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The Influence of Bioflavonoids from the Radix of *Scutellaria baicalensis* on Liver Function of Laboratory Rats Fed with Fresh and Oxidized Fats

Wpływ bioflawonoidów zawartych w korzeniu tarczycy bajkalskiej na funkcję wątroby szczurów doświadczalnych karmionych paszą zawierającą świeże i utlenione tłuszcze

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Abstract

Background. The overall daily food intake of humans always includes some oxidized fats, which form during culinary processes. It is possible to prevent the unhealthy effects of the activity of oxidation products by introducing anti-oxidants to the diet, for example bioflavonoids.
Objectives. The aim of this study was to assess the influence of an extract from the radix of *Scutellaria baicalensis* on liver function under conditions of oxidative stress caused by consumed oxidized fats.
Material and Methods. Using rats fed a diet with an 8% fat (fresh or oxidized) content and a 0.5% cholesterol supplement, we assessed the influence of bioflavonoids from the radix of *Scutellaria baicalensis* on body mass gain and the activities of alanine (ALT) and aspartate (AST) aminotransferases and alkaline phosphatase (ALP) in serum and in liver tissue extracts. The source of pro-oxidants in the rats' diet was either oxidized sunflower oil or oxidized lard. The four-week experiment involved 80 male Buffalo rats, of which 40 received a 0.075% radix extract as a supplement to their diet.
Results: In the group of rats diets containing oxidized lard, the extract had a statistically significant influence on the increase in the activity of ALP in the serum. The activity of AST increased in the rats fed a diet containing fresh lard and extract.
Conclusion: Bioflavonoids did not display a hepatoprotective activity in rats given fodder containing oxidized fats. We also found that fodder containing lard, especially oxidized lard, had a negative influence on liver function in rats, taking the form of a significant increase in the activity of ALP in the serum of those animals. In all the groups given fodder containing lard, regardless of its quality, the value of the de Ritis index indicated steatosis (**Adv Clin Exp Med 2007, 16, 1, 13–20**).

Key words: *Scutellaria baicalensis* radix, oxidized fats, rats, liver enzymes.

Streszczenie

Wprowadzenie. W całodziennych racjach pokarmowych człowieka stale są obecne tłuszcze utlenione powstające w wyniku procesów kulinarnych. Niekorzystnym skutkom działania produktów utlenienia w organizmie można zapobiegać przez wprowadzenie do diety antyoksydantów, np. bioflawonoidów.
Cel pracy. Ocena wpływu ekstraktu z korzenia tarczycy bajkalskiej na funkcje wątroby w warunkach stresu oksydacyjnego wywołanego utlenionymi tłuszczami spożywczymi.
Materiał i metody. W grupach szczurów karmionych paszami z 8% zawartością tłuszczu i 0,5% dodatkiem cholesterolu, oceniono wpływ bioflawonoidów z korzenia tarczycy bajkalskiej na: przyrosty masy ciała, hematokryt, hemoglobinę i aktywność amonotransferaz alaninowej (ALT), asparaginianowej (AST) oraz fosfatazy zasadowej (ALP) w surowicy i ekstraktach z tkanki wątrobowej. Źródłem prooksydantów w diecie były utlenione: olej słonecznikowy lub smalec. Czterotygodniowe doświadczenie przeprowadzono na 80 szczurach, samcach, rasy Buffalo, z których połowa otrzymywała 0,075% dodatek ekstraktu do paszy.

Wyniki. W grupie zwierząt na diecie z utlenionym smalcem dodatek ekstraktu powodował istotny statystycznie wzrost aktywności fosfatazy zasadowej w surowicy. Aktywność aminotransferazy asparaginianowej wzrosła w surowicy szczurów na diecie ze świeżym smalcem i ekstraktem.
Wnioski. Bioflawonoidy nie wykazywały działania hepatoprotekcyjnego u zwierząt karmionych paszą z utlenionymi tłuszczami. Obserwowano negatywny wpływ na funkcję wątroby paszy zawierającej smalec, zwłaszcza utleniony, co miało swój wyraz w znaczącym wzroście aktywności fosfatazy zasadowej w surowicy badanych zwierząt. We wszystkich grupach zwierząt karmionych paszą ze smalcem, niezależnie od jego jakości, wartość wskaźnika de Ritisa wskazywała na stłuszczenie wątroby (**Adv Clin Exp Med 2007, 16, 1, 13–20**).

