International Journal of Phytomedicine 5 (2013) 233-242
http://www.arjournals.org/index.php/ijpm/index

Original Research Article

ISSN: 0975-0185

Effect of rutin and telmisartam on metabolic syndrome x

M Renuka[1], G Rajani[2]*, K Haritha[2], M Swathi[3], AB Raju[3],

*Corresponding author:

G Rajani

[1]Research associate in Preclinical toxicology, Sipra Labs, Hyderabad, AP, India
[2]Department of Pharmacy Practice KLR Pharmacy College, Paloncha, AP, India
[3]Department of Pharmacology St. Peters Institute of Pharmaceutical Sciences, Hanamkonda,Warangal, AP, India

Abstract

The objective of the present study was to investigate the effect of rutin and telmisartan on parameters related with insulin sensitivity, glucose and fat metabolism in the high fructose consumed animal model of the ovariectomized female Sprague dawley rats.

Female Sprague Dawley (SD) rats of 180-230 g body weight were grouped and fed with either standard chow diet (NPD) or high fructose diet (HFD30% w/v). Rutin and Telmisartan were administered orally in a dose of 100 mg/kg & 5 mg/kg *p.o.* Daily for the entire study period. Normal parameters (Food intake & Body weight), Biochemical parameters (Blood glucose by GOD/POD method, Total Cholesterol by CHOD/PAP method, Triglyceride level, LDL, HDL) Metabolic Indicators (Aspartate transminase (AST), Alanine transminase (ALT)), Swimming test, Oral Glucose Tolerance Test, & Body weight were measured in all the groups. The unpaired t- test and analysis of variance (ANOVA) was used for statistical analysis.Telmisartan and Rutin significantly improved abnormal metabolic profile and glucose intolerance. Rutin reduces the body weight, plasma triglycerides, and total cholesterol .in addition rutin reduces the elevated serum glutamate pyruvate transminase levels.Improved abnormal metabolic profile and glucose intolerance indicates that rutin and telmisartan provides a scientific rationale for the use in metabolic syndrome.

Keywords: Metabolic syndrome; Peroxisome proliferator activator receptor (PPAR-γ); Diabetic mellitus; Telmisartan; Rutin

Introduction

The incidence of metabolic diseases in the modern society has reached epidemic proportions and constitutes a great public health concern. Metabolic syndrome is a growing problem. Like many syndromes in the history of medicine metabolic syndrome is a group of disorders that share a common pathway. It is commonly defined as a group of risk factors or abnormalities closely associated with insulin resistance that markedly increases risk for diabetes and it is an important prognostic factor for cardiovascular diseases. It is widely agreed that it is a growing and pressing problem for the society. Obesity corresponds to excessive fat accumulation originating from an imbalance in consumed versus expended calories. The World Health Organization (WHO) has estimated that the global incidence of type II diabetes will exceed 300 million by the year 2030, in another side India has become as the diabetic capital of the world. The "metabolic syndrome", formerly "metabolic syndrome X" [1] is composed of cluster of visceral obesity, insulin resistance, dyslipidemia, hypertension, is associated with pro-thrombotic, pro-atherogenic, and inflammatory risk factors that predispose to heart attack, stroke, heart failure and sudden cardiac death. Estrogen insufficiency in postmenopausal women is a status of permanent cessation of menstruation resulting from loss of ovarian follicular activity. Reduced estrogen levels in postmenopausal women are at risk for increased incidence of metabolic and cardiovascular abnormalities including obesity, type II diabetes mellitus, and cardiovascular diseases [2]. To mimic these metabolic complications like weight gain, dyslipidemia, moderate hyperglycemia, oxidative stress and insulin resistance, we are feeding the rats with a high calorie food in the form of high fructose diet (HFD). Besides, ovariectomy (surgically removal of ovaries) in rats leads to decline in estrogen levels which play an important role in the regulation of many processes related to the control of energy homeostasis. These include food intake and energy expenditure, insulin sensitivity in the liver and muscle, adipocyte growth and its body distribution as well as the pancreatic β cell functions. So, the ovariectomized (OVX) female rats which are fed with HFD for 6 weeks had a greater increase in energy intake, body weight, plasma glucose, serum levels of cholesterol and triglycerides.

Fasting hyperglycemia or impaired fasting glucose, impaired glucose tolerance, or insulin resistance, High blood pressure, Central obesity (also known as visceral, or apple-shaped adiposity), Decreased HDL cholesterol [< 35 mg/dLl], Elevated triglycerides [> 250]. Associated diseases and signs are: hyperuricemia, progressing to non-alcoholic fatty liver disease, polycystic ovarian syndrome (in women). Various strategies have been proposed to prevent the development of metabolic syndrome. These include increased physical activity and a healthy, reduced calorie diet.The currently existing drugs for the treatment of metabolic complications associate with insulin resistance for obese patients are hampered by adverse effects related to increased adipogenesis, weight gain, and fluid over load. Recent evidence indicates that the angiotensin receptor blocker (ARB) telmisartan

structurally resembles the insulin sensitizer pioglitazone, a thiazolidinedione Ligand of peroxisome proliferator activator receptor–γ (PPAR-γ) used for the treatment of type 2 diabetes mellitus. Peroxisome proliferator activator receptor–γ is an established therapeutic target in the treatment of insulin resistance, diabetes, and metabolic syndrome. Activation of PPAR-γ leads to the expression of key target genes that mediate beneficial effects on glucose and lipid metabolism. But there are no systemic studies carried out to evaluate the beneficial role of telmisartan on high fructose diet induced metabolic syndrome in ovariectomized female Sprague Dawley (SD) rats.

