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RESEARCH ARTICLE

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Taxonomic re-evaluation of species in *Talaromyces* section *Islandici*, using a polyphasic approach

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Key words

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Penicillium rugulosum
Penicillium variabile
Talaromyces acaricola
Talaromyces crassus
Talaromyces infraolivaceus
Talaromyces subaurantiacus

Abstract The taxonomy of *Talaromyces rugulosus*, *T. wortmannii* and closely related species, classified in *Talaromyces* sect. *Islandici*, is reviewed in this paper. The species of *Talaromyces* sect. *Islandici* have restricted growth on MEA and CYA, generally have yellow mycelia and produce rugulosin and/or skyrin. They are important in biotechnology (e.g. *T. rugulosus*, *T. wortmannii*) and in medicine (e.g. *T. piceus*, *T. radicus*). The taxonomy of sect. *Islandici* was resolved using a combination of morphological, extrolite and phylogenetic data, using the Genealogical Concordance Phylogenetic Species Recognition (GCPSR) concept, with special focus on the *T. rugulosus* and *T. wortmannii* species complexes. In this paper, we synonymise *T. variabilis*, *Penicillium concavorugulosum* and *T. sublevisporus* with *T. wortmannii*, and introduce four new species as *T. acaricola*, *T. crassus*, *T. infraolivaceus* and *T. subaurantiacus.* Finally, we provide a synoptic table for the identification of the 19 species classified in the section.

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INTRODUCTION

Penicillium s.l. is an important group of molds associated with a wide range of habitats, where it acts as degraders of organic material (Pitt 1980, Frisvad & Samson 2004). *Penicillium* subg. *Biverticillium* species are phylogenetically resolved in a well-supported monophyletic clade together with the teleomorphic genus *Talaromyces*, distinct from other *Penicillium* subgenera (LoBuglio et al. 1993, Houbraken & Samson 2011, Samson et al. 2011). Following the recent move to single name nomenclature in fungi, Samson et al. (2011) subsequently combined all accepted species belonging to *Penicillium* subg. *Biverticillium* into *Talaromyces*. Yilmaz et al. (2014), using a polyphasic approach, accepted 88 species in *Talaromyces* and based on a multi-gene phylogeny classified them into seven sections. One of these sections is sect. *Islandici* and this section is in the focus of this study.

Talaromyces sect. *Islandici* includes species that grow restrictedly on most media, have predominately yellow mycelia, and produce characteristic mycotoxins. Previously, Pitt (1980) introduced the *Penicillium* sect. *Simplicium* ser. *Islandica* for species which grow restrictedly on malt extract agar (MEA) and Czapek yeast extract agar (CYA). He included *T. brunneus*, *T. erythromellis*, *T. islandicus*, *T. loliensis*, *T. piceus*, *T. primulinus*, *T. rugulosus* and *T. variabilis*. However, a multi-gene phylogeny showed that *T. erythromellis* and *T. primulinus* are located in sect. *Trachyspermi* and *Talaromyces*, respectively (Yilmaz et al. 2014) and that the remaining species were included in sect. *Islandici* (Yilmaz et al. 2014).

This group of species typically produces rugulosin and/or skyrin (except for *T. scorteus*) (Yilmaz et al. 2014). Rugulosin is a bis-anthraquinoid pigment described by Breen et al. (1955) with a specific antibacterial activity against *Staphylococcus aureus* and moderate activity against the parasitic fungus-like Chromistan *Pythium intermedium*. Rugulosin was also indicated as a weak hepato-carcinogen (Ueno et al. 1980). A recent study showed that rugulosin extracted from *T. radicus* had antimicrobial activity against methicillin resistant *S. aureus* (Yamazaki et al. 2010a–c). Even though it has been classified as a mycotoxin, erythroskyrin was also reported to be an antitumor agent (Kenkyusho 1983). Rubroskyrin and flavoskyrin are also classified as toxins (Kawai et al. 1984, Mori et al. 1996) and are produced by some sect. *Islandici* species. The rugulovasines (Antipova et al. 2008) were referred to as mycotoxins but toxicity data are scarce (Cole & Cox 1981).

Talaromyces sect. *Islandici* includes important enzyme producers such as *T. wortmannii* (= *T. variabilis*) producing urethanase (Zhou et al. 2013) and *T. rugulosus* producing beta-rutinosidase and phosphatase (Reyes et al. 1999, Narikawa et al. 2000). *Talaromyces wortmannii* also produces high concentrations of uncharacterised bioactive natural compounds. Bara et al. (2013) showed that six compounds isolated from *T. wortmannii* exhibited antibacterial activity, predominantly directed against *S. aureus*, including (multi) drug-resistant isolates. However, other Gram-positive bacteria such as *Streptococcus*, *Enterococcus* and *Bacillus* were only moderately affected (Bara et al. 2013). Several compounds were isolated from *T. wortmannii* by Pretsch et al. (2014). A metabolite labelled as Compound C was found an effective antimicrobial against *Propionibacterium acnes* and had anti-inflammatory properties (Pretsch et al. 2014). It was thus suggested that this substance, or the crude extract, could represent alternative treatments for antibiotic/anti-inflammatory therapy for acne (Pretsch et al. 2014).

Some species of *Talaromyces* sect. *Islandici* may be potential opportunistic pathogens because of their ability to grow at 37 °C and higher (Yilmaz et al. 2014). Previous studies reported that *T. piceus* caused fungaemia (Horré et al. 2001) and rib osteomyelitis in an X-linked chronic granulomatous disease (X-CGD) (Santos et al. 2006). *Talaromyces radicus* caused a fatal infection in a German shepherd (de Vos et al. 2009). Corneal ulcer

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Table 1 Strains used for this study.

Name	Collection no.	Substrate and origin	GenBank accession numbers			
			BenA	*CaM*	ITS	*RPB2*
T. acaricola	CBS 137367 = DTO 61-H2	Air sample, beer producing factory, Kaulille, Belgium	KF984567	KF984720	KF984862	KF984953
	CBS 137369 = DTO 66-H7	Unknown, unknown	KF984568	KF984721	KF984860	KF984954
	CBS 137374 = DTO 77-C7	Apple concentrate, The Netherlands	KF984569	KF984722	KF984861	KF984955
	CBS 137386^{T} = DTO 183-B3 = DAOM 241025 = IBT 32387	Mites from *Protea repens* infructescens, Malmesbury, South Africa	JX091610	JX140729	JX091476	KF984956
	CBS 137387 = DTO 183-B4	Mites from *Protea repens* infructescens, Malmesbury, South Africa	JX091611	JX140730	JX091477	KF984957
	CBS 137388 = DTO 183-C1 = DAOM 241029	*Protea repens* infructescence, Malmesbury, South Africa	JX091612	JX140731	JX091478	KF984958
	CBS 137390 = DTO 183-E3 = DAOM 241022	Mites from *Protea repens* infructescens, Struisbaai, South Africa	JX091613	JX140732	JX091479	KF984959
T. allahabadensis	CBS 137361 = DTO 55-F9	Swab sample in vaccin producing factory, The Netherlands	KF984608	KF984763	KF984867	KF985000
	CBS 137362 = DTO 55-G3	Indoor air sample in vaccin producing factory, The Netherlands	KF984610	KF984765	KF984869	KF985002
	CBS 137373 = DTO 77-C3	Guava pure imported to The Netherlands	KF984615	KF984769	KF984871	KF985007
	CBS 137397 = DTO 245-E3	House dust, Mexico	KF984605	KF984761	KF984864	KF984998
	CBS 137399 = DTO 267-H6	House dust, Thailand	KF984607	KF984762	KF984866	KF984997
	CBS 178.81 = DTO 247-D9 = ATCC 48474 = FRR 3579 = IMI 253805	Type of *P. zacinthae*, crepis zacintha, Alicante, Spain	KF984612	KF984767	KF984863	KF985004
	CBS 441.89 = DTO 247-D5	Seed groud, Denmark	KF984613	KF984759	KF984872	KF985005
	DTO 055-G1	Indoor air sample in vaccin producing factory, The Netherlands	KF984609	KF984764	KF984868	KF985001
	DTO 067-F7	Indoor air sample in vaccin producing factory, The Netherlands	KF984611	KF984766	KF984870	KF985003
	DTO 245-I6	House dust, Mexico	KF984606	KF984760	KF984865	KF984999
	CBS 453.93^{T} = DTO 93-B7 = CBS 304.63 = ATCC 15067 = NRRL 3397 = FRR 3397 = IBT 3926 = IBT 10824	Cultivated soil, Allahabad, India	KF984614	KF984768	KF984873	KF985006
T. atricola	CBS 255.31^{T} = DTO 278-F1 = NRRL1052 = FRR 1052 = Thom 4640.439 = ATCC 52257 = IBT 4489	Unknown, unknown	KF984566	KF984719	KF984859	KF984948
T. brunneus	CBS 227.60^{T} = DTO 284-G1 = ATCC 18229 = FRR 646 = IFO 6438 = IMI 078259 = IBT 4490	Milled rice impoted into Japan, Thailand	KJ865722	KJ885264	JN899365	KM023272
T. columbinus	CBS 137393 = DTO 189-A5 = IBT 13019	Chicken feed (Unga), Nairobi, Kenya	KF984659	KF984671	KF984794	KF984897
	NRRL 58644	Air, Maryland, USA	KF196842	KF196880	KF196899	KF196987
	NRRL 62680	Corn grits, Illinois, USA	KF196844	KF196882	KF196901	KF196988
T. crassus	CBS 137379 = DTO 181-B1	*Protea repens* infructescence, Stellenbosch, South Africa	JX091606	JX140726	JX091473	KF984912
	CBS 137380 = DTO 181-B2	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	JX091607	JX140725	JX091474	KF984913
	CBS 137381^{T} = DTO 181-C5 = DAOM 241027 = IBT 32814	*Protea repens* infructescence, Stellenbosch, South Africa	JX091608	JX140727	JX091472	KF984914
T. infraolivaceus	CBS 137385^{T} = DTO 182-I2 = DAOM 241024 = IBT 32487	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	JX091615	JX140734	JX091481	KF984949
	CBS 137389 = DTO 183-D2 = DAOM 241023	Mite from *Protea repens* infructescence, Struisbaai, South Africa	JX091616	JX140735	JX091482	KF984950
	CBS 137391 = DTO 183-F1 = DAOM 241030	*Protea repens* infructescence, Struisbaai, South Africa	JX091617	JX140736	JX091483	KF984951
	CBS 137392 = DTO 183-G4	Mite from *Protea repens* infructescence, Struisbaai, South Africa	JX091618	JX140737	JX091484	KF984952
T. islandicus	CBS 338.48^{T} = DTO 107-H2 = ATTC 10127 = FRR 1036 = IMI 040042 = NRRL 1036 = IBT 14884 = IBT 4476	Unknown, Cape Town, South Africa	KF984655	KF984780	KF984885	KF985018
	CBS 117284 = DTO 2-C7	Wheat flour, The Netherlands	KF984652	KF984777	KF984882	KF985015
	CBS 165.81 = DTO 158-D6 = ATCC 42240 = IMI 253796	Type of *P. aurantioflammiferum*, spice mixture used in sausage making industry, Spain	KF984653	KF984778	KF984883	KF985016
	CBS 394.50 = DTO 93-B9	Kapok fibre, unknown	KF984656	KF984781	KF984886	KF985019
	DTO 158-D7	Air contaminant, The Netherlands	KF984654	KF984779	KF984884	KF985017
T. loliensis	CBS 172.91 = DTO 105-E9	Soil, New Zealand	KF984657	KF984782	KF984887	KF985020
	CBS 643.80^{T} = DTO 169-F7 = ATCC 52252 = FRR 1798 = IMI 216901 = NRRL 2148 = MUCL 31325 = IBT 4546	Rye grass (Lolium), New Zealand	KF984658	KF984783	KF984888	KF985021
T. piceus	CBS 116872 = DTO 247-E1	Production plant, The Netherlands	KF984660	KF984678	KF984788	KF984903
	CBS 132063 = DTO 191-C5	Straw used in horse stable, The Netherlands	KF984665	KF984674	KF984789	KF984904
	CBS 137363 = DTO 58-D1	Pectin, unknown	KF984664	KF984677	KF984787	KF984902
	CBS 137377 = DTO 178-F3	House dust, Cape Town, South Africa	KF984661	KF984676	KF984784	KF984900

