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INDIAN SOCIETY OF PLANT GENETIC RESOURCES

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- To provide a forum for organizing symposia/conferences with a view to develop close relationship among the scientists engaged and interested in plant genetic resources activities.

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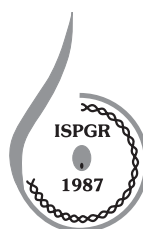
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Progress in Implementing the Biosafety Protocol: A Report on COP-MOP-6 Meeting Held in Hyderabad

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Sixth meeting of the Conference of Parties serving as the Meeting of Parties to the Cartagena Protocol on Biosafety was held in Hyderabad on 1-5 October 2012. Its agenda items included discussions on capacity-building, biosafety clearing-house, socio-economic considerations concerning living modified organisms (LMOs), reviewing effectiveness of the Protocol and taking decisions to further ensure the safe transfer, handling and use of LMOs. This paper seeks to analyse and assess the outcome of this meeting against the much awaited expectations and India's concerns.

Key Words: Biosafety, Biosafety Clearing House, Cartagena Protocol, COP-MOP 6 meeting, Compliance Committee, Liability and Redress, LMOs, Nagoya-KL Supplementary Protocol, Risk Assessment and Risk Management

Legally binding Cartagena Protocol on Biosafety (CPB) to the Convention on Biological Diversity (CBD), which entered into force on 11 September 2003, is an international treaty with 164 Parties to it, including India. It has a specific focus on transboundary movement of living modified organisms (LMOs) resulting from modern biotechnology that may have adverse effect on the conservation and sustainable use of biological diversity including risks to human health.

This Protocol also establishes an advance informed agreement (AIA) procedure for ensuring that countries are provided with the information necessary to make informed decisions before agreeing to the import of LMOs into their territory. It contains reference to a precautionary approach, reaffirming the Principle 15 of the Rio Declaration on Environment and Development, and also establishes a Biosafety Clearing-House (BCH) to facilitate the exchange of information on LMOs and to assist Parties in implementing the Protocol.

The governing body of the CPB, known as the Conference of the Parties to CBD serving as meeting of the Parties to the Protocol (COP-MOP in short) has held six meetings till date. Adoption of the Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress in 2010 notwithstanding, the progress in implementing this Protocol has been slow so far and differences among Parties on several issues still remain unresolved.

In an effort to fast track the pace, fifth meeting of the COP-MOP, held at Nagoya, Japan in 2010, adopted a 10-year Strategic Plan (2011-2020) to facilitate the implementation of the protocol. The sixth meeting of the COP-MOP was held in Hyderabad, India on 1-5 October 2012 which was attended by some 1,500 delegates from more than 100 countries. As the incoming President of COP-MOP, India must now strive to take the process forward during the next two years of its Presidency until COP-MOP-7 to be held in South Korea in 2014.

I) Key Issues before COP-MOP-6

Substantive issues arising from the programme of work and previous COP-MOP decisions were identified by the Secretariat as follows:

1. Risk assessment and risk management (Articles 15 & 16)
2. Socio-economic considerations (Article 26)
3. Handling, transport, packaging and identification (HTPI) of LMOs (Article 18)
4. Liability and redress (Article 27) – including status of Supplementary Protocol
5. Unintentional transboundary movements and emergency measures (Article 17)
6. Assessment and review (Article 35)
7. Capacity-building and the roster of experts

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II) Main Agenda Items

Secretariat's revised annotations to the provisional agenda, developed for the meeting, included the following items for consideration and guidance of COP-MOP6:

i. Risk Assessment and Risk Management

- The revised version of the "Guidance on Risk Assessment of Living Modified Organisms"
- The mechanism for updating the lists of background materials
- New specific topics of risk assessment on which to develop further guidance
- Capacity-building activities coordinated by the Secretariat and possible future activities based on elements of the Strategic Plan for the Protocol

ii. Socio-economic Considerations (Article 26)

- Review the report of the regional online conferences and the workshop on socio-economic (SE) considerations organised during the intersessional period
- Consider the need for establishing an Ad hoc Technical Expert Group (AHTEG), or other mechanisms, to continue the work on SE considerations, including (i) development of conceptual clarity on SE considerations; (ii) review of information available on specific cases of SE impacts of LMOs; and (iii) development of guidelines on SE considerations.

iii. Handling, Transport, Packaging and Identification of LMOs

- Review the progress with the implementation of HTPI requirements for LMOs destined for contained use and LMOs intended for intentional introduction into the environment and provide guidance on further action
- Review the results of a study on existing standards, methods and guidance relevant to the HTPI of LMOs and an analysis of the developments on existing rules and standards related to the HTPI of LMOs and consider the possible need for the elaboration of standards for HTPI of LMOs and/or provide guidance on the use of existing international regulations and standards.

iv. Liability and Redress (Article 27)

- Consider the status of signature, ratification or accession to the Supplementary Protocol on Liability and Redress and the activities undertaken to expedite its early entry into force

v. Unintentional Transboundary Movements and Emergency Measures

- Consider the need for, and modalities of, developing tools and guidance to facilitate Parties to take appropriate responses to unintentional transboundary movements and initiate necessary actions, including emergency measures

vi. Assessment and Review (Article 35)

- Consider the status of implementation of the Protocol and adopt measures for further improvement

vii. Capacity Building

- Review the status of capacity-building under the Protocol and adopt a new Capacity-building Framework and Action
- Adopt a revised nomination form for the roster of experts and consider measures to improve the use of the roster

III) Expectations

COP-MOP-6 meeting was expected to address and deliver on the following issues:

1. To continue its commitment to support capacity-building activities as an important tool for its effective implementation and review the updated Action Plan for this purpose to clearly set out the strategy and priorities for the coming period with robust measures relevant to the current policy context.
2. To support the work of the Compliance Committee in order to ensure full and effective implementation of the Protocol by encouraging the Committee to make full use of its new and enhanced supporting role and assist Parties in meeting their obligations on submission of national reports.
3. To encourage more active use of BCH mechanism and overcome difficulties to facilitate easy submission of information in this context.
4. To identify and support more effective ways forward for proper implementation of the risk assessment and risk management provisions under the Protocol,

based on recommendations of the Ad Hoc Technical Expert Group.

5. To promote early ratification of the Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress while also encouraging non-Parties to join the Cartagena Protocol.
6. To build up further on the progress achieved during the inter-sessional period between COP-MOP 5 and COP-MOP 6 on socio-economic considerations through regional online conferences and through the regionally-balanced workshop on capacity-building for research and information exchange on socio-economic impacts of LMOs and to identify way forward for socio-economic considerations.
7. To deliberate upon the crucial issue of the environmental risk assessment for taking decisions related to the intentional transboundary movement of LMOs in order to identify and evaluate their possible effects on the conservation and sustainable use of biological diversity taking into account risks to human health.
8. To work towards developing a consensus on the budget that is consistent with the agreed strategic priorities and programme of work for the Protocol's effective implementation and to mobilize greater resources for this purpose, including the next cycle of GEF funding.

IV) Brief Report on the Decisions taken by COP-MOP-6 Meeting

Indian Minister of Environment & Forests, Smt. Jayanthi Natarajan took over the COP-MOP-6 Presidency, from Mr Masamichi Saigo, Ministry of Agriculture, Forestry and Fisheries representing Japan's Presidency of COP-MOP-5, and stressed the need to develop a balance between health, technology and the environment and also urged upon the Parties to ratify the Supplementary Protocol on Liability and Redress. Following the inaugural session, Shri M.F. Farooqui, Spl. Secretary, Ministry of Environment & Forests, took over as the COP-MOP-6 Chair and conducted the proceedings.

The meeting adopted 16 decisions related to the following topics:

Compliance; the Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress; subsidiary bodies; cooperation with other international organizations, conventions and initiatives; the BCH; capacity building; the roster of experts; monitoring

and reporting; assessment and review; notification requirements; HTPI of LMOs; unintentional transboundary movements of LMOs; financial mechanism and resources; socio-economic considerations; risk assessment and risk management; and the budget.

1. Compliance Committee

During the closing plenary, COP-MOP Chair Shri Farooqui announced the nominations for Compliance Committee from the beginning of 2013 as follows:

Kaouthar Tliche Aloui (Tunisia) and Johansen Voker (Liberia) for the African Group; Dubravka Stepic (Croatia) and Angela Lozan (Moldova) for Central and Eastern European Group; Jimena Nieto (Colombia) and Hector Conde Almeida (Cuba) for Group of Latin American and Caribbean countries; Geoff Ridley (New Zealand and Reuben Dekker (the Netherlands) for the Western Europe and Others Group; and Rai S. Rana (India) and Banpot Napompeth (Thailand) for the Asia-Pacific Group.

COP/MOP Decision: In its decision (UNEP/CBD/BS/COP-MOP/6/L.2), the COP-MOP:

- Calls upon Parties to expedite efforts to put in place legal and administrative frameworks to meet obligations under the Protocol;
- Requests Parties that have not yet put in place operational biosafety frameworks to submit information on challenges, and on plans and timelines for taking necessary measures;
- Requests the Secretariat to compile and submit such information to the Compliance Committee for consideration and appropriate action;
- Reminds Parties experiencing challenges that they may seek assistance from the Compliance Committee; and
- Reiterates its invitation to make use of the programme of work on public awareness, education and participation on safe transfer, handling and use of LMOs.

2. Biosafety Clearing-House (BCH)

Discussions largely focused on the functionality and use of the BCH, the second phase of the UNEP-Global Environment Facility (GEF) project on capacity building support for the BCH and the project's likely extension. While many Parties supported extension, there were others who sounded caution.

COP/MOP Decision: In its decision (UNEP/CBD/BS/COP-MOP/6/L.6), the COP-MOP requests the Secretariat to:

- Collect, through the BCH national focal points and online tools made available to the BCH, feedback on existing capacity and experiences in using the BCH and the submission and retrieval of data, to take the experience into account for future improvements to the BCH;
- Continue its collaboration with other biosafety databases and platforms, such as those of the OECD and the UN Food and Agriculture Organisation (FAO), with a view to improving the utility of the BCH as a global mechanism for sharing information on biosafety;
- Continue to organize online forums and real time online conferences on topics relevant to biosafety and the implementation of the Protocol; and
- Encourages greater use of the BCH to further promote and facilitate public awareness, education and participation of relevant stakeholders in the use of LMOs.

The COP-MOP further urges Parties to fulfill their obligations under the Protocol and the decisions of the Parties by updating all incomplete published national records with the mandatory fields required by the common formats: and recommends that the COP, in adopting its guidance to the GEF, urge it to provide further support to all eligible Parties for the capacity building in the use of the BCH, based on experiences learned from the UNEP-GEF BCH-II.

3. *Financial Mechanism and Resources*

COP/MOP Decision: In the decision (UNEP/CBD/BS/COPMOP/6/L.14), the COP/MOP notes with concern the drastic decline in the level of bilateral and multilateral funding available for biosafety capacity-building activities.

The COP/MOP, *inter alia*, urges parties to give priority to national biosafety plans and projects under the GEF-STAR to ensure support for implementation of the Protocol.

On guidance to the financial mechanism, the COP/MOP recommends to the COP, in adopting further guidance with respect to financial support for implementation of the Protocol, to invite the GEF to *inter alia*:

- Further streamline, simplify and expedite the process of accessing funds from the GEF Trust Fund;
- Consider developing a new strategy for financing biosafety, incorporating the priorities and objectives of the Strategic Plan;
- Make available, in a timely manner, adequate and predictable financial resources to eligible parties to facilitate the preparation of their third national reports under the Protocol;
- Provide support to eligible parties that have not yet done so to initiate implementation of their legal, administrative and other measures for the implementation of the Protocol;
- Provide financial and technical assistance to developing country parties and parties with economies in transition to undertake the testing and capacity-building activities on risk assessment and risk management, and to implement detection and identification requirements of the Protocol;
- Make financial resources available to support awareness raising, experience-sharing and capacity-building activities to expedite the early entry into force and implementation of the Protocol and the Supplementary Protocol;
- Consider the following priorities within the four-year outcome oriented framework of programme priorities for biodiversity for GEF-6: national biosafety frameworks; risk assessment and risk management; HTPI and identification of LMOs; liability and redress, public awareness and biosafety education and training; and socio-economic considerations; and
- Consider making a notional allocation that improves the biosafety share of the biodiversity focal area to support the implementation of the Protocol during GEF-6. On mobilization of additional resources, the COP/MOP:
- Requests the Executive Secretary to include resource mobilization for the Protocol in activities to facilitate the implementation of the strategy for resource mobilization in support of the CBD; and
- Also requests the Executive Secretary to further communicate with the GEF Secretariat in order to discuss the possibility of opening a special financial support window for implementation of the Protocol.

4. Cooperation with Other Organisations, Conventions and Initiatives

COP/MOP Decision: In the decision (UNEP/CBD/BS/COPMOP/6/L.5), the COP/MOP welcomes the Executive Secretary's cooperation with a number of institutions and underlines the contribution of cooperation to the implementation of the Strategic Plan. The COP/MOP requests the Executive Secretary, subject to the availability of funds, to: further pursue cooperation with other organizations, conventions and initiatives with a view to meeting the strategic objective in Focal Area 5 of the Strategic Plan on outreach and cooperation; and continue efforts to gain observer status in those committees of the World Trade Organization that are relevant to biosafety.

5. Budget

COP/MOP Decision: In its decision on the budget (UNEP/CBD/BS/COP-MOP/6/L.17), the COP/MOP approves: a core programme budget of US\$2,922,100 for the year 2013 and US\$2,963,100 for the year 2014; and includes a contingency plan for a provisional budget for 2015 in case COP/MOP 7 takes place in 2015. The COP/MOP further notes with concern and regret that the core programme budget does not contain adequate finance for all activities identified by the parties, including the priorities of developing country parties, resulting in finance for AHTEGs being dependent on voluntary funding, which could have a deleterious effect on capacity building for developing countries. The COP/MOP further agrees to upgrade a post for implementation of the supplementary protocol for the biennium 2015-2016.

6. Capacity Building

COP/MOP Decision: In the decision (UNEP/CBD/BS/COP-MOP/6/L.7), the COP/MOP takes note of the report of the independent evaluation and the working document prepared by the Executive Secretary to facilitate the comprehensive review and possible revision of the 'Action Plan for Building Capacities for the Effective Implementation of the Protocol'; adopts and decides to review the new 'Framework and Action Plan for Capacity Building' in conjunction with the mid-term review of the Strategic Plan of the Protocol; and requests the Executive Secretary to raise awareness of the above Framework and Action Plan and encourage regional stakeholders and donors to play a greater role in supporting its implementation by parties.

The COP/MOP further invites:

- Parties, other governments, and relevant organizations to implement the Framework and Action Plan for capacity building and to share their experiences through the BCH;
- Developed country parties and donors and relevant organizations to take into account the Framework and Action Plan in providing financial and technical support to developing countries, in particular the least developed and small island developing states and countries with economies in transition; and
- The GEF to provide financial support to eligible parties to implement the Framework and Action Plan for Capacity Building.

The COP/MOP further requests the Executive Secretary to prepare reports on the status of implementation of the above Framework and Action Plan. The COP/MOP decides to review the Framework and Action Plan in conjunction with the midterm review of the Strategic Plan of the Protocol and the third assessment and review of the Protocol's effectiveness.

On strategic approaches to capacity building, the COP/MOP: takes note of the analysis of strategic approaches to capacity building prepared by the Executive Secretary; invites parties, other governments and relevant organizations to adopt the strategic approaches to improve the design, delivery, effectiveness, impact and sustainability of biosafety capacity building initiatives; and requests the Executive Secretary to provide, as appropriate, and subject to the availability of funding, technical support to parties to implement the strategic approaches to capacity building.

On coordination mechanisms, the COP/MOP decides to adopt the restructured and streamlined elements in Annex II to the decision; and invites donor countries and agencies and other organizations providing capacity support in biosafety to participate actively in the Coordination Mechanism.

Annex I to the decision outlines objectives of the Framework and Action Plan for Capacity Building for the Effective Implementation of the Protocol, including to:

- Further support the development and implementation of national regulatory and administrative systems;
- Enable parties to evaluate, apply, share and carry out risk assessments; develop capacity for HTPI of LMOs;

- Assist parties to the Protocol to establish and apply rules and procedures on liability and redress from transboundary movements of LMOs;
- Enhance capacity to facilitate public awareness and promote education on safe transfer, handling and use of LMOs; and
- Ensure that BCH is easily accessed by all stakeholders.

Annex II, on Coordination Mechanisms for Capacity-Building Efforts under the Protocol, outlines guiding principles, elements and administration of the coordination mechanism.

7. Roster of Experts

COP/MOP Decision: In the decision (UNEP/CBD/BS/COPMOP/6/L.8), the COP/MOP:

- Reiterates its earlier call to parties and other governments that have not yet done so to nominate experts to the roster;
- Adopts the revised nomination form for the roster of experts and authorizes the Executive Secretary to update the form based on operational experience;
- Decides to expand the mandate of the roster of experts to include supporting, as appropriate and upon request, the work of the Secretariat, the COP/MOP and other bodies under the Protocol, in relation to capacity building for developing countries and countries with economies in transition; and
- Invites parties and other governments to consider nominating experts on the roster to serve on the AHTEGs, informal advisory committees and other relevant bodies under the Protocol.

The COP/MOP also: invites parties, other governments, relevant organizations and the Executive Secretary to consider using experts on the roster as resource persons for capacity building activities; and reiterates its invitation to developed country parties and other donors to make contributions to the voluntary fund.

8. Handling, Transport, Packaging and Identification (HTPI)

- HTPI for LMOs destined for Contained Use or for Intentional Release
- HTPI Standards

HTPI of LMOs was discussed in WG I on Monday, Wednesday and Thursday. Delegates initially discussed two sub-items: HTPI for LMOs destined for contained use or for intentional release (UNEP/CBD/BS/COP-MOP/6/8 and INF/7); and HTPI standards (UNEP/CBD/BS/COP-MOP/6/9 and INF/24). They eventually agreed to address both items in a single decision.

COP/MOP Decision: In the decision on HTPI of LMOs (UNEP/CBD/BS/COP-MOP/6/L.12), the COP/MOP notes ongoing cooperation between the CBD Secretariat and international organizations whose work is relevant for HTPI of LMOs and, *inter alia*: requests parties and encourages other governments to continue to implement the requirements of Article 18(2)(b) and (c) and related decisions through the use of a commercial invoice or other documents required or utilized by existing documentation systems, or documentation required by domestic regulatory and/or administrative frameworks; requests the Executive Secretary to include a specific question in the third national report inquiring whether parties require the use of existing documents or stand-alone documents or both; and encourages the OECD to renew efforts to develop unique identification systems for living modified micro-organisms and animals.

The COP/MOP also requests the Executive Secretary to further examine the potential gaps and inconsistencies in HTPI standards and provide recommendations to COP/MOP 7, as appropriate.

9. Notification Requirements

COP/MOP Decision: In its decision (UNEP/CBD/BS/COPMOP/6/L.11), the COP/MOP:

- Requests parties to address gaps in domestic implementation of notification requirements related to intentional transboundary movements of LMOs;
- Decides that further review of notification requirements should only take place if there is a documented need, as indicated through national reports or other submissions;
- Invites parties, other governments and relevant organizations to consider using the LMO quick-link tool by their relevant national authorities where reference is made to a living modified organism; and
- Encourages sharing, through the BCH, best practices and experiences on implementing requirements.

10. Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress

COP/MOP Decision: In its decision (UNEP/CBD/BS/COPMOP/6/L.3), the COP/MOP:

- Calls on parties to expedite internal processes for ratification, approval, acceptance of or accession to the Supplementary Protocol;
- Calls on parties to the CBD that are not parties to the Protocol to take relevant steps to become parties so they may also become parties to the Supplementary Protocol;
- Invites parties to identify capacity-building needs and establish national priorities to implement and apply provisions of the Supplementary Protocol;
- Invites parties and relevant organizations to make financial resources available for awareness-raising, experience-sharing, and capacity-building activities to expedite entry into force and implementation; and
- Requests the Executive Secretary to encourage UNEP and the International Union for the Conservation of Nature (IUCN) to develop an explanatory guide.

11. Unintentional Transboundary Movements and Emergency Measures

COP/MOP Decision: In its decision (UNEP/CBD/BS/COP-MOP/6/L.13), the COP/MOP:

In its decision, COP-MOP encourages parties to use as guidance in their implementation of Article 17, decisions that have been or may be taken relating to Protocol Article 18 (HTPI), and the guidance on risk assessment of LMOs developed by the AHTEG; and urges parties to make relevant details of their point of contact for receiving notifications available, establish and maintain measures to prevent unilateral transboundary movement of LMOs, and establish mechanisms for emergency measures. The COP/MOP further requests parties and invites governments and relevant organizations to provide views and information to the Executive Secretary six months prior to COP/MOP 7 on any challenges and experiences relating to the implementation of Article 17; and requests the Executive Secretary to prepare a synthesis of the views.

12. Risk Assessment and Risk Management

COP/MOP Decision: In the decision (UNEP/CBD/BS/COPMOP/6/L.16), the COP/MOP, *inter alia*, states that the guidance is not prescriptive and does not impose

any obligations on parties and that the guidance will be tested nationally and regionally for further improvement. It further encourages parties, governments and other organizations to translate the guidance and provide financial and technical assistance to developing country parties and parties with economies in transition to test the guidance.

The COP/MOP requests the Executive Secretary to: develop appropriate tools to structure and focus testing of the guidance; gather and analyze feedback from the testing; and provide a report on possible improvements to the guidance. It also establishes a mechanism for regularly updating the list of background documents to the guidance and extends the open ended online forum renewable every four years.

The COP/MOP further decides to bring to a close the current AHTEG and establishes a new AHTEG to serve until COP/MOP 7. The terms of reference for the AHTEG are attached as an annex to the decision. It requests the Executive Secretary to select experts for the new AHTEG, in consultation with the COP/MOP Bureau.

On capacity building, the COP/MOP requests the Executive Secretary to, *inter alia*: convene the remaining training courses on risk assessment for the African and CEE sub-regions; follow up on training by gathering additional feedback from parties on the practicality, usefulness and utility of the guidance; and conduct workshops on risk assessment and risk management at international, regional and/or sub-regional levels.

On the identification of LMOs or specific traits that may have or are not likely to have adverse effects on the conservation and sustainable use of biological diversity, the COP/MOP requests the Executive Secretary to create sections in the BCH where such information can be submitted and easily retrieved and invites parties to provide the Executive Secretary with scientific information that may assist in the identification of LMOs that may have or are likely to have adverse effects on the conservation and sustainable use of biodiversity.

On the status of implementation of risk assessment and risk management provisions, the COP/MOP requests the Executive Secretary to conduct an online survey on the status of the implementation of Operational Objectives 1.3, 1.4 and 2.2 of the Strategic Plan with a view to establishing baselines for, and collecting data on, the indicators concerned.

13. *Subsidiary Bodies*

COP/MOP Decision: In the decision on subsidiary bodies (UNEP/CBD/BS/COP-MOP/6/L.4), the COP/MOP decides:

That at this stage there is no need to establish an open-ended subsidiary body for scientific and technical advice; to continue establishing AHTEGs with specific mandates, as needed and subject to the availability of funds; to take into account experience and lessons learned from previous AHTEGs, including the use of open-ended online expert forums; and to consider the need to establish a permanent subsidiary body at COP/MOP 8.

14. *Socio-Economic Considerations*

COP/MOP Decision: In its decision (UNEP/CBD/BS/COP-MOP/6/L.15), the COP/MOP encourages parties, other governments and relevant organizations to: conduct research on the socio-economic impact of LMOs to fill knowledge gaps and identify specific socio-economic issues, including those with positive impacts; share and exchange information on research and experience via the BCH; and build domestic capacity in socio-economic analysis of LMO impacts by engaging local institutions of higher education.

The COP/MOP further requests the Executive Secretary to compile, take stock of and review information on socioeconomic considerations arising from the impact of LMOs on the conservation and sustainable use of biodiversity, on the basis of: existing situational frameworks, legislation and policies with provisions on socio-economic considerations; capacity-building activities relating to biosafety and socioeconomic considerations; existing expertise and experience; and other policy initiatives concerning social and economic impact assessments.

The COP/MOP further decides to establish an AHTEG, subject to availability of funds, and according to the terms of reference contained in the annex to the decision. The AHTEG will develop conceptual clarity, drawing on the outcomes of: stocktaking and review by the Executive Secretary of information on socio-economic impacts of LMOs on the conservation and sustainable use of biodiversity; and online discussion groups and regional online conferences to facilitate and synthesize the exchange of views, information and experiences among

parties, governments, organizations, and indigenous and local communities. According to the annex, the AHTEG will be composed of: a minimum of five and maximum of eight experts per region, depending on funding, and nominated by parties, while maintaining a regional balance; and at least five, but no more than ten observer participants representing nonparties, UN organizations/agencies, relevant organizations, and indigenous and local communities.

15. *Monitoring and Reporting*

COP/MOP Decision: In its decision (UNEP/CBD/BS/COP-MOP/6/L.9), the COP/MOP: welcomes the high rate of submission of second national reports by parties and takes note of the analysis of responses prepared by the Secretariat; reminds parties of their obligation to submit national reports, urging those who have not done so to submit their reports and answers to all mandatory questions; and further reminds parties to make the required information available to the BCH.

The COP/MOP further requests the Secretariat to:

- Assess, on the basis of the second national reports, the discrepancies and/or gaps in information made available by parties through the BCH;
- Assist parties to submit through the BCH the updated information contained in their reports; update the reporting format, taking into account the experiences gained from analyzing the second national reports, the recommendations of the Compliance Committee and feedback from parties; and to submit the revised format to COP/MOP 7.

16. *Assessment and Review*

COP/MOP Decision: In its decision (UNEP/CBD/BS/COPMOP/6/L.10), the COP/MOP: notes the information contained in the second national reports and the analysis undertaken on the status of implementation of core elements of the Protocol; and decides that the data and information contained in the analysis shall form the baseline for measuring progress in implementing the Protocol.

The COP/MOP further requests the Executive Secretary to:

- Undertake a dedicated survey to gather information corresponding to indicators in the Strategic Plan that could not be obtained from the second national reports or through other existing mechanisms;

- Review the information gathered through the survey and make the results available to the parties prior to COP/MOP 7;
- Commission a consultant, subject to availability of funds, to develop a sound methodological approach for the third assessment and review of the effectiveness of the Protocol; and
- Provide parties with an opportunity to submit views on the methodological approach, review it in light of the views provided, and submit a proposal for consideration by COP/MOP 7.

The COP/MOP also requests the Compliance Committee, in light of the conclusions and recommendations of the AHTEG on the Second Assessment and Review of the Protocol, to evaluate the status of implementation of the Protocol as a contribution to the third evaluation of effectiveness in meeting the objectives of the Protocol; and decides that in the process of preparing for the third assessment and review of the Protocol, the experiences of the parties in complying with the Protocol shall be taken into account.

17. Closing Plenary

The Plenary adopted 15 decisions and the reports of the working groups (UNEP/CBD/BS/COP-MOP/6/L.1/Add.1 and Add.2). This was preceded by a prolonged discussion on some outstanding issues, under the Agenda items on unintentional transboundary movements and the financial mechanism and resources, to explore developing consensus over them.

Session was then suspended to allow for the finalization of the decision on the budget and reconvened at 8:30 pm. Conrod Hunte, Chair of the budget contact group, introduced the 2013-2014 draft budget decision (UNEP/CBD/BS/COP-MOP/6/L.17).

He expressed concern that priority issues expressed by Parties could not be included in the core budget, with both AHTEGs (Risk Assessment, and Socio-economic Considerations), being subject to voluntary contributions. Decision on budget was adopted without amendment. Delegates then adopted the COP/MOP 6 report (UNEP/CBD/BS/COP-MOP/6/L.1).

Progress made on risk assessment and socio-economic considerations received appreciation and Bolivia proposed to host the first AHTEG on this subject. Need for capacity building was again highlighted by the Asia-Pacific and the CEE groups. GRULAC urged retaining the two-year periodicity for COP/MOPs to fulfill the objective of the Strategic Plan.

The Republic of Korea announced its intention to host COP/MOP 7 and CBD COP 12. New Zealand offered a formal tribute to the Government and people of India, which the plenary endorsed by acclamation. Japan, recalling the challenges of the Presidency, asked delegates to support India over the next two years.

COP-MOP Chairman Shri M.F. Farooqui thanked the delegates for their positive attitude and spirit of compromise, announced financial pledges made by Norway and the Republic of Korea for the AHTEG on socioeconomic considerations, and formally closed the sixth meeting.

V) Overview

A. Progress of Implementation

The CPB is the only international agreement dealing exclusively with products of modern biotechnology. Interpretations of its Articles and their implementation have a significant bearing on biosafety regulations in both developed and developing countries. It is noteworthy that products from new technologies are assessed keeping in view the precautionary principle allowing countries to ban imports of an LMO if they feel that there is not enough scientific evidence showing that the product is safe. Its provisions also require the exporters to label clearly their shipments that contain genetically engineered commodities like maize or cotton.

At the first glance, it seems that implementation is doing well as gleaned from the fact that the Protocol now has 164 Parties and the submission rate of the national reports, which is one among the measures of implementation, reached an outstanding level of nearly 90 %. A closer look, however, reveals that the progress has been admittedly slow so far and severe differences among Parties still persist. COP-MOP-5, held at Nagoya in 2010, had adopted the 10-year Strategic Plan

(2011-2020) to speed up the Protocol's implementation and it is now left to India, as the president, to take the process forward.

All Parties are required to enact enabling legislations and to adopt necessary regulatory framework and procedures in support of the mandated safety measures. However, only about half of the Parties have actually implemented the core provisions of the Protocol by establishing an advance informed agreement procedure and supportive national biosafety frameworks.

It is noteworthy that only a small number of countries seem to be totally opposing the introduction of LMOs while more and more countries are now trying to differentiate between LMOs that they want to develop themselves and LMOs that they approve for import. As more countries become LMO exporters, their decisions regarding transboundary movements of LMOs may become inherently more complex, requiring the design of national biosafety frameworks that balance the interests of both the importers and exporters. Whereas importer countries seek to protect the environment and the biodiversity against risks associated with LMO-shipments, exporters are more concerned with creating least disruption to their international trade. During this COP-MOP meeting, demarcation of frontlines between interests of exporters and importers somehow appeared to be less pronounced on many issues.

Going by the example of HTPI, a highly contested issue of long standing, especially with regard to the documentation requirements for LMOs destined for contained use, and LMOs for intentional introduction into the environment, COP/MOP 6 was to review the use of these requirements and take necessary measures to boost implementation of Article 18. Importing countries used to take a strong stance on this issue as they see documentation as a primary means to take informed decisions with regard to LMO imports. However, rather than repeating the traditional face-off between importers and exporters, COP/MOP 6 delegates chose to neutralize most of the potentially controversial references by either deleting them or opting for flexible language. Prominent examples are the deletion of reference to the UN Model Recommendations on the Transport of Dangerous Goods, or the move away from a requirement to use stand-alone documentation, rather than existing documents such as a commercial invoice. In both cases, delegates shied away from the task of crafting finely balanced language that would have been needed to make substantive progress

towards implementation. Similar trends were observed in the discussions on notification requirements or unintended transboundary movements and emergency measures, where the decision text shrunk at an impressive speed as delegates decided to review the issue only when a problem has emerged.

Regarding the likely reasons for slow implementation, many developing countries are still faced with insufficient technical human resources, lack of institutional capacities, inadequate awareness at all levels, irrational protests and, above all, little mobilization of the required funding. These challenges have to be met in case the pace of implementation is to make a real headway.

B. Risk Assessment and Socio-Economic Considerations

Following hard negotiations, key decisions on socio-economic considerations and risk assessment were adopted, advancing thereby the work of the Protocol on these issues.

Risk assessment and risk management are the core provisions of the Cartagena Protocol and discussions on them during this meeting, in particular the revised guidance on risk management, made good progress. It was felt that since it had gone through an extensive review through online forums and the reconvened AHTEG on risk assessment, the guidance should be sufficiently mature to be endorsed by the COP/MOP as an essential resource for developing nationally adapted risk assessment approaches. However, despite numerous references emphasizing the voluntary nature of the guidance and the broad understanding that it would be tested and further revised, delegates did not agree to endorse and operationalize the guidance. Instead, they decided to call for another round of improvements by requesting a structured approach to testing and subsequent revision. While this was generally welcomed, some believed that the discussion missed out on a key point. For developed countries the guidance is a reference point in further development of their own nationally adapted approaches to risk assessment. They felt that the current guidance was too detailed and restrictive. Developing countries, on the other hand, need the guidance as a tool to start conducting risk assessments in the first place. Rather than addressing these different needs, which could have led to a more differentiated approach, the discussions focused on finding language that would satisfy all parties.

Surprisingly there was a welcome breakthrough on the issue of socio-economic considerations. This issue had long been blocked by bitter debates among those who felt that broader socio-economic considerations should not be dealt with under the Protocol, because it is limited to transboundary movements; and participants who wanted to address potential negative socio-economic impacts of LMOs, such as the loss of agricultural varieties that have cultural value. As a result, work on socio-economic considerations has been slow and mostly limited to compiling information, until a workshop on socio-economic considerations was hosted by India during the Inter-sessional period. COP/MOP 6 was able to achieve broad consensus that socio-economic considerations require substantive engagement.

The decision on socio-economic considerations established an AHTEG to develop conceptual clarity on what constitutes socio-economic considerations under Article 26 and submit its report to COP-MOP-7 in 2014. This issue had remained highly contested earlier as the developing Parties wanted to retain their right to take socio-economic considerations into account when taking a decision on importing LMOs while some developed countries opposed this by giving it low priority. Building consensus on the need to establish an AHTEG to conduct this basic work led to a reference to Operational Objective 1.7 of the Strategic Plan, which mentions the development of “guidance” or “guidelines” as a possible outcome. Many hailed this reference as the real breakthrough of the meeting since it provided a clear objective for future work on socio-economic considerations and raised its status from that of a perpetual stalemate to an actual issue for outcome-oriented deliberations. The substantive debate will, however, still face some hurdles such as drawing a line between any type of socio-economic impact of LMOs and those impacts associated with damage to the conservation and sustainable use of biodiversity that can be rightfully considered under the Protocol’s scope. However, by taking the first step at this COP-MOP, socio-economic considerations issue has now been firmly established as one of the main substantive issues to be developed at future COP/MOPs.

C. Refocusing on implementation

Being the first COP-MOP meeting after the adoption of the Nagoya-Kuala Lumpur Protocol on Liability and Redress, it faced somehow a cautious attitude of Parties delaying the rapid entry into force of this Supplementary Protocol that was intended mainly as an instrument to

protect the importing countries. The on-going negotiations revealed that the influence of importers is waning since many formerly importing countries have now become exporters and LMOs. Some exporting countries have made it clear, however, that they won’t accept any reference to the Supplementary Protocol, for example suggesting it as guidance in the context of other decisions, before its entry into force. These statements may indicate that these countries have little interest in ratifying the Supplementary Protocol. Nevertheless, adoption of this Supplementary Protocol has provided some breathing space to focus on new issues, with socioeconomic considerations poised to move into the spotlight. COP/MOP 6 also made it clear that there is a need now to refocus on Protocol’s implementation. The risks associated with LMOs may concern low probability events that could potentially create irreversible and long-term damage to biodiversity. The objective of the precautionary approach and the Protocol is precisely to avoid such risks from materializing by putting into place adequate procedures and regulatory frameworks.

VI) The Indian Scene

The COP-MOP-6 was an opportunity for the global community to discuss many of the biosafety aspects and pave the way for a more robust science-based regime across all countries in the world, including both the providers as well as users of the modern biotechnology products. It was also a testimony to India’s growing stature as it took over the COP-MOP presidency from Japan. It was reflected in the way socio-economic considerations and livelihood issues were brought forward and moved ahead in the meeting’s agenda.

It is commendable that India has notably moved forward and the private sector is now collaborating with the Government in an effort to ensure that we have one of the best science-based regulatory mechanisms in operation. In this context, India is set to enact the Biotechnology Regulatory Authority of India Bill to set up a statutory and autonomous agency with adequate powers to regulate research, import, develop, transport and use of biotechnology products with a view to maintaining safety standards. At present, there is a moratorium imposed in India on Bt brinjal (eggplant/ aubergine), imposed in February 2010, but open field trials for other GM food crops are permitted after they are approved by the Genetic Engineering Appraisal Committee subject, however, to final say by the individual states within the country.

A technical expert committee, appointed by the Supreme Court of India to advise upon open field trials for GM crops, has recently (18 October 2012) recommended a ten-year moratorium on all Bt food crops' field trials and has asked the regulatory authorities to revisit regulations afresh so as to ensure that the GM crops pose no risk to human health or the environment. This report highlights the need for specifically designated and certified field trial sites and focuses upon sufficient mechanisms to be established for monitoring trials before conducting further field trials. This recommendation is, however, yet to be considered by the Supreme Court for its acceptance. This new report apparently contradicts the advice from the Prime Minister's scientific advisory council on biotechnology and agriculture, which met on 09 October 2012, but it is in line with another report, submitted in August 2012, by a parliamentary committee on the cultivation of GM crops, recommending that GM crops' field trials should be discontinued and research on GM crops be conducted under strict regulation. Notwithstanding the urgent need for Indian evaluation of the safety status of GM crops to be of the desired standards, this experience reveals the need for convergence in policy and administrative decisions, considering that the progress of on-going research efforts may suffer for want of clear policy guidance on this subject.

VII The Way Ahead

Ensuring biosafety, when LMOs are released, is the responsibility of national governments. Problems often arise within a country when several ministries, with radically different priorities, are involved in regulation. CPB being a multilateral environment agreement, Ministry of Environment & Forests is the nodal implementing agency although the major beneficiaries may be the Ministries of Agriculture and Health. It has now become increasingly clear that the biosafety mainstay in developing countries is the precautionary principle that empowers Member States to permit imports, impose restrictions, or refuse imports even when there is no conclusive evidence of risk.

Transboundary movement of agricultural products containing LMOs is henceforth likely to be regulated through BCH, an internet-based information system. All LMOs, approved at the national level as food or feed, and which are registered with the BCH, may be exported to other Member States provided the importing country has not already imposed any restrictions based on its legal regulatory framework. Capacity building, creating awareness, clarity in procedures and transparency in decision making continue to remain the key elements in implementing this Protocol.

Use of Heuristic Approach for the Development of a Core Set from Large Germplasm Collection of Foxtail Millet (*Setaria italica* L.)

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Foxtail millet, (*Setaria italica* L.) is an ancient crop and important minor cereal. It has a long history of cultivation in India and possesses rich genetic diversity conserved in genebanks. Large germplasm collection in genebanks limit accessibility of this crop in crop breeding. It is, therefore, important to develop core sets of germplasm from large collection for conserving maximum diversity of this crop. “PowerCore” is a new approach that simplifies the generation process of a core set by significantly cutting down the number of core entries but maintaining maximum diversity. Here, we have developed a core set from 1,482 accessions of foxtail millet using data of 25 quantitative and qualitative descriptors. The newly formed core set has 59 accessions which represents 4% of the total collection. This heuristic method has proved to be an efficient tool for developing core sets from collections of unequal diversity and differentiation

Key Words: Characterization, Core collection, Foxtail millet, Minor cereal, PowerCore

Introduction

Foxtail millet [*Setaria italica* (L.) Beauv.], a member of tribe Panicoideae, is one of the oldest crops cultivated for hay, pasture and food grain. It is an important crop in China besides India, Japan and other countries in Asia and Europe. It is also grown in North and South America, Australia and Africa as a grass. It was grown fairly extensively with its cultivation extending from temperate Eurasia to tropics and sub-tropics of Asia. Presently, in India, the crop is cultivated on a very limited area in sporadic patches in many states throughout the country. Known for its drought tolerance, foxtail millet can withstand severe moisture stress and can adjust to wide range of soil condition. At the All India Coordinated Small Millet Improvement Programme (AICSMIP) at Bangalore, India, the foxtail millet collections exceeding around 1,500 accessions have been assembled and maintained, there represent good diversity from various regions within and outside the country.

Increasing size of germplasm collection in most genebanks, limits their accessibility for use in crop breeding and their subsequent management. In addition, redundant resources have become an obstacle to the

effective management and utilization of these resources. Therefore, it has been proposed that a limited set of accessions be selected capturing as much genetic diversity as possible to offer a good starting point when searching for new traits and could be used for in-depth evaluation, thus increasing the knowledge of the entire collection (Knüpffer and van Hintum, 1995). Core collections improve the management and effective use of plant genetic resources (Brown, 1989). A core collection is a subset of a large germplasm collection that contains chosen accessions capturing most of the genetic variability of the whole genebank. Such a core subset provides a proper working collection for the extensive searching of desired alleles and a point of entry to the entire germplasm collection (Holbrook and Anderson, 1995; Dussert *et al.*, 1997).

Since core sets are derived from the wide spectrum of genetic diversity of the whole collection, most of this diversity is expected to be retained. The sampling strategies for choosing core entries can be divided into two approaches, simple random sampling and stratified random sampling (Brown, 1989a and 1989b; Spagnoletti Zeuli and Qualset, 1993). The most common method employed in obtaining a core collection of desired size is

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that of stratified random sampling (Peeters and Martinelli, 1989; Chandra et al., 2002). Several strategies have also been suggested for determining the appropriate sampling fraction from each group or strata. These methods include proportional, log frequency and constant allocations (Brown, 1989a; Spagnoletti Zeuli and Qualset, 1993).

Though many of these methods have been used successfully in obtaining core sets of desired size, inequality in diversity of accessions and differentiation of stored accessions has been a recurring problem that skews the subset population. This problem has led to several attempts to build germplasm core collections through the maximization of allelic or phenotypic richness. More recently, the Rural Development Administration (RDA), South Korea, has used the advanced M (maximum) strategy with a heuristic search for establishing core sets and accordingly a programme known as “PowerCore” (<http://genebank.rda.go.kr/powercore/>) has been developed which has been used in the present study. In this paper, we discuss the development of a core set from a large germplasm collection of 1,482 accessions of foxtail millet using Heuristic Core Collection (HCC) program. The HCC program employs the advanced M-strategy using a modified heuristic algorithm. It creates subsets representing all alleles or observations classes, with the least allelic redundancy, and ensures a highly reproducible list of entries. Further, the heuristic strategy is compared with cluster analysis using Ward’s method (Ward, 1963). This approach has already been used in developing core set from large rice collection (Chung et al., 2009).

Thus, in establishing a good core collection it is very important to choose best sampling strategy that represents largest diversity of the entire collection. Earlier cluster analysis has been widely used as an important tool to group accessions for constructing core collection. Recent HCC programme 9 uses the advanced M-strategy. This modified heuristic algorithm can be applied for the selection of genotype data (allelic richness), the reduction of redundancy and the development of a more extensive analysis in the management and utilization of large collection of plant genetic resources.

