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The Oil Bodies of Liverworts: Unique and Important Organelles in Land Plants

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Oil bodies of liverworts are intracellular organelles bounded by a single unit membrane containing lipophilic globules suspended in a proteinaceous matrix. They are a prominent and highly distinctive organelle uniquely found in liverworts. Although they have been widely used in taxonomy and chemosystematics, and many of their secondary metabolites are known to be bioactive and are considered as potential sources of medicines, their origin, development and function still remain poorly understood. Recently, biochemical studies have indicated that the isoprenoid biosynthetic pathways in liverworts are similar to those of the seed plants and that oil bodies of *Marchantia polymorpha* contain a protein complex immunologically related to plastid and cytosolic enzymes of isoprenoid synthesis. Cytoplasmic lipid droplets lacking a bounding membrane have recently been recognized as important dynamic organelles playing active roles in cell physiology. Structural proteins, covering the surface of the lipid droplets and preventing them coalescing during desiccation, have been found in seed plants and also in the moss *Physcomitrella patens*. However, whether liverwort oil bodies play a dynamic role in cell metabolism, in addition to their role as sites of essential oil accumulation and sequestration, has not been formally tested. In this review, we present current knowledge on the oil bodies of liverworts on their origin and development, their role in taxonomy, chemosystematics and potential pharmaceutical applications leading to their functional significance, and we also identify avenues for future studies on this important but long-overlooked organelle.

Keywords oil body, lipophilic globule, liverwort, organelle, lipid droplet, land plant, ontogeny, homology, evolution, ecology, function

I. INTRODUCTION

Extant liverworts include approximately 8000 species and are found on all continents, occupying arid to aquatic niches. Fossil records together with evidence from phylogenetic studies

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suggest that the first land plants, which evolved approximately 470 million years ago, were liverwort-like (Kenrick and Crane, 1997; Wellman *et al.*, 2003; He-Nygrén *et al.*, 2004; Qiu *et al.*, 2006; Rubinstein *et al.*, 2010). These findings indicate that metabolic pathways essential for survival under stress conditions on land must have evolved at a very early stage. It also implies that liverworts might have played an important role in creating a more suitable environment for the subsequent evolution of other land plant groups and that they may help us to understand the vegetative and reproductive changes that favoured the successful radiation of land plants and their adaptations to life in a terrestrial environment.

Progress towards understanding liverwort origins and evolution has been made in resolving phylogenetic relationships of major groups of liverworts and identifying putative key innovations (Davis, 2004; Forrest and Crandall-Stotler, 2004; He-Nygrén *et al.*, 2004, 2006; Crandall-Stotler *et al.*, 2005; Heinrichs *et al.*, 2005; He and Glenny, 2010). These studies can well serve as a basis for further investigating adaptive strategies and exploring evolutionary mechanisms and causal factors that have led to extant diversity. One of the putative evolutionary innovations of liverworts is the presence of oil bodies in the cells of both gametophytes and sporophytes. These are intracellular organelles bounded by a single unit membrane containing lipophilic globules (called here as oil body globule, abbreviated as OB globule in the following sections) suspended in a proteinaceous matrix (Pihakaski, 1968; Duckett, 1986). The oil bodies of liverworts exhibit diverse morphological and chemical variations and have been widely used in taxonomy and chemosystematics. Numerous terpenoids as well as aromatic compounds extracted from liverwort oil bodies have been shown to be bioactive and have been suggested as potential sources of medicines (Asakawa, 1995, 2004, 2012). More than 90% of liverwort species contain oil bodies in their cells, and the phylogenetic studies (He-Nygrén *et al.*, 2004, 2006) have shown that oil bodies in some groups have been lost independently and most likely at different times during the course of evolution, although they must serve some important function since they have been preserved in most liverworts. However, since the oil bodies have been studied only for a few liverwort species, their origin and development remain poorly understood, and their evolutionary and ecological importance has long been overlooked. Hypotheses on their possible functions, such as, protection against herbivores, pathogens, cold temperatures, UV radiation and desiccation, remain mainly speculative. Recently, biochemical studies have indicated that the isoprenoid biosynthetic pathways in liverworts are similar to those of the seed plants (Nabeta *et al.*, 1994; Tazaki *et al.*, 1995; Adam *et al.*, 1998). Suire *et al.* (2000) reported that oil bodies of *Marchantia polymorpha* contain protein complex immunologically related to plastid and cytosolic enzymes of isoprenoid synthesis. However, the mechanisms of metabolic activity in liverwort oil bodies have not been investigated. While the proteins of the liverwort oil bodies are little known, the structural proteins, oleosins, found in cytoplasm lipid droplets of many vascular plant seeds, have recently been reported in the moss *Physcomitrella patens* (Huang *et al.*, 2009). These proteins cover the surface of the lipid droplets and prevent them coalescing during desiccation. Cytoplasmic lipid droplets lacking a bounding membrane appear to be ubiquitous and are found across kingdoms, although it has been suggested that liverwort oil bodies are fundamentally different in both substructure and development from lipid droplets found in all land plants or algae (Duckett, 1986), further comparative studies, looking at their origin, development, and function, would likely shed light on their evolution in liverworts and also other plant groups. In this paper, we review historical and contemporary studies on the oil bodies of liverworts on their ontogeny, their role in taxonomy, chemosystematics, potential pharmaceutical applications and their functional significance; we also identify avenues for future studies on this unique and important organelle.

II. DISCOVERY OF THE OIL BODY IN LIVERWORTS AND ITS ORIGIN

The oil bodies of liverworts were first described by Hübener (1834) in his description for leafy liverwort *Jungermannia taylorii* (now as *Mylia taylorii*) as transparent drops, with shining, membranous texture. Shortly after, Mirbel (1835) observed and illustrated what were called by him as whitish, small, and solid masses or bodies (Figure 1) scattered in the cells of *M. polymorpha*. Mirbel could not recognize the nature of these bodies, but he suspected they were starch. Gottsche (1843) also called attention on these remarkable structures naming them Zellen Körper (cell bodies) in his study on *Haplomitrium hookeri*. Von Holle (1857) changed the name to cell vesicles. In 1874, Pfeffer designated them as Ölköper (oil bodies) mainly on the account of their fatty nature. A genuine membrane surrounding the oil bodies, in which OB globules of various sizes are embedded in a medium of high refractive index, was recognized by some early workers (von Holle, 1857; Pfeffer, 1874; Müller, 1939). Galatis *et al.* (1978b) suggested that the membrane may have a particular synthesizing activity from the ultrastructure evidence of *M. palaecea*. It was suggested that protein might be present in oil bodies (Wakker, 1888; Garjeanne, 1903; Pfeffer, 1904; Pihakaski, 1966, 1968). Pihakaski (1972a, b) confirmed this with light and electron microscope studies, further stating that oil bodies are the main site of lipids in the cells of liverworts. Suire *et al.* (2000) identified that the oil body is the site of intracellular isoprenoid synthesis in liverworts, supporting the hypothesis that the oil body is an active cell compartment (Galatis *et al.*, 1978b).