	CBS 250.56 = DTO 93-G8	Sputum from a man patient, The Netherlands	KF984666	KF984679	KF984790	KF984905
	CBS 354.66 = DTO 93-C6	Unknown, United Kingdom	KF984667	KF984672	KF984791	KF984907
	CBS 361.48^{T} = DTO 93-C8 = IMI 040038 = NRRL 1051 = FRR 1051 = ATCC 10519 = Thom 5633.6 = QM 7609 = IBT 4460	Unknown, unknown	KF984668	KF984680	KF984792	KF984899
	CBS 435.62 = DTO 228-E5	Sputum, The Netherlands	KF984669	KF984681	KF984793	KF984906
	DTO 191-C4	Straw used in horse stable, The Netherlands	KF984662	KF984673	KF984785	KF984898
	DTO 191-C6	Silage, grass, The Netherlands	KF984663	KF984675	KF984786	KF984901
T. radicus	CBS 100488 = DTO 37-F6	Wheat root, New South Wales	KF984598	KF984772	KF984877	KF985012
	CBS 100489^{T} = DTO 37-F7 = FRR 4718 = IBT 14379	Root seadling, New South Wales	KF984599	KF984773	KF984878	KF985013
	CBS 100490 = DTO 37-F8	Wheat root, New South Wales	KF984600	KF984774	KF984879	KF985014
	CBS 122887 = DTO 63-C5	Ex infection dog, The Netherlands	KF984604	KF984776	KF984876	KF985011
	CBS 137382 = DTO 181-D5	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984602	KF984775	KF984875	KF985009
	DTO 181-D4	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984601	KF984770	KF984880	KF985008
	DTO 181-D7	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984603	KF984771	KF984881	KF985010
T. rotundus	CBS 369.48^{T} = DTO 105-D3 = IMI 40589 = NRRL 2107 = FRR 2107 = ATCC 10493 = IBT 4829	Wood, Panama	KJ865730	KJ885278	JN899353	KM023275
T. rugulosus	CBS 101423 = DTO 225-I6	Wood, British Columbia, Vancouver, Canada	KF984597	KF984717	KF984856	KF984944
	CBS 111.64 = DTO 93-B8	Jute, The Netherlands	KF984573	KF984691	KF984857	KF984923
	CBS 137360 = DTO 14-A2	Cardboard, Norway	KF984588	KF984703	KF984835	KF984931
	CBS 137366 = DTO 61-E8	Air sample, beer producing factory, Kaulille, Belgium	KF984572	KF984700	KF984850	KF984922
	CBS 137369 = DTO 63-C7	Jelly like product used in bakery, The Netherlands	KF984586	KF984713	KF984851	KF984947
	CBS 137372 = DTO 70-B7	Wood of crate, United Kingdom	KF984582	KF984697	KF984855	KF984943
	CBS 137378 = DTO 180-A6	House dust, Cape Town, South Africa	KF984591	KF984704	KF984838	KF984933
	CBS 137398 = DTO 254-A2	Indoor air, Utrecht, The Netherlands	KF984595	KF984695	KF984845	KF984937
	CBS 258.37 = DTO 166-A6 = NRRL 2116 = KCTC 16068	Unknown, unknown	KF984580	KF984718	KF984833	KF984928
	CBS 344.51 = DTO 93-C2 = ATCC 22352 = FRR 560	Type of *P. echinosporum*, unknown, Japan	KF984574	KF984701	KF984858	KF984924
	CBS 371.48^{T} = DTO 278-E8 = NRRL 1045 = FRR 1045 = IMI 040041 = ATCC 10128 = MUCL 31201 = IBT 4485	Roating potato tubers (solanum tuberosum), USA	KF984575	KF984702	KF984834	KF984925
	CBS 378.48 = DTO 278-F2 = NRRL1073 = FRR 1073 = IMI 040034 = ATCC 10503 = Thom 4640.444	Type of *P. tardum* & *P. elongatum*, decaying twigs, France	KF984579	KF984711	KF984832	KF984927
	DTO 066-G6	Unknown, unknown	KF984585	KF984714	KF984852	KF984940
	DTO 066-G7	Unknown, unknown	KF984584	KF984715	KF984853	KF984941
	DTO 070-B5	Wood of crate, United Kingdom	KF984583	KF984716	KF984854	KF984942
	DTO 179-I3	House dust, Cape Town, South Africa	KF984589	KF984692	KF984836	KF984932
	DTO 180-A4	House dust, Cape Town, South Africa	KF984590	KF984693	KF984837	KF984929
	DTO 180-B3	House dust, Cape Town, South Africa	KF984587	KF984705	KF984839	KF984934
	DTO 180-B9	House dust, Cape Town, South Africa	KF984570	KF984698	KF984840	KF984920
	DTO 193-I5 = IBT 10835 = IBT 3616	Unknown, unknown	KF984578	KF984706	KF984831	KF984926
	DTO 199-H3	Material for bed for milking cows, The Netherlands	KF984576	KF984707	KF984841	KF984946
	DTO 244-F6	House dust, New Zealand	KF984593	KF984709	KF984843	KF984930
	DTO 254-A1	Indoor air, Utrecht, The Netherlands	KF984594	KF984694	KF984844	KF984936
	DTO 269-G1	House dust, Cape Town, South Africa	KF984581	KF984696	KF984846	KF984938
	DTO 269-G4	House dust, Cape Town, South Africa	KF984571	KF984699	KF984847	KF984921
	DTO 278-E9 = NRRL 1053 = FRR 1053 = IMI 028259	Type of *P. chrysitis*, unknown, unknown	KF984577	KF984710	KF984848	KF984945
	DTO 61-E4	Air sample, beer producing factory, Kaulille, Belgium	KF984596	KF984712	KF984849	KF984939
T. scorteus	CBS 233.60 = DTO 278-F3 = NRRL 203 = IMI 78256 = FRR 203 = ATCC 18481 = IFO 6437	Type of *T. phialosporus*, milled Californian rice, Japan	KF984562	KF984683	KF984895	KF984917
	CBS 499.75 = DTO 247-D7 = IMI 144145	Unknown, Nigeria	KF984563	KF984685	KF984894	KF984918
	CBS 500.75 = DTO 225-I5 = IMI 152168 = KCTC 16071	Unknown, Sierra Leone	KF984564	KF984687	KF984896	KF984919
	DTO 270-A6	House dust, Thailand	KF984561	KF984686	KF984893	KF984915
	CBS 340.34^{T} = DTO 278-F4 = NRRL 1129 = FRR 1129	Military equipment, Japan	KF984565	KF984684	KF984892	KF984916
T. subaurantiacus	CBS 137383^{T} = DTO 181-I2 = DAOM 241020 = IBT 32838	Fynbos soil, Stellenbosch, South Africa	JX091609	JX140728	JX091475	KF984960

Table 1 (cont.)

Name	Collection no.	Substrate and origin	GenBank accession numbers			
			BenA	*CaM*	ITS	*RPB2*
T. tardifaciens	CBS 250.94^{T} = DTO 247-D6 = SUM 3017 = IBT 14986	Paddy soil, Bhaktapur, Nepal	KF984560	KF984682	KF984874	KF984908
T. tratensis	CBS 113146^{T} = DTO 140-G4 = KUFC 3383 = IBT 31982	Soil, Trat, Thailand	KF984559	KF984690	KF984891	KF984911
	CBS 137400 = DTO 270-F5	House dust, Mexico	KF984557	KF984688	KF984889	KF984909
	CBS 137401 = DTO 278-F6 = NRRL1013 = FRR 1013	Carbonated beverage, Washington DC, USA	KF984558	KF984689	KF984890	KF984910
T. wortmannii	CBS 100258[c] = DTO 226-A5	Unknown, unknown	KF984630	KF984723	KF984823	KF984969
	CBS 116051[a] = DTO 92-I7	Unknown, unknown	KF984640	KF984736	KF984809	KF984995
	CBS 130028[c] = DTO 208-I8	Unknown, adjacent to Cinnabar Park, Medicine Bow National Forest, Wyoming, USA	KF984631	KF984749	KF984818	KF984974
	CBS 137364[a] = DTO 58-G6	Corn kernels, imported from Brazil, The Netherlands	KF984636	KF984732	KF984806	KF984980
	CBS 137365[a] = DTO 58-H6	Wooden crate, imported from China, The Netherlands	KF984638	KF984734	KF984808	KF984993
	CBS 137368[c] = DTO 63-C6	Indoor air, The Netherlands	KF984617	KF984746	KF984815	KF984964
	CBS 137371[c] = DTO 67-F9	Indoor air sample in vaccin producing factory, The Netherlands	KF984646	KF984753	KF984825	KF984972
	CBS 137375[c] = DTO 161-G6	Bamboo, Ho Chi Minh City, Vietnam	KF984643	KF984742	KF984820	KF984976
	CBS 137376[b] = DTO 176-I7 = PF1130	Type of *T. sublevisporus*, soil, Japan	KF984632	KF984724	KF984800	KF984979
	CBS 137384[a] = DTO 181-H2 = DAOM 241019	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984622	KF984726	KF984802	KF984985
	CBS 137394[a] = DTO 189-C8 = IBT 28678	Soil, Amazonas, Brazil	KF984624	KF984729	KF984795	KF984988
	CBS 137395[a] = DTO 189-D8 = IBT 30868	Unknown, Brazil	KF984634	KF984740	KF984805	KF984990
	CBS 293.53 = DTO 108-A3	Soil, Mozambique	KF984650	KF984758	KF984827	KF984962
	CBS 316.63[c] = DTO 93-A1 = KCTC 16056	Polyvinyl acetate, The Netherlands	KF984620	KF984750	KF984819	KF984975
	CBS 319.63 = DTO 108-A4	Unknown, unknown	KF984651	KF984755	KF984828	KF984961
	CBS 385.48[c] = DTO 92-I8 = IMI 40040 = NRRL 1048 = FRR 1048 = ATCC 10508 = IFO 6111	Type of *T. variabilis*, coconut matting, Johannesburg, South Africa	KF196853	KF196878	KF196915	KF196975
	CBS 391.48^{T} = DTO 278-F5 = NRRL 1017 = IMI 040047 = FRR 1017 = ATCC 10517 = IFO 7738 = Thom 4733.126.1 = IBT 4838	Soil, Denmark	KF984648	KF984756	KF984829	KF984977
	CBS 553.72 = DTO 107-H4	Soil, France	KF984649	KF984757	KF984830	KF984978
	CBS 777.95[a] = DTO 92-I6	Rubber, Sri Lanka	KF984641	KF984741	KF984810	KF984981
	CBS 895.73[a] = DTO 92-I9 = ATCC 20201 = IFO 4683 = BCRC 31677	Unknown, Japan	KF984626	KF984737	KF984811	KF984982
	CBS 896.73[a] = DTO 93-C7 = BCRC 31676 = IFO 9136	Unknown, Japan	KF984642	KF984738	KF984799	KF984996
	CBS 898.73[a] = DTO 93-A2 = ATCC 20202 = IFO 6017 = BCRC 31678	Unknown, Japan	KF984627	KF984739	KF984812	KF984983
	DTO 1-I3[c]	Wood, Amsterdam, The Netherlands	KF984616	KF984745	KF984814	KF984963
	DTO 127-I4[c]	Sauce, The Netherlands	KF984628	KF984744	KF984813	KF984967
	DTO 181-B7[a] = DAOM 241018	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984621	KF984725	KF984801	KF984984
	DTO 181-I7[a]	Mite from *Protea repens* infructescence, Stellenbosch, South Africa	KF984623	KF984727	KF984803	KF984986
	DTO 189-C6[a] = IBT 27918 = NCB 1494	Ex passito wine grape, Italy	KF984633	KF984728	KF984804	KF984987
	DTO 189-C9[a] = IBT 28726	Soil, Amazonas, Brazil	KF984625	KF984730	KF984796	KF984989
	DTO 189-D9[a] = IBT 31242	Wheat flour, Denmark	KF984629	KF984743	KF984821	KF984968
	DTO 278-E7[c] = NRRL 2125 = FRR 2125	Weathering canvas, Panama	KF984635	KF984731	KF984797	KF984991
	DTO 55-G2[c]	Swab sample in vaccin producing factory, The Netherlands	KF984644	KF984751	KF984824	KF984970
	DTO 58-H1[a]	Corn kernels, imported from Brazil, The Netherlands	KF984637	KF984733	KF984807	KF984992
	DTO 66-H5[c]	Unknown, unknown	KF984639	KF984735	KF984798	KF984994
	DTO 67-F8[c]	Indoor air sample in vaccin producing factory, The Netherlands	KF984645	KF984752	KF984822	KF984971
	DTO 67-G1[c]	Indoor air sample in vaccin producing factory, The Netherlands	KF984647	KF984754	KF984826	KF984973
	DTO 67-G2[c]	Indoor air sample in vaccin producing factory, The Netherlands	KF984618	KF984747	KF984816	KF984965
	DTO 67-G3[c]	Indoor air sample in vaccin producing factory, The Netherlands	KF984619	KF984748	KF984817	KF984966
T. yelensis	CBS 138209^{T} = DTO 268-E5	House dust, Micronesia	KJ775210	KP119161	KJ775717	KP119163
	CBS 138210 = DTO 268-E7	House dust, Micronesia	KJ775212	KP119162	KJ775719	KP119164
Trichocoma paradoxa	CBS 788.83	Rotting stump of cut down tree, Japan	KF984556	KF984670	JN899398	JN121550

[a] Isolates previously identified as *Penicillium concavorugulosum*.
[b] Isolate previously identified as *Talaromyces sublevisporus*.
[c] Isolates previously identified as *Talaromyces variabilis*.

caused by *T. rugulosus* was reported by Swietliczkowa et al. (1984). *Talaromyces islandicus* can also grow at 37 °C, but until now has not been isolated from humans. It is more important for agriculture because it produces mycotoxins such as cyclochlorotine, islanditoxin, erythroskyrine and luteoskyrin, which are hepatotoxic agents and also carcinogenic (Uraguchi et al. 1961, 1972, Uraguchi 1962, Ueno & Ishikawa 1969, Bouhet et al. 1976, Stark et al. 1978, Pitt & Hocking 2009). This species also causes yellowing of rice in Japan (Saito et al. 1971, Sakai et al. 2005, Oh et al. 2008).

The diverse range of species in sect. *Islandici* species, and their importance in medicine, agriculture and biotechnology, make correct identifications crucial. The aim of this study, was thus to complete a multigene phylogenetic study of the section, and apply Genealogical Concordance Phylogenetic Species Recognition (GCPSR, Taylor et al. 2000) by adding to the internal transcribed spacer (ITS) and β-tubulin (*BenA*) data published in Yilmaz et al. (2014) and studying extrolites produced by the species, with a special focus on the *T. wortmannii* and *T. rugulosus* species complexes. The phylogenies resulted in the identification of four unique clades that we describe here as new species. Strains of these four new species mainly originate from a biodiversity study of Fynbos soil, *Protea repens* infructescences and air, in the Western Cape of South Africa (Visagie et al. 2009, 2013, 2014c, Visagie & Jacobs 2012). In addition to the multi-gene phylogenies, we compare the morphological characters and extrolite data of the new species with others in the section and provide notes to facilitate their identification.

MATERIAL AND METHODS

Isolates

Isolates used in this study were obtained from the culture collections of the CBS-KNAW Fungal Biodiversity Centre, Utrecht, The Netherlands; the culture collection of Center for Microbial Biotechnology at Department of Systems Biology, Technical University of Denmark, Lyngby, Denmark (IBT); the Agricultural Research Service Culture Collection, Peoria, Illinois, USA (NRRL); the Canadian Department of Agriculture – Mycology Culture Collection, Ottawa, Canada (DAOM); and isolates deposited in the working collection of the Department of Applied and Industrial Mycology (DTO), housed at CBS-KNAW. Isolates are listed in Table 1.

DNA extraction, PCR amplification and sequencing

DNA extractions were made from isolates grown for 7–14 d on MEA using the Ultraclean™ Microbial DNA isolation Kit (Mo-Bio, Solana Beach, USA) and extracted DNA was stored at -20 °C. The internal transcribed spacers, including the 5.8 S rDNA (ITS), calmodulin (*CaM*) and RNA polymerase II (*RPB2*) gene regions were amplified and sequenced using previously described methods (Yilmaz et al. 2014, Visagie et al. 2014b). For *BenA*, primer set T10 and Bt2b (Glass & Donaldson 1995) was used with annealing temperatures of 50 and 52 °C.

Phylogeny

Sequence contigs were assembled in Seqman v. 9.0.4 (DNASTar Inc.). The newly generated sequences were included in a dataset including sequences obtained from Peterson & Jurjević (2013) and Yilmaz et al. (2014). GenBank accession numbers for sequences used in the phylogenies are listed in Table 1. The dataset for each gene was aligned using Muscle software included within the MEGA5 software package (Tamura et al. 2011). The aligned ITS, *BenA*, *CaM* and *RPB2* data were concatenated in SeaView (Gouy et al. 2010) and analysed using Maximum Likelihood (ML) and Bayesian tree Inference (BI). The model for ML was selected based on the Akaike Information Criterion (AIC) calculated in MEGA5. The analysis was initiated by calculating an initial tree using BioNJ and the subsequent Heuristic done with the Nearest-Neighbour-Interchange (NNI). Bootstrap support was calculated using 1 000 replicates. The most suitable model for BI was selected based on AIC, calculated in MrModeltest v. 2.3 (Nylander et al. 2004). The analysis was run in MrBayes v. 3.2.1 (Huelsenbeck & Ronquist 2001) with two sets of four chains (one cold, three heated), until an average deviation of split frequencies reached 0.01. The sample frequency was set at 100, with 25 % of trees removed as burn-in phase.

Morphological analysis

Macroscopic characters were studied on different media and growth conditions. Cultures were plated onto Czapek yeast extract agar (CYA), CYA supplemented with 5 % NaCl (CYAS), yeast extract sucrose agar (YES), creatine sucrose agar (CREA), dichloran 18 % glycerol agar (DG18), oatmeal agar (OA) and malt extract agar (MEA; Oxoid malt). The isolates were inoculated at three points on 90 mm Petri dishes and incubated for 7 d at 25 °C in darkness. All media were prepared as described by Visagie et al. (2014b). Additional CYA plates were incubated at 37 °C for 7 d in darkness. The isolates growing at 37 °C, were also incubated at 40 °C for 7 d in darkness. After incubation, the colony diameters on the various media were measured. The density of sporulation, obverse and reverse colony colours and the production of soluble pigments were noted. Colony colour codes refer to Kornerup & Wanscher (1967). Colonies were photographed with a Canon EOS 400D. Species were characterised microscopically by preparing slides from MEA. Lactid acid was used as mounting fluid. Specimens were examined using a Zeiss AxioSkop2 plus microscope, and the NIS-Elements D software package from Nikon was used for capturing photographs and taking measurements.

