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Oxidative and physiological effects of carvedilol, a beta–blocker, in *Daphnia magna*

Efectos oxidativos fisiológicos del carvedilol, un betabloqueante, en *Daphnia magna*

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ABSTRACT

Carvedilol is a beta–blocker. Beta–blockers affect the heart and circulation. carvedilol is used to treat heart failure and hypertension. Although this substance has effects in humans and some animals, its effect on the physiological and antioxidant status of daphnids is unknown. The aim of this study was to determine the effect of carvedilol doses (0, 0.125, 0.45 and 0.90 mg·L^{-1}) on physiological activities (heart rate, postabdominal paw activity and thoracic limb movements) and oxidative stress. In *Daphnia magna*; malondialdehyde (MDA), superoxide dismutase (SOD), catalase (CAT), total glutathione (GSH) and glutathione S transferase (GST) markers were examined. The activity of physiological parameters in *D. magna* exposed to all carvedilol concentrations was found to be lower than in the control group, and the differences were statistically significant (P<0.01). Application of carvedilol to *D. magna* resulted in lower GSH content in all groups throughout the experimental period. MDA, SOD, CAT and GST activity were improved. These findings indicate that carvedilol causes physiological and biochemical changes in *D. magna*. Daphnia species have great potential to provide valuable insights into the mechanisms of human medicine. More comprehensive research is needed on this subject.

Key words: Oxidative stress; *Daphnia*; carvedilol; antioxidant enzymes; heart rate; thoracic limb activity; post–abdominal claw activity

RESUMEN

El carvedilol es un betabloqueante. Los betabloqueantes afectan el corazón y la circulación. El carvedilol se usa para tratar la insuficiencia cardíaca y la hipertensión. Aunque esta sustancia tiene efectos en humanos y algunos animales, se desconoce su efecto sobre el estado fisiológico y antioxidante de las dafnidas. El objetivo de este estudio fue determinar el efecto de dosis de carvedilol (0, 0,125, 0,45 y 0,90 mg·L^{-1}) sobre las actividades fisiológicas (frecuencia cardíaca, actividad postabdominal de las patas y movimientos de las extremidades torácicas) y el estrés oxidativo. En Dafnia magna; Se examinaron los marcadores de malondialdehído (MDA), superóxido dismutasa (SOD), catalasa (CAT), glutatión total (GSH) y glutatión S transferasa (GST). Se encontró que la actividad de los parámetros fisiológicos en *D. magna* expuesta a todas las concentraciones de carvedilol era menor que en el grupo de control, y las diferencias fueron estadísticamente significativas (P<0,01). La aplicación de carvedilol a *D. magna* resultó en un menor contenido de GSH en todos los grupos durante el período experimental. Se mejoró la actividad de MDA, SOD, CAT y GST. Estos hallazgos indican que el carvedilol provoca cambios fisiológicos y bioquímicos en *D. magna*. Las especies de Daphnia tienen un gran potencial para proporcionar información valiosa sobre los mecanismos de la medicina humana. Se necesita una investigación más exhaustiva sobre este tema.

Palabras clave: Estrés oxidativo; *Dafnia*; carvedilol; enzimas antioxidantes; ritmo cardiaco; actividad de las extremidades torácicas; actividad de las garras postabdominales

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INTRODUCTION

A Human medicines are inevitable tools for human health. It is possible to drain pharmaceuticals from city treatment plants and hospital wastes into water intermittently. However, we cannot ignore the dangers of pollution when discharged into natural waters and the negative impact it may have on all living things, such as *Daphnia magna*, living in these waters. Many medicaments are known to have harmful effects on fish and other aquatic organisms. Various effects of theirs on aquatic animals have been extensively reviewed [1, 2, 3, 4, 5, 6, 7].

