

# Evaluation of the activity of trans-Resveratrol alone and in combination with Amlodipine and Pioglitazone against Fructose induced metabolic syndrome rats

T. Anila<sup>1</sup>, A. Sudheer<sup>1\*</sup>, B. Mary Vishali<sup>1</sup>, K. Somasekhar reddy<sup>1</sup>, I. Sai Reddemma<sup>1</sup>, S. Akkulanna<sup>2</sup>, M. Sri Ramachandra<sup>3</sup>, Khusali soni<sup>4</sup>

<sup>1</sup>Department of Pharmacology, Raghavendra Institute of Pharmaceutical and Educational Research (RIPER), Anantapuramu, Andhrapradesh, India.

<sup>2</sup>Department of Botany, Phytomedicine division, Sri krishnadevaraya University, Anantapuramu, Andhra Pradesh, India.

<sup>3</sup>Department of Pharmacology, Bhaskar pharmacy college, Moinabad, Telangana, India.

<sup>4</sup>Department of Pharmacy, The M. S. University of Baroda, Vadodara, India.

Email: sudeerlegend@gmail.com

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## Abstract

Metabolic syndrome (MS) is a cluster of conditions that cause an increase in the risk of diabetes, heart disorders, and stroke.

The present research was completed in Wistar rats, in which Metabolic Syndrome (MS) was induced with a High Fructose Diet. Animals were randomly divided into 7 gatherings and the test group animals received Resveratrol (RSVT), Amlodipine (AML), Pioglitazone (PIO), Resveratrol+Amlodipine, and Resveratrol+Pioglitazone at different doses for 5 weeks. Various behavioral, biochemical, and histopathological parameters were estimated.

AML alone and along with RSVT was found to reduce diastolic and systolic pressures, there was the reduction in BP in the remaining groups. There was a significant reduction in serum insulin and Fasting glucose level (FGL) in all the treatment groups. And there was a noticeable reduction in the levels of total glycerides (TG), total cholesterol (TC) along with LDL, HDL, and VLDL when compared to the control group and HFD group. Histopathological study revealed that there was a reduction in the deposition of lipids in liver cells and aorta as compared to HFD group.

The outcomes showed the defensive mechanism of Resveratrol against fructose-induced Metabolic Syndrome. The mechanism of protection may be due to an escalation of cellular antioxidants. The activity was found to increase in combination with amlodipine and pioglitazone.

**Keywords:** Amlodipine, Fructose, Metabolic syndrome, Pioglitazone, Resveratrol.

## INTRODUCTION

International Diabetes Federation (IDF), National Cholesterol Expert Program Adult Treatment Program III (NCEP ATP III), World Health Organization (WHO)1, and harmonized criteria were used to define the term "metabolic syndrome", these criteria include central obesity, elevated triglycerides, reduced high-density lipoprotein (HDL), raised blood pressure (BP), and fasting plasma glucose (FPG) or fasting glucose levels (FGL)2. Diagnostic criteria for metabolic syndrome often include central obesity and any two of the risk variables3.

Reaven initially coined the term metabolic syndrome (MS), also known as syndrome X, in 1988 to refer to the presence of atherogenic risk factors and underlying insulin resistance. The World Health Organization (WHO) improved the definition in 1997 to refer to a particular grouping of risk factors for type 2 diabetes and cardiovascular diseases, including abdominal obesity, high blood pressure, atherogenic dyslipidemia, stroke, cardiovascular disease, hyperglycemia, insulin resistance, hyperuricemia, and proinflammatory state4,5,6.

The WHO definition stipulates the presence of three or more of the following factors: fasting blood glucose >110 mg/dl, blood pressure >130/85 mm Hg, triglycerides >150 mg/dl plasma, high-density lipoproteins (HDL) 40 mg/dl plasma, and waist circumference >102 cm for men and >88 cm for women. In the world, metabolic syndrome affects more than 25% of the population<sup>7,8</sup>. The change in dietary habits, particularly the increased intake of simple sugars, primarily fructose, which are frequently used in sugar-sweetened drinks and the food industry, is one of the most significant factors contributing to the rising prevalence of MS, obesity, and type 2 diabetes mellitus around the world<sup>9,10</sup>. Rats have frequently been used as models for high fructose intake. High fructose feeding causes hypertension, hypertriglyceridemia, insulin resistance, and hyperinsulinemia in these models<sup>11</sup>.

Environmental and genetic variables are both involved in the multifactorial etiology of metabolic syndrome. The combination of a modern sedentary lifestyle with a diet high in fat and low in dietary fiber, bioactive compounds, and micronutrients accelerates the development of metabolic syndrome. Therefore, preventing metabolic syndrome with weight loss, a good diet, exercise, medication, and bariatric surgery is crucial. However, most drugs on the market are ineffective and linked to negative drug reactions<sup>3</sup>.

A stilbenoid, or natural phenol, called resveratrol (3,5,4'-trihydroxy-trans-stilbene), is a phytoalexin produced by several plants in reaction to injury or when the plant is being attacked by pathogens like bacteria or fungi<sup>12,13</sup>. The skin of grapes, blueberries, raspberries, mulberries, and peanuts are among the food sources of resveratrol<sup>14,15</sup>. There is no solid proof that resveratrol lengthens lifespan or significantly affects any human disease, even though it is frequently used as a dietary supplement and investigated in lab models of human diseases<sup>16</sup>.

There is insufficient proof that resveratrol affects human metabolic syndrome. There isn't much support for using resveratrol to treat diabetes, according to an analysis<sup>17</sup>. A meta-analysis found scant support for resveratrol's potential impact on diabetes biomarkers<sup>18</sup>.

In one review, there was insufficient proof that resveratrol helped diabetics with fasting plasma glucose levels. According to two reviews, resveratrol administration may lower body weight and body mass index, but not fat mass or total blood cholesterol<sup>19</sup>. Resveratrol supplementation may lessen the indicators of inflammation of TNF- and C-reactive protein, according to a 2018 review<sup>20</sup>.

The body may respond to resveratrol in a variety of ways, including by widening blood arteries and decreasing blood clotting. Additionally, it might lessen pain and swelling, lower blood sugar levels, and support the body's immune system. The most typical illnesses for which resveratrol is suggested are high cholesterol, cancer, heart disease, and many more.

