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Full length Research

Air Freshener Induced Oxidative Stress and Its Adverse Effects on Immunity

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Recently, the use of 'sunlight' air freshener in Nigeria is on the increase. Some people use air freshener in the office, car as well as in their homes, thereby constantly being exposed to it without consideration of its adverse effect on human health. This study is therefore aimed at assessing the effect of "sunlight" air freshener on oxidative stress biomarkers. Thirty (30) Wistar rats were divided into three groups of ten each and kept in different rooms. Rats in group 1 were not exposed to any substance; those in groups 2 and 3 were exposed to sunlight air freshener for 8 hours daily for 28 days by inhalation. After 28 days exposure, animals in group 3 were allowed to recover for 14 days. Throughout the experiment, all animals were fed *ad libitum* with standard feed and drinking water. At the end of the experiment, rats were sacrificed after an overnight fast under diethyl ether as anesthesia. Blood samples were collected through cardiac puncture. Oxidative stress biomarkers were determined using standard methods. The result showed that air freshener significantly increased lipid peroxidation but significantly reduced glutathione concentration after 28 days of exposure. Activities of antioxidant enzymes (catalase, superoxide dismutase and glutathione peroxidase) were observed to significantly increase due to air freshener exposure when compared with those in the control group. The effects of air freshener on oxidative stress biomarkers (except glutathione) were reversed when animals exposed to air freshener for 28 days were allowed to recover for 14 days. This study revealed that sunlight air freshener induced oxidative stress, thus possesses the propensity to destroy the immune system. Frequent use of this air freshener should be discouraged.

Keywords: Air Freshener, Antioxidants, Free Radicals, Immunity, Oxidative Stress.

INTRODUCTION

Oxidative stress is the result of production of reactive oxygen species (ROS). Sometimes they are also referred to as Reactive Oxygen Intermediates (ROI). They occur as a result of metabolism of tissues (Repine and Bast, 1997). ROS acts as signaling mediators as most of the time may be beneficial. Most of the exogenous substances may activate the production of these ROS. Cigarette

smoke, UV radiation in the atmosphere, alcohol, drugs and cancer chemotherapeutic agents and radiotherapy treatments induce oxidative stress. Petroleum combustion products, heavy metals as well as pesticide particles and their metabolic end products also initiate the oxidative stress. Infections, tissue injury, and ischaemia also contribute in the elevated levels of ROS (Kasahara et al., 2001). Cigarette smoke is one of the exogenous agents that induce oxidative damage in cell line. Cigarette smoke induces oxidation of structural and functional components and also able to decrease the endothelial growth. In a recent study, Airaodion et al., (2019a) reported that hydrocarbon induced oxidative stress when animals were fed with crude oil treated-diet. Several studies have also reported that alcohol has the propensity to induce oxidative stress (Airaodion et al., 2019b, c; Ogbuagu et al., 2019).

Air fresheners are consumer products that typically emit fragrance and are used in homes or commercial interiors such as restrooms, foyers, hallways, vestibules and other smaller indoor areas, as well as larger areas such as hotel lobbies, auto dealerships, medical facilities, public arenas and other large interior spaces (Wolverton et al., 2004). There are many different methods and brands of air fresheners. Some of the different types of air fresheners include electric fan air fresheners, gravity drip hygiene odor control cleaning systems, passive non-mechanical evaporating aroma diffusers, metered aerosol time-operated mist dispensers, sprays, candles, oils, gels, beads, and plug-ins. Some air fresheners contain chemicals that provoke allergy and asthma symptoms or are toxic. Air freshening is not only limited to modern day sprays; air freshening also can involve the use of organic and everyday house hold items. Although air fresheners are primarily used for odor elimination, some people use air fresheners for the pleasant odors they emit (Wolverton et al., 2004).

