



Australian Government

**Australian Centre for
International Agricultural Research**

Final report

project

Improving the quality of pearl millet residues for livestock

project number

CIM/1999/062

date published

June 2011

prepared by

Charles Thomas Hash Jr, International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)

*co-authors/
contributors/
collaborators*

Michael Blümmel and Sunita Choudhary, International Livestock Research Institute (ILRI)

T. Nepolean, Francis R. Bidinger and B. Ramana Kumari, ICRISAT

I.S. Khairwal, All-India Coordinated Pearl Millet Improvement Project, Rajasthan Agricultural University

C.J. Dangaria, Gujarat Agricultural University, Millet Research Station

H.P. Yadav, C.C.S. Haryana Agricultural University

Srikant Sharma, Rajasthan Agricultural University, Agricultural Research Station Durgapura

approved by

Dr Paul Fox, Crop Improvement and Management Research Program Manager, ACIAR

final report number

FR2011-14

ISBN

978 1 321738 85 2

published by

ACIAR
GPO Box 1571
Canberra ACT 2601
Australia

This publication is published by ACIAR ABN 34 864 955 427. Care is taken to ensure the accuracy of the information contained in this publication. However ACIAR cannot accept responsibility for the accuracy or completeness of the information or opinions contained in the publication. You should make your own enquiries before making decisions concerning your interests.

© Australian Centre for International Agricultural Research (ACIAR) 2011- This work is copyright. Apart from any use as permitted under the *Copyright Act 1968*, no part may be reproduced by any process without prior written permission from ACIAR, GPO Box 1571, Canberra ACT 2601, Australia, aciar@aciar.gov.au.

Contents

1	Acknowledgments	4
2	Executive summary	5
3	Background	6
4	Objectives	9
5	Methodology	10
6	Achievements against activities and outputs/milestones	15
7	Key results and discussion	22
8	Impacts	31
8.1	Scientific impacts – now and in 5 years	31
8.2	Capacity impacts – now and in 5 years	33
8.3	Community impacts – now and in 5 years	34
8.4	Communication and dissemination activities	397
9	Conclusions and recommendations	408
9.1	Conclusions	408
9.2	Recommendations	419
10	References	41
10.1	References cited in report	41
10.2	List of publications produced by project	43
11	Appendixes	46
11.1	Appendix 1:	46
11.2	Appendix 2:	60
11.3	Appendix 3:	73

1 Acknowledgments

This report is dedicated to the memory of Dr Fran Bidinger, my mentor, colleague, and friend, who contributed substantially to the planning and implementation of this project until his untimely and completely unanticipated death in early April 2008. This project, and the entire ICRISAT pearl millet improvement team globally, suffered great loss with his passing. Although it has taken substantially more time to complete the analyses and interpretation of the NIRS-based recurrent selection studies in this project, which targeted improvement of dual-purpose open-pollinated varieties and topcross pollinators, than would have been required had Fran still been with us, we believe that the results are promising – provided that the base population has sufficient genetic variability for the traits of interest, and the progenies can be appropriately assessed in a reasonably uniform and representative environment, we can in fact improve both stover yield and its nutritive value without significantly reducing grain yield and/or disease resistance, and without changing crop duration to any agronomically important degree. Fran's skills as an agronomist – in getting good crop establishment and early seedling growth – combined with his keen observations of field variability, permitted us to produce usable data sets in some “pretty marginal” on-station pearl millet production environments that were not so different from those found in rainfed farmers' fields in central and western Rajasthan – a major target environment for this project. Fran's skills in debugging data sets and completing statistically rigorous analyses of these have been carried forward as he trained me and his support staff very well. However, his dedication to publication of results – in appropriate venues, be they the *International Sorghum & Millets Newsletter*, *Annals of Arid Zone*, the *Indian Journal of Genetics & Plant Breeding*, *Euphytica*, *Field Crops Research*, or *Crop Science* – is sorely missed. Last and most important, we miss him as a friend and colleague, with a broad smile on face, always eager to discuss science (or good food).

This project report is also dedicated to Dr. Bruce Stone, leader of the exploratory project on which this project was based. He dragged me kicking and screaming – “We're not ready, we're not ready ...” through proposal development and then guided us during the first three years of this project, when it became clear that although we really had not quite been ready to take this leap towards application, we really did have tantalizing evidence that in this dual-purpose “desert maize”, farmers (and plant breeders attempting to meet their varietal needs) really can “have their cake and eat it too”. With careful evaluation, it is possible to identify well-adapted hybrids, full-sibs, and composites having truly dual-purpose grain+fodder characteristics with reasonably high grain and stover yields combined with superior stover nutritional value. The anticipated role of foliar disease resistance in improving all three of these varietal characters, was borne out by the discovery of a previously undetected major dominant gene for foliar disease resistance that appears to co-segregate with the favourable allele for an introgressed QTL of one validated estimator of stover nutritive value (the volume of gas produced per unit ground dry matter in the first 24 hr of *in vitro* incubation with buffered rumen fluid under controlled laboratory conditions). Due to inadequacy of the molecular marker set available for pearl millet at the start of this project, it took longer than it should have for us to produce the finished contig line set, and the near-isogenic pairs of QTL introgression lines in elite hybrid parental line backgrounds; but proof of concept has been made for nearly all of the areas intended in this project, significantly more senior and junior pearl millet scientists have benefited from theoretical and on-the-job training in marker-assisted backcrossing and assessment of stover quality, and policy changes occurred in the way varietal release decisions are made in India (at least in part due to the efforts of project participants). Without Bruce's single-minded guidance of this project through its proposal development and initial implementation phases, none of this would have happened as soon as it has.

Finally, I thank Peter Ninnes and Oscar Riera-Lizarazu for strongly pushing me to give priority to completing this overdue report, which was to have been submitted earlier, but was delayed due to my slow recovery following a back injury that required surgery.

2 Executive summary

The aim of this project was to improve overall animal productivity in crop-livestock systems of India by increasing the nutritive value of pearl millet stover. Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is the only reliably productive cereal in rainfed areas of north-western India, where pearl millet stover accounts for over 30% of fodder resources available, so genetic improvement of pearl millet stover yield and quality is considered the only practical way to improve the productivity and economic returns of this rainfed crop-livestock production system without substantial public and/or private investments in irrigation and other infrastructure. This project was a follow-up to the Small Restricted Grant AS2/97/98: *Development and use of molecular genetic markers for enhancing the feeding value of cereal crop residues and pastures for ruminants*. Until this project, only limited attention had been paid by cereal breeders in India to either the quantity or quality of stover, despite the high value placed on this component of the crop by farmers in crop-livestock systems. This project attempted to redress this problem, by focusing on genetically improving the yield and quality of crop stover, while maintaining grain yield and other important agronomic traits of adapted, farmer-accepted dual-purpose crop cultivars.

The first project objective was to produce pearl millet hybrid parent lines with enhanced stover quality suitable for use in commercial hybrid seed production, by first identifying flanking markers for additional quantitative trait loci (QTLs) for stover quality traits known to predict animal performance with a high degree of accuracy, then transferring these to several selected hybrid seed parent maintainer lines by marker-assisted backcrossing. These first two activity areas within this objective were largely successful. The third activity area, using NIRS-based full-sib recurrent selection to improve stover quality in restorer populations adapted to the arid zone of central and western Rajasthan was not successful, either because full-sib field trials in the target region were vitiated by field variation in this marginal production environment, or because the base populations used lacked sufficient heritable variation for stover yield and quality traits. However, proof of concept of this activity was demonstrated by successful use of the same recurrent selection protocol in improving stover yield and quality of composite variety ICMV 221 (based on 124 S1 progenies from the broad-based ICRISAT Bold-Seeded Early Composite).

The second project objective was to determine the effects of individual stover quality QTLs, and combinations, in enhanced commercial hybrid parents, on *in vitro* stover quality and *in vivo* animal production. The first two activities in this area were to complete development and testcross hybrid evaluation of a series of near-isogenic chromosome segment substitution lines involving elite *Iniari* landrace-based donor parent 863B and genetically distant elite dual-purpose hybrid seed parent maintainer line ICMB 841. This took longer than expected, but will result in a Ph.D. thesis (supported with complementary funding from Indian government sources) describing the production (first activity) and successful testcross evaluation (second activity) of substitution lines for 6 of the 7 pearl millet chromosome pairs. As the final season's field trials for these "contig line" testcross studies were harvested as this report was being finalized, and data analysis of only 2 of the 4 season's trials were complete, description of the results of these two activities are limited here to presentation of graphical genotypes of the sets of segmental substitution lines produced. In addition, testcross hybrids of the original and improved versions of the maintainer lines bred in the first objective were assessed for grain and stover yield, and *in vitro* stover quality, with sufficient stover being produced of selected experimental and control entry pairs to permit *in vivo* ruminant nutritional quality assessment. As a result, a previously undetected major dominant foliar disease resistance gene was shown to be associated with a major QTL for estimated *in vitro* digestibility in buffered rumen fluid; however, only trends *in vivo* improvement in livestock performance could be detected in the first year of testing.

The final objective of this project was to strengthen awareness of the importance of stover nutritive value and to similarly strengthen national program human resources in India. This was fully successful, resulting in important policy changes in varietal release criteria.

3 Background

The aim of this project was to improve animal productivity in crop-livestock systems of India by increasing the nutritive value of pearl millet stover. The vast majority of the ~10 m ha area on which pearl millet is cultivated in South Asia is rainfed, and dependant upon in-season precipitation. As pearl millet is the only reliably productive cereal in rainfed areas of north-western India, and pearl millet stover accounts for over 30% of fodder resources available in this region, improvement of pearl millet stover yield and quality by genetic means is still considered to be the only practical way to improve the productivity and economic returns of this rainfed crop-livestock production system without substantial public and/or private investments in irrigation and other infrastructure.

This project was a follow-up to the Small Restricted Grant AS2/97/98: *Development and use of molecular genetic markers for enhancing the feeding value of cereal crop residues and pastures for ruminants*. That project was led by Prof. Bruce Stone of Latrobe University and involved ICRISAT, ILRI, CIAT, ICARDA, and NARS in India, Morocco and Tunisia. It finished on 30 June 2000, but work on some components work continued with funding provided by DfID, by ILRI, and by ICRISAT. This new project was more tightly focused than the exploratory AS2/97/98 and included only two of the original partners.

The problem

Stover (dry leaf and stem residues fed to livestock following harvest of grain crops) constitutes a major component of ruminant rations in marginal production environments, particularly during the dry season when green fodder and/or grazing are limited. Low productivity of livestock in smallholder crop-livestock systems in these environments is due in part to limited quantity and low nutritional quality of available stover (Renard 1997). Practices that might improve stover yields and stover quality, such as higher applications of nitrogen (currently averaging only 5-20 kg ha⁻¹, as manure and mineral fertilizer in the target region for this project) are considered very risky for farmers to adopt in our highly unpredictable target environment. The best option for increasing availability and quality of crop residues is genetic improvement of their yield and quality in locally-adapted cultivars.

The approach

Until this project, only limited attention had been paid by cereal breeders in India to either the quantity or quality of stover, despite the high value placed on this component of the crop by farmers in crop-livestock systems (who often based their cultivar adoption decisions as much on stover traits as on grain yield and price). This project attempted to redress this problem by focusing on genetically improving the yield and quality of crop stover, while maintaining the grain yield and other important agronomic traits of adapted, farmer-accepted dual-purpose crop cultivars. Pearl millet, a widely accepted dual-purpose plant, was used as a model system for the following reasons: (a) the high degree of genetic variability for grain and stover yield and quality traits (Kelly *et al.* 1996; Blümmel *et al.* 2003a, 2003b; Blümmel and Bidinger 2007) and demonstrated effectiveness of marker-assisted selection in the crop (Hash *et al.* 2001, 2003), (b) the extensive utilization of this cereal in crop-livestock production systems of resource-poor farmers in the driest arable areas in both South Asia and sub-Saharan Africa (Renard 1997), and (c) the very effective delivery system for improved pearl millet cultivars to smallholder farmers provided by the private sector of the hybrid seed industry in India (Hash 1994).

The country

While the problem of low quality crop residues is common in most developing countries and with most cereals, this project focused upon pearl millet (for the reasons outlined above) and on India. In India, pearl millet stover is extensively used to feed farm livestock and is sold off-farm, especially in areas where the cooperative dairy sector is prominent. A further compelling reason to focus on India is the adoption of hybrid varieties by small-

holder farmers and existence of an effective hybrid pearl millet seed production and marketing industry. Hybrids, mostly dual-purpose types, now cover nearly 70% of the ~10M ha of pearl millet sown annually in India, almost entirely by small farmers (Govila *et al.* 1997; Talukdar *et al.* 1999). ICRISAT continues to be the major supplier of new pearl millet hybrid parental materials to the private sector, assuring the effective delivery of research products from ICRISAT to intended beneficiaries.

Potential value

Of about 10M ha of pearl millet cultivated annually in India, >5M ha are in Rajasthan and 1M ha are in neighbouring Gujarat. Selected districts in these two states, and some areas in the adjoining states of Haryana, Uttar Pradesh and Madhya Pradesh, are considered suitable for the potential cultivars developed in this project (domain 1 of Underwood *et al.* 2000). The most important livestock production systems identified in this domain were millet-dairy buffaloes/cattle and millet-traction, plus millet-meat/wool in the arid areas of western Rajasthan. The combined sorghum and pearl millet domains of Underwood *et al.* (2000) constitute 33% of the 329M ha area of India and 40% of India's 142M ha of net cropped area. These domains support 221M people (>20% of India's population), 46M cattle (23%), 26M buffaloes (33%), and 58M sheep and goats (35%), producing 16M t of milk annually (29% of India's total) (averages of 1989-1995 census data). For these domains it has been calculated that a one percentage unit increase in nutritive value of crop residues of sorghum and pearl millet could result in increases in milk, meat and draught power outputs of 6-8% because pearl millet and sorghum crop stover/residues account for >50% of available fodder resources used for livestock production (Underwood *et al.* 2000). Pearl millet accounts for at least 33% of these combined figures.

Country priority

Pearl millet is the fourth most important cereal in India, and the major cereal grown in the rainfed areas of the drier regions of the country (<400-500 mm rainfall), where it provides food security in the form of a locally-produced staple cereal and essential crop residues to maintain ruminant livestock during the 8-9 month dry season. No alternative dryland cereal can reliably provide the quantities of grain and fodder required to sustain the approximately 60M people and their livestock in these areas. Improved productivity of crop-livestock systems of these areas is a high priority for agricultural research in India (Romney 2003) and was afforded prominence during ACIAR consultations with India.

In recognition of the importance of crop residues, stover yield has recently been added as a primary criterion for selection for official release among entries tested in national pearl millet trials in India. Also, as an outcome of this project and its immediate predecessor, the importance of stover quality is gaining recognition. The former project coordinator of the national pearl millet improvement project argued that research to genetically improve livestock performance per unit of crop residues fed should be given priority over research simply targeting increased crop residue yields (Dr. S.K. Bhatnagar, pers. comm.).

Beneficiaries

The primary beneficiaries of the project outputs will be low- and medium-income farm families growing pearl millet in mixed crop-livestock production systems in north-western India. Improvements in the quantity and quality of pearl millet stover will increase incomes from the sale of stover and animal products, and help maintain animal production/condition during the dry season. Other beneficiaries include members of the seed industry (growers, seed agencies and merchants), and larger-scale livestock/milk producers in pearl millet production regions, and families of labourers employed by these groups.

General strategy

The general strategy of this project was to build on existing knowledge to reach commercially exploitable outcomes suitable for, and deliverable to, smallholder crop-livestock producers in India in a shorter time, and at lower total cost, than could be

achieved by conventional breeding methods. At the same time, this project included scientific comparisons of the effectiveness of individual stover nutritional quality QTLs and of different breeding strategies in accomplishing the goals of the project. The strategy explicitly recognized that the stated aim of the project — improving the value of pearl millet stover — involves a significant number of components, including improved stover yield, a range of stover palatability and ruminant nutritional traits, and the ability to produce products that will reach farmers and meet their requirements.

Farmer-participatory research on priority traits for pearl millet breeding programs has helped to understand the needs and preferences of potential beneficiaries (Underwood *et al.* 2000). Although a high fodder value of stover was ranked overall as the second priority (vs. high grain yield) among farmers' preferences, stover yield was considered the first priority in dry years (in which government drought-relief programs typically meet peoples' needs for food grains, but are unable to meet the demand for fodder for livestock). These surveys have also indicated that farmers and livestock raisers recognize differences in stover quality, and have identified various plant and stover characteristics which they believe are indicative of better livestock feeding quality. This type of information was used in setting the project goal of improving the value (*i.e.*, yield and feed quality) of pearl millet crop residues in adapted hybrids.

Specific research strategy

- To incorporate traits that directly affect animal performance into several single-cross hybrid seed parent lines widely used by the Indian public- and private-sector hybrid seed companies (years 1–3). Marker-assisted selection (MAS) techniques are the most appropriate approach for improvement of a complex attribute such as ruminant nutritional quality in genetically homozygous (inbred) breeding materials such as pearl millet hybrid parental lines. By targeting widely used hybrid parental lines for improvement, the project maximized the probability that improved lines will be rapidly adopted by the seed industry to produce and deliver pearl millet hybrids with improved stover quality to Indian farmers. (See third dot-point below.)
- To improve specific aspects of stover quality in genetically heterogeneous (open-pollinated) breeding materials by conventional recurrent selection (MAS techniques are less cost-effective in this situation). In this, the project attempted to improve the stover quality of landrace-based topcross hybrid pollinators targeted to arid-zone pearl millet-livestock production systems where currently available commercial pearl millet hybrids are not well-adapted, and where animal products are the major source of household income. Targeting the improvement of topcross hybrid pollinators attempted to capitalized on the existing hybrid seed delivery system to get seed of improved products to farmers living in this region (years 1–3).
- To work with private-sector seed companies and the Indian national pearl millet improvement program, to make and evaluate a range of potentially commercially exploitable hybrids (utilizing the improved maintainer lines and topcross pollinators), to accelerate development and marketing of new hybrids with improved stover nutritional quality (years 3–5). ICRISAT is the major supplier of hybrid parental line germplasm to the industry and finished and unfinished products from this project were made freely available to all potential users under the terms and conditions of ICRISAT's standard Material Transfer Agreement (see Appendix 1). Support was provided to collaborators in the form of research information, experimental hybrids for evaluation, and both *in vitro* and *in vivo* stover quality evaluation data.
- To take advantage of work already well advanced in a DfID-funded project to produce and evaluate contiguous segment substitution lines (CSSLs) in the background of elite hybrid seed parent maintainer line ICMB 841, and to produce and evaluate a set of near-isogenic lines for individual stover quality QTLs in this genetic background. *In vitro* and *in vivo* evaluation of these near-isogenic lines and their hybrids provided quantitative information on the specific effects and relative value of individual stover

quality QTLs, allowing marker-assisted improvement of other maintainer lines to focus on QTL(s) with the largest beneficial effects on animal performance (years 1–3).

4 Objectives

The goal of this project was to improve overall productivity in smallholder mixed crop livestock systems in arid and semi-arid in India through the use of pearl millet hybrids with better inherent stover quality. Significant progress towards this goal was realized via the following three objectives:

Objective 1: To produce hybrid parent lines with enhanced stover quality suitable for use in commercial hybrid seed production.

This involves three specific activities:

- **Activity 1:** Identify additional markers for stover quality traits now known to predict animal performance with a high degree of accuracy.
- **Activity 2:** Transfer selected stover quality traits, by marker-assisted backcrossing, into several selected hybrid seed parent maintainer lines that have wide adaptation, good combining ability and ready acceptance by pearl millet seed producers.
- **Activity 3:** Improve stover quality in two restorer populations based on landrace materials adapted to the arid zone of NW Rajasthan, by conventional recurrent selection.

Objective 2: To determine the effects of individual stover quality QTLs, and combinations of enhanced commercial hybrid parents, on *in vitro* stover quality and *in vivo* animal production.

This involved three specific activities:

- **Activity 4:** Develop a series of near-isogenic lines, for individual stover quality QTLs, in the background of the widely adapted and economically important hybrid seed parent maintainer line 841B.
- **Activity 5:** Produce sets of experimental hybrids using the (i) the original and improved versions of the maintainer lines and restorer populations bred in objective 1, and (ii) the near-isogenic lines for individual stover yield and quality QTLs bred in activity 4, and produce sufficient stover for *in vitro* and *in vivo* ruminant nutritional quality assessment.
- **Activity 6:** Determine the improvement made in the nutritional quality of the stover of the experimental hybrids (Activity 5) and evaluate the specific effects of all individual stover nutritional quality QTLs (individually and in combination) using *in vitro*, NIRS and *in vivo* analyses.

Objective 3: Promote the use of improved parental lines to public and private seed companies.

This involved two specific activities:

- **Activity 7:** Evaluate the quality of currently marketed pearl millet hybrids, and use the results to raise awareness of fodder quality issues with the public and private seed industry.
- **Activity 8:** Encourage the public and private seed producers to use the improved parental lines by providing them information on quality characteristics and animal performance, experimental quantities of seed, and analytical assistance.

5 Methodology

Activity 1: Identify additional markers for stover quality traits now known to predict animal performance with a high degree of accuracy.

We used recent research results to identify markers for additional quality traits that are now known to predict animal performance with a high degree of accuracy. We used NIRS to repeat phenotypic assessment of stored stover samples from the mapping population used in the initial project (ICMB 841 × 863B). These stover sets consisted of 162 testcross entries (78 F_{2:4} mapping progenies × two restorers) from the mapping population grown in 1998, 1999 and 2000 in three locations and under different management conditions. Approximately 13,000 samples from these experiments had been stored safely and were available for stover quality assessments. For example, after testing 40 pearl millet stover samples within this project, protein losses/accretion in sheep fed pearl millet stover can be predicted with high accuracy ($R^2 > 0.80$) using multiple regression based on stover *in vitro* rumen fermentation characteristics, chemical constituents and morphological composition. Previously we had developed several regression equations for stover quality prediction based on a wide range of *in vitro*, chemical, structural and morphological laboratory measurements. The final choice of measurements depended on a) contribution to animal performance, b) heritability of a given measurement and c) relationship of measurement with other desirable agronomic traits. Because the mapping population progenies were evaluated in testcross form, the QTLs detected are for combining ability for stover quality. This means that they should directly predict the effectiveness of these QTLs in hybrid combination, *i.e.*, when incorporated in hybrid parents. This work was done by Dr. Nepolean and several Visiting Scientists supported by the project, working with Drs. Blümmel and Hash at ICRISAT-Patancheru, with the assistance of the field, marker laboratory, and livestock nutrition technicians funded by the project.

