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Effects of β-glucan and vitamin C on immunological and antioxidant serum parameters in rats administered with oral capecitabine

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ABSTRACT

Aim: Based main purpose of the study to evaluate the effect of β-glucan and vitamin C on antioxidant serum enzymes superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx), immunoglobulin, an antibody and the important cytokines interleukin-1β (IL-1β) and tumor necrosis factor-α (TNF-α) in rats administered capecitabine.

Method and materials: In this study, 35 Sprague Dawley rats were divided into 5 groups of seven. Tween 20 (5 ml/kg body weight/day) to the first group (Group I), 359 mg/kg body weight/day capecitabine to the second group (Group II), 100 mg/kg body weight/day β-glucan in addition to capecitabine to the third group (Group III), 100 mg/kg body weight/day vitamin C in addition to capecitabine to the fourth group (Group IV), and β-glucan and vitamin C in addition to capecitabine to the fifth group (Group V) were orally given by gastric tube for 14 days. Sera obtained from blood taken from rats on the 14th and 21st days of the experiment were stored at -80 ˚C.

Results: Serum IgG concentrations were significantly higher in Group II given capecitabine on day 14 of blood collection than in other groups. On day 21, IL-1β levels in Groups IV and V were significantly higher than Groups I and II, while IL-1β levels between Groups I, II and III were not significantly different. On the 14th blood draw day, SOD activities in Groups III, IV and V were found to be significantly lower than Groups I and II. On the 14th day of the study, serum CAT activities were found to be significantly lower in Group II given capecitabine than Group IV and V, while the levels in Group IV were found to be significantly higher than the control group (Group I). Again, on the 14th day of the study, a statistically significant difference in serum GPx activities was detected between the control group (44.3 ± 2.64 ng/ml) and Group III (28.7 ± 11.34 ng/ml). On the 14th and 21st days of the study, TNF-α concentrations were found to be significantly lower in the control group (Group I) than in the other groups.

Conclusion: It was concluded that the immunosuppressive feature of chemotherapy in this study caused certain decreases in the levels of antioxidant enzymes, as expected. It seems that chemotherapy suppressed the production of all antioxidant enzymes, but this was different for some cytokines and antibodies. It was observed that therapy with β-glucan, vitamin C and a combination of both showed different effects for each parameter or did not have a specific effect.

Keywords: Capecitabine, β-Glucan, vitamin C.

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Introduction

Millions of animals and people die every year from cancer which is one of most important health problems of our time. Many chemotherapy drugs or drugs developed for molecular targets are used in treatment of this disease. Capecitabine is a chemotherapy drug that is a precursor of 5-fluorouracil with high tumor selectivity.

Capecitabine is a chemotherapy drug that is a precursor of 5-fluorouracil with high tumor selectivity. Like many chemotherapy drugs, capecitabine has been reported to cause immunosuppression in cancer patients. A weakened immune system increases the risk of cancer patients getting infections. In addition, infections, which are considered insignificant for healthy people, are very severe in cancer patients due to the suppressed immune system. For this reason, in addition to recent chemotherapy studies, additional treatment studies aimed at strengthening the immune system have been directed.

For many years, ascorbic acid (vitamin C) has already been used as an immune booster alongside chemotherapy. In recent years, as the positive effects of glucans on the immune system have begun to be discovered, many studies have been carried out in the direction of being an additional or alternative treatment in cancer treatments. The levels of antioxidant enzymes superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx), which play a decisive role in detoxifying the reactive toxic metabolites of many toxins, decrease due to chemotherapy. In studies with 5-fluorouracil combined treatments with molecules with antioxidant properties such as carboxymethyl pachyman, it has been observed that the decreased levels of antioxidant enzymes are significantly restored (Wang *et al*., 2017). At the end of a study on rats with Poria cocos mushrooms, which is one of the medical mushrooms and contains 90% β-glucan, it was determined that both the tumor size decreased and the activities of antioxidant enzymes increased (RuiDian *et al*., 2010).

Based on numerous studies to reduce the negative effects of cancer and chemotherapy on immunity, in this study, the antioxidant serum enzymes SOD, CAT and GPx, immunoglobulin, an antibody, and the important cytokines interleukin-1β (IL-1β) and tumor necrosis factor-α (TNF-α), were investigated to see how β-glucan and vitamin C, which have immune-enhancing and antioxidant properties, affect suppressed immunity in rats given capecitabine.

