# THUNBERGIA

4

O. CONSTANTINESCU & R. MOBERG

**Computer assistance and some time-saving routines in UPSC culture collection**

DISTRIBUTED BY THE HERBARIUM, UNIVERSITY OF UPPSALA

THUNBERGIA is the continuation of "Publications from the Herbarium, University of Uppsala", of which 19 numbers numbers were issued between 1978 and 1986.

The name is given in memory of Carl Peter Thunberg (1743-1828), a disciple of Linnaeus and a famous traveller to South Africa and Japan. His collections, about twentyseven thousand sheets, were transferred to our University in 1785, and became the foundation of the botanical museum. Thunberg succeded Linnaeus filius as professor in 1785 and held the professorship to his death.

Editor: Roland Moberg

Address: The Herbarium
Uppsala University
P.O. Box. 541
S-751 21 Uppsala, Sweden



# Computer assistance and some time-saving routines in UPSC culture collection

Ovidiu Constantinescu and Roland Moberg

The Herbarium, Uppsala University
P.O.Box 541, S-751 21 Uppsala, Sweden

Abstract. The use of a computer program for producing lists, catalogue and other outputs, and for automation of the transferring schedule and printing of culture lables is presented. Both a procedure for long-term preservation of prepared culture media and a time-saving transferring routine are described.

## Introduction

During the organization of the Uppsala University culture collection of fungi (UPSC) and of the identification service all routine work had to be severily rationalized, because of the various multiple functions of the unit and a rather restricted budget. Our experience in using the computer facilities in this particular field, and in the organization of some culture collection specific activities, may be of interest to those facing similar problems.

Taking into account the aims of the collection, the budget, and the staff (one full time research assistant and one half time technical assistant) it was considered that the frequent transfer preservation method (Smith & Onions 1983) best suited our present situation. However, the acquisition of liquid nitrogen storage equipment is under consideration.

Some 1400 strains are preserved as slant cultures on media containing 2% agar. Glass, rimless, 160 x 16 mm test tubes containing 7 ml medium and stopped with aluminium caps are used. The cultures are arranged according to their accession number in propylene racks, 50 tubes in each rack, and stored in a 330 L domestic refrigerator which can hold c. 3,000 cultures. The preservation temperature is 5-7°C. The period between two transfers varies between 12-26 months. According to the present experience, most of the cultures withstand more than 18 months in these conditions. Two different culture media are used for each strain, but only one tube is preserved at a time. The media are interchanged each time the strain is transferred.

## Computer assistance

A number of methods for keeping herbarium records and producing labels were developed during the period when punchcards were employed for information storage and retrieval (Beschel & Soper 1970, Argus & Sheard 1972, Morris 1974). More efficient, terminal-based systems have later been described (Vitt et al. 1977, Jones et al. 1983, Farr 1986). Recently, microcomputer-based information storage and retrieval methods (Bryant 1983, Fletcher 1985, 1987) and a labelling system (Smith & Savege 1984) for culture collection have been proposed. These systems are meant to store data, to produce lists of preserved organisms and/or labels. A much more elaborated system, intended as a standard for culture collections, particularly for those joining the Microbial Information Network Europe, is now under consideration (Gams et al. 1987).

Our intention in using the computer was not only to facilitate the maintenance of the strain records and to produce lists or a catalogue, but also to automate the transferring schedule and the printing of updated labels, an unavoidable and time-consuming process in a culture collection maintained by periodical transfer. In the same time, we had to adapt our system to the existing computer equipment of the Faculty of Mathematics and Natural Sciences of the Uppsala University.

The following hardware and software was available: VAX 11-780 computer, Facit Twist VT100 terminal and/or Ericsson PC, Facit 4510 serial matrix printer and/or Facit 8105 daisy wheel electronic typewriter, MIMER/IR, MS WORD, and Ericsson Asynchronous Communication programs. MIMER is an information storage and retrieval program written in standard FORTRAN, and it is a product of MIMER Information Systems AB, Uppsala.

