CacaoNet Workshop on the Development of the Global Strategic Cacao Collection (GSCC), 22-24 October 2014, Trinidad

Jointly organized by Bioversity International, on behalf of the Global Cacao Genetic Resources Network (CacaoNet) and the Cocoa Research Centre (CRC)

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CONTENTS

1. BACKGROUND OF THE WORKSHOP

The creation of a network to optimize the conservation and use of cacao genetic resources worldwide for the benefit of breeders, researchers and farmers was proposed in 2005. CacaoNet was officially launched at the COPAL 15th International Cocoa Research Conference in San José, Costa Rica, in October 2006. Financial and in-kind support has been contributed from a number of organizations including Bioversity International, CRA Ltd, Mars, USDA/ARS, WCF, and COPAL which has permitted the CacaoNet steering committee and working groups to meet, and the coordination of the network. CacaoNet coordinated the development of a Global Strategy for the Conservation and Use of Cacao Genetic Resources in consultation with a wide range of experts and stakeholders. The Strategy was finalised at the end of 2012 and can be downloaded at www.cacaonet.org. CacaoNet is indebted to the research institutes and organizations who have allowed their staff to participate in the network and to the individuals who have contributed their valuable time and expertise.

No country is self-sufficient when it comes to the range of genetic diversity needed to develop improved materials. This diversity is maintained by several research institutes but only a part of that diversity is available in the public domain. Only the two international collections at the *Centro Agronómico Tropical de Investigación y Enseñanza* (CATIE) and at the Cocoa Research Centre of the University of the West Indies (CRC/UWI), have placed their cacao germplasm under the auspices of the Governing Body of the International Treaty on Plant Genetic Resources for Food and Agriculture (IT-PGRFA), with the commitment to safely conserve for the long term according to international standards and make the materials readily available to any plant breeding programmes and other *bona fide* users. In addition to CATIE and CRC, some national collections can also be considered to be within the public domain, such as the collections at CIRAD and USDA.

The collection at CATIE was initiated in 1944 in Turrialba, Costa Rica, as part of a strategy of the Inter-American Institute for Cooperation on Agriculture (IICA) to promote the exchange of germplasm of tropical crops. In 1978, CATIE's collection was registered by the International Board for Plant Genetic Resources IBPGR (now Bioversity International) as a global base collection and since 2004 it is under the auspices of the Food and Agriculture Organization of the United Nations (FAO) and covered by the ITPGRFA. In the nineties, CATIE with the support of the WCF initiated a regional cacao breeding programme. The focus of the programme is selection and generation of high-yielding and disease resistant genotypes with emphasis on moniliasis (*Moniliophthora roreri*) and black pod (*Phytophthora palmivora*) diseases, two of the major biotic factors limiting cacao production in Central America and Mexico. The original source of the experimental germplasm is the CATIE International Cacao Collection (IC3), which currently comprises 1,217 accessions collected, introduced or selected/bred by IICA/CATIE over the last 70 years with different genetic and geographic origin from Central America, Mexico, South America, the Caribbean, Asia, and Africa. WCF and USDA collaborated with CATIE towards the renovation of the collection and its genetic enrichment by introducing strategic germplasm and the first effort towards its genetic rationalization. The objectives of the renovations were to curtail further losses of accession caused by soil-born fungus by re-organizing the collection, standardizing the number of plants per accession, rejuvenate the old trees and maintain replicates of each accession at different sites for security reasons. The improvement of the collection is a priority and the further reorganization in order to improve the accuracy and efficiency of maintaining this collection. www.catie.ac.cr

CRC/UWI maintains the International Cocoa Genebank, Trinidad (ICG,T) established in 1982, by consolidating diverse earlier collections of cacao from several sites in Trinidad which included accessions from other national collections and from numerous missions to collect primary germplasm from the centre of diversity of cacao. A main source of original material for the ICG,T was Marper Farm,

established by F.J. Pound following his expeditions to the upper Amazon from 1937 to 1942. The trees at Marper though now old, have survived periods of neglect to remain as an important anchor in confirming the identity of clones in the ICG,T and in replacing material which has proved difficult to establish. Trees in the ICG,T were propagated as rooted cuttings using budwood from the original trees and, by 1994 over 2000 accessions had been planted. ICG,T genebank now contains one of the most diverse collections of cacao germplasm and consists now of 2400 accessions, representing the major groups of cacao (Forastero, Criollo, Trinitario and Refractario) as well as related species of *Theobroma*. About 40% of the accessions are in the Forastero group, 40% in the Refractario group, 10% in the Trinitario group and the remainder either Criollo, hybrids or unclassified. Recent collections of primary germplasm (still to be introduced to the ICG,T) aim to increase the representation of the Criollo group. In addition, some accessions are used in pre-breeding programmes to accumulate desirable genes especially for resistance to Black Pod and Witches' Broom diseases. The main objective of such programmes is to produce enhanced germplasm that will introduce resistance genes to conventional breeding programmes in various cocoa-producing countries throughout the world. The work of CRU/UWI is supported financially in part by CRA Ltd, the Dutch Ministry of Agriculture, Nature and Food Quality or Ministerie van Landbouw, Natuur en Voedselkwaliteit, The Netherlands (LNV) and the Ministry of Food Production (MFP), Trinidad and Tobago. http://sta.uwi.edu/cru/

The International Cocoa Quarantine Centre at Reading, UK (ICQC,R) holds cacao accessions available in the public domain and acts as the international quarantine centre for the safe movement of cacao genetic resources throughout the world. The ICQC,R, established in 1985 holds approximately 450 cacao accessions (350 clones available for exchange and a further 100 undergoing quarantine). The ICQC,R is funded by the Cocoa Research Association Ltd (CRA Ltd) and the United States Department of Agriculture (USDA) with additional funding from the Common Fund for Commodities (CFC). The ICQC,R provides pest and disease free material both for use within the University and internationally. The current quarantine procedure involves a two year visual observation period to check for latent viral infections supervised by an experienced virologist. The facilities, plus laboratories fitted with the latest equipment for molecular biology and *in vitro* culture, enable pioneering research in cocoa physiology, pathology, genetic fingerprinting and tissue culture. Since 1985 many cacao clones have passed through the ICQC,R facility mostly from CATIE and CRC but material has also been received from the wild and from national collections. In addition, ICQC,R cryopreserves frequently requested clones and it is the aim to back-up 10% of the ICQC,R collection in the coming years. www.icgd.reading.ac.uk/quarantine.php

Apart from the germplasm maintained at CATIE and CRC, the remaining collections are considered to be national assets and are generally not publicly available outside of the country holding the collection, with the exception of USDA and CIRAD's collections. In many Latin American countries possessing primary sources of cacao genetic diversity, policies restrict opportunities for newly collected materials to be put into the public domain. Unique and valuable material is conserved in these national collections and thus, collaboration is needed to secure this material and increase the access to and their use in breeding programmes.

At the heart of the Global Strategy is the development of a Global Strategic Cacao Collection (GSCC) is a virtual collection consisting of materials that have been identified as unique and interesting and that is currently available to all in the public domain. The materials in the CATIE and CRC/UWI collections will form the backbone of this GSCC complemented with priority accessions from national collections available in the public domain.

The formation of the GSCC will result from a coordinated effort of characterization and rationalization of available cacao genetic resources. Each of the participating institutes will agree to conserve these accessions according to agreed practices and standards and make them readily available to any *bona*

fide user. The objective of the GSCC is therefore to ensure the cost-effective and efficient long-term *ex situ* conservation of the entire *Theobroma* genepool and its accessibility to all current and future users.

Agreed criteria such as genetic diversity, in the form of allelic richness and the uniqueness of each genotype, in combination with measures of agronomic value will be used to identify priority accessions. A study of allelic diversity conducted using the SSR data available in 2009 identified 261 accessions amongst the genotypes studied that best represented the allelic richness observed across ten population groups, capturing the majority of the known genetic and geographic diversity held within *ex situ* collections worldwide. These genotypes are a priority for inclusion in the GSCC though many of them are not yet held in the public domain. A further set of accessions will be prioritised on the basis of key traits of interest to users such as yield, flavour characteristics and disease resistance for which agreed criteria will be developed. It is acknowledged that, even within the international genebanks, there are a large number of genotypes which have yet to be fully evaluated for these characteristics and further research may also be needed to understand the genetic basis of these traits to ensure that a particular mechanism or pathway is not under-represented, or indeed over-represented, within the GSCC.

Thus, although the GSCC will be initially composed of all the accessions held in the public domain, predominantly by the international collections at CATIE and CRC/UWI, it is expected that there will be a process of gradual refinement of its composition as more information becomes available on the genetic and phenotypic characteristics of the accessions, misidentification and unnecessary duplication is reduced and new accessions become available for inclusion.

The collection managers and breeders around the world will be responsible for comprehensive characterization, evaluation and further researching of the GSCC collection. All related information should be made available to all users through GSCC information portal – CANGIS. The specific criteria and boundary for each set of accessions would be agreed through a consultation process coordinated by CacaoNet. This assessment would be part of a rationalization plan, with clear objectives, that would take place over time as knowledge becomes available. It is proposed that CacaoNet members would be responsible for the composition of the GSCC as well as for recommending, and where possible supporting, priority actions such as detecting mislabelling, evaluation, characterization, evaluation of the potential of new technologies such as *in vitro* culture and cryopreservation for safety duplication, pre-breeding, distribution and use. This would include the participation of collection curators and the breeding community represented by INGENIC.

The 2012 Global Cacao Strategy recommended several actions to be taken, to be coordinated by CacaoNet in consultation with all its members. Therefore CacaoNet organized a first consultation on the development of the GSCC, jointly with the Cocoa Research Centre of the University of the West Indies in Trinidad, 22-24 October 2014. More details on the process of developing the GSCC are in ***Annex A*.**

2. GOAL AND OBJECTIVES OF THE WORKSHOP AND EXPECTATIONS FROM PARTICIPANTS

The GOAL of the CacaoNet workshop is to contribute to the sustainability of cocoa production through the implementation of the Global Strategic Cacao Collection (GSCC) as a means to secure the long-term conservation of cacao genetic diversity and optimise its utilisation.

The SPECIFIC OBJECTIVES proposed were:

- *Objective 1*: Review the current situation and concept: where we are with the development of the Global Strategic Cacao Collection (GSCC), its goal and proposed approach (allelic diversity and traits) and the information systems supporting the decision-making process – CANGIS and the ICGD.
- *Objective 2:* Agree on the overall approach and goal of the GSCC and future direction to secure and optimise the use of cacao genetic diversity.
- *Objective 3:* Propose ways to improve the availability of information that will contribute to the decision-making process for the GSCC, particularly for evaluation of traits of interest for users.
- *Objective 4:* Discuss the criteria to be used to identify priority accessions for the GSCC, mainly:
    A. *A first set of accessions selected on the basis of capturing the greatest possible range of allelic richness and diversity.*
    B. *A further set of accessions selected on the basis of key traits of interest to users such as yield, flavour characteristics and disease resistance for which agreed criteria will be developed.*
- *Objective 5:* Review the status of methodologies and scientific and technical advances made since the GSCC concept discussed and proposed in the Global Cacao Strategy in 2012. Particularly regarding the selection of accessions based on key traits of interest to users.
- *Objective 6:* Discuss the political aspects of conservation and accessibility of unique materials in the national collections.
- *Objective 7:* Agree on the short, medium and longer-term priorities for securing the *ex situ* conservation and identifying gaps, considering collecting priorities based on threats in the wild and on-farms.
- *Objective 8:* Agree on the general decision-making process mechanism for the GSCC, particularly for the process of refining the collection to improve its efficiency and safety duplication.
- *Objective 9:* Propose roles and responsibilities of the main partners in the GSCC, including ensuring the safety-duplication of the materials.
- *Objective 10:* Propose priorities for discussion during the follow-up CacaoNet workshop on the on-farm conservation of cacao genetic diversity (26-28 October, Guapiles, Costa Rica) and the meeting of the Latin America and Caribbean Cocoa Breeders group (CATIE, 30-31 October, Turrialba, Costa Rica).
- *Objective 11:* Make recommendations for the next steps and proposal for action:
    - Next steps from actions proposed, workshop documentation and report
    - Issues to be addressed by research
    - Action agenda for the partners (CATIE, CRC, ICQCR, and the national partners), data managers
    - Identification of agents suitable for political persuasion and process
    - Funding the process and timelines
    - Coordination and partnerships

Brigitte Laliberté, CacaoNet Scientific Advisor and workshop facilitator, presented the workshop objective listed above and the proposed details programme, included in ***Annex B***. The workshop was attended by 31 participants from around the world and 7 additional participants were connected via a video-conference GoToMeeting link. The full list of participants and email addresses is in ***Annex C***.

The participants were asked to provide feedback on expectations of the 3-day workshop. The majority expressed that they were looking forward to getting an update and clearer idea of how activities have moved on since the finalisation of the Global Strategy and were looking forward to concrete implementation and what should be the next steps and priorities. The different industry and research partners were interested in knowing more about how they can help and contribute to CacaoNet and the Strategy. The partners maintaining cacao genetic diversity were also interested in getting a better understanding of how the part of the global diversity that they preserve can contribute globally.

The participants were looking forward to open and constructive discussions towards identifying the key issues and for practical and realistic solutions to be proposed. Several participants also expressed that they were interested to understand how aspects of quality and diversity of flavour could address the loss of biodiversity and how it could be given priority for conservation.

The group also wanted to know more about how the Global Strategic Cacao Collection (GSCC) will include key traits of productivity, diseases such as Cocoa Swollen Shoot Virus (CSSV), quality and flavour and how it will link to the different regional breeders groups.

Some concerns were expressed as to how best ensure the conservation of the diversity that may not be of use in the short-term but may have resistance to diseases and tolerance to drought that would be useful in the long-term.

The workshop provides an opportunity to promote the implementation of genetic resources diversity conservation to the rest of the industry and to discuss the long-term funding of these precious resources. This includes getting a better understanding of the costing of the GSCC using modern science and best practices.

The workshop was also a good opportunity for many participant to network, exchange expertise with other genebank curators and discuss tools and solutions that can benefit the entire industry.

3. Acknowledgments

The CacaoNet workshop was made possible thanks to the financial contributions from Bioversity International and the CGIAR Research Programme on Forests, Trees and Agroforestry and to the support from the Cocoa Research Centre of the University of the West Indies. CacaoNet is grateful to Cocoa Research Association Ltd., UK (CRA Ltd.) for funding a number of participants and to the many individuals funding their own participation and their respective organisations for supporting the overall goal of the workshop. Special appreciation goes to the staff at Bioversity, namely Silvia Araujo de Lima for coordinating all logistic and participation and Karen Lehrer for the travel arrangements and at the Cocoa Research Centre, namely Prof Path Umaharan, Marissa Moses, Sophia Thompson for their excellent organisation and collaboration and to the support team at Ortinola particularly Nikita Nath for providing the participants with a wonderful setting and excellent food contributing to productive deliberations. CacaoNet would like to thank the members of the Programme Committee for their input into the development of the workshop.

4. Global Strategic Cacao Collection (GSCC) Approach, Current Situation and Future Direction

4.1 The Global Strategic Strategy for the Conservation and Use of Cacao Genetic Resources

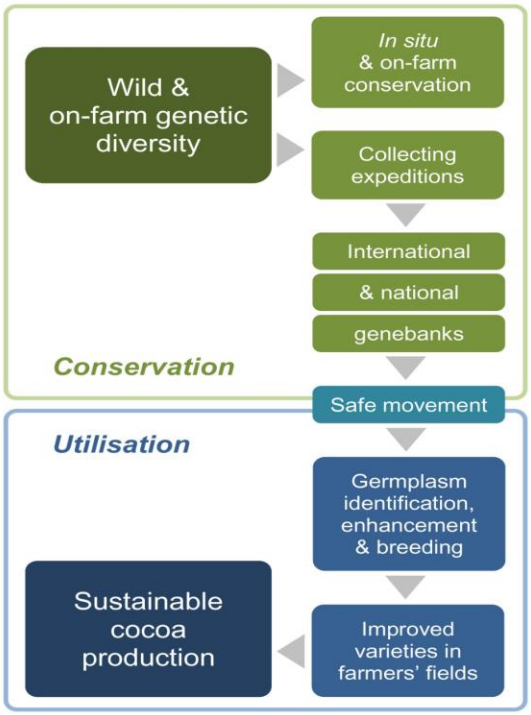
Brigitte Laliberté made a presentation on the Global Strategic Strategy for the Conservation and Use of Cacao Genetic Resources: a*s the foundation for a sustainable cocoa economy,* finalised in October 2012.

It included the following points:

Premise:

- The future of the world cocoa economy depends on the availability of a wide range of genetic diversity and the sustainable use to breed improved varieties.
- Decreasing cacao genetic diversity is a serious problem due to:
    - *Destruction of the Amazonian rainforests*
    - *Threats from natural disasters and extreme weather*
    - *Loss of traditional varieties*
- The loss of diversity increases the vulnerability of cacao to sudden changes in climate and to new pests and diseases.
- Most of the countries involved in cacao improvement and production are highly dependent on genes and varieties from other countries and regions.
- The efforts necessary to manage cacao genetic resources effectively can therefore only be carried out through international collaboration.

Extract from the Global Strategy - Figure 2. Links between the genetic diversity and sustainable cacao production (Credit: C. Turnbull, Reading University)



The Global Strategy:

- Developed by CacaoNet (Global Network for Cacao Genetic Resources)
- Result of a consultation process, drawing upon the global cocoa community's expertise in all aspects of cacao genetic resources (*over 75 individuals from 26 institutes contributed*)
- Provides a clear framework to secure funding for the most urgent needs to ensure that cacao diversity is conserved, used and provides direct benefits to the millions of small-scale farmers around the world

Where we are today:

- Over 35 collections maintain more than 24,000 samples of cacao diversity.
- Wide variation for disease resistance and quality exists in *ex situ* collections and in farmers' fields but its use it not optimised.
- Access is often restricted by lack of legal & policy framework.
- Only 2 international collections managed by CR/UWI, Trinidad and Tobago and CATIE, Costa Rica with an international agreement to maintain global collections of cacao genetic resources for the long term and to make this germplasm freely available to any *bona fide* user.
- Most collections have duplications internally and with other collections and misidentification of trees within collections can be as high as 30%.
- Only a few have strategic safety duplication of unique materials.
- Movement of germplasm brings risk of transferring pests and diseases.
- The safe global movement of cacao germplasm is through the International Cocoa Quarantine Centre at the University of Reading UK, (ICQC,R). The USDA/ARS facility in Miami offers quarantine facilities for regional transfers.

Where we want to go:

1. Securing existing *ex situ* cacao genetic resources, particularly those held in the public domain, and their distribution.
    - Highest priority to secure the conservation of the genetic diversity currently held in the public domain in ex situ collections and facilitate its safe distribution and its safety-duplication
    - Increase efficiency and effectiveness to reduce costs of conservation and increase sustainability
    - Develop a detailed fund-raising strategy and engage in dialogues with donors to secure funding for its short-term and longer-term objectives.
    - Promoting participation of all partners
2. Developing a Global Strategic Cacao Collection (GSCC).
    - Agree on the criteria for the selection of materials
    - Assess the cacao genetic diversity currently conserved in *ex situ* collections against these criteria
    - Identify those publically maintained unique accessions available for use by breeders
    - Characterize germplasm prioritized to allow assessment and recommendations for inclusion in the GSCC.
    - Field evaluation at multiple sites under controlled and recorded conditions for the proposed GSCC accessions.
    - Feasibility study on *in vitro* methods to facilitate distribution through quarantine facilities, including recommendations on type of materials (budwood or plantlets), impact and costing.
    - Costing study of the GSCC with conservation costs and associated services such as germplasm evaluation, quarantine, virus-indexing, distribution and documentation.

- Best practices for cacao collection management and develop standards.
- Ensuring the safety-duplication of the GSCC using appropriate methodology.

3. Genetic diversity gap filling in *ex situ* collections and collecting.

    - Assess the cacao genepool in the centre of diversity in situ and on-farm as a priority (Upper Amazon and Mesoamerica), including understanding the threats to genetic erosion.
    - Collect to fill gaps in *ex situ* collections, focusing on endangered wild cacao germplasm that are not yet in collections
    - Promote the establishment of a mechanism to identify and communicate threats to cacao genetic resources (vulnerability and erosion) at national and international level.

4. Ensuring the *in situ* and on-farm conservation of important genetic diversity.

    - Assess extent of genetic erosion of in situ and on-farm diversity, in centres of diversity.
    - Survey and inventory of landraces and traditional varieties in Upper Amazon and Mesoamerica
    - Develop scientific methodologies to assess impact of genetic erosion of on-farm diversity.
    - Analyze social, economic, market and cultural factors that influence farmers' maintenance of cacao diversity at the farm level and assess implications for designing in situ and on-farm conservation strategies and potential incentives such as Payments for Agro-Biodiversity Conservation Services (PACS).
    - Assess needs for in situ and on-farm conservation strategies for countries located in the cacao historical dispersal routes (Samoan islands, São Tomé and Príncipe, Reunion Island, Fernando Po Island, Sri Lanka etc.) potentially at risk as the cocoa sectors surrounding them are in significant decline.

5. Strengthening the distribution mechanism and safe movement of germplasm.

    - Support the maintenance and continued development of a network to facilitate the safe movement of cacao. This includes the ICQC,R, UK for international distribution, and regional facilities, to be established within institutes with quarantine facilities willing to play this role in the three regions.
    - Explore the feasibility of using in vitro methods for germplasm distribution through a research project.
    - Raise awareness of the new safe-movement guidelines
    - Reviewing the guidelines and incorporating any changes that have taken place regarding technologies and the distribution of pests and diseases.
    - Publishing the guidelines in French, Spanish and Portuguese.

6. Strengthening the use of the cacao genetic resources by providing support to breeders and key users through improved characterization, evaluation within collections and supporting population enhancement programmes.

    - Support a network of field trials participating in the evaluation of the GSCC materials at multiple sites.
    - Identify the most useful GSCC germplasm for distribution, adapting to the evolving needs of breeding programmes.
    - Make available a list of the main traits of accessions held in the ICQC,R which will help breeders prioritize their germplasm requests and assist them in locating material from local genebanks for inclusion in their breeding trials.

- Maintain the black pod and witches' broom enhancement programmes in CRC/UWI and frosty pod in CATIE, and continue to make available the best selections to the ICQC,R.
- Introducing accessions with diverse resistance to witches' broom and frosty pod into West Africa and South-East Asia breeding programmes.

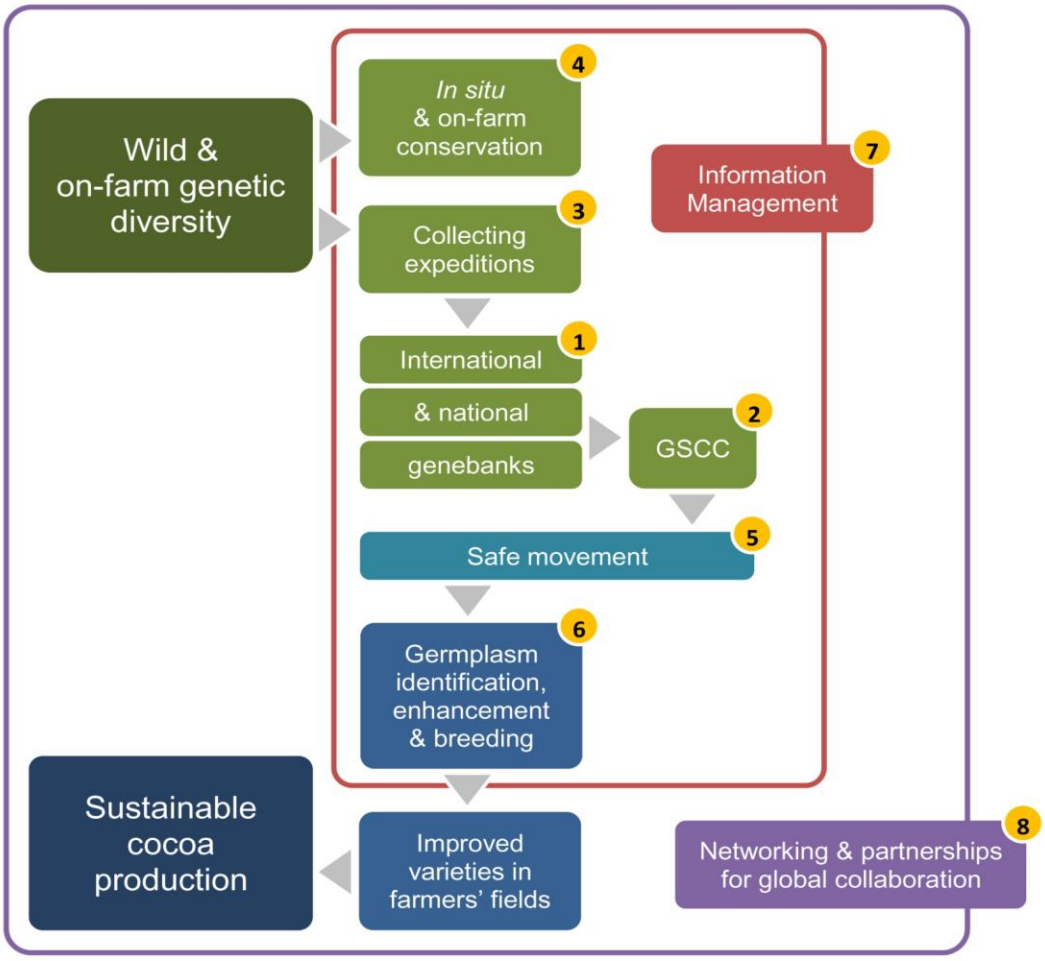
7. Improving documentation and sharing of information on germplasm.

    - Compile characterization and evaluation data from all collections (supported by molecular verification of genotypes where possible) to facilitate the identification of the GSCC including breeding and evaluation data.
    - Develop the GSCC information portal mainly based on information from the international collections held by CRC/UWI and CATIE and at ICQC,R.
    - Stimulate the rescue of historical data collected in genebanks and trials which can provide information useful to breeders.
    - Develop automated system for monitoring and updating the GSCC information portal, with particular emphasis on linking local germplasm management systems.
    - Develop a germplasm ordering and tracking systems.
    - Ensure appropriate level of record keeping in collections (working at tree level)
    - Assess the suitability of adopting GRIN-Global at collections that do not have a local information management system already by assessing minimum level of local expertise and IT equipment needed and the training requirements for initial set up (customization).

8. Strengthening the networking and partnerships for global collaboration.

    - Support the organization of regional and global workshops and expert consultations based on most critical needs.
    - Promote the availability of material in the public domain and participation in evaluation trials of GSCC materials.
    - Engage with national collections, FAO and the International Treaty to promote the placing of germplasm, particularly accessions identified for inclusion in the GSCC, in the public domain.
    - Ensure agreement on the establishment of the GSCC and is functions on behalf of all its members.
    - Engage in fund-raising for the implementation of the Global Strategy, including involvement of the private sector and international funding agencies to leverage funding for cacao genetic resources and establish of an endowment fund.
    - Play a key role in overall coordination and monitoring of the implementation of the Global Strategy and the dissemination of information.

Extract from the Global Strategy - Figure 5. The main strategic components from genetic diversity to sustainable cocoa production (Credit: C. Turnbull, Reading University).



Research and capacity building activities

- Development of *in situ* and on-farm conservation strategies.
- Diversity analysis to complement existing knowledge and to identify gaps for priority collecting.
- Research on tissue culture methods for safe movement of germplasm.
- Establishment of the regional quarantine network.
- Support for the *ex situ* collections partnering with the GSCC for linking to the GSCC information portal.

Annual recurrent management activities

- Support for the on-going maintenance of the GSCC.
- Emergency support to safeguard threatened material.
- Management of the GSCC information portal.
- Maintenance of the cacao safe movement network (quarantine facilities).
- Support for priority collecting missions.
- Network of field evaluation trials of priority GSCC materials.
- Training and capacity building for GSCC partners.
- Global partnerships towards the Strategy implementation.

4.2 Developing a Global Strategic Cacao Collection (GSCC) – Where we are today

The details of the GSCC Concept are in ***Annex A*** which includes the relevant extract from the Global Strategy. The concept is summarised here:

- No country is self-sufficient when it comes to the range of genetic diversity needed to develop improved materials.
- Wide variation for disease resistance and quality exists in ex situ collections and in farmers' fields but its use it not optimised.
- Only 2 international collections managed by CRC/UWI, Trinidad and Tobago and CATIE, Costa Rica with an international agreement to maintain global collections of cacao genetic resources for the long term and to make this germplasm freely available to any bona fide user.
- The remaining collections are considered to be national assets and are generally not publicly available outside of the country holding the collection.
- In many Latin American countries possessing primary sources of cacao genetic diversity, policies restrict opportunities for newly collected materials to be put into the public domain.
- Unique and valuable material is conserved in these national collections and thus, collaboration is needed to secure this material and increase the access to and their use in breeding programmes.
- The safe global movement of cacao germplasm is through the International Cocoa Quarantine Centre at the University of Reading UK, (ICQC,R). The USDA/ARS facility in Miami offers quarantine facilities for regional transfers.
- Most collections have duplications internally and with other collections and misidentification of trees within collections can be as high as 30%.
- Only a few have strategic safety duplication of unique materials.
- Movement of germplasm brings risk of transferring pests and diseases.

CacaoNet is working towards the establishment of a Global Strategic Cacao Collection (GSCC) as a virtual collection consisting of materials that have been identified as unique and interesting. Agreed criteria such as genetic diversity, in the form of allelic richness and the uniqueness of each genotype, in combination with measures of agronomic value will be used to identify priority accessions. Once the main part of the GSCC is formed, adding new diversity will be based on ensuring the genotype significantly increases the genetic diversity of the GSCC and/or this genotype has specific agronomic, quality or physiological traits that are of interest to users.

A first set of accessions will be selected on the basis of capturing the greatest possible range of allelic richness. These accessions would preferably be in the public domain but it is acknowledged that currently some may be maintained in collections not yet in the public domain. The Global Strategy aims to ensure that the institutes managing these accessions would conserve them for the long-term, evaluate them and take the necessary steps to make them publically available. For more details see ***Annex A1.*** *Annex 6. Description of the agreed methodology to select accessions based on allelic diversity and* ***Annex A1.*** *Annex 7. Proposed Membership of accessions for the GSCC based on allelic diversity.*

A further set of accessions will be selected on the basis of key traits of interest to users such as yield, flavour characteristics and disease resistance for which agreed criteria will be developed. Criteria for selection of genotypes may include in addition to the number of desirable traits present, the genetic diversity amongst the selected types as determined through DNA fingerprints. This part of the GSCC will complement the part selected on allelic diversity and be a dynamic and geographically dispersed collection composed primarily of wild species and populations, landraces, enhanced populations for

which characterization and evaluation data is available and used to broaden the basis on which the selection is made. This material will be in the public domain and accessible in the collections at CRC/UWI and CATIE for which considerable characterization and evaluation data are already available. Additional materials from national collections will become part of the GSCC if the governments concerned are willing to place them in the public domain.

The specific criteria and boundary for each set of accessions would be agreed through a consultation process coordinated by CacaoNet. This would include the participation of collection curators and the breeding community represented by INGENIC.

The proposed process of developing the GSCC is the following:

1. Genetic diversity in combination with measures of agronomic value will be used to identify accessions of interest.
2. A second round of selection aimed at reducing redundancy will generate the list of Priority Accessions.
3. Priority Accessions in the public domain will become part of the GSCC.
4. Public access will be requested for any Priority Accession not already in the public domain so that it can be included in the GSCC.
5. Each GSCC accession will be duplicated in another field collection for safety, and some may also be backed-up through cryopreservation (International quarantine required).
6. Material in the GSCC and all its associated information will be freely available for use in germplasm enhancement and breeding programmes, resulting in improved planting material becoming available to farmers (International and/or regional quarantine required).
7. Future collecting expeditions will target gaps in the GSCC (International and/or regional quarantine required).
8. Rationalization of the GSCC will continue as new material becomes available from collecting expeditions and breeding programmes.

*See **Annex A1** - Figure 6. Process for the development of the Global Strategic Cacao Collection (GSCC).*

The GSCC will be based on materials from the two international collections at CATIE and CRC/UWI which have placed their cacao germplasm under the auspices of the Governing Body of the ITPGRFA, the national collections that have proven records of making materials available and in public domain, such as the collections at CIRAD and USDA and from any collections willing to take the necessary steps to make their materials available particularly for use on breeding programmes. The GSCC will also rely on the critical role of the ICQC,R for the international safe movement of cacao genetic resources throughout the world.

The following specific actions were proposed:

- Agreeing on the criteria for the selection of materials (for both allelic diversity and traits of interest for breeding).
- Assessing the cacao genetic diversity currently conserved in *ex situ* collections.
- Identifying those publically maintained unique accessions that are available for use by breeders and researchers in the two international collections at CRC/UWI and CATIE and in national collections.
- Developing a proposal for reducing duplication of genetically similar clones, using genetic diversity assessment tools, with a focus on the collections at CRC/UWI and CATIE.
- Developing a process for resolving mislabelling problems in the international and national collections.

- Identifying urgent conservation support needed for the material identified for the GSCC.
- Characterizing public domain germplasm prioritized to allow assessment and recommendations for inclusion in the GSCC.
- Agreeing on field evaluation at multiple sites under controlled and recorded conditions for the proposed GSCC accessions.
- Agreeing on the safety-duplication of the GSCC in field genebanks and/or via cryopreservation.
- Conducting a detailed costing study of the GSCC with conservation costs and associated services such as germplasm evaluation, quarantine, virus-indexing, distribution and documentation.
- Promoting/holding continued discussions with the Global Crop Diversity Trust and with the private sector for possibilities of long-term funding support to the GSCC and with the Secretariat of the International Treaty and countries maintaining cacao materials targeted by the GSCC, to promote the designation of this germplasm under the Treaty following the example of CATIE and CRC/UWI.

See ***Annex A2*** – *Section 4.2* of the Strategy provide further details of the plans and the costing of the GSCC is discussed in the ***Section 8.2*** of this report.

4.3 INTRODUCTION TO THE CGIAR GLOBAL COLLECTIONS FOR LONG-TERM CONSERVATION

Brigitte Laliberté presented the example of the CGIAR global crop collections for long-term conservation and funding via the Global Crop Diversity Trust as a model for cacao. The key reference from the presentation was taken from the CGIAR publication titled*: In Trust for the International Community - Plan and partnership for managing and sustaining the CGIAR-held collections. The CGIAR CRP Research support. 2012.*

<u>The CGIAR Global Crop Collections – Introduction</u>

1. The CGIAR
2. The Crop collections – seed and clonal crops
3. Development
4. Costing study
5. Guiding principles
6. Risks
7. Management elements
8. Funding criteria
9. Decision process for funding eligibility

<u>A global partnership of organizations in research for a food secure future</u>

- Identify significant global development problems that science can help solve;
- Collect and organize knowledge related to these development problems;
- Develop research programs to fill the knowledge gaps to solve these development problems;
- Catalyze and lead putting research into practice, and policies and institutions into place, to solve these development problems;
- Lead monitoring and evaluation, share the lessons we learn and best practices we discover;
- Conserve, evaluate and share genetic diversity;
- Strengthen skills and knowledge in agricultural research for development around the world.

<u>The crop collections</u>

- 11 of the 15 centres maintain collections of 39 crops for a total of over 706,000 accessions
- Mainly seed crops but also vegetatively propagated crops such as banana, cassava, potato, sweet potato, and yam.

- Some forestry species at ICRAF
- Located around the world mainly where these crops originated from – centre of diversity

The CGIAR Crop Collection content

- 30 seed crops – 675,606 accessions – 9 Centres
  - Seed collections maintained at -18oC – low and stable cost. Monitoring of viability.
  - Safety duplication of seed collections at the Global Seed Vault Svalbard
  - Content – 59% constitute landraces and wild crop relatives
- 6 main clonal crops (plus fruit trees and Andean roots and tubers) – 35,962 accessions – 5 Centres
  - Cryopreservation – banana and plantain collection at Bioversity (100%) and cassava at CIAT and potato at CIP (but small %).
  - Field and *in vitro* – medium-term
  - *In vitro* – costly annual replacement

The Table below lists the crops, number of accessions in 2009 and the CGIAR centre responsible for the collection.

	Seed Crop	**No. of accessions**	**Centre**
1.	Barley	26,856	ICARDA
2.	Beans	35,903	CIAT
3.	Chickpea	20,267	ICRISAT – India
4.	Chickpea	13,462	ICARDA
5.	Chickpea	100	ICRISAT – Africa
6.	Cowpea	16,629	IITA
7.	Faba bean	9,181	ICARDA
8.	Forage & range plants	24,606	ICARDA
9.	Forages - tropical	18,291	ILRI
10.	Grasspea	3,210	ICARDA
11.	Groundnut	15,445	ICRISAT – India
12.	Groundnut	14,020	ICRISAT – Africa
13.	Legumes	4,346	IITA
14.	Lentils	11,008	ICARDA
15.	Maize	27,440	CIMMYT
16.	Maize	878	IITA
17.	Pea	6,075	ICARDA
18.	Pearl millet	22,211	ICRISAT – India
19.	Pearl millet	11,389	ICRISAT – Africa
20.	Pigeon pea	13,632	ICRISAT – India
21.	Pigeon pea	1,000	ICRISAT – Africa
22.	Rice	110,817	IRRI
23.	Rice	20,000	AfricaRice
24.	Small millets	10,235	ICRISAT – India
25.	Small millets	1,500	ICRISAT – Africa
26.	Sorghum	37,949	ICRISAT – India
27.	Sorghum	8,565	ICRISAT – Africa
28.	Tropical forages	23,140	CIAT
29.	Wheat	127,689	CIMMYT
30.	Wheat	39,762	ICARDA
	TOTAL seed crops	**675,606**	
	Clonal Crop	**accessions**	**Centre**
31.	Andean roots and tubers	1,174	CIP
32.	Banana, plantain	290	IITA
33.	Banana, plantain – Musa spp	1,298	Bioversity
34.	Cassava	6,592	CIAT
35.	Cassava	2,783	IITA
36.	Fruits trees – multipurpose	5,144	ICRAF
37.	Potato	7,213	CIP
38.	Sweet potato	8,108	CIP
39.	Yam	3,360	IITA
	TOTAL clonal crops	**35,962**	
	TOTAL BOTH crops	**711,568**	

Development

- 1994 – agreement with the FAO to put the collections in-trust for the benefit of the international community
- 1995 – Review the status and conditions of the collections
- 2001 – ITPGRFA and Article 15
- 2002-2005 – World Bank Funded project to upgrade the facilities

- 2006-2006 – Second Phase of World Bank funding to bring the collections to work together and build up the capacity as a system
- Main challenge – all independent centres with their own management differences and standards: transparency and accountability of budgets and activities was not easily achieved

2006-2009 Costing study

- Develop an agreed and adequate tool – Decision-Support Tool (DSC)
- Agree on the definition of costs and boundaries
- Agree on the basic custodianship operations – securing the genetic integrity of the germplasm and on the operations carried out that bring value to the materials – impact-focused
- CGIAR collections Costs - 2009 estimates:
- 15.2 M –for basic regular recurring functions
- 706,424 accessions
- 39 crops

The table below lists the centres and the total annual requirement for the long-term conservation of the crops and accessions listed in the table just above.

