

EFFECT OF FEED ENZYMES AND ENERGY LEVEL ON BROILER CHICKEN

(*Gallus domesticus*) PERFORMANCE IN KENYA

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A THESIS SUBMITTED IN PARTIAL FULFILLMENT FOR THE DEGREE OF

MASTER OF SCIENCE IN ANIMAL NUTRITION AND FEED SCIENCE

IN THE FACULTY OF VETERINARY MEDICINE

DEPARTMENT OF ANIMAL PRODUCTION

UNIVERSITY OF NAIROBI

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DECLARATION

Filling scientific gaps of scientific investigations, is a full responsibility of all researchers, I therefore do declare that, the findings in this thesis are my own and have never been published for any degree in any other university



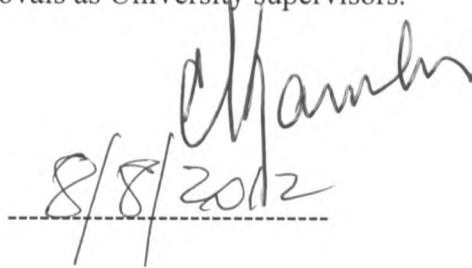
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DEDICATION

I dedicate this work to my loving wife Mary Awur Deng Anyuat, my dear daughter Sarah De-Pach, my dear sons Stephen De-Pach, Emmanuel De-Pach, Ajak and Anyuat for their understandings while away from all, for more than two years, may GOD bless you all.

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LIST OF ABBREVIATIONS

ADF: Acid Detergent Fibre

AKEFEMA: Association of Kenya Feed Manufacturers

AME: Apparent Metabolizable Energy

AOAC: Association of Analytical Chemists

ANOVA: Analysis of Variance

AXsol: Arabinoxylans soluble

AX_{insol}: Arabinoxylans insoluble

⁰C: Degree Centigrade

CF: Crude Fibre

Crude Protein

D1: Diet one high energy without enzyme addition

D2: Diet two medium energy without enzyme addition

D3: Diet three low energy without enzyme Addition

D4: Diet four high energy with Nutrase Xylla addition

D5: Diet five medium energy with Nutrase Xylla addition

D6: Diet six low energy with Nutrase Xylla addition

D7: Diet seven high energy with Allzyme Solid-state Fermentation

D8: Diet eight medium energy with Allzyme Solid State Fermentation

D9: Diet nine low energy with Allzyme Solid State Fermentation

DDGs: Dried Distilled Grain Soluble

DCP: Dicalcium Phosphate

df: Degree of Freedom

DM: Dried Matter

EE: Ether Extract

Eng. Engineer

FCR: Feed Conversion Ratio

F. Pr: Fr Probability

g: Gram

Ge: Gross Energy

GoK: Government of Kenya

H_A: Alternative Hypothesis

H₀: Null Hypothesis

HE: High Energy

IB: Infectious Bronchitis

IBD: Infectious Bursal Disease

IU: International unit

Liet. Gen.: Lieutenant General

KEBS: Kenya Bureau of Standard Specification

Kenchic : Kenya Chicken Company Limited

KShs : Kenya shilling

Kg: Kilogram

KW: Kilowatts lwt: Live Weight

ME: Medium Energy Me: Metabolizable Energy

MT: Metric ton

MOA (China): Ministry of Agriculture, China

Mg: Milligram

ms: mean of square

NDF: Nitrogen Detergent Fibre

NRC: National Research Council

NSPs: Non-Starch Polysaccharides

NSPDE: Non-starch polysaccharide degrading enzymes.

NX: Nutrase Xylla

SBM: Soya Bean Meal

Se: Standard Error of mean

SED: Standard Error of Difference

SSF: Solid state Fermentatiuon

SS: Sum of Square

T: Trace

61. Vr. Varian

ACKNOWLEDGEMENTS

I wish to express my most utmost appreciation and deep gratitude to Prof. P.N. Mbugua who offered me untiring assistance and encouragement throughout this study and preparation of the thesis. I wish also to give many thanks to Prof. C.K. Gachuri, for his guidance, suggestions and advice. Thanks are due to Prof. L.W. Kabuage for her unlimited assistance since I began this work. I gratefully acknowledge the assistance given by John. M. Kanyi during data collection, A. Kimende, Ambale and J. Kinuthia for their assistance in the laboratory work. I also wish to thank Nutrex of Belgium for assisting in analysis of soluble and insoluble NSPs of the diets fed during the feeding experiment. I would like to thanks Elizabeth W. Wainaina for her advice and assistance in data analysis, may God bless you.

I sincerely give many thanks back to Sudan People Liberation Army (SPLA) Liet. Gen. Dr. Majak A. Atem, SPLA Liet. Gen. Eng. Kuol Manyang Juuk, current Governor of Jonglei state, and the undersecretary Ministry of Animal Resources and Fisheries-Juba, Dr. Waragak G. Faguir, who all made funding to my studies possible. I also acknowledge the teaching staffs of Nairobi University (both in Kabete and Chiromo campuses) for their great effort in preparing me during my course work. May God bless you all.

Last but not the least, I would like to give all thanks to The Almighty Father for His love, care, life, strength and the blessings he has provided for me up to date. He has made it possible for me to say YES I can.

ABSTRACT

The effects of two feed enzymes, Allzyme SSF and Nutrase Xylla (NX) and dietary energy level on broiler chicken performance were studied. Three main diets, high energy (HE), medium energy (ME) and low energy (LE) were formulated to provide 100, 95 and 90%, respectively of metabolizable energy requirements of broiler chicken, based on Kenya Bureau of standards. The high energy diet contained 3000 kcal/kg metabolizable energy, while the ME and LE diets contained 2850 and 2700 kcal/kg metabolizable energy, respectively. From the three main diets, nine experimental diets were made by adding or omitting the feed enzymes. Diets 1, 2 and 3 were HE, ME and LE, respectively, without enzyme addition. Diets 4, 5 and 6 were HE, ME, and LE, all with the enzyme Nutrase Xylla added, while diets 7, 8 and 9 were HE, ME and LE, respectively, to which the enzyme Allzyme SSF was added. Both enzymes were included in the feed at the rate of 0.1gm/ kg of diet. A total of 360 day-old chicks of Arbor acres (unsexed) were randomly divided into the nine dietary treatments each comprising 40 chicks, with four replicates of ten birds per replicate. The design was 3×3 Factorial Design. The birds were reared, in deep litter pens each measuring 1m², from 0-49 days post-hatch. Feed and water were provided *ad libitum*. Data on feed consumption, weight gain, feed conversion ratio and cost of feed was collected. The data was analysed using Genstat Discovery Edition 3. The results showed that from 0-21 days, 22-49 days and for entire feeding phase (0-49 days), feed intake was significantly ($P<0.05$) improved with birds fed non-supplemented diets compared to supplemented diets with both Nutrase Xylla and Allzyme SSF, except birds fed low energy Allzyme added diets which tended to have significant ($P<0.05$) feed intake. Feed conversion ratio during starter, finisher and for entire feeding phase, was significantly ($P<0.05$) improved at low energy of non-supplemented diets, while it was significantly improved in birds fed Nutrase

Xylla throughout of growing period. The addition of exogenous enzymes was effective in overcoming anti-nutritive effects of NSPs on broiler performance only at low energy levels. Body weight gain during starter, finisher and for entire feeding phases, was not affected by enzymes inclusion as body weight gains of non-supplemented diets were higher compared to diets supplemented with exogenous enzymes at high and medium energy levels. The energy level of the diet affected the performance of the birds fed low Allzyme SSF, except FCR where low energy diets added to enzyme did not have any effect. The highest return was obtained from birds fed diets supplemented with Allzyme SSF, while Nutrase Xylla added diets resulted to loss of money.

Keywords:- Allzyme ® SSF, Nutrase Xylla, broiler chickens performance, energy levels, non-starch polysaccharides (NSPs), economic returns.

CHAPTER ONE: INTRODUCTION

1. General introduction

Cereal grains and their milling by-products are important sources of energy in poultry nutrition, while oil seed cakes are used as source of protein, Wachira, (2006). The main cereal grains utilized are maize, sorghum, wheat, barley, oats and broken rice. However, these feedstuffs contain complex carbohydrates termed as Non-Starch Polysaccharides (NSPs), which possess anti-nutritive activities even at low levels in poultry diets. Cereal milling by-products are particularly high in NSPs content associated with their high fibre levels. The by-products are nevertheless popular ingredients for poultry feeds in developing countries due to their low cost in comparison with whole grains. In Sub-Saharan Africa, the tendency is to use feedstuffs high in fibre or NSPs because of low availability and high cost of maize grain and soya bean meal, Sonaya, (2008).

1.1 Problem Statement

Non-starch polysaccharides (NSPs) affect poultry performance by lowering nutritive value of feedstuffs in poultry diets. High levels of NSPs can result in increased viscosity in the small intestine of chickens, and depress nutrients utilization and performance, Choct and Annison, (1992). The effect of NSPs on poultry performance has been widely studied in barley, rye and wheat, Choct and Annison, (1992b). Results from studies by these researchers have shown that NSPs cause depression in growth, sticky droppings and reduction in available metabolizable energy (AME) to the bird. Poultry naturally produce a wide range of enzymes to aid in digestion, but the range is not comprehensive and most of the NSPs are not digested. While animal protein sources (such as fishmeal and blood meals) used for poultry feeding can easily be digested by

birds, plant materials and their by products have certain inherent residues that are not digested by birds due to absence of a spectrum of appropriate enzymes, Ferket and Middleton, (1999).

1.2 Justification

This problem can be overcome through supplementing feeds high in NSPs with exogenous enzymes. Improvement of poultry diets through use of exogenous enzymes is well documented and has been comprehensively reviewed by several researchers such as Chessen, (1987); Evans, (1995); Campbell and Classen, (1990). From their reviews, it was reported that addition of exogenous enzymes to fibrous feeds for monogastric animals (poultry, fish, nursing calves and pigs) reduced viscosity of the ingesta in the intestine and showed a marked improvement on animal performance indices (feed intake, body weight gain and feed conversion ratio). Bedford (1996) reported that exogenous enzyme supplementation resulted in an overall improvement in nutrients digestion and reduction in endogenous amino acids losses. Also, Marsman *et al.* (1997) reported that an improvement in the nutritional value of soya bean meal could be achieved through supplementation with protease enzymes which hydrolyse undigestible proteins and render them useful to the animal.

The use of feed enzymes in poultry diets in Kenya is not widespread due to lack of data generated under local conditions. However, a number of feed enzymes are commercially available in Kenya. Allzyme ® SSF, produced through a unique solid-state fermentation (SSF) technique by non-genetically modified fungus known as *Aspergillus niger*, is one of such enzymes. It is a cocktail of seven enzymes namely, phytase, protease, pentosanase, β -glucanase, cellulase, amylase and pectinase. As their names indicate, these enzymes are capable of breaking down protein, cellulose, pentosans, phytate and starch, therefore improving digestibility and absorption of nutrients in the avian intestinal tract. Such a cocktail of enzymes may provide a

competitive strategy to improve nutrients utilization in poultry diets. Another commercially available feed enzyme is Nutrase Xylla (NX), which contains endo-1, 4- β -xylanase. It is designed for rations high in arabinoxylans and it is normally used in rations, containing high levels of wheat and its by-products, Cowieson *et al.*; (2005).

The purpose of this study was therefore; to investigate the effect of adding exogenous enzymes on the performance of broiler chickens fed on diets compounded using locally available feedstuffs.

1.3 Objectives of the study

1.3.1 Main objective

The main objective of the study was: To assess the effects of inclusion of feed enzymes on the performance of broiler chickens.

1.3.2 Specific objectives

- (i) To evaluate the effects of inclusion of feed enzymes on growth performance in broiler chickens
- (ii) To determine the effects of inclusion of feed enzymes on energy and feed utilization in broiler chickens
- (iii) To assess the cost effectiveness on use of feed enzymes in broiler rations.

1.3.3 Hypothesis to be tested

H_0 : Adding exogenous feed enzymes to broiler chicken feeds does not affect performance of the birds in relation to weight gain, feed intake, and feed conversion ratio (FCR).

H_A : Adding exogenous feed enzymes to broiler chicken feeds affects performance of the birds in relation to weight gain, feed intake and feed conversion ratio (FCR).

CHAPTER TWO: LITERATURE REVIEW

2. Overview of Poultry industry in Kenya

Poultry production is undertaken in Kenya in a multiple of ways, utilizing different sets of resources and in a wide spectrum of social, cultural and economic conditions. The types of poultry raised in Kenya include chickens, ducks, geese, turkeys, guinea fowls, pigeons, and ostriches, with chickens as predominant species. The poultry population in Kenya is estimated at 31.8 million birds, comprising 25.8 million indigenous chickens and 6 million commercial birds, MoLD, (2009). Poultry production is the most popular livestock enterprise in rural households of Kenya where over 70% (27 million) of the country's population live and derive their livelihood. Commercial poultry primarily consists of exotic hybrid chickens that include broilers, layers and the breeding stock. Hybrid layers are mostly kept near potential markets for eggs (such as cities and towns) where space is limited but ready-made commercial feeds are easily available, Wachira, (2008). Indigenous chicken are kept by 90% of rural households while broilers and layers are reared in urban and peri-urban areas, Wachira, (2008). The Kenyan poultry farmer is faced with various challenges particularly the high cost of commercial feed which are often low quality.

2.1 Global perspective and recent advances in poultry feed production

Domestic animals continue to make important contributions to global food supply and, as a result, animal feeds have become an increasingly critical component of the integrated food chain. Livestock products account for about 30 percent of the global value of agriculture and 19 percent of the value of the food production, and provide 43 percent of the protein and 16 percent of the energy consumed in the human diets, Garnsworthy and Wiseman, (2008). Meeting consumer demand for more meat, milk, eggs, and other livestock products is dependent to a large extent on

the availability of regular supplies of appropriate, cost-effective and safe animal feeds. Therefore, it is appropriate to begin defining who and what the animal feed industry is.

While there are various ways to produce feed for livestock, there are three broad production and delivery systems involved, where farmers “take in” feed to assist them in producing their livestock products, they are:

- (i) Commercial operations producing feed for sale
- (ii) Integrated operations where large pig or poultry producers in particular produce their own feedstuffs and
- (iii) Cooperative operations where farmers jointly own the feed mill or production plant that produces the feed they use.

The animal feed industry seems to be similar or different between the countries or across the borders, because the same livestock are fed (often rearing the same genetic stock); the same research trials are used to determine what animals need in order to maximize their genetic potential, the same feed formulation software is used to provide “least-cost diets in order to remain competitive in the market place. And finally the same manufacturing technology is used, the same equipment and most often the same raw materials.

Despite the strong trends of vertical integration and consumption within the industry, the world’s 10 largest feed manufacturers produce less than 65 million tons of feed per year – less than 11 percent of the global feed output, Garnsworthy, P.C; and Wiseman, J. (2006). The global feed industry therefore still remains broadly based, with many local and regional commercial feed companies as well as specialized firms. However, as the feed industry changes from being a “processor” and “transformer” of agricultural commodities and other raw materials into basic livestock diets to a sophisticated nutritional delivery system that is very much part of the food

chain, it is cognizant on governments to take greater interest in the work that the industry does and to sanction the raw material that it has to use.

