometric mean titres for each group; and
 - (iii) store sera.

At the end you should know:

- antibody titre after a single vaccination;
- antibody titre after a second vaccination;
- antibody titre that indicates resistance to challenge;
- antibody titres that will indicate chickens that have survived challenge in the field; and
- level of protection that can be expected from various vaccination regimes.

Vaccine field trial

Possible objectives:

- to confirm that the vaccine is effective under local field conditions;
- to compare different routes of administration under local field conditions;
- to determine the best intervals for re-vaccination in the trial site; and
- to train field and laboratory staff.

Personnel: Partners in trial

Village chicken farmers (female and male), village headman, local assistant, Veterinary Services staff, Extension Services staff, Laboratory staff.

Materials:

- hard cover record book for each trial site, pens, pencils;
- trial ND vaccine;
- wing tags;
- record sheets/books for records of farmers and tagged chickens;
- syringes, needles, racks, cotton wool, alcohol;
- jars with 10% formalin, jars with 50% glycerine;
- eye-droppers;
- serum tubes, Eppendorf tubes;
- · permanent markers or adhesive tape and pen; and
- coolbox and icepack for transport of sera.

Methods

i) Begin extension activities in the area well before you intend to commence the trial.

• Meet with Village Headman and village representatives to discuss the objectives of the project and to seek their cooperation. The participation of female village poultry farmers is to be encouraged.

- Ensure that villagers understand that it is a trial that is being done and that results cannot be guaranteed.
- Discuss the need to tag birds and take monthly blood samples, and the reasons for this activity.
- Explain that the vaccine will not produce 100% protection, and that the vaccine will protect against ND only.
- Check that the village chicken population is not experiencing a disease outbreak.

ii) Should the community agree to participate, select a local assistant to help with record keeping. Provide training for the local assistant and for those who wish to perform the vaccination of the birds.

iii) Allocate treatment groups to different farmer groups using a lottery draw conducted at a community meeting where representatives of all groups are present. Aim to have a minimum of 200 birds per treatment group.

iv) Decide whether or not to inform farmers which group is the control group. If farmers are not to be informed, then a mock vaccination should be undertaken in order to perform the blind trial.

v) Ensure that you allocate sufficient time to field activities, especially when starting new trials. It may be best to allow one day per treatment group initially as this would enable you to meet the farmers and work on the birds at a time convenient to the farmers.

vi) Make detailed records of:

- community meetings-comments, decisions taken, who has agreed to do what, who was present, etc.;
- vaccine usage-vaccine batch, expiry date and number of doses in the vial; how the vaccine was diluted (type of diluent used, volume of diluent, quantity of grain if used); administration route; vaccinator (Veterinary Services Department staff, farmers, etc.), vaccination dates; and
- participating farmers, number of chickens owned per farmer and tagged birds using the record sheets; endeavour to tag at least 50 birds per treatment group or 10% of birds, whichever is the greater number.
vii) Take baseline serum samples from tagged birds immediately prior to the first vaccination.

viii) Meet regularly with local trial assistant to help with any problems that may have arisen; check record books to ensure that all is in order.

ix) Ensure feedback of results to participating farmers. Results of HI tests (serum antibody titres indicating the level of protection against ND) should be discussed with farmers (Table 6).

Table 6: The immune status of birds to ND based on HI titres expressed as Log₂.

| Immune status | HI titre (Log ₂) |
|--------------------|------------------------------|
| Bird not protected | 0–2 |
| Bird protected | 3 or more |

Use visual means to convey results if necessary, e.g. to compare the HI titre of birds or different treatment groups, you can use stones, leaves, maize cobs, etc to represent one unit (Log₂). For example, one month after vaccination, the geometric mean titre (GMT) for the different treatment groups may be demonstrated by drawing on the ground with a stick to create rows for each group and indicating the level of protection by the number of stones:

| Control | • |
|----------------|---|
| Eye drop | $\bullet \bullet \bullet \bullet \bullet$ |
| Drinking water | $\bullet \bullet \bullet$ |
| Food | $\bullet \bullet$ |

x) Conduct regular community meetings. Farmers should be involved in the monitoring and evaluation of a trial. A number of indicators may be used to assess the impact of the trial, e.g. changes in flock mortality, percentage of chicks reared, changes in the number of eggs laid and their hatchability, changes in flock size, occurrence of recorded disease outbreaks, sales of chickens and eggs, changes in household income levels, demand for ND vaccine, and changes in livestock ownership patterns.

xi) Benefit:cost aspects of each of the treatment groups should be included in the community discussions as farmers will be expected to pay for further ND vaccine after the end of the trial.

xii) Always ensure that compensation of participating farmers has been discussed prior to the commencement of a trial.

xiii) Collection of post-mortem samples. It is advisable to collect samples from a representative number of birds dying in the trial area. Sample collection is detailed in Section 4 of this manual. Collect samples in formalin for histopathology and in glycerine for virus isolation if you are expecting delays in the delivery of samples to the central laboratory. It is often necessary to negotiate with farmers to obtain the samples you need.

xiv) The length of the trial will depend on the objectives. If the trial is to confirm that the vaccine is effective under field conditions, then the trial must run until there is a natural outbreak of ND. Alternatively, researchers could buy a minimum of ten birds from each treatment group, two to three months after vaccination, and take them back to the laboratory for challenge with virulent virus. If the trial seeks to determine the best intervals for re-vaccination in the area, then serology must be performed on identified birds at monthly intervals for at least one year.



Appendix 10: Sources of further information

Professor Peter Spradbrow Division of Veterinary Pathology and Anatomy The University of Queensland PO Box 125 Kenmore Q 4069 Australia Tel: +61-7-3365 5738 Fax: +61-7-3365 5600 E-mail: vppsprad@mailbox.uq.edu.au

Dr John Copland The Australian Centre for International Agricultural Research GPO Box 1571 Canberra ACT 2601 Australia Tel: +61-2-6217 0500 Fax: +61-2-6217 0501 E-mail: aciar@aciar.gov.au or copland@aciar.gov.au

Dr Robyn Alders National Veterinary Research Institute C.P. 1922 Maputo Mozambique Tel: +258-1-475171 Fax: +258-1-475172 E-mail: robyn@tropical.co.mz or robyn_alders@yahoo.co.uk

The International Network for Family Poultry Development c/o Professor Funso Sonaiya Department of Animal Science Obafemi Awolowa University Ile-Ife, Nigeria E-mail: fsonaiya@oauife.edu.ng http://www.fao.org/ag/aga/agap/Ipa/fampo1/fampo.htm Improvements in Rural Poultry in Developing Countries Website http://www.vsap.uq.edu.au/RuralPoultry