Activity 2: Transfer selected stover quality traits, by marker-assisted backcrossing, into several selected hybrid seed parent maintainer lines that have wide adaptation, good combining ability and ready acceptance by pearl millet seed producers.

A marker-assisted backcross program transferred target QTLs to three new genetic backgrounds, but this took longer than the 2–3 years originally envisaged. The three recurrent parents originally proposed to be used were: ICMB 841, the elite seed parent maintainer line used as a parent of the mapping population itself, and seed parent of popular released single-cross hybrid Pusa 23, which continues to be widely grown in central Rajasthan, with its seed being produced primarily by the Cooperative Marketing Federation in Gujarat as an irrigated summer crop so that its high quality stover becomes available during the hot, dry summer months when there are few good alternative sources of fodder for Gujarat's cooperative dairy industry; ICMB 95222, the seed parent maintainer line for two dual-purpose hybrid cultivars released in India in 2003 (HHB 146 from CCS Haryana Agricultural University, and GHB 526 (released for summer cultivation) from Gujarat Agricultural University), and ICMB 00999, seed parent maintainer of a high-yielding forage hybrid identified in ICRISAT trials in 2002. Based on discussions among informed pearl millet breeders at the project launch meeting held at ICRISAT-Patancheru in August 2004, we dropped ICMB 00999 as a recurrent parent, and replace it with ICMB 93333 (maintainer of a promising hybrid, RHB 127, from Rajasthan Agricultural University, ICMB 95111 (maintainer of several promising hybrids in advanced stages of testing in state and national program trials), and HMS 7B (maintainer of superb dual-purpose hybrid HHB 117 (from CCS Haryana Agricultural University, which is characterized by many thin stems and maintenance of green leaf area through to grain physiological maturity and beyond due to high levels of foliar disease resistance).

Transfer of stover quality QTLs from 863B to ICMB 841 background was achieved by first completing development of approximately 35 substitution lines, each having a different

segmental genomic substitution from 863B and together offering reasonably complete coverage of the genome with segmental substitutions (at least as far as the modest numbers of available SNP, SSR and STS markers for pearl millet permit us to check). This activity advanced from the BC₄F₁ generation to BC₆F₃ (with additional support from the Government of India's Department of Biotechnology, which provided support of a locally-recruited Ph.D. student, Ms. B. Ramana Kumari) and testcrosses of homozygous partial sets of substitution line were first ready for testing in the rainy season of 2008, as it took longer than expected to generate the desired substitution segregants. The parent lines for the backcrossing program had been selected after genotyping at 27 SSR and 18 RFLP marker loci in order to identify the most promising ones to be advanced to families of BC₄F₁/BC₃F₂ pairs. This procedure identified combinations of markers that impart significant improvements in stover yield and/or quality over and above that of the unmodified parent line ICMB 841.

Recurrent parent maintainer lines HMS 7B, ICMB 93333, ICMB 95111 and ICMB 95222 were improved in parallel (as completion of the work on ICMB 841 took longer than anticipated), using several combinations of stover quality markers. This provided a range of hybrids with enhanced stover quality for different ecological adaptation zones and/or with different plant types, and thereby broadened the range of genetic material available on which to base evaluations of the effects of MAS for improved stover quality on animal productivity.

This work was done at ICRISAT-Patancheru by Dr. T. Nepolean working with Visiting Scientists from the National Program (supported by ACIAR), with the CSSL study being primarily done by an ICRISAT Research Scholar (Ms. B. Raman Kumari; supported by a complementary grant provided to ICRISAT by the Government of India's Department of Biotechnology).

Activity 3. Improve stover quality in two restorer populations based on landrace materials adapted to the arid zone of NW Rajasthan, by conventional recurrent selection.

Previous demonstration that NIRS analysis can rapidly, quantitatively and accurately assess stover quality parameters in small stover samples, meant that conventional population improvement of stover nutritional quality had also become feasible as the proposal for this project was developed, where previously this was not practical when sample analysis had to be done by expensive and time-consuming conventional laboratory methods. We applied the approach outlined in Activity 1. We conducted two cycles of full-sib progeny recurrent selection for specific stover quality traits (e.g., high animal intake, high digestibility, etc.) in two pearl millet topcross pollinators (open-pollinated restorer populations) based on landrace germplasm adapted to the arid zone of NW India, as well as a broad-based improved open-pollinated variety (ICMV 88904; Witcombe *et al.* 1997) that has been released in India (as ICMV 221), Eritrea (as Kona), and was in advanced stages of evaluation prior to subsequent release in Ethiopia, Kenya, and Uganda.

This involved the making of, and field evaluation of, 200-300 progenies per population per year and NIRS analyses of 400-600 stover samples per population per year. This was intended to produce pollinator populations with improved stover quality for use to produce adapted topcross hybrids (with both improved and unimproved seed parents), which specifically target the livestock based-farming systems of the arid zone, where conventional single-cross hybrids are not well adapted. Unfortunately, the procedure was not successful in improving the two topcross pollinators (either because they had genetic bases too narrow to permit such improvement, or because of field variability at the Rajasthan rainy season evaluation sites to which we had access for producing the stover samples that were used in selecting progenies to be recombined each cycle). However, the procedure was successful in improving stover value (yield and quality) of pearl millet composite variety ICMV 221, for which the progeny trials were conducted at ICRISAT-Patancheru.

This activity provided at least a qualitative comparison of conventional selection vs. marker-assisted selection for improving pearl millet stover quality. This work was conducted by Dr. Bidinger and Ms. Sunita Choudhary — a Visiting Scientist in the project — and Dr. Blümmel in the context of the arid zone pearl millet breeding project run jointly by ICRISAT and the Central Arid Zone Research Institute, Jodhpur, India. In addition, the modes of inheritance of important stover fodder quality traits such as nitrogen content and *in vitro* digestibility were investigated by studying segregation patterns among progeny obtained by crossing parents contrasting for these traits.

Activity 4: Develop a series of near-isogenic lines, for individual stover quality QTLs, in the background of the widely adapted and economically important hybrid seed parent maintainer line 841B (= ICMB 841).

With funding from DfID we had previously initiated production of a set of contiguous segment substitution lines (referred to subsequently as contig lines or CSSLs) by ³²P-labelled RFLP- (initially), STS- and SSR- (since 2003), and SSCP-SNP- (since 2005) based marker-assisted backcross transfer of chromosome segments from donor parent 863B into the background of recurrent parent 841B = ICMB 841, with the purpose of exploring the use of such contig lines as tools for QTL detection and validation, as well as improving yield and other traits, such as disease resistance and drought tolerance, of an economically important hybrid parental line (ICMB 841). The early backcross generation materials available for development of 841B-background contigs carrying specific chromosome regions from 863B containing favorable alleles at QTLs for various aspects of stover quality identified in Activity 1, were to have been backcrossed to 841B, with marker analysis of the progeny to rapidly produce a set of near-isogenic versions of 841B for each of the specific stover quality QTLs (and materials of use for future fine-mapping of these if that proves necessary due to undesirable linkage drag associated with introgression of particular QTLs). However, delays in completing the QTL analysis meant that we just carried forward with development of the full contig line set (which by definition includes individual lines having substitutions of 863B alleles, whether favorable or unfavorable, for essentially all of the QTLs detected with the F_{2:4} mapping population testcrosses).

The sets of near-isogenic line pairs for each target QTL now allow assessment of the effects of an individual QTL on a range of specific stover quality traits (*in vitro* analysis), as well on animal performance itself (*in vivo* evaluation), providing a profile of individual QTL effects. This information is proving very useful in prioritizing QTLs for future applied use in breeding pearl millet hybrids (and hybrid parental lines) with improved stover quality.

For a given target QTL the backcrossing program using then-available partially introgressed contigs from 863B was to have been completed cost-effectively in four plant generations. In terms of this ACIAR-supported project, the parental generation was to have consisted of three typical plants each of the BC_nF₃ contig lines homozygous for a particular target introgression from 863B along with the 841B recurrent parent, and these were to have been used to produce three BC_{n+1}F₁ families. Three typical BC_{n+1}F₁ plants (limited SSR marker genotyping optional) in one of these three families were then to have been backcrossed plant × plant to typical recurrent parent 841B plants. Marker genotyping only seven BC_{n+2}F₁ plants from one or more of the three product families at loci in the vicinity of the particular target QTL was then needed to provide >99% probability of identifying at least one desirable segregant heterozygous for the target QTL (and having the frequency of 863B alleles on non-target chromosomes reduced by an average of 75% relative to the BC_nF₃ contig line used initially as the QTL donor). These selected QTL introgression heterozygotes were to have been advanced two generations with selfing to generate homozygous near-isogenic line pairs for particular target QTLs. Marker genotyping of about 25 selfed individuals from each progeny was required then to identify the homozygous pairs of near-isogenic individuals that could be advanced following harvest to produce finished near-isogenic line pairs for each particular target QTL. This

work was to have been done at ICRISAT-Patancheru, by the project-supported scientist (Dr. T. Nepolean) and Visiting Scientists from the Indian National program, but was primarily the responsibility of a Ph.D. student (Ms. B. Ramana Kumari) supported by the complementary grant from the Government of India's Department of Biotechnology, under the supervision of Dr. Hash and Dr. Nepolean, with the assistance of the project-supported field and marker-laboratory technicians.

Activity 5: Produce sets of experimental hybrids using the (i) the original and improved versions of the maintainer lines and restorer populations bred in objective 1, and (ii) the near-isogenic lines for individual stover yield and quality QTLs bred in activity 4, and produce sufficient stover for *in vitro* and *in vivo* ruminant nutritional quality assessment.

When the breeding projects described in Activities 2 and 4 were completed, the improved parental genotypes produced were testcrossed to appropriate testers (male parental lines of released hybrids produced on the male-sterile line counterparts of the elite seed parent maintainer lines using in the marker-assisted backcrossing programmes described in Activities 2 and 4 above) to produce experimental hybrids for stover quality evaluation. As the attempt in Activity 3 to improve digestibility of the restorer populations was not successful, and the Project Scientist (Dr. Fran Bidinger) leading this activity passed away unexpectedly in April 2008, we did not produce and evaluate testcrosses of the Cycle 0, Cycle 1 and Cycle 2 restorer populations and, instead, restricted our work with that material to evaluating the population cycle bulks and experimental varieties themselves. Pairs of experimental hybrids were made using both the original and improved versions of the recurrent parents from the marker-assisted backcrossing programmes, to allow quantitative assessment of the effects of introgressing specific stover quality QTLs on traits related to agronomic performance (including flowering time, plant height, grain yield and stover yield) and stover quality. The widely adapted maintainer lines used as recurrent parents (Activity 2) were testcrossed to widely used restorers (parents of successful commercial hybrids or of recently released hybrids). The 841B near-isogenic lines (Activity 4), along with their recurrent parent 841B and donor parent 863B, were testcrossed to three genetically and phenotypically diverse restorer lines (H 77/833-2, PPMI 301, and RIB 3135-18) that are male parents of released public-bred hybrids thought to be well-adapted to rainfed crop-livestock production systems the low (<400 mm mean annual precipitation) and moderate (400–500 mm) rainfall areas of the primary target environments in the Indian states of Gujarat, Haryana and Rajasthan. The improved topcross pollinators (Activity 3) were to have been crossed on male-sterile lines ICMA 91444 and ICMA 93333, which had previously been identified as having appropriate combining ability to produce hybrids adapted to the arid zone (Bidinger *et al.* 2002).

The experimental hybrids produced were sown in multi-environment trials (3–5 locations) to generate stover samples for laboratory (*in vitro* and NIRS) analyses to assess both genotype and genotype × environment effects on stover quality differences in the hybrid pairs. Subsets of these near-isogenic testcross hybrid pairs were also be grown at ICRISAT and in Rajasthan in replicated large-plot trials to produce sufficient quantities of stover for *in vivo* assessment of animal performance in both restricted and *ad libitum* feeding trials, with and without supplementation. This work was conducted by Drs. Hash and Nepolean, with the assistance of the field technician supported by this project, and (in case of the contig line testcross trials conducted in six environments across two years and two seasons at ICRISAT-Patancheru) as part of the Ph.D. thesis research program of Ms. B. Ramana Kumari (to be completed in 2011, and largely supported by a complementary grant from the Government of India's Department of Biotechnology).

Activity 6: Determine the improvement made in the nutritional quality of the stover of the experimental hybrids (Activity 5) and evaluate the specific effects of all individual stover nutritional quality QTLs (individually and in combination) using in vitro, NIRS and in vivo analyses.

Stover fodder quality ultimately can be measured only by assessing actual livestock productivity using measures such as meat and milk production, or draught power output, when the stover is provided to these animals as feed. However, livestock productivity trials cannot be used as routine tools in crop improvement because of their logistical and infrastructural demands. Never-the-less, they play an important role in validating estimates of stover fodder quality provided by simpler and more rapid methods that require smaller samples. For such estimates we require simpler quality assessment protocols that permit rapid determination of the relative fodder values of many small samples, to provide simple yet meaningful laboratory estimates of fodder quality traits in larger numbers of samples as one might find in the early segregating generations of a plant breeding programme. In the current project we investigated chemical (nitrogen, fiber and sugar), morphological (stem diameter; stem: leaf blade: leaf sheath ratios; and green leaf area), structural (grinding energy requirements) and *in vitro* (digestibility, rate and extent of fermentation, microbial efficiency) traits in 40 large pearl millet stover samples. These stover samples were extensively tested with sheep to identify those laboratory traits (and combinations of traits), that were highly correlated with the actual livestock productivity measurements. NIRS was then calibrated and validated for the selected laboratory traits. More than 800 stover samples were analyzed conventionally for developing the NIRS equations and for validating them. This work was conducted at ICRISAT-Patancheru by Dr. Blümmel, with the assistance of technicians supported by the project, and the Visiting Scientist in the 1st half of the project, Dr. G. Alexander.

Unfortunately, due to delays in completing the marker-assisted backcrossing, and subsequent evaluation of the introgression line testcrosses, we were unable to implement the originally planned multiplication and on-farm testing of selected improved hybrids with milking animals using established collaborative infrastructure with NGOs within the logistical framework of another ILRI project.

Activity 7: Evaluate the quality of currently marketed pearl millet hybrids, and use the results to raise awareness of fodder quality issues with the public and private seed industry.

ICRISAT exploited its close linkages with the public- and private-sector seed companies in India that produce or market pearl millet hybrids, to raise awareness of the potential for improvement of pearl millet stover quality. ICRISAT is the major supplier of hybrid parent germplasm (breeding lines and finished parent lines) to both sectors of the pearl millet seed industry in India, and these in turn are the major conduits by which the products of ICRISAT research reach Indian farmers. This relationship is formally recognized by the provision, by consortia of public- and private-sector seed companies, of operational support for ICRISAT's applied pearl millet, sorghum, and pigeon pea hybrid parental line breeding programs. It was also recognized by the incorporation of activities in this ACIAR project into the set of agreed activities in the ICAR-ICRISAT collaborative research project on pearl millet marker-assisted breeding for the period 2006-2009.

In anticipation of this project, ILRI procured pearl millet seed material from the private seed industry that included a wide range of popular pearl millet hybrids. The nutritive value of stover of these materials were determined in the ILRI laboratory and animal experimental facilities at ICRISAT-Patancheru, with specific results made available to owners of the hybrids and general results shared with the industry as a whole. This program provided the basis for discussions with the seed industry, including the potential for genetic improvement of pearl millet stover quality, the relationship of quality parameters to various agronomic characteristics in current commercial hybrids, and its value as a marketing tool.

We also used this program to keep the seed industry well informed of work in progress on improving pearl millet stover quality, through written reports, presentations during field days and national meetings and through existing, regular personal contacts with both public- and private-sector plant breeders. This activity involved all project personnel.

Activity 8: Encourage the public and private seed producers to use the improved parental lines by providing them information on quality characteristics and animal performance, experimental quantities of seed, and analytical assistance.

We have made the improved versions of parental lines of released pearl millet hybrids available to both public- and private-sector plant breeders for their evaluation under the terms and conditions of ICRISAT's Revised Material Transfer Agreement for products of our public-supported plant breeding research, which itself is an annex to the Standard Material Transfer Agreement for the exchange of Plant Genetic Resources for Food and Agriculture (see Appendix 1); encouraged these plant breeders to participate in the multi-environment trials of stover yield and quality of experimental hybrids made with these parents; and shared with them all data and results summaries obtained from these trials. In future we will publish results of this work in both the scientific and popular literature, and continue to make appropriate presentations at national and international scientific meetings.

Although the project has ultimately been successful in producing experimental hybrids with significantly improved stover quality, we are only now in a position to multiply seed of experimental hybrids and provide technical assistance to public and private sector organizations (at their cost) to conduct participatory evaluations with farmers of the productivity of the hybrids and of animals fed on their stover. Originally, these on-farm assessments were to have been conducted in the final two years of the project, with support of these activities by the technicians funded by the project, under the supervision of Drs. Hash and Blümmel. However, we now need to identify funding that will permit us to bring the valuable findings of this project to Indian pearl millet-producing farmers *via* on-farm trials.

6 Achievements against activities and outputs/milestones

Objective 1: To produce hybrid parent lines with enhanced stover quality suitable for use in commercial hybrid seed production

No.	Activity	Outputs/ Milestones	Completion date	Comments
1	Identify additional markers for stover quality traits now known to predict animal performance with a high degree of accuracy	Availability of complete stover quality profiles for four phenotyping data sets Sets of QTL for various aspects of stover quality	Dec 2005 Jan 2006	Completion of analysis of data sets delayed Results partially published in an ISMN article by Nepolean <i>et al.</i> (2006)
2	Transfer selected stover quality traits, by marker-assisted backcrossing, into several selected hybrid seed parent maintainer lines that have wide adaptation, good combining ability and ready acceptance by pearl millet seed producers	** BC ₁ F ₁ seed of all crosses ** BC ₂ F ₁ seed of all crosses ** BC ₃ F ₁ seed of all crosses ** BC ₅ F ₁ seed of all crosses ** BC ₅ F ₂ seed of all crosses ** BC ₅ F ₃ seed of all crosses, and their counterpart testcrosses available for initial field testing in 2006 rainy season	Not completed Not completed Not completed	Attempted four recurrent parents when only two were targeted; two of four recurrent parents proved difficult to use due to their high level of susceptibility to the ICRISAT-Patancheru field strain of pearl millet downy mildew Advanced successfully with three of four recurrent parents Three recurrent parents advanced to at least BC ₃ F ₄ and/or BC ₄ F ₃ generations Completed marker-assisted backcross QTL introgressions (producing BC ₃ F ₃ and BC ₄ F ₃ introgression homozygotes) for favourable alleles from ICMB 841 and from 863B in the genetic backgrounds of three recurrent parents (HMS 7B, ICMB 93333, and ICMB 95222) when only two recurrent parents were originally proposed Testcross seed of BC ₃ F ₃ and BC ₄ F ₃ lines was produced for use in replicated observation nurseries, replicated field trials (small plots), and in selected cases large plot replicated field trials (the latter to produce straw samples large enough for <i>in vivo</i> quality assessment in sheep feeding trials)

No.	Activity	Outputs/ Milestones	Completion date	Comments
3	Improve stover quality in two restorer populations based on landrace materials adapted to the arid zone of NW Rajasthan, by conventional recurrent selection	<p>** Identification of two populations with good intra-population variation for selected stover quality traits</p> <p>** First cycle progenies for evaluation</p> <p>** Identification of superior progenies for recombination to make cycle 1</p> <p>** Identification of superior progenies for recombination to make cycle 2</p> <p>** Completion of second cycle of recurrent selection</p>	<p>Dry season 2003 (before start of project)</p> <p>Rainy season 2003</p> <p>Post-rainy season 2003/04</p> <p>Post-rainy season 2004/05</p> <p>Dry season 2005</p>	<p>We were not able to improve the stover quality of either of the two landrace-based pollinator populations adapted to central and western Rajasthan. This failure was:</p> <ul style="list-style-type: none"> • because the requisite genetic variability was not present in these rather narrow-based populations, or • because field variability was so large that most observed variation for stover yield and quality traits in the marginal rainfed phenotyping environments used for assessing the full-sib progenies during the two cycles of recurrent selection was due to micro-environment and had no genetic basis and the three cycle bulks and related experimental varieties these environments. <p>However, full-sib recurrent selection for both stover yield and quality, while maintaining grain yield, disease resistance and growth duration, was successful in the broad-based pearl millet composite ICMV 221, for which full-sib evaluation was undertaken at Patancheru (an appropriate environment for this improved variety adapted to peninsular Indian conditions and also released in Eritrea, Ethiopia, Kenya, Mexico, and Uganda).</p>

PC = partner country

Objective 2: To determine the effects of individual stover quality QTLs, and combinations of enhanced commercial hybrid parents, on in vitro stover quality and in vivo animal production