In the modern feeding practices, feed additives are assuming a position of prime importance in poultry nutrition. Feed additives not only stimulate growth and feed efficiency but the health of birds can also be improved Panda, *et al.*, (2001). Probiotics, enzymes, amino acids supplementations and readily available minerals, are all relatively new additions to the armory of the poultry nutritionists and have a very positive effect on nutrient utilization when used with appropriate feed ingredients.

Probiotics are defined as live microbial supplements, which beneficially affect the host animal by improving its intestinal microbial balance, Panda *et al.*, (2009). These active microorganisms have a positive effect on the host organism by modifying its GI biota. The use of probiotics, in poultry feeds is gaining popularity in the recent years and large number of such preparations has been launched by different commercial organizations. In contrast, enzymes are biological catalysts, which bring about biochemical reactions without themselves undergoing change. Enzymes are not living organisms but they are products of living organisms such as bacteria, fungi, yeast and plant tissues. Commercial enzymes used as feed supplements do not contain a single enzyme but rather they are preparation of more than one enzyme.

Commercial amino acids of particular interest to poultry nutrition are L-lysine, DL-methionine, L-threonine and L-tryptophan. The use of commercial amino acids in diet formulation of poultry allows formulations to approximate more closely to an ideal amino acid balance, thereby reducing dietary nitrogen concentrations with corresponding reductions in amino acid catabolism and nitrogen pollution. The use of commercial amino acids also allows flexibility in feed formulation and more consistent prediction of amino acid availability from the diet and thus provides a consistent performance in a similar way to that obtained with feed enzymes.

2.2 The livestock feed industry in Kenya

The Government of Kenya (GoK), in its recovery strategy for wealth and employment creation, has identified agriculture as an important vehicle in realization of its objectives, which include bringing together cooperation and coordination of all business involved in the manufacturing of animal feeds and to promote, support and encourage commercial livestock production, Maina, (2011). The feed milling industry has experienced unprecedented difficulties in securing raw materials for mixed feed production. This is due to a shortage of grain (including maize), grain milling by-products and oil cakes. On average, the prices of these materials have gone up by 50-94% in the last two years, with resultant increase in the cost of animal feeds to a point where most farmers are not able to sustain their livestock. The Association of Kenya Feed Manufacturer (AKEFEMA) has been concerned with the recent shortage of raw materials used for animal feed which have resulted in sky-high prices of feeds. The current shortage of raw materials threatens livestock production and hence farmers livelihood and the people who depend directly and indirectly on the industry.

Feed manufacturers in Kenya require about 622,000 metric tons (MT) of raw materials per year to compound feeds, Maina, C, (2011). The current annual maize demand alone in the livestock industry stands at 180,000 MT. Global and local unavailability of raw materials has resulted to the sudden surge in raw material prices. Currently the availability of maize in the market is poor. Under such circumstances buyers such as feed manufacturers who must buy raw materials are held at ransom by opportunistic traders. Prices of wheat by-products have increased tremendously. This has been attributed to global increase in prices. Protein sources such as cotton cake, sunflower cake, soya cake and Omena are becoming scarce and expensive with retail prices increasing by 77%, 75%, 70% and 50%, respectively, Garnworth and Wiseman, (2006). *Omena*, a local fish, has been the only reliable source of animal proteins that feed millers

have had for feed formulation nowadays. The interest to the Kenya feed millers is the opportunity to make a low cost and good quality feed. They recognized that feed standards should be improved to be in tandem with international benchmarks. A solution to improve their cost position would be to adopt technological advancements to produce feed cost efficiently and in line with current international standards as well. Experts have suggested that by adding enzymes to raw materials, Kenyan feed millers will be able to effectively enhance the range of the raw materials available, and improve efficiency of feed utilization. Farmers using the improved feed will witness numerous benefits as well, like lower mortality, shorter production cycles and improved litter conditions. Ultimately, these savings will be passed on to poultry consumers in a region where poverty is prevalent, Onudi, (2005).

2.3 Raw materials used in poultry feed production in Kenya and their limitations

The raw materials used to compound poultry feeds in Kenya are divided into four main groups namely energy, protein and mineral sources as well as vitamin-mineral premixes. Cereal grains (maize, sorghum) as well as the milling by-products (wheat pollard, wheat bran, maize bran, maize germ meal/cake) are the important energy sources. Oil seed meals or cakes are used as the plant protein sources and include soybean meal, cotton seed cake as well as sunflower seed cake. Wheat pollard, a by-product of wheat milling, is also used as protein source. The material is less fibrous than wheat bran and has a higher feeding value. It also contains a higher proportion of gluten which is a good natural binder for pelleted feed production, Jackson *et al.*, (1982). Sunflower seed cake made from de-hulled seeds has fairly high protein content. It has also been shown to have high amounts of crude fibre, but richer in the sulfur amino-acids than soya bean meal, Jackson *et al.*, (1982).

Some feedstuffs of plant origin contain factors that negatively affect the performance of the birds. The best known examples are the trypsin inhibitors found in raw soybean which can be destroyed by heating the beans, Jackson, *et al.*, (1982). Cereal grains and cereal by-products are important sources of energy. Carbohydrates such as starch and sugars are readily utilized by poultry to furnish energy. There are however some forms of carbohydrates that negatively affect performance of chickens. These carbohydrates that are associated with poor performance of poultry and pigs are referred to as non-starch polysaccharides (NSPs). The avian digestive enzymes cannot break down the non-starch polysaccharides (NSPs), hence diets high in NSPs are associated with poor performance of poultry.

2.4 Non-starch polysaccharides (NSP) in animal feeds

2.4.1 Definition, classification and occurrence of non-starch polysaccharides

According to Choct, (1997), polysaccharides are polymers of monosaccharide joined through glycosidic linkages and are defined and classified in term of the following considerations:

- (i) Identity of the monosaccharide present
- (ii) Monosaccharide ring forms (6-membered pyranose or 5-membered furanose)
- (iii) Positions of the glycosidic linkages
- (iv) Configuration (α or β) of the glycosidic linkages
- (v) Presence or absence of non-carbohydrate substituent

2.4.2 Classification of Non-Starch Polysaccharides (NSPs)

The term non-starch polysaccharides (NSP) covers a large variety of polysaccharide molecules excluding α -glucan (starch). The classification of NSP was based originally on the methodology

used for extraction and isolation of polysaccharides. The residue remaining after a series of alkaline extractions of cell wall materials was called cellulose, and the fraction of this residue solubilised by acid was called hemicellulose. The word hemicellulose was adopted because early researchers mistakenly regarded these polysaccharides as the precursors of cellulose. This is now known to be incorrect but the term is still commonly used. Some workers have used the terms hemicellulose and pentosans interchangeably because the pentose containing polysaccharides make up the bulk of hemicelluloses, Neukom *et al.*, (1967).

Classification by differences in solubility lacks precision with respect to both chemical structures and biological functions. For example, the term crude fiber (CF) refers to the remnants of plant material after extraction with acid and alkali and includes variable portions of the insoluble NSP. On the other hand, neutral detergent fiber (NDF) refers to the insoluble portion of the NSP plus lignin while acid detergent fiber (ADF) refers to a portion of the insoluble NSP comprised largely, but not exclusively, of cellulose and lignin, Bailey, (1973). The nutritional relevance of values obtained using these methods in monogastric nutrition, therefore is questionable.

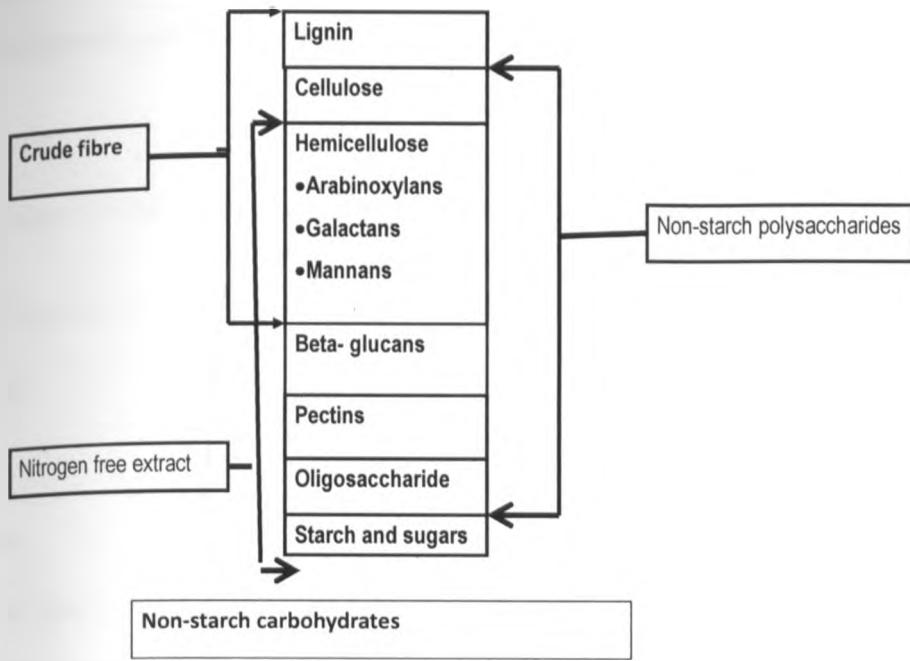


Fig. 1: Non-starch polysaccharides in plant feedstuffs

NSPs fall into three main groups, namely cellulose, non-cellulosic and pectic polysaccharides Bailey, (1973). Besides well digestible nutrients, such as starch and sugars, the carbohydrates fraction of vegetable origin includes indigestible components such as cellulose, hemicelluloses, pectins, β -glucans and lignin. All of these poorly digestible components, excluding lignin, are classified in a group referred to as non-starch polysaccharides (NSPs). NSP fraction is well known for the anti-nutritional effects it can exert.

Within the group of NSP, hemicellulose itself is a heterogeneous sub-group predominantly made xylans, arabinans, galactans, glucans and mannans. Arabinoxylan (AX) is the principal NSP fraction which is found in several of the most important feed raw materials, including wheat and corn, Choct, (1998) as shown in Table 1 below. Arabinoxylans (AX) is one of most important anti-nutritional factor found in cereals and cereal by-products. It increases water binding capacity of the digesta leading to increased viscosity of the intestinal content, resulting in decreased

digestibility and absorption of nutrients, sticky droppings and increased microbiological proliferation and finally poor performances, Choct, (1997).

Table 1: NSP components of feed ingredients (% dry matter)

Feed ingredient	AX sol	AX insol	β -glucans	Cellulose	Mannose	Galactose	NSP	AX/NSP %
Wheat	1.8	6.3	0.8	2.0	T	0.3	11.4	71
Rye	3.4	5.5	2.0	1.5	0.3	0.3	13.2	67
Maize	0.1	5.1	T	2.0	0.2	0.6	8.1	64
Wheat bran	1.1	20.8	0.4	10.7	0.4	0.8	35.3	62
Sorghum	0.12	3.8	0.2	2.0	0.1	0.15	6.45	62
Wheat DDGS	4.9	13.4	2.3	5.8	T	0.9	33.2	55
Barley	0.8	7.1	4.3	3.9	0.2	0.2	16.7	47
Corn (DDGS)	0.4	12.6	T	7.1	0.7	2.1	28.6	45
Rice bran	0.2	8.3	T	1.2	0.4	1.2	21.8	39
Rice	T	0.2	0.1	0.2	T	0.1	0.8	25

Source: Choct, M.R. (1997); T = trace, DDGS = dried distilled grain soluble.

2.4.3 Characteristics of key non-starch polysaccharides (NSP)

Non-starch polysaccharides (NSPs) include cellulose which is the most abundant compound in nature, and pentosans, (arabinoxylans) which are composed predominantly of two pentoses, arabinose and xylose, pectic polysaccharides, arabans and galactans. Cellulose is the most abundant organic compound in nature, comprising over 50% of all the carbon in vegetation, Goring and Timel, (1962). Cellulose is a linear homopolymer of (1-4)- β -glucose units and is of high molecular weight, Goring and Timell, (1962). It is believed to have identical chemical composition regardless of source. It is insoluble in water and aqueous solutions of alkalis. Cellulose in cereal grain cell walls can be recovered from the insoluble residue left after vigorous extraction of cell wall material matrix components with alkalis, Mares and Stone, (1973a).

The enzymes that can degrade cellulose are called cellulases with β -glucanase being the most common, Goring and Timel, (1962). These enzymes will hydrolyse 1, 4 - β -glycosidic bonds. The 1, 3- β - and 1, 2- β -glucanases appear to be widely distributed in both fungi and yeast, White *et al.* (1981). Endo-1, 3- β -glucanases are involved in hydrolysis of 1, 3- β -glucosidic linkages, while laminarinases hydrolyze 1,3- β - or 1,4- β -glucosidic linkages when the glucose residue whose reducing group is involved in the linkage to be hydrolyzed, Petterssons and Aman, (1988, 1989).

The production of β -glucan-degrading enzymes is a characteristic attributable to wide variety of organisms, although the fungi are most common producers of this enzyme. The many β -glucan-hydrolyzing enzymes are classified according to the type of β -glucosidic linkage(s) they cleave and their mechanism of the substrate attack, Piston *et al.* (1993). It is not known whether a similar type of interaction occurs among the various endo- β -glucanases to enhance their ability to reduce the viscosity of β -glucans and therefore, to improve the nutritional value of the diet.

2.4.3.1 Pentosans (arabinoxylans)

Cereal pentosans (arabinoxylans) are composed predominantly of two pentoses, arabinose and xylose units, and their molecular structure consists of a linear (1-4) β -xylan backbone to which substituents are attached through O₂ and O₃ atoms of the xylosyl residues, Perlin (1951). The major substituents are single arabinose residues although, in many instances, hexoses and hexuronic acids are present as minor constituents, Fincher, (1975). The molecular weights of arabinoxylans from cereal grains can be extremely high depending on the source and extraction method, Mares and Stone, (1973) and Annison *et al.*, (1992). Most of the arabinoxylans in cereal grains are insoluble in water because they are anchored in the cell wall by alkali-labile ester like cross links rather than by a simple physical entrapment, Mares and Stone, (1973).

2.4.3.2 Pectic Polysaccharides

The term pectic polysaccharides refers to galacturonans or rhamnogalacturonans in which (1-4)- α -D-galacturonan chain are interrupted at intervals by insertion of (1-2)- α -L-rhamnose residues, Aspinall and Jiang, (1974). Other constituent sugars attached as side chains include D-galactose, L-arabinose, D-xylose, and less frequently L-fucose and D-glucuronic acid. Most of these sugars occur in short side chain although D-galactose and L-arabinose are often found in multiple units. The molecular weight of the pectins has been reported to range from 30,000 to 300,000, Pilnik and Vorgen, (1970). In soya bean, the L-rhamnose residues in the rhamnogalacturonan backbone have been shown to be joined consecutively and others to alternative with galacturonic acid residues, Aspinall *et al.* (1967).

2.4.3.3 Arabinans and galactans

The arabinans are polymers of (1-5)- α -L-arabinose residues through O₂, O₃ or both positions, whereas the galactans are polymers of (1-4)- β -D-galactose residues. Galactans containing approximately 4% (1-6) - β -linkages in addition to the usual (1-4) linkages have also been found, Ghosh and Das, (1984). Finsher and Stone (1974) isolated a low molecular weight arabingalactan from wheat flour.

