COMPARATIVE ASSESSMENT OF THE REPRODUCTIVE PERFORMANCE AND NATURAL DISEASE RESISTANCE OF THREE PHENOTYPES OF VILLAGE CHICKENS IN ADAMAWA STATE

 \mathbf{BY}

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(PhD/AS/06/0169)

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IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF DEGREE OF DOCTOR OF PHILOSOPHY IN ANIMAL PRODUCTION AND MANAGEMENT

DECLARATION

I hereby declare that, this Thesis was written by me and it is a record of my own
research work. It has not been presented in any previous application for any degree
anywhere. All sources of information shown in the text are listed in the reference and all
efforts made by others are duly recognized and acknowledged.

Dr. BOBBO, Goniwa Aminu	Date

DEDICATION

This work is dedicated to my late mother Zainab - Abu and my late father Alhaji Bobbo Goniwa

APPROVAL PAGE

This Thesis entitled "comparative assessment of the reproductive performance and natural disease resistance of three phenotypes of village chickens in Adamawa State" by Aminu Goniwa Bobbo with registration no (PhD/AS/06/0169) meets the regulations governing the award of Degree of Doctor of Philosophy in Animal Production and Management, Modibbo Adama University of Technology, Yola and is approved for its contribution to knowledge and literacy presentation.

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ABSTRACT

The research was designed to study egg quality traits, fertility and hatchability traits and susceptibility of three phenotypes of village chickens to Newcastle Disease Virus (Hertz 33/56). The study was carried out at Modibbo Adama University, Yola. Three phenotypes of Sixteen (16) hens and Two (2) cockerels each (48 breeder hens and 6 cocks) were selected and assigned randomly (Expt i and ii) and Forty five (45) village chickens, made up of fifteen (15) each of Frizzle, Naked neck and Smooth feathered types were infected with NDV (Expt iii) were used for the study. A total of two hundred and seventy and three hundred and twenty four fertile eggs were collected for egg quality traits and for hatching respectively. Micrometer screw gauge, Varnier caliper and Microtitre plates were used. Egg width, albumin height and width significantly (p<0.001) differ among the phenotypes. Egg weight, yolk height, yolk index and haugh unit had a significant (p<0.01) effect on all the phenotype. Egg length, egg index and shell weight had significant (p<0.05) effect on the phenotype. Frizzle feathered consistently produced heavier, longer, wider egg and albumin and heavier shell. Similarly frizzle naked neck produced heavier albumin and yolk height and haugh unit. Furthermore naked neck produced higher egg width, egg and yolk index and yolk height. Frizzle and Smooth feathered chickens and its associate produced significant positive and negative correlations in all the egg quality traits and naked neck and its crosses produced significant correlation in all the egg quality traits studied. All the hatchability parameters were significantly different except hatchability on fertile egg and dead in shell. Hatching egg weight had significant (P<0.01) effect on all the phenotypes. Fertility, hatchability on set eggs, dead embryo, normal chicks, abnormal chicks and average chick weight had significant (P<0.05) effect on all the phenotypes. Frizzle and Naked neck and its crosses were superior in most of the hatchability parameters studied. Smooth and its crosses were superior in fertility, percent normal chicks and average chick weight. A morbidity rate of 100% and mortality rates of 58.3%, 41.7% and 75% for the frizzle, naked neck and smooth was observed respectively. The mean antibody titre of all the phenotypes before the infection was zero and naked neck started developing protective antibodies on day 7 post infection (PI). All the phenotypes, with the exception of the naked neck in the intraocular group, exhibited highest Geometric mean titre (GMT) of NDV HI antibodies on day 28 PI through both routes. The GMT in the control group remained low throughout the experiment. It could therefore be concluded that crossing between Frizzle and Naked neck produced better egg quality traits, Smooth and Frizzle feathered chicken produced better fertility and hatchability traits and the naked neck group and their crosses were the most resistant phenotype and the smooth group and their crosses were the least resistant and poor sero-converts to NDV infection. It could therefore be recommended that Frizzle naked neck be considered for table eggs, Smooth Frizzle to be considered for hatching programmes and rearing naked neck group to avoid killer disease.

Key words: Phenotypes, Frizzle, Naked neck, Smooth, Fertility, Hatchability, Newcastle disease virus (NDV), Geometric mean titre and Haemagglutination inhibition.

TABLE OF CONTENT

COV	ER PAGE	i
TITL	E PAGE	ii
DECI	LARATION iii	
DEDI	ICATION iv	
APPR	ROVAL PAGE	V
ACK	NOWLEDGEMENT vi	
ABST	TRACT vii	
TABI viii	LES OF CONTENT	
LIST xiii	OF TABLES	
LIST	OF APPENDICES xv	
CHAI	PTER ONE	
1.0	INTRODUCTION	1
1.1	Background of the study	1
1.2	Statements of problem	5
1.3	Justification of the study	6
1.4	Objectives of the study	6
1.5	Hypothesis	6
CHAPTER TWO		
2.0	LITERATURE REVIEW	8
2.1	Poultry Development in Nigeria	8

2.2	The Village Chickens	9
2.2.1	Naked neck	9
2.2.2	Frizzle 10	
2.3	Fertility and Hatchability 11	
2.4	Growths Traits 13	
2.5	Egg Production Traits 13	
2.6	Feed Consumption and Efficiency 14	
2.7	Egg Quality 14	
2.7.1	Internal egg quality 15	
2.7.1.1	Yolk quality 16	
2.7.1.2	Yolk colour 16	
2.7.1.3	Yolk firmness 18	
2.7.1.4	Yolk texture 18	
2.7.1.5	Albumin consistency 18	
2.7.1.6	Genetics (internal and external) 18	
2.7.1.7	Age and storage of the eggs 19	
2.7.1.8	Vanadium 19	

2.7.1.9 Appearance of albumin 20 2.7.2 External eggs quality 2.7.2.1 *Egg shell integrity* 20 2.7.2.2 *Egg size* 21 2.7.2.3 Age of the birds 2.7.2.4 Stress 22 2.7.2.5 Elevated environmental temperature 22 2.7.2.6 *Nutrition and water quality* 2.7.2.7 Disease of an egg 24 2.7.2.8 Management 25 2.7.2.9 *Egg shape* 26 2.7.2.10 Egg shell color 2.7.2.11Chemotherapeutic agent 26 2.7.212 Shell cleanliness 27 2.7.2.13 Nutrition and/or birds health 2.8 Newcastle Disease (ND)

27

2.8.1	History and distribution of ND 28
2.8.2	Etiology 28
2.8.3	Host range and reservoir 29
2.8.4	Clinical signs and gross lesions 29
2.8.5	Laboratory diagnosis 31
2.8.6	Influence of age and sex 31
2.8.7	Spread of ND in village chickens 32
2.8.8	Serological prevalence of ND in village chickens 33
2.8.9	Prevention and control of ND 33
2.8.10	Differential diagnosis 34
2.10.1	Geometric mean titre (GMT) 34
2.10.2	Vaccination titre 36
2.10.2.	1 The Immune response 36
2.10.2.	2 Alternatives available 37
2.10.2.	3 Sampling methods 38
2.10.2.	4 Standardization 38

2.10.2.5 Reporting results 38

2.10.2. *Interpretation* 39

CHAPTER THREE

3.0 MATERIALS AND METHODS 41

3.1 Study Area

3.1.1 Experimental chickens 41

3.1.2 Experimental design 42

3.2.0 Experiment i: Determination of internal and external egg quality characteristics 42

- **3.3.0** Experiment ii: Determination of hatchability parameters 43
- 3.4.0 Rearing of day-old chicks/brooding $\Delta \Delta$
- **3.5.0** Experiment iii: Susceptibility of three phenotypes to experimental newcastle disease virus hertz 33/56 45
- **3.5.1** Experimental birds 45
- **3.5.2** Experimental design 45
- **3.5.2**.1 *Serum samples* 45
- **3.5.2.2** Chickens red blood cells 45

3.5.1.3	46
3.5.1.4	Challenge 46
3.5.1.5	Serological test 46
3.6.1	Procedure for collecting blood from chickens 47
3.6.2	Precaution for collecting blood from chickens 47
3.7.0	Statistical analysis 47
СНАР	TER FOUR
4.0	RESULTS 49
4.1	Experiment i 49
4.2	Effect of Hen of different Phenotypes on Egg Quality 49
4.2.1	Correlation for egg quality traits for hen 52
4.2.1.1	Frizzle 52
4.2.1.2	2. Naked neck 56
4.2.1.3	S Smooth 57
4.3	Experiment ii 63
4.3.1	Effect of hen of different phenotypes on hatchability traits 63
4.3.2	Correlation of hatchability traits for hen 65

4.3.2.1 Naked neck 65 4.3.2.2 Smooth 65 4.3.2.3 *Frizzle* 70 4.4 Experimental iii 70 4.4.1 Experimental infection of three phenotypes of local chickens **CHAPTER FIVE** 5.0 **DISCUSSION** 78 **Egg Quality Traits** 5.1 5.1.1 Correlation of different egg quality traits 5.2 Effects of hen of different phenotype on hatchability traits -5.2.1 Correlation among hatchability traits 84 5.3 Experimental infection of three phenotypes of local chickens **CHAPTER SIX** 6.0 SUMMARY, CONCLUSION, RECOMMENDATION AND CONTRIBUTION TO KNOWLEDGE 87

6.1

6.2

Summary

Conclusion

87

88

6.3 Recommendations

80

6.4 Contribution to knowledge

89

REFERENCES

90

APPENDICES

104

LIST OF TABLES

- Table 1: Egg quality traits of nine phenotypes of local Chickens 50
- Table 2: Phenotypic Correlation between external and internal egg quality

traits for Frizzle

53

- Table 3: Phenotypic Correlation between internal egg quality traits for Frizzle 54
- Table 4: Phenotypic Correlation between external egg quality traits for Frizzle 55
- Table 5: Phenotypic Correlation between external and internal egg quality

traits	for	naked	neck
uans	101	Hancu	HCCK

58

Table 6: Phenotypic Correlation between internal egg quality traits for naked neck 59

Table 7: Phenotypic Correlation between external egg quality traits for naked neck 60

Table 8: Phenotypic Correlation between external and internal

egg quality traits for smooth feathered 61

Table 9: Phenotypic Correlation between internal egg quality traits

for smooth feathered

62

Table 10: Phenotypic Correlation between external egg quality

traits for smooth feathered

66

Table 11: Hatchability traits of nine phenotypes of local Chickens. 67

Table 12: The Correlation of different hatchability traits among Hens of Frizzle 68

Table 13: The Correlation of different hatchability traits among

Hens of Naked neck

69

Table 14: The Correlation of different hatchability traits

among Hens of Smooth feathered 71

Table 15: Results of the Morbidity and Mortality rates among different

Phenotypes of village chickens challenged with Hertz 33/56 NDV 72

Table 16: Sex and Route susceptibility of infected and non infected local chickens
73

Table 17: Geometric mean titre (GMT) of NDV HI antibodies of three phenotypes of village chickens infected with Hertz 33/56 NDV using intraocular route

75

Table 18: Geometric mean titre (GMT) of NDV HI antibodies of three

Phenotypes of local chickens infected with Hertz 33/56 NDV

using intranasal route
76

Table 19: Geometric mean titre (GMT) of NDV HI antibodies of control group 77

LIST OF APPENDICES

APPENDIX 1:External egg characteristics 104

APPENDIX 2: Internal eggs characteristics

106

APPENDIX3: Fertility and hatchability traits

109

APPENDIX 4: Pelletized Chicks feed 25kg/bag

112

APPENDIX 5: Pelletized Grower feed 25kg/bag

113

APPENDIX6: Pelletized Layer feed 25kg/bag

114

APPENDIX 7: Challenged Virus Information

115

APPENDIX 8: Publication Certificates

ABBREVIATIONS, DEFINATIONS AND SYMBOLS

OIE Office of International Epizootics

GMT Geometric Mean Titre

NRC National Research Council

FAO Food and Agricultural Organization

HA Haemagglutination

HI Haemagglutination Inhibition

NDV Newcastle Disease Virus

ND Newcastle Disease

CRD Chronic Respiratory Disease

ILT Infectious Laryngotrichitis

HU Haugh Unit

F Frizzle feathered chicken

S Smooth feathered chicken

N Normal feathered chicken

Nk Naked neck

M In contact chickens

CDC Centre for Disease Control

EPZ Egg Production Federation

NZFSA Food Standard Australia and Newzealand

Nacl Sodium chloride

Anova Analysis of variance

% Percent

et al Others

CHAPTER ONE

INTRODUCTION

1.1 Background of the Study

Free-ranging / Scavenging / Local / Indigenous / Runners / Backyard / Native / Bush /Rural / Family / Traditional / Village chickens (Gallus gallus domesticus), are the commonest type of chickens raised in rural, peri- urban and urban household in Nigeria. They are non descriptive and heterogeneous type of birds that differ in size, shape, colour and production according to their genetic constitution. The Local chicken population constitutes 80% of the total world chicken population. In Africa, there are over 800 million chickens and of these, more than 80% are Local chickens (Gueye, 1998). In Nigeria, it constitutes about 92% of 134 million poultry birds (Manchang et al., 2004). Of the 92%, Sonaiya and Olori (1989) reported that 75% are the smooth multicolored type,12% had frizzle feathered, 6% had naked neck and 4% are the dwarf chickens. They are economically, nutritionally and culturally important in many countries (Ekue et al., 2002) and contribute substantially to annual meat and egg production (up to 90%) for family consumption and for sale (Nwapku et al., 1999; Fayeye et al., 2005). Local chickens meat and eggs taste are preferred over those of exotic chickens (Robert, 1999). They are also manifesting a great deal of variation which is due to genetic and environmental factors (Olori and Sonaiya, 1992). Local chicken are well adapted to hash environmental conditions such as hot or cold weather, rain and periodic feed shortage. The chickens produce good eggs, hard egg shell, high dressing percentages and good meat flavor. They, therefore offer a good source of animal protein to most rural people (Johnston, 1990). The smallness of the carcass preclude for the refrigeration as in the case with sheep, goat or cattle meat. Chicken particularly with white colour have been used for many years for therapeutic purposes by traditional healer (Oh, 1987). Similarly they also provide, manure and keep the surrounding environment clean through scavenging on insects, weeds and waste that lie on the ground. They are also used as gift during traditional ceremonies. The management system of rural poultry is primarily of free type which is characterized by lack of feeding, minimal housing and little or no veterinary/husbandry services. These production systems are often called low input low out put (Pandey, 1992). The low out put is caused by range of factors, such as sub-optimal management, lack of supplementary feed, low genetic potential and disease prevalence (Pandey, 1992). Despite the constraints above, poultry continue to play a big role in rural as well as national economy in developing countries (FAO, 2000). Women and children are generally in-charge of rural poultry husbandry. The birds scavenge in the vicinity of the homestead, during, the dry whether. They may be given sorghum, millet, maize, broken grains or other waste products as supplementary feed. Breeding under the free ranging system of management is random, cock and hens mate in uncontrolled manner (Williams, 1990). It is usually the most aggressive, strong and dominant cock, which sires most offspring in the neighborhood (Williams, 1990).

The most striking feature in relation to village poultry production is low genetic potential of the birds and these include low hatchability traits, poor egg laying; slow growth rate and late attainment of sexual maturity as well as small body size (Mjojo,1983). Egg size/weight is usually below 40g, while total egg production is usually less than 120 eggs per annum. Scavenging hen may lay 30-60 eggs/hen/year (Safalaoh *et al.*, 1996). While industrialized battery hens lay up to 300 eggs annually. Egg quality parameters such as egg length, diameter, albumen height and diameter and yolk height etc. have also been reported to be lower in other breeds (Yeasmin *et al.*, 1992). Market age of more than 1kg are attained at more than Twenty weeks of age (Safalaoh *et al.*, 1996).1.6 kg at sixteen weeks of age (Bay-Peterson, 1991) and 1.9kg at 20 weeks of age (Kang *et al.*,1993). Furthermore, malnutrition, environment, management and societal pressure interact in multiple ways to influence the ultimate productivity level, the overall mortality rate and the quality of the final product (Calneck, 1998).

Fertility and hatchability are the major determinants of profitability in hatchery industry (Peters *et al.*, 2008). The two parameters appear to be very important as far

as reproduction is concerned. Hatchability is a complex age dependant traits. It comprises of several sub traits which are susceptible to genetic and environmental factors arising from various sources(Wolc and Olori, 2009). There are many factors which influence hatchability of eggs and these include storage time, fertility, temperature, relative humidity, ventilation, position of the egg, turning of the egg and candling. Similarly feed variation also affects hatchability (Mussaddeq et al, 2002). Zelleke et al. (2005) showed that both sexes are responsible for poor fertility in Rhode Island Red but, the female is responsible for the poor hatchability and this poor performance is due to the egg weight loss during incubation. Other factors that affect hatchability of a breeding hen include genetic constitution of the embryo, disease, egg size age and shell quality (King'ori, 2011). Egg weight, fertility, hatchability and late dead in germs varied greatly between feed regimes (Lariviere et al, 2009). Similarly the fertility of an egg is affected by factors directly related to the laying hen such as her ability to mate successfully, store sperm, ovulate and finally produce a suitable environment for the formation and development of embryo (Brillard, 2003). Fertility also depends on the ability of cock to mate successfully, quantity and quality of semen deposited (Wilson et al, 1979; Brillard, 2003)

Eggs and meat are amongst the most nutritious foods and eggs are rated with milk as one of the best protein foods rich in iron and vitamins (Oluyemi and Roberts 2000). The significance of the egg as a protein source for the nourishment of humans led to the consumers demand for some qualities in this nutrients (Uluocak *et al.*, 1999). Therefore monitoring and evaluation of external and internal quality of chicken eggs is important in production economy and consumers' preferences for better quality of eggs. Eggs quality has been defined by Stadlemen (1977), as the characteristics of eggs that affect its acceptability to the consumers and is more important price contributing factor in table and hatching eggs. Therefore, the economic success of a laying flock solely depends on the total number of quality eggs produced. Egg quality is a general term which refers to several standards which define both external and internal quality, such as egg weight, egg length, width, egg index, shell weight, shell thickness, albumin height, albumin width, yolk height, yolk index, haugh unit. Approximately 7-8% of the total eggs are broken

through the transfer eggs from the production to the consumer. And this results in serious economical problems both for the producers, dealers and consumers (Hamilton, 1982). Kul and Seker, (2004) reported that egg weight and egg index are determinants of egg resistance to cracking and are considered very important traits when eggs are packed to container. The free-ranging village chicken industry is characterized by low production, high mortality and losses are due to diseases and predation (Kitalyi, 1998). The disease prevalence constitutes major production constraint in developing countries (Sonaiya, 1990; Awan et al., 1994). Newcastle disease (ND) has been ranked as the number one killer disease of free-ranging local chickens. It is the most important limiting factor in local chickens farming in most developing countries of the world and a serious threat to intensively reared chickens (Echeonwu et al., 2008). The disease is endemic in Nigeria and continues to be responsible for high mortality and morbidity among village and exotic poultry (Saidu et al., 1998; Baba et al., 1998). It is currently being ranked as the most economically important disease of chickens in Nigeria and elsewhere and currently controlled by routine vaccination (Ezeokoli, 1984). High prevalence rate of 60.8% in local fowls, 57.2% in layers and 37.7% in broilers were reported by (Sadiq et al., 2011). Similarly 54% prevalence rate was recorded in Nigerian local chickens (Baba et al., 1998). Furthermore Njagi et al. (2010) reported sex seroprevalence of 11.5% for adult female and 8.3% adult male and reported seroprevalence of 6.5% and 3.6% grower female and males respectively. Local chickens are generally believed to be resistant to endemic diseases and stressful environment. This ability to endure with disease and stress is what is responsible for their higher survival under village condition when compared to other strains (Msoffe, 2003). Several research findings indicated that village chickens are not resistant to all endemic diseases (Minga et al., 1989; Lin and Lee, 1996; Okoye and Aba-Adulugba, 1998).

