Original Research Article

ANTIHYPERLIPIDEMIC STUDIES OF METHANOLIC EXTRACT OF *GOSSYPIUM HERBACEUM*: AN *IN SILICO* AND *IN VIVO* APPROACH

ABSTRACT

Aim: To evaluate anti-hyperlipidemic activity of the whole plant of *Gossypiumherbaceum*extract in Wistar Albino Rats.

Methods: The whole plant of *Gossypiumherbaceum* were collected and extracted with methanol by soxhlation. It was tested in Triton and Propylthiouracil (PTU) induced hyperlipidemic rat models and antioxidant hydrogen peroxide radical scavenging assay.

Results: Preliminary Phytochemical screening resulted in the presence of flavonoids, terpenoids, steroids, phytosterols, carbohydrates, alkaloids, tannins and phenolic compounds. The anti-hyperlipidemic effect of methanolic extract of whole plant of *Gossypiumherbaceum* was tested in Triton and Propylthiouracil (PTU) induced hyperlipidemic rat models. Treatment with MEGH (200 and 400 mg/kg, *p.o*) significantly reduced the elevated serum lipids, restored the decreased HDL compared to disease group. Histopathological examinations showed recovery of the damaged liver cells in Propylthiouracil treated group. The extract was also evaluated for its antioxidant potential by using hydrogen peroxide radical scavenging assay. Ascorbic acid was used as standard. The results demonstrated that methanolic extract of whole plant of *Gossypiumherbaceum* possessed significant antioxidant and antihyperlipidaemic activities. Docking simulation was done to PDB protein of Lecithin cholesterol acyltransferase, HMG-CoA reductase inhibitor and Antoxidant and viewed in discover studio followed by Ramachandran plot.

Conclusion: Methanolic extract of *Gossypiumherbaceum* can be used for management of hyperlipidemia and possess antioxidant activities.

Key words: Triton-X-100, Propylthiouracil, hyperlipidemia, antioxidant, *Gossypiumherbaceum*, whole plant, rats, Atrovastatin

INTRODUCTION

Coronary heart diseases (CHD) are the main cause of death in western nations and Asia. Among CHDs, ischemic heart disease (IHD) leads to the highest death rate [1].Hyperlipidemia is a medical term for abnormally high levels of lipids in the blood including cholesterol, triglycerides and lipoproteins [2]. There are over 3 million adults throughout the United States and Europe that currently have a diagnosis of hyperlipidemia, and that number continues to rise at a drastic pace. Hyperlipidaemia is typically a chronic, progressive disease process that demands lifestyle and dietary changes, with the potential need for additional lipid-lowering medications.Hyperlipidaemiacondition in which the enhanced formation of ROS is of pivotal pathogenetic importance. Propyl thiouracil and Triton X 100 are used in induction of hyperlipidaemia and Atorvastatin and ascorbic acid serves as standard in our present study. The healthcare system is structured in such a way that natural remedies are now widely perceived as inferior or something that people use when they cannot afford modern medicine.

Gossypiumherbaceum is a bushy shrub that grows height of 2-8 feet, with few branches; stem thick and rigid belongs to family *Malvaceae*.*Gossypiumherbaceum* originated in southern Africa but was first domesticated in Arabia, from where cultivated forms spread westward to Africa and eastward to India[3]. The different parts of *Gossypiumherbaceum* are used for anticonvulsant, toxicity studies, hypoglycemic, hypolipidemic activity, antidepressant, antidiabetic activity etc. The aim of our study to evaluate anti-hyperlipidemic activity and antioxidant activity of the whole plant extract of *Gossypiumherbaceum* in Wistar Albino Rats and to perform *in-silico* analysis.