Xyloglucans, another unusual group of NSP has been found in rice, Shibuya and Misaki, (1978). The structure of xyloglucans is a (1-4)- β -linkaged glucan backbone with single units of α -xylose attached to the O₆ atoms of the main chain. The physiochemical properties and nutritional activities of these NSP are not yet established, Choct, (1997).

2.5 Nutritional significance of non-starch polysaccharide

Approximately two billion tons of cereal grains and 140 million tons of legumes and oilseeds are produced throughout the world each year, with an estimated 230 million tons of fibrous material as part of a variety of by-products, Choct, (1997). The fibre component of the grain consists primarily of non-starch polysaccharides (NSP) which in cereals form part of the cell wall structure. In grain legumes which are used in monogastric diets to supply protein, it is proved that they also contain substantial amounts of NSP. Cellulose and xylans, which are the major NSP in cereal grains, are only found in hulls or husks of most legumes. The NSP in the cotyledon of legumes are pectic polysaccharides, Choct, (1997).

The role of fibre in monogastrics diets has attracted much attention in recent years, due to the fact that the soluble NSP elicit anti-nutritive effects and that the utilization of NSP as a feed material in monogastrics is very poor. These two factors are of significant importance because the world's population is increasing whereas its food production is static, Choct, (1997). More

efficient utilization of potentially utilisable nutrients for food production is therefore of paramount importance to sustainability of agriculture in the future. The importance of NSP in monogastric diets varies with the raw material considered, Marquardt *et al.*, (1996).

2.6 Types and levels of NSP in common feed ingredients

The NSP content varies not only between ingredients but also within the same ingredient due to variety and geographical location where it is grown, Choct, (1997). However, the main structural feature of the NSP in a particular ingredient is not affected by environmental or varietal factors. The NSP in cereal grains are composed predominantly of arabinoxylans (pentosans), β -glucans and cellulose. Only small amounts of pectic polysaccharides are found in the stem and leaves of cereals with the exception of rice, Shibuya and Nakane, (1984). Maize and sorghum contain very low levels of NSP, whereas wheat, rye and triticale contain substantial amounts of both soluble and insoluble NSP, Shibuya and Nakane, (1984). The main soluble NSP in these grains are pentosans or arabinoxylans. On the other hand, barley and oats contain large amounts of β -glucans. Large amounts of cereal by-products are produced and these products contain high levels of cell wall components and are therefore usually rich in NSP and low in nutritive quality. For example, rice bran, contains approximately 20-25% NSP which consists of approximately equal amounts of pentosans and cellulose, Saunders, (1986).

Table 2: The types and levels of NSP present in some cereal grains and cereal by-products (% dry matter)

	Cereal	Pentosans	β -Glucan	Cellulose	Galactans	Total
Wheat	Soluble	1.8	0.4	-	0.2	2.4
	Insoluble	6.3	0.4	2.0	0.1	8.5
Barley	Soluble	0.8	3.6	-	0.1	4.5
	Insoluble	9.5	1.5	2.5	0.4	13.9
Triticale	Soluble	1.3	0.2	-	0.1	1.6
	Insoluble	9.5	1.5	2.5	0.4	13.9
Sorghum	Soluble	0.1	0.1	-	-	-
	Insoluble	2.0	0.1	2.2	0.15	2.45
Corn	Soluble	0.1	-	-	-	-
	Insoluble	5.1	-	2.0	0.2	7.3
Wheat pollard	Soluble	1.1	0.4	-	0.1	1.5
	Insoluble	20.8	-	10.7	0.7	32.2

Source: Englyst (1989); 2 Choct *et al*, Unpublished

In addition grain legumes are also used in monogastric diets mainly to supply protein. In addition to the protein, they also contain substantial amounts of NSPs. Cellulose and xylans, which are the major NSPs in cereal grains, are only found in the hulls or husks of most legumes seeds. The NSP in the cotyledon of legumes are pectic polysaccharides, Choct, (1997).

Copra meal is used in monogastric feeds in some countries. However, its use in monogastric diets is limited due to a very high level of NSPs, Purwadaria *et al.*, (1995) because it contains about 45-60% of NSP made up predominantly of mannans (galactomannans and mannans) and some cellulose, Sattagaroon *et al.* (1983); Zamora *et al.*, (1989). Sattagaroon *et al.* (1983) reported that approximately 30% of the NSP is soluble in water, but their nutritional properties are yet to be defined which also lowers the feeding value of copra.

2.7 Role and effects of NSP in monogastric nutrition

The insoluble NSPs make up the bulk of the total fibre in the diets, but they have little or no effect on nutrient utilization in monogastric animals. Carre (1990) and Begin (1961) showed no detrimental effect, other than simple nutrient dilution, when up to 21% of cellulose was added to poultry diets. The insoluble NSP, however, are not inert and their roles in monogastric nutrition cannot be neglected. One of the most important attributes of insoluble NSP is their ability to absorb large amounts of water and maintain normal motility of the gut, Stephen and Cummings, (1979). This is essential for the consistency of the excreta in monogastric animals.

Elevated levels of insoluble fibre in the diet shorten the residence time of digesta, and some argued that this leads to lower nutrient digestibility, Kirwan *et al.*, (1974). The rationale is that the longer the feed is exposed to the digestive processes in the gut, the more complete its digestion. This, however, may not be true under all circumstances. Soluble NSP can increase gut viscosity and slow digesta transit time in chickens, which allows the proliferation of fermentative organisms in the small intestine, which is detrimental, Choct, *et al.*, (1996). It is believed that when gut viscosity is decreased and nutrient digestion and absorption are enhanced, the indigestible feed materials pass through the gut quickly and insufficient time is available for anaerobic microflora to establish in the upper part of the gut, Choct, *et al.* (1996).

In Australia, it was observed that some wheat had very low apparent metabolizable energy values when fed to broiler chickens, Mollah *et al.*, (1983). Addition of enzymes to wheat and barley based poultry feeds to hydrolyse NSPs and reduce the negative effects of anti-nutritive factors, minimized variability, and therefore improved ingredient value of the feed. The poor ME values are due to high levels of soluble NSP of the wheat, Annison, 1991 and Choct, *et al.*, (1995).

Rogel *et al.* (1987) demonstrated that adding coarsely-ground oats hulls, more than 90% NSP of which 99% is insoluble to low ME wheat diets largely ameliorated poor nutritive quality of wheat. The effect of oat hulls on digesta transit time was demonstrated, where addition of 10% oat hulls increased the rate of digesta passage significantly, Rogel (1985). Fine-grinding of the oat's hulls, however, rendered them ineffective. The water-holding capacity of fibre changes with particle size. In studies with humans, a coarse wheat bran preparation significantly shortened the digesta transit time and showed beneficial effects on colonic function, whereas a fine bran preparation was completely without effect, Kirwan *et al.* (1974). These authors demonstrated that the beneficial effect of bran was dependant on the water holding capacity, which is a function of particle size. Thus, milling the coarse bran to particle size of 1 mm almost halved the water-holding capacity from 6.15 to 3.54g of water per g of bran. It is possible that the effectiveness of the coarse fibre was due to its ability to hold large amount of water, thereby preventing increased solubilization of NSP. The net was an increased rate of digesta passage, giving little time for fermentative organisms to establish in the gut, especially in the small intestine. This highlights the possibility that perhaps at an appropriate ratio between the soluble and insoluble fractions, the anti-nutritive effect of the NSP may be minimized.

2.8 Nature and the anti-nutritive effects of NSP

The NSPs affect intestinal transit time, modification of the intestinal mucosa, and changes in hormonal regulation due to a varietal rate of nutrient absorption, Vahouny, (1982). The viscosity of NSP depends on their solubility and molecular weights. Solubility of NSP, in turn, depends on the chemical structure of the NSP and their association with the rest of the cell wall components, Edwards *et al.*, (1998); Ikegami *et al.*, (1990). Viscosity, however, is not specific to the sugar composition or linkage types present in the NSP. Generally, high gut viscosity decreases the rate

of diffusion of substrates and digestive enzymes and hinders their effective interaction at the mucosal surface, Edwards *et al.*, (1988).

2.8.1 Effects of NSPs

The effects of Soluble NSPs are their interaction with the glycocalyx of the intestinal brush boarder and thicken the rate limiting unstirred water layer of the mucosa, which reduces the efficiency of nutrient absorption through the intestinal wall, Johnson and Gee, (1981).

The fact that the viscous property of the NSP is a major factor in the anti-nutritive effect of NSP in monogastric diets is supported by the wide-spread use of enzymes in monogastric diets. The enzymes cleavage the large molecules of NSP into smaller polymers, thereby reducing the thickness of the gut content and increasing the nutritive value of the feed Bedford *et al.*, (1991) and Annison, (1992). The dry matter content of the litter of wheat or barley-fed broiler is improved (reduced sticky droppings) by adding enzymes to their diets, Wiedmer and Volker (1989); Jansson *et al.* (1990); Mohammed (1995). The reduced litter condition reduces ammonia building up in sheds and reduces the incidence of hock burns and breast blisters. Also, birds fed high-barley or high-wheat diets have been shown to have elevated intestinal weight, which negatively affects the carcass yield. This negative effect is reduced after supplementation with the appropriate enzymes, Francesh *et al.* (1989); Jeroch and Danicke (1993).

2.9 Treatments for improving nutritive value of specific cereal grains

The feeding value of nutritionally inferior cereals such as rye and barley can be substantially improved if anti-nutritive components are eliminated or their unfavourable effects on nutrients absorption are altered, Annison and Choct, (1991). Several methods have been developed over years to improve the nutritive value of cereals, especially those of rye and barley. These include:

addition of enzymes supplementation, water treatment and antibiotic supplementation, Annison and Choct, (1991). The improvement of the nutritive value of barley, corn, and wheat by a simple water treatment was reported almost five decades ago, Fry *et al.*, (1958); Lepkovsky and Furuta, (1960). Water treatment of rye improves significantly the growth and feed utilization of chicks compared with untreated rye, Fernandez *et al.*, (1973) and increased the retention of protein and the digestibility of amino acids and fat, Antoniou and Marquart, (1982). The positive effect of water treatment is well established and it is most likely the result of the removal of the water-soluble non-starch polysaccharides (pentosans and β -glucans) and the activation of endogenous enzymes capable of degrading these polysaccharides. The degree of improvement is obviously dependent on the concentration of the water-soluble non-starch polysaccharides in the cereal. Thus, Adams and Naber (1969) reported that the response to water treatment was consistently high for barley and wheat, but only occasional positive responses were seen with maize. This is because barley and wheat contain higher levels of NSP than maize, Choct, and Annison, (1990).

There is ample evidence that the anti-nutritive activity of NSP in poultry diets is related to the gut microflora of the chicken. The addition of procaine penicillin to rye-based diets resulted in marked increases in chick growth, in efficiency of feed utilization, Moran *et al.*, (1969); McAuliffe and McGinnis, (1971), as well as feed intake and retention of all nutrients, Misir and Marquardt, (1978).

The responses in performance to anti-biotic supplementation may depend on composition of the diets, notably quantity and quality of the dietary protein, Misiri and Marquardt, (1978). Although the insoluble NSP have mainly been regarded as nutrient diluents in the diet, they can also affect digestion transit time and gut motility, Annison and Choct, (1991). Another facet of the role of insoluble NSP in poultry diets that is worthy of reiteration is their ability to act as a physical

barrier to digestive enzymes, such as amylase and proteases thus reducing their efficient digestion of nutrients embedded in the cell wall matrix of grains, Annison and Choct, (1991). Evidence available appears to suggest that enzymes with affinity for insoluble NSP can elicit a positive response in growth performance of broilers, Cowan, (1995); Choct, (1998). This indicates breakdown of cell wall matrix, especially the soluble components, may facilitate easier access of digestive enzymes to their substrates within the short feed transit time in birds. Wiseman and McNab (1996) have also shown that the rate of starch digestion *in vitro* correlates closely with the apparent metabolizable energy (AME) values of different types of wheat. This suggests that the accessibility of amylolytic enzymes to starch granules differs depending on the wheat type, and some of the key factor influencing it may relate to the cell wall architecture of the wheat. Bedford (2002) demonstrated that a considerable amount of nutrients such as starch remains encapsulated in the cell walls in the small intestine of chickens and is removed upon xylanase supplementation as exogenous enzyme supplemented.

Cowieson (2005) speculated that the ability of enzymes, in particular, glycanases, to enhance the nutritive value of some corn-soy diets is probably mediated through changes in the cell wall architecture of the grain, rather than viscosity reduction as it is often the case for viscous grains. Some of the enzymes that have been used over the past several years or have potential for use in the feed industry include cellulase (β -glucanases), xylanases and associated enzymes, phytase, proteases, lipases, and galactases, Ronald, and Marquardt, (1996) Enzymes in the feed industry have mostly been used for poultry and, to a lesser degree, for pigs to neutralize the effects of the viscous, non-starch polysaccharides in cereals such as barely, wheat, rye and triticale. These enzymes are used to prevent anti-nutritive effects which are undesirable as they reduce digestion and absorption of all nutrients in the diet, especially fat and protein, Bedford, (1995).

2.10 Use of enzyme technology for improved feed utilization in monogastric animals

Cereals such as wheat, barley and rye are incorporated into animal feeds to provide a major source of energy. However, much of the energy remains unavailable to monogastrics due to the presence of non-starch polysaccharides (NSP). Most of selected carbohydrases (enzymes) will break down NSP, releasing nutrients (energy and protein), as well as reducing the viscosity of the gut contents. The overall effect is improved feed utilization and a relatively healthy digestive system of the monogastric, Choct, (1997).

The anti-nutritive activity of soluble NSP with well-defined chemical structures, e.g. arabinoxylans and glucans in cereal grains, is eliminated effectively by supplementation of feed with xylanases and glucanases which cause a partial depolymerisation of the NSP to smaller polymers so that their ability to form highly viscous digesta is greatly reduced. Enzymes capable of effectively cleaving various pectic polysaccharides are, however, yet to be produced. The fermentative capacity of the chicken is limited and therefore it absorbs most of its energy as monomeric sugars, Choct, (1997). The current enzymes are not designed to degrade NSP to monomeric sugars within the food transit time of pigs and poultry. The future challenge for the feed industry is, therefore, to produce highly efficacious enzymes which will lead to the utilization of both soluble and insoluble NSP as energy source for monogastrics animals.

2.11 The non-starch polysaccharides degrading enzymes

More wheat is used to compound poultry feeds in USA, Australia, Canada and United Kingdom than in other countries, Heldander and Inberr, (1989). A considerable amount of research has been done on broilers' responses to wheat-based diets. In some countries, commercial broiler feeds typically contain in excess of 60% wheat and the inclusion of xylanase-based enzymes in these diets is now common. Positive effects on AME, weight gain, feed conversion, protein

digestibility, fat digestibility and litter condition were observed when broiler diets containing a high proportion of wheat were supplemented with non-starch polysaccharide degrading enzymes, Helander and Inborr (1989), Jansson *et al.* (1990), Graham and Harker (1991), McNab *et al.*, (1993), Schurz *et al.*, (1993), Veldman, *et al.*, and Schutte (1995); Rajmane *et al.* (1995); Juin *et al.* (1995).

