ECRONICON OPEN ACCESS

EC PHARMACOLOGY AND TOXICOLOGY

Research Article

# Histological, Hematological and Biochemical Effects of Bee Bread on DSS-Induced Colitis Model in Rats

**Sibel Silici[1], Serhat Özer[1], Aslı Okan[2], Batuhan Aycı[3], Meltem Soylu[4] and Züleyha Doğanyiğit[2]***

[1]*Department of Agricultural Biotechnology, Seyrani Faculty of Agriculture, Erciyes University, Kayseri, Turkey*

[2]*Department of Histology and Embryology, Faculty of Medicine, Yozgat Bozok University, Erdoğan Akdağ Campus, Yozgat, Turkey*

[3]*Cerrahpasa Medical Faculty, Istanbul University-Cerrahpasa, Istanbul, Turkey*

[4]*Nutrition and Dietetic Department, Faculty of Health Sciences, Biruni University, Istanbul, Turkey*

***Corresponding Author:** Züleyha Doğanyiğit, Department of Histology and Embryology, Faculty of Medicine, Yozgat Bozok University, Erdoğan Akdağ Campus, Yozgat, Turkey.

**Received:** March 08, 2022; **Published:** March 29, 2022

## Abstract

Inflammatory bowel disease (IBD) is a systemic disease associated with oxidative stress and inflammation. Bee bread (perga, fermented pollen) is an essential bee product used in apitherapy due to its antioxidant and anti-inflammatory effects. In this study, determining the histological, hematological, and biochemical effects of bee bread was aimed on IBD in the dextran sodium sulfate (DSS) induced colitis model in rats. A colitis model was established by gavage administration of DSS rats. Fifty-six rats were randomly divided into 7 groups and the effects of low and high doses of bee bread were examined. Significant improvements in lymphocyte count and erythrocyte parameters were detected. ALT and uric acid levels also normalized. In histopathological evaluation, low dose bee bread significantly decreased intestinal damage. Bee bread may be a potential treatment agent in IBD patients. The nutrients used can improve the systemic effects of the disease by regulating hematological and biochemical values.

***Keywords:*** *Bee Bread; Colitis; Dextran Sodium Sulfate; Hemoglobin; Inflammatory Bowel Disease; Tissue Damage*

## Abbreviations

IBD: Inflammatory Bowel Disease; UC: Ulcerative Colitis; CD: Crohn's Disease; DSS: Dextran Sodium Sulfate; GGT: Gamma Glutamyl Transpeptidase; LDH: Lactate Dehydrogenase; AST: Aspartate Aminotransferase; ALT: Alanine Aminotransferase; ALP: Alkaline Phosphatase; WBC: Hematological Parameters, Leukocyte Count; RBC: Erythrocyte Count; Hgb: Hemoglobin Concentration; HTC: Hematocrit Ratio; MCV: Mean Erythrocyte Volume; MCH: Mean Erythrocyte Hemoglobin; MCHC: Mean Erythrocyte Hemoglobin Concentration; EOS: Eosinophil Amount; LYM: Lymphocyte Amount; NEU: Neutrophil Amount

## Introduction

Inflammatory bowel disease (IBD) is a systemic disease characterized by recurrent chronic intestinal inflammation. In IBD, the higher frequency of the relatives of the patients indicates a strong genetic predisposition. Basically, IBD is divided into ulcerative colitis (UC) and Crohn's disease (CD). The etiology of IBD includes a complex interaction between genetic, environmental, or microbial factors and immune responses [1-3].

One of those directing the immune response in IBD is subtypes of T cells [4,5]. However, dendritic cells and macrophages are involved in the immune response as well as T cells [6]. Cytokines released from immune cells control the inflammatory response. The imbalance between pro-inflammatory and anti-inflammatory cytokines that occur in IBD prevents the resolution of inflammation. Thus, this imbalance causes the disease to continue and tissue damage [7].

In IBD, the intestinal epithelium may deteriorate during tissue damage in the intestine. This epithelial tissue response regulates barrier function, composition of the microbiota, and immune cells residing in the intestinal tissue. Therefore, abnormal signal transmission between epithelium and immune cells may increase immune dysregulation in the disease. As a result, IBD can be triggered by defects in the integrity of the epithelial barrier [8,9]. However, reactive oxygen species affect the epithelial barrier [10].

The pathophysiology of IBD is associated with reactive oxygen species [11]. Disruption of the balance between the production of reactive oxygen species and their elimination leads to oxidative stress. This imbalance may result in chronic inflammation, as observed in IBD [12]. Related to this, oxidative stress signaling in IBD is considered among potential therapeutic approaches [13].

Different drug classes such as amino salicylates, antibiotics, immunomodulators are used therapeutically. However, there is no curative therapy method for this disease [14]. Anti-inflammatory and antioxidant properties of bee pollens, which can be candidates in this direction, have been revealed. Antioxidant activity of bee products suggests that it may be beneficial in diseases such as IBD in which the oxidative balance is impaired [15,16].

