A case for a core collection of sugarcane germplasm

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ABSTRACT

Enthusiasm and support for collection of sugarcane germplasm has not been matched by effort in its conservation, documentation and use. Rate of attrition in the USDA World Collection has been high, but germplasm in the Indian World Collection appears to be secure. Good work done in the characterization of sugarcane has been partly negated by a lack of uniformity in the descriptors used and inaccessibility of results. These problems arise from absence of an effective central body to communicate the need and requirements for uniformity and to collect and analyze data. If sugarcane germplasm collections are to be used more effectively in the future than at present, they must be rationalized, refined and structured around well-defined, representative “cores”. These core collections would represent, with a minimum of repetitiveness, the genetic diversity of Saccharum and its relatives. The genetic basis of a core collection is discussed, together with procedures for its establishment and use. Sufficient information exists to designate clones to a core collection. However, collection and analysis of the data for selection of entries is a major task. While best coordinated by ISSCT, a well-prepared proposal for this work could attract external funding. Establishment of a core collection is essential to improve conservation, documentation and use of sugarcane germplasm.

Key words: Germplasm characterization, germplasm conservation, cane breeding.

INTRODUCTION

Widespread concern in recent years that plant genetic resources are under increasing threat in a rapidly developing world has stimulated activity in germplasm collection. However, recent enthusiasm and support for germplasm collection, including sugarcane, has not been matched by effort in conservation, documentation and use. The problem is not new, as in 1962 Simmonds argued against genebanks, which he termed ‘museum collections’. The criticism has been repeated by many others in recent years, but there are divergent views, e.g. Brown developed the thesis that if plant germplasm collections are to be used
more in the future than at present; they will need to be better collections. A ‘better’ collection was considered to be one that is rationalized, refined and structured around a well-defined, representative core.

The objectives of this paper are (i) to review the present status, documentation and use of existing germplasm collections and (ii) to examine the feasibility and benefits of establishing a core collection for sugarcane.

STATUS OF SUGARCANE GERMPLASM COLLECTIONS

1. Status of Collections: Sugarcane germplasm is held principally in two ‘world’ collections, one maintained in the USA and the other in India, maintained by the Sugarcane Breeding Institute. They are to a large extent duplicates. In addition to these two collections, a number of collections comprising Saccharum species, related genera and breeding hybrids are held in several other countries and some contain accessions not present in the World Collections. While size of collections should be measured by their comprehensiveness in genetic diversity rather than their numerical size, Table 1 provides some indication the scope of the two World Collections.

TABLE 1. Clonal entries in World Collections of the Saccharum complex:

<table>
<thead>
<tr>
<th>Germplasm group</th>
<th>USDA 1984*</th>
<th>USDA 1991**</th>
<th>India 1987***</th>
<th>India 1991****</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saccharum officinarum</td>
<td>169</td>
<td>546</td>
<td>568</td>
<td>632</td>
</tr>
<tr>
<td>S. robustum</td>
<td>62</td>
<td>200</td>
<td>121</td>
<td>386</td>
</tr>
<tr>
<td>S. spontaneum</td>
<td>118</td>
<td>266</td>
<td>355</td>
<td>415</td>
</tr>
<tr>
<td>S. barbieri</td>
<td>52</td>
<td>14</td>
<td>59</td>
<td>43</td>
</tr>
<tr>
<td>S. sinense</td>
<td>80</td>
<td>84</td>
<td>37</td>
<td>29</td>
</tr>
<tr>
<td>S. edule</td>
<td>9</td>
<td>18</td>
<td>29</td>
<td>29</td>
</tr>
<tr>
<td>Allied genera</td>
<td>195</td>
<td>215</td>
<td>120</td>
<td>24</td>
</tr>
<tr>
<td>Man-made hybrids</td>
<td>168</td>
<td>174</td>
<td>219</td>
<td>1,728</td>
</tr>
<tr>
<td>Recent Indian collections</td>
<td>484</td>
<td>398</td>
<td>1,722</td>
<td>1,728</td>
</tr>
<tr>
<td>New accessions from USDA</td>
<td>2,404</td>
<td>5381</td>
<td>529</td>
<td>3,384</td>
</tr>
<tr>
<td>Unclassified</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Totals</td>
<td>3,237</td>
<td>1,787</td>
<td>1,504</td>
<td>3,438</td>
</tr>
</tbody>
</table>

