Erciyes Med J 2023; 45(2): 131–7 • DOI: 10.14744/etd.2022.55492

ORIGINAL ARTICLE – OPEN ACCESS



# The Effects of Genistein as Supplement to Oral/Enteral Nutrition on Inflammatory Cytokines in Septic ICU patients: A Prospective, Single-center, Randomized Controlled Pilot Study

Gülseren Elay[1], Kürşat Gündoğan[1,2], İnayet Güntürk[3], Şahin Temel[1], Nurhayat Tuğra Özer[2], Hilal Sipahioğlu[1], Ömer Küçük[4], Cevat Yazıcı[3], Muhammet Güven[5], Murat Sungur[1,2]

**Cite this article as:**
Elay G, Gündoğan K, Güntürk İ, Temel Ş, Özer NT, Sipahioğlu H, et al. The Effects of Genistein as Supplement to Oral/Enteral Nutrition on Inflammatory Cytokines in Septic ICU patients: A Prospective, Single-center, Randomized Controlled Pilot Study. Erciyes Med J 2023; 45(2): 131-7.

[1]Division of Intensive Care, Department of Medicine, Erciyes University Faculty of Medicine, Kayseri, Türkiye
[2]Department of Clinical Nutrition, Erciyes University, Health Science Institute, Kayseri, Türkiye
[3]Department of Clinical Biochemistry, Erciyes University Faculty of Medicine, Kayseri, Türkiye
[4]Department of Hematology and Medical Oncology, Emory University Faculty of Medicine, Atlanta, GA, USA
[5]Department of Internal Medicine and Critical Care, Lokman Hekim University, Ankara, Türkiye

**Submitted**
22.09.2021

**Revised**
12.12.2021

**Accepted**
04.04.2022

**Available Online**
14.10.2022

**Correspondence**
Kürşat Gündoğan, Erciyes University Faculty of Medicine, Department of Medicine, Division of Intensive Care, Kayseri, Türkiye
Phone: +90 352 207 66 66 /21919
e-mail: kgundogan@erciyes.edu.tr



ABSTRACT

**Objective:** Genistein has an anti-inflammatory effect that may be beneficial in many inflammatory diseases. The aim of this study was to determine the effects of supplementation of oral/enteral nutrition (EN) with genistein on the level of inflammatory cytokines in septic patients.

**Materials and Methods:** This prospective, randomized controlled study included critically ill adult patients with sepsis receiving EN or oral/EN. The patients were randomly divided into a genistein or a control group. Genistein (60 mg/day) was administered as a supplement to EN in the genistein group and the control group received only EN or oral/EN. Serum interleukin 1 beta (IL-1β), interleukin 6 (IL-6), high mobility group box 1 protein (HMGB-1), tumor necrosis factor alfa (TNF-α), procalcitonin (PCT), and C-reactive protein (CRP) concentrations were serially measured at the baseline, 24$^{th}$ hour, and 72$^{nd}$ hour after admission.

**Results:** Thirty-two participants (genistein group: 16 patients, control group: 16 patients) were included. The mean age was 56±17 years. The serum IL-1β concentration in the genistein group was significantly higher than that of the control group in follow-up (p=0.001). The control group had a significantly lower serum IL-6 value at the 72$^{nd}$ hour compared with the baseline and 24$^{th}$-hour values (p=0.001). The TNF-α concentration was significantly greater (p<0.001, both groups), while the PCT values were lower in follow-up measurements (genistein group: p=0.031; control group: p=0.004). The CRP level was higher in the genistein group than in the control group at the baseline (p=0.019) and significantly lower in follow-up measurements (p=0.028). At all of the study time points, the serum IL-6, TNF-α, HMGB-1, and PCT level of the genistein group was similar to that of the control group.

**Conclusion:** Genistein supplementation may add to the inflammation process and worsen the prognosis of sepsis patients in the acute period.

**Keywords:** Enteral nutrition, genistein, inflammatory cytokines, intensive care unit, sepsis

## INTRODUCTION

Sepsis is a clinical condition that develops in response to severe infection. Despite improvement in diagnosis and treatment, there is still a high risk of mortality. The incidence of sepsis is approximately 30% in intensive care unit (ICU) patients (1). Severe sepsis and septic shock continue to be a frequent cause of death in an ICU. The sepsis triad consists of systemic inflammation, coagulation, and impaired fibrinolysis. There is an exaggerated and irregular host response in the pathophysiology of sepsis involving microbial pathogens and the inflammatory response. Cytokines are released as a response to invading microorganisms (2, 3). Interleukin 1 beta (IL-1β) and tumor necrosis factor alfa (TNF-α) are released as early phase molecules, followed by cytokines such as interleukin 6 (IL-6) and high mobility group box 1 protein (HMGB-1). Studies have reported high serum IL-1β, IL-6, and TNF-α levels in many inflammatory diseases (cardiovascular disease, cancer, autoimmune diseases). The severity of the sepsis and peak levels of cytokines are associated with poorer outcomes (3–5).

