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SB-01/02

Genetic Resources Unit

Report on Achievements and Progresses

CIAT
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CIAT GENETIC RESOURCES PRODUCT LINE: Options of agrobiodiversity available for agricultural development worldwide **(2009-2011).**

Targets	Products	Intended user	Outcome	Impact
PRODUCT 1	Genetic Resources Collections of CIAT Germplasm commodities upgraded and maintained up to international standards	All users of the three commodity germplasms worldwide, namely in Central and South America, Africa and South East Asia	Adoption or use designated germplasm in breeding/ agronomy programs	Better varieties, requiring less expensive inputs
Product Targets 2009	• 25% of bean backlog on introduction cleared • 25 % of designated accessions with increased characterization (recovery of 'institutional memory') • 50% of designated accessions documented with digital images • Bar coding implemented for all GRU operations • Protocols for conservation of botanic seeds of cassava and wild *Manihot* species defined • DNA bank: protocols and database established	• NARS and CGIAR commodity breeding and research projects	• Bean breeders can have access to a wider bean diversity • All breeders can use all past evaluation data by CIAT specialists	• Designated germplasm accessed more quickly
Product Targets 2010	• 50% of bean backlog on introduction cleared • 50% of designated accessions with increased characterization (recovery of 'institutional memory') • Protocols for conservation of botanic seeds of cassava and wild *Manihot* species tested • Protocol for cleaned production of *Brachiaria* seed germplasm • DNA bank: 25% of in-trust bean and cassava accessions included	• NARS and CGIAR commodity breeding and research projects	• Bean breeders can have access to a wider bean diversity • All breeders can use all past evaluation data by CIAT specialists • NARS conserving *Manihot* genetic resources have an alternate conservation scheme • Forage agronomists can have access to *Brachiaria* germplasm • Geneticists can use DNA directly for beans and cassava	• Designated germplasm can be used more efficiently by using the appropriate accession(s) • Gene responsible for a particular trait can be sequenced quickly • Gene maps better documented
Product Targets 2011	• 100% of bean backlog on introduction cleared • 75% of designated accessions with increased characterization (recovery of 'institutional memory') • Protocols for conservation of botanic seeds of cassava and wild *Manihot* species implemented • DNA bank: 50% of in-trust bean and cassava accessions included	• NARS and CGIAR commodity breeding and research projects	• Bean breeders can have access to a wider bean diversity • All breeders can use all past evaluation data by CIAT specialists • NARS conserving *Manihot* genetic resources have an alternate conservation scheme • Geneticists can use DNA directly for beans and cassava	• Designated germplasm can be used more efficiently by using the appropriate accession(s) • Gene responsible for a particular trait can be sequenced quickly • Gene maps better documented

PRODUCT 2	The germplasm of *Phaseolus* beans, *Manihot* cassava and selected tropical forages is made available to users, restored to NARS, and safely duplicated	Users, including countries of origin, worldwide can obtain quality germplasm from CIAT GRU; that germplasm is systematically safely duplicated	The "Global System" foreseen by the Trust is getting concrete for beans, cassava, and tropical forages, and the NARS can rely on the CGIAR for a full backup of their national collections.	More benefits in the society (farmers, breeders, agronomists, but also university departments) because of access to genetic resources at anytime; stable and secure access because of the safety duplicates
Product Targets 2009	• On average 4-6,000 samples of the designated collections of beans, cassava and tropical forages are distributed to users annually • 3,000 accessions are replaced at CIP for the safety back-up of the cassava collection in vitro • 3,000 accessions are shipped to CIMMYT for the safety back-up of bean and tropical forages collections • 5,000 accessions are shipped to the Svalbard Global Seed Vault for the safety back-up of bean and tropical forages collections • Advances in detection of diseases of quarantine importance using the Real-Time PCR • Handling of SMTAs implemented electronically	• CGIAR and NARS breeders sensu lato, NGOs, and farmers	• Increased security for the in-trust collections • Shipment of in-trust germplasm and reporting to the Treaty Secretariat made easier • Multilateral System of the International Treaty is implemented and working for the in-trust collections kept by CIAT GRU	• More sustainable food production through either improved varieties or traditional landraces restored to farmers
Product Targets 2010	• On average 4-6,000 samples of the designated collections of beans, cassava and tropical forages are distributed to users annually • 3,000 accessions are replaced at CIP for the safety back-up of the cassava collection in vitro • 3,000 accessions are shipped to CIMMYT for the safety back-up of bean and tropical forages collections • 5,000 accessions are prepared for shipping to the Svalbard Global Seed Vault for the safety back-up of bean and tropical forages collections • Methods for the detection of diseases of quarantine importance using the Real-Time PCR implemented	• CGIAR and NARS breeders sensu lato, NGOs, and farmers	• Increased security for the in-trust collections • Shipment of in-trust germplasm and reporting to the Treaty Secretariat made easier • Multilateral System of the International Treaty is implemented and working for the in-trust collections kept by CIAT GRU	• More sustainable food production through either improved varieties or traditional landraces restored to farmers
Product Targets 2011	• On average 4-6,000 samples of the designated collections of beans, cassava and tropical forages are distributed to users annually • 3,000 accessions are replaced at CIP for the safety back-up of the cassava collection in vitro • 3,000 accessions are shipped to CIMMYT for the safety back-up of bean and tropical forages collections • 5,000 accessions are shipped to the Svalbard Global Seed Vault for the safety back-up of bean and tropical forages collections	• CGIAR and NARS breeders sensu lato, NGOs, and farmers	• Increased security for the in-trust collections • Shipment of in-trust germplasm and reporting to the Treaty Secretariat made easier • Multilateral System of the International Treaty is implemented and working for the in-trust collections kept by CIAT GRU	• More sustainable food production through either improved varieties or traditional landraces restored to farmers

PRODUCT 3	The in-trust collections are genetically and socially relevant: GRU keeps the appropriate genetic diversity so that there is genetic progress in beans, cassava, and selected tropical forages.	Users worldwide can obtain the genetic variation they need in due time, now and in the future	Varieties of beans, cassava, and tropical forages that make a breakthrough in farmers' fields	Higher income for farmers, better nutrition for users, lower costs to the environment
Product Targets 2009	• Secondary gene pools of cultivated *Phaseolus* species better defined. • Monitoring of genetic erosion implemented so that germplasm explorations are carried out in due time and at the right place. • Selected explorations are taking place for *Phaseolus* and cassava germplasm in countries that have ratified the Treaty • Selected sets of germplasm are collected and restored to NARS/ farmers so that they can have access to niche markets (e.g. popping beans)	• NARS, NGOs, farmer groups, breeders in AROs, other genebanks (e.g. USDA, EMBRAPA) are getting access to unexplored sources of variability	• Breeders can include novel traits in their breeding programmes (e.g. white mold resistance in common bean breeding, delayed root deterioration in cassava breeding).	• Increased production and incomes, improved nutritional and technological traits in the crop commodities at lower environmental costs.
Product Targets 2010	• Better appraisal of genetic relationships among *Manihot* species • Monitoring of genetic erosion implemented so that germplasm explorations are carried out in due time and at the right place. • Selected explorations are taking place for *Phaseolus* and cassava germplasm in countries that have ratified the Treaty • Selected sets of germplasm are collected and restored to NARS/ farmers so that they can have access to niche markets (e.g. popping beans, colored cassava)	• NARS, NGOs, farmer groups, breeders in AROs, other genebanks (e.g. USDA, EMBRAPA) are getting access to unexplored sources of variability	• Breeders can include novel traits in their breeding programmes (e.g. tolerance to extreme temperatures in common bean breeding, high or low HCN in cassava breeding).	• Increased production and incomes, improved nutritional and technological traits in the crop commodities at lower environmental costs.
Product Targets 2011	• Selected explorations are taking place for *Phaseolus* and cassava germplasm in countries that have ratified the Treaty, or that allow access and registration into the Multilateral System of the Treaty • Selected sets of germplasm are collected and restored to NARS/ farmers so that they can have access to niche markets (e.g. popping beans, colored cassava)	• NARS, NGOs, farmer groups, breeders in AROs, other genebanks (e.g. USDA, EMBRAPA) are getting access to unexplored sources of variability	• Breeders can include novel traits in their breeding programmes (e.g. tolerance to extreme temperatures in common bean breeding, high or low HCN in cassava breeding).	• Increased production and incomes, improved nutritional and technological traits in the crop commodities at lower environmental costs.

PRODUCT 4	Strengthened institutions that expand the scope of the conservation effort for the agricultural biological heritage	NARS in Latin America and Africa (primary target, although experience has shown that GRU training materials have been used by many other players, *including Spain*)	Improved capacity of NARS to tackle conservation problems of other sets of agrobiodiversity beyond the crop commodities handled by CIAT and the IARCs, namely along the two research drivers of GRU	More conservation for minor cereals, pulses, root crops and tropical fruit species, and hence diversification of the diet and of incomes for farmers worldwide
Product Targets 2009	• Updated handbook of GRU procedures that can serve as a basis for future training (e.g. hands-on training, distance education through e-learning) • Documents with best practices (joint activity involving several IARCs coordinated by SGRP) produced • Distance education course re-run for Latin America (pending on funding availability)	• NARS, NGOs, departments of universities involved in conservation of agrobiodiversity	• Increased capacities of partner institutions to use the updated handbooks of procedures and booklets with best practices	• The "Global System" envisioned by the Trust is progressively taking place for other crops that the ones dealt with by the CGIAR, with conservation and social benefits
Product Targets 2010	• Documents with best practices (joint activity involving *several IARCs coordinated by SGRP*) further diffused • Distance education course run for Africa/ Asia (pending on funding availability) • Participation of GRU Staff in MSc programmes including genetic resources conservation	• NARS, NGOs, departments of universities involved in conservation of agrobiodiversity	• Increased capacities of partner institutions to develop research in conservation for genetic resources not handled by IARCs	• The "Global System" envisioned by the Trust is progressively taking place for other crops that the ones dealt with by the CGIAR, with conservation and social benefits
Product Targets 2011	• Distance education course run for Africa/ Asia (pending on funding availability) • Participation of GRU Staff in MSc programmes including genetic resources conservation	• NARS, NGOs, departments of universities involved in conservation of agrobiodiversity	• Increased capacities of partner institutions to develop research in conservation for genetic resources not handled by IARCs	• The "Global System" envisioned by the Trust is progressively taking place for other crops that the ones dealt with by the CGIAR, with *conservation and social* benefits

PRODUCT 5	Improved link between conservation *ex situ* at CIAT with *in situ* conservation on farm and in the wild, resulting in larger numbers of populations of landraces and wild relatives effectively conserved	National genebanks (e.g. USDA, EMBRAPA), biodiversity institutes; major conservation agencies; national and international herbaria and museums of natural history; NGOs interested in on-farm conservation	Genetic resources of *Phaseolus* beans and *Manihot* cassava are better conserved, and hence used directly or employed in breeding programs	Improved conservation at lower costs for the societies; link between the environmental sector (i.e. protected areas) and the agricultural sector
Product Targets 2009	• Populations of *Phaseolus* beans documented in herbaria (two institutes not yet visited per year) • Populations of *Manihot* wild species documented in herbaria (two institutes not yet visited per year) • GIS mapping for at least one species of bean for all populations ever mentioned in the western hemisphere • Gene flow documented for beans/ or cassava in view of on-farm conservation	• Genebanks of USDA, Mexico, Brazil; CONABIO of Mexico, INBio of Costa Rica, I von Humboldt of Colombia • NGOs interested in on-farm conservation of landraces	• Natural history museums are integrated into genetic resources conservation for beans and cassava wild species • GIS capacity is put at work for the conservation of *Phaseolus* and *Manihot* species	• More populations of more wild species of *Phaseolus* and *Manihot* eventually conserved; thus future sources of variability conserved for progress of plant breeding
Product Targets 2010	• Populations of *Phaseolus* beans documented in herbaria (two institutes not yet visited per year) • Populations of *Manihot* wild species documented in herbaria (two institutes not yet visited per year) • Matching all populations mapped versus those conserved *ex situ* in genebanks worldwide for that bean species; conclusions about conservation priorities • GIS mapping for another species of bean for all populations ever mentioned in the western hemisphere	• Genebanks of USDA, Mexico, Brazil; CONABIO of Mexico, INBio of Costa Rica, I von Humboldt of Colombia • NGOs interested in on-farm conservation of landraces	• Natural history museums are integrated into genetic resources conservation for beans and cassava wild species • GIS capacity is put at work for the conservation of *Phaseolus* and *Manihot* species	• More populations of more wild species of *Phaseolus* and *Manihot* eventually conserved; thus future sources of variability conserved for progress of plant breeding
Product Targets 2011	• Populations of *Phaseolus* beans documented in herbaria (two institutes not yet visited per year) • Matching all populations mapped versus those conserved *ex situ* in genebanks worldwide for that second bean species; conclusions about conservation priorities • Populations of *Manihot* wild species documented in herbaria (two institutes not yet visited per year)	• Genebanks of USDA, Mexico, Brazil; CONABIO of Mexico, INBio of Costa Rica, I von Humboldt of Colombia • NGOs interested in on-farm conservation of landraces	• Natural history museums are integrated into genetic resources conservation for beans and cassava wild species • GIS capacity is put at work for the conservation of *Phaseolus* and *Manihot* species	• More populations of more wild species of *Phaseolus* and *Manihot* eventually conserved; thus future sources of variability conserved for progress of plant breeding

1. 2008 PRODUCT LINE LOGFRAME.

Targets	Products	Intended user	Outcome	Impact
PRODUCT 1	Genetic Resources Collections of CIAT Germplasm commodities upgraded and maintained up to international standards	All users of the three commodity germplasms worldwide, namely in Central and South America, Africa and South East Asia	Adoption or use designated germplasm in breeding/ agronomy programs	Better varieties, requiring less expensive inputs
Product Targets 2008	• 50% of seed accessions under long-term at -20°C • 50% of seed accessions with viability tested • 50% of accessions with health tested • 50% of accessions regenerated • 25 % of designated accessions with increased characterization (recovery of 'institutional memory') • 30% of designated accessions documented with digital images • Bar coding implemented for operations in Germplasm Health • Protocols for conservation of botanic seeds of cassava and wild *Manihot* species defined • DNA bank: protocol advanced	• NARS and CGIAR commodity breeding and research projects	• Bean breeders can have access to a wider bean diversity • All breeders can use all past evaluation data by CIAT specialists	• Designated germplasm accessed more quickly
PRODUCT 2	The germplasm of *Phaseolus* beans, *Manihot* cassava and selected tropical forages is made available to users, restored to NARS, and safely duplicated	Users, including countries of origin, worldwide can obtain quality germplasm from CIAT GRU; that germplasm is systematically safely duplicated	The "Global System" foreseen by the Trust is getting concrete for beans, cassava, and tropical forages, and the NARS can rely on the CGIAR for a full back-up of their national collections.	More benefits in the society (farmers, breeders, agronomists, but also university departments) because of access to genetic resources at anytime; stable and secure access because of the safety duplicates
Product Targets 2008	• On average 4-6,000 samples of the designated collections of beans, cassava and tropical forages are distributed to users annually • 2,800 accessions are replaced at CIP for the safety back-up of the cassava collection *in vitro* • 3,000 accessions are shipped to CIMMYT for the safety back-up of bean and tropical forages collections • 5,000 accessions are shipped to the Svalbard Global Seed Vault for the safety back-up of bean and tropical forages collections • Advances in detection of diseases of quarantine importance using the PCR reaction. • Handling of SMTAs implemented electronically	• CGIAR and NARS breeders sensu lato, NGOs, and farmers	• Increased security for the in-trust collections • Shipment of in-trust germplasm and reporting to the Treaty Secretariat made easier • Multilateral System of the International Treaty is implemented and working for the in-trust collections kept by CIAT GRU	• More sustainable food production through either improved varieties or traditional landraces restored to farmers

PRODUCT 3	The in-trust collections are genetically and socially relevant: GRU keeps the appropriate genetic diversity so that there is genetic progress in beans, cassava, and selected tropical forages.	Users worldwide can obtain the genetic variation they need in due time, now and in the future	Varieties of beans, cassava, and tropical forages that make a breakthrough in farmers' fields	Higher income for farmers, better nutrition for users, lower costs to the environment
Product Targets 2008	• Secondary gene pools of cultivated *Phaseolus* species and cassava better defined. • Monitoring of genetic erosion implemented so that germplasm explorations are carried out in due time and at the right place. • Selected explorations/ acquisitions are taking place for *Phaseolus* and cassava germplasm in countries that have ratified the Treaty. • Selected sets of germplasm are collected and restored to NARS/ farmers so that they can have access to niche markets (e.g. popping beans)	• NARS, NGOs, farmer groups, breeders in AROs, other genebanks (e.g. USDA, EMBRAPA) are getting access to unexplored sources of variability	• Breeders can include novel traits in their breeding programmes (e.g. white mold resistance in common bean breeding, delayed root deterioration in cassava breeding).	• Increased production and incomes, improved nutritional and technological traits in the crop commodities at lower environmental costs.

