

P E R S O O N I A

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UBIQUINONES IN SELECTED SPECIES OF *PENICILLIUM* AND RELATED TELEOMORPH GENERA

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The ubiquinone type of 38 species (41 strains) of *Penicillium* sensu lato and related teleomorphic genera (*Eupenicillium*, *Talaromyces*, *Thermoascus*) was determined by RPPC or RPHPLC. In eight of the ten sections of *Penicillium* ubiquinone Q-9 was found as the main type. In sect. *Geosmithia* Q-10 and traces of Q-9 were demonstrated. In sect. *Biverticillium* (= sect. *Simplicium*) and its teleomorph *Talaromyces* Q-10(H₂) was demonstrated; in one species an additional unidentified ubiquinone was found. *Thermoascus* has ubiquinone Q-9. The results and those of earlier authors support the idea that the genus *Penicillium* in the actually accepted broad concept is heterogeneous.

The main components of ubiquinone (coenzyme Q) systems have been shown to be interesting tools in the taxonomy of microscopic fungi, in particular of yeasts and yeast-like fungi (de Hoog & al., 1987). Relatively few information is available concerning ubiquinone types in filamentous fungi. Concerning *Penicillium* sensu lato, the scarce data published by Raman & al. (1965), Lavate & al. (1965), Law & al. (1971), Kuraishi & al. (1985, 1990) and Kreisel & Schubert (1990) show that different ubiquinone types occur in this genus and correlated teleomorphs. Therefore a number of additional strains and species has been analyzed.

MATERIALS AND METHODS

Forty-one strains of *Penicillium* and related teleomorphs, representing 38 species, have been taken from the culture collection of Biology Section, Ernst Moritz Arndt University of Greifswald (SBUG). Origin of these strains is mentioned below together with the results. Determination of all analyzed strains has been done or verified by H. Kreisel.

The ubiquinone analyses have been carried out by M. Schubert with the methods applied by Yamada & Kondo (1973) and Kreisel & Schubert (1990) respectively. The fungi have been cultured in surface cultures on liquid medium. The mycelia were saporified with pyrogallol and methanol, extracted with petrolether, the ubiquinones isolated and cleaned on silicagel plates and extracted with acetone.

In most cases the ubiquinone type was identified by reverse phase paper chromatography (RPPC) and by comparison with reference substances extracted from mycelia of *Galactomyces geotrichum* (Q-9), *Aspergillus fumigatus* (Q-10), *Aspergillus flavus* (Q-10(H₂)) and with ubiquinone Q-10 from Merck.

In some cases, the ubiquinone types were identified by high performance liquid chromatography (RPHPLC), using an HPLC equipment LC 1084 B (Hewlett-Packard) and ap-

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plying 65 vol-% methanol and 35 vol-% n-butanol as motile phase. This combination has been found useful for this purpose by Th. Jira who carried out the determination by RPHPLC and revealed superior to 65 vol-% methanol and 35 vol-% iso-propanol as recommended by Nakase & Suzuki (1986).

RESULTS

The following survey presents name, number, and origin of the investigated strains in alphabetical order, followed by the ubiquinone type determined by RPPC (PC) and/or RPHPLC (LC).

