

Biotech tools improve pearl millet

RIU

Validated RNRRS Output.

Genes can now be inserted into pearl millet to make them more tolerant to heat and drought, and more resistant to disease. Millet is a staple cereal and important animal feed in the hot dry regions of Sub-Saharan Africa and India. High temperatures often kill millet seedlings. This means that the crop has to be re-sown and so doesn't ripen before the rains end. When the rains end early, millet produces very little grain or leaf. Plus, up to a third of the crop is often lost to downy mildew. The genes that help plants withstand the three main threats—heat, drought and mildew—could vastly improve harvests. They are already being used in India by government and international research laboratories.

Project Ref: **PSP23:**

Topic: **1. Improving Farmers Livelihoods: Better Crops, Systems & Pest Management**

Lead Organisation: **Institute of Grassland and Environmental Science (IGER), UK**

Source: **Plant Sciences Programme**

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Description

PSP23

Research into Use

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Geographical regions included:

[India](#),

Target Audiences for this content:

[Crop farmers](#),

A. Description of the research output(s)

1. Working title of output or cluster of outputs.

Genetic Improvement of pearl millet seedling thermotolerance and terminal drought tolerance

2. Name of relevant RNRRS Programme(s) commissioning supporting research and also indicate other funding sources, if applicable.

Plant Sciences Research Programme

The outputs of this research have been stored in the MilletGenes database, which was initiated at JIC with DFID-PSRP funding (R6354; *Pearl millet bioinformatics to promote the distribution and uptake of research results*) and has now been incorporated into the BBSRC-funded UK CropNet Programme.

3. Provide relevant R numbers (and/or programme development/dissemination reference numbers covering supporting research) along with the institutional partners (with individual contact persons (if appropriate)) involved in the project activities. As with the question above, this is primarily to allow for the legacy of the RNRRS to be acknowledged during the RiUP activities.

R5487, R6451 and R7375 (1992-2002)

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4. Describe the RNRRS output or cluster of outputs being proposed and when was it produced? (max. 400 words).

This requires a clear and concise description of the output(s) and the problem the output(s) aimed to address. Please incorporate and highlight (in bold) key words that would/could be used to select your output when held in a database.

Pearl millet [*Pennisetum glaucum* (L.) R. Br.] is the staple cereal grain and fodder crop grown by subsistence farmers in the hottest, driest regions of the Sahelian zone of sub-Saharan Africa and the Indian subcontinent where rainfed crop production is possible. Early cessation of rains is the single most important factor contributing to low mean grain and straw yields and poor yield stability of this crop. Failure of seedling establishment due to extreme high soil temperatures at the start of the growing season result in the need to re-sow and an increased likelihood that the crop will be exposed to the damaging effects of post-flowering drought. These projects which were completed in 2002 resulted in the development of **molecular markers** linked to **thermotolerance**, **drought tolerance** and **disease resistance**. These were then used within a breeding programme to enable the development of elite hybrid parents. 3 separate pearl millet mapping populations were developed for this purpose (ICMP 451 × H77/833-2; H77/833-2 × PRLT 2/89-33 and 841B × 863B). In particular a major drought tolerance QTL mapping to linkage group 2 was identified. Using marker assisted backcrossing and conventional sexual crossing genomic regions (QTLs) which provide enhanced performance under terminal drought (identified in R6451) were transferred into the agronomically elite pollinator, H 77/833-2, in R7375. Drought-tolerant topcross pollinators were also bred from PRLT 2/89-33 × H77/833-2 using conventional and marker-assisted methods and evaluated. The cross ICMB 841 × 863B (again between elite parents) was used to verify the QTLs obtained, to identify new QTLs, to determine their stability over a range of test cross parents and environments, and to provide closely linked markers for the improvement of seed parent maintainer 841B. These markers were then used in R8183 to breed adapted, high-yielding, disease-resistant hybrid cultivars. The cross ICMP 451 × H77/833-2 originally developed to study seedling thermotolerance in R5487 proved excellent material for the identification of regions of the genome providing resistance to **downy mildew**. This devastating disease of pearl millet can result in a loss of 30% or more of the entire grain harvest. Marker-assisted backcross transfer of mapped QTLs from ICMP 451 to H77/833-2 was completed by a PhD students in India ensuing technology transfer of the methods developed. The research has been fully documented in a series of publications and presentations related to development and application of molecular genetic tools to assist pearl millet breeders in developing countries.

