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BIODETERIORATION CENTRE
UNIVERSITY OF ASTON
ST. PETER'S COLLEGE,
SALTLEY, BIRMINGHAM B8 3TE.

Editor-in-Chief of Biodeterioration Centre Journals
Dr. H.O.W. Egginns.
Editor
Professor T.A. Oxley.
Business Manager
Dr. D. Allsopp

The Editors are able to call upon the assistance of an Editorial Board whose members are in Britain, various countries of Europe, and the U.S.A.

NOTES FOR CONTRIBUTORS

The International Biodeterioration Bulletin is published four times per year (Spring, Summer, Autumn and Winter). Typescript contributions should be sent to the Editor, Professor T.A. Oxley, at the above address. The Bulletin acts as a vehicle for the publication of original works, including reviews, on all aspects of biodeterioration, i.e., deterioration of materials, artefacts or facilities, of economic importance by living organisms, which include microorganisms, insects, rodents, birds, higher plants, etc. Articles on biodegradation, that is conversion of materials to less objectionable, more disposable, or higher value products by living organisms, are also published.

Contributions are published only in English. Each article must be accompanied by a summary in 50–150 words which will be translated into French, German and Spanish. Native speakers of these languages are invited to submit their summaries in their own language; in certain circumstances complete articles may be submitted in French, German or Spanish and will be translated into English for publication.

Illustrations must be very clearly drawn, normally larger than the size finally desired. The suggested final size should be clearly indicated but the Editor reserves the right to vary this in the interests of economy and clarity.

As far as possible diagrams will be reduced to single column width (80 mm) or to half page (170 mm). In any event, neither these nor half tone photographs can exceed full page (260 by 170 mm). Authors should bear in mind that it is generally more convenient for readers if legends which accompany diagrams or photographs appear with them on the same page and should proportion their illustrations accordingly. Lettering on diagrams will normally be inserted by the printer; authors are therefore asked to insert lettering or symbols in pencil on the originals or in ink on a copy.

All articles are submitted by the Editor to one or more independent referees for advice on their clarity, originality, and general suitability for publication, but the final decision whether or not to publish an article rests with the editors. If articles are rejected the substance of the referee's report will usually be communicated to the author and in suitable cases the Editor will be pleased to help authors to improve their papers with a view to possible publication.

Bibliographic references are indicated in the text by author names (no initials) and year only, viz: Reese and Levinson (1952); or: Darby et al., (1968) and in the bibliography in strict alphabetical order of first author's names, thus: Reese E.T. and Levinson H.G. (1952)

Comparative study of the breakdown of cellulose by microorganisms.

Physiologica Plantarum 5: 354–366

or:

Darby R.T., Simmons E.G. and Wiley B.J. (1968)

A survey of fungi in a military aircraft fuel supply system.


References to books, conference proceedings, etc. should quote first the author(s) or editor(s), then the year of publication and title followed by the name of the publisher and the city in which it is published. As far as possible titles of journals should be given in full except for such abbreviations as 'Journ.', 'Proc.', 'Trans.' etc.

20 reprints will be sent free of charge to the first named author unless otherwise instructed. Any number (normally not more than 150) of additional reprints may be purchased if ordered sufficiently in advance. An order form and price will be sent giving about one month's notice.

ACKNOWLEDGEMENTS TO SUSTAINING ORGANISATIONS

Financial support for the Biodeterioration Centre from the following organisations is gratefully acknowledged:

- ALBRIGHT & WILSON (MFG) LTD., Oldbury Division, P.O. Box 3, Oldbury, Warrior, Worcestershire, England.
- B.D.H. CHEMICALS LIMITED, Laboratory Chemicals Division, Poole, Dorset, England; manufacturers of laboratory chemicals, biochemicals, industrial fine chemicals and microbiocides.
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- NATIONAL COAL BOARD, Coal House Lyon Road, Harrow, Middlesex, England.
BIODETERIORATION SOCIETY NEWSLETTER

U.K. Programme of meetings, 1982-1983

The Council of the Society has approved the following outline programme of meetings for the next two years.

Spring 1982 15-16 April.
Long Ashton Research Station, University of Bristol. Subject: Modern methods for detecting microbial spoilage.

Summer 1982 8 - 9 July.
Portsmouth Polytechnic. Subject: Conservation of Timber in Water. (To include a talk on the conservation of the Mary Rose).

Winter 1982 21 December.
Sheffield Polytechnic. Subject: Biodegradation of waste materials.

Spring 1982 (Date not yet fixed)
University of Nottingham. Subject: Recognition and physiology of spoilage organisms.

Summer 1983 (Date not yet fixed)
University of East Anglia. Subject: Post harvest deterioration of crops.

Winter 1983 (Date not yet fixed) (Venue not fixed)
Subject: Packaging as a means for control of biodeterioration.

Subscriptions

Members are reminded that subscriptions are due for payment on 1st April each year. The standard worldwide subscription is £2 per annum. To this is added £1.50 in U.K., as the meetings subscription, making a total for U.K. of £3.50 per annum. Other countries levy their own local meetings subscriptions. Worldwide subscriptions, together with U.K. local meetings subscriptions should be sent to the treasurer,

Dr. L.H.G. Morton,
Division of Biology,
Preston Polytechnic,
Corporation Street,
PRESTON, PR1 2TQ U.K.

Meetings Secretary for Nigeria

Dr. C.I. Ogbonna has agreed to act as local meetings secretary for Nigeria. Members interested should write to him. His address is:

Dr. C.I. Ogbonna,
Department of Botany,
University of Jos,
Private Mail Bag 2084
JOS Nigeria.

Contact with Members

The Council of the Society has considered the question of contact with members. It was felt that the present arrangement whereby U.K. members receive copies of this Newsletter by post, while overseas members receive nothing, is inequitable. It has therefore been decided that in future, after circulation of the Spring issue, this Newsletter will no longer be distributed. In its place an annual summary of events, including abstracts of papers presented at U.K. meetings, will be circulated to all members, world-wide. The Newsletter will continue to be published in the International Biodeterioration Bulletin as at present.
Physical Methods for Microbial Control

Title: Physical limits to microbial growth
Author: J.W. Hopton
Address: Department of Microbiology, University of Birmingham, P.O. Box 363, BIRMINGHAM, B15 2TT.

ABSTRACT

An array of methods is available to control microorganisms by physical means and choice is dependent on the nature and prospective use of the material and the character of the flora.

The physical conditions of some natural environments can resemble those imposed to control microorganisms. Such extreme environments are characterised by a markedly less diversity of organisms than normal environments are characterised by a markedly less diversity of organisms than normal environments and, in some, bacteria are the only inhabitants. Studies of the physiology of tolerant organisms indicate that tolerance is attributable to exclusion of the external stress, achieved through novel wall and membrane structure or to possession of tolerant macromolecules only marginally different in composition from sensitive counterparts.

Title: Ultrasonic disruption of microbial cells
Authors: W.T. Coakley and E.C. Hill
Address: Microbiology Department, University College, Newport Road, CARDIFF, CF2 1TA.

ABSTRACT

Cell disruption by ultrasound required the generation of ultrasonic cavitation bubbles in the cell suspending medium. the sound intensity threshold for cavitation generation increases with ultrasonic frequency. Consequently ultrasonic generation of 20kHz - 40kHz are favoured for cell disruption. The strong mechanical forces (shock waves and turbulence) associated with collapsing cavitation bubbles are responsible for the mechanical disruption of cell walls. The number of intact cells in a sonicated cell suspension decreases exponentially with time. The rate of decrease of the cell population depends on the mechanical strength of the cell wall. The disintegration constant is essentially independent of cell concentration up to 60 gm net weight of cells/100 ml of cell suspension. The disintegration constant for a 100 ml volume of brewers yeast exposed to 150 acoustic watts of 20kHz is about 0.0025 S^(-1), requiring 280S to disrupt half the cell population. Half the yeast population would be disrupted in a suspension flowing at 15 ml/min through a 150 watt field. Sonication is normally carried out to study enzymes and hence the cell suspension is externally cooled. If a commercial application to kill cells is envisaged it would be more appropriate to allow the temperature to rise. Ultrasound increases the permeability of cells and a synergistic effect with biocides can be demonstrated.

Title: Sterilisation with ultraviolet
Author: J.D. Bridgen
Address: Hanovia Group Ltd., 480, Bath Road, Slough, Berks. SL1 6BL.

ABSTRACT

The process of Ultra Violet sterilisation was presented as a means of disinfection of air and suitable liquids, and the surfaces of solids, primarily food products. The relationship of the various Ultra Violet energy wavelengths, produced by low and medium pressure mercury vapour arc tubes, to the cell protein absorption spectrum was demonstrated.

Examples of process applications were presented highlighting the important design parameters that must be considered to achieve a satisfactory operational efficiency. Equipment design, installation and maintenance applicable to the pharmaceutical, food and air-conditioning industries were discussed.
Title: Ethylene oxide sterilisation

Author: G. Drysdale

Address: Griffith Laboratories (U.K.) Ltd., Cotter Park Farm Industrial Estate, Somercoates, Alfreton, Derbyshire.

ABSTRACT

The use of a plate heat exchanger (PHE) means that a continuous pasteurisation system can be chosen. In this case oil is heated first by heat exchange with hot pasteurised oil, then to final temperature with steam and held at that temperature for the specified time. Cooling then occurs in the regeneration section and final cooling is by cooling water. Up to 95% heat recovery has been proved to be economical in the dairy/brewery industries.

Title: Sterilization by gamma irradiation

Author: F.J. Ley

Address: Irradiated Products Ltd., Moray Road, Elgin Industrial Estate, Swindon, Wiltshire, SN2 6DU.

ABSTRACT

Ionizing radiation has a lethal effect on microbial populations and the relationship between dose applied and numbers surviving is well established. There is a difference in radiation resistance between species and resistance is significantly influenced by a number of environmental factors. Radiation sterilization of medical devices and pharmaceutical products is practised on an industrial scale and there are many other applications of radiation processing involving microbial control. Radiation facilities are largely based on the use of gamma radiation using cobalt-60 which involves a high capital investment in the source itself and the necessary shielding and conveyor system.

Title: Pasteurisation of mineral oils

Author: A.F. Hurst

Address: Alfa Laval Co. Ltd., Great West Road, Brentford, Middlesex, TW8 9BT.

ABSTRACT

One method of controlling microbial growth in mineral oils is to heat the media to a temperature high enough to cause death; temperature and time being the controlling factors.

The simplest method is to heat a tank of media to the required temperature and maintained for the specified time. This method is very expensive in use of steam and cooling water and also takes a long time to produce a suitable kill rate.
Title: Extending coolant life by centrifuging
Author: P. Tandy
Address: Alfa Laval Co. Ltd.,
Great West Road,
Brentford, Middlesex.

ABSTRACT

The problems which arise when coolants become contaminated with dirt, metal fines, lubricating oils or bacteria were outlined. Various methods of separation were described. The speaker also described how a centrifuge works and went on to deal with dimensioning and system layouts. He also detailed some case histories in which centrifuges are being used to extend coolant life and reduce operating costs.

Title: Significance and measurement of water activity
Author: T.A. Oxley
Address: Biodeterioration Centre,
University of Aston,
St. Peter's College,
Saltley,
BIRMINGHAM, B8 3TE.

ABSTRACT

Drying is one of the most generally effective physical methods for control of biodeterioration by microorganisms. Water activity (aw) is the most generally useful measure of the availability of water in a material and hence lowered water activity is the best measure of the effectiveness of drying. Water activity was shown to be very closely analogous to relative humidity and measurement of the equilibrium relative humidity in a small volume of air in close contact with a material was considered to be the best method for evaluating water activity in solids for which freezing point and boiling point methods are inapplicable. A new commercial instrument was described which utilises the high precision chilled mirror technique for measurement of dew point (and hence equilibrium relative humidity) in a measuring head of such low energy dissipation that it can be buried in a granular or powdered solid and reach equilibrium in 15 minutes. The use of this instrument to measure aw in tobacco, tea, wheat flour and other solids was described.

