The effects of apricot on serum proteins and liver enzymes in rats

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Summary
This study aimed to investigate the effects of different rates and feeding periods of sun-dried organic apricot (SDOA) supplementation on serum proteins and liver enzymes in rats. Numbers of 120 male and 120 female rats were randomly divided into five groups. The control group was fed with normal rat chow, and the others with 1%, 2.5%, 5% and 10% SDOA-supplemented diet, respectively. At the end of the 30th day of feeding periods, blood samples of 8 rats from each gender of every group were taken. Serum samples were used for measurements of albumin (ALB), total protein (TP), alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) levels. This procedure was repeated on the 60th and 120th days. Effects of rates and periods on parameters and interactions were investigated by two-way ANOVA. As the rates of SDOA in diet increased, decreases were observed in all parameters of males, and in ALP, AST and TP parameters of females. Considering periods, an effective role was observed on ALB, ALT and TP levels in genders. However, there were no significant interactions between rates and periods. The rate of 1% had beneficial effect on parameters in genders. However, the optimal period was not determined.

Keywords
sun-dried organic apricot; serum proteins; liver enzyme; rat

Consumption of fruits and vegetables is widely recommended by nutrition experts because of their preventive and/or protective effects on several chronic diseases such as obesity, coronary heart disease, stroke, diabetes, hypertension and some cancers [1–3]. The World Health Organization has recommended that people eat at least five portions (about 400 g) of fruits and vegetables per day to prevent cancer and other chronic diseases [4]. Some fruits including apricot, banana, grape, nectarine and peach, meet the daily requirements of vitamins and minerals for humans [4–7]. In this context, it has been reported that consumption of 3–4 fresh apricots per day for adults may provide daily requirements of some essential elements, in particular K, Se, Mg and half of required P and Zn [8, 9]. Regarding apricot consumption, nutritional and mineral contents of sun-dried organic apricot (SDOA) were briefly reported by Yilmaz et al. [10].

In clinical practice, serum alanine aminotransferase (ALT), aspartate aminotransferase (AST) and alkaline phosphatase (ALP) enzyme activities have been considered to be important indicators of liver functions [11]. Fluctuations of ALT, AST and ALP levels are mainly the result of the leakage of these enzymes from the cytosol of hepatocytes into the blood stream. While an increase in these enzyme activities in serum may indicate serious liver diseases, decreased serum albumin (ALB) and total protein (TP) levels are also used as indicators of functions in that they can reflect reduced synthesis and increased protein degradation in liver diseases [12]. Therefore, for understanding alterations of liver functions, knowledge of the pathophysiology of liver enzymes is an essential guide [13].

Literature reveals no information about consumption of SDOA and its effects on serum liver enzyme levels. In order to address these topics, this study was performed on four different rates of SDOA, over three time periods. This study may open a new perspective on the investigation of nutritional values of SDOA, based on serum ALB, TP and liver enzyme levels in rats.

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MATERIALS AND METHODS

The study protocol was approved by the Ethic Comittee, Faculty of Medicine, University of Inonu, Malatya, Turkey. Rats were provided by the Experimental Animal Research and Production Center of Inonu University. During a 120-days study period, female rats were housed seperately from male rats. They had free access to chow and water during study periods, and no toxic effects, side effects or death were observed.

Animals

Numbers of 120 Sprague Dawley rats of each sex were used. They were randomly divided into five groups, leading to 24 rats in each group of both genders. The control group (group-1) was fed with standard rat chow and the others were fed with 1%, 2.5%, 5% and 10% SDOA-supplemented diet. They were housed at room temperature (21 °C ± 2 °C) with relative humidity of 53% ± 3% with a 12 h light/dark cycle. At the beginning of the study, the average body weight of male and female rats were 321 g ± 24.6 g and 210 g ± 21.4 g, respectively. On the 30th day, eight rats from each groups of both gender (totally 40 female and 40 male) were anaesthetized by inhalation of diethylether and 7–10 ml of blood samples were taken by intracardiac puncture. Blood samples were centrifuged at 3000 ×*g* for 10 min and the extracted serum samples were stored at –20 °C until analysis of AST, ALT, ALP, ALB and TP. On the 60th and 120th day, this procedure was repeated.

Diet

The normal rat chow was purchased from Korkutelim, Antalya, Turkey. The Kabaaşi variety of SDOA, which was provided from a local market in Malatya, Turkey (having organic certificate), was used as a supplementary diet. Nutrient and mineral composition of SDOA and the daily average food consumption of the male and female rats were determined as described by Yilmaz et al. [10]. The SDOA-supplemented diet was freshly prepared in 10 kg batches manually.

