

COMPUTER SYSTEM TO ASSIST WITH MANAGEMENT OF A TISSUE CULTURE LABORATORY

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ABSTRACT

This computer system provides information on each clone, maintains sub-culturing records over time, and enables comprehensive retrieval of information and error checking. The system has application in tissue culture laboratories handling a large number of clones and in the maintenance of gene-banks.

Keywords: subculturing records; computer databank; information retrieval.

INTRODUCTION

The management of a large number of records is required in a tissue culture laboratory. The minimum amount of information needed for each container is:

- (1) What medium is in the container;
- (2) When the medium was prepared;
- (3) What plant is in the container;
- (4) When the plant was placed in the container;
- (5) When the plant is due for subculturing to a new medium;
- (6) Where the plant containers are located, e.g., position on shelves in the propagation room, plant containers in cool storage.

Additional information on each clone, such as its special characteristics (e.g., variety name, frost- and salt-tolerance), source of original selection, and number of mother plants in the clone bank is included in the databank. Such information is useful for selecting plants adapted to specific sites or for answering requests from clients such as lists of all salt-tolerant clones in culture.

Historical records which show all of the subculturing operations performed on each plant also record which media the plant has been tested on and help in tracing errors and sources of contamination.

For laboratories handling a small number of clones and media a simple colour coding system is adequate, but where many clones are in culture, the number of records required is substantial. For example, in our laboratory about 200 clones are maintained (mainly *Eucalyptus* species) with at least 20 containers of each clone on a 3-week sub-

culturing interval. Each container must be labelled to indicate the medium and clone it contains. This is equivalent to over 138 000 records per year.

In this paper a computer system is described for the maintenance of subculturing records and information retrieval on the clones.

MANUAL SYSTEM OF RECORD KEEPING

The computer system arose out of an existing manual operation. A set of sequentially numbered container lids served as the basis of the manual system and the operations associated with each container were recorded, e.g., containers numbered 50 to 365 contain medium SM2 (a medium code) and containers 500 to 600 contain medium RM1 (a medium code).

When a clone was subcultured from one medium to another a record was made on the file for that clone of each subculturing operation, e.g., clone CML-52 (CML is the code for *Eucalyptus camaldulensis* Dehnh., clone number 52) was subcultured from lid range 50–55 (medium SM2) to lid range 506–525 (medium RM1) (see Fig. 1). In this way a complete history of subculturing operations was maintained for each clone (including dates, media it has been tested on, etc.).

THE COMPUTER SYSTEM

The computer system consists of two distinct parts – a databank containing all essential information stored in several files, and a set of programs allowing users to access and manipulate this information. The relationships between the programs and files and the information flow between them are outlined schematically in Fig. 2.

The system described here has been implemented on a D.E.C. PDP11/34 mini-computer using the RSX-11M operating system and RMS file support software. The system is also operating on VAX computers under VMS operating systems and a version is running on a PRIME computer. With modifications the system could run on smaller computers. The main problem in converting the system to different computers is whether they have the capability of handling indexed sequential files from FORTRAN.

The Databank

The databank consists of five files stored on magnetic disk. In order to minimise information retrieval time, an indexed sequential organisation (Hawryszkiewicz 1977) is used for all files. Generally, indexed sequential files are stored in a manner such that they can be accessed directly by the value of selected record items (fields) called key-fields. The key-fields are designated at the time of creation of files and their selection reflects the users' demand for retrieval of information. For example, in the clone file there are several clones which belong to the same species. In order to extract information about all clones of the same species an indexed sequential search using the species name as a key can be conducted. Note that the species name field, which is present on every clone record, has been declared as a key at the time of creating the clone file (see 'clone' file description). An indexed sequential search using species name as a key will only directly access records in the databank having the matching key in the appropriate field.

