EFFECT OF ESSENTIAL UNSATURATED FATTY ACIDS IN FISH OIL ON LITTERS AND COMPOSITION OF MILK OF RABBIT DOES

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ABSTRACT

The aim of the study was to determine the degree to which a 3% fish oil supplementation in female rabbit diets affected the quantitative composition of fatty acids in milk, litter size, litter weight, and birth-to-weaning mortality of young rabbits in three successive litters. Twenty New Zealand White female rabbits of the foundation stock and all their offspring were investigated. Animals were assigned to two feeding groups: group I, fed a pelleted basal diet with standard components; group II, fed a pelleted diet with 3% fish oil. The fish oil was a byproduct of fish (herring, sprat, silver carp, mackerel, cod) meal production and contained 2.9% of C18:3n-3, 7.4% of C20:5n-3 (EPA), 12.6% of C22:6n-3 (DHA) and 0.8% of C22:5n-3 (DPA). Fat milk increased from 18.4 to 22.2% in female rabbits receiving 3% fish oil compared to the control group. In the milk fat of female rabbits, there were highly significant differences between the control and experimental groups in the level of polyunsaturated fatty acids of the n-3 series (n-3 PUFA) and saturated fatty acids (SFA). The fish oil supplementation reduced the level of short-chain fatty acids in milk (C8:0-C10) and increased the level of long-chain fatty acids (C16:1–C22:6). Milk was characterized by a higher concentration of EPA and DHA. Fat supplemented diets increased kit weight at birth, 21 days and 35 days of age ($P<0.01$) in the three reproductive cycles. In the experimental group, there was a higher percentage of reared rabbits in all the litters analysed. The high mortality of young rabbits among primiparous rabbits was due to relatively low milk production, which increased with parity. The bacteriological tests of the rabbits with diarrhoea symptoms showed that the main causes of mortality were the proliferation of conditionally pathogenic *Escherichia coli*, responsible for enteritis and the presence of *Streptococci*, which are relatively or conditionally pathogenic bacteria. There were no significant differences between the milk production of female rabbits from groups I and II, which shows that the fish oil supplementation had no effect on this parameter. Milk yield was significantly affected by litter size. For this reason, the lower mortality of young rabbits may result from the higher fat content of milk rather than from the higher milk yield.

Key words: Fish oil, Fatty acids, Milk.

INTRODUCTION

Fats as energy carriers and sources of essential unsaturated fatty acids have attracted the attention of nutritionists in recent years. The activity of these acids in the animal body is reflected mainly in the activity of eicosanoids (known as tissue hormones) synthesized from arachidic acid and eicosapentaenoic acid (EPA). Owing to the mechanism of their action, they can be treated as the most peripheral first messengers, which strengthen or weaken the regulatory activity of hormones and neuromediators at cellular level. Among the four families of essential unsaturated fatty acids, specific action is mainly shown by the linoleic acid (n-6) and the α-linolenic acid families (n-3). A favourable effect of n-3 acids was observed not only in diseases but also in maintaining the growth and development of mammalian bodies before and after birth.

The intense reproductive use of rabbits has led to the overlapping of particular reproductive stages, enabling the body resources to be simultaneously used for milk synthesis, milk secretion and fetal growth. Unfortunately, the intense reproduction causes a relatively high rearing mortality resulting from the birth of rabbits characterized by low body weight or the low milk yield of females.

The health quality and composition of feed is conditional on the onset of reproductive activity, embryo and fetal survival, neonatal weight, and the quantity and biological value of the milk secreted by females.

Many experiments attempted to increase milk yield by using high-energy rations with an increased carbohydrate content, but failed to produce anticipated results.

Among the many nutrients used in feeding, special attention has recently been given to the quantity and quality of fats. Long-chain fatty acids from fish oil, especially C20:5 n-3 (EPA), C22:5n-3 (DPA) and C22:6n-3 (DHA) enrich milk and animal tissues in n-3 PUFA in a way that is physiologically most effective.

Studies on the effect of essential unsaturated fatty acids found in fish oil on gilt productivity showed considerable improvements in piglet health, higher daily gains and lower mortality. D'Ambola *et al*. (1991) reported that newborn rabbits whose mothers were fed diets enriched with fish and saffron oil had better defensive mechanisms against lung diseases.

The aim of the study was to determine the degree to which a 3% fish oil supplement in female rabbit diets affects the quantitative composition of higher fatty acids in milk, litter size, litter weight, and birth-to-weaning mortality of young rabbits in three successive litters.

MATERIALS AND METHODS

Twenty New Zealand White female rabbits of the foundation stock and all of their offspring were investigated. Animals were assigned to two feeding groups: group I – fed a pelleted basal diet with standard components, group II – fed a pelleted diet with 3% fish oil. The fish oil used was a byproduct of fish (herring, sprat, silver carp and mackerel) meal processing and contained 2.9% of C18:3n-3, 7.4% of C20:5n-3 (EPA), 12.6% of C22:6n-3 (DHA) and 0.8% of C22:5n-3 (DPA).

