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Research

Evaluation of the Anti-hypertensive Activity of Ethanolic Leaves Extract of Euterpe Oleracea Experimental Animal

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Abstract:

Hypertension, or high blood pressure, is a prevalent and serious disorder that is linked to several health consequences, including premature death and cardiovascular disease. It is the primary aetiology for angina, myocardial infarction, stroke, heart failure, and renal failure. According to the World Health Organisation, hypertension is the third leading cause of death globally, responsible for nearly one in eight deaths. Cardiovascular problems provide a considerable contribution to illness and death, especially in the Eastern Mediterranean area, where 26% of individuals have hypertension. Annually, there are four million deaths and billions of people affected. The prevalence of hypertension is increasing as a result of variables such as a growing elderly population, higher rates of smoking, and changes in nutrition and lifestyle. Despite the availability of several antihypertensive drugs, they often lead to undesirable outcomes, including renal and gastrointestinal problems. Consequently, there is a growing fascination in herbal remedies, which are both more economical and have less negative consequences. This research examines the therapeutic benefits of using ethanolic leaf extract from Euterpe oleracea to treat hypertension. Over a span of five weeks, rats were given ethanol orally at a dosage of 5 g/kg/day, leading to an elevation in their blood pressure. Afterwards, the rats were divided into groups that were administered normal saline, ethanol, advanced marine (10 mL/kg), Nifedipine (10 mg/kg), or Euterpe oleracea extract (100 mg/kg and 200 mg/kg). The findings demonstrated that ethanol had a notable impact on arterial pressure and heart rate, whereas the administration of Euterpe oleracea and Nifedipine led to a substantial decrease in mean arterial pressure. Additionally, these treatments resulted in better cholesterol levels, liver and kidney function, as well as antioxidant status in hypertensive rats. The findings suggest that the extract of Euterpe oleracea has strong antihypertensive, hypolipidemic, and antioxidant effects, making it a potential therapy for hypertension

Keyword: *Euterpe oleracea, Anti-hypertension, Histopathology and Serum Concentration of total Cholesterol*

1. INTRODUCTION

Elevated blood pressure may be indicative of a persistent illness and may be accompanied with the first symptoms are indeterminate. Patients are in good health and must attend an outstanding concert. Patients' life is affected by vascular abnormalities, leading to clinical indications and subsequent worries. This illness typically affects many organs throughout the body as an independent risk factor. Health issues that may be faced include heart failure, cerebrovascular illness, chronic renal failure, eye complications, and coronary disease. The frequently used name is derived from the Portuguese translation of the Tupian phrase ĩwasa'i, which means "[fruit that] cries or expels water." The plant's name is said to have originated from local folklore that associates the fruit with its crucial role as a main source of nourishment in the Amazon River estuary. According to folklore, the chief Itaqui ordered the mass

killing of all children due to a shortage. The tree was designated as "açaí" due to its name, Iaçá, being spoken in reverse. The tribe heavily depended on this tree for nourishment.

2. MATERIALS AND METHODS

2.1 Identification, collection and authentication of plant

The selection of leaves was based on their historical usage, and a thorough examination of the available literature was carried out to exclude any prior similar studies. A thorough literature study was undertaken from 1979 to 2018, using diverse sources including the Medicinal and Aromatic Plants Abstracts (MAPA) database, online resources, and published materials.

The leaves of Euterpe oleracea were gathered from Sardhana, District Meerut, Uttar Pradesh. Dr. Vijai Malik, the Head of the Department of Botany at CCS University in Meerut, India, identified and confirmed the effectiveness of the drugs.

2.2. EXPERIMENTAL ANIMALS

Healthy, resilient guy without any diseases the investigation on antihypertensive effect used Wistar rats weighing between 150-200 g. The Institutional Animal Ethics Committee granted permission for the use of these animals. The rats were specifically kept in the animal facility of TIPER, Meerut, for the purpose of this study. Under normal reproductive circumstances, each animal will undergo a seven-day phase of use. Each experimental group shall consist of a distinct group of rats, and every measure will be done to avoid using rats that show a particular reaction in other groups. Rats will undergo a 48-hour acclimatisation period to laboratory settings before testing procedures to reduce any inexplicable stress. The rules for all animal management practices were set by the CPCSEA, a department under the Ministry of Forest and Environment, Government of India.

