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Soils Newsletter



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OF NUCLEAR TECHNIQUES IN FOOD AND AGRICULTURE
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VOL. 20 No. 1 2

TO OUR READERS

1996 was the busiest year so far for the Soils Section, and another successful one.

As reported in the last issue of the Soils Newsletter in May 1996, a team of six agricultural scientists and research-management specialists conducted an external review of the Soils Sub-Programme. The Review Team strongly supported our new emphasis on integration of soil, water and nutrient management activities within a cropping-systems context, using nuclear techniques. One of their major recommendations was that the Sub-Programme should "seek to participate fully in the Consortia on Soil, Water and Nutrient Management being developed by International Agricultural Research Centres". The programmes of the Centres are based on the same principle as our Sub-Programme, but with very limited or no application of nuclear techniques. In order to promote future collaboration, the Joint FAO/IAEA Division contacted the Consultative Group of International Agricultural Research (CGIAR) Centres involved in the Consortia, and it was agreed that the best way to move forward was to hold a Consultants Meeting between representatives from the CGIAR Centres and staff of the Soil and Water Management and Crop Nutrition Sub-Programme. The meeting was held in Vienna, 25-29 November 1996. The purpose was to identify areas in which nuclear techniques could play an important role in the research supported by the CGIAR, and to work out a mechanism for collaboration suitable to both the Centres and the Joint Division within existing programmes and resources. After reciprocating information and useful discussions, areas of future collaboration were elaborated. There is no doubt that this co-operation will be of mutual benefit, and will add a new dimension to our activities.

Attending the meeting on behalf of the CGIAR Centres were Dr. R. Thomas, Centro Internacional de Agricultura Tropical (CIAT), Cali, Colombia; Dr. J. White, Centro Internacional de Mejoramiento de Maiz y Trigo (CIMMYT), Mexico; Dr. J. Ryan, International Centre for Agricultural Research in Dry Areas (ICARDA), Aleppo, Syria; Dr. R. Buresh, International Centre for Research in Agroforestry (ICRAF) Nairobi, Kenya; Dr. B. Myers, International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India; Dr. H. Grimme, International Institute for Tropical Agriculture (IITA), Ibadan, Nigeria; and Dr. M. Kondo, International Rice Research Institute (IRRI), Los Baños, Philippines. Dr. R. Lefroy, International Board for Soil Research and Management (IBSRAM), Bangkok, Thailand, and Dr. Hassan Nabhan, representing the Land and Water Development Division, FAO, Rome, also participated.

Two Co-ordinated Research Programmes (CRPs) were initiated in the second half of 1996: (i) "The Use of Isotope Techniques in Studies on the Management of Organic Matter and Nutrient Turnover for Increased Sustainable Agricultural Production and Environmental Preservation," with the first Research Co-ordination Meeting (RCM) held in Vienna, 7-11 October 1996, and (ii) "The Assessment of Soil Erosion Through the Use of ¹³⁷Cs and Related Techniques as a Basis for Soil Conservation, Sustainable Production and Environmental Protection," with the first RCM held also in Vienna, 11-15 November 1996. The CRP on soil erosion is being implemented in close co-operation and interaction with a CRP on sedimentation run by the Isotope Hydrology Section, Division of Physical and Chemical Sciences. Both CRPs are of great importance for soil conservation, sustainable agriculture and environmental preservation, and nuclear techniques will complement other activities; significant results are expected.

The second RCM on "The Use of Irradiated Sewage Sludge to Increase Soil Fertility and Crop Yields and to Preserve the Environment" took place in Cairo, Egypt, 14-18 September 1996, and was attended by eleven Research Contract Holders and five Research Agreement Holders. Promising results were reported and satisfactory progress described in developing management practices for the efficient use of sewage sludge as an organic fertilizer for increasing crop production in an environmental-friendly manner. Excerpts from the reports presented are published elsewhere in this issue of the Newsletter.

The CRP on "Enhancing Soil Fertility and Crop Production by Better Management of *Rhizobium*" was completed with the fourth RCM held in Vienna, 2-6 September 1996. The results will be published as a book in 1997.

A 4-week Regional Training Course for Latin America was held in Irapuato, Mexico, 2-27 September 1996, with 19 participants from 10 countries, on "The Use of Nuclear Techniques in Studies of Soil/Plant Relationships". The course was very successful.

Three Workshops were organized by the Soil Section under Technical Co-operation activities. A second workshop for the "Regional Project for Asia and the Pacific on Nuclear Techniques for the Promotion of Agroforestry Systems" involved counterparts from ten countries from the region; results from last year were discussed and plans for future activities finalized. The other workshops, one in Santiago, Chile on "Strengthening Analytical Laboratories in Latin America," and the other in Faisalabad, Pakistan on "Sustainable Utilization of Wasteland on Saline Groundwater for Plant Production" were held in October and November 1996, respectively.

On the staff front, several changes occurred during the reporting period. For detailed information, please refer to the staff section (page 4 of this Newsletter).

I would like to express my thanks to all our staff members from HQ and SSU for their invaluable contributions to our intensive activities during 1996.

As always, I thank our readers for continued interest in our Soils Newsletter. We will be pleased to receive your comments and suggestions for improvement. Also, please notify us promptly of any change in your mailing list/address. This issue was edited by Allan Eaglesham and myself, with contributions from our colleagues.

I wish you all a Merry Christmas and a very Happy New Year.

Christian Hera
Head, Soil and Water Management &
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3. Staff Changes

Dr. Chris van Kessel concluded a one-year sabbatical leave with the Section and returned in October 1996 to the University of Saskatchewan, Saskatoon, Canada. In addition to his participation in many Section activities, Chris was Project Officer for CRPs on nitrogen-fixing trees and soil organic matter. His professionalism and depth of knowledge were much appreciated by his Soils Section/Unit colleagues. Chris will soon take up a Professorship at the University of California at Davis, California, USA, and we wish him and his wife Betsy the very best in their future endeavours.

In August 1996, **Dr. Allan Eaglesham** joined the Section on temporary assignment. Allan leaves in December for Jamaica, to join the Department of Life Sciences at the University of the West Indies as Professor of Botany. We thank him very much for his excellent contributions in various aspects of our research activities, and wish him and his family the best for the future.

Dr. Zaharah Rahman, from the University of Malaysia, joined the Soil Science Unit in September 1996 for a 4-month consultancy. Zaharah assisted in implementing work on P-use efficiency and nutrient availability from crop residues, and in training fellows in soil fertility. We are grateful for her able contributions and wish her the best of future success.

At the end of November 1996, **Ms. Silke Eckert** left after 2½ years service as an Associate Professional Officer with the Soil Science Unit at the FAO/IAEA Agriculture and Biotechnology Laboratory, Seibersdorf. We appreciate Silke's contributions to our work on water use efficiency, and wish her all the best for the future.

CO-ORDINATED RESEARCH PROGRAMMES

1. FAO/IAEA Co-ordinated Research Programme on Enhancing Soil Fertility and Crop Production by Better Management of *Rhizobium* (D1-40.06).

Project Officer: Gudni Hardarson

The fourth and final Research Co-ordination Meeting was held in Vienna 2-6 September 1996. This CRP had eight Research Contract Holders, Dr. S.M. Tsai (Brazil), Dr. H. Moawad (Egypt) Dr. E. Martínez-Romero (Mexico), Dr. J.J Peña-Cabriales (Mexico), Dr. K.A. Malik (Pakistan), Dr. J.C. Mamaril (Philippines), Dr. A. Popescu (Romania), Dr. N. Teaumroong (Thailand), and six Agreement Holders, Dr. W. Ludwig (Germany), Professor D. Gareth Jones (UK), Professor A. Akkermans (Netherlands), Professor W.J. Broughton (Switzerland), Dr. J.E. Cooper (UK), and Dr. P. Young (UK). Recommendations from the scientists participating in the meeting, and excerpts from their presented reports are provided elsewhere in this newsletter.

2. FAO/IAEA/OPEC Co-ordinated Research Programme on The Use of Nuclear Techniques in the Management of Nitrogen Fixing Trees for Enhancing Soil Fertility and Soil Conservation (D1-40.05).

Project Officer: Christian Hera

The last year for this CRP is underway and the final Research Co-ordination Meeting is scheduled to be held in September 1997, in Vienna. During this final meeting, all participants will be asked to present the major results and conclusions of their component of the Programme. Currently, there

are eight Contract Holders: C. Ovalle-Molina (Chile), E.Y. Safo (Ghana), H. Shariffuddin (Malaysia), K. Malik (Pakistan), N. Mbaya (Zaire), M. Gueye (Senegal), M. de S. Liyanage (Sri Lanka), M. Bekunda (Uganda), and three Agreement Holders are participating: J.K. Ladha (Philippines), N. Sanginga (Nigeria), and D. Baker (USA).

The final batches of labelled ^{15}N fertilizers have been sent out to the Contract Holders. Furthermore, samples are being analyzed in Seibersdorf and participants should receive the data by May 1997.

3. FAO/IAEA Co-ordinated Research Programme on the Use of Nuclear Techniques for Optimizing Fertilizer Applications under Irrigated Wheat to Increase the Efficient Use of Fertilizers and Consequently Reduce Environmental Pollution (DI.40.07).

Project Officer: Pierre Moutonnet

This programme includes seventeen participants, of which five are Agreement Holders, R. Rennie (Canada), G. Vachaud (France), I. Ortiz-Monasterio (Mexico), W. Baethgen and J. Schepers (USA), and twelve are Research Contract Holders, S.M. Rahman (Bangladesh), A.E. Boaretto (Brazil), I. Vidal (Chile), X. Wen (People's Republic of China), M.A.S. Abdel Monem (Egypt), M.S. Sachdev (India), X. Uvalle-Bueno (Mexico), J.M. Sanchez-Yanez (Mexico), M. Bazza (Morocco), C. Cioban (Romania), A. Arslan (Syria), Ç. Kirda (Turkey). A Consultants Meeting established the goals and objectives of this CRP in co-operation with CIMMYT, Mexico, and IFDC, USA. The first RCM was held in Vienna in October 1994. Fertilizers labelled with ^{15}N and several technical documents to be used as guidelines were provided to the contractors. ^{15}N -enrichment measurements of plant and soil samples have been made at the Seibersdorf Laboratory and in various developing countries. The first results were discussed in detail at the second RCM, which was held in El Batan CIMMYT Research Centre, near Mexico City, on 4-8 March 1996. Excerpts from presented papers were published in the last issue of the Newsletter.

The third season of experiments is now in progress in the northern-hemisphere countries. The next RCM will be held at the Agency in Sept.-Oct. 1997, and the programme is expected to close in 1998-1999.

4. FAO/IAEA Co-ordinated Research Programme on The Use of Nuclear and Related Techniques for Evaluating the Agronomic Effectiveness of Phosphate Fertilizers, in Particular Rock Phosphates (DI-50.03).

Project Officer: Felipe Zapata

This CRP is entering its final phase of implementation. Participants are conducting field experiments to gather information on the agronomic effectiveness of phosphate fertilizers under a variety of soil and climatic conditions. Special emphasis is being given to ways and means of enhancing the availability of P from rock phosphates to crops. Data collected will be used to test the P submodel of the DSSAT family of crop models. At the third Research Co-ordination Meeting, to be held 17 - 21 March 1997 at the IAEA Headquarters in Vienna, the participants will present progress reports.

The network, funded by the IAEA Regular Budget, has ten Contractors and six Agreement Holders. Thanks to generous support from the French Government, five additional Contractors, from Hungary, Lithuania, Poland, Romania and Russia, will be included in the programme this year.

Following the IAEA - IMPHOS Co-operation Agreement, soil samples from the benchmark field sites are being analysed at CIRAD, Montpellier, France. Similar arrangements are being made for a standardized characterization of the rock phosphates that are being utilized within the network.

5. FAO/IAEA Co-ordinated Research Programme on The Use of Irradiated Sewage Sludge to Increase Soil Fertility and Crop Yields and to Preserve the Environment (D1-50.04).

Project Officer: Christian Hera

The Second Research Co-ordination Meeting on the agricultural use of irradiated sewage took place in Cairo, Egypt, from 14 to 18 September, with Dr. Rawia El Motaïum acting as the local organizer. The meeting was attended by eleven Research Contract Holders, from Argentina (C. Magnavacca), Bangladesh (S. Ahmed), P.R. China (T. Jiang), Egypt (R. El Motaïum), India (T.J. D'Souza), Indonesia (M. Mitrosuhardjo), Malaysia (A.B. Rosenani), Mexico (M. Barajas-Aceves), Pakistan (F. Azam), Portugal (E. M. Ferreira), Thailand (J. Prasatsrisupab), and five Research Agreement Holders, from Austria (F. Koch), Germany (H. Harms), Japan (K. Kumazawa), USA (A.C. Chang), and UK (S.P. McGrath). Excerpts of the presented reports are presented elsewhere in this newsletter.

Dr. I.J. Manguiat, the Research Contract Holder in the Philippines informed us that he can no longer participate in the CRP because of inadequate funds. Therefore, we note with regret that the contract is terminated.

