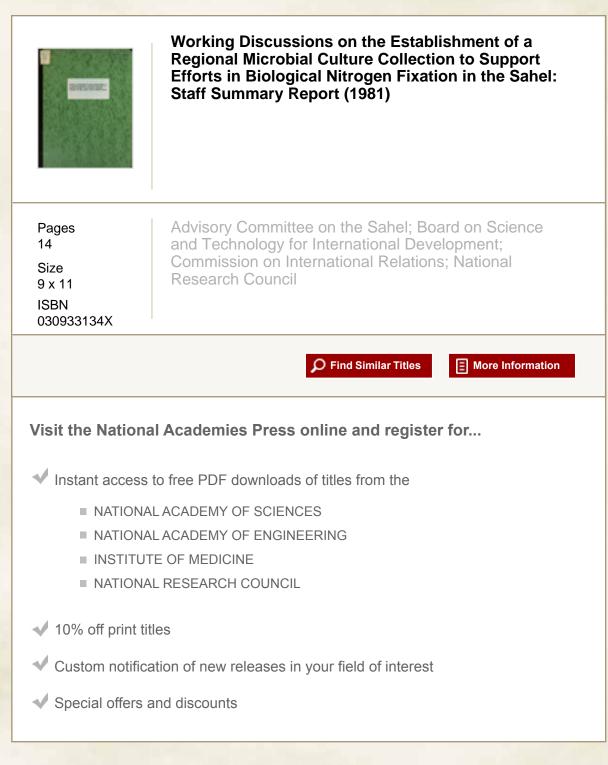
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STAFF SUMMARY REPORT

WORKING DISCUSSIONS ON THE ESTABLISHMENT OF A REGIONAL MICROBIAL CULTURE COLLECTION TO SUPPORT EFFORTS IN BIOLOGICAL NITROGEN FIXATION IN THE SAHEL

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#### BACKGROUND

BOSTID's interest in the establishment of a regional microbial culture collection for the Sahel emerged in connection with working discussions held in Mauritania in April 1979.\* In visits to the Nouakchott greenbelt and its associated nursery, BOSTID team members found that the <u>Prosopis</u> <u>chilensis</u> plantings were not nodulated and were performing poorly when transplanted. While the initial recommendations resulting from this visit focused on improved nursery techniques and a more appropriate schedule of watering, soil samples taken in the greenbelt and analyzed at the U.S. Department of Agriculture (USDA) laboratories at Beltsville, Maryland, indicated that the soil was effectively sterile.

Subsequent discussions with Sahelian foresters, officials of the Agency for International Development, Canadian International Development Research Centre (IDRC) scientists, and others have suggested that environmental degradation in the Sahel has greatly altered the soil ecology of the region. Among the victims of disturbed soil ecology have been soil microorganisms such as <u>Rhizobium</u>, <u>Frankia</u>, endomycorrhizae, ectomycorrhizae, and various free-living, nitrogen-fixing bacteria that play important roles in plant establishment, productivity, and survival.

The maintenance of essential ecological processes is tied structurally to a healthy soil environment. Hence, BOSTID has stressed increasingly the importance of linking project activities in the areas of agriculture and environmental rehabilitation with careful evaluations of soil ecology in the areas considered. Because Sahelian efforts in these areas characteristically involve various legumes, particular attention has been paid to Rhizobium-leguminous plant associations.

Since the April 1979 working discussions in Mauritania, BOSTID has explored the possibility of establishing a regional microbial culture collection in the Sahel with Jacques Diouf of the Senegalese Délégation Générale à la Recherche Scientifique et