## MATERIALS:

Sources of fine chemicals:

Fructose & casein were purchased from Hi-media Ltd., Baroda, coconut oil was purchased from National Chemicals, Baroda. Trans-resveratrol & Amlodipine were obtained from Zydus Research Centre, Ahmedabad. All other reagents and chemicals obtained were of analytical grade.

Drugs:

Trans-resveratrol (RSVT) was dissolved in DMSO (dimethyl sulphoxide) & suspended in saline so that the final concentration of DMSO would not exceed 5%. Amlodipine (AML) and Pioglitazone (PIO) were dissolved in DMSO & suspended in saline so that the final concentration of DMSO would not exceed 5%. They were given orally.

## METHODS:

### Animals:

All experiments and protocols described in the present study were approved by the Institutional Animal Ethics committee (IAEC) of M. S. University, Baroda and with the permission from Committee for Control and Supervision of Experiments on Animals (CPCSEA), Ministry of Social Justice and Empowerment, Government of India. Healthy adult rats of either sex (200-250 gms) were used for the study. Rats were housed in small cages with free access to food and water ad libitum. During the period of the experiment, the animals were fed the standard laboratory diet, unless specified. Healthy rats of either sex (200-250gm) were reused. Animals were placed in small cages and maintained under standardized conditions (12-hr light/dark cycle, 24°C, 35 to 60% humidity) and provided free access to a pelleted CHAKKAN diet (Nav Maharashtra Oil Mills Pvt. Ltd., Pune) and purified drinking water ad libitum unless specified.

### EXPERIMENTAL DESIGN:

Rats (n=30) were randomized into the following groups.

1. Control: They were administered DMSO (DMSO in saline, final conc. of DMSO was 5%) p.o for 5 weeks.
2. High Fructose Diet (HFD): They were administered DMSO (DMSO in saline, final conc. of DMSO was 5%) along with HFD (60% fructose, 20% casein, 5% coconut oil, 15% std chow diet) & water ad libitum for 5 weeks.
3. HFD + Resveratrol (RSVT): They were administered RSVT (20mg/kg/day) p.o along with HFD & water ad libitum for 5 weeks.
4. HFD + Amlodipine (AML): They were administered Amlodipine (10mg/kg/day) p.o along with HFD & water ad libitum for 5 weeks.
5. HFD + RSVT + AML: They were administered RSVT (20 mg/kg/day) p.o & amlodipine (10mg/kg/day) p.o along with HFD & water ad libitum for 5 weeks.
6. HFD + Pioglitazone (PIO): They were administered pioglitazone (20 mg/kg/day) p.o along with HFD & water ad libitum for 5 weeks.
7. HFD + RSVT + PIO: They were fed administered RSVT (20 mg/kg/day) p.o & pioglitazone (20mg/kg/day) p.o along with HFD & water ad libitum for 5 weeks.

### Estimation of hemodynamic parameters:

At the end of the study, blood pressures were monitored indirectly in conscious, prewarmed, slightly restrained rats by the tail-cuff method. All the animals were acclimatized in PANLAB Non-Invasive Tail Cuff Pressure Recorder for blood pressure measurement for 7 days during the end of the treatment. At the time of measurement, five individual readings were obtained in a rapid sequence. The highest and the lowest readings were not counted, and the average of the remaining three readings was accepted as the measurement.

### Serum parameters:

The blood samples were withdrawn from the retro-orbital plexus under light ether anesthesia without any anticoagulant and allowed to clot for 10 minutes at room temperature. It was centrifuged at 2500 rpm for 20 minutes. The serum obtained was kept at 4°C until used. The separated serum was used to estimate various biochemical parameters such as uric acid (Reckon Diagnostics Pvt. Ltd, India.), glucose (Beacon Diagnostics Pvt. Limited), serum triglycerides (TG), total cholesterol (TC), low-density lipids (LDL), and High-density lipids (HDL) were estimated by a kit obtained from Span Diagnostics Ltd. Total lipids (TL) (lipids react with vanillin in the presence of sulphuric and phosphoric acid to form a pink-colored complex). Estimation of very low-density lipids (VLDL)-cholesterol was done using the Fried Ewald formula: VLDL cholesterol = triglycerides / 5.

#### Estimation of serum insulin:

The amount of insulin in rat serum was estimated by ELISA kit as per the instruction of Merckodia ELISA kits.

#### Antioxidant Parameter Study:

The liver, kept in cold conditions (precooled in an inverted Petri dish on ice) was removed. It was cross-chopped with a surgical scalpel into fine slices and was chilled in the cold 0.25 M sucrose, quickly blotted on a filter paper. The tissue was minced and homogenized in 10 mM Tris-HCl buffer, pH 7.4 (10%w/v) with 25 strokes of tight Teflon pestle of glass homogenizer at a speed of 2500 rpm. The prolonged homogenizer under hypotonic conditions was designed to disrupt as far as possible the ventricular structure of the cells to release soluble protein and leave only membrane and nonvascular matter in a sedimental form. The clear supernatant was used for other enzyme assays.

#### Superoxide Dismutase (SOD):

SOD was estimated by the method of Mishra and Vidovich, 1972. To 0.5ml of dilute brain homogenate, added 0.5ml of ethanol followed by the addition of 0.15ml of chloroform. Shake for 1min, then Centrifuge at 2000rpm, and separate supernatant. Take 0.5ml of supernatant, add 1.5ml of carbonate buffer, and 0.5ml Of Ethylenediamine tetraacetic acid (EDTA).

The reaction was initiated by the addition of epinephrine and the change in optical.

Density/min was measured at 480 nm, taken for 3 min at 30-second intervals and results are reported in Units/ mg of protein.

#### Catalase:

Catalase was estimated by the method of Hugo E. Abe, 1987. To 2ml of diluted homogenate added 1ml of phosphate buffer pH 7 followed by the addition of 1ml of Hydrogen Peroxide (H<sub>2</sub>O<sub>2</sub>). Add H<sub>2</sub>O<sub>2</sub> just before taking absorbance at 240 nm and take the reading for 3 min with a 15-second interval and report the results in units  $\mu$  mol of H<sub>2</sub>O<sub>2</sub> evolved/ mg of protein.