carvedilol was patented in 1978 and approved for medical use in the United States in 1995. carvedilol is the first beta blocker approved for the treatment of all forms of congestive heart failure [8]. It is a non-selective β-adrenergic receptor blocker of the third generation [9]. Neuropharmacological and biochemical studies have provided convincing evidence that carvedilol has effects on animals. Previously a study demonstrated the neuroprotective effect of carvedilol in rats-at least partly- and its antioxidant effect [10]. For example, Asanuma *et al*. [11] carvedilol has vasodilator properties in the cardiovascular system, Sgobbo *et al*. [12] reported that carvedilol also has antioxidant activity. The beta-blockers were detected at higher concentrations in surface waters [13, 14]. Recently, there are common studies on the accumulation and effect of human pharmaceuticals on water, sediment, and aquatic animals [2, 5, 15, 16, 17, 18, 19, 20]. There are a few reports on the effects of the β - blockers in aquatic organisms [21, 22, 23, 24, 25, 26, 27, 28]. For example, there is a high sensitivity of algae to beta-blockers [29]. Previous studies have described the effects of β-blockers on aquatic organisms as impairing testosterone levels, reducing fertility and reproductive rates, and/or causing abnormal behavior [21, 25, 26].

Daphnia also called "water flea" is a very common planktonic invertebrate organism inhabiting freshwater ecosystems such as lakes and ponds. *D. magna* is a type of species that belongs to Phylum Arthropoda, Class Crustacea, Order Cladocera, and Family Daphniidae. The short life-span and high rate of reproduction makes *D. magna* a potential species for exploring population sequences of chemical exposures. Bioassays with *Daphnia* sp. (mostly *Daphnia magna*) are regularly used in ecotoxicological studies because they have high fertility values, easy to maintain in laboratory conditions, ubiquitous, and important bioindicators for aquatic environments due to their sensitivity to contaminants and position [30, 31, 32]. *D. magna* is a freshwater crustacean and is among the most sensitive model organisms. Therefore, they are immediately affected by environmental changes. Standard protocols recommended by the International Organization for Standardization (ISO), the Environmental Protection Agency (EPA), and the Organization for Economic Co-operation and Development (OECD) mandate the use of *D. magna* as a model organism [33, 34].

Bioindicators are species that can be used to signify the health of an ecosystem. The bioindicators in an aquatic ecosystem include algae, macrophyte, zooplankton, bivalve mollusks, seabirds and fish that can be used to assess the contaminants in aquatic system. *D. magna* is frequently used as a model organism and is considered a keystone species. In the aquatic ecosystem, *D. Magna* plays an important role as the primary consumer of the food chain. Therefore, any significant changes in population sizes can be a good indicator of aquatic toxicology. Environmental variables such as temperature and food quality and quantity can have significant effects on zooplankton *D. Magna* [35, 36, 37, 38, 39].

The ecotoxicological impacts from carvedilol are not documented at all as of this time and need significant research. Chronic toxicity testing on macroinvertebrates and aquatic plants are still lacking, and multi-generational testing for beta-blockers is also quite scarce for all aquatic organisms. Congruently, bioaccumulation and field studies for the detection of beta-blockers in aquatic biota are also scarce [40].

Processes related to oxidative stress ıt attracts great attention due to its sensitivity and prevalence. The most studied parameters in many pathophysiological conditions [41]; prooxidative nature of toxic substances (SOD), catalase (CAT), antioxidant tripeptide glutathione (GSH) and the relevant enzymes are measured. In literature searches, no previous studies on carvedilol were found regarding the physiological and antioxidant mechanisms of *D. magna*. This study was conducted to determine the effects of carvedilol on heart rate, post-abdominal paw activity, thoracic limb movements, and MDA, SOD, CAT, and GSH activities of *D. magna*.

