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THE DESIGN OF SEED STORAGE FACILITIES FOR GENETIC CONSERVATION

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DESIGN OF SEED STORAGE FACILITIES FOR GENETIC CONSERVATION

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The International Board for Plant Genetic Resources (IBPGR) is an autonomous international scientific organization under the aegis of the Consultative Group on International Agricultural Research (CGIAR). IBPGR was established by the CGIAR in 1974. The basic function of IBPGR is to promote and coordinate an international network of genetic resources centres to further the collecting, conservation, documentation, evaluation and use of plant germplasm and thereby contribute to raising the standard of living and welfare of people throughout the world. Financial support for the core programme is provided by the Governments of Australia, Austria, Belgium, Canada, China, Denmark, France, FRG, India, Italy, Japan, the Netherlands, Norway, Spain, Sweden, Switzerland, the UK and the USA, the United Nations Development Programme and the World Bank

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PREFACE TO THE 1990 REVISION

Such has been the demand for this IBPGR Handbook that it has been reprinted twice. The opportunity has been taken at this third reprinting (1990) to revise Appendix 2 (with regard to seed storage characteristics) and to provide additional information on seed storage in Appendix 4, which has arisen as the result of IBPGR-funded research at Reading, regarding the IBPGR preferred conditions for long-term seed storage for genetic conservation.

We have not attempted to update any of the original estimates of costs. Calculations based on general price inflation since 1981 are more than adequate to allow for increases in these costs in cash terms but will vary between countries. Experience has shown that it has been possible to construct genebanks to the preferred standards slightly more cheaply than the 1981 indexed estimates. Some stores have cost a lot more, but the extra costs were not essential to achieving the aims outlined here.

Those responsible for handling seed accessions in genebanks should also consult the following two IBPGR Handbooks published in 1985.

IBPGR Handbook No. 2.
R.H. Ellis, T.D. Hong and E.H. Roberts, 1985. Handbook of Seed Technology for Genebanks, Volume I. Principles and Methodology, 210 pp. International Board for Plant Genetic Resources, Rome.

IBPGR Handbook No. 3.
R.H. Ellis, T.D. Hong and E.H. Roberts, 1985. Handbook of Seed Technology for Genebanks, Volume II. Compendium of Specific Germination Information and Test Recommendations, pp. 211-667. International Board for Plant Genetic Resources, Rome.

The following IBPGR Manual was especially designed as a practical reference for technicians and staff involved in the day-to-day handling of seed accessions (and is consequently less detailed than the above). It is therefore useful to those responsible for staff training in genebanks.

J. Hanson, 1985. Procedures for Handling Seeds in Genebanks, 115 pp. International Board for Plant Genetic Resources, Rome.

In addition, the following recent IBPGR publications will be of interest to those concerned with recalcitrant seeds and the regeneration of seed accessions, respectively.

H.F. Chin, 1988. Recalcitrant Seeds - A Status Report, 28 pp. International Board for Plant Genetic Resources, Rome.

E.L. Breese, 1989. Regeneration and Multiplication of Germplasm Resources in Seed Genebanks, 69 pp. International Board for Plant Genetic Resources, Rome.

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PREFACE

The International Board for Plant Genetic Resources includes in its terms of reference 'the determination of minimum standards for conservation and regeneration of stocks of both seeds and vegetative material'.

FAO, through its Panel of Experts on Plant Exploration and Introduction, made recommendations in 1975 on the standards and procedures to be adopted in storing seeds for long-term and medium-term conservation. In order to further this work, the IBPGR established a Working Group which met in Rome during 1976 to investigate the engineering, design and constructional aspects of economical seed storage facilities. Their report[1], published in December 1976, has been in considerable demand and is now out of print. In the meanwhile there have been a number of scientific and technical developments in seed storage and more thought has been given to the operation and design of seed stores for genetic conservation. Moreover, economic circumstances have changed since 1976. All of this suggested the need for revision. While basing this publication on the original report, we have taken the opportunity of providing more detail and giving greater attention to medium-term as well as long-term storage. We have also given attention to the use of deep-freeze chests for dealing with small collections, or as a temporary measure for dealing with material while a larger facility is being built.

Estimates of costs are provided at 1981 prices. These are currently being held down by the economic recession and are expected to rise at rates greater than inflation should the world economy recover.

The draft of this report was sent for comment to members of the original Working Group, to members of the IBPGR *ad hoc* Seed Storage Committee (the composition of both is shown in Appendix 1) and also to Professor L. Kåhre (Director, Swedish State Seed Testing Station, Sweden) and Dr. D. Giacometti (Director, CENARGEN, Brazil). We received many useful comments, most of which have been incorporated. However, since some suggestions have not been included the authors must take responsibility for any shortcomings in the report which follows.

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[1] IBPGR, 1976. Report of IBPGR Working Group on Engineering, Design and Cost Aspects of Long-term Seed Storage Facilities 19 pp, International Board for Plant Genetic Resources, Rome.

Seed storage behaviour

1. Seeds can be classified into two distinct groups according to their storage physiology.

The first group is described as 'orthodox'. It includes the majority of arable and horticultural crop species. In this group seed ageing, which ultimately results in seed death, occurs as a function not only of time but also of temperature and moisture content. Consequently it is possible to influence the survival period of seeds of these species by controlling the seed storage environment. Essentially the lower the temperature and the lower the moisture content (at least down to 5%) the greater the longevity.

The second group is described as 'recalcitrant'. It includes many important tropical plantation crops, many tropical fruits, and a number of timber species of both the tropics and of the temperate latitudes. In contrast to orthodox seeds, the rate of deterioration of recalcitrant seeds is less easy to control because they are damaged by drying and since they cannot be dried neither can they be cooled to sub-zero temperatures because they would then be killed by freezing injury resulting from ice formation. Furthermore many of the tropical recalcitrant species cannot be cooled to temperatures in the region of 10°C or less since they are subject to chilling injury. At present most recalcitrant seeds can only be stored for a few weeks or months without loss of viability. Short-term storage methods usually involve treating the seeds with a fungicide and keeping them moist with access to oxygen. This can be achieved by storing in thin polythene bags in a moist, inert medium such as charcoal or sawdust.

2. Since there are no methods available yet for more than short-term storage of recalcitrant seeds, other methods of conservation have to be relied on for species in this group[1]. Consequently this report is concerned entirely with the storage of orthodox seeds.

3. Although the distinction between orthodox and recalcitrant seeds would appear to be reasonably clear, mistakes in classification have occasionally been made. For example, when the first IBPGR report on the design of seed storage facilities was published in 1976 it was thought that citrus seeds were recalcitrant, but it is now known that they are orthodox and can be stored satisfactorily at low temperatures and low moisture contents. A revised and extended list of species indicating storage and other characteristics is included in Appendix 2 of this report. In some cases it will be seen that the classification is tentative and may have to be revised as further information becomes available. **See also page 64.**

Types of seed collections

4. Genetic resources centres take responsibility for one or more of the following collections:
   (i) base collections for long-term conservation,
   (ii) active collections for (a) medium-term conservation, (b) regeneration, (c) multiplication and distribution, (d) evaluation and (e) documentation,
   (iii) duplicate collections (of base collections) for long-term conservation which are housed for security in different locations from the corresponding base collections.

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[1] M.W. King and E.H. Roberts, 1979. The Storage of Recalcitrant Seeds - Achievements and Possible Approaches. 96 pp, International Board for Plant Genetic Resources, Rome.

Breeders' working collections are regarded as outside the framework of genetic resources centres, but use of genetic resources by breeders will generate information which contributes to evaluation and documentation. (The types of collections and their synonyms are described more fully in the glossary, Appendix 12.)

5. The long-term storage methods described below are suitable for base and duplicate collections. There is no special reason why active collections or breeders' collections should not also be stored under long-term storage conditions. However, apart from expense, other operational factors discussed below suggest that medium-term storage systems may sometimes be more appropriate for active collections.

Conditions for long-term storage

6. The longevity of 'orthodox' seed may be dramatically improved by controlling the storage environment because the increase in longevity with decrease in either temperature or moisture content is approximately exponential - see the seed viability nomographs (Figures 4.1 and 4.2, Appendix 4). The 1976 Working Group accepted as a general recommendation the proposed Preferred Standards for long-term seed storage installations as included in Appendix IV (revised July, 1975) of the Report of the Sixth Session of the FAO Panel of Experts on Plant Exploration and Introduction. These preferred standards specify storage at -18°C, or less, in air-tight containers at a seed moisture content of 5 ± 1% (wb [wet basis]). In fact -18°C (or 0°F) is an arbitrary temperature. Most equipment will operate economically at -20°C and this temperature has been adopted for many calculations throughout this report. **See also page 64.**

7. Seed moisture content during storage may be controlled in two ways. The method recommended for the preferred conditions of long-term storage is to dry the seed to about 5% moisture content and then maintain that moisture content by keeping the accessions in hermetic containers. The alternative to hermetic storage would be to use open containers and control the relative humidity of the atmosphere in the coldroom, since there is an equilibrium relationship between the relative humidity of the air and seed moisture content (Appendix 3). However to achieve a satisfactory relative humidity for this purpose (i.e. approximately 10 to 15% rh [relative humidity]) is difficult at sub-zero temperatures and can be prohibitively expensive. In addition, although the hygroscopic characteristics of seeds have been extensively investigated at near ambient conditions, the relationships are less well understood at sub-zero temperatures. Most important, though, is the fact that hermetic containers provide a safer method of controlling moisture content for, if refrigeration equipment should fail, the seeds may take longer to warm up than the store air and consequently, if the seeds are in open containers, water might in certain circumstances condense on them and result in a rapid rise in moisture content (Appendix 3).

8. These preferred standards provided storage conditions which are superior to those previously in general use. The reason for specifying these standards was that increasing the storage period by improving the storage environment would reduce the frequency at which accessions would need to be both monitored and regenerated (Appendix 4), thereby reducing costs and also the difficulties and dangers of regeneration, without greatly affecting the capital and running costs as compared with less satisfactory stores (Appendix 9).

9. The simplicity of the preferred conditions, i.e. a single environment for all orthodox species, provides the advantage that a single store can accommodate very many different species. Nevertheless in a given environment the longevity of seed of different species differs greatly (Appendix 4).

10. The 1976 Working Group suggested that the temperature standard could be relaxed to -10°C if the seed bank was restricted to a few species with good storage characteristics such as the common cereals. Accordingly any store which operates at -10°C or less has been listed as a long-term store (Appendix 12). Present indications suggest that accessions stored at -10°C will require monitoring and regenerating twice as frequently as those at -18°C. Consequently it is recommended that new facilities should adopt -18°C or less, because any saving on capital and running costs of the store will be outweighed in the long-term by the increased staffing and costs required for these increased frequencies. It is suggested that -10°C only be adopted where there are constructional constraints in converting existing buildings to long-term seed stores or when refrigeration equipment is unable to maintain -18°C. **See also page 64.**

11. For a few 'orthodox' species with inherently very poor storage characteristics it may prove advisable to improve upon the preferred conditions of storage. The simplest procedure would be to reduce seed moisture content for these species still further and then store them hermetically at the preferred temperature, since this would only require the one coldroom. For example true seed of potato can be easily dried to 2.5% moisture content (e.g. in the presence of regularly regenerated silica gel) and this results in an approximate tenfold increase in subsequent longevity compared to seed at 5% moisture content. In some species, however, damage may be caused by excessive drying which results in a failure to germinate normally (Appendix 4) and, if the use of ultra-low moisture contents is to be adopted, it is important to check the response of species first if this is not already known. The second approach, not excluded by the prescription of the preferred conditions, is to reduce the temperature well below -18°C. The extreme limit currently being investigated, most notably by the National Seed Storage Laboratory at Fort Collins, is storage in liquid nitrogen at -196°C. However no design criteria are provided in this report for such stores because they are still at the developmental stage. The IBPGR ad hoc Advisory Committee on Seed Storage wishes to see a protocol developed from pilot projects and reliability confirmed before recommending liquid nitrogen storage for general adoption in certain species. **See also page 64.**

Conditions for medium-term storage

12. The accession size of active collections is likely to be larger than for base collections and the rate of depletion is likely to be more rapid since it is from these collections that material is distributed for evaluation and breeding. Regeneration may have to be instituted more often from depletion of stocks in this way than because of loss of viability. Accordingly there is not the same pressure to extend longevity as there is within base collections. Thus many active collections are housed at temperatures between 0° and 10°C and stores of this kind have been classified as medium-term stores (Appendix 12). The recommended seed moisture content can be controlled either by using hermetic containers or, if samples are withdrawn more frequently, it may be considered more convenient to use unsealed containers and control air relative humidity (although running costs are comparatively high - Appendix 9). If the latter policy is adopted then the lower the relative humidity, at least down to 15% rh, the better, but a wide variation in moisture content must be expected both between and, to a lesser extent, within species. This will require direct monitoring of seed moisture content - which may consume relatively large quantities of seed (Appendix 5), although to avoid regularly monitoring seed moisture content some gene banks add a coloured self-indicating dehydration agent to each container - which is then regularly checked and replaced. If this technique is used care should be taken that the relative humidity at which the indicator changes colour is known. (The

coloured indicator can be mixed with the cheaper, non-indicating dehydration agent in a 1:10 ratio to reduce costs.) The most commonly used indicator is silica gel containing cobalt chloride. This begins to lose its dark blue colour at about 15% rh, becomes colourless at about 30% rh and begins to turn pink at about 45% rh. An equation is provided (Appendix 3) as a rough guide to the seed moisture contents to be expected at various relative humidity values. Alternatively, if hermetic containers are used then there will be no need to control the relative humidity of the store and seed moisture content need not be monitored so frequently.

13. Sufficient data are not yet available to indicate the periods of viability to be expected for many crops but the nomographs for barley (which has good storage characteristics) and onion (which has poor storage characteristics) will provide some guidance as to the range of storage periods likely to be obtained in various types of store (Appendix 4).

Drying seed and determining moisture content

14. To satisfy the preferred storage conditions it will normally be necessary to dry accessions to 4 - 6% moisture content (wb). Thus the moisture content of accessions on receipt must be determined to ascertain whether drying is necessary, and if so to determine the loss in moisture that is required. Since different moisture determination techniques give different results, it is recommended that the oven-drying methods prescribed by the International Seed Testing Association (ISTA) are adopted. These are discussed in Appendix 5.

15. For many species a moisture content of 5% is considerably less than that of seed in commercial practice. Also the moisture content of accessions when received at the seed bank is likely to vary greatly between species (compare small grained cereal seed with that from tree fruits). Consequently the techniques, equipment and recommendations that are appropriate to commercial practice are not necessarily the most suitable for seed banks.

16. Seed bank staff require a seed drying procedure that is not only benign to the seed, but that is also simple to operate and apply to the many different seed lots of the many different species which many seed banks have to handle. Recommendations for hot-air drying techniques vary with the design of the equipment, the species and the initial seed moisture content. They are often inappropriate to poor quality and heterogeneous seed lots, and are difficult to operate under humid tropical conditions unless combined with dehumidification. Thus no single hot-air drying technique can be recommended as a standard procedure for all seed banks. Because of this complex situation and to help minimise both the gene bank workload and loss of viability during drying, equations and nomograms which show the inter-relationships between various aspects of the drying environment are provided in Appendix 3.

17. The simplest solution to the problem of drying seed to 5% moisture content is to provide a drying room maintained at about 15°C and 10-15% rh with good air recirculation. This environment can be achieved by an air dehumidifier (sorption-type) with refrigeration to lower the temperature and remove the heat generated by the air dehumidifier. Refrigeration dehumidifiers are unsuitable for this purpose. This, and alternative equipment, is discussed in Appendix 3. Seeds are exposed to this environment in thin layers on open trays. This solution to the drying problem has been adopted by several seed banks, e.g. at the Royal Botanic

Gardens, Kew at Wakehurst Place, and at the National Vegetable Research Station at Wellesbourne, UK. A similar solution is used at the Nordic Gene Bank, Lund, Sweden, but there accessions are dried within thin-woven cotton bags in order to prevent the accidental mixing of accessions that might occur with open trays. This is a satisfactory technique but, unless the bags are force ventilated, the time taken to dry the seed will be considerably increased. For many species (e.g. the small seeded vegetables) in open trays, drying to below 6% occurs fairly rapidly over about ten days. However for larger seeds, e.g. maize and beans, drying is much slower and a longer period of exposure is necessary. Table 1 provides estimates of the cost of providing drying rooms of different sizes to these specifications. It should be noted that the running costs of these rooms are higher than those of an equivalent size coldroom. This is particularly so in tropical humid climates where it is necessary to provide a series of dehumidified buffer zones to progressively reduce the dewpoint temperature to the desired level.

18. To reduce drying times for the larger seeds an alternative solution is to use a two-stage drying system in which during the first stage the drying room is maintained at 17°C with about 40-45% rh using conventional air-conditioning systems (i.e. refrigeration dehumidifiers, Appendix 3). This would dry high oil-content (e.g. groundnuts) or starchy seeds (e.g. wheat) to about 7% and 12% equilibrium moisture contents (wb) respectively. During the second stage final drying to below 6% moisture content can be achieved in a self-contained drier (sorption type) with through air recirculation but no refrigeration, say at about 30°C with 10-15% rh (Appendix 3).

19. It is advisable with either system that standby dehumidifiers or air-conditioners are available in the event of equipment failure. It is suggested that the weight of accessions is monitored during drying to determine when the required loss in moisture has been achieved (Appendix 3); although when experience has been gained with a particular type of seed, it may be sufficient to dry for a standard time.

Hermetic containers

20. After seed drying, moisture content must be maintained during long-term storage by hermetic containers (para. 7). An alternative, for medium-term stores, is to maintain the coldstore at approximately 10-15% rh and store the seed in open containers (para. 12). Any material which is impermeable to water could be used for hermetic containers. In practice, three types of material are being used and considered suitable: glass, metal and aluminium foil laminates.

21. Glass vials which are sealed by annealing the glass are relatively safe and cheap, having a low unit cost and require no capital investment. They are, however, rather inconvenient to prepare and seal, taking approximately four to ten times the period required to package seed in other containers. Glass jars with screw caps are rather more convenient but the seals are not always perfect. Natural rubber seals are reported to be reasonably reliable (provided they are replaced after opening) but some types of plastic seal show a high failure rate. Cobalt chloride paper or coloured silica gel can be included in the jars to detect an increase in moisture content (para. 12), but this requires regular visual examination of all containers in store. Some glass containers are relatively fragile.

22. Metal cans are relatively convenient. Lacquered cans should be purchased to minimise rust problems,

TABLE 1

COLD ROOM AND DRYING ROOM CAPITAL COSTS (f.o.b. European port, equipment tested and packed, 1981 prices)[1]

SIZE OF COLDSTORE				COSTS[2] US $						DRYING ROOM[3]	
(Internal) Store Vol. m^3	Floor Area[4] m^2	Mobile Shelf Area[5] m^2	Number of Accessions[6]	Coldroom		Mobile Shelving	Coldroom & Shelving			Daily moisture removal rate kg	Costs[7] US $
				temperate	tropical		temperate	tropical	per accession		
50	20	75	6,800- 10,000	17,800	18,690	7,900	25,700	26,600	2.57-3.91	5	10,800-12,000
100	40	163	14,800- 21,700	26,500	28,020	17,360	43,900	45,400	2.02-3.07	10	11,500-18,300
200	80	353	32,300- 47,000	40,370	43,000	37,556	77,900	80,600	1.66-2.50	20	12,400-28,700
300	120	556	50,800- 74,000	52,240	55,920	59,000	111,200	114,900	1.50-2.26	32	12,800-37,800
400	160	767	70,100-102,100	63,000	67,730	81,400	144,400	149,100	1.41-2.13	44	13,200-46,300
500	200	984	90,000-131,000	73,100	78,880	104,500	177,600	183,400	1.36-2.04	56	13,600-54,400

[1] i.e. excluding freight and insurance charges. Consequently it may be necessary to increase these costs by up to 50% for installations within developing countries. Note also that these costs are exclusive of the cost of providing a building to house these facilities (para. 60).

[2] Fully fitted modular coldroom for operation at -20°C with either 125 mm or 150 mm insulation (minimum recommended thicknesses for temperate and tropical locations respectively), two independent refrigeration systems and a standby generator. Costs are provided for tropical location coldrooms with 150 mm thick insulation panels - rather than the 200 mm preferred here (para. 34). Manufacturers are generally reluctant to provide quotations for 200 mm thick panels because there is little demand for this thickness.

[3] Fully fitted insulated (125 mm) modular drying room (floor area 10-30 m^2) with air lock for operation at 10°-20°C with 10%-15% rh and standby generator.

[4] i.e. internal cold store height of 2.5 m.

[5] Assuming eight shelves per mobile shelving rack.

[6] Single accessions are assumed to be contained within a 0.88 l cylindrical metal can (para.26). The number of containers that can be accommodated within a coldstore is dependent upon the number of shelves and the organisation of the containers upon each shelf. The range provided here reflects different layouts. Accessions are easier to retrieve from storage the lower the packing density. The lower and upper estimates relate to freestanding and drawer storage systems respectively, but the mobile shelving costs are based on the lower estimate of packing density.

[7] The upper price within each range includes an additional (standby) rotary sorption air drier and an additional (standby) refrigeration system. In addition, for the larger cold stores, the range of costs reflects the range in drying room sizes.

unless the cans are made from aluminium. Screw-cap, or similar, cans have the advantage, as do glass jars, of being re-usable, but they may not always form perfect seals; moreover any indicator of leakage inside the container can only be inspected by opening the container. Nevertheless, they have been used satisfactorily in a number of seed banks. Seamed, 'three piece' or 'two piece', cans are both safe and convenient, although they require the capital investment of a double-seamer to seal the cans (about US $3,000) and careful adjustment and inspection (of seam thickness using a micrometer) to ensure perfect seals. However the cans themselves are cheap.

23. Laminated aluminium foil packets are very convenient. They are inexpensive, occupy little storeroom space before use, can be cut to various sizes, and be re-used after sampling. They do, however, require the capital investment of a sealing machine (US $1,000) and careful adjustment and inspection to ensure adequate seals. Moreover the packets must have a robust specification, be obtained from a reliable manufacturer and not abused during filling and handling. These points are considered in detail in Appendix 6.

The size of accessions and the volume of the coldroom

24. The provisional recommendations of the 1976 report were that the size of each accession in base collections should, if possible, not be less than about 4,000 seeds for genetically uniform material (e.g. established varieties of self-pollinating crops, or F1 hybrids) and 12,000 seeds for heterogeneous material, and 1,000 and 3,000 seeds respectively in duplicate collections at other centres. The ad hoc Seed Storage Committee sees no reason to alter these recommendations. However, for very large seeds it may be necessary to reduce these quantities.

25. The weight of, and the volume occupied by seeds varies greatly between species, and to a lesser extent within species. In Appendix 2 we have provided some information on these values for many species, including most of those on IBPGR Priority Lists, which may assist in planning the volume required for each accession. Obviously the total number of accessions a given coldstore can hold will depend on the species to be stored and the proportion of the volume of the store to be provided to each species.

26. Wheat is a convenient species to provide an example for planning store volumes, since many other common cereals have similar 1,000 seed volumes. 12,000 wheat seeds occupy about 600 ml (Appendix 2). This volume can be readily accommodated in a cylindrical metal can of approximately 880 ml internal volume (103 mm diameter x 118 mm height). In a store one container would occupy a square prism of almost 1.3 l, with a maximum packing density of 90 cans per m^2 of shelf area.

27. However it is not possible to fill the internal volume of the coldstore with containers. First it is essential to allow sufficient cold air circulation between the inner surfaces of the coldroom and its contents to prevent unacceptable temperature gradients. There should be a minimum gap of 20 cm between the coldroom walls and shelving, 10 cm between the floor and the lowest shelf and a 50 cm gap at the top of shelves to allow, in addition to cold air circulation, for lighting fittings, and a ceiling-mounted air cooler (if installed). Second it is necessary to provide a shelving system that enables accessions to be easily located and removed from store without having to displace other accessions. It follows that the choice and layout of the shelving system determines the total number of accessions that can be accommodated within a

given store. Installing mobile shelving sytems (fabricated from steel protected by a PVC coating or stove enamelled paint), as used for example in libraries and factory storerooms, in place of static shelving, can double the number of samples which, for example, a 100 m^3 store could contain, without radically changing the refrigeration requirement (Appendix 8). Accordingly, it is recommended that manually propelled mobile shelving stacks be installed in long-term cold stores.

28. Access to material in mobile shelving is gained by providing two passageways (100-120 cm wide) at right angles to each other. The shelves (30-50 cm from front to back) should be capable of withstanding loads of 150-200 kg/m^2 and can be assembled to form trays with a raised outer edge to retain samples when the shelves are moved. The vertical spacing of individual shelves must be arranged according to container size and operator reach. The maximum convenient height is 2 m. A library kickstool will ease access to the upper shelves. Drawers or removable boxes should be incorporated, particularly for small-size accessions. These will help the orderly incorporation of accessions into, and any possible emergency evacuation of accessions out of the store, as well as preventing accessions from being dislodged when shelves are moved. Mobile shelving systems normally include a false floor (loading 1,500 kg/m^2) recessed for the shelf unit tracks and fitted with end stops to prevent wall damage. A similar reinforcement should be constructed for free-standing static shelving because of the extremely high 'point contact' loads provided by the shelf framing, which could otherwise damage the floor insulation and underfloor heating system (para. 36). This can be fabricated from embossed aluminium plates or a layer of granolithic cement.

29. Whether mobile or static shelves are installed, it is important that they do not block cold air circulation. Consequently if backplates or sideplates are fitted to shelves they should be perforated.

30. A procedure is shown in Appendix 8 as an example of the calculations necessary for providing the information in Table 1. It is indicative of the considerations required to design a store of any size.

31. As a procedure for estimating the cold storage space required when a gene bank is being established, it is suggested that an attempt is made to assess the probable number of accessions which are expected over the next, say, 10 years, and base the storage requirement on this estimate. While it is more economical to construct single large coldrooms of the required capacity than to provide the equivalent volume in several smaller coldrooms (Table 1), dividing a collection between two coldrooms does give additional protection against equipment failure. In any case it would be advisable to design the layout of the first coldroom and its associated facilities so that a second coldroom could be added in the future without difficulty, should it be required. Presumably the individual foreseeable requirements for the majority of gene banks will be between about 7,000-70,000 accessions and will thus fall within the coldroom size range 50-300 m^3 with a capital cost for coldroom and shelving between about US $4 to US $1.5 per accession respectively (Table 1).

Coldroom design: 1. Design standards

32. It is recommended that specifications for tender be based on one of the major national codes and standards. For example, the Codes and Standards of the American Society of Heating and Refrigeration Engineers (ASHRE) are helpful and widely accepted.

Coldroom design: 2. Coldroom thermal insulation and construction

33. It is recommended that the coldroom is constructed from factory prefabricated interlocking insulated panels. Those made from painted galvanised steel sheeting, with sealed lap joints, containing polyurethane foam (long-term thermal conductivity 0.017-0.023 W/m per 1°C) mixed with a fire retardant, are preferred because such panels are competitively mass produced with strict quality control. Moreover, the use of pre-formed insulating panels avoids any health hazards which are associated with in situ polyurethane foam production. Pre-formed insulating panels containing polyurethane foam present no hazard to health except in the event of fire (para. 67). Modular coldrooms, constructed from these panels, are self-supporting up to a width of 7 m and are therefore less vulnerable to earthquake damage. They can be dismantled for removal or repair work. The protective steel casing is resistant to vermin and impervious to water vapour ingress. Fastenings (bolts and screws) should not be made right through the steel facings and tightened, for polyurethane foam is a viscoelastic material which will creep in time so that the screwed fixing will become sloppy, thereby allowing ingress of the atmosphere and thus water vapour. It is essential that ingress of moisture into the store is minimised in order to prevent excessive icing of the evaporator, to maintain the integrity of the insulation, and to prevent rusting of any ferrous materials (e.g. shelving or metal containers). In other words the coldroom must be properly sealed. (A simple coldroom pressure test procedure for testing air infiltration is given in Appendix 7.) Rusting may occur if the air exceeds about 65% rh. Coldrooms will operate within the range 25-45% rh if fitted with dehumidified air-locks (para. 48) controlled at 10-15% rh. If such an air-lock is provided it is not necessary to use stainless steel fittings within the coldroom. Permanent coldrooms may be constructed from multiple layers of slab stock insulation secured by vapour seal bonding with external and internal sheet metal cladding provided that they are constructed to the same high standard; but they can be more costly to erect.

34. It is recommended that the appropriate insulation thickness be selected on the basis of "least cost operation" (Appendix 9), because the accumulated running costs of a badly insulated coldroom over a number of years, may well exceed the original capital expenditure. Coldroom panels should be preferably 200 mm (and not less than 150 mm) thick for operation at -20°C under tropical applications, but proportionally less for other climates, according to the difference between ambient and coldroom temperature (Appendix 9). It is recommended that modular coldrooms be factory tested to certify the standard of thermal insulation (Appendix 7).

35. Coldroom doors (100-120 cm wide) either hinged or sliding must have heated seals to prevent local icing, and preferably be fitted with a triple glazed inspection window (50 cm x 50 cm). Similarly, medium-term stores which operate under tropical conditions without a dehumidified air-lock should be fitted with heated door seals to prevent local condensation. To limit the flow of air out of the coldroom when the door is opened (due to the differences in air density) a nylon-reinforced transparent PVC (cold store grade) strip curtain should be suspended from the coldroom door frame. Air curtains offer no special advantage over strip PVC curtains for this application. A small gap should be made to allow inspection through the coldroom door window. Precautionary measures must be taken to prevent air pressure, produced by either cooling ambient air or opening/closing the hinged coldroom door, from causing structural damage. This problem may be resolved, for example, by attaching a non-recessed flexible seal to the underside of the coldroom door (small stores) or by fitting a heated ventilator port to equalise air pressures.

