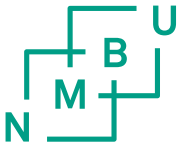
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Comparing rapeseed fed chicken with ordinary soybean oil fed chicken

Master Thesis (30 credits)

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In Feed Manufacturing Technology

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Ås 2016

Abstract

Energy is needed for the poultry in order to provide growth, egg production at a high level to allow maximal economic return for the production unit. In diet formulation, fat is added to increase palatability, increase fat soluble vitamin and to control dust in feed mill industry Soybean oil consists of high proportion of unsaturated fatty acid which is well utilized by poultry in order to meet energy requirement for fast growing broilers. The use of rapeseed oil is popular all over the world due to improvement made on it. A rapeseed line which contains the low amount of erucic acid and good balance between n-3 and n-6 fatty acids has been made. The aim of the present study was to compare fatty acid profile of breast muscle in chickens fed a diet containing rapeseed oil with ordinary soybean oil feed. In the present study, 36 chickens were included and 18 of the chicken received the diet containing rapeseed oil (diet 1) and 18 chickens received the diet containing soybean oil (diet 2).

Chickens fed with soybean oil feed had more saturated fatty acids like palmitic acid and stearic acid in muscle compared to chickens fed rapeseed oil feed. Oleic acid was significantly different between the two diets and it was less oleic acid in soybean oil than rapeseed oil. Comparing soybean oil with rapeseed oil, soybean oil contains more linoleic acid than rapeseed oil. Linoleic acid is converted to arachidonic acid in chicken through desaturation and elongation process. EPA, DPA and DHA, n-3 derivatives of α-linolenic acid are synthesized in chickens in the process similar to arachidonic acid by desaturation and elongation. That is why high amounts of fatty acids in the feed are now converted in same pattern in muscle. There is significant ($p < 0.05$) lower amount of arachidonic acid and higher amounts of the long chain fatty acids EPA, DPA and DHA in the chicken breast muscle of chicken fed diet containing rapeseed oil compared to soybean oil.

Our results show the final body weight was not different between the two diet groups. Consumption of meat from chickens fed the rapeseed oil containing feed can help us to increase the total intake of n-3 LCPUFA and reduce arachidonic acid without the consumer having to change their eating habits.

Acknowledgement

I would like to convey my sincere gratitude to my supervisor Prof. Anna Haug for her valuable guidance, scientific advice, patience, motivation, enthusiasm, and immense knowledge. Her constant guidance with esteemed comments and suggestions helped me during my thesis period. I am very grateful and deeply indebted to her. I feel I am proud to be her master student. I am also thankful to my co-supervisor Prof. Birger Svihus for his guidance during analysis of my statistics. Due to his wise advice, I was able to analyse the data. Besides this, special thanks to Department of Animal and Aquaculture Science at Norwegian University of Life Sciences (NMBU).

I am also grateful to my friend Biswas Bajgai for his continuous support, inspirations who shared the experience and help me during my study. Besides this I can never forget to my friend Parsuram Rimal, Deependra Acharya, Ganesh Kumar Upadhayayay, Dipendra K.C., Ishwor koirala, Krishna Kumar Thapa and Bhuwan Lal Meche. Lastly, my thanks to my father Shaligram Poudel, mother Tara Poudel and my dear wife Bidhy Lamsal Poudel for her encouragement during my study period in Norway.

Table of Contents

List of tables **Page**

List of figures **Page**

List of abbreviations

AA	Arachidonic acid
ALA	alpha-linolenic acid
Ca	Calcium
CHO	carbon, hydrogen, oxygen
DHA	Docosahexaenoic acid
DPA	Docosapentaenoic acid
EPA	Eicosapentaenoic acid
FAME	Fatty acid methyl esters
GC	Gas chromatography
HCl	Hydrochloric acid
Kcal	Kilocalories
Kg	kilogram
LA	Linoleic acid
LCPUFA	Long chain polyunsaturated fatty acids
MCP	Monocalcium Phosphate
ME	Metabolizable energy
MJ	Millijoules
mL	milliliters
N	Nitrogen
NSP	Non-starch Polysaccharides
P	Phosphorus
PUFA	Polyunsaturated Fatty Acid
Qn	Quantity
SBM	Soybean meal
SD	Standard deviation
T	Threonine
%	Percentage
α	alpha
°C	Degree celcius

1. Introduction

Chicken meat is regarded as a popular healthy type of meat. There is the variety of sources of fat and oil for the formulation of chicken feed. This includes rendering byproducts like tallow or poultry, vegetable oil like soybean, rapeseed, linseed, acid oil; hydrogenated fat, acid soapstock (McDonald et al. 2002). Chicken meat is one of the food items which are good carriers of long chain polyunsaturated fatty acids. The amount of fatty acid is affected by the type of fat in the chicken feed (Haug et al. 2007). During the last 50 years, poultry nutrition has remarkable change due to a dramatic change in the genetics of the bird (Svihus 2011). Furthermore, there is much more improvement in incubation and hatching, dietary formulations, health programs, management, and processing. This all improvement makes poultry one of the most economical produce (Dibner and Richards 2004).

Energy is needed for the poultry in order to provide growth as well as for the egg production at a high level to allow a maximal economic return for the production unit. In diet formulation, fat is added to increase palatability, increase fat soluble vitamin and to control dust in feed mill industry (Nutrition 1977). Broiler chickens have a limited ability to utilize fat because of an incomplete excretion of bile salts necessary for the digestion of fat (Svihus et al. 2000). They utilize polyunsaturated fat better than saturated fats and they get more energy easier from soybean oil feed when it was more polyunsaturated fats present. Soybean oil consists of a high proportion of unsaturated fatty acid which is well utilized by poultry in order to meet energy requirement for fast growing broilers.

Lipid contains minimum twice the available energy compared to carbohydrates and protein. This consequence in less heat loss and lower heat increment during feeding. Lipids are used to poultry diet in order to meet energy requirement for the fast growing broilers (Krogdahl 1985). The use of rapeseed oil is popular all over the world due to the improvement made on it. During the last forty years, rapeseed production was increased significantly due to low in erucic acid (22:1; cis-13 docosenoic acid) as well as glucosinolates (Przybylskiet al. 2005).

Furthermore, it has been reported that long chain n-3 Polyunsaturated Fatty Acid (PUFA) help in the metabolism in the body. Linoleic acid (C18:2, n-6) (linoleic acid) and alpha-linolenic acid (C18:3, n-3) can be converted in the cells to important eicosanoids. The western diet contains much more n-6 fatty acid compared to n-3 fatty acids. In the western diet, the ratio between n-6 to n-3 may be as high as 10:1 or even upto 20:1 (Haug et al. 2007).

Haug et al. (2012) reported that poultry meat can contain more long-chain n-3 fatty acids and less arachidonic acid than in the case at the present. Moreover, in order to improve the fatty acid profile of chicken meat containing less n-6 and more of n-3 can be obtained by removing soybean oil from the commercial feed, and add rapeseed and linseed oil instead.

Thus, feed should be formulated to supply the correct balance of fatty acid and of energy to allow optimum growth and performance. However, its optimum growth and performance depend upon feed intake. It is essential to make an economic decision for each company or enterprise. When formulating the feed, least-cost formulation is necessary for broilers. Also, the nutritional quality of broiler meat should be taken into account. The quality of the product is important while selecting ingredients for broiler diets, it is essential to look upon impact on gastrointestinal health and any physiological impact (Ross 2009).

The aim of the present study was to compare slaughter weight and fatty acid profile of chicken fed a diet containing rapeseed oil with ordinary soybean oil feed. Moreover, we are using the fatty acid profile to calculate fatty acid concentration in the breast meat.

2. Literature review

2.1 Digestive system of the chicken

In the chicken, the digestion of food begins in the break. Food picked up by the break enters the mouth. Break plays an important role in the crushing and cutting of feed. They are not able to chew their food because they do not have teeth. Chicken tongue allows to push the feedback of the mouth so that it can be swallowed. Saliva is mixed with the food.Thus,simply swallow and moves to the organ called crop and lead forward to proventriculus (Leeson and Summers 2001). In it, food is further mixed with number of enzymes to assist the breakdown of food. Afterwards, pass into the gizzard or grinding, mixing and mashing. The food passes through the duodenal loop and into the intestinal loop and into the small intestine. In small intestine absorption of food particles primarily occurs. The undigested particles pass through two pouches named ceca. In ceca water is absorbed from the food. The remaining undigested portions go towards colon and rectum to the cloaca in which they were excreted (Jacob et al. 2011).

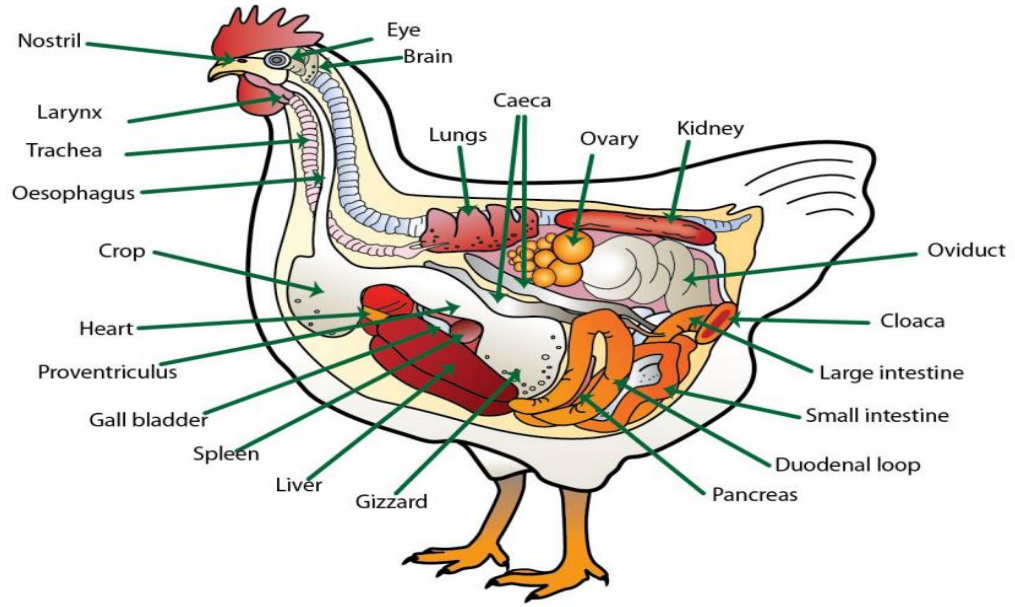


Figure 1: Internal organ of chicken (Jacob et al. 2011)

2.1.1 Fat digestion and absorption

The digestion process involves the breakdown of large and complex molecules into smaller ones that are eventually absorbed into the blood. Digestion occurs by means of physical and chemical aspects. The physical aspect involves grinding of food, i.e. stomach, gizzard, as well as a peristaltic movement which aid with passage through the intestinal tract. In the chicken, digestion of fats occurs mainly in the small intestine. Chemical digestion involves the secretion of enzymes that help to degrade food particles even further for molecular level absorption and transport (Leeson and Summers 2001).

