BEAN GERMPLASM CONSERVATION BASED ON SEED DRYING WITH SILICA GEL AND LOW MOISTURE STORAGE

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PREFACE

This volume focusses on a simple low-cost technology based on seed drying with silica gel and low moisture storage for medium-term germplasm conservation. Procedures were developed for collection, characterization, maintenance, as well as for data management of bean germplasm collections.

This volume is the seventh in a working document series that serves research on beans (*Phaseolus vulgaris*) in Africa. Working documents will include bibliographies, research reports and bean network discussion papers. These publications are intended to complement an associated series of Workshop Proceedings.

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Further information on regional research activities on bean in Africa that are part of these projects is available from:

Pan-Africa Coordinator, CIAT, P.O. Box 23294, Dar es Salaam, Tanzania.

Coordinateur Regional, CIAT, Programme Regional pour l'Amelioration du Haricot dans la Region des Grands Lacs, B.P. 259, Butare, Rwanda.

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No. 16. Cinquieme Seminaire Regional sur l'Amelioration du Haricot dans la Region des Grands Lacs, Bujumbura, Burundi, 13-17 Novembre, 1989.

No. 17. Sixieme Seminaire Regional sur l'Amelioration du Haricot dans la Region des Grands Lacs, Kigali, Rwanda, 21-25 Janvier, 1991.

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BEAN GERMPLASM CONSERVATION BASED ON SEED DRYING WITH SILICA GEL AND LOW MOISTURE STORAGE

Martin Fischler[1]

ABSTRACT

Preservation of germplasm collections with low temperature storage is problematic because of power failures and equipment breakdown. Low moisture storage is an alternative to low temperature storage for medium-term germplasm conservation of seeds of most crops. Seed drying using silica gel for medium-term storage of bean seed was investigated. Seeds of two bean cultivars were dried for 50 days with silica gel in a desiccator experiment using a gel to seed ratio of 1:2. The final moisture content was 6.1 and 6.6% for the two cultivars. Dry seeds were stored in recycled glass soda bottles with screw caps sealed with candle wax at 25°C for one year. The seed moisture content remained constant confirming that recycled glass soda bottles can be used as inexpensive seed storage containers. Germination rates after one year of storage were 97.5 and 100% for the two cultivars. It is expected that the seed can be kept in glass bottles for 10-20 years (mid-term storage). In order to dry larger amounts of seed, a drying facility using silica gel in an air-tight PVC drum was developed. Procedures were developed for collection, characterization and maintenance of bean germplasm collections, as well as for data management.

INTRODUCTION

At the recent First Crop Science Conference for Eastern and Southern Africa, a researcher responsible for a national germplasm collection stated: "... our genebanks in Uganda are merely poor seed stores due to inadequate cool storage facilities."

Like many other germplasm collections, the Uganda bean

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germplasm until recently was stored at ambient conditions (13-14% moisture content, 25°C). Regeneration of all accessions every twoto three seasons was required to retain seed viability. Apart from the heavy workload, too frequent regeneration bears the risk of genetic drift and loss of accessions due to unfavorable weather conditions, insect pests and diseases.

On the other hand, low temperature storage commonly used in genebanks is expensive and problematic where power supply is unreliable and equipment breakdown frequent. It is not uncommon that whole collections are lost due to these problems.

This paper is intended for anybody responsible for medium-term germplasm conservation. It focusses on common beans (*Phaseolus vulgaris* L.) but most of the principles are equally applicable to other crop species. The paper is presented in three parts. The first part summarizes the findings of experimentation on (silica gel) seed drying and low moisture storage. The second part reports on the development of a low-cost technology for seed drying with silica gel and low moisture storage. The third part addresses collection, characterization, maintenance, as well as data management, of national bean germplasm collections, and presents procedures which could be standardized to ease the exchange of information. These procedures are mostly compatible with those recommended by the International Board of Plant Genetic Resources (IPBGR). Exceptions are discussed.

SUMMARY ON FINDINGS ON SEED DRYING WITH SILICA GEL AND LOW MOISTURE STORAGE

Literature review and objectives

Seed ageing of most crop species[2] is not only a function of time but also of temperature and seed moisture content (MC). Harrington (1973) reports that the life of seed is doubled by each 1% reduction in MC and by each 5°C reduction in temperature. More precise recent findings indicate curvy-linear relations between MC and the logarithm of longevity (Fig. 1) and between temperature and the logarithm of longevity (Fig. 2; Cromarty *et al.*, 1985, Ellis and Roberts, 1991). Those two figures show that a reduction of 1% MC has a relatively greater effect at lower MCs, whereas the effect of a reduction of 1°C in temperature becomes less at lower temperatures. The advantages of low moisture storage has led to an increase in ultra-dry storage of seeds for germplasm conservation (Ellis and Roberts, 1991). The viability of dried seeds is preserved provided the seeds are properly rehydrated prior to germination (Zhang and Tao, 1989; Ellis *et al.*, 1990). On the other hand, refrigeration to -20°C has become less emphasized in germplasm conservation. However, there is a low moisture content limit below which seed longevity is negligibly increased or even decreased. For common beans (*Phaseolus vulgaris*) a low moisture content limit of 5.7% (± 0.6) is reported (Ellis and Roberts, 1991).

Recommended methods for seed drying differ depending on the species, initial moisture content and resources available (Ellis and Roberts, 1991). However, hot-air drying techniques are difficult to operate under humid tropical conditions and need to

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[2] Seeds are classified into two groups according to different storage behaviour (Cromarty *et al.*). The first group described as "orthodox" includes most arable and horticultural crops. Seeds can be stored at low moisture and low temperature (sub-zero) conditions. The second group described as "recalcitrant" includes many important plantation crops, tropical fruit and a number of timber species. Recalcitrant seeds can neither be dried nor stored at sub-zero temperatures.