MATERIALS AND METHODS

Test organisms and experimental procedure

Daphnia magna samples used as model organisms in the study were taken from a commercial aquarium shop and brought to the laboratory environment alive in water. *D. magna* samples were placed in 10 litre plastic containers filled with previously rested, aerated, non-chlorinated city tap water, allowing them to adapt to the laboratory environment for 5 days. Characteristics of the aquatic environment where organisms are left; temperature was measured as 25 ± 2°C, pH 7-8, dissolved oxygen > 7 mg·L^{-1}, total hardness 130-160 mg·L^{-1} $CaCO_3$, alkalinity 110-120 mg·L^{-1} $CaCO_3$. Natural lighting was applied in the laboratory. Spirulina in powder form, purchased from the market, was used to feed *D. magna*. Spirulina was diluted at 2 mg·L^{-1} and *D. magna* were fed by adding a few drops to each tank once a day. Feeding was stopped 24 hours (h) before the experiments.

In bioexperiments; In *D. magna*, to determine the oxidative and physiological effects of carvedilol at different doses (0.225, 0.45 and 0.90 mg·L^{-1}), 4 groups were used for 48 h and 96 h, including 3 experimental and 1 control (0 mg·L^{-1}) group. 4 groups were created. The study was carried out in a total of 24 glass containers (1 L), with 3 repetitions. During the adaptation of the organisms to the glass containers, 250 mL of rested, non-chlorinated city tap water with the same characteristics of the water environment (temperature, pH, dissolved oxygen) was left in the glass containers. 100 *Daphnias* were placed in each of the experimental and control groups, and a total of 2400 *Daphnias* were used.

Physiological assays

Heart rate, PCA, and thoracic limb activity (TLA) were determined according to the method of Bownik *et al*. [4]. The heart, post-abdominal paw, and thoracic limbs were observed under a microscope (Olympus CX31, Japan) for one minute, and physiological activities were determined. For these tests, 10 *Daphnias* were randomly sampled from 100 *Daphnias* in each experimental group and examined. The average value of the three observations was used to calculate heart rate, expressed as beats per minute. The calculation method here, that is, the average value of three observations, was also applied to calculate the average values of other analysis parameters.

Biochemical tests

Determination of lipid peroxidation and antioxidant parameters

To be used in biochemical tests, 10 *Daphnias* samples were taken from each of the bioexperiment groups; Homogenates were prepared separately in 0.1 M acetate buffer, pH 5.0, in the cold. Unbroken cells and cell debris were removed by centrifugation (Nüve, NF800R, Türkiye) at 700 G for 10 min. The supernatants thus obtained were stored at -20°C until use.

MDA levels, which are indicative of lipid peroxidation in the samples, were determined according to the method of Placer *et al.* [42]. Thiobarbituric acids were measured by conversion to reactive substances with high absorbance at 532 nm. MDA ratios were expressed as nmol/mg protein. The total SOD activity was measured by the nitro blue tetrazolium (NBT) method at 560 nm [43]. The enzymatic activity was calculated as U·mg^{-1} protein. One unit of SOD was defined as the amount of sample required to inhibit the rate of reduction of NBT by 50%. The CAT activity was detected by the decrease in hydrogen peroxide radical (H_2O_2) concentration at 240 nm [44]. The CAT activity (1 U) was expressed as 1 mmol of decomposed hydrogen peroxide per second per mg of protein.

Total GSH levels were analyzed according to Ellman's method [45]. Results were expressed as mol/mg protein. The GST activity was measured using the method of Habig *et al.* (46) and was calculated as µmol·mg protein^{-1}·min^{-1}.

Statistical Analysis

All data are given as mean and standard deviation. Obtained data were analyzed using the SPSS 16.0 package program by analysis of variance (ANOVA) and Tukey test, and differences between groups were determined. Statistical differences in all analyses were determined at the 95% confidence interval.

RESULTS AND DISCUSSION

Physiological parameters

Acute administration of carvedilol significantly decreased cause heart rate, post-abdominal claw activity, and thoracic limb movements in *D. magna* as compared to control. The activity of physiological parameters in *D. magna* exposed to all carvedilol concentrations was found to be lower than in the control group, and the differences were statistically significant (P<0.01). Heart rate (FIG. 1) showed a decreasing trend throughout the experiment. For the experimental periods, heart beats per minute in the control groups were recorded between 398.83 ± 14.55 and 403.83 ± 17.08.