Materials and Methods

The Indian foxtail millet germplasm collection used for developing core subset consisted of 1,482 accessions. All accessions were planted during *kharif* season (July–November) in year 2001 in the experimental field at

the Main Research Station, GKVK, Bangalore, which is situated at 13°N latitude and 77°35'E longitude at an altitude of 890 m. The average annual rainfall is around 850 mm; which is mostly received during July to October. The soil of the experimental field was red sandy loam with an acidic pH of 5.5. The soil is low in organic carbon (0.4%) with moderate availability of nitrogen (300 kg/ha) and phosphorus (185 kg/ha) and fairly rich in potash (225 kg/ha). The material was planted using an Augmented Block Design (Federer, 1956) using three standard checks.

The passport information for all the 1,482 accessions was published in a catalogue on “Evaluation of foxtail millet (*Setaria italica*) germplasm (Gowda et al., 2002)”. The germplasm collections represent diversity from 15 states of India which includes: Andhra Pradesh (164), Bihar (57), Gujarat (11), Himachal Pradesh (8), Jammu and Kashmir (12), Karnataka (139), Kerala (7), Madhya Pradesh (34), Maharashtra (3), Orissa (6), Punjab (10), Rajasthan (2), Tamil Nadu (49), Uttar Pradesh (505) and West Bengal (25). In addition to diversity from India, these collection also include exotic diversity from Africa (8), Bangladesh (3), China (25), Pakistan (1), Turkey (91), USA (39) and USSR (2). There were 373 accessions for which the passport information was not known.

All the accessions were characterised for 25 important traits as outlined by IPGRI (1985). Eleven traits were measured on a quantitative scale including days to 50% flowering, plant height, of basal tillers, flag leaf length, flag leaf width, peduncle length, ear length, panicle exertion, days to maturity, grain yield per plant and 1,000-grain weight. Fourteen qualitative characters included plant pigmentation at flowering, leaf colour, blade pubescence, sheath pubescence, degree of lodging at maturity, senescence, inflorescence lobes, inflorescence bristles, lobe compactness, inflorescence shape, inflorescence compactness, fruit colour, grain shape and apical sterility in panicle. The accession level information has been published by the All India Coordinated Small Millet Improvement Programme (AICSMIP), Bangalore, India (Gowda et al., 2002).

“Power Core” was used for developing core collection, which is a program that applies the advanced M-strategy with a heuristic search for establishing core or allele mining sets and thus possesses the power to represent all alleles or classes. It effectively simplifies the generation process of a core set while significantly

cutting down the number of core entries, maintaining 100% of the diversity as categorical variables. The validation of core sets using “PowerCore” has also been compared with the core developed using the traditional clustering method for the same number of accessions using Wards clustering method (Ward, 1963) and the distance used was Euclidean.

Core collections are considered to well represent the genetic diversity of the initial collection if the following two criteria are met: (1) no more than 20% of the traits had different means (significant at $\alpha = 0.05$) between the core collection and the initial collection and (2) Coincidence Rate (CR) was retained by the core collection in no less than 80% of the traits (Hu *et al.*, 2000). The design concept and implementation strategy of ‘PowerCore’ and the validation on the outcome in comparison with other methods has been well described by (Kim *et al.*, 2007). “PowerCore” by default classified the continuous variables into different categories based on Sturges rule (Sturges, 1926), which is described as: $K = 1 + \log_2 n$, where n = number of observed accessions. However, the software also allows modification of this rule to make desired number of classes for the continuous variables. Once classification of the continuous variables is performed, the software takes into account all classes, without omission of any of its variables. It thus, possesses the capability to cover all the distribution ranges of each class.

Results and Discussion

“PowerCore” successfully selected 59 accessions (Table 1) from the entire collection of 1,482 accessions, an even distribution of characters was observed in the core set when classified by this method. Most of these core collections were from Uttar Pradesh (28) followed by

5 accessions each from Andhra Pradesh and Karnataka, thereby indicating that the collections from Uttar Pradesh have more diversity than these from other parts of the country. In the present study, maximum possible number of classes for each variable was used in order to capture 100% diversity for each unique class in a core set. The core size was from 4% of the total collection. For the validation, the following statistical parameters were analysed to compare the mean and variance ratio between core and entire collections and are presented in Table 2.

Following four statistical parameters were analysed using “PowerCore” to compare the mean and variance ratio between core and entire collections. The percentage of the significant difference between the core sets and the entire collection was calculated for the mean difference percentage (MD, %) and the variance difference percentage (VD, %) of traits. Coincidence rate (CR, %) and variable range (VR, %) were designed to evaluate the properties of the core set against the entire collection (Hu *et al.*, 2000).

Mean Difference Percentage (MD %) – which is estimated as:

$$MD (\%) = \frac{1}{m} \sum_{j=1}^m \frac{Me - Mc}{Mc} \times 100$$

Where, M_e = Mean of entire collection; M_c = Mean of core collection, and m = number of traits.

Variance Difference (VD %) – which is estimated as:

$$VD (\%) = \frac{1}{m} \sum_{j=1}^m \frac{Ve - Vc}{Vc} \times 100$$

Where, V_e = Variance of entire collection, V_c = Variance of core collection, and m = number of traits.

Confidence ratio (CR %) – which is estimated as:

$$CR (\%) = \frac{1}{m} \sum_{j=1}^m \frac{Rc}{Re} \times 100$$

Where, R_e = Range of entire collection, R_c = Range of core collection, and m = number of traits.

CR% indicates whether the distribution ranges of each variable in the core set are well represented when

Table 1. List of core accessions identified based on 25 variables

GS20	GS374	GS820	GS1315	GS1954
GS27	GS401	GS848	GS1372	GS2025
GS77	GS545	GS872	GS1377	GS2029
GS78	GS617	GS887	GS1388	GS2035
GS84	GS639	GS889	GS1404	GS2040
GS105	GS678	GS890	GS1430	GS2076
GS160	GS717	GS892	GS1500	GS2096
GS260	GS736	GS900	GS1507	GS2100
GS275	GS739	GS958	GS1618	GS2164
GS338	GS764	GS963	GS1657	GS2248
GS364	GS766	GS1242	GS1794	GS2258
GS372	GS809	GS1308	GS1929	

Table 2. Comparative description of mean and variance components between entire and core accessions developed using “PowerCore” and clustering approach

Statistical parameters	Days to 50 percent flowering			Plant height (cm)			No. of basal tillers			Flag leaf length (cm)		
	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)
No. of accessions	1482	59	288	1482	59	288	1482	59	288	1482	59	288
Mean	49.25	48.51	49.38	142.40	133.42	142.56	3.91	4.03	3.89	27.68	29.51	27.94
Minimum	33	35	33	52.20	52.20	91.40	1	1	1	16	17	16
Maximum	69	69	69	184	174	184	12.20	12	12.20	47.50	45.25	47.50
Range	36	34	36	131.80	121.80	92.60	11.20	11	11.20	31.50	28.25	31.50
CV%	7.57	12.57	9.05	10.39	18.67	9.86	36.18	57.87	43.70	19.91	23.91	22.73

Statistical parameters	Flag leaf width (cm)			Peduncle length (cm)			Ear length (cm)			Panicle exertion		
	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)
Number of accessions	1482	59	288	1482	59	288	1482	59	288	1482	59	288
Mean	1.51	1.66	1.52	28.05	29.91	28.04	14.76	15	14.92	13.31	13.86	13.23
Minimum	0.78	0.85	0.85	13.40	15.25	13.40	3.60	3.60	3.60	1.50	2.70	1.50
Maximum	4.40	4.40	3.50	56.50	53	56.50	24.50	23.80	24.50	29	29	29
Range	3.62	3.55	2.65	43.10	37.75	43.10	20.90	20.20	20.90	27.5	26.3	27.50
CV%	18.57	35.64	19.08	15.81	25.59	19.37	16.57	21.07	20.71	24.89	36.38	29.78

Statistical parameters	Days to maturity			Grain yield per plant (g)			Thousand grain weight (g)		
	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)	Entire	Core (PowerCore)	Core (Cluster approach)
Number of accessions	1482	59	288	1482	59	288	1482	59	288
Mean	90.79	88.88	90.93	9.74	10.29	10.06	3.06	3.05	3.02
Minimum	72	72	72.00	2.10	2.10	2.10	1.90	2	2
Maximum	110	110	110	23.80	23.80	20.20	4	3.90	4
Range	38	38	38	21.70	21.70	18.10	2.10	1.90	2
CV%	7.40	8.93	7.81	39.98	43	42.25	11.86	14.30	12.91

compared to the entire collection.

Variable Rate (VR %) – which is estimated as:

$$CR (%) = \frac{1}{m} \sum_{j=1}^m \frac{R_c}{R_e} \times 100$$

Where, CV_e = Coefficient of variation of entire collection, CV_c = Coefficient of variation of core collection, and m = number of traits. VR% allows a comparison between the coefficient of variation values existing in the core collections and the entire collections and determines how well it is being represented in the core sets.

The results showed that there was no significant difference ($\alpha=0.05$) for the means of all traits between core and entire collections. The estimated values for MD% was -2.2, which indicated that there is no difference in the mean values of entire and core collections. VD% was estimated to be 30.56, the VD values indicated that the variance for the entire and the core populations are not the same. The CR% obtained was 94.83, which indicated that the core has captured all accessions from all the classes and, thus, is a representative of the entire collection. High VR % (71.04) indicated that the coefficient of variation in the core set is higher compared to entire collections for all the variables.

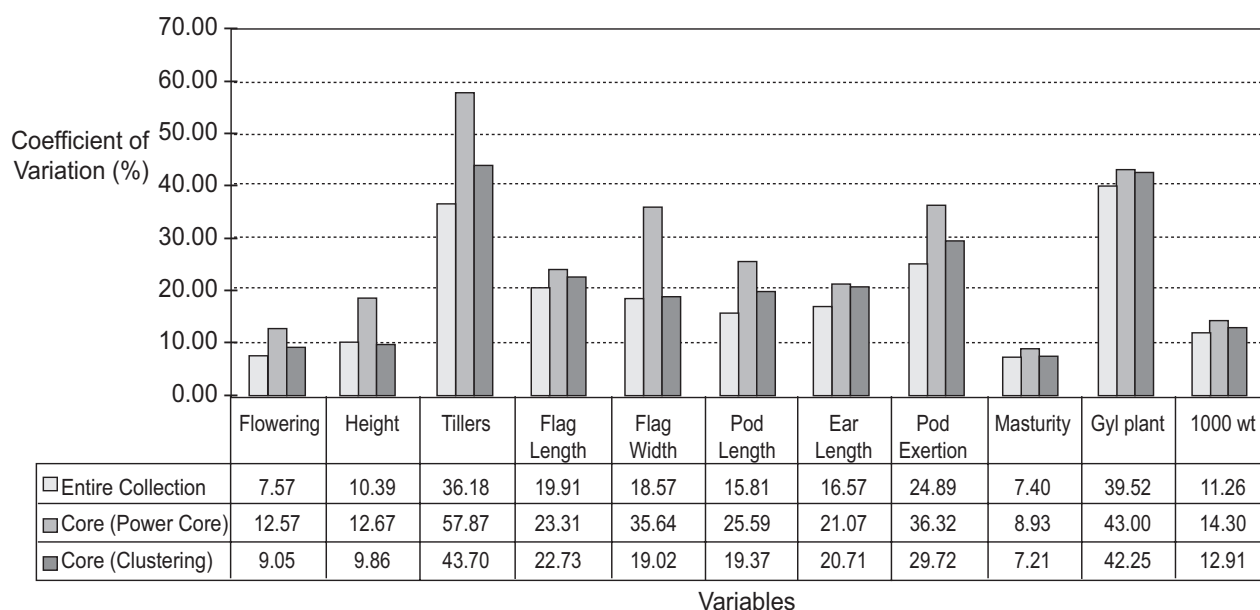


Fig. 1. Comparison of CV between entire collection, core collection developed using PowerCore and clustering approach (Wards method) for quantitative variables

In order to compare the efficiency of “PowerCore” for developing core collection over other clustering method, mean and statistical parameters for entire population, core developed using “PowerCore” and core developed using clustering method were compared. This indicated that the mean components (mean, minimum, maximum and range) are the same for the two core sets developed, except for plant height where, core developed by clustering method does not include all the classes (Table 2). The only difference between these two core sets was that the “PowerCore” core was based on only 4% of the entire population, whereas, the clustering core was based on 15.64% of entire collection. The variance in core developed using clustering method was either close to the entire collections or was higher for all the descriptors, but was never higher to the core developed using “PowerCore”. The Coefficient of Variation was also either close or higher to the entire collection for all descriptors for core developed based on clustering method, but never higher to core developed using “PowerCore” for these descriptors. Histogram comparing CV for the entire and core sets is shown in Fig. 1. High value obtained for CR % (94.83) suggests that the core attained using the heuristic approach method could be adopted as a representative of the whole collection.

A comparison of Shannon-Weiner (Shannon and Weaver, 1949) diversity index for the entire collection, core developed using “PowerCore” and core developed

using clustering method also indicated a high diversity for all the qualitative traits in core developed using “PowerCore” compared to core developed using clustering approach, except for a few variables, where it was observed at par (Table 3).

Table 3. Comparison of Shannon-weiner diversity index values for entire collection, core collection developed using “PowerCore” and clustering method (Wards method)

Variables	Core collections (powercore)	Core collections (cluster approach)	Entire collections
Plant pigmentation at flowering	0.505	0.311	0.379
Leaf colour	1.102	0.756	0.761
Blade pubescence	0.909	0.728	0.714
Sheath pubescence	0.341	0.064	0.045
Degree of lodging at maturity	0.690	0.634	0.539
Senescence	0.364	0.444	0.342
Inflorescence lobes	1.205	1.037	0.934
Inflorescence bristles	1.520	1.525	1.393
Lobe compactness	1.111	0.941	0.913
Inflorescence shape	1.473	1.368	1.349
Inflorescence compactness	0.587	0.292	0.251
Fruit colour	1.385	0.942	0.884
Grain shape	0.671	0.462	0.466
Apical sterility in panicle	0.689	0.692	0.692

Conclusions

“PowerCore” is a new and a faster approach for developing core collection, which effectively simplifies the generation process of a core set with reduced number of core entries but maintaining high percent of diversity compared to other methods used. The efficiency of “PowerCore” when compared with other clustering methods showed that in most cases the mean component for the two core sets were at par and the variance components were higher in core collection developed using “PowerCore”. Thus, it can be concluded that the core sets identified using “Power Core” are small in size with greater diversity captured compared to traditional clustering method.

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Genetic and Environmental Variability in Sorghum [*Sorghum bicolor* (L.) Moench] Germplasm Collected from Rajasthan and Madhya Pradesh

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Sorghum (*Sorghum bicolor* L. Moench) germplasm collections consisting of 80 accessions from Madhya Pradesh and Rajasthan were evaluated during *kharif* 2008 and 2009 (June-October) season for 22 agro-morphological traits using minimal descriptors developed by NBPGR. Analysis of variance showed significant differences among genotypes for 22 traits. Large variation among genotypes was found for the traits viz., days to flowering (49-85 days), leaf length (47-95 cm), leaf width (4.2-8.7 cm), number of leaves per plant (7-15), plant height (165-349 cm), panicle length (8.2-29.4 cm), panicle width (3.5-8.9 cm), 100-seed weight (1.7-3.4 g), days to maturity (101-121 days), stem fresh weight/10 plants) (575-3,225 g) and stem dry weight/10 plants (288-2188 g). Selections can be practiced for traits such as days to flowering, leaf length, leaf orientation, days to maturity and plant height as governed by additive gene action.

Key Words: Cluster analysis, GCV, PCV, Principal components, Sorghum

Introduction

Sorghum [*Sorghum bicolor* (L.) Moench] is fifth most important cereal crop worldwide after wheat, rice, maize and barley (FAO, 2004). It is grown during rainy and post-rainy seasons in semi-arid regions of the country on the marginal lands. It is better adapted to drought-prone regions with poor soils when compared to other cereal crops. In India, the crop is mostly grown in Maharashtra, Karnataka, Andhra Pradesh, Madhya Pradesh, Rajasthan, Uttar Pradesh, Gujarat and Tamil Nadu. In India it occupies 7.8 million ha. In the light of rapidly increasing human population and expansion of agriculture into marginal areas, the importance of sorghum in semi-arid regions of the world will increase in future (House, 1995). In order to meet the food requirement of the increasing population, development of high yielding varieties is essential. Farmers and breeders use local cultivars or landraces for varietal improvement programmes. They are morphologically similar or different as they are adapted to particular environments. For the present study traditional cultivars or primitive cultivars or local landraces or farmer's variety of sorghum were collected from Rajasthan and Madhya Pradesh and evaluated at the Directorate of Sorghum Research, Hyderabad.

Materials and Methods

The sorghum germplasm collections consisting of 80 accessions from Rajasthan and Madhya Pradesh were the base material used for evaluation. The germplasm

collection represented all the basic races of sorghum viz., *bicolor* (8), *durra* (40), *durra-bicolor* (6), *durra-caudatum* (24), and *guinea-caudatum* (2) (Table 1). These accessions were evaluated in *kharif* 2008 and 2009 (June-October) season at the New area field farm of Directorate of Sorghum Research, Rajendranagar, Hyderabad located at latitude 17° 19' 28.5" N and longitude 78° 24' 13.4" E at an altitude of 524 m msl. The seed material was sown in augmented block design plots with two replications in both the growing seasons. Each genotype was sown in three rows, 5 m long, with a row-to-row spacing of 45 cm, and plant-to-plant spacing within a row, of 15 cm. Recommended cultural practices were followed to raise the crop. The data were recorded in each genotype on 22 agro-morphological traits following minimal descriptor developed by National Bureau of Plant Genetic Resources (Mahajan *et al.*, 2000). Five representative plants in each accession were tagged for recording the qualitative and quantitative traits. The seedling vigour was recorded in 14-days-old seedlings. The midrib colour was recorded in the one month aged seedlings. The data on days to 50% flowering was recorded during the panicle emergence stage. The leaf orientation, leaf length (cm), and leaf width (cm), number of leaves, plant height (cm), were recorded during the physiological maturity stage. The earhead shape, compactness, length (cm) and width (cm) were counted during the maturity stage. The stem fresh weight (g), stem dry weight (g) and 100-seed weight (g) were measured after the harvesting. The glume colour,

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Table 1. List of accessions collected from two states with racial classification

Sl. No.	Genotype	National Identity Number	State	District	Race	Sl. No.	Genotype	National Identity Number	State	District	Race
1	E 1	IC 338968	Rajasthan	Udaipur	Durra	33	EJ 22	IC 339002	Rajasthan	Pali	Durra
2	E 2	IC 338969	Rajasthan	Udaipur	Durra-caudatum	34	EJ 23	IC 339003	Rajasthan	Pali	Durra-caudatum
3	E 3	IC 338970	Rajasthan	Udaipur	Durra	35	EJ 24	IC 339004	Rajasthan	Pali	Durra
4	E 4	IC 338971	Rajasthan	Udaipur	Durra-caudatum	36	EJ 25	IC 339005	Rajasthan	Pali	Durra
5	E 5	IC 338972	Rajasthan	Udaipur	Durra-caudatum	37	EJ 26	IC 339006	Rajasthan	Pali	Durra
6	E 7	IC 338973	Rajasthan	Rajsamand	Durra	38	EJ 27	IC 339007	Rajasthan	Pali	Durra
7	E 9	IC 338974	Rajasthan	Rajsamand	Durra	39	EJ 28	IC 339008	Rajasthan	Pali	Durra
8	E 11	IC 338976	Rajasthan	Rajsamand	Bicolor	40	EJ 29	IC 339009	Rajasthan	Pali	Durra-caudatum
9	E 12	IC 338977	Rajasthan	Rajsamand	Durra-caudatum	41	EJ 30	IC 339010	Rajasthan	Pali	Durra
10	E 13	IC 338978	Rajasthan	Rajsamand	Bicolor	42	EJ 31	IC 339011	Rajasthan	Pali	Durra
11	E 14	IC 338979	Rajasthan	Rajsamand	Durra	43	EJ 32	IC 339012	Rajasthan	Pali	Durra
12	E 15	IC 338980	Rajasthan	Rajsamand	Bicolor	44	EJ 33	IC 339013	Rajasthan	Pali	Durra
13	EJ 2	IC 338982	Rajasthan	Udaipur	Durra-caudatum	45	EJ 34	IC 339014	Rajasthan	Nagaur	Durra
14	EJ 3	IC 338983	Rajasthan	Pali	Durra-caudatum	46	EJ 35	IC 339015	Rajasthan	Nagaur	Durra
15	EJ 4	IC 338984	Rajasthan	Pali	Guinea-caudatum	47	EJ 36	IC 339016	Rajasthan	Nagaur	Durra bicolor
16	EJ 5	IC 338985	Rajasthan	Pali	Durra-caudatum	48	EJ 37	IC 339017	Rajasthan	Nagaur	Durra
17	EJ 6	IC 338986	Rajasthan	Pali	Durra-caudatum	49	EJ 38	IC 339018	Rajasthan	Nagaur	Durra
18	EJ 7	IC 338987	Rajasthan	Sirohi	Durra	50	EJ 39	IC 339019	Rajasthan	Ajmer	Durra
19	EJ 8	IC 338988	Rajasthan	Sirohi	Durra	51	EJ 40	IC 339020	Rajasthan	Ajmer	Durra-caudatum
20	EJ 9	IC 338989	Rajasthan	Sirohi	Durra	52	EJ 41	IC 339021	Rajasthan	Tonk	Durra-caudatum
21	EJ 10	IC 338990	Rajasthan	Sirohi	Bicolor	53	EJ 42	IC 339022	Rajasthan	Tonk	Durra bicolor
22	EJ 11	IC 338991	Rajasthan	Sirohi	Bicolor	54	EJ 43	IC 339023	Rajasthan	Tonk	Durra-caudatum
23	EJ 12	IC 338992	Rajasthan	Sirohi	Bicolor	55	EJ 44	IC 339024	Rajasthan	Bhilwara	Durra
24	EJ 13	IC 338993	Rajasthan	Sirohi	Durra	56	EJ 45	IC 339025	Rajasthan	Bhilwara	Durra bicolor
25	EJ 14	IC 338994	Rajasthan	Sirohi	Durra	57	EJ 46	IC 339026	Rajasthan	Bhilwara	Durra
26	EJ 15	IC 338995	Rajasthan	Sirohi	Durra-caudatum	58	EJ 47	IC 339027	Rajasthan	Bhilwara	Durra bicolor
27	EJ 16	IC 338996	Rajasthan	Sirohi	Durra	59	EB 1	IC 332460	Madhya Pradesh	Indore	Durra-caudatum
28	EJ 17	IC 338997	Rajasthan	Sirohi	Durra	60	EB 2	IC 332461	Madhya Pradesh	Indore	Durra-caudatum
29	EJ 18	IC 338998	Rajasthan	Sirohi	Durra bicolor	61	EB 3	IC 332462	Madhya Pradesh	Dhar	Durra-caudatum
30	EJ 19	IC 338999	Rajasthan	Pali	Durra	62	EB 4	IC 332463	Madhya Pradesh	Dhar	Durra-caudatum
31	EJ 20	IC 339000	Rajasthan	Pali	Durra-caudatum	63	EB 5	IC 332464	Madhya Pradesh	Dhar	Bicolor
32	EJ 21	IC 339001	Rajasthan	Pali	Durra						

Sl. No.	Genotype	National Identity Number	State	District	Race
64	EB 6	IC 332465	Madhya Pradesh	Dhar	Bicolor
65	EB 7	IC 332466	Madhya Pradesh	Dhar	Durra-caudatum
66	EB 8	IC 332467	Madhya Pradesh	Dhar	Durra-caudatum
67	EB 9	IC 332468	Madhya Pradesh	Dhar	Durra
68	EB 10	IC 332469	Madhya Pradesh	Dhar	Durra
69	EB 11	IC 332470	Madhya Pradesh	Dhar	Guinea-caudatum
70	EB 12	IC 332471	Madhya Pradesh	Dhar	Durra bicolor
71	EB 13	IC 332472	Madhya Pradesh	Jhabua	Durra-caudatum
72	EB 14	IC 332473	Madhya Pradesh	Jhabua	Durra
73	EB 15	IC 332474	Madhya Pradesh	Jhabua	Durra
74	EB 16	IC 332475	Madhya Pradesh	Jhabua	Durra
75	EB 17	IC 332476	Madhya Pradesh	Jhabua	Durra-caudatum
76	EB 18	IC 332477	Madhya Pradesh	Jhabua	Durra
77	EB 19	IC 332478	Madhya Pradesh	Jhabua	Durra
78	EB 20	IC 332479	Madhya Pradesh	Jhabua	Durra
79	EB 21	IC 332480	Madhya Pradesh	Jhabua	Durra-caudatum
80	EB 22	IC 332481	Madhya Pradesh	Dhar	Durra

glume coverage, presence of awns, grain color, grain size and grain lustrous were observed after the harvesting stage. For qualitative and quantitative traits data, average of five plants per genotype, were computed and these were used for statistical analyses.

Analysis of variance (ANOVA) was carried out assuming season effects as random, and cultivar effects as fixed. The homogeneity of error variance was established as per Gomez and Gomez (1984). Coefficient of variation (CV%), phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and heritability in broad-sense (h^2) were calculated according to the procedure described by Singh and Chaudhary (1985), and expected genetic advance (GA) as suggested by Johnson *et al.* (1995). The data were subjected to multivariate

analyses (Mahalanobis, 1949) and the genotypes were further grouped into different clusters based on Ward's minimum variance method (Hair *et al.*, 1987), and principal component analysis was done according to Rao (1964). All the data analysis was performed using Indostat (2004).

Results and Discussion

Genetic Variability

Analysis of variance showed significant differences among genotypes for all traits studied. Large variation among genotypes was found for the traits, days to flowering (49-85 days), leaf length (47-95 cm), leaf width (4.2-8.7 cm), number of leaves per plant (7-15), plant height (165-349 cm), panicle length (8.2-29.4 cm), panicle width (3.5-8.9 cm), 100 seed weight (1.7-3.4 g), days to maturity (101-121 days), stem fresh weight/10 plants (575-3225 g) and stem dry weight (288-2188 g) (Table 1). Analysis of variance showed significant differences among the genotypes for 22 traits. Significant GxE interactions for leaf length and leaf width of the plants were observed.

Genetic Diversity

Phenotypic and genotypic co-efficient of variations, heritability and genetic gain were calculated following Burton and De Vane (1953). Significant differences were observed among the genotypes for all the characters indicating presence of adequate genetic variability in the experimental material. The genetic constants for the characters revealed that the magnitude of phenotypic co-efficient of variation (PCV) was higher than the corresponding genotypic co-efficient of variation (GCV) denoting environmental factors influencing their expressions to some degree or other. Narrow difference between PCV and GCV suggested their relative resistance to environmental alteration. In the present study, the PCV and GCV were higher for days to 50% flowering, leaf length, leaf width, number of leaves, plant height, earhead length, earhead width, 100-seed weight, days to maturity, stem fresh weight, stem dry weight, vigour, leaf orientation, midrib colour, earhead shape, earhead compactness, glume colour, glume covering (Table 2). High amount of GCV and PCV suggested greater scope for selection of superior genotypes for these traits. The lower degree or equal GCV and PCV for presence of awns, grain size, grain color and grain lustrous indicated that improvement for such traits might be achieved only

Table 2. Genetic variability, heritability and genetic advance for different traits in sorghum germplasm collected from Rajasthan and Madhya Pradesh

Traits	Mean	C.V.	C.D 5%	Range	Variance (Genotypical)	Variance (Phenotypical)	ECV	GCV	PCV	h ² percentage (Broad sense)	Genetic advance as % of mean (5%)
Days to 50% flowering (days)	66.90	10.07	9.38	49-85	55.55	100.90	10.07	11.14	15.02	55.00	17.03
Leaf length (cm)	70.99	12.04	11.91	46.6-94.5	127.55	200.65	12.04	15.91	19.95	64.00	26.13
Leaf width (cm)	6.21	15.09	1.30	4.2-8.7	0.54	1.41	15.09	11.81	19.16	38.00	14.98
Number of leaves	10.39	15.16	2.19	7-15	3.89	6.37	15.16	18.97	24.28	61.00	30.52
Plant height (cm)	252.62	10.84	38.16	165-349.1	2258.62	3009.10	10.84	18.81	21.72	75.00	33.58
Earhead length (cm)	16.57	14.95	3.45	8.2-29.4	27.29	33.42	14.95	31.53	34.90	82.00	58.69
Earhead width (cm)	5.55	15.39	1.19	3.5-8.9	1.03	1.76	15.39	18.33	23.93	59.00	28.92
100-Seed weight (g)	2.40	20.66	0.69	1.7-3.4	0.12	0.37	20.66	14.59	25.29	33.00	17.33
Days to maturity (days)	109.16	4.83	7.34	101-121	26.55	54.31	4.83	4.72	6.75	49.00	6.80
Stem fresh weight (g)	1632.66	40.44	919.78	575-3225	338238.10	774205.00	40.44	35.62	53.89	44.00	48.50
Stem dry weight (g)	937.00	52.49	685.10	288-2188	150568.30	392442.50	52.49	41.41	66.86	38.00	52.84
Vigour	2.71	2.06	0.08	2-3	0.21	0.21	2.06	16.74	16.86	99.00	34.22
Leaf orientation	1.10	5.07	0.08	1-2	0.09	0.09	5.07	27.27	27.74	97.00	55.24
Midrib colour	1.28	4.37	0.08	1-2	0.20	0.20	4.37	35.00	35.28	99.00	71.55
Earhead shape	3.77	1.48	0.08	2-7	2.58	2.58	1.48	42.59	42.61	100.00	87.68
Earhead compactness	1.75	3.19	0.08	1-3	0.52	0.52	3.19	41.16	41.28	99.00	84.53
Glume colour	4.65	1.20	0.08	2-10	7.93	7.93	1.20	60.51	60.53	100.00	124.64
Glume covering	2.33	2.40	0.08	1-4	0.85	0.85	2.40	39.52	39.60	100.00	81.27
Awns	0.73	0.00	-	0-1	0.20	0.20	0.00	61.98	61.98	100.00	127.67
Grain size	5.83	0.00	-	3-9	3.01	3.01	0.00	29.77	29.77	100.00	61.33
Grain colour	3.09	0.00	-	1-14	9.90	9.90	0.00	101.93	101.93	100.00	209.97
Grain lustrous	0.16	0.00	-	0-1	0.14	0.14	0.00	228.45	228.45	100.00	470.61

up to some extent. Similar findings were also obtained by Warkad *et al.* (2008). Equal GCV and PCV was observed in sorghum for plant height up to base of flag leaf, stigma length, plant height, panicle length (Reddy, *et al.*, 2009). High GCV and PCV values were found for grain lustrous.

The determination of the heritable portions is not based only on the estimation of the GCV and PCV. The estimation of heritability along with the coefficient of variability would mean the amount of advance expected. Burton (1952) also suggested that GCV and heritability estimate would give better information about

the efficiency of the selection. The heritability ranged from 33 (100-seed weight) to 100 (earhead shape, glume colour, glume covering, presence of awns, grain size, grain colour and grain lustrous). The utility of heritability is increased when it is used to estimate genetic advance (Johnson *et al.*, 1995). The genetic advance has an added edge over heritability as a guiding factor to breeders in selection programme. High heritability coupled with high genetic advance and GCV was noticed for grain colour, grain lustrous, presence of awns, earhead shape and earhead compactness.

Table 3. Cluster mean values for different contributing characters

S. No	Variable	PC I	PC I	PC II	PC III	PC IV
1	Days to flowering (days)	66.71	66.71	66.66	68.70	72.75
2	Leaf length (cm)	70.69	70.69	71.88	70.24	77.05
3	Leaf width (cm)	6.18	6.18	6.23	6.30	6.53
4	Number of leaves (No)	10.36	10.36	10.54	9.95	11.75
5	Plant height (cm)	255.00	255.00	254.46	212.95	283.63
6	Earhead length (cm)	14.77	14.77	20.92	20.10	27.13
7	Earhead width (cm)	5.52	5.52	5.90	4.66	5.50
8	100-Seed weight (g)	2.45	2.45	2.28	2.22	2.53
9	Days to maturity (days)	109.32	109.32	109.10	106.65	114.00
10	Stem fresh weight (g)	1635.97	1635.97	1641.18	1440.00	2262.50
11	Stem dry weight (g)	944.58	944.58	943.01	772.50	1225.00
12	Vigour	2.81	2.81	2.46	2.40	2.00
13	Leaf orientation	1.13	1.13	1.13	1.00	1.00
14	Midrib colour	1.20	1.20	1.43	1.80	2.00
15	Earhead shape	3.24	3.24	4.93	5.15	7.00
16	Earhead compactness	1.47	1.47	2.41	2.60	3.00
17	Glume colour	4.34	4.34	4.60	8.40	7.00
18	Glume covering	2.28	2.28	2.60	1.80	4.00
19	Awns	0.87	0.87	0.53	0.00	0.00
20	Grain size	6.41	6.41	4.78	3.80	3.00
21	Grain colour	1.49	1.49	8.16	2.00	14.00
22	Grain lustrous	0.24	0.24	0.07	0.00	0.00

The 80 sorghum germplasm genotypes tested for two years during the seasons were grouped into four clusters based on D^2 values (Table 5). The distribution pattern indicated that the maximum number of genotypes were included in cluster I with 57 genotypes, followed by cluster II having 17 genotypes and cluster III having five genotypes. The cluster IV has only one genotype i.e. E 11 (IC 338976). The cluster distance ranged from 2218.3 to 4640.8 within the clusters and 5972.5 to 45279.1 between clusters (Table 6). The inter-cluster distance were higher than intra-cluster distance, which indicated

Table 4. Matrix of principal components of 22 variables for 80 sorghum germplasm

S. No	Variable	PC I	PC II	PC III	PC IV
1	Days to flowering (days)	0.00057	0.01386	0.00582	0.02258
2	Leaf length (cm)	0.00268	0.01832	0.00261	0.04537
3	Leaf width (cm)	0.00112	-0.00036	0.00006	-0.0019
4	Number of leaves (No)	0.00099	0.01327	-0.00521	0.01596
5	Plant height (cm)	-0.0014	0.017	-0.01088	0.03573
6	Earhead length (cm)	0.02035	0.00138	0.052	-0.00346
7	Earhead width (cm)	0.00017	0.01594	-0.02072	0.03526
8	100-Seed weight (g)	0.00098	0.01308	-0.00074	0.01583
9	Days to maturity (days)	0.00185	0.01422	0.00039	0.02041
10	Stem fresh weight (g)	0.00167	0.01122	-0.00007	0.01314
11	Stem dry weight (g)	0.00018	-0.00232	0.00354	-0.00118
12	Vigour	-0.02405	-0.01468	-0.03671	0.03878
13	Leaf orientation	-0.0047	-0.02125	-0.03358	0.03687
14	Midrib colour	0.01223	-0.02898	0.05501	-0.03703
15	Earhead shape	0.14817	-0.10351	0.65734	0.35008
16	Earhead compactness	0.00763	0.04441	-0.02309	-0.07413
17	Glume colour	0.06074	-0.88706	-0.33683	0.12449
18	Glume covering	-0.02576	-0.05567	-0.28149	-0.40529
19	Awns	-0.09534	0.2362	-0.16442	0.14118
20	Grain size	-0.16693	0.28583	-0.52977	0.63239
21	Grain colour	0.71544	0.2335	-0.23924	-0.30342
22	Grain lustrous	-0.65088	0.04548	0.03035	-0.41213
	Root	213350.5	40488.19	32098.16	11047.6
	% Var. Exp.	69.21899	13.1359	10.41386	3.58426
	Cum. Var. Exp.	69.21899	82.35486	92.76874	96.353

wide genetic diversity among the accessions of different groups than those of same cluster. The least intra-cluster distance among the accessions was 2218.3. In cluster I, there was lack of variation among the accessions.

The diversity among the present set of material was also supported by the appreciable amount of variation among cluster means for different characters (Table 4). The cluster which is diverse from other clusters

Table 5. Grouping of genotypes into different clusters

Cluster	Genotype Name
I	EJ 26 (IC 339006), EJ 27 (IC 339007), EJ 24 (IC 339004), E 9 (IC 338974), EJ 6 (IC 338986), EJ 15 (IC 338995), EJ 17 (IC 338997), EJ 22 (IC 339002), EJ 32 (IC 339012), EB 3 (IC 332462), EJ 19 (IC 338999), EB 16 (IC 332475), EB 21 (IC 332480), E 7 (IC 338973), EJ 30 (IC 339010), EJ 36 (339016), EJ 33 (IC 339013), EJ 21 (IC 339001), EJ 29 (IC 339009), EJ 23 (IC 339003), EJ 20 (IC 339000), EJ 5 (IC 338985), EJ 39 (IC 339019), EJ 3 (IC 338983), EJ 14 (IC 338994), EB 14 (IC 332473), EJ 8 (IC 338988), EJ 16 (IC 338996), EB 19 (IC 332478), EJ 38 (IC 339018), EJ 31 (IC 339011), EJ 35 (IC 339015), EB 9 (IC 332468), EJ 47 (339027), EJ 41 (IC 339021), EJ 7 (IC 338987), EB 1 (IC 332460), EB 15 (IC 332474), EJ 42 (IC 339022), E 2 (IC 338969), EJ 37 (IC 339017), EJ 45 (IC 339025), EB 10 (332469), E 5 (IC 338972), E 4 (IC 338971), EB 20 (IC 332479), EB 4 (IC 332463), EJ 40 (IC 339020), EB 22 (IC 332481), EB 5 (IC 332464), EJ 25 (IC 339005), EB 2 (IC 332461), EB 7 (IC 332466), EJ 34 (IC 339014), EJ 2 338982), EB 13 (IC 332472), EJ 12 (IC 332471)
II	EJ 44 (IC 339024), EJ 46 (IC 339026), EB 18 (IC 332477), E 1 (IC 338968), EJ 28 (IC 339008), E 12 (IC 338977), EB 8 (IC 332467), EB 12 (IC 332471), EB 6 (IC 332465), EB 11 (IC 332470), E 14 (IC 338979), EJ 43 (IC 339023), E 13 (IC 338978), EJ 11 (IC 338991), EJ 18 (IC 338998), EJ 10 (IC 338990), EJ 9 (IC 338989)
III	EJ 4 (IC 338984), EB 17 (IC 332476), E 3 (IC 338970), EJ 13 (IC 338993), E 15 (IC 338980)
IV	E 11 (IC 338976)

has different mean values for leaf width, number of leaves, earhead length, earhead width, 100-seed weight, stem fresh weight, stem dry weight, midrib colour, earhead shape, earhead compactness, glume colour, glume covering, awns, grain size, grain colour, and grain lustrous. Cluster IV also was diverse from other clusters due to different cluster mean values for days to flowering, leaf length, leaf orientation, days to maturity, plant height. Exploitation of genetic resources within and among these contrasting pools of diversity may provide a different source of germplasm with combination traits for utilization in the sorghum improvement programme. Landraces adapt to different climatic conditions which may be used for developing improved cultivars for yield. Selection for the traits plant height, grain weight can be practiced and incorporated easily as the traits

Table 6. Average intra (bold) and inter cluster distances for 80 genotypes of sorghum (Tocher method)

Cluster	I	II	III	IV
I	2218.26	15318.97	5972.54	45279.08
II		4640.78	11191.85	12563.12
III			3259.75	32376.30
IV				0

are governed by additive gene action. The traits glume colour, awns, grain colour, grain lustrous are governed by the non-additive gene effects and can be made use for developing distinct variety.

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Characterization of High Yielding and Drought Tolerant RILs Identified from Wheat Cross WL711 x C306 RIL Mapping Population using Drought Susceptibility Index (DSI) as Selection Criteria

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Wheat cultivars WL711 and C306 were shown to be drought susceptible and drought tolerant, respectively, in an earlier study. These two cultivars were crossed to obtain a RIL population comprising 206 RILs. The parents and the RIL mapping population was tested for drought tolerance *via* field trials conducted at Delhi from 2007 to 2010. Water stress was created by withholding irrigation. Phenotyping of the RIL population was done for phenology, yield and yield components under irrigated (Irr) and water stress (WS) treatments. The range of days to flowering in the RIL population was 78-98 and 74-96 days after sowing (DAS) under Irr and WS conditions, respectively. The range of grain yield, biomass, harvest index, 1000 grain weight, grain number in the RIL population was 335.1-720.0g/m², 1316-2413g/m², 22.3-58.6%, 24.0-45.1g, 9497-22763/m² and 219.2-568.1 g/m², 992-1759g/m², 17.3-54.8%, 23.1-39.7g, 8807-17509/m² under Irr and WS conditions respectively. Analysis of variance revealed significant interaction between genotypes and treatment for all the traits except 1000-grain weight and harvest index. Drought Susceptibility Index (DSI) was used as criteria for drought tolerance. DSI of yield and yield components showed considerable variation and transgressive segregation in the RIL population. Based on DSI of yield and yield components of the medium to late flowering RILs, 8 RILs were identified for combining yield higher than C306 with yield stability. These RILs maintained better water relations and tougher membranes under WS treatment, like C306.

Key Words: Drought susceptibility index, Recombinant inbred lines, Water stress, Wheat, Yield, Yield components

Introduction

Bread wheat (*Triticum aestivum* L.) is an important food crop in India. Drought is by far the most important environmental stress in agriculture (Cattivelli *et al.*, 2008; Mir *et al.*, 2012). In India, although 80% of the wheat is grown under irrigated conditions, only one-third receives full irrigation while the remainder is cultivated under partial irrigation with 1-2 irrigations over the cropping season. It is likely that water will become a limiting factor for sustained production of wheat in India (Joshi *et al.*, 2007). The duration and intensity of drought is location specific and can vary from year-to-year because of rainfall pattern. When monsoon and winter season rainfall in India are considered, the probability of water deficit is predictable. Wheat crop can experience water deficit stress during growth and development both in irrigated and limited irrigation environments depending upon the water availability (Sinha *et al.*, 1982). In fact wheat crop often experiences both drought and heat stress

in the post-anthesis period as it matures in increasing temperatures in North India. Growing food demand and global warming would further push wheat crop to drought and heat stress environments (Araus *et al.*, 2008; Semenov and Helford, 2009; Sinclair, 2011). Therefore, breeding for drought tolerance in wheat has been a major aim of many breeding programs both nationally and internationally in order to improve crop productivity under water-limiting conditions (Araus *et al.*, 2002; 2008; Chaves *et al.*, 2003, Richards *et al.*, 2010). If the breeding programs address factors of stress-adaptation in addition to yield under stress, then it may be possible to combine higher yield potential and drought tolerance (Blum, 2005; Richards *et al.*, 2010).

Wheat cultivars WL711 and C306 were shown to be drought susceptible and drought tolerant respectively in an earlier study involving *Triticum aestivum*, *T. durum* and *Triticale* germplasm (Sinha *et al.*, 1986). With the aim of combining drought tolerance with high yield, a

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program was initiated at the Stress Physiology Laboratory, Water Technology Centre, IARI, New Delhi in which these two varieties were crossed in order to obtain a RIL population (Patil and Khanna-Chopra, 2006). The aim of the research reported in this paper was to identify RILs combining drought tolerance with yield higher than C306 from the cross WL711 x C306 based on DSI and to analyse the basis of stability in yield in relation to physiological traits such as water relations and membrane stability in the post-anthesis stage.