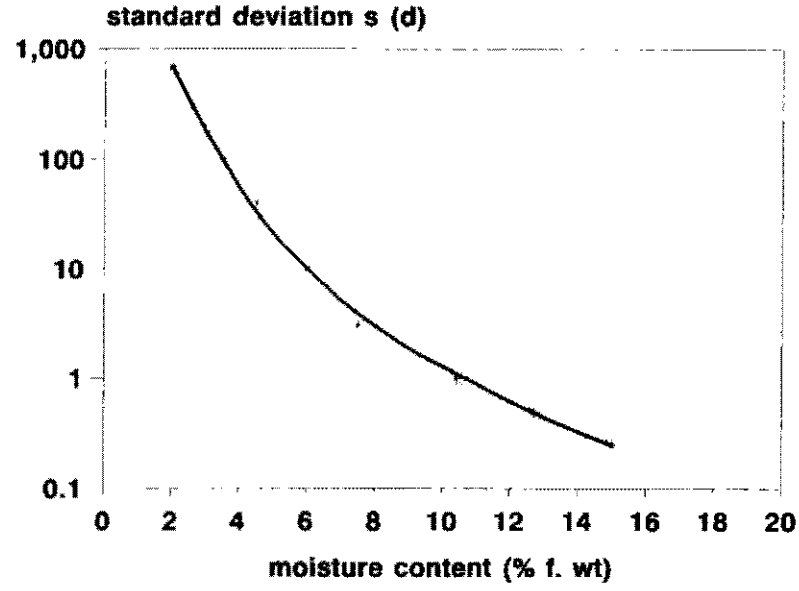

Fig. 1: The relation between seed moisture content and the standard deviation of the frequency distribution of seed deaths in time s (log scale) for *Sesamum indicum* L. in hermetic storage at 50°C (stars). The fitted line represents the logarithmic regression between seed longevity and seed moisture fitted to the stars. Modified from Ellis, Hong and Roberts (1986).

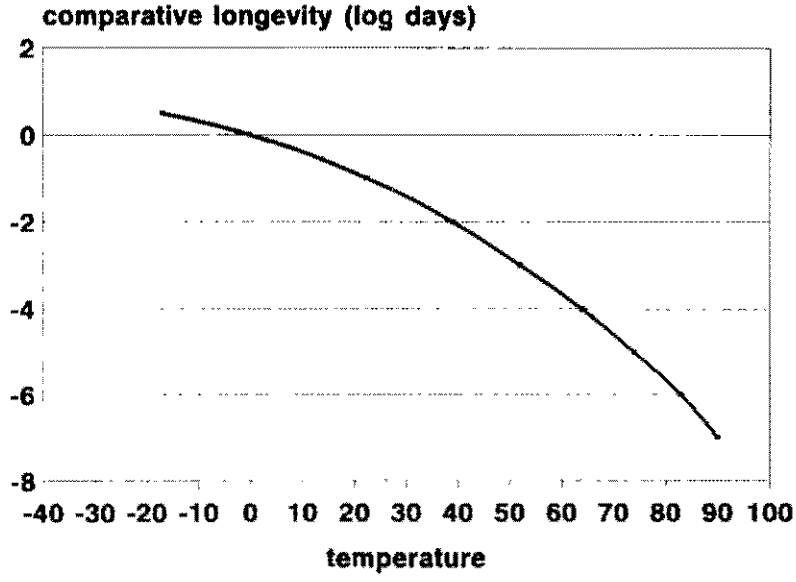

Fig. 2: The logarithmic relation between seed longevity and temperature (average values for several crops). Modified from Dickie, Ellis, Kraak, Ryder and Tompsett (1990).

be combined with dehumidification. Furthermore, hot-air drying can result in seed deterioration (Cromarty *et al.*, 1985). Instead, the use of silica gel for seed drying has been recommended, especially for genebanks which handle smaller collections of germplasm.

In view of these findings, storage of dried seed at ambient temperatures seems to be a promising alternative for genebanks which cannot provide reliable cool storage facilities for medium-term germplasm conservation.

The feasibility of bean seed drying using silica gel and seed storage aspects were studied. The objectives were:

i) to assess the seed drying behaviour of two bean cultivars with different seed size
ii) to assess the suitability of recycled glass soda bottles for medium-term conservation of bean germplasm
iii) to determine medium-term storage effects on seed viability

Materials and methods

Seed MC of two bean cultivars, White Haricot (22 g/100 seeds) and Rubona 5 (38 g/100 seeds), was measured prior to drying over silica gel using the modified high constant temperature oven method (International Seed Testing Association (ISTA), 1976). Two seed samples of 125 g of each cultivar were weighed and dried to 6.0-6.5% MC in a desiccator with a silica gel to seed ratio of 1:2. The desiccator was kept at 25°C. Silica gel with a color-changing ability (containing cobalt chloride) was replaced by the same amount of dry silica gel when 50-75% of the gel had changed its color. Seed MC was determined through weight loss after 10, 25 and 50 days. Dry seeds were put into recycled glass soda bottles (250 ml) with plastic screw caps and the bottles were sealed with candle wax. The bottles were then stored at 25°C. Seed MC was monitored through weighing every month over a period of one year. Germination tests were carried

out at seven months and one year after bottling. Prior to germination, forty seeds were rehydrated at ambient conditions (25°C; 70-90% relative humidity) for 2 and 6 days after storage of seven months and one year, respectively (imbibition damage at the first test suggested the need for a longer rehydration period).

Results and discussion

Seed MC of White Haricot and Rubona 5 dropped from 13.8% (seed equilibrium MC) to 6.1% and 6.6%, respectively, after 50 days of drying over silica gel. The silica gel had to be replaced four times during this period. The long drying period may be due to the relatively large seed size, the thick seed coat and the high protein content of bean seed. A more frequent replacement of the silica gel would decrease the drying period. Zhang and Tao (1989) indicate that 30-34 days are required to dry bean seeds from 14% to 5% MC using a silica gel to seed ratio of 1:2, replacing the silica gel when 12-23% of the silica gel had changed its colour. Alternatively, silica gel to seed ratios of 1:1 up to 3:1 could be used. However, when seeds are dried too fast physiological and mechanical damage to the seed can occur, especially if initial seed moisture is higher than 50% (Zhang and Tao, 1989).

Dry seeds stored for seven months in sealed glass bottles had a germination rate of 98% and 86% for Rubona 5 and White Haricot, respectively, compared to 100% and 98%, respectively, for non-dried seed (Table 1). When seeds were rehydrated at ambient conditions for six days instead of two days, the germination rate after one year of storage was comparable to the non-dried seed. The lower germination rate at seven months was probably a result of imbibition damage rather than seed deterioration *per se*. These results confirm findings of Zhang and Tao (1989), who did not observe a significant decrease in germination and vigour after bean seeds had been stored at 6.3% MC at 10-20 and 40°C for six months.

After one year of storage there was no difference in germination rates of dried seeds and seeds kept at seed equilibrium moisture content. However, with time the germination rate for non-dried seed is expected to decrease faster than for dried seed since deterioration of non-dried seed occurs at a faster rate.

Table 1: Germination rates (%) after seven and twelve months of storage at 25°C of two bean cultivars dried with silica gel compared with non-dried seeds (13.8 % MC)

	Germination rate (%)			
	White Haricot		Rubona 5	
Storage time	dried (6.1%MC)	non-dried	dried (6.6% MC)	non-dried
7 months (dried seed rehydrated for 2 days)	86	98	98	100
12 months (dried seed rehydrated for 6 days	98	98	100	100

Conclusion

Bean seeds can be successfully dried to 6% MC using silica gel without losing seed viability, provided the seeds are rehydrated for 6 days prior to germination tests. Recycled glass soda bottles sealed with candle wax proved to be air-tight. They are an inexpensive alternative to other seed storage containers such as laminated aluminum bags.

