

Chemotaxonomic studies on the pigmentation of North American species of *Dermocybe* (Fr.) Wünsche, section *Dermocybe* and related species*

G. Keller¹ & J. F. Ammirati²

¹Eichenweg 14, A-6460 Imst, Austria

²Department of Botany, KB-15, University of Washington, Seattle, 98195, Washington, USA

Keller, G. & J. F. Ammirati (1995). Chemotaxonomic studies on the pigmentation of North American species of *Dermocybe* (Fr.) Wünsche, section *Dermocybe* and related species. – Beih. Sydowia X: 127–136.

Pigmentation of 11 yellow and orange gilled taxa of *Dermocybe* from North America were investigated by means of thin layer chromatography. A comparison of pigment data showed more or less specific pigment patterns and six types of pigmentation within the species studied. The importance of pigmentation and chemical relationships to European *Dermocybe* species are discussed.

Keywords: Agaricales, taxonomy.

A great variety of anthraquinonic pigments are found in the sporocarps of *Dermocybe* species. Their chemical properties and structures have been extensively studied (for review see Gill & Steglich, 1987), while their occurrence and distribution have proven useful in differentiating infrageneric taxa (Gabriel, 1965; Gruber, 1970; Keller, 1982; Høiland, 1983). The relevance of chemical characters for taxonomy is discussed by Moser (1985).

In an earlier study Keller & Ammirati (1983) reported a number of anthraquinone and anthraquinonoid pigments in red gilled species of *Dermocybe*, section *Sanguineae*. Furthermore, it was shown that strong chemotaxonomic relationships exist between European and North American *Dermocybe* spp. In view of the importance of the anthraquinonic pigments as taxonomic criteria in the systematics of *Dermocybe*, the present study was carried out to determine the pigments and pigment patterns of some of the yellow and orange gilled *Dermocybe* species in North America.

This paper is dedicated to Professor M. Moser on the occasion of his seventieth birthday.

Material and methods

Several collections used in this study were made by the authors in the fall of 1980 in Oregon and Washington, USA. These and additional collections are listed in Tab. 1. Herbaria are indicated by the appropriate abbreviations from Index Herbariorum. The taxa treated in this paper are placed in the sections *Dermocybe*, *Holoxanthae* Mos. and *Malicoriae* Mos. (Moser, 1972).

Sample preparation and techniques of thin layer chromatography (TLC) of pigments are as described in Keller (1982) and Keller & Ammirati (1983) with minor modifications. Chromatography was done on precoated TLC plates (silica gel 60). Five solvent systems were employed to detect pigment patterns of each species. Further chromatographic data are given in Tab. 2. The developed chromatograms were dried and examined in daylight and UV light (365 nm). For further identification of pigments TLC plates were sprayed with reagents potassium hydroxide (5% in methanol) and magnesium acetate (5% in methanol).

Results

Chromatographic examination of sporocarps of yellow and orange gilled *Dermocybe* spp. from North America revealed the presence of a number of anthraquinone and anthraquinonoid pigments. The anthraquinone carboxylic acids endocrocin, dermolutein and dermorubin were identified with authentic reference samples, as well as the neutral anthraquinone derivatives emodin, emodin-glucoside, dermoglaucin, dermocycin and dermocycin-glucoside, and the dimeric dihydroanthracenone flavomannin-6,6'-dimethyl ether.

More than 20 pigments were distinguished on the chromatograms of the species investigated. The pigments and their chromatographic properties are listed in Tab. 3. With the exception of flavomannin-6,6'-dimethyl ether all pigments show a positive colour reaction with the reagent potassium hydroxide (and magnesium acetate), which suggests the anthraquinonic nature of the identified pigments as well as the unknown coloured compounds. It is also suggested from the chromatographic properties that many of the unknown pigments may be glycosides of the anthraquinones themselves. Furthermore, additional pigments have been detected chromatographically with weak intensities. Flavomannin-6,6'-dimethyl ether is always accompanied by two other pigments: the first is anhydroflavomannin-9,10-quinone-6,6'-dimethyl ether, which may be partly an artifact, the second is probably 7,7'-biphyscion which has been isolated as an artifact formed during storage and chromatography (Steglich & al.,

Tab. 1. – *Dermocybe* species included in this study and their origin.