36. Coldrooms constructed at ground level and operating at temperatures below 0°C in temperate regions must be fitted with underfloor heating systems to prevent frost heave, i.e. freezing of ground water which expands to distort and damage the coldroom floor. Various heating methods, ranging from underfloor ventilation to circulating oil through a grid of buried pipes, are equally satisfactory, providing they maintain a temperature of 4°C immediately under the insulated floor. It is recommended that a simple thermostatically controlled low voltage electric cable heating circuit, formed by laying a network of PVC coated heavy gauge stainless steel wire (3.25 mm diameter), laid in the concrete floor supporting a coldroom, be used. Provision must be made beneath the floor for more than one thermometer/temperature sensor pocket for monitoring and alarm purposes. A heater load of 5-10 W/m^2 of floor area (depending on soil temperature and locality) is adequate. However, neither small coldrooms (less than 50 m^3) built on raised frames nor larger coldrooms operating under tropical conditions require underfloor heating. Special action must be taken when metal stanchions (or large bolts) penetrate the coldroom floor into an unheated, unsealed concrete floor on bare earth. The high thermal conductivity of the metal plus a small temperature gradient would produce local ground freezing, capable of moving the heavy column upwards to cause structural damage, even though the stanchion is well insulated. Once again, the problem can be solved by placing a heater circuit around the base of each stanchion or large bolt. Similarly, although a false floor is essential to distribute the shelf load (para. 28), great care must be taken not to damage the frost heave protection circuits during installation.

Coldroom design: 3. Refrigeration plant

37. In view of its well tried reliability and ease of maintenance, it is recommended that conventional direct or indirect vapour-compression based refrigeration cycles are used. Such systems can be obtained as package units, factory tested for tropical or other environments. Cheaper, hermetically sealed refrigeration units may be used for small installations but their motors can be damaged if the mains voltage changes by more than ± 6-10% of the declared supply rating. Open type units (belt driven compressor with separate electric motor) are recommended because they can be easily repaired and can be progressively replaced. Refrigerants R12 and R22 or R506 are recommended for medium- and long-term storage facilities respectively. Freely ventilated air-cooled condensers (derate by 1% per 1°C rise above maximum operational temperature), which dissipate heat removed from the coldroom during refrigeration, are recommended because water-cooled condensers are liable to failure should the water supply be interrupted or contaminated by impurities. Coldroom air-coolers, incorporating the refrigeration system evaporator coil, and air recirculation fan and drainage tray, must be fitted with removable electric heater elements for defrosting if the control air temperature is below 2°C. (Incidentally, air coolers fitted in medium-term stores operating at 10°-15°C and 40% rh may contain a reheat coil for humidity control, but the evaporator may require defrosting.) The drain pipe (protected within the coldroom by a low voltage heater tape - 45 W/m run) should be connected externally to a water trap, leading to a sewer, by a length of transparent plastic tubing, arranged to prevent noxious gases from being drawn into the coldroom. The effectiveness of the cooling system can be monitored by weighing the condensate melted during successive defrost cycles. The amount of moisture entering a coldroom fitted with a dehumidified air-lock is small.

38. It is recommended that the defrost cycle be time-switch initiated and stopped by a thermostat to keep the heating period to a minimum. The refrigeration unit must be switched off during defrosting and the

air-cooler fans only restarted after the residual moisture has frozen on the evaporator, otherwise water will be blown on to the shelving. The air-cooler unit can be wired to operate only at the maximum ventilating rate when a coldroom is unoccupied; this precaution reduces the air 'chilling factor', making coldroom work more tolerable. The air recirculation rate must not be less than 5-10 air changes/h (hour) and the refrigeration thermostat should be positioned centrally to control the return air temperature to within ± 1°C of the thermostat setting. High and low refrigerant pressure gauges and a liquid line sight glasses are useful for monitoring refrigeration performance. High and low temperature alarm thermostats are also desirable.

39. Each coldroom should be provided with two independent refrigeration systems sized for a 16 h maximum design load duty. Allowance must be made for any additional refrigeration load - such as sorption-type air dehumidifiers. The independent refrigeration systems should be run alternately, for monthly periods, to ensure they remain in good working order. Refrigeration units larger than 1 kW rating should not be mounted on the coldroom structure, because long-term vibration may damage the insulated panels.

Electricity supply and maintaining gene bank services in emergency

40. Major gene bank installations require a three-phase alternating current power supply to cope with the high starting current of refrigeration equipment (use the locked-rotor current load to estimate this), but small walk-in cold-rooms and deep-freeze cabinets are usually designed for single-phase operation. It is vitally important that the local mains voltage, frequency, and its fluctuations be measured in order that this information can be clearly specified when ordering equipment - so as to prevent electric motor failure. Any plans for future expansion of gene bank activities likely to affect demand must be taken into account when calculating the total demand on the local power distribution network. The total connected load ranges from 20-100 kVA (3 times the actual maximum connected load) for coldroom sizes recommended in this report (see Table 1), but additional air conditioning plant would increase this estimate.

41. A useful precaution is to install a battery power supply (12 V with mains powered charger) within the coldstore (para. 65) and adjacent areas for emergency lighting during disruptions to the power supply.

42. Standby power generator sets (diesel) are required to maintain the minimum gene bank services - refrigeration, drying and lighting. Fuel consumption is approximately 0.142 kg/kWh of electricity generated. A 6 month reserve fuel supply should be maintained (but not adjacent to the store). The room, or building, housing the standby generator set(s) should be provided with emergency lighting (para. 41). Manual or automatic starting is available. However, manual starting permits sequential load switching on a priority basis, with due regard to existing equipment failures and electrical short circuits (blown fuses etc.), which might otherwise overload, damage or shut down automatic plant. Diesel generator sets are usually designed to work in ambient temperatures up to 40°C. Above this temperature the output must be derated by 0.5% per 1°C. Conversely, in cool climates a sump heater may be required during cold weather. A service hour counter should be fitted and plant maintenance undertaken at the intervals recommended by the manufacturers. Tractor powered generators may be used in an emergency.

43. The time available for rectifying major refrigeration or standby generator faults is related to the 'coldroom time constant', i.e. the time taken for the store temperature to rise through 63% of the range

from operating to ambient temperature. The coldroom time constant is dependent upon the insulation thickness and the coldroom internal thermal mass. For instance, a half filled 100 m^3 capacity coldroom, constructed from 125 mm polyurethane panels, would take approximately four days to heat from -20°C to 0°C when the ambient temperature averages 20°C. A completely full store would take twice as long to reheat. But this loss of temperature control would be no worse than a comparable delay before putting the seeds into storage after drying. This is because there is no evidence that temperature change itself affects viability. In fact all investigations carried out on the effect of temperature change on seed viability indicate that it is the integral of temperature which is important. Because of this temporary short-term failures will have no significant effect on storage life, providing they do not add up to a significant period at ambient temperature. Thus although standby generators are required to guard against longer-term failures, it may not be necessary to take any further precautions.

44. Nevertheless, if various direct refrigerants (solid and liquid carbon dioxide, liquid air or liquid nitrogen) are available locally it might be worth considering a further standby system which utilises one of these for emergency cooling purposes. Liquid nitrogen is usually the least expensive of the four alternatives and the refrigerant most suitable for direct refrigeration of coldrooms and deep freeze cabinets. Simple automatic control equipment (for operation during a power failure) is available to maintain a temperature of -20°C. The control temperature is maintained by injecting the requisite amount of liquid nitrogen into the storage space to give an equivalent refrigeration effect of 113 W/kg (-20°C). However, liquid nitrogen expands by 610 times during vaporisation. Consequently a suitable air/nitrogen vent must be fitted to prevent coldroom structural damage from occurring as a result of increased pressure within the store. In addition, the nitrogen vapour will diminish the concentration of oxygen and therefore the coldroom must be fully ventilated with fresh air before staff enter the cold store (see also para. 63).

Temporary or alternative seed storage facilities

45. The cold store design described here (paras. 33-39) is conventional and similar to those which many gene banks already have in operation throughout the world. However in certain circumstances, alternative facilities might be required. For example to provide temporary facilities during major collecting missions or during the construction of a conventional store; or to provide a relatively inexpensive storage facility for a very small collection; or to provide a store with lower power requirements. These special requirements are considered in Appendix 10.

Monitoring gene bank environments

46. It is advisable that the environments - temperature and relative humidity - within both the coldstore and the drying room are regularly monitored and logged. It is recommended that each coldroom should be provided with two certificated mercury-in-glass thermometers covering the storage-ambient temperature range. These should be mounted within the main shelving area. Similarly, a further two thermometers should be mounted in the main shelving area of the drying room. Secondary, remote, temperature monitoring can be done by multi-channel strip chart recorders (US $1,500-2,500), or reliable digital recorders operating on the emergency, or separate, low voltage power supply with calibrated thermocouple junction or platinum resistance

thermometers located to detect coldroom temperature extremes. An output for the underfloor temperature sensor (para. 36) could also be provided on this recorder.

47. Relative humidity - in medium-term cold stores, drying rooms and any other area in which relative humidity control is necessary - can be measured by many different empirical methods. The linear measurement of hygroscopic materials, unaspirated wet bulb thermometers and the changing electrical properties of various substances are only suitable for monitoring purposes. Observing dew formation on cooled mirrors (simple dew-point apparatus) or the expansion of an air/water vapour mixture are recommended for calibration purposes (± 1°C dew-point temperature). A sling psychrometer (whirling hygrometers) or Assimann hygrometer (or similar meteorological instrument) are very satisfactory for drying room conditions above 4°C providing both the wet- and dry-bulb thermometers are fully shielded from radiant heat sources. Great care must be taken to ensure that the appropriate conversion or calibration tables are used - particularly for sub-zero temperatures. The relative humidity can be determined from (aspirated) wet- and dry-bulb thermometer readings using a psychrometric chart (Appendix 3).

Air-lock

48. It is recommended that all sizes of walk-in coldrooms with over 20 m^3 storage capacity, be fitted with air-locks to prevent the incursion of warm moist air into the cold store(s). Air-locks comprise a cubicle enclosing the coldroom entrance with a second outer door (preferably at right angles to the first) and are constructed from a suitable vapour sealed (i.e. moisture-proof) insulating material. The outer door must always be closed before entering the main coldroom. It is advisable to provide side wall and door windows (50 cm x 50 cm). The air-lock air space may be dehumidified to the coldroom dew-point temperature (Appendix 3) and if this precaution is taken it has a secondary advantage; it can be used to allow frozen accessions to reheat to the ambient temperature without moisture condensing on containers (para. 20). A floor area of at least 6 m^2 is essential so that the coldroom door can be opened without obstructing a trolley and personnel. Access through the air-lock must also be sufficient to enable the largest items within the cold store, e.g. shelves, to be replaced.

49. A double air-lock can be provided by building the drying room and/or dehumidified packaging room adjacent to the coldroom entrance. This would be particularly advantageous in humid climates or when coldrooms are entered frequently. The air-lock can be repositioned to the side of an enlarged insulated room with one wall (two if placed between two coldrooms) formed by the coldroom front. This arrangement saves construction costs. It can be of modular construction and designed and built to form a single general purpose coldroom-drying room-packaging room complex, and is particularly suitable for small to medium size gene banks. The area of, and the equipment contained within this room will depend upon its function(s) but is likely to be between 12-30 m^2. A cabinet for coldroom protective clothing (para. 63) is required. The air recirculation rate in this dehumidified room must be not less than 10 air changes per hour, and preferably at 2 mm wg (water gauge) above atmospheric pressure. Since staff may work for considerable periods in this environment adequate artificial lighting is essential (500 lux at work level). In addition the IBPGR ad hoc committee on Seed Storage has recommended that windows (double glazed) be provided to improve the working environment.

Ancillary facilities

50. The coldroom is only one element in the provision of a long-term seed storage facility. Further elements are required to support and service the store. These are indicated in Appendix 11. Before they are placed in cold storage, accessions must be registered, cleaned, dried, packaged and assessed. During storage accessions will require temporary removal from storage to enable samples to be extracted for monitoring germination (and possibly moisture content) tests, and to satisfy requests for material and ultimately for regeneration (leaving a certain amount of seed in store in case of accidents if the regeneration procedure fails). In some institutes where seed banks are being established there may be existing laboratories, offices and equipment which can provide the necessary facilities. In other cases all the appropriate facilities may have to be built.

51. Although seed should be dried to 5% moisture content as soon as possible after receipt, in order to avoid unwittingly ageing the seed before it is stored, there could be some unavoidable delay. First material will need to be inspected, accepted, prepared (e.g. threshed), and cleaned (i.e. remove chaff, dirt, broken seeds and weed seeds from the bulk). Secondly, bottlenecks in the input procedure may delay the start of drying. It is therefore advisable either to control the environment in the cleaning and threshing areas to provide a temporary storage facility, or to provide a separate room for this purpose (Appendix 11). It may be necessary to provide a fumigation chamber in this area. The capacity of the drying room must be sufficient to enable large numbers of accessions to be received during the initial commissioning period. A preliminary design estimate would be to allow for an annual drying load of 15-30% by weight of the maximum cold store capacity.

52. The area required for packaging will depend on throughput but is unlikely to be less than 12 m^2, and there is a need to allow space for one or more balances. If possible, the relative humidity of the packaging room should be maintained at the same level as the drying room, otherwise seed moisture content will rise during packaging. This is a particular problem with very small seed, e.g. sugar cane.

53. In a small gene bank, the cleaning, drying and packaging operations could all take place in a single low humidity room, preferably adjacent to the cold store (para. 49). The arrangement of rooms to provide these facilities are discussed in Appendix 11.

54. Laboratory space will depend upon the number of staff working within the laboratory and the amount of equipment located there. The following list is provided as an indication of the minimum area required in a seed laboratory for the equipment and its associated access.

		m^2
(a)	Two people will require a floor area of 2 m x 3 m for benchspace.	6
(b)	Further bench area for miscellaneous use.	3
(c)	A (three) balance bench with space for desiccators.	3
(d)	An oven and grinder for moisture content determinations.	3
(e)	Six incubators (three for germination testing, two for seed health testing, and a domestic refrigerator).	12
(f)	An aseptic room for preparing seed health tests.	4
(g)	Washing-up area with autoclave and drying cabinet.	8

	m²
(h) Seed humidification chamber (see Appendix 3)	2
(i) Fume cupboard	2
(j) Glassware cabinets, etc.	3
Total minimum area of laboratory	46
or 40 m² if no provision is made for seed health testing.	

55. The cost of providing apparatus for this laboratory would be about US $14,000.

56. An X-ray machine for detecting empty seeds would be useful, indeed essential if certain seeds (e.g. many grass and tree species) are to be tested. Some designs of equipment require a separate, dark room (at least 4 m²). Other designs can be operated (safely) within the laboratory, but in either case a photographic dark room is required unless polaroid-type film is used or no permanent record is required. Suitable, basic equipment will cost upwards of US $12,000, though additional, optional, equipment might double this cost.

57. If two of the incubators are replaced by two walk-in germinators (each 2 m x 2 m), the domestic refrigerator (for seed dormancy breaking treatments) is replaced with a walk-in coldroom at about 5°C (2 m x 2 m), a growth room is provided for seed health investigations beyond the early stages of germination (2 m x 3 m), and bench space is provided for an additional member of staff, the required laboratory area would increase to about 70 m².

58. An office for the seed bank manager can be combined with a records office and need not occupy more than 20 m². The size of general purpose storage rooms will depend upon the choice of seed storage containers. Cans will occupy far more store room space than an equal number of laminated foil packets. Nevertheless a 40 m² provision should be adequate and enable consumable items to be ordered in sufficient quantities in advance of demand. Ten m² should be sufficient for the provision of two lavatories and wash-basins, an emergency shower and a cupboard for cleaning materials. In addition a seminar/coffee room of about 15 m² or more would enable visitors to be accommodated in comfort without disrupting the routine of the gene bank, and provide a basic staff rest facility.

59. In all but the smallest stores a machinery room (external to the building which houses the bank and well ventilated) will be required for the refrigeration plant, air-dehumidifiers and standby generator set(s). The area of this room will need to be approximately 20% of the coldroom floor area, but in any case not less than 10 m².

60. The total area of buildings required for ancillary purposes can now be estimated:

	m²
Machinery room	10-40
Seed drying room	10-30
Seed cleaning and packaging rooms	30-90
Seed testing laboratory	40-70
Offices and records room	20-40
Dark room	4

	m^2
Store room(s)	40
Toilet and service room	10-20
Seminar/rest room	15-30
Total	179-364

In addition up to 20% more floor area may be needed for circulation (e.g. entrance vestibule, corridors). In a small establishment it should be possible to reduce the above allocations without drastically affecting the efficiency of the organisation. The cost of providing a building to house both a prefabricated coldroom and the ancillary facilities will vary widely according to location. As an approximate guide in a number of countries this would be currently about US $600-1,000/m², including the provision of mains services and architects' fees.

61. This suggested provision of facilities makes no allowance for any duties other than those essential for the operation of a base collection within a gene bank. The sequence of operations is outlined in Appendix 11. Some thought should be given to whether further facilities are required, or might be required in the future, e.g. facilities for regeneration, tissue culture or research.

Safety precautions

62. Gene bank planners and operators must aim to minimise installation and operating costs without jeopardising coldroom safety laws and codes of practice. Frequent surveys and alarm system tests are recommended.

63. All persons working at low temperatures must be provided with suitable protective clothing. The in-store working period should be kept to a minimum. Additional ventilation must be provided if the coldroom respired carbon dioxide gas concentration exceeds 0.5%. It is recommended that senior staff be instructed to use gas detection apparatus for direct and indirect refrigerants. For safety, and security, only authorized personnel should be allowed within the cold store.

64. An audible personnel alarm, low voltage type, should be provided for coldroom staff. Alarm switches should be located by every door, at 45 cm above floor level, in case the worker is unable to stand. Fire alarm switches should also be located near fire exit points so that a person can raise the alarm but still have a direct escape route. The alarm, a gong or siren, must have a distinctive tone and should be located where there is always somebody in attendance. The alarm system should be controlled by a key-switch in the charge of a senior security officer. Each low temperature coldroom and drying room should be fitted with a suitable telephone link which, in the former case, can be operated by staff wearing thermal gloves. It is recommended that this communication link to a central office or exchange be used to record when staff enter or leave a coldroom facility. A log book of coldroom work should be kept and used daily.

65. Lighting must be available at all times, with light switches located within the coldroom area. Both normal and emergency - mains and low voltage (battery) - circuits are recommended. A single low wattage safety light (battery powered) can be kept on permanently in each coldroom and positioned above the

entrance, the wiring to be done with fire resistant cable. Emergency lighting must also be provided in the machinery room (paras. 41, 42) to allow emergency repair work to be undertaken. All lighting fittings and electrical motors are potential fire hazards and must be designed for low temperature service and, ideally, protected by separate fuses. Either tungsten filament or fluorescent lamp fittings (designed for low temperature operation) can be used to give an illuminated level of 100 lux at floor level. The respective electrical loading would be aproximately 45 W/m^2 or 18 W/m^2 of total floor area. An exterior red warning lamp, wired in parallel with the coldroom lighting circuit, should be fitted to indicate when the coldroom is occupied. Hand lamps, portable power tools and frost protection equipment must be low voltage (50 V) and all metalwork correctly earthed.

66. Attention to methods of escape from coldrooms is more important than in most other types of building, because of their construction and low temperature. In large stores emergency doors should be installed to give the widest field of escape via at least two routes. The machinery room should have an alternative exit point through a fire resistant external door. Emergency exits should, whenever possible, lead directly to the outside at ground level. External handles can be removed to improve security, with doors secured on the inside for quick release. The main coldroom door should have a safety latch designed to prevent staff from being locked in. Heated door seals should be used for temperature zones below 0°C to prevent icing. It is recommended that coldroom and drying room access doors contain a glazed inspection window (50 cm x 50 cm) to give an unobstructed interior view. (Coldroom [-20°C] windows must be triple glazed.)

67. The selection and use of 'first stage' fire fighting equipment should be made in consultation with local fire brigades and staff instructed accordingly. In particular it should be noted that in a fire synthetic foams - within the coldroom walls, ceiling and floor - will emit both toxic vapour and toxic fumes and thus such a fire should only be tackled by trained personnel wearing breathing apparatus. Water reservoirs may be required in rural areas. Fire extinguishers are essential but must be selected cautiously. Danger exists with water (electrocution) and carbon dioxide (asphyxiation), whereas certain chemicals are poisonous. Dry chemical powder extinguishers are to be recommended and must be positioned near access and emergency doors. Fire-proof blankets and buckets of dry sand can be used to smother small fires, while automatic water sprinkler systems outside the coldroom can protect the coldroom exterior. A major gene bank should have one set of breathing apparatus, with a self-contained compressed air supply, and arrangements must be made for expert training and fire drills.

68. Sufficient space must be allowed around gene bank buildings to give a fire brigade quick access to any point. All water hydrants should be clearly identified and must be compatible with fire brigade hoses. Additional 'fire walls' may be erected as required, particularly when existing buildings are converted for gene bank use.

69. If possible it might be worth considering the possibility of giving accessions a priority rating in case rapid removal from a coldroom is necessary, in which case thought should be given to the position of priority accessions within the store and type of storage container used for them. Alternative, temporary low temperature storage arrangements, typically in other (possibly commercial) cold stores, should be considered and kept under review.

Selection of site

70. In the majority of cases gene banks have been, and will continue to be, sited within, or adjacent to, existing research institutes. Nevertheless, there are a number of important factors which must be considered when selecting a site for a long-term storage facility. Most of these are obvious, but it is prudent to state some of the major factors:

(i) Socially stable area within reach of security personnel;
(ii) Reliability of mains electricity supply and voltage stability;
(iii) Suitability of substratum for foundations, adequate drainage, and absence of flooding;
(iv) Situation away from dangerous chemical or fuel storage areas; and
(v) Easy access to the area where seeds of active collections are threshed, dried and cleaned.

APPENDIX 1. COMPOSITION OF 1976 WORKING PARTY AND 1981 *ad hoc* SEED STORAGE COMMITTEE

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APPENDIX 2. SEED WEIGHT, VOLUME, IBPGR PRIORITY AND STORAGE CHARACTERISTICS

Seed weights and volumes

Where available, information is provided on seed weights and volumes. It is intended that these values should be used as an approximate guide to determine the volume of storage space required for accessions of the orthodox species. Note that in some cases the variation within a species is quite large. This partly reflects the influence of both genotype and provenance on seed size. If the volume (cm^3) occupied by 1,000 seeds (S_v) is unknown it can be estimated from the 1,000 seed weight (W_{1000}, g) if this is known, using the following expression (provided the seeds have no appendages):

$$S_v = q \times W_{1,000}$$

where q is a constant with a value between 1.2-1.5. Thus the number of seeds per litre is approximately

$$= 760{,}000/W_{1,000}$$

It is important to note that the weight of, and less obviously, the volume occupied by 1,000 seeds will vary with the seed moisture content. The solid volume of most seeds is reduced proportionally to the weight loss that occurs during drying. For example, a 50 cc container filled with maize seeds gave seed counts of 70, 75, 87 and 103 at 31, 26, 15 and 9% moisture content (wb) respectively.

IBPGR Priority

The IBPGR has assigned priorities to crop species as a guide to those which require immediate action.[1] The priorities only partly reflect the present or future importance of a particular crop. They are substantially influenced by the amount and quality of existing genetic resources work on it, and the current risk that important genetic resources may soon be lost. This does not, however, exclude collection of wild species and related taxa of no immediate economic importance, where they seem likely to be important for crop improvement.

Four degrees of priority have been used: first priority (1); second priority (2); third priority (3); and lesser priority (4). In addition a further priority (S) has been designated to those crops or groups of crops which the Board wishes to study further before assigning a priority.

Seed storage characteristics

The majority of crop species show orthodox seed storage characteristics (see Glossary). These species have been designated by the letter O in the Table. Some crops are believed to show recalcitrant seed storage behaviour (see Glossary) and are designated by the letter R, although often information is lacking and thus future revision may be required in some cases. The symbols O? and R? denote probably orthodox and probably recalcitrant, respectively, while ? denotes considerable doubt. Recent work at Reading suggests that seeds of certain species, possibly some of those designated O? in the Table, show some but not necessarily all of the characteristics of orthodox seeds; i.e. they can be partially dried and cooled with benefit, but low moisture contents, say less than about 10%, and sub-zero temperatures often appear to be deleterious. As a consequence, while medium-term storage is feasible under particular storage conditions, the IBPGR 'preferred conditions' for long-term storage of orthodox seeds may not be suitable, or at least not suitable for all accessions of some of these species. The two most important crops thought to belong to this category are coffee (Coffea spp.) and oil palm (Elaeis guineensis), both of which were formerly described as recalcitrant - see King and Roberts (1979) - cited at the foot of page 1.

For more information on this topic see:
R.H. Ellis, T.D. Hong and E.H. Roberts, 1990
An intermediate category of seed storage behaviour. I. Coffee.
Journal of Experimental Botany, 41, 1167-1174

[1] IBPGR, 1981. Revised priorities among crops and regions 17 pp, International Board for Plant Genetic Resources, Rome.

Species	Common Name	Approximate 1000 seed weight (g)	Approximate 1000 seed volume (cm^3)	IBPGR Priority	Storage Characteristics
Abelmoschus esculentus	okra	53		2	O
Abelmoschus manihot	okra, aibika	45-60	90	2	O
Abies alba	silver fir	140			O
Abies amabilis	pacific silver fir	40-145			O
Abies balsamea	balsam fir	8			O
Abies concolor	white fir	30-85			O
Abies fraseri	fraser fir	8			O
Abies grandis	grand fir	20-50			O
Abies homolepsis	nikko fir	15			O
Abies lasiocarpa	subalpine fir	12			O
Abies magnifica	california/shasta red fir	77			O
Abies nordmanniana		165			O
Abies procera	noble fir	33-55			O
Abies veitchii	veitch fir	8			O
Abrus precatorius	rosary pea, jumbie bead				O
Abutilion sylvaticum	flowering maple				O
Acacia spp.					O
Acer argutum		15-20			O
Acer campestre	hedge maple				O
Acer cissifolium		60-115			O
Acer crataegifolium					O?
Acer ginnala	amur maple				O
Acer griseum		29			O
Acer japonicum	fullmoon maple				O
Acer macrophyllum	bigleaf maple	55			O
Acer negundo	boxelder, negundo maple	143			R
Acer palmatum	japanese maple	40-70			O
Acer pensylvanicum	striped maple	75			O
Acer platanoides	norway maple	40			O
Acer pseudoplatanus	sycamore maple	100-260			O
Acer rubrum	red maple	65-200			R
Acer rufinerve		20			O
Acer saccharinum	silver maple	400			O
Acer saccharum	sugar maple	333			R
Acer sieboldianum		71			O
Acer spicatum	mountain maple	110			R
Acer tataricum		20			O
Achillea spp.	yarrow	55			O
Aegilops spp.					O
Aeschynomene spp.	joint vetch, ambatch				O
Aesculus hippocastanum	horse chestnut				O
Agathis australis	kauri	4,000-20,000			R
Agathis macrophylla		21			O
Agathis robusta	queensland kauri	230			O
		50			O

Agave spp.					0
Agropyron cristatum	fairway crested wheatgrass	1.5		S	0
Agropyron desertorum	standard crested wheatgrass	2.2-2.5		S	0
Agropyron elongatum	tall wheatgrass	6		S	0
Agropyron intermedium	intermediate/pubescent wheatgrass	5-10		S	0
Agropyron smithii	western wheatgrass	4		S	0
Agropyron spicatum	beardless wheatgrass	3.6		S	0
Agropyron trachycaulum	slender wheatgrass	3.4		S	0
Agrostis canina	velvet bentgrass	0.06		S	0
Agrostis castellana					0
Agrostis curtisii					0
Agrostis gigantea	redtop	0.1		S	0
Agrostis stolonifera	creeping bentgrass	0.07		S	0
Agrostis tenuis	colonial bentgrass	0.07-0.08		S	0
Ailanthus altissima	tree of heaven, ailanthus	33-56			0
Aleurites spp.	tung, candlenut	2,000-3,250		S	0
Alisma plantago-aquatica	arrowhead, duck potato				0
Allium spp.	onion, leek, etc.	2.4	2-9	2	0
Alnus cordata		9			0
Alnus glutinosa	common/black alder	4			0
Alnus incana	grey alder	3.5			0
Alnus rubra	red alder	2			0
Alnus viridis		5			0
Alocasia spp.				2	0?
Alopecurus pratensis	meadow foxtail	1.1		S	0
Althea officinalis	marsh mallow				0
Alysicarpus vaginalis	alyce clover	1.5		S	0
Alyssum spp.		0.3-1			0
Amaranthus spp.	amaranth	0.7	0.8	3,2	0
Amelanchier canadensis	juneberry	5			0
Ammophila arenaria	european beach grass				0
Amorphophallus campanulatus	elephant yam				0
Amorphophallus variabilis	acung				0
Anacardium occidentale	cashew	3,000-4,000		3	0
Ananas comosus	pineapple			3	0
Andropogon spp.	big/sand bluestem	1-5		S	0
Anemone spp.	anemony, windflower	0.7			0
Anethum graveolens	dill	1.2			0
Annona muricata	soursop				0
Annona squamosa	sugar apple				0
Anthemis tinctoria	chamomile				0
Anthoxanthum odoratum	sweet vernalgrass	0.6		S	0
Anthyllis vulneraria	kidney vetch, woundwort				0
Antirrhinum spp.	snapdragon				0
Apium graveolens	celery	0.4-0.7	2		0
Arachis hypogaea	groundnut	1,000-3,000	500	2	0
Araucaria angustifolia	parana pine	5,000			R
Araucaria araucana	monkey puzzle	2,900-5,000			R
Araucaria bidwillii	bunya-bunya	1,000			R
Araucaria columnaris	new caledonian pine	400			0

