

# Cortinarius sordidemaculatus and two new related species, *C. anisatus* and *C. neofurvolaesus*, in Fennoscandia (Basidiomycota, Agaricales)

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Two new species growing in the coniferous forests of Fennoscandia and related to *Cortinarius sordidemaculatus* Rob. Henry are described based on morphological and DNA data: *C. neofurvolaesus* Kytöv., Niskanen, Liimatainen & H. Lindstr., spec. nova and *C. anisatus* H. Lindstr., Kytöv. & Niskanen, spec. nova. *C. furvolaesus* H. Lindstr. is synonymized with *C. sordidemaculatus*. The distribution of each species in Fennoscandia is mapped, and their taxonomy, ecology, and relationships are discussed. These three species are preliminary placed in the section *Sordescentes* Melot.

Key words: *Cortinarius*, *Telamonia*, *Sordescentes*, Fennoscandia, taxonomy, ITS, DNA, POY

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## Introduction

In the coniferous forests of Fennoscandia there are several dark brown, middle- to big-sized species of *Cortinarius* subgenus *Telamonia*, which darken with age (and thumbing) and become sordid brownish but not blackish when dried. Most of them are not commonly known. Some of the species have been described (e.g. Henry 1961, Moser 1967, Henry 1981), but a good overview of this group is still lacking.

Usually these darkening species have fairly big spores (larger than  $9.0 \times 5.5 \mu\text{m}$ , unpublished), but small-spored *Cortinarius sordidemaculatus* Rob. Henry (Henry 1981) and *C. furvolaesus* H. Lindstr. (Brandrud et al. 1998) have also been described. In the present paper two new, closely

related, small-spored species are described, and their systematic position studied using the ITS regions of their rDNA.

The use of ITS regions has been successful in *Cortinarius* taxonomy at a higher level (e.g. Høiland & Holst-Jensen 2000, Garnica et al. 2003, Peintner et al. 2004), but there are only a few studies at the species level (e.g. Moser & Peintner 2002a, 2002b). In these studies, only a few collections per species were used and the type material was seldom sequenced, leaving many questions unanswered regarding taxonomy and nomenclature in this species rich genus.

Different species concepts have been used or discussed in the taxonomy of Agaricales (e.g.

Kuyper 1988, Brandrud et al. 1989–1998, Atlas des Cortinaires 1989–2004, Taylor et al. 2000). Our principals of limiting the species are discussed below.

In this study we use names which we can relate to type material or published photo collections. Also, we have tried to study several specimens from different habitats and geographical areas of Fennoscandia to get an idea of the intraspecific variation of the species.

## Material and methods

A total of about 400 specimens were studied, and 150 were identified to be the species studied here. Mostly the collections were gathered by the authors from different parts of Fennoscandia, some however were from Estonia as well as Central and South Europe (deposited in H, S, TUR, UPS and OULU), additionally the whole potential material of H and parts from TUR and OULU were included. The types of *Cortinarius sordidemaculatus* Rob. Henry 1981 (Henry no 1122, PC 0088382), *C. phaeosmus* Rob. Henry 1981 (Henry no. 80828, PC 0088381) and *C. furvolaesus* H. Lindstr. 1998 (CFP 517, S) were studied. The acronyms follow those used by Holmgren et al. (1990).

The interpretation of the names used in differential diagnosis follows *Cortinarius Flora Photographica* (Brandrud et al. 1989–98, photo collections studied). Also the type material of *C. brunneogriseus* Soop (F14331, S) was studied. Suggestive size of the spores is provided for these species based on our own measurements.

Macroscopic characteristics were observed from fresh fruitbodies and several collections were used for the descriptions. Some representative collections were also photographed in fresh condition (marked with \* in the lists of specimens examined). Colour codes were not used, but instead photographs are provided.

Microscopic characteristics were examined with a light microscope (Leitz Labourlux 12 and Leica DM/LS) at magnifications of about 625 and 1560. Spores were drawn with a drawing tube at magnifications of 2000 (Leitz) and 3000 (Fig. 5, Leica). Spores were examined from the surface view of pieces of gills of dried basidiomes, and measured (with an ocular micrometer) from the veil and gill, mounted in Melzer's reagent. Only mature spores in a perpendicularly lateral position (see Fig. 5) were measured. Young, anomalous, very small or gigantic spores were excluded, especially when measured from the gill, thus making the measurements from the gill and veil comparable.

Twenty spores per collection were measured from the cortina or from the top of the stipe of one fruitbody (8 to 15 collections per species). Additionally, 5 to 15 spores were measured from the gill of most of the collections. Scatter diagrams were drawn based on these measurements. The diagrams were used, after excluding approximately 5% of the extreme measurements, to get the size of the spores for each species (descriptions of the species and Fig. 6). Length and width were measured

from the same spore, and the length/width ratios (Q-value) were calculated for individual spores (then as above). For the mean values ( $\bar{X}$  and  $\bar{XQ}$ ), only collections with at least 15 measurements were included.

The hyphae of the gill trama were examined from the pieces of gills mounted in Melzer's reagent as well as basidia, which were measured from a few collections from the same pieces. Additionally, sterile cells, found at the edge of the gill, were measured from a few collections; however, the frequency was very variable and thus are not discussed more. Pileipellis was examined from small scalps or sections of dried basidiomes (central part of the cap) made with a razor blade, mounted in Melzer's reagent or from water-mounted fresh material. Some drawings were made of these sections, but due to the difficulty of presenting differences between species only one was chosen to represent the groups' common pileipellis structure. For the measurements of the pileipellis approximately 5 to 10 collections/ species were studied (sections). Additionally, the measurements of the epicutis hyphae as well as more observations were made from larger material (scalps). Section making is time consuming, and for large material a more effective method – scalps – is needed. In our opinion, this method is useful for studying the epicutis and hypoderm in large material and the incrustations are easier to observe from the scalps than sections.

Several collections of the studied species (n=27) from different geographical areas (Table 1, marked with \* in the lists of specimens examined) and type material of *Cortinarius sordidemaculatus* and *C. furvolaesus* were sequenced. Also *C. cf. bovinus* (plate no. III *Cortinarius* 65 upper in Moser & Jülich 1990, IB 86/172), one collection of *C. brunneus* (Pers. : Fr.) Fr. var. *brunneus* (plate B07 in Brandrud et al. 1992, CFP587, S, neotype), *C. armillatus* (Fr. : Fr.) Fr. (plate B09 in Brandrud et al. 1992, CFP584, S), *C. hinnuleus* Fr. (plate A19 in Brandrud et al. 1989, CFP332, S) and *C. armeniacus* (Schaeff. : Fr.) Fr. (plate A46 in Brandrud et al. 1989, CFP809, S) were included to get a preliminary idea of the systematic position of our species inside the subgenus *Telamonia*. *Cortinarius norrlandicus* Brandrud (plate A26 in Brandrud et al. 1989, CFP526, S, isotype) was chosen for the outgroup species, because its sequence is close to the sequence of *C. saginus* Fr. (AF325608), which belongs to the clade *Phlegmacium* in Peintner et al. (2004).

Total DNA was extracted from a few milligrams of dried material (a piece of gill) using the NucleoSpin Plant kit (Macherey-Nagel). The primers ITS 1F and ITS 4B (Gardes & Bruns 1993) were used to amplify the ITS regions of the rDNA.

PCR amplifications were performed in a 25  $\mu$ l reaction mix with about 70 ng of extracted DNA, 1.0 U Taq DNA polymerase and 1X buffer (Promega, USA), 3 mM MgCl<sub>2</sub>, 200  $\mu$ M of each dNTP and 0.4  $\mu$ M of each primer. PCR reactions were run on a PTC-200 Thermal Cycler (MJ Research, Inc.) with the following settings: denaturation for 5 min at 95°C, followed by 35 cycles of: 94°C denaturation for 30 s, annealing for 30 s at 50°C and extension at 72°C for 2 min. The PCR products were purified using a GFX PCR DNA and Gel Band Purification kit (Amersham Biosciences).

Table 1. Specimens used in the DNA study. CFP= Cortinarius Flora Photographica, BM= E. Bendiksen & K. Metsänheimo, IK= I. Kytövuori, TEB= T. E. Brandrud, TN= T. Niskanen. For acronyms of biological provinces see e.g. Hansen & Knudsen 1992: Nordic Macromycetes 2: 24, 25.

