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EFFECT OF SOYBEANS AND PALM OIL ADDITION TO THE COWS DIETS ON MILK FATTY ACID PROFILE

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Effects of soybean and palm oil supplements on milk yields and fatty acids composition with considering intermediate products of unsaturated fatty acids hydrogenation were investigated. Fifteen multiparous Holstein cows in mid-lactation were assigned to groups of 5 each. Cows of control group fed (kg): alfalfa hay — 6; corn silage — 20; grass silage — 10; wheat — 3; corn — 2; soybean meal — 1; molasses — 2. In the diet of the 1st treated group, soybean meal was replaced by 1.5 kg of extruded soybeans. Diet of 2nd treated group cows was supplemented with 0.3 kg of palm oil. All diets were equivalent in contents of nutrients (except fat). Crude fat content in the diets was 0.70, 0.96 and 0.97 kg/cow/day, respectively. Treatment lasted 2 months. Milk yield was recorded weekly for the duration of the study. The milk fatty acids methyl esters were then quantified by gas chromatography.

In the milk of cows fed diet with soybeans was found 2.2 times more of trans-11 18:1 (P<0.001) and 1.3 times more of cis-9,trans-11 18:2 (P<0.05) fatty acids compared to cows of control group. Trans-11 18:1 and cis-9,trans-11 18:2 act in animal and human tissues as antagonists of ω-6 fatty acids and, respectively, they are synergists of ω-3 fatty acids. Thus an increased content of these acids improves milk dietary value.

Cows of both treated groups had higher milk protein yield for 40 g daily (P<0.05). Milk fat yield was increased only in cows fed palm oil, it was 50 g greater than in control cows (P<0.05). Both fat supplements increased the average daily milk yields. Milk yield of cows fed diet with soybeans addition was higher by 4 % (P<0.05), and in cows fed diet with palm oil milk yield grew to 7 % (P<0.05). Fat corrected milk yield was increased only in cows fed diet with palm oil addition (P<0.05). The results of the present study indicate a benefits of feeding cows by palm oil as dietary supplement.

Keywords: COWS, MILK, FATTY ACIDS, SOYBEANS, PALM OIL

ЖИРНОКИСЛОТНИЙ СКЛАД ЛІПІДІВ МОЛОКА ЗА ДОДАВАННЯ ДО РАЦІОНУ КОРІВ СОЄВИХ БОБІВ ТА ПАЛЬМОВОЇ ОЛІЇ

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Додавання до раціону високопродуктивних корів жирових добавок позитивно впливає на обмін речовин, підвищує молочну продуктивність та якість молока. Це пояснюється високою енергетичною цінністю жирів, їх нітрогензберігаючою дією в організмі корів, використання наявних у жирі жирних кислот у синтезі ліпідів молока.

Дослідження було проведено на 3 групах корів-аналогів української чорно-рябої молочної породи, по п'ять голів у кожній, продуктивністю 20–25 кг молока за добу. Корови контрольної групи отримували стандартний збалансований за вмістом поживних речовин раціон, що містив 670 г сирого жиру. Вміст жиру у раціонах корів 2-ї та 3-ї груп збільшували на 50 % за рахунок введення до їх складу відповідно соєвих бобів або пальмової олії. Раціон контрольних корів складався з (кг): сіна люцернового — 6,0, силосу кукурудзяного — 20, сінажу — 10, дерті пшеничної — 3,0, дерті кукурудзяної — 2,0, шроту соєвого — 1,0, меляси — 2,0. У раціоні корів 1-ї дослідної групи соєвий шрот замінювали на 1,5 кг соєвих бобів, а до раціону корів 2-ї дослідної групи додавали 0,3 кг пальмової олії. Тривалість досліду становила 2 місяці. У кінці досліду відбирали вмістиме рубця, кров і молоко. Жирнокислотний склад ліпідів плазми крові досліджували методом газорідинної хроматографії на хроматографі Hewlett Packard HP-6890 з полум'яно-іонізаційним детектором, обладнаному капілярною колонкою SP-2560 довжиною 100 м.