The next RCM will be held in March 1998. The venue and exact date will be announced in the next issue.

6. FAO/IAEA Co-ordinated Research Programme on the Use of Isotope Techniques in Studies on the Management of Organic Matter and Nutrient Turnover for Increased Sustainable Agricultural Production and Environmental Preservation (D1-40.08).

Project Officer: Christian Hera

The first Research Co-ordination Meeting for this CRP was held 7-11 October 1996 in Vienna, with Chris van Kessel as Scientific Secretary. There are twelve Research Contract Holders, S. M. Rahman (Bangladesh), K. Reichardt (Brazil), E. Zagal (Chile), J.Y. Wang (P.R. China), S.K.A. Danso (Ghana), M.S.A. Safwat (Egypt), R.A. Bakar (Malaysia), J.Z. Castellanos (Mexico), D. Amara (Sierra Leone), R. Sangakkara (Sri Lanka), and N.N. Dang (Vietnam), and three Agreement Holders, D.F. Herridge (Australia), R. Merckx (Belgium), and D.S. Powlson (United Kingdom). Excerpts from presented papers are printed elsewhere in this Newsletter. The next RCM is scheduled for spring 1998.

7. FAO/IAEA Co-ordinated Research Programme on The Assessment of Soil Erosion Through the Use of Cs-137 and Related Techniques as a Basis for Soil Conservation, Sustainable Production, and Environmental Protection (D1-50.05).

Project Officer: Felipe Zapata

As has been previously reported, this programme is being implemented closely with the CRP on sedimentation [Sedimentation Assessment Studies by Environmental Radionuclides and their Application to Soil Conservation Measures (F3-10.01) Scientific Secretary: Edmundo Garcia Agudo] because of similarities in documentation techniques. Erosion and sedimentation are particularly strongly inter-related when working at the watershed level. Currently there are eight Research Contract Holders, from Brazil, China (2), Chile, Romania, Russia, Slovakia and Zimbabwe, and four Agreement Holders, from Australia, Canada, UK, and USA.

The first Research Co-ordination Meeting for both CRPs took place 11-15 November 1996 in Vienna. The purpose was to present and evaluate preliminary data on soil erosion and deposition through the measurement of fallout of Cs-137 and other radionuclides, to discuss protocols on site selection, sampling and sample processing/analysis, and to review the overall work plan. The meeting will be fully reported in the next issue.

FAO/IAEA TRAINING COURSES

1. **IAEA/FAO Regional Training Course on The Use of Nuclear Techniques in Studies of Soil/Plant Relationships (RLA/5/037). Irapuato, Gto., Mexico, 2-27 September 1996.**

Technical Officer: Felipe Zapata
Course Director: Juan José Peña-Cabriales

Held at the Centro de Investigaciones y Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN) at Irapuato, this 4-week course was very intensive, and lecturers/instructors and participants alike are to be commended for their commitment and active involvement. The language of instruction was Spanish.

There were 19 participants from 10 countries: Argentina, Brazil (2), Chile, Colombia (2), Cuba (2), Dominican Republic, El Salvador, Guatemala, Mexico (6), and Venezuela (2). There were 20 lecturers from various research institutes and universities in Mexico, and five from other Latin American countries. The laboratory, greenhouse and field facilities were of high standard, and the lecture room, library and photocopying facilities were excellent. Most of the local instructors were trained in past FAO/IAEA training courses.

A course evaluation, conducted during the final week, included questionnaires relating separately to CINVESTAV and IAEA. While concluding that the course was a great success, participants expressed concern over follow-up activities and future connection with IAEA programmes. Copies of the full training programme giving details of lectures, practicals, field excursions, other related activities, and the list of participants, are available from the Soil and Water Management & Crop Nutrition Section.

MEETINGS/WORKSHOPS

1. **FAO/IAEA Research Co-ordination Meeting on Enhancing Soil Fertility and Crop Production by Better Management of *Rhizobium* (D1-40.06). Vienna, 2-6 September 1996.**

Scientific Secretary: Gudni Hardarson

The fourth and final FAO/IAEA Research Co-ordination Meeting was held in Vienna, 2-6 September, 1996. In attendance were eight Research Contract Holders and four Agreement Holders, or their representatives, with staff members from the Section and the Seibersdorf Laboratory. After introductory remarks by Acting Section Head Mr. F. Zapata, each participant gave a talk on the current status of their research (abstracted below). Research reports were compiled and edited, and recommendations for future activities were recorded. Many of the participants have generated important data that will be published in a Programme Report.

Better Utilization of Biological Nitrogen Fixation in Cropping Systems: a formal statement to FAO/IAEA from Research Contract Holders and Agreement Holders

Firm evidence now exists to show that legumes, in particular the common bean (*Phaseolus vulgaris*), the most important legume for human consumption, can respond successfully to inoculation with rhizobia and positively contribute to the nutrient status of the soil. Although common bean has the ability to fix N₂ at levels equal to soyabean, the crop frequently fails to achieve its yield potential. Therefore, there is a pressing need to define the limitations to good growth in the field.

Developments in legume breeding, advances in molecular microbiological methods, and the availability of nuclear techniques, now offer interdisciplinary approaches to solving problems of increasing BNF contributions to crop production. In particular, a range of molecular techniques, developed in the Co-ordinated Research Programme on Enhancing Soil Fertility and Crop Production by Better Management of *Rhizobium*, allowed the monitoring in soil of introduced rhizobia in terms of

survival, colonization of rhizosphere and root, and ability to compete with indigenous rhizobia for nodulation sites. With an integrated approach, it will be possible to identify combinations of legume cultivar and rhizobial-inoculant strain for particular soil conditions, to alleviate yield constraints and enhance the sustainability of legume-based cropping systems.

The molecular methods developed in the current CRP offer possibilities for related programmes aimed at improving plant performance by manipulation of rhizosphere microbial activity. For example:

- Assessment of benefits to legumes, wheat, rice, sugarcane etc., from co-inoculation with associative N_2 fixers, plant-growth-promoting bacteria, and mycorrhizal fungi.
- Analyses of changes in soil biodiversity under particular cropping conditions.
- Quality control of microbial inoculants for crop plants.
- Manipulation of micro-organisms for enhanced symbiotic performance.
- Identification of microbial strains with increased resistance to environmental stress (drought and high temperature, salinity, acidity, high aluminium etc.) and agrochemicals, together with precise assessment of the influence of such stresses on key aspects of host-plant/micro-organism interactions.

Excerpts from presented reports:

Esperanza Martínez-Romero

Centro de Investigación sobre Fijación de Nitrógeno

Cuernavaca, Mor.

Mexico

Symbiotic performance of some modified Rhizobium strains in assays with high- N_2 -fixing beans.

Although common bean (*Phaseolus vulgaris*) originated in the Americas, it has become an important crop worldwide. *Rhizobium* species that form N_2 -fixing nodules on common bean are now characterized, and new species proposed: *R. etli* and *R. tropici*.

Common bean is reported to show low N_2 -fixing rates, and various research projects have been directed at improvement of symbiotic effectiveness. The aim of this work was to combine *Rhizobium* strains with improved nodulation ability with recently-described high- N_2 -fixing cultivars. Using *R. etli* strains with additional citrate synthase genes, we obtained strain- and cultivar-dependent differences in nodulation. However, superior nodulation was not reflected in better plant development.

We have also analyzed symbiotic performance of genetically-modified *Rhizobium* strains. A strain of *R. tropici* with a cassette on the *nodH* gene, incapable of producing sulphated Nod factors, increased nodule number with high-fixing bean. In soil, this strain was more competitive in terms of nodule number than native *R. etli*; it remains to be determined if it improves plant development and grain yield.

Siu Mui Tsai

Centre for Nuclear Energy in Agriculture

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Brazil

Molecular and evolutionary aspects of common bean-Rhizobium interactions.

Molecular markers can be used as indirect selection tools to simplify breeding or to provide information about genome evolution. Markers with limited polymorphism, such as some morphological traits or isozymes, may provide useful additional information for linkage mapping in the common bean. This work used a recently developed RFLP linkage map between BAT-93 of *meso*-American origin, and Jalo EEP558 of Andean origin, for genotypic differentiation.

Fifty F_8 - F_9 progenies were grown with mineral N supplied at either optimum (0.25 mM N) or inhibitory (5 mM N) levels for nodule formation. We identified genetic loci that affect quantitative trait

loci (QTLs) such as nodule number and have possible association with QTLs involved in responses to pathogens *Xanthomonas campestris* pv. *phaseoli* (causative of common bacterial blight) and *Pseudomonas syringae* pv. *tabaci* (causative of wildfire disease of common bean).

Nitrogen enhanced the expression of both diseases, and inhibited nodulation; the diseases were scored by quantitative analysis of lesion areas. Under high N, regions known to be associated with nodule number were detected, indicating QTLs that control host response to symbiotic and pathogenic organisms. Increased susceptibility to one or both diseases was observed among the most recently selected superior N₂-fixing lines. It is apparent that breeding for a BNF trait in common bean affects resistance to disease.

Most materials in breeding programs are highly disease-resistant, therefore this study suggests that negative selection for BNF has occurred in N-rich soils. In future breeding efforts with common bean, due consideration should be given to symbiotic characteristics as important agronomic traits.

Neung Teaumroong

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Molecular microbial ecology approaches to detect Bradyrhizobium in Thailand.

Total chromosomal DNAs were extracted from 38 *Bradyrhizobium* strains (10 from *Vigna radiata*, 10 from *Arachis hypogaea*, 18 from *Glycine max*) to serve as templates for REP, ERIC and RAPD primers in PCR analyses. The resulting patterns were highly specific for each strain, especially when analysed with intrinsic antibiotic resistance patterns, serving as a useful data base as dendrograms.

In developing technologies to detect strains of *Bradyrhizobium* applied as inocula to soil, a method for extracting DNA directly from soil was established. Recovery of 20-30 µg DNA g⁻¹ soil is possible, sufficient for PCR amplification.

Marker genes were also used to distinguish applied *Bradyrhizobium* from indigenous strains. Strains TAL 1000 (*A. hypogaea* symbiont) and TAL 379 (*G. max*) were used as recipients for the GUS transposon. Planted in sterile sand, seeds of the respective host species were inoculated with 10⁶ cells of the *gus*⁺ strains, and after 27 days nodules were stained to detect the blue dye; occupancy by both strains was approximately 90%. Similar occupancy studies in soil are in progress.

James E. Cooper

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Early bacterial and plant responses during the formation of legume-Rhizobium symbioses.

Molecular, biochemical, and chemical analytical techniques were employed to investigate plant and bacterial responses in the very early stages of communication between a legume and its rhizobial symbiont. Symbiosis-specific gene expression in white clover was analyzed by differential display reverse transcription PCR (DDRT-PCR). White clover homologues of *ENOD40* and phosphoglycerate mutase, among other cDNAs, were so detected.

Reverse transcription of PCR (RT-PCR) assays revealed the temporal expression patterns of a number of mRNAs in roots during a 5-day post-inoculation period. RT-PCR expression analyses were also performed on other genes (isoflavone reductase and cyclin) that had been isolated from root cDNA using degenerate primer PCR.

Flavonoid degradation by rhizobia was studied by means of direct GC-MS analyses of derivatized culture supernatants. Many rhizobia degrade legume flavonoids by mechanisms originating in fissions on the central (C-) ring of these polyphenolic compounds. Label tracing experiments with ^{14}C -naringenin demonstrated that C atoms from this flavanone were incorporated into the lipid moiety of a *Rhizobium leguminosarum* bv. *viciae* Nod factor.

Hyphenated separation techniques (TLC-UV, HPLC-UV, CZE-UV, and GC-MS) were used to identify flavones in seed and root exudates of *Lotus pendunculatus* and to show that new flavonoids and monocyclic aromatics are formed when exudates are incubated with *R. loti*.

Several molecular methods were applied to the detection of flavonoid-dependent gene expression in *Rhizobium* strain NGR234 - including competitive RNA hybridization, subtractive DNA hybridization, and RNA arbitrarily primed PCR (RAP-PCR). RAP-PCR of daidzein-induced and uninduced NGR234 cells revealed several daidzein-dependent products, among which were a 269 bp cDNA with 93% homology to a soyabean-cultivar specificity gene (*noiU*) from *R. fredii*, and a 308 bp cDNA with 99% homology to the polyphosphate kinase (*ppk*) gene from *E. coli*.

M. Sajjad Mirza

National Institute for Biotechnology and Genetic Engineering (NIBGE)

Faisalabad

Pakistan

Detection of diazotrophic bacteria associated with the roots of rice.

Plant growth promoting rhizobacteria (PGPR) have been shown to stimulate growth of a variety of plant species. Five PGPR strains of the genera *Azospirillum* (N-4, K01, WB-3, and WB 6) and *Zoogloea* (Ky-1) were used to inoculate seedlings of two rice (*Oryza sativa*) varieties (BAS-370 and NLAB-6). With NLAB-6, maximum N_2 fixation (38% Ndfa) was detected with Ky-1. With BAS-370, maximum (84% Ndfa) was recorded with N-4, however the greatest beneficial effect on plant growth prevailed with Ky-1.