Species	Voucher	Herb.	Locality	Sequence no.	GenBank accession number
<i>C. sordidemaculatus</i> Rob. Henry (holotype)	PC0088382	PC	France, Haut-Doubs	783-05	DQ139984
<i>C. sordidemaculatus</i>	TEB104-03	H	Norway, Hord, Ulvik	889-05	DQ139990
<i>C. furcolaesus</i> H. Lindstr. (holotype)	CFP517	S	Sweden, Upl, Uppsala-Näs	484-04	DQ139986
<i>C. sordidemaculatus</i>	TN03-597	H	Sweden, Mpd, Haverö	293-03	DQ139989
<i>C. sordidemaculatus</i>	CFP611	S	Sweden, Jmt, Hällesjö	505-04	DQ139987
<i>C. sordidemaculatus</i>	IK04-003	H	Finland, U, Kirkkonummi	648-04	DQ139991
<i>C. sordidemaculatus</i>	IK98-1758	H	Finland, ES, Kerimäki	408-04	DQ139985
<i>C. sordidemaculatus</i>	IK98-1240	H	Finland, PeP, Tornio	380-04	DQ139988
<i>C. neofurvolaesus</i> Kytöv., Niskanen, Liimatainen, H. (Lindstr. (holotype))	CFP1438	S	Sweden, Hrj, Hede	592-04	DQ139999
<i>C. neofurvolaesus</i>	IK04-002	H	Finland, U, Espoo	613-04	DQ140001
<i>C. neofurvolaesus</i>	IK04-001	H	Finland, U, Helsinki	614-04	DQ139997
<i>C. neofurvolaesus</i>	IK04-005	H	Finland, U, Nurmijärvi	612-04	DQ140000
<i>C. neofurvolaesus</i>	IK01-010	H	Finland, U, Tammisaari	172-03	DQ140002
<i>C. neofurvolaesus</i>	IK94-570	H	Finland, EK, Anjalankoski	619-04	DQ140003
<i>C. neofurvolaesus</i>	IK95-1588	H	Finland, EH, Orivesi	618-04	DQ139998
<i>C. neofurvolaesus</i>	IK02-002	H	Finland, PH, Konnevesi	291-03	DQ139996
<i>C. neofurvolaesus</i>	TN02-960	H	Finland, Ks, Kuusamo	174-03	DQ139995
<i>C. neofurvolaesus</i>	TN02-766	H	Finland, Ks, Kuusamo	147-03	DQ139992
<i>C. neofurvolaesus</i>	TN02-769	H	Finland, Ks, Kuusamo	148-03	DQ139993
<i>C. neofurvolaesus</i>	TN02-795	H	Finland, Ks, Kuusamo	173-03	DQ139994
<i>C. anisatus</i> H. Lindstr., Kytöv., Niskanen (holotype)	CFP1200	S	Sweden, Äng, Säbro	611-04	DQ117931
<i>C. anisatus</i>	BM18.9.1987	O	Sweden, Sm, Femsjö	709-05	DQ120758
<i>C. anisatus</i>	IK00-005	H	Sweden, Dlr, Idre	288-03	DQ120756
<i>C. anisatus</i>	TN03-372	H	Sweden, Hls, Bergsjö	430-04	DQ117929
<i>C. anisatus</i>	TN03-596	H	Sweden, Mpd, Haverö	428-04	DQ117930
<i>C. anisatus</i>	TN03-625	H	Sweden, Mpd, Haverö	292-03	DQ120753
<i>C. anisatus</i>	TN04-152	H	Finland, EH, Ruovesi	615-04	DQ120757
<i>C. anisatus</i>	TN04-550	H	Finland, PeP, Tornio	710-05	DQ120754
<i>C. anisatus</i>	TN04-640	H	Finland, PeP, Tornio	649-04	DQ120755
<i>C. armeniicus</i> (Shaef. : Fr.) Fr.	CFP809	S	Sweden, Äng, Häggdänger	590-04	DQ117925
<i>C. armillatus</i> (Fr. : Fr.) Fr.	CFP584	S	Sweden, Äng, Säbrå	542-04	DQ114744
<i>C. cf. bovinus</i> Fr.	IB86/172	IB	Austria, Tirol	706-05	DQ139983
<i>C. brunneus</i> (Pers. : Fr.) Fr. var <i>brunneus</i> (neotype)	CFP587	S	Sweden, Äng, Säbrå	557-04	DQ117927
<i>C. himmuleus</i> Fr.	CFP332	S	Sweden, Mpd, Torp	561-04	DQ117926
<i>C. norrlandicus</i> Brandrud (isotype)	S-F14270	S	Sweden, Äng, Häggdänger	237-03	DQ117928

Sequencing was performed on both strands using a BigDye Terminator v1.1 Sequencing kit (Applied Biosystems). Reactions were performed in 8 µl with 1 µl of PCR-product, 1.6 µM of primer (ITS 1F or ITS 4, White et al. 1990, Gardes & Bruns 1993) and 2.2 µl Terminator Ready Reaction Mix. Reactions were run for 96°C for 1 min, followed by 30 cycles with settings: 96°C for 30 s, 50°C for 15 s, 60°C for 4 min. Unincorporated dye

terminators and primers were removed by Sephadex G-50 DNA Grade Fine (Amersham Biosciences) purification and the reactions were analyzed by a MegaBace (Amersham Biosciences) automatic sequencer.

To get a rough idea of the closeness of our species, the sequences were aligned using the ClustalW 1.8 program (Thompson et al. 1994) on the European Bioinformatics Institute server (<http://www.ebi.ac.uk/clustalw/>)

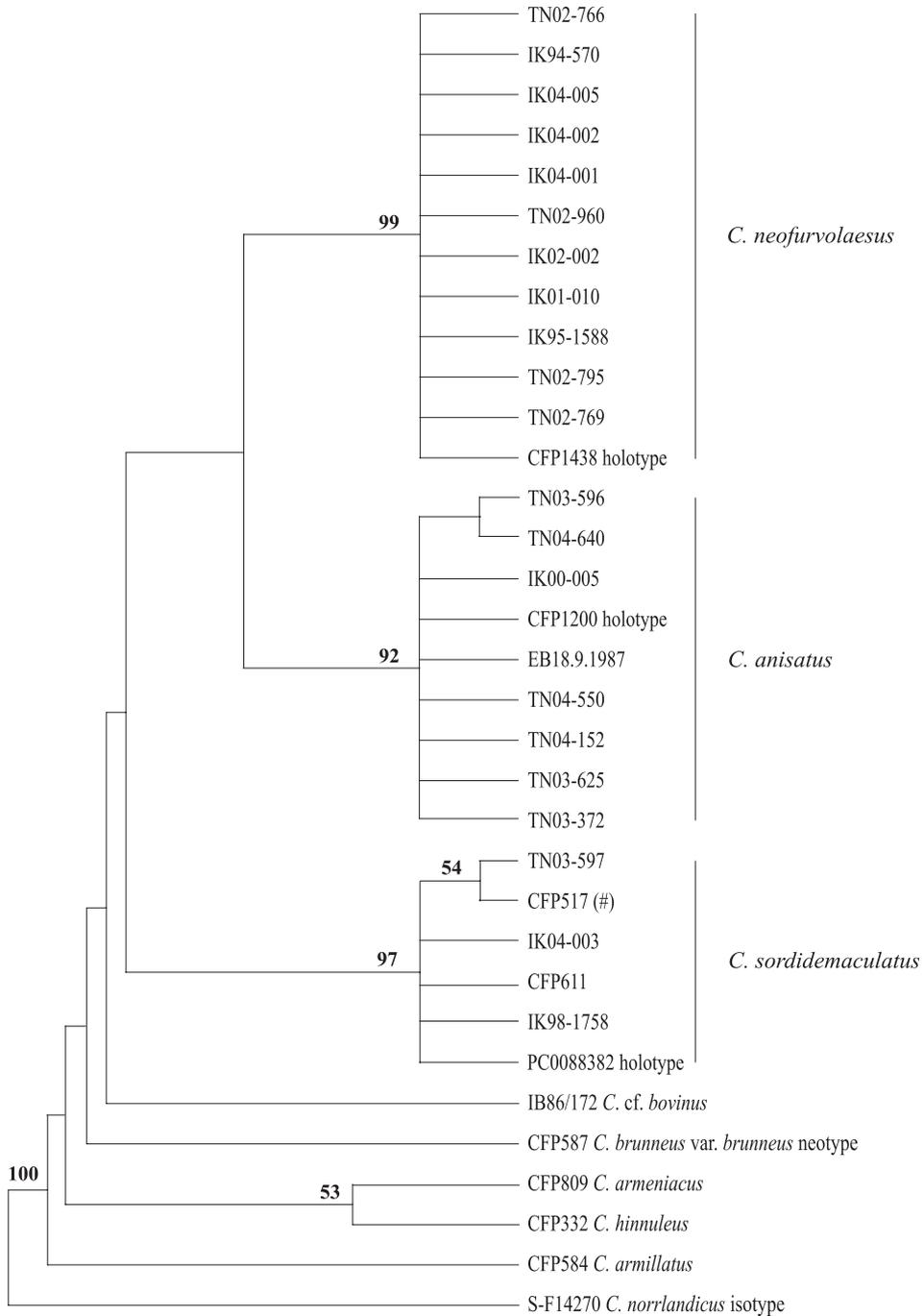


Fig. 1. POY consensus tree of eight shortest trees with a length of 361 steps for ITS sequences. Jackknife values higher than 50% are shown above the respective branches. (#) = *C. furvolaeus* CFP517 holotype.

index.html). Differences between species were counted from alignments, which of closely related species are usually fairly unambiguous, thus counting the differences between species is easy. Even though the method has limitations presenting differences between the taxa or limiting the species, it gives a rough idea of how closely related the species are.

