

# *Mucuna News*

## *Developing Multiple Uses for a Proven Green Manure/Cover Crop*

### *Update on the Progress of the Project “Increasing Mucuna’s Potential as a Food and Feed Crop”*

**CENTER FOR COVER CROPS INFORMATION AND SEED EXCHANGE  
IN AFRICA (CIEPCA)**

*Fourth issue, November 2001*

### *Progress of the Project*

By R.J. Carsky and M. Eilittä

At an approximate midpoint of the project, we have asked the investigators affiliated with the Project to give a brief summary – just a paragraph or two – on their progress. The responses made it clear that most researchers are at the point of peak field and laboratory work. *Mucuna* researchers are busy with their diverse activities all around the globe. Processing methods for *Mucuna* foods are being developed (in Guatemala City, Guatemala; in Gainesville, Florida, USA; in Kitale, Kenya; in Cotonou, Benin; in Ibadan, Nigeria; and in Coimbatore, Tamil Nadu, India), *Mucuna* feeds have been grown, some of them processed, and fed to goats, sheep, and dairy cows (in Mt. Pleasant, Zimbabwe; in Mombasa, Kenya; and near Cotonou, Benin) and to chickens (in Umudike, Nigeria; in Burlington, Vermont, USA and at Zamorano, Honduras). A genetic map of *Mucuna* is being developed in Auburn (see abstract, below), where the researcher's additional work includes a number of crosses made, including one between climbing and erect varieties. Some of these *Mucuna* types for which genetic information is available will soon be shipped a few hundred kilometers south (to Gainesville, Florida) where the exact amount of some toxic alkaloids known to be present in *Mucuna* (5-Me-DMT and serotonin (5-HT) and tryptamine) will be determined. Farther north, in Illinois, the past year's *Mucuna* seed samples from the genotype by environment trial are being analyzed, while work early next year will focus on determining whether any mutagenic compounds may be present in *Mucuna*. Eight *Mucuna* types are being grown under field conditions in ten different locations (in 6 countries) in latitudes ranging from 18° S to 38° N to evaluate whether environment, genotype or both influence their final L-dopa content. A survey of food utilization of *Mucuna* in

several African countries is initiating its pilot phase in Eastern Nigeria, while farmers in Central Benin are being surveyed on their utilization of *Mucuna* as a cattle feed. Finally, across Africa and in Gainesville, Florida and Judson, Illinois, researchers are working together by reviewing literature, making trial runs, and giving advice to make L-dopa analysis a reality in diverse institutions in Africa. Indeed, this continues to be our priority work now as we have not yet been able to locate an institution to conduct the L-dopa analyses for most of the projects in West Africa.

That all these *Mucuna* researchers are busy with their projects is good news. In addition, we have another piece of good news – one which we hope will further encourage the investigators: another *Mucuna* Workshop is being considered at the conclusion of the project. Many of the details of the workshop have not yet been worked out but we have received positive responses for informal inquiries to funding. Please see the separate announcement in this *Mucuna* News and look for further information in subsequent issues.

We are grateful for the updates submitted by the investigators and, as always, to the MOIST-CIIFAD of Cornell University for posting our newsletter on the Internet.

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## Project Update

### Midpoint Updates

In the following, we will highlight the progress of our diverse projects. In the next issue, additional projects will be featured. For further information on these projects, please send an email to the editor ([meilitta@aviso.ci](mailto:meilitta@aviso.ci)) who will help you make the contact with the particular researcher.

### Food-related Projects

**Solvent Extraction Studies for Removing L-dopa from *Mucuna* Bean:** A. Teixeira and his collaborators at the University of Florida have just completed the first phase of the project. The purpose of this phase was to study the effect of particle size on rate of L-dopa extraction from chopped *Mucuna* beans using tap water at room temperature as the solvent. These first results indicate that after 2 hours of extraction time, residual L-dopa content in whole shelled beans is reduced by only 50% and it plateaus at this level for the 24-hour duration. In contrast, the residual content in 1mm (smallest) particle size samples is reduced by nearly 80% within 2 hours and continues decreasing to reach a final reduction of 95% after 24 hours. With respect to actual levels, L-dopa is reduced from an initial content of 4.23% dry weight to 0.25% after 24 hours in tap water. In the next phase, A. Teixeira and his collaborators will work only with the 1mm particle size, studying longer times with water at room temperature, extraction rates at higher temperatures and at different pH, and with continuous flow adsorption columns.

