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***PHYSCIA BIZIANA* AND *PHYSCIA STELLARIS* CHEMICAL, ANALYTICAL AND PHARMACOLOGICAL DATA (LITERATURE REVIEW)**

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Abstract. *Physcia biziana* and *Physcia stellaris* are cosmopolitan foliose lichens of section *Stellares* whose chemistry has been documented almost exclusively through taxonomic spot tests and thin-layer chromatography rather than through dedicated phytochemical investigation. Both species share an invariant cortical β -orcinol depside, atranorin ($C_{19}H_{18}O_8$), and a K[–] medulla. Beyond this shared core, the published record is contradictory: hopane triterpenoids (zeorin, leucotylin) are reported present in some populations of both taxa and explicitly absent in others. This review compiles the primary chemical data, presents the structures of the relevant metabolites, resolves the apparent contradiction between the K spot test and triterpenoid occurrence, and identifies the analytical and pharmacological work required to place the chemistry of these two species on a modern footing.

Keywords: *Physcia biziana*, *Physcia stellaris*, atranorin, zeorin, leucotylin, antimicrobial, antioxidant and cytotoxicity.

Introduction. *Physcia* (Schreb.) Michx. is a cosmopolitan genus of foliose lichens comprising approximately 75 species. Chemically the genus operates a two-layer strategy: a cortical depside (atranorin), responsible for the diagnostic K⁺ yellow reaction, combined with a medullary suite of hopane-type triterpenoids whose composition varies between species. The genus is distinguished from most other foliose Physciaceae chiefly by the combination of a pseudoparenchymatous upper cortex with atranorin as the cortical substance [2].

Physcia biziana and *P. stellaris* both belong to Lynge's section *Stellares*, circumscribed by non-ciliate lobe margins, a pale lower cortex, and — in the classical definition — a K[–] medulla with zeorin absent [1],[3]. Both are esorediate and regularly apotheciate, and both are broadly distributed across Europe, Asia, Africa, Australasia and the Americas. Despite this ubiquity, neither has been the subject of an isolation-grade chemical study. The present

review assembles what is actually documented, exposes where the record conflicts with itself, and proposes a workflow for resolving it.

The aromatic metabolites of *Physcia* derive from the acetyl–polymalonyl pathway via orsellinic acid. Sequential O-methylation, C-methylation and oxidation of the orsellinate unit, followed by esterification of two such units, yields the β -orcinol para-depside atranorin. The intermediates are chemically informative in their own right: methyl orsellinate, the methyl ester of orsellinic acid, is a direct precursor/hydrolysis fragment of the depside series and has been detected as a discrete metabolite in *P. stellaris* [14].

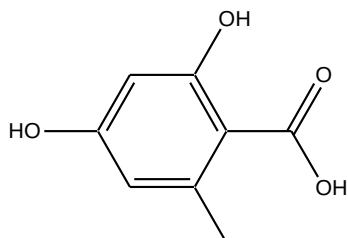


Fig. 1a. Orsellinic acid (2,4-dihydroxy-6-methylbenzoic acid)

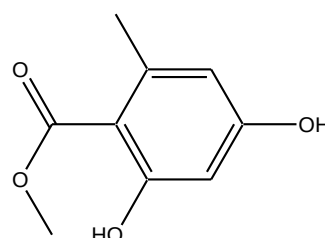


Fig. 1b. Methyl orsellinate (methyl 2,4-dihydroxy-6-methylbenzoate)

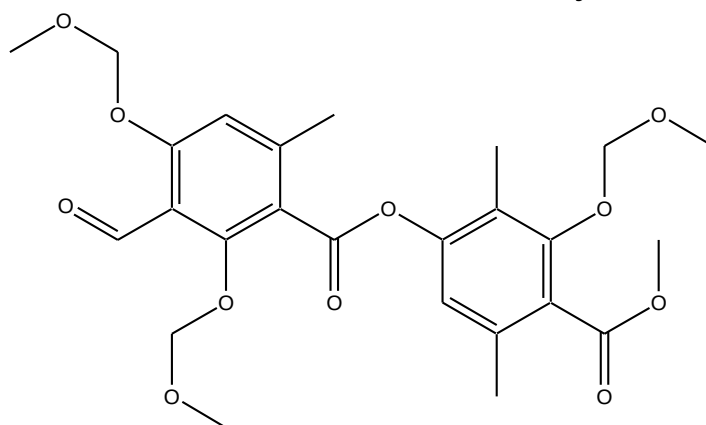


Fig. 1c. Atranorin, $C_{19}H_{18}O_8$, $M = 374.35$, CAS 479-20-9 [15].

Atranorin is deposited in the upper cortex, where it functions principally as a UV screen. A practical detection note: when atranorin is present at low concentration the K^+ reaction may be almost invisible, but the dried spot fluoresces bright yellow under long-wave UV, which markedly improves sensitivity [5].