No.	Activity	Outputs/ Milestones	Completion date	Comments
4	<p>Develop a series of near-isogenic lines, for individual stover quality QTLs, in the background of the widely adapted and economically important hybrid seed parent maintainer line 841B = ICMB 841</p>	<p>** Identification of 841B contig lines with one or more selected stover yield & stover quality QTL(s) ** 105 BC_{n+1}F₁ families ** 315 BC_{n+2}F₁ families & their paired BC_{n+1}F₂ families ** 2175 BC_{n+2}F₂ families ** Genotype information for selection of a small subset (<20) of the available 2175 BC_{n+2}F₂ families to advance to create near-isogenic pairs targeted to specific stover yield & quality QTLs ** Near-isogenic pairs for each targeted stover yield & quality QTL from the (ICMB 841 × 863B)-based mapping population, in the genetic background of elite seed parent maintainer line 841B = ICMB 841 ** Seed of testcross hybrids for these near-isogenic pairs ** Stover samples from near-isogenic pairs (and hybrids produced using these near-isogenic pairs) for lab-scale analysis; information on grain and stover yield costs associated with each targeted stover quality QTL</p>	<p>2007/08 post-rainy season for linkage group 3, 4 and 5 contig line sets; and 2008 rainy season for linkage groups 1, 6 and 7 (and part of linkage group 2)</p> <p>One season following near-isogenic set production</p> <p>Stover samples from different NIL testcross sets were produced in rainy seasons of 2008, 2009 & 2010, & dry seasons (stress & non-stress) of 2009 & 2010</p>	<p>Delay in generation of a reliable QTL analysis led to a change of plans such that instead of developing NIL sets for selected target QTLs, instead we pushed ahead attempting to develop a contiguous segment substitution line set covering the full map length of the (ICMB 841 × 863B)-derived mapping population</p> <p>The set of substitution lines for linkage group 2, which we thought we had completed during the course of a Ph.D. thesis study within an earlier DfID-supported project, were later demonstrated to not have the expected introgressions – probably as the result of tracking errors made in the penultimate or final generations (BC₅ or BC₆); so our set of contiguous segmental substitution lines with donor parent 863B introgressions in the genetic background of elite maintainer line ICMB 841 is still incomplete</p> <p>Testcross hybrid seed produced for all near-isogenic linkage group substitution line sets except for linkage group 2</p> <p>Stover analysis of samples produced through the 2010 dry season has been completed, while sample preparation of those produced during the 2010 rainy season has just begun; once the NIRS-predicted stover quality data set is complete, a full across-environment analysis will be completed for the near-isogenic testcrosses evaluated for linkage groups 1, 3, 4, 5, 6, and 7, and these results will be included in the Ph.D. thesis of Ms. B. Ramana Kumari, which is due to be submitted in Mar 2011</p>

No.	Activity	Outputs/ Milestones	Completion date	Comments
5	Produce sets of experimental hybrids using the (i) the original and improved versions of the maintainer lines and restorer populations bred in objective 1, and (ii) the near-isogenic lines for individual stover yield and quality QTLs bred in activity 4, and produce sufficient stover for <i>in vitro</i> and <i>in vivo</i> ruminant nutritional quality assessment	<p>** Set of topcross hybrids based on the original, cycle 1 and cycle 2 versions of the two arid-zone adapted restorer populations</p> <p>** Stover samples of hybrids based on the original, cycle 1 and cycle 2 versions of the two arid-zone adapted restorer populations</p> <p>** Set of near-isogenic hybrids differing for specific stover yield and quality QTLs</p> <p>** Material for <i>in vivo</i> assessment of effects of individual stover yield and quality QTLs on animal performance</p>	<p>Not achieved as no progress in improving the two restorer populations themselves was achieved</p> <p>Not achieved, as above</p> <p>Dry seasons (Jan-May) of, 2008, 2009 & 2010 (under both stress & non-stress conditions) & rainy seasons (Jun-Oct) of 2008, 2009 & 2010</p> <p>Rainy seasons of 2008 & 2009</p>	<p>As for Activity 3 above; instead produced testcross hybrids (produced on male-sterile lines ICMA 95111 and ICMA 99022) of h_xh, h_xl and l_xl full-sib progeny crosses (targeting stover nitrogen and stover <i>in vitro</i> digestibility) produced in the genetic background of pearl millet composite (open-pollinated variety) ICMV 221</p> <p>As for Activity 3 above; instead evaluated testcross hybrids (produced on male-sterile lines ICMA 95111 and ICMA 99022) of h_xh, h_xl and l_xl full-sib progeny crosses (targeting stover nitrogen and stover <i>in vitro</i> digestibility) produced in the genetic background of pearl millet composite (open-pollinated variety) ICMV 221</p> <p>Small-plot trials of the original and improved versions of the three maintainer line recurrent parents (HMS 7B, ICMB 93333 & ICMB 95222) were evaluated at ICRISAT-Patancheru and at collaborating national programme sites</p> <p>Large plot (8 rows of 100 m length spaced 50 to 75 cm apart) trials (two replications per environment) of pairs of near-isogenic hybrids were conducted at ICRISAT-Patancheru and at the AICPMIP Coordination Unit test site at Rajasthan Agricultural University's Agricultural Research Station Mandore, near Jodhpur, Rajasthan</p>

6	Determine the improvement made in the nutritional quality of the stover of the experimental hybrids (Activity 5) and evaluate the specific effects of all individual stover nutritional quality QTLs (singly and in combination) using <i>in vitro</i>, NIRS and <i>in vivo</i> analyses	<p>** Quantitative data on the effectiveness of recurrent phenotypic selection for improved stover quality in restorer populations</p> <p>** Quantitative data on the effects on laboratory quality assessment and on animal performance of individual stover yield and quality QTLs</p> <p>** Quantitative data on the effectiveness of marker-assisted backcross improvement of stover yield and quality of hybrid seed parents</p>	<p>2008/09 post-rainy season & 2009 rainy season</p> <p>2007/08 post-rainy season & 2008 rainy season</p> <p>2008/09 post-rainy season & 2009 dry season</p>	Improvement in stover fodder quality traits was clearer in laboratory trials than the <i>in vivo</i> experiments; the latter were short term due to limited access to stover for the trials. More animal work with improved cultivars is planned for 2011 under the Bill and Melinda Gates Foundation HOPE project, and under farmer conditions.
---	---	---	--	--

PC = partner country

Objective 3: To promote the use of improved parental lines to public and private seed companies

No.	Activity	Outputs/ Milestones	Completion date	Comments
7	Evaluate the quality of currently marketed pearl millet hybrids, and use the results to raise awareness of fodder quality issues with the public and private seed industry	** Ranges in stover yield and quality of currently used hybrids quantified and relationships between desirable defined	2007/08, 2008/09 & 2009/10 post-rainy seasons	Pearl millet hybrid GK 1044 was identified as having superior stover yield and quality attributes, but has subsequently succumbed to an epidemic of pearl millet blast (a disease previously considered to be only a cosmetic problem in pearl millet in India); the parental line(s) of this hybrid can now be improved by applied marker-assisted backcrossing to introduce additive and dominant genes for host plant resistance to this foliar disease that devastates stover palatability and nutritional value, provided that the private company that previously bred and marketed this hybrid expresses interest in doing so.

8	<p>Encourage the public and private seed producers to use the improved parental lines by providing them information on quality characteristics and animal performance, experimental quantities of seed, and analytical assistance</p>	<p>** Uptake of improved pearl millet hybrid parental materials promoted</p>	<p>Rainy season 2010</p>	<p>Only now do we have enough supporting evidence to be able to provide convincing arguments that we have indeed been able to produce versions of seed parent maintainer line ICMB 95222 that are capable of producing higher yields of digestible dry matter (as a result of reducing losses caused by the pearl millet blast pathogen, <i>Pyricularia grisea</i>).</p>
---	--	--	--------------------------	--

PC = partner country

7 Key results and discussion

Activity 1: Identification of additional markers for stover quality traits now known to predict animal performance with a high degree of accuracy

QTLs for stover *in vitro* organic matter digestibility (IVOMD) and metabolizable energy (ME) content were identified using stover samples from replicated field trials of testcross hybrids of the (ICMB 841-P3 x 863B-P2)-derived F_{2:4} mapping population (Nepolean *et al.* 2006). Briefly, based on calibrated NIRS analysis of stover samples from a set of testcrosses based on high-tillering, regionally-adapted tester H 77/833-2 and a set of 149 partially inbred mapping population progenies, grown in field trials (3-replications x 2 rows of 4-m length) conducted at ICRISAT-Patancheru and CAZRI-Jodhpur during the rainy seasons of 2004 and 2005. Using the highly heritable data set from the 2005 Patancheru season we identified putative QTLs for stover quality traits (as well as grain and stover yield) listed in Table 1.

Table 1: QTLs mapped for stover quality-related traits and dry stover yield sharing common genetic regions. The marker intervals cited are those flanking the peak of the LOD scan. Position is the QTL peak from the left marker given in Haldane cM. R² represents the percentage of phenotypic variance explained for each QTL (using a composite interval mapping algorithm). A positive sign of the additive effects indicates the 863B allele of the QTL had a numerically positive effect (favourable or not) on the trait.

Trait	LG	Marker interval	Position	LOD	R ²	Additive effects
GAS24 (mL/200 mg dry matter)	1	<i>Bm1RA10a</i> – <i>Xpsm761</i>	18	3.5	10.8	0.422
	2	<i>Xpsmp2066</i> – <i>Xpsm380</i>	70	5.5	16.5	0.575
	4*	<i>Xpsm1003d</i> – <i>Xpsm716</i>	18	3.1	10.1	0.379
	6	<i>Xpsm514</i> – <i>TstRA6c</i>	30	4.5	13.8	0.52
IVOMD (%)	1	<i>Xpsm761</i> – <i>Bm3RA1c</i>	20	3.1	9.7	0.294
	2	<i>Xpsmp2066</i> – <i>Xpsm380</i>	70	4.0	12.2	0.341
	6	<i>Xpsm514</i> – <i>TstRA6c</i>	30	4.2	12.8	0.348
ME (mJ/kg)	1	<i>Bm1RA10a</i> – <i>Xpsm761</i>	18	3.3	10.2	0.053
	2	<i>Xpsmp2066</i> – <i>Xpsm380</i>	70	4.1	12.5	0.063
	6	<i>TstRA6c</i> – <i>Xicmp3002</i>	34	3.1	9.6	0.059

Trait	LG	Marker interval	Position	LOD	R ²	Additive effects
NDM (%)	2	<i>Xpsmp2066</i> – <i>Xpsm380</i>	68	5.0	15.2	-0.018
	4	<i>Xpsm265</i> – <i>Xpsm344</i>	22	2.7	8.5	-0.012
	5	<i>Xctm25</i> – <i>Xpsmp2064</i>	68	5.3	15.9	0.027
	5	<i>Xpsms53</i> – <i>Xpsms70</i>	98	3.8	12.1	-0.022
	6	<i>Xpsmp2213</i> – <i>Xpsm713</i>	28	6.6	19.6	-0.019
SUGSDM (%)	2	<i>Xpsmp2066</i> – <i>Xpsm380</i>	68	4.2	13.0	0.268
	5	<i>Xpsm318</i> – <i>Xpsms18</i>	90	4.6	14.4	-0.236
	6	<i>TstRA6c</i> – <i>Xicmp3002</i>	34	4.8	14.5	0.264
DSY (kg/ha)	3	<i>Xctm10</i> – <i>Xpsm174</i>	46	5.0	14.9	74.1
	5	<i>Xpsmp2064</i> – <i>Xpsm318</i>	84	9.0	26.0	-104.9
	6	<i>TstRA6c</i> – <i>lcmp3002</i>	36	7.2	20.9	101.2
GY (kg/ha)	5	<i>Xicmp3027</i> – <i>Bm1RA1d</i>	0	3.2	10.4	39.1
	5	<i>Bm1RA5a</i> – <i>Bm3Ra1d</i>	36	2.8	8.7	-39.2
	5	<i>Xpsms70</i> – <i>Xpsm732.1</i>	104	3.5	11.3	-25.7

GAS24: gas volume generated during 24 h *in vitro* digestion of 200 mg dry matter, IVOMD: *in-vitro* organic matter digestibility, NDM: nitrogen on dry matter basis, ME: metabolizable energy, SUGSDM: sugar content, DSY: dry stover yield, GY: grain yield

* Increased gas volume from the 863B allele at this QTL is likely conditioned by a major dominant gene for host plant resistance to blast disease caused by *Pyricularia grisea* (Nepolean *et al.* 2010)

Activity 2: MAS and progress in validating QTLs of stover fodder quality traits

In anticipation of the success of that QTL-mapping exercise describe above, we initiated SSR marker-based backcross introgression of favourable alleles from both mapping population parental lines, ICMB 841-P3 or 863B-P2, by blindly crossing, and then backcrossing these two potential donors of improved stover yield and quality traits to a set of four elite hybrid parental lines selected by consensus of pearl millet breeding teams

from ICRISAT, AICPMIP, state agricultural universities (SAU) from Gujarat, Haryana and Rajasthan, and several major private-sector seed companies. The recurrent parents thus chosen included two with which we faced tremendous difficulties (ICMB 95111 and HMS 7B) because of their high level of susceptibility to the ICRISAT-Patancheru field population of the pearl millet downy mildew pathogen, *Sclerospora graminicola*. Both of these candidate recurrent parents were sufficiently susceptible so that nearly all true-to-type plants succumbed to downy mildew in the field, before flowering, and primarily outcrossed plants that could not reliably be used as recurrent parents remained for use in the crossing programme. After three field seasons attempting to work with these materials, we abandoned work on ICMB 95111 and concentrated our efforts on HMS 7B (with which we were able to make progress by moving the work into a greenhouse free from soil- and air-borne downy mildew inoculum) and the two remaining recurrent parents ICMB 93333 (female parent maintainer of a superb hybrid from RAU-Durgapura, RHB 127, which for various technical reasons has not been notified for commercial multiplication), and ICMB 95222 (female parent maintainer of one released *khariif*-adapted hybrid each from the Gujarat and Haryana Agricultural Universities, and of one failed summer-adapted hybrid from Gujarat Agricultural University's MRS Jamnagar).

As the F_1 progenies of the donor x candidate recurrent parent crosses were uniformly heterozygous, we only needed to confirm parentage of 1-3 plants per combination and backcross these to their respective recurrent parent inbreds to produce BC_1F_1 progenies in each of the recurrent parent backgrounds that were segregating 1:1 for donor and recurrent parent alleles at all loci across the genome. Genotyping 7 plants of each BC_1F_1 family to confirm their parentage (and segregation of putatively polymorphic SSR marker alleles at 20 cM intervals across all seven pearl millet linkage groups, and then backcrossing all 7 plants to their respective recurrent parents, theoretically yields sets of BC_2F_1 families for each of the donor parent x recurrent parent populations, with approximately 99% probability of the opportunity to recover donor parent heterozygotes at any position across the full length of the genome. Thus we advanced to the BC_2F_1 seed generation with recurrent parents HMS 7B, ICMB 93333, and ICMB 95222, for both donor parents (ICMB 841-P3 and 863B-P2), before completing QTL mapping of our targets for marker-assisted selection.

We then began marker-assisted foreground selection in earnest using donor parent SSR marker alleles flanking each of the putative stover quality QTLs detected in the mapping study reported by Nepolean *et al.* (2006), and accompanied this with limited background selection among foreground selected BC_2F_1 , BC_3F_1 , and BC_4F_1 individuals in order to identify 2 BC_nF_1 progenies per combination per generation that carried a particular target QTL while recovering the largest extent of recurrent parent homozygosity across the rest of the genome. In the field; all BC_nF_1 plants were both selfed and backcrossed to their respective recurrent parents. Two $BC_nF_2/BC_{n+1}F_1$ progeny from each donor parent QTL x recurrent parent combination were then selected for advance, with $BC_{n+1}F_2$ populations of 23-46 individuals (with the intention of creating near-isogenic pairs homozygous for donor and recurrent parent alleles in the target QTL region) and $BC_{n+1}F_1$ populations of at least 11 individuals per progeny being advanced for genotyping the following crop season.

Activity 3: Recurrent selection, progress in stover fodder quality traits and mode of inheritance of stover fodder quality traits

This piece of work depended on NIRS predictive equations for laboratory fodder traits validated by extensive livestock productivity trials. In 40 pearl millet stover samples fed to sheep, 73, 70, 84 and 82% of the variations in *in vivo* organic matter digestibility, organic matter intake, digestible organic matter intake and nitrogen loss/accretion were accounted for by combinations of stover nitrogen, cell wall fractions, *in vitro* digestibility, *in vitro* metabolizable energy content and morphological measurements such as stem diameter

and sheath fractions. For example, *in vitro* digestibility, nitrogen and acid detergent fibre accounted for 73, 5 and 4% of the partial variation in nitrogen loss/accretion. *In vitro* and chemical parameters were well predicted by NIRS (Table 2).

Table 2: Blind-prediction of chemical and *in vitro* traits in 434 pearl millet stover samples by NIRS.

Variable	R ² (Comparison of wet lab vs. NIRS)	Standard Error of Prediction
Nitrogen (%)	0.935	0.1
NDF (%) Cell wall	0.870	2.3
ADF (%) Cellulose	0.810	2.1
ADL (%) Lignin	0.844	0.5
<i>In vitro</i> digestibility (%)	0.827	2.5
<i>In vitro</i> metabolizable energy (MJ/kg)	0.833	0.4

Amongst 256 full-sib progenies of pearl millet variety ICMV 221 substantial ranges were observed for grain (1.5-fold difference) and stover (1.8-fold difference) yields. Stover protein content varied two-fold, ranging from 4.3 to 8.6% and stover *in vitro* digestibilities varied by more than 5%. Yield of (*in vitro*) digestible stover dry matter ranged from 1132 to 2388 kg ha⁻¹. From these full-sibs, experimental varieties were generated as the base population was subjected to two cycles of full-sib recurrent selection. Experimental varieties produced from the C0 and C1 full-sib progeny sets included a) grain yield, b) dual-purpose use, c) high stover protein; and d) high stover digestibility sub-populations. Stover from experimental varieties and population cycle bulks produced following each of the two selection cycles in 2005 (C0) and 2006 (C1) was tested with sheep for digestibility, intake, digestible organic matter intake and nitrogen balances. Generally observed variation in traits in the experimental varieties were consistent with their intended design. Through full-sib recurrent selection considerable gains were achieved in food-feed traits. For example more than a 17% increase was found in digestible organic matter intake in the dual-purpose experimental variety (15.1 g kg⁻¹ LW d⁻¹) compared to the original variety (12.9 g kg⁻¹ LW d⁻¹). Similarly, nitrogen balance improved from -0.016 g kg⁻¹ LW d⁻¹ in the sheep fed on stover from the original variety to +0.051 g kg⁻¹ LW d⁻¹ in sheep fed stover from the experimental dual-purpose variety.

From the same full-sib of pearl millet variety (ICMV 221) three high and low nitrogen/digestibility full-sib progenies were selected. Crosses were made for high × high (h×h), low × low (l×l) and high × low (h×l) full-sib combinations, and evaluated in the rainy seasons of 2007 and 2008 at Patancheru. The high and low nitrogen full-sib parents contrasted 0.85/0.72% for nitrogen, 41.7/42.3% for *in-vitro* digestibility and 2576/3695 kg

ha⁻¹ for stover dry matter yield, respectively in 2007. The stover nitrogen contents were 0.85, 0.73% and 0.80% for the h_xh, l_xl and h_xl crosses, respectively, in 2007. In 2008, the high and low digestibility full-sib parents contrasted 43.3/40.3% for *in-vitro* digestibility, 0.73/0.85% for nitrogen and 3862/2895 kg ha⁻¹ for stover dry matter yield, respectively. The stover *in-vitro* digestibilities were 43.7%, 40.3% and 42.2% for h_xh, l_xl and h_xl crosses, respectively, in 2008. The h_xh nitrogen cross showed a mean increase of 14% and the h_xh digestibility cross showed a mean increase of 3.3% over the original ICMV 221 base population (N = 0.80%, *in-vitro* digestibility = 41.3%). The intermediate results of h_xl crosses strongly indicate the additive nature of both traits suggesting the application of cyclic breeding methods to improve each of these traits will be successful. Stover digestibility was found to be positively related with stover yield ($r^2=0.9$), suggesting perhaps a positive affect of crop duration on both of these characters, at least in the testing environments used in this study. The selection for improved stover IVOMD can be a very useful approach in breeding to enhance value of crop residues for ruminant livestock nutrition.

This work was taken further by investigating the effects of two A-line testers (ICMA 9511 and ICMA 99022) on crosses of h_xh, h_xl and l_xl digestibility full-sibs high and of h_xh, h_xl and l_xl nitrogen full-sibs in 2007 and 2008. This work confirmed the prevailing role of additive gene action for stover nitrogen and stover *in vitro* digestibility. Testcrossing the h_xh, h_xl and l_xl full-sibs to these two male-sterile lines had no significant effects on stover quality traits, although time to 50% flowering was reduced by an average of 2 days (42.4 vs. 44.3) while panicle yield and stover yields were greater in the testcross hybrids than in their corresponding h_xh, h_xl and l_xl full-sib pollinators.

Activity 4: Develop a series of near-isogenic lines, for individual stover quality QTLs, in the background of the widely adapted and economically important hybrid seed parent maintainer line 841B (= ICMB 841)

This activity was the subject of an on-going Ph.D. thesis study supported by a complementary grant from the Government of India's Department of Biotechnology in its Ministry of Science & Technology. A summary of the substitution lines developed during the course of her thesis research, for six of the seven pearl millet linkage groups, is presented in Appendix 2. This summary is extracted from poster presentations authored by the Ph.D. student, Ms. B. Ramana Kumari, during the first half of 2010 (Ramana Kumari *et al.* 2010a, 2010b).