2. MATERIALS AND METHODS

The designing of methodology involves a series of steps taken in a systematic way in order to evaluate anti-hyperlipidemic activity of the whole plant extract of *Gossypiumherbaceum* in Wistar Albino Rats

2.1 Plant collection, drying & pulverization:

All parts of *Gossypiumherbaceum* was collected and identified. The crude plant material is authenticated by P. Suresh babu, Govt Degree College, Hyderabad, Telangana. The freshly collected parts of the plant were cleared from dirt and then dried under shade for about 15 days and coarsely powdered in a mixer grinder. The powdered material was stored or taken up for extraction process.

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2.2 Preparation of *Gossypiumherbaceum* extract

Soxhlet extraction is the process of continuous extraction in which the methanol solvent can be circulated through the extractor for several times. This process involves extraction followed by evaporation of the solvent. The vapours of the solvent are taken to a condenser and the condensed liquid is returned to the drug for continuous extraction

2.3 Experimental animals

Adult wistar albino rats (180 –200 g) were procured from Albino Labs Hyderabad and used for the pharmacological activities. They were kept in polypropylene cages at 25 ± 2° C, with relative humidity 45-55% under 12h light and dark cycles. All the animals were acclimatized to the laboratory conditions for a week before use. They were feed with standard animal feed and water ad libitum. All the pharmacological experimental protocols were approved by the Institutional animal ethics committee (IAEC)

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2.4 Preliminary phytochemical screening

The Methanolic extract of *Gossypiumherbaceum*was subjected to preliminary phytochemical screening to identify various phytoconstituents present in *Gossypiumherbaceum*

2.5 Acute toxicity testing

Acute toxicity study was carried out in order to check the toxic effects for methanolic extract of *Gossypiumherbaceum*. The study was performed as per Organization for Economic Cooperation and Development (OECD). The method is used to evaluate the acute oral toxicity is up and down procedure (OECD guideline-425). Up and down procedure (OECD guideline-425) acute toxicity studies were carried out as per the OECD 425 guidelines.

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2.6 *In-vitro* Anti-oxidant assay

2.6.1 H_2O_2 radical scavenging activity

Hydrogen peroxide is a week oxidizing agent and can inactivate a few enzymes directly, usually by oxidation of essential thiol (-SH) groups. Hydrogen peroxide can cross cell membranes rapidly, once inside the cell, H_2O_2 can probably react with Fe^{+2} and possibly Cu^{+2} ions to form hydroxyl radical and this may be the origin of many of its toxic effects. It is therefore biologically advantageous for cells to control the amount of hydrogen peroxide that is allowed to accumulate [4].

Procedure: A solution of hydrogen peroxide (2mmol/L) was prepared in phosphate buffer (pH 7.4). Test compounds (10-50 µg/mL) were added to hydrogen peroxide solution (0.6 mL). Absorbance of hydrogen peroxide at 230 nm was determined after 10 min against a blank solution containing phosphate buffer without hydrogen peroxide and compared with ascorbic acid, the reference compound

$$\%H_2O_2 = \text{Abs control - Abs sample} \div \text{Abs control} \times 100$$

Where 'Abs (control)' is absorbance of control 'Abs (sample) is absorbance of extract/standard [5].

2.7*In-vivo* Anti hyperlipidaemic activity

2.7.1 Propylthiouracil induced hyperlipidaemia

The proposed method of induction with PTU per oral 10 mg kg-1 bd.wt followed by induction of cholesterol solution in vegetable oil of 400 mg/kgbd.wt dose produced hyperlipidaemia animal models that have cholesterol metabolism disruption. Adult Wistar albino rats were administered with corresponding treatments for 8 days. They were divided into 5 groups with 6 animals per group. Study design of High fat diet induced hyperlipidaemia method is Group –I serves as Control (Normal saline). Group II (disease control), III, IV and V received PTU and cholesterol powder along with respective treatment, Group –III received methanolic extract of *Gossypiumherbaceum* (100mg/kg, *p.o*) and Group – IV received *Gossypiumherbaceum* (200mg/kg, *p.o*) and atorvastatin (100mg/kg, *p.o*).

