SHORT COMMUNICATION

GC/MS profiling, in vitro anti-leptospiral and haemolytic activities of Boesenbergia rotunda (L.) Mansf. used as a medicinal plant by Nicobarese of Andaman and Nicobar Islands

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Leaves of the plant Boesenbergia rotunda are used by the Nicobarese tribe of Andaman and Nicobar Islands, India, to prepare traditional medicine for treating fever, headache and body ache. In the present investigation, methanol fraction of these leaves were analysed by GC/MS that revealed the presence of 25 compounds. The anti-leptospiral activity of methanol crude extract was determined by both microdilution and macrodilution methods. The MICs of the extract were tested against 24 pathogenic leptospiral strains and ranged between 62.5–125 μg/mL in both microdilution and macrodilution. The range of MBCs was 250 and 500 μg/mL in macrodilution and microdilution respectively. The crude extract was subjected to cytotoxic studies and found to have negligible or no haemolytic activity, exhibiting IC50 values of greater than 4 mg/mL. Further in vivo studies are needed to investigate the pharmacological and toxicological properties of Boesenbergia rotunda, before it can be considered as a new anti-leptospiral agent.

Keywords: Boesenbergia rotunda; anti-leptospiral activity; Car Nicobar Island; Nicobarese

1 Introduction

Leptospirosis is a zoonosis of worldwide distribution, caused by spirochetes, belonging to the genus Leptospira. Leptospires are highly diverse, both antigenically and genetically, with more than 260 serovars belonging to 24 serogroups and about 17 species. Clinical manifestations ranged from asymptomatic (recognizable only by seroconversion without illness) to severe, potentially fatal infections that involves renal, liver and/or respiratory complications. The case fatality ratio ranged from 3% to 70% (Gouveia et al. 2008). As the rural communities lacked modern medical facilities, they utilized the local plants in their surroundings to treat various
illnesses (Tabuti et al. 2003). Traditional medicines can provide a valuable lead in the discovery of new and more efficacious drugs. Till date, very few of these are utilized in drug and pharmaceutical industries (Verma et al. 2007). Many plant species prove to be effective against various pathogens in vitro, even though the data regarding the efficacy of antibiotics or natural pharma against *Leptospira* seem to be very scanty. In the present study, antimicrobial efficacy of *Boesenbergia rotunda* (L.) Mansf. used as a medicinal plant by Nicobarese of Andaman and Nicobar Islands was tested against diverse species of leptospires.

## 2 Results and discussion

### 2.1 Anti-leptospiral acitivity

Currently, very few reports are available regarding the natural products used against leptospires (Vedhagiri et al. 2009; Chander, Kumar, et al. 2014). In the present study, methanol extract of *B. rotunda* leaves were subjected to percolation with methanol as a solvent. The resultant crude extract was utilized in the study of anti-leptospiral activity against 24 pathogenic strains. The MICs and MBCs obtained by microdilution and macrodilution method among the serovars tested are provided in Table S1. Methanol crude extract of *B. rotunda* showed an MIC$_{90}$ of 62.5–125 µg/mL in both microdilution and macrodilution, while MBC$_{90}$ was 500 and 250 µg/mL and respectively. MIC was higher for the tested plant extract compared to the standard antibiotics. However, when tested for its toxic parameter, it showed negligible haemolytic activity (greater than 4 mg/mL). These results were similar, when compared to other natural pharmacological studies carried out against *Leptospira* (Prabhu et al. 2008; Vedhagiri et al. 2009; Chander, Kumar, et al. 2014).

### 2.2 Preliminary phytochemical analysis

Freshly prepared extracts were subjected to preliminary phytochemical screening for various constituents. The results revealed the presence of alkaloids, tannins, sterols, triterpenes and flavonoids.

### 2.3 Chemical composition of methanol extracts

The GC/MS analysis of methanol extracts enabled in identifying 25 components (Table S2 and Figure S1). This volatile fraction contained a mixture of compounds from different classes. Compounds were identified on the basis of retention time, Kovats indices, fragmentation patterns and data comparison along with NIST mass spectral library, thus providing a complete structural identification.

## 3 Conclusions

Our earlier study on *Glyptopetalum calocarpum*, an endemic plant used by Nicobarese, prove to be a potential antibacterial agent against *Leptospira* and other pathogenic *Staphylococcus epidermidis, Staphylococcus aureus* and *Pseudomonas aeruginosa* (Chander, Kumar, et al. 2014, Chander, Srivani, et al. 2014). As time passed, indigenous communities learned to use the locally available plants in the treatment of various infections and diseases. This study demonstrates the anti-leptospiral efficacy of *B. rotunda*, a plant that is commonly used by Nicobarese as an herbal remedy. The plant extract was observed to have higher MIC values than the standard antibiotics. The GC/MS analysis of methanol extracts revealed few major constituents. These could be purified further and tested in an animal model to determine their
activity on Leptospires. The in vivo activity in animal models could facilitate the selection of agents for human trials.

**Supplementary material**
Experimental details relating to this paper are available online, along with Tables S1–S2 and Figure S1.

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**Disclosure statement**
No potential conflict of interest was reported by the authors.

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