The term air freshener may be misunderstood since these products do not considerably reduce air pollutants but rather add more substances to the air that have an odor strong enough to mask bad odors (Kim et al., 2015). Past Studies have shown that air fresheners emit over hundred different chemicals, including volatile organic compounds such as terpernes, benzene, formaldehyde, terpenoids, ethanol, formaldehyde, benzene, toluene, xylene and phthalate esters (BEUC, 2005; Senthikumaran et al., 2012). The components emitted from air fresheners are directly inhaled by the respiratory system through the nose to the alveoli; the eyes, nose, and skin are directly affected during the usage of air freshener (Kim et al., 2015). In addition, the VOCs emitted by air fresheners react with ozone to produce secondary pollutants such as ultrafine particles; the particles formed by the reaction affect health in a manner dependent on particle diameter. Secondary pollutants also affect the respiratory system, central nervous system, and immune response (Kim et al., 2015).

Many air fresheners employ carcinogens, volatile organic compounds and known toxins such as phthalate esters in their formulas. A Natural Resources Defense Council (NRDC) study of 13 common household air fresheners found that most of the surveyed products contain chemicals that can aggravate asthma and affect reproductive development. The NRDC called for more rigorous supervision of the manufacturers and their products, which are widely assumed to be safe (NRDC, 2007).

In 2009, Anne C. Steinemann published a study of top-selling air fresheners and laundry products (Steinemann, 2009). She found that all products tested gave off chemicals regulated as toxic or hazardous under federal laws, including carcinogens with no safe exposure level, but none of these chemicals were listed on any of the product labels or Material Safety Data Sheets. Chemicals included acetone, the active ingredient in paint thinner and nail-polish remover; chloromethane, a neurotoxicant and respiratory toxicant; and acetaldehyde and 1,4-dioxane, both carcinogens. A plug-in air freshener contained more than 20 different volatile organic compounds, with more than one-third classified as toxic or hazardous under federal laws. Even air fresheners called "organic," "green," or with "essential oils" emitted hazardous chemicals, including carcinogens (Steinemann, 2009). Sunlight air freshener (Figure 1) is the most common air freshener in popularly used in Nigeria. Therefore, this study is aimed at assessing the effect of this air freshener on oxidative stress biomarkers.

MATERIALS AND METHODS

Collection of Air Freshener

Sunlight air freshener was purchased in a super market in Douglas, Owerri, Imo State, Nigeria and

Figure 1. Sunlight Air Freshener

was kept at room temperature before and during the experiment.

Experimental Design and Animal Treatment

Thirty Wistar rats weighing between 160 and 190 g were used for this study. They were acclimatized for seven (7) days to laboratory conditions before the commencement of the experiment. During this period, they were fed *ad libitum* with standard feed and drinking water and were housed in clean cages placed in well-ventilated housing conditions (under humid tropical conditions) throughout the experiment. All the animals received humane care according to the criteria outlined in the 'Guide for the Care and Use of Laboratory Animals' prepared by the National Academy of Science and published by the National Institute of Health. After the seven days acclimatization period, the animals were weighed and recorded and were divided into three groups of ten each and kept in different rooms. Rats in group 1 were not exposed to any substance; those in groups 2 and 3 were exposed to sunlight air freshener by inhalation for 8 hours daily for 28 days following the method of Akingbade et al. (2014). After the 28 days of exposure, animals in group 3 were allowed to recover for 14 days. Throughout the experiment, all animals were fed *ad libitum* with standard feed and drinking water. At the end of the experiment, rats were weighed and recorded. They were sacrificed after an overnight fast under diethyl ether as anesthesia. Blood samples were collected via cardiac puncture.

Determination of Oxidative Stress Biomarkers

Determination of Lipid Peroxidation (LPO), Reduced Glutathione (GSH), Catalase (CAT), Superoxide Dismutase (SOD) and Glutathione peroxidase (GPx) were carried out according to the methods previously described by Airaodion et al., (2019d).

Data Analysis

Data were subjected to analysis using Microsoft Excel, 2013 and results were presented as mean standard ± deviation with n = 10.

RESULTS

Air freshener exposure for 28 days significantly decreased weight gained by animals when compared with those in the control group (Figure 2). Exposure of animals to air freshener significantly elevated the malondialdehyde (MDA) levels (Figure 3) as well as the activities of antioxidant enzymes in animals when compared to those in the control group (Figures 5-7). These effects were reversed when animals were allowed to recover from the effect of air freshener exposure for 14 days. Glutathione level was depleted in animals exposed to air freshener when compared with those in the control group, and the effect was sustained after the 14 days recovery period (Figure 4).

