MICROBIOLOGICAL CORROSION OF METALS—MARINE WOOD BORERS—RODENT ATTACKS ON STORED PRODUCTS—FOULING OF SHIPS BY BARNACLES—DETERIORATION OF STONE BY BACTERIA—ROTTING OF WOOD BY FUNGI—BACTERIAL BREAKDOWN OF ASPHALT—MILDEWING OF LEATHER—INSECT DAMAGE TO BOOKS—BIRD HAZARDS TO AIRCRAFT—FUNGI IN JET FUEL TANKS—TERMITES IN TIMBER—MICROBIOLOGICAL ATTACK ON RUBBERS PLASTICS AND PAINTS,—FUNGAL ETCHING OF GLASS.

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EDITORIAL STATEMENT

There will be four issues of the *International Biodeterioration Bulletin* in 1969 and subsequently. This quarterly production has been necessitated by the increasing number of useful articles submitted for publication, by the need to quicken the transference of information from authors to readers and by subscriber requests. The price of the Bulletin will be maintained at £1 per issue, the annual subscription thus becoming £4 ($10) for four issues.

*IBBRIS* [International Biodeterioration Bulletin Reference Index Supplement] will now cost £4 ($10) per year for four issues, instead of the previous subsidised price of £1 per year. The new cost covers the new printing charges, but not the cost of gathering and processing the information, nor the enquiry services, which are still subsidised.

The Combined Annual Subscription for four issues of the *International Biodeterioration Bulletin* and four issues of *IBBRIS* is £6 ($15) per year.

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CONTENTS

NEWS AND COMMENT 1-2

FUNGICIDES AND BACTERICIDES FOR USE IN RUBBER 3-8
B. J. Rytych

ASSESSMENT OF PAINTS FOR FUNGUS RESISTANCE 9-14
E. Hoffmann

A SIMPLE METHOD FOR THE ASSAY OF THE CELLULOLYTIC ACTIVITY OF FUNGI 15-20
J. H. Walsh and C. S. Stewart

THE UTILIZATION OF N-ALKANES BY PSEUDOMONAS AERUGINOSA UNDER CONDITIONS OF ANAEROBIOsis I. PRELIMINARY OBSERVATION 21-25
R. W. Traxler and J. M. Bernard

A NOTE ON ENDOGENOUS AND BIODETERIORATIVE FACTORS IN THE RESPIRATION OF DORMANT YAM TUBERS 27-30
D. G. Coursey and J. D. Russell

BOOK REVIEWS 31-36
TABLE DES MATIERES

Nouvelles et commentaires .................................................. 1-2
FONGICIDES ET BACTERICIDES A L'USAGE DU CAOUTCHOU ................. 3-8
B. J. Rytych
EVALUATION DES PEINTURES RESISTANT A L'ATTAQUE PAR LES CHAMPIGNONS.................................................. 9-14
E. Hoffmann
UNE METHODE SIMPLE DE CONTROLE DE L'ACTIVITE CELLULOLYTIQUE DES CHAMPIGNONS.............................................. 15-20
J. Walsh et C. S. Stewart
L'UTILISATION PAR PSEUDOMONAS AERUGINOSA DES N-ALKANES DANS DES CONDITIONS D'ANAEROBIOSE. I. OBSERVATIONS PRELIMINAIRES .................................................. 21-25
R. W. Traxler et J. M. Bernard
NOTE SUR LES Facteurs ENDogenes ET DE BIODETERIORATION DU Taux DE RESPIRATION DES TUBERCULES D'IGNAME A L'ETAT DORMANT ............. 27-30
D. G. Coursey et J. D. Russell

Revues des livres .................................................................. 31-36

INHALT

Nachrichten und Kommentar .................................................. 1-2
FUNGIZIDEN UND BAKTERIZIDEN FUR DEN GUMMIGEBRAUCH ......... 3-8
B. J. Rytych
PRÜFUNG DER WIDERSTANDSFÄHIGKEIT VON FARBEN .................. 9-14
E. Hoffmann
EIN EINFACHES VERFAHREN ZUR PRÜFUNG DER CELLULOLYTISCHEN AKTIVITÄT VON PILZEN .................................................. 15-20
J. Walsh und C. S. Stewart
DIE NUTZUNG VON N-ALKANEN DURCH PSEUDOMONAS AERUGINOSA UNTER ANAEROBEN BEDINGUNGEN. I. VORLÄUFIGE BEOBACHTUNG 21-25
R. W. Traxler und J. M. Bernard
EIN KURZER BERICHT ÜBER ENDOGENE UND DURCH ORGANISMEN HERVOR GERUFENE EINFLÜSSE AUF DIE ATMUNG VON RUHENDEN JAMSWURZELKNOLLEN 27-30
D. G. Coursey und J. D. Russell

Buchbesprechungen .................................................................. 31-36

CONTENIDO

Nuevas y comentarios .................................................. 1-2
LOS FUNGICIDAS Y LOS BACTERICIDAS EMPLEADOS PARA EL CAUCHO .................. 3-8
B. J. Rytych
AVALUACIÓN DE LAS PINTURAS PARA LA RESISTENCIA A LOS HONGOS .......... 9-14
E. Hoffmann
UN MÉTODO SENCILLO DE INDAGAR LA ACTIVIDAD CELULOLITICA DE LOS HONGOS .................................................. 15-20
J. Walsh y C. S. Stewart
LA UTILIZACIÓN DE LOS ALKANES N POR PSEUDOMONAS AERUGINOSA BAJO LAS CONDICIONES DEL ANAEROBIOsis. I. OBSERVACIÓN PRELIMINAR 21-25
R. W. Traxler y J. M. Bernard
APUNTES SOBRE LOS FACTORES ENDÓGENOS Y BIODETERIORATIVOS EN LA RESPIRACIÓN DE LOS TUBÉRCULOS INACTIVOS DEL YAM .... 27-30
D. G. Coursey y J. D. Russell

Revistas de libros .................................................................. 31-36
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FISON'S LIMITED, Cambridge Division, Saffron Walden, Essex; makers of agricultural chemicals. Research on control of weeds, plant diseases, spoilage organisms, and agricultural and livestock pests.

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GAGLIARDI RESEARCH CORPORATION, East Greenwich, Rhode Island, U.S.A.; sponsored industrial research in textile chemical dyeing and finishing products and processes.

GALLOWAY & BARTON-WRIGHT, Haldane Place, London, S.W.18.; consultants in industrial microbiology and microbiological deterioration.

GEIGY (U.K.) LIMITED, Simonsway, Manchester, 22.

J. R. GEIGY S.A., Basle, 21, Switzerland; manufacturers of dyestuffs, industrial chemicals, pharmaceuticals and agricultural chemicals.

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SCIENTIFIC CHEMICALS INC., 1637 South Kilbourne Ave., Chicago, Illinois, 60623, U.S.A.; leading manufacturers of industrial fungicides and bactericides who maintain substantial research and development facilities to assist customers in the development of final products geared to meet government and industry standards.

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NOTES FOR CONTRIBUTORS

The International Biodeterioration Bulletin is published four times per year (Spring, Summer, Autumn and Winter). Typescript contributions in triplicate should be sent to the Editor, Dr. H. O. W. Eggins, at the above address.

The Bulletin acts as a vehicle for the publication of works on all aspects of biodeterioration, i.e. the deterioration of materials of economic importance by micro-organisms, insects, rodents, etc.

Contributions may be in English, French, German or Spanish and should be submitted in triplicate on international A4 size paper (21.0 cm × 29.7 cm or 8.27 in. × 11.69 in.); typewritten on one side of the paper only. A summary of 25-100 words should accompany each contribution.

Illustrations should be clearly drawn in Indian ink or should be photographed. The reduction desired should be clearly indicated and illustrations when reduced are not to exceed 17 cm × 26 cm. Where figures are to be inserted in the text the approximate position for each one should be clearly marked in the typescript.

The bibliographic references are to be indicated in the text as, e.g.:

Reese and Levison (1952).

and in the bibliography:


Authors are requested to abbreviate journal titles according to the conventions of the World List of Scientific Periodicals.

Proofs will not be sent to authors before final publication.

25 reprints will be sent free of charge to each author. Additional reprints are obtainable on application to the Publications Editor at a charge of £5 ($12) per 100.
NEWS AND COMMENT

It is with great regret that we record the death on 20th February, 1969 of Dr. Ing. Edgar Morath, Professor of the Technical University and Secretary General of the International Academy of Wood Science in Vienna.

Dr. Ing. Morath whose subject was wood preservation and who was one of the Biodeterioration Information Centre’s Co-operating Specialists, was one of the Biodeterioration Specialists was 72.

Dr. W. P. K. Findlay retired at the end of January, 1969 from his position as Assistant Director of the Brewing Industry Research Foundation, Nutfield, England. Dr. Findlay was previously Head of the Mycology Section of the Forest Products Research Laboratory and is the author of a number of books on timber decay and preservation.

Dr. Findlay will now be able to give more time to his consulting practice which is carried on at the address below. His special fields of interest include the preservation of timber, dry rot and woodworm attack in buildings and other structures, and the legal aspects of disputes relating to these subjects.

Dr. W. P. K. Findlay,
St. George’s Hill,
New House Lane,
Salfords,
Redhill, Surrey, England.
Telephone 0293 4 2462.

We should like to congratulate Mr. D. A. G. Isaksson, Ministry of Agriculture, Fisheries and Food Pest Operator of Montgomeryshire, Wales on the award of the British Empire Medal, announced in the New Year Honours List.

A group of the ASTM (American Society for Testing and Materials) Committee on the Corrosion of Metals in the United States is presently conducting measurements by electrochemical methods of corrosion rates of five metals and alloys buried in the soil. Three government and three industrial organisations are participating in these tests which will be completed in about a year. Measurements of soil resistivity, redox potential and pH are being conducted in addition to those of the corrosion rate. Evaluation of this data may provide better understanding of the anaerobic corrosion process in which bacteria have been incriminated.

The Ecology Bookshop has recently opened at the address below and exists to serve the growing number of people whose awareness and interest in ecology, conservation and related subjects is active in one way or another. Orders from stock can be supplied by return post and any other title available will be obtained on request. A stock list is published which will be revised every six months. Opening hours are 10.00-19.00 on weekdays and 10.00-12.30 on Saturdays.

The Ecology Bookshop,
45 Lower Belgrave Street,
London S.W.1.
Telephone 01-730 8603

Rentokil’s turnover in 1968 amounted to £8,820,000, of which £1,779,000 was accounted for by the group’s overseas activities. (Abstracted from The Times).

Burmah Oil control Atlas Preservatives through their acquisition of Castrol. (Abstracted from The Times).

A new type of quick-acting, safe mouse-killer now available to the food trade and general public overcomes mice that are resistant to warfarin and dispenses with the need for prolonged baiting.

This new rodenticide, Alphakil, has been developed by scientists of Rentokil Laboratories Limited and has been successfully used by their pest control teams and by local authorities for the past three years.

Mice are selective in the type of food they eat and are often difficult to control with baits that depend on a cumulative effect over a long period of time. The mouse’s first meal of Alphakil is also its last. It drops unconscious within feet of the bait and dies as its body temperature quickly falls. The dead bodies are easily recovered for disposal.

Alphakil effectively replaces preparations that were prohibited under the Animal Cruelty Poisons Act (1964) and it has been welcomed by the Universities Federation for Animal Welfare as being humane in action. Apart from this support, the preparation has been cleared under the Pesticides Safety Precaution Scheme of the Ministry of Agriculture, Fisheries and Food. Alphakil is for use indoors only.

Obtainable from:
Rentokil Products Division, Felcourt, East Grinstead, Sussex.

May & Baker Ltd. have developed a new technique for Thin Layer Chromatography, involving the use of the special Chromalay grooved plate, similar to a conventional T.L.C. plate, except that it carries near one end and parallel to it, a rounded groove 1·5 mm. in depth. Normal coating equipment can be used to cover the plates with adsorbent, the only difference being that 50 per cent more adsorbent is needed for grooved plates.
Up to ten times more crude extract can be applied to the grooved plate as opposed to a normal plate without overloading occurring. This is because the layer of adsorbent in the groove is thicker than on the plate itself and there is less tendency for the sample to spread.

Using a method of preliminary separation on the plate itself (which eliminates the necessity for a preliminary clean-up using separate equipment), the sample is brought to a new origin above the groove. The following method describes the technique.

The plate is dipped into a relatively volatile solvent ensuring that the solvent level lies a few millimetres below the bottom of the groove. As the solvent flows up, it carries the more mobile constituents of the extract with it. When the solvent front reaches a line 1 to 2 cm. above the groove, the plate is removed and the solvent is driven off; the process is repeated until all the mobile constituents are concentrated in a narrow regular band just above the groove. Use can be made of a shallow covered glass vessel with a longitudinal slit on top to accommodate the grooved plate. This slit is about 1 cm. above the groove. As the solvent front reaches the slit evaporation occurs and the new origin is formed just beyond it.

The advantages of grooved plate T.L.C. over conventional plate are:

- up to ten times more crude sample can be applied in the groove without the spreading that would occur on an ordinary plate;

- separation and preliminary clean-up are carried out on the one plate, thus eliminating the need for extra apparatus and minimising risk of loss of extract during transfer;

- the accuracy of spotting or streaking is no longer critical, so time is saved.