5. *What is the type of output(s) being described here?*

Please tick one or more of the following options.

Product	Technology	Service	Process or Methodology	Policy	Other Please specify
x	x				

6. *What is the main commodity (ies) upon which the output(s) focussed? Could this output be applied to other commodities, if so, please comment*

Pearl Millet. Also applicable to other cereals grown in drought- prone areas, in particular sorghum.

7. *What production system(s) does/could the output(s) focus upon?*

Please tick one or more of the following options. Leave blank if not applicable

Semi-Arid	High potential	Hillsides	Forest-Agriculture	Peri-urban	Land water	Tropical moist forest	Cross-cutting
x							

8. What farming system(s) does the output(s) focus upon?

Please tick one or more of the following options (see Annex B for definitions).

Leave blank if not applicable

Smallholder rainfed humid	Irrigated	Wetland rice based	Smallholder rainfed highland	Smallholder rainfed dry/cold	Dualistic	Coastal artisanal fishing
				x	x	

9. How could value be added to the output or additional constraints faced by poor people addressed by clustering this output with research outputs from other sources (RNRRS and non RNRRS)? (**max. 300 words**). Please specify what other outputs your output(s) could be clustered. At this point you should make reference to the circulated list of RNRRS outputs for which proformas are currently being prepared.

This project links closely with projects R6667 (marker-assisted selection, QTLs and contig lines for improving downy mildew resistance of pearl millet hybrids), R6951 (saturation of the pearl millet genetic maps with molecular markers and fine mapping of regions of agronomic importance) and R7379 (Marker-assisted improvement of pearl millet downy mildew resistance in elite hybrid parental lines for Africa and Asia). This set of projects were continued as part of R8183 (Exploiting marker-assisted methods for pearl millet improvement) resulting in the development and release of HHB 67-2, a downy mildew resistant version of the popular pearl millet hybrid HHB 67. This new hybrid, the first to be bred in India by the public sector using MAS had additional benefits of improved grain yield, improved stover yield (via increased plant height) and improved ruminant nutritional quality of stover via better foliar disease resistance.

Screening data from the work on drought tolerance has also been used to identify regions of the genome controlling components of grain and stover yield. The most interesting stover yield QTL detected so far is that from 863B in linkage group 3, which segregated independently from flowering time QTLs. This offers the potential to improve stover yield potential, without delaying maturity, by marker-assisted backcross transfer of part of the linkage group 3 from 863B into the genetic background of 841B. The male-sterile line counterpart of this new version of 841B could then be used to produce higher stover yield versions of the hybrids currently produced on 841A. Another trait that has been targeted for QTL mapping and MAS is improved ruminant nutritional quality of the stover. Interestingly the major genomic region providing enhanced drought tolerance has recently been demonstrated to contribute significantly to variation in ruminant nutritional quality of pearl millet straw, with the *Iniadi*-derived parent 863B contributing the favourable allele.

Alignment of regions of the genome relating to drought tolerance in pearl millet with related species has the potential to benefit less tolerant but more economically important crops such as rice in breeding programs worldwide.

Validation

B. Validation of the research output(s)

10. How were the output(s) validated and who validated them?

Please provide brief description of method(s) used and consider application, replication, adaptation and/or adoption in the context of any partner organisation and user groups involved. In addressing the “who” component detail which group(s) did the validation e.g. end users, intermediary organisation, government department, aid organisation, private company etc... This section should also be used to detail, if applicable, to which social group, gender, income category the validation was applied and any increases in productivity observed during validation (max. 500 words).

The effect of the drought tolerance QTL was validated by ICRISAT during R8183 in two independent marker-assisted backcrossing programs in which the 30% improvement in grain yield general combining ability (GCA) expected of this QTL has been recovered in the best introgression lines.

A simple initial evaluation of the drought tolerance QTL on LG 2 as a selection criterion was made by comparing hybrids made with topcross pollinators bred from progenies selected from the original mapping population for presence of the tolerant allele at the target QTL vs. field performance in the phenotyping environments (Bidingger et al., 2005, Field Crops Research 94:14-32). A set of 36 topcross hybrids was evaluated in 21 field environments by ICRISAT, which included both non-stressed and drought stressed treatments during the flowering and grain filling stages. The QTL-based hybrids were significantly higher yielding in a series of both absolute and partial terminal stress environments, but at the cost of a lower yield in the non-stressed evaluation environments.