Genistein (soy isoflavone) is derived from legumes and appears to be a promising multifunctional agent. It displays a broad range of anti-inflammatory, antioxidant, antiviral, and antibacterial effects. The primary mechanism for the anti-inflammatory effect of genistein is related to inhibition of nuclear factor kappa-B (NF-κB) and chemokine-8 (6, 7). TNF-α plays a crucial role in inflammatory diseases by activating NF-κB. NF-κB is described as a transcription factor. It also plays a decisive role in stimulating cell growth, survival, and apoptosis. It has been reported that genistein supplementation given to 6 healthy adult males inhibited NF-κB activation in peripheral lymph cells (8). The anti-inflammatory role of genistein has been demonstrated in preclinical studies, including a report that genistein could potentially contribute to preventing Alzheimer's disease (9). Various *in vitro* studies have reported that genistein may reduce the production or release of IL-6 and TNF-α (9, 10). Research has indicated that soy isoflavones may be beneficial in efforts to prevent and heal many chronic diseases, such as inflammatory diseases, cardiovascular disorders, osteoporosis, and cancer (11–13). Genistein supplementation studies are generally preclinical studies; to the best of our knowledge, there have been no previous clinical studies that have investigated the effect of genistein in patients with sepsis.

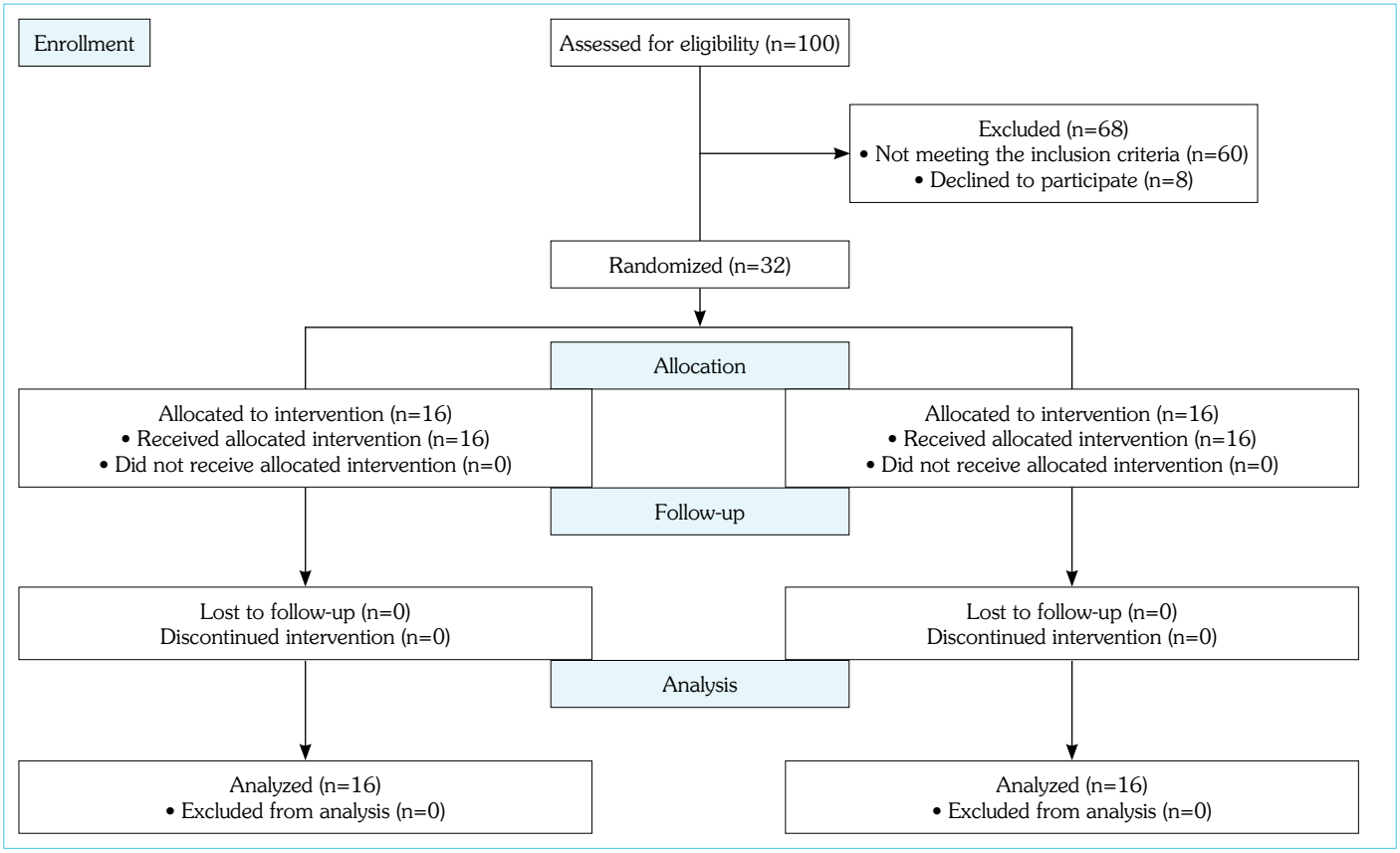


**Figure 1. Study enrollment and randomization**

It was hypothesized that the addition of genistein to enteral nutrition (EN) in critically ill patients with sepsis, including hyperinflammation, could play a valuable role in decreasing proinflammatory markers and improving the prognosis. Morbidity and mortality might be reduced with lower levels of proinflammatory cytokines in sepsis.