Зміна ліпідного та жирнокислотного складу раціону вплинула на жирнокислотний склад молочного жиру. У молоці корів 1-ї дослідної групи зменшилась кількість кислот ізо-15:0, антеізо-15:0 і 17:0 (Р<0,05–0,01) та зростала кількість 18:1 та 18:2 кислот (Р<0,01–0,001). У молоці корів 2-ї дослідної групи зросла кількість 14:0 кислоти (Р<0,05). У складі молочного жиру корів 1-ї дослідної групи виявлено у 2,2 рази більше кислоти транс-11 18:1 (Р<0,001) та у 1,3 рази більше її метаболіту — кислоти цис-9,транс-11 18:2 (Р<0,05). У корів 2-ї дослідної групи вміст транс-11 18:1 (Р<0,05) — був вищим у 1,3 рази, а вміст цис-9,транс-11 18:2 — у 1,2 рази. Частка лінолевої (цис-9,12 18:2) кислоти у молоці корів 1-ї дослідної групи зростала (Р<0,001), а 2-ї дослідної групи — зменшувалась (Р<0,01). У молочній залозі зменшувався синтез коротколанцюгових жирних кислот: С4:0, С6:0 і С8:0 (Р<0,05–0,01).

Вміст жиру у молоці корів контрольної та 1-ї і 2-ї дослідних груп становив, відповідно, 3,54; 3,37 і 3,51 %. Обидві жирові добавки підвищували середньодобові надої корів: у 1-й дослідній групі на 4 % (Р<0,05), а у 2-й — на 7 % (Р<0,05), проте у перерахунку на базисну жирність надої зростали лише у корів 2-ї дослідної групи (Р<0,05).

Ключові слова: КОРОВИ, МОЛОКО, ЖИРНІ КИСЛОТИ, СОЄВІ БОБИ, ПАЛЬМОВА ОЛІЯ

ЖИРНОКИСЛОТНЫЙ СОСТАВ ЛИПИДОВ МОЛОКА ПРИ ДОБАВЛЕНИИ К РАЦИОНУ КОРОВ СОЕВЫХ БОБОВ И ПАЛЬМОВОГО МАСЛА

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Добавление к рациону высокопродуктивных коров жировых добавок положительно влияет на обмен веществ, повышает молочную продуктивность и качество молока. Это объясняется высокой энергетической ценностью жиров, их азотсохраняющим действием в организме

коров, использование имеющихся в жире жирных кислот для синтеза липидов молока.

Исследование было проведено на 3-х группах коров-аналогов украинской черно-пестрой молочной породы, по пять голов в группе, производительностью 20–25 кг молока в сутки. Коровы контрольной группы получали стандартный сбалансированный по содержанию питательных веществ рацион, содержавший 670 г сырого жира. Содержание жира в рационах коров 2-й и 3-й групп увеличивали на 50 % за счет введения в их состав в соответствии соевых бобов или пальмового масла. Рацион контрольных коров состоял из (кг): сена люцернового — 6, силоса кукурузного — 20, сенажа — 10, дерти пшеничной — 3, дерти кукурузной — 2, шрота соевого — 1, мелассы — 2. В рационе коров 1-й опытной группы соевый шрот заменяли 1,5 кг соевых бобов, а к рациону коров 2-й опытной группы добавляли 0,3 г пальмового масла. Продолжительность опыта 2 месяца. Жирнокислотный состав липидов плазмы молока исследовали методом газожидкостной хроматографии на хроматографе Hewlett Packard HP-6890 с пламенно-ионизационным детектором, оборудованным капиллярной колонкой SP-2560 длиной 100 м.