#### Reduced glutathione:

Reduced glutathione (GSH) was estimated by the method of Moran et. al, 1979. To 1ml of homogenate added 1ml of Tri Chloro acetic acid (TCA) (10%). Cool for 10 min and centrifuged at 2000 rpm take 0.5 ml of supernatant. Take 0.5ml of supernatant, and add 4 ml 5-5'-dithiobis-2-nitrobenzoic acid (DTNB)+1.5 ml Phosphate buffer. Mix well and keep at room temperature. Read the absorbance against blank at 412 nm using a spectrophotometer and report the results in  $\mu$ g of GSH / mg of protein.

#### Lipid peroxidation:

Malondialdehyde formation (MDA) was estimated by the method of Slater and Sawyer, 1979. To 2ml of homogenate added 2 ml of TCA. Cool for 15 min and centrifuged, take the supernatant. Take 2ml of supernatant and, add 2ml of Thiobarbituric acid (TBA).

Keep in boiling water bath for 10 min. Read the absorbance of the test against blank at 535 nm using a spectrophotometer. The results are reported in nm of MDA / gm of tissue.

#### Histopathology:

Thoracic aortas & liver were collected after the rats were sacrificed. After blotting free of blood and tissue fluids. Paraffin-fixed tissues were made into 5-15 $\mu$ m thick sections on a Leitz microtome in a horizontal plane, stained by eosin and hematoxylin. Later, sections were mounted on a glass slide. The sections were observed and desired areas were photographed in an Olympus photomicroscope for morphometric studies of different cells. The sections were viewed under 40X and 100X magnification.

## RESULTS:

### Hemodynamic study:

There was a significant ( $P < 0.05$ ) increase in diastolic B.P of fructose-treated animals as compared to control animals. Trans-Resveratrol (RSVT) was found to reduce diastolic B.P significantly. Amlodipine (AML) significantly ( $p < 0.01$ ) reduced diastolic B.P. as compared to fructose treated group. RSVT combined with AML was also found to reduce diastolic B.P significantly ( $p < 0.01$ ). Pioglitazone (PIO) alone & PIO in combination with RSVT were reducing diastolic B.P significantly as compared to fructose-treated animals but not as AML group (fig 1).

There was a significant ( $P < 0.001$ ) increase in systolic B.P of fructose-treated animals as compared to control group animals. RSVT was found to reduce systolic B.P significantly. However, AML significantly ( $p < 0.001$ ) reduced systolic B.P. as compared to fructose treated group. RSVT combined with AML also was found to reduce systolic B.P significantly ( $p < 0.01$ ). PIO alone & PIO in combination with RSVT were reducing systolic B.P significantly as compared to fructose-treated animals (fig 1).

### Serum Parameters:

There was a significant increase ( $p < 0.001$ ) in serum fasting glucose level (FGL) level in HFD group as compared to the control group. RSVT was found to decrease serum FGL significantly ( $p < 0.01$ ) as compared to HFD group. AML alone & in combination with RSVT were found to reduce serum FGL significantly ( $p < 0.001$  &  $p < 0.01$ , respectively) as compared to HFD group. PIO alone & in combination with RSVT were found to reduce serum FGL significantly ( $p < 0.001$  in both groups) as compared to HFD group.

There was a significant increase ( $p < 0.001$ ) in serum insulin level of HFD group as compared to the control group. RSVT was found to decrease serum insulin significantly ( $p < 0.001$ ) as compared to HFD group. AML alone & in combination with RSVT were found to reduce serum insulin significantly ( $p < 0.001$  in both groups) as compared to HFD group. PIO alone & in combination with RSVT were found to reduce serum insulin significantly ( $p < 0.001$  in both groups) as compared to HFD group (fig 1).

In the case of serum uric acid, no significant increase was found in HFD group as compared to the control group (fig 1).

There was a significant increase ( $p < 0.001$ ) in serum TG level in HFD group as compared to the control group. RSVT was found to decrease serum TG significantly ( $p < 0.01$ ) as compared to HFD group. AML alone & in combination with RSVT were also found to reduce serum TG significantly ( $p < 0.05$  &  $p < 0.001$  respectively) as compared to HFD group. PIO alone & in combination with RSVT were found to reduce serum TG significantly ( $p < 0.001$  in both groups) as compared to HFD group (fig 1).