## MATERIALS and METHODS

### Ethical Considerations

The Erciyes University Clinical Research Ethics Committee approved this study. Written informed consent was obtained from all of the patients/their legal representatives before enrollment, and the study was registered with ClinicalTrials.gov, the international database provided by the US National Library of Medicine (no: NCT02796794).

### Study Design

This exploratory, single-center, open label, randomized controlled study was conducted in the 18-bed medical ICU of Erciyes University Hospital between June 2016 and June 2017. The inclusion criteria were age ≥18 years, diagnosis of sepsis (14) in the first 12 hours after admission to ICU, expectation of stay in the ICU of >48 hours, and use of EN (EN alone or oral+EN), Patients with thyroid dysfunction or hyperlipidemia were excluded.

Initial patient data were collected at the time of randomization. The patient demographic data, source of sepsis, and the primary reason for ICU admission were recorded upon study admission. The Acute Physiology and Chronic Health Evaluation II (APACHE II) score was calculated at 24 hours after ICU admission. The Sequential Organ Failure Assessment (SOFA) score was assessed at baseline (24$^{th}$ hour of sepsis diagnosis), and the 24$^{th}$ and 72$^{nd}$ hour. The need for renal replacement therapy and vasopressor treatment, days of mechanical ventilation (MV), length of ICU and hospital stay, and ICU mortality were also recorded. All of the patients received nutrition therapy according to recommendations of the European Society for Clinical Nutrition and Metabolism (ESPEN) and the American Society for Parenteral and Enteral Nutrition (ASPEN) guidelines for nutrition in ICU. Energy requirements were calculated at baseline, and the intake was recorded at baseline, and the 24$^{th}$ and 72$^{nd}$ hour.

### Study Randomization and Intervention

All of the study participants were randomly assigned in a 1:1 ratio to receive either 60 mg genistein (genistein group) or a placebo (control group) using the sealed envelope method.

### Outcome Measurements

Venous blood samples were drawn into an 8 mL sterile plain tube (without anticoagulant) before administering any medication (baseline), and at the 24$^{th}$ and 72$^{nd}$ hour to measure the level of inflammatory cytokines (IL-1β, IL-6, HMGB-1 protein, and TNF-α), procalcitonin (PCT), and C-reactive protein (CRP). The blood samples were centrifuged at 3000 g for 10 minutes, and the serum was collected. The samples were stored at -40 °C until the biochemical analyses were to be performed and then brought to room temperature. Serum TNF-α (cat. no: EH0302), IL-1β (cat. no: EH0185), IL-6 (cat. no: EH0201), and HMGB-1 (cat. no: EH0884) levels were measured according to the manufacturer's instructions using commercially available enzyme-linked immunosorbent assay kits (FineTest Human Elisa Kits, Wuhan Fine Biotech Co., Ltd. Wuhan, Hubei, China).

**Table 1.** Patient demographic and clinical characteristics

| Variables | Total (N=32) | Genistein group (n=16) | Control group (n=16) | p |
|---|---|---|---|---|
| Age (mean±SD) (years) | 56±17 | 54±16 | 59±17 | 0.698 |
| Gender, n (%) | | | | 0.238 |
| Male | 23 (72) | 10 (63) | 13 (37) | |
| Female | 9 (28) | 6 (81) | 3 (19) | |
| Reason for ICU admission, n (%) | | | | 0.142 |
| Respiratory failure | 17 (53) | 11 (65) | 6 (35) | |
| Septic shock | 9 (28) | 2 (22) | 7 (78) | |
| Postoperative | 1 (3) | 1 (100) | 0 (0) | |
| Other | 5 (16) | 2 (40) | 3 (60) | |
| Source of sepsis, n (%) | | | | 0.421 |
| Respiratory | 18 (56) | 8 (44) | 10 (56) | |
| Gastrointestinal system | 6 (19) | 5 (83) | 1 (17) | |
| Genitourinary system | 4 (13) | 1 (25) | 3 (75) | |
| Catheter | 2 (6) | 1 (50) | 1 (50) | |
| Other | 2 (6) | 1 (50) | 1 (50) | |
| APACHE II score (mean±SD) | 16±9 | 17±9 | 16±8 | 0.725 |
| SOFA score (mean±SD) | | | | |
| Baseline | 5±3 | 5±3 | 5±3 | 0.743 |
| 24$^{th}$ hour | 5±3 | 5±3 | 4±3 | 0.998 |
| 72$^{nd}$ hour | 4±3 | 5±3 | 4±2 | 0.322 |
| Need for renal replacement therapy, n (%) | 8 (25) | 6 (75) | 2 (25) | 0.096 |
| Need for vasopressor therapy, n (%) | 8 (25) | 5 (63) | 3 (37) | 0.412 |
| Duration of mechanical ventilation, median (min–max) (days) | 8 (1–117) | 13 (6–117) | 7 (1–12) | **0.037** |
| Length of ICU stay, median (min–max) (days) | 7.5 (3–117) | 10.5 (3–117) | 6.5 (3–24) | 0.232 |
| Length of hospital stay, median (min–max) (days) | 13.5 (5–117) | 20.5 (5–117) | 12.5 (5–30) | 0.192 |
| ICU mortality, n (%) | 12 (37.5) | 9 (56) | 3 (44) | **0.028** |