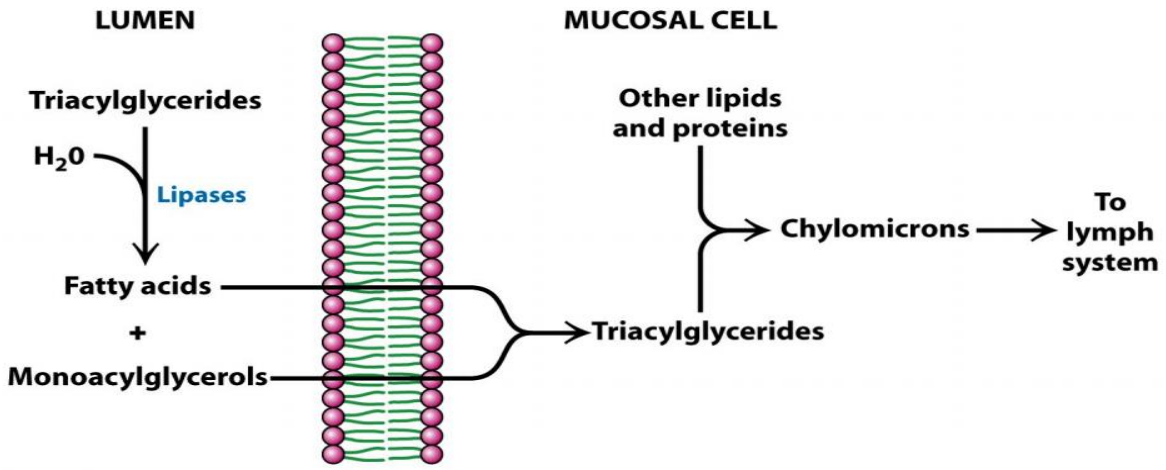


Figure 2: Fat digestion and absorption (Leeson and Summers 2001).

The digestion of fat starts after digesta enter duodenum. The fat in the duodenum stimulates cholecystokinin which further helps in pancreatic enzymes and bile secretion. From the gall bladder bile is released to emulsify fat in the chyme. The pancreas secretes pancreatic lipase that helps to break down emulsified fat into fatty acids and monoglycerides. Colipase that contains both hydrophobic and hydrophilic amino acids is essential in the action of lipase on triglyceride emulsion. The function of Colipase include i) maintaining the lipase in an active configuration at the lipid- water surface ii) it binds to the surface of lipid droplets iii) acts in anchor for lipase allowing lipase to digest triglycerides. As shown in the figure, due to the action of pancreatic lipase triglycerides are hydrolysed. Unsaturated fatty acids, medium chain fatty acids, monoglycerides and phospholipids spontaneously form mixed micelles with conjugated bile salts (Krogdahl 1985).

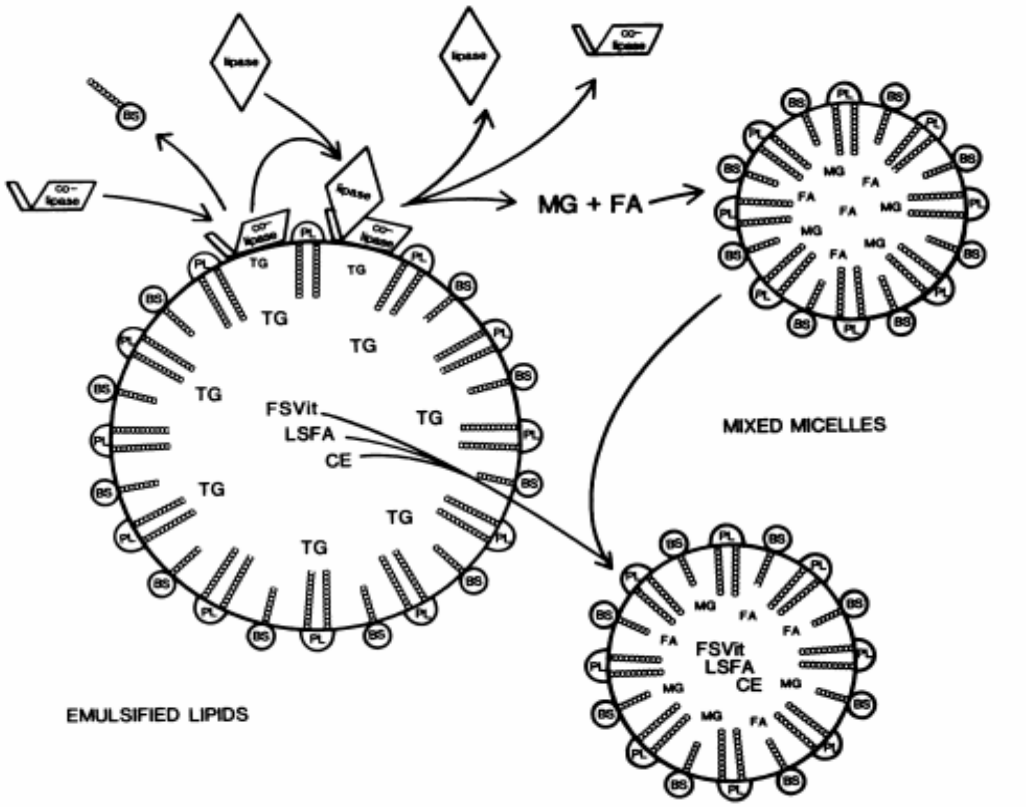


Figure 3: Sequence of events during intestinal lipolysis in poultry. (BS=bile salt, CE=cholesteryl ester, FA=free fatty acid, FSVit=fat soluble vitamin, LSFA=long chain saturated fatty acids, MG=monoglycerides, PL=phospholipid, TG=triglyceride) (Krogdahl 1985).

2.2 Role of fat in poultry diets

Fat is used as another name for lipid in the human food as well as in the ingredients for animal nutrition. Fat is generally insoluble in water but soluble in organic solvents. Fat chemical composition includes tri-esters of glycerol and fatty acids. Fatty acids those are not bound to other organic components as glycerol are known as free fatty acids (Baião and Lara 2005). The addition of fat to diets has following functions:

- Improve growth
- Supply energy
- Improves the absorption of fat-soluble vitamins
- Diminishes the pulverulence
- Increases the palatability of the rations
- Increases the effectiveness of the consumed energy
- Reduces the passage rate of the digesta in the gastrointestinal tract which allows a better absorption of all nutrients present in the diet.

2.2.1 Composition of fat

Fat are generally insoluble in water but soluble in organic solvents. Fat chemical composition includes tri-esters of glycerol and fatty acids. Fats and oils are main ingredients in feed formulation. In chicken feed, it may be added 2 to 10% fat.

Table 1: Common fatty acid nomenclature (Pond et al. 1995)

Chemical name	Trivial name	No. of carbon	No. of double bond	Short desiganation
Butanoic	Butyric	4	0	C4:0
Hexanoic	Caproic	6	0	C6:0
Octanoic	Caprylie	8	0	C8:0
Decanoic	Capric	10	0	C10:0
Dodecanoic	Lauric	12	0	C12:0
Tetradecanoic	Myristic	14	0	C14:0
Pentadecanoic	-	15	0	C15:0
Hexadecanoic	Palmitic	16	0	C16:0
Hexadecanoic	Palmitoleic	16	1	C16:1
Heptadecanoic	Margaric	17	0	C17:0
Octadecanoic	Stearic	18	0	C18:0
Octadecanoic	Oleic	18	1	C18:1
Octadecadienoic	Linoleic	18	2	C18:2
Octadecadienoic	Linolenic	18	3	C18:3
Eicosanoic	Arachidie	20	0	C20:0
Eicosatetraenoic	Arachidonic	20	4	C20:4
Docosenoic	Erucic	22	1	C22:1
Docosopentaenoic	Clupanodonic	22	5	C22:5
Tetracosanoic	Ligonoceric	24	0	C24:0

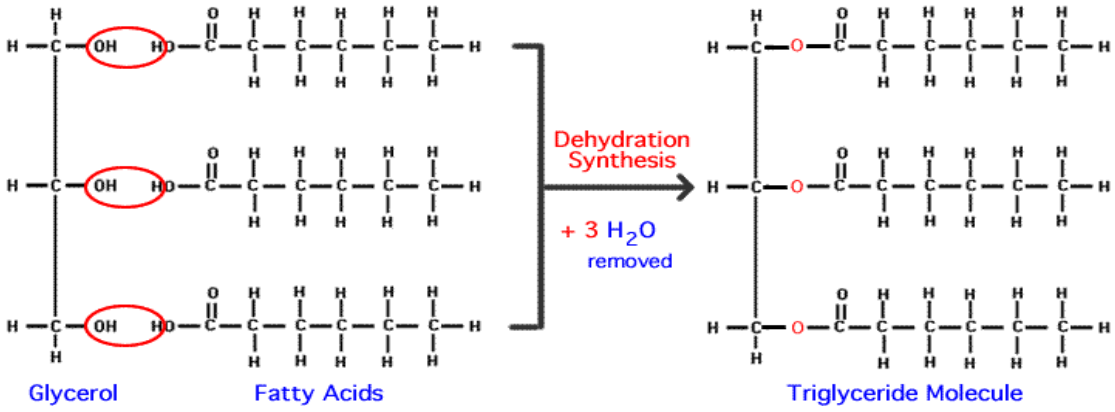
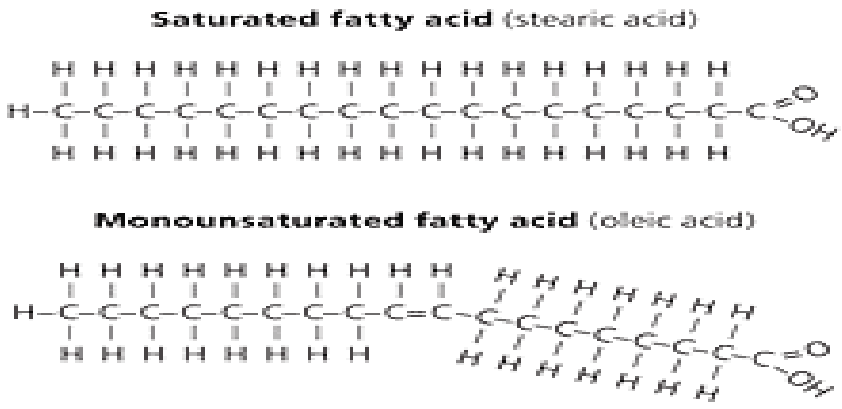


Figure 4: Triglyceride and its formation (Source: http://science.halleyhosting.com).

Pond et al. (1995) reported that lipids are classified in simple (fatty acids which are esterified with glycerol), compound (same as simple, however with other compounds attached as well), phospholipids (fats containing phosphoric acid and nitrogen), glycolipids (fatty acids compounded with CHO, but no N), derived lipids (substances from the above derived by hydrolysis) and sterols (large molecular wt. alcohols found in nature and combined with fatty acids e.g., cholesterol).The fatty acid always contains even number of carbon atoms. It contains from four to thirty two carbon (C) atom bounded in a chain. If the bond between carbon atoms is single, this fatty acid is known as saturated fatty acid. If the bond is double, this fatty acid is known as unsaturated fatty acid. If there is only one double bond, it usually is between the 9th and 10th carbon atom in the chain. When there is a second double bond, it usually occurs between the 12th and 13th carbon atoms and in the case of a third is usually between the 15^{th} and 16^{th} (Tisch 2005). The saturated fatty acids are butyric acid ($C_4H_8O_2$), Palmitic acid ($C_{16}H_{22}O_2$) and stearic acid ($C_{18}H_{36}O_2$). Unsaturated fatty acids are oleic acid ($C_{18}H_{34}O_2$), linoleic acid ($C_{18}H_{32}O_2$) and alpha linolenic acid ($C_{18}H_{30}O_2$). There are, Alpha-linolenic acid (polyunsaturated n-3 fatty acids) and linoleic acid (polyunsaturated n-6 fatty acids), are the two essential fatty acids. Thus, they cannot be produced by the body and therefore, they have to be supplied by in diets (Swanson et al. 2012).