* Data from Berding and Roach
** Data provided by Director, Sugarcane Breeding Institute, Coimbatore
*** Walker
The USDA World Collection was initially sited at Canal Point but at the time the data in Table 1 were supplied, the collection was in the process of transfer to Coral Gables, Miami. The latter site on oolitic limestone soil is an undesirable environment for most sugarcane material. Tew \textsuperscript{53} reported that many clones of \textit{S. officinarum} had been established in Hawaii from the Miami collection and most had flowered. This has enabled establishment of a seed genebank using material polycrossed in 1990. Part of the Miami collection of related grasses was discarded after collecting seed and depositing it in the seed storage facility at Fort Collins, USA\textsuperscript{54}. There has been concern at the rate of attrition that has occurred in the USA Collection following its placement at Miami in 1976\textsuperscript{55}. However, it should be borne in mind that the organizations maintaining world sugarcane collections provide the sole funding for their upkeep. They provide a free service to the world sugar industry and the USDA collection in particular has been valuable to many programs.

The World Collection in India is maintained under much more favorable conditions at three locations.

1. Sugarcane Breeding Institute, Coimbatore. Mosaic is present in the area and only the \textit{Erianthus} collection and part of the \textit{S. spontaneum} collection is maintained there.

2. SBI Research Center, Cannanore. This very favorable environment is mosaic free and the remainder of the collection, other than \textit{Miscanthus} is maintained there.

3. The International Cane and Sugar Research Institute (ICRI), IARI Research Center, Wellington. Clones of \textit{Miscanthus}, an upland species with poor survival at low altitudes, are maintained at this high altitude site in the Western Ghats.

Clones in these collections are replanted annually and losses are rare. In addition, a back-up in vitro genebank is being established for further safety\textsuperscript{56}.

Over the past decade there has been much activity in germplasm collection, particularly in Southeast Asia\textsuperscript{57} and sugarcane workers have shared in this activity. Their interest has ranged over a wide diversity of material in several regions, with almost 1,700 clones collected. However, one must question both the reality of objectives and the value of collections of this magnitude, given the past performance of sugarcane workers in maintaining, characterizing and using material previously collected.
1. Documentation

The following terms, defined by IBPGR, provide a hierarchy of increasing detail of documentation:

(i) **Passport information** consists of data recorded at the time of collection of an accession, including identifying names and numbers.

(ii) **Characterization** consists of recording details of some botanical characters that are highly heritable, are expressed in all environments and can be easily observed by eye.

(iii) **Preliminary evaluation** consists of recording a limited number of additional agronomic traits thought desirable by a consensus of users.

(iv) **Further evaluation** consists of recording other traits related to breeding programs. This is a theoretically infinite task involving designed experiments.

While there has been a number of excellent individual efforts in documentation of sugarcane germplasm, absence of a central coordinating body has resulted in some information being lost and the remainder largely inaccessible.

Characterization has been regarded as one of the most neglected tasks implicit in genetic resources activities and the two main problem areas are: (i) the quality of characterization work done and (ii) the quality of this characterization. For sugarcane, problem (ii) is perhaps the more important, as much of the work done has been on an *ad hoc* basis, lacking uniformity with other work and making aggregation of data more difficult. The problem has been recognized for a number of years and numerous procedures have been developed and recommended to improve the situation. Current confusion and lack of uniformity in characterization of sugarcane germplasm is typified by the following:

- In 1985 and 1991 the Indian Council of Agricultural Research published excellent catalogues on sugarcane genetic resources. However, the descriptor and descriptor states used were different from those suggested by IBPGR.
- In 1989 a botanist sponsored by IBPGR made descriptions of the entire World Collection at Miami. Apart from the dubious value of characterization in the adverse environment, the descriptors used differed from those suggested by IBPGR Working Group and those used in the Indian catalogues.

No report has been published so far.
The practical value of all this well-intentioned work could well be questioned. Sugarcane breeders have been slow to develop and adopt uniform descriptors, partly due to the difficulty in providing a single set of descriptors for contrasting Saccharum species. The problem is now being addressed, with descriptor sets under development and evaluation. It is important that recommendations resulting from this be communicated to individual sugarcane breeding stations, where much of the information originates and where it will be ultimately used.

3. Use of Sugarcane Germplasm

Berding and Roach summarised the extent and nature of use of sugarcane germplasm and its past benefits in improvement of the crop. Evidence that there is still adequate genetic variance in existing breeding pools for continued progress and the long and difficult processes in the use of new germplasm and continual pressure for new varieties to meet perceived needs have not favored advances in the unknown. Benefits from programs begun in the 1960s to exploit new germplasm have been slow to emerge and possible reasons were reviewed by Roach and Roach and Daniels. However, there is recent evidence of positive results in the temperate cane growing areas of Louisiana, USA and New South Wales, Australia.

