THE EFFECT OF GOLDEN JUBILEE MAIZE (GJM) ON EGG PRODUCTION AND EGG QUALITY OF LAYER CHICKENS

By

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CERTIFICATION

I hereby certify that the work herein submitted as a thesis for the Master of Science (Animal Nutrition) Degree has neither been presented nor is being concurrently submitted for any other degree elsewhere. However, work of other researchers and authors which served as sources of information are duly acknowledged.

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DEDICATION

This research is dedicated to my lovely daughter, Precious Nayram Glover and my wife, Olivia Borbi

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ABSTRACT

A total of 192 Lohmann brown pullets of twenty- three weeks of age at a production level of 50 % and with a mean initial weight of 1.48 kg were randomly allotted to three isocaloric and isonitrogenous diets ie. Normal maize (NM), Obatanpa (OBAT), and Golden Jubilee maize (GJM) over a 33 week period to assess their relative nutritive values . Feed and water were given *ad libitum.* The proximate analysis of the maize samples showed that NM, GJM and OBAT had crude protein values of 10.0, 8.6 and 9.1 % respectively and dry matter content of 89 %, 90 % and 89.50 % for NM, GJM, and OBAT respectively. The results did not show any significant difference ($p>0.05$) in feed consumption among the dietary treatments though numerical differences were observed. Hens fed GJM laid 898 and 1,056 more eggs than those fed NM and Obatanpa respectively showing a significant difference ($p<0.05$) between OBAT and GJM but not significant ($p>0.05$) between NM and GJM. Though cost of feed consumed did not differ among the three dietary treatments, GJM recorded higher values for feed per kilogram egg, feed per dozen eggs, feed cost per kilogram egg and feed cost per dozen eggs produced, an indication of higher income when GJM is used in layer diet. The internal qualities measured such as Haugh Unit (HU) and pH were within the acceptable range indicating a good quality. The mean (HU) Scores were 85.57, 87.64 and 89.64 for NM, OBAT and GJM respectively. The keeping quality of the eggs did not show any significant difference ($p>0.05$) among the dietary treatments. However, there was a deterioration of HU, albumen height, egg weight, increment in pH value and yolk percentage with storage. The cholesterol level in the egg of hens fed GJM was lowest compared with that of NM and slightly higher than that of Obatanpa. The use of GJM in the diet of layer hens showed favourable results and will be useful to farmers for maximum egg production without compromising egg quality.

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LIST OF ABBREVIATIONS

ADE	Apparent Digestible Energy
AME	Apparent Metabolizable Energy
ANOVA	Analysis of Variance
AOAC	Association of Official and Analytical Chemists
CIDA	Canadian International Development Agency
CIMMYT	Centro Internacional de Mejoramiento de Maiz y Trigo
CP	Crude Protein
CRD	Completely Randomized Design
DE	Digestible Energy
DM	Dry Matter
EE	Ether Extract
FAO	Food and Agriculture Organization
FAOSTAT	Food and Agriculture Organization Statistical Database
FASDEP	Food and Agriculture Sector Development Policy
GE	Gross Energy
GH¢	Ghana cedi
GJM	Golden Jubilee maize
GOG	Government of Ghana
Hb	Haemoglobin
HDL	High-Density Lipoprotein
HPAI	Highly Pathogenic Avian Influenza
HU	Haugh Unit

IITA	International Institute for Tropical Agriculture
LDL	Low-Density Lipoprotein
LSD	Least Significant Difference
MCA	Millennium Challenge Account
MCHC	Mean Cell Haemoglobin Concentration
ME	Metabolizable Energy
MOFA	Ministry of Food and Agriculture
NFE	Nitrogen Free Extract
NM	Normal maize
NRC	National Research Council
OBAT	Obatanpa
PLA	Polylactic Acid
PUFA	Polyunsaturated Fatty Acid
QPM	Quality Protein maize
RBC	Red Blood Cell
SFA	Saturated Fatty Acid
TGS	Triglycerides
USDA	United States Department of Agriculture
WBC	White Blood Cell

CHAPTER ONE

1.0 INTRODUCTION

Maize is a major staple cereal crop in Ghana and contributes significantly to the diets of most people. It is also a major component of poultry and swine diets in Ghana (MCA, 2009). Maize is a primary source of energy and can contribute up to 30 percent protein, 60% energy and 98% starch in animal diets (Dado, 1999).

Among all the nutrients required for effective performance of monogastric animals, carbohydrate remains the most abundant nutrient supply in a balanced diet, constituting between 45 and 60% of finished feeds (Nestel,1975; Machin, 1992). Maize has remained the major source of energy supply in the diet of livestock and poultry industry in Ghana. In the diets of monogastric animals, maize constitutes between 50 and 60 % (Omage *et al.* 2009).

The normal maize variety in the diets of livestock and poultry has two significant limitations, firstly it is low in protein (9-10%) and secondly it is deficient in some essential amino acids especially lysine and tryptophan (Okai *et al.* 2005, Vasal, 2006). Maize as a result, is supplemented with protein-rich ingredients such as fishmeal, and soyabean meal which are expensive and are not always available (Osei *et al.*1994a, Okai *et al.* 2001) and synthetic lysine to make up for the deficient lysine.

Improving maize varieties to possess an improved balance of essential amino acids can reduce the dietary inclusion of protein-rich feed ingredients thereby reducing cost of feeding and production. Mertz *et al*. (1964) discovered opaque-*2* mutant gene in maize whose lysine and tryptophan content is about twice that of normal maize but inherent agronomic characteristics of this plasm, particularly its low yield and high

susceptibility to diseases and insects discouraged breeders from further studies (Zhai and Zhang, 2007).

Through several selections and trials, breeders at the International Maize and Wheat Improvement Center (Centro Internacional de Mejoramiento de Maiz y Trigo, CIMMYT) succeeded in combining the high- lysine potential of opaque-*2* gene with a sufficient number of modifier genes to change the original soft opaque-*2* endosperm into hard vitreous type called quality protein maize (Vasal *et al*. 1980). Quality protein maize (QPM) locally called Obatanpa meaning *Good Nursing Mother* is being used in several feeding trials by researchers all over the world. According to Osei *et al.* (1998), broiler chicks fed on Obatanpa (QPM) performed significantly better than those fed normal maize. Okai *et al.* (1992); Osei *et al*. (1993) indicated that instances where maize was the only source of protein, pigs offered Obatanpa ate more feed and grew significantly faster than those fed a normal maize variety.

The bulk of maize produced world-wide is yellow and three times as much is used for livestock feed as for direct human consumption (López-Pereira and Morris, 1994). However, in Ghana white maize is the predominant variety used by the human population and as feed for livestock and poultry. The increasing pressure on the white maize led to an escalating price of maize in Ghana with the current price between GHC 0.6 and GHC 0.65/kg,(Boateng and Adjei, Personal communication).

Yellow maize varieties are known to impart yellow colour to egg yolk and yellow pigmentation to the skin and shank of broilers. Unfortunately, Ghana produces limited amount of yellow maize which cannot meet the demand of Ghanaian poultry farmers (MCA, 2009).

The Crop Research Institute, Kumasi, in 2007, developed and released four new varieties of the QPM including Golden Jubilee. Golden Jubilee maize is a dent/flint yellow QPM variety with yield potential of 5 tonnes per hectare and matures between 105 and 110 days (Ewool *et al*. Unpublished). The golden jubilee maize is being promoted for the poultry and livestock industry in Ghana (Ghanaweb, 2007).

Much work has been done by researchers to evaluate the nutritive value of Obatanpa (Osei *et al*. 1994; Osei *et al*. 1996; Osei *et al*. 1999; Okai *et al.* 1994; Okai *et al.* 2001; Okai *et al.* 2005; Akuamoa-Boateng, 2002), however there is virtually no literature on the nutritive value of Golden Jubilee maize.

According to Ewool *et al*. (Unpublished) ten layer chickens fed diets containing Golden Jubilee maize (yellow QPM) laid 600 eggs with deep yellow yolk and harder egg shell compared to 224 eggs laid by those fed the diet containing normal white maize over a twelve week period.

This experiment sought to study the effect of Golden Jubilee maize on egg production and egg quality of layer chickens.

The Specific objectives were:

1. To determine the proximate compositions of Golden Jubilee maize.
2. To evaluate the nutritive value of Golden Jubilee maize in diets for laying chickens.
3. To assess the economics of egg production when GJM is added to layer diets.

CHAPTER TWO

2.0. LITERATURE REVIEW

2.1. Importance of maize (*Zea mays*)

Maize (*Zea mays*) is a major staple cereal crop in Ghana which is cultivated throughout the country but leading producers are mainly in the middle-southern part (transitional and forest zones) with an estimated 15 percent grown in the Northern Regions of the country (MCA, 2009). In 2007, the average yield per hectare was 1.5 metric tonnes for smallholder farmers, who rely on rain-fed conditions with limited use of improved seeds, fertilizer, mechanization, and post-harvest facilities and as high as 5.0-5.5 metric tonnes for farmers who use improved seeds, fertilizer, mechanization and irrigation (MCA 2009).

Maize is widely cultivated throughout the world and higher quantity of it is produced each year than any other cereal grain. The United States produces about 41% of the world's harvest (FAO, 2009). Other top producing countries are China, Brazil, Mexico, Indonesia, India, France, South Africa, and Argentina and Ukraine as shown in Table 2.1.

Worldwide production of maize in 2009 was 817 million tonnes harvested over 159 million hectares of land with a yield of over 5 tonnes per hectare. Africa produced about 7 percent of the world total maize production (FAO, 2009), as against a production of 12.5 percent of the global maize production in 2002 where 17.4 million hectares of land were cultivated (James, 2003). The major maize producing countries in Africa are Nigeria, South Africa, Ethiopia, Kenya, Tanzania, Congo, Mozambique

Zimbabwe and Ghana (James, 2003). Table 2.2 shows maize production statistics in Sub-Sahara Africa as reported by Pingali (2001).

Table 2.1: Top World Maize Producers in 2009

Country	Production (Tons)
United States	333,010,910
China	163,118,097
Brazil	51,232,447
Mexico	20,202,600
Indonesia	17,629,740
India	17,300,000
France	15,299,900
Argentina	13,121,380
South Africa	12,050,000
Ukraine	10,486,300
Asia	233,633,476
Europe	83,958,488
Africa	56,685,857
World	817,110,509

Source: Food and Agriculture Organization of United Nations, 2010.

Globally, over 60 percent of the maize produced is used as feed for animals and only 22 percent is used for direct human consumption with the remaining 18 percent used for production of ethanol and biofuel (Morris, 1998). However in Africa, about 64 percent of the maize produced is used for human consumption, 23 percent for animal feed and an estimated 13 percent for brewing (Morris, 1998). Maize serves as the main energy source for monogastric animals where its grain constitutes more than 50 percent by weight of their diets (NARP, 1993).

Table 2.2: Maize Production Statistics in Sub-Sahara Africa (1997-1999)

Country	Area harvested ('000ha)	Yield (t/ha)	Production ('000t)	Maize area as % of Total Cereal Area	Average per Capita Maize Consumption 1995-97 (kg/year)	Average Net Imports 1996-98 ('000t)
E- S Africa[1]	15,436	1.5	23,389	41	81	127
W- C Africa[2]	9,223	1.2	11,035	21	43	184
North Africa	1,192	5.4	6,402	10	74	4,892
D- C[3]	96,062	2.9	276,325	21	66	24,426
World	140,182	4.3	600,277	20	100	-

Source: Pingali (2001)

E-S :- [1]Eastern and Southern Africa, [2]W-C:- Western and Central Africa, [3]D-C:- Developing Countries

Maize contributes substantially to the total cereal grain production in the world economy as a trade, food, feed, and industrial grain crop (Pingali, 2001; FAO, 2002). The maize grain, leaves, stalk, tassel and cob are used for food and non-food products. Maize is the largest staple crop in Africa, Latin America and Asia (NRC, 1988). The maize grain is either consumed directly or changed into other forms as desired. The utilization of maize in Sub-Sahara Africa is depicted in Table 2.3.

Table2.3: Utilization of Maize in Sub-Saharan Africa

Sub-Sahara Africa region	Utilization kg/capita/year	Food use %	Feed use %	Other uses %
Eastern and Southern Africa	76	72	19	9
West and Central Africa	43	64	13	23
Africa	62	64	23	13
World	94	22	63	15

Source: Morris (1998)

Industrially, maize has several uses obtained from 'wet milling' process (Corn Refiners Association, 2002). It is used to prepare alcoholic beverages, fuel alcohol, syrups, sweeteners, jams, and jellies. Compostable plastics, packaging films, fast food

serving utensils are made with maize-based polylactic acid (PLA) (Nielson, 2003). Maize-based PLA can be blended with cotton wool and silk to make suits and pure corn-fibre wedding dress. The maize bran, germ and gluten obtained as by-products from the industrial processing of maize are used in compounding feed for animals (Gomez, 1992).

2.2. Maize as Food for Humans

Maize is a major staple with 90 percent being used for food, and serves as a major source of calories for 50 percent, 30 percent and 13 percent of the people in Southern, Eastern, West and Central Africa respectively (Pandey, 1998). Maize, though low in protein, contributes to both protein and calories needs of people especially in the rural areas where maize intake is considerably high (FAO, 1984). Table 2.4 shows maize intake and its calorie and protein contribution to the daily diet of humans in some selected countries.

Table 2.4: Maize Intake and its Calories and Protein Contribution to the Daily Diet

Country	Intake (g/person/day)	Calories (per person/day)	Protein (g/person/day)
Botswana	209.3	665	17.5
Cape Verde	334.1	1 052	28.0
Egypt	149.7	508	13.4
Guatemala	276.2	977	15.4
Honduras	255.9	878	22.8
Kenya	286.1	808	21.3
Lesotho	315.4	1002	26.4
Malawi	468.8	1422	37.6
Mexico	328.9	1061	27.1
Nicaragua'	131.0	472	11.1
Paraguay	131.2	445	11.6
Philippines	152.1	399	8.7
Romania	128.6	373	8.6
South Africa, Rep.	314.7	961	24.6
Swaziland	381.4	1279	33.7
Togo	136.9	411	10.8
Venezuela	118.3	339	7.4
Zambia	418.6	1226	31.3
Zimbabwe	330.9	958	25.2

Source: FAO (1984)

2.4. Nutritional Values of Different Types of Maize

Different types of maize have varied nutritional composition due to genetic and environmental factors. These factors influence the weight distribution and chemical compositions of the endosperm, germ and hull of the kernel. The different types of maize include; Salpor, Crystalline, Floury, Starchy, Pop and Black. (Cortez and Wild-Altamirano 1972). Table 2.5 shows gross chemical composition of different types of maize expressed in percentage.

Table 2.5: Gross Chemical Composition of Different Types of Maize

Maize type	Moisture	Ash	Protein	Crude fibre	Ether extract	Carbohydrate
Salpor	12.2	1.2	5.8	0.8	4.1	75.9
Crystalline	10.5	1.7	10.3	2.2.	5.0	70.3
Floury	9.6	1.7	10.7	2.2	5.4	70.4
Starchy	11.2	2.9	9.1	1.8	2.2	72.8
Pop	10.4	1.7	13.7	2.5	5.7	66.0
Black	12.3	1.2	5.2	1.0	4.4	75.9

Source: Cortez and Wild- Altamirano (1972)

2.4.1. Chemical Compositions of Parts of Maize Kernel

The parts of maize kerrnel are the pericarp (outer layer of the maize kernel), endosperm (the tissue that surrounds the embryo) and the germ (the reproductive part of the maize kernel) (Watson, 1987). The maize pericarp has crude fibre content of about 87 percent which is made up of mainly hemicelluloses (67 percent), cellulose (23 percent) and lignin (0.1percent). (Burge and Duensing, 1989). The endosperm contains high level of starch (87.6 percent) and 8 percent protein with relatively low crude fat. The germ has high content of crude fat averaging about 33 percent and very high level of protein (18.4 percent) and minerals. The aleurone layer (tissue found in the endosperm that surrounds the embryo) has about 19 percent protein and high crude fibre. The endosperm and the germ contribute larger proportion of the kernel nitrogen content in maize grain. The proximate compositions of main parts of maize

which indicate its nutritive value is of great importance when maize is processed for consumption (Bressani *et al*. 1990). Table 2.6 shows the proximate composition of main parts of maize.

