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# EVALUATION OF I<sub>2</sub> THERMOSTABLE NEWCASTLE DISEASE VACCINE ON LOCAL CHICKENS IN SELECTED DISTRICTS OF WESTERN AMHARA

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ABSTRACT: Evaluation of I<sub>2</sub> thermostable Newcastle disease vaccine was conducted in three districts of four local chicken ecotypes using survey and sera analysis from 2010 to 2011. According to the survey result conducted on 160 chicken owners, the major chicken production constraint 77.5% of the area was disease and mortality of chickens by any cause from day old to adult chicken age was 44.6%, ranging from 39.9%, 45.3%, 45.8%, and 46.3% at Melohamusit, Mecha, Farta, and Tillili, respectively from which disease related mortality was 77%. Mortality of chickens due to disease outbreak was usually higher during the beginning of the rainy season, mainly in April (43.1%), May (38.8%) and June (63.8) as first, second and third priority months, respectively and there is significant deference in disease occurrence among seasons. The overall seroprevalence of Newcastle disease in village chickens using Hemagglutination inhibition test (≥1:16) was 55.8%. However, the antibody titer response to  $I_2$  thermostable vaccine was 90.4% ranging from 83.8%, 90.9%, 91.7%, 95.1% in Mecha, Tillili, Farta and Melohamusit, respectively after one vaccination and 93% ranging from 90.9%, 93.3%, 93.8%, 96%, in Mecha, Melohamusit, Tillili and Farta, respectively after booster dose vaccination. There was no significant difference in antibody titer detected between local chicken ecotypes and/ or districts before and after vaccination. However, there was significant difference in antibody titer after  $1^{st}$  (P =0.000) and booster dose (P =0.000) vaccination. A quick survey conducted after the last vaccination showed that mortality of chickens became 8.2% which is reduced by 82% than the mortality before vaccination. In conclusion this vaccine was found very appropriate and effective in reducing village chicken mortality and morbidity, so controlling of Newcastle disease using I<sub>2</sub> thermostable vaccine could be a key to the development of village chicken production.

Key words: Hemagglutination, I2 thermostable vaccine, Newcastle disease, Village chickens

# INTRODUCTION

In Ethiopia, village chickens have been reared for a long time for different purposes in addition to meat and egg production. They have a big contribution to the country's economy. This is not because they are productive but are huge in number Alemargot (1987). According to many studies constraints which restrict the potential of village chickens in Ethiopia include; the presence of diseases of various natures, low inputs of feeding, poor management, and lack of appropriate selection and breeding practices (Alemu, 1995; Ashenafi, 2000; Tadelle and Ogle, 2001).

Newcastle disease (NCD) is among the major constraint to production of village chickens in many developing countries (Spradbrow, 1988; Alexander, 2001). It is the most important viral disease recognized in tropical countries in village poultry production systems. The disease causes great losses in most scavenger and commercial flocks (Spradbrow, 1988; Alders, 2001). Recently, the highly infectious ND is reported to have almost reached 100% mortality in some African countries (Kitalyi, 1997; Tadelle and Ogle, 2001; Tadelle and Jobre, 2004; Mazengia et al., 2009).

Newcastle disease (NCD) is a highly contagious viral disease that attacks many species of domestic and wild birds Al-Garib et al. (2003). The causal agent is the Newcastle disease virus (NCDV) which is a negative sense single stranded RNA virus belonging to the family *paramyxoviridae*. The strains of Newcastle disease virus are classified into highly virulent (velogenic), intermediate (mesogenic), or avirulent (lentogenic) based on their pathogenicity in chickens Beard and Hanson (1984). NCDV infections of poultry range from latent to rapidly fatal depending upon the pathotype of virus involved Alexander (2003). The transmission of NCDV occurs through respiratory aerosols, exposure to fecal and other excretions from infected birds, through newly introduced birds, selling and giving away sick birds and contacts with contaminated feed, water, equipment and clothing Tu et al. (1998).