Słowa kluczowe: korzeń tarczycy bajkalskiej, utlenione tłuszcze, szczury, enzymy wątrobowe.

The products of fatty acid oxidation, such as peroxides and aldehydes, which form during culinary processes such as frying, roasting, and boiling, interfere with the metabolism of food components and with the function and structure of many organs, leading to various pathological conditions [1–3]. It is possible to prevent the unhealthy effects of the activity of oxidation products by introducing anti-oxidants to the diet, for example bioflavonoids. Large quantities of bioflavonoids are found in dried culinary herbs or in medicinal herbs such as *Scutellaria baicalensis* [4]. The most important biologically active chemical components of *Scutellaria baicalensis* are lipophilic flavones, such as baicalein, baicalin, and vogonin, and their glucosides. The flavonoid content of the material is very high, > 20%, of which 12–17% is baicalin and 3–4% is vogonin. Baicalin itself is poorly absorbed from the gut, but is hydrolized to baicalein by intestinal bacteria and then restored to its original form from the absorbed baicalein in the body. The bioavailability of orally administered baicalin was calculated to be 62% [5].

Existing information on the influence of bioflavonoids on liver function is not unequivocal. It is also unclear how oxidized lipids in the diet influence liver function or animal development. In the literature there are many reports on the hepatoprotective activity of flavonoids and of *Scutellaria baicalensis* itself [2, 5–9]. However, no precise investigations have been performed on its activity in protecting the liver from oxidative stress caused by consumed fats oxidized during culinary processes.

Material and Methods

The Experiment

The study was carried out on 80 male Buffalo rats, with an initial body mass of 183.6 ± 21.1 g and a final body mass of 273.5 ± 25.9 g. During the period before the experiment, all the rats received standard LSM fodder. For the four weeks of the experiment, the rats were kept in appropriate conditions (room temperature, 12 hour light-dark rhythm).

The animals were divided into eight groups of 10 animals. The plan of the experiment is given in Table 1. Half of the rats (40 rats) were given a diet with an 8% sunflower oil content, while the second half (40 rats) were given a diet with an 8% pork lard content. Within these two supergroups receiving sunflower oil or lard, half (20 rats) had a diet with oxidized sunflower oil or lard and the other half fresh. Half of the rats eating a diet with fresh fat and half of the rats eating a diet with oxidized fat also had an extract from the radix of *Scutellaria baicalensis.* The supplement came to 0.5 g per kilo of diet, with the extract having an 80% baicaline content. Ready-made extract "Oxyd" was obtained from Wroclaw's Herbalist Factory "Herbapol". The method of its preparation is copyrighted and patented. Details concerning the experiment are described in a previous study [10].

The rats had unlimited access to fodder and water. Their consumption of fodder and water was checked every two days and their body mass measured once per week. The fodder was prepared according to the method described in [11]. The oxidation conditions were established based on the study by Ziemlański et al. [12]. The fatty acid component and the content of primary and secondary products of oxidation in the fresh and oxidized fats were measured as the peroxide and anisidine values. The composition and proportion of the total fat content of the polar fraction was also determined. These results are given in Table 2.

Preparation of the Biological Material for Analysis

After completion of the feeding stage of the experiment, the rats underwent light ether anesthesia and blood was taken directly from their hearts into test tubes containing heparin. After the rats had been sacrificed, their livers were prepared. The livers were rinsed in a physiological salt solution, blotted dry, and weighed. Liver samples were taken for histopathological analysis and the results of the examination are described elsewhere [10]. Institutional approval for the described animal experiment was obtained.