DEVELOPMENT OF A LOW-COST SEED DRYING TECHNOLOGY USING SILICA GEL

Using silica gel for seed drying is not new. However, there is little information about drying containers other than desiccators. Desiccators are expensive, easily breakable and can not handle large amounts of seeds. Other containers could be used for silica gel seed drying provided they:

- are air-tight;
- allow easy filling/removal of seeds and silica gel;
- allow adequate air circulation; and
- are inexpensive.

The above criteria are met by a high density polyvinyl chloride (PVC) open head drum (diameter 0.45 m, height 0.75 m, volume 120 l; Fig. 3). An air-tight seal is achieved through a rubber gasket in the cover and a metal spanner pulling drum and cover tightly together.

A metal cylindrical grill construction is fitted to hold silica gel in the center of the drum (Fig. 3, 4) The 20 cm diameter of the central cylinder allows for a silica gel to seed ratio of approximately 1:2. Cloth bags of silica gel are put in the center and cloth bags of seed around the central cylinder (Fig. 5). Two rings near the periphery prevent over-packing of the seeds and allow good air-circulation. The silica gel is removed for drying and is replaced by an equal amount when about 25% of the silica gel has lost its dark blue color. The silica gel should be changed more frequently at lower MCs. (A second batch of silica allows for continuous drying).

This construction allows for drying of approximately 16 kg of bean seed. Bean seeds were successfully dried to 6.4% MC during a period of nine weeks, while replacing the silica gel nine times (Fig. 7). A shorter drying time can be achieved if the silica gel is replaced more frequently. Alternatively, a silica gel to seed ratio of 1:1 could be used but the cost will be increased, as silica gel, while reusable, is the costly component

Fig. 3: Open-headed PVC drum with air-tight cover for silica gel seed drying (left) and a metal construction fitted in the drum to separate silica gel from seed (right).

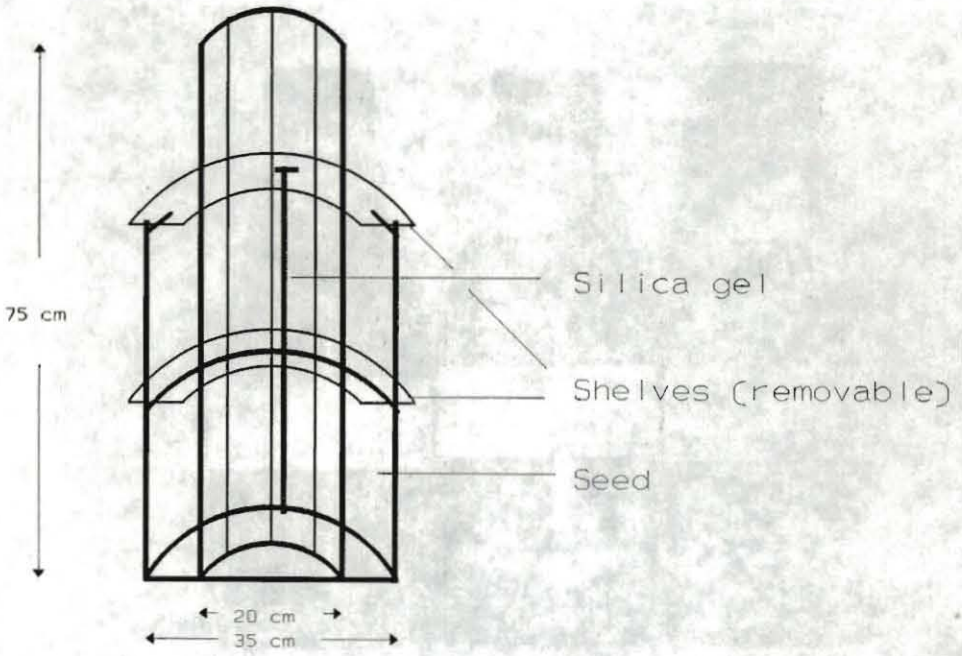

Fig. 4: Longitudinal section of metal construction to fit in the PVC drum (simplified).

Fig. 5: PVC drum filled with seed (periphery) and silica gel (central cylinder).

Fig. 6: Recycled glass soda bottles with screw cap sealed with candle wax for seed storage.

of this technology. To further reduce costs, the indicating silica gel could be mixed with the cheaper non-indicating type at a ratio of 1:10. An even less expensive alternative is the use of toasted rice (<1% MC) as a desiccant. In an experiment at the International Potato Center (CIP), soybeans were dried in two weeks from 11.4% to 5.6% MC using a rice to seed weight ratio of 1.5:1 (Sadik and White, 1982). Further experimentation with bean is necessary to confirm the suitability of toasted rice as a desiccant for bean seed drying. A cost estimate for the drying facility is given in Appendix 1.

Seed storage in glass soda bottles with screw caps is convenient. However, the seals are not always perfect. Sealing with candle wax is a quick and inexpensive method to assure hermetic sealing. The top of the bottle is simply dipped into melted candle wax so that the whole cap is covered with a layer of wax (Fig. 6).

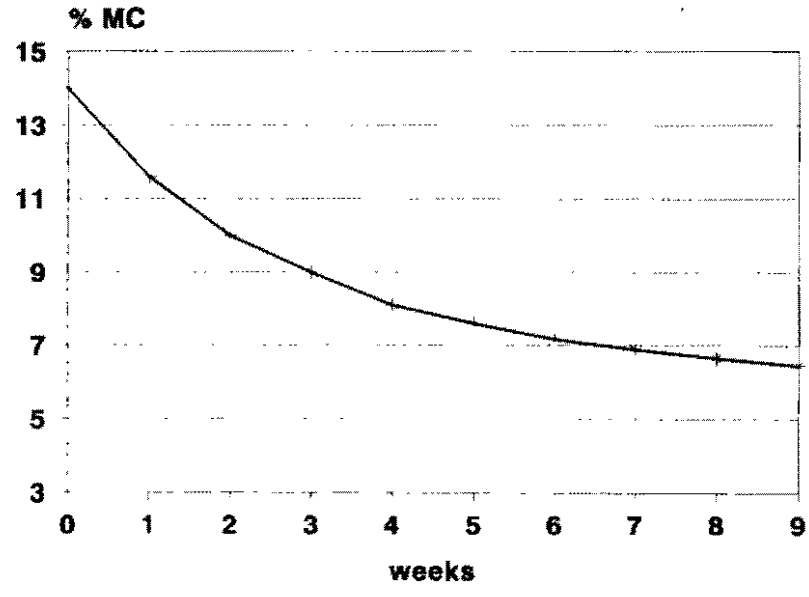

Fig. 7: Drying curve of bean seed dried with silica gel in PVC drum (% MC on fresh weight basis)