### Data format

When we started our system at the beginning of 1986 no internationally accepted format for data entry intended for culture collections was available; indeed this format is still in a project phase (Gams et al. 1987). Consequently, we have adopted a strain record rather similar to the specimen record used for the computerization of the UPS herbarium. The data entry is structured in 29 fields (Fig. 1). For explanatory reasons, both the name of the field and its label/acronym are shown in Fig. 1, although only the latter is normally displayed on the computer screen. ON refers to subcultures of the same strain kept in other collections, OM to the alternate state (anamorph or teleomorph), IS to the way the strain was isolated (single spore, single ascus, tissue etc.) and to the code of its mating type (+, -), DR indicates the herbarium where a dried reference culture is preserved, and HB is meant to indicate if and where



the herbarium voucher specimen from which the culture was obtained is deposited. MP shows the medium on which the strain is presently kept and OP if the strain is also preserved by another method than periodical transfer. The transfer period (TP) is always indicated in months. MIMER gives automatically to any new document a sequential number. A document can also be provided with another identification number. For reasons of convenience the sequential number also functions as the accession number.

| | |
|---|---|
| Docno: 2121 | |
| Other no (ON) | - |
| Species (SP) | Nectria purtonii |
| Author (AU) | (Grev.) Berk. |
| Other morph (OM) | Fusarium aquaeductuum (Radlkofer & Rabenh.) Lagerh. |
| Isol/sex (IS) | SS |
| Matrix (MA) | Eutypella stellulata |
| Country (CY) | Sweden |
| Province (PR) | Uppland |
| Locality (LO) | Dalby par., Påskhällen |
| Sender (SE) | - |
| Acc date (AD) | 1987.04 |
| Collector (CO) | K. & L. Holm |
| Coll no (CN) | 4404b |
| Coll date (CD) | 1987.03.10 |
| Isol by (IB) | O. Constantinescu |
| Isol no (IN) | P-674 |
| Isol date (ID) | 1987.03 |
| Det by (DB) | K. & L. Holm |
| Dried (DR) | UPS |
| Herb (HB) | UPS |
| Medium pres (MP) | MA |
| Medium next (MN) | OA |
| Transf last (TL) | 1987.04 |
| Transf per (TP) | 18 |
| Transf next (TN) | 1988.10 |
| Other pres (OP) | - |
| Metabol (ME) | - |
| Ref (RE) | Booth, Fusarium p. 62, 1971 |
| Notes (NO) | Ascomata on MA at 20°C and NUV light |

Fig. 1. A structured, completed strain data form. See text for explanation.

### Applications

All applications are started with a search command, followed by a predefined selection of the fields within a document and

their arrangement in a desired sequence. Further, the documents are either arranged sequentially by accession number or sorted alphabetically, and a printed output is obtained.

By using these application programs the following outputs can be obtained (Fig. 2): list of cultures arranged by accession number, alphabetically arranged list of cultures, catalogue of cultures, record file accompanying cultures delivered in exchange or for sale etc.

The lists of cultures used as a working, internal document, and the records for cultures delivered are not edited, whereas the catalogue is edited with Microsoft Word.

The most useful part of the computer assistance is the automation of the transferring schedule. Every month, the strains which have to be transferred at that particular time, i.e. those TN correspond with the current year and month, are retrieved and an application program called UPDAT is run. It contain two steps: first the present culture medium (MP) is interchanged with the next one (MN); second, the transfer dates are updated i.e. the next transfer date (TN) becomes last (TL) and to this updated last transfer date the transfer period (TP) is added in order to get an updated next transfer date. Finally, labels containing the accession number, fungus binomial, culture medium and date of transfer are printed on 50 mm wide gummed roll paper, using long life, non correctable ribbon (Fig. 2E).

**The transfer routine**

In the standard procedure the cultures to be transferred are removed from the collection and stored in separate racks. This operation is facilitated by the printing of the labels according to accession number sequence; the collection itself being arranged by accession number. Using the same printout, tubes with the necessary fresh media are prepared. The roll paper of the printout is cut into c. 25 mm wide labels which are glued on the appropriate culture media tubes, maintaining their accession number sequence. After transfer the fungi are routinely incubated at 20*C, in complete darkness for 7 days, followed by 7-10 days in a 12 h on/off alternating regime of darkness and a combined day- and near ultraviolet light. The old cultures are stored in the refrigerator. After incubation the newly transferred cultures are checked against the old ones and information concerning the survival, growth, sporulation etc., are entered into the strain data file under NO (notes) field. If a fungus does not grow or sporulate well on the new culture medium, a new inoculation from the original culture is made on a different medium.