Extrolites

Extrolites were extracted from fungal strains grown on CYA and YES at 25 °C for 7 d. In some cases, extractions were made from strains also grown on MEA and OA at 25 °C for 7 d. Three agar plugs of each medium were extracted as described in Nielsen et al. (2011) and Houbraken et al. (2012). The extracts were analysed by high performance liquid chromatography with diode-array detection (HPLC-DAD) (Frisvad & Thrane 1987) for extracts made before 2011 and by UHPLC-DAD (Houbraken et al. 2012) for extracts made after 2011. The eluted compounds were identified by comparing retention time, retention index and UV spectra measured at 200–600 nm. The UV spectra were compared to a database of UV spectra (Nielsen et al. 2011), and to data from the literature.

RESULTS

Phylogeny

A multi-gene phylogeny, based on four genes, was used to infer the relationships among species in *Talaromyces* sect. *Islandici* (Fig. 1). The aligned concatenated dataset (ITS 583 bp; *BenA* 482 bp; *CaM* 541 bp; *RPB2* 772 bp) had a total length of 2 378 bp. The most suitable model for ML was Kimura 2-parameter (K2)+Gamma distribution (G)+evolutionarily invariable (I) and the most suitable for BI was Genereal Time Reversible (GTR)+I+G. Tree topologies for ML and BI were identical. As such, the tree obtained from ML was used to show the result with both bootstrap support (bs) and posterior probabilities (pp) indicated above branches where support was higher than 80 % (bs) and/or 0.95 (pp) (Fig. 1). The same applies to phylogenies shown in Fig. 2 and 3. Based on the phylogeny (Fig. 1), *Talaromyces* sect. *Islandici* contains 19 species, including four

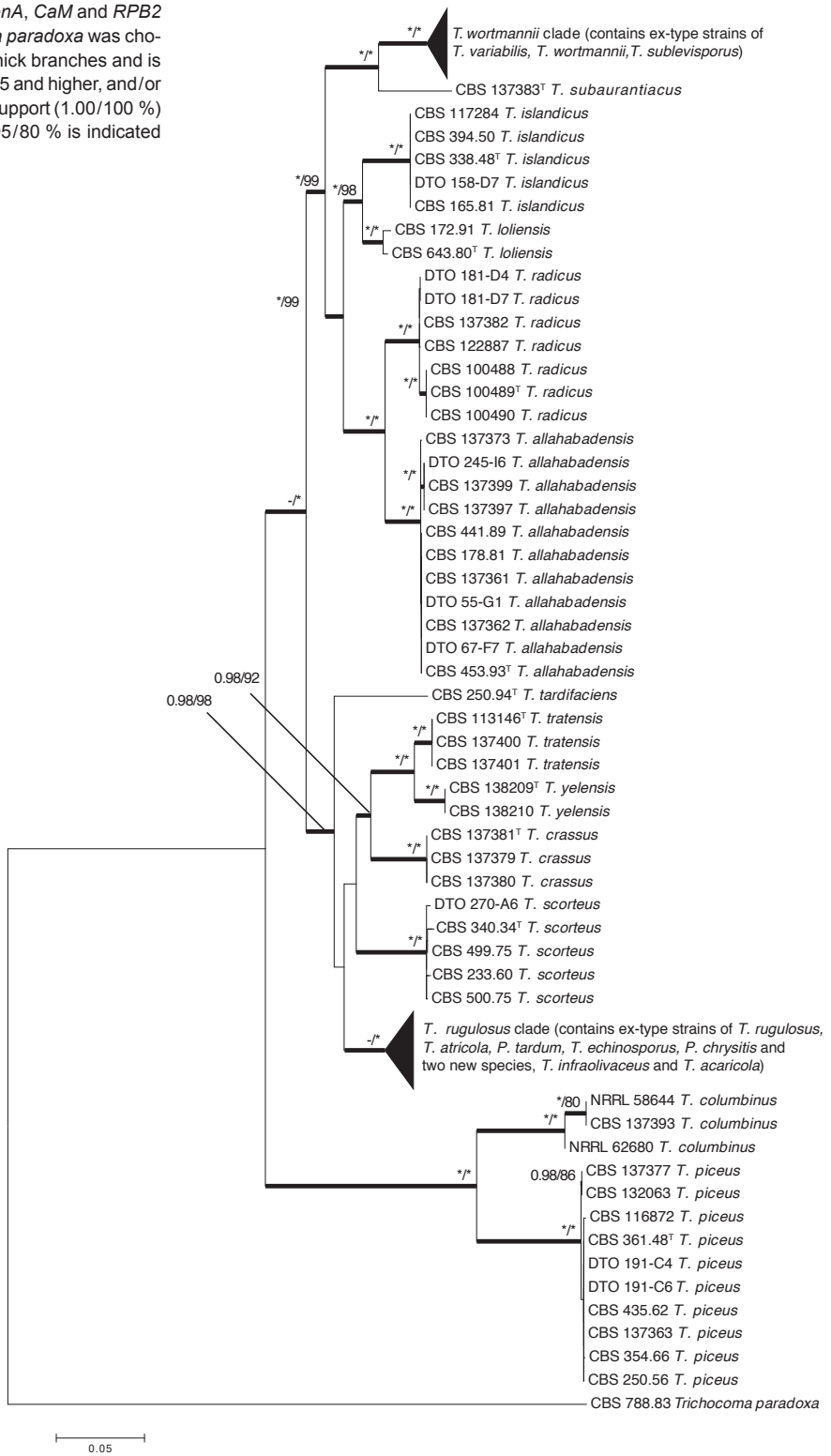

Fig. 1 Combined phylogenetic tree comparing ITS, *BenA*, *CaM* and *RPB2* of species from *Talaromyces* sect. *Islandici*. *Trichocoma paradoxa* was chosen as outgroup. Support in nodes is indicated above thick branches and is represented by posterior probabilities (BI analysis) of 0.95 and higher, and/or bootstrap values (ML analysis) of 80 % and higher. Full support (1.00/100 %) is indicated with an asterisk (*); support lower than 0.95/80 % is indicated with a dash (–). ^{T} = ex type.

species that we describe here as new. *Talaromyces infraolivaceus* and *T. acaricola* are resolved in the *T. rugulosus* complex, while *T. crassus* and *T. subaurantiacus* are closely related to *T. rotundus*, *T. tratensis*, *T. wortmannii* and *T. yelensis*. Differences between these species are discussed in the taxonomy section. Tree topologies differed between phylogenies of different genes in the *T. wortmannii* clade. As such, we adopted the GCPSR concept in this clade, and this is discussed below. GCPSR was also applied for resolving the species in the *T. rugulosus* complex.

For the *T. rugulosus* complex, the aligned datasets were 573 (ITS), 432 (*BenA*), 489 (*CaM*) and 786 (*RPB2*) bp long. The most suitable models for ML were Tamura 3-parameter (T92)+G (ITS), K2 (*BenA*), K2+G (*CaM*) and K2+G (*RPB2*). The most suitable models for BI were Hasegawa-Kishino-Yano 1985 (HKY)+I(ITS), Symmetrical (SYM) (*BenA*), HKY+G (*CaM*) and Kimura 1980 (K80)+G (*RPB2*). The phylogenies (Fig. 2) show that the *T. rugulosus* complex contains four species. *Penicillium tardum* and *P. chrysitis* are synonyms of *T. rugulosus*, confirming results of Peterson & Jurjević (2013). Pitt (1980) proposed *P. echinosporum* as a synonym of *T. rugulosus*, which we confirm here. Peterson & Jurjević (2013) showed that *P. rugulosum* var. *atricolum* is a distinct phylogenetic species and introduced the new combination *T. atricola*, which is accepted here. We also describe two new species as *T. infraolivaceus* and *T. acaricola*. Strains of the latter two species form consistently distinct clades from all other species, which was confirmed by their unique morphological and extrolite characters.

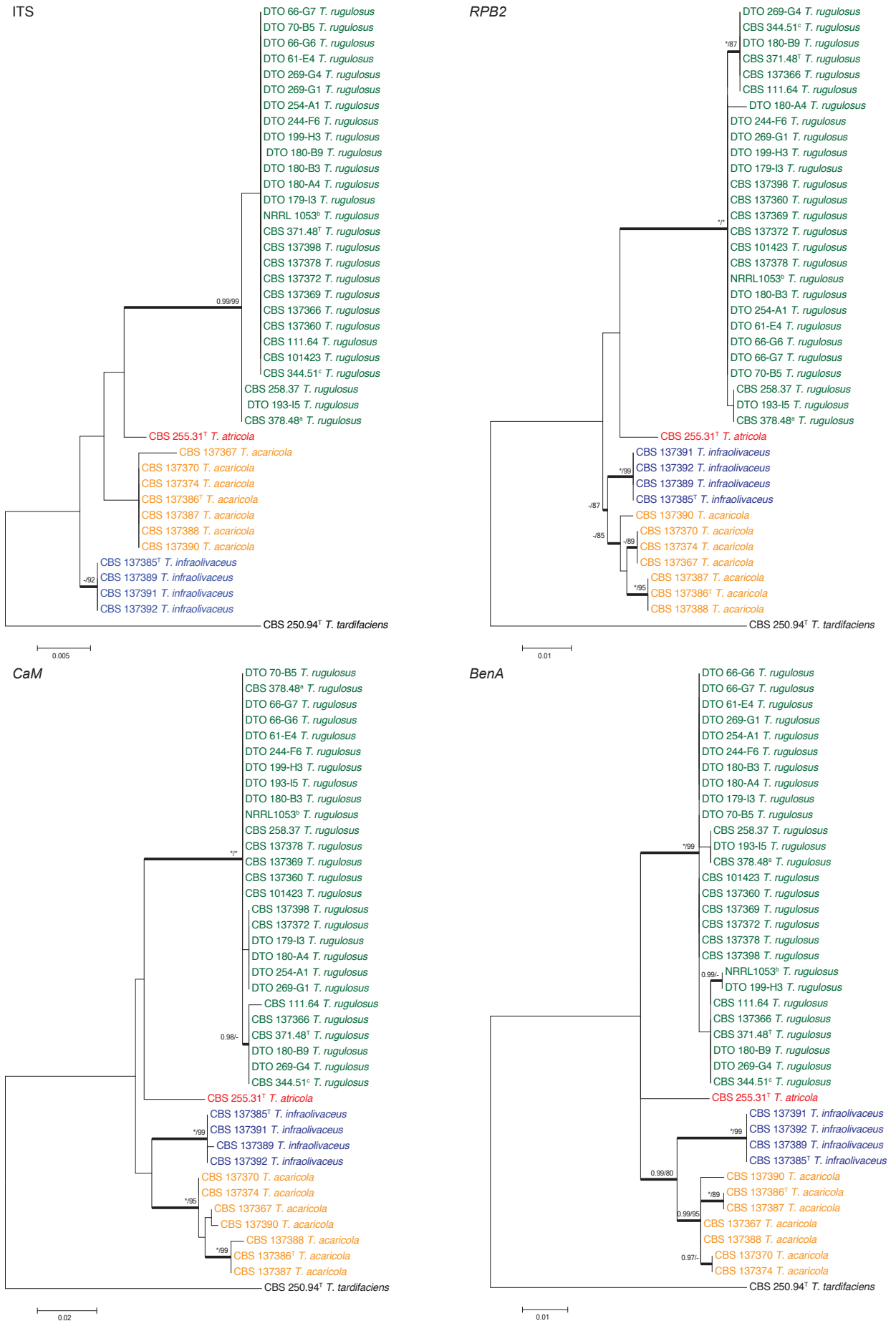

Fig. 2 Phylogenetic trees of the *ITS*, *BenA*, *CaM* and *RPB2* regions of strains in the *T. rugulosus* complex. *Talaromyces tardifaciens* was chosen as outgroup. Support in nodes is indicated above thick branches and is represented by posterior probabilities (BI analysis) of 0.95 and higher, and/or bootstrap values (ML analysis) of 80 % and higher. Full support (1.00/100 %) is indicated with an asterisk (*); support lower than 0.95/80 % is indicated with a dash (–). ^{T} = ex type. ^{a} = ex-type of *P. elongatum* and *P. tardum* (CBS 378.48 = NRRL 1073), ^{b} = ex-type of *P. chrysitis* (NRRL 1053) and ^{c} = ex-type of *P. echinosporum* (CBS 344.51). Colours are used to emphasise species in the clade.

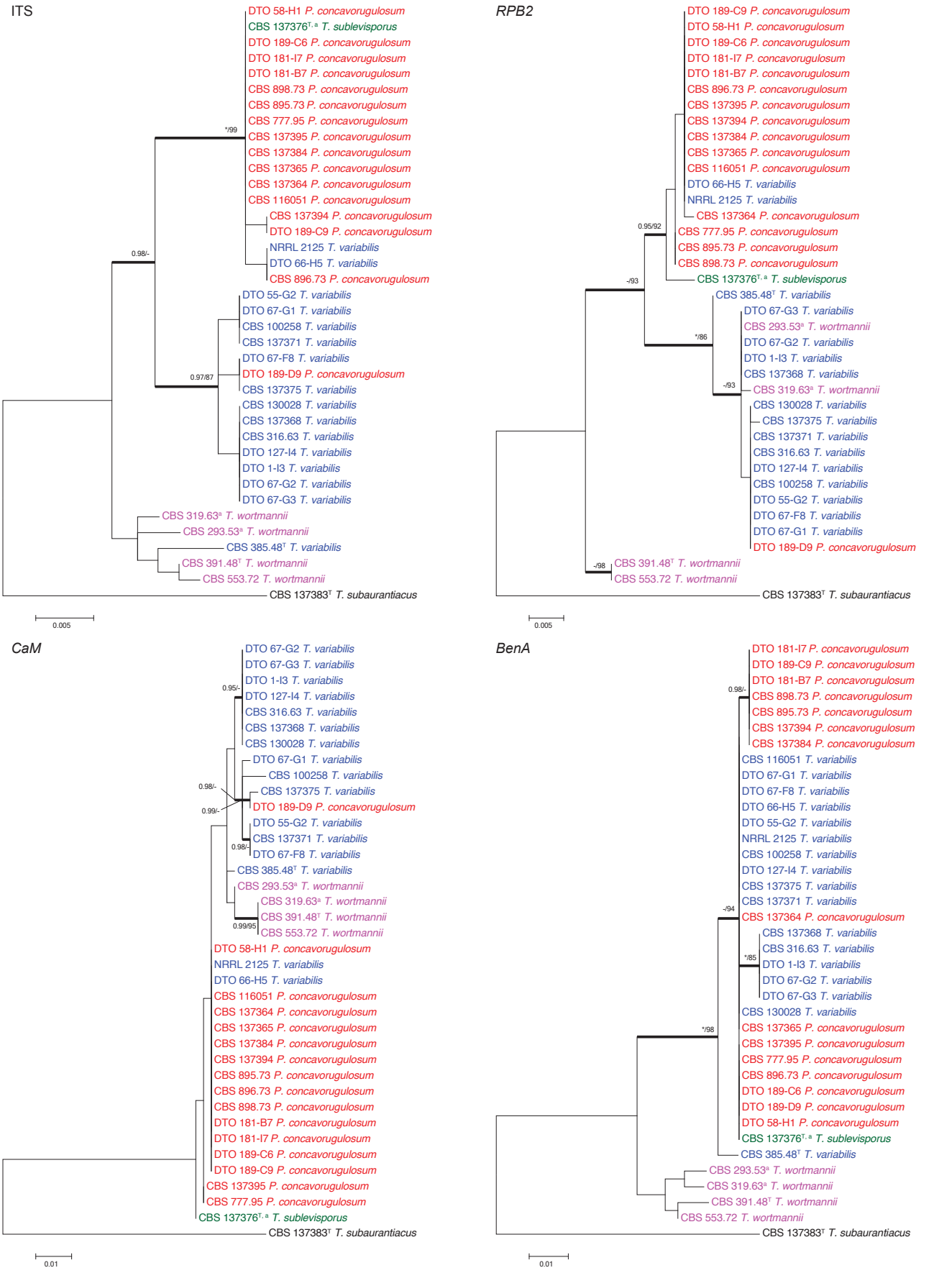

Fig. 3 Phylogenetic trees of the ITS, *BenA*, *CaM* and *RPB2* regions of strains in the *T. wortmannii* clade. *Talaromyces subaurantiacus* was chosen as outgroup. Support in nodes is indicated above thick branches and is represented by posterior probabilities (BI analysis) of 0.95 and higher, and/or bootstrap values (ML analysis) of 80 % and higher. Full support (1.00/100 %) is indicated with an asterisk (*); support lower than 0.95/80 % is indicated with a dash (–). T = ex type. Blue = isolates previously identified as *T. variabilis*; red = isolates previously identified as *P. concavorugulosum*; green = isolate of *T. sublevisporus*; purple = isolates previously identified as *T. wortmannii* and a indicates isolates which produce ascomata.