Opinions on the origin of liverwort oil bodies from the chloroplast (Wakker, 1888; Kozlowski, 1921), the mitochondrion (Bergdolt, 1926), or the vacuole (Pfeffer, 1874; Garjeanne, 1903; Dombary, 1926; Zirkle, 1932) were presented respectively based on studies at light microscopy level. Later ultrastructural evidence largely supports the hypothesis that oil bodies originate from the dilation of endoplasmic reticulum cisternae

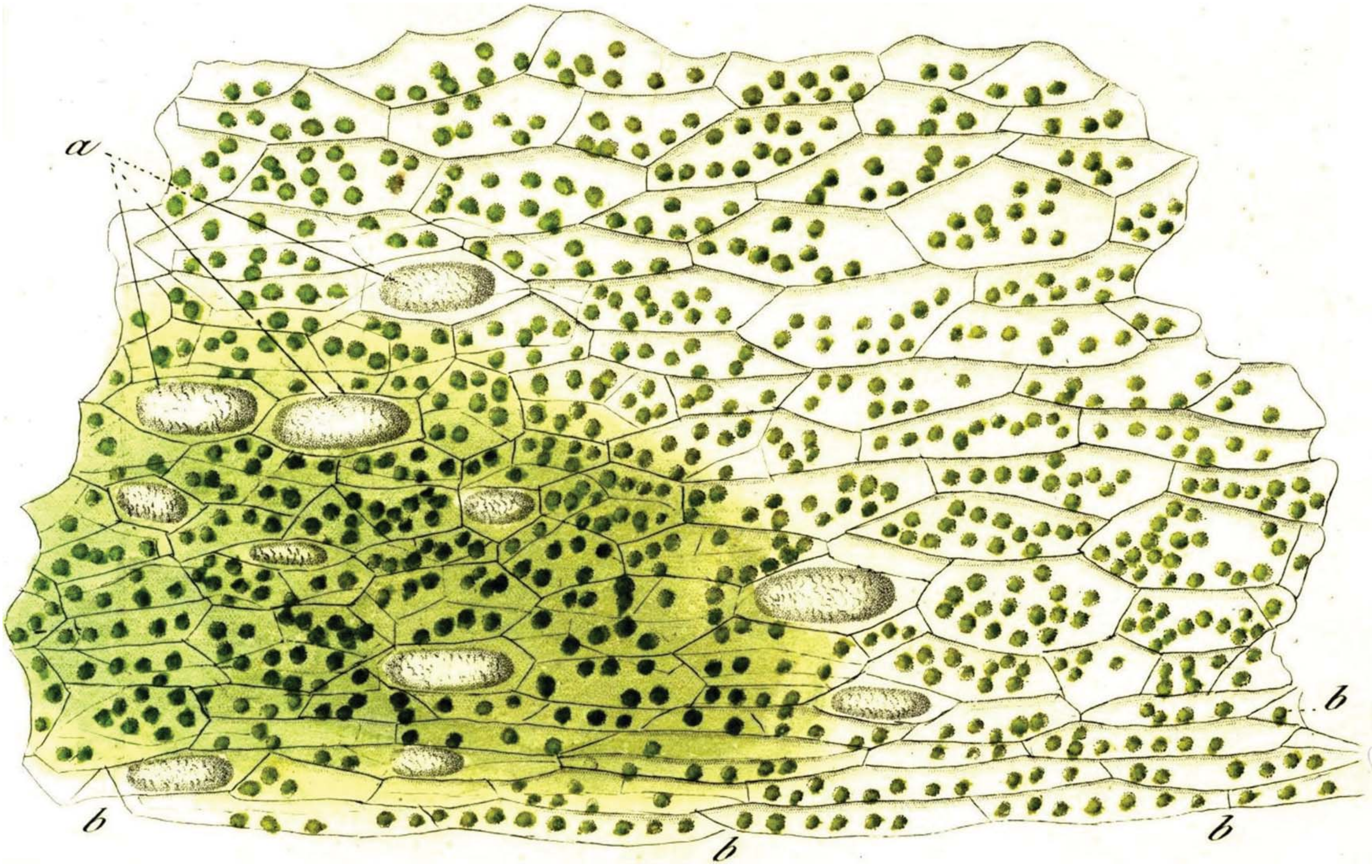

FIG. 1. Illustration of oil bodies in *Marchantia polymorpha*, a. oil bodies, b. chloroplasts (reproduced from Mirbel 1835) (color figure available online).

(Carbonnier and Gavaudan, 1960; Suire, 1970, 2000; Kronestedt, 1983; Duckett and Renzaglia, 1987; Duckett and Ligrone, 1995; Figure 2). However, hypotheses on the contribution of organelles to oil body formation are substantially different. In her studies of a few leafy liverwort species, Pihakaski (1968) suggested that in *Bazzania trilobata*, oil bodies are formed from vacuole-like structures into which the ER or Golgi apparatus secretes substances as small droplets, whereas in *Lophozia ventricosa* they are developed from fusion of cytoplasmic lipid droplets and the cytoplasm surrounding the OB globules differentiates into the matrix and a limiting membrane of the oil bodies. In contrast, Suire (1970) working with *Radula complanata* concluded that the oil bodies originate from dilated ER cisternae. This conclusion is supported with the study on *Odontoschisma denudatum* and species of *Calypogeia*, *Diplophyllum, and Scapania* by Duckett and Ligrone (1995), who rejected the hypothesis of Pihakaski (1966, 1968, 1972a, b) with an argument that the hypothesis of oil body origin from fusion of cytoplasm lipid droplets with vacuole-like structures was probably based on poorly-fixed material. Compared with the leafy liverworts, the origins of the oil bodies in complex thalloid liverworts have been shown to be markedly different. In *M. paleacea* the oil bodies were suggested to originate from

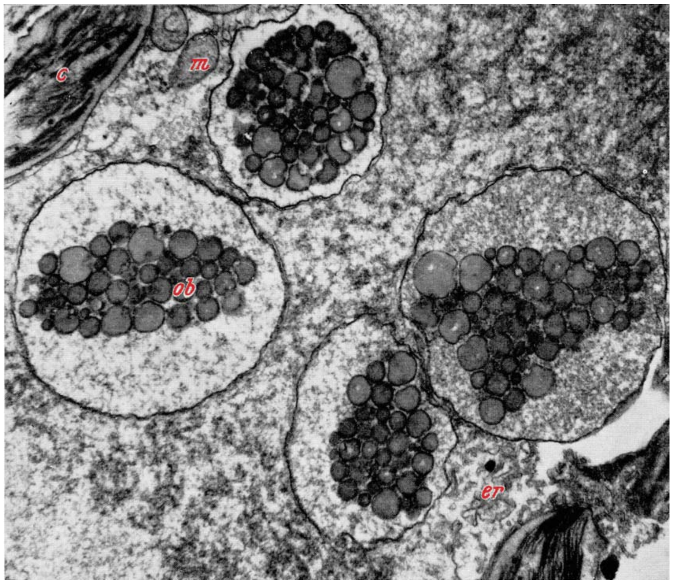

FIG. 2. Transmission electron micrograph of mature oil bodies of *Lophozia ventricosa*. c. chloroplast; er. endoplasmic reticulum; m. mitochondrion; ob. oil body with lipophilic globules enclosed. 11,000 x (reproduced from Pihakaski 1966) (color figure available online).

fusion of Golgi-derived vesicles and dilation of the ER membrane (Galatis and Apostolakos 1976; Galatis *et al.* 1978a, b). Microtubules forming a dense framework around the growing oil bodies were also reported by the authors. Furthermore, the authors underlined that the OB globules are exclusively synthesized within the oil body, while cytoplasmic lipid droplets that are free in the cytoplasm never enter the oil body. This mode of oil body development was also observed in other complex thalloid liverworts *Lunularia cruciata* and *M. polymorpha* by the same authors (Galatis *et al.*, 1978a). However, subsequent studies on complex thalloid *Riella* (Lehmann and Jaster, 1981; Lehmann and Schultz, 1982), simple thalloid Metzgeriales (Duckett and Renzaglia, 1987), and the re-examination of *L. cruciata* and *M. polymorpha* together with other fourteen species (Suire, 2000), showed no evidence for any contribution of Golgi vesicles and microtubules to oil body development. Suire (2000) concludes that "oil bodies originate from the same cell membrane system and basically develop in the same way in all the Hepaticae," and considers that *M. paleacea* presents a unique variation in the ontogenic process of the oil bodies.