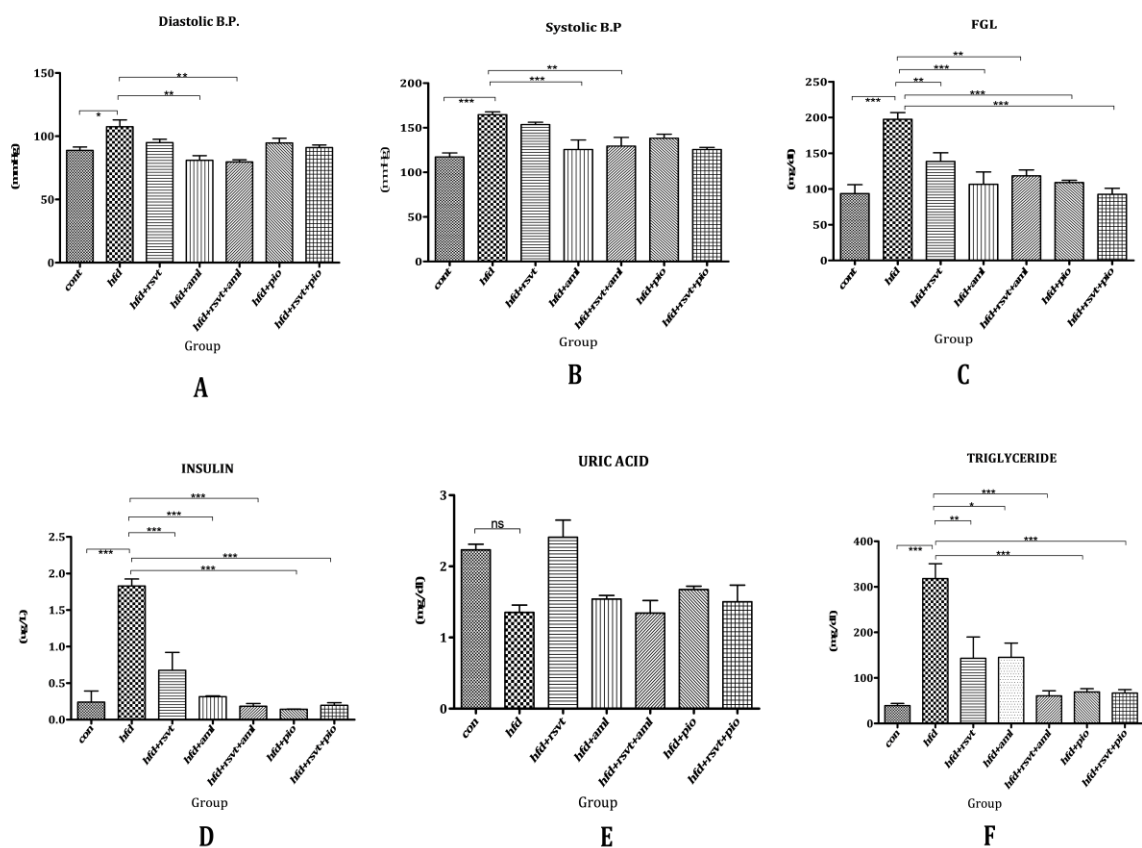


Fig. 1: Estimation of Diastolic and Systolic B.P and biochemical estimations like FGL, insulin, uric acid, Triglycerides.

There was a significant increase ( $p < 0.05$ ) in serum TC level in HFD group as compared to the control group. In this, case all of the treated groups were found to decrease serum TC significantly as compared to HFD group (fig 2).

There was a significant increase ( $p < 0.001$ ) in serum VLDL and LDL levels in HFD group as compared to the control group. RSVT was found to decrease serum VLDL significantly ( $p < 0.01$ ) as compared to HFD group. AML alone & in combination with RSVT were also found to reduce serum VLDL and LDL significantly ( $p < 0.05$  &  $p < 0.001$  respectively) as compared to HFD group. PIO alone & in combination with RSVT were found to reduce serum VLDL and LDL significantly ( $p < 0.001$  in both groups) as compared to HFD group (fig 2).

In the case of serum HDL, no significant increase was found in HFD group as compared to the control group.

There was a significant increase ( $p < 0.001$ ) in serum total lipid level in HFD group as compared to the control group. RSVT was found to decrease serum total lipid significantly ( $p < 0.05$ ) as compared to the HFD group. AML alone & in combination with RSVT were also found to reduce serum total lipid significantly ( $p < 0.001$  in both groups) as compared to HFD group. PIO alone & in combination with RSVT were found to reduce serum total lipid significantly ( $p < 0.001$  in both groups) as compared to HFD group (fig 2).

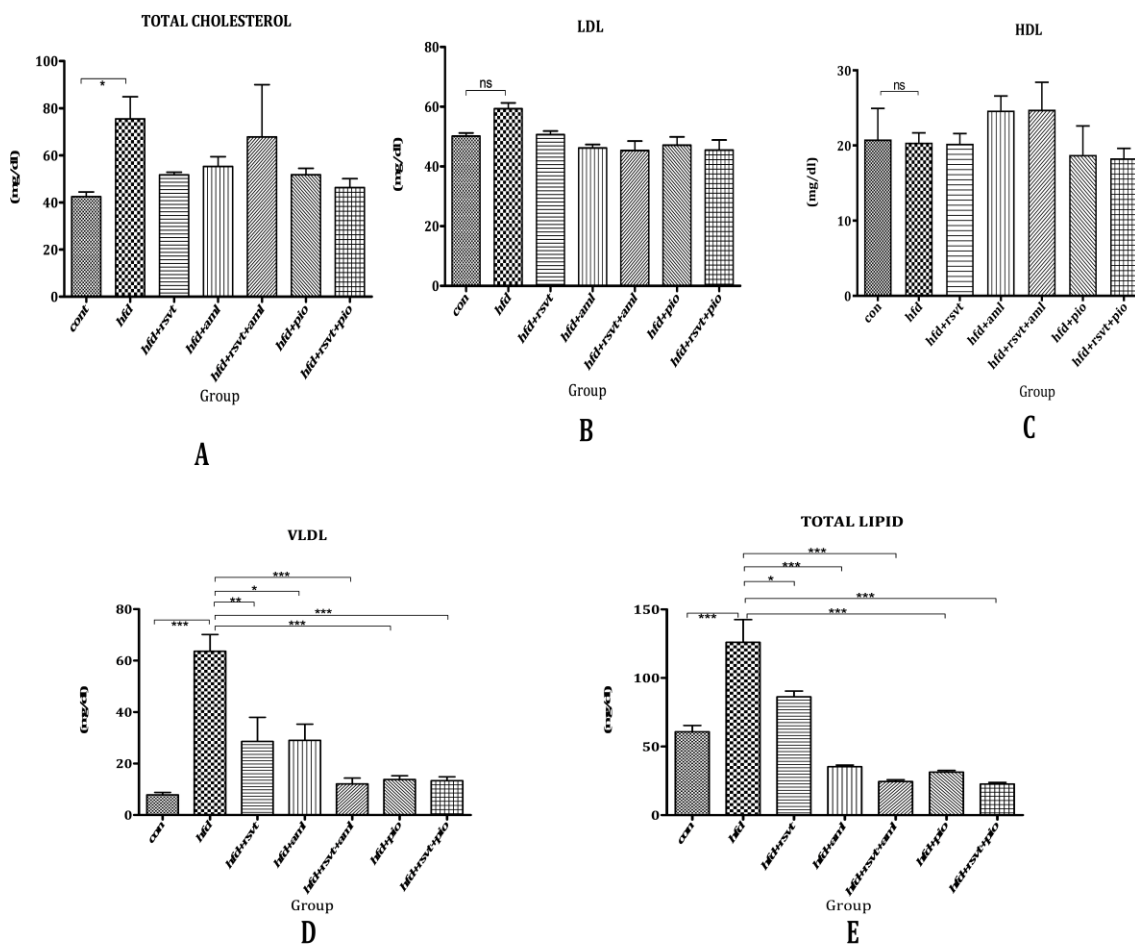


fig. 2: Biochemical estimations including total cholesterol, LDL, HDL, VLDL, total lipid.