Table 1. The plan of the experiment

Tabela 1. Schemat doświadczenia

Group 1 Control (kontrola) Fodder containing 8% sunflower oil (Pasza z 8% zawartością oleju słonecznikowego)	Group 3 Fodder containing 8% sunflower oil and the extract* (Pasza z 8% zawartością oleju słonecznikowego i z ekstraktem)	Group 5 Fodder containing 8% oxidized sunflower oil (Pasza z 8% zawartością utlenionego oleju słonecznikowego)	Group 7 Fodder containing 8% oxidized sunflower oil and the extract* (Pasza z 8% zawartością utlenionego oleju słonecznikowego i z ekstraktem)
Group 2 Control (kontrola) Fodder containing 8% lard (Pasza z 8% zawartościa smalcu)	Group 4 Fodder containing 8% lard and the extract* (Pasza z 8% zawartościa smalcu i ekstraktem)	Group 6 Fodder containing 8% oxidized lard (Pasza z 8% zawartością utlenionego smalcu)	Group 8 Fodder containing 8% oxidized lard and the extract* (Pasza z 8% zawartościa utlenionego smalcu i ekstraktem)

* Extract from the radix of *Scutellaria baicalensis.* The supplement came to 0.75 g per kilo of fodder, with the extract having an 80% baicaline content. Ready-made extract "Oxyd" was obtained from Wroclaw's Herbalist Factory "Herbapol". The method of its preparation is copyrighted and patented.

* Ekstrakt z korzenia tarczycy bajkalskiej. Ekstrakt dodawany do 1 kg paszy w ilości 0,75 g zawierał 80% bajkaliny. Preparat farmaceutyczny pochodził Wrocławskich Zakładów Zielarskich „Herbapol" i sposób jego otrzymywania jest zastrzeżony patentem.

Table 2. Fatty acid composition, expressed as a percentage of the total fatty acid content, and the quality indicators for the fresh and oxidized fats added to the rats' fodder

Tabela 2. Skład kwasów tłuszczowych, jako % sumy wszystkich kwasów, i wskaźniki jakości świeżych i utlenionych tłuszczów dodawanych do paszy szczurów

Analyzed indicator (Badane wskaźniki)	Fresh lard (Świeży smalec)	Oxidized lard (Utleniony smalec)	Fresh sunflower oil (Świeży olej słonecznikowy)	Oxidized sunflower oil (Utleniony olej słonecznikowy)
Peroxide value (Liczba nadtlenkowa) [mg O_2/100 g fat]	0.1	6.94	0.13	22.7
Anisidine value (Liczba anizydynowa)	0.7	46.0	4.8	71.8
Polar fraction [%]* (Frakcja polarna)	1.3	23.9	2.1	42.3
Fatty acid type [sum total]:				
Saturated acids (Nasycone kwasy tłuszczowe)	43.3	43.6	8.6	24.5
Monounsaturated acids (Jednonienasycone kwasy tłuszczowe)	45.1	45.5	22.3	30.6
Polyunsaturated acids (Wielonienasycone kwasy tłuszczowe)	10.7	9.8	68.2	42.9
Unidentified acids (Niezidentyfikowane kwasy tłuszczowe)	0.9	1.1	0.9	2.0

* Proportion of the total fat content of the polar fraction.

* Udział % frakcji polarnej w masie tłuszczu.

Parameters Determined for the Biological Material

The following parameters were determined from analyses of the biological material: the activities of alanine and aspartate aminotransferase via kinetic methods using the Biosystems enzymatic diagnostic tests ALT (cat. No. 11533), AST (cat. No. 11531) and alkaline phosphatase via kinetic methods using the Biochemtest enzymatic diagnostic tests and ALP (cat. No. 178152149).

