

Pharmacological Screening of Polyherbal Formulation for Diabetic Associated Hyperlipidemia

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Article History Received: 12 June 2023 Revised: 12 Sept 2023 Accepted: 02 Dec 2023 CC License CC-BY-NC-SA 4.0	Abstract Plant parts such as seeds, berries, roots, leaves, bark, or flowers can be used medicinally. This practise is known as herbal medicine, botanical medicine, or phytomedicine. Outside of mainstream treatment, herbalists have long used herbalism for conventional medicine. <i>Grewia subinaequalis</i> DC in the family Tiliaceae, only one genus, <i>Grewia</i>, yields edible fruit. <i>Saccharum officinarum</i> is a species of grass belonging to the genus <i>Saccharum</i> that is characterised by its robust growth and size. <i>Neisosperma oppositifolium</i> is a tree that typically exhibits a height range of 6 to 25 metres, although it has been observed to vary from as low as 2.5 metres to extraordinarily high heights of 45 metres, and in rare cases, even up to 60 metres. The presence of moisture in crude pharmaceuticals is an unavoidable factor that should be minimised to the greatest extent possible^{9,10}. The process of drying significantly influences both the quality and purity of the material. Key Words: Hyperlipidemia, Phytomedicine, HPTLC, Polyherbal formulation, Antidiabetic.
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Introduction

The method of making medicinal use of plant parts such as seeds, berries, roots, leaves, bark, or flowers is known as herbal medicine, botanical medicine, or phytomedicine.^{1,2} There is a long history of herbal medicine being used in contexts outside of mainstream medicine.^{3,4,5} There are many different ailments that can be prevented and treated with herbal medicine, which is often referred to as alternative medicine.⁶ Herbal remedies offer a number of benefits that cannot be found in modern synthetic or chemical compounds.⁷ Today, people all around the world are becoming more and more aware of the significance of returning to nature as a means of combating a variety of health issues⁸. Natural treatments derived from herbs are absolutely free of any adverse effects and can be found in nature⁹.

Grewia subinaequalis DC.

Among the Tiliaceae family, only one of the genera, *Grewia*, produces fruit that can be consumed. *G. subinaequalis*, which has been referred to in literature for a long time as *G. asiatica*, is the only species that of any significance¹⁰. In a country like India, where there are many different dialects of names, the vernacular name that is most commonly used is L. Phalsa. In Pakistan, the plant is specifically referred to as Phalsa.

Saccharum officinarum

Saccharum officinarum refers to a specific species of sugarcane. The Poaceae family consists of a vast and robust species of grass belonging to the *Saccharum* genus¹¹. The sturdy stems of this plant contain a high concentration of sucrose, a type of sugar that builds up in the sections between the stalks.

Neisosperma oppositifolium (Lam.)

The scientific name for *Ochrosia oppositifolia* is the Apocynaceae family consists of trees that typically reach heights ranging from 6 to 25 metres, although they can vary in size from as small as 2.5 metres to as tall as 45 metres, and in unusual cases, up to 60 metres^{12,13}.

HPTLC FINGERPRINTING ANALYSIS

HPTLC is a method employed to separate and identify various constituents. The separation process relies on the disparity in adsorption coefficients between the distinct constituents of a mixture, whereas identification is determined by comparing the R_f values^{14,15}. Compounds that have a higher degree of adsorption to the stationary phase will have a slower upward movement compared to compounds with lower adsorption, resulting in the separation of the compounds. Chromatographic investigations were conducted in accordance with references¹⁶.