#### Antioxidant study:

Liver parameters showed that fructose induces oxidative stress so, SOD, catalase (CAT), and reduced glutathione (GSH) were found to be decreasing ( $p < 0.01$  in all the cases), and lipid peroxidation was found to be increasing significantly ( $p < 0.01$ ) in HFD group. RSVT (20mg/kg) was found to increase SOD ( $p < 0.05$ ), catalase & reduced glutathione significantly as compared to HFD rats, whereas decrease in lipid peroxidation ( $p < 0.05$ ) significantly (fig 3).

Effect of RSVT (20 mg/kg) on change in liver antioxidant enzyme as well as lipid peroxidation in HFD-induced MS in rats

(Values are expressed as mean  $\pm$  SEM, P value \*  $< 0.05$ ; \*\*  $< 0.01$ , \*\*\*  $< 0.001$ )

| Group   | Lipid Peroxidation<br>(nm of MDA /gm of tissue) | Antioxidant Enzymes               |  |                         |
|---------|---|-----------------------------------|--|-------------------------|
|         |   | GSH<br>(mg of GSH / gm of tissue) | Catalase<br>( $\mu$ mol of H <sub>2</sub> O <sub>2</sub> evolved/ mg of protein) | SOD<br>(U/gm of tissue) |
| Control | 1.110 $\pm$ 0.00                                | 54.83 $\pm$ 2.598                 | 24.87 $\pm$ 1.278  | 49.33 $\pm$ 0.6667      |

|                 |                 |                  |                              |                  |
|-----------------|-----------------|------------------|------------------------------|------------------|
| <b>HFD</b>      | 2.040 ± 0.13**  | 27.33 ± 1.139*** | 15.45 ± 1.776**              | 14.67 ± 0.6677** |
| <b>HFG+RSVT</b> | 1.377 ± 0.1333* | 36.03 ± 1.560*   | 19.44 ± 0.5336 <sup>ns</sup> | 34.67 ± 7.265 *  |

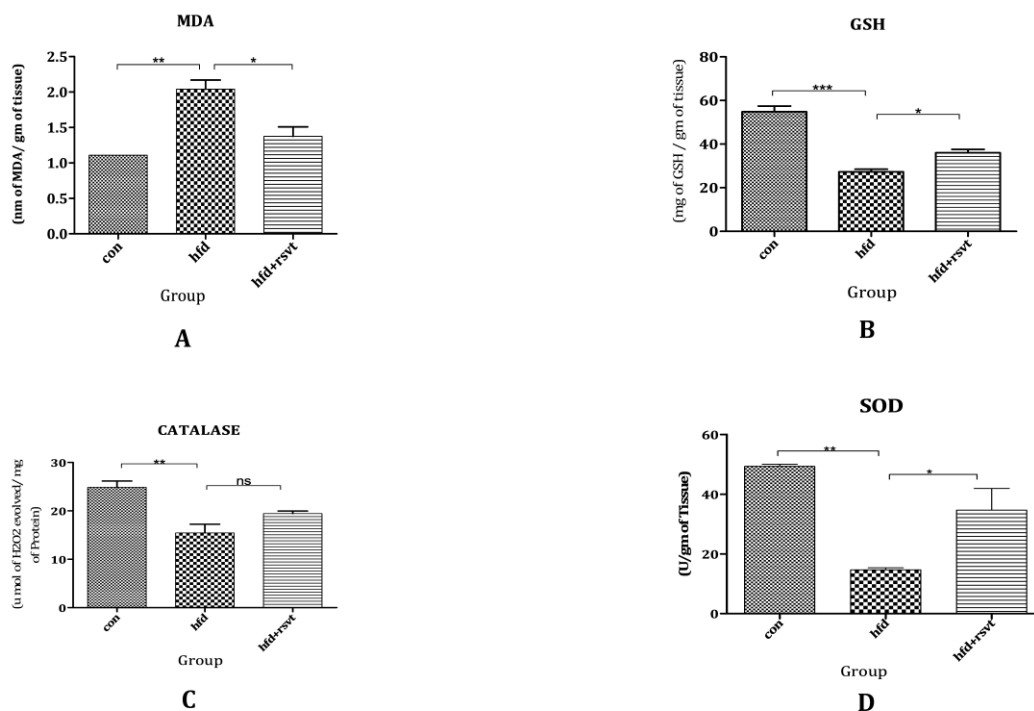
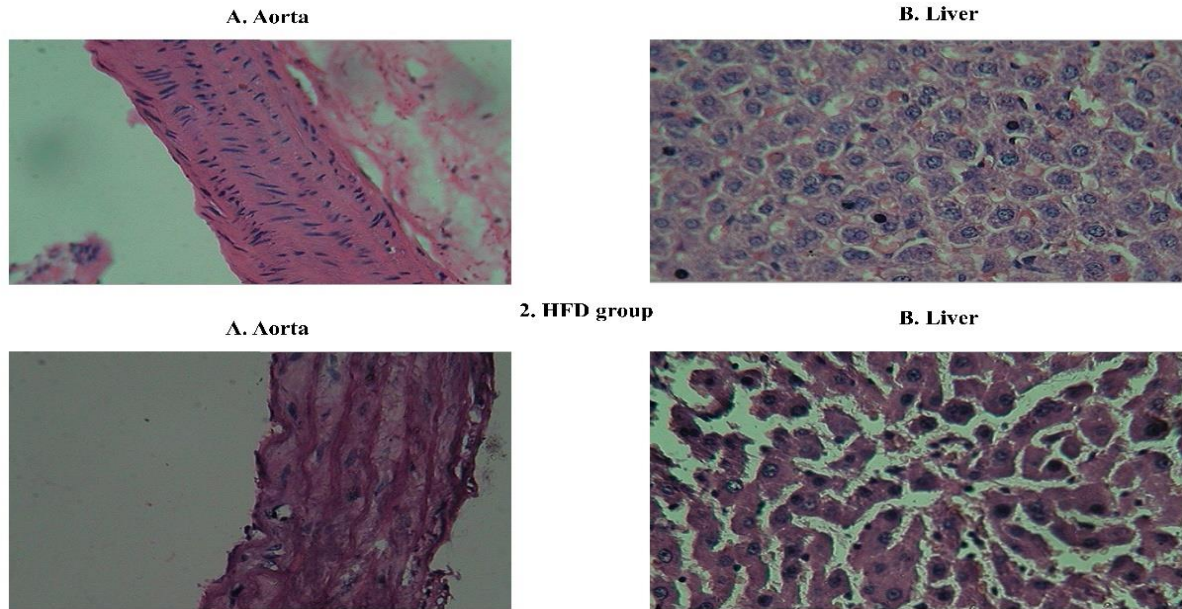


Fig. 3: Bar diagrams indicating antioxidant activity of control group, high fructose diet group and high fructose diet + RSVT group.