CGIAR Centre	**Total requirement USD - 2012**
AfricaRice	342,515
Bioversity	970,932
CIAT	2,394,585
CIMMYT	1,165,430
CIP	3,231,248
ICARDA	1,299,908
ICRISAT	2,464,419
IITA	1,130,621
ILRI	840,763
IRRI	1,393,625
Sub-total	**15,234,045**
Optimising collections	3,800,352
Regeneration project intro	1,994,564
TOTAL	**21,028,960**

Funding Guiding principles

Effective conservation system includes the following functions:

- Acquisition
- Storage / maintenance
- Safety duplication
- Regeneration / multiplication
- Characterisation
- Evaluation
- Documentation
- Distribution
- Promotion of use

NOTE: Essential but not cost vital:

- Gap analysis and collecting
- Molecular characterisation
- Evaluation of important traits
- Pre-breeding
- Training and technical backstopping
- Research on conservation methods
- Development of global information system
- Networking, international collaboration and facilitation
- Public awareness, conferences and visitor services

CGIAR Research Programme – CRP

- Existing institutes and facilities as the starting point
- Firmly based on sound scientific/technical principles in supportive political and social circumstances
- Increasing overall efficiency and development of common databases, reducing duplication, division of labour, harmonizing quality assurance standards and reporting, and strengthening collaboration
- Robust global conservation system with participation of all relevant institutions (not just those with long-term conservation roles)

CRP on the crop collections – objectives:

1. Diversity secure at perpetuity
2. Germplasm clean and available and disseminated
3. Informed and facilitated use
4. Diversity conservation is rationalised, cost effective and globalized

Rationalisation and optimization of collection:

- Eliminate unnecessary duplication
- Optimum long-term conservation methods (reducing need for regeneration and health testing)
- Harmonise information on accessions
- Improve outreach
- Quality Management System (QMS), operation manual and staff retention plan

Management elements

- Five-year strategic plan and budget for each centre
- Yearly work plans and budgets approved
- Annual programme and financial reports
- Annual certification of funds flow
- Annual meeting of all Centres to confer on technical and policy matters and address larger cross-cutting strategic and management issues
- Five-yearly comprehensive review of each Centre genebank

Risks

Major risks to the conservation and availability of plant genetic resources falls into 2 broad categories:

1. Funding – inadequate and unstable – the most serious and pervasive threats to genebank collections
2. Natural or political factors – natural disasters, civil unrest, political factors have threatened collections

Funding criteria

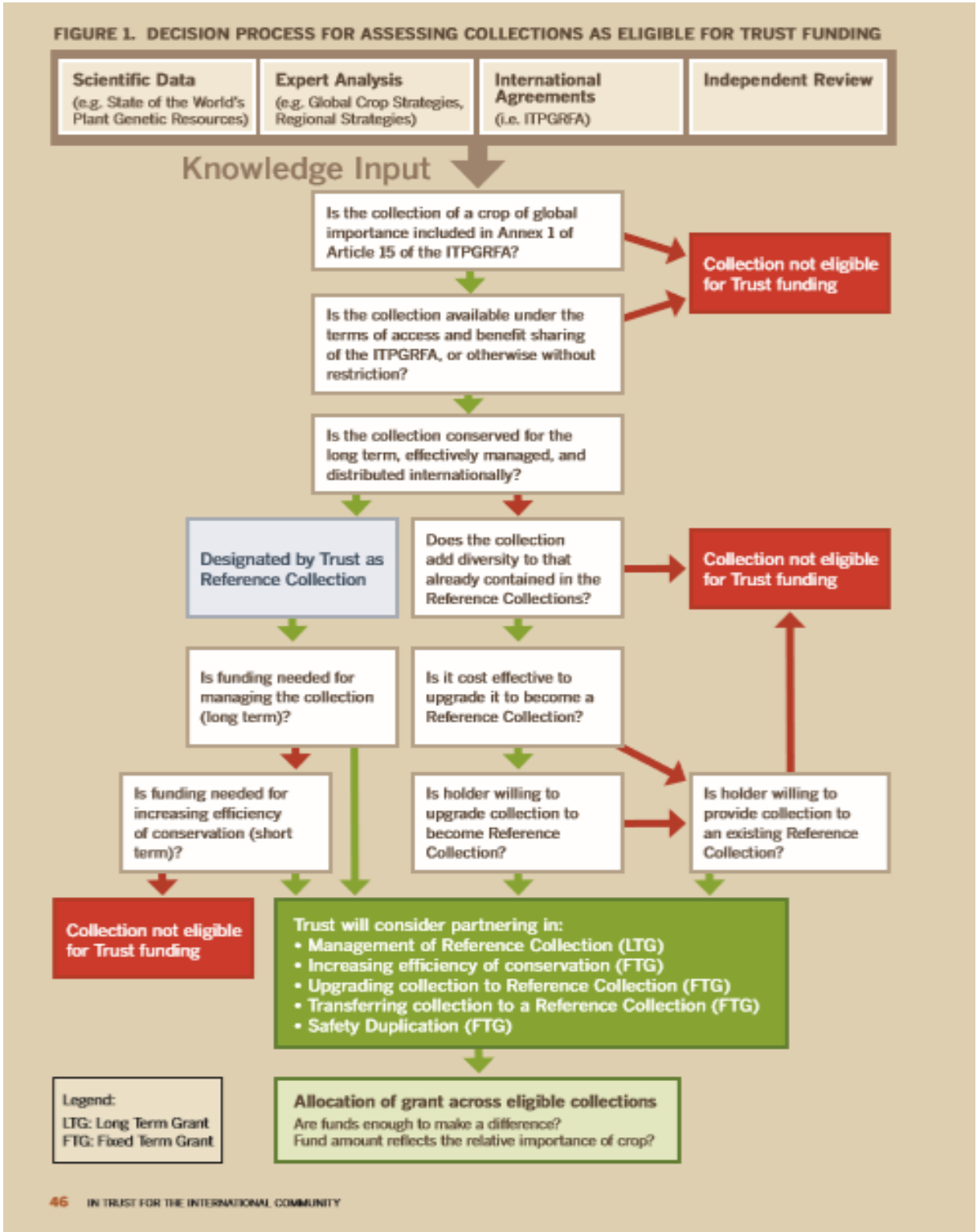
Basic principles that must be met for crop genetic resources collections to be eligible for support:

1. Of global importance – priority to crops included in Annex 1 of the ITPGRFA and Article 15.1 (b)
2. Accessible under internationally agreed terms of access and benefit sharing by the ITPGRFA and set out in the Standard Material Transfer Agreement (SMTA)
3. Holders commit themselves to long-term conservation and availability
4. Recipient of funds will work in partnership to develop an efficient and effective global conservation system

Specific criteria:

1. Recipient of funds has effective links with users
2. The genetic diversity is important or potentially important within the context of a rational global system of ex situ conservation
3. Legal status of the collection/institute enables meeting the conditions of access and benefit-sharing
4. Human resources and management systems needed to maintain the genetic resources and can demonstrate conformity with agreed scientific and technical standards of management
5. Facilities are adequate to ensure long-term conservation

Figure 1. Decision Process For Assessing Collections as Eligible for Trust Funding



Ref: CGIAR publication: In Trust for the International Community - Plan and partnership for managing and sustaining the CGIAR-held collections. The CGIAR CRP Research support. 2012.

4.4 THE UK NATIONAL FRUIT COLLECTION AS AN EXAMPLE

Paul Hadley presented the UK National Fruit Collection. It included the following points:

Decision-making processes for efficient genebank conservation and evaluation: The UK National Fruit Collection as an example

National Fruit Collections at Brogdale

- 3,500 accessions of fruit trees (principally apples, pears, plums, cherries)
- Maintained as a field collection
- Established 85 years ago
- Situated near Faversham in Kent
- Collection open to the public
- Online database (Chris Turnbull)
- Funded by DEFRA

Organisation

- Scientific curation – University of Reading
- Maintenance – Farm Advisory Services Team
- Public – Brogdale Collections
- Overseen by NFC Advisory Board
- UoR Management Committee
- Accession/Deaccession Sub-Committee
- National Fruit Collections Trust
- Supporters 'Club'

Collection Organisation

- Collections re-propagated approximately every 25 years
- Maintenance and curation activities separate from evaluation work (essential)
- Collections: 2 replications per accession
- Safety backup in Cryo
- Evaluation funded separately
- Typical of most live perennial collections in Europe

Accession/deaccession policy

- Essential for the efficient management of the collection
- Policy developed iteratively
- Accession…..

Accession policy

Find and introduce into the collection cultivars with proven:

- home garden utility
- increased yield
- higher 5-a-day nutrient content, (including fibre?)
- better 5-a-day nutrient preservation characteristics
- extended harvest periods
- disease resistance

- reduced pollinator dependence
- low winter chill requirements
- improved water stress tolerance
- Improved resilience to extreme weather events
- low input requirements

Confirmation of uniqueness prior to introduction to the NFC collections

Local/traditional varieties

- Any material offered to the NFC as traditional regional varieties will have to be accompanied by a contemporary published description highlighting traits of value.
- Varieties with value largely associated with their 'local' provenance should be considered firstly for inclusion in local variety collections with a view to recognizing these through 'virtual' inclusion in the NFC database.
- Where this is not available, the collection will be placed into the observation plot for assessment.

De-accession policy

- Duplicated accessions will generally be deaccessed.
- De-accession will generally be based upon a combination of robust genetic evidence and morphological analysis.
- Genetic evidence alone will be insufficient in the case of sports and clonal versions.
- Sports and clonal versions will be retained where potential further value can be demonstrated.
- De-accessed trees will generally be removed through the repropagation cycle.
- Where trees are to be removed a notice of deaccession will be placed on the website at least three months prior with the aim that interested parties are able to consult and potentially request graftwood if possible.

Evaluation

Climate change trial
- 25 cultivars (commercial, early/late, low/high chilling)
- 10 year duration (at least)
- Statistically robust
- Funded separately (NFC Trust)
- Platform for other research

Conclusions

- Important to separate curational and evaluation activities in any germplasm collection.
- A clear accession/deaccession policy is vital to the efficient development of a germplasm collection.

GENERAL DISCUSSION ON THE GSCC

The following point were mentioned and/or raised during the follow-up discussion:

- In addition to the 2 international collections at CATIE and CRC, the collection at USDA should be considered as a publically available collection.
- Cacao and the GSCC should learn from the CGIAR In-Trust materials and how they are managed and the use of the Standard Material Transfer Agreement.

- The question remains on how to ensure the conservation of materials that may not be a priority for the GSCC? The exclusion of materials needs to be done very carefully as it could include promising materials particularly vis a vis flavour.
- How to prioritise evaluation and link to accessions based on the main criteria and molecular and morphological characteristics. How to rationalise the evaluation needs?
- How to handle the characterisation of quality traits? Based on 2 replicates and verification.
- What need to do for cocoa to get there are:
    - Propagation
    - Removing duplicates
    - Fewer individuals but a higher standards
    - Should not need a large number of replicates
    - Industry information on traits from evaluation trials on a subset
- Subset of clones may be needed with basic information on robustness in experimental designs
- The public can provide a strong support for the conservation of the collections. The UK National Fruit Collection has a strong public awareness based on the fact that 40% of production is in home gardens. So there is public pressure to maintain the collection.
- How much information is available on the total apple diversity? And what is the collaboration with other centres that might contribute to sharing the responsibilities? Live collections can be compared looking at morphology.
- In the case of the UK Fruit collection, it took 15 years to secure the funding, 6 years to develop strategies and policies and gain confidence for long-term funding and 2 years of difficult discussions. It is important to never give up and this can be the main lesson to be drawn for cocoa.
- The key element in the case of the UK Fruit collection is that the evaluation work is separate from the conservation and there are 2 funding streams.
- In the case of cocoa, evaluation funds are often used for covering the cost of maintenance.

5. Characterisation and Evaluation of Key Traits of Importance to Users

The following sections cover the different traits and where we are on their evaluation. The objective is to provide information on the status of evaluation and how the information could be used to select and prioritise germplasm for inclusion in the GSCC.

5.1 Morphological characterisation and evaluation of traits of economic interest

Frances Bekele presented the morphological characterisation and evaluation of traits of economic interest. It included the following points:

Value

1. Identification of accessions
2. Assessment of phenotypic diversity (assignment to recognized groupings and identification of range of variation)
3. Detection of duplicates or mislabeled accessions
4. Preliminary evaluation of germplasm through assessment of traits of interest to breeders
5. Facilitation of the utilization of genetic resources within the genebank (through breeding & germplasm enhancement)

Morphological Descriptors used for characterisation at CRC (25)

FLOWER

1. Sepal length
2. Ligule width
3. Ovule number
4. Style length
5. Ligule colour
6. Filament colour
7. Pedicel colour

FRUIT and SEED (18)

1. Pod length, width
2. Wet bean weight (total)
3. Seed/Bean number
4. Cotyledon weight
5. Cotyledon length, width
6. Pod index
7. Mature pod colour (ridges)
8. Pod shape
9. Pod basal constriction
10. Pod apex form
11. Pod surface texture
12. Pod furrow disposition and separation
13. Pod hardness
14. Bean (cotyledon) colour and shape

Traits of Economic Interest

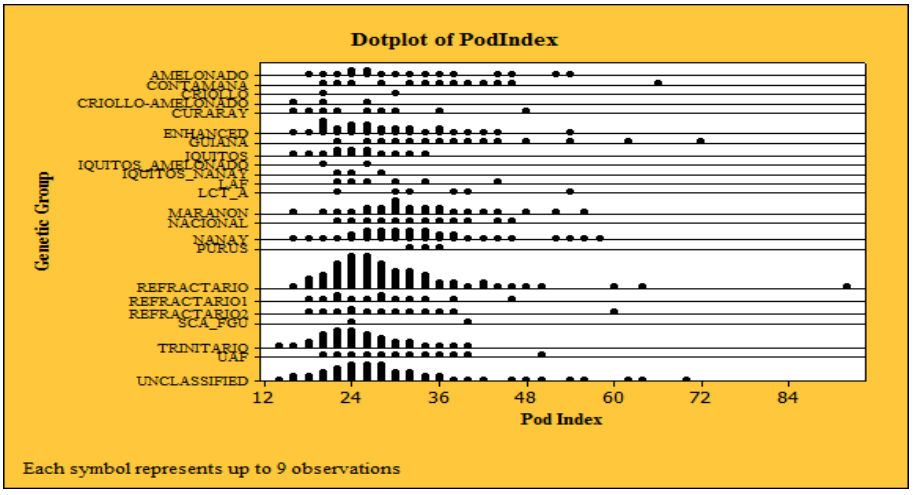
- Yield potential – Pod index (PI) (the number of pods required to produce 1 kg of dried cocoa)
- Cotyledon weight and size
- Seed/Bean number
- Disease and pest resistance
- Flavour, quality, butterfat content
- Abiotic stress resistance

Descriptive Statistics for 1979 accessions from the ICG,T

Variable	Mean and Standard Error (SE)	Coefficient of Variation	Minimum value	Maximum value
Cotyledon Weight (g)	0.984 ± 0.005	21.53	0.41	1.84 (UF 11)
Cotyledon Length (cm)	2.17 ± 0.004	8.87	1.37	2.72 (UF 11)
Cotyledon Width (cm)	1.22 ± 0.003	10.00	0.63	1.62 (JA 2/21)
Pod Index	28.04 ± 0.17	26.9	13.94 (UF 11)	92.76 (B 9/10-35)

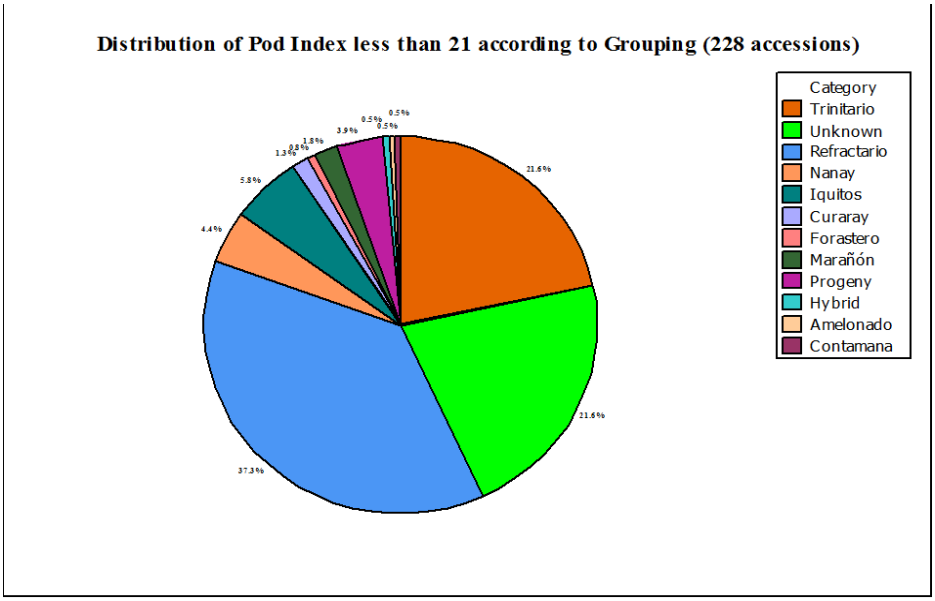
195 accessions have PI <20

Distribution of Pod Index by Genetic Group among 1979 accessions characterised at CRC



Descriptive Statistics for Pod Index for 1682 accessions

Group	N	Mean	SE Mean	StDev	Minimum	Maximum
"Criollo"	1	30.5	*	*	30.5	30.5
Amelonado	15	30.7	2.01	7.77	19.9	44.1
Contamana	19	36.9	2.34	10.18	20.3	66.1
Curaray	13	27.3	2.05	7.4	16.2	39.8
Forastero	29	26.6	1.18	6.35	16.5	44.6
Guiana	40	37.8	1.41	8.94	26.5	71.3
Hybrid	14	30.3	2.88	10.77	19.8	61.6
Iquitos	80	24.9	0.57	5.112	16	55.4
LA Forastero	10	33.6	2.98	9.43	24.1	54.1
LA Guianese Forastero	2 (ELP 40, 41)	36.1	0.02	0.03	36.1	36.1
Marañón	116	30.7	0.58	6.27	16.5	56.8
Nacional	23	33.3	1.39	6.66	24.4	46
Nanay	199	30.9	0.49	6.93	15.9	57.4
Enhanced germplasm	50	28.3	1.02	7.2	16.9	43.3
Purús	6	38.6	3.36	8.24	31.6	54.8
Refractario	620	27.5	0.29	7.14	15.5	92.8
Trinitario	223	25.1	0.35	5.22	13.9	43.3
Unknown	222	25.9	0.45	6.66	14.7	63



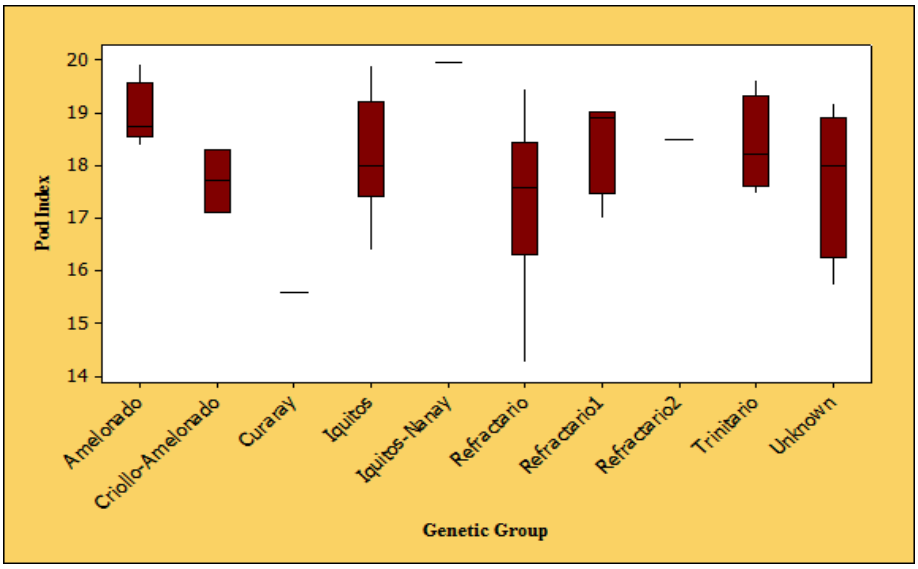
Best 81 Accessions (in terms of Pod Index value < 21) from Recognised Genetic Groups

Genetic Group	Count	% of Total
Amelonado	1	1.23
Contamana	1	1.23
Curaray	3	3.7
Iquitos	13	16.05
Marañón	4	4.94
Nanay	10	12.35
Trinitario	49	60.19
All	81	100

Note: No Purús, Criollo, Guiana and Nacional accessions

- Johnson, E.S., Bekele, F.L., Brown, S.J., Song, Q., Motamayor, J.C., Zhang, D., Meinhardt, L.W. and Schnell, R. J. (2009) Population structure and genetic diversity of the Trinitario cacao (*Theobroma cacao* L.) from Trinidad and Tobago. Crop Science 49: 564–572.
- Bekele, F.L., Bekele I., Butler, D.R.B. and Bidaisee, G.G. (2006). Patterns of morphological variation in a sample of cacao (Theobroma cacao L.) germplasm from the International Cocoa Genebank, Trinidad. Genetic Resources & Crop Evolution (Kluwer Academic Publishers, Netherlands) Volume 53 (5) (August 2006): 933-948.

Boxplot of accessions with favourable pod index (< 20) among 200 verified* accessions



**Research results of Lambert Motilal, CRC*

Conclusion

There is a good source of potentially high-yielding genotypes within the ICG,T across the range of genetic groups conserved. Selection of a diverse set of accessions for the GSCC should encompass accessions with favourable yield potential.

Future Direction

- Verify the authenticity of the trees from which data were collated through DNA fingerprinting (using molecular markers such as SNPs);
- Complete morphological characterization of uncharacterized and misidentified accessions;
- Compare phenotypic and genetic diversity of germplasm. Such a combined analysis will facilitate consideration of the value of morphological descriptors in the area of molecular genetics, and the list of morphological descriptors for routine characterisation within cacao collections may be modified;
- Pursue genomewide trait association studies (GWAS).

Acknowledgements for Funding

- The Government of The Republic of Trinidad & Tobago
- Cocoa Research Association (UK) Ltd
- RDI Fund, The UWI

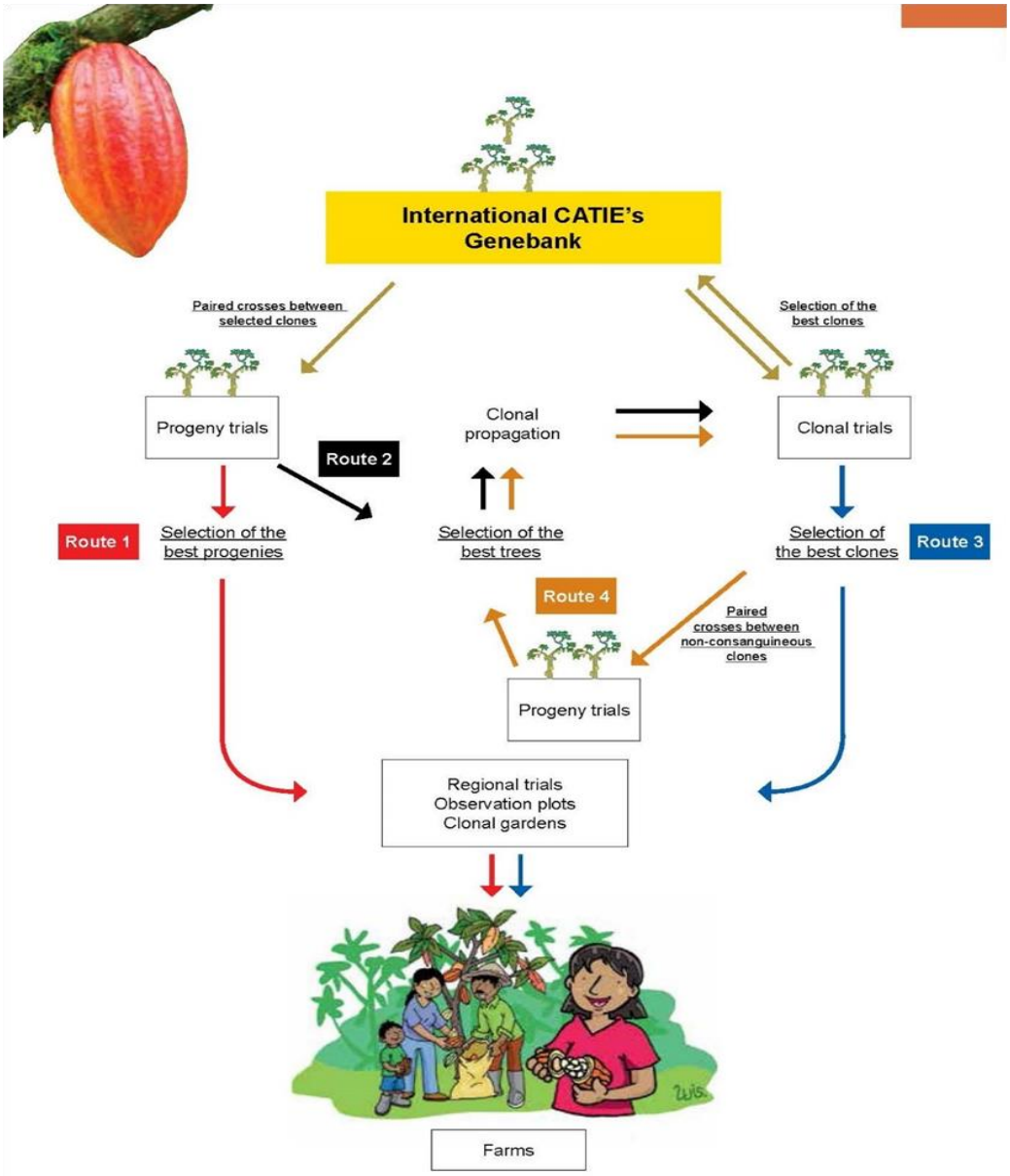
5.2 Evaluation of disease resistance traits with examples from CATIE

Wilbert Phillips presented the evaluation of disease resistance traits with examples from CATIE. It included the following points:

Frosty pod rot Up 90% pod losses and Black pod rot Up 20% pod losses

Evaluation of disease traits is a routine activity in our genebank and an essential part of our breeding strategy

A. Use of artificial inoculation methods to:
   A1. Rate the clones and identify sources of resistant in IC3.
   A2. Determine/check the reaction of selected genotypes.
B. Evaluation of natural incidence of moniliasis and black pod in our field trials.



A. Use of artificial inoculation methods to identify sources of resistant

- Categorization of confident sources of resistance is the first step in a breeding program that emphasizes disease resistance.
- To reach this goal, it is necessary to count with:
    - An adequate source of T. cacao variation.
    - Confident methodologies to select resistant genotypes.

CATIE accomplished both requirements:

- It hosts one of the two International Cacao Genebanks currently containing 1217 cacao clones.
- It implemented effective artificial methods of inoculation to test the reaction against frosty pod and black pod.

Artificial inoculation methods to indentify sources of disease resistance

- MONILIASIS (Moniliophthora roreri)
- BLACK POD (Phytophthora palmivora)

Supporting papers:

- Phillips M., W. 1986. Evaluación de la resistencia de cultivares de cacao (Theobroma cacao L.) a Moniliophthora roreri Cif. Par. IN Conf. Internacional de Investigación en cacao. 1987. Santo Domingo, República Dominicana.
- Phillips-Mora, W; Castillo, J; Krauss, U; Rodríguez, E and Wilkinson, M.J. 2005. Evaluation of cacao (Theobroma cacao) clones against seven Colombian isolates of Moniliophthora roreri from four pathogen genetic groups. Plant Pathology 54 (3): 483-490.

Advantages of using artificial inoculations methods

They permit to control:

- Pathogen stream.
- Concentration of inoculum.
- Type of propagules used.
- Age and maturity of the organ inoculated.
- Some environmental conditions such as humidity.

Biological aspects of the pathogen

- Level and distribución of genetic diversity.
- Main sources of variation.
- Predominant species (s) and stream.
- Main infective propagules.
- Conditions regulating the infective process.

P. capsici
P. megakarya
P. citrophthora
P. palmivora

Artificial inoculation method to test the reaction against Phytophthora spp.

The "Paper Disc Method was developed at CATIE in the late 80´s.

- Phillips M., W.; Galindo, J.J. 1989. Método de inoculación y evaluación de la resistencia a Phytophthora palmivora en frutos de cacao (Theobroma cacao L.). Turrialba (Costa Rica) 39(4):488-496.

Inoculation method

- Five month old pods are inoculated with a zoospore suspension (150.000 sp/ml) absorbed in a 1cm paper disc.

Evaluation

- 10 days after inoculation, the diameter of the lesion is measured.
- Clones are rated accordingly to this parameter.

Scoring

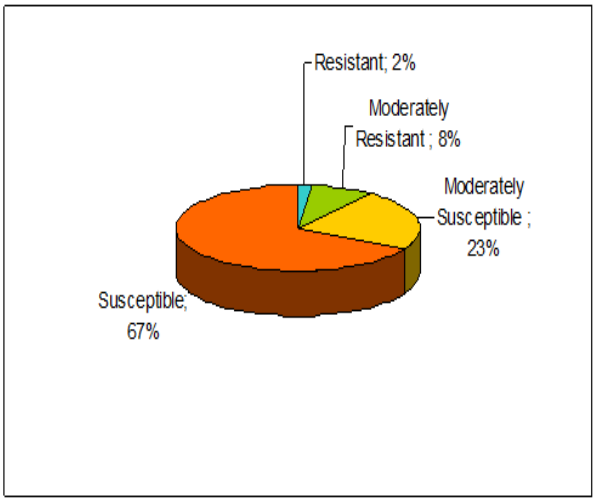
We determine the average severity based on at least 20 pods inoculated in different events and rate the clones using this scale:
Reaction / Internal severity (cm)

- Highly Resistant: 0.0 - 3.0
- Resistant: 3.1 - 6.0
- Moderately resistant: 6.1 - 9.0
- Moderately susceptible: 9.1 - 12.0
- Susceptible: 12.1 - 15.0
- Highly Susceptible: > 15.1

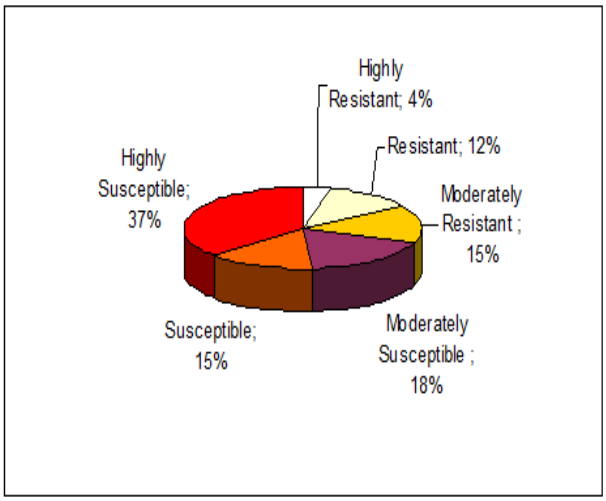
Resistant Clones to Black Pod as Determined Using Artificial Inoculations

- APA-5
- ARF (12, 14, 22, 24, 31, 32)
- BE-4
- Criollo-34
- EET (59, 272)
- ICS-47
- México-4A1
- ML-103
- Nacional-3A41
- PA (4, 51)
- PMCT (23, 35, 37, 46, 92, 93, 99)
- Pound-7
- RB-46
- SNK-12.

Reaction of 746 clones to M. *roreri*



Reaction of 819 clones to P. *palmivora*



Reaction of 819 clones to P. *palmivora*

New sources of resistance are immediately incorporated in the breeding activities

A. Resistant x Resistant genotypes to accumulate genes.
B. Resistant x High yielding, High quality or Precocious clones to accumulate desirable characters in single individuals.

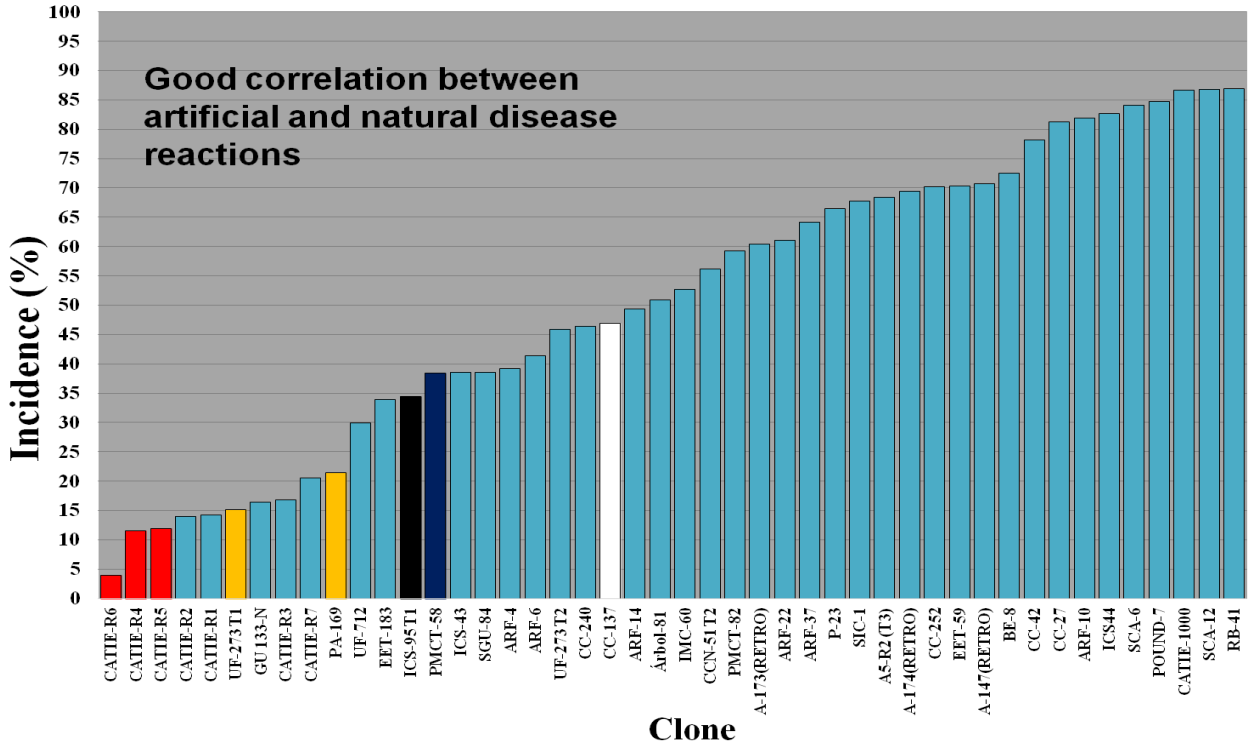
Uses of Artificial Inoculations at CATIE

- Evaluation/rating of clones from CATIE´s Genebank.
- Identification of highly resistant genotypes to be used in breeding and germplasm conservation activities.
- Virulence determination of selected isolates
- QTL studies

B. Routinely disease evaluation in CATIE´s field trials.

- We count the number of healthy and diseased pods per month and tree starting 2 years after planting.
- Data are typed in Excel spreadsheets and then corroborated.
- We calculated disease incidence and carry out data analyses.

Frosty pod incidence (avg 14 years)



5.3 Evaluation of Flavour and other quality traits

Darin Sukha presented the evaluation of flavour and other quality traits. It included the following points:

<u>Why do we need to evaluate flavour in selecting clones for the GSCC?</u>

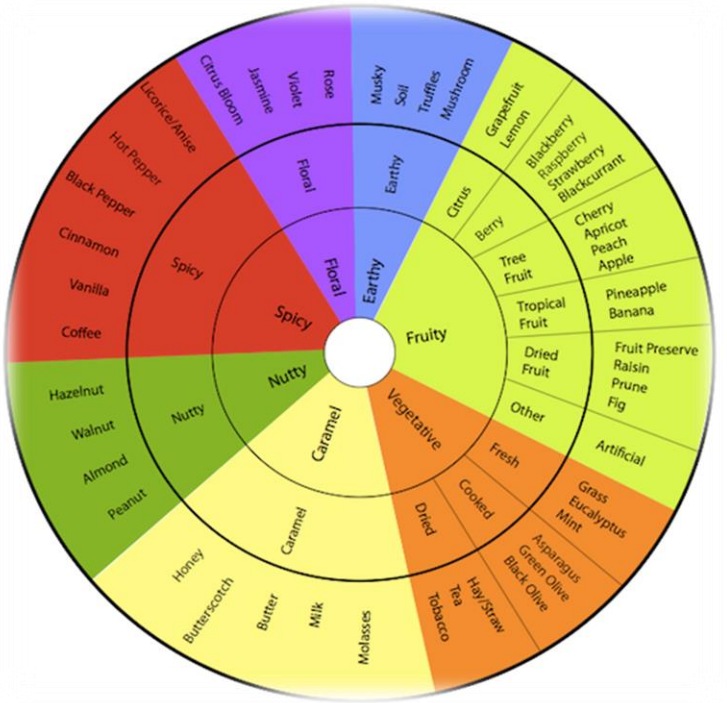
- Genetic flavour potential from each bean/variety.
- Rich flavour diversity we need to capture and preserve.

<u>Flavour Evaluation…</u>

"…a scientific method used to (1) Evoke, (2) Measure,(3)Analyse, and (4) Interpret those responses to products as perceived through the senses of sight, smell, touch, taste and hearing".

