



## The Effect of High Amounts of Wheat Gluten Meal and Corn Gluten Meal Added to the Diets on Some Serum Parameters in Rats

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ARTICLE INFO	ABSTRACT
<p><i>Research Article</i></p> <p>Received : 27/11/2019 Accepted : 13/12/2019</p> <p><b>Keywords:</b> Biochemistry Enzyme Gluten Rat Serum</p>	<p>The aim of this study was to determine the effects on some serum parameters of wheat and corn gluten diet, which is also a high protein source. A total of 24 male rats aged 20 days and divided into 3 groups, each containing 8 rats were used in the study. The majority of the protein content in the diet of soybean meal as a Control group; in the diet of wheat gluten as a Wheat group; in the diet of corn gluten as a Corn group were composed. At the end of the study, blood samples taken from all animals were analysed. In the study, serum glucose, TP, urea, creatinine and BUN levels were found to be significantly decreased in Corn group compared to Wheat group. Serum ALT levels were similar with the Control and Wheat groups, but it was found to be significantly decreased in the Corn group. Serum AST levels were significantly lower in the Corn group. Serum GGT levels were significantly lower in both Corn and Wheat groups compared to Control group. Serum Ca and P levels were similar with all groups, whereas Mg levels were significantly decreased in Wheat and Corn groups compared to the Control group. As a result, it can be said that corn gluten added to the diet has more positive effects than wheat gluten in terms of both liver enzyme activities (AST, ALT and GGT) and renal parameters (Urea and BUN).</p>

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### Introduction

It is important to use feeds with appropriate nutrients and contents according to the type, breed, age and physiological status of the animals, as well as this cost of this diet in animal nutrition. Unfortunately, feed costs of enterprises increased due to rising feed prices in recent years. In order for the animals to be balanced and healthy, the diet they consume must be appropriate in terms of nutritional values (protein, energy, etc.) type and amount of nutrients.

Soybean meal is one of the most used feedstuffs in the feed sector as a source of vegetable protein. Soy plants are very rich in essential amino acids (lysine, methionine, loysin, isoloysin, histidine, etc.) and therefore are known to be the most valuable of plant protein sources (Vynias, 2006). Since the proteins are very valuable, the parts that are not used in human food production and industry are used as a source of protein for animal feed raw material (Liu, 2004). As soybean production is very limited in our

country, most of the soy products used in the feed sector are imported and this is one of the main reasons for the increase in feed prices.

Gluten is a protein compound obtained after separation of starch and small components in cereals (wheat, barley, corn, oats, rice, etc.) 75-85% of the dry matter is protein, 5-10% is lipid and the rest is it consists of starch and non-starch substances (Lafiandra et al., 2004). Glutens, which make up a large portion of the storage proteins in wheat (80-85% of the total protein) are composed of two fractions, monomeric gliadins and polymeric glutens, which are insoluble in water or saline (Lafiandra et al., 2004; Fellstone, 2011). Corn gluten, which is obtained as a by-product in wet milling of corn, also contains high levels of protein (Almeida et al., 2011).

This study was conducted to determine the effects of wheat and corn gluten on some serum parameters used as an alternative to soybean meal in diets of rats.

## Material and Methods

### Animals, Experimental Design, and Diets

This study was approved by Sivas Cumhuriyet University Animal Experiments Local Ethics Committee (Decision No: 2017/18). In this study, 24 male rats at the age of 20 days (*Wistar albino*) obtained from Sivas Cumhuriyet University Faculty of Medicine Experimental Animals Unit were used as animal material. In the experiment, 3 groups were formed, each having an equal number (8 animals). The majority of the protein content in the diet of the Control group was composed of soybean meal, the majority of the protein content in the diet of Wheat group was from wheat gluten and the majority of protein content in the diet of Corn group was made from corn gluten. The animals used in the study were fed with feeds given in Table 1 during the study. In the study 50-day fattening experiment was applied to the animals. Feed and water were given ad libitum. Comfort temperature (22°C) was applied to all animals during the study.

### Collection and Analysis Blood Samples

Cardiac blood samples were taken from all animals under anesthesia for biochemical analysis on the last day (70 days) of the study. Blood was collected from animals into anticoagulant tubes Becton Dickinson Co. USA) for approximately 5 cc. The sera were stored at -80°C until the day of analysis.

Serum glucose, total protein (TP), urea, creatinine, blood urea nitrogen (BUN),  $\gamma$ -glutamyltransferase (GGT), aspartate transaminase (AST), alanine transaminase (ALT), alkaline phosphatase (ALP), calcium (Ca), phosphorus (P) and magnesium (Mg) levels were measured in Mindray (BS 200, PRC).

### Statistical Analysis

The data obtained were evaluated using SPSS statistical package program (SPSS, 2011). One-way ANOVA test was used to determine whether there was a statistical difference between the data obtained from groups and Duncan test was used to determine which groups were different.