Responses to enzyme supplementation depend on the bird's age which is apparently related to both the type of gut microflora present and the physiology of the bird. Older birds, because of the enhanced fermentation capacity of the microflora in their intestines, have greater capacity to deal with negative viscosity effects, Allen *et al.*, (1995), Choct *et al.*, (1995); Vukic and Wenk (1995).

While they lack the viscous nature of NSPs found in wheat, barley and rye, NSP in non-viscous cereal grains (examples) pose a physical barrier between the intestine enzymes and cell components, Hesselman and Aman, (1986). In doing so, starch, protein, oil and other nutrients are encapsulated within the plant cell. Energy gained from the complete NSP digestion of the cell walls is insignificant. Instead, the greatest nutritional value is expected from the released components inside the cell.

2.11.1 Enzyme treatments for monogastric diets based on cereals and soya bean meal

Choct, (2001) outlined other detrimental aspects of insoluble NSP of maize (corn) as one of the most cereal used for feeding livestock in USA, however, has the insoluble NSP portion of ingredients which presents the greatest challenge in most US diets. For maize (corn) grain, this would be mainly the arabinoxylans and cellulose. Researchers recently analyzed two groups of US corn grain (n= 23). The arabinoxylan content was close to four per cent, while the cellulose fell within a range of 2-4%. These two components, along with about one per cent pectins,

comprise 90% of the NSP in corn, or 9-10% of the dry matter, Malathi and Devegowda, (2001). The oligosaccharides are of concern in SBM due to their indigestible nature and level, Bach, (2001). The oligosaccharides mainly consist of α -galactosides (raffinose and stachyose) and are not digested by endogenous enzymes. These substances make up to 6% of the SBM dry matter, and are associated with wet litter due to bacterial degradation in the lower intestinal tract. The ratio of Me to gross energy (Ge) in SBM is 0.51 for poultry, NRC, (1994), indicating about 51% of the gross energy in SBM is used for metabolic functions. The removal of oligosaccharides with ethanol resulted in 10-15% or more improvement in AME, Coon *et al.*, (1990).

2.11.2 Enzymes for enhancing bioavailability of P in high phytate feedstuffs

Apart from contributing to improve nutritive value, feed enzymes can also have positive impact on the environment by allowing better use of natural resources and reducing pollution by nutrients. In areas with intensive livestock production, the phosphorus output is often very high and this can lead to environmental problems. Most (50-80%) of the phosphorus contained in feedstuffs of plant origin exists as the storage form known as phytate, or phytic acid, and is indigestible for non-ruminant animals, China MOA, (1995). These animals cannot access the phosphorus contained within these complex phytate structures, since they lack the enzyme to break down the phytate and free the phosphorus. The phytase enzyme is essential for the release of the phytate-bound phosphorus. Phytate also forms complexes with proteins, digestive enzymes and minerals, and as such is considered to be an anti-nutritional factor, Simpson and Ward, (1995). Phytase frees the phosphorus contained in cereals and oilseeds, and by breaking down the phytate structure also achieves the release of other minerals such as calcium and magnesium, as well as proteins and amino acids, which become bound to the phytate. Use of the

enzymes also has the added benefit of helping to conserve natural resources by eliminating the need to supplement feeds with sources of digestible inorganic phosphorus.

2.12 Role of enzymes in broiler chicken diets

The effect of NSP on nutritive value of feeds and effect on poultry performance has been presented. The adverse effects of NSP can be overcome, to a large extent, by use of exogenous feed enzymes. Such enzymes have been shown to improve weight gain, feed intake, apparent protein digestibility, and the AME of the diet, Bird, (1994a). When properly used, enzymes can produce many beneficial effects in the chicken and other kinds of poultry and monogastrics.

2.13 Use of Allzyme SSF in broilers diets

Allzyme ® SSF is a naturally occurring enzyme complex that improves the digestibility of feed, utilizing the power of seven enzymes. The constituent seven enzymes work synergistically to break down the different substrates resulting in more nutrients being available to the animal and giving improved physical and economic efficiency. This enzyme complex is manufactured through solid-state fermentation (SSF) by Alltech, Com. SSF involves the careful selection of a specific strain of naturally occurring fungus that expresses the required activities for a given substrate. Allzyme SSF is produced by a carefully selected strain of *Aspergillus niger*. The enzymes in the complex work synergistically to break down the different substrates such as phytate-bound phosphorus, calcium, energy, and amino acids from poultry feed. As a result, more nutrients are available to the animal, thus improving growth and economic efficiency, Pierce *et al.*, (2009).

Corn-soy bean based broiler diets have traditionally been considered highly digestible with no need of enzyme application. However, Ramesh and Devegowda (2006) investigated the effect of

reformulating corn based diets with and without Allzyme SSF on the performance of broilers. They observed that in diets containing recommended energy levels addition of Allzyme SSF resulted in improved performance in the form of increased final body weight and reduced feed conversion ratio (FCR). In palm kernel meal diets, Allzyme SSF application reduced the anti-nutritive effects of palm kernel meal, increasing the possible inclusion rate without affecting bird performance, Ravindran, (2009). The introduction of palm kernel meal into standard corn and soy bean meal diets for broilers resulted in a reduction in performance, both lower final body weight and higher feed conversion ratio. The use of Allzyme SSF, has allowed nutritionists more flexibility in their ingredient choices without affecting animal performance, at the same time it lead to lower overall diet cost. In India, Ramesh and Devegowda, (2006) demonstrated that Allzyme SSF added on the top of the diet resulted in a significant improvement ($P < 0.05$) in gain (79 g) over a 42-day period. The use of Allzyme SSF maintained performance when reformulated in the diet with 75 kcal/kg less of ME and both available phosphorus and calcium content reduced by 0.1 percent. Allzyme ® SSF has a broad range of activities on substrates, and therefore can improve the digestibility of a range of anti-nutritional factors in feed ingredients. It is effective in improving the digestibility of both common feed ingredients, like corn, wheat, sorghum and soybean meal, but also less common ingredients, including palm kernel meal, dried distillers grain soluble (DDGS), cassava, rice bran and copra meal, Ravindran, *et al.*, (2009).

2.14 Use of Nutrase Xyla (NX) in broiler diets

Nutrase ® Xyla (NX) is a bacterial endo-1,4- β -xylanase, designed for use in pig and poultry rations with a high content of arabinoxylans. It is produced by the bacteria *Bacillus subtilis* strain. It reaches its highest efficiency in rations with a high inclusion of wheat and wheat by-

products. It has an important effect on improving the digestibility of other grains, such as barley, corn, rice, rye and sorghum, Schutte et al.; (1995). Nutrase ® Xylla has many more functions, optimally under a neutral pH (between 6 and 7), since feed spends most time in the small intestine, under neutral conditions, Nutrase Xylla (NX) has a much longer time to exert its activity on the soluble and insoluble arabinoxylans (AX) fractions to release nutrients and reduce viscosity of digesta, Choct, (1998). Mathlouthi *et al.*; (2002) stated that enzymes application in poultry diets perform some or all of the benefits such as maintaining good performance on poorer quality feed, decreased formulation cost, widens range of raw materials (feedstuffs), overcome inconsistency and anti-nutritional factors of raw materials, and decrease nutrients and water excretion and encourage better economic returns. In a similar study, Choct, (1998) reported inclusion of Nutrase Xylla in wheat based-broiler diets improved digestibility of fat and enhanced the ME of the diet.

2.15 Role of feed enzymes in reducing feed cost

The use of enzymes in poultry diets is widespread, owed largely to the cost and availability of ingredients. The benefits from feed enzymes such as Nutrase Xylla, Allzyme SSF, phytase and α -galactosidase are to increase nutrients availability from a given ingredient, less of that ingredient need to be included in the diet to begin with. For example, by breaking the phytate bonds, significantly more calcium, phosphorus and amino acids can be made available from feed ingredients, Yi *et al.*; (1996). These “bonus” nutrients could in turn be included in the feed formulation parameters. Consequently, one would obtain a given formulation with lower ingredient levels, Marquardt *et al.* (1996). In feeding trial where Ramesh and Devegowda, (2006) added Allzyme SSF on top of corn-soya bean diet fed to broilers, the result was improved performance in the form of final body weight and reduced feed conversion ratio. Also the diet

was reformulated to account for the additional energy, calcium and phosphorus, released by the Allzyme SSF, reduced diet cost without affecting either weight gain or feed efficiency.

CHAPTER THREE: MATERIALS AND METHODS

3. Effects of inclusion of feed enzymes in broiler chicken diets and their impact on broiler performance and production costs of feeds

3.1 Introduction

The purpose of this study was to assess the effect of inclusion of feed enzymes in broiler chicken diets and their impact on broiler chicken performance and saving cost of feed. Globally, considerable research has been carried out with enzyme treatments in monogastric diets particularly to investigate the role of enzymes in utilization of dietary non-starch polysaccharides (NSP) and concomitant animal performance. The NSP are generally not digested by poultry and addition of enzymes is expected to increase their degradation, improve efficiency of feed utilization and thus enhance performance of birds. Although various feed enzymes might be commercially available in Kenya their use in the local poultry industry is very limited. This is due to lack of information on their utilization, effect on productivity and overall economic benefits.

This study was therefore designed to practically evaluate the effects and potential of including selected feed enzymes in broiler chicken diets. The objectives of this experiment were:

- (i) To evaluate the effect of inclusion of two enzymes namely, Allzyme SSF and Nutrase[®] Xylla on growth performance in broiler chickens
- (ii) To determine the effects of the two enzymes on energy and feed utilization in broiler chickens
- (iii) To assess the cost effectiveness of using the feed enzymes in broiler chicken rations.

To accomplish the above objectives, a broiler feeding trial was conducted for a 49 day period where the pertinent performance parameters were assessed including laboratory analysis of the experimental diets. On the other hand, cost effectiveness of use of enzymes in poultry diets, was

determined for seven days after feeding trial using three main high energy (HE) diets namely D1 (without enzyme addition), D4 supplemented with Nutrase Xylla and D7 with Allzyme SSF addition. 12 birds per each high energy diets from feeding trial were fed for seven days, and data on feed intake, body weight and prices of feeds was computed to assess economic use of feed enzyme in broiler chicken diet.

3.2 Experimental diets

The study was carried out in the poultry unit, Department of Animal Production, University of Nairobi. Two feed enzymes were used in this study, namely Allzyme SSF and Nutrase Xylla (NX). Allzyme SSFTM is an enzyme complex containing seven constituents of enzymes of different composition and functions. Nutrase Xylla is an endoxylanase argeting the degradation of arabinoxylans. The two enzymes were obtained from the appointed dealers in Nairobi. Three main diets, namely high energy (HE), medium energy (ME) and low energy (LE) were formulated to provide 100, 95 and 90% respectively, of metabolizable energy (ME) requirements of broiler chicken, based on Kenya Bureau of standards, KEBS, (1978); Scott *et al.* (1982) specification. The high energy diets contained 3000 kcal/kg metabolizable energy, while the ME and LE diets contained 2850 and 2700 kcal/kg of metabolizable energy, respectively. From the three main diets, nine experimental diets were made by adding or omitting the feed enzymes, as shown in Table 3 and 5 for both the starter and finisher phases. Diets 1, 2 and 3 were HE, ME and LE respectively without enzyme addition. Correspondingly, diets 4, 5 and 6 were HE, ME, and LE, all with the enzyme Nutrase Xylla added while diets 7, 8 and 9 were HE, ME and LE, respectively, to which the enzyme Allzyme SSF was added. Both enzymes were included in the feed at the rate of 100 gm per 1000 kg of feed for enzyme-containing diets, the enzyme was first mixed with the vitamin-mineral premix and other micro-ingredients prior to mixing with the rest

of the ingredients. The entire feed was then thoroughly mixed using horizontal animal feed mixture of 7.5 KW, with a horizontal shaft holding paddle agitators which mixes feed ingredients to ensure uniform dispersion of the enzyme in the diets.

The feed ingredients used for preparing the broiler diets in this study, were procured from commercial ingredient vendors in Nairobi. The coarse ingredients were first ground with a hammer mill to give the appropriate particle size for preparation of the diets. The feed ingredients used for feed cost analysis were mixed in same procedures used for feeding trial to mix three high energy diets namely D1, D4 and D7 respectively.

3.3 Management of experimental birds and design of experiment

Three hundred and sixty (360) day-old unsexed Arbor Acre chicks were purchased from a commercial hatchery (Kenchic Limited) in Nairobi for the feeding trial. On arrival, the chicks were weighed in groups of ten and each group randomly placed in one of 36 pens (experimental units) in a well ventilated house. Each pen had a floor space of 1 m². Each group of chicks was then randomly assigned to one of the nine experimental starter diets (Table 3). This gave nine dietary treatments with four replicates of ten chicks each, in a 3×3 factorial design. The broiler chicks were fed on the starter diets from day old to three weeks of age, and finisher diets (Table 5) from the fourth to the seventh week of age.

Prior to the arrival of the chicks, the house and the equipments were cleaned and disinfected using Omnicide® disinfectant. Wood shavings litter was spread in each pen to a thickness of 4-5 cm. Infra red bulbs were used for brooding for the first three weeks to provide a floor level temperature of 35⁰C for the first week. Thereafter, the temperature was reduced by about 3⁰C each week until the normal room temperature between 22-23 ⁰C was attained. The chicks were vaccinated against Newcastle disease and infectious bronchitis (IB) on the 10th day of age and

against infectious bursal disease (IBD) also called Gumboro diseases on 14th day of age. Feed and water were offered *ad libitum*.

Table 3: Composition of high (HE), medium (ME) and low (LE) energy broiler starter diets

Energy level	High energy (%)	Medium energy (%)	Low energy (%)
Maize	48	48	46
Wheat pollard	15	21	27
Corn oil	3	2	1
Corn gluten meal	5	4	4
Soybean meal	17	15	11
Omena ¹	10	8	8
Coccidiostat	0.1	0.1	0.1
DCP ²	0.1	0.1	0.1
Limestone	1	1	1.5
Common salt	0.5	0.5	0.5
Vitamin-mineral premix ³	0.25	0.25	0.25
Total (%)	100	100	100

¹Fish by the name *Rastrineobola argentea*, which is dried, ground whole and included in poultry feeds;

²Dicalcium phosphate; ³Vitamin-mineral premix- Composition of the premix was: Vitamin A i.u 10,000,000; Vitamin D3 i.u 2,000,000; Vitamin E i.u 24,000; Vitamin K3 mg 3,200; B1 mg 1,600; Vitamin B2 mg 5,600; Nicotinic Acid mg 32,000; Pantothenic Acid mg 8,000; Vitamin B6 mg 4,000; Vitamin B12 mg 24; Biotin mg 96; Folic Acid mg 960; Choline chloride mg 350,000; Manganese mg 150,000; Iron mg 40,000; Zinc mg 45,000; copper mg 5,000; Cobalt mg 200; Iodine mg 1,400; Selenium mg 120.

