REFERENCES

1. Kour P, Bilandi A, and Kataria MK, Swami H. Sustained release matrix tablets of miglitol: techniques implemented and patents. International Journal of Institutional Pharmacy and LifeSciences.2015; 5(1):88-100.

2. Kara D. D., Krishna V. T., Pai G. K. A Review on Manufacturing of Tablets by Using Various Granulation Techniques // J. Global Pharma Technol. – 2017. – V. 10, N 9. – P. 05–10.

3. Wachtel GS, Benzie IFF. Herbal Medicine: An Introduction to its History, Usage, Regulation, Current Trends, and Research Needs. Herbal Medicine: BiomolecularandClinicalAspects 2011.2nd edition.

4. Canter P. H, Ernst E. Herbal supplement use by persons aged over 50 years inBritain: Frequently used herbs, concomitant use of herbs, nutritional supplementsandprescriptiondrugs,rateofinformingdoctorsandpotentialfornegativeinteractions.Drugs Aging.2004;21:597–605.

5. Tilburt J. C, Kaptchuk T. J. J. Herbal medicine research and global health: An ethical analysis. Bull World Health Organ. 2008;(8):594–9.

6. Shinde, et al.: Formulation and evaluation of antidiabetic polyherbal tablets. AsianJournalofPharmaceutics 2021;15(2):250-53

7. Hasan S, Vedant Misra, Swati Singh, Garvita Arora, Sunita Sharma and Sarika Sharma,Current status of herbal drugs and their future perspectives, Biological Forum–An International Journal 2009;1(1),12-17

8. Cragg G M, Newman D J,Sander K M,Natural products in drug discovery and development, Journal of Natural Products 1997;60:52-60.

9. Prabhakar Reddy Veera Reddy.Herbal drugs and formulations. Biochem & Pharmacol 2013; 2(4):213.

10. Kwakye KO, Doriskumadoh. Dosage forms of herbal medicinal products and their stability considerations –an overview.J CritRev 2017;4(4):1-8

11. Siddiqui, Ameena & Shukla, Sudhir. (2015). Conservation of Plant Genetic Resources and Their Utilisation in Global Perspective. LS: International Journal of Life Sciences. 4. 10.5958/2319-1198.2015.00007

12. Sarangi MK, Padhi S. Novel herbal drug delivery system: An overview. Arch Med Health Sci 2018;6:171-9

13. https://blogs.baruch.cuny.edu/herbal and pharmaceutical medicines.

14. http://1epir.com/en/pages/mdex/id

15. Neeraj Choudhary, Bhupinder Singh Sekhon. An over view of advances in the standardization of herbal drugs.J PharmEducRes.2011;2(2):55-70.

16. Calixto J B. Efficacy, safety, quality control, marketing and regulatory guidelines for herbal medicines (Phytotherapeutic agents) Brazilian Journal of Medical and Biological Research 2000; 33(2):179-189.

17. http://www.pharmainfo.net/Herbal medicines and its standardization.

18. Kumadoh, Doris, Ofori-Kwakye, Kwabena. Dosage forms of herbal medicinal products and their stability considerations-an overview. Journal of Critical Reviews 2007; 4:1-8.

19. https://www.researchgate.net/publication/319242213 Dosage forms of herbal medicinal products and their stability considerations an overview.

20. Balandrin MF, Klocke JA, Wrtele EA,Boilinger WH. Content and purity of extracts Olasodine in some available species of Solanum, Science and culture 1985; 56(5) ,214-216.

21. https://www.Pharmapproach.com/advantages-and-disadvantages-of powders.

22. Knight J. Endocrine System 1: Overview of the endocrine system and hormones. Nursing Times [online] 2021; 117(5):38-42

23. Michelle A, Clark, Richard Finkel, Jose A Rey, Karen Whalen. Lippincott's Illustrated reviews Pharmacology.5th edition.

24. Williams Textbook of Endocrinology. 12th edition. Elsevier Publishers. 22

25. https://www.mayoclinic.org/diseases-conditions/diabetes/in-depth/diabetes-treatment/art/20044084

26. https://www.cdc.gov/diabetes/basics/type2.html

27. Jain R, Jain P, Jain P.A Review on Treatment and Prevention of Diabetes Mellitus.

Int J Curr Pharm Res, Vol.8,Issue 3,16-18.

28. https://www.healthline.com/health/diabetes/Treatment,diagnosis,prevention.

29. https://www.niddk.nih.gov/health-information/diabetes/overview/insulin-medicines-treatments.

30. Marín-Peñalver, J. J., Martín-Timón, I., Sevillano-Collantes, C., & Del Cañizo-Gómez, F. J. (2016). Update on the treatment of type 2 diabetes mellitus. *World journal of diabetes*, *7*(17), 354–395. https://doi.org/10.4239/wjd.v7.i17.354

31. Hussain S A, Namilikonda M G, Chandra T K, Pasha M A. A Review on Medicinal Plants with Anti-Diabetic Activity.Int.J.Adv.Res.2020; 8(03):902-917.

32. Verma S, Gupta M, Popli H, Aggarwal G. Diabetes Mellitus Treatment Using Herbal Drugs: A review. International Journal of Phytomedicine 2018;10(1):1-10

33. Abdulmohsen N, Swedan B, Amer M G, Robert A A. The Role of Herbal Medicines in the Treatment of Diabetes: A Short Review. International Journal of Science and Research (IJSR).2021;10(11):1294-97

34. Modak, M., Dixit, P., Londhe, J., Ghaskadbi, S., & Devasagayam, T. P. (2007). Indian herbs and herbal drugs used for the treatment of diabetes. *Journal of clinical biochemistry and nutrition*, *40* (3), 163–173.

35. Kambale, E. K., Quetin-Leclercq, J., Memvanga, P. B., & Beloqui, A. (2022). An Overview of Herbal-Based Antidiabetic Drug Delivery Systems: Focus on Lipid- and Inorganic-Based Nanoformulations. *Pharmaceutics*, *14*(10), 2135.

36. Kumar R, Janadri S, Kumar S, Dhanajaya DR, Swamy S. Evaluation of antidiabetic activity of alcoholic extract of flower *Sesbania grandiflora* in alloxan induced diabetic rats. Asian Journal of Pharmacy and Pharmacology 2015; 1(1):21-26.

37. Gyawali R & Acharya G & Pokharel M & Shah A & Silwal A. Evaluation of Antidiabetic Polyherbal Formulations. Advance Research Journal of Medicinal and Clinical Sciences2015; 1(1):33-37.

38. Mawlieh B, Shastry S & Chand S. Evaluation of Anti-diabetic Activity of two marketed Herbal Formulations. Research Journal of Pharmacy and Technology

2020; 13(02):664-668

39. Sharma A, Kaushik K. Formulation and Evaluation of Herbal Antidiabetic Tablet 2011; Journal of Drug Delivery &Therapeutics; 2011; 1(1):65-67.

40. Suruse P B, Kale M K, Duragkar N J, Gundawar A. Formulation and Evaluation of Antidiabetic Herbal Capsules. Research J. Pharma. Dosage Forms and Tech. 2011; 4(2):113-118.

41. Sahu MK, Singh VK, S Rao PS. Development and Evaluation of Antidiabetic Potential of Polyherbal Formulation in Streptozotocin Induced Animal Model. Int J cell Sci & Molbiol.2018;5(2):0029-0037

42. Nidhi,N.C.,Rujuta,M.,Drasti,M.,Ismail,U.S.,Ezaj,D.andVaishnavi,C.P.Formulation , Evaluation and Comparison of the Poly Herbal Anti-Diabetic Tablet with the Commercial Tablets. Journal of Pharmaceutical Research International 2021; 33 (37A):252-263.

43. Arora R,Mittal A, Jha K K. Formulation and Evaluation of Herbal Anti-diabetic Formulation containing Eugenia jambolana, Gymnema sylvestre, Tinospora cordifolia,Pterocarpus marscipum, Terminalia bellerica & Emblica officinalis.ThePharma Innovation–Journal 2013;2(5):210-216.

44. Aziz Namra,Wal P,Wal Ankita, Saxena S. Evaluation of a Polyherbal Powder for Treatment of Alam S, Baig A, Reddy S K, Reddy M K, Mohiuddin M, Reddy M V, Gupta R K. Antidiabetic and Diabetes Mellitus .Indian J Pharma Sci 2019;81(6):1070-1077.

45. Uddandrao S V V, Bramhanaidu P, Ganapathy S. Evaluation of the Antioxidant and Antidiabetic Potential of the Polyherbal Formulation: Identification of Bioactive Factors. Cardiovascular & Hematological Agents in Medicinal Chemistry. 2020; 18(2):111-123.

46. Shinde S A, Jain S, Chavan S, Shukla K. Formulation and Evaluation of Antidiabetic Polyherbal Tablets.Asian Journal of Pharmaceutics.2021;15(2):250

47. Alam S et al. Antihyperlipidemic Effects of Aqueous Extract of Polyherbal Formulation (ZiabeeteinPowder)inExperimentalAnimals.Int.J.Pharm. Phytopharmacol.Res. 2013,2(4):263-267

48. Suman M, Shivalinge G K P, Paul U, Priyanka S.Evaluation of Antidiabetic and Antihyperlipidemic Activity Of Newly Formulated Polyherbal Antidiabetic Tablets In Streptozocin-Induced Diabetes Mellitus In Rats. Asian Journal of Pharmaceutical and Clinical Research2016; 9(1):202-207.

49. Patel K, Hingorani L, Jain V. Formulation Development and Evaluation of AntidiabeticPolyherbalTablet.Int.J.Pharm.Sci.Rev.Res.2017;42(2):146-151.

50. Kumodh D, Ofori-Kwakye K. Dosage Forms of Herbal Medicinal Products and Their Stability Considerations- An Overview. Journal of Critical Reviews 2017; 4(4):1-8.

51. Parasuraman S, Thing GS, Dhanaraj SA. Polyherbal formulation: Concept of Ayurveda.Pharmacognosy Revision.2014;8(16):73-80.

52. Gupta R et.al. Formulation and evaluation of herbal effervescent granules incorporated with Calliandra haematocephala leaves extract. IndoAmerican Journal of Pharm Research.2013; 3(6):4366-4371.

53. Kothari S, Thangavelu L, Roy A. Anti-diabetic activity of Sesbania grandiflora – alpha amylase inhibitory effect.J Adv Pharm Edu Res 2017;7(4):499-502

54. Panigrahi G, Panda C, Patra A. Extract of *Sesbania grandiflora* Ameliorates Hyperglycemia in High Fat Diet-Streptozotocin Induced Experimental Diabetes Mellitus.Scientifica 2016:1-10.

55. Singh H, Arora S, Mani M, Mahaur K, Phool C. Development of multicomponent formulation of herbal drugs for evaluation of Antidiabetic activity. Der Pharmacia Lettre, 2014, 6(1):219-223.

56. Sabale K D, Sabale K D, Kathawate G S, Mane S S. Formulation and Evaluation of Herbal Antidiabetic Tablet.AsianJ.Res.Pharm.Sci.2020; 10(3):145-148.

57. Shrivastava S. Panda P, Vishwakarma D K, Verma N K, Nayak J. Formulation and evaluation of herbal tablets containing Agaricus bisporus powder. International Journal of Advances in Pharmaceutics 2017; 06(02):63-69.

