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The Beneficial Impact Of Nano-Piperine On Hormonal Balance And Antioxidant Levels In Female Wistar Rats

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Abstract A research investigation evaluated the beneficial advantages of nano-piperine (administered at 20 mg/kg of body weight) on reducing sodium benzoate's (100 mg/kg of body weight) damaging effects on female hormones and particular antioxidants. The experiment involved forty Wistar rats of female gender at 120 days old with a weight range of 200±10 g which were organized randomly into four equal treatment groups for four weeks. Distilled water was given to the control group (C) in a one-cc solution. A sodium benzoate substance served as the treatment agent for group T1. Nano-piperine constituted the second treatment protocol that the T2 group received. The experimental group receiving treatment T3 received both sodium benzoate and nano-piperine combined within a single dosage. The research trial culminated with the execution of female subjects followed by heart blood extraction for tests measuring serum LH, FSH, oestrogen concentration together with MDA, CAT and GSH levels. Measures of LH, FSH, and Oestradiol showed significant reduction in T1 while T2 and T3 presented higher levels than the control group according to the obtained findings ($p<0.05$). The concentrations of MDA increased remarkably in T1 rats and simultaneously there was a significant decrease in CAT and GSH levels yet T2 and T3 rats exhibited opposing results. The research shows that nano-piperine functions as a protective agent which counteracts sodium benzoate-induced harm to female hormones together with antioxidant balance in adult female Wistar rats.

Key word: Nanopeprine, Antioxidant, Sodium benzoate, female hormones.

INTRODUCTION

Food preservation agents use sodium benzoate as one of their most frequently applied substances. Tropical food safety and protection depend on the bacteriostatic and fungistatic actions of sodium benzoate because it dissolves easily in water (Shahani and Sharma, 2021). The cosmetic and pharmaceutical industries utilize sodium benzoate to produce their products and silage makers apply it as an additive (Baranowska et al., 2014). Food safety programs designed by the Food and Agricultural Organisation accept a 0.1 percent allowance of sodium benzoate. (Lennerz et al., 2015). The research discovered harmful sodium benzoate effects in food products thus recommending a restriction on its usage (Salviano et al., 2015). The severity of sodium benzoate exposure directly influenced the_FUNC_ oxidative stress levels in treated subjects (Dinc et al., 2023). Natural antioxidants have taken the market lead over synthetic antioxidants because consumers prefer them more than chemical antioxidants (Ni et al., 2002). Future research has intensified about functional food constituents due to their recognized health benefits and disease prevention abilities. (Ul-Haq et al., 2019). Black pepper which belongs to the Piperaceae family plays a crucial role in biological studies (Zadorozhna et al., 2019). Piperine represents the majority of black pepper molecules because it functions as an alkaline compound that brings diverse health benefits (Acharya et al., 2012; Ahmad et al., 2012). The substance exists in multiple medical

pharmaceutical formulations used for different disease treatments alongside its application for antiseptic medication and diuretic and digestive enhancement and antibacterial properties and insectical properties (Meghwal et al., 2013). This research study examined whether piperine can protect against sodium benzoate-induced damage to female hormones and antioxidants in adult rats' system.

MATERIAL AND METHODS

ETHICAL APPROVAL

The students have obtained ethical permission from the Ethical permission Committee of Al-Qadisiyah University for the inquiry.

EXPERIMENTAL ANIMALS

The research employed forty Wistar female rats weighing 200±10 grams at their 120-day maturity for four experimental condition analysis. All animals received a period for accommodation within the animal housing before starting the experiment. A well-ventilated room provided polypropylene cages as the housing space for these animals. During the experiment the rats received standard chow while having an unlimited supply of distilled water. The experimental room maintained a temperature between 23 degrees and 2 degrees Celsius under twelve hours of light and twelve hours of darkness periodicity.

EXPERIMENTAL DESIGN:

A total of forty adult female rats received a four-week treatment under four different study conditions that contained ten rats within each group. The investigators created the following treatment groups for their study:

1.The control group (C) received 1 ml of distilled water through gavage needle administration throughout oral intake.