Until the present day, fine structure investigations on the formation and development of oil bodies have been limited to only a few taxa, nearly all of which have relatively large oil bodies. Methods used for tissue fixation in early studies were usually insufficient and unsatisfactory compared with present day standards. The marked differences in the hypotheses for the formation of the oil bodies seen in previous studies suggest that further studies with a wider range of sampling are required. In particular, the sampling should include species lacking oil bodies such as *Blasia pusilla* and *Anthelia julacea*, and also species having oil bodies that difficult to distinguish from cytoplasmic lipid droplets under a light microscope. Examples are genera *Cephalozia, Drucella*, *Kurzia*, and *Metzgeria*. Comparative studies of oil bodies are likely to provide new insights into their origin and development, with comprehensible implications for taxonomy and systematics.

III. TAXONOMY AND PHYLOGENY

The oil bodies of liverworts occur not only in the gametophytes but also in the sporophytes including seta and spores (Müller, 1939; Suire, 1966, 1970; Pihakaski, 1972a, b). Their size ranges from less than 1 μm to nearly 30 μm in diameter, and they are often the most conspicuous feature of the cell. The oil bodies are best observed and may only be correctly circumscribed from living material. Upon unnaturally fast rates of drying, they disintegrate and eventually become invisible—this may even occur under the strong beam of a light microscope. In natural conditions, they seem to retain their original state even after prolonged periods of drought (Pressel *et al.*, 2009). Seen through a light microscope, oil bodies are usually easily recognizable by their high refractive index and they appear to be colorless, or various densities and tints of brown (such as *Calypogeia* and most Acrobolbaceae), and homogeneous or segmented. The oil bodies occur in all mature cells of the gametophyte and sporophyte in the Jungermanniopsida and Haplomitriales, whereas they usually are restricted to specialized idioblasts in Marchantiopsida and Treubiales. Müller (1939) was the first to suggest a system of classifying the oil bodies into different groups. He recognized nine types of oil bodies according to their appearance, size and form. Schuster (1966) adapted Müller's classification with some modification and he emphasized that the cells containing a single oil body (called an ocellus) in the complex thalloids except *Monoclea*, never bear chloroplasts, in contrast to the leafy and simple thalloid group in which chloroplasts are always present in the cells along with the oil bodies. However, ultrastructure evidence (Galatis *et al.*, 1978b, Kronestedt, 1983; Suire, 2000) showed that differentiating ocelli bear chloroplasts like any other cells. Cytoplasmic structures including chloroplasts are pushed to the periphery of the ocellus by the increasing volume of the oil body, which may become difficult to observe under a light microscope. In subsequent taxonomic studies, different schemes of oil body types have been proposed (Gradstein *et al.*, 1977; Schuster, 1992; Kis and Pócs, 1997; Zhu and So, 2001). For example, Gradstein *et al.* (1977) proposed four types of oil bodies, dividing homogeneous oil bodies into *Massula*-type and *Bazzania*-type, and segmented ones into *Jungermannia*-type and *Calypogeia*-type (Figure 3). Nevertheless, the shape, number, size, color and distribution of oil bodies vary considerably and they have been recognized as taxonomically important. For example, the oil body types were used as a primary criterion to separate genera of Ptychanthoideae of the family Lejeuneaceae (Schuster and Hattori, 1954). Distinguishing taxa at species and genus levels using oil body characters have also been widely used for leafy and simple thalloid liverworts, especially in families Lejeuneaceae, Frullaniaceae and Radulaceae. For instance, the separation of *Dendrolejeunea* from *Thysananthus* was supported by oil body chemical evidence (Sukkharak *et al.*, 2011). Taxa that lack oil bodies are present in various families, including Blasiaceae, Metzgeriaceae, Cephaloziaceae, Lepidoziaceae, and Antheliaceae. The cause for the loss of the oil bodies is not known.

So far, taxonomic characterization of oil bodies of liverworts has always been done at light microscopy level; the distinction between an oil body and a cytoplasmic lipid droplet has been questioned when oil bodies are so small that the bounding membrane is difficult to observe. Transitions between homogeneous and segmented or granular types of oil bodies are also present in some families, such as Haplomitriaceae and Lejeuneaceae (Schuster, 1992). Schuster (1992) hypothesed that the plesiomorphic condition in liverworts is that all cells contain a variable number of small oil bodies, similar in size to the chloroplasts or smaller. In a phylogenetic context, the occurrence of unstructured oil bodies in the leafy and simple thalloid groups (such as *Lembidium* of Lepidoziaceae, *Rectolejeunea* of Lejeuneaceae, and *Metzgeria* of Metzgeriaceae), seem to be in a derivative position compared with their allied groups in which structural oil bodies are found (He-Nygrén *et al.*, 2006; Wilson

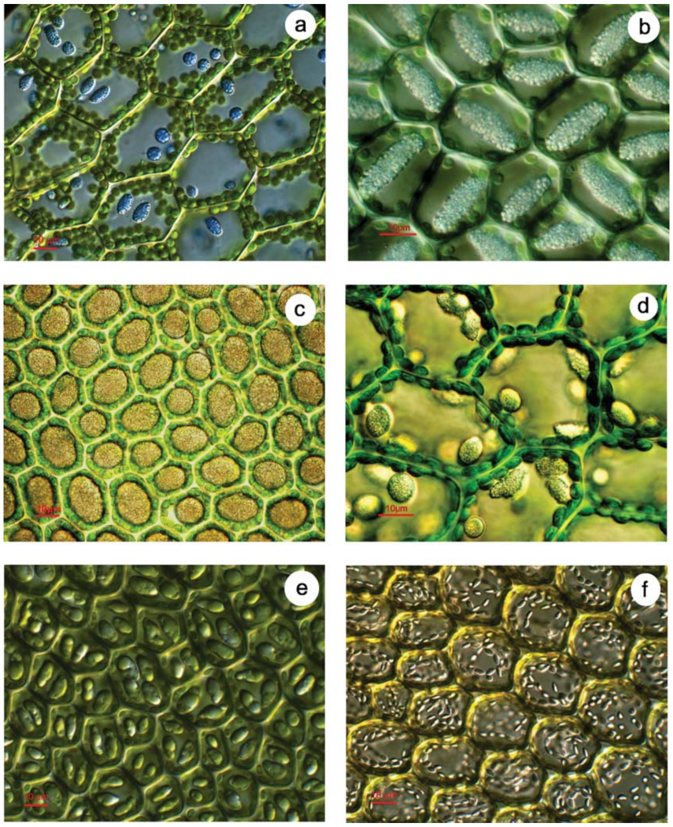

FIG. 3. Different types of oil bodies in the cells of liverworts: a–b, *Calypogeia*-type; c–d, *Jungermannia*-type; e, *Bazzania*-type; f, *Massula*-type—a. *Calypogeia azurea*, b. *Cheilolejeunea anthocarpa*, c. *Radula constricta*, d. *Solenostoma truncatum*, e. *Bazzania tridens*, f. *Trocholejeunea sandvicensis* (color figure available online).

et al., 2007; Cooper *et al.*, 2011). The ancestral type of the oil bodies is ambiguous because the earliest diverging Haplomitriopsida including *Treubia* and *Haplomitrium*, presents two different types of oil bodies, as mentioned above. Tremblay (2010) suggests that the dark, scattered cells found in Paleozoic liverwort fossils are homologous with the oil body cells of extant taxa, providing evidence for specialized oil body cells as the plesiomorphic state in liverworts. However, in this short account on oil body fossils, no detailed data and actual analyses were presented by the author. Further phylogenetic studies addressing the evolution of oil bodies are needed.