Other diseases reported in local chickens; Gumboro disease (Sonaiya, 1990; Baba *et al.*, 2004); fowl pox and coccidiosis (Sonaiya, 1990); fowl typhoid and fowl cholera; infectious coryza and chronic respiratory disease (CRD) (Melewas, 1989) and helminthosis (Permin *et al.*, 1997). Nevertheless, local chickens survive amidst all these constraints in spite of low inputs from the owners (Crawford, 1984)

Natural resistance to Newcastle and infectious bursal disease has been reported in Egyptian breeds of native chickens and also differences within and between ecotypes in immunocompetence for endemic disease like NDV has also been investigated (Hassan et al., 2004). Similarly, natural resistance to Salmonellae infections have been reported in some lines of White Leghorn chickens (Bumstead and Barrow, 1993). Village chickens are believed to be resistant to many common diseases, partly because little attention is paid to disease control measures among this group of poultry (Melewas, 1989). Previous studies have shown that local chickens appear to be more susceptible to some diseases than exotic commercial types (Okoye and Aba-Adulugba, 1998). Testing of the disease resistance potential can be by infecting the host directly with the virulent strain (Okoye and Aba-Adulugba, 1998) or by contact (Akinoluwa et al., 2012). So far there are very limited research findings and dearth of information on egg quality, reproductive performance and the disease resistance potentials of different phenotypes of village chickens in Nigeria. It is important to determine the disease resistance potentials of our village chickens as a means of using their genetic potentials to control endemic infectious diseases. This study is strictly designed to compare and assess the egg quality characteristics, reproductive performance and compare the susceptibility of three phenotypes of village chickens to Newcastle disease virus Hertz 33/56 strain and assess the utilization of the available genetic resources in the improvement of village chickens production and consumer patronage and confidence in Nigeria.

1.2 Statement of the Problem

The village chickens are characterized by slow growth rate, small body size, low egg production, low fertility and hatchability (Mjojo, 1983); low internal/external performance characteristics (Yesmin *et al.*, 1992). Local chickens laid 64 eggs per year reared in intensive system (Sazzad, 1986). But Khan (1983) found that indigenous fowl lay small eggs weighing 30 - 35g with an average egg production of 50 - 80 eggs per year, and on proper selection program the egg production of the native hens could be increased up to 135 eggs per year per bird and are vulnerable to different types of disease conditions. Their management system has made it

difficult to determine their production and productivity as well as their relative resistance to infectious diseases. The reproductive performance and disease resistance of different phenotypes of village chickens have not been investigated in this environment.

1.3 Justification of the Study

The research findings will display the egg quality, fertility, hatchability and susceptibility to different phenotypes of local chickens in Adamawa state. It will equip hatcheries, farmers, local women and poultry keepers on which phenotype to be adapted for hatching, table eggs and rearing in order to produce quality Day-old chicks and to avoid seasonal outbreaks of deadly disease like Newcastle disease virus in our homes. Furthermore findings will also assist Veterinarians in designing prevention and control programmes of Newcastle disease in Nigeria.

1.4 Aims and Objectives of the Study.

The Research was designed to achieve the following:

- i. Determine the internal and external egg quality characteristics of different phenotypes of Local chickens.
- ii. Determine the fertility and hatchability traits of the different phenotypes of Local chickens.
- iii. Compare the susceptibility of three phenotypes of Local chickens to Hertz 33/56 Newcastle disease virus.
- iv. Determine sex susceptibility among the three phenotypes of Local chickens to Hertz 33/56 Newcastle disease virus.
- v. Determine phenotype and inoculation route that gives higher seroconverts.

1.5 Hypothesis

The village chickens are well adapted to this environment and have economically sustainable productive potentials and disease resistance traits when compared to exotic/commercial chickens. These characteristics of village chickens vary among the different phenotypes of this group of birds.

CHAPTER TWO

LITERATURE RIVIEW

2.1 Poultry Development in Nigeria

Zoologically, the chickens belong to the genus Gallus of the family Phasianidae. The domestic chicken is called simply Gallus domesticus. The wild ancestors of the domestic chickens probably originated in South East Asia. Four species of wild jungle fowl are Gallus gallus (red jungle fowl), Gallus varius (the black or green jungle fowl). The red jungle fowl, Gallus gallus, has the widest distribution of the wild species and may well be the chief ancestors of modern chickens. The poultry industry has shown a tremendous growth in the last twenty five years in Nigeria; largely because of a complete and fundamental change in view point. Instead of keeping poultry chickens as a happy or a sideline for pleasure and some incidental profit, thousands of flock owners have taken up the poultry enterprise on their farms as an economic unit, a means of livelihood, a source of income by which to raise and educate family and acquire a certain degree of economic independence. Instead of keeping chickens, they have made the chickens keep them. Ideally, poultry keeping developed into a commercial enterprise involving thousands and millions of birds in Nigeria. Large poultry units replaced small ones, while more efficient strains of birds, balanced feeds, intensive housing and better poultry equipments came into use (Oluyemi and Roberts, 1979). Commercialization of poultry keeping is a recent development in the humid tropical countries of the world in contrast with the temperate countries, where the industry is less capitalized, consists of smaller units and depends more on manual labor that led to lower performance of poultry products consumption in the tropical areas (Oluyemi and Roberts, 1979). Therefore, there is no doubt whatsoever about the significant role of the poultry industry in bridging, the protein deficiency gap in the nutrition of Nigerians and indeed the tropical world at large. The components of poultry feeds include Carbohydrates, Protein, Minerals, Vitamins and diets with varying

crude protein and Metabolizable energy content and majority of management practices require instituting biosecurity measures

2.2 The Village Chickens.

The indigenous fowls have been variously referred to as the African Chickens, Bush Chickens/ scavenging chickens/Local chickens/ Back yard chickens/ Runner Chickens etc. However, distinct local varieties have been reported in Nigeria, Egypt, Cameroon, Burkina Faso, Morocco and Sudan (Gueye, 1998). Indigenous chickens tend to be robust and are well adapted to hard environmental conditions such as hot or cold weather, rain and periodic feed shortage. These birds have many advantages such as good egg and meat flavor, hard egg shells, high dressing percentages, and especially low cost with little special care required for production. However, they have some disadvantages arising from the fact that they suffer high rearing mortality (Trial, 1962), have slow growth rates and poor egg quality Layers attain sexual maturity late. Such birds can be improved genetically through selective breeding or by cross breeding with exotic stocks depending on the management conditions (Katule, 1992).

2.2.1 Naked neck gene

The naked neck gene is a single autosomal dominant gene, Na. The gene is incompletely dominant with Na/na+ birds showing an isolated tuft of feathers on the ventral side of the neck above the crop, while Na/Na birds either lack this tuft or it is reduced to just a few pinfeathers or small feathers. The resulting bare skin becomes reddish, particularly in males as they approach sexual maturity (Somes, 1990).

The relevance of the naked neck gene in the tropics lies in its association to heat tolerance. The reduction in feather coverage of 30-40% in naked neck birds facilitates better heat dissipation and improved thermoregulation resulting to a better relative heat tolerance under hot climates. Merat (1990) has reviewed several favorable effects of this gene. He states that in high temperatures, near 30°C or higher, homozygous *Na/Na* or heterozygous *Na/na*+ naked neck birds had a better

weight gain than normal na+/na+ birds. There was also an improvement in the carcass yield, laying rate, mean egg weight, eggshell strength and egg mass for the heterozygous genotype. However, he noted an increase in embryonic mortality, mostly during the last stages before hatching that was observed in the Na/Na and Na/na+ birds.

Other favourable effects of this gene in high temperatures includes higher breast weight, superior growth rate, better feed conversion ratio and carcass traits (Yalcin et al., 1997, Patra et al., 2002), reduced effect of high ambient temperatures on fertility (Ladjali et al., 1995), lowest incidence of the pathologies such as cloacal cysts, Ascites, prolapse, Mareks disease, coccidiosis, osteodystrophy and Salmonellosis (Fraga et al., 1999) and resistant to sudden death and Ascites syndrome (Gonzales et al., 1998). Further, combining the naked neck allele with other tropically relevant alleles such as frizzling has been shown to result to a favorable additive effect to various productive parameters (Yunis and Cahaner, 1999).

2.2.2 Frizzle gene

Frizzling is caused by a single incompletely dominant autosomal gene, F, restricted by an autosomal recessive modifier, mf (Hutt, 1949). The mode of action and the effect of this gene are well reviewed in the literature (Somes 1990). In unmodified homozygous frizzled birds, the rachises of all feathers are extremely recurved. These feathers are easily broken and therefore the birds appear quite bare. The modifying gene lessens the extreme aspects of the homozygotes so that they appear less woolly. Unmodified heterozygote has the feather shafts and barbs of contour feathers recurved, to a much less extent than the homozygotes. This is modified so that some birds are almost indistinguishable from the wild type. The action of the F gene has been shown to be localized in the feather follicle and does not result from a metabolic disorder. The effect of this gene on production has been shown to be favourable by an increase in egg number and egg mass, alongside reducing mortality under hot conditions (Merat, 1990). The only unfavorable effect of the gene found in the literature is mentioned by Somes (1990) who said that

hatchability of eggs laid by heterozygous frizzles is subnormal and it is still lower in eggs from homozygotes.

2.3 Fertility and Hatchability

A lot of research works have been performed in different parts of the world concerning artificial incubation of eggs. Fertility and hatchability have been reported to be influenced by genetic, physiological, social and environmental factors, male and female ratio, age, nutritional status, preferential mating, lighting, sperm quality, rate of production, quality of eggs etc. Singh et al. (1983) observed egg fertility and hatchability in White Leghorn and White Plymouth Rock. The fertility was 91.3 and 91.5% respectively. Embryonic mortality was 36.9 and 45.2% and hatchability was 63.0 and 54.7%. Ghany et al., (1969) incubated 4537 Fayoumi, 1971 Baladi and 2876 Rhode Island Red eggs and candled them individually at 7 and 14 days of age for dead embryos. Early embryonic mortality was 10% in all 3 breeds at 3rd day of incubation. In all breeds, a second increase occurred on the 12th and 14th day (4% and 6% respectively) and a third peak (20%) on 21st day. Dev et al. (1993) reported four batches of chicken eggs each comprising 2 replicates of 100, were incubated in each of the following incubators (1) James way force - draught electric incubators; (2) Doyal force draught incubators; (3) Kerosene incubator; (4) Kerosene incubator with the heater replaced by a 100w electric light bulb; (5) Rice husk incubator. They found hatchability of fertile eggs average 84.6, 73.6, 62.3, 61.5 and 62.5% in the five types of incubator, respectively and the percentage of normal chicks was 97.4, 95.3, 92.6 and 90.1 respectively; embryonic mortality was 4.6, 4.9, 15.2, 16.5 and 17.1% respectively and dead in shell 10.2, 21, 22.3, 21.1 and 20.4% respectively.

Card and Nesheim (1978) stated that hatchability, embryonic mortality, dead in shell, and normal chick hatched depend on storage of egg, position, angle of setting, humidity, temperature and O₂ and CO₂ ratio. The number of females per male, social dominance, light and sperm quality can play a significant role in the process of fertilization (Champion and Rood, 1958); Kalita (1994) studied on 1800 eggs from 850 White Leghorn and 564 Rhode Island Red fowls. Both breeds

hatchability was higher for medium size eggs 51 - 55g) than for small (45 - 50g) and large (56 - 60g) eggs. Brah and Sindhu (1981) observed embryonic mortality at different stages in two strains of White Leghorn and their crosses. Mortality up to 17 days of setting was 19.8% in pure breeds and 9.3% in cross breeds. Mortality from 18 to 22 days was 47% and 45.8% and post-piping mortality was 33.2% and 44.9% respectively.

Khalil (1960) recorded embryonic mortality in Fayoumi eggs of 6%, 3% and 14% during three weeks of incubation respectively. Hafez and Kamar (1955) observed 7.91% mortality in Fayoumi eggs during the first week, while that of the last week was 25%. Fertility and hatchability are the major determinants of profitability in poultry industry (Peters et al., 2008). The two parameters appear to be very important as far as parent stocks are kept to produce chicks. Hatchability is a complex age dependant traits. There are several factors which influence hatchability of eggs like the pre-incubation storage time, fertility and incubation condition such as temperature, humidity, ventilation, egg turning and candling. Variation in feed composition resulted in variation in hatchability (Mussaddeg et al., 2002). Zelleke et al., (2005) concluded that both sexes are responsible for the poor fertility in Rode Island Red, but female is responsible for poor hatchability and performance is mainly due to greater weight egg loss during incubation. Apart from these, other factors which can have considerable influence on hatchability include, nutrition of the breeding hens, genetic constitution of the embryo, disease, egg size, age and shell quality (King'ori, 2011). Egg weight, fertility, hatchability and late embryonic mortality varied greatly with feed regimes (Lariviere et al., 2009). Similarly the fertility of an egg is affected by the factors directly related to the laying hen such as her ability to mate successfully, store sperm, ovulate an egg cell, and finally produce a suitable environment for the formation and development of the embryo (Brillard, 2003). Fertility also depends on the ability of the cock to mate successfully, quantity and quality of semen deposited (Wilson et al., 1979; Brillard, 2003). Fertility and hatchability traits can be classified into those that are associated with the egg, e.g. fertility and hatchability of egg and those directly associated with the chicken, e.g. age at sexual maturity, which is defined as the age from hatch to the day of first egg. Kicka *et al.* (1978), obtained the highest (92.3%) fertility in Fayoumi (Fy) x Rhode Island Red (RIR), compared to Fayoumi (89%) and RIR (77.9%). Hatchability of fertile eggs as reported by many authors is influenced by genetic factors, storage, temperature, care, age, quality of eggs, age of bird, season, nutrition, pre-incubation, warming and humidity etc (Laxi,1964).

2.4 Growth Traits

Fast growing chickens have an advantage in that they reach the market weight early hence reducing production costs. It has been shown that crossbreeding the indigenous chickens with the exotic breeds improves daily gains and feed intake (Omeje and Nwosu, 1988). Under a good nutritional environment, Katule and Mgheni, (1990) reported similar growth rates of indigenous and exotic breeds to 12 weeks of age. In the study, the merits of the indigenous chickens were supported by the fact that the backcross to the indigenous strain grew faster than the exotic backcross. Various ecotypes of indigenous chickens produce meat from exotic breeds of same age and management (Obanu *et al.*, 1984). Improvement in body size and growth of indigenous chickens is important from economic considerations bordering on the need to increase chickens (Ibe, 1995). With well-designed selection programmes, this can easily be achieved in the indigenous chickens because of the appreciable additive genetic variance observed in these breeds (Olori, 1994).

2.5 Egg Production Traits

Egg number and weight are major traits of economic interest in commercial production. Egg size determines to a large extent the prices received in any market. Soltan (1993) compared the egg quality traits of the selected Sinai fowls with those of the Fayoumi indigenous breeds and found that the differences in these genotypes for these traits were not significant. In Nigeria, Omeje and Nwosu, (1988) compared egg production traits of the indigenous chickens, the Gold-Link exotic breed and crosses between them. Significant differences were observed in the egg weights of the different genotypes. The Gold-Link exotic breed and the Gold-link

sired backcross laid heavier eggs compared with the first cross, F2 and back cross populations that were sired by the indigenous chickens.

Local chickens laid 64 eggs per year reared in intensive system (Sazzad, 1986). But Khan (1983) found that indigenous fowl lay small eggs weighing 30 - 35g with an average egg production of 50 - 80 eggs per year, and on a proper selection program the egg production of the native hens could be increased up to 135 eggs per year per bird.

2.6 Feed Consumption and Efficiency

Hague *et al.* (1999) found that daily feed intake per bird averaged 64.3 and 55.2g and feed conversion ratio 5.7 and 4.9 for Fayoumi and RIR respectively in a group of 3 male and 20 females during the week 6-17 weeks.

2.7 Egg Quality

Quality has been defined by Kramer (1951) as the properties of any given food that have an influence or rejection of this food by the consumer. Egg quality is the general term which refers to general standards which define both internal and external quality. External quality is focused on shell cleanliness, texture and shape, where as the internal quality refers to an egg white (Albumin) cleanliness and viscosity, size of the air shell, yolk shape and yolk strength. Egg is one of the most complete and versatile foods available. It consists of approximately 10% shell, 58% white and 32% yolk. The average egg provides approximately 313 kilojoules of energy, of which 80% comes from the yolk. The nutritive content of an average large egg (containing 50g of edible egg) contained the following: 6.3g protein, 0.6 g carbohydrates, 5.0 g fat (0.21 g cholesterol). Egg protein is of high quality and is easily digestible. Almost all of the fat in egg is found in the yolk and is easily digestible. Eggs contain every vitamin except vitamin C. They are particularly high in vitamins A, D and B₁₂ and also contain B₁ and riboflavin. Provided that laying hens are supplemented according to the optimum vitamin nutrition concept, eggs are important vehicle to complete the essential vitamin supply to the human population. Eggs are also good source of iron and phosphorus and also supply

calcium, copper, iodine, magnesium, manganese, potassium, sodium, zinc, chlorine and sulphur. All these minerals are present as organic chelates, highly bioavailable, in the edible part of the egg.

