# Observations on the use of *Sesbania rostrata* as green manure in paddy fields

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#### Introduction

Nitrogen is one of the two major factors governing plant productivity, the other being water. The nitrogen requirements of plants are considerable: for example, in order to produce 100 kg of grain, rice requires 1.8-2 kg of nitrogen regardless of the type of soil and the date of planting (Patnaik & Rao 1979). A harvest of 4 t of rice per hectare requires 80 kg of nitrogen. Nitrogen absorbed by the plant comes from three sources: reserves of soil nitrogen (organic matter nitrogen and inorganic soil nitrogen), fertilizers supplied by the farmer, and biological nitrogen fixation. Because soil nitrogen reserves are limited, however, it is necessary to use fertilizers which are expensive, as energy requirements for their production are high (c. 1600 l natural gas is consumed in the synthesis of 1 kg of anhydrous ammonia). Moreover, imported fertilizers must often be purchased with hard currency. Because biological nitrogen fixation presents an appealing alternative to fertilizers, much work has been devoted to exploiting this process.

One solution that has been used for many centuries involves incorporating the shoots of nitrogen-fixing plants into the soil, so that the plant residues serve as an organic nitrogen fertilizer; hence the term 'green manure'. For several centuries many nitrogen-fixing plants have been used as green manures, notably in the Far East. Legumes, such as Astragalus sinicus, Sesbania aculeatea, S. paludosa, S. cannabina, Crotalaria juncea, C. striata, Vicia cracca, Medicago hispida, M. officinalis (Vachhani & Murty 1964; Patnaik & Rao 1979; Watanabe & App 1979; Allen & Allen 1981), or non-legumes, such as Coriaria sinica (Watanabe, pers. comm.) have all been used in this way. The plant that has probably been most often used as a green manure is A. sinicus, which is cultivated during the winter and incorporated into the soil 20 days before rice is planted. Incorporation of plant residues amounts to 20-30 t of fresh weight per hectare, i.e. 100 kg of nitrogen per hectare. Progress has recently been made in this field in Senegal with the discovery of S. rostrata, a stem-nodulated legume of the Papilionoidae (Dreyfus 1982). An advantage of this legume is that, because it nodulates both on roots and stems, it fixes nitrogen more actively than other known legumes.

In this review paper, we begin by presenting the state of the art of the biology of S. rostrata. Next, we present the preliminary results of experiments in the use of S. rostrata as green manure in paddy fields. Finally, we propose guidelines for field trials aimed at introducing S. rostrata into tropical agriculture.

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#### Biology of the nitrogen-fixing symbiosis S. rostrata-Rhizobium

The most distinctive characteristic of *S. rostrata* (Figs 1 and 2) is the presence of pre-determined nodulation sites on the stems. These sites resemble small points arranged in vertical lines extending up the stem. When infected by specific strains of *Rhizobium*, these sites form nitrogen-fixing nodules, known as stem nodules.

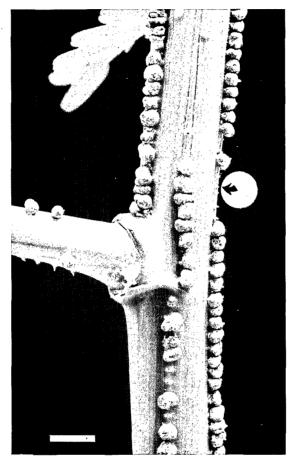


Fig. 1 Stem of *S. rostrata* exhibiting three rows of stem nodules. On the right a few nodulation sites (arrow), resembling small points, that have not been infected by the specific *Rhizobium*. Bar represents 1 cm.

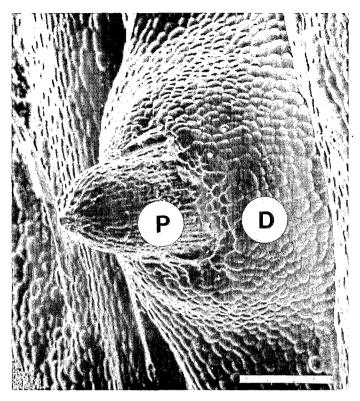
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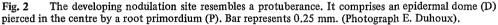
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#### Climate and soil requirements of S. rostrata

Sesbania rostrata is a wild plant that has not yet been selected for plant breeding studies. Climate and soil requirements must be taken into consideration in its cultivation.

