INTERNATIONAL
BIODETERIORATION
BULLETIN

A QUARTERLY JOURNAL OF BIODETERIORATION

BIODETERIORATION INFORMATION CENTRE
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Biodeterioration Society Newsletter

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Book Reviews
Editor-in-Chief of Biodeterioration Centre Journals
Dr. H.O.W. Eggins.

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 easily 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: 354–368

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 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:

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RENTOKIL LIMITED, East Grinstead, West Sussex, England.

Short papers invited for the Summer meeting

The Annual Summer Meeting of the Society will be held at Preston Polytechnic, Preston, Lancashire, on 10th and 11th July. The theme of the principal symposium will be

“Biodeterioration of Polymeric Materials”

Offers of papers on this subject are invited.

As usual there will, in addition, be a session devoted to short papers on a wide variety of topics which need not be related to the main theme. The Society cordially invites such papers. This is an occasion for presentation of short pieces of work, perhaps unfinished. It has frequently been the opportunity for a student’s first public presentation and discussion. Also, as short abstracts of all papers are subsequently published in this Newsletter (in the International Biodeterioration Bulletin) it provides an opportunity to bring the interests of a particular Department, or the work of a group or of a single student to world wide attention.

Offers of papers in either the Symposium or the short paper session should be sent to:

Mr. Alleyn Barr,
Catomance Ltd.,
88-96, Bridge Street,
Welwyn Garden City, Herts.
AL7 1JW

Winter meeting 1980

The winter meeting will be held at the Princes Risborough Laboratory of the Building Research Establishment on Thursday 11th December. It will take the form of a one day symposium on:

“Effects of Toxic Chemicals on Microbial Ecology”

The meeting is being organized by Dr. R. Neil Smith of the Department of Biological Sciences, Hatfield Polytechnic, P.O. Box 109 Hatfield Herts. AL10 9AB, to whom offers of papers should be sent.

For local arrangements apply to Mrs. Wendy Worley, Biodeterioration Section, Princes Risborough Laboratory, Building Research Establishment, Princes Risborough, Aylesbury, Bucks. HP17 9PX

New members of Council

The Society is glad to welcome the following members on to its Council:

Dr. Rod Eaton of Portsmouth Polytechnic.
Mr. Graham Lloyd of Sterling Industrial Ltd.
Dr. H.O.W. Eggins of the Biodeterioration Centre, University of Aston in Birmingham.
Dr. Eggins is, of course, founder and past President of the Society returning to the Council after a period out of office.

In addition, Dr. R. Norrish will attend meetings of the Council as an observer from the Health and Safety Executive.

Professor T.A. Oxley of the Biodeterioration Centre, also a past President, has been co-opted to the Council as Editor.

Membership

Membership of the Society remains a small fraction of the potential. Part of this shortfall has been attributed to the difficulties which potential members appear to have in obtaining membership application forms and finding the correct address to which to send them. We will therefore devote one page of this and each subsequent issue of the Newsletter to a membership application form which will carry the subscription rates and the name and address of the treasurer.

This must surely be one of the cheapest and best value annual subscriptions in the scientific world. If you can persuade your colleagues to join it will be possible to avoid increases.

M.Sc. Degree in biodeterioration

Portsmouth Polytechnic will again run its highly respected M.Sc. course in Biodeterioration of Materials from September 1980 to September 1981. Entry qualification is normally an appropriate degree in a biological science subject, or an equivalent qualification. Applications will also be considered from graduate members of the Institute of Biology or holders of Higher National Diploma in a biological subject.

The syllabus is listed under the following six headings:


The course is supervised by Dr. E.B. Gareth Jones and is taught by twelve members of staff plus specialist lecturers.

Fees are £780 gross for British students or £2200 for overseas students. A few S.R.C. studentship grants are available to support exceptionally well qualified candidates.

Increased facilities for testing new wood preservatives

The British Wood Preserving Association has agreed to sponsor an addition to the staff available at the Princes Risborough Laboratory of the Building Research Establishment in order to meet the growing demand for tests which the Laboratory would otherwise be unable to meet. The demand arises from firms engaged in technical development of formulations or seeking evidence for official approval schemes in various countries. Part of the demand arises from the move to new biocides to replace those which are now less acceptable in the environment. The work will be conducted under the supervision of experienced staff at the Laboratory and the results will carry the official status of Building Research Establishment tests. Results of tests will be strictly confidential to the applicant firm and materials will be handled under code. Applications for tests should be made to the head of the Biodeterioration Section of the Laboratory, Dr. A.F. Bravery, Building Research Establishment, Princes Risborough Laboratory, Princes...
Risborough, Aylesbury, Bucks. HP17 9PX. Fees will be collected by the British Wood Preserving Association whose secretary is able to give informal guidance on the likely cost. The B.W.P.A. telephone number is 01 837 8217

Abstract of a paper presented at the Symposium on "Biocides for the Preservation of Materials" held at Hatfield Polytechnic on 18th. December 1979
(This abstract was received too late for inclusion in the last number of the Newsletter)

Title: New Organo-metallic Compounds for use in Wood Preservation.

Author: C.R. Sparks

Address: Cuprinol Ltd., Adderwell, Frome, Somerset.

ABSTRACT

Copper and zinc naphthenate have been widely used in wood preservation for over 70 years. However, the availability of naphthenic acids is becoming restricted and consequently they are increasingly more expensive. One area of research conducted by Cuprinol into new biocides has centered around examination of alternatives to naphthenic acids.

Synthetic acids produced industrially either by the OXO reaction or the Koch process are amongst materials examined. Certain of these acids have been shown to be extremely promising wood preservatives when used as either the copper or zinc salt of the acid, as shown by both laboratory and field test results. In addition the toxicological evidence also suggests that these compounds, like the naphthenates will be environmentally acceptable.
**FORTHCOMING CONFERENCES, MEETINGS AND COURSES**

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| 31 May 1980        | Cellulose Decomposition  
Discussion meeting on the use of Cotton Textile Strips for Measuring Cellulose Decomposition  
(Joint meeting, Biodeterioration Society and British Ecological Society). | Birmingham ENGLAND | Dr. D. Walton,  
British Antarctic Survey,  
Madingley Road,  
CAMBRIDGE, CB3 OBT.  
Phone: 022361188 ext. 255. |
| June 1980          | Marine Corrosion and Fouling  
5th International Congress on Marine Corrosion and Fouling | Barcelona SPAIN | Secretaria del Congreso,  
Revista "CP Corrosion y Proteccion"  
Calle Londres 17  
MADRID-28, Spain. |
| 9 July 1980        | Toxic Chemicals  
One day course on "Toxic Chemicals – Changing needs in the Recognition & Avoidance of Hazards"  
Fee £36 (includes meals) | London ENGLAND | Netta Swallow,  
Short Course Unit,  
Polytechnic of Central London,  
309, Regent Street,  
LONDON W1R SAL.  
Phone: 01 580 2020 ext. 220 |
| 28-30 July 1980    | Controlled Release of Bioactive Materials  
7th International Symposium on Controlled Release of Bioactive Materials | Ft. Lauderdale, FLORIDA | Dr. Danny H. Lewis,  
Southern Research Institute,  
2000 Ninth Avenue South,  
Birmingham, Ala. 35205,  
U.S.A. |
| 31 August – 6 Sept. 1980 | Applied Microbiology  
6th International Conference on Global Impacts of Applied Microbiology.  
Sponsored by UNESCO, UNEP and the Government of Nigeria. | Lagos NIGERIA | Prof. O. Ogunbi,  
Dept. of Microbiology & Parasitology,  
Lagos University Teaching Hospital,  
P.M. Bag 12003,  
Lagos, NIGERIA. |
| 7-12 Sept. 1980    | Macromolecules  
IUPAC International Symposium on Macromolecules | Florence ITALY | Fondazione Giovanni Lorenzini  
Via Monte Napoletano 23  
20121 MILAN Italy.  
Phone: (02) 702.267  
or 783 868 |
| 7-October 1980     | Control of Insects and Rot in Buildings  
Three Day Course  
Fee: £29.75 incl. VAT | Princes Risborough (Near Aylesbury) ENGLAND | Miss Diane Poole,  
Building Research Establishment,  
Princes Risborough,  
AYLESBURY, Bucks.  
HP17 9PX.  
Phone: 084 44 3101. |
| 4-6 November 1980  | Rubber  
International Rubber Conference and Exhibition 'Rubbercon '81' | Harrogate, ENGLAND. | Richard H. Craven,  
Plastics and Rubber Institute,  
11, Hopton Place,  
London SW1W 0HL. |
| 8-12 June 1981     | Biodeterioration  
5th International Biodeterioration Symposium | Aberdeen SCOTLAND. | Dr. J.M. Shewan,  
79, Duthie Terrace,  
ABERDEEN Scotland.  
Phone: (0224) 37565. |
| 6-12 Sept. 1981    |                                                                                             |          |                                              |

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*Note: Contact information provided for organizers and organizers. Phone numbers and addresses are for reference only.*
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You will be asked to undertake that your subscription is for your own personal use and not for the Institution with which you are affiliated. Do not send money for a subscription to a journal with your Society subscription. You will be invoiced by the Biodeterioration Centre of the University of Aston which publishes the journals.
A REVIEW OF STRENGTH TESTING AS A MEASURE OF BIODETERIORATION OF WOOD AND WOOD BASED MATERIALS

Kay Hardie

Summary

Assessment of different strength properties of materials provides valuable information on their ability to withstand stress when in service. Wood possesses strength properties which may be reduced by biological agents. The patterns of deterioration by fungi and bacteria are discussed and methods of strength testing are described and reference to published literature indicates that, in the early stages of decay, loss of strength is a more sensitive criterion of deterioration than loss of weight.

