The Effects of Canola (*Brassica napus*) and Juncea (*Brassica juncea*) Meals in Diets on Broilers and Turkeys

by

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ABSTRACT

Canola and juncea meals can be used in poultry diets instead of soybean meal. The meals were fed to broilers and turkeys in digestibility trials to measure the apparent metabolizable energy (AME_n). Throughout growth trials, canola and juncea meals were fed at four levels (0, 10, 20 and 30 %) with and without a dietary enzyme cocktail (DEC). This study investigated higher inclusion levels than previously recommended for use in broilers diets. The carcass compositions, fatty acid analysis and liver parameters were measured to investigate the effects of diets on birds. The AME_n of the meals were used in formulating broiler and turkey diets. Growth trials found positive effects of DEC on performance, allowed higher inclusion levels of the meals. No negative effects of diets were observed for liver parameters and carcass compositions. The fatty acid profiles of carcass tissues were improved by increasing levels of canola oil in diets.

LIST OF ABBREVIATIONS USED

Acid detergent fiber…….	ADF
Acid insoluble ash……...	AIA
Alpha linolenic acid…….	ALA
Analysis of variance……	ANOVA
Apparent metabolisable energy(nitrogen corrected)……...	AME_n
Body weight gain…….	BWG
Body weight…….	BW
Canola Council of Canada……..	CCC
Canola meal…….	CM
Crude fat……..	CF
Crude protein……...	CP
Docosahexaenoic acid…….	DHA
Dry matter……	DM
Eicosapentaenoic acid……..	EPA
Fatty acid…….	FA
Fatty liver haemorrhagic syndrome…….	FLHS
Feed consumption…….....	FC
Feed conversion ratio……...	FCR
Gross energy……	GE
Juncea meal……..	JM
Linoleic acid……	LA
Metabolizable energy……..	ME
Mono unsaturated fatty acid……	MUFA
National research council…….....	NRC
Neutral detergent fiber……	NDF
Nitrogen corrected apparent metabolizable energy……	AME_n
Nitrogen……..	N
Non-starch polysacccharides……..	NSP
Polyunsaturated fatty acids…….	PUFA

Protein efficiency ratio……	PER
Rape seed meal……	RSM
Saturated fatty acids…….	SFA
Soybean meal……..	SBM
Thyroxin…….	T4
Triiodothyronine…….	T3
True metabolizable energy……..	TME
Trimethylsilyl…….	TMS
Volatile fatty acid…….....	VFA

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CHAPTER 1. INTRODUCTION

1.1. Introduction

Canola (*Brassica napus*) and juncea (*Brassica juncea*) are both increasing in production in western Canada for edible oil (Aider and Barbana 2011). Canola meal (CM) and juncea meal (JM) by-products of seed crushing for the extraction of oil, are good sources of protein in poultry diets with 36% of minimum protein Canola council of Canada (CCC 2009). CM is a good source of essential minerals (Bell et al. 1999).

Current recommendations for inclusion of canola and juncea meals in broiler diets are based on old studies using old varieties of canola seed containing higher levels of glucosinolates. Glocosinolates showed some negative effects on broiler growth performances (Khajali and Slominski 2012), haemorrhagic lesions in broiler livers (Yamashiro et al. 1977) and hypothyroidism in poultry (Schöne et al. 1993). Reduced glucosinolate levels in current oilseed varieties might allow higher inclusions of the meals. The other factor that limits the use of canola in larger quantity in broiler diets is high levels of dietary fiber as birds cannot utilize fiber efficiently (Newkirk et al. 1997). Many of the previous studies completed used broilers that are different from today's commercial birds for meat production. Modern, broilers have been improved genetically to produce more meat in a shorter time. Therefore, they may have different capabilities to use CM and JM.

Digestibility of nutrients in new varieties of modern CM and JM need to be re-evaluated due to the changes in both meal and bird characteristics.

Superzyme-OM ^{TM} is an enzyme which was designed for diets containing high levels of flax or canola. The combinations of enzymes in Superzyme-OM^{TM} reflect, the nature of carbohydrates found in CM. These enzymes improved the metabolizable energy of CM and growth performance (Woyengo et al. 2010 and Jia et al. 2012) in broilers.

With more research, CM and JM, which are more available in western Canada than soybean meal, could be effective alternatives as a protein source combined with Superzyme^{TM} - OM in broiler diets.

1.2. Objectives

The study will determine the AME_n content of a current commercial source of CM and JM for broilers and turkeys. This study will evaluate different levels of CM and JM (0, 10, 20 and 30%), in diets for broilers. Growth performance, liver characteristics, carcass composition and tissue fatty acid content were studied. This study evaluated the inclusion of a multicarbohydrase enzyme (Superzyme-OM^{TM}) in diets for broiler chickens.

1.3. Hypotheses

The AME_n in CM and JM will be higher for turkeys and broilers than represented in NRC (1994). AME_n content of JM and CM will be higher in turkeys compared to broilers.

Similar growth performance of broilers fed graded levels (0, 10, 20 and 30 %) of CM and JM are expected. Dietary enzyme will improve growth performance of broilers fed CM and JM. Increased tissue levels of desirable n-3 fatty acids will result from higher dietary inclusion of CM or JM. No significant changes in carcass characteristics and liver performance are expected in broilers fed graded levels of CM or JM with and without dietary enzyme addition.

CHAPTER 2. LITERATURE REVIEW

2.1. Canola meal and juncea meal

Canola (Canadian oil, low erucic acid) is rapeseed (*Brassica napus*, Argentinan canola, or *Brassica campestris*/*rapa*, Polish canola), which were bred, to have low levels of erucic acid and glucosinolates, less than 2% and 30 μmol·g^{-1}, respectively, (CCC 2009). Juncea (*Brassica juncea*) is yellow seeded mustard, which is mainly grown in western Canada. As it has less than 2% erucic acid and less than 30 μmol g^{-1} glucosinolates, it can be recognized as a canola-quality seed. Juncea has been developed to meet the requirements of the canola industry since the first low erucic acid lines of *Brassica juncea* were developed in Australia (Kirk and Oram 1981). Production of canola and juncea are both increasing in western Canada for edible oil and are considered to be the second most available source of edible oil in the world (Aider and Barbana 2011). Comparing *Brassica napus* and *Brassica juncea* under western Canadian conditions, *Brassica juncea* experiences less damage from heat and drought stress, is more resistance to diseases and matures earlier than *Brassica napus* canola (Woods et al. 1991). Jiang et al (1999) showed a high negative relationship between protein and dietary fiber contents in meals derived from black- and yellow-seeded *Brassica napus* canola, as a result, development of yellow seed has been justified to improve the quality of CM. Recently, canola-quality of *Brassica juncea* have been developed in Canada with a pure yellow seed coat (Jia et al. 2012). Canola seed production is about 9 million tonnes per year. The industry is targeting an increase to 15 million tonnes per year. by 2015 (CCC 2009). Canola seed is 1-2 mm in diameter and contains approximately 43% oil and 20% crude protein (CCC 2009). Canola oil is a good source of alpha-linolenic acid (ALA) (Ajuyah et al. 1991). Canola oil's proponents claim that it is one of the most heart-healthy oils and has been reported to reduce cholesterol levels and lower serum triglyceride levels (Davis and Melina 2000). Juncea oil generally has a similar fatty acid profile to *Brassica napus* oil with very good stability of the fatty acid profile. *Brassica juncea* oil was found to be more unsaturated than canola oil of *B. napus* (Potts et al. 1999). Canola oil is also used

for producing biofuel for powering motor vehicles, which may increase the availability of canola meal (CM) for use in poultry feed (Thacker and Petri 2007). CM, a by-product of seed crushing for the extraction of canola oil, contains approximately 36% protein on a as fed basis (CCC 2009). CM is a good source of protein in poultry diets. Comparing CM as a protein source in poultry diets to soybean meal (SBM), the most common plant protein source in poultry diets, CM contains less protein (36 vs. 48 %), more crude fiber (12 vs. 3.5%) (Table 2.1) and less metabolizable energy (2,440 kcal·kg^{-1} vs. 2,230 kcal·kg^{-1}) National Research Council (NRC 1994).

Table. 2.1. Standard Specifications for Canadian Canola Meal and Soybean Meal

	Canola meal (%)[1]	Soybean meal (%)[2]
Protein, minimum	36	47.5-49.0
Fat, minimum	2	0.5
Fiber, maximum	12	3.3-3.5
Moisture, maximum	12	12

[1] Canola Council of Canada (CCC) 2009.
[2] Canadian International Grain Institute (CIGI) 2010.

Because of the good balance of essential amino acids, canola protein is of high quality with the highest protein efficiency ratio (PER = 3.29) of all plant-based proteins in feedstuffs (Sarwar et al. 1984). CM is also a good source of essential minerals such as potassium, sulphur, calcium and iron and especially a good source of selenium and phosphorus (Bell et al. 1999).

2.1.1. Anti-nutritive factors of canola meal

2.1.1.1. Glucosinolates

Glucosinolates are a large group of sulphur-containing secondary plant metabolites, which are present in all economically important varieties of *Brassica*. There are different classes of glucosinolates, but they all share a common structure comprised of a β-D-thioglucose group (Figure 2.1). Rapeseed meal (RSM) contains three major glucosinolates with higher concentrations of progoitrin or epiprogoitrin followed by

gluconapin and glucobrassicanapin. The content and distribution of glucosinolates differs among different *Brassica* species. Glucosinolate content in plants differs in different growing conditions due to dry and hot environments. With lack of water, the synthesis of amino acids and sugars, as precursors in gluscosinolate biosynthesis, increases (Tripathi and Mishra 2007).

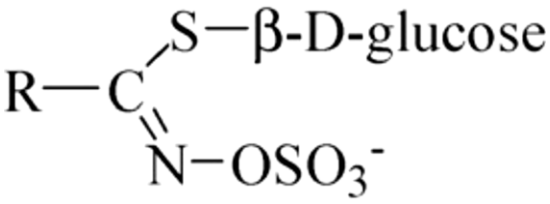

Figure. 2.1. General structure of glucosinolate. From Tripathi and Mishra (2007).

The ingestion of considerable amount of glucosinolates may have negative effects on animal health. The adverse effects are greater in monogastric animals compared to ruminants; also, young animals are more sensitive to glucosinolates than older animals (Tripathi and Mishra 2007). Glucosinolates are biologically inactive molecules, but the degradation products are biologically active. When seeds are reptured in the presence of moisture, glucosinolates are hydrolyzed by myrosinase to produce unstable aglucones which break down to produce a range of products including isothiocyanates, goitrin nitriles, and thiocyanates (Figure 2.2) that together inhibit the function of the thyroid gland and have negative effects on growth performance (Fenwick et al. 1982. McCurdy 1990).

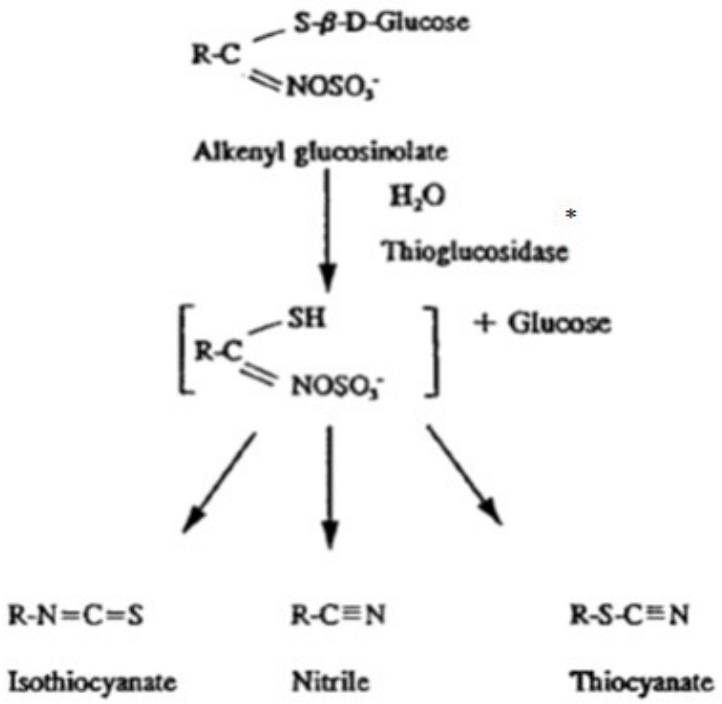

Figure. 2.2. Hydrolysis of glucosinolate. From Mithen (1992)
*Thioglucosidase: Myrosinase

Glucosinolates in plants are physically separated from the enzyme myrosinase (thioglucoside glucohydrolase). The contact of myrosinase and glucosinolates, by injury of plant tissues during processing, or digestion, causes the glucosinolates to be hydrolyzed by myrosinase in the plant and myrosinase from intestinal microflora, which releases breakdown products that have adverse effects on animals (Tripathi and Mishra 2007). Rather than the toxic effects of glucosinolate on animals, an increased incidence of leg problems with feeding diets containing high levels of glucosinolates has been shown in several studies. Summers et al. (1992) found that the sulphur level of glucosinolate can cause leg problems in broilers as sulphur interferes with calcium absorption (CCC 2009). So, it is useful to supplement the diet with extra calcium as that does not affect the feed intake (CCC 2009). RSM had 166 $\mu \cdot mol\ g^{-1}$of glucosinolates on dry oil-free meal basis during 1980's. Currently very low glucosinolate rapeseed varieties

contain less than 25 μ·mol g^{-1} (Tripathi and Mishra 2007). The glucosinolate in canola is mainly aliphatic and indolyl (85 and 15% of glucosinolates, respectively) (Newkirk et al.2003).

2.1.1.2. Sinapine

Sinapine, is the choline ester of sinapic acid. CM contains approximately 1% sinapine on dry matter basis and it may cause a fishy odor in eggs from some brown-shell layers. It is hydrolyzed to trimethylamine, which is deposited in the egg and produces a fishy odor (Butler et al. 1982). The bitterness of CM is due to the presence of sinapine, it makes the meal less palatable for animals, while no major negative effect of sinapine has been found in broiler performance. Qiao and Classen (2003) reported that the bitter taste of sinapine did not affect the feed intake and growth rate in broilers and, interestingly, they found improvement in metabolizable energy content and protein digestibility of diet by feeding purified sinapine extract to broilers.

2.1.1.3. Phytate

Phytic acid (myoinositol 1,2,3,4,5,6-hexakis dihydrogen phosphate) exists in the embryo of rapeseed. Phytic acid may form complexes as mixed salts (phytates) of Ca, Mg, Zn and K and reduces the availability of some minerals (Nwokolo and Bragg 1977). Phytates, are the main storage form of phosphorus in plant seeds (Greenwood 1990). Poultry are not able to hydrolyze the majority of phytate to release the bound phosphorus (Nahashon et al. 1994). There are some proteins bound to phytate which poultry cannot digest and can cause reduced amino acid availability (Nahashon et al. 1994). The total phosphorous in CM is about 1.22% with 0.53 % of it, as phytate bound (Bell 1993). Nwokolo and Bragg (1997) found Zn , Ca and Mg deficiency syndromes in chickens due to the phytates present in rapeseed. Based on Khajali and Slominski (2012), the total amount of phosphorus in CM is 1.02%, phytic acid content of CM is high. The proportion of phytate to the total phosphorous ranges from 36 to 70% (Khajali and Slominski

2012).Canola meal has a high level of phytate, it also has high phosphorous availability from non-phytate-phosphorous.

2.1.1.4. Tannin

Tannins can be divided into two fractions: the hydrolysable and condensed portions (Yapar and Clandinin 1972), Clandinin and Heard (1968) reported that RSM contains considerable amounts of tannin (3%). Tannin in RSM can have negative effects on growth and metabolizable energy of chickens (Vohra et al. 1966). Brown seeded rapeseed hulls usually contain more condensed tannin than yellow hulls (Theander et al. 1977). The higher amount of tannin in brown seeded varieties, gives the meal a dark color and may form complexes with protein and proteolytic enzyme in the gastrointestinal tract and, as a result, affect protein digestion (Khajali and Slominski 2012). Mansoori and Acamovic (2007) found tannic acid (water soluble tannin) was responsible for the negative effect on growth performance as it affected the amino acids in the diets, especially methionine, histidine and lysine. Poor digestibility of amino acid might be associated with an increased excretion of inactivated enzyme and glycoproteins of gastrointestinal mucosa (Mansoori and Acamovic 2007). However, because most of the tannins in canola cell walls are water insoluble, the anti-nutritional factor of tannin in CM would be small (Khajali and Slominski 2012). CM contains between 1.5- 3 % of tannins, with higher levels in brown seeds compared to yellow seeds (CCC 2009).

2.1.1.5. Erucic Acid

Rapeseed oil is different from other vegetable oils, because it contains substantial amounts of erucic acid, $C_{22:1n-9}$ monounsaturated fatty acid (MUFA). Most vegetable oils contain high amounts of 16-18 carbon chain fatty acids (C_{16} and C_{18}) (Harvey and Downey 1964). Studies showed that consuming higher erucic acid oil can cause some health risk to humans (Ecke et al. 1995). Diets with high amounts of erucic acid (more than 10%) showed some adverse effects on rats, such as, accumulation of erucic acid in tissue lipids, reduction in growth rate (Green and Innis 2000). These problems dictated

plant breeders to diminish the amount of erucic acid in rapeseed varieties. Vogtmann and Clandinin (1974), concluded that inclusion of up to 15% of low erucic acid rapeseed oils in the diets of broilers was nutritionally satisfactory.

2.1.1.6. Electrolyte balance

Inappropriate dietary ratios of minerals can affect the acid-base balance in the body. Mongin and Sauveur (1977) found dietary imbalance between (Na^+K^-Cl) affected growth and caused leg abnormalities in broilers. Optimal performance can be achieved with diets containing 250 meq kg $^{-1}$ of Na^+K^-Cl. An ideal electrolyte balance is necessary for good performance and livability in broilers. The electrolyte balance is lower in CM compared to SBM diets (307 vs. 504 meq·kg^{-1}) because of lower levels of potassium in CM (11.4 vs. 19.6 g kg^{-1}) (Khajali and Slominski 2012). As feed intake in broilers is positively correlated with cation-anion balance, the decrease in feed intake by broilers consuming CM may be related to cation and anion levels in the diet, increasing levels of dietary cations, will correct the problem. Adding extra calcium carbonate has had some negative effects on feed intake, it would likely be preferable to add potassium bicarbonate to the diet (CCC 2009). March (1984) showed an improvement in growth performance of broilers fed CM, by adding NaCl in the diet, because, CM is low in sodium. Another problem is associated with higher level of sulphur for CM compared to SBM (0.65 vs. 0.44%). Sulphur can cause leg abnormalities in broilers because it interferes with calcium absorption (Khajali and Slominski 2012). The problem can be solved by supplementing the diet with extra calcium.

2.1.1.7. Fiber

The term dietary fiber was first used to describe plant cell walls (Van Der Kamp 2004). Dietary fiber is defined as the edible parts of plants or analogous carbohydrates that are resistant to digestion and absorption in the small intestine, with partial or complete fermentation in the large intestine. It includes polysaccharides, oligosaccharides, lignin, and associated substances (Van Der Kamp 2004). Polysaccharides in feed stuffs include starch and non-starch polysaccharides (NSP). NSP covers different varieties of polysaccharide molecules. The classification of NSP can be

seen in Figure 2.3. Different feedstuffs have different amounts and structures of NSP (McNab and Boorman 2002). In soybean, the major NSP is pectic polysaccharides (Voragen et al. 2001), while in canola, β - (1-3,6)-linked galactose polymers are dominant (Siddiqui and Wood 1972).

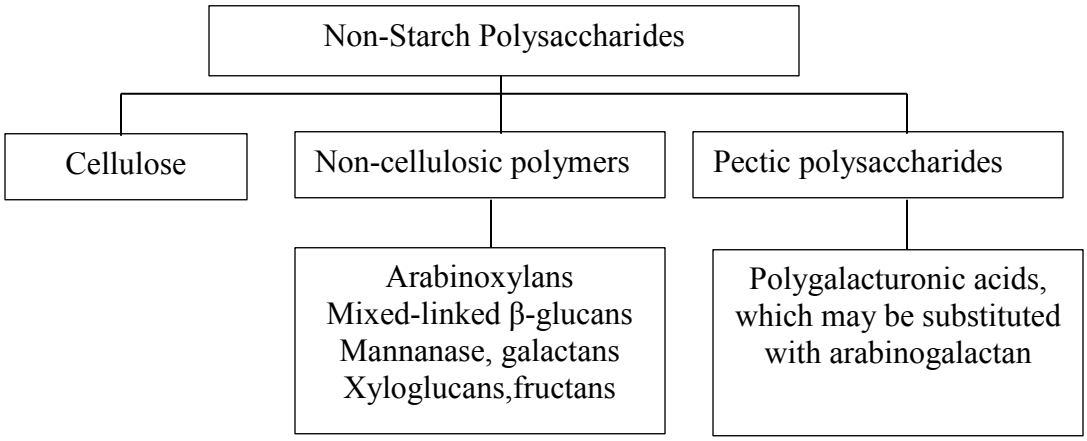

Figure. 2.3. Classification of non-starch polysaccharides. From McNab and Boorman (2002).

Dietary fiber is a significant part of all plant feedstuffs. High fiber level can be considered as the main restriction to formulate broiler diet with CM, as monogastrics cannot digest fiber. Fiber increases the passage rate of nutrients in the digestive tract and causes reduction in digestion in pigs (Imbeah and Sauer 1991). The carbohydrates in CM are protected by cell walls and it makes them less digestible (Bell 1993). Sixteen percent of the seed and 30% of the meal weight are hulls, which includes mainly fiber (Bell 1993). Stringam et al. (1974) found that yellow canola seed had lower percentage of hull than brown seeds (7.1 vs. 11.5 % of crude fiber). There are some ways to counter the negative effects of fiber in CM, like dehulling to improve the digestibility of protein or, plant breeding to breed low fiber and high protein cultivars. Dehulling, increases the glucosinolate content of the meal (Bell 1993). Hull contains a high level of non-starch

polycacccharides (NSP) and lignin, which may reduce digestibility and metabolizable energy. NSP with celulluse, β-glucans, arabinoxylans and pectin can increase the viscosity of digesta. NSP cannot be hydrolyzed by endogenous enzymes in birds and NSP may prevent access of endogenous enzymes to the nutrient content within grain cells. Some of the NSP in cereal cell walls dissolve in the digestive tract and increase viscosity (White et al. 1981). High viscosity interferes with enzymatic digestion of the nutrients and their subsequent absorption. Viscous materials can interfere with digestible enzyme and reduce the availability of nutrients to the animal (Antoniou et al. 1981). Furthermore, NSP can cause fermentation in the small intestine of broilers and be depolymerized to soluble NSP and can cause significant loss of energy in the form of heat and very violate fatty acids (VFA) these effecting the availability of energy to animals (McNab and Boorman 2002). As a result, the NSP in feed stuffs is the main target of commercial feed enzymes (Liang 2000). Enzymes cleave the NSP molecules into smaller polymers and reduce digesta viscosity. In poultry, corn and SBM do not cause viscosity, even though they contain NSP's. SBM contains approximately 6% NSP and between 8-12 % of insoluble NSP in 3.5% of total fiber content and corn has approximately 0.9 % soluble and 6% insoluble NSP of 3.22 % of fiber, (Knudsen 1997). Wheat is a good source of energy in poultry diets and it has between 7-8 % NSP on dry matter (DM) basis in 3.87% of the fiber content, the problem with wheat is associated with entrapment of wheat starch and protein by polysaccharides (Bedford and Autio 1996). Another problem associated with dietary fiber is the binding activities which are associated with some components of fiber such as pectin which can interact with dietary cations and decrease their digestibility, (Kirk and Oram 1981).

2.1.2. Canola meal processing

Processing conditions during oil extraction are key factors that influences CM feed quality. Beach and Hickling (2010) suggested that utilizable energy can be increased of 5% using current CM processing techniques. While the temperature during processing can be helpful in deactivating myrosinase enzyme in the seed to avoid break down of glucosinolates into toxic metabolites, there are some adverse effects of temperature

during processing. Amino acids, especially lysine and methionine, which are sensitive to heat, might be damaged during processing. This affects the quality of the protein in the meal (Hurrell 1984). A decrease in protein quality and digestibility is due to the Maillard reaction involving amino acids, especially lysine (Newkirk and Classen 1999). Newkirk (2009) collected samples from different stages of prepress solvent extraction to examine the nutritional values of the meal. He found that lysine content reduced ($P < 0.05$) from 6.03 to 5.50% of crude protein. Lysine digestibility decreased from 88 to 79% and the crude protein digestibility decreased from 81% to 76%. The Maillard reaction causes the production of colour and some odor during processing (Hurrell 1984).

2.1.2.1. Pre-Press Solvent Extraction Process

The most common processing method used to extract oil is prepressed solvent extraction (Figure 2.4). The process includes cleaning (according to grading standards based on maximum moisture content, seed damage and chlorophyll level), drying (to 6-7% moisture), seed preconditioning (heating to approximately 75-78˚C to prevent shattering during flaking), flaking (by roller mills to rupture a seed coat and form thin flakes), and cooking (one hour at 75-85˚C to deactivate the myrosinase enzyme and rupture oil cells which have survived flaking and prepare the seed for expelling). The next step is pressing (to remove as much oil as possible, around 50-60% of the seed oil content), solvent extraction with hexane (to remove the remaining oil), desolventization and toasting (to remove the hexane, during this process the meal is heated to 100-110 ˚C and moisture increases to 12-18 %). Toasting the meal helps to decrease the anti-nutritional glucosinolate material in this stage, drying and cooling (to approximately 12% moisture by blowing air over the meal) and the last stage is grinding and pelleting (using a hammer mill to have a uniform consistency). The resulting meal contains less than 1% oil and 12% moisture (CCC 2009). Gums are the portion of phospholipid materials in crude canola oil, which are removed in processing. In Canada, gums are added back to the meal in the desolventization and toasting stage at 1-2%. By adding back gums, the dustiness of the meal can be decreased, the phospholipid content increased and the metabolizable energy value of the meal increasers (CCC 2009). There are some

drawbacks to pre-pressed solvent extraction, such as Maillard browning reactions which occur when the meal is exposed to high temperature of the desolventization-toasting process (100 to 110°C) (CCC 2009). CM can undergo some damage during the desolventization and toasting stage. Newkirk et al. (2003) suggested that the regular temperature of toasting (107°C) can cause some protein damage, reduces protein and lysine digestibility and makes the meal darker in colour. It has been reported that limiting to 100°C can decreased lysine digestibility to levels similar to SBM (Newkirk et al. 2003).

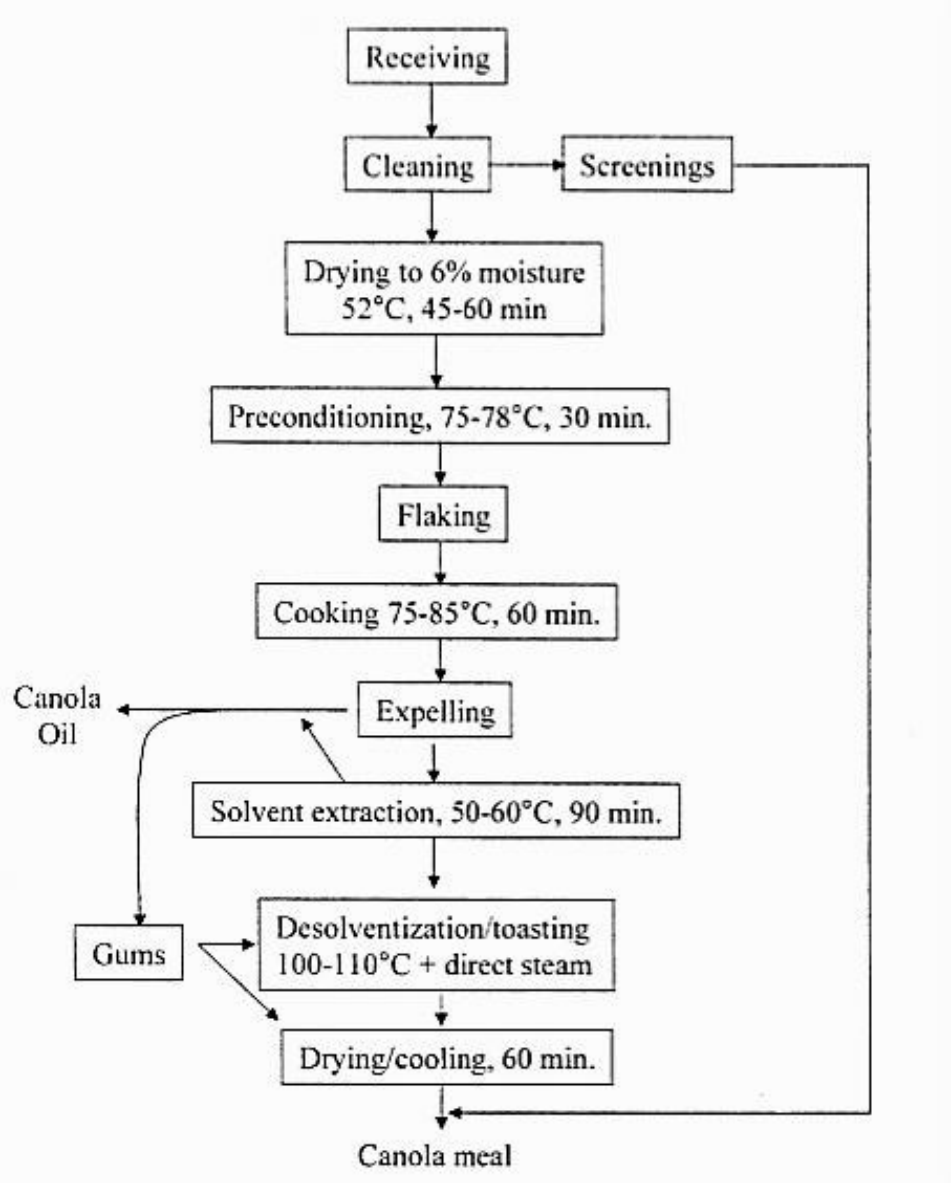

Figure. 2.4. Prepress solvent extraction of canola seed. From Newkirk et al (2003).

2.1.2.2. Other oil extraction procedures

Expelling was the first commercial oil extraction process in western Canada (Bredeson 1983). This process involves cleaning, conditioning, flaking, cooking to 130°C for 30 minutes and feeding into a screw press. Cooking at high temperatures, results in the loss of lysine content and reduced digestibility (Clandinin and Tajcnar 1960).

Double pressing is another method for oil extraction. The method is the same as single press expelling except the seed is pressed a second time to remove more of the oil. With an average residual oil content of 8-15%, the meal has higher metabolizable energy than meal from solvent extraction (2000 kcal·kg^{-1} vs. 2340 kcal·kg^{-1} respectively) (CCC 2009).

Canola oil can be extracted by cold pressing. In this method the temperature during expelling does not exceed 50°C. The main product of cold pressing extraction is a press cake with high residual oil (12-17%) and is considered as a valuable animal feed, without the use of chemical extracting agents (Ferchau 2000). This processing is done to provide oils with special characteristics. The meals available for animal feeding but have not been subjected to high heat processes with their benefits and disadvantages.

2.2. Digestibility of canola and juncea

Digestibility measurements are necessary to describe the nutrient efficiency of feed ingredient utilization. The high fiber content of CM is considered to be the primary factor resulting in the low metabolizable energy and digestibility of protein in CM. The high amount of fiber dilutes the digestible nutrients in the diets and increases the passage rate which results in lower digestion and nutrient utilization (Bell 1993).

NRC (1994) reported the AME_n value of CM for poultry to be 2000 kcal kg^{-1}, which is lower than SBM (2440 kcal·kg^{-1}). Slominski. et al (1999) reported the TME_n (kcal·kg^{-1} DM) in *Brassica napus and Brassica juncea* (yellow-seeded canola meals) as 2320 and 2152 kcal·kg^{-1}, respectively and the TME_n (kcal·kg^{-1} DM) for brown seeded CM

to be 2192 kcal·kg^{-1} by broilers. The dietary fiber ranged from 271 to 352 g kg^{-1} of dry matter. The lowest value of fiber was for yellow-seeded *Brassica napus* and the highest was for commercial meal from brown-seeded *Brassica napus* canola. This low fiber level, explains the higher metabolizable energy in yellow seeded meals.

Differences between turkey and broiler digestibility of the same the diets was reported by Slinger et al (1964). Turkeys were able to use more energy from high fiber feedstuffs than chickens. Leeson et al (1974) concluded the same thing with 8% more ME was available for poults compared to chicks when birds were feed dietary oats.

There are several ways to improve AME_n of CM and JM. They include, reduce the fiber of canola by breeding yellow seeded species, dehulling the seed prior to extraction and use of exogenous dietary enzyme (Khajali and Slominski 2012).

2.3. Growth performance

The world's poultry industry has grown at a rapid rate to meet the high consumption demands. Poultry meat production increased from 8.9 to 70.4 million tons from 1961 to 2006 (Singh 2007). Improvements in body weight gain (BWG) and feed conversion ratio (FCR) are the results of good management, genetics and nutrition of birds (Singh 2007). As feed represents around 55% of the cost of broilers production, proper feed formulation, which meets all the dietary demands of birds to maximize the FCR, is important.

CM and JM with low levels of glucosinolates can be used in poultry diets as a protein source. Balancing diets to ensure adequate levels of the essential amino acids is necessary in formulating poultry diets. Essential amino acid digestibility of CM is lower than SBM. The lower energy in CM compared to other protein sources is another factor that limits using high amount in broiler feeds. In western Canada, CM is typically used at less than 10% in wheat-based diet and a little more in corn-based diets (CCC 2009). The maximum recommended levels of low glucosinolate CM for starter and grower phase in broiler production are 10 and 20 % respectively due to low energy levels of the meal (CCC 2009).

Different studies have been conducted to evaluate the effects of different inclusion levels of CM in broilers. Lee et al. (1991) reported CM could be included at 10

to 20% of the diet for broilers without any negative effects on growth. Birds fed with these levels of CM inclusion in a corn-based diet had similar body weight (BW) and feed conversion ratios (FCR) as the birds fed the control treatment. Naseem et al. (2006b) found broiler chickens fed 25% CM, had a better body weight gain (BWG) and FCR compared to broilers fed 5, 10, 15 and 20 % CM. They suggested 25% CM inclusion in commercial broiler diets to optimize the profits in broiler production. CCC (2009) reported that low levels of glucosinolate in the current CM did not have adverse effects on broiler performance when the diets were formulated with 30% CM. The low energy level of CM is considered as a negative factor, which limits the inclusion level of CM in broiler diets compared to other plant protein sources such as SBM. Another concern about formulating broilers diets with high levels of CM is decreasing feed intake, which is suggested to be related to cation-anion balance of CM (Summers and Bedford 1994).