2.Animals in T1 received the sodium benzoate treatment with 100 mg/kg body weight amount dissolved in 1 ml of distilled water according to Boutlelis (2021).

3.The T2 group received Nano-piperine at the dose of 20 mg/kg body weight through administration of 1 ml of distilled water solution (Sahu et al., 2014).

4.The T3 treatment involved giving animals Nano-piperine (20 mg/kg body weight) together with sodium benzoate (100 mg/kg body weight) as a solution with 1 ml of distilled water.

Animal Sacrifice and Sample Collection: The animals underwent sacrifice through anesthesia after the treatment duration concluded. The researchers collected blood samples to obtain serum through centrifugation for testing LH and FSH and determining estrogen levels as well as measuring MDA, GSH, and CAT activities.

ESTIMATION OF HORMONE LEVEL :

The blood levels of FSH, LH, and Oestrogen were quantified using an ELISA test using a kit produced by Cusabio (China).

MARKERS OF OXIDATIVE STRESS AND ANTIOXIDANT PARAMETERS :

The serum levels of MDA, CAT, and GSH were measured using equipment produced by Solarbio (China).

STATISTICAL ANALYSIS

The data were examined using SPSS using a one-way analysis of variance (ANOVA) with the least significant difference (LSD) method.

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RESULTS

SERUM CONCENTRATIONS OF HORMONES

ESTROGEN

The blood oestrogen levels, shown in Fig. (1), significantly reduced ($p<0.05$) in the T1 group of female rats compared to the control group, whereas the T2 and T3 groups exhibited elevated levels ($p<0.05$) relative to the control.

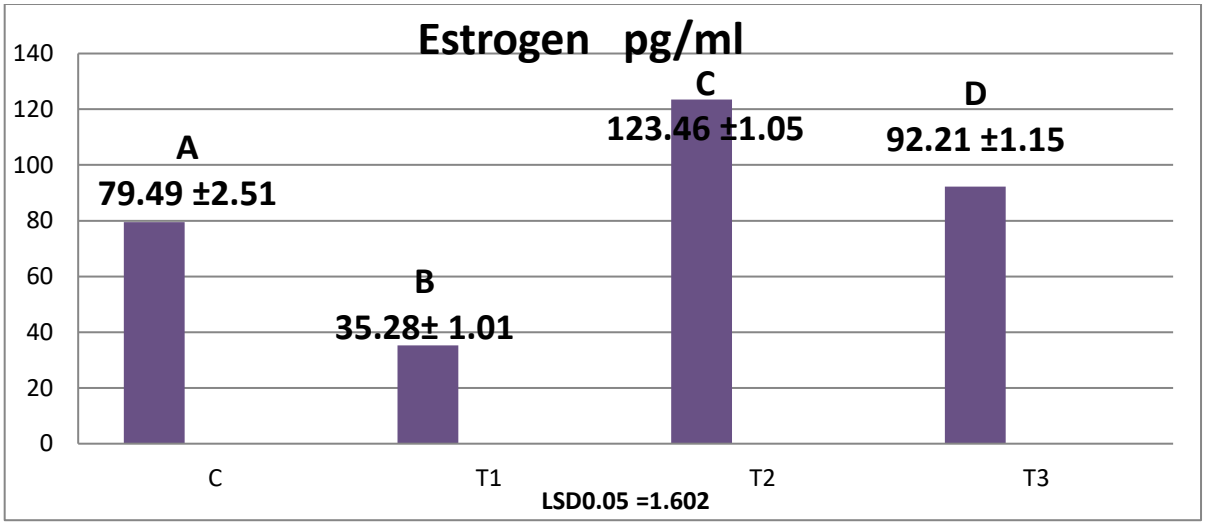

Fig.1 The Influence of Nano-piperine on Estrogen(pg/ml) in Females Rats Treated with Sodium Benzoate.

LUTEINIZING HORMONE (LH)

Figure 2 indicates that the T1 group exhibited the lowest serum LH levels ($p<0.05$) compared to the control female rats, whereas the T2 and T3 groups had the greatest concentrations ($p<0.05$) relative to the control group.