Alignments are seldom objective or repeatable when DNA sequences compared show length variation. The normal procedure in these situations is that these ambiguous parts of the alignment are not included in the analysis. To avoid these problems we used optimization alignment as implemented in the program POY 2.7 (Wheeler 1996, Gladstein & Wheeler 2001). The key feature of this approach is that alignment and search for the optimal solution are done simultaneously. We used the computers of CSC (Scientific computing, Espoo, Finland). Command line used for the analysis of POY was: `poy -random 150 -gap 1 -maxtrees 1 -holdmaxtrees 80 -fitchtrees -ratchettbr 3 -ratchettrees 1 -treefuse -fusemaxtrees 20 -fuselimit 10 names of the sequence files > name of the results file 2 > name of the program progress file`. The gap cost one was chosen because it does not weight indels (insertion/deletion) compared to base changes. Also gap cost two, which is the default value in the program, was used simply in order to see how this affects the results. Two specimens of *C. sordidemaculatus* (Kytövuori 98-1240, Brandrud 104-03) were excluded from the analysis for following reasons: the quality of the sequence at the end of the ITS 2 region was not good (98-1240) and the sequence was obtained only after the analyses were already performed (104-03).

To quicken the analyses, the sequences were cut into four different regions: ITS 1, 5.8S, start of the ITS 2 and end of the ITS 2 (the sequence cut points are available upon request from the authors).

Jackknife resampling (Farris et al. 1996) analysis was done using the command line: `poy -jackboot -random 1000 -gap 1 -maxtrees 1 -hold maxtrees 100 -fitchtrees`. Analysis was done also with gap costs of two. Jackknife was chosen, because unlike bootstrap, it does not create a matrix of characters that does not exist.

## Results of DNA studies

The analysis of the ITS sequences with the gap cost one resulted in eight most parsimonious trees with the length of 361 steps. With 150 replicates, the shortest trees were found seven times and the analysis took approximately 13 hours. The use of ratchet or treefusing did not result in a shorter tree than SPR and TBR, but treefusing resulted in three new trees with the length of 361 steps. From the eight parsimonious trees a consensus tree was produced (Fig. 1).

*Cortinarius anisatus*, *C. sordidemaculatus* and *C. neofurvolaeus* form a clade with jackknife value < 50%. *C. neofurvolaeus* is the nearest relative of *Cortinarius anisatus* (also jackknife value < 50%). Their interspecific difference is 21/30 bases (gaps treated as one change/ every gap treated as individual change). Their nearest relative in this study is *C. sordidemaculatus* differing 22/24 bases from *C. anisatus* and 24/33 from *C. neofurvolaeus*.

The gap cost two resulted in one most parsimonious tree with the length of 567 steps (tree not shown). With 150 replicates, the shortest tree was found 11 times and the analysis took approximately 16 hours. The use of ratchet or treefusing did not result in a shorter tree than SPR and TBR.

The default gap cost two in the POY analyses showed that the relationships between the studied species might be different, but again the support values are low (< 50%). *C. sordidemaculatus* and *C. neofurvolaeus* are the nearest species and their nearest relative is *C. anisatus*. In both analyses *C. cf. bovinus* is the nearest relative of these three species (jackknife value < 50%).

*Cortinarius sordidemaculatus*, *C. neofurvolaeus* and *C. anisatus* have low intraspecific variation. Based on ITS-sequences and morphological data they seem to be clearly separate species. In *C. anisatus*, the specimens *Niskanen 03-596* and *Niskanen 04-640* differ from the others by one base. These two specimens also have slightly bigger spores and both smelled like radish.

We did not find any match or even close sequences to *C. sordidemaculatus*, *C. neofurvolaeus* or *C. anisatus* from the public gene banks (GenBank: <http://www.ncbi.nlm.nih.gov/> and UNITE: <http://unite.zbi.ee/>). Among the other species used in the tree (Fig. 1) we found almost identical to *C. brunneus* (UNITE no. UDB000158), *C. armillatus* (UNITE no. UDB 000155) and *C. armeniacus* (UNITE no. UDB001010).

In some parts of our ITS-sequences there are ambiguous IUB-codes. These places may be indicative of polymorphisms in ITS-regions within the same individual, but further studies are needed.



Fig. 2. *Cortinarius sordidemaculatus*, Finland, Perä-Pohjanmaa, Tornio, Korkiamaa, 1998 *Kytövuori* 98-1240 (H). Photo I. Kytövuori.



Fig. 3. *Cortinarius neofurvolaesus*, Finland, Etelä-Karjala, Anjalankoski, Kaipiainen, 1994 *Kytövuori* 94-570 (H). Photo I. Kytövuori.

### Key to the species

Fruitbodies brown to dark, medium-sized, strongly darkening when bruised or dried, stipe with clavate-bulbous base, spores rather small, about  $7.5\text{--}9.0 \times 5.0\text{--}6.0 \mu\text{m}$ , hyphae of the gill trama usually smooth or faintly incrusted. Pileipellis with thin epicutis and rather well developed hypoderm (Fig. 9).

1. Fruitbodies with a smell of anise (like *C. odorifer*) ..... 3. *C. anisatus* (Fig. 4)
  - Fruitbodies not with a smell of anise ..... 2
2. Fruitbodies dark red brown, spores ellipsoid to ovoid-ellipsoid, with rounded apex, evenly verrucose, ornamentation fairly weak ..... 2. *C. neofurvolaesus* (Fig. 3)
  - Fruitbodies more sordid brown, spores slightly amygdaliform or ovoid, more strongly and less evenly verrucose ..... 3
3. Fruitbodies fairly stout, spores often amygdaliform, fairly thin-walled, weakly dextrinoid ..... 1. *C. sordidemaculatus* (Fig. 2)
  - Fruitbodies more slender (especially stipe), spores distinctly ovoid, somewhat thick-walled, more strongly dextrinoid ..... 3. *C. anisatus*

#### 1. *Cortinarius sordidemaculatus* Rob.

Henry – Figs. 2, 5–8

*Cortinarius sordidemaculatus* Rob. Henry, Bull. Soc. Mycol. Fr. 97(3): 196. 1981. – Type: France, sapinières du Haut-Doubs, Frasne, leg. R. Henry 1122 (PC0088382).

*Synonym:* *Cortinarius furvolaesus* H. Lindstr., Cortin. Fl. Photogr. 4: 20. 1998. – Type: Sweden, Uppland, Upplands-Näs, Dalkarlskärret, coniferous forest (*Pinus*, *Picea*), 18 Sept 1986 Lindström et al. CFP517 (S).

*Illustrations:* Cortin. Fl. Photogr. D41.



Fig. 4. *Cortinarius anisatus*, Finland, Perä-Pohjanmaa, Tornio, 2004 Niskanen 04-550 (H). Photo K. Liimatainen.

*Pileus* 4–10 cm, convex when young, soon plano-convex with a low and very broad umbo and slightly down curved margin, with age often ring-like depressed around the umbo; surface silky whitish fibrillose when young, later more apparent only in the margin, veil remnants fairly abundant in the margin; greyish brown-umber (to chestnut brown), later darkening-blackening in large spots; hygrophanous, drying up in a zone at the centre and then somewhat radially towards the margin to more yellowish brown. *Lamellae* moderately crowded (40–60 reaching the stipe), (fairly) strongly emarginate, light greyish ochraceous to light yellowish brown when young, brown to dark brown with age, middle thick, broad in later stage, edge somewhat lighter or not, fairly uneven. *Stipe*

(5)8–13 × 0.7–1.5 cm, cylindrical, at the base sub-clavate to somewhat bulbous (–3 cm), fairly coarsely (silky) fibrous, at first white to greyish white, later brown, darkening downwards. *Mycelium* white. *Veil* white to greyish white, fairly abundant in a girdle-like zone, floccose below. *Context* in cap thin towards the margin, brown, in stipe marbled yellowish-greyish brown, darker in the cortex, darkening downwards, whitish grey when dry, blue tints not observed. *Smell* indistinct, reminding a little that of *C. brunneus*. *Exsiccata* dark sordid but not black.