Yellow seeded and black seeded solvent extracted canola meals were compared for growth performance. FCR was improved for of yellow-seeded *Brassica napus* compared to the black seeded *Brassica napus* (1.53 vs. 1.60 feed $g \cdot gain^{-1}$). Birds that were consuming yellow canola had higher BWG (398 $g \cdot b^{-1} \cdot 14\ d^{-1}$) than black seeded (342 $g \cdot b^{-1} 14 \cdot d^{-1}$) in the starter period (Jiang et al. 1999). This can be associated with the lower fiber level of yellow seeded CM, which increased the metabolizable energy compared to black seeded meals.

Based on the previous studies and inclusion recommendations of CM in broilers diets, the main factor that restricts the higher inclusion rate of CM in diets, is the energy level in the diet rather than the toxic effects of glucosinolates. Higher level of dietary fiber in CM may increases the digesta passage rate and result in reduction of digestion time and consequently nutrient utilization (Khajali and Slominski 2012). By incorporating different strategies to overcome these negative effects of fiber it might be possible to feed higher levels of modern CM to the commercial broilers.

Increasing level of CM and JM to allow the current recommendations of 10% for starter and 20% for grower (CCC 2009), which are based on older studies, might be possible, as the levels of fiber and glucosinolates content of canola and juncea has been decreasing through plant breeding (CCC 2009. Furthermore the growth performance of the birds has been changed through genetic selections.

2.4. Dietary Enzymes

Plant feedstuffs contain dietary fiber. Fiber consists of carbohydrates that are not hydrolyzed by digestive enzymes. There are different variations in fiber in different of plants due to different chemical properties (Smiths and Annison. 1996). Plant cell walls consist of polysaccharides, polyphenolics, glycoproteins and glycolipids (Smiths and Annison. 1996).

In cereal based diets, non-starch polysaccharides (NSP) are poorly digested. Some dissolve in the digestive tract of the birds and can increase viscosity and consequently inhibit digestion (White et al. 1981). The slower rate of passage of digesta in the intestinal tract and the water holding capacity of NSP's can cause sticky droppings. The NSPs in CM are main1y arabinose, xylose, mannose and uronic acids (Simbaya 1996).

Some dietary enzymes have been developed to decrease the negative effects of NSP. Xylanase and β-glucanase are considered to be effective by increasing digestibility and improving birds' performance in cereal based diets (Pettersson et al. 1990). Phytate is unavailable to poultry. Supplementing commercial phytase designed to increase digestibility of phosphorus from plant ingredients, was useful in releasing phosphorous bound in phytate, making it available for broilers (Ravindran et al. 1995). Phytase can increase growth rate and improve FCR, perhaps due to the release of minerals and trace elements from complexes with phytic acid (Simons et al. 1990). Improvements in BWG and FCR accrued in a study by Slominski (1997). They discovered greater improvements by blending carbohydrase and phytase. Xylanase and phytase showed synergy to facilitate each other's activities resulting in a positive effect of blending these two enzymes in wheat-based diets fed to broilers (Ravindran et al. 1999). The AME_n, ileal amino acid digestibility, BWG and FCR all improved. Meng et al (2006) found enzyme supplementation of canola meal diets improved FCR, NSP digestion and AME_n content. Generally, a wide range of positive effects of supplementing carbohydrase enzyme has been shown in poultry. Different feedstuffs have different amount and structures of NSP, as a result, the selection of enzyme for each feed ingredient for improving the nutritional value of feed is really important (McNab and Boorman 2002)

2.4.1. Superzyme-OM^{TM}

Superzyme–OM^{TM}, made by Canadian Bio-Systems Inc. (Calgary, Alberta, Canada) , is an enzyme complex for poultry and swine feeds which was designed for diets containing high levels of flax or canola. This enzyme cocktail contains cellulase, mannanase, galactanase, xylanase, glucanase, amylase and protease. This combination reflects the nature of carbohydrates found in CM. It is recommended to use 500 grams of Superzyme-OM^{TM} per tonne of complete feed (Canadian Bio-Systems Inc., Calgary, Alberta, Canada). Woyengo et al. (2010) found supplementing broiler diets with phytase and Superzyme-OM^{TM} improved BWG and FCR. They suggested phytase supplementation with Superzyme-OM^{TM} as this enzyme mixture improved nutrient utilization and growth performance in broilers. In another study by Jia et al. (2012), multicarbohydrase enzyme addition to broiler diets, increased AME_n values of diets with CM (from 1943 to 2249 kcal kg^{-1}) and a very large increase with JM (from 1736 to 2356 kcal kg^{-1}).

2.5. Liver

2.5.1. Normal function

Liver plays a major role in the digestion, metabolism and utilization of feed ingredients. Bile secretion, carbohydrate, lipid and protein metabolism are some of the important roles carried out by the liver in chickens (Butler 1976).

Lipids come from different origins in the liver. Some physiological dietary disorders, like inhibition in some enzymes such as lipoprotein lipase, due to stress or reduction in the rate of lipid catabolism, and dietary toxic substances can cause the accumulation of fat in the liver (Butler 1976).

The liver detoxifies poisonous substances produced by the body and from feed into harmless metabolites for the body and these metabolites can be easily excreted via the kidneys. Broilers liver adapts easily to different challenges by increasing its functions

(Butler 1976).Generally, in poultry, the metabolic activity of the liver is high, as the liver is mostly responsible for fatty acid synthesis (Butler 1976).

Fatty liver hemorrhagic syndrome (FLHS) sometimes has been referred to as "fat dystrophy", "fatty degeneration" or "liver haemorrhage syndrome" mostly happens in layers and heavy breeders. Usually egg production falls and the birds have pale combs followed by an increase in flock mortality. The liver becomes enlarged, pale and friable (Figure 2.5) (Butler 1976). The main causes of this disease are related to high laying intensity, high energy to protein ratio and high energy source of the diets. FLHS is more common in hens fed corn-based compared to wheat-based diets (Dhawale 2007).

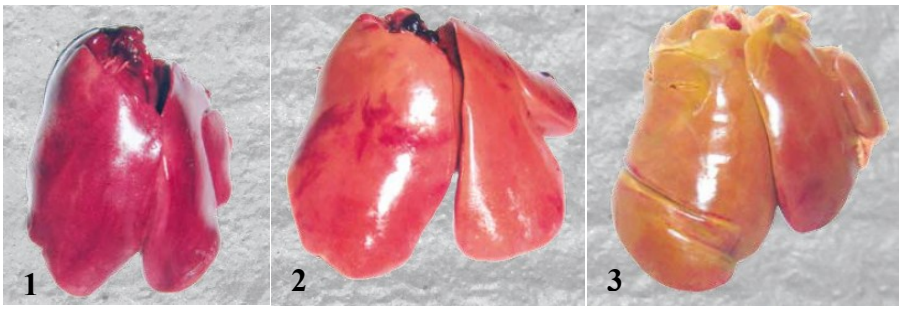

Figure. 2.5. 1) A normal liver, 2) a moderate and 3) extreme cause of fatty liver haemorrhagic syndrome.

From Dhawale (2007)

Olomu et al. (1975b) reported that liver hemorrhage could increase by feeding RSM in layers. Many factors can cause FLHS including consumption of diets high energy, because of the positive energy balance resulting in fat deposition, the presence of excessive fat in diets, or hormonal imbalances (the use of rapeseed meal in diets) (Branton et al. 1995).

Nitrile, which is a breakdown product of progoitrin glucosinolate, the major glucosinolate of rapeseed, was considered to have a negative effect on liver metabolism (Smith and Campbell 1976). Diets containing RSM with a high level of glucosinolate fed to layers, compared to diets with SBM decreased egg production (Ibrahim and Hill 1980). In general, the negative effects of glucosinolates were more severe in layers compared to broilers (Fenwick et al. 1982). The liver is threatened by microbial and chemical toxins.

These toxins can affect its functions, resulting in poor health and production (Dutta 2009).

The liver performs many functions to maintain homeostasis. However, efficiency when the liver decreases in metabolic functions and detoxification, results in poor performance of birds (Dhawale 2007).Therefore, there is a need to study liver function in fast growing birds with high metabolic liver activity.

2.6. Thyroid function in birds fed canola meal

Glucosinolates can be hydrolyzed by myrosinase enzyme and yield products such as: isothiocyanates, goitrin, nitriles, and thiocyanates, which together, interfere with the thyroid function and then have negative effects on growth performance (Khajali and Slominski 2012). Low thyroid activity causes reduction in metabolic rate, increased fat deposition and growth depression (Whittow 2000). Glucosinolate derivatives (higher than 30 μmole·g^{-1}), can lead to hypothyroidism in poultry; this can cause reduction in thyroid hormones and changes the ratio between triiodothyronine (T3) and thyroxin (T4) in the blood (Adibmoradi and Pedram 2007).Increased thyroid stimulating hormones resulting in thyroid enlargement in broilers fed rapeseed meal diets containing more than 30 μmole·g^{-1} glucosinolates has been reported by Schöne et al. (1993). Modern canola meals still have enough goitrogenic activity to result in significant increases in thyroid weight. Adibmoradi and Pedram (2007) showed that, feeding modern CM with low levels of glucosinolate (less than 30 μmole g^{-1}) at 10, 15 and 20% of diet could affect morphology of the thyroid gland. The thyroid weight increased by supplementing with CM at more than 5% of broiler diets without negative effects on bird performance (Adibmoradi and Pedram 2007.

2.7. Carcass composition

Today's consumers are more heath conscious. Lower fat content in poultry meat is more desirable. Producers try to increase the production efficiency by improving BWG and FCR in a short time to be more profitable. This can cause increased fat deposition in

carcasses. The ideal composition is one which satisfies producers in terms of having high BWG and efficient FRC, and consumers, in terms of not having an excessive amount of fat in carcasses (Singh 2007).

Poultry carcasses have moisture, protein and fat as dominant composition components, while carbohydrate, minerals and vitamins represent a small part (Moran 1986). The information on body composition analysis can be useful to evaluate growth pattern, dietary treatments, progression of chronic disease and genetic improvement (Mitchell et al. 2011). As public awareness of excess fat in foods increases, the influence of diets on carcass composition becomes more important to evaluate. Excessive amount of fat in broilers is not desirable for processors or consumers (Jackson et al. 1982). The fat content of broiler carcasses ranged from; 1.4 to 16.7 % (Fraps 1943), related to higher energy intake. In a diet with a high ratio of energy to protein, the carcass fat deposition increasedsand the water and protein content decreases (Donaldson et al. 1956). Summers et al. (1965) found by increasing levels of dietary energy, the protein content of carcass decreased from 64% to 59 % and the fat increased from 18% to 20%.

2.8. Fatty acids

Reducing carcass fat and cholesterol and improving the fatty acid profile of chicken meat can be beneficial for consumers and producers in terms of nutritional benefits. Fatty acid composition can be controlled by the amount and type of dietary fat (Ajuyah et al. 1991). Currently, oils have been used as one of the energy sources in broilers diets. Oil seeds can be used in three forms: unextracted whole seed partially defatted through mechanical processes or defatted meal. Oil seeds are processed through a combination of mechanical pressing and solvent extraction with lipid added back to dietary formulation (Ajuyah et al. 1991). Supplementing fat in diets can supply energy, help the absorption of fat-soluble vitamins, reduce the dustiness and increase palatability of the feed. Oil reduces the passage rate of digesta and provides better digestion and absorption of all diets (Shahryar et al. 2011). Canola and flax seed are two common oilseeds used in Canada. Both are high in fat, protein and alpha linolenic acid (ALA) (Leeson et al. 1978). ALA (C18:3 n-3) is an unsaturated n-3 (Omega 3) fatty acid which

has been shown to improve broiler performance (Sim 1990). Decreased lipid content in the edible portion of broilers, especially unsaturated fatty acids was observed when birds were fed canola oil supplementation (Zanini et al. 2008). Omega 3 fatty acids in canola can decrease fat deposition by reducing very low density lipoprotein circulation levels and decrease fat in blood vessels (Yang et al. 2000). In terms of human health, consuming saturated fat and n-6 (omega 6) polyunsaturated fatty acids (PUFA) increases the risk of cardiovascular disease, while, consuming n-3 PUFA can reduce the risk (Simopoulos 1997). Some vegetable oils like corn, soybean and safflower are rich n-6 PUFA sources while fish, flax seed and canola seed are some examples rich in n-3 PUFA (Schmitz and Ecker 2008). In the past, humans consumed diets ranged ratios between 1:1 to 4:1 of n-6:n-3 with higher levels of long chain eicosapentaenoic acid (20:5n-3; EPA) and docosahexaenoic acid (22:6n-3; DHA) (Simopoulos 1999). Today diets range from 10:1 to 25:1. Therefore, in terms of human nutrition, consuming more n-3 fatty acids especially EPA and DHA is recommended (Betti et al. 2009). ALA (C18:3n-3) and linoleic acid (C18:2 n-6) (LA) are essential fatty acids for humans, as we cannot synthesize them (Betti et al. 2009). Through the desaturation and elongation processes of ALA and LA are further metabolized to long chain PUFA (Gogus and Smith 2010). ALA is converted to stearidonic acid ($18:4_{n-3}$) and eicosatetraenoic acid ($20:4_{n-3}$) to form EPA. EPA is then metabolized to DHA (Schmitz and Ecker 2008). The recommended daily intake of ALA is 2,200mg and for EPA+DHA is 650mg (Simopoulos et al. 1999). Despite the ability of humans to convert some ALA to EPA and DHA, it was found that LA cannot be the only source of EPA and DHA (Wojtasik et al. 2012). ALA from the broiler diet moves to muscle fat (Rymer and Givens. 2005) and their meat can be considered as n-3 PUFA enriched if a minimum level of 300 mg per 100 g of meat is n-3 PUFA (Canadian Food Inspection Agency (CFIA) 2003). Canola and rapeseed oil contain around 9% ALA and the ratio of n-6 to n-3 is 2.5:1 (Leskanich et al. 1997). Improvement in broilers growth performance when fed PUFA from 3.5% rapeseed oil compared to soybean oil and processed fat product (Zollitsch et al. 1997) was observed.

Despite all the positive effects of PUFA on broilers, there are some negative effects that should be considered. PUFA causes a change in physical characteristics of the fat because, they are in a more liquid form and the n-3 enriched products may become

oily and sometimes unacceptable to consumers. The shelf life of the products might be reduced because of the increased chance of oxidative damage due to the more numerous of double bonds in the fatty acids (Wood and Enser 1997). Diets deficient in essential fatty acid can cause some metabolic disorders. Abnormalities in membrane structure, capillaries and skin and general immunity depression are examples of some of these disorders in poultry (Wiseman 1984).

2.9. Summary

As feed represents 60-70% of the costs of production in the poultry industry, taking advantage of lower cost potential feed ingredients is important. Canola has high production in western Canada, approximately 9 million tonnes. Production targets are focused for 15 million tonnes by 2015 (CCC 2009). With more CM available, increased inclusion levels by substituting CM or JM for SBM as a protein source in broiler diets may be economically profitable. Knowledge of the characteristics of CM is critical in formulating diets. Canola and juncea meals both have good amino acid profiles and good oil characteristics. The anti-nutritional factors in these meals should be considered in feeding birds. Anti-nutritional factors can have some adverse effects on thyroid functions, liver metabolism and consequently on growth performance. Modern canola has lower glucosinolate than the old rapeseeds which contained up to 30 $\mu mole \cdot g^{-1}$. Today the level of glucosinolate in Canadian CM is even less. The total glucosinolate content is reported as approximately 7 $\mu mole \cdot g^{-1}$ by CCC (2009). There is a need to reevaluate the effects of glucosinolate in modern varieties of CM and JM on bird performance.

Another problem that has been associated with canola is high level of fiber which makes the meal less available and the nutrients less desirable to the birds. Plant breeders tried to increase the production of yellow seed varieties of canola and juncea with lower levels of fiber. Supplementing higher fiber diets with a multicarbohydrase enzyme cocktail has become common practice in feeding broilers.

The current recommendation of CM inclusion in broiler diets (maximum of 10 and 20% in starter and grower diets respectively) (CCC 2009) is based on some older studies which observed toxic effects of glusosinolates in birds and a lower digestibility

values of CM because of the high levels of fiber in the feed were evident. Evaluating new varieties of CM or JM with lower levels of gluscosinolates by using dietary enzyme to overcome the negative effects of fiber on the new generation of commercial broilers is necessary. In order to formulate diets with CM or JM, apparent metabolizable energy determination (AME_n) of feed should be determined in these meals. There is a lack of information on AME_n content of CM and especially JM for both broilers and turkeys. These AME_n content are necessary, in order to be able to formulate diets with the meals.

CHAPTER 3: DETERMINATION OF THE APPARENT METABOLIZABLE ENERGY CONTENT OF CANOLA MEAL AND JUNCEA MEAL USING BROILER CHICKENS AND TURKEYS

3.1 Abstract

There is a difference between the ability of turkeys and chickens to digest dietary fiber. Studies have shown, turkeys are able to digest fiber more efficiently than chickens and extract more energy from diets. This study determined the nitrogen corrected apparent metabolizable energy (AME_n) content of juncea meal (JM) and canola meal (CM) for both broilers and turkeys. One hundred and eight day-old male chicks and 72 day old female turkey poults, were randomly assigned to three dietary treatments with six broilers or four poults per cage and six replicate cages per treatment. Dietary treatments for both broilers and turkeys started after a standard starter fed up to day 14. From day 15 to day 21 for broilers and from 22 to day 28 for turkeys one of three diets was fed, either a basal grower diet or the basal grower diet supplemented with 30% CM or JM. Celite® was used as an inert marker at 0.8% to determine digestibility. On days 19, 20 and 21 for broilers and days 27, 28 and 29 for turkeys, clean excreta samples were collected from underneath each cage and immediately frozen. Dried excreta and diet samples were analyzed for acid insoluble ash (AIA), gross energy (GE) and nitrogen (N). CM provided an AME_n of 2006 ± 100 kcal·kg $^{-1}$ and AME_n content of JM was 1867 ± 100 kcal·kg $^{-1}$ for broilers. For turkeys, the AME_n in CM and JM, were 2331± 200 kcal·kg $^{-1}$ and 2215 ± 200 kcal·kg $^{-1}$ respectively. The AME_n in JM and CM were not significantly different ($P \geq 0.05$) from each other within each trial. Turkeys made better use of GE in both CM or JM than broilers. There was over 300 kcal AME_n kg $^{-1}$difference between the bird species.

Key words: *AME_n, broilers, turkeys, canola, juncea, Celite®.*

3.2 Introduction

Digestibility of nutrients in new varieties of CM and JM by broilers and turkeys may not be similar to published results (NRC 1994, Lee et al 1995, Newkirk et al. 1997 and Jayaraman 2010), due to the changes in composition of the meals and the genetic improvements of the birds for high production. Consequently, new AME_n values of genetically improved CM and JM for current commercial broilers and turkeys need to be determined.

Metabolizable energy determination of feed ingredients is critical for formulating poultry diets. AME_n and true metabolizable energy (TME) are the most common ways of determining energy in feeds for poultry (NRC 1994). AME is usually corrected to the state of nitrogen equilibrium (Sibbald et al. 1980). When nitrogen is retained in the body, it yields energy containing compounds with metabolite wastethat are voided in the urine, so by using 8.22 $kcal \cdot g^{-1}$ corrections for retained nitrogen, which is the gross energy (GE) value for uric acid, a standard AME_n value can be obtained (Hill and Anderson 1958).

In AME_n determination, the test ingredient is often added at 30% of the diet, so the diet more closely approximates commercial formulations (NRC 1994). The feed is available *ad libitum.* In TME determination a precise amount of test ingredient is delivered directly to the crop. In AME_n method, the physiological changes in the bird are recalculated compared to TME measurement. The palatability of test ingredients can be determined, based on measuring feed intake . In the TME method, the test ingredient is the only feed provided to the bird. Birds are fasted for 24 hours and then tube fed the exact amount of test ingredients directly into the crop and the resulting excreta is analyzed (Sibbald et al. 1980). Digestibility markers are normally used to estimate the digestibility of dietary nutrients. In a study by Scott and Boldaji (1997), the choice of chromic oxide (Cr_2O_3) or insoluble ash marker (Celite®) were tested on wheat and barley based diets. There was no effect of the digestible marker on growth and feed efficiency, but they found chromic oxide was the least accurate method for determining AME_n, as this marker, did not show any differences in AME_n of the barley-based diet with and without dietary enzyme supplementation. While the insoluble ash marker, demonstrated

higher AME_n of barley-based diet with enzyme supplementation due to the effects of enzyme on energy availability of the diet. However, the quantity of sample needed for accurate analysis, was, at least 3 g when Celite® was used compared to 0.5 g required for chromic oxide. This is important where only a small amount of sample is available. Scott and Boldaji (1997), reported the best levels of insoluble ash marker in diets to be between 0.5 and 1.0%.

Generally, AME_n and crude protein requirements are higher for turkeys compared to broiler chicks (NRC 1994). In addition, turkeys appear to utilize more energy from certain feed ingredients, particularly those with higher fiber (Slinger et al. 1964). The AME_n for CM and JM may need to be specifically determined for both chickens and turkeys.

3.3 Objectives

The objective of this study was to determine the AME_n content of current commercial samples of CM and JM for turkeys and broiler chickens.

3.4 Hypotheses

Canola and juncea meals will have higher AME_n for turkeys and broilers compared to NRC (1994) values. Higher AME_n values will be determined in turkeys compared to broilers.

3.5 Materials and methods

3.5.1 Canola and juncea meal

Canola *(Brassica napus)* and juncea meals *(Brassica juncea)* were obtained from seed grown in western Canada. They were derived from commercial seeds crushed at the Altona, Manitoba crushing plant of Bunge Company. The amino acid profile of the meals

used in the study, show variation in the two meals (Appendix A). They were analyzed by an acid hydrolysis procedure at the University of Manitoba, Department of Animal Science according to method 982.30 (AOAC 1990).

The glucosinolate profile of CM and JM were measured by POS Bio Sciences Saskatoon, Saskatchewan, Canada**.** The method was based on the trimethylsilyl (TMS) and non-hydrolyzed glucosinolates as their TMS derivatives were measured by gas chromatography (Underhill and Kirkland 1971).

Table 3.1. Glucosinolate profile of canola (*Brassica napus*) and juncea (*Brassica juncea*)meals [1]

Glucosinolate ($\mu moles \cdot g^{-1}$)	Meal	
	Canola	Juncea
3-butenyl	1.92	10.72
4-pentenyl	0.18	0.48
2-OH-3-butenyl	4.19	0.49
2-OH-4-pentenyl	0.10	-
CH3-thiobutenyl	0.13	-
Phenylethyl	0.12	0.22
CH3-thiopentenyl	0.06	-
3-CH3-indolyl	0.27	-
4-OH-3-CH3-indolyl	1.12	0.24
Allyl	-	0.36
Total aliphatics	8.09	12.51

[1]POS Bio Sciences Saskatoon, Saskatchewan, Canada,reported on $\mu mol \cdot g^{-1}$ on an air-dry, oil-free basis

3.5.2 Broiler trial

Broiler chickens were managed under the guidance of the local animal care and use committee following the guidelines of the Canadian Council of Animal Care (CCAC 2009). Chickens were vaccinated in the hatchery prior to shipping with 0.05 mL of Marek's vaccine. In the broiler trial, one hundred eight, one-day-old male (Ross 508 × Ross 508) chicks were placed in eighteen battery cages (60 cm × 48 cm) with 6 replicates per treatment in a completely randomized experiment. The diets were formulated to meet or exceed NRC (1994) nutrient requirements (Table 3.2). Birds were fed standard starter diet, from day 1-14. Then from day 15-21 one of the treatment diets including the basal diet or basal diet plus 30% of CM or JM were fed. Celite® (Celite Corp., Lompoc, CA, 93436, USA) added at 0.8% was used as an indigestible marker in all the grower diets.

Table 3.2. Ingredient composition and calculated analyses of the starter and grower diets for broilers (%as fed).

	Starter	Grower
Ingredients		
Corn	41.79	48.32
Soybean meal	40.62	32.58
Wheat	10.00	10.00
Poultry fat	3.71	4.79
Limestone	1.80	1.67
Celite®	0.00	0.8
Mono dicalcium phosphate	0.71	0.65
MCBS5[1]	0.50	-
MCBF5[2]	-	0.50
Iodized salt	0.44	0.42
Methionine premix[3]	0.35	0.19
Coban[4]	0.05	0.05
Stafac 44[5]	0.03	0.03
Total	100	100
Calculated analyses (as fed)		
AME_n ($kcal.kg^{-1}$)	3050	3150
Protein (%)	23.00	20.00
Ether extract %	5.58	6.83
Crude fiber (%)	2.55	2.48
Calcium (%)	1.00	0.92
Total phosphorus	0.58	0.51
Lysine (%)	1.43	1.19
Methionine (%)	0.56	0.44
Met + Cys (%)	0.95	0.77
Linoleic acid (%)	1.93	2.30

[1] MCBS5, Broiler starter premix (amount per tonne), vitamin A (650×10^6 IU kg^{-1}),15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000 mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin(50%), l00g;Wheat middlings 1432g;Ground limestone (38%),500g.

[2] MCBF5, Broiler grower premix, vitamin A (650×10^6 IU kg^{-1}), 15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1532g;Ground limestone (38%),500g.

[3] Supplied kg $premix^{-1}$: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[4] Coccidiostat - Coban (active ingredient monensin sodium, 200 g kg^{-1}) Elanco Animal Health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[5] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada.

Diets were fed in mash form throughout the trial from troughs attached at the front of the cage. Broilers were fed *ad libitum* and water was provided through nipple drinkers. Lighting and temperature schedules used are shown in Appendix B. Excreta samples were collected from underneath each cage on days 19, 20 and 21. Bird weight, was measured on days 1, 14 and 21. Feed was weighed back from each cage on days 14 and 21.

3.5.3 Turkey trial

Turkey poults , were managed under the guidance of the local animal care and use committee following the guidelines of the Canadian Council of Animal Care (CCAC 2009). Seventy-two, one-day-old female (Hybrid converter) poults were placed in 18 battery cages (60 cm × 48 cm) in a completely randomized experiment with 4 poults per cage and 6 replicate per treatment. The diets were formulated to meet or exceed NRC (1994) nutrient requirements (Table 3.3). Birds were fed a standard starter diet from day 1-15 and on days 15 to 22 a common basal grower diet was fed. From days 22 to 28, basal grower with 30% of CM or JM were fed. All diets from day 22 to 28 contained 0.8% Celite®. Diets were fed in mash form throughout the trial from troughs at the front of the cage. Turkeys were fed *ad libitum* and water was provided through nipple drinkers. Lighting and temperature schedules used are shown in Appendix C. Excreta samples were collected from underneath each cage on each of days 27, 28 and 29 and immediately frozen. Bird weights were measured on days 1, 14, 22 and 28. Feed was weighed back for all cages on days 14, 22 and 28.

Table 3.3. ingredient composition and calculated analyses of the starter and grower diets for turkeys (%as fed)

	Starter	Grower
Ingredients		
Corn	36.33	34.76
Soybean meal	40.06	41.72
Wheat	10.00	10.00
Poultry fat	1.00	4.16
Poultry by- product	7.44	4.04
Limestone	1.67	1.84
Celite®	-	0.8
Mono dicalcium phosphate	1.86	1.74
ATS5[1]	1.00	-
ATF5[2]	-	0.50
Iodized salt	0.30	0.31
Methionine Premix[3]	0.34	0.13
Total	100	100
Calculated analyses (as fed)		
AME_n ($kcal.kg^{-1}$)	2851	3000
Protein (%)	28.00	26.50
Ether extract (%)	3.70	6.29
Crude fiber (%)	2.52	2.48
Calcium (%)	1.40	1.30
Total Phosphorus	0.65	0.56
Lysine (%)	1.91	1.57
Methionine (%)	0.62	0.55
Met+Cys (%)	1.09	0.99
Linoleic acid (%)	1.35	2.03

[1] ATS5, Turkey starter premix, Supplied per kg diet; vitamin A, 9750 IU; vitamin D_3, 2000 IU; vitamin E, 35 IU; vitamin K, 3.0 mg; riboflavin, 7.6 mg; Dl Ca-pantothenate, 27 mg; vitamin B_{12}, 0.015 mg; niacin, 76.2; folic acid, 4.9 mg, choline chloride, 801 mg; biotin, 0.6 mg; pyridoxine, 5.9 mg; thiamine, 2.9 mg; manganous oxide, 70. 2 mg; zinc oxide, 80.0 mg; copper sulphate, 25 mg; selenium, 0.15 mg; lysine, 29.75 mg; ethoxyquin, 50 mg; wheat middlings, 530 mg; ground limestone, 500 mg.
[2] ATF5, Turkey grower premix, Supplied per kg diet; vitamin A, 9750 IU; vitamin D_3, 2000 IU; vitamin E, 35 IU; vitamin K, 3.0 mg; riboflavin, 7.6 mg; Dl Ca-pantothenate, 27 mg; vitamin B_{12}, 0.015 mg; niacin, 76.2; folic acid, 4.9 mg, choline chloride, 801 mg; biotin, 0.6 mg; pyridoxine, 5.9 mg; thiamine, 2.9 mg; manganous oxide, 70. 2 mg; zinc oxide, 80.0 mg; copper sulphate, 25 mg; selenium, 0.15 mg; lysine, 29.75 mg; ethoxyquin, 50 mg; wheat middlings, 530 mg; ground limestone, 500 mg.
[3] Supplied/kg premix: DL-Methionine, 0.5kg; wheat middlings, 0.5kg

3.5.4 Measurements and data analysis

In these two digestibility trials, duplicate excreta samples calculated from each cage were immediately frozen at -20˚C and then freeze dried. Feed and excreta dry matter

(DM) were measured using method 935. 29 (AOAC 2005). Frozen excreta samples calculatedwere weighed in duplicate then freeze dried and then re-weighed to calculate percent dry matter. Freeze dried excreta and oven dried diet samples were analyzed for AIA with 2N HCl (Van Keulen and Young 1977). The gross energy (GE) of duplicate feed samples and excreta were measured by bomb calorimeter (Parr Instrument Company, Moline, Illinois). Nitrogen, was determined using a Leco Nitrogen analyzer (Leco Corporation, St Joseph, MI) with method 990.03 (Association of Official Analytical Chemists (AOAC) 2005) to calculate crude protein, which is equal to Nitrogen×6.25.

The AME_n was calculated as described by Leeson and Summers (2001).

AME_n = GE diet - (GE Excreta × (AIA diet/AIA excreta)) - 8.22 × N retained.

Where;

AME_n ($kcal \cdot kg^{-1}$) = N - corrected apparent metabolizable energy content of the diet

GE diet and GE excreta ($kcal \cdot kg^{-1}$) = GE of the diet and excreta

AIA diet and excreta (%) = acid insoluble ash in the diet and excreta

8.22= energy value ($kcal \cdot kg^{-1}$) of uric acid

N retained ($g\ kg^{-1}$) is the nitrogen retained by broilers per kilogram of diet consumed.

The retained Nitrogen was calculated as:

N retained=N diet - N excreta × AIA diet/AIA excreta

The AME of the test ingredients was calculated as:

AME_n of the basal diet- [(AME_n of the basal diet- AME_n of the test diet)/0.3].

3.5.5 Statistical analysis

The experimental design was completely randomized. Cages were used as the experimental units, with six replicated for each treatment. All AME_n data were subjected to the Proc Mixed procedure of SAS v.9.3 (SAS Institute Inc., Cary.NC). If treatment effect was significant, the Tukey-Kramer (Littell et al. 1996) option was used to compare differences among the least square means. The α-level for significance was 0.05.

The following model was employed for statistical analysis in the digestibility trials on the AME_n data:

$Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$

Where Y_{ij} is the variable of interest (i= the level of the factor, j= replication within the level); μ is the overall mean; τ_i is the effect of the i^{th} treatment level (i=2) and ε_{ijkl} is the random effect of error.

3.6 Results and discussion

3.6.1 Broiler trial

Results of ANOVA analysis for general performance of broilers (Table 3.4) indicated there was no significant effect of treatments on body weight gain (BWG) and feed consumption (FC) of broilers through the trial. This shows that, both JM and CM are palatable for broilers at 30% of meal inclusion, as the test ingredients, did not affect their growth performance. CM was determined to have an AME_n value of 2006 ± 100 kcal kg^{-1} and JM was determined to have an AME_n value of 1867 ± 100 kcal kg^{-1}. The AME_n of prepress-solvent extracted CM for broilers measured in other studies are compared in Table 3.5. Jayaraman (2010) found that the AME_n content of pre-press solvent extracted JM was not significantly different from CM, both CM and JM that study had higher energy values compared to the current study. The JM in a study by Newkirk et al (1997) had higher glucosinolate level than CM (34.3 vs. 21.8$\mu mol \cdot g^{-1}$). Chromic oxide was used as an indigestible marker in the study by Newkirk et al (1997). The glucosinolate content of the current study was higher for juncea compared to canola meal (Table 3.1) unlike to the study by Newkirk. Newkirk et al. (1997) found no significant differences in the AME values of *Brassica napus* and *Brassica juncea* which was in agreements with our results.

The variation among values for AME_n of canola can be due to different nutrient composition of meals as the seeds came from different places and might have different glucosinolate contents due to the plants, growth conditions. Classen et al (1991) found a 16% improvement in AME of CM with very low level of glucosinolate compared to the commercial CM fed to broilers. However, generally the effect of fiber on AME_n is more considerable (Bell et al. 1991). In this study, the glucosinolate content of both canola and

juncea meals were within the definition for canola, less than 30 μmole·g^{-1}. The difference between digestibility of CM and JM in different studies might be associated with different processing conditions, such as overheating the meal, leading to loss of some amino acids (Parsons et al.1992).