- Trained tasters: Panel and Individual
- Robust sensory design
    - Sample Coding
    - Randomization
    - Repetition
    - Flavour Descriptors

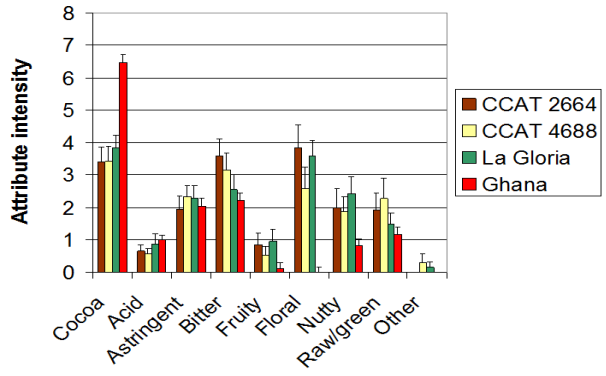
<u>Data analysis</u>



The spectrum of flavours in different cocoa types

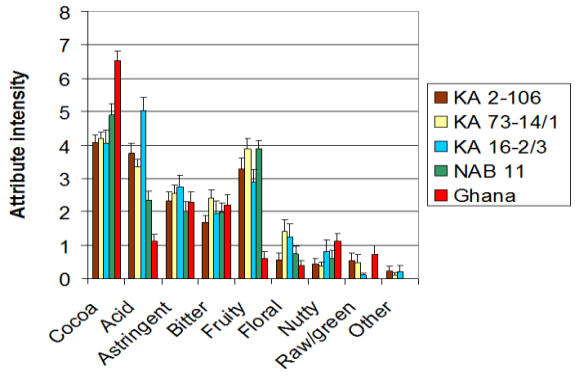
Ecuador vs Ghana

- Floral (sometimes aromatic but mainly herbal, “forest” green)
- Nutty (peanut skins)



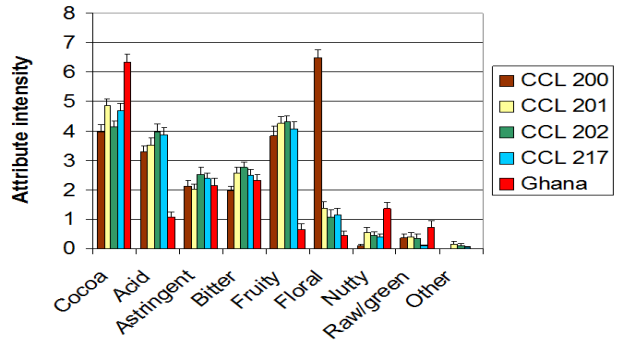
Papua New Guinea vs Ghana

- Acid
- Fruity (fresh fruit, banana)
- Floral (fragrant)
- Cocoa flavour at end



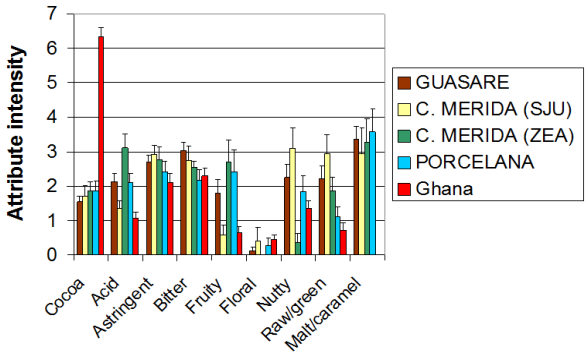
Trinidad and Tobago vs Ghana

- Fruity (brown dried fruit (raisin))
- Floral (fragrant)
- Acid (citric acid)

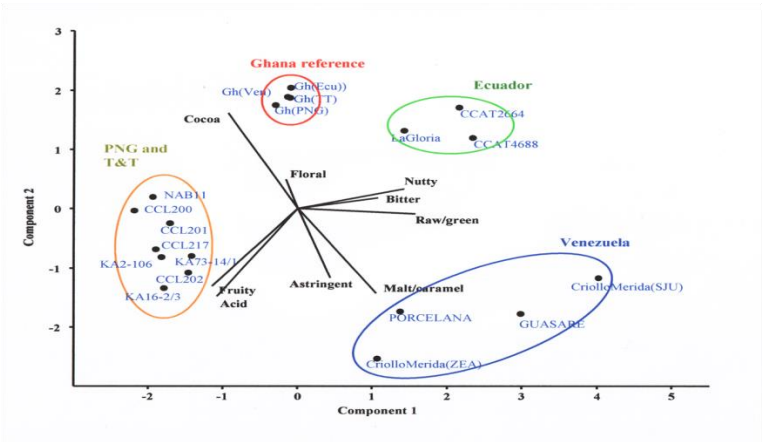


Venezuela vs Ghana

- Nutty (raw nuts)
- Caramel/fudge/malt (Panela)
- Raw/beany (green beans)



PCA plot of different country clones vs Ghana



Other quality traits include…

- Physical bean characteristics
- Cocoa butter%, FA profile and melting point
- Pulp flavour
- Fermentation requirements
- Nutraceuticals
    - Total phenolics
    - Theobromine
    - Caffeine
    - Catechins
    - Epicatechins
    - Flavanols
    - Antioxidant capacity

5.4 Screening for Yield Components and Resilience to Abiotic Stress

Paul Hadley made a presentation on screening for yield components and resilience to abiotic stress. It included the following points:

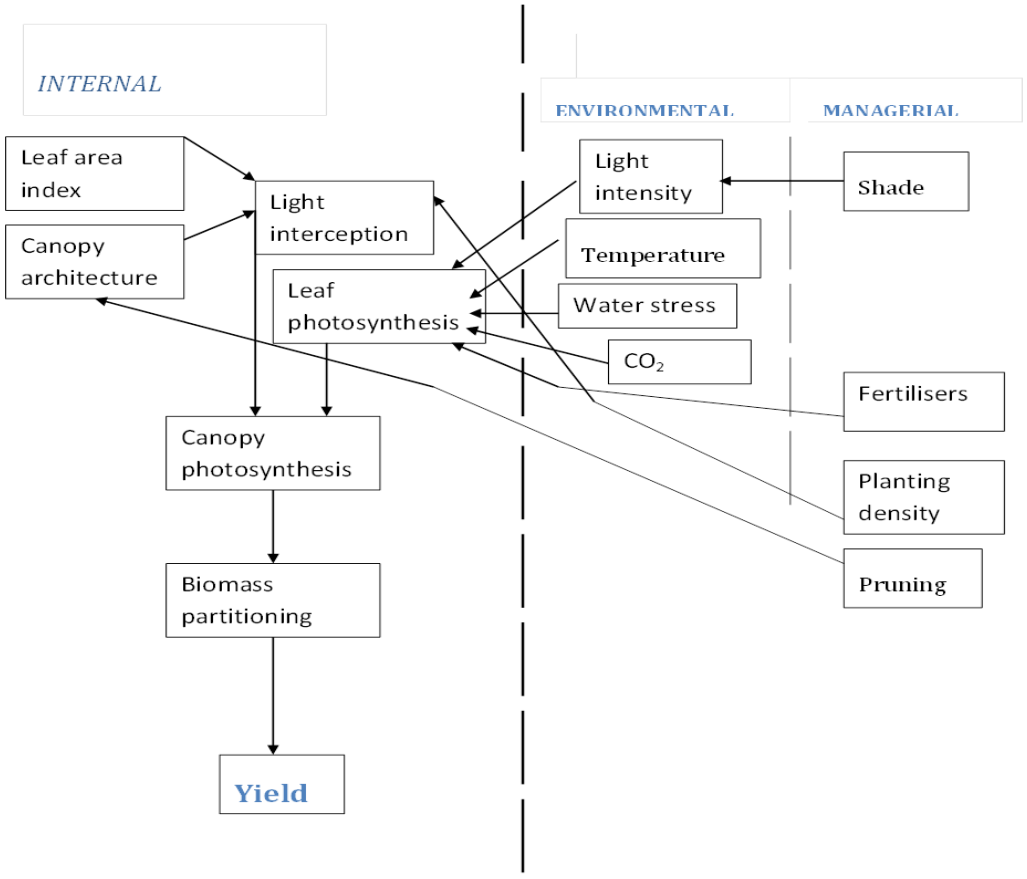


Figure 1: A schematic representation of determinants of yield in cocoa

Table 3. Comparison of studies of photosynthesis in cocoa

Internal factor	External factor	Photosynthetic rate (µmol m^{-2} s^{-1})	Reference
Canopy position		2.7 (sun leaves) 1.8 (shade leaves)	Murray, 1940
Leaf age		3.8	Lemée, 1955
Tree vigour		2.5-4.4	Hutcheon, 1977
	light	3.9	Raja Harun & Hardwick, 1988a
	temperature, water stress	0.8-1.8	Raja Harun & Hardwick, 1988b
Varietal	light, temperature, water stress	2.0-3.9	Balasimha *et al*., 1991
Leaf age	irradiance	2.0-6.5	Balasimha *et al*., 1992
Varietal		6.4-8.4	Yapp & Hadley, 1994
Varietal		2.6-3.7	Galyuon *et al*., 1996

Table 2: Comparison of studies of canopy characteristics.

Material	Proportion of radiation intercepted (I)	Leaf area index (L)	Light extinction coefficient (k)	Reference
Hybrid	0.90-0.97	3.5-5.7	0.62	Alvim (1977)
Clonal (7 clones)	0.72-0.94	2.3-3.4	0.61-0.95	Yapp & Hadley (1994)
Clonal (10 clones)	0.75-0.88	2.2-3.5	0.63-0.82	Daymond *et al*. (2002)

Table 1: Proposed visual assessment scale for light transmission to the ground under cocoa tree canopies (Eskes *et al.*, 2000)

Assessment scale	Light transmission
0	0% (no sunflecks)
1	1-5%
2	5-10%
3	10-20%
4	20-40%
5	>40%

Physiology with Environment

- Genotypic variation in photosynthetic rate
- Water use efficiency – genotypic variation in stomatal index
- Harvest index – yield partitioning
- Pod index
- Fat content
- Genotypic characteristics under different environmental regimes
  - climate change
  - drought
- Productivity and sustainability under stress
- Plasticity and stability of responses

General Discussion

The following points were mentioned and/or raised during the follow-up discussion:

- Stomatal index is simple to measure.
- Portable photosynthesis equipment can be used to make the first cut, i.e. the top 20% of varieties, to carry out further evaluation trials.

PROPOSED ACTIONS:

- Develop a short list of the most important characteristics to measure in the context of the GSCC and the equipments to be used. Paul Hadley and Andrew Daymond could do this. Could use the INGENIC newsletter to publish and consult.
- Need to develop a targeted programme to move forward, indentifying what can be done:
  - (1) in the genebank (characterisation)
  - (2) in evaluation trials

6. WHAT IS NEEDED FROM THE DIFFERENT GROUPS, PRIORITIES AND INTEREST FOR A GSCC

6.1 FROM THE INDUSTRY AND COCOAACTION

Martin Gilmour made a presentation on CocoaAction. It included the following points:

CocoaAction: Partnerships between governments, cocoa farmers, and the cocoa industry to boost productivity and strengthen community development in Côte d'Ivoire and Ghana.

Many of the slides are referenced World Cocoa Foundation (WCF).

Cocoa Sector Concerns

What needs to change?

- Numerous small, fragmented programs
- Many trained farmers, but most lack inputs (fertilizer and planting material) to drive production improvements
- Difficulties in measuring improvements at the farm level in a cost effective and consistent way
- Senior leaders in companies have not been involved as much as needed

CocoaAction

- Objective: To rejuvenate the cocoa sector at the farm/community level through closer voluntary industry and public-private collaboration, and to align sustainability efforts.
- Motivation: Cocoa farmers still tend to live in communities where poverty is prevalent, quality of life is low and cocoa yields are far below what they should be in order to secure a more prosperous future for the farmer and his/her family. As demand for chocolate rises across the globe, these factors call into question the viability of the cocoa supply chain.
- Uniting the forces: Despite the significant investment we have fallen short on achieving the required turnaround of the cocoa sector. Due to a lack of coordination and alignment, our individual projects do not have a weighty impact on the change needed in the sector.
- Set of common goals:
    - unprecedented, voluntary, industry-wide commitment to improve living conditions of cocoa farmers;
    - increase average yields;
    - strengthen value chain relationships;
    - work with existing industry and producing country programs;
    - strengthen cocoa growing communities.
- West Africa is at the center of our attention. Our effort will first focus on Côte d'Ivoire and Ghana and later expand to other cocoa producing countries.

As of May 20, 2014, the companies that have committed to CocoaAction are

- ADM
- Barry Callebaut
- Blommer
- Cargill
- ECOM Agrotrade Limited
- Ferrero
- The Hershey Company
- Mars, Incorporated
- Mondelēz International
- Nestlé
- Olam

Toward Rejuvenated Cocoa Sector

- A rejuvenated and economically viable cocoa sector, starting in Ghana and Côte d'Ivoire, that can compete with alternative crops and provide opportunities to cocoa farmers and cocoa
- Productivity package that allows: farmers are on the track to be professional farmers with at least a 100%-200% increased income.
- Indicators framework and cocoa reference standard, that helps process the strategy and assess the impact
- Community good practices packages that enable: Address the worst forms of child labor; education outcomes, gender parity and cocoa livelihoods to be improved
- Alignment with governments, donor and NGO engagement

Work streams - areas where industry can make biggest impact

- WS1: Planting breeding material and CSSV-resistant material
- WS2: Fertilizers
- WS3: Communities: align on approach to communities and community development including education, gender and child labor
- WS4: Aligned approach towards governments and donors
- WS5: Innovations/ future forms of extensions
- WS6A: Measuring what matters / indicators / aligning, reporting & shared learning
- WS6B: What is a viable and successful cocoa farmer business model?
- WS6C: Improve the effectiveness and efficiency of certification standards

Planting Material: Uniting Forces and Focus

- Companies with plant expertise are working together for the first time, and are looking to work with governments.
- Expertise already exists within the companies, e.g.:
    - Nestle: In Vitro propagation and orthotropic shoots
    - Mars: Grafting, genetics and breeding
    - Mondelez: CSSV and breeding
- Identified two areas of focus where we can add value:
    - Propagation: expand propagation capacity of origin countries to produce good quality trees
    - CSSV: the virus needs more attention and focus, e.g. on breeding new tolerant/resistant varieties and detection
- The companies will leverage the expertise within WCF and other programs to build models for distribution of improved planting material down to the farm-level.

CocoaAction's CSSV program consists of three subprojects

Subproject 1: Identification & screening of CSSV resistant material in Côte d'Ivoire and Ghana

- Part 1. Identification of existing CSSV resistant materials in national and international collections
- Part 2. Optimizing screening methods and screen materials for resistance to CSSV at one European and one Côte d'Ivoire site
- Part 3. Participative breeding: identifying CSSV resistant plants which could be used as parents

Subproject 2: Development of protection and detection measures

- Part 1: Development of protection methods against the mealybug (vector)
- Part 2: Development and local implementation of a robust detection test for the virus

Subproject 3: Support breeding for CSSV resistance in Côte d'Ivoire

- Development of breeding populations using selected parents, identification of resistant genotypes and development of markers for CSSV tolerance.
- Note: The program will work towards maintaining current CDI quality and flavour

Subproject 1: Identification & screening of CSSV resistant material in Côte d'Ivoire and Ghana

1. Identify CSSV resistant genotypes in different collections
    - Reading
    - CATIE
    - Cote d'Ivoire
    - Ghana
    - Togo
    - Trinidad
2. Introduce them into the quarantine of Reading University
3. Screening and validation of resistant genotypes
4. Provide resistant budwood or SE plants to regional institutes

Subproject 2: Development of protection and detection measures

1. Develop a test recognizing all strains of CSSV
2. Industrialize the test
3. Transfer the test to partners in Côte d'Ivoire

CocoaAction and Cacao Genetic Resources

- A few members currently support the international collection at CRC Trinidad, and the International Cacao Quarantine Centre at University of Reading.
- Via CacaoNet, WCC, WCF, we are promoting the importance and vulnerability of cacao genetic resources.
- CocoaAction has identified CSSV disease management as a priority, to meet predicted consumption needs.
- Resistance to CSSV disease is essential as one of the selection criteria for the GSCC.
- Opportunity to use CSSV disease resistance as an example of the importance of cacao genetic resources.
- Additional work needed on developing a screen for CSSV resistance, characterizing what resistance is.

Understanding the extent of molecular diversity of the complex of viral species responsible for Cacao Swollen Shoot disease in order to improve CSSV detection and identify genuine alternative host plants. (ECA/Caobisco Cocoa Productivity & Quality WG)

- Emmanuelle Muller (CIRAD), Andy Wetten (University of Reading) & Joël Allainguillaume (University of the West of England). CRIG, CNRA, and CRIN.
- Employ NGS profiles of the entire complement of CSSV species in T. cacao and putative alternative host plants to improve the molecular diagnostic for the disease.
- Characterise the geographical distribution in Côte d'Ivoire, Ghana and Nigeria of viral species responsible for CSSD.
- Through the use of mealybug transmitted CSSV isolates identify genuine alternative hosts of CSSV and confirm the 'non-host' status of species proposed for use as barrier crops (prioritising citrus, coffee and rubber). In this way non-infective plant species can be eliminated from inclusion in proposed cutting-out programmes.
- 2 years, €200

World Cocoa Foundation: Cocoa Swollen Shoot Virus (CSSV) disease Program Manager Abidjan, Côte d'Ivoire www.worldcocoafoundation.org

- WCF seeks a (Bilingual – French and English) Cocoa Swollen Shoot Virus (CSSV) disease Program Manager to lead a multicomponent R&D program aimed at developing tools for the control of CSSV and the protection of cocoa farms. The CSSV program is part of the CocoaAction strategy aimed at accelerating the long-term sustainability of cocoa farmers and the cocoa sector generally in West Africa.
- The WCF CSSV program is part of this priority area which aims at making better planting material available to farmers in order to improve productivity and farmer income. It is a five-year program, focusing on three specific areas: 1) identification, screening and sharing of existing resistant materials; 2) optimization of CSSV detection and protection against the disease; and, 3) Supporting breeding for CSSV resistance in Côte d'Ivoire.
- The position is based in Abidjan, Côte d'Ivoire. Candidates must possess proven leadership, a track record of technical project management in a cocoa producing country, experience working on both the public and private sector levels, and fluent oral and written language skill levels in both English and French. The candidate must also bring experience developing collaborative partnerships with both public and private sector stakeholders.

GENERAL DISCUSSION

The following point were mentioned and/or raised during the follow-up discussion:

- What should be the approach considering that CATIE and CRC do not have CSSV:
    - (1) Possible to identify materials in genebanks for multiplication and spread
    - (2) Identify the strains of CSSV and interactions
- Need to develop a test looking at different varieties, looking at genotypes with different varieties, looking also at the associated mealy bugs transmitting CSSV.
- The characterisation of the virus will also be important.
- A case study on CSSV can be used to raise the awareness of the industry to the importance of genebanks and collections as the long-term solutions to the industry problems.
- What about other objectives such as witches broom? CSSV is the largest threat now and there are no solutions. But this does not preclude work to be carried out on a number of priorities.
- How to become a partner in this? The CSSV Programme is still in draft form and will start in early 2015.

6.2 From the Fine Chocolate Industry

Ed Seguine made a presentation on the point of view from the Fine Chocolate Industry Association (FCIA) and the Heirloom Cacao Preservation Initiative (HCP). It included the following points:

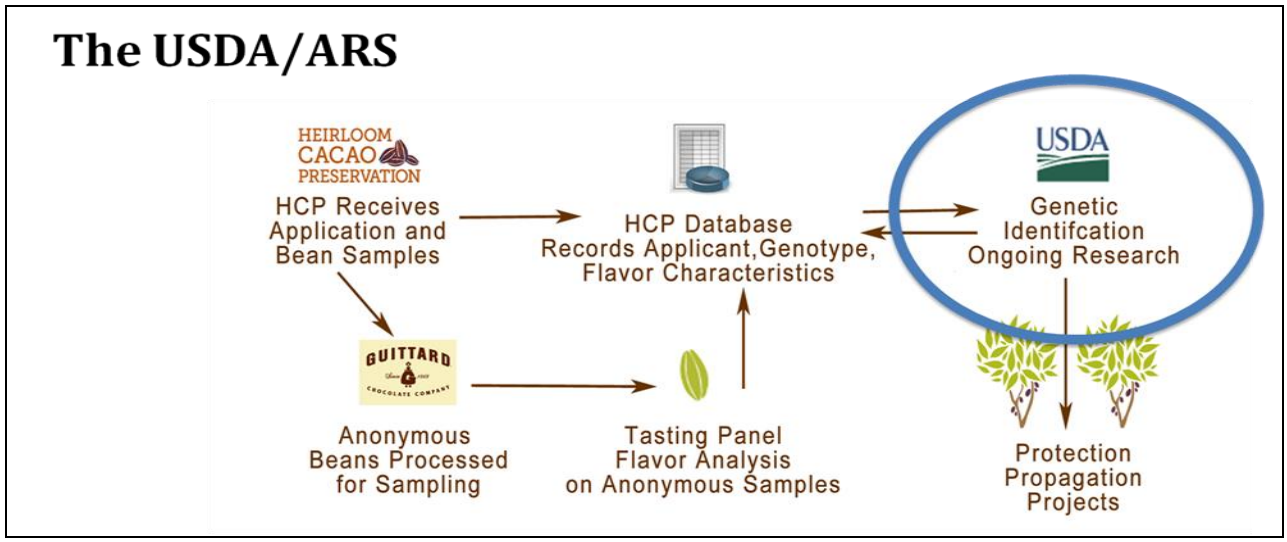
Breeding for Flavour and Agronomic Traits: You have to have it in the first place in order to have it in the end—but you need to pay attention to it!

Sector issues

- Yields have not changed
- Diseases continue to devastate
- Aging trees / exhausted soils
- Major quality declines all origins
- Not a career future
- Major volume needs in next 10 yrs. (BRICS countries)
- Preserve flavour

Our breeding track record is not so good where flavour is concerned--

- Tomatoes and strawberries that look perfect but have no flavour and roses with no perfume



Breeding programs get what they measure

- Flavour must be included from the beginning of a breeding program
- If you don't measure it You will lose it

Started 15 years ago

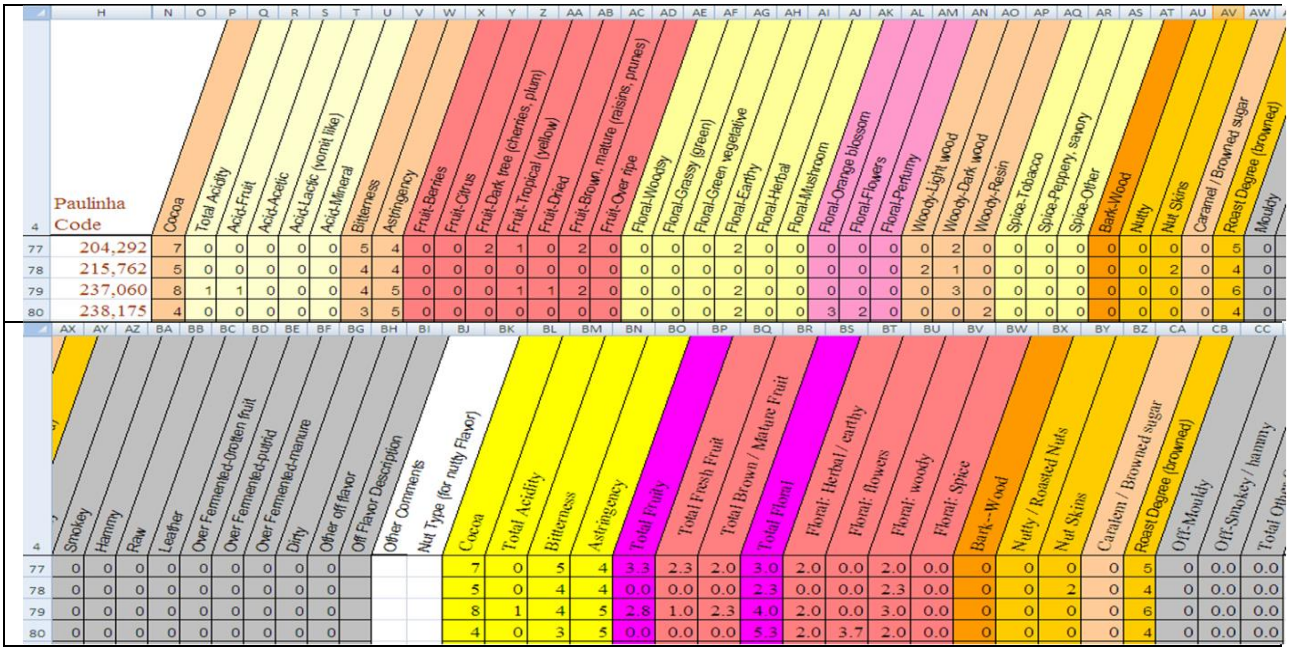
- Wilberth Phillip CATIE Breeding / Selecting
    - Adriana Arciniega
    - Allan Mata
    - Juan Carlos Motamayor (Mars)
    - Ray Schnell (USDA, now Mars)
- Flavour Evaluation and Selection—Ed Seguine (Guittard Chocolate)

Moniliophthora rorei—fungal disease

Economic losses due to Frosty Pod

- Costa Rica 1978 - exports reduced by 92% - 7000 ha of farms abandoned
- Mexico 2005 - 50% loss to disease
- Peru 1988 - 16,500 ha (50% of cacao producing areas) abandoned
- Bolivia 2011 - Construction of oil pipeline

The Taster - 55 Flavour Attributes



Paulinha Code	Cocoa	Total Acidity	Acid-Fruit	Acid-Acetic	Acid-Lactic (vomit like)	Acid-Mineral	Bitterness	Astringency	Fruit-Berries	Fruit-Citrus	Fruit-Dark tree (cherries, plum)	Fruit-Tropical (yellow)	Fruit-Dried	Fruit-Brown; mature (raisins, prunes)	Fruit-Over ripe	Floral-Woodsy	Floral-Grassy (green)	Floral-Green vegetative	Floral-Earthy	Floral-Herbal	Floral-Mushroom	Floral-Orange blossom	Floral-Flowers	Floral-Perfumy	Woody-Light wood	Woody-Dark wood	Woody-Resin	Spice-Tobacco	Spice-Peppery, savory	Spice-Other	Bark-Wood	Nutty	Nut Skins	Caramel / Browned sugar	Roast Degree (browned)	Mouldy
204,292	7	0	0	0	0	0	5	4	0	0	2	1	0	2	0	0	0	0	2	0	0	0	0	0	0	2	0	0	0	0	0	0	0	0	5	0
215,762	5	0	0	0	0	0	4	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	1	0	0	0	0	0	0	2	0	4	0
237,060	8	1	1	0	0	0	4	5	0	0	0	1	1	2	0	0	0	0	2	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	6	0
238,175	4	0	0	0	0	0	3	5	0	0	0	0	0	0	0	0	0	0	2	0	0	3	2	0	0	0	2	0	0	0	0	0	0	0	4	0

Smokey	Hammy	Raw	Leather	Over Fermented-Rotten fruit	Over Fermented-putrid	Over Fermented-manure	Dirty	Other off flavor	Off Flavor Description	Other Comments	Nut Type (for nutty Flavor)	Cocoa	Total Acidity	Bitterness	Astringency	Total Fruity	Total Fresh Fruit	Total Brown / Mature Fruit	Total Floral	Floral: Herbal / earthy	Floral: flowers	Floral: woody	Floral: Spice	Bark--Wood	Nutty / Roasted Nuts	Nut Skins	Caramel / Browned sugar	Roast Degree (browned)	Off-Muddy	Off-Smokey / hammy	Total Other
0	0	0	0	0	0	0	0	0				7	0	5	4	3.3	2.3	2.0	3.0	2.0	0.0	2.0	0.0	0	0	0	0	5	0	0.0	0.0
0	0	0	0	0	0	0	0	0				5	0	4	4	0.0	0.0	0.0	2.3	0.0	0.0	2.3	0.0	0	0	2	0	4	0	0.0	0.0
0	0	0	0	0	0	0	0	0				8	1	4	5	2.8	1.0	2.3	4.0	2.0	0.0	3.0	0.0	0	0	0	0	6	0	0.0	0.0
0	0	0	0	0	0	0	0	0				4	0	3	5	0.0	0.0	0.0	5.3	2.0	3.7	2.0	0.0	0	0	0	0	4	0	0.0	0.0

Descriptive Flavour (example of descriptive flavour)

ESS Liquor Evaluation	**ESS Chocolate Evaluation**
Pronounced cocoa intensity with mild acidity primarily fruit with a trace of acetic. Normal bitterness and slightly increased astringency. Lots of fresh fruit notes--citric, red, and tropical. Clean finish.	Color: rich brown with slight reddish hues Flavour: Smooth, moderate chocolate, slightly tart with a fruit tartness. Fresh fruit notes with slight citric and red and tropical notes. Moderate intensity finish with moderate chocolate with mild fruit. Clean profile. Bright.
Low to moderate chocolate with higher than normal bitterness and higher astringency. Overall character is a very dirty note.	Color: Moderate brown Flavour: Slightly astringent up front with lower chocolate notes. Rougher and not attractive. Slightly barky and very dirty end.
Improved cocoa intensity and lower in bitterness and astringency than 7117 but still has a not very clean finish.	Color: Similar to 7116 Flavour: slightly flat in flavour with flatter chocolate notes. The 7116 is clearly the most interesting chocolate of the three.

Final Direction – traffic lights

- Green: One or more of the suite of valued cacao flavours (cocoa, balanced bitterness/astringency, fruit, floral, nut
- Red: Problematic inferior flavour—dirty, badly out of balance astringency—remove from breeding unless it "walks on water"—let's talk about it

CATIE Released Clones

Clone	Yield Kg/ha-yr	Monilia Losses	Black Pod Losses	Flavour
R-1	1,674	15%	6%	Fresh fruit note, smooth balanced cocoa, mild bitterness
R-4	2,070	12%	1%	Bright fresh fruit, moderate cocoa, complex blend with slight browned fruit notes • Salon du Chocolat, Paris, Cocoa of Excellence winner, 2009
R-6	2,363	4%	0%	Fresh and browned fruit notes, dark wood notes, rich chocolate • Salon du Chocolat, Paris, Cocoa of Excellence winner, 2009

CATIE Released Clones—Fermentation Test

Clone	3 days ferm	5 days ferm	6 days ferm
CATIE-R1	3539 Harsh, astringent, low chocolate flavour. Not a good fermentation for this bean.	3540 Trace of acidity but this has a very good, full bodied chocolate flavour. The late taste seems to have a nice dried fruit / browned fruit (dark raisin) note along with some woody, dark wood floral notes. Very good overall flavour.	3541 Not as clean a flavour. The trace of acidity has shifted from fruity to more acetic in character. The chocolate is there but is more muddied and it has more of a bark wood note. The fruit flavour is depressed though it is slightly there
CATIE-R4	3542 Slight acidic notes that are more acetic than fruity. Astringency is present but not severe. Not a lot of chocolate flavour but not bad either. This is just sort of OK. Aftertaste has much more astringency present along with a muddy flavour.	3543 Beautiful color. Flavour is slightly on the bitter and astringent side. Has a dark wood and some bark wood flavour characters. The middle taste has a trace of acidity that gives rise to some dark fruit notes and a slight raisin not. Chocolate flavour is good but not dominant.	3544 Good chocolate flavour profile from the beginning. Has some dark wood notes though not quite as much as in the 5 day fermentation but this is offset by more chocolate. The bitterness is a clean bitter and is balanced with the astringency.
CATIE-R6	3545 Initial astringency emerges in this sample with some but much lower cocoa notes. Not a chocolate flavour but more of a cocoa note with bitter and astringent being the dominant features. Some slight wood notes. Ends bitter and astringent	3546 Rich color. Smooth initial flavour profile. As it progresses, a rich chocolate character emerges. Very good chocolate base here. Some trace notes of dark wood with low bitterness and relatively low astringency. This is an excellent base bean.	3547 Slightly more bitterness in this sample it is a more direct chocolate hit with the dark wood notes shifting to a more bark wood character. It is a "darker" and deeper overall flavour character but is still good.

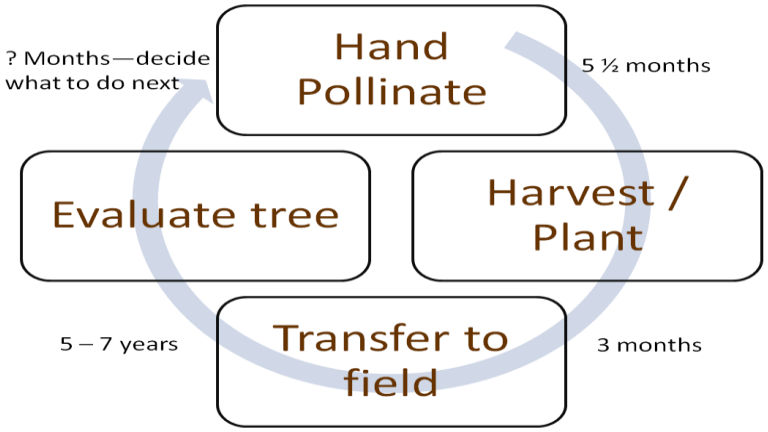
Clone	3 days ferm	5 days ferm	6 days ferm
CC-137	3548 Clearly lighter in color than the 5 day fermentation sample. This is more nutty but of a raw, more astringent nut. Astringency much, much stronger in this sample.	3549 Light color, thick liquor. This has a very interesting nutty flavour profile with no acidity, mild bitterness and moderate astringency but more of a nut-skins astringency. Some light wood notes. Very good tasting.	3550 Color seems slightly greyer. Flavour has lost some of the fresh roasted nut character of the 5 day fermentation and is more bitter with some bark wood notes and much stronger nut-skins astringency.
PMCT-58	3551 Not a good fermentation for this sample. Astringent and bitter, low / almost no chocolate flavour.	3552 Much darker in color. Initial taste is a very deep, persistent chocolate with some increased bitterness and mild astringency. Has bark wood notes along with slight dark fruit notes. Some earthy character also. This seems to be a deeper overall character and more blunt chocolate than R1, R4, and R6	3553 Thinner viscosity. Very similar to the 5 day sample. Maybe a bit better even but both are good. This is definitely a base bean. Roast on this sample seems to be a fuller roast than the other samples.

Status of Markers for Flavour

- No validated markers associated with key flavour attributes
- No specific programs in place for marker identification
- Company programs (G12 Cocoa Action) focused on productivity / disease resistance
- Freddy Amores--INIAP
- Breeding— Juan Carlos Motamayor (Mars) and Ray Schnell (USDA, now Mars)
- End breeding flavour evaluation—Ed Seguine (Mars)
    - EET 544
    - EET 558
    - EET 575
    - EET 576
- Highly precocious breeding in crosses between heterotic groups
- New cultivars at 1.5 (left) and 2.5 (right) years old in Ecuador

Breeding is a numbers game

- Combine – “Stacked Traits”
    - Black pod resistance
    - Pollen self-compatibility
    - High fat content
    - CSSV, cocoa swollen shoot virus, resistance
    - High cocoa flavour
- Chances are about 1 in 1200
- Maybe plant 12,000 trees to get 10 to be “sure” to win the bet?



Ghana—65% Cacao (from Guittard Chocolate)

- Color: Rich, warm brown.
- Aroma: Clear, direct, intense CHOCOLATE.
- Flavour: The chocolate that keeps on giving! Quick rise in chocolate that is exactly like the aroma. Character and intensity is maintained from beginning—to middle—and on to the end—and well into the aftertaste—it lasts long after the chocolate is gone from the mouth.

Ecuador Nacional—65% Cacao (from Guittard Chocolate)

- Color: Deep, darker brown.
- Aroma: Very mild chocolate aroma with a trace "woodsy" character.
- Flavour: Classic profile of an Ecuador Nacional. Mild initial flavour that builds in the mouth but goes deep. Late initial / middle taste presents green grass and mild vegetative notes along with floral flowers with orange blossom notes. Some dark wood and wood resin as well as some moderate astringency that deepens the profile—A deep flavour profile.
- Ecuador INIAP
- Released Clones: EET 544, EET 575, EET 576
- Roast: 121ºC x 23 min (Binder convection oven)
- Source: La Victoria Farm, Ecuador at scale planting, fermented by Freddy Amores, INIAP

INIAP Clone Blend—65% Cacao (from Seguine Cacao)

- Ecuador INIAP
- Released Clones: EET 544, EET 575, EET 576
- Roast: 121ºC x 23 min (Binder convection oven)
- Source: La Victoria Farm, Ecuador at scale planting, fermented by Freddy Amores, INIAP
- Color: Deep, dark brown (typical of Ecuador).
- Aroma: Mild chocolate with dark wood and mild green notes.
- Flavour: Mild chocolate with smooth, velvety astringency. Dark wood with green vegetative and trace green cut grass floral notes. Shifts to a mild herbal-like character. Finish is relatively short with chocolate fading quickly leaving a mild green character and astringent notes.

CATIE Blend—R-1, R-4, R-6, PMCT 58 (from Seguine Cacao)

- Color: light to medium brown hue with slight yellowish hues.
- Aroma: Mild chocolate, mild tropical yellow fruit with balsamic notes.

- Flavour: Early chocolate blended with smooth fruit and balsamic acidity. Has deeper overall notes. Some mild tart tropical fruits along with deep browned notes that are a fermented browned fruit, with clear balsamic notes. Finish is smooth chocolate. Mild astringency with a smooth profile. Very balanced and well blended overall.

You CAN have your flavour— But only if you INCLUDE it!

- *Theobroma cacao* – Food of the Gods

GENERAL DISCUSSION

The following point were mentioned and/or raised during the follow-up discussion:

- The "traffic light" approach is easy but requires judgement. It is mainly about assessing how it would measure if processed into a blend.
- What is important is high quality varieties with interesting traits.
- The interactions between processing and inherited quality is key.
- How to manage quality in a breeding programme? Need a priori knowledge on how it needs to be processed, considering fermentation and drying protocols and screening to select potential value.
- How to address small-scale panel for flavour evaluation? Need a holistic approach to flavour.
- Needs and opportunities to combine robustness and flavour. This can be assessed in public domain collections at CATIE, CRC and USDA.

PROPOSED ACTIONS:

- CacaoNet could produce standard protocols for flavour evaluation – Ed Seguine and Darin Sukha could publish a short paper to be used with samples.
- Could assess/process the different genetic groups at CRC as a first step to understand the differences.

6.3 From the Regional Cocoa Breeders Groups

6.3.1 West African Cocoa Breeders Group

Désiré Pokou represented the West Africa Breeders Group members in the CacaoNet meeting. He presented the following key points verbally:

- There are many introductions of cacao genotypes from international quarantine to increase new diversity of breeders' germplasm collection. But little is known about the allelic richness that are bringing in. there is a need to know what the different collections of NARS have, what have been added and and what is the need for the future in terms of diversity. The situation is uneven between the countries and between the farmers and research germplasm. Field Evaluation is very costly. It has been agreed in the scope of USIAD/USDA/MARS project to split key clones for evaluation in different countries using the same standard.
- A good example of application of markers in seed garden output is ongoing under the WCF-ACI project to check the conformity of parental clones in order to eliminate off-types from crossing and also check the fidelity of crosses in seed garden t.
- There is an important issue of misidentification in seed gardens. The -WCF-ACI project has funded the analysis using SNP markers of 2,000 samples per country with the aim of resolving this issue and

is at the data analysis stage now. The next West Africa Breeding group meeting will discuss how to address the results.