Araucaria cunninghamii	hoop pine	150			O
Araucaria heterophylla	norfolk island pine	2,000-3,000			R?
Araucaria hunsteinii	klinkii pine	540			R
Arctium lappa	burdock	13	230		O
Aristida spp.					O
Arrhenatherum elatius	tall oatgrass	2.4		S	O
Artemisia spp.	wormwood, tarragon				O
Artocarpus spp.	breadfruit, jackfruit	6,000-8,000		3	R
Asparagus officinalis	asparagus	20-40	25		O
Astragalus spp.	milk vetch, gum tragacanth				O
Atropa belladonna	belladonna				O
Avena spp.	oat	13-49	30	3	O
Axonopus affinis	carpetgrass	0.5		S	O
Baccaurea dulcis	ketupa				R
Baccaurea motleyana	rambai				R
Barbarea verna	upland cress	0.9			O
Basella alba	ceylon spinach	40		4	O
Benincasa hispida	white gourd	70			O
Berberis aggregata	salmon barberry	35			O
Berberis darwinii					O
Berberis gagnepainii		24			O
Berberis julianae	wintergreen barberry	24			O
Berberis thunbergii	japanese barberry	17-24			O
Berberis vulgaris	european barberry	12-33			O
Berberis wilsonii		40			O
Beta vulgaris	sugar/leaf beet	10-20	80	2	O
Betonica officinalis	betony, woundwort				O
Betula alleghaniensis	yellow birch	1			O
Betula lenta	sweet birch	0.7			O
Betula nigra	river birch	1.2			O
Betula papyrifera	paper birch	0.3-1.3			O
Betula pendula	european white birch	0.2-7			O
Betula populifolia	grey birch	0.1			O
Betula pubescens	downy birch	3.5			O
Betula utilis					O
Boehmeria polystachya					O
Bothriochloa ischaemum	yellow bluestem	1		S	O
Bouea macrophylla	gandaria				R
Bouteloua spp.	grama	0.6			O
Brachiaria decumbens		2		S	O
Brachiaria humidicola				S	O
Brachiaria ramosa	browntop millet	3		S	O
Brassica carinata	ethiopian mustard			3	O
Brassica chinensis	pakchoi	1.6		2	O
Brassica juncea	mustard	1.6		2	O
Brassica napus	rape, rutabaga	2.3-4.4		2	O
Brassica nigra	black mustard	0.8		2	O
Brassica oleracea	collards, kale, broccoli, cauliflower, cabbage, brussel sprouts, kohlrabi	3.2		2	O

Brassica pekinensis	chinese cabbage	1.6		2	O
Brassica perviridis	spinach mustard	1.9		2	O
Brassica rapus	turnip, rapekale	1.9-2.4		3	O
Bromus arvensis	field brome	2.2		S	O
Bromus inermis	smooth brome	3-3.3		S	O
Bromus marginatus	mountain brome	7		S	O
Bromus mollis	soft chess	1.8		S	O
Bromus unioloides	rescuegrass	8.9		S	O
Buchloe dactyloides	buffalograss	9			O
Buddleia davidii					O
Cajanus cajan	pigeon pea	70-200		3	O
Calamagrostis canadensis	bluejoint	0.1			O
Calapogonium mucunoides		15			O
Calluna vulgaris	heather				O
Campnosperma brevipetiolata		25 (depulped)			O?
Canavalia spp.	jackbean, swordbean	800-4,000	1,500-6,000	4	O
Canna edulis	edible canna				O
Cannabis sativa	hemp	19	30		O
Capsicum frutescens	peppers	3.5-6.5	15	2	O
Caragana arborescens	pea tree	40			O
Carica papaya	papaya	50		3	O?
Carpinus betulus	hornbeam	6-70			O
Carthamus tinctorius	safflower	33		3	O
Carya illinoensis	pecan	4,500			O
Cassia spp.	senna				O
Castanea spp.	chestnut	650-7,500			R
Catalpa spp.	southern/northern catalpa	20-65			O
Cedrus atlantica	atlas cedar	140			O
Cedrus deodara	deodar cedar	200			O
Cedrus libani	cedar of lebanon	200			O
Celastrus orbiculata	oriental bittersweet	8			
Celastrus scandens	american bittersweet	18			
Celosia spp.	sokoyokoto	0.2-0.8		3	O
Celtis australis		250			O
Celtis occidentalis		250			O
Cenchrus ciliaris	buffelgrass	0.5		S	O
Cercis siliquastrum	judas tree	35			O
Chaenomeles japonica	dwarf japanese quince	40			O
Chaenomeles lagenaria	japanese quince	50			O
Chamaecyparis lawsoniana	port orford cedar, lawson cypress	2-4			O
Chamaecyparis nootkatensis	alaska cedar, nootka cypress	4.2			O
Chamaecyparis obtusa	hinoki cypress	4.2			O
Chamaecyparis pisifera	sawara cypress				O
Chenopodium quinoa	quinoa	2		3	O
Chloris gayana	rhodes grass	0.2			O
Chrysanthemum coronarium					O
Chrysophyllum cainito	star apple				R
Cicer arietinum	chickpea	300-500		2	O
Cichorium spp.	chicory, endive	1-1.7	5		O

Cinnamomum zeylanicum	cinnamon				?
Citrullus vulgaris	watermelon, citron	70-180	400	4	0
Citrus spp.	lemon, lime, oranges etc.	70-140		2	0?
Clematis vitalba	old man's beard	11			0
Cnidoscolus chayamansa	chaya			4	?
Cochlearia danica					0
Cocos nucifera	coconut	$0.5-1.0\times10^6$		2	R
Coffea spp.	coffee	140-450		1	0?
Coleus spp.		0.3		4	0
Colocasia esculenta	taro, eddo, dasheen			4	0
Colocasia gigantea	padang taro			4	0
Colutea arborescens	bladder senna	20			0
Corchorus fruticosus					0
Corchorus olitorius	jute mallow	2-3	3	4,2	0
Coriandrum sativum	coriander	7-13	35		0
Cornus alba	tatarian dogwood	50			0
Cornus controversa					0
Cornus florida	flowering dogwood	250			0
Cornus kousa	kousa	100			0
Cornus mas		300			0
Cornus sanguinea	bloodtwig dogwood	70			0
Coronilla varia	crown vetch	3.3			0
Corylus avellana	hazel, common filbert	2,100		S	0
Corylus colurna		2,000		S	0
Cotinus coggygria	smoke tree	13			0
Cotoneaster acutifolia		30			0
Cotoneaster bullatus		32			0
Cotoneaster dielsianus		20			0
Cotoneaster divaricatus		26			0
Cotoneaster franchetii		19			0
Cotoneaster horizontalis		26			0
Cotoneaster multiflorus					0
Cotoneaster salicifolius					0
Cotoneaster simonsii					0
Crambe scaberrina					0
Crassocephalum crepidioides					0
Crataegus crus galli	cockspur thorn	340			0
Crataegus coccinea		200			0
Crataegus monogyna	hawthorn	50-130			0
Crataegus oxyacantha	hawthorn	165			0
Crataegus prunifolia		335			0
Crotalaria brevidens	slenderleaf crotalaria	5			0
Crotalaria incana					0
Crotalaria juncea	sunn crotalaria	29			0
Crotalaria lanceolata	lanceleaf crotalaria	2.7			0
Crotalaria pallida	smooth crotalaria	4.7			0
Crotalaria retusa					0
Crotalaria spectabilis	showy crotalaria	12.5			0
Cryptomeria japonica		4			0
Cucumis spp.	cucumber, melons, gherkin etc.	5-25	22-81	4	0

Cucurbita spp.	pumpkins, squashes, marrows	35-333	330-830	2	O
Cupressus arizonica	arizona cypress	11-28			O
Cupressus macrocarpa	monterey cypress	40			O
Curcuma spp.	tumeric, curcumas				O
Cyamopsis tetragonoloba	cluster bean	29		4	O
Cydonia oblonga	quince			2	O?
Cynara scolymus	globe artichoke	42		3	O
Cynodon dactylon	giant/bermudagrass	0.3			O
Cynosurus cristatus	crested dogtail	0.5			O
Cyperus spp.					O
Cytisus scoparius	scotch broom	12			O
Dactylis glomerata	cocksfoot	0.6-1.4			O
Daphne mezereum		200			O
Datura spp.					O
Daucus carota	carrot	0.5-3.2	3-6	4	O
Deschampsia spp.					O
Desmodium tortuosum	florida beggarweed	2.3			O
Dichondra repens	dichondra	2.1			O?
Digitaria exilis	hungry rice	0.5		2	O
Dioscorea spp.	yams			3	O
Diospyros spp.	persimmon, ebony	110-900			R
Dipsacus fullonum	fullers teasel				O
Dolichos lablab	hyacinth bean	200-330	330	4	O
Dryobalanops aromatica	borneo camphor	3,000-4,500			R
Durio kutejensis	lai			S	R?
Durio zeylanicum	durian			S	R?
Durio zibethinus	durian	19,000-20,000		S	R
Ecballium elaterium	squirting cucumber				O
Echinochloa crus-galli	barnyard millet	3.3	8	3	O
Ehrharta calycina	perennial veldtgrass	1.5			
Elaegnus angustifolia	oleaster	100-150			O
Elaeis guineensis	oil palm	2,000-4,000		3	O?
Elaeis melanococca	oil palm			2	O?
Eleusine coracana	african/finger millet	2-2.8	4.5	2	O
Elymus canadensis	canada wildrye	5.3			O
Elymus junceus	russian wildrye	2.7-2.9			O
Eragrostis abyssinica	teff	0.3	0.5	4	O
Eragrostis curvula	weeping lovegrass	0.3		S	O
Eragrostis trichodes	sand lovegrass	0.3		S	O
Eriobotrya japonica	loquat				O?
Erodium cicutarium	alfileria	2.3			
Eruca sativa	rocket salad, roquette	2			O
Erythrina poeppigiana	mountain immortelle				O
Erythroxylum coca	coca				R
Eucalyptus archeri		2			O
Eucalyptus coccifera		3.5			O
Eucalyptus dalrympleana		65			O
Eucalyptus globulus	tasmanian blue gum	200			O

Eucalyptus gunnii		3.5			O
Eucalyptus johnstonii		3.5			O
Eucalyptus niphophila		5			O
Eucalyptus nitens		3.5			O
Eucalyptus pauciflora		10			O
Eucalyptus urnigera		3.5			O
Euchlaena mexicana	teosinte				O
Eucommia ulmoides					O
Eunonymus europaeus		55			O
Euphorbia spp.		11			O
Euphoria longan	longan				R
Fagopyrum esculentum	buckwheat	22	25	S	O
Fagus sylvatica	beech	170-650			O
Fedia cornucopiae	african valerian				O
Festuca arundinacea	tall fescue	1.9-2.6		S	O
Festuca longifolia	hard fescue	0.8		S	O
Festuca ovina	sheep fescue	0.9		S	O
Festuca pratensis	meadow fescue	2		S	O
Festuca rubra	red/chewings fescue	1.1-1.2		S	O
Festuca tenuifolia	hair fescue	0.3		S	O
Ficus carica	fig			3	O
Foeniculum vulgare	fennel	1.2-5			O
Fragaria spp.	strawberries	0.4-0.7		3	O
Fraxinus americana	white ash	45			O
Fraxinus excelsior	european ash	75-130			O
Fraxinus latifolia	oregon ash	56			O
Fraxinus nigra	black ash	40			O
Fraxinus ornus	flowering ash	35			O
Fraxinus pennsylvanica	green ash	25-40			O
Garcinia dulcis	mundu				R
Garcinia mangostana	mangosteen			S	R
Ginkgo biloba	ginkgo	700-4,700			R?
Gleditsia japonicum					O
Gleditsia triacanthos	honey locust	165-205			O
Glycine max	soyabean	100-670	175-200	2	O
Gossypium spp.	cotton	63-125	180-350	2	O
Guizotia abyssinica	niger	2.4	4	4	O
Hamamelis virginia	witch hazel	300			O
Helianthus annuus	sunflower	40-200	140-700	3	O
Hevea brasiliensis	rubber	2,000-4,000		2	R
Hibiscus cannabinus	kenaf	13-33		4	O
Hippophae rhamnoides	sea buckthorn	15			O
Holcus lanatus	velvetgrass	0.3		S	O
Hopea spp.		220-400			R
Hordeum vulgare	barley	25-58	40-50	2	O
Hyoscyamus spp.	henbane				O
Hyparrhenia hirta					O

Hypericum calycinum		9			O
Ilex aquifolia	holly	45			O
Indigofera hirsuta	hairy indigo	2.3			O
Indigofera suffroticosa	indigo				O
Ipomoea aquatica	water spinach, kangkong	40-47	63	4	O
Ipomoea batatas	sweet potato			1	O
Ipomoea turbinata					O
Isatis tinctoria	woad				O
Juglans nigra	black walnut	22,200			O
Juglans regia	persian/english walnut	11,800			O
Juglans sieboldiana		8,700			O
Juniperus spp.	junipers	4-250			O
Kerria japonica		22			O
Kerstingiella geocarpa	kersting's groundnut			4	O
Koelreuteria paniculata	goldenrain tree				O
Laburnum anagyroides	golden chain, bean tree	35			O
Lactuca sativa	lettuce	0.6-1.3	2.3-2.9	4	O
Lagenaria siceraria	bottle gourd	130-150	350	4	O
Lansium domesticum	duku				R
Larix decidua	european larch	6-17			O
Larix eurolepsis	dunkeld larch	4-20			O
Larix kaempferi	japanese larch	4-10			O
Larix occidentalis	western larch	3.2			O
Larix sibirica	siberian larch	10.5			O
Lathyrus hirsutus	rough-pea	25			O
Lavendula pubescens	lavender				O
Lens culinaris	lentil	20-50	28	3	O
Lepidium sativum	garden cress	2.4			O
Lespedeza cuneata	sericea lespedeza	1.2			O
Lespedeza hedysaroides	siberian lespedeza	1.2			O
Lespedeza stipulacea	korean lespedeza	1.9			O
Lespedeza striata	common lespedeza	1.3			O
Leucaena leucocephala		36-48			O
Libocedrus decurrens	incense cedar	50			O?
Ligustrum vulgare	common privet	42			O
Linum usitatissimum	flax, linseed	5.5-7	11-12.5	4	O
Liquidambar styraciflua	sweetgum	5.5-7.5			O
Liriodendron tulipifera	tulip tree, whitewood	340			O
Litchi chinensis	lychee			S	R
Lolium multiflorum	italian ryegrass	2.2-2.5		S	O
Lolium perenne	perennial ryegrass	1.7-2.2		S	O
Lonicera tatarica	tatapian honeysuckle	4.5			O
Lotus corniculatus	birdsfoot trefoil	1.2		S	O
Lotus uliginosus	big trefoil	0.5		S	O
Luffa acutangula	angled loofah				O
Luffa cylindrica	vegetable sponge	90-103	235		O

Lupinus albus	white lupin	140		4	0
Lupinus angustifolius	blue lupin	140		4	0
Lupinus arboreus	tree lupin	25			0
Lupinus luteus	yellow lupin	110		4	0
Lycopersicon spp.	tomato	2.5		1	0
Magnolia hypoleuca		360			0
Magnolia kobus		345			0
Magnolia sieboldii		170			0
Mahonia aquifolia	holly mahonia/barberry	15			0
Malus spp.	apple	20-50	70	2	0
Mandragora autumnalis	mandrake				0
Mangifera caesia	kemang				R
Mangifera foetida	bacang				R
Mangifera indica	mango	4,000-5,000		2	R
Mangifera odorata	keweni				R
Manihot esculenta	cassava	114	190	1	0
Manilkara achras	sapodilla				R
Marrubium supinum	hoarhound				0
Matricaria spp.					0
Medicago arabica	spotted burclover	20(in bur)		S	0
Medicago lupulina	black medick	1.7		S	0
Medicago orbicularis	button-clover	3		S	0
Medicago polymorpha	californian burclover	2.7		S	0
Medicago sativa	lucerne, alfalfa	2		S	0
Melaleuca spp.					0
Melilotus alba	white sweetclover	1.8		S	0
Melilotus indica	sourclover	1.5		S	0
Melilotus officinalis	yellow sweetclover	1.8		S	0
Melinis minutiflora	molassesgrass	0.1		S	0
Momordica charantia	balsam pear, bitter gourd	60-170	470	3	0
Morus alba	white mulberry	11			0
Morus nigra	black mulberry	7			0
Mucuna spp.	velvet bean	500		4	0
Musa paradisiaca	banana, plantain			2	0
Myristica fragrans	nutmeg			S	R
Myrrhis odorata	myrrh				0
Nasturtium officinale	water cress	0.2-2.5	4.5		0
Nepeta spp.					0
Nephelium lappaceum	rambutan	190-270		S	R
Nephelium mutabile	pulasan				R
Nicotiana tabacum	tobacco	0.08	0.2		0
Nothofagus cunninghamii		40			0
Nothofagus dombeyi		40			0
Nothofagus obliqua	roble beech	40			0
Nothofagus procera	rauli beech	50			0
Nothofagus pumilio		85			0

Ocimum spp.	basil				0
Oenothera spp.	evening-primrose				0
Olea europaea	olive	250		3	0
Onobrychis viciifolia	sainfoin	20			0
Oreodoxa regia	royal palm	300			0?
Origanum spp.	marjoram				0
Oryza glaberrima	rice	18-35	45	2	0
Oryza sativa	rice	15-40	42-50	2	0
Oryzopsis hymenoides	indian ricegrass	2.8		S	0
Oryzopsis miliacea	smilograss	0.5		S	0
Ostrya carpinifolia	european hop hornbeam	14			0
Oxalis tuberosa	oca			4	0?
Pachyrrhizus erosus	yam bean	150-250	250		0
Panicum antidotale	blue panicgrass	0.7		S	0
Panicum maximum	guineagrass, green panic	0.5-0.8		S	0
Panicum miliaceum	broom corn, hog, proso millet	3-5.5	6.5	3	0
Panicum miliare	little millet			4	0
Panicum virgatum	switchgrass	1.5-2.1		S	0
Papaver spp.	poppy				0
Parkia biglobosa	locust bean	1,000			0
Paspalum dilatatum	dallisgrass	1.6		S	0
Paspalum notatum	bahiagrass	1.6		S	0
Paspalum scrobiculatum	kodo millet	5-7	11	4	0
Paspalum urvillei	vaseygrass	1		S	0
Passiflora edulis	passion fruit				0
Passiflora quadrangularis	giant granadilla				0
Pastinaca sativa	parsnip	2-3	4		0
Pennisetum glaucum	pearl/indian/african millet	5.2-7.5	10	2	0
Persea americana	avocado			3	R
Petroselinum crispum	parsley	1.5-2.2	4.8		0
Peucedanum officinale	dill				0
Phalaris arundinacea	reed canarygrass	0.8		S	0
Phalaris canariensis	canarygrass	6.7		S	0
Phalaris stenoptera	hardinggrass	1.3		S	0
Phaseolus angularis	adzuki bean	90-160	125-180	1	0
Phaseolus aureus	mung bean	30-66	43-47	1	0
Phaseolus coccineus	scarlet runner bean	800-1,300	1,000-1,500	1	0
Phaseolus limensis	lima bean	500-2,000	700-2,750	1	0
Phaseolus lunatus	sieva bean	100-1,500	130	1	0
Phaseolus mungo	black gram	100	130	1	0
Phaseolus vulgaris	kidney bean	100-1,250	140-2,000	1	0
Phellodendron amurense	cork tree	17			0
Phleum pratense	timothy	0.2-0.5		S	0
Phoenix dactylifera	date			3	0
Physalis spp.	husk tomato, cape-gooseberry	0.8-2			0
Picea abies	norway spruce	7-9			0
Picea breweriana		11			0
Picea engelmannii	engelmann spruce	3-5			0
Picea glauca	white spruce	2-5			0

Picea glehnii	sakhalin spruce	3.3			0
Picea jezoensis	yeddo spruce	2.5			0
Picea koyamai	koyama spruce	3			0
Picea mariana	black spruce	1-2			0
Picea omorika	serbian spruce	3-6			0
Picea orientalis	oriental spruce	5.7			0
Picea polita	tigertail spruce	15			0
Picea pungens	(colorado) blue spruce	4-6			0
Picea rubens	red spruce	3			0
Picea sitchensis	sitka spruce	2-3.5			0
Pimpinella saxifraga					0
Pinus albicaulis	whitebark pine	125			0
Pinus aristata	bristlecone pine	20			0
Pinus banksiana	jack pine	3.5			0
Pinus cembra	swiss stone pine	250-500			0
Pinus cembroides	mexican pinyon pine	250			0
Pinus contorta	lodgepole pine	3.3-4.4			0
Pinus coulteri	coulter/bigcone pine	330			0
Pinus densiflora	japanese red pine	10-11			0
Pinus echinata	shortleaf pine	9.5			0
Pinus elliottii	slash pine	33			0
Pinus flexilis	limber pine	100			0
Pinus halepensis	aleppo pine	18			0
Pinus heldreichii	bosnian pine	22			0
Pinus jeffreyi	jeffrey pine	143			0
Pinus koraiensis	korean pine	1,000			0
Pinus lambertiana	sugar pine	200			0
Pinus monticola	western white pine	17-40			0
Pinus mugo	(mugo) swiss mountain pine	5.5-8			0
Pinus nigra	austrian/corsican pine	14-20			0
Pinus palustris	longleaf pine	110			0
Pinus parviflora	japanese white pine	17-110			0
Pinus peuce	macedonian pine	60			0
Pinus pinaster	cluster pine				0
Pinus ponderosa	ponderosa/western yellow pine	40			0
Pinus pumila	dwarf stone pine	55			0
Pinus radiata	monterey pine	20			0
Pinus resinosa	red/norway pine	8.7			0
Pinus rigida	pitch pine	7.4			0
Pinus strobus	eastern white pine	17-20			0
Pinus sylvestris	scots pine	6.5-7.5			0
Pinus taeda	loblolly pine	25			0
Pinus thunbergii	japanese black pine	13-17			0
Pinus virginiana	virginia/scrub pine	8.7			0
Pinus wallichiana	himalayan pine	50			0
Pisum sativum	field, garden pea	90-330	230-330	3,2	0
Poa annua	annual bluegrass	0.4		S	0
Poa arachnifera	texas bluegrass	0.4		S	0
Poa bulbosa	bulbous bluegrass	1.7		S	0
Poa compressa	canada bluegrass	0.2		S	0

Poa nemoralis	wood bluegrass	0.2		S	O
Poa nevadensis	nevada bluegrass	0.4		S	O
Poa pratensis	kentucky bluegrass	0.25-0.45		S	O
Poa trivialis	rough bluegrass	0.2		S	O
Portulaca spp.	purslane	0.1			O
Prunus americana		300-830			O
Prunus amygdalus	almond	2,000-3,600			O
Prunus angustifolia	chickasaw plum	300-600			O
Prunus armeniaca	apricot	1,000		3	O
Prunus avium	cherry	165-330		3	O
Prunus besseyi	western sand cherry	110-310			O
Prunus cerasifera	cherry plum	340-580			O
Prunus cerasus	sour cherry	110-300			O
Prunus domestica	plum	500-1,000		3	O
Prunus laurocerasus	cherry laurel	770			O
Prunus mahaleb	mahaleb/st. lucie cherry	80-145			O
Prunus myrobalana		720			O
Prunus padus	bird cherry	35-70			O
Prunus pensylvanica	pin cherry	20-57			O
Prunus persica	peach	2,000-6,000		2	O
Prunus pumila	sand cherry	75-185			O
Prunus serotina	black cherry	33-190			O
Prunus spinosa	sloe, blackthorn	170-240			O
Prunus virginiana	choke cherry	54-150			O
Pseudotsuga menziesii	blue/green/grey douglas fir	10.5-11.8			O
Pseudotsuga taxifolia	douglas fir	14			O
Psidium guajava	guava				O
Psophocarpus tetragonolobus	goa, wing bean	250-500	330	2,2	O
Pterocarya fraxinifolia					O
Pueraria lobata	kudzu	12.5			O
Pyrethrum spp.					O
Pyrus spp.	pear	4-40		2	O
Quercus borealis	red oak	4,200			R
Quercus cerris	turkey oak	5,300			R
Quercus ilex	holly/holm oak	2,500			R
Quercus palustris	pin oak	1,400			R
Quercus petraea	sessile oak	4,600			R
Quercus robur	common oak	5,000			R
Raphanus sativus	radish	7-15	14-21	2	O
Rhamnus cathartica	common buckthorn	29			O
Rhamnus frangula	alder buckthorn	30			O
Rheum rhaponticum	rhubarb	17			O
Rhus typhina	staghorn sumac	18			O
Rhus verniciflua	chinese lacquer				O
Ribes spp.	currants, gooseberries	6			O
Ricinus communis	castor bean	200-700	400		O
Robinia pseudoacacia	black locust	18-28			O

Rosa canina	dog rose	19			0
Rosa laxa		17			0
Rosa multiflora	multiflora rose	10			0
Rosa rubiginosa		15			0
Rosa rubrifolia		16			0
Rosa rugosa		12			0
Rosa spinosissima	scotch rose	20			0
Rubus spp.	raspberry, blackberry etc.				0
Rumex acetosa	sorrel	1			0
Saccharum officinarum	sugar cane	0.2-0.6	0.3	2	0
Salix spp.	willow	45			0
Salvia officinalis	sage	8.3			0
Sambucus nigra	european elder	4			0
Sambucus racemosa	european red elder	4			0
Sandoricum koetjape	kecapi				R
Sanguisorba minor	little burnet	9.1			0
Satureja hortensis	summer savory	0.6			
Schizachyrium scoparium	little bluestem	1			
Sciadopitys verticillata	umbrella pine	65			0
Scolymus hispanicus	spanish oyster plant				0
Secale cereale	rye	20-37	40-47	3	0
Sechium edule	chayote, christophine			4	R
Sequoia sempervirens	redwood	5-16			0
Sequoiadendron giganteum	giant sequoia	5-27			0
Sesamum indicum	sesame	3	8.5	4	0
Sesbania exaltata	sesbania	9.5			0
Setaria italica	foxtail millet	2-3.3	5.5	2	0
Shorea spp.		60-625			R
Sida spp.					0
Sinapis alba	white mustard	6.3			0
Sisymbrium spp.					0
Skimmia japonica		125			0
Solanum melongena	eggplant, aubergine	3.3-4.4	8	2	0
Solanum quitoensis	naranjilla				0
Solanum tuberosum	potato	0.6-0.7	2.3	2	0
Sophora japonica	japanese pagoda tree				0
Sorbaria sorbifolia		200			0
Sorbus aria	whitebeam tree	33			0
Sorbus aucuparia	european mountain ash, rowan	5			0
Sorbus intermedia		31			0
Sorbus torminalis	wild service tree	34			0
Sorghastrum nutans	indiangrass	2.3-2.9		S	0
Sorghum X almum	almum sorghum	6.7		S	0
Sorghum bicolor	broom corn	17		S	0
Sorghum halepense	johnson grass	3.8		S	0
Sorghum 'Sorgrass'	sorgrass	7.4		S	0
Sorghum sudanese	sudangrass	9-12		S	0
Sorghum vulgare	sorghum	12-28	36-41	2	0
Spartium junceum	spanish/weavers broom	20			0

Spergula arvensis					O
Sphenostylis stenocarpa	african yambean			4	
Spinacia oleracea	spinach	5-10	18	4	O
Spondias dulcis	golden apple				R
Sporobolus cryptandrus	sand dropseed	0.08			
Stachys spp.					O
Stelechocarpus burahol	kepel				R
Stipa viridula	green needlegrass	2.7			O
Styrax japonica	storax, snowbell	1,250			O
Swietenia humilis		470			O
Swietenia mahagoni	mahogany	125-400			R
Syringa vulgaris	common lilac	5-11			O
Syzygium aquaeum	watery rose apple				R
Syzygium aromaticum	clove				R
Syzygium malaccense	pomerac				R
Symphoricarpos alba	snowberry, waxberry	12			O
Talinum triangulare	waterleaf	0.3			O
Tanacetum ptarmicifolium					O
Taraxacum officinale	dandelion	0.8			O
Taxodium distichum	bald cypress	125			O
Taxus baccata	english yew	100			O
Telfairia occidentalis	fluted pumpkin			4	R
Terminalia brassii		4			O
Terminalia calamansanii		17			O
Tetragonia tetragonioides	new zealand spinach	77			O
Tetragonolobus spp.					O
Thea sinensis	tea	1,970		4	R
Theobroma cacao	cacao	1,000-3,600		2	R
Thuja occidentalis	northern white cedar	1.3-2.2			O
Thuja orientalis	oriental/chinese arborvitae	20-30			O
Thuja plicata	western red cedar	1.1-1.8			O
Thymus spp.	thyme				O
Tilia cordata	common lime, small leaved linden	50			O
Tilia platyphyllos	large leaved lime	170			O
Tilia tomentosa	white/silver linden	125			O
Tragopogon porrifolius	salsify	15			O
Tricosanthes anguina	snake gourd				O
Trifolium alexandrinum	berseem clover	2.2		S	O
Trifolium campestre	large hop clover	0.2		S	O
Trifolium dubium	small hop/suckling clover	0.5		S	O
Trifolium fragiferum	strawberry clover	1.6		S	O
Trifolium glomeratum	cluster clover	0.3		S	O
Trifolium hirtum	rose clover	3		S	O
Trifolium hybridum	alsike clover	0.7		S	O
Trifolium incarnatum	crimson clover	3		S	O
Trifolium lappaceum	lappa clover	0.7		S	O
Trifolium pratense	red clover	1.7		S	O
Trifolium repens	white clover	0.5-0.7		S	O
Trifolium resupinatum	persian clover	0.7		S	O

Trifolium semipilosum	kenya clover	0.7		S	0
Trifolium subterraneum	subterranean clover	8.3		S	0
Trigonella foenum-graecum	fenugreek				0
Triticale cereale	triticale	38	48-54		0
Triticum spp.	wheat	22-57	44-50	1	0
Tropaeolum tuberosum	anu	140		4	0
Tsuga canadensis	eastern/canada hemlock	2.5-4			0
Tsuga heterophylla	western/pacific hemlock	1.5-3.5			0
Ulex europaeus	furze, gorse, whin	9			0
Ullucus tuberosus	ullucu			4	
Ulmus americana	american elm	6.7			0
Ulmus campestris	english/european elm	10			0
Ulmus glabra	scotch/wych elm	28			0
Ulmus parvifolia	chinese elm	2.8			0
Ulmus pumila	siberian elm	6.9			0
Urena lobata	aramina				0
Urtica spp.					0
Valerianella locusta	cornsalad	2.6			0
Viburnum lantana	wayfaring tree	55			0
Viburnum opulus	european cranberry bush	42			0
Vicia benghalensis	purple vetch	45			0
Vicia faba	broad bean	181-2,500	280-4,000	3,2	0
Vicia pannonica	hungarian vetch	42			0
Vicia sativa	common vetch	17-70			0
Vicia villosa	hairy/winterpod vetch	29-40			0
Vigna aconitifolia	moth bean			2	0
Vigna angularis	adzuki bean			3	0
Vigna cylindrica	catjang	105	135		0
Vigna mungo	blackgram			2	0
Vigna radiata	greengram, mungbean			2	0
Vigna sesquipedalis	asparagus / yard-long bean	125-220		2,2	0
Vigna trilobata				3	0
Vigna umbellata	rice bean			2	0
Vigna unguiculata	cowpea	100-120	130-155	2	0
Vitis vinifera	grape	43	80	3	0
Voandzeia subterranea	bambara groundnut	670		3	0
Wisteria sinensis	chinese wisteria	460			0
Zea mays	maize	290-330	360-430	3	0
Zelkova serrata		34			0
Zingiber spp.	ginger				0
Zoysia japonica	japanese lawngrass	0.8			0

APPENDIX 3. PSYCHROMETRY AND SEED DRYING

Introduction

This appendix is provided as a source of information for engineers designing seed drying facilities within gene banks and as an introduction to the quantifiable relationships between environment and seed drying rates. The general relationships described here provide a useful framework for design calculations and are approximately true for most species. However, since there are some apparent anomalies and conflicting data in the literature, seed bank managers should determine and monitor the performance of their drying equipment on the species they handle.