Further details are available from:

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The latest addition to the "Horo" range of precision ovens, incubators and heating cabinets is the large capacity "Horo" 700.

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The finish of the cabinet is white stoved enamel on steel.

United Kingdom and Eire distributors for "Horo" products are:

Beamcote Ltd.,
240 Hurst Road,
Bexley,
Kent.
Telephone 01-300 1498

The World Conference on Bird Hazards to Aircraft is to be held at Queen's University, Kingston, Ontario, Canada on 2nd-5th September, 1969. It is hoped to include papers on bird strikes, radar detection of bird movements, identification of bird remains, bird-proofing of aircraft, ecological aspects of the congregation of birds at airports and the dispersal of birds from airports.

Further information can be obtained from:

Mr. M. K. Ward,
Conference Secretary,
c/o National Research Council of Canada,
Building M-20,
Ottawa 7,
Ontario,
Canada.

The Pesticides Group of the Society of Chemical Industry propose to run a three day international Symposium on the Technological Economics of Pest Control at Stirling University, Scotland on 3rd-5th September, 1969, by invitation of Professor F. R. Bradbury, Director of the School of Technological Economics which is a main interest of this University.

The key-notes of the Symposium will be immediate and long-term objectives in pest control and how existing and future technological resources may be deployed to achieve them. It will discuss the interactions of the economic consequences of pests to the individual and to the community—losses of food and social values—with the technologies available to deal with them. This subject requires broad treatment as a background to the problems of making profitable business from manufacture of pesticides and of providing funds for research into alternative methods of control.

Further information from:

Dr. M. B. Green,
Hon. Secretary Pesticides Group
S.C.I.,
P.O. Box 13,
Imperial Chemical Industries Ltd.,
The Heath,
Runcorn,
Cheshire,
England.

2
Summary This article discusses the fungicides and bactericides used in the past and at present to protect rubber against microbial deterioration. Consideration is given to the advantages of certain fungicides as well as to the fungicidal action of additives in rubber mixtures.

1. Introduction

Investigations over the last 30 years have shown that both natural and synthetic rubber may be attacked by bacteria (ZoBell et al., 1942; Blake et al., 1949, 1950, 1953, 1955; Shaposhnikov et al., 1952; Nette et al., 1959; Taysum, 1966). Actinomycetales (Sohngen et al., 1914; Spence et al., 1936; Kalinienko, 1938; Rook, 1941; Blake et al., 1949, 1950, 1953, 1955; Shaposhnikov, 1952; Nette et al., 1959) and fungi (Scott, 1920; Kalinienko, 1938; Blake et al., 1949, 1950, 1953, 1955; Nette et al., 1959; Shaposhnikov, 1952; Ross, 1964; Calderon, 1965; Taysum, 1966).

The degree of deterioration of the rubber depends on many factors, of which the chief are:

1. Biological susceptibility of the individual constituents, the polymer, as well as additives (accelerators, curing compounds, fillers etc.).
2. Methods of preparing the raw mixture and the mutual influence of the constituents during the manufacturing processes.
3. Influence of external factors such as temperature, moisture and pH of the environment.

The economic aspects of this problem have been determined by Heuck van der Plas (1965). The annual loss of natural and synthetic rubber due to biological deterioration was estimated for U.S.A. at about 23 million dollars.

2. Requirements for fungicides and bactericides for use in rubber

Morris and Leslie Ewart (1927) mentioned paranitrophenol as a fungicide for smoked crepe and rubber sheets and since that time investigations into the selection of useful fungicides and bactericides for rubber have continued up to the present. In biodeterioration of organic materials the best results up to the date have been achieved in the protection of wood. The requirements laid down by Schultze (1939) for wood-preservatives are perhaps the most comprehensive. Having in view his requirements, which may be adapted for every fungicide for a deteriorated material, as well as requirements pointed out by Dolezel (1964) and MacLachlan et al. (1966) an ideal fungicide or bactericide for rubber must meet following requirements:

1. Good compatibility and mixing properties with the rubber mixture
2. Chemical inertness to other components of the rubber mixture
3. No influence on the physical, chemical and mechanical properties of the final rubber product
4. Low volatility and good heat-stability
5. Resistance to leaching by water
6. Easy incorporation in the manufacturing process
7. Non-toxic or at least non-hazardous to the workers in the rubber factory
8. Non-hazardous in the final rubber products
9. Durable during storing and for the service life of the product
10. Non-corrosive to metals
11. Price justifying the economical effectiveness.

As may be seen the requirements for fungicides are high and it may be concluded that up to date an ideal compound, which would fulfill all these requirements, has not been found. Every compound used as a fungicide has advantages and disadvantages. For this reason the choice of a fungicide for a given type of rubber has to be very careful.

3. List of fungicides and bactericides for rubber

Summing up the available papers on the subject of protecting rubber against microbiological deterioration for the period of the last 30 years the list of fungicides is given. Having in view the chemical characteristics of those compounds, they can be divided into seven following groups:

1 Central Mining Institute, Katowice, Plac Gwarków 1, Poland. Translated into English by Dr. B. J. Zyska.
Fungicides and bactericides for use in rubber. Bozena J. Rytych.

1. Phenol derivatives
- o-nitrophenol (Borecki et al., 1965).
- p-nitrophenol (Yohe, 1939; Heinisch, 1962; Borecki et al., 1965; Blahnik and Zanova, 1965).
- dinitrophenol (Kost et al., 1959).
- p-chlorophenol (Yohe, 1939).
- tetrachlorophenol (Dolezel, 1964).
- pentachlorophenol (Yohe, 1939; Ritzinger, 1959; Reinsch, 1962; Dolezel, 1964; Blahnik and Zanova, 1965).
- p-chloro-m-cresol (Borecki et al., 1965).
- o,p-benzylphenol (Dunlop Rubber Co., 1954; Kost et al., 1959; Brown, 1959; Rajevski and Slogov, 1964).
- 3-alkyl-2,2'-dihydroxy-3,5,5',6'-tetrachlorodiphenyl methane (Dow Chemical Co., 1950).
- o-phenylphenoxide (Heinisch, 1962).
- tetrachlorothymol (Kost et al., 1959; Blahnik and Zanova, 1965).

2. Sulphur compounds
- thiuram derivatives: tetramethylthiuram monosulphide (Du Pont 1952).
- tetramethylthiuram disulphide (Dimond and Horsfall, 1943; Blahnik and Zanova, 1965; Cousins et al., 1957).
- salts of dithiocarbamic acid: zinc diethylidithiocarbamate (Yeager, 1954; Dolezel, 1964; Blahnik and Zanova, 1965).
- zinc dibutylidithiocarbamate (Blahnik and Zanova, 1965).
- lead dimethyldithiocarbamate (Blahnik and Zanova, 1965).
- sodium dibutylidithiocarbamate (Blahnik and Zanova, 1965).
- copper dimethyldithiocarbamate (Fajfer and Jirsa, 1959; Blahnik and Zanova, 1965).
- esters of dithiocarbamic acid (Société des Usines Chimiques Rhône-Poulenc, 1950).
- mixtures of mentioned salts of dithiocarbamic acid (Bakanaukas and Prince, 1955).
- mercapto benzothiazole and its derivatives: 2-mercapto benzothiazole (Dimon and Horsfall, 1943; Dolezel, 1964; Blahnik and Zanova, 1965).
- zinc mercapto benzothiazole (Blahnik and Zanova, 1965).
- sodium mercapto benzothiazole (Vanderbilt Co., 1952).
- benzothiazole disulphide (Dolezel, 1964; Blahnik and Zanova, 1965).
- mixture of benzothiazole disulphide and copper-8-quinolinolate (Monsanto Chemical Co., 1952).
- thiobisazoyl ethers (Goodrich Co., 1953).

3. Salicyl acid derivatives
- sodium salt of salicylicthio-(or para-) toluidine (Brit. Cott. Ind. Res. Assoc., 1940).

4. Mercuric compounds
- phenylmercuric bromide (Dolezel, 1964).
- phenylmercuric chloride (Yohe, 1939; Dolezel, 1964).
- phenylmercuric acetate (Dolezel, 1964).
- phenylmercuric borate (Dolezel, 1964).
- phenylmercuric salicylate (Dolezel, 1964).
- pyridylmercuric stearate (Dolezel, 1964; Borecki et al., 1965).
- phenylmercuric o-benzosulphanilamide (Dolezel, 1964).
- mercuric-dodecyl-trimethylammonium phthalate (Dunlop Rubber Co., 1954).
- diaryl mercury compounds (Sowa, 1952).

5. Naphthalene derivatives
- zinc napthenate (Dolezel, 1964).
- tetrabrombeta naphthol (Yohe, 1939).

6. Quinoline derivatives
- 8-hydroxyquinoline (Wren, 1964).
- zinc-8-hydroxyquinolinolate (Dolezel, 1964).

7. Miscellaneous fungicides
- zinc benzoate (Blahnik and Zanova, 1965).
- cyanides (Blahnik and Zanova, 1965).
- beta-dithiocarbazyl carboxylic acid (Goodrich Co., 1953).
- di-o-toluene-quanidine (Lightbody et al., 1954; Blahnik and Zanova, 1965).
- 10-(2-mercaptobenzothiazolylthi)-5,10-dihydrophenarsazine (Biro and Parkany, 1963).
- 1-methyl-2-phenyl-3-dodecylbenzimidazolium bromide (Farben Fabr.Bayer, 1966).
- 3,4-dichlorobenzilidimethyl dichloroammonium chloride (Farben Fabr.Bayer, 1966).
- stabilised resin amines (Hercules Powder Co., 1952).
- tallium carbonate (Yohe, 1939).
- tributyl tin derivatives (Hueck van der Plas, 1966).
4. The classic fungicides

According to the frequency of quotation of fungicides for rubber in literature it can be seen that only a few compounds have found a wide use. These are compounds known for a longer period and among these the following may be listed: nitrophenol; pentachloronitrophenol; dihydroxydichlorodiphenyl methane; o-phenylphenol; salicylanilide and copper-8-quinolinate.

5. More recent fungicides

It must be admitted, that the total knowledge on the protection of rubber products against microbiological deterioration is still very small. The most frequent publications in this subject are found in English or American technical literature, but often they do not put forward any new ideas on the fungicidal properties of the compounds. Among the fungicides proposed in recent years by British firms the following should be mentioned: halogen derivatives of mercapto-benzothiazol and their salts, according to Brit. Pat. 873 602 (1958); o-phenylphenol for rubber items used in dairy equipment, according to Brit. Pat. 883 448 (1959); mixture of salicylanilide with hydroxydiphenyl methane or dihydroxydiphenyl methane or dihydroxydiphenyl sulphide, according to Brit. Pat. 916 539 (1961); and concentrates of 8-hydroxyquinoline for rubber latex to protect against deterioration by bacteria, according to Brit. Pat. 949 364 (1964). Investigations of Winner (1957) indicate the usefulness of laurie acid to protect rubber against bacteria. Dubrovin (1959) sees the possibility of use of azomethine derivatives of thiophene and Hofmann (1962) advises the use of Antimykotikum produced by Bayer.

Although information on the accelerating influence of copper salts on the ageing of rubber has been known for a long time, the copper compounds are still recommended as useful fungicides. Ritzinger (1959) advises the use of Ottacide P and Milmer 1, which are copper-8-quinolinate, for protecting neoprene. Barnes (1964) recommends the use of copper naphthenate to protect insulating materials. Glupushkin et al. (1966) recommend, apart from salicylanilide, Albichtol, which is a mixture of thiophene with hydrocarbons not exactly identified, in rubber mixtures for electrical cables in tropical climates. The paper of Huciek van der Plas (1966) gives the most recent information of fungicides for rubber. These fungicides are for sale in West European markets. It may be seen from this list, that the choice of compounds is very small. It is surprising that copper or mercury compounds are still marketed. A novelty is the recommendation of capitan and folpet to protect rubber. Phenylmercury compounds are used in greater quantities in Czechoslovakia and have the reputation of effective fungicides for rubber. Apart from phenylmercuric acetate they are insoluble in water, but easily soluble in organic solvents. Their disadvantage is that they are poisonous to human beings and corrosive to aluminium, lead and selenium. Dolezel (1964) states that this group of compounds lose their fungicidal activity after a time. Salicylanilide should not be used for the protection of rubber insulations since it absorbs moisture, resulting in decreased insulating properties of cables, however it may be recommended for other rubber products (Glupushkin, 1966).

The need to protect rubber-coated fabrics also involves the problem of investigating the influence of fungicides for cotton on the physical and chemical properties of rubber as well as on the adhesion, abrasion and strength properties of the rubber-coated product. These investigations were undertaken by Stief and Boyle (1947). The fungicides tested were: copper naphthenate; pyridylmercuric stearate; salicylanilide; pentachlorophenol and 2,2'-methylbis(4-chlorophenol). The influence of these fungicides was investigated in natural rubber, GR-S and neoprene. In aged natural rubber copper naphthenate caused rapid deterioration. Other fungicides had no influence on natural rubber. In GR-S copper naphthenate caused a remarkable loss of strength. Pyridylmercuric stearate had a slight negative effect while the other fungicides had no effect on the physical properties of GR-S. In the case of neoprene copper naphthenate caused a loss of tensile strength after ageing but on the other hand an increase in elongation and tearing strength. Similar tests were done by Hum and Leggett (1947) but here, as well as copper naphthenate, the influence of lubricating oil on six types of rubber was also evaluated. The ageing showed no effect on the rubber when oil was not present. When oil was added to the fabric the copper accelerated the deterioration only when the oil itself negatively affected the rubber.

