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Phytochemical Profiling, *In Vitro* Anti-Inflammatory Activity and GC–MS Characterization of *Picrorhiza kurroa* Royle ex Benth. Root Extract

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Abstract - Picrorhiza kurroa is a medicinal Himalayan plant traditionally used in herbal medicine and is recognized for its phytochemical and pharmacological importance. The present study evaluated the phytochemical profile of *P. kurroa* root extracts prepared with solvents of different polarity and examined *in vitro* anti-inflammatory activity, protein content and GC–MS-based chemical characterization. Dried root powder was extracted separately with ethanol, chloroform, petroleum ether and distilled water by maceration for approximately 48 h. Qualitative phytochemical screening was performed for major secondary-metabolite classes. Antimicrobial testing was documented by disc diffusion, anti-inflammatory activity was assessed using a bovine serum albumin (BSA) protein-denaturation assay, protein content was estimated by the Lowry method and GC–MS was used for chemical profiling. Ethanol extract showed the broadest qualitative phytochemical profile, with alkaloids, flavonoids, tannins, glycosides, proteins, carbohydrates, steroids, resins, phenols and terpenoids detected. The aqueous extract also showed a broad profile, whereas petroleum ether contained fewer detected classes. In the protein-denaturation assay, Test 3 showed the strongest inhibition among the tested samples and was closest to the standard according to the project record. The Lowry assay gave an optical density of approximately 0.48 for the test sample compared with approximately 0.95 for the BSA standard. GC–MS profiling tentatively identified eight major compounds: caryophyllene, spathulenol, caryophyllene oxide, tetradecanoic acid, n-hexadecanoic acid, phytol, kauren-19-oic acid and palustric acid. These findings indicate that *P. kurroa* root contains diverse phytochemical constituents and a GC–MS-detectable profile dominated by terpenoid- and fatty-acid-related compounds. The observed *in vitro* protein-denaturation inhibition supports further investigation; however, quantitative chemical analysis, standardized antimicrobial testing, compound isolation and mechanistic studies are required before therapeutic claims can be made.

Keywords - Picrorhiza kurroa, phytochemical screening, anti-inflammatory activity, GC–MS, caryophyllene, Phytol.

1. INTRODUCTION

Medicinal plants remain important sources of chemically diverse compounds investigated for pharmacological applications. *Picrorhiza kurroa* Royle ex Benth., commonly known as Kutki or Katuki, is a perennial medicinal herb of the family Plantaginaceae that occurs in high-altitude Himalayan environments [1]. Its rhizomes are widely used in traditional medicine, particularly in relation to liver and inflammatory disorders. The project report describes *P. kurroa* as containing several classes of phytochemicals, including iridoid glycosides, flavonoids, phenolic compounds, alkaloids, tannins and terpenoids [2].

The biological evaluation of medicinal plants benefits from combining preliminary phytochemical screening with functional assays and chemical profiling. GC–MS can provide a useful profile of volatile and semi-volatile constituents, while *in vitro* protein-denaturation assays can provide preliminary evidence of protein-stabilizing activity [3]. However, detection of a compound by GC–MS does not by itself establish its biological activity or causality. The present study therefore aimed to characterize *P. kurroa* root extracts using qualitative phytochemical screening and GC–MS and to document their preliminary biological properties, particularly *in vitro* anti-inflammatory activity [4].

Fig. 1. Picrorhiza kurroa plant.

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Fig. 2. P. kurroa root (left) and root powder (right).

2. MATERIALS AND METHODS

2.1 Plant Material and Processing

P. kurroa roots were obtained from an herbal supplier and authenticated by a qualified botanist; the project report states that a voucher specimen was preserved. The roots were washed, shade-dried for approximately 7–10 days, ground to powder and stored in airtight containers until extraction [5].

2.2 Extraction

Approximately 25 g of dried root powder was extracted separately with ethanol, chloroform, petroleum ether and distilled water. The mixtures were maintained at room temperature for approximately 48 h with intermittent shaking. Extracts were filtered through Whatman No. 1 filter paper, concentrated by low-temperature water-bath evaporation, and stored at 4 °C until analysis [6].