In a field experiment, total bacterial populations in rhizosphere soil continuously increased equally in inoculated and non-inoculated plots until 3 months after transplantation, whereas maximum numbers of diazotrophs were observed at 1 month.

Within plant roots, higher numbers of bacteria were detected on inoculated than on non-inoculated plots. Total bacterial counts increased until harvest, whereas diazotroph numbers again declined after 1 month. Higher acetylene reduction activities were obtained with inoculated than with non-inoculated roots. Using PCR, the maximum number of cells within plant roots was recorded as $1.4 \times 10^5 \text{ g}^{-1}$ at 1 month after seedling transplantation from the nursery.

Kaisa Haukka

Department of Biology

University of York

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UK

Diversity of 16S rRNA and symbiotic genes in rhizobia that nodulate African Acacia and Prosopis.

We studied the diversity of chromosomal 16S rRNA genes and plasmid-borne *nifH* and *nodD* genes in a collection of rhizobial strains isolated from *Acacia senegal* and *Prosopis chilensis* trees growing in the Sudan and Kenya. The strains were selected on the basis of information from previous studies using numerical taxonomy, pulsed-field gel electrophoresis and plasmid-profile analysis.

A 230-bp fragment of 16S rRNA was sequenced. In thirty strains, twelve different sequences were obtained: four identical with previously described species, and eight novel. Sequence comparisons indicated that one strain was of the "*Mesorhizobium*" phylogenetic branch, which includes *Rhizobium loti* and *R. huakuii*, whereas the others were *Sinorhizobium* close to *S. (Rhizobium) meliloti*, *S. fredii*, *S. teranga*, and *S. saheli*. The sequences indicate recombination between 16S rRNA genes among our isolates. Moreover, one isolate had two 16S rRNA sequences differing at six positions. The type strain of *S. saheli* was shown to have similar microheterogeneity among its 16S rRNA genes.

Using PCR-RFLP, twelve *nifH* banding patterns that were found among 45 strains did not correlate with 16S rRNA sequences. We chose nine Sudanese, three "*Mesorhizobium*" and eight *Sinorhizobium* isolates from other tree species and countries in Africa and Latin America, for *nifH* sequencing. Analysis of a 561-bp fragment revealed that most differences are third-base substitutions that do not affect amino-acid sequence. In a phylogenetic tree of the *nifH* proteins, the "mesorhizobia" were grouped together, and African *sinorhizobia* resembled each other although four slightly different sequences were found (one or two variable amino acids). Within the *Sinorhizobium* clusters from Africa and Latin America, different chromosomal backgrounds (16S rRNA) variously contained similar and dissimilar *nifH* copies. The *nifH* genes in Latin American *sinorhizobia* more closely resembled those from *R. etli* and *R. tropici* than from African isolates, which may mean that genes are exchanged across genera.

RFLP hybridizations using a *nodD* probe showed that most strains have two similar genes and, in some cases, one different. We now have a complete *nodD* sequence for one strain, and it is different from other known *nodD* sequences, with only a 77% similarity to *S. meliloti nodD3* and 75% to that of *S. fredii*. Partial sequences of three other *nodD* genes from Sudanese isolates were found to be similar to each other and to the complete sequence, regardless of chromosomal differences and of the species of derivation. On the basis of these results, *nodD* and *nifH* gene-sequence types do not seem to be correlated.

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PCR fingerprinting of Azospirillum and Rhizobium.

The bacteria used in this study were, *Azospirillum* sp. strain BSs 8, *A. lipoferum* TN8 and 34H, *A. brasilense* T2W, 1577, Sp 7, and rhizobial strains BJVr 12 from *Vigna radiata*, BJLl 12 and BLl 80 from *Leucaena leucocephala*, and CIAT 590 from *Centrosema pubescens*.

PCR-based fingerprint patterns of genomic DNA from the *Azospirillum* and rhizobial strains were generated using ERIC, REP and BOX AIR primers. Each pattern was unique. The numbers of bands discernible with ERIC for *Azospirillum* were eleven (strain BSs 8), five (1577), four (Sp 7), three (34H), and two (T2W), and for the rhizobia were nine (CIAT 590), seven (BJLl 12) six (BJVr 12), and two (BLl 80). The REP-PCR fingerprint patterns were more complex, revealing more bands than with ERIC, whereas fewer bands were obtained with BOX AIR. Amplified products of these generated fingerprint patterns ranged from 0.3 to 1.5 kb.

The amplified products of strain BSs 8 with ERIC will be used for the preparation of probes for monitoring and identifying BSs 8 in ecological studies.

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Nod-factor attachment, calcium fluxes, and lipid-transfer proteins in symbiotic signal transduction.

Temporal and spatial observations on the attachment of Nod-factors were made by biotinylating the reducing terminus of the lipo-chitooligosaccharides of the broad host range *Rhizobium* sp. NGR234 with the fluorescent reagent 2-amino-(6-amidobiotinyl)pyridine. Complex formation between the biotinylated Nod-factors and fluorescent streptavidin allowed localization of the binding site. At concentrations of less than 10^{-7} M, the tagged molecules bound asymmetrically within a minute to nodulation-competent root hairs, but not to root hairs of the non-host *Arabidopsis thaliana*.

Early cellular events within root hairs were studied by loading root segments with the calcium indicators Fura-2 and Fluo-3. Fluorescent ratio imaging showed that addition of NodNGR factors provoked almost immediate, plateau-like increases in intra-cellular free calcium in root hairs and other epidermal cells. Confocal laser-scanning microscopy revealed that calcium accumulation was concentrated at the tips and sides of responsive root hairs.

A gene encoding a lipid-transfer-like protein (LTP2) was isolated from a *Vigna unguiculata* root-hair cDNA bank. Levels of LTP2-transcript increased in root hairs 24 h after treatment with NGR234 or its Nod-factors. The LTP2-gene was cloned into the pMal™ expression vector, and LTP2 purified by affinity chromatography. It was unable to transfer phospholipids between liposomes and mitochondria. Anti-sense analysis in which the LTP2 coding region was cloned between the 35S promoter and terminator sequences reduced nodulation when transformed into *V. unguiculata*.

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rRNA-based identification and detection systems for rhizobia and other bacteria.

Ribosomal ribonucleic acids are excellent marker molecules for elucidation of bacterial phylogeny; they also provide useful target sites for identification and detection with nucleic-acid probes. Based on the currently available 16S rRNA sequence data, bacteria of the rhizobial phenotype fall into three moderately related phylogenetic sub-groups of the α -subclass of the *Proteobacteria*: the rhizobia group, the bradyrhizobia group, and *Azorhizobium*. All rhizobia, azo-, sino-, and brady-rhizobia are closely related and in some cases phylogenetically intermixed with non-symbiotic and/or non-N₂-fixing bacteria, i.e. *Agrobacterium*, *Chelatobacter*, species of *Blastobacter*, *Brucella*, *Mycoplasma*, species of *Nitrobacter*, *Ochrabactrum*, *Phyllobacterium*, *Rhodopseudomonas palustris*, and strains of *Zoogloea*. Especially in the case of *Bradyrhizobium japonicum* strains, the 16S rRNA sequence data indicate substantial heterogeneity. Taxonomic evaluation and revision of strains and/or species of *Rhizobium*, *Bradyrhizobium* and related genera are needed.

Specific probes have been designed and evaluated for the whole rhizobia group. A multiprobe concept for resolving specificity problems with group-specific probes was applied. Given that probe-target sites are usually comprised of only 15 to 20 nucleotides, in some cases it is not possible to design specific probes that exclusively hybridize to the target nucleic acids of the members of a given phylogenetic group of organisms, without binding to nucleic-acid target sites of non-related organisms. These common target sites in non-related organisms can be the result of multiple base changes during the course of evolution. In other cases, it is possible only to design probes that do not hybridize to non-

related organisms, but which also do not react with all members of the target group. These problems can be circumvented by designing and applying multiple probes of complementary or nested specificities. A set of probes was evaluated for the specific and reliable detection and identification of the whole rhizobia group. *In-situ* identification of rhizobia in cultures, as well as rhizobia and cyanobacteria within plant material, was achieved using such group-specific probes in combination with confocal laser-scanning microscopy.

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Contributions and limitations of symbiotic N₂-fixation in common bean (Phaseolus vulgaris) in Romania.

The effects of inoculation of common bean with 19 rhizobial strains were studied over 17 years (1977-1994) in several locations of distinct pedoclimatic conditions in Romania. Grain yield was significantly influenced by strain, year, and location. The average yield increase to inoculation ranged from 6 to 20%; four strains proved to be more stable than the others in their performance, with increases of greater than 10% in all trials.

Linear regressions demonstrated a limiting effect of average temperature on grain yield, and an interaction between rhizobial strain and increasing N application demonstrated that N₂ fixation can satisfy the crops needs. Host cultivar x strain interactions were observed for amounts of N fixed.

Studies on rhizobial strain diversity, using plasmid profile, isoprotein pattern and RFLP, indicated the presence of three groups: *R. leguminosarum* bv. *phaseoli*, *R. tropici*, and *Rhizobium* sp. (*Cicer*).

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Field assessment of response to inoculation with Rhizobium leguminosarum strains in three Egyptian winter legumes.

Indigenous rhizobia in Egyptian soils that nodulate common bean (*Phaseolus vulgaris*), berseem clover (*Trifolium alexandrinum*), and lentil (*Lens culinaris*), were characterized using intrinsic antibiotic resistance patters, plasmid profiles, serological markers, and REP-PCR fingerprinting. The large proportion were extremely poor at fixing N₂, emphasizing the need to increase the numbers of efficient strains by application of high-quality inoculants containing strains able to survive and compete successfully with indigenous strains for nodulation sites.

In field studies, inoculant strains occupied only 12 to 24% of nodules on lentil; estimates of N fixed by the isotope-dilution method ranged between 127 and 139 kg N ha⁻¹ with no effect from inoculant application. In contrast, with berseem clover, inoculant strains competed successfully and occupied 52 to 79% of nodules; in response to inoculation, estimates of N fixed increased from 118 kg N ha⁻¹ to 162 to 205 kg N ha⁻¹, with three cuts.

Preliminary results with common bean also indicate a potential for improvement in N₂ fixation, by application of inoculants containing efficient strains.

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N₂ fixation and nodule occupancy by native strains of Rhizobium on different cultivars of common bean (Phaseolus vulgaris L.).

A field experiment under rainfed conditions was conducted in Durango, to assess N₂ fixation by three cultivars of common bean using ¹⁵N methodology. In addition, diversity of rhizobial isolates from nodules was evaluated by intrinsic antibiotic resistance (IAR), ERIC-PCR, PCR-RFLP analysis of the 16S rRNA gene, and multilocus enzyme electrophoresis (MLEE). Selected isolates were used to determine acetylene reduction activity (ARA) and competitive ability under greenhouse conditions.

The three cultivars showed little variation in N₂ fixation: %Ndfa values ranged from 19 to 26. Variation in ARA among the native rhizobial isolates was high, and our data indicate differences also in competitive ability. PCR-RFLP of the 16S rRNA gene and MLEE revealed that most of the isolates are *R. etli*. IAR and ERIC-PCR showed high, whereas MLEE showed low, genetic diversity among isolates.

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Use of marker genes in competition studies of Rhizobium.

In studies of rhizobial competitiveness, marker genes have several advantages over traditional approaches. Reporter genes such as those for β-glucuronidase (*gusA*) or thermostable β-glucosidase (*celB*) allow detection of rhizobial strains in nodules still attached to the root. Analysis is simple and rapid, permitting high data throughput.

Data from nodule-occupancy studies using *gusA*-marked strains are presented. Moreover, strains separately marked with *gusA* and *celB* allow competitive-interaction studies with two or more strains, and analysis of dual occupancy.

This methodology requires no sophisticated equipment, therefore a GUS Gene Marking Kit was developed and provided to Research Contractors and Agreement Holders.

2. FAO/IAEA Research Co-ordination Meeting on The Use of Irradiated Sewage Sludge to Increase Soil Fertility and Crop Yields and to Preserve the Environment (D1-50.04). Cairo, Egypt, 14-18 September 1996.

Scientific Secretary: Christian Hera

The Second Research Co-ordination Meeting on the agricultural use of irradiated sewage sludge took place in Cairo, with Dr. Rawia El Motaïum, a Research Contract Holder, acting as the local organizer. The RCM was attended by eleven Research Contract Holders and five Research Agreement Holders.

During the first two days of the meeting, the Research Contract and Agreement Holders presented the results of their past year's research. In the last two days, a round-table discussion was organized to elaborate workplans for the coming period. During a one-day field trip, participants visited a ⁶⁰Co source and Electron Beam Accelerators for sewage-sludge irradiation at the Egyptian Atomic Energy Authority

Research Centre in Cairo, as well as an 80-year-old sewage-sludge plant near Cairo that provides solid and liquid sludge to a 2,500-ha farm supervised by the Egyptian Ministry of Agriculture. The farm visit demonstrated the potential of sewage sludge for increasing soil fertility and sustaining high crop production.