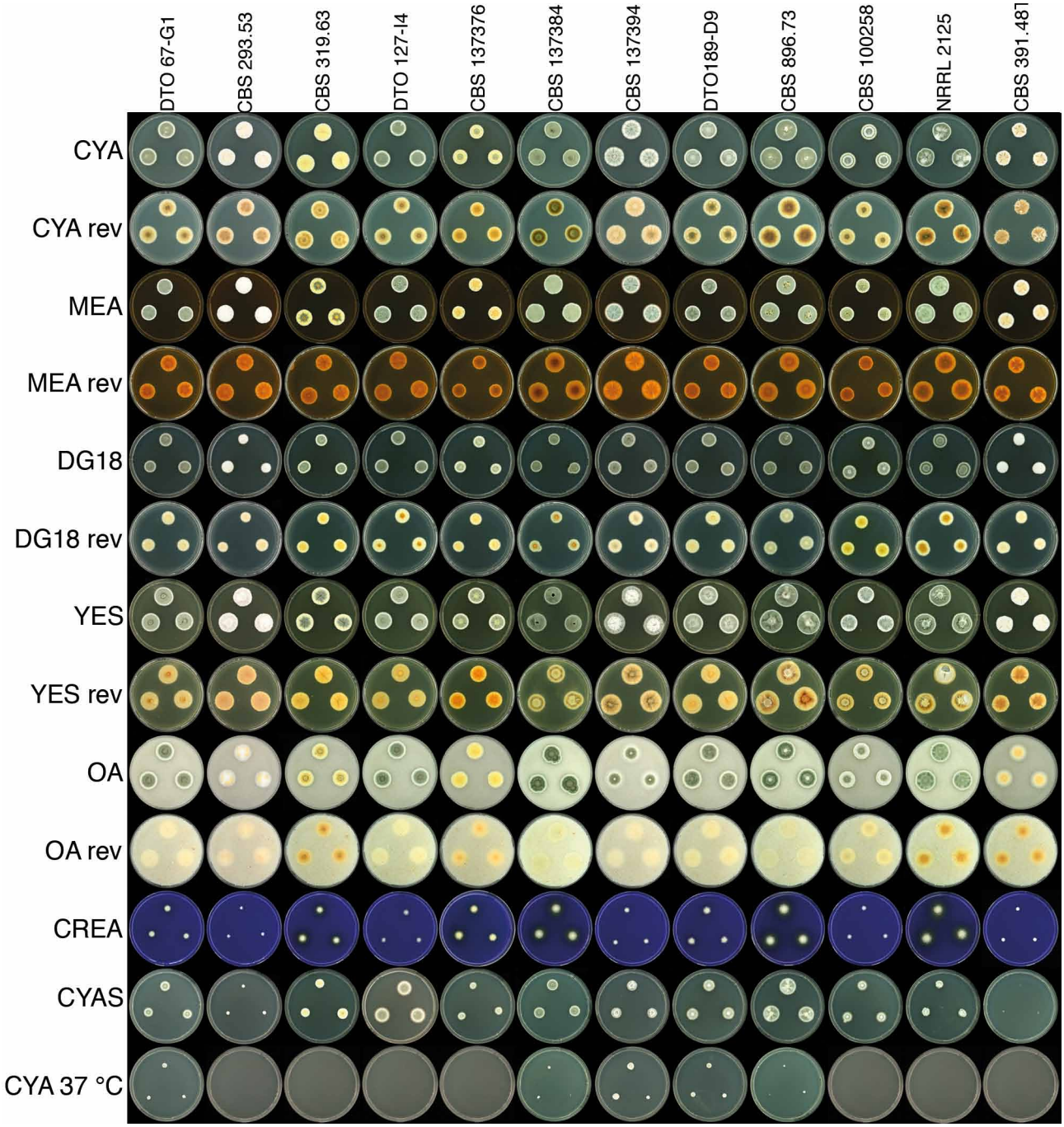

Fig. 4 *Talaromyces wortmannii* colonies grown on various media at different conditions.

For the *T. wortmannii* clade, the aligned datasets were 562 (ITS), 406 (*BenA*), 491 (*CaM*) and 766 (*RPB2*) bp long. The most suitable models for ML were T92+G (ITS), K2+G (*BenA*), K2+G (*CaM*) and K2+G (*RPB2*). The most suitable models for BI were GTR+I (ITS), K80+G (*BenA*), K80+G (*CaM*) and SYM+I (*RPB2*). Four previously described species are resolved in the *T. wortmannii* clade (Fig. 3). The four phylogenies showed different topologies between genes studied. Especially the locations of strains CBS 319.63, CBS 293.53, CBS 553.72 and CBS 391.48 varied. More noticeably, CBS 319.63 and CBS 293.53 are resolved with other *T. wortmannii* strains in all genes except for *RPB2* which resolved them with other *T. variabilis* strains. Similarly, the type of *T. variabilis* (CBS 385.48^{T}) is resolved within a clade of *T. wortmannii* for ITS. These switching positions of strains result in the only consistent branch being the one supporting the entire clade. Because this result was considered strange, DNA was extracted and strains resequenced in order to confirm the result obtained. As such, under GCPSR, strains from these four species are considered to belong to the same species. This is confirmed by our morphological studies, where conidiophores of *T. sublevisporus* and *T. wortmannii* (previously known for their teleomorphs) are identical to that of *P. concavorugulosum* and *T. variabilis*. Extrolite data also supports this. *Penicillium wortmannii* (1903) represents the oldest name in the clade and as a result we synonymise *T. variabilis*, *T. sublevisporus* and *P. concavorugulosum* with *T. wortmannii*.

Morphology

Species were compared morphologically, with characters distinguishing among species summarised in Table 2. The most important characters for identification include growth at 37 °C, colony texture, conidial colour, colony reverse, ascomata production and shape of ascospores. The new species identified by the phylogenetic analyses, displayed various distinct mor-

Table 2 Morphological characters for the identification of *Talaromyces* sect. *Islandici* species.

Talaromyces sp.	Colony diameter (mm)					Reverse coloration on CYA	Texture on MEA	Conidial colour on MEA	Acid production	Conidial size (µm)	Ascomata	Shape, ornamentation and size of ascospores (µm)	Vesiculated stipes
	MEA 25 °C	YES 25 °C	CYA 25 °C	CYA 37 °C	CYA 40 °C								
T. acaricola	15–20	13–16	10–15	NG	NG	Greyish green centre fading into greyish yellow	Velvety to floccose	Dull green	A	2.5–5.5 × 2–3	A	A	A
T. allahabadensis	20–23	22–23	20–25	23–25	NG	Orange centre fading into yellow	Velvety	Dull green	P	2.5–4.5 × 1.7–2.5	A	A	A
T. atricola	15	12	10	NG	NG	Yellowish white	Floccose	Dull green to dark green	A	2–5 × 2–5	A	A	A
T. brunneus	17–19	24–25	19–20	NG	NG	Yellowish brown center fading into golden yellow	Velvety and in the center floccose	Golden brown to yellowish brown	A	3–4(–7) × 2–4	A	A	A
T. columbinus	23–25	18–20	11–12	45–50	43	Dark brown	Velvety and floccose	Greyish green	A	2.5–3.5 × 3–4.5	A	A	P
T. crassus	17–20	15–18	14–16	NG	NG	Pale yellow	Floccose	No sporulation (yellow mycelia dominant)	A to VW	2–3 × 1.5–2.5	A	A	A
T. infraolivaceus	19–21	15–21	17–18	NG	NG	Olive brown	Velvety and loosely funiculose in the centre	Dull green	A to VW	2.5–4 × 1.5–3	A	A	A
T. islandicus	21–26	22–30	20–27	8–17	NG	Orange to brown	Velvety and loosely funiculose	Dull green to dark green	P	2.5–6 × 2–4.2	A	A	A
T. loliensis	13–15	13–15	10–13	NG	NG	Deep orange centre fading into deep yellow	Loosely funiculose to floccose	Greyish green to dark green (yellow mycelia dominant)	A to VW	3–5 × 2.4–3.5	A	A	A
T. piceus	25–27	15–20	20–27	30–35	23–27	Orange to brown	Loosely funiculose to floccose	Greyish green	A	2–3.8 × 2–4	A	A	P
T. radicus	15–25	22–25	15–22	25–30	3–11	Yellowish brown	Loosely funiculose to floccose	Greyish green	A	2–3 × 2–2.5	A	A	A
T. rotundus	15–17	9–10	9–11	NG	NG	Greyish green circle at center fading into greenish grey	No sporulation	No sporulation (white mycelia dominant and at center yellow mycelia)	A (NG)	3–5(–6.5) × 1.5 × 2.5	P (2–3 weeks)	Globose, 4–5.5 × 4–5.5, spinose	A
T. rugulosus	17–20	15–20	15–17	NG	NG	Yellowish brown	Velvety	Greyish green to dark green	A to VW	2.5–6 × 2.5–4	A	A	A
T. scorteus	10–15	7–16	8–16	NG	NG	Olive	Velvety to floccose	Dark green	A	3–5.5 × 2–3	A	A	A
T. subauranticus	20–21	17–18	16–18	7	NG	Yellowish brown to dark brown	Floccose	Dull green	A	2–3 × 2–2.5	A	A	A
T. tardifaciens	13–15	9–10	9–10	NG	NG	Light orange centre fading into greyish yellow	No sporulation	No sporulation (white mycelia dominant)	A (NG)	3–6 × 1.5–2.5	P (3 weeks)	Broadly ovoidal, 3–3.5 × 2–3, smooth	A
T. tratensis	15–20	12–18	10–12	NG	NG	Greyish yellow to brownish orange	Floccose	No sporulation (yellow mycelia dominant)	A	2–2.5 × 3–3.5	P (1–2 weeks)	Ovoidal to broadly ellipsoidal 3.5–5 × 2.5–3.5 µm, thick walled slightly roughed	A
T. wortmannii	15–25	20–30	18–28	NG to 7	NG	Reverse in various colours*	Velvety	Greyish green to dull green	A to VW	2.5–5.8 × 1.5–3.2	A to P (1–2 weeks)	Broadly ellipsoidal, 3.5–6 × 2.5–4 µm, thick walled, verrucose to smooth	A
T. yelensis	15–16	20–21	20–22	14–16	NG	Yellowish white to light yellow to brown	Floccose	No sporulation (yellow mycelia dominant)	A	2.5–3.5 × 2.5–3	A	A	A

NG No Growth.
* In some isolates centre brown fading into in some isolates reddish yellow, in some isolates greyish orange to orange, in some isolates centre yellowish brown fading into in some isolates olive and in some isolates greyish yellow in some isolates with production of ascomata yellow with dark blonde dots in centre.
A Absent.
P Present.
VW Very Weak.

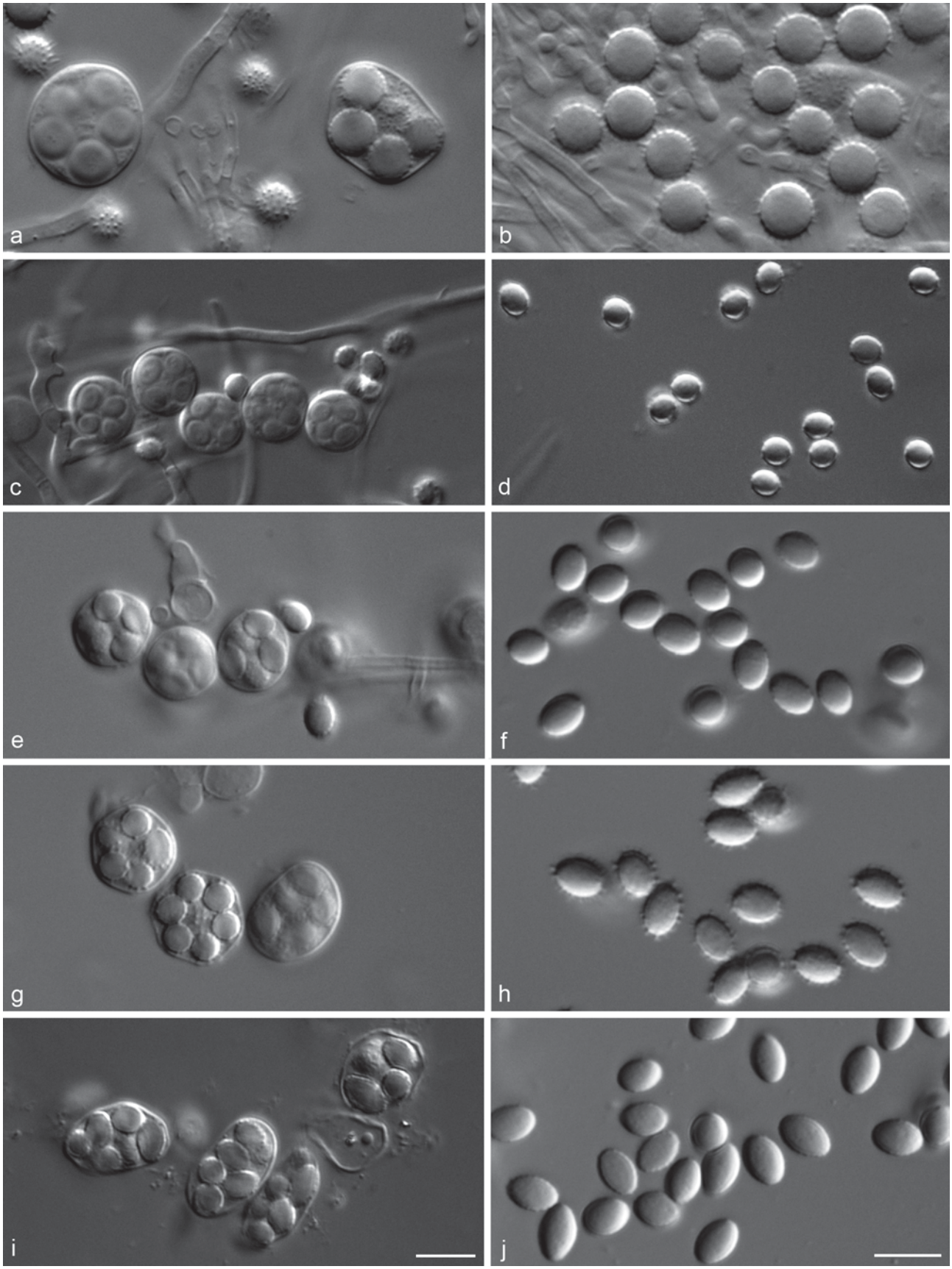

Fig 5 Variations of asci and ascospores produced by different species in *Talaromyces* sect. *Islandici*. a. Asci of *T. rotundus* (CBS 369.48^{T}); b. ascospores of *T. rotundus* (CBS 369.48^{T}); c. asci of *T. tratensis* (CBS 137401); d. ascospores of *T. tratensis* (CBS 137401); e. asci of *T. tardifaciens* (CBS 250.94^{T}); f. ascospores of *T. tardifaciens* (CBS 250.94^{T}); g. asci of *T. wortmannii* (CBS 293.53); h. ascospores of *T. wortmannii* (CBS 293.53); i. asci of *T. wortmannii* (CBS 137376 = ex-type of *T. sublevisporus*); j. ascospores of *T. wortmannii* (CBS 137376 = ex-type of *T. sublevisporus*).

phological features. Descriptions and distinguishing characters for each of the new species are presented below in the taxonomy section.