Bee bread, an apitherapy product, is a product that results from the fermentation of a mixture of pollen, nectar and bee saliva interacting with bacteria and yeasts [17]. The resulting product is a rich source of phenolic compounds associated with high antioxidant activity. Studies have reported the antioxidant properties of bee bread [18-20]. However, the anti-inflammatory properties of bee bread are known in studies [21,22]. The antioxidant and anti-inflammatory activity of bee bread is important in terms of the effectiveness of bee bread in IBD disease, which is associated with inflammation and oxidative stress.

In this study, the chemical structure of the bee bread sample was analyzed. The effect of bee bread was aimed in the experimental colitis model created in rats. Histopathological changes in the intestines of animals and liver enzymes and blood parameters were biochemically analyzed.

## Materials and Methods

### Animals

8-week-old Wistar albino male rats (n = 56, 280 - 350g) were used in the study. Rats were obtained from Erciyes University Experimental and Clinical Research Center (DEKAM). Experimental animals were kept at room temperature 21 ± 1°C throughout the study. The rats to be used in the experiment were exposed to a 12-hour light cycle and dark cycle. Rats were kept in cages in groups of four with unlimited access to standard food. According to the experimental group, dextran sodium sulfate (DSS) was added, or they had unlimited access to drinking water. Ethics committee approval required for the study was obtained from Erciyes University Animal Experiments Local Ethics Committee (decision dated 15.11.2017 and numbered 17/114).

### Chemicals

The DSS product Sigma-Aldrich (St Louis, MO, Sigma) was supplied in powder form. This chemical was dissolved in water and was given with drinking water [23]. Lansoprazole was supplied from Lansor (30 mg). Bee bread was obtained from Erciyes University Technopark (Nutral Therapy R&D company). Bee bread was dissolved in water at doses of 50 mg/kg and 100 mg/kg and administered orally via gavage. Ketamine and xylazine were used for anesthesia.

### Chemical analysis and bee bread samples

Bee bread was stored at -20°C until usage. Using the AOAC method, the moisture content of bee bread was gravimetrically determined by drying it to constant weight at 105°C in a convection oven. Crude protein value was determined using Kjeldahl's method [24]. A conversion factor of 6.25 was used to convert the percentage of nitrogen to percentage of crude protein. The crude oil was extracted according to the AOAC method using a Soxhlet apparatus and diethyl ether [25]. Crude fiber content of bee bread was analyzed according to the AOAC method. Ash content of the samples was gravimetrically determined [24]. The total carbohydrate content was calculated according to the expression: total carbohydrate = 100-(% moisture +% protein +% fat +% ash).

Aliquots of 1.5g bee bread sample was accurately weighted into a 100 mL flask. They were then dissolved in 10 ml ethanol (95%) by agitation on a vortex mixer followed by ultrasonic assisted extraction in an ultrasonic cleaning bath for 60 minutes at 40°C. The mixture was centrifuged at 2700g for 30 minutes at 40°C. The supernatant was collected in a pear-shaped flask. The extraction procedure was repeated twice, and the supernatant extraction solutions were combined into a 25 ml volumetric flask. The volume was made up to the mark with ethanol (95%). Prior to analysis, a portion of the supernatant was filtrated through a 0.45 $\mu m$ membrane and diluted to the appropriate concentration within the range of the calibration curves. The content of total phenolic compounds was performed according to the Folin-Ciocalteu method proposed [26]. 1,1-Diphenyl-2-picrylhydrazyl (DPPH) assay The DPPH assay was based on the 96-well plate assay described by Herald., *et al*. with some modifications [27].

### Creating colitis model with a DSS

Colitis model Acute colitis model was created by giving 5% (weight/volume) DSS aqueous solution as drinking water in 5 days. The chronic colitis model was obtained by applying 3% (weight/volume) DSS for the following 10 days. Wistar albino rats were not treated as negative control group, but low dose bee bread applied control group (AEDD), high dose bee bread applied control group (AEYD) was induced. Also, DSS induced colitis group (DSS), low dose bee bread after DSS induction randomly divided into 7 groups as treatment group (DSS + AEDD) in total, treatment group (DSS + AEYD) where high dose bee bread was given after DSS induction and reflor group (positive control) (DSS + Reflor) (n = 8 in each group). AEDD and AEYD groups were given 50 mg/kg and 100 mg/kg orally once a day for 2 weeks, respectively. Colitis was induced by adding 5% DSS to the drinking water of the DSS group for one week and 3% for the following 2 weeks. The DSS + AEDD and DSS + AEYD groups were given 50 mg/kg and 100 mg/kg bee bread orally once a day for 2 weeks after the induction of colitis, respectively.