*Spores* 7.7–9.1(9.3) × 5.0–5.6 μm, Q=1.50–1.72 (320 spores, 25 collections (measurements from cortina 9 collections), one fruitbody from each, Figs. 5, 6),  $\bar{X}$ =8.2–9.0 × 5.2–5.6 μm,  $\bar{X}Q$ =1.54–1.60

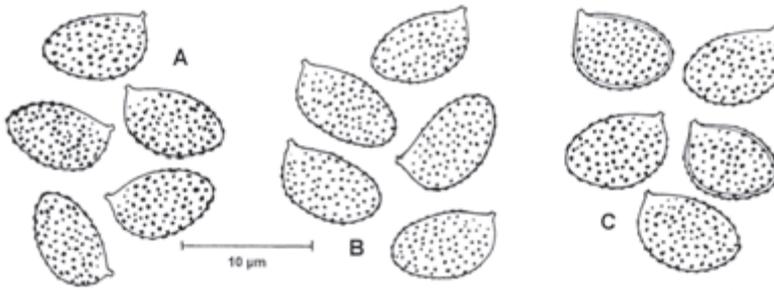


Fig. 5. Spores of A) *Cortinarius sordidemaculatus* (Henry 1122, PC, holotype), B) *C. neofurvolaeus* (Lindström et al. CFP 1438, S, holotype) and C) *C. anisatus* (Lindström et al. CFP 1200, S, holotype), wall thickness indicated in two spores. Drawings T. Niskanen.

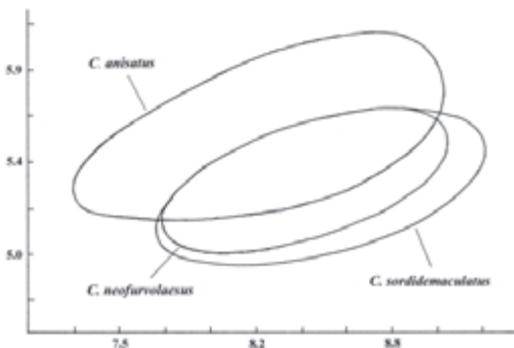


Fig. 6. Diagram showing the spore size of *Cortinarius sordidemaculatus*, *C. neofurvolaeus* and *C. anisatus*. The lines are drawn on the basis of scatter diagrams, and contain 95% of the spore measurements of each species. x axis: length of spores. y axis: width of spores.

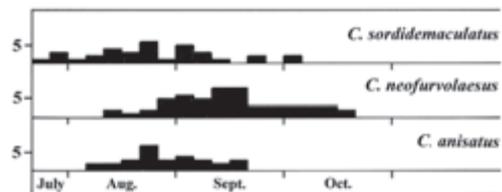


Fig. 7. Fruiting periods of *Cortinarius sordidemaculatus*, *C. neofurvolaeus* and *C. anisatus* in the material examined. x axis: the season from 21.VII. to 24.X. divided in pentads. y axis: number of specimens collected in each pentad.

Fig. 8. Distribution of *Cortinarius sordidemaculatus* in NW Europe according to the material examined.



(11 collections (measurements from cortina 9 collections), at least 15 spores measured from each), (weakly) amygdaliform, narrower at the apex than in *C. neofurvolaeus*, fairly thin-walled, weakly to moderately dextrinoid, moderately verrucose, slightly more strongly verrucose at the apex; spores measured from the gills slightly narrower than those from the cortina at the top of the stipe. *Hyphae of the gill trama* in the overall view pale olivaceous, fairly finely scabrous to scabrous. *Lamella edge* with frequent clavate to bulbous sterile cells ( $10\text{--}50 \times 6\text{--}14 \mu\text{m}$ ). *Basidia* 4-spored,  $22\text{--}33 \times 6\text{--}9 \mu\text{m}$ , with somewhat olivaceous contents. *Pileipellis* with a fairly thin, not or only poorly gelatinised epicutis of fairly short-celled hyphae ( $4\text{--}25 \mu\text{m}$ , wide hyphae characteristic and fairly frequent) with pale amber parietal or stronger intracellular granulose pigment, smooth to finely cross-striated incrustated, often not uniform with gaps exposing the hypoderm (transition to the hypoderm often not distinct); hypoderm present but sometimes fairly poorly distinctive with isodiametric to elongated elements about  $15\text{--}60 \times 5\text{--}40 \mu\text{m}$  with weakly thickened walls with parietal, pale amber thick-wall pigment (especially in the upper part); at the transition to the trama hyphae  $5\text{--}20 \mu\text{m}$  wide with distinctly incrustated amber pig-

ment. *Trama* hyphae more or less hyaline. *Clamp connections* present.

*ITS-regions* (including 5.8S region) 512–514 bases long (total 8 sequences, Table 1). Intraspecific variation occurs in four positions of the ITS-regions. Difference to *C. anisatus* 22/24 bases, to *C. neofurvolaeus* 24/33 bases.

*Ecology and distribution:* *Cortinarius sordidemaculatus* grows in boreal to hemiboreal, sub-mesic spruce forests, especially on rich or calcareous ground, but also occurs in ordinary blueberry spruce forests. It seems to be fairly common but not abundant and grows in loose groups or solitarily, usually in older forests. Fruiting of *C. sordidemaculatus* is observed from late July to early October (Fig. 7). The beginning and optimum fruiting period, in August and early September, is earlier than in *C. neofurvolaeus*. So far, we know *C. sordidemaculatus* from Fennoscandia and France (Fig. 8).

*Differential diagnosis:* *Cortinarius sordidemaculatus* is a brown, middle-sized but quite robust *Telamonina* species growing in spruce forests. It is best known by its greyish-brown to amber, plano-convex cap with low umbo, clavate-bulbous base of the stipe, and brownish context. The spores are typically slightly dextrinoid, quite

small (7.7–9.1 × 5.0–5.6 µm), and somewhat amygdaliform. Blue tints have never been observed in the fruitbodies.

Other darkening *Telamonia* species, such as related *Cortinarius anisatus*, grow in similar habitats. *C. anisatus* can be distinguished from *C. sordidemaculatus* by the characteristic smell of anise, membraneous white veil, more reddish-brown colours and somewhat thick-walled more dextrinoid and slightly broader spores. *C. illuminus* Fr., a very common spruce forest species, is usually more slender than *C. sordidemaculatus* and has a deeper reddish-brown, at the margin pellucid-striate cap, and small round spores (6.0–7.0 × 5.0–6.0 µm). In calcareous spruce forests there are also other more or less robust, dark *Telamonia* species, but these have bigger spores than *C. sordidemaculatus*.

The other closely related new species, *C. neofurvolaeus*, grows in proximity to pine, has a deeper reddish-brown cap and is slightly more slender. The spores are more dextrinoid and more ellipsoid than in *C. sordidemaculatus*.

Type specimens of *C. sordidemaculatus* Rob. Henry (Henry 1981) and *C. furvolaeus* H. Lindstr. (Brandrud et al. 1998) were studied morphologically and using their rDNA ITS-regions. The type material of *C. sordidemaculatus* was scanty and in very poor condition. It was strongly darkened, but it was not possible to make more specific macroscopic observations. The spores are abundant, and their size and shape fit well with *C. furvolaeus*. In the description of *C. sordidemaculatus* the spore size given is 8.7 × 4.3 µm, but our own measurements from the type are 7.7–8.6(9.0) × 5.0–5.7 µm (20 spores from the gill, one fruitbody).

There was a 3 base difference in DNA between the types in the ITS regions. We have identical specimens and also intermediates for both the types. Based on morphology and DNA we consider *C. furvolaeus* to be a later synonym of *C. sordidemaculatus*.

*Cortinarius furvolaeus* was first described as growing amongst pine, but this also actually included the closely related, here newly described *C. neofurvolaeus*.

**Specimens examined:** NORWAY. Hordaland: Ulvik, Brandrud 104-03\* (a part in H). – SWEDEN. Uppland: Uppsala-Näs, Dalkarlskärret, Lindström et al. CFP517\* (holotype of *C. furvolaeus*, S). Västmanland: Skinnskatteberg, Ridrarhyttan, Lindström 00.690

(UPS). Medelpad: Borgsjö, Harrån, Lindström 95.086 (UPS); Lönnån, Lindström 95.221 (UPS). Haverö, Snöberget, Niskanen et al. 03-597\* (H). Jämtland: Hällesjö, Ansjö, Lindström et al. CFP611\* (UPS). Frösö, Fillstabäcken, Lindström 04.023 (UPS). – FINLAND. Varsinais-Suomi: Kisko, Kytövuori et al. 00-007 (H). Vihti, Lintumäki, Kytövuori et al. 01-013 (H); Vesikansa, Kytövuori 04-020 (H). Uusimaa: Espoo, Nuukio National Park, Kytövuori 04-012 (H). Kirkkonummi, Meikoträsket, Kytövuori et al. 04-003\* (H). Vantaa, N of Ilola, Kytövuori 04-009 (H). Etelä-Häme: Ruovesi, Pihlajalahti, Kytövuori 04-011 (H); Siikakangas, Niskanen et al. 04-160 (H). Virrat, Monoskylä, Kytövuori 02-003 (H). Etelä-Savo: Kerimäki, Kytövuori 98-1758\* (H). Mäntyharju, Hietaniemi, Kytövuori 97-1878 (H). Pohjois-Häme: Konnevesi, Pyydyskylä, Kytövuori et al. 02-016 (H); Tökkerönmäki, Kytövuori 04-014 (H). Laukaa, Hitonhauta, Kytövuori 02-011 (H); Äijälä, Kytövuori et al. 02-014 (H), 02-015 (H). Virrat, Hauhuu, Kytövuori 04-006 (H), 04-007 (H); Jäähdyspohja, Kytövuori 04-008\* (H), 04-010 (H). Kainuu: Kuhmo, Elimyssalo, Kytövuori 99-702 (H). Puolanka, Paljakkä, Kytövuori 02-009 (H). Suomussalmi, Kiannanniemi, Kytövuori 02-006 (H), 02-007\* (H), 02-008 (H); Näljänkä, Kytövuori 97-1297 (H). Perä-Pohjanmaa: Rovaniemi rural commune, Louevaara, Niskanen et al. 04-459 (H, OULU). Tervola, Raemäki, Kytövuori 98-1304 (H). Tornio, Korkiamaa, Kytövuori 98-1240\* (H, TUR), Niskanen et al. 04-503 (H). Koillismaa: Kuusamo, Oulanka National Park, Niskanen et al. 02-263 (H). – FRANCE. Sapinières du Haut-Doubs, Frasné, Henry 1122\* (holotype, PC0088382).