Natural compounds have been proposed as potential therapeutic agents in the prevention and/or treatment of number of diseases. Being aware of the increasing prevalence of obesity/IR, it is imperative to test and identify the possible therapeutic role of bioactive compounds. Rutin (quercetin-3-rhamnosyl glucoside), a natural flavone derivative, was first discovered in buckwheat in the 19th century. It is low molecular weight polyphenolic compound that is widely distributed in vegetables and fruits. Rutin has various biological activities [3,4] that are beneficial to human health such as antioxidant effect, protective effect against hepatotoxicity, and anti-inflammatory effects. In the present study we investigate the effect of both the drugs rutin and telmisartan on high fructose diet induced metabolic complications in ovariectomized female Sprague dawley rats.

The nonpeptide ARB telmisartan was approved by the US Food and Drug Administration (FDA) in 1998 for the treatment of hypertension alone or in combination with other compounds. The ARB telmisartan interferes with the renin angiotensin system by selectively blocking the binding of angiotensin II to its receptor subtype 1. The angiotensin II receptor blocker (ARB) telmisartan has a molecular structure that confers it partial agonist properties similar to those of peroxisome proliferators-activated receptor gamma molecule, which is thought to modulate tissue response to insulin ARBs has been known to reduce the incidence of new-onset DM [5]- via the improvement of insulin resistance and prevent cardiovascular events through the development of atherosclerosis by normalizing endothelial dysfunction [6] A recent clinical trial showed that telmisartan reduces visceral fat accumulation in patients with metabolic syndrome . In animal models, several reports have shown that telmisartan protects against weight gain [7] Peroxisome proliferator–activated receptor γ is a transcription factor that controls the gene expression of several key enzymes of glucose and lipid metabolism and thereby increases insulin sensitivity. In vitro, telmisartan-induced PPAR- γ activation was AT1 receptor independent and occurred at therapeutically relevant concentrations of 1 to 5 A mol/L, whereas the potency of other ARBs to activate PPAR- γ receptors tested in this setting was considerably lower. In addition, telmisartan was just recently shown to induce adiponectin protein content in cultured adipocytes by a posttranscriptional mechanism involving a reduction of 26S proteasome activity [8].

On the other hand, because Telmisartan works as a selective PPAR γ modulator, its effects on gene expression partially differ from those of full agonists, such as thiazolidinediones, which strongly promote adipogenesis and cause weight gain. Microassay and quantitative PCR analysis showed that stimulation of Telmisartan on the expression of genes involved in adipocytic lipid storage, for example, prostacyclin receptor and glycerol kinase, was much weaker than that of pioglitazone [9]. This finding suggests that Telmisartan-induced PPAR activation is not associated with weight gain. Indeed, they observed that Telmisartan treatment decreased the weight of visceral adipose tissue without affecting food intake in diet-induced obese mice and increased the mRNA expression of uncoupling protein 1 in brown adipose tissue, accompanied by an increase in oxygen consumption [10]. Hyperglycemia, hyperinsulinemia, and hypertriglyceridemia in diet-induced obese mice all improved with Telmisartan treatment. Furthermore, Telmisartan treatment increased adiponectin mRNA in visceral adipose tissue and reduced the serum level of resistin, which impairs insulin sensitivity. Our results proposed that Telmisartan may prevent the development of obesity and related metabolic disorders by altering the levels of various adipocytokines and increasing energy expenditure. Recent reports indicate that the visceral fat area (VSA), estimated by abdominal computed tomography scan, and decreased in patients with metabolic syndrome after Telmisartan treatment.

Molecular modeling studies suggest that telmisartan might influence PPAR- γ activity by interacting with regions of the ligand-binding domain that are not typically engaged by full agonists of the receptor [10]. This partial agonist antagonized the activity of full agonist in cell culture studies and the same antagonizing character is yet to be studied in vivo experiments. PPAR-γ receptors are present in the nucleus, so to bind to these receptors the compound must reach nucleus and liphophilicity is playing a crucial role, telmisartan found to be most hydrophobic among the ARBs. Two parallel trials of telmisartan known as Ongoing telmisartan Alone and in Combination with Ramipril Global Endpoint Trial (ONTARGET) and telmisartan Randomized Assessment Study in ACE-Intolerant Subjects with Cardiovascular Disease (TRANSCEND)are in progress to evaluate cardiovascular and metabolic end points in 28,400 patients [11]. A subset of 5000 patients who are intolerant to ACE inhibitors will be Studied in the TRANSCEND trial [12].

