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Hypolipidemic activity of *Murraya koenigii* in Alloxan induced Diabetic rats

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Abstract
Type-2 diabetes mellitus is a metabolic disorder characterized by persistent hyperglycemia, abnormal lipid, carbohydrate and proteins metabolism. *Murraya koenigii* is known for its widespread use as spice/herb in Indian dishes and its therapeutic uses in Ayurveda. We studied the hypolipidemic activity of *Murraya koenigii* in alloxan induced diabetic rats. The study was conducted using eighteen wister rats of 180-250 g. Diabetes was induced by administration of 150 g/kg intraperitoneal alloxan. Animals were separated into three groups with six animals in each group. One group received saline and served as controls, whereas other two groups received Atorvastatin 10mg/kg/PO and *Murraya koenigii* at a dose of 200 mg/kg/PO for one week. Diagnostic reagent kit was used to estimate plasma glucose concentration, Plasma triglyceride concentration, and total cholesterol concentration. Statistical values were expressed as mean ± SEM. One-way ANOVA analyzed inferential statistics between mean. A two-tailed p-value was considered statistically significant. Administration of the test drug *Murraya koenigii* produced statistically significant reduction in the Triglycerides level (p<0.0001) which was 125.6 before treatment and reduced to 114.5 after treatment. Whereas the cholesterol level was 135.8 before treatment and reduced to 123 after treatment (p<0.0001). Aqueous leaf extract of *Murraya koenigii* has shown the hypolipidemic effect in diabetic rats which is comparable to Atorvastatin. Thus our study concludes, *Murraya koenigii* may have beneficial effects in the management of dyslipidemia in type -2 DM. The existing literature and easy availability of *M. koenigii* in India thus make it a smart lead molecule for further clinical research.

Keywords: *Murraya koenigii*, Atorvastatin, Type-2 diabetes mellitus, dyslipidemias

Introduction
Type-2 diabetes mellitus is a metabolic disorder, characterized by persistent hyperglycemia, abnormal lipid, carbohydrate and proteins metabolism. This will enhance the risk of cardiovascular disease (CVD) complications [1]. Hence, dyslipidemia is one of the major risk factors and treatment with statins will reduce diabetic patients' morbidity and mortality. Even though drug therapy for dyslipidemia must be individualized, it often needs treatment with multiple agents to achieve therapeutic goals. The availability of several lipid-lowering drugs in the market is effective options for achieving target lipid levels in people with diabetes [2-3]. Diabetic dyslipidemia comprised a triad of raised triglycerides, LDL and reduced HDL and it is due to insulin resistance and deficiency affects key enzymes in lipid metabolism [4]. Conventionally, several herbal medicines have been used for the treatment of type-2 diabetes as an alternative medicine/nutritional supplements [5]. Therefore, medicinal plants have the potential to impart therapeutic effect in chronic disorders like diabetes and its complications [6]. *Murraya koenigii* is known for its extensive use as spice/herb in Indian dishes and also for its therapeutic uses in Ayurveda [7]. The plant has also been studied for their various pharmacological activities like antioxidant, antibacterial, antifungal, antiprotozoal [8] hypoglycemic, anti-hyperglycemic activity [9] and hypolipidemic activity [10] Screening of medicinal plants is one of the alternatives and valid approaches in the drug development process since they contain varied phytoconstituents which may give lead component which can be effective and safe in diabetes [11]. To this rationale, we studied the hypolipidemic activity of *Murraya koenigii* in alloxan induced diabetic rats.

Methods
The study was conducted using eighteen Wister rats of 180-250 g. Institutional animal ethics committee approved the study proposal (17/2011/NMC). Diabetes was induced by administration of 150 g/kg intraperitoneal alloxan (Sigma Pvt. Ltd.). Animals were separated

into three groups with six animals in each group. One group received saline and served as controls, whereas other two groups received Atorvastatin (Cipla, 10mg/kg/PO) and *Murraya koenigii* at a dose of 200 mg/kg/PO for one week.

Plants Materials/Preparation of the Extract [12]

Fresh leaves of the *Murraya koenigii* was obtained from the local market and approved by acertified botanist from Vikrama Simhapuri University, Nellore. The fresh leaves were oven dried at 45 ^{0}C, ground into powder and 20g will be soaked in 300ml of distilled water overnight at room temperature. The filtrate obtained was evaporated in a hot-air oven at 45 ^{0}C. The extract was weighed (2g) and reconstituted in an appropriate volume of distilled water before administration to the rats. At the end of seven days of extract administration, 1ml of blood samples were collected from the tail directly into anticoagulant bottles contains sodium fluoride. Centrifugation separated the plasma. Diagnostic reagent kit was used to estimate plasma glucose concentration, Plasma triglyceride concentration, and total cholesterol concentration. The total cholesterol was estimated by using cholesterol oxidase/peroxidase standard kit consisting of enzyme reagents, diluents buffer, cholesterol standard 200mg/dl, obtained from Reck on Diagnostics Pvt. Ltd., Triglycerides estimation kit consists of enzyme reagent sand Triglycerides standard, was obtained from Trans Asia Biomedicals Ltd.