Phase I of the CRP has been completed by all Research Contract Holders participating at the second RCM. This phase included studies to quantify the availability of N to crops from sewage sludge applied at various rates, to assess crop-yield increases from sewage-sludge application, and to quantify decreases in contamination of soil by pathogenic organisms as a result of irradiation of sewage sludge compared to non-irradiated. In general, promising results were presented and satisfactory progress described in developing management practices for the efficient use of sewage sludge as an organic fertilizer for increasing crop production in an environmentally friendly manner. A full report of this meeting is available upon request.

Excerpts from presented reports:

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Comisión Nacional de Energía Atómica

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Evaluations for safe re-use of irradiated sewage sludge to preserve the environment.

The bioavailability of N from sewage sludge to sugarcane was assessed indirectly by simultaneously applying 20 kg N/ha as ¹⁵N-labeled fertilizer. Also the contamination of soil with pathogenic micro-organisms was examined, as was heavy-metal pollution due to the use of sludge.

The experiment was performed near Tucuman City (northwest Argentina) where an irradiation plant is under construction. Dried sludge from a purification plant in Tucuman was irradiated in Buenos Aires at the Ezeiza Atomic Center, and then applied in the field at Tucuman.

The N-bioavailability study was carried out on ratoon sugarcane with three levels of irradiated (3 kGy) and non-irradiated sewage sludge, equivalent to 90, 180, and 270 kg N/ha (3.2 to 9.6 t dry matter/ha). Plots were applied also with (NH₄)₂SO₄ labelled with ¹⁵N at 10% a.e.

The ¹⁵N-enrichment data from leaves sampled at the fifth month indicate poor fertilizer recovery, inversely correlated with the amount of organic amendment. Plant N contents were low; the highest levels of sludge (300% of the recommended application of N as chemical fertilizer) failed to satisfy the crop's needs, indicating a necessity to apply it to the soil well in advance of planting to allow time for nutrient release.

Irradiation of sewage sludge decreased the average total coliform count from 1.1 x 10⁵ to 4.4 x 10³/g, and the faecal coliform count fell from 1.6 x 10³/g to zero. At final harvest, seven months after planting, faecal coliforms were undetected in the soil of plots treated even with the highest level of unirradiated sludge. For the heavy-metal contamination work, atomic absorption spectrometry was found to be insufficiently sensitive, therefore neutron activation analysis and particle-induced X-ray emission are now being used.

Ferdinand Koch

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Sewage sludge utilization - Moedling case study.

The objective of the Moedling case study is the development of methods for the safe use of sewage sludge as a component of sustainable agriculture. There are two chief aspects of the programme:

the documentation of sludge quality and of the composting process, and the quantification of effects of sludge on soil and crop when used as organic fertilizer.

Currently the quality of the sewage sludge from the Moedling wastewater-treatment plant meets legislated standards for agricultural use in Austria. However, there are new guidelines to be met by 2004; concentrations of Pb, Cr, Ni, and Cd must be decreased. Levels of organic pollutants are well below current reference values.

As a result of composting dewatered sewage sludge with addition of straw, Hg concentration decreased by 20% whereas increases of 20 to 30% occurred in Zn, Cu and Co. Although the recommended duration of temperatures in excess of 50°C was not achieved, numbers of *Enterobacteriaceae* fell to almost zero.

Since 1993, raw and composted sewage sludge have been applied to a chernozem soil at the regulation rate of 2.5 t dry matter/ha/year. Maize was grown in 1994, and barley in 1995 at which time the soil was analyzed. No general tendencies were observed in terms of accumulation of heavy metals or of organic pollutants - their concentrations in the soil remained below recommended limits. In the maize, there was no significant accumulation of heavy metals above background levels, and polycyclic aromatic hydrocarbons were at concentrations typical of plants grown in non-polluted soil; dioxins were below detection limits as were organochlorine pesticides except lindane. Crop growth was improved by addition of the organic fertilizers: maize yields were 70% higher with compost addition than in unfertilized plots.

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Isotope-aided studies on the effects of radiation-processed sewage-sludge application on crop yields and bioavailability of heavy metals in Bangladesh soils.

Irradiation (5 kGy) of sewage sludge decreased bacterial counts to almost half, and eliminated the presence of pathogenic bacteria and parasites. The application of irradiated and non-irradiated sludges significantly increased wheat yields on a dark grey floodplain soil (Haplaquepts) at Myensingh, but only at the very high rates of 16.5 and 22 t/ha (equivalent to 150 and 200 kg N/ha). This result indicates that sewage sludge alone should not be used as a source of N, but rather it must be combined with chemical fertilizer to maximize yields. ¹⁵N-isotope aided studies showed that the sewage sludge contributed little N to the wheat.

There were only marginal increases in heavy-metal concentrations in sludge-treated soil after wheat harvest, and no clear trend was seen in metal levels in the grain and straw. Grain yields of mung bean planted after the wheat were positively affected by residual effects of the very high levels of sewage sludge.

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Effects of irradiation of sewage sludge on crop yield and nitrogen distribution in soil-plant system

Irradiated and non-irradiated sewage sludge were used in a field experiment on a brown earth heavy-loam paddy soil, at application rates equivalent to 75 to 300 kg N/ha. Results showed that gamma irradiation of sewage sludge at a dosage of 3 kGy increased wheat grain yield by 27% on average, as compared with non-irradiation treatments. Irradiation may have promoted the mineralization of organic

N in the sewage sludge, and thus enhanced crop growth and development. The grain yield attained with irradiated sewage sludge applied at the equivalent of 225 kg N/ha (approximately 5.2 t/ha) equaled that with 150 kg N/ha as chemical fertilizer. It is preliminarily concluded that irradiated sewage sludge can fully substitute for chemical N fertilizer in crop cultivation.

Rawia El Motaium

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Effects of sewage sludge on yields of tomato and citrus.

Experiments were conducted with tomato ('Castle Rock', in the field) and citrus ('Sour Orange', seedlings in pots) using a virgin "pure sand" soil. Sewage sludge was applied at 50, 100, 150, and 200% of the recommended rate for N. Only the highest application rates significantly increased crop yields; recoveries of N were low.

The highest level of sewage sludge resulted in insignificant increases in the concentrations of heavy metals in soil, none of which exceeded the standards recommended by the U.S. Environmental Protection Agency. Gamma irradiation (5 kGy) of sludge significantly decreased the counts of pathogenic micro-organisms.

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Metabolic fate of sewage-sludge-derived contaminants in plant-cell cultures.

Besides beneficial plant nutrients, sewage sludges contain hazardous pollutants that, with application to soil, can enter the food chain and are, therefore, of public concern. Some 330 organic pollutants have been described in the literature, of which 42 are regularly found in sewage sludge. Here are reported the metabolic fate of 4-nonylphenol (4-NP), various congeners of polychlorinated biphenyls (PCBs), and 1,4 dichlorobenzene (1,4-DCB) in plant-cell cultures.

Wheat-cell suspension cultures were incubated with ¹⁴C-labeled 4-NP, and four major radioactive fractions were isolated on Sephadex LH-20 and semipreparative HPLC. Their chemical structures were elucidated by enzymatic hydrolysis, HPLC, GC-MS, and ESI-MS/MS: they were 4-(hydroxy)- and 4-(dihydroxy)nonylphenols glucosylated at the phenolic OH-group and further glucosylated, glucuronidated, and acylated with acetic or malonic acid.

The metabolism of ten PCB congeners was tested in cell cultures of 12 plant species. The rates of metabolism were strongly species-dependent. Except for PCB 15 (4,4'-dichlorobiphenyl) a lower chlorination grade was associated with higher metabolism rate. The incubation of wheat-cell cultures with ¹⁴C-PCB 52 (2,2',5,5'-tetrachlorobiphenyl) yielded four monohydroxylated and three dihydroxylated metabolites.

Uptake, metabolism and phytotoxicity of 1,4-DCB in cell cultures of four species were investigated using a sealed system because of the substrate's volatility. 1,4-DCB was taken up by the cells of carrot (49%), soybean (50%) and *Chenopodium rubrum* (62%); extreme toxicity for tomato precluded estimation of uptake. Only the soybean cultures metabolized the compound, probably to conjugates of chlorophenols. A concentration of 0.5mM caused a 50% inhibition in the growth of the carrot and soybean cells, whereas 0.05 mM inhibited the growth of tomato cells by 50%.

These data indicate that hazardous organic compounds that may be present in sewage sludges can be taken up and metabolized by plants.

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Effects of sewage-sludge amendment on a maize/legume cropping system.

Raw and irradiated (5 kGy) sewage sludge were applied to a clay-loam inceptisol of pH 6.7, to provide organic sources of N at 50, 100, 150 and 200% of the equivalent of the recommended application (60 kg N/ha as ammonium sulphate). Residual effects of the sludge treatments were appraised with a follow-up crop of green gram, for which N₂ fixation was quantified using mustard as the non-fixing reference.

The sewage sludge applications had no effects on the growth or grain yields of maize or green gram. Yields with ammonium sulphate were at par with the sludge treatments. There were no significant trends in terms of heavy-metal levels, as accumulated by the crops or residual in the soil. Efficiency of use of sludge N will be estimated by the indirect method after completion of total-N and ¹⁵N analyses of the plant samples.

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The effects of untreated and irradiated sewage sludge on yields of Capsicum annum.

Irradiated (3.6 to 4.2 kGy) and untreated sewage sludge were applied to pepper plants (*Capsicum annum*) on an oxisol, at rates of 6 to 24 t dry matter/ha, equivalent to 50, 100, 150 and 200% of the recommended application of N (120 kg N/ha). The organic fertilizer stimulated crop growth such that the 18 and 24 t/ha treatments produced yields equal to that obtained with 120 kg N/ha applied as chemical fertilizer. The non-irradiated treatments consistently yielded more than the irradiated, although the differences were not significant. After crop harvest, soil Zn levels were slightly higher in sludge-treated plots, whereas the soil concentrations of other heavy metals were not increased by sludge application. Heavy-metal contents of the plants were low, with Ni, Cd, Pb, Cr, and Co undetectable. After harvest, counts of pathogenic micro-organisms in the soil of non-irradiated sludge-treated plots were at background levels.

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Recent research on agricultural use of sewage sludge in Japan.

The properties of composted sludge were improved by mixed fermentation with cattle waste. This "advanced compost" had a better balance of N, P, and K, and a 1:1 mix had the most beneficial effects on plant growth.

The analysis of results of two long-term experiments revealed the following relationship for accumulation of heavy metals in soils from sewage sludge:

$$m_n = (m_{n-1} + M_n)e^k$$

where m_n is the metal content in soil just before the n^{th} sludge application, M_n is the metal content increment carried by the n^{th} sludge application, and k' is the decrease rate-constant which depends on pH and other soil characteristics.

Sequential analyses have shown that heavy metals gradually change form in soil, from soluble to less soluble to insoluble. Absorption of Zn by plants decreased with time reflecting changes in solubility.

Using the $\delta^{15}\text{N}$ method, it was shown that N from sewage sludge is readily absorbed by plants early in the process of decomposition. And a series of lysimeter experiments has demonstrated that nitrate leaching can be minimized by replacing about half of the chemical fertilizer N with composted sewage sludge or mixed sludge/manure compost, without compromising crop yields.

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The use of irradiated sewage sludge for sustainable crop production in Malaysia.

Sewage sludge, irradiated (25 kGy) and non-irradiated, was applied at 10.5 to 42 t dry matter/ha, equivalent to 150, 300, 450 and 600% of the recommended chemical fertilizer N rate (140 kg N/ha) for maize. Total dry matter yields were increased by the application of sludge up to the 300% level, but high variability prevented statistical significance. Irradiation of sludge did not alter yield responses. The N-utilization of the recommended fertilizer rate was 61%, whereas most of the sludge treatments showed < 10% N-utilization, indicating that relatively small amounts were available to the first crop - hence the need for 21 t/ha to maximize positive effects on yields. The application of sludge did not significantly affect concentrations of heavy metals in the plants in comparison with those grown with chemical fertilizer only.

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Evaluation of treated sewage sludge added to agricultural soils in Mexico: heavy-metal effects on soil microbial biomass and activity.

Sewage sludge, non-irradiated and irradiated (8.7 to 12.8 kGy), was applied to a vertisol at rates equivalent to 130, 260, 390 and 520 kg N/ha prior to planting maize; the recommended rate for chemical fertilizer N is 260 kg N/ha. The concentrations of heavy metals in the sludge and sludge-amended soils were within the limits recommended by the European Union.

Soil respiration, a measure of microbial activity, was increased six- to eleven-fold by sludge treatment (both irradiated and non-irradiated). The highest levels of application of sewage sludge gave biomass C levels (expressed as a percentage of soil organic C) that were higher than equilibrium values.

The least application of sludge (equivalent to 130 kg N/ha) gave grain yields (non-irradiated 4.96 t/ha, irradiated 4.79 t/ha) superior to that obtained with the recommended rate of 260 kg N/ha (4.34 t/ha). Higher applications of sludge further increased grain yields, by up to 25%.