\*See the BOSTID staff summary report, "An Assessment of Agro-Forestry Potential within the Environmental Framework of Mauritania." More explicit statements of concern are contained in the semiannual management reports of BOSTID's Advisory Committee on the Sahel (ACOS) and in the summary report of the joint Sahel Institute-ACOS Workshop on Ecology and Environmental Programs in the Sahel held January 1980. Technique (presently the Secrétariat d'Etat à la Recherche Scientifique et Technique), Louis Sauger (formerly of the Institut Sénégalais de Recherches Agricoles), El-Hadji Sène of the Senegalese Service des Eaux et Forêts, Albassadjé Touré of the Sahel Institute, Robert Winterbottom of the Permanent Interstate Committee for Drought Control in the Sahel (CILSS), Lloyd Frederick and various AID officials, E.L. Schmidt of the University of Minnesota (involved in a collaborative effort in inoculation testing of rhizobia with ORSTOM (Office de la Recherche Scientifique et Technique Outre-Mer) and the Senegalese Centre National de Recherches Agronomiques de Bambey), Jean Gorse of the World Bank, L. Gilles Lessard and Gordon MacNeil of the IDRC, Harold Keyser and Deane Weber of the USDA, Joe Burton of NifTAL and the Nitragin Company, and others.

These discussions strongly supported the establishment of a regional microbial culture collection. Thus BOSTID proposed that further, more specific discussions involving the Sahel Institute, interested Senegalese officials, ORSTOM scientists, AID and USDA officials, and BOSTID representatives be held in Dakar in November 1979. As the need for such discussions was not widely recognized within AID's nontechnical bureaus, the discussions were postponed.

In the absence of any visible movement to establish a microbial culture collection in the Sahel, BOSTID informally linked concerned Sahelians with sources of inoculum in the United States, principally with USDA's Cell Culture and Nitrogen Fixation Laboratory and the Nitragin Company. These linkages permitted Sahelian access to U.S. collections, facilitated germ plasm exchanges that would enlarge these collections in areas of critical importance to Sahelian agriculturalists and foresters, and resulted in the distribution of some inoculum in the Sahel. This networking was intended to serve as an interim measure until a collection better adapted to Sahelian needs and conditions could be established.

This matter is being pursued with renewed urgency as a result of suggestions that inoculum viability is adversely affected by temperature fluctuations and delays in shipping. Recent shipping trials reported by Cris Stearn, based in Mayagüez, Puerto Rico, have verified the reality and seriousness of this problem. Hence, with the support of AID's Office of Sahel and Francophone West Africa Affairs, BOSTID took advantage of the occasion of an international workshop on biological nitrogen fixation, held at the Centro Internacional de Agricultura Tropical (CIAT) in Colombia, to convene working discussions on the establishment of a Sahelian inoculum collection.

#### CURRENT STATUS OF BIOLOGICAL NITROGEN FIXATION IN THE SAHEL

The most advanced efforts in biological nitrogen fixation in the Sahel are those currently directed by Y.R. Dommergues at the ORSTOM center in Dakar. ORSTOM research in root-microorganism associations in arid and semi-arid regions was prompted by the fact that trees and shrubs play particularly important roles in supporting human, livestock, and wildlife populations in the Sahel. They provide fuel, building materials, medicinal substances, and economic products, and forage and shade for livestock and wildlife, and promote environmental rehabilitation through nutrient accumulation, soil improvement, erosion control, and by intercepting precipitation and facilitating infiltration.

Aware of the serious consequences of progressive degradation, ORSTOM scientists have undertaken research in the manipulation of various elements of the soil microflora in order to improve plant growth under the harsh edaphic and climatic conditions of the Sahel region. This research has generally focused upon <u>Rhizobium</u> inoculation, endomycorrhizae, ectomycorrhizae actinorhizal plants, and biocoenosis.

Rhizobium inoculation for leguminous trees and shrubs in arid and semi-arid regions appears to be of particular importance in assuring establishment during the seedling stage. Recent research on <u>Prosopis</u> spp. at the University of California at Riverside suggests that it might also be of greater importance than previously assumed in more mature plants as well. In addition to the more familiar root associations, ORSTOM scientists have found that <u>Sesbania rostrata</u> bears stem nodules when inoculated with its specific Rhizobium, and that these nodules permit the fixation of N<sub>2</sub> in the presence of high concentrations of inorganic nitrogen. Green manuring with <u>S. rostrata</u> has resulted in a quadrupling of rice production in trials reported at the CIAT workshop.