Biochemical parameters

Hepatic enzyme concentrations were measured by Abbott clinical autoanalyser (Abbott Diagnostics, Abbott Park, Illinois, USA) using ion-selective electrode method (Architect c16000).

Statistics

All statistical analyses were performed using MedCalc version 11.4.4 (MedCalc Software, Mariakerke, Belgium). Data were expressed as mean ± standard deviation. Kolmogorov-Smirnov test was used to determine normal distribution. Multiple comparisons were carried out by One-way ANOVA or two-way ANOVA tests followed by Bonferroni post-hoc analysis. Additionally, Student's t-test was used to determine the differences between the means of gender groups. We considered statistical significance at $p < 0.05$.

RESULTS

The results of serum parameters are shown in Tab. 1–3. Serum TP levels in both sexes and ALB levels only in males were decreased by the supplementation rates ($p < 0.001$; Tab. 1). When feeding periods were taken into consideration in both genders, a decrease in serum ALB levels and different fluctuations of TP levels were identified ($p < 0.001$). While the highest TP levels were determined on the 60th day in both sexes, the lowest TP levels were determined on the 30th day in females and on 120th day in males ($p < 0.05$; Tab. 2). It was determined that serum ALT, AST

Tab. 1. Effects of different rates of SDOA supplementation on serum proteins and liver enzymes in rats.

Parameter	Control		1%		2.5%		5%		10%	
	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male
ALB [g·dl^{-1}]	3.4 ± 0.5	3.0 ± 0.5	3.4 ± 0.5	3.1 ± 0.4	3.5 ± 0.6	2.7 ± 0.6	3.3 ± 0.3	2.9 ± 0.5	3.4 ± 0.4	2.7 ± 0.5^{b}
ALP [U·l^{-1}]	162 ± 43	209 ± 60	165 ± 39	215 ± 50	161 ± 62	171 ± 54^{ab}	149 ± 38	181 ± 50	125 ± 33^{abc}	144 ± 43^{abd}
ALT [U·l^{-1}]	49 ± 8	57 ± 10	50 ± 11	56 ± 10	49 ± 12	49 ± 9^{a}	47 ± 11	54 ± 11	46 ± 12	47 ± 10^{abd}
AST [U·l^{-1}]	123 ± 22	123 ± 19	116 ± 23	118 ± 20	114 ± 18	105 ± 16^{a}	108 ± 19	115 ± 25	104 ± 24^{a}	107 ± 22
TP [g·dl^{-1}]	7.1 ± 0.3	6.7 ± 0.3	7.2 ± 0.3	6.8 ± 0.3	7.1 ± 0.3	6.6 ± 0.4	7.0 ± 0.2^{b}	6.6 ± 0.3	7.0 ± 0.2^{b}	6.6 ± 0.3

Each value is expressed as mean ± standard deviation ($n = 24$), differences at $p < 0.05$ were considered significant.
a – different than the control group; b – different than the 1% group; c – different than the 2.5% group; d – different than the 5% group.

Tab. 2. Effects of different periods of SDOA supplementation on serum proteins and liver enzymes in rats.

Parameter	30 days		60 days		120 days	
	Female	Male	Female	Male	Female	Male
ALB [g·dl^{-1}]	3.7 ± 0.4	3.2 ± 0.3	3.5 ± 0.4^{ac}	3.0 ± 0.4^{ac}	3.0 ± 0.3^{ab}	2.3 ± 0.4^{ab}
ALP [U·l^{-1}]	140 ± 38	190 ± 61	156 ± 48	192 ± 58	160 ± 50	169 ± 51
ALT [U·l^{-1}]	46 ± 9	58 ± 11	45 ± 10	52 ± 9^{a}	53 ± 11^{ab}	48 ± 9^{a}
AST [U·l^{-1}]	108 ± 21	110 ± 21	114 ± 24	116 ± 22	118 ± 19	114 ± 22
TP [g·dl^{-1}]	7.0 ± 0.3	6.7 ± 0.2	7.1 ± 0.3	6.9 ± 0.3^{ac}	7.1 ± 0.2	6.4 ± 0.3^{ab}

Each value is expressed as mean ± standard deviation (n = 40), differences at $p < 0.05$ were considered significant.
a – different than 30 days period; b – different than 60 days period; c – different than 120 days period.

Tab. 3. Differentiation of parameters in rats with regard to sex.