## 2. *Cortinarius neofurvolaeus* Kytöv., Niskanen, Liimatainen & H. Lindstr., spec. nova – Figs. 3, 5–7, 9, 10

*Cortinarius furvolaeus* H. Lindstr., Cortin. Fl. Photogr. 4: 20. 1998, p. p. not including the type.

**Illustrations:** The photograph of the type will be published in Cortin. Fl. Photogr. 5.

*Pileus* 3–7 cm, obtuse conicus, tum explanatus, late et obtuse umbonatus, laevis, glaber, margine deflexo, albosericeo; hygrophanus, udo obscure castaneobrunneus, sicco alutaceo-ochraceus, centro obscuro; plerumque radialiter fusciscente-nigrescente maculatus. Lamellae subconfertae, dein latae, pallide griseo-ochraceae. Stipes 5–12 × 0.5–1.2 cm, cylindræus, basi incrassato-bulbosus; fibrillososericeus, albidus, dein infra obscure fusciscentis, velo albedo, in zonan adpresso. Caro fuscovariæata, nunquam caerulea observata, inodora. Exsiccata brunneofusca. Sporae 7.7–9.1 × 5.0–5.7 µm, ovatoellipsoideae, paulo verrucosae. Epicutis incrustata. In silvis coniferis aridis et semiaridis, cum Pino sylvestri.

*Holotypus*: Sweden, Härjedalen, Hede parish, 1 km E of Hede, in a dry pine forest on sand (*Pinus*), 1 Oct 1999 Lindström et al. CFP 1438 (S, isotype H).

*Pileus* 3–7 cm, convex when young, soon plano-convex with a broad umbo, sometimes also low acute, slightly down-curved margin, when old somewhat umbonate-concave and often undulating, margin seldom narrowly pellucid-striate, rarely with weak fibrous veil remnants; surface matt; deeply to dark, saturated red brown, later with darkening to blackening (radial) spots, margin often silky whitish, at least when young; hygrophanous, drying up in a central zone and then somewhat radially towards the margin to a more yellowish brown, umbo remaining reddish brown. *Lamellae* moderately crowded (36–48 reaching the stipe), strongly to weakly emarginate, light (greyish) ochraceous to light yellowish brown when young, with age saturated brown, middle thick, moderately broad, edge not or somewhat lighter coloured, weakly uneven. *Stipe* (4)5–12(13) × 0.5–1.2 cm, cylindrical, at the base weakly to strongly clavate or bulbous (–2.5 cm), finely silky fibrous, at first white to greyish white, later greyish yellow brown, with age downwards dirty grey-brown. *Mycelium* white. *Veil* white, thin, sparse, often forming a distinct but thin band in the middle of the stipe. *Context* in the cap thin towards

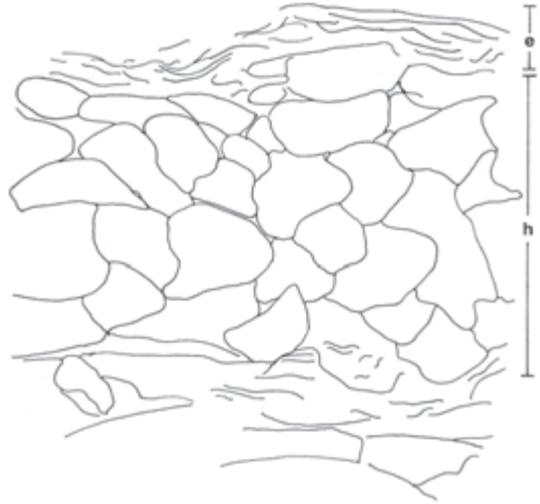


Fig. 9. Pileipellis structure in *C. neofurvolaeus* (Kytövuori 04-025, H). Section from the centre of the pileus of a dried basidiome, mounted in Melzer's reagent. e=epicutis, h=hypoderm. Drawing I. Kytövuori.

the margin, dark reddish brown, in the stipe greyish (yellow) brown to dirty brown, marbled, whitish grey when dry, bluish tints not observed. *Smell* indistinct or slightly raphanoid. *Exsiccata* variably brown, dark, not black.

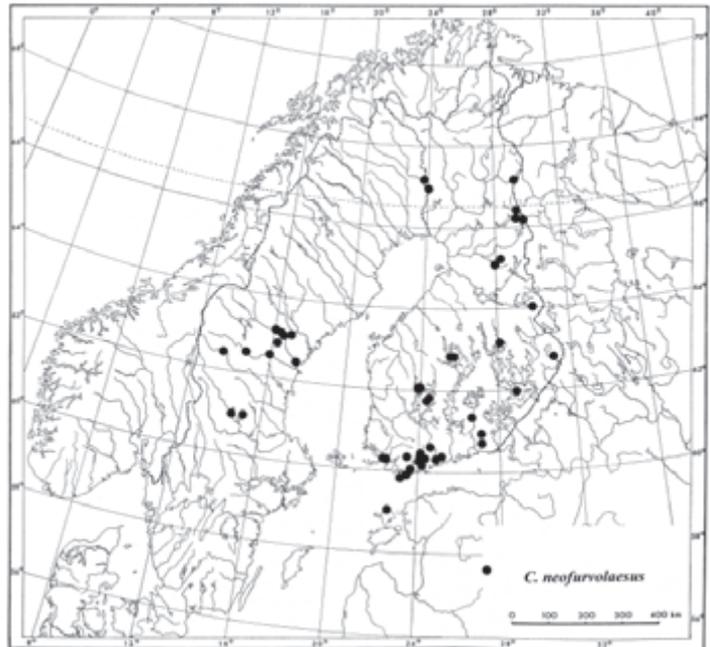


Fig. 10. Distribution of *Cortinarius neofurvolaeus* in NW Europe according to the material examined.

Spores  $7.7\text{--}9.1 \times 5.0\text{--}5.7 \mu\text{m}$ ,  $Q=1.48\text{--}1.72$  (660 spores, 56 collections (measurements from cortina 15 collections), one fruitbody from each, Figs. 5, 6),  $\bar{X}=7.8\text{--}8.5 \times 5.2\text{--}5.5 \mu\text{m}$ ,  $XQ=1.49\text{--}1.58$  (21 collections (measurements from cortina 15 collections), at least 10 spores measured from each), ellipsoid to very weakly amygdaliform with a rounded apex, fairly thin-walled, moderately dextrinoid, fairly finely (sometimes moderately) and evenly verrucose; spores measured from the gills slightly narrower than those from the cortina at the top of the stipe. *Hyphae of the gill trama* smooth to very finely scabrous, in the overall view pale olivaceous yellowish. *Lamella edge* with rather frequent clavate to bulbous ( $9\text{--}23 \times 5\text{--}12 \mu\text{m}$ ) sterile cells. *Basidia* 4-spored,  $25\text{--}32 \times 5\text{--}8 \mu\text{m}$ , with fairly dark olivaceous contents. *Pileipellis* (Fig. 9) with thin epicutis of 2–6 layers, hyphae  $3\text{--}7 \mu\text{m}$  broad (inner broader, about  $10 \mu\text{m}$ ), mostly smooth with greyish brown, amorph intracellular pigment, somewhat cemented by a gelatinous substance; hypoderm distinct, about 10 layers, elements short rectangular to roundish,  $15\text{--}70 \times 10\text{--}20(30) \mu\text{m}$ , with moderately strong, greyish brown thick-wall pigment (stronger than in *C. brunneus*); transition to the trama with longer elements ( $80\text{--}90 \times 8\text{--}15 \mu\text{m}$ ) with more distinct cross-stripped and granular incrustated pigment. *Trama hyphae* loose and irregular with a mixture of narrow ( $4\text{--}5 \mu\text{m}$ ), weakly incrustated and broader ( $20\text{--}30 \mu\text{m}$ ), hyaline elements. *Clamp connections* present.