Dr. Dommergues indicated that the ORSTOM laboratory in Dakar currently possesses a collection of some 30-40 strains of <u>Rhizobium</u>. He also noted that in some tree species, such as <u>Acacia</u> <u>holosericea</u> and <u>A.</u> <u>raddiana</u>, the inoculum is more effective if the <u>Rhizobium</u> is combined with endomycorrhizae.

Research by ORSTOM in Senegal indicates that legumes inoculated with vesicular-arbuscular endomycorrhizae increased their phosphorus uptake and, in the case of <u>Vigna</u> unguiculata, their nitrogen content. This research suggests further that endomycorrhizae inoculation can increase crop yields in plants infested with nematodes to levels recorded in noninfested plants. This quality is of particular interest because nematode infestation is a principal cause of poor crop yields in the Sahel.

The beneficial contributions of ectomycorrhizae to the establishment of <u>Pinus caribaea</u> have been demonstrated in field trials in Senegal. Similarly, laboratory experiments in Australia suggest that <u>Eucalyptus gummifera</u> might respond well to inoculation with the ectomycorrhizal fungus, <u>Pisolithus tinctorius</u>. ORSTOM researchers are presently pursuing this matter further in collaboration with the Institut Sénégalais de Recherches Agricoles (ISRA).

In their consideration of actinorhizal plants, ORSTOM research has apparently dealt almost exclusively with trees of the genus Casuarina (a genus that, according to John Torrey of Harvard University, is presently being taxonomically reevaluated). Particular attention has been devoted to C. equisetifolia, a species long utilized in coastal dune stabilization efforts in Senegal. The root system of Casuarina can be infected by an actinomycete, probably of the genus Frankia, which results in the formation of root nodules within which microbial enzyme nitrogenase occurs. The trees are presently inoculated with crushed nodules containing five strains. (There is still some question as to which strain is effective and how the process works.) It is known that such inoculation reduces stress when seedlings are transplanted. Although not currently objects of research in the Sahel, other genera of actinorhizal plants, such as Colletia, Coriaria, Datisca, and Elseagnus, are worthy of consideration. Coriara sinica, for example, is utilized as a green manure by Chinese farmers in Hunan, and BOSTID has suggested the possibility of trial plantings of Elaeagnus angustifolia in Sahelian shelterbelts.

Although there has been some interest in the Sahel in the possible contribution of biocoenosis, or associative symbiosis, to nitrogen fixation, this interest has decreased in recent years. It has been found that estimates of rhizospheric  $N_2$  fixation utilizing the acetylene reduction method have been exaggerated for various reasons, particularly because incubations are too long and the systems under study are therefore modified excessively. Australian research further suggests that environmental factors such as soil water content, combined nitrogen, and root pathogens tend to decrease rhizospheric N<sub>2</sub> fixation markedly. This research indicates that fixation through biocoenosis probably falls within the range of 0-15 kg/ha. Perhaps most importantly, research conducted at the ORSTOM laboratory in Dakar indicates that there is no relationship between rhizospheric N<sub>2</sub> fixation as measured by acetylene reduction and plant growth. It should be added, however, that it is possible that inoculation with certain strains of <u>Azospirillum</u> could be beneficial to irrigated rice culture. In this case, the effect of the inoculum has been attributed to the production of growth-stimulating substances by the bacteria introduced into the rhizosphere.

#### LIMITING FACTORS AND CONCLUSIONS

There appear to be several potential obstacles to successful implementation of an inoculation program in the Sahel, falling into the categories of production and inoculum distribution and utilization.

### Production

It became clear during discussions in the Sahel that the advantages of biological nitrogen fixation are generally less obvious to planners, officials, and conventionally trained technicians than to microbiologists. Thus development of inoculum production facilities are assigned a lower priority than the demands of existing programs. Little effort has been made to inform decision makers of the economic and environmental benefits of biological nitrogen fixation (BNF). Further, some proponents of BNF have presented the approach as an alternative to existing fertilizer programs, which it is not. BNF can sharply reduce fertilizer costs and substantially increase the ecological soundness and sustainability of agricultural production. However, in many instances supplemental nitrogen is still required and BNF contributes only indirectly to the further nutrient requirements of crops.