**Evaluation of processing methods of *Mucuna* for food in Kenya:** E. Wanjekeche from the Kenya Agricultural Research Institute-Kitale is leading an effort to assess the impact of locally available alkaline and acid materials (i.e., maize cob and bean stover ash and "Magadi soda"), ordinary and pressure cooking and germination and fermentation on nutritional and anti-nutritional factors of mature and immature *Mucuna* beans. E. Wanjekeche had completed one of the three replications in early October when M. Eilittä met with her and observed some of her processing methods at the University of Nairobi, where she conducts her laboratory analyses. The results from the first replication affirm that pre-soaking in water greatly reduces cooking time to normal softness (6.0, 4.5, and 2.5 hours for samples with no soaking, 12-hr soaking, and 24-hr soaking, respectively). After a 24-hr soaking, cooking in alkaline and acid solutions further reduced cooking time (1.4, 2.0, and 1.0 hrs with 0.5% Magadi soda, 0.25% citric acid, and maize cob ash solution in 1:3 dilution with water, respectively). Furthermore, cooking in alkaline solutions loosened the seed coat and

resulted in a very dark brown cooking liquid; in plain water, the color was only slightly brown, while in acid media, the color was clear, indicating different reactions with perhaps significant impacts on L-dopa content (which will be analyzed at the laboratory of C. Gachuiiri at the Department of Animal Production). There was a large seed-to-seed variability in water absorption but the alkaline solutions also improved uniformity of cooking. Content of important nutritional factors seemingly varies by treatment.

### **Protein Quality and Residual L-dopa and Trypsin Inhibitor Activity in Processed**

***Mucuna* Beans:** R. Bressani's project focuses on the impact of different processing methods (i.e., atmospheric and pressure cooking, roasting, germination and fermentation), on nutritional and anti-nutritional factors in *Mucuna*. Processing studies are currently being done. Roasting treatment has caused some problems in determining L-dopa content; N. Szabo has assisted in this regard. The products will be tested biologically through rat feeding studies in October. Two student technicians and two food science students are working on the project.

### **Survey of ways to utilize *Mucuna* as a food:**

J. Onweluzo from the University of Nsukka and M. Eilittä are coordinating this survey that seeks general information on ways to cultivate and utilize *Mucuna*, and specifics on its food utilization, its perceived benefits and problems, as well as on market-related issues. The pilot study is being conducted in one local government area in Eastern Nigeria, after which the survey form will be revised if necessary, and the survey implemented in a wider area in Nigeria and in Ghana, Mozambique, and Malawi.

### Germplasm Projects

***Mucuna* Germplasm Collection and Screening for Anti-Nutrients:** K. Janardhanan's team has gathered five accessions of *Mucuna* beans from the Western Ghats in Tamil Nadu and Kerala of India. At the moment, they are analysing anti-nutrients present in the raw seeds. The next phase will include formulation of processing methods to minimize or eliminate the detected antinutrients.

**Assessment of genetic diversity and initial genetic map for *Mucuna* sp.:** The purpose of L. Capo-chichi's Ph.D. research is to identify potential molecular markers linked to important morphological and agronomic traits that would be useful for developing and improving the species. The focus of his work has been an intra-specific F<sub>2</sub> population, generated after self-pollinating a cross between two *Mucuna* types, for which he has been locating a large number of AFLP

markers (see also a separate article below on this activity).

**Genotype by Environment trial:** See separate article below.

#### *Mucuna* as Feed for Ruminants

**The Effect of L-dopa in Forage on the Performance of Lactating Cows:** The work by R. Muinga and her collaborators in Mombasa, Kenya, involves assessing the long term effect (6 months) of feeding velvet bean forage on dry matter degradation in the rumen, feed intake, milk production and the relationship between animal performance and L-dopa levels in the blood. To ensure continuous supply of fresh fodder, *Mucuna* was planted at two-week intervals in April-June 2001. The 12 experimental animals were put under zero grazing system on 21 June 2001. To date, there have been no apparent differences between cows fed *Mucuna* and *Gliricidia* in animal performance, including milk yield. However, a July outbreak of Foot and Mouth Disease affected some of the animals adversely and cows per treatment were reduced to three for the *Gliricidia* and four for the *Mucuna* treatment. As with the project of E. Wanjekeche and collaborators, the L-dopa analyses will be conducted at the University of Nairobi under the leadership of C. Gachuiiri.