After completing the experimental protocol, rats were sacrificed under ketamine/xylazine anesthesia. Every effort has been made to minimize the suffering of the animals. Blood and intestinal tissue samples were collected. The samples taken with the methods described below were examined.

### Histological analysis

Intestinal tissues were fixed with 10% formaldehyde solution to be used in histological examinations. After fixation, tissues were embedded in paraffin by applying routine tissue follow-up steps. 5 - 6 $\mu m$ sections from paraffin blocks were taken on slides. The prepared slides were then diluted with graded alcohol by removing the paraffin with xylene after the oven. Hematoxylin-eosin (H&E) staining was applied to the sections taken [28]. Intestinal scoring was made by considering criteria such as villi shortening and thinning from 50 areas, presence of necrotic cells and shortening of the mucosa. According to these criteria, 0: no damage, 1: little damage, 2: moderate damage, and 3: severe damage. Examinations were carried out under a light microscope (Olimpus BX51).

### Collection and analysis of blood samples

Blood samples were collected from the animals only by the end of the 30 and 90 days of the study. The animals were fasted for 6h prior to the collection of blood samples. Due to the risk of change in biochemical parameters, the animals were not administered general anesthesia prior to the taking of the samples. The animals were maintained under light ether anesthesia, performed just before the collection of blood samples, and samples were slowly collected from each animal into tubes both with and without anticoagulants, by means of the insertion of a cannula in the heart. Blood was taken from every seven animals in each group. Analyses for the parameters to be investigated were performed on the same day, in blood samples collected into the tubes of both types.

### Statistical analysis

SPSS 22 package program (SPSS, Inc., Chicago, IL, USA) was used for statistical analysis. Results are expressed as mean ± standard deviation. A comparison was made between groups with the One-Way ANOVA test. Post-Hoc Tukey test was used for paired comparisons. A value of $p < 0.05$ was considered statistically significant.

## Results and Discussion

### Chemical analysis of bee bread sample

According to the nutrient content of bee bread used in the study, the bread contains 18.8 g/100g dietary fiber, 8.14 g/100g ash and 408 kcal/100g energy. However, bee bread was found to contain 13.56 g/100g protein, 30.6 g/100 g carbohydrate, 21.69 g/100g fat. Total phenolic content was determined as 43.44 (mg GAE/g) and antioxidant activity as 3.88 ± 0.030 (mg TEAC/g). The nutritional elements and bioactivity of bee bread are given in table 1.

| Nutrients | |
|---|---|
| Ash | 8.14 g/100g |
| Protein | 13.56 g/100g (Nx6.25) |
| Carbohydrate | 30.60 g/100g |
| Dietary fiber | 18.18 g/100g |
| Oil | 21.69 g/100g |
| Energy | 408 kcal/100g |
| Total phenolic content | 43.44 ± 0.78 (mg GAE/g) |
| Antioxidant activity (DPPH) | 3.88 ± 0.030 (mg TEAC/g) |

***Table 1:*** *The nutrient content of bee bread used in the study.*

### Hematological analysis

RBC values was found to be significantly lower in the DSS and DSS + AEDD groups ($p < 0.05$). When the groups were compared in terms of Hgb, a significant difference was found ($p < 0.05$). The lowest Hgb level was observed in the DSS group. Bee bread improved the drop in MCHC levels of the DSS-induced groups. As the MCV values of the groups were compared with the control group, a significant difference was obtained ($p < 0.05$).

The MCV values of the bee bread groups were also higher than the DSS + Reflor group. A significant decrease was detected in the DSS group in terms of MCH level ($p < 0.05$). When compared with the control group, all groups except the DSS group were found to have close

MCH levels. MCHC level was significantly higher in AEYD and DSS + Reflor groups ($p < 0.05$). The lowest MCHC value was significantly observed in the DSS group ($p < 0.05$). In addition, eosinophil levels were higher in all DSS groups. In the groups that DSS and bee bread were given together (DSS + AEDD, DSS + AEYD and DSS + Reflor), this level was found significantly decrease ($p < 0.05$). The highest lymphocyte level was measured in the AEYD group and the lowest in the DSS group. As these data were compared with the control group, a statistical difference was obtained between the experimental groups ($p < 0.05$). While no difference was found between the control and AEYD groups in terms of neutrophil levels, the lowest neutrophil level was found in the DSS group ($p < 0.05$). There was no significant difference in WBC and HTC levels. The results of the hematological analyzes obtained from the research are shown in table 2.