Taxon	collection no.	Herbarium	Date of collection	Site of collection	Collector
<i>D. crocea</i> (Schff.) Mos.	80/81	IB	13.11.1980	Mason Lake, Mason Co., Washington, USA	G. Keller
<i>D. crocea</i>	80/15	IB	23.10.1980	Mason Lake, Mason Co., Washington, USA	G. Keller
<i>D. crocea</i>	80/56	IB	8.11.1980	San Juan Island, San Juan Co., Washington, USA	H. Keller-Dilitz
<i>D. zakii</i> (Amm. & Smith) Amm.	80/29	IB	22.10.1980	Grays Harbor Co., Washington, USA	J. Rogers
<i>D. incognita</i> (Amm. & Smith) Amm.	7966	WTU	5.9.1977	Macey Lake Bog, Simcoe Co., Ontario, Canada	C. Ovrebo
<i>D. uliginosa</i> (Berk.) Mos.	80/149	IB	28.11.1980	Redmond, King Co., Washington, USA	J. F. Ammirati
<i>D. uliginosa</i>	80/30	IB	26.10.1980	Redmond, King Co., Washington, USA	S. Rehner
<i>D. cinnamomea</i> (L.: Fr.) Wünsche	80/89	IB	13.11.1980	Mason Lake, Mason Co., Washington, USA	G. Keller
<i>D. subcroceifolia</i> (Amm. & Smith) Amm.	80/68	IB	13.11.1980	Mason Lake, Mason Co., Washington, USA	G. Keller
<i>D. subcroceifolia</i>	80/100	IB	24.11.1980	Ocean Shores, Grays Harbor Co., Washington, USA	G. Keller
<i>D. subcroceifolia</i>	80/101	IB	24.11.1980	Ocean Shores, Grays Harbor Co., Washington, USA	H. Keller-Dilitz
<i>D. subcroceifolia</i>	80/22	IB	23.10.1980	Mason Lake, Mason Co., Washington, USA	G. Keller
<i>D. subcroceifolia</i>	80/108	IB	25.11.1980	Ocean City, Grays Harbor Co., Washington, USA	G. Keller
<i>D. subcroceifolia</i>	8295	WTU	25.9.1978	Dorset, Holiburton Co., Ontario, Canada	J. F. Ammirati
<i>D. tubaria</i> (Amm. & Smith) Amm.	7302	WTU	19.9.1975	Macey Lake, Simcoe Co., Ontario, Canada	J. F. Ammirati
<i>D. tubaria</i>	7909	WTU	13.9.1977	Macey Lake, Simcoe Co., Ontario, Canada	J. F. Ammirati
<i>D. aureifolia</i> (Peck) Mos.					
var. <i>hesperia</i> (Amm. & Smith) Amm.	80/163	IB	12.12.1980	Sand Lake, Tillamook Co., Oregon, USA	J. F. Ammirati
<i>D. aureifolia</i> var. <i>hesperia</i>	80/152	IB	12.12.1980	Sand Lake, Tillamook Co., Oregon, USA	G. Keller
<i>D. malicoria</i> (Fr.) Rick.	8013	WTU	17.9.1977	Clearwater Co., Minnesota, USA	W. J. Sundberg
<i>D. malicoria</i>	8282	WTU	16.9.1978	Baraga Co., Michigan, USA	J. F. Ammirati
<i>D. malicoria</i>	80/36	IB	26.10.1980	Redmond, King Co., Washington, USA	S. Rehner
<i>D. idahoensis</i> (Amm. & Smith) Amm.	80/9	IB	18.10.1980	Lake Kachess, Kittitas Co., Washington, USA	G. Keller
<i>D. idahoensis</i>	80/10	IB	18.10.1980	Lake Kachess, Kittitas Co., Washington, USA	G. Keller

Tab. 2. Chromatographic systems. Layer: precoated TLC plates silica gel 60 (MERCK), thickness of layer 0.25 mm.