Изменение липидного и жирнокислотного состава рациона повлияло на жирнокислотный состав молочного жира. В молоке коров 1-й опытной группы уменьшилось содержание кислот изо-15:0, антеизо-15:0 и 17:0 (P<0,05–0,01) и больше 18:1 и 18:2 кислот (P<0,01–0,001). В молоке коров 2-й опытной группы возросло количество 14:0 кислоты (P<0,05) . В составе молочного жира коров 1-й опытной группы обнаружено в 2,2 раза больше кислоты транс-11 18:1 (P<0,001) и в 1,3 раза больше ее метаболита — кислоты цис-9,транс-11 18:2 (P<0,05). У коров 2-й опытной группы содержание транс-11 18:1 (P<0,05) было выше в 1,3 раза, а содержание цис-9,транс-11 18:2 — в 1,2 раза. Количество линолевой (цис-9,12 18:2) кислоты в молоке коров 1-й опытной группы увеличилось (P<0,001), а 2-й опытной группы — уменьшилось (P<0,01). В молочной железе снизился синтез короткоцепочечных жирных кислот: 4:0, 6:0 и 8:0 (P<0,05-0,01).

Содержание жира в молоке коров контрольной, 1-й и 2-й опытных групп составил, соответственно, 3,54; 3,37 и 3,51 %. Обе жировые добавки повышали среднесуточные удои коров. В 1-й опытной группе на 4 % (P<0,05), а во 2-й — на 7 % (P<0,05), однако в пересчете на базисную жирность надои возросли только у коров 2-й опытной группы (P<0,05).

Ключевые слова: КОРОВЫ, МОЛОКО, ЖИРНЫЕ КИСЛОТЫ, СОЕВЫЕ БОБЫ, ПАЛЬМОВОЕ МАСЛО

Dietary sources of lipids can be added to the diet of lactating dairy cows to increase the energy density of the diet, modify milk production, and milk fatty acids profile [1–5]. Cows receiving fat supplements produce more milk per unit of consumed dry matter, due to better energy use associated with lower energy losses in the rumen, more efficient production of ATP from long-chain fatty acids than from acetate, direct inclusion of long-chain fatty acids in the fat milk [6]. Fat supplements to the cows diet decrease energy loss on thermoregulation and methanogenesis, reduce the risk of ruminal acidosis. Moreover, fat supplementation has some other effects, such as increased absorption of fat-soluble nutrients and reduced dustiness of feed [5].

Fat digestion and metabolism in ruminants has several features associated with fermentation in the rumen. Double bounds of dietary unsaturated fatty acids are isomerizes and saturated by ruminal bacteria [1–3]. In addition, ruminal bacteria synthesize specific fatty acids with odd and branched carbon chains [7]. Triglycerides of the diet are largely hydrolyzed by microbial lipases of rumen to yield free fatty acids, and double bounds of unsaturated fatty acids in turn undergo biohydrogenation to stearic acid and positional and geometrical isomers of oleic, linoleic and linolenic acids that delivered to the small intestine, absorbed to the blood and used by the organism, including for the synthesis of milk fat [1–3]. As a result, unsaturated fatty acids, including α-linolenic acid (cis-9,12,15 18:3) and linoleic acid (cis-9,12 18:2), are abundant in grass and certain other ruminant feedstuffs, yet are present at low concentrations in ruminant meat and milk.

Such intermediates of biohydrogenation as conjugated linoleic acid probably possess important metabolic properties. Cis-9,trans-11 18:2 is generally considered to be the main health-promoting

conjugated linoleic acid for human consumption [8, 9]. The *trans*-11 18:1 fatty acid, an intermediate of biohydrogenation, is also desirable as a product flowing from the rumen because *trans*-11 18:1 acts as a substrate for the formation of *cis*-9,*trans*-11 18:2 in the animal's own tissues [10]. Dietary *cis*-9,*trans*-11 conjugated linoleic acid have been shown in many animal studies to contribute to cancer prevention, decreased atherosclerosis, improved immune response, and altered protein or energy metabolism. *Trans*-10,*cis*-12 18:2 isomer reduces body fat [8, 11, 12]. Therefore, ruminant products contain potentially health-promoting conjugated linoleic acids mainly *cis*-9,*trans*-11 18:2 and *trans*-10,*cis*-12 18:2.