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Animals were orally induced with propylthiouracil of 10 mg/kgbd.wt. Dosage and 0.01% PTU in drinking water for 7 days. On day 8 test drugs were given to animals orally. One hour after test drugs administration, animals were given a solution of high dosage cholesterol in vegetable oil of 400 mg/kgbd.wt. Serum total cholesterol level was measured in every 1 h after administration of cholesterol for 6 h. After 6 h, a level of total cholesterol in the liver and faeces were measured. This method is simpler and requires less time to get hyperlipidaemia animal model. Serial measurement of serum total cholesterol level in every hour for 6 h gave a cholesterol profile that can explain different drug mechanisms in cholesterol homeostasis [6].

2.7.2 Triton induced hyperlipidaemic rat model

Hyperlipidaemia was induced in Wistar albino rats by single *intraperitoneal* injection of freshly prepared solution of Triton X-100 (100 mg/kg bd.wt) in physiological saline solution after overnight fasting for 18 h. The animals were divided into four groups of containing five rats each group. Group I was given standard pellet diet, water (1% acacia). Group II was given a single dose of Triton X-100 administered at a dose of 100 mg/kg, bd.wt, i.p. After 48 hrs of Triton injection, this group received a daily dose of 1% acacia (*p.o)* for 7 days. Group III and Group IV was administered METP at doses 200 mg/kg bd.wt and 400 mg/kg bd.wt for 7 days. Group V was administered standard atorvastatin 10 mg/kg bd.wt, *i.p*for 7 days. On the blood was collected by retro orbital sinus puncture. The collected

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samples were centrifuged for 10mins. Then serum samples were collected and used for estimation of various biochemical experiments [7].

2.8 *Insilico*analysis

2.8.1 Molecular docking

The mechanism of binding of drug with the target protein is called docking. Docking can be used to find inhibitors for specific target proteins and thus to design new stable drugs from docking results. Docking can be calculated by binding energy (energy release during protein and ligand interaction). In this project, mCule software was used for docking [8].

2.8.2 Structure based drugdesign

Initially the protein downloaded from PDB was prepared by selecting any one chain. Water molecules present in the chains are removed. Attributes selected. Later docking is performed by mCule online software *i.e* protein-ligand docking performed for protein 6MVD, 3CCZ and 1URM.

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2.8.3 mCule dockingresults

Docking indicates that some of our compounds have good binding ability with LCAT activator (PDB ID: 6MVD), HMG-CoA reductase inhibitor (PDB ID: 3CCZ) and Antoxidant (PDB: 1URM)

2.8.4 Ramachandran plot

Ramachandran plot has been generated from PROCHECK validation server which was used to access the quality of the model by looking into the allowed and disallowed regions of the plot [9].

2.9 Statistical analyses

The Results were expressed as the m1ean ± S.E.M. The significance of theresults was calculated using ANOVA andDunnett'stest and results were deliberatedstatistically noteworthy when significant $p<0.0001$, $p<0.001$, $p<0.01$, ns- non significant.

3. RESULTS

Methanolic extract of *Gossypiumherbaceum* was explored for its Anti-hyperlipidaemic activity using animal models and its antioxidant activity was screened by in-vitro antioxidant assays. All the results obtained in this study were included below.

3.1 Preparation of methanolic extract of *Gossypiumherbaceum*

The methanolic extract of *Gossypiumherbaceum* was prepared by soxhlation technique. The percentage yield of the extract was obtained about 7.3% w/w

3.2 Preliminary phytochemical analysis

The preliminary phytochemical investigation for methanolic extract of *Gossypiumherbaceum* showed the presence of flavonoids, terpenoids, tannins & phenolic compounds, alkaloids, steroids, terpenoids, phytosterols, proteins.