Introduction

The range of materials colonised by microorganisms is wide and the importance of biodeterioration to industry is unquestionable. The attack by biological agents results in economic loss, either by spoiling the material in its practical use. In order to understand why strength losses occur it is necessary to know the patterns of attack by the microorganisms. The organisms may attack a minor constituent of the material, such as plastisiers or fillers in plastics, or adhesives in plywoods and so destroy the integrity of the material. Alternatively, they may degrade the major constituents such as cellulose in wood, paper or cotton textile, and hence reduce the material’s inherent strength.

In many cases where biodeterioration causes massive economic losses, the materials under attack are engineered by man and as such their components or ratio of components are controlled as in paper, plastics and chipboards. Due to this, large numbers of replicate samples, having the same strength properties, can be produced and measurement of strength loss is particularly valuable. One natural material which has a wide variety of uses and has yet to be replaced by a manmade substitute is

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1 Biodegradation Centre, University of Aston, St. Peter’s College, College Road, Salliley, Birmingham B8 3TE
Present address: Dept. of Forestry, University of Oxford, South Parks Road, Oxford OX1 3RB
(Received, November 1979)
wood. Unfortunately, it is a complex, heterogeneous material composed of elements which are easily biodeteriorated. Its complexity and variability make it difficult to produce large numbers of replicate samples for strength testing. However, because wood possesses, to some extent, all the types of strength found separately in other materials and because biodeterioration of wood is brought about by groups of organisms which affect the various strength properties in different ways, wood is used as the prime example in this review although much of the information is relevant to other materials.

Wood Structure

Wood consists mainly of components assembled from sugar or monosaccharide molecules such as cellulose, hemicellulose and starch. The basic physical properties of wood can be attributed to its principal components, the longitudinal cellular elements, the tracheids or fibres. It is these elements which are colonised and degraded by microorganisms. Figure 1 is a diagrammatic representation of a typical softwood tracheid showing the several wall layers and the orientation of the cellulose microfibrils. The primary cell wall consists of loose microfibrils which are irregularly orientated in a predominantly shallow spiral at about 60° to the vertical axis of the cell. The secondary wall is produced once the primary wall has attained its ultimate dimensions and is formed in three stages. The first secondary wall, the \( S_1 \) layer, is thin with a shallow microfibril spiral. The \( S_1 \) layer sometimes consists of two or more lamellae spiralling in opposite directions and is morphologically and structurally intermediate between the primary and \( S_2 \) layers. The \( S_2 \) layer consists of very regularly and closely packed microfibrils at a very steep spiral angle, only 10-20° relative to the longitudinal axis. The \( S_3 \) layer consists of a thin layer of microfibrils orientated in a shallow spiral. These cell walls account for the cellulose which comprises 60% of the wood substance. The \( S_2 \) layer is always dominant and it is therefore not surprising that the longitudinal orientation of microfibrils within this layer accounts for many of the basic physical properties of wood.

Types of Decay

Wood is colonised by a wide range of microorganisms which may be classified by their patterns of attack.

Blue-stain: This type of deterioration is caused by microfungi which utilise the wood cell contents for their nutrition. Hyphae of blue-stain fungi lie in the lumen of the cell on the warty or \( S_1 \) layer. No degradation of the cell wall occurs in association with these hyphae. They traverse across cells via pits in the cell wall, or occasionally by direct penetration of the wall brought about by both a localised enzyme action and mechanical pressure. In general they cause little or no structural damage to the wood but they do produce pigments as a product of their metabolism which stain wood and reduce its economic value. This type of biodeterioration cannot be quantified by strength testing.

Soft-rot: Soft-rot is caused by fungi belonging to the ascomycetes and fungi imperfecti. They generally invade the wood through the rays (radially orientated bands of thin walled cells) or vessels from where they grow into the lumina of the adjacent fibres or tracheids. In hardwoods the warty layer and \( S_1 \) layer are often degraded and troughs under the hyphae are seen. The hyphae then attack the \( S_3 \) layer. In softwoods, the \( S_3 \) layer remains undegraded and small penetration hyphae are produced which grow through the \( S_3 \) layer. The penetration hyphae can grow transversely through several tracheids, regaining full hyphal width in each lumen. At some stage in their growth, when they reach an \( S_2 \) layer, either prior to or more frequently after, the penetration of the middle lamella a T shaped branching occurs. The branches orientate themselves to run approximately parallel to the cellulose microfibrils. The hyphae follow the fibrillar pattern of the cell and
produce large cavities with tapered ends due to the action of their ecto-enzymes. Extensive cavity formation in the $S_2$ layer reduces the strength properties of the timber.

**Brown rot:** Brown rots are caused by basidiomycete species which possess enzymes to break down the polysaccharides of the cell wall but leave the lignin matrix undigested. Hyphae penetrate through the rays and spread into the tracheids. They can penetrate through the cell walls but generally the attack starts from the cell lumen where hyphae grow on the cell wall surface. Enzyme secretion by the fungus gives rise to etching of the wall surface resulting in loss of the warty layer and $S_3$ layer. This type of decay progresses through the $S_2$ layer and results in a rapid decrease in the degree of polymerisation of the cellulose and hence a general reduction in the strength properties of wood.

**White rot:** White rot is also caused by basidiomycetes but differs from brown rot because a successive decomposition of the cell wall components occurs. They preferentially metabolise lignin and hemicelluloses and the cellulose fraction is degraded at a later stage. The hyphae grow on the surface of the warty layer and $S_2$ layer. Each hypha produces a localized enzyme action which results in an erosion trough under and around the hypha. A simultaneous dissolution of the $S_2$ and $S_3$ layers occurs and the particular microfibrillar orientation of successive layers is exposed. Wood decayed by white rot fungi shows an early reaction to attack only in the strength property known as 'toughness' because these fungi cause a general reduction in the average length of cellulose chains and do not produce such rapid depolymerisation of cellulose as the brown rot fungi.

**Bacterial attack:** Pectinolytic bacteria may be found degrading the pit membranes of cell walls, and bacteria capable of cellulolysis can cause progressive decay of the cell wall from the $S_3$ layer.

### Strength Properties

The patterns of decay which cause structural damage to the cell result in some loss of physical strength of the timber in addition to a loss in weight. In order to assess the levels of strength loss it is first necessary to distinguish the various strength properties that a material may possess. There are three basic stresses which a material may be subjected to:

1. **Compression stress** — a reduction in material volume.
2. **Tensile stress** — an increase in material volume.
3. **Shear stress** — the movement of one portion of the material with respect to another (usually referred to in a direction parallel to the grain).

The resistance of the material to these stresses is known as its strength and may be measured in a variety of ways.
Side-compression tests, applying the load at right angles to the grain, provide useful observations on the performance of timber used in situations such as railway sleepers.

Tensile testing is widely used for determining the loss in strength of textiles but a number of problems exist in testing wood samples. If rectangular strips of wood are used and gripped at each end between vice-like jaws, as is done with textile strips, then it is difficult to avoid crushing the specimens. This results in the specimen breaking at the ends where it has been weakened. Alternatively, shaped specimens may be used which can be held in special jaws without crushing (Fig. 3). This is not a complete answer to the problem since it can only be used for ‘across the grain’ testing. If specimens are cut with the grain running longitudinally then they tend to fail in shearing strength long before the tensile strength is put under duress.

Figure 3. Tension testing across the grain using a shaped specimen. (After Henderson, 1944).

Shearing strength of materials is occasionally tested and with wood there will obviously be greater strength across the grain than with the grain. It is easier to slide the wood fibres along one another in the direction indicated in figure 4 than the shear across them if the sample was turned through 90°.

Bending strength is another commonly measured criterion in physical testing and is a combination of compression, tensile and shearing stresses. This strength is tested on materials such as plastics and timber which are to be utilised in the form of beams. In these tests the sample is usually supported at its two ends and loaded, at a steady rate, in the centre of the span resulting in a bending or flexing action. Bending places the convex surface of a sample under tension and the concave surface under compression. These two opposite stresses diminish between the convex and concave faces of the beam and the centre remains virtually unstressed. Shearing stresses are set up between the various imaginary lamina that make up the specimen. Three-point loading of the beam concentrates the stress at the centre of the span while four-point loading distributes the stress more evenly along the beam length.

Strength in general is approximately proportional to the density but it may vary according to fibre length, particularly in relation to bending strength. Wood from trees which suffer a short growing season, such as those in polar regions, is often very dense but fibre length is comparatively short and its bending strength suffers accordingly. Rapid growth, producing wide annual ring spacing, also causes lower bending strength apparently because the soft, low density, spring wood compresses, but this may be coupled with an increase in ability to withstand impact loads.