The question of leachability of fungicides from rubber products is still not satisfactorily explained due to contradictory information. In the opinion of Leeflang (1963) the solubility of fungicides is necessary to enable the fungicidal action, but in this case there is danger of leaching out. For this reason soluble fungicides do not give durable protection of rubber and from this point of view the following fungicides have been tested and disqualified: dichlorophene (up to 3 per cent), p-dichlorobenzene (Chloobol) up to 5 per cent; Termittenschutzmittel P 22 (composition unknown): KA 7007 (a mixture of phenols and sulphur compounds); dichlorophene (Preventol G-D); fungicide on glycol basis (composition unknown), thymol up to 2 per cent; Arquad 18 up to 4 per cent. In the opinion of the National Coal Board (1953), which used copper naphthenate and pentachlorophenyl laurate to protect rubber coated hoses these two compounds have proved unachievable.

A very interesting problem not often mentioned in the literature is the synergism of mixtures of fungicides.

6. Advantages and disadvantages of certain fungicides

Knowledge of the advantages and disadvantages of various fungicides for rubber is at the moment insufficient but some information is available and may be reviewed.
Fungicides and bactericides for use in rubber. Bozena J. Rytych.

for rubber. It was R. T. Vanderbilt Co. Inc. (1952), who pointed out the synergic action of sodium salt of mercaptobenzothiazol and dimethyldithiocarbamate in Vanicide 51. Yeager (1954), basing on his own investigations, gives as an example the synergism of zinc dimethyldithiocarbamate and zinc 2-mercaptobenzothiazol. The synergic mixture given by Bakanauskas and Prince (1955) is close to those explained by Yeager. Here it is a mixture of zinc salts of dimethyldithiocarbamic acid with mercaptobenzothiazol and dehydroabietylammonium pentachlorophenoxide. It frequently occurs that fungicides lose their fungicidal effect when introduced into a given type of rubber. Also the effect of the environment in which the rubber will be used must be considered, but more investigations are necessary. An extensive series of tests done by Kost et al. (1959) on the influence of phenolic derivatives on micro-organisms isolated from rubber is of no significance as the toxicity of the compounds was tested on tissue-paper. In this way the inhibiting influence on the cellulolytic activity of these compounds was evaluated, but not their suitability for protecting rubber against microorganisms. According to the information given by Stief and Boyle (1947) and Hum and Leggett (1947) a fungicide suitable for one type of rubber is useless in another type of rubber.

U.S. Patent 2608551 (1949) gives the information that butadiene-acrylonitrile vulcanisate may have fungicidal properties when 100 parts by weight of the elastomer is heated with 1-5 part of dithiobisbenzothiazole, 1-5 parts of sulphur and 3-4 parts of copper 8-quinoilnolate. This method of obtaining fungicidal vulcanisate is not recommended for use in natural rubber but can be used with GR-S, neoprene and Thiokol rubbers. Similar was an earlier opinion of Phillips (1947), who found that copper 8-quinoilnolate controlled well the growth of fungi in Perbunan 26, Hycar OR-15 and Thiokol rubbers, but supported the fungus growth in natural rubber mixtures. Dolezel (1964) discussing the protection of natural rubber, chloroprene and buna S against microbiological deterioration recommends the following fungicides: pyridylmercuric stearate; salicylanilide; pentachlorophenol; dehydroabietylammonium pentachlorophenoxide and 2,2'-dichloro-5,5'-dichlorodiphenyl methane.

7. Fungicidal action of additives in rubber mixtures

The additives in rubber mixtures may influence the susceptibility of a finished rubber product to microbiological deterioration either in a positive or a negative sense. From this point of view this paper presents only a review of additives which are able to give fungicidal fungicidal properties to the rubber product. According to Blahnik (1965) there are few investigations into the fungicidal or fungastic effectiveness of the additives to be bound with rubber in finished products.

Accelerators for rubber

Dimond and Horsfall (1943) were the first to test the fungicidal properties of such accelerators as 2-mercaptobenzothiazole and tetramethylthiuram disulphide. They found that the first compound had average properties and the second excellent. According to Rook (1955) neither 2-mercaptobenzothiazole nor tetramethylthiuram disulphide, when added as accelerators to the rubber mixture in a normal amount protects rubber against micro-organisms. On the other hand high amounts of tetramethylthiuram disulphide may negatively influence the final properties of rubber, although in the opinion of Bieri (1959) it protects butyl rubber against moulds. Basing on the investigations of Yashin (1957), Blokh gives data from which it can be seen that tetramethylthiuram disulphide and 2-mercaptobenzothiazole lose their fungicidal effect when the rubber mixture is aged at 65°C for a period of 30 days.

In the Brit.P. 684 379 (1951) esters of dithiocarbamate acid, used as accelerators, show bactericidal properties. Blehnik (1965) lists the following accelerators for rubber as having fungicidal properties: zinc dimethyldithiocarbamate; lead dibutyldithiocarbamate; sodium dibutyldithiocarbamate; mercaptobenzothiazole and its zinc salt; benzothiazoldisulphide, tetramethylthiuram disulphide and di-o-toluene-guanidine. Dubrovin et al. (1958) found that Zn, Sb, and Ni salts of salicylanilide, methylphenol, or methyl-diphenol act as accelerators in the rubber mixture, but additionally they protect the mixture against micro-organisms. Later investigations of Dubrovin (1959) have shown that several azometine derivatives of thiophene are good accelerators as well as fungicides for rubber. Good thermal resistance is an additional advantage of these compounds. They are: 5-methyl-2-thienyl-p-aminophenol; 5-methyl-2-thienylnethanol and 2-thienyl-m-nitroaniline. The valuable properties of the above azometine derivatives are stated by Kuzminskii et al. (1960) and are also mentioned by Blokh (1964).

Antioxidants

As it is well known, several aromatic amines, phenols as well as quinones are used in rubber mixtures for their improvement against the oxidation. When reviewing this group of compounds little information on their fungicidal properties is available. According to U.S. Patent 2 502 708 (1950) nitrated hydrogenated cardenol is claimed to be an antioxidant having some fungicidal properties. Winner (1957) was perhaps the only investigator who tested the bacterial properties of some antioxidants for rubber. On the other hand Blahnik (1965) doubts whether the small amount of antioxidants used in the finished product are able to protect the rubber against micro-organisms.

8. Conclusion

In the past years a great effort has been made to select fungicides and bactericides for natural and synthetic rubber. The problem is not yet satisfactorily solved and several questions have to be explained and investigated. It seems that there is a need for investigation into the mechanism of the deterioration of rubber by micro-organisms, the choice of fungicides for a given type of rubber, the synergism of fungicides for rubber, and into the fungicidal properties of several additives, used in rubber mixtures.
Fungicides and bactericides for use in rubber. Bozena J. Rytchy.

References


Fungicides and bactericides for use in rubber. Bozema J. Rytynych.


Yohe, R. V. (1939). *Products,* April, Preventing mildew on rubber products.


ASSESSMENT OF PAINTS FOR FUNGUS RESISTANCE

E. Hoffmann

Summary. The testing of fungus-resistant paints is described and discussed.

Evaluation des peintures résistant à l'attaque par les champignons Description et discussion des peintures résistant à l'attaque par les champignons.

Cases of mould growth on painted surfaces occur under a wide range of climatic conditions throughout Australia, from the tropical humid areas in the north to the temperate regions in the south. This leads to considerable expense in redecoration, and it is therefore important to ensure that coatings used in areas susceptible to the growth of mould have a high degree of fungus resistance.*

Two methods are at present in use for evaluating compounds claimed to increase the fungus resistance of paint. The first is the Vicklund-Manowitz (V.M.) test or one of its numerous modifications. After many years of comparing the assessments of these laboratory tests with actual performance in the field, most workers are now convinced that no correlation exists. The method is valuable in showing whether a compound is active as a fungicide or has some effect in inhibiting fungus growth, but even then some care is required since some paint compositions which do not show fungus resistance in the V.M. test may exhibit it on exposure in the field (e.g. zinc oxide formulations).

It is not surprising that this test fails to predict actual performance when it is considered that the conditions under which the testing is carried out are widely different from those prevailing in practice. Specifically the method does not take into account the loss of active ingredients which takes place in the field. Some workers have tried to correct this by exposing the paint films to artificial weathering before carrying out the test, but correlation between artificial and natural weathering is usually so poor that scarcely anything is gained by introducing this step.

The second method used is to expose test panels at sites where mould growth occurs, and recommendations for fungicides in paint are largely based on the outcome of these tests. This is also a very unsatisfactory procedure, because in case of a failure it does not give any indication of why it occurs. For example, if a fungicide has been shown to be effective in the V.M. test but fails on exposure this could be due to loss of fungicide, to the presence of more viable or different types of mould to those which had been used, or to an exceptionally heavy deposition of plant material. The latter is common with gloss latex paint films, which are especially prone to retain matter blown on to them (Hoffman, 1967).

It is clear that if fungicides are to be used in a rational way, it is desirable to determine the stability of these compounds in paint formulations and under the conditions in which the paint will be used. For this reason it was proposed some years ago (Hoffmann and Georgoussis, 1960; Hoffmann 1963) to combine bio-assay techniques with chemical analysis and to investigate systematically the stability of fungicides in paint films. Such a study has been made for phenyl-mercury compounds (Hoffmann and Georgoussis, 1960; Hoffmann and Bursztyn, 1963, 1964; Hoffmann, Saracz, and Barned, 1966a; Hoffmann, 1964), pentachlorophenol (Hoffmann, Saracz, and Barned, 1966a), para toluene sulphonamide (Hoffmann, Saracz, and Barned, 1967), zinc oxide (Hoffmann et al., 1968), tetramethyl thiuram disulphide (Hoffmann and Saracz, 1969a), salicylanilide, barium metaborate, N-trichloromethylthio-phthalimide (Hoffmann and Saracz, 1967), and copper hydroxyquinolate. The paints are brushed out on sheets of polyethylene terephthalate polyester film, which are then usually exposed under the following conditions:

1. Outdoors at Highett, Victoria
2. In a fog room at 20°C
3. In a constant temperature room at 20°C and 65 per cent R.H.
4. In a hot room at 38°C, 30 per cent R.H.
5. Outdoors at Lae, New Guinea

At Lae, New Guinea, panels of asbestos cement painted with the same coatings are also exposed, and the mould growth is assessed at intervals.

Results obtained under these exposure conditions give a good indication of the fungicidal or fungistatic value of the compound under investigation, that is, how long the fungicidal or fungistatic action is likely to persist, and under what conditions it should be used.

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*It is difficult to give an exact figure for the damage caused by mould growth on painted surfaces, but an estimate can be made. Australia spends about $300 million per year on architectural paint and labour to apply it. From 5 to 10 per cent of this amount is spent in areas susceptible to the growth of mould, and an average improvement of 10 per cent in the lifetime of paint would therefore amount to $15 to 3 million per year.
Assessment of paints for fungus resistance. E. Hoffmann.

Para tolune sulphonamide (PTSA) is recommended as a fungicidal additive to house paints for indoor and outdoor use. However, at Lae practically all the PTSA is lost from a paint film within six months, and panels coated with paint containing PTSA show no appreciable suppression of mould growth compared with the blanks. The loss of PTSA is also very high in the fog room but very low in the constant temperature room, and it can therefore be expected that this compound would be of value in situations where mould would grow at above 70 per cent R.H. but where conditions are not so bad that water is actually running down the painted surface. This has also been confirmed (Hoffmann, Saracz, and Barned, 1967).

N-trichloromethylthio-phthalimide shows a comparatively high stability in the fog room and at 20°C and 38°C, and is therefore likely to be of value in such places as bakeries, where temperature and humidity tend to be high.

The addition of zinc oxide to paints is claimed to increase their fungus resistance, and experiments with latex paints (Hoffmann et al, 1968) showed that this is the case with some brands of zinc oxide. The increased mould resistance lasts only for about two years or less, and this could be ascribed to the adaptation of the mould to the presence of zinc oxide. Analysis of the layer of chalk on the paint film showed somewhat unexpectedly that the zinc oxide had been reduced to a very small percentage of what was originally present. For example, chalk which had been formed on a coating containing about 60 per cent zinc oxide by weight on its pigment content, was found on analysis to contain less than 5 per cent. The knowledge of this suggests an entirely different approach to the formulation of this type of fungus-resistant paint.

Copper hydroxyquinolate is more stable than the other fungicides tested so far, and panels coated with a paint containing it had very little mould on the upper side for three years but considerable mould growth was observed on the under side after six months. (The exposure racks at Lae slope at 45° and face 31° east of true north, and the mould growth on both sides of each panel is being observed). N-trichloromethylthio-phthalimide is quite stable in paint films on the under side of the panels but less stable on the upper sides. The two fungicides were therefore combined in a coating to increase the mould resistance on the under side. After 15 months there is still very little growth of mould in this position.