Materials and Methods

In the study, 35 female/male Sprague Dawley rats, aged 9-15 months, with Experimental Animals Ethics Committee Approval from Istanbul University (No: 2017-22467), were used. The experimental part of the study was carried out at Boğaziçi University, Life Sciences and Technologies Application and Research Center, Experimental Animal Production and Care Unit. The care and feeding of the rats were also done in the same center. Rats were fed *ad libitum* with standard commercial rat chow and water at 21 ˚C, 12 hours light and 12 hours dark cycle.

Rats were labeled and randomly divided into 5 groups of seven. Different combinations of capecitabine, β-glucan and vitamin C were given orally to the other 4 groups, one of which was a control. Trial groups and dose amounts were recorded (Table 1).

Table 1. Trial groups.

	Group 1	Group 2	Group 3	Group 4	Group 5
Tween 20 (10%) (5 ml/kg/day)	✓				
Capecitabine (359 mg/kg/day)		✓	✓	✓	✓
β-Glucan (100 mg/kg/day)			✓		✓
Vitamin C (100 mg/kg/day)				✓	✓

All chemicals were administered to rats by gavage together with a 10% solution of Tween 20 (Sigma-Aldrich). First, each animal was weighed. The dose amounts to be given individually according to the weight of the animals were determined. After capecitabine (Xeltabin 500 mg film-coated tablet, TEVA, India) is pounded in a mortar, after β-glucan (Imuneks, 50 mg capsule, Mustafa Nevzat Pharmaceutical Industry Corporation, Istanbul) is extracted from the capsules, and vitamin C (L-ascorbic acid, Sigma-Aldrich, Germany) using as is, the daily amounts of each individual in a sensitive scale were weighed, put into labeled tubes and made ready for application.

As indicated, capecitabine, β-glucan and vitamin C were administered to the groups for 14 days. After the last dose was given on the 14th day, blood was drawn from the rats at the end of the day. Blood collection was performed by bleed from the jugular vein without anesthesia of the rats. The rats were kept in their normal environment without any chemicals until the end of the 21st day. At the end of the 21st day, the rats were killed by isoflurane. Cardiac blood collection method was used, all rats were sacrificed and the trial phase of the study was terminated.

After the blood samples were taken on the 14th and 21st days of the experiment, they were kept in the refrigerator at +4 ˚C for 16 hours to clot. At the end of this period, they were centrifuged at 2000 rpm for 15 minutes. At the end of the centrifugation, the pellet part of the blood settled to the bottom. The sera, which were the supernatant part that appeared transparent in the tubes, were carefully taken into pre-labeled eppendorf serum storage tubes without mixing with the pellet part with the help of a micropipette. Since it will be

reworked on different days and not to freeze-thaw, the obtained serums were divided into small amounts and distributed into storage tubes. Prepared serum samples were stored at -80 ˚C until the day of the kits to be used.

After the serums were obtained, ELISA kits were used to observe the changes in immunological and antioxidant serum parameters, respectively. At the end of each application, the analysis was completed by measuring the optical density (OD) values at 450 nm with a microplate reader (Moleculer Devices, San Jose, CA). IgG antibody and IL-1β cytokine determinations using rat IgG ELISA (Cat. No: E-EL-R0518) and rat IL-1β ELISA (Cat. No: E-EL-R0012) kits from Elabscience (Wuhan, China), the determination of CAT, SOD and GPx antioxidant enzymes using rat CAT ELISA (Cat. No: 201-11-5106), rat SOD ELISA (Cat. No: 201-11-0169) and rat GPx ELISA (Cat. No: 201-1-1705) from SunRed (Shanghai, China), and TNF-α cytokine determination using rat TNF-α Platinum ELISA Kit (Cat. No: BMS622) from Affymetrix eBioscience (Vienna, Austria) were made in sera. The operation of each of these kits was based on the sandwich ELISA principle.

Statistical tests were applied to each immunological and antioxidant serum parameter separately in order to examine whether the data obtained as a result of ELISA were statistically significant. First of all, one-way analysis of variance (ANOVA) test was applied to each parameter. This test was a tool used to test whether there was a statistically significant difference between the means of independent groups (Sthle and Wold, 1989). As a result of this test, the significant differences between the groups of each parameter were examined. Then, the data obtained from the groups on the 14th and 21st days were analyzed statistically by the Tukey test, which was a multiple comparison test between the groups of each day. Values were lettered according to their significant or insignificant differences between each other and appropriate statistical tables were prepared. Finally, using Student's t test, both blood collection days were compared with each other and analyzed statistically.