*ITS-regions* (including 5.8S region) 502 bases long (total 12 sequences, Table 1). In two positions of the ITS regions (not in all sequences) two nucleotides were observed in one position (Y=C/T), otherwise there were no intraspecific variation. Difference to *C. anisatus* 21/30 bases, to *C. sordidemaculatus* 24/33 bases.

*Ecology and distribution:* *Cortinarius neofurvolaeus* is common and often abundant in boreal to hemiboreal sandy pine heath forests, but also occurs in pine forests on rocky ground, and sometimes in spruce forests mixed with pines. *C. neofurvolaeus* often grows in groups, although sometimes it is solitary. It has been found from late August to late October (Fig. 7). The species fruits comparatively late with its optimum period in September and early October, later than *C. sordidemaculatus*. So far, we know *C. neofurvolaeus* from Fennoscandia and Estonia (Fig. 10).

*Differential diagnosis:* *Cortinarius neofurvolaeus* is a common *Telamonia* species in nutrient poor pine forests and especially in lichen rich, sandy pine heath forests in boreal areas. This species is typically dark reddish-brown, with a matt cap often having blackish spots, the stipe is clavate to bulbous, the context is brown (no bluish tints have been observed), and the fruitbodies darken considerably when dried; they, however, are more brownish than those of the other species. The spores are similar to those of *C. sordidemaculatus*, small and evenly verrucose, but they are somewhat more dextrinoid, almost ellipsoid with a fairly round apex, and the cap cuticle has a more distinct hypoderm. *C. sordidemaculatus* is a more robust, greyish-brown spruce forest species (Fig. 2 and Brandrud et al. 1998, plate D41 (photo)).

Another darkening species, *C. testaceofolius* H. Lindstr. & Soop, usually grows in spruce forests, but can also occur in pine heaths. It has a whitish, often somewhat radicating stipe, more brick-reddish gills, paler context, and more ovoid, strongly dextrinoid spores ( $8.0\text{--}9.2 \times 5.0\text{--}6.0 \mu\text{m}$ ), and distinctly incrustated hyphae of the gill trama. The fruitbody of *C. biformis* Fr. is paler and often has blue tints, and the spores are smaller, narrower, and more verrucose ( $7.5\text{--}8.2 \times 4.5\text{--}5.2 \mu\text{m}$ ). *C. brunneus* var. *clarobrunneus* H. Lindstr. & Melot has blackening fruitbodies and less reddish-brown colours as well as broader, almost subglobose spores ( $7.3\text{--}8.2 \times 5.2\text{--}6.5 \mu\text{m}$ ).

In pine heaths, there also grows a very common, darkening species, *C. suberi* var. *brunneogriseus* (Soop) Soop, which is robust, more greyish, and the cap surface is strongly fibrillose. The veil on the stipe is thicker and persistently white, possibly becoming pale pinkish when dried, and the spores are of similar size ( $8.0\text{--}9.0 \times 4.8\text{--}5.4 \mu\text{m}$ ) and shape but less dextrinoid.

*Cortinarius rubricosus* (Fr.) Fr. (Fries 1838) has many similarities to our species: a dark reddish-brown cap, light ferruginous gills and a veil formed like an appressed zone “velo exacte *C. armeniaci*”. Later Fries (1851) also mentions other suitable characteristics: a mountainous coniferous forest habitat, “hactus tantum pinetomontanis”, and a stipe, “extus intusque fuscus”. The epithet was first published (Fries 1818) as a variety, epsilon, of *Agaricus castaneus*. The protologue here is very short and vague. In the later descriptions,

however, there are also characteristics that do not fit well, such as the form of the stipe, “subaequale” (Fries 1838) or “deorsum attenuatus” (Fries 1851). The name was neotypified by Reumaux (Moëne-Loccoz et al. 1990, pl. 42, f. 82) on a collection growing with *Carpinus* in SE France. We conclude that this name is too uncertain to be used on a species in such a critical group as ours. Our species is also, as far as we know, bound to coniferous forests, mainly pine (*Pinus*).

Among the large number of names published by Henry, *Cortinarius dermagnitus* (Henry 1981, p. 196) could be the name for our species, but the cap is described as “ocre-alutacé”, the spores are rather strongly verrucose, and the epicutis is different. Type material was not available.

As we have not found any reliable name for our species in the literature we here publish a new one for it, *C. neofurvolaeus*. The epithet is used according to the similarity with the earlier published *C. furvolaeus* H. Lindstr. (= *C. sordidemaculatus* Rob. Henry) with which it has been confused.

**Specimens examined:** SWEDEN. **Dalarna:** Idre, Karmoråsen, *Kytövuori et al.* 00-008 (H). Mora, Vinäsgraven, *Lindström* 04.647 (UPS). Rättvik, Rättviksheden, *Lindström* 04.651 (H). Älvdalen, Oxgrav, *Kytövuori et al.* 00-009 (H). **Härjedalen:** Hede, 1 km öster om Hede, *Lindström et al.* CFP1438\* (holotype S, isotype H). **Medelpad:** Borgsjö, Gettryggen, *Lindström* 95.049 (UPS). Tuna, Sköle, *Lindström et al.* CFP 468\* (S). **Jämtland:** Fors, Kilen, *Lindström* 98.202 (UPS). Hällesjö, N of Hällesjön, *Lindström* 98.283 (UPS). Ragunda, Krängede, *Lindström* 96.683 (UPS); Prästberget, *Lindström* 90.085 (UPS). Rätan, Handsjö, *Niskanen et al.* 03-886\* (H), 03-974 (H). **Ångermanland:** Graninge, Viksmon, *Lindström* 95.061 (UPS); Åkroken, *Lindström* 95.050 (UPS). – FINLAND. **Varsinais-Suomi:** Lohja, *Niskanen et al.* 04-864 (H). Parainen, Lemlaxön, *Vauras* 5387F\* (TUR); Petteby, 13.IX.1978 *Alava et al.* (TUR). Suomusjärvi, Lahnajärvi, *Kytövuori* 03-001 (H), 03-002 (H); Vihti, Lintumäki, *Kytövuori et al.* 01-014 (H), 01-015 (H), 01-016 (H). **Uusimaa:** Espoo, Nuukio National Park, *Kytövuori et al.* 04-002\* (H), *Kytövuori* 04-015 (H); Sikalammet, *Skytén* 1516 (H). Hanko, Tvärminneby, *Kytövuori* 04-029 (H). Kirkkonummi, Meikoträsket, *Kytövuori et al.* 04-016 (H). Nurmijärvi, Kiljava, *Kytövuori* 04-005\* (H), 04-022\* (H), 04-023\* (H), 04-024 (H). Sipoo, Hindsby, *Kytövuori et al.* 04-030 (H). Tammisaari, Jomalävik, *Kytövuori* 01-019 (H); Lappohja, *Kytövuori et al.* 04-010 (H); Skärlandet, *Kytövuori et al.* 04-017\* (H), 04-018 (H); crossing to Snapertuna *Kytövuori* 04-025 (H), 04-026 (H), 04-027 (H), 04-028 (H). Vantaa, Tolkinkylä, *Kytövuori* 04-001\* (H). Vihti, Nummela, *Kytövuori* 04-021\* (H). **Etelä-Karjala:** Anjalankoski, Kaipainen, *Kytövuori* 94-568\* (H), 94-570\* (H, TUR), 94-577b (H), 94-806 (H).

Vehkalahti, Kitula, 3.10.1970 *Fagerström* (H). **Etelä-Häme:** Orivesi, Hirsilä, *Kytövuori* 95-1588\* (H). Ruovesi, Mustajärvi, *Kytövuori* 01-018 (H). Vilppula, Lyly, *Kytövuori* 98-1423 (H). Virrat, Monoskylä, *Kytövuori* 01-017 (H). **Etelä-Savo:** Kerimäki, Ruokojärvi, *Kytövuori* 98-1712 (H), 98-1713 (H), 98-1747 (H). Mäntyharju, Hietaniemi, *Kytövuori* 94-1164 (H). **Pohjois-Häme:** Konginkangas, Kivetty, *Kytövuori et al.* 02-002\* (H). Saarijärvi, Pyhä-Häkki National Park, *Kytövuori et al.* 02-013 (H). Virrat, Paskolampi, *Kytövuori* 95-1612 (H). **Pohjois-Savo:** Nilsia, Valkeiskylä, *Kytövuori et al.* 01-011 (H). **Pohjois-Karjala:** Ilomantsi, Mekrijärvi, *Kytövuori* 97-1582 (H). **Kainuu:** Kuhmo, Kähkölä, *Kytövuori* 99-703 (H). Suomussalmi, Lohivaara, *Kytövuori* 97-1298 (H); Suolijärvi, *Kytövuori* 97-1423 (H), 97-1449 (H). **Perä-Pohjanmaa:** Pello, Orajärvi *Kytövuori* 01-020 (H). **Koillismaa:** Kuusamo, Kantojoki, *Niskanen et al.* 02-524 (H), 02-971 (TUR), 02-975 (H), 02-977 (H), 02-978 (H), 02-1133 (H), 02-138 (H), 02-1139 (H); Oulanka National Park, *Niskanen et al.* 02-650 (H), 02-720 (H), 02-771 (H), 02-766\* (H), 02-769\* (H), 02-774\* (H), 02-795\* (H), 02-800 (TUR), 02-801 (OULU), 02-1094 (H), 02-1109 (H); Paljakka, *Niskanen et al.* 02-930 (H), 02-933 (H), 02-960 (H). Salla, Naruska, *Niskanen et al.* 02-441 (H). **Kittilän Lappi:** Kolari, Lappea, *Kytövuori* 98-1134 (H). – ESTONIA. **Hiiumaa:** Tahkuna, *Kytövuori et al.* 01-019 (H). **Võrumaa:** Haanja, Meeksi, *Kytövuori* 97-1981 (H).