3.3 Acute toxicity studies

Limit test was selected in present toxicity study: Methanolic extract of *Gossypiumherbaceum* was tested on female mice at the dose of 2000 mg/kg bd.wt. *p.o*methanolic extract of *Gossypiumherbaceum* did not exhibit any signs of toxicity and mortality even up to 2000 mg/kg. bd.wt. All animals were safe even after 14 days of observation. So based on limit studies, the working dose was considered as $1/20^{th}$ i.e., 100 mg/kg bd.wt

3.4 *In vitro* Antioxidant assay

3.4.1 Hydrogen peroxide radical scavenging activity

In hydrogen peroxide radical scavenging assay, IC_{50} value for the MEGH and standard drug ascorbic acid was found to be 32 and 38 µg/mL respectively. From this result is clear that MEGH showed good antioxidant property.

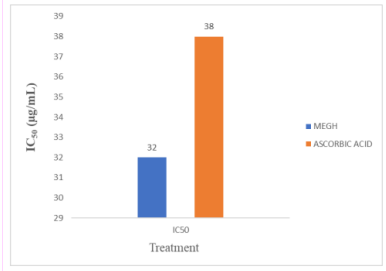

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Figure 1: Hydrogen peroxide radical assay of MEGH and Ascorbic acid

The hydrogen peroxide is extremely reactive free radical formed in the biological system has been implicated as a highly damaging species, capable of damaging almost every molecule found in the living cell. The radicals have the capacity to join the nucleoside in DNA and can cause strand breakage. In addition these species are considered to be rapid initiators of the lipid peroxidation process due to abstraction of hydrogen atom from unsaturated fatty acid. The ability of MEGH to quench hydroxyl radicals seems to directly relate to prevention of propagation of process of lipid per oxidation and the formulation seems to be good scavenger of ROS. The reducing power activity of MEGH might be due to its presence of phenolic, flavonoids compound.

3.5 *In vivo* Anti hyperlipidemic activity

Methanolic extract of *Gossypiumherbaceum* was explored for its antihyperlipidaemic activity in Propylthiouracil induced and Triton induced hyperlipidaemic rat models. All results obtained in this study were included below.

3.5.1 Propylthiouracil induced hyperlipidaemic rat model

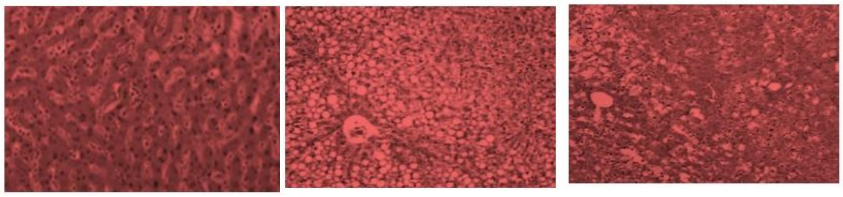
Table 1: Anti-hyperlipidaemic activity for methanolic extract of *Gossypiumherbaceum* on Propylthiouracil induced hyperlipidaemic rat model

Treatment	Lipid levels (mg/dL)				
	TC	TG	HDL	LDL	VLDL
Normal Control	98.1 ± 4.14	90.5 ± 6.96	37.7 ± 1.64	90.2 ± 1.56	22.5 ± 0.79
Disease Control	98.1 ± 4.14	236.5 ± 13.5**	17.4 ± 0.52**	156 ± 2.77**	70.4 ± 2.89**
MEGH (100 mg/kg)	127.1 ± 3.75**Bb	193.3 ± 4.64**Ab	22.1 ± 0.68**Ab	131.5 ± 3.45**Bb	50.8 ± 1.78**Bb
MEGH (200 mg/kg)	127.1 ± 3.75**Bb	165.5 ± 1.77**Ba	25.2 ± 0.69**Aa	121.6 ± 1.72**Bb	42.7 ± 0.95**Ba
Atorvastatin (10 mg/kg)	77.7 ± 0.86**B	126.7 ± 3.45**B	30.83 ± 1.7**B	102.1 ± 2.28**B	34.2 ± 0.83**B