In the experimental groups administered different doses of carvedilol (0.225, 0.45 and 0.90 mg·L^{-1}), at the end of the 48th and 96th hours, the lowest heart rates per minute were respectively (heart beats·minute^{-1}); It was determined as 245.16 ± 16.36, 210.66 ± 45.00, 145.83 ± 12.67 and 182.50 ± 17.75, 162.00 ± 23.47, 129.83 ± 18.11, (P<0.001). Accordingly, carvedilol caused immobility and a decrease in heart rate in *D. magna*. This decrease; It showed parallelism with the increase in carvedilol dose and duration.

Post-abdominal paw activity analyzed in control and experimental animal *D. magna* exposed to carvedilol for 48 h and 96 h is presented in FIG. 2. From the results, post-abdominal paw activity (beats per minute); It was observed that it was 15.83 ± 4.79 at the 48th hour and gradually decreased at the 96th hour. However, this decrease was found to be not significant. The post-abdominal claw activity of the experimental groups was found to be significantly lower during 96h. There was a reduction to the extent of 42.07, 38.98, and 75,80 percent at 48h as compared to the control group level the post abdominal claw activity shot up to a decrease up to 45.09, 67.04, and 88.99 percent, at the 96 hours. Post-abdominal claw activity decreases with higher doses of carvedilol. The 0,225 mg·L^{-1} groups resulted in a statistically significantly higher level of TLA compared with the other experimental group (FIG. 2).

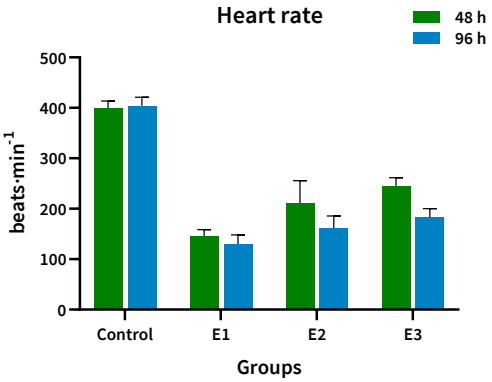

FIGURE 1. *Daphnia magna* heart rate trend

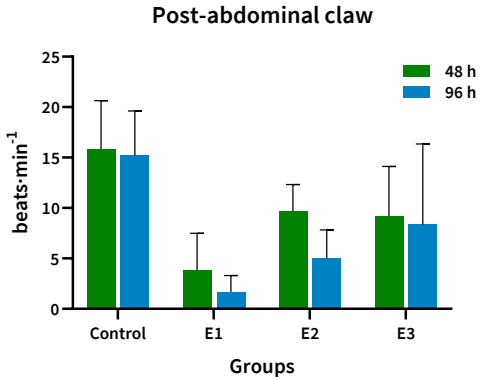

FIGURE 2. The post-abdominal claw activity (beats per minute)

It was observed that the average number of beats per minute of thorax extremity movements in the control and carvedilol groups in the different periods of 48 and 96 h of the experiment was 148.50 ± 11.59 in the control groups and 150.67 ± 15.20 in the carvedilol groups. Thoracic limb activity (TLA) at all experimental groups at 48 h was found to be lower than the initial level and there was a regular trend of variation. The TLA level as observed at the 48 h progressive decrease was observed till 96 h. Administration of acute carvedilol (0.90 mg·L^{-1}) in *D. magna* for 48 h and 96 h significantly decreased the thoracic limb movement levels (FIG. 3, P<0,01). Thoracic limb movements were significantly reduced by more than 55% at 96 h.