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Utilization of sewage sludge for enhancing agricultural productivity in Pakistan.

Applications of sewage sludge, irradiated (5 kGy) and non irradiated, were made to experimental plots as organic fertilizer for wheat (cv. Inquilab), at rates that were equivalent to 120, 180 and 240 kg N/ha. Yields were compared with control treatments of 20 and 120 kg N/ha. The sludge applications were combined with ¹⁵N-labelled fertilizer N at 20 kg N/ha (10% a.e.) so that N contributed by the organic fertilizer could be assessed by the indirect method.

Even the least of the sludge applications improved yields over that obtained with the 20 kg N/ha control, but the yield with the highest application, 16 t dry matter/ha, was less than that with 120 kg N/ha. Irradiation greatly reduced coliform counts, but did not influence yield effects.

Only about 16% of the applied organic N was assimilated by the wheat. A marked residual effect, which was particularly striking with the non-irradiated sludge treatments, was seen in terms of improved dry matter and total N production by a subsequent green manure crop, *Sesbania aculeata*.

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The use of sewage sludge as a fertilizer in pastures.

An irradiated (6.2 kGy) urban sewage sludge, collected from drying beds, was used for a field and a pot experiment with a calcareous soil, pH 7.2. The sludge was applied at rates of 5, 10, 20 and 50 t/ha (equivalent to 133 to 1,330 kg N/ha), and an 80 t/ha treatment was included in the pot experiment. *Lolium multiflorum* cv. Prima and inoculated *Trifolium subterraneum* cv. Clare were used in combination in the field, but sown separately in the pots. ¹⁵N at 5% a.e. was applied at 20 kg N/ha, one month after sowing or immediately after each cut (field: two cuts, pot: three cuts).

In both experiments, clover growth decreased as the rate of sludge increased, with the exception of the lowest application in the pot experiment. In marked contrast, the shoot dry weights of the grass increased with sludge rate. The estimates of %N derived from fixation by the clover decreased as sludge rate increased; a chemical P+K treatment without sludge gave the highest estimates of N fixed.

Irradiation drastically reduced the numbers of coliforms in the sewage sludge, and eliminated fungi.

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The use of irradiated wastewater sludge cake as a source of nitrogen for cropping systems.

A field experiment using ¹⁵N was conducted on a Reddish Brown Lateritic soil, to compare the effects of chemical and organic sources of N on growth, yield and N-uptake by maize. Applications of irradiated (2 kGy) and non-irradiated wastewater sludge cake were made at rates equivalent to 30, 60, 90 and 120 kg N/ha each with fertilizer N at 20 kg N/ha enriched at 10% a.e.

Although growth and yields of maize tended to increase with the rate of sludge applied, none of the organic treatments gave grain yields equal to that obtained with chemical fertilizer N at 60 kg N/ha. No yield effects could be assigned to the radiation treatment.

Heavy metals, pathogens and soil properties were determined; no significant differences were found between sludge-treated and non-sludge-treated soils. ¹⁵N analyses are pending.

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Cd solubility and phyto-availability in sludge-treated soil.

Sewage sludge was applied to experimental plots at 0, 22.5, 45, 90 or 180 Mg/ha/yr for six years, at which time half of the plots continued to receive these rates and sludge application to the other half was terminated. All plots were then planted to Swiss chard (*Beta vulgaris*) twice per year for 10 years, with soils sampled after each harvest.

When sewage sludge applications were terminated, the soluble Cd concentrations of treated plots were higher than those of control plots, and did not decline for 10 years. With continued sludge application, soluble Cd concentrations increased with each input for the same 10-year period. With continuous sewage-sludge inputs, organic-C contents continued to rise, and after termination organic-C contents decreased only slowly. There was no indication of increase in soluble Cd concentration or in phyto-availability of Cd in the sludge-treated soils when the organic C decreased after termination of sludge application.

Steve P. McGrath

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UK

Comparison of outcomes after acute or chronic exposure of soil bacteria to toxic metals.

The responses of soil-borne bacteria to heavy metals that may be found in sewage sludge differ depending on the nutritional status of the organism. In nutrient culture, differences between rhizobial strains in sensitivity to Zn and Cd were apparent in terms of growth rate, whereas in nutrient-poor conditions only the tolerant strain survived.

Most ecotoxicology tests are of short duration, whereas there is evidence that, in soil, moderate exposure levels may have no effect within a 12-month period. One hypothesis to explain this is that the mechanism of toxicity depends on a replication-related process; in the nutrient-limited conditions that usually prevail in soil, mean turnover rates of microbes are of the order of 12 months. In soil, small differences in growth would eventually lead to competitive dominance by strains able to maintain higher growth rates. Such outcomes cannot be predicted from short-term toxicity tests that are performed in the absence of competition. It is difficult to make any ecotoxicity test simulate what happens in soil over a long time-period

3. **FAO/IAEA Research Co-ordination Meeting on The Use of Isotope Techniques in Studies on the Management of Organic Matter and Nutrient Turnover for Increased Sustainable Agricultural Production and Environmental Preservation (D1-40.08). Vienna, 7-11 October 1996.**

Scientific Secretary: Chris van Kessel

The first Research Co-ordination Meeting for this CRP on soil organic matter and nutrient cycling involved twelve Contract Holders and two Agreement Holders. Staff members from the Section and the Seibersdorf Laboratory also attended. Following introductory remarks by the Director of the FAO/IAEA Joint Division, Dr. J.D. Dargie, each participant gave a talk on the current status of their research on soil organic matter and the use of organic amendments in crop production in her/his country. This was followed by a series of lectures on nutrient cycling in cropping systems, in particular N and C, and how the use of isotopes can contribute to a better understanding of underlying mechanisms.

A series of common experiments, to be carried out by the Contract Holders, was discussed in detail. The main objectives are to increase nutrient-use efficiency by crops and to improve cropping-system capability in retaining nutrients in organic amendments. Implementation of the research has already been initiated by some of the Contract holders.

Excerpts from presented reports:

A.B. Rosenani

Department of Soil Science
Universiti Pertanian Malaysia
Malaysia

Utilization of organic fertilizers and crop residue for sustainable maize production and effects on soil organic matter.

Malaysian highly weathered acid soils are low in soil organic matter and cation exchange capacity. Thus, in recent years, organic materials, as a source of nutrients and soil amendments (in combination with chemical fertilizers), have been recommended together with crop-residue incorporation. Palm oil mill effluent (POME) is a major waste product of the palm oil industry and is a threat to the environment. However, due to its high nutrient content, POME has been converted to an organic fertilizer. It is now recommended for use in field-crop cultivation, such as maize and groundnut. Another important and popular organic fertilizer in Malaysia is poultry waste (chicken dung), commonly used by vegetable farmers. As it is easily and abundantly available, it is also recommended for other crops, including maize.

Several studies had been conducted to show the beneficial effects of POME and chicken dung on crops, but these merely gave yield-performance data and effects on soil parameters to indicate improvement in soil fertility. Understanding is greatly lacking of organic-matter turnover in our highly weathered acid soils. In order to more efficiently manage organic-matter application to crops, we need to know more about the dynamics of soil organic matter in these soils by quantifying some of the related soil and plant parameters. Therefore, this proposed project using isotope techniques is important. The project protocol involves the following treatments, (i) POME + glyricidia + 50% recommended (rec) rate of chemical fertilizer N, (ii) POME - glyricidia + 50% rec N, (iii) chicken dung and paddy straw mixture + 50% rec N, (iv) chicken dung + 50% rec N and (v) 100% rec N. Each treatment (replicated four times) will consist of subplots with or without crop residue incorporation and microplots for ¹⁵N-labelling. Maize will be planted in rotation with groundnut. All treatment plots will be limed and receive recommended rates of P and K fertilizers.

Jia Yu Wang
Institute of Soils and Fertilizers
Zhejiang Academy of Agricultural Sciences
Hangzhou
P.R. China

Long-term isotope-tracer studies on soil organic matter in Chinese paddy soils and British arable soils.

The general objective of this research is to understand the mechanisms that govern organic matter (OM) dynamics in different climatic and soil conditions. Long-term changes in soil OM monitored using an isotope may ultimately lead to recommendations for soil OM management that will assist in attaining sustainable soil fertility and crop production.

D.S. Powelson
IACR-Rothamsted
Harpenden
Herts.
U.K.

The influence of straw incorporation and soil type on N losses.

The incorporation into soil of cereal straw having a wide C:N ratio (typically about 80) can cause immobilization of inorganic N as the soil microbial biomass proliferates. In the past this has been regarded as a potential problem, as immobilization decreases the amount of N available for the subsequent crop. However, many arable soils in northwest Europe contain an excess of nitrate in the autumn, which contributes to nitrate leaching during winter. Straw incorporation is now seen as a means of decreasing the amount of nitrate at risk to leaching.

An experiment was conducted at three sites, having clay contents of 14, 26 and 39%. The soil nitrate pool was labelled by application, in September, of either 2.5 kg N ha⁻¹ as K¹⁵NO₃ at 81 atom % excess or 50 kg N ha⁻¹ as K¹⁵NO₃ at 4.6 atom % excess. The treatments were applied to areas where wheat straw had either been burned or was immediately ploughed in. The next wheat crop was sown shortly afterwards. Visual assessments were made of the extent of soil movement and the positions of the plots (each 2 x 2 m in triplicate) were moved accordingly. This was later checked by analyzing each wheat row individually for ¹⁵N. Soil and crop samples were taken from the central area of each plot in the following April or May.

Where the very small quantity of ¹⁵N had been applied (2.5 kg N ha⁻¹) the *proportion* retained in soil, through immobilization or uptake into plant roots, was greater than for the larger application (50 kg N ha⁻¹) in the sandy and silty soils. This trend was less clear in the clay soil. For the small application, it ranged from about 20% in the sandy soil to 60% in the clay soil. The corresponding values for the larger application were 5-15% in the sandy soil to 35-60% in the clay soil. Incorporation of straw generally increased retention of ¹⁵N in soil. In the sandy soil, retention increased from 4% to 16% for the larger ¹⁵N application; the corresponding values for the silty and clay soils were 14% to 41% and 34% to 61%, respectively. In the silt and clay soils the increased retention of ¹⁵N in soil was reflected by decreased uptake in the crop. This was not so in the sandy soil, perhaps because turnover of organic N is faster in a sandy soil due to less clay protection of microbial metabolites.

In two soils the incorporation of straw decreased the total loss of ¹⁵N from the crop/soil system, but the difference was probably significant only in the sandy soil: a decrease of 82% to 72%. The overall impact of straw incorporation on nitrate leaching would appear to be small. In the long term, continued incorporation of straw will lead to an increased quantity of N (and C) in some soil pools and possibly increased mineralization of N.

M.S.A. Safwat
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Minia University
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Egypt

The effects of organic matter, water management and inoculation on nutrient turnover and biological N₂ fixation using isotope techniques.

Pots and field experiments will be conducted to study the effects of organic matter (e.g. plant residues, compost and animal manure), water management (e.g. surface and sprinkler irrigation) and inoculation on nutrient turnover, micronutrients, biological N₂ fixation, and the synchronization of nutrient release and plant-nutrient demand of various crops. Field experiments will be carried out on newly reclaimed sandy soils at Shosha.

Javier. Z. Castellanos
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Mexico

Soil organic C and N cycling under different crop rotations and tillage practices in an irrigated vertisol in central Mexico.

Vertisols are widely distributed throughout the world. In Mexico, they occupy an important area of irrigated and rainfed agriculture, mainly in the center of the country, that produces one fourth of the wheat and one fifth of the corn and sorghum. Unfortunately, for at least 50 years crop residues have been burned and the soils deeply and frequently plowed. Long-term records indicate that the levels of soil organic C have been reduced to 60 or 70 % of the original value due to these practices. Loss of organic matter in these soils has decreased the organic-N-supplying capacity to only about 20 to 40 kg N/ha per crop. Thus, these soils have severely decreased their capacity to mineralize N for crop production, to support an active microbial biomass for nutrient retention and cycling, to retain applied pesticides, and they also have lower water-holding capacity. An important additional problem is the leaching of nitrates from fertilizer N that is applied to overcome N deficiency. Leaching of nitrate into groundwater decreases the quality of the water pumped from wells that supply drinking water.

The current practice is to produce two crops per year under irrigated conditions, and to burn the residues of both to facilitate plowing. An effective alternative is proper management of the crop residues by using conservation tillage, including no till. Return of crop-residue C and the judicious use of N fertilizer should begin rebuilding the organic content and the fertility of these important soils. Currently, very few studies have been conducted to assess the effects of conservation tillage on increasing soil organic matter in the vertisols of Mexico. No studies have been conducted using ¹⁵N/¹⁴N or ¹³C/¹²C isotope-ratio technology to measure the effects of conservation tillage on N-cycling dynamics, fertilizer N-use efficiency, and crop-residue C sequestration. Such information should be of direct benefit to Mexico and other regions with similar conditions.