Table 4: Amount of enzyme used in the starter and finisher diets

Feed enzyme used	No enzyme			Nutrased Xylla			Allzyme SSF		
	HE	ME	LE	HE	ME	LE	HE	ME	LE
Energy									
Diet	1	2	3	4	5	6	7	8	9
Enzyme, (%)	0	0	0	0.1	0.1	0.1	0.1	0.1	0.1

Figures 1, 2 and 3 were assigned to HE, ME and LE no enzyme diets, 4, 5 and 6 were assigned to HE, ME and LE diets added to Nutrased Xylla, while figures 7, 8 and 9 were named as HE, ME and LE diets added to Allzyme SSF

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Table 5: Composition of high (HE), medium (ME) and low (LE) energy broiler finisher diets (%)

Energy level	High energy	Medium energy	Low energy
Maize	48	48	46
Wheat pollard	23	24	26
Corn oil	3	3	1
Wheat bran	5	8	11
Corn gluten meal	3	2.5	2.5
Soybean meal	10	7.5	4.5
Omena	6	5	5
DCP	0.1	0.1	0.1
Limestone	1.5	1.5	1.5
Common salt	0.5	0.5	0.5
Vitamin-mineral premix	0.25	0.25	0.25
	100	100	100

*Vitamin-mineral premix was as follows- Composition of the premix was Vitamin A i.u 10,000,000; Vitamin D3 i.u 2,000,000; Vitamin E i.u 24,000; Vitamin K3 mg 3,200; B1 mg 1,600; Vitamin B2 mg 5,600; Nicotinic Acid mg 32,000; Pantothenic Acid mg 8,000; Vitamin B6 mg 4,000; Vitamin B12 mg 24; Biotin mg 96; Folic Acid mg 960; Choline chloride mg 350,000; Manganese mg 150,000; Iron mg 40,000; Zinc mg 45,000; copper mg 5,000; Cobalt mg 200; Iodine mg 1,400; Selenium mg 120** Diets 1,2 and 3 had no enzyme, whiles diets 4,5 and 6 contained Nutrase-Xylla and diets 7,8 and 9 had Allzyme® SSF. Both enzymes included at the rate of 100gm per 1000 kg of feed.*

3.4 Data collection

Data on broiler performance parameters was collected over 49 days of the feeding trial. The variables measured were feed intake, body weight gain and mortality. From this data, body weight gain, feed intake and feed conversion ratio (FCR) were computed.

Feed cost benefits (Table 18) were computed among three main high energy diets, namely D1, D4 and D7 to determine the effect of enzymes in saving cost of feed.

3.4.1 Feed intake

This was calculated weekly as the difference between feed offered at the beginning of the week and feed left over at the end of the same week. The mean feed intake in grams per bird per week was obtained by dividing the total intake for a particular replicate (pen) with the total number of birds in the replicate for that week. The treatment mean was accordingly calculated as the mean of the four replicates. Feeds were kept and fed to birds inside hanging round plastic feeders which minimize feed wastage and any waste of feeds particles was not put under considerations.

3.4.2 Body weight gain

Body weight per replicate was taken every week from the first to the seventh week. Body weight gain per week was obtained as the difference in body weights per replicate between consecutive weeks. Mean weight gain per treatment was calculated as a mean of the four replicates.

3.4.3 Feed conversion Ratio (FCR)

The feed conversion ratio was calculated as the ratio of feed consumed to body weight gain (feed: gain) per replicate per treatment.

3.4.4 Mortality

The chicks were observed daily and any bird that died was recorded.

3.5 Laboratory analysis

Samples of feed ingredients and experimental diets used in the study were analysed for dry matter (DM), crude protein (CP), ether extract (EE), crude fibre (CF) and ash according to procedures of the Association of Official Analytical Chemists (AOAC, 1998). The diets were also analysed for soluble and insoluble arabinoxylans (NSP of wheat and wheat by products) according to the procedures of Englyst and Cummings, 1984. The samples of diets used during feed economic analysis, Table 18 were analysed for gross energy using adiabatic bomb calorimeter, Table 18, according to Hill, Seals and Montiegel, (1958).

3.6 Statistical Analysis

Data obtained on feed intake, body weight gain and feed conversion efficiency was entered, using Microsoft Excel 2007™, then it was analyzed using two-way Analysis of variance (ANOVA). The statistical programme used was Genstat Discovery edition 3. Standard error of differences of means (SEM) was calculated to compare variation among treatments where ANOVA showed significant differences among the means.

3.7 Economic evaluation

The market prices of feed ingredients in May, 2010 (when this study was started), were used to compute the break-even prices of formulated three high energy diets. By the end of the seven days after feeding experiment, the mean feed intake, body weight gain, sale of birds were computed per bird per main three high energy diets (D1, D4 and D7) respectively, to get economic benefits of feed enzymes.

CHAPTER FOUR: RESULTS AND DISCUSSION

The purpose of this study was to determine the effect of adding two types of feed enzymes in diets containing varying energy levels on broiler chicken performance. Feed intake and weight gain were monitored for 49 days.

4. Chemical composition of diets

The chemical composition of the diets is shown in Table 6. The slight variation in nutrient content from the formulated was attributed to the deviation of the quality of the raw materials from the expected. The analysed CP content of the starter diets was 220, 210 and 200 g/kg for HE, ME and LE diets, respectively, while for finisher it was 200, 190 and 195 g/kg. The CP levels of the diets were within the recommended standards, KEBS, (1978); Scott *et al.* (1982); NRC, (1984). The crude fibre in the starter diets were 46, 58 and 61 g/kg and 52.4, 58.9 and 57 g/kg for finisher diets for the HE, ME and LE diets respectively and were all within the recommended level. The higher ether extract (EE) contents in the high energy were due to inclusion of fat.

Table 7 shows the levels of total non-starch polysaccharides (NSPs) in broiler starter and finisher diets. The low energy diets had slightly higher levels of AX (6.93 and 9.16 for starter and finisher) compared to 6.58 and 7.72 for high energy diets. The higher NSPs in the LE diets can be attributed to the inclusion of higher levels of fibrous wheat by-products (bran and pollard). It is recognized that poultry diet contains variable levels of poorly digested NSP including arabinoxylans and pectins, Campbell and Bedford, (1992) that possess chemical cross-linking between them and are not well digested by poultry, Adams and Pough, (1993).

Table 6: Chemical composition of starter and finisher diets (g/kg DM)

Item	Starter diets			Finisher diets		
	High	Medium	Low	High	Medium	Low
Dry matter	904.0	899.0	885.0	910.0	908.0	909.0
Crude protein	210.0	220.0	216.0	200.0	190.0	195.0
Crude fibre	46.0	58.0	61.0	52.4	58.3	58.0
Crude fat	65.6	57.1	44.3	65.7	60.3	57.1
Ash	85.9	77.4	71.3	84.4	81.5	104.8
Nitrogen free extract	656.0	681.0	671.0	668.0	685.0	679.0
Total phosphorus	5.5	6.5	7.0	6.0	6.0	5.5
Calcium	11.2	11.7	11.4	11.5	11.6	11.5

The level of soluble arabinoxylans in the diets is shown in Table 8. The starter diets had lower mean content of soluble AX (0.17 mg/g) compared with the finisher diets (0.21mg/g), with an increase of approximately 19 %. Within the starter diets, the medium energy diet had lower AX while the HE and LE was similar (0.18, 0.14 and 0.19 mg/g (for HE, ME and LE respectively). There was an increase in the level of AX within the finisher diets as the level of energy decreased (0.21, 0.24 and 0.28 mg/kg) for HE, ME and LE, respectively.

The high levels of soluble arabinoxylans in the low energy diets were expected as the diets were formulated with higher levels of wheat by-products. With the inclusion of the enzymes (Nutraze Xylla and Allzyme SSF) which were expected to be capable of opening up the complex feed cell walls, allowing the animals own enzymes to digest the enclosed nutrients. Supplementing broiler

diets with separate or combinations of enzymes such as xylanase, cellulase, pectinase and β -glucanase minimizes the adverse effects of NSPs and improve the nutritive value of the diet, Campbell *et al.*, (1989); Francesch *et al.* (1989); Helander and Inberr (1989); classen *et al.* (1995) and Choct *et al.* (1995).

The impact of soluble arabinoxylans is by reducing feed digestibility and nutritional value of the raw materials in which they were present. Choct *et al.*, (1995) indicated that when levels of NSP in the digesta are high or viscous, an active microflora is present in the ileum as measured by level of short chain fatty acid production, which may have a detrimental effect on the bird. The quality and quantity of NSP both soluble and insoluble in wheat pollard are 1.1% and 20.8% dry matter, and 2.6% and 26% dry matter for wheat bran respectively, Englyst (1989).

Table 7: Total arabinoxylans (AX) in broiler starter and finisher diets (mg/g)

Energy level	Arabinose	Xylose	Mannose	Galactose	Glucose	(AX)
Starter diets						
High	3.09	4.39	0.48	1.61	43.40	6.58
Medium	2.37	2.90	0.48	1.87	46.07	4.64
Low	3.23	4.64	0.4	1.32	45.02	6.93
Finisher diets						
High	3.53	5.24	0.49	1.45	47.06	7.72
Medium	3.50	5.10	0.49	1.24	48.17	7.57
Low	4.17	6.24	0.48	1.29	43.82	9.16

Chemical analysis of total arabinoxylans (AX) in broiler starter and finisher diets (mg/g).

Table 8: Soluble arabinoxylans (AX) in broiler starter and finisher diets (mg/g)

Energy level	Arabinose	Xylose	Mannose	Galactose	Glucose	Arabinoxylans
Starter diets						
High	0.11	0.09	0.30	0.65	2.35	0.18
Medium	0.09	0.07	0.25	0.74	2.20	0.14
Low	0.11	0.11	0.28	0.52	2.14	0.19
Mean	0.1	0.09	0.28	0.64	2.23	0.17
Finisher diets						
High	0.12	0.12	0.34	0.53	2.43	0.21
Medium	0.13	0.14	0.27	0.45	2.43	0.24
Low energy	0.15	0.17	0.32	0.48	2.55	0.28
Mean	0.13	0.14	0.31	0.49	2.47	0.21

Chemical analysis of soluble arabinoxylans (AX) in broiler starter and finisher diets

4.1 Chicks performance during 1-21 days (starter phase)

The effect of supplementation of the diets with non-starch polysaccharide degrading enzymes (NSPDE) namely Allzyme SSF and Nutrase Xylla on feed intake, body weight gain and feed conversion rate (FCR) during starter phase are shown in Tables 9, 10 and 11.

Effects of feed enzymes and energy levels on the feed intakes of birds at 1-21 days of age are shown in Table 9.

Table 9: Effect of energy level and enzyme on feed intake (g/ bird) at 1 to 21 days of age

Energy level	No enzyme	Nutrase-Xylla	Allzyme ® SSF	Mean	SEM
High	1050 ^{xa}	933 ^{ya}	932 ^{ya}	971 ^a	34.3
Medium	1028 ^{xb}	871 ^{yb}	865 ^{yb}	921 ^b	
Low	816 ^{xc}	829 ^{xc}	905 ^{yc}	850 ^c	
Mean	964 ^x	877 ^z	900 ^y	914 ^y	
SEM		19.8			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different;

^{abc} Means within the column are significantly ($P < 0.05$) different.

SEM = Standard error of means.

4.1.1 Effect of energy on feed intake of broiler chicks at 1-21 days

Feed intake during starter phase (1-21 days) is shown in Table 9. The Mean feed intakes of different feeds types, decreased ($P < 0.05$) with decrease in energy level of the diets (971, 921 and 850 g/bird) for HE, ME and LE diets respectively. This was unexpected as poultry have been reported to increase feed intake with decreased metabolizable energy to satisfy their energy requirements, NRC, 1984. This trend was observed for birds fed diets without enzyme addition (1050, 1028 and 818 g/bird) for HE, ME and LE diets respectively. It was also observed with chicks fed diets added to Nutrase Xylla that the feed intake decreased with decrease in energy level (933, 871 and 829 g/bird) for HE, ME and LE diets respectively. The feed intakes of Allzyme SSF diets (932, 865 and 900 g/bird) indicate that FI of birds fed high energy diets added

to Allzyme had more than those fed diets at ME and LE diets, but LE diets had higher FI compared to ME diet.

It was observed that more feed was consumed by birds fed non-supplemented diets. This was clearly seen at both high and medium energy levels while at low energy levels, the Allzyme supplemented diets had the highest feed intake at low energy diet compared to Nutrase Xylla which was expected to hydrolyze the soluble and insoluble arabinoxylans fractions and thus, it was noted that feed intake of Nutrase Xylla supplemented diets did not affect the feed intake of broiler chicks during starter feeding phase, this was in line with findings by Teves *et al.* (1989) and Pluske *et al.* (1997) who reported that the inclusion of Allzyme SSF (Mannanase) in a copra meal diet improved feed intake of broiler chick at low energy density diet. The birds fed on Allzyme SSF supplemented low energy diet had a higher intake compared to LE diet with no enzyme. These birds might have benefited from the enzyme inclusion, which was expected to hydrolyze different kinds of NSPs of the feed ingredients used in the current study, this had shown the effect of both enzyme and energy on feed intake and thus interaction between energy and enzyme inclusion. This result is in agreement with findings of Ravindran (2009) who fed broiler chicken with palm kernel meal added to Allzyme SSF and found reduced effect of anti-nutritional effects of palm kernel meal.

4.1.2 Effect of enzymes on feed intake during starter feeding phase

There were significant ($P < 0.05$) differences among the birds fed diets supplemented with Allzyme SSF compared to Nutrase Xylla that receiving supplemented diets. Improvement of feed intake due to Allzyme over Nutrase Xylla may be due to reduction in bulkiness and intestinal viscosity which is accompanied with presence of reduced soluble arabinoxylans.

Allzyme SSF being a multipurpose enzyme, Alltech. (2010), containing glucanase, xylanase, amylase, pectinases, protease, cellulose and hemicellulases, is better in this regard than Nutrase Xylla which is a single purpose enzyme containing xylanase. Ramesh and Devegowda, (2004), reported that Allzyme SSF possess activities of seven enzymes which is capable of breaking down protein, pentosans, cellulose, phytase and starch, therefore, improving the availability and absorption of nutrients in the avian intestinal tract.

There was no clear pattern on the effect of inclusion of enzyme on feed intake. Overall, the absence of enzyme resulted in the highest intake followed by the Allzyme supplemented diet and least for the Nutrase Xylla supplemented diets. Within the different energy levels, the HE and ME had highest feed intakes for diets without enzyme while for LE diets, the Allzyme supplemented diet had the highest feed intake. The inclusion of two enzymes was expected to improve broiler chick feed intake, and to increase palatability and availability of nutrients, leading to greater feed efficiency. Decreased feed intake on addition of enzyme has been reported by Samarasinghe *et al.* (2000), Richter *et al.* (1995); Ranade and Rajmane (1992); Kadam *et al.* (1991). They found that feed intake decreased on addition of enzymes due to bird fulfill their nutrient requirement by taking less amount of feed. However, this finding differ with observations of Davey *et al.* (1998); Augelovicova and Michalik (1997); Leeson *et al.*, 1996; Pettersson and Aman (1992, 1989), who all conducted research work on Alquerzim, Roxazyme-G, and Feedzyme which were added to iso-energetic and iso-nitrogenous diets for starter and finisher chicks, they suggested increased digestibility of nutrients and partial degrading of cell wall of feed as reasons for increased feed intake on enzymatic diets.