STANDARD PROCEDURES FOR BEAN GERMPLASM COLLECTIONS

Procedures are given for bean germplasm collections including collection, preliminary evaluation and maintenance, as well as data management. Emphasis is put on simplicity of the procedures, taking into account operational constraints and limited number of trained personnel available. Standardization of procedures is recommended to facilitate exchange of information and germplasm between genebanks. Figure 8 illustrates the individual steps in handling germplasm. They include:

- Collection of accessions and passport data
- Multiplication and preliminary characterization/evaluation
- Seed cleaning
- Determination of moisture content
- Seed drying
- Determination of seed viability
- Seed storage
- Monitoring seed viability
- Multiplication for distribution
- Regeneration

Collection of accessions and passport data

Systematic collection of germplasm is costly. Therefore the Uganda Bean Programme has adopted another approach where non-government organizations, other programmes and projects, and government institutions distributed over the country are asked to collect seed. In addition, researchers from the Bean Programme collect seed on trips made for purposes other than germplasm collection. However, while cost-effective, this approach has the disadvantage that germplasm is not collected systematically and therefore leaves gaps in the collection.

A proposed format for a passport data collection sheet for bean germplasm is given in Appendix 2. A minimum of 200 seeds for pure cultivars and 500 seeds for mixtures should be collected.

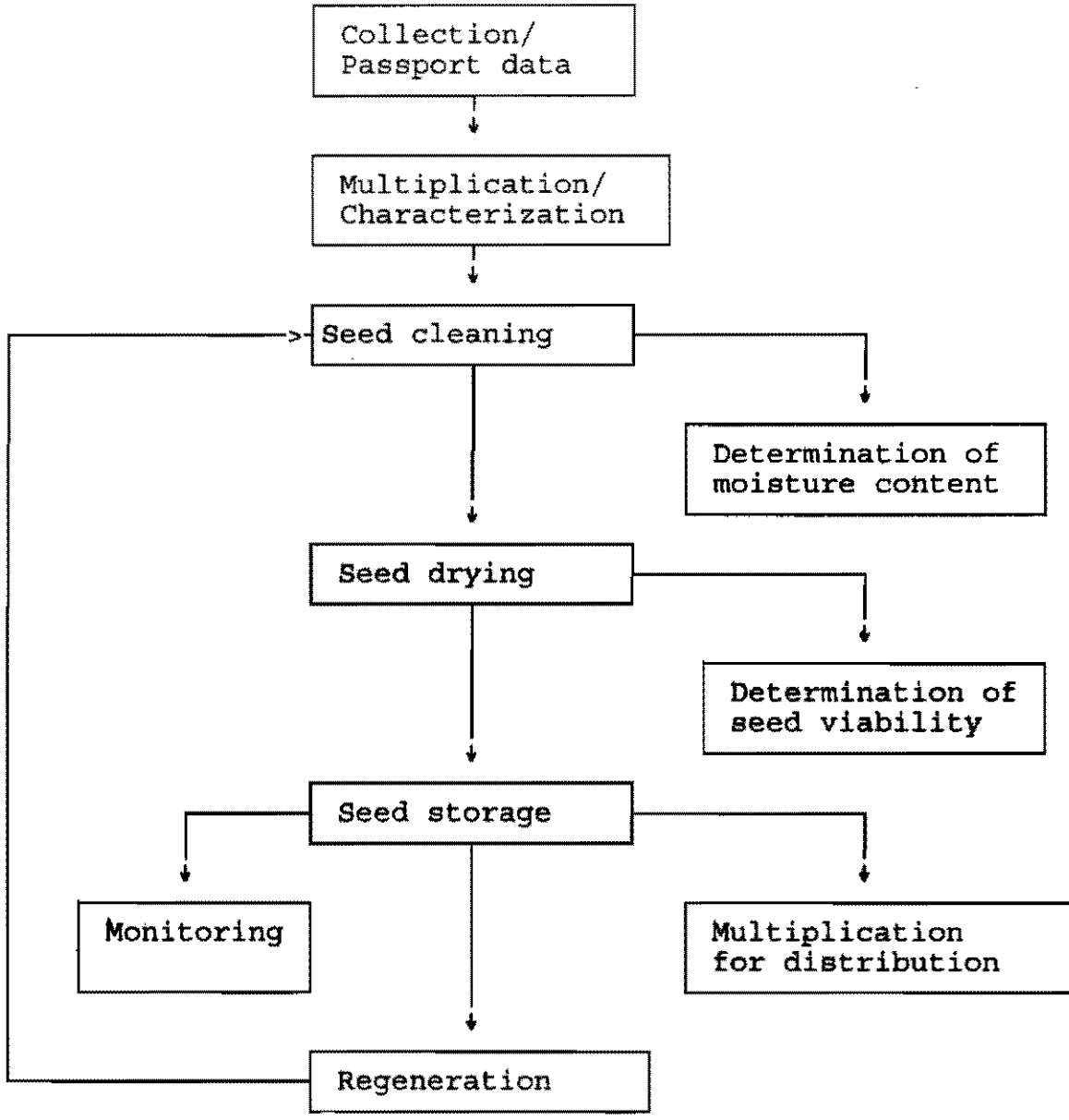

Fig. 8: Steps in handling germplasm (mod. from Hanson, 1985).

Seeds should be collected at the farm level and additional information should be obtained from the person growing the crop species/cultivar. More recently, IBPGR has embarked on a programme with emphasis on recording more ethno-botanical, social, economic and cultural information (Sperling, 1993, Toll, 1993; pers. comm.). However, at this moment there is no descriptor list or guidelines for this. An attempt to collect data on indigenous knowledge (IK) is made in points No. 13,14, 19, 20 and 21.

Data from collection (= passport data) is entered in a database (as outlined below) using the descriptors given in Appendix 3.

Multiplication and preliminary characterization/evaluation

Newly collected seed samples are checked for insect damage, diseases and shrivelled seed. A minimum of 100-120 clean bean seeds are planted for preliminary characterization/evaluation and multiplication.

For characterization, IBPGR gives a list of more than 60 descriptors (IBPGR, 1982). CIAT currently uses 25 field descriptors and six seed descriptors (Hidalgo, 1991). Evaluation of resistance to pests and diseases is carried out separately from characterization at CIAT. However, since the resources of National Programmes are often limited, characterization and evaluation of resistance for pests and diseases is carried out at the same time using a reduced descriptor list as shown in Appendix 4. The list consists of 15 field descriptors and the six seed descriptors currently used by CIAT.