### 3. *Cortinarius anisatus* H. Lindstr., Kytöv. & Niskanen, spec. nova – Figs. 4–7, 11

*Illustrations:* The photograph of the type will be published in Cortin. Fl. Photogr. 5.

*Pileus* 2.5–7 cm, convexus – obtuse conicus, dein planoconvexus, vix umbonatus, laevis, glaber, margine reflexo, sericeo, laeviter translucido; hygrophanus, udo sordide ochraceoargillaceus, usque fere testaceus, saepe punctis rubro-obscuris tinctus, sicco sordide ochraceus. Lamellae subconfertae, primo angustae, pallide argillaceo-ochraceae. Stipes 5–12 × 0.4–1(1.2) cm, cylindraceus, basi clavatobulbosus, apice fibrillosus, sordide albidus, basin versus demum griseofuscescens. Velum albidum, gracile, primo ochreiforme, dein interdum zonatum, mox evanesces. Caro ochraceobrunnea, variegata, infra fusca, nunquam caerulea observata. Odor dulcis, amoenus, anisodorus. Exsiccata sordide fusca. Sporae 7.5–9.1 × 5.0–6.1 μm, late ovatoelipsoideae, subvalde verrucosae. In silvis coniferis mediocriter humidis, cum *Picea abiete*.

*Holotypus:* Sweden, Ångermanland, Säbro parish, Hällenyland, in blueberry spruce forest (*Picea*, *Betula*), 27 Aug. 1993 *Lindström et al.* CFP 1200 (S, isotype H).

*Pileus* 2.5–7 cm, broadly conical to convex when young, soon planoconvex with a low umbo and narrowly down-curved margin (long persistent), sometimes wrinkled, when old sometimes with a reflexed outer margin, often narrowly pellucid-striate; surface glabrous, slightly glossy, with some fine, light fibrils, margin often silky whitish when young; saturated dull yellowish brown to somewhat reddish brown, sometimes more light yellowish brown to brown, sometimes with small, red dots and later dark strings or spots; hygroph-anous, drying up in a zone around the umbo and then centrifugally towards the margin to dull brownish yellow – light greyish brown, umbo resting (reddish) dark brown. *Lamellae* moderately crowded (50–60 reaching the stipe), strongly to weakly emarginate, middle thick, at first narrow then moderately broad, very light-coloured when young, then pale yellow brown, with age saturated brown, edge concolorous or white, uneven. *Stipe* 5–12 × 0.4–1 (1.2) cm, cylindrical, quite slender from the upper part, base clavate to bulbous (–2.5 cm), softening with age, top silky greyish white, downwards brown. *Mycelium* white. *Veil* white, forming a very thin, sock-like sheath, disappearing easily when bruised or with age, sometimes forming obscure zones. *Context* in pileus

reddish to yellow brown, darker in the umbo, in the stipe marbled brown, darker towards the base, bluish tints not observed. *Smell* in lamellae usually with a distinct smell of anise just like in *C. odorifer* Britzelm., sometimes missing and then slightly raphanoid. *Exsiccata* dark, sordid brown.

*Spores* 7.5–9.1 × 5.2–6.1 μm, Q=1.39–1.65 (280 spores, 17 collections (measurements from cortina 8 collections), one fruitbody from each, Figs. 5,6),  $\bar{X}$ =7.7–8.7 × 5.2–5.8 μm,  $\bar{X}Q$ =(1.38)1.44–1.51 (14 collections (measurements from cortina 8 collections), at least 10 spores measured from each), ovoid, weakly to clearly thick-walled, moderately to strongly dextrinoid, moderately verrucose, the size, and also the shape, rather variable between different collections; spores measured from the gills slightly narrower than those from the cortina at the top of the stipe. *Hyphae of the gill trama* fairly finely scabrous, in the overall view pale (olivaceous) yellowish. *Lamella edge* with fairly frequent clavate to bulbous sterile cells (11–33 × 6.5–14 μm). *Basidia* 4-spored, 30–35 × 6–10 μm, with fairly dark olivaceous contents. *Pileipellis* with moderately thin epicutis of 3–5 layers of hyphae, outer 3–5 μm, inner to 10 μm broad, almost hyaline to fairly strongly umber (intracellular pigment), smooth to fairly strongly



Fig. 11. Distribution of *Cortinarius anisatus* in NW Europe according to the material examined.

cross-striated incrustated, distinctly cemented by a gelatinous substance; hypoderm relatively well-developed, elements  $25\text{--}80 \times 10\text{--}25 \mu\text{m}$ , in the upper part colourless, hyphoid to irregular, and often fairly strongly cemented by a gelatinous substance, in the lower part more regular, angular, with pale umber thick-wall pigment; transition to the trama with distinctly incrustated hyphae. *Trama hyphae* irregular with a mixture of narrower and broader elements,  $4\text{--}14 \mu\text{m}$  wide, more or less hyaline. *Clamp connections* present.

*ITS-regions* (including 5.8S region) 508 bases long (total 9 sequences, Table 1). Intraspecific variation occurs in one position of the ITS 2 region. Difference to *C. neofurvolaeus* 21/30 bases, to *C. sordidamaculatus* 22/24 bases.

*Ecology and distribution:* According to the present material, *C. anisatus* grows in submesic spruce forests, often on calcareous ground, but also in ordinary blueberry spruce forests, often in deep moss cover. It grows in loose groups or solitarily. It has predominantly been found in older forests, but it also grows in fairly young stands. Fruitbodies occur from early August to late September (Fig. 7).

*Cortinarius anisatus* is not rare, but is sparse in suitable habitats. So far, we know it is found only in boreal and hemiboreal Fennoscandia and Estonia, and it seems more common towards the north (Fig. 11).

*Differential diagnosis:* *Cortinarius anisatus* is a middle sized, brown, with age darkening *Telamonia* species growing in spruce forests, and it is best known by its delightful, aniseed smell (like in *C. odorifer*). It also typically has a narrowly pellucid-striate cap margin, usually slender, at the base clavate stipe and a white, thin, sheath-like veil. Dried specimens can be identified by the combination of sordid dark brown colour and the rather small, ovoid, somewhat thick-walled, dextrinoid spores. It is important however, to keep in mind that fruitbodies without aniseed aroma are not uncommon, and the pellucid-striate cap margin and the characteristic shape of the spores are most diagnostic. Some correlation between raphanoid smell and large spores was observed in two collections, which were also sequenced. These had one base difference compared to the other collections. Further studies are needed.

In mixed spruce/ pine forests *C. brunneus* var. *clarobrunneus*, which can somewhat resemble *C. anisatus*, may also grow. The former is also

brown and can smell of anise, it differs from *C. anisatus* however, by having a not pellucid-striate cap margin, a thicker, not sock-like veil, and subglobose, more verrucose and only slightly dextrinoid spores ( $7.3\text{--}8.2 \times 5.2\text{--}6.5 \mu\text{m}$ ).

A related species, *C. sordidamaculatus*, also grows in rich spruce forests. It is usually more robust, lacks an aniseed smell and pellucid-striate cap margin, has a thicker veil, and has narrower, thinner-walled and less dextrinoid spores. *C. illuminus* has a pellucid-striate cap margin, but it differs from *C. anisatus* by a more saturated red cap colour, raphanoid smell, and round, small spores. *C. biformis* usually has blue tints in its fruit body, a silky-shiny cap, indistinct or slightly raphanoid smell, narrower spores, and fruitbodies darken only weakly when dried.