Statistical Assessment of the Results

The normality of the distribution in the examined groups was assessed with the Shapiro-Wilk's W test. In the case of a lack of a normal dis-

tribution, logarithmic transformation was applied. The significance of the differences between the average values for the examined groups was assessed using individual analysis variance ($p < 0.05$). Levene's test was used to check individual variance. The assessment of the influence of two independent variables on one dependent variable was done with the ANOVA two-factor analysis of variance. All the statistical calculations were done with the STATISTICA 6.0 PL program of StatSoft. Inc. USA.

Results

The Influence of the Type of Diet on Liver Function

The rats' liver function was assessed by determining the activities of alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP) in the their serum. The activity of ALT in the serum was higher (63 U/l) in the rats receiving oil in their diets than in those receiving lard (37.9 U/l). The type of fat added to the fodder did not influence the AST or ALP activities in the serum of the rats.

The influence of oxidized fats (sunflower oil and lard) on liver function was assessed by comparing the activities of selected enzymes from the serum of rats from groups 5 and 6 with rats from the appropriate controls, i.e. groups 1 and 2. An unhealthy effect of the oxidized fat on liver function was only observed in the groups fed with lard (group 6 relative to group 2), as seen in the statistically significant increase in the activity of ALP by 33.7% in the serum of the rats receiving fodder containing oxidized lard relative over that in the rats receiving fodder containing fresh lard. In the groups of rats receiving fodder containing sunflower oil, there was no observable influence of the quality of the fats on liver function.

By comparing the activity of the liver enzymes in the serum of rats from groups 1 and 3 and 2 and 4, the effect of the bioflavonoids from the radix of *Scutellaria baicalensis* on the function of the liver in rats receiving fodder containing fresh fats was assessed. In the serum of rats from group 4, an increase in AST activity by 24.3 U/l was observed compared with group 2; thus the bioflavonoids acted in a manner unhealthy for the organism. In group 3, no changes in the activity of the studied enzymes were found relative to group 1; thus the influence of the bioflavonoids was fat-type dependent.

No differences were observed between the activities of the aminotransferases in the groups on diets containing oxidized fats (groups 5 and 6) and the activities in the groups on diets containing oxidized fats and the extract from the radix of *Scutellaria baicalensis* (groups 7 and 8). The bioflavonoids in question did not display any hepatoprotective activity in this experiment. In the group receiving fodder with both oxidized lard and bioflavonoids, there was a statistically significant increase of 120.1% in the activity of alkaline phosphatase in the serum**.** Such an increase in ALP activity in the serum of the rats that received the extract in their diet may have been due to the increased level of bile secretion under the influence of bioflavonoids.

The hepatoprotective activity of bioflavonoids can also be assessed by comparing groups 7 and 8 with 3 and 4, respectively. In the serum of the rats in group 7, a statistically significant increase in ALT activity was found relative to the activity in rats in group 3. This unhealthy activity level seems to be due to the increased level of bile secretion under the influence of bioflavonoids, which abets the digestion of oxidized oil and absorption of the toxic products of oxidation. The significant increase in ALP activity in the serum of rats in groups 7 and 8 relative to that in rats in groups 3 and 4, respectively, confirms the observation regarding bile secretion under the influence of bioflavonoids, particularly strong in group 8. It is possible that bioflavonoids enhance the unhealthy effects of oxidized fats.

Discussion

The Influence of the Type of Fat (Oil, Lard) on the Function of the Liver

ALT is an index enzyme for liver damage, and it is specific to damage to that organ. AST occurs outside the liver, in organs such as the skeletal muscles, heart, kidneys, pancreas, spleen, and lungs, so an increase in AST activity in the serum is not necessarily due to damage to the hepatocytes. The de Ritis index has a diagnostic value: it is a measure of the AST/ALT activity quotient. In humans, a value of less than 0.9 indicates an acute inflammation of the liver, and a value of above 2 suggests hepatic steatosis. Alkaline phosphatase is an excretory enzyme. An increase in its activity in the serum suggests cholestasis. ALT activity was higher in the serum of the rats receiving fodder containing oil (63 U/l) than in the rats on a diet with lard (37.9 U/l). Similar results were obtained by Kawahara et al. [13].