Fig: 1. HPTLC chromatogram of Aqueous extract of *Grewia subinaequalis* DC. Heartwood

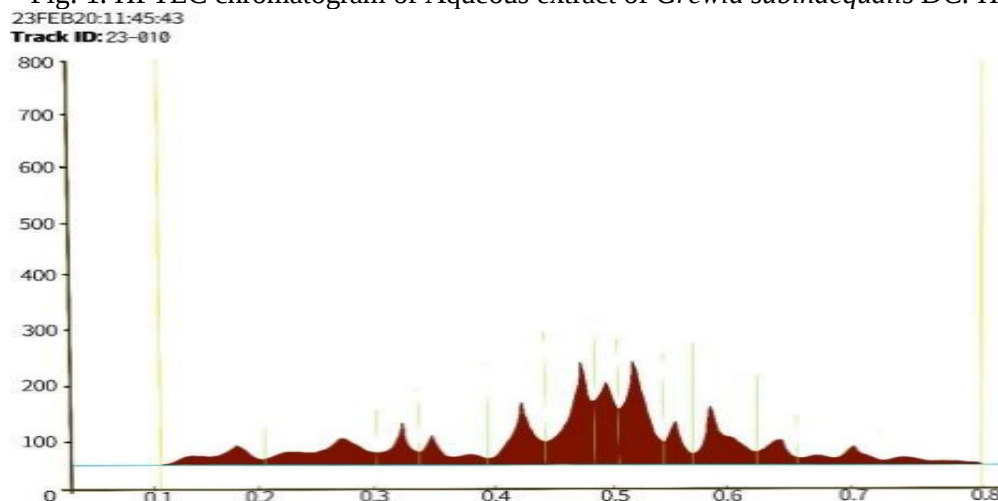
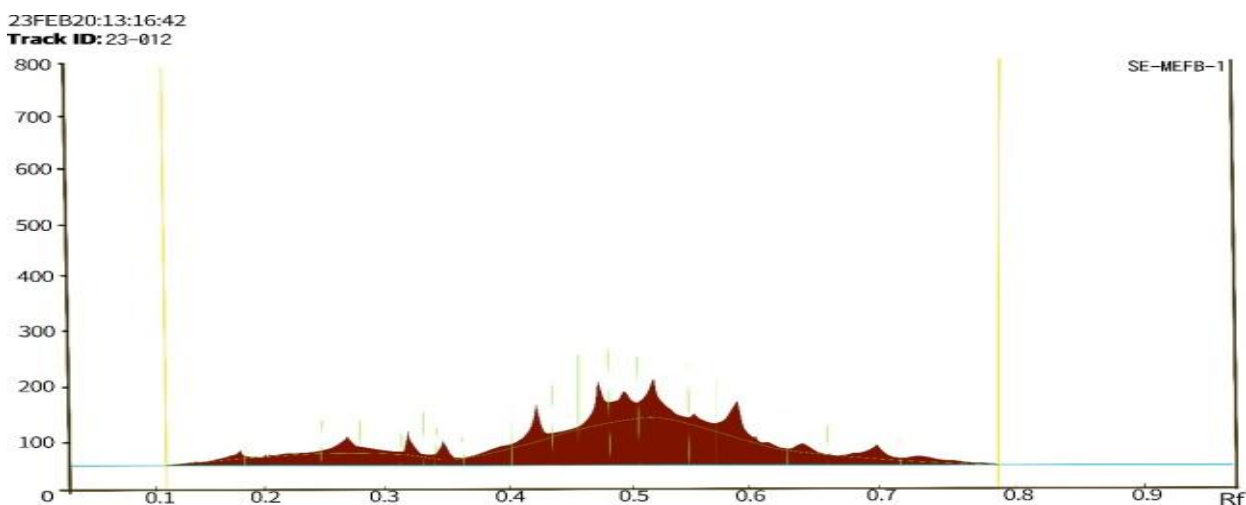
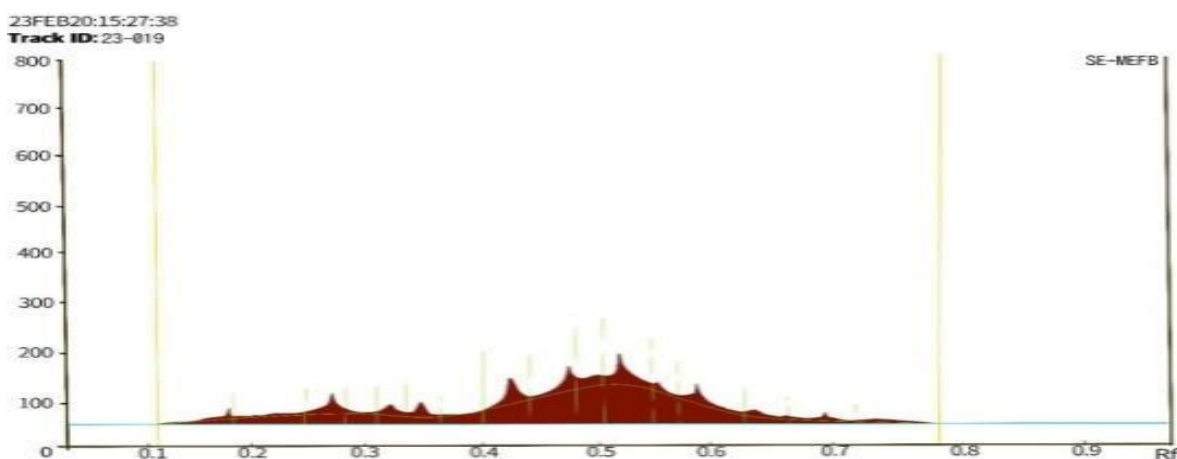