DISCUSSION

The rise in popularity of air fresheners has outpaced awareness of the potential health threats from exposure to the effects of chemicals they may contain. Air fresheners are consumer products that emit fragrance to provide an aroma to a space, to mask an odor, or both (Senthikumaran et al., 2012). They are known to contain a number of different chemical agents in order to neutralize offensive odors and create a more pleasant scent (Gilbert, 2009). A study in 2006 found that the prominent products of the reaction of terpenes found in air fresheners with ozone included formaldehyde, hydroxyl radical, and secondary ultrafine particles (EPA, 2006). In addition to this effect, it is associated with sensory irritation, most commercial air fresheners have low sensory threshold, and thus damaging health effect comes from sensory

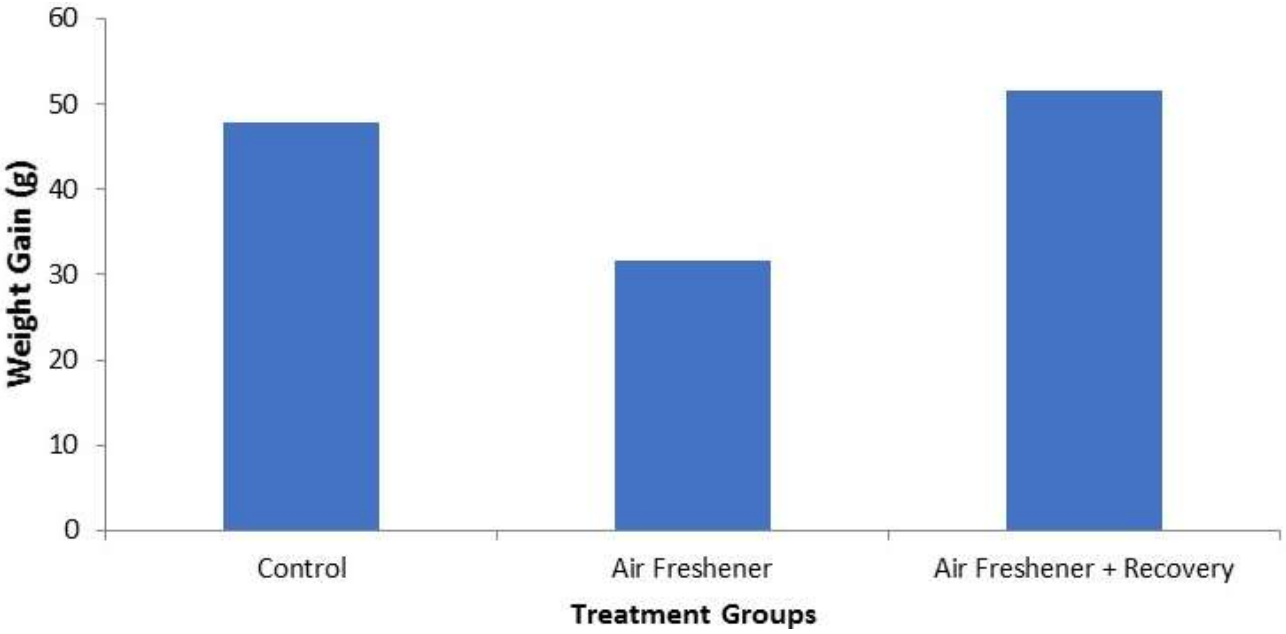

Figure 2. Effect of Air Freshener on the Weight Gained by Animals after 28 Days of Exposure.