CLONE NUMBER	PRESENT RANGE OF LID NUMBERS	TO BE PLACED ON	NEW RANGE OF LID NUMBERS	DATE	ADDITIONAL NOTES ON SUBCULTURING	NEW LOCATION (STORE)	DATA BASE DETAILS ONLY
CML-52	50 - 55	RM1	506-525	1/4/86	RM1 : 20 containers	Culture Room	3.5.7
CML-42	10861-10875	S2A	99-108	1/4/86	10 vials on S2A to cool storage	Cool Room	9.1.6
			109-118	"	10 to shelf 5	Culture room	5.2.67
		R10	606-615	"	10 containers on R10	Greenhouse	8.1.0
COMMENTS							

FIG. 1—Example of subculturing notes. The “new range of lid numbers” and “date” are completed by the operators immediately after subculturing. The “database details only” refers to the new location of the containers and is recorded when the containers are placed in their new environment.

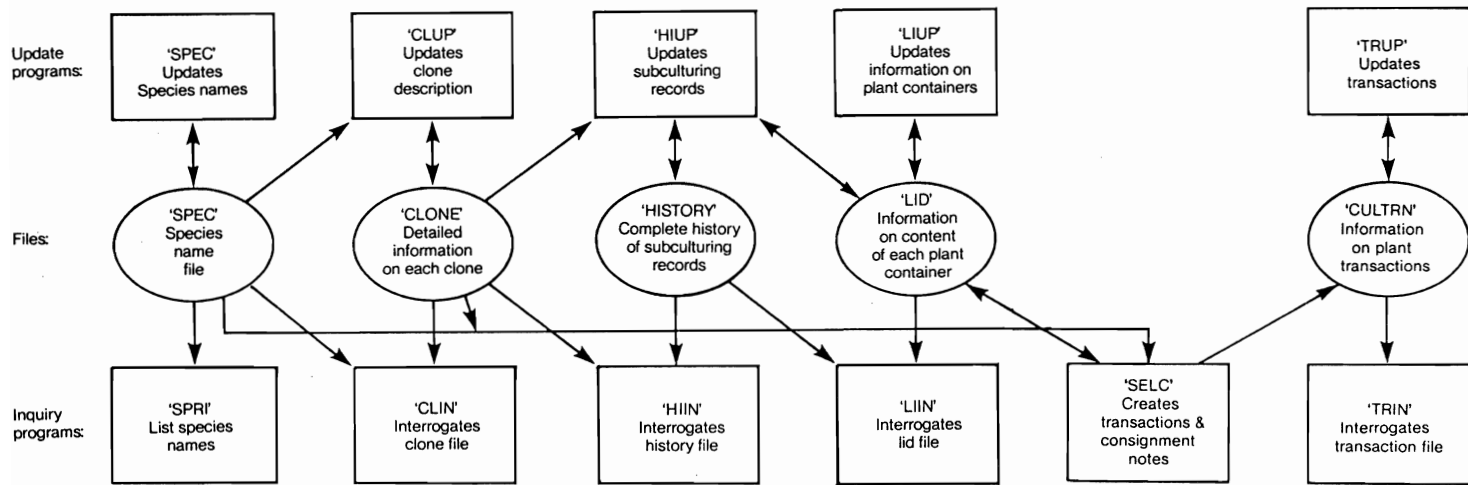


FIG. 2—Information flow diagram between programs and files.

This type of search/retrieval procedure is extremely fast as it reduces disk input/output to a minimum and it is essential for efficient machine operation with large data sets (Dunn 1979). Fields common to several files are used in cross-interrogation of these files.

Description of files

1. Species name; 'SPEC' File. The 'spec' file contains the botanical names of all species. Records within the file have two fields, a unique three-character species code and the corresponding species name, e.g., CML = *Eucalyptus camaldulensis*. The species code is the same as that used in several other CSIRO databanks (Chippendale & Wolf 1984; Wolf & Turnbull 1982). This will enable a common key between databanks in the future – for example, a databank is being developed for international field trials of tree species.

The full species name can be obtained by a single access on this file using the species code as a key. The purpose of this file is to reduce the amount of repetitive information in the databank.

2. Information on each clone; 'CLONE' File. This file contains a detailed description of all of the clones. Each clone has a record consisting of the following fields:

2.1 Species name, represented by the three-character species code (as above).

2.2 Clone number, a sequential number from 1 to n unique to each clone.

2.3 Organisation, a code to represent the organisation or individual that selected the clone, e.g., UM = University of Melbourne, WF = Forests Department, Western Australia.