Climate requirements. At the latitude of Senegal  $(15^{\circ}N)$ , S. rostrata grows well in waterlogged soils during the rainy season (June to September); it usually behaves as an annual plant and flowers in the second or third month of its growth cycle. Stem nodulation occurs rapidly (one week after inoculation) and is regular and profuse. In contrast, at the same latitude during the dry and cold season (December to March), growth is poor, flowering takes places abnormally early, and nodulation is difficult. This variation in the behaviour of S. rostrata during the different seasons can be attributed to its sensitivity to photoperiod, temperature, and humidity.





Sesbania rostrata is characterized by having a relatively long photoperiod demand (13-14 h), so that it grows poorly and flowers readily when the day length is too short. Until non-photoperiodic cultivars of S. rostrata are available, this requirement for relatively long days is probably a limiting factor to the use of this plant in equatorial climates.

Soil requirements. Sesbania rostrata grows well only in low lying land or in waterlogged or heavily irrigated soils. Apparently it tolerates a certain level of soil salinity, but the salinity threshold has not yet been determined. In wet but well-drained soils, S. rostrata is particularly susceptible to attack by nematodes, especially *Meloidogyne* sp.

#### Nodulation sites on the stems

As already indicated, nodulation sites resembling small points are distributed evenly in three or four vertical lines along the stem (Fig. 1). Formation of these sites is pre-determined and independent of the *Rhizobium* infection. A developing nodulation site appears as a protuberance on the stem. A day or two later, this protuberance swells and becomes an epidermal dome pierced in the centre by a root primordium with a length of 0.2-0.8 mm (Fig. 2).

The root primordium remains inactive on the stem until its base is infected by the specific *Rhizobium* strain, when it produces a nitrogen-fixing nodule.

Nodulation sites exhibit two unique characteristics: (1) they are pre-formed and their exact location is known in advance; (2) they are formed continuously throughout the growth of the stem and remain sensitive to *Rhizobium* infection throughout the life of the plant. Thus nodule formation can occur at any time during the growth cycle.

#### Double Nodulation of S. rostrata

Sesbania rostrata can bear nodules on roots and stems at the same time.

When grown in non-inundated soil, the young S. rostrata plant (15 days to one month old) displays two types of root nodules. At the level of the crown and at the base of the main (tap) root a cluster of large, elongated nodules appears with an apical meristem. These nodules have two or three lobes and measure from 0.2-1.5 cm in length. Another type of root nodule appears like beads on a string on lateral roots. These nodules are spherical, small (1-2 mm in diameter) and numerous. When the plant grows on inundated soil, the large nodules from the crown disappear rapidly and only those nodules located on the surface of the lateral roots remain. These roots float in water and bear many nodules with a green cortex. The total weight of these nodules remain small, (2-4 g) compared with that of the stem nodules (15-40 g).

In nature, stem nodules often appear spontaneously over the whole length of the plant, including lateral branches. Distribution of these stem nodules is often irregular, however. Dust seems to play a significant role in spontaneous nodulation of stems, as plants that grow along dirt roads are generally better nodulated than other plants. Other vectors that may be significant include insects and rain. Not all nodulation sites are spontaneously infected, and the number and weight of stem nodules may vary from one plant to another. Some nodules are spherical, while others are irregular. The red central zone is surrounded by the green cortex. Diameter of the nodules ranges from 0.3-0.8 cm, and a mature plant of 3-4 m high can bear nodules weighing up to a total of 40 g (fresh weight).

#### Genesis of stem nodules

Rhizobia never infect the nodulation sites from inside; they reach the root primordium through the fissure encircling it (Fig. 2). Nodule genesis consists of three distinct stages: 1. Development of intercellular pockets of infection and dedifferenciation of the meristematic zone. Initially rhizobia become attached to the surface of the cells located at the base of the root primordium in the circular fissure. Later, they penetrate the intercellular space when they multiply in large numbers, forming pockets of infection (Fig. 4). At the same time, a differentiation of some cortex cells of the root primordium occurs through an activating mechanism which is still not known. The resulting meristematic cells are then infected by the rhizobia.