Parameter	Female		Male		p
	n	Mean ± SD	n	Mean ± SD	
ALB [g·dl^{-1}]	119	3.4 ± 0.5	120	2.9 ± 0.5	< 0.001
ALP [U·l^{-1}]	115	152 ± 46	119	184 ± 57	< 0.001
ALT [U·l^{-1}]	117	48 ± 11	115	53 ± 11	0.001
AST [U·l^{-1}]	115	113 ± 22	116	114 ± 21	0.818
TP [g·dl^{-1}]	117	7.1 ± 0.3	117	6.7 ± 0.3	< 0.001

The numbers of rats cover all subgroups of the experiment. Comparisons were carried out by a non-paired Student's t-test.
n – number of samples, SD – standard deviation, p – statistical significance.

and ALP activities decreased as supplementation rate increased ($p < 0.005$; Tab. 1). The duration of feeding did not have any effect on enzyme activities in any sex (Tab. 2). In addition, there were no statistically significant interactions between supplementation rates and feeding periods for all parameters (Tab. 3). Regarding the effects of gender differences on test parameters, it was determined that serum ALB and TP levels of female rats were higher than those of male rats ($p < 0.001$). Serum enzyme levels of females were lower than those of males ($p < 0.05$; Tab. 3). The effects of different SDOA supplementation rates and feeding periods on serum proteins and liver enzyme activities in rats are shown as two-way ANOVA presentations in Fig. 1.

DISCUSSION

To our best knowledge, there are no scientific studies reporting on the effect of SDOA consumption on the levels of serum ALB, TP, ALT, AST and ALP in rats. This study provides intitial insights into the appropriate feeding rates and time periods that may lead to a more precise determination of ideal feeding parameters.

Hepatocellular damage due to various reasons causes increase in plasma levels of several enzymes, including ALT and AST. Normalization of these parameters is accepted as an indicator of the improvement of liver functions [13, 14]. Das et al. and Bilal et al. reported that different *Eugenia jambolana* extracts caused a decrease in serum ALP, ALT and AST activities [15, 16]. Similarly in our study, the increase in SDOA supplementation rates caused a decrease of serum ALT and AST levels. These changes may be interpreted as a benefical effect of SDOA consumption. ALP, which hydrolyses phosphate esters at alkaline pH, is present particularly in the liver, but also in other tissues such as intestines, uterus, bile ducts and bones. Clinically, a major portion of serum ALP (about 80%) originates from hepatobiliary and/or bone tissues [17, 18]. Mucous cells in bile ducts of the liver and osteoblastic cells in bone tissue have high levels of ALP activity. Therefore, serum ALP levels are used as an indicator of either liver function or bone metabolism [19]. Osteoblastic activity is at its minimum levels in adults, so the contribution of bone isoenzyme to total ALP activity during this period is minimal [13–18].

In the present study, because of completed bone development of the adult rats used, any increase in age-related levels of serum ALP was not expected. The decrease in the serum ALP levels