Activity 5: Produce sets of experimental hybrids using the (i) the original and improved versions of the maintainer lines and restorer populations bred in objective 1, and (ii) the near-isogenic lines for individual stover yield and quality QTLs bred in activity 4, and produce sufficient stover for in vitro and in vivo ruminant nutritional quality assessment

As no statistically significant progress could be demonstrated from two cycles of NIRS-based full-sib recurrent selection targeting improvement of stover yield and nutritional value in the two arid zone-adapted pearl millet restorer populations, we did not produce and evaluate the testcross hybrids of the population cycle bulks as originally planned. Instead, as reported above, we investigated the effects of two male-sterile line testers (ICMA 9511 and ICMA 99022) on crosses of ICMV 221-background h_xh, h_xl and l_xl digestibility full-sibs, and h_xh, h_xl and l_xl nitrogen full-sibs, during the rainy seasons of 2007 and 2008. This work confirmed the prevailing role of additive gene action for stover nitrogen and stover *in vitro* digestibility. Testcrossing the h_xh, h_xl and l_xl full-sibs to these two male-sterile lines had no significant effects on stover quality traits, although time to

50% flowering was reduced by an average of 2 days (42.4 vs. 44.3) while panicle yield and stover yields were greater in the testcross hybrids than in their corresponding h×h, h×l and l×l full-sib pollinators. As stover quality traits were unaffected in the testcrosses, but stover yields were improved in the testcrosses of these h×h, h×l and l×l full-sib pollinators, it is clear that stover digestible dry matter yields were improved in the testcrosses, so one should expect to obtain the highest yields of livestock products by feeding hybrids based on the h×h full-sib pollinators.

For the second half of the first portion of this activity, seed was produced for small-plot yield trials of testcross hybrids of the original and improved versions of the three maintainer line recurrent parents (HMS 7B, ICMB 93333 and ICMB 95222) used on the marker-assisted backcrossing programme. Once homozygous BC₃F₃ and BC₄F₃ near-isogenic pairs for a particular QTL × recurrent parent combination was available (see Activity 2 above), these and their donor and recurrent parents were crossed to relevant testers (RIB 3135-18 in case of recurrent parent ICMB 93333, H 77/29-2 in case of recurrent parent HMS 7B, and HTP 94/54 for recurrent parent ICMB 95222), and replicated observation nurseries (1-2 rows per entry in two replications) or trials (larger plots with 3-4 replications) were grown at ICRISAT-Patancheru (both rainy season and post-rainy season) and project partner testing sites in Gujarat, Haryana and Rajasthan (rainy season only) to identify the most promising entries. Grain and stover yield estimates were obtained from each plot, along with information on flowering date, plant height and a subjective assessment of the degree of recovery of the released hybrid background. From each test site dried stover samples were collected from each plot and sent to ICRISAT-Patancheru for grinding and subsequent NIRS-based assessment of previously validated stover quality attributes (see results from Activity 6 below).

The most promising entries, and appropriate controls (the most closely related released hybrids) were then multiplied on a larger scale (3-5 kg of seed per entry) and sown in replicated large plots (8 rows of 100 m length) at ICRISAT-Patancheru and AICPMIP-Mandore to produce stover samples during the rainy seasons of 2008 and 2009 for *in vivo* feeding trials that were conducted by the ILRI Livestock Nutrition team at ICRISAT-Patancheru.

To summarize, three generations of marker-assisted backcrossing and subsequent selfing of backcrossed progenies having target QTLs was carried out with the help of QTL-flanking microsatellite markers (SSRs). Near-isogenic single-QTL introgression lines that were homozygous for donor parent or recurrent parent alleles in target regions were identified. Improved hybrids were synthesized from these near-isogenic pairs for each target QTL × recurrent parent combination, and were evaluated along with comparable donor parent and recurrent parent hybrids, in multi-locational field trials.

The second portion of this activity is the subject of an on-going Ph.D. thesis study supported by a complementary grant from the Government of India's Department of Biotechnology, Ministry of Science & Technology. As the thesis will be submitted only in March 2011, and the final season of field trials assessing agronomic performance and stover yield and quality of the testcross hybrids of the segmental substitution lines for each linkage group are still underway (harvest of grain and straw samples of the 2010 rainy season trials was initiated late-Sept '10), summaries of the incomplete results are not included in this report. This report will be published on-line prior to submission of the Ph.D. thesis of Ms. B. Ramana Kumari and such prior publication could prejudice acceptance of her thesis by Osmania University authorities.

Activity 6: Determine the improvement made in the nutritional quality of the stover of the experimental hybrids (Activities 2 and 5) and evaluate the specific effects of all individual stover nutritional quality QTLs (individually and in combination) using in vitro, NIRS and in vivo analyses

Results from the laboratory analysis of whole plant straw samples showed that one of the improved hybrids has at least 8.5% higher ME content and 6.3% higher IVOMD compared to the control hybrid. The new hybrid produced a 10% increase in grain yield and 4% increase in stover yield. These results suggest that improved versions of existing hybrids can be developed; concomitantly improving grain and stover traits.

In the particular case referred to in the previous paragraph, field observations at Patancheru during the 2009 rainy season suggested that the improved stover quality had been at least accompanied by (a causal relationship is suggested, but not yet proven) improved host plant resistance to blast disease (caused by *Pyricularia grisea*, the same pathogen responsible for rice blast disease and finger millet blast disease) as a result of introgression of a previously undetected dominant resistance gene from linkage group 4 (LG4) of donor parent 863B (Nepolean *et al.* 2010), which had previously been detected only as a QTL for NIRS-predicted gas yield following *in vitro* digestion of ground stover samples for 24 h (Nepolean *et al.* 2006). The introgression of superior disease resistance from 863B was confirmed by screening potted seedlings of all trial entries, and their parental lines, against the ICRISAT-Patancheru field strain of the pathogen. This demonstrated that elite pollinator HTP 94/54 is resistant to the disease but that its resistance is inherited in a recessive manner, so the original version of released hybrid HHB 146 is susceptible to this disease. In contrast, pollinator RIB 3135-18 has dominantly-inherited resistance to this disease. Thus it was only in the genetic background of the cross of blast-susceptible ICMA/B 95222 × HTP 94/54 (released hybrid HHB 146) that we were able to detect the effect of this major dominant gene for foliar disease resistance from 863B, which appears to contribute to both grain and stover yield, and stover nutritional value under environmental conditions that are favourable for the pathogen. Greenhouse screening of the RIL mapping population based on cross ICMB 841-P3 (susceptible) × 863B-P2 (resistant), followed by QTL mapping using a composite interval mapping algorithm, detected a major QTL for blast resistance at a map position on LG4 (Nepolean *et al.* 2010), which coincides with that for “gas24” detected in the earlier F₂:F₄ testcross study (Nepolean *et al.* 2006), with the 863B allele(s) for increased gas generation being associated with 863B alleles for blast resistance. A poster describing this finding was presented at a national symposium at ANG Ranga Agricultural University in Feb 2010 (Nepolean *et al.* 2010) and was awarded 3rd prize. In an earlier publication, arising from the predecessor of the current project, we had indicated that marker-assisted selection for enhanced host plant resistance to foliar diseases would very likely contribute to enhanced stover nutritional value (Hash *et al.*, 2003).

We are now ready to begin on-farm assessment of this improved version of released pearl millet hybrid HHB 146 (ICMA 95222 × HTP 94/54), and are seeking cooperation from CCS Haryana Agricultural University authorities to do so, beginning in the rainy season of 2011. This should be accompanied by two years of testing in state and national trials, after which the improved hybrid would be eligible for release (with a 1-year shorter assessment programme, as it would be an “essentially-derived” cultivar based on a previously released cultivar). We have identified an appropriate livestock nutritionist to work with us, based at C.C.S. Haryana Agricultural University’s Regional Research Station Bawal. He has recently expressed interest in having ICRISAT produce up to 100 kg of seed of each of the original and improved versions of pearl millet hybrid HHB 146 for on-farm assessment (perhaps under the umbrella of the Bill & Melinda Gate Foundation-supported “HOPE” project, which is making extensive use of on-farm assessment of supposedly improved pearl millet hybrids in Gujarat, Haryana and Rajasthan during a 4-year initial phase (2009–2013).

Using testcross hybrids of a (ICMB 841 × 863B)-derived F₂:F₄ mapping population, stover quality QTLs contributing to the improvement of metabolizable energy (ME) and *in-vitro* organic matter digestibility (IVOMD) were mapped to linkage groups 2, 4 and 5. These QTLs were then introgressed into four parental lines of released dual-purpose hybrids having good agronomic performance. Three generations of marker-assisted backcrossing and two subsequent generations of selfing of individuals having target QTLs was carried out with selection based on QTL-flanking SSRs. Single-QTL introgression lines homozygous for QTL target regions were identified. Experimental hybrids were synthesized from these QTL-introgression homozygotes and their parents, and were evaluated in multilocal field trials. Results from the laboratory analysis of whole plant stover samples and animal feeding trials showed that one of the experimental hybrids had 8.5% better ME and IVOMD and 2% better digestible organic matter intake than its near-isogenic commercially cultivated control hybrid. During field evaluation it was noted that improved blast resistance from donor parent 863B was incorporated into the same hybrid, suggesting that resistance to blast was contributing to improved straw quality. These results suggest that development of improved hybrids with better stover quality is possible without compromising grain and stover yield traits. Small-scale (using straw samples from replicated 6 x 100 m plots of each hybrid) *in vivo* analysis (using sets of 6 sheep per straw sample) of the stover quality of this experimental hybrid and its near-isogenic blast susceptible released hybrid counterpart (HHB 146 = ICMA 95222 × HTP 94/54) were inconclusive, as no statistically significant improvement was detected in most of the observed measures of livestock performance.

Activity 7: Evaluate the quality of currently marketed pearl millet hybrids, and use the results to raise awareness of fodder quality issues with the public and private seed industry

Eight popular commercial hybrids of pearl millet were evaluated for stover fodder quality traits and their potential trade-off with stover and grain yield. The stover quality traits chosen and analyzed were nitrogen, *in vitro* digestibility and metabolizable energy content. Highly significant ($P < 0.01$) variations were observed for grain yields (2860 to 4220 kg/ha), stover yields (range 3760 to 4930 kg/ha), stover nitrogen (0.62 to 1.10%), stover *in vitro* digestibility (37.6 to 46.7%) and stover metabolizable energy (5.26 to 6.88 MJ/kg). Stover nitrogen content was negatively associated with grain and stover yield but no such trade-offs were observed between stover *in vitro* digestibility and metabolizable energy contents on the one hand and grain and stover yield on the other.

Hybrid GK 1044 had the lowest stover nitrogen content (0.62%), and highest stover digestibility (46.7%) and metabolizable energy content (6.88 MJ/kg); while MLBH 267 with the highest stover nitrogen content (1.10%) had the lowest stover digestibility (37.6%) and metabolizable energy content (5.26 MJ/kg). These two contrasting hybrids were re-sown on large plots the following year and their stover tested *in vivo* with sheep as a sole feed. Digestible organic matter intake was significantly higher in GK 1044 than in MBLH 267 (13.5 *versus* 12.5 grams per kg live weight) and nitrogen balance tended ($P < 0.10$) to be more favourable in GK 1044 than in MBLH 267 (−0.008 *versus* −0.10 grams per kg live weight). These results show that among commercial high-yielding pearl millet hybrids, some can be found with high grain and stover yield, combined with high stover digestibility and metabolizable energy content. Observations from the feeding trial suggested that stover *in vitro* digestibility and metabolizable energy are more important than stover nitrogen content in determining stover quality, which is in agreement with the extensive testing of experimental varieties and hybrids undertaken in this project.

In collaboration with the private seed company that produced and marketed pearl millet hybrid GK 1044, the food-feed performance of this hybrid was tested across several states in India. While the seed company could explain the high digestibility/metabolizable

energy content of GK 1044 by tracing the parentage of its pollinator line to sweet pearl millet genotypes, they observed poor stover performance of this hybrid in northern India because of its susceptibility to the foliar disease blast, caused by the fungal pathogen *Pyricularia grisea*. We will continue working with the breeding team of this seed company to better understand this problem, and to outline a crossing programme to improve the blast resistance of either or both of the parental lines of GK 1044 in order to maintain the high food-feed-value of this hybrid while eliminating negative traits that prevent its wider cultivation. Indeed, it is likely that marker-assisted backcross introgression of the dominantly inherited blast resistance from linkage group 4 of 863B (Nepolean *et al.* 2010), into the genetic background of the seed parents (A/B-pair) of GK 1044 could provide a reasonable and cost-effective short-term solution to this problem. An even shorter-term solution would be to produce and evaluate the hybrid produced by crossing the pollinator parent of GK 1044 with the improved version of ICMA/B 95222, as ICMB 95222 has in its parentage a sister line of the seed parent maintainer line (bred by ICRISAT) of GK 1044.

It was also agreed with the private seed enterprises to extend such pearl millet stover analyses (quantity and quality) to >30 popular hybrids grown at three or more different locations in India during the 2010 rainy season.

Generally this work with public- and private-sector pearl millet breeding programmes has substantially raised the level of awareness about genetic variation in food-feed traits in pearl millet, and has probably contributed to recent decisions amending cultivar release criteria to include stover traits among factors to be considered while making decisions for cultivar release.

Activity 8: Encourage the public and private seed producers to use the improved parental lines by providing them information on quality characteristics and animal performance, experimental quantities of seed, and analytical assistance.

We have made the improved versions of parental lines of released pearl millet hybrids available to both public- and private-sector plant breeders for their evaluation under the terms and conditions of ICRISAT's Revised Material Transfer Agreement for products of our public-supported plant breeding research, which itself is an annex to the Standard Material Transfer Agreement for the exchange of Plant Genetic Resources for Food and Agriculture (see Appendix 1); encouraged these plant breeders to participate in the multi-environment trials of stover yield and quality of experimental hybrids made with these parents; and shared with them all data and results summaries obtained from these trials. In future we will publish results of this work in both the scientific and popular literature, and continue to make appropriate presentations at national and international scientific meetings.

Although the project has ultimately been successful in producing experimental hybrids with significantly improved NIRS-predicted stover quality, we are only now in a position to multiply seed of experimental hybrids and provide technical assistance to public and private sector organizations (at their cost) to conduct participatory evaluations with farmers to assess the productivity of these hybrids and of animals fed on their stover. Originally, these on-farm assessments were to have been conducted in the final two years of the project, with support of these activities by the technicians funded by the project, under the supervision of Drs. Hash and Blümmel. However, we now need to identify funding that will permit us to bring the valuable findings of this project to Indian pearl millet-producing farmers *via* on-farm trials.

8 Impacts

8.1 Scientific impacts – now and in 5 years

Strategies to promote use of project outputs

The primary outputs of this project were new and/or modified hybrid pearl millet parent lines that produce stover with a higher nutritive value for ruminants. The potential benefits of these new parent lines will be made available to farmers by the public and private hybrid seed industry, with which ICRISAT has a close relationship. ICRISAT is the major supplier of both early generation and finished parental breeding materials to the public and private pearl millet seed sector and makes this material freely available. The value of this relationship is formally recognized by the provision of operational funding to ICRISAT by a consortium of the larger companies, and by the formal incorporation of activities of this project into the agreed annual work plans of the ICAR-ICRISAT collaborative project on marker-assisted breeding of pearl millet for the period 2006-2009. As outlined in Activities 7 & 8, we built upon these relationships, and will continue to exploit these to improve the likelihood of adoption of the new versions parent lines by breeding programs and the seed industry, and ultimately improved versions of currently popular pearl millet hybrids by crop-livestock producers across this project's target region in Gujarat, Haryana, and Rajasthan.

The strategy employed in this project was to:

- involve individual seed company and state agricultural university breeders at an early stage in collaboration with the major public-sector pearl millet research coordinating body in India (AICPMIP);
- build awareness of the importance of stover quality;
- communicate throughout the conduct of the project; provision of seed of parent lines and promising hybrid lines; and
- provide technical support for farmer evaluation trials

However, we were not able to reach this final phase of the project due to:

- delays in completing the marker-assisted backcrossing and subsequent identification of superior QTL introgression testcrosses warranting on-farm evaluation, and,
- failure of NIRS-based full-sib progeny recurrent selection to improve stover value (yield and/or nutritional quality) of two topcross pollinator populations adapted to the arid zone of central and western Rajasthan).

Details of the implementation of these strategies are provided above in Activities 3, 7 & 8, but some additional information is provided here.

At the outset of this project the seed industry was consulted to select appropriate maintainer lines in which to incorporate stover quality traits (the four lines recommended to us were HMS 7A/B, ICMB 93333, ICMB 95111, and ICMB 95222), and on their preferences for testers to use to make experiential hybrids (RIB 3135-18 for ICMA/B 93333, HTP 94/54 or J 2372 for ICMA/B 95222, and H 77/29-2 for HMS 7A/B) to evaluate the improved maintainers. During the five and a half years taken in implementing this project, we kept the pearl millet breeding programmes of private seed companies and state agricultural university project partners informed of work in progress and of results obtained in improving stover quality through written reports, presentations during field days and annual national pearl millet group meetings, and discussions with visiting company and state agricultural university scientists and managers. These covered topics

such as the QTLs identified for different stover quality traits, status reports on the progress in incorporating these into elite maintainer lines, and the likely benefits of these for animal producers relying on millet stover as their main source of roughage.

During the two year years, we started to encourage public- and private-sector pearl millet breeders to evaluate these new versions of elite hybrid parental lines by producing and evaluating their own experimental hybrids with the improved maintainer lines (B-lines) and their own pollinators (R-lines). Over the coming 2-3 years we will continue to do this and distribute fully converted male-sterile A-lines corresponding to the best available improved B-lines.

They also were encouraged to participate in the multi-environment trial program assessing the stover quality of the experimental hybrids, and through that involvement they will have had access to all data and results obtained from these trials, including the detailed stover quality analyses of trials conducted at their own locations, and the *in vivo* analyses done at ICRISAT-Patancheru by the ILRI Livestock Nutrition team based there.

The timeliness provision of the stover quality analysis results was a constraint, so a summary of these results will be produced over the coming 4 months, submitted in February 2011 for inclusion in the 2010/11 Annual Report of the All-India Coordinated Pearl Millet Improvement Project (AICPMIP), and presented at the annual pearl millet group meeting in March/April 2011.

We are now seeking funding to support seed multiplication and large-scale on-farm testing of the best improved versions of previously released pearl millet hybrids in the target zone of Gujarat, Haryana and Rajasthan – perhaps as part of the Bill & Melinda Gates Foundation-supported “HOPE” project.

We are also seeking funding from the Indian Council for Agricultural Research (ICAR) to support establishment of a pearl millet marker-assisted breeding network via a 5-year programme that would

- bring all major public-sector pearl millet breeding programs in India “up to speed” on marker-assisted backcrossing for maintenance breeding and value addition in elite hybrid parental lines and provide the operational resources required for contracting appropriate service laboratories to generate the necessary marker data in a cost-effective and timely manner;
- introgress at least 6 QTLs (in some instances including the pearl millet foliar disease resistance and stover quality QTLs that have been validated in this ACIAR-supported pearl millet stover project) into the genetic backgrounds of at least 25 different elite hybrid parental lines of released hybrids adapted to one or more of the four major pearl millet production environments in India;
- produce and evaluate testcross hybrids of the single- and double-QTL introgression lines in the genetic background of each of the 25 recurrent parents (developed in the course of the first 3 years of the proposed 5-year project) in 2-year multi-locational on-station field trials that are now required prior to release in India of essentially derived products based on previously released cultivars; and
- pyramiding of the single- and double-QTL introgressions in each recurrent parent genetic background to provide multiple-QTL introgression lines to be used
 - in another round of testcross hybrid evaluation, and
 - as recurrent parents for future maintenance and value-addition breeding.

In future we will also utilize linkages with selected NGOs working in the arid areas where conventional single-cross hybrids are not widely used by farmers, but where millet/livestock is the major farming system (for example BAIF, Seva Mandir, and the Urmal Trust). As the project has been not been successful in producing experimental topcross hybrids with significantly improved stover quality, we will instead furnish seed of

the best available single-cross hybrids for this region and assist these organizations to evaluate the hybrids with farmers.

We have started to present the results of this research at professional meetings in India (e.g., Nepolean *et al.* 2009, 2010; Ramana Kumari *et al.* 2010a,) and internationally (Ramana Kumari *et al.* 2010b), and are drafting journal articles to be published in regional and international refereed journals, in targeted newsletters such as open-access SAT eJournal that ICRISAT publishes, incorporating the International Sorghum and Millets Newsletter (which was previously distributed to over 1500 sorghum and millet scientists worldwide), and in the popular agricultural literature in India. These publications will continue to be complemented by presentations at national meetings (including the annual meeting of the All-India Coordinated Pearl Millet Improvement Project) at which we can provide public- and private-sector breeding programmes with status reports on progress in the research project, including the availability of hybrid parental lines with improved livestock feeding value, and on recent advances in the number and genome coverage provided by mapped genomic- and genic-SSR markers (Rajaram *et al.* 2010a, 2010b), STS markers (PCR-compatible derivatives of previously mapped RFLP markers; Gale *et al.* 2001), SSCP-SNP and CISP markers (Bertin *et al.* 2005; Feltus *et al.* 2006, Thudi *et al.* 2010), DArT markers (Kilian *et al.* 2009; Supriya *et al.* 2010), and integrated genetic linkage maps (Senthilvel *et al.* 2010) for pearl millet.

8.2 Capacity impacts – now and in 5 years

The project provided opportunities for training of 16 Visiting Scientists from collaborating Indian Universities, most of them through the All-India Coordinated Pearl Millet Improvement Project. The scientist individually spent from three months to two years actively working on the project at ICRISAT-Patancheru, during which time they were provided on-the-job training in stover quality assessment, QTL mapping, and marker-assisted backcrossing. Marker-assisted breeding has been receiving considerable attention in India, especially in the major staple cereals (particularly rice and wheat) and cash crops (cotton and oilseed mustard), so policy makers here are now aware of the potential of the technology and have been taking steps to ensure that adequate research infrastructure is in place to support applied public-sector research.