IV. CHEMOSYSTEMATICS AND PHARMACEUTICAL APPLICATION

The chemical nature of oil bodies has been one of the interests of many authors as many liverworts present fragrance, especially when crushed. Gottsche (1843) and von Holle (1857) considered that oil bodies contain a fragrant resin or the mixture of ethereal oil and a resin. Lindberg (1888) and Pfeffer (1874) assumed that they contain fatty oil and protein substance. Lohmann (1903) concluded that the content of oil bodies was terpenes, and Müller (1905) provided evidence that the terpenes were monoterpenes, either sesquiterpenes or the corresponding alcohols. However, the chemical constituents of liverworts were not investigated until 1950s mostly because of the difficulty of obtaining a large quantity of plant material for chemical analysis (Fujita *et al.*, 1956; Huneck, 1967; Huneck and Klein, 1967; Benešova and Herout, 1970; Matsuo *et al.*, 1971 a, b). Isolating pure compounds and determining chemical structures with small amount of material later became possible and easier due to gas chromatography, gas chromatography-mass spectrometry and other analytical methods. So far, about 10% of the more than 700 lipophilic compounds extracted from liverworts have been characterized. Most of the compounds found in liverworts are composed of mono-, sesqui- and diterpenoids and aromatic compounds, such as typical bibenzyls and bisbibenzyls (Asakawa, 2004, 2012). Terpenoids represent the oldest group of small molecular products synthesized by plants. Sesquiterpenoids are the largest class of secondary metabolites in liverworts. Some are also found in vascular plants, while others are unique to liverworts. Diterpenoids are the second largest class of secondary metabolites in liverworts. Pinguisane-type sesquiterpenes and sacculatane-type diterpenoids are only found in liverworts (Figure 4). For example, pinguisane-type sesquiterpenes are significant chemical constituents of *Porella* species (Asakawa *et al.*, 1981a, b; Asakawa, 1982, 1995, 2001; Hashimoto *et al.*, 2000). Sacculatane diterpenoids have been detected in *Pellia*, *Pallavicinia*, *Fossombronia*, *Trichocoleopsis,* and *Porella* (Asakawa, 2004). Many of the sesqui- and diterpenoids are enantiomers of those found in higher plants (Asakawa, 2012). Liverworts have fewer monoterpenes than sesqui- and diterpenoids and according to Asakawa (1995), they

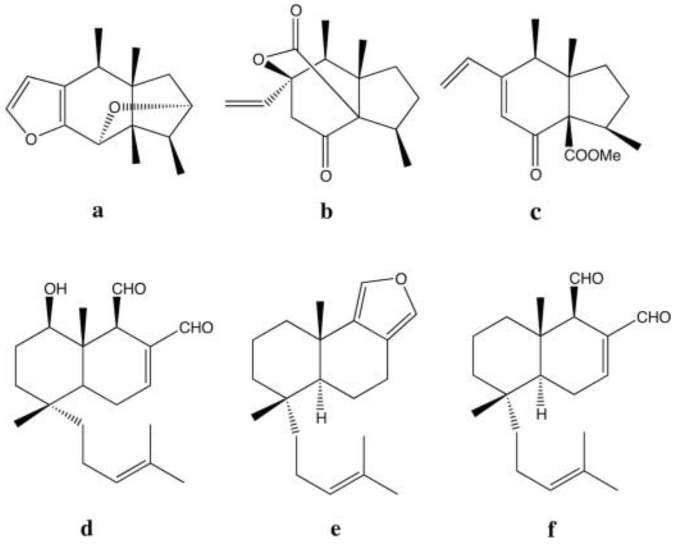

FIG. 4. Unique chemical compounds of pinguisane-type sesquiterpenes (a–c) and sacculatane-type diterpenoids in liverworts (d–f): a. *Porella platyphylla*, b–c. *Porella acutifolia* subsp. *tosana*, d–e. *Pellia endiviifolia*, f. *Trichocoleopsis sacculata*. Data sources include Asakawa 2001, 2004 and Asakawa *et al.* 2012.

are mainly responsible for the characteristic odour of liverworts. Together with sesquiterpenes, monoterpenes are the main constituents of essential oils. No terpenoids are found in *B. pusilla*, which lacks oil bodies (Asakawa, 1988; Wurzel and Becker, 1989). But terpenoids and aromatic compounds have been found in *A. julacea*, which bears no oil bodies (Asakawa *et al.*, 1980a, b). Unsaturated fatty acids, triglycerides and sterols which are elaborated in high amount in mosses, have also commonly been detected in liverworts. In mosses, terpenoids have rarely been detected.

Terpenoids and lipophilic aromatic compounds have been explored extensively as chemosystematic indicators (Asakawa *et al.*, 1979a, b, c, 1980a, b, 1981a, b; Asakawa, 2004). Asakawa (2004) discussed at length the importance of chemosystematics of liverworts in 36 families at different taxonomic levels. For example, the specific terpenoids and bis-bibenzyls, such as pinguisane sesquiterpenoids, which are the characteristic markers of the leafy liverworts, are also found in simple thalloid *Aneura* species (Asakawa, 1995). Asakawa (1995) considers that this indicates the close relationship of the two groups. The sister relationship of the leafy liverworts and the group consisting of Aneuraceae and Metzgeriaceae has been confirmed by recent molecular systematic studies (He-Nygrén *et al.*, 2006). Chemical differences have been used to distinguish family Frullaniaceae from Jubulaceae, and Lepidolaenaceae from Trichocoleaceae (Asakawa, 2004). For a detailed account of the chemosystematics of liverworts, readers are referred to the review paper by Asakawa (2004).

In liverworts, many terpenoids and lipophilic aromatic compounds have been shown to possess remarkable biological activities (Asakawa, 2012). Among others, these include cytotoxicity, anti-HIV inhibitory, antimicrobial, insect antifeedant and mortality, nematocidal activity, anti-obesity, anti-influenza activity, allergenic contact dermatitis, and vasorelaxant effects (Asakawa, 2012). Since *M. polymorpha* had long been used as traditional medicine to treat liver ailments and to reduce inflammation in China, biological investigation and pharmacological testing have focused on the complex thalloid liverworts. The well-known compounds from *Marchantia* species, marchantin A-C, bisbibenzyl ethers have been demonstrated to possess significant biological activities, such as antibacterial (Kámory *et al.*, 1995), antifungal (Niu *et al.*, 2006), anticytotoxicity (Asakawa *et al.*, 2000) and anti-cancer activity—in the latter case by triggering apoptosis of tumor cells (Shi *et al.*, 2007; Huang *et al.*, 2010). The marchantins have also been revealed to have anti-influenza activity against both influenza A and B viruses (Iwai *et al.*, 2001). Xue *et al.* (2012) showed that riccardin D, a macrocyclic bisbibenzyl isolated from *Dumortiera hirsuta*, is a novel DNA topo II inhibitor which can induce apoptosis of human leukemia cells. Thus, it has been considered as a potential chemotherapeutic agent for treatment of cancers. Sun *et al.* (2009) demonstrated that the macrocyclic bisbibenzyl dihydroptychantol A (DHA), isolated from *Asterella angusta*, showed significant multidrug resistance (MDR) reverting activity in chemo-resistant cancer cells. Within the leafy liverworts, Bukvicki *et al.* (2012) investigated chemical constituents of *Porella cordaeana* and their results demonstrated that *P. cordaeana* extracts possess significant antimicrobial potential against food micro-organisms and that high percentages of monoterpene and sesquiterpene hydrocarbons found in the species could be responsible for the strong antimicrobial activity. Guo *et al.* (2008) demonstrated that the chemical compounds of *Scapania verrucosa* have antimicrobial and antitumor activities. Burgess *et al.* (2000) showed that extracts of *Bazzania novae-zelandiae* are cytotoxic, with selective activity against human tumor cell lines. Scher *et al.* (2002) found that Glaucescenolide 1 was the most cytotoxic compound isolated from *Schistochila glaucescens* against P388 leukemia cells. Nevertheless, liverwort bioactive compounds are increasingly seen as being potentially useful for the production of new drugs. The ability of liverwort oil bodies to undergo dynamic metabolic fluxes of their matrix constituents may provide a competitive evolutionary advantage, as much of the tissue of liverworts is only one cell thick, grows closely to the substrate and has no mechanical protection such as bark or cuticle. This ability may have been a very early prerequisite for survival on land of the earliest land plants.