2.7. 1 Internal egg quality

Internal egg quality involves functional, aesthetic and microbiological properties of the egg, yolk and albumin. Egg white is formed by four components. Firstly, the chalziferos layer or chalazae, immediately surround the yolk, accounting for 3% of the white .Next is the inner thin layer, which surrounds the chalazae and accounts for 17% of the white. Third is the firm or thick layer, which provides an envelope or jacket that holds the inner thin white and the yolk. It adheres to the shell membrane at each end of the egg and accounts for 57% of the albumin. Finally, the outer thin layer lies just inside the shell membrane, except where the thick white is attached to the shell and accounts for 23% of the egg white (USDA, 2000)

Unlike external (shell) quality, internal quality of the egg begins to decline as soon as the egg is laid. Thus although factors associated with the management and nutrition of the hen do play a role in internal egg quality, egg handling and storage practices do have a significant impact on the equality of the egg reaching the consumer. Similarly, although the shell provides a unique "package" for the distribution of the egg contents to the consumer, it is in fact the internal quality of the egg that is most important to the consumer. These aspects of internal quality are significantly more difficult to observe or evaluate in the intact egg, even with the use of candling.

In addition to the, obvious, nutritional quality of the egg, internal egg quality is extremely important because of its many functional and aesthetic properties. For example, eggs are used as thickening agents in custards and puddings, egg whites are used as smoothing agents to give icings a desirable texture and egg yolks add colour and richness to food (Koelkebeck, 1999).

The EPF Code of Practice (EPF and NZFSA, 2002) listed a total of nine internal defects and these include: Blood spots, Meat sports, Watery whites, Pale yolks,

mottled yolks and discolored, discolored whites rotten eggs, Roundworms in eggs and off odors and flavors. To minimize egg quality problems two things are important: frequent egg collection, mainly in hot months and rapid storage in the cool room. The best results are obtained at the temperature of 10°C. There are six main factors affecting internal quality: disease, egg age, temperature, humidity, handling and storage.

2.7.1.2 Yolk quality

Egg yolk from newly laid eggs is round and firm. As the egg gets older, the yolk absorbs water from the egg white, increasing its size. This produces an enlargement and weakness of the vitalline, the yolk looks flat and show spots. Yolk quality is determined by the colour, texture, firmness and smell of the yolk (Jacob *et al.*, 2000).

2.7.1.3 *Yolk color*

Although yolk color is a key factor in any consumer survey relating to egg quality (Jacob *et al.*, 2000), consumer preferences for yolk colour are highly subjective and vary widely from country to country.

The primary determinant of yolk colour is the xanthophylls (plant pigment) content of the diet consumed. It is possible to manipulate the yolk colour of eggs by the addition of natural or synthetic xanthophylls to layer hen feeds. This ability to readily manipulate egg yolk colour can be an advantage in meeting market demands. However, the ease with which yolk colour can be manipulated can lead to unwanted colour changes. For example, the inclusion of higher than recommended levels or incorrect ratios of pigments can lead to orange-red yolks and diphenyl-para-phenylenediamine (antioxidant), has been reported to cause excessive deposition of pigments in the egg yolk (Coutts and Wilson, 1990).

The inclusion of more than 5 % cottonseed meal in a layer diet will result in olive or salmon colored yolks, while the inclusion of certain weeds or weed seeds may results in green yolks (Coutts and Wilson, 1990; Beyer, 2005). Similarly,

inadvertent omission of xanthophylls from the diet will lead to pale yolks. Both inadequate mixing of the diet as well as excessive mixing of the diet will also result in a heterogeneous feed, and subsequent variation in the amount of xanthophylls consumed by each hen in the flock, This will result in egg yolk colour not being uniform throughout the flock.

Pale yolks can result from any factor which alters or prevents the absorption of pigments from the diet or the deposition of these pigments in the yolk. These factors could include; worms (Coutts and Wilson, 1990); any factor which inhibits liver function, subsequent lipids metabolism and deposition of pigment in the yolk, for example, Mycotoxicosis caused by aflatoxin B1 (Zaghini *et al.*, 2005) and Coccidiosis, although this is rare in adult hens.

Mottled yolks (with many pale spots and blotches which vary in colour size and shape), occur when the contents of the albumen and yolk mix as a result of degeneration and increase permeability of the vitelline membrane (Jacob *et al.*, 2000). Factors affecting mottling were reviewed in detail by Cunningham and Sanford (1974). Dietary factors which may cause mottled yolks include; the presence of nicarbazin (an anticoccidial agent) in the feed has shown by numerous authors to cause mottling (Cunningham and Sanford, 1974; Jones *et al.*, 1990).

Deworming drugs such as Phenothiazine (Coutts and Wilson, 1990), Dibutyltin dialaurate (Coutts and Wilson, 1990; Jacob *et al.*, 2000) and Piperazine (Coutts and Wilson, 1990; Jacob *et al.*, 2000) has been shown to cause mottling of yolk.

Gossypol from cotton seed meal (Jacob *et al.*, 2000), certain antioxidants such as gallic acid (from grapes, tea and oak bark) and tannic acid (Coutts and Wilson, 1990), or tannins from grains such as sorghum (Jacob *et al.*, 2000), calcium deficient diets (Jacob et al., 2000). Storage time and temperature has also been shown to affect the degree of egg yolk mottling (Coutts and Wilson, 1990; Jacob *et al.*, 2000). Jones (2006) stated that as the internal temperature of the egg increases above 7°C, the protein structures of the thick albumen and vitelline membrane breakdown faster. As the membrane degenerates during storage, water enters the

yolk causing mottling and after prolonged storage, albumen proteins also enter the yolk increasing the severity of mottling (Jacob *et al.*, 2000). In order to reduce the rate of breakdown of the vitelline membrane, eggs should be collected regularly; reducing the time they are exposed to higher environmental temperatures and contaminants, and stored at temperatures of $7 - 13^{\circ}$ C and humidity of 50 - 60 %. In their review, Cunningham and Sanford (1974) also identified hen age, oil coating of eggs and movement of eggs as possible factors affecting mottling of eggs.

2.7.1.4 Yolk firmness:

The yolk of a freshly laid egg is round and firm (Jacob *et al.*, 2000). However, as the egg ages and the vitalline membrane degenerates, water from the albumen moves into the yolk and gives the yolk a flattened shape.

2.7.1.5 *Yolk texture:*

Rubbery yolks may be caused by severe chilling or freezing of intact eggs, the consumption of crude cottonseed oil or the seeds of some weeds (Jacob *et al.*, 2000).

2.7.1.6 *Albumin consistency:*

Albumin quality is measured in terms of Haugh Units (HU) calculated from the height of the albumin and the weight of the egg (Coutts and Wilson, 1990). However most eggs leaving the farm should be between 75 and 85 HU (Coutts and Wilson, 1990) Albumin consistency is influenced by: *Age of the hen:* HU will decrease with increasing bird age value, with HU decreasing by around 1.5 to 2 units (Coutts and Wilson, 1990) for each month in lay. Doyon *et al.* (1986), cited by Jones, (2006) stated that HU decreases at a fairly constant rate of 0.0458 units per day of lay as the hen ages.

2.7.1.7 Genetics (internal and external egg quality)

Strain of bird has also been shown to play a role in albumin consistency, with some strains consistently producing eggs with thin albumin. Curtis *et al.* (1985) reported

that brown egg layers produced eggs with higher HU, while other authors (Williams, (1992) reported that HU values were more variable within the brown egg layers compared with those that lay white eggs. High producing birds tend to lay eggs with relatively lower amounts of thick albumin and, although this can be influenced by selective breeding, egg numbers are usually considered more important. Clunies *et al.* (1992) found that hens laying thick shelled eggs retained more dietary calcium than those laying thin-shelled eggs. Although there was no difference in egg production between thick and thin shell layers, both egg and shell weight were greater for the thick shelled eggs. The production of eggs with calcium deposits on the shell (or pimples) is thought to be hereditary (Coutts and Wilson, 1990).

2.7.1.8 Age and storage of thee egg:

Raji *et al.* (2009) concluded that egg weight, albumin and yolk height, Haugh unit, and yolk indices decreased with increase in storage time, while albumin and yolk width increased. Egg length and width were not affected by storage time and methods. He therefore established that the quality of an egg is affected by the methods and length of storage. Egg kept at high temperature 40°C deteriorated in quality fast and were not fit for human consumption after two weeks. Refrigerated eggs were able to maintain their quality comparable to fresh eggs. Oiling of eggs also maintained egg quality to some extent, but oiling is not a replacement for refrigeration. It may however serve for 28 days where refrigeration facilities are not available and egg must be stored. In hot dry climate, where ambient temperature can reach 40 - 45 °C, eggs should not be stored at room temperature, for more than one week before consumption.

2.7.1.9 *Vanadium:*

Henry and Miles (2001) reviewed the effects of vanadium on poultry performance. They noted that poorer albumin quality has been reported from laying hens consuming as little as 6 ppm of vanadium (Sell *et al.*, 1982). Later reports, Sell *et al.* (1986) showed that the interior quality of eggs decreased in two strains of laying

hens fed 3 or 6 ppm added vanadium. Duyck *et al.* (1990) fed laying hens 10 ppm of vanadium for 30 days. HU from these hens averaged 71 HU after one day of storage 62 °F (16.6°C) and 60 % Relative Humidity) and 64 after seven days of storage. This was in contrast to the average of 82 and 74 HU after one and seven days storage respectively, observed for hens fed the control diet. Henry and Miles (2001) reported that the negative effects of vanadium may be overcome by feeding cottonseed meal, ascorbic acid, vitamin E or carotene, although this is dose dependant.

2.7.1.10 Appearance of albumin:

Normal albumin is transparent, with a slightly yellow green colour. Discoloration of the albumin may occur if the eggs are stored for an extended time period in poor conditions, with the albumin becoming much yellower (Coutts and Wilson, 1990). Cyclopropene fatty acids from cottonseed meal (Coutts and Wilson, 1990; Beyer, 2005) and the certain weed seeds (Coutts and Wilson, 1990) can cause albumin to turn pink after storage. Green whites are caused by excesses of riboflavin (vitamin B2) in the diet (Coutts and Wilson, 1990). Cloudy whites may be caused by the oiling of eggs within six hours of lay (Beyer, 2005)

2.7.2 External eggs quality

The Egg Producers Federation of New Zealand (Inc) (EPF) Code of Practice (EPF and NZFSA, 2002) listed 14 possible eggs shell defects, although these can be grouped into five main categories, defects associated with egg shell integrity, texture, shape, colour and cleanliness.

2.7.2.1 Egg shell integrity

Approximately 7-8% of the total eggs are broken through the transfer of eggs from the production to the consumer. And this results in serious economical problems both for the producers, dealers and consumers (Hamilton, 1982). To maintain consistency good shell quality throughout the life of the hen, it is necessary to

implement a total quality management programmed through out the production cycle.

Defects considered under the category of egg shell integrity include gross cracks, hairline cracks, star cracks and thin shelled or shell-less eggs. As cracked eggs cannot be made available for retail sale in New Zealand Food Standards Australia New Zealand, (FSANZ, 2006), high numbers of cracked eggs will have a negative impact on the profitability of any egg producer. One of the most obvious reasons for egg shell cracks (including gross cracks, hairline cracks and star cracks) is mechanical damage caused either by the birds themselves or as a result of poor management practices, such as infrequent collection of eggs, rough handling and poor design and or maintenance of the cage floor.

Egg shell strength ultimately affects the soundness of the shell, with weaker shelled eggs more prone to cracks and breakages and subsequently microbial contamination. Shell strength can be affected by a wide range of factors.

2.7.2.2 *Egg size:*

Smaller eggs have stronger shells than larger ones, as hens have a finite capacity to deposit calcium in the shell and as a result, the same amount of calcium is spread over a larger area (Butcher and Miles, 2003a).

2.7.2.3 *Age of bird:*

Older birds tend to lay bigger eggs and have a higher egg output, which impacts on shell strength as described earlier (Butcher and Miles, 2003a). Very young birds with immature shell glands may produce shell-less eggs or eggs with very thin shells. Delaying the onset of sexual maturity by one to two weeks will prevent this (Coutts and Wilson, 1990). As birds age increases, the intensity of pigment decreases. This may be due to decreasing production of pigment or increased surface area over which available pigment is distributed (Butcher and Miles, 2003b). As with shell soundness, young birds with immature shell glands may produce misshapen eggs (Coutts and Wilson, 1990).

2.7.2.4 Stress:

A single stress or disturbance to a flock of laying hens can be enough to desynchronize the process of egg formation for several days, during which time, a number of different egg quality faults may be seen (MAFF, undated). For example; any factor which results in oviposition prior to completion of shell deposition will result in soft or thin-shelled eggs. Activities which create disturbances in and around the layer shed should be minimized (Coutts and Wilson, 1990). If an egg is retained in the shell gland, any subsequent egg laid may spend less time than normal in the shell gland, resulting in insufficient shell deposition and a soft shelled or shell-less egg (MAFF, undated).

Solomon (1991) noted that once an imperfect mammillary layer has formed (as a result of stress experienced before the egg reaches the shell gland), subsequent layers are disorganized and thin or soft shelled eggs are a common phenomenon after stress.

Epinephrine, a stress hormone, will cause a delay in oviposition and cessation of shell gland cuticle formation, which can cause pale shelled eggs to be produced. Stressors may include, amongst others, high cage density, loud noise and handling (Butcher and Miles, 2003b). Body checked eggs, marked by grooves and ridges, occur when the shell of the egg break in the shell gland, during the formation process (i.e. 10 - 14 hours before the egg is laid). Although the damage can be partly repaired, a bulge forms around the egg (Coutts and Wilson, 1990; Solomon, 1991). Flat sided eggs occur where two eggs are in the shell gland at the same time (Solomon, 1991). Boefects may be caused by overcrowding, frights or other disturbances and poor lighting patterns (Koelkebeck, 1999). Jones (2006) stated that proper handling can reduce the incidence of body checks.

2.7.2.5 Elevated environmental temperature:

High (above 25°C) environmental or shed temperatures may affect the feed (and therefore calcium) intake of the bird, thus resulting in a decreased availability of calcium for shell deposition. As well as decreasing feed intake, laying hens will try

to overcome heat stress by panting (Koelkebeck, 1999). However, this causes a decrease in the amount of carbon dioxide (CO₂) in the hens' blood, a condition known as respiratory alkalosis (Koelkebeck, 1999). As egg shells are made up of 95 % calcium carbonate (CaCO₃), this decrease in blood CO₂ levels, combined with an increase in blood pH and a subsequent decrease in Ca2+ ions for shell formation leads to an increase in the number of thin or soft shelled eggs produced. Arima *et al.* (1976) found that the egg quality of older hens was more severely affected by increased temperature than younger hens.

2.7.2.6 Nutrition and water quality:

The provision of adequate dietary minerals and vitamins is essential for good eggshell quality. Similarly, as water quality varies from country to country and region, the role of drinking water in mineral and trace element supply should not be overlooked.

Calcium and phosphorous are essential macro minerals with calcium forming a significant component of the shell and phosphorous playing an important role in skeletal calcium deposition and subsequent availability of calcium for egg shell formation during the dark period (Boorman *et al.*, 1989).

Coetzee (2002), investigated the effect of calcium levels in drinking water on shell integrity in South African laying hens and demonstrated that birds supplied an additional 200 mg of calcium per litre of drinking water laid eggs with mean shell strength of 42.6 ± 9.0 . This was in comparison to those receiving un-supplemented water, whose eggs had mean shell strength of 38.9 ± 7.0 . Feeding hens high levels of calcium may interfere with the availability of other minerals (NRC, 1994). Similarly, feeding high levels of dietary phosphorous have also been shown to have a negative effect on eggshell quality (Keshavarz and Austic, 1990). It remains unclear whether excess plasma phosphorous interferes with the mobilization of skeletal phosphorous reserves or the shelling process itself (Ahmad and Balander, 2004) or if elevated levels of phosphorous increases calcium excretion (Keshavarz and Austic, 1990). Keshavarz and Austic (1990) also examined the interaction of

phosphorus and chloride and the role of chloride in egg shell integrity. Phosphorous, elevated dietary levels of chloride resulted in decreased eggshell quality and lower levels of blood acid-base indicators. This is supported by the work of Balnave *et al.* (1989) who observed an increase in shell defects with no changes in egg production, egg weight, feed or water intake, blood-acid base levels and electrolyte levels for birds provided with 2000 mg of sodium chloride (NaCl) per litre drinking water. This was in comparison to those hens provided with water with 600 mg NaCl per litre. Birds receiving 2000 mg of NaCl per litre drinking water also had an increased incidence of shell-less eggs. Egg shell defects persisted after the sodium chloride was removed from the drinking water.

Finally, care should be paid to the mixing of the diet. Thorough mixing of the feed is essential if each bird in the flock is to receive a similar amount of any given nutrient. This is particularly true for layer hen diets which frequently contain raw materials with a wide range of different densities.

2.7.2.7 Diseases of an egg

Diseases such as certain strains of Egg Drop Syndrome, Infectious Bronchitis Newcastle Disease and Infectious Laryngotracheitis (Jacob *et al.*, 2000) can all cause a decrease in albumin consistency. Similarly viruses, which affect the mucus membranes of the respiratory and reproductive tract, such as NDV and IBV, not only cause a decrease in egg production, but also cause the shell to become abnormally thin and pale (Beyer, 2005). Infectious bronchitis (IB), which is a corona virus attacks the mucus membranes of the respiratory and reproductive tracts (Jones, 2006), may result in egg defects. These include pale shelled eggs, and eggs with poor shell structure and integrity (Jones, 2006). Similarly, birds affected by Egg drop syndrome (EDS), caused by an adenovirus, initially produce pale eggs, quickly followed by thin soft-shelled or shell-less eggs (McFerran and Adair, 2003). Jewers (1990) reported that thin rubbery shells which break more readily than normal have been observed during field outbreaks of Ochratoxicosis. Similarly, in an outbreak of *T-2* toxicosis, egg breakages increased from a normal 3 % to 15 % with a further 18 % of eggs broken in transit to customers (Jewers,

1990). Zaghini et al. (2005) reported that birds fed diets containing 2.5 ppm of aflatoxin B1 had lower egg shell weights than those fed the control diet or diets supplemented with Mannanoligosaccharides. As the albumen of the egg and surrounding membranes provide the structure on which the egg shell is deposited, if the albumen quality is very poor, there is no sound foundation on which to build the shell (MAFF, undated). As a result, those diseases which result in poor albumen quality often cause an increase in the number of misshapen eggs. Examples of these are Infectious Bronchitis (Jones, 2006), Egg Drop Syndrome (Coutts and Wilson, 1990) and certain strains of Newcastle Disease (ND) or avian influenza (AI) (Coutts and Wilson, 1990). Gross cracks, Star cracks, Rough shells or Sandpaper shells, Pimples, Pinholes and Mottled or glassy-shells, Cage marks, Stained eggs, Fly marks, Fungus or Mildew on shells, Body-checked eggs, Flat sided eggs, misshapen eggs, Thin shell, are all egg shell defects associated with egg shell texture. These defects are frequently a result of bird age, but may also be caused by other factors (Coutts and Wilson, 1990). Certain diseases such as Infectious Bronchitis, infectious laryngotracheitis (ILT) (Coutts and Wilson, 1990; Beyer, 2005) and avian encephalomyelitis (AE) (Coutts and Wilson, 1990) have been implicated in the production of rough or "sandpaper" eggshells.