Materials and Methods

Material and Location

The parent cultivars WL711 [(S308 x Chris) x Kalyansona] and C306 [(Regent x Ch23) x C591 (P19 x C28)] have several unique morphological and physiological differences in addition to yield and yield components. WL711 is a semi-dwarf, high yielding, drought susceptible, medium flowering variety with medium seedling vigour, erect growth habit and medium grains which are dull brown in colour while C306 is a tall, medium yielding, drought tolerant, late flowering variety with high seedling vigour, spreading growth habit and bold grains which are light amber in colour. The present investigation comprises of 206 RILs, F_{10} generation onwards of the WL711 x C306 cross. The experiments were conducted for three consecutive years from 2007 to 2010 in the fields of Water Technology Centre (IARI), New Delhi (77°12'E, 28°40'N; 228 msl). The data presented is an average of the three trials at Delhi.

Soil Characteristics

The soil at Water Technology Centre (IARI) is sandy loam with average bulk density of 1.55 g cm^{-3} . The water retention capacity is 18% at field capacity (-1.5MPa) and 4.5% at permanent wilting point (-1.5Mpa) on w/w basis (Sinha *et al.*, 1986). The contribution of ground water through capillary action is almost negligible because the depth of water table is 4m.

Sowing

The parent cultivars WL711, C306 and 206 RIL population were sown in the field during winter seasons of 2007-08, 2008-09 and 2009-10. Seed rate was approximately 200 seeds per m^2 . Pre-sowing irrigation was applied to obtain sufficient moisture for germination. Sowing was done on 25th November in the three trials in a Randomized Complete Block Design (RCBD)

with four replications. The plot size was $3 \times 2 \text{ m}^2$ with 6 rows with a distance of 25cm between the rows. A fertilizer dose of 100:50:50 kg NPK ha^{-1} was applied in the form of urea, single super phosphate and murate of potash, respectively, as a single basal dose at the time of sowing. Weeds were removed from the field as and when they appeared.

Irrigation Treatments

There were two irrigation treatments, full irrigation (Irr) and water stress (WS). Irr treatment received 4 irrigations at tillering (DC 2.0), jointing (DC 3.0), flowering (DC 6.0), and grain filling stages (DC 7.0) while WS treatment received 2 irrigations at tillering (DC 2.0) and flowering (DC 6.0). Each irrigation was approximately equivalent to 4 cm of water. The Irr treatment received 37.7, 37.1 and 37.8cm while WS treatment received 28.9, 28.3 and 29.5cm of total available soil water in 2007-08, 2008-09 and 2009-10 respectively.

Parameters Recorded

Phenology

Days of flowering was recorded for all RILs under both water treatments as the date when 50% of the shoots had reached this stage by observing RILs every 1-2 days from February to the first week of March. Days to flowering was calculated as days after sowing (DAS).

Physiological Parameters

Relative water content (RWC), and cell membrane stability (CMS) were measured in the parents and the selected RILs in 2009-10. RWC and CMS were measured in flag leaves at post-anthesis stage (A+14) under both Irr and WS treatments following the procedure given by Barrs and Weatherly (1962) and Blum and Ebercon, (1981), respectively. The results of CMS were expressed as % injury using the formula:

$$\% \text{ Relative injury} = 1 - [1 - (T_1/T_2)] / [1 - (C_1/C_2)] \times 100.$$

Where, C and T refer to mean of treatment and controls, respectively, and the subscripts 1 and 2 refer to initial and final conductivity respectively.

Yield and Yield Components

One meter square area was harvested of each RIL at maturity for biomass (g/m^2) and grain yield (g/m^2). There were 3 replicates per treatment. 500 grains were counted at random and their weight was recorded and expressed as 1000 grain weight. Harvest index (HI) was

calculated by the formula- $HI = \text{grain yield} / \text{total above ground biomass} \times 100$. Number of grains per meter square was obtained by dividing grain yield (g/m^2) by grain weight and multiplied with 1000.

Drought Susceptibility Index (DSI)

Drought susceptibility index (DSI) and stress intensity (D) was calculated using the formula of Fischer and Maurer, (1978).

$$DSI = (1 - Y/Y_p) / D$$

Where, Y- Mean yield or yield component of a RIL under water stress condition, Y_p - Mean yield or yield components under irrigated condition. D- Stress intensity

$$D = 1 - X/X_p$$

Where, X- Mean Y of all RILs, X_p - Mean Y_p of all RILs

DSI is a measure of drought tolerance. If DSI is < 1 , the cultivar is drought tolerant and $DSI > 1$ indicates that cultivar is drought susceptible.

Statistical Analysis

Analysis of variance (ANOVA) was estimated for all traits separately for estimating coefficient of variation (CV) and critical difference (CD) for evaluation the significance of treatment and trial effects on the parents and the WL711 x C306 RIL population. ANOVA was done using the three factor factorial analysis of the statistical programme MSTAT-C, version 1.41, Michigan State University, USA. Correlation analysis was performed using MSTAT-C.

Results and Discussion

Weather

The average $T_{\max}$ and $T_{\min}$ during the crop season were $24.83 \pm 1.56^\circ\text{C}$ and $10.96 \pm 0.85^\circ\text{C}$ respectively. Total rainfall recorded during the 2007-08, 2008-09 and 2009-10 crop seasons were was 25.6 mm, 37mm and 30.6 mm, respectively. There was no rainfall in the month of March and April i.e. during grain filling to maturity. Meager rainfall and higher ET in both the years helped in development of severe water stress during post-anthesis period.

Phenology

WL711 and C306 flowered 88 DAS and 96 DAS under Irr treatment and 82 DAS and 92 DAS under WS treatment. Flowering in the RIL population started from mid February (78 DAS) and continued up to the first week of March (98 DAS). Under WS treatment, both parents and the RIL population flowered earlier than the irrigated control (Table 1). Phenology plays an important role in response to drought stress as growth duration determines water requirement and probability of exposure to water stress (Araus *et al.*, 2008). Water stress developed in early March and consequently medium to late flowering RILs (85 -98 DAS) are likely to experience water deficit stress, in the post-anthesis period more than the early flowering RILs.

Yield and Yield Components

The parents, WL711 and C306, differed significantly in yield and yield components (Table 1). WL711 is a high yielding variety ($673.9 \pm 57.3 \text{ g/m}^2$) while C306 is a medium yielding variety ($435.8 \pm 23.2 \text{ g/m}^2$). WL711 showed higher biomass ($1856 \pm 120.7 \text{ g/m}^2$), grain number ($18657 \pm 825 / \text{m}^2$) and HI ($39.5 \pm 1.7\%$) than C306 ($1688 \pm 102.0 / \text{m}^2$, $11017 \pm 541 / \text{m}^2$ and $26.7 \pm 0.9\%$ respectively). C306 has bolder grains and higher 1000-grain weight ($37.52 \pm 0.31 \text{ g}$) than WL711 ($33.31 \pm 0.62 \text{ g}$). The RIL population showed normal distribution and transgressive segregation for grain yield under Irr and WS treatments (Fig 1). The mean value of RILs

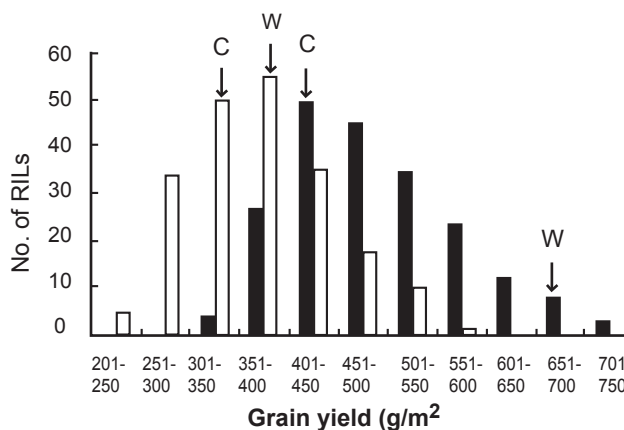


Fig. 1. Frequency distribution of grain yield (g/m^2) of WL711 x C306 wheat RIL population under irrigated and water stress conditions based on the mean values of the three trials conducted at Delhi (2007-2010). The histograms with the solid and the open bars represent the frequencies of grain yield of RILs under irrigated and water deficit stress conditions respectively. W- WL711, C- C306

Table 1. Means ($\pm$ SE) and ranges of grain yield and yield components in the parents, the selected RILs and the WL711 x C306 wheat RIL population under irrigated and water stress environments at Delhi from 2007-2010. Irr- irrigated treatment, WS- water stress treatment; NS –non-significant

Selected RILs	Days to flowering (DAS)		Grain yield (g/m ²)		Biomass (g/m ²)		Harvest index (%)		1000-grain weight (g)		Grain number (/m ²)	
	Irr	WS	Irr	WS	Irr	WS	Irr	WS	Irr	WS	Irr	WS
WL711	87 $\pm$ 2	81 $\pm$ 1	673.9 $\pm$ 57.3	435.8 $\pm$ 23.2	1856 $\pm$ 120	1329 $\pm$ 72	39.5 $\pm$ 1.7	27.6 $\pm$ 2.6	33.3 $\pm$ 0.6	28.1 $\pm$ 0.5	18657 $\pm$ 825	14375 $\pm$ 1018
C306	96 $\pm$ 3	89 $\pm$ 2	435.8 $\pm$ 23.2	347.2 $\pm$ 17.7	1688 $\pm$ 102	1412 $\pm$ 63	26.7 $\pm$ 0.9	26.7 $\pm$ 2.1 ^{NS}	37.5 $\pm$ 0.3	36.4 $\pm$ 1.0	11017 $\pm$ 541	10642 $\pm$ 876 ^{NS}
R-19	91 $\pm$ 2	86 $\pm$ 3	578.8 $\pm$ 36.3	458.3 $\pm$ 36.1	1855 $\pm$ 109	1473 $\pm$ 59	32.2 $\pm$ 3.8	30.4 $\pm$ 2.2	33.8 $\pm$ 1.5	32.4 $\pm$ 1.9	16161 $\pm$ 830	13994 $\pm$ 766
R-46	94 $\pm$ 3	91 $\pm$ 2	540.0 $\pm$ 28.1	405.0 $\pm$ 16.9	1633 $\pm$ 74	1424 $\pm$ 84	31.6 $\pm$ 1.5	29.5 $\pm$ 2.0	37.6 $\pm$ 0.6	34.2 $\pm$ 1.1	14302 $\pm$ 683	13539 $\pm$ 921
R-61	91 $\pm$ 3	87 $\pm$ 3	614.8 $\pm$ 43.8	421.0 $\pm$ 13.1	2040 $\pm$ 129	1480 $\pm$ 40	31.9 $\pm$ 1.6	29.1 $\pm$ 1.6	33.5 $\pm$ 0.5	32.5 $\pm$ 1.3	18286 $\pm$ 982	15092 $\pm$ 745
R-208	90 $\pm$ 2	86 $\pm$ 4	617.6 $\pm$ 36.4	471.6 $\pm$ 16.8	2109 $\pm$ 163	1478 $\pm$ 69	34.3 $\pm$ 4.1	33.1 $\pm$ 1.0	36.1 $\pm$ 0.4	34.7 $\pm$ 1.3	16443 $\pm$ 795	14831 $\pm$ 820
R-25	89 $\pm$ 3	84 $\pm$ 3	659.9 $\pm$ 40.7	461.6 $\pm$ 9.7	2008 $\pm$ 130	1701 $\pm$ 75	35.7 $\pm$ 3.0	27.3 $\pm$ 1.7	36.7 $\pm$ 0.8	36.5 $\pm$ 1.0 ^{NS}	17562 $\pm$ 887	14309 $\pm$ 829
R-109	91 $\pm$ 2	84 $\pm$ 4	673.6 $\pm$ 53.1	498.2 $\pm$ 43.2	2285 $\pm$ 153	1530 $\pm$ 61	33.0 $\pm$ 2.5	31.2 $\pm$ 2.5	35.5 $\pm$ 1.0	35.0 $\pm$ 1.7 ^{NS}	17721 $\pm$ 836	14575 $\pm$ 1154
R-128	94 $\pm$ 1	88 $\pm$ 2	603.1 $\pm$ 44.0	427.2 $\pm$ 27.2	1903 $\pm$ 118	1393 $\pm$ 42	33.6 $\pm$ 2.7	28.2 $\pm$ 2.3	38.0 $\pm$ 1.5	36.0 $\pm$ 1.5	15955 $\pm$ 584	12617 $\pm$ 1035
R-7	86 $\pm$ 3	80 $\pm$ 5	642.1 $\pm$ 35.3	460.2 $\pm$ 21.6	2375 $\pm$ 209	1699 $\pm$ 122	35.5 $\pm$ 1.6	26.2 $\pm$ 2.5	37.4 $\pm$ 0.6	36.3 $\pm$ 1.9	17437 $\pm$ 496	14692 $\pm$ 956
RIL Population Mean	86 $\pm$ 3	82 $\pm$ 2	538.0 $\pm$ 27	368.6 $\pm$ 21	1729 $\pm$ 65	1324 $\pm$ 43	41.2 $\pm$ 2.8	34.2 $\pm$ 2.4	34.3 $\pm$ 1.7	31.6 $\pm$ 1.8	14706 $\pm$ 678	12273 $\pm$ 735
RIL Population Range	78–98	74-95	335.1 –720.0	219.2 – 568.1	1316 – 2413	992 –1759	22.3 – 58.6	17.3 – 54.8	24.0 – 45.1	23.1 – 39.7	9497– 22763	8807 – 17509

Table 2. Analysis of variance for yield and yield components in the WL711 x C306 RIL population at Delhi 2007-2010. NS-non-significant; CV-coefficient of variation; CD-critical difference at 0.05% significance level

Source of variation	df	Mean squares					
		Grain yield (gm ⁻²)	Biomass (gm ⁻²)	Harvest index (%)	1000-grain weight (g)	Grain number (m ⁻²)	Days to flowering
Replicates	3	58715.4	216255.9	3.64	1.13	12149013.1	28.6
Genotypes	210	90164*	278255.3*	61.5*	119.6*	63619182*	151.2*
Treatment	1	18895550.8*	125641914.9*	1614.3*	3148.2*	5293351989*	28729.7*
Genotype x treatment	210	14896.6*	73081.7*	55.7 ^{NS}	10.76 ^{NS}	23821076.3*	16.2*
Trial	2	2384898*	23368523.1*	6919.8*	6005*	3087407217.1*	20662.0
Genotype x trial	420	16485.9*	91768.6*	64.1*	29.37*	22114740.9*	39.0*
Treatment x trial	2	219518.6*	8409674.1*	11054.3*	757.5*	2707242163.7*	2048.5*
G x treatment x trial	420	7256.7*	48390.6*	60*	11.1*	2722673.9*	11.1*
Error	1267	1779.8	17612.8	41	0.304	11783228.3	0.62
CV (%)		9.40%	8.81%	10%	5.20%	24%	0.92%
CD		48.07	151.2	6.06	1.8	1815.5	0.92

was intermediate of the parents for most of the traits. Transgressive segregation was observed for all the traits in the RIL mapping population (Table 1). Analysis of variance revealed significant interaction between genotypes and treatment for all the traits except 1000-grain weight and harvest index (Table 2).

Drought Susceptibility Index (DSI)

C306 was more stable (0.71 ± 0.17) than WL711 (1.39 ± 0.15) for grain yield. Hence C306 is drought tolerant while WL711 is drought susceptible as reported earlier (Sinha *et al.*, 1986; Table 3). DSI has been used as a criterion for evaluating genotypes and mapping populations for drought tolerance in wheat (Ehdaie *et al.*, 1988; Blum *et al.*, 1989; Lazar *et al.*, 1995; Dencic *et al.*, 2000; Foulkes *et al.*, 2007; Kirigwi *et al.*, 2007; Semenov *et al.*, 2009). Considerable variation for DSI for grain yield was observed in the RIL population. Variation for grain yield stability has been observed in other wheat RIL populations under water stress environment (Foulkes *et al.*, 2007; Kirigwi *et al.*, 2007). C306 showed more stability for all yield components compared to WL711. The RILs also showed considerable variation in DSI for biomass, HI, 1000-grain weight and grain number (Table 3). DSI showed higher correlation with grain yield under Irr condition ($r = 0.364^{**}$) than WS condition ($r = 0.113^{**}$).

To identify RILs combining higher yield than C306 with yield stability under WS conditions, only medium and late flowering RILs were analyzed. Out of 182 RILs, 48 RILs were found to be stable for grain yield. Among these RILs, only 8 RILs exhibited higher yield than C306 and yield stability (Table 1, Fig 2). RIL R-19 was stable for four yield components, RILs R-46, R-61, R-208, R-25 and R-109 were stable for three yield components i.e. grain number, 1000-grain weight and HI while RILs R-128 and R-7 showed stability

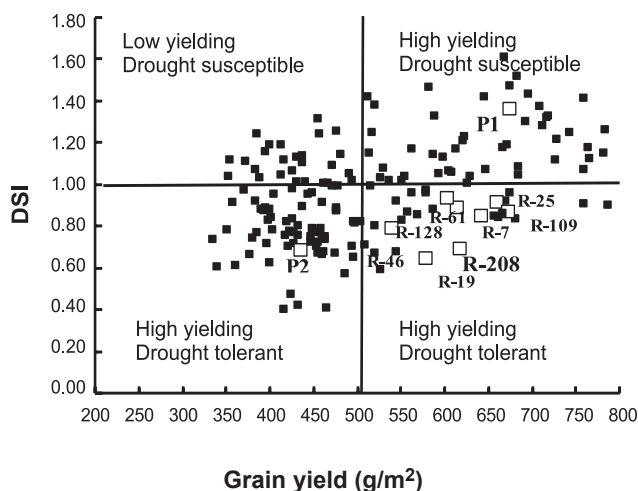


Fig. 2. Relationship between grain yield (g/m^2) and drought susceptibility index (DSI) of grain yield of medium to late flowering RILs from the wheat cross WL711 x C306. Symbol ($\square$) indicates selected RILs, P1-WL711, P2-C306

Table 3. Means ($\pm$ SE) of DSI of grain yield and yield components of the parents and the selected RILs from the wheat cross WL711 x C306 at Delhi from 2007-2010

Parents and selected RILs	Drought susceptibility index				
	Grain yield (g/m^2)	Biomass (g/m^2)	Harvest index (%)	1000grain weight (g)	Grain number ($/\text{m}^2$)
WL711	1.39 ± 0.15	1.18 ± 0.08	2.26 ± 0.11	1.99 ± 0.03	1.29 ± 0.09
C306	0.71 ± 0.17	0.69 ± 0.06	0.20 ± 0.05	0.29 ± 0.11	0.69 ± 0.10
R-19	0.63 ± 0.08	0.79 ± 0.13	0.36 ± 0.10	0.24 ± 0.11	0.73 ± 0.11
R-46	0.80 ± 0.10	0.62 ± 0.06	0.34 ± 0.06	1.23 ± 0.02	0.31 ± 0.06
R-61	0.82 ± 0.04	1.16 ± 0.06	0.99 ± 0.09	0.24 ± 0.09	0.97 ± 0.19
R-208	0.67 ± 0.14	1.06 ± 0.14	0.70 ± 0.08	0.55 ± 0.17	0.55 ± 0.04
R-25	0.92 ± 0.16	0.65 ± 0.17	2.30 ± 0.03	0.11 ± 0.08	1.03 ± 0.12
R-109	0.92 ± 0.04	1.18 ± 0.05	0.29 ± 0.02	0.24 ± 0.10	1.00 ± 0.15
R-128	0.98 ± 0.12	0.92 ± 0.14	1.06 ± 0.07	0.66 ± 0.19	1.23 ± 0.11
R-7	0.83 ± 0.14	1.13 ± 0.13	2.50 ± 0.05	0.42 ± 0.12	0.89 ± 0.11
Range in RIL population	0.27 – 1.84	0.31 – 1.98	-1.8 – 2.6	0.12 – 1.69	0.34 – 1.90

for only two yield components (Table 3). The selected RILs, except RIL R-46 were able to maintain 1000-grain weight under WS treatment and thus showed stability for this character like C306 (Table 1). C306 is known to mobilize pre-anthesis stored carbohydrates in the stems to the developing grains under post-anthesis water stress condition resulting in stable 1000-grain weight and consequently stable grain yield (Aggarwal and Sinha, 1984, Khanna-Chopra *et al.*, 1994). It may be inferred from the performance of selected RILs under WS treatment that some of them may have acquired this trait from C306. The range of grain yield, biomass, grain number, 1000-grain weight and HI of the selected RILs was 540.0-673.6 g/m², 1633-2375 g/m², 14302-18286 / m², 33.5-38.0g and 31.6-35.7 % respectively.

RWC and CMS have been reported to be associated with drought tolerance (Blum, 2005). C306 had higher

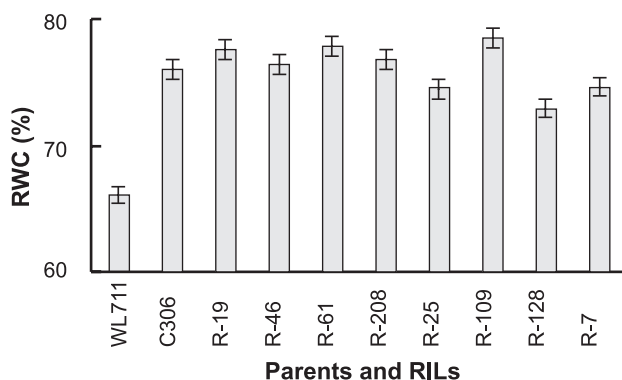


Fig. 3A

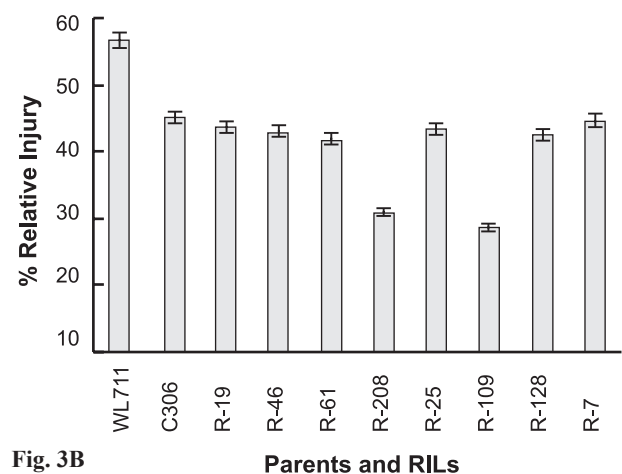


Fig. 3B

Fig. 3. Relative water content (RWC, A) and cell membrane stability (CMS, B) in the flag leaf at post-anthesis stage (A+14) of the parents and the selected RILs from the wheat cross WL711 x C306 under water stress conditions. Vertical bars represent $\pm$ SE (n=3)

RWC and membrane stability as compared to WL711 under WS treatment (Fig. 3A). RILs R-19, R-46, R-109 and R-128 maintained higher RWC than C306 under WS treatment. RILs R-19, R-46, R-61, R-25, R-128 and R-7 were similar to C306 in CMS; however R-208 and R-109 maintained higher CMS than C306 under WS treatments (Fig. 3B). Grain yield of the selected RILs was significantly correlated with RWC ($R^2=0.165$, $p<0.05$), and CMS ($R^2=0.461$) under WS treatment. In previous studies also, grain yield under water stress was shown to be significantly correlated to RWC (Sharma and Kumar, 2010) and CMS (Geravandi *et al.*, 2011). Favourable expression of water relation traits are indicators of the capacity of the crop to access water from deeper layers of soil under water stress condition as a result of deep and dense root system (Reynolds *et al.*, 2005; Blum *et al.*, 1989). Deep root system increases the total water availability to the crop under water stress condition and is associated with improved drought tolerance (Reynolds *et al.*, 2007).

In conclusion we report certain RILs from WL711 x C306 cross showing desirable recombination of traits resulting in yield improvement compared to C306 and showing stability in yield and some yield components under water deficit stress. The selected RILs are medium to late in flowering and some RILs also have amber coloured grains, a desirable character of grain quality. The selected RILs maintained high RWC and tougher membranes under water stress conditions like C306. Four of these RILs (R-7, R-61, R-19 and R-208) have been registered at NBPGR as drought tolerant wheat germplasm (Registration no. INGR11037, INGR11038, INGR11039 and INGR11040 respectively). These RILs can be used as genetic material for transferring the desirable drought tolerance traits acquired from C306.

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Genetic Diversity of Pear (*Pyrus* spp.) in Uttarakhand Himalayas

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Survey of two pear growing districts of Uttarakhand (Champawat and Pithoragarh) was conducted during the fruiting season (July-August) of two consecutive years (2010 – 2011). A total of 38 accessions with distinct traits of horticultural importance were identified. Scion wood of the identified accessions was collected during following winter dormant period (January-February) and grafted on suitable rootstock for conservation in the field genebank. Prior to collection of scion wood of a plant, passport data and data pertaining to flowering and fruiting characters were recorded. Fruit length ranged from 3.6 – 8.2 cm, fruit width 2.6 – 6.7 cm, fruit weight 65 – 290 g, TSS 8.5 – 14.5 °Brix, vitamin 'C' 2.3 – 6.3 mg.100 ml⁻¹, organoleptic value 4 – 9 on 10 point scale and post harvest life of fruits ranged from 5 – 13 days. The accessions collected from different sites were found to have fairly rich diversity in parameters under study.

Key Words: *Pyrus* spp., Total soluble solids, Flowering, Fruit size, Genetic diversity

Introduction

The genus *Pyrus* has originated in Central Asia, the mountainous regions of western and southern China, from Asia Minor to India and further diversified and moved both in eastern and western directions from primary centre of origin (Watkins, 1976). The genus *Pyrus* is classified into more than 20 primary diploid species distributed over Europe and Asia (Zohary and Hopf, 1988) and six naturally occurring inter-specific hybrids (Bell *et al.*, 1996). On the basis of origin and commercial fruit production pear is classified into three main groups, viz., European pear (*Pyrus communis* L.), Japanese pear (*P. pyrifolia* Burm.) and Chinese pear (*P. bretschneideri* Rehd. and *P. ussuriensis* Maxim). Furthermore, the genus is also divided into two native groups, i.e. occidental and oriental pears. The occidental pears include over 20 species mostly found in Europe, Northern Africa, Asia Minor, Iran and Central Asia. The oriental pears include 12 to 15 species distributed from the Tian-Shan and Hindu Kush mountains eastward to Japan. Speciation has occurred mainly in eastern and central Asia in the Himalayas, Caucasus, Asia Minor and Eastern Europe. There are three centres diversity for cultivated pears i.e. Chinese centre, Central Asiatic centre and Near Eastern centre (Vavilov, 1951). Zeven and Zhukovsky (1975) have proposed the fourth centre of diversity as European Siberian centre.

Pyrus spp. grows in a wide range of climatic conditions and can tolerate temperature as low as -26°C in dormant stage and as high as 45°C in growing period. A large number of pear varieties require about 800 to 1200 h below 7°C during winter to complete their chilling requirements to flower and fruit satisfactorily. About 72% of all commercially cultivated species of genus *Pyrus* are native to Asia. People's Republic of China, United States, Italy, Argentina, Spain, India, Turkey, South Africa, South Korea and Belgium are top ten pear producing countries in the world with India producing 3,82,000 tonnes (FAO, 2010). Himalayan region in Asia is known for its biological richness and has always been a botanist's paradise. Its diversified landforms, relief and environmental conditions support a wide range of vegetations. Rich diversity of flora in general and temperate fruits in particular is available in Uttarakhand Himalayas which need to be collected and characterized for further utilization. Genetic diversity is the key component of any agricultural production system. The value of genetic diversity, in its various forms has been extensively discussed (Smale, 2006; Rausser and Small, 2011). Moreover, plant breeders require genetic variation (genotypes) for crop/ plant improvement. In spite of suitable climatic conditions for production of temperate fruits in general and pear in particular, production of pear in Uttarakhand is not keeping pace with the current

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demand. Pear is widely distributed in Champawat and Pithoragarh district of Uttarakhand. Pear germplasm have rich source of genetic variability as accumulated through hybridization, mutation and naturally seed based propagation. *Pyrus* species particularly wild populations are threatened worldwide with about more than 85% pear varieties lost in the 19th century, continuing even today (Sindelar, 2002). Furthermore, due to various insect-pests and pathogens, valuable and highly resistant to various abiotic stresses pear germplasm has been lost (Fowler and Mooney, 1990). Proper attention for exploration, collection, conservation and characterization of genetic resources of pear is an urgent need. Volk *et al.* (2006) reported that a high diversity exists in *P. communis* at the molecular level but that has not been exploited properly. Keeping this in view, the present investigation was undertaken to collect and evaluate the available genetic diversity of pear in two pear growing districts of Uttarakhand i.e., Champawat and Pithoragarh.

Materials and Methods

Four exploration tours (two survey and identification tours during the fruiting season in July-August and two scion wood collection tours during winter dormant stage in January – February) were conducted during 2010 – 2012 in pear growing areas of district Champawat and

Pithoragarh (Uttarakhand) (Fig. 1) to collect available genetic diversity of *Pyrus* spp. Champawat district is situated between 29°5' - 29°30' N latitude and 79°59' - 80°3' E longitude. Pithoragarh district lies between 29°4' - 30°3' N latitude and 80° - 81°E longitude. Formal and informal conversation with local farmers was adopted as a strategy to collect the information about the pear germplasm available in the area. In each village three four farmers were consulted before identification of a genotype for collection. Accessions were selected randomly at fruit maturity stage from 38 sites of these districts and earmarked with durable label (aluminium sheet) during survey. The available diversity of primitive cultivars frequently found in the cultivated habitats of the area was collected from population through selective sampling technique along with passport information. Only disease-free plants bearing fruits with unique traits of horticultural importance were identified for collection. Precocity of flowering, fruit shape, fruit colour, ripening time, fruit size, fruit weight, total soluble solids (TSS) (by refractometer) in the fruit pulp were the main parameters for identifying a genotype for collection. Fruits of identified accessions were collected and vitamin C content was estimated in the laboratory following the method of Davies and Masten (1991). Observations were recorded for growth habit of plants (upright, pyramidal

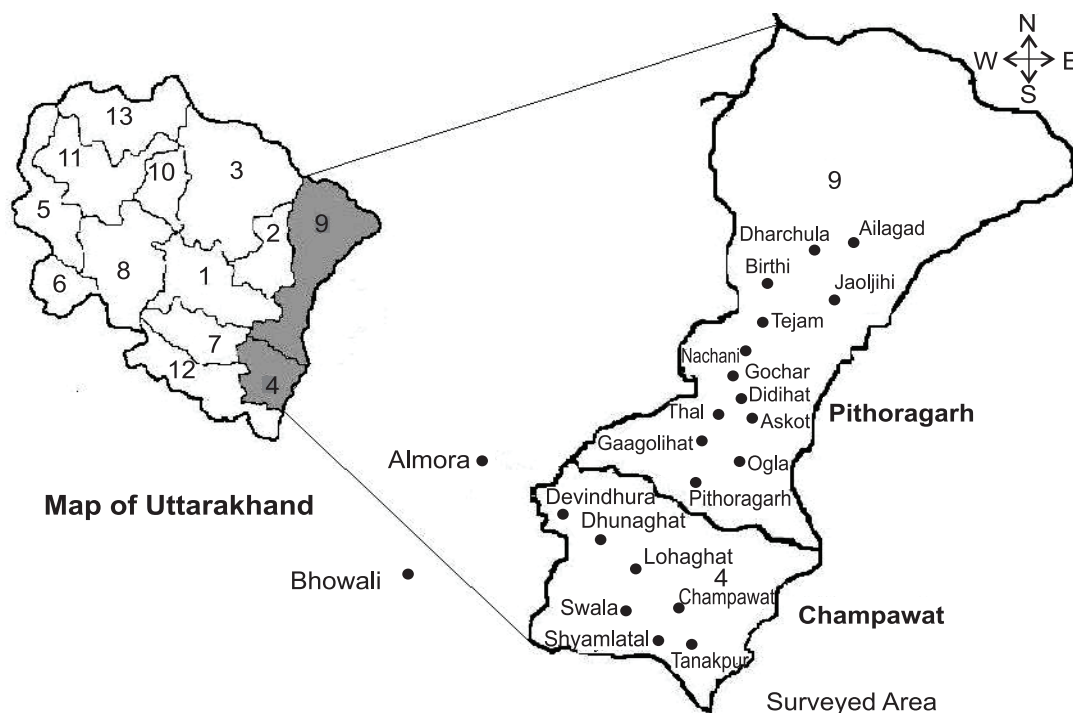


Fig. 1. Sites of *Pyrus* species germplasm collection in Uttarakhand Himalayas

and broad spreading tree) and time of ripening (early to medium, medium and medium to late) in *in situ*. Post-harvest life of ready to eat fruits was calculated by storing them at ambient room temperature. The quantitative data collected were subjected to statistical analysis following two ways analysis of variance considering year and accession as two independent variables. The difference between the two groups was assessed by computation of least significant difference taking 't' values for error at the 5% level of significance.

Table 1. Passport data of pear germplasm collected during exploration

S. No.	Collector No.	Botanical name	Vernacular name	District	Altitude (m)	Latitude	Longitude
1.	AKT/VC-01 (2011)	<i>Pyrus communis</i>	Jagnel	Champawat	1930	29° 23' 34.3" N	80° 08' 43.6" E
2.	AKT/VC-02 (2011)	<i>Pyrus communis</i>	Babboogosa	Champawat	2040	29° 24' 34.0" N	79° 52' 19.9" E
3.	AKT/VC-03 (2011)	<i>Pyrus communis</i>	Garmehel	Champawat	2040	29° 24' 34.0" N	79° 52' 19.9" E
4.	AKT/VC-04 (2011)	<i>Pyrus communis</i>	Garmehel	Champawat	2035	29° 24' 48.5" N	79° 52' 49.8" E
5.	AKT/VC-05 (2011)	<i>Pyrus communis</i>	Garmehel	Champawat	2060	29° 22' 56.2" N	80° 00' 43.8" E
6.	AKT/VC-06 (2011)	<i>Pyrus communis</i>	Garmehel	Champawat	1950	29° 23' 54.0" N	80° 09' 46.7" E
7.	AKT/VC-07 (2011)	<i>Pyrus communis</i>	Chusnee	Champawat	1935	29° 21' 55.4" N	80° 10' 40.1" E
8.	AKT/VC-08 (2011)	<i>Pyrus communis</i>	Kakariya	Champawat	1950	29° 23' 14.3" N	80° 04' 28.0" E
9.	AKT/VC-09 (2011)	<i>Pyrus communis</i>	Kakariya	Champawat	1720	29° 23' 14.9" N	80° 04' 30.0" E
10.	AKT/VC-10 (2011)	<i>Pyrus communis</i>	Jagnel	Champawat	1720	29° 23' 14.9" N	80° 04' 30.0" E
11.	AKT/VC-11 (2011)	<i>Pyrus communis</i>	Garhmehel	Champawat	1720	29° 23' 14.9" N	80° 04' 30.0" E
12.	AKT/VC-12 (2011)	<i>Pyrus communis</i>	Garhmehel	Champawat	1720	29° 23' 14.9" N	80° 04' 30.0" E
13.	AKT/VC-13 (2011)	<i>Pyrus communis</i>	Garhmehel	Champawat	1845	29° 23' 15.6" N	80° 04' 36.0" E
14.	AKT/VC-14 (2011)	<i>Pyrus communis</i>	Garhmehel	Champawat	2060	29° 23' 14.1" N	80° 04' 18.0" E
15.	AKT/VC-15 (2011)	<i>Pyrus communis</i>	Kakariya	Champawat	2060	29° 23' 14.1" N	80° 04' 18.0" E
16.	AKT/VC-16 (2011)	<i>Pyrus communis</i>	Jagnel	Champawat	2060	29° 23' 14.1" N	80° 04' 18.0" E
17.	AKT/VC-17 (2011)	<i>Pyrus communis</i>	Garhmehel	Pithoragarh	1850	29° 37' 11.0" N	80° 15' 38.4" E
18.	AKT/VC-18 (2011)	<i>Pyrus communis</i>	Kakariya	Pithoragarh	1970	29° 37' 31.9" N	80° 16' 26.4" E
19.	AKT/VC-19 (2011)	<i>Pyrus communis</i>	Jagnel	Pithoragarh	1935	29° 38' 25.4" N	80° 18' 32.6" E
20.	AKT/VC-20 (2011)	<i>Pyrus communis</i>	Jagnel	Pithoragarh	1740	29° 36' 55.8" N	80° 11.4' 52" E
21.	AKT/VC-21 (2011)	<i>Pyrus communis</i>	Garhmehel	Pithoragarh	1740	29° 36' 55.8" N	80° 11.4' 52" E
22.	AKT/VC-22 (2011)	<i>Pyrus communis</i>	Garhmehel	Pithoragarh	1740	29° 36' 55.8" N	80° 11.4' 52" E
23.	AKT/VC-23 (2011)	<i>Pyrus communis</i>	Garhmehel	Pithoragarh	1740	29° 36' 55.8" N	80° 11.4' 52" E
24.	AKT/VC-24 (2011)	<i>Pyrus communis</i>	Kakariya	Pithoragarh	1640	30° 00' 00.0" N	80° 54' 22.5" E
25.	AKT/VC-05 (2012)	<i>Pyrus communis</i>	Gola	Champawat	1350	29° 03' 38.9" N	80° 07' 8.2" E
26.	AKT/VC-06 (2012)	<i>Pyrus communis</i>	Garmehal	Champawat	887	29° 13' 32.5" N	80° 02' 53.0" E
27.	AKT/VC-07 (2012)	<i>Pyrus communis</i>	Garmehal	Champawat	820	29° 21' 55.4" N	80° 10' 40.1" E
28.	AKT/VC-16 (2012)	<i>Pyrus communis</i>	Garmehal	Pithoragarh	1787	29° 42' 52.1" N	80° 02' 34.1" E
29.	AKT/VC-17 (2012)	<i>Pyrus communis</i>	Jagnel	Pithoragarh	1781	29° 43' 32.1" N	80° 02' 12.9" E
30.	AKT/VC-18 (2012)	<i>Pyrus communis</i>	Jagnel	Pithoragarh	1781	29° 43' 32.1" N	80° 02' 12.9" E
31.	AKT/VC-19 (2012)	<i>Pyrus communis</i>	Garmehal	Pithoragarh	1722	29° 47' 46.3" N	80° 02' 19.9" E
32.	AKT/VC-20 (2012)	<i>Pyrus communis</i>	Garmehal	Pithoragarh	1292	29° 50' 11.4" N	80° 10' 58" E
33.	AKT/VC-21 (2012)	<i>Pyrus communis</i>	Jagnel	Pithoragarh	1360	29° 46.06' 3" N	80° 19.44' 9" E
34.	AKT/VC-22 (2012)	<i>Pyrus communis</i>	Gola	Pithoragarh	1360	29° 46.06' 3" N	80° 19.44' 9" E
35.	AKT/VC-23 (2012)	<i>Pyrus communis</i>	Gola	Pithoragarh	793	29° 47' 48.2" N	80° 24' 18.3" E
36.	AKT/VC-16 (2012)	<i>Pyrus communis</i>	Gola	Pithoragarh	809	29° 47' 51.5" N	80° 24' 27.9" E
37.	AKT/VC-24 (2012)	<i>Pyrus communis</i>	Garmehal	Pithoragarh	818	29° 52' 37.3" N	80° 33.44' 7" E
38.	AKT/VC-25 (2012)	<i>Pyrus communis</i>	Gola	Pithoragarh	820	29° 52' 37.3" N	80° 33.44' 7" E

Results and Discussion

The list of 38 genotypes of pear collected from cultivated habitats of surveyed area are shown in Table 1. Genotypes collected show upright or pyramidal growth habit with more vertical branching which helps in proper light interception and growth of plants. Diversity in precocity of flowering ranged from early to late and intensity in flowering ranged medium to high in most of the accessions collected. Variability in morphological characters is considered as an important prerequisite for

characterization and evaluation of pear genotypes for breeding and research. Therefore, variability available with respect to these traits was collected up to maximum possible extent.

On the basis of flowering time, genotypes of pear are classified into three main groups, viz., early, medium and late flowering types (Table 2). Although, flowering time

is under phylogenetic control (Jung and Muller, 2009), variability exists in flowering period (Jung and Muller, 2009). Flowering time is also affected by environmental factors like mean temperature, differences in chilling requirement for breaking bud dormancy, age and vigour of tree as well as natural factors/ calamities like heavy rainfall and hail storms. Variation in other quantitative

Table 2. Growth habit, flowering and fruiting characters of pear germplasm collected during exploration

S. No.	Vernacular name	Growth habit	Precocity of flowering*	Fruit shape	Fruit colour	Ripening time **
1.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
2.	Babboogosa	Pyramidal	Late	Pear shaped	Green to light yellow	Late
3.	Garmehel	Upright	Early	Round	Yellow with brown russet	Late
4.	Garmehel	Upright	Early	Round	Yellow with brown russet	Late
5.	Garmehel	Upright	Early	Round	Yellow with brown russet	Late
6.	Garmehel	Upright	Early	Round	Yellow with brown russet	Late
7.	Chusnee	Upright	Early	Pear shaped	Smooth green to yellow	Early
8.	Kakariya	Pyramidal	Late	Oblong	Green to light yellow	Late
9.	Kakariya	Pyramidal	Late	Oblong	Green to light yellow	Late
10.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
11.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
12.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
13.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
14.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
15.	Kakariya	Pyramidal	Late	Oblong	Green to light yellow	Late
16.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
17.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
18.	Kakariya	Pyramidal	Late	Oblong	Green to light yellow	Late
19.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
20.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
21.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
22.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
23.	Garhmehel	Upright	Early	Round	Yellow with brown russet	Late
24.	Kakariya	Pyramidal	Late	Oblong	Green to light yellow	Late
25.	Gola	Upright	Early	Round	Dark brown	Early
26.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
27.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
28.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
29.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
30.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
31.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
32.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
33.	Jagnel	Pyramidal	Medium	Pear shaped	Green to light yellow	Medium
34.	Gola	Upright	Early	Round	Dark brown	Early
35.	Gola	Upright	Early	Round	Dark brown	Early
36.	Gola	Upright	Early	Round	Dark brown	Early
37.	Garmehal	Upright	Early	Round	Yellow with brown russet	Late
38.	Gola	Upright	Early	Round	Dark brown	Early

* Precocity of flowering: early- flowering in April second fortnight, medium-flowering in May first fortnight, late-flowering in May second fortnight.