Chromatographic system	Solvent (v/v)
I	benzene acetic acid (glacial) 2 1
II	ethyl acetate methanol water 100 16.5 13.5
III	toluene ethyl formate formic acid 160 40 1
VI	toluene ethyl formate formic acid 50 40 10
VII	ethyl acetate ethyl methyl ketone formic acid water 50 30 10 10

1972). Distribution of pigments and pigment patterns in the *Dermocybe* species investigated are shown in Tab. 4.

D. crocea, *D. crocea* var. 80/81, *D. incognita*. – Chromatographic examination of these yellow gilled *Dermocybes* showed that flavomanin-6,6'-dimethyl ether is the major coloured compound. Endocrocin and dermolutein were detected with medium intensities. Dermorubin was observed only with weak intensity in *D. crocea* var. 80/81 but with medium to high intensities in the other taxa. Pigmentation of these species is similar to that of *D. crocea* found in Europe (Keller, 1982).

D. uliginosa. – Chromatographic examination of this species revealed a *cinnamomea*-type pigmentation. Flavomannin-6,6'-dimethyl ether and dermorubin were found to be the major pigments, whereas dermolutein and endocrocin were observed with lower intensities. There is no significant difference between pigment patterns of North American collections and those found in Europe (Gruber, 1970; Keller, 1982; Høiland, 1983).

D. cinnamomea, *D. subcroceofolia*, *D. zakii*. – The major pigment of these orange gilled species has been identified as flavomannin-6,6'-dimethyl ether. Dermorubin has been detected as the major anthraquinone carboxylic acid, whereas endocrocin and dermolutein have been found with low to medium intensities. The pigment patterns are very similar between the taxa investigated and to those of specimens of *D. cinnamomea* collected in Europe (Gruber, 1970; Keller, 1982; Høiland, 1983).

Dermocybe tubaria. – The major pigments of this species have been identified as endocrocin and dermolutein as aglycones and possibly as glycosides. None or only small amounts of dermorubin have been detected. Dermoglaucin, possibly dermoglaucin-glycoside

Tab. 3 – Chromatographic data of anthraquinonic pigments detected in *Dermocybe* species. Chromatographic conditions are given in the Experimental section and in Tab. 2. (?) indicates identity of pigment not certain. Abbreviations: FDM: Flavomannin-6,6'-dimethyl ether, AFDM: Anhydroflavomannin-9,10-quinone-6,6'-dimethyl ether.

Pigment	Rf values × 100 in chromatographic systems							Colour in daylight	Colour under UV light	Colour reaction with KOH
	I	II	III	VI	VII					
7,7'-Biphyscion (?)	82	80	60	78	98		yellow	orange		purple red
Emodin	77						yellow	brown		orange
AFDM	64	77	32	62	98		yellow	orange		purple red
Dermoglaucin	58	70	08	49	95		yellow	dark brown		purple red
Dermocybin	50-62	55-63	00-22	50-60	95		brown	dark		greyish brown
	40-60	15-30	00-18	50-60	93		purple red	dark violet		purplish violet
	39						orange	orange		orange red
FDM	34	30-55	02	37	90		lemon	orange		yellow
	31	23	02	47	92		pink	pink orange		violet
Endocrocin	30	24	02	45	90		yellow	orange		pink
5-Cl-dermolutein (?)	18	12	00				yellow	orange		purplish violet
5-Cl-dermorubin	17	10	00				red	purple		purple violet
Dermolutein	16	16	00				yellow	orange		purple red
Dermorubin	12	14	00	33	88		red	red orange		purple violet
	05	41	00	07	60		yellow	orange		orange red
Emodin-glucoside	05						yellow	dark		greyish violet
Dermoglaucin-glycoside (?)	05	28-32	00	04	44		brown	dark violet		purplish violet
Dermocybin-glycoside	04	00-18	00	03	42		purple red	orange red		red orange
Endocrocin-glycoside (?)	02	07	00	00	48		yellow	pink orange		pink
Dermolutein-glycoside (?)	02	04	00	00	38		yellow	purple		purple
Dermorubin-glycoside (?)	02	03	00	00	36		red			

and flavomannin-6,6'-dimethyl ether were found with low to medium intensities. There is no significant difference between pigment patterns of North American specimens and those found in Europe [Keller, 1982 (= *D. sphagnetii*)].