Histopathological study:

HFD showed an increase in lipid deposition and endothelial dysfunction as compared to the normal animal aorta. RSVT, AML, and PIO as well as a combination of AML & PIO with RSVT showed decreased lipid deposition as well as endothelial dysfunction after 35 days of treatment (fig 4.- fig 7.).

**1. CONTROL group**



**2. HFD group**

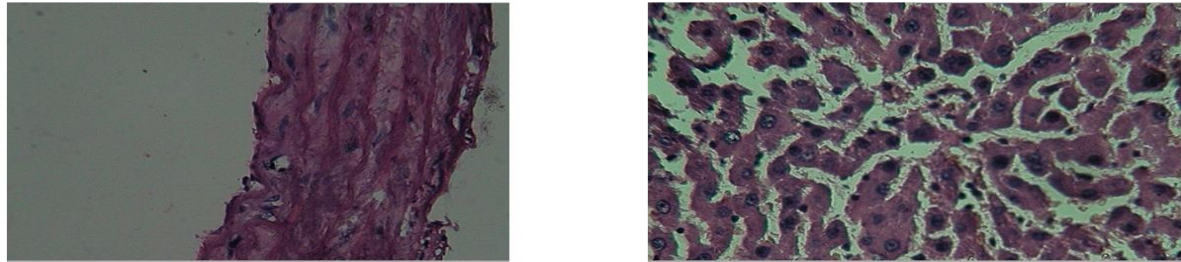
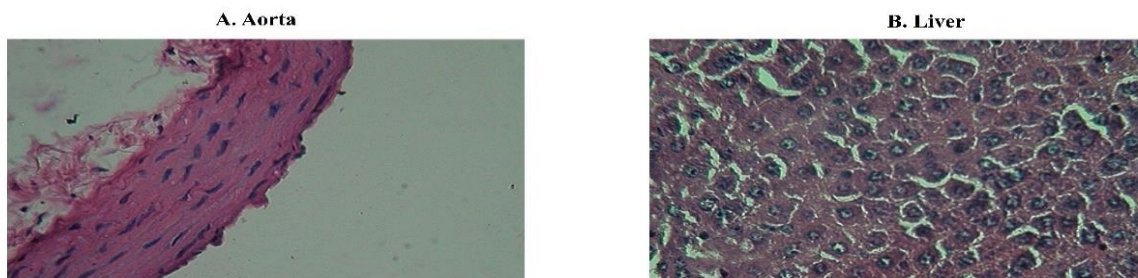


Fig 4: (1. Control group) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with Standard laboratory diet (Control group) and HFD group respectively. (2. HFD group) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD.

**3. HFD+RSVT**



**4. HFD+AML**

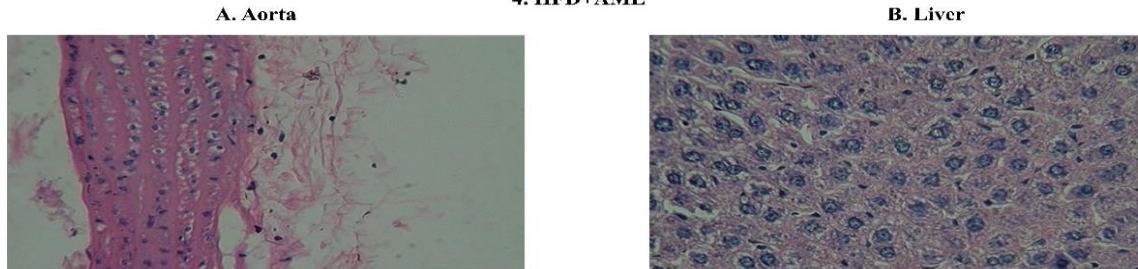


Fig 5: (3. HFD+RSVT) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD + RSVT (20 mg/kg). (4. HFD+AML) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD + AML (10 mg/kg).

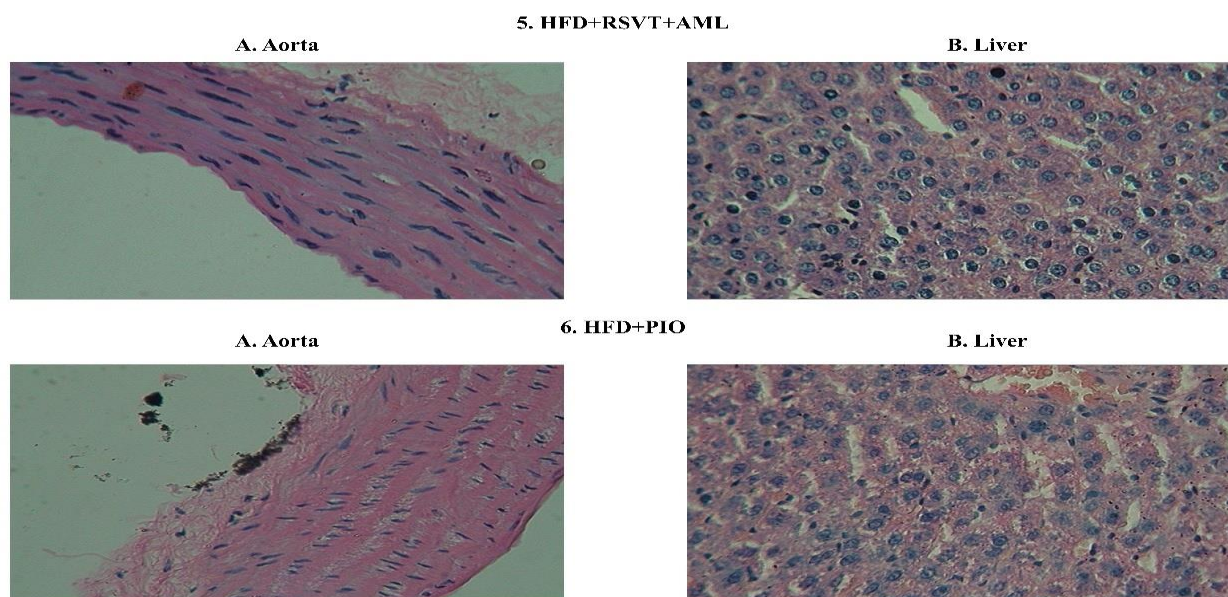


Fig 6. (5. HFD+RSVT+AML) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD + AML (10 mg/kg) + RSVT (20mg/kg). 6. HFD+PIO - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD + PIO (20mg/kg).