**A**
2123 Acremonium butyri - Populus tremula wood
2124 Quaternaria dissepta - Ulmus glabra
2125 Pithya cupressina - Juniperus sp.
2126 Lophiotrema nucula - Salix fragilis

**B**
Discostroma corticola 1937 Cornus mas
Ditiola radicata 1524 Pinus sylvestris
Ditopella ditopa 1863 Alnus glutinosa
Doratomyces nanus 1982 marten dung

**E**
UPSC 1665
Apiospora
montagnei
MA 1987.07

UPSC 1719
Penicillium
roqueforti
CZM 1987.07

**C**
Nectria purtonii (Grev.) Berk.
anamorph: Fusarium aquaeductuum (Radlkofer & Rabenh.) Lagerh.
UPSC 2121, SS
Substrate: Eutypella stellulata
Place of origin: Sweden, Uppland, Dalby par., Påskhällen
Acc. date: 1987.04
Coll/No/Date: K. & L. Holm, 4404b, 1987.03.10
Isol. by/No/Date: O. Constantinescu, P-674, 1987.03
Ident. by: K. & L. Holm
Voucher herb. specimen: UPS

**D**
**Mycogone rosea** Link
1588 Entoloma prunuloides, Sweden, 1985.08
1600 Tricholoma terreum, Sweden, 1986.08
1906 Inocybe friesii, Sweden, 1986.08
**Myrothecium roridum** Tode: Fr.
2007 plate contaminant, Sweden, 1986.12
**Myxotrichum berkeleyi** Apinis
968 Pinus plank, Sweden, 1983.10
**Nectria purtonii** (Grev.) Berk., anamorph: Fusarium aquaeductuum (Radlkofer & Rabenh.) Lagerheim
2121 SS, Eutypella stellulata, Sweden, 1987.04

Fig. 2. Various computer printout. A - List of cultures arranged by accession number; B - List of cultures arranged alphabetically; C - Form accompanying the strains delivered; D - A section of the catalogue (in preparation); E - Updated labels used for the newly transferred cultures.

## Preservation of culture media

Because fungi received for identification belong to almost all taxonomic groups, a large variety of media have to be ready for use at any time. This is particularly important for less common media which are only needed rarely, and consequently have to be preserved for long periods of time. The following procedure is a modification of the method described by Watson et al. (1966): after medium preparation 12 ml aliquots are distributed with a manual dispenser in 180 x 18 mm test tubes, stopped with aluminium caps and autoclaved. After cooling the tubes are introduced into a double polypropylene bag, and a cotton wad moistened with c. one ml propylene oxide is added. The bag is sealed with a rubber band, stored for minimum 48 h at room temperature in a fumigation cabinet, open for c. 30 min. in order to eliminate the propylene oxide in excess, sealed again and stored in refrigerator. When needed the tubes are extracted from the bags, heated on a water bath to melt the medium and poured into the Petri dishes. By using this procedure c. 40 different culture media are permanently in stock, stored in two 330 L refrigerators. No contamination has occurred even after more than three years storage. Tubes processed in the same way but not treated with propylene oxide become contaminated after a few months. Moulds grow first on the exterior of the tube, and later within the tube. The media used for the preservation of cultures are run on in the same way.

## Conclusions

Compared with other computer assistance procedures used in culture collections maintained by frequent transfer, the one presented here has the advantage that it supplements the usual storage and retrieval systems with an automated input of variable information (i.e. the transfer period), and completely eliminates the manual writing of the labels, a permanent source of error. Another advantage is that the transferring operations and the preparation of media can be planned in advance. The system used at Uppsala is applied to a small collection; it would provide much greater advantages if used with larger collection. It can also be of use to other types of collections where periodical renewal, updating or inspection is needed.

By using the computer, and following the transfer routine described, the time necessary for processing a culture, from the retrieval of the old culture till the placement of the newly transferred one into collection, could be reduced to less than 3 minutes (Tab. 1).



The storage of a large assortment of media for longer periods saves both time and money. This is particularly important for those laboratories where the diagnosis of a wide variety of microorganisms with different culture requirements is an almost daily routine.

Table 1

Estimation of the time spent for processing one culture 1)

| | |
|---|---|
| Retrieve, update, type label | 9" |
| Glue labels on tubes | 14" |
| Remove culture from collection | 14" |
| Transfer | 50" |
| Check old and new culture, enter data | 70" |
| Place the new culture into collection | 15" |
| Total | 2'52" |

1) Not including the time spent to prepare the tubes with fresh medium, and the incubation period.

## Acknowledgements

We thank Mr Torbjörn Wiktorin for writing the UPDAT application program, Miss Wiltrud Neumann for help with the entering of the data, and Dr. John Packham for correcting the English text. This work was supported by grants from the Swedish Natural Science Research Council, Swedish Council for Forestry and Agricultural Research, and Knut and Alice Wallenberg Foundation

## References

Argus, G. W. & Sheard, J. W. 1972. Two simple labeling and data retrieval systems for herbaria. - Can. J. Bot. 50: 2197-2209.