The benefits and limitations of biological nitrogen fixation should be more rigorously assessed and reconciled with the established needs of Sahelian agriculture and forestry.



The appropriate institutional affiliations of a regional microbial culture collection must also be determined more clearly. It is generally agreed that such a collection should be located in Bambey or Dakar. This agreement is based on considerations of communications, research facilities, dependable power supply in relation to collection storage, and inoculum distribution. On this basis, Dakar appears to be the more appropriate site. Similarly, there has been general agreement that the collection should be housed within the framework of ISRA. ISRA, which is institutionally responsible for both agricultural and forestry research in Senegal, has expressed interest in producing ectomycorrhizae inoculum and in conducting Rhizobium research. Further, ORSTOM and the University of Minnesota (USDA Grant No. 801-15-67) are already involved in collaborative efforts in BNF with ISRA, and the IDRC's experience in the inoculation of Acacia senegal is known to ISRA researchers (although carried out under the auspices of the Ministry of Rural Development).

The establishment of Microbiological Resource Centers (MIRCENs) was proposed to a group of microbiologists by M.K. Tolba of the United Nations Environment Programme (UNEP) in 1974. Financial support for implementation of this proposal was provided by UNEP and UNESCO. One of the aims of MIRCENs is to provide the infrastructure for a network that would incorporate regional and interregional units geared to the management, distribution, and utilization of microbial gene pools.

The first step in initiating this concept was establishment of the World Data Center (WDC)\* as a MIRCEN, which was undertaken in close collaboration with the World Federation for Culture Collections. This MIRCEN serves as a pivotal unit for the formation of culture collections in developing countries and for providing data services to centers acting in liaison with the WDC.

In 1975, a MIRCEN was proposed for Senegal. Although no action has been taken on the proposal, some individuals feel that it is wise to establish a MIRCEN in Francophone West Africa. Such a center would be complementary to the MIRCEN associated with the Department of Soil Science at the University of Nairobi in East Africa. In discussions during the CIAT workshop, Edgar da Silva of

\*The WDC maintains a computerized listing of the strains held by various institutions and their characteristics. UNESCO supported establishment of a MIRCEN in Senegal. Similarly, S.O. Keya, the director of the Nairobi MIRCEN, was receptive to the establishment of such a center and would be an excellent resource person to assist with the details of such an effort.

It is suggested that a West African MIRCEN be established in Dakar, and housed within the framework of ISRA, possibly utilizing the facilities of the Centre National de Recherches Forestières (CNRF). It is suggested further that this effort be coordinated with ORSTOM, USDA/AID, and IDRC activities in the area of biological nitrogen fixation and be reconciled with the regional responsibilities of the Sahel Institute.

With regard to staffing, it was noted in discussions that relatively few Sahelians are trained as microbiologists. ISRA (CNRF), together with the Service des Eaux et Forêts, supports a Laval- and ORSTOM-trained microbiologist, Ibrahima Gueye, who is currently working at the CNRF station in Dakar. ORSTOM is currently supporting and training two additional soil microbiologists, Alphonse Kabré from Upper Volta and Mamadou Gueye from Senegal. Both of the latter trainees are regarded as being particularly promising. In addition to these individuals, discussants at the CIAT workshop were aware of two Malian microbiologists and a small number of scientists from Upper Volta. Most of these individuals are relatively inexperienced.

The establishment of a regional microbial culture collection would require further training for those Sahelian scientists responsible for maintenance of the collection. During the initial development stages, it would almost certainly be necessary to recruit staff members from outside the Sahel.

### Inoculum Distribution and Utilization

Problems of inoculum distribution and utilization within the Sahel region should perhaps be explored within the CILSS framework. Within this framework, the Sahel Institute remains responsible for the dissemination of scientific and technical information, technology transfer, coordination of scientific research, and training.

BOSTID is prepared to assist the Institute and/or other interested parties with the establishment of a regional microbial culture collection. It is similarly prepared to assist with the organization of the delivery systems and address the various problems of utilization associated with such a collection.

## DISCUSSANTS

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