Mean total feed intake (914 g/bird) Table 9, was not affected by enzyme supplementation in the diets which shows that total feed intake means of the diets supplemented with both Nutrase Xylla and Allzyme SSF were low with means of 877.7 and 900.7 g/bird respectively. Non-

supplemented diet had final mean feed intake of 964.7 g/bird. Supplementation of Allzyme SSF and Nutrase Xylla did affect feed intakes of the birds fed low energy diets (905 and 829 g/bird respectively) compared to feed intake of non-supplemented diet which had the lowest intake of (816 g/bird). The final feed intake was depressed ($P < 0.05$) with enzymes inclusion (both Allzyme SSF and Nutrase Xylla). The insignificant effects of the added enzymes on feed intake of broiler chicks during this feeding phase indicates that the enzymes at this level of inclusion (0.1/kg) did not elicit any beneficial response on utilization of NSPs in the diets fed to broiler chicks. In addition, the two enzymes did not completely hydrolyzed different kinds of NSPs such as soluble and insoluble arabinoxylans which are the most important factors in cereals and cereal by-products used in livestock feeds and have negative effect on feed intake. Feed intake during this feeding phase however, was independent of the inclusion of NSPs and their enzymes supplementation. The results coincide with the findings of Abbas *et al.*, (1998) and Naqvi *et al.* 2004, who noticed non-significant difference in feed consumption among diets with or without enzymes supplementation. In contrast to this study by Marquardt *et al.*, (1994) showed that enzyme supplementation resulted in increased feed consumption.

There was significant ($P < 0.05$) interactions between low energy diets and the enzyme supplementation of both Allzyme SSF (904 g/bird) and Nutrase Xylla (829 g/bird) on feed intakes of the birds, compared to low energy diet of non-supplemented (816 g/bird). On the other hand, there was no significant ($P < 0.05$) interactions between the energy levels of the diets and the enzymes supplementation on feed intake of the bird fed HE and ME energy diets with enzymes as feed intakes of birds fed He and ME diets without enzymes were significantly higher compared to those with exogenous enzymes addition.

Effects of supplementing diets with both (Allzyme and Nutrase Xylla) and Non-supplemented diet on body weight of broiler chickens fed at different levels of energy during 1-21 days of age are shown in Table 10.

Table 10: Effect of energy level and enzyme inclusion on weight gain (g/bird) from 1 to 21 days of age

Energy level	No enzyme	Nutrase Xylla	Allzyme SSF	Mean	SEM
High energy	418 ^{xb}	450 ^{ya}	408 ^{xb}	425.3 ^a	
Medium energy	456 ^{xa}	444 ^{xa}	400 ^{yb}	433.3 ^a	30.7
Low energy	447 ^{xa}	410 ^{yb}	449 ^{xa}	445.3 ^a	
Mean	440.3 ^x	434.6 ^x	419 ^y	434.6 ^x	
SEM		17.7			

^{xy}Means with different superscript within the rows are significantly ($P < 0.05$) different

^{ab}Means with different superscripts within the columns are significantly ($P < 0.05$) different

SEM = Standard error of mean.

4.2.1 Effect of energy on weight gain during 1-21 days

Body weight gains (g/kg) during starter phase (1-21 days) are shown in Table 10. The mean body weight gains at different energy levels were not significantly different (425, 433 and 445 g/kg for HE, ME and LE, respectively). The addition of enzyme, irrespective of the energy level, seemed to decrease the body weight gain (450, 435 and 419g/bird for No enzyme, Nutrase Xylla and Allzyme SSF, respectively). The body weight gains of birds fed low energy Allzyme added diet had high body weight gain than No enzyme added and Nutrase Xylla diets. Allzyme fed birds might have benefited from various nutrients released by enzymes inclusion. Unexpected

condition with bird fed non-supplemented high and medium energy diets was observed, where the birds appeared with higher weight gains than enzymes added diets at same energy level. This was attributed to high feed intake of birds fed non-supplemented diet, Table 9. Non-starch polysaccharides degrading enzymes did not show significant effect on weight gain during this feeding phase.

4.2.2 Effect of enzymes on weight gain

Within the high energy diets, birds receiving Nutrase Xylla supplementation had a higher ($P<0.05$) weight gain compared to the others, which indicates that there is interaction between the enzyme and energy level. Within the ME diet, the Allzyme supplemented diet had the lowest ($P<0.05$) weight gain while within the LE diet, Nutrase Xylla had the lowest ($P<0.05$). The expectation was an improvement in performance of birds on ME and LE diets on addition of enzyme compared with the non-supplemented ME and LE diets. This does not appear to have happened. The depressed growth observed in broilers fed low energy Nutrase Xylla added diet, might be due to incomplete hydrolysis of the soluble arabinoxylans (arabinose and xylose) which were more at low energy diets, might have increased viscosity of intestinal digesta. The viscosity caused by water soluble NSPs dramatically reduces bird performance, Choct and Annison (1992b). The high fibrous content of low energy diets might have not been completely hydrolyzed by Nutrase Xylla addition and has been the reason for depressed weight gain, and subsequently poor absorption of nutrients from the feed and thus reduced body weight gain. The weight gain of birds fed Nutrase Xylla added diets at high and medium energy showed numerical high weight gains compared to Allzyme fed birds at high and medium energy diets. This was

associated with a significant increase in feed consumption of birds fed Nutrase Xylla added diets, Table 9.

The body weight gain of Nutrase Xylla decreased slightly with decreased in energy levels of the diets. This can be explained that, the diets contained Nutrase Xylla, did not compensate reduced energy content of the rations. Nutrase Xylla inclusion was expected to break down soluble and insoluble arabinoxylans to reduce viscosity and liberate nutrients. The failure to do this may be attributed to the fact that Nutrase Xylla at the level at which it was included (100g/MT) did not affect broiler chickens weight gains, Sekoni *et al.*, (2008), reported that body weight gains of broiler chicks were depressed by the treatment diets, despite the supplementation of Nutrase Xylla at 10g/100 kg, 20g/100kg, 30g/100kg, 40 g/100kg and 50g/100kg per diets respectively. This finding agrees with that of Sekoni, *et al.*, (2008), who reported that broilers fed on African yam bean meal based diets with and without Nutrase Xylla supplementation body weight gain was significantly ($P<0.05$) depressed by enzyme supplementation. On the other hand, in this study, it was observed that body weight gains of chicks fed Nutrase Xylla supplemented diets was higher with birds fed HE and ME diets, compared to those fed on HE and ME Allzyme supplemented diets. This showed that Nutrase Xylla resulted in better performance than Allzyme SSF during this feeding phase. It is suggested that Nutrase Xylla was effective with diets with low fibre content which was added at HE and ME diets than to do with diet added to high fibre added at low energy diets or with more fibre, this implies that Nutrase Xylla did not affect the body weight gain of bird fed low energy diet. It can be concluded that, birds supplemented with Nutrase Xylla had improved ($P<0.05$) body weight gain at HE and ME diets, but not the case with birds fed low energy diet. This agrees with findings of Schang *et al.* (1997) who added Vegpro enzyme at 300 and 200 g/tonne for starter and finisher respectively, they argued that bird fed low nutrient density diet had lower body weight gain (2.59 vs 2.86) than birds fed high

nutrient density diet. There was an interaction between the addition of enzyme (Nutrased Xylla) and energy levels of the diets as the body weight of birds fed high and medium energy diets was increased with increase in energy density of the diets.

Effects of dietary energy and enzymes on feed conversion ratio of broiler chicks at 1-21 days of age are shown in Table 11.

Table 11: Effects of dietary energy levels and enzymes on Feed Conversion Ratio (FCR) per chick during starter phase (1-21 days of age)

Energy level	No enzyme	Nutrased Xylla	Allzyme SSF	Mean	SEM
High energy	2.5 ^{ax}	2.1 ^{az}	2.4 ^{ay}	2.3 ^a	
Medium energy	2.3 ^{bx}	2.0 ^{cz}	2.2 ^{by}	2.1 ^b	0.29
Low energy	1.8 ^{cy}	2.1 ^{ax}	2.1 ^{cx}	2.0 ^c	
Mean	2.2 ^x	2.1 ^y	2.2 ^x	2.2	
SEM		0.17			

^{xy} Means with different superscript within the rows are significantly ($P < 0.05$) different;

^{abc} Means with different superscripts within the columns are significantly ($P < 0.05$) different;

SEM = Standard error of means.

4.3.1 Effects of energy density of diets on broiler chicken during 1-21 days

Feed conversion ratio in terms of feed intake per unit body weight gain during 1-21 days, was improved with decrease in energy levels of the diets (2.3, 2.1 and 2) at HE, ME and LE for non-supplemented, Nutrased xylla and Allzyme SSF diets respectively. This trend was observed with

birds fed Allzyme SSF (2.4, 2.2 and 2.2) per bird for HE, ME and LE respectively, non-supplemented birds had same trend of FCR improved with decrease in energy level of the diet (2.5, 2.3 and 1.8) per bird fed HE, ME and LE diets respectively. On the other hand, Nutrase Xylla had inconsistent trend of feed conversion ratio where HE and LE had FCR of 2.1 per bird and 2 as FCR per bird fed diet containing Nutrase Xylla at ME diet. This explains that Nutrase Xylla had significantly improved FCR at ME energy diet (2) implying better feed utilization. Within HE and ME diets of two diets (no enzyme and Allzyme SSF), FCR is high and show poor FCR, while within LE diets, No enzyme diet significantly improved feed conversion ratio of the bird with ratio of 1.8, implying better FCR. This may be attributed to the fact that bird fed LE no enzyme diet consumed significantly more feed and had higher weight gain and consequently lowered FCR than those fed with enzymes added diets. Supplementation of both enzymes tended to increase FCR during this feeding phase. These findings agree with observation of Paul *et al.*, (2007) who fed laying hens at pullet stage with maize-soy diets supplemented with a composite microbial enzyme, Allzyme SSF and found that enzyme did not affect feed efficiency. On the other hand, the findings differed with Cowan (1990) who reported a 2-3% increase in efficiency of feed utilization by birds fed diets containing enzymes compared to those fed non-supplemented diets.

4.3.2 Effect of enzymes on feed conversion ratio of bird during 1-21 days

Within HE diets, Allzyme SSF had improved FCR with decrease in energy level of diets, while Nutrase Xylla had improved FCR at ME diet (2). Implying that within the diets added to Nutrase Xylla, FCR was significantly ($P < 0.05$) improved at HE, ME and LE diets which have 2.1, 2 and 2.1 respectively. Within LE, all three diets showed better feed conversion ratio per broiler chick,

compared to medium and high energy diets, but no enzyme diet showed the best feed efficiency ratio of 1.8.

There is significant ($P < 0.05$) interaction effect between the enzymes and low energy levels of the diets, as FCR of LE diets show better feed utilization.

4.4. Broiler performance during finisher phase (22-49 days)

The effect of varying dietary energy and enzyme supplementation on feed intake, body weight gain and feed conversion ratio of broilers during the finisher phase are shown in Tables 12, 13 and 14.

The effects of energy density and enzymes inclusion are shown in Table 12, during finisher feeding phase (22- 49 days).

Table 12: Effect of energy level and enzymes inclusion on feed consumption**(g/bird) per finisher bird fed from 22-49 days old**

Energy level	No enzyme	Nutrase Xylla	Allzyme ® SSF	Mean	SEM
High	4455 ^{ax}	4026 ^{ay}	4111 ^{ay}	4197 ^a	
Medium	4534 ^{ax}	3889 ^{az}	4112 ^{ay}	4178 ^a	206.1
Low	3995 ^{bx}	3668 ^{bx}	4332 ^{az}	3998 ^b	
Mean	4328 ^x	3861 ^y	4185 ^z	4125	
SEM		119			

^{abc} Means with different superscript within the rows are significantly ($P < 0.05$) different.

^{xyz} Means with different superscripts within the columns are significantly ($p < 0.05$) different.

SEM = Standard error of means

4.4.1 Effect of energy on feed intake of birds at 22-49 days old

The mean feed intake decreased with decrease in energy level of the diet (4197, 4178 and 3998 g/bird), Table 12, for H.E, M.E and L.E diets respectively, LE diet feed intake was significantly lowered ($P < 0.05$). This observation was not expected, as birds are expected to consume more feed at low energy concentration, to compensate for energy requirements. Scott and Associates, 1976 reported that chicks consume more food largely to satisfy an inner need for energy. This could be due to bulkiness of feed in the gut and may have relationships with reduction in feed intake observed during this feeding phase. The same trend was observed with feed intake of birds fed Nutrase Xylla diets, which showed that feed intake decreased with decrease in energy level of the diet. The energy significantly ($P < 0.05$) reduced feed intake of birds fed diets with inclusion of Nutrase Xylla resulting in the lowest intake (4026, 3889, 3668 g/bird). On the other

hand, energy significantly ($P < 0.05$) affected feed intake of birds fed Allzyme SSF added diets, which showed increased feed intake at low energy diet compared to HE and ME diets. Enzymes are expected to facilitate the breakdown of larger molecular structures of the feed ingredients into smaller ones by their specific action and making these nutrients readily available to the digestive system for better absorption. This is an attempt to help the chicken to extract more nutrients from feedstuffs, Ghazi *et al.*, (2003). The decrease in intake with decrease in energy content was for diets with no enzyme added, where LE (3995 g/bird) was lower ($P < 0.05$) than ME and HE, which were similar (4535 and 4455 g/bird respectively). Chicks fed Nutrase Xylla supplemented diets consumed equal amounts of feed irrespective of energy level, 4026, 3889 and 3668 g/bird for HE, ME and LE respectively. However there was gradual, though non-significant, decrease in feed intake with decrease in energy content among the birds fed these diets. Birds fed Allzyme SSF supplemented diets increased their feed intake with the decrease in energy levels (4332, 4112 and 4111 g/bird) for L.E, M.E and H.E respectively with LE being significantly higher ($P < 0.05$). The improved feed intake observed with bird fed low energy Allzyme was expected as the enzyme improves the performance of the bird at low energy added diet. Nutrase Xylla had increase feed intakes with decrease in energy level (4026, 3886 and 3669). The better feed intakes observed with birds fed Allzyme SSF over feed intakes of birds fed Nutrase Xylla might be due to multipurpose nature of Allzyme SSF (Alltch, Inc., Nicholasville, KY, the manufacturer), which make it's diet with improved nutritive value than Nutrase Xylla, which is a single purpose enzyme. This increase in feed consumption of bird on low energy diet has been observed and may be due to tendency of bird to eat more feed to compensate for low energy diet, Oluyemi, *et al.*, (2000). As such increase in intake for Allzyme supplemented birds with reduced energy can be two prong; effect of enzyme on digestion and effect of energy level. In contrast, the decrease in feed consumption of bird fed Nutrase Xylla

added diet could be due to the fact that Nutrase Xylla is a single purpose enzyme targeting one anti-nutritional factor and might have not completely hydrolysed or degraded large percentage of NSPs and oligo-saccharide components of the diet. There was no good explanation to results of feed intake values of birds fed non-supplemented diets as high and medium energy diets (4455 and 4534 g/bird respectively) were superior to those of birds fed supplemented diets, low energy non-supplemented bird had feed intake of 3995 g/ bird which was higher than feed intake of bird fed low energy diet supplemented with Nutrase Xylla (3668 g/bird), but not to LE feed intake (4332 g/bird) of bird fed low Allzyme SSF supplemented diet.