Impact bending strength is also known as ‘shock resistance’ or ‘toughness’ and impact tests may be used on materials subjected to sudden loads. It is a combined measurement of bending strength and plasticity of the material. Toughness can be estimated by measuring the amount of work done up to the point of maximum
load and may be determined by subjecting samples to the impact of a falling hammer. The hammer is raised higher for each successive blow. It is this strength property which is particularly affected by the white rot fungi.

Only one further measure of strength may be applied and that is a test of the property known as 'hardness'. This is the ability of a material to resist penetration and is usually measured by the force required for a standard metal sphere to penetrate to half its diameter. If timber is tested by this method, orientation of the sample is important as end-grain surfaces are harder than radial or tangential surfaces.

**Measurement of Strength Loss**

Strength loss determinations are carried out on materials both as a measure of the inherent properties of the material and as a measure of deterioration. The strength properties of test materials have frequently been found to be affected earlier in the decay process than measurable changes in weight. Liese and Pechmann (1959) reported a 50% decrease of impact bending strength of wood for a 5% reduction of weight. In the same year, Armstrong and Savory (1959) found that beech, for a 5% weight loss, showed a decrease of impact bending strength and static bending strength of 60% and 18% respectively. Cartwright and Findlay (1950) listed bending strength, compression, hardness and elasticity as the approximate order in which mechanical strengths decrease during decay of wood.

It appears therefore that the strength properties of timber are affected to a much greater extent in early microbial degradation than the weight of the wood. In view of this it is surprising that the assessment of strength as a criterion for microbial activity has been used so infrequently. The answer may lie in the difficulty of obtaining consistent results. Part of the problem in the past lay in the scarcity of machines for carrying out strength tests in the laboratory. Machines which were available were generally large, expensive and designed for use on large dimensional structural materials. Simple, inexpensive devices have been designed by various workers for laboratory use (Fig. 5). For example (Mills, Allsopp & Eggins, 1972) depended on the lowering of a bar onto paper or textile strips until they broke. Such devices suffer from the disadvantage of lack of precision in rate and direction of loading and strain.

In recent years more sophisticated machines have become commercially available, for example the Tensometer and the Instron laboratory scale tester. Either machine may be motor driven to apply strain to the samples at a steady rate and may be used to test a wide variety of strength properties on a range of materials by virtue of a number of detachable jaws. A range of spring beams and scales supplied with the machine allows the Tensometer to be used for measuring strengths of up to two tons. The force required to break a test sample can either be read directly from a mercury scale or a special attachment will produce a line trace on heat sensitive paper which gives a permanent record of the strength of a material. Alternatively, the more sophisticated Instron machine may be used to apply a load at a constant rate.

Since strength testing is destructive it is only possible to measure strength loss by reference, on a statistical basis, to suitable controls. In the case of man-made materials it is usually easy to obtain large numbers of control specimens with relatively small variation between them. Wood is more heterogeneous and in order to reduce the variations between controls, and between these and the undeteriorated test specimens, it is necessary to select the material carefully, avoiding all visible defects such as knots and ensuring, as far as possible, equal growth rates and grain parallel to one dimension of the specimens. This is not difficult but may be wasteful of material owing to the need to reject many potential specimens.

The strength of cellulose fibres is greatly affected by their moisture content. It is important, therefore, that all tests of controls and test specimens are conducted at the same moisture content. Solid wood is not greatly affected by moisture content and may be used structurally, or tested, in fully saturated or permanently submerged conditions, but the use of hardboard, even tempered
hardboard, in similar conditions would be inadvisable due to its great loss of strength with increased moisture content (Chan, 1979). Other cellulosic materials are also affected, e.g. paper. The hydrogen bonding which holds fibres together in paper is almost completely lost at saturation. For such materials it is usual to equilibrate all specimens to a common relative humidity and to conduct the test at the same humidity. This requirement may introduce a considerable delay because it can take several weeks for even a small specimen to adjust itself to a common water activity throughout.

Strength Loss versus Weight Loss

One of the main advantages with strength loss determinations is the rapidity with which results can be obtained in the laboratory. Mills, Allsopp and Eggins (1972) showed the rapid loss of strength in paper strips exposed to cultures of thermophilic fungi (Fig. 6). One to five days incubation was all that was required to produce strength losses of over 75%.

Walchli (1968) reported rapid loss of tensile strength in cotton cloth buried in soil for three, seven or fourteen days. In the same paper the results of soil burial tests on beech wood impregnated with preservative at various concentrations were recorded. Where no, or low, salt concentrations were used deterioration of the wood was extensive and bending strength loss was high after a twelve week incubation period (Fig. 7). Weight loss, on the other hand, was much lower, although, as salt concentration was increased and hence the extent of deterioration lowered, the strength and weight loss decreased to similar proportionate levels.

Bravery and Grant (1971) showed the rapid decrease in strength which accompanied an increase in decay in thin strips of pine sapwood. Strips were treated with different concentrations of a copper/chrome/arsenic type preservative and exposed to Coniophora cerebella for five weeks. Tensile strength decreased less with an increase in preservative concentration as would be expected with a successful treatment. The appearance of the wood strips after incubation and strength testing
Figure 8. The appearance of wood strips previously treated with concentrations of copper/chrome/arsenic after incubation with *Coniophora cerebella* for five weeks. The percentage figures indicated the concentration (w/v) of Tanalith CT 106. In each case the control strip (sterile) is on the right.

(Reproduced by permission of the Building Research Station, Princes Risborough Laboratory, from Bravery and Grant, 1971)

<table>
<thead>
<tr>
<th>0·0%</th>
<th>0·5%</th>
<th>1·0%</th>
<th>2·0%</th>
<th>5·0%</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image.png" alt="Strip image" /></td>
<td><img src="image.png" alt="Strip image" /></td>
<td><img src="image.png" alt="Strip image" /></td>
<td><img src="image.png" alt="Strip image" /></td>
<td><img src="image.png" alt="Strip image" /></td>
</tr>
</tbody>
</table>

Bravery and Lavers (1971) investigated strength and weight loss of miniature test beams of Baltic redwood after exposure to *Coniophora cerebella*. They observed that the greater the length of incubation the greater the decline in impact strength after forty-two days of incubation the impact strength loss was as high as 90%, at which time the weight loss had reached only 20%.

An earlier study of strength loss in wood was carried out by Armstrong and Savory (1959). They investigated the loss of impact and bending strength brought about by fungal decay in beech wood. The wood was exposed to the soft rot organisms, *Chaetomium globosum*, and an immediate reduction in impact strength ('toughness') was observed before any appreciable loss in dry weight was recorded (Fig. 9). Toughness continued to decrease rapidly until it was reduced to about 50%, at a time when the loss in dry weight was only 2%. The bending strength was affected less dramatically but the rate of reduction was fairly uniform, just beginning to show

ey to slow down at higher weight losses.

A strong and Savory (1959) carried out further experiments on impact strength and weight loss of beech with three different fungi: a white rot, a brown rot and a soft-rot organism. All three fungi caused a marked reduction in strength at very low weight losses. The brown rot caused the greater loss of impact strength for any given weight loss, while losses caused by the white rot and soft-rot were similar.

The relationship between bending strength loss and weight loss of beech, pine and lime sapwood decayed by *Chaetomium globosum* was also reported by the present author (Hardie, 1979). The results were similar to those of Armstrong and Savory (1959) and showed an initial rapid loss of strength at low weight loss during the early stages of decay. This was then followed by a gradual decrease in the rate of strength loss at higher weight losses.

Conclusion

In summary, strength loss appears to be a useful criterion for assessing decay by microorganism especially during initial decay where weight losses are low. The advantages of this criterion lie in the rapidity and magnitude with which it appears and also the information
Figure 9. Effect of Chaetomium globosum on bending strength and toughness of beech (Fagus sylvatica). Note that toughness, especially shows very great reduction before weight loss is appreciable. 
(Reproduced by permission of Building Research Establishment, Princes Risborough Laboratory, from Armstrong and Savory, 1959)

References

Armstrong F.H. and Savory J.G. (1959)  
The influence of fungal decay on the properties of timber.  
Holzforschung 13

Bravery A.F. and Grant C. (1971)  
Preliminary investigations into the use of a thin strip tensile strength test for the rapid evaluation of wood preservatives against basidiomycete fungi.  
International Biodeterioration Bulletin 7 (4)

Bravery A.F. and Lavers G.H. (1971)  
Strength properties of decayed softwood measured on miniature test beams.  
International Biodeterioration Bulletin 7 (3)

Cartwright K.St.G. and Findlay W.P.K. (1950)  
Decay of timber and its prevention, H.M.S.O. London

Chan W.W.L. (1979)  
Strength properties and structural use of tempered hardboard.  

Hardie K. (1979)  
The effects of nitrogenous compounds on decay of wood by soft-rot fungi.  

Henderson F.Y. (1944)  
Timber, its properties, pests and preservation. Crosby Lockwood and Son. Ltd. London.

Liese W. and Pechmann H. (1959)  
Experiments on the effect of soft-rot fungi on wood strength.  
Forstwiss. Zentralblatt 78

Some new developments in cellulosic material testing using perfusion techniques.  

Wälchli O. (1968)  
Biodeterioration test methodology.  

it imparts on the performance of a material when in service. The existence of several different strength properties enables a choice of test to be made according to the practical use of the material.