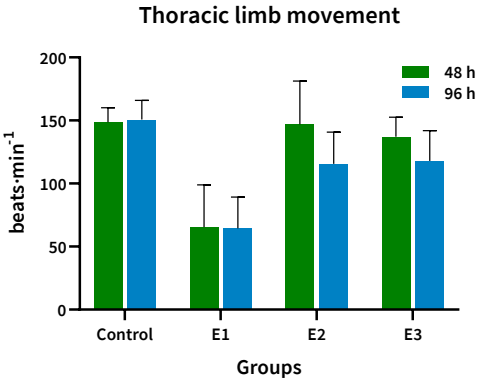

FIGURE 3. Thoracic limb activity of *Daphnia magna* exposed to various concentrations of carvedilol.

The MDA level increased in the experimental groups at both 48 and 96 h (FIG. 4). The MDA results of *D. magna* from control, experimental groups were found to be significant variations from each other at the 48 h (FIG. 4). Similar data were also reported for 96 h exposures. In control groups, the MDA level was stable during all the exposure times. Moreover, in experimental groups, among experimental times a not significant change in the MDA content of *D. magna* was recorded.

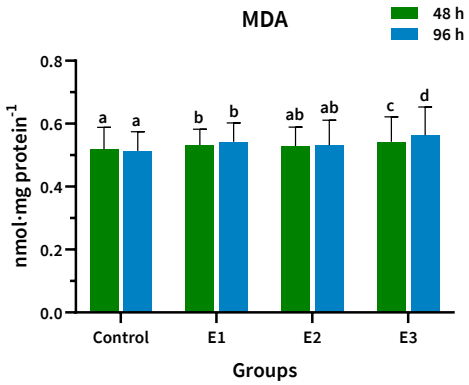

FIGURE 4. The MDA level. a, b, c, d: The difference between the mean (±SE) values indicated by different letters on the same line is statistically significant (*P*<0.05)

D. magna from experimental groups showed a progressive increase in the activity of SOD until 48 h of treatment, giving the increment according to control the group. After, at the end of 96 h, the SOD level increase was 3.20, 19.72, and 0.89 percent, respectively. The increase in the SOD level of *D. magna* from the E2 group was more pronounced than that of its counterpart from the E1 and E2 groups (FIG. 5).

FIG. 6 presents the data on the changes in whole–body CAT level of *D. magna* treated with different concentrations of carvedilol in the experimental groups. The CAT in the 0.45 mg·L^{-1} carvedilol group showed an increase over the control data. In this group, the increase in the CAT level at 48 h was 8.02 percent. There was a progressive and stable increase in the CAT in all the experiments up to 96 h, displaying a peak depletion of 2.60, 11.18, and 2.16 percent, respectively.

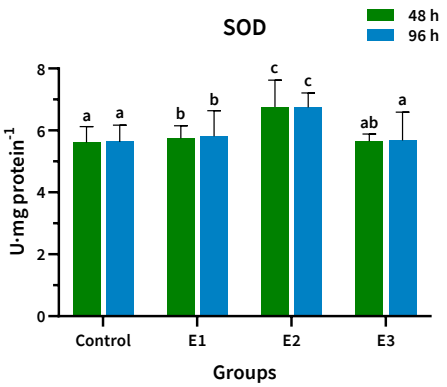

FIGURE 5. *Daphnia magna,* SOD activity, a, b, c: The difference between the mean (±SE) values indicated by different letters on the same line is statistically significant *P*<0.05)

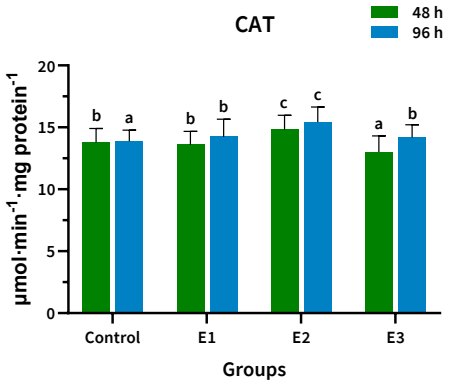

FIGURE 6. Whole body catalase level of *Daphnia magna.* a, b, c: The difference between the mean (±SE) values indicated by different letters on the same line is statistically significant (*P*<0.05)

The total GSH in samples was 0.86 and 0.76 µmol·g^{-1} protein in the control group during the experimental period. Due to the exposure to carvedilol, the total GSH activity in the *D. magna* showed a moderate depletion trend. No statistical difference was found between the groups. The values are presented in FIG. 7.