NCD is mentioned as one of the disease problems in farms and backyard chickens in most parts of Ethiopia. It has many different local names in different areas and the most common one is "Fengil'" (Nasser, 1998; Ashenafi, 2000; Tadelle and Ogle, 2001), which means sudden dorsal prostration and signifies the acuteness and severity of the disease. It is possible to say that currently there are no low risk areas for NCD remaining in Ethiopia. The disease has already become endemic in village poultry population and thus it recurs every year inflicting heavy losses (Tadelle and Jobre, 2004; Mazengia et al., 2009). It is estimated that annual outbreaks of NCD kill 70–80% of unvaccinated village chickens Spradbrow (1995). Outbreaks are unpredictable and discourage villagers from paying proper attention to the husbandry and welfare of their chickens.

Vaccination is the most important method of disease control particularly to decrease mortality from NCD. Vaccination results in a quite significant increase in chick survival from 30% to 60% Udo (1997). Conventional vaccines are unsuitable for sustained use in village chicken production system because of their cost, large dose presentation and thermolability. The importance of village chickens to the rural and peri-urban poor in developing countries is not contested. Another universal truth is that these flocks are less productive, and cost-effective remedies should be available. Thermostable/heat stable/ Newcastle disease vaccines, suitably applied, have proved effective in many trials under laboratory conditions and in villages Spradbrow (2011).

So the objective of this study is to introduce  $I_2$  thermostable vaccines in village chickens and evaluate the effectiveness of  $I_2$  thermostable Newcastle disease vaccine and to reduce the mortality of village chickens due to Newcastle disease and to increase the awareness of households in particular women about NCD and the control options.

# MATERIALS AND METHODS

#### Study areas

These studies were conducted at three districts (Tillili /Guagusa-Shikudad/, Mecha and Farta districts), located in the North Western part of the country. Local chicken ecotypes collected from these study districts showed relatively better egg and meat production potential when managed intensively at Andassa Livestock Research Center and were recommended for further improvement by Halima (2007). Therefore these districts were selected purposively.

#### **Questionnaire survey**

A questionnaire survey was conducted on 160 respondents before the beginning of vaccination to assess the prevailing chicken production system of village chickens and major constraints of the system in selected districts. And after the fifth vaccination a quick survey was conducted on 122 respondents to assess the effect of I<sub>2</sub> thermostable vaccine in reducing the morbidity and mortality of local chickens of the study area.

#### **Vaccination of animals**

Before the beginning of vaccination Couple training\_was given on poultry disease and health management to farmers of the area who have an experience of rearing poultry. After the training vaccination site were selected and practical training were given to selected farmers or community vaccinators on how to vaccinate chickens and how to handle the vaccine.

Chickens were vaccinated with I<sub>2</sub> thermostable Newcastle disease vaccine produced by National Veterinary Institute (NVI), Debre zeit Ethiopia. The vaccines were administered once, every 3 months. Vaccination was given to the whole chicken population in selected villages by community vaccinators/selected farmer/.

Vaccines were diluted using the formula:-

Amount of water required in ml = <u>1ml X number of doses that the vial contain</u>

Number of drops per 1ml of the dropper/ syringe

Vaccination was given through ocular route and the vaccine costs only 0.15 Ethiopian cents per head/bird. Administration of the eye drop to the bird was done with the dropper in a vertical position to make sure that drops of a uniform size are produced. Chickens were vaccinated five times.

#### Serum collection

About 2-3ml blood was collected once before vaccination and twice after 3 weeks of each vaccination regime from the wing vein of chicks of all age groups with 5ml disposable syringe/ non-heparinized vaccutainer tube of 5ml and 23G (32mm) needle. The syringe/ tube containing the blood was kept at room temperature overnight in slanting position until the blood clot and the blood was centrifuged with hematocrite centrifuge, then the serum was transferred into a sterile plain tube. The tubes were labeled and stored at -20°C until analysis.