Strain	Ubiquinone type
<i>Eupenicillium euglaucum</i> (van Beyma) Stolk & Samson, Anam. <i>Penicillium citreonigrum</i> Dierckx, SBUG M-915, isolated 1988 by F. Schauer from soil contaminated with butyl caoutchouk, Berlin	Q-9 PC
<i>Eupenicillium limoneum</i> Gochenaur & Zlatner, Anam. <i>Penicillium lagenae</i> (Delitsch) Stolk & Samson, SBUG M-997, ex CBS 382.64, isolated by M. Christenen from soil, USA (type strain of <i>Monocillium humicola</i> var. <i>brunneum</i>)	Q-9 PC
<i>Penicillium arenicola</i> Chalabuda, SBUG M-994, ex CBS 220.66, type strain from pine forest soil, Soviet Union	Q-9 PC
<i>Penicillium aurantiogriseum</i> Dierckx, SBUG M-275, isolated 1943 by Müller from lemon, Karlsruhe	Q-9 PC
<i>Penicillium brasiliense</i> Batista (= <i>P. simplicissimum</i> sensu Pitt), SBUG M-564, isolated 1979 by J. Sandoval from grassland soil near Greifswald	Q-9 PC
<i>Penicillium brevicompactum</i> Dierckx, SBUG M-881, ex CBS 257.29 = IMI 40225 = ATCC 10418, isolated by P. Biourge	Q-9 LC
<i>Penicillium camemberti</i> Thom, SBUG M-414, commercial strain ex VEB Ostra, Dresden	Q-9 PC
<i>Penicillium canescens</i> Sopp, SBUG M-537, isolated 1978 by J. Sandoval from grassland soil near Greifswald	Q-9 PC
<i>Penicillium citrinum</i> Thom, SBUG M-260, isolated 1965 by H. Kreisel from ascoma of <i>Gyromitra gigas</i> , Stralsund	Q-9 PC
<i>Penicillium clavigerum</i> Demelius, SBUG M-970, ex MW Weimar	Q-9 LC, PC
<i>Penicillium crustosum</i> Thom, SBUG M-823, isolated 1987 by V. Ernst from deciduous forest soil near Greifswald	Q-9 PC
<i>Penicillium cylindrosporium</i> G. Smith (= <i>Geosmithia cylindrospora</i> (G. Smith) Pitt), SBUG M-900, ex CCM F-439, isolated by L. Marvanová from technical oil	Q-9 + Q-10 LC
<i>Penicillium digitatum</i> (Pers.: Fr.) Sacc., SBUG M-937, isolated 1988 by H. Marko from air in hospital, Rostock	Q-9 LC, PC
<i>Penicillium glabrum</i> (Wehmer) Westling, SBUG M-1000, isolated 1989 by M. Schubert from <i>Crassula arborescens</i> , Greifswald	Q-9 PC
<i>Penicillium glandicola</i> (Oudem.) Seifert & Samson, SBUG M-973, ex MW i 488 (as <i>P. granulatatum</i>)	Q-9 PC
<i>Penicillium inflatum</i> Stolk & Malla, SBUG M-1018, ex CCM 8036	Q-9 LC
<i>Penicillium islandicum</i> Sopp, SBUG M-108, ex CCM F-473 = ATCC 26535, isolated by K. Ishii from wheat flour	Q-10(H ₂) LC, PC