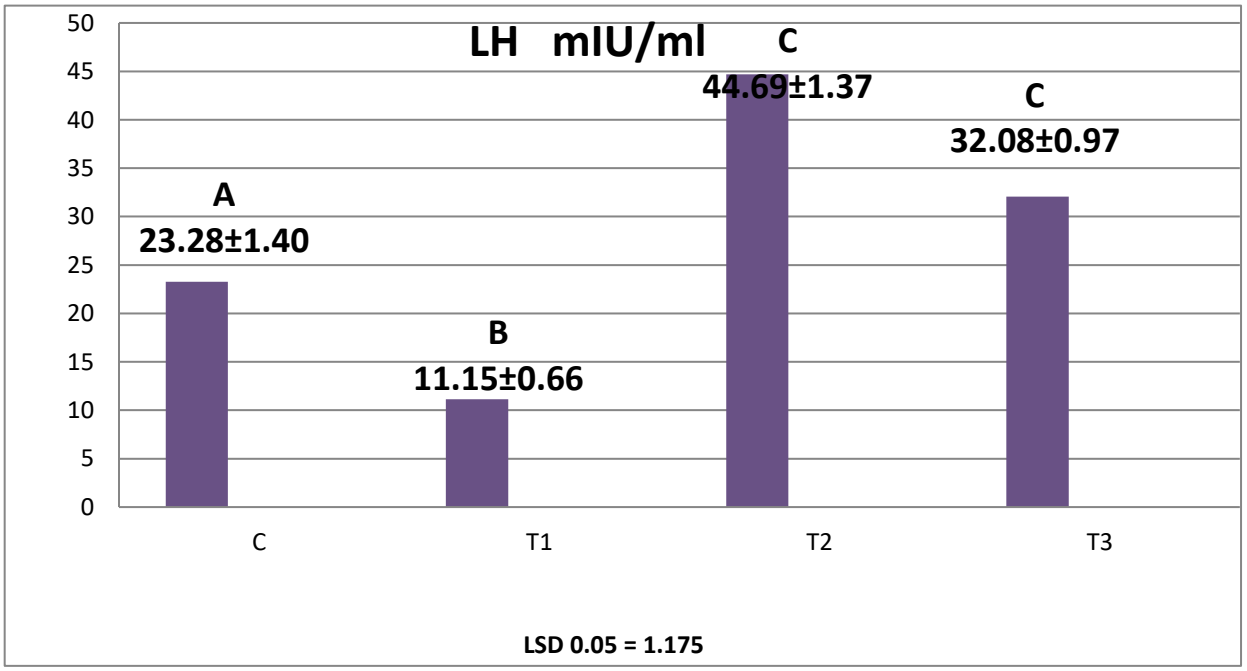

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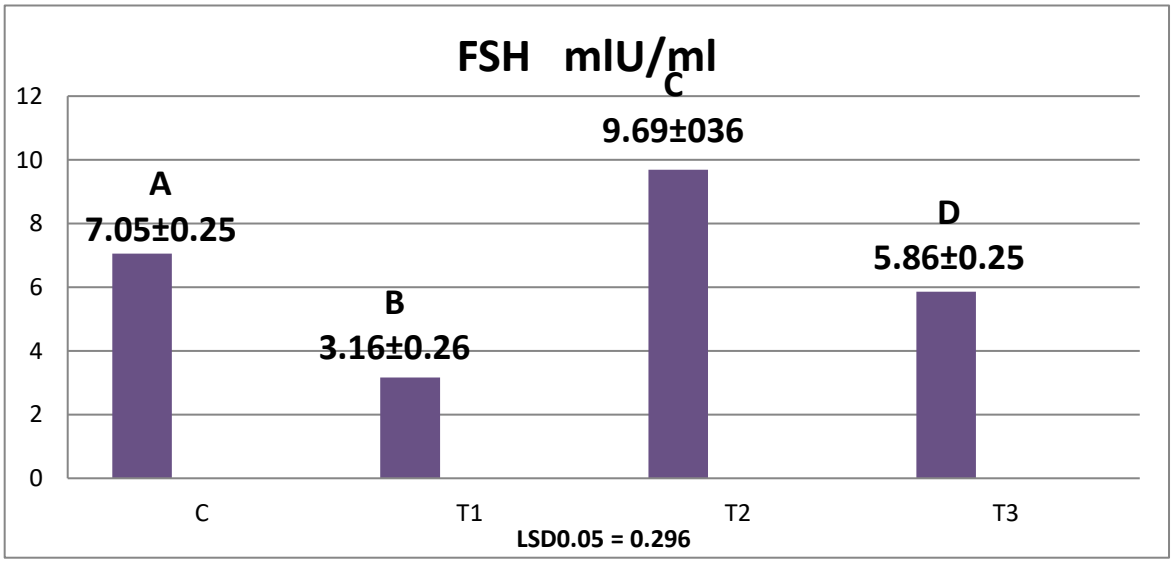
Fig.2 The Influence of Nano-piperine on LH(mlU/ml) in Females Rats Treated with Sodium Benzoate.