| Groups | Control | DSS | AEDD | AEYD | DSS+AEDD | DSS+AEYD | DSS+Reflor |
|---|---|---|---|---|---|---|---|
| WBC | 9.66 ± 3.80 | 11.42 ± 0.88 | 6.48 ± 2.10 | 6.91 ± 1.63 | 10.15 ± 0.52 | 9.38 ± 1.63 | 9.93 ± 5.49 |
| RBC | $9.40 \pm 0.37^{b}$ | $8.28 \pm 0.32^{a}$ | $9.10 \pm 0.67^{b}$ | $9.18 \pm 0.28^{b}$ | $8.33 \pm 0.12^{a}$ | $9.36 \pm 0.51^{b}$ | $9.16 \pm 0.25^{b}$ |
| HGB | $16.00 \pm 0.44^{b}$ | $12.15 \pm 1.13^{a}$ | $16.30 \pm 0.36^{b}$ | $16.37 \pm 0.46^{b}$ | $15.75 \pm 0.34^{b}$ | $16.23 \pm 0.39^{b}$ | $15.47 \pm 0.67^{b}$ |
| HTC | 51.37 ± 1.82 | 53.97 ± 2.09 | 54.67 ± 2.00 | 53.15 ± 1.10 | 53.97 ± 1.77 | 53.37 ± 1.82 | 52.20 ± 1.31 |
| MCV | $60.35 \pm 1.36^{c}$ | $50.23 \pm 0.16^{a}$ | $57.95 \pm 1.23^{bc}$ | $60.23 \pm 2.74^{c}$ | $57.73 \pm 0.64^{bc}$ | $57.07 \pm 1.72^{b}$ | $55.60 \pm 2.43^{b}$ |
| MCH | $17.85 \pm 0.39^{b}$ | $15.21 \pm 0.29^{a}$ | $17.38 \pm 0.96^{b}$ | $17.68 \pm 0.52^{b}$ | $17.33 \pm 0.29^{b}$ | $17.50 \pm 0.70^{b}$ | $17.67 \pm 0.06^{b}$ |
| MCHC | $29.57 \pm 0,68^{c}$ | $26.17 \pm 0.14^{a}$ | $28.80 \pm 0.51^{b}$ | $30.53 \pm 0.68^{d}$ | $30.03 \pm 0.21^{cd}$ | $30.67 \pm 0.29^{d}$ | $30.67 \pm 0.40^{d}$ |
| EOS | $0.07 \pm 0.06^{a}$ | $0.83 \pm 0.74^{b}$ | $0.12 \pm 0.10^{a}$ | $0.07 \pm 0.06^{a}$ | $0.23 \pm 0.21^{ab}$ | $0.28 \pm 0.16^{ab}$ | $0.62 \pm 0.59^{ab}$ |
| LYM | $74.65 \pm 5.39^{bc}$ | $56.77 \pm 6.37^{a}$ | $69.85 \pm 4.96^{b}$ | $81.60 \pm 3.99^{c}$ | $74.93 \pm 5.10^{bc}$ | $75.35 \pm 7.10^{bc}$ | $69.63 \pm 2.29^{b}$ |
| NEU | $28.60 \pm 2.14^{b}$ | $18.53 \pm 0.95^{a}$ | $20.63 \pm 6.75^{ab}$ | $30.10 \pm 4.89^{b}$ | $20.43 \pm 5.93^{ab}$ | $21.82 \pm 6.71^{ab}$ | $16.00 \pm 3.36^{a}$ |

***Table 2:*** *Hematological analysis results obtained from the study.*

*Data are expressed as mean ± standard deviation. P < 0.05 was considered significant. There is no significant difference between the groups containing the same letter (a-d).*

Anemia was discussed to be observed in colitis patients [30]. In our study, hemoglobin levels increased significantly after bee bread. Bee bread statistically improved the MCV, MCH and MCHC values. In addition, high dose bee bread significantly increased the RBC parameter. In a clinical study, intravenous iron supplementation was given to UC patients in the first phase. In the second phase, erythropoietin hormone was administered. Most of the patients responded to treatment in the first phase. In the second phase, effective treatment response was obtained from patients that did not respond in the first phase [31]. In a clinical study conducted on patients with CH, erythropoietin was given along with intravenous iron. According to the study, a significant response was obtained with erythropoietin. The measured hematological responses increase the quality of life. Most patients with CD-related anemia are said to respond to intravenous iron alone. In addition, erythropoietin has been reported to have additional effects on hemoglobin concentrations [32]. Based on these data, bee bread can be a potential treatment method that preserves tissue integrity in patients with CD accompanied by anemia. The improvement of bee bread in anemia can positively affect the quality of life of colitis patients. In our colitis study, no significant difference was found in WBC levels. Studies have found that the WBC levels of colitis patients generally increase [33]. The DSS-induced colitis model in our study may cause a difference.

In our study, neutrophil levels were statistically decreased in the DSS model. In addition, bee bread significantly increased this parameter despite the decrease in the number of lymphocytes. In a study conducted in UC, the lymphocyte count of the patients decreased while the neutrophil values increased [34]. UC clinical study was noted that neutrophil counts increased and the number of lymphocytes decreased [35]. The use of the DSS model in our study may have a restrictive role on neutrophil count. Bee bread was predicted to contribute for the regulation of the immune system by increasing the lymphocyte count.