1. Psychrometry - the study of the properties and behaviour of air and water vapour mixtures

The composition of air is generally accepted as being 78% nitrogen, 21% oxygen, 0.9% argon and 0.03% carbon dioxide. However air also contains varying amounts of water vapour. The drying process is dependent on the amount of water vapour held in the air and the temperature of an air/water vapour mixture. Since seeds are dried in air/water vapour mixtures, a basic understanding of psychrometry is useful to those designing and/or operating seed drying equipment. Indeed it is particularly important when seeds are to be dried to very low moisture contents.

There is a limit to the maximum amount of moisture that dry air can absorb at a given temperature. The higher the temperature the greater is the maximum amount of moisture that can be held within a given volume of air. Broadly speaking, the maximum amount of moisture held doubles for each 10°C temperature rise. However, the weight of water vapour that can be absorbed by dry air is small; for example at 0°C and 30°C it is less than 0.004 kg and 0.028 kg moisture/kg of dry air (i.e. 0.4% and 2.8% respectively). Moreover, air is usually only partially saturated; that is the amounts of water vapour held are less than these values. Psychrometric charts (Figure 3.1) have been constructed to show the properties of air and water vapour mixtures. They enable simple air conditioning problems to be solved graphically and are therefore suitable for calculating suitable coldroom and drying equipment designs.

The axes of the psychrometric chart (Figure 3.1A) are defined as follows (clockwise order starting from the top).

PERCENTAGE SATURATION

The ratio of the actual moisture content of the air (kg/kg) to the maximum (saturated) moisture content of air (kg/kg) at the same temperature. This is the *relative humidity*. The ratio can also be expressed in terms of water vapour pressures, i.e. the ratio of the actual water vapour pressure to the saturated water vapour pressure at the same temperature.

MOISTURE CONTENT (AIR)

The weight of water vapour per unit weight of dry air (kg/kg). If it is required air moisture content *(W)* can be converted to vapour pressure using either the following expressions,

$$\text{vapour pressure (mb)} = 1013.2 \times W / (0.662 + W) \quad (3.1)$$

or more approximately " " $= 1590 \times W$ (3.2)

or the vapour pressure nomogram (Figure 3.1B).

DRY-BULB TEMPERATURE

The temperature of the air measured with an ordinary thermometer. (The thermometer should be screened to prevent radiant heat sources from affecting the reading).

WET-BULB TEMPERATURE

The temperature measured with a thermometer whose bulb is covered by a wetted wick exposed to a moving current of air (ideally 5 m/second). Note that the chart will be inaccurate if "still air" wet-bulb measurements are used. The difference between the temperature readings of the dry- and wet-bulb thermometers is called the wet-bulb depression. The greater the depression for a given dry-bulb temperature, the drier the air.

SPECIFIC ENTHALPY

This is a thermodynamic quantity. It is the combined total of sensible and latent heat fractions of the air mixture relative to an arbitrary datum temperature (0°C) and is frequently called Total Heat or Heat Content. Sensible heat is that which changes the temperature of the air/water vapour mixture but not its state. Latent heat is that which changes the state of the air mixture but not the temperature.

SPECIFIC VOLUME

The volume of air per unit weight of dry air (m^3/kg), that is the reciprocal of the density of dry air.

DEW-POINT TEMPERATURE

The temperature to which air must be cooled to condense moisture contained in the air. It is not shown in the psychrometric charts (Figure 3.1) but it can be calculated easily using the chart. An example determination of the dew-point temperature is provided here, first to assist those learning to use the chart, and second to emphasize an important precaution in seed bank management.

Suppose that the (air-dry) temperature of a laboratory is 25°C and a wet-bulb thermometer reads 18°C. These points of these two axes of the psychrometric chart (Figure 3.1A) cross where the moisture content of the air is 0.01 kg/kg. Now this moisture content line can be continued until it reaches the 100% saturation curve. At this point the wet-bulb temperature is 14°C. Thus 14°C is the dew-point temperature since at this temperature the air would be fully saturated. Note that the point of intersection of the dry-bulb and wet-bulb temperatures (25° and 18°C respectively) is on the 50% relative humidity curve. That is the

Fig. 3.1 Psychrometric charts and vapour pressure nomogram.
A. Psychrometric chart, -10° to 60°C. This chart is reproduced with the permission of the Chartered Institution of Building Services from whom pads of charts sized A3 for permanent records may be obtained.

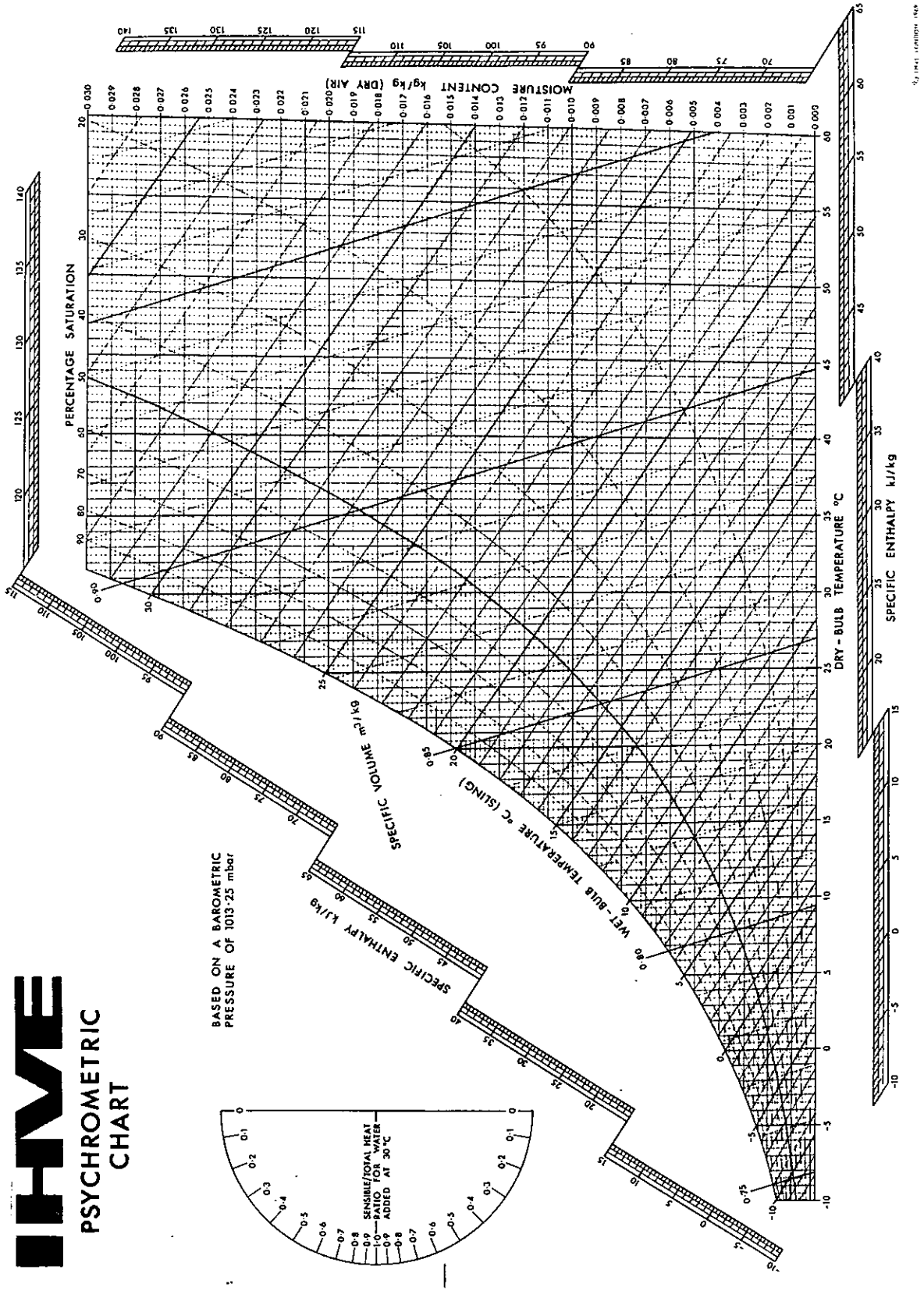
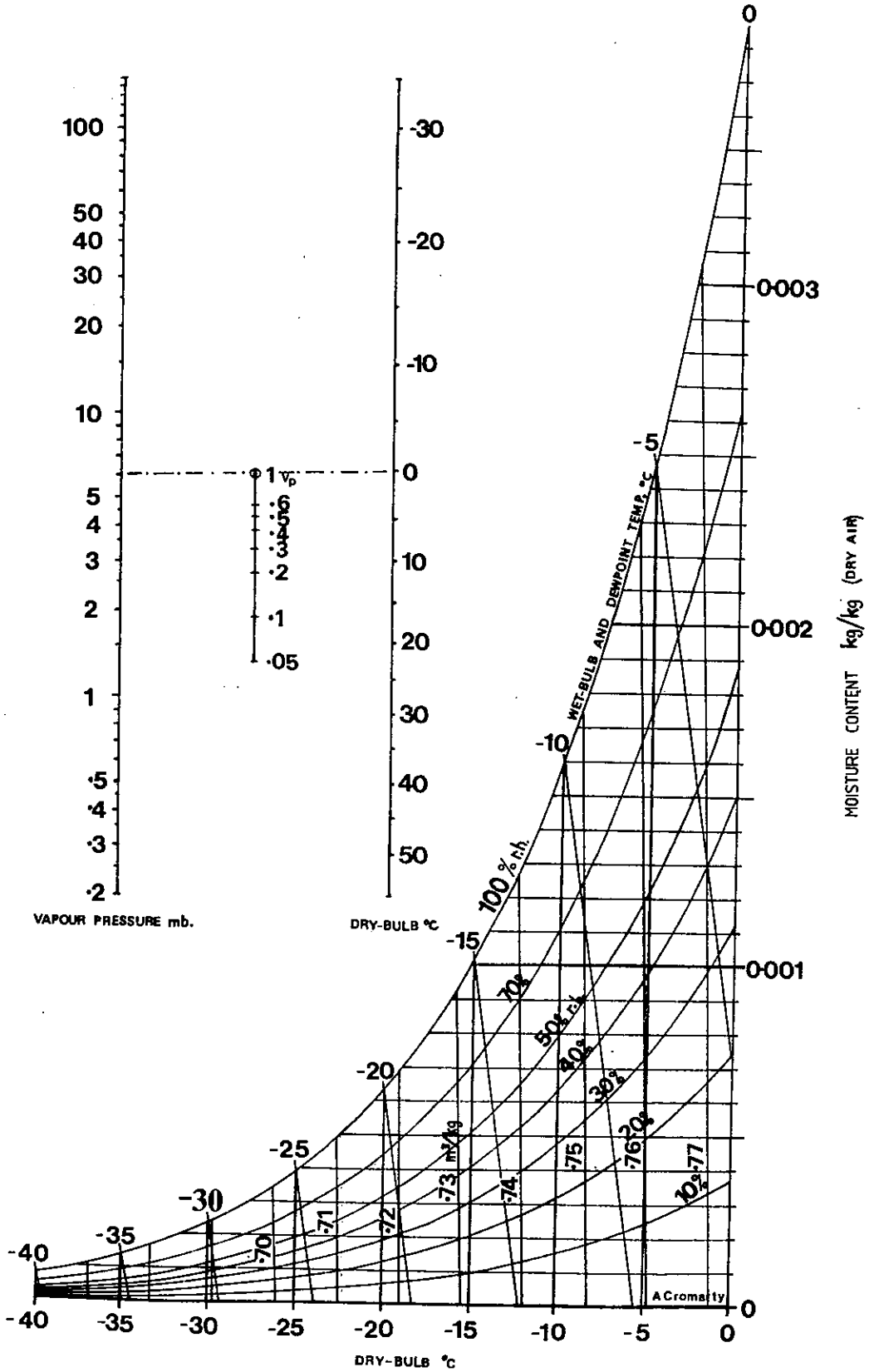

IHVE
PSYCHROMETRIC CHART
BASED ON A BAROMETRIC PRESSURE OF 1013·25 mbar
PERCENTAGE SATURATION
MOISTURE CONTENT kg/kg (DRY AIR)
SPECIFIC VOLUME m³/kg
WET-BULB TEMPERATURE °C (SLING)
DRY-BULB TEMPERATURE °C
SPECIFIC ENTHALPY kJ/kg
SENSIBLE/TOTAL HEAT RATIO FOR WATER ADDED AT 30°C

Figure 3.1 A

Figure 3.1 B


VAPOUR PRESSURE mb.
DRY-BULB °C
WET-BULB AND DEWPOINT TEMP. °C
MOISTURE CONTENT kg/kg (DRY AIR)
100 % r.h.
70%
50% r.h.
30%
10%
m³/kg
.70
.71
.72
.73
.74
.75
.76
.77
A Cromarty
DRY-BULB °C

relative humidity of the laboratory is 50%. If the dew-point temperature of the laboratory atmosphere is 14°C then moisture from the atmosphere will condense on the surface of any article with a temperature of 14°C or below which is placed in the laboratory. Thus, for example, moisture will condense on the surface of containers from either a medium-term store (0°C) or a long-term store (-20°C). Moreover if the containers are opened moisture will condense on the containers' contents thereby increasing their moisture content. Thus two precautions are necessary in seed banks. First waterproof labelling should be used on containers. Secondly containers should not be opened until the temperature of the seed within the container has risen above the dew-point temperature of the atmosphere. This is considerably longer than the time required for the temperature of the surface of the container to rise to ambient temperature, since bulks of seed are self-insulating and heat or cool slowly. The following empirical expression is provided to estimate the nominal safe reheating period, H (hours), for seed within cylindrical containers with radius r (mm):

$$H = r^{1.57}/50 \qquad (3.3)$$

For example, a full 0.9 l can of 103 mm diameter will take at least 9 hours to reheat from -20° to 20°C.

Before the psychrometric chart can be used to design seed drying conditions it is necessary to understand the relationship between seed moisture content and air moisture content. Given sufficient time in a closed system the moisture contents of seeds and the atmosphere with which they are in contact reach an equilibrium.

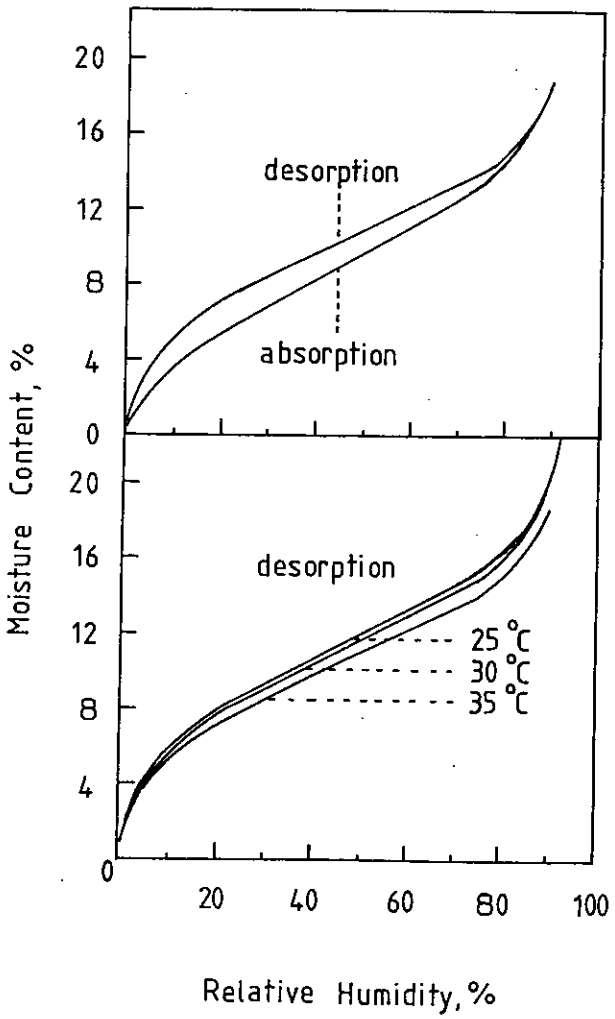
2. *Equilibrium relationships between seed moisture content and relative humidity*

The relationship between seed moisture content and the relative humidity of the atmosphere when equilibrium has been reached is positive, but not linear (Figure 3.2). Moreover two curves are necessary to define the relationship at a given temperature because of a hysteresis effect; at a given relative humidity the moisture content of seeds on desorption (i.e. being dried) is greater (often by 1-2 percent) than that of seeds on absorption (i.e. being humidified) (Figure 3.2). However, since we are interested only in seed drying we can ignore the absorption curve and consider desorption curves only. Temperature has a small effect on the moisture content at any given relative humidity: the lower the temperature the greater the moisture content of the seeds (Figure 3.3). In general, the increase in equilibrium moisture content at a given relative humidity which results from a given reduction in temperature becomes less at lower temperatures (Figure 3.3). Although there are definable equilibrium relationships between relative humidity and seed moisture content of a given species, it is important to note that the composition of the seed (which can be affected by both genotype and the growing environment) has a slight effect on the equilibria. For example, flint maize can have a higher equilibrium moisture content at a given temperature and relative humidity than dent maize. It should also be noted that the equilibrium hygroscopic relationships can differ within a seed (i.e. between the embryo and endosperm) but this is unimportant practically.

Fig. 3.1 Psychrometric charts and vapour pressure nomogram (continued).
B. Low temperature, -40° to 0°C, skeleton psychrometric chart and vapour pressure nomogram (A.S. Cromarty, unpublished). To use the vapour pressure nomogram join points on the vapour pressure scale (left), the vapour pressure ratio (or relative humidity expressed as a decimal) scale (centre), and the dry-bulb temperature scale (right) with a straight line. In the example given a fully saturated atmosphere (i.e. Vp = 1) at 0°C has a vapour pressure of 6 mb.

Fig. 3.2 Hysteresis in moisture sorption in wheat. Desorption and absorption isotherms at 35°C. (From J.E. Hubbard, F.R. Earle and F.R. Senti, 1957. Moisture relations in wheat and corn. Cereal Chemistry, 34, 422-433.)

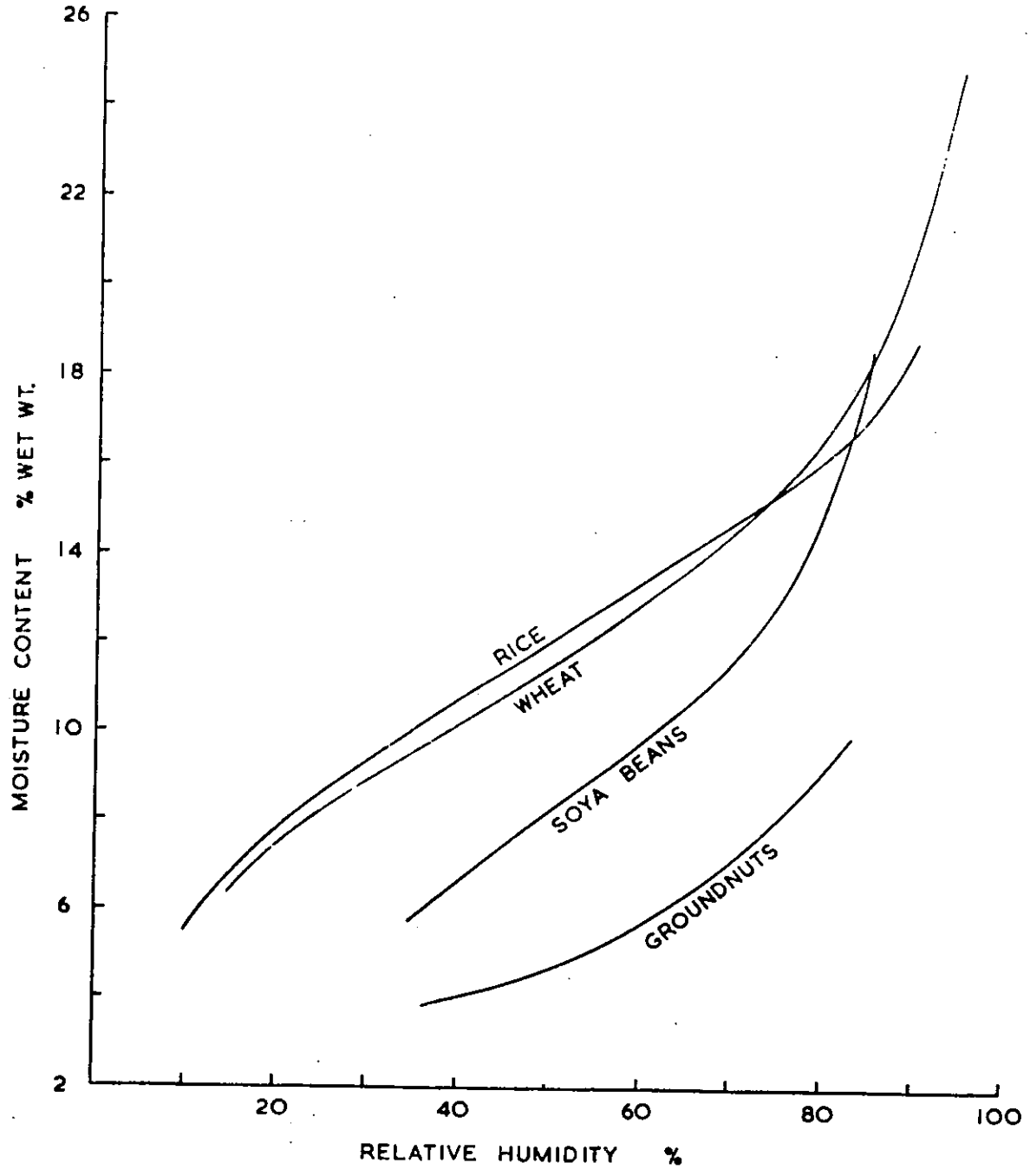
Fig. 3.3. Desorption isotherms for wheat at 25°, 30° and 35°C. (Source: as for Figure 3.2).



Differences between the equilibrium hygroscopic relationships for seed of different species, however, can be substantial (Figure 3.4). As a result exposure to a given relative humidity until equilibrium is reached will not result in the seed of all species having the same moisture content. Nevertheless, the same environment can be used if those species which have a low equilibrium moisture content are removed from the drying environment before that equilibrium is reached: that is, drying of very many different species to the same moisture content can be achieved in a single environment by altering the drying treatment period, provided that the relative humidity is sufficiently low for the species with the highest equilibrium seed moisture content.

Most of the differences between the species hygroscopic equilibria demonstrated in Figure 3.4 are related to seed oil content. The greater the oil content, the lower the seed equilibrium moisture content. But relative humidity and temperature also affect equilibrium moisture contents. When there are no reliable experimental data available for a particular species it is suggested that the following expression (A.S. Cromarty, unpublished) be used for seed drying applications within the temperature and humidity ranges of 0°-40°C and 10-70% rh for starchy seeds (e.g. cereals), but only 15°-25°C and 10-70% rh for oilseeds,

Figure 3.4. The relative humidity equilibrium relationship of seed of various species which shows the difference between oily and non-oily seeds. Each curve was prepared from the results of several workers by S.W. Pixton (Moisture content - its significance and measurement in stored products. Journal of Stored Products Research, 1967, 3, 35-47) and is reproduced with permission. These curves are shown to illustrate the difference between species. This information - and that in Figures 3.2 and 3.3 - however, cannot be used as calibration curves for each species since the form of the curve within a species is dependent upon seed composition, the number of times the seed have been wetted and dried, and the experimental procedure used to determine these equilibria.



$$(1 - R) = e^{-[\{Me \times (1.1 + T/90)/(1 - D_o)\}^2/440]} \quad (3.4)$$

where

R = relative humidity (expressed as a decimal, not %)

Me = equilibrium percentage moisture content (db)

T = temperature of the air, and also the seed at equilibrium, °C

D_o = oil content of the seed, db (expressed as a decimal)

[e = constant 2.71828]

In this expression the equilibrium moisture content is expressed as db (dry basis). If, as is likely, the preferred moisture content is expressed as wb (wet basis) then this should first be converted to the equivalent db value using either the equation (5.3) or the scale (Figure 5.1) provided in Appendix 5.

The following list of species is provided as a guide to typical seed oil content values, expressed as a percentage of the total dry weight of the seed.[1]

Parkia biglobosa	(locust bean)	1.3
Pisum sativum	(garden pea)	1.5
Vicia faba	(field bean)	1.5
Phaseolus lunatus	(butter bean)	1.5
Hordeum vulgare	(barley)	1.7
Secale cereale	(rye)	1.7
Oryza sativa	(rice)	1.8
Triticum aestivum	(wheat)	1.9
Lens culinaris	(lentil)	1.9
Quercus robur	(oak)	2.4
Fagopyrum esculentum	(buckwheat)	2.6
Sorghum vulgare	(sorghum)	3.8
Pennisetum glaucum	(pearl millet)	3.9
Zea mays	(maize)	4.4
Avena sativa	(oats)	4.8
Glycine max	(soyabean)	18.0
Gossypium hirsutum	(cotton)	21.5
Allium cepa	(onion)	24.0
Fagus sylvatica	(beech)	27.5
Helianthus annuus	(sunflower)	32.5
Linum usitatissimum	(linseed)	36.6
Arachis hypogaea	(groundnut)	44.9
Brassica napus	(rape)	45.2
Sesamum indicum	(sesame)	47.5

As an example in the use of the above equation we shall determine the relative humidity necessary for maize seed (4.4% oil content) to equilibrate at 5% moisture content (wb) at 15°C. Now 5% (wb) is equivalent to 5.3% (db) (eqn. 5.3).

---

[1] For a more detailed examination of the influence of relative humidity and temperature on the moisture content of biological materials see:
P.O. Nepoddy and F.W. Bukker-Arkema, 1970. A generalized theory of sorption phenomena in biological materials. Transactions of the ASAE 13, 612-617.
and C.S. Chen and J.T. Clayton, 1971. The effect of temperature on sorption isotherms of biological materials. Transactions of the ASAE 14, 927-929.

Thus,

$$(1 - R) = e^{-[\{5.3 \times (1.1 + 15/90)/(1 - 0.044)\}^2/440]}$$

$$= e^{-0.1121}$$

Therefore $(1 - R) = 0.90$

and $R = 0.10$ or 10% relative humidity

Alternatively at 30°C the equation becomes

$$(1 - R) = e^{-[\{5.3 \times (1.1 + 30/90)/(1 - 0.044)\}^2/440]}$$

$$= e^{-0.1435}$$

Therefore $(1 - R) = 0.87$

and $R = 0.13$ or 13% relative humidity

Eqn. (3.4) can be rewritten to estimate equilibrium moisture contents (db) at a given temperature and relative humidity as follows:

$$Me = [(1 - D_o)\sqrt{\{-440 \times \ln(1 - R)\}}] / \{1.1 + (T/90)\} \quad (3.5)$$

3. Drying rates

Once it is known that a given environment will provide the required equilibrium moisture content the only question remaining unanswered is how long will it take to dry the seeds to this value? The drying rate is controlled by internal moisture diffusion and the ability of the air close to the seed to absorb the released water vapour. Increase in temperature will increase the rate of moisture diffusion from the centre of the seed to the seed surface and the difference in vapour pressures between the seed surface and the atmosphere, thereby shortening the time to equilibrium, whereas increase in seed size increases the distance through which moisture must diffuse before it reaches the seed surface, thereby increasing the time to equilibrium. Initial seed moisture content affects this period, but since seeds are being dried to low moisture contents and the rate of moisture loss declines dramatically the closer actual seed moisture content approaches the equilibrium value, the effect of initial seed moisture content is small.