APACHE II: Acute Physiology and Chronic Health Evaluation II; ICU: Intensive care unit; SOFA: Sequential Organ Failure Assessment; SD: Standard deviation

The serum PCT level was measured using the electrochemiluminescence immunoassay method on a Cobas e801 autoanalyzer (Roche Diagnostics, Basel, Switzerland). The serum CRP concentration was analyzed using a Siemens Dade Behring BN II nephelometer (Siemens Healthineers GmbH, Erlangen, Germany). Microorganism growth was recorded from routine culture analyses during ICU follow-up.

### Statistical Analysis

All of the data were analyzed with IBM SPSS Statistics for Windows, Version 22.0 software (IBM Corp., Armonk, NY, USA). Continuous data were presented as the mean±SD or the median (min–max). The Shapiro-Wilk test was performed to test normality. Comparisons between 2 groups of continuous variables were conducted using Student's t-test for normally distributed data and the Mann-Whitney U test for data with a non-normal distribution.

Two-way repeated measures analysis of variance (ANOVA) was conducted to evaluate the effect of time (baseline, 24$^{th}$, and 72$^{nd}$ hour) and group interaction between variables. The Friedman test was used to evaluate the effect of time for non-normally distributed variables. A chi-squared test or Fischer's exact test was used to determine significant differences between categorical variables. A p value of <0.05 was accepted as statistically significant.

## RESULTS

### Baseline Characteristics

In all, 100 patients were screened; 68 were ineligible, and 32 were enrolled. The patients accepted into the study were divided into 2 equal groups: 16 participants in the genistein group and 16 participants in the control group (Fig. 1). The demographic and clinical data of the patients are provided in Table 1. The mean age of the sample was 56±17 years. The most common reasons for ICU admission were respiratory failure in 17 patients (53.0%) and septic shock in 9 patients (28.0%).

### Primary Outcome

Repeated measures ANOVA results revealed no statistically significant interaction according to group or time in the serum IL-1β concentration (p=0.318). During the follow-up period, there was

**Table 2.** Inflammatory cytokine levels during follow-up

| Variables | Total (N=32) | Genistein group (n=16) | Control group (n=16) | p* (group) | p* (time) | p* (int.) |
|---|---|---|---|---|---|---|
| IL-1β (mean±SD) (pg/mL) | | | | **0.001** | 0.140 | 0.318 |
| Baseline | 31.4±11.5 | 39.7±7.7[a] | 23.0±8.1[b] | | | |
| 24th hour | 38.2±16.0 | 41.4±14.6[a] | 34.9±17.1[b] | | | |
| 72nd hour | 33.1±16.5 | 37.6±17.6[a] | 28.6±14.4[b] | | | |
| IL-6 (mean±SD) (pg/mL) | | | | 0.167 | **0.001** | **<0.001** |
| Baseline | 28.9±13.3 | 29.5±17.8[A] | 28.2±7.1[A] | | | |
| 24th hour | 32.0±15.7 | 28.4±10.6[A] | 35.6±19.3[A] | | | |
| 72nd hour | 21.2±15.3 | 31.6±14.9[A] | 10.8±5.3[B] | | | |
| TNF-α (mean±SD) (pg/mL) | | | | 0.092 | **<0.001** | 0.025 |
| Baseline | 68.1±11.7 | 68.3±12.3[A] | 67.7±11.3[A] | | | |
| 24th hour | 76.1±16.0 | 74.5±13.7[AB] | 77.7±18.3[A] | | | |
| 72nd hour | 95.4±21.6 | 86.3±15.1[B] | 104.6±23.6[B] | | | |
| HMGB-1 (mean±SD) (pg/mL) | | | | 0.902 | 0.640 | 0.451 |
| Baseline | 68.9±38.9 | 67.4±35.8 | 70.4±42.9 | | | |
| 24th hour | 73.2±35.6 | 78.5±40.5 | 67.8±30.4 | | | |
| 72nd hour | 74.8±41.9 | 73.1±3.2 | 76.4±41.9 | | | |
| | | | | | **p** (group)** | |
| Procalcitonin (median [min–max]) (ng/mL) | | | | | | |
| Baseline | 5.9 (0.05–119) | 5.5 (0.10–119)[A] | 6.5 (0.05–114)[A] | | 0.925 | |
| 24th hour | 3.7 (0.05–200) | 4.3 (0.07–200)[AB] | 3.8 (0.05–80)[AB] | | 0.940 | |
| 72nd hour | 2.1 (0.05–200) | 2.5 (0.07–200)[B] | 1.3 (0.05–12)[B] | | 0.298 | |
| p*** (time) | | **0.031** | **0.004** | | | |
| CRP (median [min–max]) | | | | | | |
| Baseline | 169.5 (5–380) | 201.5 (76–380)[aA] | 131 (5–251)[b] | | 0.019 | |
| 24th hour | 149 (4–413) | 154.5 (14–413)[AB] | 141.5 (4–338) | | 0.250 | |
| 72nd hour | 118 (3–316) | 144 (31–316)[B] | 89.5 (3–206) | | 0.105 | |
| p*** (time) | | **0.028** | 0.052 | | | |