D. aureifolia var. *hesperia*. – Chromatographic examination of this *Dermocybe* revealed a special pigmentation. Flavomannin-6,6'-dimethyl ether is the major colouring principle. Endocrocin, dermolutein and dermorubin were found with medium intensities. Furthermore, emodin and an unknown pink pigment (Rf 0.31, System I) were readily detected with medium intensities. The latter compound was never observed on chromatograms of other species included in this study. These results are not in accordance with those from a pigment analysis of *D. aureifolia* var. *aureifolia* by Høiland (1983). However, no flavomannin-6,6'-dimethyl ether or emodin were found by TLC in the type material of this *Dermocybe* species.

D. malicoria. – Emodin and emodin-glucoside are the predominant coloured compounds of this bright orange gilled *Dermocybe* species. Flavomannin-6,6'-dimethyl ether has been observed as a major pigment with intensities comparable to species of the *cinnamomea*-complex. Dermolutein has been detected with medium intensities whereas endocrocin and dermorubin are minor constituents. Furthermore, only low amounts of dermoglaucin were found. Similar pigment patterns have been reported for European collections of this species but dermoglaucin was not detected (Gruber, 1970; Keller, 1982; Høiland, 1983).

D. idahoensis. – It is apparent from the chromatographic data that *D. idahoensis* is a very complex species. Flavomannin-6,6'-dimethyl ether, emodin, dermoglaucin and dermolutein are the major pigments of this species whereas dermocycin, endocrocin, dermolutein and an unknown orange compound (Rf 0.39, System I) were observed with low or medium intensities. These results correspond with a pigment analysis of *D. idahoensis* by Høiland (1983).

Discussion

Chromatographic examination of yellow and orange gilled *Dermocybe* species from North America reveals that a variety of anthraquinonic pigments are present in sporocarps of the species investigated. A comparison of pigmentation data shows that each

Tab. 4. —Distribution of pigments and pigment patterns in *Dermocybe* species investigated. Symbols: +, ++, +++, +++++ indicate relative intensity, (+) indicates traces of pigment detectable, — indicates pigment not detected, ≤ indicates area of detection covered by another pigment. (?) indicates identity of pigment not certain. Abbreviations: FDM: Flavomannin-6,6'-dimethyl ether, AFDM: Anhydroflavomannin-9,10-quinone-6,6'-dimethyl ether.