Results and Discussion

The comparison with each other of 5 groups in each blood collection day and the comparison with each other of 2 blood collection days within each group were recorded (Table 2) in terms of serum immune and antioxidant enzyme parameters.

Serum IgG concentrations were significantly higher in Group II given capecitabine on day 14 of blood collection than in other groups. On the 21st day, the concentrations were found to be significantly higher in Group II given capecitabine than in the other 3 groups except the control group. On day 21, no significant differences were found between the control group and the other groups. There were no significant changes in IgG levels between the two blood draws in either of the 5 groups. There were no significant differences between the groups in terms of serum IL-1β concentrations on the 14th day of chemotherapy. On the 21st day, IL-1β levels in Group IV and Group V were found to be significantly higher than Groups I and II, while IL-1β levels between Groups I, II and III were not found to be significantly different.

Table 2. The comparison with each other of 5 groups in each blood collection day and the comparison with each other of 2 blood collection days within each group, in terms of concentrations of serum immune and antioxidant enzyme parameters (n=7).

Indices	Days	Group I	Group II	Group III	Group IV	Group V
IgG (ng/ml)	Day 14	3.30 ± 1.32^{a}	8.87 ± 2.27^{b}	3.42 ± 1.86^{a}	5.39 ± 1.75^{a}	2.63 ± 1.14^{a}
	Day 21	4.71 ± 0.53^{ab}	7.80 ± 3.56^{a}	2.56 ± 0.94^{b}	3.59 ± 2.11^{b}	3.73 ± 1.39^{b}
IL-1β (pg/ml)	Day 14	638 ± 181.0*	518 ± 82.3*	568 ± 103.4*	518 ± 136.5	405 ± 95.27
	Day 21	268 ± 66.4^{a}*	255 ± 58.5^{a}*	340 ± 89.7^{ac}*	491 ± 135.9^{bc}	518 ± 122.8^{b}
TNF-α (pg/ml)	Day 14	30.2 ± 13.6^{a}	211.2 ± 64.8^{b}	190.0 ± 30.6^{b}*	165.1 ± 42.4^{b}*	143.2 ± 47.7^{b}
	Day 21	19.6 ± 12.0^{a}	130.8 ± 37.7^{b}	112.5 ± 43.6^{b}*	97.2 ± 24.0^{b}*	90.1 ± 16.9^{b}
SOD (ng/ml)	Day 14	42.3 ± 1.33^{a}	38.1 ± 3.58^{ab}*	35.8 ± 2.39^{b}	35.1 ± 2.18^{b}*	37.3 ± 4.70^{b}
	Day 21	34.2 ± 4.16^{ab}	32.5 ± 2.22^{a}*	37.0 ± 3.36^{b}	38.5 ± 1.90^{b}*	37.4 ± 1.51^{b}
CAT (ng/ml)	Day 14	164.7 ± 6.2^{bc}	154.3 ± 20.2^{c}	172,8 ± 14.9^{ac}	194.0 ± 16.5^{a}	185.0 ± 14.4^{ab}
	Day 21	161.6 ± 28.2	163.0 ± 22.9	164.9 ± 20.3	177.5 ± 10.3	179.6 ± 14.5
GPx (ng/ml)	Day 14	44.3 ± 2.64^{a}*	35.6 ± 5.18^{ab}	28.7 ± 11.34^{b}	39.5 ± 5.37^{ab}	36.3 ± 6.21^{ab}
	Day 21	37.0 ± 6.38*	27.2 ± 16.35	34.4 ± 9.51	40.8 ± 9.37	36.7 ± 10.12

IgG: Immunoglobulin G, IL-1β: Interleukin-1β, TNF-α: Tumour necrosis factor-α, SOD: Superoxide dismutase, CAT: Catalase, GPx: Glutathione peroxidase, Group I: Control group, Group II: Rats given capecitabine, Group III: Rats given capecitabine and β-glucan, Group IV: Rats given capecitabine and vitamin C, Group V: Rats given capecitabine, β-glucan and vitamin C. Mean ± standard deviation. Different letters (a, b, c) in the same line show statistically significant differences between groups ($p<0.05$). The * sign in some groups and parameters indicates that there is a statistically significant difference between the 14th and 21st days ($p<0.05$).