The following general expression (A.S. Cromarty, unpublished), which relates to test work on seed moisture equilibrium relationships, is provided as a guide to the major effects of temperature and seed size on the time taken to reach equilibrium moisture contents in still air:

$$Pe = 3650/(GS)^{2/3} \times Ps \quad (3.6)$$

where Pe = number of days to equilibrium

GS = number of seeds per gramme

Ps = saturated vapour pressure (mb) at the drying temperature

(use Figure 3.1A and B or Figure 3.1A and eqn. (3.1))

Table 3.1. Estimated time in days for seed to dry to equilibrium in still air at various temperatures [from eqn. (3.6)]. These figures can also be calculated from the seed drying nomogram (Figure 3.5).

Temperature °C	Cabbage (GS[1] = 315)	Maize (GS = 4)	Onion (GS = 341)	Sorghum (GS = 50)
0	13	240	12	44
5	9	170	9	31
10	7	120	6	22
15	5	86	4.5	16
20	3.5	62	3	11
25	2.5	45	2.5	8
30	2	33	1.5	6
35	1.5	26	1.5	5

[1] GS = no. of seeds per gramme

To emphasise the influence of seed size and temperature, estimates of Pe derived from this expression are provided in Table 3.1. These calculations assume that seeds are in shallow layers (just a few seeds thick) over perforated bases, but with no ventilation. They do not apply to situations where seed bulks remain packed within bags. It is clear from Table 3.1 that a drying temperature of about 15°C provides suitable practical drying periods for the small seeded species and to a lesser extent for sorghum, whereas for maize the periods are substantial. Note that through ventilation would significantly reduce these periods (see Figure 3.5).

The figure of 26 days for maize seed to dry to equilibrium at 35°C (Table 3.1) may seem barely credible to those concerned with commercial seed drying, because maize seed is regularly dried at 35°-40°C in considerably less time. Why the apparent discrepancy? First, commercial seed driers are ventilated, and secondly, the seeds are only dried part way to equilibrium. The change in seed moisture content during drying slows down exponentially as the equilibrium is approached and can be plotted linearly on log/linear graph paper if this change is expressed in terms of the moisture content ratio (MCR). The moisture content ratio is the ratio of the difference between the moisture content (M) after H hours drying and the equilibrium moisture content (Me) to the difference between the original moisture content before drying (Mo) and the equilibrium moisture content:

that is
$$MCR = (M - Me)/(Mo - Me) \qquad (3.7)$$

where seed moisture content is expressed on the dry basis. For drying seed in a seed bank the value of the moisture content ratio will be low because the value of M will generally be close to the equilibrium moisture content.

No single expression can be universally applied to describe thin layer drying characteristics. Nevertheless the following general drying equation for agricultural seeds (A.S. Cromarty, unpublished) serves to illustrate important features of the underlying process:

$$MCR = A\, e^{-(HPG)} + B\, e^{-(HPG\,\pi^2)} \qquad (3.8)$$

where

H = drying time, hours

$P = Ps/2667(1 - D_O)(1 - [0.115 \times V])$

$G = (GS)^{2/3}$

Ps = saturated vapour pressure (mb) based on seed temperature

D_O = oil content (db), decimal

V = air velocity, m^3/second per m^2 of bed (m/s) (range 0.05-2.5 m/s)

and A and B are constants with the values 0.6 and 0.4 respectively (corrected for use with Me).

The second term of equation (3.8) can be disregarded when the MCR is less than 0.35, and this allows the remainder of the expression to be rearranged to calculate the drying time (hours) to low moisture contents:

$$H = [\ln(MCR/A)] \;/\; -PG \qquad (3.9)$$

The equations (3.8, 3.9) have some limitations since they assume that all seeds are spheres whereas they are often cylindrical or disc-shaped and, further, the number of seeds per gramme (GS) changes during drying and therefore only an average value can be used. Nevertheless the equations are of fairly general application and can be used for most crop species. An example calculation is shown here for maize seed (which is relatively slow drying) to estimate the time taken to dry from 15% to 6% moisture content (wb) in thin layers at 30°C with 10% rh and an air flow velocity of 2.5 m/s.

Now $P = 43.6/2667\ (1 - 0.044)(1 - [0.115 \times 2.5]) = 0.024$

$G = [4]^{2/3} = 2.52$

We can use equation 3.5 to determine the equilibrium moisture content (Me) at 30°C with 10% relative humidity.

$$Me = [(1 - 0.044)\sqrt{\{-440 \times \ln(1 - 0.1)\}}\,] \Big/ \{1.1 + (30/90)\}$$

Therefore $Me = 4.54\%$ (db)(= 4.34% wb)

15% and 6% (wb) are equal to 17.65% and 6.38% (db) respectively, and consequently the moisture content ratio

$= (6.38 - 4.54)/(17.65 - 4.54) = 0.140$

Substituting in equation 3.9 gives

$H = [\ln(0.140/0.6)] \;/\; (-0.024 \times 2.52)$

$= 24.1$ hours

Thus in a drier where maize seed are in a shallow layer and ventilated with air at 30°C and 10% relative humidity at an air flow rate of 2.5 m/s it will take about a day to dry from 15% to 6% (wb), whereas in still air at the same conditions it would take about a month (Table 3.1) to dry the seed to 4.3% (wb), the

equilibrium moisture content (equation 3.4). These calculations are manually tedious, but can be readily solved by computer. The nomograms (Figure 3.5) are sufficiently accurate for gene bank design purposes and provide a rapid method for solving these problems graphically. However, the equations underestimate the drying time required if equilibrium relative humidity levels are lowered below 35%, because the remaining free moisture is held tenaciously.

4. Limiting damage to seed during drying

The purpose of drying seed is to limit the subsequent rate of seed deterioration during long-term storage. However the drying process itself can also be considered as a storage treatment during which seed ageing can occur at a rate dependent on time, temperature and moisture. Now the equation which describes this relationship (Appendix 4) is only appropriate for constant environments, whereas when seed are dried their moisture content and, less obviously, temperature change. Nevertheless, the viability equation can be applied if the drying period is treated as a succession of very short discrete periods at very many different, but constant, storage environments. Commercial recommendations for air temperatures, air flow rates and drying periods have been determined empirically in temperate countries to minimise damage during drying. Now these recommendations vary between species and depend upon initial seed moisture content[1], but more importantly they are unsuitable for poor quality seed lots and heterogeneous seed lots and where final moisture contents are as low as 5%. Further, in tropical environments hot air driers are unable to dry seed sufficiently because the moisture content of ambient air is too high. Thus these recommendations cannot be applied in gene banks. However, it is important that gene bank drying procedures are also designed to minimise seed deterioration.

This we have attempted to do using the equations provided in this appendix to determine change in seed moisture content over time (3.8, 3.9), and the seed viability equation (4.1) (Appendix 4). The criterion for minimising damage during drying is to limit the change in probit percentage viability caused by any drying regime (see Appendix 4). This can be done using a digital computer and iterative programming techniques so that the fall in probit percentage viability is progressively recalculated as seed temperature and seed moisture content change. A close approximation of the computer simulation has been devised for gene bank use,

[1] For example, see those tabulated by E.H. Roberts and D.L. Roberts (Moisture content of seeds. In Viability of Seeds (ed. E.H. Roberts) 424-437, Chapman and Hall, London, 1972).

Figure 3.5. Seed drying nomograms

A. This nomogram (A.S. Cromarty, unpublished) provides a solution to the general thin layer drying equation (3.8) by graphical anamophosis. The example provided is that given numerically in the text. It is for maize seed dried at 30°C and ventilated at 2.5 m/s. The nomogram helps to illustrate the relative importance of drying temperature, oil content, seed weight, and air velocity on drying rate - as expressed by the moisture content ratio (MCR) after a given time. The displaced line below MCR 0.01 (EQUI. ESTIMATE MAX.) can be used to estimate the maximum time taken for seed moisture content to approach the equilibrium value under still air conditions, eqn. (3.6). In this case the appropriate value of the air velocity is 0 m/s - the uppermost line. This diagram is useful for general design work as a preliminary to more detailed calculation.

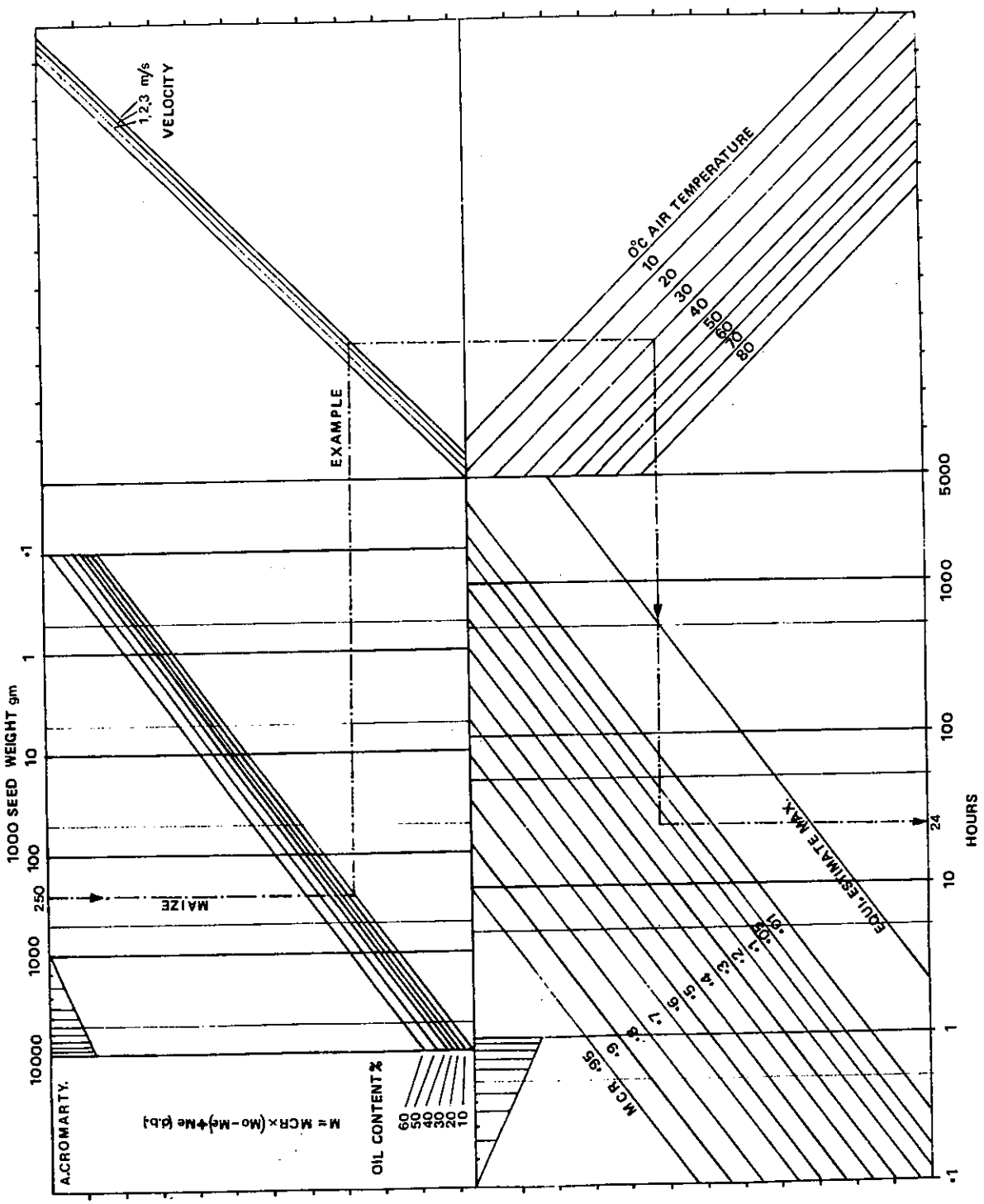

1,2,3 m/s
VELOCITY
EXAMPLE
0°C AIR TEMPERATURE
10
20
30
40
50
60
70
80
1000 SEED WEIGHT gm
10000
1000
250
100
10
1
·1
MAIZE
A.CROMARTY.
M = MCR×(Mo-Me)÷Ne (d.b.)
OIL CONTENT %
60
50
40
30
20
10
MCR
·95
·9
·8
·7
·6
·5
·4
·3
·2
·1
·05
EQUIL. ESTIMATE MAX
HOURS
5000
1000
100
24
10
1
·1

Figure 3.5 A

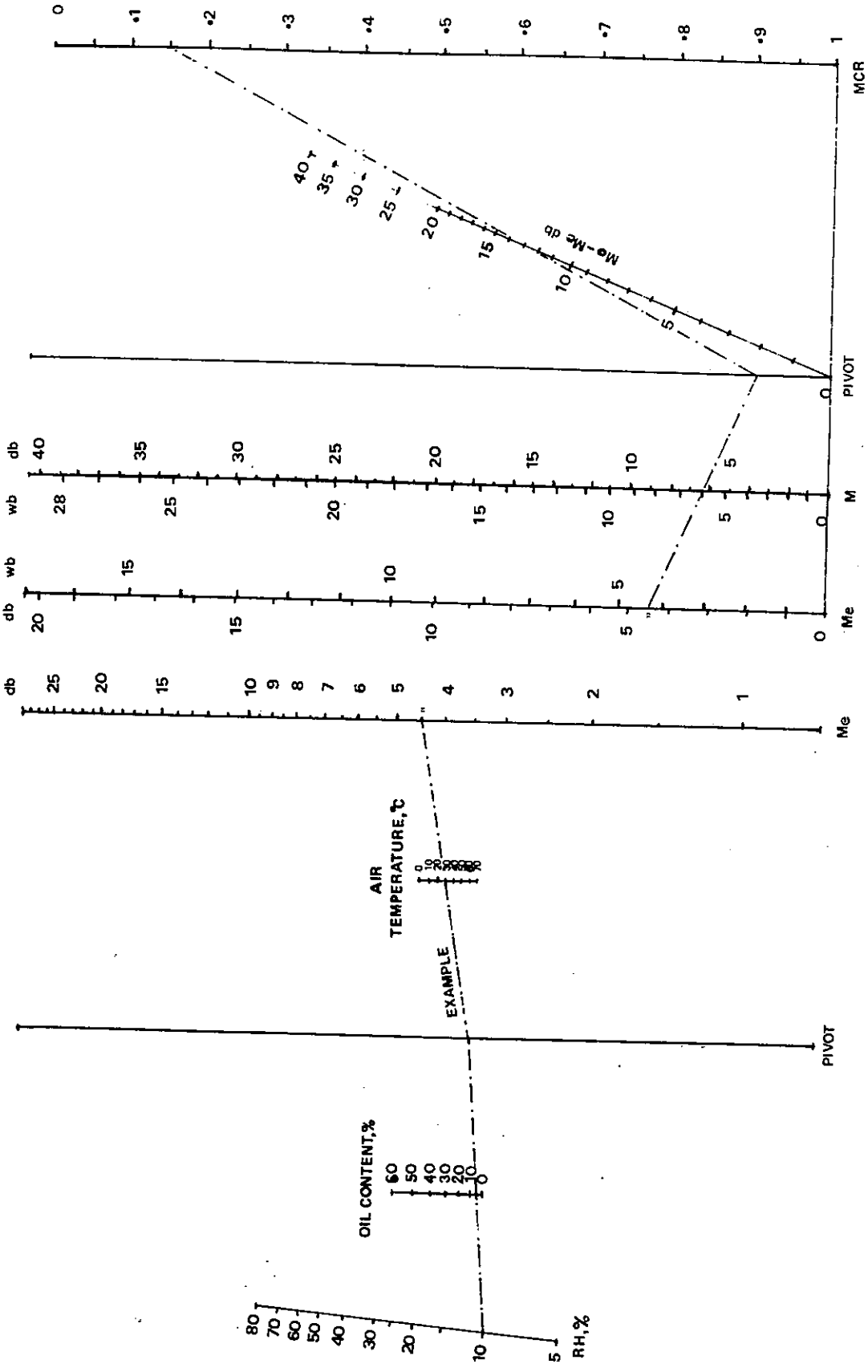

MCR
PIVOT
M
Me
Me - Mc db
db
wb
AIR TEMPERATURE, °C
EXAMPLE
OIL CONTENT, %
RH, %

Figure 3.5 B

where an 'effective moisture content' is determined which corrects for the change in seed moisture content that occurs during the drying period. This value and the time taken to dry the seed are then inserted into the seed viability equation (Appendix 4).

The calculation sequence to estimate the loss in probit percentage viability that occurs during drying is as follows:

1. Define the drying temperature, air relative humidity, the initial and final seed moisture contents, the 1,000 seed weight and seed oil content.

2. Calculate the equilibrium moisture content (db) using eqn. (3.5) or Figures 3.5B.

3. Convert initial and final moisture contents to db and calculate the moisture content ratio (MCR) using eqn. (3.7) or Figure 3.5B.

4. Calculate the 'effective moisture content' (m) using the following expression (A.S. Cromarty, unpublished);

$$m = [\{0.48 + (0.48 \times MCR)\}\{Mo - Me\}] + Me \qquad (3.10)$$

where Me and Mo are the equilibrium and original seed moisture contents (db) respectively.

5. Convert the value for the effective seed moisture content during drying from db to wb using eqn. (5.4) (Appendix 5).

6. Calculate the time necessary to dry the seed to obtain the calculated MCR using eqn. (3.8) or Figure 3.5A. Convert the drying time from hours to the equivalent value in days.

7. Substitute the drying temperature for t, the value of the drying time in days (calculated in 6) for p, and the 'effective moisture content' (wb) (calculated in 4 and 5) for m in the viability equation (4.2) (Appendix 4) to estimate the probit loss in viability occurring during drying. Alternatively, for a given seed lot where the value of the seed lot constant (K_i) is known, equation (4.1) can be used to determine probit percentage viability after drying.

Seeds are cooled when moisture is lost as a result of the latent heat of evaporation. Consequently seed temperatures are lower than temperatures during the early stages of drying. Thus the use of the drying temperature in eqn. (4.2) provides a small safety margin in these calculations. For seed lots of 50% viability

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Figure 3.5. Seed drying nomograms (continued)

B. These nomograms are provided to allow the equilibrium moisture content (Me), eqn. (3.5), and the moisture content ratio (MCR), eqn. (3.7), to be estimated, where Mo and M are the original and final seed moisture contents respectively. The example provided is that given in the text for maize seed where the required moisture content after drying is 6% (wb).

the safety margin is between 2°-7°C, but the safety margin is smaller for seed lots of higher viability - as will generally be the case in gene banks. It is therefore recommended that this approach be utilized for general drying and drying room design work for gene banks.

The results of analyses using these procedures with data for onion and soyabean (two species in which loss in viability is comparatively rapid) suggests that a theoretical optimum drying temperature exists and apparently covers a narrow band close to 10°C, although at temperatures between 0° and 20°C the loss is not appreciably greater. However, as has been shown, the more starchy and larger-seeded cereal species take much longer to dry at these temperatures. Consequently optimum temperatures are generally higher but this depends very greatly on the initial moisture content; the higher the initial moisture content the lower the optimum drying temperature. The problem is that these values span a very wide temperature range from those temperatures optimal for onion (above), to the maximum air temperatures recommended for commercial seed drying (c. 60°C). To overcome this problem a two-stage drying procedure is recommended for large-seeded species (para. 18).

There are certain conditions in which the viability equations are no longer valid and these must be noted at this point before recommendations regarding drying regimes can be made since they concern seed maintained at moisture contents in excess of 20% to 25% (wb). Contrary to the viability equation, seed at moisture contents close to fully imbibed can maintain viability for considerable periods provided oxygen is present, germination is prevented, fungal deterioration can be avoided, and the temperature is not excessively high. It is only in the transition from the fully hydrated maturing seed to the air-dry seed that the very rapid rates of deterioration occur. In the majority of crop species this phase of drying occurs naturally on the plant prior to harvest. However in those species where this does not occur before harvest (e.g. tree fruits) it is preferable to maintain seed at moisture contents close to fully imbibed and aerate during drying bottlenecks (rather than allow the seed to dry very slowly, as would occur in a very humid environment), and then to dry the seed rapidly when the drying capacity is available.

It should also be noted that very moist seed should not be exposed to extremes of temperature. First, at temperatures below 0°C freezing damage will occur. Secondly, at high temperatures secondary dormancy can be induced. For example in apple and pear seed this occurs if moist seed are stored (or dried) at temperatures above 17°C, and this phenomenon has been reported in moist seed of many other species.

Further moist seed is liable to be attacked by insects, mites and fungi. Damage from insects and mites can largely be avoided by cooling seed to below 17°C. However to prevent fungal deterioration a lower seed temperature (5°C) is necessary unless the seed are in equilibrium with an atmosphere of less than 70% relative humidity. Thus to prevent insect, mite and fungal damage a pre-drying holding environment of < 17°C with $< 70\%$ relative humidity is necessary.

Structural damage to the seed as a result of drying can also be a problem. Superficial cracking of the seed coat is common in many species and frequently occurs in the field before drying. It may not always be visible to the naked eye but can be detected with appropriate staining techniques and microscopic examination. Seed cracking can result in damage from excessively rapid imbibition during germination tests, in particular for low moisture content seed. This problem occurs in a diverse range of species (e.g. cotton, peas and sorghum) but can be successfully avoided when germinating seed by first allowing seed moisture content to

rise slowly without allowing the seed to be in contact with liquid water. This is done by placing the seed in a humid atmosphere for some time before setting the seed to germinate (whether in the laboratory or the field). Drying seed at the conditions recommended in this appendix will minimise the increase in the incidence of seed coat cracking above that caused during drying on the plant.

In seed of many leguminous species a different phenomenon occurs during drying; namely the seed coat becomes impermeable to the ingress of moisture, i.e. hardseededness. This makes it difficult to germinate the seed since they fail to imbibe, but a variety of laboratory techniques are available to overcome this problem.

In soyabean a further problem is caused by cleavage damage, where the seed coat splits creating a gap between the cotyledons. If the seed are handled carefully and humidified before testing for germination then damage to the resultant seedlings can be avoided, but the seeds are extremely brittle in this situation and the problem is better avoided completely. If soyabean seeds are dried at low temperatures, as will be recommended here, cracking will only start to occur on any scale at about 9% moisture content (wb) if dried rapidly. If a slightly higher relative humidity of 40% is employed then the seed will dry more slowly and drying to about 8% (wb) without cleavage is possible. Consequently it is suggested that the preferred conditions for storage might be relaxed for soyabean and that they be stored at -20°C with 8%-9% moisture content (wb). The figures provided here relate to American soyabean varieties produced in the USA. It may be necessary to modify this recommendation for accessions of different genotypes produced elsewhere.

5. Recommendations for seed drying conditions

From the foregoing it should be clear that an environment of 10°-15°C with 10%-15% relative humidity is optimal for drying the small-seeded species. Accordingly it is recommended that a seed drying room be provided in which such an environment is maintained, with adequate ventilation (10 air changes per hour) and a suitable shelf layout where seed are exposed to this environment in thin layers. Seeds must not be left to dry in deep layers or sacks unless ventilated at 0.0015 m^3 per second per kg of seed (1.5 cfm per lb). A suitable fan for this purpose must be capable of operating against a static pressure of 2 cm (wg).

For many gene banks an additional temporary store may be necessary to enable peak seasonal inputs to be held temporarily prior to drying. In view of the points outlined in the preceding section an environment of 17°C with 40%-45% relative humidity is recommended, particularly since this environment can be economically maintained using conventional air-conditioning equipment (refrigeration dehumidifiers, see next section) with air flow rates of 0.2-0.4 m/s.

In addition this environment can function as the first stage in a two-stage drying procedure where this is necessary for larger-seeded species, e.g. peas and maize. (Of course it could also function as such for the small-seeded species.) Equilibrium moisture contents for such species with relatively low oil contents will be about 10%-12% (wb). Moreover very wet seed will dry comparatively quickly; for example, even maize seed would be expected to dry from 25% to 15% moisture content (wb) in approximately 36 hours (using the equations provided in section 3). For these species the second-stage environment could be as high as 30°C with 10% relative humidity without causing a serious loss in viability, provided the air flow velocity is

2.5 m/s or greater.

6. Dehumidification equipment

a) Sorption systems

Moisture can be extracted from the air by chemicals in two ways, by absorption or adsorption (see Glossary, Appendix 12). Lithium chloride is an absorbant whereas silica gel is an adsorbant. In both cases the materials are regenerated for re-use by heating. Continuous dehumidification is usually achieved by passing the moist air through one half of a rotating drum containing the desiccant. Heated air is passed through the other half to regenerate the desiccant, and exhausted to the outside air. An alternative procedure is to switch between two separate machines, one dehumidifying while the other is regenerated (i.e. a batch system).

It is recommended that a rotary absorption dehumidifier with secondary refrigeration equipment be used to provide an environment of 15°C with 10% relative humidity to dry seed. A typical system, capable of extracting 8 kg H_2O per day at 15°-20°C, costs about US $3,600, but in a major gene bank facility this would need to be duplicated. It is recommended that a rotary absorption dehumidifier be used without refrigeration to provide an environment of 30°C with 10% relative humidity for the second stage in the two-stage drying of large-seeded species. A typical unit currently costs about US $2,500.

A less expensive system might be required if a seed drying room is not yet available. A simple approach is to use relatively large amounts of desiccants in a closed container, and regenerate batches of the desiccant as required.

Silica gel has a moisture adsorption capacity which increases linearly from zero by 0.0066 kg H_2O/kg dry silica gel per 1% rh increment at 20°C up to 50% rh.

The weight of desiccant necessary can be determined in a manner similar to that described below for barley.

Suppose 0.5 kg of barley is to be dried from 16% to 5% moisture content (wb). At 16% the dry weight of seed is 0.420 kg and the weight of water 0.08 kg. At 5% the weight of water will be only 0.022 kg and thus the required loss of moisture is 0.058 kg. At 20°C the equilibrium relative humidity for barley seed at 5% is approximately 12% rh. Now the moisture holding capacity of silica gel at 12% rh is

$$= 12 \times 0.0066 \text{ kg/kg} = 0.0792 \text{ kg/kg}$$

Thus the weight of silica gel required is

$$= 0.058/0.0792 \text{ kg} = 0.73 \text{ kg}$$

Drying can either be achieved in a closed container without air movement (although this will require considerable time with large seeds, see Figure 3.5) or in a closed system with air being forced through the seed. In either case it should be noted that the uptake of water by the desiccant is associated with heat

release, which results in a slight increase in temperature, but this is partly redressed by evaporative cooling of the seed. The silica gel can be regenerated by heating at 175°C or 125°C for 6 or 16 hours respectively, but must be allowed to cool (in a moisture-proof container) to ambient temperature before re-use.

b) Refrigeration systems

It can be seen from the psychrometric charts (Figure 3.1) that the maximum saturated air moisture content is very low at low temperatures. Moreover, water vapour can be condensed out of the air by cooling below the dewpoint temperature. This is how refrigeration dehumidifiers work. The cold saturated air is then reheated, using the latent heat removed by condensing air moisture, to provide a lower relative humidity. The mode of operation can be understood most easily from the psychrometric chart (Figure 3.1A). Air at 17°C with 70% relative humidity has a moisture content of 0.0086 kg H_2O/kg air. The dewpoint temperature for this moisture content is 14°C. Consequently, if the air is passed across an evaporator maintained at 5°C moisture will condense out of the air. The saturated air moisture content at 5°C is 0.0054 kg/kg, and thus the moisture will be extracted at the rate of 0.0032 kg/kg. If this air is now reheated to 17°C the air moisture content will remain at 0.0054 kg/kg and thus the relative humidity of the reheated air will be about 45%.

Note that a higher moisture extraction rate would be achieved if the air input were at a relative humidity above 70%. Quoted moisture extraction rates are often given for 95% rh, and these rates will not apply to seed drying applications. Consequently refrigeration dehumidifiers operate very effectively where the air moisture content is high, that is in warm, humid conditions. They are generally considered to be uneconomic for air temperatures below 15°C or relative humidities less than about 40%-45%. They are relatively cheap to install and have large capacity fans which give favourable air circulation. It is recommended that refrigeration dehumidifiers be used to provide temporary storage prior to drying, where this is required, and to provide first-stage drying of moist seed of large seeded species (para. 18). One useful facet of this equipment is that the condensed water vapour can be collected to provide a direct measure of "drying performance". Refrigeration dehumidifiers, fitted with reheat coils (electric heaters or hot refrigerants), are sometimes used for controlling the air relative humidity in working collection coldrooms operating between -10° to 5°C, with or without open storage, but these are outside the framework of this report. Separate, special low temperature refrigeration dehumidifiers can be used in gene bank coldrooms without reheat coils providing the moisture ingress does not exceed recommended limits.

8. Monitoring seed moisture content during drying

It will be necessary to monitor seed moisture content during drying to determine when accessions can be removed from the drying room to be first packaged and then stored. To do this by determining moisture contents regularly using standard procedures (Appendix 6) would be to waste resources - both in terms of seed and labour. Instead it is recommended that this be done by determining the initial moisture content and weight, and then subsequently weighing the seed during drying.

The change in weight and moisture content by the addition or removal of moisture is given by

$$[(100 - Mo)/100] \times Wo = [(100 - Mf)/100] \times Wf \qquad (3.11)$$

where Wo and Wf and Mo and Mf are initial and final weights and moisture contents (%, wb) respectively.