Materials and Methods

Animals

female Sprague Dawley (SD) rats of 180-230 g body weight were procured from the Mahaveer Enterprises, Hyderabad, India. The animals were maintained under controlled room temperature (22±2 °C) and humidity (55 ± 5%) with a 12-h light / dark cycle. Animals were housed in three animals per cage and allowed to food and water *ad libitum*. Rats were grouped and fed with either standard chow diet (NPD) or high fructose diet (HFD30% w/v) The protocol of this experiment was approved by The committee for the purpose of control and supervision of experiments on animals (CPCSEA)

vide approval no 1516/PO/a/11/CPCSEA KLR Pharmacy College, and the experiments were carried out in accordance with IAEC guidelines on animal experimentation.

Drugs and chemicals

Rutin and telmisartan is procured from Sigma-Aldrich Company. Glucometer was manufactured by aspen diagnostics PVT Ltd and procured from S.S Pharma, Hanamakonda. Total cholesterol and HDL kit, trigycerides kit, were procured from SS pharma. All the solvents and other chemicals were procured from SS pharma, Hanamkonda and they were of analytical grade quality.

Experimental procedure

Female Sprague dawley (SD) rats were divided into the following groups. Each group consists of 6 animals.
Group 01: CONTROL (Fed with normal diet chow)
Group 02: SHAM CONTROL (Female SD Rats were fed with High Fat Diet or HFD)
Group 03: HFD control
Group 04: HFD+RUTIN treated animals
Group 05: HFD+TELMISARTAN treated animals
Female SD rats were Bilateral ovariectomized(OVX). Surgical procedure was performed under anesthesia. Animals were allowed to recover for 7 days after surgery.
Group 06: OVX control
Group 07: OVX+RUTIN treated animals
Group 08: OVX+TEL treated animals
Female SD rats were Bilateral ovariectomized and fed with HFD
Group 09: OVX+HFD
Group10: OVX+HFD+RUTIN
Group11: OVX+HFD+TELMISARTAN

Preparation of high fructose diet

30 % w/v solution was prepared by dissolving 300g of fructose in 1 liter water.

Treatment Protocol

Rutin was administered orally in a dose of 100 mg/kg, *p.o.* Daily for the entire study period.
Telmisartan was administered orally in a dose of 5 mg/kg, *p.o.* Daily for the entire study period.
Following Parameters were studied: Normal parameters: Food intake & Body weight, Biochemical parameters: Blood glucose, Total Cholesterol & Triglyceride level, LDL, HDL, Metabolic Indictors: Aspartate transminase (AST) & Alanine transminase (ALT)

Swimming test

The exercise treatment consisted of swimming the rats in 33-gallon plasticbarrels (diameter, 60 mm). Depth of the water was approximately 76 mm, and the temperature ranged between31±33_C (thermo-neutral temperature as described by Mc Cardle [6]. The swimming protocol consisted of a 4-week adaptation period and 4-week training period. It began 2 days after delivery as follows: 5 days per week (Monday to Friday) in the morning, at the beginning of the light period. Animals swam for 5 min the first day, and their swim time was increased by 5 min per day until40 min was attained for a 2 week period. Duration was increased to 60 min for 1 week, then 75 min for the remaining 4 weeks to ensure an aerobic training effect. Rats learning quickly to float without much effort and to intermittently sink to the bottom to rest then bob to the top. When barrel was crowded, rats tended to remain at the top and compete for space. Thus, bobbing was decreased and hind and fore-leg activity was optimized by this protocol. Furthermore, the rats were constantly supervised and made to keep moving in the water by gently prodding them if needed. Rats swam freely with no additional weights (unloaded). The exercise oxygen consumption of non- weighted swimming rats has previously been determined to be 2-7 times the resting value [6], which is considered to be moderate intensity exercise for rats. After the daily swimming session, the rats were towel-dried and returned to their cages. They were watched carefully after being placed in their cages to ensure that they did not shiver indicating hypothermia stress.

Results

Effect of Rutin and Telmisartan on body weight of rats

The body weight (gm.) was recorded from 0 to 6 week in all the groups and we found that there is no increase in body weight in rutin and telmisartan treated groups when compared with untreated groups. (Table 1)

Effect of Rutin and Telmisartan on Food Intake

Total food intake over 6 week study period in all the groups were listed in table-2. We observed that rutin and telmisartan has not shown marked effect on food intake.

Effect of Rutin and Telmisartan Oral glucose tolerance test

Oral glucose tolerance test in all groups were measured as shown in table 3 and it was found that glucose levels were decreased with respect to time in groups containing OVX and HFD rats when treated with rutin and telmisartan.

Effect of Rutin and Telmisartan Plasma glucose level

Plasma glucose levels in all the groups were measured and it was found that rutin and telmisartan decreases plasma glucose levels in all the treatment groups. Telmisartan has more pronounced effect on plasma glucose levels when compared to rutin as mentioned in table-4.

Effect of Rutin and Telmisartan Plasma triglyceride level

Plasma triglyceride levels were measured in all the groups as listed in table-5 and it was found that rutin and telmisartan reduces plasma triglyceride levels in group-9 to normal level.

Effect of Rutin and Telmisartan Plasma total cholesterol levels

Plasma total cholesterol levels were measured and listed in table-6 and it was found that rutin and telmisartan has beneficial effect in reducing total cholesterol levels in all the treated groups.