Table 3. The increase in body mass [g] of rats and the influence of the applied diets on the activity of selected enzymes in the serum of the rats and in liver samples taken from the rats [x ± *SD*; average ± standard deviation]

Tabela 3. Przyrost masy ciała [g] i wpływ stosowanych diet na aktywność wybranych enzymów wątrobowych w surowicy i ekstraktach z wątrób szczurów doświadczalnych [x ± SD; średnia ± odchylenie standardowe]

Variable (zmienna) x ± SD	Sunflower oil – control (Olej słonecznikowy – kontrola)	Lard – control (Smalec – kontrola)	Sunflower oil + extract (Olej słonecznikowy + ekstrakt)	Lard + extract (Smalec + ekstrakt)	Oxidized sunflower oil (Utleniony olej słonecznikowy)	Oxidized lard (Utleniony smalec)	Oxidized sunflower oil + extract (Utleniony olej słonecznikowy + ekstrakt)	Oxidized lard + extract (Utleniony smalec + ekstrakt)
	group 1 (grupa 1)	group 2 (grupa 2)	group 3 (grupa 3)	group 4 (grupa 4)	group 5 (grupa 5)	group 6 (grupa 6)	group 7 (grupa 7)	group 8 (grupa 8)
Body weight gain (Przyrost m.c.)	80.5 ± 18.0[e]	107 ± 10.9[e]	102.5 ± 9.8[d]	103 ± 20.3	64.5 ± 20.2	98 ± 13.8	77 ± 17.0[d]	87 ± 16.2
Serum:								
AST [U/l];	92.1 ± 14.6	109.6 ± 19.2[a]	94.9 ± 22.1	133.9 ± 15.2[a]	87.7 ± 12.9	119.7 ± 13.3	86.6 ± 10.4	123.2 ± 22.2
ALT [U/l];	63 ± 15.4[e]	37.9 ± 4.6[e]	46.7 ± 14.5[d]	43.7 ± 7.3	68.2 ± 17	47.1 ± 11.1	75.4 ± 16.4[d]	48.1 ± 26.6
de Ritis Index;	1.5 ± 0.4[e]	2.9 ± 0.6[e]	2.1 ± 0.5[d]	3.1 ± 0.6	1.4 ± 0.4	2.6 ± 0.5	1.2 ± 0.3[d]	3.0 ± 1.0
ALP [U/l];	64.8 ± 7.5	68.5 ± 3.9[b]	55.4 ± 7.3[d]	58.6 ± 5.7[d]	74.5± 15.3	91.6 ± 12.9[b, c]	85.5 ± 11.8[d]	202 ± 34.8[c, d]
Liver:								
AST [U/g liver]	26.6 ± 4.5[a, e]	39.6 ± 12.9[b, e]	42.8 ± 4.9[a, d]	48.2 ± 7.3[d]	24.5 ± 4.5[c]	44.1 ± 16.2[b]	19.0 ± 4.0[c, d]	39.1 ± 8.0[d]
ALT [U/g liver]	28.4 ± 5.4[a, e]	41.5 ± 14.4[e]	3 9.3 ± 4.7[a]	50.0 ± 8.2[d]	28.5 ± 5.2	44.4 ± 17.6	30.4 ± 8.7	39.6 ± 10.5[d]
ALP [U/g liver]	0.15 ± 0.02[e]	0.18 ± 0.02[b, e]	0.17 ± 0.04	0.19 ± 0.02	0.14 ± 0.02[c]	0.25 ± 0.03[b, c]	0.18 ± 0.03[c]	0.20 ± 0.02[c]

a – statistically significant difference between groups 1 and 3 and between 2 and 4.
b – statistically significant difference between groups 1 and 5 and between 2 and 6.
c – statistically significant difference between groups 5 and 7 and between 6 and 8.
d – statistically significant difference between groups 3 and 7 and between 4 and 8.
e – statistically significant difference between groups 1 and 2.
The results of *t*-test for independent samples were considered significant at $p < 0.05$.