- In summary, there is a need to highlight the gaps in diversity between local collections and clones of interest identified in the international collections.

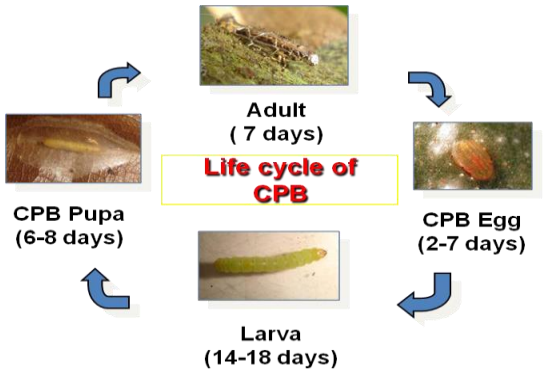
6.3.2 Asia and Pacific Cocoa Breeders Group

Haya Ramba is the Chair of the Asia and Pacific Cocoa Breeders Group and represented the members in the CacaoNet meeting. He made a presentation also on behalf of Sapirah Bakar with the following key points.

<u>Screening cocoa clones for cocoa pod borer tolerance</u>

- Cocoa Pod Borer (CPB) Conopomorpha cramerella Snellen
- Lepidoptera: Gracillariidae
- The most serious pest of cocoa in Malaysia. Damage is caused by larvae that bore into pods. Inside the pod larvae mine and feed within the placenta and in the interbean spaces. Attacks on young pods may result in the malformed and clumping of beans making them unextractable during harvest (Azhar et al., 1995) and low quality beans (*Tay, 1982*).
- 1980 - Tawau region, 5000 ha.
- 1986 - Malacca, 700 ha

<u>Malaysia, Indonesia, Philippines, Papua New Guinea (PNG)</u>



- Mean total days from egg to adult emergence 27.6 days (*Lim, GT et al., 1982*)

<u>Methods of CPB management</u>

- Chemical control
  - Insecticides – regular application, high cost and develop insect resistant to chemical
- Biological control (ants and egg parasitoids)
- Host plant resistance (HPR)
- Cultural and mechanical control (rampasan, pod sleeving)
- Integrated pest management (IPM)

(Azhar, 2000; Anon, 2008)

Definition of Tolerance

- Tolerance is a 'defence' mechanism by which plants have the ability to recover from attacks of insects and produce an acceptable yield despite supporting an insect population that would cause more damage to susceptible plants of the same species (*Kogan, 1975*).
- However, under natural field conditions, the effect of tolerance is very difficult to detect among plant genotypes because it often occurs in combination with other resistance mechanism for the ultimate expression of resistance (*Horber, 1980; Smith, 1989; Panda and Khush, 1995*).
- In view of this, the mechanism of resistance cannot unequivocally be assigned to specific categories, and this could explain why the tolerance mechanism is often confused with moderate or low resistance (*Horber, 1980; Smith, 1989*).
- In cocoa, tolerance to CPB is measured by comparing their respective damage as wet beans losses (*Lim and Phua, 1986*) or an average damage severity index (*Azhar, 1988; Azhar et al., 1995). The observed differences* in damage among clones indicate the degree of pod tolerance to CPB attack**.**

Screening and selecting resistance cocoa clones

- Various approaches have been attempted to select and screen cocoa clones despite a lack of understanding of the underlying mechanisms that provide resistance to CPB.
- Selection criteria include determining resistance on the basis of wet bean losses (Lim and Phua, 1986) and the hardness of the sclerotic layer (Day and Mumford, 1983; Azhar and Lim, 1987; Bekele, 1996; Haya et al., 2007).
- The first large scale attempt at screening for CPB resistance in Malaysia was initiated in 1986 by comparing pod infestation rates and wet beans loss using 59 clones (Lim and Phua, 1986). The study found that dried beans from infested pods weighed less than those from healthy pods because the thickness of beans from infested cocoa pods was reduced significantly. Clones PA7, UA30, UA12, UA9 and NA34 were found to be more tolerant to CPB attack, whereas clones ICS98, NGK16, I594, I206 and WA331 were most susceptible.

Average damage severity index

Five major categories:

- Healthy
- Slight
- Light
- Medium
- Heavy / heavy spoon / germinated beans

Score code description

- 0 – Healthy: Healthy (no larva penetrated the sclerotic layer (SCL) and all beans extractable)
- 1 – Slight - Slight damage (Larvae penetrated the SCL with sign of infestation inside the pod, such as the frass and cell growth on the inner endodermis, but all beans are extractable)
- 2 – Light - Light damage (<20% of the beans are unextractable)
- 3 – Medium - Moderate damage (21-50% of the beans are unextractable)
- 4 – Heavy - Heavy damage (>50% of the beans are unextractable)

Infestation category

- Healthy
- Slight
- Light
- Medium
- Heavy
- Germinated beans

Average damage severity index

Degree of infestation is obtained from ADSI (Average Damage Severity Index) values

[(0xn1) + (1xn2) + (2xn3) + (3xn4) + (4xn5)]/N, where

- n1: number of pods in category 0 (Healthy)
- n2: number of pods in category 1 (Slight)
- n3: number of pods in category 2 (Light)
- n4: number of pods in category 3 (Medium)
- n5: number of pods in category 4 (Heavy)
- N : total number of pods examined

Infestation category

- 0-0.9: Healthy to slight - Tolerant
- 1-1.9: Slight to light - Moderate
- 2-2.9: Light to medium - Susceptible
- 3-4: Medium to heavy -Susceptible

Average damage severity index

- Example of 1000 assessed pods
- (250 healthy, 50 slight, 400 light, 200 medium, 100 heavy)
- [(0x250) + (1x50) + (2x400) + (3x200) + (4x100)]/1000
- = 0 + 50 + 800 + 600 + 400 / 1000
- = 1850 / 1000
- = 1.85

Crop loss (CL) and pod infestation (PI)

$$CL = \frac{[(He \times 0) + (S \times 0.093) + (M \times 0.297) + H] \times 100}{TP}$$

$$PI = \frac{[S + M + H] \times 100}{TP}$$

Where,

He = 0 % Not pod infested by CPB

S = 0 – 20 % unextractable but pod infested by CPB

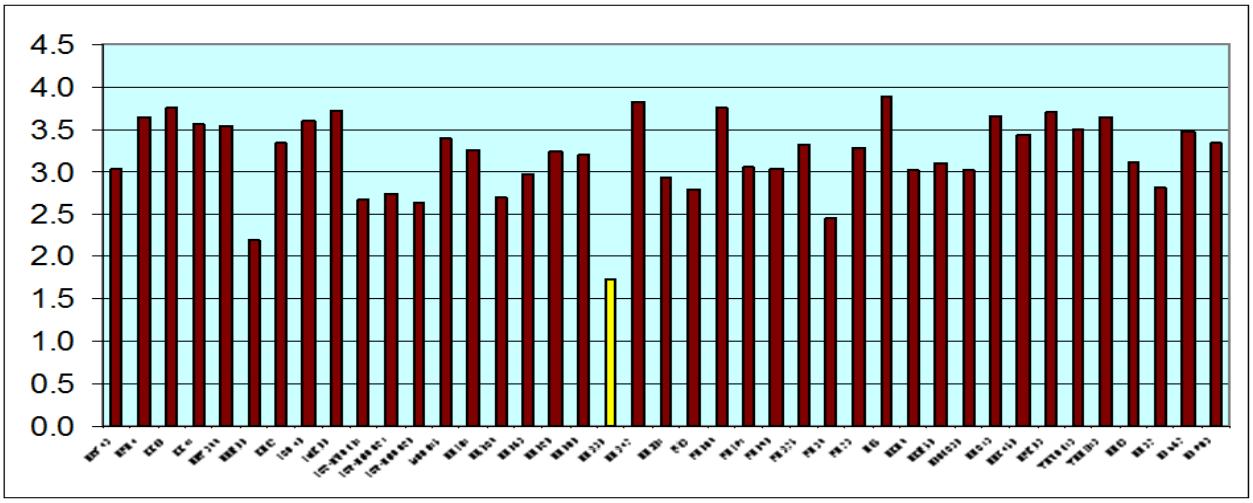
M = 21 – 50 % unextractable

H = > 50 % unextractable

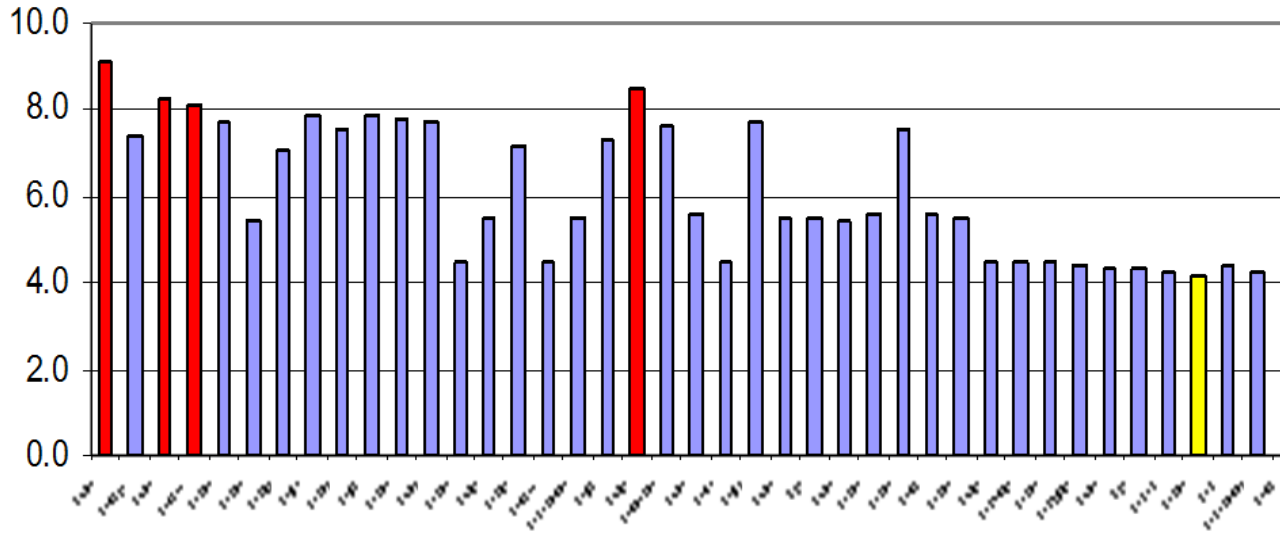
TP = Total Pod

Source : Manjit Sidhu

Average damage severity index (ADSI)



Pod hardness



Pod Hardness – Sclerotic layer thickness • APA 4 • MOQ 216 • IMC 55 • SIAL 325	Pod Hardness – ADSI • SIAL 325 • LCT-EEN 37I • UA 13
Entry Holes – ADSI • NA 325 • EQX 80 • LCT-EEN 241	Exit Holes – ADSI • NA 325 • EQX 80 • LCT-EEN 37E • NA 162

Clones categorized into various CPB resistant categories based on Resistance Index (RI)

Tolerant

- SIAL 325
- NA 311
- LCT-EEN 37I
- MOQ 216
- PA 171
- UA 13
- NA 162
- APA 4
- IMC 55
- P 13
- PA 109
- L-E 37E
- UF 667

Moderately tolerant

- NA 180
- PA 29
- PA 276
- SGU 60
- PA 73
- NA 139
- EET 399
- NA 101
- CC 18
- NA 247
- ACT 43
- NA 235
- SNK 12
- NA 170
- CC 41

Susceptible

- SNK 60
- R 16
- PA 198
- TSH 1112
- TSAN 812
- EQX 80
- ICS 40
- LCT-EEN 241
- SPC 52
- SCA 9
- GS 17
- UA 37
- UF 703

Conclusions

Both methods only provide early indicators for the degree of CPB infestation, other factors such as economic threshold/economic injury level should be considered when screening of the clone.

6.3.3 Latin America and Caribbean Cocoa Breeders Group

Wilbert Phillips represented the Latin America and Caribbean Cocoa Breeders Group that will be established during the upcoming meeting at CATIE 30-31 October 2014. He presented the following key points verbally:

- We need to secure urgently the germplasm that we have.
- We have old sources of resistance and it is hard to make progress.
- We need to define a strategy to complete the gaps in the collection and carry out actions.
- We need to share information and methodologies for evaluation.
- We need to find materials to establish evaluation trials addressing the issues of climate change.
- A key issue is how to fund and budget for these activities.

GENERAL DISCUSSION

The following point were mentioned and/or raised during the follow-up discussion:

- Nobody and no country is self-sufficient when it comes to cacao genetic resources and we need the international collection to have access to the diversity.
- We need pre-breeding populations that captures diversity and traits that can be used.
- The GSCC can develop a list of priority traits for evaluation.
- Need to prioritise materials at the International Cocoa Quarantine Centre at Reading (ICQCR) but how should this be done?
    - Primary germplasm at ICQCR and CRC
    - Cocoa pod borer and witches broom resistance
- We need to find out if the CFC collection is being used and what is the status of this programme and ensure that the CFC collection goes through the ICQCR.

- We need funds to continue maintaining or continue the evaluation trials.
- The Asia Pacific Breeders group collects data on yield.
- We need more information on the heritability of yield component.
- How important is compatibility in yield? Important and some examples in Brazil of this. Compatibility
- The Peru collection carries out a heavy metals extraction trait trials.

6.4 From the Different Cocoa-Producing Countries

Participants from different cocoa-producing countries provided a summary of the key traits of most importance to complement the discussions from the regional breeders groups. The countries represented were the following:

- Brazil
- Ecuador
- Costa Rica
- Cote d'Ivoire
- Hawaii
- Malaysia
- Peru
- Puerto Rico
- Trinidad and Tobago

GENERAL DISCUSSION

The following point were mentioned and/or raised during the follow-up discussion:

- The biggest cost for cocoa production is the labour cost. Therefore traits that reduce this cost are needed and may be even more important than yield.
- We need to develop smaller plants. For example, moniliasis depends on size.
- The shape of the tree is also important and should make it easy for mechanisation.
- In Trinidad and Tobago for example the cost of labour is very high and therefore less pruning is a key factor. Witches broom is under control now so labour cost is the most important thing.
- In Brazil the cost of post harvest is estimated at 30% of the total cost.
- Management efficiency, uniformity of production and harvest is very important.
- Bean size and fat content is important for international markets but pest and diseases are more important economically.
- The minimum bean size and fat content for industry requirement are 1g/bean and 45%, respectively, which are the benchmarks across the world.
- Number of beans per pod and beans to husk ratio is important.
- Cocoa butter content is at the top, and then high flavonoids/polyphenols and cadmium content are some of the priorities. In the Caribbean, cadmium is a very important issue.
- Small chocolate companies may have different requirements.
- Other important factors are related to adaptability to grow in specific, new and/or marginal areas affected by conditions such as drought. In Trinidad and Tobago for example, cocoa is being pushed into lands less suitable for it that require high fertilisers and other input. We need criteria for ability to extract nutrients for more efficiency.
- We need to breed for common pest and diseases such as CSSV and megakarya in West Africa but also preventive breeding for pests and diseases that may come one day from other regions.
- We need agreement on the evaluation of selected physiological characters and look at the ICQCR materials with this focus. Resources are needed to do this and develop evaluation projects.

- The target should be to preserve all the existing diversity. This is the challenge since we do not know the future.
- We need to prioritise the morphological and molecular characterisation of germplasm.
- We need to agree on a number of priority traits relevant to the following categories and if these are global or region-specific.
- Screening methods are needed to identify a subset of materials for further evaluation. We need to also distinguish between identifying a snapshot and when deep thorough evaluation has been carried out.
- We may need to also consider what is easy to measure even if not relevant to all.
- Should consider the totality of the cocoa world the key issues for sustainability and not only what is driving a part of the marker today.
- We need to identify core sets for pests and diseases resistance, for flavour and for a range of key characteristics.

7. Information on Evaluation Contributing to the GSCC Decision-Making Process

7.1 The ICGD and CANGIS Information Systems

Chris Turnbull made a presentation, also on behalf of Max Ruas, on the International Cocoa Germplasm Database (ICGD) and the CacaoNet Germplasm Information System (CANGIS) as the information systems to document the GSCC. It included the following points:

Information Systems to document the GSCC

Improving the documentation and sharing of information on germplasm is a key element of the Global Strategy for the conservation and use of cacao genetic resources.

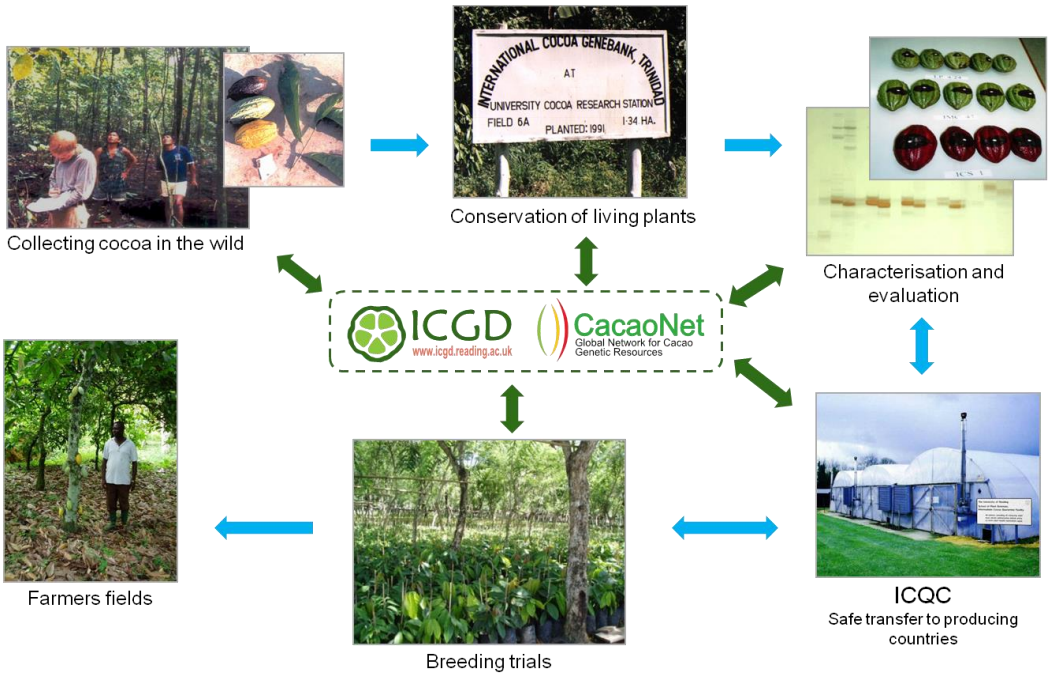
The International Cocoa Germplasm Database (ICGD)

ICGD is the principal data provider for the global cocoa research community

- Over 29,500 cocoa variety names, including synonyms
- Agronomic traits (including quality, yield and disease reactions)
- Morphological data
- Origins and locations of varieties
- Over 1600 photographs and drawings
- More than 3000 genetic fingerprints
- Fully referenced

Funded by NYSE Liffe and CRA Ltd.

ICGD: Supporting the Global Breeding Effort

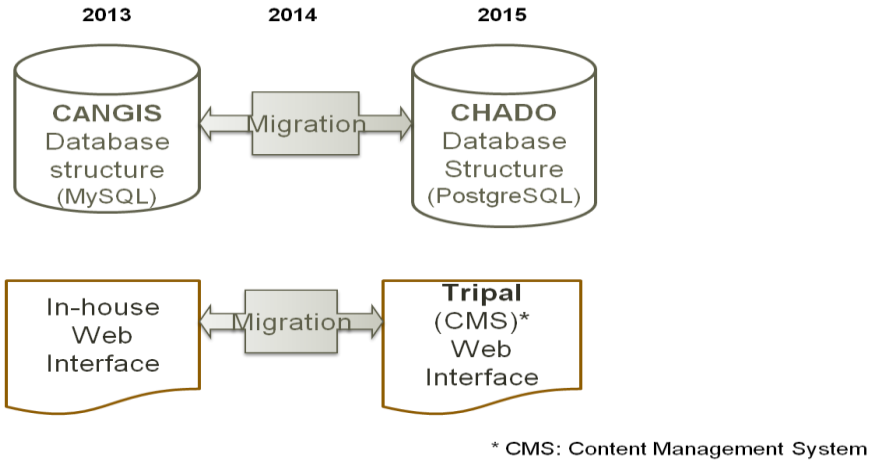


- ICGD: example - www.icgd.rdg.ac.uk
- ICGD: GSCC tool

CANGIS - CacaoNet Germplasm Information System

- Web-based, accession level information database
- Deals exclusively with germplasm in the GSCC and the information about their inclusion
- Modelled on the new version of MGIS (Musa Germplasm Information System)
  - Reduced development time
  - Backed by a wealth of expertise
  - Other crops (e.g. Coconut) are also using MGIS as a model, making the future integration of CANGIS with other systems an easier prospect

See Annex 3 - Figure 7. Components of the GSCC information portal (C. Turnbull, Reading University).



- Chado is a relational database schema
  - Capable of representing many of the general classes of data frequently encountered in modern biology such as sequence, sequence comparisons, phenotypes, genotypes, ontologies, publications, and phylogeny
  - Chado manages multi site evaluation data
  - Crop Ontology integration facilitated
- Tripal is a web front end for Chado databases
  - based on the Drupal content management system
- Chado & Tripal supported by the GMOD (Generic Model Organism Database) project

Data

There is more data to enter into ICGD and CANGIS:

- Data received/obtained, but yet to be entered
- Incomplete datasets (e.g. those linked to publications)
- Data yet to be received (e.g. accession lists)
- Unavailable data (e.g. progeny trials)
- New data, yet to be generated (e.g. flavour)
- Unaware of some data (e.g. CSSV)

Your help is needed to increase the amount of information available to aid the decision making processes for the GSCC

- Check the ICGD reference search

7.2 A GLOBAL PLANT GENEBANK INFORMATION MANAGEMENT SYSTEM – GRIN-GLOBAL

Pete Cyr made a presentation on GRIN-Global, the global plant genebank information management system. It included the following points:

So what is GRIN-Global?

- GRIN-Global (GG) is a software suite that enables genebanks to store and manage information associated with plant genetic resources (or germplasm) and deliver that information globally.

Why GRIN-Global?

- Genebanks can tailor a powerful information management system to meet their specific requirements because GRIN-Global has been developed with free or open source software and its source code is available.

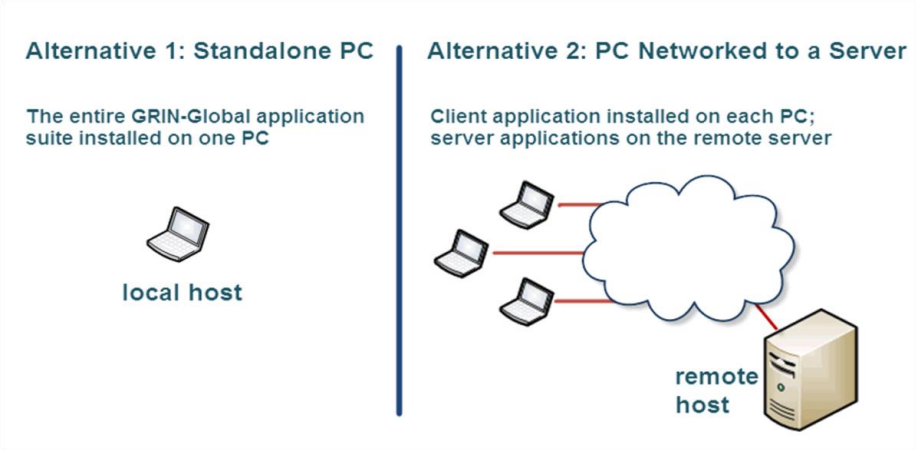
GRIN-Global manages many different types of data…

- Passport and Provenance (Accession ID, Taxonomy, Accession name, Origin, Material Type, Maintained By, Availability, Intellectual Property, Material Transfer Agreement Status)
- Phenotypic and Genetic Marker Observations
- Images of germplasm
- Germplasm inventory (e.g. number of seeds or plants)
- Requests for germplasm and order fulfillment
- and much more…

GRIN-Global & Interoperability

- GRIN-Global will interoperate with databases that provide specialized genomic, ecogeographic, and many other types of information needed for multidisciplinary research objectives.

Alternative GRIN-Global Configurations



Curator Tool

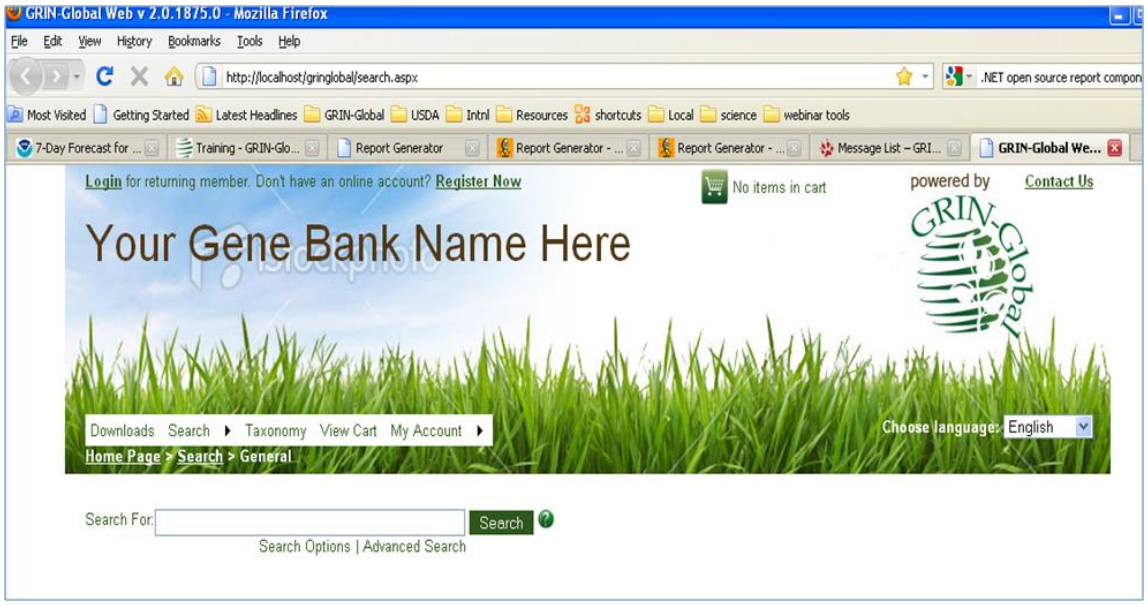
- The Curator Tool is a GRIN-Global application designed primarily for curators and genebank workers creating and managing their genebank's data

Search Tool

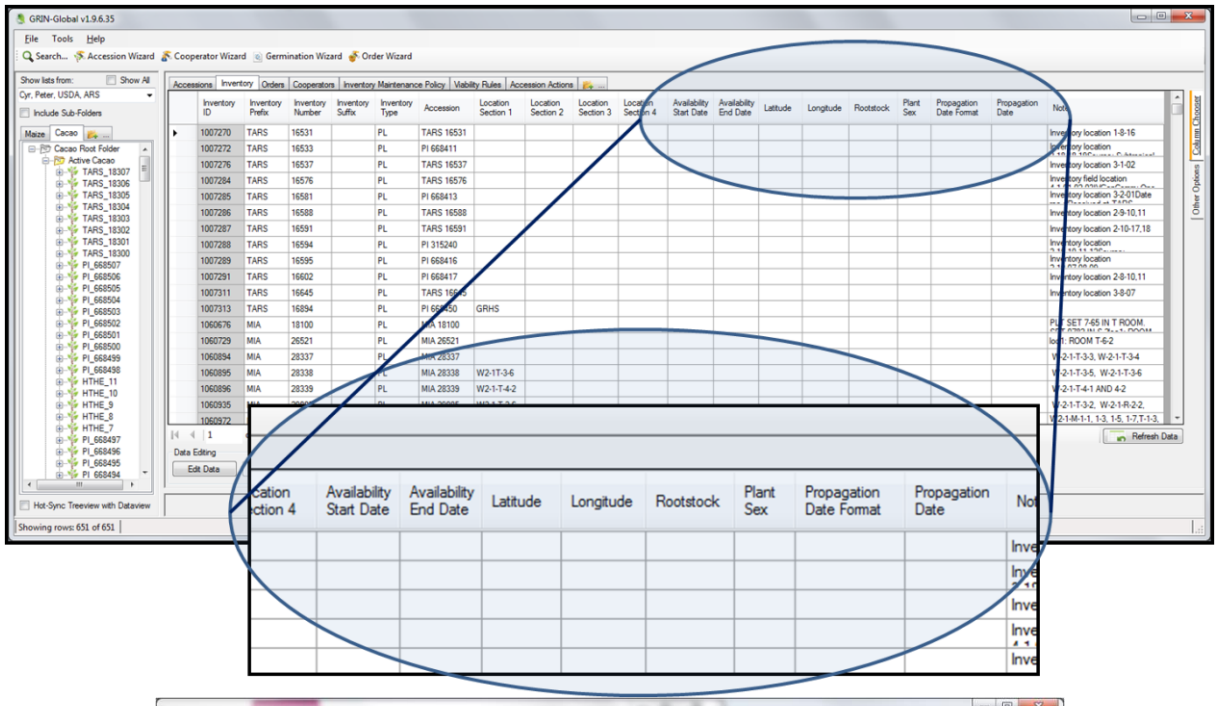
- The Search Tool can be launched from the Curator Tool, or launched directly from the Windows Start Programs list

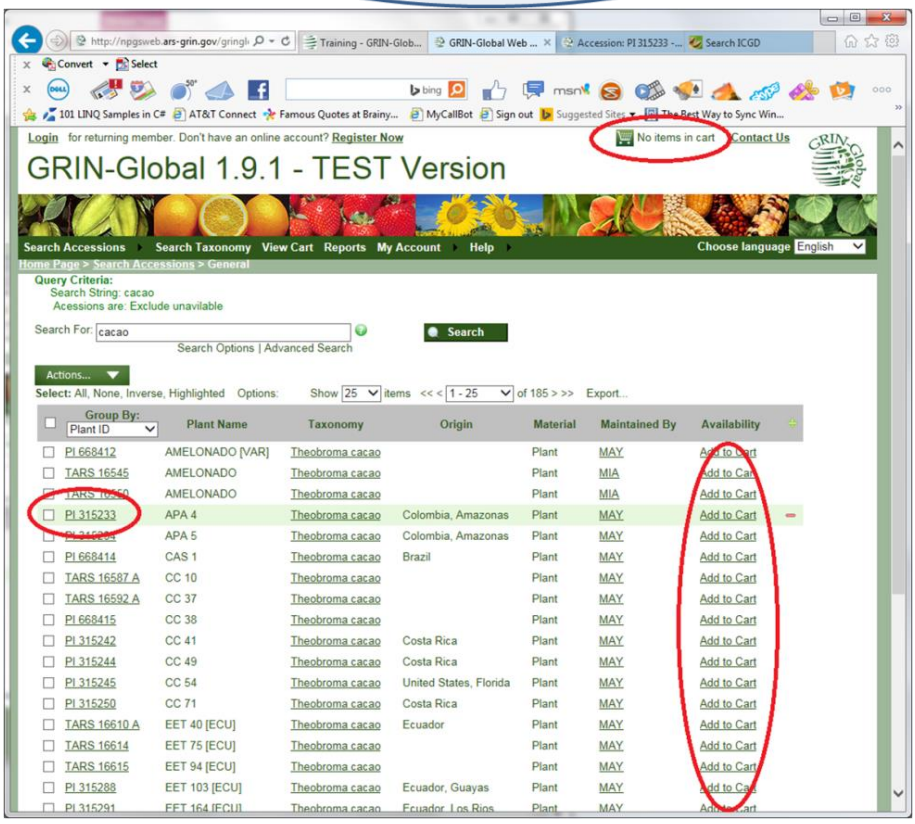
Public Website

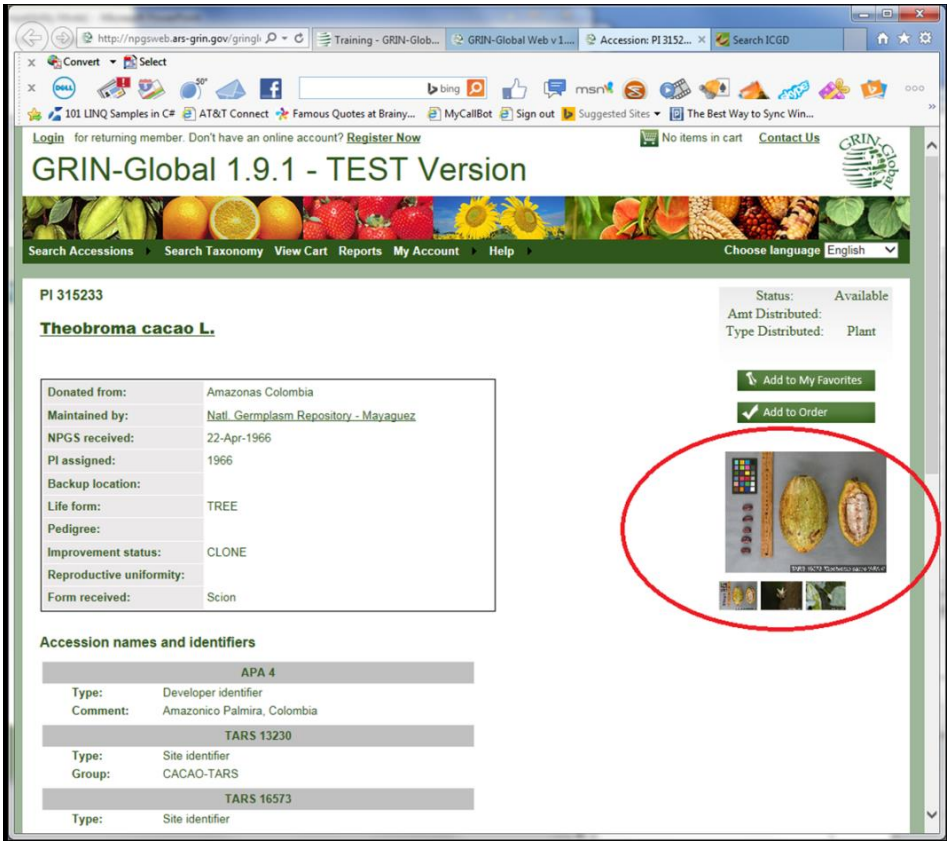
- Through the Public Website, clientele can search the GG database, access germplasm information, and order germplasm through a "shopping cart facility."



New Inventory Columns for Clonal Germplasm






Login for returning member. Don't have an online account? Register Now
No items in cart
Contact Us
GRIN-Global 1.9.1 - TEST Version
Search Accessions
Search Taxonomy
View Cart
Reports
My Account
Help
Choose language English
PI 315233
Theobroma cacao L.
Status: Available
Amt Distributed:
Type Distributed: Plant
Add to My Favorites
Add to Order
Donated from: Amazonas Colombia
Maintained by: Natl. Germplasm Repository - Mayaguez
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PI assigned: 1966
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Life form: TREE
Pedigree:
Improvement status: CLONE
Reproductive uniformity:
Form received: Scion
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Group: CACAO-TARS
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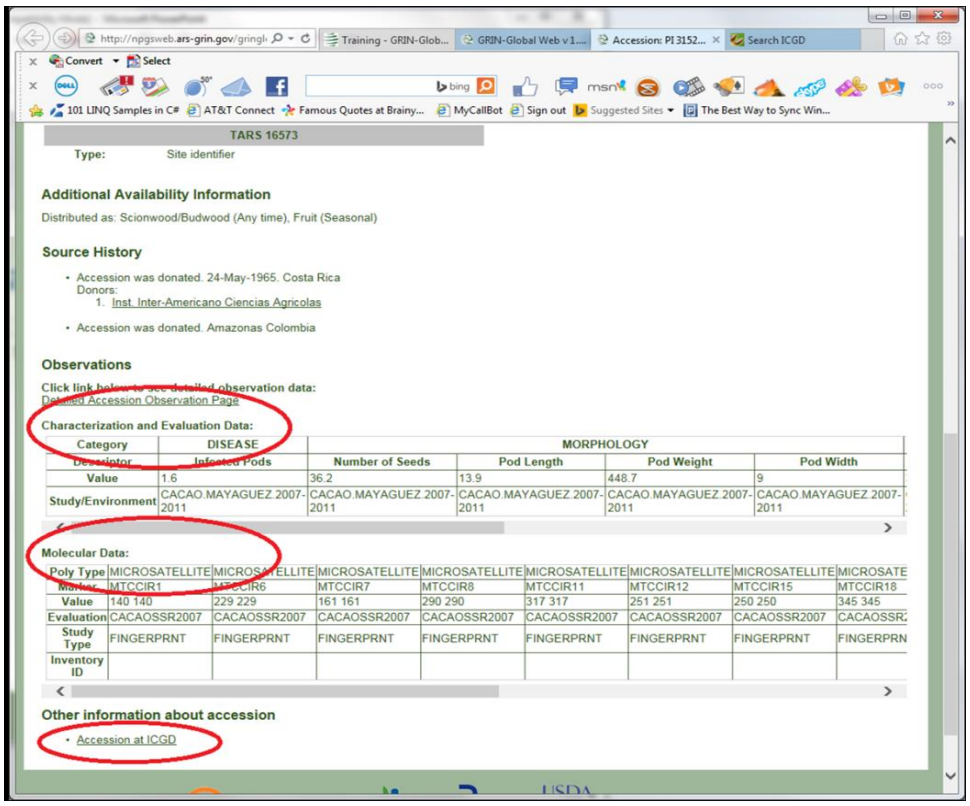
TARS 16573
Type: Site identifier
Additional Availability Information
Distributed as: Scionwood/Budwood (Any time), Fruit (Seasonal)
Source History
Accession was donated. 24-May-1965. Costa Rica
Donors:
1. Inst. Inter-Americano Ciencias Agricolas
Accession was donated. Amazonas Colombia
Observations
Click link below to see detailed observation data:
Detailed Accession Observation Page
Characterization and Evaluation Data:
Category DISEASE MORPHOLOGY
Descriptor Infected Pods Number of Seeds Pod Length Pod Weight Pod Width
Value 1.6 36.2 13.9 448.7 9
Study/Environment CACAO.MAYAGUEZ.2007-2011 CACAO.MAYAGUEZ.2007-2011 CACAO.MAYAGUEZ.2007-2011 CACAO.MAYAGUEZ.2007-2011 CACAO.MAYAGUEZ.2007-2011
Molecular Data:
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Marker MTCCIR1 MTCCIR6 MTCCIR7 MTCCIR8 MTCCIR11 MTCCIR12 MTCCIR15 MTCCIR18
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Evaluation CACAOSSR2007 CACAOSSR2007 CACAOSSR2007 CACAOSSR2007 CACAOSSR2007 CACAOSSR2007 CACAOSSR2007 CACAOSSR2
Study Type FINGERPRNT FINGERPRNT FINGERPRNT FINGERPRNT FINGERPRNT FINGERPRNT FINGERPRNT FINGERPRN
Inventory ID
Other information about accession
Accession at ICGD

Current Status of GRIN-Global

- The GRIN-Global Development team is currently testing:
  - GRIN-Global Server v1.9.2
  - GRIN-Global Client v1.9.6.35
- The source code (and compiled programs) have been made available to Matija Obreza at the Global Crop Trust
- The GRIN-Global Team's intention is that the GRIN-Global v2.0 (complete with source code) will be given to the Public Domain (with no restrictions on usage)
- Until GRIN-Global v2.0 is made available we are providing early release code with a letter of transmittal

For additional information, please visit:

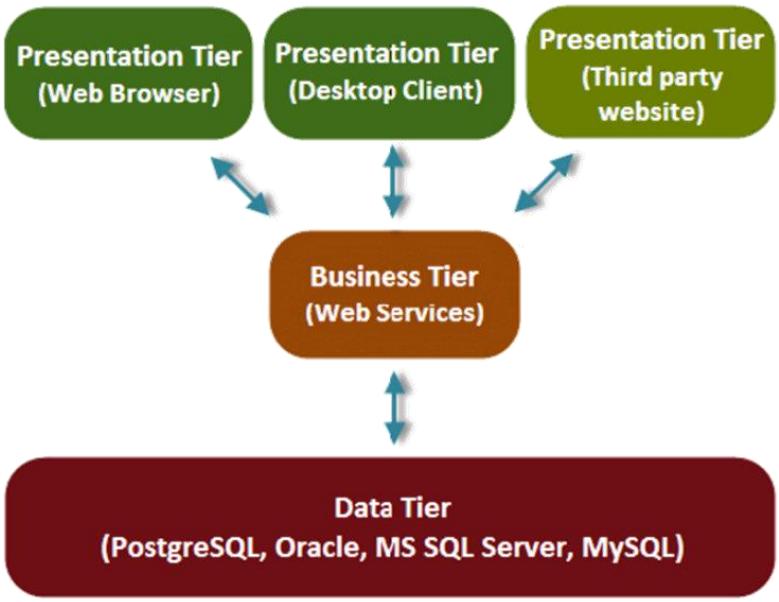
- www.grin-global.org (project background)
- www.grin-global.org/index.php/Training (training and documentation references including videos, exercises, and presentations)

GRIN-Global Design

- New applications written with the .NET framework and browser-based components comprise a system flexible enough to handle the needs of both small and large organizations.