2.3 Acute Oral Toxicity Study

Prior to administration of the Ethanolic leaves extract (2,500 mg/ml in 10% dimethyl sulfoxide, DMSO) following the OECD guideline for chemical testing 420, the animals were subjected to a fasting period of 16-16 hours without water. The extract was administered orally to rats at a dosage of 2,000 mg per kilogramme of body weight. The vehicle was only allocated to the control group. The animals are monitored for signs of poisoning for duration of 14 days. The research examined the effects of toxin administration on body weight and several markers, such as normal behaviour, respiratory function, heart activity, motor abilities, and changes in skin and hair. The indicators were evaluated on a daily basis for the next 14 days, as well as during the first, second, fourth, and sixth hours after administration. In addition, the rates of death were analysed.

2.4 Evaluation of Hypertension parameters:

2.4.1 BP EVALUATION

In the end, the researchers recorded the heart rate and arterial blood pressure of each rodent. In essence, the animals were rendered incapacitated by administering a peritoneal injection of 1.5g/kg urethane. The trachea was exposed and prepared to enable automatic respiration. Blood pressure was assessed by connecting a cannula vein to a Biopac Student Lab hemodynamic recorder and a weight transducer. This objective was achieved by employing the carotid artery. The measurements were performed using a personal computer and an MP35 device.

2.4.2 SERUM AND ORGAN COLLECTION EXAMINATION TESTS FOR OXIDATIVE STRESS MARKERS:

The subsequent hemodynamic parameters were measured, and plasma samples are obtained from the abdominal circulation and then centrifuged at a speed of 3000 revolutions per minute for duration of 15 minutes. The plasma detection was kept at a temperature of 20 ℃ in a chemical test. Afterwards, the heart was gathered, preserved with salt, measured, and kept for the purpose of assessing Oxidative pressure indicators.

2.4.3 ASSESSMENT OF BIOCHEMICAL PARAMETERS

Total cholesterol, High Density Lipoprotein (HDL), and triglyceride concentrations in blood serum were measured using commercially accessible tools as part of the hyperlipidemia investigation. The units of measurement used were milligrams per deciliter (mg/dl).

2.4.4 Examination of Serum Concentration of total Cholesterol (TC)

To determine total cholesterol levels, the CHOD-POD method—which stands for cholesterol oxide peroxidase technique—was used. The Goa, India-based Coral Clinical System provided the commercially accessible kit that was used for this. In order to get the test ready, combine 1000 µl of cholesterol reagent with 10 µl of normal cholesterol (200 mg/dl), 10 µl of clean water, and 10 µl of serum. All samples, including test, normal, and empty ones, should undergo this procedure. An ad hoc chamber was used to hold the test cylinders. A length of ten minutes. Spectrophotometric analysis made it possible to see the absorption at 505 nm in both the experimental and high-resolution samples.

The serum cholesterol was measured using the formula:

$$\text{Total cholesterol (mg/dl)} = \frac{\text{Absorbance of test}}{\text{Absorbance of standard}} \times 200$$

2.4.5 Serum Triglycerides (TG) Estimation

As part of its glycerol-phosphate oxidase peroxidase (GPO-PAP) blood triglyceride testing, the Coral Clinical System in Goa, India used a commercial kit. In order to carry out the test, a 1000 µl volume of enzyme reagent will be added to 10 µl of serum, 10 µl of normal (200 mg/dl), and 10 µl of clean water, respectively, for testing, standardisation, and blank control. For 10 minutes, the test tubes were safely contained in the makeshift container. Without the presence of any substances, the absorbance and standard samples were determined using spectrophotometry at a wavelength of 505 nm.