Polyunsaturated fatty acid (α linolenic acid)

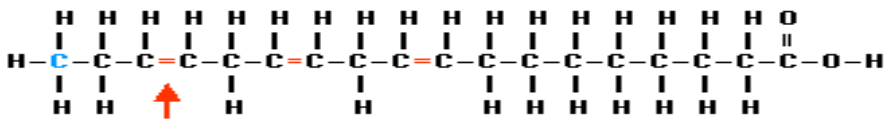


Figure 5: Saturated fatty acid stearic acid (C18:0) and unsaturated fatty acids (C18:1c9) and polyunsaturated fatty acid linoleic acid http://www.aafp.org) and polyunsaturated fatty acid α linolenic acid C18:3 (http://users.rcn.com/jkimball.ma.ultranet/BiologyPages/F/Fats.html).

Fats and oils are originated from the plant and animal. Plant fats are rich in unsaturated fatty acids while the fats from the animal origin are rich in saturated fatty acids. Fats and oils are produced from three different sources- animal (pigs, cows and sheep), vegetables (wheat, barley, oats, seeds, olives, beans) and fish (trout, mackerel, salmon, herring).

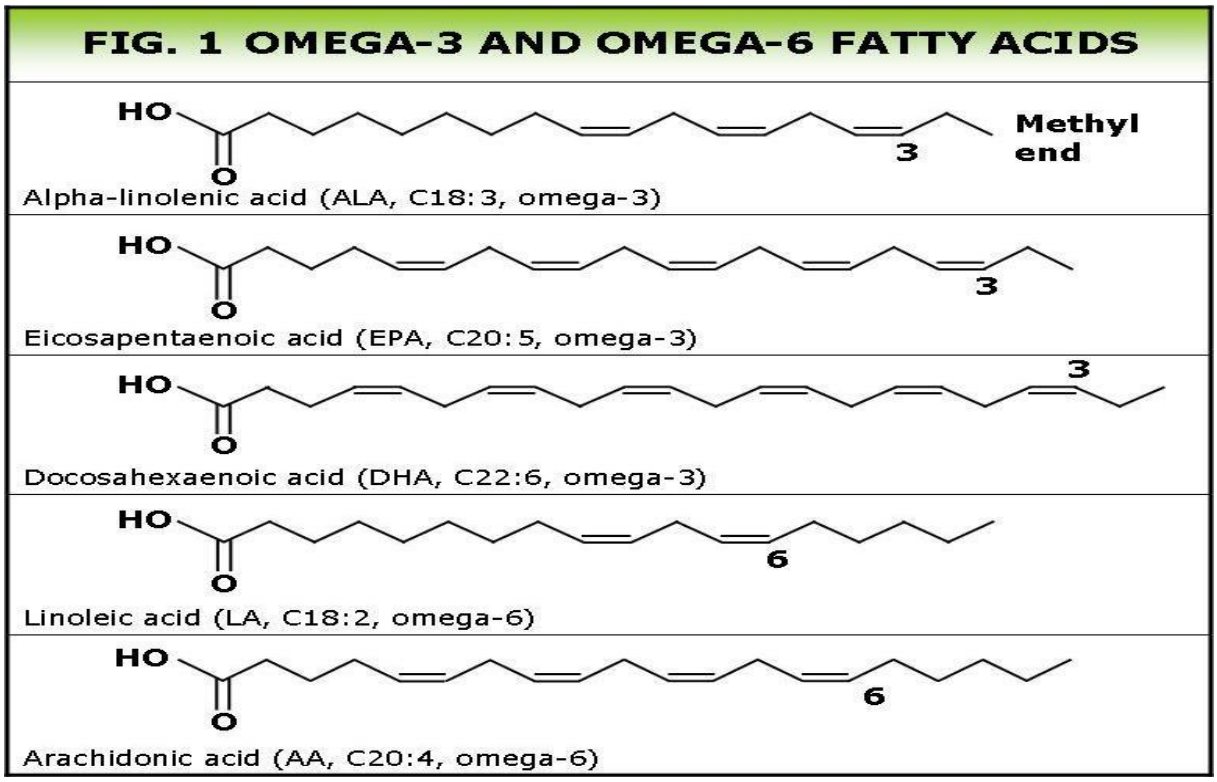


Figure 6: Structure of the n-3 and n-6 fatty acid(Source: http://www.eufic.org).

2.3 Essential Fatty Acids

Essential fatty acids are PUFA which contain the first double bond on their third (n-3 series) or on the sixth (n-6 series) carbon atom from the methyl group. Essential fatty acids are not synthesised by mammals. Therefore, chicken is completely dependent on dietary intake. They are necessary for the development of cell membranes, the development of nerve tissue and they also form the precursors of major endocrine signaling (Glaser et al. 2010).

The essential fatty acids (n-6 fatty acid linoleic acid and n-3 fatty acid α-linolenic acid) can extend, desaturated or change to give extra-long n-6 and n-3 fatty acids. Enzymes, elongase and desaturase (Δ5 and Δ6) are used during this transformation. This is the same for both acids and it will thus be a competition between the fatty acids (Preedy et al. 2011).

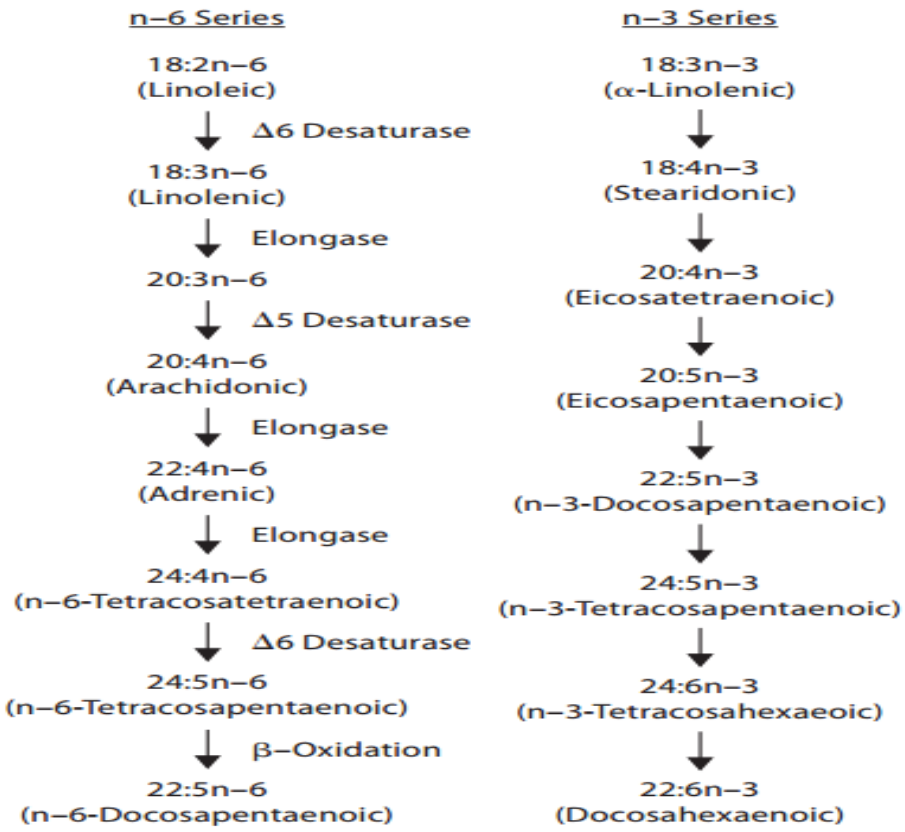


Figure 7: Schematic pathway of n–6 and n–3 fatty acid metabolism (Ratnayake and Galli 2009).

As shown in the figure 7 the LA is converted to AA, while ALA is converted to EPA, DPA and DHA. When we look the enzyme Δ 6 desaturase included in the desaturation of both LA and ALA. This enzyme has a greater affinity for ALA than the LA. However during the comparison the concentration of LA is high and it will further lead to a greater conversion of LA to n-6 LCPUFA than ALA to n-3 LCPUFA. The both pathways (i.e. n-6 and n-3 series) are independent of each other and there are no cross reactions and both pathways use the same desaturation and the elongation enzymes (Ratnayake and Galli 2009).

The first step involves insertion of a double bond at the Δ 6 positions of LA and ALA by the action of Δ 6 desaturase. This is followed by chain elongation by 2 carbon units by elongase and an insertion of another double bond at the Δ 5 position by Δ 5 desaturase to form AA (20:4n-6) and EPA (20:5n-3), respectively. In the next step, AA and EPA are elongated by 2 carbon units to 22:4n-6 and 22:5n-3, respectively. These C24 PUFAs are then desaturated by the Δ 6 desaturase to give 24:5n-6 and the 24:6n-3. Same desaturase enzymes are used that desaturates LA and ALA. DHA is formed from the 24:6n-3 by chain shortening by 2 carbon

units by means of one cycle of the beta-oxidation pathway. By the similar chain shortening system, 22:5n-6 is produced from 24:5n-6 (Ratnayake and Galli 2009).

2.4 Types of oil used

2.4.1Soybean oil

Liu (2012) reported that Soybean (*Glycine max* L.) is one of the most important field crops. Soybean contains a major source of edible oil and protein for human, livestock and other applications. Soybean is regarded as a high protein yielding crop per unit area than any other protein crops. So, the soybean crop plays an important role in the world to reduce the hunger. The other importance of soybean is preventing and treating chronic disease due to the presence of isoflavones (Nawar 1996). Soybean seeds contain about 40% protein, 20% oil, and 35% carbohydrate. Due to soybean composition, it can be used in multiple ways in the industry. They are crushed and dehulled to extract oil.

At the time of seed development, the lipid is stored in the form of triglycerides. In the soybean, most of the fatty acids are found to be unsaturated. It is found that highest percentage of fatty acid in linoleic acid followed in decreasing order by oleic, palmitic, linolenic and stearic acid respectively. However, it also contains minor fatty acids, including arachidic, behenic, palmitoleic and myristic acid (Nawar 1996). Major components of crude oil are triglycerides (96%), phospholipids (2%), unsaponifiables materials (1.6%), free fatty acids (0.5%), minute amounts of carotenoid pigments, and trace metals. The unsaponifiable material consists of tocopherols, phytosterols, and hydrocarbons. The concentration of these minor compounds is reduced after typical oil processing (Berk 1992). The crude oil needs further treatment in order to convert bland, stable as well as to provide nutrition.

Table 2: Fatty acid composition in soybean oil (Erickson et al. 1980).