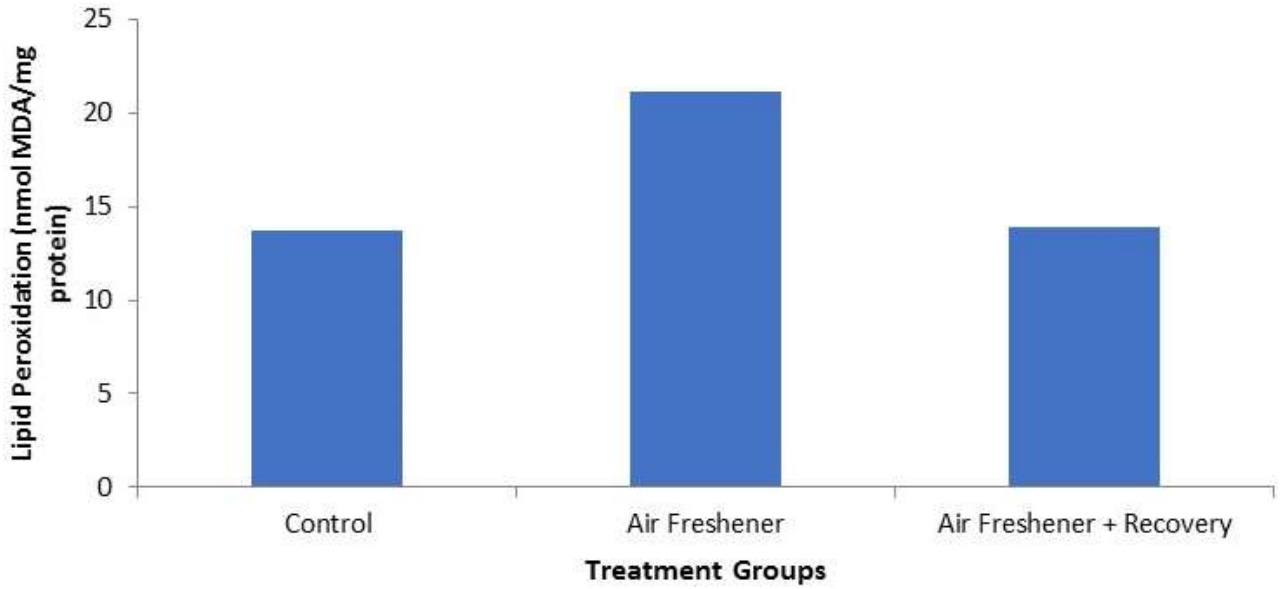

Figure 3. Effect of Air Freshener on the Lipid Peroxidation in Animals after 28 Days of Exposure.

irritations that are caused by constant use of air fresheners (Wilkins et al., 2007).

Exposure of animals to air freshener in this study caused a significant decrease in weight gained by experimental animals when compared with those of the control animals (Figure 2). This might be attributed to decrease in food intake as observed during the experimental period. It might have also resulted from the effects of fragrance and perfume ingredients that caused interruption in absorption and metabolism of food nutrients essential for health (Gentry et al., 2004). Similar decrease in weight