58. Saifi A, Chauhan R, Dwivedi J. Development of a polyherbal formulation FMST and evaluation for antidiabetic activity in alloxan induced diabetic rats. Asian J. Pharm. Res.2017; 7(1):1-7.

59. Chauhan L, Vashisht S. Formulation and evaluation of novel herbal antidiabetic transdermal patch.Innov Pharm Pharmacother 2018;6(4):61-64.

60. Gauttam V K, Kalia A N. Development of polyherbal antidiabetic formulation encapsulated in the phospholipids vesicle system. JAdvPharmTechnol Res. 2013; 4(2):108-17.

61. Jyothi D, Koland M, Sneh P, James J P.Formulation of Herbal Capsule Containing Trigonella Foenum-Graecum Seed Extract for the Treatment of Diabetes.Journal of Young Pharmacists, 2017; 9(3):352-356.

62. Telapolu S, Kalachavedu M, Punnoose AM, Bilikere D. MD-1, a poly herbal formulation indicated in diabetes mellitus ameliorates glucose uptake and inhibits adipogenesis - an invitro study. BMC Complement Altern Med. 2018 Apr2; 18(1):113.

63. Petchi R R, Chockalingam V, Parasuraman S. Antidiabetic Activity of Polyherbal Formulation in Streptozotocin – Nicotinamide Induced Diabetic Wistar Rats. Journal of Traditional and Complementary Medicine, 2014; 4(2):108-117.

64. Panda A, Jena S, Sahu PK, Nayak S, Padhi P. Effect of Polyherbal Mixtures on theTreatment of Diabetes.ISRN Endocrinology 2013:1-5.

65. Farghaly U, Kamel M S, Elzahwey A S, Mangoura S A. Design and Formulation of Novel Pharmaceutical Capsules of Herbal Origin for Diabetes Mellitus. International Journal of PharmaceuticalSciences and Research, 2014;5(4):1474-81.

66. Mandlik R V, Desai S K, Naik S R. Antidiabetic activity of a polyherbal formulation (DRF/AY/5001). Indian Journal of Experimental Biology, 2008; 46(8): 599-606.

67. Majumdar P, Paridhavi M. Physiocochemical Standardization and Formulation Development of Poly-herbal Tablet for Diabetes. British Journal of Pharmaceutical Research.2016; 12(3):1-17.

68. Mandal, I., Jannat, H., Rahman, S., Jahan, R., Khan, T., Mou, S. M., & Rahmatullah, M. (2014). Antihyperglycemic and Antinociceptive Activity Tests with Beta Vulgaris L. SSP. Vulgaris Roots: A Preliminary Report. *World Journal of Pharmaceutical Research*, *3*(9), 109-118.

69. Al-Harbi LN, Alshammari, Al-Dossari AM, Subash-babu P, Binobead MA, AlhussainMH, Alsedairy SA, Al-Nouri Doha, Shamlan G. *Beta vulgaris* L. (Beetroot) Methanolic Extract Prevents Hepatic Steatosis and Liver Damage in T2DM Rats by Hypoglycemic, Insulin-Sensitizing, Antioxidant Effects, and Upregulation of PPARα. MDPI biology.2021; 10(12):1306.

70. Kumar S, Shachi K, Dubey NK. Anti-Diabetic and Haematinic Effects of Beet Root Juice(Beta vulgaris L.) in Alloxan Induced Type-1 Diabetic Albino Rats. Journal of DiabetesResearchandTherapy.2020;6(1):1-3.

71. Chauhan N N, Mistry R, Mandale D, Ismail U S, Ezaj D, Patel V C. Formulation, Evaluation and Comparison of the Polyherbal Anti-Diabetic Tablet with the Commercial Tablets.Journal of Pharmaceutical Research International. 2021; 33(37A):252-263.

72. Harshmal Kadt, Sakunthala H S. Evaluation of the effect and efficacy of herbal powder preparation derived from 'Thalpathe Piliyam'in the management of type II Diabetes mellitus. Young Ayurveda Researchers' and Innovators' Symposium (YARIS – 2019), Institutional Research Committee, Gampaha Wickramarachchi Ayurveda Institute, University of Kelaniya, Yakkala, and SriLanka.2019:40.

73. Manekar S S, Rangari V D, Agrawal M N, Rathod S M. Formulation and Anti-Diabetic Activity Studies of Herbomineral Formulation for Treatment of Diabetes. International Journal of Pharmaceutical Sciences and Research.2014;5(9):3912-3917.

74. Jain S, Pandhi P,Singh A P,Malhotra S. Efficacy of Standardized Herbal Extracts in Type 1 Diabetes - An Experimental Study.Afr.J.Traditional,Complementary and Alternative Medicines.2006; 3(4):23-33.

75. Kumar, Ajeet & Kumar, Rajesh.Formulation and evaluation of ayurvedic anti-diabetic drug. International Journal of Pharmaceutical Sciences and Drug Analysis 2022; 2(1): 30-37

76. Mahajan N, Lokhande B, Thenge R, Gangane P, Dumore N.Polyherbal formulation containing antioxidants may serve as a prophylactic measure to diabetic cataract: Preclinical investigations in rat model. Pharmacognosy

Magazine. 2018; 14(58): 572-577.

77. Gilchrist M, Winyard P G, Fulford J, Angela C A. Shore, Benjamin N. Dietary nitrate supplementation improves reaction time in type 2 diabetes: Development and application of a novel nitrate-depleted beetroot juice placebo.Elsevier.2014;40:67-74.

78. Thissera B, Visvanathan R, Khanfar M A, Qader M M, Hassan M H A, Hassan H M, Bawazeer M, Behery F A, Yaseen M, Liyanage R, Abdelmohsen U R, Rateb M E. Sesbania grandiflora L. Poir leaves: A dietary supplement to alleviate type 2 diabetes through metabolic enzymes inhibition, South African Journal of Botany. Elsevier 2020; 130:282-299.

79. Reddy KS, Sudheer A, Pradeepkumar B, Reddy CS. Effect of a polyherbal formulation in streptozotocin-induced diabetic nephropathy in wistar rats.Indian J Pharmacol 2019;51:330-6

80. L Pari, R Ramakrishnan, S Venkateswaran,ntihyperglycaemic effect of Diamed, a herbal formulation, in experimental diabetes in rats, Journal of Pharmacy and Pharmacology, 200;53(8):1139–1143.

81. Mamatha M K, Suma US, Annegowda HV. The Ascent of Polyherbal Formulation in the Treatment of Diabetes Mellitus. Res.J.Pharmacognosy and Phytochem.2020; 12(4):256-260.

82. Debnath B, Manna K. Formulating anti-diabetic nutraceutical tablets based on edible plants from Tripura, India.Foods and Raw Materials.2022; 10(2):227–234.

83. Panigrahi G, Panda C, Patra A. Extract of Sesbania grandiflora Ameliorates Hyperglycemia in High Fat Diet-Streptozotocin Induced Experimental Diabetes Mellitus.Scientifica (Cairo).2016; 2016:4083568.

84. Kumar R, Janadri S, Kumar S, Swamy S. Evaluation of antidiabetic activity of alcoholic extract of flower Sesbania grandiflora in alloxan induced diabetic rats. Asian Journal of Pharmacy and Pharmacology.2015;1(1):21-6.

85. Ramasubbu K, Padmanabhan S, Al-Ghanim K A, Nicoletti M, Govindarajan M, Sachivkina N, Rajeswari V D. Green Synthesis of Copper Oxide Nanoparticles Using Sesbania grandiflora Leaf Extract and Their Evaluation of Anti-

Diabetic,Cytotoxic,Anti-Microbial,andAnti-Inflammatory Properties in an In-Vitro Approach. Fermentation.2023Mar27;9(4):332.

86. Abdullah R, Arshad H, Kaleem A, Iqtedar M, Aftab M, Saleem F. Assessment of angiotens in converting enzyme inhibitory activity and quality attributes of yoghurt enriched with Cinnamomum verum, Elettaria cardamomum, Beta vulgaris and Brassica oleracea.Saudi Journal of Biological Sciences. 2023 Feb1;30(2):103556.

87. Al-Harbi L N, Alshammari G M, Al-Dossari A M, Subash-Babu P, Binobead M A, Alhussain M H, AlSedairy S A, Al-Nouri D M, Shamlan G. *Beta vulgaris* L. (Beetroot) Methanolic Extract Prevents Hepatic Steatosis and Liver Damage in T2DM Rats by Hypoglycemic, Insulin-Sensitizing, Antioxidant Effects, and Upregulation of PPARα. *Biology*.2021; 10(12):1306.

88. Dubey, Nagina Kumar & Kumar, Sanjeev &Shachi, Kumari & Dubey, Usha. (2020).Anti-Diabetic and Haematinic Effects of Beet Root Juice (Beta vulgaris L.) in Alloxan Induced Type-1 Diabetic Albino Rats. Journal of Diabetes Research and Therapy. 6.10.16966/2380-5544.150.

89. Türkyilmaz I, Bayrak B, Saçan Ö, Kabasakal L, Sener G, Yanardağ R.Chard (beta vulgaris l. varcicla) extract inhibits polyopathway and hyperglycaemia-induced oxidative stress in ratlens.farmacia.2022;70(1).

90. Isabela Micheletti Lorizola, Josiane Érica Miyamoto, Ana Luiza FigueiredoVieira, Beatriz Rocchetti Sumere, Rosângela Maria Neves Bezerra, Marcio Alberto Torsoni, Adriana Souza Torsoni, Mauricio Ariel Rostagno, Marciane Milanski, Caroline Dário Capitani, Beet (Beta vulgaris L.) stalk and leaf supplementation changes the glucose homeostasis and inflammatory markers in the liver of mice exposed to a high-fat diet, Food Chemistry:Molecular Sciences,2021;2:2666-5662.

91. Helmy, S. A., Morsy, N. F. S., Elaby, S. M., & Ghaly, M. A. H. A. (2024). Antidiabetic Effect of Combined Leaf Extracts of Portulaca oleracea L., Beta vulgaris L., and Cichorium intybus L. in Streptozotocin-Induced Diabetic Rats. *Journal of Medicinal Food*, *27*(4), 339-347.

92. Han, Haewook & Segal, Adam & Seifter, Julian & Dwyer, Johanna. (2015). Nutritional Management of Kidney Stones (Nephrolithiasis). Clinical nutrition

research. 4. 137-52. 10.7762/cnr.2015.4.3.137.

93. https://ntbg.org/database/plants/detail

94. https://www.planetayurveda.com/library.

95. Sushrut Samhita S.S.Su.46/281,282

96. Hasan,N.,Osman,H.,Mohamad,S.,Chong,W.K.,Awang,K.,& Zahariluddin S.M. (2012). The Chemical Components of *Sesbania grandiflora* Root and their Antituberculosis Activity.Pharmaceuticals, 5(8), 882–889.

97. Aung, A.A. Chemical investigation and antimicrobial lactivities of Sesbania grandiflora L.UnivResJ, 2011;4(1),337-350.

98. Arfan, Nafisa & Julie, Azima & Mohiuddin, Abdul Kader & Khan, Shah. (2016). Medicinal Properties of the *Sesbania grandiflora* Leaves. Ibnosina J Med BS. Ibnosina J Med BS 2016. 271-277. 10.4103/1947-489X.210243.