The publication of these abstracts by the Biodeterioration Society does not constitute publication in the usual sense. No reprints are available. Those wishing to pursue these subjects further should write directly to the authors at the addresses shown and not to the Society.
MICROFOULING ON METAL SURFACES EXPOSED TO SEAWATER

Sharon G. Berk, Ralph Mitchell, Ronald J. Bobbie, Janet S. Nickels, and David C. White

Summary

The formation of films that retard the efficient transfer of heat through metallic surfaces exposed to seawater is a complex process beginning with formation of a primary film that attracts microorganisms of increasingly complex morphology. These organisms and their extracellular polymers form the biofouling film. The morphology of this film appears to depend on the surface of the metallic surface but not on its chemical composition. The biomass and community composition of the microbial film can be quantitatively assayed with biochemical measures that can be correlated with resistance to heat transfer. This information can be used to perfect antifouling design modifications and chemical or mechanical cleaning countermeasures.

Microsalissures à la surface de métaux exposés à l' eau de mer

La formation de films retardant un transfert efficace de chaleur au travers de surfaces métalliques exposées à l'eau de mer est un processus complexe commençant par la formation d'un film primaire qui attire des microorganismes d'incroyable complexité morphologique. Ces organismes et leurs polymères extracellulaires forment le film de biooffsetures. Apparemment, la morphologie de ce film ne dépend pas de la texture de la surface métallique mais de sa composition chimique. La biomasse et la composition de l'ensemble du film microbien peut être quantitativement estimée par des mesures biochimiques que l'on peut utiliser à la résistance au transfert de chaleur. On peut utiliser cette information pour améliorer les modifications de conception anti-salissures ainsi que les moyens de nettoyage chimique ou mécanique.

Introduction

The development of films of microorganisms on electric power plant condenser metallic surfaces exposed to seawater imposes a severe load on heat transfer efficiency. This "microfouling" layer forms rapidly in nutrient-rich coastal waters, insulating the metal from the cooling effect of the seawater.

The formation of the microfouling film is usually inhibited by chlorinating the water, but this may lead to the formation of unacceptable pollutants. At present no completely acceptable alternative control for biofouling has been developed, although mechanical brushing can often be combined with chlorination for better control. Methods of microfouling control which are more environmentally acceptable will depend on understanding the mechanisms of film formation on heat exchanger metallic surfaces.

Degradacion Sobre Superficies De Metales Expostos A Agua De Mar

La formación de películas que retardan la transformación eficiente de calor a través de superficies de metal expuestas a agua de mar es un proceso complejo que empieza con la formación de una película primaria que atrae microorganismos de morfología de complejidad creciente. Estos microorganismos y sus polímeros extra celulares forman la película biodegradante. La morfología de esta película no depende aparentemente de la textura metálica sino de su composición química. La biomasa y la composición de la película microbiana puede ser cuantitativamente determinada con medidas químicas que pueden relacionarse con la resistencia al transferencia de calor. Esta información puede ser usada para diseñar modificaciones anti contaminantes y medidas de limpieza mecánicas o químicas.

Study of these films reveals a complex microbial community of bacteria, protozoa, fungi, and microalgae that become enmeshed in the polymeric fibrils of the primary film (Mitchell, 1978). The diversity of the microbial community is illustrated by scanning electron micrographs of various metallic surfaces exposed off Fort Lauderdale, Florida, and analyzed at the Harvard Laboratory of Applied Microbiology. The biochemical nature of the microfouling film from pipe sections exposed to seawater flowing at 1.8m. sec⁻¹ (6ft. sec⁻¹) in the Ocean Thermal Energy Conversion (OTEC) condenser simulating facility at the Naval Coastal Systems Center, Panama City, Florida (Braswell, et al., 1979) was analyzed at Florida State University (Bobbie et al., 1979).

The process which leads to the primary attachment of the microbial film is initiated by immediate coating of the surface by a layer of biopolymers produced by

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1 Presently at: Department of Environmental Sciences, University of Virginia, Charlottesville, VA 22903.

2 Laboratory of Applied Microbiology, Division of Applied Sciences, Harvard University, Cambridge, MA 02138.

3 Department of Biological Science, Florida State University, Tallahassee, Florida, 32306.

(Received December 1980)
organisms living in the sea (Baier, 1980). The organic layer is believed to attract microbes (Young and Mitchell, 1973). The adsorbed layer of biopolymer, which consists of proteins, polysaccharides and lipids, has a lower surface tension than the metal surface which it covers and thus increases the adhesion of the organisms. Free swimming bacteria are believed to be attracted to the higher nutrient concentrations of the film and attach reversibly. Shortly thereafter irreversible attachment and the secretion of acidic polysaccharide extracellular polymeric films begins (Marshall, 1976). Different surfaces have been reported to select fouling populations with different morphologies (Marsalek et al., 1979).

Effect of the Metallic Surface Texture on the Morphology of the Microfouling Organisms

One-inch square metal sheets were exposed to the sea at Nova University Ocean Sciences Center, Fort Lauderdale, Florida, two meters below high, and 0.5 meters below the low, tide level, and collected after 6, 10 and 15 days. Half the samples were sandblasted before immersion to test for effects of surface texture. The specimens were fixed in gluteraldehyde, dehydrated in acetone, and critical point dried, prior to scanning electron microscopy. The surface metallic texture apparently had little effect on the rates of microfouling. Comparison of rough and smooth surfaces of copper, stainless steel, and aluminum, titanium or stainless steel and at 15 days bacteria are the only organisms visible (Figure 1A). The fouling of stainless steel illustrates a progression with time showing bacteria (10 days) and protozoa (15 days). Aluminum surface corroded very rapidly and was covered by aggregates of bacteria with an occasional flagellate protozoan visible within 6 days (Figure 2). Longer exposure allowed development of stalked algae and sessile protozoans which are themselves coated with bacteria (Figure 3). The copper surface induced bacteria to form an extracellular film which can be seen peeling off the metallic surface (Figure 4). The peeling may be a fixation artefact.

Titanium fouls most rapidly of the metals tested and forms a diverse microbial community (Figure 5). The film contains microorganisms that do not appear on other metals.

Microfouling in Flowing Seawater Systems

Scanning electron microscopy shows that the microfouling film which forms on the surface of 5052 aluminum or titanium after 29 days' exposure to flowing seawater at Panama City, Florida, has a complex morphology (Figure 6). The resistance to heat transfer for the aluminum and titanium pipes was 5.8 x 10^-4 units when not fouled but increased to 8.0 x 10^-4 units after this time.*

Quantification of Biomass and Community Composition

To provide a quantitative measure of the biomass and community composition a series of biochemical analyses were applied as described by Bobbie et al. (1979) and summarized below.

The microfouling film was extracted, using sections of fouled pipe, with the single phase chloroform-methanol solvent which removes the lipid contents of the film. The lipids were then hydrolyzed and specific fragments purified by thin layer chromatography, assayed by capillary gas liquid chromatography, and identified by electron impact-mass spectrometry (Bobbie and White, 1980).

Examination of the data in Table 1 shows a number of significant differences between the biomass and community composition of the microfouling films on titanium and aluminum illustrated in Figure 6. Biomass can be estimated from the extractable lipid phosphate, and the total extractable palmitic acid (PA). All microbial cells contain membranes in which phospholipids are essential. Consequently measurement of the total extractable lipid phosphate measures the absolute amount of phospholipid in the microfouling film. Lipids are not only found in membranes but may accumulate in cells as a reserve energy storage material such as fat or triglyceride. PA is found in all lipids, phospholipid in membranes and in fats and is thus a measure of all lipids. The microbial animals (protozoa, metazoa) and plants (algae, fungi) contain fats the bacteria do not. Since all microbes contain phospholipids the ratio of lipid phosphate to palmitic acid can give an estimate of the ratio of the bacteria to the microfauna and microflora. The biomass of the bacteria is three times larger in the film from titanium than from aluminum. The microfauna and microflora, but not the bacteria, also contain polyenoic fatty acids (fatty acids with three or more double bonds). Examination of the increase in total extractable palmitate, the ratio of lipid phosphate, to palmitic acid and the total extractable polyenoic fatty acids, shows that the ratio of lipid phosphate to palmitic acid is formed only by bacteria containing the anaerobic desaturating pathway. Examination of the absolute amounts of these fatty acids again indicates that the microfouling film from titanium contains more bacteria of the types containing these fatty acids than the film on aluminum.

* The units for resistance to heat transfer are: hours X feet^2 X degrees F/ BTU X 10^-4. The equivalent values in S.I. units are: Unfouled pipe: 0.93 degrees C. m^2/w X 10^-4. After 29 days in flowing seawater: 1.28 degrees C. m^2/w X 10^-4.
Figure 1. Scanning electron micrographs of metal surfaces exposed to seawater for 10 days. A) rough copper; B) smooth copper; C) rough stainless steel; D) smooth stainless steel; E) rough aluminium; F) smooth aluminium. All scale bars = 10 μm.
The examination of the proportions of these fatty acids relative to the PA allows comparisons between the components making up the community composition that is independent of the total biomass.

The relative proportions of the bacterial short-branched, the cyclopropane fatty acids from the bacteria and the polyenoic fatty acids characteristic of the microflora and microfauna are significantly larger in the film from the titanium pipes.

Extracellular Polymer Formation

The organisms of the microfouling community secrete extracellular polymeric plaque (similar to that which forms on teeth). This can be estimated as the ratio of total organic carbon to extractable PA. In free fouling the aluminium and titanium surfaces contain a small proportion of extracellular polymer. The residual community left after brushing contains significantly more extracellular polymer.

In further applications of these techniques, the consequences of mechanical brushing of the film have been shown to be selectively to remove the components from both titanium and aluminium, and to change the community composition of the residual film. The residual film left on aluminium is greatly increased in extracellular polymer plaque compared to the film remaining on titanium and both are enriched in particular bacterial components. The titanium fouls faster but is more easily than the aluminium. Filamentous bacteria with extracellular polymers can be detected morphologically and biochemically in and under the corrosion gel in mechanically cleaned aluminium tubes. This can possibly account for the difficulty in cleaning aluminium surfaces. These data have been summarized and are, or will shortly be, published (Nickels et al., 1981a; 1981b).

Conclusions

1. The formation of microfouling film on metal surfaces suspended in seawater is unaffected by the texture of the surface.

2. The chemical composition of the metallic surface is an important factor in the quality and quantity of the film.
3. It appears that there is correlation between the large bacterial community on aluminium and the rapid rate of corrosion.

4. Titanium surfaces are colonized by a rich, diverse and unusual population when suspended in the sea.

5. Microfouling films formed in rapidly flowing seawater can be quantitatively examined as to biomass and community composition by biochemical measures.

6. The combination of morphological and biochemical methods can show insight into the microfouling community that can be used to optimize cleaning and prevention designs and procedure.

Acknowledgements

A portion of this work was supported by contracts from the Department of Energy administered by Dr. Peter H. Benson, Argonne National Laboratories, and was presented on June 3, 1980, at the 7th Ocean Energy Conference, Washington D.C. We would like to thank Mr. Ed Seling of the Museum of Comparative Zoology's Electron Microscope Facility, Harvard University, for his excellent technical assistance in obtaining many of the electron micrographs. We also wish to acknowledge NOVA University Ocean Science Center, Ft. Lauderdale, Florida, for facilities used during sampling periods.

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Characterization of benthic microbial community structure by high resolution gas chromatography

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Nickels, J.S., Bobbie, R.J., Lott, D.F., Martz, R.F., Benson, P.H. and White, D.C. (1981a)
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Applied and Environmental Microbiology, 41 (6):

Effect of cleaning with flow-driven (M.A.N.) brushes on the biomass and community composition of the marine microfouling film on aluminium and titanium surfaces.

Assay and correlation between microbial fouling and O.T.E.C. cleaning of surfaces exposed to seawater.
Figure 3. A) *Zoothamnium* colony on smooth aluminium surface after 15 days' exposure to seawater; B) higher magnification showing bacteria attached to the protozoan surfaces. Scale bars = 10 µm.

Young, L.Y. and Mitchell, R. (1973)
The role of chemotactic responses in primary film formation.
Northwestern University Press, Evanston, Ill. USA.

Figure 4. A) Sheets of material covering copper surfaces after 15 days' exposure to seawater; B) higher magnification of the material with bacteria on the surface. Scale bars = 10 µm.
Table 1
Biomass and community composition of the microfouling film on aluminium and titanium pipes in flowing seawater for 4 weeks.