Table 2.6: Proximate Composition of Main Parts of Maize Kernel (%)

Chemical component	Pericarp	Endosperm	Germ
Protein	3.70	8.00	18.40
Ether extract	1.00	0.8	33.20
Crude fibre	86.70	2.70	8.8
Ash	0.80	0.30	10.50
Starch	7.30	87.60	8.30
Sugar	0.34	0.62	10.80

Source: Watson (1987)

2.4.2 Carbohydrate Content of Normal Maize

Starch constitutes a major chemical component of the maize kernel (72 to 73 percent). Other carbohydrates present include, glucose, sucrose and fructose in amounts that vary from 1 to 3 percent of the kernel (Boyer and Shannon, 1987).

Maize starch accumulates in the endosperm tissue in the form of insoluble granules which are made of two glucose polymers: amylose (linear molecule) and amylopectin (branched molecule) (Pollak and Scott, 2005). Starch of dent or flint types of endosperm is made up of 25 to 30 percent amylose and 70 to 75 percent amylopectin while starch in waxy maize contains up to 100 percent amylopectin (Boyer and Shannon, 1987). Amylose is important in plant energy storage and is also an important thickener, water binder, emulsion stabilizer, and gelling agent in both industrial and food-based contexts (Zhong *et al*. 2006).

The digestible energy of maize starch ranges from 3.75 to 4.17 kcal/g dry matter which makes maize one of the highest energy cereal grains (Fetuga *et al.* 1979). Maize has relatively higher metabolizable energy compared to other cereals.

2.4.3 Oil and Fatty Acid Content of Normal Maize Grain

Maize oil provides a concentrated source of energy for animals (Pollak and Scott, 2005) and improves the palatability of the grain (Duffus and Slaughter, 1980). Tocopherol and carotenoids are the main lipids contained in maize grain. Germ oil provides relatively high levels of fatty acids (Weber, 1987; Bressani *et al.* 1990).

Carotenoids in yellow maize contain about 22 percent beta-carotenes and 51 percent cryptoxanthin (Squibb *et al.* 1957). The carotenes in the yellow maize are the precursor of vitamin A, and thus, helpful in overcoming vitamin A deficiency especially xerophthalmia which leads to childhood blindness (NRC, 1988). Yellow xanthophylls cause yellow pigmentation of skins and shanks of broilers and egg yolk (Squibb *et al.* 1957).

Maize oil contains about 10 percent palmitic, 2 percent stearic, 25 percent oleic, 62 percent linoleic acid and 1 percent linolenic acids with total of 12 percent saturated fatty acid (Pollak and Scott, 2005).

Refined corn oil is composed of 99 percent triacylglycerols with 59 percent polyunsaturated fatty acids (PUFA), 24 percent monounsaturated fatty acids and 13 percent saturated fatty acids (SFA) (Corn Refiners, 2002). Corn oil is a good source of ubiquinone, alpha-and gamma-tocopherols which function to activate a yellow enzyme called cytochrome reductase, which plays an important role in oxidation processes involving the transfer of energy in the cells of the body. It is also necessary

for maintaining the structure and function of muscles and of peripheral blood vessels (Adom and Liu, 2002).

Maize oil has cholesterol lowering effect because of its high content of polyunsaturated fatty acids (Adom and Liu, 2002).

Maize has the highest total antioxidant activity (181.42 micromol of vitamin C equiv/g) of grain compared with wheat, oats and rice with 76.70 micromol, 74.67 micromol and 55.77 micromol respectively (Adom and Liu, 2002). The high total antioxidant activity in maize is due to bound phytochemical which could help survive stomach and intestinal indigestion to reach the colon explaining partly the mechanism of grain consumption in the prevention of colon cancer (Adom and Liu, 2002).

Low level of linolenic acid coupled with high level of antioxidant activity in maize prevents rancidity in storage (Martinez *et al.* 1996). Gǖdǖ *et al.* (2008) reported that at the same inclusion level of different dietary oils, layers fed on diet containing maize oil laid eggs with thin shells indicating that high maize oil is detrimental to egg production.

2.4.4. Protein content of normal maize grain

Protein content varies in common varieties from about 8 to 11 percent of the kernel weight, most of which is found in the endosperm (Akalu *et al*. 2001) The maize endosperm is made up of different fractions, such as 7 percent albumins, 5 percent globulins and 6 percent non-protein nitrogen (Landry and Moureaux, 1982). The prolamine fraction which is soluble in isopropanol contributes 52 percent of the nitrogen in the kernel with prolamine 1 or zein 1 found in the largest concentration, about 42 percent , with 10 percent provided by prolamine *2* or zein *2* percent (Landry and Moureaux,1980, 1982).

All fractions other than zeins are balanced in amino acid content and are quite rich in lysine and tryptophan (Salamini *et al*. 1983). Zeins are devoid of lysine and tryptophan (Nelson *et al*. 1965), they reduce the distribution of these essential amino acids from the other types of endosperm which are collectively referred to as non-zeins (Vasal, 2000; Prasanna *et al*. 2001). The zeins and non-zeins constitute the prolamine which predominate the total protein in maize kernel. The zein fraction in normal maize normally contains higher proportion of leucine (18.7%), phenylalanine (5.2%) isoleucine (3.8%), valine (3.6%) and tyrosine (3.5%), but smaller amounts of other essential amino acids such as threonine (3%), histidine and cysteine (1%), methionine (0.9%), lysine (0.1%) and is essentially devoid of tryptophan as it is absent from the major prolamin fraction (a-zeins) of maize kernel (Vasal, 2000).

Considering the whole maize kernel, the essential amino acid content is a reflection of the amino acid in the protein of the endosperm (Bressani, 1971). Germ proteins contribute a relatively high amount of certain amino acids, although not enough to provide a higher quality of protein in the whole kernel (Bressani, 1971). The essential amino acids content of germ protein and endosperm protein is shown in Table 2.7.

Table 2.7: Essential Amino Acid Content of Germ Protein and Endosperm Protein

Amino acid	Endosperm		Germ	
	mg %	mg/g N	mg %	mg/g N
Tryptophan	48	38	144	62
Threonine	315	249	622	268
Isoleucine	365	289	578	249
Leucine	1 024	810	1 030	444
Lysine	228	180	791	341
Total sulphur amino acids	249	197	362	156
Phenylaianine	359	284	483	208
Tyrosine	483	382	343	148
Valine	403	319	789	340

Source: Orr and Watt (1957)

Due to the low level of protein in normal maize grain, maize based diets fail to supply the needed essential amino acids such as lysine and tryptophan for both human and animals and are supplemented with other protein rich sources to augment its nutritive value (Bressani *et al.* 1971).

2.4.5 Mineral content of normal maize

The concentration of ash in the maize kernel is about 1.3 percent (Bressani *et al.* 1962). The germ is relatively rich in minerals, with an average value of 11 percent as compared with less than 1 percent in the endosperm. The germ provides about 78 percent of the whole kernel minerals. The most abundant minerals in the maize grain are phosphorus and potassium. However, the phosphorus in normal maize is found in a bound form (phytate) which is not available to monogastric animals as they lack the enzyme phytase to break down the phytate to liberate the phosphorus (Maner, 1975). All of the phosphorus is found in the embryo, with values in common maize of about 0.90 percent. As with most cereal grains, maize is low in calcium content and also low in trace minerals. Table 2.8 below depicts the mineral content of maize.

Table 2.8: Mineral Content of Maize

Mineral	Concentration (mg/100 g)
Phosphorus	299.6 ± 57.8
Potassium	324.8 ± 33.9
Calcium	48.3 ± 12.3
Magnesium	107.9 ± 9.4
Sodium	59.2 ± 4.1
Iron	4.8 ± 1.9
Copper	1.3 ± 0.2
Manganese	1.0 ± 0.2
Zinc	4.6 ± 1.2

Source: Bressani, *et al*. (1962)

2.4.6. Vitamin content of maize grain

Maize grains contain both fat-soluble and water-soluble vitamins concentrated mainly in the aleurome layer and the endosperm. The fat-soluble vitamins consist of provitamin A (carotenoids) and vitamin E (Squibb *et al.* 1957). The carotene is mainly found in the yellow maize grain with beta-carotene content ranging from 6.4 to 11.3 µg per gram and with vitamin A activity of 1.2 to 2.6µg per gram (Squibb *et al.* 1957). The carotenoids however are susceptible to destruction after storage. Watson, (1987) reported values of 4.8 mg per kg carotenoids in maize which decreased to 1.0 mg per kg after thirty-six months of storage.

The water-soluble vitamins are found mostly in the aleurone layer of the maize kernel. Maize grain contains a considerable amount of thiamine, riboflavin, panthothemic acid, pyridoxine and choline sufficient enough to satisfy requirement of most livestock (NRC, 1988). Maize grain contains a pyridoxine level of 2.69 mg per kg (Yen *et al.* 1976).

Maize contains low level of niacin with an average of about 20 µg per gram. This however, is in a bound form (niacytin) and therefore not available to monogastric animals (Christianson *et al.* 1968). High incidence of pellagra (disease caused by dietary deficiency of niacin) in countries where maize consumption is considerably high is also associated with low level of available niacin (Gopalan and Rao, 1975; Patterson *et al*. 1980).

2.5. Improving protein quality of normal maize

The low nutritive value of maize with respect to its protein quality could be improved upon, thereby improving the biological utilization of the nutrients it contains. This could be done by genetic manipulation, processing and fortification.

Genetically, the protein content of maize could be increased from 10.9 to 26.6 percent in the high-protein strain after 65 generations of selection (Dudley, *et al*. 1974). Dudley, *et al*. (1974) and Dudley *et al*. (1977) demonstrated that the protein content of standard inbred lines could be increased by crossing with the high protein strain. Woodworth and Jugenheimer (1948) concluded that total protein content could be increased by selection in an open-pollinated variety or by crossing standard inbred lines with high protein strain followed by backcrossing and selection in segregating populations.

Processing maize could help achieve the maximum potential nutritional value of maize. Processing though could lead to losses when optimum conditions are exceeded; it stabilizes nutrients in the maize grain (Bressani, 1983). Lime-cooking of maize induces important nutritional changes by improving upon the calcium, amino acids and niacin content of the maize (Norad *et al*. 1986).

Lime-cooking involves the addition of one part whole maize to two parts of approximately I percent lime (Calcium hydroxide) solution and the mixture heated to 80°C for 20 to 45 minutes and then allowed to stand overnight (Khan *et al*. 1982). The cooking liquor is decanted and the maize is washed two or three times with water to remove the seed-coats, the tip caps, excess lime and any impurities in the grain. This process converts the maize in to tortillas.

Due to the use of calcium hydroxide in converting maize into tortillas, the calcium content of the product increases significantly, up to about 400 percent (Braham and Bressani, 1966). In a study by Bressani and Scrimshaw, (1958) using in *vitro* enzymatic digestions with pepsin and trypsin they reported that the amount of alpha-amino nitrogen (a measure of total amino-acids) which form the building blocks of protein as a percentage of total digested nitrogen was twice as high from tortilla (43 percent) as from maize (21.4 percent) and levels of histidine, isoleucine, leucine, lysine methionine, phenylalanine, threonine and tryptophan were higher from the tortilla hydrolysate than from maize indicating the amino acid balance induced by lime cooking. Pearson *et al*. (1957) also reported that lime cooking destroyed the pellagragenic factor in maize suggesting that lime treatment resulted in the release of bound niacin in maize. Natural fermentation of maize, results in B-vitamin concentration and protein quality (Wang and Field, 1978).

Maize based diets can be fortified by the addition of protein rich sources supplemented with synthetic amino acids in small quantities to boost the protein quality and amino acid content of such diets (Bressani, 1988). Blending maize based diets with an appropriate animal protein source; plant isolates and oil seed products help overcome the nutritional shortfalls of maize based diets. Protein complementation of legumes and oilseed cakes can be done to improve the methionine level of such sources by addition of maize grain which contains enough of methionine .(Bressani, 1988)

2.6. Development of quality protein maize (QPM)

The high consumption of maize by the human population in a number of countries especially in Latin America and Africa and the well-established lysine and tryptophan deficiencies in maize motivated the search for maize kernel with higher concentration of these essential amino acids in its protein. The first breakthrough in this endeavour was the discovery of the effect of the opaque-*2* and floury-*2* mutant on lysine and tryptophan content in maize endosperm protein (Mertz *et al.* 1964). The new maize, opaque-2 had the same amount of protein as conventional maize (Bressani, 1988). The initial feeding trials indicated that the new maize could significantly reverse protein deficiency, malnutrition as well as prevent pellagra (Harpstead, 1971, Pradilla *et al*. 1975).

The discovery of opaque-*2* however, came with several drawbacks of the opaque-*2* maize germplasm. Noticeable among them were reduced yield, soft grain, with a chalky appearance; slower dry-down in the field and higher susceptibility to pest and disease than the normal maize. These inherent agronomic defects of the opaque-2, discouraged many breeders from further investigation (Vasal *et al*.1980). Eventually, researchers at the International Maize and Wheat Improvement Center (CIMMYT) successfully combined the high-lysine potential of the *opaque-2* gene with sufficient number of modifier genes to change the original soft *opaque-2* endosperm into hard vitreous type called quality protein maize (Vasal *et al*. 1980).

Quality protein maize has superior lysine content (0.43 percent compared to 0.23 percent in normal maize) and high yield and agronomic characteristics similar to those of normal maize (Ortega *et al.* 1986). Osei *et al*. (1999) also reported lysine level of 0.24 and 0.32 percent for NM and QPM respectively. The QPM has smaller, denser,

harder kernel than the normal maize grain. The QPM also has faster drying time compared to the *opaque-2* thereby reducing its possibility of going mouldy.

2.7. Agronomic characteristics of QPM

The major agronomic characteristics of QPM include grain yield, kernel type, moisture content and resistance to diseases and pests, drought tolerance and storage characteristics (NRC, 1988). QPM have yields comparable to NM counterparts in many locations in the world (NRC, 1988).

There was a change in kernel of opaque-2 when converted into QPM. The trait for soft opaque, floury endosperm was changed into hard vitreous type (Vasal, 1980). The endosperm is tightly packed with few spaces around the starch granules and as shiny and transparent as those of traditional flint or dent maize varieties (NRC, 1988). The hard vitreous type kernel of QPM is resistant to pest infestation.

Excessive moisture of the mature *opaque-2* maize, about 2-4 percent higher than that of normal varieties prolongs the drying time making it more susceptible to mould infestation (NRC, 1988). QPM varieties have little of the slow drying characteristics that limited *opaque-2*. It dries at a rate comparable to that of normal maize varieties but still shows high incidence of ear rot (Vasal *et al*. 1980).

2.7.1 Changes in Chemical Composition and Nutritive Value during Grain Development

Changes in chemical composition of maize grain upon maturation are important. Ingle *et al.* (1965) reported a decrease in nitrogen, crude fibre and ash on a dry-weight basis and an increase in starch and ether extract. The alcohol-soluble proteins increase rapidly as the kernel matures, while acid-soluble and alkali-soluble proteins decrease. Arginine, isoleucine, leucine and phenylalanine (expressed as mg per g N) increase,

while lysine, methionine and tryptophan decrease with maturation (Gómez *et al.* 1992). They also reported a decrease in protein quality upon maturation.

2.8 Nutritional value of QPM

The low protein quality of normal maize stems mainly from deficiency of essential amino acids-lysine and tryptophan (Vasal, 1980). QPM retains in essence all of the high quality nutritional components that were in the *opaque-2* maize. QPM grain has the same amount of protein as common maize but twice the useable protein because the quality and biological value of its protein is much higher than that of the normal maize (Young *et al*. 1971). The normal maize grain protein has 40 percent of the biological value of milk protein while that of QPM protein is about 90 percent of that of milk protein (Bressani *et al.* 1969).