Effect of Rutin and Telmisartan Plasma High Density Lipoprotein

Plasma High Density Lipoprotein levels were measured in all the groups and listed in table-7. Based on the results we found that rutin and telmisartan has no significant effect on high density lipoprotein level in treatment groups.

Effect of Rutin and Telmisartan Plasma low Density Lipoprotein

Plasma low Density Lipoprotein levels were measured in all the groups as listed in table-8 and from the results we found that rutin and telmisartan has more beneficial effect in reducing plasma low density lipoprotein levels in treated groups.

Effect of Rutin and Telmisartan Plasma very low Density Lipoprotein

Plasma very low Density Lipoprotein levels were measured in all the groups and it was found that rutin and telmisartan reduces plasma low density lipoprotein levels to normal level in treated groups. The values were represented in table-9.

Swimming Test

Table 10 represents rat weights (g) after swimming test over the 6-week study period in NPD, SHAM, OVX, OVX+ex, OVX+RTN+ex , OVX+RTN, OVX+TEL, OVX+TEL+ex groups. From the results it was found that rutin and telmisartan treatment along with swimming reduces body weight in treated groups as compared to untreated groups.

Effect of Rutin and Telmisartan Serum glutamate pyruvate transaminase

Serum glutamate pyruvate transminase levels in all the groups were measured and listed in table-11 and it was found that rutin and telmisartan has a protective effect in reducing serum glutamate pyruvate transaminas levels in treated groups to normal level.

Effect of Rutin and Telmisartan Serum glutamate oxalo acetic transminase

Serum glutamate oxalo acetic transminase levels were measured in all the groups and listed in table-12 and from the results we found that telmisartan has more beneficial effect in reducing serum glutamate oxalo acetic transaminase levels when compared to rutin in treated groups

Table 1: Body weight

GROUP	Body Weight (g)						
	Owk	lwk	2wk	3wk	4wk	5wk	6wk
NPD	200±2.84	210±3.25	218±2.42	225±3.26	234±2.84	240±2.54	250±3.27
SHAM	205±2.50	207±3.48	216±2.81	220±3.43	232±2.83	238±2.54	245±3.24
OVX	198±2.58	218±3.27	230±4.26	245±5.21	258±3.42	261±2.84	270±6.54
HFD	200±2.80	220±5.60	238±2.40.	244±2.53	256±2.54	262±6.12	272±2.52
OVX+HFD	200±3.21	225±4.62	242±6.85	260±3.24	275±4.22	285±7.62	300±3.32
OVX+RTN	200±2.80	212±6.21	218±2.63	232±3.40	240±4.68	250±3.44	255±6.32
HFD+RTN	200±3.50	215±2.50	225±4.50	235±5.20	245±6.23	252±3.21	260±3.85
OVX+HFD+RTN	200±3.12	210±4.12	215±3.82	230±4.12	235±2.54	240±3.14	245±2.81
OVX+TEL	20012.85	210±3.15	215±4.16	230±4.54	240±6.53	235±2.54	245±6.12
HFD+TEL	200±2.14	212±2.45	217±3.12	232±2.45	242±3.12	240±2.54	237±2.12
OVX+HFD+TEL	200±2.17	215±3.12	218±4.12	220±2.54	230±3.12	245±3.14	245±2.18

Table 1 : Body weight changes from basal to 6 week study period in NPD, SHAM, OVX, HFD, OVX+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL AND OVX+HFD+TEL groups, Each data point is represented as MEAN + SEM (n = 6). a* vs NPD, b*vs. OVX, c*vs. HFD, d*vs OVX+HFD Date was analyzed by One way ANOVA followed by Dunett's multiple comparison test. *P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 2: Food intake

Group	Food intake (g)
NPD	2900
SHAM	2700
OVX	3400
HFD	2900
OVX+HFD	3000
OVX+RTN	3300
HFD+RTN	2900
OVX+HFD+RTN	3000
OVX+TEL	3200
HFD+TEL	2900
OVX+HFD+TEL	2900

Total food intake over week study period in NPD, SHAM, OVX, HFD,OVX + RTN, HFD + RTN, OVX + HFD + RTN, OVX + TEL, HFD + TEL and OVX + HFD + TEL groups.

Table 4: Plasma glucose levels:

GROUP	MEAN ± SEM (mg/dl)
	Plasma glucose levels
NPD	99.8± 1.30
SHAM	99.8 ± 1.30 c***
OVX	134 ± 1.54 a***
HFD	140 ± 1.83 a***
OVX+HFD	157 ±2.77 a***
OVX+RTN	115 ± 5.49 b**
HFD+RTN	126 ± 2.29 c**
OVX+HFD+RTN	133 ± 1.73 d*
OVX+TEL	116 ± 3.68 b*
HFD+TEL	124 ± 3.87 c**
OVX+HFD+TEL	117± 4..51d***

Plasma glucose levels in NPD, SHAM, OVX, HFD, OVX + HFD, OVX + RTN, HFD + RTN, OVX + HFD + RTN, OVX + TEL, HFD + TEL, OVX + HFD + TEL groups. Each data point is represented as MEAN ± SEM (n=6). ***p < 0.001 a Vs NPD. b Vs OVX, cVs HFD d Vs OVX+HFD Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests *P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 3: Oral glucose tolerance test