## Results and Discussion

In this study, data on blood parameters are given in Table 2. It was found that glucose ( $P<0.01$ ), total protein (TP), urea, creatinine and blood urea nitrogen (BUN) levels in Corn group decreased significantly compared to Wheat group ( $P<0.05$ ) as shown in Table 2. Serum alanine transaminase (ALT) levels were similar with the Control and Wheat groups ( $P>0.05$ ), but it was found to be significantly decreased in the Corn group ( $P<0.05$ ). Serum aspartate transaminase (AST) levels were also significantly decreased in the Corn group ( $P<0.01$ ). Serum alkaline phosphatase (ALP) levels were found to be statistically similar with all groups ( $P>0.05$ ). Serum gamma-glutamyl transferase (GGT) levels were significantly lower in both Corn and Wheat groups compared to the Control group ( $P<0.05$ ). Serum calcium and phosphorus levels were similar with all groups ( $P>0.05$ ) and magnesium levels were significantly decreased in Wheat and Corn groups compared to the control group ( $P<0.05$ ) (Table 3).

The nutrients they receive while feeding are able to change some metabolic parameters in the body. Serum parameters may also be affected by nutrition as biochemically. In this study, the effects of wheat and corn gluten, which are widely used as protein source in diet, on some parameters in rats were investigated. It is known that gluten, which constitutes a large portion of the protein contained in cereals, meets the protein needs of people and increases the quality of the products made from these cereals. On the other hand, it is known that some people consuming gluten-containing foods (people with HLA-DQ2 and HLA-DQ8 genes called human leukocyte antigens) are affected severely by autoimmune system, blood cytokine and reproductive parameters, especially the intestinal system, and the disease is called celiac disease. (Sapone et al., 2012; Hudacko et al., 2013; Soya and Ün, 2014). In this study, the effects of wheat and corn gluten, which are widely used as protein source in diet of rats on some parameters in serum were investigated.

Table 1. Dietary composition of different experimental groups of rats

The composition of the diet, %	Groups		
	Control	Corn	Wheat
Wheat bran	3.24	4.55	1.8
Oat, %11 CP	62.11	64.00	68
Sunflower meal, % 28 CP	6	13	13
Corn gluten meal, % 62 CP	-	16.8	-
Wheat gluten meal, % 75 CP	-	-	24.85
Soybean meal, % 51 CP	24.85	-	-
Animal fat	2.8	0.65	2.2
Vitamin-mineral premix*	1	1	1
	Nutrient composition (calculated)		
Metabolizable energy energy, (kcal kg <sup>-1</sup> )	2598	2598	2599
Crude protein, %	22	22	22

\*The vitamin-mineral premix provides the following (per kg): vitamin A 6.000.000 IU; vitamin D3 800.000 IU; vitamin E 8000 mg; vitamin K3 2000 mg; vitamin B1 1200 mg; vitamin B2 3000 mg; vitamin B6 2000 mg; vitamin B12 8 mg; niacin 10000 mg; folic acid 400 mg; d-biotin 20 mg; choline chloride 160.000 mg; manganese 32000 mg; iron 16000 mg; zinc 24.000 mg; copper 2000 mg; iodine 800 mg; cobalt 200 mg; selenium 60 mg; Cal-D-Pan. 4000 mg; antioxidant 4000 mg. CP: Crude protein.

Table 2. Results of blood serum analysis

Parameters	Groups			P-Value
	Control	Corn	Wheat	
Glucose, mg/dL	177.875±7.626 <sup>a</sup>	124.000±9.582 <sup>b</sup>	178.125±13.34 <sup>a</sup>	**
TP, g/dL	6.857±0.170 <sup>ab</sup>	5.957±0.534 <sup>b</sup>	7.213±0.161 <sup>a</sup>	*
Creatinin, mg/dL	1.020±0.083 <sup>ab</sup>	0.959±0.044 <sup>b</sup>	1.184±0.027 <sup>a</sup>	*
Urea, mg/dL	42.133±4.442 <sup>ab</sup>	29.864±4.218 <sup>b</sup>	45.395±4.065 <sup>a</sup>	*
BUN, mg/dL	17.500±2.639 <sup>ab</sup>	14.000±2.024 <sup>b</sup>	21.250±1.916 <sup>a</sup>	*
AST, U/L	205.875±24.620 <sup>a</sup>	135.429±3.866 <sup>b</sup>	246.875±26.678 <sup>a</sup>	**
ALT, U/L	103.788±5.519 <sup>a</sup>	75.500±9.166 <sup>b</sup>	95.863±7.199 <sup>ab</sup>	*
AST / ALT	1.98	1.79	2.58	
ALP, U/L	381.857±22.430	365.000±42.538	409.250±30.741	ns
GGT, U/L	4.667±0.211 <sup>a</sup>	3.571±0.297 <sup>b</sup>	3.125±0.441 <sup>b</sup>	*

All values are given as mean ± standard error of mean (SE), (n=8). ns: not significant (P>0.05); <sup>a,b</sup>: A letter in the same line means significantly different (\*: P<0.05), (\*\*: P<0.01); TP: total protein, BUN: blood urea nitrogen, AST: aspartate transaminase, ALT: alanine transaminase, ALP: alkaline phosphatase, GGT:  $\gamma$ -glutamyltransferase.