The biological value of QPM protein was also studied by Young *et al*. (1971). Egg protein was used as reference, fed at intakes of 2.64 to 3.95 g nitrogen per day. The authors calculated true protein digestibility and biological value from the faecal metabolic nitrogen and urinary endogenous nitrogen. The protein digestibility of *opaque-2* maize protein varied from 67 to 106 percent, with an average for the eight individuals in the study of 92 percent, while the variability for egg protein was from 78 to 103 percent with an average of 96 percent. The average biological value for QPM was 80 percent, and for egg the average was 96 percent as shown in the Table 2.9

Table 2.9: The relative quantities of maize protein

Quality measures	**Maize**		**QPM Protein Scale to Egg albumin, %**
	Normal	QPM	
True Protein	82-91	92	96
Digestibility / Biological value	40-47	80	96

Source: Young *et al.* (1971)

QPM has impact on human health and on the productivity of swine and poultry. It serves as an excellent source of quality protein for malnourished children (Pradilla *et al*. 1975). CIMMYT, (1985) showed that babies in the second year of life grew normally when QPM was fed as the only source of protein.

The nutritional benefits of QPM for people who depend on maize for their energy and protein needs are quite significant (Prasanna *et al*. 2001). Quality protein maize has superior protein quality and protein digestibility over normal maize (Paes and Bicudo, 1995; Graham *et al*. 1980). QPM also has higher niacin content and lower leucine content than the normal maize (Graham *et al*. 1980). The yellow version of QPM has considerable level of carotenoids which helps overcome xeropthalmia, a vitamin A deficiency that is a primary cause of childhood blindness in many developing countries (NRC, 1988).

2.9 Factors affecting the chemical composition of maize grain

The chemical composition of maize is affected by both genetic (Mertz *et al*. 1964) and environmental factors (Bressani *et al*. 1962).

2.9.1. Genetic factors

Genetic variability for most traits in maize is high and amenable to enhancement (Prasanna *et al*. 2001). Maize plants accumulate starch in endosperm which is subject to genetic manipulation. The waxy gene in waxy maize controls amylopectin in the endosperm up to 100% with very low amount of amylose (Creech, 1965). The amylose extender gene increased the amylose fraction of the starch from 27 percent to 50 percent (Vineyard *et al*. 1958). Maize varieties with waxy or sugary genes have better nutritional value for monogastric animals due to their greater digestibility of the starch they produce (Sandstead *et al*. 1968).

The protein content of the different varieties could be increased by genetically manipulating the genes from 10.9 percent to 26.6 percent (Dudley *et al*. 1974). Apart from the starch and protein, maize oil can also be increased by genetic influence (Jellum and Marion, 1966; Leibovit and Ruckentein, 1983), Dudley and Lambert, (1969) reported an increase in maize oil from 4 percent to 15 percent by increasing the size of the germ, where the oil is concentrated.

2.9.2. Environmental factors

Nitrogen content of the soils could be enhanced by the addition of appropriate level of nitrogen fertilizer which improves upon the protein in maize. Nitrogen application of maize increased protein due to increases in prolamine (Tsai *et al*. 1980; Tsai *et al*. 1983). Mitchell *et al*. (1952) reported that increase in the zein fraction of high protein maize resulted in high protein content but lower biological value.

2.10. Development of Obatanpa (QPM)

Following the successful discovery of QPM with hard kernel, good taste and other consumer flavouring characteristics by the researchers at CIMMYT, Ghanaian Scientists at the Crop Research Institute (CSIR), Fumesua, improved upon the nutritional value of maize in the early 1990s.

The CSIR was assisted by Canadian International Development Agency (CIDA), the International Institute for Tropical Agriculture (IITA), the CIMMYT and the Sasakawa Global 2000 (SG2000) to evaluate the vast and diverse amount of CIMMYT's QPM materials including open-pollinated varieties, inbred lines and experimental hybrid in several parts of Ghana. They succeeded in developing a new improved Quality Protein Maize (QPM) variety and named it Obatanpa, a Ghanaian word in Twi meaning "A Good Nursing Mother" (Twumasi-Afriyie *et al*. 1992).

Obatanpa, an intermediate maize variety, has about 10 percent protein just like the normal maize. It has 70 percent higher level of the essential amino acids, tryptophan and lysine. QPM has nutritive value of about 90 percent that of milk protein while the corresponding value for NM is only about 40 percent (Akuamoa-Boateng, 2002). The grain yield of QPM is almost the same as that of the normal maize. Table 2.10 shows the mean yield grain of QPM and normal maize.

Table 2.10: Mean grain yield of QPM and NM Varieties

Variety	Yield(tonnes/ha)
Obatanpa (QPM)	3.1
Aburotia (NM)	2.6
Okomasa (NM)	3.3
Abeleehi (NM)	2.7
Dobidi (NM)	3.3

Source: Twumasi-Afriyie *et al*. (1992)

2.11. Nutritional Evaluation of QPM

Nutritional evaluations of QPM in various locations have proved the superiority of QPM over normal maize in the feeding of various categories of animals (Ortega *et al*. 1986; Sproule *et al*. 1988; Sullivan *et al*. 1989; Burgoon *et al*. 1992; Osei *et al.* 1999; and Zhai, 2002). In all these studies, the lysine level was relatively higher compared to that of normal maize. The lysine content of QPM ranged from 0.32 to 0.43 percent and 0.24 to 0.33 percent for the normal maize and the tryptophan content ranged from 0.06 to 0.10 percent as shown in Table 2.11. Methionine levels in both maize varieties were below 0.30 percent. Apart from Sproule *et al.* (1988), all the other researchers recorded similar values for both NM and QPM. The ether extract of QPM was relatively higher compared with that of NM. As indicated in Table 2.11, ether extract of NM ranges from 4.2 to 4.48 percent while values of 5.10 and 5.12 percent were

reported for QPM. The crude protein content of NM was reported to range from 8.92 to 11.00 percent for NM and 9.8 to 11.3 percent for QPM in all the studies.

Table 2.11: Comparison of the nutritional composition of quality protein maize (QPM) and normal maize (NM) (dry basis)

Source	Ortega *et al.* 1986		Sproule *et al.*1988		Osei *et al*. 1999		Zhai , 2002	
	NM	QPM	NM	QPM	NM	QPM	NM	QPM
GE, MJ/kg	-	-	17.39	17.26	14.71	16.76	18.85	18.80
CP %	9.8	9.8	11.0	11.3	8.92	9.11	-	-
EE %	-	-	4.2	5.1	4.48	5.12	-	-
CF %	-	-	-	-	1.92	2.14	-	-
Ash %	-	-	1.3	1.6	1.90	1.60	-	-
NFE%	-	-	72.3	72.3	71.52	71.37	-	-
Lys %	0.27	0.43	0.28	0.42	0.24	0.32	0.33	0.43
Trp %	0.06	0.10	-	-	0.06	0.08	-	-
Met %	0.22	0.21	0.28	0.19	0.19	0.18	0.19	0.18
Cyst %	-	-	-	-	0.19	0.25	-	-
Ala %	0.82	0.68	0.78	0.62	-	-	0.72	0.69
Arg %	0.42	0.75	0.49	0.66	0.40	0.50	0.44	0.52
Asp %	0.62	0.78	1.51	1.62	-	-	0.64	0.78
Glu %	1.94	1.77	2.11	1.67	-	-	1.92	1.80
Gly %	0.37	0.55	0.40	0.47	0.34	0.42	0.42	0.46
Hist %	0.33	0.47	0.31	0.42	-	-	0.37	0.49
Ile %	0.36	0.36	0.38	0.35	0.34	0.31	0.36	0.33
Leu %	1.34	0.96	1.45	1.00	1.18	0.92	1.21	0.98
Phe %	0.54	0.47	0.53	0.51	0.46	0.39	0.50	0.39
Pro %	0.78	0.83	1.02	1.04	-	-	0.80	0.97
Ser %	0.53	0.55	-	-	0.48	0.45	0.40	0.40
Thr %	0.38	0.45	0.35	0.38	0.29	0.31	0.34	0.34
Tyr %	0.33	0.41	0.45	0.35	-	-	0.51	0.45
Val %	0.50	0.57	0.57	0.55	0.46	0.49	0.43	0.43

2.11.1. Energy availability

Guang-Hai *et al*. (2006), found no significant differences in gross energy (GE), apparent metabolizable energy (AME) for poultry and apparent digestible energy (ADE) for pigs when fed NM and QPM. The results showed that though NM has higher GE content than QPM, its AME for poultry and ADE for pigs were lower than those of QPM indicating that available energy from QPM is a little higher than that from NM (P>0.01).

2.11.2. Evaluation of QPM in humans

The nutritional and biological evaluation of QPM in human beings conducted in Ghana and elsewhere, proved the superiority of QPM over NM. In a metabolic-balance trial by Viteri *et al*. (1972) as cited in NRC, (1988) children, who consumed maize as the only source of protein, increased their nitrogen retention by 50-100 percent when they switched from NM to nutritionally improved maize. These increases could translate into equivalent rates of weight gain, growth in stature and protection from the manifestation of protein deficiency. Braham and Bressani, (1966) found no significant differences in nitrogen retention among children fed diets based on milk and on alkali-processed *opaque-2* maize.

Akuamoa-Boateng, (2002), in infant feeding trials in Ghana, reported no significant difference between the mean weight gain of children fed on QPM and NM varieties over a 12-month period. The weight gains reported were 2.92 and 2.93 kg for Obatanpa and Normal maize. The results, however, showed significant differences in average height in favour of QPM (14.76 and 12.37 cm for Obatanpa and Normal maize respectively). These results were in agreement with earlier results by Graham *et al*. (1980); Graham *et al.* (1990) and Bressani *et al*. (1990). Singh *et al.* (1980): Singh and Chandra (2010), however, reported that in preschool children fed QPM continuously for six months showed a significant increase in weight and arm circumference with marginal increase in height. The report also indicated that QPM fed to pregnant women from first trimester up to last trimester showed very good impact upon the health of the babies and mothers.

In the study of QPM on convalescent malnourished children, (Graham *et al*. 1980; Pradilla., *et al.* 1975) reported that children were restored to normal health when fed with nutritionally improved maize.

2.11.3. Poultry trials

Various experiments with poultry have shown the superiority of QPM over NM (Osei *et al*. 1999; Bai, 2002; Gao, 2002, Zhai, 2002). Broilers fed with QPM-based diets performed significantly better than their counterparts fed the NM-based diet. They consumed more feed and used it more efficiently to attain live body weight of 708 g after 8 weeks while those fed the NM-based diet weighed 532 g over the same time period (Okai *et al.* Unpublished). Osei *et al*. (1998), in an experiment where QPM was the sole source of protein, observed that, QPM-fed birds on the average consumed 14 percent more feed and gained weight at 1.7 times that of the NM group and the feed conversion efficiency was 20 percent better in the QPM group, but noted that QPM cannot be the sole source of protein to broilers because birds which received a balanced diet significantly out-performed those on the QPM diet.

In laying hen trials, Zhai, (2002), noted that replacing NM with QPM significantly enhanced feed intake, 113.95 g and 116.69 g for NM+lysine and QPM. The results also showed enhanced egg production as QPM-fed hens recorded 90.97 percent as against 89.63 percent by those fed NM+lysine but recorded no noticeable effect on egg weight (59.21 and 59.07 for NM+lysine and QPM respectively), FCR, soft and broken eggs or Haugh unit. In addition, Osei *et al.* (1999) carried out an evaluation of QPM for layer pullets in two phases:

1. Grower phase (8 to 18 weeks)
2. Layer phase (from 19 to 51 weeks)

The results of the grower phase suggested that when QPM was added to pullet diets, protein level could be reduced to 14 percent without any adverse effects on their performance. The performance of birds fed NM was lower compared to those on

QPM. The addition of QPM to layer diets had significant effect on the age at first egg, age when 50 percent egg production was achieved and on the daily production of housed hens. Pullets fed QPM diets laid their first egg at mean age 117 days ($P<0.01$), attained 5 percent egg production at a mean age of 124 days ($P<0.05$) and 50 percent egg production at mean age of 130 days ($P<0.05$), all of which occurred earlier than those fed NM-based diet. Pullets fed NM-based diet laid their first egg at a mean age of 123 days ($P<0.05$) and attained 50 % egg production at a mean age of 141 days ($P<0.05$).

These results indicated that QPM could be used in layer diet to cut down cost on the use of fishmeal and results in good financial benefits without compromising performance.

2.11.4. Pig trials

In experiments conducted by (Okai *et al*. 2001; Okai *et al*. 2003), where QPM was used in complete diets for starter-finisher pigs and phase feeding of pigs, the results showed that the growth performance and carcass traits were similar ($P<0.05$). In effect, there was no significant difference ($P> 0.05$) in the carcass dressing percentage of pigs. These results were similar to earlier experiment conducted in other countries (Maner *et al*.1971; Knabe *et al.* 1992).

Sullivan *et al*. (1989) and Burgoon *et al.* (1992) also reported similar findings where there was no significant difference in the carcass dressing percentage where QPM was used as a replacement for NM. Guang-Hai *et al.* (2006), found no significant effect on back fact thickness contradicting earlier findings by Jin *et al*. (1998) that using QPM rather than NM in the diet could decrease back fat thickness. However, Okai *et al*.

(2001) reported a reduction in the back fat thickness as the level of fishmeal in the diet was reduced from 14 percent to 10 percent.

2.12. Economic assessment of QPM

In animal feeding, QPM provides a cheaper source of obtaining balanced animal feed which can be calculated in monetary terms. Lopez-Pereira (1993), showed relatively modest reduction in feed cost for poultry; 2.8 percent for broilers and 2.6 percent for layers when QPM to was used to compound their diets.

Nyanamba *et al.* (2003), in a study carried out in Kenya, found a 5 percent cost reduction from substituting QPM for normal maize in broiler ration. Okai *et al*. (2001) reported about 12.4 percent reduction in feed cost in Obatanpa-based diet due to the corresponding decrease in inclusion level of fishmeal, without any significant adverse effects on growth performance of pigs. Osei *et al*. (1994b) in broiler experiment, reported as high as 29.4 percent reduction in feed cost when the QPM inclusion level was increased with corresponding decrease in fishmeal.

Obatanpa-based diet therefore provides savings as a result of reduction in feed cost due to lower inclusion rate of expensive protein sources without compromising performance of the animals.

2.13. Development of Golden Jubilee Maize (GJM)

Golden Jubilee Maize is a yellow variety of Obatanpa developed from QPM germplasm using the conventional maize breeding methods. It was bred to have high level of carotene giving it the yellow appearance. GJM is an intermediate maturing (105-110 days), dent/flint QPM variety with a yield potential of 5tonnes per hectare. It has about 10 percent protein with 0.2 µg/g and 0.8 µg/g of alpha-carotene and beta-

carotene respectively and pro-vitamin A activity of 1.2µg/g (Ewool *et al*. Unpublished).

Yellow maize varieties give deeper yellow colour of egg yolk when fed to poultry. Golden Jubilee maize was therefore developed to meet the demand of the poultry farmers and also help to boost the poultry industry in Ghana (Ewool *et al*. Unpublished). It is resistant to pest and diseases and has a desirable lodging tolerance. Table 2.12 compares some chemical contents of Golden Jubilee maize and local white variety.

Table 2.12: Some chemical contents of Golden Jubilee Maize and local white variety

Variety	Protein	Fat	Beta-cryptoxantin	Alpha-carotene	Beta-carotene	Provitamin A
	…… %.....		……µg/g……			
GJM	9.8	3.3	0.6	0.2	0.8	1.2
NM	11.8	3.0	0	0	0	0

Source: Ewool *et al*. (unpublished)

Ewool *et al*. (Unpublished) ten layer chickens fed diets containing Golden Jubilee maize (yellow QPM) laid 600 eggs with deep yellow yolk and harder egg shell compared to 224 eggs laid by those fed the diet containing normal white maize over a twelve week period. According CIMMYT (2004), layer chickens fed yellow QPM laid more eggs than those fed the normal maize. The report also indicated that the hens fed yellow QPM were healthier.

