

## ABSTRACT

### Studies on Acid/Alkali Production and Tolerance to Soil Acidity by Cowpea Rhizobia

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The growth and acid/alkali production by cowpea rhizobia strains IRC256 and IRC291 were examined by growing rhizobia in media containing various combinations of nitrogen (urea, ammonium sulphate, yeast extract or sodium nitrate) and carbon sources (glucose, mannose, ribose or mannitol). While growth of cowpea rhizobia in medium containing ammonium sulphate always caused an acid pH change, growth in medium containing urea mostly caused an alkali pH change. When sodium nitrate and yeast extract were used as nitrogen source, cowpea rhizobia showed variable pH changes dependent on the carbon source used. As a carbon source, ribose often caused an acidic pH change irrespective of nitrogen source.

When cowpea rhizobia grown in medium (containing ammonium sulphate) until the pH became acidic (3.3) were transferred to fresh medium (containing ammonium sulphate) pre-adjusted to acidic pH (3.3), neither any growth nor a further pH change was observed. Similarly, when rhizobia grown in medium (containing urea) until

pH became alkali (pH 8.0) was transferred to fresh medium (containing urea) pre-adjusted to alkali pH (8.0), only little growth but no further pH change was observed. On the other hand, when rhizobial culture grown in medium (containing ammonium sulphate) until pH becomes acidic (pH 3.3) was transferred to fresh medium (containing urea as nitrogen source instead of ammonium sulphate) pre-adjusted to acidic pH (3.3) immediate growth and pH increase were observed.

When ammonia was assayed in the supernatant of rhizobial culture after their growth, it was seen that cultures grown in medium containing urea accumulated  $\text{NH}_4^+$  while cultures grown in medium containing ammonium sulphate consumed  $\text{NH}_4^+$ . However, there was no significant ammonia accumulation in culture grown in medium with yeast extract or sodium nitrate as nitrogen source.

Cultures of IRC256 and IRC291, when grown in medium containing ammonium sulphate, showed reduced oxygen consumption and activities of key metabolic enzymes when compared with rhizobial cultures grown in YEMB. Oxygen consumption by rhizobia grown in culture medium containing urea, however, was in general, higher than for cultures grown in medium containing yeast extract or ammonium sulphate.

While both strains were able to survive for 28 days in broth culture at pH 3.3, they showed poor survival in an acidic soil (pH 4.0). Thirteen strains of cowpea rhizobia studied showed tolerance to both low pH (4.5) and low phosphate (10  $\mu$ M) as opposed to four fast growing rhizobial strains. Tolerance to Al and Mn among cowpea rhizobia strains showed variation. In general, strains isolated from Jamaica (JRC strains) were more sensitive to both, than strains isolated from West Africa (IRC strains). Calcium, and to a much lesser extent, urea were able to ameliorate the toxic effects of Mn and Al. Six cowpea rhizobia strains tested were able to nodulate cowpea in nutrient solution at pH 4.5, but only three could nodulate cowpea with the added stress of the addition of 200  $\mu$ M Al.