Table 3.4. Performance of broilers fed canola and juncea meals in a digestibility trial (least square mean ±SE)

	Body weight gain (g·b^{-1}·d^{-1})		Feed Consumption (g·b^{-1}·d^{-1})	
Treatments	Days			
	0-14	15-21	0-14	15-21
Basal diet	20±1	52±3	39±2	87±4
Canola meal	17±1	48±3	39±2	78±4
Juncea meal	19±1	55±3	34±2	85±4

ANOVA *P*-values General performance of broilers in digestibility trial

	Body weight gain (g·b^{-1})			Feed Consumption (g·b^{-1})		
Effect	Day					
	0-14	15-21	0-21	0-14	15-21	0-21
Meal	0.191	0.310	0.260	0.260	0.297	0.260

Table 3.5. Apparent metabolizable energy content of canola meal and juncea meal compared to the literature for broilers

	n[1]	Meal (AME_n kcal·kg^{-1})	
		Canola	Juncea
Current study	6	2006±100	1867 ±100
Jayaraman (2010)	16	2462±53	2443±52
Newkirk et al. (1997)	6	1832±79	2216 ±79
Lee et al. (1995)	NA[2]	2050±140	-
NRC (1994)	NA[2]	2000	-

[1] Number of samples
[2]Not available

3.6.2 Turkey trial

ANOVA for general performance of turkeys (Table 3.6), showed there was no effect of treatments on BWG and FC. No significant differences ($P \geq 0.05$) were found in BWG or FC. Table 3.6 shows 30% of JM and CM inclusion are palatable for turkeys without having an adverse effect on growth performance. CM was determined to have an AME_n value of 2331±200 kcal·kg^{-1} and JM was determined to have an AME_n value of

2215±200 kcal kg^{1}. The AME_n value of JM and CM were not significantly different (P= 0.6605) from each other.

No previous information on the AME_n values of JM and CM is available for turkeys to compare the results of the current study. The AME_n content for turkeys fed JM and CM in this study, were higher than broilers which might be associated with their ability to utilize fiber in diets more efficiently than broiler chickens (Slinger et al. 1964).

Table 3.6. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) and Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) of turkeys fed canola meal and juncea meal in digestibility trial (least square mean ±SE)

Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$)			
Treatments	Days		
	0-14	14-21	21-28
Basal diet	19±0	46±0	68±2
Canola meal	19±0	44±0	64±2
Juncea meal	20±0	49±0	63±2
Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$)			
	Days		
	0-14	15-21	22-28
Basal diet	28±0.5	60±3.0	128±10
Canola meal	28±0.5	61±3.0	121±10
Juncea meal	29±0.5	66±3.0	110±10

ANOVA P-values General performance of turkeys in digestibility trial

	Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$)			Feed Consumption ($g \cdot b^{-1} \cdot d^{-1}$)		
Effect	Days					
	0-14	15-21	22-28	0-14	15-21	22-28
Meal	0.236	0.085	0.085	0.531	0.413	0.407

3.7 Conclusions

Determination of the AME_n values, for both the CM and JM, can result in more efficient feed formulation for poultry. The AME_n values of CM and JM for broilers were 2006 and 1867 kcal kg^{-1}, respectively. For turkeys the AME_n values were 2331 and 2215 kcal kg^{-1} for CM and JM respectively.

CHAPTER 4: THE EFFECT OF DIFFERENT LEVEL OF CANOLA MEAL (BRASSICA NAPUS) AND JUNCEA MEAL (BRASSICA JUNCEA) IN BROILER DIETS SUPPLEMENTED WITHOUT OR WITH A DIETARY ENZYME COCKTAIL: GROWTH PERFORMANCE

4.1. Abstract

Canola meal (CM) and juncea meal (JM) are both produced from *Brassica* species with low levels of glucosinolates. Newer varieties have lower anti nutritive factors, however, it has been reported that high inclusion levels of these meals in broiler diets can lead to negative effects on growth performance. In this study, the effects of increasing levels of the meals (CM and JM) with and without a multicarbohydrase enzyme (Superzyme- OM ^{TM}) were evaluated on growth performance in corn- and wheat-based diets. A 2×4×2 factorial analysis with dietary inclusion level of CM or JM (0, 10, 20 and 30 %) and enzyme supplementation (absent or present) was designed using 2560 day-old broiler chicks (Ross 508 × Ross 508), in each trial. Diets were formulated to be isoenergetic and isonitrogenous and fed in mash form. Growth performance data was subjected to analysis of variance using Proc Mixed of the statistical analysis systems. In corn-based diets, improvements in feed conversion ratio (FCR) were found in starter and finisher periods when Superzyme- OM ^{TM} was added. In all the periods for the corn-based trial, supplementing CM compared to JM improved ($P \leq 0.05$) FCR. In the wheat-based trial, improvement ($P \leq 0.05$) in FCR occurred when Superzyme-OM ^{TM} was added compared with the birds which did not consume dietary enzyme. Birds showed better FCR in starter diets when CM was fed compared to JM. In grower diets, birds fed CM at 30% inclusion had better FCR compared those fed JM .In the finisher diets CM had more efficient use of feed than JM. Enzyme supplementation improved ($P \leq 0.05$) BW in all growth periods at 0, 10 and 20% but not at 30% meal inclusion during the grower phase, for both corn and wheat-based diets. Based on the current study, it is recommended to supplement broiler diets with Superzyme-OM ^{TM} in all the growth periods. Thirty percent inclusion level of CM for starter period in both corn and wheat-based diets are recommended. Thirty percent and 20% CM in grower for corn and wheat-

based diets, respectively. A level of 10% CM inclusion for corn and 20% for wheat based diets in finisher are recommended.

Key words: *Growth performance, broilers, canola, juncea, Superzyme-OM* ^{TM}

4.2. Introduction

Knowledge of nutrient content of feedstuffs and subsequent bird performance is necessary for effective utilization of alternative feedstuffs. CM and JM are both produced from *Brassica* species with levels of glucosinolates less than 30 μmol.g^{-1} (CCC 2009). Glucosinolates are considered to have some negative effects on the growth performance of broilers by interfering with the function of the thyroid gland (Khajali and Slominski 2012). Feeding high levels of glucosinolate in wheat-based diets to broilers including 100 or 200 g·kg^{-1}, of solvent extracted modern rapeseed meals (RSM) (*Brassica napus*) caused reduction in feed intake and growth rate (from 645 g of weight gain to 596 g in the starter period) and increased mortality (McNeill et al. 2004). The glucosinolate content of modern canola is only around one-twelfth of older RSM (Khajali and Slominski 2012). In terms of nutrient quality for poultry, CM has high levels of methionine and cysteine which make it a good source of protein with a good amino acid balance (Bell et al. 2000). While, CM is a good source of protein, metabolizable energy and amino acid digestibility of CM is lower than SBM (NRC 1994). An important factor that limits the use of canola in large quantity for broiler diets is high levels of dietary fiber because birds are unable to digest and utilize fiber efficiently. A high level of fiber in the diet can reduce AME and decrease ileal protein digestibility (Newkirk et al. 1997). Supplementation of broiler diets with multicarbohydrase enzymes can overcome the adverse effects of fiber in canola and juncea meal. Meng et al. (2006) indicated the use of a multicarbohydrase enzyme improved the feeding value of canola for broilers by degrading non-starch polysaccharides, therefore, increasing energy utilization.

Currently, the maximum recommended levels of CM for starter and grower in broiler diets are 10 and 20 %, respectively (CCC 2009). Considering the decreasing levels of glucosinolate and fiber in the new varieties and controlling the possible adverse effects of fiber by supplementing with Superzyme-OM^{TM}, it might be possible to include

higher levels of CM or JM in broilers diets. In western Canada, CM is typically included at less than 10% in wheat-based diets and slightly higher in corn-based diets (CCC 2009). Thacker (2005) studied the differences between wheat- and corn-based diets in broilers and found no differences in weight gain or feed intake between broilers fed diets based on wheat or corn. There is a need to look at canola and juncea meals in corn and wheat-based diets as two different common energy sources in poultry feeding.

4.3 Objectives

The objective of this research was to determine the effects of various levels of CM or JM on the broiler growth performance included in corn or wheat-based diets with or without multicarbohydrase enzyme (Superzyme-OM^{TM}).

4.4. Hypotheses

Growth performance of broilers fed graded levels of CM and JM will be similar to control diets and dietary enzymes will improve growth performance of broiler chickens.

4.5. Materials and methods

4.*5.1. Animal experiment*

Broiler chickens were managed under the guidance of the local animal care and use committee following the guidelines of the Canadian Council of Animal Care (CCAC 2009). In each of two trials, 2560 day-old broilers (Ross 508 × Ross 508) were placed randomly in 64 (2.13 m × 1.40 m) floor pens with wood shaving litter and a stocking density of 35 birds per pen (0.85 m^2·bird) at the Atlantic Poultry Research Centre in Truro, NS. Chickens , were vaccinated in the hatchery prior to shipping with 0.05 mL of Marek's vaccine (Intervet/Schering . Plough, Kirkland, QC). .The Product description of Superzuyme-OM^{TM} is listed in Appendix I.

Table 4.1. Ingredient composition, calculated analyses and analyzed composition for starter corn-based broiler diets (%as fed).

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Corn	42.4	39.4	36.4	33.3	42.4	40.2	38.2	36.0
Soy bean meal	40.5	32.8	25.0	17.2	40.5	32.9	25.2	17.6
Canola meal	-	10.0	20.0	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.0	30.0
Wheat	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Canola oil	3.3	4.2	5.1	6.1	3.3	3.3	3.2	3.2
Limestone	1.8	1.7	1.6	1.5	1.8	1.7	1.6	1.5
MonoDicalPO$_4$	0.7	0.7	0.7	0.7	0.7	0.7	0.7	0.6
MCBS5[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Methionine premix[1]	0.4	0.3	0.2	0.2	0.4	0.3	0.2	0.2
Coban[3]	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Stafac 44[4]	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03
Lysine HCL	-	-	-	0.03	-	-	-	0.01
Total	100.1	100.1	100	100	100.1	100.1	100.1	100.1
Calculated composition								
AME$_n$(kcal·kg^{-1})	3050	3050	3050	3050	3050	3050	3050	3050
Protein (%)	23.0	23.0	23.0	23.0	23.0	23.0	23.0	23.0
Crude fiber(%)	2.5	3.4	4.3	5.2	2.5	3.4	4.3	5.2
Lysine (%)	1.43	1.40	1.37	1.35	1.43	1.40	1.37	1.35
Methionine (%)	0.56	0.53	0.52	0.52	0.56	0.53	0.52	0.52
Met +Cys (%)	0.95	0.95	0.97	0.99	0.95	0.95	0.97	1.00
Ether extract (%)	5.26	6.20	7.16	8.15	5.26	5.38	5.52	5.69
Calcium (%)	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Total phosphorus	0.5	0.6	0.7	0.7	0.5	0.4	0.7	0.4
Linoleic Acid (%)	1.0	0.9	0.9	0.8	1.0	0.9	0.9	0.9
Analyzed values (as fed)								
Dry matter (%)	88.98	89.52	89.48	89.50	88.98	89.11	89.02	89.46
Crude protein (%)	24.10	23.59	22.16	23.59	24.10	23.57	24.61	23.30
Calcium (%)	0.96	0.93	0.97	0.95	0.96	0.91	0.96	1.02
Phosphorous (%)	0.55	0.57	0.63	0.68	0.55	0.61	0.69	0.72
Sodium (%)	0.17	0.12	0.16	0.20	0.17	0.20	0.21	0.19
Potassium (%)	0.06	1.03	0.93	0.84	0.06	0.95	0.92	0.87
Magnesium (%)	0.18	0.21	0.23	0.27	0.18	0.21	0.26	0.29
Manganese (ppm)	92	105	91	106	92	93	96	101
Copper(ppm)	34	31	27	36	34	29	35	41
Zinc (ppm)	107	103	105	110	107	110	119	119
Crude Fat (%)	5.82	6.72	7.90	8.85	5.82	5.89	5.66	6.01

[1] MCBS5, Broiler starter premix (amount per tonne), vitamin A (650×10^6 IU kg^{-1}),15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca-pentothenate (45%), 30g; vitamin B_{12} (l000 mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1432g; Ground limestone(38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g kg^{-1}) Elanco Animal Health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

Table 4.2. Ingredient composition, calculated analyses and analyzed composition for starter wheat-based broiler diets (%as fed)

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Wheat	59.5	55.9	52.0	48.2	59.5	56.7	53.8	51.0
Soy bean meal	32.2	25.3	18.4	11.5	32.2	25.3	18.5	11.6
Canola meal	-	10.0	20.0	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.0	30.0
Canola oil	4.0	4.9	5.8	6.7	4.0	4.0	3.9	3.9
Limestone	1.7	1.6	1.6	1.5	1.7	1.7	1.6	1.5
MonoDicalPO$_4$	1.1	1.0	0.9	0.8	1.1	1.0	0.9	0.8
MCBS5[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.5	0.5	0.4	0.5	0.5	0.5	0.4	0.4
Methionine premix[2]	0.5	0.4	0.3	0.2	0.5	0.4	0.4	0.2
Coban[3]	0.05	0.05	0.05	0.05	0.05	0.05	0.06	0.05
Stafac 44[4]	0.03	0.03	0.03	0.03	0.03	0.04	0.05	0.03
Lysine HCL	0.02	0.02	0.02	0.02	0.03	0.02	0.02	0.02
Total	100.1	100.2	100	100	100.1	100.2	100.1	100
Calculated composition								
AME$_n$ (kcal·kg^{-1})	3050	3050	3050	3050	3050	3050	3050	3050
Protein (%)	23.0	23.0	23.0	23.0	23.0	23.0	23.0	23.0
Crude fiber (%)	2.6	3.5	4.4	5.3	2.6	3.5	4.4	5.4
Lysine (%)	1.35	1.35	1.35	1.35	1.35	1.35	1.35	1.35
Methionine (%)	0.57	0.55	0.52	0.52	0.57	0.56	0.54	0.53
Met +Cys (%)	0.95	0.95	0.95	0.97	0.95	0.95	0.95	0.95
Ether extract (%)	5.05	6.01	7.00	7.97	5.05	5.21	5.39	5.56
Calcium (%)	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Total phosphorus	0.6	0.7	0.7	0.8	0.6	0.7	0.7	0.8
Linoleic acid (%)	0.4	0.4	0.4	0.4	0.4	0.4	4.4	0.4
Analyzed values (as fed)								
Dry matter (%)	89.83	89.77	90.76	90.96	89.83	89.91	90.60	91.47
Crude protein (%)	23.63	23.65	24.52	24.90	23.63	23.09	25.01	24.29
Calcium (%)	1.26	1.19	1.20	1.24	1.26	1.15	1.24	1.17
Phosphorous (%)	0.63	0.70	0.73	0.83	0.63	0.73	0.77	0.81
Sodium (%)	0.18	0.21	0.23	0.25	0.18	0.20	0.23	0.22
Potassium (%)	0.91	0.95	0.88	0.86	0.91	0.94	0.92	0.87
Magnesium (%)	0.18	0.22	0.25	0.29	0.18	0.22	0.27	0.32
Manganese (ppm)	113	128	140	141	113	133	136	143
Copper(ppm)	29	31	32	39	29	36	39	39
Zinc (ppm)	104	114	116	127	104	111	122	119
Crude Fat (%)	5.65	6.83	7.83	9.34	5.65	5.97	6.38	6.41

[1] MCBS5, Broiler starter premix (amount per tonne), vitamin A (650×10^6 IU kg^{-1}),15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca-pentothenate (45%), 30g; vitamin B_{12} (l000 mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1432g; Ground limestone(38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g·kg^{-1}) Elanco Animal Health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g ·kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

Table 4.3. Ingredient composition, calculated analyses and analyzed composition for grower corn-based broiler diets (%as fed)

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Corn	52.0	48.6	45.2	41.7	52.0	49.4	46.7	44.1
Soy bean meal	30.7	23.4	16.1	8.8	30.7	23.5	16.4	9.2
Canola meal	-	10.0	20.0	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.0	30.0
Wheat	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Canola oil	3.6	4.6	5.6	6.6	3.6	3.7	3.7	3.8
Limestone	1.6	1.5	1.4	1.3	1.5	1.5	1.4	1.3
MonoDicalPO$_4$	1.0	0.9	0.8	0.7	1.0	0.9	0.84	0.74
MCBF5[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.4	0.4	0.3	0.3	0.4	0.3	0.3	0.3
Methionine premix[2]	0.2	0.1	0.1	0.09	0.2	0.16	0.12	0.07
Coban[3]	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.05
Stafac 44[4]	0.02		0.02	0.03	0.02	0.02	0.02	0.02
Lysine HCL	-	-	0.0 3	0.03	-	-	-	0.03
Total	100.1	100	100.1	100.1	100.1	100	100	100.1
Calculated composition								
AME$_n$(kcal·kg^{-1})	3150	3150	3150	3150	3150	3150	3150	3150
Protein (%)	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0
Crude Fiber (%)	2.6	3.3	4.2	5.1	2.6	3.4	4.33	5.23
Lysine (%)	0.14	1.12	1.10	1.10	0.14	1.12	1.11	1.10
Methionine (%)	0.44	0.44	0.44	0.44	0.44	0.44	0.44	0.44
Met+ Cys (%)	0.76	0.79	0.82	0.85	0.76	0.79	0.82	0.86
Ether Extract (%)	5.97	6.96	7.96	8.96	5.97	6.14	6.31	6.51
Calcium (%)	0.92	0.92	0.92	0.92	0.92	0.92	0.92	0.92
Total Phosphorus	0.58	0.63	0.68	0.73	0.58	0.63	0.67	0.72
Linoleic acid(%)	1.16	1.10	1.05	0.99	1.16	1.12	1.08	1.04
Analyzed values (as fed)								
Dry matter (%)	88.43	88.79	88.96	89.17	88.43	88.69	88.43	88.39
Crude protein (%)	19.02	19.09	20.11	20.63	19.02	20.11	20.40	20.50
Calcium (%)	1.08	1.09	0.88	0.96	1.08	1.04	1.02	0.91
Phosphorous (%)	0.64	0.61	0.61	0.70	0.64	0.68	0.67	0.70
Sodium (%)	0.22	0.19	0.18	0.20	0.22	0.21	0.19	0.16
Potassium (%)	0.88	0.83	0.78	0.74	0.88	0.88	0.83	0.77
Magnesium (%)	0.16	0.20	0.22	0.26	0.16	0.21	0.25	0.29
Manganese (ppm)	108	123	115	122	108	116	111	115
Copper(ppm)	32	32	29	91	32	30	27	28
Zinc (ppm)	117	126	118	117	117	116	117	121
Crude fat (%)	6.99	8.25	8.94	10.00	6.99	7.56	7.66	7.25

[1] MCBF5, Broiler grower premix, vitamin A (650×10^6 IU kg^{-1}), 15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1532g; Ground limestone (38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g·kg^{-1}) Elanco Animal health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

Table 4.4. Ingredient composition, calculated analyses and analyzed composition for grower wheat-based broiler diets (%as fed).

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Wheat	67.9	64.2	60.2	56.4	67.9	65.1	62.1	57.5
Soy bean meal	23.9	17.0	10.25	3.4	23.9	17.1	10.3	3.5
Canola meal	-	10.0	20.0	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.00	30.0
Canola oil	4.6	5.5	6.4	7.3	4.6	4.5	4.5	4.5
Limestone	1.6	1.5	1.5	1.4	1.6	1.5	1.5	3.4
MonoDicalPO$_4$	0.9	0.8	0.7	0.6	0.9	0.8	0.7	0.6
MCBF5[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.4	0.3	0.3	0.3	0.4	0.3	0.3	0.3
Methionine premix[2]	0.2	0.2	0.1	0.1	0.4	0.2	0.1	0.1
Coban[3]	0.04	0.04	0.04	0.04	0.05	0.04	0.05	0.05
Stafac 44[4]	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02
Lysine HCL	0.03	0.03	0.03	0.03	0.03	0.007	0.02	0.03
Total	100.1	100.1	100	100.1	100.1	100.1	100.1	100.4
Calculated composition								
AME$_n$ (kcal·kg^{-1})	3150	3150	3150	3150	3150	3150	3150	3150
Protein (%)	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0
Crude fiber (%)	2.6	3.5	4.4	5.3	2.6	3.5	4.4	5.3
Lysine (%)	1.1	1.1	1.1	1.1	1.1	1.1	1.1	1.1
Methionine (%)	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Met+ Cys (%)	0.7	0.7	0.8	0.8	0.7	0.7	0.8	0.8
Ether extract (%)	5.7	6.7	7.7	8.7	5.7	5.9	6.1	6.3
Calcium (%)	0.9	0.9	0.9	0.9	0.9	0.9	0.9	0.9
Total phosphorus	0.6	0.6	0.7	0.7	0.6	0.6	0.7	0.7
Linoleic acid (%)	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.4
Analyzed values (as fed)								
Dry matter (%)	88.26	88.61	88.99	88.89	88.26	88.70	88.73	88.84
Crude protein (%)	21.09	21.91	21.02	21.46	21.09	21.18	20.82	21.10
Calcium (%)	0.93	0.94	0.89	0.90	0.93	0.89	0.99	0.94
Phosphorous (%)	0.64	0.69	0.69	0.70	0.64	0.67	0.73	0.77
Sodium (%)	0.16	0.24	0.20	0.21	0.16	0.15	0.21	0.21
Potassium (%)	0.79	0.74	0.69	0.66	0.79	0.78	0.70	0.69
Magnesium (%)	0.19	0.21	0.24	0.27	0.19	0.21	0.26	0.30
Manganese(ppm)	118	122	113	122	118	112	117	110
Copper (ppm)	27	36	37	26	27	27	30	28
Zinc (ppm)	109	116	110	114	107	112	106	166
Crude fat (%)	6.40	8.03	8.56	9.36	6.40	6.84	6.98	6.12

[1] MCBF5, Broiler grower premix, vitamin A (650×10^6 IU kg^{-1}), 15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g; Wheat middlings 1532g; Ground limestone (38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g ·kg^{-1}) Elanco Animal Health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

Table 4.5. Ingredient composition, calculated analyses and analyzed composition for finisher corn-based broiler diets (% as fed)

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Corn	57.2	53.8	50.4	47.0	57.2	54.5	52.0	49.3
Soy bean meal	25.9	18.5	11.2	3.9	25.9	18.7	11.5	4.36
Canola meal	-	10.0	20.0	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.0	30.0
Wheat	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Canola oil	3.5	4.4	5.4	6.3	3.5	3.6	3.6	3.6
Limestone	1.5	1.5	1.4	1.3	1.5	1.5	1.4	1.3
MonoDicalPO$_4$	0.9	0.9	0.8	0.7	0.9	0.8	0.7	0.6
MCBF5[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.4	0.3	0.3	0.3	0.4	0.3	0.3	0.3
Methionine premix[2]	0.1	0.07	0.03	-	0.1	0.06	0.02	-
Coban[3]	0.04	0.04	0.04	0.04	0.04	0.04	0.04	0.04
Stafac 44[4]	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.02
Total	100.1	100	100.1	100.1	100.1	100	100.1	100
Calculated composition								
AME$_n$ (kcal·kg^{-1})	3200	3200	3200	3200	3200	3200	3200	3200
Protein (%)	18.0	18.0	18.0	18.0	18.0	18.0	18.0	18.0
Crude fiber (%)	2.49	3.37	4.26	5.15	2.49	3.40	4.31	5.22
Lysine (%)	0.99	0.97	0.95	0.93	0.99	0.98	0.96	0.95
Methionine (%)	0.37	0.37	0.37	0.37	0.37	0.37	0.37	0.38
Met+ Cys (%)	0.65	0.69	0.72	0.75	0.65	0.69	0.72	0.76
Ether extract (%)	6.02	6.95	7.87	8.80	6.02	6.20	6.36	6.54
Calcium (%)	0.90	0.90	0.90	0.90	0.90	0.90	0.90	0.90
Total phosphorus	0.55	0.60	0.65	0.70	0.55	0.60	0.64	0.69
Linoleic acid (%)	1.23	1.18	1.13	1.07	1.23	1.19	1.16	1.12
Analyzed values (as fed)								
Dry matter (%)	87.63	88.04	88.76	89.13	87.63	88.01	88.14	88.23
Crude protein (%)	18.80	18.60	18.59	18.45	18.80	18.74	19.18	19.35
Calcium (%)	1.02	0.88	0.91	0.94	1.02	0.92	0.99	1.00
Phosphorous (%)	0.54	0.58	0.58	0.64	0.54	0.62	0.65	0.69
Sodium (%)	0.16	0.20	0.16	0.16	0.16	0.20	0.18	0.17
Potassium (%)	0.77	0.72	0.66	0.63	0.77	0.72	0.73	0.69
Magnesium (%)	0.16	0.18	0.21	0.26	0.16	0.20	0.25	0.29
Manganese (ppm)	106	102	117	114	106	120	105	116
Copper(ppm)	31	32	28	30	31	39	22	30
Zinc (ppm)	104	105	107	106	104	108	112	112
Crude fat (%)	6.77	7.74	8.26	9.58	6.77	7.07	6.92	6.80

[1] MCBF5, Broiler grower premix, vitamin A (650×10^6 IU kg^{-1}), 15g, vitamin D_3 permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1532g; Ground limestone (38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g·kg^{-1}) Elanco Animal Health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

Table 4.6. Ingredient composition calculated analyses and analyzed composition for finisher wheat-based broiler diets (%as fed)

	Canola meal				Juncea meal			
Levels	0	10	20	30	0	10	20	30
Ingredients								
Wheat	54.8	51.3	47.5	43.6	54.8	52.3	49.2	46.3
Corn	16.8	16.7	16.7	16.7	16.8	16.7	16.7	16.7
Soy bean meal	20.6	13.7	20.0	0	20.6	13.7	6.9	0
Canola meal	-	10.0	6.8	30.0	-	-	-	-
Juncea meal	-	-	-	-	-	10.0	20.0	30.0
Canola oil	4.2	5.0	5.9	6.8	4.2	4.2	4.1	4.1
Limestone	1.6	1.5	1.5	1.4	1.6	1.5	1.5	1.4
MonoDicalPO$_4$	0.9	0.8	0.7	0.6	0.9	0.7	0.7	0.6
MCBF5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Iodized salt	0.4	0.3	0.3	0.3	0.4	0.3	0.3	0.3
Methionine Premix [2]	0.17	0.12	0.07	0.03	0.17	0.12	0.07	0.03
Coban[3]	0.05	0.04	0.04	0.04	0.05	0.04	0.04	0.04
Stafac 44[4]	0.03	0.02	0.02	0.02	0.03	0.02	0.02	0.02
Lysine HCL	-	0.030	0.031	0.031	-	0.007	0.020	0.033
Total	100.1	100	100.1	100	100.1	100	100.1	100
Calculated composition								
AME$_n$ (kcal kg^{-1})	3200	3200	3200	3200	3200	3200	3200	3200
Protein (%)	18	18	18	18	18	18	18	18
Crude Fiber (%)	2.6	3.5	4.3	5.2	2.6	3.5	4.4	5.3
Lysine (%)	0.9	0.9	0.9	0.9	1.1	0.9	0.9	0.9
Methionine (%)	0.3	0.3	0.3	0.3	0.4	0.3	0.3	0.3
Meth+Cyst (%)	0.6	0.6	0.7	0.7	0.7	0.6	0.6	0.7
Ether Extract (%)	5.7	6.7	7.6	8.6	5.7	5.9	6.1	6.3
Calcium (%)	0.9	0.9	0.9	0.9	0.9	0.9	0.9	0.9
Total Phosphorus	0.5	0.6	0.6	0.7	0.6	0.6	0.6	0.7
Linoleic Acid (%)	0.6	0.6	0.6	0.6	0.4	0.6	0.6	0.6
Analyzed values (as fed)								
Dry matter (%)	87.23	87.41	87.54	87.65	87.23	87.61	87.67	87.37
Crude Protein (%)	18.96	19.07	17.95	18.23	18.96	19.33	18.83	18.23
Calcium (%)	0.95	0.98	0.90	0.91	0.95	1.10	0.98	0.88
Phosphorous (%)	0.56	0.61	0.63	0.65	0.56	0.66	0.64	0.67
Sodium (%)	0.20	0.19	0.18	0.19	0.20	0.22	0.18	0.14
Potassium (%)	0.76	0.73	0.68	0.63	0.76	0.71	0.67	0.62
Magnesium (%)	0.17	0.19	0.23	0.25	0.17	0.21	0.24	0.27
Manganese (ppm)	109.29	128.19	131.08	126.65	109.29	120.83	114.31	117.52
Copper(ppm)	29.29	32.55	25.49	35.71	29.29	42.22	32.88	31.53
Zinc (ppm)	107.32	111.97	117.46	114.08	107.32	119.77	109.96	109.11
Crude Fat (%)	6.14	7.08	8.30	8.95	6.14	6.60	6.56	6.23

[1] MCBF5, Broiler grower premix, vitamin A (650×10^6 IU kg^{-1}), 15g, vitamin D $_3$ permix (50×10^6 IU kg^{-1}), 40g; vitamin E (500,000 IU kg^{-1}), 50 g; vitamin K (33%), 9 g; Riboflavin (95%), 8g; DL Ca- pentothenate (45%), 30g; vitamin B_{12} (l000mg kg^{-1}), 23g; Niacin (99%), 30; Folic acid (3%), 133g; Choline chloride (60%), 1335g; Biotin (0.04%), 750 g; Pyridoxine (990,000 mg kg^{-1}), 5g; Thiamin (970,000 mg kg^{-1}), 3g; Manganous oxide (60%), 117g; Zinc oxide (80%), l00g; Copper sulphate (25%), l00g; Selenium premix (675 mg kg^{-1}), 220g; Ethoxyquin (50%), l00g;Wheat middlings 1532g; Ground limestone (38%),500g.

[2] Supplied kg premix^{-1}: DL-Methionine, 0.5kg; wheat middlings, 0.5kg.

[3] Coccidiostat - Coban (active ingredient monensin sodium, 200 g kg $^{-1}$) Elanco Animal health, Division Eli Lilly Canada Inc., Guelph, ON, Canada.

[4] Antibiotic - Stafac 44 (active ingredient virginiamycin, 44 g kg^{-1}) Phibro Animal Health Ltd., Regina, SK, Canada

The diets were formulated to meet or exceed NRC (1994) nutrient requirements for three growth periods; starter (day 0-14) (Table 4.1 and Table 4.2), grower (day 15-24) (Table 4.3 and Table 4.4), and finisher (day 25-35) (Table 4.5 and Table 4.6). Diets within a period were isoenergetic and isonitrogenous in mash form with different primary grains for each trial, (corn, Trial 1 or wheat, Trial 2). All birds had *ad libitum* access to the feed and water. Feed was provided in (51cm × 43cm) cardboard trays for the first week and from tube feeders for the duration of the experiment, water was provided via nipple drinkers for the entire experiment. Some chicks in each pen were introduced to water by dipping their beaks in water immediately after they were placed on the floor. Lighting and temperature schedules used are shown in Appendix D. Analyzed composition of CM and JM are reported in Appendix E.

4.5.2. Growth performance analysis

For each trial on days 1, 14, 24 and 36, birds were weighed by pen and feed was weighed back to determine consumption for the period. On day 35, for both trials, all birds were slaughtered and processed for further analysis. Throughout the trials, feed was weighed when delivered and when mortality occurred. Based on the recorded data, feed consumption (FC), FCR, BW and BWG and % mortality were calculated.

4.5.3. Statistical analysis

Each trial (corn and wheat based) was designed as a 2×4×2 factorial experiment with dietary inclusion level of CM and JM (0,10,20 and 30%) and enzyme supplementation (either present or absent). The experimental design was a randomized complete block with four blocks. The blocks were rooms with all treatments equally repeated in each room. Pens were used as the experimental units. All the growth performance data and percent of mortality were subjected to analysis of variance using the Proc Mixed procedure of the SAS v.9.3 (SAS Inc., Cary NC). Growth data were analyzed as repeated measures with day as a factor. compound symmetry covariance structure were used for repeated measures. Where interactions with day were significant

(P=0.05) data was sliced by day and analyzed separately. If significant effects were found, the Tukey-Kramer option (Littell et al. 1996) was used to compare differences among the least square means. The α-level for significant difference was 0.05.
The model statement for both trials was:

$$Y_{ijklm}=\mu+\rho_i+\alpha_j+\beta_k+\gamma_l+\delta_m+\alpha\beta_{jk}+\alpha\gamma_{jl}+\alpha\delta_{jm}+\beta\gamma_{kl}+\beta\delta_{km}+\gamma\delta_{lm}+\alpha\beta\gamma_{jkl}+\alpha\beta\delta_{jkm}+\beta\gamma\delta_{klm}+\alpha\beta\gamma\delta_{jklm}+\varepsilon_{ijklm}$$

Where Y_{ijklm} is the variable of interest; μ is the overall mean; ρ_i is the effect of the i^{th} block (i=1-4); α_j is the effect of j^{th} meal(j=1-2); β_k is the k^{th} level of meal inclusion (k=1-4); γ_l is the effect of l^{th} enzyme (l=1-2);δ_m is the effect of m^{th} day (m=1-4); $\alpha\beta_{jk}$ is the effect of the interaction between meal and the level of inclusion, $\alpha\gamma_{jl}$ is the effect of the interaction between meal and enzyme, $\alpha\delta_{jm}$ is the effect of the interaction between meal and day, $\beta\gamma_{kl}$ is the effect of interaction between level of inclusion and enzyme; $\beta\delta_{km}$ is the effect of interaction between level inclusion and day, $\gamma\delta_{lm}$ is the effect of interaction between enzyme and day, $\alpha\beta\gamma_{jkl}$ is the effect of interaction between meal, meal inclusion level and enzyme; $\beta\gamma\delta_{klm}$ γ_{jkl} is the effect of interaction between level, enzyme and day, $\alpha\beta\gamma\delta_{jklm}$ is the effect of interaction between meal, level, enzyme and day and ε_{ijkl} is the random effect of error.

4.6. Results and discussion

4.6.1. Corn-based trial

ANOVA for BW and BWG (Table 4.7) indicated significant effects of inclusion level × enzyme on BW in grower and finisher periods and BWG in the grower period. The effect of type of meal × enzyme was significant for BWG in the grower period.

Table 4.7. ANOVA *P*-values for body weight ($g \cdot b^{-1}$) and body weight gain ($g \cdot b^{-1} d^{1}$) in broilers fed canola or juncea meals in corn-based diet

	Body weight ($g \cdot b^{-1}$)			Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$)		
	Day					
Effect	14	24	35	0-14	15-24	25-35
Meal	0.493	0.904	0.786	0.504	0.788	0.510
Level	0.022	0.075	0.001	0.028	0.028	0.039
Meal × Level	0.598	0.693	0.190	0.531	0.265	0.696
Enzyme	<.0001	<.0001	<.0001	<.0001	<.0001	0.051
Meal × Enzyme	0.425	0.062	0.071	0.458	0.018	0.664
Level × Enzyme	0.124	<.0001	<.0001	0.128	<.0001	0.424
Meal × Level × Enzyme	0.397	0.783	0.806	0.524	0.546	0.568

The data of the starter period for BW (Table 4.8) and BWG (Table 4.9) show that adding Superzyme-OM^{TM} to the diet improved bird weights regardless of level of CM or JM. Ahmadauli et al (2008) showed improvement in BW by adding dietary multicarbohydrase enzyme in corn-based diets with different levels of CM (0, 20, 30 and 40%). This agrees with Kermanshahi and Abbasi Pour (2006), who added 0.025% of NSP-degrading enzyme to RSM in a corn-based diet. This resulted in improvement in BWG of broiler chickens during starter period.