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<th>Aluminium</th>
<th>Titanium</th>
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<tr>
<td><strong>Biomass</strong></td>
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<tr>
<td>Lipid phosphate a</td>
<td>1.41 (0.69)</td>
<td>4.21 (0.26)</td>
<td>***</td>
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<tr>
<td>Total palmitic acid (PA) a</td>
<td>0.45 (0.06)</td>
<td>0.60 (0.08)</td>
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<tr>
<td>Lipid phosphate/PA</td>
<td>3.06 (1.12)</td>
<td>7.22 (1.38)</td>
<td>***</td>
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<tr>
<td>Pentadecanoic acid</td>
<td>0.047 (0.002)</td>
<td>0.074 (0.001)</td>
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<tr>
<td>Anteiso + iso pentadecanoic acid</td>
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<td>0.382 (0.025)</td>
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<td>Linoleic acid</td>
<td>0.021 (0.003)</td>
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<td>Total polyenoic acids</td>
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<tr>
<td><strong>Community Composition</strong></td>
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<tr>
<td><strong>Bacterial</strong></td>
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<td>Anteiso + iso pentadecanoic acid/ pentadecanoic acid</td>
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<td>5.47 (0.45)</td>
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<td>Linoleic acid/PA</td>
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a  \( \bar{X} \) (± S.D.) in μmoles/36 inch pipe section;

b  *, **, *** indicate statistical significance by analysis of variance at the 0.1, 0.05 and 0.01 level of probability respectively.
Figure 5. Organisms found on titanium surfaces after 11 days' exposure to seawater. A) and B) peritrich protozoan covered with bacteria. Unidentified organisms C, D, E. A mulberry-surfaced bacterium, (F), has been observed only on titanium surfaces. Scale bars: A, B, C, and E = μm; D and F = 1 μm.
Figure 6. Scanning electron micrographs of the microfouling film on the 5052 aluminium surface (A and B) and the film on the titanium surface (C and D) that were fouled in flowing seawater for 29 days. Bar in A = 100 μm; B = 10 μm.
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BIODETERIOGENIC FUNGI IN TWO CANADIAN HISTORIC HOUSES SUBJECTED TO DIFFERENT ENVIRONMENTAL CONTROLS

J. David Miller and Harold Holland

Summary

Two buildings in an historical village in New Brunswick, subjected respectively to a minimum temperature and a maximum humidity control, were examined for the presence of potential biodeteriorogenic fungi. Thirty-nine species, many of them cellulolytische, were isolated from various surfaces in the two buildings. No differences attributable to the different types of environmental control could be discerned. Cellulosic substrates supported the greatest density and diversity of fungi.

Introduction

Fungal damage of historical objects in the Canadian climate has received little attention. In museum situations attempts are made to control environmental conditions to eliminate chemical and biological deterioration (Brommelle, 1968), but in an historical village such controls are not generally feasible.

Established in 1967, Kings Landing is an historical village 45 km from Fredericton, New Brunswick. Approximately 100,000 people visit the site per year. The 3.3 ha site consists of 60 buildings recreating all aspects of a nineteenth century Loyalist Village. Within the houses, shops and miscellaneous buildings are valuable historical objects. These include everything needed for life in rural nineteenth century Canada. Many of the cloth objects are made from wool and linen prepared on site.

In September, the village is closed and the buildings prepared for winter. All of the major buildings have hidden central heating systems, and these are turned on and the windows are insulated. The heating systems are controlled by conventional thermostats set to maintain a minimum temperature of 8°C.

In the past ten years, January temperatures have averaged from -6.4°C to -12.4°C and average minimum temperatures were -18°C (Anon, 1980). Heating this air to 8°C results in low indoor relative humidities and hence in undesirably low equilibrium moisture contents of materials within the buildings. Wooden objects are then particularly susceptible to damage by cracking (MacLeod, 1975). At the same time, the climate is influenced by maritime weather systems and temperatures can go above freezing and short period of rain are possible followed by extended cold. Under these circumstances, relative humidities within the buildings can become higher than the desired 50% (Garver, 1968), and fungal damage of the surfaces and artefacts becomes a possibility (Brommelle, 1968; MacLeod, 1975).

In an effort to combat these problems, the heating system of one building, the Long House, was converted in the autumn of 1979 to be controlled by a humidistat set at 50% R.H. This report describes the isolation of fungi from a building with a conventional heating system (Ingram House) and Long House over a 15 week period during winter. The purpose of the study was to determine the presence of biodeteriogenic fungi on various surfaces in the buildings and note any differences between the two treatments.

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2 New Brunswick Provincial Archives, P.O. Box 6000, Fredericton, N.B. E38 5H1.

(Received September 1980. In final form April 1981)
Materials and Methods

Surface swabs were taken of six representative areas in various rooms of each building as detailed in Table 1. The method used was similar to that proposed in Upsher (1977). Wet sterile discs (13 mm) of pure absorbent paper 'Schuler and Schuell 740-E) were carefully swabbed over a 5 cm path with forceps and each placed in a sterile 125 ml Erlenmeyer flask containing 10 ml distilled water with Tween 80 (10^(-3) %) stoppered with a cotton wool plug. Care was taken to sample within a space of 0.1 m² for a given sampling site, but never twice in exactly the same spot. The samples were returned to the laboratory and processed within one hour. The flasks were placed on a rotary shaker (250 rpm) for 15 minutes. One millilitre aliquots of the fluid were then plated using the spread plate method, on each of three plates of potato dextrose agar (PDA), and Park's (1973) modification of Eggins and Pugh (1962) cellulose agar (CA). These were incubated for two weeks at 20°C. The number of colonies and species were recorded in each case and isolations were made for identification and cellulolytic assay. Samples were taken in each building every two weeks from 18 December 1979 to 1 April 1980.

Species isolated were assayed for cellulolytic activity using the agar diffusion method (Rosenberg and Oberkotter, 1977). Walseth cellulose at a concentration of 1.25% was used in the agar, 18 ml of which was poured into culture tubes, 15 x 125 mm, and sterilized. Each of six tubes of the agar was inoculated with a 6 mm disc cut from the leading edge of a malt extract agar (2.5%) plate culture of the test isolate. The tubes were incubated at 23°C for six weeks and the depths of the clearing zones measured.

Temperature and humidity in the two houses were recorded on Belfort instruments (1600 South Clinton, Baltimore, U.S.A. 21224).

Walls, plaster and wallpaper, were examined for fungal growth using a portable UV light and visual inspection, and affected areas were photographed in the winter of 1980.

Results

Details of the sampling sites and average plate counts per ml sample on the two media are given in Table 1. No statistically significant differences were observed between the plate counts of the eight samples of any site.

Table 1 also shows average plate counts per ml sample of the three typical surfaces: cloth, wallpaper, and plaster. Cloth, such as that on a chair seat-cover, showed the highest densities of fungi, followed by wallpaper and plaster. No significant difference was observed between the plate counts per ml sample on PDA and cellulose agar.

Table 2 shows the species found on the three typical surfaces and also the results of the cellulolytic activity tests. No differences could be discerned between the species isolated in Long House and Ingram House. Approximately 62% of the species were isolated on cellulose agar. Cloth supported the most diverse mycoflora followed by wallpaper and then plaster. All species tested showed some cellulolytic activity and about half showed clearing zones equal to, or greater than, that of Trichoderma viride, a noted cellulolytic species.

Table 3 shows the temperature and humidity measurements in the two houses for the month of January 1980 and similar data for the environment.

Figure 1(a) shows the presence of fungi growing on wallpaper and plaster in a corner of a bedroom of Ingram House. No sign of direct water leakage in the attic above could be seen. Figure 1(b) shows the effect of a water leak near a chimney in Long House. Ultraviolet light showed the presence of fungal hyphae on the plaster and wallpaper.
Table 1

<table>
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<tr>
<th>House</th>
<th>Sample site</th>
<th>Material</th>
<th>PDA count/ml †</th>
<th>Cellulose count/ml †</th>
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<tr>
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<td>living room</td>
<td>sofa (cloth)</td>
<td>84 ± 27.2</td>
<td>145 ± 38.9</td>
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<td>(Humidistat</td>
<td>living room</td>
<td>wallpaper</td>
<td>26 ± 9.1</td>
<td>63 ± 15.9</td>
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<tr>
<td>controlled)</td>
<td>living room</td>
<td>wallpaper</td>
<td>21 ± 7.4</td>
<td>39 ± 13.8</td>
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<tr>
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<td>kitchen</td>
<td>plaster</td>
<td>12 ± 7.1</td>
<td>8 ± 2.8</td>
</tr>
<tr>
<td></td>
<td>kitchen</td>
<td>plaster</td>
<td>16 ± 6.0</td>
<td>36 ± 18.4</td>
</tr>
<tr>
<td></td>
<td>kitchen</td>
<td>sofa (cloth)</td>
<td>139 ± 25.1</td>
<td>106 ± 8.8</td>
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<tr>
<td>Ingram House</td>
<td>living room</td>
<td>chair (cloth)</td>
<td>48 ± 16.6</td>
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<td>(Thermostat</td>
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<td>wallpaper</td>
<td>97 ± 34.3</td>
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<td>bedroom No. 1</td>
<td>bed (cloth)</td>
<td>45 ± 11.0</td>
<td>107 ± 37.5</td>
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<td>plaster</td>
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<td>bedroom No. 2</td>
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<td>† Mean (all sites) cloth</td>
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<td>107</td>
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<td></td>
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<tr>
<td>† Mean (all sites) wallpaper</td>
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<td></td>
</tr>
<tr>
<td>† Mean (all sites) plaster</td>
<td>15</td>
<td>10</td>
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<td></td>
</tr>
</tbody>
</table>

† Average for the 8 samples ± standard error of the mean.

* Significant difference at the 95% level with a paired t-test between the noted pairs.

Means are taken from original data and are not precisely equal to means derived from the data in the upper part of the table.

Discussion

The swabbing method for the isolation of the fungi provided an indication of the numbers of propagules present and enables an approximate comparison of propagule numbers on each of the three general surfaces. It may be argued that the numbers of fungal propagules found on the various surfaces (Table 1) are reflective of activity, as is frequently done in soil mycology (Montegut, 1960). It is likely that the cloth surfaces are the most susceptible to biodeterioration, followed by wallpaper and then plaster, and the relative numbers of propagules may reflect this. It may also be that the nature of the surfaces facilitated accumulations of propagules.

One purpose of the study was to determine the presence of biodeteriogenic fungi. Most of the species isolated have previously been found to be biodeteriogenic (Table 2). The species which were most frequently isolated on cloth are all cellulolytic to varying degrees as shown in the same table. A number of other species isolated on cloth (Aspergillus repens, Penicillium frequentans, and P. simplicissimum) are noted for their ability to grow under low moisture conditions (Ayerst, 1969; Raper and Thom, 1968). The species isolated from plaster, Alternaria alternata and Cladosporium herbarum, are often found growing on the surface of plaster under damp conditions.

The materials used in the Kings Landing houses are original or crafted according to the original methods and contain no specific antifungal agents. The presence of these fungi thus attains more significance than they would if found on modern materials which may be treated with antifungal agents or fabricated from non-biodegradable components. Figure 1 shows two manifestations of the fungal problems outlined in this paper. First is the evidence that in some corners and other partially-enclosed locations, enough moisture can accumulate to allow the growth of fungi on the walls. The spot shown in Figure 1(a) increased in size over the one year study period. The second kind of problem is demonstrated in Figure 1(b). Liquid water can enter buildings in the winter (when they are generally unattended) and the data suggest that ample inoculum is present to allow fungal damage in addition to water damage.
The humidity data for Ingram House listed in Table 3 show that after a period of higher outside temperature and humidity, at least one week is required to reduce the inside humidity to the desired 50%. Since there is a moisture content desorption lag of materials, the return to the desired equilibrium moisture is further lengthened. The lower average temperature of Ingram House may also affect the problem as materials have increased equilibrium moisture contents at lower temperatures in relation to relative humidity.

In the humidity-controlled Long House, material equilibrium moisture contents were presumably more stable, which is advantageous for conservation (Coremans 1968), and were lower overall, presumably resulting in lower levels of material moisture content. However, the moisture content of the air in the humidity-controlled environment was higher than that in the temperature-controlled Ingram House, which would tend to predispose towards condensation on the walls in certain circumstances.