The major disadvantage of strength loss assessment lies in the initial outlay required to buy a strength testing machine for accurately measuring the losses. Other disadvantages may lie in the careful selection of samples required in order to reduce inter-sample variability to a level at which small strength losses become significant.
SURVEY OF THE MICROORGANISMS ASSOCIATED WITH CEREAL GRAINS AND THEIR MILLING FRACTIONS IN INDIA. II INDIGENOUS WHEAT

B.S. Mehrotra1 and Indra Kala Kunwar1

Summary

Samples from sixteen flour mills in different places in India were used to investigate the microorganisms associated with indigenous wheats and their milling fractions. Numbers of bacteria greatly exceeded those of fungi which in turn exceeded the numbers of actinomycetes. Compared with imported wheat, reported in a previous paper, the numbers of mesophilic bacteria were much higher, possibly as a result of improper drying and storage. As in imported wheat, some psychrophilic and thermophilic organisms were found, but unlike imported wheat, thermophilic bacteria were more numerous in the grain than in the milling fractions. Some acid producing bacteria were also found. The effect of normal washing procedure at the mills was generally slight but occasionally resulted in a very great increase in fungal infestation.

Contribucion al estudio de microorganismos asociados con granos de cereales y sus moliendas en la India.
II. Trigos Indigenas

Para la investigacion sobre microorganismos asociados con trigos indigenas y sus moliendas, se usaron dioses muestras de harina de diferentes sitios en la India.

El numero de bacterias excede mucho a los de hongos que a su vez excede al numero de actinomycetes. Al comparar trigo importado, publicado en una previa publicacion, el numero de las bacterias mesofilas fue mucho mas alto, posiblemente como resultado de la desecacion y almacenamiento inadecuados. Como en el trigo importado, se encuentra organismos psicrófilos y termofilos, pero a diferencia del trigo importado, las bacterias termofilas fueron mas numerosas en el grano que en las fracciones de molienda. Tambien se encontraron algunas bacterias productoras de acidez. El efecto del lavado normal sobre las moliendas era generalmente ligero pero ocasionalmente origina un gran incremento en la infeccion por hongos.

Introduction

Like any other substance with some moisture and organic matter, wheat and its milling fractions are subject to infestation by microorganisms during the process of storage and milling. Very large numbers of microorganisms are known to be present in association with wheat grains and their milling fractions (Bell, 1909; Morgenbahr, 1918; Geltinger, 1921; Kent-Jones and Amos, 1930; Gustafson and Parfitt, 1933; Soenen and Pinguair, 1937; James et al., 1946; Christensen, 1946; James and Smith, 1948; Thatcher et al., 1953; Poison and Guibot, 1956; Graves and Heseltine, 1966; Heseltine and Graves, 1966; Flannigan, 1970; Mehrotra and Basu, 1975).

A systematic investigation of the microflora of cereal grains and their milling fractions was undertaken in the senior author's laboratory during a period of six years. The first paper in this series (Mehrotra and Basu, 1975) reported the extent of microbial infestation with special reference to bacteria, actinomycetes, and fungi, of mostly imported wheats and their milling fractions collected from roller flour mills in India. The present paper reports similar studies on indigenous wheat and its milling fractions.

Material and Methods

The samples were collected from sixteen mills located at different regions in the country during the period

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Dr. Kunwar's name is now: Dr. (Mrs) Indra Kala Palni (Received October 1979)
November 1970 to March 1973. The methods employed for the screening of the samples were as given in the earlier paper (Mehrotra and Basu, 1975). Incubation was at 10°C (Psychrophiles), 25°C and 37°C (Mesophiles), and 55°C (Thermophiles). Identification of the fungi found is given in Mehrotra and Basu, 1976.

Results

The extent of microbial infestation in wheat grains and its milling fractions was found to be heavy. In both grain and milling fractions the maximum counts (number of colonies per gram) were those of bacteria followed by fungi and actinomycetes. The minimum, maximum, and average counts of bacteria, actinomycetes and fungi are given in Table 1. The maximum microbial counts reached in each case are also presented graphically in Figures 1–3.
### Table 1

Infestation of wheat grains and their milling fractions by bacteria, actinomycetes and fungi.
Numbers of colonies per gram of sample developing at 10°C, 25°C, 37°C and 55°C

<table>
<thead>
<tr>
<th>Samples</th>
<th>Organisms</th>
<th>10°C</th>
<th>25°C</th>
<th>37°C</th>
<th>55°C</th>
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<tr>
<td></td>
<td></td>
<td>Total count</td>
<td>Acid producers</td>
<td>Osmophilic producers</td>
<td>Total count</td>
</tr>
<tr>
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<td></td>
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<td>25°C</td>
<td>37°C</td>
<td>55°C</td>
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<td>2,068</td>
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<tr>
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<td>5,935</td>
<td></td>
<td>2,068</td>
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</tr>
<tr>
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<tr>
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NT = Not tested. Acid producers tested for bacteria only. Actinomycetes not tested for osmophilic growth.
Bacteria

Mesophilic and osmophilic bacteria were present in all the grain and milling fraction samples. Psychrophilic and thermophilic bacteria were found respectively in 75.8% and 82.7% of grain, and in 96.1% and 74.0% of the milling fraction samples. Acid producing bacteria were less frequently encountered: 20.6% of the grain and 26.6% of the milling fractions had mesophilic, and 3.4% of the grain and 4.5% of the milling fractions had thermophilic acid producers. From table 1 it is evident that the highest average counts of bacteria were found in bran samples followed by wheat grain, flour, finer flour, and semolina.

Fungi

Usually fungal population was much less than the bacterial. Mesophilic and osmophilic fungi were present in most of the grain and milling fraction samples. Psychrophilic fungi were present in 75.8% of the grain and 83% of the milling fraction samples. Thermophilic fungi were observed only in one grain and one flour sample. The average count of mesophilic fungi was highest in bran and that of psychrophilic and osmophilic fungi in semolina samples.

Actinomycetes

The counts and frequency of these organisms were much lower than those of bacteria and fungi. Psychrophilic actinomycetes were not observed in any of the samples. Mesophilic actinomycetes were observed in 31.0% of the grains and 36.2% of the milling fractions.

Although the frequency of occurrence of actinomycetes was lower in grain samples, their counts were higher in these than in the milling fractions. Thermophilic actinomycetes were rarely encountered.

The average mesophilic count of actinomycetes was found to be highest in the bran samples, followed by grain, finer flour, semolina and finally flour.

Counts of microorganisms in grains before washing and after the normal washing process in the mills were also recorded. These are summarised and presented, as total counts at all four temperatures, in figure 4.

Figure 4. The effect of a standard washing process at the mill. The numbers of bacteria and fungi found in wheat before and after the washing process at each of nine mills. Open histograms - before washing. Solid histograms - after washing. The numbers shown are the totals, per gram, at each of the four temperatures, 10°, 25°, 37° and 55° C.
Discussion

From this survey of the microflora of indigenous stored wheat and its milled products, collected from different parts of India during different seasons for nearly two and half years, certain facts have been brought to light:

1. The microflora associated with wheat grains was found to vary considerably, probably depending upon the climatic, particularly micro-climatic, conditions during their storage. This variation in the microflora was attributed mainly to the uncontrolled environmental conditions and storage practices in the country by Mehrotra and Basu (1975). Part of this microflora is in association with the grains right from the fields while they are maturing, a fact which is substantiated by the studies conducted in this laboratory (Mehrotra and Dwivedi, 1976). A sizable number contaminate the grains during and soon after harvest because of the natural contact with the soil and exposure to dust. Further increase in the microflora occurs during transportation and in storage.

2. That bacteria constitute the most numerous in the microflora of both wheat grains and its milling products in India is now well established; such has also been the observation of workers in other countries. The much speedier division, the smaller size of bacterial cells and dusty environment which is closely associated with storage conditions, are accounted by Mehrotra and Basu (1975) as the plausible reasons for their high percentage occurrence.

3. That actinomycetes occur infrequently and in smaller numbers in indigenous wheat grain and its milled products. This was also the finding of Mehrotra and Basu (1975). The actinomycetes are handicapped by their slow growth and greater specificity for the substrate.

4. That the fungal population, although considerably lower than the bacterial, is higher than that of the actinomycetes. This finding accords with that of Mehrotra and Basu (1975) who have attributed it to the incapability of the fungi in competing with bacteria in multiplication rate, capacity to colonise a substratum and more favourable pH of the substrate for bacterial than for fungal growth.

5. In the present study, as in that of Mehrotra and Basu (1975), microorganisms found associated could be classified into the following five broad categories:

- Psychrophiles
- Mesophiles
- Thermophiles
- Acid producing bacteria
- Osmophiles

These categories are now considered separately.

Psychrophiles

As generally psychrophiles are generally not found frequently in nature, their infrequent occurrence in the present study is not surprising. The frequency of occurrence (75.8%) and the counts (up to 86,580 per gram) of psychrophilic bacteria in wheat grains were lower than in the milling fractions (96.1% and up to 94,920 per gram), contrary to the findings of Graves et al., (1967) and Mehrotra and Basu (1975). Psychrophilic fungi were present in 75.8% of the grain and 83% of the fraction samples. Their population was greater in milling fractions than in the grains. This agrees with the finding of Mehrotra and Basu (1975) with imported wheat and its fractions. But even small numbers of these psychrophiles in foodstuffs may be of importance because such contaminated foodstuffs cannot be preserved by low temperatures. Psychrophilic actinomycetes in general are in nature, therefore their absence in these substrates is not surprising; a similar observation to that of Mehrotra and Basu (1975).