A more rigorous evaluation of the drought tolerance QTL was completed by ICRISAT using new near-isogenic versions of H 77/833-2 into which various putative drought tolerance QTL segments were introgressed from donor parent PRLT 2/89-33 by marker-assisted backcrossing. BC4F3 progenies from selected BC4F2 plants homozygous for various portions of the LG2 target region were crossed to each of five different seed parents, and the resulting hybrids were evaluated under a range of moisture regimes (non-stressed control, early-onset, medium-onset and late-onset terminal drought stress). The hybrids exhibited a large variation in yield component expression and yield response to the moisture regimes, but there was a consistent yield advantage in hybrids carrying alleles from the drought tolerant donor parent PRLT 2/89-33 in the vicinity of the target QTL. Several of the QTL introgression lines (ICMR 01029 and ICMR 01031 in particular) had a significant, positive general combining ability (across all testcrosses) for grain yield under terminal stress, which was associated with a higher panicle harvest index.

A line-source, gradient stress experiment was conducted to further compare the performance of test crosses (on four seed parents) of the two pollinators with the highest GCA for yield under stress (ICMR 01029 and ICMR 01031), one pollinator with a negative GCA, and both mapping population parents. The results confirmed the previous findings of yield advantage of ICMR 01029 and ICMR 01031 compared to parents H-77/833-2 and PRLT-2/89-33. Leaf gas exchange measurements indicated that photosynthetic rate was significantly correlated with grain yield ($r^2=0.80$), indicating the existence of genotypic differences in the response of leaf gas exchange to drought, which could open new opportunities for better understanding of the plant water relations and selection

of drought tolerant varieties in pearl millet.

Similarly, a total of 13 lines are available in the background of ICMB 841 with introgressions of various portions of LG2 from donor parent 863B, and one of these (ICML 03050) appears to be particularly promising. 3-years of drought nursery evaluations by ICRISAT of testcrosses of these lines show that a 30% improvement in grain yield GCA was obtained using ICML 03050.

11. *Where and when* have the output(s) been validated? Please indicate the places(s) and country(ies), any particular social group targeted and also indicate in which production system and farming system, using the options provided in questions 7 and 8 respectively, above (**max 300 words**).

The outputs have been validated in semi-arid dualistic farming systems at ICRISAT-Patancheru and in drought-prone Northern India (CAZRI-Jodhpur and RAU-RSS Nagaur) in 2002, 2003, 2004 and 2005.

Current Situation

C. *Current situation*

12. *How and by whom* are the outputs currently being used? Please give a brief description (**max. 250 words**).

Our success has made available favourable alleles for this drought tolerance QTL on both sides (seed parent and pollinator) of the parentage of elite Indian pearl millet hybrids. This has subsequently been exploited by a number of projects.

The Department of Biotechnology (DBT) of the Government of India has funded 12 national institutions in a project entitled "Development and applications of biotechnological tools for millet improvement" This includes the Central Arid Zone Research Institute in Rajasthan to incorporate drought tolerance and downy mildew resistance into parental material of the most common millet hybrids grown in India using marker assisted selection.

The Generation Challenge program SP2 commissioned research in 2005 entitled 'Stress response-enriched EST resources for targeted species - Pearl Millet' in which HHB 67 and its more drought tolerant counterpart 843A x ICMR 01029 were used to identify expressed sequences relating to drought tolerance.

The Australian Centre for International Agricultural Research has funded a project on improving the quality of pearl millet residues for livestock using pearl millet mapping population ICMB 841 x 863B (01/01/2004 - 31/12/2008) at ICRISAT in collaboration with the International Livestock Research Institute, India

The Indian Council for Agricultural Research has funded ICRISAT in 2006/7 to advance several of the mapping populations to F6 RIL sets and transfer SSR marker-based maps from the earlier F2:4 populations to the F6 populations. This will also finish RFLP- & SSR-marker-assisted pyramiding of the drought tolerance QTL and two DMR QTLs in H 77/833-2 background and produce testcross hybrid seeds of the multiple-QTL homozygotes for multi-locational testing in 2007. It also provides training to several young scientists in the national program during this period.

13. *Where* are the outputs currently being used? As with Question 11 please indicate place(s) and countries where

the outputs are being used (max. 250 words).

The outputs are being used predominantly in India by several public-sector research laboratories including that at the Central Arid Zone Research Institute in Jodhpur, Rajasthan, India, which serves the hottest, driest, most marginal pearl millet production environments in India, and at the International Crops Research Institute for the Semi-Arid Tropics.

