Über das Vorkommen von Pristan in der Anisfrucht

In den reifen Früchten von *Pimpinella anisum* L. haben wir Pristan (2,6,10,14-Tetramethylpentadecan) neben Squalen nachgewiesen. Pristan ist hiermit erstmals in einer Pflanze aufgefunden worden. Innerhalb der Familie der Umbelliferen war das Vorkommen von Squalen bisher nur in Blättern und Wurzeln von *Daucus carota* L. bekannt[1].

Die Isolierung des Pristans erfolgte aus dem Unverseifbaren des Hexanextraktes. Auch im unverseiften Hexanextrakt ist es vorhanden, so dass ein Entstehen des Pristans als Artefakt beim Aufarbeiten des Unverseifbaren mit Sicherheit ausgeschlossen werden kann. Aus dem Unverseifbaren werden zunächst die *n*-Alkane als Harnstoffaddukte entfernt. Der Rückstand wird anschliessend über eine Kieselgelsäule chromatographiert, wobei Petroläther eine Fraktion, bestehend aus Anethol, Squalen und Pristan, eluiert. Hieraus werden Squalen und Pristan als Thioharnstoffaddukte[2] ausgefällt. Nach Regeneration der beiden Kohlenwasserstoffe wird Squalen als Hexahydrochlorid[3] abgetrennt. Pristan haben wir durch Gas-Flüssig-Chromatographie an zwei Säulen unterschiedlicher Polarität, durch Mischchromatogramme mit authentischer Substanz, durch das IR-Spektrum, die Dichte und den Brechungsindex identifiziert.

TSUJIMOTO[4] fand diesen gesättigten Kohlenwasserstoff erstmals neben Squalen im Leberöl des Haies *Cetorhinus maximus* Gunner. TOYAMA[5] bezeichnete ihn als Pristan. Pristan kommt ausser in verschiedenen Haien[6,7] noch in Walen[8], Heringen[9] und anderen Seefischen[10,11] vor. Im Lebensraum des Meeres ist Pristan ferner in Plankton-Crustaceen enthalten[12]. Es ist auch Bestandteil der Lipidfraktion des Seewassers[13]. In verschiedenen Erdölen[14,15] und fossilen Sedimenten[16] wurde es nachgewiesen. MOLD et al.[17] berichten erstmals über das Vorkommen von Pristan im Wollwachs als einem Stoff nicht marinen Ursprungs.

Pristan zeigt, was die Anordnung der Methylgruppen anbetrifft, Übereinstimmung mit dem Chlorophyllbestandteil Phytol. Es könnte daher ebenso wie der nächst höhere homologe methylverzweigte Kohlenwasserstoff Phytan (2,6,10,14-Tetramethylhexadecan) aus dem ungesättigten Diterpenalkohol entstanden sein. Ähnliches gilt möglicherweise für die 2,6,10,14-Tetramethylpentadecansäure, die HANSEN[18] im Butterfett aufgefunden hat.

Summary. Pristane (2, 6, 10, 14-tetramethylpentadecane), as well as squalene, was found in a hexane-extract of Anise fruit (*Pimpinella anisum* L.). This is the first case in which pristane has been reported in the plant kingdom.

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Effects of Light, Temperature and Nutrients on the Production of Conidia and Sclerotia by Forms of *Aspergillus japonicus*

Aspergillus japonicus Saito (previously reported as *Aspergillus luchuensis* Inui) has been commonly isolated from oil palm produce in Nigeria[1]. Under ordinary maintenance conditions it was observed that the proportions of Conidia and Sclerotia varied from culture to culture. Investigating this phenomenon with cultures of a strain of the fungus grown at 25°C on a 2% agar medium containing 0.3% malt extract, 0.3% yeast extract, 0.5% mycological peptone, 1% glucose, and 0.2% potassium dihydrogen phosphate, it was found that the cream coloured Sclerotia were formed only in the dark, whereas the purple-brown Conidia were produced when a light stimulus was given. Furthermore, this phenomenon only occurred over the temperature range 22–30°C; between 18 (minimum for growth) and 22°C, and between 30 and 36°C (maximum for growth), only the purple-brown Conidia were formed, either in the light or dark.

At 25°C, 1 foot candle of constant illumination was found to be sufficient to inhibit sclerotial production, Conidia being formed. At the same temperature, and with 10 foot candles intensity, green and blue light inhibited sclerotial production, while red and orange filters removed the inhibiting effect from light.