2. Development of infection threads. Infection threads originate from the intercellular pockets of infection (Fig. 3). These threads divide into several branches that penetrate the meristematic cells.

3. Intracellular infection. Rhizobia are released from the infection threads into the cell cytoplasm five or six days after infection starts (Fig. 3). When enclosed in the peribacteroid membrane (Fig. 4), the rhizobia begin to fix nitrogen symbiotically. When the rhizobia have reached this stage (known as the bacteroid stage), the nodules exhibit the characteristic red colour of leghaemoglobin, and they begin fixing nitrogen.

The mode of infection of *S. rostrata* stems is thus unique among the legumes, as it involves both an intercellular invasion by rhizobia, as in peanuts, and the development of infection threads, as in temperate legumes, although in contrast with the latter, however, infection in *S. rostrata* does not occur through root hairs.

#### Rhizobium strains of S. rostrata

Two types of strains have been isolated: stem strains, capable of nodulating both stems and roots, and root strains, which nodulate roots only. Although the stem strains (type ORS571)



Fig. 3 Multiplication of rhizobia in intercellular spaces at the base of the root primordium three days after inoculation. Bar represents  $12 \ \mu m$  (Tsien *et al.* 1983).

are fast-growing, they differ from temperate fast-growing and tropical slow-growing rhizobia. Thus they probably constitute a new species.

A physiological study showed that stem strain ORS571 is able to grow with atmospheric nitrogen as the sole nitrogen source (Dreyfus *et al.* 1983). This property, unique among all other known rhizobia, has allowed us to obtain  $nif^-$  mutants of strain ORS571 by using standard genetic methods. Nitrogen fixation genes of the wild strains have been cloned in a plasmid. Following conjugation and transfer of this plasmid to a  $nif^-$  mutant, genetic complementation was observed both *ex planta* and *in planta* (Elmerich *et al.* 1982).

#### Medium for growing strain ORS571

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The liquid nutrient medium for the stem strain ORS571 is composed as follows: Na lactate, 10 g;  $(NH_4)_2SO_4$ , 1 g;  $K_2HPO_4$ , 1.67 g;  $KH_2PO_4$ , 0.87 g; MgSO\_4, 0.2 g NaCl, 0.1 g FeCl<sub>3</sub>, 4 mg; yeast extract, 1 g. The pH is adjusted to 6.8 and the medium is autoclaved at 120°C for 20 min. The same medium is used with agar for cultures in Petri dishes.

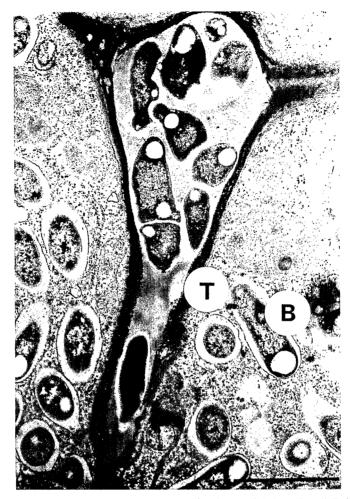


Fig. 4 Extremity of an infection thread (T) with a *Rhizobium* cell being released. Released *Rhizobium* cells (B) are enclosed individually within the peribacteroid membrane. Bar represents 3  $\mu$ m (Tsien, unpubl. photo.)

#### Inoculation

As we have indicated above spontaneous inoculation of stems is always irregular. Thus, inoculation of stems is generally recommended, as is root inoculation although the latter is not necessary when soils already harbour native rhizobia capable of nodulating *S. rostrata*. We recommend strain 0RS571, which is both competitive and effective in fixing nitrogen.