Thus the weight of an accession when dried is given by

$$Wf = Wo \times (100 - Mo)/(100 - Mf) \qquad (3.12)$$

For example for 1 kg of seed at 16% moisture content, the required weight of seed when dried to 5% moisture content

$$= 1 \times (100 - 16)/(100 - 5) \text{ kg} = 0.884 \text{ kg}$$

Thus the 1 kg sample is placed in the drying room and weighed regularly, but only removed when the weight has fallen to 0.884 kg. Of course this system for monitoring moisture content can only be used if care is taken not to spill seed, otherwise the true seed moisture content will be underestimated. Once sufficient experience has been obtained with the drying room it may be possible to dispense with the regular weighings and rely instead on standard timings in the drying room for seed of each species. This would enable the maximum number of accessions to be put into store with the minimum of effort.

APPENDIX 4. SEED DETERIORATION AND STORAGE ENVIRONMENT

Orthodox seeds in air-dry storage deteriorate. Eventually this deterioration results in loss of viability of individual seeds - that is the seeds fail to germinate under optimum conditions. Within an accession individual seeds die at different times. Consequently the viability of an accession is assessed as the proportion (expressed as a percentage) of viable seeds. The proportion of live seeds within an accession is also an indicator of the accumulation of deterioration within the seeds which remain alive. For example as percentage viability declines the time taken by surviving seeds to germinate increases quite markedly[1], and the surviving seeds are also more likely to fail to germinate in sub-optimal environments[2]. There is also, however, a more subtle aspect to this deterioration within the surviving seed that must be noted for genetic resources conservation; loss of viability is correlated with an accumulation of chromosome damage in surviving seed[3,4] and with hereditable damage in succeeding generations[5].

Although seed deterioration in air-dry storage can thus be considered as a form of ageing it does not only occur as a function of time. The rate of deterioration is a positive function of both seed moisture content and temperature. Thus deterioration occurs relatively slowly at low moisture and temperature, which is why the preferred conditions for long-term storage (para. 6) are considerably cooler and drier than those of normal commercial practice.

The decline in percentage viability observed in accessions of orthodox seeds under any constant storage environment is sigmoidal. This can be described by a negative cumulative normal distribution which indicates that the life-spans of individual seeds are distributed normally. Consequently these sigmoid survival curves can be linearised by converting percentage viability values to their corresponding probit (or normal equivalent deviate) values. (Figure 4.1 (scale h) provides a conversion scale that can be used for this purpose). It has been shown that the survival curves of different accessions of a species in the same constant storage environment have the same slope when transformed in this way[2,6]. Thus this device simplifies the prediction of loss in seed viability and is used in an equation which has been developed to predict the relationship between percentage viability (v, expressed as a probit), time in storage (p, days), seed moisture content (m, % wb), and temperature (t, °C). The equation[7],

$$v = K_i - p/10^{K_E - C_W \log m - C_H t - C_Q t^2} \qquad (4.1)$$

can be used for any accession of a species for which the constant values K_E, C_W, C_H and C_Q have been determined. The constant K_i is a constant for the accession, and its value can be estimated by taking the probit value of the result of an initial viability test.

Since the values of the four constants K_E, C_W, C_H and C_Q are the same for all accessions of a species[6,7] the loss in probit viability $(K_i - v)$,

where $$(K_i - v) = p/10^{K_E - C_W \log m - C_H t - C_Q t^2} \qquad (4.2)$$

after a given period of storage is also the same for all accessions of a species if they are stored in the same environment (see Figure 4.1). This is because the various constants and variables on the right hand side of the above expression have the same values for all accessions. As a consequence it is possible to compare directly different storage environments and to determine the loss in probit viability for all accessions in thos

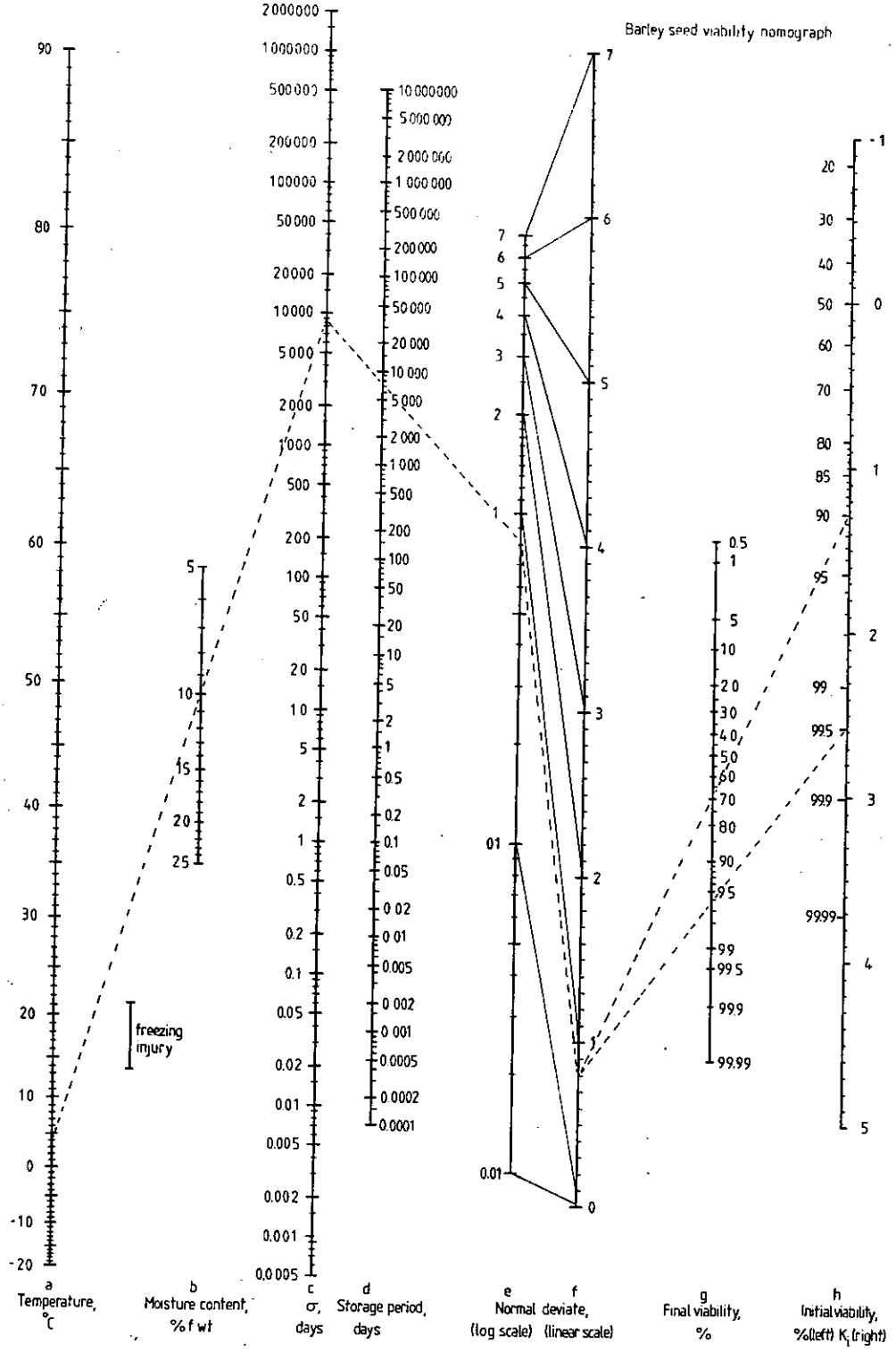
environments. Moreover where seeds experience more than a single constant environment during storage, the total loss in probit viability can be obtained simply by summing up the individual values for each period in each separate environment. In this way it is also possible to compare the likely damage caused by different drying environments, for example to consider whether a short period of exposure to a high temperature or a longer period of exposure to a lower temperature results in the smaller loss in probit viability even though they both dry seed to the required moisture content. This is considered further in Appendix 3.

At present, values of the four species constants have been determined for only five species (Table 4.1). Seed viability nomographs, constructed from equation (4.1) and constant values from Table 4.1 (Figures 4.1 and 4.2), enable seed longevity calculations to be made easily and rapidly. These nomographs also illustrate the general form of the relationship between storage environment and loss in seed viability. Roughly speaking the logarithm of the storage period over which a given loss in viability takes place decreases linearly with increase in either the temperature or the moisture content of storage; hence the values for storage period on scale d of the nomographs are plotted on logarithmic scales. However this relationship is only approximate for it can be seen that the temperature (a) and moisture content (b) scales are not in fact linear.

On the temperature scale (a) of these nomographs it can be seen that the divisions (1°C) are closer together the lower the temperature. This is indicative of a diminishing temperature coefficient; the lower the temperature the smaller the increase in the log value of longevity that results from further reduction in temperature (although in terms of an ordinary time scale there continues to be a greater improvement in longevity with each unit decrease in temperature). Results from very many orthodox species - including cereal, flower and vegetable species - indicate that over the same temperature ranges the longevity of all orthodox species have a similar relative response to change in temperature[2]. (Differences in the viability constant values of C_H and C_Q between the three grain legume species in Table 4.1 reflect the difficulties in accurately quantifying this non-linear relationship and are not significant[6].) Consequently scales (a) and (c) can be used to demonstrate for all orthodox species the relative benefit resulting from a reduction in storage temperature (at a constant moisture content). For example a reduction from 1 to -20°C will result in a four-fold (approximately) increase in longevity (Figure 4.1). Thus a store at -20°C would be expected to require only one quarter of the manpower and land to monitor and regenerate accessions compared to a store at 1°C, and this benefit can be obtained for minimal additional expenditure (Appendix 9). It is believed that -20°C

Figure 4.1. Seed viability nomograph for barley[7] which solves the viability equation (4.1) for the constant values provided in Table 4.1. The nomograph may be used in many ways. For example, the broken line illustrates a calculation to predict viability after 20 years storage at 4°C with 10% moisture content (wb). Place a ruler at 4°C (scale a) and 10% mc (scale b), and note the resultant value (8,400 d) on scale c. Use this point on scale c as a pivot and move the ruler to indicate 7,300 d (20 years) on scale d. Note the corresponding value (0.8) on scale e and transfer this value to scale f. Finally, by connecting this value on scale f with points on scales g and h by ruler, the viability of an accession initially 99.5% viable (scale h) would be predicted to fall to 96% (scale g) whilst the viability of another accession initally 90% viable would be predicted to fall to 70% after the same combination of temperature, moisture and storage period. Scale h can also be used to convert percentage viability values (left hand side of the scale) to the appropriate probit values (right hand side of the scale) or vice-versa. For example, if percentage viability is 98% then probit percentage viability (v, equation 4.1) is 2, and when probit percentage viability is -0.5 percentage viability is slightly more than 30%. Scales e and f provide an estimate of loss in probit percentage viability, that is $K_i - v$ (equation 4.2). This value is the same for different accessions when stored for the same period in the same environment - as can be seen in the example calculation. Consequently this value provides a rational basis that can be used to compare the effect of different environments on the loss in viability to be expected for all accessions.

Figure 4.1


Barley seed viability nomograph
a
Temperature, °C
b
Moisture content, % f wt
freezing injury
c
σ, days
d
Storage period, days
e
Normal deviate, (log scale)
f
(linear scale)
g
Final viability, %
h
Initial viability, %(left) K_i (right)

represents a sensible compromise temperature for long-term storage. Below -20°C it is uncertain by what factor seed longevity may be increased by further reduction in temperature whereas below this point the costs of providing cold storage can increase sharply.

On the moisture content scale (b) of the nomographs it can be seen that the divisions (1% moisture content) are further apart the lower the moisture content. That is the increase in longevity which results from reduction in moisture content is more than exponential. Five ± 1% moisture content (wb) was chosen by FAO as the preferred moisture content of storage rather than a lower value because of the practical difficulties encountered in drying seed of certain species below this level (Appendix 3) and there are some species whose seed may be damaged by extreme desiccation. However, for those species whose seed can be dried below this level without difficulty the resultant increase in longevity can be substantial (para. 11) and very worthwhile.

The response of seed longevity to storage at different moisture contents varies between species (e.g. Table 4.1). This appears to be mainly a function of seed oil content; the longevity of oily seeds (e.g. onion and soyabean) is less sensitive to a given difference in moisture content than seeds with a very low oil content (e.g. barley). Hence in Table 4.1 the value of constant C_W for onion and soyabean is similar but considerably less than the value for barley. However, because the potential longevity of seeds with a high oil content is relatively low (compare the values of constant K_E) it is vital that these species seed be dried despite the smaller relative beneficial effect.

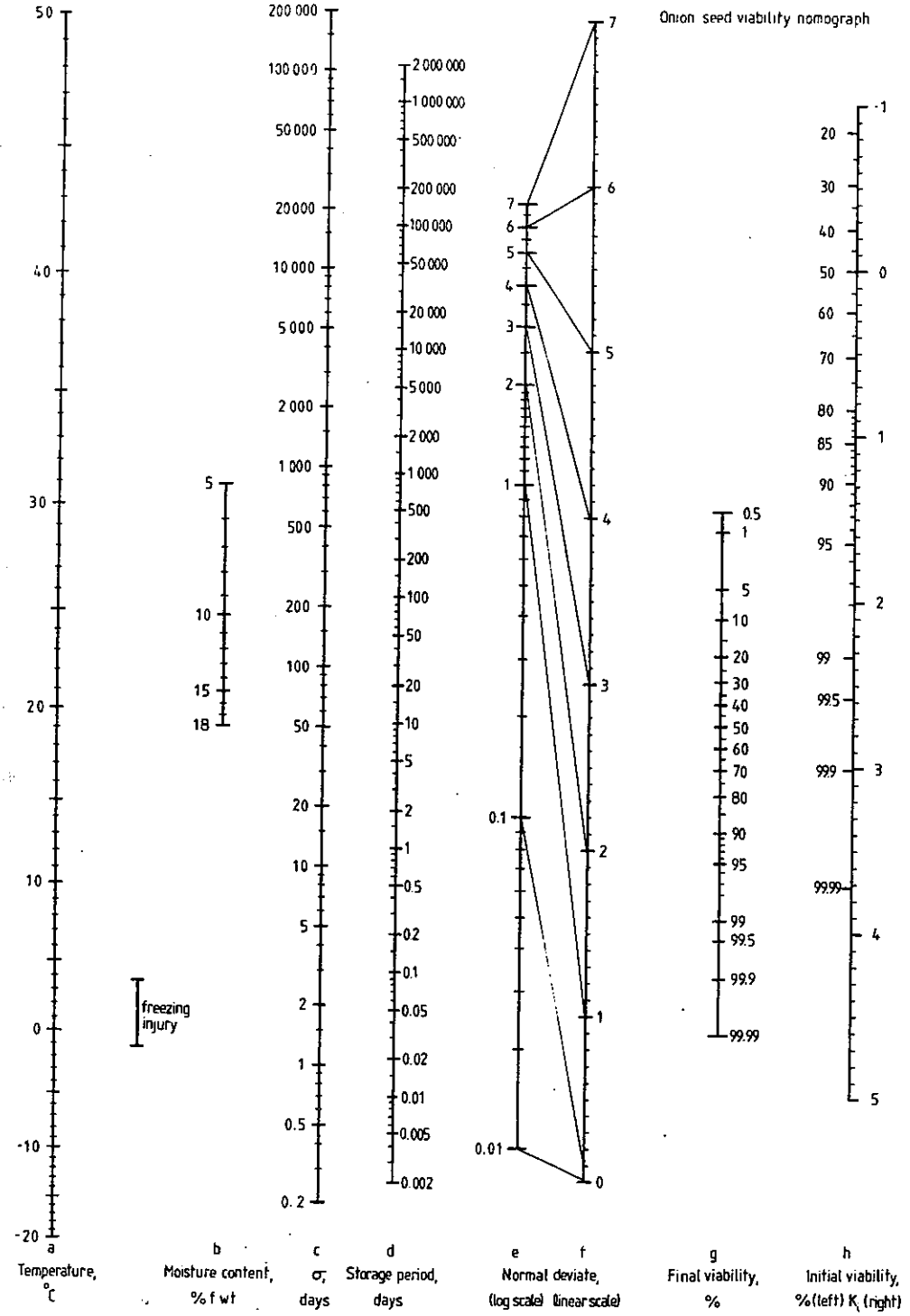
Since seed longevity will differ between species in a given storage environment (due to differences in the values of constants K_E and C_W between species) it is difficult to define medium-term storage conditions. It is suggested here that the nomographs for barley (Figure 4.1) and onion (Figure 4.2) are used as some indication of the required environments for species with good and poor storage characteristics respectively. Many species are likely to fall within the range represented by these two species.

One aspect of the storage environment that is excluded from the preceding equation is the influence of oxygen partial pressure on seed longevity. At very high temperatures there is evidence that increase in oxygen partial pressure reduces seed longevity. Conversely at very high moisture contents increase in oxygen partial pressure increases longevity. However at combinations of low temperatures and low moisture contents it would appear that the effect of oxygen is negligible and difficult to discern. Hence in long-term storage packaging in inert atmospheres has little or no effect on longevity and is unnecessary[8]. Sealing seed in air at atmospheric pressure is satisfactory and is simple. Vacuum sealing is a more complicated technique which is at least as satisfactory and is used by some seed banks. It is doubtful, however, whether the additional costs are worthwhile.

It can be seen from the nomographs that, even within a species, different accessions (which have different K_i values) have different longevities in the same environment. Thus in a seed bank it is expected that some accessions of a species will require regeneration much earlier than other seed lots. Procedures for

Figure 4.2. Seed viability nomograph for onion[2] which solves the viability equation (4.1) for the constant values provided in Table 4.1.

Figure 4.2


Onion seed viability nomograph
a
Temperature, °C
b
Moisture content, % f wt
freezing injury
c
σ, days
d
Storage period, days
e
Normal deviate, (log scale)
f
(linear scale)
g
Final viability, %
h
Initial viability, %(left) K (right)

managing accessions within seed banks are briefly considered here insofar as they affect the design and provision of long-term seed storage facilities. A separate IBPGR publication is being prepared which will examine these procedures in detail.

An accession represents a population of seeds. Within the population genetic mutation accumulates in the surviving seeds as viability declines. In addition different genotypes within a species often show different periods of longevity; consequently when an accession is genetically heterogenous, there will be a tendency to select against short-lived types as the accession loses viability. To reduce the accumulation of mutation and minimise genetic selection due to loss of viability in storage it is necessary to regenerate a sample from the accession to produce fresh seed before viability has fallen very far, i.e. to the regeneration standard[9]. However any estimate of viability derived from a sample of the accession is subject to error, the greater the sample size the less the error, but the greater the sample size the sooner the accession will be depleted.

To maximise the accuracy of tests whilst minimising the total number of seeds used when monitoring accession viability the IBPGR Seed Storage Committee recommends the adoption of a sequential test procedure in which only a small number of seeds are tested at one time, but a sequence of tests comprise the complete procedure - the sequential probability ratio test[9]. This requires a succession of separate samplings from the accession over a period of weeks. To minimise the possibility of the accession bulk increasing in moisture content during these samplings it is suggested that they are carried out in the dehumidified packaging room (para. 52). In addition the frequent sampling during the sequential test procedure will waste containers unless re-usable containers (e.g. laminated aluminium foil packets or screw-capped cans or jars) are used. *

Provided the preferred conditions are met, the rates of decline in viability in most species are likely to be slow. The monitoring tests are designed to indicate when accession viability has fallen to the regeneration standard. Estimates of regeneration intervals (the time taken for viability to fall to the regeneration standard) will soon be available for many of the major crops and for these species it should be possible to base the monitoring interval (the storage period between each monitoring test) on such estimates. These estimates differ between accessions[9]. For safety, and to allow for errors, the monitoring interval will be some fraction of the estimated regeneration interval. In other species no estimates are likely to be available in the near future. In such cases monitoring intervals will have to be guessed, and subsequently modified in the light of experience. For many of these accessions monitoring intervals will probably approach 10 years. However, in a newly established store it will probably be advisable to begin with a shorter monitoring interval (perhaps on a small proportion of accessions, preferably those of low initial viability) in order to check that the system is working as expected, the first tests being carried out after 3 to 5 years. Clearly more laboratory facilities for seed testing will be required where the gene bank is maintaining relatively short-lived species (e.g. onion) than one of the same size maintaining relatively long-lived species (e.g. cereals).

*Research and development work after the original report was completed has provided an improved sequential probability ratio test most suitable for application in genebanks. This test, the truncated sequential probability ratio test, is described in detail in the following paper.
R.H. Ellis and J. Whitehead, 1987. Open, truncated and triangular seed testing procedures. Seed Science and Technology, 15, 1-17.

Table 4.1 Values of the seed viability constants

Crop	K_E	C_W	C_H	C_Q
barley [7]	9.983	5.896	0.040	0.000428
chickpea [6]	9.070	4.829	0.045	0.000324
cowpea [6]	8.690	4.715	0.026	0.000498
onion [2]	6.975	3.470	0.040	0.000428
soyabean [6]	7.748	3.979	0.053	0.000228

References

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IBPGR PREFERRED CONDITIONS FOR LONG-TERM STORAGE OF SEED ACCESSIONS: AN UPDATE

When the preferred conditions for the long-term storage of seed accessions (-18°C, or less, with 5 ± 1% moisture content) were recommended by IBPGR in 1976 they were to some extent a compromise based on the limited information available at that time. They were a compromise simply because although it was known that reduction in seed storage temperature or moisture content increased longevity, the quantitative nature of either relation and the extent to which either relation was subject to limits was not known then. The evidence now available suggests that those recommendations represent remarkably effective compromises with regard to both variables, bearing in mind the wide range of orthodox species to which they have been applied.

Temperature. As previously noted (page 58), the semi-logarithmic relation between temperature and seed longevity is curved - such that each further reduction in seed storage temperature has progressively less effect in relative terms. A recent comparison[1] among eight species with very contrasting taxonomy (covering four of the ten super-orders of flowering plants) showed no significant differences in the values of the viability constants C_H and C_Q. That is, the relative effect of temperature on the longevity of seed of all eight contrasting species was the same and so the conclusion (see page 58) that, over the same temperature ranges, the longevity of seed of all orthodox species have a similar relative response to temperature is further supported[1]. The values of C_H and C_Q derived recently from the very comprehensive set of data on which this conclusion was based can be applied to provide our best estimates to date of the relative benefit to the longevity of seed accessions from various reductions in seed storage temperature. Thus, it is estimated that longevity is increased by a factor of almost 3 if storage temperature is reduced from 20°C to 10°C; by 2.4 from 10°C to 0°C; by 1.9 from 0°C to -10°C; and by only 1.5 from -10°C to -20°C.

Moisture content. Further research since the first printing of this report has confirmed that in some species the logarithmic relation can continue down to very low moisture contents. For example, in sesame (Sesamum indicum L.) the logarithmic relation has been shown to continue down to moisture contents as low as 2%[2]. The effect of a reduction to this value from 5% moisture content is considerable: about a forty-fold increase in longevity[2]. Indeed, it has been pointed out that ultra-dry seed storage has the potential to reduce the costs of conservation by reducing the need for refrigeration since, for example, calculations for sesame seed accessions show that longevity at 20°C with 2% moisture content is expected to be similar to that at the IBPGR preferred conditions of -20°C with 5% moisture content[2].

There is, however, a low-moisture-limit to the negative logarithmic relation between seed longevity and moisture[3-5]. This value varies greatly between orthodox species: for example, from around 2% in very oily seeds such as groundnut (Arachis hypogaea L.)[5] and (presumably) sesame, to just above 6% moisture content in very starchy seeds such as pea (Pisum sativum L.)[4]. Further reduction in moisture content immediately below these respective values of the low-moisture-limits does not, or does not greatly, influence longevity[3-5]. Nevertheless, from the point-of-view of seed storage these values represent the practical limits for desiccation.

Although the variation in the above gravimetric values among species may at first seem disconcerting, it does in fact reflect considerable uniformity amongst all the species so far investigated. Since the composition of seeds varies amongst species of seed (especially in percentage oil content), so does the relative humidity in equilibrium with any given moisture content (see Appendix 3). The common feature of the different moisture contents amongst species below which there is no further improvement in seed longevity is that they are all in equilibrium with approximately 10.5% r.h. [4,5]. Accordingly, we are currently investigating whether advice on recommended seed storage moisture contents should be specified not gravimetrically but as an equilibrium relative humidity of approximately 10-11%. A small pilot study with a genebank has recently begun to test the advisability of such a recommendation in practice. In the meantime it would be sensible to interpret the earlier recommendation of 5 ± 1% moisture content as around 6% for those species with starchy seeds which are difficult to dry much below 6% moisture content in the recommended drying regime of 15°C with 10-15% r.h. (see paragraph 17, pp. 4-5), but 4% for those species with oily and very oily seeds which dry comparatively rapidly to 4% moisture content in these conditions.

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APPENDIX 5. THE DETERMINATION OF SEED MOISTURE CONTENT

The moisture content of a sample of seed is determined as the loss in weight of a sample when it is dried. In seed technology it is expressed as a percentage on the wet or fresh-weight basis (wb, see Appendix 12). However, since the loss in weight of a given sample of seed will vary according to the temperature and duration of the drying treatment it is necessary to use standardised drying procedures in order to obtain results that are both repeatable and reproducible and that can therefore be used for comparative purposes. The methods prescribed by the International Seed Testing Association are designed to reduce oxidation, decomposition or the loss of other volatile substances whilst ensuring the removal of as much moisture as possible. They are widely applied in seed technology and are thus most suitable for gene banks. The ISTA rules for the determination of moisture content are summarised below. The rules and annexes to the rules for determining moisture content can be found in *Seed Science and Technology*, 1976, Volume 4, pages 40-43 and 160-163 respectively, with subsequent amendments in Volume 9, pages 307-8 and 334[1].

1. The first imperative throughout any drying procedure is to minimise the period during which the sample is exposed to the laboratory atmosphere.

2. Determinations are carried out in duplicate on two independently drawn samples from thoroughly mixed accessions.

3. It is necessary to predry very moist seed. The samples are weighed in previously weighed containers, predried and then reweighed to determine the loss in weight (moisture). If moisture content is more than 30% samples are predried overnight on the top of a heated oven. If moisture content is between 17% to 30% and the seeds are large, samples are predried at 130°C for 5-10 minutes followed by 2 hours exposure to the laboratory.

4. Large seeds are then ground. (A list of species for which grinding is obligatory is provided in the annexes to ISTA rules.)

5. The drying containers are made from non-corrosive metal or glass, with snug fitting covers to minimise gain or loss of moisture. Before use they should be dried in an oven at 130°C for 1 hour and then allowed to cool in a desiccator.

6. 4.5 to 5 g[2] of sample are weighed into previously weighed containers. Seeds with a high oil content and seeds of all tree species are dried for 17 ± 1 hours at 103° ± 2°C. Seed of other species are dried for

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[1] The amendments have been consolidated in Amendments to International Rules for Seed Testing 1976, 54 pp, Zurich, 1981.

[2] When the supply of seed is severely limited, it may be impossible to spare the 9-10 g prescribed by ISTA. However satisfactory, but less accurate, determinations can often be made with as little as 1 g of seed (2 replicates of 0.5 g each). If this is done it is necessary to use a four decimal place balance and to reduce to an absolute minimum the periods during which samples are exposed to ambient rh.

1 h at 130°-133°C, except maize and other cereals where the exposure period is 4 h and 2 h respectively. The drying ovens must be force draught ventilated. After drying the containers are covered, cooled in a desiccator for 30-45 minutes, and then re-weighed.

7. Moisture content (wb) is calculated as loss in weight/original weight and expressed as a percentage to one decimal place. If M_1 is the weight of the container (with cover), M_2 the weight of container and contents before drying, and M_3 the weight of container and contents after drying, then the moisture content (%)

$$= 100 \times (M_2 - M_3)/(M_2 - M_1) \quad (5.1)$$

In a two stage drying procedure the moisture content is calculated from the results obtained in the first (predrying) and the second stages of the procedure as follows. If S_1 and S_2 are the percentage loss of moisture in the first and second stages respectively, then the original percentage moisture content is:

$$M_W = (S_1 + S_2) - [(S_1 \times S_2)/100] \quad (5.2)$$

Although seed moisture content is expressed on the wet basis in agriculture and seed technology, in other fields it may be expressed on the dry basis (see Appendix 12). The percentage seed moisture content value determined on the wet basis (M_W) can, if it is wished, be converted to the equivalent value on the dry basis (M_D) using the following expression

$$M_D = 100 \times M_W/(100 - M_W) \quad (5.3)$$

Conversely moisture contents expressed on the dry basis can be converted to the equivalent wet basis value using the following expression

$$M_W = 100 \times M_D/(100 + M_D) \quad (5.4)$$

Figure 5.1 provides an easy-to-use scale for converting moisture content values determined on either basis.

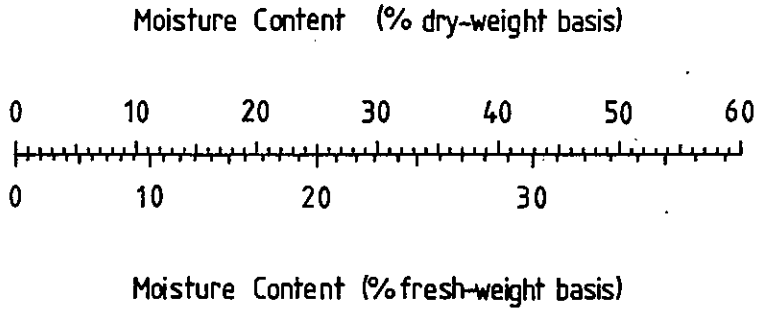


Figure 5.1. Scale for converting moisture content values between dry basis and wet basis (fresh-weight basis).