The rabbits were fed a complete standard diet, which contained dried meadow grass, soybean meal, wheat bran, ground maize, milk replacer, NaCl, and a mineral and vitamin supplement The feeds were analysed for dry matter, crude ash, crude protein, crude fat, crude fibre, N-free extractives, and the level of higher fatty acids using gas chromatography.

Females of the foundation stock were subjected to rationed feeding and received daily rations ranging from 150 g feed during the non-coupling period to 300 g during gestation and rearing of the young to 21 days. After this period, the rations were increased to meet the nutrient requirements of growing rabbits.

Females were kept singly in cages on deep litter from wheat straw. The experiment covered three successive litters. On the fifth day of the third lactation, milk samples were taken from 6 females of each group by hand milking of all active teats. Basic milk composition was determined using conventional methods (MilkoScan 104, A/S N. Foss Electric, Denmark). The composition of milk fatty acids was analysed using gas chromatography.

To determine productivity, females were analysed for fertility, prolificacy and other parameters such as the body weight of rabbits on the day of birth and at 21 and 35 days of age, and the milk yield of females using a formula given by Niedźwiadek (1981): $M=[(C_2-C_1):(21xC_2)]x100$ where M – coefficient of milk yield, C_1 – litter weight (g) after birth, C_2 – litter weight (g) at 21 days of age.

The results were analysed statistically by analysis of variance and Duncan's test, using Statgraphics Plus 4.0 software.

RESULTS AND DISCUSSION

Table 1 presents the results of basic analysis for complete pelleted diets. The addition of fish oil mixture increased the proportion of crude fat from 2.42% to 5.19%.

Table 1: Nutrient content of the feeds (%)

Group	Dry matter	Crude ash	Crude protein	Crude fat	Crude fibre	N-free extractives
I	89.4	5.83	16.7	2.42	12.2	58.1
II	87.7	5.21	16.9	5.19	12.4	53.2

In the chemical composition of milk from female rabbits receiving a 3% fish oil supplement, the proportion of fat increased from 18.44 to 22.23% compared to the control group (Table 2). The type of diet had no effect on milk protein content, probably due to the constant level and source of protein.

Table 2: Chemical composition and pH of rabbit milk (%)

Components	Group I	Group II	SE
Fat	18.44A	22.23B	1.12
Protein	12.65	12.28	0.98
Lactose	1.98	2.12	1.40
pH	5.5	5.8	0.95

Table 3 presents the profile of higher fatty acids in feed and milk fat from rabbits in their third lactation (day 5). The increase in digestible energy content of the diet, induced by a fish oil supplement, caused highly significant changes in the majority of fatty acids found in milk fat.

Table 3: Determination of the profile of higher fatty acids in the diets and milk fat of female rabbits (% of total acids)

Item	Feed		Milk		SE
	Group I	Group II	Group I	Group II	
C8:0	0.00	0.00	33.6A	16.4B	0.95
C10:0	0.00	0.00	19.9A	12.3B	1.12
C12:0	0.00	0.00	1.62	1.54	0.84
C14:0	0.18	0.76	2.72A	1.94B	1.12
C16:0	16.3	12.2	7.76A	12.9B	1.30
C16:1	0.22	0.97	2.33	2.40	0.95
C18:0	2.13	1.92	1.99a	2.20b	0.98
C18:1	20.2	33.8	7.84A	11.5B	1.16
C18:2n-6	52.8	37.2	22.0A	31.2B	1.34
C18:3n-3	6.74	9.04	1.42A	2.96B	1.11
C20:0	0.56	0.57	0.03a	0.06b	0.46
CLAc9t11	0.06	0.11	0.12A	0.55B	0.97
CLAt10c12	0.04	0.09	0.00	0.01	1.24
CLAc9c11	0.00	0.00	0.01	0.01	1.45
CLAt9t11	0.08	0.04	0.01A	0.05B	1.20
C20:4n-6	0.00	0.00	0.11a	0.21b	0.87
C20:5n-3(EPA)	0.00	1.91	0.03A	0.91B	1.34
C22:6n-3(DHA)	0.00	2.80	0.03A	2.55B	1.33
C22:0	0.49	0.43	0.01A	0.07B	0.95
C22:1	0.10	0.39	0.01a	0.04b	0.45
SFA	19.6	15.8	67.6A	47.4B	0.89
UFA	80.4	84.1	33.9A	52.5B	1.10
MUFA	20.6	35.2	10.1A	13.9B	0.95
PUFA	59.7	49.9	23.8A	38.6B	1.12
PUFAn-6	52.8	37.2	22.1A	31.5B	1.46
PUFAn-3	6.74	13.7	1.50A	6.43B	1.12
PUFA/SFA	3.04	3.08	0.36A	0.81B	0.98

a,b – means with different letters differ significantly at $P<0.05$, A,B at $P<0.01$

As regards the milk fat of female rabbits, highly significant differences were found between the control and experimental groups in the level of polyunsaturated fatty acids of the n-3 series (n-3 PUFA) and in saturated fatty acids (SFA). The fish oil supplement reduced the level of short-chain fatty acids in milk (C8:0-C10) and increased the level of long-chain fatty acids (C16:1 – C22:6). The milk was characterized by a higher concentration of EPA and DHA.