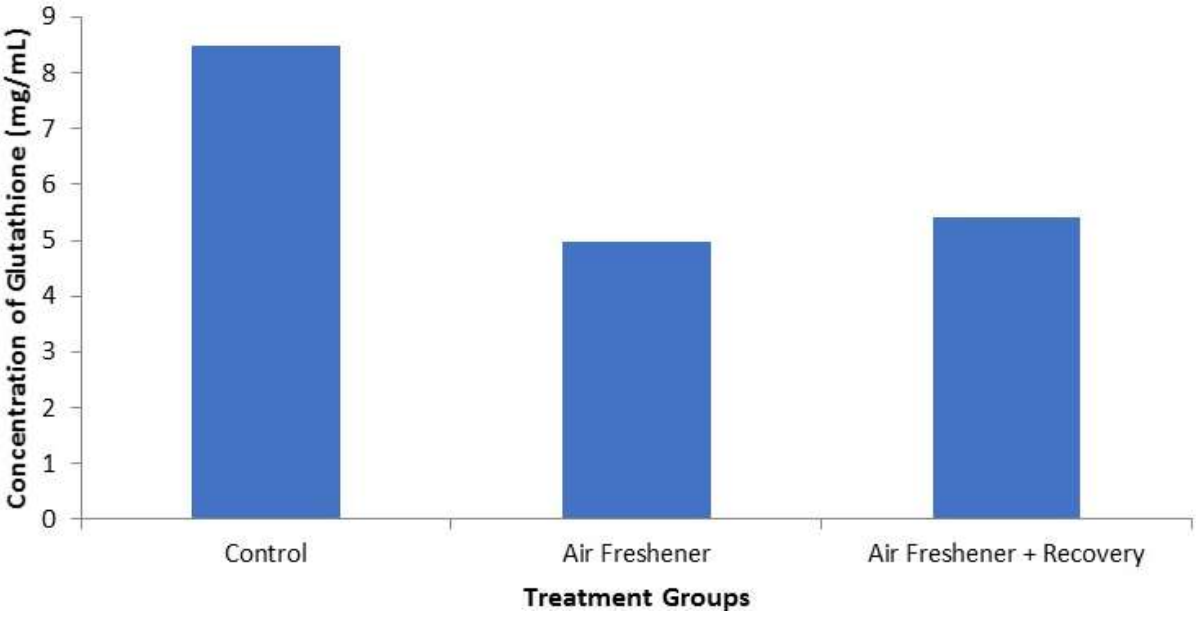

Figure 4. Effect of Air Freshener on the Concentration of Reduced Glutathione (GSH) in Animals after 28 Days of Exposure.

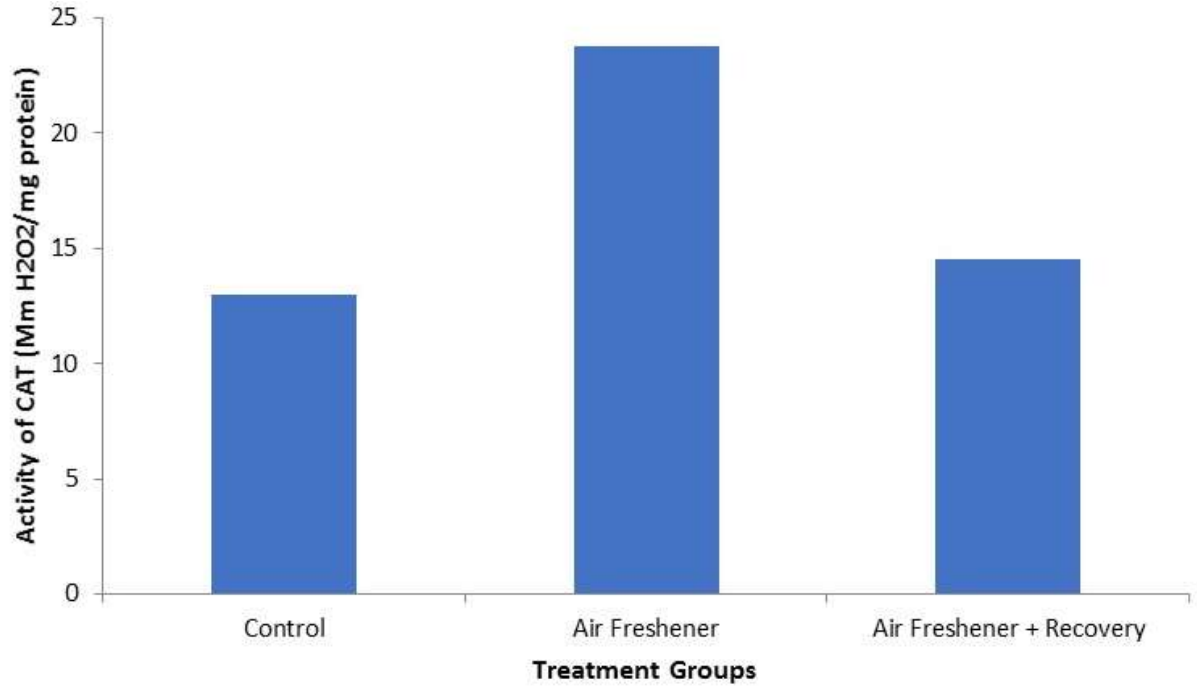

Figure 5. Effect of Air Freshener on the Activity of Catalase (CAT) in Animals after 28 Days of Exposure.

gained by animals has been recorded by Akingbade et al., (2014) after exposing albino rats to incense air fresheners for four weeks. In this present investigation, it was also observed that the activity of the experimental animals in terms of movement within the cage changes from normal to sluggish