Phan thi Cong
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Vietnam

The use of isotope techniques in studies on management of organic matter for sustainable agricultural production in south Vietnam.

Due to intensive and prolonged weathering, most of the soils in Vietnam are poor in nutrients. Decline in soil organic matter (SOM) content after deforestation causes severe reduction in the mineralization of nutrients for crops and in the nutrient-sorption capacity of the soil, and threatens the collapse of weakly-structured soils such as Acrisols. Decrease in SOM, therefore, threatens the sustainability of upland ecosystems.

The objectives of this study are: i) to increase the quantity of nutrients available to crops with a judicious mixture of inorganic and organic sources, ii) to assess the effectiveness of organic residue and inorganic fertilizers on the building of SOM and the release of plant nutrients through isotopic labelling, iii) to investigate whether soil organic fractions can be used as early and sensitive indicators of sustainability in upland ecosystems, and iv) to improve process-level understanding of C and nutrient flows by the use of isotopic techniques and computer models, so that management recommendations can be extrapolated to a wide range of environments.

In order to achieve these objectives, the following activities will be implemented: 1) install microplots with ^{15}N , and doubly-labelled (^{15}N - ^{14}C) plant material into existing field experiments for C-dynamics monitoring; 2) investigate the decomposition and transformation of different organic sources into soil organic matter; 3) study the chrono- and topo-sequence with clear-cut C_3 - C_4 transitions using ^{13}C ; and 4) look into the transformation of P in soils with low P-availability (Haplic Acrisol and Rhodic Ferralisol) using ^{32}P -labelled fertilizer and plant material.

Erick Zagal
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Chillan
Chile

Decomposition of ^{15}N and ^{14}C labelled residues in volcanic soils of the Chilean central plain.

The central plain is the main agricultural region in Chile, producing wheat, maize, common bean, potato, and sugar beet. The soil types are of alluvial or volcanic origin, with volcanic-ash-derived soils (Andepts and Andosols) dominating the latter. Studies on organic-matter dynamics show that the allophane content in ash-derived soils inhibits the degradation of added carbonaceous materials as compared to non-allophanic soils. The mechanisms are unclear.

A project is proposed with the general aim of studying decomposition of ^{15}N - and ^{14}C -labelled residues in volcanic soils under irrigation, with different crop rotations and levels of fertilization. Specific aims are: i) to determine rate of decomposition of labelled materials of different quality (*i.e.* maize stems and roots); ii) to evaluate possible nutrient limitations (N, P) in the process of decomposition; iii) to quantify gross mineralization-immobilization rates.

The approach will be a field experiment located on a volcanic soil in an irrigated valley with a Mediterranean climate, complemented with laboratory incubations. Microplots will be installed within larger plots of an established study of rotations with two levels of fertilization. The levels of fertilization are: high and medium. Four treatments (rotations) with and without livestock are considered: i) Sugar beet / wheat / red clover / red clover with livestock; ii) Sugar beet / wheat / common bean / barley without livestock; iii) Maize / wheat / red clover / red clover with livestock; iv) Maize / wheat / common bean / barley without livestock.

Maize plants are being labelled in rotations iii) and iv) during spring season 96 (first week of November). Winter-wheat plants will be labelled during autumn 97 (April/May). After harvest, labelled material is applied to microplots different from those used for labelling. ^{15}N -labelled maize residues are to be applied during autumn 1997 before winter wheat. ^{15}N -labelled wheat residues are to be applied during autumn 1998 before clover. Doubly-labelled ($^{14}\text{C}/^{15}\text{N}$) wheat residues are to be added in 1-2 kg plastic tubes within the microplots. Samples of plants, soil and remaining residues will be collected at 3 weeks, 3, 6 and 12 months, and for the duration of the experiment. Collected materials will be analyzed for total N, ^{15}N , total C and ^{14}C . Determinations of soil biomass and inorganic N will be made.

S. M. Rahman
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Mymensingh
Bangladesh

Organic matter management for increased and sustainable agricultural production in Bangladesh.

For several years, agronomic experiments (non-isotopic) were conducted in a grey floodplain soil (Haplaquepts) in Bangladesh to study tillage and manuring effects on crop yields, soil physical properties, and water relations. The objectives included studies on the effects of organic manures (crop residues and decomposed farmyard manure, FYM) incorporated into the soil alone or in combination with chemical fertilizers under different tillage practices, on soil organic matter (SOM), nutrient status, and crop yield. A neutron probe was used to study the water relations at various stages.

Results indicated no significant effect on wheat yield due to tillage practice. The highest yield was recorded with the locally recommended dose of NPKSZn fertilizers, followed by treatments receiving FYM or rice straw each at 6 t ha⁻¹ along with half of the recommended fertilizer. FYM and/or rice straw applied singly or in combination did not produce comparable yields. Residual effects of manuring were not observed in summer mung bean, but subsequent rice yield increased considerably without manure and/or chemical fertilizer except urea, indicating a substantial contribution of nutrients from manuring and tillage practices performed on the first crop (wheat). Soil water-holding capacity also showed no significant changes, remaining between 45 and 55%.

SOM was always highest at the 0-1.0 cm depth, and considerably decreased at 20-30 cm. It would be premature to discuss changes in SOM at the end of only a three-year study, although it has been reported that SOM declines dramatically when tropical soils are continuously cultivated.

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Piracicaba
Brazil

Geostatistical studies applied to Amazonian soils: organic matter related properties in a forest-pasture succession.

This landscape-scale study used geostatistical methodologies to analyze and characterize the spatial variability of parameters related to soil organic matter in a forest-pasture succession, in the State of Rondonia, Brazil. The great majority of soil-nutrient storage estimates on a global or regional scale are based on the extrapolation of averages considering soil classes or categories and vegetation types. There is much uncertainty in these estimates. The use of Geographic Information Systems (GIS) is improving estimates, but there is still need to consider the spatial variability within each soil unit. Geostatistical characterization of several physico-chemical properties of the soil, which is the scope of this project, will certainly improve the data. This information should bring better understand the dynamics of organic matter in Amazonian forest-pasture systems, in order to enhance soil productivity and assure sustainability.

This first report presents a general overview of the C cycle, and details for sugarcane and pasture systems. Sampling locations, scales and schemes are shown in relation to geostatistical analyses that will be used in future. Preliminary results are presented in the form of semivariograms, for some of the measured soil properties, e.g. pH, bulk density, ¹³C, moisture, total C, and exchangeable cations.

Denis Amara
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Institute of Agricultural Research
Freetown
Sierra Leone

Studies on organic matter turnover and build-up in ultisols and oxisols in Sierra Leone.

The majority of the upland soils in Sierra Leone are ultisols or oxisols, which are generally low in fertility as a consequence of intensive weathering and leaching of nutrients by heavy rainfall. The soil-fertility problem is exacerbated by shortening of the bush-fallow period and increasing demands on agricultural land. Despite this, very few farmers use fertilizers to replenish soil fertility, and even the large amounts of crop residues obtained at the end of the cropping season are not properly managed as a source of nutrients and soil organic matter.

Since crop residues and the prunings of multipurpose trees are potential sources of nutrients and organic matter for cropping systems, the following experiments have been designed to provide an understanding of how best they may provide nutrients and build up soil organic matter: (i) improve residue management in a crop rotation, (ii) determine the N uptake from organic (crop residues) and inorganic sources, and (iii) determine the decomposition of ¹⁵N and ¹⁴C labelled crop residues and prunings of multipurpose trees. It is anticipated that results obtained from this research will provide guidance in the management of crop residues and tree prunings to maintain the physical, chemical, and biological health of soils in the upland agro-ecosystems of Sierra Leone.

Ravi Sangakkara
Faculty of Agriculture
University of Peradeniya
Sri Lanka

Management of soil organic matter to enhance soil productivity and sustainability in Sri Lanka.

The agricultural production systems of Sri Lanka consist of two well-defined components. One is the plantation sector established during the colonial era, and the other is the smallholder subsistence sector producing food crops. While the traditional Sri Lankan agricultural system was based on organic manures and amendments, the advent of the green revolution and the availability of chemical fertilizers at low prices enhanced the use of agro-chemicals at the expense of recycled organic matter. This has led to depletion of soil organic matter, especially under humid tropical conditions, resulting in low productivity of current chemical-based agricultural sectors, along with low soil fertility and lack of sustainability of farming systems. Furthermore, farmers using organic matter apply different rates haphazardly, leading to the inefficient use of added materials.

4. FAO/IAEA Research Co-ordination Meeting on The Assessment of Soil Erosion through the Use of ¹³⁷Cs and Related Techniques as a Basis for Soil Conservation, Sustainable Production, and Environmental Protection (D1-50.05). Vienna, 11-15 November 1996.

Project Officer: Felipe Zapata

The first Research Co-ordination Meeting was implemented as scheduled, and will be reported in the next issue.

5. Workshop for the Regional Project for Asia and the Pacific on Nuclear Techniques for the Promotion of Agroforestry Systems (RAS/5/029). Faisalabad, Pakistan, 28 July-1 August 1996.

Scientific Secretary: C. van Kessel

The second workshop for this regional project involved counterparts from ten member states. The first two days were spent on presentations by counterparts describing progress made since the last workshop, held in Thailand in the spring of 1995. The last days of the workshop were devoted to discussion and finalization of future plans.

Visits were made to field stations located near Faisalabad and Lahore, and to the laboratories of NIAB and, on the same campus, of the National Institute for Biotechnology and Genetic Engineering (NIBGE).

Excerpts from presented reports:

S.U. Patwary

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Mymensingh
Bangladesh

Nuclear techniques for the promotion of agroforestry systems.

Six legume-tree species were evaluated for N₂-fixing ability, namely *Samanea saman*, *Tamarindus indica*, *Dalbergia sissoo*, *Acacia farnesiana*, *Acacia auriculaeformis*, and *Delonix regia*. The non-fixing reference species were *Swietenia mahogani*, *Terminalia arguna*, and *Azadirachta indica*. Three-month-old inoculated plants were square-planted in the field with a 1-m spacing; fixing and non-fixing trees were planted alternately. ¹⁵N-labelled fertilizer (10% a.e.) was applied in three split doses for a total of 20 kg N/ha as (NH₄)₂SO₄. Each 36-m² isotope sub-plot was separated from guard areas by placing two layers of polythene into the soil to a depth of 1 m.

All plants in the isotope sub-plots were harvested after 8 months. Leaves, stems+branches and roots were separated for determination of dry weight, total N and ¹⁵N enrichment. *S. saman* and *A. auriculaeformis* were of maximum height and canopy size, and yielded well in terms of dry matter and total N, although *A. farnesiana* gave higher dry weight than *S. saman*. The highest estimates of N₂ fixation were obtained with *S. saman* (59 to 71% depending on reference species, mean value of 66% of N derived from fixation). The next highest was *D. sissoo* for which 48% of N was derived from fixation. Therefore, *S. saman* shows promise for use in agroforestry.

Zhongze Zhang

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A comparison of N₂ fixation by trees for use in low-fertility and drought-stressed soils in China.

We selected for study five species already in agroforestry use: two legumes, *Robinia pseudacacia* and *Amorpha fruticosa*, and three N₂-fixing non-legumes, *Hippophae rhamnoides*, *Alnus tinctoria* and *Eleagnus angustifolia*. And four species were chosen as non-fixing references: *Gleditsia sinensis*, *Prunus persica*, *Ligustrum japonicum*, and *Syringa oblata*. Three split doses of fertilizer N enriched with ¹⁵N at 10% a.e. were applied, for a total of 20 kg N/ha. The trees were harvested at 11 months after transplanting.

H. rhamnoides and *E. angustifolia*, which are drought-tolerant, grew slowly and showed no evidence of N₂ fixation; they may perform better if allowed to establish over a longer period of time. *R. pseudacacia* was the superior species in terms of height, dry weight and N₂ fixed.

Elsje. L. Sisworo

Centre for Application of Isotopes and Radiation

Jakarta

Indonesia

The use of ^{15}N to estimate N_2 fixation in trees.

Estimates of fixation were made with six species: *Gliricidia sepium*, *Sesbania sesban*, *Calliandra tetragona*, *Flemingia congesta*, *Acacia mangium*, and *Leucaena leucocephala*. *Eucalyptus alba* was used as the non-fixing reference species. One-month-old seedlings were transplanted in the field (soil pH 5.2), and trees were harvested 10 months later. Three split doses of fertilizer enriched with ^{15}N at 10% a.e. were applied, for a total of 20 kg N/ha.

The highest dry weight accumulations were obtained with *S. sesban* (2.34 kg/plant) and *C. tetragona* (2.11 kg/plant), and the highest estimates of N fixed were with *C. tetragona* (19.9 g N/plant) and *L. leucocephala* (17.4 g N/plant).

Wan Rasidah Kadir

Forest Research Institute of Malaysia

Kuala Lumpur

Malaysia

Identifying tree species for high biomass production and high N_2 -fixation capacity.