Dietary factors have the greatest effect on milk CLA content, although the stage of lactation, breed and age may also have some effect. The supplementation of female rabbit diet with fish oil caused a highly significant increase in the isomers of linoleic acid CLAc9t11, which were shown in studies with model animals to inhibit the growth of cancerous cells (Roche, 2001; Corl *et al*. 2003).

Table 4 shows reproductive results for three successive litters of female rabbits.

Table 4: Reproductive performance

Item	Litter I			Litter II			Litter III		
	Group I	Group II	SE	Group I	Group II	SE	Group I	Group II	SE
Mean litter size after birth (head)	6.6	6.2	6.13	6.8	6.9	7.10	6.8A	7.5B	9.11
Mean weight of 1 rabbit after birth (g)	61.5A	69.9B	2.50	62.1A	72.1B	3.52	62.7A	72.8B	4.52
No. animals at 21 d (head)	6.1	6.0	4.90	6.1A	6.7B	5.80	6.5	7.4	8.50
% of rabbits reared to 21 days	92.4	96.8		89.7	97.1		95.6	98.7	
Mean weight of 1 rabbit at 21 d (g)	282.0A	330.1B	9.20	304.0a	320.6b	13.4	307.6A	336.8B	16.4
No. of animals at 35 d (head)	5.4A	5.9B	3.20	5.9A	6.6B	6.8	6.1A	7.3B	6.8
Mean weight of 1 rabbit at 35 d (g)	717.3A	791.8B	11.0	724.3A	795.6B	13.6	723.5A	810.5B	12.5
% of rabbits reared	81.8	95.2		86.8	95.6		89.7	97.3	
Coefficient of milk yield (kg)	3.8	3.9	11.7	4.3	4.4	10.7	4.3	4.5	9.5

a,b – means with different letters differ significantly at $P<0.05$, A,B at $P<0.01$

The addition of fat to the dietary ration of female rabbits had a positive effect on increasing the neonatal weight of rabbits, which differed highly significantly between the analysed groups for all the litters analysed. This trend also persisted at 21 and 35 days of age. A higher percentage of reared young rabbits was found for all the litters analysed in the experimental group.

The high mortality of young rabbits among primiparous females was due to relatively low milk production, which increased with parity. The bacteriological tests of the rabbits with diarrhoea symptoms showed that the main causes of mortality were the proliferation of conditionally pathogenic *Escherichia coli*, responsible for enteritis, and the presence of beta-hemolytic *Streptococci*, which are relatively or conditionally pathogenic bacteria. The activation of both bacteria may result from the action of resistance-reducing factors such as hyperthermia, malnutrition, improper feed, vitamin-deficiency disease, invasion of coccidiosis, stress or overstocking.

According to Padilha *et al*. (1995), the number of *Escherichia coli*, which is 10^8 x g of caecum digesta in suckling rabbits, decreases linearly with age. This decrease may be due to the increased level of volatile fatty acids and reduced pH of intestinal digesta, which is determined by a change of food. Nutritional changes may modify fermented caecum digesta, and thus the activity and stability of bacterial flora. The proliferation of *Escherichia coli* is inhibited mainly by food that is rich in starch and contains digestible fibre, which is degraded mostly in the caecum. Mista *et al*. (2004) reduced *Escherichia coli* count by adding a mineral and fat supplement to the feed. Therefore, the higher percentage of young rabbits reared in the experimental group may be due to the favourable effect of fish oil fatty acids on the composition of caceum bacterial flora.

There were no significant differences in milk yield between females from groups I and II, which shows that the addition of fish oil has no effect on this parameter. Because milk yield is significantly affected by litter size, the observed reduction in the mortality of young rabbits may be due to the higher milk fat content rather than to the higher milk yield. As a result, although litter weight at 21 days of lactation is a good indicator of milk yield, when high-fat diets are used this may be due to the higher dietary fat content and milk yield may be overestimated. These results suggest that milk production should be determined using milk adjusted for fat or milk energy.

CONCLUSIONS

In the experiment with female rabbits fed a complete diet with 3% fish oil supplement, quantitatively and qualitatively better milk fat content, higher fertility and prolificacy values, higher body weight of young rabbits at birth and at 21 and 35 days of age, and lower mortality were obtained.

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