Beschel, R. E. & Soper, J. H. 1970. The automation and standardization of certain herbarium procedures. - Can. J. Bot. 48: 547-554.

Bryant, T. N. 1983. A microcomputer-based information storage and retrieval system for the maintenance of records for culture collection. - J. Appl. Bact. 54: 101-107.

Farr, D. F. 1986. Experience in computerizing the National Fungus Collections' specimen label data. - Mycotaxon 25: 175-182.

Fletcher, H. J. 1985. Data base manipulation in Mycology. - Bull. Br. Mycol. Soc. 19: 140-143.

Fletcher, H. J. 1987. A microcomputer catalogue of cultures. - Bull. Br. Mycol. Soc. (The Mycologist) 21: 117-118.

Gams, W., Hennebert, G. L., Stalpers, J. A., Janssens, D., Schipper, M.A.A., Smith, J. & Yarrow, D. 1987. Structuring strain data for storage and retrieval of information on fungi and yeasts in MINE, the Microbial Information Network Europe. - MINE Technical Publications No. 1 (manuscript).

Jones, S. B., Coile, N. C. & Martin, R. 1983. The University of Georgia Herbarium (GA) optical-scan data encoding system. - Taxon 32: 47-50.

Morris, J. W. 1974. Progress in the computerization of herbarium procedures. - Bothalia 11: 349-353.

Smith, D. & Onions, A. H. 1983. The Preservation and Maintenance of Living Fungi. - Commonwealth Mycol. Institute, Kew.

Smith, P. & Savege, A. 1984. Labelling of stock cultures using a microcomputer. - Trans. Br. Mycol. Soc. 82: 749-751.

Vitt, D. H., Horton, D. G. & Johnston, P. 1977. A computer program for printing herbarium labels. - Taxon 26: 425-428.

Watson, R. D., Carley, H. E. & Huber, D. M. 1966. Storage of culture media for laboratory and field use. - Phytopathology 56: 352.

**Publications from the Herbarium, University of Uppsala.**

1. L. Tibell, Caliciales exsiccatae, fasc. I (Nos 1-25), 1978
2. K. Vanky, Ustilaginales, fasc. I-V (Nos 1-125), 1979
3. K. Vanky, Ustilaginales, fasc. VI-X (Nos 126-250), 1979
4. L. Tibell, Caliciales exsiccatae, fasc. II (Nos 26-50), 1979
5. K. Vanky, Ustilaginales, fasc. XI-XIII (Nos 251-325), 1980
6. Fungi Exsiccati Suecici, fasc. LXI (Nos 3001-3050), 1981
7. Fungi Exsiccati Suecici, fasc. LXII (Nos 3051-3100), 1981
8. N. Lundqvist, Fungi Fimicoli Exs., fasc. I-II (Nos 1-50), 1981
9. K. Vanky, Ustilaginales, fasc. XIV-XV (Nos 326-375), 1982
10. L. Tibell, Caliciales exsiccatae, fasc. III (Nos 51-75), 1982
11. K. Vanky, Ustilaginales fasc. XVI-XVIII (Nos 376-450), 1983
12. L. Tibell, Caliciales exsiccatae, fasc. IV (Nos 76-100), 1984
13. R. Santesson, Fungi Lichenicoli Exs., fasc. I-II (Nos1-50), 1984
14. Fungi Exsiccati Suecici, fasc. LXIII (Nos 3101-3150), 1984
15. Fungi Exsiccati Suecici, fasc. LXIV (Nos 3151-3200), 1984
16. L. Tibell, Caliciales exsiccatae, fasc. V (Nos 101-125), 1985
17. Fungi Exsiccati Suecici, fasc. LXV (Nos 3201-3250), 1985
18. Fungi Exsiccati Suecici, fasc. LXVI (Nos 3251-3300), 1985
19. R. Moberg, Lichen exsiccata and other collections in UPS, 1986

**THUNBERGIA**

1. L. Tibell, Caliciales exsiccatae, fasc. 6 (Nos 126-150), 1986
2. R. Santesson, Fungi Lichenicoli Exs., fasc. 3-4 (Nos51-100), 1986
3. R. Moberg, Lichenes Sel. Exs. Upsal., fasc. 1 (Nos 1-25), 1986

Published Dec. 1. 1987

ISSN-0283-2275

*Reprocentralen HSC*, 1987