APPENDIX 6. LAMINATED ALUMINIUM FOIL PACKETS; THEIR SPECIFICATION AND PROPERTIES

Compared to metal cans or sealed glass vials, laminated aluminium foil packets are a relatively new seed packaging medium. They also differ from the first two forms of containers because they are a composite of three different materials - polyester on the outside, aluminium foil in the middle and polyethylene on the inside. The polyethylene provides the sealing property, the aluminium provides a barrier to moisture and the polyester protects the thin layer of aluminium from damage and oxidation. Sometimes there is an additional outer layer of paper, but this is not essential.

Exact packet specifications differ between manufacturers and are being improved all the time, but at present a typical laminated aluminium foil packet specification would be:

an outer layer of	17 g/m² Melinex (syn. Terylene),
	4 g/m² lacquer,
a middle layer of	33 g/m² (12 µm) aluminium foil,
	4 g/m² lacquer,
and an inner layer of	63 g/m² polyethylene.

Most packet specifications include 9-15 µm aluminium foil and 50-75 µm (46-69 g/m²) polyethylene.

A warning: It is essential to obtain packets with a robust specification from a reliable manufacturer. Paper packets lined with polyethylene or polyethylene packets painted an aluminium colour are not laminated aluminium foil packets. Neither is suitable for hermetic storage. It is the aluminium foil which provides a barrier to moisture.

In an undamaged state, aluminium foil can offer a total barrier to moisture. However, in practice, foils used in packaging will exhibit some damage. Sources of foil damage are threefold: rolling holes, conversion damage and abuse. The number of rolling holes present in virgin foil is largely proportional to the thickness of the foil. Foil 30 µm thick is 'commercial pin-hole free' whilst 40 µm foil is guaranteed hole free. However in practice thinner foil is generally used to construct laminated aluminium foil packets (presumably for malleability). Conversion damage occurs during the laminating process and is often typified by creases, or score marks in the foil packet. Such packets should be discarded. Abuse damage is that occurring during the filling, sealing, handling, transit and storage of seed packets. Hard sharp seeds may penetrate the foil. A tough grade of polyethylene as the inside ply provides foil support, resistance to seed penetration and appropriate heat sealing properties. In general, the greater the volume of the completed laminated foil packet, the more vulnerable it is to abuse damage. It is suggested that seed volumes of about 600 ml or more should be stored in more rigid containers.

It is evident, then, that the aluminium thicknesses used to construct laminated aluminium foil packets may contain small pinholes, through which water can permeate, although the polyester and polyethylene layers will minimise these. In fact moisture can enter a laminated aluminium foil packet via two routes: through the laminated body of the packet; and through the seals around the edge of the packet. In the former case this will depend upon the combined properties of the laminated foil constituents, the relative water concentrations on the two sides of the film, the temperature, and the cross-sectional area of the packet. In the

latter case this will depend upon the polyethylene thickness, the effective length of the seal, the width of the seal, the relative water concentrations inside and outside the pack, and the temperature.

To calculate the ingress of moisture into a seed packet via these two routes we have assumed a 100 mm square (internal) packet made from a laminate of 12 μm polyester/9 μm aluminium foil/50 μm polyethylene. Now 9 μm aluminium foil has between 100-8,000 pinholes per m^2. If the worst figure is assumed (8,000 per m^2), and it is further assumed that this density is doubled during the laminating process (conversion damage) and further doubled during packet filling (abuse damage) then there will be 32,000 holes per m^2. The total area of laminated foil is 0.02 m^2 (both sides of the packet) and thus the total number of holes in a packet is 640 holes. If it is assumed that the average pinhole diameter is 50 μm but that the effective permeation through a pinhole is three times the true area then the effective hole area is 3.77 mm^2. Through this area of the pack only the polyester and polyethylene constituents of the laminate are present to limit moisture ingress. Now at 25°C if outside rh is 100% and that in equilibrium with the seed is 75% the rate of water ingress through 12 μm polyester/50 μm polyethylene will be 2.2 g/m^2 per day. Therefore water ingress into the pack would be in the order of 0.003 grammes per year.

The second source of moisture ingress is through the seal. A packet 100 mm square will have a seal length of 400 mm. The thickness of the polyethylene will be 50 μm (2 x 25 μm). The seal width would be about 6 mm. Assuming the same environment as before, the water vapour permeability will be about 0.01875 g/m^2 per day for a 6 mm seal width. This gives a rate of moisture ingress through the seals of this pack of 0.00014 grammes per year.

The total rate of moisture ingress through both routes is therefore 0.00314 grammes per year. Now a packet this size could accommodate about 100 grammes of seed. If the seed moisture content was originally 6% (wb) then at this rate of moisture ingress the seed moisture content would rise to 7% (wb) after 343 years. This estimate by calculation involves several very pessimistic assumptions, and the packaging industry has been unable to substantiate them by experiment because the actual rates of moisture ingress into packets at 25°C are several orders of magnitude less than their measuring techniques can detect. Obviously, then, good quality laminated aluminium foil packets are suitable for short-term storage at ambient temperatures.

They would also seem to be suitable for medium-term and long-term storage because at lower temperatures, and in particular at -20°C, the rate of moisture ingress will be substantially less than at ambient temperatures. First the permeability coefficient of the polyester/polyethylene laminate falls with reduction in temperature. Secondly the vapour pressure of the surrounding atmosphere will be greatly reduced (see Figure 3.1B). Moreover, ageing of polyethylene at -20°C is expected to be negligible.

Gene banks using laminated aluminium foil packets must ensure that the packets are adequately sealed after filling. There are two main designs of sealing machines. Both rely on heat and pressure to form a seal with the two faces of the polyethylene inner layer.

In the first, static design, the side of the packet to be sealed is positioned between two heated plates which are closed together like jaws by the operator using a hand or foot controlled lever. The operator then releases the pressure, the jaws open, and the packet removed. The problem with this design, unless it is automated, is that the operator must ensure that equal pressure is applied to each packet for the same period.

In the second design the side of the packet to be sealed is drawn between two heated plates by rubber belts and then compressed between two contra-rotating wheels. Thus with this design the period of heating and the pressure applied is automatically the same for all packets. The pressure applied to seal packets is adjusted by altering the distance between the contra-rotating wheels. Obviously if these are too far apart the seal will not be formed. However, and rather less obviously, if the wheels are too close the aluminium foil may be scored by the wheels and water will then be able to permeate through these scores.

The temperature of the heated plates is adjustable and thermostatically controlled. The correct temperature setting for each packet specification must be determined empirically. If the temperature is too hot the outer layer of polyester will be damaged (it will shrink) and thus it will fail to protect the aluminium foil. If the temperature is too cool the inner layer of polyethylene will not melt and thus it will fail to seal correctly. The seals can be examined visually if the packets are cut in half and the two faces of the packet pulled apart. The area of the polyethylene that has been sealed (before being pulled apart) will be opaque, whereas the polyethylene within the sealed circumference will be clear. Compare the seal formed in the machine with that of another side formed by the manufacturer. Make sure that this opaque area of the seal is even in width and continuous. It is important that the sealing technique used in the store is tested at regular intervals and that the procedure is also checked whenever a new batch of packets is obtained from the manufacturer.

We wish to thank Mr. K.D. Jeffs (Metal Box Limited) and Mr. M.L. Lomax (Rubber and Plastics Research Association of Great Britain) for providing technical information on the properties of laminates.

APPENDIX 7. COLDROOM TEST PROCEDURE

Two simple tests can be conducted, prior to commissioning a coldroom, to minimise air/water vapour infiltration and to determine the effectiveness of thermal insulation.

A. Coldroom air/water vapour infiltration test

Unless properly constructed and sealed air/water vapour will enter a coldroom through badly made joints causing excessive icing of the air cooler and a loss of refrigeration capacity. A convenient way of detecting this potential problem in advance is to pressure test the structure in order to determine air leakage. Accordingly the following test is recommended for all new coldrooms once shelving has been fitted - because this can distort a false floor. The test is also suitable for certain types of drying room construction. The equipment required for the purpose is a small blower (a cylinder type vacuum cleaner is suitable) which is capable of raising (or lowering) the coldroom air pressure by approximately 30 mm wg (water gauge) and a manometer (a simple clear plastic or glass U-shaped tube [100 mm] filled to a depth of at least 50 mm) to measure pressure change.

Possible points of air leakage around pipes and cable entry and exits must be made good before commencing the test. Air-vents etc. must also be sealed.

The blower and manometer must be connected directly to the store interior and the former provided with a shut-off valve. The test is made by running the blower until a store-ambient pressure gradient of approximately 20 mm wg (i.e. the difference in height of the two water columns) has been established. If this fails to occur then the store or drying room system has a major air leakage which must be located and repaired before the test is repeated. The blower is switched off and delivery line valve closed once the desired pressure difference has been achieved. The time taken for the air pressure to fall from 19 mm to 13 mm wg (0.75 in. to 0.5 in. wg) should be greater than 7 minutes, irrespective of coldroom size, for the standard of construction to be satisfactory for seed store operation.

Small air leaks can be detected by noise, a candle flame or gas tracer technique. The total applied test pressure must not exceed 30 mm wg otherwise structural damage may occur. Note that although drying rooms may be tested in a similar manner, moisture ingress can normally be controlled by pressurization, i.e. dry room air pressure held at 2 mm wg above atmospheric pressure.

B. Coldroom heat leak test

The conditions of thermal insulation in new and old coldroom facilities can be measured, but with some difficulty, by direct or indirect refrigeration based calorimetry. A simpler and more convenient approach is to establish the heat leak rate using conventional resistance electric heaters before filling a new store or for example testing an existing coldroom prior to low temperature operation. Such tests can be undertaken using thermostatically controlled fan heaters adjusted to maintain the coldroom 10°-15°C above ambient dry-bulb air temperature. The actual electricity consumption per degree temperature difference after 4-6 days preheating gives a direct measure of the heat leak rate. Ambient air should be circulated between the coldroom exterior and the surrounding building.

APPENDIX 8. COLDROOM STORAGE CAPACITY

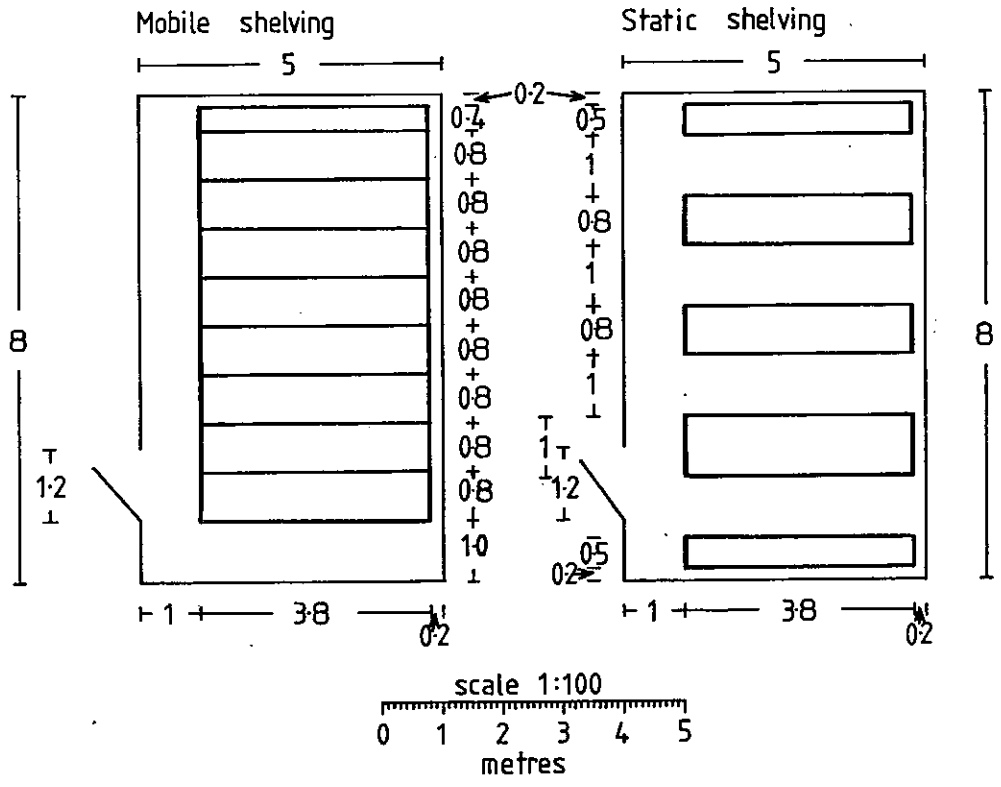
1. *Cost comparison between mobile and static shelving*

A. *CAPITAL COSTS*

There are two ways to compare the costs of providing storage space within cold stores using mobile or static shelving. Either one can compare the different areas of shelving provided by the two systems within a given coldroom volume, or the different volumes of coldroom necessary to provide the same area of shelving.

The first cost comparison between mobile and static shelving considers the relative expense of fitting out a 100 m^3 coldroom. Such a room would have an internal floor area of 40 m^2. If the shelf units are six shelves high then static shelving can provide 82 m^2 of shelf space, whereas mobile shelving can provide 155 m^2 of shelf space (Figure 8.1). Now, mobile shelving costs approximately twice as much as industrial grade static steel shelving per unit area, typically about US \$110/m^2 compared with about US \$50/m^2. The cost of a 100 m^3 coldroom with 125 mm thick insulation panels is about US \$26,500 (Table 1). Thus the

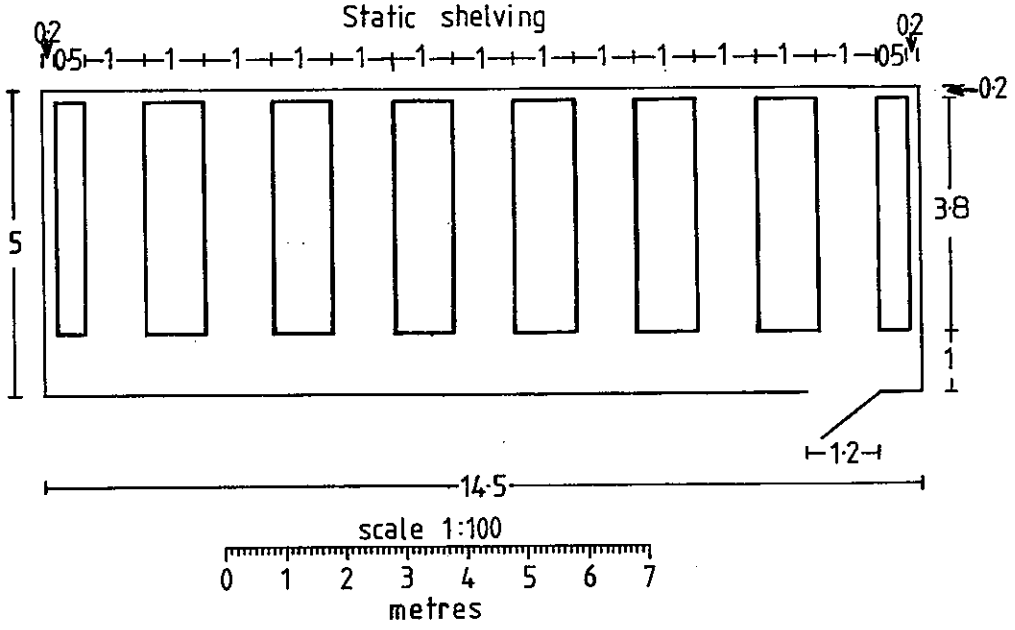
Figure 8.1. Plan of shelving systems within 100 m^3 coldrooms with 40 m^2 floor area (5 x 8 m). Passageways are 1 m wide, and the gaps between shelving and walls are 0.2 m. Left: mobile shelves. Shelving racks occupy a floor area of 3.8 m x 6.8 m = 25.84 m^2. Since there are six shelves per rack, the total shelf area is 155 m^2. Right: static shelves. Shelving racks occupy a floor area of 3.6 m x 3.8 m = 13.68 m^2. Since there are six shelves per rack, the total shelf area is 82 m^2.



total cost of coldroom and shelving is US $43,550 with mobile shelving but only US $31,000 with static shelving. However, the costs per unit shelf area are US $281/m² and US $373/m² respectively. Thus in this example the capital cost of a coldroom with mobile shelving per unit shelf area is approximately three-quarters that of a coldroom with static shelving.

The second cost comparison considers the relative expense of providing 155 m² of shelf space assuming that both systems are six shelves high. With mobile shelving this requires a 100 m³ coldroom (Figure 8.1). With static shelving a 181 m³ coldroom is required to provide 160 m² shelf space (Figure 8.2). The cost of a coldroom of this size would be about US $38,000 (by interpolation from Table 1). The cost of providing shelving would be 160 x US $50, about US $8,000 making the combined cost of shelving and coldroom US $46,000, or US $288/m² of shelf space. This figure is comparable to that for mobile shelving for a similar shelf area.

Figure 8.2. Plan of static shelving within a 181.3 m³ coldroom with 72.5 m² floor area (14.5 m x 5 m). Shelving racks occupy a floor area of 7 m x 3.8 m = 26.6 m². Since there are six shelves per rack, the total shelf area is 159.6 m²



Thus, in general, if a coldroom is to be constructed within an existing building where space is not a limiting factor the cost of providing a given area of shelf space will be very similar whether it is designed for mobile or static shelving systems.

In the case of a cold store being constructed in a new building (or a coldroom being installed in an existing building where space is limited) mobile shelving will be more economic, because the larger building will cost more to erect. Allowing a figure of US $600/m² for building costs (para. 60), the total cost of providing 155-160 m² of shelf space, including coldroom, shelving and a building to house the cold store would be about US $500/m² of shelf space for mobile shelving and US $620/m² of shelf space for static shelving.

These figures include an allowance for an air-lock and a machinery room. If the coldroom insulation is thicker than the 125 mm used in these calculations then the economic advantages of mobile shelving would be greater.

B. RUNNING COSTS

In general electricity costs per unit shelf area will be about half as much again for coldrooms with static shelving as for coldrooms with mobile shelving. Thus if the coldroom is designed for least cost operation it is essential to install mobile shelving.

2. Estimate of maximum coldroom capacity using mobile shelving

The following assumptions were made to estimate the maximum capacity of a 50 m^3 coldroom (floor area 5 m x 4 m and height 2.5 m) fitted with mobile shelving (Table 1).

1. Each accession is contained within a single cylindrical container measuring 103 mm diameter and 118 mm high (para. 26).

2. Sixteen such accessions are located upright within a removable open top drawer (as a 4 x 4 grid pattern) measuring 430 x 430 x 120 mm externally.

3. Each drawer occupies a 0.45 m run of a 0.45 m deep shelf.

4. Six mobile shelf stacks, each 2 m tall, 3.6 m long and 0.45 m wide, bolted back to back as 3 pairs for convenience, are installed.

5. Two passageways are provided, each 1.2 m wide, at right angles to each other with a 200 mm gap between the stacks and adjacent coldroom walls to allow for air circulation.

6. There is a gap of 100 mm between the lowest shelf and floor, 146 mm between each of 13 shelves and 500 mm between the shelf stack top and coldroom ceiling.

7. The total linear shelf length is 280.8 m, i.e. 13 shelves 3.6 m long, in each of 6 stacks.

8. The total number of drawers is 624 (280.8/0.45).

9. The maximum number of accessions is 9,984 (624 x 16).

Therefore the maximum calculated capacity of the 50 m^3 coldroom is 9,984 accessions (0.88 l each). Note that only 18% of the coldroom volume is effectively available for accession storage.

APPENDIX 9. ECONOMICS OF COLDROOM CONSTRUCTION AND OPERATION

a) Long-term stores (para. 6)

The design of "low temperature" gene banks differs significantly from normal commercial practice (e.g. meatstores) and consequently requires detailed examination. In a gene bank the product load is small, intrinsically valuable, with a comparatively low spoilage rate, and the expected life of both the structure and refrigeration equipment must be long. They are likely to be situated in rural areas without easy access to urban service industries. Moreover although they will be free from the usual capital and interest charges associated with new buildings they are, nevertheless, unlikely to be protected from inflation and the consequent escalation of running costs. For these reasons it is recommended that the coldroom be constructed for least cost operation.

The main considerations involved are deceptively simple. Increasing coldroom insulation thickness decreases the size of the refrigeration unit (and standby generator, if installed) and thus operational costs, but increases the original cost of coldroom construction.

A straightforward method for calculating the theoretical minimum economic insulation thickness for a modern 100 m^3 capacity coldroom (surface area of 143 m^2) held at -20°C is provided here. The basic calculations can be applied to a store of any size operating at -20°C in any ambient temperature up to a daily maximum of 40°C.

The following assumptions have been made.

1. Prefabricated polyurethane panels (k = 0.023 W/m per °C after 5 years) which range in thickness from 75 mm to 200 mm require the same amount of labour to manufacture and erect.

2. They are purchased on the basis of an inside-outside temperature difference of 45°C, i.e. a coldstore temperature of -20°C and an ambient temperature of 25°C.

3. They have a variable cost which is directly related to the thickness of insulation.

4. Two package (factory tested) refrigeration systems are installed, each of which is capable of maintaining the specified coldroom conditions. (Two systems are recommended to allow for the repair or replacement of one defective system - particularly for gene banks operating in remote regions - whilst maintaining the control temperature with the second system.)

5. The cost of the refrigeration equipment (within the range of 0.5-20 kW) can be predicted to an accuracy of ± 5% by the numerical expression given later (C_R).

6. A standby diesel power generator adequate to maintain one refrigeration unit only is provided.

7. Items such as temperature recorders, frost protection heaters, lighting, air-locks etc. present a fixed cost which is independent of the thickness of insulation and can therefore be excluded from the calculations.

8. Prices exclude freight charges, local taxes, and are f.o.b. a European port.

9. Items 1-6 comprise only 30-40% of the construction cost of a modular coldroom (erected within an existing building).

U THE OVERALL HEAT TRANSMISSION COEFFICIENT

If the specific heat conductivity, k, of polyurethane is 0.023 W/m dec. C and the insulation efficiency of polyurethane panels is only 67% (due to local heat gains along metal seams etc.) then the overall heat transmission per unit area of panel is

$$U = 0.023/S \times 0.67 = 0.0343/S \quad W/m^2 \text{ per } °C$$

where S is the panel thickness (metres).

Q THE HOURLY HEAT LEAK

The hourly heat leak for a total surface area of 143 m^2 and 45°C temperature difference is

$$Q = (0.0343 \times 143 \times 45)/(S \times 1000) = 0.2207/S \quad kW$$

Q_R THE OVERALL REFRIGERATION CAPACITY

The refrigeration plant is selected to cover the maximum daily design cooling load during a 16 h period. If it is assumed that the power requirement of secondary equipment - such as the air-cooled condenser and coldroom air cooler fans - is directly related to, and 11% of, the cooling load then the overall refrigeration capacity is

$$Q_R = (1.11 \times Q)/0.67 = 0.36567/S \quad kW$$

C THE ANNUAL ELECTRICITY CONSUMPTION

The power consumed to provide this refrigeration capacity will depend on the efficiency of the motor and the refrigeration compressor. It is expected that the refrigeration system will have a coefficient of performance for refrigeration (ratio of cooling capacity to power input) of 1.9, be driven by an 85% efficient electric motor, with fan power consumption equal to 22% of the compressor load. The annual electricity consumption for a 50% load factor (4380 h/annum) is then

$$C = (Q_R/(0.85 \times 1.9)) \times 1.22 \times 4380 = 1210/S \text{ kWh/annum}$$

C_E THE ANNUAL ELECTRICITY COST

A unit price of E US $/kWh gives an annual charge of:

$$C_E = C \times E = 1210 \times E/S \text{ US \$/annum}$$

C_I THE COST OF COLDROOM INSULATION

The basic variable cost for the polyurethane insulation panels is typically US $140/m³. Thus the cost per unit thickness is

$$C_I = 140 \times 143 \times S = 20020 \times S \text{ US \$/m}$$

C_R THE COST OF PACKAGE REFRIGERATION UNITS

The variable cost for two self-contained refrigeration systems (accuracy ± 5%) is

$$C_R = 2 \times 993 \times Q_R = 1986 \times 0.36567/S = 726.2/S \text{ US \$}$$

S_I THE THEORETICAL ECONOMIC INSULATION THICKNESS

The theoretical economic insulation thickness (S_I) is that thickness of insulation which provides the lowest combined cost of coldroom insulation, package refrigeration units and operation. Obviously the value of S_I obtained will depend on the number of years of operation included in the calculation and the expected cost of electricity in the future. If only the first year of operation is taken into account and the cost of electricity is expected to be US $0.1/kWh then the theoretical economic insulation thickness in metres (m) is found by solving for S when

$$C_I + C_R = C_E$$

Thus $$(20020 \times S) + (726.2/S) = (1210 \times E/S)$$

and therefore $$S_I = \sqrt{[(1210 \times 0.1) + 726.2]/20020} = 0.206 \text{ m}$$

This represents a minimum economic thickness since only one year's operational costs are taken into account. This thickness is double the commercial recommendations for small frozen meat cold stores. This is because two refrigeration systems and a standby generator are required for long-term seed stores and this cost is included in the calculation of S_I. Theoretically if operational costs for the first 10 years are included in this calculation the economic insulation thickness is 50% greater (i.e. approximately 0.3 m). At present this thickness is outside manufacturing limits, but nonetheless highly desirable should panels of this thickness become available.

If the expected cost of electricity is higher than the value used here then the minimum economic thickness will be greater, whereas it will be lower if ambient temperature is below 25°C. The following generalised expression should help to clarify the relative importance of the more important variables:

$$\text{least cost thickness} = \sqrt{\frac{\text{electricity cost x thermal conductivity x temperature difference}}{\text{expected life of store x insulation cost per unit depth}}}$$

b) Medium-term stores (para. 12)

The construction of coldrooms for medium-term storage can be simplified because the control temperature is above freezing point. Dual refrigeration systems and standby generators are recommended for tropical locations. However in temperate climates they are generally unnecessary, provided that more than one coldroom is operational. Accessions within an active collection maintained in a medium-term store may be sampled quite frequently; the use of static shelving and open storage would considerably ease the sampling work load. However these two measures will adversely affect running costs. First, the overall heat leak per accession is increased because, for a given number of accessions, a larger volume coldroom is required for static, compared to mobile shelving (see Appendix 8). Secondly the running costs of a dehumidifier and auxiliary refrigeration equipment (for cooling recirculated air) necessary to control the relative humidity for open storage are considerable and must be added to the main refrigeration cost.

Repeating the basic calculations made in the previous section for a 100 m^3 medium-term store, controlled at 5°C and applying a correction for improved refrigeration capacity because of the reduced temperature gradient, gives a theoretical economic insulation thickness of 0.133 m. The comparative estimated annual running cost (as specified) would be:

Long-term storage	1210 x 0.1/0.206	= US $587.3
Medium-term storage	340.53 x 0.1/0.133	= US $256.0
		Difference US $331.3

However, providing dehumidification (sorption) plus auxiliary refrigeration would cost between US $648 and $955 per annum per kg of moisture removed daily. A minimum design load of 2.5 kg/dayfor a 100 m^3 store gives an upper running cost for dehumidification of 7.2 times the difference between the refrigeration costs at the two temperatures. For sealed storage the running cost at 5°C is approximately half that for -20°C. However the difference (US $331.3) represents an insignificant proportion of the total gene bank operational costs and must always be considered in relation to the increase in seed storage life (and consequent reduction in the costs of monitoring and regeneration) that will result from storage at the lower temperature (see Appendix 4).

Since the design criteria and hence the original capital cost can influence the subsequent electricity costs for maintaining both low and medium temperature cold stores, it is recommended that quotations for new cold stores be compared on an overall economic basis, which may include the cost of monitoring and regenerating accessions, rather than the lowest price tender.

APPENDIX 10. ALTERNATIVE OR TEMPORARY SEED STORAGE FACILITIES

a) Deep-freeze cabinets

Domestic deep-freeze cabinets are entirely suitable for the preservation of small collections of seed, where major expenditure on a small purpose-built cold store is not justified. They can also provide temporary storage of air-dry seed before final drying during collection and/or to reduce the seasonal load on drying equipment.

Deep-freeze cabinets can be positioned in any suitable vacant space with the number of cabinets installed closely matching the storage requirement. Moreover, expansion of storage capacity can be readily and rapidly achieved by purchasing additional units from local suppliers. However, both the capital and running costs (per unit volume) of deep-freeze cabinets are greater than a prefabricated coldroom for large seed collections. The capital cost of deep-freeze cabinets is between US $920-2,250 per m^3 (Table 11.1) but only US $150-360 per m^3 for a basic prefabricated coldroom (Table 1). Running costs of deep-freeze cabinets are approximately double those of conventional coldrooms (per m^3) because deep-freeze cabinets have only a minimum insulation thickness for competitive commercial reasons. The break-even point where the cost of a prefabricated cold store begins to be cheaper than deep-freeze cabinets is about 10-15 m^3. However, 10 m^3 of storage space would require 17 x 600 litre capacity deep-freeze cabinets and these would occupy a considerable amount of floor space.

Generally all domestic deep-freeze cabinets are capable of maintaining the preferred temperature of -18°C or less, in ambient temperatures up to 32°C. Above this temperature it is important to obtain cabinets designed for tropical conditions. On many deep-freeze cabinets a variable controller allows temperatures within the range -18°C to -21°C to be selected and maintained. Purpose-built ice-cream freezers normally maintain a slightly colder temperature (-22°C). Either horizontal (chest) or vertical cabinets can be obtained. The latter are more economical on floorspace and readily accessible making the location and removal of accessions comparatively easy. However, they suffer from cold air fallout when the doors are opened and are generally more expensive. Consequently chest deep-freeze cabinets are superior in terms of internal temperature stability, reliability and relative absence of frost accumulation. Where floor space is not a limiting factor the purchase of chest deep-freeze cabinets is therefore advised in preference to upright cabinets. Ice-cream freezers of classic design have a light, folding inserted lid; these lids do not form very good seals with the body of the cabinet, allowing frost to accumulate inside. This design of cabinet consequently requires defrosting far more regularly than those domestic chest cabinets with a heavy cantilevered lid; the purchase of the latter design is therefore preferable.