The main aim of investigations into ruminal biohydrogenation is to create healthier ruminant products. Increased milk concentration of both conjugated linoleic acid can be obtained by feeding of oilseeds and other fat supplements [14–17]. The objective of this experiment was to compare the effects of soybeans and palm oil feed supplement on cows' milk production and composition. Conjugated linoleic acid has drawn significant attention in the last two decades for its variety of biologically beneficial effects. At the same time, some researchers are skeptical of positive actions based on traditional ideas about the impact of trans fatty acids, so further research is needed to determine the health effects of *trans*-11 18:1 and *cis*-9,*trans*-11 18:2 acids [18, 19].

Materials and methods

Fifteen multiparous Holstein cows in midlactation were assigned to five treatments. The study was preceded by a 3 weeks randomized covariate period. All cows were fed the same diet during the covariate period and had free access to water. Diets were fed twice daily in equal. Cow's diets consisted of (kg): alfalfa hay — 6; corn silage — 20; grass silage — 10; wheat — 3; corn — 2; soybean meal — 1; molasses — 2. After that cows were assigned to groups of 5 each. Cows of control group further fed the same diet. In the diet of 1st experimental group soybean

meal was replaced by 1.5 kg of extruded at 130 °C exit temperature soybeans. Diet of 2nd experimental group cows was supplemented with 0.3 kg of palm oil. All diets were equivalent in contents of nutrients (except fat): crude protein — 3.2, crude fiber — 4.5, starch — 2.9, sugar — 1.7. Crude fat content in these diets was 0.70, 0.96, 0.97 kg/cow/day, respectively. Experimental period lasted 2 months.

Milk yield was recorded weekly for the duration of the study. Samples collected for the determination of protein, fat, and lactose were taken from two consecutive milkings of each week. Milk samples for the determination of fatty acid profiles were collected during the last week of test periods. Milk fat, protein, and lactose were determined by milk analyzer Ecomilk (Bulgaria).

Fatty acids methyl esters were prepared by ISO 15884:2002 [20]. Milk fatty acids were measured as described in ISO 15885:2002 [21]. Fatty acids were expressed as percentage of total fatty acids. The fatty acids methyl esters were then quantified by gas chromatograph Hewlett Packard HP-6890 with capillary column HP-88 (Agilent Technologies) 100 m lengths, 0.25 mm diameter, stationary phase film thickness 0.2 mcm. The temperature settings were 280 °C for the injector and 290 °C for the detector, and the column oven was temperature programmed from 40 to 260 °C. Flow rate for the carrier gas (He) was 1.2 ml/min. Peaks were quantified by peak area comparisons with a known amount of an internal standard (heptadecanoic acid, Sigma). Peaks were identified by comparison with known commercially prepared standards (37 component FAME mix, Supelco, № 47885-U; CLA isomers mix, Sigma-Aldrich, № 05632).

The SEM were calculated in Excel by dividing the SD by the square root of sample size. Statistics between control and each of experimental groups were calculated by ttest function in the Excel. Differences were considered significant at P<0.05.

Results and discussion

Changes in fatty acid composition of cows diet affected the fatty acid profile of milk fat (tabl. 1). Increasing of dietary fat content decreased the synthesis of some short-

chain fatty acids (particularly: butyric (4:0), caproic (6:0) and caprylic (8:0) (P<0.05-0.01)) by mammary gland of cows both treated groups. Additionally, milk of cows fed soybeans contained less myristic (14:0) acid (P<0.01), which is caused by more intensive usage of dietary fatty acids for milk synthesis in cows received fat supplements. Increased level of typical for palm oil myristic (14:0) acid (P<0.05) was found in a large amount in the milk of 2nd experimental group cows.