2.7.2.8 Management:

Good management practices will help reduce the number of dirty eggs. These practices include frequent collection of eggs, as well as regular replacement of litter material in nest boxes or regular maintenance and cleaning of cage floors and roll out trays. Whilst less common, fly stains, water stains and grease or oil stains may occur, and can be prevented by good shed and equipment maintenance or management (Coutts and Wilson, 1990).

Overcrowding of birds, changes in the lighting programme, poor shed ventilation and inadequate water supply can contribute to increased incidence of shell defects associated with egg texture (Coutts and Wilson, 1990).

2.7.2.9 *Egg shape*:

Misshapen eggs have a shape which differs from the smooth normal shape (for example, flat sided eggs and body checked eggs).

2.7.2.10 Egg shell color

The color of an egg shell is determined primarily by the genetics of the hen, with white feathered hens laying white eggs and brown feathered hens laying brown eggs (Jacob *et al.*, 2000). During the process of egg shell formation in brown egg layers, the epithelial cells lining the surface of the shell gland synthesize and accumulate pigments, such as biliverdin-IX, its zinc chelate and protoporphyrin-IX (Butcher and Miles, 2003). In the final three to four hours of shell formation these pigments are transferred to the viscous, protein rich cuticle. The quantity of pigment in the cuticle determines the colour of the egg (Butcher and Miles, 2003). As the cuticle is deposited onto the eggshell at the same time that shell deposition reaches a plateau (approximately 90 minutes prior to oviposition), pigment distribution is not uniform throughout the shell, with very little pigment contained in the shell itself (Butcher and Miles, 2003). Thus, any factor which causes a disruption, either in the ability of the epithelial cells to synthesize pigment or in the deposition of the cuticle, will affect the colour of the egg shell. These factors include:

2.7.2.11 Chemotherapeutic agents:

Certain drugs have been shown to affect egg shell colour. For example, nicarbazin (anticoccidial) fed at a level of 5 mg per day can result in the production of pale eggs within 24 hours, while higher doses can lead to complete depigmentation (Butcher and Miles, 2003b). Chlortetracycline (600 - 800 ppm) may also result in yellow egg shells (Beyer, 2005).

2.7.2.12 Shell cleanliness:

Cleanliness is probably the easiest aspect of egg shell quality to control, and good management plays an important role. Most eggs are clean when laid and subsequently become contaminated with faecal material or other contaminants. Defects listed in the EPF Code of Practice (EPF and NZFSA, 2002), which fall into this category, include cage marks, stained eggs and fly marks. Although fungus or mildew on shells is listed as a defect, it is only likely to occur in poor conditions (Coutts and Wilson, 1990).

2.7.2.13 Nutrition and/birds' health:

Any factor which causes diarrhea in the birds, (for example high dietary salt levels), will also result in an increase in the number of dirty eggs collected. Blood smears on eggs can be minimized by good pullet management, including weight for age, lighting and beak trimming if necessary (Coutts and Wilson, 1990).

2.9 Newcastle disease

Newcastle disease (NDV) is caused by viruses of avian paramyxovirus type 1 (APMV 1) serotype of the genus Avulavirus belonging to the family paramyxoviridae. There are nine serotype of the avian paramyxoviridae designated as APMV1 APMV 9. It is a highly contagious and fatal disease affecting both domestics and wild birds (Adu *et al.*, 1990). It is a serious viral disease which is present all over the world. In many tropical and subtropical countries virulent strains of Newcastle disease virus (NDV) is endemic (Spradbrow, 1990). In most developing countries, NDV is the most important infectious disease, affecting village chickens (Aini *et al.*, 1990). The disease is a recognized problem of village chickens (Durojaye and Adene 1988). The disease has been reported to be characterized by a dramatic decline in egg production both quality and quantity (Copland, 1987). It causes great economic losses

The disease is endemic in Nigeria (Saidu et al., 1998; Baba et al., 1998). The first documented outbreak of the disease occurred in Ibadan in 1952 (Hill et al.,

1953). Since then the disease has been the most economically important disease of chickens in Nigeria (Ezeokoli *et al.*, 1984). Currently, the disease is controlled by routine vaccination (Ezeokoli *et al.*, 1984). Newcastle continues to be a serious economic threat to the poultry industry resulting in increased morbidity and mortality rates and loss of egg for both breeding and human consumption (Abdu *et al.*, 1992).

In Nigeria, the disease continues to be responsible for high mortality and morbidity among village and exotics poultry (Baba *et al.*, 1998). 52.3% prevalence of ND was reported in Maiduguri, Borno state, North Eastern, Nigeria and peak outbreaks was recorded in November, December and January (Sadiq *et al.*, 2011).

2.8.1 History and distribution of ND

In 1926, ND was recognized in two widely separated locations: Java, Indonesia and in Newcastle-upon-Tyne, England (Alexander, 1997). In 1927, ND was reported as Ranichet in India, and in 1930 it appeared on a number of farms in Australia. Today ND is present all over the world. Newcastle disease (ND) is caused by a group of closely related viruses that form the avian *Paramyxoviruses* type 1 (APMV-1) serotype.

There are 3 theories on the origin of NDV (Alexander, 1988). The first possibility is that a major mutation of a precursor virus of low virulence took place resulting in virulent NDV. The second, is that the disease was present in poultry in South East Asia for a long time, but because it affected only poultry raised at a village level, it afforded little attention and it was only with the development of large scale poultry operations that the disease and the enormous economic losses attributable to it were noticed. The third - and more attractive - possibility is that NDV was present as enzootic in species different from poultry, and it was only when these species and poultry were brought together by chance that the disease emerged.

2.8.2 Etiology

NDV belongs to the Paramyxoviridae family, subfamily Paramyxovirinae and under genus *Avulavirus*. The genus Avulavirus is formed from mumps virus,

human Para influenza virus viruses 2 and 4, Newcastle disease virus (PMV-1) and the avian paramyxoviruses PMV-2 to PMV-3 (Alexander, 1997).

2.8.3 Host range and reservoir

The vast majority of birds are susceptible to infection with NDV, although the disease may vary enormously from one species to another. Chickens are highly susceptible, but other poultry species can be infected with NDV, and may play a role in the spread of NDV in extensively managed poultry. Approximately from 800 known avian species, only about 236 species have a record of NDV isolation. The disease is seen most frequently in domestic poultry, including guinea fowl, a species more susceptible than turkey and peafowl. Ducks, geese, partridge, and quail are relatively resistant. The most resistant species appear to be aquatic birds; while the most susceptible are gregarious birds forming temporary or permanent flocks, the birds include ducks, geese, turkeys, doves, and guinea fowl. Such birds can become infected with NDV, shed the virus, and act as a source of infection for chickens, even if they do not develop clinical signs (Martin, 1992).

2. 8.4 Clinical signs and gross lesions

The incubation period of NDV after natural exposure has been reported to vary from 2-15 days or longer, with an average of 5-6 days. Clinical signs, and the speed of which they appear vary widely, and depend upon infectivity and dose of the virus, the species, age, immune status of the host, infections with other organisms, environmental conditions and the route of exposure (Alexander, 1997). Infection with NDV displays a complete continuum from very rapid fatal disease to unapparent infection. NDV is grouped into five pathotypes Viscerotropic velogenic, Neurotropic velogenic, Mesogenic, Lentogenic and Asymptomatic enteric), by Beard and Hanson (1984) on the basis of predominant signs in affected chickens. This grouping is useful for descriptive purpose, but attention should be drawn to the fact that some overlap occurs, and thus it is difficult to classify some viruses into a specific pathotype group, recognized the presence of the a virulent strains of the virus, while many of the island states were apparently free from all path types of NDV. A sequence of events following introduction of NDV into

chickens is initiated by multiplication of the virus at the site of introduction. Initially the virus replicates in the mucosal epithelium of the upper respiratory and intestinal tracts. Then follows release of the virus into the bloodstream, a second cycle of multiplication occurs in visceral organs producing a secondary viremia. This leads to infection of other target organs such as lungs, intestine and central nervous system. Signs of the disease and liberation of the virus into the environment are associated with the second release of the virus into the bloodstream. An exception occurs with large airborne exposures where the virus replicates in and is disseminated from the epithelium of the respiratory tract (Beard and Hanson, 1984).

Pathologic changes associated with ND vary greatly from bird to bird, flock to flock and from one geographic region to another. Gross lesions vary depending on virus and may also be absent. Cadavers of birds that died because of virulent NDV, usually have a dehydrated appearance. Little evidence of gross lesions is found in the central nervous system even in birds showing neurological signs prior to death. Gross pathological lesions are usually present in the respiratory tract in birds with respiratory illness. They consist predominantly of hemorrhagic lesion and congestion of the trachea and in addition air sacculitis may be evident. Egg peritonitis is often seen in laying hens affected with virulent NDV (Beard and Hanson, 1984). Histopathologically, hyperemia, edema, hemorrhage, and other changes in blood vessels are found in various organs. In general, in most tissues and organs involved, the lesions include hyperemia, necrosis, cellular infiltration, and edema. Lesions in the central nervous system are characterized by non purulent encephalomyelitis (Beard and Hanson, 1984). For virus isolation in the head, spleen and long bones from acutely sick birds should be collected after disinfection in flame. The brain, bone marrow and spleen tissues are crushed into pieces using pestle and mortar with 5 mL broth, 2000 I.U. of penicillin and 5 mg of streptomycin. In addition to virus isolation other tests conducted to confirm the disease include: haemagglutination test, haemagglutination inhibition (HI) test, virus neutralization test, fluorescent antibody technique (FAT) and complement fixation test (CFT).

2.8.5 Laboratory diagnosis

Diagnosis can be made on the basis of a number of direct and indirect methods. Indirect methods include among others haemagglutination inhibition test (HI), enzyme-linked immunosorbent assays (ELISA), virus plaque neutralization (VN), complement fixation (CFT)], (OIE, 1996).

Direct methods include virus isolation, electron microscopy and virus characterization. Samples from dead birds should consist of oro-nasal swabs, as well as sample collected from lungs, air sac, intestine (including contents), spleen, brain, liver and heart tissues collected separately or as a pool and placed in phosphate buffer isotonic saline (7.0-7.4 containing antibiotic) (Alexander, 2000).

The importance and impact of NDV is directly related to the virulence of that isolate. It is however necessary to carry out laboratory assessment in order to determine the pathogenicity of the virus isolate. At present, three in vivo tests are used for this purpose, Mean death time (MDT), intracerebral pathogenicity index (ICPI) and Intra venous pathogenicity index (IVPI) (Njagi *et al* 2010).

2.8.6 Influence of age and sex

Ezeokoli *et al.* (1984) concluded that in backyard management systems birds around 6-24 weeks of age had the highest risk of NDV infection. Furthermore, studies have suggested, that the sex of the birds may influence the morbidity and mortality of NDV. Female chickens had higher mean NDV titres compared to the male birds. Hens and pullets may therefore be playing a significant role in the carriage and maintenance of the virus in these rural poultry populations. Previous studies by Kutubuddin (1973) indicated that male birds were more affected by NDV than female birds. However, in the study the females had higher viral load than male birds in all age groups. Huchzermeyer (1993) noted that brooding hens and hens with chicks that were kept segregated could also escape infection. Chickens that may have survived previous ND outbreaks produce chicks, which become susceptible to ND after the maternal antibodies have waned

(Huchzermeyer, 1993). The actual cause of this apparent sex related differences in NDV carriage is not yet understood.

2.8.7 Spread of ND in village chickens

Inhalation of aerosols is considered to be the primary mode of transmission of NDV within a flock. Approximately 2 days after exposure, and a full day before showing clinical sign, the infected birds begin to liberate virus into air, and continue to do so for several days (Hanson, 1988), resulting in a fine aerosol and/or larger droplets which transmit the virus to other birds in the house. This mode of transmission may also occur in extensively managed poultry, especially in semi-intensive systems, if the birds are housed overnight. However, it is not likely to be nearly as important an infection route as in intensively managed poultry (Martin, 1992).

The oral route of infection seems to be more common for the transmission of NDV in free range scavenging poultry (Martin, 1992). Rural poultry commonly eat infected faeces and material likely to be contaminated with faeces.

Live infected chickens are the most likely means of introduction of NDV into village populations, and the live bird markets are probably the major source of infection (Alexander, 1988b; Martin, 1992; Nguyen, 1992). In many developing countries, farmers may try to sell their chickens when they show signs of disease. This attitude is reported from Vietnam (Nguyen, 1992) and Kenya (Muslime, 1992).

The role of wild birds in the spread of NDV is not fully understood (Martin, 1992). However, for village poultry, the chance of contact with wild birds is higher than for intensive poultry. The migratory nature of waterfowl and the reported isolation of pathogenic NDV from passerines during an epidemic in chickens indicate their potential involvement in the spread of NDV (Alexander, 1988).

Clothing, hair and footgear of people and equipment such as crates, egg flats, feed sacks and vehicles undoubtedly act as mechanical vectors. In this way, vaccinating

crews have contributed to the spread of velogenic, viscerotropic ND in California (Utterback and Schwartz, 1973).

Field investigations in Malaysia, Thailand and the Philippines have shown that the presence of busy roads, intervening paddy fields and other factors are likely to affect the spread of NDV in village poultry populations (Jackson, 1992).

2.8.8 Serological prevalence of NDV in village poultry

Positive serological results in unvaccinated adult birds are clear evidence of exposure to NDV. Serological studies in village chickens were made in several countries. In the Khon Kaen province in Thailand, a health and productivity study in native village chickens was carried out in two villages from September 1987 to August 1988 (Thitisak *et al.*, 1989). Monthly blood samples were taken from 920 unvaccinated wing tagged birds and 448 offspring. The mean HI titre for ND was high in newly hatched chicks, and declined as maternal antibodies disappeared at about 90 days. Thereafter, mean titres rose steadily as the age of birds increased, peaking in birds 3 years of age. However, the proportion of seropositive samples was not stated. ND occurred during the study period, but no information on the pattern of occurrence was provided.

In Nigeria, Baba *et al.* (1998) in a slaughter house survey for antibodies against ND in 242 village chickens recorded a prevalence rate of 26%.

2.8.9 Prevention and control

Effective control of Newcastle disease requires good sanitation, management, quarantine, an appropriate vaccination program, and monitoring system, including serotyping and pathogenicity testing of isolated virus. A minimum of 70% of flocks in high risk areas must be included in sanitary and combined vaccination programs if control is to be effective. Vaccination against ND can be performed using either live or inactivated vaccines. The effectiveness of ND vaccines in the control of the disease, whether under closed commercial, semi-closed intensive, or under free

range rural systems in tropical countries, depends on the virulence of the field strain, immunological state of the birds and the method of vaccine application.

2.9.10 Differential diagnosis

Fowl cholera, highly pathogenic avian influenza, Infectious Laryngotracheitis, Fowl pox (diphtheritic form), Psittacosis (psittacine birds), Mycoplasmosis, Infectious bronchitis, Aspergillosis, Also management errors such as deprivation of water, lack of or nutritionally deficient feed and poor ventilation. In pet birds: Pacheco's parrot disease (psittacine birds), salmonellosis, adenovirus, and other paramyxoviruses.

2.10.1 Geometric mean

The geometric mean, in mathematics is a type of mean or average which indicates the central tendency or typical value of a set of numbers. A geometric mean is often used when comparing different items- finding a single "figure of merit" for these items- when each item has multiple properties that have different numeric ranges. For example, the geometric mean can give a meaningful "average" to compare two companies which are each rated at 0 to 5 for their environmental sustainability, and are rated at 0 to 100 for their financial viability. If an arithmetic mean was used instead of a geometric mean, the financial viability is given more weight because its numeric range is larger- so a small percentage change in the financial rating (e.g. going from 80 to 90) makes a much larger difference in the arithmetic mean than a large percentage change in environmental sustainability (e.g. going from 2 to 5). The use of a geometric mean "normalizes" the ranges being averaged, so that no range dominates the weighting, and a given percentage change in any of the properties has the same effect on the geometric mean. So, a 20% change in environmental sustainability from 4 to 4.8 has the same effect on the geometric mean as a 20% change in financial viability from 60 to 72.

The geometric mean is similar to the arithmetic mean except that the numbers are multiplied and then the n^{th} root (where n is the count of numbers in the set) of the resulting product is taken.

For instance, the geometric mean of two numbers, say 2 and 8, is just the square root of their product; that is $\sqrt[2]{2 \times 8} = 4$. As another example, the geometric mean of the three numbers 4, 1, and 1/32 is the cube root of their product (1/8), which is 1/2; that is $\sqrt[3]{4 \times 1 \times 1/32} = 1/2$.

More generally, if the numbers are x_1, \ldots, x_n , the geometric mean G satisfies

$$G = \sqrt[n]{x_1 x_2 \cdots x_n},$$

and hence

$$\log G = \frac{1}{n} \sum_{i=1}^{n} \log x_i.$$

The latter expression states that the log of the geometric mean is the arithmetic mean of the logs of the numbers.

The geometric mean can also be understood in terms of geometry. The geometric mean of two numbers, a and b is the length of one side of a square whose area is equal to the area of a rectangle with sides of lengths a and b. Similarly, the geometric mean of three numbers, a, b, and c, is the length of one side of a cube whose volume is the same as that of a cuboid with sides whose lengths are equal to the three given numbers.

The geometric mean applies only to positive numbers. It is also often used for a set of numbers whose values are meant to be multiplied together or are exponential in nature, such as data on the growth of the human population or interest rates of a financial investment.

The geometric mean is also one of the three classical Pythagorean mean together with the aforementioned arithmetic mean and the harmonic mean. For all positive data sets containing at least one pair of unequal values, the harmonic mean is always the least of the three means, while the arithmetic mean is always the

greatest of the three and the geometric mean is always in between (Encyclopedia, 2012).

2.10.2 Vaccination titre

Titration is the method or process by which the strength or concentration of a substance in a solution is measured. The result obtained is called the titre of the solution. The word titre is often used in connection with vaccination of poultry. It is, however, used to refer to two quite distinct subjects. On the one hand we have the titre of the vaccine in use, and on the other the titre of antibody produced in the bird by way of vaccination. Most of the live vaccines are titrated by successive dilution to find the minimal dose necessary to bring about a given effect in a biological system. The titre of a given batch of vaccine is usually a good measure of its ability to immunize animals. It must be understood that the titre necessary is specific for each product. The methods used in titration can have a considerable effect on the titre obtained. Titres are found by successive dilution of the serum to find the minimal quantity capable of producing a given effect in a biological system. Effects often used in titration of antibody are inhibition of haemagglutination and neutralization of infectivity (McMullin, 1984).