Table 1. HPTLC chromatogram of Aqueous extract of *Grewia subinaequalis* DC. Heartwood

Peak	Start point	Maximum R _f	End point	Peak height (AU)	Area (AU)	Percentage area (%)
1	0.15	0.18	0.21	91.5	549	7.81
2	0.26	0.27	0.3	109.3	437.2	6.22
3	0.3	0.32	0.33	130.7	392.1	5.58
4	0.33	0.35	0.36	112.4	337.2	4.8
5	0.39	0.42	0.44	184.2	921	13.11
6	0.44	0.47	0.48	243.5	974	13.86
7	0.48	0.49	0.5	207.1	414.2	5.89
8	0.5	0.51	0.54	249.6	998.4	14.21

Fig: 2. HPTLC chromatogram of Aqueous extract of *Saccharum officinarum* L. LeavesTable: 2. HPTLC chromatogram of Aqueous extract of *Saccharum officinarum* L. leaves

Peak	Start point	Maximum Rf	End point	Peak height (AU)	Area (AU)	Percentage area (%)
1	0.17	0.18	0.2	100.5	301.5	3.902
2	0.26	0.28	0.3	109.8	439.2	5.68
3	0.3	0.2	0.33	124.1	372.3	4.81
4	0.33	0.35	0.36	153.7	461.1	5.96
5	0.38	0.39	0.39	92.3	92.3	1.19
6	0.39	0.42	0.44	238.4	1192	15.42
7	0.45	0.47	0.48	316.2	948.6	12.27
8	0.48	0.49	0.5	256.9	513.8	6.65

Fig: 3. HPTLC chromatogram of Aqueous extract of *Neisosperma oppositifolium* (Lam.) Fosb BarkTable 3. HPTLC chromatogram of Aqueous extract of *Neisosperma oppositifolium* (Lam.) Fosb Bark

Peak	Start point	Maximum Rf	End point	Peak height (AU)	Area (AU)	Percentage area (%)
1	0.14	0.17	0.18	91.2	364.8	6.81
2	0.24	0.26	0.28	108.6	434.4	8.11

3	0.31	0.32	0.33	117.4	234.8	4.38
4	0.34	0.35	0.36	101.2	202.4	3.78
5	0.4	0.42	0.43	177.1	531.3	9.92
6	0.46	0.47	0.48	209.8	419.6	7.84
7	0.48	0.49	0.5	193.9	387.8	7.24
8	0.5	0.51	0.54	212.4	849.6	15.87