Another approach in formulating a fungus-resistant paint with copper hydroxyquinolate suggests itself. The reason for the small activity on the under side of the panels could be that the paint film does not chalk sufficiently where it is not exposed to sunlight. Some decomposition of the copper hydroxyquinolate is likely to take place on the top layer of the film, but as this compound is not very soluble the loss cannot be replenished by diffusion, and hence the film becomes less fungus-resistant. Sufficient chalking takes place on the top layer to expose enough copper hydroxyquinolate to retain the fungus resistance. The activity of this fungicide may therefore be increased by formulating the paint so that it chalks slightly, and this could be done by replacing part of the rutile titanium dioxide with zinc oxide. To do this efficiently it is necessary to know quantitatively how much the chalking is increased with given amounts of this pigment, and how this depends on the prevailing weather conditions. Work on this project, which is also important for a better understanding of the weathering process of paint films, is under way (Hoffmann and Saracz, 1969b).

It must be emphasised that a V.M. test combined with stability determinations does not replace a field test of the chosen formulation. For example, copper hydroxyquinolate shows up as active on the V.M. test and is also very stable, but in certain areas the paint containing it is not satisfactory. What the proposed approach* to testing of fungus-resistant paints allows is to choose from the very many possible paint formulations those that are likely to succeed in a given environment.

It has already been mentioned that panel exposures are a reasonable indication of the fungus resistance of paints that are to be used outdoors. Such a procedure is useless for coatings intended for indoor use. If painted panels are attached to walls on which mould growth is known to occur, it will often be found that the paint films show little or no mould, although the walls around the panels may be heavily infested. The reason for this is that less water vapour will condense on the panel, which is better insulated against heat loss than the wall. It follows that to test a paint for indoor use it must be applied directly to the surface which is to be decorated, and this should be done as shown in Fig. 1. Three paints were applied to this wall (Saracz and Hoffmann, 1968)—A, a chlorinated rubber paint; B, a latex paint containing only rutile titanium dioxide as pigment; and C, a latex paint in which part of the titanium dioxide was replaced by zinc oxide. The figure shows the mould growth after four years, and it is evident that only the paint containing zinc oxide is effective.

It might be thought that it would be sufficient to divide the walls into three parts and to apply one type of paint to each part, but this could be very misleading, as can be seen from Fig. 2. The three paints A, B, and C were applied to the wall and its appearance after two years is shown in the figure. If paint B had been tested under two different names, believing them to apply to two different compositions, B1 and B2, it would have been concluded that B1 had a higher fungus resistance than B2, whereas in fact they have the same compositions. The difference is probably due to the slightly higher humidity at the wall surface which is under the sloping roof.

*The usefulness of the analytical method would in some cases be increased by following the loss of the compound in question in the various layers of the paint films. This would be of interest for fungicides which decompose very slowly, e.g. copper hydroxyquinolate.
Assessment of paints for fungus resistance. E. Hoffmann.

Many workers still seem to believe that it is possible to develop a reliable accelerated laboratory test (other than the V.M. test) which will allow an assessment of the fungus resistance of paint. The author considers that this has little hope of success, because it is difficult to accelerate the loss of the mould-inhibiting compound of the paint in a realistic way. It has been suggested (O'Neill and Skinner, 1966) that painted specimens should be exposed in a weatherometer for 250 hours and then tested for mould resistance in a cabinet which has recently been devised (Hendy, 1962). However, it is impossible to deduce from this how long the paint film will remain resistant to fungus, since nothing is disclosed about the rate of loss of the fungicidal compound.

The search for accelerated testing is understandable but must be futile unless a great deal is known about the mechanism of the change in properties to be measured. The stability of a fungicide is one of the most important features to be known in determining the fungus resistance of a paint film.

No detailed analytical procedure can be given, as this will vary with the compound to be analysed. However, two general principles have been followed: (1) The procedure must be quick, and (2) the apparatus used must be as simple as possible. Fungicides often contain elements such as mercury, chlorine, and sulphur which can be easily determined. An oxygen flasks technique was used to obtain the element in a suitable form for analyses, and this was combined with an appropriate microchemical method to make a very quick estimation of the element in question.

It is sometimes necessary to modify known procedures, as is described, e.g. by Hoffmann and Saracz (1964). The equipment used in the combustion flask technique is shown in Fig. 3. A suitable absorbent, e.g. nitric acid for the determination of mercury, is added to the flask which is then filled with oxygen. A sample of paint film (50-100 mg) is wrapped in a paper carrier and placed in the platinum holder. The fuse is ignited and the platinum holder quickly inserted into the flask, after which the absorbent is analysed for the liberated element. With two of these flasks it is possible to make two determinations of mercury in one hour.

The combustion flask method was supplemented by extracting the fungicide from the paint film and determining its ultraviolet absorption spectrum. The techniques used in investigations for phenylmercury compounds (Hoffmann, 1964, 1965; Hoffmann and Saracz, 1964), pentachloro phenol (Hoffmann, Bursztyn and Saracz, 1965), N-trichloromethylthio-phthalimide (Hoffmann, Bursztyn, and Saracz, 1965), salicylanilide and para toluene sulphonamide (Hoffmann, Saracz, and Bursztyn, 1964), copper hydroxyquinolate (Hoffmann, 1966), and barium metaborate (Hoffmann, 1968) have been described and probably can serve as a model for similar cases.

Neglect to take into account the stability of fungicidal compounds in paint formulations has given rise to contradictory opinions as to the effectiveness of these materials and a great deal of unnecessary work, and also to the production of inferior paints. The paint companies who are the users of fungicidal additives cannot be entirely blamed for this state of affairs. In the author's opinion the onus falls on the supplier to establish the stability of the compounds, and to develop and describe quick and simple analytical techniques for their determination*. Paint companies should refuse to handle a product if these data are not supplied (Hoffmann and Saracz, 1967). A step in the right direction is exemplified by a recent advertisement in the Journal of the Oil and Colour Chemists' Association (Vol. 51 No. 6 p.viii), in which the rate of decomposition of a phenylmercury compound (55 per cent lost in 4 months) is compared with the rate of loss (90 per cent in 4 months) of phenylmercury dodecyl succinate. This is important technical information, and the buyer knows what he is getting. It is then up to him to decide whether or not the improvement is significant for his purposes.

References


*It may be pertinent to illustrate what is meant by a simple technique. It has been proposed by Taylor (1965) that the loss of mercury in paint films should be measured by a radioactive method. The method may be the simplest one for those who have facilities to prepare radioactive tagged compounds and the necessary equipment, but it certainly would not be a simple technique for a paint manufacturer's laboratory.
Assessment of paints for fungus resistance. E. Hoffmann.


Fig. 1. Tests of fungicidal paints on the wall of a dye works.
Growth of mould in 4 years.
A—Chlorinated rubber paint
B—Latex paint pigmented with rutile titanium dioxide
C—Latex paint pigmented with rutile titanium dioxide and zinc oxide
Assessment of paints for fungus resistance. E. Hoffmann.

Fig. 2. Tests of fungicidal paints on the wall of a dye works.
Growth of mould in 2 years.
A—Chlorinated rubber paint
B1 & B2—Latex paint pigmented with rutile titanium dioxide
C—Latex paint pigmented with rutile titanium dioxide and zinc oxide.

Fig. 3. Oxygen combustion flask.
A simple method for the assay of the cellulolytic activity of fungi.

A SIMPLE METHOD FOR THE ASSAY OF THE CELLULOLYTIC ACTIVITY OF FUNGI

J. H. Walsh and C. S. Stewart

Summary. A method for measuring the cellulolytic activity of fungi by the degree of clearing when the fungus is grown on a nutrient medium containing finely divided cellulose is described. This is estimated by measuring the area of the microscope field obscured by cellulose particles before and after fungal growth.

Introduction

Numerous methods have been used to measure the cellulolytic activity of fungi, and of these many have involved determinations of the diameters of cleared zones formed when fungi are grown on nutrient media containing finely divided cellulose.

A major problem encountered in methods involving the study of clearing of cellulose is that fungal hyphae and spore clumps often obscure the clear zones. Two techniques have been used to overcome this problem. Aschan and Norkrans (1953) and Savory, Mather, Maitland and Selby (1967) incorporated growth inhibitors into the cellulose medium and inoculated this with borings from a healthy culture. Rautela and Cowling (1966) relied upon anaerobic conditions below the surface of a cellulose medium in test tubes to check fungal spread.

A consideration of these existing methods suggests to us that different criteria of cellulolytic activity are used according to whether the diameter of the cleared zone produced by a growing colony is measured or the width of the cleared zone around or under an inhibited colony. In the first case the measurement reflects the total cellulose destroyed per colony and this depends on the rate of colony spread, as well as on the activity of the cellulose degrading system. In the second case the activity of the cellulose degrading system is measured, and the effect of lateral spread largely excluded. To achieve this however the cleared zone which is measured is produced by cellulose enzymes working at a distance from the mycelium, so that factors such as the diffusibility and stability of the enzyme may be involved.

Method

As a substrate, the nutrient medium of Eggins and Pugh (1962) was used, with incorporation of 1% ball milled cellulose, as suggested by the authors. Two types of cellulose powder were studied. Firstly, 72 hour ball milled Whatman standard grade cellulose powder (CF 11) as suggested by the authors, and secondly a powder derived from a cotton yarn which had proved on analysis to contain 10% non-cellulosic material.

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As a refinement, it was found that a sterile cover slip could be placed on top of the water agar layer before pouring the cellulose medium. For counting the cover slip and the nutrient medium were removed, inverted and placed on a glass slide, thus forming a mount of the cellulose agar with the particles uppermost directly beneath the cover slip. The advantage of this refinement was that a much clearer image was obtained when the particles were viewed under the microscope than with the usual method. Convenience apart however both methods gave the same results, and the results quoted in this paper were obtained with the original method.

Results

As a preliminary, counts were made on a large number of uninoculated plates in order to characterise the amount of variation which could be expected from the controls. A selection of representative counts obtained on uninoculated plates is shown in Table I together with estimates of standard deviation, each value quoted in the Table being the average of two counts. Since three replicate plates were later to be studied in the case of each organism tested, the control values shown in Table I are grouped into random triplets, as would normally occur upon inoculation of the plates.

### TABLE I—Control values obtained before inoculation

<table>
<thead>
<tr>
<th>Triplet No.</th>
<th>Average % Cover Plate</th>
<th>Overall Average</th>
<th>Standard Deviation</th>
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<tr>
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<td>2</td>
<td>3</td>
</tr>
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</tr>
<tr>
<td>6</td>
<td>91</td>
<td>93</td>
<td>95</td>
</tr>
</tbody>
</table>

The results quoted in Table I, together with most of the work quoted in this paper, were obtained using a cellulose powder derived from cotton yarn. This was done because we wished to compare estimates of cellulolytic activity obtained by the particle counting method with those obtained from measurements of tensile strength losses of cotton yarn. Due to the high level (approx. 10%) of impurities present in the yarn, it was felt that such comparisons would be more valid if powdered yarn were used for the particle counting studies.

Since it is much easier to prepare ball milled cellulose from Whatman cellulose powder, tests were made to see to what extent this would give results similar to those for the powdered yarn. Since the particles are of the same size in each case, no modification of the method is needed, whichever powder is used.

Five organisms which showed rather sparse hyphal growth were selected for a comparison of the utilisation of ball milled Whatman cellulose powder (CF 11) and cotton yarn powder. In these cases, successive determinations of particle counts could be made on the same plates after various time intervals, since it was not necessary to remove the fungal growth before counting. The results obtained with two of the fungi studied, Trichoderma koningi and an unidentified Aspergillus sp., are shown in Fig. 1. The percentage clearing after 10 days for all five organisms studied is listed in Table 2.

Figure 1 Comparison of utilisation of ball milled Whatman cellulose powder CF 11, and cotton yarn powder.
Incubation at 25°C.

TABLE 2—Comparison of utilisation of Whatman cellulose powder and cotton yarn powder; % clearing after 10 days at 25°C.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Initial % cover, before inoculation</th>
<th>% clearing after 10 days at 25°C</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Whatman CF 11</td>
</tr>
<tr>
<td><em>Aspergillus</em> sp. No. 9</td>
<td>93</td>
<td>11</td>
</tr>
<tr>
<td><em>Chaetomium globosum</em></td>
<td>90</td>
<td>83</td>
</tr>
<tr>
<td><em>Fusarium moniliforme</em></td>
<td>91</td>
<td>62</td>
</tr>
<tr>
<td><em>Gliomastix</em> sp.</td>
<td>95</td>
<td>73</td>
</tr>
<tr>
<td><em>Trichoderma koningi</em></td>
<td>90</td>
<td>77</td>
</tr>
</tbody>
</table>

From these results, it can be seen that the attack of cotton yarn powder is higher throughout, although by an inconsistent margin. The results obtained were similar however, in that when fungi were arranged in sequence of their cellulolytic activity, the same order was obtained, regardless of which powder was used for the assay.

The cellulolytic activity of a large number of fungal isolates was then measured by the particle counting method, using yarn cellulose powder. Simultaneously, the cellulolytic activity was estimated by the decrease in tensile strength of cotton yarn after the same period of incubation. For the tensile strength studies cotton yarn was incubated in test tubes, half immersed in the nutrient liquid recommended by Marsh, Bollenbacher, Butler and Raper (1949). The isolates used in these experiments were all derived from PVC covered cotton conveyor belt exposed in coal mines. The results are given in Table 3.