Statistical Analysis

Values were expressed as mean ± SEM. One-way ANOVA analyzed inferential statistics between mean. A two-tailed p-value was considered statistically significant. Microsoft excel-2007 and Graph Pad prism were used for data management and statistical analysis respectively.

Results

Triglycerides: In the control group, the Triglycerides values were 123.8 and 124.6 respectively before and after administration of control (normal saline). There was no significant increase in the Triglycerides level indicating that vehicle (normal saline) used to dissolve test and standard drugs was not having any Triglycerides lowering action. In atorvastatin group, Triglycerides levels decreased from 127.3 to 99.6 after seven days of treatment which was statistically highly significant. Administration of the test drug *Murraya koenigii* produced statistically significant reduction in the Triglycerides level (p<0.0001) which was 125.6 before treatment and reduced to 114.5 after treatment. Thus, overall the Triglycerides lowering action was more with Atorvastatin >*Murraya koenigii* >Normal Saline.

LDL- Cholesterol: In the control group, the cholesterol values were 134.5 and 135.3 respectively before and after administration of control (normal saline). There was no significant increase in the Triglycerides level indicating that vehicle (normal saline) used to dissolve test and standard drugs was not having any cholesterol lowering action. In atorvastatin group, cholesterol level decreased from 135.1 to 103 after seven days of treatment which was statistically highly significant. Administration of the test drug *Murraya koenigii* produced statistically significant reduction in the cholesterol level (p<0.0001) which was 135.8 before treatment and reduced to 123 after treatment. Thus, overall the LDL lowering action was more with Atorvastatin >*Murraya koenigii* >Normal Saline.

Table: Glucose, Triglycerides & Cholesterol Before and After Test and Standard Drug Administration

	Control		Atorvastatin		*Murraya koenigii*	
	Before	**After**	**Before**	**After**	**Before**	**After**
Glucose (mg/dl)	257 ± 8.9	288 ± 10.2	235.1 ± 10.9	233.33 ± 11.2	240.1 ± 6.6	224 ± 7.9***
TG (mg/dl)	124.6 ± 2.0	134.2± 6.0	127.3 ± 5.7	99.6 ± 6.6***	125.6 ± 6.2	114.5±9.6**
TC (mg/dl)	135.3 ± 3.7	144.8 ± 4.2	135.1 ± 7.3	103 ± 9.2***	135.8 ± 5.2	123 ± 8.0**

*** P<0.001, ** P<0.01when compared to before drug administration using paired t-test

Discussion

In the traditional system of Murraya was used as an antiemetic, anti-diarrhoeal, dysentery, febrifuge, blood purifier, tonic, stomachic, a flavoring agent in curries and chutneys [13]. Active compounds like dichloromethane and ethyl acetate extracts of *Murraya koenigii* leaves significantly reduced the body weight gain, plasma total cholesterol and triglyceride levels significantly when given orally at a dose of 300 mg/kg/day to the high-fat diet-induced obese rats for two weeks [14].

Kesari *et al.* [15] observed the hypoglycemic activity of the aqueous extract of the Murraya keonigii leaves in normal as well as streptozotocin-induced diabetic rats and confirmed Murraya has got the favourable effect in bringing down the severe complications of diabetes.

Dineshkumar *et al.* [16] who has also demonstrated that *Murraya koenigii* exhibited antidiabetic and hypolipidemic effects in streptozotocin-induced diabetic rats. In his study, treatment with *Murraya koenigii* (at a dose 200 mg/kg, p.o) for seven days significantly reduced total cholesterol, triglycerides compared to the control, the weight loss in diabetic rats may be associated with lipid lowering activity of *Murraya koenigii* or due to its influence on various lipid regulation systems.

Birari R *et al.* [17] also confirmed that the body weight gain, plasma total cholesterol, and triglyceride levels were significantly reduced by ethyl acetate extracts of Murraya keonigii when given orally at a dose of 300 mg/kg per day to the high-fat-diet-induced obese rats for two weeks.

Our study results were also supported by Khan BA *et al.* [18-19] who has studied extensively, the biochemical response, hematological and histological studies in rats by supplementation of Murraya keonigii and mustard to their diet.

Conclusion

Aqueous leaf extract of *Murraya koenigii* has shown the hypolipidemic effect in diabetic rats which is comparable to Atorvastatin. Thus our study put forwards, *Murraya koenigii* may have beneficial effects in the management of dyslipidemia in type -2 DM. The existing literature and easy availability of *M. koenigii* in India thus make it a smart molecule for further clinical research. *Murraya koenigii* leaves can be used as add-on therapy along with standard pharmacology therapy for dyslipidemias.

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