14. What is the scale of current use? Indicating how quickly use was established and whether usage is still spreading (max 250 words).

Usage is spreading but requires additional funding

15. In your experience what programmes, platforms, policy, institutional structures exist that have assisted with the promotion and/or adoption of the output(s) proposed here and in terms of capacity strengthening what do you see as the key facts of success? (max 350 words).

Consistent long term funding has resulted in outputs being made available. The public- and private-sectors of the Indian pearl millet improvement programme, in combination with the applied pearl millet hybrid parents breeding program at ICRISAT-Patancheru, provide the institutional platform from which the promotion, application, and adoption of outputs from R8183 and its predecessors must be made for south Asia. The MS Swaminathan Applied Genomics Laboratory, and its Center of Excellence for high-throughput marker data generation (being established with funding from the Government of India's Department of Biotechnology), can provide service lab facilities for marker data generation for all applied plant breeding in India. A DArT marker platform will soon be established at ICRISAT-Patancheru as part of this Center of Excellence, and application of this to high-throughput marker genotyping for pearl millet has the potential to completely revolutionize breeding of pearl millet hybrids for this region. The BecA facility, hosted by ILRI in Nairobi, Kenya, is expected to play a similar role for crop and livestock improvement programmes (both public and private) in eastern and central Africa, and similar facilities are under development in southern Africa and in western Africa. A Syngenta Foundation-sponsored project to develop marker-assisted breeding tools for pearl millet and sorghum in sub-Saharan Africa should also be considered as an important mechanism to be supported to broaden the scope of application of R8183 outputs to poverty alleviation and livelihood enhancement in developing countries.

Current Promotion

D. Current promotion/uptake pathways

16. Where is promotion currently taking place? Please indicate for each country specified detail what promotion is taking place, by whom and indicate the scale of current promotion (max 200 words).

The outputs of this research have been stored in the MilletGenes database, which was initiated at JIC with DFID-PSRP funding (R6354; *Pearl millet bioinformatics to promote the distribution and uptake of research results*) and has now been incorporated into the BBSRC-funded UK CropNet Programme. The database contains information on probes, DNA polymorphisms, gel images, genetic maps and QTL data. ICRISAT and National Agricultural Research Systems (NARS) scientists are now using this database to assist their MAS breeding programmes.

The Indian Council of Agricultural Research is providing ICRISAT with US\$100K during fiscal year 2006/07 for application of marker-assisted breeding for improvement of pearl millet hybrids for downy mildew resistance and terminal drought tolerance. In addition, the Government of India's Department of Biotechnology is supporting a Millets Biotechnology Network Project that includes funding to support staff training and limited marker lab development at the Central Arid Zone Research Institute and Project Coordination Unit of the All-India Coordinated Pearl Millet Improvement Project.

17. What are the current barriers preventing or slowing the adoption of the output(s)? Cover here institutional issues, those relating to policy, marketing, infrastructure, social exclusion etc. (max 200 words).

One barrier has been the feeling among both public- and private-sector pearl millet breeding programmes that the technology is too expensive to apply to a relatively low-value commodity like pearl millet. However the success with development, testing, release, multiplication, and dissemination of pearl millet hybrid "HHB 67 Improved" has gone a long way to overcoming this hesitation.

Whereas uptake of new varieties and hybrids is excellent in much of India, in Africa, by contrast, most farmers continue to plant traditional landrace cultivars. Improved varieties that are adapted to target environments and both farmer and market needs have been developed but have not been disseminated. One of the major limitations of the technology delivery system is lack of an efficient and effective seed multiplication and supply system. While there are signs of interest in new open-pollinated cultivars, private seed companies do not believe this area is profitable, and public sector investments in seed production are limited. The widespread promotion of hybrids in Asia has encouraged private investment in seed production, but the prospects for hybrid adoption in Africa remain unknown. The costs of distributing hybrid seed are higher than in Asia (because population densities are lower), and the willingness of the often poorer African farmer to purchase hybrid seed remains untested. However, given the low seed requirement and the low production costs (because of high multiplication rates), even poor pearl millet producers in Africa, similar to their counterparts in Asia, may find it worthwhile to invest in improved seed, either hybrid or open-pollinated. The extension systems in most African countries are also weak, thus causing a bottleneck in technology dissemination.