2.10.2.1 The Immune response

When a vaccine is applied to a bird, a complex biological mechanism is set in motion which normally results in the elevation of the birds' specific defenses against the disease in question, and it may also raise non-specifically its defenses against other diseases. The main components of this defence system are:

- 1. Antibodies circulating in the blood.
- 2. Antibodies secreted on the mucosae (including conjunctiva, trachea, intestine etc.).
- 3. Lymphocytes and macrophages sensitized to the antigen.
- 4. Interferon measured.

2.10.2.2 Alternatives available

Although some methods of studying sensitized lymphocytes and mucosal antibodies have been developed for research purposes, under normal circumstances these methods have not been applied to commercial poultry. There are basically two methods which are used to evaluate birds' response to vaccination: titration of specific antibodies circulating in the blood, and measurement of the effect of a challenge exposure to one or more virulent strains of organisms. Challenge has sometimes been portrayed as the "ideal" method of evaluating the effectiveness of a vaccine. On the surface this may seem correct but in fact this approach has a number of serious defects if we are thinking about routine flock monitoring (as opposed to quality control of batches of vaccine in the laboratory). Facilities capable of safely containing the challenge organisms throughout large numbers of tests are expensive and not widely available to the industry. Simple live/die challenge models may greatly underestimate the effects of disease on productivity parameters in supposedly protected birds (especially layers). It may also underestimate the importance of virulent viral replication, in apparently healthy birds. In view of these defects of challenge techniques, emphasis is being placed increasingly on titration of circulating antibody levels as a guide to response to vaccination in commercial poultry. Among the many laboratory methods available to measure antibody the choice should be determined by availability of reagents, economy, and the accumulated experience in interpreting results. When a new test is introduced it is often necessary to start "from scratch" when it comes to interpretation what the results mean. It is sometimes necessary to estimate changes in antibody titre by the judicious use of qualitative tests. This is possible because it is generally true that the greater the proportion of birds positive in a qualitative test, the higher will be the mean titre of the group. This strategy has been used with the agar gel precipitation test for infectious bronchitis in Holland for a number of years (McMullin, 1984).

2.10.2.3 *Sampling methods*

In view of the fact that we often use very small samples to infer about the condition of a large flock of birds. Appropriate statistical techniques are available to determine the number of samples necessary for a given purpose. It is often the economic and practical considerations which actually determine the number of samples taken. Sampling should generally be stratified. That is, the larger the degree of variability we expect in a given class of birds, the greater the proportion which must be sampled. Methods of sampling must be practical, easy to teach to farm staff, and produce serum of appropriate quality for the intended test(s) (McMullin, 1984).

2.10.2.4 Standardization

There has been some discussion on the necessity of standardizing laboratory procedures so that results from one laboratory are comparable with those produced by others. While the objective is obviously desirable, the means by which it may be obtained is not so obvious. Not all laboratories have exactly the same physical facilities or equipment, and sometimes subjective judgment can have an effect on the results. Emphasis must be placed on standardization of results and not necessarily the means by which they are obtained. If co operating laboratories exchange their internal reference sera, and better yet, if all use the same national or international reference serum then there will be a strong tendency towards uniformity of results produced. Obviously there will be considerable modification of the methods in use in some laboratories so that they produce the "correct" result with the reference serum. It is perfectly possible, to produce comparable results from a given set of samples, using quite different equipment, and sometimes with small variations on the method (McMullin, 1984).

2.10.2.5 Reporting results

The way in which results of serological titrations are reported has often been a source of confusion. The term G.M.T. (Geometric Mean Titre) is often used. In fact

this is nothing more than the simple arithmetic mean of the logarithms of the last positive dilution of each serum. By using a doubling dilution sequence beginning at 2, the number of the last positive tube or well is equal to the logarithm to the base 2 of the dilution. This makes reading and calculation of the mean titre very simple, and is the reason for the increasing popularity of this type of dilution sequence. In view of the fact that in many cases it is not enough to have good titres - it is also necessary that they should be reasonably uniform within the flock - some measure of variability should be included when reporting the results. We have used the standard deviations of the logs of the titres for this purpose (McMullin, 1984).

2.10.2.6 Interpretation

Unfortunately there is not always a direct relationship between the titre of circulating antibody and how birds will behave when subject to challenge. In view of the complex nature of the immune response it would be surprising if such a rigid relationship did exist. There are however data in the literature which correlate circulating antibody with results of challenge, especially when the latter is carried out under carefully controlled conditions. It must be remembered that birds with quite low titres of circulating antibody may be quite well protected by local immunity, and, on the other hand, birds with high titres may, in the absence of clinical signs, excrete large quantities of virulent virus, due to a lack of local immunity. Any laboratory test, care must be taken in interpreting the results. Routine monitoring of serum antibody response to vaccination is most appropriate for comparison between flocks immunized with the same type and route of vaccine application. Rather than relying on published data to interpret the results obtained, it is recommended that each organization establish a normal base-line curve for response to vaccination using its laboratory and normal flocks immunized with the customary program. This entails intensive initial testing to determine the absolute values for antibody titre post-vaccination, and the degree of variability obtainable within and between flocks. Such a curve for HI(haemagglutination -inhibition) response of broilers on a large commercial farm on which there was intensive vaccination against Newcastle Disease, Subsequent testing can and should be

directed to age groups or types of birds found to have greater variability of response and to cases of suspect vaccine failure. The results then obtained are compared to the base-line curve, but always remember that an apparent improvement in vaccine response may be simply the result of exposure to field virus (McMullin, 1984).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Study Area

The study was carried out in the Teaching and Research Farm of the Modibbo Adama University of Technology, Yola in Adamawa State, located at the North Eastern part of Nigeria. It lies at Latitude 9° and 14¹ North and Longitude 12° and 28¹ East and altitude of about 152 Metres above sea level. Adamawa State is within the Guinea Savanna Zones of West Africa characterized by short period of rainy season. The climate is tropical with distinct dry and wet season. The rainfall starts in April and ends in October while Dry season starts in November to March. The state mean annual rainfall ranged about 700mm-1600mm and relative humidity ranges from 5%-42% with the mean minimum temperature of 15.2°C average temperature of 39°C (Adebayo and Tukur, 1999).

3.1.1 Experimental chickens

Different phenotypes of experimental chickens (grower) were obtained from villages without the history of crossbreeding programmes involving exotic chickens and these represented the foundation stock. The chickens were placed on broad spectrum antibiotics, dewormed using Piperazine salt and treated for ectoparasites. From each phenotype 16 pullet layers and 2 cockerels were randomly selected for the study. The birds were housed on concrete floor full of wood shavings at 16 hens per pen. The birds were exposed to natural day light feeding of about 12 hours per day. All the pullets were placed on grower diet and fed at the rate of 80 - 90 g /bird

/day. The grower feed contained 15% crude protein and 2550Kcal Metabolizable energy (ME) per Kg of feed. Grower feed were gradually replaced with layer mash at point of lay (24 weeks). The layer mash contained 16.5% crude proteins and 2650 kcal/kg ME of feed. The chickens were provided with wooden laying boxes for laying. Eggs were collected twice a day at 10.00 am and 3.00 pm (Appendix 4 and 5).

3.1.2 Experimental design

Three phenotypes of Sixteen (16) hens and Two (2) cockerels each (48 breeder hens and 6 cocks) were selected and assigned randomly for the study. Each group was mated with two cockerels eight times in a completely randomized design (CRD).

3.2.0 Experiment i: Determination of internal and external egg quality characteristics

A total of two hundred and seventy fertile eggs were collected from three phenotypes. Determination of external and internal egg quality traits were carried out immediately after collection, as described by (Fayeye *et al.*, 2005). The parameters included egg weight, egg length, egg width, egg index, yolk weight, yolk diameter, yolk index, shell weight, shell thickness, albumen height, albumen width and haugh unit. Digital electric balance was used in weighing the eggs. Egg length and width was measured with the aid of pair of Varnier caliper (mm). The values of egg length and egg weight was used to determine the eggs index. The thickness of each shell was determined using the micrometer screw gauge (mm). The yolk and albumen height was determined using a pair of Varnier caliper calibrated (mm). Accuracy of shell thickness was determined by measuring shell samples at the broad and middle portion and narrow end of the shell. The average shell thickness was then recorded (mm). Yolk index was determined as a ratio of the yolk height to yolk width. The egg shape index, yolk index and haugh unit was calculated using the formulae below.

Egg shape index (%) = [egg width (cm)/egg length (cm)] x100%

Yolk index (%) = [yolk height (mm)/yolk diameter (mm)] x100%

Haugh unit (Hu) = $100 \log (H+/G(30 \text{w o.}37-100)+19]/100\%$

Where

HU = Haugh units

HA = observed albumin height (mm)

G = gravitational constant 32.2

W = observed weight of egg

3.3.0 Experiment ii: Determination of hatchability parameters

A total of three hundred and twenty four hatchable eggs were selected. The selection was done based on the uniform size, good shape, clean shell, no cracks and the weight of the all hatchable eggs before setting was recorded in gram using digital electrical balance. The eggs were disinfected by rubbing with a wet cloth dipped in a powerful disinfectant (Morigad^R) before storage. The hatching eggs were then fumigated using potassium permanganate before setting. The incubator was test run for 24 hours to observe if there were any defects. Temperature of 39.4°C (103 ° F) and 75% RH was maintained through out the incubation. Eggs were turned at 2 or 3 hours intervals, at least 5 times daily and candled at Day 7 days and 18 to remove the infertile ones and dead embryo (dead in germs) respectively. Turning was stopped immediately after the last candling. Optimum humidity was ensured three days before hatching.

At the end of day 21 of incubation, the incubator was opened. The number of hatched out chicks including the normal, abnormal chicks, dead in shell un-hatched

eggs were carefully counted and recorded. Chicks which were under sized, poorly feathered, parrot beaked, blind, lame, open navel etc was considered as abnormal. Hatched out chicks was weighed and recorded using digital electric balance. Estimation of fertility, hatchability on fertile eggs, hatchability on set eggs, embryonic mortality, dead in shell, normal and abnormal chicks hatched and the weight of each day old chicks were recorded in gram. The percentage fertility and hatchability were estimated using the relationship below:

Fertility (%) = No of fertile eggs/ Total no of eggs produced x 100%

Hatchability on fertile eggs (%) = No of eggs hatched out/Total no of fertile eggs x 100%

Hatchability on set eggs (%) = No of eggs hatched out/Total no of eggs set x 100%

3. 4.0 Rearing of day-old chicks/brooding

All the successfully hatched out chicks were kept in a small pen at a control rearing temperature of 35°C for the first one week and gradually reviewed down as the chick's ages. They were placed on Antibiotics and Multivitamin (Aliseryl)^{R.} for the fist one week. The chicks were also fed (*ad-lib*) chick mash containing 18% crude protein for the fist nine weeks of age (Appendix 3). No vaccination was carried out throughout the rearing period.

3.5.0 Experiment iii: Susceptibility of three phenotypes to experimental hertz 33/56 newcastle disease virus

3.5.1 Experimental birds

Forty five village chickens, made up of fifteen (15) each of frizzle, naked neck and smooth feathered types were hatched and raised up to twenty weeks of age.

3.5.2 Experimental design

Forty five village chickens comprising fifteen (15) each of frizzles, naked necks and smooth feathered types were used in this study. Thirty (30) chickens comprising 10 of each phenotype were assigned to two groups A and B and 9 chickens comprising 3 of each phenotype out of the remaining 15 were assigned to group C. The remaining 6 chickens, two (2) of each phenotype were introduced to groups A and B to serve as in-contact. Group A and B were challenged with Newcastle virus strain Hertz 33/56 of chicken origin using intraocular (i/o) and intranasal (i/n) routes respectively. While group C served as uninfected control group. 0.2mls of the diluted virus were instilled in to the eyes and nostril of each chicken.

3.5.2.1 Serum samples

The experimental birds were bled on days 0, 3, 7, 14, 21 and 28 post infections. Three milliliters (3 ml) of blood was collected from each experimental bird through the wing vein using sterile needle and 21G needle into sterile vacutainer tubes. The blood samples collected in the vacutainer tubes were kept at room temperature to clot. Serum samples were harvested after centrifuging the vacutainer tubes at 1,500 rpm for 10 minutes, into sterile cryotubes and kept at -20 °C until tested.

3.5.2.2 Chicken red blood cells

Four (4) ml of chicken red blood cells (RBC) was drawn onto 1 ml of Alsevers solution (anticoagulant) in a sterile syringe. The RBC was washed 3 times first with Alsevers solution and subsequently with Phosphate Buffered Saline (PBS), PH 7.4

and centrifuged at 1,000 rpm for 5 minutes. A 1% suspension of the chicken RBC in PBS was prepared for use in Haemagglutination (HA) and Haemagglutination inhibition (HI) tests (Allan and Gough, 1974).

3.5.1.3 *Antigen*

Newcastle disease vaccine 'Lasota' obtained from National Veterinary Research Institute (NVRI) Diagnostic Laboratory Yola, was used as an antigen for the HA and HI tests. The HA titre of the ND vaccine Lasota was determined as described by Allan and Gough (1974) and appropriately diluted to obtain the 4HA unit used in the HI test. The HI titre for each chicken serum was determined and expressed as the reciprocal of the last dilution that completely inhibits agglutination of the chicken RBC.

3.5.1.4 *Challenge virus:*

The virus was diluted with phosphate buffered saline (PBS) of pH7.2 at the ratio of 1:10 to give virus Lethal Dose fifty (LD₅₀) of $10^{6.5}$ ml⁻¹. And 0.2 ml of the dilution was used to challenge the chickens. The chickens were monitored daily for clinical signs, morbidity and mortality for 28 days post infection. Postmortem examination was carried out on chickens that died. Gross lesions observed were recorded and some organs were frozen at -20 0 C for viral isolation and identification.

3.5.1.5 Serological test

Haemagglutination inhibition (HI) test was used for the detection and quantification of antibodies against NDV as described by (Allan and Gough, 1974). The HI test was performed using beta-techniques (constant virus and varying serum) against 4 HAUs computed from the result of the HA titration. Briefly, $25\mu l$ of PBS was added to all the wells of a microtitre plate and $25\mu l$ of the test serum was added into the first well (A1) and diluted across the plate by transferring $25\mu l$ from the first well to the second well and up to the last well (A12). Twenty five microlitres ($25\mu l$) of appropriately diluted antigen (4HAU) was added to all the wells across the plate. Finally, $25\mu l$ of 1% chicken RBC was added to all the wells and the plate

incubated at room temperature for 30 minutes. Positive and negative NDV sera were also included on the plate. The titre was taken as the reciprocal dilutions where there was complete inhibition of agglutination of the chicken RBC.

3.6.1 Procedure for collection of blood from chickens

The birds were restrained physically with the help of an assistant who held the legs and placed it horizontally on its back. The assistant placed his other hand under the back to support it and the wing of the chicken was pulled out towards me, while the site for bleeding was disinfected by swabbing with 70% alcohol, a syringe of 5mls and 25 gauge was inserted under the tendon and directed the needle into the wing vein in the direction of the flow of blood. Precautions were taken not to damage the Ulnar nerves and hygienic conditions were also maintained through out the bleeding periods. Once the tip of the needle is in the vein, I gently pulled the plunger of the syringe, blood flow into the syringe. If, blood does not flow, release the plunger and make a very slide adjustment to reposition the end of the needle. Patient and a gentle suction withdraw the blood. Chicken veins collapses easily and hematoma can be formed. After collecting blood a pressure cotton swab was applied to discourage further bleeding (FAO, Undated)..

3.6.2 Precautions for collection of blood from chickens

The chickens were gently handled. Whole blood was collected quickly. Care was taken not to damage the wing vein which would lead to hematoma. Lost of blood, trauma to the chickens and stress during handling were minimized.

3.7.0 Statistical analysis.

All the data generated were subjected to Analysis of variance (ANOVA) using statistical package of SSPS 13.0 Relevant means were separated using Duncan Multiple Range, where significant differences exist. The Correlation analysis was carried out using Pearson Correlation Coefficient. The Geometric mean titre of Newcastle disease antibodies was calculated using descriptive statistics (CDC,

1998).Briefly, the geometric mean of a sample of n positive observations was defined as the nth root of the product of the n numbers:

$$x \text{ geo} = n \sqrt{x_1, x_2, \dots, x_n}$$

Thus, the geometric mean was calculated as x geo = antilog₁₀ (1/n \sum log10 x₁) where x₁ = the reciprocal of dilution.