One result of such increased emphasis of India's policy makers on marker-assisted breeding during the course of this ACIAR project is that ICRISAT's M.S. Swaminathan Applied Genomics Laboratory at Patancheru was awarded a 4-year project by the Ministry of Science & Technology's Department of Biotechnology (DBT) to establish a Centre of Excellence in Genomics (CEG). That DBT-supported project has permitted ICRISAT to substantially strengthen opportunities for impact of this ACIAR-supported project by

- developing a Genotyping Service Laboratory that provides SSR marker data for crop, livestock and microbial researchers (including ICRISAT scientists) in India on a full-cost, non-profit basis; and,
- conducting a 2-3 week short-course on molecular marker technology and its application in plant breeding, twice each year (2006-2010), for 20-30 participants from public- and private-sector plant breeding programs in India. Normally 2-4 of the participants in each of these "CEG marker-assisted breeding courses" have been pearl millet breeders, with another 3-5 of the participants being sorghum breeders, and we have typically be able to offer such scientists a further 1-2 weeks of training in applications of the technology in ICRISAT's sorghum and pearl millet marker-assisted backcrossing programmes. Private company staff interested in taking the course have been permitted to join on a payment basis, as have public-sector researchers from ICRISAT's target environments outside India.

Such opportunities for training at established labs such as those at ICRISAT-Patancheru are highly valued by young Indian scientists, particularly when they are long enough (3 weeks or more) to count towards their promotion.

The existence of CEG's Genotyping Service Laboratory has meant that it is no longer necessary for ICRISAT, national program, or private company plant breeders to expend a great deal of time and effort in generating the marker data, purchasing supplies for this purpose, and maintaining expensive equipment that rapidly becomes obsolete as the most cost-effective technologies for marker data generation evolve. Instead, the plant breeding teams need only concern themselves with the biological questions that they are attempting to address with these modern tools: planning their marker-assisted breeding experiments (for diversity assessment, linkage map development, QTL mapping, marker-assisted selection, and/or fingerprinting for establishment of the unique identity of new breeding products for the purposes of plant variety protection), collecting DNA or tissue samples from the required numbers of plants or progenies to ensure a high probability of success in their marker-assisted breeding objectives and providing these to the Genotyping Service Laboratory, keeping good quality records so that sample identities are maintained, performing the best possible job of phenotyping sets of genetic stocks or breeding products, and then interpreting the marker data sets received from the Service Laboratory and using the results in their basic, strategic and applied research programmes. Because of this, and because of the substantially larger numbers of young pearl millet breeders that we have been able to provide with an introduction to marker-assisted breeding techniques, there is now the required critical mass of interested pearl millet scientists to apply these methods for marker-assisted improvement of pearl millet disease resistance, drought tolerance, grain yield and micronutrient content, and stover yield and quality – once they have access to the required operational resources (and a concept note was recently submitted to the Indian Council of Agricultural Research seeking funding for establishment of a Pearl Millet Marker-Assisted Breeding Network that would ensure the required operational resources for key public-sector pearl millet breeding programmes throughout the country).

As this project has been largely successful, it has added the improvement of stover quality to ICRISAT's own millet breeding program capabilities (not only in India, but also in West Africa) and, through this, to the research programs of public-sector research organizations as well as major seed companies who see such value-addition as a market advantage for their proprietary hybrids. ICRISAT has been assisting such companies in acquiring or contracting out the capability to both pursue marker-assisted selection to improve host plant resistance to major diseases, so it will now be a small step further to helping them use these tools to enhance stover quality components.

We expect that within three years it will be possible to complete the testing required for currently available marker-assisted breeding products having stover with enhanced ruminant nutritional value to be officially released (as essentially-derived versions of previously released hybrids) and enter the pearl millet hybrid seed multiplication chain. Only once seed of such value-added versions of currently grown hybrids becomes widely available to farmers will it be possible for project AS2/1999/062 to have significant economic impacts at the farm level.

8.3 Community impacts – now and in 5 years

We do expect most such impact to occur in the economic and social spheres. However, we feel that the project outputs will also contribute to better integration of crop-livestock production activities through improved and more sustainable feed resources. Improved feed resources have previously been shown to alleviate pressure on natural resources such as common property and forest lands, and also to reduce drudgery for women and children by reducing the need for foraging, herding and fodder collection.

8.3.1 Economic impacts

An *ex-ante* assessment of returns to research to improve the yield and digestibility of sorghum and pearl millet crop residues in appropriate climatic domains (Kristjanson and Zerbini 1999) indicated a net present value of US\$42-208M, with predicted internal rates of return of 28-43%, and benefit-cost ratios of 15-69:1. Disaggregated figures for pearl millet are not readily available from that study, but it is reasonable to assume that pearl millet research would provide at least one-third of these total benefits. This will be less than that from sorghum because pearl millet stover yield and ruminant nutritional quality are both lower than for sorghum, which is typically grown in more favourable environments. However, benefits would accrue more rapidly from pearl millet research because of the higher level of adoption of hybrid cultivars for pearl millet. Since the recommendation domain for work proposed in this proposal constitutes 2/3rds of the pearl millet area in that earlier *ex-ante* study (*i.e.* about 6M ha), the net present value in India of the research proposed can be estimated at about US\$10M.

Assumptions in the *ex-ante* study included a 10% level of adoption of improved pearl millet cultivars in the target region within 10 years of the initiation of the research program. We considered these to be conservative figures, given that this project has provided improved versions of seed parent pair 841A/B that can immediately be exploited to produce improved versions of currently popular pearl millet hybrids at no extra cost to seed producers or smallholder farmers, and similarly improved versions of ICMA/B 95222 that can be exploited in a similar manner. We anticipate a 50% replacement of 841A-based hybrids within 5 years of the first commercialization of hybrids based on the new version of 841A/B (with such commercialization to begin approximately 3 years from now). This time frame for initial adoption will depend on how recent changes in Indian seed legislation are implemented, but should require no more than three years from release of the new parent lines if a crop variety registration system replaces the current truthful label system (that would require only two years for initial adoption). Spill-overs to non-target countries are not likely in the short term, however, as India is the only major producer of dual-purpose pearl millet cultivars where hybrids are produced.

Thus we expect that within three years it will be possible to complete the testing required for currently available marker-assisted breeding products having stover with enhanced ruminant nutritional value to be officially released (as essentially-derived versions of previously released hybrids) and enter the pearl millet hybrid seed multiplication chain. Only once seed of such value-added versions of currently grown hybrids becomes widely available to farmers will it be possible for project AS2/1999/062 to have significant economic impacts at the farm level.

If recent experience with marker-assisted breeding product “HHB 67 Improved” provides a reasonable basis for expectations of the rate and extent of adoption of the marker-assisted breeding products from for project AS2/1999/062 can be expected to take 2-3 years following their entry into the pearl millet hybrid seed multiplication chain before they are widely adopted as replacements for the original version of currently popular hybrid cultivars. The rate of adoption of subsequent generations of marker-assisted breeding products (in which improved stover yield and/or quality is combined with improved host plant resistance to downy mildew and tolerance to drought, salinity or micronutrient deficiency), can be expected to be higher as the seed industry and farmers gain experience with the products of marker-assisted QTL introgression breeding programmes.

In the case of India’s first pearl millet marker-assisted breeding product “HHB 67 Improved”, which was notified for release by the Government of India in late 2005, too late for significant quantities of F1 hybrid seed to be multiplied for sale during the 2006 rainy season, the seed industry and farmers have now largely adopted the new version of the hybrid as a higher-yielding and more disease resistant version of their now disease-susceptible old friend “HHB 67”, and the value of the new version to Indian farmers is estimated conservatively to exceed US\$10 million per year due to losses avoided as a result of its improved downy mildew resistance. The actual increased value to Indian

farmers from cultivation of this single hybrid could exceed US\$30 million annually from grain and stover production on at least 500,000 hectares. In the case of hybrids with improved stover quality – these will reach market if they exhibit clearly superior resistance to foliar diseases, which farmers recognize as dramatically reducing stover feeding value. Assuming that we pyramid major genes for blast resistance (such as that identified as the likely underlying cause of the linkage group 4 stover quality QTL with favourable alleles from 863B) with those for blast and downy mildew resistance, then farmers will be able to easily see the advantages of the improved hybrids and will clamour for seed – providing the demand necessary to get public-sector seed agencies such as the National Seeds Corporation, the Rajasthan State Seeds Corporation, and the Gujcomasol marketing federation in Gujarat to take up multiplication of these improved hybrids in a big way. Of course, private seed companies willing to take the risk to multiply and market these products before substantial demand is in place, will have opportunities to capture increased market share and so will find it attractive to enter the market with such products earlier than the public-sector seed agencies.

8.3.2 Social impacts

This project specifically targeted improvement of the productivity and economic value of a major source of cash income of small crop-livestock producers in marginal agricultural environments of one of the world's poorest regions. The targeted beneficiaries of project outputs were the low- and medium-income farm families growing pearl millet in mixed crop-livestock production systems of north-western India. We do not expect any significant barriers to the ability of these target groups to access and utilize the new genetic materials or to share in the potential benefits that could accrue to the target communities if this project is successful. These communities have already largely adopted pearl millet hybrid cultivars (even in the margins of the arid zone, where "HHB 67 Improved" is now demanded by more farmers than the public-sector seed distribution system can effectively serve – providing a market opportunity for small- and medium-sized seed companies interested in meeting this demand), and prior PRA studies (Underwood *et al.* 2000) with these communities indicate their appreciation of differences in the nutritional quality of the stover of different millet genotypes, their keen interest in having higher yields of more nutritious stover for their livestock, and the economic benefits they will receive from higher value stover.

8.3.3 Environmental impacts

Adoption of outputs from this project are not expected to entail any environmental risks and are unlikely to have any significant positive or negative environmental impacts. As the changes resulting from the project are likely to be in the form of improved versions of existing cultivars, there will be little or no impact on either the social or physical environment. However, there exists concern about trade-off effects between use of stover (more generally biomass) for livestock feeding and soil improvement through, for example, Conservation Agriculture. On the bright side, multi-dimensional crop improvement as explored in the current project will contribute to mitigate such trade-offs; for example, by breeding and selecting cultivars with not only high grain yield but also high stover yield. In addition, there are some core areas where improved dual-purpose pearl millet cultivars can have a substantial favourable environmental impact. For example, the Gujarat dairy sector is very important economically, but is very inefficient in terms of water use. In Gujarat, the production of 1 litre of milk currently requires an average of 3,400 litres of water, with the vast majority of this water used to irrigate forage crops such as alfalfa. Improving pearl millet stover feed value can substantially reduce this water requirement, by reducing dairy requirements for less water-use-efficient leguminous forage crops.

In India, the use of RFLP-based molecular marker technology (in which the RFLP probe sequences are maintained as plasmid inserts in transgenic bacteria) as a selection tool in plant breeding falls under the biosafety regulations of the Ministry of Environment's

Department of Biotechnology. ICRISAT is one of >100 government-recognized research centres having the necessary infrastructure (including an Institutional BioSafety Committee) to allow safe use of the relevant technologies. The use of PCR-based marker techniques such as sequence-tagged microsatellite (STMS) markers is effectively unregulated in India as it is not considered to pose any potential biosafety hazards. There was complete compliance with the relevant government regulations at ICRISAT-Patancheru during the research phase of this project, and during the course of this project ICRISAT's pearl millet marker-assisted breeding activities stopped the use of RFLP markers as sufficient numbers of sequence-tagged site (STS), simple sequence repeat (SSR), single nucleotide polymorphism (SNP), and Diversity Array Technology™ (DArT™) markers became available to replace RFLP markers across the entire length of the mapped pearl millet nuclear genome.

8.4 Communication and dissemination activities

This project generated several key-findings relating to: 1) breeding methods and 2) improved whole plant utilization resulting in a proposed change in paradigm in pearl millet improvement. Both findings were aggressively disseminated using scientific papers and books, crop and livestock conferences, the annual group meeting of the All-India pearl millet research team (attended by research scientists and research managers from both the public-sector and the private-sector), and the ICRISAT hybrid seed consortium. The pearl millet project contributed also to the ILRI CGIAR Outcome Story (which was about Indian crop and animal scientists deciding to join forces in changing crop improvement approaches towards whole plant optimization); the CGIAR Science Council rated this outcome as the highest among all IARCs, achieving a score of 9.4 out of 10. In addition a special issue on Food-Feed Crops will be published (proofs have been corrected) by the journal *Animal Nutrition and Feed Technology*, essentially devoted to further disseminate core messages of this project. Along similar lines, the United Nations Food and Agriculture Organization (FAO) approached us for a think piece on further institutionalizing and up-scaling of food-feed crop work.

9 Conclusions and recommendations

9.1 Conclusions

The aim of this project was to improve overall productivity in crop-livestock systems of India by increasing the nutritive value of pearl millet stover. Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is the only reliably productive cereal in rainfed areas of north-western India, where pearl millet stover accounts for over 30% of fodder resources available, so genetic improvement of pearl millet stover yield and quality is considered the only practical way to improve the productivity and economic returns of this rainfed crop-livestock production system without substantial public and/or private investments in irrigation and other infrastructure. This project was a follow-up to the Small Restricted Grant AS2/97/98: *Development and use of molecular genetic markers for enhancing the feeding value of cereal crop residues and pastures for ruminants*. Until this project, only limited attention had been paid by cereal breeders in India to either the quantity or quality of stover, despite the high value placed on this component of the crop by farmers in crop-livestock systems. The project attempted to redress this problem, by focusing on genetically improving the yield and quality of crop stover, while maintaining grain yield and other important agronomic traits of adapted, farmer-accepted dual-purpose crop cultivars.

The first project objective was to produce pearl millet hybrid parent lines with enhanced stover quality suitable for use in commercial hybrid seed production, by first identifying flanking markers for additional quantitative trait loci (QTLs) for stover quality traits known to predict animal performance with a high degree of accuracy, then transferring these to several selected hybrid seed parent maintainer lines by marker-assisted backcrossing. The first two activity areas within this objective were largely successful. The third activity area, using NIRS-based full-sib recurrent selection to improve stover quality in restorer populations adapted to the arid zone of central and western Rajasthan was not successful, either because full-sib field trials in the target region were vitiated by field variation in this marginal production environment, or because the base populations used lacked sufficient heritable variation for stover yield and quality traits. However, proof of concept of this activity was demonstrated by successful use of the same recurrent selection protocol in improving stover yield and quality of composite variety ICMV 221 (based on 124 S1 progenies from the broad-based ICRISAT Bold-Seeded Early Composite).

The second project objective was to determine the effects of individual stover quality QTLs, and combinations, in enhanced commercial hybrid parents, on *in vitro* stover quality and *in vivo* animal production. The first two activities in this area were to complete development and testcross hybrid evaluation of a series of near-isogenic chromosome segment substitution lines involving elite *Iniari* landrace-based donor parent 863B and genetically distant elite dual-purpose hybrid seed parent maintainer line ICMB 841. This took longer than expected, but will result in a Ph.D. thesis (supported with complementary funding from Indian government sources) describing the production (first activity) and successful testcross evaluation (second activity) of substitution lines for 6 of the 7 pearl millet chromosome pairs. As the final season's field trials for these "contig line" testcross studies were harvested as this report was being finalized, and data analysis of only 2 of the 4 season's trials were complete, description of the results of these two activities are limited here to presentation of graphical genotypes of the sets of segmental substitution lines produced. In addition, testcross hybrids of the original and improved versions of the maintainer lines bred in the first objective were assessed for grain and stover yield, and *in*

in vitro stover quality, with sufficient stover being produced of selected experimental and control entry pairs to permit *in vivo* ruminant nutritional quality assessment. As a result, a previously undetected major dominant foliar disease resistance gene was shown to be associated with a major QTL for *in vitro* digestibility estimates in buffered rumen fluid; however, no *in vivo* improvement in livestock performance could be detected.

Several of the testcross hybrids of the “improved” seed parent maintainer lines that had been developed by marker-assisted backcross introgression of favorable alleles for stover quality traits from donor parents ICMB 841 or 863B demonstrated significant changes (compared to their near-isogenic released counterparts) in flowering date, plant height, grain and stover yield components, grain and stover yield, and/or various *in vitro* or NIRS-predicted measures of stover quality. Many of these significant changes in agronomic traits were unfavorable; however, several promising stover quality QTL introgression lines were developed, not least of which was a BC₄F₃ line based recurrent parent ICMB 95222, with a linkage group 4 introgression from donor parent 863B-P2 that includes a major dominant gene for host plant resistance to blast disease. Compared to its released near-isogenic counterpart hybrid HHB 146, the testcross of this introgression line demonstrated improvement in NIRS-predicted stover quality as well as at least marginal improvements in grain yield, 1000-grain mass, grain number per panicle, and panicle harvest index (Nepolean *et al.* 2010).

The third and final objective of this project was to strengthen awareness of the importance of stover nutritive value and to similarly strengthen national program human resources in India. This was fully successful, resulting in important policy changes in varietal release criteria.

The five key results of this project were as follows:

- 1) improvement in hybrid performance (at least for NIRS-predicted stover quality traits) as reported by Nepolean *et al.* (2010);
- 2) progress in 2 cycles of full-sib recurrent selection for stover yield and quality traits (based on NIRS-predicted stover quality traits) as reported by Bidinger *et al.* (2010a, 2010b) in the genetic background of widely-cultivated pearl millet composite ICMV 221 (released and cultivated in Eritrea, Ethiopia, India, Kenya, Mexico and Uganda);
- 3) demonstration of additive to partially dominant mode of inheritance for stover nitrogen content and stover *in vitro* digestibility in ICMV 221 (Bidinger *et al.* 2010a, 2010b), and that dominant host plant resistance to an important foliar disease caused by the fungal pathogen *Pyricularia grisea* is associated with NIRS-predicted improvement of stover quality in the genetic background of released pearl millet hybrid HHB 146 (Nepolean *et al.* 2010);
- 4) characterization of the considerable genetic variation in food feed traits that is present among widely cultivated commercial pearl millet hybrids; and
- 5) substantial national programme human resource development in the form of short-term (<1 month), medium-term (>1 month to 3 months), and long-term (up to 2 years) Visiting Scientist training in aspects of marker-assisted backcrossing and/or stover quality assessment for 16 Indian scientists (Appendix 3).

9.2 Recommendations

Although this project has ultimately been successful in producing experimental pearl millet hybrids with significantly improved stover quality, we are only now in a position to multiply seed of the best of these experimental hybrids and provide technical assistance to public-

and private-sector organizations (at their cost) to conduct participatory evaluations with farmers assessing the productivity of these hybrids and of animals fed on their stover. Originally, these on-farm assessments were to have been conducted in the final two years of the project, with support of these activities by the technicians funded by the project, under the supervision of Drs. Hash and Blümmel. However, we now need to identify funding that will permit us to bring the delayed but never-the-less valuable findings of this project to Indian pearl millet-producing farmers via participatory on-farm trials.

In a wider context, the project has shown that significant variation exists in stover quantity and quality among cultivars of pearl millet, and we propose that this variation should be considered in breeding, selection and release of new cultivars by public- and private-sector cereal breeding programs globally and it is clear from parallel studies that such variation exists in elite genetic backgrounds in other cereals as well. The current project has contributed to the creation of a NIRS platform where national institutes can phenotype their cultivars for fodder related traits. There is a high probability that this can and will happen because 1) the national system has already undertaken the 1st step by considering stover quantity in new cultivar release decisions (and as this project has clearly demonstrated that, at least in pearl millet, stover quantity and quality are positively associated); and 2) new pearl millet improvement as envisaged and outlined in the new ICRISAT-led CRP(3) on Dryland Cereals, in which improved stover fodder quality is a major target trait.

Finally, substantial effort should now be made to completing pending data analyses (such as that associated with the Ph.D. thesis research programme of Ms. B. Ramana Kumari) and preparation, submission and publication of journal article manuscripts summarizing the most pertinent results from this generally successful research project so that they can be more effectively and more widely disseminated.