There were no differences in BW or BWG for birds given the lowest (10%) and highest (30%) meal levels in starter periods. This is in agreement with Naseem et al (2006a), where there was no negative effect in BWG of broilers fed with different levels of CM (5 to 25%). Ahmadauli et al (2008) on the other hand, found that increasing the dietary level of CM negatively affected, BWG, FCR and FC. While in the current study 20% of CM inclusion increased BW in grower period compared to 0%. The BW during grower and finisher periods (Table 4.10 and 4.11) improved by supplementing Superzyme-OM^{TM} in all meal inclusion levels, except at 30% in grower. This supports the positive effect of enzyme on nutrient digestibly, which can increase the availability of nutrients in the meals. 10, 20 and 30% inclusion levels of CM or JM did not affect BW up to the grower period. This is similar to the study by Lee et al (1991) who did not find any significant differences in BW of broilers fed 10 or 20% CM in corn-based diets. The decrease in BW can be observed in the finisher period from the interaction effects of level × enzyme (Table 4.11). Birds had significantly lower BW when 20 and 30% of

meal inclusion was supplemented with Superzyme-OM™ compared to 0 and 10% when enzyme was present. Based on these results, it is beneficial to broilers fed CM or JM diets to supplement with Superzyme-OM™, in the starter and grower phase. No negative effects of level can be found in these periods. Supplementing the high inclusion level of meals without any adverse effects, shows that the glucosinolate level of the meals which was in an acceptable range for poultry.

Table 4.8. Body weight ($g \cdot b^{-1}$) of broiler chickens fed corn-based starter diets (day 14) containing different levels of canola meal or juncea meal with and without Superzyme – OM™ (least square mean ±SE)

Day 14					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	320±10	386±10	332±10	374±10	353 ±5*b*
10%	332±10	377±10	332±10	384±10	356±5*ab*
20%	336±10	397±10	365±10	397±10	374±5*a*
30%	361±10	376±10	350 ±10	378±10	366±5*ab*
Enzyme	NO		YES		
	341±4*b*		384±4*a*		

[a-b] Means ± SEM with different letters for levels and enzyme are significantly different (α=0.05)

Table 4.9. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed corn-based starter diets (0-14) containing different levels of canola meal or juncea meal with and without Superzyme – OM™ (least square mean ± SE)

0-14 days					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	20±1	25±1	21±1	24±1	22±0*b*
10%	20±1	24±1	21±1	24±1	23±0*ab*
20%	21±1	25±1	23±1	25±1	24±0*a*
30%	23±1	14±1	22±1	24±1	23±0*ab*
Enzyme	NO		YES		
	21±0*b*		24±0*a*		

[a-b] Means ± SEM with different letters for levels and enzyme are significantly different (α=0.05)

Table 4.10. Body weight ($g \cdot b^{-1}$) of broiler chickens fed corn-based grower diets (day 24) containing different levels of canola meal or juncea meal with and without Superzyme– OM ™ (least square mean ±SE)

	Day 24					
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme Levels	NO	YES	NO	YES	NO	YES
0%	885±21	1104±21	898±21	1088±21	892±15*d*	1096±15*a*
10%	928±21	1109±21	965±21	1110±21	946 ±15*cd*	1110±15*a*
20%	942±21	1111±21	982±21	1077±21	962±15*c*	1094±15*a*
30%	988±21	1057±21	981±21	1033±21	985±15*bc*	1045±15*ab*

[a-c] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

Table 4.11. Body weight ($g \cdot b^{-1}$) of broiler chickens fed corn-based finisher diets (day 35) containing different levels of canola meal or juncea meal with and without dietary enzyme (least square mean ±SE)

	Day 35					
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme Levels	NO	YES	NO	YES	NO	YES
0%	1881±36	2279±36	1900±36	2289±36	1891±25 *d*	2284±25*a*
10%	1942±36	2272±36	2029±36	2289±36	1985±25*cd*	2280±25*a*
20%	1917±36	2183±36	1981±36	2142±36	1949±25 *d*	2163±25*b*
30%	1967±36	2142±36	1949±36	2044±36	1958±25*d*	2093±25*bc*

[a-c] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

The interaction of inclusion level of oilseed meals and enzyme for BWG (Table 4.12) in the grower period, shows that birds fed 0, 10 and 20% of dietary meal inclusion with enzyme supplementation had the highest BWG. The interaction effect of enzyme by meal showed that the enzyme effects were different depending on the meal. Enzyme supplementation increased BWG. On average, enzyme had a greater effect on BWG of birds fed canola than on birds fed juncea (Table 4.12).In the finisher period (Table 4.13), birds with the lowest dietary inclusion of either CM or JM (10%) had the highest BWG compared to the 20 and 30% inclusion level. But there is no significant difference (P>0.05) among control diets (0% of meal inclusion) and 20 and 30 % meal inclusion, showing supplementing diets with high inclusion levels of canola or juncea meals was acceptable. On the other hand, Woyengo et al. (2010) observed a linear decrease in BWG when they increased the levels of CM in a corn-based diet. Ahmadauli et al (2008) found that BWG was reduced when 20 to 40% of CM was included.

Based on the results of this study and the data from other researchers in finisher period not more than 10% of meal inclusion is recommended. Formulating diets based on ileal amino acid digestibility of diets can be beneficial for further studies

Table 4.12. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed corn-based grower diets (15-24) containing different levels of canola meal or juncea meal with and without Superzyme– OM™ (least square mean ±SE)

15-24 day						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme	NO	YES	NO	YES	NO	YES
Levels						
0%	57±1	72±1	57±1	71±1	57±1*e*	72±1*a*
10%	60±1	73 ±1	63±1	73±1	61 ±1*d*	73±1*a*
20%	61±1	71±1	62±1	68±1	61±1*d*	70±1*ab*
30%	63±1	68±1	63±1	66±1	63±1*cd*	67±1*bc*
Enzyme × Meal	60±1*b*	71 ±1*a*	61±1*b*	69±1*a*		

[a-d] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

Table 4.13. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed corn-based finisher diets (25-35) containing different levels of canola meal or juncea meal with and without Superzyme– OM™ (least square mean ±SE)

25-35 day					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	91±8	107±8	91±8	109±8	99±4*ab*
10%	123±8	106±8	97±8	107±8	108±4*a*
20%	89±8	97±8	91±8	97±8	94±4*b*
30%	89±8	99±8	88±8	92±8	92±4*b*

[a-b] Means ± SEM with different letters for levels are significantly different (α=0.05)

ANOVA for FC and FCR (Table 4.14) indicated the significant effects of enzyme on FC throughout the whole study and its effect on FCR on starter and finisher periods. The effect of types of meal was significant for FC and FCR through the whole study.

Table 4.14. ANOVA *P*-values for feed Consumption ($g \cdot b^{-1} \cdot d^{-1}$) and feed conversion ratio in broilers fed canola or juncea meals in corn-based diet

	Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$)			Feed Conversion ratio		
	Day					
Effect	0-14	15-24	25-35	0-14	15-25	25-35
Meal	0.001	0.000	0.005	0.036	0.000	0.031
Level	0.202	0.341	0.094	0.538	0.412	0.119
Meal × Level	0.108	0.587	0.983	0.109	0.275	0.637
Enzyme	<.001	<.001	0.046	0.008	0.377	0.011
Meal × Enzyme	0.537	0.914	0.690	0.508	0.359	0.845
Level × Enzyme	0.496	0.095	0.250	0.919	0.866	0.439
Meal × Level × Enzyme	0.099	0.968	0.459	0.538	0.931	0.459

During days 0-14 (Table 4.15) birds fed JM consumed more feed than birds fed CM ($P \leq 0.05$). Birds with dietary enzyme inclusion had higher FC ($P \leq 0.05$). The same pattern occurred during days 15-24 (Table 4.16) and 25-35 (Table 4.17).Unlike the current study, Woyengo et al (2010) found that feeding birds with increasing inclusion levels of CM resulted in a linear decrease in FC. Even the highest inclusion level of 30%, did not affect consumption in the current study.

FCR in the starter period (Table 4.18) indicated birds consuming CM were more efficient than the birds consuming JM. Birds with dietary enzyme supplementation had better FCR than without enzyme. In the grower phase (Table 4.19) the same pattern for the effect of type of meal can be observed, as birds with CM had a better ($P \leq 0.05$) FCR than JM. The same results that were obtained from starter phase can be observed for the finisher period (Table 4.20) for FCR. The difference of FCR between types of meal may be a result of a difference between glucosinolates profiles of CM and JM, (Table 3.2). 3-butenyl the most common glucosinolate in JM (10.72 vs.1.92 $\mu mole \cdot g^{-1}$) is found to be more toxic to the birds and might affect growth performance (Slominski et al. 2012). It has been reported that JM has a higher amount of NSP in the fiber fraction of meal than

CM (Simbaya 1996). This difference in fiber could contribute to differences observed in performance of birds consuming CM compared to JM.

Table 4.15. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) by broiler chickens fed corn-based starter diets (0-14) containing different levels of canola meal and juncea meal with and without Superzyme– OM ™ (least square mean ±SE)

0-14 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	30±0	34 ±0	31±0	35±0
10%	30±0	34 ±0	32±0	35±0
20%	32±0	35±0	33±0	36±0
30%	32±0	32±0	34±0	38±0
Meal	32±0*b*		34±0*a*	
Enzyme	NO		YES	
	32±0*b*		35±0*a*	

[a-b] Means ± SEM with different letters for meals and enzyme are significantly different (α=0.05)

Table 4.16. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) by broiler chickens fed corn-based grower diets (15-24) containing different levels of canola meal and juncea meal with and without Superzyme – OM™ (least square mean ±SE)

15-24 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	88±5	111±5	101±5	126±5
10%	100±5	115±5	105±5	119±5
20%	101±5	112±5	113±5	121±5
30%	99±5	104±5	106±5	115±5
Meal	104 ±2*b*		113±2*a*	
Enzyme	NO		YES	
	101 ±2*b*		115±2*a*	

[a-b] Means ± SEM with different letters for meals and enzyme are significantly different (α=0.05)

Table 4.17. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) by broiler chickens fed corn-based finisher diets (25-35) containing different levels of canola meal and juncea meal with and without Superzyme– OM ™ (least square mean ±SE)

25-35 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	183±8	198±8	199±8	203±8
10%	170±8	199±8	188±8	199±8
20%	191±8	181±8	197±8	201±8
30%	173±8	177±8	185±8	192±8
Meal	184±3*b*		196±3*a*	
Enzyme	NO		YES	
	186±3*b*		194±3*a*	

[a-b] Means ± SEM with different letters for meals and enzyme are significantly different (α=0.05)

Table 4.18. Feed conversion ratio of broiler chickens fed corn-based starter diets (0-14 days) containing different levels of canola meal and juncea meal with and without Superzyme– OM ™ (least square mean ±SE)

0-14 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.51±0.1	1.41±0.1	1.52±0.1	1.47±0.1
10%	1.47±0.1	1.43±0.1	1.57±0.1	1.43±0.1
20%	1.50±0.1	1.39±0.1	1.44±0.1	1.41±0.1
30%	1.41±0.1	1.33±0.1	1.53±0.1	1.53±0.1
Meal	1.43±0.0*b*		1.49±0.0*a*	
Enzyme	NO		YES	
	1.49±0.0*a*		1.42±0.0*b*	

[a-b] Means ± SEM with different letters for meals and enzyme are significantly different (α=0.05)

Table 4.19. Feed conversion ratio of broiler chickens fed corn-based grower diets (15-25 days) containing different levels of canola meal and juncea meal with and without dietary enzyme (least square mean ±SE)

15-25 day				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	1.55±0.1	1.54±0.1	1.78±0.1	1.76±0.1
10%	1.67±0.1	1.57±0.1	1.65±0.1	1.65±0.1
20%	1.67±0.1	1.57±0.1	1.83±0.1	1.78±0.1
30%	1.58±0.1	1.52±0.1	1.68±0.1	1.75±0.1
Meal	1.58±0.03*b*		1.74±0.03*a*	

[a-b] Means ± SEM with different letters for meals are significantly different (α=0.05)

Table 4.20. Feed conversion ratio of broiler chickens fed corn-based finisher diets (25-35 days) containing different levels of canola meal and juncea meal with and without Superzyme– OM ™ (least square mean ±SE)

25-35 day				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	2.02±0.1	1.86±0.1	2.19±0.1	1.87±0.1
10%	1.85±0.1	1.88±0.1	1.96±0.1	1.86±0.1
20%	2.18±0.1	1.86±0.1	2.17±0.1	2.08±0.1
30%	1.95±0.1	1.79±0.1	2.11±0.1	2.09±0.1
Meal	1.92±0.04*b*		2.04±0.04*a*	
Enzyme	NO		YES	
	2.05±0.04*a*		1.91±0.04*b*	

[a-b] Means ± SEM with different letters for meals and enzyme are significantly different (α=0.05)

In the current study, FCR shows that there is no adverse effect of adding different levels of CM or JM in broiler diets. Lee et al. (1991) did not find any differences in FCR for broilers fed 10 and 20% CM in a corn-based diet. Contrary to the results of the current study, Ahmadauli et al. (2008) did not find improvement from adding dietary multicarbohydrase enzyme (xylanase, β-glucanase, celluase,pectinas, proteas and manosidase) for FC and FCR of birds fed different levels of CM in corn-based diets. They found negative effects of inclusion of 40% CM on FCR and FC compared to 20%.

Kermanshahi and Abbasi Pour (2006) did not find improvement to FCR by adding NSP-degrading enzyme for a RSM, but a depression in FC of broilers consuming 30% of RSM was found. This current study did not find any effects of inclusion levels for either type of meal for FC.

Enzyme supplementation significantly affected (P=0.04) mortality in the finisher period (Table 4.21). Birds fed dietary Superzyme -OM ^{TM} had lower levels of mortality compared to the ones without dietary enzyme (1.66±0.44 *a vs* 2.94±0.44 *b.*). The reason for this result is not clear. The average mortality within periods in the corn-based trial was highest in the finisher period. Mortality increased as the birds got older (1.6% in starter, 1.5% in grower and 2.3% in finisher periods). The total mortality calculated as the sum of mortality through the study was 5.4%. The most common reason for mortality in the finisher period was leg abnormalitie (25% of mortalities in finisher period) .

Table 4.21. ANOVA *P*-values for the percent of mortality in broilers fed canola or juncea meals in corn-based diets

Day			
Effect	0-14	15-24	25-35
Meal	0.810	0.760	0.100
Level	0.455	0.987	0.439
Meal × Level	0.655	0.858	0.473
Enzyme	0.753	0.829	0.045
Meal × Enzyme	0.329	0.947	0.123
Level × Enzyme	0.431	0.691	0.061
Meal × Level × Enzyme	0.181	0.112	0.894

4.6.2. Wheat-based trial

ANOVA for BW and BWG (Table 4.22) indicated that the interaction effect of level × enzyme was significant ($P < 0.05$) throughout the study for BW and BWG. The effect of meal × level was significant in the finisher period for BW and BWG. The main effect of meal for the starter and grower periods was significant for BW and BWG.

Table 4.22. ANOVA *P*-values for Body weight ($g \cdot b^{-1}$) and Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) in broilers fed canola or juncea meals in corn-based diets

	Body weight ($g \cdot b^{-1}$)			Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$)		
	Day					
Effect	14	24	35	0-14	15-24	25-35
Meal	0.003	0.003	<.0001	0.004	0.003	<.0001
Level	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Meal × Level	0.436	0.443	0.038	0.423	0.451	0.015
Enzyme	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Meal × Enzyme	0.370	0.403	0.460	0.386	0.407	0.702
Level × Enzyme	0.009	0.001	<.0001	0.009	0.001	0.001
Meal × Level × Enzyme	0.626	0.857	0.739	0.625	0.844	0.572

In both starter and grower phases birds fed CM had higher BW (Table 4.23) and BWG (Table 4.24) than birds fed JM. In the finisher periods, the highest levels of meal inclusion (20 and 30%) resulted in birds consuming CM having higher BW (Table 4.25) and BWG (Table 4.26) compared to JM.

For BW (Table 4.23 and 4.25) the interaction of meal inclusion level and enzyme, showed ranking of the levels is different depending on whether enzyme is included or not. In all levels in starter and 0, 10 and 20% in grower and finisher, feeding Superzyme-OM™ improved BW. BWG data (Tables 4.24 and 4.26) indicated enzyme supplementation did not make a difference for 30% meal inclusion throughout the study. The interaction of meal inclusion level and enzyme in BWG during the finisher period (Table 4.26) indicates that the BWG decreased for 30% inclusion and adding enzyme at this level did not have the beneficial affect for either meal. The improvement in BWG by supplementing with a mixture of Superzyme-OM™ and phytase was reported in a study by Józefiak et al (2010) with wheat-based diets and full fat rapeseed (6% in starter and

12% in grower). These results were attributed to the positive effects of Superzyme-OM^{TM} on the meal which made nutrients more available to the birds.

Table 4.23. Body weight (g·b^{-1}) of broiler chickens fed wheat-based starter diets (0-14) and grower (15-24) containing different levels of juncea meal or canola meal with and without Superzyme-OM ^{TM} (Least square mean ± SE)

Day 14						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme	NO	YES	NO	YES	NO	YES
Levels						
0%	314±9	376±9	304±9	382±9	309±7*e*	379±7*bcd*
10%	367±9	417±9	347±9	408 ±9	357±7*d*	413±7*a*
20%	365±9	416 ±9	353±9	391±9	359±7*d*	404±7*ab*
30%	384±9	398±9	351±9	384±9	359±7*d*	391±7*abc*
Meal	380±3.3*a*		365±3.3*b*			
Day 24						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme	NO	YES	NO	YES	NO	YES
Levels						
0%	902 ±20	1079±20	902±20	1078±20	905±14*e*	1078±14*ab*
10%	999±20	1147±20	969±20	1111±20	984±14*d*	1129±14*a*
20%	968 ±20	1079±20	910. ±20	1049±20	939±14*de*	1064±14*bc*
30%	986±20	1013±20	922±20	989±20	954±14*de*	1001±14*cd*
Meal	1023±7*a*		991±7*b*			

[a- d] Means ± SEM with different letters within interaction means and meals are significantly different (α=0.05)

Table 4.24. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed wheat-based starter diets (0-14) and grower (15-24) containing different levels of canola meal or juncea meal with and without Superzyme-OM ™ (least square mean ± SE)

0-14 day						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme Levels	NO	YES	NO	YES	NO	YES
0%	19±1	24±1	19±1	24±1	19±1*e*	24±1*bcd*
10%	23±1	27±1	23±1	26±1	22±1*d*	26±1*a*
20%	23±1	27±1	22±1	25±1	22±1*d*	26±1*ab*
30%	24±1	25±1	22±1	24±1	23±1*cd*	25±1*abc*
Meal	24±0.24*a*		23±0.24*b*			
15-24 day						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme Levels	NO	YES	NO	YES	NO	YES
0%	89±1	105±1	88±1	105±1	89±1 *e*	105±1*ab*
10%	98±1	112±1	95±1	109±1	96±1*d*	110±1*a*
20%	95±1	105±1	89±1	102±1	92±1*de*	104±1*bc*
30%	96±1	99±1	90±1	96±1	93±1*de*	98±1*cd*
Meal	100±1*a*		100±1*b*			

[a-d] Means ± SEM with different letters within the same columns are significantly different (α=0.05)

Table 4.25. Body weight ($g \cdot b^{-1}$) of broiler chickens fed wheat-based finisher diets (day 35) containing different levels of juncea meal or canola meal with and without Superzyme– OM ™ (least square mean ± SE)

Day 35						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme Levels	NO	YES	NO	YES	NO	YES
0%	1944 ±33	2214±33	1915±33	2237±33	1929 ±23*cd*	2226±23*a*
10%	2033 ±33	2268±33	1965±33	2165±33	1999 ±23*bc*	2217±23*a*
20%	1909 ±33	2132±33	1765±33	2005±33	1837 ±23*de*	2068±23*b*
30%	1861±33	1885±33	1721±33	1806±33	1791 ±23*e*	1846 ±23*de*

Meal ×Level	Canola meal	Juncea meal
0%	2079±23*ab*	2076±23*ab*
10%	2151±23*a*	2065 ±23*ab*
20%	2020±23*b*	1885±23*c*
30%	1873±23*c*	1764±23*d*

[a-e] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

Table 4.26. Body weight gain ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed wheat-based finisher diets (24-35) fed different levels of juncea meal and canola meal with and without Superzyme– OM ^{TM} (least square mean ± SE)

25-35 day						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme	NO	YES	NO	YES	NO	YES
Levels						
0%	94±2	103±2	92±2	105±2	93±1*b*	104±1*a*
10%	94±2	102±2	90±2	96±2	92±1*b*	99±1*a*
20%	86±2	96±2	78±2	87±2	82±1*c*	91±1*b*
30%	80±2	79±2	73±2	74±2	76±1*c*	77±1*c*
Meal ×Level	Canola meal		Juncea meal			
0%	99±1*a*		99±1*a*			
10%	98±1*a*		93±1*ab*			
20%	91±1*b*		82±1*c*			
30%	79±1*c*		73±1*d*			

[a-d] Means ± SEM with different letters within the interaction means are significantly different (α=0.05)

From the interaction effect of meal by levels in BWG in the finisher period (Table 4.26), there was a significantly lower BWG with meal inclusion levels of 20 and 30 % compared with the 0% level and 10% of either CM or JM. McNeill et al. (2004) repoeted similar results using a wheat-based diet with RSM. Olomu et al (1975a) also reported depression in BWG in broilers by feeding above 20% of rapeseed in a wheat-based diet. Based on reduced BW and BWG at 30% of CM inclusion, not more than 20% of meal inclusion, preferably CM, in grower, and not more than 10% of either meals in finisher is recommended, the enzyme inclusion improved the BW and BWG of birds throughout the study except at 30%.

ANOVA for FC and FCR (Table 4.27) indicated the significant effect of meal inclusion level and enzyme in the grower period and the main effect of enzyme in starter and grower periods on FC. The effects of type of meal and meal inclusion level were significant in the grower period for FCR**,** the main effect of meal and meal inclusion level

were significant in starter and finisher periods for FCR. The main effect of enzyme was significant for starter in FC.

Table 4.27. ANOVA *P*-values for feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) and feed conversion ratio in broilers fed canola or juncea meals in wheat-based diets

	Feed consumption ($g.b^{-1}$ d^{-1})			Feed Conversion Ratio		
Day						
Effect	0-14	15-24	25-35	0.14	15.24	25-35
Meal	0.554	0.143	0.407	0.000	0.002	0.000
Level	0.175	0.089	0.769	0.001	<.0001	<.0001
Meal × Level	0.094	0.058	0.587	0.268	0.002	0.186
Enzyme	0.002	0.000	0.010	<.0001	0.230	0.495
Meal × Enzyme	0.423	0.210	0.132	0.276	0.327	0.470
Level × Enzyme	0.084	0.030	0.943	0.378	0.161	0.847
Meal × Level× Enzyme	0.164	0.366	0.178	0.686	0.067	0.715

Based on FC data in starter (Table 4.28) and grower (Table 4.29) periods , birds fed enzyme supplemented diets consumed more feed.The interaction of meal inclusion level and enzyme in the grower phase shows that only birds consuming the control diet consumed more feed , as a result of dietary enzyme inclusion. McNeill et al (2004) found a reduction in FC by feeding solvent-extracted RSM in wheat-based diets to broilers with no adverse effect of meal inclusion level. Enzyme supplementation increased FC in the finisher phase (Table 4.30). There was no adverse effect of CM or JM in the current study on FC. Therefore, antinutritional factors in CM and JM did not affect consumption in the broilers.

Table 4.28. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) of broiler chickens fed wheat-based starter diets (0-14) containing different levels of canola meal and juncea meal with and without Superzyme-OM™ (least square mean ± SE)

0-14 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	35±1	34±1	31±1	34±1
10%	31±1	35±1	33±1	35±1
20%	32±1	35±1	32±1	35±1
30%	34±1	34±1	35±1	35±1
Enzyme	NO		YES	
	33±0 *b*		35±0. *a*	

[a-b] Means ± SEM with different letters for enzyme are significantly different (α=0.05)

Table 4.29. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) by broiler chickens fed wheat-based grower diets (15-24) containing different levels of canola meal and juncea meal with and without Superzyme-OM ™ (least square mean ± SE)

15-24 day						
Meal	Canola meal		Juncea meal		Level × Enzyme	
Enzyme	NO	YES	NO	YES	NO	YES
Levels						
0%	101±5	115±5	101±5	120±5	95±4*b*	118±4*a*
10%	109±5	120±5	107±5	118±5	108±4*ab*	119±4*a*
20%	105±5	114±5	112±5	116±5	109±4*ab*	115±4*a*
30%	104-±5	112±5	119±5	128±5	116±4*a*	116±4*a*

[a-b] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

Table 4.30. Feed consumption ($g \cdot b^{-1} \cdot d^{-1}$) by broiler chickens fed wheat-based finisher diets (25-35) containing different levels of juncea meal and canola meal with and without Superzyme-OM™ (least square mean ± SE)

25-35 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	172±9	181±9	173±9	193±9
10%	175 ±9	191±9	178±9	182±9
20%	170±9	181±9	164±9	185±9
30%	180±9	166±9	169±9	202±9
Enzyme	NO		YES	
	177±3*b*		185±3*a*	

[a-b] Means ± SEM with different letters for enzyme are significantly different (α=0.05)

In the starter (Table 4.31) period, feeding CM compared to JM, improved FCR. There was an interaction of type of meal inclusion by level inclusion in the grower period (Table 4.32) on FCR. The highest inclusion level of 30% for CM resulted in improved FCR compared to JM. The glucosinolate profile of JM compared to CM maybe the reason for this (Slominski et al. 2012). The same pattern that was observed for the starter period repeated in the finisher period (Table 4.33). Inclusion of enzyme had a beneficial effect on FCR in the starter period (Table 4.31). In the starter period (Table 4.31), FCR improved for diets with the test ingredients at 10 to 20%. This finding is contrary to Olomu et al (1975a) who found adverse effects of feeding rapeseed meal above 20% in broilers fed wheat-based diets. The results of the current study are in agreement with a study by Józefiak et al (2010) who found that supplementing the mixture of Superzyme-OM™ and phytase in wheat-based diets, improved FCR when full fat rapeseed meal was included at 6 and 12% of the diet for the grower and finisher periods .

Table 4.31. Feed conversion ratio of broiler chickens fed wheat-based starter diets (0-14) containing different levels of juncea meal and canola meal with and without Superzyme– OM ™ (least square mean ± SE)

0-14 day					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	1.6±0.1	1.5±0.1	1.7±0.1	1.4±0.1	1.5±0.0*b*
10%	1.4±0.1	1.3±0.1	1.5±0.1	1.4±0.1	1.4±0.0*a*
20%	1.4±0.1	1.3±0.1	1.5±0.1	1.4±0.1	1.4±0.0*a*
30%	1.5±0.1	1.3±0.1	1.6±0.1	1.5±0.1	1.5±0.0*ab*
Meal					
	1.4±0.0*a*		1.5±0.0*b*		
Enzyme	NO		YES		
	1.5±0.0*b*		1.4±0.0*a*		

[a-c] Means ± SEM with different letters for levels and enzyme are significantly different (α=0.05)

Table 4.32. Feed conversion ratio of broiler chickens fed wheat-based grower diets (15-24) containing different levels of juncea meal and canola meal with and without Superzyme– OM ™ (least square mean ± SE)

15-24 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.7±0.1	1.64±0.1	1.5±0.1	1.7±0.1
10%	1.7±0.1	1.65±0.1	1.7±0.1	1.7±0.1
20%	1.7±0.1	1.72±0.1	2.0±0.1	1.8±0.1
30%	1.9±0.1	1.70±0.1	2.1±0.1	2.1±0.1
Meal × Level				
0%	1.7±0.1*c*		1.6±0.1*c*	
10%	1.7±0.1*bc*		1.7±0.1*bc*	
20%	1.7±0.1*bc*		1.9±0.1*ab*	
30%	1.8±0.1*bc*		2.1±0.1*a*	

[a-c] Means ± SEM with different letters within the interaction means are significantly different (α=0.05)

Table 4.33. Feed Conversion Ratio of broiler chickens fed wheat-based finisher diets (25-35) containing different levels of canola meal and juncea meal with and without Superzyme– OM ™ (least square mean ± SE)

25-35 day					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	1.8±0.1	1.7±0.1	1.9±0.1	1.8±0.1	1.8±0.0*a*
10%	1.9±0.1	1.9±0.1	2.0±0.1	1.9±0.1	1.9±0.0*bc*
20%	2.0±0.1	1.9±0.1	2.1±0.1	2.1±0.1	2.0±0.0*c*
30%	2.1±0.1	2.1±0.1	2.3±0.1	2.4±0.1	2.3±0.0*c*
Meal	1.9±0.0*a*		2.1±0.0*b*		

[a-c] Means ± SEM with different letters for levels and meals are significantly different (α=0.05)

ANOVA for mortality (Table 4.34) indicated the significant effects of meal × enzyme in starter period. All other treatments did not have any effects on mortality in different periods.

Table 4.34. ANOVA *P*-value for mortality in broilers fed canola or juncea meals in whaer-based diets

Day			
Effect	0-14	15-24	25-35
Meal	0.209	0.061	0.327
Level	0.275	0.320	0.560
Meal × Level	0.133	0.613	0.601
Enzyme	0.205	0.308	0.359
Meal × Enzyme	0.011	0.377	0.370
Level × Enzyme	0.246	0.189	0.330
Meal × Level × Enzyme	0.788	0.355	0.809

There were negative effects of enzyme by meal on mortalities in starter period. Birds fed JM without dietary supplementation of Superzyme-OM^{TM} had the highest percent of mortality (Table 4.35). The reason for this result might be attributed to the toxic profile of glucosinolate to broilers especially in starter phase when birds are not able to handle toxic materials as well as older birds. As a result, substituting JM in bird diets with SBM in starter period is not recommended as it affected mortality rate. Further studies are required to make the final conclusions as the glucosinolate content of JM is still in the canola definition (less than 30 μmol·g^{-1}). The glucosinolate content of JM which was fed in this experiment was calculated to be 3.8 μmol·g-^{1} in the 30% of JM inclusion. The major cause of mortality in starter period was associated with dehydration. The high percentage of 5.7 of mortality in 30% of JM inclusion without enzyme supplementation might be only associated to high numbers of birds died from dehydration in this period. No significant effects of treatments were found on mortality for grower and finisher periods (Table 4.35).

The average mortality within periods in wheat -based was 1.2 % in the starter, 2.1% in the grower and 1.3% in the finisher periods. The total mortality through the study was 4.6 %. The most common reason for mortality in the grower period with highest rate of mortality was associated with liver problems (swollen livers).

Table 4.35. Mortality (%) by broiler chickens fed wheat-based starter (0-14), grower (15-24) and finisher (25-35) diets containing different levels of juncea meal and canola meal with and without Superzyme– OM ^{TM} (least square mean ± SE)

0-14 day				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.7±1.1	0.0±1.1	1.9±1.1	0.0±1.1
10%	0.0±1.1	2.5±1.1	0.0±1.1	0.0±1.1
20%	0.6±1.1	1.9±1.1	2.5±1.1	0.6±1.1
30%	0.6±1.1	0.6±1.1	5.7±1.1	1.3±1.1
Enzyme × Meal	Canola meal		Juncea meal	
	NO	YES	NO	YES
	0.5±0.6*b*	1.3±0.6*ab*	2.7±0.6*a*	0.5±0.6*b*

[a-b] Means ± SEM with different letters within the same row are significantly different (α=0.05)

4.7. General discussion

Superzyme-OM^{TM} is beneficial in terms of making the nutrients in meals more available for broilers by affecting different components of fiber in meals. To sum up, the effect of Superzyme- OM^{TM} on meal cell wall degradation, more nutrients was available to broilers and the birds performed better. Adding dietary enzyme to their diets helped the birds overcome the negative effects of fiber in JM or CM, as a result, the reported data indicates the benefit of supplementing Superzyme-OM^{TM} in broiler diets containing either CM or JM, because of the improvements in all growth parameters (BW, BWG, FC and FCR) in both trials. In this study, in some cases, the highest levels of meal inclusion had adverse effects on growth performance, despite the fact that all the meals in the current study had low levels of glucosinolates (0.81,1.62 and 2.43 μmole·g^{-1} for 0,10 , 20 and 30% of CM and 1.3,2.5 and 3.5 μmole·g^{-1} for 0,10,20 and 30% of JM). It is possible that the anti-nutritional factors of glucosinolate in higher meal inclusion levels, is responsible for poor growth performance of the birds. More study is required, to investigate the effects of glucosinolates profiles of JM and CM on the broilers performance. Some of the differences in the performance of birds in corn and wheat based trials can be due to different NSP content of corn and wheat and the ability of Superzyme-OM^{TM} to break them down.

4.8. Conclusions

Based on the results of the current study, these recommendations are suggested for inclusion levels and Superzyme-OM^{TM} supplementations in different growth periods of broilers. In corn-based diets, 30% CM with dietary enzyme for starter and grower and 10% of CM with enzyme for finisher are recommended. In wheat- based trial, 30% of CM with enzyme in starter, 20% of CM in grower and 20% of CM with enzyme in finisher are recommended. These levels are higher than the previous recommendations of 10% in starter period (CCC 2009). In case of including JM in the diets, 30% meal inclusion in both corn- and wheat-based diets for starter phase , 30% and 20% of JM in

grower phase for corn and wheat-based diets respectively and 10% in finisher phase for both corn- and wheat based diets are recommended.

CHAPTER 5: THE EFFECT OF DIFFERENT LEVEL OF CANOLA MEAL (BRASSICA NAPUS) AND JUNCEA MEAL (BRASSICA JUNCEA) IN BROILER DIETS SUPPLEMENTED WITHOUT OR WITH A DIETARY ENZYME COCKTAIL: CARCASS COMPOSITION

5.1. Abstract

A multicarbohydrase enzyme (Superzyme-OM^{TM}) was fed to broilers with different inclusion levels of CM or JM inclusion (0, 10, 20 or 30%) for 35 days in two trials based on either, corn or wheat. The effects of meals, inclusion levels and enzyme on edible carcass compositions were studied. In corn-based diets Superzyme-OM^{TM} increased the percentage of DM (P<0.01), fat (P<0.01) and protein (P<0.01) in carcasses. In wheat-based diets, Superzyme-OM^{TM} increased the percentage of DM (P =0.04). The carcasses of the birds fed JM had higher (P=0.04) percent ash compared to birds fed CM (5.6 ± 0.11 vs. 5.3 ± 0.11 respectively). The effect of inclusion level was significant (P=0.03) for the percentage of protein in carcasses. Birds in the control group had higher protein compared to 30% JM or CM inclusion (51.3±0.79 % vs. 47.9±0.82%).