*: Repeated measure two-way analysis of variance; **: Mann-Whitney U test; ***: Friedman test; a,b,c… lowercase and A,B,C… uppercase letters indicate the Bonferroni post-hoc test results for groups and time points, respectively. Letters indicate the difference between groups and time points. CRP: C-reactive protein; HMGB-1: High mobility group box 1 protein; IL1-β: Interleukin 1 beta; IL-6: Interleukin 6; TNF-α: Tumor necrosis factor alpha; SD: Standard deviation

no statistically significant difference between time points in the 2 groups (p=0.140). The serum IL-1β level in the genistein group was significantly higher than that of the control group at all time points (p=0.001) (Table 2).

Statistically significant differences were observed in ANOVA results for serum IL-6 concentration in group*time interaction analysis (p<0.001). The genistein group had a similar serum level of IL-6 to that of the control group at all time points (p=0.167). However, the serum IL-6 concentration of the control group at 72 hours was significantly lower than the values seen at baseline and the 24th hour after admission (p=0.001) (Table 2).

Also shown in Table 2, repeated measures ANOVA results yielded a significant group*time interaction for the serum TNF-α concentration (p=0.025). During follow-up, there was no statistically significant difference between the genistein group and the control group (p=0.092). The serum TNF-α concentration was significantly elevated during follow-up in both groups (p<0.001).

The results revealed no significant group*time interaction for serum HMGB-1 protein values (p=0.451). The HMGB-1 protein level was similar in both groups (p=0.902) and did not change during follow-up (p=0.640) (Table 2).

The PCT value did not present a significant difference between groups at any time point of the study (p=0.925, p=0.940, and p=0.298). The serum PCT level was lower during follow-up in both the genistein and the control group (genistein group: p=0.031; control group: p=0.004).

**Table 3.** Microbiological culture findings

| Type of culture | Genistein group | Control group |
|---|---|---|
| Blood | *Staphylococcus epidermidis* (n=5) | *Staphylococcus epidermidis* (n=5) |
| | *Escherichia coli* (n=1) | *Diphtheroid bacilli* (n=1) |
| | *Klebsiella pneumoniae* (n=1) | *Escherichia coli* (n=1) |
| | *Pseudomonas aeruginosa* (n=1 | *Pseudomonas aeruginosa* (n=1) |
| | *Streptococcus sanguineous* (1) | |
| Catheter | *Staphylococcus epidermidis* (n=4) | *Staphylococcus epidermidis* (n=2) |
| | *Escherichia coli* (n=1) | |
| | *Klebsiella pneumoniae* (n=2) | |
| | *Acinetobacter baumannii* (n=1) | |
| | *Enterococcus faecalis* (n=1) | |
| | *Staphylococcus aureus* (n=3) | |
| Respiratory secretions | *Staphylococcus epidermidis* (n=1) | Yeast (n=2) |
| | *Acinetobacter baumannii* (n=2) | *Haemophilus influenzae* (n=1) |
| | *Enterococcus faecalis* (n=1) | |
| | Non-hemolytic streptococci (n=1) | |
| | *Pseudomonas aeruginosa* (n=3) | |
| | *Stenotrophomonas maltophilia* (n=1) | |
| Urine | *Escherichia coli* (n=4) | Yeast (n=1) |
| | *Klebsiella pneumoniae* (n=2) | *Klebsiella pneumoniae* (n=2) |
| | *Pseudomonas aeruginosa* (n=1) | *Polymicrobial* (n=1) |
| | *Acinetobacter baumannii* (n=1) | |
| | *Enterobacter species* (n=1) | |
| | Yeast (n=3) | |
| Wound site | *Staphylococcus epidermidis* (n=1) | Yeast (n=1) |
| | *Escherichia coli* (n=1) | |
| | *Klebsiella pneumoniae* (n=1) | |
| | *Staphylococcus aureus* (n=1) | |

The genistein group had higher CRP values than the control group at baseline (201.5 mg/L [range: 76–380 mg/L] vs. 131 mg/L [range: 5–251 mg/L]; p=0.019). The CRP values were significantly lower at the 24$^{th}$ hour and the 72$^{nd}$ hour in the genistein group compared with baseline measurements (p=0.028) (Table 2).