Composition	Fatty acid composition, weight %	
	Range	Average
Saturated		
Lauric C12:0	-	0.1
Myristic C14:0	<0.5	0.2
Palmitic C16:0	7-12	10.7
Stearic C18:0	2-5.5	3.9
Arachidic C20:0	<1.0	0.2
Behenic C22:0	<0.5	-
Total	10-19	15
Unsaturated		
Palmitoleic C16:1	<0.5	0.3
Oleic C18:1	20-50	22.8
Linoleic C18:2 n-6	35-60	50.8
Alpha Linoleic C18:3 n-3	2-13	6.8
Eicosenoic C22:1	<1	-
Total	-	80.7

2.4.2 Rapeseed oil

Proskina et al. (2011) reported that rapeseed is an important source of vegetable oil after soybean. Rapeseed is widely used in Europe due to lower relative cost and better adaptability than other oil crops. It is characterized by high nutritional value containing about 35.0 to 45.0% crude protein, 14.0-15.0% of crude fat, 1,736 kcal kg^{-1} of metabolic energy, 8.0-10.0% of n-3 and 20.0-26.0% of n-6 fatty acids. The edible fat and oil are composed of triacylglycerols.

Table 3: Fatty acid composition in weight % of Rapeseed oil (Gunstone 2007).

	C16:0	C18:0	C18:1	C18:2	C18:3	C20:1
Low-erucic acid rapeseed	4	2	56	26	10	2

Rapeseed oil is removed by crushing, solvent extraction or by the combination of both techniques. Its quality depends upon the method of oil removal. Low or zero varieties of rapeseed has been developed to minimize both erucic acid and glucosinolates. Rapeseed has expanded its acceptances all over the world due to its major improvements in the seed oil and the meal quality. In order to increase nutritional and functional properties, rapeseeds with different type with a modified fatty acid composition are available for different purposes. It has been reported that in the past forty years it has seen significant growth of rapeseed production is observed due to the introduction of food rapeseed (canola), low in erucic acid (22:1; cis-13 docosenoic acid) and glucosinolates (Smulikowska et al. 1998).

Leonard (1994) reported that plant breeding is the tool to make rapeseed oil more competitive in various segments of the food and industrial oil markets. Firstly, the maximum content of the desired fatty acid will decrease the amount of the waste. Secondly, significant savings in the processing costs. Rapeseed oil with high erucic acid with alarger than 80% 22:1 level is desired to decrease the cost of producing this fatty acid and its derivatives as a renewable and environment- friendly industrial feedstock. Due to the effort of plant breeder, modifications of fatty acids composition are possible in order to improve oxidative stability or crystallization properties. There is the development in low linolenic acid canola oil (2% vs 9%), high oleic acid canola oil (69-77% vs 60%), high palmitic acid canola oil (10% vs 4 %), high steric acid canola oil (30% vs 2%),lauric acid canola oil (33%), y- linoleic acid canola oil (37% vs 1%) (Gunstone 2011).

2.5 Poultry diet

There are large numbers of classes of feedstuffs available for the use of poultry (Choct and Hughes 1997). The nutrient composition is presented in the table 4 below. The poultry diet is formulated based on the Metabolizable Energy (ME) which represents the energy. In Norway poultry diet is based on cereal grain and soybean meal along with small amounts of fat, calcium, phosphorus, salt, vitamins and trace minerals.

2.5.1 Cereals

2.5.1.1 Barley

Barley ranks fourth in the world production of cereal crops. Barley is commonly used in the poultry diets. It is being popular in some regions of Canada and Europe. Barley is grown on both irrigated and dry land. It is considered as medium-energy grain. It has low starch content and high fiber content. Barley also contains gluten and possesses glycemic properties (Salih et al. 1991). The nutrient composition is presented in the table 4.

2.5.1.2 Oats

Oats is a versatile crop and can be grown in the different growing system. Oats crop should be stored carefully and it needs to be dried to a maximum of 14% moisture. Oats have high fiber content. Likewise, they contain more oil than other cereals. The oil is rich in unsaturated fatty acids, including the essential fatty acid like linoleic acid. It has been reported that an increased starch digestibility has been observed when oat hulls are added to the broiler diets. In addition, of oat hulls does not reduce nutrient digestibility. Moreover, increased gut volume and a faster feed passage may be the significant view for an increased feed consumption in diets containing oat hulls (Hetland and Svihus 2001).

2.5.1.3 Wheat

Wheat is one of the important cereal grain grown for human consumption. However the sizable amount is used in poultry feed industry and it should be limited to 50%. High gluten content improves the pellet quality in poultry diets and non-starch polysaccharide (NSP) content may limit inclusion rate in unless using enzymes. In the feed mill, grinding is avoided and a fine meal was made. This is done because of dust hazard problem at the time of processing. It is essential to avoid over processing because over processing reduces the palatability in wheat. Wheat has a tendency to flour and forms the small fine particle (Chris 2008).

Table 4: Metabolisable energy (ME) and nutrient composition in wheat, barley and oat.

(http://www.poultryhub.org/nutrition/feed-ingredients/)

Ingredients	Protein (%)	ME (kcal/kg)	Ca (%)	Available P (%)	Lysine (%)
Wheat	13	3153	0.05	0.2	0.5
Barley	11.5	2795	0.1	0.2	0.4
Oats	12	2756	0.1	0.2	0.4

2.5.2 Animal protein

2.5.2.1 Fish meal

Fish meal is prepared from dried and ground fish and fish by-products. It represents a well-balanced as well as highly digestible protein with 60% protein. Excess amount of fish meal will result fishy flavor in the egg and meat. Likewise feeds with high amount of fish meal will harm the environment due to excess amounts of phosphorus (Rumsey 1993).

2.5.3 Plant protein

2.5.3.1 Soybean meal

Soybean meal is regarded as the most commonly used plant protein source containing about 48 % protein. It is a very palatable supplement, highly digestible and contains well-balanced amino acids. It is usually the most economical protein source for animal diets. It contains various anti-nutritive factors which affect protein and energy utilization in diets for poultry (Perilla et al. 1997). Protease inhibitors and lectins are heat labile anti-nutritive factors can be inactivated by toasting or extruding. Soybean meal over processing leads to reduce the amount of lysine and cystine digestibility and subsequently reduce growth performance. Thus, by means of adding microbial phytase to diets increase apparent digestibility of protein and amino acids (Kocher et al. 2002).

2.5.4 Minerals

Minerals are the essential inorganic compounds, which is required in small amounts for growth, maintenance, reproduction and lactation (McDowell 2003). Calcium is one of the most important factors which influence the quality of eggs and bones. Due to which calcium

must be added to the diets. They are essential in blood coagulation, nerve and muscle function. Their deficiency cause retarded growth, deformed bones in young chicken and soft-shelled eggs (Leeson et al. 2005).

2.5.5 Vitamins

Vitamins are essential organic nutrients which are required in small amounts and it cannot be synthesized by the body. They are generally classified into fat soluble-vitamins (A, D, E, and K) and water-soluble vitamins (B-complex and vitamin C). Vitamin D for poultry is provided in the form of vitamin D_3. It is found naturally in the fish liver oil. It may be synthesized by the irradiation of an animal sterol (Joint and Organization 2005).

Table 5: The function of vitamin A, vitamin D, vitamin E and vitamin K (Joint and Organization 2005).

Vitamin	function	deficiency	sources
A	Development on healthy skin and nerve tissue and bone formation	retarded growth at the young stage and abnormal condition around the eyes are seen	carotene, legume forages, animal body oils
D	Accurate utilization of calcium and phosphorus to produce healthy bones.	Retarded growth, osteoporosis, misshapen bones and lameness.	forage crops, fish liver oils, irradiated yeast
E	Normal reproduction	Poor growth and muscular dystrophy. They are rapidly destroyed in rancid or spoiled fats. Due to which they cause white muscle disease.	cereal grains and wheat germ oil, green forages and oil seeds
K	Maintenance of normal blood coagulation	Serious hemorrhages can cause light wounds or bruises.	green leafy forages, fish meal, liver,

2.5.6 Enzyme

Phytase is widely accepted in practice for utilization of phosphorus and other nutrients bound in phytate complex in diets for poultry. For the successful use of phytase, enzyme diet should contain sufficient amount of phytate phosphorus. It has been reported that about 50% of the phytine phosphorus from feeds can be released by the use of phytase. Application of phytase results positive effect on the digestibility and the availability of calcium, iron, magnesium, zinc and protein with the use of this enzyme (Lukić et al. 2009).

3 Material and method

3.1 Chicken experiment

3.1.1 Birds and housing

In the present study, 36 chickens, Ross 308, (Samvirkekylling Norway) were included in order to study the effect of two different types of feed on breast muscle fatty acid composition and final body weight. The 36 broiler chickens were randomly selected and kept in room number 3 at center for husdyrforsøk, Ås Norway. Total number of cage used was 12. In each cage 3 birds were kept. In cage 1, 2, 3, 7, 8 and 9, diet 1 were fed to the chicken whereas in cage 4, 5, 6, 10, 11 and 12, diet 2 (Kromat, Felleskjøpet) were fed to the chicken. The feeding regime from 7 to 14 days of age consisted of feed being available for ad libitum consumption from 08.00 to 09.00, 12.00 to 13.00, 16.30 to 17.30 and 21.00 until light goes o at 23.00 hour. There was darkness from 23.00 to 03.00 and from 04.00 to 08.00. From 14 days of age until termination of the experiment at 35 days of age. Feed was available for ad libitum consumption from 08.00 to 09.00, 12.30 to 13.30, 17.30 to 18.30 and 22.00 to 23.00. At day 35, the birds were weighed and slaughtered. Breast muscle was taken out and frozen at -20°C for fatty acid determination.

Diet 1: Barley/Oats/ Wheat-based experimental diet with rapeseed oil

Diet 2: Commercial diet containing soybean oil (Kromat)

3.2 Diet formulation, diet 1 (with rapeseed oil)

Table 6: Diet 1

ingredients	QN	ME (MJ/kg)
Demand	1	13.2
Supply	1	4.745
Diet	g/kg	
Barley	200	12.4
Oats	200	
Wheat	260	
Fish meal	60	14.6
Rapeseed oil	30	35
SBM	212	13.3
Limestone	10	0
MCP	10	0
L-lysin HCL	2	0
DL -Metionin	2	0
L- Threonin	1	0
Salt	2.5	0
Mineral premix	1.5	0
Vitamin A	0.5	0
Vitamin ADKB	1	0
Vitamin D_3	0.8	0
Vitamin E	0.5	0
Chlorinechloride	1.2	0
Phytase/ glucanase/ xylanase		
Titanium oxide	5	0
	1000	

3.3 Diet 2 (Kromat)

Table 7. Diet 2

Ingredients	%
Crude protein	21.2
Fiber	2.2
Crude fat	4.8
Ash	5.1
Calcium	0.76
Phosphorus	0.58
Sodium	0.14
Lysine	1.19
Methionine	0.53
Selenium	0.40 mg / kg

The ingredient used were wheat, soybean extracted, Maize grits, Corn Gluten, Oats husked, Vegetable fat, fish meal, oats, ensiled protein, ground limestone, Mono Calcium Phosphate, Aminopremix ,Vitamin premix, Sodium bicarbonate, Taste Spread Mix. The vitamin used were E672 Vitamin A 9000 ie, E671 Vitamin D3 5000 IU Vitamin E 80 mg. In addition, micro mineral used were E1 Iron as iron (II) fumarate 52 mg, E2 Iodine as calcium iodate 1.0 mg, E4 copper as copper (II) sulfate 15 mg, E5 Manganese as manganese sulphate 127 mg, E6 Zinc as zinc sulfate 82 mg, E8 Selenium as sodium selenite 0.32 mg. The enzyme used were 4a1640 6-phytase EC 3.1.3.26 500 FTU, E1641 Endo-1,4 -betaxylanase EC 3.2.1.8 70 AXC, E1634 Endo-1,3 (4) -betaglukanase EC 3.2.1.6 100 AGL.