Serum triglyceride was measured using the following formula:

$$\text{Triglycerides (mg/dl)} = \frac{\text{Absorbance of test}}{\text{Absorbance of standard}} \times 200$$

2.4.6 High Density Lipoprotein (HDL) Estimation

The HDL levels were determined using the cholesterol oxide peroxidase technique CHOD-POD using a commercially available kit from Coral Clinical Systems, located in Goa, India.

The mixture of 200µl of serum and 300µl of the produced reagent was well mixed in a centrifuge tube before being centrifuged at 3000 rpm for 10 minutes to get the maximum apparent strength. 100µl of the first step's supernatant, 100µl of the 50 mg/dl HDL cholesterol standard, and 100µl of test water (normal and empty) would each be added individually to 1000µl of cholesterol reagent. Ten minutes is all it takes to keep each tube at room temperature. Spectrophotometry was used to measure the absorbance at 505 nm, which produced non-zero results for both the standard experiments and the samples. Centrifugation and an active serum reagent both release lipoprotein, but the HDL fraction stays in the supernatant. Serumtriglyceride is calculated using the following formula:

$$\text{HDL cholesterol (mg/dl)} = \frac{\text{Absorbance of test}}{\text{Absorbance of standard}} \times 50$$

2.4.7 Low Density Lipoprotein (LDL) Cholesterol determination

The LDL was defined by the Friedewald's equation:

2.5 ASSESSMENT OF OXIDATIVE STRESS MARKER

2.5.1 ASSESSMENT OF (MDA) MALONDIALDEHYDE

Using the process of *Wilbur et al.* Malondialdehyde (MDA) was defined.

2.5.2 EVALUATION OF SUPEROXIDE DISMUTASE (SOD)

Using the technique designated by Misra and Fridovich. Superoxide dismutase (SOD) was defined.

2.5.3 ASSESSMENT OF CATALASE

Catalase was defined according to *Sinha*

2.5.4 ASSESSMENT OF GLUTATHIONE (GSH)

The method described by *Ellman.* decreased glutathione (GSH) was assessed

2.6 Statistical Analysis

The data are presented as a textual representation of the standard error of the mean (± SEM). To examine the differences between the groups, a one-way analysis of variance (ANOVA) was employed in conjunction with Tukey's post hoc analysis. In most cases, statistical significance is defined as a p-value below 0.05. Graph Pad Prism 5.03 is the only application used in the review.

3. RESULTS AND DISCUSSION

3.1 PLANT AUTHENTICATION

The selection of leaves was based on their traditional uses, and a thorough literature assessment was carried out to exclude any prior studies that were comparable in nature. A comprehensive literature review was carried out between 1979 and 2018, using books, internet sources, and the Medicinal and Aromatic Plants Abstracts (MAPA). The Euterpe oleracea was obtained from Sardhana, located in the Meerut District of Uttar Pradesh. Dr. Vijai Malik, the Botany Department Chair of CCS University in Meerut, India, verified and confirmed the authenticity of the medications.

3.2 Effects of the Ethanolic leave extract of *Euterpe oleracea* leaves on Heamodynamic parameters in Hypertensive rats*:*

Table 1: results of Ethanolic abstract of *Euterpe oleracea leaves* on hemodynamic parameters in HTR*:*

Parameter	N T R	H T R	Nif 10	E.O. 100	E.O. 200
SBP (mmHg)	118.30± 0.70	178.04± 2.99	124.06±3.34*	105.22±2,90*	116.40±1.23*
DBP (mmHg)	104.37± 2.07	159.24± 2.87	107.44±2.77*	95.32 ± 2.88*	109.21±2.90*
MBP (mmHg)	108.66± 1.62	168.45± 2.55	115.35±0.77*	99.04 ± 2.34*	111.24±1.08*
HR (BPM)	3324.09±2.65	345.22± 2.99	347.63±2.66*	348.34±2.32*	347.09±2.53*