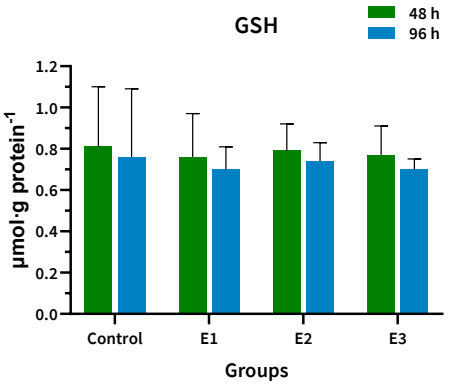

FIGURE 7. Total GSH activity in the *Daphnia magna*

The GST content of the test organisms was found to be on an increasing trend. The observed GST level was at 0.014 in the control group whereas 0.014, 0.017, and 0.024 U·/mg protein^{-1}, respectively, in experimental groups at the 48 h. In group E1, very little fluctuation was observed. In the experimental samples, the maximum GST content was found maximum 0.014 U·/mg protein^{-1} while in control samples it was recorded at 0.025 U·/mg protein^{-1} in the E3 group at 96 h. The GST values are presented in FIG. 8.

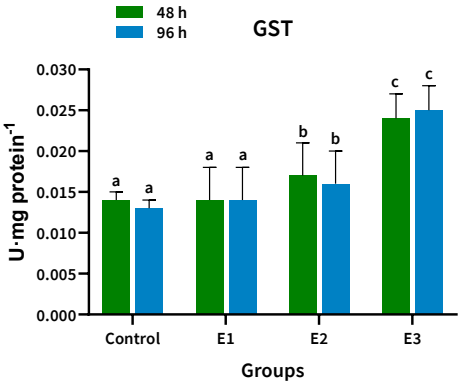

FIGURE 8. The GST content of *Daphnia magna*. a, b, c : The difference between the mean (± SE) values indicated by different letters on the same line is statistically significant *P*<0.05)

Behaviour tests

In the current study, heart rates were roughly similar between control groups during the experimental periods. However, compared to control groups, heart rate values in *D. magna* gradually decreased up to 48 h and 96 h during short-term exposure to different doses of carvedilol.

After 48 and 96 h of exposure to different concentrations of carvedilol, a decrease in heart rate of *D. magna* was observed in parallel with the increase in concentration. However, the percent suppression of heartbeat was much greater in the *D. magna* exposed to high concentration, and it increased with the increase in the exposure period. Whereas in low concentrations percent suppression was relatively less, and it decreased with the increase in the exposure period. The results showed an irreversible suppression of physiological activities of *D. magna* exposed to high concentration and an initial suppression accompanied by a gradual recovery in those exposed to low concentration of carvedilol.

In the present study, the level of post-abdominal claw showed there were considerable changes in control and experimental groups in the administration of carvedilol. All the values are highly significant at *P*<0.001 level except 48 h. In the current study, we found a reduction in post-abdominal claw level. Several authors have observed a fluctuation in the post-abdominal claw level in *D. magna* when exposed to human drugs [47, 48]. A similar study conducted in the *D. magna* exposed to apomorphine, a dopamine agonist, by Bownik *et al.* [4], observed a fluctuation in post-abdominal claw level

It is well known that the heart of Daphnids is myogenic and the best originates at any point of the heart, whether the concentrations are local or complete. It is also known that Acetylcholine, which has a stimulating function such as regulating heart contractions and blood pressure, and catechins, which have high antioxidant potential, can inhibit heart rate and stimulate heartbeat [49]. Changes in heart rate have been used as sensitive indicators of stress in crustaceans. It has been explained in many studies that acetylcholine synthesis or catecholamine production increases heart rate in *Daphnia* species [4, 23, 24, 35, 36, 47, 50]. In the current study; The decrease in heart rate in Daphnia exposed to different dosages of carvedilol is an indication that acetylcholine synthesis or catecholamine production slows down.