Mesophiles

The high frequency (100%) and numbers of mesophiles are to be expected as they constitute the major flora in nature, but the very high numbers observed (Table 1) may possibly be due to improper drying and storage of the indigenous grains and their milled products. Workers in other lands have also found substantial counts of mesophilic bacteria (Morgenthaler, 1918; Gellinger, 1921; Turley, 1922; Kent-Jones and Amos, 1930; Gustafson and Parfitt, 1933; Soenen and Pinquier, 1937; James et al., 1946; James and Smith, 1948; Thatcher et al., 1953; Poisson and Guilbot, 1956; Graves et al., 1967; Hesseltine, 1968). In the present work the counts of mesophilic actinomycetes in wheat grains were higher than in the milling fractions; Graves et al., (1967) and Mehrotra and Basu (1975), however, found higher counts in milling fractions than in the grains. Higher counts of fungi in flour samples (up to 18,500 per gram) than in wheat grains (up to 7,200 per gram) have also been reported by Morgenthaler (1918), Cohen and Christensen (1947), Christensen and Cohen (1950), Thatcher et al., (1953), Graves and Hesseltine (1966), Hesseltine (1968), and Mehrotra and Basu (1975). The greater number of fungi in milling fractions than in the grains could possibly be due to additional contamination in the mills, especially during the milling process.

Thermophiles

Thermophilic bacteria are less frequent in nature and were found in less numbers in the present substrate also. However, the frequency of occurrence was higher (82.7%) in wheat samples than in milling fractions (74.0%). Similar results were obtained by Graves et al., (1967) and Hesseltine (1968). Thermophilic actinomycetes were encountered rather infrequently and their numbers were greater in grains than in bran samples. This finding is in concurrence with that of Mehrotra and Basu (1975) on imported wheat and its fractions. Thermophilic fungi were even rarer which correlates
with their infrequent occurrence in nature. Flannigan (1970) and Mehrotra and Basu (1975) also found very few of these fungi in their samples.

**Acid Producing Bacteria**

Among the acid producing bacteria, mesophilic ones were more common and were observed both in the case of grains and their milled products. Their frequency was higher (26.6%) in milling fractions than in wheat grains (20.6%), but their counts were higher in the wheat grains which is contrary to the findings of Mehrotra and Basu (1975) who found higher counts of mesophilic acid producing bacteria in milling fractions. The presence of acid producers was also shown by James and Smith (1948).

Like other thermophiles, thermophilic acid producers were also few. They were less in number in wheat than in its fractions but their frequency of occurrence was almost similar in grains (3.4%) and milling fractions (4.5%). Thatcher et al. (1953) reported counts up to 32 in wheat flour and Graves et al. (1967) also found more thermophilic acid producers in flour samples (730 per gram) than in wheat (45 per gram). In contrast to this Mehrotra and Basu (1975) found the population (up to 500 per gram) in both cases, but observed that the frequency was higher in grains (20%) than in the milling fractions (10%).

**Osmophiles**

A sizable number of bacteria from wheat grains and milling fractions were found to be osmophilic which accords with the observations of Mehrotra and Basu (1975) in imported wheat and fractions. Osmophilic bacteria were isolated from most of the samples. Osmophilic fungi were present in all the samples. In both cases the numbers found were rather greater than those found by Mehrotra and Basu (1975). It was observed that the microflora in bran samples was generally higher than in other milling fraction samples and sometimes even greater than in the wheat samples. This agrees with the findings of Mehrotra and Basu (1975) who suggest that it may possibly be due to the different nutritive composition of bran from that of other milling fractions, also its greater surface area and greater amount of entrapped air.

**Washing Treatment at the Mills**

The effect of the washing treatment given to wheat at the mills is generally slight. In most instances it tended to reduce the bacterial count and to increase the fungal count, but there exceptions to both of these tendencies. In a few instances the fungal count is increased enormously by the washing treatment. Mehrotra and Basu (1975) also found one instance of an enormous increase in fungal count as a result of washing, and they found increases in both bacterial count and fungal count in nearly every case. They also point out that the object of the washing treatment given to wheat at mills is primarily to condition the wheat to a moisture content suitable for milling, not to clean it.

From the present investigations and those of Mehrotra and Basu (1975) it may be concluded that under the present storage conditions prevailing in the country, a substantial microflora is associated with the indigenous and imported wheat grains and the milling fractions made from them. Naturally these associated microorganisms pass on to their milled products and to food preparations made from them, and this microflora must influence in various ways the food value and appearance of the grains and finally the health of the consumer. Thorough drying and proper storage of wheat grains may lessen the extent of microbial infestation.

**References**


Gustafson, C.B., Parfitt, E.H. (1933)
Effect of numbers of bacteria on the development of rancidity in soft wheat flour
Cereal Chemistry 10: 233–238

Hesseltine, C.W. (1968)
Flour and wheat: research on their microbiological flora
Bakers’ Digest 42(3) 40–42, 66.

Hesseltine, C.W., Graves, R.R. (1966)
Microbiology of flours
Economic Botany 20(2): 156–168

James, N., Smith K.N. (1948)
Studies on the microflora of flour
Canadian Journal of Research 26C: 479–485

James, N., Wilson, Joyce, Stark, Egon (1946)
The microflora of stored wheat
Canadian Journal of Research 24C: 224–233

Kent-Jones, D.W., Amos, A.J. (1930)
Preliminary studies in the bacteriology of wheat and flour Analyst; 55: 248–268

Mehrotra, B.S., Basu, Monica. (1975)
Survey of the microorganisms associated with cereal grains and their milling fractions in India. Part 1. Imported wheat.

Mehrotra, B.S., Basu, Monica, (1976)
Fungi associated with wheat and milling fractions in India
Indian Journal of Mycology and Plant Pathology 6 (1): 43–50

Mehrotra, B.S., Dwivedi, P.K. (1977)
Subepidermal fungi of wheat grains in India

Morgenthaler, O. (1918)
Über die Mikroflora des normales und muffigen Getreides.
Landwirtschaft Jahrb. Schweiz 32: 548–570

Poisson, J., Guilbot, M.A. (1956)
Microflora des farines francaises
Meunerie Francaise 42–52

Soenen, M., Pinguair, R. (1937)
A bacteriological study of flour and its importance in milling and baking.

Thatcher, F.S., Coutu, C., Stevens, F. (1953)
The sanitation of Canadian flour mills and its relationship to the microbial content of flour.
Cereal Chemistry 30: 71–102
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CONTROL OF SOME FUNGI CAPABLE OF DEGRADING CELLULOSE AND ALSO WATER BASED POSTER COLOURS

J.N. Dholakia¹ and H.S. Chhatpar¹

Summary

Aspergillus sp. and Fusarium sp. were isolated from water based poster colours and found to be responsible for their degradation and spoilage. The culture filtrate of Aspergillus sp. 1 was found to degrade blue poster colour specifically and was also shown to be cellulolytic, although not capable of utilizing cotton or filter paper except in the presence of small amounts of glucose. 8-hydroxy-quinoline, pentachlorophenol, dimethyl formamide, phenyl mercury acetate and boric acid each found to be effective in inhibiting the growth of Aspergillus sp. without affecting the properties and qualities of the poster colour.

Methods and Materials

Isolation of fungal cultures

The fungal cultures were isolated by streaking spoiled colours on Saboraud’s agar plates and incubating at 30°C for 72 hours. The composition of Saboraud’s agar used was (in grams per 100 ml): Glucose 2.5, Peptone 1.0, NaCl 0.5 and Agar (Centron) 2.0. The pH was adjusted to 5.5 Fungal isolates were maintained on Saboraud’s agar slants supplemented with colours (20 mg in 100 ml = 0.02%).

Measurement of the colour

Most of the investigations were carried out using blue poster colour. The spectrum of the colour showed maximum absorbancy at 660 nm. Therefore the colour in aqueous solution was measured by observing the optical density at 660 nm.

Degradation of colour by Aspergillus sp. 1

Aspergillus sp. 1 was grown in a synthetic medium supplemented with blue poster colour and incubated at 30°C for six days on a rotary shaker at 250 rpm. At the end of the growth period, the mycelium was harvested by filtration through two layers of cheese cloth and dried at 50°C until constant weight was obtained. The mycelial growth was expressed in mgs dry weight per 100 ml flask. The colour remaining in the medium

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(Received April 1979; in final form, August 1979)
was compared with controls (uninoculated flasks) by observing the decrease in optical density at 660 nm.

The composition of the synthetic medium used was (in grams per 100 ml): Glucose 1.0, NH₄Cl 0.5, NH₄NO₃ 0.1, Na₂SO₄ 0.02, K₂HPO₄ 0.1, MgSO₄ 7H₂O 0.01, Blue poster colour 0.02. pH was adjusted to 5.5

**Colour degrading enzyme system**

The culture filtrate, after dialysis for 4–5 hours at 0–5°C, was used as an enzyme for the study of degradation of the colour. The test system for the degradation of the colour contained: phosphate buffer (pH 7.0) 100 μmoles, colour 0.2 mgs and a suitable amount of enzyme in a total reaction mixture of 3.0 ml. The system was incubated at 30°C for 8 hours. The reaction was terminated by addition of 1 ml of 30% cold trichloracetic acid (TCA). The change in optical density was observed at 660 nm.