PRODUCT 4	Strengthened institutions that expand the scope of the conservation effort for the agricultural biological heritage	NARS in Latin America and Africa (primary target, although experience has shown that GRU training materials have been used by many other players, including Spain)	Improved capacity of NARS to tackle conservation problems of other sets of agrobiodiversity beyond the crop commodities handled by CIAT and the IARCs, namely along the two research drivers of GRU	More conservation for minor cereals, pulses, root crops and tropical fruit species, and hence diversification of the diet and of incomes for farmers worldwide
Product Targets 2008	• Updated handbook of GRU procedures that can serve as a basis for future training (e.g. hands-on training, distance education through e-learning) • Documents with best practices (joint activity involving several IARCs coordinated by SGRP) produced • Distance education course re-run for Latin America (pending on funding availability)	• NARS, NGOs, departments of universities involved in conservation of agrobiodiversity	• Increased capacities of partner institutions to use the updated handbooks of procedures and booklets with best practices	• The "Global System" envisioned by the Trust is progressively taking place for other crops that the ones dealt with by the CGIAR, with conservation and social benefits

PRODUCT 5	Improved link between conservation *ex situ* at CIAT with *in situ* conservation on farm and in the wild, resulting in larger numbers of populations of landraces and wild relatives effectively conserved	National genebanks (e.g. USDA, EMBRAPA); biodiversity institutes; major conservation agencies; national and international herbaria and museums of natural history; NGOs interested in on-farm conservation	Genetic resources of *Phaseolus* beans and *Manihot* cassava are better conserved, and hence used directly or employed in breeding programs	Improved conservation at lower costs for the societies; link between the environmental sector (i.e. protected areas) and the agricultural sector

2. List of 2008 Output Targets.

TARGETS 2008	Fully Achieved	75% Achieved	>50% Achieved	<50% Achieved	Cancelled	Deferred	VERIFICATION/ EXPLANATION
PRODUCT 1 (international standards)							Verification: GRU Annual Report 2008, and 2008 Report to the Trust.
50% of seed accessions under long-term at -20°C	X						51% of designated seed accessions in long-term at CIAT.
50% of seed accessions with viability tested	X						60% of designated seed accessions tested for viability.
50% of seed accessions with health tested	X						75% of designated accessions have been tested.
50% of seed accessions regenerated50% of seed accessions regenerated	X						55% of forage accessions regenerated (91% in beans).
25% of seed accessions characterized	X						30% of seed accessions characterized.
30% of accessions with digital images	X						27,932 digital images available on the web site.
Bar coding implemented in Germplasm Health Lab	X						Bar coding pending in seed reception. Harvest of botanic seeds of different *Manihot* species in progress; reduction of area in Quilichao is a problem.
Protocols for conservation of botanic seeds of *Manihot* species DNA bank protocol advanced	X			X			Protocol established for beans and cassava.

PRODUCT 2 (GR distributed and safely replicated)							Verification: GRU Annual Report 2008, 2008 Report to the Trust, and GPG2 Report for 2008.
• On average 4-6,000 samples of the designated collections of beans, cassava and tropical forages are distributed to users annually	X						5,720 samples of beans, 332 samples of tropical forages, and 1,348 samples of cassava distributed to users in 2008.
• 2,800 accessions are replaced at CIP for the safety back-up of the cassava collection *in vitro*.	X						2,119 materials were sent to CIP, in reply to their evaluation, meaning that the slow-growth is working.
• 3,000 accessions are shipped to CIMMYT for the safety back-up of bean and tropical forages collections.	X						4,023 accessions (2,543 of beans and 1,480 of forages) were sent to CIMMYT in 2008.
• 5,000 accessions are shipped to the Svalbard Global Seed Vault for the safety back-up of bean and tropical forages collections.	X						30,911 accessions (21,698 of beans and 9,213 of forages) were sent to Svalbard in 2008; this work initiated in 1997 and went strong since 2003.
• Advances in detection of diseases of quarantine importance using the CR reaction.	X						A PCR detection test for Frog Skin Disease of cassava was published in 2008.
• Handling of SMTAs implemented electronically.	X						This has been implemented and has served as model for CIAT Breeding Projects.

PRODUCT 3 (Genetic relevance of designated collections)							Verification: GRU Annual Report 2008; http://www.ciat.cgiar.org/urg.
• Secondary gene pools of cultivated *Phaseolus* species and cassava better defined.	X						A phylogeny of 28 wild species of *Manihot* and 3 subspecies of cassava was published in 2008.
• Monitoring of genetic erosion implemented, so that germplasm explorations are carried out in due time and at the right place.						X	While CONABIO of Mexico got the data, they could not hire people this year.
• Selected explorations/ acquisitions are taking place for *Phaseolus* and cassava germplasm in countries that have ratified the Treaty.	X						140 accessions (landraces) introduced from Indonesia; introduction of 16 clones from high altitude location in Colombia.

PRODUCT 4 (Strengthened institutions)							Verification: GRU Annual Report 2008, GPG2 Report for 2008; http://www.ciat.cgiar.org/urg.
• Updated handbook of GRU procedures that can serve as a basis for future training (e.g. hands-on training, distance education through e-learning).	X						Handbook of procedures for *in vitro* and germplasm health on GRU web site.
• Documents with best practices (joint activity involving several IARCs coordinated by SGRP) produced.	X						Handbook of procedures for germplasm production and conservation in writing phase.
• Distance education course re-run for Latin America (pending on funding availability).	X						No course because of lack of funding, but 17 Professionals from Latin America, Nigeria, Spain, trained by individual training.

PRODUCT 5 (Improved link *ex situ/ in situ*)							Verification: GRU Annual Report 2008; 2008 Report to the Trust; http://www.ciat.cgiar.org/urg.
• Populations of *Phaseolus* beans documented in herbaria (two institutes not yet visited per year).	X						In 2008, the following herbaria were studied: CAS, CS, DAV, FI, IBUG, IEB, O and TEX.
• GIS mapping for at least one species of bean for all populations ever mentioned in the western hemisphere.	X						Done for the *Brevilegumeni* for Costa Rica for the publication of a new species
• Gene flow documented for beans/ or cassava in view of on-farm conservation.		X					Done for two groups of Lima beans from Mexico and Peru, respectively, in view of a future publication.

3. Research Highlights 2008

A published result has been the definition of a PCR protocol for the detection of Frog Skin Disease in cassava. This viral disease endemic in all countries of Amazonia can harm cassava production very much, with no symptoms expressed on the aerial part in most cases but no root. The regular testing by grafting a hypersensitive clone on the rootstock to be tested takes 13 months, although a minigrafting improved by GRU in the past takes 21 weeks. With the rt-PCR test, the result is obtained in 5 days. The parallel testing grafting and PCR performed on 508 clones from 21 countries has shown an excellent correlation, with a plus for the rt-PCR in sensitiveness of detection. Apart from applications in cassava research, this protocol is very useful in germplasm movement, because it can be done on *in vitro* materials, thus at the very beginning of any introduction process. It is also very safe for tropical countries where introduction of the viral disease is feared.

DNA sequencing on two cpDNA intergenic spacers (*trn*L-*trn*F and *atp*B-*rbc*L) and the ITS/5.8S region was performed to investigate the genetic relationships among wild and domesticated Lima beans. The Neighbor-Joining topologies built on the bases of cpDNA and ITS/5.8S polymorphisms reveal three gene pools in this crop, while only two have long been recognized by many authors. One gene pool called 'AI' with wild forms from SW Ecuador and NW Peru and large-seeded cultivated forms from the same region, confirms the domestication of the latter from the former. A second gene pool called 'MI' includes mostly wild forms from Mexico west of Tehuantepec and small-seeded domesticated forms from the same region and from Central America and South America, evidencing a possible domestication west of Tehuantepec in Mexico. A third gene pool called 'MII' includes mostly wild forms from Mexico east of Tehuantepec, different countries of Central America, Cuba, Colombia, eastern Peru, and Argentina, and two small-seeded domesticated accessions from Brazil, opening up the possibility of a third domestication event, the precise location of which now depends on further exploration of this gene pool.

4. Project Outcome: Safety backup of CIAT designated seed collections (beans and tropical forages) into the Svalbard Global Seed Vault, resulting in a worldwide recognition of the role of the CGIAR in saving agricultural biodiversity.

It is wise practice in genebanks to deposit copies of genetic collections as safety backups, in appropriate facilities (Product 2 of GRU Product Line); making safety backups was listed in MTP 2004-2006 and MTP 2006-2008 (it was also a priority within the GPG1 and GPG2 collective efforts of Upgrading the CGIAR genebanks, supported by the World Bank). It is responsible practice when as in the case of CIAT the agricultural heritage of 141 countries is kept in trust. For filling such objectives, it was necessary to produce all that seed, clean it, check viability and health status, and then pack according to the highest quality standards. On December 19, 2007, CIAT made an agreement with the Ministry of Agriculture and Food of Norway to deposit a copy of the seed collections of beans (35,896 accessions to date) and tropical forages (23,140 accessions) into the Svalbard Global Seed Vault. Two shipments (30,911 on Jan 30, 2008, and 3,200 on Feb 9, 2009) have been made to date, securing a total of 34,111 accessions (23,812 of beans and 10,299 of forages). CIAT was third in the number of accessions deposited among the first twenty depositors on the Opening of the Vault (the first two depositors: IRRI with 70,180, CIMMYT with 57,721, and

CIAT with 30,911), on February 26, 2008. The contribution by CIAT was key for filling the CGIAR target of 200,000 accessions at Svalbard for the Opening, as agreed with the Global Crop Diversity Trust that sponsored the initiative. It was an interesting and unmatched case of cooperation, from near the equator to the artic polar circle, involving the Centers in places as distant as Palmira, Los Baños, Texcoco, Aleppo, Ibadan, Lima, Addis, the secretariat of the Trust in Rome, and the Nordic genebank in Oslo and in Longyearbyen.

The outcome was three-fold: an increased security for the in-trust collections, a responsible implementation of the agreement of 16 October 2006 between CIAT and the Governing Body of the International Treaty, and a worldwide recognition of the role of the CGIAR in saving agricultural biodiversity for present and future generations. During the opening and afterwards, there were over 100 articles (in journals such as Time, the New York Times, International Herald Tribune, US News, The Guardian, Science, Popular Science), press releases, interviews covering the work of CIAT and sister Centers towards that goal. A compilation of many of the press articles, specially in Latin American countries, can be found in http://www.ciat.cgiar.org/urg, then GRU Files ("CIAT at Svalbard: a report"). The visibility of the CGIAR towards the world media was second to that when Dr Borlaug won the Nobel prize. The Vault was evaluated as the 6th Best Inventions of the Year 2008 by Time Magazine of November 10, 2008, and without the seeds from the CGIAR, there would have been no start of a functional Vault in Svalbard. Beyond the technical aspects, and as stressed by Mr Stoltenberg of Norway and Mr Barroso of EU, a lasting impact of the Vault and the seeds therein is the message to the world community that our survival depends on conservation.

5. A list of 2008 publications

5.1. Articles in refereed journals

Calvert, L.A., **M. Cuervo**, I. Lozano, N. Villareal & J. Arroyave. 2008. Identification of three strains of a virus associated with cassava plants affected by frogskin disease. J. Phytopathology **156**: 647-653.

Chacón, J., S. Madriñán, **D.G. Debouck**, F. Rodríguez & J. Tohme. 2008. Phylogenetic patterns in the genus *Manihot* (Euphorbiaceae) inferred from analyses of nuclear and chloroplast regions. Molec. Phylogen. Evol. **49** (1): 260-267.

Gaiji, S. & **D.G. Debouck.** 2008. Flujos de germoplasma en las Américas: 30 años de distribución de muestras de frijol por parte del Centro Internacional de Agricultura Tropical. Recursos Naturales y Ambiente **53:** 54-61.

Montoya, C.A., P. Leterme, N.F. Victoria, **O. Toro**, W.B. Souffrant, S. Beebe & J.-P. Lallès. 2008. Susceptibility of phaseolin to *in vitro* proteolysis is highly variable across common bean varieties (*Phaseolus vulgaris*). J. Agric. Food Chem. **56:** 2183-2191.

Reichel, H., **Cuervo, M.** & F. Morales. 2008. Caracterización parcial de un potexvirus aislado de *Musa coccinea* afectada por rayado neurótico en Colombia. Agronomía Colombiana **26** (2): 285-291.

5.2. Articles in non-refereed journals

D.G. Debouck, R. Herrera T. & R. Araya V. 2008. New populations of wild common bean disclosed in Nicaragua. Annu. Rept. Bean Improvement Coop. (USA) **51**: 120-121.

Ocampo, C. H. & O. Toro. 2008. Phaseolin diversity of Nicaraguan common bean germplasm held at CIAT. Annu. Rept. Bean Improvement Coop. (USA) **51**: 122-123

5.3. Papers presented at formal conferences and workshops with external attendance

Aranzales, E. Lima, Perú, 22-23 November 2008, invited REDARFIT meeting: "Conservación *in vitro* de germoplasma del género *Manihot."*

Aranzales, E. Palmira, Colombia, 30 October 2008, invited Universidad Nacional de Colombia: "Aplicaciones prácticas de las técnicas de cultivo de tejidos vegetales."

Cuervo, M., Lozano, I., and Morales, F. Palmira, Colombia, 26 November 2008, invited internal seminal in CIAT: "Historia, caracterización y manejo de la enfermedad viral más importante de la especie *Manihot esculenta* en su centro de origen."

Debouck, D.G. Palmira, Colombia, 3 December 2008, invited internal seminar in CIAT: "1,2,3: a new strategic look at a great crop."

Debouck, D.G. Davis, California, USA, 15 September 2008, invited conference at the Harlan II Symposium on the domestication of crop plants and domestic animals: "Domestication of Lima beans: a new look at an old problem."

Debouck, D.G. Celaya, México, 22 May 2008, invited conference at the 1st international bean congress: "Recursos genéticos y patentes: el caso del fríjol común."

Debouck, D.G. Quito, Ecuador, 9 May 2008, invited seminar at the regional workshop of IUCN on certificate of origin: "Perspectivas desde un Centro Internacional sobre la Trazabilidad."

Debouck, D.G. Lima, Perú, 6 May 2008, invited conference at the 13th Latinamerican Congress of Genetics: "Experiencias con los Recursos Fitogenéticos de Fríjol y Yuca."

Debouck, D.G. Palmira, Colombia, 7 March 2008, invited internal seminar in CIAT: "Implications of the International Treaty on Plant Genetic Resources for Food and Agriculture for the work of the Center."

Toro, J.O. 2008. Banco Mundial de *Phaseolus* spp. L., CIAT – URG, Aspectos relevantes en regeneración. Presentación en Power point (PDF), REDARFIT 22-23 de Noviembre, Lima, Perú.

6. List of proposals funded in 2008, should be prepared, including total dollar value of each contract and donor, and the amount going directly to CIAT for implementation.

Project	Title	Acronym	Donor	Starting Date	Ending Date	Outcome Line	Total Budget	CIAT Budget
GRA52	The Long-Term conservation and sustainable utilization of the ex situ collection of Bean and Cassava germplasm held by CIAT	GCDT	The Global Crop Diversity Trust.	1-Jan-08	31-Dec-12	SBA-5	120,000	120,000
GRA50	The Long-Term conservation and sustainable utilization of the ex situ collection of Bean and Cassava germplasm held by CIAT	GCDT	The Global Crop Diversity Trust.	1-Jan-08	31-Dec-12	SBA-5	150,000	150,000
GRG57	Development and refinement of cryopreservation protocols for the long-term conservation on vegetatively propagated crops	GCDT	The Global Crop Diversity Trust.	15-Jul-08	30-Sep-09	SBA-5	37,950	37,950
CPA70	Industrial Uses of Cassava-Waxy Starch	TTDI	Thailand Tapioca Development Institute.	01-Jan-08	31-Dic-12	SBA-2	62,501	62,501
GRC3	Rehabilitation of International Public Goods; CGIAR Genebanks Upgrading Project, Global Public Goods, Phase 2).	WB	World Bank	01-Jan-08	31-Dec-08	SBA-5	208,010	208,010

7. Staff list: (indicate % time assignment)
Daniel G. Debouck, Head, PhD (100%)
Celia Lima, Agr. Engineer, M.Sc. (100%)
Graciela Mafla, Biologist (100%)
Maritza Cuervo, Agr. Engineer, M.Sc. (100%)
César Ocampo, Biologist, M.Sc. (100%)
Orlando Toro, Technician (100%)
Arsenio Ciprián, Technician (100%)
Roosevelt Escobar, Biologist, M.Sc. (50%)
Ericson Aranzales, Ing. Biotec. (100%)
Maria del Socorro Balcázar, Bacteriologist (100%)
Ángela Marcela Hernández, Information System Analyst (100%)
Josefina Martínez, Secretary (100%)
Eliana Urquijo, Secretary (100%)

8. Summary budget prepared by Finances:

Source	Amount (US$)	Proportion (%)
Unrestricted core	721,659	58.6
Sub Total	**721,659**	
Special projects		
Long-Term conservation-Cassava	120,000	9.7
Long-Term conservation-Beans	150,000	12.2
Development and refinement of cryoconservation	14,000	1.1
Upgrading Plan Operations 2	208,010	16.9
Cassava genetic resources (TTDI)	16,667	1.3
Sub Total	**508,677**	
TOTAL	**1,230,336**	**100.0**

Product 1. The International Standards

1.1. Backlogs of received materials processed

1.1.1. Introduction of germoplasm into the genebank processes

Within the process of documenting the 'Institutional Memory', thirty-six new accessions were added to the bank from the elite lines produced by the Bean Improvement project of CIAT. Of these, twenty correspond to lines with bc3 resistant gene to BCMV and resistance gene to race NL3 of BCMV, twenty-four present high tolerance to anthracnose (*Colletotrichum lindemuthianum*), with reaction 1 (no disease sympthoms visible) in conditions of artificial inoculation, and thirtheen have yield rates of over 2,000 Kg/Ha. Table 1 reports the new introductions.

Table 1. New elite lines from CIAT Bean Improvement Program introduced in the genebank in 2008.

GNumber	Code	Release Name or other Identification	Observation
G51506	AFR 724		bc3 resistant gene to BCMV
G51507	BRC 2		bc3 resistant gene to BCMV
G51508	BRC 4		bc3 resistant gene to BCMV
G51509	BRC 6		bc3 resistant gene to BCMV
G51510	BRC 9		bc3 resistant gene to BCMV
G51511	BRC 12		bc3 resistant gene to BCMV
G51512	BRC 13		bc3 resistant gene to BCMV
G51513	BRC 14		bc3 resistant gene to BCMV
G51514	BRC 15		bc3 resistant gene to BCMV
G51515	BRC 17		bc3 resistant gene to BCMV
G51516	BRC 18		bc3 resistant gene to BCMV
G51517	BRC 20		bc3 resistant gene to BCMV
G51518	BRC 21		bc3 resistant gene to BCMV
G51519	BRC 23		bc3 resistant gene to BCMV
G51520	BRC 24		bc3 resistant gene to BCMV
G51521	BRC 25		bc3 resistant gene to BCMV
G51522	BRC 28		bc3 resistant gene to BCMV
G51523	BRC 29		bc3 resistant gene to BCMV
G51524	BRC 33		bc3 resistant gene to BCMV
G51525	BRC 34		bc3 resistant gene to BCMV
G51526	CAR 47		Tolerance to anthracnose
G51527	CAL 153		High yield rate
G51528	CAL 154		High yield rate
G51529	CAL 155		High yield rate
G51530	CAL 156		High yield rate
G51531	CAL 160		Tolerance to anthracnose
G51532	CAL 179		Good performance
G51533	CAL 164		Tolerance to anthracnose
G51534	CAL 167		Has I gene (strain NL3)
G51535	CAL 172		Tolerance to anthracnose
G51536	CAL 178		Good performance
G51537	CAB 2		Commercial variety in Rwanda
G51538	CAB 19		Commercial variety in Rwanda

G51539	ZAA 12	High yield rate
G51540	ZAA 54	High yield rate
G51541	ZAA 55	High yield rate

1.2. Backlog materials

During 2008, a total of 281 accessions from the 'historic backlog' were handled following scarification methods - disinfection – pre-germination, combined with substrates such as germination paper, sand, soil, petri-dishes and others. Similarly, we continue with *in vitro* culture methods for both whole seeds and embryos to rescue wild accessions of *Phaseolus sensu stricto* and cultivated materials with germination problems. Table 2 reports progress made in 2008.