Seed cleaning

Seeds should be cleaned immediately after threshing. Plant debris, non-viable seeds, and seeds damaged by insect pests and diseases should be destroyed to prevent the spread of infection.

Determination of seed moisture content (MC)

Seed moisture content can be accurately predicted if the storage conditions are known. The seed MC will equilibrate with the relative humidity of the surrounding air. This so called equilibrium moisture content is constant for any species at known air temperature and relative humidity and can be used as an approximation of the actual MC. For *Phaseolus spp.* the equilibrium moisture contents at 25°C are 9.2%, 11.0% and 13.8% at 45%, 60% and 75% relative humidity, respectively (Roberts, 1972).

A more precise method is by oven determination of the MC as described by the International Seed Testing Association (ISTA, 1976). The methods differ depending on the species and MC. For beans (*Phaseolus spp.*), about 4 g of seed (pre-dried if MC is >17%) is crushed into pieces not larger than 4 mm, weighed to 0.0001g using an analytical balance, and then dried in the oven for 1 hour at 133°C. Percent MC is calculated on a wet weight basis and expressed to one decimal place.

Alternatively, the MC can be measured using a high quality seed moisture meter. However, its precision should be checked comparing its readings with the oven method.

Seed drying

Cleaned seed is pre-dried in the shade to a MC below 15% (sun drying of very moist seed can lead to seed deterioration). Seeds kept at interim MCs of 15-20% lose viability most rapidly (Toll 1993, pers. comm.). After weighing, seeds are dried with silica gel in a PVC drum to a MC of 6%. About 400 g of seed per accession is put in a cloth bag and placed at the periphery of the drum (around the central metal cylinder). The silica gel is put in fine cloth bags and placed in the central cylinder (Fig. 5). The drum is then closed.

The silica gel is removed for drying when about 25% has lost its dark blue color (about every 4-6 days) and replaced by an equal amount of dry silica gel. The silica gel can be made into permanent packages of 2 kg in fine cloth bags. These can be dried in a drying oven at 110-120°C without damage and are easier to handle than loose silica gel. After drying the silica gel should be left to cool before being placed in the drum.

The MC is best monitored by monitoring weight loss using the formula:

$$\%MC_t = 100 - [\frac{W_o}{W_t} \times (100 - \%MC_o)]$$

where as:
% MC_t = moisture content at measuring time
W_o = initial seed weight
W_t = seed weight at measuring time
% MC_o = initial seed moisture content

Determination of seed viability

The initial viability should be determined, before seeds are stored, with a germination test. IBPGR recommends a fixed sample size of 200 seeds for initial viability testing. However, in order to save seed for the collection a modified sequential germination test using a maximum of 80 seeds is suggested here for bean germplasm (Fig. 9).

In order to prevent imbibition damage, dry seeds should be rehydrated for 6 days at ambient conditions. Seeds are germinated in petri-dishes on filter paper moistened with distilled water. The number of germinated seeds are counted and the percentage germination calculated. Accessions with less than 85% viability should be regenerated (see Fig. 9).

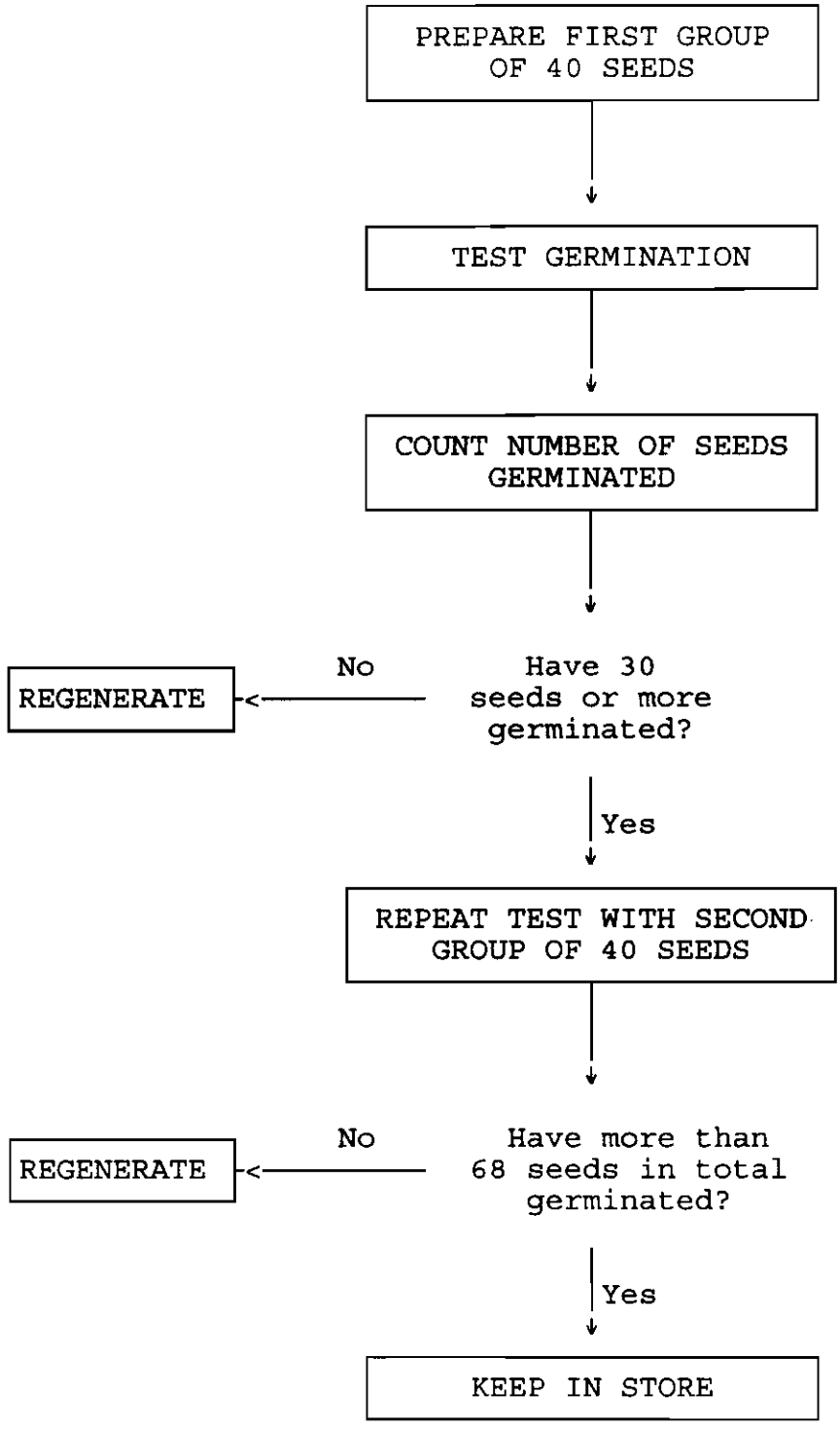

Fig. 9: Sequential germination test (mod. from Hanson, 1985)

Seed Storage

Storage containers. Dry seeds should be stored in moisture-proof containers. For medium-term storage glass, thick plastic or metal containers could be used as an alternative to more expensive laminated aluminum bags. However, they should be tested in advance in order to assure that they are moisture proof (e.g. with oven-dried salt). To improve sealing, petroleum jelly can be put around the thread and the cap sealed with candle wax. Metal containers should be rust-proof. The Uganda bean germplasm is stored in recycled glass soda bottles (see above).