4. THE CONCEPT AND REASONS FOR A CORE COLLECTION

Franke proposed that a germplasm collection could be pruned to what he termed a 'core collection' which would represent 'with a minimum of repetitiveness the genetic diversity of a crop species and its relatives'. Accessions not included in the core collection would not be discarded but retained, apart from duplicates, in a 'reserve' collection.

Brown discussed the concept, constitution and use of core collections, providing examples of such collections established in Australia. He considered that germplasm collections were now so large as to deter their extensive use for all characters which are readily and reliably distinguished on single plants, e.g. some morphological traits, major genes for disease resistance. Greater use of a collection could be made, particularly for a wider range of characters, if a smaller number of accessions were given priority in evaluation and hybridization. The designation of a core in a collection can provide a ready reference set
of accessions for assessing redundancy. Brown also considered designation of a core necessary for testing general combining ability with locally adapted germplasm in the search for yield enhancement. This latter concept is similar in principle to the work initiated and reported by Roach in selection and hybridization of representative clones of species of the Saccharum complex. Finally, Brown considered that documentation and communication of results informally and through the scientific literature would be facilitated by concentration on a reduced number of accessions.

STATISTICAL AND GENETIC BASIS OF CORE COLLECTIONS

Any sample of germplasm for a core collection cannot include all the genetic variation, but excluded variation is not lost if the reserve collection is maintained. Brown used random sampling theory and various allele frequency profiles to analyze baseline expectations. On the basis of this analysis for sexually propagated species, he suggested that a sensible target size for a core collection should be around 10% of the total collection, up to about 3,000 entries.

There is good evidence that the genetic diversity of a species is not randomly dispersed within and among populations, but is organized to varying degrees. This basic structure of species variation means that carefully chosen sub-samples of a species will contain more of the species genetic diversity than predicted by random sampling theory. This was the basis for partition by Roach of Saccharum species into sub-groups and use of representatives of these for hybridization. However, the same basic structure of species variation means that carelessly chosen sub-samples could well reduce the level of diversity in the core below that expected in a random fraction. This can happen when choice is governed unduly by any character state, e.g. rust resistance, or by one geographic region, e.g. a center of genetic diversity.

PROCEDURES FOR ESTABLISHING A CORE COLLECTION OF SUGARCANE GERmplASM

1. Size of Collection

Options are a proportion of existing accessions or an upper limit. The numbers of clonal entries in the World Collections of sugarcane are not large in relation to numbers of accessions in major seed-propagated crops. However, to fulfill its purpose, a core collection of sugarcane germplasm requires clonal maintenance in the field. Management must be of a high standard to eliminate losses and errors of identity and to provide material in suitable condition for meaningful characterization. A collection of 500 clones should be manageable, somewhat
more than 10% of the 4,000 or so clones in the World Collection in India. However, establishment of a core collection of sugarcane germplasm would, of necessity, need to be progressive as data were collected and analyzed, to enable selection of clones for entry to the collection. This process would provide a much better indication of the size of collection necessary to adequately represent the genetic variation of Saccharum and its relatives.

2: Classification of Collection for Selection of Core Entries

Classification consists of analysis of the collection to identify degrees of genetic similarity among the accessions. The operation is essentially a hierarchical cluster analysis, where each accession is sorted into a related sub-group from which a representative sample will be drawn. Multivariate methods such as numerical cluster analysis, principal component analysis and/or network analysis are valuable tools, particularly when many characters have to be evaluated on most accessions. The three types of data which can be used are listed below, together with an assessment of the availability of such data for accessions in sugarcane germplasm collections.

(i) Origin of accessions. The system of passport information comprising accession data and collection data accepted at the IBPGR Working Group meeting in 1981 included 17 categories of information. Much of the information has been recorded by collectors of sugarcane germplasm, but some details, e.g. latitude and longitude of collection have rarely been recorded. However, it is generally possible to determine from collector’s notes or other sources reasonably precise details of geographic origin of a clone. This, together with ecological information, enables an important initial separation of collection accessions.

(ii) Characterization data. Most clones in sugarcane germplasm collections can be reliably allocated to a taxonomic group and this, together with geographical origin, enables ready and reliable separation of accessions into sub-groups based on these descriptors. Further separation relies on characterization data which will vary greatly in its availability, its nature according to species, its uniformity and reliability. However, the answer to these problems is not to ignore the assembled data and choose accessions at random. Nor should a group necessarily be represented in the core in the proportion to which it occurs in the collection. Preference could be given to clones with more extensive or reliable data, provided that overall representation is maintained.