One unit is defined as the amount of enzyme required to degrade 1 μg of the colour at 30°C per hour.

**Effect of some anti-fungal compounds on the growth of Aspergillus sp.1**

The mould was grown in the synthetic medium with or without various anti-fungal compounds which are widely used in the paint industry as preservatives. These were: 8(OH) quinoline, dimethyl formamide, boric acid, pentachlorophenol, and phenyl mercury acetate. They were used at the concentrations indicated in the results. The growth was measured as indicated earlier.

**Cellulolytic activity of Aspergillus sp.1**

Cellulase was assayed according to the method of Miller (1959). The test system contained: sodium acetate buffer (pH 5.0) 100 μmoles, carboxy methyl cellulose (CMC) (or cotton or filter paper) 10 mg, and a suitable amount of enzyme protein in a total reaction mixture or 2.0 ml. Incubation was at 50°C for one hour and the reaction was terminated by addition of 110 ml 3–5 dinitro salicylic acid and boiling for 10 minutes. The unit is defined as the amount of enzyme which liberates 1 mg of reducing sugar at 50°C per hour.

**The poster colours**

The poster colours were manufactured by Camlin Pte. Ltd. of Bombay – Andheri. Gum arabic is used as the binder but the composition of the pigment is not revealed by the manufacturers who state only that they use a variety of pigments drawn from mineral organic as well as inorganic sources.

**Results**

Two Aspergillus spp. and one Fusarium sp. were isolated from blue, red, and yellow poster colours (Table 1). Further studies were concentrated on the Aspergillus sp. 1 which was isolated from the blue poster colour.

<table>
<thead>
<tr>
<th>Fungal isolate from Camlin poster colours</th>
</tr>
</thead>
<tbody>
<tr>
<td>Poster colour</td>
</tr>
<tr>
<td>Blue colour</td>
</tr>
<tr>
<td>Yellow colour</td>
</tr>
<tr>
<td>Red colour</td>
</tr>
</tbody>
</table>

**Degradation of colour**

When the fungus was grown in the synthetic medium supplemented with blue colour, the colour was degraded1 When the mould was grown in the synthetic medium with glucose and colour, the colour was also found to be degraded, but there was no correlation between the amount of growth and colour degradation (Table 2);

<table>
<thead>
<tr>
<th>Synthetic medium plus glucose (%)</th>
<th>Mycelial dry weight (mg/100 ml flask)</th>
<th>Colour decomposed (mg/100 ml flask)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>60</td>
<td>-</td>
</tr>
<tr>
<td>0.5</td>
<td>248</td>
<td>19.94</td>
</tr>
<tr>
<td>1.0</td>
<td>250</td>
<td>20.60</td>
</tr>
<tr>
<td>2.0</td>
<td>460</td>
<td>18.88</td>
</tr>
<tr>
<td>4.0</td>
<td>510</td>
<td>16.10</td>
</tr>
</tbody>
</table>

for example, maximum growth occurred with 4% glucose addition but the colour degradation was found to be greatest when the mould was grown with only 1% glucose. It seemed, therefore, that the mould must be producing an extracellular enzyme system which may be responsible for the degradation of the colour. To study this, mould was grown in the synthetic medium supplemented with blue colour. After the 6th day the growth was harvested and the culture filtrate after dialysis was used as a crude enzyme system. When this crude enzyme was incubated with colour the colour was degraded, but boiled enzyme was not able to degrade the colour. This enzyme was specific for this

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1 The authors submitted a colour photograph which confirmed this but it was not possible to reproduce this here. Editor.
Fungi degrading cellulose and water based poster colours J.N. Dholakia and H.S. Chhatpar

blue colour since it was not able to degrade some other dyes which were tested, namely: methylene blue, safranine, and 2,6-dichlorophenol-indophenol. The colour degradation was not due to oxidation or reduction since the incubation of colour with ascorbic acid, diphospho pyridine nucleotide (DPN) or reduced diphospho pyridine nucleotide (DPNH), did not cause any change in colour (Table 3). These data indicate that the mould produces an enzyme system which is responsible for degradation of the colour.

Table 3

<table>
<thead>
<tr>
<th>System</th>
<th>Colour degradation activity (units)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blue colour + buffer</td>
<td>n.d.</td>
</tr>
<tr>
<td>Blue colour + buffer + boiled enzyme</td>
<td>n.d.</td>
</tr>
<tr>
<td>Blue colour + buffer + enzyme</td>
<td>20</td>
</tr>
<tr>
<td>Methylene blue + enzyme</td>
<td>n.d.</td>
</tr>
<tr>
<td>Safranine + enzyme</td>
<td>n.d.</td>
</tr>
<tr>
<td>2,6-dichlorophenol-indophenol + enzyme</td>
<td>n.d.</td>
</tr>
<tr>
<td>Blue colour + ascorbic acid (0.2 g)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Blue colour + DPN (0.1 μmoles)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Blue colour + DPNH (0.1 μmoles)</td>
<td>n.d.</td>
</tr>
</tbody>
</table>

n.d. = no colour degradation

Chemical control

Attempts were made to find a chemical which would inhibit the growth of the mould without affecting the properties of the colour. The materials tested were all without effect on the colour. They are named, and the results given, in table 4 and figures 1 and 2.

Cellulolytic activity

The activity of the enzyme in degrading CMC, cotton and filter paper was found to be greatest when the mould was grown in the presence of glucose (0.1%) and CMC (1%) (Table 5). Cellulase activity was not detectable when glucose was supplied as the sole source of carbon which shows that glucose may be causing catabolite repression of the enzyme and CMC may be needed for its induction. However, a low concentration of glucose is needed initially for growth. There was insufficient growth when cotton or filter paper were supplied as sole carbon sources. The maximum levels of production of the cellulolytic enzymes occurred on the fourth day of growth. The kinetic constants of CMCase showed Km value of 10 grams per litre and Vmax as 0.833 (Fig. 5).

Table 4

<table>
<thead>
<tr>
<th>Concentration of the inhibitor (mg)</th>
<th>Mycelial Dry Weight (mg/100 ml flask)</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 (OH) quinoline</td>
<td>PMA</td>
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<tr>
<td>0.0</td>
<td>343.5</td>
</tr>
<tr>
<td>0.2</td>
<td>n.d.</td>
</tr>
<tr>
<td>0.5</td>
<td>n.d.</td>
</tr>
<tr>
<td>0.8</td>
<td>n.d.</td>
</tr>
<tr>
<td>1.0</td>
<td>n.d.</td>
</tr>
<tr>
<td>2.0</td>
<td>n.d.</td>
</tr>
</tbody>
</table>

n.d. = indicates no growth

Figure 1 Effect of boric acid on the growth of Aspergillus sp.1

Boric Acid (g %)

Mycelial dry wt. (mg)

0.0 0.25 0.50 0.75 1.00 1.25 1.50 1.75 2.00

Fig. 5
Figure 2  Effect of dimethyl formamide on the growth of *Aspergillus* sp.

Table 5  
Cellulase activity of *Aspergillus* sp.1 grown in synthetic medium containing glucose and various cellulytic carbon sources

<table>
<thead>
<tr>
<th>Carbon source</th>
<th>Cellulase activity (units/100 ml flask)</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>CMCase</td>
</tr>
<tr>
<td>Glucose 1%</td>
<td>ND</td>
</tr>
<tr>
<td>Glucose 0.5% + CMC 1%</td>
<td>356</td>
</tr>
<tr>
<td>Glucose 1% + CMC 1%</td>
<td>531</td>
</tr>
<tr>
<td>CMC 1%</td>
<td>451</td>
</tr>
<tr>
<td>Cotton 1%</td>
<td>6</td>
</tr>
<tr>
<td>Filter paper 1%</td>
<td>4</td>
</tr>
</tbody>
</table>

ND = Not detectable  
CMCase = Carboxymethyl cellulase

Figure 3  Cellulase production by *Aspergillus* sp.1 during incubation for various periods of days.

Figure 4  Lineweaver-Burk plot of CMCase from *Aspergillus* sp.1  
\[
\frac{1}{V} = \frac{1}{V_{max}} + \frac{K_m}{V_{max}} \\
\frac{1}{V_{max}} = 1.2 \quad V_{max} = 0.833
\]
Discussion

Aspergillus sp. was found to be responsible for the spoilage of blue poster colour. Aspergillus, Phoma, Alternaria, Pullularia, and Cladosporium species have all been reported to be responsible for the biodeterioration of paint films (Drescher, 1958; Krumperman, 1958; Reynolds, 1950, and Horvath, 1976). Extracellular enzymes degrading paint films were also reported by Winters and Guidetti in 1976.

We have also seen that Aspergillus sp I shows a high cellulase activity. As this colour is used on drawing paper for painting and since this fungus also has the ability to degrade poster colour, this may create problems for storage of paintings especially in museums.