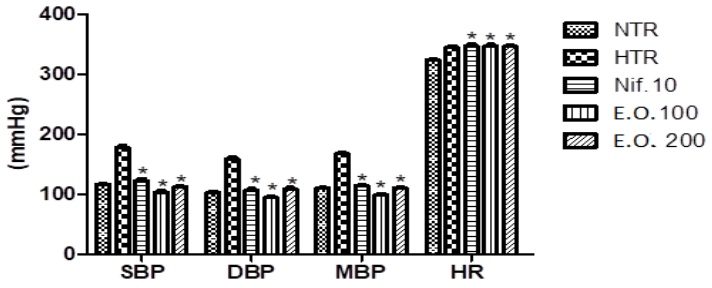

Figure 1: results of Ethanolic abstract of *Euterpe oleracea leaves* on hemodynamic parameters in HTR*:*

Each figure reflects the average value plus or minus the standard error of the average (SEM) for a certain sample size of n = 5. The heart rate (HR) is measured in beats per minute (BPM), whereas blood pressure is measured by diastolic blood pressure (DPM) and systolic blood pressure (SBP). The research used hypertensive rats that received alcohol (5 g/kg) and distilled water (10 ml/kg), as well as normotensive animals that received just distilled water (10 ml/kg). A statistical analysis revealed a significant disparity (*P<0.05) between the two groups, indicating that the impact of alcohol was more noticeable in hypertension rodents compared to normotensive rats. The name "E. O. 100" refers to a group of mice with hypertension that were given a dosage of 100 mg/kg of Euterpe oleracea leaf

extract. E. O. 200 refers to a separate set of rats with high blood pressure that were given a dosage of 200 mg/kg of Euterpe oleracea leaf extract. A group of rats with high blood pressure were given Nifedipine at a dosage of 10 mg per kilogramme, known as Nif 10.

3.3 Results of Ethanolic leave extract of *Euterpe oleracea* *leaves* on Lipid Parameters in HTR*:*

Table 2: results of Ethanolic extract of *Euterpe oleracea* leaves on Lipid parameters in Hypertensive rats.

Parameter	Cholesterol (mg/ml)	TG (mg/ml)	LDL (mg/ml)	HDL (mg/ml)
NTR	151.22 ± 8.99	66.45 ± 0.72	77.04 ± 9.62	61.08 ± 2.40
HTR	226.00 ± 7.32	103.40 ± 0.46	176.22 ± 5.68	30.00 ± 0.56
Nif.10	155.84 ± 8.66*	70.99 ± 0.33*	90.10 ± 9.45*	53.02 ± 1.35*
E.O. 100	186.04 ± 7.88*	68.88± 1.97*	140.08± 7.82*	32.01 ± 0.67*
E.O. 200	178.56 ± 6.92*	67.14 ± 0.21*	123.66 ± 7.07*	42.22 ± 0.73*

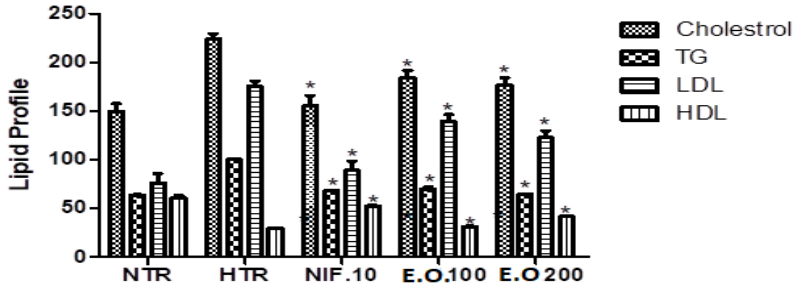

Figure 2: The mean±SEM (n = 5) is represented by each result. The difference from normotensive rodents is statistically significant, as evidenced by a p-value of less than 0.05.