The medullary chemistry of *Physciaceae* is dominated by C30 pentacyclic hopanoids — four fused six-membered rings (A–D) plus a cyclopentane ring E bearing a C-21 side chain. Oxygenation at C-6, C-16, C-20 and C-22, and acetylation of those hydroxyls, generate the species-specific chemosyndromes used in modern *Physcia* taxonomy [9],[10]; for the wider distribution of terpenoids and sterols across lichens see the recent review of the class [13]. The structure of leucotylin was established by Yosioka, Nakanishi and Kitagawa from *Parmelia leucotyliza* [11].

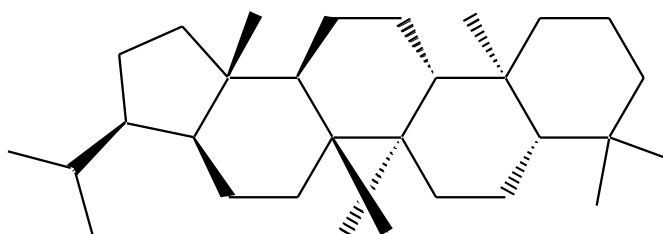


Fig. 2a. Hopane (A'-neogammacerane), $C_{30}H_{52}$, $M = 412.75$ — the parent skeleton.

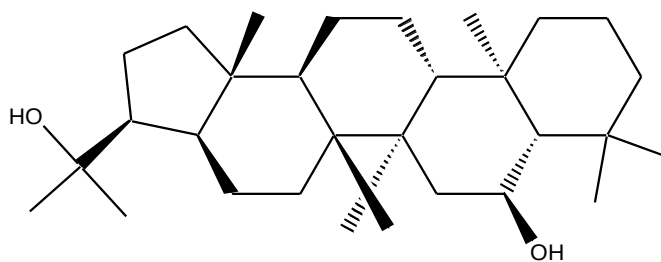


Fig. 2b. Zeorin = hopane-6 α ,22-diol, $C_{30}H_{52}O_2$, $M = 444.74$.

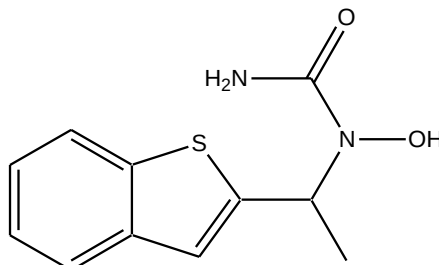


Fig. 3. Leucotylin = hopane-6 α ,16 β ,22-triol, $C_{30}H_{52}O_3$, $M = 460.74$.

Table 1.

Reference data for the metabolites discussed. Molecular formulae and masses computed from the drawn constitutions; systematic names follow the standardised lichen-substance catalogue [12].

Compound	Systematic name	Formula	Class
Orsellinic acid	2,4-Dihydroxy-6-methylbenzoic acid	$C_8H_8O_4$	Monoaromatic polyketide
Methyl orsellinate	Methyl 2,4-dihydroxy-6-methylbenzoate	$C_9H_{10}O_4$	Depside precursor
Atranorin	(3-Hydroxy-4-methoxycarbonyl-2,5-dimethylphenyl) 3-formyl-2,4-dihydroxy-6-methylbenzoate	$C_{19}H_{18}O_8$	β -Orcinol para-depside
Zeorin	Hopane-6 α ,22-diol	$C_{30}H_{52}O_2$	Hopane triterpenoid
Leucotylin	Hopane-6 α ,16 β ,22-triol	$C_{30}H_{52}O_3$	Hopane triterpenoid
20 α -Acetoxyzeorin	20 α -Acetoxyhopane-6 α ,22-diol	$C_{32}H_{54}O_4$	Acetylated hopanoid
—	6 α -Acetoxyhopane-16 β ,22-diol	$C_{32}H_{54}O_4$	Acetylated hopanoid
—	16 β -Acetoxyhopane-6 α ,22-diol	$C_{32}H_{54}O_4$	Acetylated hopanoid
Aipolic acid	6 α -Acetoxy-22-hydroxyhopane-25-oic acid	$C_{32}H_{52}O_5$	Hopanoic acid

Physcia biziana (A. Massal.) Zahlbr synonyms: *Squamaria biziana* A. Massal., Misc. Lichenol. 5 (1856). Type: Ragusa, Dalmatia (Dubrovnik, Croatia), on rock [2]; for the full

synonymy see Moberg [4]. The most detailed modern account is the Australian Physciaceae treatment [2], which reports for *P. biziana*: upper cortex K⁺ yellow, medulla K[−], containing atranorin. In the accompanying identification key the species is placed in the couplet defined by crenulate apothecial margins and the absence of triterpenes, and is separated on that basis from the morphologically similar *P. austrostellaris*, which has smooth apothecial margins and contains 20 α -acetoxypezizic acid [2]. This is consistent with the placement of the species in section *Stellares*, defined by a K[−] medulla and absent zeorin [3].