Fig. 4. HPTLC chromatogram of Aqueous extract of polyherbal tablet

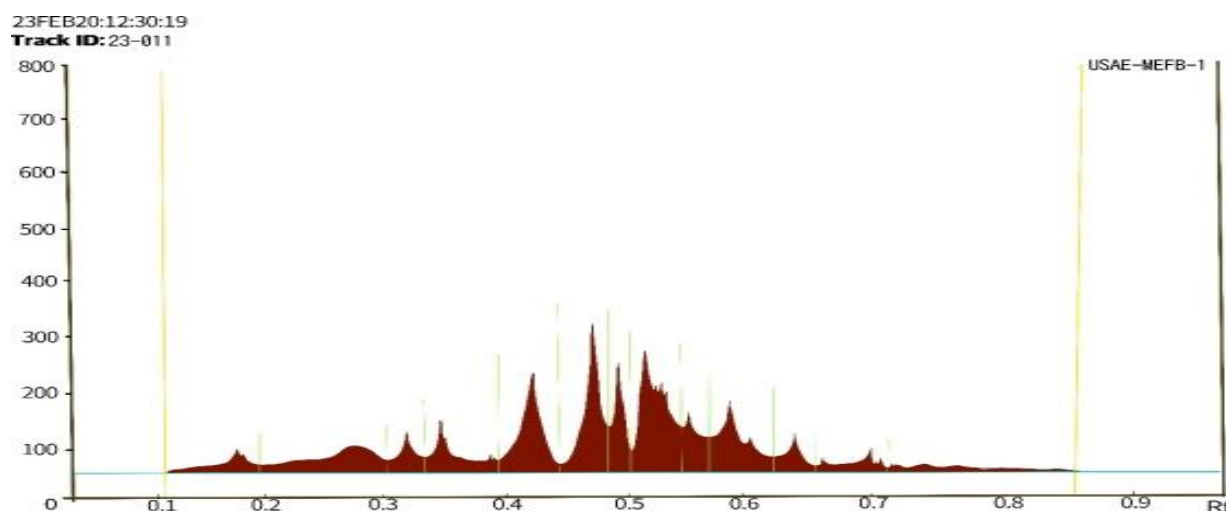


Table 4. HPTLC chromatogram of Aqueous extract of polyherbal tablet

Peak	Start point	Maximum Rf	End point	Peak height (AU)	Area (AU)	Percentage area (%)
1	0.16	0.17	0.18	95.7	191.4	4.60
2	0.25	0.27	0.28	114.4	343.2	8.25
3	0.31	0.32	0.33	98.2	196.4	4.72
4	0.34	0.35	0.37	101.6	304.8	7.33
5	0.4	0.42	0.44	150.4	601.6	14.47
6	0.45	0.47	0.48	178.2	534.6	12.86
7	0.5	0.52	0.54	199.4	797.6	19.18
8	0.54	0.55	0.57	132.2	396.6	9.54

Results and Discussion

Acute oral toxicity studies of aqueous extracts

The body weight of the rats was measured both before and after the medication was given, and it was found that there were no changes in the respiratory, circulatory, autonomic, central nervous system, eyes, mucous membranes, skin and fur, motor activity, or behaviour patterns. There were also no signs of tremors, convulsions, salivation, diarrhoea, lethargy, sleep, or coma. There was also no observation of the beginning or indicators of toxicity. Further investigation revealed that these amounts did not cause any toxicity or fatalities.