Design…3 Tiers

- The system architecture uses three tiers - database, business, and presentation – that can be developed and modified independently.



Design…Web Services

- The GRIN-Global Application Program Interface (API) provides web services–software modules accessible over the internet via standard messaging protocols.
- The web services allow the data to be retrieved in various formats – XML, CSV, TXT – making it platform independent. Any application that can make HTTP requests can use the GRIN-Global API.

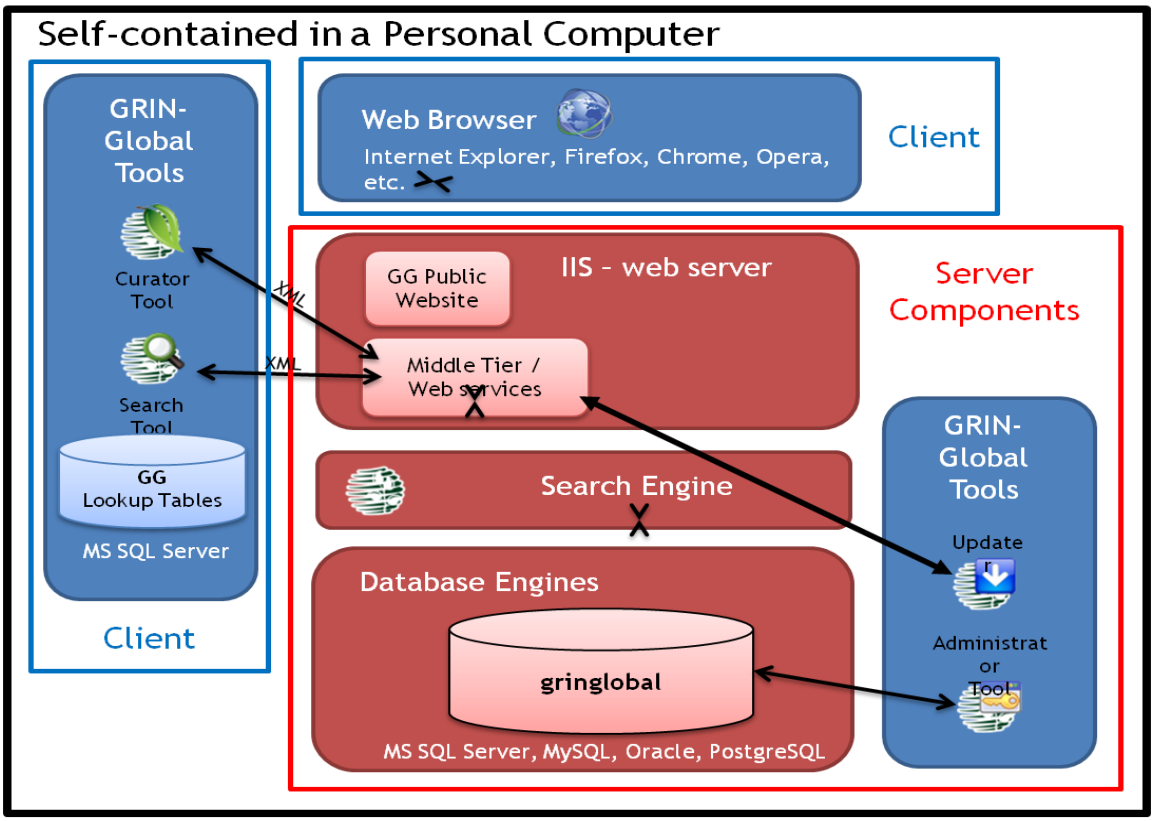
Design…Open Source

- Supports free or open source database management systems:
    - MySQL, PostgreSQL, Oracle Express, or Microsoft SQL Server Express
- By providing all source code for system components, GRIN-Global enables genebanks to tailor the application to meet their specific requirements

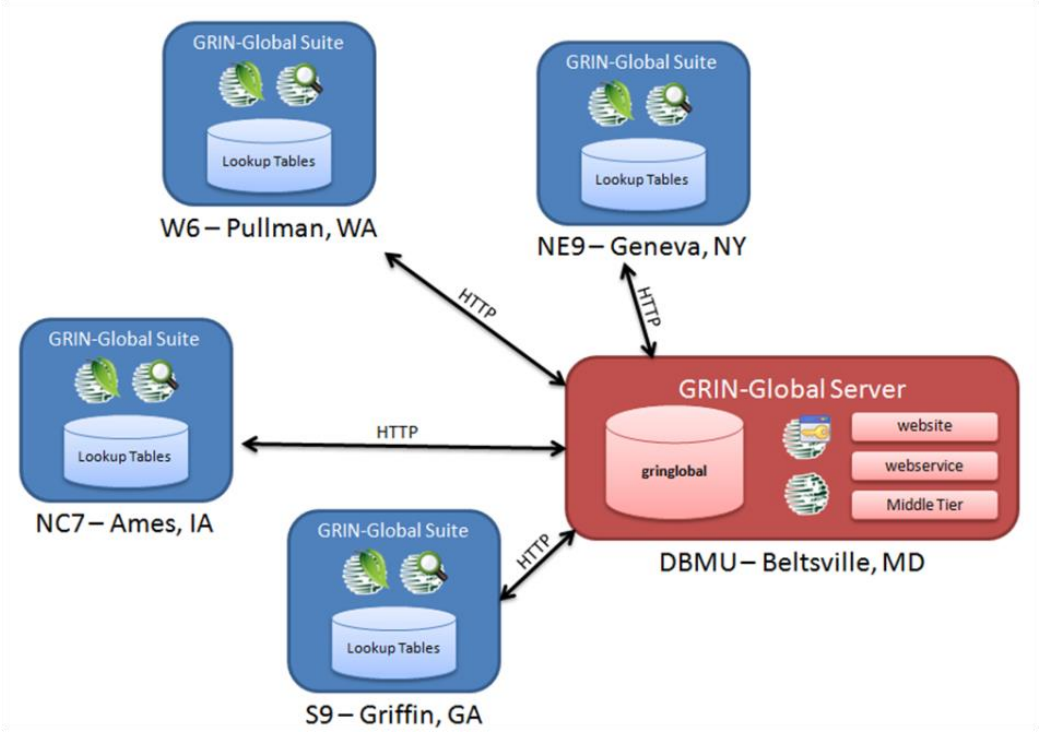
GRIN-Global Environment

- The GRIN-Global (GG) program suite runs in a Windows environment. Besides the basic Operating System, GG also requires the Windows IIS.
- One of the four supported database engines (SQL Server, Oracle, MySQL, or PostgreSQL) is required. The database software houses the many GRIN-Global data tables. GG is a relational database system -- these tables are related by key fields. The major related "families" of tables include Accessions, Inventory, Taxonomy, and Orders.
- Users access the data via the GRIN-Global applications: the Curator Tool (CT), the Search Tool, and the Public Website. The CT and the Search Tool are closely integrated – the CT has a Search button which invokes the Search Tool; however, the Search Tool can also run as a stand-alone application.

GG System Architecture Overview – Single PC

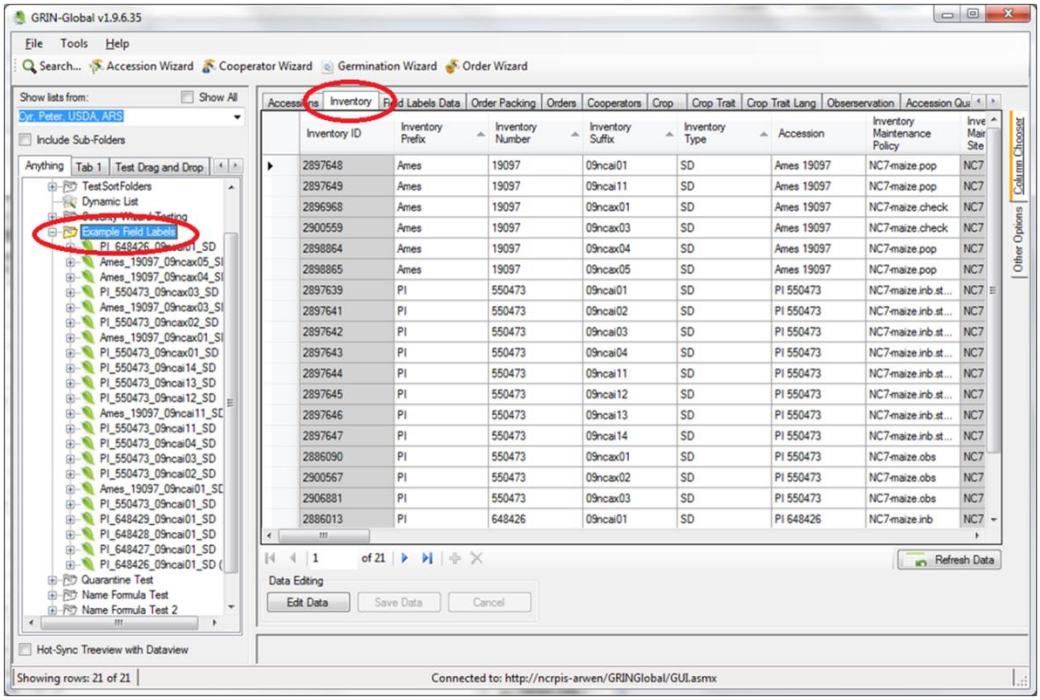


GRIN-Global System Architecture Overview – Networked to a Server

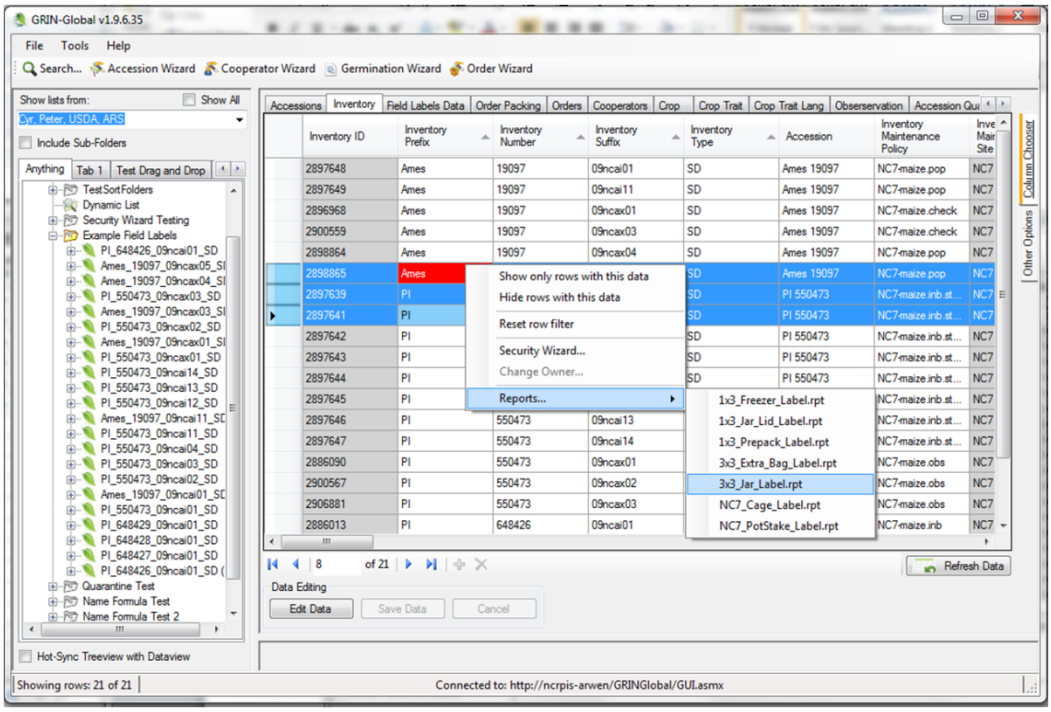


The following was sent out by Pete Cyr by email following the question about the printing of accession labels:

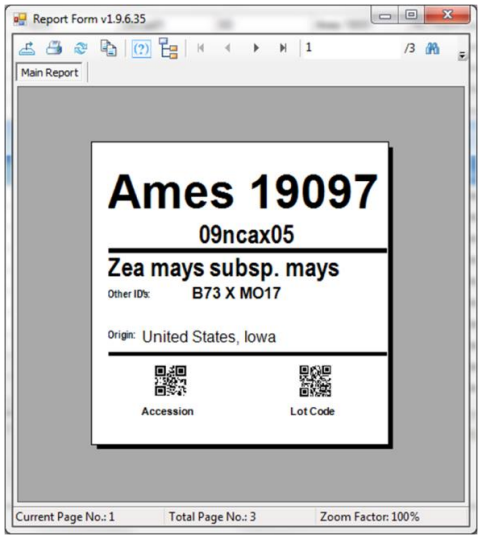
Selected a "List Folder" on left side of CT and then chose the Inventory Tab on the right side of the CT



Selected 3 rows (out of the 21 visible) then right-clicked anywhere on the blue highlighted rows to view the reports menu (I selected the “3x3 Jar Label” we use in cold storage)



Which brings up the Report Viewer where you can print or export the jar labels



Here is an example of our “Field Label” made using the same process on the same data

7.3 Integrated Breeding Platform (IBP) and Breeding Management System (BMS)

Elizabeth Arnaud made a presentation, also on behalf of Chris Turnbull, introducing the GCP Integrated Breeding Platform (IBP) crop ontologies, Trait Dictionaries and objectives/uses of Breeding Management System (BMS). It included the following points:

Integrated Breeding Platform and Crop Ontology www.integratedbreeding.net

Slides courtesy of Jean-Marcel Ribaut

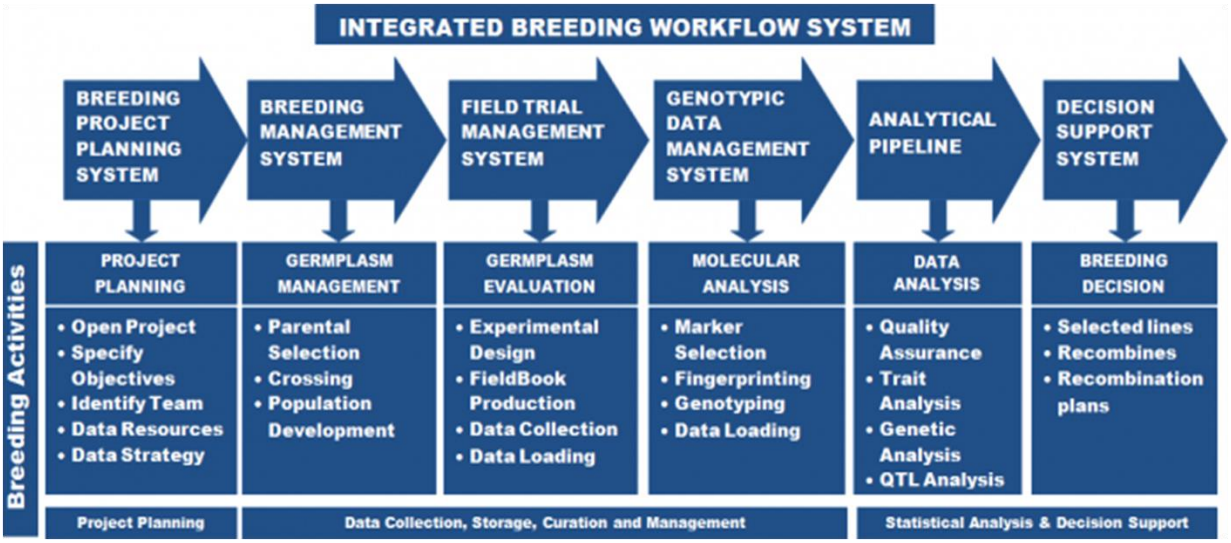
IBP Overall Objective

Improve the efficiency of plant breeding programmes in developing countries by enabling plant breeders to access modern breeding technologies, breeding materials and related information in a centralised, integrated and practical manner.

- Crop groups –Community of Practice, involving private sector
- Breeding management system : workbench
- Expertise and services
- Training and capacity building (partnership with Universities)

Breeding Management System

- 9 crop databases with historical data and trait dictionaries: bean, cassava, chickpea, cowpea, groundnut, maize, rice, sorghum and wheat
- Up next will be: barley, lentil, potato, soya beans and sweet potato and more to come
- Empty DB available for all crops. Trait Dictionary is the 1st product to develop
- A comprehensive, all-in-one suite of tools to effectively manage your breeding activities from project planning to final decision-making:



Version 3 – Sept 2014

Breeding Management System (BMS) Product Concept

- Targets routine breeding activities
- Simple and easy-to-use application containing all informatics tools needed by a breeder
- Seamless flow of data between applications
- Accumulation, sharing and re-use of breeding data

- Will allow integration of users own tools into the system
- Implementable as a standalone system - Access to local DB, as well as the BMS on a local PC
- Implementable as a LAN-based system - Data synchronization with central DB

https://www.integratedbreeding.net/161/training/bms-user-manual/

BMS Core Applications

Breeding activities:

- Manage lists
- Make crosses
- Manage nurseries
- Manage trials
- File format

Information management:

- Germplasm import
- Manage genotyping data
- Browse studies
- Adapted germplasm query
- Manage ontologies

Marker-assisted breeding:

- Molecular breeding design tool
- OptiMAS
- Molecular Breeding Planner

Statistical Analyses:

- Single site analysis
- Multi-site analysis
- Multi-site multi-year analysis
- QTL analysis

Breeding View (BV) is a graphical user interface to a statistical analysis package that permits you to conduct phenotypic and genotypic analyses of the field trial observations that you upload to the Breeding Management System (BMS).

The IBP User Support Services

- Scientific Support :
    - Client-oriented breeding support service
    - Capacity building support
        - comprehensive training in orientation to, and using, the tools
        - On line e-learning and curriculum for breeders
    - Interaction with peers through social networks and Communities of Practices available through the IBP portal
- Technical Support to be provided at two levels to all users:
    - Level 1: installation technical support - To overcome any difficulties in downloading, installing and getting started with the BMS and related tools
    - Level 2: Regional technical support - for users that might encounter problems in day-to-day use of the BMS and related tools
- Genotyping Support Services
    - Access to third party commercial laboratories for submission of genotyping projects at agreed preferential prices supporting both discovery and implementation projects
    - SNP – LGC Genomics
    - SSR – BecA & ICRISAT
- Phenotyping Support Services
    - Access to service providers for phenomics and metabolomics analysis
    - Access to phenotyping protocols and trait measurements under stress conditions
- GIS
    - Free access to seasonal private weather profile to better segment, understand, analyze and interpret trial results

Support for deployment - IBP Regional Hubs Network

Current Hubs:

- Africa Rice
- Biotec
- IITA
- CAAS
- BeCA
- ICRISAT
- CERAAS/CORAF
- Others Coming

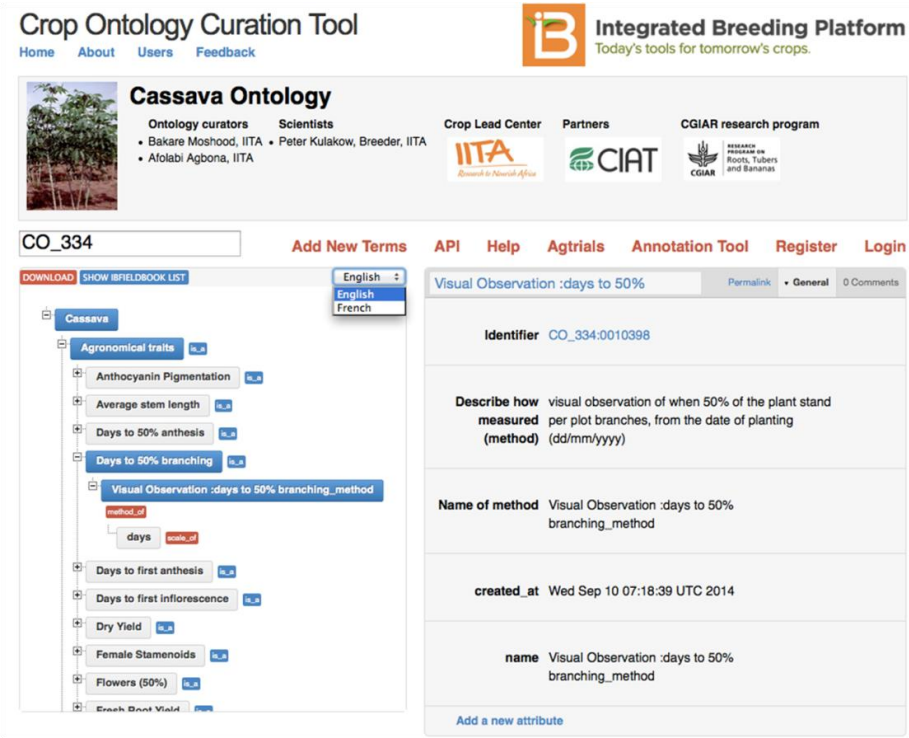
The Crop Ontology - www.cropontology.org

Unique ontology for traits of crop breeders including the methods and scales of measurement

- Common concepts supporting Integration of genetic and phenotypic data
- Creation of standard fieldbooks of BMS in Integrated Breeding Platform
- Community-based development with a methodology and a workflow
    - 8 CGIAR Crop Lead Centers and partners, CoP
    - 18 TD validated for all regions where evaluation is performed for a crop
- Annotations of data sets in 3rd party databases and web sites
    - BMS, Agtrials (CCAFS), Solanaceae breeding database in Wageningen, NGS Cassava - Cornell, Australian Phenotyping Platform, …
- Forum for scientists to discuss their variables across disciplines, regions

Fieldbook for Participatory Varietal Selection (PVS)

- Online visualization of Trait dictionaries
- Consensus trait classes



<u>Creating content for new crop database in the Breeding Management System (BMS) and fieldbook</u>

Six steps to adoption

- Assess the feasibility of adopting BMS
- Build an implementation team
- Develop an implementation plan
- Verify the plan and resources
- Setup customized program resources
- Deploy BMS in day to day operations

Baseline data

- Crop, farming practice and geographic region
- Program name and objectives
- Number of breeders, technicians and field staff
- Current information management practices
- Breeding methodologies - traditional, molecular, DH
- Program size - population development
- Program size - line evaluation
- Productivity of the program over the past five years
- Expected changes over the next five years

<u>Possible follow up</u>

- Developing the Trait Dictionary – needs consultations
- Follow up in key meetings and workshop
- Small group of early adopters

7.4 AGROGEN BREEDING MANAGEMENT SYSTEM

Uilson Lopes made a presentation on AgroGen, a database system for plant breeders that he developed in Brazil. It included the following points:

AgroGen: A Database System for Plant Breeders

Objectives:

- AgroGen – A Database system for breeders
- AgroForm – A system for collecting data on cheap (<US150,00) cell phones

Breeding Programs

- Data Storage
- Data Typing

Few Available + Expensive (Leasing?)

- 4 systems (Customizable to any crop)
    - PRISM (USA)
    - Agrobase (Canada): US$ 8,000/year
    - Doriane/Lakey (France)
    - NBS (China)
- Specific (sugarcane)

AgroGen – Database Management for Breeders database

- Version 1.0 : Release in March-April/2015
- Characteristics:
    - Web platform or a single PC
    - Multi-user (other institutions)
    - Any plant species
    - Import/Export data: text/Excel, dataloggers (including cell ´phones)
    - Basic analysis
    - Password protected + differencial access
    - Same platform:
        - Trial description
        - Variables, unit
        - Phenotypic + Molecular data
        - Diary
        - Field layouts
        - Germplam pedigrees+ pictures

Windows Based (Customization)

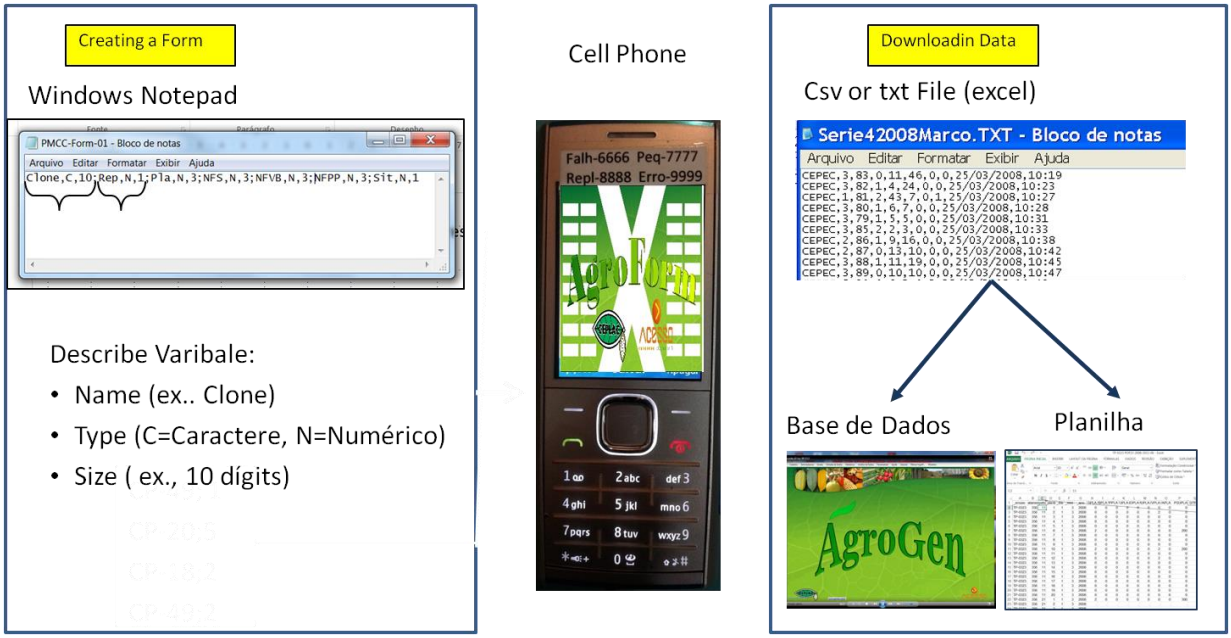
- Passwords
- Trial Description
- Germplasm Description
- Germplasm Pictures
- Field Layouts
- Typing Faciltator (Collection Order)
- Typing Facilitator (Virtual Forms)

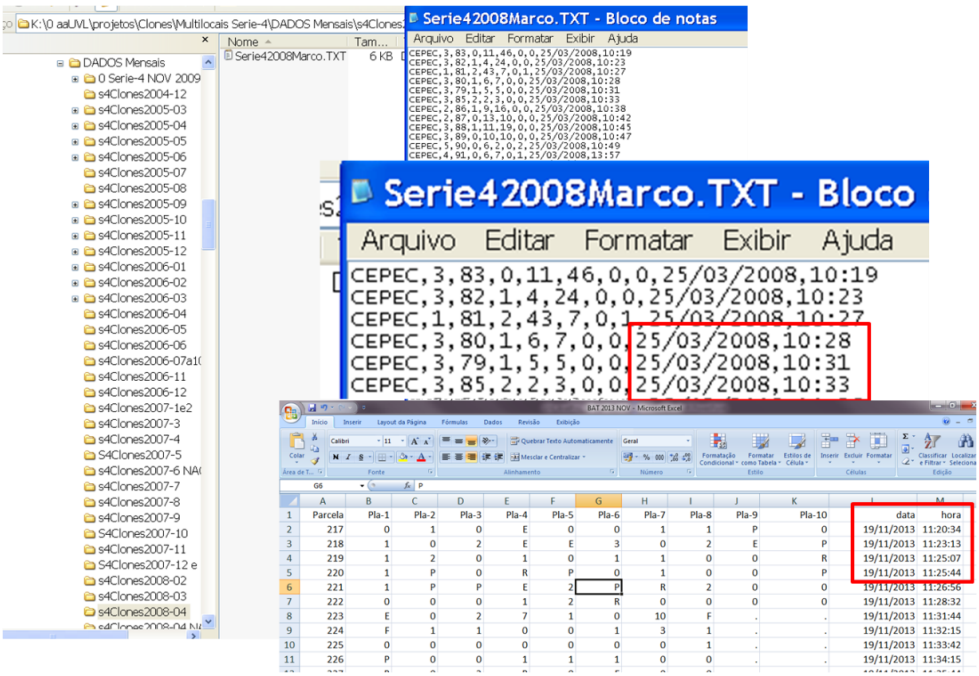
AgroForm – A Data Collection System for Cell Phones

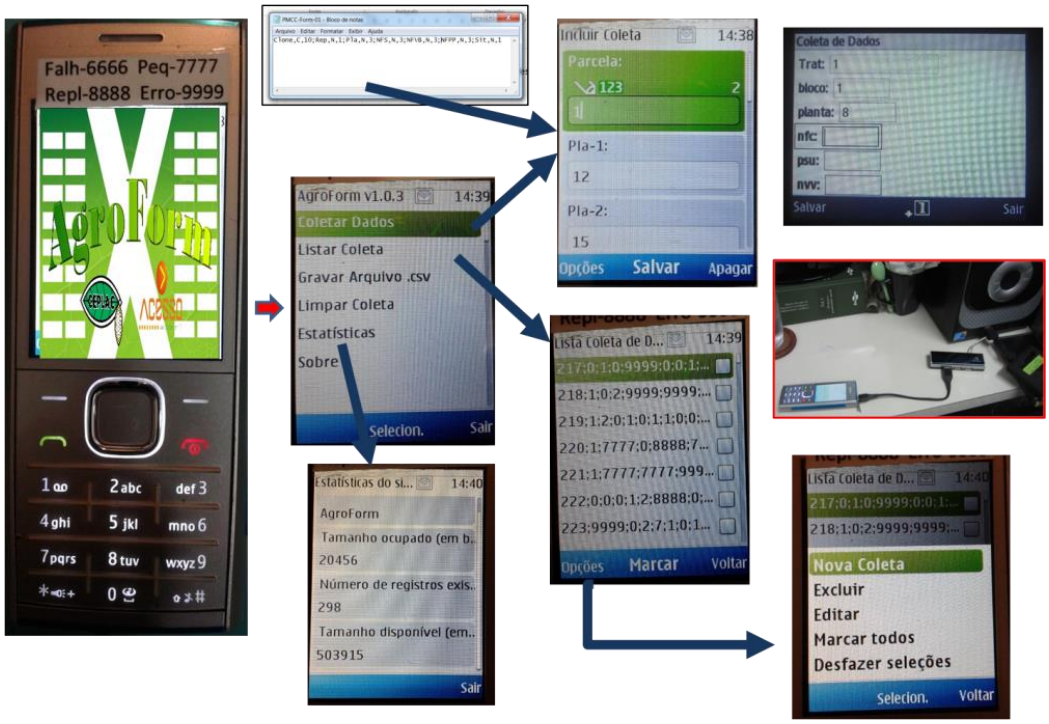
Characteristics:

- Easy to design the customized forms (Windows Notepad/txt)

- Any variable: Name, Number or form (Numeric vs Alphanumeric)
- Allows stablishing the “breeders ideal route”)
- Upload dificult to type data (treatments, reps, ...)
- Transfer data through USB cable (initially) ou GPRS (under development)
- Stores thousands datapoints in 2 MB card
- Future:
    - Taking info to the field (plant selection)
- Based on low cost cell phones (< US$ 100-150)
- Easy customization (Just use Windows notepad)







<u>Some Dreams</u>

Cell Phones

- Catalogs
  - Portable Field Germplasm (AgroCPC) (Catálogo Portátil de Cultivares)
  - Deficiency Systems
- AgroSafra (Farm Yield Forecast)
  - Tablets

AgroGen & AgroForm para Tablet

8. SECURING *EX SITU* CONSERVATION – FUNDING AND SAFETY-DUPLICATION

8.1 CACAONET TASK FORCE ON SUSTAINABLE FUNDING MECHANISMS FOR CACAO GENETIC RESOURCES

Brigitte Laliberté provided an on the establishment by CacaoNet of a Task Force on Sustainable Funding Mechanisms for Cacao Genetic Resources. The Task Force is focusing on the long-term conservation and use of the Global Cacao Strategic Collection (GSCC), as part of the Implementation of the Global Strategy for the Conservation and Use of Cacao Genetic Resources. The Task Force consists of a small group of dedicated partners to research the different options and propose a long-term funding model for the priorities of the Global Strategy. The Objectives of the Task Force are to:

- Review the current funding structures.
- Evaluate the long-term funding needs.
- Explore options of funding mechanisms and governance.
- Formulate recommendations for funding options of the different components of the Global Strategy.
- Make information available for wider consultation through CacaoNet and the WCF partnership meetings.

The first priority of the Global Strategy is to find a solution for the long-term sustainable funding to secure existing *ex situ* cacao genetic resources and their distribution (component 1 of the Global Strategy) supporting the development of the Global Strategic Cacao Collection (component 2 of the Global Strategy).

The Members of the Task Force are the following:

1. Brigitte Laliberté, Bioversity International, Italy, Task Force coordinator
2. Alison Branch, Mondelez International, UK
3. Gary Guittard, Guittard Chocolate Company
4. Martin Gilmour, Mars Incorporated, UK
5. Michelle End, The Cocoa Research Association Limited, UK
6. Path Umaharan, Cocoa Research Centre (CRC), Trinidad and Tobago
7. Paul Hadley, International Cocoa Quarantine Centre, Reading (ICQC,R) and Reading University, UK
8. Stephan Weise, Bioversity International, Italy
9. Virginia Sopyla, World Cocoa Foundation (WCF)
10. Wilbert Phillips, Collection Manager CATIE, Costa Rica

In order to seek funding, the Task Force recommended developing communication materials and agreeing on the key messages to be conveyed to potential donors. Another key task is to fine-tune the cost estimates of conservation. And the Task Force has been working with CATIE, CRC and ICQCR on developing costing studies, under the expert guidance of Daniela Horna, in order to provide a more detailed and accurate cost estimate of the management of the Global Strategic Cacao Collection (GSCC). Further details on the GSCC costing study is presented by Daniela Horna below.

The next steps of the Task Force is to start approaching potential donors to discuss the possible terms and conditions that the different types of funders might be interested and willing to contribute to. This feasibility phase will help determine the most appropriate funding mechanism to establish and the partners best placed to manage it for the long-term, taking into consideration the range of funding needs (short-medium-long terms activities).