a – różnice istotne statystycznie między grupami 1 i 3 oraz 2 i 4.
b – różnice istotne statystycznie między grupami 1 i 5 oraz 2 i 6.
c – różnice istotne statystycznie między grupami 5 i 7 oraz 6 i 8.
d – różnice istotne statystycznie między grupami 3 i 7, oraz 4 i 8.
e – różnice istotne statystycznie między grupami 1 i 2).
Poziom istotności statystycznej ustalono przy $p < 0,05$.
m.c. – masa ciała

The influence of an extract from the radix of *Scutellaria baicalensis* in the diet on the function of the liver was assessed on the basis of measurements of the activities of ALT, AST, and ALP in the serum of the rats**.** ALT and AST aminotransferases are indicator enzymes. They enter the serum not only in the case of damage to the liver, but also when the permeability of the cell membrane increases. The permeability of the cell membrane increases with increasing unsaturated fatty acid content in the diet. ALT, an enzyme occurring in the cytoplasm, moves more easily from the cell interiors to the serum than AST, which is mainly located in the mitochondria. The value of the de Ritis index for the group receiving fresh lard in their fodder (group 2) indicated that fat was accumulating in the livers of those rats.

The Influence of the Quality of the Fat (Fresh or Oxidized) on the Function of the Liver

The unhealthy effect of oxidized lard was probably due to the large amount of readily absorbed secondary products of oxidation, i.e. aldehydes and ketones from the polar fraction, and their toxic activity on the liver. The polar fraction makes up almost a quarter of the mass of the oxidized lard added to the fodder, while the aldehyde and ketone contents were 65-fold higher than in the fodder containing fresh lard (Table 2). The increase in ALP activity in the serum and the value of the de Ritis value seem to indicate that this group of rats underwent cholestasis.

In the groups of rats receiving oxidized sunflower oil in their fodder, no influence of the quality of the fat on the function of the liver was observed. It can be assumed that the polymers and toxic components of the polar fraction formed in the oxidation process were not absorbed from the digestive systems of the rats. The polar fraction was almost half of the mass of the oxidized oil added to the fodder, but the secondary product content only rose 15-fold relative to the fresh oil (Table 2). This is possibly why the quality of the oil did not seem to influence the function of the rats' livers so clearly.

Hayam et al. [2], after seven weeks of feeding rats a diet containing fresh or oxidized soybean oil, obtained different results. They found a significant increase in the activity of the aminotransferases in the serum of the rats fed a diet with oxidized oil, which indicated hepatocyte damage. These differences between their results and those of the present study could be due to the period of the study as well as a different content of the polar fraction and its toxic components added to the fodder in fat.

The Influence of Bioflavonoids on Liver Function

Juśkiewicz et al. [14] studied the influence of an extract from grapefruit on the function of the liver of rats. They saw a fall in ALT activity in the groups of rats that received flavonoids in their diet relative to the activity for the control group. They also found an increase in ALP activity in the serum of the rats that had received flavonoids in their diet, and the activity of the enzyme increased together with the dose of the extract. This indicates bile secretion under the influence of bioflavonoids. In the study of Zduńczyk et al. [15], rats were given a high-fat fodder, with a mixture of soybean oil and beef fat as the fat source, with or without an added extract from *Scutellaria baicalensis*. In the group receiving bioflavonoids in their diet, there was an increase in aminotransferase activity in the serum relative to the control group, which was also found in the present experiments for the group given fodder with lard. This confirms the observation of the present study on the influence of bioflavonoids being dependent on the type of fat in the diet.

There are conflicting results of earlier studies on the hepatoprotective activity of bioflavonoids in animals subjected to the activity of various compounds that damage the liver. Wada et al. [16, 17], Niedworok et al. [18], and Jian Zhang et al. [19] found that there was a reduction in the activity of toxic chemical compounds such as D-galactosamines, carbon tetrachloride, lead ions, and bromobenzene. Juśkiewicz et al. [14] found that grapefruit flavonoids had a protective activity in rats given fodder containing oxidized fats. Madej [20] did not find any protective activity of an extract from the radix of *Scutellaria baicalensis* in rats injected with lead and cadmium. In most of the studies in which flavonoids were found to have a hepatoprotective activity, the dose of flavonoids was higher than in this study.