Together with the results from these studies, Table 3 lists the cellulolytic activity of these fungi as given by Siu (1951) compiled from the results of previous workers.

Discussion

In comparing the present method with results obtained from tensile strength studies, two features are of major importance. The first of these is the reliability of the results obtained. Statistical analysis (A.S.T.M., 1957) of the tensile strength loss results indicates that at the level of variation encountered in this study, using 5 replicates, differences of 10-15% in average tensile strength losses were significant. In the case of the particle counting results, differences of between 20-25% were significant in a similar analysis. This comparatively low reliability is however increased considerably if the 5 particle counting results (marked (x) in Table 3) which showed excessive variance, are deleted. In this case differences in particle count of 15% are found to be significant, a figure which compares well with that from tensile strength studies, in which more replicates were studied.

The second important feature in which methods might be compared, is the ease of operation. In this respect, the particle counting method involves no specialised equipment, and thus has a considerable advantage over tensile strength studies. On the other hand it is more time-consuming.

It is apparent from Table 3 that for many isolates, particle counting and tensile strength studies result in similar estimates of activity, indicating that the present method is useful for classifying isolates with regard to activity, particularly in comparative studies of a range of organisms, as studied here. In addition both methods show a good agreement with Siu's qualitative data (Siu 1951), though the subjective nature of Siu's terms makes this a more difficult comparison.

In a few cases there is more disparity, notably with *Verticillium lateritium*, *Aspergillus niger*, *A. nidulans* and *A. ustus*, which all show higher activity when assayed by tensile strength losses. Conversely *Penicillium cyclopium* and *Aspergillus oryzae* show higher activity when assayed by particle counting. Although the latter effect may be due to degradation during ball milling, or the finely divided form of the cellulose, the former is more difficult to explain. It is suggested that reduced activity in particle counting estimates may result from the accumulation of inhibitory products around the culture in the petri dish studies, which would be expected to drain away or be dispersed in the tensile strength studies in which the yarns were incubated vertically in a liquid medium. Bravery (1968) has shown the effect of non-cellulosic carbon sources in Eggins and Pugh medium in delaying the development of the cellulolytic activity of certain isolates, but this factor is probably not involved here. The cotton yarn used is impure and contains 8% of non-cellulosic carbon compounds (fats, waxes, pectins, organic acids, free sugars etc.) so that differences in the non-cellulosic carbon content of the media for particle counting, and for tensile strength studies are probably not important.

**TABLE 3—Cellulolytic activity of fungi; decrease in yard particle count, compared to yarn tensile strength losses**

(Incubation 10 days at 25°C)

<table>
<thead>
<tr>
<th>Organism</th>
<th>Average Initial Count</th>
<th>Replicate Final Count</th>
<th>Average Final Count</th>
<th>% Decrease</th>
<th>% Loss of* Tensile Strength</th>
<th>Shi's Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>None (Control)</td>
<td>89</td>
<td>90 90 91</td>
<td>90</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Aspergillus nidulans</td>
<td>89</td>
<td>70 69 74</td>
<td>71</td>
<td>21</td>
<td>41</td>
<td>Weak</td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>89</td>
<td>78 80 79</td>
<td>79</td>
<td>11</td>
<td>33</td>
<td>None†</td>
</tr>
<tr>
<td>Aspergillus oryzae</td>
<td>92</td>
<td>50 41 44</td>
<td>45</td>
<td>51</td>
<td>12</td>
<td>None (Jermyn, 1952)</td>
</tr>
<tr>
<td>Aspergillus recurvatus</td>
<td>90</td>
<td>67 61 58</td>
<td>62</td>
<td>31</td>
<td>30</td>
<td>Not studied</td>
</tr>
<tr>
<td>Aspergillus usus</td>
<td>91</td>
<td>75 70 53(x)</td>
<td>66</td>
<td>27</td>
<td>54</td>
<td>Positive</td>
</tr>
<tr>
<td>Cephalosporium sp.</td>
<td>91</td>
<td>2 14 21(x)</td>
<td>12</td>
<td>87</td>
<td>92</td>
<td>Moderate (some strains) Very strong</td>
</tr>
<tr>
<td>Chaetomium globosum</td>
<td>88</td>
<td>1 9 —</td>
<td>5</td>
<td>94</td>
<td>86</td>
<td></td>
</tr>
<tr>
<td>Circinella mucoroides</td>
<td>90</td>
<td>83 78 88</td>
<td>83</td>
<td>8</td>
<td>5</td>
<td>None (other Circ. species) Not studied</td>
</tr>
<tr>
<td>Doratomyces purpureofuscus</td>
<td>91</td>
<td>35 19 17(x)</td>
<td>24</td>
<td>74</td>
<td>88</td>
<td></td>
</tr>
<tr>
<td>Fusarium culmorum</td>
<td>90</td>
<td>21 15 15</td>
<td>17</td>
<td>81</td>
<td>66</td>
<td>Definite</td>
</tr>
<tr>
<td>Fusarium moniliforme</td>
<td>89</td>
<td>17 12 11</td>
<td>13</td>
<td>85</td>
<td>73</td>
<td>Moderate</td>
</tr>
<tr>
<td>Fusarium solani</td>
<td>90</td>
<td>26 34 30</td>
<td>30</td>
<td>66</td>
<td>75</td>
<td>Moderate</td>
</tr>
<tr>
<td>Geomyces cretaceus</td>
<td>89</td>
<td>29 33 29</td>
<td>30</td>
<td>66</td>
<td>53</td>
<td>Not studied</td>
</tr>
<tr>
<td>Gliocladium roseum</td>
<td>91</td>
<td>16 14 8</td>
<td>13</td>
<td>86</td>
<td>87</td>
<td>Moderate</td>
</tr>
<tr>
<td>Glomastix sp.</td>
<td>91</td>
<td>4 0 1</td>
<td>2</td>
<td>98</td>
<td>82</td>
<td>Strong (G. convoluta)</td>
</tr>
<tr>
<td>Memnoniella echinata</td>
<td>88</td>
<td>0 4 0</td>
<td>1</td>
<td>98</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Mucor racemosus</td>
<td>92</td>
<td>86 86 90</td>
<td>87</td>
<td>6</td>
<td>0</td>
<td>None</td>
</tr>
<tr>
<td>Penicillium crustosum</td>
<td>88</td>
<td>79 76 80</td>
<td>78</td>
<td>11</td>
<td>11</td>
<td>Not studied</td>
</tr>
<tr>
<td>Penicillium cyclopium</td>
<td>90</td>
<td>70 63 73</td>
<td>69</td>
<td>23</td>
<td>5</td>
<td>Not studied</td>
</tr>
<tr>
<td>Penicillium notatum</td>
<td>90</td>
<td>82 77 82</td>
<td>80</td>
<td>11</td>
<td>26</td>
<td>None</td>
</tr>
<tr>
<td>Rhizopus oryzae</td>
<td>91</td>
<td>87 92 89</td>
<td>89</td>
<td>2</td>
<td>12</td>
<td>None</td>
</tr>
<tr>
<td>Stachybotrys atra</td>
<td>92</td>
<td>9 9 30(x)</td>
<td>16</td>
<td>83</td>
<td>91</td>
<td>Strong</td>
</tr>
<tr>
<td>Streptomycies sp.</td>
<td>90</td>
<td>89 91 91</td>
<td>90</td>
<td>0</td>
<td>0</td>
<td>Varies according to species Strong</td>
</tr>
<tr>
<td>Trichoderma koningi</td>
<td>88</td>
<td>2 6 0</td>
<td>3</td>
<td>97</td>
<td>75</td>
<td></td>
</tr>
<tr>
<td>Trichirachium roseum</td>
<td>90</td>
<td>83 87 84</td>
<td>85</td>
<td>6</td>
<td>14</td>
<td>None</td>
</tr>
<tr>
<td>Verticillium lateritium</td>
<td>93</td>
<td>66 61 50(x)</td>
<td>59</td>
<td>37</td>
<td>92</td>
<td>Definite</td>
</tr>
</tbody>
</table>

(x) = excessive variation
* = average of 5 determinations
† = Simpson and March (1964) found some strains were active.

These disparities noted above serve to emphasise that in all probability, no single method yields an absolute value for cellulolytic activity. The technique used for any particular method may modify the results obtained. This is reaffirmed by the results in Table 2, since had Whatman cellulose powder CF 11 been used in the general comparison listed in Table 3, a different set of figures would have been obtained, although the order of activity of the isolates relative to each other would have been much the same.

Bearing in mind the many factors which affect the relevance of results obtained by the methods available, it is felt that the present method gives results as valid as those obtained by any other method, avoiding the criticisms of existing clearing methods. At its best, the replication obtained is of a similar order to that of tensile strength studies on yarn using 5 replicates, a method involving much more specialised apparatus. Although the method was subject to occasional (5/78) erratic results, this is not thought to detract from the usefulness of the results. Some other methods (e.g. respirometric studies, Kaplan, 1964) have also shown this tendency, but have yielded many useful results.

Although the emphasis in this paper has been on the use of yarn cellulose powder, it is felt that the results quoted in the comparison of ball milled yarn and Whatman cellulose powder establish that the method is equally useful when the latter type of particles are used. Because of the ease of preparation of ball-milled Whatman cellulose powder, it is felt that this is probably the better substrate to use, where a simple comparison of one culture with another is required without reference to estimates of activity by other methods.

One point which has arisen in writing up where it is felt the method might be improved, lies in the selection of the area to be counted. In the present study, measurements were made 1 cm. from the colony edge. It may be that the counting region would be better situated at a fraction (e.g. 1/2) of the colony radius from the edge, so that the hyphae in the area investigated are of equal age in all cultures.

Acknowledgements

Identifications of the fungi studied were carried out at the C.M.I. (Kew) by J. J. Elphick, Dr. A. H. S. Onions and Dr. C. Booth, and we are grateful for their expert advice.

We wish to record our appreciation of a grant in aid of research which was donated by the National Coal Board. The views expressed, however, are those of the authors, and not necessarily those of the Board.

References


THE UTILIZATION OF N-ALKANES BY PSEUDOMONAS AERUGINOSA UNDER CONDITIONS OF ANAEROBIOSIS.

I. PRELIMINARY OBSERVATION.

R. W. Traxler and J. M. Bernard

Summary. This paper describes the utilization of n-octane and n-hexadecane by Pseudomonas aeruginosa strain 196Aa under anaerobic conditions as well as the reduction of nitrate by this organism.

There have been innumerable papers which describe the utilization of n-alkanes by micro-organisms in the presence of atmospheric oxygen. It seems logical that such a reduced substrate as an alkane should be oxidized by some micro-organism in the absence of molecular oxygen if the alkane can be induced to lose hydrogen so as to form a double bond between two adjacent carbon atoms. Azoulay, et al., (1963) have shown Pseudomonas aeruginosa strain Sol 20 to possess the capacity to cause a dehydrogenation of n-heptane. This finding suggests that indeed biochemical mechanisms are available whereby a bacterium could cause oxidation of an n-alkane in the absence of molecular oxygen. Hansen and Kallio (1957) report the inability of Pseudomonas stutzeri to reduce sulfate under conditions where one would expect to find a lack of gaseous oxygen.

Pseudomonas aeruginosa strain 196Aa was isolated from contaminated JP-4 fuel. It has been shown to utilize n-alkanes and fatty acids. It will not utilize any of a variety of aromatic hydrocarbons. This organism is a typical pseudomonad with one polar flagellum as shown in Figure 1.

Table I. Composition of mineral salts medium

<table>
<thead>
<tr>
<th>Component</th>
<th>Mg per liter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Na&lt;sub&gt;2&lt;/sub&gt;HPO&lt;sub&gt;4&lt;/sub&gt;</td>
<td>1000</td>
</tr>
<tr>
<td>KH&lt;sub&gt;2&lt;/sub&gt;PO&lt;sub&gt;4&lt;/sub&gt;</td>
<td>500</td>
</tr>
<tr>
<td>NH&lt;sub&gt;4&lt;/sub&gt;N&lt;sub&gt;3&lt;/sub&gt;</td>
<td>2500</td>
</tr>
<tr>
<td>MgSO&lt;sub&gt;4&lt;/sub&gt; · 7H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>500</td>
</tr>
<tr>
<td>CaCl&lt;sub&gt;2&lt;/sub&gt; · 2H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>10</td>
</tr>
<tr>
<td>Fe&lt;sub&gt;2&lt;/sub&gt;(SO&lt;sub&gt;4&lt;/sub&gt;)&lt;sub&gt;3&lt;/sub&gt; · 6H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>10</td>
</tr>
<tr>
<td>MnSO&lt;sub&gt;4&lt;/sub&gt; · H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>0·01</td>
</tr>
<tr>
<td>Co(NO&lt;sub&gt;3&lt;/sub&gt;)&lt;sub&gt;2&lt;/sub&gt; · 6H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>0·005</td>
</tr>
<tr>
<td>(NH&lt;sub&gt;4&lt;/sub&gt;)&lt;sub&gt;2&lt;/sub&gt;MoO&lt;sub&gt;4&lt;/sub&gt; · 4H&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>0·1</td>
</tr>
</tbody>
</table>

pH 6·8-7·0; Filter sterilized

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The growth of this organism was compared in mineral salts medium with glucose or with n-octane as the carbon source (Table 1). These initial experiments demonstrated an interesting phenomenon which is shown in Figure 2. The growth response with glucose was much better if the culture was shaken than if it was allowed to incubate under static conditions. However, when hydrocarbon served as the sole source of carbon and energy the reverse situation was found. The aeration efficiency of these aerobic cultures was determined by the sulfite method (Cooper *et al.*, 1944) and found to be 0.85 mM O$_2$/L/min for the shaken cultures and 0.52 mM O$_2$/L/min for the cultures incubated without shaking. The superior yield of cells from n-alkane with reduced aeration indicated either there was accumulation of a toxic product under higher oxygen tension or the organism preferred to utilize a non-oxygen requiring system. We do know that this organism is sensitive to high concentrations (0.1 %) of fatty acids and therefore the accumulation of fatty acids from n-octane could be responsible for the lower cell yields.

