

## ANNEX III

### DIAGNOSTIC PROCEDURES FOR THE CONFIRMATION AND DIFFERENTIAL DIAGNOSIS OF NEWCASTLE DISEASE

#### CHAPTER 4

##### **Rapid tests for detection of Newcastle-disease virus and antibodies**

##### 2. Detection of antibodies in unvaccinated birds

The majority of laboratories involved in Newcastle disease diagnosis are familiar with the haemagglutination inhibition test and the recommendation described below relate to this test for the measurement of antibodies to the virus. However, enzyme-linked immunosorbent assays (Elisa) may be successfully used to detect antibodies to the virus. It is suggested that if there is a wish to employ an Elisa test at regional laboratory level the test should be monitored by the national laboratory referred to in Annex II.

##### (a) Samples

Blood samples should be taken from all birds if the flock size is less than 20 and from 20 birds from larger flocks (this will give a 99 % probability of detecting at least one positive serum if 25 % or more of the flock is positive, regardless of flock size). The blood should be allowed to clot and serum removed for testing.

##### (b) Examination for antibodies

Individual serum samples should be tested for their ability to inhibit Newcastle disease virus haemagglutinating antigen in standard haemagglutination inhibition tests as defined in Chapter 6.

There is some debate as to whether 4 or 8 haemagglutinin units should be used for the HI test. It would appear that either is valid and the choice should be left to the discretion of the national laboratories. However, the antigen used will affect the level at which a serum is considered positive: for 4 HAU a positive serum is any showing a titre of  $2^4$  or greater and for 8 HAU a positive serum is any showing a titre of  $2^3$  or greater.