Fig. 3. Maceration/soaking of P. kurroa root powder in different solvents.

Fig. 4. Filtration of P. kurroa root extracts.

2.3 Qualitative Phytochemical Screening

Extracts were screened qualitatively for alkaloids, flavonoids, phenols, tannins, glycosides, proteins, carbohydrates, saponins, steroids, resins and terpenoids using standard colour and precipitation-based tests described in the project report, including Wagner's and Mayer's tests for alkaloids, Shinoda and lead-acetate tests for flavonoids, ferric-chloride and related tests for phenolics, ferric-chloride testing for tannins, Legal's test for glycosides, Xanthoproteic testing for proteins, Molisch/Fehling/Benedict tests for carbohydrates, foam testing for saponins and appropriate tests for steroids, resins and terpenoids [7].

2.4 Antimicrobial Testing

The project report describes disc-diffusion testing on Mueller–Hinton agar with incubation at 37 °C for 24 h. The available results record inhibition-zone diameters for antibiotic discs. Because the report does not provide extract concentration, microbial species or strain identifiers, extract-specific zone measurements, replicate number or statistical analysis, extract-specific antimicrobial efficacy is not interpreted as a primary quantitative outcome in this manuscript [8].

2.5 In Vitro Anti-Inflammatory Assay

Protein-denaturation inhibition was evaluated using bovine serum albumin (BSA). Test mixtures containing plant extract and BSA in phosphate buffer (pH 6.4) were incubated at 37 °C for 15 min and heated at 70 °C for 5 min. After cooling, absorbance was measured at 660 nm. The project report states that Test 3 showed the highest inhibitory effect among the test samples and was closest to the standard [9].

2.6 Protein Estimation

Protein content was estimated by the Lowry method using BSA as the standard. Absorbance was measured at 660 nm. The project report records an optical density of approximately 0.95 for the standard and approximately 0.48 for the test sample [10].

2.7 GC–MS Analysis

The concentrated plant extract was filtered and introduced into a GC–MS instrument. Components were separated chromatographically and identified by comparison of mass spectra with the NIST library [11]. The report lists retention time and compound name for eight major detected compounds. Instrument model, column specification, carrier gas, temperature programme, injection conditions, library match scores and peak-area percentages were not provided in the available project text and should be added from the original instrument record before journal submission [12].

3. RESULTS

3.1 Phytochemical Profile

Qualitative screening demonstrated solvent-dependent differences in the detected phytochemical classes. Ethanol produced the broadest profile, with ten of the eleven screened classes detected; saponins were not detected. The aqueous extract also showed a broad profile, whereas petroleum ether showed only steroids, resins and terpenoids [13].

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Fig. 5. Qualitative phytochemical analysis of *P. kurroa* root extracts.

Table 1. Qualitative phytochemical screening of P. kurroa root extracts. + = detected; – = not detected.

Phyto chemical	Ethanol	Chloro form	Petroleum ether	Distilled water
Alkaloids	+	+	–	–
Flavonoids	+	+	–	+
Tannins	+	–	–	+
Glycosides	+	+	–	+
Protein	+	+	–	+
Carbohydrates	+	–	–	+
Saponins	–	+	–	+
Steroids	+	+	+	+
Resins	+	+	+	–
Phenols	+	–	–	+
Terpenoids	+	+	+	+

3.2 Antimicrobial Testing Record

The project records inhibition-zone diameters for antibiotic discs rather than extract-specific quantitative results. Streptomycin showed the largest recorded zone (~14 mm), followed by vancomycin (~13 mm), fluconazole and kanamycin (~12 mm each), ampicillin (~11 mm), penicillin-G (~10 mm), erythromycin (~9 mm), and methicillin (~8 mm). These values describe the antibiotic controls or standards documented in the project and should not be reported as inhibition produced by P. kurroa extract [14].

