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Solvent-Dependent Phytochemical Diversity in Justicia Tranquebariensis: A Comparative Analysis of Methanol and Ethyl Acetate Extractions

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ABSTRACT:

This study investigates the impact of solvent polarity on the extraction efficiency and phytochemical profiles of *Justicia tranquebariensis*, a plant traditionally valued for its medicinal properties. Bioactive compounds were extracted from dried plant material using methanol (polar) and ethyl acetate (semi-polar) via Soxhlet extraction. Methanol yielded a significantly higher extraction rate (24.17%) compared to ethyl acetate (11.82%) and a broader range of phytochemicals, including alkaloids, flavonoids, steroids, terpenoids, phenols, tannins, and carbohydrates. In contrast, the ethyl acetate extract was characterized by flavonoids, steroids, terpenoids, carbohydrates, and oils/resins. These findings highlight the crucial role of solvent selection in determining the quantity and diversity of extracted secondary metabolites from *Justicia tranquebariensis*, providing valuable insights for targeted phytochemical investigations and potential drug discovery efforts.

Keywords: *Justicia tranquebariensis*, Phytochemical Analysis, Solvent Polarity, Methanol Extraction, Ethyl Acetate Extraction, Medicinal Plants, Natural Products, Secondary Metabolites

INTRODUCTION:

Globally recognized as invaluable reservoirs of novel pharmaceuticals and bioactive molecules, medicinal plants have historically underpinned a significant portion of modern pharmacotherapy [1][2]. The isolation and characterization of numerous secondary metabolites from these botanical sources have unveiled potent pharmacological activities, underscoring the importance of traditional herbal remedies in drug discovery. *Justicia tranquebariensis*, a low-growing subshrub belonging to the Acanthaceae family, has emerged as a subject of interest due to its documented therapeutic applications. Morphologically distinguished by its diminutive stature, delicate leaves measuring approximately 2.5–3 cm, and a characteristic bilabiate corolla adorned with pink markings on the lower petal, this species is locally known by names such as Sivanervembu or Thavasimurungai in certain regions [3][4]. Traditionally, various parts of *J. tranquebariensis* have been employed for medicinal purposes; for instance, its leaf juice is used as a cooling agent and laxative, and to treat ailments like smallpox and jaundice. Furthermore, a paste prepared from its leaves is applied externally to diminish swelling, relieve pain, and even as a traditional remedy for cobra bites [5][6][7]. In an era marked by escalating demands for natural health products and the increasing vulnerability of plant resources due to habitat destruction and unsustainable harvesting



practices, a comprehensive evaluation focused on optimizing the extraction and analysis of the valuable phytochemical constituents of *J. tranquebariensis* becomes strategically crucial for both its conservation and its potential contribution to future drug development. The present study, therefore, aims to address this need by comparatively assessing the efficacy of two distinct solvent systems—methanol and ethyl acetate—in extracting key phytochemicals from this medicinally important plant.



LITERATURE REVIEW:

The efficacy of segregating biologically active composites from factory matrices is critically dependent on the physicochemical properties of the birth detergent. Being literature underscores the significant influence of solvent opposition on both the yield and the spectrum of uprooted phytochemicals. Largely polar detergents, similar to methanol, are extensively honored for their effectiveness in rooting polar composites, including polyphenols, flavonoids, and tannins[8][9]. Again, semi-polar detergents like ethyl acetate tend to preferentially solubilize less polar ingredients, similar to certain terpenoids, canvases, and resins[8][9]. Studies in [10] and [11] have empirically demonstrated these solvent-dependent variations in birth issues across different factory species. Likewise, a broader review of medicinal factory exploration highlights that the choice of detergent is vital for accurate phytochemical profiling, which latterly impacts the evaluation of pivotal bioactivities like antioxidant, antimicrobial, and anticancer properties [12][13]. Parallels drawn from exploration on other factory species suggest an implicit correlation between an advanced yield of uprooted secondary metabolites and enhanced pharmacological exertion[14][15]. This present disquisition builds upon this established body of knowledge by conducting a direct relative analysis of methanol and ethyl acetate as birth detergents for Justicia tranquebariensis. The end is to interpret the qualitative and quantitative differences in the uprooted phytochemical biographies, thereby furnishing precious perceptivity to guide unborn pharmacological evaluations of this important medicinal factory.

METHODOLOGY:

Collection of Materials

Fresh samples of *Justicia tranquebariensis* were randomly collected from the Katpadi area. The plant materials, known for their traditional medicinal uses, were thoroughly washed with running tap water, air dried, and ground into a fine powder. The powdered material was stored in airtight bottles in a refrigerator until further processing.



Preparation of Extract

Crude extracts were prepared using the Soxhlet extraction method. Approximately 20 g of the homogenized powder was packed into a thimble and extracted separately with 250 mL of methanol and ethyl acetate. The extraction was carried out continuously for 24 hours or until the siphon tube's solvent became colorless. The extracts were then concentrated on a hot plate at 30–40°C until complete solvent evaporation. The dried extracts were stored at 4°C for further analysis.