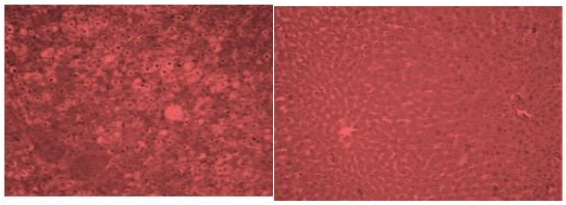
Values are expressed as Mean ± SEM, (n=6). Statistical analysis was performed by using ANOVA followed by Dunnett's test. (* = $p<0.0005$, ** = $p<0.0001$) when compared to control group, (A = $p<0.0001$, B=$p<0.001$), when compared to disease control group, (a =$p<0.0001$, b =$p<0.001$) when compared to standard group.

The propylthiouracil induced hyperlipidaemic rat model showed significant increase in serum biochemical parameters like TC, TG, LDL, VLDL levels and decrease in serum HDL when compared to normal control group. The MEGH treated group at 100 mg/kg showed significant decrease in TC, TG, LDL, VLDL and increase in HDL when compared to disease control group. The MEGH treated group at 200 mg/kg decreased TC, TG, LDL, VLDL and increased HDL. The standard group showed significant decrease in TC, TG, LDL, VLDL and increase in HDL.

3.5.2 Histopathology of Liver



a) Control group b) Disease control group c) MEGH (100 mg/kg)



d) MEGH (200 mg/kg) e) Atorvastatin (10 mg/kg)

Fig 2: Histopathology of liver in Propylthiouracil induced hyperlipidemic rat model

Control group showed following parameters where Bile duct appeared normal, no inflammation or fibrosis noticed surrounding the portal region of liver. Kupffer cells and sinusoids are normal. No evidence of fatty change and fibrosis. Hyperlipdemic group disease control showed Cord pattern of hepatocytes. Few periportal lymphocytes in focal area fibrosis noticed in periportal region of liver. Fatty change found in cytoplasm and fibrosis. Histopathology of rat liver treated with MEGH 100 mg/kg revealed moderate sinusoidal space dilatation along with haemorrhages in the sinusoidal space of liver and few periportal lymphocytes in focal area and Histopathology of rat liver treated with MEGH 200 mg/kg showed Mild Cord pattern of hepatocytes. Mild sinusoidal space dilation along with haemorrhage.Kupffer cells are normal. Histopathology of rat liver treated with atorvastatin 10 mg/kg showed Hepatocytes, Kupffer cells and sinusoids appeared normal and no evidence of fat deposition.

3.5.3Triton X-100 induced hyperlipidemic rat model

Table 2: Anti-hyperlipidemic activity for methanolic extract of Gossypiumherbaceum on Triton X-100 induced hyperlipidemic rat model

Treatment	Lipid levels (mg/dL)				
	TC	TG	HDL	LDL	VLDL
Normal Control	178.2± 1.84	124.5±2.4	56±3.01	98±0.30	24.6±2.18
Disease	291 ±	241.3 ±	15.9 ±	177 ±	72.63 ±

Control	2.91*	6.24*	1.2*	4.65*	3.21*
MEGH (100 mg/kg)	260.3 ± 3.03*Ab	193.7 ± 1.24*Ab	25.08 ± 0.45*Ab	121.8 ± 3.63*Ab	58.5 ± 1.57*Ab
MEGH (200 mg/kg)	250.2 ± 1.24*Ab	169.2 ± 4.06*Aa	28.08 ± 0.87*Ab	112.9 ± 2.95**Ab	55 ± 1.43*A
Atorvastatin (10 mg/kg)	220.98 ± 2.7*A	149.6 ± 1.39**A	40.3 ± 0.73*A	73.2 ± 1.53*A	38.3 ± 3.27**A

Values are expressed as Mean ± SEM, (n=6). Statistical analysis was performed by using ANOVA followed by Dunnett's test. (* = p< 0.0001, ** = p<0.0005) when compared to control group, (A = *P*=0.001) when compared to disease control group, (a = *P*=0.0005, b = *P*=0.0001) when compared to standard group.