Table 3. Serum calcium (Ca), phosphorus (P) and magnesium (Mg) values of the groups

Parameters	Groups			P-Value
	Control	Corn	Wheat	
Ca, mg/dL	10.328±0.394	10.396±0.181	10.438±0.167	ns
P, mg/dL	10.735±0.542	11.164±0.525	11.156±0.662	ns
Mg, mg/dL	3.403±0.148 <sup>a</sup>	2.832±0.115 <sup>b</sup>	2.880±0.139 <sup>b</sup>	*

All values are given as mean ± standard error of mean (SE), (n=8). ns: not significant (P>0.05); <sup>a,b</sup>: A letter in the same line means significantly different (\*: P<0.05).

Protein fractions in corn are divided into four different groups as albumin, globulin, glutelin and zein, whereas zein type proteins are the most abundant group and they are very poor in lysine and tryptophan amino acids (Osborne, 1897; Vasal, 2002). Zein constitutes 50% or more of the total proteins found in the corn endosperm. Since urea, which is the final product of protein metabolism in mammals, is a small water-soluble molecule, it can enter freely into cell membranes and can pass into all tissues, cells and fluids (blood, milk, urine) in the body (Gustafsson and Palmquist, 1993; O'callaghan and Boland, 1999). In this study, it was observed that serum total protein, urea, BUN and creatinine levels decreased significantly in the corn group compared to the wheat group. High levels of urea and creatinine for a long time are considered as indicators of renal failure (Lameire et al., 2005; Ögütmen, 2011). Accordingly, it can be stated that the negative effects of corn gluten on kidneys are less.

AST and ALT activities in blood serum are generally considered as an index of liver damage and this trend is known to be different in rodents (Ha et al., 2001). It was also emphasized that the enzyme activities of AST, ALT, ALP and GGT increased in serum when the cell integrity was affected and degeneration of parenchymal cells (İmren and Turan, 1985; Huseby and Ingebretsen, 1993). Aspartate aminotransferase (AST) / alanine aminotransferase (ALT) ratio higher than 2 has been reported to indicate liver damage (De Bruyn and Graviss, 2001; Giannini et al., 2003). In this study, both the AST and ALT levels in the corn group and the AST / ALT ratio of less than 2 indicate that the negative effects of corn gluten on the liver were lower than wheat gluten and the low GGT level in the corn group supported this (De Bruyn and Graviss, 2001; Giannini et al., 2003). Similar to the results obtained in this study, Göze et al. (2019) in their study of rats in animals with liver damage, serum AST, ALT and GGT levels have been reported to increase and AST / ALT ratio is over 2.

It has been reported that cereal proteins play an important role in the emergence of autoimmune disease Type 1 diabetes (Antvorskov et al., 2014). It is reported that the incidence of Type 1 diabetes is higher in dietary animals with a high gluten content (Hoorfar et al., 1991). On the other hand, it has been reported that a gluten-free diet may reduce the incidence of diabetes in animals and the progression of type 1 diabetes (Elliott and Martin, 1984; Funda et al., 1999; Pastore et al., 2003). However, the hydrolyzate derived from corn protein, zein, has been reported to have a positive function in the fight against type 1 diabetes (Hira et al., 2009). Similar to the above information, in this study, it was found that the lowest serum glucose level was in the group using corn gluten in the diet.

Magnesium is an important element for biological systems, biochemical enzymatic and metabolic processes involved in various (Fischer and Giroux, 1991). It is also involved in the transport of ions in the body in various mechanisms (pump, carrier channel, etc.) and thus modulates signal transduction and cytosolic calcium, potassium and sodium ions concentrations (Herroeder et al., 2011). In this study, while serum Ca and P levels were not different between the groups, Mg levels were significantly decreased in both wheat gluten and corn gluten groups.

## Conclusions

As a result; It can be said that corn gluten added to the diet has more positive effects than wheat gluten in terms of both liver enzyme activities (AST, ALT and GGT) and kidney parameters (Urea and BUN). It can be stated that there is no difference in terms of Ca and P levels in terms of mineral metabolism and Mg level is affected by gluten in diet. However, there are no studies on this subject in the literature, it is thought that new studies are needed to evaluate the results more accurately.

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