A 2023 taxonomic study reporting *P. biziana* as a new record for Pakistan (Parachinar, Kurram district, Khyber Pakhtunkhwa, moist temperate forest, on bark) recorded atranorin and zeorin by thin-layer chromatography for the specimen described, alongside a K⁺ cortical reaction [7].

Cortex: atranorin (major); K⁺ yellow; Medulla: K[−]; triterpenoids reported absent in Australian and European material [2],[3]

Regionally reported additional constituent: zeorin (Pakistan) — requires confirmation [7]. Chemically relevant ecology: an arid-zone corticolous species of open situations with a densely pruinose upper surface; the pruina (calcium oxalate) supplements atranorin as a photoprotective layer [2]

The classical European account gives cortex K⁺ yellow, medulla K[−], and distinguishes *P. stellaris* from the frequently co-occurring *P. aipolia* by its less conspicuous white maculae, its K[−] medulla and the absence of zeorin [5]. Central European and North American diagnostic resources apply the same criterion, contrasting the K[−] medulla of *P. stellaris* with the K⁺ yellow, zeorin-containing medulla of *P. aipolia* [16],[17],[23].

The New Zealand treatment, however, reports for *P. stellaris*: cortex K⁺ yellow, medulla K[−], with atranorin, leucotylin, zeorin and two unidentified compounds present [6].

A 2020 screening study examining lichen extracts across the phylogeny for effects on fungal sporulation, hyphal growth and specialised metabolite production purified methyl orsellinate (Fig. 1b) as an active compound from *P. stellaris*, using flash chromatography over silica followed by HPLC, with characterisation by LC–MS/MS [14].

Methyl orsellinate is a monoaromatic orsellinate ester — a depside precursor rather than an assembled depside — and its accumulation indicates an active orsellinic-acid polyketide pathway in the *P. stellaris* mycobiont whose products are not exhausted by atranorin. Minor phenolics of this class are routinely missed by taxonomic TLC because they migrate near the solvent front and stain weakly. The finding implies that the conventional “atranorin + triterpenes” description substantially understates the metabolite inventory of the species.

Cortex: atranorin (major); K⁺ yellow [5],[6]. Medulla: K[−]; zeorin and leucotylin reported present in New Zealand material [6], reported absent in British and Central European material [5],[17]. Additional identified constituent: methyl orsellinate [14]. Unresolved: at least two unidentified compounds reported in New Zealand material [6].

Table 2.

Comparative chemistry and diagnostic morphology. AU = Australia, EU = Europe, PK = Pakistan, NZ = New Zealand. Data from refs [2],[3],[5]–[7],[14],[17].

Character	<i>P. biziana</i>	<i>P. stellaris</i>
Section	Stellares	Stellares
Cortex, K	+ yellow (atranorin)	+ yellow (atranorin)
Medulla, K	–	–
Atranorin	Major, invariant	Major, invariant
Zeorin	Absent (AU, EU); reported (PK)	Absent (UK, C. EU); reported (NZ)
Leucotylin	Not reported	Reported (NZ)
Other phenolics	Not investigated	Methyl orsellinate
Thallus surface	Emaculate, densely pruinose	Emaculate, epruinose
Apothecial margin	Crenulate when immature	Entire to subcrenulate
Habitat	Bark, open/arid situations	Bark of deciduous trees; heliophilic, nitrophilous

The essential point is that the two species are separated morphologically rather than chemically. Both express the same core profile — cortical atranorin over a K– medulla — and are distinguished in practice by the dense pruina and initially crenulate apothecial margins of *P. biziana* against the epruinose thallus of *P. stellaris*. This makes the pair a poor subject for chemotaxonomic separation and an excellent one for the study of intraspecific chemical variation.

Neither species has been the subject of a dedicated pharmacological study. What exists is activity data for their individual constituents, obtained from other source organisms and extrapolated here.

Endolichenic microorganisms are wholly uninvestigated in these two taxa. Endolichenic fungi and bacteria are now recognised as independent sources of novel scaffolds, making both species accessible targets.

No crude-extract bioactivity data. Baseline antimicrobial, antioxidant (DPPH, FRAP, ABTS) and cytotoxicity screening of acetone and methanol extracts is a straightforward and publishable first step.

Conclusion

Physcia biziana and *P. stellaris* share a conservative and rather minimalist chemical profile: cortical atranorin as the invariant major metabolite, a K– medulla, and a variable, poorly documented trace inventory of hopane triterpenoids. The published record for both taxa rests almost entirely on spot tests and TLC performed for taxonomic rather than phytochemical purposes, and the contradictions within that record — zeorin present or absent depending on the source — remain unresolved. Given that atranorin is itself a compound of demonstrated pharmacological interest, and that at least one non-depside polyketide has

already emerged from *P. stellaris*, both species warrant a proper modern chemical investigation using HPLC–DAD, LC–MS/MS and NMR on sequence-verified material.

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