2.14. Inferences from Literature Review

Most poultry and pig farmers use normal maize varieties because of its desirable nutritional characteristics such as high energy content, high palatability and digestibility as well as its low fibre content (NRC, 1988). Normal maize varieties

though deficient in lysine and tryptophan, contribute some proportion of protein in the diets of poultry.

Maize-based diets are supplemented with rich protein sources to meet the protein requirement of poultry or fortified with synthetic amino acid boost the lysine and tryptophan levels. Identifying desirable traits in *opaque-2* and *floury-2* with elevated level of lysine and tryptophan by Mertz *et al.* (1964) was a great breakthrough to improving upon the lysine and tryptophan deficiencies of normal maize variety.

Successful combination of maize high-lysine potential of the opaque-2 gene with sufficient number of modifier genes to change the original soft opaque-2 endosperm into hard vitreous type led to the breeding of quality protein maize (QPM) (Vasal *et al.* 1980). The lysine and tryptophan content of the opaque-2 is about twice that of normal maize (NRC, 1988).

Research conducted by several researchers in Ghana and elsewhere have shown the nutritional superiority of QPM over normal maize in animal (monogastric) nutrition (Osei *et al.*1994a*:* Osei *et al.*1994b Osei *et al.* 1994c; Osei *et al.* 1999; Okai *et al.* 2001; Okai *et al.* 2005; Sproule *et al.* 1988 and Knabe *et al.* 1992;) and in human nutrition (Bressani *et al.* 1980; Pradilla *et al.* 1975; Graham *et al.* 1980; Graham *et al.* 1990; Akuamoa- Boateng, 2002).

In addition the economic benefit of using QPM in the diet of monogastric animals is quite significant due to the reduction of fishmeal in the QPM-based diets (Lopez-Perera, 1993; Osei *et al.* 1994; Okai *et al.* 2001; Nyanamba *et al.* 2003)

The initial trial of Golden Jubilee Maize (GJM) by Ewool *et al.* (Unpublished) indicated that layers fed GJM-based diet laid more eggs than their counterparts fed

with NM. This was attributed to the higher lysine and tryptophan which might have contributed to a better feed efficiency. Again, the results also showed hens fed GJM laid egg with harder egg shell with deep yellow egg yolk.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Study Area

The experiment was carried out at the Poultry Section of Animal Science Department of Kwame Nkrumah University of Science and Technology (KNUST), Kumasi. The experiment was conducted for thirty-three weeks, from December 2009 to August 2010. The study area is located within the semi-deciduous humid forest zone of Ghana characterized by bimodal rainfall pattern with annual rainfall of 1300mm. Daily temperatures range from 20^{o}C to 35^{o}C with average of 26^{o}C. The relative humidity varies from 97 percent during the morning of wet season to as low as 20 percent during the late afternoon in the dry season. (Meteorological report, Unpublished).

3.2 Source of Maize and Experimental Diets

Normal maize (NM), Obatanpa (OBAT), and Golden Jubilee maize (GJM) used to formulate the experimental diets were provided by Alpha Seed Company Ltd, Kumasi. Other ingredients including fishmeal, soybean meal, oyster shell and vitamin premix were purchased from the open market in Kumasi.

Each maize type was ground in a hammer mill with a sieve mesh size of 70 mm. The diets were designated as NM, OBAT, and GJM for normal maize, Obatanpa and Golden Jubilee maize respectively.

The diets were formulated to meet all requirements for essential nutrients as recommended by the National Research Council (NRC, 1988). All diets were formulated to be isocaloric and isonitrogenous. Table 3.1 shows the dietary and nutrient composition of the layer diets fed throughout the experimental period.

Table 3.1: Composition and nutrient content of Layer Diets

Ingredients	NM%	OBAT%	GJM%
NM	56	-	-
OBAT	-	56	-
GJM	-	-	56
Fishmeal	7	7	7
Soyabean meal	11	11	11
Wheatbran	17	17	17
Oyster shell	8	8	8
Premix	0.5	0.5	0.5
Common salt	0.5	0.5	0.5
Total	**100**	**100**	**100**
Calculated compositions			
Crude protein %	17.24	17.13	16.80
Calcium	3.14	3.14	3.14
Phosphorus	0.48	0.48	0.48
Lysine	0.84	0.91	0.91
Cystine	0.27	0.31	0.31
Tryptophan	0.14	0.22	0.22
Methionine	0.57	0.56	0.56
ME/kgcal/kg	2630.34	2630.34	2630.34
Analysed composition (%)			
Crude protein	16.70	16.90	16.80
Dry matter	89	90.00	89.50
Ether extract	6.00	2.50	4.00
Crude fibre	2.63	2.62	2.63
Ash	10.0	9.50	10.50

Vitamin Trace Mineral Premix: Inclusion rate is 2.5kg/tonne.

To supply Vit. A = 8000, 000 IU, Vit. D = 500,000 IU, Vit. E = 2,500mg, Vit. K_3 = 1000mg Vit. B_2 = 2000mg, Vit. B_{12} = 5mg, Folic Acid = 500mg, Nicotinic Acid = 8000mg, Calcium Panthotenate = 2,000mg, Choline Chloride = 50,000mg, Manganese = 50,000mg, Zinc = 40, 000mg, Copper = 4, 500mg, Cobalt = 100mg, iodine 1000mg, Selenium = 100mg

3.3 Source of Experimental birds

One hundred and ninety-two Lohmann brown pullets of twenty-two weeks old were acquired from Akate Farms in Kumasi at the production level of approximately 50 percent (Hen-day production).

3.4 Housing

The pullets were raised in open-sided deep litter house partitioned into pens measuring 1.82 m by 1.75 m by 0.75 m giving a floor space of 0.20 square metres per bird. The pens were thoroughly cleaned and disinfected. Wood shavings were spread on the floor about 5 cm in depth to provide litter for the birds. Feed trough, water trough, and laying nest were placed at vantage points for the birds to have easy access. The house was roofed with galvanised aluminium roofing sheets in a gable style.

3.5 Feeding and Watering

The birds were fed with diet containing normal maize for four days to remove residues of previously fed diet from their digestive tract.

Feed was given *ad libitum.* The birds were fed twice daily (7 am and 4 pm) .The water troughs were washed every morning and fresh clean water offered to the birds *ad libitum.*

3.6. Management practices

The water troughs were washed daily and feed troughs cleaned every week after the left over feed was measured. The pens were cleaned periodically by removing the cobwebs and the droppings at the back of the pens every two weeks by scooping it with a spade. The wood shaving used as litter material was changed every six weeks or anytime it became wet. The birds were dewormed every 28 days using *Pepirazine citrate.* The laying nests were also sprayed to get rid of fowl lice.

3.7. Parameters measured

The parameters measured included feed consumption, egg weight, feed conversion efficiency (feed per dozen eggs, feed per kg egg), initial and final body weight, haematology, and egg yolk colour, egg pH, whipping volume of whole egg, yolk weight, shell weight, and albumen weight. Other parameters such as shell thickness, shell, yolk and albumen percentages, albumen height, hen-day egg production, hen-house egg production, egg biochemical parameters, economics of production and mortality were also taken.

3.8. Feed consumption

At the end of every week, the feeding trough was emptied and the orts weighed. The weight of the leftover feed was then subtracted from total feed offered to get the weekly feed intake. Daily feed intake per replicate was determined by dividing the weekly intake by seven, which is the number of days making one experimental week. Daily feed intake per bird was also determined by dividing the result of the daily feed intake per replicate by the number of birds in each replicate.

3.9. Egg production

Eggs were collected twice daily, morning (8 am) and in the evening (5 pm). The eggs were weighed and sorted according to their sizes. Cracked eggs, soiled eggs and misshapen eggs were recorded as they occurred.

Hen-day egg production was calculated as the number of eggs produced as a percentage of the number that should have been produced if each of the birds alive produced one egg each day. This was recorded on daily basis. The hen-house production was also measured as the number of eggs collected daily and expressed as a percentage of birds at the beginning of the laying phase.

$$Hen-day\ production\ (\%) = \frac{Number\ of\ eggs\ produced}{Number\ of\ hen-days} \times 100$$

$$Hen-house\ production\ (\%) \quad = \frac{Number\ of\ egg\ produced}{Number\ of\ hens\ housed \times Number\ of\ days} \times 100$$

3.10 Feed per kilogram Eggs

Feed per kg egg is the amount of feed consumed in kg to produce 1kg of eggs. This was calculated by dividing the total feed intake in kg per each replicate by total egg weight in kg for that week.

$$Feed/kg\ egg = \frac{Total\ feed\ intake\ (kg)}{total\ egg\ weight\ (kg)}$$

3.11 Feed per Dozen Eggs

This is defined as the quantity of feed consumed to produce a dozen eggs. It was calculated by dividing total feed intake by the number of dozen eggs and was recorded in grams. The total egg number divided by 12 gives the dozen eggs.

3.12 Albumen Height

Eggs were weighed using electronic digital scale (OHAUS, USA) with 400 g/0.01 g precision and then broken unto a Petri dish and the albumen height measured at its widest part at a position half way between the yolk and the outer margin using adjustable tripod micrometer (Plowright Hinton Limited, (P.H Ltd), United Kingdom) with 0.01 precision.

3.13. Albumen Weight

The albumen weight was determined as the difference of egg weight, yolk weight and shell weight (Parmar *et al.* 2006).

Albumen weight (g) = whole egg weight – (yolk weight+ shell weight)

3.14 Haugh Unit

The Haugh unit is a measure of egg protein quality (egg freshness) based on the height of its albumen (Monira *et al*. 2003). It can be calculated if the albumen height and the weight of the egg are known.

The Haugh unit score was computed using the relation; **HU = $100\log_{10}$ ($h-1.7W^{0.37}+7.6$)** proposed by Raymond Haugh in 1937, where **HU** = Haugh unit, **h** = albumen height, **W** =egg weight where **0.37, 1.7** and **7.6** are constants. The log scale is used in HU because the albumen height declines with storage in a logarithmic manner (Haugh, 1937).

3.15 Egg pH

The pH also measures egg freshness. The pH of both fresh and aged whole egg (eggs stored for 7 and 21 days), yolk only, and albumen only were measured using a pH meter manufactured by Suntex manufacturers (Suntex pH/mv/temp.Meter) at the Soil Science Department laboratory, KNUST. The eggs were broken and the yolk carefully separated from the albumen into a beaker and the pH electrode gently lowered in the content to measure its pH. Whole eggs were broken into a beaker and whipped before measuring the pH. In all a total of 642 eggs were used for the analysis.

3.16 Yolk colour

The yolk colour was determined using the Roche Colour Fan (Dynamic Source Manufacturing Inc. (DSM), Switzerland). The eggs were cracked open at the centre onto a Petri dish placed against plain background. The colour fan was brought near the Petri dish and the yolk colour was scored by visual comparison with the various colours of the Roche colour fan and the number of the particular colour fan which

corresponded with the yolk colour was then recorded for that replicate. A total of 214 eggs per treatment were analysed.

3.17. Yolk weight

At each sampling time, the eggs weight was measured within 2-4 hours of being laid (fresh) and after storage at room temperature and refrigerator for 7 and 21 days. In all, a total of 642 eggs consisting of 214 eggs per treatment were sampled.

The yolk was separated from the albumen, rolled on a wet paper to remove any trace of albumen and chalaziferous membrane before weighing using the digital electronic weighing scale (OHAUS, USA) with 400 g/0.01 g precision at the Department of Animal Science, KNUST.

3.18. Shell weight

The shell was washed and air dried for 24 hours and weighed using the digital electronic weighing scale at the Animal Science Department KNUST.

3.19. Shell thickness

The egg shell thickness was measured at three different random points in the equatorial shell zone after the shell membranes had been removed using micrometre screw gauge (Kalkum Ezquerra, Spain) (precision 0.01mm). A total of 642 eggs were analysed and the calculated mean was then used.

3.20 Storage quality

The eggs collected were weighed and refrigerated at a temperature of 4-6^{o}C for 7 and 21 days and also stored in a well ventilated room at room temperature of 21-25^{o}C. Parameters such as egg weight, yolk weight, albumen height and weight, and egg pH

were measured. 16 eggs per treatment were sampled at a time and repeated for 5 times making a total of 80 eggs per treatment.

3.21 Egg protein and cholesterol

Biochemical analysis was carried out on the yolk and albumen fraction of whole egg to determine the total cholesterol using the calorimetric end point method as described by Richmond (1973) and the total protein using the Biuret method as described by Henry *et al*. (1974). At the last quarter of the experiment, 48 eggs consisting of four eggs per replicate were sampled to analyse for their protein and cholesterol levels.

3.22. Haematology

Blood samples of 48 birds, consisting of four birds selected at random from each replicate were taken at the beginning and at the end of the experiment by wing vein puncture to measure the haematological and biochemical parameters. Red blood cell, Haemoglobin, White blood cell, Mean cell haemoglobin, Mean cell haemoglobin concentration, Mean corpuscular volume, Total protein, Albumin and Cholesterol.

Approximately 5ml of blood per bird were drawn into vacutainer tubes containing ethylene diamine tetracetate anticoagulant to prevent blood clotting. Haematological parameters were measured using the auto-haematology analyser, Sysmex KX-2IN (Sysmex Corporation, Japan). All samples were kept in refrigerator at a temperature of 4-6^{o}C until utilised.

3.23. Mortality

Mortalities during the experimental period were recorded as they occurred. Post-mortem examinations were carried out on the birds at the Animal Science Department and at the Ashanti Regional Veterinary laboratory in Kumasi to determine the cause of death.

3.24. Data analysis

Data collected were subjected to statistical analysis using analysis of variance (ANOVA) for completely randomised design using general linear model procedure of GenStat Statistical package Discovery Edition 3 (VSN International). Least Significant Difference (LSD) was used to determine significant differences among treatment means at ($P<0.05$).

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Proximate composition of Normal maize (NM), Obatanpa (OBAT) and Golden Jubilee maize (GJM)

The proximate compositions of the three maize varieties are shown in Table 4.1.

Table 4.1 Proximate composition of NM, OBAT and GJM

Parameter (%)	NM	OBAT	GJM	LSD	Level of sig.
CP	10.00	8.60	9.10	3.91	ns
E E	4.00	4.90	5.50	3.65	ns
CF	1.06[a]	1.04[a]	1.58[b]	0.07	*
Ash	1.50[ab]	2.00[b]	1.00[a]	0.70	*
Moisture	11.00	10.00	10.50	1.76	ns
Dry matter	89.00	90.00	89.50	1.76	ns
NFE	72.44	73.46	72.32	-	-

Each value is the mean of triplicate determinations.

a,b: Means in a row with different superscripts are significantly different *($p<0.05$)

CP; Crude Protein, EE; Ether Extract, CF; Crude Fibre, NFE; Nitrogen Free Extract.

The two quality protein maize varieties; Obatanpa (90 %) and Golden Jubilee maize (89.5 %) were slightly higher in dry matter content than the normal maize (89 %). These differences were, however, not statistically significant ($P>0.05$). Obatanpa and Golden Jubilee recorded lower crude protein and higher ether extract compared with normal maize. These findings are in agreement with earlier findings by Dei (1997), who reported 89.3 and 88.7 percent dry matter for QPM and NM respectively and Burgoon *et al.* (1992) who reported 88.9 percent and 87.3 percent for QPM and NM and concluded that QPM contains higher dry matter, crude protein, crude fibre and ether extract than normal maize.