**Nutritive Evaluation of Velvetbeans as Feed Ingredients for Small Ruminants:** The general objective of the project of M. Titterton, N. Ngongoni, and B. Maasdorp (University of Zimbabwe) is to conduct a nutritional characterization of *Mucuna* seed, evaluate the impact of ensiling on the anti-nutritional factors of *Mucuna*, and to assess the response (i.e., milk yield, reproductive performance, content of L-dopa in blood and milk) of goat does and sheep. *Mucuna* seed has been ensiled with maize and, when ready, will be analysed for nutritional and fermentation quality as well as for L-dopa content. The does have been flushed and are ready for synchronization of oestrus so that they kid in January. L-dopa analysis is being worked out at the university, with the help of R. Myhrman and N. Szabo. An M.Sc. student is working on the project.

#### *Mucuna* as a Feed for Non-Ruminants

**Studies on the effect of heated, water extracted and extruded *Mucuna* and of methionine and lysine supplementation in diets for broilers:** The initiation of S.N. Ukachukwu's work at the Michael Okpara University of Agriculture in Eastern Nigerian town of Umudike was delayed due to difficult communications. But, in the past months, he has

made quick progress by purchasing the necessary supplies and processing the seed samples for analysis. The experimental pens have been readied and the animal study is about to start.

**The Effects of Heated Velvet Beans (*Mucuna pruriens*) on Blood Chemistry and Organ Size in Growing Chickens:** The work of L. Carew (University of Vermont) and collaborators focuses on the impact of raw and processed (heating at 130°C for 30 min.) *Mucuna* feed on organ size, blood chemistry, and histology of growing chickens. He reported the results on organ size at the Poultry Science meetings in July. Intake of raw *Mucuna* beans has marked effects on the growth of certain organs (i.e., increased size of small and large intestines and ceca, pancreas, gizzard and proventriculus). In some cases this is modified by heating the beans, presumably because raw *Mucuna* beans contain growth factors or growth properties that are altered by heat treatment. Where heating of the beans was only partially effective in altering organ growth, as with lengths of the small and large intestines, and gizzard and proventriculus, the residual effect may be due to the increased presence of fiber. In the next phase, the investigators will focus on the analysis of blood samples and on studying the histological changes.

#### Health-related Projects

**Assessment of the levels of alkaloids in different parts of *Mucuna* plant:** N. Szabo's study is to be conducted in late October-November with accessions that L. Capo-chichi has genetically mapped using molecular methods. Genetically diverse materials are used in the assessment of alkaloids, to attempt to establish the range in the quantities of alkaloids present.

**Mutagenic characterization of *Mucuna*:** This work will initiate in January 2002.

#### G by E Trial Update

While the seeds from last year's trials are being analyzed at Judson College, *Mucuna* seems to have flourished in this year's trials. We have photos available from a number of sites; to view them, please contact the editor (meilitta@aviso.ci). Unfortunately, "Deeringiana" accession seemingly had a problem with germination, and in many locations, one of the replications was lost for this type. In two locations – in Zimbabwe and in one location in Columbia – *Mucuna* has not yet been planted. To accommodate these different growing seasons and to leave sufficient time for data analysis and writing, we have asked for a no-cost

extension for this project until December 31, 2002.

Here is a brief update on the various locations:

**Alabama, USA:** In September, all accessions were growing well, but none of them had flowered. Our collaborator and the trial coordinator L. Capo-chichi has a number of other *Mucuna* types also growing in Auburn as a part of his Ph.D. project.

**Columbia:** Problems with seed import and a three-month drought delayed the planting in Columbia. However, our CIAT collaborator M. Peters has already planted in one, acid, site and plans to plant the other – alkaline - site in March 2002.

**Benin:** A. Etèka from CIEPCA reports that the trial, which was planted in June, looks good. The “Deeringiana” type had to be replanted. In September, both “Rajada” and “Ghana” types had many pods; the other accessions had not yet flowered.

**Florida, USA:** Planting was done in April but in September, there was no flowering yet. Our collaborator R. Gilberts tells that one “Deeringiana” replication did not emerge and one “IRZ” replication died of an unknown disease. The growth of the other plants is lush, despite Japanese Beetle attacks.

**California, USA:** Accessions there were planted in a greenhouse in March, then transplanted to field in June. All accessions were flowering and many had pods in September and – in the words of our collaborator S. Temple the *Mucuna* there “is fantastic!!”

**Honduras:** The trial site is about three hours from the capital, at Escuela Nacional de Agricultura. They were planted in July, and have been growing well in spite of some pest problems in the beginning of the growing season. In September, one accession was flowering while the others were about to flower.