CHAPTER FOUR

RESULTS

4.1 Experiment I

4.1.1 Effect of hen of different phenotypes on egg quality:

Results of egg quality traits of nine phenotypes of local chickens are presented in Table 1. The results indicated that all the parameters were significantly affected by the type of phenotypes except yolk width and shell thickness. Frizzle feathered consistently produced heavier, wider egg and albumin and heavier shell. Similarly naked neck produced higher egg index yolk height and yolk index. Egg weight had a significant (p<0.01) effect on all the phenotype with 45.04, 41.90, 40.82, 41.59, 41.66, 39.64, 40.71, 37.80 and 38.44g for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Egg length had a significant (p<0.05) effect on the phenotype being 5.05, 4.98, 4.96, 5.28, 5.38, 4.96, 5.04, 4.85 and 4.94cm for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Egg width similarly had a significant (p<0.001) effect on the phenotype with 3.95, 3.78, 3.76, 3.94, 3.70, 3.74, 3.76, 3.66 and 3.69cm for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Egg index had a significance (p<0.05.) effect on all the phenotype studied with 77.61,

Table 1: Egg Quality Traits of Nine Phenotypes of Local Chickens

Parameters	Treatment								
	1	2	3	4	5	6	7	8	9 SEM
Egg weight [g]	45.04 ^a	41.90 ^b	40.81 ^{bc}	41.59 ^b	41.66 ^b	39.64 ^{bc}	40.71 ^{bc}	37.80°	38.44 ^{bc} 2.14**
Egg length [cm]	5.05 ^a	4.98 ^{bc}	4.96 ^{bc}	5.29 ^{ab}	5.38 ^a	4.96 ^{bc}	5.04 ^{abc}	4.85°	4.92 ^{bc} 0.17 *
Egg width [cm]	3.95 ^a	3.78 ^b	3.76 ^b	3.94 ^a	3.70 ^b	3.74 ^b	3.76 ^b	3.66 ^b	3.69 ^b 0.10**
Egg index [%]	77.61 ^{ab}	76.25 ^{ab}	76.09 ^{ab}	79.69 ^a	74.18 ^b	74.71 ^b	75.90 ^{ab}	73.80 ^b	74.79 ^b 1.85*
Albumin height [mm]	6.51 ^b	9.19 ^a	7.41 ^b	6.80 ^b	7.43 ^b	7.51 ^b	6.66 ^b	7.11 ^b	7.03 ^b 0.79**
Albumin width [mm]	6.25 ^a	5.31 ^{cd}	5.70 ^{bc}	6.01 ^{ab}	5.76 ^b	5.84 ^{ab}	5.86 ^{ab}	5.26 ^d	5.19 ^d 0.36**
York height [mm]	1.60 ^{ab}	1.67 ^a	1.46 ^{bc}	1.67 ^a	1.45 ^c	1.60 ^{ab}	1.60 ^{ab}	1.53 ^{ab}	1.55 ^{abc} 0.08**
York width [mm]	3.90	3.90	4.00	3.86	3.90	3.85	3.84	3.90	3.86 0.05N
York index [%]	41.64 ^{abc}	43.61 ^{ab}	36.45 ^d	45.92 ^a	37.78 ^{cd}	42.89 ^{abc}	41.72 ^{abc}	39.35 ^{bcd}	39.99 ^{bcd} ^{2.97*}
Shell weight [g]	5.11 ^a	4.06 ^b	4.01 ^b	4.01 ^b	4.24 ^b	3.69 ^b	4.13 ^b	3.63 ^b	3.99 ^b 0.43*
Shell thickness [mm]	.45	.41	.59	.53	.53	.54	.45	.53	.54 .06NS
Haugh	85.13°	100.50 ^a	91.30 ^{bc}	91.86 ^c	91.06 ^{bc}	92.18 ^b	90.35 ^{bc}	90.54 ^{bc}	89.96 ^{bc} 3.99 ^x

Key: 1 = Frizzle, 2 = Frizzle naked neck, 3 = Frizzle smooth, 4 = Naked neck, 5 = Naked neck frizzle, 6 = Naked neck smooth, 7 = Smooth, 8 = Smooth naked neck, 9 = Smooth frizzle Means on the row with different superscripts are significantly different. *: P < 0.05, **: P < 0.01, ***: P < 0.001

76.25, 76.09, 79.69, 74.18, 74.71, 75.90, 73.80 and 74.79% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Albumin height and albumin width had a significant (p<0.001) effect on the phenotypes with 6.51, 9.19, 7.41, 6.80, 7.43, 7.51, 6.66, 7.11, 7.03, and 6.25, 5.31, 5.70, 6.01, 5.76, 5.84, 5.86, 5.26 and 5.19mm for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Yolk height had a significant (p<0.01) effect on all the phenotypes with 1.60, 1.67, 1.46, 1.67, 1.45, 1.60, 1.60, 1.53 and 1.56mm respectively for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Yolk width was not statistically significant in all phenotypes studied. Yolk index had a significant (p<0.01) effect on all the phenotypes with 41.64, 43.61, 36.45, 45.92, 37.78, 42.89, 41.72, 39.35 and 39.99% respectively for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Shell weight had a significant (p>0.01) effect on all the phenotypes studied with 5.11 4.06, 4.01, 4.01, 4.27, 3.69, 4.13, 3.63 and 3.99g respectively for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck,

smooth x frizzle respectively. Haugh unit also had a significant (p>0.01) effect with 85.13, 100.50, 91.30, 91.86, 91.06, 92.18, 90.35, 90.54, 89.96 respectively for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively (Table 1).

4.2.1 Correlation of egg quality traits for hen

4.2.1.1 *Frizzle*

Significant negative correlations were found between haugh unit and egg weight (p<0.05), albumin height and egg width (p<0.01), haugh unit and egg width (p<0.01), haugh unit and shell weight (p<0.01), yolk height and shell thickness (p<0.05), yolk index and shell thickness (p<0.05), (Table 2).

Significant positive correlations were found between haugh unit and albumin height (p<0.01), yolk index and yolk height (p<0.01) and some significant negative correlations between albumin width and albumin height (p<0.01), haugh unit and albumin width (p<0.01), yolk index and yolk width (p<0.01), (Table 3).

Significant positive correlations were found between egg length and egg weight (p< 0.01), egg width and egg weight (p< 0.01), shell weight and egg weight (p< 0.01), egg width and egg length (p<0.05), shell weight and egg length (p<0.01), egg index and egg width (p<0.05), shell weight and egg width (p<0.01) and a significant negative correlation was observed between egg index and egg length (p<0.05), (Table 4).

Table 2: Phenotypic Correlation between External and Internal Egg Quality Traits for Frizzle

Internal Egg		Ex	kternal egg Trai	ts		
Traits						
	Egg	Egg	Egg	Egg	Shell	Shell
	weight	length	width	index	weight	thickness
Albumin height	-0.38	-0.16	-0.59 **	-0.37	-0.36	-0.24
Albumin width	-0.16	-0.05	0.30	0.27	0.35	0.10
Yolk height	0.34	0.26	0.30	0.05	0.07	-0.41*
Yolk width	0.19	0.40	0.30	-0.40	0.26	0.17
Yolk index	0.22	-0.06	-0.01	0.27	-0.09	-0.44 *
Haugh unit	-0.47 *	-0.24	-0.66 **	-0.35	-0.44 *	-0.19

^{*:} P<0.05, **: P<0.01

Table 3: Phenotypic Correlation between Internal Egg Quality Traits for Frizzle

Internal Egg Traits	3	Internal Egg Traits						
	Albumin height	Albumin width	Yolk height	Yolk width	Yolk index	Haugh unit		
Albumin height	1.00							
Albumin width	-0.64 **	1.00						
Yolk height	-0.25	-0.21	1.00					
Yolk width	-0.11	-0.02	-0.29	1.00				
Yolk index	0.28	-0.14	0.83 **	-0.76**	1.00			
Haugh unit	0.98 **	-0.64 **	0.20	-0.12	0.25	1.00		

^{*:} P<0.05, **: P<0.01

Table 4: Phenotypic Correlation between External Egg Quality Traits for Frizzle

External Egg Tra	its					
	Egg weight	Egg length	Egg width	Egg index	Shell weight	Shell thickness
Egg weight (g)	1.00					
Egg length	0.85 **	1.00				
Egg width	0.83 **	0.51 *	1.00			
Egg index	-0.03	-0.49 *	0.44 *	1.00		
Shell weight	0.85 **	0.82 **	0.60 **	-0.25	1.00	
Shell thickness	-0.19	-0.04	-0.13	-0.04	-0.05	1.00

^{*:} P<0.05, * *: P<0.01

4.2.1.2 Naked neck

Significant positive correlations were found between albumin height and egg weight (p<0.01), yolk height and egg weight (p<0.01), yolk width and egg weight (p<0.01), haugh unit and egg weight (p<0.01), yolk width and egg length (p<0.01), albumin height and egg width (p<0.01), yolk height and egg width (p<0.01), yolk width and egg width (p<0.01), yolk index and egg width (p<0.01), haugh unit and egg width (p<0.01), yolk height and egg index (p<0.05), haugh unit and egg index (p<0.01), albumin height and shell weight (p<0.01), yolk width and shell weight (p<0.05), haugh unit and shell thickness (p<0.01), yolk width and shell thickness (p<0.01), yolk width and shell thickness (p<0.01), yolk width and shell thickness (p<0.01), (Table 5).

Significant positive correlations were found between yolk height and albumin height (p<0.05), haugh unit and albumin height (p<0.01), yolk width and albumin width (p<0.01), haugh unit and yolk height (p<0.05), yolk index and yolk width (p<0.01), haugh unit and yolk width (p<0.05), haugh unit and yolk index (p>0.01), (Table 6).

Significant positive correlations were found between egg length and egg weight (p<0.01), egg width and egg weight (p<0.01), shell weight and egg weight (p>0.01), shell thickness and egg weight (p>0.01), egg width and egg length (p<0.05), shell weight and egg length (p<0.05), egg index and egg

Table 5: Phenotypic Correlation between External and Internal Egg Quality Traits for Naked Neck

Internal Egg Traits		External Egg Traits								
	Egg weight	Egg length	Egg Width	Egg index	Shell weight	Shell thickness				
Albumin height	0.67 **	0.23	0.45 **	0.29	0.71 **	0.54 **				
Albumin width	0.21	0.25	0.36	0.29	-0.08	-0.04				
Yolk height	0.54 **	0.13	0.69 **	0.42 *	0.37	0.34				
Yolk width	0.63 **	0.55 **	0.58 **	0.36	0.46 *	0.41 *				
Yolk index	0.26	0.03	0.56 **	0.37	0.06	0.09				
Haugh unit	0.73 **	0.35	0.74 **	0.57 **	0.68 **	0.54 **				

^{*:} P<0.05, **: P<0.01

Table 6: Phenotypic Correlation between Internal Egg Quality Traits for Naked Neck

Internal Egg Traits Albumin Albumin Yolk Yolk Haugh Yolk height width height width index unit Albumin height 1.00 -0.08 Albumin width 1.00 Yolk height 0.48 * 0.12 1.00 Yolk width 0.30 0.53b** 1.00 0.19 Yolk index 0.23 0.04 -0.09 0.88** 1.00 Haugh unit 0.08 0.46 * 0.41** 0.91 ** 0.63* 1.00

^{*:} P<0.05, **: P<0.01

width (p<0.01), shell weight and egg width (p<0.01), shell thickness and egg width (p<0.05), shell thickness and shell weight (p<0.01), (Table 7).

4.1.2.3 Smooth

Significant positive correlations were found between albumin width and egg weight (p<0.01), yolk height and egg weight (p<0.05), albumin width and egg width (p<0.05), albumin width and shell weight (p<0.01), albumin height and shell thickness (p<0.01) and also some significant negative correlations between albumin height and egg length (p<0.05) haugh unit and egg length (p<0.01), albumin height and shell weight (p<0.05), (Table 8).

Significant positive correlations were found between haugh unit and albumin height (p<0.01), yolk index and yolk height (p<0.01) and also some significant negative correlations between albumin width and albumin height (p<0.01), yolk width and yolk height (p<0.05), yolk index and yolk width (p<0.01), (Table 9).

Significant positive correlations were found between egg length and egg weight (p<0.01), egg width and egg weight (p<0.01), shell weight and egg weight (p<0.05), egg index and egg width (p<0.01), shell weight and egg width (p<0.01) and some significant negative correlations between shell thickness and egg weight (p<0.05), shell thickness and egg length (p<0.01), (Table 10).

Table7: Phenotypic Correlation between External Egg Quality Traits for Naked Neck

External Egg Tr	raits					
	Egg weight	Egg length	Egg width	Egg index	Shell weight	Shell thickness
Egg weight (g)	1.00	lengui	widii	muex	weigiii	unckness
Egg length	0.60**	1.00				
Egg width	0.71**	0.50*	1.00			
Egg index	0.29	0.09	0.72**	1.00		
Shell weight	0.91**	0.49*	0.56**	0.24	1.00	
Shell thickness	0.68**	0.26	0.48**	0.39	0.77**	1.00

^{*:} P<0.05, * *: P<0.01

Table 8: Phenotypic Correlation between External and Internal Egg Quality Traits for Smooth feathered

Internal Egg Traits	External E	External Egg Traits							
-	Egg weight	Egg length	Egg width	Egg index	Shell weight	Shell thickness			
Albumin height	-0.34	-0.41*	-0.30	0.15	-0.41*	0.61 **			
Albumin width	0.69 **	0.38	0.50*	0.01	0.65**	-0.39			
Yolk height	0.43 *	-0.08	0.33	0.17	0.26	0.09			
Yolk width	0.18	0.26	0.18	-0.21	0.04	-0.31			
Yolk index	0.15	-0.17	0.19	0.29	0.09	0.19			
Haugh unit	-0.16	-0.44**	-0.10	0.10	-0.29	0.59 **			

^{*:} P<0.05, **: P<0.01

Table 9: Phenotypic Correlation between Internal Egg Quality Traits for Smooth feathered

Internal Egg Trai	ts					
	Albumin height	Albumin width	Yolk height	Yolk width	Yolk index	Haugh unit
Albumin height	1.00					
Albumin width	-0.55 **	1.00				
Yolk height	-0.07	0.29	1.00			
Yolk width	-0.33	0.13	-0.45*	1.00		
Yolk index	0.15	0.10	0.83**	-0.83**	1.00	
Haugh unit	0.90 **	-0.35	0.10	-0.39	0.29	1.00

^{*:} P<0.05, **: P<0.01

Table10: Phenotypic Correlation between External Egg Quality Traits for Smooth feathered

External Egg Tr	External Egg Traits									
	Egg weight	Egg length	Egg width	Egg index	Shell weight	Shell thickness				
Egg weight (g)	1.00									
Egg length	0.53 **	1.00								
Egg width	0.79 **	-0.26	1.00							
Egg index	0.21	0.27	0.56 **	1.00						
Shell weight	0.47 *	0.40	0.41 **	0.28	1.00					
Shell thickness	-0.41 *	-0.76 **	-0.24	0.19	-0.33	1.00				

^{*:} P<0.05, * *: P<0.01

4.3 Experiment II

4.3.1 *Effect of hen of different phenotypes on hatchability traits:*

The hatchability traits of nine different phenotypes of local chickens are presented in Table 11. All the hatchability traits were significant except hatchability on fertile egg and dead in shell. Hatching egg weight had significant (P<0.01) effect with 36.30, 40.06, 44.50, 44.48, 40.69, 40.85, 36.91, 38.34 and 40.29g for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Naked neck produced heaviest egg (44.48) and frizzle produced the lightest (36.00). Fertility had significant (P<0.05) effect on all the phenotypes with 12.49, 50.00, 58.20, 55.35, 43.75, 20.8, 32.55, 66.68, and 64.81% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Smooth naked neck (66.68) was the most fertile chicken, followed by smooth frizzle (64.81).and by frizzle (ff) least (12.49). Hatchability on set eggs had a significant (P<0.05) effect with 8.33, 0.00, 14.86, 12.51, 39.58, 16.66, 9.48, 20.84 and 23.29% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Naked neck frizzle produced highest number of day old chicks on hatchability on set than any phenotype. There was no significant

Table 11: Hatchability Traits of Nine Phenotypes of Local Chickens Treatment

	-									_
Parameters	1	2	3	4	5	6	7	8	9	SEM
Egg weight (g)	36.30 ^d	40.06 ^{bc}	44.50 ^a	44.48 ^a	40.69 ^b	40.85 ^b	36.91 ^{cd}	38.34 ^{bcd}	40.29 ^b	1.06***
Fertility (%)	12.49 ^c	50.00 ^{ab}	58.20 ^a	55.35 ^{ab}	43.75 ^{abc}	20.83 ^{bc}	32.55 ^{abc}	66.68 ^a	64.81 ^a	11.37*
Hatchability on set eggs [%]	8.33 ^{bc}	0.00°	14.86 ^{bc}	12.51 ^{bc}	39.58 ^a	16.66 ^{bc}	9.48 ^{bc}	20.84 ^{abc}	23.29 ^{ab}	6.98*
Hatchability on fertile eggs [%]	25.00 ^{ab}	0.00 ^b	21.23 ^{ab}	19.16 ^{ab}	56.25 ^a	37.50 ^{ab}	22.74 ^{ab}	33.34 ^{ab}	37.03 ^{ab}	12.58 ^{NS}
Dead embryo [%]	12.50 ^b	25.00 ^b	62.98 ^a	27.50 ^{ab}	12.50 ^b	0.00^{b}	37.33 ^{ab}	16.68 ^b	37.44 ^{ab}	11.57*
Dead shell [%]	0.00^{b}	25.00 ^{ab}	15.81 ^{ab}	40.84 ^a	6.25 ^{ab}	12.50 ^{ab}	26.71 ^{ab}	33.33 ^{ab}	25.55 ^{ab}	10.92NS
Normal chicks	25.00 ^{bc}	0.00°	21.23 ^{bc}	19.16 ^{bc}	56.25 ^{ab}	37.50 ^{bc}	31.08 ^{bc}	37.50 ^{bc}	83.34 ^a	14.14*
Abnormal chicks	0.00 ^b	0.00 ^b	28.78 ^a	28.34 ^a	6.25 ^{ab}	0.00^{b}	18.93 ^{ab}	0.00 ^b	4.16 ^b	7.40*
Average chick weigh [g]	7.00 ^{bc}	0.00°	15.98 ^{ab}	16.04 ^{ab}	17.46 ^{ab}	12.60 ^{abc}	13.61 ^{abc}	11.03 ^{abc}	25.63 ^a	4.93*

Key: 1 = Frizzle, 2 = Frizzle naked neck, 3 = Frizzle smooth, 4 = Naked neck, 5 = Naked neck frizzle, 6 = Naked neck smooth, 7 = Smooth, 8 = Smooth naked neck, 9 = Smooth frizzle. Means on the row with different superscripts are significantly different.* P < 0.05, ** P < 0.01 *** P < 0.001

difference between the hatchability on fertile eggs on all the phenotypes studied, but apparently naked neck frizzle again produced highest no of day old chicks than any phenotype. Dead embryo had a significant (P<0.05) effect on the phenotypes with 12.50, 25.00, 62.98, 27.50, 12.50, 0.00, 37.33, 16.68, and 37.44% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Normal chick had a significant (P<0.05) effect with 25.00, 0.00, 12.30, 19.16, 56.25,37.50,31.08, 37.50 and 83.34% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Smooth frizzle produced highest (P<0.05) percentage of normal day old chicks. Abnormal chicks had a significant (P<0.05) effect with 0.00, 0.00, 28.78, 28.34, 6.25, 18.93, 0.00 and 4.16% for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth, smooth x naked neck, smooth x frizzle respectively. Average chick weight had a significant (P<0.05) effect with 7.00, 0.00, 15.98, 16.04, 17.46, 12.60, 13.61, 11.03, 25.63g, for frizzle x frizzle, frizzle x naked neck, frizzle x smooth, naked neck x naked neck, naked neck x frizzle, naked neck x smooth, smooth x smooth x naked neck, smooth x frizzle respectively. Smooth frizzle chickens produced heavier (P<0.05) than the other phenotypes (Table 11).

4.3.2. Correlation of hatchability traits for hen

4.3.2.2.1 *Naked neck*

Significant positive correlations were found between fertility and hatchability on set eggs (p<0.01), fertility and hatchability on fertile eggs (P<0.05), fertility and dead in shell (p<0.05), fertility and normal chick (p<0.05), fertility and average chick weight (p<0.01), hatchability on set eggs and hatchability on fertile eggs (p<0.01), hatchability on fertile eggs and chick weight (p<0.01), hatchability on set eggs and normal chick (p<0.01), hatchability onset eggs and average chick weight (p<0.01), hatchability of fertile egg and normal chick (p<0.05), normal chick and average chick weight (p<0.01), (Table 12).