Table 2. Backlog pending and processed in 2008.

Description	Beans
Germplasm pending in 2007	7,988
Germplasm processed in 2008	281
Germplasm pending	7,707

1.2.1. Material cleared by quarantine authorities

Visits to CIAT station in Popayan, by the staff of Germplasm Health Laboratory of URG were made to certify the health cleanliness of germplasm produced there. Additionally, all these materials go post harvest through monitoring by the Germplasm Health Laboratory, to certify its quality. In 2007-2008, a total of 4,067 materials were inspected in compliance with these standards.

1.2.2. Materials multiplied/ regenerated

The processes of regeneration and multiplication were severely affected by "climate change", namely by the high levels of rain that occurred during the entire period 2008, with levels above 3,500 mm per year. Table 3 reports the number of accessions handled by GRU in the different stations.

Table 3. Number of accessions handled by species and locality to December 2008.

	Palmira		Popayán		Tenerife		Total	
Species	Delivered	Goal fulfilled	Delivered	Goal fulfilled	Delivered	Goal fulfilled	Delivered	Goal fulfilled
P. vulgaris	70	5	3,616	1,782	9	5	2,669	1,792
Complex *coccineus*			1	1			1	1
P. lunatus	279	226	45	37	1		325	263
Otras spp.	27	16	8	3	12	8	47	27
Total	376	247	3,670	1,823	22	13	3,041	2,083

Table 4. Forage germplasm planted for multiplication and regeneration under greenhouse/mesh-house and field conditions (number of accessions).

Localities	Legumes	Grasses	Total
Greenhouse/Mesh-house	1,827	77	1,904
Quilichao	1,593	134	1,727
Palmira	1,095	242	1,337
Popayán	120	612	732
Total	4,635	1,065	5,700

Table 5. New forage germplasm installed during 2008.

	Quilichao	Palmira	Popayan	Total
Sown during 2008	1,678	1,043	42	2,763

Table 6. New forage germplasm characterized during 2008.

	Legumes	Grasses	Total
Characterized during the process	2,065	4	2,069

Contributors: O. Toro, A. Ciprián.

Status of designated germplasm at the GRU in 2008

Manihot cassava: 6,467 (out of which 5,584 of *M. esculenta*)
Phaseolus beans: 35,896
Tropical forages: 23,140
Total: 65,503

1.2.3. Periodical subculturing of the FAO designate cassava collection

This year, 8,261 accessions of *Manihot* were subcultured by the nodal cutting technique. A total of 1,348 accessions (3,462 *in vitro* plants) were propagated for the distribution to users, 292 accessions (362 *in vitro* plants) were propagated for indexing tests, and 2,119 accessions (6,326 *in vitro* plants) were propagated for the security backup.

Contributors: G. Mafla, E. Aranzales

Ouput 1.3. Materials processed into final packing

Activity 1.3.1. Final drying and temporary storage

Table 7 indicates the amount of accessions for beans (4,575) and forages (1,574), respectively, (total 6,149), which have been harvested, cleaned, dried, and stored at 5°C, awaiting the results from viability and health tests.

Table 7. Germplasm in seed processing during 2008.

	Beans	Forages
Seed selection / temporal storage	4,575	1,574
Total	**4,575**	**1,574**

Contributors: C. Lima, A. Ciprián, O. Toro

Activity 1.3.2. Viability testing

Table 8 indicates flows of materials during 2008 for viability testing. It shows the importance of good drying and other procedures following the gene bank standards (FAO/IPGRI, 1994). Ranges of germination were chosen because they indicated priorities for regeneration in the field/ glass-houses.

Table 8. Viability testing for *Phaseolus* beans and tropical forages during 2008.

	BEANS		FORAGES	
Class	**Germination (%)**	**No. Accessions**	**Germination (%)**	**No. Accessions**
Already stored materials	1-64 65-84 85-100	69 147 725	1-64 65-84 85-100	6 4 18
Sub-Total		**941**		**28**
Short Term stored materials to Long Term Storage	1-64 65-84 85-100	87 128 1,642		
Recently multiplied materials	1-64 65-84 85-100	18 49 1,874	1-64 65-84 85-100	84 191 1,422
Sub-Total		**3,798**		**1,627**
TOTAL		**4,739**		**1,725**

Literature cited

FAO/IPGRI 1994. Genebank standards. Rome, Food and Agriculture Organization of the United Nations and Institute Plant Genetic Resources Institute. 17 p.

Contributor: C. Lima.

Activity 1.3.3. Final packing and seed germplasm orientation into the five conservation targets.

Once cleared by the Viability Lab (for viability above 85%) and by the Germplasm Health Lab (for absence of diseases of quarantine importance), the materials are processed into the five conservation purposes: long-term, safe duplicates, restoration, periodical monitoring, and distribution (Tables 9-10).

Table 9. Final storage and packing of *Phaseolus* beans processed during 2008 (number of accessions).

	Beans
Base, duplicates, repatriation, monitoring, and distribution	4,007
Refreshed seed	2,628
Total	**6,635**

Table 10. Final storage and packing of tropical forages processed during 2008 (number of accessions).

	Total
Base, duplicates, repatriation, monitoring, distribution	1,480
Refreshed seed	151
Total	**1,631**

Contributor: C. Lima

Activity 1.3.4. Monitoring the viability of conserved seed germplasm of beans and forages.

In 2008 we tested the monitoring period of *Phaseolus vulgaris* seeds that were conserved for a period of 10 years at long-term storage. Three groups of seeds were conserved: Ten1995B, Ten1996A and Ten1996B. A total of 1,690 accessions were tested and the results are shown in Table 11.

After 10years of conservation the mean of germination increased 0.55 units for Ten1995B, 1.02 for Ten1996A and decreased 1.65 for Ten1996B. In Ten 1995B the difference was not statistically significant. The difference is statistically significant in Ten1996A and Ten1996B cases with confidence interval of 95%.

Table 11. Paired T-Test for monitoring bean seeds after 10 years of long-term conservation.

Source	% Germ	Mean	StDev	SE Mean	N	Difference	Std.Dv.Diff	T-value	P-Value
Ten1995B	Initial	97.315	5.383	0.212					
	Monitored	97.866	5.737	0.226	642	-0.551	7.527	-1.86	0.064
Ten1996A	Initial	97.526	3.349	0.136					
	Monitored	98.551	3.723	0.151	606	-1.025	4.889	-5.16	0.000
Ten1996B	Initial	97.471	3.346	0.159					
	Monitored	98.538	2.672	0.127	442	-0.168	3.792	-5.92	0.000

Similarly, the monitoring test was done for forage germplasm conserved after a 10 and 5 years period. A group of 251 forages species were conserved during 1998 and 453 during 2003. The results are shown in Table 12 and Table 13. The difference of the mean germination was of 2.16 units in the group conserved during 10 years and of 4.78 in the groups conserved during 5 years. These differences are statistically significant with confidence interval of 95%.

According to these results, for the seed lots monitored a total of 28 (1.7%) accessions of beans conserved in 1998 need to be refreshed by seed multiplication following the protocols for seed conservation of germination above 85%. For the forage seeds group conserved in 1998 a total of 31 (12.3%) needed to be refreshed and for the group conserved in 2003 there were 96 (21.2%) accessions above 85%.

Table 12. Paired T-Test for monitoring forage seeds after 10 years of long-term conservation.

% Germ	Mean	StDev	SE Mean	N	Difference	Std.Dv.Diff	T-value	P-Value
Initial	93.012	6.178	0.390					
Monitored	90.398	10.759	0.679	251	2.164	10.982	3.77	0.000

Table 13. Paired T-Test for monitoring forage seeds after 5 years of long-term conservation.

% Germ	Mean	StDev	SE Mean	N	Difference	Std.Dv.Diff	T-value	P-Value
Initial	92.821	4.377	0.206					
Monitored	88.035	14.300	0.672	453	4.786	13.863	7.35	0.000

Literature cited

FAO/IPGRI (1994). Genebank standards. Rome, Italy, Food and Agriculture Organization of The United Nations and Institute Plant Genetic Resources Institute: 17 p.

Contributors: M.C. Lima

Product 2. GR distributed and safely replicated

Output 2.1 FAO designate collections cleaned against seed borne diseases

Activity 2.1.1 Indexing and cleaning the cassava collection

We continued with indexing activities of clones of the Cassava World Collection maintained under *in vitro* conditions at CIAT. The final objective of this activity is to make available the accessions for distribution, once the collections checked for the three viruses currently known of quarantine importance, following the FAO/IPGRI recommendations for the safe movement of cassava clones at national and international levels.

The plants developed *in vitro* after the thermotherapy (Mafla, G. et. al., 1984) are tested for cassava virus diseases: Cassava Common Mosaic Virus (CsCMV), Cassava X Virus (CsXV) and Cassava Frog Skin Disease (CFSD). The following diagnosis techniques are used: ELISA for CsCMV and CsXV, and grafting with a hypersensitive clone (MCOL 2063) for the CFSD. This year the GHL also used a molecular method for testing CFSD for the analysis by RT-PCR.

The total Cassava World Collection kept in GRU designated by the FAO is of 6.467 accessions, of which 5.184 (80.2%) corresponds to landraces, 400 (6.2%) breeding material and 883 (13.6%) wild species. Of these, 5420 are available for distribution corresponding to 86.8 % (Figure 1).

The total of negative clones evaluated for the three viruses in 2008 is 305 (CsCMV (28), CsXV (28), CFSV through grafting (95), and CFSD by RT-PCR (424)).

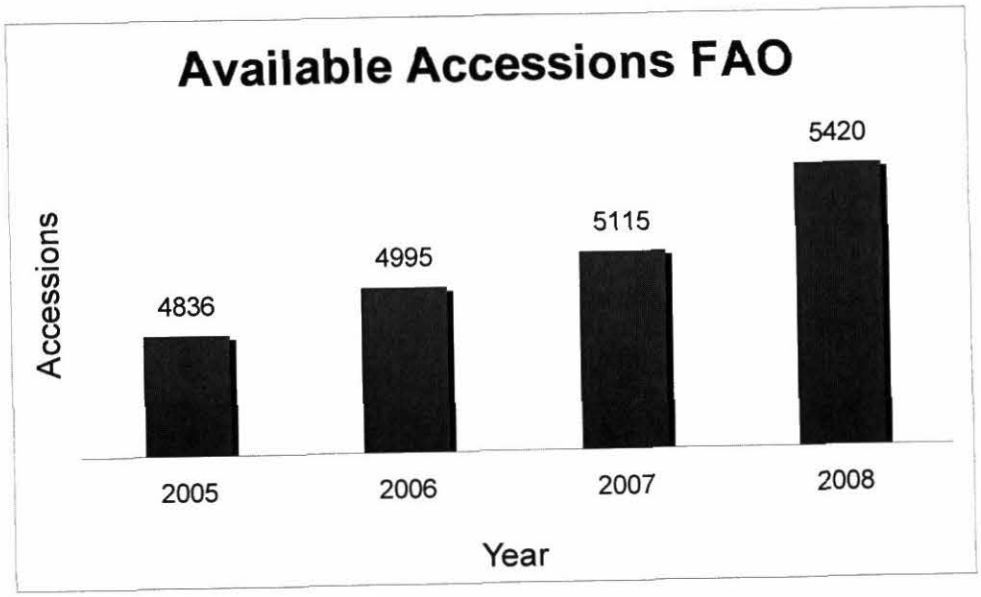

Figure 1. Available accessions of the cassava germplasm collection.

The total progress in the indexing of the cassava collection (negatives clones for three viruses) in the previous years is shown in figure 2.

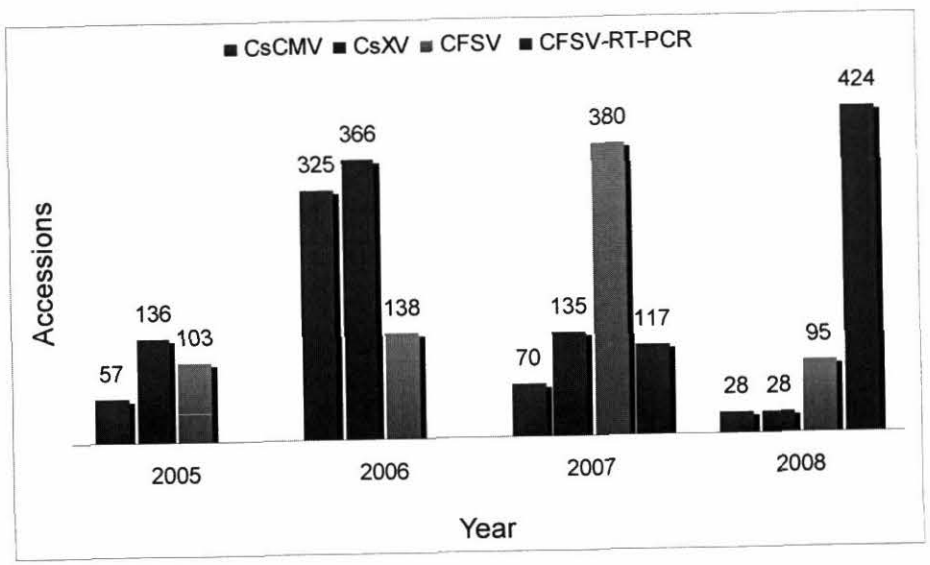

Figure 2. Number of clones evaluated for each virus 2005 – 2008.

As shown in Figure 3, 5420 materials are available for distribution, out of which 400 (7.4 %) are Breeding material, 82 (1.5 %) are Wild Species and 4964 (91.5 %) are Landraces.

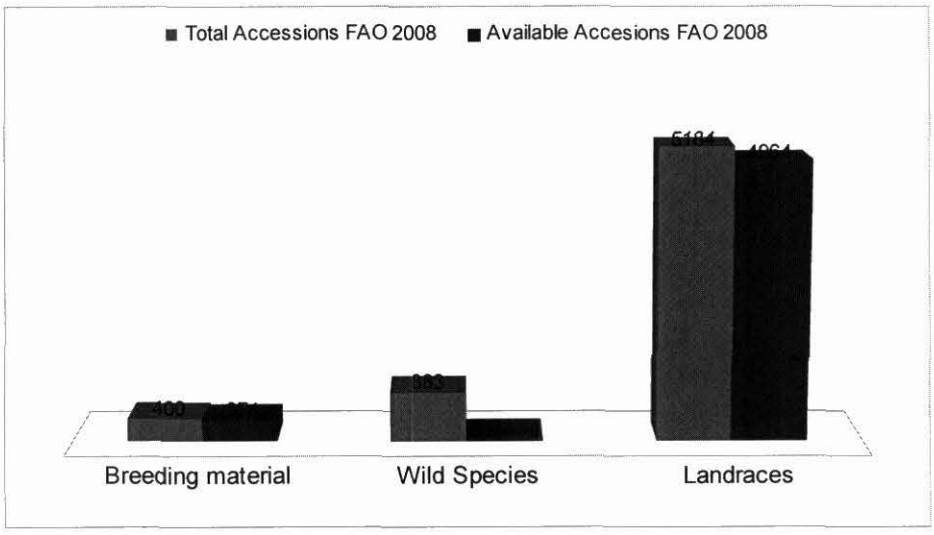

Figure 3. Availability status of the cassava germplasm collection.

A total of 99.5% (5,556) of the collection of *Manihot esculenta* (breeding material: 400; landraces: 5,184) have been evaluated by the PCR technology and are available for distribution.

Activity 2.1.2 Application of a molecular technique for the detection of Cassava Frog Skin Virus (CFSV).

Detection of Cassava Frogskin Virus (CFSV) has so far been made by grafting. This process takes an average of three months for plants to have the right size for the grafting, and three weeks to observe the characteristic symptoms in the Secundina clones MCOL 2063 (highly susceptible to viruses). In some cases, it takes longer because some varieties cannot be easily established in the greenhouse, making it difficult to complete the indexing of the entire collection of cassava germplasm (Flor et al, 2003). With the purpose of reducing the time, a molecular method (rt-PCR) has been implemented for the detection of CsFSV in the Virology lab, which only takes three days and is more sensitive and specific (Cuervo 2006).

The implementation and use of this molecular method (rt-PCR) is of great importance, not only for the production of material free of the cassava frogskin virus, but also for the conservation and distribution of the genetic resources of cassava deposited in the Germplasm Bank of CIAT. To that end both methodologies available are compared for the detection of the virus.

Over 500 accessions of cassava from the genebank and from different countries were evaluated using both the grafting and the rt-PCR technique, in order to compare their reliability and relative advantages.

Clearly 501 out of 508 evaluated accessions were free of the virus, and only 4 accessions were carriers of the virus according to both used methods, which shows the high sensibility of both evaluated methodologies. Nevertheless, three of the samples evaluated as free of the virus by the grafting test, were detected as infected by the CFSV by rt-PCR, which demonstrates the higher reliability of this new methodology.

Table 14. Report of samples indexed by rt-PCR .