Probably, the stress conditions developed within the organs of the animal might have induced a low Internal pressure, which in turn might have dropped the rate of heartbeat. Interestingly, the gradual drastic decrease in the rate of the heartbeat from 48 h of exposure to 96 h was shown. Probably, on very prolonged exposure, the high concentrations of carvedilol may cause cessation of the heartbeat leading to the death of the *D. magna*. Postmes *et al.* [51] It was observed that the heart rate did not change in *D. magna* after administration of metoprolol, a beta-blocking drug like carvedilol. Villegas-Navarro *et al.* [35] observed no change heart rate of *D. magna* in groups of high-dose metoprolol administration. However, they reported that increased heart rate at low concentrations. *Daphnia pulex* were exposed to containing 29.6 mg·L^{-1} propranolol for acute administration. The treated groups showed a significantly reduced heart rate in *D. pulex* [36]. In the present study, reductions in heart rate were seen at all of the low and high doses.

In the literature research, no information was found about the effects of carvedilol on the thoracic limb movement of *D. magna*. However, there are some studies describing other chemicals such as titanium dioxide [47]; fluoroquinolones [48]; Apomorphine [4] and Diltiazem [52]. In this study, it was observed that even the lowest dose of carvedilol (0.225 mg·L^{-1}) reduced activity at the thoracic limb level. With this result; contribution will be made to the literature. Likewise, in the 0.45 and 0.90 mg·L^{-1}, high doses groups, carvedilol decreased the number of thoracic limb activities. Thoracic limbs play an important role in the feeding and oxygen consumption of daphnids [53]. Perhaps the disturbance in oxygen consumption levels after Daphnid's exposure to carvedilol may have reduced ventilation, resulting in hypoxia and biochemical degradation. Maybe, the decrease of thoracic limb activity in response to exposure to carvedilol may be due to the decrease in internal pressure mediated by the decrease in heart rate.

Biochemicals assays

The present study for the first time estimates in Daphnids the modification of oxidative stress parameters by carvedilol. The effect on the antioxidant status of *D. magna* of carvedilol had not been studied. The present work is the first report on *D. magna* of carvedilol and the results obtained suggest a uniform susceptibility of *D. magna* to reactive oxygen species menace with time.

One of the harmful effects of oxyradicals is lipid peroxidation [54]. Lipid peroxides easily decay to release highly reactive carbonyl fragments. The most prominent of these are the malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE). Tissue TBA reactive substances (TBARS) are usually considered to disclose tissue levels of MDA or the extent of lipid peroxidation [55]. Therefore, the measurement of tissue TBARS levels is one method of appraising the content of tissue damage due to free radicals. *D. magna* demonstrated increased production of MDA in the presence of carvedilol after 48 h with a gradual increase thereafter. After 96 h there was a significant drastic increase in MDA production in other test groups. A progressive increase in the

MDA levels with time indicates a parallel depletion of unsaturated fatty acids from membrane lipids. Nkoom *et al.* [56] investigated the antioxidant effect of diclofenac on the oxidative stress response and lipid peroxidation in Daphnids. *D. magna* exposed to diclofenac has significantly increased the MDA level. The MDA content increment was in a concentration-dependent manner from low doses as compared to the control, then decreased at high doses as compared to the low dose treatment. Also, De Felice *et al.* [57] found enhanced lipid peroxidation in *D. magna* exposed to Methamphetamine for 21 days.

There are SOD and CAT important tasks in scavenging toxic intermediates. They also play an important role in the body's defense mechanism against the detrimental effects of oxygen-free radicals in biological systems. A decrease in the activity of these enzymes can lead to the formation of O_2 and H_2O_2 which in turn can form hydroxyl radicals (OH). This radical can bring about several reactions which can be harmful to the tissues. We observed that the maximum SOD activity in acute exposure to carvedilol was 6.75 ± 0.87 units·mg^{-1} and 6.74 ± 0.47 in the E2 group. The magnitude of increase was 2.32, 20.32, and 0.71 percent, respectively, in groups of administration acute carvedilol according to the control group at 48 h.