Seeds are inoculated with a culture that is either liquid or adsorbed on a carrier such as peat (Vincent 1970) or entrapped in a polymer (Jung *et al.* 1982). Seed inoculation induces complete root nodulation but only partial stem nodulation. Thus, the shoots must also be inoculated in order to obtain satisfactory stem nodulation. Either of the following methods can be used: (1) Spraying liquid culture on the shoots. When S. rostrata plants have attained a height of 50-80 cm, a culture of the strain ORS571 containing c.  $10^8$  bacteria/ml is sprayed on the shoots using a standard sprayer. (2) Spraying colloidal solution of rhizobia in a polymer (alginate type). The inoculum made of rhizobia cells entrapped in a polymer and dried

as a powder (Jung *et al.* 1982) is mixed with water to obtain a colloidal solution, which is then sprayed on the shoots of *S. rostrata*. An insecticide can be added to the colloidal solution to protect the plants from insects.<sup>1</sup>

#### Nitrogen fixation

Nitrogen fixation by stem nodules of S. rostrata exhibits two unique properties. The first is that the plant has a high nitrogen-fixing potential. Acetylene reduction takes place at about 600  $\mu$ mol C<sub>2</sub>H<sub>4</sub>/h/plant, a much higher rate than that of soybean, which ranges from 14-120  $\mu$ mol C<sub>2</sub>H<sub>4</sub>/h/plant. When the difference method and the nitrogen balance method are used, nitrogen fixation by S. rostrata has been shown to be about 200 kg N<sub>2</sub>/ha in 50 days, which indicates that this legume is one of the most powerful of nitrogen-fixing systems (Rinaudo et al. 1982a, b). The second unique property of S. rostrata is its ability to nodulate and fix nitrogen even when the rates of combined nitrogen in the soil are high (c. 200 kg N/ha). Thus S. rostrata is capable of assimilating both soil and atmospheric nitrogen, which constitutes a significant advantage (Dreyfus & Dommergues 1980).

#### Seed production and seed treatment

In Senegal S. rostrata produces seeds in abundance as soon as the days become shorter (September, October). It is advisable to harvest the seeds as soon as they mature and to treat them with insecticide.

As for many tropical legumes, S. rostrata seeds germinate rapidly and homogeneously only when they are treated chemically or scarified in a mortar. Chemical treatment consists of submerging seeds in concentrated sulphuric acid for 30 min and then rinsing them quickly in a large amount of water. Seeds can also be scarified by mixing them with an equivalent quantity of siliceous coarse sand and then pouring this mixture into a mortar and delicately rotating the pestle in a scraping motion until the grains become pock-marked. The seeding density is usually 100 seeds  $(2 \text{ g})/\text{m}^2$  or 20 kg/ha.

#### Pests

Pests that attack S. rostrata are not well known. However, we have observed that young plants can be attacked by a black aphid, flowers can be eaten by a beetle, and seeds can be destroyed by a fly. The destructive effect of these pests is greatly reduced in the summer, when environmental conditions are more favourable to growth of the plants.

#### Preliminary findings on the use of S. rostrata as green manure in paddy fields

Seeds of S. rostrata were sown in rows in wet soil. When the plants had attained a height of 30 cm, the paddy fields were flooded. Stems were then inoculated by spraying with a culture of the specific *Rhizobium* strain 20-30 days after sowing. When the plants had attained a height of 1.5 m (six to seven weeks after sowing), the paddy field was drained and the plants uprooted and chopped into pieces 10-20 cm long which were then incorporated into the soil at a depth of 10-15 cm. In the first trials (1 m<sup>2</sup> microplots), rice was planted two weeks after incorporation of S. rostrata into the soil. Later trials (25 m<sup>2</sup> plots) showed that rice could be planted immediately after incorporation of the legume, significantly reducing the duration of the Sesbania-rice sequence.

<sup>1</sup> Patent pending.

Based on microplot trials conducted at the Bel Air ORSTOM Station in Dakar in the summer of 1980, the use of *S. rostrata* as green manure doubled rice yields. These results were later confirmed in trials conducted at the ISRA station of Djibelor on  $25 \text{ m}^2$  plots.

