

Chapter 2

Investigating *Cyperus compressus* L. (Cyperaceae) as a natural antibacterial support for gut health in livestock

Neelakshi Sharma¹, Sanjeet Kumar², Rajkumari Supriya Devi², Sweta Mishra², Sugimani Marndi² and Sumitra Jethy^{2*}

¹Royal School of Pharmacy, The Assam Royal Global University, Betkuchi, Guwahati, Assam, India

²Ambika Prasad Research Foundation, Odisha India

*Email-Id: sumitrajethy19@gmail.com

ABSTRACT

The increasing misuse of antibiotics in veterinary and livestock practices has significantly contributed to the rise of antimicrobial resistance and disruption of natural gut microflora. This growing concern highlights the urgent need for safer, plant-based alternatives that can support animal health while reducing dependency on synthetic drugs. *Cyperus compressus* in this context was evaluated for its phytochemical composition and antibacterial activity against *Escherichia coli*, a common gut inhabitant and opportunistic pathogen in both humans and animals. Qualitative phytochemical screening revealed the presence of key secondary metabolites, particularly in polar extract and antibacterial assay demonstrated a noticeable inhibitory effect against *E. coli*, supporting the traditional claims of its therapeutic usefulness. These finding not only validate the ethnomedicinal relevance of this lesser studied sedge but also emphasizes its potential as a natural antimicrobial and immune supporting agent in veterinary applications.

Keywords: Antibacterial resistance, gut health, sedges

INTRODUCTION

Animal gut is considered a microbiome as it homes trillions of microorganisms including bacteria, fungi, viruses and archea that helps in maintenance and management of digestion, immunity, and metabolism (Quigley, 2013). It is also

connected to the brain through the gut-brain axis which is a complex bidirectional network of nerves, hormones and immune signals which not only impact behaviors like stress, anxiety and social motivation but also acts as a protective barrier against pathogens (Carabotti *et al.*, 2015). *Escherichia coli* is a very common microbe found in the gut of many animals. It is one of the first microbes to colonize the gut post birth and usually considered harmless and sometimes, even aid digestion (Martinson and Walk, 2020). As being a part of the gut microbiota in mammals, it helps maintain a healthy gut environment but its pathogenic strains can cause a range of diseases in both humans and animals (Pokharel *et al.*, 2023). Diseases caused by enteropathogenic, enter invasive and cytotoxic (EPEC) strains of *E. coli* can range from mild to severe (Evans and Evans, 1996). Because it's a facultative anaerobe, wild and domestic animals, particularly ruminants like cattle are a natural reservoir for its pathogenic strains (Tabaran *et al.*, 2022). This cause diseases like Diarrhea, Enteritis, Septicemia (in young animals) and reduced immunity. Gut imbalance and pathogenic *E. coli* threatens animal health. While we may think up of animal health from a human perspective, animals instinctively consume certain plants for self-medication known as zoopharmacognosy (Domínguez-Martín *et al.*, 2020). Many herbivores, even carnivores change their dietary habits when sick, like grazing plants with known medicinal properties like high in tannins or other secondary metabolites (Yoshimura *et al.*, 2021). It not only helps in gaining back their metabolism but also serve as a natural form of deworming, contributing towards gaining resilience against future pathogenic health threat (Foley and Moore, 2005). Hence, the role of secondary metabolites like presence of tannins, phenols, flavonoids, etc. becomes essential in maintain ecological balance. Common plant groups like cyperaceae constitute a major part of the food for the grazing strata of the food pyramid. They are rich in both primary and secondary metabolite which adds to the nutritional and defense aspect respectively (Roy and Devi, 2025). *Cyperus compressus* L. (POWO, 2025) common to seasonally dry tropical habitats are annual sedge which is found among grasslands, agricultural fields, river and lake sides, and open fields especially with sandy, alluvial or clay soils. Similar members of the Cyperaceae family have reported high content of secondary metabolites like alkaloids, phenols, tannins, saponins and flavonoids in them which makes them an important part of the diet of grazing and semi aquatic animals linking their phytochemistry directly to gut health (Roy and Devi, 2025). Currently, the overuse of antibiotics in the veterinary and livestock sectors has become one of the major reasons behind increasing antimicrobial resistance (Economou and Gousia, 2015). It not only promotes the emergence of resistant microbial strains but also disrupts the natural gut microflora. Therefore, alternative plant-based solutions, when

combined with existing veterinary practices, may offer a safer and more cost-effective approach. In Indian folk medicine practices, the powdered roots of *C. compressus* have been used orally in the treatment of helminthiasis by the Santhal tribes (Soren *et al.*, 2020; Taheri *et al.*, 2021), pointing toward long standing folk practices that remain



Figure 1: inflorescence of *Cyperus compressus*

largely unexplored today. Present study was therefore undertaken to profile the phytochemical constituents of *C. compressus* and to evaluate its antibacterial

activity against *E. coli*, to validate its traditional uses and generate baseline data for its potential application in veterinary health and livestock management.