Key words: *Carcass composition, Broilers, Canola, Juncea,* Superzyme- OM ^{TM}

5.2. Introduction

Carcass chemical composition, such as protein, fat, ash and dry matter are important economically, because they are directly related to the efficiency in meat production (Nascimento Nunes et al .2011)

As poultry breeders improve the growth rate of broilers, excess fat in the edible parts of carcasses is more likely to increase. This is a result of the relationship between growth rate and the fat deposition in the body (Griffiths et al. 1977). Currently, consumers awareness of increasing dietary fat has been recognized. As a result, the effect of diets on carcass composition should be taken into account. Proper utilization of nutrients by broilers should be considered, in order to have a high amount of edible protein and low levels of fat in carcasses. CM or JM can be used as a source of nutrients in poultry diets that maybe enhanced with supplementing dietary enzymes. There is lack

of information on the effects of graded levels of CM or JM in broiler diets with multicarbohydrase enzyme supplementation on chemical composition of the edible parts of carcasses.

5.3. Objectives

The objectives were to determine the effects of various levels of CM or JM, on carcass composition with and without multicarbohydrase enzyme (Superzyme-OM^{TM}) on corn or wheat- based diets fed to broilers.

5.4. Hypotheses

It is hypothesized that different inclusion levels of CM or JM in broiler diets will not result in any significant changes in carcass composition. It is expected to see slightly higher percentage of fat and protein in carcasses of the birds supplemented with Superzyme- OM$^{TM.}$, resulting from increased nutrient availability of feed ingredients.

5.5. Materials and methods

5.5.1. Sample collections and carcass composition analysis

On day 35 of Trial 1 and Trial 2, two birds per pen were randomly selected and commercially slaughtered for whole carcass analysis after 12 hours of fasting. The eviscerated carcasses were identified by wing tags, placed in plastic bags and stored at -20 ° C until being processed for carcass composition. The carcasses were thawed at 4 °C and individually ground using a commercial meat grinder (Hobart Meat grinder, Model 4246S, Ohio, USA) using a plate with 3 mm holes, and weighed. The ground carcass were frozen again at -20°C untill freeze dried. Ground carcasses, were weighed prior to and after freeze drying to calculate DM (Method 935. 29 (AOAC 2005)). representation from the freeze dried carcasses were ground in a coffee grinder (Lancaster Coffee Grinder, 43-1964-8). Duplicate samples were analyzed for ash by muffle furnace at

525°C, crude protein (CP) (% N×6.25) using a Leco $^{FP-528®}$ Nitrogen analyzer (Leco Corporation, St Joseph, MI) method 990.03 (AOAC 2005) and crude fat (CF) by ether extraction method using Ankom$^{XT15®}$ (Ankom Technology Corporation, Macedon, USA) and petroleum ether as the solvent.

5.5.2. Statistical analysis

Carcass ash, DM, CP and CF were subjected to analysis of variance using the Proc Mixed procedure of the SAS v.9.3 (SAS Institute Inc., Cary.NC) (Littell et al. 1996). The following model was employed for statistical analysis:

$Y_{ijkl}=\mu+\rho_i+\alpha_j+\beta_k+\gamma_{l+}\alpha\beta_{jk}+\alpha\gamma_{jl}+\beta\gamma_{kl}+\alpha\beta\gamma_{jkl}+\varepsilon_{ijkl}$

Where Y_{ijkl} is the variable of interest; μ is the overall mean; ρ_i is the effect of the i^{th} block (i=1-4); α_j is the effect of j^{th} meal(j=1-2); β_k is the k^{th} level of inclusion (k=1-4); γ_l is the effect of l^{th} enzyme (l=1-2); $\alpha\beta_{jk}$ is the effect of the interaction between meal and the level of inclusion, $\alpha\gamma_{jl}$ is the effect of the interaction between meal and enzyme, $\beta\gamma_{kl}$ is the effect of interaction between level of inclusion and enzyme; $\alpha\beta\gamma_{jkl}$ is the effect of interaction between meal, level and enzyme and ε_{ijkl} is the random effect of error. Significant main or interaction effects (α=0.05) were separated using Tukey-Kramer test (Littell et al. 1996) to differentiate the means.

5.6. Results and discussion

5.6.1. Corn-based trial

ANOVA for the percentage of DM, ash, fat and protein are reported in Table 5.1. The enzyme mixture influenced dry matter (P<0.01) and fat percentage (P<0.01) and protein ($P=0.01$) of carcasses of broilers fed a corn-based diet. There were no treatment effects of levels and meal on carcass composition.

Table 5.1. ANOVA *P*-values for carcass dry matter, ash, fat and protein (%) in broilers fed canola or juncea meals in corn-based diets

	Dry matter	Ash	Fat	Protein
Effect				
Meal	0.679	0.187	0.878	0.992
Level	0.552	0.257	0.069	0.103
Meal × Level	0.352	0.411	0.330	0.185
Enzyme	0.000	0.931	0.001	0.011
Meal × Enzyme	0.499	0.160	0.431	0.509
Level × Enzyme	0.959	0.266	0.279	0.327
Meal × Level × Enzyme	0.716	0.649	0.516	0.777

Superzyme-OM^{TM} supplementation increased the percentage of DM (Table 5.2), fat and decreased protein of carcasses (Table 5.3). Increase fat percentage by supplementing with enzymes was accompanied by increasing BW of birds when Superzyme- OM ^{TM} was added in the diets (Tables 4.8, 4.10 and 4.11). Possibly, the birds had increased access to more energy from the meals resulting in an increase in the percentage of fat in the carcass. Jackson et al (1982) reported increasing fat content of broiler carcasses with increased weight gain by the birds. No significant effects of treatments were observed for the percentage of ash in the carcasses (Table 5.2).

Hajati (2010) did not find that multicarbohydrase enzyme supplementation affected carcass composition in broilers fed a blended corn-soybean meal in a wheat-based diet. The percentage of carcass ash in our study (5.15 ±0.3) was lower than the average (9.11 ± 0.50) of Hajati (2010). The percentage of DM in the current study (32.4±0.2) was higher than that found by Hajati (2010) (28.48±0.52) and the percentage of fat (40±0.6) and protein (51.25±0.5) were in the same ranges as this study (40.75±0.58 and 50.07±0.57 respectively). The differences between the two trials can be associated with the different breeds of broilers (Cobb 500 vs Ross 508) and slaughter age (44 days) of the birds and different diets (corn-soybean meal-wheat diets) that were fed.

Table 5.2. Dry matter and ash percentage (on dry matter basis) in carcasses of broilers fed corn-based diets with different levels of canola meal or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Dry matter %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	31.8±0.6	33.0±0.6	31.4±0.6	32.3±0.6
10%	31.6±0.6	33.0±0.6	32.3±0.6	33.9±0.6
20%	31.2±0.6	33.4±0.6	31.9±0.6	32.4±0.6
30%	32.3±0.6	33.6±0.6	31.7±0.6	32.8±0.6
Enzyme	NO		YES	
	31.8±0.2*b*		33.0±0.2*a*	
Ash %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	5.2±0.3	5.3±0.3	5.0±0.3	5.1±0.3
10%	5.2±0.3	4.7±0.3	5.1±0.3	5.3±0.3
20%	5.3±0.3	5.0±0.3	5.7±0.3	5.4±0.3
30%	4.9±0.3	4.9±0.3	4.8±0.3	5.5±0.3

[a-b] Means ± SEM with different postscripts for enzyme in dry matter content of carcass are significantly different (α= 0.05)

Table 5.3. Fat and protein percentage (on dry matter basis) of carcasses of broilers fed corn-based diets with different levels of canola meal or juncea meal with and without Superzyme-OM™(least square mean ±SE)

Fat %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	37.0±1.6	41.1±1.6	34.9±1.6	41.2±1.6
10%	38.1±1.6	40.4±1.6	40.5±1.6	42.3±1.6
20%	37.9±1.6	41.3±1.6	39.2±1.6	40.3±1.6
30%	40.8±1.6	44.0±1.6	41.1±1.6	40.1±1.6
Enzyme	NO		YES	
	38.7±0.6*b*		41.3±0.6*a*	
Protein %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	54.1±1.5	50.1±1.5	55.7±1.5	50.8±1.5
10%	53.5±1.5	51.2±1.5	50.3±1.5	49.6±1.5
20%	52.8±1.5	50.3±1.5	50.5±1.5	51.3±1.5
30%	49.6±1.5	48.3±1.5	51.6±1.5	50.2±1.5
Enzyme	NO		YES	
	52.3±0.5*a*		50.2 ±0.5*b*	

[a-b] Means ± SEM with different postscripts for enzyme are significantly different (α= 0.05)

5.6.2. Wheat-based trial

ANOVA (Table 5.4), indicated the main effects of meal on the percentage of ash (*P*=0.04), the effect of meal inclusion level (*P*=0.03) on the percentage of protein and the effect of enzyme inclusion (*P*=0.04) on the percentage of dry matter in carcasses of broilers fed wheat-based diets.

Table 5.4. ANOVA *P*-values for carcass dry matter, ash, fat and protein (%) in broilers fed canola or juncea meals in wheat -based diets

	Dry matter	Ash	Fat	Protein
Effect				
Meal	0.837	0.042	0.081	0.311
Level	0.544	0.278	0.274	0.029
Meal × level	0.760	0.114	0.905	0.421
Enzyme	0.039	0.067	0.059	0.226
Meal × Enzyme	0.377	0.279	0.568	0.936
Level × Enzyme	0.476	0.996	0.961	0.972
Meal × Level × Enzyme	0.306	0.527	0.582	0.789

Superzyme -OM^{TM} supplementation increased the percentage of DM (Table 5.5), of carcasses. Birds with diets supplemented with JM had higher percentage of ash in the carcasses than the birds fed CM (Table 5.5). No significant effects of treatments were observed for the percentage of fat (Table 5.5). The birds in control groups (Table 5.6) had higher percentage of protein ($P \leq 0.05$), compared with the highest meal inclusion of 30%. This is supported by BW data on day 35 (Table 4.25), indicated heavier birds with 0% meal inclusion compared to 30% .This result was opposite to Olomu et al. (1975a), who found higher protein percentage in broilers carcasses fed 20% of raw RS compared to the wheat-based control diets. They found 30 % of rapeseed meal increased the protein percentage and decreased percentage of fat in carcasses.

The results of this study can be compared to Hajati (2010) who fed broilers with ablended diet of corn-soybean meal and wheat. Hajati (2010) found higher DM and lower ash percentage compared to our study (28.48 ±0.52 and 9.11±0.50 respectively), and the same range of fat and protein percentage (40.75±0.58 and 50.07±0.57 respectively).

It is suggested to conduct more research to investigate the effects of different levels of Superzyme- OM^{TM} on the percentage of fat in carcasses. Perhaps , if lower inclusion levels of enzyme can reduce the fat content of carcasses while still, having a positive influence on the growth performance of birds

Table 5.5. Dry matter, fat and ash percentage (on dry matter basis) in carcasses of broilers fed wheat-based diets with different levels of canola meal or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Dry matter %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	30.2±1.19	32.1±1.19	30.8±1.19	30.9±1.19
10%	30.7±1.19	30.7±1.19	30.8±1.19	31.2±1.38
20%	32.0±1.38	32.7±1.19	30. 8±1.19	32.8±1.19
30%	28.4±1.19	33.5±1.19	32.0±1.19	32.4±1.38
Enzyme	NO		YES	
	30.74±0.43*b*		32.05±0.44*a*	
Fat %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	40.6±1.63	41.7±1.63	39.1±1.63	41.2±1.63
10%	42.3±1.63	41.6±1.63	39.4±1.63	42.7±1.82
20%	43.2±1.63	43.7±1.63	40.0±1.63	42.0±1.63
30%	41.9±1.63	45.6±1.63	41.5±1.63	42.9±1.88
Ash %				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	4.8±0.30	5.2±0.30	5.4±0.30	5.6±0.30
10%	5.1±0.30	5.9±0.30	5.3±0.30	5.2±0.35
20%	4.9±0.35	5.4±0.30	5.9±0.30	5.5±0.30
30%	5.5±0.30	5.6±0.30	5.5±0.30	5.1±0.30
Meal	5.3±0.11*b*		5.6±0.11*a*	

[a-b] Means ± SEM with different postscripts for enzyme in dry matter and ash contents of carcassed are significantly different (α= 0.05)

Table 5.6. Protein percentage (on dry matter basis) in carcasses percentage (on dry matter basis) of broilers fed wheat-based diets with different levels of canola meal or juncea meal with and without Superzyme -OM^{TM} (least square mean ±SE)

Protein %					
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	51.1±1.57	50.7±1.57	51.8±1.57	51.5±1.57	51.3±0.79*a*
10%	51.4±1.57	50.6±1.57	50.5±1.57	48.7±1.82	50.3±0.82*ab*
20%	48.6±1.82	48.1±1.57	51.2±1.57	49.2±1.57	49.3±0.82*ab*
30%	48.1±1.57	45.5±1.57	48.8±1.57	49.1±1.82	47.9±0.82*b*

[a-b] Means ± SEM with different postscripts for levels are significantly different (α= 0.05)

Reported data on carcass compositions in broilers (Hajat 2010 and Chambers and Fortin 1984) confirms that the carcasses chemical compositions of the birds in our study were in acceptable ranges (Table 5.7). Different variables, such as breed, age, sex, nutrition, can affect the chemical characteristics of carcasses (Bogosavljevic-Boskovic et al.2010)

Table 5.7. The averages of fat, protein , ash and dry matter in carcasses compared to the literature data for broilers

	Chambers and Fortin (1984)	Hajati Corn-wheat (2010)	Corn- based trial (2013)	Wheat-based trial (2013)
Traits				
Fat %	41.5	40.8	40.0	41.8
Protein %	50.0	50.2	51.2	49.7
Ash%	7.5	9.11	5.15	5.4
Dry matter %	35.1	28.5	32.4	31.4

5.7. Conclusions

In corn-based diets, when Superzyme-OM^{TM} was supplemented the content of fat in carcasses increased while the enzyme did not change the percentage of fat in broilers fed wheat-based diets. The percentage of carcass protein was higher in the birds fed 30% of wheat-based diets compared to 30% of CM or JM inclusions. Based on the results of carcass compositions of birds, it is recommended, to include up to 20% of either JM or CM in wheat-based diets for broilers.

CHAPTER 6: THE EFFECT OF DIFFERENT LEVEL OF CANOLA MEAL (BRASSICA NAPUS) AND JUNCEA MEAL (BRASSICA JUNCEA) IN BROILER DIETS SUPPLEMENTED WITHOUT OR WITH A DIETARY ENZYME COCKTAIL: LIVER PARAMETERS

6.1 Abstract

The glucosinolates in canola and juncea meal can have negative effects on broiler livers, hypertrophy and hemorrhage. In this study, the effect of different inclusion levels of canola and juncea meals, (0, 10, 20 and 30 %), with and without Superzyme-OM^{TM}, in corn- and wheat-based trials, was examined. The weight, visual scoring of colour and texture, as well as colorimeter data were measured for two birds from each pen at the end of each trial. In the corn-based trial, feeding canola meal resulted in increased (P=0.023) liver weight, compared to juncea meal (44.93 g vs. 41.89 g), while Superzyme-OM^{TM} supplementation increased (P=0.049) liver weight (44.71 g vs. 42.11 g). In wheat-based diets, liver weight was reduced (P=0.005) when 30% (40.48 g) inclusion was provided, compared to 0 and 10 % (45.51 g and 47.02 g, respectively). Birds fed dietary enzyme had larger (P=0.041) livers (45.43 g) than those without enzyme (42.74 g). No significant treatment effects were observed on visual scoring and colorimeter data ($P\geq$0.05) except for lightness (L^* score) in wheat-based diets. L^* score was higher (P=0.048) when JM was fed compared to CM (25.68 vs. 24.60 respectively). There was no treatment effect on visual scoring in either trial ($P\geq$0.05). Overall, enzyme inclusion in both trials resulted in higher ($P\leq$0.05) liver weight, which can be associated with higher metabolic activity due to additional nutrient release from the digesta. In general, canola meal and juncea meal did not have negative effects on livers in both trials. Further study is needed to relate the percentage of fat in livers to colour.

Key words: *Liver, Broilers, Canola, Juncea,* Superzyme – OM^{TM}

6.2 Introduction

Almost all fatty acid synthesis in avian species is carried out in the liver and only a very little in adipose tissue; as a result, the metabolic activity of the liver is high (Butler 1976). The lipids in the liver come from different sources , including, synthesis from carbohydrates, adipose tissues and dietary fats. Also, a variety of dietary and toxic substances can cause the accumulation of fat in the liver (Butler 1976).

High glucosinolate levels in layer hens have been implicated in hemorrhagic syndrome in several studies (Khajali and Slominski 2012). Fatty liver hemorrhagic syndrome (FLHS), a metabolic condition found in laying hens, results from accumulation of liver fat in commercial flocks (Couch 1956). Lipid synthesis in laying hens is higher than in broilers due to increased production of triglyceride and secretion of very low-density lipoprotein for egg production. Yamashiro et al. (1977) reported the adverse effect of high glucosinolate levels on broilers; livers, such as haemorrhagic lesions and hepatocytic necrosis, when they were fed a diet supplemented with 50% of either RSM or rapeseed full fat seed in corn-based diets.

6.3 Objectives

The objectives of this trial were to determine the effects of different levels of canola meal or juncea meal, with and without Superzyme-OM^{TM} supplementation, on liver characteristics in corn and wheat based diets.

6.4 Hypothesis

It is hypothesized that birds receiving high inclusion levels of either CM or JM in their feed will develop liver diseases due to the negative effects of the glucosinolate content of the meals on the livers.

6.5 Materials and methods

6.5.1 Sample collection and analysis

On day 35 of corn- and wheat- based trials, livers were taken from four slaughtered birds from each pen and were weighed on a Sartorius type Universal U6100S balance, accurate to 0.01 g of readability. The texture score of livers were determined visually by one person , with 1 as firm and 2 as friable. The colour of livers, post mortem, was determined visually using the scale where, 1 was dark and 2 was pale. Immediately following visual scoring, the hand held Hunter Lab Miniscan XE^{TM} (HunterLab. 2008) (CIE, illuminant D6, observer angle 10°) colorimeter, with a trichromatic system, was used to evaluate colour. Calculations were done automatically by the computer. D65/10° set up was used as the illuminant/observer combination. The system uses L* (lightness) in which 0 is black, 100 is white. a*(red-green) has positive values as red and negative values as green and 0 is neutral and b* (blue-yellow), has positive values as yellow, negative values as blue and 0 is neutral (Figure 1.5). This colour system is beneficial, as it has a uniform colour scale and provides the opportunity to compare colours among samples (Marcus 1998).

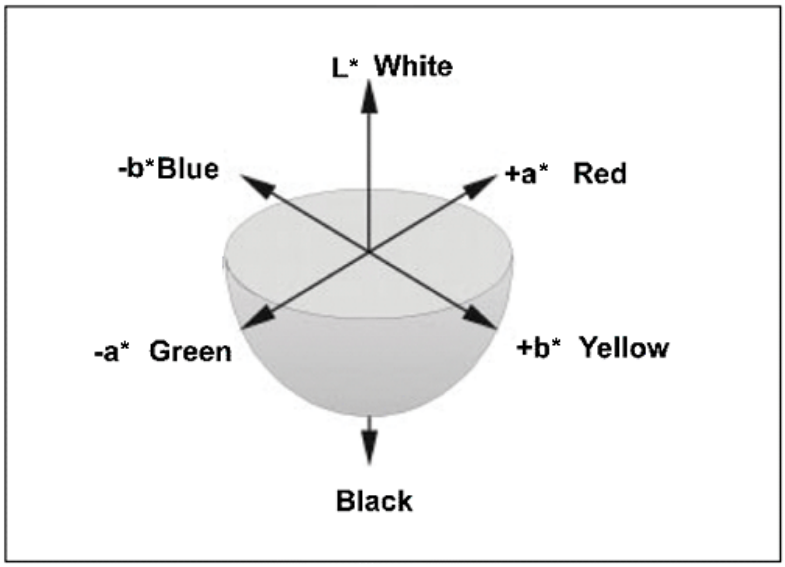

Figure1.5: Colour score system from Heildelberg (1999)

6.5.2 Statistical analysis

Liver weight and colorimeter data were analyzed using proc Mixed ANOVA in SAS 9.3 (SAS Institute Inc., Cary, NC. 1996). Significant main or interaction effects (α=0.05) were analyzed using Tukey-Kramer (Littell et al. 1996) test to differentiate the means.

Categorical data of visual scoring of colour and texture were analyzed using the Chi-square test, using 2-way contingency tables in Minitab®16 (Minitab, Inc., USA. 2010) statistical analysis software.

Statistical Model for Liver Measurement Analysis**:**

$$Y_{ijkl}=\mu+\rho_i+\alpha_j+\beta_k+\gamma_{l+}\alpha\beta_{jk}+\alpha\gamma_{jl}+\beta\gamma_{kl}+\alpha\beta\gamma_{jkl}+\varepsilon_{ijkl}$$

Where Y_{ijkl} is the response of the parameter being measured; μ is the overall mean; ρ_i is the effect of the i^{th} block (i=1-4); α_j is the effect of j^{th} meal (j=1-2); β_k is the k^{th} level of inclusion (k=1-4); γ_l is the effect of l^{th} enzyme (l=1-2); $\alpha\beta_{jk}$ is the effect of the interaction between meal and the level of inclusion, $\alpha\gamma_{jl}$ is the effect of the interaction between meal and enzyme, $\beta\gamma_{kl}$ is the effect of interaction between level of inclusion and enzyme; $\alpha\beta\gamma_{jkl}$ is the effect of interaction between meal, level and enzyme and ε_{ijkl} is the random effect of error.

6.6. Results and discussion

6.6.1 Corn-based trial

Results of ANOVA analysis for liver weights indicated the significant main effects of enzyme and meal on liver weight (Table 6.1). Colorimeter scores (Table 6.2) showed that there was no difference ($P \geq 0.05$) in varying levels of the two meals, with or without the enzyme, in a^*, b^* and L^* colour scores. In general, all of L^* and a^* scores in the control diets were lower and the b^* scores were higher, compared to the study by Trampel et al. (2005) (L*=54.41, a* =15.32 and b* = 7.44) and Northcutt et al. (1997)

(L*=35.22, a* =17.78 and b* =7.45). The higher L* scores in these studies, which showed lighter and fattier livers compared to the present study, might be due to the slaughter age, as the birds were older (48 and 43 days, respectively) in the two previous trials, compared to our study. Another factor can be the effects of feed withdrawal time on liver colour. In Trampel et al. (2005) study, broilers with an access to feed prior to slaughter had lighter colour compared the ones that experienced fasting. Therefore, darker livers (with lower L* score) in the current study compared to liver colours in Trampel et al. (2005) and Northcutt et al. (1997) might be due to feed withdrawal prior to slaughtering in our study (12 hours). During fasting, triglyceride synthesis is reduced in chicken livers and rapid loss of livers lipid occurred during fasting period (Hasegawa et al. 1994). This is showing that the differences between colours in our studies and others is more likely associated to the factor of fasting prior to the slaughter rather than the treatment effects.

Table 6.1. ANOVA *P*-values for liver weight in broilers fed canola or juncea meals in corn-based diets

Effect	Weight
Meal	0.023
Level	0.306
Meal × Level	0.102
Enzyme	0.049
Meal × Enzyme	0.472
Level × Enzyme	0.317
Meal × Level × Enzyme	0.675

Table 6.2. Liver weight(g) of broiler chickens fed different levels of canola meal and juncea meal with and without Superzyme – OM™ supplementation in corn-based diets (least square mean ± SE)

Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	42.58±2.58	44.35±2.58	41.21±2.58	45.11±2.58
10%	46.95±2.58	45.98±2.58	45.23±2.58	43.55±2.58
20%	43.37±2.58	43.38±2.58	38.76±2.58	45.79±2.58
30%	43.47±2.58	49.35±2.58	35.30±2.58	40.22±2.58
Meal	44.93±0.91*a*		41.89±0.91*b*	
Enzyme	NO		YES	
	42.11±0.91*b*		44.71±0.91*a*	

[a-b] Means ± SEM with different postscripts for meals and enzyme are significantly different (α=0.05)

Superzyme-OM™ supplementation, and feeding canola meal, compared to juncea meal, increased ($P \leq 0.05$) liver weight (Table 6.2). Increasing liver weight by adding enzyme can be attributed to higher metabolic activities of liver to detoxify the anti-nutritional components (Woyengo et al. 2011), especially glucosinolate in the meal, which becomes more available to birds when enzyme is supplemented. This result is in agreement with the study by Zakaria et al. (2010) who found an increase ($P \leq 0.05$) in liver percent to the whole carcass when broilers were fed commercial multicarbohydrase enzyme at 250 and 500 g. tonne^{-1} of feed, compared to the control diet. Heavier liver weight, due to added enzyme, might be the result of the effect of dietary enzyme on improving the metabolizable energy content of the diet (Meng et al. 2006) which increases the percentage of fat in the liver. Although the relation of BW to liver weight should be considered, the interaction effect of enzyme × level on BW in day 35 (Table 4.11) shows that birds consuming enzyme at all inclusion levels had heavier BW. It is accepted that birds with heavier body weights have heavier liver weight. The effect of Superzyme-OM™ supplementation on liver weight might just be associated with birds BW.

The results of categorical analysis on the corn based trial (Table 6.4) shows that Chi-square is not a good estimate of distribution of colour scores for the effects of the meals, levels and enzyme. This is because the expected count of pale livers in the 2×2 contingency meal table was less than 5 and also, the average of livers which received a score of 2 (pale) for different levels (0,10, 20 and 30%) of the 2×4 contingency table, was less than one. Because, all of the liver samples in the corn based trial had firm texture for all treatments, it is possible to say that there is no effect on texture for enzyme, meals or levels.

No published results were found which compared the liver visual scoring of birds using the Chi-square contingency tests to compare the results and to comprehend the best statistical analysis for visual scoring.

Table 6.3. Liver colour scores from Hunter Lab Miniscan XE™ colorimeter of broiler chickens fed different levels of canola meal and juncea with and without Superzyme- OM ™ supplementation in corn- based diets (least square mean ± SE)

a*Score				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	13.73±0.73	14.61±0.73	12.53±0.73	14.18±0.73
10%	13.25±0.73	13.25±0.73	14.19±0.73	14.66±0.73
20%	12.10±0.73	13.60±0.73	13.58±0.73	14.06±0.73
30%	13.73±0.73	12.64±0.73	13.27±0.73	13.35±0.73
b*Score				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	10.13±0.67	10.48±0.67	8.81±0.67	9.02±0.67
10%	9.30±0.67	8.51±0.67	9.83±0.67	9.51±0.67
20%	8.71±0.67	8.82±0.67	9.99±0.67	9.44±0.67
30%	10.12±0.67	8.95±0.67	9.35±0.67	9.08±0.67
L*Score				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	24.60±1.15	26.16±1.15	26.97±1.15	24.27±1.15
10%	25.66±1.15	26.18±1.15	24.83±1.15	24.54±1.15
20%	25.45±1.15	26.33±1.15	27.32±1.15	25.71±1.15
30%	25.63±1.15	25.08±1.15	26.78±1.15	26.24±1.115

Table 6.4. Chi-Square analysis of liver colour and texture scores of livers from broilers fed corn based diets

	Colour		
	Meal[1]	Level[2]	Enzyme
X^2	4.063	2.243	0.000
DF	1	3	1
P-value	0.044	-	1.000

[1]Two cells within expected counts lees than 5
[2]Four cells with expected counts less than 1- Chi-Square approximation probably invalid

6.6.2 Wheat-based trial

Results of ANOVA analysis for colour scores and liver weights are shown in Table 6.5.

Colour analysis of livers indicated that there was an effect of meal on lightness (L* score), as birds which were fed JM had lighter livers compared to those which were fed CM ($P \leq 0.05$); the lightness can be associated with higher lipid concentrations, as it was shown in a study by Trampel et al. (2005) who found a relationship between liver lightness and hepatic lipid concentration and higher total liver lipid. No significant differences were found ($P \geq 0.05$) in a* and b* colour scores on livers for levels, meals and enzyme inclusion (Table 6.6).

Table 6.5. ANOVA *P*-values for liver a*, b* and L* score and liver weigh in broilers fed canola or juncea meals in wheat-based diets

Effect	a* score	b* score	L* score	Weight
Meal	0.454	0.379	0.048	0.534
Level	0.366	0.864	0.143	0.005
Meal × Level	0.743	0.791	0.505	0.989
Enzyme	0.757	0.135	0.515	0.041
Meal × Enzyme	0.204	0.620	0.110	0.965
Level × Enzyme	0.560	0.706	0.799	0.114
Meal × Level × Enzyme	0.273	0.331	0.084	0.497

Table 6.6. Liver weight (g) of broiler chickens subjected to different levels of canola meal and juncea meal with and without Superzyme-OM ™ supplementation in wheat-based diets (least square mean ± SE)

Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	44.21±2.55	47.88±2.55	45.04±2.55	44.93±2.55	45.51±1.28 *a*
10%	44.70±2.55	49.77±2.55	41.23±2.55	52.32±2.55	47.02±1.28 *a*
20%	43.71±2.55	44.31±2.55	42.01±2.55	43.23±2.55	43.31±1.28 *ab*
30%	39.83±2.55	41.47±2.55	41.19±2.55	39.45±2.55	40.48±1.28 *b*

Enzyme	NO	YES
	42.74±0.90 *b*	45.43±0.90 *a*

[a-b] Means ± SEM with different postscripts for levels and enzyme are significantly different (α=0.05)

There was a reduction (*P*=0.005) in liver weight, when 30% of meal inclusion was provided compared to 0 and 10 % (Table 6.6), which is contrary to the study by Woyengo et al. (2011) where they found increasing the dietary level of expeller-extracted canola meal from 0 to 40% resulted in linear increasing liver weight (*P*<0.001) relative to live body weight. The difference between the results of the previous study and this study can be attributed to different meals, dietary composition or different basal diets, as Woyengo et al. (2011) used corn as a source of energy. Different basal diets can be associated with differences in the composition of gut microbes which hydrolyze glucosinolates and therefore results in differences in the severity of the toxicity of the

glucosinolate degradation products (Woyengo et al. 2011). As it is suggested by Talebali and Farzinpour (2005), higher liver weights for control and low inclusion level of 10% might be due to the lower fat content of the diets compared to other . Dietary fat can reduce liver activity for lipogenesis which results in the liver producing more energy from carbohydrates. Higher liver weight in broilers fed the control diet compared to different inclusion levels, is in agreement with the study by Talebali and Farzinpour (2005) who tested the effect of different dietary levels of full fat canola seed in broilers.

Birds fed dietary enzyme had higher (P=0.041) liver weight than those without enzyme inclusion (Table 6.6). These results are in agreement with both our results in the corn based trial and those of Zakaria et al. (2010). The BW data on day 35 (Table 4.25) indicated the effect of levels and enzyme on BW as the birds fed with enzyme and lower meal inclusion levels had higher BW. The reason for heavier livers by adding enzyme and meal inclusion levels of 0 and 10% might be associated with heavier BW.

In the 2 × 4 contingency table (Table 6.8) of colour by level the average of expected count for pale liver was less than 5 and also in the 2 ×2 contingency table of colour by enzyme one cell with expected counts had less than 5 livers which indicates that in the mentioned cases Chi-square is not a good estimate of distribution. No effects of treatments were found for other treatment effects (P≥0.05). There were no published results found to compare the liver visual scoring of birds using Chi-square contingency tests.

Table 6.7. Liver colour scores from Hunter Lab Miniscan XE™ colorimeter of broiler chickens fed different levels of canola meal and juncea meal with and without Superzyme– OM ™ supplementation in wheat- based diets (least square mean ± SE)

	a*Score			
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	12.81±0.72	13.49±0.83	13.66±0.72	14.11±0.83
10%	12.79±0.83	13.33±0.83	13.77±0.83	13.57±0.84
20%	12.66±1.03	12.61±0.83	12.39±0.72	13.83±0.72
30%	14.40±0.83	11.38±0.83	12.73±1.02	12.64±0.72
	b*Score			
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	8.16±1.01	8.23±1.17	8.98±1.01	9.24±1.17
10%	7.93±1.17	9.17±1.17	10.46±1.17	7.61±1.17
20%	8.17±1.17	8.17±1.17	10.45±1.01	9.07±1.01
30%	10.17±1.17	7.51±1.117	8.71±1.43	8.06±1.01
	L*Score			
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	23.17±1.09	23.56±1.26	25.69±1.09	23.86±1.26
10%	23.51±1.26	26.22±1.26	27.07±1.26	26.22±1.26
20%	25.26±1.70	24.11±1.26	23.87±1.26	24.11±1.26
30%	28.87±1.26	25.42±1.26	26.63±1.26	25.42±1.26
Meal	24.60±0.44*b*		25.86±0.43*a*	

[a-b] Means ± SEM with different postscripts for meals in L score are significantly different (α=0.05)

Table 6.8. Chi-Square analysis of colour and texture scores for livers from birds fed wheat-based diets

	Colour			Texture		
	Meal	Level [1]	Enzyme[2]	Meal	Level	Enzyme
X^2	1.665	1.725	3.294	1261	0016	2693
DF	1	3	1	1	3	1
P-value	0.197	0.631	0.070	0262	0999	0.101

[1]4 cells with expected counts less than 5
[2]1 cell with expected counts less than 5

6.7 Conclusions

Overall, there were no negative effect of different meal inclusion levels with adding Superzyme-OM^{TM} on livers. Further studies should examine to analyze the visual and colour scoring in broilers liver.