3.4 Results of *Euterpe oleracea* leaves Ethanolic leaves extract on Markers of Oxidative stress parameters in HTR:

Table 3: Effects of *Euterpe oleracea* on the heart of HTR:

Parameters	MDA (nmol/g of tissue)
NTR	0.628 ± 0.066
HTR	0.733 ± 0.061
Nif.10	0.706 ± 0.042*
E.O. 100	0.661 ± 0.031*
E.O. 200	0.421 ± 0.012*

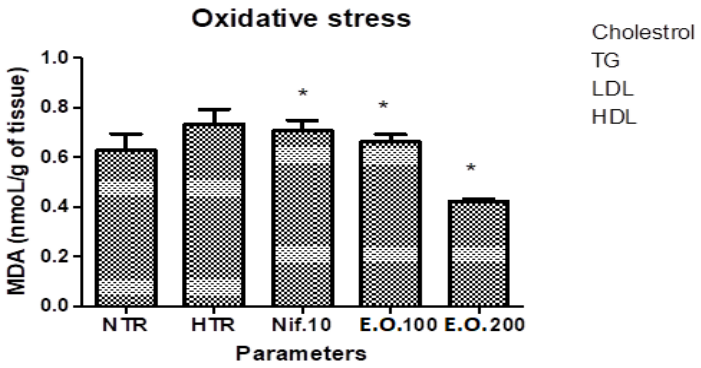

Figure 3: Each result reflects the average value plus or minus the standard deviation of the average value, based on a sample size of 5. *: p <0.05 denotes a statistically significant alteration in comparison to normotensive rats

3.5 SUPEROXIDE DISMUTASE (SOD):

Table 4: The impact of SUPEROXIDE DISMUTASE (SOD) activity on the heart of hypertensive rats:

LIMITS	SOD (Unit /mg protein)
NTR	5.62 ± 5,01
HTR	22.13 ± 1.92
Nif.10	50.44 ± 4.49*
E.O. 100	39.52± 3.54*
E.O. 200	33.82 ± 3.77*

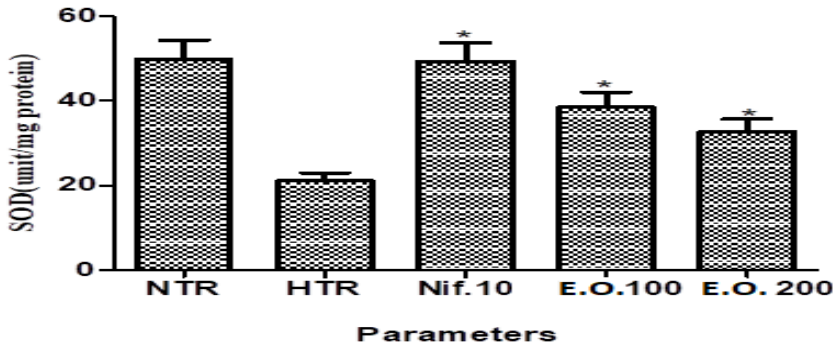

Figure 4: Each number represents the mean value plus or minus the standard error of the mean (n = 5). *: p <0.05 indicates a significant deviation from the normotensive rats in terms of variance.

3.6: CATALASE (CAT)

Table 5: Effect of CATALASE (CAT) activity in the Cardiac of HTR:

Parameters	Catalase (Unit /mg protein)
NTR	9.03± 0.86
HTR	3.10± 0.09
Nif.10	13.21 ± 1.09*
E.O. 100	6.06 ± 0.47*
E.O. 200	4.23 ± 0.25*

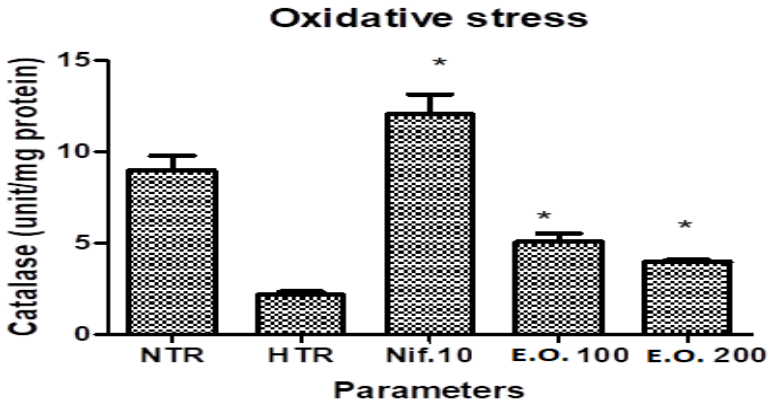

Figure 5: Each value signifies the mean±SEM (n = 5). *: *p* <0.05 substantial variance from normotensive rats.