### Secondary Outcomes

While the SOFA score decreased significantly in the control group over time (p=0.005), there was no statistically significant difference observed in the genistein group (p=0.216).

The number of MV days was significantly greater in the genistein group (p=0.037), and the genistein group had a higher mortality rate than the control group (56% vs. 44%) (p=0.028) (Table 1).

Microorganisms grown from cultures taken during ICU follow-up of patients revealed 46 microorganisms in the genistein group and 18 microorganisms in the control group. The most common microorganisms were *Staphylococcus epidermidis* (n=11, 69%), *Escherichia coli* (n=7, 44%), and *Klebsiella pneumoniae* (n=6, 38%) in the genistein group, and *Staphylococcus epidermidis* (n=7, 44%) and yeast (n=4, 25%) in the control group (Table 3).

## DISCUSSION

Our results indicated that patients receiving genistein supplementation had a significantly higher serum IL-1β level during follow-up and a higher CRP level at baseline than the control group. Compared with the baseline and 24$^{th}$-hour measurements, the control group had a significantly lower serum IL-6 value at 72 hours. The TNF-α concentration was significantly higher and the PCT and CRP levels were lower in both groups during follow-up. No significant difference was observed in the serum IL-6, TNF-α, HMGB-1 and PCT levels between the groups.

There is only limited research of genistein use in humans and rats with sepsis. To our knowledge, there have been no studies that demonstrated the effect of genistein on inflammation in patients with sepsis in an ICU. Previous studies have found that genistein reduced inflammatory parameters in chronic inflammatory diseases such as cancer and cardiovascular diseases (15–19).

TNF-α, IL-1β, and IL-6 are released from monocytes in sepsis. These cytokines, which contribute significantly to the fight against local infection, are synthesized in large quantities and cause wide-

spread endothelial cell damage. *In vitro* and rat studies have noted that genistein may decrease macrophage and TNF-α levels in inflammatory diseases by inhibiting NF-κB activation (13, 20, 21). In contrast, in a study on birds injected with *Escherichia coli* and given genistein, the TNF-α and IL-6 levels in the genistein group increased (22). In our study, inflammatory cytokines, one of the components of sepsis, were generally higher in the genistein group during follow-up.

A strong correlation exists between uncontrolled inflammation and both CRP and PCT. They are considered descriptive biomarkers that can show the prognosis of sepsis (23, 24). In this study, the PCT level in the control group decreased more than that of the genistein group. The CRP level at baseline was higher in the genistein group and showed a smaller decrease than that of the control group.

Unlike experimental studies, our results revealed a poorer prognosis for the inflammatory process in the genistein group in comparison with the control group. One possible explanation may be the size of the dose of genistein given to patients. It was previously reported that the dose we used in our study did not have adverse effects, and was noted as the optimal dose in another study (25, 26). The optimal dose may be higher in septic patients.

Sepsis is likely to be associated with a poor prognosis in ICU patients, including morbidity and mortality (3).The baseline SOFA score of the 2 groups was similar, but while the SOFA score improved in the control group during the follow-up period, there was no significant change in the genistein group. This raises the question that the administration of genistein in the acute phase of sepsis may have a negative effect on morbidity. It may be important to consider the clinical implications.

The need for MV may increase due to endothelial damage caused by sepsis-associated inflammatory cytokines. The results of this study indicated that the genistein group had a longer MV duration than the control group. However, other research of the effect of genistein on airway inflammation have yielded different results. Guinea pigs were given intraperitoneal ovalbumin to induce bronchospasm and the administration of genistein (15 mg/kg intraperitoneal) markedly inhibited ovalbumin-induced acute bronchoconstriction. Inhibition of the tyrosine kinase signaling pathway, which has a vital role in the activation of inflammatory cells, produced a therapeutic result that could potentially be useful (19).