Strain	Ubiquinone type
<i>Penicillium italicum</i> Wehmer, SBUG M-928, isolated 1988 by M. Schubert from Uruguayan orange, Greifswald	Q-9 PC
<i>Penicillium janczewskii</i> Zaleski, SBUG M-413, isolated 1973 by G. Salzsieder from deciduous forest soil, Niederhof near Stralsund	Q-9 PC
<i>Penicillium janczewskii</i> Zaleski, SBUG M-594, isolated 1981 by A. Klement from wheat phylloplane, Greifswald	Q-9 PC
<i>Penicillium lanosum</i> Westling (= <i>P. puberulum</i> sensu Pitt), SBUG M-287, ex E.A.N. Sacavem (as <i>P. expansum</i>)	Q-9 PC
<i>Penicillium</i> cf. <i>lividum</i> Westling, SBUG M-588, isolated 1979 by J. Sandoval from grassland soil near Greifswald	Q-9 PC
<i>Penicillium olsonii</i> Bain. & Sart., SBUG M-991, ex CBS 232.60, isolated from <i>Picea abies</i> root, Austria	Q-9 PC
<i>Penicillium oxalicum</i> Currie & Thom, SBUG M-828, isolated 1987 by P. Neubauer from Elbe river water, Pirna	Q-9 PC
<i>Penicillium piceum</i> Raper & Fennell, SBUG M-905, isolated 1988 by H. Böhm in plant cell culture, Halle	Q-10(H ₂) PC
<i>Penicillium pinophilum</i> Hedgcock, SBUG M-899, ex CBS 303.67, type strain of <i>P. proteolyticum</i> Kamyschko ¹	Q-10(H ₂) PC
<i>Penicillium purpurogenum</i> Stoll, SBUG M-67, ex CCM F-709 = CP 187, isolated by R.A. Hill from corn	Q-10(H ₂) LC
<i>Penicillium purpurogenum</i> Stoll, SBUG M-370, ex CCM F-199, isolated by M. Polster	Q-10(H ₂) LC
<i>Penicillium restrictum</i> Gilman & Abbott, SBUG M-429, isolated 1973 by G. Salzsieder from deciduous forest soil, Niederhof near Stralsund	Q-9 PC
<i>Penicillium roqueforti</i> Thom, SBUG M-982, isolated 1989 by E. Retzlaff from Roquefort cheese, Greifswald	Q-9 LC, PC
<i>Penicillium rugulosum</i> Thom, SBUG M-955, isolated 1988 by H. Marko from air in hospital, Rostock	Q-10(H ₂) PC
<i>Penicillium sacculum</i> Dale (= <i>Eladia saccula</i> (Dale) G. Smith), SBUG M-582, isolated 1979 by J. Sandoval from meadow soil near Greifswald	Q-9 LC, PC
<i>Penicillium variabile</i> Sopp, SBUG M-818, isolated 1986 by E.D. Erdenschimeg from soil at filling station, Greifswald	Q-10(H ₂) PC
<i>Penicillium variabile</i> Sopp, SBUG M-1009, isolated 1989 by M. Schubert from alpine soil, 2000 m s. m. near Mürren, Switzerland	Q-10(H ₂) PC
<i>Penicillium</i> cf. <i>variabile</i> Sopp, SBUG M-985, isolated 1989 by M. Schubert from mouldy sausage, Greifswald	Q-10(H ₂) and Q-x LC, PC
<i>Penicillium vulpinum</i> (Cooke & Masee) Seifert & Samson, SBUG M-967, ex MW i 490, isolated by G.R.W. Arnold from <i>Nymphaea</i> leaves (as <i>P. claviforme</i>).	Q-9 PC
<i>Talaromyces byssochlamydoides</i> Stolk & Samson, Anam. <i>Paecilomyces byssochlamydoides</i> Stolk & Samson, SBUG M-855 (det. R.A. Samson), isolated 1987 by K. Zimmermann from glasshouse soil, Oberlausitz, Saxonia	Q-10(H ₂) PC

¹ *Penicillium proteolyticum* Kamyschko was treated as a synonym of *P. verruculosum* Peyronel by Pitt (1979b), but as a synonym of *P. funiculosum* Thom by Samson (in CBS List of Cultures, 30th ed., 1983). The type culture received from CBS shows colonies on MEA exceeding 22 mm diam. after 7 d, on CzA exceeding 12 mm diam., aerial mycelium white to yellow, stipes 90–120 µm long, conidia smooth walled, grey green in mass; therefore it keys out as *P. pinophilum* Hedgcock.

Strain	Ubiquinone type
<i>Talaromyces flavus</i> (Klöcker) Stolk & Samson, Anam. <i>Penicillium dan-gardii</i> Pitt, SBUG M-941, ex WM (as <i>P. spiculisorum</i>)	Q-10(H ₂) PC
<i>Talaromyces stipitatus</i> (Thom) C.R. Benjamin, Anam. <i>Penicillium em-monsii</i> Pitt, SBUG M-271 ex CCM Brno F-174 = CBS 375.48 = ATCC 10500 (type strain of <i>P. stipitatum</i> Thom), isolated by K.B. Raper from wood, U.S.A.	Q-10(H ₂) PC
<i>Talaromyces wortmannii</i> Klöcker, Anam. <i>Penicillium kloeckeri</i> Pitt, SBUG M-410, isolated 1972 by G. Salzsieder from deciduous forest soil, Niederhof near Stralsund	Q-10(H ₂) LC
<i>Thermoascus thermophilus</i> (Sopp) v. Arx, Anam. <i>Polypaecilium spec.</i> , SBUG M-859, isolated 1987 by K. Zimmermann from glasshouse soil, Ober-lausitz, Saxonia	Q-9 PC

DISCUSSION

Von Arx (1987: 282 and 289) regarded the genus *Penicillium* in the classic circumscription (Raper & Thom, 1949; Pitt 1979b) as polyphyletic, stressing the particular position of the section *Biverticillium* 'which should be transferred to *Paecilomyces*', but he avoided to make any new combinations with *Paecilomyces*.