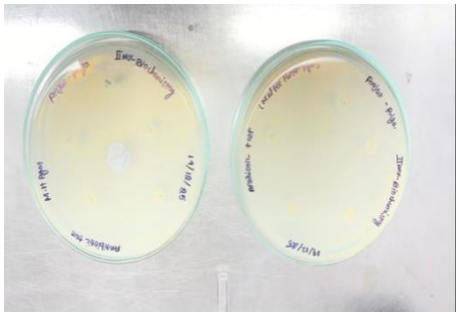

Fig. 6. Antimicrobial assay before incubation.

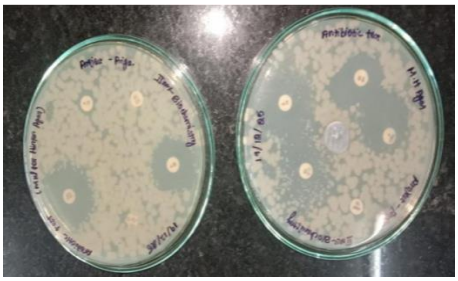

Fig. 7. Antimicrobial assay after incubation.

Table 2. Inhibition-zone measurements reported for antibiotic discs in the project record.

S. No.	Retention time (min)	Compound
1	12.8289	Caryophyllene
2	14.8245	Spathulenol
3	14.9082	Caryophyllene oxide
4	16.7836	Tetradecanoic acid
5	18.8717	n-Hexadecanoic acid
6	20.2940	Phytol
7	22.9996	Kauren-19-oic acid
8	23.1088	Palustric acid

3.3 *In Vitro* Anti-Inflammatory Activity

The protein-denaturation assay showed lower optical-density values for Test 1, Test 2 and Test 3 than the control, according to the project record. Test 3 showed the strongest inhibition among the test samples and was closest to the standard. Exact replicate values, standard identity and concentration and percentage-inhibition values are not supplied in the project text and should be recovered from the original laboratory data before submission [16].

Table 3. Compounds reported as identified by GC–MS in the project. Identification should be described as tentative unless confirmed by authentic standards and/or orthogonal analytical data.

Antibiotic disc	Zone diameter (mm)
Penicillin-G	~10
Fluconazole	~12
Streptomycin	~14
Vancomycin	~13
Ampicillin	~11
Erythromycin	~9
Methicillin	~8
Kanamycin	~12

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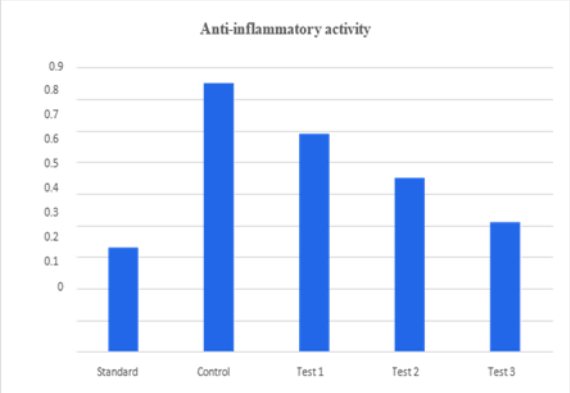

Fig. 8. Protein-denaturation assay results recorded as anti-inflammatory activity.

3.4 Protein Estimation

The Lowry assay produced an optical density of approximately 0.95 for the BSA standard and approximately 0.48 for the plant test sample. The project therefore confirms detectable protein-associated colour development in the tested sample [15].

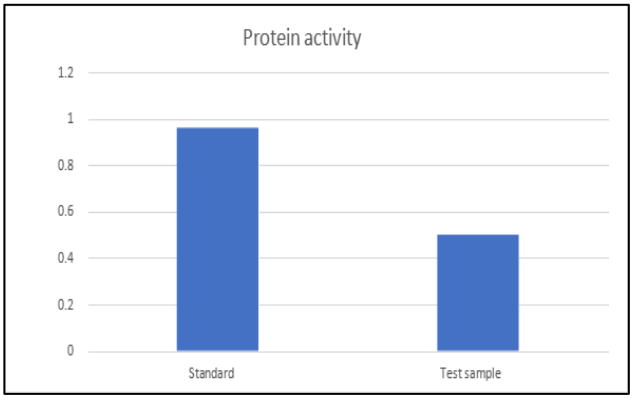

Fig. 9. Protein estimation by the Lowry assay.