Storage conditions. IBPGR recommends a storage temperature of -20°C for base collections and 0°C to 10°C for medium-term storage. However, if no cooling equipment is available or power supply is unreliable, storage at ambient conditions can be accepted as a compromise for medium-term storage. Preferably, seeds should be stored in a cool place with no major temperature fluctuations (e.g. cellar or basement).

Number of seeds stored. A minimum accession size of 3,000 seeds for genetically homogeneous accessions is recommended by IBPGR (1985). However, for large seed species it may be necessary to reduce the number of seeds stored. The accession size of the Uganda bean germplasm collection is currently at least 300 g per accession (= 500 to 2000 seeds assuming a range of the 100-seed weight from 15 to 60 g). The small sample size has the following implications:

1. no seed is immediately available for distribution, but has to be first multiplied, and
2. a sequential germination test only using 80 seeds is used for initial viability determination and monitoring.

Monitoring the accessions

The viability of seeds should be checked at regular intervals. This interval depends on the species, initial viability, moisture content and storage temperature. As a guideline seeds in medium-term storage or seed with poor initial viability should be tested every three to five years using the modified sequential germination test.

Regeneration of accessions

Regeneration is the renewal of an accession by sowing a random sample of seeds and growing the resulting plants under conditions so that the seeds harvested will have the same characteristics as the original accession (Hanson, 1985).

Regeneration of an accession is necessary when the number of seeds falls below a critical level and if the seed viability falls below 85%. Seed for at least three regeneration cycles should be stored (in case of loss due to unfavorable weather conditions, insect pests and diseases). For beans a minimum of 300 seeds should be available for regeneration.

Data management

Modern genebanks handling large germplasm collections use advanced software for data management. CIAT is currently using a mainframe program called "Oracle" (Tohme, 1993; pers. comm.). However, if data has to be efficiently handled by national programmes with fewer facilities, a simple PC-based database programme is recommended. A widely used and easy to learn database programme is DBASE III Plus (Copyright Ashton-Tate, 1984-86). The programme takes only 0.55 MB space and can even be run from a floppy diskette. Some countries are already handling (bean) germplasm data with DBASE. Appendix 5 gives the format

currently used for the Uganda bean germplasm collection. The use of a standard format would ease the exchange of information between collections of different countries.

Passport data and preliminary characterization/evaluation data is kept in two separate files (PASS-UGA.DBF and CHAR-UGA.DBF, respectively). Accession number and name are the same for both files (common fields).

DBASE is menu-driven but after some experience it is more convenient to work from the command prompt. A list of the most commonly used commands is given is Appendix 6. For more information a DBASE handbook should be consulted. Small programmes can be written in DBASE to speed up routine commands and even to further simplify the use of the programme.

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APPENDIX 1: Cost estimate for seed drying and storage facility using silica gel in an air-tight PVC drum. The cost for storage space is not considered.

Item	US $
PVC drum (120 l)	50
Fitting of metal cylinder	20
Silica gel (20 kg x US $ 20)	400[1]
Bottles (for 1000 Accessions)	80[2]
Total:	550

[1] Cost will be less than half if cheaper non-indicating silica gel is mixed with self-indicating gel at a ratio 10:1.

[2] Recycled bottles can often be purchased for free. However, it may be necessary to offer a small amount to get the bottles (e.g. UShs. 100 = 8 US cents)

APPENDIX 2: Format for "passport data collection sheet for bean germplasm"

Assigned Accession No.: _____
(leave blank!)

1. Collector's Name (last name): _____

2. Collector's sample No.: _____

3. Date of collection (month/day/year): _____

4. Country of collection: _____

5. District/County: _____

6. Village (or parish)/town: _____
(village/town or km and direction from nearest village/town, e.g. Hoima7N)

7. Latitude of collection site: _____
(Degrees and minutes followed by N or S, e.g. 1030 S)

8. Longitude of collection site: _____
(Degrees and minutes followed by E or W, e.g. 0545 E)

9. Altitude of collection site: _____
(Elevation above sea level in meters)

10. Collection source: _____
(1=wild; 2=farm land; 3=farm store; 4=local market; 5=commercial m.; 6=institute; 7=other)

11. Name of donor person (indicate sex): _____

12. Name of variety (indicate other names): _____

13. Translation/meaning of name: _____

14. Ethnic group and language: _____

15. No. of plants sampled (at least 5 plants): _____
(Collect at least 200 seeds for pure lines and 500 seeds for mixtures)

16. Type of sample: _____
(1=landrace pure; 2=landrace mixture; 3=bred cultivar pure; 4=bred cult. mixture; 5=other)

17. Growth habit: _____
(b=bush; scl=semi-climber; cl=climber)

18. Seed colour: _____
(1=white; 2=cream-beige; 3=yellow; 4=coffee (brown); 6=red; 7=purple; 8=black; 9=other)

19. Susceptibility to: (3=low susceptibility; 5=medium s.; 7=high s.)

a) pests (specify if known): _____
b) diseases (specify if known): _____
c) drought: _____
d) low soil fertility: _____

20. Cultivation: _____
(1=monoculture; 2=mixed with maize; 3=mixed w. cassava; 4=mixed w. banana; 5=mixed w. others)

21. Ask farmer why s/he is growing this variety (use, preference, specialties)

_____

_____

_____

APPENDIX 3: Descriptor list for the passport data file of the Uganda bean germplasm collection