3.4 Fatty acid composition of feed

Table 8 shows the fatty acid composition of diet 1 and diet 2. The fatty acids C12:0, C14:0, C15:0, C16:1 c9; C18:1 c9, C18:1 c11, C20:0, C20:1 n-9, C20:2 n-6, C20:3 n-3, C20:4 (n-6/C22:1, n-9), C20:4 n-3, C20:5n-3 ,C22:5n-3and C22:6, n-3 were higher in diet 1 than diet 2. Whereas remaining fatty acids C16:0, C17:0, C18:0, C18:2, n-6, C18:3, n-3, C22:0 were higher in diet 2 than diet 1.

Table 8: Fatty acid composition of diet 1 and diet 2.

Common name	fatty acid composition,% FAME of feed	Diet 1 (rape seed)	Diet 2 (kromat)
Lauric acid	C12:0	0.02	0.01
Mysteric acid	C14:0	0.82	0.21
Pentadecylic acid	C15:0	0.08	0.05
Palmitic acid	C16:0	11.28	13.43
Palmitoleic acid	C16:1,c9	0.62	0.24
Margaric acid	C17:0	0.08	0.10
Stearic acid	C18:0	1.46	2.17
Oleic acid	C18:1, c9	39.97	24.75
Vaccenic acid	C18:1, c11	2.23	1.65
Linoleic acid	C18:2, n-6	27.29	47.61
Arachidic acid	C20:0	0.32	0.27
Alpha-linolenic acid	C18:3, n-3	5.97	6.08
Eicosenoic acid	C20:1, n-9	1.67	0.50
Eicosadienoic acid	C20:2, n-6	0.09	0.06
Behenic acid	C22:0	0.20	0.23
Eicosatrienoic acid	C20:3, n-3	1.34	0.17
AA+Erucic acid	C20:4,n-6+C22:1,n-9	0.27	0.04
Eicosatetraenoic acid (ETA)	C20:4, n-3	0.06	0.04
EPA	C20:5, n-3	0.69	0.13
Docosapentaenoic acid (DPA)	C22:5, n-3	0.11	0.02
DHA	C22:6, n-3	1.21	0.27
LA/ALA		4.57	7.86
n-6/n-3		3.46	7.36
AA/EPA		0.39	0.29
C16:1c9/C16:0		0.06	0.02
C18:1c9/C18:0		27.32	11.4

3.5 Amount of fatty acid in feed (mg/ 100 g feed)

Amount of fatty acid in feed (mg/ 100 g feed) was calculated by using C13:0 internal standards, table 9. The sum of fatty acid shows that it was 5.1 g fatty acid in 100 g feed of diet 1 and 4.7 g fatty acid in 100 g feed of diet 2. The fatty acids C12:0, C14:0, C15:0, C16:1 c9,C18:1, c9, C18:1, c11, C20:0, C20:1 n-9, C20:2 n-6, C20:3 n-3,C20:4 (n-6/C22:1, n-9), C20:4 n-3, C20:5n-3, C22:5 n-3 and C22:6 n-3 were higher in diet 1 than diet 2 and remaining fatty acids were lower in diet 1 than diet 2.

Table 9: Amount of fatty acid in feed (mg/ 100 g feed).

	Diet 1	Diet 2
C12:0	1.1	0.6
C14:0	43.7	10.4
C15:0	4.0	2.2
C16:0	602.3	648.0
C16:1,c9	33.3	11.5
C17:0	4.2	4.6
C18:0	78.2	104.6
C18:1, c9	2134.6	1193.3
C18:1, c11	119.4	79.6
C18:2, n-6	1457.4	2295.4
C20:0	17.2	12.8
C18:3, n-3	318.7	292.2
C20:1, n-9	89,1	24.3
C20:2, n-6	4.8	2.9
C20:3, n-3	71.6	8.4
C20:4, n-6 + C22:1, n-9	14.2	1.8
C20:4, n-3	3.2	1.8
C20:5, n-3	36.6	6.2
C22:5, n-3	5.8	0.7
C22:6, n-3	64.6	13.2
Sum	5115	4726

LA/ALA	4.6	7.9
n-6/n-3	3.0	7.1
AA/EPA	0.4	0.3
sum n-6	1477	2300.1
sum n-3	501	323

3.6 Procedure during analysis

A direct method for fatty acid methyl ester (FAME) was done during analysis (O'Fallon et al. 2007). The steps during analysis were listed below.

- Sample 1 g muscle was were weight and the sample was placed into a 16×125mm screw cap Pyrex culture tube to which 1 mL of the C:13 internal standards (0.5 mg of C13:0/mL of MeOH) and it was followed by putting 0.7 mL of 10 N Potassium hydroxide (KOH) in water
- Afterwards 5.3 mL of Methyl hydroxide (MeOH) were added
- The tube was incubated in a 55°C water bath for one and half hours with vigorous hand shaking for five sec every twenty minutes
- It was cooled below the room temperature in a cold tap water bath and 0.58 mL of 24 N Sulfuric acid (H_2SO_4)was added
- It was precipitated with Potassium sulfate (K_2SO_4) and incubated again in a 55°C water bath for one and half hours. Properly hand shaking for five seconds every minute was done.
- After FAME synthesis, the tube was cooled in a cold tap water bath
- Three milliliters of hexane was added and the tube was vortex-mixed for five minutes on a multi tube vortex.
- The tube was centrifuge for five minutes in a table top centrifuge, and the hexane layer, the containing the FAME was placed into a GC vial.
- The Vial was capped and placed at -20°C until GC analysis.
- Afterwards the fatty acid composition of the FAME was determined by capillary GC gas chromatography.

4 Result

4.1 Final body weight

Final body weight at day 35 was not different between the two diet groups. The final body weight was about 2.63 kg for diet 1 and 2.66 kg for diet 2 as shown in table 10.

Table 10: Final body weight

	Diet 1		Diet 2		T-test
	Mean	SD	Mean	SD	
Final body weight, g	2632	146	2664	127	0.691

4.2 Fatty acid profile of chicken breast muscle, % FAME

% FAME of chicken breast muscle was shown in table 11. The table shows the mean results, SD and t-test of breast muscle from chicken fed diet 1 and diet 2.

Table 11: Fatty acid profile of chicken breast muscle, % FAME

	Diet 1		Diet 2		
% FAME	Mean	SD	Mean	SD	T-test
C12:0	0.02	0.00	0.02	0.00	0.000
C14:0	0.61	0.02	0.39	0.06	0.000
C15:0	0.10	0.00	0.08	0.00	0.000
C16:0	18.50	0.37	20.24	0.89	0.001
C16:1,c9	2.85	0.18	3.22	0.73	0.244
C17:0	0.12	0.01	0.12	0.01	0.858
C18:0	6.35	0.51	7.09	1.19	0.191
C18:1, c9	36.77	1.17	27.99	2.34	0.000
C18:1, c11	2.99	0.06	2.66	0.26	0.013
C18:2, n-6	14.81	0.34	22.72	1.13	0.000
C20:0	0.07	0.00	0.05	0.00	0.000
C18:3, n-6	0.10	0.01	0.16	0.02	0.000

C18:3, n-3	2.82	0.12	2.24	0.35	0.003
C20:1, n-9	0.82	0.04	0.38	0.01	0.000
C20:2, n-6	0.36	0.04	0.57	0.15	0.009
C20:3, n-6	0.48	0.06	0.60	0.17	0.121
C20:3, n-3	0.29	0.02	0.13	0.03	0.000
C20:4, n-6+C22:1, n-9	1.67	0.20	2.95	1.08	0.018
C20:4, n-3	0.19	0.02	0.17	0.06	0.540
C20:5, n-3	0.72	0.07	0.32	0.12	0.000
C22:5, n-3	1.39	0.13	0.95	0.38	0.024
C22:6, n-3	2.29	0.21	1.01	0.43	0.000
Unknown	5.68	0.37	5.93	1.26	0.657
LA/ALA	5.33	0.21	10.50	1.50	0.000
n-6/n-3*	2.28	0.06	5.69	0.64	0.000
AA/EPA	2.28	0.16	9.06	0.72	0.000
16:1c9/16:0	0.15	0.01	0.16	0.03	0.733
18:1c9/18:0	5.97	0.60	4.15	0.92	0.002

*n6/n3 = (18:2n-6 +18:3 n-5 +20:2 n-620:3 n-6+ 20:4 n-6)/(18:3 n-3 + 20:3 n-3+20:4 n-3 + 20:5 n-3+22:5 n-3 + 22:6 n-3).

4.3 Overall showing fatty acid profile, % FAME, of chicken breast muscle

The fatty acid composition of chicken breast muscle (% FAME) is shown in table 12. The fatty acid composition is shown as the mean composition of 3 birds in each cage. In diet 1 number of cages was 6, and in each cage there were 3 birds. Thus total number of birds was 18 in diet 1. Likewise in diet 2, number of cages was 6, and in each cage there were 3 birds. Thus, equal number of birds in diet 1 and diet 2 that was 18. We calculate mean, SD from diet 1 and diet 2 and t-test were used to compare fatty acid profile of breast muscle in birds fed diet1 and diet2.

It is found that C12:0, C14:0, C15:0, C16:0, C18:1, c9, , C18:1, c11, , C18:2, n-6 , C20:0, C18:3, n-6 , C18:3, n-3, C20:1, n-9 , C20:2, n-6 , C20:3, n-3 , C20:4, n-6/C22:1, n-9 , C20:5, n-3, C22:5, n-3 , C22:6, n-3 , LA/ALA, n-6/n-3, AA/EPA, 18:1c9/18:0 were significant different between two diet groups. However, it was found that C16:1, c9, C17:0, C18:0,

C20:3, n-6, C20:4, n-3, 16:1c9/16:0 were not significantly different. In addition, fatty acids composition in chicken breast muscle in different cages within the same diet group was very similar as shown in table 12.