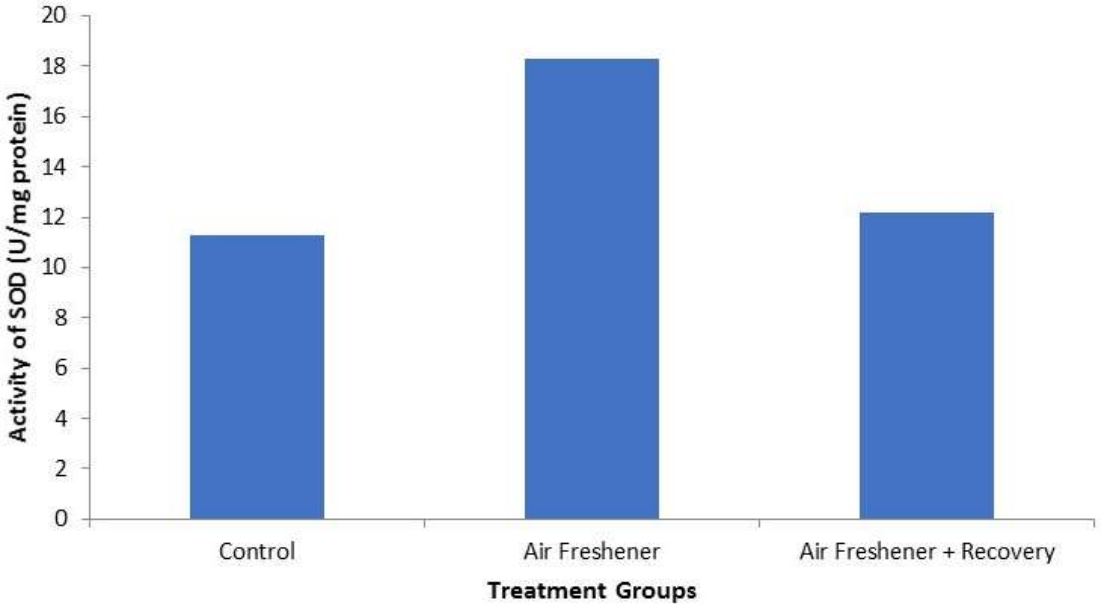

Figure 6. Effect of Air Freshener on the Activity of Superoxide Dismutase (SOD) in Animals after 28 Days of Exposure.

during the period of exposure. This result is also consistent with the observation of Mohammed and Yakasai (2017) who studied the sub-acute toxicity of some brands of air fresheners sold in Kano on Swiss albino rats. This might be due to the effect of fragrance chemicals that causes a host of physical and neurological problems such as dizziness, headache, and fatigue (William, 2004).

Exposure of animals to air freshener significantly elevated the malondialdehyde (MDA) levels in animals indicating enhanced peroxidation and breakdown of the antioxidant defense mechanisms (Airaodion et al., 2020a). Decomposition products of lipid hydroperoxide such as malanoldehyde and 4-hydroxynonenal can cause chaotic cross-linkage with proteins and nucleic acids, which plays an important role in the process of carcinogenesis. Furthermore, extensive damage to tissues in a free radical mediated lipid peroxidation (LPO) results in membrane damage and subsequently decreases the membrane fluid content (Oyenihi et al., 2016). However, this effect was reversed when animals were allowed to recover from the effect of air freshener exposure for 14 days. This was observed as there was no significant difference in level of malondialdehyde in animals exposed to air freshener for 28 days and allowed to recover for 14 days when compared to those in control animals.

Glutathione (GSH) is a tripeptide (L-α-glutamylcysteinol glycine) which is highly abundant in all cell compartments and it is the major soluble antioxidant. Glutathione directly quenches ROS such as lipid peroxides, and also plays a major role in xenobiotic metabolism (Airaodion et al., 2019e). Glutathione detoxifies hydrogen peroxide and lipid peroxide by donating electron to hydrogen peroxide to reduce it to water and oxygen protecting macromolecules such as lipids from oxidation. In this study, the decrease in the reduced glutathione level in animals exposed to air freshener for 28 days might be connected to air pollution-induced oxidative stress and direct conjugation of GSH with reactive intermediates of pollutant oxidation (Airaodion et al., 2020b). The effect of air freshener on glutathione level was sustained when animals were allowed to recover for 14 days (Figure 4). This might be an indication that the toxic chemicals in the air freshener have permanently inhibited the synthesis of glutathione.