As noted previously, a great deal of work has been done in characterizing sugarcane germplasm. While value of the data is reduced by lack of uniformity, the greatest problem is its inaccessibility. Whether a core collection of sugarcane is established or not, collection, collation and analysis of characterization data for sugar cane is crucial to long-term improvement of the crop.
The nature of characterization data for sugarcane varies widely. Mention was made previously of some large-scale morphological studies and similar data are available from other sources. From 1986-1989 Tyrell28 studied 500 clones from the USDA collection, using botanical descriptors for species and genera not strongly influenced by environment. Biochemical marker studies include triterpenes30, leaf flavonoids30, 13, 33, 34, 60 and isoenzymes12, 16, 55. In a recent major study, Wood62 surveyed approximately 1,500 accessions in the USDA World Collection for variation in 10 enzyme systems. Variation in nuclear ribosomal DNA in sugarcane has recently been studied by Glaszmann et al. 19, 20 and Wood61.

There is obviously an abundance of data for meaningful classification of sugarcane germplasm at the tertiary level, i.e. below geographical origin and species. Such tertiary classification has already been done by multivariate analysis for clones of S. spontaneum from Japan29, 30. Groups identified showed close association with islands of origin and representatives of these groups should be included in any core collection of sugarcane germplasm.

(iii) **Evaluation data.** While sugarcane literature contains an abundance of evaluation data, the high level of genotype x environment interaction for most characters influencing yield limits the value of these data. Such data are of limited practical value for wild germplasm, which is utilized via introgression rather than directly and where phenotype is a poor guide to its breeding value for yield40. Evaluation data are obviously of greater importance for classification of commercial type hybrids, if these are to be included in the core collection.

(iv) **Evolution of the core collection.** It is not necessary that the total core set of accessions be designated at once; accessions can be included in the core as justified by additional information. The core set of accessions should be dynamic rather than static and changes in content and size are to be anticipated with time41. However, alteration of the core should be a relatively rare process. A major aim of the core is to build up a body of information on a restricted reference set of clones and rapid movement of accessions through the core would defeat this aim41.

(v) **Location and use of the core collection.** To be effective, the core needs to be incorporated with the major germplasm collection for the crop and in a location where risk of loss of accessions is minimal and reliable data on accessions can be collected. The World Collection in India meets these requirements, but the World Collection at Miami does not.

The objective of the core collection is to make available a well-documented, well-characterized set of accessions covering most of the genetic variation of the total collection. Main use of the core will be either through on-site studies of the core or through breeders and other researchers obtaining sub-sets of the core for study and
use in their own environment. In recent years, some countries, e.g. Brazil and Cuba, have acquired large collections of sugarcane germplasm and Cuba has indicated its desire to have its own 'world collection'. Progressive acquisition by such countries of clones in the designated core collection would provide them with better collections than could be obtained by more or less random acquisition from the entire collection. They would have a set of clones which in total more fully represented the genetic variation of the crop and individually were better characterized. Such countries could add considerably to overall value of the core by characterization and evaluation in their own environments.

MAINTENANCE AND ROLE OF THE RESERVE COLLECTION

It is important that if a core is designated for a collection that the remaining accessions do not suffer from neglect or become vulnerable to short-sighted economies. To prevent this, the reserve must be seen as part of the genetic resources of the collection, serving at least three functions:

(i) As a back-up resource which can be screened for a needed variant, if such is not available in the core.
(ii) Alternative sources of the same character, possibly in different genetic backgrounds.
(iii) A reserve source of seed for a clone in the core collection.

To fulfill these functions for sugarcane germplasm, a well-maintained clonal field collection is necessary, as presently exists in India. In vitro collections can provide very valuable back-up, as done for cassava by CIAT. As noted before, an in vitro gene bank is being created in India as a back-up to the field genebank of sugarcane germplasm.

Seed genebanks provide the basis for conservation of genetic resources of many crops and the necessary technology and management procedures are well documented. Seed storage has been advocated for conservation of genetic resources of sugarcane and reference was made earlier in this paper to use of this procedure to maintain some of the genetic variation from the USDA collection. However, a seed genebank cannot maintain the full spectrum of genetic variation in a sugarcane germplasm collection for the following reasons:

(i) Clones of the Saccharum complex require a diversity of environments for flowering and some do not flower at all. Artificial induction of flowering would be impractical for large numbers of very dissimilar clones.
(ii) Sugarcane clones flower over a lengthy period of several months and artificial synchronization of flowering is impractical for large numbers of clones.
(iii) Even if synchronized flowering were achieved, problems of male and female sterility remain. Such problems may affect a whole group or species, e.g. *S. sinense* Roxb.

The rationale for use of a seed genebank for sugarcane is that the objective is to maintain genes rather than individual clones. However, for *Saccharum* and its relatives, seed storage cannot meet the objective of maintaining a representative sample of the total genetic variation.