In the system of Stolk & Samson (1985), *Penicillium* appears again in a very broad concept, divided in 10 sections, whereas Pitt (1979b) had arranged it in four subgenera with nine sections, excluding *Geosmithia* and *Merimbla* (= *Raperia*) as separate anamorphic genera. The analysis of ubiquinone types provides new suggestions for the taxonomic treatment of the mentioned genera.

It should be noted that results obtained by RPPC and by RPHPLC from the same strain were identical, as well as results from different strains of the same species, even if obtained independently by different authors.

Ubiquinone Q-9 was found in the great majority of the investigated strains, i.e. in 27 of the 38 species. Including the results of previous authors, Q-9 has been demonstrated in all investigated species of the teleomorphic genus *Eupenicillium* (Eurotiaceae, teste von Arx, 1987) and – with one exception – of *Penicillium* sect. *Torulomyces*, *Aspergilloides*, *Eladia*, *Divaricatum*, *Inornata*, *Ramosum*, *Penicillium*, and *Coremigenum* in the system of Stolk & Samson (1985). The one exception is *P. megasporum* (sect. *Eladia*), in which ubiquinone Q-10 was found by Kuraishi & al. (1985).

Ubiquinone Q-10(H₂) was found in all investigated species of the teleomorphic genus *Talaromyces* (Onygenaceae) and of *Penicillium* sect. *Biverticillium* (*P. islandicum*, *P. piceum*, *P. pinophilum*, *P. purpurogenum*, *P. rugulosum*, *P. variabile*). This is coincident with results of previous authors (Lavate & al., 1965 for *T. stipitatus*, Kuraishi & al., 1985 for *T. flavus* and *P. islandicum*). Consequently, section *Biverticillium* with its teleomorph *Talaromyces* occupies from the chemotaxonomic point of view a rather separate position within the genus *Penicillium* and deserves a higher taxonomic rank than most of the other sections. Pitt (1979b) recognized a subgenus *Biverticillium*, including the sections *Coremigenum* and *Simplicium*

(= *Biverticillium* s. str.), but our analysis of two representative species of sect. *Coremigenum* (*P. clavigerum*, *P. vulpinum*), both with Q-9, does not support such an arrangement.

An interesting exception in section *Biverticillium* is one doubtfully determined strain of *P. cf. variable*, which has Q-10(H₂) and in somewhat minor quantity another, unidentified ubiquinone, possibly Q-10(H₄), but no reference substance was available.

Ubiquinone Q-10 (main component) and Q-9 (minor component) were found in *P. cylindrosporum*, hitherto the only investigated species of section *Geosmithia* resp. of the genus *Geosmithia*, segregated from *Penicillium* by Pitt (1979a).

Hamigera (Onygenaceae) with its anamorph *Raperia* (*Merimbla*) is a small genus. The ubiquinone of *H. avellanea*, Anam. *Raparia ingelheimensis* (= *Penicillium ingelheimense*), was determined as Q-10 by Kuraishi & al. (1985). This supports a generic separation of *Hamigera* from both *Eupenicillium* and *Talaromyces*.

Finally, the investigated representant of the genus *Thermoascus* (Onygenaceae), *Th. thermophilus*, has coenzyme Q-9. The same type was found in another species, *Th. auranthiacus*, by Kuraishi & al. (1985).

The results obtained in this paper and by previous authors support the concept that *Geosmithia* and *Biverticillium* s. str. should be separated from *Penicillium* s. str. (which contains the majority of the species) on generic level. But definite taxonomic decisions (new combinations) should be made only after determination of ubiquinone types in more species and strains of *Penicillium* s.l.

The authors of this paper could not consider the oral communication by Pitt & Samson on ubiquinone types in *Penicillium*, during the Fourth International Mycological Congress in Regensburg, 1990.

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