FOLLICLE STIMULATING HORMONE (FSH)

The blood FSH levels in T1 female rats of the experiment group measured at a minimum quantity compared to both T1 rats of the control group and T1 rats of the experiment group ($p<0.05$) as depicted in figure 3. The serum FSH concentration in the T2 and T3 groups registered the highest values with statistically significant results compared to the control group ($p<0.05$).

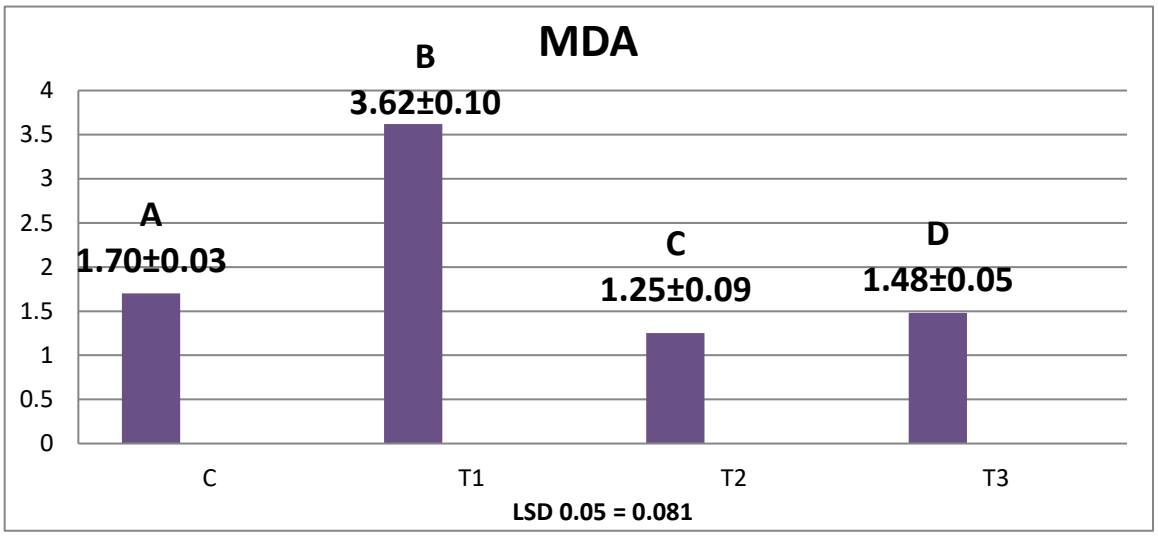
Fig.3 The Influence of Nano-piperine on FSH(mlU/ml) in Females Rats Treated with Sodium Benzoate



SERUM CONCENTRATION OF MALONDIALDEHYDE (MDA)

The results in figure (4) showed that T1 group participants had elevated MDA serum levels above the control values with statistica significance ($p<0.05$). The experimental data from the control group was compared to this set of measurements. T2 and T3 groups showed decreased MDA concentration than the T1 group.

Fig.4 The Influence of nanopiperine on MDA in Females Rats Treated with Sodium Benzoate



SERUM ACTIVITY OF GLUTATHIONE (GSH)

A significant reduction ($p<0.05$) emerged in GSH serum levels of the T1 group when compared to levels observed in the control group according to figure (5). The rats from T2 and T3 groups showed a significant enhancement ($p<0.05$) against the control group measurements.

