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Molecular characterization of microsymbionts associated with root nodules of *Crotalaria burhia* Buch.-Ham. ex Benth., a native keystone legume species from Thar Desert of India

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Establishment of legume-rhizobia symbiosis has ample agronomic and ecological significance. Characterization of native rhizobia could enhance our understanding of their natural distribution and co-evolution. The Great Indian Thar Desert is an ecologically significant unique habitat with its flora and fauna. *Crotalaria* spp. is an economically important legume widely distributed in the Thar Desert and can be considered its one of the bioresources, particularly for biological nitrogen fixation with their symbiotic rhizobia. Here, we examined the legume *Crotalaria burhia* Buch.-Ham. ex Benth. in search of potential novel rhizobial species. Out of 72 root nodule bacterial (RNB) strains isolated from *C. burhia*, 51 rhizobia-like strains were examined for genetic diversity based on ARDRA and RAPD patterns. BLASTn sequence similarity results based on 16S rRNA gene of selective thirteen strains representing four ARDRA types revealed that they were related to genera *Ensifer*, *Rhizobium* and *Bradyrhizobium*. In 16S rRNA gene phylogeny, five (CB5, CB17, CB36, CB44, CB56) strains were closer to *Ensifer kostiensis*, three (CB6, CB12, CB32) to *E. terangae* and CB11 showed similarity with *E. kostiensis* and *E. saheli*. Strain CB4 was similar to *Bradyrhizobium yuanmingense* and three (CB29, CB31, CB46) strains were closer to species of *Rhizobium* (*R. etli*, *R. sullae* and *R. borbori* respectively). Symbiotic (*nodA* and *nifH*) genes phylogeny of *Ensifer* sp. CB56 was incongruent and showed close similarity with *E. fredii* whereas *sym* gene phylogeny of *Bradyrhizobium* sp. CB4 was congruent with 16S rRNA gene phylogeny. In *Rhizobium* strains *sym* genes could not be amplified and they failed to nodulate host. Our study suggests that *C. burhia* is nodulated by diverse strains of *Ensifer* and *Bradyrhizobium* in alkaline soil of Thar Desert and these strains effectively cross-nodulated crop *Vigna radiata*.

Keywords: ARDRA, *Bradyrhizobium*, Burhia Rattlepod, *Ensifer*, *nifH*, Nitrogen fixation, *nodA*, Nodulation, Root nodule bacterial (RNB) strains