

ical composition (total protein, amino acid profile, crude fibre, and carbohydrate, etc.) of green pods, and dry seeds will also be studied.

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USE OF A MODIFIED GIBSON'S TUBE METHOD  
FOR ASSESSMENT OF NODULATION IN BEANS

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In our Soil Microbiology Section at CENA, we have developed a new technique to test bean plants in sterile conditions mainly for nodulation, which was partly modified after orientation by Emeritus Professor J. M. Vincent, an expert of IAEA who spent two months at our institution.

This method is a modification of the Gibson's partly enclosed seedling method (Gibson, 1963), as the bean is a big plant and needs larger containers for its N<sub>2</sub>-fixation tests. The test is as follows:

- a. Introduce a thin glass tube (3 x 35mm) into an empty test tube (30 x 300 mm). (The thin tube is used for watering.)
- b. Fill the big tube with washed and oven-dry vermiculite until 5/6 of its portion.
- c. Add 60 ml of seedling nutrient solution minus N (McKnight, 1949). The solution's level must be at 1/3 of total volume of the tube.
- d. Put a cotton wool plug in the entrance of the thin tube and cover it with an aluminum cap.
- e. Wrap the upper part with aluminum foil.
- f. Autoclave the tubes.
- g. Sterilize seeds (Vincent, 1970) and put two seeds per tube, cover with a thin layer of sterile sand (5 mm depth).
- h. Thin to one plant per tube after germination (3-5 days later) and inoculate the plants.
- i. Cover with another 1.5 cm depth sterile sand.
- j. Avoid direct illumination of roots by wrapping each tube with aluminum foil.
- k. When necessary, refill the tube with sterile tap water to maintain the solution's level.

Figure 1 shows the assembly and the watering bottle.

The technique was also tested for nodulation with Glycine max and Vigna sinensis and can be recommended for short-term experiments, as in all tests the following points were taken into account: freedom from nodules in

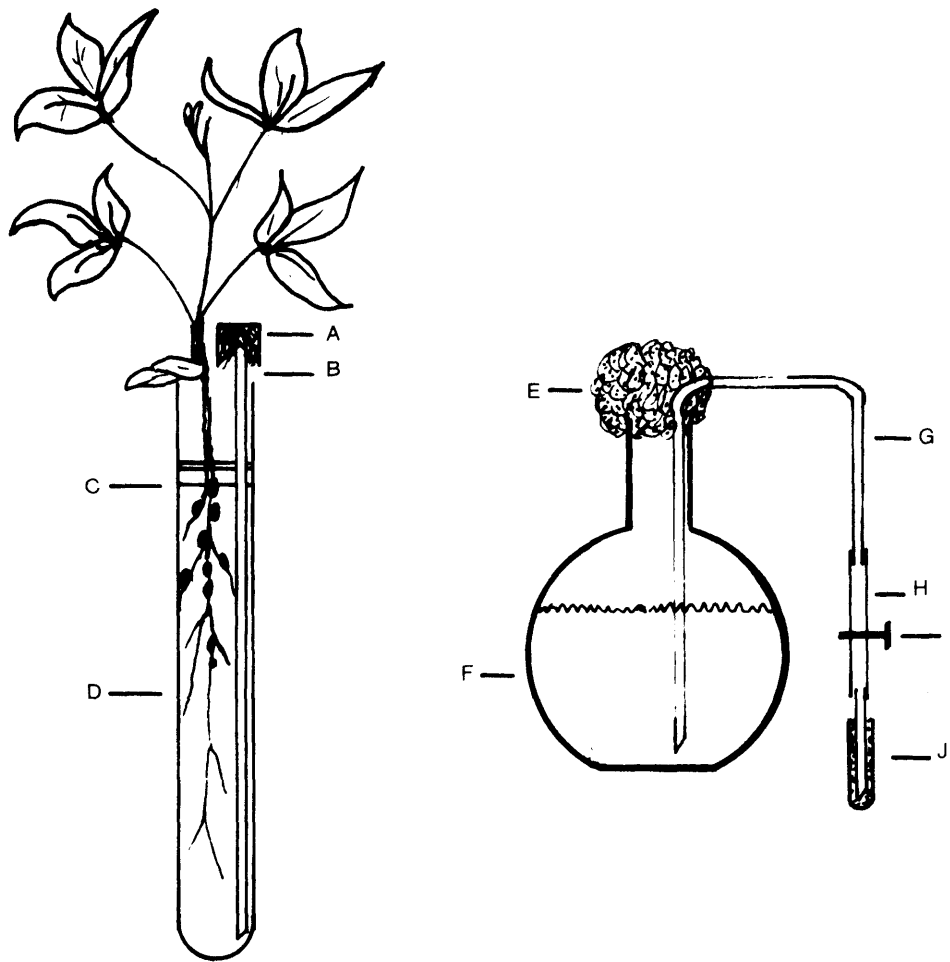


Figure 1

- A. Cotton plug covered with aluminum cap for watering
- B. Thin glass tube
- C. Sterile sand
- D. Vermiculite plus seedling nutrient solution
- E. Cotton plug
- F. Sterile tap water
- G. Thin glass tube (syphon)
- H. Autoclaveable latex tube
- I. Tubing clamp
- J. Glass tip covered with small test tube (cotton wool inside)

uninoculated controls, reliability of nodule formation, reasonable reproductibility of measured nitrogenase activity after 3 weeks growth and reliability as a predictor of the effectiveness of the association.

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### EFFICIENCY OF N<sub>2</sub>-FIXATION OF R. phaseoli STRAINS INOCULATED IN THE BEAN CULTIVAR VENEZUELA-350

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A comparison of the N<sub>2</sub>-fixation abilities of 18 R. phaseoli strains in the bean cultivar Venezuela-350 was made under aseptic conditions in a greenhouse (12 hours of light, 22°-28°C). The origins of the strains are listed in Table I, all of them being previously tested for purity and effectiveness for nodulation. The experiment was divided into two parts and strain 127-K17 (Nitragin Co., U.S.A.) was used as control. Tables II and III show the inoculation effect on the weight and total N of the plants and in the nodulation (weight and nitrogenase-ethylene-activities).

Strains C-01 and C-19 were superior to the others in increased weight and total N of the plants. Plants inoculated with 127-K17 presented good nodulation and N<sub>2</sub>-activity (Table II), comparable to plants inoculated with C-05 and C-23 (Table III). At this growth stage (5-6 weeks after germination), the nodular mass is only a reasonable predictor of N<sub>2</sub>-fixation efficiency (Saito and Cardoso, 1977) and in some cases, mostly in the intermediately nodulated plants, the contribution of N from N<sub>2</sub>-fixation is not apparent. Table III shows a good correlation between total N x N<sub>2</sub>-ase activities.

Strains CIAT-57, C-12, C-13, F-300, F-310 and CIAT-351 did not cause a significant increase in plant nitrogen.

All the other strains are intermediate for N<sub>2</sub>-fixation, as compared with the uninoculated plants.

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