99. Padmalochana, K., &Rajan, M. D. (2014). Antimicrobial activity of Aqueous, Ethanol and Acetone extracts of *Sesbania grandiflora* leaves and its phytochemical characterization. Int. J. of Pharma Sciences and Research, 2014; 5(12):957-962.

100. Mohiuddin, A. K. Medicinal and therapeutic values of *Sesbania grandiflora*. J Pharm Sci Exp Pharmacol, 2019, 81-86.

101. Ananta Worasakul, P., Hamamoto H., Sekimizu K., & Okonogi, S.(2017). Biological activities and antibacterial biomarker of Sesbania grandiflora bark extract.Drug Discoveries & Therapeutics,11(2),70–77.

102. Rateb, Mostafa E. *Sesbania grandiflora* L. Poir leaves: A dietary supplement to alleviate type 2 diabetes through metabolic enzymes inhibition. South African Journal of Botany,2020;130:282–299

103. Ninfali, P., & Angelino, D. (2013). Nutritional and functional potential of Beta vulgaris cicla and rubra.Fitoterapia,89,188–199

104. El-Sohaimy Sobhy, Eman Mohamed Abdo, Omayma El-Sayed Shaltout, Ahmed Elsaid Abdalla, & AhmedM. Zeitoun. (2020). Nutritional Evaluation of Beetroots (Beta vulgaris L.) and Its Potential Application in a Functional Beverage.Plants, 9(12), 1752–1752.https://doi.org/10.3390/plants 9121752

105. Rowe, R.C., Sheskey, P.J. and Quinn, M.E. (2009) Handbook of Pharmaceutical Excipients. 6th Edition, Pharmaceutical Press, 359,404,767.

106. Khadabadi SS, Deore SL. Experimental pharmacognosy, a comprehensive guide. Nirali Prakashan. 2013; 2: 1.3-1.4.

107. Khandelwal KR. Practical Pharmacognosy –Technique and Experiments. Nirali Prakashan. 2015; 25:1-25.

108. The Ayurvedic Pharmacopoeia of India. Part- I, 1st Edition, Vol-II, Government of India, Ministry of Health And Family Welfare Department Of Indian system of medicine and homoeopathy. 1989; 142-143.

109. Lohar DR. Protocol for Testing Ayurvedic, Siddha and Unani Medicine,Government of India, Department of AYUSH, Ministry Of Health And Family Welfare. Pharmacopoeia Laboratory for Indian Medicines Ghaziabad New Delhi.

110. Quality Control Methods for Medicinal Plant Materials. World Health Organization. Geneva WHO. 1998; 8-30.

111. Waterhouse, A. L. (2002). Determination of total phenolics. *Current protocols in food analytical chemistry*, *6*(1), I1-1.

112. Shirazi, O. U., Khattak, M. M. A. K., & Shukri, N. A. M. (2014). Determination of total phenolic, flavonoid content and free radical scavenging activities of common herbs and spices. *Journal of pharmacognosy and phytochemistry*, *3*(3), 104-108.

113. Harborne JB. Phytochemical Methods: A Guide to Modern Technique of Plant Analysis. Chapamn and Hall: London. 2007; 1(37)69:125-75

114. Thakker, V. Y., Shah, V. N., Shah, U. D., & Suthar, M. P. (2011). Simultaneous estimation of gallicacid, curcumin and quercetin by HPTLC method.*J Adv Pharm Educ Res*, *1*, 70-80.

115. Wagh, V.D., Wagh, K.V., Tandale, Y.N., & Salve,S.A.(2009). Phytochemical, pharmacological and phytopharmaceutics aspects of Sesbania grandiflora (Hadga): A review. Journal of Pharmacy Research, 2(5), 889-892.

116. Sethuraman, V., Janakiraman, K., Krishnaswami, V., Natesan, S., & Kandasamy,

R. (2021).Combinatorial analysis of quercetin and resveratrol by HPTLC in Sesbania grandiflora phyto-based nanoformulations. *Natural product research*, *35*(13), 2243-2248.

117. Jain, R., Hait, M., & Jain, S. K. (2022). Quantification of β-Sitosterol in Sesbania grandiflora Bark using High Performance Thin Layer Chromatography. *ES Food & Agroforestry*, *9*, 39-44.

118. Gauttam VK, Kalia AN. Development of polyherbal antidiabetic formulation encapsulated in the phospholipids vesicle system. J Adv Pharm Technol Res. 2013 Apr;4(2):108-17. doi: 10.4103/2231-4040.111527. PMID: 23833751; PMCID: PMC3696222.

119. Manik, S., Gauttam, V., & Kalia, A. N. (2013). Anti-diabetic and antihyperlipidemic effect of allopolyherbal formulation in OGTT and STZ-induced diabetic rat model.

120. S. Parasuraman,Toxicological screening,J Pharmacol Pharmacother, 2 (2011), pp. 74-79

121. ECD (2002), Test No. 423: Acute Oral toxicity - Acute Toxic Class Method, OECD Guidelines for the Testing of Chemicals, Section 4, OECD Publishing, Paris, https://doi.org/10.1787/9789264071001

122. Ghauri, A. O., Mohiuddin, E., Rehman, T., & Siddiqui, H. S. M. (2022). Acute and subacute toxicity studies of a poly herbal formulation used for diabetes. Pakistan Journal of medical sciences, 38(6), 1668–1673. https://doi.org/10.12669/pjms.38.6.5928

123. Majhi, S., Singh, L., Verma, M., Chauhan, I., & Sharma, M. (2022). In-vivo evaluation and formulation development of polyherbal extract in streptozotocin-induced diabetic rat. *Phytomedicine Plus*, *2*(4), 100337.

124. Chaudhuri, A., & Sharma, S. (2016). Evaluation of antidiabetic activity of polyherbal formulation in streptozotocin-induced diabetic rats. *Pharmaceutical and Biosciences Journal*, 01-06.

125. Kumar, C. H., Kumar, J. S., Ishaq, B. M., Rani, G. U., & Prkash, K. V. (2010). Antidiabetic activity of a polyherbal preparation. Pharmacologyonline, 2(1), 780-

87.

126. Petchi, R. R., Vijaya, C., & Parasuraman, S. (2014). Antidiabetic activity of polyherbal formulation in streptozotocin–nicotinamide induced diabetic Wistar rats. Journal of Traditional and Complementary medicine, 4(2), 108-117.

127. Arijit Chaudhuri & Shalini Sharma. (2016). Evaluation of Antidiabetic Activity of Polyherbal Formulation in Streptozotocin-Induced Diabetic Rats. Pharmaceutical and Biosciences Journal, 01-06. https://doi.org/10.20510/ukjpb/4/i5/113983

128. Ghanshyam Panigrahi, Chhayakanta Panda, Arjun Patra, & Extract of Sesbania grandiflora Ameliorates Hyperglycemia in High Fat Diet-Streptozotocin Induced Experimental Diabetes Mellitus & Scientifica, vol. 2016, Article ID 4083568, 10 pages, 2016. https://doi.org/10.1155/2016/4083568

129. Wroblewska,M.,Juskiewicz,J.,&Wiczkowski,W.(2011).Physiological properties of beetroot crisps applied in standard and dyslipidaemic diets of rats. Lipids in Health and Disease,10(1),178

130. Lorizola I, Furlan C, Portovedo M, Milanski M, Botelho P, Bezerra R, SumereB, Rostagno M, Capitani C. 2018. Beet stalks and leaves (Beta vulgaris L.) protect against high-fat diet-induced oxidative damage in the liver in mice. Nutrients 10 (7):872.

131. Rasica,L,Porcelli,S.,Marzorati,M.,Salvadego,D.,Vezzoli,A.,Agosti,F.,Grassi, B. (2018). Ergogenic effects of beetroot juice supplementation during severe-intensity exercise in obese adolescents. American Journal of Physiology Regulatory, Integrative and Comparative Physiology, 315(3), R453–R460.

132. El Gamal, A. A., AlSaid, M. S., Raish, M., Al-Sohaibani, M., Al-Massarani, S.M., Ahmad, A., Hefnawy, M., Al-Yahya, M., Basoudan, O. A., & Rafatullah, S. (2014). Beetroot (Beta vulgaris L.) extract ameliorates gentamicin-induced nephrotoxicity associated oxidative stress, inflammation, and apoptosis in rodent model.Mediators of inflammation, 2014, 983952.

133. Rateb, Mostafa E. *Sesbania grandiflora* L. Poir leaves: A dietary supplement to alleviate type 2 diabetes through metabolic enzymes inhibition. South African Journal of Botany,2020;130:282–299

134. Visvanathan, Rizliya; Khanfar, Mohammad A.; Qader, M. Mallique; Hassan,Marwa H.A.; Hassan, Hossam M.; Bawazeer, Majed; Behery, Fathy A.; Yaseen,Mohammed; Liyanage, Ruvini; Abdelmohsen, Usama R.; Rateb, Mostafa E. (2020). Sesbania grandiflora L. Poir leaves: A dietary supplement to alleviate type 2 diabetes through metabolic enzymes inhibition. South African Journal of Botany, 130, 282–299.

135. Thissera, B., Visvanathan, R., Khanfar, M. A., Qader, M. M., Hassan, M. H., Hassan, H. M., & Rateb, M. E. (2020). *Sesbania grandiflora* L. Poir leaves: A dietary supplement to alleviate type 2 diabetes through metabolic enzymes inhibition. *South African Journal of Botany*, *130*,282-299.

136. Beals, J.W., Binns, S.E., Davis, J.L., Giordano, G.R., Klochak, A.L., Paris,H. L., Bell, C. (2017). Concurrent beet juice and carbohydrate ingestion: Influence on glucose tolerance in obese and nonobese adults. Journal of Nutrition and Metabolism, 2017, 6436783.

137. Karole, S., Shrivastava, S., Thomas, S., Soni, B.,Khan, S., Dubey, J., Dubey, S.P., Khan, N., & Jain, D. K. (2019). Polyherbal Formulation Concept for Synergic Action: A Review. *Journal of Drug Delivery and Therapeutics*, *9*(1-s), 453-466.https://doi.org/10.22270/jddt.v9i1-s.2339

138. Anantaworasakul, P., Hamamoto, H., Sekimizu, K., & Okonogi, S. (2017). In vitro antibacterial activity and in vivo therapeutic effect of Sesbania grandiflora in bacterial infected silkworms. *Pharmaceutical biology*, *55*(1), 1256–1262. https://doi.org/10.1080/13880209.2017.1297467

139. Bolkent, Ş., Yanardağ, R., Tabakoğlu-Oğuz, A., &Özsoy-Saçan, Ö. (2000). Effects of chard (Beta vulgaris L. var. cicla) extract on pancreatic B cells instreptozotocin-diabetic rats: a morphological and biochemical study. Journal of ethnopharmacology, 73(1-2), 251-259.

140. Leon Lachman, Herbert A. Lieberman, Joseph L. Kanig: The hypothesis and Practice of Modern Drug store, Varghese distribution house, 3d release, 1990, 293-373

141. Mahapatra, S. K. &Verma, S. (2023).Formulation and Evaluation of Polyherbal

Tablet for Better Therapeutic Efficacy. *Research Journal of Pharmacy and Technology*, *16*(2), 835-838.