3.5 GC–MS Profile

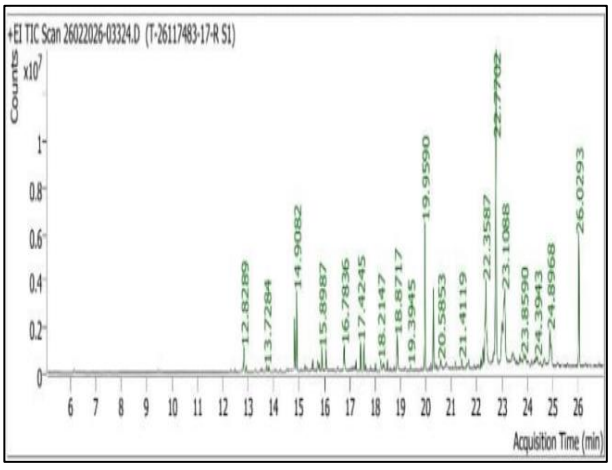

Fig. 10. GC–MS chromatogram of *P. kurroa* root extract.

4. DISCUSSION

The solvent-dependent phytochemical pattern is consistent with the principle that solvent polarity influences the classes of constituents recovered from plant material [17]. In the present dataset, ethanol showed the broadest qualitative profile, whereas petroleum ether detected only three classes. The aqueous extract also displayed a relatively broad profile. These observations support the use of solvent selection as an important variable in preliminary medicinal-plant investigations [18].

The anti-inflammatory experiment provides preliminary evidence that the extract can reduce protein denaturation under the assay conditions used [19]. Test 3 was reported as the most active sample and closest to the standard. Nevertheless, protein-denaturation assays are screening-level in vitro models and do not establish inhibition of inflammatory pathways in cells or organisms. The absence of replicate-level data and percentage-inhibition values in the report prevents a robust statistical comparison [20].

The GC–MS analysis reported eight compounds, principally terpenoid- and fatty-acid-related constituents. Caryophyllene, caryophyllene oxide, spathulenol, and phytol are chemically plausible constituents of plant extracts and have been associated in the literature with biological activities [21]. In this study, however, their detection should be interpreted as a chemical-profile observation rather than proof that any individual compound caused the measured activity. The project itself appropriately identifies HPLC/LC–MS quantification, compound isolation, and mechanistic studies as necessary next steps [22].

A major limitation concerns the antimicrobial dataset. Although disc-diffusion experiments are described, the reported table contains antibiotic-disc zones and does not provide the extract concentration, microbial strain details, extract-specific inhibition zones, or replication and statistical information. Therefore, the present manuscript deliberately avoids making a quantitative claim that the P. kurroa extract inhibited the tested organisms. This correction improves scientific accuracy and will be important during peer review [23].

5. CONCLUSION

P. kurroa root extracts showed clear solvent-dependent qualitative differences in phytochemical composition, with ethanol producing the broadest profile in the present dataset. The in vitro protein-denaturation assay indicated preliminary anti-inflammatory potential, with Test 3 reported as the strongest test sample. GC–MS profiling reported eight major constituents, including caryophyllene, spathulenol, caryophyllene oxide, tetradecanoic acid, n-hexadecanoic acid, phytol, kauren-19-oic acid, and palustric acid. Collectively, these findings justify further phytochemical standardization and mechanistic investigation, but they are not sufficient to establish therapeutic efficacy. Quantitative chemical analysis, standardized biological assays, and compound-level validation are recommended.

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6. DECLARATIONS

Ethics approval: Not applicable to the experiments described in the available project record.

Conflict of interest: To be completed by the authors.

Data availability: The data reported in this manuscript are derived from the available project record; missing experimental details should be recovered from the original laboratory records before submission.

7. REFERENCES

[1] A. W. Bauer, W. M. Kirby, J. C. Sherris, and M. Turck, "Antibiotic susceptibility testing by a standardized single disk method," American Journal of Clinical Pathology, 1966.