4.3.2.2 Smooth

Significant positive correlations were found between fertility and dead in shell (p<0.05), hatchability on set eggs and hatchability on fertile eggs (p<0.01), hatchability on set eggs and normal chicks (p<0.01), hatchability on set eggs and average chick weight (p<0.01), hatchability of fertile eggs and normal chick (p<0.01), hatchability on fertile eggs and average chick weight (p<0.01), normal chick and average chick weight (p<0.05). Some significant negative correlation between hatchability on set eggs and dead embryo (p<0.05), hatchability on fertile eggs and dead embryo (p<0.05), (Table 13).

Table 12: The Correlation of different Hatchability Traits among Hens of Naked Neck

Parameters	1	2	3	4	5	6	7	8	9
1	1.00								
2	0.33	1.00							
3	-0.11	0.59**	1.00						
4	-0.16	0.42*	0.82**	1.00					
5	0.20	0.09	-0.09	-0.33	1.00				
6	0.21	0.46*	-0.21	-0.22	-0.21	1.00			
7	-0.15	0.42*	0.82**	1.00**	-0.33	-0.20	1.00		
8	0.17	0.38	0.03	-0.00	-0.14	0.00	-0.00	1.00	
9	0.00	0.54**	0.70**	0.85**	-0.36	0.11	0.85**	0.62**	1.0

Key: 1=Egg weight [g], 2= Fertility [%], 3 = Hatchability on set eggs [%], 4 = Hatchability on fertile eggs [%], 5 = Dead embryo [%], 6 = Dead shell [%], 7= Normal chicks, 8 = Abnormal chicks [%], 9 = Chick weight[g]. P < 0.05, **p < 0.01,

Table 13: The Correlation of different Hatchability Traits among Hens of Smooth feathered

Parameters	1	2	3	4	5	6	7	8	9
1	1.00								
2	0.29	1.00							
3	0.10	0.31	1.00						
4	-0.08	0.09	0.90**	1.00					
5	-0.20	-0.07	-0.49*	-0.49*	1.00				
6	0.15	0.51*	-0.21	-0.33	-0.25	1.00			
7	0.13	0.18	0.82**	0.83**	-0.29	-0.29	1.00		
8	-0.18	0.01	0.04	0.04	-0.07	0.20	-0.14	1.00	
9	0.09	0.18	0.76**	0.77**	-0.31	-0.18	0.87**	0.33	1.00

Key: 1= Egg weight [g], 2 = Fertility [%], 3 = Hatchability on set eggs [%], 4 = Hatchability on fertile eggs [%], 5 = Dead embryo [%], 6 = Dead shell [%], 7 = Normal chicks [%], 8 = Abnormal chicks [%], 9 = Average chick weight [g].* p < 0.05, **p < 0.01.

4.3.2.3 Frizzle

Significant positive correlations were found between fertility and dead embryo (p<0.05), fertility and dead in shell (p<0.01), hatchability on set eggs and hatchability on fertile eggs (p<0.01), hatchability on set eggs and normal chick (p<0.01), hatchability on set eggs and average chick weight (p<0.01), hatchability on fertile eggs and average chick weight (p<0.01), hatchability on fertile eggs and average chick weight (p<0.01), (Table 14).

4.4 Experiment iii

4.4.1 Experimental infection of three phenotypes of local chickens:

The results of experimental infection of three phenotypes of village chickens with NDV strain 33/56 indicated that all the infected birds came down with the disease as from day three (3) after infection. The onset of mortality started from day 4 PI for Frizzles and day 5 PI for naked necks and smooth feathered. A percentage mortality of 58.3%, 41.7% and 75% was observed for frizzle, naked neck and smooth feathered birds respectively. Morbidity rate of 100% was demonstrated for all the phenotypes except the in-contact chickens. The duration of the disease was 7, 9 and 10 days for Frizzle, Naked neck and Smooth feathered types respectively (Table 15). The males were more susceptible than the females in all the three phenotypes studied. Furthermore the naked and smooth feathered are more susceptible to intraocular routes of infection and frizzle feathered are susceptible to intranasal routes of infection (Table 16).

Table 14: The Correlation of different Hatchability Traits among Hens of Frizzle

Parameters	1	2	3	4	5	6	7	8	9
1	1.00								
2	0.24	1.00							
3	0.12	0.27	1.00						
4	-0.05	0.13	0.89**	1.00					
5	0.27	0.50*	-0.10	-0.17	1.00				
6	0.21	0.52**	-0.15	-0.18	-0.15	1.00			
7	-0.15	-0.13	0.89**	0.94**	-0.17	-0.18	1.00		
8	0.39	0.31	0.48*	0.25	0.20	-0.04	0.29	1.00	
9	0.20	0.26	0.86**	0.83**	0.01	-0.15	0.83**	0.78**	1.00

Key: 1 = Egg weight [g], 2 = Fertility [%], 3 = Hatchability on set eggs [%], 4 = Hatchability on fertile eggs [%], 5 = Dead embryo [%], 6 = Dead shell [%], 7 = Normal chicks [%] 8 = Abnormal chicks [%], 9 = Average chick weight [g].

^{*} p < 0.05, **p < 0.01

Table 15: Results of the Morbidity, Mortality rates among different Phenotypes of Village Chickens challenged with Hertz 33/56 NDV

Parameters	Phe	enotypes	
	Frizzle	Naked neck	Smooth
Onset of c/ signs (days)	3	4	3
Onset of mortality (days)	4	5	5
Morbidity (%)	100	100	100
Mortality (%)	58.3	41.7	75
Duration of the disease (days)	7	9	10

Table 16: Sex and Route susceptibility of infected and non infected Local Chickens

S/N	Phenotype	Total	No (%) Mortality				
			M	f	i/o	i/n	
1	F	10	5(100)	2(40)	3(60)	4(80)	
2	NK	10	1(50)	3(37.5)	3(60)	1(20)	
3	S	10	4(100)	3(50)	4(80)	3(60)	
4	MF	2	0(0)	0(0)	0(0)	0(0)	
5	MNK	2	1(100)	0(0)	0(0)	1(100)	
6	MS	2	2(100)	0(0)	1(100)	1(100)	

 $Key: M = Male, \ F = Female, \ I/O = Intraocular, \ I/N = Intranasal,$

The geometric mean antibody titre for experimental infection through intraocular route indicated that most of the phenotypes started developing protective antibody titre as from Day 7 post infection (Table 17). Similarly most of the phenotypes inoculated intranasally started developing antibodies as from day 3 except the in contact chickens (Table 18). None of the chickens in the control group developed protective HI antibody through out the experiment (Table 19).

Table 17: Geometric Mean Titre (GMT) of NDV HI Antibodies of Three Phenotypes of Village Chickens infected with Hertz 33/56 NDV using intraocular route

S/N	P/types	Total No Tested	GMT OF HI Antibody					
			D_0	D ₃	D_7	D ₁₄	D ₂₁	D ₂₈
1	NK	5	0	0	4	16	64	32
2	N	5	0	0	0	4	0	3.2
3	F	5	0	0	0	32	128	181
4	M	3	0	0	14	362	362	512

Key: HI= Haemagglutination inhibition Test, F = Frizzle, NK = Naked Neck, N = Smooth Feathered types, M = In-contact chicken, NDV = Newcastle diseases virus P/type= Phenotypes

Table18: Geometric Mean Titre (GMT) of NDV HI Antibodies of Three Phenotypes of Village Chickens infected with Hertz 33/56 NDV using intranasal route

S/N	Phenotypes	Total No Tested	GMT OF H1 Antibody					
			D_0	D_3	D_7	D ₁₄	D ₂₁	D_{28}
1	NK	5	0	2.6	10.1	12.7	40.3	50.8
2	N	5	0	2.3	3.2	45.3	128	128
3	F	5	0	1.5	0	0	0	1024
4	M	3	0	0	1.4	181	90.5	512

Key: H1 Haemagglutination inhibition Test, F = Frizzle, NK = Naked neck S = Smooth Feathered types, M = In-contact chicken NCDV = Newcastle diseases virus

Table 19: Geometric Mean Titre (GMT) of NDV HI Antibodies of Control group

S/N	P/types	Total No	GMT OF HI Antibody					
			D_0	\mathbf{D}_3	\mathbf{D}_7	D ₁₄	D_{21}	D_{28}
1	CNK	3	0	0	0	0	0	2
2	CN	3	0	0	0	1.3	0	0
3	CF	3	0	0	2	0	0	3.2

 $Key: HI = Haemagglutination \ Inhibition \ test, \ CNK = Control \ Naked \ Neck,$

CN = Control Smooth Feathered, CF = Control Frizzle, P/type= Phenotypes

CHAPTER FIVE

DISCUSSION

5.1 Egg Quality Traits

The average mean egg weight (45.04) produced by Frizzle phenotype obtained in this study is higher than the results reported by (Parmar et al. (2006) and Momoh et al. (2010), who studied egg quality traits in Kadaknath breeds and ecotype in Nigerian local chickens respectively. The result is lower than values obtained in Egyptian Bandra and Gimmizah local chickens (Yousria et al., 2010). Raji et al. (2009) also reported higher values in unspecified Nigerian local chickens. Age, feed protein levels and temperature are some of the factors adduced to affect the egg size (Benerjee, 1992). The heavier egg produced by frizzle feathered chicken was probably due to the favorable gene effect on production (Merat, 1990). The mean egg length obtained by naked neck and naked neck frizzle in the study is slightly higher than value (5.15) reported by Yakubu et al. (2008), in Nigerian naked neck and it compares favorably with the values recorded for heavy and light ecotypes and their F_i crosses in Nigeria (Momoh et al., 2010). The result is also slightly higher than values (4.83) reported for Bangladesh indigenous chickens (Islam and Ripon, 2010). The mean value of egg width produced by frizzle feathered in the present study is slightly higher than (Yakubu et al., 2008). Egg shape index is an indicator of external egg quality. The values produced by naked neck was higher (74.68), (72.60) for naked neck and normal feathered respectively (Yakubu et al., 2008). The higher shape index in the study was due to the factors earlier mentioned above and also due to heat dissipation and improved thermoregulation resulting to a better relative heat tolerance under hot climates and of genetic constitution of the chicken. Furthermore the higher shape index produced by the naked neck further consolidated their superiority over the other local chicken phenotypes in Nigeria. The mean yolk index produced by the naked neck in this study is slightly higher than values reported (Parmar et al., 2006; Momoh et al., 2010; Yousria et al., 2010), but slightly lower than values reported elsewhere (Ikeobi *et al.*, 1999), from unnamed phenotypes in Nigerian local chickens and closely similar to the result reported elsewhere (Yousria *et al.*, 2010; Olurede and Longe, 2002; Chineke, 2001; Ukachukwu and Akpan, 2007). The mean albumin height in the study is higher than the results reported by (Fayeye *et al.*, 2005; Nonga *et al.*, 2010; Momoh *et al.*, 2010). Generally albumin has a major influence on overall in terms of egg quality and large proportion of thick white indicating high quality and it ultimately will have high haugh unit.

Highest yolk index produced by naked neck obtained in this study is far higher than values reported Parmar *et al.*, (2006), but compared favorably with the values reported for heavy, and light ecotypes and its reciprocal crosses Momoh *et al.*, (2010). The result is also similar to values reported for Egyptian Bandara and Gimmizah, but slightly lower than Bandara and Gimmizah crosses.

The mean value of shell weight obtained in frizzle feathered in the present study is lower than values Yousria *et al.* (2010) reported on Egyptian strains of local chickens, but the present findings compare favorably with others elsewhere (Momoh *et al.*, 2010; Nonga *et al.*, 2010 and Islam and Ripon, 2010). The lower weight recorded in this study was probably due to the efficacy of the weighing Machine used, the methods of drying of the shell employed and probably differences in rearing systems. The main chemical component of egg shell is calcium which may have different levels in the feed.

Shell thickness did not show significant difference among the phenotypes studied. However, eggs with thick shell wall are desirable to withstand externally applied force, thus preventing breakage of egg and this is an economic indicator for commercially poultry producers and consumers. The result obtained in this study is higher than the values reported (Padhi *et al.*, 1998, Yakubu *et al.*, 2008; Momoh *et al.*, 2010; Parmar *et al.*, 2010; Yousria *et al.*, 2010). Naked neck produced heavier shell weight than the other phenotypes. The result obtained in this study is comparable to light ecotype reported (Momoh *et al.*, 2010; Nonga *et al.*, 2010), but slightly lower than values produced by heavy ecotypes (Momoh *et al.*, 2010). The

results was also slightly lower than the result reported by (Yousria *et al.*, 2010) in Egyptian Bandra and Gimmizah and its crosses.

Highest haugh unit produced by frizzle naked neck obtained in this study is far higher than the average haugh unit reported by (Parmar *et al.*, 2006) in Kadaknath breeds. The result is also higher than values reported for some indigenous backyard poultry elsewhere (Ikeobi *et al.*, 1999; Chatterjii *et al.*, 2006; Niranjan *et al.*, 2008; Yousria *et al.*, 2010; Momoh *et al.*, 2010). Since haugh unit is the measure of albumin quality which determines the quality of the egg. The higher haugh unit obtained in this study indicated superior albumin in all the phenotypes studied. Similarly, it is an indication that the research was conducted on middle age class chickens with good quality fresh eggs and also free from infectious diseases like NDV. Haugh unit and yolk index are the indicators of internal egg quality (Isikwenu *et al.*, 1999). The higher the yolk index (Ayorinde, 1987), and the haugh unit, the more the desirable the egg quality (Fayeye *et al.*, 2005).

5.1.1 Correlation of different egg quality traits

Statistically significant positive correlation recorded in the present study between albumin height, yolk width and haugh unit with egg weight; yolk width and egg length; albumin height, yolk height, yolk width; haugh with egg width; albumin height, yolk width, haugh unit with shell weight, yolk width and shell thickness are in agreement with (Yakubu et al. 2008) in naked neck and smooth feathered type of local chickens in Nigeria. Similarly significant positive correlation obtained in naked neck and smooth feathered chickens in this study between albumin height and shell thickness and haugh and shell thickness are in harmony with (Yakubu et al. 2008). Furthermore the result recorded in this study in smooth feathered chickens between albumin width and egg weight, albumin width and egg width and albumin width and shell weight are consistent with the finding of Olawumi and Ogunlade, (2008). The negative correlation value between albumin height and shell weight in smooth feathered phenotype concurs with (Yakubu et al., 2008). Similarly the negative correlation obtained between haugh unit and egg length is

comparable with (Yousria *et al.*, 2010), who reported non significant, but negative correlation value.

Significant positive correlation between egg length and egg weight, egg width and egg weight, shell weight and egg weight, shell weight and egg width, egg index and egg width, in all the three phenotypes compares favorably with (Yakubu *et al.*, 2008, Olawumi and Ogunlade, 2008, Yousria *et al.*, 2010). In this study egg width was indicated to be good estimator of egg shape index. Yannakopoulos and Tserveni-Gousi (1986) reported that egg shape index could be used as a criterion for determining stiffness of eggshell. Furthermore the values between egg width and egg length in naked neck and frizzle feathered chickens agreed with (Yakubu *et al.*, 2008; Olawumi and Ogunlade, 2008 and Yousria *et al.*, 2010).

The values recorded between shell weight and egg length in naked neck and frizzle agreed with (Olawumi and Ogunlade, 2008; Yousria et al., 2010). Also the result obtained between egg weight and shell weight, egg weight and shell thickness in naked neck is inconsonance with (Yakubu et al., 2008). Kul and Seker (2004) reported that egg weight has an indirect relationship with shell quality of the egg. Thus it has been stated by most of the researchers that the egg shell thickness has a direct relation with the egg weight (Choi et al., 1983) and has positive significant correlations with the shell weight (Farooq et al., 2001). It has been considered that the eggshell quality would be determined by using egg weight values due to the positive and significant correlation determined between the egg weight and shell thickness and the shell weight. Similarly, Ozcelik (2002) in his study had reported that the egg weight values would be used instead of determining the shell quality, because the shell thickness and the shell weight would be measured after breaking the egg. The phenotypic correlation value obtained between the egg weight and shell thickness in the study is higher compared to 0.05 reported by (Olawumi and Ogunlade, 2008) in exotic Isa brown layers and higher than 0.26 reported by (Stadelman, 1986). This implies that egg weight has a strong association with shell thickness in this phenotype of chickens. The values recorded between shape index and the shell weight in naked neck and smooth feathered types is consistent with

(Olawumi and Ogunlade, 2008). Therefore shell weight could not be considered as good estimator of egg shape index.

The negatively correlated values in this study recorded between albumin width and albumin height, in frizzle and smooth feathered agreed with (Olawumi and Ogunlade, 2008). Similarly, yolk index and yolk width is in agreement with (Yakubu *et al.*, 2008). Furthermore albumin and yolk height with haugh unit in all the three phenotypes is in agreement with (Yakubu *et al.*, 2008) in Nigerian smooth and naked neck chickens. Also the result obtained between haugh unit and albumin height in frizzle chicken agreed with (Olawumi and Ogunlade, 2008). Yolk width and yolk height in smooth feathered chickens disagreed with (Yakubu *et al.*, 2008). The values between haugh unit and yolk index in naked neck is in consonance with Kul and Seker (2004), in Japanese quails.