CHAPTER 7: THE EFFECT OF DIFFERENT LEVEL OF CANOLA MEAL (BRASSICA NAPUS) AND JUNCEA MEAL (BRASSICA JUNCEA) IN BROILER DIETS SUPPLEMENTED WITHOUT OR WITH A DIETARY ENZYME COCKTAIL: FATTY ACID PROFILE OF BROILER MEAT

7.1. Abstract

The fatty acid content and composition of poultry meat is an important factor in terms of acceptability of broiler carcasses. In this study, the effect of different inclusion levels of CM and JM (0, 10, 20 and 30 %), in corn- and wheat-based diets on fatty acid profiles of broiler tissues, was examined. Additionally the inclusion of Superzyme-OM^{TM} in the diets was investigated. The fatty acid analysis of the diets in wing, breast and thigh samples from two birds from each pen at the end of each trial was measured. For corn-based diets PUFA, MUFA, LA and Omega 6 fatty acids increased ($P \leq 0.05$) in all samples for birds fed CM compared to JM. The Omega 6:Omega 3 ratio was lower in breast and wing meat samples of broilers fed supplemented Superzyme- OM^{TM} compared to those without dietary enzyme. In the wheat-based trial, the amount of SFA decreased in breast samples when enzyme was added and in thigh samples when CM was supplemented. For wheat-based diets, PUFA, MUFA, LA, ALA, EPA, DHA, Omega 3 and Omega 6 fatty acids were lower ($P \leq 0.05$) in breast meat when Superzyme-OM^{TM} was supplemented. The Omega 6:Omega 3 ratio was lower in breast meat when CM was fed to birds compared to JM. Overall, most of the muscle samples in both trials fed CM diets improved fatty acid profiles compared to JM. incorpration of CM and JM both had positive effects on fatty acid profiles of carcass composition.

Key words: Fatty acid, Broilers, Canola, Juncea, Superzyme– OM ^{TM}

7.2. Introduction

Animal or vegetable fats have been commonly used in broiler diets as a source of energy, some PUFA's in vegetable oils are considered essential for broilers and their deficiency causes some disorders such as growth depression (Balnave 1970). The requirement of essential fatty acids for poultry refers to LA this requirement has been estimated as 1.0 percent of the diet (Balnave 1970). ALA dietary need has not been demonstrated for birds (NRC 1994). However, 0.05 to 0.1% of dietary dry matter is suggested (Hovenier et al. 2006). Fat with a higher percentage of unsaturated fatty acids are better absorbed than highly saturated fatty acids (Zollitsch et al. 1997), however, there is a problem of oxidation associated with unsaturated fatty acids. Zollitsch et al. (1997) studied the effects of different dietary fat sources on broilers. These authors analyzed the fatty composition in broiler droppings and identified that the saturated long chain fatty acids could not be metabolized by broilers. Animal performance and carcass quality should both be taken into account when supplementing fat in diets. The composition and quantity of fatty acids in the diet affects fatty acid profiles of the meat. Szymczyk et al (2001) reported that LA in diets reduced the content of MUFA and PUFA in broiler meat. The effects of adding canola oil in diets in Zanini et al. (2008) study showed that canola oil supplementation in broiler diets reduced the lipid content in carcasses, especially the amount of SFA. Adding vegetable sources of Omega 3 such as full fat canola seed and full fat flax seed to broiler chicken diets increased Omega 3 fatty acid content of broiler meat (Rahimi et al. 2011).

7.3. Objectives

The current study investigated the fatty acid composition of wing, breast and thigh samples from broilers fed graded levels of either CM or JM with and without dietary Superzyme- OM^{TM} supplementation in corn and wheat based diets.

7.4. Hypotheses

Lower levels of saturated fat and higher levels of unsaturated fatty acids in different cut of meats of birds fed higher levels of meals with supplementary canola oil is expected. It is also hypothesized that by supplementing enzyme in a diet the levels of total fat in the meat will increase due to the effects of enzyme on availability of energy in the feed.

7.5. Materials and methods

7.5.1. Sample collection and analysis

On day 35 of the corn and wheat-based trials, two birds per pen were randomly selected and commercially slaughtered for fatty acid analysis of wings, breasts and thighs. The samples were stored in sealed plastic bags at -20°C until analysis. The fatty acid content of the skinless meat parts and the dietary diets were analyzed by the Coastal Zones Research Institute Inc. (CZRI), Université de Moncton, Shippagan Campus, New Brunswick, Canada. The samples were subjected to a trans-esterification to produce fatty acid methyl esters (Lepage and Roy 1984). The fatty acid methyl esters were then analysed using gas chromatography, on a CP-3900 GC-FID (Varian) with a ZB-WAX capillary column, 20m × 0.18mm × 0.18µm (Phenomenex). The carrier gas used was hydrogen. The gas chromatography oven to achieve a good separation of the fatty acid methyl esters. The identification of the individual fatty acid methyl esters was done by injection of known mixture of fatty acid methyl esters. The quantification was performed using nonadecanoic acid as an internal standard.

7.5.2. Statistical analysis

Saturated fatty acids (SFA), polyunsaturated fatty acids (PUFA), mono unsaturated fatty acid (MUFA), linoleic acid (LA), alpha linolenic acid (ALA), docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA), Omega 6, Omega 3, Omega 6 :Omega 3 ratio of breast, thigh and wing samples were subjected to analysis of variance using the SAS v.9.3 (SAS Inc., Cary NC) (Littell et al. 1996). The following model was

employed for statistical analysis:

$Y_{ijkl}=\mu+\rho_i+\alpha_j+\beta_k+\gamma_{l+}\alpha\beta_{jk}+\alpha\gamma_{jl}+\beta\gamma_{kl}+\alpha\beta\gamma_{jkl}+\varepsilon_{ijkl}$

Where Y_{ijkl} is the variable of interest; μ is the overall mean; ρ_i is the effect of the i^{th} block (i=1-4); α_j is the effect of j^{th} meal(j=1-2); β_k is the k^{th} level of inclusion (k=1-4); γ_l is the effect of l^{th} enzyme (l=1-2); $\alpha\beta_{jk}$ is the effect of the interaction between meal and the level of meal inclusion, $\alpha\gamma_{jl}$ is the effect of the interaction between meal and enzyme, $\beta\gamma_{kl}$ is the effect of interaction between meal level of inclusion and enzyme; $\alpha\beta\gamma_{jkl}$ is the effect of interaction between meal, level and enzyme and ε_{ijkl} is the random effect of error. If treatment effect was significant, the Tukey-Kramer option was used to compare differences among the least square means. The α-level of significance was 0.05.

7.6. Results and discussions

7.6.1. Corn-based trial

Fatty acid compositions of corn-based diets changed from the starter to the finisher diets (Table 7.1). ALA and Omega 3 content of the diets increased in all periods as the level of meal inclusion increased. Omega 6:Omega 3 ratio was decreased by increasing the levels of either CM or JM.

Fatty acid composition of corn-based diets as a percentage of total fat content of diet can be seen in Appendix G.1.

Table 7.1. Fatty acid content of sample (mg·g^{-1}) of corn-based diets with increasing levels of canola or juncea meals (AS FED BASIS)

Starter								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg·g^{-1})								
LA[1]	20.79	23.62	25.13	26.14	20.79	20.50	20.96	20.27
ALA[2]	3.74	4.93	6.03	6.37	3.74	3.75	3.88	4.01
DHA[3]	0.00	0.00	0.02	0.03	0.00	0.02	0.01	0.02
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Omega 6[5]	20.82	23.66	25.18	26.19	20.82	20.54	21.00	20.30
Omega 3	3.74	4.93	6.06	6.43	3.74	3.78	3.90	3.91
Omega 6:Omega 3	5.57	4.80	4.16	4.08	5.57	5.43	5.39	5.20
SFA[5]	7.18	7.83	8.44	8.77	7.18	6.89	7.04	7.00
PUFA[6]	24.74	28.76	31.47	32.85	24.74	24.52	25.09	24.42
MUFA[7]	27.52	37.78	47.33	51.64	27.52	27.66	29.86	31.34
Total fatty acids	59.44	74.37	87.24	93.26	59.44	59.07	61.94	62.76
Grower								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg·g^{-1})								
LA[1]	22.60	26.16	25.45	27.74	22.60	22.41	22.21	22.15
ALA[2]	4.01	4.94	5.41	6.71	4.01	4.03	4.10	4.28
DHA[3]	0.00	0.00	0.01	0.00	0.00	0.00	0.01	0.02
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Omega 6[5]	22.63	26.20	25.50	27.79	22.63	22.45	22.26	22.19
Omega 3	4.01	4.94	5.43	6.73	4.01	4.03	4.12	4.30
Omega 6:Omega 3	5.64	5.31	4.69	4.13	5.64	5.57	5.41	5.16
SFA[5]	7.59	8.54	8.47	9.28	7.59	7.53	7.53	7.66
PUFA[6]	26.80	31.35	31.12	34.75	26.80	26.65	26.54	26.68
MUFA[7]	30.00	39.65	44.41	55.89	30.00	31.72	33.51	36.15
Total fatty acids[8]	64.39	79.54	84.35	99.89	64.39	65.9	67.58	70.49
Finisher								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg·g^{-1})								
LA[1]	23.76	25.78	29.03	27.16	23.76	23.39	21.63	22.51
ALA[2]	3.74	4.60	6.04	6.09	3.74	3.90	3.62	3.76
DHA[3]	0.00	0.00	0.00	0.04	0.00	0.00	0.00	0.00
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Omega 6	23.79	25.82	29.08	27.21	23.79	23.42	21.67	22.54
Omega 3	3.74	4.60	6.04	6.17	3.74	3.90	3.62	3.76
Omega 6:Omega 3	6.36	5.61	4.81	4.41	6.36	6.01	5.99	6.00
SFA[5]	7.56	8.13	9.16	8.82	7.56	7.42	7.00	7.23
PUFA[6]	27.65	30.58	35.33	33.51	27.65	27.47	25.44	26.46
MUFA[7]	30.00	37.92	50.80	52.22	30.00	32.27	30.61	33.26
Total fatty acids[8]	65.21	76.63	95.29	94.55	65.21	67.16	63.05	66.95

[1]LA, Linoleic acid,
[2]ALA, Alpha -Linolenic acid,
[3] DHA, docosahexaenoic acid,
[4] EPA, eicosapentaenoic acid,
[5] SFA, Saturated fatty acids,
[6] PUFA, Poly unsaturated fatty acids,
[7] MUFA, Mono unsaturated fatty acids
[8]Total fatty acids was calculated as SFA+PUFA+MUFA

ANOVA for SFA (Table 7.2) indicated the significant interaction effects of type of meal × meal inclusion level on SFA content of wing samples. The main effect of enzyme on SFA content of breast samples was significantly different.

Table 7.2. ANOVA *P*-values for saturated fatty acids for carcass parts of broilers fed corn-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.379	0.642	0.315
Level	0.380	0.005	0.084
Meal × Level	0.451	0.030	0.299
Enzyme	0.005	0.428	0.069
Meal × Enzyme	0.158	0.389	0.255
Level × Enzyme	0.574	0.733	0.244
Meal × Level × Enzyme	0.376	0.455	0.898

Supplementing Superzyme-OM^{TM} in diets decreased the SFA content of breast samples (Table 7.3). This is a desirable effect of enzyme because of the relationships between SFA and cardiovascular diseases in human, lower SFA in the meat is more desirable for consumers (Simopoulos 1997). This result might be associated with the effect of enzyme on nutrient digestibility, which can increase the availability of nutrients in the meals including the canola and juncea with a good fatty acid profile composition.

SFA content of breast, thigh and wing samples as a percentage of total fat in a diet is reported in Appendix G.2

The interaction effect of meal by meal inclusion level indicated (Table 7.4) that the ranking of level is different depending on whether CM or JM was inclided, SFA content of wing samples at the highest inclusion level of canola meal (30%) decreased compared to the control diet.

Table 7.3. Saturated fatty acids (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	30.58±2.74	22.18±2.38	24.36±2.38	20.99±2.38
10%	26.68±2.13	20.74±2.38	21.69±2.38	19.93±2.38
20%	18.07±2.38	18.84±2.38	21.30±2.38	20.82±2.38
30%	22.85±2.38	20.99±2.38	19.93±2.38	21.36±2.38
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	7.08±1.49	3.70±1.67	6.23±1.67	3.38±1.67
10%	5.98±1.67	4.22±1.93	9.73±1.67	4.02±1.67
20%	3.55±1.67	4.23±1.67	9.21±1.67	3.40±1.67
30%	4.53±1.67	3.83±1.67	3.76±1.67	3.36±1.67
Enzyme	NO		YES	
	6.26±0.58*a*		3.77±0.60*b*	

[a-b] Means ± SEM with different postscripts for enzyme within cut of meat are significantly different (α=0.05)

Table 7.4. Saturated fatty acids (mg·g^{-1}) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM}(least square mean ±SE)

	Wing					
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	29.15±2.40	22.18±2.40	24.36±2.40	20.82±2.40	29.27±1.28*a*	25.12±1.28*ab*
10%	27.13±2.40	20.74±2.40	24.36±2.40	19.93±2.40	25.61±1.28*ab*	24.24±1.28*ab*
20%	18.07±2.40	18.84±2.40	20.82±2.40	21.30±2.40	21.34±1.28*b*	25.05±1.28*ab*
30%	22.85±2.40	20.99±2.40	19.93±2.40	21.36±2.40	22.69±1.28*b*	22.81±1.28*b*

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

ANOVA for PUFA (Table 7.5) indicated the significant effects of meal on the content of PUFA in breast, wing and thigh samples. All the other parameters were not significant.

Table 7.5. ANOVA *P*-values for poly unsaturated fatty acids for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.013	0.003	0.010
Level	0.488	0.471	0.269
Meal × Level	0.927	0.483	0.517
Enzyme	0.934	0.294	0.140
Meal × Enzyme	0.092	0.949	0.427
Level × Enzyme	0.847	0.546	0.371
Meal × Level × Enzyme	0.697	0.739	0.647

The PUFA content of thigh, breast and wing samples increased by supplementing CM compared to JM in diets. (Table 7.6). These results can be explained by higher PUFA content of CM in the diets compared to JM (Table 7.1), because higher percentage of canola oil added to the diets while CM was supplemented.

Table 7.6. Polyunsaturated fatty acids (mg·g^{-1}) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM}(least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	24.61±2.34	20.77±2.34	23.07±2.34	20.30±2.34
10%	28.19±2.34	21.65±2.34	22.26±2.34	20.11±2.34
20%	22.48±2.34	24.25±2.34	21.71±2.34	20.76±2.34
30%	29.09±2.34	26.90±2.34	20.96±2.34	23.56±2.34
Meal	24.74±0.83*a*		21.59±0.83*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	8.60±1.28	6.37±1.28	4.88±1.28	6.77±1.28
10%	8.92±1.28	7.95±1.28	5.75±1.28	7.10±1.28
20%	6.80±1.28	6.20±1.28	5.95±1.28	5.16±1.28
30%	8.12±1.28	7.73±1.28	4.82±1.28	6.10±1.28
Meal	7.58±0.45*a*		5.93±0.45*b*	
Wings				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	26.47±1.97	29.08±1.97	24.31±1.97	26.94±1.97
10%	27.71±1.97	30.41±1.97	26.01±1.97	27.87±1.97
20%	30.75±1.97	28.32±1.97	26.90±1.97	27.70±1.97
30%	30.89±1.97	32.46±1.97	26.59±1.97	25.24±1.97
Meal	29.51±0.70*a*		26.44±0.69*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

PUFA content of breast, thigh and wing samples as a percentage of total fat in a diet can be seen in Appendix G.3.

ANOVA for MUFA (Table 7.7) indicated the significant main effect of meal on MUFA content of breast, wing and thigh samples. All the other parameters were not significantly different.

The MUFA content of thigh, breast and wing samples were higher in the birds fed CM compared to the ones fed JM (Table 7.8). The MUFA content of diets supplemented

with CM were higher than those with JM (Table 7.1), because higher percentage of canola oil added to the diets while CM was supplemented.

Table 7.7. ANOVA *P*-values for monounsaturated fatty acids for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.012	0.016	0.015
Level	0.395	0.607	0.275
Meal ×Level	0.792	0.250	0.464
Enzyme	0.846	0.276	0.168
Meal × Enzyme	0.064	0.796	0.362
Level × Enzyme	0.786	0.717	0.269
Meal × Level × Enzyme	0.670	0.440	0.846

Table 7.8. Monounsaturated fatty acid (mg·g^{-1}) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	60.42±6.22	47.60±6.22	54.09±6.22	44.83±6.22
10%	67.17±6.22	51.33±6.22	52.61±6.22	48.27±6.22
20%	50.29±6.22	54.38±6.22	49.15±6.21	50.80±6.22
30%	68.88±6.22	64.59±6.22	48.24±6.22	54.21±6.22
Meal	58.08±2.20*a*		50.27±2.20*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	19.70±3.10	12.60±3.10	9.08±3.10	13.14±3.10
10%	18.61±3.10	17.06±3.10	11.26±3.10	15.16±3.10
20%	12.90±3.10	11.85±3.10	11.44±3.10	10.66±3.10
30%	16.77±3.10	15.93±3.10	8.30±3.10	14.07±3.10
Meal	15.68±1.10*a*		11.64±1.10*b*	
Wings				
Meal	Canola meal		Juncea meal	
Enzyme Levels	NO	YES	NO	YES
0%	66.27±4.94	68.07±4.94	58.72±4.94	61.07±4.94
10%	64.30±4.94	73.30±4.94	62.17±4.94	68.06±4.94
20%	69.01±4.94	63.37±4.94	61.65±4.94	70.70±4.94
30%	73.42±4.94	76.58±4.94	63.11±4.94	59.28±4.94
Meal	69.29±1.75*a*		63.09±1.75*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

MUFA content of breast, thigh and wing samples as a percentage of total fat in a diet can be seen in Appendix G.4.The effects of meal ×level was significantly different in brest, thigh and wing samples of broilers meat.

ANOVA for LA (Table 7.9) indicated the significant main effect of type of meal on LA content of breast, wing and thigh samples. All the other parameters were not significantly different (P >0.05).

Table 7.9. ANOVA *P*-values for Linoleic acid for carcass parts of broilers fed corn-based diet

Effect	Tissues		
	Breast	Wing	Thigh
Meal	0.017	0.007	0.012
Level	0.463	0.788	0.347
Meal × Level	0.907	0.550	0.565
Enzyme	0.928	0.291	0.141
Meal × Enzyme	0.079	0.919	0.404
Level × Enzyme	0.830	0.504	0.363
Meal × Level × Enzyme	0.673	0.748	0.665

The LA content of thigh, breast and wing samples were higher in the birds fed CM compared to those fed JM (Table 7.10). The LA content of diets supplemented with CM was higher than the ones with JM (Table 7.1). These results agreed with the study by Zanini et al (2008) in corn-based diets, that addition of LA in the diet resulted in LA deposition in tissue samples of broilers.

Table 7.10. Linoleic acid ($mg \cdot g^{-1}$) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	19.37±1.92	16.25±1.92	17.92±1.92	15.92±1.92
10%	22.44±1.92	16.76±1.92	17.39±1.92	15.67±1.92
20%	17.31±1.92	18.62±1.92	16.77±1.92	15.94±1.92
30%	22.41±1.92	20.91±1.92	16.17±1.92	18.20±1.92
Meal	19.26±0.68*a*		16.5±0.68*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	6.31±1.01	4.50±1.01	3.24±1.01	4.75±1.01
10%	6.45±1.01	5.70±1.01	3.98±1.01	5.10±1.01
20%	4.70±1.01	4.21±1.01	4.17±1.01	3.52±1.01
30%	5.72±1.01	5.31±1.01	3.11±1.01	4.97±1.01
Meal	5.36±0.36*a*		4.11±0.36*b*	
Wings				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	20.59±1.54	22.48±1.54	18.73±1.54	20.95±1.54
10%	21.07±1.54	23.22±1.54	19.87±1.54	21.50±1.54
20%	23.29±1.54	21.20±1.54	20.56±1.54	21.22±1.54
30%	23.24±1.54	24.26±1.54	20.13±1.54	19.22±1.54
Meal	22.42±0.54*a*		20.27±0.54*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

ANOVA for ALA (Table 7.11) indicated the significant interaction effects of meal × level on the ALA content of wing samples. Enzyme had a significant effect on ALA content of wing samples. Meal used significantly affected ALA content of breast and thigh samples.

Table 7.11. ANOVA *P*-values for Alpha linolenic acid for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.002	<.0001	0.007
Level	0.382	0.000	0.034
Meal ×Level	0.684	0.012	0.516
Enzyme	0.633	0.048	0.402
Meal × Enzyme	0.175	0.712	0.727
Level × Enzyme	0.810	0.594	0.625
Meal × Level × Enzyme	0.769	0.654	0.527

ALA content of thigh samples of the birds fed 30% meal inclusion were higher than the control group (Table 7.12). Both breast and thigh samples in birds fed CM hadhigher in ALA compared to those fed JM (Table 7.12).

Table 7. 12. Alpha linolenic acid (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh					
Meal	Canola meal		Juncea meal		
Enzyme	NO	YES	NO	YES	Levels
Levels					
0%	2.81±0.38	2.57±0.38	2.71±0.38	2.31±0.38	2.60±0.19*b*
10%	3.23±0.38	2.77±0.38	2.77±0.38	2.29±0.38	2.76±0.19*ab*
20%	3.02±0.38	3.39±0.38	2.70±0.38	2.69±0.38	2.95±0.19*ab*
30%	4.15±0.38	3.58±0.38	2.63±0.38	3.14±0.38	3.37±0.19*a*
Meal	3.19±0.13*a*		2.65±0.13*b*		
Breast					
Meal	Canola meal		Juncea meal		
Enzyme	NO	YES	NO	YES	
Levels					
0%	0.88±0.20	0.68±0.20	0.45±0.20	0.73±0.20	
10%	1.13±0.20	0.80±0.20	0.60±0.20	0.80±0.20	
20%	0.83±0.20	0.80±0.20	0.63±0.20	0.53±0.20	
30%	1.08±0.20	1.13±0.20	0.43±0.20	0.78±0.20	
Meal	0.93±0.07*a*		0.62±0.07*b*		

[a-b] Means ± SEM with different postscripts for meals in thigh and breast and for levels in thigh are significantly different (α=0.05)

The interaction effect of meal × level (Table 7.13) indicated a difference between ALA of wing samples between meals when birds were supplemented with 30% canola or juncea meals. Levels of inclusion was different depending on type of meals. Higher ALA content was associated with the birds fed CM compared to JM.

Table 7.13. Alpha linolenic acid (mg·g^{-1}) in wing meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM^{TM} (least square mean ±SE)

Wing						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	3.23±0.31	3.85±0.31	3.00±0.31	3.58±0.31	3.54±0.22*c*	3.29±0.22*c*
10%	3.94±0.31	4.38±0.31	3.46±0.31	3.74±0.31	4.16±0.22*bc*	3.60±0.22*c*
20%	4.76±0.31	4.54±0.31	3.66±0.31	3.87±0.31	4.65±0.22*ab*	3.76±0.22*bc*
30%	4.89±0.31	5.54±0.31	3.53±0.31	3.49±0.31	5.22±0.22*a*	3.51±0.22*c*

[a-c] Means ± SEM with different postscripts for interaction means for meals within cut of meat are significantly different (α=0.05)

ALA content of breast, thigh and wing samples as a percentage of total fat in a diet can be seen in Appendix G.5.The main effect of meal in thigh samples and the interaction effect of meal ×level in breast and wing samples were significantly different.

ANOVA for DHA (Table 7.14) indicated a significant effect of meal × level on the DHA content of breast and thigh samples. Enzyme had a significant effect on DHA content of thigh samples. DHA in wing samples were not affected by dietary treatments ($P > 0.05$).

Table 7.14.ANOVA *P*-values for docosahexaenoic acid for carcass parts of broilers fed corn-based diet			
Effect	Tissues		
	Breast	Wing	Thigh
Meal	0.401	0.303	0.248
Level	0.080	0.705	0.031
Meal ×Level	0.016	0.618	0.033
Enzyme	0.361	0.130	0.003
Meal × Enzyme	0.555	0.117	0.978
Level × Enzyme	0.975	0.835	0.875
Meal × Level × Enzyme	0.670	0.427	0.805

Breast samples of birds that consumed 30% CM diets had more ($P \leq 0.05$) DHA compared to 20% JM. In thigh samples, birds with 30% CM inclusion had DHA content that was higher compared to the control group. The effect of enzyme in thigh samples indicated that supplementing with enzyme decreased the DHA content of the meat (Table 7.15), the reason for this result is not clear.

Table 7.15. Docosahexaenoic acid (mg·g^{-1}) in breast and thigh meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM}(least square mean ±SE)

Breast						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	0.11±0.00	0.10±0.00	0.13±0.00	0.12±0.00	0.10±0.01*ab*	0.13±0.01*ab*
10%	0.11±0.00	0.11±0.00	0.11±0.00	0.10±0.00	0.11±0.01*ab*	0.11±0.01*ab*
20%	0.13±0.00	0.12±0.00	0.10±0.00	0.01±0.00	0.12±0.01*ab*	0.10±0.01*b*
30%	0.13±0.01	0.14±0.00	0.13±0.00	0.11±0.00	0.13±0.01*a*	0.12±0.01*ab*
Thigh						
					Meal × Level	
Meal	Canola meal		Juncea meal			
Enzyme	NO	YES	NO	YES	Canola meal	Juncea meal
Levels						
0%	0.14±0.01	0.13±0.01	0.17±0.01	0.14±0.01	0.13±0.01*b*	0.16±0.01*ab*
10%	0.16±0.01	0.13±0.01	0.14±0.01	0.13±0.01	0.15±0.01*ab*	0.14±0.01*ab*
20%	0.17±0.01	0.15±0.01	0.15±0.01	0.13±0.01	0.16±0.01*ab*	0.14±0.01*ab*
30%	0.18±0.01	0.17±0.01	0.16±0.01	0.15±0.01	0.17±0.01*a*	0.16±0.01*ab*
Enzyme	NO		YES			
	0.16±0.00*a*		0.14±0.00*b*			

[a-b] Means ± SEM with different letters for interaction means in breast and thigh and for enzyme in thigh are significantly different (α=0.05)

The DHA content of wing samples (Table 7.16) was not affected by dietary treatments.

Table 7.16. Docosahexaenoic acid (mg·g^{-1}) wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

	Wing			
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.16±0.02	0.16±0.02	0.18±0.02	0.16±0.02
10%	0.17±0.02	0.17±0.02	0.16±0.02	0.15±0.02
20%	0.19±0.02	0.17±0.02	0.18±0.02	0.16±0.02
30%	0.17±0.02	0.19±0.02	0.19±0.02	0.14±0.02

DHA content of breast and wing samples as a percentage of total fat in a diet can be seen in Appendix G.6. The main effect of enzyme in breat meat samples and the interaction effect of level ×enzyme was significantly different.

ANOVA for EPA (Table 7.17) indicated a significant effect of meal on EPA content of breast, wing and thigh samples and a significant effect of level on breast samples. All the other parameters were not significantly different (P >0.05).

Table 7.17. ANOVA *P*-values for eicosapentaenoic acid for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.001	0.003	0.006
Level	0.003	0.471	0.786
Meal ×Level	0.390	0.483	0.568
Enzyme	0.498	0.294	0.272
Meal × Enzyme	0.881	0.949	0.439
Level × Enzyme	0.769	0.546	0.354
Meal × Level × Enzyme	0.352	0.739	0.899

CM increased the content of EPA in thigh, wing and breast samples compared to JM (Table 7.18 and Table 7.19). This may be a results of higher ALA levels in CM diets (Table 7.1) compared to JM diet. ALA is a precursor of EPA.

Table 7.18. Eicosapentaenoic acid (mg·g^{-1}) in thigh and wing meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.09±0.01	0.08±0.01	0.08±0.01	0.07±0.01
10%	0.09±0.01	0.08±0.01	0.09±0.01	0.07±0.01
20%	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
30%	0.09±0.01	0.08±0.01	0.07±0.01	0.08±0.01
Meal	0.09±0.00*a*		0.07±0.00*b*	
Wing				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.11±0.01	0.12±0.01	0.10±0.01	0.10±0.01
10%	0.11±0.01	0.12±0.01	0.10±0.01	0.10±0.01
20%	0.12±0.01	0.12±0.01	0.11±0.01	0.11±0.01
30%	1.13±0.01	0.13±0.01	0.12±0.01	0.09±0.01
Meal	0.12±0.01*a*		0.10±0.01*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

Birds fed control diets showed higher EPA in their breast meat compared to birds fed 20 and 30% meal inclusion (Table 7.19). The birds fed CM had higher amount of EPA in their breast samples compared to JM (Table 7.19). The reason for this result is unclear.

Table 7.19. Eicosapentaenoic acid (mg·g^{-1}) in breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

	Breast				
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	0.06±0.00	0.06±0.00	0.06±0.00	0.06±0.00	0.06±0.00*a*
10%	0.06±0.00	0.06±0.00	0.05±0.00	0.05±0.00	0.05±0.00*ab*
20%	0.06±0.00	0.05±0.00	0.05±0.00	0.05±0.00	0.05±0.00*b*
30%	0.05±0.00	0.06±0.00	0.05±0.00	0.04±0.00	0.05±0.00*b*
Meal	0.06±0.00*a*		0.05±0.00*b*		

[a-b] Means ± SEM with different postscripts for meals and levels within cut of meat are significantly different (α=0.05)

EPA content of breast samples as a percentage of total fat in a diet can be seen in Appendix G.7.The interaction effect of meal ×enzyme was significant in breast samples.

ANOVA for Omega 6 fatty acids (Table 7.20) indicated type of meal influenced Omega 6 content of breast, wing and thigh samples. All the other parameters were not significantly different (P >0.05).

Table 7.20. ANOVA *P*-values for Omega 6 for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.019	0.011	0.014
Level	0.481	0.799	0.363
Meal ×Level	0.918	0.636	0.539
Enzyme	0.991	0.359	0.129
Meal × Enzyme	0.080	0.994	0.397
Level × Enzyme	0.842	0.512	0.357
Meal × Level × Enzyme	0.655	0.766	0.651

The effect of meal on Omega 6 content of breast, thigh and wing samples shows the birds fed CM had higher levels of Omega 6 compared to birds fed JM (Table 7.21).

The same pattern of higher Omega 6 content of CM compared to JM can be observed in the diets (Table 7.1).

Table 7.21. Omega 6 (mg·g^{-1}) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	21.02±1.99	17.59±1.99	19.63±1.99	17.36±1.99
10%	24.17±1.99	18.20±1.99	18.79±1.99	17.18±1.99
20%	18.74±1.99	20.07±1.99	18.31±1.99	17.40±1.99
30%	24.01±1.99	22.51±1.99	17.64±1.99	19.67±1.99
Meal	20.79±0.70*a*		18.25±0.70*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.26±0.22	1.03±0.22	0.79±0.22	1.11±0.22
10%	1.52±0.22	1.51±0.22	0.93±0.22	1.14±0.22
20%	1.23±0.22	1.18±0.22	0.94±0.22	0.86±0.22
30%	1.52±0.22	1.55±0.22	0.79±0.22	1.15±0.22
Meal	1.33±0.08*a*		0.96±0.08*b*	
Wings				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	22.32±1.63	24.34±1.63	20.40±1.63	22.47±1.63
10%	22.81±1.63	25.05±1.63	21.68±1.63	23.27±1.63
20%	24.98±1.63	22.82±1.63	22.31±1.63	22.89±1.63
30%	25.00±1.63	25.86±1.63	22.07±1.63	20.88±1.63
Meal	24.15±0.58*a*		21.10±0.58*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

ANOVA for Omega 3 (Table 7.22) indicated a significant effect of meal × level on Omega 3 content of wing samples. The main effect of meal type was significantly different for Omega 3 content of breast and thigh samplesThe main effect of level was significantly different for Omega 3 content of thigh samples.

Table 7.22. ANOVA *P*-values for Omega 3 for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.002	<.0001	0.004
Level	0.383	0.001	0.021
Meal × Level	0.680	0.024	0.416
Enzyme	0.648	0.100	0.308
Meal × Enzyme	0.176	0.634	0.675
Level × Enzyme	0.874	0.655	0.571
Meal × Level × Enzyme	0.833	0.656	0.483

The Omega 3 content of wing samples was higher for the birds that consumed 30% CM compared to the ones consuming 30% JM (Table 7.23). CM resulted in higher Omega 3 content in thigh and breast samples compared to JM (Table 7.24).

Table 7.23. Omega 3 ($mg \cdot g^{-1}$) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

	Wing					
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	3.93±0.35	4.58±0.35	3.71±0.35	4.25±0.35	4.25±0.25*c*	3.98±0.25*c*
10%	4.69±0.35	5.16±0.35	4.15±0.35	4.42±0.35	4.92±0.25*bc*	4.29±0.25*c*
20%	5.56±0.35	5.30±0.35	4.40±0.35	4.59±0.35	5.43±0.25*ab*	4.49±0.25*bc*
30%	5.72±0.35	6.38±0.35	4.32±0.35	4.15±0.35	6.05±0.25*a*	4.24±0.25*c*

[a-c] Means ± SEM with different postscripts for interaction means within cut of meat are significantly different (α=0.05)

Table 7.24. Omega 3 ($mg \cdot g^{-1}$) in thigh and breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	3.44±0.40	3.06±0.40	3.31±0.40	2.84±0.40
10%	3.87±0.40	3.34±0.40	3.34±0.40	2.81±0.40
20%	3.64±0.40	4.06±0.40	3.28±0.40	3.24±0.40
30%	4.94±0.40	4.26±0.40	3.20±0.40	3.75±0.40
Meal	3.83±0.14*a*		3.22±0.14*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.26±0.22	1.10±0.22	0.79±0.22	1.03±0.22
10%	1.51±0.22	1.35±0.22	0.93±0.22	1.14±0.22
20%	1.23±0.22	1.17±0.22	0.93±0.22	0.86±0.22
30%	1.52±0.22	1.55±0.22	0.80±0.22	0.79±0.22
Meal	1.33±0.08*a*		0.96±0.08*b*	

[a-b] Means ± SEM with different postscripts for meals within cut of meat are significantly different (α=0.05)

Omega 3 content of breast and thigh samples as a percentage of total fat in a diet can be seen in Appendix G.8.the interaction effects of meal × level and meal × enzyme was significantly different in breast and thigh samples.

Results of ANOVA analysis for the ratio of Omega 6:Omega 3 (Table 7.25) indicated a significant effect of meal × level on breast and wing samples. The main effect of level was significantly different for thigh samples. Enzyme supplementation had an effect on the ratio of Omega 6: Omega 3 of breast and wing samples. All other parameters were not significantly different (P>0.05).

Table 7.25. ANOVA *P*-values for Omega 6:Omega 3 for carcass parts of broilers fed corn-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	<.0001	<.0001	0.409
Level	<.0001	<.0001	0.008
Meal × Level	<.0001	<.0001	0.754
Enzyme	0.008	0.016	0.525
Meal × Enzyme	0.088	0.298	0.472
Level × Enzyme	0.764	0.428	0.981
Meal × Level × Enzyme	0.599	0.988	0.305

In thigh samples Omega 6:Omega 3 ratio was lower for 20 and 30 % meal diets compared to the control group (Table 7.26). The reason for these result is not clear since the level of Omega 6:Omega 3 decreased in the diets. We expected to observe the same trend in the ratio of Omega 6:Omega 3 in meat samples.