Strains from the *T. wortmannii* clade were compared morphologically (Fig. 4, 5). In our study, the only strains that produced the characteristic yellow ascomata were CBS 137376, CBS 319.63, CBS 293.53 and CBS 391.48^{T} (Fig. 4). Ascospores of these strains are generally rough-walled. However, CBS 137376^{T}, previously described as *T. wortmannii* var. *sublevisporus* (Yaguchi et al. 1994), produces smooth to finely roughened ascospores (Fig. 5). Yaguchi et al. (1994) mentioned that other characters

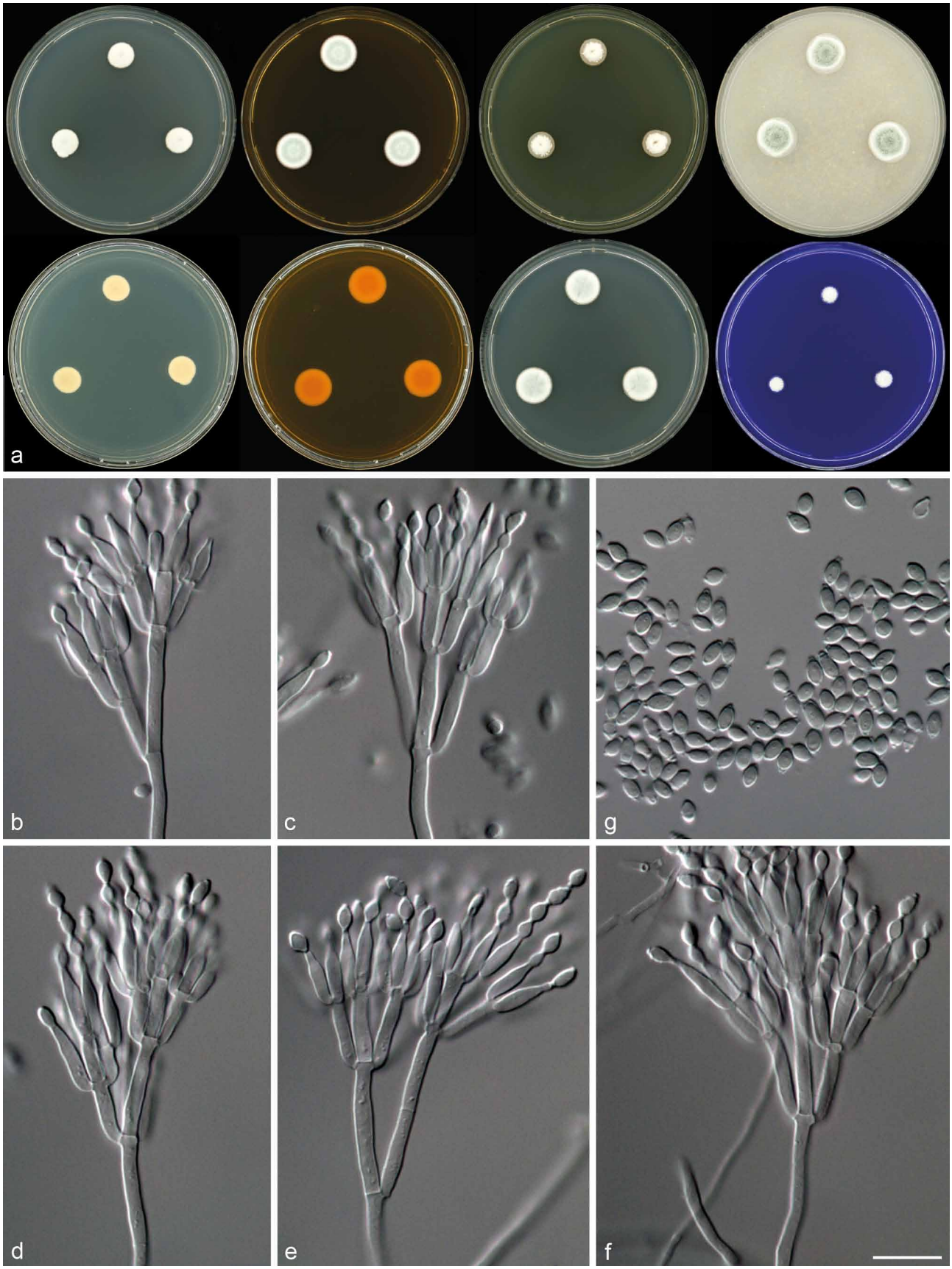

Fig. 6 Morphological characters of *Talaromyces acaricola* (CBS 137386^{T}). a. Colonies from left to right (top row) CYA, MEA, YES and OA; (bottom row) CYA reverse, MEA reverse, DG18 and CREA; b–f. conidiophores; g. conidia. — Scale bar: f = 10 µm, applies to b–g.

of *T. wortmannii* var. *sublevisporus* and *T. wortmannii* were almost identical and this is clear from colony characters shown in Fig. 4. Some strains in the clade lacked ascomata and only produced conidiophores. This was typically of strains previously identified as *P. concavorugulosum* and *T. variabilis*. Abe (1956) never provided a Latin diagnosis for *P. concavorugulosum* species and did not make type material available. As such we synonymise this invalid name based on many strains received from other collections identified as *P. concavorugulosum* by their extrolite profiles. Strains of *T. wortmannii* characteristically produced rugulovasines, rugulosins, skyrin, wortmannilactones E, F, G and H, and mitorubrins. These exometabolites were found in several strains of isolates formerly identified as either *Penicillium variabile* (= *T. variabilis*), *P. concavorugulosum* or *T. wortmannii*.

TAXONOMY

Talaromyces acaricola Visagie, Yilmaz & K. Jacobs, *sp. nov.* — MycoBank MB810899, Fig. 6

ITS barcode. JX091476.
Alternative markers. JX091610 (*BenA*), JX140729 (*CaM*), KF984956 (*RPB2*).

Etymology. Latin (*acarus* = mite), *acaricola*: meaning resident on mites, in reference strains isolated from mites inside *Protea repens* infructescences.

Typus. South Africa, Western Cape, Malmesbury, mite isolated from *Protea repens* infructescence, 2009, collected by *C.M. Visagie* (CBS-H 21632, holotype, culture ex-type CBS 137386 = DTO 183-B3 = DAOM 241025 = IBT 32387).

Diagnosis — Colonies CYA 10–15 mm, MEA 15–20 mm, acid not produced. Conidiophores biverticillate, a minor proportion with subterminal branches; phialides acerose to ampulliform; conidia rough-walled, sometimes forming ridges, ellipsoidal, 2.5–5.5 × 2–3 µm.

Colony diam, 7 d (mm) — CYA 10–15; CYA 37 °C No growth; MEA 15–20; DG18 12–17; CYAS 4–6; OA 10–20; CREA 6–10; YES 13–16.

Colony characters — CYA, 25 °C, 7 d: Colonies raised in the centre, concentrically sulcate; margins narrow (1 mm), low, entire, plane; mycelium white and yellow; texture velvety, in some isolates centre floccose; sporulation moderately dense; conidia *en masse* dull green to dark green (28E4–28F4); exudates absent; soluble pigment absent; reverse centre greyish green (1C4) fading into greyish yellow (1B4). MEA, 25 °C, 7 d: Colonies slightly raised in the centre, crateriform, sulcate; margins narrow (1 mm), low, entire, plane; mycelium white and yellow; texture velvety to floccose; sporulation moderately dense to dense; conidia *en masse* dull green (26D4–27D4); exudates absent; soluble pigment absent; reverse centre olive (1E4–1E5) fading into brownish yellow (5C7–5C8). YES 25 °C, 7 d: Colonies raised at centre, crateriform; margins narrow (1 mm), low, entire, plane; mycelium white and in some isolates yellow; texture velvety and floccose; sporulation sparse to moderately dense; conidia *en masse* dull green (27D4–27E4); exudates clear or yellow droplets (except CBS 137367 and CBS 137374); soluble pigment absent; reverse light yellow to greyish yellow (4A5–4B5), centre greyish green (1C4) in some isolates. DG18, 25 °C, 7 d: Colonies raised in the centre, crateriform, sulcate; margins narrow (1 mm), low, entire, plane; mycelium white and yellow; texture velvety, centre floccose in some isolates; sporulation dense; conidia *en masse* greyish green to dull green (25C4–25C5 to 25D4–25D5); exudates absent; soluble pigment absent; in some isolates reverse centre greyish green (1C5–1D5), in others reddish yellow to greenish yellow (4A6–4B6), fading into light yellow to greyish yellow (1A5–1B5). OA, 25 °C, 7 d: Colonies low, plane; margins narrow (1–2 mm), low, entire, plane, in some isolates with yeast like slimy margins; mycelium white and yellow; texture velvety and loosely funiculose; sporulation dense; conidia *en masse* greyish green to dull green (27C4–27C5 to 27D4–27D5); exudates absent; soluble pigment absent; reverse yellow to greenish yellow. CREA, 25 °C, 7 d: Acid not produced.

Micromorphology — Conidiophores biverticillate, a minor proportion with subterminal branches; stipes smooth-walled, 40–160 × 2–3 µm, branches 2–3 per stipe, 14–22 × 2–3 µm; metulae 3–5, 7.5–12 × 2–3 µm; phialides acerose to ampulliform, 3–5 per metulae, 6.5–9.5 × 2–3 µm; conidia rough-walled, sometimes forming ridges, ellipsoidal, 2.5–5.5 × 2–3 µm.

Extrolites — *Talaromyces acaricola* produces mitorubrins, rugulosin, skyrin, ukulactones and a polar metabolite with a chromophore similar to calbistrins.

Distinguishing characters — *Talaromyces acaricola* is characterised by typically floccose colonies especially on CYA and YES. The phylogenies resolve *T. acaricola* in the *T. rugulosus* complex (Fig. 2), closely related to *T. rugulosus*, *T. atricola* and *T. infraolivaceus* (Fig. 2). *Talaromyces acaricola* differs from *T. rugulosus* by the production of lightly coloured conidia *en masse* and MEA colonies that are more floccose in *T. acaricola* compared to the velvety colonies of *T. rugulosus*. It differs from *T. infraolivaceus* by greyish green or greyish yellow rather than dark olive reverse pigmentation and grows faster than *T. atricola* on most media.

Talaromyces crassus Visagie, Yilmaz & K. Jacobs, *sp. nov.* — MycoBank MB810900, Fig. 7

ITS barcode. JX091472.
Alternative markers. JX091608 (*BenA*), JX140727 (*CaM*), KF984914 (*RPB2*).

Etymology. Latin, *crassus*: meaning thick, in reference to the thick deep colonies produced.

Typus. South Africa, Western Cape, Stellenbosch, *Protea repens* infructescence, 2009, collected by *C.M. Visagie* (CBS-H 21631, holotype, culture ex-type CBS 137381 = DTO 181-C5 = DAOM 241027 = IBT 32814).

Diagnosis — Colonies on CYA 14–16 mm, MEA 17–20 mm. Acid generally not produced, some isolates weakly positive. Thick, deep, fluffy, yellow colonies on MEA. Conidiophores biverticillate, a minor proportion with subterminal branches; phialides acerose; conidia smooth-walled, ellipsoidal, 2–3 × 1.5–2.5 µm.

Colony diam, 7 d (mm) — CYA 14–16; CYA 37 °C No growth; MEA 17–20; DG18 12–16; CYAS 8–10; OA 16–20; CREA 6–10; YES 15–18.

Colony characters — CYA 25 °C, 7 d: Colonies low, plane; margins narrow (1 mm), low, entire, plane; mycelium white and predominately pale yellow; sporulation moderately dense to dense, especially in the centre; texture velvety to funiculose, conidiophores borne from aerial hyphae especially in the centre; conidia *en masse* dull green (25D4); exudates absent; soluble pigment absent; reverse pale yellow (4A3), in some isolates the centre greyish orange (5B3–5B4). MEA, 25 °C, 7 d: Colonies slightly raised in the centre, slightly concentrically sulcate and crateriform; margins narrow (1 mm), low, entire, plane; mycelium white and predominately yellow; sporulation none to sparse (very difficult to determine the conidia colour); texture floccose; exudates absent; soluble pigment absent; reverse brownish yellow (5C7–5C8). YES, 25 °C, 7 d: Colonies slightly raised in the centre, slightly crateriform and very slightly sulcate; margins narrow (1–2 mm), low, entire, plane; mycelium white and yellow; sporulation sparse to moderately dense; texture floccose; conidia *en masse* dull green (25D4–26D4); exudates small and clear droplets; soluble pigment absent; reverse butter yellow (4A5). DG18, 25 °C, 7 d: Colonies slightly raised in the centre, slightly sulcate; margins narrow (1 mm), low, entire, plane; myce-

lium white; sporulation none, not enough to determine colour; texture floccose; exudates absent; soluble pigment absent; reverse yellowish white and in some isolates greenish grey (1A2 and sometimes 1B2). OA, 25 °C, 7 d: Colonies low, plane; margins narrow (1 mm), low, entire, plane; mycelium white and predominately yellow; sporulation moderately dense to dense, especially in the centre; conidia *en masse* dull green (29E3–29E4); texture floccose and funiculose, conidiophores borne from aerial hyphae especially in the centre; exudates small and clear droplets; soluble pigment absent; reverse very pale light yellow, in some isolates centre dark green. CREA, 25 °C, 7 d:

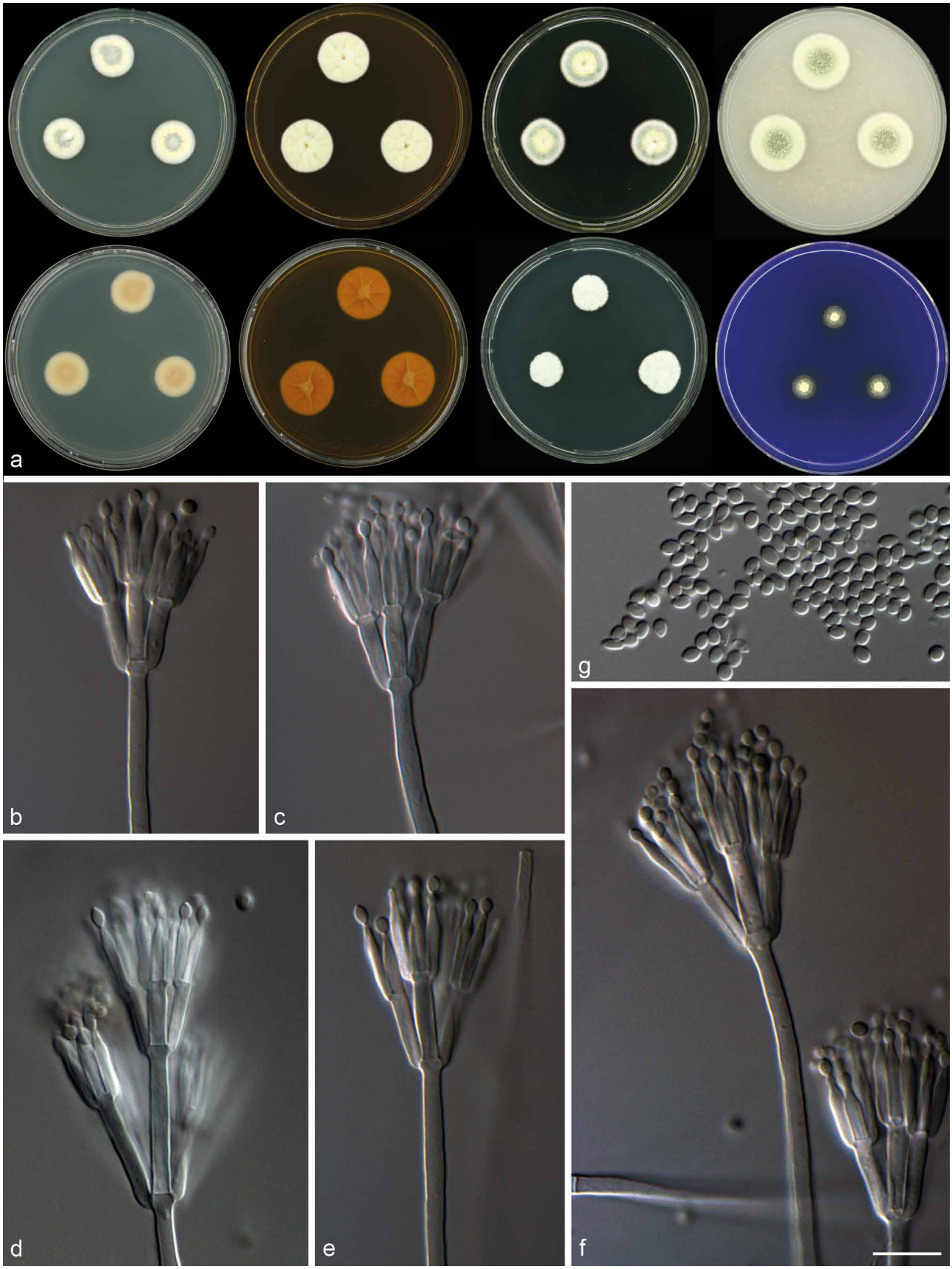

Fig. 7 Morphological characters of *Talaromyces crassus* (CBS 137381). a. Colonies from left to right (top row) CYA, MEA, YES and OA; (bottom row) CYA reverse, MEA reverse, DG18 and CREA; b–f. conidiophores; g. conidia. — Scale bar: f = 10 µm, applies to b–g.

Acid production generally absent, in some isolates very weak acid production (CBS 137380).

Micromorphology — Conidiophores biverticillate, a minor proportion with subterminal branches; stipes smooth-walled, 130–390 × 2.5–3.5 µm; branches 2–3 per stipe, 13–17 × 2.5–3.5 µm, metulae 3–6, 9.5–14 × 2.5–3 µm; phialides acerose, number per metulae 3–6, 8.5–11.5 × 1.5–2.5 µm; conidia smooth-walled, ellipsoidal, 2–3 × 1.5–2.5 µm.

Extrolites — The ex-type isolate CBS 137381^{T}, CBS 137379 and CBS 137380 only produced mitorubrins.

Distinguishing characters — *Talaromyces crassus* has restricted growth on most media, similar to other sect. *Islandici*

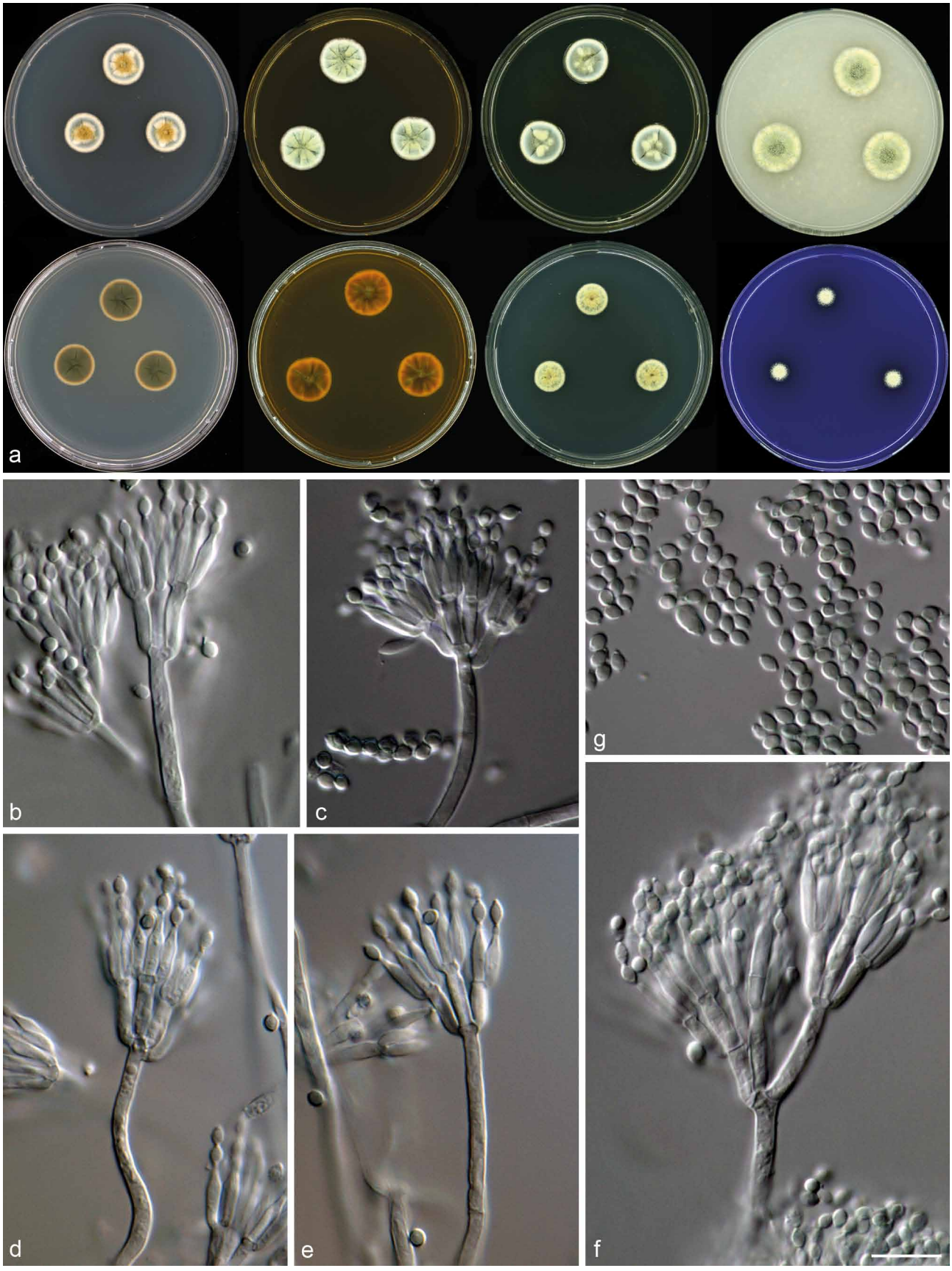

Fig. 8 Morphological characters of *Talaromyces infraolivaceus* (CBS 137385^{T}). a. Colonies from left to right (top row) CYA, MEA, YES and OA; (bottom row) CYA reverse, MEA reverse, DG18 and CREA; b–f. conidiophores; g. conidia. — Scale bar: f = 10 µm, applies to b–g.

species. Colonies are characteristically deep and consist of light yellowish mycelia that produce a fluffy texture. It cannot grow at 37 °C. Based on the multi-gene phylogeny, *T. crassus* is closely related to *T. tratensis* and the recently described *T. yelensis* (Visagie et al. 2014a) (Fig. 1). Morphologically all three species produce deep, fluffy yellow colonies. However, *T. tratensis* produces ascomata and spiny ascospores, which are absent in *T. crassus* and *T. yelensis*. *Talaromyces yelensis* is able to grow at 37 °C (14–16 mm), distinguishing it from *T. crassus*.

Talaromyces infraolivaceus Visagie, Yilmaz & K. Jacobs, *sp. nov*. — MycoBank MB810901, Fig. 8

ITS barcode. JX091481.
Alternative markers. JX091615 (*BenA*), JX140734 (*CaM*), KF984949 (*RPB2*).

Etymology. Latin, *infraolivaceus*: meaning below olive, in reference to the olive reverse of colonies.

Typus. South Africa, Western Cape, Malmesbury, mite isolated from *Protea repens* infructescence, 2009, collected by *C.M. Visagie* (CBS-H 21633, holotype, culture ex-type CBS 137385 = DTO 182-I2 = DAOM 241024 = IBT 32487).

Diagnosis — Colonies CYA 17–18 mm, MEA 19–21 mm. Acid generally not produced, some isolates weakly positive. Consistent production of deep olive reverses on most media. Conidiophores biverticillate, a minor proportion with subterminal branches and sometimes terverticillate; phialides acerose to ampulliform; conidia rough-walled, sometimes in ridges, ellipsoidal, 2.5–4 × 1.5–3 µm.

Colony diam, 7 d (mm) — CYA 17–18; CYA 37 °C No growth; MEA 19–21; DG18 12–13; CYAS 8–10; OA 18–20; CREA 5–8; YES 15–21.

Colony characters — CYA 25 °C, 7 d: Colonies slightly raised at centre, crateriform, radially sulcate; margins narrow (1 mm), low, entire, plane; mycelium white and very pale yellow; sporulation dense; texture velvety and loosely funiculose at centre, conidiophores borne from aerial hyphae especially in the centre; conidia *en masse* dull green (26D4–26E4–27E4); exudates absent; soluble pigment absent; reverse centre olive brown (4F5) fading into golden brown to light brown (5D7). MEA 25 °C, 7 d: Colonies slightly raised at centre, sulcate and in some isolates crateriform; margins narrow (1 mm), low, entire, plane; mycelium white and very pale yellow; sporulation moderately dense to dense; texture velvety and loosely funiculose, conidiophores borne from aerial hyphae especially in the centre; conidia *en masse* dull green (26D4–26E4–27E4); exudates absent; soluble pigment absent; reverse centre olive brown (4F5) fading into golden brown to light brown (5D7). YES 25 °C, 7 d: Colonies raised at centre, crateriform, sulcate; margins narrow (1–2 mm), low, entire, plane; mycelium white and very pale yellow; sporulation dense; texture velvety; conidia *en masse* dull green (26D4–26E4–27E4); exudates absent; soluble pigment absent; reverse centre olive brown (4F3–4F4 to 4D5–4E5). DG18, 25 °C, 7 d: Colonies raised in the centre, in some isolates crateriform, sulcate; margins narrow (1 mm), low, entire, plane; mycelium white and very pale yellow; texture loosely funiculose, conidiophores borne from aerial hyphae especially at centre; sporulation sparse to dense (CBS 137392, CBS 137389); conidia *en masse* dull green (26E4–27E4); exudates absent (except CBS 137391); soluble pigment absent; reverse centre olive brown (4F5) fading into golden brown to light brown (5D7). OA 25 °C, 7 d: Colonies low, plane; margins narrow (1–2 mm), low, entire, plane; mycelium white and very pale yellow; sporulation dense; texture velvety and loosely funiculose; conidia *en masse* dull green (26D4–26E4–27E4); exudates absent and in some isolates small clear droplets; soluble pigment absent; reverse brownish olive green fading into brownish yellow. CREA, 25 °C, 7 d: Acid generally not produced, some isolates weakly positive (CBS 137385^T and CBS 137389).

Micromorphology — Conidiophores biverticillate, a minor proportion terverticillate and with subterminal branches; stipes smooth-walled, 12–100 × 2–3 µm, branches 2–3 per stipe when present, 11–15 × 2–3 µm; metulae 4–6, 7.5–11.5 × 2–3 µm; phialides acerose to ampulliform, 3–4 per metulae, 7–10 × 1.5–2.5 µm; conidia rough-walled, sometimes in ridges, ellipsoidal, 2.5–4 × 1.5–3 µm.

Extrolites — Isolates in this species produce mitorubrins, viomellein, vioxanthin and xanthomegnin. This is the first report of production of the xanthomegnin in *Talaromyces*. Xanthomegnins have formerly been found in *Penicillium* spp., *Aspergillus* spp., *Trichophyton* spp. and similar genera. In addition, CBS 137389 and CBS 137385^T produce a compound suggesting a polar calbistrin.

Distinguishing characters — *Talaromyces infraolivaceus* is characterised by a consistent production of deep olive reverse on most media. Based on the phylogenies, *T. infraolivaceus* is resolved in the *T. rugulosus* complex (Fig. 1), closely related to *T. rugulosus*, *T. atricola* and *T. acaricola* (Fig. 2). *Talaromyces infraolivaceus* differs from *T. rugulosus* by the production of lightly coloured conidia *en masse* and MEA colonies that are more floccose. The most distinct feature, however, is the dark olive reverse on most media. This dark reverse was not observed in any other species from this clade.

Talaromyces subaurantiacus Visagie, Yilmaz & K. Jacobs, *sp. nov*. — MycoBank MB810902, Fig. 9

ITS barcode. JX091475.
Alternative markers. JX091609 (*BenA*), JX140728 (*CaM*), KF984960 (*RPB2*).

Etymology. Latin, *subaurantiacus*: named in reference to the light orange mycelium produced by this species.

Typus. South Africa, Western Cape, Stellenbosch, Fynbos soil, 2009, collected by *C.M. Visagie* (CBS-H 21630, holotype, culture ex-type CBS 137383 = DTO 181-I2 = DAOM 241020 = IBT 32383).

Diagnosis — Colonies CYA 16–18 mm, MEA 20–21 mm, CYA at 37 °C 7 mm. Acid not produced. Colonies produce orange mycelia on MEA and CYA. Conidiophores biverticillate; phialides acerose; conidia finely rough-walled, ellipsoidal, 2–3 × 2–2.5 µm.

Colony diam, 7 d (mm) — CYA 16–18; CYA 37 °C 7; MEA 20–21; DG18 15–17; CYAS 9–12; OA 17–18; CREA 3–4; YES 17–18.