The experiment was carried out on a Petroferric Tropudult of pH 3.9. Biological N_2 -fixing capacity of six species was estimated: *Acacia mangium*, *Leucaena leucocephala*, *Gliricidia sepium*, *Parkia speciosa*, *Azadirachta excelsa*, and *Paraserianthes falcataria*. The non-fixing reference trees were *Khaya ivorensis*, *Shorea leprosula*, and *Hopea odorata*. Because of the low pH, lime was added at 2 t/ha. Each plot had an area of 100 m²; ^{15}N applications were made to 36-m² microplots that were trenched and lined with two layers of thick plastic sheeting and covered back with soil. Nitrogen was applied in three split doses for a total of 20 kg N/ha and, at transplanting, 500 g of phosphate rock (13% P) were applied per planting hole. All species were planted randomly in each plot.

At six months after transplantation, the tallest trees were *P. falcataria* (3.6 m) and *L. leucocephala* (2.8 m), and the highest leaf N concentrations were in *G. sepium* (3.4%) and *L. leucocephala* (3.2%). Final destructive sampling is planned for November 1996.

Mazhar H. Naqvi

Nuclear Institute for Agriculture and Biotechnology

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Pakistan

Selection of tree species for high biomass production and high N_2 fixation.

The experiment was conducted at two sites, one of which has a salt-affected soil. The tree species selected were (N_2 -fixing) *Acacia nilotica*, *Acacia ampliceps*, *Albizia lebbek*, *Leucaena leucocephala*, and *Prosopis juliflora*, and (non-fixing reference) *Eucalyptus camaldulensis*, and *Tamarix aphylla*. *T. aphylla* plants were raised from cuttings, and the others from seed. All of the N_2 -fixing plants were inoculated with broth cultures prepared from nodules. Total N contents of the transplants were determined, and ^{15}N -labelled ammonium sulphate was applied in three equal splits for a total of 20 kg N/ha.

Plants in the non-saline soil, including roots, were harvested at 26 weeks after transplanting. *L. leucocephala* produced the highest dry weight (759 g/plant), followed by *A. ampliceps* (538 g/plant). *L. leucocephala* also fixed the most N (6.81 g/plant), whereas the *Acacia* species fixed approximately 1.1 g N/plant, *A. lebbek* 0.97 g N/plant, and *P. juliflora* fixed the least at 0.16 g N/plant. The data from the salt-affected soil are pending.

Reynaldo G. Palis

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N₂-fixing trees for agroforestry in the Philippines.

Comparisons were made among six N₂-fixing tree species, *Gliricidia sepium*, *Leucaena pallida*, *L. taramba*, *L. involucrata*, *L. diversifolia*, and a hybrid of *L. diversifolia*. The non-fixing reference species were jackfruit, cashew, and bauhinia. Labelled ammonium sulphate fertilizer (10% a.e.) was applied as a spray in three split doses, at 3, 6, and 8 months after transplanting to give a total of 20 kg N/ha.

At 6 months after transplanting, *L. diversifolia* had the greatest height (2.1 m). To elucidate trends in fertilizer N uptake and total N assimilation, leaf samples were collected at 20, 25 and 30 weeks. The final harvest was at 12 months after transplanting. Dry weight determinations, and total N and ¹⁵N analyses, are pending.

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Biological N₂-fixation capacity of six leguminous tree species.

A field experiment was conducted from July 1995 to April 1996, to estimate the biological N₂-fixing capacity of six leguminous trees using the isotope-dilution technology. *Gliricidia sepium*, *Calliandra calothyrsis*, *Leucaena leucocephala*, *Erythrina subumbranse*, *Albizia falcata*, and *Acacia mangium* were the fixing species, and *Cassia siamea*, *C. spectabilis* and *Michalia champica* were the non-fixing reference species. Seedlings raised in 12.5 x 17.5 cm polythene bags were field-planted at one-month old. Each plot consisted of 36 plants (three fixing and six non-fixing) square-planted with a 1-m spacing; there were four replicate plots. Two weeks after field planting, P and K were applied at 21 kg/ha of each. A solution of ammonium sulphate enriched with 10.36 atom % ¹⁵N was sprayed onto the soil at a rate of 20 kg N/ha in three split applications at 3, 6, and 8 months after transplanting.

The highest total dry weight (4.58 kg/plant) was obtained with the non-fixing legume reference species *C. spectabilis*. Of the fixing species *C. calothyrses* recorded the highest dry weight (2.52 kg/plant, not significantly different from those of *A. falcataria* or *A. mangium*). *C. spectabilis* also had the highest total N content (70.7 g N/plant); *C. calothyrsis* accumulated 39.5 g N/plant, not significantly greater than the N contents of *L. leucocephala* or *G. sepium*. Estimates for N fixed varied with the reference species, but with each reference the differences between the amounts fixed by *C. calothyrses* (average 21.9 g N/plant) and those by *L. leucocephala* (average 18.5 g N/plant) were insignificant. The average estimates for N fixed (g N/plant) by the other species were 12.7 (*G. sepium*), 7.64 (*A. falcataria*), 7.38 (*E. subumbranse*), and 1.11 (*A. mangium*).

Sawaeng Ruaysoongnern

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Nitrogen fixation by leguminous tree species in the northeast of Thailand.

Eleven N₂-fixing tree species were planted in open-ended cement cylinders buried in a soil typical of northeast Thailand: *Azelia macrocarpa*, *Pterocarpus macrocarpus*, *Xylia xylocarpa*, *Sesbania*

grandiflora, *Peltophorum inerme*, *Pithecellobium dulce*, *Samanea saman*, *Leucaena leucocephala* var. *K-7*, *Cajanus cajan*, *Acacia auriculiformis*, and *Acacia mangium*. Four non-fixing reference trees were included in the study: *Azadirachta indica*, *Dipterocarpus alata*, *Cassia siamea*, and *Tamarindus indica*. Transplanted seedlings differed in age, but were similar in size. Fertilizer N, labelled with ^{15}N , was applied at 2 g N/m². Final harvest was at 9 months after transplanting.

The highest dry-matter production was obtained with the non-fixing reference species *C. siamea* (1.08 kg/plant); the other non-fixers produced only 0.042 to 0.098 kg/plant. The best of the fixing species were *A. mangium*, *A. auriculiformis*, and *C. cajan* (0.70, 0.63, and 0.38 kg/plant, respectively). Total N and ^{15}N analyses are pending.

Do Dinh Sam

Forest Research Institute of Vietnam

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Vietnam

Biological N₂ fixation in some tree species in Vietnam.

Six species of N₂-fixing trees were chosen for the experiment: *Albizia samal*, *Acacia auriculiformis*, *Acacia mangium*, *Indigofera teysmannii*, *Tephrosia candida*, *Calliandra calothyrsus*. And three species were included as non-fixing reference species: *Cassia siamea*, *Anisoptera cochinchinensis*, and *Hopea odorata*. Three-month-old seedlings were transplanted in the field (soil pH 5.2), and treated with fertilizer N in four split doses at 1, 2, 4, and 6 months after transplanting, for a total of 20 kg N/ha labelled with ^{15}N at 10% a.e. Phosphorus, K, and lime were applied at locally-recommended rates.

Trees were destructively harvested after 8 months. The highest dry weights were obtained with *I. teysmannii* (669 g/plant), *C. calothyrsus* (631 g/plant) and *T. candida* (475 g/plant); these species also had the highest accumulations of N (9.61 to 11.7 g/plant). The estimates of N fixed were similar using *A. cochinchinensis* and *H. odorata* as the reference crops, e.g. for *I. teysmannii* the respective values were 77 and 83% of N from fixation. Lower estimates resulted with *C. siamea* (e.g., 10% of N from fixation for *I. teysmannii*), and negative values were obtained for *A. samal*, *A. auriculiformis*, *T. candida*, and *C. calothyrsus*.

6. Regional Co-ordination Meeting of the Technical Co-operation Project on Plant Nutrition and Soil-Water Management (RLA/5/036). Irapuato, Gto., Mexico, 30 September - 4 October 1996.

Scientific Secretary: Felipe Zapata

As mentioned in the last issue, this project was initiated this year. One of the first activities was the organization of the first co-ordination meeting at the Centro de Investigaciones y Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), at Irapuato.

The objectives of the first Regional Co-ordination Meeting were as follows:

i) To assess the status of the use of nuclear techniques in studies on soil/plant/water relationships in each participating country through a review of results and achievements from IAEA Technical Co-operation projects implemented over the last ten years.

ii) To identify the main problems in soil fertility and water management affecting agricultural production in the region, and to prioritize them.

iii) To elaborate a detailed Programme of Activities for implementation during the biennium 1997-98, in accordance with the objectives set out for the project.

iv) To discuss mechanisms for strengthening regional co-operation and co-ordination in the planning and implementation of the project, as well as to promote free exchange of information.

Based on the conditions set for participation in the project, eight countries from the region were selected: Argentina, Brazil, Cuba, Chile, Guatemala, Mexico, Uruguay and Venezuela. The project co-ordinators who participated in the meeting were: N. Barbaro (Argentina), T. Muraoka (Brazil), O. Muñoz (Cuba), I. Pino (Chile), M.A. Alfaro (Guatemala), J.J. Peña-Cabiales (Mexico), J. Zamalvide (Uruguay)

and R. Delgado (Venezuela). Mr. J. Morales, Head of the ARCAL (Arreglos Cooperativos para la Promocion de la Ciencia y la Tecnologia Nucleares en America Latina) Unit, and Mr. F. Zapata, Technical Officer, both of the IAEA, also participated.

A full technical report was prepared, containing an assessment of the situation in the region, the detailed programme of activities for 1997-98, experimental plans for the regional network, and conclusions and recommendations. This report (in Spanish) is available from Mr. F. Zapata at the Soil and Water Management and Crop Nutrition Section.

7. Pre-project Workshop under the Inter-regional Technical Co-operation Project on Sustainable Utilization of Wasteland and Saline Groundwater for Plant Production, (INT/1997/001/PL) Faisalabad, Pakistan, 6-13 October 1996.

Responsible Officers: M.H. Naqvi (NIAB); C. Hera (FAO/IAEA); E. Garcia Agudo (RIPC)

The objectives of the pre-project workshop, organized at the Nuclear Institute for Agriculture and Biology (NIAB), were:

- to prepare detailed work plans for the project for each participating counterpart
- to exchange information and materials
- to demonstrate nuclear and other techniques to be used in the project
- to elaborate implementation strategies, standardized uniform techniques and plans, with time schedules and review procedures.

In attendance were representatives of seven countries: Egypt, Iran, Iraq, Morocco, Pakistan, Syria and Tunis. Each participant presented a country report and work-plan suggestions.

The workshop was organized by the IAEA Department of Technical Co-operation in conjunction with the Soil and Water Management & Crop Nutrition Section of the Joint FAO/IAEA Division, with the Isotope Hydrology Section of the Division of Physical and Chemical Sciences, and with the NIAB.

8. FAO/IAEA Annual Co-ordination Meeting of the Regional Technical Co-operation Project for West Asia on Water Balance and Fertigation for Crop Improvement (RAW/5/002). Amman, Jordan, 10-13 November 1996.

Responsible Officer: Pierre Moutonnet

The second Annual Co-ordination Meeting was implemented as scheduled, and will be fully reported in the next issue.

9. Workshop on Strengthening Analytical Laboratories in Latin America (RLA/5/036.001). Santiago, Chile, 11-15 November 1996.

Responsible Officer: I. Pino (CCHEN, Chile)

This workshop was implemented as scheduled, and will be reported in the next issue.

Planned

1. Third Research Co-ordination Meeting of the FAO/IAEA CRP on The Use of Nuclear and Related Techniques for Evaluating the Agronomic Effectiveness of Phosphate Fertilizers, in Particular Rock Phosphates (D1-50.03). Vienna, Austria, 17-21 March 1997.

Responsible Officer: F. Zapata

2. **Fourth and final Research Co-ordination Meeting of the FAO/IAEA CRP on The Use of Nuclear Techniques in the Management of Nitrogen Fixing Trees for Enhancing Soil Fertility and Soil Conservation (D1-40.05). Vienna, Austria, September 1997.**

Responsible Officer: (To be decided)

3. **Third Research Co-ordination Meeting of the FAO/IAEA CRP on The Use of Nuclear Techniques for Optimizing Fertilizer Applications under Irrigated Wheat to Increase the Efficient Use of Fertilizers and Consequently Reduce Environmental Pollution (D1-40.07). Vienna, Austria, 22-26 September 1997.**

Scientific Secretary: P. Moutonnet

4. **FAO/IAEA Regional Seminar for Latin America on Nuclear Techniques for Optimizing the Use of Nutrients and Water for Maximizing Plant Productivity and Environmental Preservation (D1-SR-203). Piracicaba, Sao Paulo, Brazil, 27-31 October 1997.**

Responsible Officer: F. Zapata

5. **FAO/IAEA Group Training under the Regional Technical Co-operation Project for West Asia on Water Balance and Fertigation for Crop Improvement (RAW/5/002). Nicosia, Cyprus, March 1997.**

Responsible Officer: P. Moutonnet

6. **FAO/IAEA Consultants Meeting on the Use of Nuclear Techniques in Rainfed Agriculture, 1997 (exact date to be decided).**

Scientific Secretary: P. Moutonnet

TECHNICAL CO-OPERATION PROJECTS

In this issue we provide information on two regional TC Projects, and list the TCPs that are planned for 1997-98.