Table 7.26. Omega 6 :Omega 3 in thigh meat of broilers fed different levels of juncea meal or canola meal with and without Superzyme-OM™ (least square mean ±SE)

	Thigh				
Meal	Canola meal		Juncea meal		Level
Enzyme	NO	YES	NO	YES	
Levels					
0%	6.13±0.36	5.78±0.36	5.97±0.36	6.13±0.36	6.00±0.18a
10%	6.37±0.36	5.56±0.36	5.61±0.36	6.05±0.36	5.90±0.18*ab*
20%	5.17±0.36	5.06±0.36	5.63±0.36	5.39±0.36	5.31±0.18*b*
30%	5.03±0.36	5.33±0.36	5.56±0.36	5.27±0.36	5.29±0.18*b*

[a-b] Means ± SEM with different postscripts for levels within cut of meat are significantly different (α=0.05)

The ratio of Omega 6:Omega 3 was lowest in breast and wing samples of the birds fed 20 and 30 % of CM. (Table 7.27) Enzyme supplementation in both breast and wing samples decreased this ratio (Table 7.27).

Table 7.27. Omega 6 :Omega 3 in breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	5.74±0.16	5.18±0.16	5.16±0.16	5.10±0.16	5.46±0.11*a*	5.13±0.11*ab*
10%	5.03±0.16	4.84±0.16	5.23±0.16	5.20±0.16	4.93±0.11*b*	5.21±0.11*ab*
20%	4.53±0.16	4.25±0.16	5.30±0.16	4.10±0.16	4.39±0.11*c*	5.15±0.11*ab*
30%	4.37±0.16	3.96±0.16	4.98±0.16	5.03±0.16	4.16±0.11*c*	5.00±0.11*ab*
Enzyme	NO		YES			
	5.04±0.06*a*		4.82±0.11*b*			
Wing						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	5.69±0.12	5.33±0.12	5.52±0.12	5.30±0.12	5.51±0.08*a*	5.41±0.08*ab*
10%	4.89±0.12	4.85±0.12	5.25±0.12	5.27±0.12	4.87±0.08*c*	5.26±0.08*ab*
20%	4.50±0.12	4.32±0.12	5.09±0.12	5.02±0.12	4.41±0.08*d*	5.05±0.08*bc*
30%	4.33±0.12	4.08±0.12	5.12±0.12	5.04±0.12	4.20±0.08*d*	5.08±0.08*bc*
Enzyme	NO		YES			
	5.05±0.04*a*		4.90±0.04*b*			

[a-b] Means ± SEM with different postscripts for interaction means and enzyme within cut of meat are significantly different (α=0.05)

The results of a study by Ajuyah et al (1991) on the fatty acid profile of meat from broiler chickens fed canola oil in corn-based diets agree with the current study. Increasing the LA content of feed increased the LA content in broiler meat. Birds fed full fat canola seed in their diets showed an increase in EPA compared to the control groups. Similarly Omega 3 fatty acid levels increased in tissues parallel to the increas EPA in the diets. Tissue accumulation of ALA, EPA, DHA were in relation to the fat sources and the level of fats in the diets. Marion and Woodroof (1965) showed that highly unsaturated fats in diets results in more unsaturated fat in carcasses. The same results were reported by Phetteplace and Watkins (1989).

Higher canola oil was included in the starter (Table 4.1), grower (Table 4.3) and finisher (Table 4.5) corn-based diets supplemented with graded levels of CM compared to JM due to the AME_n estimate used to formulate the diet. This is the reason for a higher content of PUFA, MUFA, LA and Omega 6 ($P \leq 0.05$) in all cuts of meat in broilers fed CM compared to JM in this study.

The effects of supplementing Superzyme-OM^{TM} in fatty acid profiles of different cuts of meat might be associated with the effect of enzyme on nutrient digestibility. Enzyme, can increase the availability of nutrients in the diets, including canola and juncea meals, with their good fatty acid profiles. There was a lower Omega 6:Omega 3 ratio in breast (P=0.01) and wing (P=0.02) samples of broilers fed supplemented Superzyme- OM^{TM} compared to birds without dietary enzyme. This is agreed with Slominski et al (2006) where the addition of multicarbohydrase enzyme in corn-based diets of broilers containing oilseeds improved oil utilization. Enzyme increased NSP digestibility in canola. In a study by Jia et al (2008) on layers fed flax seed, enzyme addition increased the n-3 fatty acid deposition in eggs. Improving the fatty acid profile of birds fed Superzyme-OM^{TM} might be due to the effects of enzyme on cell wall digestion. Resulting in the release of oil making it more available for digestive enzymes.

In general, the EPA and DHA in broiler meat shows the ability of birds to make this fatty acids from canola oil as occured in our experimental diets. The dietary levels of EPA and DHA were 0.00 mg g^{-1} this is in agreement with Ajuyah et al. (1991).

7.6.2. Wheat-based trial

Fatty acid compositions of wheat-based diets (Table 7.28) show ALA and Omega 6, and Omega 3 of the diets which included either CM or JM are higher than the control diets for starter, grower and finisher periods.

Fatty acid composition of wheat-based diets as a percentage of total fat content of diet reported in Appendix H.1

Table 7. 28. Fatty acid content (mg·g^{-1}) of wheat-based diets supplemented with canola or juncea meals

Starter								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	18.63	20.15	23.26	23.30	18.63	19.92	18.56	19.10
ALA[2]	3.89	4.80	6.23	6.55	3.89	4.86	4.23	4.42
DHA[3]	0.00	0.00	0.00	0.02	0.00	0.00	0.03	0.05
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.01
Omega 6[5]	18.67	20.19	23.32	23.09	18.67	19.97	18.61	19.14
Omega 3	3.95	4.84	6.29	6.60	3.95	4.88	4.31	4.53
Omega 6:Omega 3	2.73	2.85	3.26	3.10	2.73	2.66	2.62	2.77
SFA[5]	7.16	7.55	8.80	8.67	7.16	7.38	9.64	7.26
PUFA[6]	22.81	22.26	29.88	29.95	22.81	25.05	23.11	23.89
MUFA[7]	27.82	35.55	49.25	50.08	27.82	32.56	30.75	34.43
Total fatty acids	57.79	65.36	87.93	88.70	57.79	64.99	63.50	65.58
Grower								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	20.68	21.85	22.86	24.94	20.68	21.05	21.61	19.26
ALA[2]	4.51	5.25	5.99	7.04	4.51	4.89	5.19	4.38
DHA[3]	0.00	0.00	0.00	0.00	0.00	0.02	0.01	0.01
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.03	0.00	0.00
Omega 6[5]	20.73	21.91	22.93	25.01	20.73	21.10	21.67	19.32
Omega 3	4.53	5.29	6.02	7.06	4.53	4.98	5.25	4.44
Omega 6:Omega 3	2.98	3.06	3.20	3.47	2.98	2.96	3.10	2.83
SFA[5]	7.47	7.75	8.20	8.94	7.47	7.55	7.81	7.03
PUFA[6]	25.48	27.44	29.24	32.42	25.48	26.28	27.10	23.96
MUFA[7]	32.25	38.82	46.65	56.69	32.25	36.12	40.06	34.11
Total fatty acids	65.20	74.01	84.09	98.05	65.20	69.95	74.97	65.10
Finisher								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	21.32	22.49	24.79	25.36	21.32	20.85	20.76	20.49
ALA[2]	4.22	4.98	5.99	6.46	4.22	4.32	4.37	4.24
DHA[3]	0.00	0.00	0.02	0.01	0.00	0.00	0.00	0.00
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01
Omega 6[5]	21.37	22.54	24.86	25.42	21.37	20.92	20.81	20.54
Omega 3	4.22	4.98	6.05	6.50	4.22	4.32	4.40	4.35
Omega 6:Omega 3	3.31	3.42	3.67	3.72	3.31	3.22	3.22	3.20
SFA[5]	7.67	8.00	8.80	8.95	7.67	7.38	7.45	7.38
PUFA[6]	25.75	27.75	31.08	32.17	27.75	25.39	25.38	25.06
MUFA[7]	32.00	39.90	49.29	54.11	32.00	33.70	35.39	35.37
Total fatty acids	65.42	75.65	89.17	95.23	67.42	66.47	98.22	67.81

[1]LA, Linoleic acid,
[2]ALA, Alpha -Linolenic acid,
[3] DHA, docosahexaenoic acid,
[4] EPA, eicosapentaenoic acid,
[5] SFA, Saturated fatty acids,
[6] PUFA, Poly unsaturated fatty acids,
[7] MUFA, Mono unsaturated fatty acids
[8]Total fatty acids was calculated as SFA+PUFA+MUFA

ANOVA for SFA (Table 7.29) indicated the significant effect of meal and level on SFA content of wing samples and the effect of enzyme on the SFA content of breast samples. All the other parameters were not significantly different.

Table 7.29. ANOVA *P*-values for saturated fatty acids for carcass parts of broilers fed wheat-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.437	0.001	0.765
Level	0.320	0.012	0.670
Meal × level	0.460	0.396	0.671
Enzyme	0.005	0.116	0.850
Meal × enzyme	0.148	0.304	0.279
Level × enzyme	0.594	0.836	0.551
Meal × level × enzyme	0.342	0.280	0.348

Supplementing dietary Superzyme-OM^{TM} to the diets decreased the amount of SFA in breast samples (Table 7.30). Supplementing CM to the diets compared to JM caused a reduction of SFA content in wing meat and also 30% of meal supplementation in diet resulted in lower SFA content in the wing samples compared to 10%., which is a desirable effect of enzyme at this point, due to the negative effects of SFA on human cardiovascular disease (Simopoulos 1997).

Table 7.30. Saturated fatty acids (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	22.49±0.65	22.36±0.56	21.79±0.56	23.28±0.56
10%	20.95±0.56	20.78±0.56	21.92±0.56	21.95±0.56
20%	17.85±0.56	18.77±0.56	20.68±0.56	21.49±0.56
30%	16.23±0.56	16.55±0.56	20.75±0.56	21.59±0.56
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	7.32±1.67	3.70±1.67	6.23±1.67	3.38±1.67
10%	5.98±1.67	4.69±1.67	9.73±1.67	4.02±1.67
20%	3.55±1.67	4.23±1.67	9.21±1.67	3.40±1.67
30%	4.53±1.67	3.83±1.67	3.76±1.67	3.36±1.67
Enzyme	NO		YES	
	6.29±0.59*a*		3.82±0.59*b*	

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

The SFA content of wing samples increased when JM were supplemented (Table 7.31). The reason for this result is not clear.

Table 7.31. Saturated fatty acids (mg·g^{-1}) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Wing					
Meal	Canola meal		Juncea meal		Levels
Enzyme	NO	YES	NO	YES	
Levels					
0%	24.11±1.88	23.63±1.62	24.08±1.62	26.37±1.62	24.55±0.85*b*
10%	23.72±1.62	24.99±1.62	25.02±1.62	27.09±1.62	25.21±0.81*a*
20%	20.48±1.62	19.78±1.62	21.93±1.62	27.31±1.62	22.38±0.81*ab*
30%	18.77±1.62	20.54±1.62	24.46±1.62	23.39±1.62	21.79±0.81*b*
Meal	22.00±0.59*b*		24.96±0.59*a*		

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

SFA content of breast and thigh samples as a percentage of total fat in a diet can be seen in Appendix H.2.

ANOVA for PUFA (Table 7.32) indicated the significant main effect of enzyme on the PUFA content of breast samples and the main effect of meal inclusion level on the PUFA content of wing samples. No differences could be detected for the least squares means for other treatments (P.> 0.05).

Table 7.32. ANOVA *P* -values for polyunsaturated fatty acids for carcass parts of broilers fed wheat-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.934	0.104	0.239
Level	0.559	0.005	0.390
Meal × level	0.334	0.194	0.876
Enzyme	0.003	0.094	0.895
Meal × enzyme	0.158	0.981	0.425
Level × enzyme	0.702	0.517	0.582
Meal × level × enzyme	0.260	0.287	0.250

The PUFA content of wing samples of birds supplemented with 20 and 30% of meal inclusions was higher than for the birds fed the control diet (Table 7.34). The fatty acid analysis of diets (Table 7.28) indicated that the control diets had lower PUFA content than 20 and 30 % of meal inclusion. The PUFA content of control diets were lower than diets supplemented with 20 and 30 % of meal. Supplementing Superzyme-OM^{TM} to the diets decreased the amount of PUFA in the breast samples of birds ($P \leq 0.05$).

Table 7.34. Polyunsaturated fatty acids (mg·g^{-1}) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Wing					
Meal	Canola meal		Juncea meal		Levels
Enzyme	NO	YES	NO	YES	
Levels					
0%	22.14±1.75	22.98±1.51	23.40±1.51	23.05±1.51	22.89±0.79*b*
10%	23.39±1.51	26.85±1.51	23.39±1.51	25.44±1.51	24.53±0.76*ab*
20%	27.20±1.51	25.95±1.51	24.21±1.51	27.77±1.51	26.28±0.76*a*
30%	27.43±1.51	29.67±1.51	25.11±1.51	24.05±1.51	26.56±0.76*a*

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

The PUFA content of breast samples increased when canola meal was added compared to JM (Table 7.35). Finisher diets with CM supplemented had higher PUFA content compared to control and JM diets (Table 7.28) because higher percentage of canola oil added to the diets while cM was supplemented.

Table 7.35. Polyunsaturated fatty acids (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	7.78±1.13	5.94±1.13	6.31±1.13	6.67±1.13
10%	7.82±1.13	6.54±1.13	6.27±1.13	7.06±1.13
20%	7.24±1.13	10.17±1.13	7.32±1.13	7.15±1.13
30%	8.20±1.13	6.88±1.13	6.64±1.13	7.78±1.13
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	6.56±1.44	3.45±1.44	5.48±1.44	3.16±1.44
10%	5.74±1.44	4.66±1.44	8.83±1.44	3.79±1.44
20%	4.30±1.44	4.72±1.44	8.81±1.44	3.35±1.44
30%	5.92±1.44	4.70±1.44	3.77±1.44	3.34±1.44
Enzyme	NO		YES	
	6.18±0.51*a*		3.90±0.51*b*	

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

PUFA content of wing samples as a percentage of total fat in a diet can be seen in Appendix H.2.

ANOVA for MUFA (Table 7.36) indicated the significant increase due to enzyme on MUFA content of breast. All the other parameters were not significantly different (P>0.05).

Table 7.36. ANOVA *P* -values for monounsaturated fatty acids for carcass parts of broilers fed wheat-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.663	0.894	0.471
Level	0.383	0.221	0.797
Meal × Level	0.345	0.438	0.950
Enzyme	0.003	0.609	0.922
Meal × Enzyme	0.137	0.600	0.313
Level × Enzyme	0.603	0.843	0.533
Meal × Level × Enzyme	0.300	0.127	0.340

Enzyme supplementation decreased the MUFA content of breast meat compared to the birds that did not receive Superzyme-OM^{TM} supplementation (Table 7.37). The reason for this result is not clear. It might be related to SFA and PUFA in breast meat that increased by supplementing Superzyme-OM^{TM}.

Table 7.37. Monounsaturated fatty acids (mg·g^{-1}) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	18.58±3.33	12.43±3.33	13.19±3.33	14.43±3.33
10%	19.04±3.33	13.42±3.33	14.50±3.33	16.38±3.33
20%	14.26±3.33	21.79±3.33	15.39±3.33	15.67±3.33
30%	16.43±3.33	13.21±3.33	13.57±3.33	16.32±3.33
Wing				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	61.29±5.35	59.25±4.63	63.29±4.63	63.81±4.63
10%	65.51±4.63	68.73±4.63	66.10±4.63	71.22±4.63
20%	66.77±4.63	60.09±4.63	59.53±4.63	72.00±4.63
30%	68.23±4.63	73.62±4.63	68.30±4.63	60.85±4.63
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	15.54±4.17	6.65±4.17	12.10±4.17	5.95±4.17
10%	13.39±4.17	9.69±4.17	23.23±4.17	7.81±4.17
20%	7.78±4.17	9.24±4.17	21.42±4.17	6.18±4.17
30%	7.04±4.17	9.11±4.17	7.04±4.17	5.77±4.17
Enzyme	NO		YES	
	14.14±1.47*a*		7.55±1.47*b*	

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

MUFA content of wing samples as a percentage of total fat in a diet can be seen in Appendix H.3.The main effect of enzyme and the interaction level of meal ×enzyme was significantly different in wing samples.

Results of ANOVA analysis for LA (Table 7.38) indicated a significant main effect of enzyme on LA content of breast samples. No differences could be detected for the least squares means of meals, levels or their interactions (P <0.05).

Table 7.38. ANOVA *P* -values for Linoleic acid for carcass parts of broilers fed wheat-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.868	0.109	0.294
Level	0.515	0.067	0.555
Meal × Level	0.343	0.225	0.896
Enzyme	0.004	0.182	0.829
Meal × Enzyme	0.153	0.798	0.386
Level × Enzyme	0.666	0.416	0.587
Meal × Level × Enzyme	0.263	0.110	0.260

Table 7.39. Linoleic acid ($mg \cdot g^{-1}$) in thigh, breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	4.60±1.11	2.26±1.11	3.78±1.11	1.97±1.11
10%	3.92±1.11	3.09±1.11	6.35±1.11	2.49±1.11
20%	2.69±1.11	3.12±1.11	6.21±1.11	2.09±1.11
30%	3.88±1.11	3.03±1.11	2.33±1.11	2.11±1.11
Enzyme	NO		YES	
	4.22±0.39*a*		2.52±0.39*b*	
Wing				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	16.67±1.27	16.88±1.01	17.55±1.27	17.38±1.01
10%	17.27±1.01	19.98±1.01	16.75±1.01	18.76±1.01
20%	20.08±1.01	18.05±1.01	16.87±1.01	19.69±1.01
30%	19.79±1.01	21.33±1.01	18.45±1.01	17.36±1.01
Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	5.58±0.88	4.46±0.88	4.33±0.88	4.68±0.88
10%	5.53±0.88	4.52±0.88	4.35±0.88	5.01±0.88
20%	4.92±0.88	7.18±0.88	5.05±0.88	4.99±0.88
30%	5.57±0.88	4.59±0.88	4.48±0.88	5.43±0.88

[a-b] Means ± SEM with different superscripts for enzyme in breast are significantly different (α=0.05)

Supezyme-OM™ supplementation decreased the LA content of breast samples (Table 7.39).

LA content of wing samples as a percentage of total fat in a diet can be seen in Appendix H.4.The main effects of level and meal were significantly different in wing samples

ANOVA for ALA (Table 7.40) indicated the interaction effects of meal × level on wing samples and the significant main effect of enzyme on ALA content of breast samples.

Table 7.40. ANOVA *P*-values for alpha linolenic acid for carcass parts of broilers fed wheat-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.868	<.0001	0.056
Level	0.515	<.0001	0.212
Meal × Level	0.343	0.007	0.758
Enzyme	0.004	0.378	0.862
Meal × Enzyme	0.153	0.853	0.462
Level × Enzyme	0.666	0.466	0.529
Meal × Level× Enzyme	0.263	0.161	0.225

The ALA content of breast samples decreased when Superzyme-OM™ was supplemented (Table 7.41).

Table 7.41. Alpha linolenic acid (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.96±0.20	0.64±0.20	0.67±0.20	0.75±0.20
10%	1.05±0.20	0.83±0.20	0.74±0.20	0.86±0.20
20%	0.96±0.20	1.53±0.20	0.88±0.20	0.85±0.20
30%	1.15±0.20	0.89±0.20	0.75±0.20	0.94±0.20
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.88±0.26	0.32±0.26	0.66±0.26	0.28±0.26
10%	0.79±0.26	0.60±0.26	1.30±0.26	0.40±0.26
20%	0.50±0.26	0.59±0.26	1.29±0.26	0.30±0.26
30%	0.61±0.26	0.61±0.26	0.37±0.26	0.29±0.25
Enzyme	NO		YES	
	0.82±0.09*a*		0.42±0.09*b*	

[a-b] Means ± SEM with different superscripts for enzyme in are significantly different (α=0.05)

Superzyme-OM™ supplementation decreased the amount of ALA in breast samples (Table 7.41). thirty percent CM incorporation in diets resulted in higher ALA content in wing samples compared to control diets and all JM inclusion and 10% of CM inclusion (Table 7.42). ALA content of 30% CM in the diets was also higher than all other inclusions levels (Table 7.28)

Table 7.42. Alpha linolenic acid ($mg \cdot g^{-1}$) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Wing						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	3.43±0.32	3.56±0.28	3.54±0.28	3.52±0.28	3.50±0.21*c*	3.53±0.20*c*
10%	4.03±0.28	4.62±0.28	3.59±0.28	4.02±0.28	4.33±0.20*bc*	3.81±0.20*bc*
20%	4.89±0.28	4.34±0.28	3.67±0.28	4.25±0.28	4.62±0.20*ab*	3.96±0.20*bc*
30%	5.24±0.28	5.47±0.28	4.10±0.28	3.72±0.28	5.35±0.20*a*	3.91±0.20*bc*

[a-b] Means ± SEM with different superscripts for interaction means within cut of meat are significantly different (α=0.05)

ALA content of wing, thigh and breast samples as a percentage of total fat in a diet can be seen in Appendix H.5. The mean effect of level and meal were significantly different in thigh samples and the interaction effects of level ×enzyme and meal ×level were significantly different in breast samples.

ANOVA for DHA (Table 7.43) indicated interaction effect for meal × level on DHA content of thigh samples, the main effect of meal inclusion level and the main effect of enzyme on DHA content of wing sample.

Table 7.43. ANOVA *P*-values for docosahexaenoic acid for carcass parts of broilers fed wheat-based diet

	Tissue		
Effect	Breast	Wing	Thigh
Meal	0.141	0.917	0.039
Level	0.081	0.002	<.0001
Meal × Level	0.476	0.708	0.014
Enzyme	0.017	0.770	0.354
Meal × Enzyme	0.673	0.672	0.572
Level × Enzyme	0.328	0.148	0.138
Meal × Level × Enzyme	0.782	0.893	0.804

Twenty percent and 30% inclusion of CM caused higher incorporation of DHA in thigh meat compared to chickens fed 0 and 10% CM (Table 7.44) this is because of the highest levels of canola oil in 20 and % of CM inclusion. In wing samples 30%, meal inclusion levels increased the DHA content of meat compared to 0 and 10% (Table 7.34). Superzyme-OM^{TM} decreased the DHA in breast samples (Table 7.45).

Table 7.44. Docosahexaenoic acid (mg·g^{-1}) in thigh meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	0.01±0.01	0.01±0.01	0.11±0.01	0.11±0.01	0.10±0.01*c*	0.11±0.01*bc*
10%	0.01±0.01	0.10±0.01	0.09±0.01	0.10±0.01	0.10±0.01*c*	0.09±0.01*c*
20%	0.15±0.01	0.14±0.01	0.13±0.01	0.10±0.01	0.14±0.01*ab*	0.12±0.01*bc*
30%	0.15±0.01	0.15±0.01	0.13±0.01	0.12±0.01	0.15±0.01*a*	0.12±0.01*abc*
Wing						
Meal	Canola meal		Juncea meal		Level	
Enzyme	NO	YES	NO	YES		
Levels						
0%	0.12±0.02	0.13±0.01	0.13±0.01	0.13±0.01	0.13±0.01*bc*	
10%	0.11±0.01	0.13±0.01	0.11±0.01	0.13±0.01	0.12±0.01*c*	
20%	0.17±0.01	0.15±0.01	0.14±0.01	0.14±0.01	0.15±0.01*ab*	
30%	0.16±0.01	0.14±0.01	0.16±0.01	0.15±0.01	0.15±0.01*a*	

[a-b] Means ± SEM with different superscripts for interaction means in thigh and for levels in wing are significantly different (α=0.05)

Table 7.45. Docosahexaenoic acid (mg·g^{-1}) in breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.10±0.01	0.08±0.01	0.08±0.01	0.09±0.01
10%	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
20%	0.12±0.01	0.09±0.01	0.11±0.01	0.08±0.01
30%	0.12±0.01	0.10±0.01	0.10±0.01	0.08±0.01
Enzyme	NO		YES	
	1.00. ±0.00*a*		0.08±0.00*b*	

[a-b] Means ± SEM with different superscripts for enzyme within cut of meat are significantly different (α=0.05)

Results of ANOVA analysis for EPA (Table 7.46) indicated the main effect of level was significant for the EPA content of wing samples and the main effect of enzyme was significant for EPA content of breast and thigh samples.

Table 7.46. ANOVA *P*-values for eicosapentaenoic acid for carcass parts of broilers fed wheat-based diet

Effect	Tissues		
	Breast	Wing	Thigh
Meal	0.319	0.786	0.845
Level	0.454	<.001	0.738
Meal × Level	0.364	0.340	0.986
Enzyme	0.000	0.068	0.043
Meal × Enzyme	0.261	0.177	0.072
Level × Enzyme	0.485	0.418	0.542
Meal × Level × Enzyme	0.165	0.575	0.426

The EPA content of thigh and breast muscles decreased in birds fed Superzyme-OM^{TM} (Table 7.47). Birds fed 20 and 30% of meal inclusion had higher amount of EPA in their wing muscles compared to the control group (Table7.48).

Table 7.47. Eicosapentaenoic acid (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM^{TM} (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.07±0.00	0.06±0.00	0.06±0.00	0.07±0.00
10%	0.07±0.00	0.06±0.00	0.07±0.00	0.06±0.00
20%	0.07±0.00	0.07±0.00	0.07±0.00	0.07±0.00
30%	0.07±0.00	0.06±0.00	0.06±0.00	0.07±0.00
Enzyme	NO		YES	
	0.07±0.00*a*		0.06±0.00*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.06±0.00	0.05±0.00	0.05±0.00	0.05±0.00
10%	0.06±0.00	0.05±0.00	0.06±0.00	0.05±0.00
20%	0.06±0.00	0.05±0.00	0.06±0.00	0.05±0.00
30%	0.06±0.00	0.05±0.00	0.04±0.00	0.05±0.00
Enzyme	NO		YES	
	0.06±0.00*a*		0.05±0.00*b*	

[a-b] Means ± SEM with different superscripts for enzyme within cut of meat are significantly different (α=0.05)

Table 7.48. Eicosapentaenoic acid (mg·g^{-1}) in wing meat of broilers fed different levels of juncea meal or canola meal with and without Superzyme-OM^{TM} (least square mean ±SE)

Wing					
Meal	Canola meal		Juncea meal		Levels
Enzyme	NO	YES	NO	YES	
Levels					
0%	0.10±0.01	0.09±0.01	0.10±0.01	0.10±0.01	0.10±0.00*c*
10%	0.11±0.01	0.10±0.01	0.11±0.01	0.11±0.01	0.11±0.00*bc*
20%	0.12±0.01	0.11±0.01	0.11±0.01	0.12±0.01	0.11±0.00*ab*
30%	0.14±0.01	0.11±0.01	0.12±0.01	0.11±0.01	0.12±0.00*a*

[a-b] Means ± SEM with different superscripts for levels within cut of meat are significantly different (α=0.05)

EPA content of breast and wing samples as a percentage of total fat in a diet can be seen in Appendix H.6. The effect of meal × level × enzyme was significantly different in breast samples and the main effect of level and meal were also significantly different in thigh samples.

ANOVA for Omega 3 (Table 7.49) indicated a significant effect of meal × level on Omega 3 content of wing samples, the main effect of enzyme on breast samples and the main effect of meal on Omega 3 in thigh samples. No differences were be detected for the other least squares means using Tukey-Kramer.

Table 7.49. ANOVA *P*-values for Omega 3 for carcass parts of broilers fed wheat-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.730	<.0001	0.038
Level	0.518	<.0001	0.087
Meal × Level	0.290	0.008	0.668
Enzyme	0.002	0.524	0.977
Meal × Enzyme	0.190	0.847	0.485
Level × Enzyme	0.721	0.430	0.564
Mea l × Level × Enzyme	0.236	0.164	0.194

The effect of CM and JM on wing samples (Table 7.50) and thigh samples indicated that the birds fed CM had higher Omega 3 content than birds fed JM. The lower Omega 3 content in breast samples were observed for diets supplemented with Superzyme-OM^{TM}. The interaction effect of meal × level showed that diets with 30% CM with the highest dietary level of Omega 3 had higher amount of Omega 3 in wing samples compared to all inclusion levels of JM and 0 and 10% of CM (Table 7.51).

Table 7.50. Omega 3 (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.31±0.21	0.95±0.21	1.04±0.21	1.08±0.21
10%	1.43±0.21	1.17±0.21	1.07±0.21	1.19±0.21
20%	1.39±0.21	1.98±0.21	1.29±0.21	1.20±0.21
30%	1.63±0.21	1.35±0.21	1.15±0.21	1.35±0.21
Meal	1.40±0.08*a*		1.17±0.08*b*	
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	1.21±0.28	0.55±0.28	0.94±0.28	0.54±0.28
10%	1.01±0.28	0.87±0.28	1.65±0.28	0.66±0.28
20%	0.86±0.28	0.89±0.28	1.67±0.28	0.56±0.28
30%	1.21±0.28	0.94±0.28	1.20±0.28	0.55±0.28
Enzyme	NO		YES	
	1.16±0.01*a*		0.70±0.01*b*	

[a-b] Means ± SEM with different superscripts for meal in thigh and for enzyme in breast within the same columns are significantly different (α=0.05)

Table 7.51. Omega 3 (mg·g^{-1}) in thigh meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Wing						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	4.07±0.35	4.13±0.30	4.22±0.30	4.13±0.30	4.01±0.23*c*	4.17±0.21*c*
10%	4.69±0.30	5.31±0.30	4.26±0.30	4.68±0.30	5.00±0.21*bc*	4.47±0.21*bc*
20%	5.63±0.30	5.06±0.30	4.36±0.30	4.99±0.30	5.35±0.21*ab*	4.67±0.21*bc*
30%	6.04±0.30	6.21±0.30	4.87±0.30	4.41±0.30	6.13±0.21*a*	4.64±0.21*bc*

[a-b] Means ± SEM with different superscripts for interaction means within cut of meat are significantly different (α=0.05)

Omega3 content of wing and thigh samples as a percentage of total fat in a diet can be seen in Appendix H.7. The interaction effect of meal × level in wing and the main effect of level in thigh samples were significantly different.

ANOVA for Omega 6 (Table 7.52) indicated a significant main effect of enzyme on Omega 6 content of breast meat. All other effects were not significantly different (P>0.05).

Table 7.52. ANOVA *P*-values for Omega 6 for carcass parts of broilers fed wheat-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.857	0.148	0.326
Level	0.558	0.056	0.480
Meal × Level	0.346	0.215	0.892
Enzyme	0.003	0.219	0.863
Meal × Enzyme	0.154	0.847	0.410
Level × Enzyme	0.696	0.399	0.589
Meal × Level × Enzyme	0.268	0.113	0.265

Enzyme supplementation decreased the amount of Omega 6 in breast samples (Table 7.53). Thirty percent meal inclusion increased the Omega 6 content in wing samples compared to the control diets (Table 7.54).Omega 6, content of control diets was lower than the highest meal inclusion level (Table 7.28).

Table 7.53. Omega 6 (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM}(least square mean ±SE)

Thigh				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	6.33±0.90	4.87±0.90	5.15±0.90	5.45±0.90
10%	6.26±0.90	5.25±0.90	5.08±0.90	5.75±0.90
20%	5.74±0.90	8.05±0.90	5.90±0.90	5.81±0.90
30%	6.44±0.90	5.42±0.90	5.37±0.90	6.29±0.90
Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	5.23±1.15	2.81±1.15	4.42±1.15	2.53±1.15
10%	4.53±1.15	3.70±1.15	7.05±1.15	3.04±1.15
20%	3.35±1.15	3.72±1.15	6.98±1.15	2.70±1.15
30%	4.61±1.15	3.66±1.15	3.00±1.15	2.70±1.15
Enzyme	NO		YES	
	4.89±0.41*a*		3.11±0.41*b*	

[a-b] Means ± SEM with different post scripts for enzyme in breast are significantly different (α=0.05)

Table 7.54. Omega 6 (mg·g^{-1}) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM^{TM} (least square mean ±SE)

Wing					
Meal	Canola meal		Juncea meal		Levels
Enzyme	NO	YES	NO	YES	
Levels					
0%	17.79±1.30	17.96±1.13	18.90±1.13	18.65±1.13	18.32±0.59b
10%	18.44±1.13	21.28±1.13	17.95±1.13	19.87±1.13	19.38±0.56ab
20%	21.31±1.13	19.28±1.13	18.15±1.13	21.02±1.13	19.94±0.56ab
30%	21.14±1.13	22.56±1.13	19.89±1.13	18.63±1.13	20.55±0.56a

[a-b] Means ± SEM with different post scripts for enzyme in breast are significantly different (α=0.05)

Omega6 content of wing samples as a percentage of total fat in a diet can be seen in Appendix H.8.The main effects of level and meal are significantly different in wing samples.

ANOVA for the ratio of Omega 6:Omega 3 (Table 7.55) indicated a significant effect of meal × level on the ratio of Omega 6:Omega 3 for wing and thigh samples. The main effects of meal and level were also significant for the ratio of Omega 6:Omega 3 in breast samples. Enzyme supplementation significantly affected the Omega 6:Omega3 ratio of breast and wing samples.

Table 7.55. ANOVA *P*-values for Omega 6:Omega3 (mg.g^{-1}) for carcass parts of broilers fed wheat-based diet

Effect	Tissue		
	Breast	Wing	Thigh
Meal	0.000	<.0001	<.0001
Level	0.012	<.0001	<.0001
Meal × Level	0.125	0.000	<.0001
Enzyme	0.004	0.070	0.136
Mea l× Enzyme	0.915	0.961	0.985
Level × Enzyme	0.776	0.660	0.965
Meal× Level× Enzyme	0.111	0.975	0.221

The meal × level interaction effect of Omega 6:Omega 3 in thigh samples indicated that birds fed 20 or 30 % of CM had the lowest. Meal × level interaction effect in wing samples, showed 30% of CM inclusion in diets, resulted in the lowest Omega 6: Omega 3 ratio (Table 7.56). Breast samples from birds given CM and Superzyme-OM^{TM} had a reduced ratio of Omega 6: Omega 3 (Table 7.57). Meal inclusion levels for Omega 6: Omega 3 in breast samples indicated that diets supplemented with 20% and 30% of meal had a significantly lower ratio compared to the control group (Table 7.57).