142. Gaikwad, B. B., Rane, B. R., & Jain, A. S. (2022). Formulation and Evaluation of Orodispersible Tablet of Sulindac. *European Journal of Pharmaceutical Research*, *2*(3), 11–19. https://doi.org/10.24018/ejpharma.2022.2.3.41

143. Sadaf Anwar, Mohd. Adnan Kausar, KehkashanParveen, Aqeela Zahra, Abrar Ali,Riadh Badraoui, Mejdi Snoussi, Waseem A. Siddiqui, Mohd Saeed, Polyherbal formulation: The studies towards identification of composition and their biological activities, Journal of King Saud University - Science, Volume 34, Issue 7,2022,102256,ISSN 1018-3647

144. Pradhan, A., Subba, M., Asif, M., & Sharma, C. (2022).Formulation and evaluation of Polyherbal tablet using Carica papaya, Emblica officinalis, Foeniculum vulgare. *Journal of Pharmacognosy and Phytochemistry*, *11*(5), 211-214.

145. Gunde, M., Sonule, R., & Suruse, P. (2022, November). Formulation and Evaluation of Polyherbal Antidiabetic Tablet. *2022 International Conference on Emerging Trends in Engineering and Medical Sciences (ICETEMS)* (pp.5-8). IEEE.

146. Paul,S.,Dey,T.,Koirala,P.,Tamang,S.,Bhattacharya,S.,&Das,R.(2023). Formulation and evaluation of Polyherbal tablet by using Neem, Tulsi, Turmeric and Ginger extract. *Journal of Drug Delivery and Therapeutics*, *13* (7), 46-51.

147. Umesh, A, Kumudhavalli, M.V., Kumar, M. & Venkateswarlu,B.S.Formulation And Evaluation Of Polyherbal Formulation Containing Indigenous Medicinal Plants.

148. Ray, A., Prasad, S., &Yadav, R.(2022).Formulation and Evaluation of Polyherbal Formulation of Aegle Marmelos & Pedalium Murex Extract for Anti-Diabetic Activity.*Journal of Pharmaceutical Negative Results*,7708-7720.

149. Mishra, R., Ray, A., Singh, A., Tripathy, S., Prasad, S., & Yadav, R. (2023). Development And Evaluation Of Anti Diabetic Activity In Polyherbal Tablets Of Local Herbs. *Journal of Pharmaceutical Negative Results*, 1418-1426.

150. ICH harmonized tripartite guideline. Stab Test New Drug Subst Prod. Q1A (R2). Geneva: International Conference of Harmonization; 2009.

151. Ferraro, P. M., Bargagli, M., Trinchieri, A., & Gambaro, G. (2020). Risk of Kidney Stones: Influence of Dietary Factors, Dietary Patterns, and Vegetarian-Vegan Diets. *Nutrients*, *12*(3), 779. https://doi.org/10.3390/nu12030779

152. https://www.mayoclinic.org/diseases-conditions/hyperoxaluria/symptoms-causes/syc-20352254.

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In-vivo Anti-diabetic activity of Polyherbal formulation on Streptozotocin Induced diabetic Wistar Rats

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Abstract

Objective: To investigate the *In-vivo* antidiabetic activity of a polyherbal mixture in streptozotocin-induced diabetic Wistar albino rats. **Methods:** Optimization of Polyherbal formulation combinations was done by testing for oral glucose tolerance test (OGTT) (2 h) in non-diabetic rats and Antidiabetic activity (28 days) in non-diabetic and STZ-induced diabetic Wistar albino rats. Five sets of Wistar albino rats (n = 6) were used in this investigation. Male Wistar rats were given intraperitoneal injections of streptozotocin to induce diabetes. Upon confirmation of diabetes, the animals were given oral treatments for 30 days, consisting of 200 or 400 mg/kg body weight of extracts or distilled water. **Results:** The result of investigation revealed that PHF2 significantly decreases blood glucose level as compared to PHF1 and PHF3 ,no crystals of Uric acid and Calcium oxalte crystals was observed and was further selected for antidiabetic in-vivo activity. The optimized Polyherbal formulation combination (PHF2) administered at a dosage of 400 mg/kg followed by 200 mg/kg shows significant antidiabetic activity. This investigation of the antidiabetic and biochemical effects of polyherbal formulation (PHF) was carried out on diabetic rats induced with streptozotocin (STZ). In the experimental animals, biochemical parameters such as hemoglobin, glycosylated hemoglobin, high-density lipoprotein, low-density lipoprotein, glucose, creatinine, serum cholesterol, serum triglyceride, and so on were also measured. It was concluded that PHF 2 had strong antihyperglycemic effects. Methanol extract of PHF 2 treatment brought the elevated biochemical parameters significantly ($P<0.05$) back to normal in diabetic rats.

Keywords: Diabetes, Biochemical and Hematological parameters Polyherbal formulation, Streptozotocin , Wistar Albino Rats.

Introduction

Globally, the number of people with diabetes has more than doubled during the last 20 years. One of the most concerning aspects associated with this sharp rise is the rise in type 2 diabetes in children, adolescents, and young adults. [1] Uncontrolled diabetes can lead to problems in many different organs. Severe macro vascular complications like heart attacks, strokes, kidney failure, damage to small and large blood vessels, and nerve injury are the most alarming trends[2,3]. Insulin and a number of oral hypoglycemic drugs, including biguanides and sulfonylureas, are currently the available treatments for diabetes mellitus. Although they have some disadvantages, such as side effects and high rates of secondary failure, these drugs are used to treat diabetes mellitus. To meet this need, the diverse traditional plant kingdom offers many promising therapeutic uses. A plethora of natural remedies have been recommended to treat diabetes [4]. The World Health Organization (WHO) defines a medicinal plant as one that "contains substances in one or more of its organs that can be used for therapeutic purposes, or which are precursors for chemo-pharmaceutical semi-synthesis." The idea of polyhedral formulation is well documented in ancient literature. Compared to a single plant, the polyherbal formulation has a larger and more comprehensive medicinal potential. The goal of the current study was to evaluate the therapeutic benefits of a plant known to have antidiabetic action in order to create and standardize a polyherbal formulation. [5]

The medicine formulation in Ayurveda is based on two principles:

1. Several herbs are combined to create a single product in polyherbal formulations (PHF). It blends a variety of herbs to achieve therapeutic efficacy.

2. PHF has a synergistic, broad therapeutic index, making it safe at high doses while still being effective at low doses (better risk to benefit ratio) than allopathic hypoglycemic medications, which have a narrow therapeutic range. This treatments are ideal due to their efficacy, safety, affordability, acceptability, and accessibility.[6] Prior studies have demonstrated the presence of antioxidant and antiurolithiatic properties in *S. grandiflora*, along with chemopreventive and anticancer properties, anxiolytic and anticonvulsant effects, hepatoprotective properties, cardioprotective properties, antiulcer properties, antimicrobial properties, analgesic and antipyretic properties, diuretic properties, CNS depressant and laxative hypolipidemic properties, and anthelmintic properties. After a careful analysis of the literature, it was found that there hasn't been much research done on the leaves' potential to prevent diabetes [7-8]. While a number

of pharmacological effects, such as anti-inflammatory, antioxidant, neuroprotective, hyperglycemic, and anticancer, have been demonstrated for the genus *Beta vulgaris* L. Additionally, earlier studies have demonstrated the anticancer *activity of Beta vulgaris* L. against tumor cells, particularly breast cancer cells. Many illnesses, such as leukemia, esophageal cancer, glandular cancer, prostate cancer, and breast cancer, are treated with it in traditional medicine.[9-12]

Material and Method

Collection of Plants

The fresh leaves of *Sesbania Grandiflora* and root of *Beta Vulgaris* was collected from local area of Ale, Junnar ,Pune ,Maharashtra .Taxonomically leaves of Sesbania *Grandiflora* and root of *Beta Vulgaris* was identified and authenticated by Dr.R.K Chaudhary,Scientist, Agharkar Research Institute, Autonomous Body under DST, GOI, Pune. Herbarium specimens have been preserved in the laboratory.

Extraction and Lyophilization

The collected plant material were shade dried, coarsely powdered using mixer grinder and passed through 100 number sieve and stored in an airtight container.100 grams of each powder were extracted with methanol by Soxhlet Extraction procedure till complete powder color disappears .The extract were concentrated under vacuum using rotary evaporator at 40℃.The concentrated extract was freeze dried at -20℃ for 12h then lyophilized using lyophilizer .These lyophilized extract was stored in air tight container and kept in desiccators for further study.

Chemicals: Streptozotocin was procured from Sigma Chemical Laboratories, Shree Chemicals, Pune. Glibenclamide Tablet (5mg) was purchased from Aventis Pharma, Citrate Buffer, Glucose was purchased from Scientific Chemicals, Mumbai.

Animals

Adult male Wistar rats (180-250 g) were procured from Lachmi Biofarm Pvt.Ltd,,Pune ,Maharashtra India. The animals were housed in large, spacious polyacrylic cages at an ambient room temperature with 12-h light/12-h dark cycle. Rats had free access to water and rodent pellets diet (Nutrivate Pvt. Ltd, Bangalore, India). The study was approved by the Institute

Animal Ethics Committee of the Vishal Institute of Pharmaceutical Education and Research Ale with Reg. No. 1409/PO/RE/S/11/IAEC/2020-2021/07/01 were used for the study and all the animal exper*iments were carried out according to the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA) guidelines, Ministry of Environment and Forests, Government of India.

Development of Polyherbal formulation [13,14]

The lyophilized powder of methanolic extract was evaluated for antihyperglycemic potential using the OGTT model in Wistar rats for a single dose of 1000 mg/Kg. As a result, by altering the ratios, several extract combinations were developed for the formulation design .The three distinct batches of polyherbal formulation, as listed in Table No. 1 below, contain methanolic extracts of leaves of *Sesbania Grandiflora* and root of *Beta Vulgaris* in varying ratios. According to WHO guidelines for herbal medicine quality control, batches were tested for quality. The batch optimized after OGTT was chosen for further *in vivo* activity on diabetes.

Table no:1 Polyherbal Formulation design

Formulation	Drug combination	Ratio
PHF 1	MEBG+MESV	2:1
PHF2	MEBG+MESV	1:1
PHF3	MEBG+MESV	1:2

PHF: Polyherbal Formulation, MESG: Methanolic extract of *Sesbania Grandiflora* leaves, MEBV- Methanolic extract of *Beta Vulgaris* Root.

Optimization of the formulation as per Oral Glucose Tolerance Test Model

The OGTT study was conducted on overnight fasted glucose (2g/kg)induced hypoglycemic normal rats. The rats were divided into five groups (n=6)

Group I-Normal Group-0.5 % w/v carboxymethylcellulose (CMC) solution pretreated rats.

Group II-Glucose Load- Glibenclamide 5mg/kg

Group III-Single dose levels (1000 mg/kg per oral) serving as bPHF1-Animal treated with combination of MESG and MEBV with ratio 2:1

Group IV-Single dose levels (1000 mg/kg per oral) serving as PHF2-Animal treated with combination of equal amount of extracts of MESG and MEBV i.e with ratio 1:1

Group V-Single dose levels (1000 mg/kg per oral) serving as PHF2-Animal treated with combination of equal amount of extracts of MESG and MEBV i.e with ratio 1:2

The blood sample was withdrawn from the tail vein before and 0,30,60,90,120 min after glucose administration .The serum glucose level was estimated within 30 mins of withdrawal of the blood sample. Also the urine samples collected was microscopically examined for presence of crystals of Uric Acid and Calcium Oxalate.