V. FUNCTION

The possible functions of liverwort oil bodies suggested have been protection from herbivores (Stahl, 1888), pathogens, cold temperatures, excessive light (Hieronymus, 1892), and UV radiation and desiccation (Gavaudan, 1927; Chalaud, 1931). These suggestions made very early on are still been stressed today, although cases that are against these assumptions can all be found. For instance, *Anthelia* usually grows in sites with intensive light and sometimes in very arid conditions, but it does not bear oil bodies; xeric *Riccia* species don't bear oil bodies. Müller (1939) suggested that the oil bodies may have had a physiological function in the past that no longer is expressed today. Pihakaski (1968) assumed that oil bodies have a metabolic function based on their structure and the close connection with the endoplasmic reticulum. As yet effort on testing these assumptions directly or indirectly has been limited. Recently, Pressel *et al.* (2009) investigated physiological and cytological changes associated with dehydration and rehydration in six liverwort species and they found that the oil bodies of *Southbya nigrella* retained their original volume during desiccation. According to the authors, the oil bodies collapse during rehydration only to regain their original volume some 48 h after rewetting. It is suggested that the collapse during rehydration indicates a shifting of carbohydrates into the cytosol. Therefore, these authors assume that the oil bodies contain highly concentrated soluble carbohydrate or other moieties whose osmoticum resists water loss and that the oil bodies may have a crucial role in desiccation tolerance.

Fabón *et al.* (2012) demonstrated that *Jungermannia exsertifolia* subsp. *cordifolia* showed a dynamic protection and acclimation capacity to the irradiance level and spectral characteristics of the radiation received by studying the global amount of UV-absorbing compounds (as the bulk UV absorbance of methanolic extracts). Arróniz-Crespo *et al.* (2004) based on their study of bryophytes collected in Spain, and Otero *et al.* (2008) based on their Argentina collections, have drawn the same conclusion that the accumulation of UV-absorbing compounds might often increase protection against UV radiation in liverworts, but rarely in mosses. Martínez-Abaigar and Núñez-Olivera (2011) concluded that liverworts seem to have higher amounts of both constitutive and inducible UV-absorbing compounds than mosses. It has been suggested that the accumulation can reduce UV penetration and damage to potential targets (Otero *et al.*, 2008). Thus, the efficient accumulation of UV-absorbing compounds in liverworts could have been one of the factors favoring their success in the colonization of land. UV fluxes were higher during the earlier phases of evolution of land plants, due to the thinner ozone layer (Cockwell and Knowland, 1999), therefore the development of UV-screening compounds might have played a major role in the evolution of plants. However, whether the oil bodies of liverworts have UV-absorbing compounds, has not been investigated.

Whether the oil bodies of liverworts act as an energy storage organelle is not well known. This was discussed in the past but the authors all denied the possibility that the oil bodies could store excessive nutrients. The ground for this was that oil bodies were not detected in the sporophytes (Zwickel, 1932) or that they were found in the cells of sporophytes but there were no chloroplasts in those cells (Pfeffer, 1874; von Küster, 1894; Stewart, 1978). Both oil bodies and photosynthetic activities have been detected in liverwort sporophytes in subsequent studies. Stewart (1978) considered that terpenes, which occur in many plants, do not reserve food.

Cytoplasmic lipid droplets are particularly conspicuous in storage cells in vascular plants. For example, most vascular plant seeds store lipid droplets as a food reserve for germination and post-germinative growth (Huang *et al.*, 2009). Cytoplasmic lipid droplets have recently been recognized as important dynamic organelles playing active roles in cell physiology (Beller *et al.*, 2010; Chapman *et al.*, 2012; Fujimoto and Parton, 2011). However, whether oil bodies of liverworts play a dynamic role in cell metabolism, in addition to their role as sites of essential oil accumulation and sequestration, has not been formally tested. The current hypothesis for lipid droplet biogenesis is that the organelles are derived from the ER. Lipid droplets consist of a hydrophobic core of stored fat, mostly neutral lipids such as triacylglycerols but also sterols and sterol esters, which are surrounded by a protein-bearing phospholipid hemimembrane (Farese and Walther, 2009; Beller *et al.*, 2010; Digel *et al.*, 2010). Recently, major proteins that associate with lipid droplets in some seed plant and algal groups have become known (Murphy, 2001; Abell *et al.*, 2002; Li *et al.*, 2002; Nguyen *et al.*, 2011; Huang *et al.*, 2009; Vieler *et al.*, 2012). Structural proteins, oleosins, have been found in seed plants and also in the moss *Physcomitrella patens*. These proteins cover the surfaces of lipid droplets and prevent them coalescing during desiccation (Siloto *et al.*, 2006; Shimada *et al.*, 2008; Huang *et al.*, 2009). Oleosins are not present in green algae, although oleosin-like proteins with functions analogous to plant oleosins have been identified (Moellering and Benning, 2010; Peled *et al.*, 2011; Vieler *et al.*, 2012). The proteins of liverwort oil bodies are little known.

VI. FUTURE DIRECTIONS AND PERSPECTIVES

Without doubt, oil bodies are one of most important organelles in liverworts. It is regrettable that while our understanding of lipid droplets in other plant groups, fungi and animals is rapidly evolving, fundamental questions pertaining to the evolution and function of oil bodies in liverworts still remain unanswered. These include: (1) Are the OB globules of the oil bodies of liverworts and the lipid droplets of other plant groups homologous and how are the OB globules and cytoplasmic lipid droplets related within the liverworts? (2) Do oleosin or oleosin-like proteins occur in liverwort oil bodies? (3) Have the oil bodies of liverworts followed a completely different evolutionary pathway by evolving a single unit membrane surrounding the OB globules instead of a structural protein? (4) What is the composition of the liverwort oil body membrane? (5) What are the mechanisms of metabolic activity in the oil bodies of liverworts? It will be challenging to deal with such an organelle with largely unknown morphology, development, function, and effects of many of its secondary compounds. One way forward may be to conduct proteomic studies in order to characterize the major proteins of the oil bodies and also identify the proteins that are involved in metabolism. Novel tools for visualizing and exploring oil body origin, development and dynamics, and enhancements of imaging techniques such as high resolution electron microscopy, would likely accelerate the deciphering of the biology and evolution of the oil bodies of the liverworts. We believe that the results would provide important information about the mechanisms for the origin of compartmentalization in liverwort oil bodies and would eventually fill the gap in the understanding of lipid droplet evolution in green plants. Furthermore, the elucidation of liverwort oil body metabolism will provide essential knowledge for potential pharmaceutical applications.

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REFERENCES

Abell, B. M., High, S., and Moloney, M. M. 2002. Membrane protein topology of oleosin is constrained by its long hydrophobic domain. *J. Biol. Chem.* **277**: 8602–8610.

Adam, K. P., Thiel, R., Zapp, J., and Becker, H. 1998. Involvement of the mevalonic acid pathway and the glyceraldehydepyruvate pathway in terpenoid biosynthesis of the liverworts *Ricciocarpos natans* and *Conocephalum conicum*. *Arch. Biochem. Biophys.* **353**: 181–187.

Arróniz-Crespo, M., Núñez-Olivera, E., Martínez-Abaigar, J., and Tomás, R. 2004. A survey of the distribution of UV-absorbing compounds in aquatic bryophytes from a mountain stream. *The Bryologist.* **107**: 202–208.

Asakawa, Y., Hattori, S., Mizutani, M., Tokunaga, N., and Takemoto, T. 1979c. Chemosystematics of bryophytes. III. Terpenoids of the primitive Hepaticae, *Takakia* and *Haplomitrium*. *J. Hattori Bot. Lab.* **46**: 77–90.

Asakawa, Y., Ludwiczuk, A., and Nagashima, F. 2012. Phytochemical and biological studies of bryophytes. *Phytochemistry*: http://dx.doi.org/10.1016/j.phytochem.2012.04.012.

Asakawa, Y., Matsuda, R., Takemoto, T., Hattori, S., Mizutani, M., Inoue, H., Suire, C., and Huneck, S. 1981a. Chemosystematics of bryophytes. VII. The distribution of terpenoids and aromatic compounds in some European and Japanese Hepaticae. *J. Hattori Bot. Lab.* **50**: 107–122.