Landraces		Grafting result	rt-PCR result
Argentina	34	34 Negative	34 Negative
Brazil	91	88 Negative	88 Negative
		2 Positive	2 Positive
		1 Negative	1 Positive
Colombia	91	89 Negative	89 Negative
		2 Negative	2 Positive
Costa Rica	2	2 Negative	2 Negative
Ecuador	20	20 Negative	20 Negative
FJI	3	3 Negative	3 Negative
Guatemala	11	11 Negatives	11 Negatives
Indonesia	88	88 Negative	88 Negative
Malaysia	6	6 Negative	6 Negative
Mexico	8	8 Negative	8 Negative
Panama	8	8 Negative	8 Negative
Paraguay	23	23 Negative	23 Negative
Peru	15	15 Negative	15 Negative
Venezuela	13	13 Negative	13 Negative
Vietnam	1	1 Negative	1 Negative
Philippines	2	2 Negative	2 Negative
Thailand	3	3 Negative	3 Negative
USA	1	1 Negative	1 Negative
Cuba	6	6 Negative	6 Negative
Dominican Republic	1	1 Negative	1 Negative
Nigeria	2	2 Negative	2 Negative
Others	2	2 Positive	2 Positive
Wild species	43	43 Negative	43 Negative
Breeding material	34	34 Negative	34 Negative
Positive controls	8	8 Positive	8 Positive
Negative controls	8	8 Negative	8 Negative
Negative by Grafting test and rt-PCR	501		
Positive by Grafting test and rt-PCR	4		
Positive by Grafting test and rt-PCR Positive controls	8		
Negative by Grafting test and rt-PCR Negative controls	8		
Negative by Grafting test and Positive by rt-PCR	3		
Total evaluated	524		

Contributors: M. Cuervo I., G. Mafla, E. Aranzales.

Activity 2.1.3 Establishment of a "Bonsai" collection as safety back-up of the whole Cassava Collection.

Since October 2001 we began to establish one copy of the whole cassava collection under greenhouse conditions. Along our current agreement with FAO, this backup is necessary for safely maintained the entire collection of Cassava. At the moment, in the Bonsai Greenhouses we are keeping 885 clones of different sources (Figure 4, Table 15).

Figure 4. Cassava plants established as "Bonsai" collection as back up for the In Vitro Bank.

Activity 2.1.4. Updating the Cassava ORACLE database

We continue updating the ORACLE database with the new results of indexing (to CCMV, CsXV and FSDA) and with the dates of the new bonsai clones.

References

Flor. N.C., Mafla G., Danny M. Montero. Annual Report 2003. CIAT Project on Saving Agrobiodiversity SB 01/02. 2003.

Cuervo M. Masters Thesis: Caracterizacion molecular de algunos aislamientos del virus del cuero de sapo de la yuca recolectados en diferentes zonas de Colombia. Universidad Nacional de Colombia, sede Palmira. 2006.

Table 15. Cassava plants established as "Bonsai" collection

Landraces	
ARGENTINA	37
BRASIL	206
COLOMBIA	246
COSTA RICA	15
CUBA	6
REPUBLICA DOMINICANA	1
ECUADOR	12
GUATEMALA	10
INDONESIA	80
MALASIA	9
MEXICO	17
NIGERIA	5
PANAMA	8
PARAGUAY	60
PERU	31
Philippines	2
Puerto Rico	2
USA	1
TAILANDIA	6
VENEZUELA	26
Breeding material	
SG	4
SM	9
CG	12
CM	29
Wild Species	
CTH *(Manihot carthaginensis)*	13
CAE *(Manihot caerulescens)*	1
FLA *(Manihot esculenta subsp. Flabellifolia)*	20
FMT *(Manihot filamentosa)*	4
GLA *(Manihot glaziovi)*	1
GUT *(Manihot guaranitica)*	3
JAC	3
PNT *(Manihot pentaphylla)*	1
PSE *(Manihot pseudoglaziovii)*	1
RUB *(Manihot rubricaulis)*	1
TST *(Manihot tristis)*	3
TOTAL	885

2.1.5. Germplasm health control in seed germplasm

Introduction

The main responsibility of the Germplasm Health Laboratory (GHL) at the International Center of Tropical Agriculture (CIAT) is to test the health status of beans, tropical pastures and cassava germplasm stored at CIAT's genebank. The purpose of the Lab is to ensure that the designated germplasm is kept under the international phytosanitary standards for each crop commodity, and also to ensure that the germplasm distributed by the Genetic Resources Unit (GRU) is free of diseases of quarantine importance.

To minimize the phytosanitary risks associated with the movement of germplasm, especially concerning the inadvertent transport of pathogens and pests of significance quarantine, *CIAT follows a regulatory* and quarantine program, in close cooperation with the plant quarantine authority of its host country the Instituto Colombiano Agropecuario (ICA).

During the period January 2008- December 2008, the GHL tested 7,708 seed samples (4,713 bean seeds samples, 1,805 legume forages and tropical grasses seed samples from GRU seed increases. We also tested 1,127 samples of bean seeds and 63 of tropical grasses and legumes from projects such as the Mesoamerican Bean Genetics, Andean Bean Genetics, Tropical grasses and legumes, and others.

Materials and Methods

CIAT's facilities for germplasm health testing are designed as a multifunctional laboratory to test seeds and tissues for fungi, bacteria, viruses, and occasionally nematodes (Table 16).

The GHL practices phytosanitary inspections on multiplication plots (fields and glass-houses), and apply indexing procedures in the laboratory to ensure that the germplasm is free of seed borne diseases that could affect its longevity during the storage and prohibit its distribution to users.

Accessions are tested in the GHL using accepted methodologies to identify seed-borne pathogens as fungi, bacteria and viruses according with those pathogens recorded in seed production areas. To detect pathogens of quarantine significance, the GHL uses the methodologies recommended by CIAT's pathologists and virologists. When a recipient country request additional statements, the GHL executes additional tests whenever possible to comply the specific quarantine regulations of the recipient country. All seed health lab results are stored in the GRU database.

Table 16. Pathogens of quarantine significance for Beans and Tropical Pastures tested by the GHL.

Bean	Tropical Pastures
Fungi	
Alternaria tenuis	*Alternaria tenuis*
Ascochyta spp.	*Ascochyta spp.*
Botrytis cinerea	*Botrytis cinerea*
Cercospora canescens	*Cercospora canescens*
Colletotrichum gloeosporioides	*Colletotrichum gloeosporioides*
Colletotrichum lindemuthianum	*Colletotrichum truncatum*
Colletotrichum truncatum	*Curvularia sp.*
Curvularia sp.	*Drechslera sp.*
Fusarium oxisporum	*Helminthosporium sp.*
Macrophoma sp.	*Macrophoma sp.*
Macrophomina phaseolina	*Macrophomina phaseolina*
Pestalotia sp.	*Phoma exigua var. diversipora.*
Phomopsis sp.	*Phomopsis sp.*
Phoma exigua var. diversipora.	*Pestalotia sp.*
Phaeoisariopsis griseola	*Pyricularia sp.*
Rhizoctonia solani	*Rhizoctonia solani*
Sclerotinia sclerotiorum	*Sclerotium rolfsii*
	Sphaceloma arachidis
	Sphacelia sp.
Bacteria	
Xanthomonas axonopodis pv. phaseoli	*Xanthomonas sp.*
Pseudomonas syringae pv. Phaseolicola	*Pseudomonas fluorescens*
Curtobacterium flaccumfasciens	*Curtobacterium flaccumfasciens*
Viruses	
Potyvirus Group	*Potyvirus Group*
Bean common mosaic virus (BCMV)	*Bean common mosaic virus (BCMV)*
Bean southern mosaic virus (BSMV)	*Bean southern mosaic virus (BSMV)*
Insects	
Acantoscelides sp.	*Acantoscelides sp.*
Zabrotes spp.	*Zabrotes spp.*
Nematodos	
Meloidogyne spp.)	

The Seed Health Testing Methods of CIAT's GHL for the following pests are as follows:

1. Fungi: Two incubation methods are used: blotter test and agar plate. Under the agar plate method, seeds

are treated with 1% sodium hypochlorite for two minutes, plated in petri dishes on a suitable agar media, generally potato dextrose agar with lactic acid 25% and 100 microliters/Lt of media. After plating both procedures include the placing of seeds in culture plates and then incubation under fluorescent light with a 12 hr. cycle light and dark at 20 - 27°C. After eight days of incubation the seeds are examined using a stereobinocular microscope and microscope. The fungi are identified by morphology.

2. Bacteria: The detection and identification of seedborne bacteria at the GHL is accomplished by isolation of bacteria in culture media and their serology. The agar plate dilution technique is the preferred method for this purpose. Seeds are washed thoroughly in running tap water and then they are placed inside a plastic bag with sterilized saline solution. Samples are incubated overnight at +6 °C under dark conditions. Aliquots of the seed leachate are removed and serially diluted 10 fold to 1:100, one, and 100 microliters from each dilution are spread onto the surface of YDC Agar plates. Plates are incubated at 27 °C for three days and then examined for colonies. A semiselective media (MXP) described by Claflin et al. (1987) is used for *Xanthomonas axonopodis pv. phaseoli.* that hydrolyzes starch leading to characteristic halos around the colonies.

Bacterial identification is initially accomplished by using color description and colony morphology and finally with serological reactions with a specific antiserum.

Detection of *P. syringae pv. phaseolicola* has an extraction phase similar to X. a. pv. phaseoli but aliquots are spread onto the surface of King's B medium (Saettler et al., 1989). Plates are incubated at 27°C for three days. The *P. syringae pv. phaseolicola* is identified by presence of the fluorescent pigment and with the serologic agglutination test.

The detection of *Curtobacterium flaccumfasciens pv. flaccumfasciens* (*Corynebacterium flaccumfasciens fsp flaccumfasciens*, also has the same extraction phase. The medium for these bacteria is NBY at 27 °C. Bacteria are identified using color and morphology of the colonies and the gram stain reaction. They are the only gram positive bacteria of quarantine importance.

3. Viruses: The ELISA test is used for the examination of bean and tropical legumes seeds in order to check absence of the seedborne viruses: bean common mosaic virus (BCMV) and bean southern mosaic virus (BSMV). For the detection of common mosaic virus we use the Indirect ELISA with a monoclonal antiserum against the potyvirus group. In order to check the samples against BSMV we use the DAS Elisa Reagent set (Clark et al. 1977).

Results

Beans (*Phaseolus* spp.)

Seed samples of 4,713 accessions of beans were tested, some of them for export to specific countries, and most for conservation in GRU. Their health status showed 3,756 samples without pathogens of quarantine importance (Figure 5).

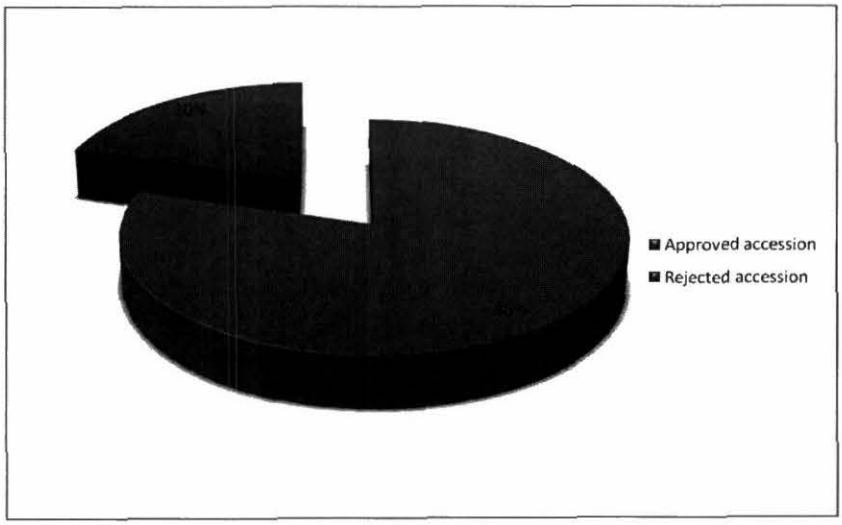

Figure 5. Percent of *Phaseolus* sp. seed samples disapproved or accepted after GHL analysis.

The 20 % of samples of *Phaseolus* sp. showed pathogens of quarantine importance (Table 17, Figure 6).

Seedborne viral infections were detected at very high frequency in this year. The Southern bean mosaic virus (SBMV) was detected in 678 samples and Potyviruses were detected in 291 samples (Table 17).

The seedborne fungi as *Macrophomina phaseolina, Macrophoma* sp, Ascochyta spp., *Botrytis cinerea, Rhizoctonia solani, Colletotrichum gloeosporioides, Colletotrichum lindemuthianum, Colletotrichum truncatum, Phomosis sp.*, and *Sclerotium rolfsii* were detected.

In this year *Pseudomonas syringae pv. phaseolicola* were detected in 60 samples and Xanthomonas axonopodis pv. phaseoli were detected only in one accession.

Virus SBMV was increased at a very high percentage as compared with recent years. Due to this, a specific research will be conducted in order to lower the level of incidence of this virus. Eighty-seven percent of samples have been rejected for viruses, mainly Southern Bean Mosaic virus (Figure 6).

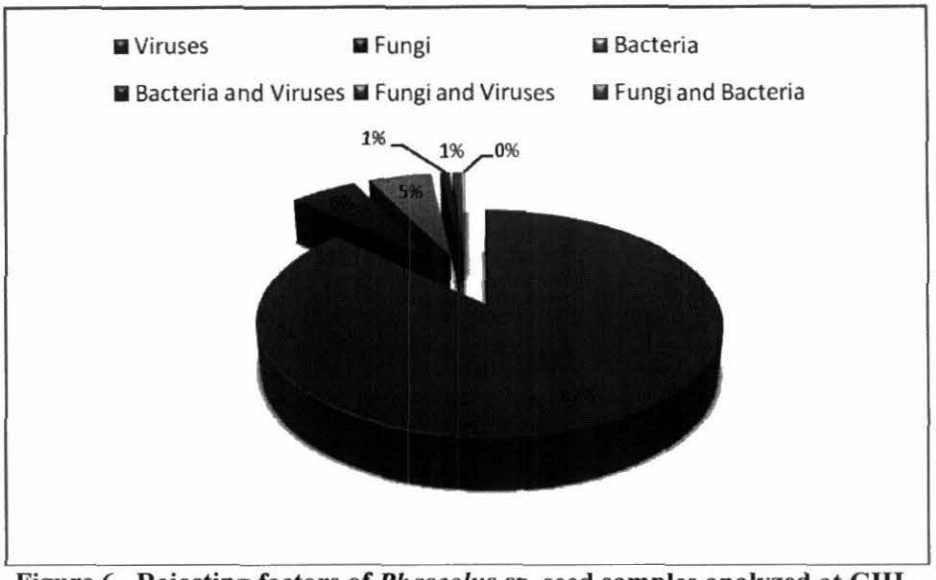

Figure 6. Rejecting factors of *Phaseolus sp.* seed samples analyzed at GHL.

Table 17. Rejecting factors of *Phaseolus* seed samples analyzed at GHL.

Reject Factors	**Affected accessions**	**%**
SBMV	544	56,8
Potyvirus	167	17,5
Potyvirus, SBMV	122	12,7
Pseudomonas syringae pv. Phaseolicola	51	5,3
Macrophomina phaseolina.	31	3,2
Macrophoma sp.	6	0,6
Pseudomonas syringae pv. Phaseolicola, SBMV	6	0,6
Ascochyta spp.	5	0,5
Botrytis cinerea.	4	0,4
Macrophomina phaseolina., SBMV	4	0,4
Rhizoctonia solani.	4	0,4
Colletotrichum gloeosporioides	3	0,3
Pseudomonas syringae pv. Phaseolicola, Potyvirus	2	0,2
Rhizoctonia solani., SBMV	2	0,2
Colletotrichum lindemuthianum, Pseudomonas syringae pv. Phaseolicola.	1	0,1
Colletotrichum lindemuthianum, SBMV	1	0,1
Colletotrichum truncatum, SBMV	1	0,1
Phomosis sp.	1	0,1
Sclerotium rolfsii	1	0,1
Xanthomonas axonopodis pv. phaseoli	1	0,1
Total	**957**	**100,0**

Tropical grasses and legumes

Seed samples of 1,805 accessions of tropical grasses and legumes were tested. Their health status showed 1,315 samples without pathogens of quarantine importance (Figure 7). The 27 % of samples of tropical grasses and legumes showed pathogens of quarantine importance (Table 18, Figure 8).

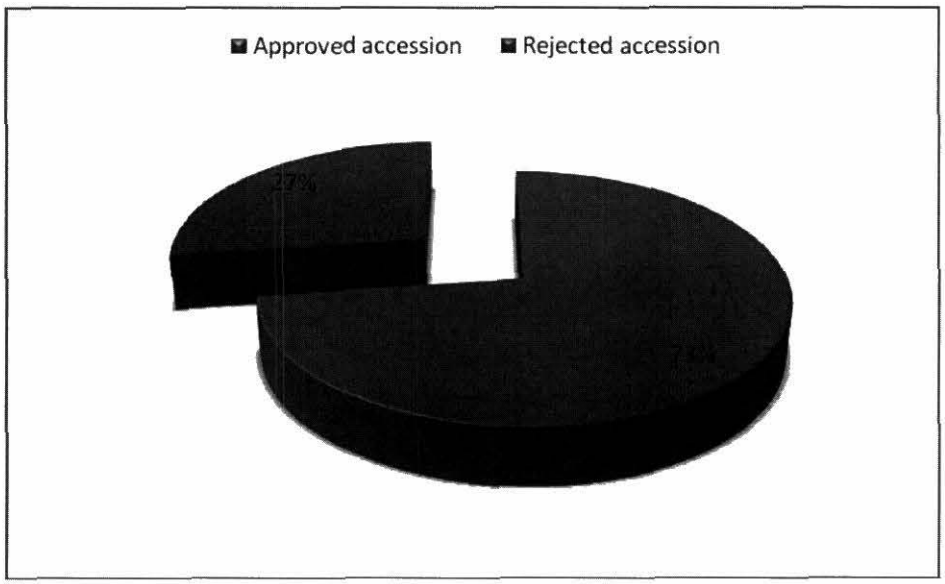

Figure 7. Number of Tropical grasses and legumes seed samples rejected or accepted after analysis by GHL.