** Ripening time: early-June second fortnight, medium-July first fortnight, late-August first fortnight.

and qualitative characteristics of fruit like nutritional quality traits viz., TSS, total sugars, vitamins C and texture/ post-harvest life also exist among genotypes (Table 3). Post-harvest life is a crucial trait for collection of genetic variability of fruits which are to be grown in remote areas (Sharma *et al.*, 2012). In conformity with the present findings Muratovic *et al.* (1990) have also emphasized that high yielding cultivars of pear should be selected and conserved in field in mountainous areas

for crop improvement. Exploration and collection of germplasm and morphological characterization provides a base and raw picture of germplasm of specific plant species about variability and genetic diversity. These characters can be used as a base for further study at molecular level.

The green revolution has resulted in farmers planting fewer varieties of a plant that they grow so that they can focus on use of high yielding varieties. In addition,

Table 3. Fruit size, fruit weight and fruit quality traits of *Pyrus* spp. germplasm collected during exploration

S. No.	Vernacular Name	Fruit size		Fruit weight (g)	Total soluble solids ($^{\circ}$ Brix)	Total sugars (%)	Vitamin 'C' (mg/ 100 ml juice)	Organoleptic evaluation (1-10 Scale)	Post harvest Life (days)
		Length (cm)	Width (cm)						
1.	Jagnel	6.2	4.1	225	13.5	8.5	3.2	8.5	8
2.	Babboogosa	5.8	5.3	265	12.2	8.8	4.2	7.5	13
3.	Garmehel	4.8	4.1	210	8.5	9.6	4.1	5	7
4.	Garmehel	4.7	3.8	200	9.0	9.3	4.6	5	5
5.	Garmehel	5.0	4.2	195	9.6	10.0	4.5	5	5
6.	Garmehel	5.1	4.3	205	9.5	10.2	4.8	5	6
7.	Chusnee	3.6	2.6	65	14.5	12.2	2.3	9	5
8.	Kakariya	7.6	5.3	270	12.5	9.6	2.4	7.5	11
9.	Kakariya	8.2	5.6	255	12.8	10.0	2.7	7.5	13
10.	Jagnel	6.0	4.6	185	11.8	8.4	3.4	8.5	9
11.	Garhmehel	4.6	4.1	180	9.6	8.6	4.8	5	6
12.	Garmehel	4.9	4.3	195	10.0	8.9	5.1	5	8
13.	Garhmehel	5.3	4.5	210	10.3	9.0	5.0	5	8
14.	Garhmehel	5.2	4.4	220	10.8	9.0	5.3	5	9
15.	Kakariya	8.0	6.1	280	12.6	10.0	2.9	7.5	12
16.	Jagnel	6.2	4.6	210	11.4	8.7	3.0	8.5	10
17.	Garhmehel	5.3	5.0	180	8.8	10.2	5.6	5	8
18.	Kakariya	7.6	5.8	290	12.2	11.0	3.1	7.5	10
19.	Jagnel	5.9	4.3	240	10.6	8.3	2.8	8.5	9
20.	Jagnel	6.1	4.6	200	10.2	8.8	2.6	8.5	10
21.	Garhmehel	5.2	5.0	180	8.5	9.8	4.3	5	7
22.	Garhmehel	5.1	4.6	190	8.8	9.7	4.8	5	7
23.	Garhmehel	5.0	4.9	210	9.4	9.6	4.6	5	8
24.	Kakariya	8.2	6.7	285	11.6	10.6	3.0	7.5	12
25.	Gola	4.3	3.6	85	10.2	7.2	4.9	4	10
26.	Garmehal	5.4	3.8	170	10.6	10.5	4.9	5	8
27.	Garmehal	5.4	4.1	190	10.8	10.8	4.9	5	9
28.	Garmehal	5.0	4.0	175	11.0	9.8	5.0	5	7
29.	Jagnel	6.6	4.6	160	13.2	8.6	3.1	8.5	9
30.	Jagnel	6.4	4.5	180	12.8	9.2	2.5	8.5	10
31.	Garmehal	4.7	3.9	210	9.6	11.1	5.2	5	7
32.	Garmehal	5.2	4.3	200	10.2	11.4	4.7	5	7
33.	Jagnel	6.0	4.6	160	11.0	9.7	3.4	8.5	9
34.	Gola	4.3	4.2	90	10.2	7.8	5.8	4	10
35.	Gola	4.0	4.5	100	10.8	8.6	6.3	4	11
36.	Gola	4.4	3.3	75	11.4	8.2	5.2	4	12
37.	Garmehal	5.0	4.1	160	8.8	10.3	4.8	5	8
38.	Gola	4.5	3.9	110	11.6	8.5	5.9	4	11
Mean $\pm$ S.E.		5.54 ± 0.47	4.47 ± 0.31	189.73 ± 13.73	10.81 ± 0.97	9.48 ± 0.88	4.20 ± 0.37	6.10 ± 0.59	8.78 ± 0.77
Range		3.6-8.2	2.6-6.7	65-290	8.5-14.5	7.2-12.2	2.3-6.3	4.0-9.0	5-13
LSD ≤ 0.05		2.4	1.3	21	2.6	3.1	0.90	1.2	1.8

the varieties that are planted have been bred to a high degree of genetic uniformity within each variety. Both of these approaches are a change from past practices, in which farmers planted a large number of different, often locally-adapted, varieties, each of which generally contained a large number of different genotypes. Moreover, valuable genetic resources of plants especially fruits are eroding rapidly which results in large scale depletion of variability. There are many reasons for such depletion including deforestation, road laying, urbanization and land slides in mountainous terrain. Genetically uniform modern varieties are replacing the highly diverse primitive cultivars/landraces in traditional agro-ecosystem even in far and remote areas. Breeders are much interested in germplasm having early ripening time, big fruit size, more weight per fruit, attractive shape, appealing colour and high organoleptic ranking. These traits were taken into consideration during the survey and collection of *Pyrus* spp. germplasm (Table 3). Conservation of indigenous fruit species serves as a stock and source gene bank for fruit breeding. Low level of variability in cultivated pear genotypes is alarming the need to widen the genetic base to conserve an adequate level of genetic diversity for breeding programmes (Wunsch and Hormaza, 2007). Cultivation of primitive cultivars having resistance to abiotic and biotic stresses as well as desirable fruit quality traits is in decreasing trend. Therefore, there is an urgent need to collect the available genetic diversity and conserve in the field genebank for use in crop improvement programme and other research purposes.

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Elite Wheat Genetic Stocks for Improvement of End Product Quality in Wheat (*Triticum aestivum* L.)

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Plant genetic resources for quality components are most vital for orienting the breeding programme for developing superior end product quality wheat cultivars. Majority of superior quality wheat genotypes are in non-adaptive background. VL 858 (IC 546940), developed through selection from a cross OPATA/RAYON//KAUZ has excellent chapati quality. VL 852 (IC 565011), developed through pedigree method of selection from a winter x spring wheat cross Al Frog#4/HD 30// CPAN 3031 has excellent *chapati* quality. VL 876 (IC 565010), developed through pedigree method of selection from a winter x spring wheat cross SPINEBILL 'S'/CPAN 2045 has high bread loaf volume (581 ml), good quality ratio of bread loaf volume (ml)/dough weight (g) and very good bread quality. These genetic stocks are in well adapted background. To safe guard these national resources with respect to Intellectual Property Rights these genetic stocks were registered with NBPGR, New Delhi. Diversification of germplasm using these genetic stocks by involving them in hybridization would prove useful in improving quality components in wheat improvement programme.

Key Words: Bread loaf volume, Bread quality, *Chapati* quality, Germplasm, Quality

Introduction

Wheat (*Triticum aestivum* L.) is the most important winter cereal crop of India. India has witnessed phenomenal increase in wheat production and is now the second largest producer of wheat in the world. Recently the quality of wheat has been of great concern and there is demand for specific quality traits for suitable end products. Bread wheat has different end product uses like chapati, bread, biscuits etc. Specific quality traits need to be taken care in a breeding programme for *chapati*, bread and biscuits. For a successful breeding programme, it is essential to have germplasm for novel genetic variability and its judicious utilization. These novel sources can be used as donors in developing new varieties. The donors become more useful when they are in desirable genetic background. This investigation aims to study, elite wheat genetic stocks for quality components developed by VPKAS and registered at the national level.

Materials and Methods

Grain samples received from different test sites of All India Coordinated Wheat Improvement Programme (to avoid G x E interactions) were analyzed at Quality

laboratory of Directorate of Wheat Research, Karnal for their suitability for various end products. The following procedures were followed for assessing *chapati* and bread quality.

Chapati quality: For chapati quality evaluation, various parameters like water absorption, dough nature and colour (before and after maturation), chapati appearance and its colour, aroma, pliability, taste, puffing height and water loss just after baking and after 4 hours of baking were considered and the score was given out of 10. The chapati quality is considered poor, average, good, very good and excellent when the score are <5.0, 5.1-6.0, 6.1-7.0, 7.1-8.0 and >8.0, respectively.

Bread loaf volume and bread quality: For bread quality evaluation, parameters like loaf volume, stickiness, appearance, crust colour, crumb colour, texture, taste and aroma are considered and the score is in a scale of 0-10. Among all these parameters, loaf volume is considered most important and is given maximum weightage while evaluating bread quality. The loaf volume is considered poor, average, good, very good and excellent when the scores are <500, 501-600, 601-700, 701-800 and >800,

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respectively. The bread quality is considered poor, average, good, very good and excellent when the score are <5.0, 5.1-6.0, 6.1-7.0, 7.1-8.0 and >8.0, respectively.

Results and Discussion

Chapati quality data of VL 858 and VL 852 are presented in Table 1 and 2, respectively, whereas, bread quality data of VL 876 are presented in Table 3. All the three genetic stocks have been registered by the Plant Germplasm Registration Committee of Indian Council of Agricultural Research, NBPGR, New Delhi.

VL 858 (IC 546940; INGR 07002): a genetic stock for excellent chapati quality

VL 858, an elite wheat genetic stock has been developed through selection from a cross OPATA/RAYON//KAUZ. It has excellent *chapati* quality (8.11 on 0 to 10 scale) under irrigated and very good (7.83 on 0 to 10 scale) under rainfed conditions (Table 1) (Gupta *et al.*, 2003; 2004). In addition to excellent *chapati* quality, VL 858 possesses high hectoliter weight (79.15 and 80.43 kg/hl), high carotene (4.6 and 4.1), iron (33.0 and 36.6), zinc (3.5 and 24.0) and manganese (57.4 and 29.7) ppm under rainfed and irrigated conditions, respectively. It is a spring wheat, having intermediate growth habit and green coleoptile. The average plant height is 95–100 cm. It takes an average of 160-170 days to mature. It has dull white colour ears with amber coloured, ovoid shape grains and 38-39 g for thousand-grain weight. It was tested in the All India Coordinated yield evaluation trials of Northern Hills Zone from 2002-03 to 2004-05 and yielded a mean grain yield of 28.0 and 45.3 q/ha under rainfed and irrigated conditions, respectively. In addition it possesses high degree of field resistance to yellow and brown rust diseases. Further, it was also tested in plains of Uttarakhand in the State Varietal Trials from 2005-06 to 2007-08 and yielded 47.0 q/ha out yielding checks PBW 343 and UP 2338 by 5.05 and 3.59%, respectively, under irrigated conditions. Therefore, besides the quality parameters, this genotype carries very good yield potential.

VL 852 (IC 565011; INGR 09056): a diverse source of excellent chapati quality

VL 852, an elite wheat genetic stock has been developed through pedigree method of selection from a winter x spring wheat cross Al Frog#4/HD 30//CPAN 3031. VL 852 has excellent *chapati* quality (8.17 on 0 to 10 scale) under late sown with pre-sown irrigation conditions

of Northern hills zone (Table 2) (Gupta *et al.*, 2002, 2003, 2004). In addition, it possesses high hectoliter weight (79.67 kg/hl) and protein content (11.96%). It is a spring wheat, having semi-erect growth habit and green coleoptile. It's average plant height is 75-85 cm. It takes an average 135-144 days to mature. It has white colour ears with amber coloured ovoid shape grains and 39 g thousand-grain weight. It was tested in All India Coordinated yield evaluation trials of Northern Hills Zone from 2001-02 to 2003-04 and recorded a mean grain yield of 28.0 q/ha under late sown with pre-sown irrigation conditions.

VL 852 possesses high degree of field resistance to yellow and brown rust diseases. It is a true slow rust line (AUDPC=101-200) against yellow and brown rust (Gupta *et al.*, 2002, 2003) and thus, infers long lasting field resistance.

VL 876 (IC 565010; INGR 09055): high bread loaf volume and good quality bread

VL 876, an elite wheat genetic stock has been developed through pedigree method of selection from a winter x spring wheat cross, SPINEBILL 'S'/CPAN 2045. VL 876 has high bread loaf volume (581 ml), with the good quality ratio of bread loaf volume (ml)/dough weight (g) of 3.47 and very good bread quality (7.65 out of 10 scale) under early sown rainfed conditions (Table 3) (Gupta *et al.*, 2004, 2005, 2006). For evaluation of bread quality, the loaf volume is considered as the most important trait and is given maximum weightage. Bread loaf volume of >575 ml is highly desirable for good quality bread making. The ratio of bread loaf volume (ml)/dough weight (g) is considered important, while evaluating bread quality and a ratio of > 3.5 is considered appropriate for good quality bread. VL 876 is facultative wheat, having spreading growth habit and green coleoptile colour. Its average plant height is 90-105 cm. It takes an average 186-198 days to maturity. It has white colour ears with amber coloured grain, ovoid shape and 38 g thousand-grain weight. It has been tested in the All India Coordinated yield evaluation trials of the Northern hills zone from 2003-04 to 2005-06 and yielded grain yield of 33.63 q/ha under early sown rainfed conditions. In addition to high bread loaf volume and very good bread quality, it possesses high hectoliter weight (79.5 kg/hl), protein content (12.44%), iron (35.95), zinc (45.2) and copper (4.28) ppm under early sown rainfed conditions. It also possesses HMWGS 5+10 at Glu-D1 with Glu score 9,

Table 1. Comparison of VL 858 with the elite wheat cultivars for chapati quality

Production condition	Year	VL 858	HS 240	VL 738	Sonalika	VL 804
Timely sown Rainfed low Fertility	2003-04	7.64	7.27	7.09	7.22	7.16
	2004-05	8.01	7.21	7.53	-	7.24
	Mean	7.83 (Very Good)	7.24	7.31	7.22	7.20
Timely sown Irrigated high Fertility	2003-04	8.13	7.14	7.13	7.18	6.84
	2004-05	8.09	7.44	7.29	-	7.09
	Mean	8.11 (Excellent)	7.29	7.21	7.18	6.97

Table 2. Comparison of VL 852 with the elite wheat cultivars for chapati quality

Production condition	Years	VL 852 (P)	HS 295 (C1)	Sonalika (C2)	HS 420 (C3)
Late sown	2001-02	7.92	7.11	6.32	7.79
Restricted	2002-03	8.21	7.08	6.17	7.67
Irrigation	2003-04	8.38	7.02	6.67	7.47
	Mean	8.11 (Excellent)	7.07	6.39	7.64

Chapati score > 8 is considered as excellent for chapati quality.

Table 3. Comparison of VL 876 with the elite wheat cultivars for bread quality

Quality Characteristics	Years	VL 876 (P)	HS 277 (C1)	VL 616 (C2)	VL 829 (C3)
Bread Loaf Volume (ml)	2003-04	610	530	510	540
	2004-05	-	507	493	513
	Mean	581*	520	508.3	523.3
Bread Loaf Volume (ml)/ Dough weight (g)	2003-04	3.64	3.16	3.04	3.22
	2004-05	-	3.02	2.95	3.06
	Mean	3.47#	3.10	3.03	3.12
Bread Quality (Max score-10)	2003-04	8.37	6.05	5.62	6.63
	2004-05	-	5.44	5.03	5.55
	Mean	7.65\$	5.89	5.57	6.04

*Bread loaf volume > 575 ml highly desirable for good quality bread making.

#Bread loaf volume (ml)/ Dough weight (g)>3.5 is considered appropriate for a good quality bread.

\$Bread quality score between 7.1-8.0 (out of 10) is very good for good quality bread.

**VL 858- A genetic stock for excellent chapati quality****VL 852- A diverse source for excellent chapati quality**



VL 876- A genetic stock for high bread loaf volume and good quality bread

therefore, highly suitable for very good quality bread. Besides, VL 876 possesses high degree of field resistance to yellow and brown rust diseases.

The problem with majority of genetic stocks is that invariably they are in non-adoptive background which makes it difficult to use them in breeding programmes. VL 858, VL 852 and VL 876 are with very good agronomic background having rust resistance as well as

other desirable agronomic traits. Therefore, diversification of germplasm with these genetic stocks and involving them in future hybridization programme would prove useful in wheat improvement programme of India.

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Characterization of Sorghum Germplasm Collected from Gujarat

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The preliminary characterization of 107 accessions of sorghum landraces was carried out at DSR, Hyderabad and SDAU, Deesa (Gujarat) during *kharif* 2008 to 2010. The data on qualitative and quantitative descriptors were recorded using minimal descriptors. The accessions studied, exhibited good variability in both qualitative and quantitative traits. The preliminary characterization of sorghum landraces revealed that stem fresh and dry weight, plant height, ear head length and width, days to 50% flowering and days to maturity are the most variable characters. The qualitative characters have also shown high variability. High positive and significant correlation was observed among days to flowering, plant height, number of leaves/plant, stem thickness, stem fresh and dry weight, leaf length and days to maturity which help to identify early genotypes with high biomass. The positive significant correlation of grain yield/plant with 100-seed weight may help to select the high grain yielding genotypes. Based on the results of the mean over the environment (location x year) some potential accessions identified for significantly high brix per cent (E 147, E 151, ERN 22, E 148 and E159) for sweet sorghum breeding, the fodder lines with high stem fresh and dry weight (ERN 9, ERN 11, ERN 17, ERN 7 and ERN 16), for high grain yield (E 145, EJN 13, ERN 31 and EJN 31) and early maturity (EJN 7, EJN 9, EJN11, EJN 15 and EJN 24). These potential germplasm may be used in the forage, dual purpose, grain and sweet sorghum varietal improvement programme.

Key Words: Evaluation, Genetic Resource, Sorghum

Introduction

Sorghum [*Sorghum bicolor* (L.) Moench] is an important semi-arid crop which can tolerate any adverse climatic conditions. It originated and domesticated in Africa about 5,000-8,000 years ago (De Candolle, 1884). Indian subcontinent is the secondary centre of origin of this important cereal. Sorghum is popularly known as "Jowar" in India. It is a multipurpose and bioenergy crop; the grain, stem and glume are the useful parts. It is used as human food, livestock feed, in alcohol production, fuel, malt and other industrial productions (Elangovan, 2005). The crop in the country stands at the third place in context of importance after wheat and rice. Sorghum is cultivated in 7.53 mha, of which 2.89 mha is cultivated during rainy and *kharif* season with a production of 3.05 mt in India (Anonymous 2010). The major sorghum growing states are Andhra Pradesh, Gujarat, Karnataka, Madhya Pradesh, Maharashtra, Rajasthan and Tamil Nadu.

Plant genetic resources represent the inter and intra-specific reservoir of potentially useful genetic material. Landraces or farmer varieties constitute the basic material for developing any variety or hybrid. An autochthonous

landrace is a variety with a high capacity to tolerate biotic and abiotic stress, resulting in high yield stability and an intermediate yield level under a low input agricultural system. It is well established fact that the progress in improvement of a crop depends on the degree of variability in the desired character in the base material *vis-a-vis* germplasm collection. The study of relationships among quantitative traits is important for assessing the feasibility of joint selection of two or more traits and hence for evaluating the effect of selection for secondary traits on genetic gain for the primary trait under consideration. A positive genetic correlation between two desirable traits makes the job of the plant breeder easy for improving both traits simultaneously. High diversity in sorghum is distributed through out India, including Gujarat. Collection and characterization of sorghum germplasm is an important activity for identifying potential germplasm for utilization in the varietal improvement programme and to avoid duplication. Hence, in order to capture the landrace diversity from unsurveyed and surveyed areas, exploration was undertaken during 2008 to 2010 in Gujarat and a total of 107 accessions collected were characterized to access the variability and identify the promising accessions for different traits.

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Materials and Methods

One hundred seven sorghum genotypes collected from different district of Gujarat were evaluated at Directorate of Sorghum Research, Hyderabad and Sorghum Research Station, Sardarkrushinagar Dantiwada Agricultural University, Deesa (Gujarat) during *kharif* 2008 to *kharif* 2010. All the accessions were evaluated in Augmented Block Design with three check varieties *viz.*, CSV 15, SPV 462 and CSV 25 in each block. DSR, Hyderabad located at latitude of 27.19° N and longitude 78.24° E and at an elevation of 538 M msl and Deesa is situated at latitude of 24.5° N and longitude 72° E and at an elevation of 136 M msl. The soil of the field was sandy to deep sandy loam in texture with pH value of 7.5 to 8.5 having good physical and chemical properties. The experimental unit was a single-row plot of 6.75 m long, spaced at 0.60 m apart. NPK 120:40:00 fertilizers was applied as half basal dose of nitrogen and full dose of phosphorus at the time of sowing and half nitrogen applied after one month of sowing. Plots were thinned down after two weeks of crop emergence and plant-to-plant distance of 0.10 m was maintained. The experimental years showed different temperature regimes, humidity, rain fall and sunshine hours during the crop durations. All other recommended agronomical practices were followed to raise a good crop in both the seasons. The data on qualitative and quantitative characters were recorded using the minimal descriptor developed by NBPGR (Mahajan *et al.*, 2000) and list of sorghum descriptors released by Anonymous (1993). Five representative plants in each accession were tagged for recording the qualitative and quantitative characters. Brix percentage of each accession was estimated by refractometer. Descriptive statistical analysis was done for the quantitative characteristics.

Results and Discussion

The data of characterization and preliminary evaluation of 107 sorghum accessions recorded for 28 agromorphological and bio-chemical characters revealed a wide range of variability in both qualitative and quantitative characters. The range of variability and frequency observed in qualitative traits are given in Table 1. Majority of the accessions showed gray purple leaf pigmentation (66 acc.), dark green leaf colour (40 acc.), drooping leaf orientation (33 acc.), white midrib colour (60 acc.), semi compact earheads (54 acc.) and symmetric earhead shape (59 acc.), grayed red glume colour (56 acc.) with medium glume covering

Table 1. Characterization and preliminary evaluation of sorghum germplasm

S.No.	Characters	Variations	Frequency
1.	Seedling vigour	Good	20
		Poor	19
		Very good	33
		Unclassified	35
2.	Leaf pigmentation	Grayed purple	66
		Yellow green	41
3.	Leaf colour	Dark green	40
		Unclassified	67
4.	Leaf orientation	Drooping	33
		Erect	7
		Unclassified	67
5.	Midrib colour	Dull green	2
		Grayed purple	1
		White	60
		Yellow green	44
6.	Ear head shape	Panicle broader in lower part	9
		Panicle broader in upper part	1
		Pyramidal	19
		Reversed pyramid	10
		Symmetric	59
		Unclassified	9
7.	Ear head compactness	Compact	25
		Loose	7
		Semi compact	54
		Semi loose	11
		Very loose	1
		Unclassified	9
8.	Glume colour	Grayed orange	4
		Grayed purple	29
		Grayed red	56
		Yellow white	8
		Unclassified	10
9.	Glume covering	Long	22
		Medium	44
		Short	25
		Very long	6
		Very short	1
		Unclassified	9
10.	Presence of awns	Absent	24
		Present	78
		Unclassified	5
11.	Seed size	Bold	56
		Medium	9
		Small	1
		Very bold	1
		Unclassified	40
12.	Grain colour	Grayed orange	20
		Grayed white	3
		White	49
		Yellow orange	14
		Yellow white	14
		Unclassified	7
13.	Stay-green	Stay green	67
		Unclassified	40

S. No.	Characters	Variations	Frequency
14.	Races	Bicolor	5
		Caudatum	3
		Durra	37
		Durra bicolor	8
		Durra caudatum	9
		Guinea	2
		Guinea caudatum	1
		Kafir	2
15.	Lustrous	Unclassified	40
		Lustrous	10
		Non-lustrous	90
16.	Remarks	Unclassified	7
		Early flowering	7

(44 acc.), bold seeded (56 acc.), stay green (67 acc.) (non-senescence), grain colour white (49 acc.) and durra race (37 acc.). Sorghum landraces are consistent for morphological characters, and midrib color, grain color, grain size, glume color, glume hairiness, and grain shape were used by the farmers in naming the sorghum landraces (Teshome *et al.*, 1997).

The quantitative characters also showed wide variation in the evaluated sorghum germplasm. The results of descriptive statistical analysis are presented in Table 2. In quantitative traits, the days to 50% flowering

Table 2. Descriptive statistical analysis of quantitative characters in sorghum germplasm (over the environments)

Characters	Range pooled over the environment		Mean	SD	SE	Variance	CV(%)
	Minimum	Maximum					
Days to 50% flowering (days)	46	106	76	16.00	1.55	256.13	21.05
Number of leaves	5	21	15	3.54	0.34	12.53	23.60
Leaf length (cm)	50.33	89.80	71.76	9.16	0.89	83.90	12.76
Leaf width (cm)	2.52	11.00	7.95	1.37	0.13	1.88	17.23
Plant height (cm)	145	478	318	79.83	7.72	6373.57	25.10
Ear head length (cm)	7.93	55.00	18.72	9.63	1.04	92.81	51.44
Ear head width (cm)	0.65	11.06	5.33	1.86	0.20	3.45	34.89
Stem thickness (cm)	1.14	3.00	2.01	0.35	0.03	0.12	17.41
Stem fresh weight (g/plant)	120	2000	784	423.51	49.91	179364.61	54.02
Stem dry weight (g/plant)	30	1083	368	247.48	23.92	61247.24	67.25
Brix (%)	4	18	12	2.74	0.32	7.53	23.17
Grain yield (g/plant)	4	108	25	20.85	2.03	434.51	83.40
100-seed weight(g)	1.12	3.08	1.92	0.45	0.06	0.20	23.43
Days to 85 maturity (days)	165	118	16.25	1.78	263.97	13.77	

(46 – 106 days), plant height (145 – 478 cm), number of leaves per plant (5 - 21), leaf length (5.33 - 89.80 cm), leaf width (2.52 – 11.00 cm), earhead length (7.93 - 55 cm), earhead width (0.65 – 11.06 cm), stem thickness (1.134 – 3.00 cm), stem fresh weight (120 – 2000 g/plant), stem dry weight (30 – 1083 g/plant), Brix (4-18 %) and grain yield (0.00 – 108 g/plant), 100-seed weight (1.12 – 3.08 g) and days to maturity (85 – 165 days) showed wider range. The preliminary characterization of sorghum landraces revealed that stem dry weight, stem fresh weight, plant height, ear head width and ear head length and 50% flowering and days to maturity are the most variable characters because they showed higher standard deviation and variance. The qualitative characters have also shown good variability. Earlier reports by Elangovan *et al.* (2007, 2009) and Jadav *et al.* (2011) have also showed the presence of variation for the quantitative traits in sorghum accessions. Appa Rao *et al.* (1999) evaluated over 4,000 accessions from 11 major sorghum growing states in India for morphological and agronomical characters. They found that days to flowering, plant height, panicle length, erect and compact panicles are more frequent. The present study also observed variability in the different agro-morphological characteristics.

Fodder yield and seed yield are the complex characters controlled by several components which reflect positive and negative effect on these traits. Thus, to achieve rational improvement in the yield and their component traits, knowledge of mechanism of correlation provides a basis for formulating suitable breeding methods for yield improvement. On the basis of pooled data, the genotypic and phenotypic correlation coefficients among different characters were almost similar, including stem fresh and dry weight, grain yield/plant and their related characters. Hence we are presenting here the genotypic correlation coefficient only (Table 3). High positive and significant correlation was observed among days to flowering with, plant height, number of leaves/plant, stem thickness, stem fresh and dry weight, leaf length and days to maturity which help to identify early genotypes with high biomass. The positive significant correlation of grain yield/plant with 100-seed weight may help to select the high grain yielding genotypes.

These traits also showed i-nter correlation with each other. Elangovan *et al.* (2009) and Jain *et al.* (2011) also reported similar type of associations for one or more traits in sorghum germplasm.

Table 3. Correlation coefficient in yield and yield contributed traits in sorghum germplasm (pooled)

Characters	DFL	NOL	LL	LW	PH	EHL	EHW	ST	SFW	SDW	BX	GY	SW	DM
Days to 50% flowering (days)	1.00													
Number of leaves	0.69**	1.00												
Leaf length (cm)	0.52**	0.37**	1.00											
Leaf width (cm)	0.46**	0.47**	0.53**	1.00										
Plant height (cm)	0.71**	0.73**	0.42**	0.46**	1.00									
Earhead length (cm)	-0.04	-0.07	0.41**	-0.21	-0.03	1.00								
Earhead width (cm)	-0.04	-0.01	0.54**	0.19	-0.02	0.41**	1.00							
Stem thickness (cm)	0.57**	0.51**	0.68**	0.73**	0.46**	0.17	0.33**	1.00						
Stem fresh weight (g)	0.69**	0.61**	0.64**	0.50**	0.62**	0.03	-0.04	0.56**	1.00					
Stem dry weight (g)	0.69**	0.54**	0.61**	0.47**	0.51**	0.00	0.22	0.61**	0.94**	1.00				
Brix (%)	0.16	0.20	0.10	-0.06	0.15	-0.06	-0.27	0.08	0.17	0.21	1.00			
Grain yield (g/plant)	-0.29	-0.09	-0.19	0.20	-0.02	0.09	0.08	-0.08	-0.20	-0.34**	-0.26	1.00		
100-seed weight (g)	0.09	0.00	0.04	0.14	-0.07	-0.11	0.03	0.08	0.26	0.22	-0.01	0.35**	1.00	
Days to maturity (days)	0.54**	0.41**	0.45**	0.42**	0.50**	-0.05	0.31**	0.55**	0.27	0.47**	0.12	-0.13	-0.08	1.00

Based on the results of the mean over the environment (location x year) some of the accessions identified as positional donors for different characters are presented in Table 4. These accessions showed significantly higher mean values over the environments than the best check. The identified positional donors for the high brix per cent are E 147 and E 151 with 18 % ; ERN 22, E 148 and E 159 with 17 % for sweet sorghum breeding; the fodder line with stem fresh and dry weight are ERN 9 (2000 and 1083.3 g/plant), ERN 11 (1833.33 and 1083.3 g/plant), ERN 17 (1583.3 and 1000 g/plant), ERN 7 (1666.7 and 1000 g/plant) and ERN 16 (1566.7 and 666.7 g/plant). The high grain yield accessions are E 145(108 g/plant), E (73

g/plant), EJN 13 (69 g/plant), ERN 31(67 g/plant) and EJN 31(66 g/plant). Some accessions viz., E 145 (3.1 g), E 150(2.8 g), ERN 3 (2.8 g), ERN 24(2.7 g) and E 176 (2.6 g) showed high 100-seed weight. The early maturing entries were EJN 7, EJN 9, EJN 11, EJN 15 and EJN 24 which matured in 85 days. These potential germplasm may be used in the forage, dual purpose, grain and sweet sorghum varietal improvement programme.

As a prerequisite for efficient utilization of the germplasm, it must be properly evaluated, characterized and documented with a workable retrieval system so that any group of entries carrying any desired characteristics could be easily retrieved and used in breeding programme

Table 4. Five top ranking accessions selected on the basis of average of different characters (Mean pooled over environments and significantly superior then the best check)

S. No.	Days to 50 % flowering	Plant height (cm)	Stem fresh weight (g)	Stem dry weight (g)	Grain yield / plant (g)	1000-Seed weight (g)	Days to maturity	Brix (%)
1	EJN 15 (IC 585185) 46	E 152 (IC 568369) 478	ERN 9 (IC 568524) 2000	ERN 9 (IC 568524) 1083.3	E 145 (IC 568362) 108	E 145 (IC 568362) 3.1	EJN 7 (IC 585177) 85	E 147 (IC 568364) 18
2	EJN 9 (IC 585179) 48	EJN 2 (IC 585172) 450	ERN 11 (IC 568526) 1833.33	ERN 11 (IC 568526) 1083.3	E 171 (IC 568388) 73	E 150 (IC 568367) 2.8	EJN 9 (IC 585179) 85	E 151 (IC 568368) 18
3	EJN 11 (IC 585181) 48	EJN 1 (IC 585171) 436	ERN 17 (IC 568532) 1583.3	ERN 17 (IC 568532) 1000	EJN 13 (IC 585183) 69	ERN 3 (IC 568518) 2.8	EJN 11 (IC 585181) 85	ERN 22 (IC 568547) 17
4	EJN 4 (IC 585174) 50	ERN 6 (IC 568521) 435	ERN 7 (IC 568522) 1666.7	ERN 7 (IC 568522) 1000	ERN 31 (IC 568546) 67	ERN 24 (IC 568539) 2.7	EJN 15 (IC 585185) 85	E 148 (IC 568365) 17
5	EJN 7 (IC 585177) 50	ERN 8 (IC 568523) 432	ERN 16 (IC 568531) 1566.7	ERN 16 (IC 568531) 666.7	EJN 31 (IC 585201) 66	E 176 (IC 568393) 2.6	EJN 24 (IC 585194) 85	E 159 (IC 568376) 17

(Gebrekidan, 1982). The present study revealed sufficient genetic variability for quantitative, qualitative and morphological characters among the germplasm collected from Gujarat, which can be exploited for developing superior varieties and hybrids in sorghum. Different accessions have different promising traits. Therefore, a gene pool can be generated by crossing the germplasm lines of interest which can be further used as source material to develop promising lines in sorghum.

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Identification of Photosensitive Lines in Black Gram and their Characterization

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One hundred and sixty accessions of black gram (*Vigna mungo*) germplasm were characterized and evaluated for 22 qualitative and quantitative agro-morphological traits during *kharif* (rainy) and *rabi* (post-rainy) seasons of 2005 and 2006. Two accessions, viz., IC343880 and IC426765 did not flower during the rainy season of 2005-06 and 2006-07, while they produced flowers and pods in the successive post-rainy season and, therefore, were identified as photosensitive. These two accessions were studied for their qualitative and quantitative traits in comparison with the check varieties (LBG-20, T-9 and PU-19). A comparative account of morphological traits of these identified germplasm lines with that of published literature on photosensitive released variety (LBG-17) are also provided. The uses of these potential genotypes in black gram improvement as also their alternative use as green manure and cover crop, are discussed.

Key Words: Black gram, Germplasm, Photosensitivity, *Vigna mungo*

Introduction

Black gram [*Vigna mungo* (L) Hepper] is an important short duration pulse crop grown in many parts of India, in an area of about 3.18 m ha with a total production of 1.47 million tons with an average productivity of 461 kg/ha. Andhra Pradesh leads with the highest area and production among states with an annual production of 3.5 lakh tons from 4.97 lakh has with an average productivity of 704 kg/ha (www.dacnet.nic.in/pulses/APR,06-07.pdf).

Most of the varieties released are photo-insensitive and, therefore, can be grown during any time of the year. However, one photosensitive line (LBG-17) is known to exist in black gram, which flowers under short-day conditions (NORV database, www.nbpgr.ernet.in). There is a need to identify more photosensitive lines in order to have parental diversity for varietal development, keeping the location specific requirements like combining resistance to numerous pests and diseases and other desirable traits (seed quality, plant type and photoperiod sensitivity) as reported in cowpea (Timko and Singh, 2008)

Material and Methods

One hundred and sixty accessions of black gram germplasm lines, collected during exploration from different agro-climatic zones of Andhra Pradesh, India,

were grown at the NBPGR RS experimental farm (17° 19'N, 78° 23'E, 542 m above msl) Hyderabad, India, during rainy 2005, 2006 and post-rainy seasons 2005-06, 2006-07 respectively. The crop was raised in Augmented Block Design (ABD) with three checks viz. T-9, LBG-20 and PU-19. The crop was raised following standard package of practices (Anonymous, 1991). The qualitative and quantitative traits as per minimal descriptors (Srivastava *et al.*, 2001) were recorded. Freely downloadable Web Agri Stat Package (WASP2) developed by ICAR Research Complex Goa, was used for statistical analysis (<http://icargoa.res.in/wasp2.0/index.php>).

Result and Discussion

Wide range of variation was observed for days to 50% flowering during rainy (34 to 52 days) and post-rainy (46 to 60 days) seasons. Interestingly, two accessions, IC343880 (SKN-71) and IC426765 (BAR-62), did not flower at all during the rainy seasons of both the years. However, these two accessions exhibited 50% flowering by 46 and 49 days, respectively, during the post-rainy season and produced pods, indicating their photosensitive nature. The passport data of these accessions show that, accession IC343880 was collected from Pathapatnam (18°45' N & 84°05'E), in Srikakulam district of Andhra Pradesh during December 2001 (post-rainy crop) from farmer's field and IC426765 was collected from Mandasa

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(18°51' N & 84°05'E) in Srikakulam district in April 2003 (post-rainy crop).

The performance of these photosensitive lines was analyzed in comparison to released check varieties in the post-rainy 2005-06 and 2006-07 (Table 1). ANOVA revealed that there was significant difference ($p < 0.05$) among these photosensitive lines and released checks with respect to days to 50% flowering, 100-seed weight and pod length. In post rainy 2005-06, accession IC343880, performed better than checks T-9 and LBG-20 at CD 0.05 with respect to peduncle length. However, in post-rainy 2006-07, significant variation ($p < 0.05$) with respect to days to 50% flowering, plant height, pods per cluster, seeds per pod and 100-seed weight were recorded. Accession IC426765 performed better than LBG-20 with respect to plant height and 100-seed weight (CD 0.05) and IC343880 was superior to PU-19 with respect to seeds per pod. The superior performance of these accessions in some of the quantitative traits *viz.*, seeds per pod and 100-seed weight over released checks indicate their potential in the breeding programmes for black gram crop improvement.

The qualitative traits of these photosensitive lines and the local checks including a photosensitive variety, LBG-17 are presented in Table 2. Both the germplasm accessions (IC343880 and IC426765) had indeterminate growth habit and dense pod pubescence. These qualitative

characters indicate that they are primitive landraces. The comparative study further supports the fact that these photosensitive accessions are distinctly different from the local check varieties with respect to primary leaf shape, pod pubescence and leaf pubescence (see Table 2). These two accessions, IC343880 (SKN-71) and IC426765 (BAR-62), are new reports of photosensitive black gram germplasm lines. The dominance of photoperiod insensitivity over photoperiod sensitivity has already been reported (Verma, 1971) It is well documented that, the photosensitive trait in any plant is a primitive character as loss of photoperiodic response is considered to be a sign of domestication (Paroda *et al.*, 1988). Further, the alternate use of these accessions as a forage, cover crop and green manure crop can also be exploited during the rainy season by carrying out detailed studies on the biomass yield and nitrogen fixation in the field in view of the prolonged nodulation in the absence of reproductive phase. Disintegration of the nodules has been correlated with the onset of pod formation stage (Singh, 1974). Therefore, these lines can be utilized in the pulses breeding programmes for developing multipurpose varieties of blackgram suitable for different agro-climatic zones of Andhra Pradesh. As these photosensitive lines are adapted to a particular climatic condition, they perform the best under that condition and this quality should be effectively exploited by assessing these lines in alternative cropping systems.

Table 1. Performance of the photosensitive black gram germplasm lines as compared to released varieties

Blackgram/Traits		Year	1	2	3	4	5	6	7	8	9	10	
Photosensitive lines	IC-343880	2005	46.0	26.500	3.800	14.400	3.6	6.000	6.132	4.120	11.400	35.400	
		2006	49.0	14.400	3.800	9.200	2.800	5.400	2.900	4.120	5.620	18.200	
	IC-426765	2005	49.0	29.580	3.200	9.400	4.800	5.930	6.062	3.680	7.760	23.800	
		2006	49.0	11.440	2.400	5.600	2.400	3.600	3.560	3.820	3.840	11.000	
Released varieties	T-9	2005	46.2	27.088	2.200	7.452	3.664	5.926	5.746	4.168	8.188	14.464	
		2006	48.4	18.292	2.862	7.328	3.262	4.528	3.896	4.410	3.620	11.860	
	PU-19	2005	46.0	26.676	2.864	8.464	4.528	5.812	5.640	4.232	10.116	20.732	
		2006	47.2	17.206	2.930	7.194	2.528	4.262	3.336	4.308	3.992	13.128	
	LBG-20	2005	46.8	32.504	3.260	12.664	4.600	6.088	5.994	4.672	8.012	29.864	
		2006	46.2	19.792	3.660	7.930	3.530	4.460	2.832	4.082	4.756	10.730	
	CD (0.05)	2005	0.512	NS	NS	NS	NS	NS	NS	NS	0.361	2.653	NS
		2006	1.500	4.718	NS	NS	0.800	1.105	0.627	NS	1.141	NS	

Legend: 1. Days to 50% flowering; 2. Plant height (cm); 3. Primary branches per plant (no.); 4. Clusters per plant (no.); 5. Pods per cluster (no.); 6. Seeds per pod (no.); 7. 100-seed weight (g); 8. Pod length (cm); 9. Peduncle length (cm); 10. Pods per plant (no.).

Table 2. Comparison of photosensitive blackgram germplasm with released varieties

Trait	Photosensitive lines		Released varieties (local checks)			Released variety (photosensitive)
	IC343880	IC426765	T-9	PU-19	LBG-20	Krishnayya (LBG-17)
Plant habit	Indeterminate	Indeterminate	Determinate	Determinate	Determinate	Determinate
Primary leaf shape	Lanceolate	Ovate lanceolate	Deltoid	Lanceolate	Ovate	Ovate
Pod pubescence	Dense	Dense	Puberulant	Moderate	Puberulant	Moderate
Leaf pubescence	Moderate	Dense	Sparse	Sparse	Dense	Moderate
Leaf colour	Dark green	Dark green	Dark green	Green	Green	Light green
Primary leaf shape	Deltoid	Cuneate	Ovate lanceolate	Ovate	Ovate	Ovate
Seed colour	Black	Black	Dull black	Black brown	Shiny Black	Greyish shiny black

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Establishing Distinctiveness in Local Maize (*Zea mays* L.) Varieties using RAPD Marker as Additional Descriptor for Protection under PPV&FR Act

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The molecular identification profiles were established in the present work for 24 local maize varieties with 10 RAPD markers to incorporate the fingerprint, as complementary information to the standard registration data on DUS morphological descriptors. It resulted in 172 informative RAPD loci. UPGMA analysis of RAPD marker loci could discriminate all the varieties except Khusalpur Local-4 and Nunawala Local-5. These primers could establish unique molecular identification profiles (MIPs) for a total of 14 varieties. Mean polymorphic information content (PIC), average expected gene diversity, average resolving power (Rp) and diversity index (DI) of RAPD markers used were high which reflected that RAPD marker is an efficient tool to establish distinctiveness among the present set of experimental material. Principal component analysis (PCA) of RAPD data supported UPGMA clustering results. Thus, the present investigation reflects that molecular markers are new possibilities as additional descriptor to characterize the plant cultivars for registration purposes under PPV&FR Act.