There were significant differences (P<0.05) in CF and Ash content among all the dietary treatment. The values recorded for CF were 1.06, 1.04 and 1.58 for NM, OBAT and GJM respectively. The ether extract content recorded in this study showed non-significant differences (P>0.05) among all the varieties. The values recorded in this study however compare favourably with the findings of Osei *et al*. (1999) who reported values of 4.2 for NM and 5.12 percent for QPM and Sproule *et al*. (1988) who reported 4.2 and 5.1 percent for NM and QPM respectively.

4.2 Feed intake

As shown in Table 4.2, the different dietary treatments did not show any significant differences (P>0.05) in the mean daily feed intake of the birds. Birds on GJM however, recorded slightly higher value of 118.75 g/bird/day compared with NM and OBAT which recorded118.50 and 117.50 g/bird/day respectively. These values are similar to what was reported by Osei *et al.* (1999) that birds fed QPM-based diets consumed between 112.3 and 121.3 g while those fed NM-based diets consumed between 120 and 121.1 g. Zhai (2002) reported that hens fed QPM-based diet significantly (P<0.10) consumed more feed than those fed NM. Consumption for QPM and NM+lys was 116.69 and 113.95 g and attributed the difference to a possible appetizer in QPM.

The largely non-significant differences in feed intake among the dietary treatments in this study however, could be attributed to the same energy levels of the dietary treatments used as most animals tend to eat to satisfy their energy requirement (Pond *et al*. 1995). Dietary energy level informs feed intake in chickens (Scott *et al*. 1982). Low dietary energy level results in higher feed intake of chicken to meet their physiological needs. The depression in feed intake of birds fed on OBAT is unclear

since the energy level of the diets were the same. This may however be open for further study.

Table 4.2 Effect of dietary treatments on the performance of laying hens

Parameters	NM	OBAT	GJM	LSD	Level of significance
Mean Total egg laid	2357[ab]	2320[b]	2580[a]	240.50	*
Total egg weight (kg)	135.40	132.00	146.20	14.30	ns
Mean egg weight (g)	57.43[a]	56.91[ab]	56.67[b]	0.56	*
Feed/dozen egg (g)	2178	2179	1976	241.80	ns
Feed/kg egg	3.16	3.19	2.91	0.36	ns
Total feed cost (GH¢)	221.40	219.00	221.30	6.63	ns
Feed cost/dozen egg (GH¢)	1.13	1.13	1.03	0.13	ns
Feed cost/kg egg	1.64	1.66	1.51	0.19	ns
Hen-Day production %	66.80[b]	66.00[b]	71.00[a]	2.62	*
Hen-house production %	63.70[ab]	62.80[b]	69.80[a]	6.48	*
Total feed intake (kg)	425.00	420.40	424.80	12.73	ns
Mean feed intake (g/day/bird)	118.50	117.50	118.75	4.15	ns
Haugh unit score	85.57	87.64	89.64	6.35	ns
Shell thickness	0.343	0.352	0.362	0.03	ns
Initial body weight (kg/bird)	1.49	1.47	1.48	0.36	ns
Final body weight (kg/bird)	1.74	1.71	1.67	0.07	ns
Weight gain (kg)	0.25	0.24	0.19	0.09	ns
Mortality (%)	4.69	4.69	3.13	-	-

a,b,: Means in a row with different superscripts are significantly different *($P \leq 0.05$)

4.3 Egg production

The pattern of egg-lay in terms of (hen-day and hen-house production) was not consistent, but hens fed GJM recorded higher egg production than those fed Obatanpa and NM.

Figure 4.1 shows the total eggs laid by the chickens on the various dietary treatments over the thirty-three week period of the study. Hens fed GJM laid 10,320 compared with 9422 and 9264 by those fed NM and OBAT diets, sixty four (64) birds were involved per treatment. There was a significant difference ($P<0.05$) between total eggs laid by GJM and OBAT-fed hens but there was no significant ($P>0.05$)

difference between those laid by GJM and NM-fed. Zhai (2002) reported that replacing NM by QPM significantly ($P<0.10$) enhanced egg production. In a study by Osei *et al*. (1999) where varying levels of QPM were fed to pullets, pullets fed QPM2 diet containing 58.5 percent QPM laid their first eggs at mean age of 117 days ($p<0.01$), and attained 5 percent egg production at a mean age of 124 days ($P<0.05$) and 50 percent egg production at mean age of 130 days ($P<0.05$) all of which occurred a week earlier than the other dietary treatments. Pullets fed NM-based diet laid their first egg at a mean age of 123 days ($P<0.05$) and attained 50 % production at a mean age of 141 days ($P<0.05$). CIMMYT (2004), reported that layer chickens fed yellow QPM in Nicaragua, laid more eggs and exhibited better health condition than those fed normal maize.

Both GJM and OBAT would have enhanced egg production as both are QPM varieties and would have contained higher levels of lysine and tryptophan as indicated by Ortega *et al*. (1986), Sproule *et al*. (1988), Osei *et al*. (1999), and Zhai (2002). The superior performance by birds fed GJM-based diet over that of OBAT is therefore difficult to explain. Further chemical analysis of GJM may be carried out to ascertain the reason for which birds fed GJM laid most eggs.

These differences, notwithstanding, the results agree with a preliminary work by Ewool *et al*. (Unpublished) who reported that ten hens fed GJM laid a total of 600 eggs compared with 224 eggs laid by those fed normal white maize over a twelve-week period.

Though hens fed GJM laid more eggs, their mean egg weight was relatively smaller compared to those fed Obatanpa and normal maize. The mean shell thickness values of 0.343, 0.352 and 0.362 mm for NM, OBAT and GJM respectively were not

significantly different (P>0.05) among the dietary treatments. The findings however agree with Dei (1997), who reported values of 0.351 to 0.360 mm for laying hens and compare favourably with the results of other researchers, (Nelson, 2003; Diarra and Usman, 2008; Vashan *et al.* 2008; and Moula *et al*. 2009) who reported values in the range of 0.312 to 0.380 mm. The average egg shell thickness of a fowl is about 0.33 mm (Oluyemi and Roberts 1992).

The results on the other hand, showed that shell thickness decreased with increase in the egg size which explains the reason why hens fed GJM recorded thicker egg shell having laid most eggs with relatively smaller egg size. Iken *et al*. (2002), reported that newly improved yellow dent maize in Nigeria contain higher calcium (73.3 mg/100g) than the local white maize (43.6 mg/100g). This could possibly support yet another reason GJM fed hens laid eggs with thicker egg shell though not significant.

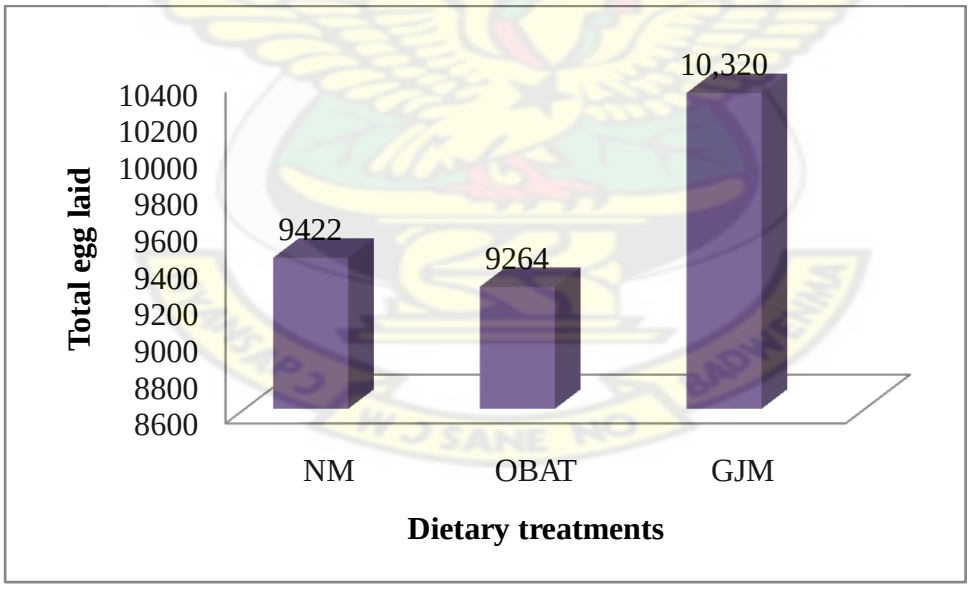

Figure 4.1: Effect of Dietary treatments on total egg laid

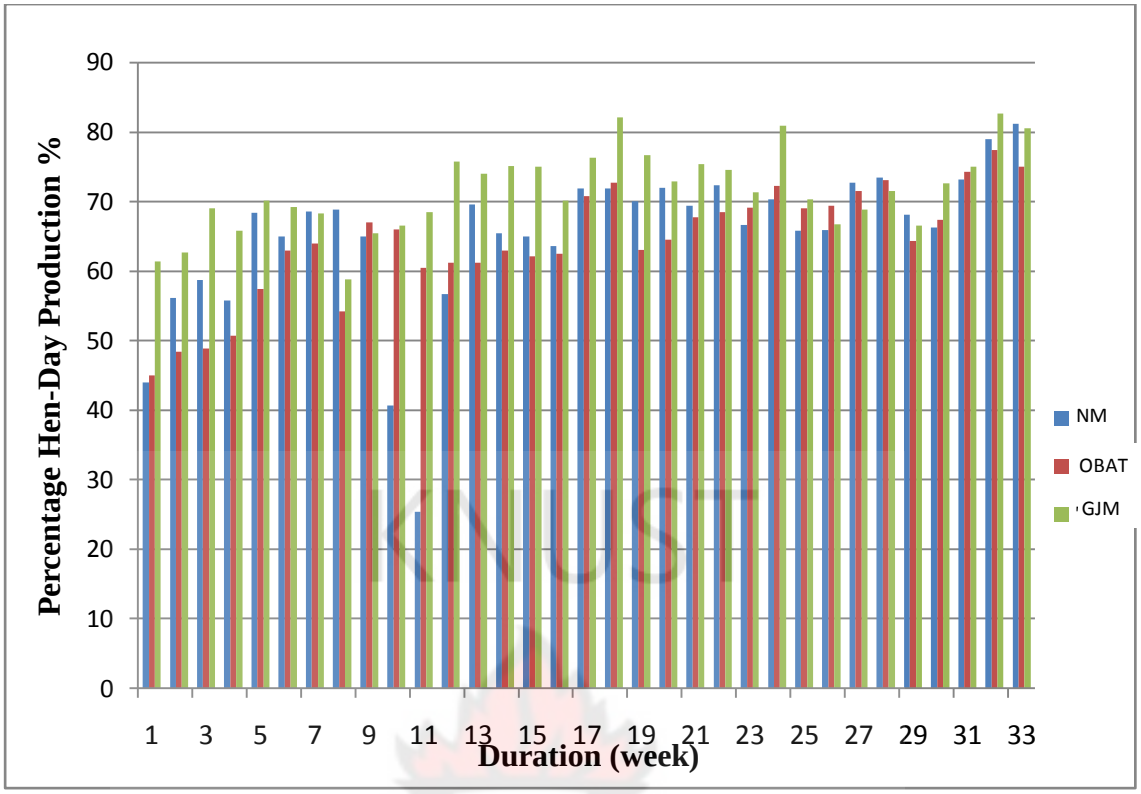

Figure 4.2: Effect of Dietary Treatments on Hen-Day Egg Production

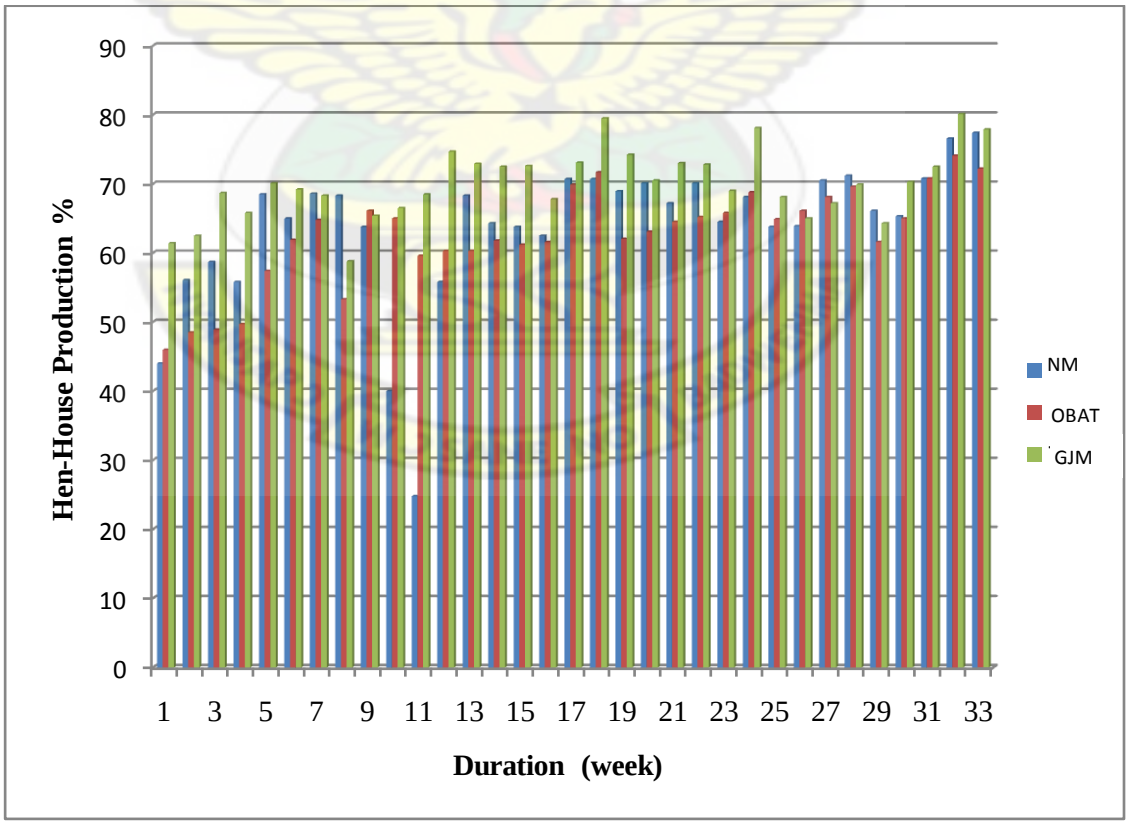

Figure 4.3: Effect of Dietary Treatments on Hen-House Egg Production

4.4 Feed conversion ratio

Feed consumed per kilogram egg produced presented in Table 4.2 was not significantly different (P>0.05) among the dietary treatments. However, numerically, hens fed GJM recorded better feed conversion value of 2.91 compared with those fed NM and OBAT which recorded values of 3.16 and 3.19 respectively. These values were higher than what was reported by Osei *et al.* (1999) who reported values between 1.99 and 2.09 for QPM and NM in that order. Hens on GJM consumed less than 2000 g (1976 g) feed to produce a dozen eggs while their counterparts fed NM and OBAT consumed 2178 and 2179 g respectively to produce a dozen eggs. Consequently, hens fed with GJM showed better feed cost per kilogram egg produced as well as feed cost per dozen egg produced due to higher egg production. The results of the current study, therefore, suggest that GJM when used in the ration of layers could improve upon the income of poultry farmers and should be encouraged.

4.5 Mortality

Throughout the experimental period, 8 mortalities were recorded; 3 each for birds fed NM and OBAT and 2 for those fed GJM. Of the eight, seven were due to pecking while only one case of mortality was attributable to bacterial infection (*Coli bacillosis*).

4.6 Egg Quality

Table 4.3 shows the effect of dietary treatment on egg quality. The albumen height was greatest (8.80 mm) during the first 8 weeks and lowest (6.58 mm) at week 32. Mean egg weight also increased with the age of the hens resulting in the decrease of Haugh unit with the age of the hens.

It was also observed that larger eggs recorded lower albumen height hence lower HU. The Haugh unit values for the various treatments did not show any significant differences (P>0.05) throughout the study period. This result agrees with Zhai, (2002), and Osei *et al*. (1999) who found no statistical difference in HU of hens fed either QPM or NM. Similar results were reported by (Silversides and Villeneuve, 1994; Jones and Musgrove, 2005).