METHODOLOGY

Whole plants of *Cyperus compressus* (Figure 1) were collected from riverside areas of Mahanadi River of Cuttack, Odisha, India and identified based on using standard flora (Saxena and Brahman, 1996). The collected samples were thoroughly washed under running tap water to remove debris and surface contaminants. The cleaned plant materials were shade dried at room temperature for a week to avoid thermal degradation of sensitive compounds. The dried material (Figure 2) was coarsely powdered using a mechanical grinder and stored in sterile, airtight container until further use.



Figure 2: Dried plant parts of *C. compressus*

Then, powdered samples were subjected to solvent extraction using three different solvents of varying polarity: n-hexane (non-polar), ethanol (polar), and aqueous (highly polar). For each solvent, 10 g of plant powder was macerated in 100 mL of solvent for 48 hours with occasional shaking (Devi *et al.*, 2023). The extracts were filtered through Whatman no. 1 filter paper and filtrates were concentrated by air drying at room temperature. Dried crude extracts were stored in airtight glass vials at 4 °C until further analysis. Preliminary phytochemical analysis of *C. compressus* extracts was carried out to detect the presence of primary and secondary metabolites. Standard phytochemical procedures (Jena *et al.*, 2025) were followed to qualitatively screen the bioactive compound. The antibacterial activity of the extracts was assessed against *Escherichia coli* (MTCC 45) using a broth micro dilution approach in accordance with recognized standards (Jena *et al.*, 2025). Briefly, standardized inocula of *E. coli* were exposed to a range of extract concentrations (100, 50, 25 and 12.5) mg/mL and incubated under ideal growth conditions. Appropriate controls were included to validate the assay. After incubation, bacterial growth was evaluated and the Minimum Inhibitory Concentration (MIC) was defined as the lowest concentration of extract that prevented visible microbial growth.

RESULTS AND DISCUSSION

Phytochemical Screening: The presence of bioactive compounds showed solvent dependent distribution. Phytochemical screening for primary metabolites detected protein and carbohydrates in both ethanolic and aqueous extract. Starch was only detected in the ethanolic extract whereas none of the tested bioactive compounds were found to be present in the n-hexane (non-polar) extract. A similar trend was observed in case of qualitative secondary metabolite screening which revealed no active compounds in the non-polar extract whereas, tannin and reducing sugars were detected in both the polar extracts (aqueous and ethanol). These observations showed that polar solvent was phytochemically rich as it contained comparatively diverse array of bioactive compounds in comparison to the non-polar solvent which was screened to be inert (Table 1-2).

Antibacterial assay: Hydroethanolic (1:1) extract of the *C. compressus* was tested against *E. coli* to estimate its Minimum Inhibitory Concentration (MIC). Four different concentrations (100, 50, 25 and 12.5) mg/mL were tested out of which, bacterial inhibition was observed at 25 mg/mL showcasing MIC. Microbial broth

showed visible microbial growth while nutrient broth and positive control (Kanamycin) showcased clear inhibition (Table 3).

Table 1: Qualitative phytochemical analysis for detection of primary metabolites

Polarity	Extract	Detected primary metabolites
Non-Polar  Polar	n-Hexane	No tested metabolites were detected
	Ethanol	Protein, Starch and Carbohydrates
	Aqueous	Protein and Carbohydrates

Table 2: Qualitative phytochemical analysis for detection of secondary metabolites

Polarity	Extract	Detected secondary metabolites
Non-Polar  Polar	n-Hexane	No tested metabolites were detected
	Ethanol	Tannin and Reducing sugars
	Aqueous	

Table 3: Estimation of Minimum Inhibitory Concentration (MIC)

Hydroethanolic Extract	Concentration (mg/mL)				MIC (mg/mL)
	100	50	25	12.5	
<i>C. compressus</i>	NG	NG	NG	G	25
Nutrient broth	NG	NG	NG	NG	-
Microbial broth	G	G	G	G	-
Antibiotics (Kanamycin)	NG	NG	NG	NG	-

(NG: No growth of bacteria; G: Growth)

Ecological link: Grazing herbivores regularly come in contact with sedges and often rely on them as a part of their natural diet in wetland and grassland ecosystems (Elliott and Henry, 2011). This suggests that their gut health and microbial balance are directly influenced by the plants they consume. The observed antibacterial activity of *C. compressus* against *E. coli* therefore highlights its possible role in supporting gut health in grazing animals in a natural and preventive manner.

CONCLUSION

This study sheds light on the hidden potential of commonly overlooked sedge, revealing how something so ordinary in appearance can hold remarkable biological value. The selective extraction of compounds using polar solvents highlighted the phytochemical richness of *C. compressus*, while its ability to inhibit *E. coli* growth adds scientific support to the traditional medicinal practices. These findings suggested that this plant is not just a component of wetland vegetation, but a silent contributor to natural health regulation in ecosystems where grazing animals constantly interact with it. In an age where antibiotic resistance is becoming a global concern, the relevance of such plant-based alternatives becomes even more significant. The relationship between sedges, grazing herbivores, and gut microbial balance opens a window into nature's self-sustaining design. What was once regarded as a humble grass like plant now emerges as a promising candidate for further exploration in veterinary health and sustainable disease management. By looking back at nature and indigenous knowledge, we may discover solutions for the challenges of the future, solutions that are rooted not in laboratories alone, but in living landscapes.

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