The Triton X-100 induced hyperlipidemic rat model showed significant increase in serum biochemical parameters like TC, LDL, VLDL levels and decrease in HDL level when compared to normal control group. The MEGH treated group at 100 mg/kg showed significant decrease in TC, TG, LDL, VLDL and increase in HDL when compared to disease control group. The MEGH treated group at 200 mg/kg decreased TC, TG, LDL, VLDLand increased HDL. The standard group showed significant decrease in TC, TG, LDL, VLDL and increase in HDL.

3.6 *In silico* analysis

3.6.1 Molecular docking

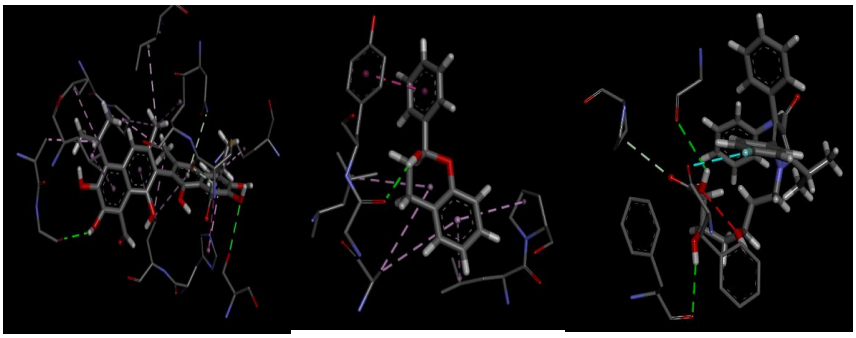
Table 3: Docking score of chemical constituents and amlodipine with protein 6MVD, 3CCZ, 1URM.

Compounds	6MVD	3CCZ	1URM
Cyanidin	-6.9	-7.5	-5.6
Delphinidin	-7.1	-6.7	-5.5
Flavon-3-ol	-7.7	-6.8	-6.0
Catechin	-7.3	-7.4	-5.6
Gallocatechin	-7.4	-6.5	-5.4
Epicatechin	-7.3	-7.4	-5.6
Epigallocatechin	-7.4	-6.5	-5.5
Gossypol	-8.1	-6.8	-6.0

Lactic acid	-4.1	-3.5	-3.1
Palmitic acid	-5.6	-4.4	-3.7
Stearic acid	-5.9	-4.0	-3.4
Atorvastatin	-4.7	-6.9	-5.5

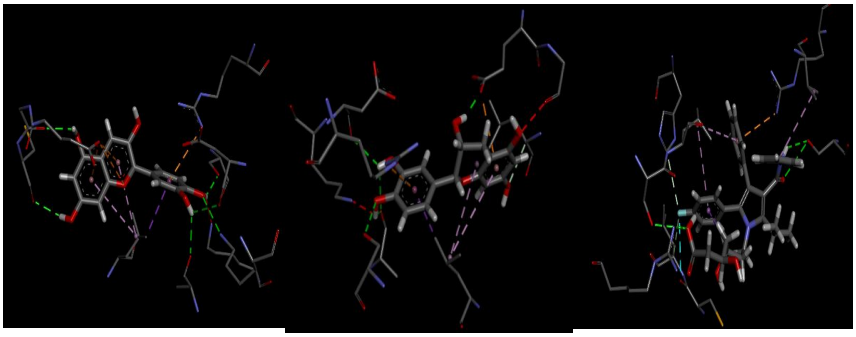
G score = glide score, the more negative the Glide score, the more favourable the binding.