Table: 5. Acute Toxicity test studies of AEGS

Sl. No	Groups	Dose/kg (body weight) p.o	Weight of Rats		Signs of Toxicity	Onset of Toxicity	Duration of Study
			Before Test (g)	After Test (g)			

1.	AEGS	2000 mg	176	178	No signs of Toxicity	Nil	14 days
2.	AEGS	2000 mg	172	173	No signs of Toxicity	Nil	14 days
3.	AEGS	2000 mg	221	220	No signs of Toxicity	Nil	14 days

AEGS- Water Extract of *Grewia subinaequalis* DC. Heartwood

Table: 6. Acute toxicity studies of AESO

Sl. No	Groups	Dose/kg (body weight) p.o	Weight of Rats		Signs of Toxicity	Onset of Toxicity	Duration of Study
			Before Test (g)	After Test (g)			
1.	AESO	2000 mg	175	174	No signs of Toxicity	Nil	14 days
2.	AESO	2000 mg	150	148	No signs of Toxicity	Nil	14 days
3.	AESO	2000 mg	190	194	No signs of Toxicity	Nil	14 days

AESO- Aqueous Extract of *Saccharum officinarum* L. leaves

Table: 7. Acute toxicity studies of AENO

Sl. No	Groups	Dose/kg (body weight) p.o	Weight of Rats		Signs of Toxicity	Onset of Toxicity	Duration of Study
			Before Test (g)	After Test (g)			
1.	AENO	2000 mg	176	178	No signs of Toxicity	Nil	14 days
2.	AENO	2000 mg	179	175	No signs of Toxicity	Nil	14 days
3.	AENO	2000 mg	185	183	No signs of Toxicity	Nil	14 days

AENO- Aqueous Extract of *Neisosperma oppositifolium* (Lam.) Fosb Bark

Conclusion

Weight gain and hypoglycemia are two conditions that can occur as a result of taking sulfonylureas, which are both for oral anti-diabetic drugs that are currently in using in the medication. In order to bring the increased glucose levels back down to normal, the extracts and the prescription that was taken on a regular basis were helpful.¹⁷⁻²⁰ As compared to the usual group of rats, the water extracts at a dosage of 250mg/kg revealed a glucose level of 193mg/dL, which is not that important²¹⁻²³. When rats were given AESO at a dosage of 250 mg/kg, their glucose levels dropped to 130 mg/dL, which is not statistically important when compared to normal rats. Water extracts AENO, on the other hand, demonstrated a substantial reduction in glucose levels to 120 mg/dL when given at a dosage of 250 mg/kg. In the other hand, the Herbal tablet at 250mg/kg resulted in a substantial reduction in glucose to normal levels. The same outcomes were observed in the groups given the regular synthetic medication. This group performed higher than any extract in terms of lowering blood glucose levels. The extracts, on the other hand, were professional enough to illustrate comparable behavior. In contrast to the diabetic control group, the overall lipid profile in serum (TG, TC, HDL, LDL, and VLDL) of STZ mediated diabetes animals treated with Polyherbal tablets was significantly increased. These findings indicate that PHT can inhibit the cholesterol synthesis pathway, and that the increased HDL/LDL ratio is due to the activation of LDL receptors in hepatocytes, which are responsible for absorbing LDL into the liver and lowering serum LDL levels.

REFERENCES

1. Agarwal SS, Paridhavi M. Extraction, isolation and analysis of phytopharmaceuticals. Herbal drug technology. 2007:324-6.