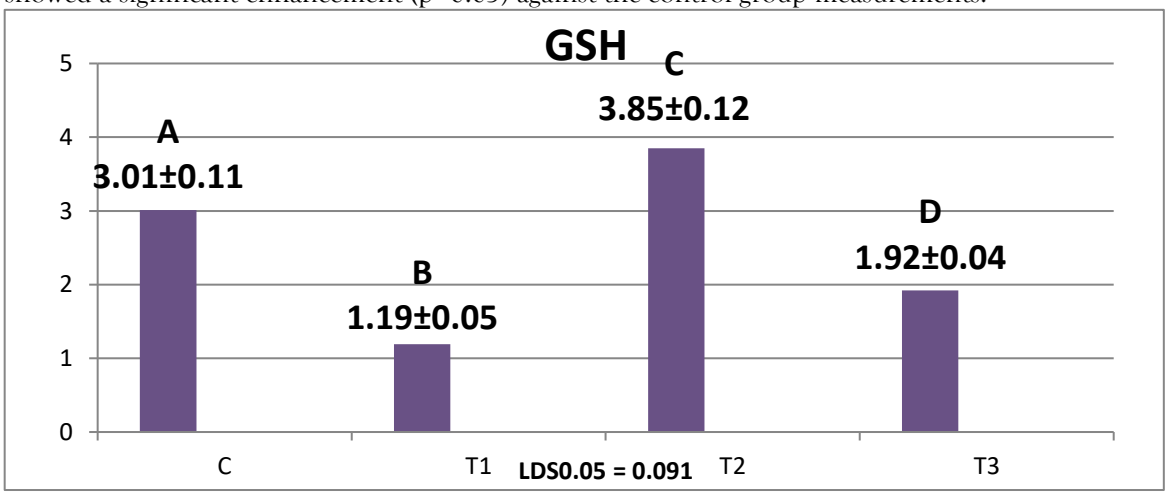


Fig.5 The Influence of nanopiperine on GSH in Females Rats Treated with Sodium Benzoate

SERUM CATALASE CONCENTRATION (CAT)

Data in Figure 6 established a statistically significant decrease ($p<0.05$) of CAT activity within the T1 group versus the control group. CAT activity measured higher levels in T2 and T3 groups than it did in the control group.