Colony characters — CYA 25 °C, 7 d: Colonies raised at centre, crateriform; margins very narrow (1 mm), low, entire, plane; mycelium white and pale yellow to light orange in the centre; texture floccose; sporulation sparse; conidia *en masse* dull green (26D4–26E4 to 27D4–27E4); exudates absent; soluble pigment absent; reverse yellowish brown to dark brown (5F5–6F5). MEA 25 °C, 7 d: Colonies low, sulcate; margins very narrow (1 mm), low, entire, plane; mycelium white and pale light orange in the centre; texture floccose; sporulation moderately dense; conidia *en masse* dull green (26D4–26E4 to 27D4–27E4); exudates absent; soluble pigment absent; reverse brown (6E5–6E6). YES 25 °C, 7 d: Colonies raised at centre, crateriform; margins very narrow (1 mm), low, entire, plane; mycelium white; texture floccose; sporulation moderately dense; conidia *en masse* dull green (26D4–26E4 to 27D4–27E4); exudates absent; soluble pigment absent; reverse yellowish brown (5E4) in the centre fading into greyish yellow (4B4). DG18 25 °C, 7 d: Colonies, slightly raised at centre, slightly sulcate; margins very narrow (1 mm), low, entire, plane; mycelium white; texture velvety and in the centre floccose; sporulation moderately dense; conidia *en masse* greyish green

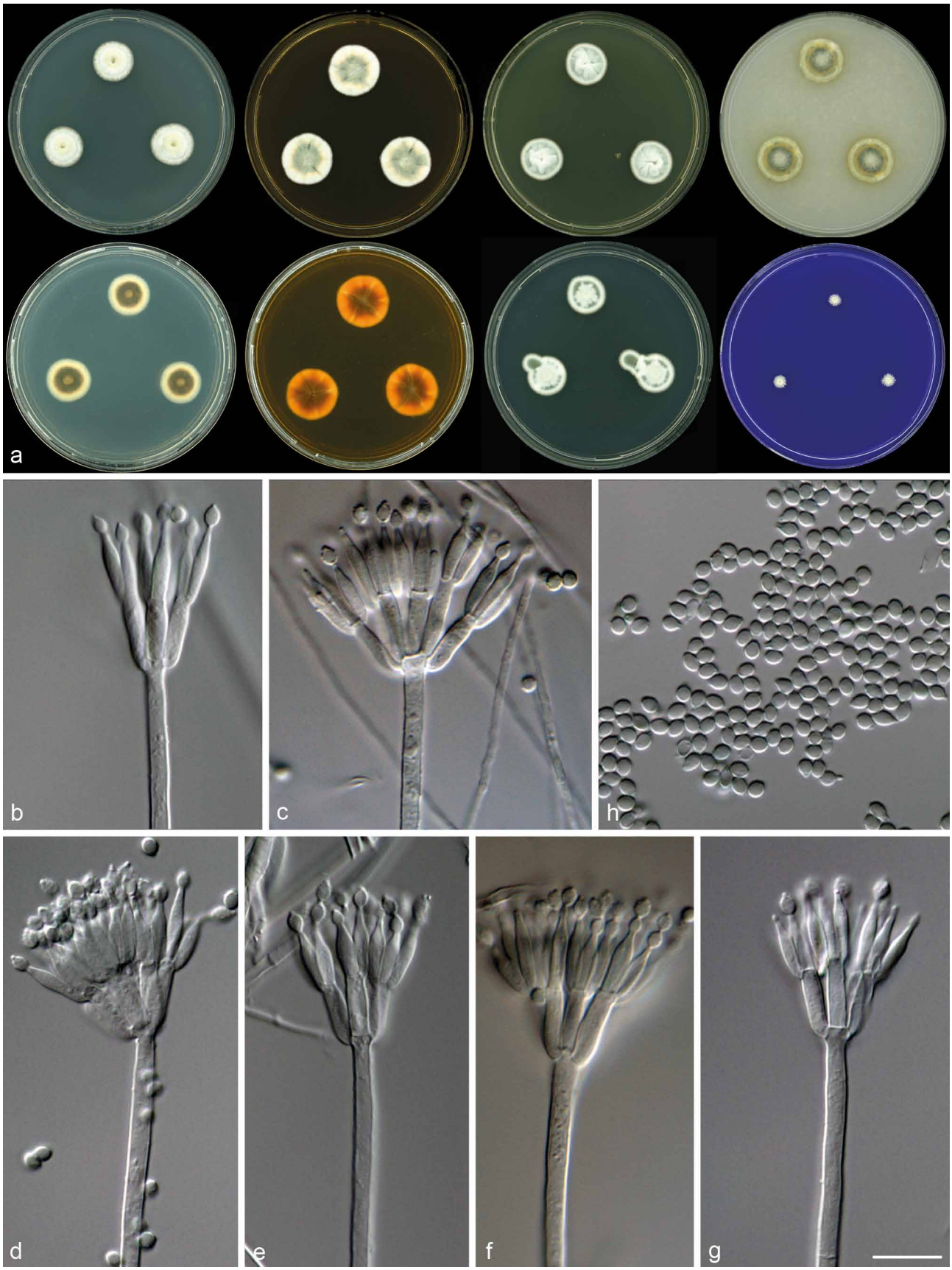

Fig. 9 Morphological characters of *Talaromyces subaurantiacus* (CBS 137383^{T}) a. Colonies from left to right (top row) CYA, MEA, YES and OA; (bottom row) CYA reverse, MEA reverse, DG18 and CREA; b–g. conidiophores; h. conidia. — Scale bar: g = 10 µm, applies to b–h.

to dull green (27E4–27E5)*;* exudates absent; soluble pigment absent; reverse greyish orange to brownish orange (6B4–6C4) in the centre, fading into light yellow to greyish yellow (3A5–3B5). OA 25 °C, 7 d: Colonies low, plane; margins very narrow (1 mm), low, entire, plane; mycelium white and yellow; texture velvety and in the centre floccose, conidiophores borne from aerial hyphae; sporulation moderately dense; conidia *en masse* dark green (25F5); exudates absent; soluble pigment absent; reverse bright orange yellow. CREA, 25 °C, 7 d: Acid not produced.

Micromorphology — Conidiophores biverticillate; stipes smooth-walled, 50–285 × 2.5–3.5 µm; metulae 3–6, 9–13 ×

2–3.5 µm; phialides acerose, 3–6 per metulae, 8.5–11 × 2–3 µm; conidia finely rough-walled, ellipsoidal, 2–3 × 2–2.5 µm.

Extrolites — *Talaromyces subaurantiacus* produces rugulovasine and an azaphilone extrolite related to sclerotiorin.

Distinguishing characters — *Talaromyces subaurantiacus* grows restrictedly on agar media, especially on CYA. Based on the multi-gene phylogeny, *T. subaurantiacus* is closely related to *T. wortmannii* (Fig. 1). However, orange mycelia, floccose texture on MEA and more appressed conidiophores distinguish the new species from *T. wortmannii.*

DISCUSSION

In this study we revised the taxonomy of *Talaromyces* sect. *Islandici*, a group easily recognised by its slow or restricted growth and conspicuous yellow aerial mycelium, using morphology, phylogeny (under GCPSR) and extrolite data. Based on our GCPSR results, sect. *Islandici* includes 19 species, including the four new species. These include *T. acaricola* with its typically floccose colonies on CYA and YES, *T. crassus* producing the typical deep, fluffy colonies with abundance of yellowish mycelia but unable to grow at 37 °C, *T. infraolivaceus* with a unique deep olive reverse on most media, and *T. subaurantiacus* with its generally restricted colonies, especially on CYA. The distinguishing characters for all accepted species are listed in Table 2.

Most species of sect. *Islandici* produce the mycotoxins rugulosin and/or skyrin, the only exceptions being *T. subaurantiacus*, *T. scorteus* and *T. infraolivaceus*, which seem to have lost the ability to produce these bisanthraquinones during their evolution. However, *T. infraolivaceus* has acquired/retained the ability to produce xanthomegnin, viomellein and vioxanthin, which are absent in all other *Talaromyces* species. Rugulosin/skyrin are only produced by species in this section and not in other *Talaromyces* species (Frisvad et al. 1990), except for *T. rubicundus* (Reenen-Hoekstra et al. 1990). The azaphilones known as mitorubrins are produced by nearly all species of *Talaromyces* but are not produced by *T. subaurantiacus*, *T. rotundus* and *T. columbinus*.

Generally speaking, species able to grow at body temperature (37 °C) can be considered a risk as opportunistic pathogens. *Talaromyces allahabadensis*, *T. columbinus*, *T. islandicus*, *T. piceus*, *T. radicus*, *T. subaurantiacus* and *T. yelensis* are able to grow at 37 °C and some strains of *T. wortmannii* and its synonyms *T. variabilis*, *T. sublevisporus* and *P. concavorugulosum. Talaromyces piceus*, *T. columbinus* and *T. radicus* are able to grow at 40 °C (Table 2). Some opportunistic pathogen cases for *T. piceus* and *T. radicus* have been previously reported (Horré et al. 2001, Santos et al. 2006, de Vos et al. 2009).

Previous studies showed a close relationship between *T. wortmannii*, *T. variabilis*, *T. sublevisporus* and *P. concavorugulosum* (Frisvad et al. 1990, LoBuglio et al. 1993, Hocking et al. 1998, Samson et al. 2011, Yilmaz et al. 2014). Hocking et al. (1998) revealed a very high similarity between *T. variabilis* and *T. wortmannii* by using a RAPD-PCR (random amplification of polymorphic DNA). Frisvad et al. (1990) and Yilmaz et al. (2014) showed that these species have many metabolites in common. Peterson & Jurjević (2013), using an *RPB2* phylogeny of only ex-type strains, considered these four species distinct, but hinted that additional analyses of more isolates and more loci were required to establish a robust phylogeny for this complex. We provide this phylogeny here applying GCPSR and reveal that *P. concavorugulosum*, *T. sublevisporus* and *T. variabilis* should be considered synonyms of *T. wortmannii* (Fig. 3). In Fig. 3, it is clear that ascosporic and non-ascosporic strains are mixed within the different clades. Furthermore, Raper & Thom (1949) reported that *T. variabilis* (= *P. variabile*) strains such as NRRL 2125 were received as non-ascosporic cultures, but after numerous transfers, yellow ascomata developed in colonies after three to four weeks. According to the International Code of Nomenclature for algae, fungi and plants (ICN), after 2011, priority is given to the oldest name irrespective of whether the

Table 3 Overview of taxonomic treatments on *Talaromyces* sect. *Islandici.*

Original names	Raper & Thom (1949)	Pitt (1980)	Samson et al. (2011)	Peterson & Jurjević (2013)	Current study
T. acaricola (current study)	–	–	–	–	*T. acaricola*
P. allahabadense (Mehrotra & Kumar 1962)	–	*P. pinophilum*	*T. allahabadensis*	*T. allahabadensis*	*T. allahabadensis*
P. rugulosum var. atricolum (Thom 1930)	*P. tardum*	*P. rugulosum*	*T. rugulosus*	*T. atricola*	*T. atricola*
P. brunneum (Udagawa 1959)	–	*P. brunneum*	*T. brunneus*	not studied	*T. brunneus*
T. columbinus (Peterson & Jurjević 2013)	–	–	–	*T. columbinus*	*T. columbinus*
T. crassus (current study)	–	–	–	–	*T. crassus*
T. infaolivaceus (current study)	–	–	–	–	*T. infaolivaceus*
P. islandicum (Sopp 1912)	*P. islandicum*	*P. islandicum*	*T. islandicus*	*T. islandicus*	*T. islandicus*
P. loliense (Pitt 1980)	–	*P. loliense*	*T. loliensis*	*T. loliensis*	*T. loliensis*
P. piceum (Raper & Fennell 1948)	–	*P. piceum*	*T. piceus*	*T. piceus*	*T. piceus*
P. radicum (Hocking et al. 1998)	–	–	*T. radicus*	*T. radicus*	*T. radicus*
P. rotundum (Raper & Fennell 1948)	*P. rotundum*	*T. rotundus*	*T. rotundus*	*T. rotundus*	*T. rotundus*
P. rugulosum (Thom 1910)	*P. rugulosum*	*P. rugulosum*	*T. rugulosus*	*T. rugulosus*	*T. rugulosus*
P. echinosporum (Nehira 1933)	not studied	*P. rugulosum*	*T. echinosporus*	not studied	*T. rugulosus*
P. tardum (Thom 1930)	*P. tardum*	*P. rugulosum*	*T. rugulosus*	*T. rugulosus*	*T. rugulosus*
P. elongatum (Bainier 1907)	*P. tardum*	*P. rugulosum*	*T. rugulosus*	not studied	*T. rugulosus*
P. chrysitis (Biourge 1923)	*P. rugulosum*	*P. rugulosum*	*T. rugulosus*	*T. rugulosus*	*T. rugulosus*
P. scorteum (Takedo et al. 1934)	*P. tardum*	*P. rugulosum*	*T. rugulosus*	*T. scorteus*	*T. scorteus*
P. phialosporum (Udagawa 1959)	–	*P. rugulosum*	*T. phialosporus*	*T. scorteus*	*T. scorteus*
T. subaurantiacus (current study)	–	–	–	–	*T. subaurantiacus*
T. tardifaciens (Udagawa 1993)	–	–	*T. tardifaciens*	not studied	*T. tardifaciens*
T. tratensis (Manoch et al. 2013)	–	–	–	not studied	*T. tratensis*
T. wortmannii var. *sublevisporus* (Yaguchi et al. 1994)	–	–	*T. sublevisporus*	not studied	*T. wortmannii*
P. concavorugulosum (Abe 1956, nom. Inval., art. 36)	–	*P. rugulosum*	*P. concavorugulosum**	*P. concavorugulosum*	*T. wortmannii*
P. variabile (Sopp 1912)	*P. variabile*	*P. variabile*	*T. variabilis*	*T. variabilis*	*T. wortmannii*
P. wortmannii (Klöcker 1903)	*P. wortmannii*	*T. wortmannii*	*T. wortmannii*	*T. wortmannii*	*T. wortmannii*
T. yelensis (Visagie et al. 2014a)	–	–	–	–	*T. yelensis*

* Samson et al. (2011) listed *P. concavorugulosum* in the 'Taxa which need further taxonomic study' list.
– species which were described later than the study.

species was described as an anamorph or teleomorph (McNeil et al. 2012). In this case *P. wortmannii*, described by Klöcker (1903), is the oldest name in the clade. *Talaromyces wortmannii* was later introduced for the sexual state of *P. wortmannii* (Benjamin 1955). As such, *T. wortmannii* (Klöcker) C.R. Benj. (≡ *Penicillium wortmannii* Klöcker, ≡ *Penicillium kloeckeri* Pitt, = *Talaromyces sublevisporus* (Yaguchi & Udagawa) Samson, Yilmaz & Frisvad ≡ *Talaromyces wortmannii* var. *sublevisporus* Yaguchi & Udagawa, = *Talaromyces variabilis* (Sopp) Samson et al. ≡ *Penicillium variabile* Sopp = *Penicillium concavorugulosum* S. Abe (nom. inval., Art. 36)) is considered the correct name for this clade.

Pitt (1980) considered *P. rugulosum* var. *atricolum*, *P. scorteum*, *P. concavorugulosum* and *P. phialosporum* to be synonyms of *T. rugulosus*. However, Peterson & Jurjević (2013) showed that *P. scorteum* and *T. phialosporus* are the same species, with *P. scorteum* an older name, and hence the name *T. scorteus* was introduced. Peterson & Jurjević (2013) showed that *P. rugulosum* var. *atricolum* is not a synonym of *T. rugulosus* and introduced the new combination *T. atricola*. Pitt (1980) also synonymised *P. echinosporum* (CBS 344.51^T), *P. elongatum* (CBS 378.48^T), *P. tardum* (NRRL 1073^T) and *P. chrysitis* (NRRL 1053^T) with *T. rugulosus* and our phylogenetic results confirm their synonymy with *T. rugulosus* (Fig. 2). Described species and associated taxonomic conclusions of different authors are summarised in Table 3. Two of the new species, *T. infraolivaceus* and *T. acaricola*, are consistently resolved in distinct clades correlating with morphological characters discussed in the taxonomy section.