3.6 The impact of GLUTATHIONE (GSH) activity on the heart of rats with hypertension:

CATALASE	GSH (Mm/g tissue)
NTR	1.97 ± 0.18
HTR	1.28 ± 0.12
Nif 10	1.83± 0.21*
E.O. 100	1.65 ± 0.19*
E.O. 200	0.60 ± 0.33*

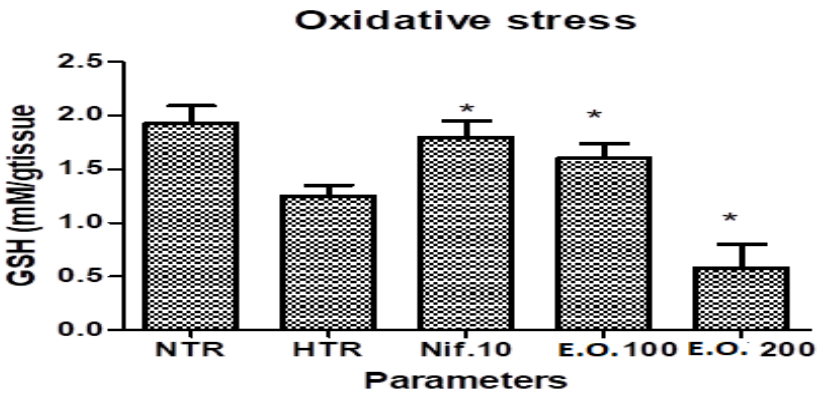

Figure 6: With n = 5, each value represents the mean±SEM. *: Significant difference from rats with normal blood pressure ($p < 0.05$).

4. DISCUSSION

The study investigated the antihypertensive effects of ethanolic leaf extracts from Euterpe oleracea on rats who were given ethanol. Alcohol has been shown to worsen the hemodynamic factors that contribute to high blood pressure (HBP). Administering ethanol at a dose of 5 grammes per kilogramme per day for a period of 5 weeks led to a notable rise in systolic, diastolic, mean blood pressure, and coronary heart rate in this study. The deployment of HTR is the outcome of a limited set of elements. Chronic alcohol intake has been shown to have a negative impact on the proper operation of the heart and blood arteries, and is a significant factor in causing high blood pressure. Ethanol has particular and detailed effects on the arteries, leading to constriction and fragmentation of the flexible layers, which in turn affects the flexibility of the blood vessels. This has been shown in several investigations. Ethanol-induced vascular dysfunction may lead to a decrease in nitric oxide production and a shift in the endothelium towards a relaxed state. When hypertensive rats are given Nifedipine together with Euterpe oleracea leaf extract, their blood pressure decreases significantly. The findings of our study indicate that the ethanolic extracts derived from the leaves of Euterpe oleracea has the capability to efficiently regulate high blood pressure caused by cross-border activities.

No rise in blood pressure was detected in persons who were not taking medication for hormone replacement therapy (HTR). This lack of increase in blood pressure was accompanied with a decrease in body weight and levels of cardiac proteins in the heart. This might be attributed to the deleterious effects of alcohol on cellular architecture. Prior study has showed that alcohol hinders the absorption of vitamins by inducing the death of stomach-protecting cells and affecting the digestive tract. Alcohol hinders the release of chemicals by decreasing the flow of bile via the pancreas. Unlike hypertensive mice given plain water, the administration of Ethanolic leaves extract of Euterpe oleracea leaves resulted in elevated protein levels in several organs and caused weight gain at the end of the therapy, comparable to the effects of Nifedipine. The findings suggest that the ethanolic extract of Euterpe oleracea leaves may protect the cellular structure from the harmful effects of atoms found in ethanol metabolism. Dyslipidemia is a significant and impartial factor that increases the chance of developing coronary heart disease. Atherosclerosis is promoted by a significant reduction in high-density lipoprotein (HDL) levels and an elevation in

triglycerides, total cholesterol (TC), and low-density lipoprotein (LDL) levels in animals exposed to ethanol. The findings of this research indicate that the progression of hypertension, induced by prolonged intake of liquor and sugar, is linked to increased levels of LDL cholesterol and disturbed lipid profiles. Dyslipidemia is a leading cause of high blood pressure and a major risk factor for the development of atherosclerosis.