#### Microplot trials at the Bel Air ORSTOM Station (Table 1)

Table 1 E	Effect of S.	rostrata green	mature or	ı yield ar	nd nitrogen a	nd content of rice.
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Treatments	Dry weig	ht (g/m <sup>2</sup> )	Nitrogen content (%)		
	Grain	Straw	Grain	Straw	
S. rostrata green manure	584 a	767 a	1.74 a	0.96 a	
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> *	381 b	484 b	1.27 b	0.49 b	
Control	212 c	276 с	1.14 b	0.58 b	

Numbers with the same letters in columns do not differ significantly, P = 0.05. \* 60 kg N/ha

Yields of dry matter (grain and straw) were more than double those of controls in plots where S. rostrata green manure was used. The effect of inorganic nitrogen fertilization was significantly less pronounced than that of S. rostrata green manure. Moreover, the nitrogen content of grain and straw in the S. rostrata plots was significantly higher by (50%) than that of control plots.

Rice was again planted on 1 m<sup>2</sup>. plots the following year, without using *S. rostrata* green manure. The residual effect of the green manure from the preceding year resulted in a 50% yield increase over control plots (328 and 230 g/m<sup>2</sup>, respectively).

#### Trials on 25 $m^2$ plots at the ISRA Station at Djibelor (Table 2)

 Table 2
 Effect of S. rostrata green manure and application of organic matter on grain yields of rice

Treatments	Grain yield (kg/25m <sup>2</sup> )			
	1981	1982		
S. rostrata green manure	7.30	11.25		
Organic matter	3.44	7.30		
Control	2.99	5.75		

Note: Each figure represents the average of six replications.

Results obtained at the ISRA Station at Djibelor in the summers of 1981 and 1982 showed that the use of *S. rostrata* green manure doubled the grain yields in comparison with the control plots. The application of organic matter (compost + farmyard manure) to the soil had a less marked effect.

#### Cause of observed effects

The large input of nitrogen to the soil resulting from the incorporation of S. rostrata shoots is responsible for most of the increase in the rice yield. However, one should not overlook two secondary effects of this green manure.

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First, the green manure provides an input of 2 kg organic matter (dry weight)/ $m^2$  or 20 t/ha. For soils low in organic matter, such an input significantly improves soil structure.

Second, in some circumstances *S. rostrata* appears to limit the pathogenic nematode population. The rice cultivated after *S. rostrata* incorporation grows in an environment where the infestation rate is low enough to protect the crop against harmful precocious attacks by nematodes. It has also been suggested that *S. rostrata* acts as a plant trap for at least two nematode species (*Hirschmaniella oryzae* and *H. spinicaudata* (Germani *et al.* 1983; Germani, pers. comm.).

#### Guidelines for S. rostrata trials

We have seen that S. rostrata is a promising plant when used as green manure in Western Africa. We have no information, however, about the behaviour of this legume in other tropical countries, so it appears necessary to study this problem by conducting trials in two successive stages. The first stage would consist in determining whether it is possible to introduce S. rostrata in a given geographic region. If the results of these trials are positive, one should proceed to the second stage, which involves assessing the effect of S. rostrata green manure on rice yields under irrigated conditions.

#### Stage 1: Assessment of S. rostrata behaviour in a given geographic region

The first objective would be to determine whether S. rostrata thrives under the climatic and soil conditions of the region. If so, it would be necessary to determine the optimal time or times for planting in order to maximize biomass, nodulation, and seed production. Finally one should examine the problems posed by various pests.

For each of the soil types to be studied (e.g. soils with different levels of salt or organic matter content), a plot of  $2 \times 6$  m divided into four subplots of  $2 \times 1.5$  m should be delimited. *Sesbania rostrata* should be planted on each subplot at 3-month intervals (Fig. 5).