In this study, it was shown that the mean CAT activity in Daphnids increased especially at the 96th h. The magnitude of the increase was 2.59, 11.18, and 2.16 percent in the experimental groups, respectively. A significant increase in CAT activity was observed by Franzellitti *et al.* [28] in the *Mytilus galloprovincialis* exposed to low-dose propranolol. In addition, Contardo-Jara *et al.* [27], found *Dreissena polymorpha* exposed to different concentrations of metoprolol for 4 and 7 days. The authors showed that CAT activity increased. However, some study findings have been reported to be in the opposite direction of these data. Oliveira *et al.* [58] reported that propranolol, a beta-blocker drug, did not affect CAT activity in *D. magna*. Similar observations were also reported in mussels exposed to propranolol [59]. In this study, a dose-related decrease in CAT activity was seen. Asayama *et al.* [60] described CAT activity reduction of atenolol, arotinolol, and carteolol administration, beta-blocker drugs, in the cardiac muscle of the hyperthyroid rat.

Lipid peroxidation requires oxygen (O_2) uptake and involves the production of superoxide radicals. The production of superoxide dismutase and catalase was caused to possibly result in the accumulation of O_2 and H_2O_2 in *D. magna* exposed to carvedilol. The possible accumulation of O_2 and H_2O_2 may increase the level of lipid peroxidation in *D. magna*. Higher levels of lipid peroxidation in experimental groups may be due to the higher uptake of oxygen and greater induction of SOD and CAT levels.

Antioxidant defense either directly or indirectly depends on GSH. Hence the decline in the level of GSH results in an imbalance in the functioning of antioxidant capacity. The study shows that there is a considerable amount of decrease in GSH. However, there was no significant depression of GSH content. This decrease led to increased oxidative stress which is supported by the elevated level of lipid peroxides in the carvedilol-treated *D. magna*. The status of the antioxidant defense mechanism in *D. magna* was also altered following exposure to carvedilol. This depression in GSH content may be the outcome of the removal of H_2O_2 by GSH [61] under stress conditions. Also, MDA and glutathione levels have a strong significant correlation which means that the higher the MDA level, the lower the level of glutathione [62].

Glutathione S-transferases (GST) are cytosolic enzymes that catalyze the conjugation of glutathione with a substrate, which is hydrophobic and electrophilic [63]. The activity of GST level declined during carvedilol treatment. This decrease may be associated with the decreased availability of GSH, which acts as a cofactor for the catalytic activity of GST. GST may inhibition has been declerated by previous studies. A study reported that the GST activity in *D. magna* was exposed to diclofenac [56]. The decrease in GST activity is associated with an increase in the MDA level. In this study, it was determined that the MDA level of MDA increased. The experimental organisms showed an increased level of MDA. The results are in agreement with the results of Quinn *et al.* [64] and Schmidt *et al.* [65] in which exposure to *D. polymorpha* and *Mytilus* sp. diclofenac, and gemfibrozil increased the GST activity.

CONCLUSION

Full evaluation of the effects of contaminants or human drugs; More comprehensive research and analyzes are needed to evaluate cellular, biochemical and non-biochemical aspects. The data presented above, showing significant alterations in *D. magna* oxidative stress parameters following carvedilol exposure, demonstrate clear biochemical suppression and provide a clear indication that the antioxidant enzyme systems of these organisms are modulated by environmental stressors. More studies are needed to explain the relationship between human drugs and the suppression of the biochemical mechanism of *D. magna*.

Consent for publications

The author read and approved the final manuscript for publication.

Ethics approval and consent to participate

No human or animals were used in the present research.

Availability of data and material

The data that support the findings of this study are available from the corresponding author upon reasonable request

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