5.2.0 Effect of hen of different phenotypes on hatchability traits:

The mean hatchable egg weight in the present study was lower than values reported for Lohmann silver and Potchefstroom Koekoek breeds of chicken (Wondmeneh et al., 2011). Similarly the result is also lower than values reported for Barred Plymouth Rock, White Leghorn, Rhode Island Red (Islam et al., 2002) on Bangladesh local chickens. But the mean egg weight for most phenotypes compares favorably with figures reported for Horro and Fayoumi, breeds of chickens reported elsewhere (Wondmeneh et al., 2011). Furthermore the findings is higher than the values reported (Joseph and Oduntan, 1999; Fayeye et al., 2005) from unclassified Nigerian local chickens and Fulani- ecotype chicken respectively. The result is also slightly higher than values reported by (Yakubu et al., 2008) for naked neck chicken but, slightly lower than values reported for Smooth feathered chickens. The reason adduced for these differences could be the type of feed used and age of the chickens. Age, feed, protein level and temperature are some of the factors that affect egg size in chickens (Banerjee, 1992). Fertility (66.68%) obtained in this study is slightly lower than those reported by other authors (Islam et al., 2002; Fayeye et al., 2005; Wondmeneh et al., 2011). The lower fertility could be probably due to heat stress encountered during incubation, as greater part of the

study was carried out during the dry season (February –July). Similarly the fertility of an egg is affected by the factors directly related to the laying hen such as her ability to mate successfully, store sperm, ovulation and finally produced a suitable environment for the formation and development of the embryo (Brillard, 2003). Furthermore fertility also depends on the ability of the cock to mate successfully, quantity and quality of semen deposited (Wilson et al., 1979; Brillard, 2003), male and female ratio, age, preferential mating, lightening. Highest fertility, normal chicks and heaviest weight of the day old chicks recorded in this study by smooth feathered is in agreement with Sonaiya and Olori, (1989) who stated that,75% of the total chickens population in Nigeria are smooth multicolored and this suggest the supremacy of the phenotype over the others. The values obtained on hatchability on set eggs in this study is far lower than values obtained else where Islam et al. (2002) and it is also lower than values reported (Wondmeneh et al., 2011) on Ethiopian local chickens. The lower hatchability in this study was probably due to excessive thickness of the eggs. Poor hatchability generally in this study was probably due to fluctuation in environmental temperature, relative humidity and ventilation stress, which is a common phenomenon in this part of the world. Similar observations were made by (Bibek, 2003) elsewhere. Other factors that have considerable influence on hatchability include, nutrition of the breeding hens, genetic constitution of the embryo, disease, egg size, age and shell quality (King'ori, 2011). Egg weight, fertility, hatchability and late embryonic mortality varied greatly with feed regimes (Lariviere et al., 2009). Highest percentage of dead embryo obtained in the study is far higher than the results reported by (Islam et al., 2002), but tended to agree with (Singh et al., 1983) who reported 36.9% and 45.2% for white leghorn and white Plymouth Rock respectively. This is probably due to efficacy of the incubator used. Highest production of percent normal chicks in this study is slightly lower than (Wondmeneh et al., 2011), who reported 91.72%, 95.98%, 89.48% and 94.18% for Horo, Fayoumi, Lohmann, Silver and Potchefstroom Koekoek breeds of chickens respectively.

Live healthy chickens obtained in the study are lower than (Wondmeneh *et al.*, 2011). The authors obtained higher percentage of healthy normal chicks in

Ethiopian breeds. Increased production of sound chicks is an indication of successful hatchability. Average chicks weight recorded in the study is slightly lower than that reported elsewhere (Islam *et al.*, 2002; Wondmeneh *et al.*, 2011) for Bangladesh and Ethiopian breed of local chickens respectively. Normal feathered produced heaviest and healthiest chicks and this support the earlier work by Kalita (1994) and Abiola *et al.* (2008) who showed that the best values were achieved with medium size eggs and comparatively large size eggs always not resulted in heavier chicks and phenotypes may have a significant role (Islam *et al.*, 2002). Thus the result also signifies that chick weight was not just a function of egg weight, but it was also altered by genetic background. Egg weight is a phenotypic characteristic.

5.2.1 Correlation among hatchability traits:

The present result indicated significant positive correlations were obtained for all the phenotypes between fertility and dead in shell; hatchability on set eggs and hatchability of fertile eggs; hatchability on set eggs and normal chick; hatchability on set eggs and chick weight; hatchability on fertile eggs and chick weight and dead in shell and normal chick, the afore mentioned findings compare favorably with Islam et al. (2002), except fertility and dead in shell and dead in shell and normal chicks which contradicts (Islam et al. 2002), low relative humidity during incubation is associated with these types of hatchability traits. The non significant correlation found between the egg weight and all the hatchability parameters in all the three phenotypes studied, disagreed with (Islam et al. 2002) who reported significant positive correlation between egg weight and some hatchability parameters on Bangladesh chickens. Naked neck showed significant positive correlation than others for fertility and hatchability on set eggs, fertility and hatchability on fertile eggs, fertility and normal chicks, and fertility and chick weight. The result of the study is comparable with that of Islam et al., (2002) who reported on white leg horn and Rode Island Red. The negative value recorded between hatchability on set eggs and dead embryo and hatchability on fertile eggs and dead embryo recorded by Smooth feathered chickens in the study is similar to

the results obtained by (Islam *et al.*, 2002) on Barred Plymouth Rock, White Leghorn, Rhode Island Red and White Rock chickens elsewhere. On the contrast significant positive correlation obtained in the present study between fertility and dead embryo by Frizzle feathered chickens was compared favorably with those reported by (Islam *et al.*, 2002) on white Leghorn hens.

5.3.0 Experimental infection of three phenotypes of local chickens

The onset of clinical signs on the third and fourth day post infection recorded in this study agreed with the findings of Msoffe et al. (2002) and Fayeye et al. (2011). Similarly the onset of mortality recorded in this study concurs with the report of Msoffe et al. (2002). The variable mortality rates obtained among the three phenotypes tend to agree with Akinoluwa et al. (2012) who obtained 40%, 30% and 70% for Yoruba frizzle, Yoruba naked neck and Yoruba smooth feathered respectively. The findings disagreed with the report of Fayeye et al. (2011) who recorded 100% and 91% respectively for some Nigerian Fulani and Ilorin ecotypes. Similarly the research findings also contradicted Msoffe et al. (2002) on four ecotype chickens in Tanzania. The relatively higher mortality observed in this study for smooth feathered chickens compared to other phenotypes compared favorably with earlier report by Akinoluwa et al. (2012). The least mortality observed among the naked neck chickens is in harmony with El-Safty et al. (2006) who reported that naked neck has a better ability to secret Acute Phase protein (APP) secreted by liver cells, which gives protection to the birds against infection or any invasion. Furthermore, lower incidence of pathology was also observed in naked neck compared to other phenotypes and these suggest greater disease resistance associated with Na gene (Fraga et al., 1999; Gonzales et al., 1998). The duration of the disease among the three phenotypes observed in this study is within the range reported by Saidu et al. (2006) for three breeds of local chickens in Nigeria.

Higher mortality in males compared to female chickens recorded among the three phenotypes in this study contradicts Njagi *et al.* (2010), who reported that female chickens were more affected compared to males, but agrees with Kutubuddin (1973), who reported that male chickens were more affected with NDV than

females. Ezeokole et al. (1984), suggested that sex of birds may influence their morbidity and mortality. The differences could be due to the fact that in the present study all the phenotypes were infected using different routes of infection. Similarly this study also showed that mingling and housing NDV infected chickens and non infected can result to infection, the findings is in agreement with Akinoluwa et al. (2012) on Nigerian local chickens ecotypes elsewhere. The 80% mortality observed among the smooth feathered chickens in the intraocular route, which is further confirmed by GMT of HI antibodies and failure to develop protective immunity in the first seven (7) days of infection is in agreement with OIE (2000) that antibody titre less than Log₂ 2² may not be protective and this is probably the reason why this phenotype recorded highest mortality. The HI antibody titre recorded in this study with all the three phenotypes and through both routes of infection recorded except in smooth feathered group inoculated through intraocular routes is far higher than that reported by Akinoluwa et al. (2012) in three Yoruba ecotypes. The reasons adduced for this difference is that, in this study all the chickens were infected with NDV isolates and this triggers more antibody response and also in Akinoluwa et.al.(2012) reported that all the chickens had residual antibody pre contact, though not protective. This is probably interfering with the production of solid immunity.

CHAPTER SIX

SUMMARY, CONCLUSION, RECOMMENDATIONS AND CONTRIBUTION TO KNOWLEDGE

6.1 Summary:

All the egg quality traits were significantly affected by the type of phenotypes except yolk width and shell thickness. Frizzle feathered consistently produced heavier, longer, wider egg and albumin and heavier shell. Similarly frizzle naked neck produced higher albumin and yolk height and haugh unit. Furthermore naked neck produced higher egg width, egg and yolk index and yolk height. Egg weight, yolk height, yolk index, shell weight and haugh unit had a significant (p<0.01) effect on all the phenotype. Egg length and egg index, had a significant (p<0.05) effect on the phenotypes. Egg width, Albumin height and albumin width had a significance (p<0.001) effect on the phenotypes.

Frizzle and Smooth feathered chickens produced significant positive and negative correlations in egg quality traits. Naked neck produced significant positive correlation in all the egg quality traits studied.

All the hatchability traits were significant except hatchability on fertile egg and dead in shell. Smooth Frizzle feathered chicken produced highest percent fertility, percent normal chick and average chick weight. Where as Frizzle Smooth produced heaviest egg weight, percent fertility and dead embryo. Naked neck produced heaviest egg weight and dead in shell. Naked neck Frizzle produced highest percent hatchability on set and fertile eggs. Hatching egg weight had significant (P<0.001) effect on all the phenotypes studied. Fertility, Hatchability on set eggs, Dead embryo, Normal chick, Average chick weight had significant (P<0.05) effect on all the phenotypes. Naked neck and Frizzle produced significant positive correlation among the hatchability traits and Smooth feathered produced significant positive and negative correlations among the hatchability traits.

The results of experimental infection of three phenotypes with strain 33/56 NDV indicated that all the three phenotypes comprising of thirty chickens came down with the disease from the 3rd day after infection depending on the phenotype, except the in-contact chickens which developed the disease much later. Frizzle and Smooth had shorter incubation period of three days, while, Naked neck had a slightly longer incubation period of four days. Onset of mortality was on day 4, 5 and 5 for Frizzle, Naked neck and Smooth respectively. The percent mortality for the three phenotypes was 58.3%, 41.7% and 75% respectively for Frizzle, Naked neck and Smooth. The morbidity rate was 100% for all the phenotypes except the in-contact chickens. The duration of the disease was 7, 9 and 10 days for Frizzle, Naked neck and Smooth feathered respectively.

6.2 Conclusion:

The egg quality traits were significantly affected by the type of phenotypes except yolk width and shell thickness. Frizzle feathered consistently produced heavier, longer, wider egg and albumin and heavier shell. Similarly frizzle naked neck produced higher albumin and yolk height and haugh unit. Furthermore naked neck produced higher egg width, egg and yolk index and yolk height. Frizzle and smooth feathered chickens produced significant positive and negative correlations in egg quality traits and naked neck produced significant positive correlation in all the egg quality traits studied.

There was no significant effect of hatchable egg weight on all the three phenotypes and in all the hatchability parameters studied. On the contrast there was strong significant positive correlation between dead in shell and fertility, hatchability on set eggs and hatchability of fertile eggs, hatchability on set eggs and normal chicks, hatchability on set eggs and chick weight, hatchability on fertile eggs and normal chick, hatchability on fertile egg and chick weight, normal chick and chick weight in the three phenotypes studied. Furthermore there was also significant positive correlation between hatchability on set eggs and fertility, hatchability on fertile eggs and hatchability on set eggs, normal chick and fertility, chick weight and

fertility in naked neck chickens. Strong significant positive correlation was also established between dead embryo and fertility in frizzle chickens.

It could therefore be concluded that crossing between Frizzle and Naked neck produced better egg quality traits required. Similarly crossing between Smooth and Frizzle feathered chicken produced good fertility and hatchability traits and naked neck group and their crosses were the most resistant phenotype, followed by frizzle feathered and their crosses and the smooth feathered and their crosses were the least resistant and poor sero converts to NDV infection.

6.3 Recommendations

Since there is no single phenotype that combines the ability of good egg quality traits, fertility, hatchability, survivability and high egg production. It could therefore be recommended that frizzle naked neck chickens should be considered for table eggs, smooth frizzle chickens to be considered and integrated for hatching programmes. Naked neck and its crosses should be adapted in rearing programmes in rural areas as Newcastle virus resistant phenotype to avoid or reduce the effects of the number one killer disease.

Further studies should be carried out on why male chickens died more than the female ones.

6.4 **Contribution to Knowledge**

The research findings displayed the egg quality, fertility, hatchability and susceptibility status of local chickens in Adamawa state. The findings made it clear that Medium and Small size eggs are better in terms of albumin quality than the Jumbo size therefore, it empowers the egg buyers economically. It is possible to produce local day-old chicks in mass using locally fabricated kerosene incubators. The findings can equip farmers, local women and poultry keepers on which phenotype to be adapted for rearing in order to avoid seasonal outbreaks in our homes

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APPENDICES

APPENDIX 1: External egg characteristics.

(i)	Analysis	of	variance	for	egg	weight
(1)	7 Milary 515	OI	variance	101	55	WCISII

SV	DF	SS	MS	F	P	
Treatment	8	291.635	36.454	3.743	0.001	**
Error	63	545.363	9.739			
Total	71	907.037				

(ii) Analysis of variance for egg length

SV	DF	SS	MS	F	P	*
Treatment	8	1.886	0.236	2.203	0.041	
Error	63	5.994	0.107			
Total	71	8.820				

(iii) Analysis of variance for egg width

SV	DF	SS	MS	F	P	***
Treatment	8	0.678	0.085	4.707	0.000	
Error	63	1.008	0.018			
Total	71	1.799				

(iv)Analysis of Variance for egg index

SV	DF	SS	MS	F	P	*
Treatment	8	219.744	27.468	2.241	0.038	
Error	63	686.282	12.255			
Total	71	954.063				

(vi) Analysis of variance for shell weight

SV	DF	SS	MS	F	Sig	
Treatment	8	11.758	1.470	2.982	0.007	**
Error	63	27.598	0.493			
Total	71	41.789				

(vii) Analysis of variance for shell thickness

SV	DF	SS	MS	F	Sig	
Treatment	8	0.207	0.026	0.803	0.602	NS
Error	63	1.807	0.032			
Total	71	2.220				

APPENDIX 2: Internal eggs characteristics

$(I) \qquad \textbf{Analysis of variance for albumin height} \\$

SV	DF	SS	MS	F	Sig ***
Treatment	8	40.185	5.023	5.456	0.000
Error	63	51.559	0.921		
Total	71	103.898			

$(ii) \qquad \textbf{Analysis of variance for albumin width} \\$

SV	DF	SS	MS	F	Sig
Treatment	8	8.448	1.056	6.409	0.000
Error	63	9.227	0.165		
Total	71	18.931			

(iii) Analysis of variance for yolk height

SV	DF	SS	MS	F	Sig	
Treatment	8	0.423	0.53	3.221	0.004	**
Error	63	0.919	0.16			
Total	71	1.409				

(iv) Analysis of variance for yolk width

SV F Sig DF SS MS Treatment 8 0.147 0.18 0.235 0.982 NS 4.369 0.78 Error 63 Total 71 4.883

(v) Analysis of variance for yolk index

SVDF SS MS F Sig 0.003 Treatment 8 563.301 70.413 3.374 Error 63 1168.697 20.870 Total 71 1871.351

(vi) Analysis of Variance for Haugh unit

Sig ** SVDF SS MS F Treatment 8 1016.228 127.028 3.458 0.003 Error 63 2057.181 36.735 Total 71 3433.313

APPENDIX 3: Fertility and hatchability traits

(vii) Analysis of Variance for the Average egg weight (g)

SV	DF	SS	MS	F	Sig	
Treatment	8	535.178	66.897	7.400	0.000	**
Error	63	506.280	9,041			
Total	71	1076.177				

(viii) Analysis of Variance for Fertility

SV	DF	SS	MS	F	Sig	
Treatment	8	23733.984	2966.748	2.867	0.010	*
Error	63	57946.083	1034.751			
Total	71	83091.911				

$(ix) \qquad \textbf{Analysis of Variance for Hatchability on set eggs}$

SV	DF	SS	MS	F	Sig	
Treatment	8	8027.230	1003.440	2.573	0.018	*
Error	63	21841.968	390.035			
Total	71	33712.189				

(x) Analysis of Variance for Hatchability on fertile eggs

SVDF SS MS F Sig Treatment 8 0.166 NS 15543.534 1942.942 1.534 Error 63 70914..346 1266.328 Total 71 101232.080

(xi) Analysis of Variance for Dead embryo

SV DF SS MS F Sig Treatment 8 22051.553 2756.444 2.572 0.018 * Error 63 60020.705 1071.798 Total 71 93386.657

(xii) Analysis of Variance for Dead in shell

SV DF SS MS F Sig Treatment 8 10971.852 1371.481 1.435 0.202 NS Error 63 53506.097 955.466 Total 71 69752.423

(xiii) Analysis of Variance for Normal Chick

SVDF SS MS F Sig Treatment 8 36638.604 4579.825 2.863 0.009 ** Error 63 100775.37 1599.609 Total 71 137413.971

(xiv) Analysis of Variance for Abnormal Chick

Sig SVDF SS MS F Treatment 8 9721.270 0.01 1215.159 2.797 Error 63 27371.368 434.466 Total 71 37092.638

(xv) Analysis of Variance for Average Chick weight (g)

SV DF SS MS F Sig Treatment 8 3249.682 406.210 2.094 0.052 Error 63 10865.734 194.031 Total 71 16203.553

APPENDIX 4: Pelletized chicks feed 25kg/bag

S/n	Ingredients	Quantity (%)
1	Crude protein	18% (min)
2	Fat	6% (max)
3	Crude fibre	9% (max)
4	Calcium	1% (min)
5	Available phosphorus	0.40% (min)
6	Metabolizable Energy	2650kcal/kg (min)

Feed (ad-lib) for the first 9 weeks of age

APPENDIX 5: Pelletized Grower feed 25kg/bag

S/N	Ingredients	Quantity (%)
1	Crude protein	15% (min)
2	Fat	10% (max)
3	Crude fibre	10% (max)
4	Calcium	1% (min)
5	Available phosphorus	0.35% (min)
6	Metabolizable Energy	2550kcal/kg (mi

Feed from about 9 weeks of age

APPENDIX6: Pelletized Layer feed 25kg/bag

S/n	ingredients	Quantity (%)
1	Crude protein	16.5% (min)
2	Fat	4% (max)
3	Crude fibre	6.5% (max)
4	Calcium	3.60% (min)
5	Available phosphorus	0.40% (min)
6	Metabolizable Energy	2650kcal/kg (min

Feed from about 20 weeks of age till culling

APPENDIX 7: Challenged Virus information

Reagent Name: Newcastle Disease Challenge Virus

- i. **Strain or Source:** Hertz 33/56 Newcastle Disease Virus (Hertz 33/56)
- ii. **Intended Use:** For use as a challenge virus in Newcastle disease assay.
- iii. 4. LD₅₀ titre: 10^{7.5} per ml
- iv. **5. Instruction for Use:** The virus was diluted with phosphate buffered saline (PBS) of Ph7.2 at the ratio of 1:10 to give virus Lethal Dose fifty (LD50) of 10^{6.5} per mil. 0.2 mils of the dilution were used to challenge the chickens.
- v. 6. Storage Container Size and Type: Plastic cryogenic vial of 2mls
- vi. 7. Storage Conditions: Store at $-20^{\circ}\text{C} + 4^{\circ}\text{C}$
- **vii. 8. Technical Contacts:** For additional information, contact the Avian Viruses Research and Diagnostic Laboratory, Viral Research Division, National Veterinary Research Institute, Vom. **GSM:** 08037024280
- viii. **8. Method of Preparation:** The virus was prepared in 10-day-old chicken embryonated eggs from minimal disease free flock.

APPENDIX 8: Publication Certificates