This is relevant to our experiment, which includes the use of a chemical in unusual amounts that promotes the development of fatty deposits in arteries. The ethanol-based extract derived from the leaves of Euterpe oleracea has been shown to lower levels of blood triglycerides, total cholesterol, and LDL cholesterol, while simultaneously boosting the concentration of HDL cholesterol. This effect is comparable to that of Nifedipine. The findings suggest that extracts from Euterpe oleracea leaves contain chemicals that might be effective in treating dyslipidemia in hypertension caused by ethanol. Oxidative stress is often associated with chronic diseases, such as hypertension.

Oxidative stress and higher levels of free radicals, which are linked to excessive alcohol intake, are thought to have a substantial impact on the development of high blood pressure (HBP). An investigation of oxidative stress thresholds reveals a significant rise in malondialdehyde levels in the plasma of rats exposed to alcohol compared to those subjected to untreated water. MDA is a reliable measure of lipid peroxidation on HTR and reflects the process of cell tissue remodeling, leading to increased protein deficiency in the bloodstream.

The tissue glutathione levels in hypertensive rodents showed a significant decline. Oxidative stress is mainly induced by the production of oxygen-responsive molecules, which is closely linked to the process of alcohol metabolism. Glutathione depletion is the term used to describe the decrease in non-enzymatic antioxidants when they are in their oxidized state. This mechanism works by directly capturing reactive oxygen species.

The research found a clear correlation between the stress caused by ethanol and the reduction in glutathione (GSH) levels. Ethanol facilitates the formation of acetaldehyde and other reactive chemicals inside the cell. The use of herbal extracts and high-temperature treatment (HTR) indicates that the concentrations of glutathione (GSH) are increased and malondialdehyde (MDA) is decreased in tissues. This suggests that the extract derived from Euterpe oleracea leaves may work by directly scavenging free radicals and decreasing their formation, thereby lowering the consumption of GSH. The observed outcome is a direct consequence of adhering to various activities, which have shown a clear connection between the antioxidant properties of certain plant extracts and an elevation in GSH levels. Our studies further show that ethanol significantly decreases the activity of catalase and superoxide dismutase (SOD). Catalase and superoxide dismutase (SOD) protect cells against the harmful effects of highly reactive oxygen species, such as O2- and H2O2. The overproduction of reactive oxygen species (ROS) is likely the main reason for the decline in their efficacy. The antioxidant status in HTR was improved by the administration of Nifedipine and the ethanolic extract of Euterpe oleracea leaves. The findings suggest that the leaves of Euterpe oleracea possess antioxidants that protect the delicate tissue from the harmful impact of free radicals produced during ethanol metabolism.

6. Conclusion

The research demonstrated the significant antioxidant and antihypertensive effects of ethanolic leaf extracts from Euterpe oleracea in hypertensive animals caused by ethanol treatment. The findings indicated that long-term intake of ethanol led to elevated systolic, diastolic, and mean blood pressure, together with an increased heart rate, thereby playing a role in the development of hypertension. The administration of Euterpe oleracea leaf extract led to a significant drop in blood pressure measures and an enhancement in lipid profiles, characterised by a reduction in total cholesterol, LDL, and triglycerides, along with an elevation in HDL values. In addition, the extract reduced the harmful effects of ethanol-induced oxidative damage by lowering the levels of malondialdehyde (MDA) and boosting the activity of glutathione (GSH), superoxide dismutase (SOD), and catalase. This resulted in an improvement in oxidative stress indicators. The results suggest that the extract from Euterpe oleracea leaves has strong characteristics in reducing high blood pressure, lowering cholesterol levels, and preventing oxidative damage. This makes it a promising natural treatment option for controlling hypertension and its related consequences. Further study is required to explore the possible therapeutic uses and underlying mechanisms of action.

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