**Chiapas, Mexico:** *Mucuna* is growing well in Chiapas, too, as evidenced by some nice photos sent by R. Quiroga, our collaborator there. Only one “Deeringiana” replication has germinated, however.

**Yucatan, Mexico:** Our collaborator J. Castillo has sent some pictures of very lush *Mucuna* growing into nearby trees. In addition to the trial accessions, he is growing three other *Mucuna* types: a dwarf type, “African” type, and “Pinto” type.

### *Fermentation Study in Benin: The Latest Mucuna Project Activity*

This summer saw the initiation of the latest – and last - of the *Mucuna* Project activities:

I. Egounlety at the Faculté des Sciences Agronomiques de l'Université Nationale du Bénin is conducting a study to determine whether fermentation may be a promising food processing method for *Mucuna*. Fermentation is a widely practised traditional processing method and brings about a number of beneficial biochemical, nutritional and organoleptic changes including the breakdown of certain constituents, the reduction of the anti-nutritional factors in grain legumes and the synthesis of  $\beta$ -vitamins.

In I. Egounlety's study, *Mucuna* beans will be pretreated and subjected to bacterial or fungal fermentation. A number of fermented *Mucuna* foods will be produced. A condiment will be prepared by bacterial fermentation with *Bacillus* that is done in calabash; this method resembles the processing of locust bean. *Mucuna tempe* will be produced with *tempe* starter containing *Rhizopus oligosporus*. *Mucuna* seed is also co-fermented with maize, which provides the bacteria and yeasts necessary for the fermentation. The pH will be closely monitored and soluble and non-protein nitrogen and level of residual L-dopa will be determined.

### *Mucuna Workshop Considered*

The current *Mucuna* Project initiated with a workshop “Food and Feed From *Mucuna*: Current Uses and the Way Forward” which was organized in April 2000 in Tegucigalpa, Honduras, by CIDICCO, CIEPCA, and Judson College. The workshop outlined the state of knowledge on the subject and proposed issues that should be the foci of future research efforts. We are now considering a second workshop to review the results of the project's activities. In this workshop, development implications of the research results would be given a greater emphasis and therefore many development organizations working on related themes would be invited. For further information please see the next issue of *Mucuna News* or contact M. Eilittä.

### *Mucuna Project to Provide Input into a Kit for GMCCs in Mexico*

*Mucuna* Project participants are invited to collaborate on a chapter on food and feed utilization of green manure/cover crops (GMCCs) in a book to be produced by RED/gac. This chapter will appear in a manual (i.e., kit) describing how to integrate green manure/cover crops in farming systems of (mainly southern)

Mexico. This is a good possibility to address field-level needs and concerns with the knowledge that we have generated during the April 2000 workshop and the *Mucuna* Project.

As described in *Mucuna* News no. 1, RED/gac (Network/Cover Agriculture Group) is a network of non-governmental organizations and academic institutions working on green manure/cover crops in Mexico. The network dates back to 1992, when it first started functioning as a network of projects that were funded by the Rockefeller Foundation, and more independently since 1997. One of the issues that the group is currently working on is the development of a kit on green manure/cover crop (GMCC) systems for Mexico. This kit, produced in participative workshops together with farmers and project staff, will give detailed information on the systems used, including farmer management and the advantages and disadvantages of GMCCs.

The kit will also include chapters that focus on particular themes associated with GMCCs. One such theme is food and feed utilization. In the participatory workshops, this issue is discussed with the farmers from a number of perspectives, including the ways that they utilize these crops as food and feed and their benefits and limitations. Farmers' concerns and questions regarding these crops will be recorded and those issues are addressed in the particular chapter of the kit.

*Mucuna* is the GMCC that has been most promoted and researched in Mexico and whose use as food and feed presents most problems. Without undermining the other GMCCs, the chapter will therefore give detailed attention to *Mucuna*. In November, 2001, a participatory workshop in Oaxaca, Mexico, will result in a listing of farmers' issues, concerns and questions with *Mucuna* and the other GMCCs. These issues will form the guiding light for the chapter that will be developed by February 2002. For further information, please contact M. Eilittä ([meilitta@aviso.ci](mailto:meilitta@aviso.ci)).

## Other News

### Genetic map of *Mucuna*

One of the exciting ongoing studies on *Mucuna* is that by L. Capo-chichi at Auburn University on taxonomy. His Ph.D. studies are progressing well and he presented some of the work that he has conducted at this year's annual meeting of ASA-CSSA-SSSA in Charlotte, North Carolina. An expanded abstract from the meeting is presented below. For further information contact L. Capo-chichi ([cludovic@acesag.auburn.edu](mailto:cludovic@acesag.auburn.edu)), D. Weaver ([dweaver@acesag.auburn.edu](mailto:dweaver@acesag.auburn.edu)), or C. Morton ([MORTOCY@auburn.edu](mailto:MORTOCY@auburn.edu)).