In the disapproved samples we detected some seedborne fungi of quarantine importance as *Curvularia sp., Phomosis sp., Phoma exigua var. diversipora, Drecshlera spp., Colletotrichum gloeosporioides, Pestalotia sp., Ascochyta spp., Macrophoma* sp., *Phomosis* and *Colletotrichum truncatum* (Table 18, Figure 8).

The Potyviruses were detected in 38 samples, and Southern bean mosaic virus (SBMV) was detected in 65 samples (Table 18).

Pseudomonas fluorescens were detected in 9 accessions and *Curtobacterium flaccumfasciens* pv. *flaccumfasciens* were detected in 8 accessions. The *Xanthomonas campestris pv. graminis* were not detected.

In tropical grasses, the highest percentage factor of rejecting is for the accessions affected by fungus, in contrast with the bean situation (Figure 9).

Some seed borne fungi are of common occurrence in the regeneration fields of *Brachiaria* ssp. in Santa Rosa Station, and they affect yield and health quality of the germplasm. In some previous studies carried out under field conditions in the station of Santa Rosa (Popayán) we determined the existence of a fungous complex, which principal components were fungi of *Sphacelia* sp., *Drechslera* spp., *Phoma* spp., *Epicoccum* sp. (*Cerebella* sp.), affecting *Brachiaria* spp. seed quality production (Garcia y Pineda 2000; Garcia et al, 2001).

In October 2008, a student started a thesis titled: "Evaluación agronómica y sanitaria de 31 accesiones de Brachiaria en tres localidades y, comparación de métodos alternativos para el control de enfermedades

fúngicas en el germoplasma de Brachiaria brizantha." This thesis is going on to study the agronomic behavior and the health of germplasm of 31 accessions of *Brachiaria* in three localities (Palmira, Santander de Quilichao and Popayán). Simultaneously we study the effectiveness of different methods from biological and chemical control for the main fungi that affect the health of germplasm of *Brachiaria brizantha* through tests in vitro and in field in the locality of Popayán.

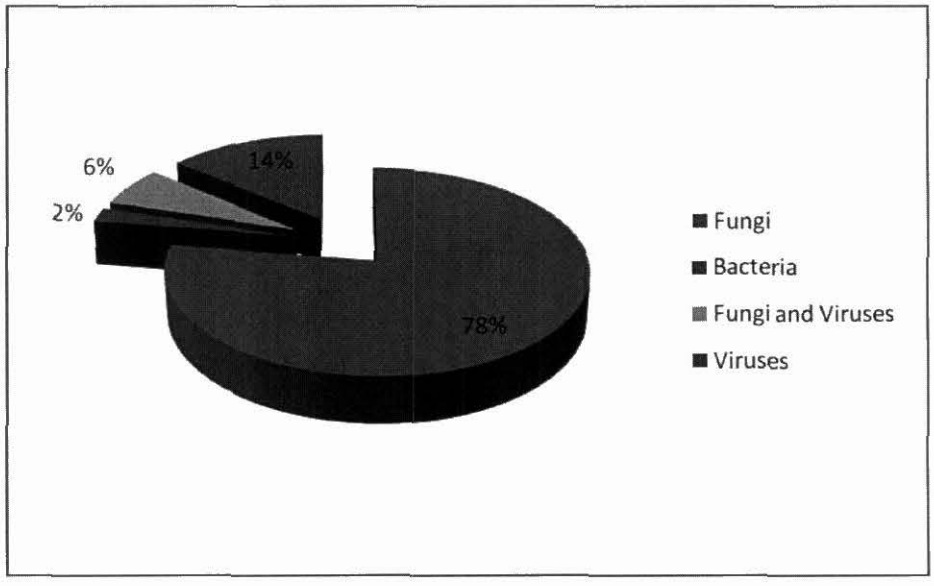

Figure 8. Rejecting factors of Tropical grasses and legumes seed samples analyzed at GHL

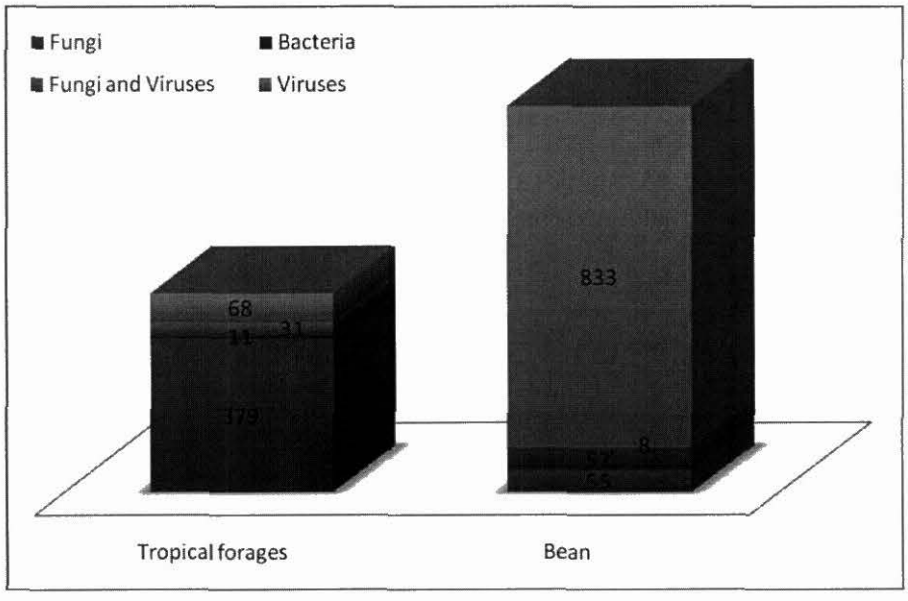

Figure 9. Comparison of rejecting factors between tropical forages and beans.

Table 18. Rejecting factors of legume and tropical grasses seed samples analyzed at GHL.

Reject Factors	Affected accessions	%
Curvularia sp.	62	12.7
Phomosis sp.	44	9.0
SBMV	43	8.8
Phoma exigua var. diversipora	34	6.9
Drechslera sp.	29	5.9
Colletotrichum gloeosporioides	26	5.3
Potyvirus	25	5.1
Pestalotia sp.	24	4.9
Ascochyta spp.	18	3.7
Macrophoma phaseolina	18	3.7
Drechslera sp., *Phoma exigua var. diversipora*	12	2.4
Curvularia sp., *Drechslera* spp.	8	1.6
Curvularia sp., *Phoma exigua var. diversipora*	7	1.4
Curtobacterium flaccumfasciens	6	1.2
Macrophoma sp., *Phomosis* sp.	6	1.2
Pseudomonas fluorescens	5	1.0
Curvularia sp., *Drechslera* spp, *Phoma exigua var. diversipora*	5	1.0
Curvularia sp., *Phomosis* sp.	5	1.0
Pestalotia sp., *Phoma exigua var. diversipora*	5	1.0
Rhizoctonia solani	5	1.0
Drechslera spp., SBMV	5	1.0
Colletotrichum gloeosporioides, *Phomosis* sp.	4	0.8
Pestalotia sp, *Phomosis* sp.	4	0.8
Phoma exigua var. diversipora, *Phomosis* sp.	4	0.8
Colletotrichum gloeosporioides, *Curvularia* sp.	3	0.6
Colletotrichum gloeosporioides, *Macrophoma* sp., *Phomosis* sp.	3	0.6
Curvularia sp., *Macrophoma* sp.	3	0.6
Curvularia sp., *Pestalotia* sp.	3	0.6
Macrophoma sp., *Pestalotia* sp.	3	0.6
Macrophomina phaseolina, *Phomosis* sp.	3	0.6
Colletotrichum gloeosporioides, Potyvirus	3	0.6
Curvularia sp., SBMV	3	0.6
Ascochyta spp., *Phoma exigua var. diversipora*, *Phomopsis* sp.	2	0.4
Ascochyta spp., *Phomosis* sp.	2	0.4
Colletotrichum gloeosporioides, *Macrophoma* sp.	2	0.4
Macrophomina phaseolina, *Pestalotia* sp.	2	0.4
Phomosis sp., *Pseudomonas fluorescens*	2	0.4
Drechslera spp., *Phoma exigua var. diversipora*, SBMV	2	0.4
Macrophomina phaseolina, Potyvirus	2	0.4
Others	48	9.8
Total	490	100.0

Service of germplasm health certification for other CIAT projects

Seed samples of 1,190 accessions from the projects Andean Bean Genetics, Mesoamerican Bean Genetics and Tropical grasses and legumes were analyzed (Figure 10).

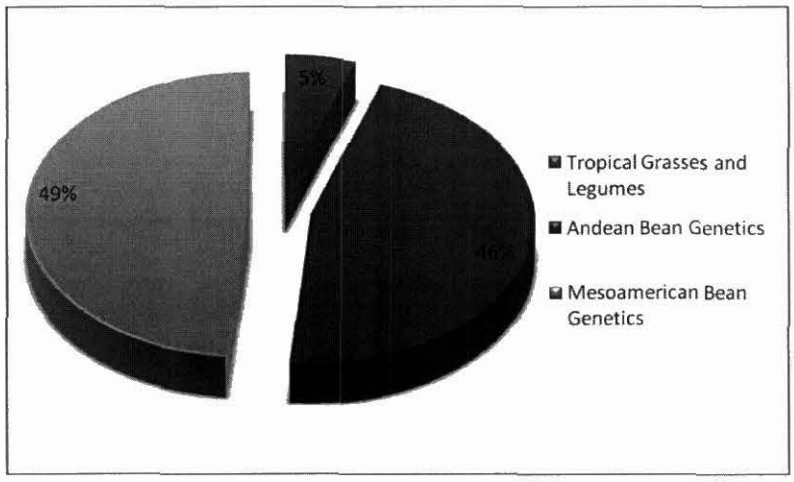

Figure 10. Number of accessions tested for other projects in 2008.

Their health status showed 52.0% samples without pathogens of quarantine importance. The 27 % of samples showed pathogens of quarantine importance (Figure 11).

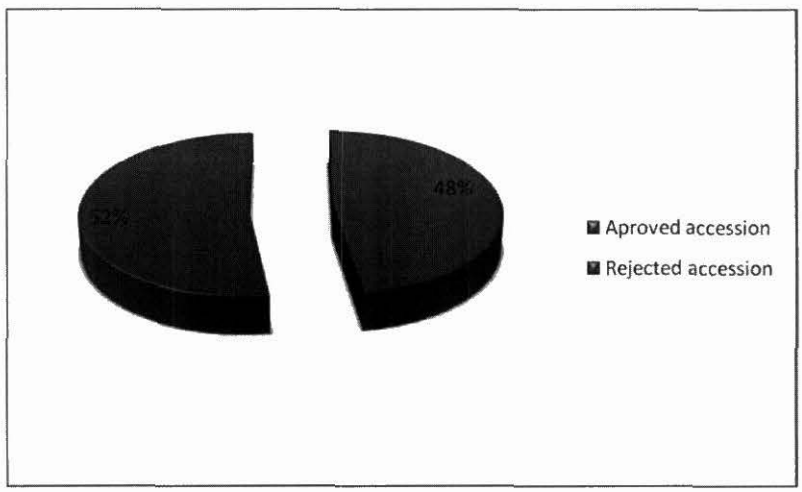

Figure 11. Number of seed samples of other projects rejected or accepted after analysis by GHL.

In rejected samples we detected some seedborne fungi of quarantine importance in 469 accessions (*Macrophomina phaseolina, Colletotrichum gloeospor*ioides, *Rhizoctonia* sp. *Ascochyta* spp., *Dreschslera* spp., *Phoma exigua var. diversipora*).

The Potyviruses were detected in 39 samples and Southern bean mosaic virus (SBMV) was detected in 113 samples (Figure 12, Table 19).

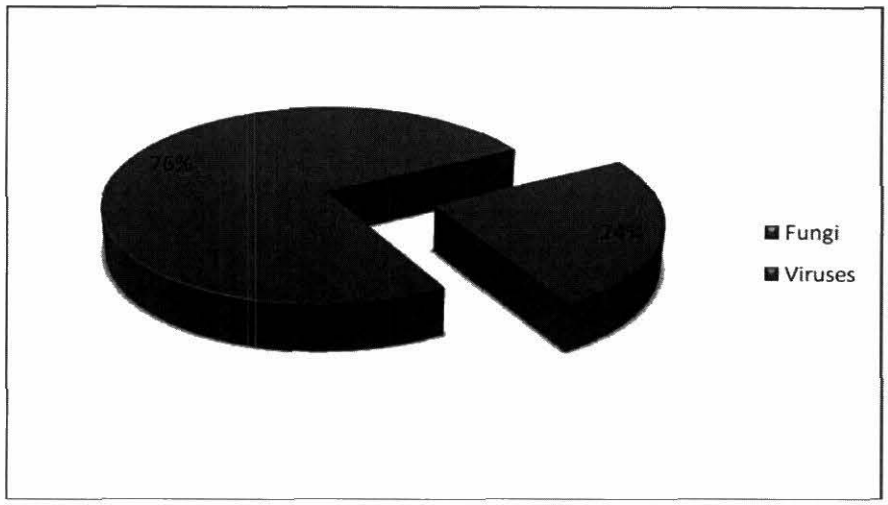

Figure 12. Rejecting factors of other projects seed samples analyzed at GHL.

Table 19. Rejecting factors of other projects seed samples analyzed at GHL

Rejecting factors	Affected accessions	%
SBMV	113	18,2
Macrophomina sp.	449	72,3
Potyvirus	39	6,3
Dreschslera spp., Phoma exigua var. diversipora	4	0,6
Phoma exigua var. diversipora	4	0,6
Rhizoctonia sp.	4	0,6
Ascochyta spp.	3	0,5
Colletotrichum gloeosporioides	2	0,3
Macrophomina sp., *Rhizoctonia* sp.	1	0,2
Macrophomina sp.,*Colletotrichum gloeosporioides*	1	0,2
Curvularia sp.	1	0,2
Total	**621**	**100**

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García, S. X., Pineda, B. 2000. Reconocimiento de enfermedades fungosas transmitidas por semillas en germoplasma de *Brachiaria* spp. Fitopatología Colombiana. 24(2): 39-46.

Contributors: M. Cuervo I., M. del S. Balcázar, J.L. Ramirez.

2.2. Germplasm, passport and characterization data available to users

2.2.1. Support and improve of GRU information system

The information system implemented at the GRU is in constant evolution; therefore it presents additions and periodical changes.

GRU Web page has been updated with digital report explorations in PDF format, and lots of new images have been added to be used with the internal Web portal, supporting information located and helping on a fast identification of the material.

Our next step is to continue adding and updating information completely available for everyone, inside and outside CIAT. We are planning to renovate the GRU portal appearance, making it more attractive and easy to navigate within. Also, we hope to increase the number of descriptors and implement new ways to search data into the web page. We have seen the need to track record of opinions by Web page users and we have developed a survey that permits such a feedback. Table 20 and 21 reports the number of visitors into GRU web page during 2007 and 2008.

Table 20. Report Web Site GRU 2007. (243 days).

	All days	Average per day
Hits	569.243	3.796,17
Page views	390.285	2.253,39
Visitors	2.903	----

Table 21. Report Web Site GRU 2008 (366 days).

	All days	Average per day
Hits	1.015.669	8.333,62
Page views	717.492	5.890,88
Visitors	7.501	----

The internal information system has been updated with new information and reports, as there were new requirements to improve the germplasm flowchart, where the internal user could insert or update the information system. In 2008 the GRU users of internal system were trained in the new web version of Discoverer 4i.

Development and implementation of bar code system at Germplasm Health Laboratory

The implementation of bar coding continues to progress. In 2008 a bar code system for the Germplasm Health Laboratory has been developed. This system allows taking data of bean and forage evaluations (fungi, bacteria and virus) and reduce the risk of error during identification of materials by Staff. The transfer of data

to the central database is also quickly and immediately updated. The introduction of the bar code system into laboratory processes has been gradually, many tests were made to get better results and to optimize the data capturing. Therefore, the training to Staff for using the bar coding devices has been successful. Right now we are working to replace the beta version of the developed software with the version we hope is going to be the final release.

Technical input:

Hardware:

- Handheld PSION WorkAbout PRO
- Printer Zebra TLP2844Z

Software

- Windows CE Operation System
- Java and SWT custom developing
- HSQLDB database
- Synchronization through cradle




We already analyzed seed reception from field, and one of the next year plans is to bring barcode processing here. Also we will continue to implementing bar coding in field, and improve capturing data characterization (growth habit, flower color), and harvest.

Contributor: A.M. Hernández.

2.2. Distribution of germplasm from designate collections to end-users

Achievement: 7,400 accessions of the three commodity FAO designate collections distributed to germplasm users.

As it can be seen in Tables 22 and 23 and Figures 13 to 18, a total of 7,400 accessions were distributed, through 174 requests attended during 2008 for beans, forages and cassava. The main recipients were CGIAR Centers with 3,905 accessions and 3,495 to others institutions. NARS and universities were another important recipients. Pending on recipient type, the main purposes of the requests were: basic research, breeding, and applied research.

Table 22. Distribution of germplasm during 2008 by kind of institution.

	Beans		Forages		Cassava	
Institution type	**Shipments**	**Accessions**	**Shipments**	**Accessions**	**Shipments**	**Accessions**
CGIAR centers	24	2,629	8	103	25	1,173
Commercial companies	6	899	4	20	2	5
Farmers			43	70		
Gene banks						
NARS	5	926	14	55	7	51
NGOs			1	1	2	5
Regional organizations			1	8	2	7
Universities	9	1266	6	75	13	96
Germplasm networks						
Others	1	3			1	11
Total	**45**	**5,720**	77	**332**	**52**	**1,348**

Table 23. Distribution of germplasm during 2008 by purpose .