***In-vivo* Antidiabetic Effect of Polyherbal Formulation in Streptozotocin Induced Diabetic Rats[13-17]**

Administration of Glibenclamide (GLB) and Streptozotocin (STZ)

A single intraperitoneal (i.p.) dose of freshly prepared Streptozotocin (STZ) 45 mg/kg in 0.1 M citrate buffer (pH 4.5) was given to overnight-fasted Wistar Albino rats to induce diabetes. In order to prevent hypoglycemia-related death, the rats were given 5 % w/v glucose solution and given access to a standard diet after receiving STZ for 24 hours. The animals treated with STZ were found to have diabetes when their fasting blood glucose levels were measured 48 hours after induction. The standard dosage of Glibenclamide was given orally once a day for 30 days in a suspension of 0.5% w/w distilled water. **[13-15]**

Administration of Polyherbal Formulation

PHF2 extract was suspended in 5 ml of sterile water and administered orally for 30 days, while the control group received water as a vehicle. After 4 hours of Polyherbal formulation administration, the rats were allowed free access to food (standard rodent pellet).

Experimental Design

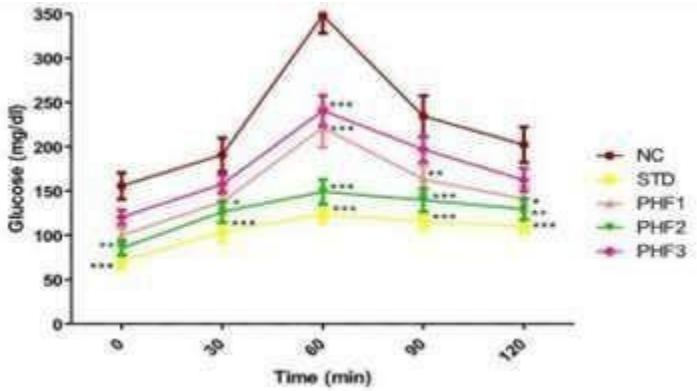
After 48 hours of induction, fasting blood glucose levels were measured to confirm the presence of diabetes in the STZ-treated animals. Wistar albino rats were randomly assigned to Group II– Group V after their blood glucose levels exceeded 200 mg/dl, which was considered the threshold for diabetes

Table no: 2 Experimental Design of Antidiabetic Polyherbal Formulation

Group	Codes	Route and Dose of drug
Group I	Normal control(NC)	Orally with vehicle (1 ml/kg BW)
Group II	Diabetic Control(DC)	Orally with STZ (45mg/kg BW)
Group III	Test solution (F 200)	Orally with vehicle (200 mg/kg BW)
Group IV	Test solution (F 400)	Orally with vehicle (400 mg/kg BW)
Group V	Standard control(STD)	Orally with Glibenclamide (5 mg/kg BW)

Diabetes was produced in overnight starved rats with a single intraperitoneal (i.p.) injection of freshly prepared Streptozotocin (STZ) 45 mg/kg b.w., in 0.1 M citrate buffer (pH 4.5) in a volume of 0.5 ml/kg b.wt. Diabetes was confirmed in STZ rats after 48 hours of induction by assessing fasting blood glucose levels. To prevent hypoglycemia mortality, the rats were administered 5% w/v glucose solution (2 ml/kg b.w.) after STZ injection. Diabetic rats had fasting blood glucose levels of greater than 200 mg/dl and were randomly assigned to one of four groups. The standard (Glibenclamide) and Polyherbal formulation were suspended in 1% w/v carboxymethyl cellulose (CMC) and given orally once daily for 21 days. Blood samples were taken by pricking the tail vein of rats on the first, seventh, fourteenth, and twenty-first days of therapy and were immediately utilized to estimate blood glucose with a Glucometer. All of the experimental animals' weekly body weight fluctuations were tracked.[16,17,18] At the conclusion of the examination, blood was collected from all of the experimental animals through retro-orbital plexus puncture for further biochemical studies.

Result



Effect of PHF 1, PHF 2, PHF 3 extract on blood glucose level (mg/dl) in experimental group of rats receiving an oral glucose load. Values are Expressed As Mean ± SEM (n=6) analyzed by two-way Anova***represent significance At $p < 0.001$

***Invivo* Antidiabetic Effect of Polyherbal Formulation on Biochemical parameters in Streptozotocin Induced Diabetic Rats**

Biochemical Parameters

Blood Glucose (mg/dl)

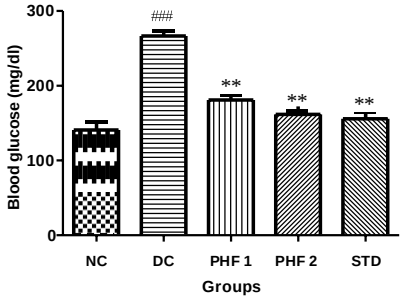


Figure 1: Effect of PHF 200 and 400 on Blood glucose level (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increase in blood glucose level when compared with DC rats. However, the treatment of rats with PHF (200 and

400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.01$; $p<0.001$) decrease in blood glucose level when compared with DC rats.

Serum Creatinine (mg/dl)

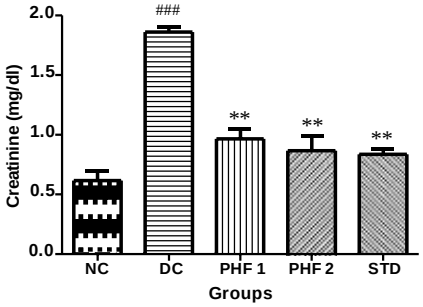


Figure 2 : Effect of PHF 200 and 400 on creatinine level (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increase in creatinine level when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.001$) decrease in creatinine level when compared with DC rats.

Serum Protein (g/dl)

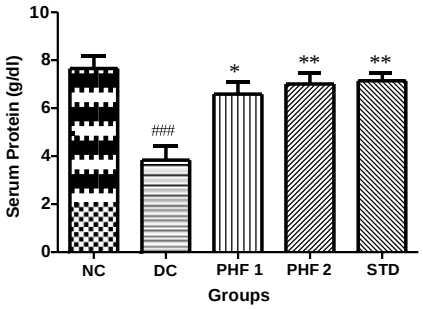


Figure 3: Effect of PHF 200 and 400 on serum proteins level (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) decrement in protein levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.01$; $p<0.001$) increment in protein levels when compared with DC rats.

Alanine transaminase (IU/L)

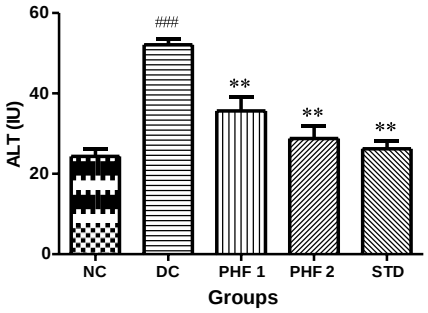


Figure 4: Effect of PHF 200 and 400 on Alanine transaminase level (IU/L) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in alanine transaminase levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.001$) decrement in alanine transaminase levels when compared with DC rats.

Aspartate aminotransferase (IU/L)

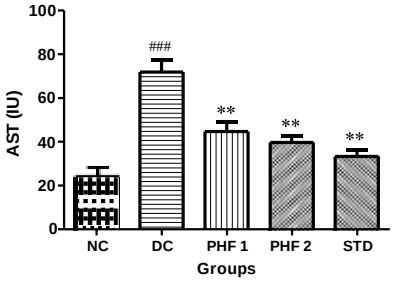


Figure 5: Effect of PHF 200 and 400 on Aspartate transaminase level (IU/L) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in aspartate transaminase levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.001$) decrement in aspartate transaminase levels when compared with DC rats.

Blood Urea Nitrogen (mg/dl)

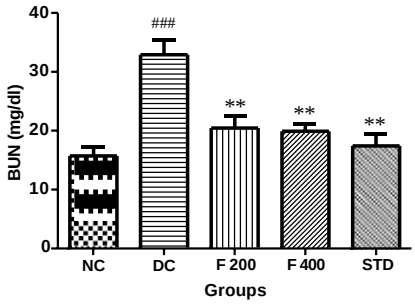


Figure 6: Effect of PHF 200 and 400 on BUN (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in BUN levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.001$) decrement in BUN levels when compared with DC rats.

Total Cholesterol (mg/dl)

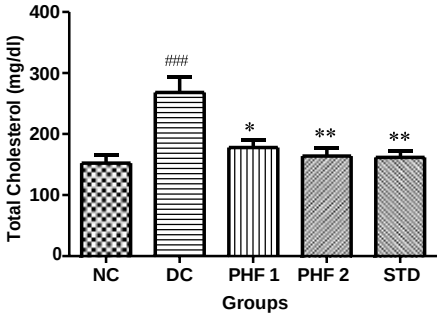


Figure 7

Figure 7: Effect of PHF 200 and 400 on Total Cholesterol (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The effects of PHF 200 and 400 on total cholesterol (mg/dl) in STZ induced diabetes in rats are shown in Figure 7. The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in total cholesterol levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.01$; $p<0.001$) decrease in total cholesterol levels when compared with DC rats.

Triglycerides (mg/dl)

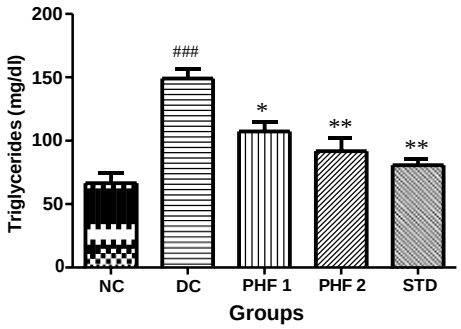


Figure 8

Figure 8: Effect of PHF 200 and 400 on Triglycerides (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in triglycerides levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.01$; $p<0.001$) decrement in triglycerides levels when compared with DC rats.

HDL Cholesterol (mg/dl)

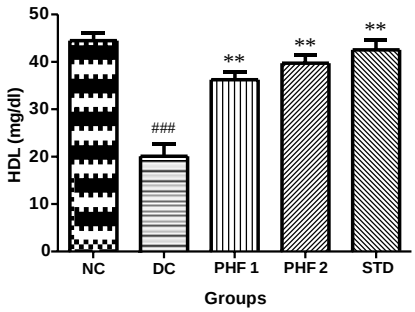


Figure 9: Effect of PHF 200 and 400 on High Density Lipoprotein (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant (p<0.001) decrement in HDL levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant (p<0.001) increment in HDL levels when compared with DC rats.

LDL Cholesterol (mg/dl)

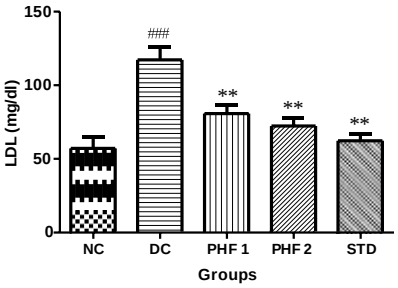


Figure 10: Effect of PHF 200 and 400 on Low Density Lipoprotein (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant (p<0.001) increment in LDL levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant (p<0.001) decrement in LDL levels when compared with DC rats.