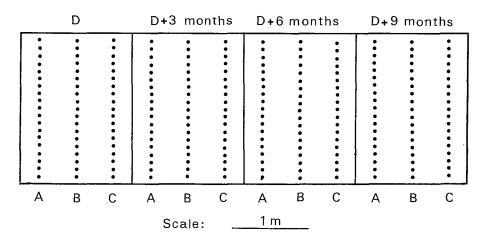


Fig. 5 Assessment of S. rostrata behaviour. For each of the soil types to be studied, a plot of  $2 \times 6$  m divided into four subplots of  $2 \times 1.5$  m should be delimited. Sesbania rostrata should be planted on each subplot at three-month intervals. In the first subplot, seeding should take place on D-day; in the second, on D + three months; in the third, D + six months; and in the fourth, D + nine months. Each subplot should contain three rows of S. rostrata (designated A, B, and C). Seeds should be spaced at 10 cm apart in each row, with a total of 19 seeds per row. The plants in row A should be cut 50 days, and plants in row C 100 days after sowing. Plants in row B should be maintained throughout the growth cycle so that the seeds can be harvested.

Each subplot would contain three rows of *S. rostrata* (designated A, B and C). Row A should be harvested 50 days and row C 100 days after seeding in order to assess biomass production. *Sesbania rostrata* in row B should be maintained throughout the growth cycle so that the seeds can be harvested. Because these trials are primarily exploratory, replications are not essential.

The soil should be ploughed and PK fertilizer applied at the standard rates used in the region. (For example, the following rates are used in Senegal: 30 kg P/ha; 8 kg K/ha). Before planting, the seeds should be treated as indicated earlier. They should then be inoculated according to the technique outlined above (this inoculation induces nodulation on the root system and at the bottom of the stem). Seeding density is shown in Fig. 5 (one seed every 10 cm on rows 50 cm apart). Sesbania rostrata should be seeded in wet, but not flooded, soil.

When the plants have attained a height of more than 30 cm, the soil should be flooded. When the plants have reached a height of 50-80 cm, shoots should be inoculated by spraying a culture of an appropriate strain of *Rhizobium* as indicated above.

Data on each experiment should include the following items:

1. Characteristics of the experimental site (latitude, longitude, soil type, climate).

2. Date of seeding.

3. Nodulation: date of stem inoculation; date of appearance of stem nodules; number and weight (fresh, dry) of stem nodules at harvest.

4. Biomass and N content of stem and leaves at harvest (that is 50 or 100 days after sowing for rows A and C); height of stems (if possible every two weeks and at harvest)

5. Flowering and fructification: date of beginning and end of flowering and seed maturation; weight of seeds (rows B).

6. Pests: dates and conditions of pest attacks; dates of treatments.

## Stage 2: Assessment of the effect of S. rostrata green manure on rice yields under irrigated conditions

The objectives are to assess the increase of rice yields (grain) resulting from the use of S. rostrata as green manure, to compare this response to that observed with three different levels of added N fertilizer (e.g. 50, 100, and 150 kg N/ha), and to assess the after-effects of the green manure.

The trial design should include the following three treatments:

Treatment S: S. rostrata green manure.

Treatment N: Nitrogen fertilizer at three levels.

Treatment T: Control.

The trial should be conducted using randomized blocks with five to six replications. The minimum surface of each plot should be  $20 \text{ m}^2$ .

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The PK fertilizer should be applied at the standard rates used in the region, as indicated earlier. For plots where treatment N is used, urea nitrogen fertilizer should be used, as other forms of fertilizer introduce other elements, such as S or P, which would distort the results. Application should be staggered.

When S. rostrata is 50 days old and has reached a height of 1.5 m, the paddy field should be drained. Plants should be uprooted or severed at soil level when the soil has been sufficiently drained (about five to seven days later). Plants should be chopped into segments 10-20 cm long and incorporated into the soil at a depth of 10-15 cm. While incorporation of S. rostrata into the soil has been carried out by hand up to now, it would be advantageous to mechanize this operation.

Rice can be planted shortly (within two weeks) after S. rostrata is incorporated into the

soil. Data collection should include items 1, 2, 3 and 6 already mentioned in the previous section. Other items should be:

7. Biomass and N content of stem and leaves; height of shoots measured before incorporation of S. rostrata into the soil.

8. Date of planting and harvest of rice; rice yield in kg grains/ha and percentage of nitrogen grains in the various treatments.