![Figure 1: Germanium shadowed *Pseudomonas aeruginosa* strain 196Aa.](image-url)

Figure 2: Growth response of *Pseudomonas aeruginosa* on octane (2% u/v) and glucose (1% w/v) under static (---) and shake ( . . . ) conditions, at 30°C.

With the low aeration efficiency demonstrated for the static cultures it was possible that the metabolism of the organism was essentially anaerobic rather than aerobic. The medium contained nitrate (as \( \text{NH}_4\text{NO}_3 \)) which could serve as an electron acceptor and which was shown to disappear from the medium. Table 2 summarizes the growth response obtained when the organism was grown under aerobic and anaerobic conditions on n-octane. The lower the oxygen tension in the culture medium the greater the cell yield, but the greater the time required to reach maximum growth.

The fate of ammonia and nitrate was followed in the aerobic cultures under static conditions. Figure 3 demonstrates that ammonia was used initially by the lag phase and early exponential phase culture. As the cell mass increased and oxygen became limiting in the latter portion of the exponential phase of growth nitrate was rapidly reduced to nitrite. The late increase in ammonia shown in this experiment could be due either to the reduction of nitrite to ammonia or to the endogenous release of ammonia by the stationary phase cells. This point has not been clarified at this time.

**TABLE 2**

Growth yield under different aeration conditions

<table>
<thead>
<tr>
<th>Culture Conditions</th>
<th>Maximum Growth</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Time (hours)</td>
</tr>
<tr>
<td>Shake-aerobic</td>
<td>18</td>
</tr>
<tr>
<td>Static-aerobic</td>
<td>50</td>
</tr>
<tr>
<td>Static-anaerobic</td>
<td>96</td>
</tr>
</tbody>
</table>

Substrate, 2% n-octane; Temperature 30°C; \( K_{LaC} \) in mM \( O_2 \)/liter/minute.

**Figure 3:** Relationship between ammonia concentration (---), nitrite accumulation (----), and growth (-----) of *Pseudomonas aeruginosa* strain 196Aa on n-octane under static conditions of culture at 30°C.

Anaerobic cultures were grown in a National Appliance Company, Model 3640, Anaerobic Incubator. The incubator was evacuated to 28 inches of mercury and back-filled with pure helium gas. The cycle was repeated four times at which point the percentage of oxygen remaining in the chamber was less than 0.0006 per cent. In some experiments nitrogen was used for replacement rather than helium. The growth response was the same with either gas. Anaerobic controls were a tube of *Bacillus megaterium* and *Clostridium bifermotens* placed in the chamber and a tube of decolorized methylene blue. The experiment was not considered valid unless there was growth of the *Clostridium*, no growth of the *Bacillus*, and the methylene blue solution remained colorless. All hydrocarbon cultures were inoculated with 1 ml of a 200 Klett Unit n-octane grown cell suspension. Octane was provided to the organism in the vapour phase and hexadecane added directly to the medium.

Three different organisms have been tested for the capacity to utilize n-alkanes in the absence of gaseous oxygen. *Pseudomonas aeruginosa* strain 196Aa from our laboratory, *Pseudomonas aeruginosa* strain Sol 20 obtained from Dr. J. C. Senez (CNRS, Marseille, France), and *Bacterium 7E1C* obtained from the stock culture collection of the Department of Microbiology, The University of Texas. Organism 7E1C would not grow under anaerobic conditions in our test system but the two pseudomonads gave the growth response shown in Figure 4. The growth with the Sol 20 strain is feeble and may not be considered representative of alkane utilization. The response of the 196Aa strain is satisfactory evidence for growth obtained from an n-alkane under these conditions.

Figure 4: Growth of *Pseudomonas aeruginosa* strain 196Aa (---) and *Pseudomonas aeruginosa* strain Sol 20 (....) on hexadecane under anaerobic conditions obtained with helium.

It is concluded from these results that *Pseudomonas aeruginosa* is capable of utilizing either n-octane or n-hexadecane in the absence of molecular oxygen. Nitrate undoubtedly serves as the terminal hydrogen (electron) acceptor in the respiratory scheme rather than oxygen. Oxygen is probably introduced into the hydrocarbon molecule from water via a hydration mechanism, at a double bond formed by the dehydrogenation of the alkane molecule. Proof of this assumption rests of course on the isolation and identification of intermediates such as an alkene and the demonstration of enzymes capable of carrying out the proposed mechanism. Work is now underway which it is hoped will provide such proof.

References


A note on endogenous and biodeteriorative factors in the respiration of dormant yam tubers.

A NOTE ON ENDOGENOUS AND BIODETERIORATIVE FACTORS IN THE RESPIRATION OF DORMANT YAM TUBERS

Summary. Yam tubers (Dioscorea rotundata) were stored at 25°C until decay or sprouting occurred. Weight and CO₂ output were determined at intervals. The results suggest that loss of dry matter as CO₂ was the major factor influencing weight loss. The tubers could be divided into two groups with low and high respiration rates: the former remained sound while the latter quickly rotted. Decayed tubers were infected with Botryodiplodia theobromae Pat. and, in one case with Penicillium cyclopium Westling.

The tubers of yams, principally Dioscorea rotundata Poir., and D. alata L., are important foods in many tropical countries, the production being of the order of 20 million tons/annum (Coursey, 1967a).

However, little attention has been given to their post-harvest behaviour. Storage methods are generally primitive, and losses are severe, amounting to more than a million tons/annum globally (Coursey, 1967b). These losses appear to arise (a) from the natural metabolic processes of the dormant tubers and (b) from attack by micro-organisms causing decay.

Preliminary respiratory studies on whole D. rotundata tubers have been described (Hayward, 1959; Hayward and Onyeanu, 1962). The first indicates that the carbon dioxide output may vary between zero and 50 mg. CO₂/kg. fresh weight/hour, but the techniques are open to criticism; while the second suggests 5 to 20 mg./kg./hr. Gane and Robinson (unpublished) found rates of the order of 100 mg./kg./hr., but their material was old and highly deteriorated, and this high apparent respiration may have been due mainly to biodeteriogens. The respiration of slices of yam tuber tissue has been studied by Coursey et al. (1966). The organisms responsible for decay in yam have been studied by several workers (reviewed by Coursey, 1967b).

Experiments were conducted using D. rotundata cv. "Labreko" purchased in Accra, Ghana. The material was recently harvested (ca. 3 days) and consisted of "first crop" tubers (i.e., slightly immature tubers, harvested from the growing plant). Ten tubers (weights between 2287 g. and 3786 g., mean 2859 g.) were selected to be as uniform as possible, and free from blemishes or decay.

These were stored at 25°C. The tubers were individually weighed at weekly or fortnightly intervals. Respiratory activity was determined at similar intervals by measuring CO₂ output by absorption in N/10 Ba(OH)₂ and titration with standard HCl. The experiments were continued with each tuber until it either exhibited marked decay, or began to sprout. The results of respiratory measurements together with changes in weight, are given in the Table. Tubers which decayed during the experiment were found to be infected with Botryodiplodia theobromae Pat. (IMI 134625 to 134628) and in one case with Penicillium cyclopium Westling (IMI 134629).

A statistical analysis of the results was made by examining the regressions of weight loss on respiration rate. Each correlation coefficient was found to be significant at the 95% level of confidence, while using all the observations, a correlation coefficient of + 0.746 was obtained, which may be regarded as highly significant.

Loss of dry matter as CO₂ appears therefore to be the major factor influencing weight loss in yams, as has been suggested elsewhere (Coursey, 1961; 1967a; 1967b). This loss of dry matter is accompanied by moisture loss, as the dry matter content of the yams varies little during storage (Coursey, 1961).

---

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(Copy received 19th December, 1968.)
A note on endogenous and biodeteriorative factors in the respiration of dormant yam tubers.
D. G. Coursey and J. D. Russell

Inspection of the results shows that the tubers may be divided between two groups. Group I includes those of comparatively low respiration rates, varying only slowly with time. Group II includes those with high respiration rates, which tended to vary erratically. All the tubers in Group II, although initially sound, quickly became rotten, while those of Group I remained sound until the end of the experiment, when sprouting occurred. Three individual tubers in Group I suddenly developed enhanced, fluctuating respiration rates during the course of the experiment, and, shortly afterwards, became rotten; they are therefore regarded as having moved into Group II (see Figure).

It was noted that respiration rates in excess of about 65 mg./kg./hr. were associated with visible decay.

The marked difference in behaviour exhibited by the two groups of tubers is in agreement with earlier field experiments (Anon, 1937; Coursey, 1961) where it was shown that visible decay is associated with enhanced weight loss. Similar effects are, of course, well known in many types of produce.

Of the two fungi isolated, *B. theobromane* has previously been reported as a storage rot of yams in several parts of West Africa (Coursey, 1967b). *P. cyclopium* has not previously been reported in yams, but is common in association with decay in the hypogenous organs of other Liliales (Raper and Thom, 1949).

Acknowledgements
The collection and supply of the experimental material was arranged by Prof. J. S. Matthews of the University of Ghana.

Mycological isolations were made by Mrs. H. C. Aldridge, and identifications by Mrs. A. Wallbridge; these were confirmed by the Commonwealth Mycological Institute. Statistical analyses were conducted by Mr. R. Pope.

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References
Relationship of respiratory activity of dormant yam tubers to duration of storage.
The full lines represent envelopes to curves of results obtained with individual tubers, enclosing areas I and II (see text). Dashed lines indicate behaviour of three individual tubers which moved from Group I to Group II during the experiment. The horizontal dashed line indicates the level of respiratory activity associated with obvious decay.
Arrow indicates time at which sprouting of the tubers was observed.
### Table 1
Respiratory Activity and Weight Losses on Yam Tubers