Domestic chest deep-freeze cabinets are available in a very wide range of sizes from about 140 litres (gross) (external dimensions, height x weight x depth, 850 x 600 x 600 mm) to 600 litres (gross) (900 x 1,750 x 650). The potential capacity is difficult to define since it will depend upon the number of seeds in each accession and the size of the seeds (Appendix 2). In addition allowance has to be made for the packaging in which the accessions are sealed and a filing system within the deep-freeze cabinet to enable individual accessions to be located rapidly and retrieved. For example a 600 litre (gross) cabinet has a net capacity of some 580 litres. Using a commercially available filing system it is possible to store 162 cylindrical metal cans of 103 mm diameter, 115 mm height and an internal volume 880 ml within this capacity cabinet. This

size of container can accommodate approximately 2,000 maize seeds, 16,000 seeds of wheat, rice, barley, rye or triticale, 20,000 sorghum seeds or 25,000 oat seeds. With a smaller container of 130 ml internal volume (50 mm diameter x 70 mm height) the same filing system can accommodate 1,620 accessions. This would allow for approximately 13,000 onion seeds per accession or 25,000 brassica seeds per accession. With the large container size 162 x 880 ml of the 580 litres are used to store seeds, that is about 25% of the available volume. With the smaller container size 1,620 x 130 ml of the 580 litres are used, about 36% of the available volume. It should be possible to increase both these proportions if a purpose-built filing system is constructed which takes into account both the container size and shape and the internal dimensions of the cabinet. Figure 10.1 provides details of two methods of providing filing systems within deep-freeze cabinets. If purpose-built filing systems are constructed care should be taken to avoid insulating accessions from the walls and base of the deep-freeze cabinet, and to allow for movement of air within the deep-freeze cabinet. As a general guide we suggest that 25%-40% of the internal volume of the cabinet is assumed to be available for the accession containers when planning the size required. In addition a further allowance for future material is desirable. Details of refrigeration performance and electricity consumption should be obtained from the manufacturers. As a guide, the normal correct electrical load (single phase) will be between 500-800 W/m^3. However, the starting current is approximately six times the running current; this will limit the number of deep-freeze cabinets that can be connected to a single ring main. The daily power load is greatly influenced by ambient temperature; rising from about 6 kWh per m^3 per 24 h at 20°C to 8 kWh per m^3 per 24 h at 30°C.

Although it is assumed that the deep-freeze cabinet is to be installed in existing buildings some thought should be given to location. It is advisable to ensure that the temperature of the room in which the deep-freeze cabinet is located is not substantially greater than the external ambient temperature. For example, where possible the room should be ventilated, shaded from the sun and not contain too much additional electrical equipment that is in continuous operation. Of course these points are less important in temperate countries. Further thought should be given to the following: is the location secure; are the doors and corridors wide enough to install the cabinet (allow about 60 mm over the depth for hinges, and allow for turning room)? Finally it is worth thinking about how the deep-freeze cabinet, or the contents at least, could be evacuated in an emergency, e.g. fire. If the cabinet is equipped with heavy-duty swivel castors and located on a ground floor near a door then it should be possible for a single person to evacuate it (and thus its contents) from the building very rapidly. Most domestic chest deep-freeze cabinets are provided with a lock and key, which will prevent unauthorized persons tampering with the collection.

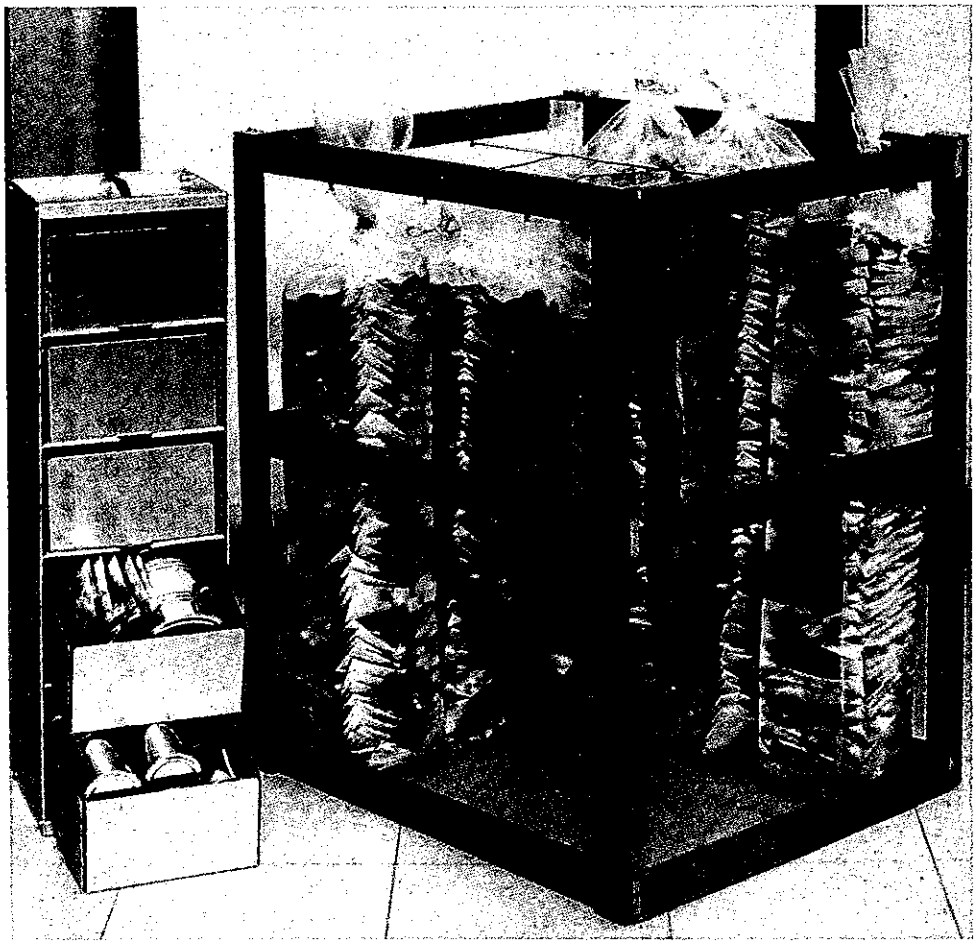
Most deep-freeze cabinets have an operational life in excess of 6-10 years depending on the refrigeration load (as reflected by the accumulated hours of compressor-motor operation) and the quality of thermal insulation and construction. Provided the seals around the lid are adequate and the cabinet is opened infrequently, chest cabinets will only require defrosting every few years, if then. It is normally unnecessary to have a spare deep-freeze cabinet in reserve for temporary accession storage during defrosting provided the accessions are in sealed containers. They should be stored at the lowest temperature at which space is available and be insulated, for example with a blanket. If accessions must be kept at ambient temperatures for a day, this will be no more serious than a delay of one day when putting the seeds into store on arrival (Appendix 4). Containers must be wiped dry of condensation before being returned to the deep-freeze cabinet.

By far the most serious factor affecting the reliability of the storage system will be the mains

Figure 10.1 Methods of filing accessions within chest deep-freeze cabinets

On the left of the photograph is a 55 cm tall aluminium file (see text). A 580 litre (net) deep-freeze cabinet can accommodate 27 of these files. Each can be separately lifted out using the handle on top (use gloves), the appropriate drawer selected and the accession required removed. The bottom drawer contains ten 130 ml capacity metal cans (see text); the drawer above contains one 880 ml metal can (see text) and five laminated aluminium foil packets. The drawers could also accommodate small screw top jars or similar containers.

To the right of the photograph is a purpose-built filing system constructed from wood and wire with polythene bags providing the vertical files. Each file contains up to 60 laminated aluminium foil packets, and there is space for 12 vertical files providing a total capacity of 720 accessions. This filing system almost fills a 210 litre (net) cabinet. The only drawback with this system is that it is comparatively time-consuming to locate and remove the required accession from the polythene bag vertical file before the remaining accessions within this file can be returned to the deep-freeze cabinet.



electricity supply. The first essential is to be aware that a disruption in power supply has occurred. This can be achieved with little cost by fitting a buzz plug to the mains lead of the freezer. If the power supply is cut the plug will emit a loud buzz for approximately 48 hours (that is until the battery within the plus is exhausted). Note that this form of device will inform you of a very local disruption in supply (for example failure of the ring main). The rate of temperature rise within the deep-freeze cabinet during a prolonged interruption to the power supply can be minimised by insulating the freezer with, for example, blankets. These should be removed immediately the power is restored to avoid damaging the refrigeration unit. A

small standby electrical generator would be advisable in locations where prolonged disruption of the power supply is expected.

Some means of determining the temperature within the deep-freeze cabinet is required. The simplest and cheapest procedure is to place a sub-zero thermometer within the deep-freeze cabinet. However this means that the deep-freeze cabinet has to be opened to determine the temperature. Consequently some form of remote temperature record might be preferred. The temperature within the deep-freeze cabinet must be checked regularly because thermostat adjustment may be required to compensate for changes in ambient temperature. A persistent and gradual rise in temperature may warn of an impending refrigeration unit failure.

Table 10.1

Guideline of approximate costs of deep-freeze cabinets and associated equipment

	US $
Domestic chest deep-freeze cabinets (-20°C)	
120 litres	270
420 litres	440
600 litres	550
Heavy duty swivel castors	30
Buzz plug (power failure alarm device)	20
-80°C to +30°C thermometer (mercury-in-glass)	20
[Thermally insulated low temperature gloves	90][1]
[Comprehensive aluminium filing system (for 600 litre cabinet)	1,200][1]
[Dial thermometer	40][1]
[1.5 kVA standby electrical generator	800][1]

[1] Optional

b) Portable cold stores

If a cold store is required urgently and the size necessary is greater than can be reasonably accommodated within deep-freeze cabinets then the purchase (or lease, if the requirement is temporary) of a portable cold store should be considered. This can be positioned close to research facilities and will not require any provision of buildings or ground preparation. A portable cold store can also provide temporary or long-term storage facilities in areas remote from major regional gene banks.

Portable cold stores are constructed for road transport with full weather protection. They are designed

to maintain temperatures down to -20°C using refrigeration equipment and thermal insulation material identical to that proposed in this report for coldroom construction (para. 34 and Appendix 9). Portable cold stores range in capacity from 25 to 55 m^3. They cost between US $800-1100/$m^3$ (f.o.b. European ports), including mobile shelving and two independent refrigeration systems. The price of portable cold stores for operation at 0°C, i.e. suitable for medium-term stores (para. 12), is 10%-15% cheaper.

International freight regulations impose certain limitations on portable cold store size. This means that additional equipment can only be incorporated within the main frame for a loss of useful refrigerated space. It is more satisfactory, therefore, to position auxiliary equipment externally and to purchase these items locally. The insulated shell would be expected to last at least 10-20 years with the refrigeration plant being progressively replaced after 10 years. A three-phase power supply - 6 to 14 kVA (about 250-350 W/m^3 of total volume) - is necessary for normal running with an allowance for a starting current of between 70-140 A depending on cold store capacity. In addition a standby generator is required which can satisfy this power demand.

c) Inexpensive medium-term stores

Where the cost of a prefabricated cold store is prohibitive, a less expensive medium-term store can be provided using local materials. The store can be built with a cavity wall from local materials. It can be insulated with a wide range of natural dried substances, e.g. cotton/rice seed hulls, oleander seed, wood shavings or corkboard (k = 0.06 W/m per °C). These should be placed in the cavity wall and above the ceiling within a heavy plastic sheet lining (15-25 cm thickness for tropical conditions within 500-1,000 gauge lining). Note that vapour barriers must always be placed on the warm side of the insulation. It is, of course, important to ensure that the building is vermin-proof, moisture-proof and not prone to flooding, in particular during the wet season. The room can be cooled to operate at 10°-15°C using a package-type air conditioning unit. This unit should be rated on a heat leak basis and not the floor area rating specified by manufacturers for domestic purposes.

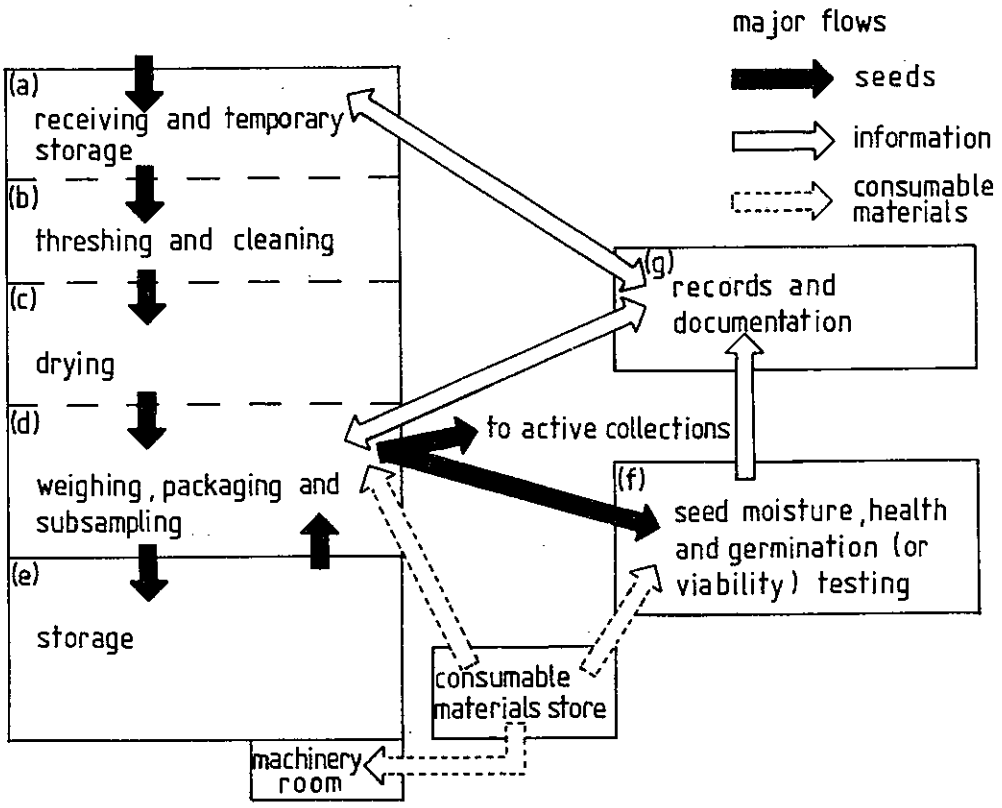
d) Natural rock stores

Limestone caves have been successfully adapted to cold storage in North America and Northern Europe. They can be inexpensively converted, because a permanently frosted zone extending at least 10 m into the rock (approximately 80 times the coldroom wall insulation thickness), formed by an initial 6-9 month period of continuous refrigeration, after which part of the refrigeration machinery is removed, gives the necessary insulation. Running costs, after the pull-down period, are comparatively low and standby power generators are not required because of the protecting frosted strata. The cave entrance must be sealed by a conventional cold store door and air-lock. However, poor access and underground streams, which cause periodic flooding, may hinder the operation of such stores.

APPENDIX 11. OUTLINE OF FLOW OF SEED, INFORMATION AND CONSUMABLE MATERIALS IN A GENE BANK

When designing the layout of a gene bank it is necessary to take into account the flow of seed, information and consumable items within the bank. An outline of these flows is provided in Figure 11.1.

Figure 11.1 Outline of major flows of seeds, information and consumable materials within a gene bank maintaining a base collection.



The space allocated to pre-storage activities (paras. 51-53) will depend on the expected peak loads of the receipt of material. Even if such matters are carefully planned it may not be possible to employ sufficient staff to deal with these peaks of input. Consequently it is necessary to plan for temporary storage where bottlenecks are likely to occur.

Since bottlenecks are likely to feed back to initial activities it is particularly important to allow for temporary storage on receipt and to minimise seed deterioration during this period. Seed arriving in a damp condition can deteriorate rapidly at ambient temperature (Appendix 4). Thus the seed should be stored at low relative humidity and preferably at a cool temperature before threshing and cleaning, particularly if there is likely to be any delay in handling the material. Similarly, threshing and cleaning should be carried out in a similar environment but this is not so important if there are no delays at this stage and, after treatment, the

seed is immediately passed on to drying. In this room a dust extraction unit should be installed if large quantities of material are to be handled. The extractors should expel dust from this area and not be of the type which attracts and collects dust. The drying room will need to be kept at a low relative humidity (10%-15% rh) and preferably at a cool temperature (10°-15°C) (paras. 17, 18), unless a two-stage drying procedure is to be used (para. 18). It is preferable that the packaging room is also maintained at 10%-15% rh (para. 52), but at a comfortable temperature for work.

If necessary activities (a) and (b) (Figure 11.1) could be combined in one room, and (c) and (d) in another. If (a) and (b) are combined then the conditions in this room would be governed by the need for temporary storage whilst providing satisfactory working conditions. Thus the relative humidity should be low and the temperature should be cool, but not cold. An air conditioning unit might be satisfactory here, say 17°C with 40% rh (see Appendix 3). If (c) and (d) are combined the provision of the seed drying facility demands a relative humidity of 10%-15% and the temperature should be controlled at about 15°C. If activities are combined within rooms it is obviously an advantage, so far as possible, to partition the various activities.

Since the seed testing laboratory is likely to contain a considerable amount of equipment (paras. 54-57), the likely location of this equipment within the laboratory should be taken into account when planning the position and layout of the laboratory. Finally the office for records and the gene bank manager (para. 58) should be in the vicinity of both this laboratory and activities (a), (b), (c) and (d) to ensure that the gene bank can function effectively.

APPENDIX 12. GLOSSARY

absorption — Process by which a fluid penetrates into the body of another material, e.g. the penetration of moisture into the body of another substance.

accession — A sample of seed representing a cultivar, breeding line or collected field sample which is held in storage for conservation.

accession size — The number of seeds contained in each accession. For genetically uniform (homogeneous) accessions typical numbers considered ideal for base, duplicate and active collections are 4,000, 1,000 and 3,000 seeds, and for genetically variable (heterogeneous) accessions 12,000, 3,000 and 5,000 seeds, respectively.

active collection — A collection of seed accessions for medium-term storage. It is assumed that a centre holding such a collection takes responsibility for regeneration, multiplication and distribution, evaluation and documentation. (Previously termed working collection).

adsorption — Process in which a fluid (e.g. moisture) is attached to the surface of a solid. In porous substances the effective surface is extended internally.

air-change rate — The ratio of the volume of air introduced or recirculated, per unit of time, to the gross volume of the ventilated space.

air cooler — A finned heat exchanger in which air and a cooling medium (refrigerant) are subject to thermal exchange.

air curtain — A high velocity air barrier limiting the exchange of air between adjacent zones.

air-dry — A widely used term which simply indicates that the seed has been dried (c.f. fresh (moist) seed) and is in equilibrium with ambient air. However the moisture content of 'air-dry seed' can vary greatly (see Appendix 3).

air-lock — A secondary cubicle, with sealed door, surrounding the entrance of a coldroom or drying room, to permit access with minimal air infiltration, arranged so that only one door is open at any one time.

ambient (condition) — The prevailing (condition) of the atmosphere surrounding the component under consideration.

base collection — A collection of seed accessions for long-term conservation. It is assumed that a centre holding such a collection assumes responsibility for viability tests, records and other administrative matters - particularly links with active collections for regeneration and evaluation. The IBPGR has requested certain institutes to accept responsibility for storing major base collections as 'world' or 'regional' repositories. (Previously termed a conservation centre).

breeders' working collection — A collection of seed accessions for immediate use by breeders. Such collections are regarded as outside the framework of genetic resources centres, but use of genetic resources by breeders will generate information relevant to evaluation and documentation. (See *active* and *base collection*).

capacity of a refrigeration unit — The refrigeration effect (kw) determined by testing under prescribed conditions.

chilling factor — Metabolic heat loss rate, relative to environmental conditions, based on human comfort.

coefficient of performance — The refrigeration effect produced per unit of work supplied based on common energy units.

current (locked rotor) — Electric motor current measured with the drive shaft in a fixed position.

condensing unit — A built-up unit of refrigeration plant components including a power driven compressor, condenser and liquid receiver.

conservation centre — See *base collection*.

defrosting — Removal, normally by heat or by mechanical means, of precipitated atmospheric moisture after it has been frozen.

dehumidifier — An apparatus for removing moisture from a substance (air), either by precipitation on reduction of temperature or by the use of a hygroscopic substance.

de-rating — Reducing the achievable performance of a machine to allow for operational conditions outside the manufacturer's test specification.

dormancy

As applied to seeds, the condition of a viable seed which prevents germination when supplied with the conditions normally considered to be suitable for germination, viz. adequate moisture, a suitable temperature and adequate aeration. Unless appropriate techniques are adopted, a dormant seed may be mistaken for a non-viable seed in a germination test.

duplicate collection

A duplicate collection of a base collection stored under similar conditions to the base collection, i.e.for long-term conservation, but at a different location as an insurance against accidental loss of material from the base collection.

duty (refrigeration)

The quantity of heat which the plant is expected to extract under specified conditions of temperature and time.

enthalpy

Thermodynamic property of a substance (air), frequently called total heat or heat content.

equilibrium moisture content/ relative humidity relationship

The relationship between seed moisture content and the relative humidity of the air. The relationship is different for different seeds and depends on their physical and chemical composition. Oil content is an important factor: the greater the oil content the lower the moisture content at a given relative humidity. The relationship also shows hysteresis and is temperature dependent (see Appendix 3).

evaporator

That part of a refrigeration system in which refrigerant is vaporized to produce cooling.

frost heave

Upthrust of the ground caused by the formation of ice layers in the subsoil.

genetic resources centres

Institutes which have accepted responsibility for the conservation of major base collections as a 'world' or 'regional' repository.

germination

Ultimately the production of a seedling from a seed, but germination begins from the first metabolic processes during imbibition. Radicle emergence is normally the first visible sign that germination has commenced. However, radicle emergence may sometimes be followed by no further growth or by abnormal growth and development. Thus according to ISTA rules, only seedlings which show normal morphology - indicating that the seedlings are capable of developing into mature plants given favourable conditions - are considered to have germinated (normal germination).

germination test

A laboratory test to estimate the proportion of germinable seeds in an accession. The result is normally expressed as a percentage. According to ISTA rules the test is carried out on a fixed sample size of 400 seeds, but an alternative procedure to determine the appropriate sample size - the sequential probability ratio test - is available to seed banks. ISTA rules (or similar) prescribe test conditions that are designed "to give the most regular, rapid and complete germination for the majority of samples of a particular kind of seed". Additional treatments may be required to remove seed dormancy.

hard seeds

Seeds which fail to imbibe when placed on moist medium because the seed coats are impermeable to water. Consequently they do not germinate. Sometimes considered as a form of dormancy.

hermetically-sealed compressor unit

A refrigerating compressor and a directly connected electric motor hermetically sealed in a welded casing.

hermetically-sealed refrigeration system

A completely sealed refrigeration system fabricated by the original manufacturer.

hermetic storage

Storage in an air-tight, moisture-proof, container.

laminated aluminium foil packets

Packets constructed of a laminate consisting of an inner layer of polyethylene, a middle layer of aluminium foil and an outer layer of polyester (usually melamine). There is sometimes an additional paper layer outside this. The packets are sealed by heat and pressure.

load (refrigeration)

The immediate refrigeration duty at the time considered.

long-term storage

The storage of seed for long-term periods as, for example, in base and duplicate collections. See '*preferred storage conditions*'. The period of storage before seeds need to be regenerated (see *regeneration* and *regeneration standards)* will vary between crops, but is expected to be at least several decades and in some cases possibly a century or more for high quality seeds of certain species. Stores operated at sub-zero temperatures have been classed as long-term stores.[1]

medium-term conservation

The storage of seed for medium-term periods as is often used for active or breeders' collections. Under the same conditions of storage the seeds of different species will have different periods of longevity. Thus it is difficult to define precisely the period envisaged by 'medium-term'. A period of 10 years or more in

(medium-term conservation - continued)

which there would be little loss of viability is generally assumed. As a guide stores which are run at temperatures between 0° and 10°C have been listed as medium-term stores.[1]

moisture content (db, i.e. dry basis)

The weight of free moisture divided by the weight of dry matter expressed as a percentage. It is normally determined by oven-drying methods (Appendix 5). The relationship between percentage moisture content dry basis and wet basis is shown in Appendix 5.

moisture content (wb, i.e. wet basis)

The weight of free moisture divided by the weight of water plus dry matter expressed as a percentage. Seed moisture content in seed testing is expressed on the wet basis. It is sometimes described as fresh-weight basis. It is normally determined by oven-drying methods (Appendix 5).

monitoring interval

As applied to accessions of seed, the period of storage between monitoring tests.

monitoring test

As applied to accessions of seed, a test (viability and/or moisture content) carried out on a sample taken from an accession in storage to ascertain whether to continue storing the accession or to regenerate.

multiplication

A representative sample of an accession is grown to increase (i.e. multiply) the supply of seed available for distribution.

open unit (refrigeration)

An indirectly (belt) driven refrigerant compressor with separate motor.

orthodox seeds

Seeds which can be dried to low moisture contents and low temperatures without damage and in which longevity is increased by so doing. Percentage viability after any given storage period can be related mathematically to temperature and seed moisture content. The majority of crop species fall into this category (see Appendix 2).

preferred storage conditions

The storage conditions recommended by FAO[2] and IBPGR[3] for long-term seed conservation, viz. storage at -18°C or less in air-tight containers at a seed moisture content of 5 ± 1% (wb).

primary dormancy

The dormancy present in a seed at harvest which is normally maintained for some time after harvest.

recalcitrant seed

Seeds which are not orthodox with regard to their storage characteristics (see *orthodox seeds*). So far only one type has been recognised: i.e. seeds which cannot be dried without injury e.g. cocoa and rubber seeds. Because they cannot be dried, recalcitrant seeds cannot be stored at sub-zero temperatures without freezing, and many tropical examples also show chilling injury (damage at about 10°C or less). No methods are so far available for the medium or long-term storage of recalcitrant seeds.

refrigerant

The medium for conveying heat in a refrigeration system, being evaporated by absorbing heat at a lower temperature, and liquefied by surrendering heat at a high temperature. Either direct - where the refrigerant is wasted (e.g. liquid nitrogen) - or indirect - where the refrigerant is recovered by recirculation (e.g. as in conventional refrigeration systems).

regeneration

The production of a fresh stock of seeds by growing plants from a representative sample of seeds from an accession in order to replenish stocks when they have been depleted (by use in viability monitoring tests, or by distribution) or when loss of viability to the level of the regeneration standard has been detected.

regeneration standard

The percentage seed viability (normally determined from a germination test) at, or below, which a decision is made to regenerate the accession as soon as possible[4].

relative humidity

The ratio of the weight of water vapour present in the atmosphere to the weight which would saturate the atmosphere at that temperature. It is also the ratio of the water vapour pressure to the saturated vapour pressure at the same temperature. (See psychrometric charts, Appendix 3).

saturated vapour pressure

The total amount of water vapour that can be held in the atmosphere at a given temperature, expressed as a pressure. The value increases at an increasing rate with temperature e.g. from 6.1 mb (millibars) at 0°C to 17.0 mb at 15°C to 42.4 mb at 30°C, and a barometric pressure of 1,000 mb. (See psychrometric charts and vapour pressure nomogram, Appendix 3).

secondary dormancy

The dormancy within a seed induced by post-harvest environment.

seed lot

A large bulk of seed of the same genotype, or genotype population, which has been produced in a single environment and subsequently treated uniformly (i.e. harvested at the same time, then dried and stored as a single bulk).

semi-hermetic motor compressor unit

A refrigerating compressor and a directly connected electric motor in a casing that can be dismantled.

sequential probability ratio test

A series of discrete tests of individuals (where in this instance an individual is a seed), or small groups of individuals, where the decision to test further individuals or stop the test depends upon the cumulative result. Thus the number of individuals tested depends upon the results obtained and is not pre-determined. A sequential germination test procedure has been developed for use in gene banks[4]. See also the reference provided on page 62.

stacks

Self-supporting, multiple shelf, mobile storage units with small wheels and tracks.

stanchion

A structural load bearing column.

thermal conductivity

The time rate of transfer of heat by conduction, through unit thickness, across unit area for a unit difference of temperature. k is the thermal conductivity and has a constant value depending upon the nature of the substance. It is expressed as watts per metre per degree Celsius.

topographical tetrazolium test

A test for viability in which moist seeds are soaked in a solution of triphenyl tetrazolium chloride. It depends on the reduction of the colourless soluble triphenyl tetrazolium chloride to a red-coloured insoluble formazan by dehydrogenases present within living tissue. The rate of this reaction is affected by pH, temperature, atmospheric pressure and the concentration of the tetrazolium salt. The interpretation of the result depends on the staining pattern of the various organs (hence topographical). The particular pattern which indicates viability has to be worked out in advance for each species and skill is needed in interpreting the result. The test takes longer than a germination test to prepare and evaluate (although the result is obtained sooner) and in many cases is not as reliable, but it may have to be used when dormancy is an insuperable problem.

viability test

A test on a sample of seed from an accession designed to estimate the percentage viability of the accession. Where dormancy would interfere with the result of a germination test special treatments are given to remove dormancy, or a topographical tetrazolium test may sometimes be used. In forestry a viability test is often synonymous with a tetrazolium test.

viable seed — A seed which is alive and capable of germination when set to germinate, providing it has lost dormancy. Seeds which do not germinate when given sufficient time to do so in a germination test are either dormant or non-viable.

working collection — See *active collection*.

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