GROUP	Mean ± SEM glucose levels (mg/dl)				
	0	15	30	60	120
NPD	100±0.57	208±1.24	182 ±4.17	162±1.01	106±0.99
SHAM	98±0.67	199±2.48	176±4.9	158±1.33	99±0.74
OVX	109±1.16	249±1.47	189±1.77	118±1.12	101±2.28
HFD	109 ±1.16	250±1.47	185±3.54	117±1.26	100±2.62
OVX+HFD	110±3.06	305±1.84	248±2.42	185±3.54	120±1.63
OVX+RTN	105±1.15	223±1.58	170±2.20	137±1.93	104±0.65
HFD+RTN	106±1.24	226±2.29	171±1.82	138±1.74	105±0.79
OVX+HFD+RTN	106±2.49	231±2.70	234±3.37	184±2.75	112±2.74
OVX+TEL	103 ±1.41	231±3.70	171±4.90	131±2.08	105±1.09
HFD+TEL	102 ±0.99	234±3.93	179±1.63	138±2.39	98.5±1.65
OVX+HFD+TEL	108 ±2.16	259±4.4	235±3.7	183±2.93	107±3.33

Oral glucose tolerance test in NPD, SHAM, OVX, HFD, OVX+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL and OVX+HFD+TEL groups. Each data point is represented as MEAN ± SEM (n = 6).

Table 5: Plasma triglyceride levels:

GROUP	MEAN ± SEM (mg/dl)
	Plasma triglyceride levels
NPD	88.2-± 1.58
SHAM	88.5 ± 1.54 e***
OVX	123 ± 1.09 a***
HFD	146 ± 1.62 a***
OVX+HFD	171 ±2.96 a***
OVX+RTN	96.5 ±0.56 b***
HFD+RTN	104 ± 0.56 c***
OVX+HFD+RTN	107 ± 0.66 c***
OVX+TEL	89.2 ± 1.58 d***
HFD+TEL	102 ± 0.65 c***
OVX+HFD+TEL	102 ± 1.51d***

plasma triglyceride levels in NPD, SHAM, OVX, HFD, OVX+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL and OVX+HFD+TEL groups. Each data point is represented as MEAN ± SEM (n=6). ***p < 0.001 a Vs NPD. b Vs OVX, c Vs HFD d Vs OVX+HFD Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 6: Total cholesterol levels

Table 6 : Plasma total cholesterol levels in NPD, SHAM, OVX, HFD, OVX+HFD.

GROUP	MEAN ± SEM (mg/dl)
	Plasma Total Cholestrol Levels
NPD	90.8 ± 2.06
SHAM	91 ± 2.2 c***
OVX	120 ± 2.76 a***
HFD	149 ± 2.37 a***
OVX+HFD	189 ± 2.43 a***
OVX+RTN	104 ± 0.85 b***
HFD+RTN	97.8 ±0.83 c***
OVX+HFD+RTN	111 ± 1.32 d**
OVX+TEL	94.5 ±0.99 b***
HFD+TEL	105 ± 1.15 c***
OVX+HFD+TEL	102 ± 1.53 d***

Effect of rutin and telmisartan treatment on the plasma cholesterol levels in OVX+RTN, HFD+RTN,OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, OVX+HFD+TEL, data shown are MEAN ± SEM. Mean values are significantly different for the following comparisons:a *** vs NPD (p<0.001), b*** vs OVX (P<0.001) , c***vs HFD (P<0.001), d*** vs OVX+HFD (P<0.001), d**(p<0.01). Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 7: High density lipoprotein levels

GROUP	MEAN ± SEM (mg/**dl**)
	Plasma High Density Lipoprotein levels.
NPD	30.7 ±1.99
SHAM	30.8 ± 2.40 ns
OVX	32.3 ± 1.43 ans
HFD	32.0 ±1.24 ans^{s}
OVX+HFD	42.2 ± 1.64 ans
OVX+RTN	41.8 ±1.92 b***
HFD+RTN	41.8 ±1.98 cns
OVX+HFD+RTN	39.0 ±0.73 d**
OVX+TEL	32.5 ± 1.57 bns
HFD+TEL	34.3 ± 2.69 cns
OVX+HFD+TEL	38.8 ± 0.98 dms

Plasma High Density Lipoprotein levels in NPD, SHAM, OVX, HFD, OVX+HFD. Effect of rutin and telmisartan treatment on the HDL levels in OVX+RTN, HFD+RTN, OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, OVX+HFD+TEL, data shown are MEAN ±SEM. Mean values are significantly different for the following comparisons: a vs NPD (p<0.001), b** vs OVX (P<0.01), c vs HFD, d *vs OVX+HFD. Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Discussion

The role of estrogen in the regulation of energy homeostasis in females is well established. They seems to have an influence on both carbohydrate and lipid metabolism on the other hand it is also well known that physical activity has beneficial effects on the metabolic syndrome. it is generally accepted that diets high in fat contribute to obesity in both humans and animal models.[13] High dietary fat intake is widely accepted to be associated with a higher risk for obesity and chronic diseases such as cardiovascular diseases, some types of cancer, diabetes, hyperlipidaemia, and