| Week No. | 1   | 2   | 3   | 4   | 5   | 6   | 7   | 8   | 10  | 12  | 14  | 16  | 18  | 20  | 22  | 24  | 26  | 28  | 30  | 31  | 32  | 33  | 34  | 35  |
|----------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Tuber No. 1 | Initial Weight 2344 g. | 13·0 | 15·6 | 19·5 | 23·7 | 28·8 | 38·2 | 46·7 | 55·2 | 73·7 | 107·2 | 44·2 | 29·3 | 13·3 | 20·2 | 37·3 | 39·6 | 63·5 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·5 | 99·0 | 98·4 | 97·6 | 97·0 | 96·2 | 95·0 | 92·9 | 90·6 | 88·5 | 86·4 | 84·6 | 82·8 | 80·8 | 78·6 | 75·8 |
| Weight (as % of original) | |
| Tuber No. 2 | Initial Weight 2807 g. | 16·0 | 26·0 | 17·6 | 14·9 | 14·1 | 13·0 | 14·7 | 16·2 | 142·0 | 132·0 | <220·0 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·5 | 98·9 | 98·3 | 97·9 | 97·3 | 96·9 | 96·3 | 95·4 | 94·0 | 92·5 | 90·3 | 87·6 | 84·4 | 81·0 | 77·0 | 73·2 | 71·2 |
| Weight (as % of original) | |
| Tuber No. 3 | Initial Weight 3449 g. | 18·6 | 17·5 | 16·2 | 12·8 | 14·2 | 13·0 | 9·8 | 12·4 | 11·4 | 11·8 | 9·2 | 9·2 | 40·3 | 54·4 | 76·0 | 75·2 |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·3 | 98·7 | 98·4 | 97·9 | 97·4 | 97·0 | 96·6 | 95·8 | 95·2 | 94·3 | 93·5 | 92·6 | 90·9 | 87·7 | 84·2 | 80·8 |
| Weight (as % of original) | |
| Tuber No. 4 | Initial Weight 3005 g. | 17·4 | 54·3 | 58·8 | 49·1 | 29·3 | 39·5 | 50·4 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 98·3 | 96·5 | 94·4 | 92·5 | 90·2 | 87·6 |
| Weight (as % of original) | |
| Tuber No. 5 | Initial Weight 2828 g. | 23·0 | 20·1 | 21·5 | 11·5 | 15·1 | 14·0 | 12·5 | 11·6 | 11·3 | 8·6 | 7·9 | 11·0 | 14·1 | 12·6 | 15·3 | 19·2 | 21·1 | 20·8 | 17·6 | 20·0 | 21·6 | 24·4 | 29·2 |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·3 | 98·7 | 98·2 | 97·6 | 97·2 | 96·8 | 95·6 | 94·7 | 94·0 | 93·4 | 92·2 | 90·3 | 88·1 | 85·4 | 82·6 | 81·0 | 79·6 | 78·0 | 76·8 | 75·4 | 74·2 |
| Weight (as % of original) | |
| Tuber No. 6 | Initial Weight 3787 g. | 19·8 | 12·1 | 14·9 | 13·2 | 11·9 | 11·1 | 11·2 | 11·6 | 9·8 | 11·3 | 9·0 | 8·1 | 9·4 | 11·7 | 13·1 | 16·5 | 19·0 | 20·0 | 17·5 | 26·4 | 20·4 | 22·3 | 24·0 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·5 | 99·0 | 98·5 | 98·1 | 97·9 | 97·6 | 97·3 | 96·6 | 95·3 | 94·2 | 93·6 | 92·8 | 91·4 | 89·4 | 87·3 | 85·0 | 83·8 | 82·5 | 81·4 | 80·4 | 79·1 | 77·8 | 76·7 | 75·6 | 74·6 |
| Weight (as % of original) | |
| Tuber No. 7 | Initial Weight 2420 g. | 12·4 | 15·4 | 15·0 | 12·0 | 11·2 | 10·5 | 10·7 | 9·0 | 8·8 | 8·8 | 7·8 | 8·6 | 8·8 | 10·5 | 12·9 | 18·3 | 19·9 | 21·0 | 17·2 | 18·1 | 20·2 | 20·2 | 23·0 | 27·7 | 23·4 | 22·2 |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·4 | 99·0 | 98·6 | 98·2 | 97·8 | 97·5 | 97·4 | 96·6 | 96·2 | 95·7 | 95·2 | 94·6 | 93·9 | 91·6 | 89·0 | 88·3 | 85·0 | 83·8 | 82·4 | 81·7 | 80·8 | 79·4 | 77·8 | 76·6 | 75·6 | 74·6 |
| Weight (as % of original) | |
| Tuber No. 8 | Initial Weight 2837 g. | 45·5 | 38·3 | 54·2 | 48·7 | 41·0 | 59·0 | 59·3 | 57·0 | 59·0 | 43·9 | 39·4 | 41·2 | 20·0 | 26·1 | 34·9 | 47·3 | 50·4 | 66·6 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 98·5 | 97·1 | 95·6 | 94·0 | 92·6 | 91·1 | 89·9 | 86·5 | 83·3 | 80·0 | 76·5 | 73·0 | 69·4 | 65·8 | 62·4 | 59·1 | 57·7 |
| Weight (as % of original) | |
| Tuber No. 9 | Initial Weight 2625 g. | 40·3 | 34·3 | 32·7 | 33·6 | 33·5 | 40·4 | 41·4 | 43·9 | 47·0 | 32·1 | 33·6 | 44·4 | 68·0 | 60·7 | 53·2 | 49·6 | 48·8 | 64·6 | Discontinued |
| Respiratory Rate (mg CO₂/kg/hr.) | 100 | 99·0 | 98·0 | 97·2 | 96·3 | 95·3 | 94·5 | 93·3 | 91·3 | 89·1 | 86·9 | 84·5 | 81·6 | 78·0 | 74·3 | 70·7 | 67·2 | 65·4 |
BOOK REVIEWS

BIODETERIORATION OF MATERIALS. MICROBIOLOGICAL AND ALLIED ASPECTS
Edited by A. Harry Walters and John J. Elphick.


This book records the proceedings of the 1st. International Biodeterioration Symposium held in September 1968, and I must first indicate my pleasure at being able to review this book while the Symposium itself remains fresh in the memory. The editors are to be congratulated for organising a very large amount of material—67 separate papers in all—in so short a time. An editorial note apologises for any errors which may have slipped through due to rapid processing and at the same time pays tribute to the publishers and printers. The apology is hardly necessary, the tribute well earned, for this book has been-produced to a very high standard and is pleasant to handle and easy to read.

Several other features of this book which may be attributable to editorial or publishing policy make the contents easy to assimilate. All the contributions are commendably brief, none exceeds ten pages of text if tables and illustrations are excluded. Within a brief space each manages to include a great deal of material and a ready understanding of this material is greatly assisted by a uniform policy of splitting each paper into short subsections. Each such sub-section is clearly headed so that the content of the article can be assessed at a glance. Where further subdivisions are necessary these too are clearly picked out in heavy type. A second feature which assists both understanding and brevity is the reproduction of large numbers of tables and diagrams, together with appreciable numbers of photographs which are particularly valuable where topics or materials unfamiliar to the majority of readers are dealt with. In short the overall effect is that of a well produced textbook, and this cannot be said of many symposium proceedings.

From the point of view of content this book is a very valuable summary of the sort of problems faced, and the sort of work being done in biodeterioration today. Its 700-odd pages contain a great deal of information and the contents list which accompanies this review shows the width of the field covered. A by-product of the large number of relatively short papers is the large number of references which they cite when taken together. The result is a comprehensive and up-to-date bibliography of biodeterioration work. While not detracting from the great value of this volume however, I do question whether it fully achieves the aims of the Symposium Organising Committee. From the number of articles and the wide interests covered, I get the impression that the intention was to produce a comprehensive account of biodeterioration. This was a very sound aim in view of the recent establishment of biodeterioration as a separate discipline. A comprehensive account would serve both as an introduction for workers in allied disciplines, and as a statement of the 'state of the art' for workers in biodeterioration. However I question whether the balance and arrangement of papers in this book will give a correct impression of biodeterioration to outsiders, and whether there are sufficient papers attempting to increase the awareness amongst biodeteriorationists of the significance of some aspects of fundamental microbiology to their work.

Taking the point of balance first. The symposium adopted the policy of having longer contributions at the morning sessions outlining large areas of biodeterioration work (Parts I, II and V) with supporting detail in the shorter contributions of the afternoon. The balance of the longer review articles is about right, fairly reflecting the areas of interest in biodeterioration. Two of them particularly (Dr. Iverson on the microbial corrosion of metals, and Drs. Traxler and Flannery on hydrocarbon degradation) provide excellent brief summaries of topics which are not easily available elsewhere. Dr. Hueck's appraisal of the whole field is valuable in setting the tone of the symposium, but one or two of the others perhaps put too much emphasis on theoretical aspects of their subject. Dr. Selby's account of cellulose biodegradation on the other hand, by dealing with a more restricted field than the other reviews, incorporates a very full account of practical results in this field.

It is in the other sections which cover the afternoon sessions that I feel an unbalanced view may be presented to the outsider. Aspects of timber decay occupy almost a quarter of the book, whilst consideration of all other materials occupies only one third. The account of timber decay is admittedly a good one. The section on ecological aspects of biodeterioration, and the section on enzymes and physiology of wood-destroying organisms, taken together with Dr. Selby's article, provides an excellent account of the subject. I feel that a similar comprehensive coverage could have been given to many other topics, such as hydrocarbon microbiology, if space had allowed. The fact that such coverage has been given only to timber may create the impression in an outsider that biodeterioration work is still mainly orientated towards traditional natural materials.

It is in these sections too that I feel the arrangement could be improved. It is unfortunate that no attempt was made in publication to separate the two styles of article which are included in these afternoon sessions. Most such contributions can be classified either as authoritative reviews of a limited area of work—for example deterioration problems in specific industries, or in the use of particular materials—or as detailed accounts of personal results. The inclusion of the latter with the review type of article perhaps gives these particular pieces of work undue emphasis. This is particularly true where a review of deterioration of paint and paint films is followed.
The biodeterioration of materials—an appraisal distinct and legitimate branch of science. As a result the comment 'Biodeterioration! What's that?' may be Jess.

The role of Government in international co-operative research mechanisms of biodegradation of cellulose mechanisms of marine fouling ecological aspects of biodeterioration aspects of taxonomy with versatility to allow them to intrude significantly in the most unexpected situations. Finally, for those already in and textbooks. It is obviously involved here, but I feel that one or two topics such as these might well have replaced some of the research papers since the tenor of this symposium is slanted toward a review treatment anyway.

In one other respect I would question the choice of contents. The title of this book bears the subscript 'microbiological and allied aspects' and the latter seem to be represented by marine fouling. While I do not question the status of marine fouling as an important aspect of deterioration I do wonder how closely it is allied to microbiology, if one considers the relevance of results with microorganisms to marine fouling, and vice versa. Certainly the paper of Drs. Dolgopolskaya and Gurevich relates fouling to the growth of bacterial slime, but the other papers in this section seem to have more in common with invertebrate biology, an impression confirmed by the references they cite. There is a large sector of biodeterioration concerned with damage caused by invertebrates, excluded in this symposium but perhaps to be considered in the future. It is surely with this sector of biodeterioration that marine fouling is more naturally allied.

Despite these criticisms this symposium has produced a very valuable book, and one which I welcome as perhaps the first book to describe the whole field of microbial degradation since 'The Deterioration of Materials' by G. A. Greathouse and C. J. Wessel, published in 1954. As such it is to be hoped that it will come to the notice of workers outside the immediate field of biodeterioration. General microbiologists may be surprised at the extent of this field of applied microbiology which is given scant recognition in the usually medically orientated courses and textbooks. It may also come as a surprise to some engineers and chemists that microorganisms show sufficient versatility to allow them to intrude significantly in the most unexpected situations. Finally, for those already in the field of biodeterioration, the production of such a professional volume may help to establish their work as a distinct and legitimate branch of science. As a result the comment 'Biodeterioration! What's that?' may be less often heard in the future.

J. Walsh.

CONTENTS

PART I FUNDAMENTALS OF BIODETERIORATION
The role of Government in international co-operative research
J. KNOX (U.K.)
The biodeterioration of materials—an appraisal
H. J. HUECK (NETHERLANDS)
Aspects of taxonomy with respect to biodeterioration
T. G. MITCHELL and J. M. SHEWEN (U.K.)
Ecological aspects of biodeterioration
H. O. W. EGGBS (U.K.)

PART II MECHANISMS OF BIODETERIORATION
Mechanisms of microbial corrosion
W. F. IVESON (U.S.A.)
Mechanisms of hydrocarbon degradation
R. W. TRAXLER and W. L. FLANNERY (U.S.A.)
Mechanisms of marine fouling
D. R. HOUGHTON (U.K.)
Mechanisms of biodegradation of cellulose
K. SELBY (U.K.)

PART III
ECOLOGICAL ASPECTS OF BIODETERIORATION
Factors in establishing microbial populations on biologically inert surfaces
R. A. RASMUSEN, R. S. HUTTON and R. J. GARNER (U.S.A.)
Protection of sensitive components from microbial contamination
M. H. BENGSON and J. R. GILLIS (U.S.A.)
Microbiological deterioration of materials in deep mines
B. J. ZYSKA (POLAND)
Some techniques to investigate the colonization of cellulosic and wood substrates
Fungal spora of the air at the Joint Tropical Research Unit, Innisfail, Queensland
F. J. UPSHER (AUSTRALIA)
The selective isolation of Sphaerotilus
J. M. SHARPLEY (U.S.A.)
32
PART IV
TESTING FOR BIODETERIORATION RESISTANCE

Laboratory and service tests of PVC
G. C. YEAGER (U.S.A.)

A rapid method of determining degradation of plasticized PVC by micro-organisms
J. J. CAVETT and M. N. WOODROW (U.K.)

The evaluation of rot resistance of cellulose textiles
A. O. LLOYO (U.K.)

L'expérience des normes Françaises d'essai de résistance des matériaux contre la dégradation par les microorganismes. Avantages des cultures mixtes de champignons dans les essais d'inoculation
Y. LE GRAND (FRANCE)

Some Problems posed by quality screening for biodeterioration
EUNICE S. L. JONES (U.K.)

PART V CONTROL OF BIODETERIORATION

The control of biodeterioration by fungicides—philosophy
A. M. KAPLAN (U.S.A.)

Protection of timber
G. GARGANI (ITALY)

Prevention of biodeterioration by control of environmental conditions
G. AYERAT (I.R.A.C.)

Biodeterioration test methodology
Q. WÄCHTL (SWITZERLAND)

PART VI DETERIORATION AND PROTECTION OF MATERIALS

Section 1

Fungus contamination of Florence art-masterpieces before and after the 1966 disaster
G. GARGANI (ITALY)

Facteurs Biologiques de l'alteration des Pierres
J. POCHON (FRANCE)

The microbiological deterioration of cosmetics and pharmaceutical products
N. J. BUTLER (U.K.)

Microbiological deterioration in the paper, printing and packaging industries
R. L. HUGOES (U.K.)

Infections of industrial waters
E. E. GARGO (NETHERLANDS)

Biodeterioration in the leather industry
A. ORLTA (CZECHOSLOVAKIA)

Section 2

Investigations into the effects of micro-organisms on PVC pressure-sensitive adhesive tape and its constituents
EILEEN E. FANKHURST and MARGARET J. DAVIES (U.K.)

Biodeterioration of paint and paint films
R. T. ROSS, J. R. SLADEN and L. A. WIENART (U.S.A.)

Fungicides in latex paints
R. ENNONGA and W. J. BORDS (NETHERLANDS)

The anti-microbial and rot-proofing properties of cotton textiles containing the cadmium salts of modified cellulose
L. B. QUIENSEL (U.K.)

PART VII HYDROCARBON MICROBIOLOGY AND METALLIC CORROSION

Metal-organic acid corrosion and some mechanisms associated with these corrosion processes
G. H. CALDERON, E. L. STAFFELOT and G. B. COLEMAN (U.S.A.)