Sectors appeared frequently in the cultures; two of these, 8M1 and 8M2, were isolated and investigated further. At the above temperature and concentration of nutrients, no light conditions were found which would induce 8M1 to produce Sclerotia, while abundant Conidia were formed under all light conditions and also in the

[1] D. G. COURSEY and H. O. W. EGGINS, Oléagineux *16*, 541 (1961).

dark. Strain 8M2 was found to produce Sclerotia in the dark over a wider range of temperature than the parent strain (20–33°C) and even a few in the light at 25°C. Outside these temperatures, however, only Conidia appeared, although their production was far less abundant than in strains 8 and 8M1 under similar conditions.

Cultures of the parent strain 8, and the variants 8M1 and 8M2 were also grown in the light and dark at 25°C on agar media with varying concentrations of glucose and mycological peptone. Under conditions of low glucose and peptone concentrations all three strains produced some Conidia in either the light or dark, but no Sclerotia. High concentrations of glucose and peptone were conducive to the production by all three strains of Conidia in the light and of Sclerotia in the dark; however, 8M2 still produced far fewer Conidia in the light than 8 or 8M1, and 8M1 produced only a few Sclerotia in the dark, compared to their abundant production under these conditions by 8 and 8M2.

Thus by varying simple cultural conditions it is possible to induce the parent strain to produce colonies with only Conidia, or Conidia and Sclerotia, or only Sclerotia; furthermore, variants have been easily selected in which the ability to produce principally either Conidia or Sclerotia has predominated. It appears that this fungus could provide a valuable tool for morphogenetic studies.

Zusammenfassung. *Aspergillus japonicus* bildet je nach den Belichtungs-, Temperatur- und Ernährungsbedingungen, unter denen er wächst, entweder nur Konidien oder nur Sklerotien oder beide zusammen.

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Fungi on the Surface of Legume Root Nodules and Phosphate Solubilization

While the importance of fungi on the root surface of plants has been recognized[1–3], the presence of fungi such as *Cephalosporium* sp., *Alternaria* sp., *Aspergillus* sp., and *Penicillium* sp. on the surface of root nodules of some legumes, and the antibiotic activity of some of them towards rhizobia, has recently been reported[4]. When *Cephalosporium* sp. and *Alternaria* sp. were screened for antibiotic activity towards phosphate-dissolving bacteria, it was found that the culture filtrate of *Cephalosporium* sp. inhibited the growth of four species of phosphate-dissolving bacteria on solid agar medium. As a natural corollary to this finding, the extent to which the fungus affected the phosphate-dissolving ability of these bacteria was determined by allowing the bacteria to interact individually with *Cephalosporium* sp. on a liquid substrate containing 5 g of Ca_3PO_4/l[5]. The solubilized

Table I. Effect of *Cephalosporium* sp. on phosphate-dissolving ability of bacteria

Treatment	mg P_2O_5 solubilized/ 50 ml medium (average of 4 replicates)	mg P_2O_5 solubilized over control	% P_2O_5[a] solubilized
Non-inoculated medium (control)	11.69	–	–
Bacillus circulans	18.58	6.89	6.70
B. circulans + *Cephalosporium* sp.	15.47[b]	3.78	3.68
Escherichia freundii	20.49	8.80	8.56
E. freundii + *Cephalosporium* sp.	18.45[b]	6.76	6.58
Bacillus megatherium var. *phosphaticum* (isolated from phosphobacterin, a bacterial fertiliser from USSR)	19.24	7.55	7.34
B. megatherium var. *phosphaticum* + *Cephalosporium* sp.	18.31	6.62	6.44
B. megatherium (Indian strain)	17.92	6.23	6.06
B. megatherium (Ind.) + *Cephalosporium* sp.	18.12	6.43	6.25
Cephalosporium sp.	18.32	6.63	6.45

[a] Figures obtained after deducting the amount of phosphorus solubilized due to autoclaving in the non-inoculated control medium.
[b] Significant inhibition. S.E. 0.708; C.D. at 5% level is 2.04.

Table II. Phosphate-dissolving ability of fungi on the surface of legume root nodules

Fungi	mg P_2O_5 solubilized/ 50 ml medium (average of 4 replicates)	mg P_2O_5 solubilized over control	% P_2O_5[a] solubilized
Non-inoculated medium (control)	11.92	–	–
Cephalosporium sp.	18.22	6.30	6.14
Alternaria sp.	18.98	7.06	6.88
Aspergillus sp.	18.67	6.75	6.58
Penicillium sp.	74.50	68.58	61.01

[a] Figures obtained after deducting the amount of phosphorus solubilized due to autoclaving in the non-inoculated control medium. S.E. 1.09; C.D. at 5% level is 3.28.

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