Table 8 : Low density lipoprotein levels

GROUP	MEAN ± SEM (mg/dl)
	Plasma Low Density Lipoprotein levels.
NPD	43.1 ±3.01
SHAM	39.6 ± 2.88 c**
OVX	51.1 ± 2.60 ans
HFD	48.8 ± 0.60 ans
OVX+HFD	64.6 ± 5.16 a***
OVX+RTN	39.1 ±2.92 b*
HFD+RTN	35.5 ±2.37 c*
OVX+HFD+RTN	35.5 ± 2.37 d***
OVX+TEL	39.3 ± 3.18 b**
HFD+TEL	36.6 ± 3.04 c**
OVX+HFD+TEL	39.3 ± 3.26 d***

TABLE 8: Plasma low Density Lipoprotein levels in NPD, SHAM, OVX, HFD, OVX+HFD. Effect of rutin and telmisartan treatment on the LDL levels in OVX+RTN, HFD+RTN, OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, OVX+HFD+TEL, data shown are means ±sem. Mean values are significantly different for the following comparisons: *** vs NPD (p<0.001), b vs OVX , c vs HFD , d*** OVX+HFD (P<0.001),. Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table 9: Very low density lipoprotein levels

GROUP	MEAN ± SEM (mg/dl)
	Plasma very Low Density Lipoprotein levels.
NPD	19.3 ±1.63
SHAM	18.5 ±1.67
OVX	21.8 ± 1.60ans
HFD	20.0 ± 1.26ans
OVX+HFD	25.3 ± 1.38a*
OVX+RTN	19.5 ± 0.91 bns
HFD+RTN	17.0 ±1.41 c*
OVX+HFD+RTN	19.5 ± 1.71d**
OVX+TEL	17.8 ± l..ll bns
HFD+TEL	17.0 ± 0.96 c*
OVX+HFD+TEL	19.3 ± 1.84 d**

Table 9: Plasma very low Density Lipoprotein levels in NPD, SHAM, OVX, HFD, OVX+HFD. Effect of rutin and telmisartan treatment on the VLDL levels in OVX+RTN, HFD+RTN, OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, OVX+HFD+TEL, data shown are MEAN ± SEM. Mean values are significantly different for the following comparisons a vs. NPD (p<0.001), b vs OVX, c vs HFD, d vs. OVX+HFD (P<0.001). Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests. *P<0.05, **P<0.01, ***P<0.001, ns=non significant

hypertension [14]. For example high fructose consumption is related to the prevalence of metabolic syndrome.

In the present study we investigated the consumption of high fructose solution induced metabolic syndrome in ovariectomized female Sprague dawley rats. As expected Ovariectomized female Sprague dawley rats which are fed with high fructose solution aggravated the metabolic complications like increased plasma triglycerides, plasma cholesterol and plasma glucose levels. Our study demonstrated the beneficial effects of Telmisartan and rutin on abnormal metabolic characters. Initially we have validated the prediabetic insulin resistance rat model by feeding high fructose diet and performing an ovariectomy to the female Sprague Dawley rats. Development of metabolic syndrome and glucose intolerance was assessed by performing the plasma biochemical analysis such as plasma glucose, triglyceride, and total cholesterol levels [15]. Glucose intolerance was assessed by performing an oral glucose tolerance test, in accordance with our previous reports HFD fed rats exhibited the cluster of metabolic syndrome characters such as obesity, hyperglycemia, hypertriglyceridimia, hypercholesterolemia and insulin resistance together with reduced glucose disappearance rate, a condition similar to prediabetic, insulin-resistant state in humans compensatory hyperinsulinemia.

Our results showed changes in body weight of the groups during the experiments. Feeding a high fructose solution caused a marked increase in body weight as compared to feeding a normal diet. Likewise, it is well recognized that Ovariectomy results in a substantial decrease in circulating 17 beta estradiol level and that OVX animals become hyperphagic and gain weight (Latour MG 2001) ovariectomization also significantly increases body weight compared to those of sham opearated animals. Ovariectomized rats which are fed with high fructose solution for 6 weeks of the present study exhibited much increased/elevated body weights when compared to those of high fructose fed rats or simply ovariectomized animals alone. However, we observe that orally administered rutin and telmisartan significantly reduces the body weight gain induced by the both ovariectomization and high fructose supplementation. [Graph 1]

Table 10 Swimming test

GROUP	Swimming Test Body Weight (g)						
	Owk	1wk	2wk	3wk	4wk	5wk	6wk
NPD	200±3.57	210±6.24	218±5.12	225±3.01	234±4.28	240±3.83	250±4.5
SHAM	205±2.81	207±6.12	216±5.41	220±3.44	232± 2.83	238±3.24	245±2.24
OVX	198±1.82	218±5.84	230±5.42	245±3.12	258±2..32	267±1.84	270±2..32
OVX + ex	200±2.82	212±4.54	218±3.54	230±4.26	240±2.43	250±4.51	255±2.8
OVX +RTN +ex	200±3.06	215±4.74	218±3.42	232±3.54	245±2.22	255±2.23	240±2.23
OVX + RTN	200±2.82	214±4.52	218±5.1	230±4.62	240±5.12	250±5.12	255±2.1
OVX + TEL	200±2.1	212±5.4	215±3.8	230±2.62	235±3.82	240±2.12	237±1.8
OVX +TEL + ex	200±1.85	212±2.5	216±3.2	227±4.2	236±5.23	238±3.82	245±4.3