	Beans		Forages		Cassava	
Purpose	**Shipments**	**Accessions**	**Shipments**	**Accessions**	**Shipments**	**Accessions**
Breeding	1	9			8	65
Agronomy			43	80	11	70
Applied research	22	4,586	32	213	2	21
Basic research	17	859	1	1	30	1,181
Training	4	263	1	38		
Other	1	3			1	11
Total	45	5,720	77	332	52	1,348

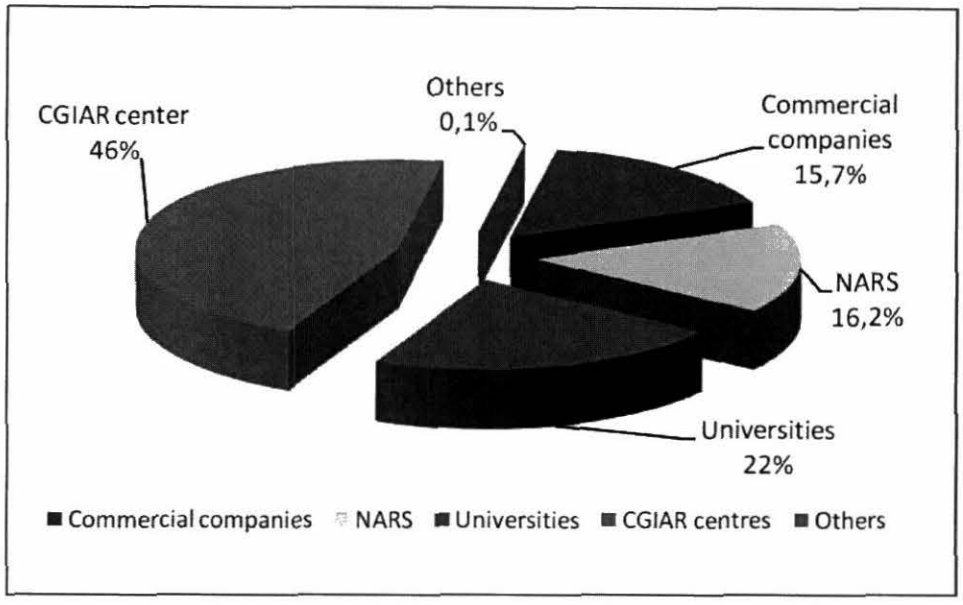

Figure 13. Distribution of bean seed germplasm by kind of users.

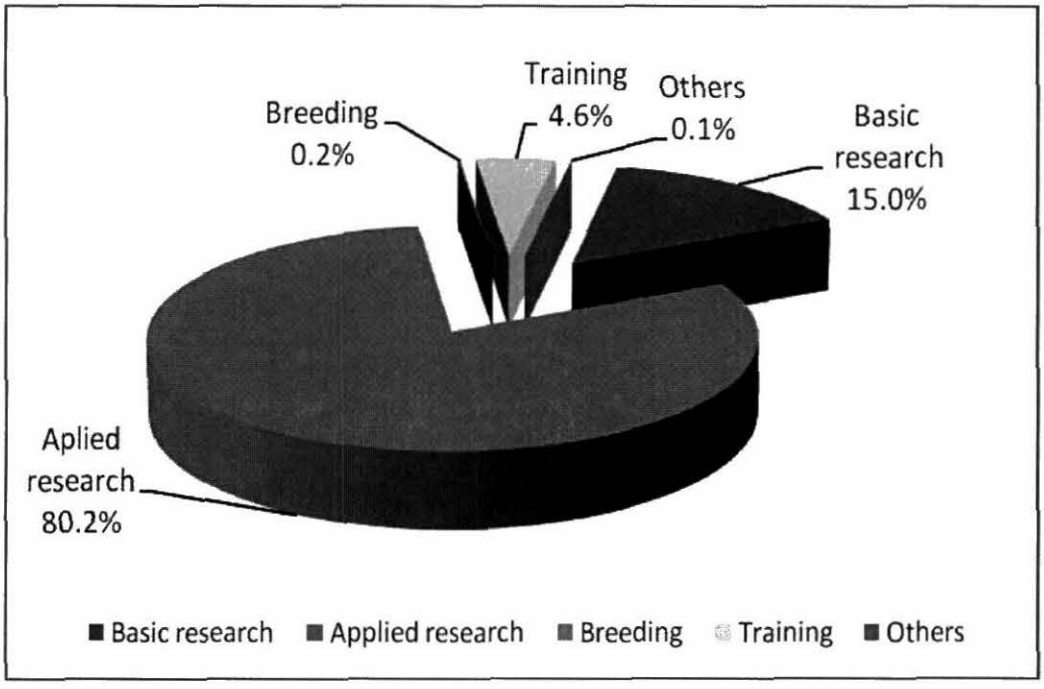

Figure 14. Distribution of bean seed germplasm by purposes.

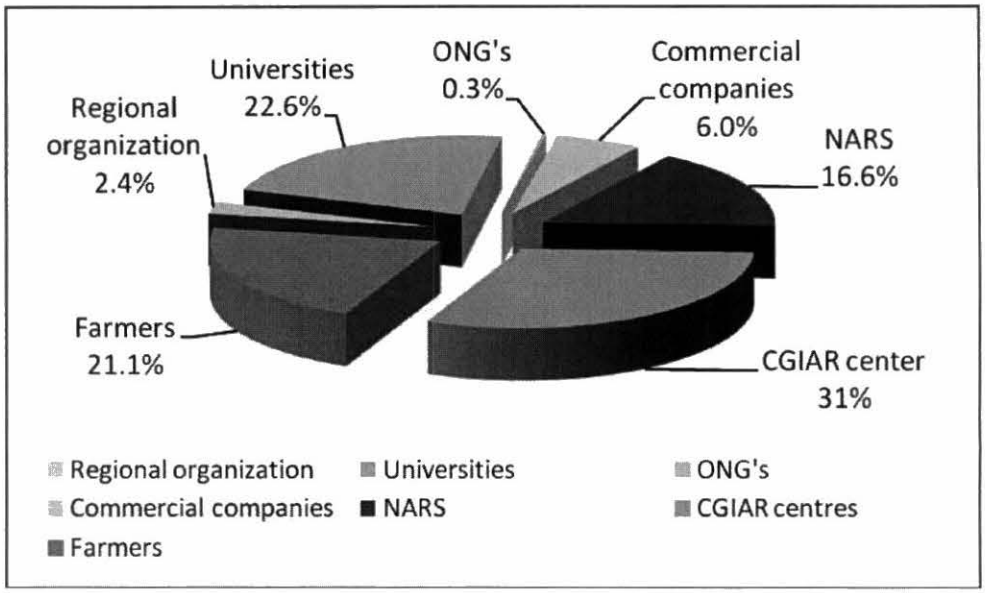

Figure 15. Distribution of forage seed germplasm by kind of users.

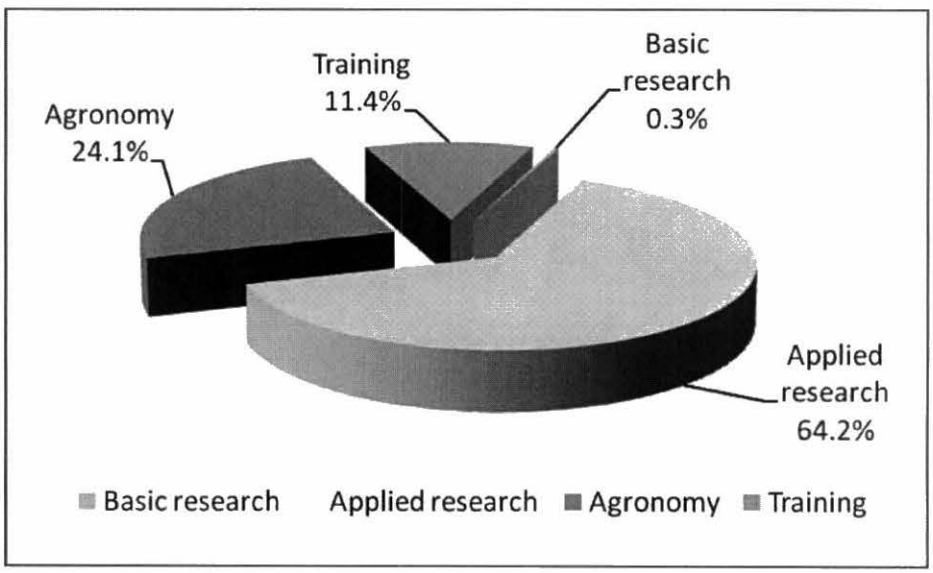

Figure 16. Distribution of forage seed germplasm by purposes.

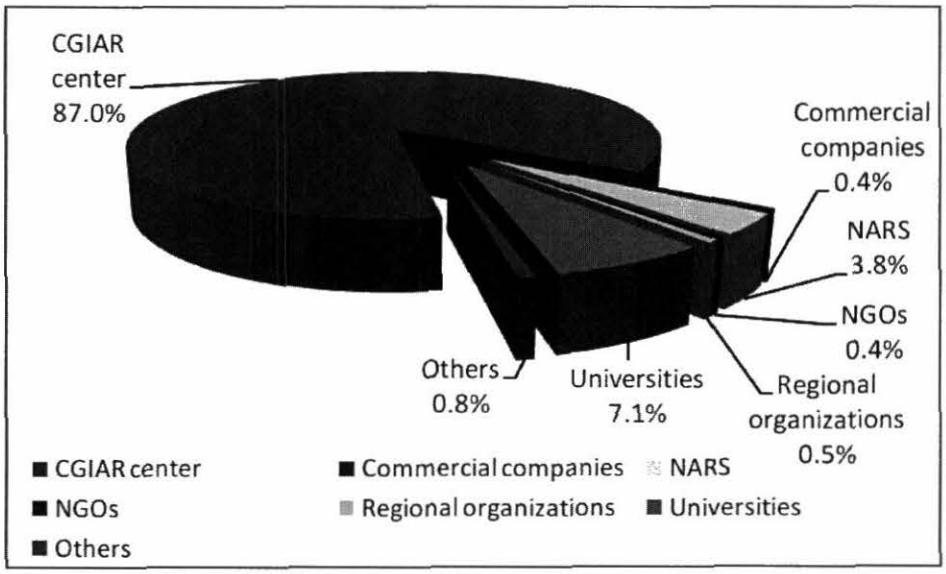

Figure 17. Distribution of *in vitro* cassava germplasm by kind of users.

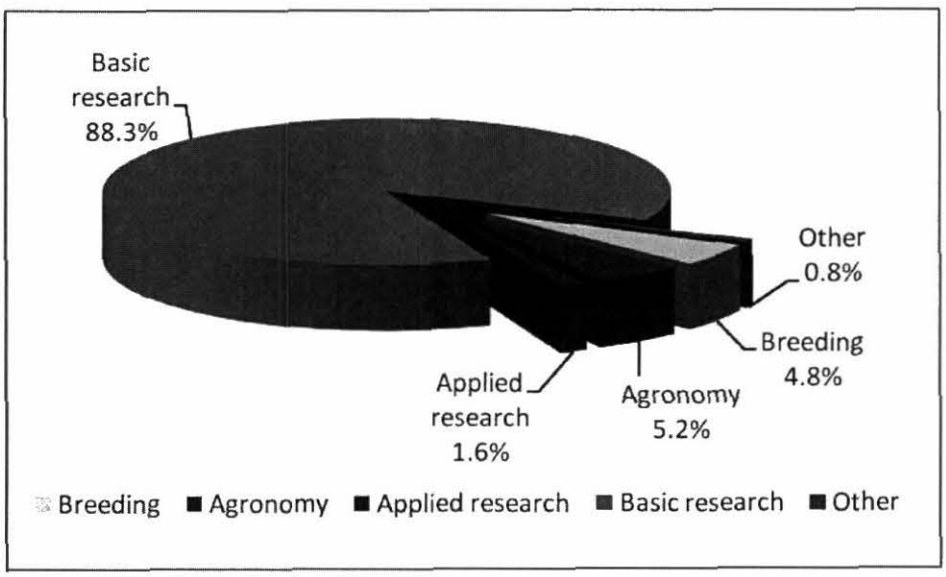

Figure 18. Distribution of *in vitro* cassava germplasm by purposes.

Contributors: C. Lima, G. Mafla, E. Aranzales, A.M. Hernández, D.G. Debouck.

2.3. National collections restored to countries

During 2008, the GRU did not receive any request for the restoration of germplasm collections.

Output 2.4. FAO designated collections safely duplicated

Activity 2.4.1. Shipment of germplasm collections for security backups

In 2008 we have shipped to CIP 2,119 accessions (6,326 tubes) of the *in vitro* cassava collection as part of the safety backup agreement with CIP. As reciprocate service, we have received 3,854 accessions (7,708 tubes) of *in vitro* sweet potato sent by CIP.

Contributors: G. Mafla, E. Aranzales

On December 19, 2007, CIAT made an agreement with the Ministry of Agriculture and Food of Norway to deposit a copy of the seed collections of beans (35,896 accessions to date) and tropical forages (23,140 accessions) into the Svalbard Global Seed Vault. On January 30, 2008, GRU made a shipment of 30,911 seed accessions (21,698 of beans and 9,213 of tropical forages). CIAT was third in the number of accessions deposited among the first twenty depositors on the Opening of the Vault (the first two depositors: IRRI with 70,180, CIMMYT with 57,721, and CIAT with 30,911), on February 26, 2008. The contribution by CIAT was key for filling the CGIAR target of 200,000 accessions at Svalbard for the Opening.

Contributors: M.C. Lima, M. Cuervo, M.S. Balcázar, J. Martínez, D.G. Debouck

Output 2.5. Refined core collections

Activity 2.5.1. Biochemical characterization of *Phaseolus* germplasm bank for improved and refined collections.

In 2008, 830 populations with number G and 1,144 genotypes of *Phaseolus vulgaris* L. were analyzed for diversity of seed storage proteins using ID-SDS-PAGE electrophoresis. Similarly, were analyzed 72 populations with number G and 87 genotypes of *Phaseolus lunatus* L. This analysis together with morphoagronomic characterization is a requisite for improving the representativeness of the designated collection and the refinement of the core collection, and to improve progressively the quality of the characterization data available on the web.

Contributors: C.H. Ocampo, O. Toro.

Product 3: The genetic and social relevance of the conservation

Output 3.1. Designate collections better characterized

In 2008, 3,700 accessions were handled in Popayán, and about 500 in Palmira. All these accessions were morphoagronomically characterized (Figure 19).

Figure 19. Field Characterization - Popayan.

Output 3.2. The image bank as complement information of characterization on CIAT website

The gathering of digital images had continued this year adding 3,503 new images for a total of 28,208, accessed through CIAT web site (Figure 20, Table 24). Since many of the images are taken at time of sowing the seeds, now an image is taken at the time of receiving the seed, and then replace it with one with a new seed that best expresses its phenotype. This "maintenance" to the bank of images involves a large additional effort. Also, according to the morphogenotype expressions in the environment, where accessions are multiplied, all variants considered to be separated, and kept are documented with images and entered to the bank.

Figure 20. Image digital Bank.

Table 24. Report of images available in the Webpage of CIAT-GRU to December 2008.

Species	No. of Images
P. vulgaris	24,161
P. coccineus	865
P. lunatus	1,683
P. acutifolius	345
wild *sensu stricto*	317
Additional vulgaris (Nuñas)	402
Additional vulgaris	128
Other lunatus	40
Other coccineus	85
Other wild forms	33
Other stocks	139
Other (Biochemistry) *	10
TOTAL	**28,208**

Images of all globulins and some isozymes for Tepari beans, which should be displayed for the entire collection. Additionally, there are some images of arcelins.

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Establishment of a Germplasm Genetic Quality Laboratory as an International Standard to be Considered in the *Ex situ* Conservation.

Activity 3.3. Biochemical and molecular fingerprinting of a cassava collection grown in CENICAÑA

Introduction

Introductions of germplasm in recent years have increased the accessions number and the availability of the cassava germplasm collection held at CIAT. So there is a need for the fingerprinting of these uncharacterized accessions. The fingerprinting is both through biochemical-isozymes and molecular markers (Ocampo et al. 1993; Ocampo et al. 1995; CIAT, 2008). For the isozyme fingerprinting, the current techniques for characterization of field grown plants cannot readily be applied to in vitro culture, because the isozyme patterns of field plants do not correspond to their in vitro counterparts. We did biochemical and molecular fingerprinting on 233 accessions assembled at CENICAÑA, maintained as field grown plants.

Materials and Methods

Plant material. Two thousand accessions were planted in separate plot in CENICAÑA (Candelaria-Valle), out of which two hundred and thirty-three uncharacterized accessions were used. Two plants per accession in greenhouse conditions were used.

Isozymes. The methodology for isozyme analysis was the one reported by Ramirez et al. (1987).

Microssatelites (SSR). A set of seven SSR markers, carefully chosen to represent coverage of the cassava genome with moderate to high polymorphism information content (PIC) and robust amplification, were used in this study (Table 25).

Results and Discussion

In 2008 233 uncharacterized accessions were biochemical fingerprinted using the α-esterase isozymes and ß-esterase. These accessions from 15 countries, mainly of Brazil, Colombia, Argentina, Paraguay and CIAT Hybrids. A total of 223 different banding patterns were found among the 233 accessions. A large number of patterns (214) of patterns were represented by only one accession, while among the remaining 9 patterns were found 2 and 3 accessions with the same isozyme pattern (Table 26). The former group, 214 accessions, represents unique genotypes within this fingerprinted field collection.

Table 25. Different primer combinations of primer evaluated in this study. Parameters of number of alleles per locus and polymorphism information content (PIC) are including (Marin et al 2003).

Primer		Alleles per locus	Polymorphism information content (PIC)
SSRY100	498	17	**0.828**
SSRY82	381	11	**0.813**
SSRY106	507	14	**0.784**
SSRY69	313	16	**0.729**
SSRY59	249	17	**0.672**
SSRY105	506	13	**0.538**

The accessions in the latter group may be possible genetic copies, because shows the same isozyme patterns. Table 28 shows the frequency distribution of 22 isozyme alleles in these 233 characterized accessions. No

allele has clear geographic distribution, although many have more or less similar frequencies in many countries. This is because the polymorphis of α-esterase isozymes and ß-esterase in cassava identifies and separates closely related accessions genetically. It is not suitable for separating or grouping geographic populations. If we compare this Frequency (%) of isozyme alleles with those of the cassava worldwide collection (Ocampo et al. 1992), we see that the frequencies for the most frequent alleles are unchanged, as are the alleles 10, 15, 22. Other important alleles, although less frequently have changed little, as are the alleles 1, 2, 11, 12, 14, 16, 17, 18, 19, 20, 21. By contrast, there are some alleles that changed significantly with respect to the reported frequency in 1992 for the whole cassava collection, which reported 4304 fingerprinted accessions. These alleles are 3, 4, 5, 6, 7, 8, 9, and 13. Therefore there are major changes in the frequency for 8 of the 22 isozyme alleles reported for cassava, which is equal to less than 36% of the genetic load used for cassava fingerprinting. It is possible that these changes have happened in the Brazilian collection, which has the largest number of accessions (137) and also where are reported the largest number of accessions with the same isozyme pattern or possible genetic copies

Molecular fingerprinting to identify genetic copies (genetic redundancy). The molecular fingerprinting analysis confirms 19 duplicate accessions, with 10 redundant accessions, which represents 8 % of the biochemical characterized accessions. Additionally, 214 accessions that are uniquely separated using biochemical markers represent unique genotypes. Finally are confirmed 10 potential genetic copies, using the biochemical and molecular analysis and supplemented with passport information (Table 27).