Table 1

Fatty acid profile of cows milk (% of total fatty acids), Σ±SEM (n=5)

Fatty acids	Cows groups		
	Control	1st experimental (soybeans)	2nd experimental (palm oil)
C 4:0	3.12±0.06	2.95±0.08	2.74±0.13*
C 6:0	1.44±0.04	1.21±0.09*	1.29±0.04*
C 8:0	1.95±0.06	1.56±0.06**	1.52±0.07*
C 10:0	5.23±0.26	5.29±0.34	5.44±0.24
C 12:0	5.4 10.08	4.59±0.20**	5.93±0.30
C 14:0	11.75±0.78	10.88±0.43	13.99±0.46*
Iso-C15:0	0.34±0.01	0.18±0.02*	0.21±0.02*
Anteiso-C15:0	0.33±0.03	0.21±0.02**	0.35±0.03
C 14:1	0.86±0.04	1.11±0.04**	1.05±0.04**
C 15:0	1.08±0.09	0.96±0.04	0.92±0.06
C 16:0	26.83±1.26	23.24±1.02	25.66±0.44
Iso-C17:0	0.30±0.03	0.28±0.04	0.37±0.03
C 16:1	1.72±0.06	1.64±0.08	1.73±0.06
Anteiso-C17:0	0.25±0.02	0.21±0.02	0.26±0.02
C 17:0	0.55±0.06	0.39±0.04**	0.52±0.09
C 17:1	0.15±0.02	0.20±0.01	0.18±0.01
C 18:0	11.18±0.53	11.88±0.65	10.57±0.47
C 18:1 6t	0.20±0.01	0.24±0.02	0.22±0.01
C 18:1 9t	0.14±0.01	0.17±0.02	0.19±0.03**
C 18:1 10t	0.48±0.03	0.53±0.04	0.47±0.02
C 18:1n 11t	0.82±0.03	1.84±0.04***	1.09±0.05*
C 18:1 6c	0.35±0.03	0.33±0.05	0.25±0.03*
C 18:1 9c	18.03±1.11	21.10±0.41*	18.19±1.06
C 18:1 11c	0.38±0.03	0.45±0.04	0.36±0.04
C 18:1 12c	0.31±0.03	0.35±0.03	0.42±0.02*
C 18:2 10t,12c	0.15±0.02	0.13±0.01	0.12±0.01
C 18:2 9c,12c	2.81±0.06	3.97±0.05***	2.21±0.06**
C 18:2 9c,11t	1.04±0.07	1.30±0.12*	1.21±0.08
C 19:0	0.10±0.01	0.08±0.01	0.09±0.01
C 20:0	0.11±0.01	0.10±0.01	0.12±0.01
C 18:3n3	1.66±0.06	1.72±0.14	1.45±0.07*
C 20:1n9	0.13±0.02	0.13±0.03	0.10±0.01
C 22:0	0.15±0.03	0.17±0.02	0.14±0.02
C 20:3n9	0.14±0.02	0.16±0.02	0.18±0.03
C 20:4n6	0.50±0.01	0.43±0.06	0.45±0.04
Odd-chain	3.11±0.05	2.50±0.08***	2.90±0.08*
Branched-chain	1.23±0.03	0.88±0.06***	1.19±0.03
Saturated	70.14±1.06	64.19±0.51**	70.13±1.08
Polyunsaturated	6.30±0.08	7.71±0.24**	5.62±0.17**
Total *trans* 18:1	1.63±0.07	2.79±0.07**	1.98±0.04**
Total of *cis* 18:1	19.07±1.10	22.24±0.41	19.23±1.09
Total of *cis* 18:1 without 9*cis*	1.05±0.02	1.14±0.08	1.04±0.06
20:3n9/20:4n6	0.28±0.04	0.42±0.10	0.42±0.09
18:2 9c,11t/*trans*-11 18:1	1.28±0.08	0.70±0.05**	1.12±0.07

Note: Statistical probabilities of treatment differences compared to the control group: * — *P*<0.05; ** — *P*<0.01; *** — *P*<0.001

Content of other dominant acid — palmitic (16:0) was about the same in the milk of all groups what can be explained by an efficient synthesis of palmitic acid by mammary gland of cows and therefore less influence of diet on this fatty acid presence in the milk.

Milk from cows of 1st experimental group had fewer iso-15:0, anteiso-15:0 and margaric (17:0) acids (P<0.05–0.01), which are synthesized by ruminal bacteria and enter the blood through the digestive tract. Milk of 1st experimental group cows has higher quantity of 18:1 and 18:2 acids because these fatty acids dominate in soybean lipids (P<0.01-0.001).