Evaluating biocidal fuel additives for intermittent use in aircraft fuel systems
J. J. ELPHECK and S. R. P. HUNTER (U.K.)

The soil as a natural source of Cladosporium resinae
D. G. FARRERY (AUSTRALIA)

Microbial degradation of lubricant oils and emulsions and its engineering significance
E. C. HILL (U.K.)

Control of cutting oil deterioration with gamma radiation
H. ROSSMOORE and J. O. BRAZIN (U.S.A.)

PART VIII ECOLOGICAL ASPECTS OF TIMBER DECAY

Microbial attack of timber and allied constructional materials
J. G. SAVORY (U.K.)

Ecology of decay fungi in birch and aspen pulpwood
H. HUNNINGHSON (SWEDEN)

Studies on the ecology of fungi in wooden fence posts
J. F. LEVY (U.K.)

Microbial associations in the deterioration of wood under long term exposure
J. G. GRAVES and J. F. LEVY (U.K.)

Ecology of fungi infecting untreated and preservative-treated sapwood of Pinus radiata D. Don
J. A. BUTCHER (NEW ZEALAND)

The distribution of marine fungi on wood submersed in the sea
B. GARETH JONES (U.K.)

PART IX BIOCIDES

Fungitoxic action of nonmetallic organic fungicides
R. J. LUCKENS (U.S.A.)

Action mechanisms of some organometallic preservatives
R. A. RICHARDSON (U.K.)

Biocide development—the manufacturer's problems
G. A. THOMAS (U.K.)

Some data on the relationship between fungicidal protection and pH
J. M. C. WEVERS and D. M. M. ADEMA (NETHERLANDS)

The role of cell membrane permeability in determining the antifungal activity of 2, 4, 6-trichlorophenol at pH 6 and pH 8
P. A. WOLF and M. M. SCHAEFFER (U.S.A.)

Investigations on the biological activity of pentachlorophenol esters
H. J. HUECK, J. LA BRUN, J. A. COPPER and J. VAN HAM (NETHERLANDS)

PART X ENZYMES AND PHYSIOLOGY OF WOOD-DESTROYING ORGANISMS

Some aspects of cellulose degradation in lignified cell walls
P. J. STRIEF, W. LIEZ and R. BÖSch (GERMANY)

Degradation of wood-cell components by extracellular enzymes of Coniophora cerebelle
N. J. KING (U.K.)

Lignolytic action of Schizophyllum commune
L. JURASEK (CZECHOSLOVAKIA)

Influence of extractives on cellulose and xylanase activities of Schizophyllum commune
R. SOKRO (CZECHOSLOVAKIA)

The effect of carbon to nitrogen ratio of substrate on the growth, composition, cellulase production, and wood-degrading capacity of Polyergus versicolor
M. P. LEVY and E. R. COWLING (U.S.A.)

Growth of selected cellulolytic fungi on wood pulp
D. S. CHAHAL and D. GRAY (INDIA)

Degradative activities on filamentous marine fungi
S. P. MEYERS (U.S.A.)

PART XI MARINE FOULING

Some factors affecting the underwater testing of wood-resisting antifouling paints
C. E. PEARSON (U.K.)

The prevention of fouling by localized chlorine generation
T. LOVEGROVE and T. W. ROBINSON (deed.) (U.K.)

Studies on possible mechanisms for the control of moulding and metamorphosis in barnacle larvae
A. M. MORTLOCK (U.K.)

Marine boring and fouling organisms in open water off Monterey Bay, California
E. C. HARDERLE (U.S.A.)

Biological and physicochemical factors influencing the efficacy of antifouling paints
M. A. DOLGOFELSAYA and E. S. GURVICH (U.S.S.R.)

PART XII STORED PRODUCTS MICROBIOLOGY

Effect of biocidal residues on wine making
A. M. ADAMS (CANADA)

A new look at mouldy cocoa
C. E. BROADBENT and J. O. GYENINRA (NIGERIA)

Biodeterioration of groundnut oil by Aspergilli
R. J. TOMLESS and R. J. TOWNSEND (U.K.)

Biodeterioration of harvested sugar cane
R. H. TILBURY (U.K.)

Méthode d'appréciation de la détérioration biologique des grains par mesure du Temps Moyen de Germination
J. L. MULTON, P. LECOURSE and A. GIULIOT (FRANCE)
Biodeterioration of Materials

Microbiological and Allied Aspects

Edited by
A. HARRY WALTERS and JOHN J. ELPHICK

This volume contains full reports of the 67 papers presented at the First International Biodeterioration Symposium, held at Southampton in September 1968, and organized in association with the Society of Chemical Industry and the Organization for Economic Development and Cooperation to consider all aspects of biological action leading to the deterioration of materials, products or processes of economic importance.

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CONTENTS

Part I Fundamentals of Biodeterioration
Part II Mechanisms of Biodeterioration
Part III Ecological Aspects of Biodeterioration
Part IV Testing for Biodeterioration Resistance
Part V Control of Biodeterioration
Part VI Deterioration and Protection of Materials
Part VII Hydrocarbon Microbiology and Metallic Corrosion
Part VIII Ecological Aspects of Timber Decay
Part IX Biocides
Part X Enzymes and Physiology of Wood-destroying Organisms
Part XI Marine Fouling
Part XII Stored Products Microbiology

RADIATION PRESERVATION OF FOODS

Edited by Edward S. Josephson and J. Harry Frankfort


This volume performs a useful function in drawing together specialised information that would otherwise be lost in the very wide selection of journals publishing work on food irradiation. This collection of research papers, the proceedings of a conference, is not suitable for the reader requiring an introduction to the subject. It does not indicate whether commercial exploitation is imminent or wholesomeness problems overcome, subjects which one might expect to find discussed in a book with such a general title.

There are 6 papers on various aspects of dosimetry, sources, general engineering and design matters; 4 papers on the irradiation of beef; 3 on fruit or vegetables; 1 on fish and one paper on radiation effects in aqueous solutions of polyamino acids. Seven of the papers originated from or were written by scientists financially supported by the U.S. Army Natick Laboratories with a further three papers from other U.S. Government Laboratories.

The very impressive irradiation facilities at the Natick Laboratories are described in relation to the main Army effort to provide sterile meat for the armed services which needs no refrigeration "yet resembles fresh food when prepared." The U.S. Atomic Energy Commission program is also described and a paper on gamma irradiator design completes a trio of reports reiterating much material already published.

The main problem of using this technique to sterilise meat is the severe off-odour and off-flavour induced by radiation treatment. One paper describes the effect of different temperatures during irradiation on model amino acid and dipeptide systems. The production of carbonyl compounds and mercaptans was less at -196°C than at higher temperatures. Irradiation of beef steaks resulted in organoleptically satisfactory products if irradiation was performed at -196°C. Chemical findings and organoleptic results could not be correlated. Two other investigations, in which gas chromatographic techniques were used, showed that over 30 compounds of both protein and lipid origin could be isolated from the volatile material from irradiated meat. Amino acid destruction in beef was examined and cystine was found to be the most sensitive residue. The authors of this paper also describe
what they claim is a difference in amino acid destruction due to differences in the energy level of the radiation. The most interesting observation from the experiments on irradiated poly a-L glutamic acid is the optical inversion to D-glutamate particularly when oxygen was absent. The paper on radiation pasteurisation of fish and shellfish outlines suitable processing conditions for a number of species and also considers the question of survival and growth at various storage temperatures of resistant bacteria. A number of biochemical points are also considered including the interesting topic of enzyme irradiation by radiation. A good paper on surface treatment of spherical objects, the example taken being the orange, and a paper on biochemical changes in irradiated fruit and vegetables completes the volume. There is an adequate index.

This volume should be in the library for reference, but it does not add greatly to what is already known about the radiation preservation of foods.

G. Glew.

INSECT AND MARINE BORER DAMAGE TO TIMBER AND WOODWORK: RECOGNITION PREVENTION AND ERADICATION
J. D. Bletchley

This publication replaces an earlier Forest Products Research Laboratory bulletin which was called "Beetles Injurious to Timber and Furniture." It deals briefly with the biology of wood-boring insects and marine borers and describes the organisms and the damage that they cause. Emphasis is laid on the need to be able to recognise the cause of damage from the appearance of the infested wood, since the insects themselves are often absent or hard to find. A useful feature of the book is the inclusion of a table summarising for easy reference the diagnostic characters of damage caused by the commoner wood-boring insects and marine borers. It is also helpful to have, in an appendix, descriptions of insects likely to be mistaken for wood-boring species, such as plaster beetles, flour beetles and carpet beetles. Remedial treatments for existing infestations are given after each insect, and there is a whole chapter on protection of timber and woodwork, dealing mainly with treatments with chemical preservatives.

Another chapter deals briefly with the inspection of buildings with particular reference to churches; and a final chapter deals with the eradication of wood-boring insects by (1) fumigation with toxic gases, (2) heat, (3) liquid insecticides, and (4) insecticidal smokes.

The text is well arranged and clearly written. It is splendidly illustrated with quite excellent photographs both of the insects and of the damage that they cause.

The very large format (30 × 21 cm.) is inconvenient for a book of reference which might have to be taken around by an inspector. This size is quite unnecessary as there are no full page photographs or plans. The layout also seems needlessly extravagant—on page 63, for instance, two small photos are tucked up in one corner, occupying only about 15% of the page. Possibly this is one reason for the very high price (35/-) which is some 23 times more than the 1940 bulletin.

W. P. K. Findlay.

INSECTS AND HYGIENE
J. R. Busvine

Since its publication in 1951, Professor Busvine's work has been the standard textbook on insect pests of medical and domestic importance in Britain and similar temperate countries, a valuable source of reference to all those, in the Public Health and other fields, in any way concerned with insects and their more direct associations with man. A glance at the references, listed at the end of each chapter of this second edition, will show how thorough has been the task of revision; 50 per cent of those cited are post-1951, whilst for the chapter on chemical control measures, which has been completely re-written, the figure is 80 per cent.

The first chapter introduces the subject of insects as pests of hygiene, and includes some interesting, and amusing, historical notes. It is followed by three chapters dealing with the structure and classification of insects (and Arachnids), their anatomy and physiology, and their ecology.

The subject of control is introduced by a discussion of the organisation of preventive and control measures, incorporating useful summaries of courses of technical information and legislation in Britain relating to domestic insect pests. There follows a short account of mechanical, physical and biological control measures such as screening, hot air disinfection, asphyxiants, dehydrants, electrocution and the more recent techniques of γ-radiation and sterilisation procedures based on radioactivity and the use of chemosterilants.
The most noteworthy feature of the new edition is the revised chapter on chemical control measures the introduction to which includes a useful list of commercial designations of insecticides and their B.S.I. equivalents. Detailed information on pesticides is presented under the headings stomach poisons, contact poisons, fumigants, chemosterilants, repellents and attractants. Practical considerations follow, with a discussion of insecticide application in which details are given of equipment and formulations, and the chapter concludes with sections on insecticide resistance and toxic hazards of pesticides.

The remaining chapters, representing well over half of the book, are devoted to a detailed account of the characteristics, life histories and control of specific pests, arranged according to habit. Of particular interest to workers in the field of biodeterioration will be those chapters dealing with houseflies and blowflies, pests (insects and mites) of foodstuffs, insect pests in waste products, clothes moths and carpet beetles and wood boring beetles.

The book concludes with a “Biological Appendix”, the larger part of which is composed of simple keys for the identification of many of the pests described in the foregoing chapters, both in the larval and adult stage.

A book as wide in its scope and wealth of detail as this would be unique if it were completely free of errors, and some have been noted, albeit mostly of a minor nature as, for example, “carbonate”, not carbamate, with reference to the secondary constituent of the fumigant tablets from which phosphine is liberated (p. 110), “mercaptor” for mercator (p. 310), “Ephestia and Anagaster” for Ephestia and Anagaster (p. 321), and (uncorrected from the first edition) “Antheribidae” for Anthribidae (p. 455).

In the introductory remarks on fumigants (p. 108) the reader is confused by the statement that “they do not normally leave residues and therefore give no protection from re-infestation”. The latter part of this statement indicates, correctly, that a fumigation treatment will impart no residual pest control effect, but it is well known that it may leave residues as, for example, the inorganic bromide detectable in stored foodstuffs following fumigation with methyl bromide.

In the chapter on pests of foodstuffs, information on the Tropical Warehouse Moth, Cadra cautella is wrongly inserted under Oecophoridae (p. 321), although the species is first mentioned, correctly, as a member of the family Phycitidae (p. 319).

Additional couplets have been inserted at the beginning of the key to adult beetles (p. 454) to include the Cerambycid, Hylotrupes and the Curculionids, Euophryum and Pentarthrum, and it is unfortunate that the author has failed to amend three of the couplets which follow; for example, couplet 5 should lead to couplets 6 and 8 (not 3 and 5), the first part of couplet 13, Ptinidae, to couplet 14 (not 11) and the second part of couplet 28 to couplet 30 (not 27).

These are not serious criticisms, however, and do not detract from the value of a book which is both highly informative and well produced; it is well worthy of a place on the shelves of any reference library.

P. F. Prevett.
GRAY, T. R. G. & BAXBY, P.
Chitin decomposition of soil. II. The ecology of chitinoclastic micro-organisms in forest soil. [Fungi. Bacteria. Actinomycetes.]

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Biodeterioration in the leather industry. [Fungi.]

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