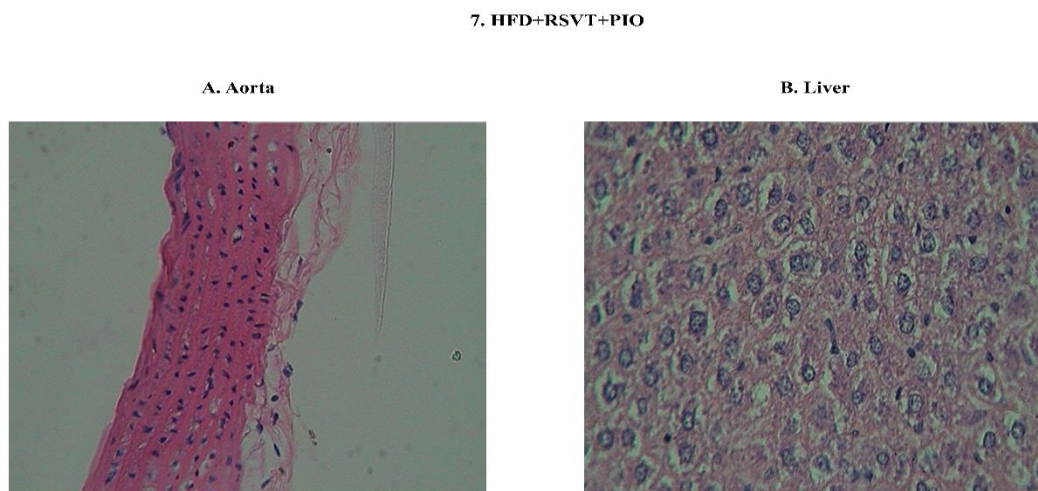


Fig 7. (7. HFD+RSVT+PIO) - Histopathological sections of Aorta and Liver (stained with Haematoxylin & Eosin) of rat fed with HFD + PIO (20 mg/kg) + RSVT (20mg/kg)

Statistical Analysis: All the data were expressed as mean  $\pm$  SEM (standard error of the mean). Statistical significance between more than two groups was tested using one-way ANOVA followed by the Bonferroni multiple comparisons test as appropriate using a computer-based fitting program (Prism, GraphPad). Differences were considered to be statistically significant when  $p < 0.05$  (\* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ , non-significant(ns)  $p > 0.05$ ).

## DISCUSSION:

Metabolic syndrome (MS) has gained attention as a global public health issue. Since most patients find it challenging to adhere to exercise or dietary plans, it has become crucial to look into potential treatments for the harmful effects of metabolic syndrome. A constellation of abnormalities, including abdominal obesity, dyslipidemia, and insulin resistance, make up metabolic syndrome and lead to hypertension in the end. The patient is at a greater risk of acquiring type 2 diabetes mellitus and cardiovascular morbidities as a result of these abnormalities<sup>21</sup>. Over the past few decades, there has been a correlation between high fructose consumption and an increase in MS cases<sup>22</sup>. In studies, fructose has received widespread acceptance as an animal model for metabolic syndrome (MS) that replicates the symptoms affecting human participants<sup>23</sup>.

The current study showed that keeping rats on HFD for 5 weeks caused the classic symptoms of MS, which were manifested by a significant rise in systolic BP, diastolic BP, a significant rise in blood glucose and serum insulin levels, a significant rise in total cholesterol, triglycerides, and LDL-C, and a significant decline in HDL-C compared to control rats.

RSVT decreased the serum glucose level maybe by SIRT1 activation can also encourage GLUT4 (glucose transporter type-4) translocation into cell membranes; resveratrol binding to estrogen receptors can facilitate GLUT4 translocation which leads to a reduction in serum glucose levels<sup>24</sup>. The observed lipid-lowering effect by RSVT may be due to increased plasma lipid uptake by the liver and adipose tissue or by decreased hepatic fatty acid synthesis. Also, RSVT is a strong antioxidant. RSVT acts by changing the oxidative stress of the diabetic tissues and thus improves the functional states of the metabolic machinery of the cells & the improved cellular redox status helps maintain the normal function of the mediators involved in insulin signaling and promotes vasodilation by enhancing nitric oxide production or improved nitric oxide bioavailability by the reduction of hydrogen peroxide RSVT decreased BP.

AML increases the hormone-sensitive triglyceride lipase to cause rapid breakdown of triglycerides and mobilization of free fatty acids, thereby leading to a fall in levels of serum triglycerides. AML acts by relaxing the smooth muscle in the arterial wall decreasing total peripheral resistance and hence reducing B.P.

PIO showed significant change in systolic and diastolic BP. PIO selectively stimulates the nuclear receptor peroxisome proliferator-activated receptor gamma (PPAR-Y). PIO reduces insulin resistance in the liver and peripheral tissues.

## CONCLUSION:

Resveratrol has (20mg/kg) reduced serum TG, insulin, FGL & total lipids significantly. AML & PIO also reduced all the above serum parameters. There was a significant change of action of drugs with a combination of RSVT was found. In case of BP, RSVT decreased significantly. AML & AML+RSVT were found to decrease both systolic as well as diastolic B.P. In the case of the antioxidant enzyme of the liver, RSVT significantly increased liver SOD, catalase, and GSH as compared to HFD animals. It also reduced liver lipid peroxidation significantly. Histopathological study of liver & aorta showed that RSVT, PIO & AML alone & their combination with RSVT prevented damage to tissue from HFD to animals.