The egg pH also had no significant effect (P>0.05) among the treatments. The various dietary treatments recorded almost same values for the whole egg pH that is 7.44, 7.41 and 7.40 for NM, OBAT and GJM respectively. The albumen and yolk pH were also not statistically different (p>0.05). The normal pH of fresh egg yolk is about 6.00 while that of albumen ranges from 7.6 to 8.5 (Heath, 1977). The values recorded in this study for egg yolk were 5.92, 5.95 and 5.96 while the albumen pH values were 8.89, 8.90 and 8.94 for hens fed NM, OBAT, GJM-based diets. This finding agrees with an earlier report by Moula *et al*. (2009) who reported that the pH of fresh egg yolk ranges from 5.93 to 6.03. However, the albumen pH value recorded were higher compared to what was reported by Moula *et al*. (2009) who indicated albumen pH values of 7.99 to 8.06. Factors which accounted for the difference could however not be explained.

The shell percentage (weight of egg shell expressed as a percentage of the weight of whole egg) showed a significant difference (P<0.05) between NM and GJM for the first 112 days of the study and showed no significant difference (P>0.05) thereafter. This difference in the shell percentage for the first 112 days was due to the larger size of eggs laid by hens fed NM-based diet. The values recorded throughout the study period tended to increase with the age of the hens.

Table 4.3. Effect of dietary treatments on egg quality parameters

Parameters		Dietary Treatments				Level of Significance
	Hen age	NM	OBAT	GJM	LSD	
Mean egg weight (g)	30	56.87	53.99	53.54	3.54	ns
	38	58.38	566.62	57.00	5.37	ns
	46	59.67	59.54	55.44	5.50	ns
	54	59.35	59.19	58.19	4.00	ns
Shell Thickness (mm)	30	0.336^{b}	0.372^{a}	0.372^{ab}	0.04	ns
	38	0.352	0.352	0.351	0.04	ns
	46	0.332^{ab}	0.336^{b}	0.365^{a}	0.03	*
	54	0.353	0.347	0.360	0.04	ns
Shell weight (g)	30	6.06	5.63	5.72	0.51	ns
	38	6.30	5.86	6.22	0.57	ns
	46	6.29	6.83	6.13	1.21	ns
	54	6.38	6.33	6.39	0.81	ns
Shell %	30	10.66	10.43	10.67	0.44	ns
	38	10.79	10.36	10.94	1.37	ns
	46	10.55	11.44	11.06	1.58	ns
	54	10.74	10.68	10.98	0.79	ns
Yolk weight (g)	30	18.57^{a}	16.72^{b}	16.15^{b}	1.53	*
	38	17.65	16.26	16.85	1.53	ns
	46	18.60^{a}	18.26^{ab}	16.78^{b}	1.73	*
	54	18.11	17.58	17.63	1.25	ns
Yolk (% of egg)	30	31.59^{a}	30.98^{ab}	30.19^{b}	1.04	*
	38	30.08^{a}	28.70^{a}	29.61^{ab}	0.92	*
	46	31.13	30.72	30.28	2.01	ns
	54	30.52	29.69	30.29	1.07	ns
Yolk colour		1^{b}	1^{b}	4.75^{a}	0.50	*
Albumen Height (mm)	30	8.04	8.80	8.83	0.92	ns
	38	7.42	7.69	7.55	0.97	ns
	46	7.07^{b}	7.17^{ab}	8.29^{a}	1.18	*
	54	6.58	6 .74	6.83	0.57	ns
Albumen weight (g)	30	32.24	31.64	31.67	2.67	ns
	38	34.42	34.52	33.93	4.04	ns
	46	34.78	34.44	32.53	3.48	ns
	54	34.87	35.29	34.17	2.20	ns
Albumen (% of egg)	30	56.00^{b}	58.59^{a}	59.14^{a}	2.31	ns
	38	58.98	60.94	59.45	2.20	ns
	46	58.33	57.84	58.66	1.25	ns
	54	58.74	59.63	58.73	0.96	ns
HU score	30	90.38	95.23	95.48	5.20	ns
	38	86.70	88.60	87.70	6.97	ns
	46	84.10	84.60	92.30	8.48	ns
	54	81.11	82.14	83.09	4.75	ns
Egg pH	54	7.44	7.41	7.40	0.34	ns
Albumen pH	54	8.89	8.90	8.94	0.17	ns
Yolk pH	54	5.92	5.95	5.96	0.11	ns

a,b,: Means in a row with different superscripts are significantly different *($p \leq 0.05$)

4.7 Yolk colour

Yolk colour differed significantly ($P<0.05$) between NM and GJM, OBAT and GJM but not significant ($P>0.05$) between NM and OBAT. The difference observed between NM and GJM and that between OBAT and GJM was due to the dietary response of the hens to the yellow carotenoid present in the GJM as reported by Ewool *et al*. (Unpublished) Birds fed NM and OBAT recorded a mean yolk score of 1.00 which corresponded with pale yellow while those fed GJM diet recorded 4.75 which corresponded with deep yellow. This finding is in agreement with the earlier findings by Ewool *et al*. (Unpublished) who observed that hens fed GJM laid eggs with deeper yellow colouration. The results also agreed with Ponsano *et al*. (2004) who reported that hens fed *Rhodcyclus gelatinosus* improved yolk pigmentation when fed at varying levels. Karadas *et al*. (2006) also indicated that xanthophylls from lucerne, marigold and tomato enhanced yolk pigmentation. Yolk colour is used as a quality determination factor but is nearly entirely dependent on the diet and is easily manipulated (Ingram *et al*. 2008).

Consumer preferences for egg yolk pigmentation vary among countries and even between regions of the same country (Golabart *et al*. 2004). Plate 4.1 shows the yolk colour of egg laid by hens fed the three dietary treatments.

NM OBAT GJM

Plate 4.1: Effect of Dietary Treatment on Egg Yolk Colouration

4.8 Haematological and Biochemical Parameters

The results of the haematological and biochemical analysis are presented in Table 4.4

Table 4.4 Biochemical and Haematological Parameters

Parameter	Dietary Treatments			LSD	Level of Sign.
	NM	OBAT	GJM		
Total Protein (g/dl)	4.80	5.00	5.06	0.51	ns
Albumin (g/dl)	2.21^{b}	2.32^{ab}	2.36^{a}	0.15	*
Globulin (g/dl)	2.60	2.68	2.71	0.45	ns
Total cholesterol (mmol/L)	2.60^{a}	2.60^{a}	2.41^{b}	0.14	*
HDL (mmol/L)[1]	0.825^{a}	0.75^{ab}	0.713^{b}	0.11	*
LDL (mmol/L)[2]	0.95	0.938	0.885	0.10	ns
Triglycerides (mmol/L)	0.70	0.70	0.663	0.28	ns
Haemoglobin count (g/dl)	9.41	8.66	9.95	1.11	ns
Haematocrit (%)	30.37^{ab}	28.28^{b}	32.33^{a}	3.33	*
MCH (pg)[3]	40.19	40.00	37.72	4.35	ns
MCHC(g/dl)[4]	30.94	300.61	30.80	0.60	ns
Mean Cell Volume (fL)	100.30	130.80	131.00	58.06	ns
RBC($\times 10^{12}$/L)[5]	2.32^{ab}	2.20^{b}	2.47^{a}	0.27	*
WBC($\times 10^{9}$/L)[6]	258.10^{ab}	248.50^{b}	264.90^{a}	16.23	*

Each value is mean of two determinations.

Means in a row with different superscripts are significantly different* ($p<0.05$).

[1]HDL- high-density lipoprotein, [2]LDL- Low-density lipoprotein,[3]MCH- Mean Cell Haemoglobin [4]MCHC- Mean cell haemoglobin concentration, [5]RBC- Red blood cell, [6]WBC- White blood cell. fL- fectolitre

4.9 Blood biochemical results

4.9.1 Total serum protein

In avian species, serum total protein consists of albumins and globulins which are commonly used in nutritional studies (Bunchasak *et al*. 2005). The dietary treatments had no significant effect ($P>0.05$) in the total serum protein levels. The values recorded in this study were 4.80, 5.00 and 5.06 g/dl for NM, OBAT and GJM diets respectively. These values fall within the range of 4 to 6 g/dl reported for laying hens

by Aengwanich *et al*. (2004) and compare favourably to the reference values of 3 to 4.9 g/dl for domestic fowl as reported by Jain (1993) cited in Aengwanich *et al*. (2004). In a study by Diarra and Usman (2008), the serum total protein in the blood of black Australorp laying hens fed varying levels of soaked sesame seed meal was reported to range from 5.27 to 5.36 g/dl. These values are slightly higher compared with the results of this study and the difference could be attributed to differences in breed as stated by Bell (1971) that serum protein is influenced by breed, age, physiological state and environment. In a comparative study by Gyenis *et al*. (2006) a range value of 2.8 to 5.0 g/dl was reported for Leghorn layers and 3.2 to 5.0 g/dl for heavy body hens.

The dietary treatments did not significantly ($p>0.05$) affect the amount of globulin in the blood serum of the laying hens. There were, however, numerical differences among the various treatments showing values of 2.60, 2.68 and 2.71 g/dl for NM, OBAT, and GJM fed hens respectively. These values were similar to what was reported by Bunchasak *et al*.(2005) who recorded values ranging between 2.20 and 2.74 g/dl and indicated that globulin level of laying hens increased with increasing dietary protein. Diarra and Usman (2008) recorded values ranging from 2.21 to 2.81 g/dl when varying levels of sesame seed meal were fed to laying hens. Their report showed that the serum globulin levels were reduced ($P<0.05$) above 25 percent level of replacement of soyabean meal by sesame seed meal but not affected by dietary protein as indicated by Bunchasak *et al*. (2005).

Serum globulin contains antibodies produced by lymphocytes in the blood, liver, spleen, bone marrow, and lymph glands which fight invading microorganisms (Bunchasak *et al*. 2005). The globulins usually elevate during the acute phase of inflammatory disease, and therefore are helpful in the diagnosis and monitoring of

many infectious diseases and other causes of chronic inflammation (Margaret, 2001). Though hens fed GJM recorded relatively higher level of globulin, there were no observable symptoms of inflammatory disease throughout the experimental period. Globulin levels therefore indicate the probability of developing infection (Margaret, 2001) as serum globulin is considered as the principal site of the circulating antibodies –immunoglobulins.

The mean values of the serum albumin showed no significant differences (P>0.05) among the dietary treatments. The values recorded are 2.21, 2.32, 2.36 g/dl for birds fed NM, OBAT and GJM diets which are slightly higher than that of Bunchasak *et al*. (2005) who reported values ranging from 1.4 to1.31 g/dl. This may be attributed to differences in breeds of the birds used. The results of this experiment however, are within the range of 1.63 to 2.58 g/dl as reported by Diarra and Usman (2008). Albumin serves as the major reservoir of protein and is important in regulating blood volume by maintaining colloidal osmostic pressure, acid-base balance, and it acts as a transport carrier for small molecules such as vitamins, mineral, hormones and fatty acids (Margaret, 2001). The high serum albumin recorded by birds fed GJM diets partly accounted for their high egg production as serum albumin is noted to increase with egg production (Hunt and Hunsaker, 1965).

4.9.2 Total Serum Cholesterol

The serum total cholesterol levels were significantly different (P<0.05) between dietary treatments NM and GJM, OBAT and GJM but not significant (P>0.05) between NM and OBAT. The values recorded are 2.60 mmol/L for hens fed NM and OBAT and 2.41 mmol/L for those fed GJM. High-density lipoprotein (HDL) showed a statistical significant difference between OBAT and GJM. HDL reduces the

incidence of artherosclerosis (hardening of the arteries) and is often referred to as "good" cholesterol while Low-density lipoprotein (LDL) on the hand appears to promote artherosclerosis (Gennet and Libby, 2011). The LDL values were not significantly different (P>0.05) among all the treatments though there were numerical differences with GJM recording the least value of 0.89 mmol/L.

The triglycerides (TGS) values were also not significantly different (P>0.05). In female avian species, fat synthesis and accumulation in the liver are mainly affected by oestrogen which is synthesized from the ovary (Akiba *et al*. 1982). High TGS content in the blood of laying hens is caused by high oestrogen synthesis in the ovary in order to support high egg production (Bunchasak *et al.* 2005). However, birds fed GJM recorded the least TGS value of 0.66 mmol/L compared to that of those fed NM and Obatanpa which recorded 0.70 mmol/L in both cases but laid more eggs than the other two dietary treatments. What accounted for the high egg production by hens fed GJM diets though recorded the least triglycerides is however difficult to explain.

4.10 Haematological Parameters

The haemoglobin (Hb) value did not show significant difference (P>0.05) among the various dietary treatments. Hb gives the red blood its characteristic red colour and is responsible for transport of oxygen, carbon dioxide and nitric acid (Margaret, 2001)
The nitric oxide helps regulate blood pressure by relaxing the walls of the blood vessels, thereby increasing blood flow (Margaret, 2001). Hb controls the expansions and contractions of blood vessels, and thus the blood pressure by regulating the amount of nitric oxide to which the vessels are exposed. The values recorded in this experiment were 9.95 g/dl, 9.41 and 8.66 g/dl for GJM, NM and OBAT. These values agree with earlier report by Diarra and Usman, (2008) that the haemoglobin level of laying hens are in the range of 8.93 to 10.38 g/dl for domestic fowl but lower than

what was reported by Duroteye *et al*. (2000) who reported 11.33 g/dl for domestic fowls. Islam *et al*. (2004) reported the haemoglobin count in commercial and local chickens ranges from 7.06 to 9.37 g/dl. The findings of this study fall within the normal reference value of 7.00 to 13.00 g/dl for laying hens as reported by Jain (1993) in Aengwanich *et al*. (2004).

There were statistical differences ($P<0.05$) in the haematocrit compositions between OBAT and GJM. The values recorded were 30.37, 28.38 and 32.33 % for NM, OBAT, and GJM in that order. These results are in agreement with what was reported by Duroteye *et al*. (2000) who reported a haematocrit composition of 32.25 to 33.3 % for domestic fowls. The haematocrit measures the blood sample that consists of red blood cell after the blood has been centrifuged and the cell compacted. The red blood cell is the oxygen carrying component of the blood.

Mean red blood cell count showed a significant difference ($P<0.05$) between OBAT and GJM. The values recorded in this study were 2.32×10^{12}, 2.12×10^{12} and 2.47×10^{12}/L for NM, OBAT and GJM respectively. These values are lower than what was reported by Epelle, (1982) for domestic fowl who reported that normal red blood cell in the domestic fowl ranges from 2.6 to 3.3×10^{12}/L. The results agree with earlier finding of Duroteye *et al*. (2000) who reported red blood cell count of 2.21 to 2.48×10^{12}/L. The result showed dietary effect on white blood cell between OBAT and GJM. The reason for this effect was not clear. The other parameters namely mean cell haemoglobin, mean cell haemoglobin concentration and mean cell volume produced no significant ($P>0.05$) difference among the dietary treatments.

4.11 Biochemical result of fresh egg

The results of the biochemical analysis of fresh eggs are presented in Table 4.5

Table4.5 Biochemical composition of fresh egg

Parameters	Dietary treatments				Level of significance
	NM	OBAT	GJM	LSD	
Total Alb. Chol (mmol/L)	10.00[a]	8.00[ab]	6.75[b]	2.98	*
Total Alb. protein(g/dl)	44.50	46.00	49.00	18.61	ns
Total yolk chol.(mmol/L)	15.50	10.00	11.00	8.09	ns
Total yolk protein(g/dl)	41.00	36.00	39.00	8.16	ns

a,b,: Means in a row with different superscripts are significantly different* ($p<0.05$)

Note : Chol:- Cholesterol, Alb:- Albumen

Total albumen cholesterol was significantly different ($P<0.05$) between NM and GJM. The values recorded in this experiment are 10.00, 8.00 and 6.75 (mmol/L) for NM, OBAT and GJM respectively. Total yolk cholesterol, total yolk protein and albumen protein did not show any significant ($P>0.05$) difference among all the dietary treatments. This result is contrary to what was reported by Vashan *et al*. (2008) who reported a range of total yolk cholesterol of 12.30 to 13.95 mmol/L. The yolk recorded higher value for cholesterol than that of the albumen among the three dietary treatments. Again, the albumen recorded higher values for its protein content compared with that of the yolk for all the dietary treatments.