For short periods the higher temperatures in the Long House may have favoured fungal growth as higher temperatures enable fungi to grow in lower humidities in the mesophilic range (Ayers!, 1969). The present data do not provide an indication as to which treatment is preferable from the point of view of prevention of biodeterioration.

References
Anon (1980) Weather-le Temps, January 1980. Agriculture Canada Research Station P.O. Box 20280 Fredericton, N.B.
### Table 3
Temperature and humidity data for January 1981

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<tr>
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<th>Ambient Temperature °C</th>
<th>Ambient Relative Humidity %</th>
<th>Ambient Temperature °C</th>
<th>Ambient Relative Humidity %</th>
<th>Ambient Temperature °C</th>
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### Table 2
Fungi from various surfaces isolated on PDA and cellulose agar, and their cellulolytic activity

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<td>KL1-8</td>
<td>+</td>
<td>1 &amp; 3</td>
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<tr>
<td></td>
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<tr>
<td></td>
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<tr>
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<td>++</td>
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<td>KL2-12</td>
<td>+</td>
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</tr>
<tr>
<td>sterile</td>
<td>wallpaper</td>
<td>X</td>
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<td>wallpaper</td>
<td>X</td>
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<td>++</td>
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Notes to the table:

1. PDA = isolated on potato dextrose agar
2. Cell = isolated on Park's (1973) cellulose agar
3. Over 50% = numbers of samples in which the species accounted for more than 50% of the colonies
4. reported as biodeteriogenic
   1 = Coremans (1968)
   2 = Eggins and Pugh (1962)
   3 = Garver (1968)
   4 = Raper and Thom (1968)

Cellulolytic activity is indicated by the diameter of the diffusion test clearing zone:

+ = 1-3 mm
++ = 3-6 mm
+++ = 6-9 mm
++++ = 9-12 mm
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University of ASTON

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Dr. D. Allsopp, Biodeterioration Centre,
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St. Peter's College, College Road,
Birmingham B3 3TE England
PHYSIOLOGICAL STUDIES ON CYLINDROCARPON TONKINENSE BUGN. AN ISOLATE FROM DECAYING ARECANUT HUSK

N. Saraswathy

Summary
The effects of pH, carbon, nitrogen and C/N ratio on growth and sporulation of Cylindrocarpon tonkinense isolated from decaying arecanut husk were studied. The fungus was adapted to a wide range of pH though it grew best a pH 7.0. Of the carbon sources tested cellulose gave the maximum mycelial dry weight followed by sucrose, then lactose. Starch was a poor source of carbon. Eleven nitrogen sources were tested. Of these calcium nitrate yielded the greatest mycelial dry weight followed by potassium nitrate, sodium nitrate and peptone, in that order. Ammonium sulphate and ammonium phosphate (dibasic) were the poorest sources of nitrogen.

Seven different concentrations of carbon and nitrogen were studied yielding C/N ratios from 2.4 to 616. The higher concentrations of carbon produced abundant mycelial growth but a significant reduction in sporulation was noticed. The actual concentration of carbon was more significant than the carbon/nitrogen ratio.

Materials and Methods
A 7 day old monosporic culture of the fungus was grown in Czapek's liquid medium through the experiments. To study the effect of pH, the fungus was grown in Czapek's liquid medium adjusted to 8 different levels of pH ranging from 3.0 to 7.0 by addition of either 0.1 N HCl or 0.1 N NaOH as required. This experiment was repeated twice. To study the effect of carbon and nitrogen sources Czapek's liquid medium was modified by substitution of either a carbon or a nitrogen source in place of that contained in the basic medium in a quantity of the
Physiological studies on Cylindrocarpon tonkinense

N. Saraswathy

element equal to that which it replaced. Except for pH experiments the pH of the medium was adjusted to 7.0 before autoclaving.

The effect of carbon/nitrogen ratio was studied by adding different concentrations of sucrose or sodium nitrate. For all experiments three replicates were maintained for each treatment. Controls were kept on Czapek's medium from which either carbon or nitrogen was omitted as appropriate. The flask was seeded with a 5 mm fungal disc and incubated at room temperature (28 - 32°C).

Average mycelial dry weight was ascertained by harvesting the fungus on Whatman No.42 filter paper under reduced pressure on a Buchner funnel at the end of the incubation period (20 days). The final pH of the medium in all the experiments was recorded at the time of filtration. Sporulation of the fungus in all the above tests was observed by examining under a low power microscope and ranked:

- = no spores
+ = poor sporulation
++ = fair sporulation
+++ = good sporulation
++++ = excellent sporulation

Results and Discussion

Effect of pH

The fungus appeared to be adapted to a wide range of pH although more growth was observed at pH 7.0. Differences in mycelial dry weight between pH levels were not significant. Sporulation, also, was good at all pH levels except pH 3.0. In all the pH experiments the fungus changed the pH in an alkaline direction although the change was small at the higher initial levels of pH.

Effect of Carbon Source

Among the different carbon sources tested the greatest mycelial dry weight was recorded with cellulose (pure cellulose powder) (Table 1). Agarwal and Sarbhoy (1978) recorded excellent growth and sporulation of Fusarium oxysporum with cellulose as the carbon source. Glucose, cellobiose, xylose and galactose were almost on a par with each other in respect of growth and sporulation. Starch did not favour good growth, as has been reported for other fungi (Cochrane, 1958; Lilly and Barnett, 1951). Poor utilization of starch by C. tonkinense may indicate an inability to elaborate the enzyme amylase under these, or perhaps any, conditions. It is known that sucrose and maltose are good sources of carbon for most fungi, while lactose favours growth depending upon the length of the incubation period. The good growth obtained with C. tonkinense in lactose medium in the present study may be attributable to the fairly long incubation period.

Effect of Nitrate versus Ammonium Nitrogen

The fungus preferred the nitrate form of nitrogen to the ammoniacal form (Table 2). This preferential use of nitrate nitrogen rather than ammonium nitrogen by certain species of Colletotrichum has also been reported by Chaturvedi (1964); Kurtz and Fergus (1964); Cochrane (1958) and Srivastava and Saksena (1967). Calcium nitrate gave maximum mycelial growth while little or no sporulation was recorded in ammonium phosphate (dibasic) or ammonium sulphate. There was a drop in pH of the culture medium in all the nitrogen source experiments except where ammoniacal nitrogen was used. This may be due to ammonium assimilation from sulphates, nitrates, or chlorides, which is rapid and is often followed by a large drop in pH resulting in reduced growth (Cochrane, 1958). The fungus grew well with potassium nitrate and sodium nitrate, followed by sodium nitrite and peptone as nitrogen sources.

Effect of carbon and nitrogen concentrations and ratio

Seven different concentrations of each of carbon and nitrogen were used, ranging from one eighth to twice the concentration of each in Czapek's medium. In the basic medium, which contains 12.63 g/litre of carbon and 0.33 g/litre of nitrogen, the C/N ratio is 38.3. The ratios in the experiments ranged from 2.4 to 616. See figure 1. At the lower levels of carbon concentration, differences in growth and sporulation relative to nitrogen concentration were not significant. High concentrations of carbon induced profuse mycelial growth, but sporulation was reduced to a great extent. A similar increase in mycelial weight at higher concentrations of carbon has been reported for Helminthosporium oryzae (Das and Baruah, 1946) and for Fusarium spp (Agarwal and Sarbhoy, 1978).

Fig. 1 Effect of C:N ratio on the growth of Cylindrocarpon tonkinense

The fungus produced abundant four celled spherical spores at higher concentrations of carbon or nitrogen. The growth and sporulation of the fungus were altered more by carbon than by nitrogen concentrations. This indicates that the variation in growth and sporulation of the fungus may be due more to the actual concentrations of carbon and nitrogen than to their
Table 1

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<th>Carbon source</th>
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<th>Sporulation (mean of three replicates)</th>
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<td>Glucose</td>
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<td>Cellulose</td>
<td>258.3</td>
<td>+++</td>
</tr>
<tr>
<td>Starch</td>
<td>86.3</td>
<td>++</td>
</tr>
<tr>
<td>Control (no added carbon)</td>
<td>16.0</td>
<td>+</td>
</tr>
</tbody>
</table>

S.E. 14.61
L.S.D. (P = 0.05) 25.07

Table 2

<table>
<thead>
<tr>
<th>Nitrogen source</th>
<th>Dry wt of mycelium (milligrams)</th>
<th>Sporulation (mean of three replicates)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ammonium phosphate (monobasic)</td>
<td>138.3</td>
<td>+</td>
</tr>
<tr>
<td>Ammonium phosphate (dibasic)</td>
<td>88.3</td>
<td>-</td>
</tr>
<tr>
<td>Ammonium sulphate</td>
<td>70.0</td>
<td>-</td>
</tr>
<tr>
<td>Ammonium tartrate</td>
<td>143.0</td>
<td>+++</td>
</tr>
<tr>
<td>Calcium nitrate</td>
<td>200.0</td>
<td>+++</td>
</tr>
<tr>
<td>Potassium nitrate</td>
<td>191.7</td>
<td>+++++</td>
</tr>
<tr>
<td>Sodium nitrate</td>
<td>188.3</td>
<td>+++++</td>
</tr>
<tr>
<td>Sodium nitrite</td>
<td>186.0</td>
<td>+++</td>
</tr>
<tr>
<td>Urea</td>
<td>135.0</td>
<td>+++</td>
</tr>
<tr>
<td>Peptone</td>
<td>175.0</td>
<td>+++++</td>
</tr>
<tr>
<td>Control (no added nitrogen)</td>
<td>41.3</td>
<td>-</td>
</tr>
</tbody>
</table>

S.E. 17.51
L.S.D. (P = 0.05) 25.07


Acknowledgements

The author is thankful to Dr. M. Koti Reddy for help and guidance in completing the work and also acknowledges the help rendered by Mr. B.P. Nair, CPCRI (R.S.), Vittal, in statistical analysis.

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BN Preservation ... 3
BS Chemical ... 3
BT Toxins ... 3
CA Biological ... 4
CA Biocides ... 4
CP Plant ... 4
CR Animals ... 4
CF Edible ... 4
CG Inedible ... 4
CH Combined ... 4

EX Animal fibres ... EX Animal fibres ... EX Animal fibres ...
EV Adhesives ... EV Adhesives ... EV Adhesives ...
FE Lignin ... FE Lignin ... FE Lignin ...
FG Cellulose ... FG Cellulose ... FG Cellulose ...
FH Vegetable fibres ... FH Vegetable fibres ... FH Vegetable fibres ...
FI Timber ... FI Timber ... FI Timber ...
FJ Bacterial attack ... FJ Bacterial attack ...
FM Marine/aquatic fungal attack ... FM Marine/aquatic fungal attack ...
FO Insect attack ... FO Insect attack ...
FP Marine borer attack ... FP Marine borer attack ...
FR Preservation ... FR Preservation ...
FT Wood pulp ... FT Wood pulp ...
FU Paper ... FU Paper ...
FX Books, etc... FX Books, etc...
FY Toxins ... FY Toxins ...

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ISOLATION OF THERMOPHILIC AND THERMOTOLERANT FUNGI FROM STORED GROUNDNUTS IN NIGERIA AND DETERMINATION OF THEIR LIPOLYTIC ACTIVITY

Vincent W. Ogundero

Summary

Nine species of thermotolerant and thermophilic fungi were obtained from stored groundnuts in Nigeria. The former had a high occurrence at 45°C while the latter were more readily isolated at 50°C. Humicola lanuginosa (Griffon & Maublanc) Bunce, Mucor pusillus Lindt and Thermoaucus aurantiacus Miehe are known zoopathogens and are potential human pathogens. All the isolates were able to induce changes in the oil and free fatty acid content of groundnuts when grown on the blended seeds. A decrease in the oil content of the seeds due to mould activity was accompanied by a rapid increase in their free fatty acid content as period of incubation increased. The isolates caused high percentage increases in the free fatty acid contents of various natural oils used as sources of carbon and made appreciable growth on the sodium salts of these acids. This demonstration of their lipolytic nature shows that they are able to participate in the spoilage of groundnuts during storage.

Introduction

The groundnut, Arachis hypogea Linn. is an economically important export crop of Nigeria. The seeds are edible and the oil (groundnut oil) extracted from them is used extensively in soap manufacture. Mould infestation of the pods pre-harvest and during storage is responsible for the potent mycotoxins (e.g. aflatoxin) often detected in the seeds on analysis (Bampton, 1963; Eldridge et al., 1965; McDonald and Harkness, 1965), and to the formation of free fatty acids from the constituent glycerides (Ogundero, 1980a). The free fatty acids impart a rancid flavour to the extracted oil thus reducing its market value since the quality of the oil depends on the free fatty acid content.