#### Hemagglutination-Inhibition (HAI)

HAI test was done according to the procedures of OIE (2004). The test was conducted at the National Veterinary Institute (NVI), Debre zeit-Ethiopia. The test was carried out by running two fold dilutions of equal volumes ( $25\mu$ I) of Phosphate Buffered Saline (PBS) and test serum ( $25\mu$ I) in U-bottomed micro titer plates. 4 Hemagglutination units of (HAU) the viral antigen of LaSota (I<sub>2</sub>) strain obtained from France was added to each well and the plates were left at room temperature for a minimum of 30 minutes. Finally  $25\mu$ I of 1% (v/v) chicken RBCs collected from four chickens older than 3-weeks and serologically negative to NCD was added to each well and, after gentle mixing, the RBCs were allowed to settle for about 40 minutes at room temperature. The HI titer was read from the highest dilution of serum causing complete inhibition of 4 HAU of antigen. Those wells that showed sedimentation of RBC as the control wells (containing only 25µl RBCs and 5µl PBS) were considered as inhibition. A titer greater than or equal to 1:16 was taken as positive.

#### Data management and statistical analysis

Basic data entry and handling were done using SPSS software version 16. Descriptive statistics and chi-square tests were employed to summarize the data. Tests were considered significant at p < 0.05.

# RESULTS

According to the survey result the prevailing chicken production constraint of the area was disease (77.5%) (Table 1), predator (80.6%) and Feed shortage (82.5%) as first, second and third priority problem, respectively. The average mortality rate of chickens by any cause from day old to adult age was 44.6%, ranging from 39.9%, 45.3%, 45.8%, and 46.3% at Melohamusit, Mecha, Farta, and Tillili, respectively from which disease related mortality was 77%. Most of respondents witness high occurrence of chicken diseases, among which 93% of them says Newcastle disease locally known as "Fengil" or "Meyaz", was the major and economically important constraint for the existing chicken production system of the study district. According to interviewed chicken owners, mortality of birds due to disease outbreaks was usually higher during the beginning of the rainy season, mainly in April (43.1%), May (38.8%) and June (63.8) as first, second and third priority months, respectively (Table 2). There is significant difference in disease occurrence among seasons (P =0.000, df=7,  $x^2$ =189.1).

The overall seroprevalence of Newcastle disease in village chickens using Hemagglutination inhibition test ( $\geq$ 1:16) was 55.8% (Table 3). However, the antibody titer of Newcastle disease in response to I<sub>2</sub> thermostable vaccine was 90.4% (255/282) and 93% (265/285) after 1<sup>st</sup> vaccination and booster dose vaccination respectively (Table 4 and figure 1).

Table 1 - First priority village chicken production constraints in the study district								
	Tillili		Mecha		Farta		Total	
Constraints	Frequency	%	Frequency	%	Frequency	%	Frequency	%
Disease	29	72.5	39	97.5	56	93.3	124	77.5
Feed shortage	2	5.0	0	0.0	2	3.3	4	2.5
Predator problem	7	17.5	1	2.5	20	33.3	28	17.5
Market Problem	0	0.0	0	0.0	2	3.3	2	1.3
Lack of land	1	2.5	0	0.0	0	0.0	1	0.6
Lack of capital	1	2.5	0	0.0	0	0.0	1	0.6

Table 2 - Major seasons of the year when Newcastle disease appear as an outbreak Months Frequency Percent Valid Percent March 15 9.4 9.4 April 69 43.1 43.1% 17 10.6 10.6 May 39 24.4 24.4 June July 13 8.1 8.1 August 2 1.2 1.2 2 1.2 October 1.2 September 3 1.9 1.9 Total 160 100.0 100.0

	Seroprevalence of Newcastle disease			
Local chicken ecotypes	Positive (≥1:16 )	<b>Negative (&lt;1:16 )</b>		
	N (%)	N (%)		
Tillili	20 (50%)	20 (50%)		
Aecha	34 (53.1%)	30 (46.9%)		
Farta	33 (60%)	22 (40%)		
Melohamusit	34 (58.6%)	24 (41.4%)		
Total	121 (55.8%)	96 (44.2%)		

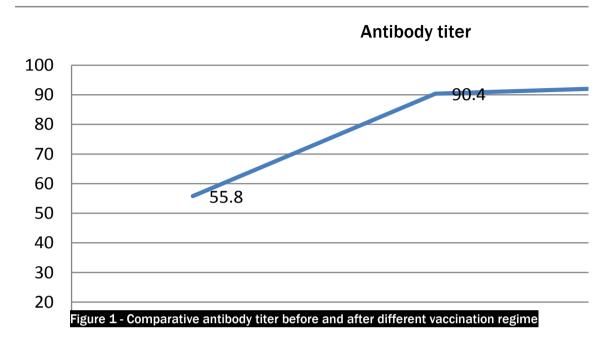
# Table 4 - Antibody titer of Newcastle disease in four local chicken ecotypes in response to the first vaccination