VLDL Cholesterol (mg/dl)

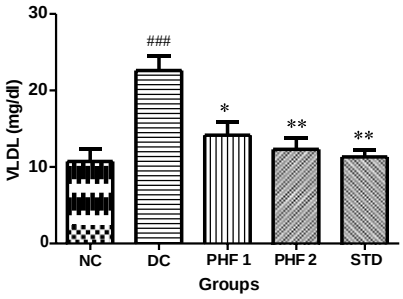


Figure 11

Figure 11: Effect of PHF 200 and 400 on Very Low Density Lipoprotein (mg/dl) in STZ induced Diabetes in rats. Values are expressed as Mean ± SEM (n = 6) & analyzed by One-way ANOVA followed by Tukey's Kramer test. $^{###}p<0.001$ versus NC rats and $^{***}p<0.001$ versus DC rats.

The treatment of rats with STZ (45 mg/kg, i.p.) induced significant ($p<0.001$) increment in VLDL levels when compared with DC rats. However, the treatment of rats with PHF (200 and 400 mg/kg, p.o.) and Glibenclamide (5 mg/kg, p.o.) exhibited significant ($p<0.01$; $p<0.001$) decrement in VLDL levels when compared with DC rats.

Conclusion

When compared to normal control, the results showed a progressive loss of body weight in diabetic control. This could be the result of an excessive breakdown of fatty acids and tissue proteins brought on by a drop in plasma insulin levels. A lack of insulin can slow down the synthesis of proteins and quicken the breakdown of metabolites, raising blood levels of amino acids that are used in the process of gluconeogenesis. Body weight increased following administration of PHF 400 mg/kg of the extract compared to Group 2. .. , The treatment of rats with PHF2 (200 and 400 mg/kg, p.o.) and Glibenclamide exhibited significant ($p<0.001$) changes in Biochemical parameters such as hemoglobin, glycosylated hemoglobin, high-density lipoprotein, low-density lipoprotein, glucose, urea, creatinine, serum cholesterol, serum triglyceride, and it was discovered that PHF's methanol extracts had strong antihyperglycemic effects. We appeal to the conclusion that the plant fraction and extract that were tested for their antidiabetic properties significantly reduced serum glucose levels and other diabetes-related

complications. The results of this study lend support to the use of this plant in conventional antidiabetic preparations; formulations based on the plant's fraction and identified effective extract may be more effective than those currently on the market that use crude aqueous extract.

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The author is thankful to Dr.Rajesh Khatjuriya, for giving me proper guidance .I also sincerely thanks to College management and Dr.Suresh L Jadhav for providing technical facilities and assistance required for this work.

Conflict of interest

No conflict of interest in the present study

References

1. Roglic, G. (2016). WHO Global report on diabetes: A summary. *International Journal of Noncommunicable Diseases*, *1*(1), 3-8.
2. Cole, J. B., & Florez, J. C. (2020). Genetics of diabetes mellitus and diabetes complications. *Nature reviews nephrology*, *16*(7), 377-390.
3. Zimmet, P. Z., Magliano, D. J., Herman, W. H., & Shaw, J. E. (2014). Diabetes: a 21st century challenge. *The lancet Diabetes & endocrinology*, *2*(1), 56-64.
4. Eidi A, Eidi M, Esmaeili E. Antidiabetic effect of garlic (Allium sativum L.) in normal and streptozotocin-induced diabetic rats. Phytomedicine 2006;13(9-10):624-9
5. Petchi, R. R., Vijaya, C., & Parasuraman, S. (2014). Antidiabetic activity of polyherbal formulation in streptozotocin - nicotinamide induced diabetic wistar rats. *Journal of traditional and complementary medicine*, *4*(2), 108–117. https://doi.org/10.4103/2225-4110.126174
6. Karthikeyan P, Suresh V, Suresh A, Aldrin bright J, Senthil S, Arunachalam G. 2010. Wound healing activity of Sesbania grandiflora (L.) Poir. Bark. International Journal of Pharmacy Research and Development, 3(2): 87-93
7. Sagarika Majhi, Lubhan Singh, Madhu Verma, Iti Chauhan, Raj kumari, Meenakshi Sharma,In-vivo evaluation and formulation development of polyherbal extract in streptozotocin-induced diabetic rat,Phytomedicine Plus,Volume 2, Issue 4, 2022,100337,ISSN 2667-0313, https://doi.org/10.1016/j.phyplu.2022.100337.
8. Karthikeyan P, Suresh V, Arunachalam G. 2011. In vitro anthelmintic activity of (L.) *Sesbania grandiflora poir* bark. International Journal of Pharmacy and Technology, 3 (1): 1548-1553.
9. Kazimierczak, R.; Hallmann, E.; Lipowski, J.; Drela, N.; Kowalik, A.; Püssa, T.; Matt, D.; Luik, A.; Gozdowski, D.; Rembiałkowska, E. Beetroot (*Beta vulgaris* L.) and naturally fermented beetroot juices from organic and conventional production: Metabolomics, antioxidant levels and anticancer activity. *J. Sci. Food Agric.* **2014**, *94*, 2618–2629.
10. Nade, V.S.; Kawale, L.A.; Zambre, S.S.; Kapure, A.B. Neuroprotective potential of *Beta vulgaris* L. in Parkinson's disease. *Ind. J. Pharmacol*. **2015**, *47*, 403.

11. Oztay, F.; Sacan, O.; Kayalar, O.; Bolkent, S.; Ipci, Y.; Kabasakal, L.; Sener, G.; Yanardag, R. Chard (*Beta vulgaris* var. *cicla*) extract improved hyperglycemia-induced oxidative stress and surfactant-associated protein alterations in rat lungs. *Pharm. Biol.* **2015**, *53*, 1639–1646.
12. Citores, L.; Iglesias, R.; Gay, C.; Ferreras, J.M. Antifungal activity of the ribosome-inactivating protein BE 27 from sugar beet (*Beta vulgaris* L.) against the green mould *Penicillium digitatum*. *Mol. Plant Pathol.* **2016**, *17*, 261–271.
13. Gauttam, V. K., & Kalia, A. N. (2013). Development of polyherbal antidiabetic formulation encapsulated in the phospholipids vesicle system. *Journal of advanced pharmaceutical technology & research*, *4*(2), 108–117. https://doi.org/10.4103/2231-4040.111527
14. Baron AD. Postprandial hyperglycaemia and α-glucosidase inhibitors. *Diab Res Clin Pract.* 1998;40: S51–5.
15. S. Parasuraman,Toxicological screening,J Pharmacol Pharmacother, 2 (2011), pp. 74-79
16. Al-Harbi, L. N., Alshammari, G. M., Al-Dossari, A. M., Subash-Babu, P., Binobead, M. A.,Alhussain, M. H., ... & Shamlan, G. (2021). Beta vulgaris L.(beetroot) methanolic extract prevents hepatic steatosis and liver damage in T2DM rats by hypoglycemic, insulin-sensitizing, antioxidant effects, and upregulation of PPARα. Biology, 10(12), 1306.
17. Qian, K., Zhong, S., Xie, K., Yu, D., Yang, R. and Gong, D. W. (2015). Hepatic ALT isoenzymes are elevated in gluconeogenic conditions including diabetes and suppressed by insulin at the protein level. Diabetes/Metabolism Research and Reviews, 31(6): 562 – 571
18. Annadurai T, Muralidharan AR, Joseph T, Hsu MJ, Thomas PA, Geraldine P. Antihyperglycemic and antioxidant effects of a flavanone, naringenin, in streptozotocin-nicotinamide-induced experimental diabetic rats. *J Physiol Biochem.* 2012;68:307–18.
19. Aladodo, R. A., Muhammad, N. O., & Balogun, E. A. (2013). Effects of aqueous root extract of Jatropha curcas on hyperglycaemic and haematological indices in alloxan-induced diabetic rats. *Fountain Journal of natural and applied sciences*, *2*(1).

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INTERNATIONAL JOURNAL OF CREATIVE RESEARCH THOUGHTS (IJCRT)

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QUALITATIVE ANALYSIS OF ROOT OF *BETA VULGARIS L.*

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ABSTRACT:

A significant member of the Chenopodiaceous family is the therapeutic herb Beta vulgaris Linn. It is often referred to as garden beet or beet root. In Indian traditional medical practices, the root is primarily used to treat issues with the reproductive system, high blood pressure, cancer, and the urinary tract. The standardization of the herb's roots was deemed to be important due to the herb's widespread use in Indian traditional medical systems. The morphological and microscopic characteristics, physical-chemical parameters, such as plant extractive with different solvents, ash levels, foreign organic matter, loss on drying, and Ph of aqueous solution, were used as quality control measures. The present study deals with the phytochemical investigation on root of *Beta Vulgaris L* for presence of saponins, tannins, terpenoids, flavonoids, polyphenols, steroids etc. Extraction is performed using the Soxhelt extraction apparatus, as well as macro and microscopical parameters are studied. Various root sections were taken to investigate and photograph the anatomical properties. Physiochemical investigation revealed loss on drying (11.74±0.32 %), total ash (10.84±0.43), water soluble ash (1.70± 0.24), acid insoluble ash (0.21± 0.31), alcohol soluble extractive (11.21± 0.18), water soluble extractive (21.31± 0.35). The HPTLC technique was used for qualitative determination of components from methanolic extracts of root solvent system Toluene; Ethyl Acetate; Formic acid Volume 5:4:1 that revealed the Rf values for terpenoids, flavonoids, gallic acid, quercitine. The HPTLC technique of alcoholic extract showed the presence of six and seven spots at 254 nm and 366 nm. The study done will provide relevant data used for proper identification and authentication of used herbal plant.

KEYWORDS: *Beta Vulgaris L,* Ayurveda, Pharmacognostic, Physicochemical Evaluation, HPTLC.

I. INTRODUCTION:

Beetroot, often known as red beet, is the taproot of the beet plant (Beta vulgaris L.). In many countries around the world, including India, beetroot is a widely consumed vegetable. High concentrations of bioactive compounds in beetroot, such as betalains and inorganic nitrate, may have a variability of positive health effects.[1] Because it is a perishable vegetable, it can be dehydrated, and its mineral content can rise due to the loss of water mass. And it is heavy in both fiber and sugars. The colour of food can play an important part in lavor perception. Food colouring is classified as follows four classifications [2]

A. Natural Colour

B. Nature-Identical Inorganic Colour

C. Mayank is A Synthetic Colour

Betalains is a kind of pigments that are commonly used as natural food colorants used in several culinary industries. Beetroot is high in nutrients such as vitamins (B complex and C), minerals, fibre, proteins, and a diversity of bioactive phenolic substances, primarily betalains, and other antioxidant-rich components such as coumarins, carotenoids, sesquiterpenoids, triterpenes, and flavonoids. Beetroot has been employed as an ingredient or preservative in food processing due to its firmness, nontoxic, noncarcinogenic, and nonpoisonous properties. Beetroot and its biochemical components have been shown to have antioxidant, anti- inflammatory, antiapoptotic, antibacterial, antiviral, and other properties. [3]

Figure 1: Beet Plant

I. MATERIALS AND METHODS:

A. Collection of Plant Material:

The material was purchased from market of Ale, Junnar. The material was Identified and Authenticated from Botanist Dr Ranangdale Savita Sanjaykumar, M.Sc. Ph.D., FIAAT, FAA BSc,Department of Botany, Balasaheb Jadhav College of Art,Commerce & Science , Ale Junnar, Pune University Maharashtra with Herbarium collection number 619-*Beta Vulgaris*.Also authenticated by Dr.R.K Chaudhary,Scientist, Agharkar Research Institute, Autonomous Body under DST, GOI, Pune. Herbarium specimen has been preserved in laboratory voucher specimen specimen no.23-94 For Beta Vulgaris. [4]

B. Method:

- **Morphological and Microscopical evaluation**

  The root of *B.Vulgaris L* was examined for various organoleptic properties. These studies include parameters such as taste, odour, shape, margin, venation, size, surface and apex. The microscopically study of *B.Vulgaris L* was done with the help of Swift Ives camera lucida microscope. The air-dried plant material was then, pulverized into a coarse powder and used for research work. [5]

- **Determination of Physicochemical constants**:

  Physicochemical constants of B.Vulgaris L root were determined water soluble ash, total ash, acid insoluble ash, alcohol soluble extractive value and water soluble extractive value as per the method described in Pharmacopoeias. [6]

- **Ultraviolet screening of leaf powder of *Beta Vulgaris L***

  The fluorescence study of powder with different reagents under ordinary day light and UV light (long & short) shows distinct characteristics fluorescence. [6]

III. PREPARATION OF EXTRACTS:

Beta Vulgaris L root were cleaned under running water and dried in the shade for seven days. Dried root were mechanically crushed to a coarse powder, sieved, and stored at room temperature in an airtight container. The extraction method was chosen based on the presence of active ingredients in the medicine. By using the soxhlet extraction method, dried powder (500 g) was extracted with Acetone, ethanol, methanol and distilled water. The extracts were dried by distilling the solvent at low temperatures with a rotary evaporator. The extracts were kept in a refrigerator at 4^0 C.