## Biochemical analysis

ALP levels were significantly higher in DSS-induced groups (DSS, DSS + AEDD, DSS + AEYD and DSS + Reflor) compared to bee bread and control groups ($p < 0.05$). The highest ALT levels were found in the DSS group. However, uric acid levels were found to be significantly higher in DSS, DSS + AEDD and DSS + Reflor groups compared to other groups ($p < 0.05$). Group given high doses of bee bread (DSS + AEYD) significantly decreased ALT and uric acid levels compared to the DSS group ($p < 0.05$). The results of the biochemical analysis obtained from the study are shown in table 3.

| Groups | Control | DSS | AEDD | AEYD | DSS+AEDD | DSS+AEYD | DSS+Reflor |
|---|---|---|---|---|---|---|---|
| ALP | $119.33 \pm 21.30^{a}$ | $162.33 \pm 33.30^{b}$ | $112.75 \pm 13.60^{a}$ | $107.80 \pm 12.80^{a}$ | $146.17 \pm 23.20^{b}$ | $140.80 \pm 15.62^{b}$ | $147.25 \pm 18.60^{b}$ |
| ALT | $145.70 \pm 13.30^{ab}$ | $227.00 \pm 21.30^{c}$ | $138.20 \pm 10.66^{b}$ | $126.20 \pm 5.74^{a}$ | $182.14 \pm 15.80^{bc}$ | $149.00 \pm 18.30^{ab}$ | $187.50 \pm 23.80^{bc}$ |
| AST | $58.50 \pm 7.03$ | $78.25 \pm 11.20$ | $54.00 \pm 9.05$ | $51.00 \pm 8.12$ | $59.67 \pm 6.05$ | $59.40 \pm 8.30$ | $69.40 \pm 7.02$ |
| Uric acid | $0.28 \pm 0.08^{a}$ | $0.48 \pm 0.09^{b}$ | $0.27 \pm 0.04^{a}$ | $0.28^{a}$ | $0.46 \pm 0.06^{b}$ | $0.36 \pm 0.06^{a}$ | $0.48 \pm 0.14^{b}$ |
| Creatinine | $1.07 \pm 0.19$ | $1.50 \pm 0.56$ | $1.24 \pm 0.12$ | $1.00 \pm 0.08$ | $1.44 \pm 0.34$ | $1.10 \pm 0.31$ | $1.50 \pm 0.59$ |

***Table 3:*** *Results of biochemical analysis obtained from the study.*

*Data are expressed as mean ± standard deviation. P < 0.05 was considered significant. There is no significant difference between the groups containing the same letter (a-c).*

According to the results we obtained in our study, DSS increased ALT levels of rats while bee bread at high doses significantly decreased ALT levels. In the DSS-induced colitis model, the effect of vitex, which is a flavonoid, on liver enzymes was investigated. In the animal model, AST and ALT increased due to DSS, and ALT levels decreased in the vitex extract groups [36]. In another DSS-mediated animal study, the effects of Dendrobium officinale polysaccharides on the liver were investigated. In the study conducted, AST and ALT parameters increased after DSS [37]. Accordingly, bee bread can be suggested to have a normalizing effect. This effect might be used therapeutically for ALT disruptions in IBD patients and may contribute to systemic improvements.

In our study, uric acid levels significantly increased in the DSS-induced colitis group, while high-dose bee bread decreased the uric acid level. In a retrospective study, uric acid increased in the serum of UC patients [31]. In another study conducted in IBD patients, serum uric acid levels increased [32]. In the light of these data, bee bread can regulate uric acid metabolism in IBD. Reducing uric acid levels with bee bread can be effective for reducing the activity of the disease.

## Histological analysis

Only in the DSS group, shortening of the neck of the villi, necrosis of the surface epithelial cells, shortening of the mucosa layer and shrinkage of the glands were observed. In relation to this, a significant amount of damage increased in the DSS group compared to the control ($p < 0.05$) (Figure 1). A near control image was observed in AEDD, AEYD and DSS + Reflor groups. A significant reduction in damage was found in the DSS + AEDD group compared to the control ($p < 0.05$). The histological analysis results obtained from the study are expressed in table 4.

| Groups | Control | DSS | AEDD | AEYD | DSS+AEDD | DSS+AEYD | RF | p |
|---|---|---|---|---|---|---|---|---|
| Score | $0.13 \pm 0.34^{a}$ | $0.96 \pm 0.88^{b}$ | $0.16 \pm 0.37^{a}$ | $0.36 \pm 0.66^{a}$ | $0.30 \pm 0.53^{a}$ | $0.56 \pm 0.77^{ab}$ | $0.20 \pm 0.40^{ab}$ | 0,001 |

***Table 4:*** *Results of histological analysis obtained from the study.*

*Data are expressed as mean ± standard deviation. P <0.05 was considered significant. There is no significant difference between the groups containing the same letter (a-b).*