Rat weights (g) over the 6-week study period in NPD, SHAM, OVX, OVX+ex, OVX+RTN+ex ,OVX+RTN, OVX+TEL, OVX+TEL+ex groups. Each data point is the mean weight of each group.OVX + noex was significantly different from all other groups

Table 11: Plasma alanine transminase levels

GROUP	MEAN ± SEM (mg/dl)
	Serum glutamate pyruvate transminaseactivity (IU/L).
NPD	31.0 ±2.13
SHAM	29.7 ±2.29
OVX	38.0 ± 2.83 ans
HFD	35.0 ± 2.77 ans
OVX+HFD	42.0± 4.04 a**
OVX+RTN	33.5±1.89 bns
HFD+RTN	33.7 ±2.56 c
OVX+HFD+RTN	39.8 ± 2.82 d*
OVX+TEL	33.0 ± 2.45 bns
HFD+TEL	32.5 ± 1.78 c*
OVX+HFD+TEL	32.0± 1.84 dns

Serum glutamate pyruvate transminase levels in NPD, SHAM, OVX, HFD, OVX+HFD. Effect of rutin and telmisartan treatment on the SGPT levels in OVX+RTN, HFD+RTN, OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, QVX+HFD+TEL, data shown are MEAN ± SEM. Mean values are significantly different for the following comparisons a vs NPD (p<0.001), b vs OVX, c vs HFD, d vs. DVX+HFD (P<0.001). Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant.

Table12: Plasma aspartate transminase levels

GROUP	MEAN ± SEM (mg/dl)
	serum glutamate oxalo acetic Transminase activity (IU/L).
NPD	51.2 ±3.11
SHAM	50.8 ±1.62
OVX	52.2 ± 1.99 a*
HFD	51.0 ± 1.88 ans
OVX+HFD	60.5 ± 4.96 a**
OVX+RTN	47.5± 1.45 bns
HFD+RTN	49.3 ± 1.76 c*
OVX+HFD+RTN	57.8 ± 2.24 dns
OVX+TEL	43.3 ± 1.82 bns
HFD+TEL	47.7 ± 1.36 c*
OVX+HFD+TEL	53.3. ± 3.74 d**

Serum glutamate oxalo acetic transminase levels in NPD, SHAM, OVX, HFD, OVX+HFD. Effect of rutin and telmisartan treatment on the SGPT levels in OVX+RTN, HFD+RTN, OVX+HFD+RTN, HFD+RTN, OVX+HFD+RTN, OVX+TEL, HFD+TEL, OVX+HFD+TEL, data shown are MEAN ± SEM. Mean values are significantlssy different for the following comparisons a vs NPD (p<0.001), b vs. OVX, c vs. HFD, d vs. OVX+HFD (P<0.001). Data was analyzed by One way ANOVA followed by Dunnett's multiple comparison tests.*P<0.05, **P<0.01, ***P<0.001, ns=non significant

Food intake during the experiments was weighed. Feeding a high fructose diet caused a marked decrease in food intake as compared to feeding normal diet, but increased food intake in ovariectomized rats. We have shown that oral administration of telmisartan decreased food intake and reduces body weight gain in both ovariectomized and high fructose fed rats.

In the present study, we demonstrate the effectiveness of swimming in preventing some of the weight gain associated with Ovariectomy. Swimming rats had lower final weight than rats that did not swim (graph 13). It is not surprising that both ovariectomized and sham rats that underwent the swimming treatment had significantly less body weight compared with non-swimming rats. This result agrees with that of [16].ovariectomized rats which are treated with rutin and telmisartan also significantly reduces body weight when they underwent for swimming training. But in our study there was not much reduced body weight difference observed between the ovariectomized exercised rats group and ovariectomized rats treated with rutin and telmisartan.

As an essential component of the metabolic syndrome blood lipid profiles also investigated in this study. Ovariectomy resulted in an increase in serum lipid levels. High fructose supplementation also increases in serum lipid levels. Ovariectomized animals which are supplemented with high fructose solution represented significantly increased cholesterol, triglyceride and Low density lipoprotein levels. However Administration of telmisartan significantly reduced serum triglycerides, LDL and total cholesterol levels. Whereas the serum levels of HDL remains unaffected. However Administration of rutin significantly reduced serum triglycerides, LDL and total cholesterol levels, but shows insignificant changes in HDL levels. Whereas several studies showed that an increase in HDL cholesterol is associated with decrease in coronary risk. It is interesting to find that in the present study rutin and telmisartan not only lowered the total cholesterol, LDL and triglycerides levels. But also enhanced the cardio protective lipid HDL levels after 6 week treatment. These would definitely reduce the incidence of coronary events.[17,18]

Plasma levels of glucose in response to OGTT are shown in graph 4. The basal levels of glucose at zero time point were not significantly different among the groups. After glucose challenge, plasma glucose levels in OVX+HFD rats were significantly higher than those in the other experimental groups at the 15 minute time and 30 minute time points. These differences were not observed after 30 minutes. At the 15 minute time point plasma glucose levels in OVX and HFD were significantly higher than those in the NPD and SHAM operated animals. At the At the 15 min and 30 min time point the plasma glucose levels were significantly reduced in OVX, HFD and OVX+HFD groups that received rutin and telmisartan treatment.