Phytochemical Screening

Preliminary qualitative phytochemical analysis—following the standard protocols outlined by Evans (1996) and Brain and Turner (1975)—was conducted on both the methanol and ethyl acetate extracts. The following standard tests were used:

- <u>Alkaloids</u>: Mayer's test (formation of a cream-colored precipitate indicates presence).
- <u>Flavonoids</u>: Sulphuric acid test (reddish brown/orange precipitate indicates presence).
- <u>Steroids</u>: Liebermann-Burchard test (violet to blue/green coloration indicates presence).
- Terpenoids: Salkowski's test (reddish brown precipitate indicates presence).
- <u>Anthraquinones</u>: Borntrager's test (pink coloration indicates presence).
- <u>Phenols:</u> Ferric chloride test (bluish-black/deep blue coloration indicates presence).
- <u>Saponins</u>: Froth test (stable, persistent froth indicates presence).
- <u>Tannins</u>: Ferric chloride test (brownish-green/blue-black precipitation indicates presence).
- <u>Carbohydrates</u>: Fehling's test (color changes indicative of sugars).
- <u>Oils and Resins</u>: Spot test on filter paper (transparent appearance indicates presence).

RESULT:

The yield from the methanol extract was approximately 40% higher than that of the ethyl acetate extract, thus supporting the selection of methanol for subsequent detailed analyses.

S. No	Methanol Extract	Ethyl Acetate Extract
1	24.17	11.82

Table 1 Yield Obtained in Justicia tranquebariensis:

Phytochemical Analysis

Phytochemical screening results for both extracts are presented in Tables 2 and 3.Test results in Figs 1 and 2. Overall, the methanol extract showed a broader range of positive tests, indicating higher concentrations of alkaloids, flavonoids, steroids, terpenoids, phenols, tannins, and carbohydrates, compared to the ethyl acetate extract

Phyto chemicals	Observations	Ethyl Acetate Extract
Alkaloids Mayer's test	Cream colour	-
Flavonoid s H ₂ SO4 test	Reddish brown / Orange colour precipitate	+

Table 2 phytochemical analysis in Ethyl Acetate extract



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Steroids		+	
Liebermann-Burchard test	Violet to blue or Green colour formation	т Т	
Terpenoids		+	
Salkowski test	Reddish brown precipitate	+	
Anthraquinone			
Borntrager's test	Pink colour	-	
Phenols			
Ferric chloride test	Deep blue to Black colour formation	-	
Saponin	Stalls annistant		
Froth test	Stable persistent	-	
Tannin	Brownish groon / Blue bleek	-	
Ferric chloride test	Brownish green / Blue black		
Carbohydrates	Vallandaran adam	+	
Fehling's test	Yellow/brownish/blue/green colour		
Oils & Resins	Filter mener method	+	
Spot test	Filter paper method		

Table 3 Phytochemical Analysis in Methanol Extract

Phyto	Observations	Methanol Extract
chemicals		
Alkaloids		+
Mayer's test	Cream colour	
Flavonoids		+
H ₂ SO ₄ test	Reddish brown / Orange colour	
	precipitate	
Steroids		+
Liebermann-Burchard	Violet to blue or green colour formation	
test		
Terpenoids		+
Salkowski test	Reddish brown precipitate	
Anthraquinone		-
Borntrager's test	Pink colour	
Phenols		+
Ferric chloride test	Deep blue to Black colour formation	
Saponin	Stable persistent	-
Froth test		
Tannin	Brownish green / Blue black	+
Ferric chloride test		
Carbohydrates	Yellow/brownish/blue/green colour	+
Fehling's test		
Oils & Resins	Filter paper method	-
Spot test		



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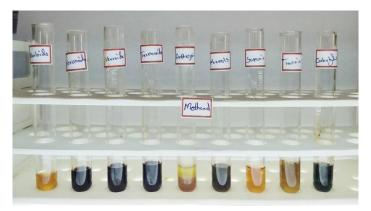


Fig 1: Methanol Extract



Fig 2: Ethyl Acetate Extract

DISCUSSION:

The findings indicate that solvent polarity critically affects both the yield and diversity of phytoconstituents extracted from *J. tranquebariensis*. The methanol extract yielded 24.17% extract mass, which is significantly higher (approximately 40% greater) than the 11.82% yield from ethyl acetate. Furthermore, qualitative analysis revealed that the methanol extract retained a wider array of bioactive compounds, including alkaloids and tannins that were not detectable in the ethyl acetate extract.

These results underscore the importance of using polar solvents like methanol to maximally extract polar phytochemicals such as phenols, flavonoids, and carbohydrates. Such compounds are associated with a range of therapeutic properties, including antioxidant, anti-inflammatory, and antimicrobial effects [15]. In contrast, the ethyl acetate extract was more selective, recovering key constituents such as flavonoids, steroids, terpenoids, carbohydrates, and oils/resins—yet it did not capture the full spectrum of bioactive secondary metabolites. This may influence the choice of solvent when preparing extracts for further pharmaceutical applications.

CONCLUSION:

This study provides a comprehensive comparison between methanol and ethyl acetate as extraction solvents for *Justicia tranquebariensis*. Key findings include:

- **Yield Efficiency:** Methanol extraction yielded 24.17%, which is approximately 40% higher than ethyl acetate extraction.
- **Phytochemical Richness:** The methanol extract demonstrated a fuller phytochemical profile, showing positive results for alkaloids, flavonoids, steroids, terpenoids, phenols, tannins, and carbohydrates.



• **Therapeutic Implications:** The broader compound profile in the methanol extract suggests its potential for greater therapeutic efficacy and supports its use in further pharmacological evaluations and drug development.

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