Asakawa, Y., Matsuda, R., Toyota, T., Suire, C., Takemoto, T., Inoue, H., Hattori, S., and Mizutani, M. 1981b. Chemosystematics of bryophytes. VIII. The distribution of terpenoids and aromatic compounds in Japanese Hepaticae and Anthocerotae. *J. Hattori Bot. Lab.* **50**: 165–177.

Asakawa, Y., Suire, C., Toyota, M., Tokunaga, N., Takemoto, T., Hattori, S., and Mizutani, M. 1980b. Chemosystematics of bryophytes. V. The distribution of terpenoids and aromatic compounds in European and Japanese Hepaticae. *J. Hattori Bot. Lab.* **48**: 285–303.

Asakawa, Y., Tokunaga, N., Takemoto, T., Hattori, S., Mizutani, M., and Suire, C. 1980a. Chemosystematics of bryophytes. IV. The distribution of terpenoids and aromatic compounds in Hepaticae and Anthocerotae. *J. Hattori Bot. Lab.* **47**: 153–164.

Asakawa, Y., Tokunaga, N., Toyota, M., Takemoto, T., Hattori, S., Mizutani, M., and Suire, C. 1979b. Chemosystematics of bryophytes. II. The distribution of terpenoids in Hepaticae and Anthocerotae. *J. Hattori Bot. Lab.* **46**: 67–76.

Asakawa, Y., Tokunaga, N., Toyota, M., Takemoto, T., and Suire, C. 1979a. Chemosystematics of bryophytes. I. The distribution of terpenoid of bryophytes. *J. Hattori Bot. Lab.* **45**: 395–407.

Asakawa, Y., Toyota, M., Tori, M., and Hasimoto, T. 2000. Chemical structures of macrocyclic bis(bibenzyls) isolated from liverworts (Hepaticae). *Spectroscopy* **14**: 149–175.

Asakawa, Y. 1982. Terpenoids and aromatic compounds as chemosystematic indicators in Hepaticae and Anthocerotae. *J. Hattori Bot. Lab.* **53**: 283–293.

Asakawa, Y. 1988. Chemical evolution of mono- and sesquiterpenoids of liverworts. *J. Hattori Bot. Lab.* **64**: 97–108.

Asakawa, Y. 1995. Chemical constituents of the bryophytes. **In:** Herz, W., Kirby, W. B., Moore, R. E., Steglich, W., and Tamm, C. (Eds.) *Progress in the Chemistry of Organic Natural Products*. Springer, Wien, NewYork.

Asakawa, Y. 2001. Recent advances in phytochemistry of bryophytes-acetogenins, terpenoids and bis (bibenzyl)s from selected Japanese, Taiwanesis, New Zealand, Argentinen and European liverworts. *Phytochemistry* **56**: 297–312.

Asakawa, Y. 2004. Chemosystematics of the Hepaticae. *Phytochemistry* **65**: 623–669.

Asakawa, Y. 2012. Liverworts—Potential Source of Medicinal Compounds. *Med. Arom. Pl.* **1**: e114. doi:10.4172/map.1000e114.

Beller, M., Thiel, K., Thul, P. J., and Jackle, H. 2010. Lipid droplets: a dynamic organelle moves into focus. *FEBS Lett.* **584**: 2176–2182.

Benešova, V. and Herout, V. 1970. Plant subsances. XXXII. Isolation and the structure of pellepiphyllin, 2-hydroxy-3-methoxy-4'-methoxydihydrostibene from liverwort *Pellia epiphylla* (L.) Dum. *Coll. Czech. Chem. Commun.* **35**: 1926–1929.

Bergdolt, E. 1926. Untersuchungen über Marchantiaceen. *Goebel's Bot. Abh.* **10**: 1–86

Bukvicki, D., Gottardi, D., Veljic, M., Marin, P. D., Vannini, L., and Guerzoni, M. E. 2012. Identification of volatile components of liverwort (*Porella cordaeana*) extracts using GC/MS-SPME and their antimicrobial activity. *Molecules* **17**: 6982–6995.

Burgess, E. J., Larsen, L., and Perry, N. B. 2000. A cytotoxic sesquiterpene caffeate from the liverwort *Bazzania novae-zelandiae*. *J. Nat. Prod.* **63**: 537–539.

Carbonnier, M. L. and Gavaudan, P. 1960. Le problem des éléoplastes de la cellule des Hépatiques. *C. R. Acad. Sci.* **154**: 1597–1601.

Chalaud, G. 1931. Germination des spores et formation du gametophyte chez *Lophocolea cuspidata* et *Chiloscyphus polyanthus*. *Ann. Bryol.* **4**: 49–78.

Chapman, K. D., Dyer, J. M., and Mullen, R. T. 2012. Biogenesis and functions of lipid droplets in plants. *J. Lipid Res.* **53**: 215–226.

Cockwell, C. S. and Knowland, J. 1999. Ultraviolet radiation screening compounds. *Biol. Rev.* **74**: 311–345.

Cooper, E. D., Shaw, A. J., Shaw, B., Henwood, M. J., Heslewood, M. M., and Brown, E. A. 2011. A multi-locus molecular phylogeny of the Lepidoziaceae: laying the foundations for a stable classification. *Mol. Phyl. Evol.* **59**: 489–509.

Crandall-Stotler, B. J., Forrest, L. L., and Stotler, R. E. 2005. Evolutionary trends in the simple thalloid liverworts (Marchantiophyta, Jungermanniopsida subclass Metzgeriidae). *Taxon* **54**: 299–316.

Davis, E. C. 2004. A molecular phylogeny of leafy liverworts (Jungermanniidae: Marchantiophyta). *Monogr. Syst. Bot. Mo. Bot. Gard.* **98**: 61–86.

Digel, M., Ehehalt, R., and Füllekrug, J. 2010. Lipid droplets lighting up: Insights from live microscopy. *FEBS Lett.* **584**: 2168–2175.

Dombray, P. 1926. Contrib. à l'étude des corps oléiformes des hépatiques des environs de Nancy. Thèse, Paris.

Duckett, J. G. and Ligrone, R. 1995. The formation of catenate foliar gemmae and the origin of oil bodies in the liverwort *Odontoschisma denudatum* (Mart.) Dum. (Jungermanniales): a light and electron microscope study. *Ann. Bot.* **76**: 405–419.

Duckett, J. G. and Renzaglia, K. S. 1987. The ultrastructure of oil body development in Aneuraceae: games with cytochemistry and systematics. *Bull. Brit. Bryol. Soc.* **49**: 17–19.

Duckett, J. G. 1986. Ultrastructure in bryophyte systematics and evolution: an evaluation. *J. Bryol.* **14**: 25–42.

Fabón, G., Monforte, L., Tomás-Las-Heras, R., Núñez-Olivera, E., and Martínez-Abaigar, J. 2012. Dynamic response of UV-absorbing compounds, quantum yield and the xanthophyll cycle to diel changes in UV-B and photosynthetic radiations in an aquatic liverwort. *J. Pl. Physiol.* **169**: 20–26.

Farese, R. V. and Walther, T. C. 2009. Lipid Droplets Finally Get a Little R-E-S-P-E-C-T. *Cell* **139**: 855–860.

Forrest, L. L. and Crandall-Stotler, B. J. 2004. A phylogeny of the simple thalloid liverworts (Jungermanniopsida, subclass Metzgeriales) as inferred from the chloroplast genome. *Monogr. Syst. Bot. Mo. Bot. Gard.* **98**: 119–140.

Fujimoto, T. and Parton, R. G. 2011. Not just fat: the structure and function of the lipid droplet. *Cold Spring Harb. Perspect. Biol.* **3**: a004838.

Fujita, Y., Ueda, T., and Ono, T. 1956. Study on essential oils of Hepaticae 1. Essential oils of *Bazzania pompeana*. *J. Jap. Chem. Soc.* **77**: 400–401.