Pigment [Rf-value, colour, System I]	<i>Dermocybe</i> species									
	<i>crocea</i> var. 80/81	<i>crocea</i>	<i>incognita</i>	<i>uliginosa</i>	<i>zakii</i>	<i>cinnamomea subcroceifolia</i>	<i>tubaria</i>	<i>aureifolia</i> var. <i>hesperia</i>	<i>malicoria</i>	<i>idahoensis</i>
7,7'-Biphsycion (?)	—	++++	—	+/+++	—	—	—	+/+++	+/++	+
[0.77, yellow]	—	—	—	—	—	—	—	—	—/(+)	(+)/+
Emodin	—	—	—	—	—	—	—	+/++	++++	+++
AFDM	—	++++	—	+/++	—	+/+++	(+)/+	+/++	++	++
Dermoglaucin	—	—	—	—	—	—	+	—	+	—
Dermocyanin	—	—	—	—	—	—	—	—	—	—
[0.39, orange]	—	—	—	—	—	—	—	—	—	—
FDM	—	—	—	+/++	—	+/+++	(+)	—	+/+++	—
[0.31, pink]	—	—	—	—	—	—	—	—	—	—
Endocrocin	—	+	—	(+)/+	+/++	(+)/+	+/++	—	+	—
5-Cl-dermolutein (?)	—	—/+	—	—	—	—/(+)	(+)	—	—/+	—
5-Cl-dermorubin	(+)/+	—/+	(+)	+/++	+	—/+	—	(+)/+	—/(+)	+/++
Dermolutein	+	++	+	(+)/+	(+)/+	+/++	+++	++	++	(+)/+
Dermorubin	(+)/+	++	+++	+/+++	++	+/+++	—/(+)	++	(+)/+	+++
Emodin-glucoside	—	—	—	—	—	—	—	—	+++	++
Dermoglaucin-glycoside (?)	—	—	—	—	—	—	(+)/+	—	—	++
Dermocyanin-glycoside	—	—	—	—	—	—	—	—	—	—
Endocrocin-glycoside (?)	+	+/++	+	(+)/+	+	(+)/+	+/++	—	—	+
Dermolutein-glycoside (?)	(+)	(+)/+	(+)	—/+	+	(+)/+	+/++	—/+	+	(+)
Dermorubin-glycoside (?)	—	—	+	—/+	+	(+)/+	—	—/+	—/+	—

species has a more or less specific pigment pattern. The resulting chemotaxonomic relationships of the species are shown in Tab. 5. Two supraspecific levels of pigmentation are found in the species investigated, which all contain flavomannin-6,6'-dimethyl ether and anthraquinone carboxylic acids. Another group comprises *D. tubaria*, *D. aureifolia* var. *hesperia*, *D. malicoria* and *D. idahoensis*, which contain neutral anthraquinone derivatives, whereas the remaining species form a second group lacking this type of pigment. These groups have direct relationships to established infrageneric categories of *Dermocybe* since the present taxonomic structure of this genus is strongly influenced by chemotaxonomic data.

The species lacking neutral anthraquinone pigments share a close chemical relationship and can only be separated into two subgroups depending on the intensity of dermorubin. In *D. crocea* var. 80/81 only small amounts of dermorubin have been detected. In the remaining taxa dermorubin is recorded with medium or high intensities as it is reported from European collections of *D. crocea* and *D. cinnamomea* (Keller, 1982; Høiland, 1983). Although there is some gross morphological variation only relatively small differences are observed in pigment patterns between taxa of this subgroup.

Within the group of species that do contain neutral anthraquinonic derivatives somewhat more diversity in pigmentation can be recognized. The following four types of pigmentation have been distinguished depending on the occurrence of neutral anthraquinone derivatives. In *D. tubaria* dermoglaucin is the only neutral anthraquinone pigment that is reported to be a specific compound in European collections of this species [Gruber, 1970 (= *D. palustris*); Keller, 1982; Høiland, 1983]. In *D. aureifolia* var. *hesperia* emodin has been detected in medium amounts. This is not in accordance with chromatographic data from a study by Høiland (1983), where type specimens of *D. aureifolia* var. *aureifolia* collected from a different location lack emodin and even flavomannin-6,6'-dimethyl ether. *D. aureifolia* is not known from Europe. In *D. malicoria* emodin has been observed as the major pigment and is accompanied by dermoglaucin. This species shows a pigment pattern that is very similar to those reported for *Cortinarius sommerfeltii* Høil. found in Europe (Keller, 1982; Høiland, 1983; Arnold & al., 1991). Emodin, but no dermoglaucin, was detected in European material of *D. malicoria* with similar intensities (Keller, 1982; Høiland, 1983). Of the taxa studied *D. idahoensis* revealed one of the most interesting pigment patterns. The presence of the neutral anthraquinone derivatives emodin, dermoglaucin and dermocycin were found to be specific for this *Dermocybe*. No chemical counterpart from Europe is known to this unique orange-ochraceous gilled *Dermocybe* species.