4.12 Effects of Dietary Treatments on Storage Quality of Eggs

4.12.1 Eggs Stored in a Refrigerator at (4-6^{o}C)

The characteristics of the eggs studied are presented in Table 4.6.

Table 4.6 Effect of egg refrigeration on egg quality

Eggs Refrigerated for 7 Days (4-6^{o}C)					
	DIETARY TREATMENTS				Level of
ITEM	**NM**	**OBAT**	**GJM**	**LSD**	significance
Initial egg weight (g)	59.46^{a}	53.07^{b}	54.84^{b}	4.18	*
Final egg weight (g)	59.01^{a}	52.73^{b}	54.64^{b}	4.17	*
Weight loss(g)	0.45^{a}	0.337^{ab}	0.21^{b}	0.23	*
Weight loss (%)	0.75	0.64	0.38	0.38	ns
Albumen pH	8.85^{a}	8.81^{a}	8.38^{b}	0.19	*
Yolk pH	5.92	5.84	5.81	0.29	ns
Albumen ht(mm)	6.63^{b}	6.92^{ab}	6.97^{a}	0.24	*
HU score	81.59^{b}	85.41^{a}	85.10^{a}	2.57	*
Albumen wt(g)	34.70^{a}	31.03^{b}	32.28^{ab}	2.81	*
Albumen %	58.70	58.84	59.09	0.87	ns
Y:A Ratio	0.52	0.52	0.51	0.02	ns
Shell weight (g)	6.37^{b}	5.72^{a}	5.84^{a}	0.40	*
Shell (%)	10.78	10.85	10.70	0.51	ns
Yolk weight (g)	17.95^{a}	15.98^{b}	16.52^{b}	1.16	*
Yolk (%)	30.43	30.31	30.21	0.88	ns
Eggs Refrigerated for 21 Days (4-6^{o}C)					
Initial egg weight (g)	62.06	58.31	58.70	4.47	ns
Final egg weight (g)	56.66	53.77	54.48	4.40	ns
Weight loss(g)	5.40	4.47	4.21	1.65	ns
Weight loss (%)	8.65	7.70	7.14	2.70	ns
Albumen pH	9.20	9.17	9.17	0.09	ns
Yolk pH	6.19	6.17	6.18	0.15	ns
Albumen ht(mm)	4.38^{b}	4.23^{a}	4.27^{ab}	0.13	*
HU score	68.73^{b}	73.69^{a}	71.42^{ab}	4.73	ns
Albumen wt(g)	31.99	30.83	31.14	2.59	ns
Albumen %	56.48	57.32	57.15	1.64	ns
Y:A Ratio	0.58	0.55	0.56	0.04	ns
Shell weight (g)	6.16	6.04	5.95	0.77	ns
Shell (%)	10.87	11.24	10.91	0.64	ns
Yolk weight (g)	18.51^{b}	16.90^{a}	17.39^{ab}	1.45	*
Yolk (%)	32.66	31.44	31.94	1.40	ns

a,b,: Means in a row with different superscripts are significantly different* ($p<0.05$)

There was no significant difference ($P>0.05$) in egg weight in cold storage (4^{o}C-6^{o}C). There was a marginal decrease in the albumen height which led to corresponding decrease in HU after 21 days of cold storage (4^{o}C-6^{o}C). The Haugh unit score and egg

pH recorded showed that eggs stored in refrigerator for 7 and 21 days recorded values which are within the domain of that of fresh eggs.

The HU values recorded were 81.59, 85.4 and 85.10 for 7 days after storage and 78.22, 77.10 and 78.72 for 21 days for NM, OBAT and GJM diets in that order. The decline in the albumen height and subsequent decrease in HU follow the trend reported by Jones and Musgrove (2005) and Silversides and Villeneuve (1994). Silversides and Villeneuve (1994) reported that with increasing age of hens, the weight of the eggs increased in quadratic fashion while the albumen height decreased linearly resulting in the decline of the HU of eggs. According to United States Department of Agriculture (USDA)-Agricultural Marketing Service guidelines (USDA, 2000), eggs with HU scores from 72 and above could be graded AA.

Albumen pH indicated significant differences ($P<0.05$) between NM and GJM, OBAT and GJM. There was no significant difference in yolk pH among the various dietary treatments but there was a slight increase with storage. According to Hauver and Hamann (1961), the increase in pH with storage was as result of carbon dioxide diffusing out of the egg. The carbon dioxide, a product of the metabolic pathways in the chicken, forms carbonic acid and bicarbonate buffers. These no longer exist when it diffuses out thereby increasing the alkalinity.

Increase in both yolk and albumen pH with storage was also reported by Moula *et al*. (2009) and Silversides and Budgell (2004).

Yolk-albumen ratio though non-significant ($P>0.05$), also showed a marginal increase with storage. The yolk percentage also increased with storage but not significant among the three dietary treatments. Higher yolk percentage is important in nutrition because is linked to a higher dry matter content of the egg and higher content in essential fatty acids (Benabljelil and Merat, 1995) as cited by Moula *et al*. (2009).

4.12.2 Eggs Stored at Room Temperature (21-25^{o}C)

The effects of dietary treatments on egg keeping quality are presented in Table 4.7.

Table 4.7 Effect of Storage on Egg Quality

Stored at Room Temperature for 7days (21-25^{o}C)					
	DIETARY TREATMENTS				Level of
Parameter	**NM**	**OBAT**	**GJM**	**LSD**	significance
Initial egg weight (g)	58.37	58.71	57.72	1.94	ns
Final egg weight (g)	57.16	57.50	56.74	1.70	ns
Weight loss(g)	1.22	1.21	0.98	0.28	ns
Weight loss (%)	2.09	2.06	1.70	0.45	ns
Albumen pH	8.85^{a}	8.81^{a}	8.3^{b}	0.19	*
Yolk pH	6.12	6.11	6.02	0.14	ns
Albumen ht(mm)	5.46	5.61	5.72	0.27	ns
HU score	74.49	74.78	75.94	2.74	ns
Albumen wt(g)	33.36	33.60	33.23	1.40	ns
Albumen %	58.36	58.43	58.56	0.18	ns
Y:A Ratio	0.53	0.53	0.53	0.04	ns
Shell weight (g)	6.10	6.19	5.96	0.34	ns
Shell (%)	10.68	10.76	10.51	0.59	ns
Yolk weight (g)	17.70	17.72	17.55	1.04	ns
Yolk (%)	30.97	30.81	30.93	1.28	ns
Eggs Stored at Room Temperature for 21days (21-25^{o}C)					
Initial egg weight (g)	62.06	58.31	58.70	4.47	ns
Final egg weight (g)	58.28	53.77	54.82	5.29	ns
Weight loss(g)	3.77	4.54	3.87	1.83	ns
Weight loss (%)	6.08	7.81	6.59	3.37	ns
Albumen pH	9.20	9.17	9.17	0.09	ns
Yolk pH	6.19	6.17	6.18	0.15	ns
Albumen ht(mm)	4.93	5.29	5.07	0.32	ns
HU score	64.37	64.34	64.42	2.39	*
Albumen wt(g)	33.62	30.83	31.48	3.30	ns
Albumen %	57.69	57.32	57.40	1.24	ns
Y:A Ratio	0.55	0.54	0.55	0.032	ns
Shell weight (g)	6.16	6.04	5.95	0.77	ns
Shell (%)	10.57^{b}	11.24^{a}	10.84^{ab}	0.55	*
Yolk weight (g)	18.51^{a}	16.69^{b}	17.39^{ab}	1.45	*
Yolk (%)	31.75	31.44	31.76	1.20	ns

a,b,: Means in a row with different superscripts are significantly different* ($p<0.05$)

The HU score deteriorated with storage from 74.49, 74.78, and 75.94 for NM, OBAT and GJM respectively for the first 7 days in storage to 64.37, 64.34 and 64.42 when stored for 21 days. It was however observed that albumen height, HU and egg weight,

decreased with the length of storage. Similar results were also reported by Nelson (2003), Jones and Musgrove, (2005), Moula *et al*. (2009) Raji *et al*. (2009). The deterioration was due to evaporative loss and protein degradation resulting in the albumen being liquefied (Moula *et al*. 2009). Haugh units are measures of albumen thickness upon breakage of the egg following a standardized procedure. Lower HU indicates lesser freshness.

Both yolk and albumen pH also increased with storage with the albumen pH becoming more alkaline. The yolk pH recorded after 21 days of storage are 6.19, 6.17 and 6.18 for NM, OBAT, and GJM diets. Kirunda and Mckee, (2000) reported a yolk pH value of 6.12 after 2 weeks of storage at room temperature (21-25^{o}C). The albumen pH values recorded are 9.20 for NM and 9.17 for hens fed OBAT and GJM diet respectively. The values are lower compared with 9.44 reported by Kirunda and Mckee, (2000) and 9.26 by Silversides and Budgell (2004) after 10 days of storage at a temperature of (21-25^{o}C). The reason for the lesser pH value recorded in this study could be as a result of lower evaporative loss (higher conservation ability) of the eggs used in the study. This implies that the eggs used in the study could be stored for relatively longer period of time.

The pH of an egg is reported to be a good measure for the follow-up of egg freshness. The pH value rises with storage as a result of evaporation and Carbon dioxide exchange (Jones and Musgrove, 2005). Temperature and humidity are known to affect the degradation speed of eggs (the rate at which eggs are deteriorated), (Silversides and Budgell, 2004; Jones and Musgrove, 2005,and Samli *et al*. (2005).

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATION

From the study, GJM in the diet of laying hens improved egg production with acceptable internal and external egg qualities such as, Haugh unit score, egg pH and shell thickness which did not show any significant difference among the various dietary treatments. Findings of this experiment suggest that it is also more economical to use GJM as the major source of carbohydrate as it would help generate more income from the sale of the eggs.

It can therefore be concluded that Golden Jubilee maize improved upon egg production and internal qualities of the egg. On the basis of these findings, farmers may consider using Golden jubilee maize for egg production.

The eggs of birds fed GJM recorded the least cholesterol level but relatively higher protein.

The major limitation here is the lack of literature on the use of GJM to compare the results of this study with. It is important therefore that more studies should be done on layers using GJM for the purposes of comparison.

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APPENDICES

Appendix 1: ANOVA FOR LAYING PERFORMANCE

Appendix 1A: Anova for total feed intake

Source of Variation	**d.f**	**s.s**	**m.s**	**v.r**	**F.pr**
Rep. Stratum	3	144.39	48.13	0.89	
Treatment	2	55.54	27.77	0.51	0.623
Residual	6	324.82	54.14		
Total	11	524.75			

Appendix 1B: Anova for total egg weight

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	249.69	83.23	1.22	
Treatment	2	440.50	220.25	3.22	0.112
Residual	6	409.79	68.30		
Total	11	1099.98			

Appendix 1C Anova for hen -day production

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	39.71	13.24	0.96	
Treatment	2	58.85	29.43	2.14	0.198
Residual	6	13.72	82.34		
Total	11	180.90			

Appendix 1D: Anova for hen-house production

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	57.24	19.08	1.36	
Treatment	2	116.35	58.18	4.15	0.074
Residual	6	84.16	14.03		
Total	11	257.75			

Appendix 1E: Anova for egg number

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	79101.	26367.	1.36	
Treatment	2	158388.	79194.	4.10	0.075
Residual	6	115932.	19322.		
Total	11	353421.			

Appendix 1F: Anova for dozen eggs

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	549.3	183.1	1.36	
Treatment	2	1099.9	550.0	4.10	0.075
Residual	6	805.1	134.2		
Total	11	2454.3			

Appendix 2: ANOVA TABLES FOR BIOCHEMICAL PARAMETERS

Appendix 2A: Anova for serum total protein

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.18229	0.06076	0.71	
Treatment	2	0.15042	0.07521	0.88	0.463
Residual	6	0.51333	0.08556		
Total	11	0.84604			

Appendix 2B: Anova for serum albumin

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.004358	0.62	0.013073	
Treatment	2	3.48	0.048750	0.024375	0.099
Residual	6	0.042083	0.007014		
Total	11	0.103906			

Appendix 2C: Anova for serum globulin

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.25266	0.08422	1.27	
Treatment	2	0.02792	0.01396	0.816	0.21
Residual	6	0.39875	0.06646		
Total	11	0.67932			

Appendix 2D: Anova for serum total cholesterol

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.25827	0.08609	2.25	
Treatment	2	0.09127	0.04563	1.19	0.366
Residual	6	0.22913	0.03819		
Total	11	0.57867			

Appendix 2E: Anova for high density lipoprotein

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.097292	0.032431	8.81	
Treatment	2	0.026250	0.013125	0.095	3.57
Residual	6	0.022083	0.003681		
Total	11	0.145625			

Appendix 2F: Anova for low density lipoprotein

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.002103	0.000701	0.22	
Treatment	2	0.009674	0.004837	1.54	0.289
Residual	6	0.018855	0.003143		
Total	11	0.030632			

Appendix 2G: Anova for serum triglycerides

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.02021	0.00674	0.26	
Treatment	2	0.00375	0.00187	0.07	0.930
Residual	6	0.15292	0.02549		
Total	11	0.17688			

Appendix 3: ANOVA FOR HAEMATOLOGICAL PARAMETERS

Appendix 3A: Anova for haemoglobin

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	1.0593	0.3531	0.86	
Treatment	2	3.3454	1.6727	4.08	0.076
Residual	6	2.4628	0.4105		
Total	11	6.8675			

Appendix 3B: Anova for haematocrit

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	14.644	4.881	1.32	
Treatment	2	31.206	15.603	4.21	0.072
Residual	6	22.215	3.702		
Total	11	68.065			

Appendix 3C: Anova for mean cell haemoglobin

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	15.074	5.025	0.79	
Treatment	2	15.096	7.548	1.19	0.366
Residual	6	37.949	6.325		
Total	11	68.120			

Appendix 3D: Anova for mean cell haemoglobin concentration

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.6898	0.2299	1.93	
Treatment	2	0.2216	0.1108	0.93	0.444
Residual	6	0.7136	0.1189		
Total	11	1.6249			

Appendix 3E: Anova for mean cell volume

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	3367.	1122.	1.00	
Treatment	2	2490.	1245.	1.11	0.390
Residual	6	6755.	1126.		
Total	11	12612.			