Key Words: Diversity index, Maize, Molecular identification profile, PPV&FR Act, RAPD

Introduction

Maize (*Zea mays* L.) an allogamous crop is an important cereal used as food for human consumption, feed for animal and poultry and as an important source of edible oil and starch. In India during 2010, it occupied an area of 7.18 million hectares with production of 14.06 million tonnes and productivity of 1958.22 kg/ha (<http://faostat.fao.org>). India has an immense wealth of maize varieties growing in all the different agroclimatic regions. There is a need in the country to protect such a vast variability present in the species, which is conserved by farmers as local indigenous varieties. The matter of protection of varieties started only after an international body UPOV was established in Paris in 1961 and it entered into force in 1968. UPOV aimed to ensure protection of varieties by the grant of an exclusive right on the protected new plant variety on the basis of a set of principles (Dutfield, 2001). On the UPOV pattern India has enacted a *sui generis* form of protection as Protection of Plant Varieties and Farmers' Rights Act (PPV&FR Act), 2001. Under PPV&FR Act a variety must fulfill the criteria of distinctiveness, uniformity, stability (DUS) and novelty (if newly developed) so as to get protection under the Act (Anonymous, 2001). As per DUS guidelines of PPV&FR Act Authority there are 31 morpho-physiological DUS descriptors for maize which are species-specific and recommended procedures for conducting DUS trials

(Anonymous, 2007). Plant morphological descriptors have been the universally undisputed traits applied for DUS testing of crop varieties but serious problems may arise in future for establishing distinctiveness of variety only on morpho-physiological DUS descriptors especially in closely related local cultivars (Patra *et al.*, 2010). These morphological descriptors suffer from many drawbacks, such as influence of environment on trait expression, limited in number and increasing number of candidate varieties with decreased variability which enforces to look for alternatives (Joshi *et al.*, 2009). Biochemical markers *viz.* isozymes and storage proteins have been mentioned to complement morphological markers under UPOV guidelines (Anonymous, 1999). But results obtained from them may not be up to the mark by the general consideration since only a minor portion of the genome is represented by these markers (Stuber *et al.*, 1988). Genetic diversity studies on maize using traditional morphological and biochemical markers are common and routinely used. However, with the availability of large number of polymorphic molecular markers *viz.* RAPD, RFLP and SSRs created interest in their use for varietal identification (Krevich *et al.*, 1992; Mackill, 1995; Mc Gregor *et al.*, 2000). Among the diverse range of molecular marker techniques available for evaluating genetic diversity, RAPDs are well known for their potentially high information content and versatility

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(Pejic *et al.*, 1998; Singh *et al.*, 2009). RAPD analysis in aromatic and basmati rice varieties revealed high degree of polymorphism and were shown to complement DUS morphological descriptors in establishing distinctiveness of varieties (Patra *et al.*, 2010; Joshi *et al.*, 2010). In the present study PCR based molecular marker RAPD was used as additional descriptor to evaluate the genetic diversity and to develop DNA profiles of 24 local maize varieties representing different geographical locations of Uttarakhand. DNA fingerprinting has rendered genotype characterization highly efficient enabling reliable distinction of even closely related accessions. This study was conducted in anticipation, if morpho-physiological DUS descriptors are not able to discriminate closely related varieties, then molecular markers can be considered as additional descriptors for establishing the distinctiveness.

Material and Methods

Plant Material

A total of 24 maize varieties were selected; 22 varieties from wide range of geographical locations of Uttarakhand and two reference varieties from Karnal and Pantnagar were selected. The details of the cultivars are given in Table 1.

Molecular Characterization

The genomic DNA was extracted from 14-day-old etiolated seedlings following CTAB method (Doyle and Doyle, 1990) with minor modifications. The quantification of DNA in RNA free sample was done using a UV visible spectrophotometer (ELICO Ltd.). The quality and quantity of DNA was checked by 1% agarose gel electrophoresis using standard containing 100 ng/μl genomic DNA. RAPD analysis was performed in a 0.2 ml reaction tube in volume of 25 μl containing 10 X Assay Buffer, 0.5 unit of *Taq* DNA polymerase, 200 μm each of dNTPs, 50 ng/μl reaction of random primer and 50 ng of template DNA (standardized). Molecular characterization was conducted with 10 random primers as shown in Table 2. PCR was performed in 'Eppendorf Thermocycler' by initial denaturation at 94°C for 5 min followed by 39 cycles of denaturation at 94°C for one min, annealing at 32°C for one min, extension at 72°C for two min and final elongation at 72°C for 7 min. The PCR products were electrophoresed on 1.2% agarose gel, prepared in 1X TBE buffer containing 0.5 μg /ml of the ethidium bromide at 80V for 3h with cooling. The gel was photographed under

Table 1. Details of maize varieties studied with their origin

Sl. No.	Genotype	Areas of cultivation
1	Dharap Local-4	Nainital
2	Kirtinagar Local-4	Dehradun
3	Purvi Khera Local-1	Haldwani
4	Paschim Khera Local-2	Haldwani
5	Manacot	Garhwal
6	Khusalpur Local-1	Garhwal
7	Khusalpur Local-3	Garhwal
8	Khusalpur Local-4	Garhwal
9	Khusalpur Local-6	Garhwal
10	Nunawala Local-1	Haridwar
11	Nunawala Local-5	Haridwar
12	Nunawala Local-6	Haridwar
13	Doiwala Local-2	Dehradun
14	Bailparow Local-4	Nainital
15	Bhadarabad Local-4	Haridwar
16	Bhadarabad Local-5	Haridwar
17	Bhadarabad Local-8	Haridwar
18	Bagauli Local-1	Haridwar
19	Bishanpur Local-2	Haldwani
20	Khera Local	Haldwani
21	Chamoli Local	Dehradun
22	Quarali Local-1	Garhwal
23	Pragati	Pantnagar
24	HQPM 1	Karnal

UV transilluminator. The PCR reaction was repeated twice for each primer. The non-reproducible bands, weak or smeared bands were not counted and also not included in the comparative analysis. Profiles for each cultivar and marker system were constructed by scoring in a binary matrix as "0" and "1" for the absence and presence of fragments respectively and the final data sets included both polymorphic and monomorphic fragments. Cluster analysis was performed using SHAN module of the NTSYS-pc (Numerical Taxonomy System, version 2.0) (Rohlf, 2002). Similarities between varieties were estimated using the SIMQUAL program to calculate the Jaccard's coefficient by Unweighted Pair Group Method on Arithmetic Averages (UPGMA), a common estimator of genetic identity (Jaccards, 1908). Strength of the clusters was analyzed by bootstrap values calculated by using 1000 sampling with Win Boot Software (Yap *et al.*, 1995). It performs analysis of binary data to determine the confidence limits of UPGMA-based dendrogram. PIC that provides an estimate of the discriminatory power of a locus or loci, by taking into account, not only the number of alleles that are expressed, but also relative

Table 2. Details of RAPD primers used for the molecular characterization of 24 maize varieties

Sl.No.	Primer code	Primer sequence	Amplified product (Kb)	Total bands	Mono-morphic bands	Poly-morphic	% Polymorphism	PIC	Average expected gene diversity (Hi)	Resolving power (Rp)
1	P1	GTAGCACTCC	0.2 Kb- 2.0 Kb	17	5	12	70.6%	0.5	0.1	16.83
2	P2	TCGGCAGCA	0.3 Kb- 1.7 Kb	22	1	21	95.5%	0.4	0.3	19.58
3	P3	GTGTCTCAGG	0.3 Kb- 2.0 Kb	16	0	16	100%	0.7	0.2	22.08
4	P4	GTCCATGCCA	0.3 Kb- 2.0 Kb	16	1	15	93.8%	0.4	0.3	14.33
5	P5	ACATCGCCCA	0.3 Kb- 4.0 Kb	18	10	8	44.5%	0.3	0.1	9.00
6	P6	CCAGCCGAAC	0.3 Kb- 1.7 Kb	17	2	15	88.2%	0.5	0.3	16.00
7	P7	GGAAGCCAAC	0.2 Kb- 3.0 Kb	18	5	13	72.2%	0.4	0.2	15.92
8	P8	CCAAGCTGCC	0.3 Kb- 3.0 Kb	11	4	7	63.7%	0.5	0.1	10.92
9	P9	TGCGGCTGAG	0.3 Kb- 2.5 Kb	16	9	7	43.8%	0.2	0.1	5.17
10	P10	GACGGATCAG	0.4 Kb- 2.5 Kb	21	4	17	81%	0.4	0.2	17.58
Average				17.2	4.1	13.1	75.33	0.43	0.19	14.8

frequencies of those alleles was calculated using the formula $PIC = 1 - \sum p_i^2$, where p_i is the frequency of the i^{th} allele. The Rp to distinguish among the studied genotypes of a primer was calculated using the formula $Rp = \sum Ib$ where Ib is band informativeness. It takes the value of: $1 - [2 \times (0.5 - p)]$, p being the proportion of the 24 maize varieties containing the band (Torre *et al.*, 2006). The Jaccard's similarity matrix was subjected to principal component analysis (PCA) (Yadav *et al.*, 2010). The first three principal components were used for 3-dimensional plotting, amongst each other using module PROJ and MXPLOT of NTSYSpc. This method makes use of multi-dimensional solution of the observed relationships based on the genetic distance.

Results and Discussion

The molecular markers play a relevant role in determining cultivar diversity and hence leading to intellectual property rights protection. The accurate description of maize varieties and distinctiveness is crucial for registration under PPV&FR Act. The identity/profile of a variety is to be established by using a set of morphological characteristics prescribed in the DUS test guidelines on maize. On the basis of DUS morphological characters only 11 varieties could be discriminated. Thus, morpho-physiological DUS descriptors alone were not sufficient for establishing the distinctiveness especially in closely related varieties or similar indigenous local varieties grown in a particular niche. Hence, in the present study molecular markers were considered for establishing the distinctiveness of maize varieties.

RAPD Analysis

RAPD marker analysis was conducted on DNA extracted from the 24 varieties using 10 oligonucleotide random

primers. The total number of bands (TNB), number of polymorphic bands (NPB), percentage of polymorphic bands (P%), PIC, Rp and DI are shown in Table 2. All the primers amplified 172 RAPD loci (average of 17.2 bands per primer) across the 24 genotypes studied, out of which 131 loci were polymorphic and 41 were monomorphic. Amplification products ranged in size from 4.0 kb (by the primer P-5) to 0.2 kb (by the primers P-1 and P-7). Maximum number of 22 amplification products were obtained with the primer P-2, followed by 21 products with primer P-10 while minimum number of 11 RAPD loci were generated with primer P-8. One primer produced 100% polymorphism and 2 primers showed more than 90% polymorphism. However other primers together accounted for 75.3% polymorphism. The unique MIPs were created from different RAPD primers, which could identify the fourteen varieties. Out of 14 varieties, maximum number of 4 unique bands were observed in Bhadarabad Local-5 and Bishanpur Local-2 while Kirtinagar Local-4, Purvi Khera Local-1 and Khera Local showed 3 unique bands in each variety. Dharap Local-4, Nunawala Local-6, Doiwala Local-2 and Bagauli Local-1 showed 2 unique bands in each variety while Paschim Khera Local-2, Khusalpur Local-6, Bailparow Local-4, Chamoli Local and HQPM 1 showed 1 unique band in each variety. The unique band appear in Doiwala Local-2 by P-5 is shown in Fig. 2. However, for ten varieties no unique MIPs were obtained by any of the primers studied. The size and number of these exclusive or genotypic specific bands amplified in the mentioned varieties are presented in Table 3. These unique bands generated in mentioned varieties reveal distinctiveness of varieties. The PIC value ranged from 0.2 for primer P-9

Table 3. Number and size of genotype specific bands amplified by RAPD markers in 24 maize varieties

Sl. No.	Varieties	RAPD		
		Primer code	No. of exclusive loci	Size of exclusive loci
1	Dharap Local-4	P3	1	0.4 Kb
		P10	1	0.45 Kb
		P2	1	1.4 Kb
2	Kirtinagar Local-4	P7	1	0.48 Kb
		P8	1	0.3 Kb
3	Purvi Khera Local-1	P1	1	0.55 Kb
		P3	1	0.70 Kb
		P10	1	0.65 Kb
4	Paschim Khera Local-2	P10	1	0.7 Kb
5	Manacot	-	-	-
6	Khusalpur Local-1	-	-	-
7	Khusalpur Local-3	-	-	-
8	Khusalpur Local-4	-	-	-
9	Khusalpur Local-6	P6	1	1.0 Kb
10	Nunawala Local-1	-	-	-
11	Nunawala Local-5	-	-	-
12	Nunawala Local-6	P9	1	0.35 Kb
		P10	1	0.95 Kb
13	Doiwala Local-2	P5	1	0.5 Kb
		P10	1	1.7 Kb
14	Bailparow Local-4	P8	1	0.45 Kb
15	Bhadarabad Local-4	-	-	-
16	Bhadarabad Local-5	P1	1	0.55 Kb
		P1	1	0.40 Kb
		P3	1	1.8 Kb
		P7	1	1.5 Kb
17	Bhadarabad Local-8	-	-	-
18	Bagauli Local-1	P1	1	1.5 Kb
		P7	1	0.5 Kb
19	Bishanpur Local-2	P1	1	0.65 Kb
		P4	1	0.35 Kb
		P7	1	0.9 Kb
		P8	1	0.7 Kb
20	Khera Local	P2	1	1.1 Kb
		P2	1	0.52 Kb
		P3	1	0.90 Kb
21	Chamoli Local	P6	1	1.1 Kb
22	Quarali Local- 1	-	-	-
23	Pragati	-	-	-
24	HQPM 1	P2	1	0.3 Kb

to 0.7 for primer P-3 with an average of 0.43 for all the ten primers which shows the ability of different primers to discriminate among the maize cultivars. 'Prevost and Wilkinson' reported the Rp as the capacity of a given primer to discriminate among different genotypes (Prevost A., 1999). Seven RAPD primers viz. P-1, P-2, P-3, P-4, P-6, P-7 and P-10 having high Rp of 16.83, 19.58, 22.08,

14.33, 16, 15.92 and 17.58, respectively, were able to discriminate majority of the varieties. The 3D depiction of PCA results are shown in Fig. 1. According to PCA results, the 24 varieties were well separated and RAPD data also well supported their UPGMA clustering. PC1, PC2 and PC3 accounted for 11.43%, 7.65% and 6.3% of the variation, respectively and the cumulative variation of these three principal components was 25.38%.

Genetic Relationships of Varieties

The Jaccard's coefficients for the genetic similarities among the 24 varieties are presented in Table 4. The similarity coefficient values varied from 0.57 between Purvi Khera Local-1 and Doiwala Local-2 to 0.85 between Khusalpur Local-4 and Nunawala Local-5. The

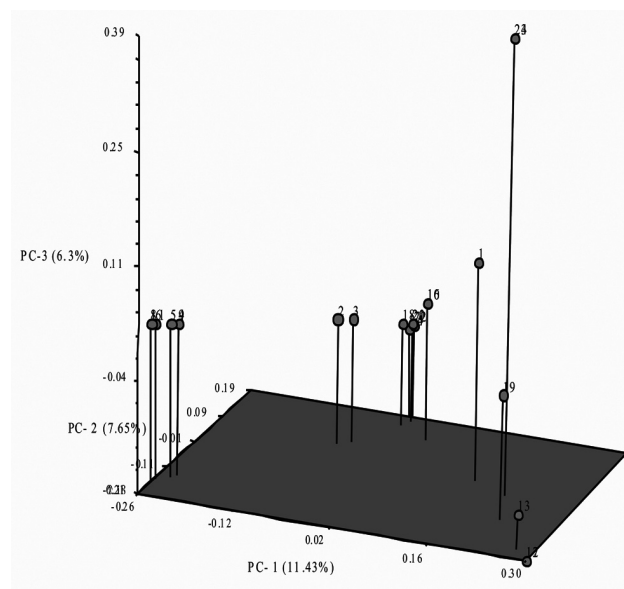


Fig. 1. Three dimensional plot based on Principal Components 1, 2 and 3 of 24 maize varieties using RAPD marker. The three axes represent the first 3 principal components (PC) (1-24: Names of varieties as mentioned in Table 1)

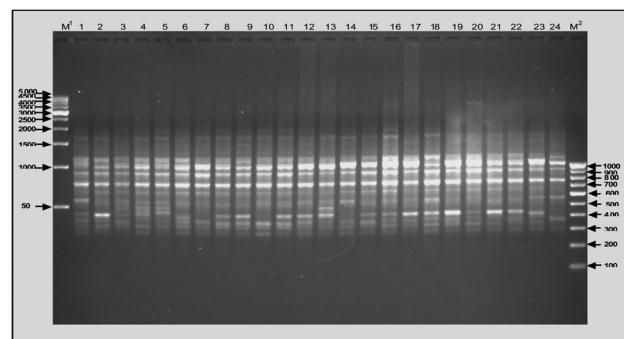


Fig. 2. Molecular diversity generated among the 24 maize varieties by RAPD primer P-5. (1-24: Names of varieties as mentioned in Table 1)

Table 4. Genetic similarity matrix of 24 maize varieties

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
1	1.00																							
2	0.70	1.00																						
3	0.66	0.74	1.00																					
4	0.68	0.74	0.72	1.00																				
5	0.71	0.71	0.70	0.82	1.00																			
6	0.75	0.76	0.75	0.79	0.81	1.00																		
7	0.68	0.68	0.73	0.74	0.70	0.79	1.00																	
8	0.72	0.76	0.75	0.82	0.84	0.84	0.81	1.00																
9	0.68	0.74	0.72	0.83	0.80	0.78	0.76	0.84	1.00															
10	0.68	0.69	0.68	0.68	0.71	0.76	0.74	0.77	0.71	1.00														
11	0.67	0.75	0.77	0.77	0.81	0.83	0.75	0.85	0.81	0.78	1.00													
12	0.66	0.63	0.68	0.68	0.69	0.72	0.68	0.69	0.64	0.69	0.68	1.00												
13	0.65	0.61	0.57	0.65	0.63	0.68	0.62	0.66	0.61	0.63	0.61	0.75	1.00											
14	0.69	0.67	0.64	0.69	0.73	0.70	0.74	0.77	0.70	0.66	0.71	0.69	0.67	1.00										
15	0.72	0.71	0.73	0.73	0.76	0.78	0.75	0.76	0.70	0.73	0.75	0.72	0.67	0.79	1.00									
16	0.62	0.64	0.66	0.69	0.73	0.73	0.71	0.71	0.73	0.73	0.77	0.68	0.58	0.69	0.76	1.00								
17	0.65	0.65	0.67	0.71	0.69	0.72	0.73	0.72	0.69	0.66	0.69	0.79	0.73	0.70	0.73	0.64	1.00							
18	0.65	0.67	0.68	0.71	0.74	0.79	0.73	0.76	0.71	0.69	0.76	0.68	0.65	0.72	0.77	0.74	0.71	1.00						
19	0.69	0.71	0.67	0.70	0.69	0.72	0.69	0.78	0.67	0.70	0.72	0.70	0.68	0.68	0.70	0.66	0.71	0.69	1.00					
20	0.61	0.67	0.69	0.71	0.72	0.75	0.73	0.77	0.75	0.66	0.72	0.66	0.59	0.67	0.73	0.73	0.66	0.72	0.67	1.00				
21	0.63	0.66	0.70	0.72	0.67	0.72	0.74	0.73	0.74	0.66	0.69	0.64	0.62	0.68	0.74	0.66	0.64	0.69	0.64	0.75	1.00			
22	0.63	0.68	0.72	0.69	0.69	0.77	0.76	0.74	0.73	0.68	0.73	0.66	0.59	0.67	0.71	0.72	0.67	0.74	0.68	0.75	0.75	1.00		
23	0.63	0.59	0.65	0.68	0.66	0.69	0.68	0.67	0.66	0.61	0.63	0.66	0.60	0.60	0.67	0.65	0.63	0.63	0.59	0.68	0.73	0.71	1.00	
24	0.62	0.62	0.67	0.62	0.61	0.69	0.67	0.64	0.63	0.65	0.60	0.67	0.62	0.63	0.68	0.62	0.65	0.64	0.63	0.65	0.68	0.68	0.65	1.00

Note: Serial number (1 to 24) of the local maize varieties are same as given in Table 1.

high degree of genetic similarity (0.85) among the maize varieties indicated their possible relatedness though they belong to different geographical locations. It might be due to migration by local farmers' or cultivation of varieties in different areas. The dendrogram (Fig. 3) using UPGMA based on the Jaccard similarity co-efficient divided 24

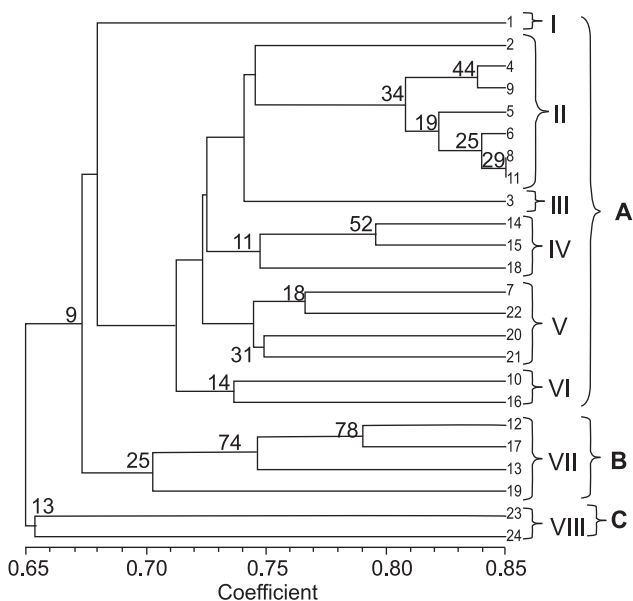


Fig. 3. UPGMA cluster analysis of 24 maize varieties on the basis of RAPD profiles. The values mentioned in the nodes of the diagram are the Bootstrap values. (1-24: Names of varieties as mentioned in Table 1)

varieties into 3 major clusters (A,B,C) and these major clusters were divided into 8 subgroups. The major cluster A was further divided into 6 subgroups. The subgroup I consists of only one variety Dharap Local-4. The uniqueness of variety was also observed by P-3 and P-10 random primers that they generated unique MIP for this variety. The subgroup II consists of 7 varieties in which Khusalpur Local-4 and Nunawala Local-5 showed most similar varieties (Bootstrap value 29) although they are from different geographical locations. Paschim Khera Local-2 and Khusalpur Local-6 also showed closeness to each other but Kirtinagar Local-4 is non-clustered in subgroup II and also well defined by PCA value (Fig 1). Purvi Khera Local-1 clustered individually in subgroup 3 and showed unique MIPs from P-1, P-3 and P-10 primers also. Bailparow Local-4 and Bhadarabad Local-4, though they are from different geographical locations but clustered together (52%) along with Bagauli Local-1 in subgroup IV. Khusalpur Local-3, Khera Local, Chamoli Local and Quarali Local-1 were clustered together in subgroup V. Nunawala Local-1 and Bhadarabad Local-5 were clustered together in subgroup VI. The major cluster B consists of only one subgroup and having Nunawala Local-6, Bhadarabad Local-8 with similarity 78% and Doiwala Local-2 and Bishanpur Local-2. The reference varieties Pragati and HQPM1 were clustered together

but their uniqueness was shown in three dimensional graph of PCA (Fig 1).

Thus, out of a total of 24 varieties unique identification profiles were obtained for 14 varieties while with DUS morphological descriptors only 11 varieties could be discriminated. However, when both morphological and RAPD molecular markers were taken together then 17 varieties could be discriminated. By this study, it can be concluded that in situations where the morpho-physiological DUS descriptors are not able to establish distinctiveness of a variety then molecular markers may be used as additional or complementary descriptors for resolving distinctiveness of maize varieties for granting plant variety protection under PPV&FR Act.

Results of this study provided sufficient evidence that molecular markers would increase the standards of DUS testing if these are included as additional descriptors and the same had been observed in earlier studies on rice (Patra *et al.*, 2010; Joshi *et al.*, 2010) sorghum (Joshi *et al.*, 2011) and maize inbred lines (Gunjaca *et al.*, 2008). Higher discrimination power of molecular marker is generated by more balanced distribution of allele frequencies. These results which have been tested and confirmed, shows that molecular markers are ideal additional descriptors for establishing distinctiveness of closely related maize varieties, that serve the purpose of protection and registration of plant variety. The molecular profiling and unique molecular fingerprinting carried out in the present study could be utilized for germplasm conservation, authentication, purification and identification of varieties for registration to protect plant varieties and farmers' rights.

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Development of Upland Rice Varieties for Drought Tolerance through Drought Susceptibility Index and Stability Parameters

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Breeding for upland rice having high yield potential coupled with drought tolerance should be the strategy for a successful breeding programme. Twenty seven genotypes were evaluated for drought susceptibility index (DSI) and stability under three water regimes. Genotype, environment and their interactions influenced significantly the phenotypes for all characteristics of genotypes. CBT 3-06 and Anjali were identified as stable genotypes by Eberhart & Russell model. The use of DSI is likely to be most beneficial in selecting parents for development of drought resistant populations, particularly when yield potential vary greatly among the tested genotypes. Lalsar, RR 348-6, CR 143-2-2, Kalinga III, Brown Gora and CB 0-13-1 were recorded lowest DSI ($DSI < 1$) for seed yield over the conditions. This contrast CR 143-2-2 Brown Gora and CB 0-13-1 recorded good score for EVV, tip drying, leaf rolling and drought score and suggested for inclusion into hybridization to obtain improved recombinants for water stress environments. Based on the various parameters, the genotypes Kalinga III CB 0-13-1 may be exploited for commercial cultivation under different water regimes after some adjustment to stabilize the yield.

Key Words: Drought Tolerance, DSI and Stability, Selection Parameters, Upland rice

Introduction

Rice in Asia is cultivated by many farm families and closely linked with the social harmony, prosperity, national food security and political stability of many countries (Hossain and Fischer, 1995). Demand for rice is expected to grow faster than the production in most countries (Swaminathan, 1998). Nearly 100 million people now depend on upland rice as their daily staple food. Upland rice is usually grown in systems where little or no fertilizer is applied, and is direct-seeded into unpuddled, unsaturated soil (Atlin *et al.*, 2004). Most traditional upland rice varieties are low-yielding and prone to lodging, but are adapted to non-flooded soils (Atlin *et al.*, 2006). Upland rice encompasses 12 per cent of global rice production area and is generally the lowest yielding ecosystem (Khush, 1997). Climate related natural disasters are the principal sources of risk and uncertainty in rice farming. Drought is one of the major constraints to low and unstable rice production in Asia. At least 23 million ha area of rice in Asia is drought prone and India accounts for the largest share (59%) of total drought-prone rice area in Asia (Pandey *et al.*, 2007).

Irrigation is not a viable option to alleviate drought problems in rainfed rice growing ecosystem. Drought mitigation through improved drought resistant rice

varieties and complementary management practices, represent an important exit pathway from poverty (Serraj and Atlin, 2008). Recently, improved upland rice varieties with higher harvest index, improved input responsiveness and consequently higher yield potential have been developed at IRRI, in Brazil and several Asian countries. Such “aerobic rice” varieties, combine aerobic adaptations of traditional upland varieties with input-responsiveness, lodging tolerance and yield potential of irrigated varieties (Atlin *et al.*, 2006). Thus, the present study was taken up to identify drought tolerant cultivars combined with high yield potential for drought prone upland.

Materials and Methods

Field screening experiments for large number of entries (216 from Observational Yield Trial under IIDBN programme) were conducted to identify drought tolerant genotypes at Central Rice Research Institute, Cuttack. On the basis of performance, 27 promising genotypes selected from different centers including CRRI, Cuttack and International Rice Research Institute, Philippines, were grown under field condition along with three check varieties *viz.*, Anjali, Kalinga III and Vandana selected on the basis of performance under drought prone upland condition.

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The experiment was laid out in Alpha Lattice Design with three replications under three water regimes: (a) irrigated control (E1) (b) moderate stress (E2) and (c) severe stress (E3) conditions at vegetative stage during dry season in the year 2008. These three water regimes were always apart from each other to avoid water interference. Plants were grown under adequate soil moisture for 30 days after germination under both the conditions. The irrigation was withdrawn for 30 days and beyond, till the susceptible check shows permanent wilting in the severe stress field, subsequently the plot was re-watered for recovery. Soil moisture content (SMC) during stress period was monitored through periodical soil sampling at 0-15, 15-30 cm soil depth after suspension water. The experimental field under irrigated condition was designed to maintain assured soil moisture by keeping 5 cm standing water. Peizometers were installed in all the treatments to monitor the ground water table. Each plot was 5 m long and 3 m wide, row to row distance was 20 cm and plant to plant distance was 15 cm each plot. Rice varieties under all the conditions were dry direct seeded at 2-3 cm soil depth by hand plough with the seed rate of 60 kg ha⁻¹ to maintain 3-4 seeds hill⁻¹. This method gave uniform seedling emergence for all the plots in 6-8 days. Recommended package of agronomic practices were followed.

The drought scores and recovery observations were taken as per SES method, on a scale of 1 to 9 (IRRI, 1996). Plant samples above the ground were collected at maturity. Observations were recorded on seed yield (t ha⁻¹), days to 50 per cent flowering, days to maturity and plant height. The effect of stress was assessed as percentage reduction in mean performance of a characteristic under stress condition relative to the performance of the same trait under continuously saturated soil moisture condition. Drought Susceptibility Index (DSI) for each trait was calculated on the basis of mean data of severe stress and irrigated condition experiments, following Fischer and Maurer (1978). The data were analyzed by appropriate statistical analysis (Gomez and Gomez, 1984) using CropStat 7.2 programme (IRRI, 2009). Pooled analysis of variance over three environments was estimated as per the model suggested by Eberhart and Russell (1966) and followed to estimate the three stability parameters viz., mean, regression coefficient (bi) and mean squared deviation (S²di) for each genotype.

Results and Discussion

Drought Susceptibility Index (DSI)

The managed water stress protocol resulted in considerable reduction in yield compared to yield under irrigated conditions and trials achieved mean reduction of 30-50% under moderate stress and 65-80% yield reduction compared to control under severe stress. Experiments were conducted during dry season, rain fall was nil during stress period, so experiment expose to desired level of stress. 66% genotypes (including checks) recorded high yield potential (>4.5) but dramatically reduced the yield under both water stress condition. Comparison across the stress conditions indicated that the genotypes Lalsar and CR 143-2-2 emerged as tolerant genotypes for grain yield under both the conditions. Results indicted the severity of stress.

The use of the DSI can help to distinguish suitable variety for drought stress from phenology and yield potential. Large DSI values indicate greater drought susceptibility (Chauhan *et al.*, 2007). Low DSI mean values (DSI<1) observed for seed yield indicated that this character is relatively resistant to stress. The Lalsar, RR 348-6, CR 143-2-2, Kalinga III, Brown Gora and CB 0-13-1 recorded lowest DSI (DSI≤1) for seed yield over the conditions, thereby indicating that the genotypes were tolerant to vegetative drought stress conditions. Further, more tolerant genotypes especially CR 143-2-2, Brown Gora and CB 0-13-1 also showed good score for early vegetative vigour in all the conditions, leaf rolling and tip drying in both stress condition and drought score in severe stress condition (Table 1). Early vigour in upland rice is associated with improved ability to tolerate weed competition, which is a major constraint in direct seeded rice crops (Namuco *et al.*, 2009). Weed competitiveness was negatively but weakly correlated with yield potential, and positively, with crop duration (Dingkuhn *et al.*, 1999). Specific leaf area and tillering ability, which are major determinants of vegetative vigour, and crop duration, which affects the ability to recover from early competition, are useful traits in the selection of weed competitive rice, particularly in breeding programme. Much larger gains should be expected from use of genotypes with below average DSI in future breeding for drought tolerant rice.

Table 1. Estimates of Drought Susceptible Index (DSI) for seed yield ($t\ ha^{-1}$) and Early Vegetative Vigour (EVV), Drought score, Leaf Rolling (LR) and Tip Drying (TD) of genotypes under different environments

S. No		Genotypes		Seed Yield		E1		E2		E3	
				(t ha-1)		LR		D		LR	
		DSI (E2)	DSI (E3)	EVV	EVV	LR	TD	EVV	D Score	LR	TD
1	ASD 17	1.00	1.03	2	1	3	2	1	3	2	1
2	Ashoka 228	1.09	1.04	2	1	3	3	1	6	4	3
3	Birsa Gora	1.02	0.89	2	2	3	2	2	2	1	1
4	Brown Gora	0.93	1.00	2	2	1	2	2	3	1	1
5	CB 0-13-1	0.94	1.00	1	1	2	2	1	3	1	1
6	CBT 3-05	1.02	1.00	1	2	3	3	2	3	2	2
7	CBT 3-06	0.91	1.01	1	3	3	2	2	3	2	2
8	CR 143-2-2	0.68	0.96	1	3	1	1	3	2	1	1
9	IR 76569-259-1-1-3	1.09	1.01	2	2	3	2	2	3	2	1
10	Kakro	1.02	1.00	1	2	3	2	1	3	2	1
11	Lalsar	0.30	0.85	1	3	2	2	2	3	2	1
12	N 22	1.03	1.04	1	2	3	3	1	3	2	1
13	RR 222-1	1.10	1.03	2	3	4	3	1	6	4	3
14	RR 267-9	1.05	0.98	1	3	3	2	2	4	2	2
15	RR 345-2	1.12	1.00	2	3	3	2	2	4	3	2
16	RR 348-6	0.62	1.00	1	3	2	1	2	4	3	2
17	RR363-3	1.03	0.94	2	2	3	2	1	3	1	1
18	RR366-5	1.04	0.97	2	2	4	3	2	3	2	2
19	RR 372-2	1.06	1.03	2	1	4	2	2	4	3	2
20	RR 383-2	1.04	1.00	2	2	2	1	2	2	1	1
21	RR433-2	1.08	1.05	2	2	3	2	2	6	4	3
22	RR 440-167-2-13	1.05	0.90	2	2	4	2	2	3	2	1
23	Sathi 34-36	1.06	1.01	1	2	4	2	1	4	2	2
24	Thara	0.89	1.01	2	1	3	3	3	3	2	1
25	Anjali (Check)	1.05	1.03	1	3	3	2	2	6	4	2
26	Kalinga III (Check)	0.91	0.97	2	2	4	3	2	3	2	2

E1: Control, E2: Vegetative Stage Moderate Stress and E3: Vegetative Stage Severe Stress

Analysis of Variance for Stability Analysis

Highly significant variances due to genotype for all the traits indicated the presence of genetic variance in the plant material. Mean squares due to environment were found significant for all the characters, indicating

differences between environments and their influence on genotypes for expression of these characters. This is in accordance with previous reports on rice by Honarnejad *et al.* (2000) and Sedghi-Azar *et al.* (2008). Mean square due to G x E interaction was significant only for seed yield ($t\ ha^{-1}$). The G x E interaction mean squares were further partitioned into two components *viz.*, G x E (linear) and pooled deviation (non linear) for all the traits. Pooled deviation showed significant differences for all the traits except seed yield ($t\ ha^{-1}$) while, G x E (linear) significant for seed yield ($t\ ha^{-1}$). Linear and non-linear components of G x E interaction were significant for all the characters, confirming the findings of Panwar *et al.* (2008) and Nayak *et al.* (2003). Simultaneously, “F” value of all the characters under study were found significant for Environment + (G x E) and Environments (Lin.).

Stability Parameters

Mean value (μ), regression coefficient (b_i) and deviation from regression (S^2_{di}) are presented in Table 2. Twelve genotypes recorded higher yield over grand mean of $1.99\ t\ ha^{-1}$. Only six genotypes i.e. RR 372-2, Kakro, RR 383-2, CBT 3-06, IR 76569-259-1-1-3 and Sathi 34-36 showed b_i values close to unity. The significant values of S^2_{di} were observed in two genotypes. The remaining genotypes recorded non significant deviation from regression. Deviation from regression for plant height was found to be non significant in six genotypes. Of 27 genotypes, 16 genotypes registered higher mean over grand mean for plant height while, only five genotypes recorded b_i values near unity.

The genotype ASD 17 ($2.79\ t\ ha^{-1}$), Kalinga III ($2.74\ t\ ha^{-1}$), CB 0-13-1 ($2.67\ t\ ha^{-1}$), Ashoka 228 ($2.56\ t\ ha^{-1}$) are having highest mean yield along with RR 267-9 ($2.43\ t\ ha^{-1}$), N 22 ($2.36\ t\ ha^{-1}$), CBT 3-05 ($2.30\ t\ ha^{-1}$) and RR 222-1 ($2.29\ t\ ha^{-1}$) but having b_i value more than one, indicating that these genotypes are having below average stability of performance and are specifically adapted to favorable environments, however, these are sensitive to environmental changes. Similar results have also been reported by Vidhu *et al.* (2005) and Panwar *et al.* (2008). While, only Thara yielded more than $2\ t\ ha^{-1}$ but having b_i value less than 1 do not respond favorable condition hence, it could be regarded as specially adapted to unfavorable environments. It was observed from the present study that majority of the high yielding genotypes have either above average ($b_i > 1$) or below average ($b_i < 1$)

Table 2. Genotypic means and stability parameters for different characters in rice

SN	Genotypes	Days to 50 per cent flowering			Days to Maturity			Plant Height (cm)			Seed yield (t ha ⁻¹)		
		Mean	Bi	S ² d	Mean	Bi	S ² d	Mean	bi	S ² d	Mean	bi	S ² d
1	ASD 17	72.78	1.38	4.23	103.22	1.62	11.55**	88.17	1.77	-5.54	2.79	1.48	-0.08
2	Ashoka 228	72.78	1.66	11.92*	103.22	1.91	21.91**	82.65	1.66	47.37	2.56	1.49	-0.04
3	Birsa Gora	70.56	0.68	-2.29	100.67	0.80	-1.46	78.93	0.94	-21.77	1.72	0.76	0.01
4	Brown Gora	69.67	0.55	1.04	99.22	0.26	-1.02	83.99	0.95	-18.86	1.46	0.69	-0.07
5	CB 0-13-1	72.22	0.23	18.16**	102.22	0.28	19.08**	80.92	1.70	42.73	2.67	1.27	-0.05
6	CBT 3-05	70.44	0.93	-0.93	100.78	1.11	-0.36	72.36	0.83	86.10*	2.30	1.19	-0.08
7	CBT 3-06	72.89	1.54	6.11	103.56	1.89	17.65**	85.91	1.46	70.07	2.17	1.00	0.01
8	CR 143-2-2	68.00	0.16	5.18	98.67	0.45	2.59	64.56	0.89	-24.51	1.64	0.58	0.20
9	IR 76569-259-1-1-3	73.44	1.91	-2.19	104.33	2.53	-0.45	90.18	0.62	-21.29	1.89	1.03	-0.03
10	Kakro	70.67	1.54	-2.38	100.33	1.13	-1.46	84.38	1.35	24.14	1.96	1.01	-0.08
11	Lalsar	49.89	0.59	-2.59	80.44	0.27	-1.48	63.74	0.15	131.06*	0.94	0.16	0.32*
12	N 22	67.00	1.76	3.67	96.89	1.79	4.90*	88.95	1.29	-14.24	2.36	1.31	-0.09
13	RR 222-1	70.78	0.84	4.61	101.22	0.74	17.81**	79.41	1.02	-0.02	2.29	1.33	-0.02
14	RR 267-9	71.11	1.10	-2.60	100.56	0.72	-1.19	74.19	1.07	-15.92	2.43	1.26	-0.01
15	RR 345-2	69.11	0.73	5.37	99.22	0.72	5.78*	88.76	1.16	-23.18	2.09	1.18	0.08
16	RR 348-6	64.67	0.87	-2.64	95.11	0.94	-1.48	57.18	0.43	-1.22	1.77	0.63	0.50*
17	RR363-3	70.56	0.07	0.43	100.45	0.04	2.34	89.75	0.81	-21.88	1.53	0.73	-0/05
18	RR366-5	70.44	0.98	-2.51	100.56	1.08	-1.47	86.06	0.38	32.25	1.57	0.79	-0.06
19	RR 372-2	54.78	2.12	34.14**	86.00	2.28	43.80**	85.16	0.57	216.74**	1.77	0.98	-0.08
20	RR 383-2	70.67	0.58	-2.20	99.89	0.09	1.59	97.84	1.43	58.61	1.95	1.02	-0.07
21	RR433-2	73.44	1.04	-1.27	104.33	1.32	-1.23	77.19	1.07	51.91	1.96	1.14	-0.07
22	RR 440-167-2-13	67.89	0.14	3.79	98.78	0.38	2.38	83.15	0.29	121.75*	1.45	0.67	0.01
23	Sathi 34-36	76.78	1.61	5.74	107.22	1.62	11.55**	76.26	0.71	99.55*	1.99	1.03	-0.05
24	Thara	66.89	0.61	0.81	96.22	0.22	-1.17	89.81	1.10	18.54	2.05	0.95	0.02
25	Anjali	69.33	0.73	1.81	99.44	0.60	3.04	80.74	1.35	-14.83	2.12	1.00	0.02
26	Kalinga III	65.89	2.16	-1.69	97.22	1.1	1.69	88.83	1.15	92.84*	2.74	1.20	-0.05
27	Vandana	70.33	0.50	3.05	100.45	1.08	0.84	87.48	0.84	18.19	1.66	0.85	-0.09
	Mean	69.00			99.27			81.72			1.99		

responses. Mishra and Mahapatra (1998) suggested to evaluate these genotypes in irrigated as well as drought stress condition and to carry out the regression analysis separately to identify genotypes combining high yield potential with wider array of adaptation to variable environments.

Among the 27 genotypes, only two genotype CBT 3-06 (2.17 t ha⁻¹) and Anjali (2.12 t ha⁻¹) registered bi=1 and showed non significant deviation from regression near to zero. Therefore, these genotypes were stable for yield in all the environments. Genotype RR 372 (1.77 t ha⁻¹), Kakro (1.96 t ha⁻¹) and RR 383-2 (1.95 t ha⁻¹) along with IR 76569-259-1-1-3 (1.89 t ha⁻¹) and Sathi 34-36 (1.99 t ha⁻¹) are having relatively higher mean yield, regression coefficient near unity and about no

deviation from regression. Indicating that these genotypes are having average stability and above average yields in most of the environments. Moreover, these genotypes, particularly RR 372, Kakro and RR 383-2 possess b value slightly less than unity for yield, indicating that these should also respond to the more favorable environments (Bhakta and Das 2008). In other words these genotypes are grouped as stable genotypes with general adaptability. These genotypes are not following any consistent stability (values of bi & S²di) trend for other traits, *i.e.* days to maturity and plant height, under studied. Tall stature during vegetative stage is prerequisite for upland rice to compete the weed. It seems that for stabilizing the yield these genotypes are making some morpho-physiological adjustments leading to below average or above average

stability of performance of the genotypes for yield contributing traits other than yield. Moreover, yield is the most preferred and reliable criterion for selection of genotypes under stress condition than secondary trait selections (Serraj and Atlin, 2008), thus can be the sole criterion for identification of suitable genotypes.