18. What changes are needed to remove/reduce these barriers to adoption? This section could be used to identify perceived capacity related issues (max 200 words).

The development of cheaper to use markers for pearl millet, and establishment of networks for rapidly and reliably moving pearl millet DNA samples to regional service labs that can provide marker data in a timely and cost-effective manner, and move marker data from these labs back to the breeding programmes, will permit application of the technology even in much of sub-Saharan Africa where improved open-pollinated varieties may remain the preferred pearl millet cultivar type for many years to come. This must be combined with large-scale training of pearl millet breeding teams in the application of markers to their breeding program objectives.

19. What lessons have you learnt about the best ways to get the outputs used by the largest number of poor people? (max 300 words).

Success has resulted from developing cultivars that are already popular but with specific weaknesses and identification of markers associated with genes to improve those traits that have then been used in marker assisted crossing programmes to address these weaknesses. Involve local breeding teams, the seed delivery infrastructure, and target farm families in identification of the problems, development and testing of possible

solutions, and product delivery.

Impacts On Poverty

E. Impacts on poverty to date

20. *Where have impact studies on poverty in relation to this output or cluster of outputs taken place? This should include any formal poverty impact studies (and it is appreciated that these will not be commonplace) and any less formal studies including any poverty mapping-type or monitoring work which allow for some analysis on impact on poverty to be made. Details of any cost-benefit analyses may also be detailed at this point. Please list studies here.*

No formal studies have yet been completed however, it is estimated that about £2.6 million worth of additional grain yield could be expected annually from a 10% increase in grain yield of the best new versions of pearl millet hybrid HHB 67. In years of severe drought stress this would be even more valuable as both grain and straw prices are higher.

21. *Based on the evidence in the studies listed above, for each country detail how the poor have benefited from the application and/or adoption of the output(s) (max. 500 words):*

It is premature to assess direct benefits to the poor.

- *What positive impacts on livelihoods have been recorded and over what time period have these impacts been observed? These impacts should be recorded against the capital assets (human, social, natural, physical and, financial) of the livelihoods framework;*
 - *For whom i.e. which type of person (gender, poverty group (see glossary for definitions) has there been a positive impact;*
 - *Indicate the number of people who have realised a positive impact on their livelihood;*
 - *Using whatever appropriate indicator was used detail what was the average percentage increase recorded*
-

Environmental Impact

H. Environmental impact

24. *What are the direct and indirect environmental benefits related to the output(s) and their outcome(s)? (max 300 words)*

Pearl millet is the staple cereal grain and fodder crop grown by subsistence farmers in the hottest, driest regions of the Sahelian zone of sub-Saharan Africa and the Indian subcontinent where rainfed crop production is possible. Rainfall in these regions is scant and unreliable. Early cessation of rains is the single most important

factor contributing to low mean grain and straw yields and poor yield stability of this crop. Enhancing the reliability of pearl millet grain and straw yields in such environments will contribute directly and sustainably to the alleviation of poverty and improvement of food security of farm households in the poorest parts of Africa and Asia. Predicted climate change scenarios indicate that water shortages and shorter effective growing season lengths will be increasingly likely in sub-Saharan Africa increasing the need for short-duration crops with enhanced drought tolerance. Pearl millet, which is more tolerant of high temperatures than any other cereal, is the ideal crop for such conditions. Efficient breeding for drought-prone environments requires selection strategies that are sufficiently independent of the variation inherent in the natural environment to permit identification and use of true genetic differences. Molecular markers linked closely to traits such as pearl millet grain and stover yield during drought will provide increased reliability for selecting and breeding of these traits. Developing new drought-resistant varieties however only reduces risk if resource-poor farmers have reliable access to high-quality seed at low cost.

This could include direct benefits from the application of the technology or policy action with local governments or multinational agencies to create environmentally sound policies or programmes. Any supporting and appropriate evidence can be provided in the form of an annex.

25. Are there any adverse environmental impacts related to the output(s) and their outcome(s)? (max 100 words)

26. Do the outputs increase the capacity of poor people to cope with the effects of climate change, reduce the risks of natural disasters and increase their resilience? (max 200 words)

Predicted climate change scenarios indicate that water shortages and shorter effective growing season lengths will be increasingly likely in sub-Saharan Africa increasing the need for short-duration crops such as pearl millet with enhanced drought tolerance. Development of crops not only able to withstand such conditions but also able to provide a usable yield will increase the resilience of poor people inhabiting those regions.