2.4 Seedlot number. This number is included if the clone was derived from a seedlot number of a particular organisation. It enables access to additional information which may be available from the seedlot number, e.g., details of soil type at seed collection site.

2.5 Notes. A code describing special characteristics of each clone, e.g., SL = screened for salt tolerance under laboratory conditions.

2.6 Ortets. The number of mother-plants of each clone available as a seedling, as cuttings, as disease-indexed plants in tissue culture, or as cultures in cool storage.

2.7 Latitude, longitude, and altitude. Refers to the original site of the seedlot, for clones derived from seedlings, or the original location of the ortet.

Species name, organisation, seedlot number, notes, latitude, longitude, and altitude have been designated as key fields for use in indexed sequential searches. This reflects the high demand for searches on these particular attributes.

3. Description of each subculturing process for each clone; 'HISTORY' File. Each subculturing process is represented by a unique record consisting of the following fields:

3.1 Process number, a sequential number from 1 to n, unique to each subculturing event. This number is automatically generated by the computer program at the time of adding a record to the history file.

- 3.2 Species name (described in 2.1, CLONE file).
- 3.3 Clone number (described in 2.2, CLONE file).
- 3.4 Origin range. The number range of the containers from which the clones originated for that particular subculturing process. If the clone was being established in culture from the ortet the origin range is zero.
- 3.5 Destination range. The number range of the containers to which the clone is being subcultured.
- 3.6 Medium. A code for the medium in the 'destination range' of containers.
- 3.7 Date. The date of the subculturing process.
- 3.8 Date Due. The date the clone is next due to be subcultured.
- 3.9 Location. The position of the numbered containers in the propagation room, cool room, etc. The location is divided into three parts – the bank of shelves, the shelf number, and the position on each shelf.

The process number, species name, clone number, date, date due, and location were designated as keys to cater for information-retrieval demands. The species name and clone number provide a cross-referencing link between the 'history' and 'clone' files.

4. Information on each container; 'LID' File. The 'lid' file contains a detailed description of each container and its contents. Each container is represented by a unique record consisting of the following fields:

- 4.1 Lid number. A sequential number from 1 to n unique to each container. In our laboratory the lid numbers are printed on computer labels using the program 'LBLS' (see utility programs below). It is also possible to use permanently labelled containers or containers labelled with a computer-readable bar code.
- 4.2 Medium. A code for the medium in the container. Empty containers have zero as a medium code.
- 4.3 Date. The date the medium was placed in the container.
- 4.4 Process number (described in 3.1, HISTORY file). This number is automatically generated by the computer at the time of subculturing. Containers which have a medium in them but no plants have a process number of zero.
- 4.5 Species name (described in 2.1, CLONE file).
- 4.6 Clone number (described in 2.2, CLONE file).

Lid number, medium, species name, and clone number, have been designated as key-fields. The lid number, process number, and medium, are used to cross-reference the 'lid' and 'history' files.

5. Details of each commercial transaction; 'CULTRN' File. The 'cultrn' file contains a detailed description of commercial transactions to customers. Each transaction is represented by a unique record consisting of the following fields.

- 5.1 Transaction number. A sequential number from 1 to n automatically generated by the computer to uniquely identify each transaction.
- 5.2 Customer. A brief identification of a customer.

5.3 Country. Country of destination.

5.4 Clone (described in 2.1 and 2.2, CLONE file).

5.5 Date. The date the transaction took place.

5.6 Lid range. A range of lid numbers for the plant containers of a particular transaction.

5.7 Total price. The price per container (unit price) is entered into the computer interactively when recording a transaction. The total price is calculated from the number of containers and their unit price.

Transaction number, customer, country, date, and clone were designated as keys to speed up information retrieval on transactions.

Programs

All programs are written in Digital Fortran IV. Programs are well structured and suitable for modification and expansion.