Column	Abbreviation	Description and legend (Data has to be entered exactly as specified)
1	ACCNO	Accession number: "u" followed by a number assigned to each accession entering into the collection, e.g. u125. **The ACCNO is always the same for each accession in both the passport and the characterization file.** Mixtures are entered both as one accession and separated into components. Components bear the accession number followed by the component number separated with a decimal point (e.g. 1.1, 1.2 = first two components of mixture one.)
2	CONA	Collector's name (last name) of original sample.
3	DATE	Date of collection of original sample (mm/dd/yy = month/day/year)
4	CTRY	Country of collected sample. Use the three-digit abbreviations defined by UN and IBPGR: BND = Burundi; CMR = Cameroon; ETP = Ethiopia; KYA = Kenya; MWI = Malawi; RWN = Rwanda; SUD = Sudan; TZA = Tanzania; UGA = Uganda; ZRE = Zaire; ZBA = Zambia
5	DRCT	Name of district/province (first geopolitical subdivision at the national level).
6	CNTY	Name of county (second geopolit. subdivision).
7	LOCN	Geographical location of collection site. Indicate village/town, or number of km and direction from nearest town/village (e.g. Hoima7N means 7 km north of Hoima)
8	LATI	Latitude of collection site (Degrees and minutes followed by North or South, e.g. 1030S).
9	LONG	Longitude of collection site (Degrees and minutes followed by West or East, e.g. 0545E).
10	ALTI	Altitude. Elevation above sea level in metres.
11	SRCE	Collection source (1=wild; 2=farm land; 3=farm store; 4=local market; 5=commercial market; 6=institute; 7=other).
12	DOPO	Name of donor person
13	DSEX	Sex of donor person

APPENDIX 3 (cont.)

Column	Abbre-viation	Description and legend (Data has to be entered exactly as specified)
14	NAME	Local vernacular name of accession. Other names for the same variety in brackets, e.g. K20 (nambale).
15	TRAN	Translation/meaning of local vernacular name.
16	ETHN	Ethnic group of donor person (or ethnic group living in the area where sample was collected).
17	LANG	Language spoken by ethnic group indicated
18	TYPE	Type of sample (1=landrace pure; 2=landrace mixture; 3=bred cultivar pure; 4=bred cultivar mixture; 5=other).
19	GRHB	Growth habit (b=bush; cl=climber).
20	COLO	Primary seed colour (1=white; 2=cream-beige; 3=yellow; 4=coffee (brown); 5=pink; 6=red; 7=purple; 8=black; 9=other).
21	PEST	Susceptibility to pests in general (specify if known!). Scale 1-9 where: 3=low susceptibility; 5=medium susceptibility; 7=high susceptibility).
22	DSEA	Susceptibility to diseases in general (specify if known!). Scale as for pests.
23	DGHT	Susceptibility to drought. Scale as for pests.
24	LSFY	Susceptibility to low soil fertility. Scale as for pests.
25	CULT	If under cultivation, cropped as: 1=monoculture; 2=mixed with maize; 3=mixed with cassava; 4=mixed with banana; 5=mixed with others.
26	ACCS	Accession size (in g)
27	LREG	Date of last regeneration (last planted in Kawanda; year and season, e.g. 1992B = second season in 1992).
28	NREG	No. of times accession was regenerated (=planted in Kawanda) since collection.
29	NOTE	Farmers comments about the variety including the pests and diseases from above if specified.

APPENDIX 4: Descriptor list for the characterization/preliminary evaluation data file of the Uganda bean germplasm collection

Column	Abbreviation	Description and legend (Data has to be entered exactly as specified)
1	ACCNO	As for passport data file.
2	NAME	Local vernacular name of accession
3	PGH	Plant growth habit: 1=bush determinate, type I; 2=bush indeterminate, type II; 3=semi-climber/prostrate indeterminate, type III 4=climber indeterminate, type IV
4	FCW	Flower colour of wings (1=purple; 2=red; 3=pink; 4=yellow; 5=white; 6=green).
5	FCS	Flower colour of standard (colour codes as FCW)
6	DFF	Number of days from emergence to stage where 50% of plants started flowering.
7	DEF	Number of days from emergence to stage where 50% of plants ended flowering.
8	DPM	Days from emergence to physiological maturity.
9	PLH	Plant height (average, in centimetres, at maturity from 5 plants, measured from cotyledon scar to tip of plant).
10	NFR	Nodes on stem to first raceme, measured at physiological maturity.
11	POP	Position of pods (1=high; 2=low; 3=evenly distributed, uniform).
12	PFC	Pod fibre content in fully expanded immature pods (0=stringless; 3=few strings; 5=moderately stringy; 7=very stringy).
13	PCO	Pod colour at physiological maturity (1=dark purple; 2=red; 3=pink; 4=yellow; 5=cream; 6=brown; 7= persistent green).
14	DHA	Days to harvest (Number of days from emergence until 90% of pods are dry)
15	PPP	Number of pods per plant (Average of 10 plants at harvest).
16	SPP	Number of seeds per pod (Average number of seeds from one pod taken from 10 plants).

APPENDIX 4 (cont.)

Column	Abbre-viation	Description and legend (Data has to be entered exactly as specified)
17	PSC	Primary seed colour (Background seed colour; 1=white; 2=cream-beige; 3=yellow; 4=coffee; 5=pink; 6=red; 7=purple; 8=black; 9=other).
18	SCP	Seed coat pattern (1=plain; 2=mottled; 3=striped; 4=spotted; 5=speckled; 6=ringed).
19	SSC	Secondary seed colour (colour of pattern; colours as for PSC).
20	SSH	Seed shape (1=round; 2=ovoid; 3=cuboid; 4=kidney shaped; 5=truncate fastigate.
21	SCL	Seed coat lustre (brilliance; 1=dull (matt); 2=medium; 3=shiny).
22	HSW	100 seed weight (in g to first decimal place at 12-14% seed moisture content).
23	YIE	Seed yield (g/plot)
24	BCM	Bean Common Mosaic Virus, BCMV (scores 1-9 where: 0=no symptoms; 3=low susceptibility; 5=medium susceptibility; 7=high susceptibility).
25	CBB	Common Bacterial Blight (scores as for BCMV).
26	ALS	Angular Leaf Spot (scores as for BCMV).
27	RST	Rust (scores as for BCMV).
28	ASC	Ascochyta (scores as for BCMV).
29	BSM	Bean Stem Maggot (=Beanfly; scores as for BCMV).