4.4.2 Effect of enzymes on feed intake at 22-49 days of age

Within LE diet added to Allzyme, enzyme supplementation significantly ($P < 0.05$) increased feed intake (4332 g/bird) compared to ME and HE diets which had 4112 and 4111 g/bird respectively. Feed intake was lowered with bird fed LE Nutrase Xylla supplemented diet (3669 g/bird), compared to that of HE and ME diets which numerically shows 4026 and 3889 g/birds respectively. There was gradual decrease in feed intake of birds fed Nutrase Xylla added diets with the decrease in energy level of the diet. The possible explanation to poor feed intake of birds fed diets added to Nutrase Xylla may be attributed to incomplete hydrolysis of NSPs of the diets. Feed intake of birds fed non-supplemented diets at ME and HE diets was higher than those of HE, ME and LE diets (Nutrase Xylla) except LE diet feed intake of bird fed Allzyme added diet, which was significantly ($P < 0.05$) different to feed intake of birds fed LE diets both fed non-supplemented and Nutrase Xylla diets. During finisher phase, the enzyme tended to decrease feed intake except at low energy Allzyme added diet. However, feed intake during finisher phase was not affected ($P < 0.05$) with enzyme supplementation.

Mean feed intake during finisher phase shows that feed intake did not improve with supplementation with Allzyme SSF and Nutrase Xylla. There was significant ($P < 0.05$) difference between the non supplemented (4328 g/bird) and Nutrase Xylla (3861 g/bird) supplemented diets, also there was a small significant difference of non-supplemented diet to Allzyme SSF which had a mean of 4185 g/bird). The results are in agreement with findings of Ranade and Rajmane (1992) who reported a lower feed intake of broilers fed reduced energy supplemented diets with enzyme preparation containing cellulose, protease, xylanase beta-glucanase and alpha amylase activities. Also, Meng, *et al.*; (2004) observed that broilers fed diets supplemented with amylase, lipase and protease generally consumed lesser amount of feed compared to that of control diet. However, feed intake of birds was independence of different energy levels of the experimental diets and their enzymes supplementation.

There was significant ($P < 0.05$) interactions between low energy diet and Allzyme SSF which was added to that diet. The increased feed intake of bird fed Allzyme SSF low energy diet, compared to Nutrase and Non-supplemented diets, is expected as this enzyme has a power of multiple enzymes which degrade the NSPs into soluble metabolizable products and thus increased energy intake of the bird. It was also noted that there was no significant interaction between energy levels and Nutrase Xylla diets, except for high energy diet which tended to have great improvement in feed intake than low energy non-added diet.

The effects of energy density and enzymes inclusion on the body weight of broiler chickens fed during 22-49 days are shown in Table 13.

Table 13: Effect of energy density and enzyme inclusion on the weight gain (g/bird) fed as from 22-49 days

Energy level	No enzyme	Nutrased Xylla	Allzyme ® SSF	Mean	SEM
High	1863 ^{xa}	1749 ^{ya}	1566 ^{zb}	1726 ^a	
Medium	1716 ^{xb}	1667 ^{xb}	1624 ^{xb}	1669 ^a	118.7
Low	1904 ^{xc}	1622 ^{zc}	1748 ^{ya}	1758 ^a	
Mean	1828 ^x	1679 ^x	1646 ^y	1718 ^x	
SEM		68.6			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different.;

^{abc} Mean with different superscripts within the columns are significantly ($P < 0.05$) different.

SEM = Standard error of means.

4.4.3 Effect of energy on body weight of birds at 22-49 days old

The body weight gains of broiler chickens fed during 22-49 days Table 13 showed that the overall body weight gain of birds fed diets of varying energy density tended to increase with reduction in energy diet density (1758 for LE versus 1669 and 1726 g/bird for ME and HE respectively). This overall trend was observed with birds fed non-supplemented diets. In this group, the highest gain was recorded for LE (1904) followed by HE (1863) with ME (1716g/bird) having the least gain. Nutrased Xylla supplemented diets had average body weight gains of 1749, 1667 and 1622 g/bird for HE, ME and LE diets. The body weight gain decreased with decrease in energy level of the diet. The decrease in body weight gain with the reduction of

level of energy in the diet of broiler is a result of reduced energy content of the ration. This is an indication that the expected increase in energy released from the NSPs by the enzyme in these low energy diets did not materialize. The lower body weight gain for the different energy levels compared with the control might have been due to low feed intake, or could be due to the decreased nutrient digestibility in the diets added to Nutrase Xylla. Results of these findings are accordance with the findings of Jacob *et al.* (2000) and Banerjee, (1992), who reported that Nutrase Xylla supplementation to broiler diet, was particularly useful for diets containing cereals with high non-starch polysaccharide (NSP) level, such as wheat, barley and oats. Probably this might be due to the use of specific enzyme (Nutrase Xylla or Xylanase) for wheat used in broiler diets, whereas, various feed ingredients having different kind of NSPs used in this experiment, did not had any effect from enzyme (Nutrase Xylla) which did not yield any beneficial response on utilization of NSPs in broilers.

4.4.4 Effect of enzymes on body weight

The effect of enzymes on body weight of broiler chickens were increased with reduction of energy in Allzyme SSF added diets (1566, 1624 and 1748 g/bird) for HE, ME and LE diets respectively. This was expected as the enzymes were to improve the performance of the bird at low energy diets. Although the non-supplemented diets resulted into numerically high body weight compared to those supplemented to enzymes, addition of a mixture of enzymes (Allzyme SSF) considering the composition of NSPs in a given diet may yield better response compared to supplementation of a single enzyme xylanase (Nutrase Xylla). Although the weight gain of non-supplemented diet was higher than enzyme added ones, the findings suggest the role of Allzyme SSF enzyme in degrading different kinds of NSPs present in the basal diets fed to birds during

this feeding phase resulted to significant weight gain over birds fed Nutrase Xylla supplemented diets. The result indicates that the body weight of bird fed added Allzyme SSF low energy diet was greater than that of high energy diet. This agrees with the result obtained by Schutte and Sundu *et al.*, (2005) and Chesson (2001). Stated authors however, emphasized that with application of enzyme on low energy diet, positive results are achieved, especially in starter young chickens, where as in older categories these positive effects are less expressed. Contrary to these result, Peric *et al.*, (2002) did not record similar result on effect of enzymes addition in mixtures with lower level of energy and protein. Also the result is inconsistent with findings of McCracken and Quintin, (2000) reported that there was no significant effect of enzyme addition on the measured AME content of broiler diet. Similarly, Ravindran *et al.* (2001) reported a significant linear effect on weight gain by feeding broilers with microbial phytase enzyme in broilers' diet. The effect of Nutrase Xylla on body weight gain of birds was not efficient at low energy diet which recorded 1622 (g) per bird, while with birds fed HE and ME diets (1749 and 1667 g/bird), the effect of Nutrase Xylla on weight gain was numerically higher than HE and ME (1566 and 1624 g/bird) added to Allzyme. It was observed that among enzyme treated groups, there was decrease in weight gain with level of reduction in energy density of the diets. Though high and medium energy non-supplemented diets did not differ significant from enzyme added groups, low energy Allzyme diet recorded higher weight gain than Nutrase Xylla groups.

Mean total body weight indicated that birds fed non-supplemented diets gained significant ($P<0.05$) mean weight gain of 1827 g/bird compared to enzymes (Nutrase Xylla and Allzyme SSF) supplemented diets which had 1679 and 1646 g/bird respectively. This implies that body weight at finisher phase was significantly ($P<0.05$) depressed with enzymes supplementation of either Nutrase Xylla or Allzyme SSF which did not had significant difference. This could be attributable to residual anti-nutritional factors and reduced feed intake, which have been reported

to have negative effects on weight of bird, Choct, and Annison (1992b). There was no significant interactions ($P < 0.05$) between energy level and the enzyme addition as the overall total mean body weight gain of non-supplemented diet was significantly higher than those of enzymes added diets. Among the enzymes added diets, there was a significant interaction between low energy diet and Allzyme existed for body weight gain in finisher during this feeding phase which resulted to greater body weight. This may be due to high feed intake of the bird and improved digestibility of nutrients in gastrointestinal system of the bird.

Effects of energy density and enzymes inclusion in broiler finisher diets fed during 22-49 days are shown in Table 14.

Table 14: Effect of energy density and enzyme inclusion in broiler diets on Feed Conversion Ratio (FCR) from 22 to 49 days of age

Energy level	No enzyme	Nutrase Xylla	Allzyme ® SSF	Mean	SEM
High	2.2 ^{ax}	2.0 ^{ay}	2.8 ^{az}	2.3 ^a	
Medium	2.3 ^{ax}	2.1 ^{ay}	2.4 ^{bz}	2.5 ^b	0.29
Low	1.7 ^{bx}	2.2 ^{by}	2.2 ^{cy}	2.3 ^c	
Mean	2.0 ^y	2.0 ^y	2.4 ^x	2.3 ^x	
SEM		0.17			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different;

^{abc} Mean with different superscripts within the columns are significantly ($P < 0.05$) different.

SEM = Standard error of means.

4.5.1 Effect of energy and enzymes inclusion on the feed conversion ratio of bird during

22-49 days

The overall means effect of energy density and inclusion of enzymes on feed conversion ratio during finisher feeding phase is inconsistent as HE and LE means of FCR per bird are the same (2.3). Medium energy fed birds tended to have higher FCR of (2.5). This explains that both HE and LE diets had better feed to gain ratio. The trend was observed with birds fed no enzyme diets

which showed higher FCR in bird fed ME, while HE and LE fed birds had 2.2 and 1.7 respectively. LE fed birds had the best feed conversion ratio (FCR), which is attributable to better feed utilization. This could be explained, that the birds fed low energy diets had efficiently used the variability in nutrient density in the formulated diets. On the other hand, there seemed to be consistency of FCR with enzymes supplementation. Nutrase Xylla improved FCR at all energy levels (HE, ME and LE), thus had improved FCR among other two diets (Allzyme SSF and No enzymes added diets) except in low energy non-supplemented bird which seemed to have the best FCR among all birds fed different diets with different energy density. Also Allzyme added diets show increased of FCR value with decreased energy level of the diets (2.8, 2.4 and 2.2). Among the two enzymes, Nutrase Xylla seemed to have better feed to gain ratio than Allzyme fed birds which did not utilized the feed efficiently. Literature reports that addition of enzymes to poultry diets has been shown to improve weight gain, feed to gain ratio, apparent nutrient digestibility and the AME of the diet, Marquardt, (1996). However, enzyme addition in the present study did not affect FCR of the birds, and was found to be non-significantly affected among the supplemented diets. The results are not in line with findings of Espino *et al.* (2000) who reported a significant improvement on feed efficiency of broilers with microbial protease, lipase and amylase fed diets after 40 days of feeding.

Mean total FCR showed lowered FCR of 2 for both non-supplemented and Nutrase Xylla added diets, while was 2.4 for Allzyme added diet. This was explained that feed to gain ratio was best with birds fed no enzyme and Nutrase Xylla diets.

The interaction between enzymes and energy level was not significant. Although it seemed there is an interaction between energy level and inclusion of Allzyme SSF as feed conversion ratio decrease with decrease in energy level.

4.6 Birds performance during overall phase (0-49 days)

Results of effects of exogenous enzymes and energy levels on broiler chicken performance during the overall phase of feeding growth are shown in tables 15, 16 and 17.

The effects of energy density and enzymes inclusion on feed intake of birds fed starter and finisher phase (entire feeding phase), are presented in Table 15.

Table 15: Effect of energy level and enzyme on feed consumption (g) per broiler chickens fed from 0-49 days old.

Energy level	No enzyme	Nutrase Xylla	Allzyme ® SSF	Mean	SEM
High	5505 ^{xa}	4959 ^{za}	5044 ^{yb}	5169 ^x	
Medium	5561 ^{xa}	4759 ^{zb}	4978 ^{yc}	5099 ^x	206.4
Low	4811 ^{yb}	4886 ^{ya}	5238 ^{xa}	4978 ^x	
Mean	5292.3 ^x	4868 ^z	5086.7 ^y	5082 ^x	
SEM		119.1			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different;

^{abc} Means with different superscripts within the columns are significantly ($P < 0.05$) different.

SEM = standard error of means.

4.6.1 Effect of energy on feed intake of broiler chickens during overall feeding phase

Over the entire feeding phase of 1-49 days of feeding broiler chickens, with non-starch polysaccharide degrading enzymes (Allzyme SSF and Nutrase Xylla), overall feed intake was observed to decrease with decrease in energy levels of the diets. The feed intake was 5169, 5099 and 4978 g/bird for HE, ME and LE diets respectively, this was not expected from birds, as the enzymes were expected to increase feed intakes of birds fed supplemented diets. The increased feed intake with increasing levels of energy in the diets during overall feeding phase might be due to the critical need of the birds for energy. Results on feed intake of broiler chickens fed Allzyme SSF added diets during overall phase, showed that feed intake significantly increased at low energy diet (5238 g/bird) compared to feed intake of birds fed HE and LE diets (5044 and 4978 g/bird respectively). This difference in feed intake may be attributed to the improvement in value of Allzyme SSF enzyme which might have hydrolysed high crude fibre in low energy diets during finisher phase. The same trend was seen with bird fed Nutrase Xylla supplemented diet which have high feed intake (4886 g/bird) at low energy diet compared to ME diet (4759 g/bird) but not higher than HE diet which have feed intake of (4959 g/bird). The results indicated that reduction of energy level of the diets added to enzymes increased the feed consumption of broiler during this feeding period. Among the two added enzymes, Allzyme SSF showed the best feed intake compared to Nutrase Xylla. Results on feed intake of birds fed non-supplemented diets showed that bird fed ME diet consumed more (5561 g/bird) feed than bird fed HE diet which had 5505 g/bird) which both appeared to be significantly different ($P < 0.05$) compared to feed intake of chick fed LE diet (4811 g/bird). Final feed intake means of different feed types with their different energy levels indicated that non-supplemented diet had more feed intake compared to enzymes added diets with FI values of (5292, 4868 and 5086 g/bird) respectively.

Results of this study suggested that enzymes supplementation to broiler chickens diet did had significant effect. Comparing the mean feed intake of the added enzymes, Allzyme bird had high feed intake than Nutrase Xylla bird. There was interaction between the energy level of the diet and the enzymes added.

Effect of energy level and enzymes inclusion on body weight per broiler chicken fed from overall feeding phase is shown in Table 16.

Table 16: Effect of energy level and enzyme on weight gain (g) per bird fed from (0-49 days) old.

Energy level	No enzyme	Nutrase Xylla	Allzyme ® SSF	Mean	SEM
High	2295 ^{xa}	2199 ^{xa}	1974 ^{zc}	2156 ^a	
Medium	2172 ^{xb}	2111 ^{yb}	2025 ^{zb}	2102 ^a	107.8
Low	2351 ^{xa}	2032 ^{zc}	2196 ^{ya}	2193 ^a	
Mean	2273 ^x	2114 ^y	2065 ^y	2151 ^x	
SEM		62.2			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different;
^{abc} Mean with different superscripts within the columns are significantly ($P < 0.05$) different.
SEM = Standard error of means.