Table 26. Number of cassava accessions from field collection (CENICAÑA) and characterized by isozymes.

Origin	No. of Acc.	Isoenzyme Fingerprinted		
		Distinct Isozyme patterns		
		No. of acc. for each pattern	No. of distinct Isozyme patterns	Total No. of Accessions
Argentina	22	1	16	16
		2	3	6
Brazil	137	1	126	126
		2	4	8
		3	1	3
Colombia	26	1	24	24
		2	1	2
Costa Rica	2	1	2	2
Cuba	1	1	1	1
Ecuador	1	1	1	1
Guatemala	1	1	1	1
Indonesia	1	1	1	1
Malaysia	1	1	1	1
México	6	1	6	6
Panama	2	1	2	2
Paraguay	11	1	11	11
Peru	3	1	3	3
Thailand	2	1	2	2
Venezuela	1	1	1	1
Hybrids CM	11	1	11	11
Hybrids SM	5	1	5	5
TOTAL	233	Total	223	233

Table 27. Description of the biochemical/molecular genetic copies of the field cassava germplasm collection held at CENICAÑA.

No. of Accessions	Genetic Duplicates
2	Arg 29 --- Arg 44
2	Arg 82 --- Arg 83
2	Arg 100 --- Arg 115
2	Bra 965 --- Bra 975
2	Bra 1056 -- Bra 1065
2	Bra 1159 --- Bra 1161
2	Bra 1168 --- Bra 1169
3	Bra 1197 --- Bra 1200 --- Bra 1204
2	Col 2534 --- Col 2632
Total: 19 duplicate accessions, with 10 redundant accessions.	

Table 28. Frequency (%) of isozyme alleles in cassava field collection (CENICAÑA), December 2008.

Origin	Isozyme alleles																						Total of Access.
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	
Argentina	0	0	0	95	32	45	50	0	86	41	0	0	18	0	91	14	0	55	9	9	50	68	22
Brazil	0	7	0	94	15	24	62	0	72	75	0	4	25	0	47	1	0	4	26	27	32	64	137
Colombia	0	8	0	96	35	31	31	0	31	26	0	19	42	0	65	0	0	4	23	38	46	58	26
Costa Rica	0	0	0	50	100	50	100	0	100	100	0	0	50	50	0	0	0	0	50	0	50	50	2
Cuba	0	100	0	100	100	0	0	0	0	100	0	0	100	0	100	0	0	0	0	0	100	100	1
Ecuador	0	0	0	100	0	0	100	0	0	100	0	0	0	0	100	0	0	0	0	0	100	0	1
Guatemala	0	100	0	0	100	0	100	0	100	100	0	0	100	0	100	0	0	0	0	0	100	100	1
Indonesia	0	0	0	100	0	0	100	0	100	100	0	0	0	0	100	0	0	0	100	0	0	100	1
Malaysia	0	0	0	100	0	0	100	0	100	100	0	0	0	0	100	0	0	0	100	0	0	100	1
Mexico	0	0	0	67	33	0	67	0	33	100	0	0	67	0	83	0	0	0	67	33	17	83	6
Panama	0	0	0	100	50	0	100	0	100	100	0	0	50	0	100	0	0	0	0	0	100	50	2
Paraguay	0	9	0	91	0	18	45	0	64	73	0	9	27	0	55	18	9	9	55	36	18	36	11
Peru	0	0	0	100	67	0	33	0	33	100	0	33	33	0	33	0	0	0	0	33	0	67	3
Thailand	0	0	0	100	0	0	50	0	0	100	0	50	50	0	50	0	0	0	0	0	50	50	2
Venezuela	0	100	0	100	100	0	0	0	0	100	0	0	0	0	100	0	0	0	0	100	0	0	1
Hybrids CM	0	18	0	73	45	18	45	0	36	55	0	27	45	0	64	0	0	18	9	18	36	64	11
Hybrids SM	0	20	0	100	80	20	40	0	80	100	0	0	20	0	60	0	0	0	20	0	20	60	5
TOTAL (%)	0	8	0	92	24	25	56	0	64	76	0	7	29	0.4	57	3	0.4	9	25	25	35	62	233

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Activity 3.4. Phaseolin fingerprinting of the *Phaseolus vulgaris* L. collection: distribution of the phaseolin types according to regions and biological status.

Introduction

The genetic diversity for a single trait – phaseolin or seed storage protein – is very informative with respect to the origin, domestication and dissemination patterns of common bean and complements archaeological, historical, linguistic, morphoagronomic and genetic data on this subject. Several characteristics of phaseolin account for this observation: such as polymorphism, environmental stability, and the most important characteristic is the complexity of phaseolin at the molecular level. The latter makes every phaseolin type is probably unique and has appeared only once in the evolutionary history of the common bean (Gepts, 1988). The evidence before making the phaseolin is considered as an evolutionary marker that will always be a need to consider for any study involving the *Phaseolus vulgaris* L. germplasm. Therefore now we propose to develop the phaseolin fingerprinting of the *Phaseolus vulgaris* L. world collection held in the GRU (CIAT) for to know the world distribution of the phaseolin types according to continental regions and its biological status.

Materials and Methods

The accessions, which are reported here, were obtained from the *Phaseolus* world-wide collection held in CIAT. The samples were analyzed in ID-SDS-PAGE (Brown et al. 1981) and confirmed later in 2D-IEF-SDS-PAGE (O'Farrel, 1975).

Results and Discussion

Gepts in 1988 use the phaseolin as an evolutionary marker to follow the domestication and dissemination patterns of the common bean both for the Americas, as other the world continents. However, Gepts only analyze 668 accessions from different bean-growing regions of the world, not including other continents such as Asia and Oceania. Furthermore only present results for 6 phaseolin types and not include all common bean biological status. Now we are presenting results of 62 phaseolin types for 6065 accessions from *Phaseolus vulgaris* L world wide collection held at CIAT, which include the different bean-growing regions of both primary centers and secondary across the whole world (Table 29). In general our results show an extensive phaseolin variability from Mexico to Southern Andes and multiple domestications have occurred along this extended distribution range, as can be inferred from the phaseolin data. In contrast for secondary centers there is a phaseolin reduced variability and restricted to the cultivated biological status, noting the phaseolin greater variability in Europe and Africa. For the Mesoamerican cultivars there is a strong phaseolin bottlenecks with respect to its wild ancestor, because 76 % of analyzed genotypes only have three phaseolin types (S, Sb and Sd). Change in Mesoamerican wild has 34 phaseolin types. The Mesoamerican common beans (wild, weedy and cultivated) exhibited the novel 'Tel' and 'Dur' types (Ocampo and Toro, 2005), which reinforces the genes flow in Mesoamerica (Gonzalez et al. 2003).

The distribution of the different phaseolin types in Colombia provides evidence of a meeting zone of the two largest American gene pools This meeting area can be found in different biological status present in the Colombian common bean. In Colombia, there are phaseolins unique to this country, Such as L, CAR, Mu, Qui, LI, HE, TI1, TI2 phaseolins, which reinforces the theory that proposes this country as a new domestication center and genetic diversity of common bean (Chacón et al. 1999). However, we must clarify that some of the Colombian phaseolins are presented in Andean gene pools, such as CAR, L, and Qui, but are present in cultivated materials, possibly brought from Colombia. In Colombia, the phaseolin distribution in all phases of the biological status, suggests a strong genes flow, much higher than that observed in other american gene pools (González et al. 2003). Contrary to what happens in the Mesoamerican genepool, for Andean gene pool, there is a smaller phaseolin bottlenecks with respect to its wild ancestor. In general there was greater phaseolin variability in the southern Andes that in the Northern Andes. Contrary to what is happening in Colombia, the phaseolin data suggests that exchanges between Middle America and Andes gene pools have been limited primarily to cultivated materials. In the secondary centers is observed the predominance of Mesoamerican phaseolins in South American lowlands (Venezuela and Brazil) and North American (USA). The Mesoamerican phaseolin also has predominance on the continents of Asia and Oceania. In contrast, the predominance Andean phaseolins in Europa and Africa is consistent with the Andean origin of the Europeans cultivars and Africans (Gepts and Bliss, 1988). In conclusión, this complete screening of phaseolin variability (as recommended Gepts in 1988) was necessary to have an overall view of the reality of this important evolutionary marker for the common bean germplasm. Really confirms that many of the events already known for the common bean diversity, but it also gives us new contributions to knowledge of the common bean, as is the case of the Colombian common bean diversity

Table 29. Geographical distribution of phaseolin types among accessions from the *Phaseolus vulgaris* L. world collection held in the GRU (CIAT).

Region[a]	n[c]	Biological Status/ Phaseolin type[b]				
		Wild	Weedy	Escape	Hybrid[d]	Cultivated
American Genepools: (Primary Center)						
Middle America	1203	CH(13), Dur(2), S(89), S/M (2), M,M1-M26 (270), Sb(8), Sd(6), Tel(3)	CH(2), Dur(6), C(1), H1(2),M1-M26 (41), S(41),Sb(1),Tel(2)	S(1)		B(24), C(3), CH(77), M(44), M1(4), M13(5), M15(3), M18(1), M20(3), M4(5), S(480), Sb(141), Sd(70), T(43), Tel(7)
Colombia	1583	B(103), C(16), Ca(1), CH(121), H(3),H1(2), H2(2), L (14), M1(1), M11(11),M13(1), M16(2), Mu(2), S(10), T(6)	B(60),C(38),Ca(2), CAR(4),CH(42),H(4),H1(6),H2(2),L(12),M11(12),M15(1), M6(3),Mu(6),S(34), T(19),Tel(2),To1(1)	B(1),C(6), CAR(1),CH(4),H1(1),H2(1), L(4),S(3),T(5), Tel(1),To1(1)	B(1),Ca(2), CAR(2), CH(2),L(2), M11(11), T(1),Tel(1)	B(214), LI(2), C(202), Ca(9), Ca1(21), CAR(112), CH(80), H(51), H1(118), H2(24), HE(2), L (34), LI(3), M11(3), M6(1),Mu(3),P1(1),Qui(1), S(301),Sb(5),Sd(5),T(440), Tel(3), TI1(1), TI2(1), To1(27)
Northern Andes[e]	537	C(1),I(17),T(2)	C(1),I(3),T(5)	S(1),T(2)		B(14),C(89),Ca(14),CAR(1), CH(7),H(77),H1(12),H2 (3), L(7),Nu(2),Pa(10),S(14), T(289),To1(9),Tel(2)
Southern Andes[f]	821	A(2),B(2),C(14),Ca(5),CH(1),H(6),H1(6),H2(9),I(1),J(1),J1(1), J2(7), J3(3),J4(8),K(2),P1(2),Pa(4), S(1),T(78),Ta(3),To1(5)	C(12),Ca(8),CH(1), H(9),H1(5),H2 (2), M6(1),Pa(3),S(2), T(39),To1(7)	C(18),Ca(1), CH(1),H(13), H1(3),H2(1), Pa(1),S(3), T(42),To1(1)	C(1), H2(1)	A(6),A1(2),B(12),C(56), Ca(24),CH(1),H(59),H1(3), H2(3),Ko(11),L(2),M21(1), Nu(2),Pa(18),Qui(1),Tel(1), S(67),T(388),To1(18),To2(2)

[a] The different regions are listed in north-south direction for both the Americas and other world continents.
[b] Values in parentheses represent phaseolin genotypes number in each region according to their biological status.
[c] Total number of accessions with number G, tested in each region.
[d] These are interspecific hybrids.
[e] The Northern Andes genepool includes Ecuador and Northern Peru (the provinces of Tumbes, Piura, Lambayeque, Cajamarca, Amozonas, Sane Martin, Loreto and La Libertad).
[f] The Southern Andes genepool includes the rest of Peru, plus Bolivia and Argentina.

Table 29. (Continuation). Geographical distribution of phaseolin types among accessions from the *Phaseolus vulgaris* L. world collection held in the GRU (CIAT).

Region[a]	n[c]	Biological Status/ Phaseolin type[b]				
		Wild	Weedy	Escape	Hybrid[d]	Cultivated
Secondary centers in the Americas						
Canada	5					B(1),C(1),S(1),T(2)
United States	152					B(14),C(10),S(95),Sb(6),Sd(1),T(29),Tel(2)
Caribbean	159					B(9),CH(1),S(66),Sb(3),Sd(3),T(77)
Venezuela	33					B(1),S(32)
Brazil	152					B(7),C(1),CH(3),S(91),Sb(8),T(46)
Chile	382					C(100),H1(28),S(185),Sb(4),Sd(3),T(76),
Secondary centers in other continents						
Europe	544					B(16),C(127),CH(1),H(17),H1(5),K(2),M(1),M15(1),S(100),Sb(5), Sd(4),T(284),To1(1)
Asia	186					B(7),C(19),CH(3),H(6),Pa(1),S(110),Sb(5),T(39)
Africa	304					B(22),C(14),H(6),H1(4),M20(1),Pa(1),S(65),Sb(8),Sd(5),T(183),Tel(2), To1(1)
Oceania	4					S(2),Sb(1),T(1)

[a] The different regions are listed in north-south direction for both the Americas and other world continents.
[b] Values in parentheses represent phaseolin genotypes number in each region according to their biological status.
[c] Total number of accessions with G number, tested in each region.
[d] These are interspecific hybrids.
[e] The Northern Andes genepool includes Ecuador and Northern Peru (the provinces of Tumbes, Piura, Lambayeque, Cajamarca, Amozonas, San Martin, Loreto and La Libertad).
[f] The Southern Andes genepool includes the rest of Peru, plus Bolivia and Argentina.

Table 30. Types of seed proteins from the collection designated to FAO characterized until January 2009.

Seed Protein	Taxon	Total Accessions
Phaseolins	*P. vulgaris*	6,993
Globulins	*P. acutifolius*	339
Lectins	*P. lunatus*	977
Total proteins	Other spp.	126
		8,435

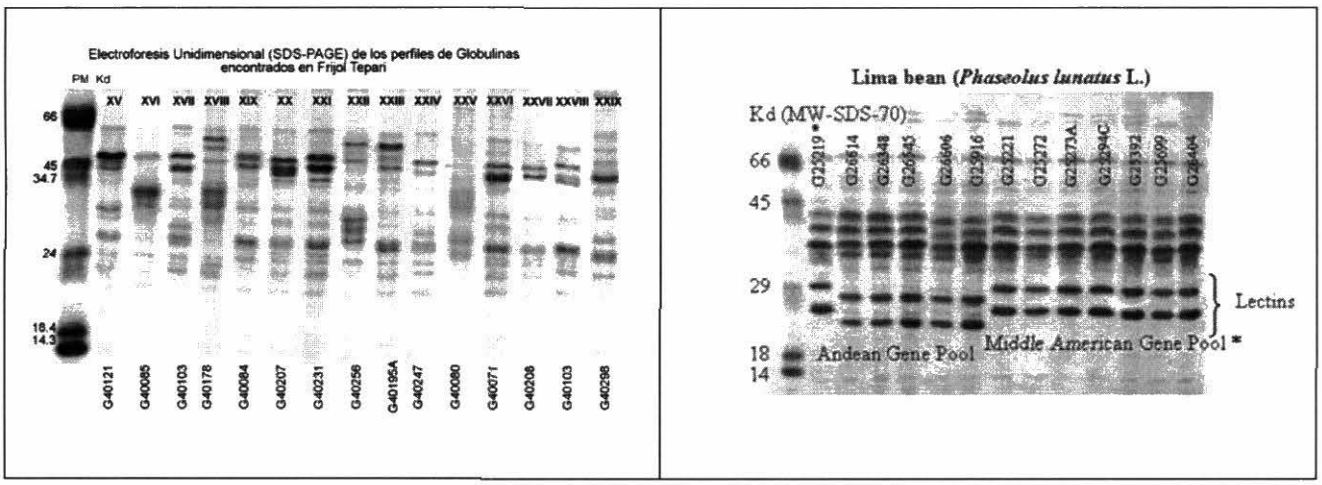

Figure 21. Examples of biochemical characterization for Globulins and Lectins.

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Output 3.4. Genetic erosion monitored

Activity 3.3.1. Monitoring practices to limit genetic drift and erosion in cultivated bean accessions: the case of the Chilean common bean collection

Introduction

Once a new accession arrives at CIAT genebank, its original seed is multiplied in a quarantine greenhouse, and the next generation in a field. During this process, a serious genetic drift (i.e., change of gene frequency) could occur. Using the wild bean model, the GRU has already gained experience and knowledge to limit the genetic drift and erosion, so that this information will help make decisions for the management and maintenance of the genetic diversity present in the genebank. (Guzmán et al. 2001). However, the genetic drift in cultivated bean germplasm collections that have enough time in *ex situ* conservation has so far has not been monitored. The Chilean common bean collection offered us some insight on this question, since most of the accessions of the Chilean collection have original seed. The analysis began with the phaseolin, because this is a marker that can work with old seed or even non-viable seed, something that could not be done with isozyme markers.

Materials and Methods

A total of 213 accessions from the Chilean common bean collection with original seed have been studied, with a range of 143 to 3 original seeds. For the phaseolin analysis, one seed of each accession was used, both for original seed and multiplied. However, for some accessions where there is phaseolin heterogeneity as reported by Paredes and Gepts (1995) and GRU-CIAT (2008), we have used more than one seed.