APPENDIX 5: Database structures for the passport and characterization/preliminary evaluation data file of the Uganda bean germplasm collection.

a) passport data file

No.	Field Name	Type	Width	Dec
1.	ACCNO	Character	8	
2.	CONA	Character	10	
3.	DATE	Date	8	
4.	CTRY	Character	3	
5.	DRCT	Character	10	
6.	CNTY	Character	12	
7.	LOCN	Character	15	
8.	LATI	Character	5	
9.	LONG	Character	5	
10.	ALTI	Numeric	4	0
11.	SRCE	Character	1	
12.	DOPO	Character	12	
13.	DSEX	Character	1	
14.	NAME	Character	23	
15.	TRAN	Character	20	
16.	ETHN	Character	12	
17.	LANG	Character	12	
18.	TYPE	Character	1	
19.	GRHB	Character	2	
20.	COLO	Character	3	
21.	PEST	Character	1	
22.	DSEA	Character	1	
23.	DGHT	Character	1	
24.	LSFY	Character	1	
25.	CULT	Character	5	
26.	ACCS	Numeric	3	0
27.	LREG	Character	5	
28.	NREG	Character	1	
29.	NOTE	Character	100	

b) preliminary characterization/evaluation data file

No.	Field Name	Type	Width	Dec
1.	ACCNO	Character	8	
2.	NAME	Character	23	
3.	PGH	Character	1	
4.	FCW	Character	1	
5.	FCS	Character	1	
6.	DFF	Numeric	2	0
7.	DEF	Numeric	2	0
8.	DPM	Numeric	2	0
9.	PLH	Numeric	2	0
10.	NFR	Numeric	2	0
11.	POP	Character	1	
12.	PFC	Character	1	
13.	PCO	Character	1	
14.	DHA	Numeric	2	0
15.	PPP	Numeric	2	0
16.	SPP	Numeric	1	0
17.	PSC	Character	1	
18.	SCP	Character	1	
19.	SSC	Character	1	
20.	SSH	Character	1	
21.	SCL	Character	1	
22.	HSW	Numeric	4	1
23.	YIE	Numeric	5	1
24.	BCM	Character	1	
25.	CBB	Character	1	
26.	ALS	Character	1	
27.	RST	Character	1	
28.	ASC	Character	1	
29.	BSM	Character	1	

APPENDIX 6: List of common commands (from command prompt) in DBASE

Dbase is menu-driven. It is recommended to use the menu for creating a database. However, after some experience it is more convenient to manage the data from the command prompt (press ESC while in the menu mode). Return to the menu "assist" typed at the command prompt. The command line at the bottom of the screen displays drive, file opened, current/total record number.

Operation	COMMAND (Press 'ENTER' to execute)	Description
Create new files		
Create a database	CREATE	Creates a database with the name specified (with Extension .DBF)
Create a print format	CREATE REPORT	Creates a format for printing with the columns specified. 'Exit', 'Save' to save the report format (Extension .FRM).
Create labels	CREATE LABEL	Creates labels with information from the database. (Can be used to label stored seed).
Open a database		
Open a database	USE [filename]	Opens a database with the name specified.
Open a database with an index	USE [filename] INDEX [indexfile], [ind..]	Opens a database which is sorted according to the index specified. The database has to be indexed before (see "sort data" below).
Modify a database etc.		
Modify a database	MODIFY STRUCTURE	Edits the structure of the database and changes can be made. Press 'Ctrl End' to save changes.
Modify a print format	MODIFY REPORT	Edits the print format specified and changes can be made.
Modify labels	MODIFY LABEL	Edits the label format specified and changes can be made.
Find/locate records		
Find a record No.	GOTO X GOTO TOP GOTO BOTTOM	Go to the record No. X Go to beginning of database Go to end of database
Locate a record	LOCATE FOR	Locates a record with specific data. Use the same syntax as for the DELETE command. Type EDIT to edit the record located.

Example:
LOCATE FOR DRCT = 'kabale'
---> locates the first record with district (DRCT) = kabale

Add/edit data

Go into database and add/delete data	EDIT	Edits individual records and data can be added/ deleted.
	APPEND	Cursor is positioned at the end of the database on top of the next new record to enter data.
	BROWSE	The whole database is edited (each record on one line) and data can be added/deleted.

Delete/undelete records/data

Delete an edited record	Crtl U	Marks an edited record for deletion ('Del' appears at the bottom right of the status line).
Delete a record (from command prompt)	DELETE	Marks the current record for deletion (with a *).
Delete specified	DELETE FOR [field] [separator] [data]	Deletes data in a column data (field) which meets the range specified. Separators are =, <, >, <=, >=, <>. Data in character field has to be within ' '. Fields can be combined with .AND./.OR. .

Examples:

DELETE FOR DRCT = 'kabale'
---> Deletes all records which have 'kabale' in the field DRCT (District)

DELETE FOR DRCT = 'kabale' .AND. FOR ACCS < 200
---> Deletes all records from kabale with accession sizes (ACCS) < 200 g

DELETE FOR DRCT = 'kabale' .OR. FOR ACCS < 200
---> Deletes all records from kabale or all records with accessions < 200g
(Note the difference caused by AND/OR!)

Undelete data/records	RECALL, RECALL FOR..	Undeletes (restores) data. The same syntax as for the delete command has to be used. **Data can not be restored after the PACK command has been used.**
Final deletion of records/data	PACK	Deletes records/data marked for deletion with the DELETE command.

Replace data

Replace data in records/fields	REPLACE [field] WITH [new] FOR [field] [separator] [old]	Replaces data in all records for the field indicated and the range specified. Fields can be combined with .AND./.OR. Separators are =. <, >, <=, >=, <>.

Example:
REPLACE DRCT WITH 'kisoro' for CNTY = 'bufumbira' .AND. ETHN = 'bafumbira'
---> replaces the district (DRCT) name with 'kisoro' in all the records with 'bufumbira' as county (CNTY) name and 'bafumbira' as ethnic group (ETHN).

Sort data/file

Sort data	SORT	Sorts data according to the fields specified (first field = primary key) and stores sorted data in a new file specified. The original file is not changed.
Sort data with an index	INDEX	Sorts original file according to fields specified. Several fields can be combined separated with ", ". The first field specified is the master index (primary key). The original file is sorted. The index fields are stored in the index file specified (Extension .NDX.

---> To keep the index up to date the index file has to be opened every time the file is opened (USE [file] INDEX [file]. Alternatively, the index can be created every time a database is sorted. (The INDEX command is faster than the SORT command.

Print data

Print directly	DISPLAY TO PRINT	Prints the current record.
	DISPLAY ALL TO PRINT	Prints all records.
	DISP NEXT X TO PRINT	Prints specified number of records.
	DISP [field], [field] etc. ALL TO PRINT	Prints specified columns of all records
	DISP [field], [field] NEXT X TO PRINT	Prints specified columns of number of records indicated.

---> The record # is printed in the first column when printed directly.
---> Without TO PRINT the data is shown on the screen only.

Print from a report format	REPORT	Uses the report format indicated for printing. No Record # is displayed in the first column.

Miscellaneous

set default drive	SET DEFAULT TO A: (or C:, B:)	Specifies drives where files are retrieved from and saved to.
Interrupt a command	Press 'Esc'	Interrupts command and goes back to command prompt.
Print on-line	SET PRINT ON/OFF	Prints all commands/operations directly if in ON mode.
Exit DBASE	QUIT	

Note: Only the four first letters of the commands have to be typed (e. g. MODI STRU for MODIFY STRUCTURE).