1) 6MVD



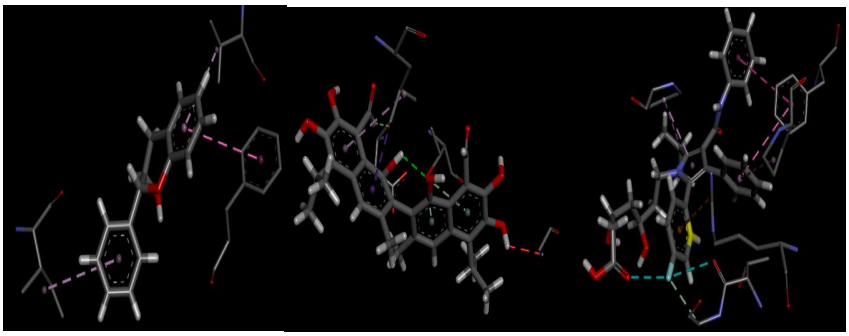
a) Gossypol -8.1 **b) Flavon-3-ol -7.7** **c) Atorvastatin -4.7**

2) 3CCZ



a) Cyanidin -7.5 **b) Catechin -7.4c) Atorvastatin -6.9**

3) 1URM



a) Flavon-3-ol -6.0 **b) Gossypol -6.0** **c) Atorvastatin -5.5**

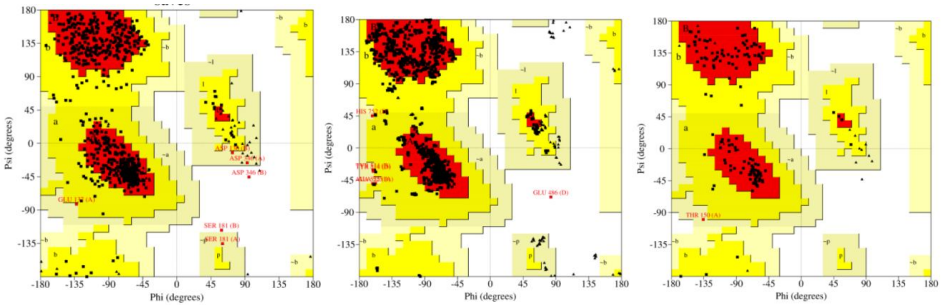
Figure 3: Hydrophobic bond interactions of ligands with 6MVD, 3CCZ, 1URM protein

3.6.2Ramachandran plot Analysis

Protein 6MVD, 3CCZ and1URM were analysed for Ramachandran plot to know amino acid presence in different regions of respective protein tabulated in table 4 and pictorial representation by figure below.

Table 4: Ramachandran plot statuswith protein with 6MVD, 3CCZ and 1URM.

Residues	6MVD	3CCZ	1URM
Most favourable region (%)	86.1	91.1	89.4
Additional allowed regions (%)	12.99	8.5	9.8
Generously allowed regions (%)	0.6	0.4	0.8
Disallowed regions (%)	0.3	0.1	0.0



a) 6MVD **b) 3CCZ** **c) 1URM**

Figure 4: Ramachandran plot of protein 6MVD, 3CCZ and 1URM protein

4. DISCUSSION

High levels of triglycerides reflect increased concentrations of the triglyceride rich lipoproteins, chylomicrons and VLDL, and their remnants. Hyperlipidaemia relates to increased oxidative stress causing significant production of oxygen free radicals, which may lead to oxidative modifications in low density lipoproteins, which presents a significant function in the initiation and progression of atherosclerosis and associated cardiovascular diseases.Antioxidants are compounds that act as inhibitors of the oxidation process and are found to inhibit oxidant chain reactions at small concentrations and thereby eliminate the threat of pathological processes [10]. Methanolic extract of *Gossypiumherbaceum*shown better IC 50 value in comparision to standard ascorbic acid.

Propylthiouracil is used for treatment of hyperthyroidism accompanied by hypercholesterolemia, an increase of total cholesterol, LDL and triglycerides in blood serum that have cholesterol metabolism disruption. Triton X-100, a non-ionic detergent provokes acute hyperlipidaemia, by increasing hepatic cholesterol biosynthesis [11].