The samples were analyzed in ID-SDS-PAGE (Brown et al. 1981) and confirmed later in 2D-IEF-SDS-PAGE (O'Farrel 1975). To seed multiplied, the phaseolin be done with seed destructive testing for proteins extraction. In contrast to the original seed will be using seed nondestructive test of seed for proteins extraction, Later these seeds were planted in greenhouse, which made possible to conserve its genotype and to guarantee repeatability of the results obtained with this study.

Results and Discussion

Six phaseolin types were found both for original and multiplied seed, 'S', 'C', 'T', 'H_1', 'Sd' and 'Sb', in decreasing order of frequency for both seed types (Table 31). Our results show that there was no change in the phaseolin pattern between the original and multiplied seed for 213 accessions. This represents 61 % of the Chilean common bean collection held at CIAT. Three accessions (G1088, G1505 and G5326) were heterogeneous for the original seed phaseolin type; however, there was no phaseolin difference between the original and multiplied seed (Table 32).

Table 31. Comparing the phaseolin variability between the original seed and multiplied from accessions of the Chilean common bean collection held in CIAT.

Seed type	n[b] (%)[c]	Phaseolin type[a]							
		Middle America				Andes			
		S	Sb	Sd	Total	C	T	H1	Total
Original seed	213 (100 %)	88 (41 %)	0	1 (0.5 %)	89 (41.5 %)	65 (30.5 %)	46 (22 %)	13 (6 %)	124 (58.5 %)
Multiplied seed	213 (100 %)	88 (41 %)	0	1 (0.5 %)	89 (41.5 %)	65 (30.5 %)	46 (22 %)	13 (6 %)	124 (58.5 %)

[a] The numeric values and in parentheses represent the number and percentages of phaseolin types of each seed type according to their gene pool.
[b] Total number of accessions with number G, tested in each seed type.
[c] The percentages in parentheses added 100 for each seed type.

Table 32. Comparing the phaseolin type between original seed and multiplied in a sample of 81 accessions of the Chilean common bean collection held in the GRU (CIAT).

Accession No. G	Phaseolin Type		Accession	Phaseolin Type		Accession	Phaseolin Type	
	Original Seed	Multiplied seed		Original Seed	Multiplied seed		Original Seed	Multiplied seed
97	S	S	5790	T	T	98	C	C
1063	S	S	5794	S	S	921	S	S
1065	S	S	5795	T	T	1509	Sd	Sd
1073	H1	H1	5799	C	C	1064	S	S
1078	C	C	5800	C	C	1066	S	S
1084	T	T	5807	C	C	1067	C	C
1086	C	C	5820	C	C	1068	C	C
1092	C	C	5822	S	S	1072	C	C
1094	C	C	5827	S	S	1075	C	C
1096	C	C	5832	C	H1	1076	S	S
1102	C	C	5839	H1	H1	1077	S	S
1105	C	C	5846	C	H1	1082	S	S
1106	S	S	5848	C	C	1085	S	S
1112	T	T	5849	H1	H1	1088	C, T, S	S
1113	C	C	5856	T	T	1095	C	C
1159	S	S	5858	T	T	1100	S	S
1529	S	S	5865	C	H1	1101	T	T
1533	S	S	5868	T	T	1103	T	T
1539	C	C	5873	S	S	1104	C	C
3758	S	S	5875	S	S	1107	T	T
4474	S	S	5877	C	C	1109	S	S
4844	S	S	5881	T	T	1503	T	T
5781	H1	C	5883	H1	H1	1504	S	S
5783	S	S	5886	H1	H1	1505	S, T, C	T
5784	H1	H1	5910	H1	H1	1506	S	S
5785	H1	H1	7161	S	S	1511	S	S
5789	T	T	96	C	C	5326	S, C	C

References

Guzmán F.A., O.Toro, C. Ocampo, I. Sánchez, H. Cárdenas & D.G. Debouck. 2001. Observation about risks of genetic erosion and drift during multiplication and regeneration of germplasm, using wild common bean as model. Annu. Rept. Bean Improvement Coop. (USA) 44: 29-30.

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O'Farrel, P. H. 1975. High resolution two-dimensional electrophoresis of proteins. The Journal of Biological Chemistry. 250 (10): 4007-4021.

Paredes, O. M. & Gepts, P. 1995. Extensive introgression of Middle American germplasm into Chilean common bean cultivars. Genet. Res. Crop Evol. 42:29-41.

GRU-CIAT. 2008. Databases for *Phaseolus* world-wide collection held in CIAT.

Contributors: C.H. Ocampo, O. Toro.

Activity 3.3.2. Understanding the structure of genepools in Lima beans

DNA sequencing on two cpDNA intergenic spacers (*trn*L-*trn*F and *atp*B-*rbc*L) and the ITS/5.8S region was performed to investigate the genetic relationships among wild and domesticated Lima beans. The Neighbor-Joining topologies built on the bases of cpDNA and ITS/5.8S polymorphisms reveal three gene pools in this crop, while only two have long been recognized by many authors. One gene pool called 'AI' with wild forms from SW Ecuador and NW Peru and large-seeded cultivated forms from the same region, confirms the domestication of the latter from the former. A second gene pool called 'MI' includes mostly wild forms from Mexico west of Tehuantepec and small-seeded domesticated forms from the same region and from Central America and South America, evidencing a possible domestication west of Tehuantepec in Mexico. A third gene pool called 'MII' includes mostly wild forms from Mexico east of Tehuantepec, different countries of Central America, Cuba, Colombia, eastern Peru, and Argentina, and two small-seeded domesticated accessions from Brazil, opening up the possibility of a third domestication event, the precise location of which now depends on further exploration of this gene pool.

Contributors: M.I. Chacón , J.R. Motta-Aldana, M.L. Serrano-Serrano, D.G. Debouck.

Product 4. Strengthened institutions

Output 4.1. Handbook of GRU procedures

We have already put on the GRU web site the handbook of procedures for in vitro and for germplasm health. The documents on best practices for seed regeneration and conservation are in the writing phase.

Output 4.2. Training/ courses

No course could be carried out in 2008 because of lack of funding, but 17 professionals from different countries of Latin America, Nigeria and Spain were given an individual training.

Product 5. Improved link *ex situ/in situ*

Output 5.1. Contribution made towards protected areas in Latin America: databases about distribution of wild relatives of crops

We have continued with the establishment of databases about the geographic distribution of wild relatives for the so-called CIAT mandate crops. The objectives of that work are:

i. correct identification of materials collected and kept in *ex situ* conservation facilities (namely CIAT genebank, and other collaborating institutions). An output of this work is the taking of digital images of vouchers and to make them available through our web site (a service acclaimed by the Botanical Society of Colombia).

ii. geographic distribution of wild relatives of direct interest in breeding activities (namely acquisition of germplasm useful to the breeders).

iii. distribution of wild relatives genetically compatible with the crop, in view of introduction and management of transgenical crops.

iv. monitoring of modification/ destruction of natural habitats and disappearance of populations.

In 2008 we have collated information in the following herbaria: CAS, CS, DAV, FI, IBUG, IEB, O and TEX. That information has been 'repatriated' to CONABIO of Mexico and INBio of Costa Rica. The information 'Cahiers de Phaséologie' has been put on CIAT web site for the sections: *Acutifolii, Chiapasana, Coriacei, glabellus, microcarpus*, *Minkelersia*, *Revoluti*, and *Rugosi*.

Contributor: D.G. Debouck

6. Annexes

6.1. List of publications by Project Staff in 2008

A. In refereed journals:

Calvert, L.A., **M. Cuervo**, I. Lozano, N. Villareal & J. Arroyave. 2008. Identification of three strains of a virus associated with cassava plants affected by frogskin disease. J. Phytopathology **156**: 647-653.

Chacón, J., S. Madriñán, **D.G. Debouck**, F. Rodríguez & J. Tohme. 2008. Phylogenetic patterns in the genus *Manihot* (Euphorbiaceae) inferred from analyses of nuclear and chloroplast regions. Molec. Phylogen. Evol. **49** (1): 260-267.

Gaiji, S. & **D.G. Debouck.** 2008. Flujos de germoplasma en las Américas: 30 años de distribución de muestras de fríjol por parte del Centro Internacional de Agricultura Tropical. Recursos Naturales y Ambiente **53:** 54-61.

Montoya, C.A., P. Leterme, N.F. Victoria, **O. Toro**, W.B. Souffrant, S. Beebe & J.-P. Lallès. 2008. Susceptibility of phaseolin to *in vitro* proteolysis is highly variable across common bean varieties (*Phaseolus vulgaris*). J. Agric. Food Chem. **56:** 2183-2191.

Reichel, H., **Cuervo, M.** & F. Morales. 2008. Caracterización parcial de un potexvirus aislado de *Musa coccinea* afectada por rayado neurótico en Colombia. Agronomía Colombiana **26** (2): 285-291.

B. In non-refereed journals:

D.G. Debouck, R. Herrera T. & R. Araya V. 2008. New populations of wild common bean disclosed in Nicaragua. Annu. Rept. Bean Improvement Coop. (USA) **51:** 120-121.

Ocampo, C. H. & O. Toro. 2008. Phaseolin diversity of Nicaraguan common bean germplasm held at CIAT. Annu. Rept. Bean Improvement Coop. (USA) **51:** 122-123

6.2. List of thesis research supervised by Project Staff in 2008

César Augusto Medina. Pregraduate thesis student: "Evaluación agronómica y sanitaria de 31 accesiones de Brachiaria en tres localidades y, comparación de métodos alternativos para el control de enfermedades fúngicas en el germoplasma de Brachiaria brizantha."

6.3. List of conferences and scientific communications presented by Project Staff in 2008

Aranzales, E. Lima, Perú, 22-23 November 2008, invited REDARFIT meeting: "Conservación *in vitro* de germoplasma del género *Manihot.*"

Aranzales, E. Palmira, Colombia, 30 October 2008, invited Universidad Nacional de Colombia: "Aplicaciones prácticas de las técnicas de cultivo de tejidos vegetales."

Cuervo, M., Lozano, I., and Morales, F. Palmira, Colombia, 26 November 2008, invited internal seminal in CIAT: "Historia, caracterización y manejo de la enfermedad viral más importante de la especie *Manihot esculenta* en su centro de origen."

Debouck, D.G. Palmira, Colombia, 3 December 2008, invited internal seminar in CIAT: "1,2,3: a new strategic look at a great crop."

Debouck, D.G. Davis, California, USA, 15 September 2008, invited conference at the Harlan II Symposium on the domestication of crop plants and domestic animals: "Domestication of Lima beans: a new look at an old problem."

Debouck, D.G. Celaya, México, 22 May 2008, invited conference at the 1st international bean congress: "Recursos genéticos y patentes: el caso del fríjol común."

Debouck, D.G. Quito, Ecuador, 9 May 2008, invited seminar at the regional workshop of IUCN on certificate of origin: "Perspectivas desde un Centro Internacional sobre la Trazabilidad."

Debouck, D.G. Lima, Perú, 6 May 2008, invited conference at the 13th Latinamerican Congress of Genetics: "Experiencias con los Recursos Fitogenéticos de Fríjol y Yuca."

Debouck, D.G. Palmira, Colombia, 7 March 2008, invited internal seminar in CIAT: "Implications of the International Treaty on Plant Genetic Resources for Food and Agriculture for the work of the Center."

Toro, J.O. 2008. Banco Mundial de *Phaseolus* spp. L., CIAT – URG, Aspectos Relevantes en Regeneración. Presentación en Power point (PDF), REDARFIT 22-23 de Noviembre, Lima, Perú.

6.4. List of trainees trained by Project Staff in 2008

In Seed Conservation

Ing. Napoleón Edgardo Paz Quevedo, Profesor, Universidad de El Salvador, February 19.

Ing. Ricardo Augusto Luna M., Samir Antonio Zambrano M., Universidad Técnica de Quevedo, Ecuador, September 16.

In Health Testing

Dr. Nnamdi Eke-Okoro, Shuaibo Suleiman Khaya, NARs scientists, Nigeria, February 4-8.

Teodulo Batista, Magdalena Poveda, Jose Caballero, MIDA Panamá, July 4.

Ana Cristina Bolaños, Universidad del Valle, Cali, Colombia, August 25-29.

In vitro Lab

Nnamdi Eke-Okoro, Shuaibo Suleiman Khaya. Subculturing in vitro, distribution in vitro, conservation under slow growth. NARs scientists, Nigeria, Feb. 4-8.

Vacca, Orlando. Training in meristem culture. CIAT, Colombia. 1- 2 August 2008.

Bolaños, Ana Cristina. Training in conservation and management of in vitro cassava germplasm. Universidad del Valle, Colombia, 25-29 August 2008.

Toro, Esteban. Training in conservation and management of *in vitro* cassava germplasm. CASD, Colombia. 29 August 2008.

Vargas, Carlos. Training in conservation and management of *in vitro* cassava germplasm. CASD, Colombia. 29 August 2008.

Aguirre, Angie. Training in conservation and management of *in vitro* cassava germplasm. CASD, Colombia. 29 August 2008.

Fernandez, Pedro. Training in conservation and management of *in vitro* cassava germplasm. CASD, Colombia. 29 August 2008.

Perez, Elena. Training in conservation and management of *in vitro* cassava germplasm. Servicio Regional de Investigación y Desarrollo Agroalimentario, SERIDA, Asturias, España. 12 September 10/November 11 2008.

Genetic Quality Lab

Lucy Díaz, Alvaro Soler Garzón. Training in 1D-SDS-PAGE technique for phaseolin. CIAT Bean Project, Palmira, Colombia. 18-29 February 2008.

Ana Cristina Bolaños. Training in Biochemical and Molecular Markers. Universidad del Valle, Cali, Colombia, 28 August 2008.

Elena Pérez Vega. Training in conservation and management of *Phaseolus* germplasm, including Training in Biochemical and Molecular Markers to develop a *joint* research project between the CIAT and SERIDA. Servicio Regional de Investigación y Desarrollo Agroalimentario, SERIDA, Asturias, España. 12 September 10/November 11 2008.

6.5. Posters

Balcázar, M.S.; Rivera, A.L.; Pineda L., B. 2008. Preliminary results of *in vitro* antagonist bacteria on development of fungi isolated from *brachiaria brizantha* seeds. 2nd International Symposium on Biological Control of Bacterial Plant Diseases, Orlando, FL, USA, 4-7 November 2008.

Motta-Aldana, J.R.; Serrano-Serrano, M.L.; Hernández Torres, J.; Castillo-Villamizar, G.; **Debouck, D.G.**; Chacón S., M.I. 2008. Domestication patterns in wild Lima bean (*Phaseolus lunatus* L.) from the Americas. International Symposium Harlan II, University of California, Davis, CA, USA, 14-18 September 2008.

Serrano-Serrano, M.L.; Hernández Torres, J.; Castillo-Villamizar, G.; **Debouck, D.G.**; Chacón S., M.I. 2008. Gene pools in wild Lima bean (*Phaseolus lunatus* L.) and their origin. International Symposium Harlan II, University of California, Davis, CA, USA, 14-18 September 2008

Mafla, G.; Roa, J.C.; Flor, N.C.; **Aranzales, E.**; Moreno, M.G.; **Cuervo, M.; Debouck. D.G**. 2008. Distribution of cassava germplasm from an international genebank: a service to the global agriculture. Scientific Meeting of GCP21, Ghent, Belgium, 21-25 July 2008.

Sánchez, T.; Ceballos, H.; Dufour, D.; **Mafla, G.;** Calle, F.; **Debouck, D.**; Pérez, J.C.; Morante, N.; Tohme, J. Variation in starch and root quality traits in cassava. Cassava Meeting the Challenges of the new millennium, Ghent, Belgium, July 21-25 2008.

Calle, F.; Lenis, J.I.; Pérez, J.C.; **Mafla, G.; Debouck, D.**; Ceballos, H.; Tohme, J.; Morante, N. Multilocation evaluation of the cassava core collection from CIAT. Cassava Meeting the Challenges of the new millennium. July 21-25, Ghent, Belgium, 2008.

Lima, M.C.; Ciprián, A.; Toro, O.; Debouck, D.G. 2008. Distribution of bean and tropical forage germplasm from an international genebank: a service to the global agriculture. 30th Anniversary of CIAT GRU, Palmira, Colombia, 18 June 2008.

Lima, M.C.; Ciprián, A.; Toro, O.; Debouck, D.G. 2008. Distribución de germoplasma de fríjoles y forrajes tropicales desde un banco de germoplasma internacional como un servicio a la agricultura global. 30th Anniversary of CIAT GRU, Palmira, Colombia, 18 June 2008.

Lima, M.C.; Ciprián, A.; Toro, O.; Debouck, D.G. 2008. La colección del CIAT en el Depósito Global de Semillas de Svalbard. 30 Aniversario de CIAT URG, Palmira, Colombia, 18 June 2008.

Mafla, G.; Roa, J.C.; Flor, N.C.; **Aranzales, E.**; Moreno, M.G.; **Cuervo, M.; Debouck. D.G.** 2008. Distribución de germoplasma de yuca desde un banco de genes internacional: un servicio a la agricultura mundial. 30 Aniversario de CIAT URG, Palmira, Colombia, 18 June 2008.

Mafla, G.; Roa, J.C.; Flor, N.C.; **Aranzales, E.**; Moreno, M.G.; **Cuervo, M.; Debouck. D.G.** 2008. Distribution of cassava germplasm from an international genebank: a service to the global agriculture. 30th Anniversary of CIAT GRU, Palmira, Colombia, 18 June 2008.

Debouck, D.G. 2008. La solicitud de revisión de la patente sobre el frijol Amarillo 'Enola'. 30 Aniversario de CIAT URG, Palmira, Colombia, 18 June 2008.

6.6. Awards

Best Scientific Poster in Knowledge Sharing Week, People's Choice category: Additional evidence suggests a new map for the distribution of wild-weed-crop complexes of common bean in Colombia. International Center for Tropical Agriculture, Colombia, 7-12 April 2008.

6.7. Visitors

The Professional Staff of the Genetic Resource Unit attended the visit of 581 people from different government bodies, institutions, companies, etc. A total of 241 students from six different universities of Colombia visited the Genetic Resource Unit, on May 9 and October 24 2008 through the "Open House" days coordinated by CIAT Training Office.