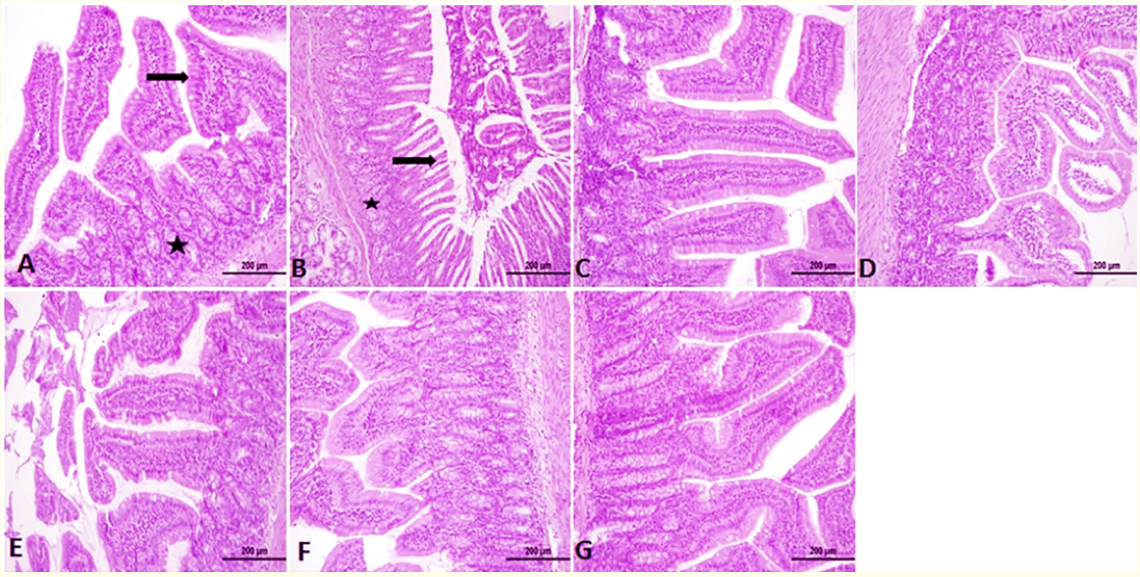


***Figure 1:*** *Intestinal tissue stained with hematoxylin and eosin. Control group (A) normal bowel structure is shown with asterisks and villi with asterisks, DSS group (B) thinning of the villi in the intestine with arrows and asterisks showing shortening of the glands, PD group (C), PY group (D), PY + DSS group (E), PD + DSS group (F), RF group (G). Images are magnified at X200 level.*

Inflammation-related tissue damage occurs in IBD. Bee products are used to reduce this damage [38]. In our study, low dose bee bread significantly reduced tissue damage in colitis. There are studies on the effect of propolis in bee products on colitis. In a study conducted in Egypt, the effect of propolis in rats, created as a UC model, was examined. In the research, propolis regulates the macroscopic and microscopic view. In the evaluation, the ulcerative index and lesion score decreased by bee product. In measurements, propolis has been shown to reduce oxidative markers and inflammatory mediator molecules. However, propolis, one of the bee products, has been suggested to be a powerful free radical scavenger [39]. The effects of propolis belonging to two countries were examined by causing colitis in rats through DSS. Propolis has reduced colitis disease activity index. Resistance to DSS-induced colonic oxidative stress increased in the groups given propolis. Bee product suppressed inflammatory markers. Based on this, propolis was considered to have a protective role in DSS-induced colitis [40]. The effect of different doses of propolis was examined in rats with colitis. The 0.3% propolis resulted in a lower disease activity index and an increase in colon length/weight ratio. A better distal colon tissue was observed in histological examination. However, compared to the control group, the symptoms of colitis were significantly improved with bee product intervention. In the study, propolis was assumed to attenuate DSS-induced colitis [41]. The effects of propolis were investigated in the colitis model created by giving DSS. Bee product reduced the severity of colitis as assessed by body weight, spleen weight and colon length. Propolis caused a decrease in inflammatory cytokines and infiltration of immune cells. In addition, loss of goblet cells and antibody reactivity to inflammatory markers decreased after bee product treatment [42]. In our study, the effect of propolis was similar to previous tries, another bee product, on tissue damage in colitis can be attributed to the antioxidant and anti-inflammatory activity of both bee products. However, the use of bee bread with propolis may contribute to the reduction of tissue damage and tissue improvement. Bee bread can positively influence markers associated with inflammation. The fact that only low doses reduce tissue damage indicates that bee bread should be consumed in accordance with the dose.