**CONCLUSIONS**

Existing breeding pools may satisfy the immediate needs of sugarcane breeders, although there is little evidence of significant recent yield improvement of the crop. In the longer term, use of the large reservoir of available germplasm will be necessary for continued progress but occasional random sampling of this germplasm is unlikely to be very productive.

Effective use of germplasm in World Collections would be greatly facilitated by establishment of a small, well-characterized 'core' collection which represented, with a minimum of duplication, the genetic diversity of *Saccharum* and its relatives. Collection and analysis of characterization data to enable designation of clones to a core collection for the *Saccharum* complex is a matter of high priority for improvement of the crop. While there are problems with lack of uniformity of the large volume of existing characterization data, the main obstacles to use of these data at present are lack of accessibility and lack of organization.

An effective basic core collection for the *Saccharum* complex could be established using existing data and the necessary collection, collation and analysis of these data could attract funding outside the cane sugar industry. This core could be progressively refined and enlarged, if necessary, to further improve its effectiveness in the future.

**REFERENCES**


PLAIDOIRIE POUR UNE COLLECTION STRUCTUREE DU PATRIMOINE GENETIQUE DE LA CANNE A SUCRE

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RESUME

L'enthousiasme et le support dégagés pour la collecte du patrimoine génétique de la canne à sucre n'ont pas été suivis par un effort similaire quant à sa conservation, sa documentation et son utilisation. La collection mondiale à l'USDA est soumise à un fort taux d'usure, alors que celle de l'Inde semble être en sûreté. Le bon travail accompli dans le catalogage de la canne a été partiellement anéanti par un manque d'uniformité des descripteurs et le manque d'accès aux résultats. Ces problèmes surgissent par l'absence d'un corps central capable de communiquer les espérances et les besoins pour l'uniformisation, la collection et l'analyse des données. Si les collections mondiales sont appelées à être mieux exploiter à l'avenir, elles doivent être rationalisées, redéfinies et restructurées autour des noyaux représentatifs. Ces organismes pourraient représenter la diversité génétique du genre *Saccharum* et les genres apparents avec un minimum de répétition. La base génétique d'un tel groupement structuré est discutée ainsi que les étapes pour sa création et son utilisation. Il existe suffisamment d'information pour allouer les blocs à des groupes spécifiques. Cependant, le choix des entrées à partir de la collection et l'analyse des données reste un exercice majeur. Bien qu'elle pourrait être gérée le mieux par l'ISSCT, une proposition bien formulée dans ce sens, pourrait bénéficier des investissements extérieurs. La création d'un organisme pour une collection structurée est indispensable si l'on veut améliorer la conservation, la documentation et l'utilisation du patrimoine génétique de la canne à sucre.

Mots clés: Patrimoine génétique de la canne à sucre, caractérisation, conservation, amélioration de la canne à sucre.
EL CASO PARA UNA COLECCION FUNDAMENTAL DEL GERMOPLASMA DE CAÑA DE AZUCAR

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RESUMEN
El entusiasmo y apoyo para la colección del germoplasma de caña de azúcar no ha sido correspondido por un esfuerzo en su conservación, documentación y uso. Ha sido grande el grado de abandono de la Colección Mundial en USDA, mientras que el germoplasma en la Colección Mundial de India parece estar seguro. No se ha hecho un buen trabajo en la caracterización debido parcialmente a la carencia de uniformidad en los descriptores usados e inaccesibilidad de los resultados. Estos problemas resultan por la ausencia de un cuerpo central eficaz que comunique las necesidades y requerimientos de uniformidad y que recoja y analice la información. Si las colecciones del germoplasma de caña de azúcar están para usarse más efectivamente en el futuro de lo que son en el presente, ellas deben ser racionalizadas, redefinidas y estructuradas alrededor de representantes bien definidos por grupos élitis. Estas colecciones élitis representarían, con un mínimo de repetibilidad, la diversidad genética del género *Saccharum* y los géneros relacionados. Se discuten las bases genéticas de la colección fundamental, al mismo tiempo que los procedimientos para su establecimiento y uso. Existe suficiente información para asignar clones a una colección élitis. Sin embargo es un gran trabajo la colección y análisis de la información para la selección de éstos. Una proprusta bien preparada y coordinada por ISSCT para este trabajo, podría atraer ayuda externa de fondos. El establecimiento de una colección élitis es esencial para mejorar la conservación, documentación y uso del germoplasma de caña de azúcar.

Palabras claves: Caracterización de germoplasma, conservación del germoplasma, mejoramiento genético de caña de azúcar.