RSVT possesses antioxidant activity. Also, it has anti-hyperglycemic anti hyper triglyceridemic & anti hyperinsulinemic effects but we haven't found any drastic increase in the efficacy when given with combinatios.

## REFERENCES

1. Alberti KGMM, Eckel RH, Grundy SM, Zimmet PZ, Cleeman JI, Donato KA, et al. Harmonizing the metabolic syndrome: a joint interim statement of the international diabetes federation task force on epidemiology and prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International. *Circulation*. 2009;120(16):1640–5.
2. International Diabetes Federation. The IDF consensus worldwide definition of Metabolic Syndrome. 2006.
3. Nur Hidayah Reshidan, Suhaila Abd Muid and Norshalizah Mamikutty. The effects of Pandanus amaryllifolius (Roxb.) leaf water extracts on fructose-induced metabolic syndrome rat model. *BMC Complementary and Alternative Medicine*. 2019 19:232.
4. Moreno-Fernandez, S., et al., 2018. High fat/high glucose diet induces metabolic syndrome in an experimental rat model. *Nutrients*, 10 (10), 1502.
5. Ford ES. Risks for all-cause mortality, cardiovascular disease, and diabetes. *Diabetes Care*. 2005;28(7):1769–78.
6. Renuka M, Rajani G, Haritha K, Swathi M, Raju A. Effect of rutin and telmissartam on metabolic syndrome x. *Int J Phytomedicine* 5. 2012; 4:567–72.
7. Kaur, J., 2014. A comprehensive review on metabolic syndrome. *Cardiology research and practice*, 2014, 943162.

8. Toth, P.P., et al., 2019. Elevated triglycerides (150 mg/dL) and high triglycerides (200–499 mg/dL) are significant predictors of hospitalization for new-onset kidney disease: a real-world analysis of high-risk statin-treated patients. *Cardiorenal medicine*, 9 (6), 400–407.
9. Tappy, L., et al., 2010. Fructose and metabolic diseases: new findings, new questions. *Nutrition*, 26 (11–12), 1044–1049.
10. Taskinen, M., Packard, C.J., and Boren, J., 2019. Dietary fructose and the metabolic syndrome. *Nutrients*, 11 (9), 1987.
11. Mona A. Said, Naglaa Y. Nafeh & Hend A. Abdallah (2021): Spexin alleviates hypertension, hyperuricemia, dyslipidemia and insulin resistance in high fructose diet induced metabolic syndrome in rats via enhancing PPAR- $\gamma$  and AMPK and inhibiting IL-6 and TNF- $\alpha$ , *Archives of Physiology and Biochemistry*, DOI: 10.1080/13813455.2021.1899242.
12. "Resveratrol". Micronutrient Information Center, Linus Pauling Institute, Oregon State University, Corvallis, OR. 11 June 2015. Retrieved 26 August 2019.
13. Fremont, Lucie (January 2000). "Biological Effects of Resveratrol". *Life Sciences*. 66 (8): 663–673. doi:10.1016/S0024-3205(99)00410-5. PMID 10680575.
14. Jasiński M, Jasińska L, Ogródowczyk M; Jasińska; Ogródowczyk (August 2013). "Resveratrol in prostate diseases - a short review". *Cent European J Urol*. 66 (2): 144–149. doi:10.5173/cej.2013.02.art8. PMC 3936154. PMID 24579014.
15. "Stilbenes-resveratrol in foods and beverages, version 3.6". Phenol-Explorer. 2016. Retrieved 13 May 2016.
16. "Resveratrol". MedlinePlus, National Library of Medicine, US National Institutes of Health. 1 April 2019. Retrieved 22 September 2019.
17. De Ligt, M; Timmers, S; Schrauwen, P (2015). "Resveratrol and obesity: Can resveratrol relieve metabolic disturbances?". *Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease*. 1852 (6): 1137–1144. doi:10.1016/j.bbdis.2014.11.012. PMID 25446988.
18. Hausenblas HA, Schoulda JA, Smoliga JM (2015). "Resveratrol treatment as an adjunct to pharmacological management in type 2 diabetes mellitus--systematic review and meta-analysis". *Mol Nutr Food Res*. 59 (1): 147–159. doi:10.1002/mnfr.201400173. PMID 25138371.
19. Asgary, Sedigheh; Karimi, Raheleh; Momtaz, Saideh; Naseri, Rozita; Farzaei, Mohammad Hosein (1 June 2019). "Effect of resveratrol on metabolic syndrome components: A systematic review and meta-analysis". *Reviews in Endocrine and Metabolic Disorders*. 20 (2): 173–186. doi:10.1007/s11154-019-09494-z. PMID 31065943. S2CID 146806930
20. Koushki, Mehdi; Dashatan, Nasrin Amiri; Meshkani, Reza (July 2018). "Effect of Resveratrol Supplementation on Inflammatory Markers: A Systematic Review and Meta-analysis of Randomized Controlled Trials". *Clinical Therapeutics*. 40 (7): 1180–1192.e5. doi:10.1016/j.clinthera.2018.05.015. PMID 30017172. S2CID 51677307.
21. Al-Sawalha, N.A., et al., 2019. The effects of l-Carnosine on development of metabolic syndrome in rats. *Life sciences*, 237, 116905.
22. Elliott, S.S., et al., 2002. Fructose, weight gain, and the insulin resistance syndrome. *The American journal of clinical nutrition*, 76 (5), 911–922.
23. Tran, L.T., Yuen, V.G., and McNeill, J.H., 2009. The fructose-fed rat: a review on the mechanisms of fructose-induced insulin resistance and hypertension. *Molecular and cellular biochemistry*, 332 (1–2), 145–159.
24. Resveratrol Counteracts Insulin Resistance—Potential Role of the Circulation - Scientific Figure on ResearchGate. Available from: [https://www.researchgate.net/figure/Multiple-mechanisms-by-which-resveratrol-may-counteract-insulin-resistance-activation-of\\_fig1\\_327217688](https://www.researchgate.net/figure/Multiple-mechanisms-by-which-resveratrol-may-counteract-insulin-resistance-activation-of_fig1_327217688) [accessed 28 Jul, 2022].
25. may-counteract-insulin-resistance-activation-of\_fig1\_327217688 [accessed 28 Jul, 2022].