## Conclusion

IBH is a chronic inflammatory disease. Bee bread has anti-inflammatory and antioxidant properties. In this study, a colitis model was established by gavage administration of DSS to rats. Histological, hematological, and biochemical effects of bee bread were analyzed on IBD. Erythrocyte parameters and lymphocyte count were significantly improved. ALT and uric acid levels were also found to be normalized. In histopathological evaluation, intestinal damage was significantly decreased with low dose bee bread. Bee bread might be a potential therapeutic target for IBD patients. The nutrient can regulate hematological and biochemical values, improving the symptoms of the disease. Bee bread may have a healing role in tissue damage of colitis. This healing can create a systemic effect by modulating liver and blood parameters.

## Acknowledgements

The study was supported by the ERÜ-BAP unit with the project code numbered FYL-2018-7894.

## Conflict of Interest

The authors declare no conflict of interest.

## Bibliography

1. Zhang Yi-Zhen and Yong-Yu Li. "Inflammatory bowel disease: pathogenesis". *World Journal of Gastroenterology* 20.1 (2014): 91-99.
2. Loddo Italia and Claudio Romano. "Inflammatory bowel disease: genetics, epigenetics, and pathogenesis". *Frontiers in Immunology* 6 (2015): 551.
3. Garber Ari and Miguel Regueiro. "Extraintestinal manifestations of inflammatory bowel disease: epidemiology, etiopathogenesis, and management". *Current Gastroenterology Reports* 21.7 (2019): 31.
4. Shen Hongxing and Lewis Zhichang Shi. "Metabolic regulation of TH17 cells". *Molecular Immunology* 109 (2019): 81-87.
5. Giuffrida Paolo and Antonio Di Sabatino. "Targeting T cells in inflammatory bowel disease". *Pharmacological Research* 159 (2020): 105040.
6. Kmieć Zbigniew., *et al*. "Cells of the innate and adaptive immunity and their interactions in inflammatory bowel disease". *Advances in Medical Sciences* 62.1 (2017): 1-16.
7. Neurath Markus F. "Cytokines in inflammatory bowel disease". *Nature Reviews. Immunology* 14.5 (2014): 329-342.
8. Martini Eva., *et al*. "Mend Your Fences: The Epithelial Barrier and its Relationship With Mucosal Immunity in Inflammatory Bowel Disease". *Cellular and Molecular Gastroenterology and Hepatology* 4.1 (2017): 33-46.
9. Yu Linda Chia-Hui. "Microbiota dysbiosis and barrier dysfunction in inflammatory bowel disease and colorectal cancers: exploring a common ground hypothesis". *Journal of Biomedical Science* 25.1 (2018): 79.
10. Novak Elizabeth A and Kevin P Mollen. "Mitochondrial dysfunction in inflammatory bowel disease". *Frontiers in Cell and Developmental Biology* 3 (2015): 62.

11. Pavlick Kevin P., *et al*. "Role of reactive metabolites of oxygen and nitrogen in inflammatory bowel disease". *Free Radical Biology and Medicine* 33.3 (2002): 311-322.

12. Hussain Tarique., *et al*. "Oxidative Stress and Inflammation: What Polyphenols Can Do for Us?". *Oxidative Medicine and Cellular Longevity* (2016): 7432797.

13. Tian Tian., *et al*. "Pathomechanisms of Oxidative Stress in Inflammatory Bowel Disease and Potential Antioxidant Therapies". *Oxidative Medicine and Cellular Longevity* (2017): 4535194.

14. Abraham Bincy P., *et al*. "Inflammatory Bowel Disease: Pathophysiology and Current Therapeutic Approaches". *Handbook of Experimental Pharmacology* 239 (2017): 115-146.

15. Li Qiang-Qiang., *et al*. "Nutrient-rich bee pollen: A treasure trove of active natural metabolites". *Journal of Functional Foods* 49 (2018): 472-484.

16. Alvarenga Livia., *et al*. "To bee or not to bee? The bee extract propolis as a bioactive compound in the burden of lifestyle diseases". *Nutrition (Burbank, Los Angeles County, Calif.)* 83 (2021): 111094.

17. Khalifa Shaden AM., *et al*. "Recent insights into chemical and pharmacological studies of bee bread". Trends in *Food Science and Technology* 97 (2020): 300-316.

18. Baltrušaitytė Vilma., *et al*. "Radical scavenging activity of different floral origin honey and beebread phenolic extracts". *Food Chemistry* 101.2 (2007): 502-514.

19. Silva João Carlos., *et al*. "Antimicrobial activity, phenolic profile and role in the inflammation of propolis". *Food and Chemical Toxicology: An International Journal Published for the British Industrial Biological Research Association* 50.5 (2012): 1790-1795.

20. Borycka Katarzyna., *et al*. "Antioxidant and antibacterial properties of commercial bee pollen products". *Journal of Apicultural Research* 54.5 (2015): 491-502.

21. Rzepecka-Stojko Anna., *et al*. "Polyphenols from Bee Pollen: Structure, Absorption, Metabolism and Biological Activity". *Molecules (Basel, Switzerland)* 20.12 (2015): 21732-21749.