A. Phytochemical Screening:

The Phytochemical screening of the extracts were assessed to detect the presence of different phytoconstituents such as alkaloids, flavanoids, saponins, triterpenoids, steroids, carbohydrate, tannin, coumarins, phenols, carboxylic acid, amino acid and proteins by performing chemical tests.[9,10]

B. HPTLC analysis of extract [11-16]

HPTLC analysis of methanolic extract of root of *Beta Vulgaris L* was done by lane analysis. HPTLC analysis was done to access presence of components.

C. TLC instrumentation and conditions

- **Sample Preparation**: Sample Dissolved in methanol & incubated over night for 24 hrs to 48 hrs followed by concentrating the sample by Rotary evaporator method.
- **Sample loading:** About 5 µl of extract of Beta Vulgaris L is diluted with methanol and standard solution of Quercetin and Gallic acid were loaded as 6.0 mm 60F 254 TLC plate with use of

Hamilton Syringe.

- **Scanning:** TLC developed was dried to evaporate solvent and then placed in photo documentation chamber and images were captured at 254nm and 366nm.
- **Band Size** : 5mm
- **Analysis Type :** Lane Analysis
- **Seperation Technique**: Ascending
- **Test :** Methanolic extract of Beta Vulgaris L
- **Standards:** Quercetin, Gallic acid
- **Mobile Phase**: Toluene; Ethyl Acetate; Formic Acid (5:4:1)

IV. RESULTS AND DISCUSSION MICROSCOPIC CHARACTERS:

The transverse section of Beta Vulgaris L in Figure 1(a, b, c, d, e, f) displays deep red-colored cell vacuoles of the cortical parenchyma and the pith. The conductive vessel and rhizodermis are colourless. Starch is absent from the roots of sugar beetroot and red beetroot. A secondary structure of roots is present, comprising a concentric circle of conductive tissues that are created from cells with cellulose walls and are penetrated by large rays of parenchyma. The cross section of the beetroot revealed a well-represented xylem vessel, with the root centre (to pith) showing evidence of proto-xylem and outward association with meta-xylem. Under the phloemis cambium, supplied by the pericycle, pith, or pith rays. This cambium, which produces more secondary xylem than secondary phloem, is responsible for the majority of the root thickness. Beet vascular bundles are collateral type, the phloem is located in the back. Between two primary tissues persists meristematic tissues namely cambium from which secondary xylem and phloem are forming.

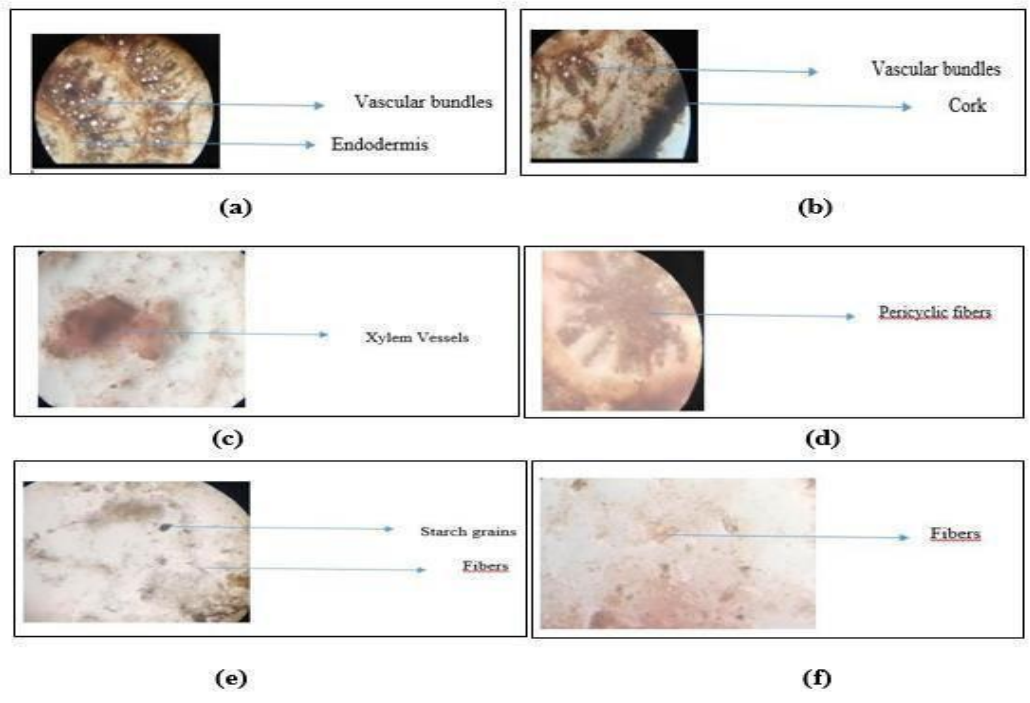

Figure 2: Microscopic Evaluation of *Beta Vulgaris L* Root

Table 1: Description of *Beta Vulgaris root*

Plant	An erec, sprawling perennial plant up to 60 cm (2 ft) high
Root	Dark green, leathery, shiny rosette root with wavy & rough triangular lower root and narrow and oval upper root. grow 20–40 cm (7.9–15.7 in) in length
Fruit	enclosed by the leathery and incurved perianth, and is immersed in the swollen, hardened perianth base
Root	swollen and fleshy long main red root
Flowers	green and tiny with the sepals thickening and hardening reach 1–2 m (3.3–6.6 ft) in height
Seed	The horizontal seed is lenticular, 2–3 mm, with a red-brown, shiny seed coat. The seed contains an annular embryo and copious perisperm

a. Ultraviolet screening of *Beta Vulgaris L* root powder

The fluorescence study of powder with different reagents under ordinary day light and UV light shows distinct characteristic fluorescence

Table 2: UV fluorescence studies of root powder of *Beta Vulgaris L*

Powder + reagent	Ordinary light	UV short wave (254nm)	UV long wave (365nm)
Only Powder	Dark red	Green	Dark Green
Powder + 5 % NaOH	Green	Dark green	Yellowish Brown
Powder +Chloroform	Yellow	Red	Green
Powder +1% KOH	Reddish yellow	Blue	Dark blue
Powder +conc. HNO3	Pale yellow	Green	Black
Powder +H2SO4	Brown	Dark brown	Brown
Powder +conc.HCl	Purple	Blue	Blue
Powder + Acetone	Light green	Red	Light green
Powder + FeCl3	Black	Blue	Blue
Powder + H2SO4	Light brown	Brown	Brown

b. Physical Analysis of Root powder of *Beta Vulgaris*

Various physical analyses are done for various parameters like foreign organic matter, loss on drying, ash content, extractive values.

Table 3: Physical Analysis of Root powder of Beta Vulgaris

Sr no	Parameters	Results
1	Foreign Organic matter	0.74 % (w/w)
2	LOD 110^0C	11.74 % (w/w)
3	Total Ash Content	10.84 % (w/w)
4	Water soluble ash	1.70 % (w/w)
5	Acid insoluble Ash	0.21 % (w/w)
6	Ethanol extractive value	11.21% (w/w)
7	Aqueous extractive value	21.31% (w/w)
8	pH	7.1

c. Extraction of root of *Beta Vulgaris L*

The % extraction yield of in aqueous, Ethanol, Acetone and Methanol are 23.61% w/w, 21.49 % w/w, 9.7% w/w and 25.37% w/w respectively. Figure 2 indicates that % w/w yield of root extract is higher in methanolic extract.

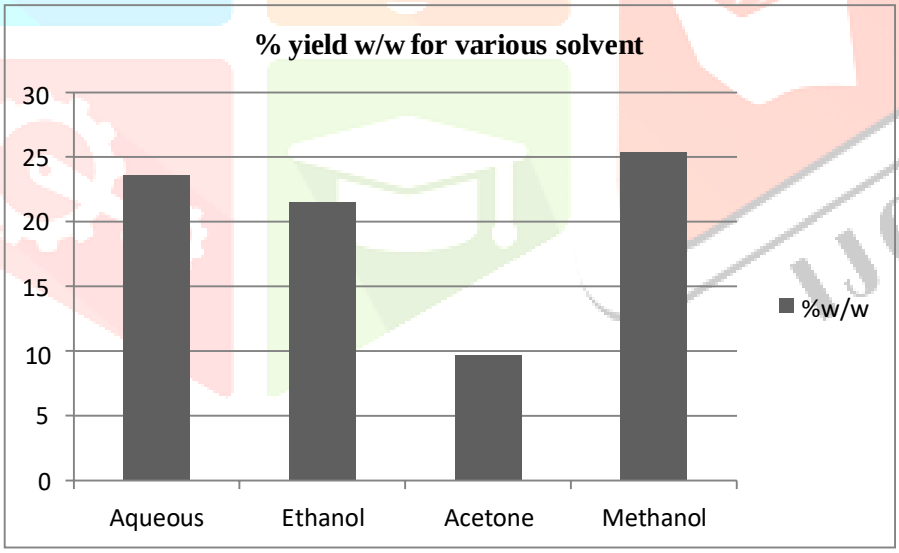

Figure 3: Extraction yield percentage in different solvents

d. Phytochemical Screening of *Beta Vulgaris L.*

Preliminary phytochemical screening of various extract of *Beta Vulgaris* linn root revealed phytoconstituents i.e. Alkaloids, Glycosides, Saponins, Phytosterols ,Tannins, Flavonoids, Carbohydrates, Proteins & amino acids, Fixed oils & fats, Gums and Mucilage, Volatile oil. Methanolic extracts shows the presence of most numbers of components Methanolic root extract has shown that it has extracted most of the compounds. Table clearly indicates clearly that methanol can be used as a principle extracting solvent and also evaporation of methanol is easy phytochemical constituents detected in crude extracts of root of *Beta Vulgari L*