8.2 GSCC Costing Study and Collection Management Operations

Daniela Horna presented, also on behalf of P. Umaharan, W. Phillips, A. Mata and B. Laliberté, on the GSCC costing study and collection management operations targeting an endowment vs project/research funding. It included the following points:

Costing Ex-situ Conservation - Cacao

Background on Costing

- CG system genebanks (2000)
- Value of Diversity ➔ Funding
- Information for decision making ➔ rationalization
- Different funding environment

Tool Development

- Different from annual budgets
- Flow of Operations
- No previous information
- Standardization for comparison

Decision Support Tool:
http://cropgenebank.sgrp.cgiar.org/index.php?option=com_content&view=article&id=45&Itemid=142

- Excel
- Information
    - Operations defined
    - Number of accession / year / operation
    - Inputs:
        - Facilities / equipment (fixed)
        - Researchers (quasi-fixed)
        - Supplies (variable)

Report – Total Costs – example from Rice, IRRI, 2008

Activities	No. Acc.	Total capital cost	Total quasi-fixed cost	Total variable labor costs	Total non-labor costs
Acquisition	2,899	103.65	4,211.15	65.19	241.34
Characterization	2,216	3,524.92	26,306.60	5,653.48	32,422.47
Safety duplication	1,300	270.82	7,528.03	814.90	302.56
Long term storage	676	23,952.96	4,954.15	437.82	100.57
Medium term storage	2,820	10,131.97	5,625.85	437.82	1,115.66
Germination testing	17,980	35,645.85	18,817.61	2,274.50	417.31
Regeneration	3,467	2,542.00	22,801.27	13,305.76	30,309.65
Seed processing	4,357	17,113.99	28,099.94	3,596.38	2,341.75
Seed health testing	3,840	880.46	10,991.34	1,407.28	48,318.21
Distribution	18,159	669.26	25,304.13	1,563.64	46,217.69
Information management	106,319	795.01	43,759.43	0.00	40,193.22
General management	106,319	5,648.36	73,363.48	0.00	17,646.94
Total	**N.A.**	**101,279.25**	**271,762.98**	**29,556.76**	**219,627.37**

Report – Average Costs – example from Rice, IRRI, 2008

Activities	Average capital cost	Average quasi-fixed cost	Average variable labor cost	Average variable non-labor costs	Total AC
Acquisition	0.04	1.45	0.02	0.08	**1.56**
Characterization	1.59	11.87	2.55	14.63	**29.05**
Safety duplication	0.21	5.79	0.63	0.23	**6.65**
Long term storage	35.43	7.33	0.65	0.15	**8.13**
Medium term storage	3.59	1.99	0.16	0.40	**2.55**
Germination testing	1.98	1.05	0.13	0.02	**1.20**
Regeneration	0.73	6.58	3.84	8.74	**19.16**
Seed processing	3.93	6.45	0.83	0.54	**7.81**
Seed health testing	0.23	2.86	0.37	12.58	**15.81**
Distribution	0.04	1.39	0.09	2.55	**4.02**
Information management	0.01	0.41	0.00	0.38	**0.79**
General management	0.05	0.69	0.00	0.17	**0.86**

Cocoa – GSCC

- CRC and CATIE
- Adjust operations to cacao
- Total annual and in-perpetuity

Standardization

A genebank needs to perform a number of operations to guarantee the proper conservation of an accession. The table below lists the operations included in the costing study and presents a short explanation of what each operation entails.

Activity	Explanation
1. Acquisition	This may involve the collection activities in the fields or the activities related to receiving and processing newly introduced accessions.
2. Greenhouse	Greenhouse or shade house management for propagation and multiplication of materials for planting in the field.
3. Field maintenance	The main activity of a cacao genebank – possibly considered “medium-term” storage
4. Characterization - Morphological	Morphological characterization of each accession.
5. Characterization - Molecular	Verification of identification of the materials.
6. Propagation	Routine propagation for replacement of dead or injured field plants in the field.
7. Health testing	This activity involves the testing of plant health, often carried out upon acquisition or during regeneration process.
8. Distribution / dissemination	Sending accessions upon request (e.g., preparation, shipment, etc.).
9. Safety duplication	Replication of the accessions in another field collection.
10. Cryopreservation	Cryopreservation / long term storage.
11. *In vitro* conservation	*In vitro* culture mainly use for the preparation of material for safe distribution and shipping.
12. Information and data management	This activity includes data entering, processing and management, including catalogue preparation and descriptor development. It also includes database management such as DBGERMO-WEB the international software currently used at CATIE

Activity	Explanation
13. General management	This is the activity that is difficult to allocate to specific activity (e.g., genebank manager's work)
14. Training	Activities related to the training of staff carrying out any of the activities of the collection management
15. Evaluation	Evaluation of the materials for flavour, disease resistance (etc.)

Results – Conservation

OPERATIONS	Ave. Costs	N° of access.	GLOBAL COLLECTION		
			Total Costs		In Perpetuity Costs (Recurring)
			Annual Recurring	One-Off	
Acquisition	87	20	1,745	-	-
Greenhouse	294	200	58,772	-	1,394,498
Field maintenance	48	2,500	119,926	-	2,845,519
Characterization - Morphological	951	100	95,131	-	2,257,214
Characterization - Molecular	358	200	128,565	-	3,050,496
Propagation	145.80	200	29,160	-	691,892
Health testing	46.74	200	9,348	-	221,803
Distribution	45.76	150	6,863	-	162,849
Cryopreservation-Introduction	1,500	-	-	-	-
Cryopreservation-Maintenance	4.00	-	-	-	-
Information and data management	15.84	2,500	39,596	-	939,516
General management	49.61	2,500	124,024	-	2,942,750
Training	9.28	2,500	23,200	-	550,484
Evaluation- flavour– pathology	292.10	200	-	58,420	-
TOTAL			**636,332**	**58,421**	**15,098,436**

Results -Safety Duplication

OPERATIONS	Ave. Costs	N° of access	SAFETY DUPPLICATION COLLECTION		
			Total Costs		In Perpetuity Costs (Recurring)
			Annual Recurring	One-Off	
Acquisition	87.27	50	4,364	-	103,535
Greenhouse	293.86	20	5,877	-	139,450
Field maintenance	47.97	1,000	47,970	-	1,138,208
Characterization - Morphological	951.32	-	-	-	-
Characterization - Molecular	358.56	-	-	-	-
Propagation	145.80	100	14,580	-	345,946
Health testing	46.74	100	4,674	-	110,901
Distribution	45.76	-	-	-	-
Cryopreservation-Introduction	1,500	500	-	750,000	-
Cryopreservation-Maintenance	4.00	500	2,000	-	47,455
Information / data management	15.84	1,000	15,839	-	375,807
General management	49.61	1,000	49,610	-	1,177,100
Training	9.28	1,000	9,280	-	220,194
Evaluation- flavour– pathology	292	-	-	-	-
TOTAL			**154,194**	**750,000**	**3,658,595**

Types of Operations

Operations	By Periodicity		By Scope	
	Routine	One-off	Custodianship	Impact-focused
Acquisition	X	X	X	
Greenhouse	X		X	
Field maintenance	X		X	
Characterization - Morphological	X	X	X	
Characterization - Molecular	X	X	X	X
Propagation	X		X	
Health testing	X		X	
Distribution	X		X	
Cryopreservation-Introduction		X	X	
Cryopreservation-Maintenance	X		X	
Information and data management	X		X	
General management	X		X	
Training	X			X
Evaluation- flavour– pathology		X	X	X

Implications for Funding

- Information about resources available and needed
- Explore scenarios
- Funding strategies
    - Who should fund what?
    - Endowment fund for custodianship operations?
    - Impact operations funded by project?
    - Technology change?
- Self funding?

Additional comments during the follow-up discussion:

- CATIE and CRC are conserved so far because of all the efforts so far (projects, sales etc) but the funds need to be secured.
- The total cost reflects how people are willing to pay for it.

8.3 Cryopreservation for Long-Term Safety Duplication

Anne Buchwalder made a presentation on cryopreservation at the Nestlé Research and Development Centre, Tours, France. It included the following points:

Cryopreservation at the Nestlé R&D Centre, Tours, France.

R&D Tours

- 55 to 60 people (including long term trainees)
- Historically specialized in plant and culture *in vitro*
- Today 4 groups:
    - Tissue culture
    - Molecular biology
    - Biochemistry
    - Agronomy & sustainability

Skills and Know-how

- *In vitro* culture
    - Multiplication of plant tissue in aseptic way
    - Plant cell suspensions
    - Cryo preservation
- Molecular biology
    - Finger printing for traceability
    - Molecular assisted selection to help plant breeding
- Biochemistry
    - Quantification of compounds (HPLC, UPLC, GC- MS)
    - Near Infra Red predictive tool Sensory analysis

Strong experience in cryo preservation

Different species:

- Coffee (Robusta and Arabica)
- Cocoa
- Chicory
- Hevea
- Several vegetables

Different explants:

- Embryogenic calli
- Roots
- Zygotic embryos
- Meristems

Cryo-preservation of cocoa at R&D Tours

1. Cell suspension – calli
2. Pre treatment
3. Cryo tubes
4. Storage
5. Thawing +4-0oC
6. Re-hydration
7. Growth

Effort spent on cryo preservation

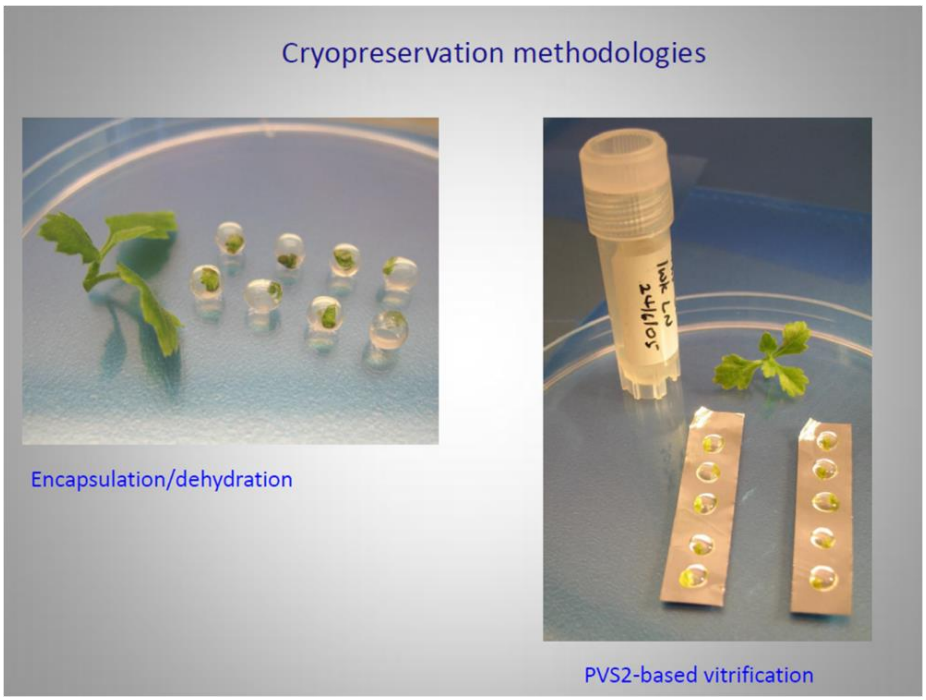
- 122 accessions in a Core Collection
- 2 experts: they spend between 15% and 20% of their time
- In the past 3 years:
    - Incorporation of15 to 20 new clones/year stored in liquid N2
    - 38 batches thawed/year
    - 0,5FTE spent / year
- Today status:
    - 34% - In progress
    - 7% - recalcitrants
    - 59% - In liquid N2

Current application of cryo preservation

- Multi-location evaluation in a core collection - Constitution of IdCards for each clones with common control.
- Germplasm Exchange of material with different areas - Reduction of risk for propagating diseases.
- Reduction variations caused by multiple transplanting.

Paul Hadley, on behalf of Andy Wetten, made a presentation on cocoa cryopreservation at the University of Reading, UK. It included the following points:

Cocoa Cryopreservation

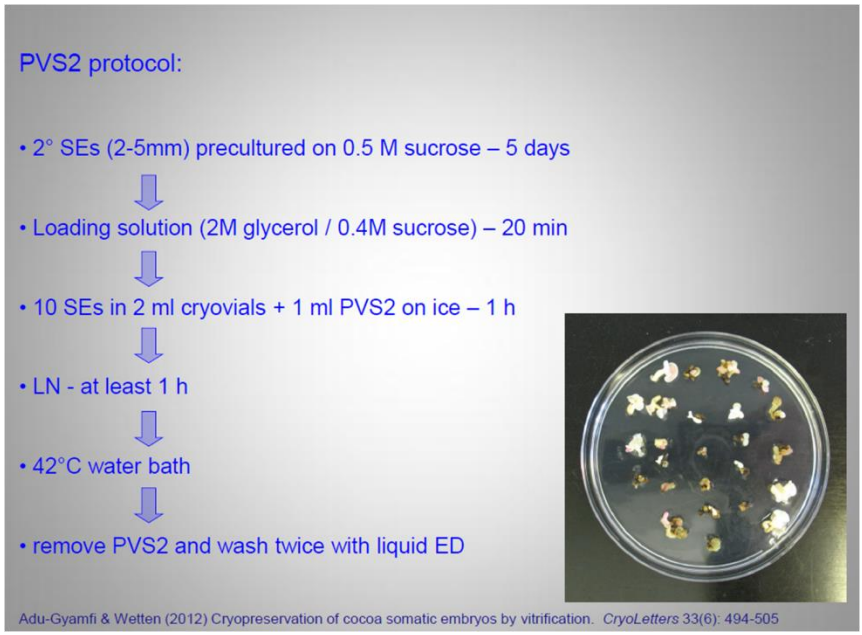


CryoLetters **33** (6), 494-505 (2012)
CryoLetters, businessoffice@cryoletters.org

CRYOPRESERVATION OF COCOA (*Theobroma cacao* L.) SOMATIC EMBRYOS BY VITRIFICATION

Raphael Adu-Gyamfi[1, 2*] and Andy Wetten[1]

[1]School of Biological Sciences, University of Reading, Whiteknights, Reading, RG6 6AS, UK.
[2]Agronomy Department, University for Development Studies, P. O. Box TL 1882, Tamale, Ghana
*Corresponding author e-mail: raphpat205@yahoo.com



GENERAL DISCUSSION

The following point were mentioned and/or raised during the follow-up discussion:

- The cost estimated at 0.5 full-time employment per year is a fairly high cost but comparable to materials in vitro in a lab environments. The advantage of cryopreservation is the long-term conservation of the germplasm.
- The main difference between the clones is the capacity to produce callus, not the cryopreservation treatment. And there are no differences in the different genotypes for regeneration.
- Need to select the best lines and the callus should be from less than 6 months old lines and cell lines of less than a year old.
- This can be done in any lab but it is best to use fresh materials. The costs will be the same of liquid nitrogen and the main thing it the knowledge on how to thaw/cool before storing in liquid nitrogen.
- Nestle maintains the materials as a safety back up.
- Cryopreservation could be used for a core collection as a good starting point.
- A good assessment of the current situation with safety back-up of collection would be an important task.

9. Political Aspects of Accessibility of Germplasm

9.1 Current Situation of Cacao Germplasm Access

As previously highlighted, a considerable range of cacao genetic diversity is currently held in national and international genebanks, but access to these resources is often restricted by the lack of a clear institutional legal and policy framework for the exchange of materials.

At present the global arrangement for the exchange of cacao genetic resources relies mainly on the two international collections held by CATIE and CRC/UWI that have formally placed their cacao collection under the auspices of the Governing Body of the International Treaty. Other collections, such as the one in CIRAD (Montpellier and French Guyana) and USDA, also make their materials available for international distribution. With the exception of these collections, there is little international exchange of germplasm. The rest of the diversity is maintained by many national programmes. Most of these national collections are operating under the premise of the Convention on Biological Diversity (CBD), usually without having specific Access and Benefit Sharing (ABS) legislation in place. This situation has resulted in fragmented approaches, informal exchanges of germplasm and thus, the benefits that are generated in germplasm providing countries are not recognized, at least not to those authorities that make policy decisions in the respective countries.

Much of the cacao genetic diversity is also conserved *in situ* and on farms. These play a crucial part but the sites can be vulnerable and threatened and the resources need to be safety duplicated not to be lost. The main purpose of a genebank collection is to facilitate the use of cacao genetic resources and make it available to breeders for evaluation projects. But the funding of many of the national collections is not secure and it is urgent to secure the resources. For example, the ITC collection in Peru is funded until December 2014 but the future is uncertain. It is challenging to work on very long-term goals but on a 2-year funding. And furthermore, it is challenging to conserve the material when some of the governments do not fully appreciate the value of the resources. Another important aspect is how to transfer these materials for famers. The challenge of conserving and evaluating is important and this is why it is important to separate these activities.

9.2 Proposed Solutions to Facilitate Access and Secure Conservation

It was proposed to look more closely at the Nagoya protocol of the CBD to possibly provide a legal framework for benefit-sharing for the national collection (Full reference: Nagoya Protocol on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits Arising from their Utilization - http://www.cbd.int/abs/about/default.shtml). The Nagoya Protocol is an international agreement which aims at sharing the benefits arising from the utilization of genetic resources in a fair and equitable way, including by appropriate access to genetic resources and by appropriate transfer of relevant technologies, taking into account all rights over those resources and to technologies, and by appropriate funding, thereby contributing to the conservation of biological diversity and the sustainable use of its components. It was adopted by the Conference of the Parties to the Convention on Biological Diversity at its tenth meeting on 29 October 2010 in Nagoya, Japan. The Protocol currently has 54 parties and 92 signatures. It is being implemented in the EU since October 2014.

Given the risks and threats to the many of the national collections, part of the solution would be to secure these as part of the GSCC. And specific terms and conditions for distribution and access could be further developed. For example, at CATIE, some clones are distributed with restricted conditions. New varieties can be distributed but not the second generation. There are different breeding strategies and

in some cases, some of the materials will be lost in the final varieties. The same situation is in Ecuador where F_1 can be used to create varieties but these F_1 cannot be distributed. There are also exchanges of materials for evaluation in other part of the work that have specific conditions and through research permits. It was also proposed that pollen of breeding materials and crosses could be distributed so that the genes could be used but not the genotypes. It would be worth exploring further developing agreements for exchange of materials for research that may have restrictions but would allow securing the resources at the same time.

Another option is to consider safety-duplication agreements as "black-box" agreement where the recipient commits to only maintain and secure the material and not distribute further. For example a country could send materials to CATIE and CRC for safety deposit only. Information may be available in a database but any request for the material would be directed to the donor institute. There could also be a combination of options with black-box safety-duplication and use in a number of very specific breeding programmes.

An assessment could be made, looking at the information from the breeding programme and the pedigrees, to demonstrate how much germplasm has come from other countries in the varieties developed. This is related to demonstrating that no country is self-sufficient when it comes to cacao genetic resources for their own immediate and further needs.

It was proposed that the 2 projects[1] funded by the Common Fund for Commodities (CFC) the International Cocoa Organization (ICCO) and implemented by Bioversity (then IPRGI) might be a good example of agreements between research partners for the sharing of germplasm for evaluation and use. Another example is to look at the current best practices of sharing materials in Asia and Pacific through the regional cocoa breeders group with a very simple material transfer agreement (MTA). This can be further discussed within INGENIC breeders groups as these can provide an important forum to discuss, raise the issues and propose solutions. In the Americas, the newly established cocoa breeders group could be the starting point and can help to convince national authorities to move forward. It may be challenging to go against government policies but in collaboration, things can be proposed and moved forward.

In recognition of the difficulty that some countries face in placing cacao genetic resources in the global public domain, it was suggested that a pragmatic approach could be adopted to allow the exchange of accessions given the many suggestions listed above. A paper, the "Ortinola protocol", including all proposed options could be developed with input from all participants. It would be important to also find out more about the Nagoya Protocol as a possible tool for sharing cacao genetic resources.

[1] *CFC/ICCO/IPGRI Cocoa Germplasm Utilization and Conservation: a Global Approach (1998-2004) and CFC/ICCO/Bioversity Cocoa Productivity and Quality Improvement: a Participatory Approach (2004-2010).*

10. DECISIONS-MAKING PROCESS FOR THE GSCC

The group discussed the general decision-making process mechanism for the GSCC, particularly for the process of refining the collection to improve its efficiency and safety duplication.

The GSCC concept was further clarified. The GSCC is a virtual collection, meaning that it will be the sum of a number of accessions located in different collections including CATIE, CRC and some national collections willing to provide access to the resources.

The key issue with cacao genetic resources is that nobody knows what will be needed in the future and this is the main challenge to form a strategic collection today for the long-term. But the main principle is to preserve the entire cacao genepool through a core set capturing the allelic diversity *(see the proposed list of 261 accessions in **Annex A1**-Annex 7),* and to add to this core set genotypes with traits that are of interest to users. And the most urgent need is to identify the materials that have priority for conservation.

10.1 CAPTURING THE ALLELIC DIVERSITY OF THE ENTIRE GENEPOOL AND GAP FILLING

Regarding the core set capturing the allelic diversity of the entire cacao genepool, the group proposed that the identified 261 accessions be the starting point since there is more information and variability on these materials. As information comes in from new populations and markers, more materials could be included. The methodology used to identify these accessions should be updated since there is better datasets now from Motamayor and verified with what is currently available in CATIE and CRC. It should be compared also to what is only available at CATIE and CRC. This means that the same methodology should be used for the entire diversity and also for what is only in the public domain to identify the gaps (what is there and what is missing). This subset based on allelic diversity can be used to assess what we have and add traits that will be prioritised by the regional cocoa breeders groups.

A discussion followed on the current status of knowledge on the total diversity that exists and the proportion of that diversity in *ex situ* collections. For example, it is estimated that about 50% of these 261 accessions are currently in the 2 international collections. But it there is insufficient information to assess the total diversity. For example in Brazil, only about 20% of the Amazon area is believed to be conserved in *ex situ* collections. In the case of Peru, the proportion is higher, estimated at 80% of the diversity conserved. But it is not entirely clear what these percentages really mean since parts of the diversity in Brazil and in the Amazon is not fully known and there is not necessary access to all the areas to sample the complete genepool.

A follow-up question was how much of the 10 groups described by Motamayor *et al* (2008) is currently in the collections at CATIE and CRC. And it is believed that in many cases these may be only represented by a few trees and only a fraction of the whole populations may be represented. A key task would be to carry out a global diversity analysis to assess what exists in the wild, on-farms and in collections and what part of the global diversity is in the collections at CATIE and CRC including black box agreements. This requires trust and clear and agreed restrictions for use. It was proposed that Bioversity would be well placed with a neutral position to get an agreement between all countries to provide access to study the diversity.

The group stressed the point that collecting needs to continue and a diversity assessment study could guide the gap filling priorities. For example CATIE is expanding to include wild materials as much as possible. For gap filling, GIS expertise is needed to assess where materials have been collected, map out what exists and determine what may be missing including assessing the different ecological zones to

know what has not been collected and where. Information from collecting missions is key for such exercise. It was also noted that gap filling can be with the objectives of completing the diversity and as well as ensuring that specific traits are captured.

It was therefore proposed to resubmit a project to explore specific area for assessing diversity. The national programmes should ensure the security of the diversity and should be encouraged and provided support to do so. It is also needed to streamline what exists and identify criteria to prioritise its conservation based on the assessment of the total diversity that exists. An Expert Panel on Allelic Diversity was proposed to further work on this aspect.

10.2 Capturing the Diversity Based on Key Traits

The purpose of any conservation effort is the use of the diversity. But we need to move towards a more efficient global system. Combining the allelic diversity with additional traits should ensure that we cover at least 90% of the existing diversity.

The first step is to identify the traits and attributes that should be characterised. The second step it to identify the genotypes that have the traits. And the third step is to assess the amount of diversity captured on this basis. It was suggested to also look at populations and their adaptation including locally adapted populations referred to in Thomas *et al*[2].

It was proposed that CacaoNet works closely with INGENIC and the Regional Cocoa Breeders Groups to develop a list of traits. The process that was followed for the CFC/ICCO/Bioversity project could be a good model.

The following actions were proposed in order to prioritise evaluation:

1. Develop a list of all traits
2. Assess what has been characterised in the different genebank collections
3. Assess what has been evaluated in field trials versus evaluated
4. Assess what breeders want from this list involving the regional breeding groups
5. Prioritise based on interest and available information

Using traits such as flavour, CSSV resistance and low cadmium uptake could be used as models to initiate the process. It was proposed that the GSCC focuses on traits that are needed now, as they are based on a clear need. It was also suggested that the risks of prioritisation should be assessed.

The process should also be linked to the Global Cocoa Agenda adopted during the First World Cocoa Conference (WCC2) in Abidjan in October 2012. Its implementation was reviewed during the Second World Cocoa Conference in Amsterdam in June 2014. The follow-up to the Global Cocoa Agenda is the development of National Cocoa Plans. WCC2 identified 8 priority areas as next steps and the following 3 have particular interest to cacao genetic diversity:

1. National Cocoa Plan to undertake an inventory of cocoa resources.
2. Establishment of several thematic working groups, by the ICCO Consultative Board, including one on cocoa genetic resources.
3. International coordination improved by building on existing initiatives such as CacaoNet and INGENIC among others.

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[2] *Thomas E, van Zonneveld, M, Loo J, Hodgkin T, Galluzi G, van Etten J. 2012. Reflections on spatial diversity patterns of Theobroma cacao L. in Tropical America.*

10.3 POSSIBLE APPROACHES TO DEVELOP THE GSCC

It was proposed that the GSCC should aim at capturing all the genetic diversity that exists. There is a need for a process to get us from now to where we want to be. And to do this, there are 2 basic approaches possible to get us moving from a disorganized to an organized set of accessions in the GSCC, each with advantages and disadvantages and each for a different process but both getting to the same result.

From large to small: An approach is to include all that is currently available in the public domain, i.e. at CATIE, CRC and some of the national collections such as USDA and CIRAD. And from this large set, reduce the number of accessions based on information about possible duplications and introducing new diversity that would be made publically available via the GSCC. This approach is starting large and getting to a reduced number of accessions over time. The main advantage of this approach is that priority would be to secure the exiting diversity first with the understanding that as information becomes available, the GSCC would be rationalized to a fairly restricted number of accessions that will not change majorly over time. Donors will be interested in knowing how the GSCC will be rationalised. As the exercise moves forward, donors would be partners in the rationalization and decision-making process. This approach acknowledges that reducing the collection will reply on the existing knowledge of the materials and of the total diversity. If the knowledge does not exist, the collection should not be reduced apart from redundancies. The main disadvantage of this approach is the danger that donors put pressure for restricting the GSCC to a very small set of core germplasm. Furthermore, reducing numbers can be a slow process. Going from large to small, we may lose the ability to find new things.

From small to large: An alternative approach would be to start with a small core collection based on allelic diversity and gradually add materials with traits of interest for which there are sufficient information to determine their current and future value, coming from evaluation trials. Accessions would only be added to the GSCC if not already included and contributing to new diversity. These would be subsets of CATIE and CRC current collections, creating an increased number and capturing the traits. The first cut can be done rapidly but we do not have all the characterisation we may need to determine the additional materials. The main advantage of this approach is that current diversity would be secured while rationalization is being proposed. The potential donors would be encouraged to follow the process towards a more organized and smaller set of accessions that would capture the most important diversity for the future. The main disadvantage of this approach is that it could delay the rationalization process if there is not funding pressure to reduce the number of accessions and get to a more global efficient and cost-effective situation. The risk is also that donors will focus on this small set and not be interested in an increase.

Both of these approaches would present different initial costing. But as the process evolves over time and the GSCC is refined, the costing would be the same with the only difference of either increasing of decreasing in the process. What we end up should be the same, whatever approach is taken.

The industry may be mainly interested in what is available that can be used by breeders. But a lot of other donors may be interested in the whole diversity aspects conserved in national collections first. There are different kinds of donors interested in the different parts of the global system for cacao genetic resources conservation and use. Some will be interested in specific geographical areas and different ecologies such as Peru, Ecuador, Colombia, and Brazil. Some donors will be interested in securing the diversity for climate change adaptation.

Cacao genebanks are expensive because of the number of trees per accessions, not the number of genotypes. The Policy Committee could also make recommendation for re-propagation of collections with a reduced number of trees (4) but with a higher management regime.

A Policy Committee is proposed to oversee the process of proposing materials in the GSCC including bringing things in and the reducing duplication. We are all also aware that curation is about numbers and genebanks need advice on how to reduce and eliminate materials. The process of getting more information and identify redundancies would also require funding. The Committee would draft a policy for managing the materials coming in and out of the GSCC including duplication and with an agreed list of traits for move forward and describe the risks associated to prioritization. The proposed members are: Wilbert Philips, Path Umaharan, Paul Hadley and Michelle End.

11. RECOMMENDATIONS FOR NEXT STEPS AND ACTIONS

Based on the presentations and group discussions, below are the recommendations for next steps and actions proposed.

11.1 SECURING THE EXISTING *EX SITU* CONSERVATION

The highest priority of the 2012 Global Cacao Strategy is to secure the conservation of the genetic diversity currently held in the public domain in *ex situ* collections and facilitate its safe distribution and its safety-duplication.

The Strategy recommends increasing the efficiency of *ex situ* collections to reduce cost and increase sustainability and developing a detailed fund-raising strategy, engaging in dialogues with donors to secure funding for its short-term and longer-term objectives.

CacaoNet established a Task Force on Sustainable Funding Mechanisms for Cacao Genetic Resources focusing on the long-term conservation and use of the Global Cacao Strategic Collection (GSCC), promoting the participation of all partners. The Task Force is convening regularly in conference calls. The Task Force developed communication materials and key messages to be conveyed to potential donors.

There is an important role to be played by all involved in and impacted by cocoa production which is to raise awareness of the importance of cacao genetic resources and how the urgent need for the diversity to be conserved so that it can be accessed and used in breeding programmes.

Aligning with the priorities of the different sectors (farmers, governments, research and industry) is essential. CocoaAction and it priorities to address the challenges of cocoa productivity in West Africa and particularly using genetic diversity to find a solution to CSSV is recommended as a case study that can demonstrate why and how the GSCC should be developed.

It worked closely with CATIE, CRC and ICQCR on developing costing studies, under the expert guidance of Daniela Horna, in order to provide a more detailed and accurate cost estimate of the management of the Global Strategic Cacao Collection (GSCC). The next steps of the CacaoNet Funding Task Force are to approach targeted donors and develop a funding mechanism that would both suit the donors' requirements and the needs for CacaoNet and the scientific community to guide the funding priorities.

The next steps of the Task Force is to fine-tune the costing of the GSCC and start approaching potential donors to discuss the possible terms and conditions that the different types of funders might be interested and willing to contribute to. This feasibility phase will help determine the most appropriate funding mechanism to establish and the partners best placed to manage it for the long-term, taking into consideration the range of funding needs (short-medium-long terms activities).

11.2 PRIORITIES AND NEEDS FROM THE DIFFERENT STAKEHOLDERS

11.2.1 THE INDUSTRY AND COCOAACTION

- CocoaAction, partnerships between the industry, governments and cocoa farmers aims to boost productivity and strengthen community development in Côte d'Ivoire and Ghana.
- The objective is to rejuvenate the cocoa sector at the farm/community level through closer voluntary industry and public-private collaboration, and to align sustainability efforts.
- West Africa is at the center of CocoaAction's attention first, with a focus on Côte d'Ivoire and Ghana and later will be expanded to other cocoa producing countries.
- Workstream 1 on planting materials is of direct relevance to cacao genetic resources work with the 2 areas of focus identified to add value:
    - Propagation: expand propagation capacity of origin countries to produce good quality trees
    - CSSV: the virus needs more attention and focus, e.g. on breeding new tolerant/resistant varieties and detection
- CocoaAction's CSSV program consists of three subprojects:
    1. Identification of existing CSSV resistant materials in national and international collections, optimizing screening methods and screen materials for resistance to CSSV at one European and one Côte d'Ivoire site and participative breeding: identifying CSSV resistant plants which could be used as parents.
    2. Development of protection methods against the mealybug (vector) and local implementation of a robust detection test for the virus
    3. Support breeding for CSSV resistance in Côte d'Ivoire through the development of breeding populations using selected parents, identification of resistant genotypes and development of markers for CSSV tolerance. The program will work towards maintaining current CDI quality and flavour.
- CocoaAction identified CSSV disease management as a priority, to meet predicted consumption needs and therefore resistance to CSSV disease is essential as one of the selection criteria for the GSCC.
- Need to develop a test looking at different varieties, at genotypes within the different varieties and at the associated mealy bugs transmitting CSSV. The characterisation of the virus will also be important.
- The critical role of CATIE, CRC and other collections willing to provide access to their germplasm will be in the identification of materials for multiplication and spread.
- CSSV is the largest threat now and there are no solutions. But this does not preclude work to be carried out on a number of priorities such as moniliasis, black pod, cocoa pod borer etc.
- Opportunity to use CSSV disease resistance as an example of the importance of cacao genetic resources and a case study to raise the awareness of the industry to the importance of genebanks and collections as the long-term solutions to the industry problems. Via CacaoNet, WCC, WCF, we are promoting the importance and vulnerability of cacao genetic resources.
- CocoaAction can have a positive impact on increasing the support to the international collections (CATIE and CRC) and the International Cocoa Quarantine Centre at Reading and securing the conservation of threatened diversity by demonstrating the value of these resources to address an urgent issue such as CSSV in West Africa.
- The process should also be linked to the Global Cocoa Agenda adopted during the First World Cocoa Conference (WCC2) in Abidjan in October 2012.

11.2.2 THE FINE CHOCOLATE INDUSTRY

- Breeding for flavour and for agronomic traits needs to pay particular attention to flavour right from the start and therefore needs to be there at the beginning of the breeding process.
- One of the key sector's issues is the preservation of flavour.
- The Heirloom Cacao Preservation (HCP) collects information on applicants, genotype and flavour characteristic. HCP receives applications and bean samples which is anonymously processed by Guittard Chocolate for flavour analysis by a tasting panel. THCP collaborates with USDA for genetic identification and ongoing research. The aim is to eventually develop protection propagation projects of the varieties.
- Work is carried out on flavour at CATIE and CRC breeding programmes.
- There are flavour attributes developed for tasters and a system of traffic lights used for evaluating materials for their flavour potential in improvement progammes. Green meaning that there is one or more of the suite of valued cacao flavours (cocoa, balanced bitterness/astringency, fruit, floral, nut, and red indicates a problematic inferior flavour (dirty, badly out of balance astringency) that should be removed from breeding unless it has other specific attributes that would override the importance of flavour. The "traffic light" approach is easy but requires judgement. It is mainly about assessing how it would measure if processed into a blend.
- CATIE for example releases clones indicating the flavour characteristics. For example: R-1 has a yield of 1,674 Kg/ha-yr, 15% monilia Losses, 6% black pod Losses and flavour characteristics of fresh fruit note, smooth balanced cocoa, mild bitterness. It evaluates the samples at different fermentation protocols (3-5-6 days) and records the results.
- There are currently no validated markers associated with key flavour attributes and no specific programs in place for marker identification.
- Highly precocious breeding in crosses between heterotic groups. The proposed process is to combine/stack traits such as: black pod resistance, pollen self-compatibility, high fat content, CSSV resistance and high cocoa flavour.
- What is important is high quality varieties with interesting traits and the interactions between processing and inherited quality is key.
- Need a priori knowledge on how the material needs to be processed, considering fermentation and drying protocols and screening to select potential value to manage quality in breeding programmes.
- There are needs and opportunities to combine robustness and flavour and these should be assessed in public domain collections at CATIE, CRC and USDA.
- The different genetic groups at CRC could be assessed and processed as a first step to understand the differences.
- CacaoNet could produce standard protocols for flavour evaluation and Ed Seguine and Darin Sukha could publish a short paper to be used with samples.

11.2.3 THE REGIONAL COCOA BREEDERS GROUPS

- There are many introductions to increase new diversity in the West African breeding programme but the key issue is to know what is being introduced and to understand the diversity that already exist in the region.
- The situation is uneven between the countries in West Africa and between the farmers and there are important gaps.

- Since evaluation is very costly it is recommended to split key clones for evaluation in different countries using the same standards and in West Africa a good example of this is the WCF seed garden output.
- There is an important issue of misidentification in the collections and gardens and the IITA-WCF project with 2,000 samples from the countries is aiming at resolving this issue. The next West Africa Breeding group meeting will discuss how to address the results.
- There are gaps in diversity in local collections and there is a need for information on clones and verification of true-to-type of individuals.
- In Asia and the Pacific, there is exchange of agreed materials for multisite evaluation.
- There is an urgent need to secure the germplasm that is available for breeding and complete the gaps in the collection and carry out actions.
- Most breeding programmes have old sources of resistance and it is hard to make progress.
- We need more information on the heritability of yield component. The Asia Pacific Breeders group collects data on yield. Compatibility is also important in yield and there are examples in Brazil of this.
- The Peru collection carries out a heavy metals extraction trait trials.
- There is a need to share information and methodologies for evaluation and find materials to establish evaluation trials addressing issues such as climate change.
- We need pre-breeding populations that capture diversity and traits that can be used.
- Nobody and no country is self-sufficient when it comes to cacao genetic resources and we need the international collection to have access to the diversity.
- We need to prioritise materials at the International Cocoa Quarantine Centre at Reading (ICQCR) but a process to do so should be proposed.
- We need to find out if the CFC collection is being used and the status of this programme and ensure that the CFC collection goes through the ICQCR.
- The GSCC should develop a list of priority traits for evaluation.
- A key issue is funding support for these activities. We need funds to continue maintaining or continue the evaluation trials.

11.2.4 THE COCOA-PRODUCING COUNTRIES

- The biggest cost for cocoa production is the labour cost. Therefore traits that reduce this cost are needed and may be even more important than yield.
- We need to develop smaller plants. For example, moniliasis depends on size.
- The shape of the tree is also important and should make it easy for mechanisation.
- In Trinidad and Tobago for example the cost of labour is very high and therefore less pruning is a key factor. Witches broom is under control now so labour cost is the most important thing.
- In Brazil the cost of post harvest is estimated at 30% of the total cost.
- Management efficiency, uniformity of production and harvest is very important.
- Bean size and fat content is important for international markets but pest and diseases are more important economically.
- The minimum bean size and fat content for industry requirement is 1g/bean which is the benchmark across the world.
- Number of beans per pod and beans to husk ratio is important.
- Cocoa butter content is at the top, and then high flavonoids/polyphenols and cadmium content are some of the priorities. In the Caribbean, cadmium is a very important issue.
- Small chocolate companies may have different requirements.

- Other important factors are related to adaptability to grow in specific, new and/or marginal areas affected by conditions such as drought. In Trinidad and Tobago for example, cocoa is being pushed into lands less suitable for it that require high fertilisers and other inputs. We need criteria for ability to extract nutrients for more efficiency.
- We need to breed for common pest and diseases such as CSSV and megakarya in West Africa but also preventive breeding for pests and diseases that may come one day from other regions.
- We need agreement on the evaluation of selected physiological characters and look at the ICQCR materials with this focus. Resources are needed to do this and develop evaluation projects.
- The target should be to preserve all the existing diversity. This is the challenge since we do not know the future.
- We need to prioritise the morphological and molecular characterisation of germplasm.
- We need to agree on a number of priority traits relevant to the following categories and if these are global or region-specific.
- Screening methods are needed to identify a subset of materials for further evaluation. We need to also distinguish between identifying a snapshot and when deep thorough evaluation has been carried out.
- We may need to also consider what is easy to measure even if not relevant to all.
- Should consider the totality of the cocoa world, the key issues for sustainability and not only what is driving a part of the market today.
- We need to identify core sets for pests and diseases resistance, for flavour and for a range of key characteristics.

11.3 Next Steps on Characterisation and Evaluation of Key Traits

11.3.1 Morphological characterisation

The value of morphological characterization is mainly for the following:

- Identification of accessions
- Assessment of phenotypic diversity (assignment to recognized groupings and identification of range of variation)
- Detection of duplicates or mislabeled accessions
- Preliminary evaluation of germplasm through assessment of traits of interest to breeders
- Facilitation of the utilization of genetic resources within the genebank (through breeding & germplasm enhancement)

There are 25 morphological descriptors used by CRC for characterisation of flowers, fruits and seeds and leaf flush.