Both inorganic and organic compounds are used in the paint industry as preservatives. Inorganic compounds which can be used include zinc oxide, cuprous oxide and barium metaborate (Pauli 1972). Of these, boric acid was tested and found to inhibit the growth of Aspergillus sp I. The organic compounds used are phenyl mercury derivatives (Hoffman, 1960); it is noteworthy that mercury compounds possess fungicidal as well as bactericidal activity. Phenyl mercury acetate does not have any effect on the properties of blue poster colour and 0.2 mg per 100 ml was required to inhibit the growth of the mould. Pentachlorophenol was also used as preservative for paint by Hoffmann in 1966. 8(OH)quinoline and dimethyl formamide were also found to inhibit the growth of Aspergillus sp I. Hence these compounds, also, can be used as preservatives of these colours.

Identity of Aspergillus Sp I

This paper was accepted by the referee subject to specific identification of Aspergillus sp I or provision of a cultural and morphological description. This was received from the authors in February 1980 and reads as follows:

Cultural characteristics Colonies woolly, white to yellow at first, turning black later on with reverse white.

Microscopic morphology Mycelia septate and unbranched, conidiophore arising from a specialized foot cell. The conidiophore is enlarged at the tip forming a rounded vesicle which is completely covered with flask shaped sterigmata that produce chains of round, smooth, conidia.

Microscopic morphology of sterigmata Double cover entire vesicle forming radiate head.

The authors consider this to be an isolate of the Aspergillus niger group.

References


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AN Preservation - chemical ............................................... 1
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BM Physical ........................................................................... 3
BN Chemical ........................................................................... 3
BG Toxins ............................................................................. 3
BS Bacterial ........................................................................... 4
BT Fungal ............................................................................... 4
BW Animal Foods ..................................................................... 5
CA Carbohydrates ................................................................... 6
CC Grain .................................................................................. 6
CD Barley, oats, rye ............................................................. 6
CE Maize ............................................................................... 6
CF Rice ................................................................................... 6
CH ......................................................................................... 6

EX Animal fibres .....................................................................
FY Adhesives ...........................................................................
FG Lignin ..............................................................................
FE Cellulose ...........................................................................
FG Vegetable fibres .............................................................
FI Timber ..............................................................................
FJ Bacterial attack .....................................................................
FM Terrestrial fungal attack ..............................................
FN Marine/aquatic fungal attack ....................................
FO Insect attack ......................................................................
FP Marine borer attack ......................................................
FR Preservation ....................................................................... 14
FT Food pulp ...........................................................................
FU Paper ..................................................................................
FY Books, etc. ........................................................................
FZ Porcine ..............................................................................

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Corrosion by microorganisms of jet aircraft integral fuel tanks

Part I: Analysis of fungal contamination

Daniel Cabral

Summary

Twenty-four samples of jet fuel (kerosene) from storage tanks, hose tips, and aircraft integral fuel tanks, were analyzed. It has been possible to verify the presence of Cladosporium resinae f. avellaneum as the principal contaminant in these systems.

Comparison between Hazzard and Kuster's technique (1962) and visual examination of the interior of the tanks sampled has allowed an estimation of the levels of contamination which differentiate "normal contamination" from "abnormal proliferation." From the results obtained it is considered that tanks with less than 500 viable particles of C. resinae per liter exhibit "internal contamination" but not "proliferation." Results above this number indicate a possible "proliferation".

Introduction

Contamination of jet aircraft fuel systems by microorganisms has been studied by several workers in different parts of the world, including Edmon and Cooney, 1967; Roger and Caplan, 1964; Darby et al., 1968; in the United States, and Hazzard, 1961, 1962, 1963; and Parbery, 1970, 1971, in Australia, and Park 1975; Scott, 1971; Scott and Hill, 1971; Scott and Forsyth, 1976; in Great Britain, and Sheridan and Soteros, 1974; Sheridan, 1975; Sheridan et al., 1973; in New Zealand, among others). Results of research in this field in Southern South America are almost nil.

The subject of this paper is a report on the incidence of fungal contamination in fuel systems of jet aircraft in Argentina, estimating by an indirect technique, the existence of a "normal contamination" caused by the inevitable introduction of conidia with fuel or a degree of "fungal proliferation" in the interior of fuel tanks such as to justify their being opened up, cleaned, and overhauled. This analysis was undertaken in tanks of only one type of aircraft in which previous problems of fungal contamination have been recorded.

Materials and Methods

Samples of kerosene jet AI are taken at three different points in the fuel system, namely: a) storage tanks, b) hose-tips, and c) integral fuel tanks. In the first two cases one litre sterilized bottles were employed. In the case of the integral fuel tanks samples were taken through drainage valves. To minimize outside contamination (Sheridan and Sheridan, 1972), the apparatus shown in figure I was used.
International Biodeterioration Bulletin ISSN 0020–6164 16 (1) Spring 1980

Valve trigger

Rubber-foam protector against air-contamination

Fuel-collecting funnel

Fuel entry perforations

Rubber protector

Perforated rubber stopper

Copper tube allowing passage of fuel

Bottle receiving fuel sample

Perforations allowing fuel flow into bottle

Rubber protector

Figure 1. Sampling Apparatus

The samples were analyzed by the method described by Hazzard and Huster (1962) which makes it possible to measure the degree of contamination as a comparative value. Such a method appeared to be the best suited to this type of analysis, notwithstanding its inconveniences (Hazzard and Huster, 1962; Sheridan et al., 1973). A Millipore 047 03 filtering flask with an Oxoid 0.45μm pore size membrane was used to retain fungal particles. In the first analysis of each sample, 50 and 100 ml were filtered in duplicate, but these quantities were reduced or increased in later samples, according to the degree of contamination of the system and to permit a better colony count. Samples were vigorously shaken before each filtration. After all the kerosene had passed through the membrane, the filter was washed with two 10 ml aliquots of a 0.5% v/v solution of detergent (Lutensol AP 10) sterilized by filtration through a Millipore membrane followed by two 20 ml aliquots of 0.85% w/v of sterile NaCl solution. The membrane was then placed on a medium of malt extract agar in petri dishes and incubated at 25°C. The samples were processed, when possible, within 48 hours of sampling; when the process is delayed, the results are usually poorer (Sheridan et al., 1973). Whenever feasible, a visual inspection of the interior of the sampled tanks was made, to verify the presence or absence of fungal growth. The solid samples obtained were examined with the optical microscope.

All strains isolated were tested for their capacity to metabolize kerosene, in order to evaluate the degree to which these organisms are involved in the formation of "biological sludge" inside the tanks. Erlenmeyer flasks of 125 ml capacity were used, to which were added 40 ml of nutritive solution and 20 ml of Jet Al kerosene sterilized by filtration. Two nutritive solutions were tested: Bushnell-Haas (1941) and a modified Klausmeir's, (Park, 1975). Growth was evaluated visually every 5 days (Parbery, 1965) during 55 days. The strains were inoculated as a suspension of spores, and incubated at 25°C.

Results

Table 1 shows the list of fungi isolated, their percentage frequency of appearance, and the maximum and the minimum levels of contamination expressed as the number of viable particles (colony count) per litre of fuel. The predominant fungi were Cladosporium resinae f. avellaneum (imperfect state of Amorphotheca resinae, Parbery), and Penicillium spp., isolated from 65% and 48% of the samples, respectively, followed by Alternaria alternata (30%), Cladosporium cladosporioides (17%), and Acremonium spp. (13%). (Acremonium spp was identified by Dr. W. Gams as an ill defined species, probably intermediate between Acremonium and Phialophora).

The presence of Acremonium spp. was detected only in samples from storage tanks, whereas the other species exhibiting high frequency of appearance were isolated from all three sources of samples analyzed.

Cladosporium resinae f. avellaneum was the only form of this species found as a contaminant in the fuel systems. Of all the organisms isolated, whose capacity to metabolize kerosene was tested, only C. resinae f. avellaneum showed active growth in both solutions. Of the remaining isolates, only Acremonium spp. exhibited incipient growth in both nutritive solutions. These results differ notably from those obtained by Darby et al., 1968; Park, 1975; and Sheridan and Soteros, 1974.

All the strains of C. resinae exhibited fast and profuse growth, and a high degree of sporulation which, together with the great sporulation observed in the fungal samples from the interior of the tanks, shows the great adaptability of all strains to kerosene assimilation. These results show a high degree of contamination (500 viable particles/ litre), which differs from the figures obtained by other workers (Sheridan, et al., 1973).

Since none of the fungi isolated, excepting C. resinae, exhibited growth on cultures having kerosene as the sole carbohydrate source, mixed cultures of the different
Table 1
Fungi isolated from aircraft fuel systems.