Galatis, B., Apostolakos, P., and Katsros, C. 1978a. Histochemical studies on the oil bodies of *Marchantia paleacea* Bert. *Protoplasma.* **97**: 13–29.

Galatis, B., Apostolakos, P., and Katsros, C. 1978b. Ultrastructural studies on the oil bodies of *Marchantia paleacea* Bert I. Early stages of oil-body cell differentiation: origination of the oil body. *Can. J. Bot.* **56**: 2252–2267.

Galatis, B. and Apostolakos, P. 1976. Associations between microbodies and a system of cytoplasmic tubules in oil-body cells of *Marchantia*. *Planta* **131**: 217–221.

Garjeanne, A. J. M. 1903. Die Ölkörper der Jungermanniales. *Flora* **92**: 457–482.

Gavaudan, P. 1927. Sur l'origine et les charactères des éléments oléifères des Jungermanniales. *C.R. Acad. Sci.* **184**: 1473–1475.

Gottsche, C. M. 1843. Anatomische-physiologische Untersuchungen über *Haplomitrium Hookeri* N. v. E. mit Vergleichung anderer Lebermoose. *Nov. Actorum Acad. Caes. Leop.-Carol. Nat. Cur.* **20**: 267–289.

Gradstein, S. R., Cleef, A. M., and Fulford, M. 1977. Studies on Colombian cryptogams. IIA. Hepaticae—oil body structure an ecological distribution of selected species of tropical Andean Jungermanniales. *Proc. Kon. Ned. Akad. Wetensch. C.* **80**: 377–393.

Guo, L., Wu, J. Z., Han, T., Cao, T., Rahman, K., and Qin, L. P. 2008. Chemical composition, antifungal and antitumor properties of ether extracts of *Scapania verrucosa* Heeg. and its endophytic fungus *Chaetomium fusiforme*. *Molecules* **13**: 2114–2125.

Hashimoto, T., Irita, H., Takaoka, S., Tanaka, M., and Asakawa, Y. 2000. New chlorinated cyclic bis(bibenzyls) from the liverworts *Herbertus sakuraii* and *Mastigophora diclados*. *Tetrahedron* **56**, 3153–3159.

He, X. and Glenny, D. 2010. *Perssoniella* and the genera of Schistochilaceae: a new classification based on molecular phylogenies. *Aust. Syst. Bot.* **23**: 229–238.

He-Nygrén, X., Ahonen, I., Julén, A., Glenny, D., and Piippo, S. 2004. Phylogeny of liverworts—beyond a leaf and a thallus. *Monogr. Syst. Bot. Mo. Bot. Gard.* **98**: 87–118.

He-Nygrén, X., Juslén, A., Ahonen, I., Glenny, D., and Piippo, S. 2006. Illuminating evolutionary history of liverworts (Marchantiophyta) – towards a natural classification. *Cladistics* **22**: 1–31.

Heinrichs, J., Gradstein, S. R., Wilson, R., and Schneider, H. 2005. Towards a natural classification of liverworts (Marchantiophyta) based on the chloroplast gene rbcl. *Crypt. Bryol.* **26**: 131–150.

Hieronymus, G. 1892. Beitrage zur morphologie und biologie der Algen. *Beitr. Biol. Pflanz.* **5**: 461–495.

von Holle, G. 1857. Ueber die Zellenbläschen der Lebermoose. Heidelberg, 26 pp.

Huang, C. Y., Chung, C. I., Lin, Y. C., Hsing, Y. L. C., and Huang, A. H. C. 2009. Oil bodies and oleosins in *Physcomitrella* possess characteristics representative of early trends in evolution. *Pl. Physiol.* **150**: 1192–1203.

Huang W. J., Wu, C. L., Lin, C. W., Chi, L. L., Chen, P. Y., Chiu, C. J., Huang, C. Y., Chen, C. N. 2010. Marchantin A, a cyclic bis(bibenzyl ether), isolated from the liverwort *Marchantia emarginata* subsp. *tosana* induces apoptosis in human MCF-7 breast cancer cells. *Cancer Lett.* **1**: 108–119.

Hübener, I. W. P. 1834. Hepaticologia Cermania. Mannheim.

Huneck, S. and Klein, E. 1967. Inhaltsstoffe der moose—III. : Über die vergleichende gas- und dünnschicht-chromatographische untersuchung der ätherischen öle einiger lebermoose und die isolierung von (−)-longifolen und (−)-longiborneol aus *Scapania undulata* (L.) Dum. *Phytochemistry* **6**: 383–390.

Huneck, S. 1967. Substances contained in moss. IV. Isolation of drimenol from *Bazzania trilobata* (L.) LINDBERG. *Z. Naturforsch. B.* **22**: 462–463.

Iwai, L. K., Duranti, M. A., Abel, L. C. J., Juliano, M. A., Kalil, J., Juliano, L., and Cunha-Neto, E. 2001. Retro-inverso peptide analogues of *Trypanosoma cruzi* B13 protein epitopes fail to be recognized by human sera and peripheral blood mononuclear cells. *Peptides* **22**: 853–860.

Kámory, E., Keseru, G. M., and Papp, B. 1995. Isolation and antibacterial activity of marchantin A, a cyclic bis(bibenzyl) constituent of Hungarian *Marchantia polymorpha*. *Planta Medica* **61**: 387–388.

Kenrick, P. and Crane, P. R. 1997. The origin and early evolution of land plants. *Nature* **389**: 33–39.

Kis, G. and Pócs, T. 1997. Oil body studies on African Hepaticae. *J. Hattori Bot. Lab.* **81**: 175–242.

Kozlowski, A. 1921. Sur l'origine des oléoleucites chez les hépatiques à leuilles. *C. R. Acad. Sci. Paris* **173**: 497–499.

Kronestedt, E. 1983. Cytology of oil-body cells in *Ricciocarpus natans*. *Nord. J. Bot.* **3**: 547–558.

Lehman, H. and Schulz, D. 1982. Die verwendung der semisimultan fixierung zur darstellung der feinstruktur spezialisierter mooszellen. *Mikroskopie* **39**: 285–291.

Lehman, J. and Jaster, B. 1981. Feinstrukturelle untersuchungen zür entwicklung der olzellen be idem lebermoos. *Riella*. *Protoplasma* **106**: 109–119.

Li, M., Murphy, D. J., Lee, K. H., Wilson, R., Smith, L. J., Clark, D. C., and Sung, J. Y. 2002. Purification and structural characterization of the central hydrophobic domain of oleosin. *J. Biol. Chem.* **277**: 37888–37895.

Lindberg, S. O. 1888. Bidrag till nordens mossflora. I. *Meddel. Soc. Fauna Flora Fennica* **14**: 63–77.

Lohmann, C. E. J. 1903. Beitrag zur chemie und biologie der leber moose. *Beih. Bot. Centralb*. **15**: 215–256.

Martínez-Abaigar, J. and Núñez-Olivera, E. 2011. Aquatic bryophytes under ultraviolet radiation. **In:** *Bryophyte ecology and climate change*. pp. 115–146. Tuba, Z, Slack, N. G., and Stark, L. R., Eds., Cambridge University Press, Cambridge, UK.

Matsuo, A., Nakayama, M., and Hayashi, S. 1971a. Fatty acid esters in the volatile oil from the liverwort, *Pellia fabbroniana*. *Phytochemistry* **10**: 430–432.

Matsuo, A., Nakayama, M., Ishida, Y., and Hayashi, S. 1971b. Isolation of Nonacosan-10-ol and cuparene from the liverwort, *Bazzannia pompeana*. *Z. Naturforsch. B.* **26**: 32–33.

Mirbel, M. 1835. Recherches anatomiques et physiologiques sur le *Marchantia polymorpha*. (Anatomical and Physiological Research on Marchantia polymorpha). *Mem. Acad. Sci. Inst. Fr.* **13**: 337–436.