Traits of economic interest are measured such as the following:

- Yield potential – Pod index (the number of pods required to produce 1 kg of dried cocoa)
- Cotyledon weight and size
- Seed/Bean number
- Disease and pest resistance
- Flavour, quality, butterfat content
- Abiotic stress resistance

There is a good source of potentially high-yielding genotypes within the international collections.

The proposed actions are:

- Verify the authenticity of the trees from which data were collated using molecular markers such as SNPs;
- Complete morphological characterization of uncharacterized and misidentified accessions;
- Compare phenotypic and genetic diversity of germplasm. Such a combined analysis will facilitate consideration of the value of morphological descriptors in the area of molecular genetics, and the list of morphological descriptors for routine characterisation within cacao collections may be modified;
- Pursue genome wide trait association studies (GWAS).

11.3.2 Evaluation of disease resistance traits

Evaluation of disease traits is a routine activity at CATIE and an essential part of the breeding strategy. There are 3 major processes used:

- Paired crosses between selected clones from the international collection go through progeny trial and the selection of the best progenies go to regional trials, observation plots and clonal gardens. And then go to farmers.
- The selection of the best clones from the international collection go through clonal trials. The best clones go to regional trials, observation plots and clonal gardens. And then go to farmers.
- Paired crosses between non-consanguineous clones go through progeny trials. Selections of the best trees from the progeny trials go for clonal propagation and clonal trials. The best clones go to regional trials, observation plots and clonal gardens. And then go to farmers.

11.3.3 Evaluation of Flavour and other quality traits

- We need to evaluate flavour in selecting clones for the GSCC because there is a genetic flavour potential from each bean/variety and rich flavour diversity that needs to be captured and preserved.
- Flavour evaluation is defined as a scientific method used to evoke, measure, analyze, and interpret those responses to products as perceived through the senses of sight, smell, touch, taste and hearing.
- This evaluation is carried out by trained tasters as a panel or as individuals.
- The methodology uses robust sensory design with sample coding, randomization, repetition and flavour descriptors.
- Other quality traits include: physical bean characteristics, cocoa butter%, FA profile and melting point, pulp flavour, fermentation requirements and nutraceuticals such as total phenolics, theobromine, caffeine, catechins, epicatechins, flavanols and antioxidant capacity.

11.3.4 Yield Components and Resilience to Abiotic Stress

Internal determinant of yield in cocoa include:

- Leaf area index
- Canopy architecture
- Light interception
- Leaf photosynthesis
- Canopy photosynthesis
- Biomass partitioning

Environmental factors impacting on leaf photosynthesis

- Light intensity
- Temperature
- Water stress
- CO_2

Managerial determinants

- Shade impacting on light intensity and leaf photosynthesis
- Fertilisers and planting density impacting on leaf photosynthesis
- Pruning impact on canopy architecture

Physiology with Environment

- Genotypic variation in photosynthetic rate
- Water use efficiency – genotypic variation in stomatal index
- Harvest index – yield partitioning
- Pod index
- Fat content
- Genotypic characteristics under different environmental regimes - climate change - drought
- Productivity and sustainability under stress
- Plasticity and stability of responses

The following points were mentioned and/or raised during the follow-up discussion:

- Stomatal index is simple to measure.
- Portable photosynthesis equipment can be used to make the first cut, i.e. the top 20% of varieties, to carry out further evaluation trials.

PROPOSED ACTIONS:

- Develop a short list of the most important characteristics to measure in the context of the GSCC and the equipments to be used. Paul Hadley and Andrew Daymond could do this. Could use the INGENIC newsletter to publish and consult.
- Need to develop a targeted programme to move forward, indentifying what can be done: (1) in the genebank (characterisation) and (2) in evaluation trials.

Table proposed for starting the process to list and agree on key traits for priority evaluation and inclusion in the GSCC.

Categories	Traits	Global	Region-specific	Ease of measurement
Quality				
Flavour				
Productivity				
Pests				
Diseases				
Yield				
Adaptability to new areas				
Climate change adaptation				
Heavy metals - Cadmium				
Other?				

11.4 Decision-Making Process for the Global Strategic Cacao Collection (GSCC)

11.4.1 Capturing the Allelic Diversity in the GSCC

- Wide variation for disease resistance and quality exists in *ex situ* collections and in farmers' fields but its use it not optimised. Unique and valuable material is conserved in CATIE, CRC and in national collections and thus, collaboration is needed to secure this material and increase the access to and their use in breeding programmes.
- The group proposed that the identified 261 accessions capturing the allelic diversity of the entire cacao genepool be the starting point since there is more information and variability on these materials. As information comes in from new populations and markers, more materials could be included.
- The methodology used to identify these 261 accessions should be updated since there is better datasets now from Motamayor and verified with what is currently available in CATIE and CRC. It should be compared also to what is only available at CATIE and CRC. This means that the same methodology should be used for the entire diversity and also for what is only in the public domain to identify the gaps (what is there and what is missing).
- This subset based on allelic diversity can be used to assess what we have and add traits that will be prioritised by the regional cocoa breeders groups.

11.4.2 Capturing the Diversity Based on Key Traits

- It was proposed that CacaoNet works closely with INGENIC and the Regional Cocoa Breeders Groups to develop a list of traits. The process that was followed for the CFC/ICCO/Bioversity project could be a good model.
- The following actions were proposed:
  - Develop a list of all the traits of interest
  - Assess what has been characterised in the different genebank collections
  - Assess what has been evaluated in field trials versus evaluated
  - Assess what breeders want from this list involving the regional breeding groups
  - Prioritise based on interest and available information and identify the genotypes that have the priority traits for inclusion in the GSCC.
  - Assess the amount of diversity captured on this basis.
- Using traits such as flavour, CSSV and cadmium could be used as models to initiate the process. It was also suggested that the risks of prioritization should be assessed.

11.4.3 Assessing the Genetic Diversity Conserved in Ex Situ Collections

- It was proposed to carry out a global diversity analysis to assess what exists in the wild, on-farms and in *ex situ* collections since the total diversity is not known.
- From the total diversity, the next step would be to identify those publically maintained unique accessions in the 2 international collections at CRC/UWI and CATIE (including black box agreements).
- Assessing the cacao genepool in the centre of diversity in situ and on-farm would be as a priority. It was proposed to resubmit a project to explore specific area for assessing diversity.
- It was proposed to assess how many of the 10 groups described by Motamayor *et al* (2008) are currently in the collections at CATIE and CRC, including many individuals are representing the populations.

- The group stressed the point that collecting and filling in the gaps should be continued based on these diversity assessment studies. GIS expertise is needed to assess where materials have been collected, map out what exists and determine what may be missing including assessing the different ecological zones to know what has not been collected and where. Information from collecting missions is key for such exercise.
- An Expert Panel on Allelic Diversity was proposed to further work on this aspect.
- This requires trust and clear and agreed restrictions for use. It was proposed that Bioversity would be well placed with a neutral position to get an agreement between all countries to provide access to study the diversity.

11.4.4 Ensuring the Safety-Duplication of the GSCC

- Most collections have duplications internally and with other collections but o a few have strategic safety duplication of unique materials.
- The safety-duplication of the GSCC materials should be agreed and could be maintained in a different field genebanks (preferably in a different country) and/or via cryopreservation.
- The Nestlé Research and Development Centre, Tours, France has a strong expertise in cryopreservation on different species including cacao and with different explants. They currently maintain a core collection of 122 accessions. And they have 2 experts that spend between 15% and 20% of their time (also estimated at 0,5FTE spent / year) on cryo-preservation activities. The clones are used for multi-location evaluation and exchange of materials in different areas (reduction of risk for propagating diseases and of variations caused by multiple transplanting).
- Cacao cryopreservation activities and research also takes place at the University of Reading, UK. The technique is used to cryopreserve frequently requested clones at the ICQC,R and to back up the collection.
- The main difference between the clones is the capacity to produce callus, not the cryopreservation treatment. And there are no differences in the different genotypes for regeneration.
- The cost estimated at 0.5 full-time employment per year is a fairly high cost but the advantage is the long-term conservation of the germplasm.
- Cryopreservation could be used for a core collection as a good starting point.
- A good assessment of the current situation with safety back-up of collection would be an important task.

11.4.5 Possible Approaches to develop the GSCC

- There was an agreement that the GSCC should aim at capturing all the genetic diversity that exist.
- There are 2 basic approaches possible with advantages and disadvantages and each for a different process but what we end up should be the same, whatever approach is taken. Both approaches would have different initial costing but the final result and costing should be the same.
    1. <u>From large to small:</u> An approach is to include all that is currently available in the public domain, i.e. at CATIE, CRC and some of the national collections such as USDA and CIRAD. And from this large set, reduce the number of accessions based on information about possible duplications and introducing new diversity that would be made publically available via the GSCC.
    2. <u>From small to large:</u> An alternative approach would be to start with a small core collection based on allelic diversity and gradually add materials with traits of interest for which there are sufficient information to determine their current and future value, coming from

evaluation trials. Accessions would only be added to the GSCC if not already included and contributing to new diversity.

- There are different kinds of donors interested in the different parts of the global system for cacao genetic resources conservation and use. The industry may be mainly interested in what is available that can be used by breeders. Some will be interested in specific geographical areas and different ecologies such as Peru, Ecuador, Colombia, and Brazil. Some donors will be interested in securing the diversity for climate change adaptation. Some will be interested in the whole diversity conserved in national collections first.
- Cacao genebanks are expensive mainly because of the number of trees per accessions, not because of the total of genotypes or accessions.
- The process of getting more information and identify redundancies would also require funding.
- A Policy Committee is proposed to oversee the process of proposing materials in the GSCC including bringing things in and the reducing duplication.
- The Committee would draft a policy for managing the materials coming in and out of the GSCC including duplication and with an agreed list of traits for move forward and describe the risks associated to prioritization.
- The Policy Committee could also make recommendation for re-propagation of collections with a reduced number of trees (4) but with a higher management regime.
- The proposed members are: Wilbert Philips, Path Umaharan, Paul Hadley and Michelle End.

11.4.6 Looking at Other Crops as Possible Models

- The development of the GSCC could learn from what has been done for a number of other crops such as the collections in the CGIAR and the UK National Fruit Collection among others.
- The CGIAR Global Collections for Long-Term Conservation have agreed to basic principles that must be met for crop genetic resources collections to be eligible for funding such as the resources should be accessible under internationally agreed terms of access and benefit sharing by the ITPGRFA and set out in the Standard Material Transfer Agreement (SMTA), the holders commit themselves to long-term conservation and availability and the recipient of funds will work in partnership to develop an efficient and effective global conservation system.
- The decision-making processes for efficient genebank conservation and evaluation of the UK National Fruit Collection can also provide a useful guide, and particularly its difference organisation mechanisms. For example the scientific curation, access to the public, management and policy committees, the National Fruit Collection Trust and the supporters club and the safety back up in cryo.
- The collections at USDA could also provide a useful model of management and networking between the different sites and crops.
- In all of the cases (CGIAR crops and the UK Fruit collections), curational and evaluation activities are separated and funded by different funding streams.
- A clear accession management policy (what comes in and what goes out) is vital to the efficient development of a germplasm collection.
- Cacao and the GSCC should learn from the CGIAR In-Trust materials and how they are managed and the use of the Standard Material Transfer Agreement.
- The exclusion of materials from the GSCC needs to be done very carefully as it could include promising materials particularly vis a vis flavour.
- The GSCC Committee will need to address how to prioritise evaluation and link to accessions based on the main criteria and molecular and morphological characteristics.

- The public can provide a strong support for the conservation of the collections as seen with the UK Fruit Collections.
- The long-term funding mechanisms of the CGIAR crops via the Global Crop Diversity Trust (endowment fund) and of the UK Fruit collections are useful models and lessons learn for cacao genetic resources.

11.4.7 COSTING STUDY OF THE GSCC

- The CacaoNet Funding Task Force is working with CATIE, CRC and ICQCR on developing costing studies, under the expert guidance of Daniela Horna, to provide a more detailed and accurate cost estimate of the management of the Global Strategic Cacao Collection (GSCC).
- The detailed costing study of the GSCC should focus on the conservation costs but also include associated services such as germplasm evaluation, quarantine, virus-indexing, distribution and documentation.
- The costing is done using the following Decision Support Tool developed for a range of crops and adapted for cacao for this exercise.
- The main purpose of the GSCC costing study is to have a clear understand of the financial requirements over the long-term and to put an estimated figure on the current value of this diversity. It allows the CacaoNet Funding Task Force also to develop the different funding mechanism required to cover the range of conservation activities and associated services.
- Costing genebank operations is a very different exercise than budgeting. Costing is what the ideal situation would require. Budgeting is what could be afforded of this ideal situation based on fund availability.
- The costing exercise also allowed a comparison to be made between the operations at CATIE and CRC and a clear understanding of their uniqueness, similarities and differences.
- The GSCC costing includes annual costs (recurring and one-off) and recurring costs at perpetuity for the development of an endowment fund.
- The costs of conservation between *ex situ* collections will mainly vary based on the total number of accessions and the number of trees per accessions.
- It is important to separate the strictly long-term conservation operation from the activities that bring value to the material such as evaluation and research.
- The annual recurring cost of conserving of 2,500 accessions (estimated size of the GSCC) is calculated at 636,332 USD per year. The perpetuity cost means that an endowment of approximately 15 M USD would be needed to generate the interest to cover the annual costs for ever (at perpetuity). These are draft figures that provide an estimate of the resources needed for the long-term conservation of the GSCC.
- The safety-duplication of the GSCC estimated at a core set of 500 accessions in cryopreservation and 1,000 accessions duplicated in other field collections is estimated at 154,194 USD recurring annual cost. This would require an endowment of 3,658,595 USD to cover the annual cost at perpetuity.
- The data collected and the tool used allow the exploration of different scenarios based on the current information.
- The funding strategy would need to define the different components to be funded. It is recommended that only the essential long-term conservation operations should target an endowment fund. Other activities such as evaluation and other research should be funded on a project basis. It may be interesting to also explore how the collection could be self funded by having some of the services paid for.

- CATIE and CRC are conserved so far because of all the efforts so far (projects, sales etc) but the funds need to be secured. The total cost reflects how much people are willing to pay for it.
- A draft GSCC costing report is being finalised and will be reviewed by the CacaoNet Task Force members shortly. Once agreed the GSCC funding requirements will be communicated and further discussed with a wider group of people.

11.4.8 Information on Evaluation Contributing to the GSCC Decision-Making Process

- Improving the documentation and sharing of information on germplasm is a key element of the Global Strategy for the conservation and use of cacao genetic resources.
- The GSCC information portal, the CacaoNet Germplasm Information System (CANGIS), will draw mainly on the information from the International Cocoa Germplasm Database (ICGD), the international collections held by CRC/UWI and CATIE and at ICQC,R.
- The help of all cacao germplasm collection curators and breeders is needed to increase the amount of information available to aid the decision making processes for the GSCC.
- Some of the cacao germplasm collections may benefit from adopting a more robust germplasm management documentation system such as the global plant genebank information management system GRIN-GLOBAL developed by USDA. GRIN-Global enables genebanks to store and manage information associated with plant genetic resources (or germplasm) and deliver that information globally. GRIN-Global is developed with free or open source software and its source code is available, and is being tested at the USDA cacao collection of Mayaguez, Porto Rico.
- An important task would therefore be to assess the suitability of adopting GRIN-Global by assessing minimum level of local expertise and IT equipment needed and the training requirements for initial set up (customization).
- In order to support the decision-making process of the GSCC, information on evaluation will need to be compiled from all available sources, supported by molecular verification of genotypes where possible. Tools such as the Integrated Breeding Platform (IBP) crop ontologies, Trait Dictionaries and Breeding Management System (BMS) of the Generation Challenge Programme of the CGIAR (GCP) as well as the AgroGen Breeding Management System from CEPLAC are available to improve the efficiency of plant breeding programmes by enabling breeders to access modern breeding technologies, materials and related information in a centralised, integrated and practical manner. These tools therefore facilitate the documentation of breeding trials and the sharing of information on evaluation.
- The GCP/IBP developed breeding management systems for 9 crops (bean, cassava, chickpea, cowpea, groundnut, maize, rice, sorghum and wheat) supported by crop groups and community of practice involving private sector. It provides a workbench, expertise, services, training and capacity building (in partnership with Universities). It is being developed for 5 more crops (barley, lentil, potato, soya beans and sweet potato).
- The GCP/IBP has been approached to include cacao and is interested in making all the tools available to the cacao community for a possible development. There are a number of IBP regional hubs providing support for development and a few already established in West Africa.
- The first step and product to develop a dictionary of traits including the methods and scales of measurement to support the integration of genetic and phenotypic data and the creation of fieldbooks. These are done through providing a forum for scientists to discuss their variables across disciplines and regions and come to a consensus.

- Creating a cacao breeding management system and fieldbook would entail building an implementation team, developing a plan with resources, setting-up customized programme resources and deploying the BMS in day to day operations.
- The proposed next step is therefore to developing the trait dictionary in consultation with a small group interested in its application and follow up in key meetings and workshops.
- INGENIC and the Regional Cocoa Breeders Groups in the Americas, Africa, Asia and the Pacific are best placed to guide the development of such tools and support a network of field trials participating in the evaluation of the GSCC materials at multiple sites.

11.4.9 Political Aspects of Accessibility of Germplasm

- The two international cacao collections at CATIE and CRC have formally placed their collection under the auspices of the Governing Body of the International Treaty. Other collections, such as the ones in CIRAD and USDA, also make their materials available for international distribution.
- Most of the national collections however are operating under the premise of the Convention on Biological Diversity (CBD), usually without having specific Access and Benefit Sharing (ABS) legislation in place. This situation has resulted in fragmented approaches and informal exchanges of germplasm.
- It was proposed to look more closely at the Nagoya protocol of the CBD (on Access to Genetic Resources and the Fair and Equitable Sharing of Benefits) to possibly provide a legal framework for benefit-sharing for the national collection. The Nagoya Protocol has 54 parties and 92 signatures and is being implemented in the EU since October 2014.
- The funding of many of the national collections is not secure. And some governments do not fully appreciate the value of the resources. Given the risks and threats to the many of the national collections, part of the solution would be to secure these as part of the GSCC. And specific terms and conditions for distribution and access could be further developed following examples of exchanges of materials for evaluation and develop agreements for exchange of materials for research that may have restrictions but would allow securing the resources at the same time. It was also proposed that pollen could be distributed of breeding materials and crosses so that the genes could be used but not the genotypes.
- Safety-duplication agreements as “black-box” should be further explored to urgently secure the material. There could also be a combination of options with black-box safety-duplication and use in a number of very specific breeding programmes.
- An assessment could be made, looking at the information from the breeding programme and the pedigrees, to demonstrate how much germplasm has come from other countries in the varieties developed.
- It was proposed that the 2 CFC/ICCO/Bioversity projects might be a good example of agreements between research partners for the sharing of germplasm for evaluation and use.
- Another example is to look at the current best practices of sharing materials in Asia and Pacific through the regional cocoa breeders group with a very simple material transfer agreement (MTA).
- This can be further discussed within INGENIC breeders groups as these can provide an important forum to discuss, raise the issues and propose solutions.
- A paper, the “Ortinola protocol”, including all proposed options could be developed with input from all participants.

11.5 WORKING GROUPS AND COORDINATION OF ACTIONS

CacaoNet Coordination will ensure the overall agreement on the establishment of the GSCC and its functions on behalf of all members. It was proposed that the following 5 thematic Working Groups be established to carry out preliminary work on behalf of all members and propose the next steps (tasks with proposed names and timelines). Each Group should develop a plan of what needs to be done with current resources and what additional tasks and resources would be needed. It was recommended to learn from other species and how they dealt with some of the issues raised. The work to be carried should take into considerations the needs and priorities of the farmers/producers, the producing countries, the regional cocoa breeders groups and the industry small and large as described in ***Section 6*** and summarized in ***Section 11.2***.

A - Best Practices Working Group

- Make recommendations on cacao germplasm management standards for the GSCC - see list of cacao genebank operations in ***Section 8.2*****.**
- This would include characterization and documentation such as the development of standard morphological descriptors and the adoption of a robust genebank information system and linking to the ICGD and CANGIS described in ***Sections 7.1 and 7.2.***
- See next steps proposed in ***Sections 11.3.1 and 11.4.8***

B - Diversity and Gap Filling Working Group

- Propose a way to assess the entire genetic diversity *in situ*, on-farm and in *ex situ* collections to know what is secured, and what are the gaps in *ex situ* collections.
- Prioritize of the conservation of the allelic diversity for the GSCC by reviewing the methodology and recommendations for actions.
- It was proposed that it includes all *Theobroma* species.
- See next steps proposed in ***Sections 10.1, 11.4.1 and 11.4.3***

C – Evaluation Traits Working Group

- Work closely with INGENIC and the Regional Cocoa Breeders Groups to identify the key evaluation traits for selecting materials in the GSCC, involving the ICGD, breeders, industry, curators and physiologists and provide feedback on what to prioritize and the gaps to fill.
- Assess the physiological characters of materials currently in the International Cocoa Quarantine Centre at Reading.
- This group should link to other crops to learn from other groups and assess tools such as the Integrated Breeding Platform (IBP) and the Breeding Management System (BMS).
- See next steps proposed in ***Sections 10.3, 11.3 and 11.4***

D - Policy Working Group

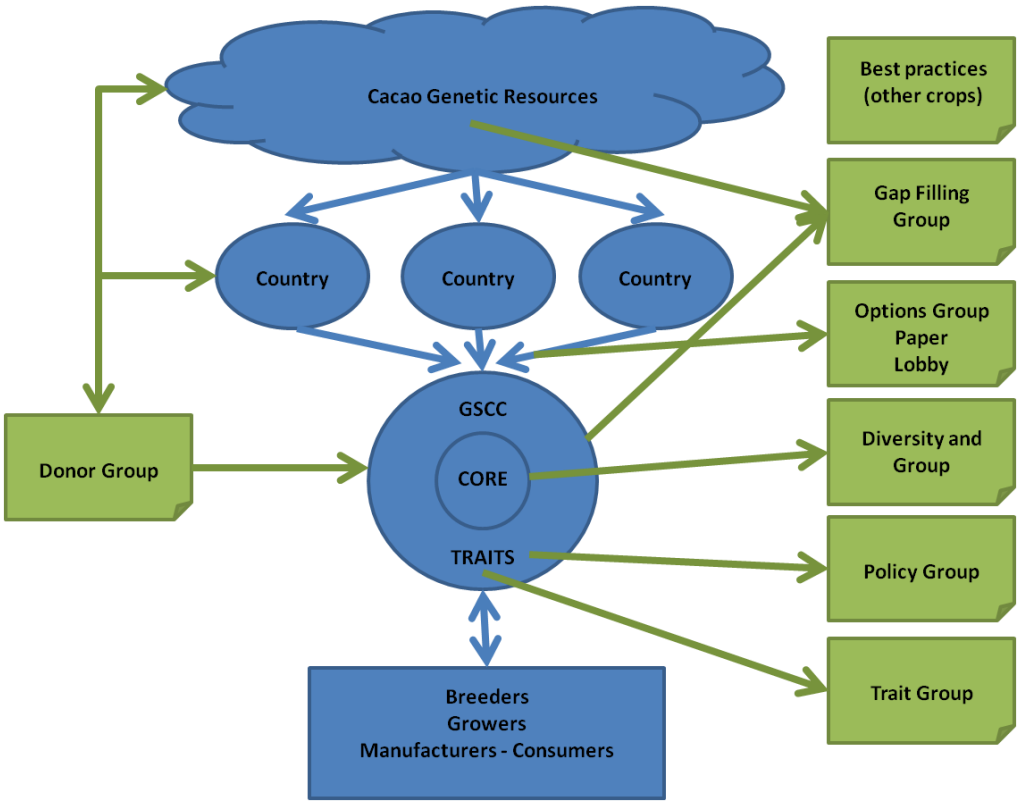
- Assess the different policy options and propose solutions to promote the accessibility and availability of material in the public domain and participation in evaluation trials of GSCC materials.
- Engage with national collections, FAO and the International Treaty to promote the placing of germplasm, particularly the accessions identified for inclusion in the GSCC, in the public domain.
- Review the 2 main approaches for developing the GSCC including the pros and cons of starting from a small to a larger set of accessions for the GSCC and vice versa.
- Collaboration for the long-term safety-duplication of the GSCC materials including the use of cryopreservation.

- See the work described in ***Sections 8.3, 9 and 10*** and the next steps proposed in ***Sections 11.4.4, 11.4.5 and 11.4.9.***

E - Donor Group Working Group

- Secure the *ex situ* conservation and engage in fundraising for the implementation of the Global Strategy, including involvement of the private sector and international funding agencies to leverage funding for cacao genetic resources and establish of an endowment fund.
- The work of this group involves fine-tuning the costing exercise of the GSCC – see Sections 8.2 and
- This group needs to indentify the donors and target the messages.
- This work is currently being coordinated by the CacaoNet Task Force on Sustainable Funding Mechanisms for Cacao Genetic Resources –
- See the work described in ***Section 8.1*** and the next steps proposed in ***Sections 11.4.7 and 11.4.9.***

Drawing summarising the next steps – as presented by Uma



ANNEX A. EXTRACTS FROM THE GLOBAL CACAO STRATEGY RELEVANT TO THE GSCC

The following Annexes are extracts from the detailed long version of the Global Strategy for the Conservation and Use of Cacao Genetic Resources, coordinated by CacaoNet and published at the end of 2012. The specific goal is to optimize the conservation and maximize the use of cacao genetic resources as the foundation of a sustainable cocoa economy. This it does by bringing together national and international players in public and private sectors.

- *The full version of the Global Strategy can be downloaded at:* ***Full document*** *(PDF 2.4Mb)*
- *A booklet version of the Global Strategy can be downloaded at:* ***Booklet*** *(PDF 2.5Mb)*

ANNEX A1 – GLOBAL STRATEGY SECTION 3.2 DEVELOPING THE GLOBAL STRATEGIC CACAO COLLECTION (GSCC) – *WHERE WE ARE NOW*

Only the two international collections at CATIE and CRC/UWI have placed their cacao germplasm under the auspices of the Governing Body of the ITPGRFA, with the commitment to safely conserve for the long term according to international standards and make the materials readily available to any plant breeding programmes and other *bona fide* users. However, some national collections can also be considered to be within the public domain, such as the collections at CIRAD and USDA. In addition, the ICQC,R holds cacao accessions available in the public domain and is the only international quarantine centre for the safe movement of cacao genetic resources throughout the world.

The remaining collections are considered to be a national asset and are generally not publicly available outside of the country holding the collection. In many Latin American countries possessing primary sources of cacao genetic diversity, policies restrict opportunities for newly collected materials to be put into the public domain. Unique and valuable material is conserved in these national collections and thus, collaboration is needed to secure this material and increase the access to and their use in breeding programmes.

But most importantly, no country is self-sufficient when it comes to the range of genetic diversity needed to develop improved materials. This diversity is maintained by several research institutes but only a part of that diversity is in the public domain.

CacaoNet is working towards the establishment of a Global Strategic Cacao Collection (GSCC) as a virtual collection consisting of materials that have been identified as unique and interesting. Each of the participating institutes will agree to conserve these accessions according to agreed practices and standards and make them readily available to any *bona fide* user. The objective of the GSCC is therefore to ensure the cost-effective and efficient long-term *ex situ* conservation of the entire *Theobroma* genepool and its accessibility to all current and future users. The formation of the GSCC will result from a coordinated effort of characterization and rationalization of available cacao genetic resources. The materials in the CATIE and CRC/UWI collections will form the backbone of this GSCC complemented with priority accessions from national collections.

Agreed criteria such as genetic diversity, in the form of allelic richness and the uniqueness of each genotype, in combination with measures of agronomic value will be used to identify priority accessions. Once the main part of the GSCC is formed, adding new diversity will be based on ensuring the genotype

significantly increases the genetic diversity of the GSCC and/or this genotype has specific agronomic, quality or physiological traits that are of interest to users.

A first set of accessions will be selected on the basis of capturing the greatest possible range of allelic richness. These accessions would preferably be in the public domain but it is acknowledged that currently some may be maintained in collections not yet in the public domain. The Global Strategy aims to ensure that the institutes managing these accessions would conserve them for the long-term, evaluate them and take the necessary steps to make them publically available. Two agreed core selection methods are employed. Both use grouping and selection of an optimum set of accessions followed by a further iteration designed to reduce the redundancy of the core selection (*van Raamsdonk & Wijnker, 2000; van Treuren et al., 2008 & 2009*). The method is illustrated by way of reference to a large public dataset of simple sequence repeat (SSR) information (microsatellite markers). The detail of the method is included in *Annex 6* (Description of the agreed methodology to select accessions based on allelic diversity). It is now possible to characterize gene sequence and nucleotide polymorphism at large numbers of loci. Following a comprehensive literature review to identify those traits known to be under well-defined allelic control, the part of the GSCC based on allelic diversity should be subject to a detailed analysis of gene coding and regulatory regions, to catalogue the variation present at those loci in order to encourage the exploitation of this resource. However, the method is not limited to the analysis of molecular genetics data but can combine both discrete categorical and continuous variable data on botanical or agronomic traits. The GSCC currently identifies 261 accessions that represent the maximum allelic richness observed across ten population groups, capturing the majority of the known genetic and geographic diversity held within *ex situ* collections worldwide. The proposed accessions are listed in *Annex 7* (Membership of accession for the GSCC based on allelic diversity). Also, considering that some of these collections have accumulated a large amount of data (e.g. 70% of CEPEC`s collection was well evaluated for several years for resistance to witches' broom disease and many yield components) it is an opportunity to find associations between markers and important traits.

A further set of accessions will be selected on the basis of key traits of interest to users such as yield, flavour characteristics and disease resistance for which agreed criteria will be developed. Criteria for selection of genotypes may include in addition to the number of desirable traits present, the genetic diversity amongst the selected types as determined through DNA fingerprints. This part of the GSCC will complement the part selected on allelic diversity and be a dynamic and geographically dispersed collection composed primarily of wild species and populations, landraces, enhanced populations for which characterization and evaluation data is available and used to broaden the basis on which the selection is made. This material will be in the public domain and accessible in the collections at CRC/UWI and CATIE for which considerable characterization and evaluation data are already available. Additional materials from national collections will become part of the GSCC if the governments concerned are willing to place them in the public domain. A first step will be the development of a list of priority genotypes identified with known agronomic/economic value. This set would be dynamic in nature, adapting to current and potential future needs.

As far as possible, genetically similar genotypes should be avoided to reduce redundancy. There may not be sufficient information on all publically available accessions to allow the identification of priority materials for the GSCC, so a comprehensive assessment of individual identity, verification of a given accession to be true-to-type and population structure, are high priority tasks. The assessment of the complementarities and duplications between the two international collections should be a priority for the establishment of the GSCC.

Since information on accessions is likely to continuously improve over time, the GSCC should be a dynamic evolving collection, to which individuals may be added or, if really necessary, deleted from time to time.

New technologies for cacao, such as *in vitro* culture and cryopreservation, could be used to complement field genebanks to ensure the duplication of the GSCC providing they can be shown to be cost-effective. Cryopreservation is a technology that is far enough advanced to be applied to a large number of genotypes, despite some genotype-dependency for somatic embryogenesis. However, the method is expensive so the application of this method might initially focus on the safety duplication of priority accessions. CacaoNet would lead the process of consultation on safety-duplication and the use of cryopreservation with all its members and develop agreements on behalf of its members.

The accessions within the GSCC should be prioritized for distribution to other international, regional or national genebanks as well as to breeders, via intermediate quarantine. The collection managers and breeders around the world will be responsible for comprehensive characterization, evaluation and further researching of the GSCC collection. All related information should be made available to all users through GSCC information portal to allow the selection of materials for inclusion in the GSCC (*see details in Section 3.7*).

The specific criteria and boundary for each set of accessions would be agreed through a consultation process coordinated by CacaoNet. This assessment would be part of a rationalization plan, with clear objectives, that would take place over time as knowledge becomes available. CacaoNet members would be responsible for the composition of the GSCC as well as for recommending and where possible supporting priority actions such as detecting mislabelling, evaluation, characterization, pre-breeding, distribution and use. This would include the participation of collection curators and the breeding community represented by INGENIC.

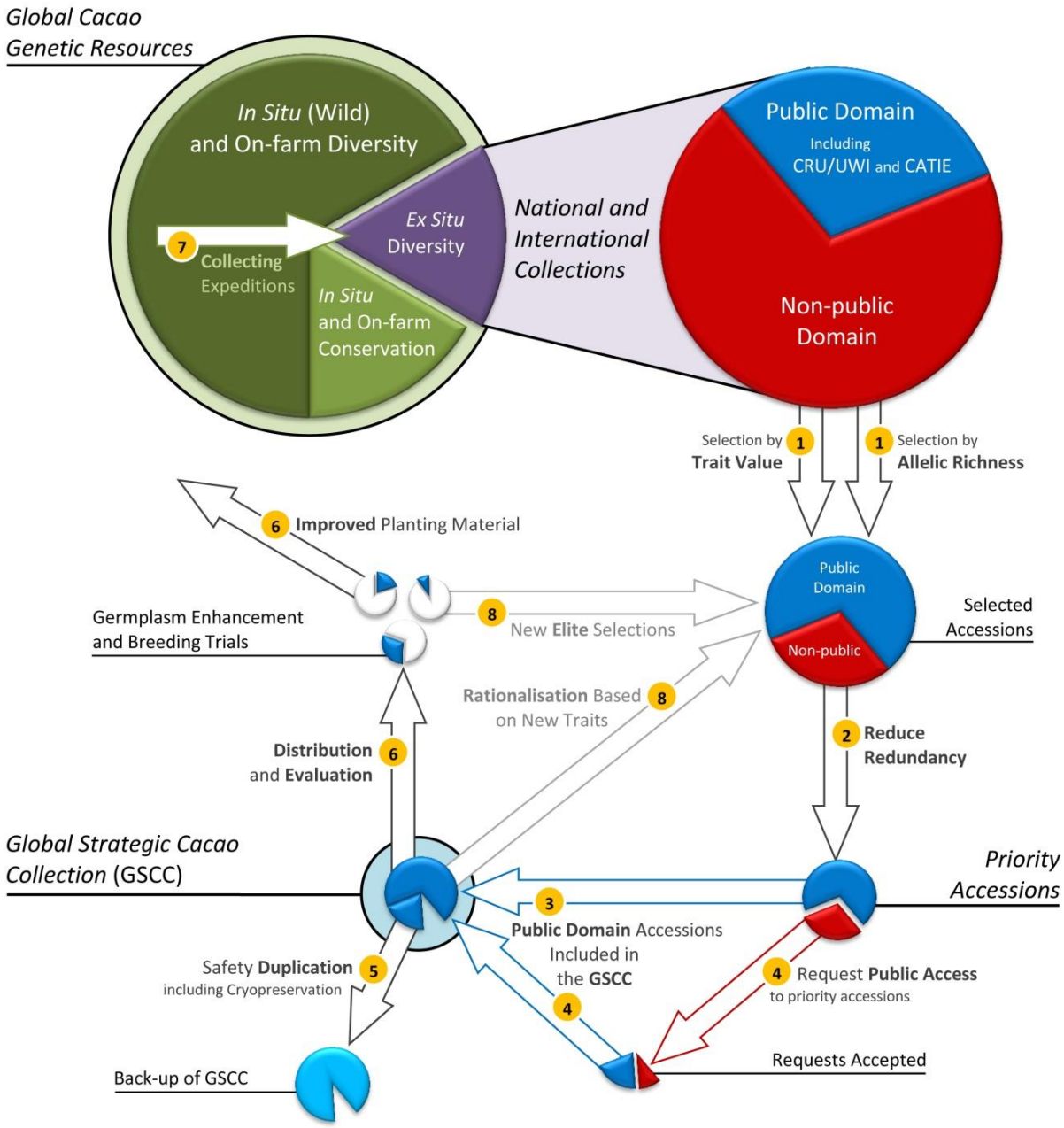
CacaoNet will also ensure the continuing development of the GSCC in consultation with all its members. Partners will agree on how to share responsibilities for conserving and distributing material from the GSCC. The management responsibility of the identified accessions would reside with the various genebank curators. Long-term funding will be discussed with the Global Crop Diversity Trust, other international donors and with the private sector. CacaoNet expects to continue to facilitate the dialogue between the ITPGRFA and the countries that are maintaining cacao materials targeted by the GSCC, in order to encourage countries to follow the example of CATIE and CRC/UWI and place selected accessions under the Treaty.

The process of developing the GSCC is represented in Figure 6 (see next page).

9. Genetic diversity in combination with measures of agronomic value will be used to identify accessions of interest.
10. A second round of selection aimed at reducing redundancy will generate the list of Priority Accessions.
11. Priority Accessions in the public domain will become part of the GSCC.
12. Public access will be requested for any Priority Accession not already in the public domain so that it can be included in the GSCC.
13. Each GSCC accession will be duplicated in another field collection for safety, and some may also be backed-up through cryopreservation (International quarantine required).
14. Material in the GSCC and all its associated information will be freely available for use in germplasm enhancement and breeding programmes, resulting in improved planting material becoming available to farmers (International and/or regional quarantine required).

15. Future collecting expeditions will target gaps in the GSCC (International and/or regional quarantine required).
16. Rationalization of the GSCC will continue as new material becomes available from collecting expeditions and breeding programmes.

Figure 6. Process for the development of the Global Strategic Cacao Collection (GSCC)



Annex 6. Description of the agreed methodology to select accessions based on allelic diversity

Two traditional core selection methods that use grouping and selection of an optimum set of accessions were employed, followed by a further iteration designed to reduce the redundancy of the core selection (*van Raamsdonk & Wijnker, 2000; van Treuren et al., 2008; van Treuren et al., 2009*). The method is illustrated by way of reference to a large public dataset of simple sequence repeat (SSR) information (microsatellite markers). However, the method is not limited to the analysis of molecular genetics data but can combine both discrete categorical and continuous variable data classes. Importantly, initial population differentiation and selection of representative subsets from a population need not make use of only a single data type across the whole cacao resource; a consistent data type is only required for the final selection of the non-redundant core collection. However, for clarity and due to the incomplete nature and inconsistencies in data entry format for morphological, physiological, and agronomically valued information in this example only molecular data has been used. The primary dataset used as an example to demonstrate the utility of this model made use of a public dataset first published by Motamayor et al. in 2008 (http://www.ars.usda.gov/Research/docs.htm?docid=16432). The method follows an iterative approach employing a three step procedure; first the genetic resource is stratified into distinct groups; second, an optimum set is selected from within each group; third, the combined selections from each group are combined and subject to a further round of selection to reduce the redundancy within the sum of the selections from the representative groups.