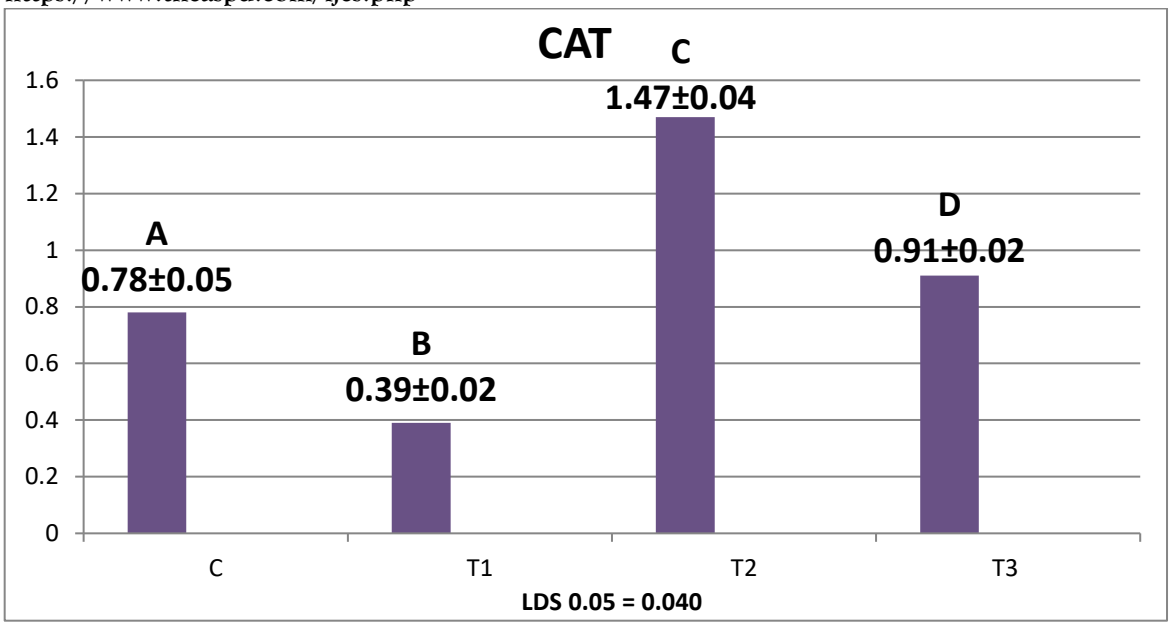


Fig.6 The Influence of nanopiperine on CAT in Females Rats Treated with Sodium Benzoate

DISCUSSION

ESTRADIOL

The research data showed that sodium benzoate consumption by T1 group female rats resulted in notable hormone oestradiol level decreases. The decline of reproductive ability resulted from oxidative stress which reduces the hypothalamus's capacity to release Gonadotropin-releasing hormone (GnRH). A decrease happens in LH and FSH production inside the pituitary gland due to this process and it ultimately damages ovarian operations and sex hormone synthesis (Ojeda and Skinner, 2006). The research data matches data observed by Jyothi (2021).

Female rats given nano-piperine exposure showed a major increase in their oestradiol hormone levels. The authors credited nano-piperine with improving hormone regulation because it enhances LH and FSH levels which lead to elevated ovarian function and subsequent steroid hormone production. The oestradiol levels in T3 group rats were significantly elevated possibly due to the antioxidant activities of nano-piperine. The antioxidant function of nano-piperine becomes stronger because it fights free radicals while controlling oxidative stress which enhances ovarian stimulation processes and boosts oestradiol production.

GONADOTROPINS (LH and FSH)

The results of this training demonstrated a decrease in LH and FSH levels in rats administered sodium benzoate. This result aligns with the findings of the research conducted by El-Shennawy (2020). This outcome may be elucidated by the influence of sodium benzoate on the hypothalamus, which inhibits the synthesis and secretion of gonadotropins or adversely affects the pituitary gland, resulting in an inadequate response to gonadotropin-releasing hormone (GnRH), thereby leading to a reduction in gonadotropins, specifically LH and FSH. Piperine positively impacts oxidative stress by diminishing radicals and lipid peroxidation (Mittal, 2000), which accounts for the increase in LH and FSH levels in the T2 and T3 groups. This outcome aligned with the findings of Mansoor et al. (2023).

OXIDANT-ANTIOXIDANT STATUS

Research findings of Chatterjee (2016) reveal that ROS and antioxidants which generate oxidative stress exhibit an imbalance. The major end product of free radicals stems from MDA which refers to chemical damage of biological components. Polyunsaturated fatty acids in cellular membranes have their double bonds as primary points for free radical-induced damage. Madiepins' acid formation occurs during lipid peroxidative reactions due to free radical-induced oxidation of fatty acids. Tests for chemical indicators of lipid peroxidation provide evidence about stress conditions as well as oxidative damage and unidentified changes in antioxidant functions (Montjean et al., 2010). The experimental T1 group demonstrated oxidative stress by elevated serum MDA levels and reduced GSH and CAT activity that laboratory results confirmed. The consumption of sodium benzoate boosted body lipid damage while reducing natural antioxidant capacity. Dog cells treated with cell benzoate show elevated lipid peroxidation levels accompanied by reduced activities of SOD and glutathione S-transferase together with CAT antioxidant enzymes according to Martina El-Shennawy (2020). Research confirms female rats in T2 and T3 groups presented lower serum MDA readings than controls because piperine behaves as a powerful antioxidant and anti-radical agent which stops the formation of toxins and protects against lipid peroxidation throughout various organs (Mittal, 2000). The study findings establish piperine as an effective substance which offsets the detrimental effects of sodium benzoate. The scientific community has proven piperine serves as an antioxidant chemical that protects cells against peroxidative damage to lipid chains (Vijayakumar et al., 2004). The study results validate the findings reported by Park et al. (2019).

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