10References

10.1 References cited in report

- Bertin, I., J.H. Zhu, and M.D. Gale. 2005. SSCP-SNP in pearl millet—a new marker system for comparative genetics. *Theoretical and Applied Genetics* 110: 1467–1472.
- Bidinger, F.R., O.P. Yadav, and M.M. Sharma. 2002. Male-sterile parents for breeding landrace-based topcross hybrids of pearl millet for arid conditions. I. Productivity, responsiveness and stability. *Indian Journal of Genetics and Plant Breeding* 62: 121–127.
- Blummel, M., and F.R. Bidinger. 2007. Management and cultivar effects on ruminant nutritional quality of pearl millet (*Pennisetum glaucum* (L.) R. Br.) stover. II Effects of genotype choice on stover quality and productivity. *Field Crops Research* 130: 129–138.
- Blümmel, M., E. Zerbini, B.V.S. Reddy, C.T. Hash, F. Bidinger and R. Devulapalli. 2003a. Improving the production and utilization of sorghum and pearl millet as livestock feed: methodological problems and possible solutions. *Field Crops Research* 84: 123–142.
- Blümmel M., E. Zerbini, B.V.S. Reddy, C.T. Hash, F. Bidinger, and A. Khan. 2003b. Improving the production and utilization of sorghum and pearl millet as livestock feed: progress towards dual-purpose genotypes. *Field Crops Research* 84: 143–158.
- Gale, M.D., K.M. Devos, J.H. Zhu, S. Allouis, M.S. Couchman, H. Liu, T.S. Pittaway, X.Q. Qi, M. Kolesnikova-Allen and C.T. Hash. 2001. New molecular marker technologies for pearl millet improvement. *International Sorghum and Millets Newsletter* 42: 17–22.
- Feltus, F.A., H.P. Singh, H.C. Lohithaswa, S.R. Schulze, T.D. Silva, and A.H. Paterson. 2006. A comparative genomics strategy for targeted discovery of single-nucleotide polymorphisms and conserved-noncoding sequences in orphan crops. *Plant Physiology* 140: 1183–1191.
- Govila, O.P., K.N. Rai, K.R. Chopra, D.J. Andrews, and W.D. Stegmeier. 1997. Breeding pearl millet hybrids for developing countries: Indian experience. Pages 97–118 *in* Proceedings of the International Conference in Genetic Improvement of Sorghum and Pearl Millet, 23-27 Sep 1996, Lubbock, Texas. INTSORMIL Publication no. 97-5. USAID Title XII Collaborative Research Support Program on Sorghum and Pearl Millet (INTSORMIL): Lincoln, Nebraska, USA.
- Hash, C.T., Jr. 1994. Current status and strategy for promoting hybrid sorghum and pearl millet technology. Pages 46–60 *in* Hybrid Research and Development Needs in Major Cereals in the Asia-Pacific Region (Paroda, R.S. and Mangala Rai, eds.). Bangkok, Thailand: Food and Agriculture Organization of the United Nations, Regional Office for Asia and the Pacific.
- Hash, C.T., M.D. Abdu Rahman, A.G. Bhasker Raj, and E. Zerbini. 2001. Molecular markers for improving nutritional quality of crop residues for ruminants. Pages 203–217 *in* Molecular Breeding of Forage Crops. Proceedings of the 2nd International Symposium, Molecular Breeding of Forage Crops, Lorne and Hamilton, Victoria, Australia November 19-24, 2000 (G. Spangenberg, ed.). Dordrecht, The Netherlands: Kluwer Academic Publishers.
- Hash, C.T., A.G. Bhasker Raj, S. Lindup, A. Sharma, C.R. Beniwal, R.T. Folkertsma, V. Mahalakshmi, E. Zerbini, and M. Blümmel. 2003. Opportunities for marker-assisted

- selection (MAS) to improve the feed quality of crop residues in pearl millet and sorghum. *Field Crops Research* 84: 79–88.
- Kelly T.G., P. Parthasarathy Rao, E. Weltzien-R., and M.L. Purohit. 1996. Adoption of improved cultivars of pearl millet in an arid environment, straw quality and quality considerations in Western Rajasthan. *Experimental Agriculture* 32: 161–171.
- Kristjansson, P.M., and E. Zerbini. 1999. Genetic enhancement of sorghum and millet residues fed to ruminants. ILRI Impact Assessment Series 3, ILRI Nairobi.
- Nepolean, T., C.T. Hash, M. Blümmel, R.P. Thakur, R. Sharma, C.J. Dangaria, H.P. Yadav, B.S. Rajpurohit, and I.S. Khairwal. 2010. P-118: Marker-assisted backcrossing (MABC) to improve pearl millet stover quality traits simultaneously improves blast resistance. Page 162 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Nepolean, T., M. Blümmel, and C.T. Hash. 2009. Improving straw quality traits through QTL mapping and marker-assisted selection in pearl millet. *Forage Symposium 2009 - Emerging Trends in Forage Research and Livestock Production*, February 16-17th 2009. Page 12.
- Rajaram, V., R.K. Varshney, V. Vadez, T. Nepolean, S. Senthilvel, J. Kholová, S. Choudhary, Supriya, S. Kumar, R.P. Thakur, R. Sharma, V. Pandurangarao, K.N. Raj, G. Velu, K.L. Sahrawat, A.G. Bhasker Raj, M. Blümmel, M. Lakshmi Narasu, M. Kocová, P.B. Kavi Kishor, R.C. Yadav, Govind Singh, and C.T. Hash. 2010a. Development of EST resources in pearl millet and their use in development and mapping of EST-SSRs in four RIL populations. P373. Poster presented at PAG–XVIII, 9-13 Jan 2010, San Diego, CA, USA.
- Rajaram, V., R.K. Varshney, V. Vadez, T. Nepolean, S. Senthilvel, Supriya A., S. Kumar, M. Lakshmi Narasu, R.C. Yadav, Govind Singh, and C.T. Hash. 2010b. P-122: Mapping EST-SSRs, developed from ESTs of stressed root and leaf tissues, in four pearl millet RIL populations. Page 166 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Rama Devi, K., R. Bandyopadhyay, A.J. Hall, S. Indira, S. Pande, and P. Jaiswal. 2000. Farmers' Perception of the Effects of Plant Diseases on the Nutritive Value of Crop Residues used for Peri-Urban Dairy Production on the Deccan Plateau: Findings from Participatory Rural Appraisals. *Information Bulletin No. 60*. International Crops Research Institute for the Semi-Arid Tropics, Patancheru 502 324.
- Ramana Kumari, B., M.A. Kolesnikova-Allen, S. Senthilvel, T. Nepolean, P.B. Kavi Kishor, J.R. Witcombe, and C.T. Hash. 2010a. P-124: Development of contiguous segment substitution lines (CSSLs) in pearl millet [*Pennisetum glaucum* (L.) R. Br.]. Page 168 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Ramana Kumari, B., M. Kolesnikova-Allen, S. Senthilvel, T. Nepolean, V. Vadez, A.G. Bhasker Raj, P.B. Kavi Kishor, J.R. Witcombe, and C.T. Hash. 2010b. Development and agronomic evaluation of contiguous segment substitution lines (CSSLs) in pearl millet [*Pennisetum glaucum* (L.) R. Br.]. Poster abstract P-249. 2nd International

- Symposium on Genomics of Plant Genetic Resources. 24-27 April 2010, Bologna, Italy.
- Renard, C. (ed.). 1997. *Crop Residues in Sustainable Mixed Crop/Livestock Farming Systems*. CAB International, Wallingford, UK.
- Romney, D. 2003. *Consultancy Report India for DfID-Project: Enhancing livelihoods of poor livestock keepers through increasing use of fodder*. ILRI, Nairobi, Consultancy Report.
- Senthilvel, S., Nepolean T., Supriya A., Eshwar K., Rajaram V., S. Kumar, T. Hash, A. Kilian, R.C. Yadav, L.M. Narasu, and Govind Singh. 2010. Development of a molecular linkage map of pearl millet integrating DArT and SSR markers. P368. Poster presented at PAG–XVIII, 9-13 Jan 2010, San Diego, CA, USA.
- Supriya, S., Senthilvel, T. Nepolean, C. Tom Hash, V. Rajaram, K. Eshwar, Rajan Sharma, R.P. Thakur, V. Panduranga Rao, and R.C. Yadav. 2010. P-126: Identification of quantitative trait loci associated with resistance to rust in pearl millet using a DArT- and SSR-based linkage map. Page 170 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Talukdar B.S., I.S. Khairwal, and Rattan Singh. 1999. Hybrid breeding. Pages 269–301 in *Pearl Millet Breeding* (I.S. Khairwal, K.N. Rai, D.J. Andrews, and G. Harinarayana, eds.). Oxford & IBH: New Delhi.
- Thudi, M., S. Senthilvel, A. Bottley, C.T. Hash, A.R. Reddy, A.F. Feltus, A.H. Paterson, D.A. Hoisington, and R.K. Varshney. 2010. A comparative assessment of the utility of PCR-based marker systems in pearl millet. *Euphytica* 174: 253–260. DOI 10.1007/s10681-010-0148-5
- Underwood M.P., A.J. Hall, and E. Zerbin. 2000. Genetic enhancement of sorghum and millet residues fed to ruminants: Farmers' perception of fodder quality in livelihood systems. Summary report of PRA case studies in Andhra Pradesh, Gujerat (*sic*), Maharashtra, Karnataka and Rajasthan states. ILRI (International Livestock Research Institute), Nairobi, Kenya and ICRISAT (International Crop Research Institute for the Semi-Arid Tropics), Patancheru, India. 68 pp.
- Witcombe, J.R., M.N.V.R. Rao, A.G.B. Raj, and C.T. Hash. 1997. Registration of 'ICMV 88904' pearl millet. *Crop Science* 37:1022–1023.

10.2 List of publications produced by project (thus far)

- Bidinger, F.R., M. Blümmel, and C.T. Hash. 2006. Response to recurrent selection for stover feeding value in pearl millet variety ICMV 221. *International Sorghum and Millets Newsletter* 47: 113–116.
- Bidinger F. R. (the Late), M. Blümmel, C.T. Hash and S. Choudhary. 2010a. Genetic enhancement for superior food-feed traits in a pearl millet (*Pennisetum glaucum* (L.) R. Br.) variety by recurrent selection. *Animal Nutrition and Feed Technology* (Special Issue Food-Feed-Crops): 49–56.
- Bidinger, F.R. (the Late), S. Choudhary, C.T. Hash, V. Vadez, Rekha B., and M. Blümmel. 2010b. Gene action governing pearl millet stover nitrogen and digestibility and their improvement. National symposium on "Genomics and Crop Improvement: Relevance and Reservations, 25-27 Feb 2010, ANGRAU, Hyderabad, India. Abstract book page 157.

- Blümmel, M., A.A. Khan, V. Vadez, C.T. Hash, and K.N. Rai. 2010. Variability in stover quality traits in commercial hybrids of pearl millet (*Pennisetum glaucum* (L.) R. Br.) Animal Nutrition and Feed Technology (Special Issue Food-Feed-Crops): 18–26.
- Blümmel, M., N. Seetharama, K.V.S.V. Prasad, Ch. Ramakrishna, A.A. Khan, S. Anandan, C.T. Hash, Belum Reddy, S. Nigam, and V. Vadez. 2009. Food-feed crop research and multidimensional crop improvement in India. Pages 17–19 in Proceedings of Animal Nutrition Association World Conference, 14-17 Feb 2009, New Delhi, India. Volume I, Lead Papers.
- Hash, C.T. 2010. Opportunities for rapid step-wise improvement of elite pure-line varieties and hybrid parental lines by marker-assisted backcrossing : Examples from sorghum and pearl millet. Pp. 6-7 in: Book of Abstracts. Indo-US Bilateral Workshop: Plant Genomics in Crop Improvement with Reference to Biotic and Abiotic Stresses. Feb 25-27, 2010, Department of Biotechnology and Molecular Biology, CCS Haryana Agricultural University, Hisar - 125 004, India in collaboration with College of Agriculture and Natural Resources, Michigan State University, East Lansing, USA.
- Hash, C.T. 2009. Application of molecular markers by ICRISAT in pearl millet and sorghum improvement. 2009 International Conference on Agricultural Biotech Frontiers, National Chiayi University, Taiwan, 30-31 October 2009. Page 18.
- Hash, C.T. 2007. Recent developments in sorghum and pearl millet related to marker-assisted selection (MAS). Pages 18–19 in: Abstracts: National Symposium on Plant Molecular Biology : Perspectives, Nov 16-17, 2007, Centre for Plant Molecular Biology, Osmania University, Hyderabad 500007, AP, India.
- Hash, C.T. 2005. Opportunities for application of molecular markers for sustainable crop production in stress environments: Sorghum and pearl millet. Page 13 in Abstracts, International Conference on Sustainable Crop Production in Stress Environments: Management and Genetic Options, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur (M.P.), India, February 9-12, 2005.
- Hash, C.T. and S. Senthilvel. 2008. DNA markers and marker-assisted selection -- Application of theory. Pages 180–201 in Sorghum Improvement in the New Millennium (Reddy, B.V.S., S. Ramesh, A. Ashok Kumar and C.L.L. Gowda, eds.). Patancheru 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics (ICRISAT).
- Hash, C.T., M. Blümmel, and F.R. Bidinger. 2006. Genotype x environment interactions in food-feed traits in pearl millet cultivars. International Sorghum and Millets Newsletter 47: 153–157.
- Hash, C.T., V. Rajaram, R.K. Varshney, T. Nepolean, S. Senthilvel, V. Vadez, I.S. Khairwal, O.P. Yadav, P.C. Gupta and M. Blümmel. 2008. Strengthening pearl millet genomics tools. Poster abstract 2.5, GCP Annual Research Meeting, 16-20 Sep 2008, Bangkok, Thailand.
- Hash, C.T., A. Sharma, T. Nepolean, and S. Senthilvel. 2006. Development and application of tools for marker-assisted breeding in pearl millet. Plant & Animal Genome XIV, Jan 14-18 2006, Town & Country Hotel, San Diego, CA, USA. Final Abstracts Guide. W66. <http://www.intl-pag.org/14/abstracts/>
- Kilian, Andrzej, Ling Xia, Peter Wenzl, Eric Huttner, David Jordan, Emma Mace, Sophie Bouchet, Jean-Francois Rami, Jean-Christophe Glaszmann, Rajeev Varshney, Tom Hash, and S Senthilvel 2009. DArT genome profiling and dedicated information technologies for modern breeding of sorghum and pearl millet. W468. Abstract of oral presentation at PAG–XVII, 10-14 Jan 2009, San Diego, CA, USA.
- Nepolean, T., M. Blümmel, A.G. Bhasker Raj, V. Rajaram, S. Senthilvel and C.T. Hash. 2006. QTLs controlling yield and stover quality traits in pearl millet. International Sorghum and Millets Newsletter 47: 149–152.

- Nepolean, T., C.T. Hash, M. Blümmel, R.P. Thakur, R. Sharma, C.J. Dangaria, H.P. Yadav, B.S. Rajpurohit, and I.S. Khairwal. 2010. P-118: Marker-assisted backcrossing (MABC) to improve pearl millet stover quality traits simultaneously improves blast resistance. Page 162 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Nepolean, T., M. Blümmel, and C.T. Hash. 2009 Improving straw quality traits through QTL mapping and marker-assisted selection in pearl millet. Forage Symposium 2009 - Emerging Trends in Forage Research and Livestock Production, February 16-17th 2009. Page 12.
- Nepolean, T., M. Blümmel, V. Rajaram, S. Senthilvel, R.S. Yadav, C.J. Howarth, K.M. Devos, S. Sivaramkrishnan, E. Zerbini, and C.T. Hash. 2005. QTLs controlling stover quality traits in pearl millet (*Pennisetum glaucum* L. R. Br.) and opportunities for marker assisted selection. Page 194 *in* Abstracts, International Conference on "Plant genomics and biotechnology: Challenges & opportunities", October 26-28, Indira Gandhi Agricultural University, Raipur, Chhattisgarh, India.
- Prasad, K.V.S.V., D. Ravi, F. Bidinger, C.T. Hash, and M. Blümmel. 2006. Relations between laboratory traits of 14 pearl millet stover and their digestibility, intake and nitrogen balance in sheep. Proceedings XIIth Animal Nutrition Conference in Technological Interventions in Animal Nutrition for Rural Prosperity, 7th-9th Jan. 2006, College of Veterinary Science and Animal Husbandry, Anand Agricultural University, Anand - 388 110, Gujrat (*sic*), India. FFR 25.
- Ramana Kumari, B., M.A. Kolesnikova-Allen, S. Senthilvel, T. Nepolean, P.B. Kavi Kishor, J.R. Witcombe, and C.T. Hash. 2010a. P-124: Development of contiguous segment substitution lines (CSSLs) in pearl millet [*Pennisetum glaucum* (L.) R. Br.]. Page 168 *in*: Abstracts: National Symposium on Genomics and Crop Improvement: Relevance and Reservations. Feb 25-27, 2010, Acharya N.G. Ranga Agricultural University, Indian Council of Agricultural Research, and Department of Biotechnology. Institute of Biotechnology, Acharya N.G. Ranga Agricultural University, Rajendranagar, Hyderabad 500 030.
- Ramana Kumari, B., M. Kolesnikova-Allen, S. Senthilvel, T. Nepolean, V. Vadez, A.G. Bhasker Raj, P.B. Kavi Kishor, J.R. Witcombe, and C.T. Hash. 2010b. Development and agronomic evaluation of contiguous segment substitution lines (CSSLs) in pearl millet [*Pennisetum glaucum* (L.) R. Br.]. Poster abstract P-249. 2nd International Symposium on Genomics of Plant Genetic Resources. 24-27 April 2010, Bologna, Italy.
- Ravi D., S. Anandan, A.A. Khan, F.R. Bidinger, T. Nepolean, C.T. Hash, and M. Blümmel. 2010. Morphological, chemical and *in vitro* traits for prediction of stover quality in pearl millet for use in multidimensional crop improvement. Animal Nutrition and Feed Technology (Special Issue Food-Feed-Crops): 37–48.
- Vellaikumar, S., A. Khan, F. Bidinger, C.T. Hash, and M. Blümmel. 2006. Variations in fodder value of pearl millet stover when fed to sheep. Proceedings XIIth Animal Nutrition Conference in Technological Interventions in Animal Nutrition for Rural Prosperity, 7th-9th Jan. 2006, College of Veterinary Science and Animal Husbandry, Anand Agricultural University, Anand - 388 110, Gujrat (*sic*), India. FFR 27.

11 Appendixes

11.1 Appendix 1:

STANDARD MATERIAL TRANSFER AGREEMENT

**(Applicable for all exchange of “Plant Genetic Resources for Food and Agriculture”
involving CG Centres globally)**

and

Revised Material Transfer Agreement for ICRISAT-bred materials

Yellow-highlighted sections in the two proformas are to be completed by the Recipient

STANDARD MATERIAL TRANSFER AGREEMENT*

PREAMBLE

WHEREAS

The International Treaty on Plant Genetic Resources for Food and Agriculture (hereinafter referred to as "the Treaty")¹ was adopted by the Thirty-first session of the FAO Conference on 3 November 2001 and entered into force on 29 June 2004;

The objectives of the Treaty are the conservation and sustainable use of **Plant Genetic Resources for Food and Agriculture** and the fair and equitable sharing of the benefits arising out of their use, in harmony with the Convention on Biological Diversity, for sustainable agriculture and food security;

The Contracting Parties to the Treaty, in the exercise of their sovereign rights over their **Plant Genetic Resources for Food and Agriculture**, have established a **Multilateral System** both to facilitate access to **Plant Genetic Resources for Food and Agriculture** and to share, in a fair and equitable way, the benefits arising from the utilization of these resources, on a complementary and mutually reinforcing basis;

Articles 4, 11, 12.4 and 12.5 of the Treaty are borne in mind;

The diversity of the legal systems of the Contracting Parties with respect to their national procedural rules governing access to courts and to arbitration, and the obligations arising from international and regional conventions applicable to these procedural rules, are recognized;

Article 12.4 of the Treaty provides that facilitated access under the **Multilateral System** shall be provided pursuant to a Standard Material Transfer Agreement, and the **Governing Body** of the Treaty, in its Resolution 1/2006 of 16 June 2006, adopted the Standard Material Transfer Agreement.

¹ Note by the Secretariat: as suggested by the Legal Working Group during the Contact Group for the Drafting of the Standard Material Transfer Agreement, defined terms have, for clarity, been put in bold throughout.

* In the event that the SMTA is used for the transfer of Plant Genetic Resources for Food and Agriculture other than those listed in Annex 1 of the Treaty:

The references in the SMTA to the "Multilateral System" shall not be interpreted as limiting the application of the SMTA to Annex 1 Plant Genetic Resources for Food and Agriculture, and in the case of Article 6.2 of the SMTA shall mean "under this Agreement";

The reference in Article 6.11 and Annex 3 of the SMTA to "Plant Genetic Resources for Food and Agriculture belonging to the same crop, as set out in Annex 1 to the Treaty" shall be taken to mean "Plant Genetic Resources for Food and Agriculture belonging to the same crop".

ARTICLE 1 — PARTIES TO THE AGREEMENT

1.1 The present Material Transfer Agreement (hereinafter referred to as “**this Agreement**”) is the Standard Material Transfer Agreement referred to in Article 12.4 of the **Treaty**.

1.2 **This Agreement** is:

BETWEEN:

Name : Dr CLL Gowda
Designation: Global Theme Leader-Crop improvement
Institution : ICRISAT (International Crops Research Institution for the Semi-Arid Tropics)
Address : ICRISAT-Patancheru P.O., Hyderabad 502 324, Andhra Pradesh, India

(hereinafter referred to as “the **Provider**”),

AND:

Name :
Designation:
Institution :
Address :

(hereinafter referred to as “the **Recipient**”).

1.3 The parties to **this Agreement** hereby agree as follows:

ARTICLE 2 — DEFINITIONS

In **this Agreement** the expressions set out below shall have the following meaning:

“**Available without restriction**”: a **Product** is considered to be available without restriction to others for further research and breeding when it is available for research and breeding without any legal or contractual obligations, or technological restrictions, that would preclude using it in the manner specified in the **Treaty**.

“**Genetic material**” means any material of plant origin, including reproductive and vegetative propagating material, containing functional units of heredity.

“**Governing Body**” means the **Governing Body** of the **Treaty**.

“**Multilateral System**” means the **Multilateral System** established under Article 10.2 of the **Treaty**.

“**Plant Genetic Resources for Food and Agriculture**” means any **genetic material** of plant origin of actual or potential value for food and agriculture.

“**Plant Genetic Resources for Food and Agriculture under Development**” means material derived from the **Material**, and hence distinct from it, that is not yet ready for **commercialization** and which the developer intends to further develop or to transfer to another person or entity for further development. The period of development for the **Plant Genetic Resources for Food and Agriculture under Development** shall be deemed to have ceased when those resources are **commercialized** as a **Product**.

“**Product**” means **Plant Genetic Resources for Food and Agriculture** that incorporate¹ the **Material** or any of its genetic parts or components thereof that are ready for **commercialization**, excluding commodities and other products used for food, feed and processing.

“**Sales**” means the gross income resulting from the **commercialization** of a **Product** or **Products**, by the **Recipient**, its affiliates, contractors, licensees and lessees.

“**To commercialize**” means to sell a **Product** or **Products** for monetary consideration on the open market, and “**commercialization**” has a corresponding meaning.

Commercialization shall not include any form of transfer of **Plant Genetic Resources for Food and Agriculture under Development**.

ARTICLE 3 — SUBJECT MATTER OF THE MATERIAL TRANSFER AGREEMENT

The **Plant Genetic Resources for Food and Agriculture** specified in *Annex 1* to **this Agreement** (hereinafter referred to as the “**Material**”) and the available related information referred to in Article 5b and in *Annex 1* are hereby transferred from the **Provider** to the **Recipient** subject to the terms and conditions set out in **this Agreement**.

ARTICLE 4 — GENERAL PROVISIONS

4.1 **This Agreement** is entered into within the framework of the **Multilateral System** and shall be implemented and interpreted in accordance with the objectives and provisions of the **Treaty**.