In control group, Group II and Group III, serum IL-1β concentrations on day 14 were significantly higher than on day 21. On the 14th and 21st days of the study, TNF-α concentrations were found to be significantly lower in the control group (Group I) than in the other groups. No significant differences were found between the other groups. Serum TNF-α concentrations of Group III and Group IV on day 14 were significantly higher than those on day 21.

In a study, the pro-apoptotic effects of high-dose vitamin C (1.25-20 mM) with anti-cancer agents (eribulin mesylate, tamoxifen, fulvestrant or trastuzumab) were studied on various breast cancer cell lines (MCF-7, SK-BR3 and MDA-MB-231) as well as normal breast epithelial cells (MCF10A). High-dose vitamin C (≥10 mM) significantly reduced the viability of all breast cancer cell lines, especially MCF-7 cells. The CAT activities of MCF-7 and MDA-MD-231 cells were lower than those of MCF10A cells. In addition, cell viability of both MCF-7 and MDA-MD-231 cells was further reduced when high-dose vitamin C and eribulin mesylate were combined. Likewise, when vitamin C was combined with tamoxifen or fulvestrant, a decrease in the viability of MCF-7 cells was observed. The combination of high-dose vitamin C and trastuzumab was effective on SK-BR3 cells (Lee *et al*., 2019). Therefore, it is thought that the combination of high-dose vitamin C and conventional anti-cancer drugs will have therapeutic advantages against breast cancer cells.

Methotrexate, another anti-cancer drug, causes liver and kidney damage when used in cancer treatments. Methotrexate-induced cytotoxicity is associated with increased oxidative stress and caspase activation. Increased H_2O_2 and caspase-9/caspase-3 activation by methotrexate administration causes cell death in hepatocellular carcinoma Hep3B cells. In the study, 5 µM vitamin C combined with 0.01 µM methotrexate increased the cytotoxic effect, while it was seen that vitamin C had no effect on cytotoxicity in cells treated with 10 µM methotrexate (Yiang *et al*., 2014). This suggests that vitamin C may only have an effect on low-dose anticancer drugs.

On the other hand, the use of β-glucan together with anticancer drugs may cause antitumor and antimetastatic effects. It has been observed that reductions in body weight, spleen weight, and the number of immunocompetent cells such as macrophages and natural killer cells in cisplatin-treated mice were prevented by the combination of β-glucan (8.0 mg/kg/day, intraperitoneally). β-glucan also induced IL-12p70 production by splenocytes, resulting in increased natural killer cell activity in cisplatin-treated mice. In splenocytes, GM-CSF, G-CSF, M-CSF, IFN-γ and IL-12 showed their effect by significantly increasing the mRNA expression of p40. In addition, treatment with β-glucan significantly reduced cisplatin-induced nephrotoxicity of renal tubular cells, accompanied by increases in serum creatinine, necrosis, and apoptosis (Masuda *et al*., 2009). Concomitant use of bortezomib with β-glucan (75 mg/kg, intraperitoneal) to examine the effects of bortezomib on liver parameters in rats resulted in overstimulation of endogenous SOD activity and normalized the change in lipid peroxidation level. In addition, it was observed that β-glucan was effective on reversible tissue damage caused by bortezomib in rat liver by regulating Kupffer cell activation and the development of non-specific reactive hepatitis via NF-kB (Keles *et al*., 2014).

When IgG concentrations, which is one of the important and most studied antibodies, were examined, the highest concentration was found in Group II, which was given only capecitabine at the end of chemotherapy. The effect of chemotherapy is evident. In the groups receiving β-glucan and vitamin C supplements, values very close to the control group were obtained. In some studies with cancer patients, decreases in IgG concentrations have been observed during or one week after chemotherapy (Cui *et al*., 2020).