Thermophilic fungi, as defined by Cooney and Emerson (1964) are fungi which grow at a minimum temperature of 20°C and above, and a maximum temperature of 50°C and above. Fungi with minimum growth temperatures below 20°C and maxima near 50°C are regarded as thermotolerant. The activity of these fungi in relation to the spoilage of agricultural products during storage has been infrequently studied in the past. Thermophilic and thermotolerant fungi

1 Department of Biological Sciences, University of Ilorin. Present address: Department of Botany, University of Ibadan, Ibadan, Nigeria. (Received September 1980; in revised form January 1981)
have been variously implicated in the deterioration of products such as tobacco (Pounds and Lucas, 1972; Ogundero, 1980b), palm produce (Eggins and Coursey, 1964; Oso, 1974a), snuff (Tansey, 1975) and moist barley grains (Mulinge and Apinis, 1969). A few species have also been reported from stored groundnuts (Taber and Petit, 1975).

The high revenue-yielding nature of Nigerian groundnuts, coupled with the potential health risks which the presence of mycotoxins in the seeds pose to the consumers, makes further studies of the microorganisms responsible for the deterioration of this product during storage necessary. This study deals with the isolation, and a determination of the lipolytic activity, of the thermophilic and thermotolerant fungi associated with stored groundnuts in Nigeria.

**Materials and Methods**

**Isolations**

Random samples of approximately 25g. each, of groundnuts were collected from 50 locations in the groundnut growing areas of northern Nigeria and from storage depots at the wharf in the humid south. The seeds were washed thoroughly with distilled water and surface sterilized with 0.01%, w/vol. Hg,C12 for 5 minutes. They were then rinsed in several changes of distilled water and plated (5 seeds per plate) on a 2% (w/vol) malt-yeast-extract agar medium as follows:

- malt extract: 20 g
- yeast extract: 2.5 g
- NaNO₃: 0.2 g
- KCl: 0.05 g
- sodium glycerophosphate: 0.05 g
- FeSO₄.7H₂O: 0.01 g
- Agar: 15 g
- distilled water: 1000 ml
- pH 6.9

and also on a vegetable oil medium selective for the growth of lipolytic fungi (Eggins and Coursey, 1964). Six plates of each sample were incubated at 45°C and six others at 50°C. The plates were examined daily for fungal growth. Mycelia from the separate colonies were transferred with a sterile mounted needle to fresh malt-yeast-extract agar plates until pure cultures were obtained. The frequency of occurrence of each fungal colony was also determined. The different isolates were identified and subsequently sent to the Commonwealth Mycological Institute, Kew, Surrey, England, for confirmation. These were maintained on malt-yeast-extract agar slants and deposited at the Mycological Collection Unit, Botany Department, University of Ibadan, Nigeria.

**Determination of growth-temperature requirements**

The various isolates were grown on malt-yeast-extract agar plates at temperatures of 10°C, 15°C, 20°C, 37°C, 45°C and 50°C ± 1°C for 5 days (5 days for each isolate and at each temperature) and the growth was scored according to the scheme given in the footnote to table 2. This method is particularly useful in determining the growth-temperature relations of filamentous fungi and has been extensively used (Bracanto and Golden, 1953; Trinci, 1969; Milner, 1977; Ogundero, 1980c) since trace growths can be detected by subjecting the petri dishes to microscopic examination. Agar-mycelial discs of each isolate, 5mm diameter, were obtained from young mycelia of 5-day old cultures on malt-yeast-extract agar plates and used as inocula.

**Degradation of blended groundnut seeds**

This was carried out by growth of the various isolates on a groundnut seed medium at temperatures which had previously been found to be optimal, namely, 37°C for thermotolerant isolates, and 45°C for the thermophilic isolates. Because it would have been difficult to work with whole groundnut seeds a blended medium was preferred. Freshly harvested, uninoculated groundnut seeds were hand-picked, rinsed in several changes of water and macerated in a Waring blender for 3 minutes. The blended stuffs were dried in the oven at 80°C for 24 hours. Twenty gram portions of the blended material were placed in each of several 250 ml conical flasks and 20 ml of distilled water added before autoclaving at 1.02 kg/cm² for 15 minutes. The pH of this medium after autoclaving was 6.9. The flasks were inoculated each with a 5 mm diameter agar- mycelial disc of each of the isolates and incubated at the appropriate temperatures. At 4-day intervals the pH of the contents of each flask was determined with a pH meter. The oil content was also determined by drying and extraction in a Soxhlet apparatus for 4 hours with boiling petroleum ether (B.P. 60°- 80°C). The free fatty acid content of a known volume (1 ml) of the extracted oil (to which 10 ml of 95% ethanol had been added) was titrated with 0.02 N NaOH using phenolphthalein (1g/500ml of 50% v/v ethanol) as indicator (Coursey and Eggins, 1861; Kuku and Adeniji, 1976). Uninoculated flasks of the groundnut medium, similarly treated, served as control.

**Hydrolysis of vegetable oils**

The various isolates were grown for 7 days on inoculation into 30 ml portions of a medium containing the following:

- vegetable oil: 20 g
- NaNO₃: 0.2 g
- K₂HPO₄: 1.0 g
- MgSO₄.7H₂O: 0.5 g
- biotin: 5 ug
- thiamine: 100 ug
- water: 1 litre

The various isolates were grown for 7 days on inoculation into 30 ml portions of a medium containing the following:
To this is added 1 ml of a micronutrient solution as follows:

- ZnSO₄·7H₂O - 439 mg
- Fe(NO₃)₃·9H₂O - 725.5 mg
- MgSO₄·4H₂O - 203 mg

distilled water to 1 litre.

This medium (Ogundero and Oso, 1980) was autoclaved and the pH determined before and afterwards. The inoculated media were incubated at the appropriate temperatures. The vegetable oils used were: groundnut oil, palm kernel oil, palm oil and corn oil. At the end of the incubation period the free fatty acid content of each flask was determined titrimetrically with 0.02 N NaOH using phenolphthalein as indicator (Kuku and Adeniji, 1976).

Utilization of free fatty acids as sole sources of carbon for growth was determined by growth on a medium containing each of palmitic, stearic, oleic and lauric acids. The appropriate weights of each to yield 1 g of carbon per litre was suspended in 250 ml of distilled water and 0.2 N NaOH added while warming until a pH of 12 was attained to form the sodium salts. The pH was then brought down to 6.9 by the addition of 0.2 N HCl before autoclaving (Oso, 1974b). This procedure was adopted because it is difficult to disperse the fatty acids themselves in an aqueous medium. The pH of the medium did not change on autoclaving. Portions, each of 10 ml, were added aseptically to 20 ml of the sterile basal medium described above, and inoculated with the isolates. The cultures were incubated for 7 days, the mycelia produced harvested by suction filtration through pre-weighed and dried sintered glass crucibles, oven dried (80°C for 24 hours) and weighed.

Results and Discussion

Isolations

Nine species of fungi were obtained from the various samples analysed (Table 1). Aspergillus fischeri, Chaetomium globosum and Chrysosporium thermophilum are thermotolerant (Table 2) with trace growths near 50°C and fair growths at 15°C. The other species are thermophilic, with no growth recorded below 20°C and good growth at 50°C (Cooney and Emerson, 1964). Humicola lanuginosa, Mucor pusillus and Thermoascus auranticus are known zoopathogens and are potential human pathogens (Cooney and Emerson, 1964; Lacey, 1975; Ogundero, 1979). Their high occurrence on stored groundnuts as reported here is of concern because of the health risks posed to consumers.

Degradation of blended groundnut seeds

All the isolates were able to induce changes in the blended groundnuts (Table 3) leading to a rapid decrease in the oil content and a corresponding rise in the free fatty acids of the medium. With increase in the period of incubation, a general decrease in the pH of the medium was also recorded (Table 3). This is due to the increased production of free fatty acids with period of growth.

Hydrolysis of vegetable oils and sodium salts of fatty acids

Results in Table 4 show that a high increase in the free fatty acid contents of the natural oils were obtained while appreciable mycelium production occurred on the sodium salts of the fatty acids. The ability of the isolates to liberate free fatty acids from the vegetable oils is an indication of their lipolytic nature and, indeed, of their ability to produce lipases extracellularly. In an earlier study (Ogundero, 1980a) the extracellular lipases of the thermophilic natural glycerides and sodium salts to free fatty acids between a pH range of 5.5-6.0 and a temperature range of 45°C - 50°C.

The high occurrence of these fungi on stored groundnuts, coupled with their ability to hydrolyse the oils in blended groundnuts, is an indication of their participatory roles in the spoilage of these products during storage.

Acknowledgements

The author is grateful to the following members of staff of the Commonwealth Mycological Institute for identifying the various specimens: Dr. J.E.M. Mordue, Mrs. J.A. Lunn, Dr. A.H.S. Onions, Dr. B.C. Sutton and Dr. M.B. Ellis. A grant from the Senate research fund of the University of Ilorin made this study possible.

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Table 1

Thermophilic fungi from mouldy groundnuts
Percent occurrence in 40 samples at two temperatures

<table>
<thead>
<tr>
<th>Species</th>
<th>IMI*</th>
<th>Incubation temperature</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>45°C</td>
</tr>
<tr>
<td>Aspergillus fischeri</td>
<td>204909</td>
<td>36%</td>
</tr>
<tr>
<td>Chaetomium globosum</td>
<td>196748</td>
<td>17%</td>
</tr>
<tr>
<td>Chrysosporium thermophilum</td>
<td>196674</td>
<td>14%</td>
</tr>
<tr>
<td>Humicola grisea Traen var.</td>
<td>204924</td>
<td>16%</td>
</tr>
<tr>
<td>Humicola lanuginosa</td>
<td>196682</td>
<td>2%</td>
</tr>
<tr>
<td>Mucor pusillus</td>
<td>204905</td>
<td>18%</td>
</tr>
<tr>
<td>Talaromyces thermophilus</td>
<td>204907</td>
<td>9%</td>
</tr>
<tr>
<td>Thermoascus aurantiacus</td>
<td>196722</td>
<td>4%</td>
</tr>
<tr>
<td>Thermoascus crustaceus</td>
<td>204916</td>
<td>17%</td>
</tr>
</tbody>
</table>

* IMI = Commonwealth Mycological Institute identification numbers
** Thermotolerant species

Table 2

Growth-temperature relations of fungi from stored groundnuts

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature (± 1°C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10°</td>
</tr>
<tr>
<td>Aspergillus fischeri</td>
<td>++</td>
</tr>
<tr>
<td>Chaetomium globosum</td>
<td>++</td>
</tr>
<tr>
<td>Chrysosporium thermophilum</td>
<td>++</td>
</tr>
<tr>
<td>Humicola grisea var. thermola</td>
<td>-</td>
</tr>
<tr>
<td>Humicola lanuginosa</td>
<td>-</td>
</tr>
<tr>
<td>Mucor pusillus</td>
<td>-</td>
</tr>
<tr>
<td>Talaromyces thermophilus</td>
<td>-</td>
</tr>
<tr>
<td>Thermoascus aurantiacus</td>
<td>-</td>
</tr>
<tr>
<td>Thermoascus crustaceus</td>
<td>-</td>
</tr>
</tbody>
</table>

Key to growth scores:
- = no growth                     ++ = fair growth
+ = trace of growth               +++ = very good growth

Table 3

Mould induced changes in groundnuts during incubation at 37°C or 45°C. Each figure is the mean of 3 readings.

<table>
<thead>
<tr>
<th>Species</th>
<th>Incubation temperature</th>
<th>Days of incubation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>37°C OC</td>
<td>4 8 12</td>
</tr>
<tr>
<td>Aspergillus fischeri</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Chaetomium globosum</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Chrysosporium thermophilum</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Humicola grisea var. thermola</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Humicola lanuginosa</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Mucor pusillus</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Talaromyces thermophilus</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Thermoascus aurantiacus</td>
<td>ffa</td>
<td>phr</td>
</tr>
<tr>
<td>Thermoascus crustaceus</td>
<td>ffa</td>
<td>phr</td>
</tr>
</tbody>
</table>

CONTROL 45°C OC 29.64 29.65 29.65
ffa 6.26 6.34 6.31
phr 6.94 6.92 6.94
Table 4
Hydrolysis of oils and fatty acids by the isolates on incubation at 37°C or 45°C for 7 days.
Each figure is the mean of 3 readings.