Table 12: Detailed showing fatty acid profile, % FAME, of chicken breast muscle

% FAME	Diet 1 (rapeseed oil feed group)								Diet 2 (soybean oil feed group)								
cage	1	2	3	7	8	9	Mean	SD	4	5	6	10	11	12	Mean	SD	T - test
C12:0	0.02	0.02	0.02	0.02	0.02	0.02	0.02	0.00	0.02	0.02	0.01	0.01	0.02	0.02	0.02	0.00	0.00
C14:0	0.62	0.58	0.62	0.62	0.58	0.62	0.61	0.02	0.41	0.47	0.33	0.33	0.40	0.42	0.39	0.06	0.00
C15:0	0.10	0.09	0.09	0.09	0.10	0.10	0.10	0.00	0.08	0.09	0.08	0.07	0.08	0.08	0.08	0.00	0.00
C16:0	18.69	18.43	18.42	18.68	17.84	18.93	18.50	0.37	20.64	21.18	18.94	19.31	20.62	20.75	20.24	0.89	0.00
C16:1.c9	2.89	2.87	2.97	2.93	2.49	2.92	2.85	0.18	3.95	3.63	2.14	2.48	3.56	3.57	3.22	0.73	0.24
C17:0	0.11	0.11	0.12	0.11	0.13	0.12	0.12	0.01	0.11	0.12	0.13	0.11	0.11	0.11	0.12	0.01	0.86
C18:0	6.79	6.75	5.50	6.03	6.70	6.35	6.35	0.51	6.11	6.03	8.67	8.51	6.73	6.52	7.09	1.19	0.19
C18:1, c9	35.54	36.13	38.52	37.87	35.98	36.61	36.77	1.17	29.33	29.12	25.09	24.94	29.23	30.25	27.99	2.34	0.00
C18:1, c11	3.04	3.01	2.93	2.95	3.06	2.93	2.99	0.06	2.51	2.38	2.92	3.04	2.61	2.49	2.66	0.26	0.01
C18:2, n-6	14.61	14.46	14.97	14.84	15.40	14.57	14.81	0.34	23.64	24.14	22.71	20.91	22.21	22.71	22.72	1.13	0.00
C20:0	0.07	0.07	0.07	0.07	0.06	0.06	0.07	0.00	0.06	0.05	0.05	0.05	0.06	0.06	0.05	0.00	0.00
C18:3, n-6	0.10	0.11	0.11	0.10	0.09	0.09	0.10	0.01	0.19	0.18	0.15	0.14	0.15	0.16	0.16	0.02	0.00
C18:3, n-3	2.74	2.65	3.00	2.88	2.88	2.77	2.82	0.12	2.41	2.56	1.88	1.74	2.31	2.51	2.24	0.35	0.00
C20:1, n-9	0.82	0.77	0.83	0.87	0.81	0.84	0.82	0.04	0.39	0.38	0.38	0.36	0.37	0.37	0.38	0.01	0.00
C20:2, n-6	0.40	0.37	0.29	0.35	0.39	0.38	0.36	0.04	0.52	0.48	0.71	0.80	0.47	0.43	0.57	0.15	0.01
C20:3, n-6	0.51	0.58	0.40	0.44	0.47	0.49	0.48	0.06	0.53	0.44	0.74	0.87	0.55	0.49	0.60	0.17	0.12

C20:3, n-3	0.30	0.27	0.26	0.32	0.29	0.29	0.29	0.02	0.12	0.11	0.14	0.18	0.11	0.10	0.13	0.03	0.00
C20:4,n-6+C22:1, n-9	1.75	1.81	1.47	1.40	1.91	1.71	1.67	0.20	2.14	2.04	4.20	4.43	2.65	2.24	2.95	1.08	0.02
C20:4, n-3	0.17	0.19	0.15	0.18	0.19	0.23	0.19	0.02	0.13	0.11	0.23	0.26	0.16	0.13	0.17	0.06	0.54
C20:5, n-3	0.76	0.79	0.59	0.68	0.75	0.76	0.72	0.07	0.24	0.22	0.41	0.53	0.30	0.25	0.32	0.12	0.00
C22:5, n-3	1.39	1.53	1.29	1.24	1.55	1.34	1.39	0.13	0.67	0.70	1.40	1.49	0.78	0.68	0.95	0.38	0.02
C22:6, n-3	2.58	2.43	2.03	2.11	2.25	2.35	2.29	0.21	0.68	0.69	1.53	1.60	0.86	0.72	1.01	0.43	0.00
LA/ALA	5.43	5.46	4.99	5.16	5.52	5.40	5.33	0.21	9.86	9.43	12.23	12.57	9.72	9.16	10.50	1.50	0.00
n-6/n-3	2.25	2.20	2.36	2.31	2.33	2.26	2.28	0.06	6.36	6.23	5.12	4.75	5.76	5.96	5.69	0.64	0.00
AA/EPA	2.21	2.30	2.49	2.05	2.42	2.19	2.28	0.16	8.60	9.19	10.44	8.94	8.69	8.51	9.06	0.72	0.00
16:1c9/16:0	0.15	0.16	0.16	0.16	0.14	0.15	0.15	0.01	0.19	0.17	0.11	0.13	0.17	0.17	0.16	0.03	0.73
18:1c9/18:0	5.49	5.40	7.01	6.29	5.71	5.94	5.97	0.60	4.89	4.83	2.91	3.04	4.48	4.72	4.15	0.92	0.00

4.4 Mg fatty acid/100 g breast muscle

Fatty acid concentration (mg /100 g muscle) was calculated by using IS C13:0 as shown in table 13. The table shows the mean results, SD and t-test.

Table 13: Fatty acid concentration (mg /100 g muscles) in breast muscle from birds fed diet 1 and diet 2.

	Diet 1		Diet 2		
	Mean	SD	Mean	SD	T-test
C12:0	0.28	0.03	0.21	0.09	0.13
C14:0	8.03	1.04	5.33	2.31	0.03
C15:0	1.24	0.15	1.07	0.38	0.32
C16:0	238.40	24.31	263.19	94.70	0.55
C16:1,c9	38.28	4.98	44.60	21.07	0.49
C17:0	1.47	0.18	1.45	0.45	0.92
C18:0	77.42	3.89	84.32	18.24	0.39
C18:1, c9	477.65	54.55	368.22	144.27	0.11
C18:1, c11	37.23	3.32	32.44	7.97	0.20
C18:2, n-6	180.49	27.65	294.78	104.63	0.03
C20:0	0.91	0.10	0.71	0.26	0.10
C18:3, n-6	1.28	0.14	2.15	0.88	0.04
C18:3, n-3	37.44	5.11	30.59	13.67	0.28

C20:1, n-9	10.72	1.34	4.79	1.60	0.00
C20:2, n-6	4.32	0.29	6.45	0.88	0.00
C20:3, n-6	5.69	0.23	6.68	0.65	0.01
C20:3, n-3	3.66	0.43	1.46	0.23	0.00
C20:4, n-6 + C22:1, n-9	19.16	1.05	31.62	2.44	0.00
C20:4, n-3	2.19	0.25	1.84	0.12	0.01
C20:5, n-3	8.60	0.33	3.57	0.26	0.00
C22:5, n-3	16.23	1.10	10.24	1.02	0.00
C22:6, n-3	26.61	1.46	10.79	1.29	0.00
sum	1197.30	116.84	1206.52	408.87	0.96
LA/ALA	5.21	0.30	10.50	1.50	0.00
n-6/n-3	2.22	0.18	5.69	0.64	0.00
AA/EPA	2.28	0.16	9.06	0.72	0.00
sum n-6	210.94	28.35	341.69	106.54	0.02
sum n-3	94.73	6.90	58.49	13.30	0.00

4.5 Mg fatty acids/100gbreast muscle of the chickens

Fatty acid concentration (mg/100gm muscle) was calculated by using IS C13:0 as shown in table 14. The fatty acid composition is shown as the mean composition of 3 birds in each cage. In diet 1 and diet 2, numbers of cages were 6 and in each cage there were 3 birds. Thus total number of birds was 18 in diet 1 and in diet 2. We calculate mean, SD from diet 1 and diet 2 and t-test were used to compare diet1 and diet 2.It is found that C14:0, C18:2 n-3, C18:3 n-3, C20:1 n-9, C20:2 n-6, C20:3n-6, C20:4 (n-6)/C22:1 n-9, C20:4 n-3, C20:5 n-3, C22:5 n-3,C22:6n-3 , LA/ALA, n-6/n-3, AA/EPAwere significant different between two groups. However, It was found that C12:0,C15:0, C16:0, C16:1,c9 , C17:0, C18:0, C18:1 c9, 18:1 c11, C20:0,C18:3 n-3 were non significantly different.

Table 14: Detailed showing, mg/100g muscle of the chicken

Cage	1	2	3	7	8	9	Mean	SD	4	5	6	10	11	12	Mean	SD	t-test
C12:0	0.29	0.22	0.30	0.27	0.29	0.29	0.28	0.03	0.26	0.29	0.12	0.10	0.22	0.29	0.21	0.09	0.13
C14:0	8.96	6.08	8.85	7.98	8.05	8.23	8.03	1.04	6.14	7.45	2.84	2.35	5.34	7.87	5.33	2.31	0.03
C15:0	1.32	0.95	1.34	1.22	1.32	1.31	1.24	0.15	1.21	1.40	0.73	0.52	1.07	1.47	1.07	0.38	0.32
C16:0	255.08	193.09	260.75	241.84	232.46	247.21	238.40	24.31	300.51	333.42	163.05	136.19	271.94	374.01	263.19	94.70	0.55
C16:1,c9	44.03	30.44	42.35	38.09	35.00	39.79	38.28	4.98	58.63	57.02	18.27	18.37	49.06	66.25	44.60	21.07	0.49
C17:0	1.49	1.14	1.65	1.44	1.62	1.48	1.47	0.18	1.50	1.89	1.14	0.78	1.44	1.94	1.45	0.45	0.92
C18:0	81.19	69.98	77.97	77.70	77.92	79.77	77.42	3.89	83.93	94.77	73.80	57.80	84.10	111.54	84.32	18.24	0.39
C18:1,	488.7	377.	545.3	488.7	483.0	482.4	477.6	54.5	418.0	458.6	216.	179.	382.1	554.3	368.2	144.	0.1

c9	7	63	1	1	0	7	5	5	9	3	44	70	5	4	2	27	1
C18:1, c11	38.65	31.33	41.40	37.98	37.31	36.70	37.23	3.32	35.49	37.45	24.91	20.94	33.81	42.07	32.44	7.97	0.20
C18:2, n-6	142.28	151.08	212.37	191.11	198.18	187.91	180.49	27.65	338.32	379.27	196.96	148.01	290.58	415.52	294.78	104.63	0.03
C20:0	0.99	0.77	1.02	0.92	0.94	0.83	0.91	0.10	0.80	0.84	0.44	0.37	0.76	1.04	0.71	0.26	0.10
C18:3, n-6	1.25	1.17	1.54	1.32	1.24	1.15	1.28	0.14	2.76	2.86	1.26	0.99	1.96	3.07	2.15	0.88	0.04
C18:3, n-3	39.80	27.91	42.64	37.19	40.09	37.01	37.44	5.11	35.22	40.23	16.45	12.74	30.96	47.95	30.59	13.67	0.28
C20:1, n-9	11.29	8.03	11.67	11.31	11.02	10.98	10.72	1.34	5.57	6.02	3.23	2.54	4.80	6.57	4.79	1.60	0.00
C20:2, n-6	4.47	3.83	4.10	4.53	4.53	4.46	4.32	0.29	7.19	7.60	6.03	5.29	5.85	6.77	6.45	0.88	0.00
C20:3, n-6	5.72	5.95	5.63	5.63	5.32	5.92	5.69	0.23	7.25	6.83	6.05	5.77	6.80	7.40	6.68	0.65	0.01
C20:3, n-3	3.92	2.86	3.64	4.10	3.79	3.66	3.66	0.43	1.71	1.72	1.21	1.21	1.36	1.55	1.46	0.23	0.00
C20:4, n-6/C22:1, n-9	18.32	18.70	20.64	17.91	19.41	20.02	19.16	1.05	28.30	31.70	35.38	29.73	32.46	32.16	31.62	2.44	0.00
C20:4, n-3	1.99	1.92	2.14	2.36	2.11	2.59	2.19	0.25	1.79	1.71	1.96	1.71	1.95	1.93	1.84	0.12	0.01
C20:5, n-3	8.67	8.23	8.29	8.74	8.56	9.15	8.60	0.33	3.33	3.48	3.40	3.44	3.77	4.00	3.57	0.26	0.00
C22:5, n-3	14.93	15.91	18.18	15.82	16.61	15.92	16.23	1.10	8.80	10.86	11.81	9.94	9.82	10.21	10.24	1.02	0.00
C22:6, n-3	26.67	25.22	28.35	27.06	24.57	27.77	26.61	1.46	9.14	10.74	13.11	10.55	10.79	10.41	10.79	1.29	0.00
Sum	1200.	982.	1340.	1223.	1213.	1224.	1197.	116.	1355.	1496.	798.	649.	1231.	1708.	1206.	408.	0.9