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#### References

ALLEN, O.N. & ALLEN, E.K. 1981 The Leguminosae. Madison: University of Wisconsin Press.

- DREYFUS, B. 1982 La symbiose entre *Rhizobium* et *Sesbania rostrata*, légumineuse à nodules caulinaires. Thèse de doctorat d'état, Université Paris VII.
- DREYFUS, B. & DOMMERGUES, Y.R. 1980 Non-inhibition de la fixation d'azote atmosphérique par l'azote combiné chez une légumineuse à nodules caulinaires, Sesbania rostrata. Comptes Rendus de l'Académie des Sciences, Paris, D 291, 767-770.
- DREYFUS, B. & DOMMERGUES, Y.R. 1981 Nitrogen-fixing nodules induced by *Rhizobium* on the stem of the tropical legume Sesbania rostrata. FEMS Microbiology Letters 10, 313-317.
- DREYFUS, B., ELMERICH, C. & DOMMERGUES, Y.R. 1983 Free-living Rhizobium strain to grow under N<sub>2</sub> as the sole nitrogen source. Applied and Environmental Microbiology 45, 711-713.
- DUHOUX, E. & DREYFUS, B. 1982 Nature des sites d'infection par le Rhizobium de la tige de la légumineuse, Sesbania rostrata. Comptes Rendus de l'Académie des Sciences, Paris, ser III 294, 407-411.
- ELMERICH, C., DREYFUS, B., REYSSET, G. & AUBERT, J.P. 1982 Genetic analysis of nitrogen fixation in a fast-growing *Rhizobium*. Embo Journal 1, 499-503.
- GERMANI, G., REVERSAT, G. & LUC, M. 1983 Effect of Sesbania rostrata on Hirschmanniella oryzae in flooded rice. Journal of Nematology 15, 269-271.
- JUNG, G., MUGNIER, J., DIEM, H.G. & DOMMERGUES, Y.R. 1982 Polymer-entrapped *Rhizobium* as an inoculant for legumes. *Plant and Soil* 65, 219-231.
- PATNAIK, S. & RAO, M.V. 1979 Sources of nitrogen for rice production. In *Nitrogen and Rice*, pp. 25–43. Los Baños, Philippines: International Rice Research Institute.
- RINAUDO, G., DREYFUS, B. & DOMMERGUES, Y.R. 1982a Sesbania rostrata as a greenmanure for rice in West Africa. In Biological Nitrogen Fixation Technology for Tropical Agriculture, ed. Graham, P.H. & Harris, S.C. pp. 441-445. Cali, Colombia: Centro Internacional de Agricultura Tropical.
- RINAUDO, G., DREYFUS, B. & DOMMERGUES, Y.R. 1982b Influence of Sesbania rostrata green-manure on the nitrogen content of rice crop and soil. Soil Biology and Biochemistry 15, 111-113.
- TSIEN, B. DREYFUS, B. & SCHMIDT, E.L. 1983 Initial stages in the morphogenesis of the nitrogen-fixing stem nodules of Sesbania rostrata. Journal of Bacteriology 156, 888-897.
- VACHHANI, M.V. & MURTY, K.S. 1964 Green manuring for rice. Indian Council of Agricultural Research Reports Series No. 17, 50pp.
- WATANABE, I. & APP, A. 1979 Research needs for management of nitrogen fixation in flooded rice crop system. In *Nitrogen and Rice*, pp. 485-409. Los Baños, Philippines: International Rice Research Institute.

#### Summary

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Sesbania rostrata is a wild annual legume that grows in West Africa in waterlogged soils during