4.6.2 Effect of energy on body weight gain during overall feeding phase

Results on effect of energy levels and enzymes on body weight gain (g) per bird during overall feeding phase show that, body weight gain increased significantly ($P < 0.05$) with decrease in energy level of the diet, with values of (2193, 2102 and 2156 g/bird) for LE, ME and HE diets respectively. This shows that addition of NSP exogenous enzymes Allzyme SSF and Nutrase Xylla did not have a clear effect on growth performance under the conditions tested in this study (Table, 16). This trend was also noted with birds fed Allzyme SSF added diets which showed increased body weight gain with decreased energy level of the diet (1974, 2025, 2196 g/bird) for HE, ME and LE diets. Allzyme SSF improved weight gain at low energy diet compared to Nutrase Xylla. The result suggested that Allzyme SSF performed better at LE diet and improved body weight gain compared to Nutrase Xylla (2032 g/bird). On the other hand, birds fed Nutrase Xylla diets, had decreased body weight gain with decreased energy density of the diets (2199, 2111, 2032 g/bird) for HE, ME and LE diets respectively. This indicates that weight gain of the bird fed Allzyme SSF supplemented diet was significantly ($P < 0.05$) different compared to that of birds fed Nutrase Xylla added diets. The reduction in weight gain of bird fed Nutrase Xylla diet could be due to incomplete hydrolysis of non-starch polysaccharides fractions in the diets, and thus had less weight gains than Allzyme SSF fed birds.

Effect of energy level and enzymes inclusion on feed conversion ratio of broiler finisher fed during overall feeding phase is shown in Table 17

Table 17: Effect of energy level and enzyme on feed conversion ratio of broiler chicken fed from 0 - 49 days of age

Energy level	No enzyme	Nutrase Xylla	Allzyme ® SSF	Mean	SEM
High	2.1 ^{zb}	2.0 ^{xa}	2.8 ^{zc}	2.3 ^b	
Medium	2.3 ^{yc}	2.1 ^{xb}	2.4 ^{yb}	2.5 ^c	0.32
Low	1.7 ^{xa}	2.2 ^{zc}	2.1 ^{xa}	2.0 ^a	
Mean	2.1 ^x	2.1 ^x	2.4 ^y	2.3	
SEM		0.18			

^{xyz} Means with different superscript within the rows are significantly ($P < 0.05$) different;
^{abc} Mean with different superscripts within the columns are significantly ($P < 0.05$) different.
SEM = Standard error of means.

4.6.3 Effect of energy and enzymes inclusion on feed conversion ratio during overall feeding phase

Results on feed conversion ratio during the overall phase of feeding broiler chicken are shown in Table 17. Overall, the mean feed conversion ratio (FCR) for the low energy diets was 2, while it increased within ME and HE diets which had feed conversion ratios of 2.5 and 2.3 respectively. A similar trend was observed with birds fed Allzyme SSF diets which showed increased FCR

with increased energy level (2.8, 2.4 and 2.1) for HE, ME and LE diets respectively. This shows that bird fed low energy diet had better FCR than HE and ME diets which had poor feed to gain ratios. The Feed to gain ratios of broiler chickens fed Nutrase Xylla supplemented diets showed that FCR improved with increased energy level of the diet (2, 2.1 and 2.2 per bird) fed HE, ME and LE diets respectively. The effect of Nutrase Xylla supplemented diet on feed to gain ratio of 2.0 was comparable ($p < 0.05$) to the feed to gain ratio of 2.8 obtained on bird fed Allzyme SSF on high energy diets. There was significant difference between the effects of the two enzymes on feed-to-gain ratio although Nutrase Xylla seems to have better effect than Allzyme SSF. Non-supplemented diets show inconsistent results as FCR was obtained to 1.7 with bird fed low energy diet, and (2.3 and 2.1) for ME and HE diets. The best FCR during this study was noted with bird fed low energy diet without enzyme supplementation. It was suggested that over the entire trial period, the enzyme supplement tended to decrease FCR, and thus had no significant effects on feed conversion ratio. Overall mean FCR was noted with 2.3, while Nutrase Xylla and No enzyme addition showed the best FCR of 2.1 and Allzyme SSF had the declined feed conversion ratio of 2.4 among all diets fed during this phase.

4.7 Cost benefits analysis of inclusion of feed enzymes in broiler diets

The results of cost benefit analysis of adding feed enzymes to broilers diets are shown in

Table 18.

Table 18: Cost benefit analysis of inclusion of enzymes in broilers diets

Diets	Diet containing		
	No enzyme	Nutrased Xylla	Allzyme SSF
Feed intake (kg/bird) [*]	5.3	4.9	5.1
Price of feed (Ksh/kg)	27	28	28
Body weight gain/bird ^{**}	2.3	2.1	2.6
Sale of bird @ 170 ^{***}	391	357	442
Cost of feed Ksh/ bird ^{****}	143.1	137.2	142.8
Return Ksh /diet ^a	247.9	219.8	299.2
Cost benefit Ksh/ diet ^b	0	-28.1	51.3
Gross energy	3.57	3.57	3.57

^{*}amount of feed consumed over the entire feeding period

^{**} Lwt (kg) at end of feeding period-the birds were sold on Lwt basis

^{***} Return per bird at Ksh 170 per kg lwt

^{****}Cost of feed consumed only

^a Earning per bird when cost of feed removed

^bCompare advantage of enzyme addition in relation to control

Results of cost benefits analysis from addition of enzymes in broiler feed are shown in Table 21.

Nutrased Xylla and Allzyme were priced at Kshs 1000/kg with an inclusion rate of 100 gm/ton raising the cost of feed by Kshs 1/kg compared to control. Ojewola *et al.* (2000), argued that the

relative advantage of using any feed enzyme is determined by its price at the time of use and the current price of the product (live or dressed chickens).

Birds fed the Allzyme supplemented diet gave a better return (Kshs 51.3/bird) compared to the control while in the case of Nutrase supplemented diets, there was a net loss (Kshs 28.1/bird). The loss for Nutrase Xylla fed birds, compared to control, was due to low body weight gain, Table 15 and low feed conversion ratio, Table 16 during overall feeding phase.

The higher return of Allzyme supplemented diet was attributed to better feed conversion rate (Table 16) resulting in higher body weight gain, Table 15, which resulted from better dietary nutrients utilization. Results of this study agree with findings of Kadam *et al.*, (1991) who reported increased profit by 16.83% in groups fed on 0.1 g/kg Roxazyme enzyme-G, which is similar to Allzyme containing glucanase, xylanase and cellulose and some traces of amylase, hemicellulases, pectinases and proteases, supplemented commercial broiler feed than control. The better returns from broilers fed control diet (D1) over Nutrase Xylla diet is difficult to explain, but was a result of better body weight gain, Table 15 of birds fed control diet. The better performance of Allzyme over Nutrase is due to its multiple enzyme content (β -glucanases, hemicellulases, amylases, proteases, lipases and pectinases) targeting different anti-nutritional factors (NSPs) of the feed ingredients broiler diets, while Nutrase Xylla is a single enzyme (xylanase only) targeting a single NSP in the diet. Bedford, (1996) and Marsmen, (1997) reported that multi enzymes support nutrients availability better than single enzymes. It is suggested that the variation in the performance of broilers on addition of enzymes to both diets (D4 and D7) might be due to variations in the concentration and composition of NSP in the diets and/or activity of the added enzyme. Nutrase Xylla did not perform better, which would indicate that the level of soluble NSPs fractions (xylose, arabinose and mannose) was low in both broiler starter and finisher diets, Table 8. This might be the cause of depressed body weight observed in

broiler fed Nutrase Xylla added diet, Table 16, and thus affected cost return of the bird compared to control fed bird.

CHAPTER FIVE: CONCLUSIONS

The effect of supplementing broiler diets with two exogenous enzymes was studied. This was based on the hypothesis that addition of exogenous enzymes would improve the utilization of non-starch polysaccharides (NSPs). The following were the conclusions from this study.

1. The broiler diets formulated using the common raw materials found in Kenya were relatively low in arabinoxylans which range from 6.96 to 9.16 mg/g in the starter and finisher diets.
2. Supplementing the diets with Nutrase Xylla had no beneficial effect on the performance of the birds. However, Allzyme SSF improved weight gain in birds fed on medium and low energy diets, implying that this enzyme could be beneficial in low energy high fibre diets.
3. It is also likely that this enzyme not only improved utilization of NSPs but also made minerals such as phosphorus and calcium available due to presence of phytase in the enzyme complex.
4. Use of Allzyme SSF improved the returns per bird by Kshs 51.30, while use of Nutrase Xylla resulted in loss of money.
5. The choice of a feed enzyme to use in broiler feeds should be based on the raw materials used to compound the feed.

CHAPTER SIX: RECOMMENDATIONS

1. From this study, it is recommended that a more detailed investigation be carried out to determine the types of NSPs in the Kenyan feed ingredients.
2. With a better understanding of the NSPS, an appropriate enzyme blend can be identified to improve the utilization of the fibrous feeds ingredients common in Kenyan poultry feeds.

CHAPTER SEVEN: LITERATURE CITED

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CHAPTER EIGHT: APPENDICE

APPENDIX 1: ANALYSIS OF VARIANCE FOR BROILER CHICKS BODY WEIGHT GAIN (1-21 DAYS)

SOURCE OF VARIATION	d.f	s.s	m.s	V.r.	F.Pr.
Energy	2	672	336	0.09	0.915
Enzyme	2	2856	1428	0.36	0.687
Energy vs Enzyme	4	11774	2943	0.78	0.546
Residual	27	101477	3758		
Total	35	11677			

Energy, feed types (enzymes), and energy versus enzyme interaction were not significant

**APPENDIX 2: ANALYSIS OF VARIANCE FOR BROILER FINISHER BODY WEIGHT
GAIN (22-49 DAYS).**

SOURCE OF VARIATION	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	48097	24049	0.85	0.437
Enzyme	2	224094	112047	3.97	0.031
Energy vs Enzyme	4	131281	32820	1.16	0.349
Residual	27	761331	28197		
Total	35	1164803			

Enzyme (feed types) was significant, while energy and enzymes interaction were no significant different.

APPENDIX 3: ANALYSIS OF VARIANCE FOR BROILER CHICKENS OVERALL

BODY WEIGHT GAIN (0-49 DAYS).

SOURCE OF VARIATION	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	49314	24657	0.54	0.594
Enzyme	2	283344	141672	3.05	0.064
Energy vs Enzyme	4	181709	45427	0.98	0.436
Residual	27	1254032	46446		
Total	35	1768399			

Energy was not significantly different while feed enzymes (Nutrase and Allzyme SSF) were significantly different.

APPENDIX 4: ANALYSIS OF VARIANCE FOR BROILER STARTER FEED INTAKE DURING (1-21 DAYS) FEEDING PHASE.

SOURCE OF VARIATION	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	89387	44693	0.85	<0.01
Enzyme	2	48668	24334	5.16	0.013
Energy vs enzyme	4	74572	18643	3.95	0.012
Residual	27	127294	4715		
Total	35	339920			

Energy was highly significant different at all levels, while feed types (enzyme), interaction between energy and feed enzyme was observed to be significantly different.

APPENDIX 5: ANALYSIS OF VARIANCE FOR BROILER FINISHER FEED INTAKE (22-49 DAYS)

SOURCE OF VARIATION	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	289739	144869	0.85	0.437
Enzyme	2	1375310	687655	4.05	0.029
Energy vs Enzyme	4	779549	194887	1.15	0.356
Residual	27	4587600	169911		
Total	35	7032197			

Enzyme (feed types) was significant, while energy levels, energy versus enzyme interaction was not significantly different.

**APPENDIX 6: ANALYSIS OF VARIANCE FOR BROILER CHICKENS FEED INTAKE
DURING OVERALL PHASE (0-49 DAYS)**

SOURCE OF VARIATION	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	224020	112010	0.66	0.526
Enzyme	2	1081498	540749	3.17	0.058
Energy vs Enzyme	4	1400602	350150	2.06	0.115
Residual	27	4599100	170337		
Total	35	7305219			

Enzyme (feed types) show significant, while energy, energy versus enzymes interaction were not significant.

**APPENDIX 7: ANALYSIS OF VARIANCE FOR BROILER STARTER FEED
CONVERSION RATIO DURING 1-21 DAYS OF AGE**

Source of Variation	d.f	s.s	m.s.	v.r.	F.Pr.
Energy	2	0.5809	0.2904	1.62	0.217
Enzyme	2	0.1661	0.0831	0.46	0.635
Enzyme versus energy	4	0.4841	0.1210	0.67	0.616
Residual	27	4.8522	0.1297		
Total	35	6.0833			

Energy was significant different, while both enzymes and energy enzymes interaction were not significant.

**APPENDIX 8: ANALYSIS OF VARIANT FOR BROILER FINISHER FEED
CONVERSION RATIO (FCR) FED DURING 22-49 DAYS OF AGE.**

Source of variation	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	0.3604	0.1802	1.02	0.375
Enzyme	2	0.5573	0.2786	1.57	0.226
Energy versus Enzyme	4	0.3689	0.0922	0.52	0.721
Residual	27	4.7834	0.1772		
Total	35	6.0699			

Both energy and enzyme (feed types) were significantly different, while energy versus enzymes interaction were not significantly different.

**APPENDIX 9: ANALYSIS OF VARIANCE FOR BROILER FINISHER FEED
CONVERSION RATIO (FCR) FED DURING 0-49 DAYS.**

Source of variation	d.f.	s.s.	m.s.	v.r.	F. Pr.
Energy	2	0.6455	0.3227	0.80	0.458
Enzyme	2	1.0575	0.528	1.32	0.284
Energy versus Enzyme	4	1.0203	0.2551	0.61	0.64
Residual	27	10.8336	0.4012		
Total	35	13.5568			

Both energy and enzymes inclusion were significant, while enzymes versus energy interaction was not significant.

APPENDIX 10: COST OF FEED INGREDIENTS (KSH) AT THE**TIME OF THE EXPERIMENT**

<u>Ingredient</u>	<u>Amount (kg)</u>	<u>Kshs/kg</u>	<u>Total cost (Kshs)</u>
Maize	1150	20	23000
Pollard	560	13	7280
Vegetable oil	12	280	3360
Wheat bran	120	6	720
Soya bean meal	270	51	13770
Omena	170	65	11050
Lime stone	42	3	126
Vitamin mineral premix	6.5	270	1755
DCP	3.5	300	1050
Coccidiostat	1	495	495
Comon salt	13	7	91
Enzymes	1.2	1000	1200
Total cost	2337.5		63,897

Cost of feed ingredients as sold to the Author at the time of the experiment.

The cost of feed ingredients in Kenya shillings for the whole experiment was sixty three thousand eight hundred and ninety seven shillings (63,897 Kshs).

Estimated cost of one kilogram of feed was 27.33 Kshs for non-supplemented diets, and at 32 Kshs for diets supplemented with enzymes (Nutrase Xylla and Allzyme SSF).