[2] M. M. Cowan, "Plant products as antimicrobial agents," Clinical Microbiology Reviews, vol. 12, no. 4, pp. 564–582, 1999.

[3] W. C. Evans, Trease and Evans Pharmacognosy, 15th ed. London, U.K.: W. B. Saunders, 2002.

[4] D. S. Fabricant and N. R. Farnsworth, "The value of plants used in traditional medicine for drug discovery," Environmental Health Perspectives, vol. 109, suppl. 1, pp. 69–75, 2001.

[5] P. D. Gupta *et al*., "Phytochemical and pharmacological importance of medicinal plants," Journal of Pharmacognosy and Phytochemistry, vol. 6, no. 5, pp. 349–354, 2017.

[6] J. B. Harborne, Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. London, U.K.: Chapman & Hall, 1998.

[7] K. R. Khandelwal, Practical Pharmacognosy: Techniques and Experiments, 19th ed. Pune, India: Nirali Prakashan, 2008.

[8] K. R. Kirtikar and B. D. Basu, Indian Medicinal Plants, 2nd ed. International Book Distributors, 2005.

[9] V. Kumar *et al*., "Pharmacological properties of Picrorhiza kurroa: A review," International Journal of Pharmaceutical Sciences Review and Research, vol. 20, no. 1, pp. 148–155, 2013.

[10] O. H. Lowry et al., "Protein measurement with the Folin-phenol reagent," Journal of Biological Chemistry, 1951.

[11] Y. Mizushima and M. Kobayashi, "Interaction of anti-inflammatory drugs with serum proteins," Journal of Pharmacy and Pharmacology, 1968.

[12] P. K. Mukherjee, Quality Control and Evaluation of Herbal Drugs. Elsevier, 2019.

[13] K. M. Nadkarni, Indian Materia Medica, 3rd ed. Mumbai, India: Popular Prakashan, 2009.

[14] D. J. Newman and G. M. Cragg, "Natural products as sources of new drugs over the nearly four decades from 1981 through 2019," Journal of Natural Products, vol. 83, no. 3, pp. 770–803, 2020.

[15] G. B. Singh *et al*., "Traditional uses and pharmacological properties of Picrorhiza kurroa," Journal of Ethnopharmacology, vol. 136, no. 1, pp. 1–12, 2011.

[16] O. D. Sparkman, Z. Penton, and F. G. Kitson, Gas Chromatography and Mass Spectrometry: A Practical Guide. Academic Press, 2011.

[17] G. E. Trease and W. C. Evans, Pharmacognosy, 16th ed. Saunders Elsevier, 2009.

[18] S. Sudha, B. Chitra, and S. Arif Nisha, "Evaluation of neuroprotective effect and identification of active compounds from *Nigella sativa* Linn. through bioactive guided fractionation," Research Journal of Biotechnology, vol. 19, no. 3, pp. 39–47, 2024.

[19] S. Sudha, B. Chitra, and S. Arif Nisha, "Evaluation of neuroprotective effect of *Nigella sativa* Linn. seed using zebrafish model," Uttar Pradesh Journal of Zoology, vol. 45, no. 6, pp. 150–163, 2024.

[20] S. Sudha, B. Chitra, and S. Arif Nisha, "Molecular docking studies of *Nigella sativa* Linn. seed compound against Alzheimer's disease: An *in silico* study," Asian Journal of Advances in Medical Science, vol. 6, no. 1, pp. 32–42, 2024.

[21] S. Sudha, B. Chitra, and S. Arif Nisha, "Ameliorating Alzheimer's diseases: Novel therapeutic approaches of traditional medicines," Biodiversity and Sustainable Resource Management, pp. 271–283, 2021.

[22] S. Sudha, B. Chitra, C. Janani, and S. Arif Nisha, "Assessment of antioxidant and anticholinesterase activity of methanolic extract from *Nigella sativa* Linn.," Gorteria Journal, vol. 34, no. 6, pp. 858–865, 2021.

[23] S. Sudha and S. Arif Nisha, "Assessment of antioxidant and anticholinesterase activity of methanolic extract of *Nigella sativa*," International Journal of Life Science and Pharma Research, 2020.