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature of incubation</th>
<th>Sodium salt of Natural fats</th>
<th>Dry wt. of mycelium mg/30 ml</th>
<th>Free fatty acid % increase</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Palmitic acid</td>
<td>Oleic acid</td>
<td>Stearic acid</td>
</tr>
<tr>
<td>Aspergillus fischeri</td>
<td>37°C</td>
<td>68</td>
<td>96</td>
<td>48</td>
</tr>
<tr>
<td>Chaetomium globosum</td>
<td>37°C</td>
<td>124</td>
<td>98</td>
<td>84</td>
</tr>
<tr>
<td>Chrysosporium thermophilum</td>
<td>37°C</td>
<td>69</td>
<td>142</td>
<td>126</td>
</tr>
<tr>
<td>Humicola grisea v.thermoideas</td>
<td>45°C</td>
<td>101</td>
<td>69</td>
<td>88</td>
</tr>
<tr>
<td>Humicola lanuginosa</td>
<td>45°C</td>
<td>72</td>
<td>49</td>
<td>106</td>
</tr>
<tr>
<td>Mucor pusillus</td>
<td>45°C</td>
<td>82</td>
<td>48</td>
<td>54</td>
</tr>
<tr>
<td>Talaromyces thermophilus</td>
<td>45°C</td>
<td>72</td>
<td>65</td>
<td>69</td>
</tr>
<tr>
<td>Thermoascus aurantiacus</td>
<td>45°C</td>
<td>88</td>
<td>46</td>
<td>63</td>
</tr>
<tr>
<td>Thermoascus crustaceus</td>
<td>45°C</td>
<td>56</td>
<td>64</td>
<td>75</td>
</tr>
</tbody>
</table>
THE ROLE OF FUNGI AND BACTERIA IN THE CONSOLIDATION OF BOOKS

Alicja B. Strzelczyk and Stanisława Leznicka

Summary

Both fungi and bacteria participated actively in the consolidation of fragments of books from the XVIIth and XIXth centuries. A large proportion of the bacteria isolated produced slimes. Under experimental conditions damage identical with the appearance of consolidated ancient books was obtained and this process was shown to be due to the growth of microorganisms which show a progressive change in species composition as the process proceeds. The intensity of the degradation of paper in XVIIth and XIXth century books and the consolidation of the samples is highly affected by the fibre content of the paper and the type of glue used.

Le rôle des champignons et des bactéries dans la consolidation des livres.

Les champignons et les bactéries ont participé activement à la consolidation de fragments de livres du XVII et XIXèmes siècles. Un grand nombre des bactéries isolées produisaient des boues. Dans des conditions expérimentales, on a obtenu des dommages identiques en aspect à celui des anciens livres consolidés et on a vu que ce processus était du à la croissance de microorganismes qui présentent un changement progressif dans la composition des espèces au cours de ce processus. L’intensité de la dégradation du papier dans les livres des XVII et XIXèmes siècles et la consolidation des échantillons est grandement affectée de la teneur en fibres du papier et le type de colle utilisées.

Introduction

Among the most serious types of damage to ancient library collections is the "consolidation" of books. This damage affects volumes which have been flooded or heavily moistened. A "consolidated" block of a book is usually overgrown by microorganisms and its leaves are glued together and stained. The growth is heaviest at the edges and is accompanied by the strongest consolidation of the leaves. The edge part of the book becomes thin and, on drying, is strongly deformed. The middle and back parts are usually much less damaged. Paper in a consolidated book is swollen, weak, and loses its leafy structure, becoming a solidified mass. Figure 1.

The type of damage in consolidated books suggests the role of cellulolytic microorganisms in this process. The interaction of different groups of microorganisms with varied cellulolytic ability may result in complete decomposition of this paper.

The intensity with which paper is degraded by cellulolytic microorganisms is affected by the presence of natural glues in the paper (Belaya et al. 1964; Nuksha, 1964). Delignified, short cellulose fibres are degraded more easily than long fibres with a higher lignin content (Basu and Ghose, 1962; Cowning and Brown, 1969; Berg et al., 1972).

The characteristics of cellulolytic enzymes of fungi which destroy ancient paper have been described elsewhere (Laznicka, in press). The complete degradation of cellulose in paper is effected by two groups of cellulases: endoglucanases which hydrolyse internal beta-1, 4-glucoside linkages and exoglucanases which split off mono- or di-saccharide units from the non-reducing end of a cellulose chain (Bergheim and Pettersson, 1973; Eriksson and Pettersson, 1975; Shikata and Nizisawa, 1975).

The intermediates of cellulose degradation have been found to be oligosaccharides with mucose properties (Gascoigne and Gascoigne, 1960; Berg et al., 1972; Ross and Strzelczyk, 1979). Moreover, on substrates rich in sugar, microorganisms form secondary products of a slimy nature. This phenomenon has been observed mainly in paper mills; slime of biological origin causes staining of the cellulose mass, tearing of paper tape in paper machines and clogging of paper mill pipes (Nelson, 1962; Szwarestain, 1968; Wolfson and Michalski, 1964). Organisms isolated from, and dominating in the slime include fungi and cellulolytic bacteria as well as bacteria forming muclaginous capsules (Martin and Dobson, 1945; Turner, 1967; Hughes, 1968; Gascoigne and Gascoigne, 1960).

Die Rolle von Pilzen und Bakterien beim "Verbacken" von Büchern


La función de hongos y bacterias en la consolidación de los libros

Hongos y bacterias participaron activamente en la consolidación de fragmentos de libros pertenecientes a los siglos XVII y XIX. Una proporción grande de las bacterias aisladast produjeron mucosidad. Se obtuvo bajo condiciones experimentales una distorsión identica en su apariencia a la de los libros antiguos consolidados y este proceso parecía ser debido al crecimiento de los microorganismos que muestran un cambio progresivo en la composición de las especies mientras el proceso tiene lugar. El contenido en fibra del papel y el tipo de cola usada afecta mucho a la densidad de degradación en los libros mencionados y la consolidación de las muestras.

Laboratory of Paper and Leather Conservation, Institute of Conservation and Restoration, N. Copernicus University, Torun, Poland.

1 The author used the term "Petrifaction" in the title and throughout. This term is not considered by the referees or the Editor to be suitable because its literal meaning in English is "turned to stone". The Editor is responsible for the use of the term "consolidation". (Received January 1981)

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Capsular mucus is produced abundantly by bacteria on media in which the amount of carbon compounds is greatly in excess of the amount of salts or nitrogen compounds. The production of jelly-like and swelling slime by bacteria depends on the source of carbon (Berg et al., 1972).

Fungi may also produce large amounts of viscous substances, depending on the source of carbon (Sanborn, 1965; Gascoigne and Gascoigne, 1960; Ross and Strzelczyk, 1979). These substances may be assumed to play a role in the consolidation of a book into a block. The role of fungi and bacteria in the consolidation of books has not yet been studied. The aim of the studies presented herein was to investigate the role of fungi and bacteria in the consolidation of books made from different papers.

Materials and Methods

The material studied consisted of fragments of books from the XVIIth and XIXth centuries. The samples used were taken from the destruct of a XVIIth century book lacking title page and the first and last pages. Their use in these studies was agreed with the Head of the Division of Old Prints of the N. Copernicus University Library, Torun. The library contains, in addition to this destruct, three identical undamaged copies.

Determination of fibre content and type of glueing

The fibre content of the book fragments chosen for study was determined on the basis of morphological differences between fibres and their staining by Herzberg’s reagent (Modrzejewski et al. 1966). The type of glueing was determined according to the methods recommended by Browning (1969) and used in the cellulose-paper industry (Modrzejewski et al., 1966).

Preparation of XVIIth and XIXth century book samples for consolidation studies

Samples of books from the XVIIth century with dimensions 135 by 156 mm and thickness 35 mm were tied with nylon thread to ensure good compaction of the leaves. (Figure 2A) The samples were treated as follows:

Sample 1 control, not moistened.

Samples 2-4 moistened for 10 minutes by immersion of one side in water. This procedure was repeated every 2 weeks.

Sample 5 moistened as above in distilled water containing 40 μg sodium pentachlorophenolate (PCFNa) per ml as a preservative.

Sample 6 moistened as above in distilled water with 10μg phenyl mercuric acetate (PMA) per ml as a preservative.

This experiment was carried out for six months.

Samples of the book from the XIXth century with dimensions 95 by 125 mm and thickness 15 mm were tied with nylon thread as above and subjected to a similar treatment, except that no preservatives were employed, i.e. no samples equivalent to 5 and 6 above.

Once a month from the beginning of the experiment microbiological analyses of paper samples taken from two sites 10 cm apart were carried out. The paper samples, after preparing a suspension, were examined by plating out on:

a. Malt extract medium (Strzelczyk, 1967) and Martin’s medium (Martin 1950) to determine the generic composition of the fungi.

b. Nutrient agar to determine the total number of bacteria. The percentage of capsulated bacteria was also estimated.

c. Medium for cellulolytic bacteria (Berg et al., 1971) in which cellulose powder was substituted by a disc of filter paper as the sole source of carbon.

Platings b and c above were begun after 3 months duration of the experiment.

Identification of fungi growing on the book fragments

Identification of the dominant strains of fungi on the book fragments was made directly by microscopical examination, whereas the accompanying fungi were identified by an indirect method.

Samples of paper were put into sterile, stoppered, centrifuge tubes containing 10 ml distilled water. They were then left for one hour, with frequent mixing, until the paper was defibrated. This ‘initial suspension’ was then diluted 100 and 1000 fold and 1 ml portions were plated on malt extract and Martin’s agar. Fungal colonies were identified after fructification at 23°C. The colonies of the identified fungi were compared with the appearance of the fungi on the books.

Determination of the composition of bacteria growing on book fragments

The initial suspension mentioned above was analysed. The suspension (in sterile stoppered centrifuge tubes) was centrifuged for 30 mins. at 3000 x g. One ml of the supernatant was transferred to a sterile Petri plate to which 9 ml nutrient agar was added. Dilutions of the supernatant were chosen so as to obtain not more than...
20 colonies per plate. After 3 day incubation at 23°C, the colonies were analysed and the percentage of encapsulated bacteria determined. The latter were identified in nigrosin preparations. The results were calculated per 100 mg dry weight of the paper taken for the analysis.

From the platings made at the monthly stages of growth (stages III - VI) 10 strains of bacteria were isolated to determine the generic composition of the dominant organisms. The strains isolated were determined according to Skerman (1967).

**Determination of the presence of cellulolytic bacteria**

The fungal, spore-free initial suspension (above) was diluted and used to inoculate medium with filter paper to test for cellulolytic bacteria. The surface of the medium was inoculated at 3 points with drops of the fluid. The presence of cellulolytic bacteria was determined after 10 day incubation at 23°C.

**Cellulolytic enzyme activity in paper from consolidated books**

The purpose of this analysis was to determine the activity of cellulases released to the environment by microorganisms responsible for the damage. Five grams of paper taken from the most heavily overgrown part of the XVIIth and XIXth century books were put into 150 ml of 0.2 M acetate buffer, pH 5.5 and left for 24 hours in a refrigerator with frequent mixing. The suspensions were then centrifuged for 30 mins. at 3000 x g, and cellulase activity in the supernatant was determined: exoglucanases according to Nelson and Somogyi (Mejbbaum-Katzenellenbogen & Mocznacka, 1960) and endoglucanases by the method of Horton and Keen (Szajer et al., 1969). Protein content was determined according to Lowry (Brzeski and Kanuga, 1968). The results were calculated per 1 g dry weight of paper taken for analysis.

**Effect of paper from XVIIth century book and Whatman paper on changes in cellulolytic activity of Penicillium notatum**

The purpose of this experiment was to seek an explanation for the absence of exoglucanase activity in consolidated XVIIth century paper samples since exoglucanase had been suspected to adsorb to the fibrous structure of cellulose.