22. Bakour Meryem., *et al*. "Antioxidant activity and protective effect of bee bread (honey and pollen) in aluminum-induced anemia, elevation of inflammatory makers and hepato-renal toxicity". *Journal of Food Science and Technology* 54.13 (2017): 4205-4212.

23. Egger B., *et al*. "Characterisation of acute murine dextran sodium sulphate colitis: cytokine profile and dose dependency". *Digestion* 62.4 (2000): 240-248.

24. Horwitz William and George W Latimer. "Official methods of analysis of AOAC International". Gaithersburg, Md: AOAC International. 18th edition (2005).

25. Williams Sidney. Official methods of analysis of the Association of official analytical chemists. Arlington (Va.): Association of official analytical chemists. 14th edition (1984).

26. Magalhães Luís M., *et al*. "Rapid microplate high-throughput methodology for assessment of Folin-Ciocalteu reducing capacity". *Talanta* 83.2 (2010): 441-447.

27. Herald Thomas J., *et al*. "High-throughput micro plate assays for screening flavonoid content and DPPH-scavenging activity in sorghum bran and flour". *Journal of the Science of Food and Agriculture* 92.11 (2012): 2326-2331.

28. Doğanyiğit Z., *et al*. "The cardiotoxic effects of acute and chronic grayanotoxin-III in rats". *Human and Experimental Toxicology* 39.3 (2020): 374-383.

29. Eraslan Gökhan., *et al*. "Effect of carbaryl on some biochemical changes in rats: the ameliorative effect of bee pollen". *Food and Chemical Toxicology: An International Journal Published for the British Industrial Biological Research Association* 47.1 (2009): 86-91.

30. Jacobson-Kelly Amanda E., *et al*. "Trends in Anemia, Iron, Therapy, and Transfusion in Hospitalized Pediatric Patients with Inflammatory Bowel Disease". *The Journal of Pediatrics* 222 (2020): 141-145.e1.

31. Gasche C., *et al*. "Sequential treatment of anemia in ulcerative colitis with intravenous iron and erythropoietin". *Digestion* 60.3 (1999): 262-267.

32. Gasché C., *et al*. "Intravenous iron and erythropoietin for anemia associated with Crohn disease. A randomized, controlled trial". *Annals of Internal Medicine* 126.10 (1997): 782-787.

33. Torun Serkan., *et al*. "Assessment of neutrophil-lymphocyte ratio in ulcerative colitis: a promising marker in predicting disease severity". *Clinics and Research in Hepatology and Gastroenterology* 36.5 (2012): 491-497.

34. Hanai Hiroyuki., *et al*. "Relationship between fecal calprotectin, intestinal inflammation, and peripheral blood neutrophils in patients with active ulcerative colitis". *Digestive Diseases and Sciences* 49.9 (2004): 1438-1443.

35. Okba AM., *et al*. "Neutrophil/lymphocyte ratio and lymphocyte/monocyte ratio in ulcerative colitis as non-invasive biomarkers of disease activity and severity". *Autoimmunity Highlights* 10.1 (2019): 4.

36. Duan Shuni., *et al*. "Effect of vitexin on alleviating liver inflammation in a dextran sulfate sodium (DSS)-induced colitis model". *Biomedicine and Pharmacotherapy = Biomedecine and Pharmacotherapie* 121 (2020): 109683.

37. Liang Jian., *et al*. "Protective roles and mechanisms of Dendrobium officinal polysaccharides on secondary liver injury in acute colitis". *International Journal of Biological Macromolecules* 107.B (2018): 2201-2210.

38. Barbosa Bezerra Gislaine., *et al*. "Hydroalcoholic extract of Brazilian red propolis exerts protective effects on acetic acid-induced ulcerative colitis in a rodent model". *Biomedicine and Pharmacotherapy = Biomedecine and Pharmacotherapie* 85 (2017): 687-696.

39. Atta Attia H., *et al*. "Phytochemical studies and anti-ulcerative colitis effect of Moringa oleifera seeds and Egyptian propolis methanol extracts in a rat model". *Asian Pacific Journal of Tropical Biomedicine* 9.3 (2019): 98.

40. Wang Kai., *et al*. "Propolis from Different Geographic Origins Decreases Intestinal Inflammation and Bacteroides spp. Populations in a Model of DSS-Induced Colitis". *Molecular Nutrition and Food Research* 62.17 (2018): e1800080.

41. Wang Kai., *et al*. "Dietary Propolis Ameliorates Dextran Sulfate Sodium-Induced Colitis and Modulates the Gut Microbiota in Rats Fed a Western Diet". *Nutrients* 9.8 (2017): 875.

42. Hwang Soonjae., *et al*. "Oral administration of Korean propolis extract ameliorates DSS-induced colitis in BALB/c mice". *International Journal of Medical Sciences* 17.13 (2020): 1984-1991.

**Volume 10 Issue 4 April 2022**