4.2 The parties recognize that they are subject to the applicable legal measures and procedures, that have been adopted by the Contracting Parties to the **Treaty**, in conformity with the **Treaty**, in particular those taken in conformity with Articles 4, 12.2 and 12.5 of the **Treaty**.²

4.3 The parties to **this Agreement** agree that (*the entity designated by the **Governing Body***),³ acting on behalf of the **Governing Body** of the **Treaty** and its **Multilateral System**, is the third party beneficiary under **this Agreement**.

4.4 The third party beneficiary has the right to request the appropriate information as required in Articles 5e, 6.5c, 8.3 and *Annex, 2 paragraph 3*, to **this Agreement**.

4.5 The rights granted to the (*the entity designated by the **Governing Body***) above do not prevent the **Provider** and the **Recipient** from exercising their rights under **this Agreement**.

¹ As evidenced, for example, by pedigree or notation of gene insertion.

² *In the case of the International Agricultural Research Centres of the Consultative Group on International Agricultural Research (CGIAR) and other international institutions, the Agreement between the Governing Body and the CGIAR Centres and other relevant institutions will be applicable.*

³ Note by the Secretariat: *by Resolution 2/2006, the Governing Body “invite[d] the Food and Agriculture Organization of the United Nations, as the Third Party Beneficiary, to carry out the roles and responsibilities as identified and prescribed in the Standard Material Transfer Agreement, under the direction of the Governing Body, in accordance with the procedures to be established by the Governing Body at its next session”. Upon acceptance by the FAO of this invitation, the term, “the entity designated by the Governing Body”, will be replaced throughout the document by the term, “the Food and Agriculture Organization of the United Nations”.*

ARTICLE 5 — RIGHTS AND OBLIGATIONS OF THE PROVIDER

The **Provider** undertakes that the **Material** is transferred in accordance with the following provisions of the **Treaty**:

- a) Access shall be accorded expeditiously, without the need to track individual accessions and free of charge, or, when a fee is charged, it shall not exceed the minimal cost involved;
- b) All available passport data and, subject to applicable law, any other associated available non-confidential descriptive information, shall be made available with the **Plant Genetic Resources for Food and Agriculture** provided;
- c) Access to **Plant Genetic Resources for Food and Agriculture under Development**, including material being developed by farmers, shall be at the discretion of its developer, during the period of its development;
- d) Access to **Plant Genetic Resources for Food and Agriculture** protected by intellectual and other property rights shall be consistent with relevant international agreements, and with relevant national laws;
- e) The **Provider** shall periodically inform the **Governing Body** about the Material Transfer Agreements entered into, according to a schedule to be established by the **Governing Body**. This information shall be made available by the **Governing Body** to the third party beneficiary.⁴

ARTICLE 6 — RIGHTS AND OBLIGATIONS OF THE RECIPIENT

6.1 The Recipient undertakes that the Material shall be used or conserved only for the purposes of research, breeding and training for food and agriculture. Such purposes shall not include chemical, pharmaceutical and/or other non-food/feed industrial uses.

6.2 The Recipient shall not claim any intellectual property or other rights that limit the facilitated access to the **Material** provided under **this Agreement**, or its genetic parts or components, in the form received from the **Multilateral System**.

6.3 In the case that the Recipient conserves the **Material** supplied, the Recipient shall make the **Material**, and the related information referred to in Article 5b, available to the **Multilateral System** using the Standard Material Transfer Agreement.

6.4 In the case that the Recipient transfers the **Material** supplied under **this Agreement** to another person or entity (hereinafter referred to as “the **subsequent recipient**”), the Recipient shall

- a) do so under the terms and conditions of the Standard Material Transfer Agreement, through a new material transfer agreement; and
- b) notify the **Governing Body**, in accordance with Article 5e.

⁴ Note by the Secretariat: *The Standard Material Transfer Agreement makes provision for information to be provided to the **Governing Body**, in the following Articles: 5e, 6.4b, 6.5c and 6.11h, as well as in Annex 2, paragraph 3, Annex 3, paragraph 4, and in Annex 4. Such information should be submitted to:*

On compliance with the above, the Recipient shall have no further obligations regarding the actions of the subsequent recipient.

6.5 In the case that the **Recipient** transfers a **Plant Genetic Resource for Food and Agriculture under Development** to another person or entity, the **Recipient** shall:

- a) do so under the terms and conditions of the Standard Material Transfer Agreement, through a new material transfer agreement, provided that Article 5a of the Standard Material Transfer Agreement shall not apply;
- b) identify, in *Annex 1* to the new material transfer agreement, the **Material** received from the **Multilateral System**, and specify that the **Plant Genetic Resources for Food and Agriculture under Development** being transferred are derived from the **Material**;
- c) notify the **Governing Body**, in accordance with Article 5e; and
- d) have no further obligations regarding the actions of any **subsequent recipient**.

6.6 Entering into a material transfer agreement under paragraph 6.5 shall be without prejudice to the right of the parties to attach additional conditions, relating to further product development, including, as appropriate, the payment of monetary consideration.

6.7 In the case that the **Recipient commercializes** a **Product** that is a **Plant Genetic Resource for Food and Agriculture** and that incorporates **Material** as referred to in Article 3 of **this Agreement**, and where such **Product** is not **available without restriction** to others for further research and breeding, the **Recipient** shall pay a fixed percentage of the **Sales** of the **commercialized Product** into the mechanism established by the **Governing Body** for this purpose, in accordance with *Annex 2* to **this Agreement**.

6.8 In the case that the **Recipient commercializes** a **Product** that is a **Plant Genetic Resource for Food and Agriculture** and that incorporates **Material** as referred to in Article 3 of **this Agreement** and where that **Product** is **available without restriction** to others for further research and breeding, the **Recipient** is encouraged to make voluntary payments into the mechanism established by the **Governing Body** for this purpose in accordance with *Annex 2* to **this Agreement**.

6.9 The **Recipient** shall make available to the **Multilateral System**, through the information system provided for in Article 17 of the **Treaty**, all non-confidential information that results from research and development carried out on the **Material**, and is encouraged to share through the **Multilateral System** non-monetary benefits expressly identified in Article 13.2 of the **Treaty** that result from such research and development. After the expiry or abandonment of the protection period of an intellectual property right on a **Product** that incorporates the **Material**, the **Recipient** is encouraged to place a sample of this **Product** into a collection that is part of the **Multilateral System**, for research and breeding.

6.10 A **Recipient** who obtains intellectual property rights on any **Products** developed from the **Material** or its components, obtained from the **Multilateral System**, and assigns such intellectual property rights to a third party, shall transfer the benefit-sharing obligations of **this Agreement** to that third party.

6.11 The Recipient may opt as per Annex 4, as an alternative to payments under Article 6.7, for the following system of payments:

- a) The Recipient shall make payments at a discounted rate during the period of validity of the option;**
- b) The period of validity of the option shall be ten years renewable in accordance with Annex 3 to this Agreement;**
- c) The payments shall be based on the Sales of any Products and of the sales of any other products that are Plant Genetic Resources for Food and Agriculture belonging to the same crop, as set out in Annex 1 to the Treaty, to which the Material referred to in Annex 1 to this Agreement belongs;**
- d) The payments to be made are independent of whether or not the Product is available without restriction;**
- e) The rates of payment and other terms and conditions applicable to this option, including the discounted rates are set out in Annex 3 to this Agreement;**
- f) The Recipient shall be relieved of any obligation to make payments under Article 6.7 of this Agreement or any previous or subsequent Standard Material Transfer Agreements entered into in respect of the same crop;**
- g) After the end of the period of validity of this option the Recipient shall make payments on any Products that incorporate Material received during the period in which this Article was in force, and where such Products are not available without restriction. These payments will be calculated at the same rate as in paragraph (a) above;**
- h) The Recipient shall notify the Governing Body that he has opted for this modality of payment. If no notification is provided the alternative modality of payment specified in Article 6.7 will apply.**

ARTICLE 7 — APPLICABLE LAW

The applicable law shall be General Principles of Law, including the UNIDROIT Principles of International Commercial Contracts 2004, the objectives and the relevant provisions of the **Treaty**, and, when necessary for interpretation, the decisions of the **Governing Body**.

ARTICLE 8 — DISPUTE SETTLEMENT

8.1 Dispute settlement may be initiated by the **Provider** or the **Recipient** or the (*the entity designated by the **Governing Body***), acting on behalf of the **Governing Body** of the **Treaty** and its **Multilateral System**.

8.2 The parties to **this Agreement** agree that the (*the entity designated by the **Governing Body***), representing the **Governing Body** and the **Multilateral System**, has the right, as a third party beneficiary, to initiate dispute settlement procedures regarding rights and obligations of the **Provider** and the **Recipient** under **this Agreement**.

8.3 The third party beneficiary has the right to request that the appropriate information, including samples as necessary, be made available by the **Provider** and the **Recipient**, regarding their obligations in the context of **this Agreement**. Any information or samples so requested shall be provided by the **Provider** and the **Recipient**, as the case may be.

8.4 Any dispute arising from this Agreement shall be resolved in the following manner:

- a) **Amicable dispute settlement: The parties shall attempt in good faith to resolve the dispute by negotiation.**
- b) Mediation: If the dispute is not resolved by negotiation, the parties may choose mediation through a neutral third party mediator, to be mutually agreed.
- c) Arbitration: If the dispute has not been settled by negotiation or mediation, any party may submit the dispute for arbitration under the Arbitration Rules of an international body as agreed by the parties to the dispute. Failing such agreement, the dispute shall be finally settled under the Rules of Arbitration of the International Chamber of Commerce, by one or more arbitrators appointed in accordance with the said Rules. Either party to the dispute may, if it so chooses, appoint its arbitrator from such list of experts as the Governing Body may establish for this purpose; both parties, or the arbitrators appointed by them, may agree to appoint a sole arbitrator, or presiding arbitrator as the case may be, from such list of experts. The result of such arbitration shall be binding.

ARTICLE 9 — ADDITIONAL ITEMS

Warranty

9.1 The **Provider** makes no warranties as to the safety of or title to the **Material**, nor as to the accuracy or correctness of any passport or other data provided with the **Material**. Neither does it make any warranties as to the quality, viability, or purity (genetic or mechanical) of the **Material** being furnished. The phytosanitary condition of the **Material** is warranted only as described in any attached phytosanitary certificate. The **Recipient** assumes full responsibility for complying with the recipient nation's quarantine and biosafety regulations and rules as to import or release of **genetic material**.

Duration of Agreement

9.2 **This Agreement** shall remain in force so long as the **Treaty** remains in force.

ARTICLE 10 — SIGNATURE/ACCEPTANCE

The **Provider** and the **Recipient** may choose the method of acceptance unless either party requires **this Agreement** to be signed.

Option 1 –Signature*

I, (Dr CLL Gowda), represent and warrant that I have the authority to execute **this Agreement** on behalf of the **Provider** and acknowledge my institution's responsibility and obligation to abide by the provisions of **this Agreement**, both by letter and in principle, in order to promote the conservation and sustainable use of **Plant Genetic Resources for Food and Agriculture**.

Signature.......... Date.....

Name of the **Provider** : Dr C L Laxmipathi Gowda

I, (), represent and warrant that I have the authority to execute **this Agreement** on behalf of the **Recipient** and acknowledge my institution's responsibility and obligation to abide by the provisions of **this Agreement**, both by letter and in principle, in order to promote the conservation and sustainable use of **Plant Genetic Resources for Food and Agriculture**.

Signature..... Date.....

Name of the **Recipient**.....

Option 2 – Shrink-wrap Standard Material Transfer Agreements*

The **Material** is provided conditional on acceptance of the terms of **this Agreement**. The provision of the **Material** by the **Provider** and the **Recipient's** acceptance and use of the **Material** constitutes acceptance of the terms of **this Agreement**.

Option 3 – Click-wrap Standard Material Transfer Agreement*

- I hereby agree to the above conditions.

* Where the **Provider** chooses signature, only the wording in Option 1 will appear in the Standard Material Transfer Agreement. Similarly where the **Provider** chooses either shrink-wrap or click-wrap, only the wording in Option 2 or Option 3, as appropriate, will appear in the Standard Material Transfer Agreement. Where the "click-wrap" form is chosen, the **Material** should also be accompanied by a written copy of the Standard Material Transfer Agreement.

Annex 1

LIST OF MATERIALS PROVIDED

This *Annex* contains a list of the **Material** provided under **this Agreement**, including the associated information referred to in Article 5b.

This information is either provided below or can be obtained at the following website: (*URL*).

The following information is included for each **Material** listed: all available passport data and, subject to applicable law, any other associated, available, non-confidential descriptive information.

(*List*)

Annex 2

RATE AND MODALITIES OF PAYMENT UNDER ARTICLE 6.7 OF THIS AGREEMENT

1. If a **Recipient**, its affiliates, contractors, licensees, and lessees, **commercializes a Product or Products**, then the **Recipient** shall pay one point-one percent (1.1 %) of the **Sales** of the **Product or Products** less thirty percent (30%); except that no payment shall be due on any **Product or Products** that:
 - (a) are **available without restriction** to others for further research and breeding in accordance with Article 2 of **this Agreement**;
 - (b) have been purchased or otherwise obtained from another person or entity who either has already made payment on the **Product or Products** or is exempt from the obligation to make payment pursuant to subparagraph (a) above;
 - (c) are sold or traded as a commodity.
2. Where a **Product** contains a **Plant Genetic Resource for Food and Agriculture** accessed from the **Multilateral System** under two or more material transfer agreements based on the Standard Material Transfer Agreement only one payment shall be required under paragraph 1 above.
3. The **Recipient** shall submit to the **Governing Body**, within sixty (60) days after each calendar year ending December 31st, an annual report setting forth:
 - (a) the **Sales** of the **Product or Products** by the **Recipient**, its affiliates, contractors, licensees and lessees, for the twelve (12) month period ending on December 31st;
 - (b) the amount of the payment due; and
 - (c) information that allows for the identification of any restrictions that have given rise to the benefit-sharing payment.
4. Payment shall be due and payable upon submission of each annual report. All payments due to the **Governing Body** shall be payable in (US\$)⁵ for the account of (*the Trust Account or other mechanism established by the **Governing Body** in accordance with Article 19.3f of the **Treaty***).⁶

⁵ Note by the Secretariat: *The Governing Body has not yet considered the question of currency of payment. Until it does so, Standard Material Transfer Agreements should specify United States dollars (US\$).*

⁶ Note by the Secretariat: *This is the Trust Account provided for in Article 6.3 of the Financial Rules, as approved by the Governing Body (Appendix E to this Report). The details of the Trust Account when established, will be introduced here, and communicated to Contract Parties.*

Annex 3

TERMS AND CONDITIONS OF THE ALTERNATIVE PAYMENTS SCHEME UNDER ARTICLE 6.11 OF THIS AGREEMENT

1. The discounted rate for payments made under **Article 6.11** shall be zero point five percent (0.5 %) of the **Sales** of any **Products** and of the sales of any other products that are **Plant Genetic Resources for Food and Agriculture** belonging to the same crop, as set out in **Annex 1 to the Treaty**, to which the **Material** referred to in **Annex 1 to this Agreement** belong.
2. **Payment** shall be made in accordance with the banking instructions set out in paragraph 4 of **Annex 2 to this Agreement**.
3. When the **Recipient transfers Plant Genetic Resources for Food and Agriculture under Development**, the transfer shall be made on the condition that the subsequent recipient shall pay into the mechanism established by the **Governing Body** under **Article 19.3f of the Treaty** zero point five percent (0.5 %) of the **Sales** of any **Product** derived from such **Plant Genetic Resources for Food and Agriculture under Development**, whether the **Product** is available or not without restriction.
4. At least six months before the expiry of a period of ten years counted from the date of signature of **this Agreement** and, thereafter, six months before the expiry of subsequent periods of five years, the **Recipient** may notify the **Governing Body** of his decision to opt out from the application of **this Article** as of the end of any of those periods. In the case the **Recipient** has entered into other **Standard Material Transfer Agreements**, the ten years period will commence on the date of signature of the first **Standard Material Transfer Agreement** where an option for **this Article** has been made.
5. Where the **Recipient** has entered or enters in the future into other **Standard Material Transfer Agreements** in relation to material belonging to the same crop[s], the **Recipient** shall only pay into the referred mechanism the percentage of sales as determined in accordance with **this Article** or the same **Article** of any other **Standard Material Transfer Agreement**. No cumulative payments will be required.

Annex 4

**OPTION FOR CROP-BASED PAYMENTS UNDER THE ALTERNATIVE PAYMENTS
SCHEME UNDER ARTICLE 6.11 OF THIS AGREEMENT**

I () declare to opt for payment in accordance with Article 6.11 of **this Agreement**.

Signature.....

Material Transfer Agreement (MTA) for Breeding Materials under Development by ICRISAT⁷

ICRISAT develops and shares improved breeding materials of mandate crops (sorghum, pearl millet, chickpea, pigeonpea and groundnut) with partners who use these materials to develop improved cultivars for adoption by farmers to enhance production and productivity of mandate crops in the SAT.

The breeding materials (from both conventional and biotechnology-assisted breeding) are products of research carried out wholly or in part by ICRISAT and are the property of ICRISAT. The breeding materials (hereafter referred to as the “material”) in the attached list are being furnished by ICRISAT, under the following additional conditions, as allowed in the Standard Material Transfer Agreement (SMTA) for products under development by the Center:

1. ICRISAT is making the material available (as per the Recipient's request) for purposes of research, breeding, and training for food and agriculture, for the exclusive use of _____ (the Recipient).
2. The Recipient will not claim ownership over the material, nor seek Intellectual Property Rights (IPRs) over the material, or its genetic parts or components, in the form received. The Recipient also agrees not to seek IPRs over related information received⁸
3. The Recipient will ensure that the material is exclusively used for purposes of research, breeding and training purposes of food and agriculture in their organization/institution.
4. ICRISAT reserves the right to distribute the materials to other parties.
5. ICRISAT makes no warranties as to the safety or title of the material, as described in Article 9.1 of SMTA.
6. Recipients are requested to provide ICRISAT with related data and information collected during evaluation and utilization of the material.
7. All other conditions, as outlined in the SMTA (copy attached) and agreed to by the Recipient, are applicable, except Article 5(a).

Signature: _____ Designation: _____

Name: _____ Institution: _____

Address: _____

⁷ MTA for Annex-I material (sorghum, pearl millet, pigeonpea and chickpea).

⁸ This does not prevent the recipient from releasing the material (or its products) to farmers for cultivation, provided that the other conditions set out in this MTA are complied with. Materials released should be acknowledged and ICRISAT should be informed of the details.

11.2 Appendix 2:

Graphical Genotypes (for Target Linkage Groups Only) of Pearl Millet Contiguous Segmental Substitution Lines Generated by Ms. B. Ramana Kumari in the course of her Ph.D. Thesis Research Program (Ramana Kumari *et al.* 2010a, 2010b)

Linkage Group 1 Contig Lines and their recurrent and donor parents

Plot no.	Seed source	Pot no. (DNA)	Xpsms86	Xpsmp2069	Xpsmp2273	Xpsms35	Xpsms38	Xpsms58	Xicmp3080	Xicmp3017	Xicmp3032	Xctm12	Comments
91001	ICMB 841-P3	841B-P3	B	B	B	B	B	B	B	B	B	B	Recurrent parent
91002	863B-P2	863B-P2	A	A	A	A	A	A	A	A	A	A	Donor parent
91003	S2005 RP 11B: 05036-04 self	71066	B	-	B	-	B	-	B	B	B	B	Parental
91004	S2005 RP 11B: 05036-12 self	71074	-	-	B	B	A	-	B	B	B	B	Segmental introgression
91005	S2005 RP 11B: 05038-02 self	71080	A	A	A	B	B	B	B	B	B	B	Segmental introgression
91006	S2005 RP 11B: 05038-05 self	71083	-	B	B	B	B	-	B	B	B	B	Parental
91007	S2005 RP 11B: 05039-01 self	71085	-	A	A	B	B	-	B	B	B	B	Segmental introgression
91008	S2005 RP 11B: 05039-04 self	71088	B	B	B	B	B	B	B	B	B	B	Parental
91009	S2005 RP 11B: 05040-11 self	71117	B	B	B	B	B	B	B	B	B	B	Parental
91010	S2005 RP 11B: 05040-12 self	71118	B	A	A	A	A	A	A	A		B	Segmental introgression
91011	S2005 RP 11B: 05048-01 self	71134	-	A	A	A	-	-	B	B	B	B	Segmental introgression
91012	S2005 RP 11B: 05048-06 self	71139	-	B	B	B	-	B	B	B	B	B	Parental
91013	S2005 RP 11B: 05050-16 self	71174	-	A	A	-	-	-	B	B	B	B	Segmental introgression
91014	S2005 RP 11B: 05051-04 self	71178	-	A	A				B	-	B	B	Segmental introgression
91015	S2005 RP 11B: 05051-07 self	71181	-	B	B	B	B	B	B	B	B	B	Parental
91016	S2005 RP 11B: 05055-03 self	71218	B	B	B	B	B	A	A	A	A	B	Segmental introgression,

91017	S2005 RP 11B: 05055-05 self	71220	B	B	B	B	B	A	A	A	A	B	Segmental introgression,
91018	S2005 RP 11B: 05055-11 self	71226	B	B	B	B	B	B	B	B	B	B	Parental
91019	S2005 RP 11B: 05060-01 self	71268	B	A	A	B	A	B	B	B	B	B	Segmental introgression,
91020	S2005 RP 11B: 05060-06 self	71273	B	A	A	A	A	B	B	B	B	B	Segmental introgression,
91021	S2005 RP 11B: 05060-14 self	71281	B	A	A	A	A	B	B	B	B	B	Segmental introgression,