The programs assist the user to perform routine daily tasks. They are specifically designed for users with little or no knowledge of computers. All programs are fully interactive, are "menu driven", and communicate with the user in English. Operator entries are monitored and, where possible, checked for errors. When an error is detected a message is displayed on the terminal indicating the nature of the error and a recovery procedure is initiated. For example, when subculturing, a check is made that the 'origin range' of containers contain the specified clone, and that the 'destination range' of containers are empty (process number = 0) and contain the medium specified. If these conditions are not met, an appropriate message is displayed at the terminal and the subculturing information is not entered into the history file.

A control program written in 'job-control' language links the individual programs together into one interactive package. This control program enables the user to interactively branch to individual programs and supervises the performance of the programs.

The user logs on to the computer and selects the database system. The system then responds with a menu of options (Table 1). Communication between the user and the database system is interactive for the choice of options in the main menu, and within all programs.

Functionally, the programs can be classified into three groups:

- (a) Update programs, which provide the user with the ability to add, alter, and delete information in various parts of the databank. The use of all update programs is restricted by passwords to authorised users only.
- (b) Inquiry programs, which select information from the databank according to user-specified criteria.
- (c) Utility programs, which perform special functions such as printing reports or preparing labels for the plant containers.

Update programs

1. Species name program; 'SPEC'. The program SPEC enables the user to add or replace species names in the species file. Note that the species name must be present

TABLE 1—List of options available when the control program is activated

SPEC	Enables entry of new species to species file
SPRI	Lists species on printer
*	
CLUP	Updates clone databank
CLIN	Information retrieval system for clone data
*	
LIUP	Updates lid databank
LIIN	Information retrieval system for lid data
*	
HIUP	Updates history databank
HIIN	Information retrieval system for history data
*	
BACK	Back-up of private databanks on RL01 (small) disk or mag tape
*	
LBSL	Produces self-adhesive labels for lids (works on DECWRITER)
*	
SELC	Stores transactions and prints consignment note
TRIN	Information retrieval system for transaction data
TRUP	Updates transaction databank
*	
STOP	Stops run

in the species file before a clone of that species can be recorded in the clone file.

2. History update program; 'HIUP'. This program enables the user to add, delete, or alter subculturing records in the history file. Records marked for deletion are first displayed on the terminal and then the user confirms or rejects the actual deletion. The user can interactively alter any field on any record in the file. All new records are interactively created by the user and extensive checks are carried out before the actual addition takes place. For example, the program checks the containers from which the plants originate, whether the plants in them belong to the specified clone, that the 'destination' containers are empty (process number = 0), and that they contain the specified medium. If these conditions are met then the program generates a new process number and stores the new record into the history file. The same process number is then recorded in the lid file to indicate that the container has plants in it.

If requested, the program clears the containers from which the plants were subcultured by setting their process number, medium, and date to zero in the lid file.

3. Clone update program; 'CLUP'. This program enables the user to add, delete, and alter clone description records in the clone file. The records selected for deletion are first displayed on the terminal and, if confirmed by the user, deleted from the database. Any information on any record in the file can be altered. New records are created and checked for errors before being recorded in the databank.

4. Container update program; 'LIUP'. This program enables the user to add, delete, and alter records for the plant containers in the lid file. The program works on the range of specified lid numbers rather than on individual records. The range must be contiguous. The program can clear (set process number to zero) those containers from which the plants originated and add a process number to occupied containers.

5. Transactions update program; 'TRUP'. This program enables the user to delete or alter transaction records. New records are created and added to the transaction file only by program SELC (see utility program 3).

Inquiry programs

1. Information retrieval on subculturing records; 'HIIN' program. This program enables the user to search the database for all subculturing records which satisfy specified criteria. The criteria can be expressed in terms of clone number, location in the propagation room, a range of creation or due dates, and a range of process numbers. Criteria can be selected in any order and in any combination. This allows the user to formulate complex retrieval criteria. For example, the program can be asked to find all the plants due for subculturing between 1 June 1985 and 24 September 1985 which belong to *E. camaldulensis* clone number 87. The output from the search can be presented as a summary on a visual display terminal, as complete information on the central printer, or both.