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Characterization of Landraces of Rice from Eastern India

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For the establishment of the distinctness, a set of 51 landraces of rice were characterized for 46 characters following standard Distinctiveness, Uniformity and Stability (DUS) testing guidelines for three seasons. Out of 51 varieties studied, 27 were found to be distinctive on the basis of 22 essential and 24 additional characters. The present study will be useful for breeders, researchers and farmers to identify valuable germplasm and their conservation as well as utilization in rice improvement as well as also to seek protection under Protection of Plant Varieties and Farmers' Rights Act.

Key Words: Characterization, DUS test, PPV&FR Act, Rice landraces, Varieties

Introduction

To meet the continuously expanding needs of varietal improvement, the assemblage, evaluation, preservation and characterization of the entire existing germplasm are essential to more rewarding breeding efforts. Until a collection has been properly evaluated it has little practical use (Chang, 1976). India has a rich and diverse genetic wealth of rice and it has been estimated from various surveys that nearly 50,000 types of rice are still being grown in the country (Roy *et al.* 1985). The switch over to high yielding varieties with the spread of modern agriculture, has posed a great threat to the security of the age old practice of growing traditional varieties and landraces which may have immense potential for different important traits (Richharia, 1979; Sharma *et al.*, 1987; Patra, 2000). In order to prevent any further gene erosion, collection and conservation of such invaluable genetic resources of rice is essential. Characterization of variety is useful to identify and avoid duplication. Characterization should eventually lead to a system of recording and storing useful data that can be readily retrieved and made available to others and help in planning breeding programmes (Dabas *et al.*, 1994). Qualitative traits being more stable over generations (Raut, 2003) are reliable for characterization of varieties.

Being signatory to the General Agreement on Trade and Tariffs, Government of India has enacted its *sui-generis* system, Protection of Plant Varieties and Farmers' Right Act (PPV&FRA), 2001 for providing protection to plant varieties based on distinctiveness, uniformity and stability (DUS) test, apart from novelty (Anonymous,

2001). Therefore, the characterization of a variety is a prerequisite and identification of plant varieties of common knowledge is essential for the protection of new plant varieties. Article 15.3(b) of the PPV&FRA states that the new variety must be clearly distinguishable by one or more essential characters from any variety whose existence is a matter of common knowledge at the time of seeking protection. The uniqueness of a variety is to be established by following standard DUS testing guidelines.

Keeping in view these facts, the present investigation was planned to characterize a set of *aman* rice genotypes of West Bengal, to understand *in situ* variability of different agro-morphological traits and interrelationship among them.

Materials and Methods

Fifty one landraces of rice were grown in a Randomized Complete Block Design with two replications at the research farm of Zonal Adaptive Research Station, (23°24'N latitude and 88°31'E longitude with an altitude of 9.75 meters msl) Krishnagar, Nadia, West Bengal, India during *kharif* season for three consecutive years of 2008, 2009 and 2010. The soil is slightly acidic with pH of 6.0, low soluble salts (EC of 0.15 dS m⁻¹), medium organic carbon content (0.57%), Total N (0.056%), medium in available P (25.28 kg ha⁻¹) and K (148.77 kg ha⁻¹). The experimental site belongs to tropical humid climate having the average rainfall of 1464 mm, most of the amount falls between June to September. The minimum temperature reaches 7.6°C in the month of January and the maximum 41.1°C in the month of May. It has been

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observed that 74.7% of the annual rainfall is obtained during June to September and more than 83.6% during June to October. The materials were transplanted in $3.0 \times 2.85 \text{ m}^2$ plot with plant to plant spacing 15 cm within a row and row to row spacing of 20 cm, plot to plot distance was 60 cm. A random sample of five competitive plants was used for observations on different traits under study. Crop was raised following recommended package of practices. Fertilizers (N:P₂O₅:K₂O) @ 50:25:25 kg ha⁻¹ were applied.

Among the qualitative trait, 46 visually assessed characteristics were observed according to the National Test Guidelines for DUS test in rice developed by Directorate of Rice Research, Hyderabad in consultation with the national core group experts for development of national guidelines in crop plants and also with the rice experts. The observation of various characteristics was recorded at different stages of growth with appropriate procedures as per the DUS test guidelines of PPV&FRA, (Shobha Rani *et al.*, 2004).

Results and Discussion

Qualitative characters are considered as marker characters in the identification of landraces of rice, which are less influenced by environmental fluctuations. The work on inheritance and linkage studies of qualitative characters was reviewed by Raut (2003). The published work (Satyavathi and Subramanyam, 2004) also substantiated that the flower petal colour, presence and absence of pod hair, colour of hair, seed colour were the most stable characters across the agroclimatic zones. To establish distinctiveness among rice cultivars, data on 46 characters have been recorded (Table 1).

For leaf characteristics, intensity of green colour was dark in 17 cultivars. Out of 51 cultivars, 25 cultivars had leaf anthocyanin colouration. Among 25, variety G28 is distinct for having uniform distribution of anthocyanin colouration. Anthocyanin colouration in leaf sheath was present in 18 cultivars, out of which, 3 cultivars (G6, G36 and G41) had medium, 5 cultivars (G7, G2, G20, G49 and G29) had weak intensity of anthocyanin colouration. Three cultivars (G22, G28 and G48) were found to be distinct for having strong pubescence while 7 cultivars were marked for absence of pubescence in leaf blade.

Seven cultivars were distinguished for absence of leaf auricle (G7, G8, G16, G18, G23, G25 and G45).

Out of rest 44 varieties, 6 varieties (G17, G21, G24, G39, G41 and G510) were marked for having purpled coloured auricle. Leaf collar was absent in two cultivars *viz.*, G20 and G45. Out of 49 cultivars, 6 cultivars (G5, G21, G30, G41, G46 and G51) were distinguished for having anthocyanin colouration both in leaf auricle and leaf collar (Table 1).

Regarding leaf ligule shape, variety G49 was distinct for having truncate ligule shape. Six varieties (G12, G14, G15, G24, G42 and G45) were distinguished for having acute ligule shape. Rest of the varieties had split shaped ligule. Two cultivars *viz.* G20 and G45 were marked for absence of leaf ligule among 51 cultivars. Six cultivars (G6, G7, G21, G23, G37 and G51) were distinguished for having purple coloured ligule (Table 1).

Cultivars *viz.*, G5, G11, G12, G16, G27 G28, G42 and G45 were found to possess long leaf length and broad leaf breadth along with erect early flag leaf attitude. Since, erect flag leaf angle was associated with high yield in rice (Chang and Tagumpay, 1970), hence, cultivars with erect flag leaf are more distinguished here. Increasing light penetration into crop canopy has been suggested as one of the ways of obtaining higher grain yields. Duncan (1971) showed that increased penetration of light into canopy would increase photosynthetic rate and perhaps enhance grain yield.

There was no deflexed flag leaf at early observations but 5 cultivars (G15, G18, G22, G31 and G49) showed deflexed flag leaf attitude during the late observations (Table 1).

In case of time of heading, 7 cultivars (G7, G16, G18, G25, G27, G41 and G45) were distinguished for early category while 7 cultivars *viz.*, G28, G29, G30, G32, G36, G44 and G48 showed very late time of heading. It was very interesting to find out that varieties G30 and G44 with spreading culm attitude showed very late time of heading while cultivars *viz.*, G7, G16, G27 and G41 having early time of heading showed erect culm attitude (Table 1).

Regarding anthocyanin colouration in lemma of spikelet, 3 cultivars *viz.*, G22, G47 and G50 had strong colouration in keel, two cultivars *viz.*, G17 and G50 had strong colouration at the area below apex and three cultivars *viz.*, G5, G23 and G50 had strong anthocyanin colouration at the apex. Cultivars G38 and G51 were distinguished for having purple coloured stigma (Table 1).

Table 1. Characterization of the cultivars (total 51) as per DUS guidelines

Sl. No.	Cultivars	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46
G1	Ranisal	C	G	M	P	IBO	A	-	W	P	C	P	A	P	S	LP	M	M	SE	Ho	SE	L	VW	A	A	w	M	M	A	-	A	M	Ss	M	A	w	Sw	A	-	-	-	P	W	E-S	PE	L	I
G2	Badhabna	G	G	M	P	IBO	P	VW	M	P	C	P	A	P	S	LP	M	M	SE	Ho	SE	L	W	W	A	LG	M	M	A	-	A	M	Ss	M	A	w	Sw	A	-	-	-	P	S	SE	ME	M	E
G3	Machkata	C	G	D	P	IBO	A	-	W	P	C	P	A	P	S	LP	M	M	Ho	Ho	SE	L	W	W	A	LG	M	Sh	A	-	A	M	Ss	M	M	b	GG	A	-	-	-	P	S	E-S	ME	L	L
G4	Laldhula	C	G	M	P	IBO	A	-	M	P	C	P	A	P	S	LP	M	M	SE	Ho	E	M	VW	A	A	w	Tn	M	A	-	A	Lg	Ss	M	M	B	GG	A	-	-	-	P	S	E-S	PE	M	I
G5	Dhula-dhan	P	G	D	A	-	A	-	A	P	C	P	P	P	S	LP	L	B	E	SE	E	L	VW	W	S	LG	M	M	A	-	P	Lg	s	F	S	B	b	A	-	-	-	P	S	E-S	PE	L	L
G6	Dhuri	C	G	M	P	IBO	P	M	M	P	C	P	A	P	S	P	M	M	SE	Ho	O	M	VW	A	A	LG	M	M	A	-	P	M	s	m	W	w	Sw	A	-	-	P	W	E-S	PE	M	E	
G7	Kalam-kathi	P	G	M	P	IBO	P	W	W	A	-	P	A	P	S	P	S	N	E	SE	E	E	VW	A	A	LG	M	Sh	A	-	A	VSh	s	M	W	Y	Sw	A	-	-	-	P	W	E	ME	E	E
G8	Suakalma	C	G	M	A	-	A	-	M	A	-	P	A	P	S	LP	M	M	SE	Ho	E	L	W	M	A	LG	Tn	Sh	A	-	P	M	D	m	W	Y	By	A	-	-	-	P	W	E-S	ME	L	E
G9	Nakrasal	C	G	D	A	-	A	-	W	P	C	P	A	P	S	LP	M	M	SE	Ho	E	L	VW	A	A	LG	Tn	M	A	-	A	Lg	D	M	W	Y	Bf	A	-	-	-	P	W	E-S	ME	L	E
G10	Asanlaya (White)	C	G	M	A	-	P	VW	W	P	C	P	A	P	S	LP	M	M	E	SE	E	L	VW	A	A	LG	Tn	Lg	A	-	A	Lg	s	F	W	w	Sw	A	-	-	-	P	W	E-S	ME	L	E
G11	Asanlaya (Red)	C	G	M	P	IBO	A	-	M	P	C	P	A	P	S	LP	L	B	E	SE	E	L	VW	A	A	LG	Tn	Lg	A	-	A	Lg	s	F	A	w	Sw	A	-	-	-	P	S	E-S	PE	L	E
G12	Pubalgara	C	G	M	A	-	P	W	A	P	LP	P	A	P	Ac	LP	L	B	E	SE	E	L	VW	A	A	w	Tk	Lg	A	-	A	Sh	Ss	M	S	Y	Bs	P	Yw	L	UHO	P	W	E-S	ME	L	L
G13	Daharn-agra	G	LP	M	A	-	A	-	A	P	LP	P	A	P	S	LP	M	M	SE	Ho	E	L	VW	A	A	w	Tn	A	-	A	M	Ss	F	S	Y	Bf	A	-	-	-	P	S	E-S	ME	L	I	
G14	Kalonunia	C	G	D	P	OMO	A	-	W	P	C	P	A	P	Ac	LP	M	M	SE	Ho	SE	L	VW	A	A	w	Tn	VSh	A	-	A	M	Ss	M	W	Y	GG	A	-	-	-	P	S	E-S	ME	L	I
G15	Tulisbhog	C	G	L	A	-	A	-	W	p	LP	P	A	P	Ac	LP	S	N	SE	DF	E	L	VW	A	A	w	M	M	A	-	A	VLg	s	M	W	Y	GG	P	b	M	TO	P	W	E-S	WE	L	I
G16	Mahishi-adan	C	G	M	A	-	A	-	M	A	-	P	A	P	S	LP	L	B	E	SE	E	E	W	A	A	LG	M	Lg	A	-	A	M	s	m	M	B	Bf	P	b	VSh	WL	P	W	E-S	ME	E	L
G17	Dudhik-alma	C	G	M	A	-	A	-	M	P	P	P	A	P	S	LP	M	M	SE	Ho	SE	L	M	S	A	LG	Tk	Vl	A	-	A	M	s	M	W	b	Pf	A	-	-	-	P	S	E-S	PE	L	L
G18	Sankarsal	G	LP	L	A	-	A	-	W	A	-	P	A	P	S	LP	S	N	Ho	DF	Sp	E	VW	A	A	w	Tn	Sh	A	-	A	Sh	D	F	W	w	Sw	A	-	-	-	P	W	E	ME	E	E
G19	Badsa-bhog	C	G	D	A	-	A	-	W	P	C	P	A	P	S	LP	M	M	Ho	Ho	O	L	VW	A	A	w	M	Lg	A	-	A	M	Ss	M	M	w	Bt	A	-	-	-	P	W	E-S	PE	L	I
G20	Agnisal	C	G	M	P	OMO	P	W	M	P	C	P	A	A	S	G	M	M	SE	SE	SE	L	VW	A	W	LG	M	M	A	-	A	Lg	Ss	F	S	B	Bf	A	-	-	-	P	S	E-S	WE	L	I
G21	Chandra-kanta	C	G	M	A	-	A	-	W	P	P	P	P	P	S	P	M	M	SE	Ho	Sp	M	VW	A	A	LG	M	Lg	A	-	A	M	Ss	F	M	B	Bf	P	Yw	L	TO	P	S	E-S	ME	M	I
G22	Muktasal	G	G	D	P	OMO	P	VW	S	P	LP	P	A	P	S	LP	L	B	SE	DF	E	M	S	W	A	LG	Tk	Vlg	A	W	A	M	s	F	M	Y	Sw	P	Yb	L	TO	P	W	E-S	ME	M	L
G23	Punjabul	P	PL	M	A	-	A	-	W	A	-	P	A	P	S	P	M	M	SE	Ho	Sp	M	VW	W	S	LG	M	VSh	A	-	A	Sh	s	M	M	P	Bt	P	Yw	M	WL	P	W	E-S	ME	M	L
G24	Sitasal	C	G	D	P	IBO	A	-	W	P	P	P	A	P	Ac	LP	L	B	SE	Ho	E	L	VW	A	A	LG	Tk	M	A	-	A	Lg	s	M	W	Y	Sw	A	-	-	-	P	S	E	PE	L	L
G25	Behalsal	C	G	D	A	-	A	-	M	A	-	P	A	P	S	LP	M	M	E	SE	SE	E	VW	A	A	LG	Tn	M	A	-	A	Sh	Ss	F	W	w	GG	P	Yw	Vlg	TO	P	W	E	PE	E	E
G26	Kabirajsal	C	G	M	A	-	A	-	A	P	C	P	A	P	S	LP	S	N	SE	Ho	E	L	VW	A	A	w	M	M	A	-	A	Lg	Ss	M	W	w	GG	A	-	-	-	P	W	E-S	ME	L	I
G27	Laldhusri	C	G	D	P	OMO	A	-	M	P	C	P	A	P	S	LP	L	B	E	SE	E	E	W	A	A	LG	M	M	A	-	A	Vlg	s	F	W	w	GG	A	-	-	-	P	W	E	PE	E	I
G28	Malliksal	G	G	D	P	U	A	-	S	P	C	P	A	P	S	LP	L	B	E	SE	E	VL	VW	A	A	w	Tk	Lg	A	-	A	Lg	s	M	W	w	GG	P	Yw	Vlg	TO	P	S	E-S	ME	VL	I
G29	Baidjhulur	G	G	D	P	OMO	P	VW	M	P	LP	P	A	P	S	LP	L	B	SE	Ho	E	VL	VW	A	A	w	Tn	Sh	A	-	A	Lg	Ss	M	W	Y	Sw	A	-	-	-	P	W	E-S	ME	VL	I

G30	Jhulur	G	G	M	P	OMO	A	-	M	P	C	P	P	S	G	M	M	E	Ho	Sp	VL	VW	A	A	LG	Tk	VLg	A	-	A	Lg	Ss	M	M	Y	Bf	A	-	-	-	P	W	E	PE	VLL	
G31	Manikanchan	C	G	L	A	-	A	-	W	P	C	P	A	P	S	G	S	N	Ho	DF	O	M	VW	A	A	w	Tk	VLg	A	-	A	Sh	D	M	W	w	Sw	A	-	-	-	P	W	S-S	ME	M L
G32	Nagra	G	G	M	A	-	P	VW	M	P	C	P	A	P	S	LP	M	M	SE	SE	Sp	VL	W	A	A	w	M	M	A	-	A	Lg	s	F	W	w	GG	A	-	-	-	P	W	E-S	ME	VLI
G33	Danaguri	G	G	M	A	-	P	VW	M	P	C	P	A	P	S	LP	S	N	SE	SE	O	M	VW	W	A	LG	Tn	Sh	A	-	P	M	Ss	M	W	w	Sw	A	-	-	-	P	W	E	ME	M I
G34	Majhisal	C	G	M	A	-	P	VW	W	P	C	P	A	P	S	LP	M	M	SE	SE	O	M	VW	A	A	LG	Tk	Lg	A	-	A	M	s	F	S	B	GG	A	-	-	-	P	W	E-S	PE	M I
G35	Basnotti-local	C	G	M	A	-	P	VW	W	P	C	P	A	P	S	LP	S	N	SE	SE	O	M	VW	A	M	w	Tk	Sh	A	-	A	M	Ss	M	M	Y	Bt	A	-	-	-	P	S	E-S	ME	M I
G36	Netaisal	C	G	M	P	OMO	P	M	W	P	C	P	A	P	S	LP	M	M	SE	SE	SE	VL	VW	W	A	Y	Tn	M	A	-	A	VLg	Ss	m	W	Y	Bt	A	-	-	-	P	W	E	ME	VLI
G37	Sankar Kalma	C	G	M	P	OMO	A	-	M	P	LP	P	A	P	S	P	M	M	E	E	L	VW	A	A	w	M	Lg	A	-	A	Lg	Ss	m	M	B	Bt	P	B	M	TO	P	W	E	PE	L I	
G38	Rupsal	C	G	L	A	-	A	-	A	P	C	P	A	P	S	G	M	M	SE	SE	Sp	L	VW	A	A	P	Tk	Lg	A	-	A	Lg	Ss	M	W	P	Bs	A	-	-	-	P	W	E	PE	L L
G39	Jhingsal	C	G	M	P	IBO	A	-	M	P	P	P	A	P	S	LP	M	M	E	E	O	L	VW	A	VS	LG	Tk	Lg	A	-	A	Lg	Ss	M	W	Y	Bt	A	-	-	-	P	S	E	PE	L L
G40	Sungakalma	PL	M	P	OMO	A	-	W	P	C	P	A	P	S	LP	L	B	SE	SE	E	L	VW	A	A	w	M	M	A	-	A	Lg	s	M	M	w	Bs	A	-	-	-	P	W	E-S	PE	L L	
G41	Jhuli	P	PL	M	P	IBO	P	M	A	P	P	P	P	S	LP	L	B	SE	SE	E	E	W	A	A	w	M	M	A	-	A	VLg	Ss	m	M	w	Bs	A	-	-	-	P	W	E-S	PE	E I	
G42	Raja Badsa	C	PL	D	A	-	A	-	W	P	C	P	A	P	Ac	LP	L	B	E	SE	O	L	M	A	A	w	Tk	Lg	P	S	P	Lg	Ss	m	W	Y	Sw	A	-	-	-	P	W	E-S	ME	L E
G43	Kalma	C	PL	M	P	OMO	P	VW	M	P	C	P	A	P	S	LP	M	M	E	SE	Sp	M	VW	W	VS	LG	M	Lg	A	-	A	Lg	Ss	F	M	P	Pf	P	P	M	TO	P	W	E-S	PE	M E
G44	Sunga Nagra	C	PL	D	P	OMO	P	VW	M	p	C	P	A	P	S	LP	M	M	SE	SE	Sp	VL	VW	A	A	LG	M	M	A	-	A	VLg	Ss	F	W	w	Sw	P	P	M	TO	P	W	E	WE	VLI
G45	Kerala Sundari	C	PL	M	A	-	A	-	A	-	A	-	P	A	Ac	LP	L	B	E	SE	E	E	VW	A	A	w	M	M	A	S	P	VLg	s	M	M	Y	Sw	P	Yw	M	TO	P	W	E-S	PE	E I
G46	Balaramsal	C	PL	D	P	OMO	P	VW	M	P	C	P	P	P	S	LP	M	M	SE	SE	Sp	L	VW	A	A	LG	Tk	M	A	-	A	Lg	D	M	W	Y	Bs	A	-	-	-	P	W	E-S	WE	L I
G47	Danga	C	PL	M	A	-	A	-	M	P	C	P	A	P	S	G	L	B	SE	SE	SE	L	S	W	A	LG	Tk	M	A	-	A	Lg	Ss	M	M	B	Bf	P	Yw	L	TO	P	W	E-S	WE	L I
G48	Asanlaya	C	PL	D	A	-	A	-	S	P	C	P	A	P	S	G	M	M	E	SE	E	VL	VW	A	W	LG	Tk	M	A	-	A	Sh	s	F	W	B	GG	A	-	-	-	P	W	E-S	ME	VEI
G49	Lalhusri	C	PL	M	A	OMO	P	W	W	P	LP	P	A	P	Tr	LP	S	M	SE	DF	Sp	L	VW	A	W	Y	M	Lg	P	W	P	M	D	M	M	Y	Bs	A	-	-	-	P	S	Sr	WE	L L
G50	Annada	C	PL	D	A	IBO	A	-	M	P	C	P	A	P	S	LP	M	M	E	SE	SE	M	S	S	S	LP	Tk	Lg	A	-	A	Lg	s	M	M	B	Bt	P	P	VLg	WL	P	W	S-S	WE	M L
G51	Sarkele aman	G	PL	D	A	IBO	A	-	M	p	P	P	P	P	S	P	M	M	SE	Ho	O	L	VW	W	VWP	Tk	Lg	P	W	P	Lg	s	M	M	R	Bf	P	Yw	M	TO	P	S	E-S	PE	L L	

N.B. Place of collection: Zonal Adaptive Research Station (Z.A.R.S.), Krishnagar, Nadia, West Bengal

1.Coleoptile Colour, 2.Basal leaf sheath colour, 3.Intensity of green colour, 4.Anthocyanin colouration, 5.Distribution of anthocyanin colouration, 6.Leaf sheath anthocyanin colouration, 7.Intensity of anthocyanin-colouration, 8.Pubesence of blade surface, 9.Auricle, 10.Anthocyanin colouration of auricle11.Leaf collar, 12.Anthocyanin colouration of collar, 13.Leaf ligule, 14.Ligule shape, 15.Ligule colour, 16.Length of leaf blade, 17.Width of leaf blade, 18.Flag leaf (early observation), 19.Flag Leaf (Late observation), 20.Culm-stem attitude, 21.Time of heading, 22.Anthocyanin colouration of keel, 23.Anthocyanin colouration of area below apex, 24.Anthocyanin colouration of stigma, 25.Colour of stigma, 26.Stem, thickness, 27.Length, 28.Anthocyanin colouration of the node, 29.Intensity of anthocyanin colouration the node, 30.Intensity of anthocyanin colouration the internode, 31.Length of main axis, 32.Curvature of main axis, 33.Number per plant, 34.Density of pubescence of lemma, 35.Colour of tip of lemma, 36.Colour of lemma and pelea in spikelet, 37.Awns, 38.Colour of awns, 39.Length of longest awn,40.Distribution of awn,41.Presence of secondary branching, 42.Secondary branching, 43.Attitude of branches, 44.Exsertion, 45.Time of maturity, 46.Leaf senescence.

A=Absent, Ac=Acute, B=broad, B=black, By=Brown tawny, BS=Brown spot, BF=Brown furrows, PF=Purple furrows, C=Colourless, D=Drooping, D=Dark, DF=deflexed, E=erect, F=Few, G=Green, GG=Gold & Gold furrows, Ho=Horizontal, LP=Light purple, L=Long, LG=long, M=medium, ME=Mostly exserted, N=Narrow, O=Open, OTO=On Tip Only, OMO=On Margins Only, OBO=On Blotches Only, P=purple, PL=Purple Lines, P=Present, PE=Partly exserted, R=Red, S=Strong, SP=Split, SH=Short, SE=Semiexsert, S-E=Semiexsert to Spreading, S=Straight, SS=Semi Short, TR=Truncate, U=Uniform, VW=Very Weak, VE=Very Early, VLg=Very Long, VL=Very Late, W=White, W=weak, WE=Well exserted, Y=Yellowish.

In case of stem, cultivars with thick and very long stem were G17, G30 and G31. Besides, cultivars G12, G34, G38, G39, G42, G50 and G51 were distinguished for having thick stem with long stem length. Anthocyanin colouration was found in node of three cultivars *viz.*, G22, G42 and G49, among which G22 had strong colouration. Cultivars G42 and G49 have anthocyanin colouration in internode along G5, G6, G8, G33, G45 and G51.

In explaining the panicle features, cultivars G36 and G41 were distinguished for having very long panicle length along with many panicle numbers/plant with semi straight curvature on the main axis. Few cultivars like G16, G37 and G42 although having many panicle numbers/plant, have medium to long panicle length of main axis. Drooping curvature on main axis were found in 6 cultivars *viz.*, G8, G9, G18, G31, G46 and G49.

In case of density of pubescence on lemma on spikelets, it was absent in 5 cultivars *viz.*, G1, G2, G11, G24 and G25 but it was very strong in cultivars like G5, G12, G13, G20 and G34 which are very much distinct from the rest varieties. Also, colour of tip of the lemma was purple in varieties like G23, G38 and G43 but red in G51.

A large amount of variation in colour have been found in the colouration of lemma and palea (Table 6). Awn was present in 15 cultivars, out of which, very long awn was found in G25 and G50, long in G12, G21, G22 and G47 and very short in G16. The rest 36 cultivars were lacking this awn. Much variation was found in colouration of the awn (Table 1). Awns were distributed on whole length in G16, G23 and G50, only on upper half in G12 and on tips only in the rest of the cultivars (Table 1).

Thus, variations in anthocyanin pigmentation have been found in the plant parts, such as, apiculous, awn, leaf blade, coleoptiles, collar, hull (lemma and palea), internode, leaf sheath, leaf tip and margin, ligule, midrib and stigma. The colour variations were colourless (white), green pink, red and several shades of purple.

The genes controlling anthocyanin pigmentations are basically three complementary genes: C5, A and P6. The gene C is basic for producing chromagen, while A controls the conversion of chromagen into anthocyanin and P controls the distribution or localization of anthocyanin in specific plant organ or organs. Non-anthocyanin pigmentation generally involves a single pair or a series of alleles, such as *Bf* alleles for brown furrows on hull

at maturity, *Bh* genes for black hull and *gh* for gold hull (Anonymous, 1959).

Secondary branching was present in all the cultivars but strong branching was found in 16 cultivars. Well exertion of panicle was found in 7 cultivars like G15, G20, G44, G46, G47, G49 and G50. Cultivars like G36 and G48 were distinguished for very early time of maturity while 5 cultivars *viz.*, G23, G24, G28, G29, G30, G32 and G44 were distinguished for very late time of maturity. Seven cultivars had early but maximum accessions showed late time of maturity. Leaf senescence was early in 11 cultivars while it was late in 16 cultivars. Cultivars G28, G29, G32, G44 and G48 had intermediate while G30 had late leaf senescence (Table 1).

Studies on quantitative traits have earlier been made by Chakravorty and Ghosh (2011). Maximum plant height (43.0 cm) was obtained in variety G51 while minimum (24.0 cm) in variety G15. Data on 1000 grain weight among varieties varied from 10.4 g (in G26) to 29.91 g (in G51). G23 and G24 recorded the late maturity in days (172.5 days) while it was recorded to be shortest with G36 (116 days).

Thus, it is concluded that out of 51 landraces of rice, 27 cultivars were found to be distinctive on the basis of 22 essential and 24 additional characters.

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Systematic Studies on *Crotalaria tetragona* Roxb. ex Andr. (Fabaceae-Crotalarieae): A Wild Relative of Sunnhemp

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Crotalaria tetragona Roxb. ex Andr. (Fabaceae) considered to be close wild relative of cultivated *C. juncea* L. (sunnhemp) occurs in the tropical Asian region. Systematic study based on floral morphology and developmental aspects of *tetragona* was attempted. The study pointed out gross structural similarity of floral parts of this species with that of *C. juncea* but differences were apparent in development of floral parts.

Key Words: Androecium, *Crotalaria tetragona*, Developmental aspects, Electron microscopy, Dimorphic stamens, Fabaceae, Keel, Pollen morphology, Standard, Stigma, Systematic study, Taxonomy, Wild relative

Introduction

Floral traits are excellent subject of study for basic and applied research on plant systematics, plant breeding and reproductive biology (Hoc *et al.*, 2003; Ford and Johnson, 2008; Koul and Bhatnagar, 2007; Parsons and Luise, 2006). Data from floral morphological studies have provided baseline information on phenology, breeding, pollination biology, phylogeny, crop genepool study and plant systematics, patterns of evolution within species and rates of divergence between species (Etcheverry *et al.*, 2008; Polhill, 1982; Striton, 1981; Uga *et al.*, 2003).

Flowering plants display extraordinary diversity in morphology of floral traits, yet the functional significance of these variations is not well understood in many important taxa. Studies on variation in floral morphology have thrown light in understanding the pattern of adaptability in flowering and fruiting (Etcheverry *et al.*, 2008; Jacobi *et al.*, 2005; Patterson, 1982). The study of morphological structure of complex flower has helped in understanding the mating system, compatibility and seed set in members of Fabaceae including *Crotalaria* (Free, 1970; Kalisz *et al.*, 1999; Etcheverry, 2001b; Etcheverry and Aleman, 2005; Jacobi *et al.*, 2005).

Despite wide knowledge available on various aspects of floral biology of Fabaceae, the genus *Crotalaria* is least worked taxa for systematic potential especially for many indigenous taxa (Pandey and Nayar, 1994; Ansari, 2008). Morphology of floral parts has provided data of

relevance to taxonomy in different species of *Crotalaria* (Pandey and Nayar, 1994; Polhill, 1982). *Crotalaria juncea* L. known as sunnhemp, a source of the oldest cultivated fibre from Indo-Pakistan region (Watt, 1989) does not occur wild. *Crotalaria tetragona* Roxb. ex Andr., presumably a wild relative of *C. juncea* is a tall shrub distributed in tropical Asia. This species occurs as wild and semi-domesticated population and reported for ethnobotanical use (Bhatt *et al.*, 2009). It has gross morphological resemblance to *C. juncea* (Babu, 1977; Pandey and Nayar, 1994) but can be distinguished from the latter in general habit, tetragonous stem, linear-lanceolate leaves, longer pods (3.5x1.4 cm as compared to *C. juncea* with 2.1x1.2 cm) and larger seeds (0.51x0.43 cm as compared to *C. juncea* with 0.4x3.8 cm). Overlapping of many of the floral traits with *C. juncea* was suggestive of a close relationship between the two taxa and, thus, needs investigation on systematics (Babu, 1977; Baker, 1876). Earlier studies on *C. tetragona* have been carried out with respect to floristics (Baker, 1876; Babu, 1977; Ansari, 2008), anatomy (Mangotra, 1991), chemotaxonomy, karyobiology (Mangotra and Bhargava, 1989) and phenetics (Raj *et al.*, 2011). The present investigation was aimed at study of *C. tetragona* with respect to morphology and development of the floral parts more prominent in corolla and androecium characters to support plant taxonomy and systematic of the taxa.

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Materials and Methods

Seeds of *C. tetragona* collected from disturbed habitat of Kolasib district in Mizoram, India were grown in the experimental pots in the net-house at National Bureau of Plant Genetic Resources (NBPGR), New Delhi. The studies were undertaken for three consecutive years (2009, 2010 and 2011). The seeds were sown in earthen pots of 40 x 35cm size filled with standard potting mixture (containing sand, soil and farmyard manure in equal proportion). Watering was done every day once during July-October and at weekly interval during October-November. Fertilizers were applied with different doses of NPK (30:10:30) during vegetative phase (August-October) and NPK (10:30:10) during flowering season (October-December). Plants were continued to grow in the net-house until they started flowering after 3 months of germination. For floral observations inflorescence from middle internodes and flowers in the mid season were used. The data on floral morphology and structural developments were recorded from 9.0 am to 5.30 pm from mid October to mid December months continuously. Inflorescence length and number of flowers were recorded on primary and secondary branches during the entire experimental period. A total of 50 plants were selected for recording the observations on selected parameters from stages of bud initiation to flower senescence during peak flowering period (July-November). Estimates of number of flowers per plant were made by counts of total number of inflorescence per plant (using 50 plants) and flowers per inflorescence (50 inflorescences). To relate floral morphology and structural development within the inflorescence 50 flower buds of same size were marked (tagged) and observed thrice a day (9, 13 and 17 hours). Developmental stages of the flower were marked arbitrarily as six stages (day 0 to day 5) to record, correlate and interpret results (Table 1). Voucher specimens were dried according to standard herbarium techniques and deposited in the National Herbarium of Cultivated Plants (NHCP), NBPGR, New Delhi and herbarium of the Botanical Survey of India, Sikkim Himalayan Circle, Gangtok.

Data on inflorescence length, number of inflorescence, inter-floral distance, number of flowers per inflorescence and mean numbers of flower opened per day, flower size, standard size, wing size, keel size and ovary size were recorded and mean values calculated $\pm$ standard error (S.E.). Observations on weather data were recorded periodically for temperature (minimum and maximum

Table 1. Stages (days to open) of flower development recorded in *C. tetragona*

Stage (days to open)	Relative time to flower opening (hours)	Development recorded
0	-48	Bud primordial development
1	-24	Bud elongation with wide splitting of calyx lobes
2	0	Initiation of flower anthesis
3	12	Corolla half opened
4	24	Corolla fully opened with standard erected at right angle (flower fully developed); pollen burst from two types of anthers
5	48	Flower maturity with maroonish yellow corolla (initiation of flower senescence and pod primordia development)

average value) and rainfall pattern during course of study (meteorological data for 2009-11) and data presented as mean value. Pollen of the two species were collected from ten selected flowers from dehiscing anthers of two types of stamens i.e. short (SS) and long stamens (LS) separately at different growth stages (freshly opened to fully matured flowers) (day 2 onwards) and studied for micro-morphological characters. Floral morphology of the taxa was studied with freshly harvested selected buds/flowers and data recorded for morphology of separated floral parts in the inflorescence for characters. Measurement of floral parts was done to the nearest 0.5 mm. Androecium size was measured at different growth stages and calibrated image captured using Stereoscopic Zoom Microscope (SMZ1000/800; 10X-1000X magnification). Pollen and stigma structures were fixed in 2.5 per cent glutaraldehyde in 0.1 M phosphate buffer (pH 7.3) followed by ascending series of alcohol and allowed to air dry (Dykstra, 2003). Air dried samples were fixed on the SEM specimen stubs with a double-sided adhesive carbon tape and placed on the revolving discs of JEOL fine coat ion sputter (JEOL JFC 1100) for coating with gold. Each sample was uniformly coated with 20-30 nm thick gold. Images were taken under Scanning Electron Microscope (JEOL-JSM 840A) operated at accelerating voltage of 5-10 Kv. Structural details of micromorphology and dimension of pollen of two types of stamens were recorded. Observations on different growth stages of stigma and pollen were examined using compound microscope (Olympus CH40; 100-1000x magnification).

Results

Selected seeds were grown for three consecutive years in the months of July-August (2009-2011) (average maximum and minimum temperature 30.67 and 20.83°C; rainfall 3.87 mm). The first blossom appeared in end of September and continued till December. However, peak flowering was recorded during October-November. Floral characters recorded for six arbitrary stages from initiation of bud to pod initiation are given in Table 1. The observations showed that: a) Bud to flower senescence took 72-90 hours (over five days); b) Anthesis started in the morning at 9 hours (second day onwards) and completed by 24 hours (on fourth day); c) Flowers showed a distinct characteristic corolla (standard and keel); and d) Stamens had dimorphic anthers and showed distinct morphology and developmental aspects.

Structures and developmental aspects

The total number of flowers produced per plant were 95.23 ± 0.257 (N=50). The inflorescence was lax with terminal and axillary racemes, flowers arranged in acropetal maturation (base to apex). Terminal inflorescence was longer (260.3 ± 1.269 mm; N=50) than the axillary inflorescence (122.45 ± 0.61 mm; N=50). The terminal ones appeared during the first month of flowering followed by axillary inflorescence that were more in number more towards the second half of the December in the experimental period. The flowers were distantly arranged with inter-floral distance of 25.66 ± 0.096 mm (N=50) in terminal and lateral inflorescences. Single plant produced 7.16 ± 0.165 (N=50) inflorescences and total number of flowers per inflorescence was 13.30 ± 0.35 (N=50). Mean number of flowers opened at a time was 2.8 ± 0.26 (N=20) per inflorescence.

The bud primordia of *C. tetragona* increased from 3-9 mm to 20-25 mm within 24 hours before flower opening (stage 0; Fig.1a). The mature flower buds are beaked, lemonish-green, linearly elongated during one day before flower opening (stage 2; Fig.1b). The flower is beautiful, attractive lemon yellow colour, zygomorphic, protandrous and 39.60 ± 0.06 mm (N=20) long on drooping peduncle (clavate shaped, 1.4 cm; 1x3-1x4 mm). It was narrow (1mm) at base broadening (4mm) towards top and beared three markings, two on each side and one on the top. The calyx is bilipped and corolla has wing-keel complex with erect standard (vexillum) on the upper side (stage3-4; Fig.1c-d).

The bracts were setaceous (two, each 3 mm long), minute, located at base of peduncle; bracteolate (two; each 2.109 mm long). Calyx is gamosepalous, five sepals, lemonish green, soft brown, tomentose, with five teeth of variable length; two upper laterals are connate at base (20x3 mm) resting on the standard, two lower laterals and a lower median united laterally from base to half of the length (21x4 mm). Corolla is slightly exserted, size of standard 27.10 ± 0.1 mm x 25.45 ± 0.069 mm (N=20), suborbicular, apex mucronate, turned backwards in completely open flower (stage 4 onwards), spotted or streaked, a little wider than length, maroonish, ribbed on maturity; wings are ovate-oblong ($20.1 \pm 0.1 \times 10.20 \pm 0.06$ mm, N=20), spatulate, claw thick (ca 3 mm), dull lemon

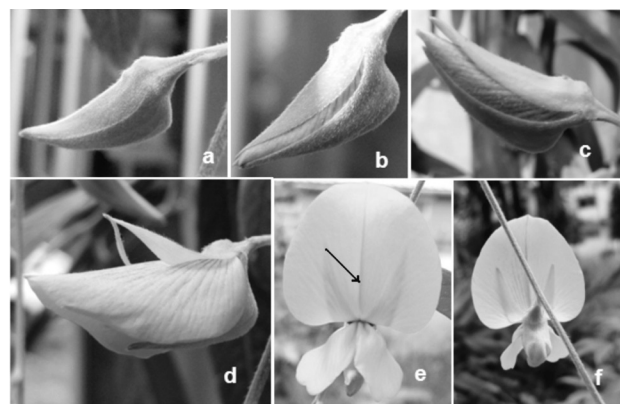


Fig. 1. (a-f) Stages of flower in *C. tetragona* a. One day old flower bud. b. Initiation of splitting of the calyx. c. Calyx splitting upto peduncle point (bilipped), lower half with three lobes and upper with two lobes. d. Corolla half open with calyx connate only at tip. e. Standard erect in fully opened flower with prominent wings, drooping on lower side and keel in the middle. f. Back side view of fully opened flower showing upper two lobes free and three lower lobes of calyx fused (for stages 0-5 refer to Table 1).

yellow with unique curvature; keel is lemon (20.50 ± 0.1 mm x 15.05 ± 0.1 mm; N=20), outcurved, conical on lower half, beaked with twisted tip; ovary subsessile (7.50 ± 0.02 x 2.6 ± 0.01 mm; N=20), tomentose, 10-18 ovules, stigma hairy; gynophore (a short stalk holding ovary; 4mm) and the furrow forming a chamber for nectar accumulation. The wing and the keel on the dorsal mid rib had thicker dark maroon coloured line and other radiating lines and folds on the lower half between veins (Fig. 3 d, e, f; see arrow). The basal part and blades of keel are fused on the lower margin upto tip by a genuine fusion. Terminal end of the keel is straight in bud but gradually twists leaving a small pore (ca 1 mm aperture) opening at the tip in mature flower (Fig. 2e,f). The bilipped corolla at base of gynophores, keel, ovary and staminal furrow formed a cavity to facilitate access by insect pollinators.

Androecium

Androecium is monoadelphous, composed of ten dimorphic, basifixed anthers with short stamens (SS) alternating with long stamens (LS). Short stamens (SS) have shorter filaments but longer anther lobes and dehisce during the 12 hours (initiation of anthesis; stage 3). They alternate with five long stamens (LS), having longer filaments but round anther lobes and dehisce 24 hours after anthesis (stage 4 onwards). The two types of anthers dehisce longitudinally and showed differential growth. However, the dimensions of the pollen grain from two types of stamens did not show any significant difference.

The filament of both types of stamens remained turgid up to third day (stage 4) but afterwards floral senescence started with initiation and twisting of filament of SS (Fig. 3d). Pollen was tri-colpate, 3-zonocolporate with microperforate to faintly reticulate exine (Fig. 2). The shape of pollen grain was prolate and size $34.3 \times 14.3 \mu\text{m}$ (Fig. 2).



Fig. 2. a. Pollen grains of *C. tetragona*. b. Enlarged pollen grains with markings on exine surface. c. Microperforate ornamentation on exine wall

In young buds, SS have larger primordia than the LS (Fig. 3a). At the time of flower anthesis (stage 2), filaments of the SS are slightly longer than the entire LS (Fig. 3b). Filaments of all the stamens join into a furrow surrounding the ovary. From second day onwards there is significant increase in filament length of both types of stamens; the filament of the LS supersedes the SS (stage 3). By third day, both the filaments and anthers of the two types showed further development: the filament of SS grows from 1.85 mm in the bud to *ca* 9.55 mm in 24 hours after anthesis whereas the LS grew from 2.0 mm to 12.8 mm in the same period and further more up to 14.15 mm (Fig. 4). Dehiscence of anthers took place on the day 2 in SS and on day 3 onwards in LS, forcing the mass of pollen progressively into the conical and twisted tip of the keel around style. The size of anther lobes of SS was five time bigger than that of the SS and produced more quantity of pollen grains that remained viable for longer floral cycle as compared to that of the

LS. The filament of LS grew around the stigma and shed all pollen leaving shrivelled anthers.