Genistein is known as an immunomodulatory agent that may reduce morbidity and mortality in inflammatory diseases, cardiovascular diseases, and cancer (12, 27). It could be considered an immunonutrient. Two important studies of immunonutrient supplementation for critically ill patients are the work of Heyland et al. (28), who reported that the use of anti-inflammatory immunonutrients (glutamine, omega-3 fatty acids, selenium, and antioxidants) increased mortality in 1223 critical patients (378 patients, 30.9% diagnosed with sepsis), and the findings of Van Zanten et al. (29), who found that among 301 critically ill patients (22% diagnosed with sepsis), mortality was higher among those given immunonutrients (glutamine, omega-3 fatty acid, and antioxidant) orally/enterally in medical ICUs. Similarly, ICU mortality was higher in the group that received genistein in our study. The adverse effect on inflammatory processes, the morbidity rate, and the greater frequency of microorganisms in patients receiving genistein were factors contributing to mortality. This study presents intriguing results on the use of genistein in patients with sepsis. However, the research has some limitations to consider. The study sample was a small group from a single medical ICU. In addition, genistein supplementation was administered to our study subjects for a short period of time. Additional research could prove valuable.

## CONCLUSION

Genistein has been evaluated for a protective role in many diseases and disorders, and it continues to be a compound of interest that may still have benefits yet to be evaluated and understood. This research revealed new information that genistein supplementation for sepsis patients in the acute period may harm the inflammation process and worsen the prognosis. However, additional research to fully identify the mechanism may provide valuable new information.

**Ethics Committee Approval:** The Erciyes University Clinical Research Ethics Committee granted approval for this study (date: 23.05.2014, number: 2014/341).

**Informed Consent:** Written informed consent was obtained from patients who participated in this study.

**Peer-review:** Externally peer-reviewed.

**Author Contributions:** Concept – GE, KG, İG, ŞT, NTÖ, HS, MS, CY, ÖK; Design – GE, KG, İG, ŞT, NTÖ, HS, MS, CY, ÖK; Supervision – GE, KG, İG, ŞT, NTÖ, HS, MS, CY, ÖK; Resource – MS; Materials – GE, KG, İG, CY; Data Collection and/or Processing – GE, KG, HS, ŞT, MS; Analysis and/or Interpretation – İG, CY; Literature Search – KG, NTÖ; Writing – GE, KG, NTÖ; Critical Reviews – KG, NTÖ, ŞT, MS.

**Conflict of Interest:** The authors have no conflict of interest to declare.

**Financial Disclosure:** The authors declared that this study has received no financial support.

## REFERENCES

1. Sakr Y, Jaschinski U, Wittebole X, Szakmany T, Lipman J, Ñamendys-Silva SA, et al. Sepsis in intensive care unit patients: worldwide data from the intensive care over nations audit. Open Forum Infectious Dis 2018; 5(12): ofy313. [CrossRef]
2. Faix JD. Biomarkers of sepsis. Crit Rev Clin Lab Sci 2013; 50(1): 23–36. [CrossRef]
3. Chousterman BG, Swirski FK, Weber GF. Cytokine storm and sepsis disease pathogenesis. Semin Immunopathol 2017; 39(5): 517–28.
4. Gouel-Chéron A, Allaouchiche B, Guignant C, Davin F, Floccard B, Monneret G; AzuRea Group. Early interleukin-6 and slope of monocyte human leukocyte antigen-DR: a powerful association to predict the development of sepsis after major trauma. PLoS One 2012; 7(3): e33095. [CrossRef]
5. Riedemann NC, Guo RF, Hollmann TJ, Gao H, Neff TA, Reuben JS, et al. Regulatory role of C5a in LPS-induced IL-6 production by neutrophils during sepsis. FASEB J 2004; 18(2): 370–2. [CrossRef]
6. Smolińska E, Moskot M, Jakóbkiewicz-Banecka J, Węgrzyn G, Banecki B, Szczerkowska-Dobosz A, et al. Molecular action of isoflavone genistein in the human epithelial cell line HaCaT. PLoS One 2018; 13(2): e0192297. [CrossRef]