mg/100 g	06	42	12	23	33	61	30	84	95	15	58	04	01	38	52	87	6
LA/ALA	4.75	5.46	4.99	5.16	5.52	5.40	5.21	0.30	9.86	9.43	12.23	12.57	9.72	9.16	10.50	1.50	0.00
n-6/n-3	1.87	2.20	2.36	2.31	2.33	2.26	2.22	0.18	6.36	6.23	5.12	4.75	5.76	5.96	5.69	0.64	0.00
AA/EPA	2.21	2.30	2.49	2.05	2.42	2.19	2.28	0.16	8.60	9.19	10.44	8.94	8.69	8.51	9.06	0.72	0.00
sum n-6	172.03	180.72	244.28	220.50	228.68	219.46	210.94	28.35	383.82	428.25	245.69	189.79	337.66	464.91	341.69	106.54	0.02
sum n-3	95.97	82.04	103.24	95.27	95.72	96.10	94.73	6.90	59.99	68.73	47.93	39.58	58.65	76.05	58.49	13.30	0.00

5. Discussion

5.1 Final body weight

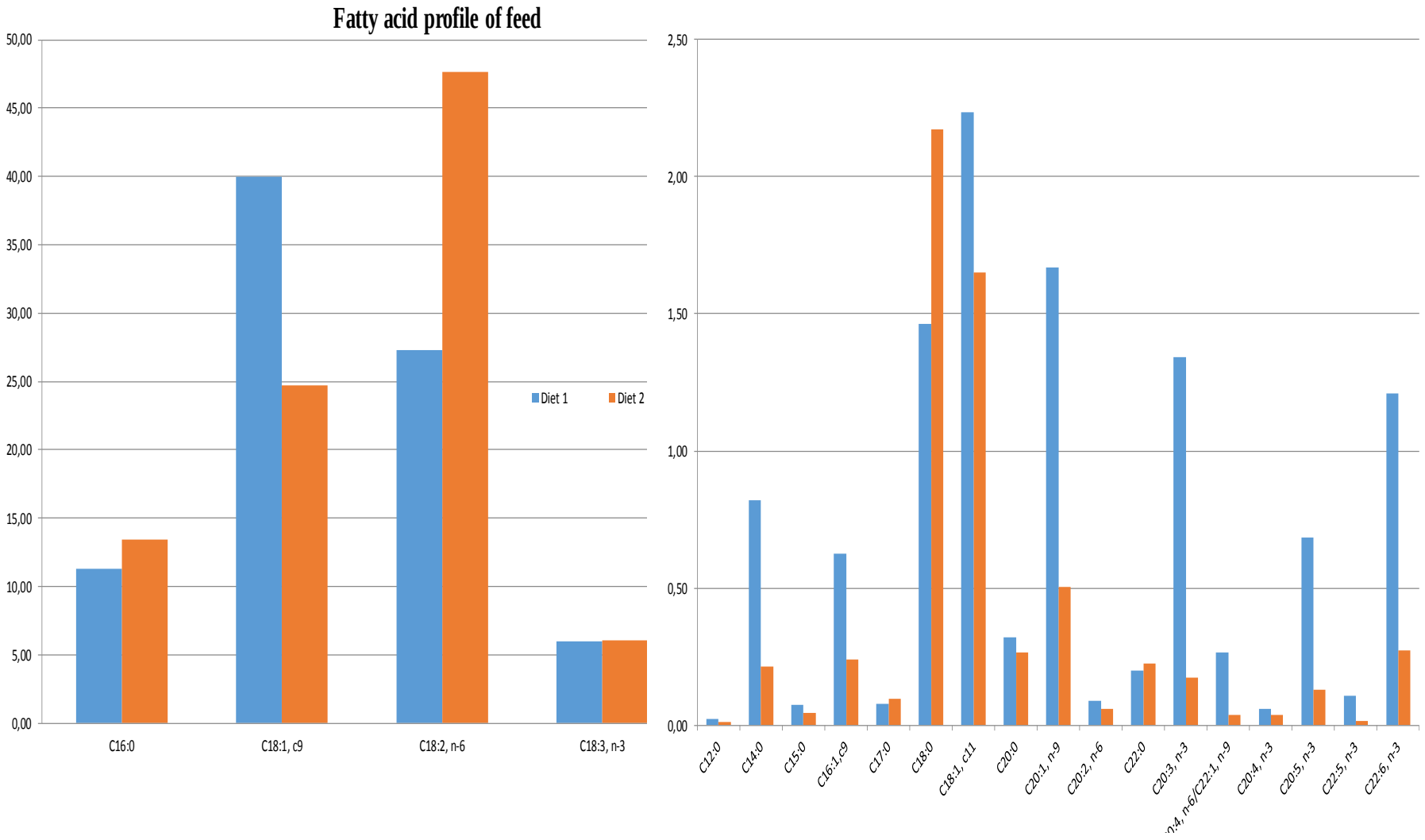
The final body weights were not different in the two groups of the chickens inspite of two different diets were given to the chicken as presented in table 10. Both diets gave the good growth. This finding makes it easier to compare the fatty acid concentration of two feed groups. Commercial feed was not better than the experimental feed we made. As shown in table 9 total fatty acid were 5.1g/100g in breast muscle from chicken fed diet 1 and 4.7g in diet 2. Sharifi et al. (2012) conduct the experiment and found that soybean oil in feed resulted increased body weight of the chicken. However, our finding is in contrast to Sharifi et al. (2012)

5.2 Fatty acid composition of feeds and breast muscle

The main finding in the present study was that the diet containing rapeseed oil (diet 1) resulted in a large reduction in arachidonic acid in chicken breast muscle to 19.2 mg/100g compared to 31.6 mg/100g in chicken fed diet 2, containing soybean oil. Such a reduction is also shown by others (Haug et al. 2012).

The other important finding in the present study is that the concentration of EPA, DPA and DHA were doubled in the breast muscle from the chickens given diet 1 compared to diet 2. This has also been shown in other studies where rapeseed oil is used instead of soybean oil in the feed (Haug et al. 2012). However, diet 1 contained also fish meal, as a source of long chain n-3 fatty acids, and the increase in EPA, DPA and DHA may be caused by the higher content of these fatty acids compared to diet 2.

In our study fatty acid composition of feeds, the fatty acids C12:0, C14:0, C15:0, C16:1 c9; C18:1 c9, C18:1 c11, C20:0, C20:1 n-9, C20:2 n-6, C20:3 n-3, C20:4 (n-6/C22:1, n-9), C20:4 n-3, C20:5n-3 ,C22:5n-3and C22:6, n-3 were higher in diet 1 than diet 2 whereas remaining fatty acids C16:0, C17:0, C18:0, C18:2 n-6, C18:3 n-3, C22:0 were higher in diet 2 than diet 1. Fatty acid profile of feed: C16:0, C18:1 c9, C18:2 n-6, C18:3 n-3 shows mirror with fatty acid profile of chicken breast muscle as shown in figure 8. However, linoleic acid can be converted to arachidonic acid in chicken through desaturation and elongation process. EPA, DPA and DHA, n-3 derivatives of α-linolenic acid and can be synthesized in chicken through desaturation and elongation.


Fatty acid profile of feed
50,00
45,00
40,00
35,00
30,00
25,00
20,00
15,00
10,00
5,00
0,00
C16:0
C18:1, c9
C18:2, n-6
C18:3, n-3
Diet 1
Diet 2
2,50
2,00
1,50
1,00
0,50
0,00
C12:0
C14:0
C15:0
C16:1,c9
C17:0
C18:0
C18:1, c11
C20:0
C20:1, n-9
C20:2, n-6
C22:0
C20:3, n-3
C20:4, n-6/C22:1, n-9
C20:4, n-3
C20:5, n-3
C22:5, n-3
C22:6, n-3