In *C. tetragona* at stage 1 filament of the two types started elongating, but anthers were not above carpels. At the stage 2 filaments partially elongated, anther beginning to emerge body, but not above. When after 12-16 hours at stage 3 filaments were elongated, anther above carpels tops but anthers did not dehisce. Lastly at stage 4 onwards, filaments were fully elongated, occur parallel to the body of style, and both anthers showed anther dehiscence (SS bursts first followed by LS). At stage 5, the anthers were abscised with senescence of other flower parts such as sepals, petals, etc.

Gynoecium

The gynoecium of *C. tetragona* had short gynophores at bud stage. The ovary is fully covered with long, pubescent and trichomatous golden hairs. During the floral cycle, style at the base formed a notch (more notched when senescence followed) from straight (180° at 0 stage) to an acute/ right angle ($80-90^\circ$) at stage 4 (Fig. 3a-c). Along the inner side of style long golden hairs occurred uniformly. A tuft of peristigmatic whorl of hairs surrounded the stigma (Fig. 4a-b). The stigma

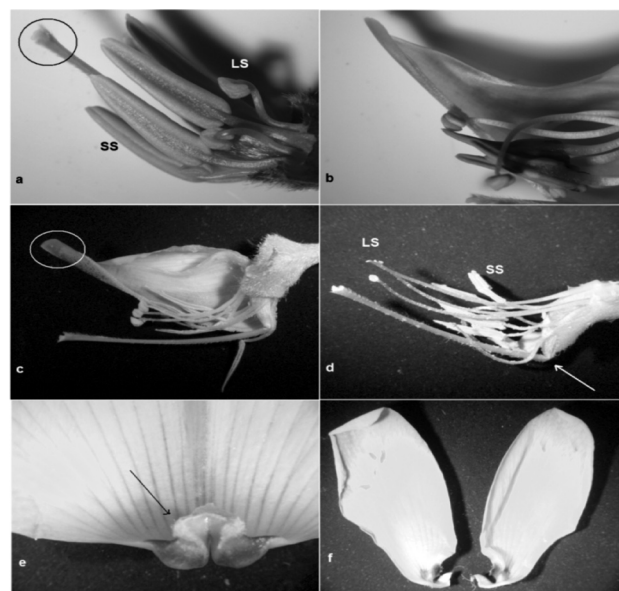


Fig. 3 (a-f) Floral parts of *C. tetragona* : a. Bud showing SS longer than the LS surrounding the style at straight angle, stigma (encircled). b-d Elongation of LS, twisted keel in flowers at stage 3, 4 (encircled), notched style (arrow); LS reaching near the stylar end, see hairy stigma and style covered with pollen. e Standard in the close up; see small claws (arrow), mid rib and radiating veins. Wings with coloured markings and deep curvature; periodic change in angle of style (from straight to 90° degrees) and hairy stigma (see in stages a-d)

was obliquely terminal and relatively small ($386 \mu\text{m} \pm 5.64$, $N=10$) (Fig. 4a). Stigmatic surface was composed of two types of epidermal papillae, the longer ones in centre showed increase in length at different stage of gynoecium development (Fig. 4c). Gynoecium showed a growth from 12 mm at bud stage to 24 mm full maturity (Table 2).

Discussion

In the present study, we investigated the floral morphology with special emphasis to the structure of androecium and gynoecium development in the entire cycle (bud-flower senescence) of *C. tetragona*. These floral structures and developmental stages were distinct when compared with related *C. juncea* (Pandey and Nayar, 1994; Pandey *et al.*, data unpublished). Despite of much resemblance

in flower morphology of the two taxa, structure and development of anther and stigma were very distinct. The key observations on the investigation carried out are discussed below in terms of application in taxonomic and systematic study.

General Floral Morphology

Floral morphology and study of development of parts have provided a better understanding of relationship and evolutionary pattern in different taxa under order Fabales (Bellow *et al.*, 2007; Etcheverry *et al.*, 2008). The investigation made on morphology and floral development revealed the complex structure of *C. tetragona* flower exhibiting sequence of changes in floral parts *viz.* petals, androecium and gynoecium (Pandey and Nayar, 1994; Ansari, 2008).

Characteristic floral morphology with deeply cleft two lipped (bilipped) calyx, as long as corolla, protracted downwards, keel tip spirally twisted from inception, corolla appendages restricted to the blade, standard marked with reddish-brown lines, and style curved (notched) with conical lower half have shown the similarity with *C. juncea*. Similar observations were reported by other workers (Ansari, 2008; Pandey and Nayar, 1994). These observations supported taxonomic placement of *C. tetragona* under section Calycinae. Data on floristic studies and presence of flavonoids have also supported placement under section Calycinae (Polhill, 1982; Mangotra and Bhargava, 1989; Ansari, 2008).

Inflorescence, Calyx and Corolla Structure

Inflorescence of *C. tetragona* was distinctly lax with more number of flowers. Inflorescence were observed to be terminal or lateral, lax with long peduncle and 120-360mm long as recorded by other workers (Singh *et al.*, 2000, 2002; Ansari, 2008). In general in *Crotalaria* species the inflorescence are terminal, lateral or both, rarely capitate (Ansari, 2008). The flowers were arranged in acropetal succession of opening that increased chances of pollination under unsuitable environment. This observation was also recorded in many Papilionaceous flowers (Suzanne Koptur, 1983; Etcheverry, 2001a; Caruso, 2006). Slow pace between the opening of consecutive flowers (2-3 flowers opened at one point of time) in contrast to related species *C. juncea* where the time gap between the consecutive flower was lesser (at one time almost half the flowers of a mature inflorescence

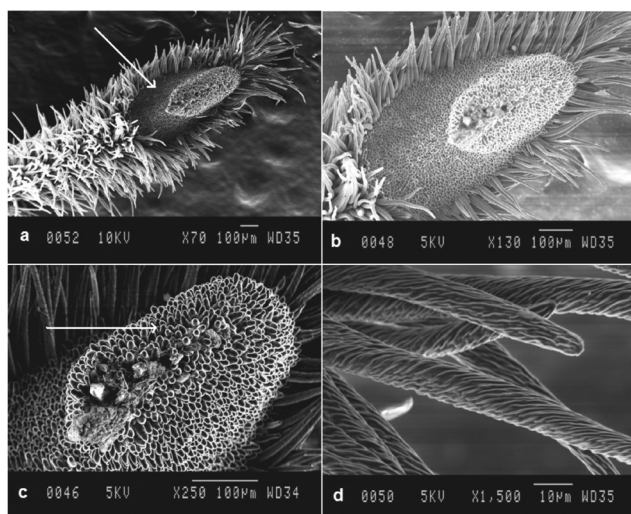


Fig. 4. SEM photomicrographs of the stigma of *C. tetragona* (a-d). a Stigma at stage 2 showing peristigmatic hairs (arrow). b Closer view of stigma showing distinct long and short epidermal papillae on the surface. c Stigmatic surface with epidermal papillae in enlarged view (arrow). d Peristigmatic hairs in closer view

Table 2. Average growth and development of androecium and gynoecium of *C. tetragona* at different stages

Stage (days to open)	Relative time to flower opening (hours)	Filament of short stamen (SS) mm	Filament of long stamen (LS) mm	Length of gynoecium (mm)
0	-48	1.85	2.0	12.40
1	-24	3.7	2.3	13.05
2	0	5.5	10.1	21.85
3	12	7.0	11.1	22.90
4	24	9.55	12.8	23.65
5	48	9.55	14.15	24.70

were opened) were distinguishable feature (Pandey *et al.*, data unpublished). These differences could be related to distribution of two species.

Development of Floral Parts

The flower of *C. tetragona* showed a typical keel blossom type of corolla with floral morphological adaptations such as bright colour of petal and wings structure, presence of nectar, and odoriferous flowers. Similar observations pointed out in other taxa by Westerkamp (1997) were indicative of adaption for pollinators. In many Papilionaceae including *Crotalaria micans*, *C. incana*, *C. pumila* and *C. stipularia* micro-morphology of wings exhibited modification into a pocket-like sculpturing as footholds during the pollinators foraging activities (Etcheverry, 2001c; Solomon and Purnachandra, 2006). In this species standard erect backwards in fully opened flower (stage 4) with prominent markings on the wing, drooping on lower side support the sitting of pollinator and keel in the middle which coincides with its body (Fig. 1e). In *C. juncea* the flowers were not drooping, and in fully opened flower the standard turned at right angle position. In various members of tribe Vicieae the flowers showed distinct structural morphology of androecium and pistil (Gunn and Kluve, 1976).

Androecium

In Fabaceae development of keel (gradual twisting) and androecium were suggestive of economic utilization of pollen that co-evolved with the pollinators (Hoc *et al.*, 2003; Prenner, 2004; Etcheverry *et al.*, 2003; 2008). In *C. tetragona* structural changes linked to differential maturity of androecium and pollen grains in two types of stamens were also observed by Etcheverry *et al.*, (2003). These changes coincided with corolla growth (stage 3) may be adaptive features that suggested of pollinator-mediated efficient mechanism, especially under unsuitable environmental conditions. Despite of morphological similarity of anthers of *C. tetragona* with that of *C. juncea*, in the latter species long anthers burst much before the opening of flower at 2 stages (Pandey *et al.*; data unpublished).

The pollen grains from both types of anthers were structurally similar as observed by many authors (Etcheverry *et al.*, 2003; Ansari, 2008). Pollen grains were prolate in shape, tri-colpate and 3-zonocolporate with faintly reticulate exine. Nair (1965) has reported 3-zonocolporate type of pollen grains being most common among family Fabaceae.

Gynoecium

Stigma architecture provides data for taxonomic delimitation and classification at infra-family levels in various taxa under Fabaceae (Ghosh and Shivanna, 1982; Venkata Lakshmi and Shivanna, 1985; Shivanna and Owens, 1989; Owens and Lewis, 1989, 1996; Etcheverry *et al.*, 2003). Some closely related species are distinguishable on the basis of pollen brush type of style and thus have proven taxonomic utility (Lavin and Delgado, 1990; Kang and Zhang, 2009).

In *C. tetragona* the stigmatic surface showed epidermal papillae in the centre forming highly receptive area (Fig. 4). Similar structure was described in *Crotalaria micans* by Etcheverry *et al.* (2003). A tuft of peristigmatic whorl of hairs surrounded the relatively small stigma, gynoecium forming a typical notch at the place of ovary and style junction and trend of hairiness along the inner side of style were characteristic features of this species. *C. juncea* could be distinguished by shorter peristigmatic hairs surrounding small stigma with protruding tip and thin hairs present only on the inner side of style. Similar observations were helpful in distinguishing different taxa under *Crotalaria* (Pandey and Nayar 1994; Ansari 2008).

Conclusion

On the basis of above observations we might interpret the two taxa, *C. tetragona* and *C. juncea* closer as pointed out by several workers (Babu, 1977; Ansari, 2008; Pandey and Nayar, 1994). In various legume taxa similar observations have been reported by Tucker (2003). Currently there is insufficient information on floral aspects of related taxa especially in genus *Crotalaria* to evaluate them for taxonomic interpretation. The study could be further extended for larger crop genepool of *Crotalaria* from the Indian subcontinent showing tremendous diversity and endemism.

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Diversity Distribution Pattern in Collected Germplasm of Rapeseed-Mustard using GIS in India

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A total of 6,923 accessions of rapeseed-mustard germplasm were collected during 1976-2011 through crop-specific and multi-crop explorations in collaboration with Indian Council of Agricultural Research (ICAR) crop-based institutes and State Agricultural Universities (SAUs) from different diversity rich areas of the country. Out of 6,923 accessions, 5,358 were examined for the status of diversity distribution pattern, which showed representation of maximum accessions from Gangetic, north-western plains and western Himalayan regions of the country. The geo-referenced map of collected diversity also indicated that parts of north-western plains (mainly western Uttar Pradesh), Himachal Pradesh and Uttarakhand have been extensively surveyed for collections of *Brassica* germplasm. In addition, grid mapping technique (GIS based) was also used to know the diversity rich areas and occurrence of trait-specific germplasm of rapeseed-mustard in different parts of the country. The priority areas and diversity distributed in different agro-ecological regions identified for further collections have been discussed in the present communication.

Key Words: Diversity distribution, GIS, Grid map, Rapeseed-mustard, Trait-specific germplasm

Introduction

The genus *Brassica* L. (Brassicaceae) possess different annual or biennial species, is considered to be the potential source of edible oils, vegetables, condiments and pharmaceuticals. In this genus about 37 species are distributed across the globe (Warwick *et al.*, 2000; Kumar *et al.*, 2004). *Brassica* is a prospective, economically important genus contributing to meet the challenges related to climate change, enhanced production and productivity as well as consumer's need (Singh and Sharma, 2007). Two important cultivated brassicas in India viz., *Brassica juncea* (L.) Czern. and *B. rapa* L. exhibit rich variability mainly in parts of northern, North-western, central and North-eastern regions (Singh and Sharma, 2007; Kumar *et al.*, 2004).

In India, rapeseed-mustard includes six important annual oilseed brassicas namely Indian mustard (*B. juncea*) commonly known as rai, raya or laha, three ecotypes of Indian rapeseed (*B. rapa* syn *B. campestris*) viz., toria, brown sarson (lotni and tora types) and yellow sarson, Swede rape or gobhi sarson (*B. napus* L.), Ethiopian mustard or karan rai (*B. carinata* Braun.) and black mustard [*B. nigra* (L.) Koch]. The other minor oilseed brassicas grown in dry areas of Rajasthan and Haryana are taramira or tara (*Eruca sativa* Mill.) and

wild turnip (*Brassica tournefortii* Gouan) (Chopra and Prakash, 1991; Minnich and Sanders, 2000; Hanelt, 2001). In Indian sub-continent, *B. juncea* is the dominant species grown along with *B. rapa* that constitute the major sources of edible oil, while *B. oleracea* L. is used as vegetable in most parts of the country. Rapeseed-mustard has the ability to grow under varied agro-climatic conditions relatively under low temperature as well as in the extremes of temperate regions (Downey, 1983; Hanelt, 2001). Since long history of their cultivation and acclimatization in India, remarkable variability has been generated in rapeseed-mustard (Rana and Singh, 1992; Duhoon and Koppa 1998). The Himalayan region is considered to be the secondary centre of diversity for *B. rapa* (Zeven and de Wet, 1982). Two exotic species namely gobhi sarson (*B. napus*) and karan rai (*B. carinata*) have become popular among the farmers in those areas where winter spell is longer.

In India, so far limited studies have been conducted on the diversity distribution pattern of genus *Brassica*. Keeping this in view, DIVA-Geographic Information System (GIS) was used to analyse the passport data of collected rapeseed-mustard germplasm to identify the diversity rich pockets in the country. Similar studies have been conducted by various workers on wild potatoes

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(Hijmans *et al.*, 2000), *Jatropha curcas* L. (Sunil *et al.*, 2009), *Vigna mungo* (L.) Hepper (Abraham *et al.*, 2010), etc. in different parts of world. Recently, application of GIS has earned recognition as a useful tool for effective management of plant genetic resources (Jarvis *et al.*, 2002; Dutta, 2008).

Materials and Methods

The germplasm collected from different agro-ecological regions belonging to more than 3,000 collection sites was examined using information recorded in plant exploration database, plant collection reporter, annual reports and other published literature (Plant Germplasm Reporter 2008-09; Annual Reports NBPGR 1976-2011; Annual Report Directorate of Rapeseed Mustard Research, 2011). To shortlist the accessions with required passport information, a rigorous screening of 6,923 accessions of rapeseed-mustard was done and a total of 5,358 accessions were short-listed with details on state, district, village, collector number, Latitude (N) and Longitude (E). Mainly six *Brassica* species viz., *B. juncea*, *B. napus*, *B. nigra*, *B. oleracea*, *B. rapa* and *B. tournefortii* were used for present analysis. Geo-referenced maps were prepared using WGS84 datum and Everest projection systems. In order to know the spatial distribution and assessment of richness, DIVA-GIS version 7.3 was used for point to grid analysis using simple-circular neighborhood method (Hijmans *et al.*, 2001; Saran *et al.*, 2010). The tool for conversion of the point data into grids which represents the germplasm collection sites was used. A grid of 1° x 1° cells (111 x 111 kms) to assign point to grid cells to map collected germplasm diversity was used for the country level data analysis (Hijmans *et al.*, 2000).

Results and Discussion

Germplasm Collection

The collected diversity includes *B. juncea* (2,078 accessions), *B. napus* (157 accessions), *B. nigra* (53 accessions), *B. oleracea* (485 accessions), *B. rapa* (2,573 accessions) and *B. tournefortii* (12 accessions) mainly from northern and north-western states of the country. The collection of rapeseed-mustard gained momentum under mission mode sub-project on Sustainable Management of Plant Diversity of the World Bank-funded National Agricultural Technology Project (NATP) during 1999-2005 in collaboration with crop-based institutes of ICAR and State Agricultural Universities. Considering the representation of aforesaid diversity from all surveyed

states in India, Uttar Pradesh (838), Uttarakhand (673), Himachal Pradesh (499) and Bihar (328) were most explored states in terms of availability of germplasm accessions.

In oleiferous brassicas, *B. rapa* germplasm was assembled from Assam plains, parts of Bihar, Jharkhand, Chhattisgarh, Madhya Pradesh, West Bengal, Himachal Pradesh, Rajasthan, Uttar Pradesh and Uttarakhand; *B. juncea* from parts of Andhra Pradesh, Madhya Pradesh, Uttar Pradesh and Uttarakhand. *B. nigra* from Punjab, Haryana, Andhra Pradesh, Himachal Pradesh and Rajasthan; *B. tournefortii* from sandy tracts of Rajasthan and Haryana. The collected accessions were also pooled according to phyto-geographical/agro-ecological regions in order to know the region that represents maximum collections (Arora, 1991). It was observed that the highest collections were made from Gangetic, north-western plains and western Himalayan regions as compared to other regions of the country (Fig. 1).

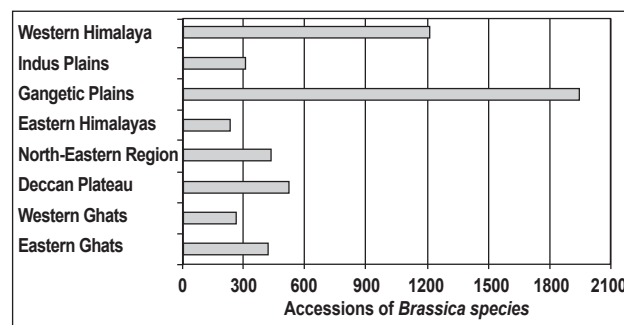


Fig. 1. Rapeseed-mustard germplasm collected from different phyto-geographical/agro-ecological regions of India

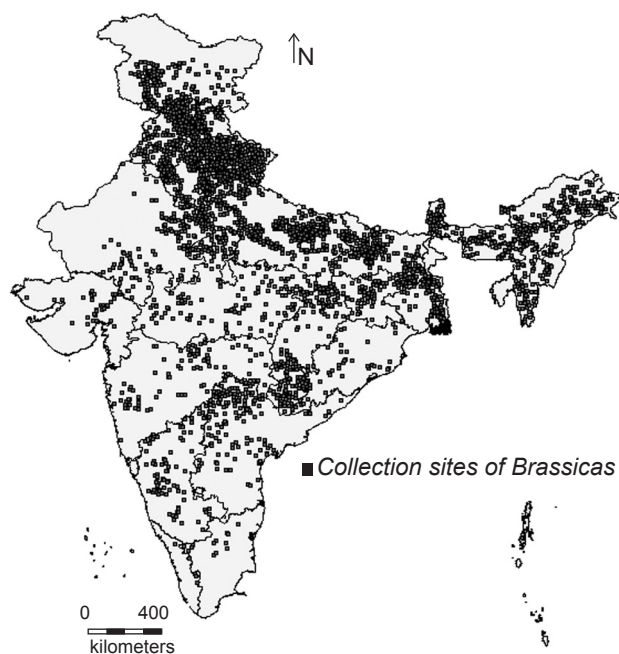
Some of the unique collections made during the period include yellow-seeded *toria*, dwarf mustard, dwarf and early *toria*, white flowered yellow *sarson* etc. (Singh and Sharma, 2007). The details of species-wise accessions collected from different agro-ecological regions are given in Table 1.

Mapping of Collected Diversity and Areas Surveyed

To know the eco-geographic diversity distribution of rapeseed-mustard through mapping the sites, passport data of collected germplasm were short listed in order to find out the gaps in exploration and germplasm collection (Fig. 2a). Mapping of diversity collected indicates that some of the diversity rich areas viz. parts of north-western plains (foothill regions of Himachal Pradesh, Uttarakhand and Western Uttar Pradesh); eastern and north-eastern parts (Bihar, Jharkhand, Assam and West Bengal) were extensively surveyed for germplasm collection.

Table 1. Species-wise germplasm diversity in rapeseed - mustard collected (Total number of accessions given in parenthesis for each species separately) from different states in India

Species (acc.)	Status of areas explored		
	Extensively explored	Moderately explored	Less explored
<i>B. juncea</i> (2,078)	Andhra Pradesh (170), Bihar (104), Haryana (103), Himachal Pradesh (92), Madhya Pradesh (156), Punjab (81), Uttar Pradesh (439) and Uttarakhand (265)	Assam (56), Chhattisgarh (53), Gujarat (38), Jharkhand (57), Karnataka (84), Maharashtra (41), Mizoram (69), Rajasthan (80) and West Bengal (76)	Arunachal Pradesh (33), Meghalaya (30), Nagaland (26) and Sikkim (22)
<i>B. napus</i> (157)	Himachal Pradesh (72) and Punjab (27)	Jammu & Kashmir (21) Uttar Pradesh (24)	Jharkhand (5) and Uttarakhand (8)
<i>B. nigra</i> (53)	Andhra Pradesh (10) Himachal Pradesh (16) and Rajasthan (8)	Haryana (7) and Tamil Nadu (7)	Jammu & Kashmir (5)
<i>B. oleracea</i> (485)	Jammu & Kashmir (154), Uttar Pradesh (104), Himachal Pradesh (63),	Arunachal Pradesh (37), Assam (19), Chhattisgarh (11), Maharashtra (30), Odisha (11), Punjab (18), Rajasthan (28), and Uttarakhand (22)	Meghalaya (12), West Bengal (6)
<i>B. rapa</i> (2,573)	Assam (135), Bihar (224), Chhattisgarh (188), Haryana (98), Himachal Pradesh (256), Jharkhand (168), Madhya Pradesh (116), Uttar Pradesh (271) Uttarakhand (348) and West Bengal (94)	Arunachal Pradesh (75), Gujarat (15), Mizoram (28), Maharashtra (26), Odisha (61) Punjab (33), Rajasthan (58), and Sikkim (68)	Kerala (4) Manipur (15), Meghalaya (13), and Tripura (4)
<i>B. tournefortii</i> (12)	–	Haryana (7)	Rajasthan (5)

**Fig. 2a. Geo-referencing of germplasm collection sites of rapeseed - mustard in India**

The grid map shows that the highest numbers of accessions in rapeseed-mustard were collected from Uttar Pradesh (Table 1) covering 51 districts whereas remaining 24 districts may be considered under explored, depending on the availability and richness of crop diversity. Grid maps were generated for identifying the species richness

areas or niches where rapeseed-mustard is predominantly grown. In the map, grid nos. 3-3 and 4-5 shows that the maximum number of *Brassica* species are distributed in this area as compared to other regions (Fig. 2b). Grid map also clearly shows that Gangetic plains, Western Himalaya and parts of North-eastern hill region would be target areas for future collections of rapeseed-mustard germplasm.

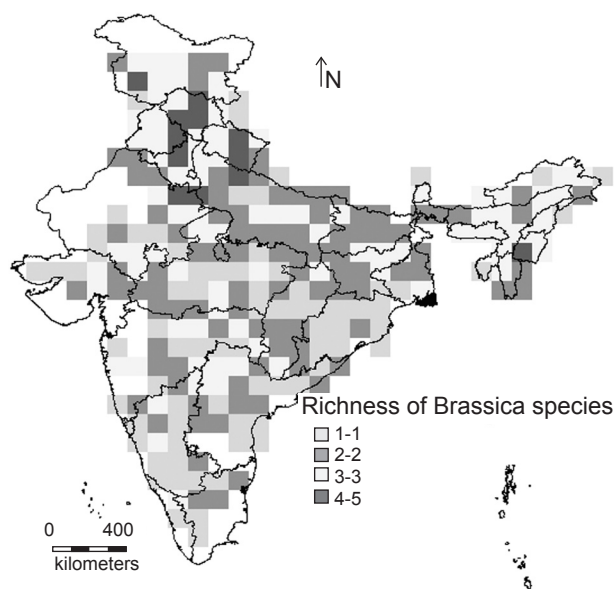
**Fig. 2b. Grid map showing species richness in rapeseed-mustard germplasm collected from different regions of India**

Table 2. Areas identified for trait-specific germplasm of rapeseed-mustard

Traits	Species	Accessions	State(s)
Dwarf habit	<i>B. juncea</i>	IC212077	Uttarakhand
	<i>B. rapa</i>	IC212022, IC267713, IC268307, IC374358, IC395569	Bihar (2), Chhattisgarh, Jharkhand, Uttar Pradesh
Early maturity	<i>B. rapa</i>	IC214824, IC267713, IC268307, IC76659, IC261641, IC261646, IC395569, IC212077, IC212022, IC212032	Andhra Pradesh, Bihar, Chhattisgarh, Haryana, Uttar Pradesh (3), Uttarakhand, West Bengal (2)
	<i>B. nigra</i>	IC426329, IC426354	Andhra Pradesh
High oil content	<i>B. juncea</i>	IC342780, IC203215	Bihar, Jharkhand
	<i>B. rapa</i>	IC248980, IC248990, IC426303, IC23003, IC23084, IC76659, IC35198, IC19955, IC343121, IC385678	Bihar (2), Chhattisgarh (2), Haryana, Himachal Pradesh, Jharkhand (2), Uttar Pradesh (2)
	<i>B. nigra</i>	IC426303	Andhra Pradesh
Long siliqua	<i>B. juncea</i>	IC385781	Jharkhand
	<i>B. rapa</i>	IC268327, IC343122, IC385663, IC385664	Madhya Pradesh, Uttar Pradesh (3)
Multi-locular pods	<i>B. rapa</i>	IC267710	Bihar

(Source: NBPGR Annual Reports, 1976 – 2011)

Variability Observed and Trait-specific Germplasm Collected

During germplasm collection, rich variability in different agro-morphological traits of indigenous rapeseed-mustard germplasm was observed in various forms viz., plant height, crop duration, branching pattern (basal, medium and top), appressed, semi-appressed, siliqua length, number of seeds/siliqua, seeds size (small and bold), seed colour (brown, dark brown, reddish brown and yellow), etc. Dwarf and early types were collected from north-eastern hill region of India, whereas late, tall and small seeded types from north-western Himalayas. In yellow sarson, germplasm with compact and tall habit, deep green, dissected leaves and waxiness texture at flowering and maturity stages was mainly from eastern Uttar Pradesh, Bihar and West Bengal (Table 2). The collected germplasm was characterized using minimal descriptors and the variability for superior traits in rapeseed-mustard germplasm was observed for plant height, maturity period, high oil content, multilocular pendants and long siliqua (Singh and Sharma, 2007).

On the basis of areas identified (Table 2) for collection of trait-specific germplasm in rapeseed-mustard, area nos. 4-4 and 5-5 in grid map represent regions with high diversity/ more number of promising traits (Fig. 3) while green areas represent regions with low diversity or occurrence of minimum promising traits. Grid map also depicts diversity pattern for collection of early types in few pockets from Chhattisgarh, Haryana, Uttar Pradesh, Uttarakhand and West Bengal; tribal dominated areas in Bihar, Jharkhand and Chhattisgarh for high oil content, and for leafy types from Uttarakhand and Himachal Pradesh. The collected germplasm exhibits good diversity for plant types, seed size, siliqua length and maturity period in *B. juncea* mainly from northern parts of India. There was outsized variation reported in plant habit, flowering period, siliqua size, seed size and number, maturity period and oil content in yellow sarson. Moreover, variability in *B. juncea* and *B. nigra* was also collected from drier tracts of Andhra Pradesh, Haryana and Rajasthan showed variation in seed size and seed colour and siliqua size. *B. tournefortii* was collected with seed colour (brown, dull brown and yellow), compact type with upright branches, leaves and pods, bushy or spreading type with horizontal branches, siliqua and leaves, early to late maturity, dwarf and tall types from parts of Rajasthan and Haryana (Plant Germplasm Reporter 2008; NBPGR Annual Reports, 2008 - 2011).

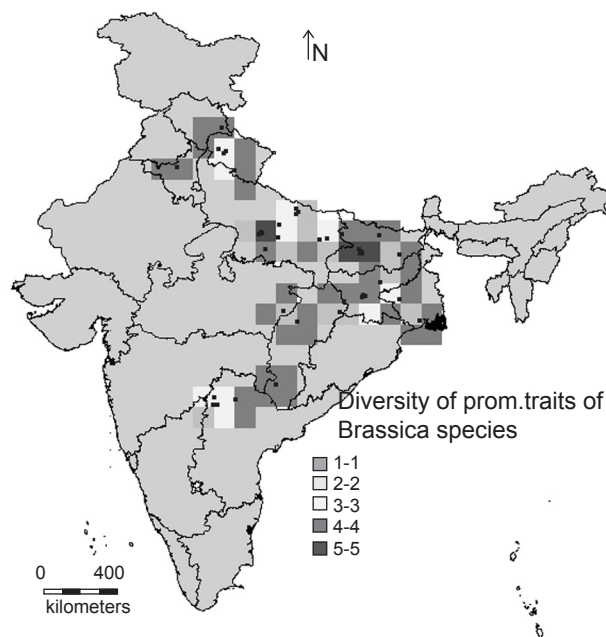


Fig. 3. Grid map showing trait-specific germplasm of rapeseed-mustard from different regions of India

Diminishing Areas under *B. nigra* Cultivation: Current Observations

The analysis of passport data of rapeseed-mustard indicated the reduction in areas of *Brassica nigra* mainly due to changing patterns of food habits, which was cultivated sporadically as a condiment in Kumaon region of Uttarakhand and peninsular region of Karnataka, Andhra Pradesh and Tamilnadu. At present, this species has completely been replaced by small seeded *B. juncea*. In North and southern districts of Karnataka and North-western districts of Andhra Pradesh, rapeseed-mustard is grown as mixed crop with other cereals, pulses and oilseed crops during *kharif*. Substantial reduction in areas under cultivation of rapeseed (*B. rapa*) and black mustard (*B. nigra*) was observed due to their susceptibility to diseases and pests and low productivity resulting in their replacement by brown mustard (*B. juncea*). In the states of Punjab, Haryana and Rajasthan, *B. tournefortii* was a common weed in drier parts up to 1960, but now it is rare in occurrence. It is distributed in dry tracts of Rajasthan, Haryana and Punjab with high concentration of diversity mainly on parched sand dunes. Disappearance of *B. tournefortii* from growing areas was recorded in last three decades as commercial hybrids have been adopted for large scale production.

Conclusions

Different agro-ecological regions of the country are rich in rapeseed-mustard germplasm diversity. Keeping in view the diversity assessment of rapeseed-mustard *vis-à-vis* germplasm collecting, the following areas have been suggested for future exploration and collection of *Brassica* spp. germplasm:

- Wild and weedy relatives/related species/taxa of rapeseed-mustard are very limited. There is an urgent need for strengthening the germplasm of wild and weedy relatives such as *Crambe*, *Sinapis* etc. from temperate, alpine region of Jammu & Kashmir, Himachal Pradesh and Uttarakhand.
 - Trait- specific *Brassica* germplasm have been identified for future germplasm collection based on their presence in respective priority areas.
1. Yellow sarson (*B. rapa* var. yellow sarson) early types and dwarf types need to be collected from tribal dominated Bastar region of Chhattisgarh.
 2. Black mustard (*B. nigra*) germplasm should be collected from Bageshwar and Pithoragarh districts of Uttarakhand.

3. Toria (*B. rapa* var. *toria*) need to be collected from Banka, Bhagalpur and Katihar districts of Bihar.
4. Leafy mustard (*B. juncea* var. *rugosa*) need to be collected from East Siang, West Siang and Kurung Kumey districts of Arunachal Pradesh; Almora and Pithoragarh districts of Uttarakhand.
5. Germplasm of *B. tournefortii* required to be collected from Fatehabad and Sirsa districts of Haryana; Churu, Barmer and Sikar districts of Rajasthan.

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SHORT COMMUNICATION

Variability Studies in Seed Morphology of Castor (*Ricinus communis* L.) Collected from North-eastern Region of India

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A total of 90 accessions of castor germplasm represented from north-eastern region of India were studied for seed morphological characteristics viz. seed length, seed breadth, caruncle size, mottling, seed surface texture, seed coat thickness and seed coat colour. Morphological variations were observed among different accessions for seed characters and significant correlation was recorded among seed morphological traits.

Key Words: Castor, Genetic diversity, Mottling, Seed Morphological Characters, Variability

Castor (*Ricinus communis* L., family Euphorbiaceae) is an important non-edible oilseed crop considered to be native of tropical Africa (Vavilov, 1951; Zeven and Zhukovsky, 1975) and now extensively cultivated in tropical and subtropical regions of world. The genus *Ricinus* is considered monotypic, with significant variation in plant habits, seed morphology and oil content (Kulkarni and Ramanamurthy, 1977; Li *et al.*, 2008).

India is the world's largest producer of castor seed with 65 per cent contribution of total world production (<http://www.crnindia.com/commodity/castor.html>). The Indo-gangetic plains are one of the centres of variability of castor. The species is common throughout the country particularly in the drier areas but cultivated commercially for extraction of oil from seeds and rearing silk worm for production of silk in north-eastern region especially in Assam and Meghalaya (Sarmah, 2010). The species depicts wide variability in cultivated and wild germplasm in plant habit and seed characters from diverse habitats. Variability studies on this species have been carried out on germplasm collected from the Upper Gangetic plains and adjoining regions of the Indian subcontinent (Anjani *et al.*, 1993; Duhoon *et al.*, 1996; Pandey and Radhamani, 2006).

The present study was undertaken on 90 accessions from different states of north-eastern region viz. Assam, Arunachal Pradesh, Meghalaya, Manipur and Nagaland assembled during 2004-2006 and available as representative vouchers in the National Herbarium of Cultivated Plants (NHCP), National Bureau of Plant Genetic Resources (NBPGR), New Delhi. A total

of seven morphological traits (seed length, breadth, caruncle size, mottling, colour, seed coat thickness and seed surface) were studied. The seeds were categorized on the basis of length and breadth ratio. Seed length, breadth and caruncle size were measured with the help of digital vernier calipers. Other morphological characters viz., mottling, seed coat colour, and seed surface were assessed visually. To quantify the data coding was done for mottling (nil-0, less mottled-1, medium mottled-2 and maximum mottled-3); seed coat colour (creamish brown-1, light brown-2 and dark brown-3); seed surface (dull-1, medium shining-2 and maximum shining-3); seed shape (categorized and coded as elongated-1, rhomboidal-2 and oval-3). For measuring seed coat thickness, seed diameter with or without seed coat was measured using vernier calipers and difference in dimensions were recorded, categorized and coded. The analysis of variance among different accessions for seed length, seed breadth and caruncle was done using ANOVA with completely randomize design (CRD) in 5 replication of each seed lot. A two tailed Pearson Correlation Coefficient was calculated between different seed characters to understand interrelationship between them (MS Excel).

Analysis of variation revealed significant difference among all accessions for seed length, breadth and caruncle size (Fig. 1). According to Kulkarni and Ramanamurthy (1977), seed size of castor generally ranges between 7.05 mm x 5.30 mm and 20.95 mm x 13.50 mm. In the present study, seed size varied between 7.73 mm x 5.03 mm and 15.23 mm x 10.28 mm, indicating good variability

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of castor seeds in the north-eastern region (Table 1). Seed length/breadth ratio showed range of variation from 0.43 to 0.81. Based on this ratio 90 accessions were categorized into three categories as small seeded type (0.43-0.55), medium seeded type (0.56-0.68) and large seeded type (0.69-0.81). Most of the castor seeds were small (49 accessions) followed by medium sized seeds (36 accessions) and the large seeded types were not common (5 accessions). According to Kulkarni and Ramanamurthy (1977), medium and small sized seeds are the two groups to which most of the cultivated varieties of the Indian castor belong. There are multiple factors in nature which are responsible for variation in seed size, of which small seeded types may be favoured in natural selection for wider adaptability and effective dispersal capacity (Rao *et al.*, 2008). Small seeds of castor have also been recorded correlating to high oil content (Anjani and Jain, 2004; Pandey and Radhamani, 2006).

All accessions, categorized as large, medium and small types of seeds showed a well developed caruncle. Caruncle size also varied from 0.33- 2.33 mm, but in some genotypes the caruncle was very flat and could not be measured. Castor bean seeds contain caruncle which are fleshy structures and have different shapes (<http://www.hybridcastorseeds.in/blog/>). Caruncle shape also showed morphological variation i.e., flat (17 accessions), small (18 accessions), elevated (23 accessions) and irregular shape (28 accessions). Elevated or irregular shapes of caruncle were most common among the studied lot.

Table 1. Variation in seed size with different variables

Parameter	Range	LSD
Seed length (mm)	7.73 – 15.23	0.039
Seed breadth (mm)	5.03 – 10.28	0.026
Caruncle size (mm)	0.33 – 2.33	0.081
Seed breadth/length ratio	0.43 – 0.81	–

LSD at 0.5

As reported by Kulkarni and Ramanamurthy (1977) caruncle is always present in Indian varieties and helps in absorbing moisture during castor seed germination.

Enormous variability was observed in seeds of castor which showed wide range of mottling i.e. less mottled (39 acc.), medium mottled (39 accessions), maximum mottled (7 accessions) and without mottling (5 accessions) (Table 2). Seed coat colour also exhibited different colours: dark

brown was most common (62 accessions) followed by light brown (27 accessions) and creamish brown (one accession). Seed coat thickness was divided into three categories. Most of the castor seed possess thick seed coat (64 accessions) as compared to less and medium seed coat (18 accessions each). Surface of mature castor seeds showed variation from less shining to maximum shining seed coat. 41 accessions had less shining seed coat and in contrast maximum shining seed coat was

Table 2. Variation in seed morphological characters of castor seeds

S. No.	Seed morphological traits	Categories	Number of accessions
1	Mottling	Nil	5
		Less mottled	39
		Medium mottled	39
		Maximum mottled	7
2	Seed coat colour	Creamish brown	1
		Light brown	27
		Dark Brown	62
3	Seed coat thickness	Less	18
		Medium	18
		Maximum	64
4	Seed surface	Less shining	41
		Medium shining	31
		Maximum shining	18
5	Seed shape	Elongated	10
		Rhomboidal	45
		Oval	35
6	Caruncle size	Flat	17
		Small	18
		Elevated	23
		Irregular	28

observed only in 18 accessions. Study of seed structure variability is important for identification of different genotypes in any germplasm which also plays a major role in classification of different *Ricinus* genotypes (Varier *et al.*, 1999).

Seed shape showed large morphological variations. These shapes were elongated (10 accessions), rhomboidal (45 accessions) and oval (35 accessions). Rhomboidal shape was most commonly occurring among accessions and elongated shape was less common.

Correlation coefficient revealed relationship between various seed traits. Among the morphological characters, positive and negative correlations were observed (Table 3). Seed length and breadth were significantly

Table 3. Correlation between different seed characters of castor

Characters	Breadth	Length	Caruncle	Mottling	Seed coat colour	Seed coat thickness	Seed surface	Seed shape
Length	0.767**	1.000	-	-	-	-	-	-
Caruncle	0.138	0.077	1.000	-	-	-	-	-
Mottling	0.248*	0.411**	0.228*	1.000	-	-	-	-
Seed coat colour	0.152	0.113	0.111	0.272**	1.000	-	-	-
Seed coat thickness	-0.411**	-0.471**	-0.071	-0.286**	0.014	1.000	-	-
Seed surface	-0.099	0.012	0.001	0.368**	0.195	0.210*	1.000	-
Seed shape	-0.239*	-0.218*	-0.038	-0.009	0.110	0.159	-0.024	1.000
Caruncle shape	0.007	0.025	0.731**	0.137	0.070	0.030	-0.093	-0.027

**Correlation is significant at the 0.01 level; *correlation is significant at the 0.05 level.

positively correlated with each other ($P > 0.01$). Positive correlation of seed length with seed breadth and thickness was also reported by Kaushik *et al.*, (2007) in *Jatropha curcas*. Seed surface also showed positive correlation with seed mottling and seed coat thickness but seed shape showed significant negative correlation with seed length and breadth. Caruncle size and caruncle shape are highly correlated with each other. Seed coat colour also showed positive correlation with mottling.

The present study undertaken on morphological variation in castor seeds showed maximum representation of dark brown, small sized, hard seed coated, rhomboidal shaped, less mottled seeds with elevated and irregular caruncle in the north-eastern region. Previous studies conducted on the indigenous variability in castor seed morphology from western Himalayan region (Himachal Pradesh, Jammu & Kashmir) by Pandey and Radhamani (2006) and from trans-gangetic plains (Punjab and Haryana) by Duhoon *et al.* (1996) also showed higher

representation of small sized seeds mostly the wild types. This species has been prominently growing as a wild/weedy type all over India across different regions of the country. Further studies on germplasm from north-eastern region would add more information on diversity pattern in seed.

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Fig. 1. Variability in seed morphological characters of castor germplasm

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Plant Germplasm Registration Notice*

The Plant Germplasm Registration Committee of ICAR in its XXIVth meeting held on November 29, 2011 at the National Bureau of Plant Genetic Resources, New Delhi, approved the registration of following 52 germplasm lines out of 104 proposals considered. The information on registered germplasm is published with

the purpose to disseminate the information to respective breeders for utilization of these genetic stocks in their crop improvement programmes. Upon request, the developer(s)/author(s) is/are obliged to distribute the material for crop improvement programme of National Agricultural Research System.

1. WCF 12-7, WCF 12-61, WCF 12-19 and WCF 12-208 (IC0594376 - IC0594379; INGR11037-INGR11040), Wheat (*Triticum aestivum*) Germplasm with Drought Tolerance

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Morpho-physiological and agronomic characteristics of the RILS selected from the wheat cross WL711 X C306

The parent cultivars WL711 and C306 have several unique morphological and physiological differences in addition to yield and yield components such as: WL711 is a semi-dwarf, high yielding, drought susceptible, medium flowering variety with medium seedling vigour, erect growth habit and medium grains which are dull brown in colour while C306 is a tall, medium yielding, drought tolerant, late flowering variety with high seedling vigour, spreading growth habit and bold grains which are light amber in colour. Results presented here are based on the experiments conducted in the fields of Water Technology Centre, IARI, New Delhi during winter of 2007-08, 2008-09 and 2009-10. Water variable environments were created using differential irrigation.