Linkage Group 3 Contig Lines and their recurrent and donor parents

				<i>Xpsmp108</i>	<i>Xpsms61</i>	<i>Xpsms31</i>	<i>Xpsms60</i>	<i>Xpsmp2214</i>	<i>Xpsmp2249</i>	<i>Xctm10</i>	<i>Xpsmp2227</i>	
Plot no.	Pot No.(DNA)	Origin of seed sown (from RP11B S2005)	Generation sown									
93001	841B-P3	ICMB 841-P3	Recurrent Parent	B (350bp)	B(560bp)	B(380bp)	B(502bp)	B(240bp)	B(130bp)	B(190bp)	B(194bp)	B(27
93002	863B-P2	863B-P2	Donor Parent	A (410bp)	A(540bp)	A(400bp)	A(505bp)	A(244bp)	A(154bp)	A(186bp)	A(196bp)	A(24
93003	73074	S2005 RP 11B: 05107-15 self	BC5F3	B	B	B	B	B	B	B	B	B
93004	73078	S2005 RP 11B: 05103-02 self	BC5F3	B	B	B	B	B	B	B	B	B
93005	73083	S2005 RP 11B: 05103-07 self	BC5F3	B	B	B	B	B	B	B	B	B
93006	73079	S2005 RP 11B: 05103-03 self	BC5F3	A	B	B	A	B	B	B	B	B
93007	73052	S2005 RP 11B: 05084-01 self	BC5F3	A	B	B	B	B	B	B	B	B
93008	73014	S2005 RP 11B: 05089-02 self	BC5F3	B	B	B	B	B	B	B	B	B
93009	73023	S2005 RP 11B: 05089-11 self	BC5F3	B	B	B	B	A	A	A	A	A
93010	73029	S2005 RP 11B: 05091-04 self	BC5F3	B	B	B	B	A	A	A	A	A
93011	73046	S2005 RP 11B: 05093-08 self	BC5F3	B	B	B	B	A	A	A	A	A
93012	73048	S2005 RP 11B: 05093-10 self	BC5F3	B	B	B	A	A	A	A	A	A

93013	73082	S2005 RP 11B: 05103-06 self	BC5F3	B	B	B	A	A	A	A	A	A	E
93014	73086	S2005 RP 11B: 05103-10 self	BC5F3	B	B	B	A	A	A	A	A	A	E
93015	73087	S2005 RP 11B: 05103-11 self	BC5F3	B	B	B	A	B	B	B	B	B	E

Linkage Group 4 Contig Lines and their recurrent and donor parents

Plot no	Seed source		Pot no. (DNA)	<i>psmp716</i>	<i>psmp2076</i>	<i>psmp2081</i>	<i>psmp305</i>	<i>psms27</i>	<i>psmp2084</i>	<i>psms16</i>	Comment
94001	ICMB 841-P3		841B-P3	B(1000Ap)	B(160Ap)	B(200Ap)	B(900Ap)	B(246bp)	B(229Ap)	B(460bp)	Recurrent parent
94002	863B-P2		863B-P2	A(650Ap)	A(148Ap)	A(211Ap)	A(800Ap)	A(237bp)	A(206Ap)	A(470bp)	Donor parent
94003	S2005 RP 11B: 05139-01 self	84010	74011	B	B	O	A	A	A	A	Short LG4 introgression
94004	S2005 RP 11B: 05139-04 self	84011	74014	B	B	B	A	A	A	A	Short LG4 introgression
94005	S2005 RP 11B: 05139-10 self	84009	74020	B	B	B	B	B	B	B	Recurrent parent home (control)
94006	S2005 RP 11B: 05139-05 self	84012	74015	B	B	B	A	O	A	A	Short LG4 introgression
94007	S2005 RP 11B: 05182-05 self	84017	74135	B	B	B	A	A	A	B	Short LG4 introgression
94008	S2005 RP 11B: 05212-06 self	84025	74382	A	A	A	B	B	B	B	Short LG4 introgression
94009	S2005 RP 11B: 05212-07 self	84026	74383	A	A	A	B	B	B	B	Short LG4 introgression
94010	S2005 RP 11B: 05214-08 self	84028	74412	B	B	B	B	B	B	B	Recurrent parent home (control)
94011	S2005 RP 11B: 05214-07 self	84029	74411	A	A	A	B	B	B	B	Short LG4 introgression
94012	S2005 RP 11B: 05214-12 self	84030	74416	A	A	A	B	B	B	B	Short LG4 introgression
94013	S2005 RP 11B: 05216-12 self	84031	74448	A	A	O	B	B	B	B	Short LG4 introgression
94014	S2005 RP 11B: 05216-13 self	84032	74449	A	A	O	B	B	B	B	Short LG4 introgression
94015	S2005 RP 11B: 05218-15 self	84036	74484	B	B	B	B	B	B	B	Recurrent parent home (control)

94016	S2005 RP 11B: 05218-03 self	84037	74472	A	A	A	B	B	B	B	Short LG4 introgression
94017	S2005 RP 11B: 05218-09 self	84038	74478	A	A	A	B	B	B	B	Short LG4 introgression
94018	S2005 RP 11B: 05218-12 self	84039	74481	A	A	A	B	B	B	B	Short LG4 introgression
94019	S2005 RP 11B: 05218-13 self	84040	74482	A	A	A	B	B	B	B	Short LG4 introgression
94020	S2005 RP 11B: 05221-02 self	84041	74523	A	A	A	B	B	B	B	Short LG4 introgression
94021	S2005 RP 11B: 05221-05 self	84042	74526	A	A	A	B	B	B	B	Short LG4 introgression
94022	S2005 RP 11B: 05221-13 self	84043	74534	A	A	A	B	B	B	B	Short LG4 introgression
94023	S2005 RP 11B: 05218-07 self	84044	74541	B	B	B	B	B	B	B	Recurrent parent home (control)
94024	S2005 RP 11B: 05218-03 self	84049	74537	A	A	A	B	B	B	B	Short LG4 introgression
94025	S2005 RP 11B: 05218-05 self	84050	74539	A	A	A	B	B	B	B	Short LG4 introgression
94026	S2005 RP 11B: 05218-09 self	84051	74543	A	A	A	B	B	B	B	Short LG4 introgression
94027	S2005 RP 11B: 05218-15 self	84052	74549	A	A	A	B	B	B	B	Short LG4 introgression
94028	S2005 RP 11B: 05218-01 self	84053	74535	A	B	B	B	B	B	B	Short LG4 introgression
94029	S2005 RP 11B: 05227-21 self	84057	74573	B	B	B	B	B	B	B	Recurrent parent home (control)
94030	S2005 RP 11B: 05227-18 self	84058	74570	A	A	A	B	B	B	A	Two short LG4 introgr
94031	S2005 RP 11B: 05227-19 self	84059	74571	A	A	A	B	B	B	A	Two short LG4 introgr
94032	S2005 RP 11B: 05227-15 self	84060	74567	A	A	A	B	B	B	NA	Short LG4 introgression
94033	S2005 RP 11B: 05229-18 self	84063	74591	B	B	B	B	B	B	B	Recurrent parent home (control)
94034	S2005 RP 11B: 05229-01 self	84064	74574	A	B	B	B	B	B	B	Short LG4 introgression
94035	S2005 RP 11B: 05229-05 self	84065	74578	A	B	B	B	B	B	B	Short LG4 introgression
94036	S2005 RP 11B: 05229-12 self	84066	74585	A	B	B	B	B	B	B	Short LG4 introgression
94037	S2005 RP 11B: 05232-07 self	84067	74601	B	B	B	B	B	B	A	Short LG4 introgression
94038	S2005 RP 11B: 05232-13 self	84068	74607	B	B	B	B	B	B	A	Short LG4 introgression
94039	S2005 RP 11B: 05232-11 self	84069	74605	A	A	A	B	B	B	A	Two short LG4 introgr

94040	S2005 RP 11B: 05233-07 self	84070	74618	A	A	A	B	B	B	A	Two short LG4 introgression
94041	S2005 RP 11B: 05233-09 self	84071	74620	A	A	A	B	B	B	A	Two short LG4 introgression
94042	S2005 RP 11B: 05233-03 self	84072	74614	A	B	B	B	B	B	B	Short LG4 introgression
94043	S2005 RP 11B: 05240-06 self	84073	74633	B	B	B	O	B	B	A	Short LG4 introgression
94044	S2005 RP 11B: 05245-15 self	84074	74692	B	B	B	B	B	B	B	Recurrent parent home (control)
94045	S2005 RP 11B: 05245-12 self	84075	74689	A	A	A	O	B	B	A	Two short LG4 introgression
94046	S2005 RP 11B: 05245-08 self	84076	74685	A	A	A	B	B	B	B	Short LG4 introgression
94047	S2005 RP 11B: 05251-02 self	84077	74698	A	A	A	B	B	B	A	Two short LG4 introgression
94048	S2005 RP 11B: 05251-18 self	84078	74714	A	A	A	B	B	B	A	Two short LG4 introgression
94049	S2005 RP 11B: 05255-01 self	84079	74779	A	A	A	B	B	B	B	Short LG4 introgression
94050	S2005 RP 11B: 05261-11 self	84081	74874	B	B	B	B	B	B	B	Recurrent parent home (control)
94051	S2005 RP 11B: 05261-12 self	84082	74875	A	B	B	B	B	B	B	Short LG4 introgression

Linkage Group 5 Contig Lines and their recurrent and donor parents

Plot no	Seed source	Pot no. (DNA)	<i>icmp 3027</i>	<i>icmp3078</i>	<i>psmp2202</i>	<i>psms74</i>	<i>psmp318</i>		<i>psmp2274</i>	<i>ctm25</i>	<i>psms56</i>	<i>psmp2078</i>	<i>psms70</i>	<i>psmp2229.1</i>
95001	ICMB 841-P3	841B-P3	B(18 6bp)	B(260bp)	B(161bp)	B(825bp)	B(950bp)			B(270bp)	B(620bp)	B(168bp)	B(325bp)	B(250bp)
95002	863B-P2	863B-P2	A(20 5bp)	A(265bp)	A(145bp)	A(820bp)	A(900bp)			A(286bp)	A(605bp)	A(180bp)	A(320bp)	A(225bp)
95003	S2005 RP 11B: 05270-12 Self	75022	A	A	A	A	A			A	A	A	A	B
95004	S2005 RP 11B: 05270-13 Self	75023	A	A	0	A	A			A	A	B	B	
95005	S2005 RP 11B: 05271-05 Self	75034	B	B	B	B	B			B	A	A	A	H
95006	S2005 RP 11B: 05271-06 Self	75035	B	B	B	B	B			B	A	A	A	H
95007	S2005 RP 11B: 05272-04 Self	75048	B	B	B	B	B			B	A	A	A	H
95008	S2005 RP 11B: 05272-10 Self	75054	B	B	B	B	B			B	A	A	A	H
95009	S2005 RP 11B: 05273-08 Self	75064	B	B	B	B	H			B	A	A	A	B
95010	S2005 RP 11B: 05273-17 Self	75073	H	B	B	B	B			B	A	A	B	B
95011	S2005 RP 11B: 05273-18 Self	75074	B	B	B	B	B			B	A	A	A	B
95012	S2005 RP 11B: 05274-01 Self	75075	A	A	A	A	A			A	N	A	A	B
95013	S2005 RP 11B: 05274-19 Self	75093	A	A	A	A	A			A	B	A	A	B
95014	S2005 RP 11B: 05275-03 Self	75098	B	B	B	B	B			A	B	A	A	B
95015	S2005 RP 11B: 05276-10 Self	75114	A	A	A	A	A/?			A	A	A	H/A	H
95016	S2005 RP 11B: 05276-13 Self	75117	A	A	A	A	A			0	A	A	A	

95017	S2005 RP 11B: 05277-11 Self	75132	B	B	B	B	B			B	B	B	B	B
95018	S2005 RP 11B: 05277-12 Self	75133	A	A	A	A	A			A	B	H	A	B
95019	S2005 RP 11B: 05277-13 Self	75134	B	B	B	A	B			B	B	H	H	B
95020	S2005 RP 11B: 05277-05 Self	75126	B	B	B	B	A			B	B	B	B	B
95021	S2005 RP 11B: 05278-12 Self	75146	B	B	B	B	B			B	B	0	H/B	B
95022	S2005 RP 11B: 05278-09 Self	75143	A	A	A	H	A			0	A	A	A	B
95023	S2005 RP 11B: 05279-11 Self	75159	A	A	A	A	A			A	A	A	A	B
95024	S2005 RP 11B: 05279-08 Self	75156	B	B	B	B	B/?			B	H	H	H	B
95025	S2005 RP 11B: 05280-02 Self	75164	B	B	B	B	B			B	B	B	B	B
95026	S2005 RP 11B: 05280-11 Self	75173	A	A	A	A	A			A	B	B	B	B
95027	S2005 RP 11B: 05281-03 Self	75180	H	B	B	B	B			A	B	B	B	B
95028	S2005 RP 11B: 05281-09 Self	75186	B	B	B	B	B			A	A	A	B	B
95029	S2005 RP 11B: 05281-13 Self	75190	B	B	B	B	B			B	A	A	B	B
95030	S2005 RP 11B: 05282-09 Self	75201	A	A	A	A	A			B	A	0	H	B
95031	S2005 RP 11B: 05283-04 Self	75209	B	B	B	B	B			B	B	B	A	B
95032	S2005 RP 11B: 05283-13 Self	75218	B	A	B	B	B			B	B	B	A	B
95033	S2005 RP 11B: 05283-03 Self	75208	H	B	B	B	B			B	B	B	B	B
95034	S2005 RP 11B: 05288-04 Self	75289	B	B	B	A	A			A	A	B	B	B
95035	S2005 RP 11B: 05288-06 Self	75291	B	B	B	A	A			A	B	B	B	B

Linkage Group 6 Contig Lines and their recurrent and donor parents

Plot no.	Seed source	Pot No. (DNA)	<i>psmp2270</i>	<i>psmp2213</i>	<i>psms59</i>	<i>icmp3002</i>	<i>icmp3058</i>	<i>icmp3086</i>	<i>icmp3050</i>	<i>icmp3038</i>	<i>psms41</i>	Comments
96001	ICMB 841-P3	841B	B(163)	B(195)	B(260,270,280)	B(193)	B(200)	B(148)	B(225)	B(147)	B(300)	
96002	863-P2	863B	A(146)	A(197)	A(262,272,282)	A(186)	A(180)	A(151)	A(204)	A(189)	A(210)	
96003	S2005 RP 11B: 05294-04self	86014	B	B	B	A	B	B	B	A	A	Segmental introgression
96004	S2005 RP 11B: 05294-09self	86019	A	A	B	B	B	B	B	B	B	Segmental introgression
96005	S2005 RP 11B: 05294-15self	86025	A	A	B	B	B	B	B	B	B	Segmental introgression
96006	S2005 RP 11B: 05294-17self	86027	A	A	B	B	B	B	B	B	B	Segmental introgression
96007	S2005 RP 11B: 05294-05self	86015	B	B	B	B	B	B	B	B	B	Recurrent parent homo. (control)
96008	S2005 RP 11B: 05299-02self	86042	B	B	B	B	B	B	B	A	B	Segmental introgression
96009	S2005 RP 11B: 05299-12self	86052	A	A	A	A	A	A	A	A	B	Segmental introgression
96010	S2005 RP 11B: 05309-01self	86053	A	A	A	A	A	A	A	A	B	Segmental introgression
96011	S2005 RP 11B: 05309-12self	86064	A	A	A	A	A	A	A	A	B	Segmental introgression
96012	S2005 RP 11B: 05358-02self	86066	A	A	A	A	A	A	A	B	B	Segmental introgression
96013	S2005 RP 11B: 05371-08self	86083	B	B	B	B	B	B	B	B	B	Recurrent parent homo. (control)
96014	S2005 RP 11B: 05373-02self	86088	B	B	A	H	A	A	A	B	B	Segmental introgression
96015	S2005 RP 11B: 05373-10self	86096	B	B	A	A	A	A	A	B	B	Segmental introgression
96016	S2005 RP 11B: 05373-14self	86100	B	B	A	A	A	A	A	B	B	Segmental introgression
96017	S2005 RP 11B: 05373-06self	86092	B	B	B	B	B	B	B	B	B	Recurrent parent homo. (control)

Linkage Group 7 Contig Lines and their recurrent and donor parents

Plot no.	Seed source	Pot no. (DNA)	<i>icmp3048</i>	<i>psmp2224</i>	<i>psmp2229</i>	<i>psmp2074</i>	<i>psmp2271</i>	<i>psmp2263</i>	<i>psmp2063</i>	<i>psmp2203</i>	<i>icmp3043</i>	
97001	841B-P3	841B	B(229,245bp)	B(146,158bp)	B(225bp)	B(209bp)	B(184bp)	B(240bp)	B(171bp)	B(357bp)	B(221,235bp)	B/H
97002	863B-P2	863B	A(233,250bp)	A(144,156bp)	A(250bp)	A(210bp)	A(180bp)	A(219bp)	A(175bp)	A(355bp)	A(216,230bp)	A
97003	S2005 RP 11B: 05442-01 self	87001	B	B	A	B		B	B	B		
97004	S2005 RP 11B: 05442-02 self	87002	B	B	B	A	A	A	A	A	A	
97005	S2005 RP 11B: 05442-05 self	87005	B	A	A	A		A	A			
97006	S2005 RP 11B: 05442-16 self	87016	B	B	B	B	B	B	B	B	B	
97007	S2005 RP 11B: 05450-12 self	87032	B	B	B	A	A	B	A	B	B	
97008	S2005 RP 11B: 05450-15 self	87035	B	B	B	B	B		B	B	B	
97009	S2005 RP 11B: 05468-05 self	87041	B	B	B	B	B	B	B	B	B	
97010	S2005 RP 11B: 05468-13 self	87049	A	A	A	A	A	A	A	A	A	
97011	S2005 RP 11B: 05470-04 self	87056	B	B	B	B	B	B	B	B	B	
97012	S2005 RP 11B: 05472-07 self	87070	A	A	A	A	A	A	A	A	A	
97013	S2005 RP 11B: 05472-08 self	87071	A	A	A	B	B	B	B	B	B	
97014	S2005 RP 11B: 05472-18 self	87081	B	A		A	A	A	A	A	A	
97015	S2005 RP 11B: 05476-__	87104	B	B			B	B		B	B	

97016	S2005 RP 11B: 05477-__	87117	B	B	B	B	B	B	B	B	B	B
97017	S2005 RP 11B: 05478-__	87130	B	B		A				A		
97018	S2005 RP 11B: 05478-__	87134	B	B	B	A	A	A	A	A	A	A
97019	S2005 RP 11B: 05479-__	87148	B	B	B	A	B	A	A	A	A	A
97020	S2005 RP 11B: 05479-__	87150	B	B	B	B	B	B	B	B	B	B
97021	S2005 RP 11B: 05481-__	87157	B	B	B	B	B	B	B	B	B	B
97022	S2005 RP 11B: 05481-__	87167	B	A	A	A	A	A	A	A	A	A
97023	S2005 RP 11B: 05481-__	87170	B	B	B	B	B	B	B	B	B	B
97024	S2005 RP 11B: 05484-__	87174	B	B	A	B	B	B	B	B	B	B
97025	S2005 RP 11B: 05484-__	87188	B	A		A	A	A	A	A	A	A
97026	S2005 RP 11B: 05487-__	87195	B	A	A	A	A	B	A	A	A	A
97027	S2005 RP 11B: 05509-09 self	87209	B	B	B	B	B	B	B	B	B	B
97028	S2005 RP 11B: 05510-04 self	87214	B	B	B	B	B	A	B	A	A	A
97029	S2005 RP 11B: 05510-07 self	87217	B	B	B	B	B	A	B	A	A	A
97030	S2005 RP 11B: 05510-12 self	87222	B	B	B	B	B	A	B	A	A	A
97031	S2005 RP 11B: 05510-16 self	87226	B	B	B	B	B	A	B	A	A	A

11.3 Appendix 3:

List of Visiting Scientists participating in various project-sponsored training programs

Ser. No.	Name and Institutional Affiliation	Duration (dd/mm/yy)	
		Start	End
1	Mr. Rakesh Pathak; Central Arid Zone Research Institute, Jodhpur, Rajasthan	08/12/2005	24/12/2005
2	Mr. Manohar Singh, Central Arid Zone Research Institute, Jodhpur, Rajasthan	08/12/2005	24/12/2005
3	Ms. Bindu Nirwan, Project Coordination Unit, All-India Coordinated Pearl Millet Improvement Project, Jodhpur, Rajasthan	01/05/2006	30/06/2006
4	Dr. B.S. Rajpurohit, Project Coordination Unit, All-India Coordinated Pearl Millet Improvement Project, Jodhpur, Rajasthan	01/11/2006	15/01/2007
5	Dr. C.J. Dangaria, Pearl Millet Research Station, Gujarat Agricultural University, Jamnagar, Gujarat	02/01/2007	15/03/2007
6	Dr. Manoj Srivastava., Punjab Agricultural University, Ludhiana	02/01/2007	01/04/2007
7	Dr. Sita Ram Kumar, Rajasthan Agriculture University Bikaner	04/01/2007	03/04/2007
8	Dr. Kannan Baskaran, International Crops Research Institute for the Semi-Arid Tropics, Hyderabad, Andhra Pradesh	01/02/2007	31/01/2009
9	Ms. Dimple Kachhawa, Project Coordination Unit, All-India Coordinated Pearl Millet Improvement Project, Jodhpur, Rajasthan	02/02/2007	01/05/2007
10	Dr. L.D. Sharma, Agricultural Research Station Durgapura, Rajasthan Agricultural University, Jaipur, Rajasthan	28/11/2007	31/03/2008
11	Dr. C. Tara Satyavathi, Indian Agricultural Research Institute, New Delhi, Delhi	25/02/2008	24/04/2008
13	Dr. Yogendra P. Khedikar, University of Saskatchewan, Canada	03/03/2008	24/12/2009
14	Dr. O.P. Yadav, Central Arid Zone Research Institute, Jodhpur, Rajasthan	11/09/2006	30/09/2006
15	Dr. G. Alexander, Indian Veterinary Research Institute, Palampur	06/8/2005	06/9/2007
16	Ms. Sunita Choudhary, International Crops Research Institute for the Semi-Arid Tropics, Hyderabad, Andhra Pradesh	08/2009	project end