In the present study, METP was investigated for hypolipidemic effect by Triton X-100 and propylthiouracil induced models. This study deals with the effect of MEGH (100 & 200 mg/kg bd.wt) in hyperlipidaemia rats shown significant reduction in plasma and hepatic lipid profiles along with elevation in plasma HDL in MEGH treated as compared to hyperlipidaemia rats, thus indicating the efficacy of MEGH in preventing the elevation seen in various components of lipid profile under PTU & Triton X 100 experimentally induced hyperlipidaemia. Epidemiological studies have shown that a higher level of HDL in plasma reduces the risk of coronary artery disease. Flavonoids in MEGH are reported to increase HDL and decrease LDL and VLDL levels in hypercholesteremic rats. Statins decrease total cholesterol and LDL-C via inhibition of 3-hydroxy-3- methylglutaryl-coenzyme A (HMG-CoA) reductase. Multiple studies have clearly shown that statins can decrease LDL-C levels, thereby decrease mortality and disease progression among patients with clinical ASCVD [12]. Atorvastatin showed decrease in TG, TC, LDL, VLDL and increase in HDL compared to disease control group.

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Hence the hypolipidemic effect of MEGH *i.e*decrease in TG, TCL, LDL, VLDL and elevation in HDL facilitates the transport of triglyceride or cholesterol from serum to liver where it is catabolized and excreted out of the body and produce an antioxidant effect. *Insilico*analysis by molecular docking revealed plant constituents showed response to LCAT activator, Inhibited HMG-CoA reductase and antioxidant activity. Lecithin cholesterol acyltransferase (LCAT) is one of the major modulators of plasma high-density lipoprotein

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cholesterol (HDL-C) and plays a central role in the reverse cholesterol transport (RCT) process. LCAT is a plasma enzyme that circulates mostly in association with the high density lipoproteins (HDL) and is responsible for the synthesis of cholesterol esters present in human plasma. Cholesterol esterification catalysed by LCAT also reduces the amount of unesterified cholesterol in plasma [13]. Flavone 3-ol, Gossypol, Cyanidin andcatechin showed good docking score compared to other compounds. Statins remain the most prescribed drugs for the treatment of hyperlipidaemia. They competitively inhibit 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase. Deleterious effect of postprandial hyperlipidemia is mediated via induction of oxidative stress [14].Ramachadran plot showed the presence of amino acid in most favourable region is greater than 86%. In the present study the superposition of Flavone 3- ol, Gossypol, Cyanidin, catechinand other compounds docking found withLCAT activator (PDB ID: 6MVD), HMG-CoA reductase inhibitor (PDB ID: 3CCZ) and Antioxidant (PDB: 1URM) have validated the accuracy of our docking study and Ramachandran plot.Flavonoids are a major class of phenolic compounds present in MEGH and are found to have a potential role in prevention of hyperlipidaemia through their antioxidant activity.

5. CONCLUSION

Methanolic extract of *Gossypiumherbaceum* has shown significant antioxidant action againsthydrogen peroxide radical assay andsignificantly lowered the triglycerides, total cholesterol levels, LDL and VLDL levels and increased HDL when results were compared to control group and disease control group in PTU induced and Triton X-100 induced hyperlipidemia models. The histopathology study of *Gossypiumherbaceum* showed recovery of the damaged liver cells in Propylthiouracil treated group. The cells of the intoxicated liver were reformed. The degree of vascularization was also reduced as compared to hyperlipidemic group that may due to presence of cyanidin, delphinidin, gossypol, catechin and flavonoids. The phytoconstituents present in MEGH showed good docking score compared to standard atorvastatin. Ramachandran plot showed the presence of aminogroups in favourable region is >86%. Hence it can be concluded that Methanolic extract of *Gossypiumherbaceum* has showed significant antihyperlipidemic activity.

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