A Comparison of the Activity of Aminotransferases in the Serum and the Liver of the Studied Rats

A dependency was found between the activities of the enzymes in the serum and their activities in liver extracts. In the groups of rats given fodder with sunflower oil (groups 1, 3, 5, 7), the increase in ALT activity was related to a fall in its

activity in the liver per gram of liver tissue. It can be assumed that this was caused by an increase in the permeability of the cell membranes due to the incorporation of unsaturated fatty acids into them and the consequent escape of the enzyme from the cytoplasm into the serum. In the groups given fodder with fresh lard (groups 2 and 4), the increase in ALT activity in the serum was concurrent with an increase in its activity in the liver. It can be assumed that the components of the diet did not influence the function of the liver in these groups. In the groups given fodder with oxidized lard (groups 6 and 8), there was an observed increase in the enzyme's activity in the serum and a decrease in its activity in the liver. Presumably, the oxidized lard significantly influenced the metabolic reactions in the livers of those rats.

The high concurrence between the changes in AST activity in the liver and serum of rats given fodder containing oil is noteworthy. The increase or decrease in the AST activity in the liver was commensurate with the changes in activity in the serum. It seems that there was no damage to the cellular structures (mitochondria) with which this enzyme is associated in the liver. Deep damage to the hepatocytes, for example during acute necrosis of the liver, is associated with an increase in AST activity in the serum and a parallel reduction of its activity in the liver. There was a similar dependency seen in three of the groups given fodder containing lard. Only in group 8 was the increase in AST activity in the serum related to a decrease in its activity in the liver. However, the changes in AST activity in the serum and the liver in this group of rats were not statistically significant.

A Discussion of the Changes in the Activity of Alkaline Phosphatase (ALP) in the Livers of the Rats (Table 3)

Due to the scarcity of reports in the literature on the activity of alkaline phosphatase in the liver, the interpretation of the obtained results was very difficult. The activity of ALP in the livers of the rats depended on the type of fat in the diet, and was higher for group 2, where the fodder contained lard, than for group 1, where the fodder contained sunflower oil. The activity of alkaline phosphatase in the liver depended on the quality of the fat only in those groups given fodder containing lard without the bioflavonoids (groups 2 and 6), and it was higher in the group given fodder with oxidized lard. Within the groups given fodder containing oxidized fats, ALP activity also depended on the presence or absence of bioflavonoids in the fodder. This bioflavonoid influence had a trend dependent on the type of fat, with group 7 showing an increase and group 8 showing a decline in ALP activity in the liver.

Under the experimental conditions, bioflavonoids from the radix of *Scutellaria baicalensis* did not have a hepatoprotective effect in the rats given fodder containing oxidized fats. Adding this type of bioflavonoid to the fodder led to the unhealthy effects of consumption of oxidized fats that did not appear in the rats fed fodder containing oxidized fats but no bioflavonoids. Specifically, in the rats with fodder containing bioflavonoids and oxidized fats, the development and the function of the liver was negatively affected, along with a significantly smaller increase in body mass.

The unhealthy effect of the applied diets on the function of the liver was manifested in the significant increase in the activity of alkaline phosphatase in the serum of the rats given fodder containing oxidized fats, especially in the group given oxidized lard and bioflavonoids. The bioflavonoids probably increased the secretion of bile from the livers of the rats, which made it easier for the rats to digest oxidized fats and absorb the toxic products of oxidation from the polar fraction.

An unhealthy influence of the fodder containing lard on the functions of the rats' livers was also found. In all the groups given fodder containing lard, regardless of its quality, the value of the de Ritis index was indicative of steatosis.

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