2. Saravanan R, Dhachinamoorthi. D, Senthilkumar. K and Thamizhvanan. K. Antimicrobial activity of various extracts from various parts of *Calophyllum inophyllum* L. Journal of Applied Pharmaceutical Science. 2011; 01 (03): 102-106
3. Sharma A, Shanker C, Tyagi LK, Singh M, Rao CV. Herbal medicine for market potential in India: an overview. Acad J Plant Sci. 2008;1(2):26-36.
4. Aneesh TP, Hisham M, Sekhar S, Madhu M, Deepa TV. International market scenario of traditional Indian herbal drugs–India declining.... International Journal of Green Pharmacy (IJGP). 2009;3(3).
5. Annapurna A., & D. Mahalakshmi, & Kota, Muralikrishna. Antidiabetic of a polyherbal preparation (tincture of punchparna) in normal and diabetic rats". Indian journal of experimental biology, 2001; 39: 500-2.
6. Moore PH, Paterson AH, Tew T. Sugarcane: the crop, the plant, and domestication. Sugarcane: physiology, biochemistry, and functional biology. 2013 Dec 6:1-7.
7. Balakrishnaiah, P., & Satyanarayana, T. Preparation and evaluation of polyherbal formulation for its antidiabetic activity against streptozotocin induced diabetes rat model. Pharmaceutical and Biological Evaluations. 4(111) 10.26510/2394-0859.pbe.2017.17.
8. Cantrill DJ, Poole I. Taxonomic turnover and abundance in Cretaceous to Tertiary wood floras of Antarctica: implications for changes in forest ecology. Palaeogeography, Palaeoclimatology, Palaeoecology. 2005 Jan 6;215(3-4):205-19.
9. Chandra A, Mahdi AA, Ahmad S, Singh RK. Indian herbs result in hypoglycemic responses in streptozotocin-induced diabetic rats. Nutrition research. 2007 Mar 1;27(3):161-8.
10. Chowdary KP, Koteswara Rao N, Malathi K. Ethyl cellulose microspheres of glipizide: Characterization, in vitro and in vivo evaluation. Indian Journal of pharmaceutical sciences. 2004;66(4):412-6.
11. Chowdary KP, Balatripura Sundari G. Design and evaluation of mucoadhesive controlled release oral tablets of glipizide. Indian journal of pharmaceutical sciences. 2003;65(6):591-4.
12. Edwin J, Balakrishnan JS, Chandra JD. Diabetes and herbal medicines. Iranian Journal of Pharmacology and therapeutics. 2008;7(1):97-106.
13. Evans WC. Trease and Evans' pharmacognosy. Elsevier Health Sciences; 2009 May 27.
14. Frier B. M., Fisher M. Diabetes mellitus, In: David Son's, Principles & Practice of medicine, edited by Nicholas A. Boon, Nicki R. Colledge, Brain P. Walker, John A. A. Hunter, 20th edition, Churchill Livingstone Elsevier, Philadelphia, USA. 2006; Pg: 905-847.
15. Frode TS, Medeiros YS. Animal models to test drugs with potential antidiabetic activity. Journal of ethnopharmacology. 2008 Jan 17;115(2):173-83.
16. Ghorbani A, Shafiee-Nick R, Rakhshandeh H, Borji A. Antihyperlipidemic effect of a polyherbal mixture in streptozotocin-induced diabetic rats. Journal of lipids. 2013 Dec 5;2013.
17. Garcia J, Ghaly ES. Evaluation of bioadhesive glipizide spheres and compacts from spheres prepared by extruder/marumerizer technique. Pharmaceutical development and technology. 2001 Jan 1;6(3):407-17.
18. Haque MR, Ansari SH. Antihyperlipidemic activity of a unani formulation in high fat diet-induced obese murine model. Trends in Phytochemical Research. 2018 Jun 1;2(2):83-90.
19. Ismail IS, Gezawa ID, Yahya MF, Chedi B, Nafisatu K, AA I. Anti-hyperglycemic effect of gasca d herbal formulation on alloxan induced diabetic rats. Diabetes Management. 2018;8(4):85-93.
20. Agarwal SS, Paridhavi M. Extraction, isolation and analysis of phytopharmaceuticals. Herbal drug technology. 2007:324-6.
21. Sharma A, Shanker C, Tyagi LK, Singh M, Rao CV. Herbal medicine for market potential in India: an overview. Acad J Plant Sci. 2008;1(2):26-36.
22. Shanmugarathinam A, Mallikarjun V, Saravanan R.. Phytocompounds used as excipients in mucoadhesive systems, an outline on buccal tablets and films,. Int J Ind Herbs Drugs 2017; 2(3): 44-48.
23. Balakrishnaiah, P., & Satyanarayana, T., (2017). Preparation and evaluation of polyherbal formulation for its antidiabetic activity against streptozotocin induced diabetes rat model. Pharmaceutical and Biological Evaluations. 4(111) 10.26510/2394-0859.pbe.2017.17.