1. **Regional Technical Co-operation Project for West Asia on Water Balance and Fertigation for Crop Improvement (RAW/5/002).**

Project Officer: Pierre Moutonnet

This project was initiated in 1995, for a two-year period, with the objective of improving efficiency of use of irrigation water and N-fertilizer in the arid and semi-arid conditions that exist in most countries of the Middle East. **Fertigation (fertilization applied through, mainly, drip irrigation)** is recognized as an effective way to save water and nutrients, and to increase crop production on marginal calcareous soils. It has been emphasized that fertigation is particularly important in the Middle East, since irrigated agriculture is expanding rapidly and modern irrigation technology is being broadly adopted. However, fertigation is practiced often without any technical background and/or control. Nuclear techniques, the soil-moisture neutron probe and ¹⁵N-labelled fertilizers, are powerful research tools to illustrate the substantial benefits of fertigation when properly applied.

Six countries are at present involved in this regional TCP: Iran (Nuclear Research Centre for Medicine and Agriculture, Karaj), Jordan (The University of Jordan, Amman; The Jordan University of Science and Technology, Irbid; The National Centre for Agriculture Research and Technology),

Lebanon (National Council for Scientific Research; American University of Beirut; Lebanese University of Beirut), Saudi Arabia (King Abdul Azziz City for Science and Technology), Syria (Syrian Atomic Energy Commission), United Arab Emirates (Ministry of Agriculture and Fisheries, Dubai). Furthermore, scientists from Turkey (Turkish Atomic Energy Authority) and Cyprus (Agricultural Research Institute, Nicosia) are associated, as consultants, with the programme. The 1996 Annual Co-ordination Meeting was held in Amman, 10-13 November, and will be reported in the next issue. The first Group Training in fertigation studies is planned for Cyprus in March 1997.

2. Regional Technical Co-operation Project for Latin America (ARCAL XXII) on Plant Nutrition, Soil and Water management (RLA/5/036)

Project Officer: Felipe Zapata

The first phase of this project (1996-1998) started implementation this year with the first Meeting of the Project Co-ordinators, which was held in Irapuato, Mexico from 30 September to 4 October 1996. The first phase is designed to gather the basic knowledge obtained so far using nuclear techniques by national institutes in the region under past IAEA Technical Co-operation and apply it to generate technologies which will be tried preferably with the end users to demonstrate its technical and economic feasibility. Nuclear techniques will be applied where appropriate to monitor nutrient supply and soil water status in a network of field trials.

Eight countries have fulfilled the requirements for participation in this TCP, i.e., Argentina (National Atomic Energy Commission), Brazil (Centre for Nuclear Applications in Agriculture), Cuba (Institute of Soils), Guatemala (General Directorate of Nuclear Energy), Mexico (Centre for Investigations and Advanced Studies), Uruguay (Faculty of Agriculture, University of the Republic) and Venezuela (National Foundation for Investigations in Agriculture and Livestock).

The report of the first meeting (in Spanish) contains: i) an assessment of the main problems and use of nuclear techniques in soil/plant research, ii) a detailed plan of activities for the biennium 1997-1998, iii) experimental plans for the network of field trials and iv) conclusions and recommendations.

Among the activities scheduled for 1997, there are two regional training workshops:

- i) FAO/IAEA Regional Training Workshop on the Use of Nuclear Techniques to Increase the Efficiency of Chemical Fertilizer N, Biofertilizers and Green Manures in Latin America (C7-RLA/5/036-002). Havana, Cuba, 21-25 April 1997.
- ii) FAO/IAEA Regional Training Workshop on the Use of Neutron Probes and Water and Nutrient Balance (C7-RLA/5/036-003). Piracicaba, Sao Paulo, Brazil, 20-24 October 1997.

3. Technical Co-operation Projects Operational in 1997-98.

	Technical Officer
Sustainable Utilization of Saline Groundwater and Wastelands for Plant Production (INT1997001PL).	Christian Hera
Increasing Food Security in Sub-Saharan Africa (RAF5036).	M. Peter Salema
Water Balance and Fertigation for Crop Improvement (RAW5002).	Pierre Moutonnet
Plant Nutrition and Soil-Water Management (ARCAL XXII) (RLA5036).	Felipe Zapata
Soil Fertility Studies in the Pampa Region (ARG5008).	Felipe Zapata
Biofertilizers for Increased Legume Production (BGD5017).	Felipe Zapata
Rational Use of Nutrients in Sustainable Agricultural Systems (CHI5020).	Felipe Zapata
Improving Plant Nutrition Systems to Increase Crop Prod'n (CUB5015).	Felipe Zapata
Nuclear Techniques in Desert Farming (EGY5024).	Edward Weck & Pierre Moutonnet
Plant Nutrient Management in Traditional Crops (GUA5011).	Felipe Zapata
Effective Use of Fertilizers in Rice Production (INS5024).	Felipe Zapata
Production of Biofertilizers to Improve Legume and Grain Yields (IRQ1997001PL).	Pierre Moutonnet

Field Performance of Selected Mutants of Sorghum and Rice (ML15014).	Beant Ahloowalia & Felipe Zapata
Nuclear Techniques to Improve Production of Wheat and Legumes (MON1997003PL).	Miroslav Maluszynski & Pierre Moutonnet
Identifying N-fixing Trees in Low Phosphate Soils (MOR5022).	Felipe Zapata
Fertilizer and Water Management (NAM5004).	Pierre Moutonnet
Improving Soil Management Practices to Increase Crop Yield (NIC5005).	Felipe Zapata
Use of Nuclear Techniques to Improve Crop Productivity in Arid Areas (PAK1997013PL).	Pierre Moutonnet
Nitrogen Fixation in Pasture Legumes (PHI1997006PL).	Christian Hera
Increasing Nitrogen Fixation Potential of Niébé (SEN1997002PL).	Pierre Moutonnet
Use of Isotopes to Increase Rice Productivity (SIL5004).	Felipe Zapata
Isotope Techniques for Efficient Use of Local Rock Phosphate (SRL1997006PL).	Felipe Zapata
Management of Nutrients, Water and Organic Matter (SUD5021).	Pierre Moutonnet
Improved Sugarcane Production Using Nuclear Techniques (THA5042).	Christian Hera
Use of Nuclear Techniques to Improve Water and Soil Management Practices (TUN1997002PL).	Pierre Moutonnet
Biofertilizers for Increased Crop Production in the Small Scale Farming Sector (ZIM1997004PL).	M. Peter Salema

FROM OUR READERS

ESNA Meeting

At the XXVIth Annual Meeting of ESNA, held at Busteni, Romania, 12 - 16 September 1996, Dr. Martin Gerzabeck (Austrian Research Centre at Seibersdorf) served as Chairman of the Working Groups on Radionuclide Soil-to-Plant Transfer. Dr. Gerzabeck has kindly provided the following report.

The Working Groups encompassed four sessions in which 35 papers were presented by scientists from 13 countries. In the session *Nitrogen in the Soil-plant System*, a series of papers given by Turkish colleagues highlighted aspects of fertilizer-N use in various regions of their country, on potato, tomato, wheat and sugar beet. In many cases N-fertilizers are being applied in excessive amounts, leading to luxury consumption by plants and to nitrate pollution of the groundwater. For example, Halitligil reported an economical optimum of 600 kg N/ha for potato in sandy soils, which results in detectable concentrations of ¹⁵N-labelled nitrate at a soil depth of 2 m within a short time period. And Brohi described problems related to excessive N supply to sugar beet: applications of up to 450 kg N/ha are common, causing problems in sugar processing. According to Durak, ammonium fixation may also be a problem in some Turkish soils.

Two papers (Brohi/Turkey and Nankova/Bulgaria) dealt with the effects of organic manures on N uptake by plants. Four papers on the N-nutrition of tomato showed that vegetative organs are the main sinks for N, and that there is opportunity for improvement in N-utilization efficiency. Gheorge (Romania) and Milev (Bulgaria) presented extensive studies on N₂ fixation by various legumes and their ameliorative effect on subsequent wheat and rye.

In the session *Soil Science and Plant Nutrition*, Licina (Yugoslavia) highlighted the importance of the micronutrient selenium for human nutrition. Although problems arise from Se-deficient soils, fertilization strategies should be evaluated carefully because of the narrow range between Se-deficiency and toxicity for humans. Other presentations in this session covered the leaching of cations and anions from irrigated and unirrigated arable land (Saltali/Turkey), a comparison of methods for the determination of lime requirements for Turkish soils (Sûrûcû), and the effects of minimum tillage on winter wheat in Bulgaria (Yankov). Iliev (Bulgaria) described the technical aspects of a soil sampler mounted on a light vehicle, and Hoza (Romania) documented the influence of the rooting substrate on the nursery production of tomato transplants.

In the *Microbiology* session, Middleboe (Denmark) proposed a standardized method for the long-term observation of soil microbial activity, using doubly-labelled phosphate. This method is advantageous in providing values integrated over a period of time (e.g. three months), avoiding the high numbers of samples that are otherwise necessary to precisely determine short-term averages. Tardioli (Italy) showed significant adverse effects of olive-mill wastewater on soil fungal community composition. Turcu (Romania) presented data on the degradation of the new herbicide imazethapyr in comparison to ^{14}C -labelled atrazin: the former is three times more mobile under comparable conditions but is more quickly degraded and, therefore, may be regarded as less harmful ecologically.

In two papers during the session on *Radioecology* chaired by Dr. N. Mitchell, the influence of K on the soil-to-plant transfer of radiocaesium was discussed. Klemt (Germany) used Langmuir's equation, taking into account concentration effects of frayed-edge sites, to describe the behaviour of ^{137}Cs in seminatural environments. Mocanu (Romania) described a laboratory method using plantlets to estimate K fertilizer effectiveness in decreasing the uptake of Cs by plants. This method covers soil-chemical as well as plant-physiological effects of the K treatment. Jones (UK) presented a basic study on ^{137}Cs -cycling in upland plants. He quantified the translocation of radiocaesium to newly-formed plant organs, losses from the plants, and re-uptake from the peat soil. It was also shown that within-plant translocation increases with improved nutrient status.

Kravets (Ukraine) proposed a model from which limit values can be deduced for radionuclide contamination of different soil types. In a second paper, he showed that high-density sowing of agricultural plants decreased the effective half-lives of ^{137}Cs - and ^{90}Sr in topsoil. In the Chernobyl exclusion zone, ^{90}Sr concentrations are increasing in pasture plants, due to decomposition of fuel particles - clearly demonstrated by Goncharova (Belarus). Russkikh (Belarus) evaluated an alternative farming system for highly contaminated areas around the Chernobyl power plant, showing that soyabean seed production is possible with acceptable seed quality. Skarlou (Greece) showed that soil-to-citrus transfer of Cs is highly affected by soil properties, and problems from dry deposition of ^{134}Cs on olive trees are significant only during the first year after contamination. Pietsak-Flis (Poland) discussed the contribution of less-well investigated radionuclides when calculating ingestion dose from plant foodstuffs; ^{226}Ra and ^{228}Th especially should be taken into account. Gerzabek (Austria) and Wadey (UK) discussed annual fluxes of ^{137}Cs , ^{60}Co and ^{226}Ra from contaminated topsoil to groundwater, as determined using lysimeters. Their preliminary results indicate that ^{60}Co is the most mobile. In a second paper they described the root uptake of radiocaesium by winter wheat and perennial ryegrass from soil horizons near the water table, and their introduction of a plant compartment into previously presented models for solute transport in soil.

The radioecology session ended with a lively discussion on modelling, after a short introduction by Mitchell (UK). The inductive and deductive methods of model development were compared, taking complexity, including area size, into consideration as is necessary for certain objectives. Limitations inherent in making site-specific predictions were discussed.

The proceedings will be published in December, and the next ESNA-meeting will be held in Ghent (Belgium), 29 August - 2 September 1997.

Letters

In the last issue, we announced the sad news of the death of our valued colleague and dear friend Dr. Saliya Kumarasinghe on 17 June 1996. Letters from bereaved colleagues continue to arrive at the Agency:

"I was saddened to learn of Saliya's death. I express my sincere condolences to all at the Joint Division particularly in the Soils Section, and to the Kumarasinghe family" (Mamadou Gyueye, Sénégal).

"We recently learned the shocking news of Saliya Kumarasinghe's sudden demise, from the Soils Newsletter. We are deeply sorry and cannot accept that he is no longer with us. No words can express our sadness at this terrible occurrence. Our condolences to his friends in the Section and especially to his family" (Le Dac Lieu, Vietnam).

"The news of the sad demise of Saliya conveyed through the IAEA Soils Newsletter of July 1996 distressed us all. He was such a nice guy. I convey my deepest condolence to all his friends and colleagues at the Agency" (Raziuddin Ansari, Pakistan).

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