Velvetbean (*Mucuna* sp., n=11), a self-pollinated species, is an important legume used in tropical agricultural systems in rotation with other crops for nematode management and/or soil improvement. A genetic map of velvetbean was constructed in order to identify potential molecular markers linked to important morphological and agronomic traits that would be particularly useful for developing and improving the species. Traits such as maturity, seed coat color, pod color, and pod pubescence were among the main parameters used in a process of genetic diversity estimation. Traits were measured in experiments during a two-year season. Two velvetbean accessions, PI364362 and Edgar Farm White, a land race from Alabama, were used to make the intraspecific F<sub>1</sub> hybrid. The AFLP (amplified fragment polymorphism length) detected an average of five polymorphic fragments per primer combination between the two parents. As expected for dominant markers, a sum of all AFLP bands from both parents was generally observed to be present in AFLP profiles of F<sub>1</sub> progeny, indicating full penetrance and dominant nature of AFLP markers. The F<sub>1</sub> hybrid should have one set of chromosomes from PI364362 and the other from Edgar Farm White. A F<sub>2</sub> population was generated from self-pollinating a single F<sub>1</sub> plant. Most of the AFLP markers were inherited and segregated in Mendelian ratios. Of the 163 polymorphic markers generated from 25 AFLP primer combinations assayed, 137 (77%) segregated in a 3:1 ratio, the remaining 26 (23%) were both segregating and scorable. However, the 26 scorable types displayed different segregation patterns. A genetic linkage map was constructed using 82 F<sub>2</sub> plants and 137 markers segregating in a 3:1 ratio. The paternal parent map (PI364362) consisted of 71 markers partitioned within 10 linkage groups (387.0 cM) and the maternal parent map (Edgar Farm White) had 66 markers assembled into 6 linkage groups (1085.7 cM). The combination of all 137 markers were partitioned within 13 linkage groups (1270.7 cM). The PCR-based molecular markers allowed the construction of genetic maps, thus ensuring a good coverage of the velvetbean genome for further QTL detection and mapping studies. AFLP is an efficient marker system for mapping plant species such as velvetbean. The construction of this linkage map will in the future facilitate the mapping of genes controlling agronomically important traits and cultivar development.





Photo 1. Some *Mucuna* accessions grown by L. Capo-chichi. Photo by L. St-Laurent.

### Selected Bibliography of *Mucuna*

#### Introduction

In this issue, we present articles on *Mucuna*'s feed uses that are either based on animal studies or have a distinctive animal-focus. Please note that many articles presented in the previous issues (which focused on nutritional characterization and alkaloids and L-dopa in *Mucuna* seeds) also include relevant information. As earlier, we will list the most recent articles first.

Unlike with some other *Mucuna*-related articles, there is a wealth of information on the subject due to the extensive use of *Mucuna* as a feed in the southern United States and various tropical countries between 1890s and 1950s. Below we present only a few of those reports and articles; if you are interested in the subject, many more are available from M. Eilittä. All forage textbooks produced in the United States in the first half of the 20<sup>th</sup> century also contain information on *Mucuna*. Of the more recent *Mucuna* feed studies, many student projects on the subject have been conducted at Zamorano in Honduras and at the Universidad Autonoma de Yucatan in Mexico (some of the work was featured in *Mucuna* News no. 2). Most of these theses are in Spanish; for details, contact M. Eilittä. In the following, we mainly highlight the English sources, and present a handful of the hundreds

of articles and reports published in the United States in the early 20<sup>th</sup> century.

As always, if you are unable to locate any of these materials please contact M. Eilittä (meilitta@aviso.ci). Additionally, please inform her of any significant materials not listed here.

#### Part III. *Mucuna* as a Feed

Burgos, I. Matamoros, and E. Toro. Evaluation of velvetbean (*Mucuna pruriens*) meal and Enterolobium ciclocarpum fruit meal as replacements for soybean meal in diets for dual-purpose cows. Forthcoming. Food and Feed From *Mucuna*: Current Uses and the Way Forward. Proceedings of a workshop held in Tegucigalpa, Honduras, April 26-29, 2000. CIDICCO, Honduras. (Please watch future issues of this bulletin for further information.)

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