Table 4: Phytochemical screening of *Beta Vulgaris* root extrac

Sr. No.	Tests	Aqueous	Ethanol	Methanol	Acetone
1	**Tests for Alkaloids**				
	Mayer's test	-	+	+	-
	Wagner's test	-	+	+	-
	Hager's tests	-	+	+	-
	Dragendorff's test	-	+	+	-
2	**Tests for Carbohydrates**				
	Molish test	+	+	+	-
	Fehling test	+	+	+	-
	Barfoed's test	+	+	+	-
	Benedict's test		+	+	-
3.	**Tests for Glycosides**				
	Borntrager's test	+	+	+	+
	Legal's test	+	+	+	+
4.	**Test for Saponins**				
	Test solution+20ml distilled H20	+	+		+
5.	**Tests for Proteins & amino acids**				
	Millon's test		-		
	Biuret test		-		
	Ninhydrin test		-		
6.	**Test for Phytosterol**				
	Libermann-Burchard's test	+		+	
7.	**Tests for Fixed oils & fats**				
	Spot test				
	Saponification test				
8.	**Tests for Tannins**				
	Ferric chloride test	+		+	+
	Gelatin test	+		+	+
	Aqueous bromine test	+		+	+
9.	**Tests for Flavonoids**				
	Lead acetate	+	+	+	+
	Alkaline reagent test	+	+		
10.	**Test for Gums &Mucilages**				
	Ext. + dis. H_2O +abs. alc. + stirring				
11.	**Test for Volatile oil**				
	50 gm. of powder subjected to hydro-distillation	-	-	-	-

Various tests were performed to find phytochemical constituents present in extracted material of different solvents. Various tests have been performed to find out the phytochemical constituents. Methanolic extracts shows the presence of most numbers of components.

Methanolic root extract has shown that it has extracted most of the compounds. Above table clearly indicates clearly that methanol can be used as a principle extracting solvent and also evaporation of methanol is easy.

e. HPTLC analysis of Methanolic extract

Phytochemical profiling and quantification of Quercetin and Gallic acid in the methanolic extract of root of *Beta Vulgaris L* was studied and results were obtained in the form chromatograms depicted in figures 3and 4. Chromatograms from standards and test samples were obtained by CAMAG TLC scanner III at short (254 nm) (fig. 3) and long (366 nm) (fig.4) wavelength.

Table 4: HPTLC details of methanolic extract of root *of Beta Vulgaris L*

Band number	Rf Value		Assigned substances
	254 nm	366 nm	
1	0.526,0.265,0.126,0.210	0.692,0.542,0.471, 0.238	Methanolic extract of *Beta Vulgaris L* root
2	0.202	0.238,0.455, 0.452,	Gallic acid
3	0.526,0.412,	0.551,0.452	Quercetin
4	0.455	0.455	Flavanoids

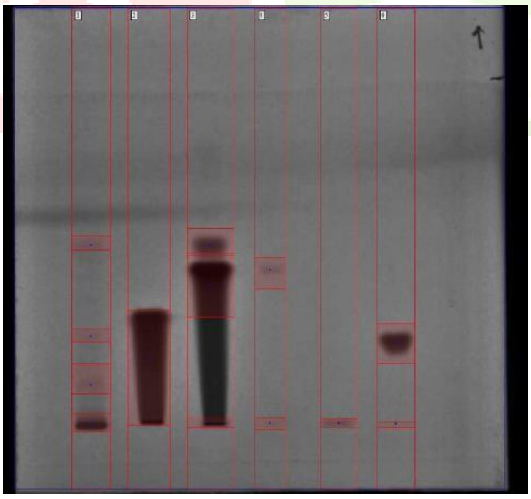

Figure 4-image of TLC plate at 254nm

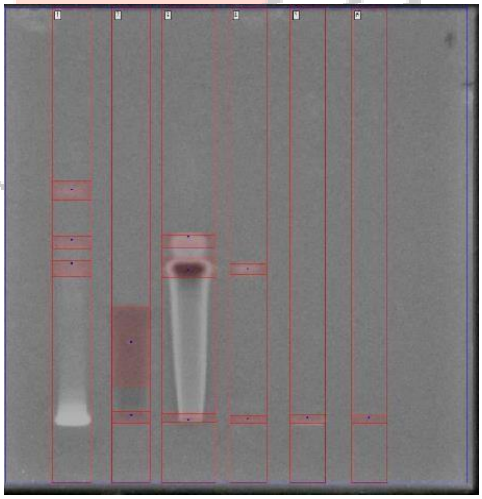

Figure 5-image of TLC plate at 366nm

Figure 4 and 5 indicates the presence of Gallic Acid and Quercetin in the methanolic extract of root of *Beta Vulgaris L*.

V. CONCLUSION:

The current study advances science by making pioneering preliminary findings regarding physicochemical properties, the presence of useful constituents through HPTLC, microscopic diagnostic characteristics through powder microscopy, and sufficient scientific material to initiate future studies As HPTLC phytochemical profiling reveals the presence of several bioactive compounds like Quercetin and gallic acid in the methanolic extract of Beta Vulgaris reveals that can be further explored up to their identification and future application in pharmacological treatment.

VI. FUTURE SCOPE:

The current study may contribute to research pioneering preliminary study with respect to pharmacognostical physiochemical, phytochemical and advanced parameters like HPTLC so that benefits of Beetroot reaches out their therapeutic values.

VII. ACKNOWLEDGEMENT:

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REFERENCES:

1) Afiomah, C. S., & Iwuozor, K. (2020). Nutritional and phytochemical properties of Beta vulgaris Linnaeus (Chenopodiaceae)–a review. Nigerian Journal of Pharmaceutical and Applied Science Research, 9(4), 38-44.Khadabadi SS, Deore SL. Experimental pharmacognosy, a comprehensive guide.Nirali Prakashan. 2013; 2: 1.3-1.4.
2) Asadi, S. Z., & Khan, M. A. (2021). The effect of beetroot (Beta vulgaris L.) root powder on nutritional, textural, sensorial and antioxidant properties of cookies. Journal of Culinary Science & Technology, 19(5), 424-438.
3) Bangar, S. P., Sharma, N., Sanwal, N., Lorenzo, J. M., & Sahu, J. K. (2022). Bioactive potential of beetroot (Beta vulgaris). Food Research International, 158, 111556.
4) Dlim, M. M., Alsabri, S. G., Mohamed, S. S., Zetrini, A. E., Salem, A. A., Auzi, A. A., & Mohamed, S. B. (2013). Use of Beta vulgaris as natural coloring agent for foods and cosmetics in Libya. J. Chem. Pharm. Res, 5(11), 340-345.
5) Indirani K et.al.(2021) Evaluation of functional properties of beetroot powder (beta vulgaris) and its suitability in developing a candy by incorporate, International Journal of Science and Healthcare Research Vol.6; Issue: 1, ISSN: 2455-7587,144-147
6) Jayet, Edziri & Jaziri, Raouf & Haddad, Ons & Anthonissen, R & Aouni, Mahjoub & Mastouri, Maha & Verschaeve, Luc. (2019). Phytochemical analysis, antioxidant, anticoagulant and in vitro toxicity and genotoxicity testing of methanolic and juice extracts of Beta vulgaris L.. South African Journal of Botany. 126. 10.1016/j.sajb.2019.01.017.
7) John, S., Monica, J., Priyadarshini, S., Sivaraj, C., & Arumugam, P. (2017). Antioxidant and antibacterial activities of beta vulgaris l. peel extracts. Int. J. Pharma Res. Health Sci, 5(6), 1974-79.
8) Odoh, U. E., Ezugwu, C. O., & Okoro, E. C. (2012). Quantitative phytochemical, proximate/nutritive composition analysis of Beta Vulgaris Linnaeus (Chenopodiceae). Planta Medica, 78(11), PI116.
9) Oghogho, U. I., Ekugum, E., Ogbeide, O. K., Idagan, M., Uadia, J. O., & Falodun, A. (2022). Phytochemical Assessment, Anti- inflammatory and Antimalarial Activities of Beta vulgaris (Chenopodiaceae) Root Extract: http://www.doi.org/10.26538/tjpps/v1i1.3. Tropical Journal of Phytochemistry and Pharmaceutical Sciences, 1(1), 3–8.
10) Patel, M., & Patel, P. (2022). HPTLC fingerprinting analysis of phytoconstituents from Bixa orellana and Beta vulgaris plant pigment. J Adv Biotechnol Exp Ther, 5(2), 292-306.
11) Pratimasari, D., & Puspitasari, D. (2022). Identification of Flavonoid Compounds from Purified Extract of Beetroot Root (Beta Vulgaris L). Indonesian Journal of Global Health Research, 4(4), 811-816.
12) Pratimasari, D., & Puspitasari, D. (2022). Identification of Flavonoid Compounds from Purified Extract of Beetroot Root (Beta Vulgaris L). Indonesian Journal of Global Health Research, 4(4), 811-816.
13) Rotich, V., Wangila, P., & Cherutoi, J. (2022). Method Validation and Characterization of Red Pigment in Beta vulgaris Peels and Pomaces by HPLC-UV and UHPLC-MS/MS. Journal of Analytical Methods in Chemistry, 2022.
14) Thiruvengadam, M., Chung, I. M., Samynathan, R., Chandar, S. H., Venkidasamy, B., Sarkar, T., ... & Simal-Gandara, J. (2022). A comprehensive review of beetroot (Beta vulgaris L.) bioactive components in the food and pharmaceutical industries. Critical Reviews in Food Science and

Nutrition, 1-33.
15) Thiruvengadam, M., Chung, I. M., Samynathan, R., Chandar, S. H., Venkidasamy, B., Sarkar, T., & Simal-Gandara, J. (2022). A comprehensive review of beetroot (Beta vulgaris L.) bioactive components in the food and pharmaceutical industries. Critical Reviews in Food Science and Nutrition, 1-33.
16) Zia, P., Sunita, M., & Sneha, S. (2021). Extraction of natural colour from beet root (Beta vulgaris) its phytochemical analysis and antibacterial activity. EAS Journal of Nutrition and Food Sciences, 3(4), 80-85.

Conference Certificates

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Online National Seminar

On

Intellectual Property Rights In Indian Context And Its Present Relevance

08 September, 2020

Certificate Awarded to

Dr./Mr./Ms. **Trupti Bhagwanrao Shevante**

of Pacific College of Pharmacy

for participation in this Online National Seminar on "Intellectual Property Rights In Indian Context And Its Present Relevance" Organized by Department of Post Graduate Studies & IQAC Cell, Pacific Academy of Higher Education and Research University, Udaipur.

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This is to Certify that,

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participated & completed as a delegate & presented Paper titled

Phytochemical and HPTLC Studies of Sesbania Grandiflora L. during International Conference held at SMBT Campus on January 06th-07th, 2023. We duly acknowledge his/her participation.

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CERTIFICATE

OF PARTICIPATION

Ms. Shevante Trupti B.

has participated in the International Faculty Development Program (iFDP) **"Emerging Trends in Pharmaceutical Research"** organized by Dr. D. Y. Patil Institute of Pharmaceutical Sciences and Research, Pimpri, Pune - 411 018 (MH), INDIA from 20th Feb to 15th May 2021. His / Her participation in this iFDP Programme is highly appreciated.

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