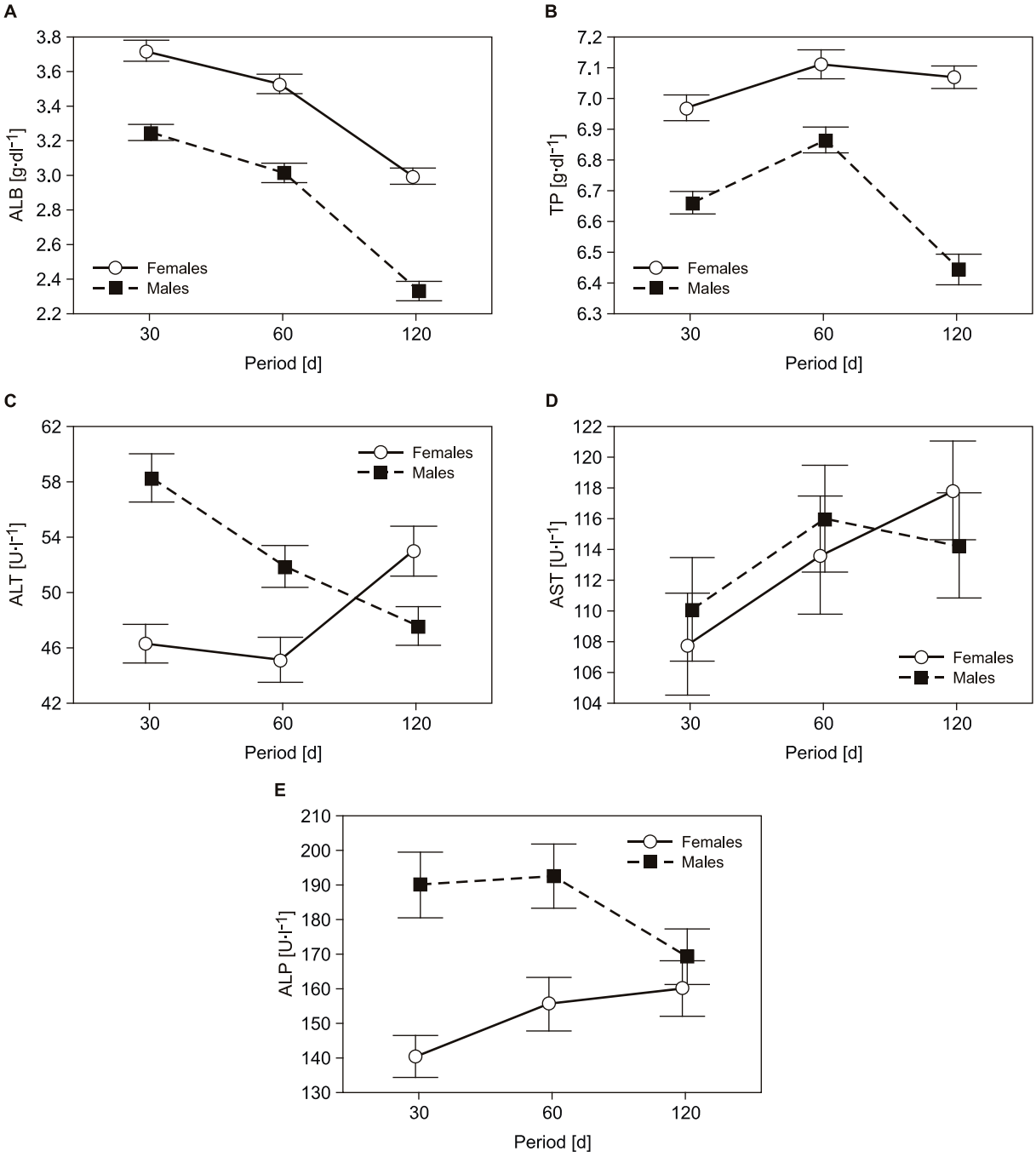

Fig. 1. Effects of sex and feeding periods on serum proteins and liver enzymes in rats.

A, B, C, D and E indicate serum ALB, TP, ALT, AST and ALP differentiation, respectivelly.

was affected only by increasing rates of SDOA supplementation, just as ALT and AST levels were, indicating an increase in healthy liver function. In healthy individuals who have completed biological development and do not have any kidney or gastrointestinal disease, any decrease of serum ALB and TP levels may indicate insufficient hepatocyte biosynthesis [19]. One of the major reasons for a decline in biosynthetic functions is feeding with a low protein diet [19]. Hence, low protein content of SDOA may be the reason of decrease in serum protein levels. In addition, rats have a strong immune system that causes an increase in the synthesis of immune globulins at the expense of decreased serum ALB levels [12]. This hypothesis is also a possible explanation for the

weaker reduction in serum TP levels compared to the serum ALB levels in our study [12]. In the present study, it was determined that the serum ALB levels of male rats were more affected than females' by the increase of SDOA supplementation rates. This can be attributed to the combination of the low protein content of SDOA and with the fact that male rats have higher energy requirements than females [20]. Despite being in the age of fertility, the females were not yet nulliparous. A rapid decrease in ALB levels in male rats was also suggested by MAC QUEEN and colleagues, who determined an age-related decrease in serum ALB and TP levels of rats, which is in concordance with our results [21]. In this regard, lower serum ALB and TP levels in males than in females may be acceptable physiologically [22]. On the other hand, in the case of enzymes, SDOA led to a more pronounced increase of liver functions in females than in males. This is supported by the fact that the enzyme levels in female rats were more affected by the increase of SDOA supplementation rates than males.

Consequently, 1% SDOA supplementation can be considered as the most suitable rate due to the combination of a positive effect on enzyme levels and a moderately decreasing effect on serum TP levels. However, taking into account the continuity of nutrition, balanced nutrition criteria, metabolism and natural eating habits of rats, although a 30-day duration appears as the most appropriate feeding period, our data do not facilitate to conclude that this is the ideal SDOA feeding period.

Acknowledgements

This study was supported by Inonu University Scientific Research Fund (Project number: 2010/74). We would like to thank the Inonu University Scientific Research Projects Unit for the financial support.

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Received 25 March 2013; revised 10 April 2013; accepted 11 April 2013.