The fungal strain used in the experiment was *Penicillium notatum* isolated in earlier studies (Lesnica, in press). A medium with cellulose powder according to Aschan and Norkrans (Szajer et al., 1969) was used. Fifty ml portions of the medium were placed in Erlenmeyer flasks, autoclaved, and inoculated with 0.5 ml of an aqueous suspension of spores of the chosen fungal strain per ml. After incubation for 14 days at 23°C the medium with mycelium was centrifuged at 3000 x g. The culture fluids from three centrifuged samples were pooled and then divided into three 50 ml aliquots. The following combinations were employed:

- 50 ml culture fluid with 2 g of XVIIth century paper
- 50 ml culture fluid with 2 g Whatman No. 1 paper
- 50 ml culture fluid without any additions (Control)

After 24 hours in a refrigerator the exo- and endoglucanase activity and protein content in each sample was determined.

**Effect of microflora growing on XVIIth and XIXth century books on the chemical properties of the paper**

a. Determination of alpha-cellulose content in the paper

The copper index is a measure of reducing ability of cellulose which reflects the number of free substrate ends. Pieces of paper from the control samples, and from the most contaminated book fragments of XVIIth and XIXth century were shredded and dried to constant weight at 105°C. Determinations were made according to Schwab and Haggland (Modrzejewski et al., 1965).

b. Determination of copper index.

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associated fungal populations. The fungi growing on book fragments were accompanied by numerous bacteria whose generic composition is given in Table 1. The following genera were present: Bacillus, Pseudomonas, Escherichia, Streptococcus, and Micrococcus. After some time typical aerobic cellulolytic bacteria - Sporocytophaga and Cytophaga - appeared. Their presence in the samples was evident in the form of mucose yellow or orange spots.

With time the proportion of capsulated bacteria greatly increased compared to total bacterial number. The percentage of these bacteria more than doubled from the third to the fifth month (Table 2).

The intensity of growth of microorganisms on the moistened edges of the book fragments was very high and the morphological changes in the samples caused by their development were very considerable. After termination of the experiment and drying of the samples, severe consolidation of the moistened side was observed. The initial 35 mm thickness of the samples decreased to 15 m (Figure 2B). Attempts to separate the leaves resulted in their tearing outside the glued part. The whole edge remained intact or separated in the form of thick pieces of several leaves glued together (Fig. 2C). The leaves themselves were brittle, frail, and sometimes crumbled; they were heavily stained by the fungal colonies.

The samples moistened with water containing preservatives (samples 5 and 6) were not attacked by the microorganisms.

The aqueous extract from the book fragments did not show any exoglucanase activity but had considerable endoglucanase activity (Table 3). The increase in protein content in the consolidated samples was not high compared with the control. However, in view of the fact that a great part of the glue originally used in the manufacture of the paper was utilised by the microorganisms, the increase in protein content was significant (Table 3). The absence of exoglucanases in the consolidated paper samples, as shown in later experiments, was caused by their adsorption to the paper of the books. The degree of adsorption was much greater on the sections from the XVIIth century book than on Whatman paper (Table 4). It was also found that the endoglucanase activity in filtrates supplemented with paper from the test book and Whatman paper was lower than in the filtrate without any additions (Table 4).

Growth of the microorganisms resulted in decreased alpha-cellulose content and accompanying great increase in the copper index (Table 5).

![Fig. 1 Ancient fungus-infested book. Formation of consolidated block of book.](image1)

![Fig. 2 Damage of XVIIth century book caused by microorganisms](image2)

A control sample  
B consolidation of book fragments after 6 months growth of microorganisms  
C attempts to separate the leaves of the book. Visible tears outside the part consolidated.
Fig. 3 Damage of XIXth century book caused by microorganisms

A control sample
B appearance of the samples after 6 months growth of microorganisms. No visible consolidation.

Dynamics of fungal and bacterial growth on XIXth century moistened book fragments

Study of the fibre content and type of paper gluing in the XIXth century book showed that its paper was produced from coniferous wood pulp glued with colophony and casein glue.

Table 6 presents the dynamics of growth of the fungi and bacteria during the 6 months period of the experiment. During this whole period the fragments of paper from the book were dominated by *Penicillium* sp., and after 4 months also by *Verticillium cinnabarum*. This in turn was dominated in the fifth and sixth months by *Chaetomium* sp and *Penicillium* sp. The accompanying fungi included *Trichoderma* sp. and *Cladosporium* sp.

The growth of the isolated fungi was poor and delicate, in the form of single, narrow, colonies. On drying the leaves were found not to have consolidated to a single unit.

The fungi developing on the paper samples were accompanied by numerous bacteria whose generic composition is presented in Table 6. The bacteria *Pseudomonas fluorescens*, *Streptococcus* sp. and *Bacillus subtilis* were accompanied, from the fifth month, by *Sporocytophaga myxococoides*.

A comparison of the succession of microorganisms on samples of XVIIth century paper with that on XIXth century paper reveals considerable similarity in the composition of the populations though the intensity of their growth was much poorer on the XIXth century paper.

The total number of bacteria in paper samples from the XIXth century book after 6 months of growth was about one eleventh of that in samples from the XVIIth century book (Tables 2 and 7). The percentage of capsulated bacteria in the whole bacterial population increased with the moistening of the samples (Table 7) but their percentage was only half that in the paper samples from the XVIIth century book (Tables 2 and 7).

The isolated microorganisms did not produce active cellulases and the protein content in the control and moistened samples was similar (Table 8). It can be assumed that the increase in microbial protein was compensated for by reduced casein glue content. Moreover, the alpha-cellulose content in the control and contaminated samples did not show any significant difference and the copper index was only slightly increased (Table 8).

Discussion

The decomposition of paper in ancient books depends to a major extent on the enzymatic activity of microorganisms. The intensity of their growth and resultant damage is also determined by the fibre content of the paper and the type of its gluing. Two of the books chosen for these studies differed greatly in these characteristics. This profoundly affected the degree of composition of paper and the consolidation of the book into a block. Most of the genera of fungi isolated from the fragments of XIXth and XIXth century books were identical with the fungi isolated from destroyed ancient books and studied earlier (Leznicka, in press). The fungi concerned were cellulytic organisms with varied capacity for the degradation of cellulose.

Fungi isolated from samples of the XVIIth century book became dominant in a definite order (Table 1). The first fungi to appear utilised first of all the gluten glue used in the manufacture of the paper, e.g. *Verticillium* sp. This fungus is known to have poor cellulose degrading abilities (Gupta and Heale, 1970). This confirms the earlier studies of Rosa and Strzelczyk, (1979) who found the degradation of cellulose to be inhibited to a certain extent in the presence of glue in the paper. The next set of fungi to appear on the samples had a full set of cellulytic enzymes (*Chaetomium* sp., *Trichoderma viride*, and *Penicillium* sp.) The genera to appear last were adapted to the utilisation of the products of cellulose degradation (*Rhizopus* sp. and *Cladosporium* sp.) The domination by *Chaetomium* sp. and *Trichoderma viride* was probably connected with the release by these
fungi of antibiotics such as chaetomin and viridin (Kadis et al., 1971), which in our experiment was manifested by poorer growth of Verticillium and relegation to the role of 'accompanying fungi' of other species. Most of the strains of fungi isolated from fragments of the XVIIth century book also occur in biological slime in cellulose mills. Firpi and Mazzucchetti (1963) isolated the following strains from slime: Fusarium, Penicillium, Trichoderma, Chaetomium, Cladosporium and Aspergillus. Sanborn (1965) isolated from slime Trichoderma lignorum and Cladosporium herbarum; Kirchen and Sladen (1965) isolated Aspergillus niger. The similar composition of the fungal populations isolated by us, first from damaged books (Leznicka, in press) and later from consolidated books, with those isolated in paper mills is not accidental. This observation indicates that the fungi are characterized by active degradation of cellulose in paper and the formation of substances of a slimy nature.

The increase in the proportion of capsulated bacteria in the total bacterial count during the overgrowing of the samples and their consolidation (Table 2) is also not by chance. The bacteria isolated included the following capsulated strains: Bacillus, Pseudomonas, and Micrococcus (Table 1). The main mucus producing bacteria are Sporocytophaga and Cytophaga (Berg et al., 1972) and these were also isolated from our book fragments.

Most of the bacteria isolated by us were also found in paper mill slime. Firpi and Mazzucchetti (1963) isolated Cytophaga and Cellvibrio. Oppermann and Wolfson (1961) isolated Aerobacter, Chromobacterium, Clostridium, Flavobacterium, and Klebsiella as well as those which we isolated in our studies, namely: Proteus, Escherichia and Pseudomonas (Table 1). The bacteria carry fimbriae which are thought to play a role in adsorption to cellulose fibres.

The intensive growth of microorganisms on fragments of the XVIIth century book resulted in a greatly decreased degree of polymerization which was correlated with a very high increase in the copper index (Table 5). Studies on the thermal ageing of paper carried out in our laboratory indicate that a 6 percent drop in alpha-cellulose content is accompanied by complete destruction of paper (Rosa, 1980).

The great degradation of cellulose in the fragments was not fully reflected in the level of cellulolytic enzymes (Table 3). However, as mentioned, this phenomenon is connected with their adsorption to cellulose (Table 4). The adsorption of cellulases to cellulose was also observed by Pettersson et al., (1977).

The 6 month period of growth of microorganisms on the samples resulted in the same degree of consolidation and changes in the structure of paper as are usually met in ancient, moulded books.

The vigour with which microorganisms grew on samples from the XIXth century book was very poor.

This poor growth of the microorganisms despite their known cellulolytic activity was caused by the presence of colophony and certain amounts of lignin in the paper. In view of the above it can be concluded that the process of the consolidation of moistened fragments of books was brought about by intensive growth of fungi and bacteria. These microorganisms produce cellulolytic enzymes which cause the degradation of cellulose, produce slime, and overgrow the separate leaves of a book which may result in consolidation as described.

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<table>
<thead>
<tr>
<th>Period of growth in months</th>
<th>Generic composition of fungi</th>
<th>Generic composition of bacteria</th>
<th>Manifestations of microbial growth on paper</th>
</tr>
</thead>
<tbody>
<tr>
<td>II</td>
<td>Dominant: <em>Verticillium</em> sp., <em>Chaetomium</em> sp. Accompanying fungi: <em>Rhizopus</em> sp., <em>Trichoderma viride</em></td>
<td>Not studied</td>
<td>Growth of <em>Verticillium</em> sp. slightly suppressed by heavy growth of <em>Chaetomium</em> sp. However, <em>Chaetomium</em> stains the medium yellow-green.</td>
</tr>
<tr>
<td>III</td>
<td>Dominant: <em>Chaetomium</em> sp., <em>Trichoderma viride</em> Accompanying fungi: <em>Verticillium</em> sp., <em>Penicillium</em> sp., <em>Rhizopus</em> sp. Appearance of: <em>Cladosporium</em> sp. (Streptomyces)</td>
<td><em>Bacillus subtilis</em>, <em>Pseudomonas fluorescens</em>, <em>Escherichia coli</em>, <em>Streptococcus</em> sp., <em>Micrococcus</em> sp.</td>
<td>Visible very dark and numerous peridia of <em>Chaetomium</em> and intensive, nodular growth of <em>Trichoderma viride</em>. Much larger range of the yellow-green stains. Appearance of dark staining of the medium caused by growth of <em>Cladosporium</em> sp. The thickness of the moistened edge begins to grow smaller due to the glueing together of the separate leaves.</td>
</tr>
<tr>
<td>V</td>
<td>Dominant: <em>Chaetomium</em> sp., <em>Trichoderma viride</em> Accompanying fungi: <em>Verticillium</em> sp., <em>Penicillium</em> sp., <em>Cladosporium</em> sp., <em>Fusarium</em> sp.</td>
<td><em>Bacillus subtilis</em>, <em>Pseudomonas fluorescens</em>, <em>Escherichia coli</em>, <em>Streptococcus</em> sp., <em>Micrococcus</em> sp., <em>Sporocytophaga</em> sp.</td>
<td>At this stage no significant changes in growth of the fungi. Increasing range of fungal and bacterial staining. Strongly visible process of consolidation of the leaves. Losses and holes in the paper.</td>
</tr>
<tr>
<td>VI</td>
<td>Dominant: <em>Chaetomium</em> sp., <em>Trichoderma viride</em> Accompanying fungi: <em>Verticillium</em> sp., <em>Penicillium</em> sp., <em>Cladosporium</em> sp., <em>Fusarium</em> sp.</td>
<td><em>Bacillus subtilis</em>, <em>Pseudomonas fluorescens</em>, <em>Escherichia coli</em>, <em>Streptococcus</em> sp., <em>Micrococcus</em> sp., <em>Sporocytophaga</em> sp. Appearance of: <em>Cytophaga</em> sp.</td>
<td>Whole edges covered with <em>Chaetomium</em> with very numerous peridia. Very wide range of varicoloured stains: yellow-green, olive-green, black and red. Far wider range of orange and yellow mucose stains with bacterial origin than in the 4th and 5th months. Moisten edges much thinner. On drying their thickness dropped from 35 mm to 15 mm. The thin edges gave rise to compact consolidated block of the book.</td>
</tr>
</tbody>
</table>
Table 2

Number of encapsulated bacteria in relation to the total number of bacteria on samples of XVIIth century book

<table>
<thead>
<tr>
<th>Time of experiment in months</th>
<th>Total number of bacteria in thousands per 100 mg dry weight of paper samples</th>
<th>Percentage of encapsulated bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>III</td>
<td>1238 ± 100</td>
<td>43.5 ± 2.5</td>
</tr>
<tr>
<td>IV</td>
<td>5944 ± 70</td>
<td>79.4 ± 5.3</td>
</tr>
<tr>
<td>V</td>
<td>6711 ± 250</td>
<td>87.2 ± 6.0</td>
</tr>
<tr>
<td>VI</td>
<td>6425 ± 110</td>
<td>79.6 ± 4.2</td>
</tr>
</tbody>
</table>

Each result is the mean (± standard deviation) of 3 experiments.