Conclusion

In conclusion we have presented evidence that both ovariectomy and over nutrition leads to the development of a systemic metabolic condition representing phenotype features of the insulin resistance syndrome including increased visceral fat content, abnormal serum lipid profile, and impaired glucose tolerance. In this study we have found that telmisartan and rutin significantly improved abnormal metabolic profile and glucose intolerance. Rutin reduces the body weight, plasma triglycerides, and total cholesterol .in addition rutin reduces the elevated serum glutamate pyruvate transminase levels. Besides, by sharing the antioxidative, anti inflammatory and antiproliferative mechanisms telmisartan showed decrease in weightgain, indicating the beneficial role of partial PPAR-γ agonist in obesity and metabolic syndrome. We predicted that swimming could be a very effective strategy to prevent the development of metabolic syndrome induced by ovariectomy. Rutin might be helpful in clinical practice. However, further studies are necessary to improve our understanding of the role of rutin in metabolic syndrome.

.

References

[1]. Reaven GM. Banting Role of insulin resistance in human disease. Diabetes 1988,37: 1595- 1600.

[2]. Sternfeld B, Bhat AK, Wang H, Sharp T, Q Menopause, physical avtivity, body compositiom/fat distribution in midlife women, Med Sci Sports Exer 2005, 37: 1195-1201.

[3]. Kamalakkannan N, Prince PSM. Rutin improves the antioxidant status in streptozotocin-induced diabetic rat tissues. Mol Cell Biochem 2006,293: 211-219.

[4]. Kurtz T. W et al. Treating the metabolic syndrome: telmisartan as a peroxisome proliferator activated receptor-gamma activator, Acta Diabetol 2005,42: S9–S16.

[5]. Dahlöf B, Burke TA, Krobot K, Carides GW, Edelman JM, Devereux RB, Diener HC. Population impact of losartan use on stroke in the European Union (EU): projections from the Losartan Intervention for Endpoint reduction in hypertension (LIFE) study. J Hum Hypertens, 2004,18(6):367-73.

[6]. raki K, Masaki T, Katsuragi I, Tanaka K, Kakuma T, Yoshimatsu H Telmisartan presents obesity and increases the expression of uncoupling protein 1 in diet-induced obese mice. Hypertension 2006,48: 51-57.

[7]. Sugimoto K, Qi NR, Kazdová L, Pravenec M, Ogihara T, Kurtz TW., Telmisartan but not valsartan increases caloric expenditure and protects against weight gain and hepatic steatosis. Hypertension. 2006;47(5):1003-9

[8]. Clasen R, Schupp M, Foryst-Ludwig A, Sprang C, Clemenz M, Krikov M, Thöne-

Reineke C, Unger T, Kintscher U, PPARgamma-activating angiotensin type-1 receptor blockers induce adiponectin. Hypertension. 2005,46(1):137-43.

[9]. Schupp M, Curtin JC, Kim RJ, Billin AN, Lazar MA. A widely used retinoic acid receptor antagonist induces peroxisome proliferator-activated receptor-gamma activity. Mol Pharmacol. 2007,71(5):1251-7.

[10]. Yamagishi S, Nakamura K, Imaizumi T. advanced glycation end products (AGEs) and diabetic vascular complications. Curr Diabetes Rev. 2005, 1(1):93-106.

[11]. Yusuf S. From the HOPE to the ontarget and the transcend studies: challenges in improving prognosis. Am J Cardiol. 2002, 24;89(2A):18A-25

[12]. Mc Cardle, W.D. Metabolic stress of endurance swim-ming in the laboratory rat Journal of Applied Physiology 1967,22: 50-54.

[13]. Astrup A, Rössner S, Lessons from obesity management programmes: greater initial weight loss improves long-term maintenance. Obes Rev. 2000, 1(1):17-9.

[14]. Sanders TA. Food safety and risk assessment: naturally occurring potential toxicants and anti-nutritive compounds in plant foods, Forum Nutr. 2003, 56: 407-9.

[15]. Srinivasan K, Patole PS, Kaul CL, Ramarao P. Reversal of glucose intolerance by by pioglitazone in high fat diet-fed rats. Methods Find Exp Clin Pharmacol. 2004,26(5):327-33.

[16]. Latour MG, Shinoda M, Lavoie JM .Metabolic effects of physical training in ovariectom- ized and hyperestrogenic rats. J Physiol 2001,90: 235-241.

[17]. Larue-Achagiotis C., Martin C., Verger C., Chabert M. & Louis-Sylvestre J Effects of acute treadmill exercise and delayed access to food on rats Physiolo and Behavior 1993,53: 403-408.

[18]. Rodgers PT, Fuke DC. New and emerging strategies for reducing cardiometabolic risk factors. Pharmacotherapy. 2006,26, 13-31