Contrary to the studies in the literature, high IgG concentration was observed in the capecitabine group only in this study, while lower IgG concentrations were observed in the control group and other groups. The reason for this, in our opinion, is that the experimental setup of this study was based on healthy rats, although most of the studies in the literature were already conducted on tumor-fighting subjects. It is expected that IgG, an antibody, is already low in healthy individuals. However, diarrhea, nutrient deficiency due to loss of appetite, and liver damage, which are side effects of chemotherapy, cause IgG concentrations to increase in the body (Fischbach and Dunning, 2009). Despite of suppression due to chemotherapy, the detection of higher serum IgG levels in Group II than the control group is an expected result. However, in groups that received β-glucan and vitamin C supplements in addition to capecitabine,

values very close to the control group were observed. The reason for this may be that Groups III, IV and V completed the study more healthily than Group II, which received only chemotherapy treatment. Those in these groups may have eliminated the side effects from chemotherapy. Those in these groups may have eliminated the side effects from chemotherapy. For this reason, an increase in IgG levels may not have been observed.

When the concentrations of IL-1β and TNF-α cytokines were examined in the study, different results were obtained in both cytokines. While IL-1β concentrations were observed to be lower than the control group after chemotherapy, a significantly higher concentration was observed in TNF-α value. Responses of cytokines may differ as they are time and dose dependent. As in the study of Topuz *et al*. (2008), changes in IL-1β and TNF-α concentrations may not be compatible with each other.

IL-1β is produced and released by many different immune or non-immune cell types in response to inflammatory signals. It has also been shown to commonly cause fever, chills, myalgia, and headache. In many studies, it has been observed that serum cytokines increase with chemotherapy. However, in this study, contrary to other studies, IL-1β concentrations were observed to be slightly lower in the groups receiving capecitabine compared to the control group. This is in line with the results found by Pusztai *et al*. (2004). In the study of Pusztai *et al*. (2004), each cytokine showed unique changes after chemotherapy. The researchers argued that the peak changes with 5-FU due to the different induction kinetics of cytokines may occur earlier or later than seen, and therefore the optimum sampling time may be different for each cytokine.

From the data obtained, it was concluded that suppression could not be prevented in the groups that received β-glucan and vitamin C supplements separately. One week after chemotherapy, it is observed that IL-1β concentrations decreased in the control group, in the group that received only capecitabine, and in the group that received beta-glucan supplementation in addition to capecitabine. The first two-week rise in the control group may be due to stress from gavage, even if they did not take any supplements. In the next week, since the rats continued their normal lives and were not exposed to any stress, their cytokine values may have decreased to normal levels. However, beyond the expected, no significant change in IL-1β levels was observed in the resting period after chemotherapy in the groups that received only vitamin C and both β-glucan and vitamin C supplements. The reason for this may be that the given supplements induce the release of IL-1β and this effect continues in the body for a while. In addition, it should be taken into account that different cytokine releases may be caused by individual differences such as age and gender.

One of the cytokines that has an important role in chronic inflammation caused by many diseases such as cancer is TNF-α. As expected in this study, significantly lower TNF-α concentrations were obtained in the healthy control group compared to the other groups. The value of 30.23 ± 13.64 pg/ml detected on the 14th day in the control group may have been caused by stress originating from gavage. High TNF-α concentrations were achieved in other groups due to capecitabine-induced inflammation. However, there was no significant difference between the groups that were given β-glucan and/or vitamin C supplements in addition to capecitabine and the group that took only capecitabine. Day 21 measurements show that after discontinuation of capecitabine and supplements on day 14, TNF-α concentrations decreased as expected in all groups, with reduction in inflammation and stress. However, on the 21st day data, it was not concluded that β-glucan and vitamin C supplements had a significant effect on TNF-α concentration decreases.

In a study examining the effect of different doses of vitamin C on TNF-α, MCF-7 human breast cancer cells were used. In the study, tamoxifen, a selective estrogen receptor modulator used in breast cancer, was administered to MCF-7 cells and healthy cells, and then TNF-α mRNA expressions in the cells by supplementing with 50 µM and 500 µM vitamin C were examined. When tamoxifen-treated cells were compared with control cells, it was observed that mRNA expressions of TNF-α were significantly increased in tamoxifen-treated cells. The mRNA expression of TNF-α increased 351-fold in cells treated with tamoxifen compared to control cells. In the study, in order to examine the effect of vitamin C on the mRNA expression of TNF-α, cells were incubated with vitamin C for 30 minutes, then tamoxifen was given, and after 24 hours, the mRNA expression levels of TNF-α were examined. The mRNA expressions of TNF-α were

found to be lower in cells incubated with vitamin C compared to other cells. Although this decrease was not obvious in cells incubated with 50 µM vitamin C, it was observed that mRNA expression of TNF-α decreased by 67.5% in cells pre-incubated with 500 µM vitamin C. It has been concluded that by increasing dose of vitamin C, which is used as a complement to chemotherapy, side effects caused by chemotherapy can be prevented (Subramani *et al*., 2014).