1. WCF12-7

Morpho-agronomic Characteristics

- a.) Tall (122 ± 5 cm)
- b.) Erect in habit, lodging
- c.) Pubescent ears, ear tip fertile
- d.) Medium flowering (86 ± 3 DAS)
- e.) High yielding (grain yield= 642 ± 35 g/m²)
- f.) High grain weight (37.41 ± 0.62 g 1000⁻¹ grains), bold and amber coloured grains

- g.) Medium total grain protein content (12.48 ± 0.36 %)

- h.) Medium gluten content (26.0 ± 0.3 %). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110 012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought tolerant under water variable environments (DSI= 0.83 ± 0.14)
- b.) High relative water content under water deficit stress (RWC= 74.7 ± 4.0 %; Barrs and Weatherly, 1962)
- c.) High cellular tolerance under water deficit stress (%Injury= 44.3 ± 0.78 %; Blum and Ebercon, 1981)

2. WCF12-19

Morpho-agronomic Characteristics

- a.) Tall (122 ± 2 cm)
- b.) Erect in habit, lodging
- c.) Pubescent ears, ear tip sterile
- d.) Medium flowering (91 ± 2 DAS)
- e.) High yielding (grain yield= 579 ± 36 g/m²)
- f.) Medium grain weight (33.83 ± 1.52 g 1000⁻¹ grains), amber coloured grains

*Compiled and edited by: Anjali Kak and RK Tyagi, Division of Germplasm Conservation, National Bureau of Plant Genetic Resources, Pusa Campus, New Delhi-110 012

- g.) High total grain protein content on dry weight basis ($14.75 \pm 0.38\%$)
- h.) High gluten content ($35.30 \pm 0.4\%$). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought tolerant under water variable environments ($DSI=0.63 \pm 0.08$)
- b.) High relative water content under water deficit stress ($RWC=77.6 \pm 4.2\%$; Barrs and Weatherly, 1962)
- c.) High cellular tolerance under water deficit stress (% Injury= $43.7 \pm 0.61\%$; Blum and Ebercon, 1981)

3. WCF12-61

Morpho-agronomic Characteristics

- a.) Semi-dwarf ($103 \pm 3\text{cm}$)
- b.) Erect in habit, non-lodging
- c.) Glabrous ears, ear tip fertile
- d.) Medium flowering (91 ± 3 DAS)
- e.) High yielding (grain yield= $615 \pm 43\text{g/m}^2$)
- f.) Medium grain weight ($33.55 \pm 0.5\text{g}$ 1000⁻¹ grains), brown coloured grains
- g.) High total grain protein content on dry weight basis ($14.10 \pm 0.21\%$)
- h.) High gluten content ($31.90 \pm 0.6\%$). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought tolerant under water variable environments ($DSI=0.82 \pm 0.04$)
- d.) High relative water content under water deficit stress ($RWC=77.9 \pm 2.7\%$; Barrs and Weatherly, 1962)
- e.) High cellular tolerance under water deficit stress (%Injury= $42.1 \pm 0.78\%$; Blum and Ebercon, 1981)

4. WCF12-208

Morpho-agronomic Characteristics

- a.) Tall ($118 \pm 1\text{cm}$)

- b.) Spreading in habit, lodging
- c.) Glabrous ears, ear tip sterile
- d.) Medium flowering (90 ± 2 DAS)
- e.) High yielding (grain yield= $618 \pm 36\text{g/m}^2$)
- f.) High grain weight ($36.12 \pm 0.44\text{g}$ 1000⁻¹ grains), bold and brown coloured grains
- g.) High total grain protein content on dry weight basis ($14.26 \pm 0.53\%$)
- h.) High gluten content ($36.60 \pm 0.5\%$). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought tolerant under water variable environments ($DSI=0.67 \pm 0.14$)
- b.) High relative water content under water deficit stress ($RWC=76.9 \pm 3.5\%$; Barrs and Weatherly, 1962)
- c.) High cellular tolerance under water deficit stress (%Injury= $31.1 \pm 0.8\%$; Blum and Ebercon, 1981)

Morpho-physiological and agronomic characteristics of the parents of the selected RILs from the wheat cross WL711 x C306

Parent P1- WL711

Morpho-agronomic Characteristics

- a.) Semi-dwarf ($102 \pm 2\text{cm}$)
- b.) Erect in habit, non-lodging
- c.) Glabrous ears, ear tip fertile
- d.) Medium flowering (88 ± 1 DAS)
- e.) High yielding (grain yield= $674 \pm 57\text{g/m}^2$)
- f.) Medium grain weight ($33.10 \pm 0.62\text{g}$ 1000⁻¹ grains), brown coloured grains
- g.) Medium total grain protein content ($11.10 \pm 0.07\%$)
- h.) Medium gluten content ($25.40 \pm 0.4\%$). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought susceptible under water variable environments ($DSI=1.39 \pm 0.15$)

- b.) Low relative water content under water deficit stress (RWC=66.2 $\pm$ 1.6%; Barrs and Weatherly, 1962)
- c.) Low cellular tolerance under water deficit stress (%Injury=56.9 $\pm$ 0.98%; Blum and Ebercon, 1981)

Parent P2-C306

Morpho-agronomic Characteristics

- a.) Tall (128 $\pm$ 1cm)
- b.) Spreading in habit, lodging
- c.) Pubescent ears, ear tip sterile
- d.) Late flowering (96 $\pm$ 1 DAS)
- e.) Medium yielding (grain yield=436 $\pm$ 23g/m²)
- f.) High grain weight (37.52 $\pm$ 0.31g 1000⁻¹ grains), bold and amber coloured grains
- g.) High total grain protein content (13.46 $\pm$ 0.19%)
- d.) High gluten content (33.40 $\pm$ 0.2%). Grain quality parameters analyzed in grain quality lab, IARI, New Delhi-110012

Associated Characters and Cultivated Practices

- a.) Drought tolerance is based on drought susceptibility index (Fischer and Maurer, 1978). Drought tolerant under water variable environments (DSI=0.71 $\pm$ 0.17)
- b.) High relative water content under water deficit stress (RWC=76.1 $\pm$ 1.4%; Barrs and Weatherly, 1962)
- c.) High cellular tolerance under water deficit stress (%Injury=45.2 $\pm$ 0.74%; Blum and Ebercon, 1981)

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2. DMR-PFSR-1 and DMR-PFSR-9 (IC0590094-IC0590095; INGR11041-INGR11042), a Maize (*Zea mays*) Germplasm, Resistant to Post Flowering Stalk Rots caused by *Macrophomina phaseolina* and *Fusarium moniliforme*, with Stiff, Strong and Stay Green Character of Stalk

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1. DMR - PFSR-1: (SW 93D-313-23-Pop-49-S4 (Z Path) -1-3-1-1-1-2-1-2-1) is an inbred line, source of resistance to post flowering stalk rots of maize caused by *Macrophomina phaseolina* and *Fusarium moniliforme* (Rakshit *et al*, 2011). The primary source of this material was received from CIMMYT. The inbred was developed at DMR, New Delhi, for desirable characters following pedigree breeding methodology. The germplasm /parental material was received from CIMMYT under collaborative programme of evaluation inbred lines against PFSR during 2001-2004. The multi-location evaluation of the genotype was done in hot spot locations at Hyderabad, Udaipur, Delhi and Ludhiana against PFSR from 2006 – 2010 under artificial epiphytotic condition. Maintenance

and multiplication of inbred line was done at Directorate of Maize Research, New Delhi and winter nursery at Hyderabad.

Morpho-agronomic Characteristics

PFSR-R1 exhibited strong robust stiff and green stem with purple brace root, sparse spikelet. Plant length is short, conical ear with yellow, round, flint grain, desirable plant type and good agronomic traits like optimum ear placement, stiff stalk, good pollen shed and exhibited consistently resistant reaction against PFSR during four years evaluation. The disease reaction recorded from 1.0 to 4.2 on 1-9 rating scale (1 is highly resistant, 9 is highly susceptible) across the location.

Associated Characters and Cultivated Practices

Stem is green, stiff robust. Resistant against PFSR, is an economically important disease causes reduction of 18.7% in cob weight and 11.2% in 1000-grain weight in infected plants (Cook, 1978). The losses due to this disease in India have also been calculated to range from 10 to 42% (Payak and Sharma, 1978; Desai *et al.*, 1991; Harlapur *et al.*, 2002).

2. **DMR – PFSR – 9:** (JCY3-7-1-2-1(Z Path) -b-2-1-3-1) is inbred line, source of resistance to post flowering stalk rots of maize caused by *Macrophomina phaseolina* and *Fusarium moniliforme* (Rakshit *et al.*, 2011). The primary source of this material is JCY series from PAU, Ludhiana. The inbred was developed at DMR New Delhi, for desirable character following pedigree breeding methodology. The germplasm /parental material was received from PAU, Ludhiana under collaborative programme of evaluation inbred lines against PFSR during 2001-2004. The multi-location evaluation of the genotype was done in hot spot locations at Hyderabad, Udaipur, Delhi and Ludhiana against PFSR from 2006 – 2010 under artificial epiphytotic condition. Maintenance and multiplication of inbred line was done at Directorate of Maize Research, New Delhi and winter nursery at Hyderabad.

Morpho-agronomic Characteristics

Plant exhibited stay-green character, stiff, strong stem, purple brace root, with light purple silk, sparse spikelets, purple silk, plant length – long, ear conico- cylindrical; Grains are dent, yellow round in shape with desirable plant type and good agronomic traits like optimum ear

placement, stiff stalk, good pollen shed. The inbred was consistently resistant against PFSR. The disease reaction recorded from 1.0 to 4.2 on 1-9 rating scale (1 is highly resistant, 9 is highly susceptible) across the location during 4 years of testing.

Associated Characters and Cultivated Practices

Stem is green, stiff robust. Resistant against PFSR which is an economically important disease causes reduction of 18.7% in cob weight and 11.2% in 1000-grain weight in infected plants (Cook, 1978). The losses due to this disease in India have also been calculated to range from 10 to 42% (Payak and Sharma, 1978; Desai *et al.*, 1991; Harlapur *et al.*, 2002).

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3. IPM 205-7 (IC0589309-IC0589310; INGR11043-INGR11044), a Mung bean (*Vigna radiata* (L.) Wilczek) Germplasm with Super Early Maturity

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IPM 205-7: Mungbean [*Vigna radiata* (L.) Wilczek] is an important short duration grain legume having wider adaptability and low input requirements. It is widely grown in the sub-tropical countries of the South and Southeast Asia, Australia, West Indies, South and North America and Tropical and Subtropical Africa. India is the largest producer of mungbean and alone accounts for 65% of the world acreage and 54% of the world

production. In India, it is grown in *kharif* (monsoon), *rabi* (winter) and spring/summer seasons in different agro-ecological regions. While comparatively longer duration genotypes (65-75 days maturity) are suitable for cultivation in the *kharif* season, short duration genotypes (<60 days maturity) are desirable for spring/summer seasons. Because of scarcity of irrigation water and intense heat wave during the months of April and May

coinciding with the reproductive phase of this crop in spring/summer sown crop, short duration genotypes are urgently required which can mature in about 50 days. Such genotypes when sown after the harvest of *rabi* crops can complete their life cycle well before the onset of intense heat wave escaping extreme temperatures during pod filling and maturity stage, giving good harvest. Such genotypes can also fit well in different crop rotations and cropping systems. Keeping this in view, an extra early maturing genotype, IPM 205-7 (IC0589309), was developed at Indian Institute of Pulses Research, Kanpur. This genotype has been derived from a cross 'IPM 02-1 x EC 398889' following pedigree method of breeding. It has short-statured and erect plants with green, ovate and entire leaves and a green stem with purple splashes. The flowers are of light yellow color while the pod habit is intermediate. Pods are short, straight and black on maturity while the seeds are green and shining. IPM 205-7 has synchronous maturity and it is resistant to Mungbean Yellow Mosaic Virus. This genotype matured 11-19 days earlier than the different check varieties when sown under Kanpur conditions (20° 27' N latitude, 80° 14' E longitude, and 152.4 meter above the mean sea level). While IPM 205-7 matured in 46-48 days, the check varieties matured in 55-67 days. This genotype can be used as a donor for transfer of earliness in agronomically superior backgrounds.

IPM 409-4: Mungbean [*Vigna radiata* (L.) Wilczek] is economically one of the most important pulse crops of the *Vigna* group and it is cultivated since prehistoric period in India. It is grown throughout Asia, Australia, West Indies, South and North America, Tropical and Subtropical Africa. In India, it is cultivated in different

seasons including spring, summer, *rabi* and *kharif*. Development of early maturing genotypes is one of the prime breeding objectives in mungbean improvement programme because such genotypes can fit well in different crop rotations and multiple cropping systems. During spring/summer season, their cultivation after the harvest of wheat in North and Central India may save at least one to two irrigations and one pesticide spray leading to considerable savings. Besides this, it may also help the crop escape from terminal heat wave, which can otherwise lead to premature flower drop and significant yield loss due to lesser pod set. Keeping this in view, an extra early maturing genotype, IPM 409-4 (IC0589310), has been developed at Indian Institute of Pulses Research, Kanpur. This genotype derived from a cross 'PDM 288 x IPM 03-1', following pedigree method of breeding, has short-statured, erect and determinate plant type. The leaves are dark green, ovate and medium-sized with greenish purple veins while the flowers are light yellow in colour. The pods are present above the canopy and are short, black and curved while the seeds are green, shiny and oval in shape. When sown under Kanpur conditions (20° 27' N latitude, 80° 14' E longitude, and 152.4 meter above the mean sea level), this genotype matured 10-19 days earlier than the check varieties. While IPM 409-4 matured in 46-48 days, the check matured in 55-67 days. This genotype has synchronous maturity and is also resistant to Mungbean Yellow Mosaic Virus. Keeping in view its higher yield, resistance to MYMV and extra early maturity, it can be evaluated for possible release besides using it as donor in hybridization programme for development of early maturing and high yielding varieties of mungbean.

4. VBG-09-012 (INGR11045), an Urd Bean (*Vigna mungo*) Germplasm with Multi-Pod Formation at Base of Peduncle, Leaf Axils and Base of Clusters

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It is derived from *Vigna mungo* ADT3 x *V. mungo* var. *silvestris*. It is unique plant type with multipod formation at base of peduncle, leaf axils and base of clusters.

5. VBG-04-014 (IC0589272; INGR11046), an Urd Bean (*Vigna mungo*) Germplasm with Unique Plant Type

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It is derived from *Vigna mungo* (VBN1) x *V. mungo* var. *silvestris* L. After flowering, pods comes above the plants.

6. Phule G96006 (IC0589349; INGR11047), a Chickpea (*Cicer arietinum*) Germplasm with Drought Tolerance

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Good yield performance (1,793 kg/ha) under rainfed conditions. Highest Proline content (2,160 mg/100 gm) which imparts high drought tolerance efficiency (DTE 71.36%). Stay green nature, the leaves remain green during grain filling stage even under moisture stress and good yield. Medium size grain with test weight of 18.6 g/100 seed. Resistant to *Fusarium wilt* (6.50%). Protein content (23.9%).

7. BPR-349-9 (IC0589778; INGR11048), an Indian Mustard (*Brassica juncea*) Germplasm with Thermo- Tolerance at Juvenile Stage

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The experiments were conducted at five locations (Hisar, Kanpur, Ludhiana, Bharatpur and SK Nagar) during 2009-10 and 2010-11 with 44 and 43 genotypes, respectively for screening against high temperature stress during juvenile stage by growing in plastic trays containing 5kg soil, which was thoroughly mixed with known volume of de-ionized water for germination. Sowing was in rows, with replication for genotype in a random order to avoid minor microclimatic variation inside the tray and the seed germinator. Seedlings were initially allowed to grow at 25±1°C for days at 70-80% RH and then exposed to 45±1°C at 30% RH for four hours daily with field capacity of 90-95%. The high temperature treatment was given up to three days. The tolerance was characterized on the basis of percent seedling mortality. Lower the mortality higher the tolerance. The proposed strain showed <20%

seedling mortality at out of 10 locations during 2009-10 and 2010-11. It also showed lower seedling mortality then the registered check RH-8814 at 6 out of 10 locations during 2009-10 and 2010-11 (Table 1).

Table 1. Seedling mortality (%) under high temperature at juvenile stage in BPR-349-9 and checks

Centre	2009-10			2010-11	
	BPR-349-9	RH 8814 (Registered)	NRCDR-02	BPR-349-9	BPR-543-2 (Registered)
Hisar	20.0	18.5	18.2	4.9	-
Kanpur	15.8	33.9	26.0	28.4	21.8
Ludhiana	37.3	30.2	27.2	18.4	22.0
Bharatpur	17.6	32.4	17.5	10.5	17.0
SK Nagar	12.7	12.3	10.7	18.8	23.4
Mean	20.7	25.5	19.9	16.0	21.1

8. MCA 1 (IC0589777 and IC0590093; INGR11049), a Karan rai (*Brassica carinata*) Germplasm with Cytoplasmic Male Sterility

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Ethiopian mustard (*Brassica carinata* A. Braun) is among the oldest oil crops cultivated in Ethiopia. Owing to its drought and heat tolerance, the crop is now being considered as an alternative to *B. napus* and *B. juncea* in dryer areas of Canada and as a potential oil crop in India. Heterosis has been reported for seed yield as well as for early maturity in this crop. A cytoplasmic male sterility system called moricandia cytoplasmic male sterility (CMS) was developed in *B. juncea* through protoplast fusion of *Moricandia arvensis* and *B. juncea* (Prakash *et al.*, 1998). This CMS system has been used to develop the hybrids in Indian mustard (*B. juncea*) with the release of first public bred hybrid NRCHB 506. Looking the possibility of developing hybrid cultivars in Ethiopian mustard, efforts were initiated to transfer moricandia cytoplasm from a male sterile line of *B. juncea* into *B. carinata*.

The F₁ progeny of the cross MJA1 (CMS line of *B. juncea*) and KR9 (fertile line of *B. carinata*) showed an intermediate phenotype between *B. juncea* and *B. carinata*. The flowers were male sterile with well developed nectaries, style, stigma and gynoecium. Continuous backcrosses with *B. carinata* genotype (recurrent parent used as pollen parent) resulted in genome substitution from *B. juncea* to *B. carinata*. Visual observation on presence/absence of pollen grains revealed the absence of pollen grains in anthers of MCA 1 which was confirmed by the no seed set in selfed buds. On the other hand, pollen grains were present and found viable as depicted by stained pollens viewed under light microscope. Further the seed set under selfed buds confirmed the presence and viability of pollen grains of MCB 1. It confirmed the development of stable male sterile line of *Brassica carinata* after eight backcrosses. This line was named as MCA 1 having moricandia cytoplasm that possesses male sterility and its counterpart fertile line was named as MCB 1. t-statistics used for making comparison between developed cytoplasmic male sterile line MCA 1 and its maintainer line MCB 1, revealed that both A and B lines were statistically at par

for days to flower initiation, days to maturity, number of primary branches, length of main raceme, siliquae on main raceme, siliqua length, plant height, petal length, petal width and style length (Table 1). Numerically longer main raceme and more number of siliquae are expected in male sterile line than that of maintainer line due to non production of pollen grains. The significant difference was observed for stamen length being low in male sterile line than its counterpart maintainer line. Underdeveloped stamens are the outcome of interaction between alien cytoplasm from *Moricandia arvensis* and nuclear genome of *B. carinata* resulting into male sterility. However, female fertility has not been affected as depicted by equal number of seeds per siliqua (which are through outcrossing in male sterile line) and siliqua length of MCA 1 and MCB 1. The developed CMS line MCA 1 will pave the way for hybrid development in *B. carinata*.

Table 1. Characterization of MCA 1 (CMS line) and its maintainer (B) line for agronomic and floral traits

Trait	MCA 1	MCB 1	t value	p value
Days to flower initiation	57	58		
Days to maturity	138	140		
Primary branch	10.2	9.6	0.34	0.37
Plant height (cm)	195	193	-0.53	0.31
Length of main raceme (cm)	90	78	1.67	0.06
Siliquae on main raceme	54.8	45.2	1.75	0.06
Siliqua length (cm)	4.9	5.1	-0.71	0.25
Seeds per siliqua	14.2	14.0	0.34	0.37
Petal length	1.42	1.38	1.63	0.08
Petal width	0.62	0.64	-1	0.18
Stamen length	0.64*	0.76	-6	0.001
Style length	0.76	0.72	1	0.18

* Significantly different from respective maintainer line at p= 0.05

Reference

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9. E 1-3 (IC0590089; INGR11050), a Potato (*Solanum tuberosum* (+) *S. etuberosum*) Tetraploid Somatic Male Fertile Hybrid Carrying Resistance to Potato Virus Introgressed from *S. etuberosum*

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Interspecific potato somatic hybrids between the cultivated potato *Solanum tuberosum* L., dihaploid C-13 and wild species *S. etuberosum* Lindl. were produced by protoplasts electro-fusion. Since, the wild species is not crossable with cultivated potato therefore somatic fusion was attempted to transfer Potato virus Y (PVY) resistance from the wild species into the cultivated potato as previously investigated by Thieme *et al.* (2004). Post-fusion products were cultured in VKM medium followed by regeneration of calli in MS₁₃ K medium at 20°C under a 16-h photoperiod, and regenerants were multiplied on MS medium. Twenty-one somatic hybrids were confirmed by RAPD, SSR and cytoplasm (chloroplast/mitochondria) type analysis possessing species-specific diagnostic bands of corresponding parents. Tetraploid nature of the somatic hybrid was determined through flow cytometry analysis as described by Arumuganathan and Earle (1991). Somatic hybrid showed intermediate phenotypes (plant, leaves and floral morphology) to their parents

in glasshouse grown plants and male fertile following DUS descriptors by Gopal *et al.* (2008). ELISA assay of somatic hybrid E 1-3 after artificial inoculation of PVY infection reveals high resistance (Anonymous 2007). The tetraploid somatic hybrid E 1-3 can be exploited in the future potato breeding programmes.

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10. P-7 (IC0590090; INGR11051), a Potato (*Solanum tuberosum* (+) *S. pinnatisectum* Tetraploid, Somatic Male Fertile Hybrid Carrying Resistance to Potato Late Blight Introgressed from *S. pinnatisectum*

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Interspecific somatic hybrids between the dihaploid of cultivated potato (*Solanum tuberosum*) and the diploid wild species *S. pinnatisectum* Dun. were produced via protoplast fusion. Since, the wild species is not crossable with cultivated potato because of genetic barriers therefore somatic hybridization was attempted to transfer late blight resistance from wild species into cultivated (Thieme *et al.* 2010; Polzerová *et al.* 2011). Protoplast isolation, electrofusion, culture of post-fusion products and regeneration of calli/shoots were undertaken following

optimized protocols. Regenerants were characterized for hybridity, ploidy and resistance to *Phytophthora infestans* (Mont.) de Bery, causal fungal pathogen of late blight disease. From a total of 126 regenerated macrocalli, 12 somatic hybrids were confirmed by possessing species specific diagnostic bands of their corresponding parents as revealed by RAPD, SSRs and cytoplasmic-DNA analyses. Tetraploid status of the somatic hybrid was determined using flow cytometry analysis as described by Arumuganathan and Earle (1991). Intermediate

phenotypes for leaf, flower, and tuber characteristics and high male fertility were observed in field-grown hybrid plants following DUS descriptors discussed by Gopal *et al.* (2008). Hybrids were highly resistant to foliage late blight based on field assessment for two seasons using methods of Kaushik *et al.* (2007). In contrast, moderate level of resistance to foliage blight of the somatic hybrid P-7 was observed in hybrids based on the detached leaf assay under laboratory conditions. Overall, somatic hybrid P-7 with moderate levels of resistance to foliage blight was identified, and these will be useful for in situ hybridization in potato breeding efforts.

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11. PS-52 (IC0590839; INGR11052), a Shisham (*Dalbergia sissoo* Roxb.) Germplasm with Straight Bole and High Height and Diameter

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Shisham (*Dalbergia sissoo* Roxb.) is one of the most important multipurpose plantation tree of northern India. It grows naturally and it is also planted on alluvial soils. It is cultivated more extensively than any other species except teak. In its natural habitat the tree grows rigorously in the new alluviums formed deposits of sand, boulders and gravels in the sub-Himalayan river beds and river banks. It prefers well drained sandy loam soils with adequate moisture supply. Shisham is a deciduous tree and coppices vigorously and produces profuse root suckers. The roots including taproots and lateral roots are covered with nitrogen fixing bacteria *Rhizobium*.

PS-52 has been selected as plus tree from natural population from Gonda district of Uttar Pradesh. It has shown superiority for straight bole and high height (13.70 m) and diameter (15.40 cm) growth at the age of 10

years and registered for these parameters, besides shown resistance to mortality (Tewari and Kaushal, 2006). PS-52 plants may be propagated through clonal propagation (sprout cuttings and root suckers). Nicked root plants may be planted in January - February while poly-packed plants may be planted in July – August. Germplasm PS-52 has been evaluated in agroforestry coordinated trials for over ten years and found consistent for its traits expression (AICRP on Agroforestry, 2011).

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12. PP-5 (IC0590840; INGR11053), a Poplar (*Populus deltoids* Bartr.) Germplasm with High Height and Diameter

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Poplar (*Populus deltoids* Bartr.) is one of the most important agroforestry tree species in the states of northern India. Owing to fast growth rate and ability to grow well over large areas in the alluvial plains, poplar contributes greatly towards production of wood for industrialization and other commercial purposes, besides maintaining ecological balance and also as an excellent source of income to the farmers. It has assumed the status of being commercially most useful tree species of the genus *Populus* in India. The genus *Populus* belongs to family Salicaceae and its wood is used in manufacture of packing cases, hardboard, veneers, sport goods, pulpwood and poles. Besides this, poplar wood is also used in matchbox industry for manufacturing match splints and boxes.

PP-5, a mutant has been developed through mutation breeding using parental line L-89-4. Germplasm PP-5 has shown its superiority and registered for higher height growth (22.3%) and diameter growth (24.27%)

than the check G 48. It has also shown resistance to stem borer, clear bole length and winter bud shape and colour in comparison to check (Chaudhary and Tewari, 2006). Large number of uniform plants can be produced through clonal propagation (stem cuttings). Germplasm PP-5 has been evaluated in agroforestry coordinated trials for about ten years and found consistent for its traits expression and shown its stability (AICRP on Agroforestry, 2006). PP-5 has high commercial value and may be planted under agroforestry system (as inter-crop and on field boundary) in whole of northern states viz. Punjab, Haryana, western Uttar Pradesh and Uttarakhand between January 15 to February 15.

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13. NRCGCS-15 (IC0589174; INGR11054) Multiple Disease Resistant Spanish Bunch Groundnut Genotype

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NRCGCS-15 (IC0589174; INGR11054) was selected from advanced generation of cross (CT7-1 x SBXI) x *A. pusilla*. The genotype was developed by pedigree selection from interspecific progenies developed at the Cytogenetics Section, Directorate of Groundnut Research, Junagadh, Gujarat. The plant has erect growth habit, produces 50% flowering in 37 days after sowing (DAS) and matures in 118 DAS during rainy season. The genotype produces an average pod yield of 97.66 g per square metre with 67% shelling out turn. Pods are slightly constricted with prominent beak and deep reticulation; mostly two seeded with rose colour kernels. Kernels are

medium in size with hundred kernel mass of 44.6 g and contain 51.5% oil. The severity of foliar diseases in the genotype under field conditions are 2.8 for rust, 5.0 for early leaf spot and 5.0 for late leaf spot on a modified 9-point scale, and an incidence of 6.9% for peanut bud necrosis disease (PBND) and 5.2% for stem rot with almost immunity to *Alternaria* leaf blight disease has been recorded. The genotype has been identified as donor for multiple disease resistance in groundnut for peanut bud necrosis diseases, stem rot, late leaf spot, rust and *Alternaria* leaf blight.

14. NRCGCS-74 (INGR11055) a Multiple Disease Resistant Spanish Bunch Groundnut Genotype

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NRCGCS-74 (INGR11055) was selected from advanced generation of cross (CT7-1 x SBXI) x *A. pusilla*. The genotype was developed by pedigree selection from interspecific progenies developed at the Cytogenetics Section, Directorate of Groundnut Research, Junagadh, Gujarat. The plant has erect growth habit, produces 50% flowering in 36 days after sowing (DAS) and matures in 121 DAS during rainy season. The genotype produces an average pod yield of 81.16 g per square metre with 72% shelling out turn. Pods are slightly constricted with moderate beak and deep reticulation; mostly two seeded

with rose colour kernels. Kernels are medium in size with hundred kernel mass of 37.8 g and contain 45.5% oil. The severity of foliar diseases in the genotype under field conditions are 2.0 for rust, 4.0 for early leaf spot and 4.0 for late leaf spot on a modified 9-point scale, and an incidence of 16% for peanut bud necrosis disease (PBND) and 12.7% for stem rot with almost immunity to *Alternaria* leaf blight disease has been recorded. The genotype has been identified as donor for multiple disease resistance in groundnut for peanut bud necrosis diseases, stem rot, late leaf spot, rust and *Alternaria* leaf blight.

15. NRCGCS-186 (INGR11056) a Multiple Disease Resistant Virginia Runner Groundnut Genotype

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NRCGCS-186 was selected from advanced generation of cross (C-364 x PBDR 25) x *A. oteroi*. The genotype was developed by pedigree selection from inter-specific progenies developed at the Cytogenetics Section, Directorate of Groundnut Research, Junagadh, Gujarat. The plant has Virginia runner growth habit, produces 50% flowering in 32 days after sowing (DAS) and matures in 123 DAS during rainy season. The genotype produces an average pod yield of 46.83 g per square metre with 65% shelling out turn. Pods are slightly constricted with moderate beak and reticulation; mostly two seeded with

rose colour kernels. Kernels are medium in size with hundred kernel mass of 32.5 g and contain 50% oil. The severity of foliar diseases in the genotype under field conditions are 3.0 for rust, 4.0 for early leaf spot and 4.0 for late leaf spot on a modified 9-point scale, and an incidence of 21.1% for peanut bud necrosis disease (PBND) and 12.8% for stem rot with almost immunity to *Alternaria* leaf blight disease has been recorded. The genotype has been identified as donor for multiple disease resistance in groundnut for peanut bud necrosis diseases, stem rot, late leaf spot, rust and *Alternaria* leaf blight.

16. NRCGCS-196 (IC0589180; INGR11057) a Multiple Disease Resistant Virginia Bunch Groundnut Genotype

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NRCGCS-196 was selected from advanced generation of cross (GAUG-10 X CGC-4018) *A. correntina*. The genotype was developed by pedigree selection from inter-specific progenies developed at the Cytogenetics Section, Directorate of Groundnut Research, Junagadh, Gujarat. The plant has Virginia bunch growth habit, produces 50% flowering in 40 days after sowing (DAS) and matures in 120 DAS during rainy season. The genotype produces an average pod yield of 160.9 g per square metre with 74% shelling out turn. Pods are prominently constricted with prominent beak and deep reticulation; mostly two seeded with rose colour kernels. Kernels are

medium in size with hundred kernel mass of 32.5 g and contain 50.7% oil. The severity of foliar diseases in the genotype under field conditions are 3.0 for rust, 3.0 for early leaf spot and 3.0 for late leaf spot on a modified 9-point scale, and an incidence of 9.6% for peanut bud necrosis disease (PBND) and 8.6% for stem rot with almost immunity to *Alternaria* leaf blight disease has been recorded. The genotype has been identified as donor for multiple disease resistance in groundnut for peanut bud necrosis diseases, stem rot, late leaf spot, rust and *Alternaria* leaf blight.

17. TGM-112 (IC0585932 INGR11058), a Groundnut (*Arachis hypogaea*) Germplasm with White to Light Orange Flower Colour Mutation

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Groundnut has been treated extensively to different mutagens for induction of genetic variability. Several studies showed number of mutations affecting leaf size, shape and colour, plant height, plant habit, flower colour, pod and seed traits. Groundnut has five distinct flower colours (white, yellow, orange, burnt orange and amber). Of these, yellow and orange flowers are most common. Seeds of groundnut cultivar tag 24 (Patil *et al.*, 1995) were treated with 150, 250 and 350 Gy gamma rays during rainy season 2000 and the M₂ plants were grown at the Bhabha Atomic Research Centre, Mumbai. Groundnut mutant, TGM 112, was isolated with white to light orange flower from the 250 Gy treatment with a frequency of 0.02% based on M₂ plant population (Badigannavar, 2007).

In the induced groundnut mutant TGM 112, the colour of petals, namely, standard, wing and keel ranged from different grades of white to light-orange (hence it was referred as light-orange) as compared to orange petals

found in the parent variety, TAG 24. Further, the central crescent area of the standard was also light-orange in mutant, while it was orange in the parent. At any given time, the mutant had either all the flowers in white colour or a combination of white and light-orange flowers. The mutant was bred true in the M₃ and its true breeding behaviour was confirmed up to the M₈ generation.

Associated Characters and Cultivated Practices:

In the crosses between the parent variety and mutant, all the F₁ plants had orange flowers indicating dominance of orange flower over light-orange. The F₂ plant population segregated to the 3:1 ratio for orange: light-orange flowered plants. Reciprocal crosses also did not differ from the expected 3:1 ratio, indicating absence of maternal effect for this trait. The F₃ progenies were classified on the basis of plants with orange and light-orange flowers with a good fit to the ratio of 1 (all plants with orange flowers) : 2 (3 orange: 1 light-orange) : 1 (all plants

with light-orange flowers). Thus, both phenotypic and genotypic segregation in F₂ and F₃ generations confirmed that the light-orange flower colour was due to a single recessive gene. Mutant had Spanish bunch growth and branching habit with sequential flower pattern. It matured in 100 days like its parent. The mutant trait is a very good genetic marker for an easy identification.

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18. Yellow Stem Mutant Castor (IC0587750; INGR11059), a Castor (*Ricinus communis* L.) Germplasm with Unique Colour and Single Bloom. Facilitating Development of Short Duration Varieties with Synchronous Maturity of Single Harvest

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The castor mutant is characterized by the yellow stem colour with single bloom. While the young seedlings have expressed very light green stem colour which later changing to yellow, it was observed that all other parts of the plants viz., leaves, petiole, inflorescence, peduncle and capsules also gradually turning from light green to yellow. This yellow stem colour mutant of castor is a spontaneously occurred mutant isolated from the breeding line DCS 99 during *kharif* 2009 at the Narkhuda farm of the Directorate of Oilseeds Research (DOR), Hyderabad.

The selfed progenies of the yellow stem mutant plant have been raised during *rabi* 2009-10 and found to be true breeding. Similarly, the selfed progenies of the progeny plants of 2009-10 have been evaluated during

immediate *kharif* 2010 and subsequently in advanced generation during *rabi* 2010-11, *kharif* 2011 and *rabi* 2011-12 through selfing. The progenies have not shown any deviation in all morphological traits including the yellow stem colour, in five successive generations and hence the mutant is stable in expressing the trait of interest *i.e.*, yellow stem colour. This unique mutant has not been discovered or reported earlier in castor. This mutant will be of immense use not only in understanding several biochemical and physiological mechanisms related to growth and development of castor plant but also in unraveling the evolutionary aspects of different coloured capsule variants in castor. Further, the yellow stem mutant castor can be used as a garden plant with high ornamental value.

19. 96-508-2-90 (IC0588696; INGR11060), a Safflower (*Carthamus tinctorius* L.) Germplasm with Drought Tolerance and Resistance to *Fusarium* wilt

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The safflower breeding line 96-508-2-90 is developed through multiple crosses at the Directorate of Oilseeds Research, Hyderabad. It is a drought and *Fusarium* wilt resistant high yielding breeding line. It was found to

be immune to all *Fusarium* isolates collected across the safflower growing areas in the country when tested in wilt sick pots and plot. It could yield higher than the highest yielding national check variety A1 over years.

20. CNH 301 (IC0587405; INGR11061), a Cotton (*Gossypium hirsutum*) Germplasm with Drought Tolerant Nature and Yield Stability

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The advance culture was developed through pedigree breeding from a single cross between SRT 1 x 1301 DD. F1 and F2's were evaluated under both rainfed and irrigated condition followed by selection under rainfed condition. The single plant selections were further tested under both the conditions and under simulated drought in pots. Drought susceptibility index was estimated and progenies were selected with low susceptibility index.

CNH 301 is one of the progeny which recorded high seed cotton yield and maintained high leaf water potential through higher stomatal resistance and lower rate of transcription, high chlorophyll stability and low % fall in pH. It possesses small thick leaves, compact plant body with yellow flowers. All these character are associated with drought tolerance.

21. IGFRI-CcSx -08/1 (IC0590889; INGR11062), an Anjan grass (*Cenchrus ciliaris* L.) Germplasm with a Rare Obligate Sexual Plant

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Buffel grass (*Cenchrus ciliaris* L.), also known as Anjan grass, is an important forage grass grown throughout the semi-arid tropics. It reproduces predominantly by aposporous apomixis (Yadav *et al.*, 2011). Apomixis provides a means of clonal propagation through seeds by producing progenies which are genetically identical to the mother plant. Absence of sexual reproduction in *C. ciliaris* has severely limited the possibilities of genetic improvement of this species. Clonal mode of reproduction further complicates molecular studies for apomixis. Generally, apomixis is dominant over sexuality due to which occurrence of obligate sexual plants in natural population becomes rare, and over a period of time apomictic individuals outnumber sexual ones. Since *C. ciliaris* is protogynous, open pollination leads to fertilization by neighbouring apomictic plants that gives rise to either facultative or obligate apomictic types. On screening the germplasm collection of *C. ciliaris* at IGFRI, a natural variant of Indian accession was identified to be exclusively sexually reproducing as examined by pistil-clearing technique and embryo sac analysis (Young *et al.*, 1979). In contrast to the source plant being an apomict, the variant was found to be obligate sexual and self-incompatible.

Embryo sac analysis in two constitutive years confirmed that the plant bears eight-nucleated sexual embryo sacs. The sexual plant was observed to be very short in stature with distinct morphology. The plant bears awnless panicles of smaller size with a fewer florets. Leaves are comparatively thick, smaller in size and placed at shorter internodes. The plant was characterized as protogynous, self-incompatible with poor seed setting on open pollination (Kumar *et al.*, 2010).

Flow cytometric analysis confirmed it to be a tetraploid ($2n=4x=36$). Being perennial in nature, it is maintained by vegetative propagation using root-slips. Under natural conditions the plant shows very poor growth and survival. In pots with intensive care the plant grows well and flowers 3-4 times in a year. Due to self-incompatibility the plant does not produce seeds in isolation or on selfing, however seeds were obtained on open pollination. As per our knowledge, this is the only obligate sexual *Cenchrus ciliaris* plant available in the country. The plant is very useful for genetic improvement/studies of this species by hybridization, studying phylogenetic relationship, genome mapping and identification of gene(s) for apomixis.

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23. TV-3, TV-4, TV-5, TV-6, TV-7, TV-8, TV-9, TV-10, TV-11, TV-12, TV-13, TV-14, TV-15, TV-16, TV-18, TV-21, TV-22, TV-23, TV-24, TV-25, TV-26, TV-27, TV-28, TV-29, TV-30 and TV-31 (IC0587385-IC0587404 and IC0587772-IC0587777; INGR11063-INGR11088), Tea (*Camellia sinensis*) Germplasm

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TV-3: High Assam flavour tea. Leaf base round, leaf apex acute, small leaves.

TV-4: Temperature tolerant and resistance to tea thrips. Leaf base highly acute, venation not prominent, large leaves.

TV-5: Assam flavour with different genetic background, resistance to tea jassids. Leaves highly ovate, leaf blade rough, venation not prominent.

TV-6: Mild liquor with high flavour Assam variety, resistance to red rust. Bullation very high, leaf apex down turn, leaf base round.

TV-7: Darjeeling flavour, multiple stem, leaves very small, venation not prominent.

TV-8: Large leaf Assam variety with less fibre. Resistant to Red Spider mite. Leaf apex highly acute, leaf base highly acute, leaves concave type.

TV-9: Tolerant to water logging. Early flusher. Leaves blackish green colour, highly lanceolate, serration very high.

TV-10: Assam flavour and bright liquor with different genetic background. High leaf chlorophyll and resistance to Pink mite infestation. Leaves ovate, leaf apex mucronate type, serration very low.

TV-11: Assam variety with high flavour and bright liquor, Resistance to thorny stem blight infection. Leaves lanceolate – ovate, leaf blade curved upward, venation not prominent.

TV-12: High rooting performance of cuttings. Bright liquor. Leaves boat shaped, serration very high, leaves hard.

TV 13: Large leaf Assam flavour variety with different genetic background; Soft shoot and less fibre. Leaf base acute, leaf blade curved downward, pubescence dense.

TV 14: High leaf water potential, bright infusion. leaf apex bluntly acuminate, pubescence sparse, serration very low.

TV 15: Assam hybrid, high flavour with different genetic background. Resistant to black root rot. Leaf shape ovate, bullation not prominent and pubescence dense.

TV 16: Radiation tolerant and Sun scorch resistance. Leaf blade expanded (leathery), high serration and pubescence is sparse.

TV 18: Minty flavor. Dark pink pigmentation during autumn at petiole region, leaf blade lanceolate.

TV 21: Quality clone with unique Assam flavour, light leaf Assam variety. Leaf apex acuminate, leaf blade expanded (large), bullation absent.

TV 22: Very high yielding, drought tolerant with different genetic background. High photosynthesis and resistance to Eelworm infestation. Leaf blade expanded (small), leaf shape ovate, leaf base shape obtuse.

TV 23: Very high yield, vigorous growth, drought tolerant with different genetic background, termite resistant. Leaves glossy, leaf margin wavy, serration very high.

TV 24: High leaf proline, cold resistant. Leaf venation not prominent, leaf blader margin entire, leaf apex acute.

TV 25: High yield with high leaf area index. Tolerant to drought with different genetic background. High water use efficiency and resistance to cockchafer grub. Leaf apex macronulate, leaf apex habit down turned, bullation prominent.

TV 26: High yield, tolerant to drought with different genetic background. Low transpiration, Resistance to Root knot nematode. Leaf apex acute, leaf apex habit down turned, Leaf bullation absent.

TV 27: High yield a, high shoot density and resistant to black rot. Leaf shape lanceolate, leaf base semi erect (50° - 70°), leaf colour intermediate.

TV 28: High yield and tolerant to drought with different genetic background. Resistance to blister blight. Leaf shape ovate, leaf base erect ($<50^{\circ}$), leaf colour light green.

TV 29: High yielding variety, Triploid ($3n=45$), resistant to root diseases. Leaf shape elliptical, leaf colour dark and medium glossy, large shoot size.

TV 30: High yield with different genetic background. Resistance to blister blight and tea mites. Leaf shape ovate, light pink pigmentation during winter and moisture stress conditions.

TV 31: Coppery yellow shoot; Resistance to tea mosquito bug. Coppery yellow young shoot, leaf shape elliptic, leaf base semi erect (50° - 70°).

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