Table 7.56. Omega 6:Omega 3 in thigh and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Thigh						
					Meal × Level	
Meal	Canola meal		Juncea meal		Canola meal	Juncea meal
Enzyme	NO	YES	NO	YES		
Levels						
0%	4.94±0.10	5.14±0.10	5.02±0.10	5.03±0.10	5.04±0.07*a*	5.03±0.07*ab*
10%	4.45±0.10	4.49±0.10	4.78±0.10	4.84±0.10	4.47±0.07*d*	4.81±0.07*abc*
20%	4.16±0.10	4.09±0.10	4.60±0.10	4.86±0.10	4.12±0.07*e*	4.73±0.07*bcd*
30%	3.95±0.10	4.07±0.10	4.70±0.10	4.67±0.10	4.01±0.07*e*	4.68±0.07*cd*
Wing						
Meal	Canola meal		Juncea meal		Meal × Level	
Enzyme	NO	YES	NO	YES	Canola meal	Juncea meal
Levels						
0%	4.38±0.07	3.39±0.07	4.51±0.07	4.51±0.07	4.39±0.05*ab*	4.50±0.05a
10%	3.94±0.07	4.03±0.07	4.22±0.07	4.26±0.07	3.98±0.05*de*	4.24±0.05*bc*
20%	3.78±0.07	3.81±0.07	4.16±0.07	4.21±0.07	3.80±0.05*e*	4.19±0.05*bcd*
30%	3.51±0.07	3.64±0.07	4.09±0.07	4.21±0.07	3.57±0.05*f*	4.16±0.05*cd*

[a-d] Means ± SEM with different postscripts for interaction means within cut of meat are significantly different (α=0.05)

Table 7.57. Omega6:Omega3 in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast					
Meal	Canola meal		Juncea meal		Levels
Enzyme	NO	YES	NO	YES	
Levels					
0%	4.29±0.20	5.11±0.20	4.84±0.20	4.87±0.20	4.75±0.01*a*
10%	4.28±0.20	4.25±0.20	4.34±0.20	4.74±0.20	4.39±0.01*ab*
20%	3.90±0.20	4.22±0.20	4.31±0.20	4.84±0.20	4.32±0.01*b*
30%	3.93±0.20	4.03±0.20	4.61±0.20	4.88±0.20	4.36±0.01*b*
Meal	4.25±0.07*b*		4.67±0.07*a*		
Enzyme	NO		YES		
	4.31±0.07*b*		4.61±0.07*a*		

[a-b] Means ± SEM with different postscripts for level, meal and enzyme within cut of meat are significantly different (α=0.05)

The fatty acid profile of samples from birds fed CM compared to JM in wheat-based diets can be explained in similar fashion to corn-based diet. Supplementation of increased levels of canola oil in the starter (Table 4.2), grower (Table 4.4) and finisher (Table 4.6) diets with CM impacted the comparison of CM to JM.

As ALA was the only source of Omega 3 fatty acids in the diets and the amount of EPA and DHA in the diets were 0.00 mg g^{-1} , the presence of EPA and DHA in meat samples shows the production of these fatty acids in the birds' bodies from the precursor ALA. This supports the findings by Betti et al. (2009). The content of PUFA, MUFA, LA, ALA, EPA, DHA, Omega 3 and Omega 6 were lower ($P \leq 0.05$) in breast meat when Superzyme-OM^{TM} was not supplemented in feed. The reason for this is not clear and it is contrary to the study by Meng et al. (2006) and Slominski et al. (2006) who indicated the addition of multicarbohydrase enzyme in the diet of broilers containing oilseeds can improve oil utilization. The reason for failure of the enzyme addition to improve the fatty acid profiles of the meat was probably due to higher viscosity effects of wheat-based diets and the lack of enzyme to overcome the viscosity activity to improve oil utilization from diets (Meng et al. 2006).

7.7. General discussion

The effects of fatty acid composition of diets on fatty acid compositions in meat samples can be observed in the results of the current study. The synthesis of two fatty acids EPA and DHA, in the meat samples was evidence that birds had the ability to convert ALA in diets to EPA and DHA .These fatty acids are required in human diets (Simopoulos 1999).

The increase in desirable fatty acids through supplementation more CM in diets compared with JM was observed in some of our results. This is likely due to addition of more canola oil to the diets supplemented with CM. Further investigation are required in order to analyze the fatty acid profiles of JM or CM to make sure CM has different fatty acid profile with more omega 3 fatty acids than JM.

Different research showed the reduction in fat digestion due to high digesta viscosity in birds (Meng et al. 2004). As fat digestion improves by combination of bile

acids, lipase and colipase in birds, supplementing lipase to the diets might facilitate fat digestion and make canola oil more available to birds.

7.8. Conclusions

Supplementing CM and JM both had positive effects on fatty acid profiles of different tissues. When adding Superzyme-OM^{TM}, more attention is required in wheat-based diets due to some negative effects of enzyme on fatty acid profiles in meat samples. Supplementing corn-based diets with Superzyme-OM^{TM} had a positive effect on fatty acid profiles in the meat. Studies are required to evaluate the effects of oxidation and shelf life of the meat when the n-3 fatty acids increase in the meat samples.The results showed that the fatty acid content of meat samples can be modified by dietary addition of canola or juncea meals.

CHAPTER 8: CONCLUSIONS

Canola meal or JM can be included in broiler diets with Superzyme- OM^{TM} at higher levels than the current recommendation levels of 10% in starter and 20% in grower (CCC 2009). The levels recommended to inclusions in broiler diets based on the current studies are 30% of CM for the starter period in both corn- and wheat-based diets, 30% and 20% CM in the grower for both corn and wheat-based diets, respectively. Ten percent CM for corn-and 20% wheat-based diets during the finisher phase. For turkeys, the AME_n in CM and JM, were 2331± 200 kcal kg $^{-1}$ and 2215 ± 200 kcal kg $^{-1}$ respectively. Turkeys are able to use more AME_n from both meals than chickens. The AME_n values of CM and JM for broilers were 2006 and 1867 kcal kg^{-1}, respectively Liver weight, visual scoring of colour and texture and colorimeter data indicated that enzyme inclusion in both trials resulted in higher ($P \leq 0.05$) liver weight. Carcass composition measured as fat, protein, ash and dry matter indicated, increasing the DM, fat and protein in the carcasses of broilers fed corn-based diets .DM in carcasses of birds fed wheat-based diet when Superzyme-OM^{TM} was supplemented. Fatty acid analyses indicated higher amounts of PUFA, MUFA, LA and Omega 6 in wing, thigh and breast of birds fed CM compared to JM in corn-based diets. The Omega 6: Omega 3 ratio was lower in breast and wing samples of broilers fed supplemented Superzyme- OM^{TM} compared to those without dietary enzyme in corn-based diets. In wheat based diets, SFA decreased in breast samples when enzyme was added and in thigh samples when CM was included. The Omega 6: Omega 3 ratio was lower in breast samples when CM was fed to bird compared to JM in wheat-based diets. Most of muscle samples of birds in both trials supplemented with CM had the better fatty acid profile than those supplemented with JM.

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Appendix A. Amino acid profile (% as fed) of canola meal (Brassica napus) and juncea meal (Brassica juncea)

	Meals	
Amino acids	Canola	Juncea
Aspartic acid	3.30	3.23
Threonine	2.33	2.42
Serine	2.44	2.26
Glutamic acid	4.67	4.59
Proline	1.91	1.72
Glycine	3.37	3.43
Alanine	2.32	2.33
Cysteine	0.88	0.76
Valine	0.78	0.76
Methionine	0.78	0.76
Isoleucine	1.49	1.60
Leucine	2.81	2.89
Tyrosine	0.71	1.11
Phenylalanine	1.66	1.68
Histidine	1.09	1.08
Lysine	2.38	2.23
Arginine	2.36	2.64
Tryptophan	0.49	0.46

[1]Analyzed by an acid hydrolysis procedure at the University of Manitoba, Department of Animal Science

Appendix B. Lighting and temperature schedules for broiler chickens housed at the Atlantic Poultry Research Centre during digestibility trial

Days post hatch	Temperature (°C)	Light Hours	Light Intensity (lux)
1-2	32	24	20
3-4	31	23	20
5-6	30	16	20
7-9	30	16	15
10-11	29	16	10
12-13	28	16	10
14-16	27	16	10
17-18	26	16	10
19-20	25	16	10
21	24	16	10

Appendix C. Lighting and temperature schedules for turkeys housed at the Poultry Research Centre during digestibility trial

Days post hatch	Temperature (°C)	Light Hours	Light Intensity (lux)
1-2	35	24	20
3-4	3.5	23	20
5	32.5	16	20
6-9	32	16	15
10-11	31	16	10
12-14	30	16	5
15-16	29	16	5
17-18	28	16	5
19-20	27	16	5
21-23	26	16	5
24-25	25	16	5
26-28	24	16	5

Appendix D. Lighting and temperature schedules for broiler chickens housed at the Atlantic poultry institute during broilers grower trial (corn and wheat-based)

Days post hatch	Temperature (°C)	Light Hours	Light Intensity (lux)
1-2	30	24	20
3-4	29	23	20
5-6	28	16	20
7-9	28	16	15
10-11	27	16	10
12-13	27	16	10
14-16	26	16	10
17-18-	25	16	10
19-20	24	16	10
21-23	23	16	10
24-27	23	16	10
28	22.5	16	10
29-32	22.5	17	10
33-35	22.5	18	10

Appendix E. Analyzed values for Canola meal (*Brassica napus*) and Juncea (Brassica juncea) meal (% dry matter basis)

	Canola meal	Juncea meal
Crude Protein[1]	43.80	43.92
Moisture	11.80	10.79
Crude Fat	2.17	1.93
Crude Fiber	9.64	8.25
Ash	8.79	8.23
ADF	20.44	15.08
NDF	30.60	22.27
Starch	0.00	1.86
Calcium	0.75	0.80
Phosphorus	1.43	1.57
Taurine	0.09	0.09
Hydroxyproline	0.36	0.26
Aspartic Acid	3.03	3.31
Threonine	1.83	1.80
Serine	1.71	1.63
Glutamic Acid	6.86	6.88
Proline	2.48	2.30
Lanthionine	0.02	0.02
Glycine	2.12	2.20
Alanine	1.89	1.97
Cysteine	1.04	0.93
Valine	1.94	2.23
Methionine	0.86	0.83
Isoleucine	1.46	1.76
Leucine	2.94	3.15
Tyrosine	1.17	1.20
Phenylalanine	1.64	1.74
Hydroxylysine	0.15	0.06
Ornithine	0.01	0.01
Lysine	2.27	2.25
Histidine	1.09	1.12
Arginine	2.54	2.84
Tryptophan	0.49	0.46
Available Lysine	2.05	2.08

[1] Percentage N X 6.25. W/W%= grams per 100 grams of sample.
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Appendix F. ANOVA *P*-values for liver a*, b* and L* score in corn based diet (Corn–based trial)

Effect	a* score	b* score	L* score
Meal	0.441	0.994	0.736
Level	0.591	0.867	0.679
Meal × Level	0.287	0.062	0.500
Enzyme	0.264	0.369	0.556
Meal × Enzyme	0.491	0.830	0.107
Level × Enzyme	0.459	0.733	0.973
Meal × Level × Enzyme	0.960	0.853	0.566

AppendixG.1. Fatty acid content of sample (% of total fat) of corn-based diets supplemented with canola or juncea meals

Starter								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	34.98	31.76	28.80	28.04	34.98	34.72	33.81	32.30
ALA[2]	6.29	6.63	6.91	6.83	6.29	6.34	6.26	6.18
DHA[3]	0.00	0.00	0.03	0.04	0.00	0.03	0.02	0.03
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.01	0.00	0.00
Omega 6[5]	35.04	31.81	28.86	28.09	35.04	34.77	33.87	32.36
Omega 3	6.29	6.63	6.95	6.89	6.29	6.41	6.29	6.22
Omega 6:Omega 3	5.57	4.80	4.16	4.08	5.57	5.43	5.39	5.20
SFA[5]	12.08	10.53	9.68	9.41	12.08	11.66	11.36	11.16
PUFA[6]	41.63	38.66	36.07	35.23	41.63	41.51	40.24	38.91
MUFA[7]	46.30	50.80	54.25	55.36	46.30	46.83	48.16	49.93
Grower								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	35.10	32.89	30.30	27.76	35.10	34.01	32.87	31.41
ALA[2]	6.23	6.21	6.44	6.72	6.23	6.11	6.07	6.07
DHA[3]	0.00	0.00	0.02	0.00	0.00	0.00	0.02	0.02
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Omega 6[5]	35.15	32.94	30.36	27.82	35.15	34.07	32.94	31.47
Omega 3	35.10	32.89	30.30	27.76	35.10	34.01	32.87	31.41
Omega 6:Omega 3	5.64	5.31	4.69	4.13	5.64	5.57	5.41	5.16
SFA[5]	11.79	10.74	10.08	9.29	11.79	11.43	11.15	10.86
PUFA[6]	41.62	39.41	37.05	34.77	41.62	40.44	39.27	37.85
MUFA[7]	46.58	49.85	52.87	55.93	46.58	48.13	49.58	51.29
Finisher								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	36.43	33.65	30.47	28.73	36.43	36.79	34.82	34.32
ALA[2]	5.73	6.01	6.34	6.44	5.73	5.81	5.74	5.61
DHA[3]	0.00	0.00	0.00	0.05	0.00	0.00	0.00	0.00
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Omega 6	36.48	33.70	30.53	28.78	36.48	34.87	34.38	33.68
Omega 3	36.43	33.65	30.47	28.73	36.43	34.82	34.32	33.63
Omega 6:Omega 3	6.36	5.61	4.81	4.41	6.36	6.01	5.99	6.00
SFA[5]	11.60	10.61	9.61	9.32	11.60	11.04	11.11	10.80
PUFA[6]	37.85	39.92	37.08	35.44	37.85	40.90	40.36	39.53
MUFA[7]	45.99	49.47	53.30	55.23	45.99	48.05	48.53	49.67

[1]LA, Linoleic acid,
[2]ALA, Alpha -Linolenic acid,
[3] DHA, docosahexaenoic acid,
[4] EPA, eicosapentaenoic acid,
[5] SFA, Saturated fatty acids,
[6] PUFA, Poly unsaturated fatty acids,
[7] MUFA, Mono unsaturated fatty acids
[8]Total fatty acids was calculated as SFA+PUFA+MUFA

AppendixG.2. Saturated fatty acids (% of total fat) in breast, thigh and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast		
	Meal × Level	
	Canola meal	Juncea meal
Levels		
0%	26.06±0.46*a*	25.32±0.46*ab*
10%	23.48±0.46*bc*	23.74±0.46*b*
20%	21.47±0.46*cd*	24.48±0.46*ab*
30%	20.28±0.46*d*	23.94±0.46*b*
Thigh		
	Meal × Level	
Levels	Canola meal	Juncea meal
0%	25.07±0.34*a*	24.19±0.34*ab*
10%	22.15±0.34*c*	22.49±0.34*c*
20%	19.59±0.34*d*	22.83±0.34*bc*
30%	18.71±0.34*d*	21.96±0.34*c*
Wing		
	Meal × Level	
Levels	Canola meal	Juncea meal
0%	23.58±0.34*a*	22.72±0.34*a*
10%	20.78±0.34*b*	20.88±0.34*b*
20%	18.30±0.35*c*	21.08±0.37*b*
30%	17.36±0.34*c*	20.75±0.3*b*

ANOVA *P*-value			
Meal × Level	Breast	Thigh	Wing
	<.001	<.001	<.001

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

AppendixG.3. Poly unsaturated fatty acids (mg g^{-1}) in wing ,thigh and breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Wing		
	Meal × Level	
	Canola meal	Juncea meal
Levels		
0%	2331±0.45*c*	23.09±0.45*bc*
10%	23.57±0.45*abc*	23.23±0.45*abc*
20%	25.17±0.45*a*	23.22±0.45*abc*
30%	24.64±0.45 *ab*	23.62±0.45*abc*
Thigh		
	Meal × Level	
	Canola meal	Juncea meal
Level		
0%	22.29±0.46*b*	23.10±0.46*ab*
10%	23.13±0.46*ab*	23.09±0.46*ab*
20%	24.91±0.46*a*	23.16±0.46*ab*
30%	24.13±0.46*ab*	23.81±0.46*ab*
Breast		
	Meal × Level	
Level	Canola meal	Juncea meal
0%	24.20±0.52*b*	25.81±0.52*ab*
10%	25.35±0.52*ab*	25.25±0.52*ab*
20%	27.18±0.52*a*	25.61±0.52*ab*
30%	26.27±0.52*ab*	26.93±0.52*a*

Enzyme × Meal				
	Canola meal		Juncea meal	
	NO	YES	NO	YES
	25.60±0.37*a*	25.90±0.37*a*	26.56±0.37*a*	25.24±0.37*a*

ANOVA –*P* value			
	Breast	Wing	Thigh
Meal × Level	0.027	0.029	0.056
Enzyme × Meal	0.033	0.499	0.436

[a-c] Means ± SEM with different postscripts for interaction means for meals within cut of meat are significantly different (α=0.05)

AppendixG.4. Monounsaturated fatty acids (% of total fat) in breast, wing and thigh meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast		
Meal × Level		
Meal	Canola meal	Juncea meal
Level		
0%	49.74±0.78*b*	48.87±0.78*b*
10%	51.17±0.78*ab*	51.02±0.78*ab*
20%	51.35±0.78*ab*	49.91±0.78*b*
30%	53.45±0.78*a*	49.13±0.78*b*
Wing		
Meal × Level		
Meal	Canola meal	Juncea meal
Level		
0%	54.11±0.38*c*	54.19±0.4*c*
10%	55.65±0.38*bc*	55.89±0.38*b*
20%	56.53±0.38*ab*	55.70±0.38*bc*
30%	58.00±0.38*a*	55.64±0.38*bc*
Thigh		
Meal × Level		
Meal	Canola meal	Juncea meal
Level		
0%	52.65±0.42*c*	52.71±0.42*c*
10%	54.71±0.42*b*	54.42±0.42*bc*
20%	55.50±0.42*ab*	54.02±0.42*bc*
30%	57.16±0.42*a*	54.24±0.42*bc*

ANOVA –*P* value			
	Breast	Wing	Thigh
Meal ×Level	0.05	0.00	0.00

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

AppendixG.5. Alpha linolenic acid (% of total fat) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Thigh			
Levels			
0%		2.66±0.10b	
10%		2.78±0.10b	
20%		3.17±0.10a	
30%		3.19±0.10a	
Breast			
Meal ×Level			
	Canola meal		Juncea meal
Levels			
0%	2.48±0.11*c*		2.58±0.11*c*
10%	2.91±0.11*bc*		2.67±0.11*c*
20%	3.37±0.11*ab*		2.63±0.11*c*
30%	3.57±0.11*a*		2.67±0.11*c*
Enzyme	NO		YES
	2.75±0.05b		2.97±0.06a
Wing			
Meal ×Level			
	Canola meal		Juncea meal
Levels			
0%	2.85±0.09*c*		2.96±0.09*c*
10%	3.37±0.09*b*		3.01±0.09*bc*
20%	3.96±0.09*a*		3.18±0.09*bc*
30%	4.07±0.09*a*		3.20±0.09*bc*
Enzyme	NO		YES
	3.27±0.05*b*		3.40±0.05*a*
ANOVA –*P* value			
	Breast	Wing	Thigh
Level	0.103	0.126	0.126
Meal ×Level	0.118	0.080	0.104

[a-b] Means ± SEM with different postscripts for meals in thigh and breast and for levels in thigh are significantly different (α=0.05)

AppendixG.6. Docosahexaenoic acid (% of total fat) in breast and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Enzyme × Meal	0.43±0.04*b*	0.46±0.04*b*	0.63±0.04*a*	0.44±0.04*b*
Wing				
Enzyme	NO		YES	
	0.15±0.00*a*		0.14±0.00*b*	
ANOVA –*P* value				
	Breast	Wing	Thigh	
Enzyme	0.057	0.013	0.477	
Meal × Enzyme	0.017	0.073	0.731	

[a-d] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

AppendixG.7. Eicosapentaenoic acid (% of total fat) in breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

	Breast			
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Enzyme × Meal	0.20±0.02*a*	0.23±0.02*a*	0.26±0.02*a*	0.20±0.02*a*

ANOVA –*P* value			
	Breast	Wing	Thigh
Meal × Enzyme	0.051	0.067	0.522

[a-d] Means ± SEM with different letters within interaction means are significantly different (α=0.05)

AppendixG.8. Omega 3 (% of total fat) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast				
	Meal × Level			
Meal	Canola meal		Juncea meal	
Levels				
0%	3.75±0.11*c*		4.19±0.11*b*	
10%	4.25±0.11*bc*		4.05±0.11*bc*	
20%	5.02±0.11*a*		4.14±0.11*bc*	
30%	5.09±0.11*a*		4.48±0.11*b*	
Meal × Enzyme	Canola meal		Juncea meal	
	NO	YES	NO	YES
	4.38±0.08*ab*	4.68±0.08*a*	4.30±0.08*b*	4.13±0.08*b*
Thigh				
	Meal × Level			
Canola meal			Juncea meal	
Levels				
0%	3.75±0.11*c*		4.19±0.11*bc*	
10%	4.25±0.11b*c*		4.05±0.11*bc*	
20%	5.02±0.11*a*		4.14±0.11*bc*	
30%	5.09±0.11*a*		4.48±0.11*b*	
	Canola meal		Juncea meal	
Meal × Enzyme	NO	YES	NO	YES
	4.38±0.08*ab*	4.68±0.08*a*	4.30±0.08*b*	4.13±0.08*b*
ANOVA –*P* value				
	Breast	Wing	Thigh	
Meal × Enzyme	0.005	0.284	0.005	
Meal × Level	<.001	<.001	<.001	

[a-c] Means ± SEM with different postscripts for interaction means within cut of meat are significantly different (α=0.05)

Appendix H.1. Fatty acid content (% of total fat) of wheat-based diets supplemented with canola or juncea meals

Starter								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	32.24	29.47	24.46	25.96	32.24	30.66	30.53	29.13
ALA[2]	6.73	7.02	7.08	7.38	6.73	7.47	6.95	6.74
DHA[3]	0.00	0.00	0.00	0.00	0.00	0.00	0.05	0.07
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.04	0.02
Omega 6[5]	32.31	29.55	26.53	26.03	32.31	30.74	30.60	29.20
Omega 3	6.83	7.09	7.16	7.44	6.83	7.51	7.09	6.91
Omega 6:Omega 3	4.73	4.17	3.71	3.50	4.73	4.09	4.31	4.23
SFA[5]	12.38	11.05	10.01	9.77	12.38	11.35	11.41	11.07
PUFA[6]	39.47	36.95	33.99	33.76	39.47	38.55	38.02	36.43
MUFA[7]	48.14	52.00	56.00	56.46	48.14	50.10	50.57	52.51
Grower								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	31.72	29.53	27.19	25.44	31.72	30.10	28.83	29.59
ALA[2]	6.92	7.09	7.12	7.18	6.92	7.00	6.93	6.72
DHA[3]	0.00	0.00	0.00	0.00	0.00	0.03	0.02	0.02
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.04	0.00	0.00
Omega 6[5]	31.80	29.60	27.27	25.51	31.80	30.17	28.91	29.67
Omega 3	6.95	7.15	7.16	7.20	6.95	7.12	7.00	6.82
Omega 6:Omega 3	4.57	4.14	3.81	3.54	4.57	4.24	4.13	4.35
SFA[5]	11.46	10.47	9.75	9.12	11.46	10.79	10.42	10.80
PUFA[6]	39.09	37.08	34.78	33.06	39.09	37.57	36.15	36.80
MUFA[7]	49.46	52.45	55.48	57.82	49.46	51.64	53.43	52.39
Finisher								
Meal	Canola meal				Juncea meal			
Levels (%)	0	10	20	30	0	10	20	30
Fatty acids (mg g^{-1})								
LA[1]	32.58	29.73	27.80	26.62	32.58	31.36	30.44	30.21
ALA[2]	6.45	6.59	6.72	6.78	6.45	6.50	6.40	6.26
DHA[3]	0.00	0.00	0.03	0.01	0.00	0.00	0.00	0.07
EPA[4]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.01
Omega 6[5]	32.66	29.80	27.88	26.99	32.66	31.46	30.51	30.29
Omega 3	6.45	6.59	6.78	6.83	6.45	6.50	6.45	6.41
Omega 6:Omega 3	5.06	4.52	4.11	3.91	5.06	4.84	4.73	4.72
SFA[5]	11.73	10.57	9.87	9.40	11.73	11.10	10.93	10.88
PUFA[6]	39.36	36.96	34.86	33.78	39.36	38.19	37.20	36.96
MUFA[7]	48.91	52.74	55.27	56.82	48.91	50.71	51.87	52.16

[1]LA, Linoleic acid,
[2]ALA, Alpha -Linolenic acid,
[3] DHA, docosahexaenoic acid,
[4] EPA, eicosapentaenoic acid,
[5] SFA, Saturated fatty acids,
[6] PUFA, Poly unsaturated fatty acids,
[7] MUFA, Mono unsaturated fatty acids
[8]Total fatty acids was calculated as SFA+PUFA+MUFA

Appendix H.2. Saturated fatty acids (% of total fat) in breast, thigh and wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Breast			
	Meal × Level		
Meal	Canola meal		Juncea meal
Levels			
0%	22.42±0.43*a*		22.53±0.40*a*
10%	20.86±0.40*a*		21.93±0.40*a*
20%	18.31±0.40*b*		21.08±0.40*a*
30%	16.39±0.40*c*		21.17±0.40*a*
Thigh			
Meal	Canola meal		Juncea meal
	23.01±0.37*b*		24.33±0.38*a*
ANOVA –*P* value			
	Breast	Wing	Thigh
Meal	0.263	<.001	0.015
Meal × Level	0.192	<.001	0.725

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

Appendix H.2. Poly unsaturated fatty acids (% of total fat) in wing ,thigh and breast meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Wing			
Level			
0%		20.93±0.39*b*	
10%		20.86±0.38*b*	
20%		23.28±0.38*a*	
30%		22.99±0.38*a*	
Meal	Canola meal		Juncea meal
	22.76±0.27*a*		21.27±0.27*b*
ANOVA –*P* value			
	Breast	Wing	Thigh
Level	0.246	<.001	0.181

[a-b] Means ± SEM with different postscripts for interaction means for meals within cut of meat are significantly different (α=0.05)

Appendix H.3. Mono unsaturated fatty acids (% of total fat) wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Wing			
Meal × Level			
Meal	Canola meal		Juncea meal
Level			
0%	56.47±0.45*b*		56.71±0.42*b*
10%	57.54±0.42*b*		57.95±0.42*ab*
20%	57.53±0.42*b*		56.52±0.42*b*
30%	59.42±0.42*a*		57.03±0.42*b*
Enzyme	NO		YES
	57.83±0.21*a*		56.96±0.21*b*
ANOVA –*P* value			
	Breast	Wing	Thigh
Enzyme	0.513	0.006	0.554
Meal × Level	0.168	0.006	0.939

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

Appendix H.4. Linoleic acid (% of total fat) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Wing			
Meal Levels	Levels		
0%	15.66±0.25*bc*		
10%	15.46±0.24*c*		
20%	16.55±0.24*ab*		
30%	16.61±0.24*a*		
Meal	Canola meal		Juncea meal
	16.61±0.17*a*		15.54±0.17*b*
ANOVA *P*-value			
	Breast	Wing	Thigh
Level	0.250	0.001	0.368
Meal	0.865	<.001	0.225

[a-b] Means ± SEM with different postscripts within the same columns are significantly different (α=0.05)

Appendix H.5. Alpha linolenic acid (% of total fat) in thigh and breast meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

Thigh		
Levels		
0%	2.73±0.11*b*	
10%	3.15±0.11*a*	
20%	3.05±0.11*ab*	
30%	3.00±0.11*ab*	
Meal	Canoal meal	Juncea meal
	3.13±0.07*a*	2.83±0.08*b*
Breast		
Level ×Enzyme		
Enzyme	NO	YES
Levels		
0%	2.55±0.19*a*	2.79±0.19*a*
10%	2.53±1.19*a*	3.16±0.19*a*
20%	2.50±0.19*a*	2.79±0.19*a*
30%	3.08±0.19*a*	2.48±0.19*a*
Enzyme	NO	YES
	2.75±0.05*b*	2.97±0.06*a*
Wing		
Meal ×Level		
Meal	Canola meal	Juncea meal
Levels		
0%	3.27±0.09*c*	3.15±0.08*c*
10%	3.71±0.08*b*	3.19±0.08*c*
20%	4.20±0.08*a*	3.42±0.08*bc*
30%	4.52±0.08*a*	3.46±0.08*bc*

ANOVA –*P* value			
	Breast	Wing	Thigh
Levels	0.708	<.001	0.044
Level ×Enzyme	0.019	0.185	0.795
Meal ×Level	0.309	<.001	0.925

[a-b] Means ± SEM with different postscripts for meals in thigh and breast and for levels in thigh are significantly different (α=0.05)

Appendix H.6. Eicosapentaenoic acid (% of total fat) in breast and wing meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Breast				
Meal	Canola meal		Juncea meal	
Enzyme	NO	YES	NO	YES
Levels				
0%	0.46±0.06*ab*	0.26±0.06*ab*	0.30±0.56*ab*	0.38±0.06*ab*
10%	0.25±0.06*ab*	0.29±0.06*ab*	0.50±0.06*a*	0.29±0.06*ab*
20%	0.27±0.06*ab*	0.36±0.06*ab*	0.32±0.06*ab*	0.21±0.06*b*
30%	0.30±0.06*ab*	0.39±0.06*ab*	0.31±0.06*ab*	0.40±0.06*ab*

Wing	
Level	
0%	0.09±0.00*b*
10%	0.09±0.00*b*
20%	0.10±0.00*ab*
30%	0.11±0.00*a*

Enzyme	NO	YES
	0.10±0.00*a*	0.09±0.00*b*

ANOVA –*P* value	Breast	Wing	Thigh
Level	0.388	0.004	0.186
Enzyme	0.541	0.035	0.216
Meal × Level × Enzyme	0.007	0.154	0.191

[a-c] Means ± SEM with different postscripts for interaction means within cut of meat are significantly different (α=0.05)

Appendix H.7. Omega 3 (% of total fat) in wing and thigh meat of broilers fed different levels of canola or juncea meal with and without Superzyme-OM™ (least square mean ±SE)

Wing		
Meal × Level		
Meal	Canola meal	Juncea meal
Levels		
0%	3.83±0.10*c*	3.73±0.09*c*
10%	4.29±0.09*b*	3.75±0.09*c*
20%	4.86±0.09*a*	4.04±0.09*bc*
30%	5.18±0.09*a*	4.11±0.09*bc*
Thigh		
Level		
0%	4.03±0.14*b*	
10%	4.52±0.14*ab*	
20%	4.34±0.14*ab*	
30%	4.59±0.14*a*	

ANOVA –*P* value			
	Breast	Wing	Thigh
Level	0.210	<.0001	0.030
Meal × Level	0.795	<.0001	0.551

[a-b] Means ± SEM with different postscripts within interaction means are significantly different (α=0.05)

Appendix H.8.Omega 6 (% of total fat) in wing meat of broilers fed different levels of canola or juncea meals with and without Superzyme-OM™ (least square mean ±SE)

	Wing		
Levels			
0%	16.76±0.28*ab*		
10%	16.49±0.27*b*		
20%	17.68±0.27*a*		
30%	17.78±0.27*a*		
Meal	Canola meal		Juncea meal
	17.69±0.19*a*		16.66±0.19*b*
ANOVA –*P* value			
	Breast	Wing	Thigh
Meal	0.992	0.000	0.591
Level	0.351	0.002	0.267

[a-b] Means ± SEM with different postscripts for interaction means within cut of meat are significantly different (α=0.05)

Appendix I. Product description of Superzuyme-OM™

Superzyme-OM™

Reg # 982528

Enzyme supplement for poultry feeds

DIRECTIONS FOR USE

Poultry: use 500 grams per tonne of complete feed

Conditioning and pelleting process may reduce detectable enzyme activity in feed.

GUARANTEED ANALYSIS

Crude Protein:	minimum 12%
Crude Fiber:	maximum 5%
Moisture:	maximum 11%
Cellulase:	minimum 2800 CMC units/g[1]
Mannanase:	minimum 400 MAN units/g[2]
Galactanase:	minimum 50 GAL units/g[3]
Xylanase:	minimum 1000 XYL units/g[4]
Glucanase:	minimum 600 GLU units/g[5]
Amylase:	minimum 2500 FAA units/g[6]
Protease:	minimum 200 HUT units/g[7]

[1] One CMC unit is the amount of enzyme which will produce 1 mg of reducing sugar (expressed as glucose) per hour at 37°C and pH 4.6.

[2] One MAN unit corresponds to the amount of enzyme which will produce 1 micromole of mannose reducing sugar equivalents per minute at 40°C and pH 4.0.

[3] One GAL unit corresponds to the amount of enzyme which will produce 1 micromole of galactose reducing sugar equivalents per minute at 40°C and pH 4.0.

[4] One XYL unit corresponds to the amount of enzyme which will produce 1 micromole of reducing sugar (expressed as xylose) per minute at 40°C and pH 4.5.

[5] One GLU unit is the amount of enzyme which will produce 1 mg of reducing sugar (expressed as maltose) per minute at 50°C and pH 5.0.

[6] One FAA unit corresponds to the amount of enzyme which breaks down 5.26mg of starch per hour at 40°C and pH 5.0

[7] One HUT unit corresponds to the amount of enzyme that produces in one minute, under specified conditions, a hydrolysate whose absorbance at 275nm is the same as that of a solution containing 1.1 microgram/ml of tyrosine in 0.006N HCl. Results are expressed as "hemoglobin units on a tyrosine basis".

INGREDIENTS

Wheat flour less than 1.5% fibre, Dried *Aspergillus oryzae* fermentation extract, dried *Aspergillus niger* fermentation extract, dried *Saccharomyces cerevisiae* fermentation solubles extract, dried *Trichoderma reesei* fermentation extract, enzyme source Reg # 981362, silicon dioxide SiO_2 , calcium carbonate $CaCO_3$, soya oil.

This product is free of antimicrobial activity and is not a source of viable microbial cells.This product may cause dermal and respiratory irritation and/or sensitivity. Appropriate protective equipment should be worn during handling

Manufactured and registered by

CANADIAN BIO-SYSTEMS INC.

CALGARY, ALBERTA

Lot #

Expiry Date: Six months from date of manufacture.

Storage: Stability is ensured for 6 months at room temperature and under appropriate storage conditions.

cbs CANADIAN BIO-SYSTEMS

25 Kg net weight