On 14th blood draw day, SOD activities in Groups III, IV and V were found to be significantly lower than Groups I and II. On 21st day, levels in Group II, which were given only capecitabine, were found to be significantly lower than other groups except the control group. The SOD activities of 14th day in Group II and the 21st day in Group IV were found to be significantly higher than other blood draw. On 14th day of study, serum CAT activities were found to be significantly lower than Groups IV and V in Group II given capecitabine, while levels in Group IV were found to be significantly higher than control group (Group I). On the 21st day, CAT activities between groups were not found to be significantly different. It was determined that comparisons of CAT activities between 14th and 21st days within each group were not statistically significant. A statistically significant difference in serum GPx activities was detected between control group (44.3 ± 2.64 ng/ml) and Group III (28.7 ± 11.34 ng/ml) at blood draw on 14th day of study. No significant differences were found between the other groups. Similarly, there were no statistically significant differences between the groups in the blood draw on the 21st day. Only in Group I, the control group, serum GPx activities on day 14 were significantly higher than the levels on day 21.

The immunosuppressive property of chemotherapy which is one of most effective cancer treatments, is known. For this reason, additional treatment methods to support immune system are applied in addition to chemotherapy. Free radicals cause fragmentation of DNA and chromosomes as well as contribute to undesired oncogene activation. Many chemotherapy drugs, such as capecitabine, give free radicals to environment as a result of their metabolic activities. The secondary defense of immune system's defense mechanism against reactive oxygen species includes transport and deactivation of existing reactive oxygen species. Enzymes such as SOD, CAT, GPx are known as antioxidant enzymes and are involved in the secondary defense against reactive oxygen species (Kaya *et al*., 2005).

A significant decrease was observed in SOD, CAT and GPx enzyme activities in rats treated with chemotherapy, while a further decrease in activity of these antioxidant enzymes occurred in adult male wistar rats administered capecitabine (500 mg/kg orally) (Rtibi *et al*., 2018). In another study, oral administration of 30 mg/kg capecitabine in Wistar rats resulted in a decrease in SOD, CAT and GPx activities (46.46%, 44.67% and 28.16%, respectively), while as a result of antioxidant use, negative effect of capecitabine on antioxidant enzyme activity decreased (Olayinka *et al*., 2017). Similarly, decrease in SOD, CAT and GPx enzyme activities due to the administration of another anticancer drug, cisplatin, was tried to be increased with vitamin C supplementation. While no change was observed in the activities of these enzymes with administration of 250 mg/kg vitamin C in male Swiss Albino mice, the activity of each enzyme increased with combination of vitamin C and cisplatin administered at 500 mg/kg (Ajith *et al*., 2007).

In this study, CAT and GPx concentrations were measured higher in the groups receiving vitamin C supplementation than in the other capecitabine groups. It is seen that vitamin C, an antioxidant, has a positive effect on the production of these enzymes despite chemotherapy. Although not as much as vitamin C, the CAT concentration in the group that received β-glucan supplementation was found to be higher than the negative control group that took only capecitabine. However, same effect on GPx was not observed. In this study, no positive or negative effects of immune-supporting β-glucan and vitamin C on SOD were observed on 14th day. However, one week after chemotherapy and supplements were discontinued, SOD levels of supplementation groups were significantly higher than those of capecitabine-only group. It appeared that β-glucan and vitamin C supplementation had a significant effect on achieving normal SOD levels in rats after capecitabine was discontinued.

Conclusion

In conclusion, the immunosuppressive feature of chemotherapy in this study caused certain decreases in the levels of antioxidant enzymes, as expected. Enzyme concentrations of the groups that received only capecitabine 14 days after the start of

chemotherapy were found to be lower than the control group. It seems that chemotherapy suppressed the production of all antioxidant enzymes, but this was different for some cytokines and antibodies. It has been observed that therapy with β-glucan, vitamin C and a combination of both have different effects for each parameter or do not have a specific effect.

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