Table 3

Cellulolytic activity of extracts of consolidated book samples (XVIIth century)

<table>
<thead>
<tr>
<th>Sample</th>
<th>Protein content in mg/g of dry weight paper sample</th>
<th>Exoglucanase activity</th>
<th>Endoglucanase activity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Total activity</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>units/min</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Specific activity</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>in units/min per mg</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>of protein</td>
</tr>
<tr>
<td>Not moistened (control)</td>
<td>5.10 ± 0.15</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Moistened (Consolidated)</td>
<td>6.90 ± 0.26</td>
<td>0.0</td>
<td>47.33 ± 0.15</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>6.83 ± 0.23</td>
</tr>
</tbody>
</table>

The results are the mean (± standard deviation) of 3 experiments.

Table 4

The influence of incorporation of XVIIth century paper or Whatman No.1 filter paper on the cellulolytic activity of *Penicillium notatum* supernatant.

<table>
<thead>
<tr>
<th>Supplements</th>
<th>Protein content in mg/50 ml supernatant</th>
<th>Exoglucanase activity</th>
<th>Endoglucanase activity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Total activity units/hour</td>
<td>Specific activity units/hour per mg of protein</td>
</tr>
<tr>
<td>Supernatant with 2.0 g of XVIIth century paper</td>
<td>1.55 ± 0.07</td>
<td>447.60 ± 37.23</td>
<td>287.65 ± 10.85</td>
</tr>
<tr>
<td>Supernatant with 2.0 g of Whatman No.1 filter paper</td>
<td>1.55 ± 0.07</td>
<td>580.20 ± 25.00</td>
<td>373.32 ± 12.80</td>
</tr>
<tr>
<td>Supernatant only</td>
<td>1.55 ± 0.07</td>
<td>918.75 ± 37.23</td>
<td>593.90 ± 51.20</td>
</tr>
</tbody>
</table>

The results are the mean (± standard deviation) of 3 experiments.
<table>
<thead>
<tr>
<th>Period of growth in months</th>
<th>Generic composition of fungi</th>
<th>Generic composition of bacteria</th>
<th>Manifestations of microbial growth on paper</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Dominant: <em>Penicillium</em> sp.</td>
<td>not investigated</td>
<td>Trace growth of named fungi in the form of single, point colonies.</td>
</tr>
<tr>
<td></td>
<td>Accompanying fungi:</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Trichoderma</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>Dominant: <em>Penicillium</em> sp.</td>
<td>not investigated</td>
<td>Presence of vari-coloured stains on the edges and surface of the moistened paper: red stains caused by growth of <em>Penicillium</em>, yellow-green caused by <em>Chaetomium</em> and <em>Trichoderma</em>. Intensity of growth of the mentioned fungi - very low.</td>
</tr>
<tr>
<td></td>
<td>Accompanying fungi:</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Trichoderma</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Chaetomium</em> cinnabarinux</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Cladosporium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>III</td>
<td>Dominants: <em>Streptococcus</em> sp. <em>Pseudomonas fluorescens Bacillus subtilis</em></td>
<td>Range of stains slightly greater than after 2 months of growth.</td>
<td>Intensity of growth of the named fungi - very low. Visible surface, point colonies. Range of stains slightly greater than after 2 months of growth.</td>
</tr>
<tr>
<td></td>
<td><em>Penicillium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Verticillium cinnabarinux</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Chaetomium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Cladosporium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IV</td>
<td>Dominants: <em>Streptococcus</em> sp. <em>Pseudomonas fluorescens Bacillus subtilis</em></td>
<td>Range of stains slightly greater than after 3 months of growth. Intensiveness of growth of the individual fungal colonies somewhat better but still very poor.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Penicillium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Verticillium cinnabarinux</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Trichoderma</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Chaetomium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Cladosporium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>V</td>
<td>Dominants: <em>Streptococcus</em> sp. <em>Pseudomonas fluorescens Bacillus subtilis Appearance of: Sporocytophaga myxococcoides</em></td>
<td>In the last two stages slightly better growth of the fungi than in the earlier stages. However, the analysed colonies still meagre and delicate. Orange-coloured mucose stains caused by the presence of <em>Sporocytophaga</em>. Range of fungal stains greater than in the earlier months though only on the surface and edges where moistening of the paper was heavier. Leaves of the book from the side of the moistened edge loose and not glued together. On drying no consolidation of the leaves into a single unit observed.</td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Penicillium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Chaetomium</em> sp.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Verticillium cinnabarinux</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td><em>Cladosporium</em> sp.</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Table 5

Alpha-cellulose content and copper index in XVIIth century control and consolidated paper examples

<table>
<thead>
<tr>
<th>Sample</th>
<th>Alpha-cellulose content %</th>
<th>Copper index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not moistened (control)</td>
<td>86.28 ± 1.80</td>
<td>1.463 ± 0.050</td>
</tr>
<tr>
<td>Moistened (consolidated)</td>
<td>77.38 ± 3.83</td>
<td>2.544 ± 0.050</td>
</tr>
</tbody>
</table>

The results are the mean (± standard deviation) of 5 experiments.

### Table 7

Number of encapsulated bacteria in relation to the total number of bacteria on samples of XIXth century book

<table>
<thead>
<tr>
<th>Time of experiment in months</th>
<th>Total number of bacteria in thousands per 100 mg dry weight of paper sample</th>
<th>Percentage of encapsulated bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>III</td>
<td>267 ± 25</td>
<td>21.3 ± 2.0</td>
</tr>
<tr>
<td>IV</td>
<td>535 ± 10</td>
<td>33.3 ± 3.3</td>
</tr>
<tr>
<td>V</td>
<td>547 ± 18</td>
<td>37.0 ± 2.8</td>
</tr>
<tr>
<td>VI</td>
<td>563 ± 14</td>
<td>40.0 ± 3.4</td>
</tr>
</tbody>
</table>

Each result is the mean (± standard deviation) of 3 experiments.

### Table 8


Influence of the microbial infection on changes in alpha-cellulose content and copper index.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Protein content in mg/g of dry weight</th>
<th>Exoglucanase activity paper sample</th>
<th>Endoglucanase activity</th>
<th>Alpha-cellulose content %</th>
<th>Copper index</th>
</tr>
</thead>
<tbody>
<tr>
<td>Not moistened (control)</td>
<td>2.70 ± 0.10</td>
<td>0.0</td>
<td>0.0</td>
<td>69.50 ± 1.70</td>
<td>1.883 ± 0.050</td>
</tr>
<tr>
<td>Moistened (slightly overgrown)</td>
<td>2.70 ± 0.10</td>
<td>0.0</td>
<td>0.0</td>
<td>69.66 ± 1.45</td>
<td>1.908 ± 0.134</td>
</tr>
</tbody>
</table>

The results are the mean (± standard deviation) of 5 experiments.
BOOK REVIEWS

PROCEEDINGS OF THE THIRD NATIONAL WEEDS CONFERENCE OF SOUTH AFRICA 1979

Balkema, P.O.Box 1675 Rotterdam US $34.50 or £12.50.

This 214 page volume, edited by S. Noser and A.L.P. Cairns is based on 26 papers presented at a conference held in Pretoria, August 1979. The text is produced, somewhat reduced, from type script, but well bound and a preface and species/herbicide index have been included.

The papers are predominantly of agricultural interest but there is more emphasis on ecology and less on herbicide use than in most such conferences. An introductory paper on "Herbicides: synthesis to reality" emphasises the increasing cost and difficulty of introducing new herbicidal products. The range of active compounds is no longer expanding rapidly and any new materials, to satisfy environmental safety requirements are likely to have relatively short residual life in the soil. Hence the control of weeds in industrial situations will continue to depend mainly on those compounds already available.

Almost half the papers are concerned with the menace of introduced weeds - their ecology, distribution and control. In the most relevant of these (for this journal) "Woody plant invaders of the central Transvaal" there is a detailed survey of the introduced trees and shrubs occurring in the area embracing Johannesburg and Pretoria, many of these originating as escapes from gardens. There is no mention of damage to roads, but Salix babylonica and Populus alba are particularly serious invaders of water courses. Another paper deals with the problem of an introduced Myriophyllum sp in water bodies proving a great deal more aggressive than the endemic species. And another concerns an introduced ornamental Sesbania sp which is now flourishing in swamps and along river banks.

Biological control is often effective on introduced weed species and the strength of this approach in South Africa is well demonstrated in the papers on Sesbania sp, Acacia spp, Hakea spp and Opuntia. There are also several excellent papers dealing with techniques of surveying distribution of introduced weeds, as the basis for control programmes. " Bramble control in Natal" discusses alternative methods of control involving mowing and the herbicides glyphosate, 2,4,5-T and picloram.

Some of the concern over introduced species is based on fears for the unique native vegetation of South Africa but this volume illustrates how wide a range of economic problems can arise when exotic plants are introduced whether deliberately or otherwise.

C. Parker

DRYING AND STORAGE OF AGRICULTURAL CROPS

Carl W. Hall (and associate authors)


This book is an expanded version of the principal author's earlier books, Drying Farm Crops and Drying Cereal Grains. Its interest from the point of view of the specialist in biodeterioration lies in the fact that, in practice, the principal measure of control of the deterioration of farm crops is drying.

In the first chapter, on the importance of drying, the biological effects of drying, or failure to dry, are fully acknowledged and to some extent documented. At many other points in the text the effects of water activity (though this term is not used) on growth of molds, mites and insects are referred to.

The text is a mixture of theory and rule of thumb practice supported by extensive tables and diagrams which make it a useful source for relevant data. However, some of the data are old, many dating from the 1940's, and in some cases are no longer acceptable as correct. For example, Lela Barton's data on relative humidity equilibrium moisture contents of a number of seeds are almost certainly invalid because of inadequate equilibration times. Similar criticisms can be made of some of the other data assembled but provided that older sources are evaluated critically, this book will be valuable on the shelves for reference to methods, principles and facts.

Four chapters, constituting about one third of the book, are contributed by other authors. Of these, the longest on moisture control and storage for vegetable crops by Denny C. Davis is especially useful for those concerned with the deterioration of these crops. However, it duplicates some information on fan and duct design which is already covered in the earlier parts of the book. This chapter might be publishable as a short book on its own.

This is a useful teaching book. Each chapter concludes with some questions to test students' understanding. They are not examination questions for in most cases they require reference to the book, but they will be useful to teachers. It will be important that students are guided through the data and cautioned to evaluate them critically; this in itself could be a useful teaching exercise.

All units are given in American and S.I. units, but they are rounded to the former, perhaps an inevitable transition stage to the newer system. Useful conversions are given in the appendix.

T.A. Oxley
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</tr>
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<td>£27 or $ 68</td>
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