2. Information retrieval on clones; 'CLIN' program. The search criteria for CLIN are species name, organisation, seedlot number, notes, and a range of latitudes, longitudes, and altitudes (see the description under CLONE file for details). These criteria can also be selected in any order and combination. This program is very useful for clone selection as it can identify clones suitable for planting in certain environments. For example, the program can find all the clones of *E. camaldulensis* (species code = CML) which are salt-tolerant (notes = SL) and originated from a natural population located above an altitude of 400 m. The output from the search can be presented in two forms as for HIIN.

3. Information retrieval on containers; 'LIIN' program. The search criteria available are the medium, the range of container lid numbers, and the clone. The user can also specify whether the search is to retrieve those containers which do not contain plants (process number = 0) or containers with plants (process number > 0). Searching the lid file using the clone option enables records of all containers holding a particular clone to be retrieved. This is very useful at the time of subculturing and for making comparisons between responses on several media.

The process number is used to access the corresponding subculturing record in the HISTORY file. The information from the LID and HISTORY files is then combined into a single report for output to the terminal or printer. For each container the report shows the clone it contains (if any), medium, date the medium was prepared, process number, date of subculturing, date next subculturing is due, and location in the tissue culture room.

This report can also be presented on a visual display terminal, a printer, or both.

4. Information retrieval on commercial transactions; 'TRIN' program. This program enables the user to search transaction file for records satisfying selected criteria. The available criteria are the transaction number, customer, country, clone, and date. As in all inquiry programs these criteria can be selected in any order and combination. The outcome of the search is again presented in two forms as for HIIN.

Utility programs

1. Listing of species names and codes; 'SPRI' program. This program provides the user with all the species codes and their botanical names which are stored in the SPEC file.
2. Printing of numbers for containers; 'LBLS' program. This program produces a set of numbers on self-adhesive labels for the containers.
3. Printing of consignment notes for commercial transactions; 'SELC' program. This program enables the user to select plants for sale to a specific customer and produces a standard consignment note for despatch with the plants. A brief clone report is also produced and attached to the consignment note. Details of the transaction are automatically recorded in the transaction file.

Several other utility programs are in preparation. One will provide a typewritten description of each clone covering details which are summarised in the CLONE file, including which organisation selected the clone, what were the methods and results of the selection, where field trials have been established and their results, organisations and addresses for correspondence. This information can be updated as results become available and will accompany each delivery of clones.

DISCUSSION

The computer system described here has reduced the time and labour involved in maintaining subculturing records and retrieving information, and has detected a number of errors which would have been made using the manual system.

The main reason for the reduction in labour is that the data pertinent to each subculturing operation (Fig. 1) has to be entered only once into the computer. The database system then carries out a number of checks for errors, notifies the user if errors have occurred, and enters the subculturing data into the History and Lid files. It is then possible to interrogate and retrieve information from the files in many ways without the need to maintain several lists as was necessary with our manual system. For example, the files can be interrogated using the inquiry programs to obtain information on all containers that are due for subculturing on a particular date, or for giving a complete history of subculturing records for a particular clone. More complex information retrieval is also possible. To obtain such information from a manual system would require the maintenance of several lists.

Errors detected include placing plants on to the wrong medium and misidentification of clones. Such errors are more likely when a large number of clones are being handled but could have serious consequences in a situation where a laboratory is producing certified lines or varieties of a particular plant.

The descriptions in the clone file reflect our present needs, but the records could be easily modified to meet the needs of other users. For example, with ornamental plants descriptions such as flowering time, size, colour, or application of the plant for specific purposes might be more appropriate. Similarly, minor additions to the system would make it possible to forecast the number of containers required in order to meet future orders.

Linking with other databank systems can extend the amount of information available. For example, the above system could be linked into our Seeds Centre databank to obtain additional details about the clone such as the soil and meteorological conditions of the collection site. It will also be possible to link into another databank which is co-ordinating the results of field trials around the world.

Computer systems are of great benefit in the management of gene banks and seed stores (Wolf & Turnbull 1982). The conservation and the international exchange of plants in tissue culture is becoming increasingly important (International Board of Plant Genetic Resources 1982; Kahn 1978) and for such purposes the above computer system could be very useful.

Further information and listings of the above computer system may be obtained by writing to the authors.

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