For this type of anonymous looking *Telamonia* species it is not easy to find a name among the classical authors' short descriptions, especially if the most striking characteristic, the smell, is not indicated. Velenovský's *Hydrocybe sordida* (Velenovský 1921, p. 477), has a distinct, mild aromatic smell, and may be similar to our species. Other macroscopic characteristics reported, also fit rather well, but Velenovský's species is vernal (March to May), and the spore size reported ( $8\text{--}10 \mu\text{m}$ ) is rather large compared to ours (despite Velenovský usually reports too large size for spores, own experience). In a later description (Velenovský 1939, p. 113), his species is quite different with bigger cap size and smaller spores. As we have no access to type material, we find this name to be doubtful, especially since we know that aromatic smell is not restricted to just one darkening *Telamonia* species.

Henry (1981, p. 250) published *C. phaeosmus* with the cap colour like *Pluteus cervinus*, stipe abruptly bulbous, a fibrous veil, and a smell like rhubarb or parsley. This fungus, however, is found in deciduous forests, and the spores are different from the ones of our species (too large and of the wrong shape). Henry reports a spore size of  $8.7\text{--}10.8 \times 3.9\text{--}5 \mu\text{m}$ , but our measurements from the type (Henry no. 80828, PC 0088381) are  $8.6\text{--}9.5 \times 4.9\text{--}5.2$  ( $5.6$ )  $\mu\text{m}$  (20 spores from the gill, one fruitbody) and the shape is elongated obovoid. *C. phaeosmus* seems to be a taxon close to *C. rheubarbarinus* Rob. Henry 1956 (Henry 1956, p. 229). An interpretation of *C. phaeosmus* is also found in Atlas des Cortinaires pars X (Bidaud et al. 2000, pl. 310, f. 484).

*Cortinarius pseudocaninus* Rob. Henry (Henry 1981, p. 219) is described as having characteristics like our species, but the gills are very distant and spores are too large ( $8.3\text{--}10.8 \times 4.7\text{--}6.5 \mu\text{m}$ , Henry 1981). Type material of this name was not available. *C. sacchariosmus* Beller & Bon 1975 (Bon 1975, p. 7) has a similar smell but different spores ( $9.0\text{--}10.0 \times 4.5\text{--}5.5 \mu\text{m}$ ), and seems to belong to the section *Hinnulei*.

Thus we give a new name, *C. anisatus* to our species. The epithet refers to the similarity to the smell of the *Pimpinella anisum* fruits.

**Specimens examined:** SWEDEN. Småland. Femsjö, Södra Färgen, 18.IX.1987 *Bendiksen & Metsänheimo\** (O). Dalarna: Idre, Karmoråsen, *Kytövuori et al. 00-005\** (H). Hälsingland: Bergsjö, Ede, *Niskanen et al. 03-369* (TUR), *03-372\** (H). Medelpad: Haverö, Snöberget, *Niskanen et al. 03-558\** (H), *03-567\** (H), *03-596\** (H), *03-603* (TUR), *03-625\** (H). Parteboda, Puttrå, *Niskanen et al. 03-643* (H). Skön, Petersvik, *Lindström 98.869* (UPS). Torp, Hästnäset, *Lindström 99.148* (UPS). Jämtland: Frösö, near Frösö zoo, *Lindström 00.002* (UPS). Håsjö, Kvarnån, *Lindström 95.010* (UPS); Lövsjön, *Lindström et al. CFP1292\** (UPS). Lockne, Tandbyn, *Niskanen et al. 03-797* (H). Ragunda, Hoo, *Lindström 93.042* (UPS). Ångermanland Stigsjö, Usland, *Lindström 95.030* (UPS). Säbro, Hällenyland, *Lindström et al. CFP1200\** (holotype S, isotype H). – FINLAND. Varsinais-Suomi: Suomensjärvi-Kisko, Lemulanrinne, *Kytövuori 98-2196* (H). Uusimaa: Sipoo, Paippinen, *Niskanen et al. 04-807* (H). Satakunta: Ikaalinen, Seitsemäinen National Park, *Niskanen et al. 04-200* (H). Etelä-Häme: Juupajoki, Hyytiälä, *Niskanen et al. 04-075* (H). Ruovesi, Sii-kakangas, *Niskanen et al. 04-130* (OULU), *04-152\** (H). Pohjois-Häme: Konginkangas, Kivetty, *Kytövuori et al. 02-012* (H). Konnevesi, Kodanovinen, 8.IX.1979 *Issakainen* (H); Tuhola, *Kytövuori et al. 02-017* (H); Töhrkerönmäki *Kytövuori 04-013* (H). Kainuu: Puolanka, Pihlajavaara, *Kytövuori 02-010* (H); Väyrylä, *Kytövuori et al. 02-004\** (H). Suomussalmi, Kiannanniemi, *Kytövuori 02-005* (H). Perä-Pohjanmaa: Rovaniemi rural commune, Louevaara, *Niskanen et al. 04-383* (H), *04-467* (H). Tervola, Peura, *Niskanen et al. 04-380* (H). Tornio, Kalkkimaa, *Niskanen et al. 04-594* (H), *04-640\** (H); Korkiamaa, *Niskanen et al. 04-550* (H, TUR, OULU). Koillismaa: Kuusamo, Oulanka National Park, *Niskanen et al. 02-811* (H), *02-1031* (H), *02-1063* (H). – ESTONIA. Hiiumaa: Tahkuna, *Kytövuori et al. 01-012* (H).

## Discussion

In this paper we use a species concept based on the following idea: between species there mainly is no genetic flow. This causes discontinuous genetic variation and with time differences in

morphology and ecology. We try to find these differences (genetic, morphological and ecological) and use them for the classification and identification of the species.

Genetic flow occurs below the species level. Populations in different areas can be somewhat different, but between these areas intermediates can be observed, and when growing in the same place and habitat they can not stay separate. Therefore, if we find dissimilar fruitbodies growing in the same habitat they can not belong to the taxa below the species level, but they can be different-looking individuals of the same species. If similar fruitbodies however, are found in different geographical areas, and are still possible to separate by the same characteristics (more than one to eliminate the possibility that the difference is caused by different alleles of one species), they must belong to different species.

Because there are so many species of *Cortinarius*, most characteristics are strongly overlapping. Most important is to try to differentiate between variation due to real intermediates, and overlapping characteristics. To study this we must find a characteristic (e.g. color, smell, spore size, chemical reactions, mycorrhizal partner, etc.) with which we can preliminary divide the collections. DNA has proved very useful for this preliminary division. Correlation with other characteristics can then be studied. If we can identify a taxon and separate it continuously from the others it is a species, despite how big the differences are or how many differences are found. Our species concept is based on discontinuous variation and correlation between the characteristics. However, mating of fungi is poorly known, and conclusions are based on current knowledge.

*Cortinarius sordidemaculatus*, *C. neofurvo-laesus* and *C. anisatus* are closely related species, but they are clearly separate by more than one morphological characteristic. They are also well supported by differences in the ITS-sequences: the genetic intraspecific variation in each taxon is low, and the differences between the species are clear (discontinuous variation). Inside the clade of these three species the relationships are not well supported or stable. The relationships can not be resolved however, with the ITS-regions alone, and more data is needed for further conclusions.

The higher systematic position of *C. sordidemaculatus*, *C. neofurvo-laesus* and *C. anisatus*

is uncertain. Studying the systematic position is difficult because there is no widely accepted classification of *Cortinarius* subgenus *Telamonia*, and most of the groups created earlier have proved to be artificial (unpublished). Also, this group of darkening *Telamonia* species is poorly studied, and many of the species, among them the closest relatives, are not yet sufficiently known (unpublished).

In Brandrud et al. (1998) *C. furvolaesus* (= *C. sordidemaculatus*) was preliminary placed in section *Brunnei* Kühner & Romagn. ex Melot (Melot 1990). Morphologically the dark colour of the fruitbodies and the fairly dark outlook of the spores in the microscope tend to support this placing. The most striking characteristics of the *Brunnei* species however, the strongly (greyish-) blackening fruitbodies when dried, is absent from our species, which become only sordid brown. The latter characteristic is most typical to section *Sordescentes* Melot (Melot 1990). The smooth or only very weakly incrustated hyphae in the gill trama also fit well with the section *Sordescentes*. Henry has placed his *C. sordidemaculatus* in subgenus *Hydrotelamonia* in the *C. biveloides* group (Henry 1981).

The results of DNA studies (Fig. 1) show that in our study of selected taxa the closest relative is not *C. brunneus*, but *C. cf. bovinus* (photo collection in Moser & Jülich 1990), although the support value is low (< 50%). *C. bovinus* Fr. is interpreted in different ways by different authors and includes several species. We have chosen the photo plate *Cortinarius* 65 in Moser & Jülich (1990) to represent the section *Sordescentes*. Based on morphological and DNA studies we place *C. sordidemaculatus*, *C. anisatus* and *C. neofurvolaesus* within the section *Sordescentes*. Since DNA results are strongly affecting the taxonomy above the species level in *Cortinarius*, it is not necessary at the present time to discuss the sections in greater detail.

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