7. Kim JM, Uehara Y, Choi YJ, Ha YM, Ye BH, Yu BP, et al. Mechanism of attenuation of pro-inflammatory Ang II-induced NF-κB activation by genistein in the kidneys of male rats during aging. Biogerontology 2011; 12(6): 537–50. [CrossRef]
8. Davis JN, Kucuk O, Sarkar FH. Genistein inhibits NF-kappa B activation in prostate cancer cells. Nutr Cancer 1999; 35(2): 167–74. [CrossRef]
9. Valles SL, Dolz-Gaiton P, Gambini J, Borras C, Lloret A, Pallardo FV, et al. Estradiol or genistein prevent Alzheimer's disease-associated inflammation correlating with an increase PPAR gamma expression in cultured astrocytes. Brain Res 2010;1312: 138–44. [CrossRef]
10. Hämäläinen M, Nieminen R, Vuorela P, Heinonen M, Moilanen E. Anti-inflammatory effects of flavonoids: genistein, kaempferol, quercetin, and daidzein inhibit STAT-1 and NF-kappaB activations, whereas flavone, isorhamnetin, naringenin, and pelargonidin inhibit only NF-kappaB activation along with their inhibitory effect on iNOS expression and NO production in activated macrophages. Mediators Inflamm 2007; 2007: 45673. [CrossRef]
11. Vitale DC, Piazza C, Melilli B, Drago F, Salomone S. Isoflavones: estrogenic activity, biological effect and bioavailability. Eur J Drug Metab Pharmacokinet 2013; 38(1): 15–25. [CrossRef]
12. Messina M. Soy and health update: Evaluation of the clinical and epidemiologic literature. Nutrients 2016; 8(12): 754. [CrossRef]
13. Verdrengh M, Jonsson IM, Holmdahl R, Tarkowski A. Genistein as an anti-inflammatory agent. Inflamm Res 2003; 52(8): 341–6. [CrossRef]
14. Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, et al. The third international consensus definitions for sepsis and septic shock (Sepsis-3). JAMA 2016; 315(8): 801–10. [CrossRef]
15. Lazarevic B, Hammarström C, Yang J, Ramberg H, Diep LM, Karlsen SJ, et al. The effects of short-term genistein intervention on prostate biomarker expression in patients with localised prostate cancer before radical prostatectomy. Br J Nutr 2012; 108(12): 2138–47. [CrossRef]
16. Yan L, Spitznagel EL. Soy consumption and prostate cancer risk in men: a revisit of a meta-analysis. Am J Clin Nutr 2009; 89(4): 1155–63.
17. Lazarevic B, Boezelijn G, Diep LM, Kvernrod K, Ogren O, Ramberg H, et al. Efficacy and safety of short-term genistein intervention in patients with localized prostate cancer prior to radical prostatectomy: a randomized, placebo-controlled, double-blind Phase 2 clinical trial. Nutr Cancer 2011; 63(6): 889–98. [CrossRef]
18. Bime C, Wei CY, Holbrook J, Smith LJ, Wise RA. Association of dietary soy genistein intake with lung function and asthma control: a post-hoc analysis of patients enrolled in a prospective multicentre clinical trial. Prim Care Respir J 2012; 21(4): 398–404. [CrossRef]
19. Duan W, Kuo IC, Selvarajan S, Chua KY, Bay BH, Wong WS. Antiinflammatory effects of genistein, a tyrosine kinase inhibitor, on a guinea pig model of asthma. Am J Respir Crit Care Med 2003; 167(2): 185–92. [CrossRef]
20. Vanden Braber NL, Novotny Nuñez I, Bohl L, Porporatto C, Nazar FN, Montenegro MA, et al. Soy genistein administered in soluble chitosan microcapsules maintains antioxidant activity and limits intestinal inflammation. J Nutr Biochem 2018; 62: 50–8. [CrossRef]
21. Liu FC, Wang CC, Lu JW, Lee CH, Chen SC, Ho YJ, et al. Chondroprotective effects of genistein against osteoarthritis ınduced joint ınflammation. Nutrients 2019; 11(5): 1180. [CrossRef]
22. Zhang M, Kou J, Wu Y, Wang M, Zhou X, Yang Y, et al. Dietary genistein supplementation improves intestinal mucosal barrier function in Escherichia coli O78-challenged broilers. J Nutr Biochem 2020; 77: 108267. [CrossRef]
23. Wacker C, Prkno A, Brunkhorst FM, Schlattmann P. Procalcitonin as a diagnostic marker for sepsis: a systematic review and meta-analysis. Lancet Infect Dis 2013; 13(5): 426–35. [CrossRef]
24. Cui N, Zhang H, Chen Z, Yu Z. Prognostic significance of PCT and CRP evaluation for adult ICU patients with sepsis and septic shock: retrospective analysis of 59 cases. J Int Med Res 2019; 47(4): 1573–9.
25. Ullmann U, Metzner J, Frank T, Cohn W, Riegger C. Safety, tolerability, and pharmacokinetics of single ascending doses of synthetic genistein (Bonistein) in healthy volunteers. Adv Ther 2005; 22(1): 65–78.
26. Ferrari A. Soy extract phytoestrogens with high dose of isoflavones for menopausal symptoms. J Obstet Gynaecol Res 2009; 35(6): 1083–90.
27. Mukund V, Mukund D, Sharma V, Mannarapu M, Alam A. Genistein: Its role in metabolic diseases and cancer. Crit Rev Oncol Hematol 2017; 119: 13–22. [CrossRef]
28. Heyland D, Muscedere J, Wischmeyer PE, Cook D, Jones G, Albert M, et al; Canadian Critical Care Trials Group. A randomized trial of glutamine and antioxidants in critically ill patients. N Engl J Med 2013; 368(16): 1489–97. Erratum in: N Engl J Med 2013; 368(19): 1853.
29. van Zanten AR, Sztark F, Kaisers UX, Zielmann S, Felbinger TW, Sablotzki AR, et al. High-protein enteral nutrition enriched with immune-modulating nutrients vs standard high-protein enteral nutrition and nosocomial infections in the ICU: a randomized clinical trial. JAMA 2014; 312(5): 514–24. [CrossRef]