Local chicken ecotypes	Antibody titer after first vaccination				
	Positive (≥1:16)	Negative (<1:16)			
	n (%)	n (%)			
Tillili	50 (90.9%)	5 (9.1%)			
Mecha	62 (83.8%)	12 (16.2%)			
Farta	66 (91.7%)	6 (8.3%)			
Melohamusit	77 (95.1%)	4 (4.9%)			
Total	255 (90.4%)	27 (9.6%)			



There was no significant difference in antibody titer detected between local chicken ecotypes and/or districts before vaccination (P >0.05, df = 3,  $x^2 = 1.3$ ), after first vaccination (P > 0.05, df = 3,  $x^2 = 5.9$ ) and after booster dose vaccination (P>0.05, df = 3,  $x^2 = 1.5$ ). But There was significant difference in antibody titer after 1<sup>st</sup> (P=0.000, df = 1,  $x^2=184.3$ ) and booster dose vaccination (P = 0.000, df = 1,  $x^2=210.6$ ).

In addition, according to a quick survey after vaccination the mean morbidity and mortality of chickens was 13.9% and 8.2% respectively, this shows that mortality of chickens was reduced by 82% as compared to the mortality of chickens before vaccination.



#### DISCUSSION

The mortality rate of chicken in this study from day-old to adult chicken age was (44.6%), from which disease related mortality is 77%. This finding is in line with the previous reports from both Ethiopia (Alemargot, 1987; Mazengia and Eshetie, 2008) and other countries Farooq (2001) the current disease related mortality from day old to adult chicken age is estimated between 20% and 80%.

On the other hand, the overall seroprevalence of Newcastle disease in village chickens in this study was 55.8%. This finding is in line with the previous reports in Ethiopia by Mazengia et al. (2010). However, this finding is higher than the previous reports in central high lands of Ethiopia by Ashenafi (2000) and the reports of Zeleke et al. (2005) in Rift valley areas of Ethiopia. Similarly higher seroprevalence Newcastle disease was reported by Ezeokoli et al. (1984) who recorded 62.9% seroprevalence in Nigeria.

The overall population with protective antibody titer ( $\geq 1:16$ ) after first and booster dose vaccination was 90.4% and 93% in the study districts, respectively. Which is higher than the reports by Mazengia et al. (2009) in day old-chicks in which the overall population with protective antibody titer ( $\geq 1:8$ ) was (71.1%) in the study districts, this could be l<sub>2</sub> thermostable vaccine may have a higher capacity of inducing antibody production than the conventional vaccines, or may also be due to challenges in keeping the cold chains of conventional vaccines during vaccination. And this finding is concurrent with the epidemic theory which suggests that if 70% of the population is immune, the disease outbreak is unlikely to occur because there are not enough susceptible to propagate an epidemic (Thrusfield, 1995; Young et al., 2001).

#### CONCLUSION AND RECOMMENDATION

The major poultry production constraint and causes of mortality in the study area was Newcastle disease locally known as "Fengil" which mostly occurs as an outbreak during the beginning of the rainy season in April and may. I<sub>2</sub> thermostable vaccine have similar response for all type of local chicken Ecotypes and can reach a protective level at one vaccination regime without the need for booster dose vaccination. Despite chickens were vaccinated and vaccines were handled by community vaccinators, I<sub>2</sub> thermostable vaccine is highly suitable and effective in reducing village chicken mortality and morbidity and control of Newcastle disease using I<sub>2</sub> thermostable vaccine is the key to the development of village chicken production. Wider use of this vaccine needs further training of farmers and the adoption of suitable extension methods.

So, emphasis should be given on extensive use of I<sub>2</sub> thermostable vaccine in village chickens in reducing the mortality and improving their productivity and Vaccination programs should be continual and sustainable but if it is not possible chickens should be vaccinated at least once every year before April which may reduce heavy chicken losses. Wider use of this vaccine should be practiced through establishment of community vaccinators and further training of farmers.

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