Talaromyces columbinus was described by Peterson & Jurjević (2013). They isolated their strains from air samples and corn grits from the USA. One of our strains was isolated from chicken feed from Nairobi, Kenya. Our results confirm Peterson & Jurjević's (2013) findings that the isolate IMI 392509, isolated by Santos et al. (2006) from a human and identified as *T. piceus*, is in fact *T. columbinus*. Also, Peterson & Jurjević (2013) considered CBS 102383, which was isolated from a case of fungemia and previously identified as *T. piceus*, as an isolate of *T. columbinus*. Both *T. piceus* and *T. columbinus* are able to grow at 40 °C and have vesiculate stipes. However, *T. columbinus* grows faster than *T. piceus* at 37 and 40 °C. In addition, colonies of *T. columbinus* have dark brown reverses and soluble pigments on YES, whereas *T. piceus* has orange to brownish orange reverses and lacks soluble pigments on YES.

Peterson & Jurjević (2013) mentioned problems with the amplification of *BenA* paralogues when using primer pairs Bt2a & Bt2b or BT2f & T22. In our study, a similar result was observed, with gel-electrophoresis revealing one band with primers Bt2a & Bt2b, but subsequent sequences with mixed electropherograms. As a result, we recommend primer set T10 & Bt2b (Glass & Donaldson 1995), at annealing temperatures of 50 or 52 °C, for the amplification and sequencing of *BenA* in this group of species.

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REFERENCES

Abe S. 1956. Studies on the classification of the Penicillia. Journal of General and Applied Microbiology Tokyo 2: 1–344.

Antipova TV, Zhelifonova VP, Kochkina GA, et al. 2008. Growth and biosynthesis of rugulovasines in Penicillium variabile Sopp 1912. Microbiology 77: 446–450.

Bainier G. 1907. Mycothèque de l'École de Pharmacie, IX–XI. Bulletin de la Société Mycologique de France 23: 9–27.

Bara R, Zerfass I, Aly AH, et al. 2013. Atropisomeric dihydroanthracenones as inhibitors of multiresistant Staphylococcus aureus. Journal of Medical Chemistry 56: 3257–3272.

Benjamin CR. 1955. Ascocarps of Aspergillus and Penicillium. Mycologia 47: 669–687.

Biourge P. 1923. Les moisissures de groupe Penicillium Link. Cellule 33: 7–331.

Bouhet J-C, Van Chuong PP, Toma F, et al. 1976. Isolation and characterization of luteoskyrin and rugulosin, two hepatotoxic anthraquinonoids from Penicillium islandicum Sopp and Penicillium rugulosum Thom. Journal of Agricultural and Food Chemistry 24: 964–972.

Breen J, Dacre JC, Raistrick H, et al. 1955. Studies in biochemistry of microorganisms 95. Rugulosin, a crystalline colouring matter of Penicillium rugulosum Thom. Biochemical Journal 60: 618–626.

Cole RJ, Cox RH. 1981. Handbook of toxic fungal metabolites. Academic Press, New York.

Frisvad JC, Filtenborg O, Samson RA, et al. 1990. Chemotaxonomy of the genus Talaromyces. Antonie van Leeuwenhoek 57: 179–189.

Frisvad JC, Samson RA. 2004. Polyphasic taxonomy of Penicillium subgenus Penicillium. A guide to identification of food and air-borne terverticillate Penicillia and their mycotoxins. Studies in Mycology 49: 1–174.

Frisvad JC, Thrane U. 1987. Standardized high-performance liquid chromatography of 182 mycotoxins and other fungal metabolites based on alkylphenone retention indices and UV-VIS spectra (diode-array detection). Journal of Chromatography 404: 195–214.

Glass NL, Donaldson GC. 1995. Development of primer sets designed for use with the PCR to amplify conserved genes from filamentous Ascomycetes. Applied and Environmental Microbiology 61: 1323–1330.

Gouy M, Guindon S, Gascuel O. 2010. SeaView Version 4: A multiplatform graphical user interface for sequence alignment and phylogenetic tree building. Molecular Biology and Evolution 27: 221–224.

Hocking AD, Whitelaw M, Harden TJ. 1998. Penicillium radicum sp. nov. from rhizosphere of Australian wheat. Mycological Research 102: 801–806.

Horré R, Gilges S, Breig P, et al. 2001. Case report. Fungaemia due to Penicillium piceum, a member of the Penicillium marneffei complex. Mycoses 44: 502–504.

Houbraken J, Samson RA. 2011. Phylogeny of Penicillium and the segregation of Trichocomaceae into three families. Studies in Mycology 70: 1–51.

Houbraken J, Spierenburg H, Frisvad JC. 2012. Rasamsonia, a new genus comprising thermotolerant and thermophilic Talaromyces and Geosmithia species. Antonie van Leeuwenhoek 101: 403–421.

Huelsenbeck JP, Ronquist F. 2001. MrBayes: Bayesian inference of phylogenetic trees. Bioinformatics Applications Note 17: 754–755.

Kawai K, Kato T, Mori H, et al. 1984. A comparative study on cytotoxicities and biochemical properties of anthraquinone mycotoxins emodin and skyrin from Penicillium islandicum Sopp. Toxicology Letters 20: 155–160.

Kenkyusho K. 1983. Antitumor agents – comprising pyrano compound obtained by culturing a Penicillium islandicum Sopp. JP 5804392-A and JP 85026372-B. Patent. Derwent Primary Accession nr. 1983-38413K.

Klöcker A. 1903. Sur la classification du genre Penicillium et description d'une espèce nouvelle formant des asques. Comptes Rendus des Travaux du Laboratoire Carlsberg: serie Physiologique 6: 92–102.

Kornerup A, Wanscher JH. 1967. Methuen handbook of colour. 2nd edn. Sankt Jørgen Tryk, Copenhagen, Denmark.

LoBuglio KF, Pitt JI, Taylor JW. 1993. Phylogenetic analysis of two ribosomal DNA regions indicates multiple independent losses of a sexual Talaromyces state among asexual Penicillium species in subgenus Biverticillium. Mycologia 85: 592–604.

Manoch L, Dethoup T, Yilmaz N, et al. 2013. Two new Talaromyces species from soil in Thailand. Mycoscience 54: 335–342.

McNeil J, BArrie FF, Buck WR, et al. (eds). 2012. International Code of Nomenclature for algae, fungi and plants (Melbourne Code). Koeltz Scientific Books, Königstein. [Regnum vegetabile no. 154].

Mehrotra BS, Kumar DA. 1962. A new species of Penicillium from India. Canadian Journal of Botany 40: 1399–1400.

Mori S, Sugihara Y, Kitagawa A, et al. 1996. The respiration-impairing effect of rubroskyrin, a toxic metabolite of Penicillium islandicum, on isolated mitochondria. Mycotoxin Research 12: 91–98.

Narikawa T, Shinoyama H, Fujii T. 2000. A beta-rutinosidase from Penicillium rugulosum IFO 7242 that is a peculiar flavonoid glycosidase. Bioscience, Biotechnology and Biochemistry 64: 1317–1319.
Nehira T. 1933. On the genus Penicillium in Japan. Journal of Fermentation Technology Osaka 11: 849–866.
Nielsen KF, Månsson M, Rank C, et al. 2011. Dereplication of microbial natural products by LC-DAD-TOFMS. Journal of Natural Products 74: 2338–2348.
Nylander AJJ, Ronquist F, Huelsenbeck JP, et al. 2004. Bayesian phylogenetic analysis of combined data. Systematic Biology 53: 47–67.
Oh JY, Kim EN, Ryoo MI, et al. 2008. Morphological and molecular identification of Penicillium islandicum isolate KU101 from stored rice. Plant Pathology Journal 24: 469–473.
Peterson SW, Jurjević Z. 2013. Talaromyces columbinus sp. nov., and genealogical concordance analysis in Talaromyces clade 2a. PLoS ONE 8: e78084.
Pitt JI. 1980. The genus Penicillium and its teleomorphic states Eupenicillium and Talaromyces. Academic Press Inc., London, England.
Pitt JI, Hocking AD. 2009. Fungi and food spoilage, 3rd ed. Springer, Dordrecht / Heidelberg.
Pretsch A, Nagl M, Schwendinger K, et al. 2014. Antimicrobial and anti-inflammatory activities of endophytic fungi Talaromyces wortmannii extracts against acne-inducing bacteria. PLoS ONE 9: e97929.
Raper KB, Fennell DI. 1948. New species of Penicillium. Mycologia 40: 507–546.
Raper KB, Thom C. 1949. Manual of the Penicillia. Williams & Wilkins, Baltimore, USA.
Reenen-Hoekstra ES van, Frisvad JC, Samson RA, et al. 1990. The Penicillium funiculosum complex – well defined species and problematic taxa. In: Samson RA, Pitt JI (eds), Modern concepts in Penicillum and Aspergillus classification: 173–191. Plenum, New York.
Reyes I, Bernier L, Simard RR, et al. 1999. Characteristics of phosphate solubilization by an isolate of a tropical Penicillium rugulosum and two UV-induced mutants. FEMS Microbiology Ecology 28: 291–295.
Saito M, Enomoto M, Tatsuno T. 1971. Yellowed rice toxins. Luteoskyrin and related compounds, chlorine-containing compounds, and citrinin. In: Ciegler A, Kadis S, Ajl SJ (ed), Microbial toxins: 299–308. Academic Press Inc., New York.
Sakai A, Tanaka H, Konishi Y, et al. 2005. Mycological examination of domestic unpolished rice and mycotoxin production by isolated Penicillium islandicum. Journal of the Food Hygienic Society of Japan 46: 205–212.
Samson RA, Yilmaz N, Houbraken J, et al. 2011. Phylogeny and nomenclature of the genus Talaromyces and taxa accommodated in Penicillium subgenus Biverticillium. Studies in Mycology 70: 159–183.
Santos PE, Piontelli E, Shea YR, et al. 2006. Penicillium piceum infection: diagnosis and successful treatment in chronic granulomatous disease. Medical Mycology 44: 749–753.
Sopp OJ. 1912. Monographie der Pilzgruppe Penicillium mit besonderer Berücksichtigung der in Norwegen gefundenen Arten. Skrifter udgivne af Videnskabs-Selskabet i Christiania. Mathematisk-Naturvidenskabelig Klasse 11: 1–208.
Stark AA, Townsend JM, Wogan GN, et al. 1978. Mutagenicity and antibacterial activity of mycotoxins produced by Penicillium islandicum Sopp and Penicillium rugulosum. Journal of Environmental Pathology and Toxicology 2: 313–324.
Swietliczkowa I, Szusterowska-Martinowa E, Braciak W. 1984. Clinical evaluation of 1% cloteimazol ointment in the treatment of corneal mycoses. Klinika Oczna 86: 221–223.
Takedo Y, Suematsu S. Nakazawa R. 1934. The moulds on military instruments II. Journal of the Agricultural Chemical Society of Japan 10: 95–121.
Tamura K, Peterson D, Peterson N, et al. 2011. MEGA5: Molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molecular Biology and Evolution 28: 2731–2739.
Taylor JW, Jacobson DJ, Kroken S, et al. 2000. Phylogenetic species recognition and species concepts in fungi. Fungal Genetics and Biology 31: 21–32.
Thom C. 1910. Cultural studies of species of Penicillium. The Bureau of Animal Industry, US Department of Agriculture, Washington, Government Printing Office.
Thom C. 1930. The Penicillia. Baltimore, The Williams & Wilkins Company.
Udagawa S. 1959. Taxonomic studies of fungi on stored rice grains. III. Penicillium group (penicillia and related genera) 2. Journal of Agricultural Science Tokyo Nogyo Daigaku 5: 5–21.
Udagawa S. 1993. Three new species of Talaromyces from Nepal. Mycotaxon 48: 141–156.
Ueno Y, Ishikawa I. 1969. Production of luteoskyrin, a hepatotoxic pigment, by Penicillium islandicum Sopp. Applied Microbiology 18: 406–409.
Ueno Y, Sato N, Ito T, et al. 1980. Chronic toxicity and hepatocarcinogenicity of (+) rugulosin, an anthraquinoid mycotoxin from Penicillium species: preliminary surveys in mice. The Journal of Toxicological Sciences 5: 295–302.
Uraguchi K. 1962. Malignant hepatoma and so-called carcinogens, with special reference to the toxicity of luteoskyrin in a small dose. Folia Pharmacologica Japonica 58: 19.
Uraguchi K, Miyake M, Shikata T, et al. 1961. Isolation of 2 toxic agents, luteoskyrin and chlorione-containing peptide, from metabolites of Penicillium islandicum Sopp, with properties thereof. Japanese Journal of Experimental Medicine 31: 19–46.
Uraguchi K, Saito M, Noguchi Y, et al. 1972. Chronic toxicity and carcinogenicity in mice of the purified mycotoxins, luteoskyrin and cyclochlorotine. Food and Cosmetics Toxicology 10: 193–207.
Visagie CM, Hirooka Y, Tanney JB, et al. 2014a. Aspergillus, Penicillium and Talaromyces isolated from house dust samples collected around the world. Studies in Mycology 78: 63–139.
Visagie CM, Houbraken J, Frisvad JC, et al. 2014b. Identification and nomenclature of the genus Penicillium. Studies in Mycology 78: 343–371.
Visagie CM, Houbraken J, Rodriques C, et al. 2013. Five new Penicillium species in section Sclerotiora: a tribute to the Dutch Royal family. Persoonia 31: 42–62.
Visagie CM, Jacobs K. 2012. Three new additions to the genus Talaromyces isolated from Atlantis sandveld fynbos soils. Persoonia 28: 14–24.
Visagie CM, Roets F, Jacobs K. 2009. A new species of Penicillium, P. ramulosum sp. nov., from the natural environment. Mycologia 101: 888–895.
Visagie CM, Seifert KA, Houbraken J, et al. 2014c. Diversity of Penicillium section Citrina within the fynbos biome of South Africa, including a new species from a Protea repens infructescence. Mycologia 106: 537–552.
Vos JP de, Garderen E van, Hensen H, et al. 2009. Disseminated Penicillium radicum infection in a dog, clinically resembling multicentric malignant lymphoma. Vlaams Diergeneeskundig Tijdschrift 78: 183–188.
Yaguchi T, Someya A, Miyadoh S, et al. 1994. A new variety of Talaromyces wortmannii and some observation on Talaromyces assiutensis. Mycoscience 35: 63–68.
Yamazaki H, Koyama N, Omura S, et al. 2010a. New rugulosins, Anti-MRSA antibiotics, produced by Penicillium radicum FKI-3765-2. Organic Letters 12 : 1572–1575.
Yamazaki H, Omura S, Tomoda H. 2010b. Xanthoradone C, a new potentiator of imipenem activity against methicillin-resistant Staphylococcus aureus, produced by Penicillium radicum FKI-3765-2. Journal of Antibiotics 63: 329–330.
Yamazaki H, Omura S, Tomoda H. 2010c. 6'-Hydroxy-3'-methoxy-mitorubrin, a new potentiator of antifungal miconazole activity, produced by Penicillium radicum FKI-3765-2. Chemical and Pharmaceutical Bulletin 58: 829–832.
Yilmaz N, Visagie CM, Houbraken J, et al. 2014. Polyphasic taxonomy of the genus Talaromyces. Studies in Mycology 78: 175–341.
Zhou ND, Gu XL, Tian YP. 2013. Isolation and characterization of urethanase from Penicillium variabile and its application to reduce ethyl carbamate contamination in Chinese rice wine. Applied Biochemistry and Biotechnology 170: 718–728.