Tab. 5. – Criteria in pigmentation used in the separation of groups and in the circumscription of types of pigmentation.

flavomannin-6,6'-dimethyl ether and anthraquinone carboxylic acids present			
neutral anthraquinone derivatives absent		neutral anthraquinone derivatives present	
dermorubin weak	dermorubin with high intensities	emodin absent dermoglaucin present	emodin present
		emodin with medium intensities	emodin with high intensities dermoglaucin present
		dermocyanin absent	dermocyanin present
<i>D. crocea</i> var. 80/81	<i>D. crocea</i> <i>D. zakii</i> <i>D. incognita</i> <i>D. uliginosa</i> <i>D. subcroceifolia</i> <i>D. cinnamomea</i>	<i>D. tubaria</i>	<i>D. malicoria</i> <i>D. idahoensis</i>
<i>crocea</i> -type	<i>cinnamomea</i> -type	<i>tubaria</i> -type	<i>malicoria</i> -type <i>idahoensis</i> -type
type of pigmentation			

Acknowledgments

This work was supported in part by a grant from the Bundesministerium für Wissenschaft und Forschung, Vienna and a grant from the National Science Foundation (DEB-8118972). We wish to thank Dr. M. Moser, Institut für Mikrobiologie, Universität Innsbruck, for his interest and his help in this study. We also wish to thank Dr. W. Steglich, Chemisches Institut, Universität Bonn, for the generous supply of anthraquinone derivatives utilized as reference samples in this study. We are indebted to the mycologists listed in Tab. 1 for collecting and shipping specimens of investigated species.

References

- Arnold, N., H. Besl, A. Bresinsky & H. Kemmer (1991). Notizen zur Chemotaxonomie der Gattung *Dermocybe* (Agaricales) und zu ihrem Vorkommen in Bayern. – Z. Mykol. 53: 187–194.
- Gabriel, M. (1965). Contribution a la Chimiotaxonomie des Agaricales. Pigments des Bolets et des Cortinaires. – Thèse, Lyon.
- Gill, M. & W. Steglich (1987). Pigments of fungi (Macromycetes). – Fortschr. Chem. Org. Naturst. 51: 1–317.
- Gruber, I. (1970). Anthrachinonfarbstoffe in der Gattung *Dermocybe* und Versuch ihrer Auswertung für die Systematik. – Z. Pilzk. 36: 95–112.
- Høiland, K. (1983). *Cortinarius* subgenus *Dermocybe* with special reference to the species in the Nordic countries. – Opera Botanica 71: 1–112.
- Keller, G. (1982). Pigmentationsuntersuchungen bei europäischen Arten aus der Gattung *Dermocybe* (Fr.) Wünsche. – Sydowia 35: 110–126.
- & J. F. Ammirati (1983). Chemotaxonomic significance of anthraquinone derivatives in North American species of *Dermocybe*, section *Sanguineae*. Mycotaxon 18: 357–377.
- Moser, M. (1972). Die Gattung *Dermocybe* (Fr.) Wünsche (Die Hautköpfe). Schweiz. Zeitschr. f. Pilzk. 50: 153–167.
- (1985). The relevance of chemical characters for the taxonomy of Agaricales. – Proc. Indian Acad. Sci. 94: 381–386.
- Steglich, W., E. Töpfer-Petersen, W. Reininger, K. Gluchoff & N. Arpin (1972). Isolation of flavomannin-6,6'-dimethyl ether and one of its racemates from higher fungi. – Phytochemistry 11: 3299–3304.

(Manuscript accepted 20th June 1994)

ZOBODAT - www.zobodat.at

Zoologisch-Botanische Datenbank/Zoological-Botanical Database

Digitale Literatur/Digital Literature

Zeitschrift/Journal: [Sydowia Beihefte](#)

Jahr/Year: 1995

Band/Volume: [10](#)

Autor(en)/Author(s): Keller Gerwin, Ammirati Joseph F.

Artikel/Article: [Chemotaxonomic studies on the pigmentation of North American species of Dermocybe \(Fr.\) Wiinsche, section Dermocybe and related species 127-136](#)