In this investigation, air freshener was observed to significantly increase the activities of antioxidant enzymes investigated when compared to those in their respective control group (Figures 5-7). These enzymes include: catalase (CAT), superoxide dismutase (SOD) and glutathione peroxidase (GPx). SOD plays an important role in reducing the effect

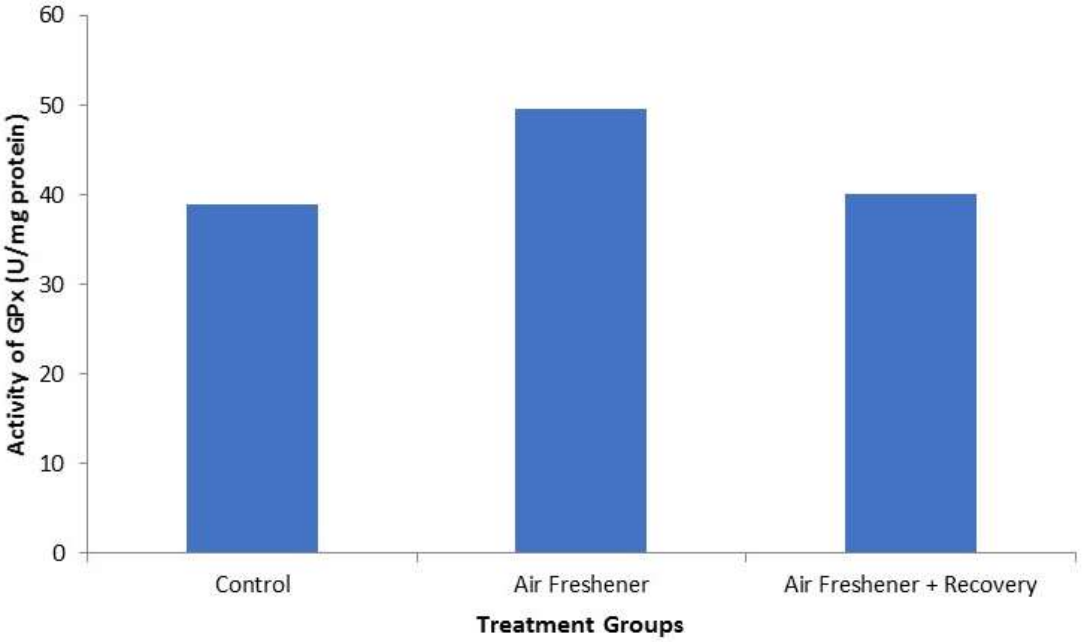

Figure 7. Effect of Air Freshener on the Activity of Glutathione Peroxidase (GPx) in Animals after 28 Days of Exposure.

of free radicals' attack, and SOD is the only enzymatic system quenching O_2- to oxygen and H_2O_2 and plays a significant role against oxidative stress (Airaodion et al., 2019e). These radicals have been reported to be deleterious to polyunsaturated fatty acids and proteins (Airaodion et al., 2019a, 2020b). CAT and GPx are other enzymatic antioxidants that act as a defense mechanism against oxidative stress (Airaodion et al., 2020b). The significant increase in the activities of antioxidant enzymes in animals exposed to air freshener might be an indication that air freshener increased the generation of free radicals which these enzymes tend to combat, thereby increasing their activities (Airaodion et al., 2020a). The mechanism in which air freshener increased the activities of these enzymes is unclear but Airaodion et al., (2020b) reported that common household insecticides significantly increased the activities of these enzymes after 21 days of exposure. Thus, the mechanism of action of air freshener in this study might be similar to that of insecticides. It is interesting to note that the effect of air freshener exposure for 28 days on the activities of antioxidant enzymes was reversed after 14 days recovery period. This was observed as there was no significant difference in the activities of antioxidant enzymes in animals exposed to air freshener for 28 days and allowed to recover for 14 days when compared with those in control animals.

CONCLUSION

This study revealed that sunlight air freshener induced oxidative stress, thus possesses the propensity to destroy the immune system. Frequent use of this air freshener should be discouraged.

Conflict of Interest

Authors wish to declare that no conflict of interest exists in this study and publication.

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