<table>
<thead>
<tr>
<th>Genera and species</th>
<th>Percentage of contaminated samples. (Out of 24 samples analysed)</th>
<th>Range of levels of contamination (Particles per liter of fuel)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acremonium sp.</td>
<td>13</td>
<td>20–260</td>
</tr>
<tr>
<td>Alternaria alternata</td>
<td>30</td>
<td>6–50</td>
</tr>
<tr>
<td>Cladosporium resinae f. avelaneum</td>
<td>65</td>
<td>10–5000</td>
</tr>
<tr>
<td>Cladosporium cladosporioides</td>
<td>17</td>
<td>20–40</td>
</tr>
<tr>
<td>Cladosporium sphaerospermum</td>
<td>4.2</td>
<td>40</td>
</tr>
<tr>
<td>Cladosporium sp.</td>
<td>4.2</td>
<td>10</td>
</tr>
<tr>
<td>Epicoccum purpurascens</td>
<td>4.2</td>
<td>100</td>
</tr>
<tr>
<td>Fusarium sp.</td>
<td>4.2</td>
<td>15</td>
</tr>
<tr>
<td>Paecilomyces variotii</td>
<td>4.2</td>
<td>200</td>
</tr>
<tr>
<td>Ulocladium chartarum</td>
<td>4.2</td>
<td>10</td>
</tr>
<tr>
<td>Penicillium spp.</td>
<td>43</td>
<td>50–2500</td>
</tr>
</tbody>
</table>

strains were made to verify the possible existence of synergic effects. It was thought appropriate to carry out this test since, in two samples of solid material obtained from the inside of integral aircraft fuel tanks, evidence was found of the growth of Penicillium spp. The mixed cultures were prepared in the same way as indicated for those of a single species, with the difference that in the inoculation the same amount of spore suspension of both fungi was introduced. The inoculations made were as follows:

C. resinae – Acremonium sp
C. resinae – Alternaria alternata
C. resinae – Penicillium sp.
Alternaria alternata – Acremonium sp.
A. alternata – Penicillium sp.
Acremonium sp. – Penicillium sp.

Growth, as verified visually, was only observed in those cultures of which one of the components was C. resinae. The microscopic observation of such growth only revealed the existence of vegetative and reproductive structures of C. resinae.

According to the results obtained by other workers and to those obtained in the present research, in Table 2 only the extreme variations in the number of viable particles per litre of C. resinae were taken as indices of contamination. For integral jet aircraft tanks the results obtained during visual inspection are recorded separately.

Conclusions

The predominance and high frequency of C. resinae in the samples agrees with results obtained in different parts of the world and confirms the importance of this species as a principal contaminant of jet fuel systems. Regarding the other species, the frequencies of appearance and levels of contamination given by various authors show high discrepancies (see Sheridan et al, 1973 and Darby et al, 1968). Similarly, our own results are at variance with those of most other authors, although there is agreement for a few species. We wish to stress, in our results, the absence of Aspergillus spp., and the low frequency of Paecilomyces variotii. These differences from other authors may perhaps be explained by the predominance, in different parts of the world, of different kerosene-tolerant strains, such as those found in Alternaria spp. in New Zealand and in the U.S. by Sheridan and Soteros (1974).

As mentioned in the Introduction, the main objective of the present work was to evaluate by Hazzard and Kuster's method, the values of contamination that indicate, with a minimum of error, the fungal proliferation inside a jet fuel tank.

Although we met with difficulties in the inspection of the jet fuel tanks which had been sampled, the results given in Table 2 show a great difference between the
### Table 2

Range of level of contamination (number of particles of *Cladosporium resinae f. melaleuca* per liter of fuel) in the various points of the system analysed

<table>
<thead>
<tr>
<th>Sites sampled</th>
<th>Range of levels of contamination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Storage tanks</td>
<td>0–50</td>
</tr>
<tr>
<td>Hose tips</td>
<td>0–110</td>
</tr>
<tr>
<td>Visibly contaminated jet-fuel tanks</td>
<td>1170–4990</td>
</tr>
<tr>
<td>Non-visibly contaminated jet-fuel tanks</td>
<td>0–450</td>
</tr>
</tbody>
</table>

**Notes to table 2:**

1. Storage tanks presented only slight visible contamination, but it was impossible to verify the real visual contamination due to the tank pattern and to problems with the firms supplying the fuel.

2. At the hose tips no visible contamination was observed for the same reasons as above.

3. The hose tips belong to the vehicles transporting fuel from storage tanks to the aircraft. The higher degree of contamination compared with storage tanks, as shown in the table (when it ought to be similar, since it is the same fuel) is probably due to the homogenization that is produced upon changing fuel from one vessel to another; this effect does not exist in the stationary liquid within the storage tanks.

Contamination data obtained from tanks where proliferation was observed and from those that did not exhibit it. From these results we consider that tanks showing a number of viable particles not higher than 500 per litre exhibit contamination but not proliferation. Results higher than this would indicate a probable proliferation and a visual inspection of such tanks would be highly desirable.

Some erroneous results obtained by this method might be due to fungal development without sporulation; however, none of the solid samples examined, even with incipient proliferation, showed even a low level of sporulation. This appears to reduce to a minimum such an objection to this method of evaluation, at least under the present conditions of analysis.

**Acknowledgements**

The author wishes to thank Dr. Jorge E. Wright very especially, for his valuable orientation, reading of the MS, and translation of the paper into English. Thanks are also due to Dr. J.L. Deangells, head of the Testing of Materials Laboratory, First Air Brigade, El Palomar, and to Mr. Lize for his cooperation without which this research would not have been possible.

The author also wishes to express his most sincere thanks to Dr. D.G. Parbery, School of Agriculture, University of Melbourne, Australia, for confirming the identity of the isolates of *Cladosporium resinae* and to Dr. W. Gams, Centraalbureau voor Schimmelcultures, Baarn, Netherlands, for identification of one of the species of *Acremonium*.

**References**

Bushnell, L.D. and Haas, H.F. (1941)  
The utilization of hydrocarbons by microorganisms.  
Journal of Bacteriology 41: 653–673

Darby, R.T., Simmons, E.G. and Wiley, B.J. (1968)  
A survey of fungi in a military aircraft fuel supply system.  

Edmond, P., and Cooney, J.J. (1967)  
Identification of microorganisms isolated from jet fuel systems.  
Applied Microbiology 15(2): 411–416

Hazzard, G.F. (1961)  
Defence Standards Laboratory (Australia) Report 252

Hazzard, G.F. (1963)  
Defence Standards Laboratory (Australia) Report 252

Fungal growths in aviation fuel systems. Part 2: Test methods.  
Defence Standards Laboratory (Australia) Report 252

The kerosene fungus.  

Parbery, D.G. (1971)  
Biological problems in jet aviation fuel and the biology of *Amorphotheca resinae*.  
Material und Organismen 6: 161–208

Park, P.B. (1975)  
Biodeterioration in aircraft fuel systems.  
Academic Press, London
Corrosion by microorganisms of aircraft fuel tanks. I Analysis of fungal contamination. Daniel Cabral

Roger, M.R. and Kaplan, A.M. (1964)
A survey of the microbiological contamination in a military fuel distribution system.
Developments in Industrial Microbiology 6: 80–94

Scott, J.A. (1971)
Microbiological contamination of aircraft fuel tanks—airframe considerations.
S.A.E. Papers No. 710438

Microbiological aspects of subsonic and supersonic aircraft.

Scott, J.A. and Forsyth, T.J. (1976)
Thermophilic microorganisms in aircraft fuel.
International Biodeterioration Bulletin 12(1): 1–4

Sheridan, J.E. (1975)
Note on occurrence of Cladosporium resinae in New Zealand jet fuel.
New Zealand Journal of Science 18: 209–210

Sheridan, J.E. and Sheridan, M. (1972)
Periodicity of "kerosene fungus" Amorphotheca resinae Parbery, conidial state Cladosporium resinae (Lindau) de Vries, in the atmosphere over Wellington, New Zealand, in 1971
Search 3 (10): 385–386

Sheridan, J.E., Soteros, J.J. and Sheridan, M. (1973)
Cladosporium resinae in jet fuel.
New Zealand Journal of Science 16: 523–528

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HOUSEHOLD PESTS: A GUIDE TO THE IDENTIFICATION AND CONTROL OF INSECT, RODENT, DAMP AND FUNGOID PROBLEMS IN THE HOME. Peter L.G. Bateman. pp. 176 SBN 0 7137 0915 4 Blandford Press, POOLE, Dorset, England. £5.95

The note on the fly-leaf claims that householders may learn all they need to know from this book to deal with infestation problems in the home. Fortunately, in the text, the reader is clearly advised to seek qualified opinion on many of the potentially harmful problems created in buildings by organisms.

In Part (1) there is an outline of the reasons for controlling infestation including a summary of existing legislation. The reader is also given some idea of the economic damage which may be caused by pests.

The author, in the major part of the book, valiantly attempts to describe some of the vast array of pest species which, given the correct circumstances, may invade the home. He frequently suggests ways to eradicate these pests using pesticides or proofing and housekeeping methods. Much of the section on crawling insects would leave the average householder as bewildered as the confused flour beetle! This is because it is extremely difficult to adequately describe a small beetle without the use of illustrations so that it may be identified by the layman. However, on balance the author has successfully selected the narrow path between too much and too little information.

The final part of this volume contains a haphazard selection of topics; from the latest interpretation of the Pied Piper of Hamelin legend to a rather meaningless section entitled “Identification Service”. The shorter life cycle of insects with more generations per given period, when compared with vertebrates, is held as the main reason for the greater diversity in insects. Surely it is the larger numbers of offspring produced by the insects during their life cycles which produces the enormous potential for mutation?

In conclusion, this is an interesting book which will be of value to those studying environmental health and catering, rather than to householders.

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