Population definition

As a stratification method, the software tool STRUCTURE 2.3 (Pritchard) was used to differentiate accessions into population groups. We employed 200,000 burn-in iterations and 200,000 iterations under an admixture model. Similar run parameters have been used elsewhere for the analysis of cacao population structure (*Motamayor et al., 2008*).

Minimum core size and optimized composition selected for maximum allelic richness

A method of maximizing allelic richness is employed to select a subset of accessions of minimal size by replacement based on the number of represented classes of marker variables for the number of accessions within the sample (*Gouesnard et al. 2001*). We employed the Shannon index as a measure of allelic diversity:

$$IShannon,j = - \Sigma(pij \ln pij)$$

where p_{ij} represents the i^{th} class frequency of the j^{th} variable.

This sampling strategy, based on allele frequency, favours core collections with fairly distributed allelic classes, rather than a biased selection of rare alleles. This method has previously been shown to provide the optimum solution for selection of core collections utilizing SSR data *(Escribano et al., 2008).* For comparison we plotted sample scores for an equal number of randomly composed core sets of equal size. We employed 20 replicate runs and 10,000 iterations within each replicate for each population group.

Once the minimal sample size required to contain maximum allelic richness has been determined for each population group, core selections of this size are constructed by replacement to identify the specific accessions contained within the optimum set. We employed 100 replicate runs and 1000 iterations within each replicate ensure adequate sampling given the size of each group and the magnitude of the selection to be made from it in each case.

Annex 7. Membership of accessions for the GSCC based on allelic diversity

The list below includes a total of 261 accessions.

NOTE: This draft list is based exclusively on DNA derived genetic data and makes no use of agronomical or morphological trait information. Population groupings employ the same geographically derived naming system employed by Motamayor et al. 2008.

Accession	Population
LCTEEN 302	Amelonado
MA 11	Amelonado
LCTEEN 26	Amelonado
MA 14 PL9	Amelonado
CAB 0733	Amelonado
SPEC 41/6 18	Amelonado
CAB 36	Amelonado
BE 8	Amelonado
MA 12	Amelonado
YAL 5A	Amelonado
CJ 5	Amelonado
BE 2	Amelonado
U 59	Contamana
CAB 183	Contamana
U 66	Contamana
U 49	Contamana
CAB 190	Contamana
U 28	Contamana
U 38	Contamana
U 15	Contamana
U 37	Contamana
U 31	Contamana
U 56	Contamana
U 57	Contamana
TAP 3	Contamana
U 39	Contamana
CAB 185	Contamana
U 68	Contamana
CAB 186	Contamana
U 36	Contamana
SCA 5	Contamana
U 5	Contamana
U 2	Contamana
UCA 1	Contamana
CAB 188	Contamana
U 58	Contamana
CAB 184	Contamana
U 4	Contamana
U 70	Contamana
CRIOLLO 13	Criollo
PER 2	Criollo
CHA 13	Criollo

Accession	Population
TC 3	Criollo
TC 1	Criollo
TC 9	Criollo
B 48	Criollo
CHA 20	Criollo
CHA 18	Criollo
CA S5	Criollo
SJU 1	Criollo
STA MARIA 2	Criollo
LIB 2	Criollo
LCTEEN 134	Curaray
LCTEEN 334	Curaray
LCTEEN 390	Curaray
LCTEEN 121	Curaray
LCTEEN 123	Curaray
LCTEEN 386	Curaray
LCTEEN 434	Curaray
LCTEEN 329	Curaray
LCTEEN 94	Curaray
LCTEEN 403	Curaray
CURIS	Curaray
LCTEEN 261/S 4	Curaray
LCTEEN 281	Curaray
LCTEEN 87	Curaray
LCTEEN 432	Curaray
LCTEEN 389	Curaray
LCTEEN 188	Curaray
LCTEEN 234	Curaray
LCTEEN 257	Curaray
LCTEEN 189	Curaray
LCTEEN 219	Curaray
LCTEEN 193	Curaray
LCTEEN 80	Curaray
LCTEEN 122	Curaray
LCTEEN 180	Curaray
NAP 25	Curaray
LCTEEN 195	Curaray
LCTEEN 325	Curaray
LCTEEN 255	Curaray
LCTEEN 57	Curaray
LCTEEN 152	Curaray
LCTEEN 421	Curaray

Accession	Population
LCTEEN 333	Curaray
NAP 3	Curaray
LCTEEN 227	Curaray
LCTEEN 60	Curaray
KER 1 L	Guiana
CJ 4	Guiana
CJ 2	Guiana
GU 156B	Guiana
KER 3	Guiana
B7 B3	Guiana
ELP 20 A	Guiana
ELP 32 A	Guiana
KER 11 1 L	Guiana
GU 134B	Guiana
CAB 0517	Iquitos
U 10	Iquitos
SPEC 54/1	Iquitos
CAB 0330	Iquitos
CAB 0367	Iquitos
CAB 0531	Iquitos
AMAZ 10	Iquitos
NA 68	Iquitos
AMAZ15/15[CHA]	Iquitos
CAB 0516	Iquitos
COCA3370/5[CHA]	Iquitos
AMAZ 2	Iquitos
NA 268	Iquitos
CAB 0527	Iquitos
CAB 0328	Iquitos
C.Sul 1	Iquitos
AMAZ 13	Iquitos
AMAZ 5/2 [CHA]	Iquitos
AMAZ 15 [CHA]	Iquitos
NA 409	Iquitos
CAB 0331	Iquitos
CAB 0324	Iquitos
PA 98	Marañón
PA 175	Marañón
CAB 17	Marañón
CAB 19	Marañón
PA 52	Marañón
CAB 0224	Marañón

Accession	Population
CAB 0776	Marañón
PA 135	Marañón
PA 18	Marañón
PA 202	Marañón
CAB 23	Marañón
PA 294	Marañón
CAB 0219	Marañón
PA 139	Marañón
PA 1	Marañón
CAB 0251	Marañón
CAB 0777	Marañón
PA 187	Marañón
CAB 0440	Marañón
PA 4	Marañón
PA 179	Marañón
PA 82	Marañón
CAB 0764	Marañón
CAB 0466	Marañón
PA 88	Marañón
CAB 0422	Marañón
CAB 0458	Marañón
CAB 0459	Marañón
CAB 21	Marañón
PA 30	Marañón
PA 188	Marañón
CAB 0452	Marañón
PA 165	Marañón
CAB 0783	Marañón
PA 310	Marañón
CAB 0749	Marañón
LCTEEN 73	Nacional
LCTEEN 91	Nacional
MO 90	Nacional
LCTEEN 312	Nacional
LasBrisas 13 13	Nacional
MO 122	Nacional
TAP 8	Nacional
NA 712	Nacional
TAP 5	Nacional
MO 84	Nacional
MO 125	Nacional
UCA 3	Nacional
MO 20	Nacional
U 22	Nacional
NA 672	Nanay
NA 406	Nanay
NA 768	Nanay
NA 326	Nanay
NA 227	Nanay

Accession	Population
NA 232	Nanay
U 9	Nanay
NA 206	Nanay
POUND 10/B	Nanay
NA 79	Nanay
NA 702	Nanay
NA 435	Nanay
NA 753	Nanay
NA 283	Nanay
NA 841	Nanay
NA 92	Nanay
NA 279	Nanay
EBC 114	Purús
CAB 67	Purús
CAB 0514	Purús
CAB 0484	Purús
C.Sul 5	Purús
CAB 198	Purús
CAB 0495	Purús
CAB 148	Purús
EBC 142	Purús
CAB 70	Purús
CAB 77 PL5	Purús
RB 40	Purús
LCTEEN 369	Purús
CAB 0334	Purús
CAB 128	Purús
C.Sul 9	Purús
CAB 130	Purús
CAB 195	Purús
CAB 151	Purús
CAB 0369	Purús
CAB 197	Purús
CAB 200	Purús
CAB 0368	Purús
EBC 138	Purús
CAB 154	Purús
LCTEEN 368	Purús
EBC 121	Purús
CAB 0344	Purús
EBC 136	Purús
CAB 193	Purús
CAB 194	Purús
CAB 0341	Purús
LCTEEN 371	Purús
CAB 150	Purús
CAB 0475	Purús
LCTEEN 406	Purús
CAB 0357	Purús

Accession	Population
LCTEEN 412	Purús
LCTEEN 415	Purús
LCTEEN 409	Purús
CAB 152	Purús
CAB 0342	Purús
CAB 0211	Purús
CAB 181	Purús
CAB 0236	Purús
CAB 0213	Purús
SIC 961	Amelonado
VILLANO 2 [CHA]	Curaray
NA 249	Nanay
FSC 7	Amelondo
TAP 2	Nacional
SIC 801	Amelonado
BOB 8 [CHA]	Nacional
TAP 1	Nacional
NA 950	Nanay
AGU 8	Curaray
IMC 27	Iquitos
NA 254	Nanay
PA 289	Maranon
NA 3	Nanay
NA 12	Nanay
WILD	#N/A
NA 337	Nanay
ICS 100	Trinitario
ICS 80	Trinitario
ICS 46	Trinitario
ICS 35	Trinitario
ICS 86	Trinitario
ICS 10	Trinitario
ICS 14	Trinitario
ICS 95	Trinitario
ICS 40	Trinitario
ICS 65	Trinitario
ICS 71	Trinitario

ANNEX A2 - GLOBAL STRATEGY SECTION 4.2 DEVELOPING THE GLOBAL STRATEGIC CACAO COLLECTION (GSCC) - WHERE WE WANT TO GO

CacaoNet will coordinate the development and establishment of a Global Strategic Cacao Collection (GSCC) as a virtual collection consisting of the most unique and valuable materials according to agreed practices and standards and readily available to any *bona fide* user. The GSCC will be based on materials from the two international collections at CATIE and CRC/UWI which have placed their cacao germplasm under the auspices of the Governing Body of the ITPGRFA, the national collections that have proven records of making materials available and in public domain, such as the collections at CIRAD and USDA and from any collections willing to take the necessary steps to make their materials available particularly for use on breeding programmes. The GSCC will also rely on the critical role of the ICQC,R for the international safe movement of cacao genetic resources throughout the world.

The objective of the GSCC is therefore to ensure the cost-effective and efficient long-term *ex situ* conservation of the cacao genepool and its accessibility to all current and future users. Agreed criteria of genetic diversity richness and uniqueness, and measures of agronomic value to be used to identify priority accessions are described in *Section 3.2*. The process of developing the GSCC is represented in *Figure 6*.

The GSCC materials should be prioritized for distribution. Collection managers and breeders using the material would participate in comprehensive characterization, evaluation and further researching of the GSCC collection. All related information will be made available through GSCC information portal. The feasibility of using *in vitro* culture and cryopreservation will be considered for cost-effective duplication of the GSCC.

The consultation process for the development of the GSCC would be led by CacaoNet with all its members and with INGENIC and would take place over a number of consultation meetings. Long-term funding will be discussed with the Global Crop Diversity Trust, other international donors and with the private sector (*see Section 4.1*).

The following specific actions are proposed:

Short-term actions – in the next three years

- Agreeing on the criteria for the selection of materials (for both allelic diversity and traits of interest for breeding).
- Assessing the cacao genetic diversity currently conserved in *ex situ* collections.
- Identifying those publically maintained unique accessions that are available for use by breeders and researchers in the two international collections at CRC/UWI and CATIE and in national collections.
- Developing a proposal for reducing duplication of genetically similar clones, using genetic diversity assessment tools, with a focus on the collections at CRC/UWI and CATIE.
- Developing a process for resolving mislabelling problems in the international and national collections.
- Agreeing on the sharing of responsibilities for conserving and distributing the GSCC materials between the cacao collections in the public domain (roles and responsibilities of the network of partners).
- Identifying urgent conservation support needed for the material identified for the GSCC.
- Characterizing public domain germplasm prioritized to allow assessment and recommendations for inclusion in the GSCC.

- Agreeing on field evaluation at multiple sites under controlled and recorded conditions for the proposed GSCC accessions (see Section 4.4 below).
- Conducting a feasibility study on *in vitro* methods to facilitate distribution through quarantine facilities, including recommendations on type of materials (budwood or plantlets), impact and costing.
- Agreeing on the safety-duplication of the GSCC in field genebanks and/or via cryopreservation (roles of implementing partners and network of collections).
- Agreeing on the development of CANGIS (see details in Section 3.7).
- Conducting a detailed costing study of the GSCC with conservation costs and associated services such as germplasm evaluation, quarantine, virus-indexing, distribution and documentation.
- Promoting/holding continued discussions with the Global Crop Diversity Trust and with the private sector for possibilities of long-term funding support to the GSCC.
- Promoting/holding continued discussions with the Secretariat of the International Treaty and countries maintaining cacao materials targeted by the GSCC, to promote the designation of this germplasm under the Treaty following the example of CATIE and CRC/UWI.
- Agreeing on best practices for cacao collection management and develop standards.
- Promoting the implementation of genebank management standards and dissemination of germplasm and related information.

Long-term actions - beyond three years

- Continuing coordination of the GSCC including support for documentation.
- Continuing effort to characterise and evaluate germplasm in international (priority) and national genebanks towards the establishment of GSCC.
- Identifying duplicates based on characterisation of priority materials.
- Rationalizing international and national collections within the framework of the GSCC.
- Identifying a priority set of accessions for *in vitro* culture to facilitate distribution through quarantine facilities, if the feasibility study validates the use of this technology.
- Ensuring the safety-duplication of the GSCC using appropriate methodology.

In order to provide exact costing figures, a detailed and comparable costing study of CATIE and CRC/UWI cacao genebank operations and an analysis of duplication should be carried out, using the tool developed by the CGIAR, as well as an assessment of the diversity currently conserved in all collections. Therefore, the budget required for the development of the GSCC is calculated here based on estimated costs, estimated number of accessions and activities to be carried out by a network of partners.

The GSCC will be composed of accessions representing the cacao genepool based on allelic richness and on key traits of interests to breeders (*criteria described in Section 3.*2). The estimated number of accessions is mainly based on the CATIE collection (1,146 accessions) and CRC/UWI collection (2,400 accessions), and additional unique materials in other currently publicly available collections such as USDA (estimated 200 accessions) and CIRAD (French Guyana and Montpellier) (646 accessions), current total in the public domain is estimated at about 4,400 accessions. If we estimate that a 10% level of duplication may exist within each of the collections (440 accessions) and 30% level of duplication between these accessions (about 1,300 accessions), it is estimated that the GSCC would initially comprise of between 2,500 to 3,000 accessions (about 12% of the current global holdings 24,000 accessions) and would gradually be reduced as research on germplasm identity progresses and priorities are further defined. It is therefore proposed that cost estimates for the development and management of the GSCC be based on an estimated 2,500 accessions.

The estimates used for costing the GSCC are based on the collection management activities defined in Annex 10 (*Definition of cacao collection management activities following the model of the CGIAR Decision-Support Tool, Table 4 of the CGIAR costing study*) and the specific costs detailed in the 2010 CGIAR costing study for the vegetatively propagated crops (adjusted for inflation at a rate of 5% per year). The calculations are explained in the footnotes for each cost estimate in Annex 11 (*Summary of the costs of the GSCC with footnotes detailing the data source).*

It should be noted that the techniques used to introduce cacao accessions into *in vitro* culture as somatic embryos for the purposes of multiplication, distribution and cryopreservation are rather different to those already widely used for other vegetatively propagated crops. Research is ongoing into improving the applicability of somatic embryogenesis techniques for a wider range of cacao genotypes and it is anticipated that the review of multiplication of planting materials currently being undertaken by Bioversity will provide updated information on the prospects and costs of such techniques. Meanwhile the costs of preparing somatic embryos of an estimated 75 accessions per year and then subsequently introduced and maintained (target 50 accessions) in cryopreservation is estimated at 1,000 USD and 500 USD respectively per accession (estimated based on the experience at Reading University). The cost of maintenance in cryopreservation in a flask (minimum containing 500 accessions) is estimated at 2,000 USD per year.

Table 7. Summary of the annual costs of the GSCC – including capital costs (USD).

Operation (as described in Annex 10)	**No of accessions / year**	**USD / year**
Acquisition	20	5,620
Field maintenance	2,500	102,678
Characterization - morphological	200	5 000
Characterization - molecular	200	22,000
Identification of duplicates and integrity	200	41,200
Regeneration	200	22,600
Health testing	200	67,600
ANNUAL COSTS - based on 2009 estimates	TOTAL	266,698
ANNUAL COSTS 2012 adjustment for inflation at 5% per year	**TOTAL**	**308,736**
Introduction/ multiplication of accession *in vitro*	75	75,000
Cryopreservation (introduction)	50	25,000
Cryopreservation (maintenance)	Up to 500	2,000
GRAND TOTAL		**410,736**

Notes: It is anticipated that the annual cost would be reduced over time as the size and composition of the GSCC is revised following the diversity analysis study. On-going efforts to conserve much of the material which will initially form the GSCC are currently supported by public and private sector funding for the collections at CRC/UWI and CATIE, representing an estimated 80% of the GSCC, though this is only assured on a short-term basis.

The budget for coordination of and consultation for the development of the GSCC is included in *Section 4.8.* Strengthening the networking and partnerships for global collaboration, in the budget allocated for meetings of the CacaoNet Working Groups and stakeholders consultations (150,000 USD/year). Support for national partners is also included in *Section 4.8*.

ANNEX A3 - GLOBAL STRATEGY SECTION 3.7 IMPROVING DOCUMENTATION AND SHARING OF INFORMATION – WHERE WE ARE NOW

A simple yet robust information management system, that combines comprehensive and accurate information on the origins, conservation locations, availability and characteristics of individual accessions, will be the portal to accessing all relevant information and be a key component in the establishment, management and use of the Global Strategic Cacao Collection (GSCC).

As part of the GSCC information portal, a central database, CANGIS (CacaoNet Germplasm Information System), will bring together all the genebanks and other service providers that collectively form the GSCC and facilitate their effective management. CANGIS will be a relatively small online database that will maintain specific, high quality data (including passport descriptors and the characters supporting an accession's inclusion in the GSCC) on all the individual accessions that make up the GSCC, and provide a means for users to access this germplasm. This information will be based on individual trees located at specific sites. CANGIS will link to existing international databases, such as ICGD and TropGENE (utilizing standardized variety identification codes), in order to access additional information that is of interest to potential users of the germplasm. This will take the form of either a direct link (taking the user to the other database) or a web service (where information is retrieved from another database, but integrated into the host's output). Users of these international databases will also be able to link back to CANGIS in order to access information on individual accessions in the GSCC and their availability.

In order to ensure that a minimum standard of record keeping is maintained for the entire 'virtual' GSCC, local germplasm management systems need to be in place at each of the genebanks holding accessions that are part of the GSCC. Some genebanks already have such a system established, or are under development, whilst others will need to install one. These local germplasm management systems must facilitate a good flow of information back to the GSCC information portal to allow effective management of the GSCC as a whole. However, they must also work at the level of the individual genebank, which may hold collections of other crops and be spread over several locations, and not require a significant amount of additional time or computer skills to use. In order to link CANGIS to the accessions maintained at each collaborating genebank, a minimum standard of record keeping is required at the local genebank level. The local genebank documentation system must be able to link passport data, plus any additional characterization or evaluation information, to specific trees in the field and then make this information more widely available.

A prototype version of CANGIS has been developed using information from ICGD on International Clone Trial (ICT) accessions (Eskes and Efron, 2006) held in the international collections at CRC/UWI and CATIE. CANGIS already includes a link to ICGD, taking the user directly to a page of evaluation data for a specific genotype, and more links are being developed.

Movement of material into and out of the 'virtual' GSCC should be monitored, including transfer for safety duplication. This would provide up-to-date information on the management of accessions within the GSCC and how they are being used. The monitoring system would link to the network of local genebank management systems, providing up-to-date information on the location and availability (e.g. quarantine status) of each of the widely distributed accessions in the GSCC. See Figure 7.

A likely future development for the GSCC information portal is the provision of a germplasm ordering system that would allow the user to select the most appropriate germplasm accessions in the collections accessible in the public domain based on passport, characterization and evaluation data. The germplasm ordering system would take account of the guidelines for safe movement of cacao germplasm and link

accessions to a MTA. The ordering system would also serve the purpose of tracking movement of germplasm in a similar way that the global information system for the International Treaty does.

Technologies such as GPS and barcoding are not widely used in cacao collections at the current time, but they are becoming less expensive and more widely available, and likely to be increasingly important in managing genetic resources in the future. In some countries like Brazil, this process has already started. Under a new law, all germplasm accessions of all species will have to be geo-referenced (GPS positioned). The use of GPS equipment during collecting missions should also be encouraged.

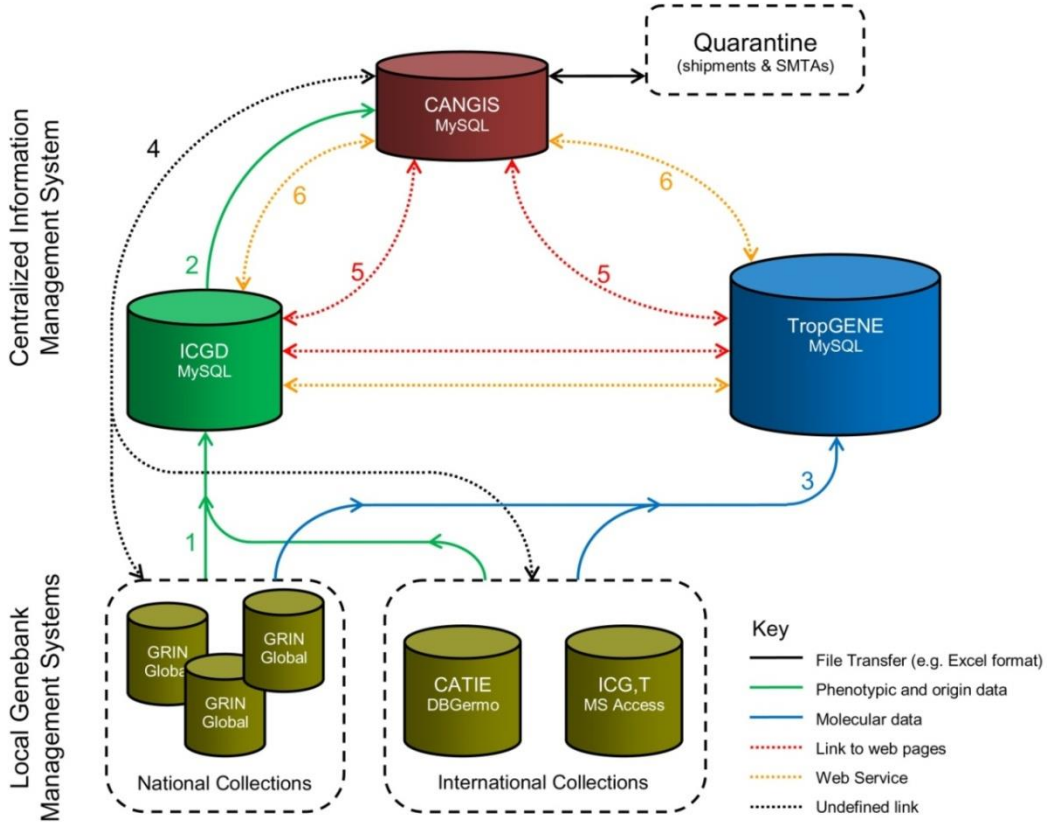


Figure 7. Components of the GSCC information portal (C. Turnbull, Reading University).

1 Characterization and evaluation data are sent to ICGD (includes non-CacaoNet accessions and information).

2 Once checked and standardized, information on the Global Strategic Cacao Collection accessions is entered into CANGIS.

3 Molecular data are sent to TropGENE (includes non-CacaoNet accessions and information).

4 A degree of direct networking between Global Strategic Cacao Collection IMS and the local genebank management systems is required for monitoring/tracking accessions in the base and active collections. The form this will take will largely depend on the genebank management systems that are adopted (e.g. GRIN-Global).

5 In order to access additional information available from one of the other databases, the user can be linked directly to the relevant page on the collaborating website (all of the databases use the same variety identification codes).

6 Web services allow an information management system to query distributed databases and integrate the results with its own output, removing the need to physically transfer the user to the other database.

The successful integration of a network of local genebank management systems into the GSCC information portal is particularly important due to the widely dispersed nature of the accessions that will make up the "virtual" GSCC. The international collections at CATIE and CRC/UWI already utilize computerized genebank management systems, but many other collections do not yet have such well-developed information management systems in place. As most collections are already aware (CacaoNet surveys 2008-2012), a robust and easy to use local management system would also be of direct benefit to local staff.

Collection curators may be encouraged to adopt the same standardized system, such as GRIN-Global. GRIN-Global is a project whose mission is to create a new, scalable version of the USDA's Germplasm Resources Information Network system (GRIN) suitable for use by any interested genebank in the world. It is being developed in a joint effort with the Global Crop Diversity Trust, Bioversity International, and ARS/USDA. The project's goal is to provide the world's crop genebanks with a powerful, flexible, easy-to-use global plant genetic resource (PGR) information management system. The database and interface(s) will be designed to accommodate both commercial and open-source programming tools, to be database-flexible, and to require no licensing fees for genebank use. However GRIN-GLOBAL may need to be customised for managing cacao germplasm.

During the transition phase of adopting GRIN-Global, Excel templates will be developed to help curators provide standardized information whenever possible, though ICGD would continue to accept data in any format. In addition, since the system records the donor of each accession, it would help identify, trace and restrict the impact of mislabelling events. For this reason, each accession's verification-status would also be indicated.

The main areas of capacity building are associated with the local information management system. If a collection chooses to use GRIN-Global, training and support will be required for staff who will maintain the system. The initial set up of the system will likely require additional training and may best be carried out at a regional or international level, with experts on GRIN-Global carrying out this initial set-up in collaboration with local staff.

Any local information management system will require a minimum level of hardware (computers and networking) and general skills in information technology. There may also be a need for additional training on data collecting and input, particularly where standard descriptors are used.

The CacaoNet website (www.cacaonet.org) will provide a single point of access for all the elements of the GSCC information portal, providing also a basic overview of cacao (including the centre of diversity, the main production areas, spread of pests and diseases, etc.) and explains the importance of cacao globally, highlighting public awareness issues. Information on CacaoNet would also be available, targeted to different user groups (including potential donors, existing partners and the general public).

ANNEX A4 - GLOBAL STRATEGY SECTION 4.7 IMPROVING THE DOCUMENTATION AND SHARING OF INFORMATION ON GERMPLASM – WHERE WE WANT TO GO

The objective is to develop a portal for accessing all relevant information and be a key component in the establishment, management and use of the GSCC. This GSCC information portal will document all data related to the conservation, evaluation and management of the germplasm and would be made publically available at all times. This includes building on its central database CANGIS, linking to the ICGD, TropGENE and all the genebanks and service providers that collectively form the GSCC, with newly developed GRIN-Global. Users will be able to access information on individual trees located at specific sites (GSCC accessions) and their availability. This will include some capacity building of key collections to develop or adopt new local genebank management systems (such as GRIN-Global) to ensure a minimum standard of record keeping. This may involve a minimum level of hardware (computers and networking) and general skills in information technology. Information would be recorded on movement of GSCC accession from one location to another including the quarantines (See Figure 7). The GSCC information portal would also provide the users with the possibility to request material on-line through a germplasm ordering system with MTAs. The CacaoNet website will provide a single point for accessing information on GSCC, and provide a basic overview of cacao and its importance globally, highlighting public awareness issues.

The following specific actions are proposed:

Short-term actions – in the next three years

- Coordinating the compilation of characterization and evaluation data from all collections (data to be supported by the molecular verification of genotypes where possible) to facilitate the identification of the GSCC including breeding and evaluation data.
- Developing the GSCC information portal, including CANGIS in particular, by requesting information from the international collections held by CRC/UWI and CATIE (e.g. accession and tree numbers and passport data not already maintained by existing databases), including information on the collection at ICQC,R, adding links and web services to other online databases and contact details to request material.
- Introducing tree identifiers and accession numbers to ICGD, allowing specific evaluation data in the ICGD to be linked to CANGIS.
- Stimulating the rescue of historical data collected in genebanks and eventually trials which can provide information useful to breeders.
- Developing automated system for monitoring and updating the GSCC information portal, with particular emphasis on linking local systems to CANGIS.
- Developing a germplasm ordering and tracking systems.
- Making CANGIS available to cacao community (online) and request feedback.
- Assessing the suitability of adopting GRIN-Global at collections that do not have a local information management system already by assessing minimum level of local expertise and IT equipment needed and the training requirements for initial set up (customization).

Long-term actions - beyond three years

- Ensuring appropriate level of record keeping in collections (working at tree level) and potential uptake of GRIN-Global.

The development of the GSCC information portal will initially focus on compiling characterisation and evaluation information on *ex situ* cacao germplasm for the identification of priority materials for the

establishment of the GSCC. The identification of the unique materials would be done through an analysis of duplicates based on characterisation data. Support would be provided to cacao collection institutes (genebanks) for linking information to the GSCC information portal at an estimated 15,000 USD per genebank for at least 10 collections. The cost estimate is based on the initial cost of development of CANGIS, the support to cacao collections for linking to the GSCC information portal (including CANGIS) and the annual management of the system. The development and maintenance of the GSCC information portal includes support towards the ICGD, TropGene, GRIN-Global and local germplasm management systems in some of the collections with GSCC accessions. An estimated 40% of these costs are currently covered by NYSE Liffe/CRA Ltd who provides support for the ICGD project. These costs may decrease with time as the information systems are established and information management capacity is built within national genebanks.

ANNEX B. WORKSHOP PROGRAMME

DAY 1	Wednesday 22 October
09:00-10:00	**WELCOME AND INTRODUCTION TO THE WORKSHOP** • **Welcome address** from hosts Cocoa Research Centre of the University of the West Indies (CRC), Bioversity International and CacaoNet – **Path Umaharan**, Director, Cocoa Research Centre – *10 minutes – Path Umaharan and Nadine Mootoo* ○ **Background and Objectives of the Workshop- Brigitte Laliberté**, CacaoNet/Bioversity International - *Facilitator – 10 minutes* ○ **Introductions of participants** – *15 minutes* ○ **Agreement on the proposed programme**– *15 minutes* ○ **Logistic information - *Marissa Moses***- *5 minutes*
10:00-10:30	***Coffee/tea break***
10:30-11:30	**SESSION 1: REVIEW CURRENT SITUATION AND CONCEPT** ***Objective 1:*** Review the current situation and concept: where we are with the development of the Global Strategic Cacao Collection (GSCC), its goal and proposed approach (allelic diversity and traits) and the information systems supporting the decision-making process – CANGIS and the ICGD. ○ **Where we are with the Global Strategic Cacao Collection (GSCC)** – **Brigitte Laliberté** – *30 minutes* ○ **GENERAL DISCUSSION** – *30 minutes*
11:30-12:30	**SESSION 2: GSCC APPROACH AND FUTURE DIRECTION** ***Objective 2:*** Agree on the overall approach and goal of the GSCC and future direction to secure and optimise the use of cacao genetic diversity. **Background** • The CGIAR: Example of global collections for long-term conservation and funding via the Global Crop Diversity Trust – **Brigitte Laliberté** - *10 minutes* • Decision-making processes for efficient genebank conservation and evaluation: The UK National Fruit Collection as an example – **Paul Hadley** – *10 minutes* • **GENERAL DISCUSSION** - *10 minutes*
12:30-14:00	***Lunch break – Ortinola restaurant – 90 minutes***
14:00-15:30	**SESSION 2: GSCC APPROACH AND FUTURE DIRECTION - *continued*** • Morphological characterisation and evaluation of traits of economic interest - **Frances Bekele** – *5 minutes* • Evaluation of disease resistance traits with examples from CATIE **– Wilbert Phillips** – *5 minutes* • Evaluation of Flavour and other quality traits **- Darin Sukha –** *5 minutes* • Yield: Yield components and effect of abiotic stress **– Paul Hadley –** *5 minutes* • **GENERAL DISCUSSION** - *10 minutes*
15:30-16:00	***Coffee/tea break***
16:00-18:00	**SESSION 3: INFORMATION ON EVALUATION FOR DECISION-MAKING** ***Objective 3:*** Propose ways to improve the availability of information that will contribute to the decision-making process for the GSCC, particularly for evaluation of traits of interest for users. • Presentation on CANGIS and the ICGD as the information systems to document the GSCC – **Chris Turnbull** – *10 minutes* • Presentation on GRIN-GLOBAL **– Pete Cyr** - *10 minutes* • Introduction to the GCP Integrated Breeding Platform (IBP) crop ontologies, Trait Dictionaries and objectives/uses of Breeding Management System (BMS) – **Elizabeth Arnaud** – *10 minutes* • AgroGen – breeding management system from Brazil – **Uilson Lopes** – *10 minutes* • **GENERAL DISCUSSION** - *30 minutes*

DAY 2	Thursday 23 October
09:00-10:00	**<u>SESSION 2: GSCC APPROACH AND FUTURE DIRECTION – *continued*</u>** **<u>What is needed from the different groups</u>** *(large, medium and small chocolate makers, breeding groups and producing countries) and what are the priorities and interest for a GSCC* • **From CocoaAction: cooperation between the cocoa and chocolate industry and the Ivorian and Ghanaian governments for productivity enhancements and community development interventions** *- example of developing CSSV resistant-materials and how the GSCC can contribute -* **Martin Gilmour** *– 10 minutes including WHERE WE ARE ON CSSV* • **From the Fine Chocolate Industry Association (FCIA) and Heirloom Cacao Preservation Initiative (HCP) - Ed Seguine** – *10 minutes including WHERE WE ARE ON METHODOLOGY* • **From the Regional Cocoa Breeders Groups** – *20 minutes* o West Africa o Asia/Pacific o Latin America and Caribbean • **From cocoa producing countries** – *20 minutes* o Brazil o Ecuador o Costa Rica o Cote d'Ivoire o Peru • **GENERAL DISCUSSION** *- 20 minutes*
10:00-10:30	***Coffee/tea break***
10:30-11:30	**<u>SESSION 4: CRITERIA FOR INCLUSION OF GERMPLASM ACCESSIONS IN THE GSCC</u>** ***Objective 4:*** Discuss the criteria to be used to identify priority accessions for the GSCC, mainly: • *A - A first set of accessions selected on the basis of capturing the greatest possible range of* ***allelic richness*** *and diversity.* • *B - A further set of accessions selected on the basis of* ***key traits of interest to users*** *(e.g. yield, flavour, disease resistance etc).*
11:30-12:30	**<u>SESSION 5: REVIEW OF METHODOLOGIES, SCIENTIFIC/TECHNICAL ADVANCES FOR EVALUATION OF KEY TRAITS</u>** ***Objective 5*:** Review the status of methodologies and scientific and technical advances made since the GSCC concept discussed and proposed in the Global Cacao Strategy in 2012. Particularly regarding the selection of accessions based on key traits of interest to users. **SMALL GROUP DISCUSSION on the following guiding questions –** *30 minutes* **PLENARY DISCUSSION** – *30 minutes* **Conclusions and next steps on the criteria based on key traits of interest to users** – *60 minutes*
12:30-14:00	***Lunch break – Ortinola restaurant – 90 minutes***
14:00-15:30	**<u>SESSION 6: POLITICAL ASPECTS OF ACCESSIBILTY OF GERMPLASM</u>** ***Objective 6:*** Discuss the political aspects of conservation and accessibility of unique materials in the national collections that may not be accessible outside the countries. • **SMALL GROUP DISCUSSSION on possible approaches to facilitate access for use** – *20 minutes* • **PLENARY DISCUSSION** – *40 minutes*
15:30-16:00	***Coffee/tea break***
16:00-17:00	**<u>SESSION 7: PRIORITIES FOR SECURING *EX SITU* CONSERVATION</u>** ***Objective 7:*** Agree on the short, medium and longer-term priorities for securing the *ex situ* conservation and identifying gaps, considering collecting priorities based on threats in the wild and on-farms. **This session could cover the following aspects:** **1.** Securing the long-term funding and the work of the CacaoNet Funding Task force **2.** Update on the GSCC costing study and collection management operations targeting an endowment vs project/research funding – **Daniela Horna** – *10 minutes*

DAY 3	Friday 24 October
07:45-10:45	**07:45 - Visit of the Cocoa Research Centre facilities – *30 minutes*** **08:15 - Bus transportation from CRC to field genebank – *45 minutes*** **09:00 - Visit of the International field collection at the genebank – *60 minutes*** **10:00 - Bus transportation from field genebank to Ortinola – *45 minutes***
10:45-11:00	***Coffee/tea break – 15 minutes***
11:00-11:30	**SUMMARY OF DAY 2, conclusions and discussion on remaining issues**
11:30-12:00	**Safety duplication – principles and update** • Cryopreservation at Nestlé R&D, Tours, France – **Anne Buchwalder** – *10 minutes* • Cryopreservation at the University of Reading, UK– **Paul Hadley** – *10 minutes*
12:00-13:00	**SESSION 8: DECISION-MAKING PROCESS FOR THE GSCC** ***Objective 8:*** Agree on the general decision-making process mechanism for the GSCC, particularly for the process of refining the collection to improve its efficiency and safety duplication. ○ **GSCC in context** – ***Tony Lass*** – *10 minutes*
13:00-14:00	***Lunch break – Ortinola restaurant***
14:00-15:00	**SESSION 9: ROLES AND RESPONSAIBILIES OF PARTNERS** ***Objective 9:*** Propose roles and responsibilities of the main partners in the GSCC, including ensuring the safety-duplication of the materials.
15:00-15:30	**SESSION 10: LINKS BETWEEN THE GSCC AND THE ON-FARM CONSERVATION OF DIVERSITY** ***Objective 10:*** Propose priorities for discussion during the follow-up CacaoNet workshop on the on-farm conservation of cacao genetic diversity (*26-28 October 2014, Guapiles, Costa Rica*).
15:30-16:00	***Coffee/tea break***
16:00-17:00	**SESSION 11: RECOMMENDATIONS FOR NEXT STEPS AND ACTIONS** ***Objective 11:*** Make recommendations for the next steps and proposal for action: • Next steps with the workshop in relations to the objectives of the workshop – documentation and report • Issues to be addressed by research • Action agenda for the partners (CATIE, CRC, ICQCR, and the national partners), data managers • Identification of agents suitable for political persuasion and process • Funding the process and timelines • Coordination and partnerships
17:00-17:30	**WORKSHOP FEEDBACK AND CLOSING** • Workshop evaluation and participants feedback • Closing of CacaoNet workshop on the GSCC

ANNEX C. WORKSHOP PARTICIPANTS

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