Supplementation of cows diet by fat with different fatty acids profile differently influenced the unsaturated fatty acids hydrogenation in the rumen and therefore on the ratio of unsaturated fatty acids isomers in milk.

Milk fat of cows of 1st experimental group contains 2.2 times more *trans*-11 18:1 (P<0.001) and 1.3 times more its metabolite — *cis*-9,*trans*-11 18:2 (P<0.05) compared to the milk fat of control group cows.

These acids performing the function of ω-6 fatty acids antagonists and are synergists of ω-3 fatty acids in animals and human tissues, so increased level of *trans*-11 18:1 and *cis*-9,*trans*-11 18:2 improves biological value of milk fat.

The level of these 18:1 and 18:2 isomers in the milk of cows of the 2nd experimental group increased also but a much lesser degree. The contents of *trans*-11 18:1 and *cis*-9,*trans*-11 18:2were 1.3 and 1.2 times higher than in control group but changes were statistically significant only for *trans*-11 18:1

(P<0.05). Another important change in the fatty acid composition of unsaturated fatty acids is a significantly increased content of linoleic (*cis*-9,12 18:2) acids in the milk of 1st experimental group cows (P<0.001) and decrease its level in the milk of 2nd experimental group cows (P<0.01). Apparently, it's due to different contents of this acid in soybean and palm oils.

The other significant change in milk fat composition was reduced total amounts of branch- and odd-chain fatty acids in cows fed soybeans (P<0.001). These acids are typical for bacteria of rumen, thus reduced its quantity indicates the suppression of ruminal microflora by polyunsaturated fatty acids of soybeans. Another reason for these effects is a reduced synthesis of fatty acids by bacteria when a large amount of dietary fat is receiving; in this case the bacteria partially used exogenous fatty acids. By this we may also explain the slightly decreased odd-chain fatty acids in milk of cows fed palm oil (P<0.05).

Milk fat of cows fed soybeans contains more polyunsaturated (P<0.01) and less saturated (P<0.01) fatty acids than milk fat of control cows. However, content of polyunsaturated acids in the milk fat of cows fed palm oil was lower than in controls group (P<0.01). This is due to differences in the fatty acid composition of the investigated fat supplements.

Both fat supplements increased the average daily milk yields (Table 2), in the 1st experimental group by 4 % (P<0.05) and in the 2nd experimental group by 7 % (P<0.05), but fat corrected milk yields were higher only in cows fed palm oil (P<0.05).

Table 2

Milk production, Σ±SEM (n=5)

Parameters	Cows groups		
	Control	1st experimental (soybeans)	2nd experimental (palm oil)
Daily milk yield, kg	24.40±0.77	25.48±0.83*	26.04±0.61*
Fat corrected milk yield (3.4%), kg	25.31±0.64	25.25±1.10	26.84±0.39*
Milk protein, %	3.30±0.05	3.33±0.05	3.28±0.03
milk fat, %	3.54±0.12	3.37±0.09*	3.51±0.07
Lactose, %	4.35±0.12	4.36±0.08	4.37±0.14
Daily protein yield, kg	0.81±0.03	0.85±0.04*	0.85±0.01*
Daily fat yield, kg	0.86±0.02	0.86±0.04	0.91±0.02*
Daily lactose yield, kg	1.06±0.05	1.11±0.02	1.14±0.04

Conclusions

Replacement of dietary soybean meal by full-fat soybeans or palm oil addition to the diet modifies cow's milk fatty acid composition. Polyunsaturated lipids of soybeans enhanced the content of healthy beneficial *trans*-11 isomers of octadecenic and octadecadienoic fatty acids in the milk fat. However, soybeans have reduced the milk fat synthesis. Palm oil is low in polyunsaturated fatty acids, so it is less affected the formation of *trans*- isomers of fatty acids and didn't inhibit the synthesis of milk fat. Both fat supplements increased milk yield of cows, but fat corrected milk yield has increased only by palm oil addition. Obtained results points out the effectiveness of the use of palm oil as a fat supplement for the diet of lactating cows.

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