the rainy season. This plant can bear  $N_2$ -fixing nodules on roots and stems at the same time. Stem nodules result from the infection of predetermined sites (dormant root primordia distributed on the stems) by a specific strain of *Rhizobium*. The mode of infection of *S. rostrata* is unique among the legumes, as it involves both an intercellular invasion by rhizobia and the development of infection threads. Inoculation of the stems is achieved by spraying a liquid culture of the specific *Rhizobium* on the shoots. Properly inoculated *S. rostrata* can fix up to 200 kg N<sub>2</sub>/ha in 50 days. A unique property of *S. rostrata* is to nodulate and fix nitrogen even when the amount of combined nitrogen in the soil is high. Based on 1 m<sup>2</sup> microplot trials conducted in Senegal during the summer the use of *S. rostrata* as green manure doubled rice yields. These results were later confirmed in trials conducted on 25 m<sup>2</sup> plots. In winter, when the day length is shorter, *S. rostrata* grows poorly but flowers readily. *Sesbania rostrata* trials should be conducted in two successive stages: (1) assessment of *S. rostrata* behaviour in a given geographic region; (2) assessment of the effect of *S. rostrata* green manure on rice yields.

#### Résumé

Observations sur l'utilisation de Sesbania rostrata comme engrais vert dans les rizières Sesbania rostrata est une légumineuse annuelle poussant dans les sols inondés de l'Afrique de l'Ouest pendant la saison des pluies. Cette plante peut porter des nodules fixateurs d'azote à la fois sur les racines et sur les tiges. Les nodules de tige proviennent de l'infection de sites prédéterminés (primordia de racines répartis sur les tiges) par une souche spécifique de Rhizobium. Le mode d'infection de S. rostrata est unique parmi les légumineuses; il comporte en effet deux étapes: invasion intercellulaire par les Rhizobiums et développement de cordons d'infection. On inocule les tiges en pulvérisant sur les parties aériennes une culture liquide du Rhizobium spécifique. S'il est convenablement inoculé, S. rostrata peut fixer jusqu'à 200 kg  $N_2$ /ha en 50 jours. Une caractéristique remarquable de S. rostrata est de noduler et de fixer l'azote même en présence de doses élevées d'azote combiné dans le sol. Des expériences conduites pendant l'été sur des microparcelles de 1 m<sup>2</sup> au Sénégal ont montré que l'utilisation de S. rostrata comme engrais vert pourrait doubler les rendements du riz. Ces résultats ont été confirmés ultérieurement sur des parcelles de 25 m<sup>2</sup>. En hivers, lorsque la durée du jour est plus courte, S. rostrata se développe mal et fleurit facilement. Les essais à conduire sur S. rostrata doivent comporter deux étapes: (1) évaluation du comportement de S. rostrata dans une région géographique donnée; (2) évaluation de l'effet de l'emploi de l'engrais vert S. rostrata sur les rendements du riz.

#### Resumen

#### Observaciones sobre el uso de Sesbania rostrata como abono verde en arrozales

Sesbania rostrata es una planta leguminosa anual que crece espontaneamente en el Oeste de Africa en suelos inundados durante la temporada de lluvias. Esta planta puede formar nódulos fijadores de nitrógeno en raíces y tallos simultaneamente. Los nódulos en tallos resultan de la infección por una cepa específica de Rhizobium de lugares predeterminados (primordios de raíces latentes, distribuidos en el tallo). El tipo de infección que ocurre en S. rostrata es único en leguminosas ya que aparte de una invasión intercelular de Rhizobium conlleva el desarrollo de hebras de infección. La inoculación de los tallos se realiza pulverizando sobre estos una cultura líquida del Rhizobium específico mediante un spray. Sesbania rostrata adecuadamente inoculada puede fijar hasta 200 kg/ha de nitrógeno en cincuenta días. Una propiedad singular de esta planta es su capacidad para nodular y fijar nitrógeno aún cuando el nitrógeno combinado existente en el suelo sea elevado. En experiencias realizadas durante el verano en Senegal en pequeñas parcelas de un metro cuadrado el uso de S. rostrata como abono verde dobló la cosecha de arroz. Estos resultados se confirmarén más tarde en parcelas experimentales de 25 m<sup>2</sup>. Durante el invierno cuando el día se acorta nel crecimiento de S. rostrata se ve frenado y tiene lugar la floración. Los próximos ensayos con S. rostrata deberían de realizarse a dos nivels: (1) estudio del comportamiento de S. rostrata en una determinada área geográfica; (1) estudio del efecto de la utilización de S. rostrata como abono verde en arrozales.