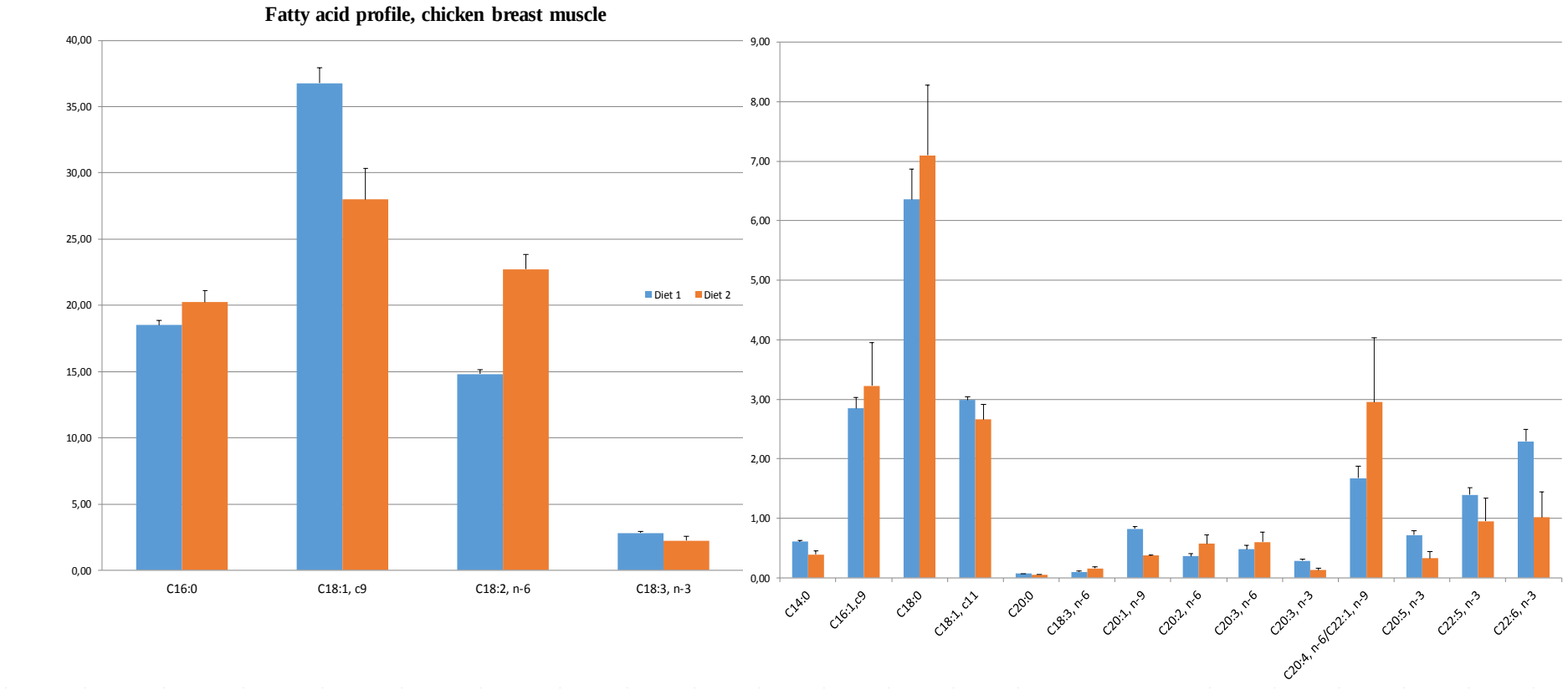


Figure 8: Fatty acid composition of feeds and breast muscle

Diet 2, Soybean oil feed contained more of the saturated fatty acids palmitic acid (C16: 0) and stearic acid (C18: 0) than rapeseed oil feed (diet 1). This can be explained by soybean oil containing 10.5 % Palmitic acid, while rapeseed oil consists of 4.3% palmitic (National Agricultural Library, 2015). It is a higher concentration of linoleic acid (n -6) and about the same concentration of α-linolenic acid (n-3) in diet 2, soybean oil feed compared to diet 1, rapeseed oil feed. Soya oil contains 50% linoleic acid, while rapeseed oil only consists of about 20% linoleic acid. There were lower amounts of AA, EPA, DPA and DHA in diet 2 compared to diet 1. Higher EPA, DPA and DHA are due to addition of 60 g fish meal/kg in diet 1. Soybean oil feed consists of more linoleic acid and this lead to a less favorable ratio of n-6 / n-3 in soy oil feed, diet 2. Ratio between n-6 / n-3 is important as in our study we got 7.4 for diet 2 and 3.5 for diet 1, rapeseed oil feed group. There is a surplus of n-6 in the western diet, and change of use of oils in feed for livestock could contribute to a shift to a more favourable ratio of polyunsaturated fatty acids.

According to Scaife et al. (1994) there is strong correlation between the fatty acid compositions of feed and fatty acid composition of muscle of chicken. Chickens fed with diet 2, soybean oil feed, had more saturated fatty acids like palmitic acid and stearic acid in muscle compared to chickens fed rapeseed oil feed. The large amount of palmitic acid may be either by de novo synthesis and elongated to stearic acid or from feed intake (Ratnayake and Galli 2009). Palmitic and stearic acids have no double bond in their molecules. Due to this reason, oil with high saturated fatty acid content has higher oxidative stability than compared to unsaturated fatty acids. High content of saturated fatty acids like primarily palmitic acid which has being claimed to have negative effects on health relating to cholesterol and heart disease (Wilson 2004).

However, C12:0 (Lauric acid) was highest in diet 1 but in muscle there were no difference the two diet groups. The reason to this is not easy to find out. Likewise, eicosadienoic acid C20:2, n-6 was highest in diet 1 but in muscle there were no mirror. The reason to this is not easy to find out.

Oleic acid (C18:1, c9) was significantly different between the two diets. It was less oleic acid in diet 2, and it was less oleic acid in breast muscle from diet 2 fed birds compared to diet 1, containing the rapeseed oil. This is because rapeseed oil contains much (61%) oleic acid (National Agricultural Library, 2015). Oleic acid in feed was mirroring the oleic acid content of breast muscle of chicken. Oleic acid can compete to α-linolenic acid, EPA and DHA and

arachidonic acid on the same space in membranes. This means that the oleic acid is one of the fatty acids which can regulate the amount of arachidonic acid found in the body's fat membranes and thus the amount of arachidonic acid which can be released and is available for formation of prostaglandins 2 series (Haug et al. 2010). Rapeseed contained more oleic acid than soy oil and will possibly reduce overproduction of 2 series prostaglandins in an outbreak of disease and thus help in lower inflammation. Monounsaturated fat is liked because the oil is more stable at high cooking temperature for greater use in food and industrial products. Moreover, high oleic acid concentration improves the oxidative stability of the oil, and reduces the need for hydrogenation which generates trans-fats that are negatively associated with heart health in humans (Wilson 2004).

Comparing soybean oil with rapeseed oil, soybean oil contains more linoleic acid than rapeseed oil. Linoleic acid is converted to arachidonic acid in chicken through desaturation and elongation process as shown in figure 7 (Ratnayake and Galli 2009). Thus it is as expected that soybean oil fed chicken has more arachidonic acid in breast muscle than rapeseed fed oil chicken. Less arachidonic acid in chickens that were fed the rapeseed diet may be due to competition between linoleic acid and α-linolenic acid at Δ 5 and Δ6 desaturase. When it was less linoleic acid in rapeseed oil, there will be α-linolenic acid have an advantage over linoleic acid, forming less n-6 and more n-3. EPA, DPA and DHA, n-3 derivatives of α-linolenic acid synthesized in chicken similar to arachidonic acid by desaturation and elongation as explained in the figure 7 (Ratnayake and Galli 2009).

In our study, the concentration of α-linolenic acid is almost similar in the two diets. EPA, DPA and DHA, n-3 derivatives of α-linolenic acid synthesized in chicken similar to arachidonic acid by desaturation and elongation (Ratnayake and Galli 2009). In chicken fed diet 1 with rapeseed oil, 60g fish meal is added to the diet containing EPA, DPA and DHA. We got same pattern of EPA, DPA and DHA in chickens' breast muscle and diet. Increased levels of n-3 polyunsaturated fatty acids in chickens that have ingested oils and thus a good proportion α-linolenic acid has been shown by (Lopez-Ferrer et al. 2001).This corresponds with our finding when we found significant greater amounts of the long chain fatty acids EPA,DPA and DHA. The more double bonds presence there in the molecules, the more susceptible the fatty acids are oxidation (Chow 2007). However, when used for cooking at high temperature, oils high in polyunsaturated fatty are unstable, easily oxidized and cause off-flavors (Lee et al. 2009). Moreover, it reduces shelf life which limits the storage time of manufactured food products (Warner and Fehr 2008). In a study conducted by (Harper et al.

2006) was given 3 g α-linolenic acid per day. α-linolenic acid incorporated rapidly into lipoproteins, and provide increased levels of EPA and DPA that is why it is important to get this EPA, DHA and DPA in meat from chicken fed α-linolenic acid.

The ALA intake in western countries should be at least doubled to reach an acceptable level for C18:2 n-6/C18:3 n-3 ratios have been reported by (Weill, Schmitt et al. 2002). We want higher amount of ALA but not too much LA (Simopoulos et al. 1999). In western countries, the ratio between LA/ALA is from 5:1 to 20:1. AA and EPA compete for enzymes elongation of eicosanoids and an increased intake of EPA in the diet can reduce the cells' capacity to synthesize eicosanoids from AA. Thus, the balance between AA and EPA in the cell membrane is essential and low ratio of these two fatty acids is beneficial. In the present study, the ratio AA/EPA is 2.28:1 for the rapeseed oil fed group and 9.06:1 for the soybean oil fed group. Since eicosanoids from AA has inflammatory and allergenic effects, it would be advantageous to reduce the formation of this effect (Ratnayake and Galli 2009). This can be achieved by reducing the intake of LA or increased intake of ALA and EPA.

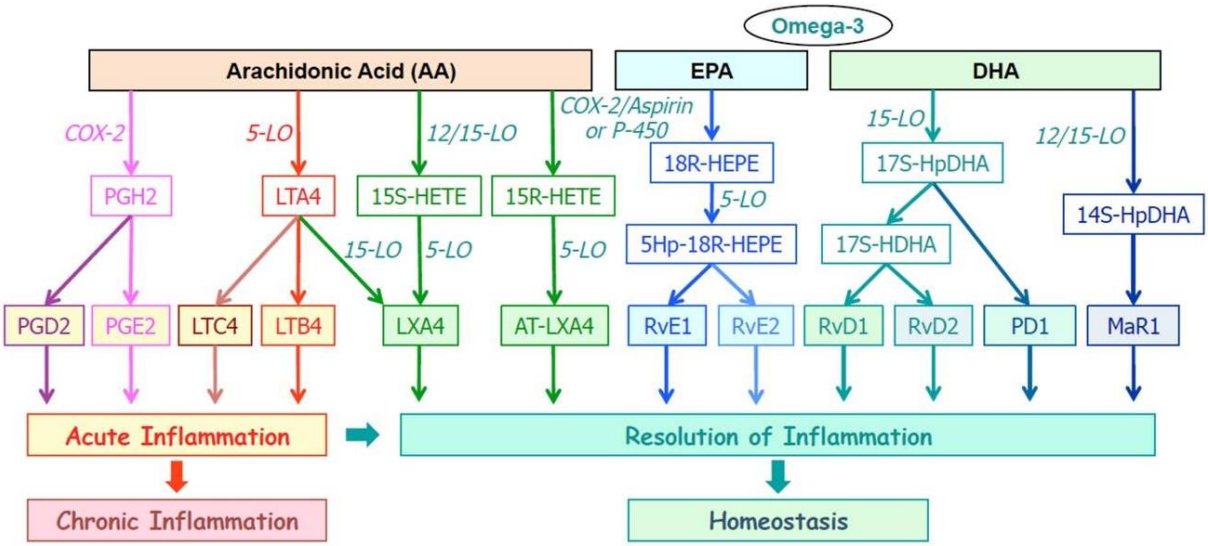


Figure 9: Biosynthetic of selected lipid mediators derived from AA, EPA, and DHA (Serhan & Petasis 2011).

AA is synthesis from linoleic acid in the animal and linoleic acid was highest in diet 2 (soybean oil fed group). In the rapeseed feed the concentration of AA was higher compared to soybean feed. But in muscle the concentration of AA was highest in soybean fed animals. This may be due to higher synthesis of AA from linoleic acid in the soybean oil fed animals.

Strandvik (2011) points out the importance of lowering the intake of n-6 instead of increasing the intake of n-3. This implies a paradigm shift in the government's recommendations regarding intake of oils rich in n-6. A reduction in the n-6 intake would also seem efficient on n-3 need; thereby prevent pillaging of the sea for its marine sources of long chain n-3 fatty acids. Access to marine n-3 LCPUFA is limited worldwide, and it will be positive to find other sources of n-3 LCPUFA than marine sources.

There is a challenge by changing the fat source in diets of soybean oil and instead to use rapeseed oil. The n-3 unsaturated fatty acids are more easily oxidizable than n-6 fatty acids. Unsaturated fatty acids are vulnerable to rancidity, and the larger content of unsaturated fatty acids in the diet, the greater the chance of rancidity (McDonald 2002). Furthermore rapeseed is rather similar in price compared to soybean oil, and the price of the concentrates will not change much. However, it is estimated that this increased feed costs are many times lower than the price of health gains in the population when they get adequate intake of the long n-3 fatty acids. The most important thing to consider is human health and nutrition.

6 Conclusion

The present study was designed to compare fatty acid profile of chicken breast muscle fed a diet containing rapeseed oil with ordinary soybean oil feed. Overall, the results showed that the fatty acid composition in chicken meat reflected the fatty acid composition in the two different diets. The final body weight was not different between the two diet groups. Overall, this study suggests that it is possible to increase the consumer's intake of n-3 fatty acids EPA, DPA and DHA and to lower the intake of n-6 fatty acid AA without the consumer having to change their eating habits.

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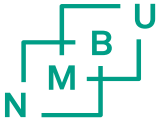

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