Appendix 3F:Anova for red blood cell

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.06909	0.02303	0.98	
Treatment	2	0.15495	0.07748	0.108	3.30
Residual	6	0.14078	0.02346		
Total	11	0.36483			

Appendix 3G: Anova for white blood cell

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	400.93	133.64	1.52	
Treatment	2	541.32	270.66	3.08	0.120
Residual	6	528.05	88.01		
Total	11	1470.31			

Appendix 4: ANOVA FOR EGG QUALITY CHARACTERISTICS

Appendix 4A: Anova for egg weight

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	1.161	0.387	0.07	
Treatment	2	3.190	1.595	0.30	0.749
Residual	6	31.546	5.258		
Total	11	35.897			

Appendix 4B: Anova for albumen height

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.1233	0.0411	0.38	
Treatment	2	0.1260	0.0630	0.58	0.590
Residual	6	0.6552	0.1092		
Total	11	0.9045			

Appendix 4C:Anova for albumen weight

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	2.243	0.748	0.46	
Treatment	2	2.573	1.287	0.80	0.493
Residual	6	9.689	1.615		
Total	11	14.505			

Appendix 4D: Anova for albumen percentage

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	2.6668	0.8889	2.92	
Treatment	2	2.1285	1.0643	3.49	0.099
Residual	6	1.8296	0.3049		
Total	11	6.6249			

Appendix 4E: Anova for haugh unit score

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	6.008	2.003	0.27	
Treatment	2	7.780	3.890	0.52	0.621
Residual	6	45.147	7.525		
Total	11	58.935			

Appendix 4F:Anova for egg shell thickness

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.0003377	0.0001126	0.19	
Treatment	2	0.0005195	0.0002597	0.43	0.669
Residual	6	0.0036178	0.0006030		
Total	11	0.0044750			

Appendix 4G:Anova for egg shell weight

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.1416	0.0472	0.21	
Treatment	2	0.0100	0.0050	0.02	0.977
Residual	6	1.3178	0.2196		
Total	11	1.4695			

Appendix 4H: Anova for egg shell percentage

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.5166	0.1722	0.84	
Treatment	2	0.2038	0.1019	0.50	0.632
Residual	6	1.2346	0.2058		
Total	11	1.9550			

Appendix 4I: Anova for yolk weight

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.1321	0.0440	0.08	
Treatment	2	0.6783	0.3392	0.65	0.556
Residual	6	3.1438	0.5240		
Total	11	3.9542			

Appendix 4J: Anova for egg yolk percentage

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	1.0080	0.3360	0.88	
Treatment	2	1.4553	0.7276	1.90	0.230
Residual	6	2.3011	0.3835		
Total	11	4.7644			

Appendix 4K:Anova for egg yolk colour

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.25000	0.08333	1.00	
Treatment	2	37.50000	18.75000	225.00	<.001
Residual	6	0.50000	0.08333		
Total	11	38.25000			

Appendix 4L: Anova for yolk-albumen ratio

Source of Variation	d.f	s.s	m.s	v.r	F.pr
Rep. Stratum	3	0.0009536	0.0003179	1.55	
Treatment	2	0.0010720	0.0005360	2.62	0.152
Residual	6	0.0012289	0.0002048		
Total	11	0.0032544			

Appendix 5: Mean weekly total feed intake per treatment (kg)

Week	NM	OBAT	GJM	LSD
1	12.48	12.52	12.47	0.07
2	13.10	12.94	12.98	0.20
3	12.85	12.87	12.84	0.17
4	12.82	12.90	12.98	0.17
5	12.93	12.94	13.04	0.07
6	13.64	13.49	13.52	0.45
7	13.58	13.56	13.62	0.23
8	13.29	13.22	13.44	0.83
9	13.32	13.15	13.70	0.88
10	13.35	13.05	13.51	0.83
11	13.02	13.05	13.54	0.68
12	12.93	12.75	12.83	0.78
13	12.97	12.74	12.88	0.93
14	13.00	12.76	12.66	0.79
15	12.76	12.79	12.64	0.97
16	12.88	12.73	12.60	0.67
17	12.95	12.85	12.56	0.63
18	12.93	12.79	12.65	0.63
19	12.95	12.63	12.65	0.69
20	12.75	12.38	12.69	0.64
21	12.65	12.53	12.68	0.66
22	12.68	12.48	12.70	0.67
23	12.71	12.56	12.67	0.72
24	12.68	12.54	12.70	0.75
25	12.71	12.48	12.69	0.71
26	12.70	12.47	12.71	0.70
27	12.69	12.51	12.68	0.74
28	12.67	12.49	12.70	0.76
29	12.67	12.46	12.64	0.69
30	12.66	12.47	12.64	0.73
31	12.66	12.47	12.65	0.74
32	12.66	12.52	12.66	0.72
33	12.46	12.47	12.66	0.87

Appendix 6: Weekly Cost of feed consumed per treatment (GH¢)

Week	NM	OBAT	GJM	LSD
1	6.50	6.52	6.50	0.13
2	6.82	6.74	6.76	0.10
3	6.70	6.71	6.69	0.91
4	6.68	6.72	6.76	0.61
5	6.73	6.74	6.79	0.04
6	7.10	7.03	7.04	0.23
7	7.07	6.96	7.09	0.12
8	6.98	6.89	7.00	0.43
9	6.94	6.85	7.14	0.46
10	6.95	6.80	7.04	0.43
11	6.78	6.80	7.05	0.35
12	6.74	6.64	6.69	0.41
13	6.76	6.64	6.71	0.49
14	6.78	6.65	6.60	0.41
15	6.65	6.67	6.59	0.51
16	6.71	6.63	6.57	0.35
17	6.75	6.70	6.54	0.33
18	6.74	6.67	6.59	0.36
19	6.75	6.58	6.59	0.36
20	6.64	6.45	6.61	0.33
21	6.60	6.30	6.61	0.35
22	6.61	6.50	6.62	0.35
23	6.63	6.55	6.60	0.38
24	6.61	6.54	6.62	0.39
25	6.62	6.50	6.61	0.37
26	6.62	6.50	6.62	0.37
27	6.61	6.52	6.61	0.39
28	6.60	6.51	6.62	0.39
29	6.60	6.49	6.59	0.36
30	6.60	6.50	6.58	0.38
31	6.60	6.50	6.59	0.38
32	6.60	6.52	6.60	0.37
33	6.49	6.50	6.60	0.45

Appendix 7: Mean weekly total eggs laid

Week	NM	OBAT	GJM	LSD
1	49.00	52.01	68.6	0.54
2	63.04	55.00	70.03	19.62
3	65.82	55.00	76.9	3.04
4	65.70	57.01	74.11	19.68
5	76.12	66.10	76.21	20.10
6	73.00	71.03	77.80	13.93
7	77.10	71.91	77.18	11.50
8	77.00	58.01	66.12	8.06
9	72.11	74.00	73.14	9.55
10	45.00	72.82	75.01	15.10
11	28.31	67.00	76.12	11.15
12	63.11	68.01	82.00	12.88
13	76.13	67.11	79.65	11.17
14	72.01	67.81	80.04	7.22
15	71.89	69.21	79.89	7.76
16	70.21	69.11	77.00	14.31
17	79.00	78.00	81.01	8.48
18	79.11	80.12	87.01	7.56
19	77.00	69.50	81.75	5.45
20	78.00	70.80	79.00	9.47
21	75.25	70.00	81.75	5.54
22	81.20	70.50	84.20	8.49
23	72.20	76.20	77.20	10.76
24	76.00	77.00	92.50	15.29
25	75.80	73.50	76.50	16.56
26	71.80	74.20	72.80	7.18
27	79.00	76.20	75.20	12.62
28	79.80	78.00	78.20	13.31
29	74.20	69.00	72.00	11.43
30	72.00	72.80	78.80	9.04
31	79.20	79.20	81.20	8.73
32	85.75	83.00	89.75	5.12
33	86.80	80.80	87.20	7.06

Appendix 8: Effect of dietary treatments on dozen eggs laid

Week	NM	OBAT	GJM	LSD
1	4.10	4.31	5.73	0.99
2	5.25	4.54	5.85	1.65
3	5.46	4.56	6.40	1.94
4	5.37	4.73	6.15	1.64
5	6.35	5.46	6.31	1.68
6	6.06	5.88	6.46	1.16
7	6.40	5.96	6.38	0.96
8	6.38	4.81	5.50	0.67
9	5.90	6.17	6.10	0.80
10	3.75	6.06	6.21	1.26
11	2.33	5.56	6.31	0.93
12	5.21	5.62	6.82	1.07
13	6.38	5.58	6.67	0.93
14	6.00	5.75	6.65	0.60
15	5.96	5.71	6.67	0.65
16	5.83	5.75	6.38	1.19
17	6.60	6.52	6.77	0.71
18	6.58	6.69	7.27	0.63
19	6.42	5.79	6.81	0.45
20	6.50	5.90	6.58	0.79
21	6.27	5.8	6.81	0.46
22	6.77	5.88	7.02	0.71
23	6.02	6.35	6.44	0.90
24	6.33	6.42	7.71	1.27
25	6.31	6.12	6.38	1.38
26	5.98	6.19	6.06	0.60
27	6.58	6.35	6.27	1.05
28	6.65	6.50	6.52	1.11
29	6.19	5.75	6.00	0.95
30	6.00	6.06	6.56	0.75
31	6.60	6.60	6.77	0.73
32	7.15	6.92	7.48	0.43
33	7.23	6.73	7.27	0.59

Appendix 9: Weekly Feed consumed per dozen eggs produced (g)

Week	NM	OBAT	GJM	LSD
1	3082	2938	2181	676.50
2	2624	2928	2231	1030.50
3	2456	2919	2011	1074.00
4	2467	2787	2123	867.90
5	2069	2414	2082	653.80
6	2261	2320	2103	456.10
7	2128	2264	2144	333.00
8	2112	2782	2454	343.30
9	2254	2157	2253	304.60
10	3773	2154	2185	1142.80
11	6225	2355	2147	1948.30
12	2552	2278	1879	481.80
13	2083	2292	1933	392.40
14	2178	2221	1908	289.40
15	2165	2251	1897	290.30
16	2239	2266	1983	487.30
17	1964	1979	1862	226.10
18	1973	1921	1743	201.30
19	2024	2186	1859	144.30
20	1969	2110	1931	260.60
21	2018	2150	1866	100.70
22	1881	2139	1814	206.40
23	2122	1995	1969	296.70
24	2012	1957	1673	320.40
25	2039	2053	2013	443.20
26	2132	2025	2102	204.10
27	1952	1974	2029	385.20
28	1926	1930	1963	373.70
29	2070	2174	2118	343.50
30	2129	2060	1926	260.90
31	1929	1894	1876	238.40
32	1774	1811	1694	109.20
33	1726	1855	1742	132.10

Appendix 10: Weekly Cost of feed consumed per dozen egg produced (GH¢)

Week	NM	OBAT	GJM	LSD
1	1.61	1.53	1.14	0.35
2	1.37	1.53	1.16	0.54
3	1.28	1.52	1.05	0.56
4	1.29	1.45	1.06	0.45
5	1.10	1.26	1.09	0.34
6	1.18	1.21	1.10	0.24
7	1.10	1.18	1.12	0.17
8	1.10	1.45	1.28	0.18
9	1.18	1.12	1.17	0.16
10	1.97	1.22	1.14	0.60
11	3.24	1.23	1.12	1.02
12	1.33	1.19	0.98	0.25
13	1.09	1.19	1.01	0.21
14	1.14	1.16	0.99	0.15
15	1.13	1.17	0.99	0.15
16	1.17	1.18	1.03	0.25
17	1.02	1.03	0.97	0.12
18	1.03	1.00	0.91	0.11
19	1.05	1.14	0.97	0.08
20	1.03	1.10	1.00	0.14
21	1.05	1.12	0.97	0.05
22	0.98	1.11	0.95	0.11
23	1.11	1.04	1.03	0.16
24	1.05	1.02	0.87	0.17
25	1.06	1.07	1.05	0.23
26	1.11	1.06	1.10	0.10
27	1.02	1.03	1.06	0.20
28	1.00	1.01	1.02	0.20
29	1.08	1.13	1.10	0.17
30	1.11	1.07	1.00	0.44
31	1.01	0.99	0.98	0.12
32	0.92	0.94	0.88	0.06
33	0.90	0.97	0.91	0.07

Appendix 11: Weekly Feed consumed per kg egg produced

Week	NM	OBAT	GJM	LSD
1	4.91	4.77	3.50	1.04
2	4.01	4.67	3.54	1.61
3	3.68	4.56	3.16	1.55
4	3.69	4.26	3.28	1.18
5	3.04	3.72	3.01	0.07
6	3.28	3.55	3.16	0.64
7	3.20	3.40	3.19	0.47
8	3.10	4.07	3.61	0.62
9	3.28	3.23	3.30	0.39
10	5.55	3.17	3.18	1.60
11	9.12	3.49	3.17	2.86
12	3.72	3.38	2.78	0.79
13	3.06	3.38	2.25	0.62
14	3.19	3.26	2.81	0.45
15	3.10	3.13	2.80	0.48
16	3.18	3.31	2.98	0.59
17	2.82	2.87	2.73	0.31
18	2.84	2.78	2.57	0.31
19	2.90	3.18	2.71	0.24
20	2.80	3.06	2.82	0.40
21	2.88	2.99	2.67	0.30
22	2.79	2.97	2.69	0.38
23	3.02	2.91	2.82	0.36
24	2.88	2.79	2.49	0.46
25	3.08	2.95	2.89	0.74
26	3.04	2.89	3.02	0.39
27	2.76	2.79	2.96	0.63
28	2.75	2.70	2.82	0.55
29	3.06	3.06	3.03	0.65
30	3.05	2.89	2.73	0.44
31	2.74	2.62	2.65	0.37
32	2.49	2.51	2.42	0.17
33	2.42	2.57	2.47	0.20

Appendix 12: Weekly Cost of feed consumed per unit kg of eggs produced by laying hens (GH¢)

Week	NM	OBAT	GJM	LSD
1	2.56	2.49	1.82	0.54
2	2.09	2.43	1.84	0.84
3	1.92	2.37	1.65	0.81
4	1.92	2.22	1.71	0.61
5	1.58	1.94	1.57	0.44
6	1.71	1.85	1.65	0.34
7	1.67	1.77	1.66	0.24
8	1.61	2.12	1.88	0.33
9	1.71	1.68	1.72	0.21
10	2.89	1.65	1.66	0.83
11	4.78	1.82	1.65	1.49
12	1.94	1.76	1.45	0.16
13	1.59	1.76	1.48	0.32
14	1.66	1.69	1.46	0.23
15	1.66	1.73	1.46	0.25
16	1.66	1.73	1.55	0.31
17	1.47	1.49	1.42	0.16
18	1.48	1.45	1.34	0.16
19	1.50	1.66	1.41	0.13
20	1.46	1.59	1.47	0.21
21	1.50	1.56	1.39	0.16
22	1.45	1.55	1.40	0.20
23	1.58	1.52	1.47	0.19
24	1.50	1.45	1.30	0.24
25	1.61	1.54	1.51	0.39
26	1.59	1.50	1.57	0.22
27	1.44	1.45	1.54	0.33
28	1.43	1.41	1.47	0.29
29	1.60	1.60	1.58	0.34
30	1.59	1.50	1.42	0.23
31	1.43	1.37	1.38	0.19
32	1.30	1.30	1.26	0.08
33	1.26	1.34	1.29	0.11

Appendix 13: Mean weekly egg weight (g)

Week	NM	OBAT	GJM	LSD
1	52.36	51.29	51.98	2.57
2	54.35	52.39	52.63	3.10
3	55.44	53.38	52.97	3.04
4	55.56	54.40	53.99	2.83
5	56.7	54.2	57.7	6.79
6	55.95	54.53	55.50	3.26
7	55.85	55.55	55.99	1.54
8	56.01	57.03	56.84	5.79
9	57.31	55.73	56.97	2.32
10	56.65	56.58	57.18	2.77
11	56.61	56.27	56.56	1.92
12	57.35	56.15	56.24	1.94
13	56.93	56.50	56.59	1.46
14	56.96	56.78	56.59	1.30
15	56.83	56.60	56.44	1.48
16	58.48	56.96	55.67	0.35
17	58.08	57.48	56.77	1.27
18	57.93	57.64	56.52	1.74
19	58.29	57.32	57.30	1.23
20	58.57	57.60	57.01	1.02
21	58.40	60.10	58.20	6.38
22	56.40	60.58	56.32	5.56
23	58.60	57.10	58.2	6.13
24	58.30	58.60	55.9	6.44
25	55.46	58.13	57.98	4.15
26	58.48	58.51	58.10	2.24
27	58.90	59.02	57.22	2.12
28	58.49	59.57	58.00	1.06
29	56.63	59.18	58.32	3.21
30	58.30	59.49	58.76	1.71
31	58.79	60.17	59.00	1.45
32	59.49	60.09	58.25	1.68
33	59.42	60.03	58.74	2.07