Moellering, E. R. and Benning, C. 2010. RNA interference silencing of a major lipid droplet protein affects lipid droplet size in *Chlamydomonas reinhardtii*. *Eukaryot Cell* **9**: 97–106.

Müller, K. 1905. Beitrag zur Kenntnis der ätherischen Öle bei Lebermoosen. *Z. Physiol. Chem.* **45**: 299–319.

Müller, K. 1939. Untersuchungen über die Ölkörper der Lebermoose. *Ber. Dtsch. Bot. Ges.* **57**: 326–370.

Murphy, D. J. 2001. The biogenesis and functions of lipid bodies in animals, plants and microorganisms. *Prog. Lipid Res.* **40**: 325–438.

Nabeta, K., Mototani, Y., Tazaki, H., and Okuyama, H. 1994. Biosynthesis of sesquiterpenes of cadinane type in cultured cells of *Heteroscyphus planus*. *Phytochemistry* **35**: 915–920.

Nguyen, L. N., Hamari, Z., Kadereit, B., Trofa, D., Agovino, M., Martinez, L. R., Gacser, A., Silver, D. L., and Nosanchuk, J. D. 2011. Candida parapsilosis fat storage-inducing transmembrane (FIT) protein 2 regulates lipid droplet formation and impacts virulence. *Microb. Infect.* **13**: 663–672.

Niu, C., Qu, J. B., and Lou, H. X. 2006. Antifungal bis[bibenzyls] from the Chinese liverwort *Marchantia polymorpha* L. *Chem. Biodiv.* **3**: 34–40.

Otero, S., Martinez-Abaigar, K. C. J., and Nunez-Olivera, E. 2008. Ultraviolet-absorbing capacity of aquatic bryophytes from Tierra del Fuego (Argentina). *J. Bryol.* **30**: 290–296.

Peled, E., Leu, S., Zarka, A., Weiss, M., Pick, U., Khozin-Goldberg, I., and Boussiba, S. 2011. Isolation of a novel oil globule protein from the green alga *Haematococcus pluvialis* (Chlorophyceae). *Lipids* **46**: 851–8861.

Pfeffer, W. 1874. Die Ölkörper der Lebermoose. *Flora* **57**: 2–6, 17–27, 33–43.

Pfeffer, W. 1904. Pflanzenphysiologie. II. Aufl. Verlag von W. Engelmann. Leipzig.

Pihakaski, K. 1966. An electron microscopy study on the oil bodies of two Hepatic species. *Protoplasma* **62**: 393–399.

Pihakaski, K. 1968. A study of the ultrastructure of the shoot apex and leaf cells in two liverworts, with special reference to the oil bodies. *Protoplasma* **66**: 79–103.

Pihakaski, K. 1972a. Histochemical studies on the oil bodies of the liverworts *Pellia epiphylla* and *Bazzania tribobata*. *Ann. Bot. Fenn.* **9**: 65–76.

Pihakaski, K. 1972b. Studies on the hepatic oil bodies, with special reference to the vacuole-like oil bodies of *Pellia epiphylla*. *Univ. Turku Publ. Bot.* **170**: 1–25.

Pressel, S., Duckett, J. G., Ligrone, R., and Proctor, M. C. F. 2009. Effects of De- and Rehydration in Desiccation-Tolerant Liverworts: A Cytological and Physiological Study. *Int. J. Plant Sci.* **170**: 182–199.

Qiu, Y. L., Li, L., Wang, B., Chen, Z., Knoop, V., Groth-Malonek, M., Dombrovska, O., Lee, J., Kent, L., Rest, J., Estabrook, G. F., Hendry, T. A., Taylor,

D. W., Testa, C. M., Ambros, M., Crandall-Stotler, B., Duff, R. J., Stech, M., Frey, W., Quandt, D., and Davis, C. C. 2006. The deepest divergence in land plants inferred from phylogenetic evidence. *PNAS* **17**: 15511–15516.

Rubinstein, C. V., Gerrienne, P., de la Puente, G. S., Astini, R. A., and Steemans, P. 2010. Early Middle Ordovician evidence for land plants in Argentina (eastern Gondwana). *New Phytol.* **188**: 365–369.

Scher, J. M., Burgess, E. J., Lorimer, S. D., and Perry, N. B. 2002. A cytotoxic sesquiterpene and unprecedented sesquiterpenebisbibenzyl compounds from the liverwort *Schistochila glaucescens*. *Tetrahedron* **58**: 7875–7882.

Schuster, R. M. and Hattori, S. 1954. The oil-bodies of the Hepaticae. II. The Lejeuneoideae. *J. Hattori Bot. Lab.* **11**: 11–86.

Schuster, R. M. 1966. The Hepaticae and Anthocerotae of North America. I. Columbia Univ. Press, 802 pp., New York, London.

Schuster, R. M. 1992. The oil-bodies of the Hepaticae. I. Introduction. *J. Hattori Bot. Lab.* **72**: 151–164.

Shi, Q. W, Dong, M., Huo, C. H., Su, X. H., Li, C. F., Zhang, X. P., Wang, Y. F., and Kiyota, H. 2007. New 14-hydroxy-taxane and 2α,20-epoxy-11(15$\rightarrow$1) abeotaxane from the needles of *Taxus canadensis*. *Biosci. Biotech. Biochem.* **71**: 1777–1780.

Shimada, T. L., Shimada, T., Takahashi, H., Fukao, Y., and Hara-Nishimura, I. 2008. A novel role for oleosins in freezing tolerance of oilseeds in *Arabidopsis thaliana*. *Plant J.* **55**: 798–809.

Siloto, R. M. P., Findlay, K., Lopez-Villalobos, A., Yeung, E. C., Nykiforuk, C. L., and Moloney, M. M. 2006. The accumulation of oleosins determines the size of seed oilbodies in *Arabidopsis*. *Plant Cell* **18**: 1961–1974.

Stahl, E. 1888. Pflanzen und Schnecken. *Jena. Z. Med. Naturwiss.* **22**: 557–584.

Stewart, G. 1978. Oil bodies of the New Zealand leafy Hepaticae (Jungermanniales). *New Zealand J. Bot.* **16**: 185–205.

Suire, C., Bouvier, F., Backhaus, R. A., Bégu, D., Bonneu, M., and Camara, B. 2000. Cellular localization of isoprenoid biosynthetic enzymes in *Marchantia polymorpha*. Uncovering a new role of oil bodies. *Pl. Physiol.* **124**: 971–978.

Suire, C. 1966. Contribution à la connaissance cytologique de quelques hépatiques. Thèse : Biologie végétale. Phytobiologie cellulaire et cryptogamie, Université de Bordeaux, 107 pp.

Suire, C. 1970. Recherches cytologiques sur deux Hépatiques: *Pellia epiphylla* (L.) Corda (Metzgeriale) et *Radula complanata* (L.) Dum. (Jungermanniale). Ergastome, sporogénèse et spermatogénèse. *Le Botaniste* **53**: 125–392.

Suire, C. 2000. A comparative, transmission-electron microscopic study on the formation of oil-bodies in liverworts. *J. Hattori Bot. Lab.* **89**: 209–232

Sukkharak, P., Ludwiczuk, A., Asakawa, Y., and Gradstein, R. 2011. Studies on the genus *Thysananthus* (Marchantiophyta, Lejeuneaceae) 3. Terpenoid chemistry and chemotaxonomy of selected species of *Thysananthus* and *Dendrolejeunea fruticosa*. *Crypt. Bryol.* **32**: 199–209.

Sun, B., Yuan, H. Q., Xi, G. M., Ma, Y. D., and Lou, H. X. 2009. Synthesis and multidrug resistance reversal activity of dihydroptychantol A and its novel derivatives. *Bioorg. Med. Chem.* **17**: 4981–4989.

Tazaki, H., Nabeta, K., Okuyama, H., and Becker, H. 1995. Biosynthesis of pinguisone in an axenic culture of the liverwort *Aneura pinguis*. *Biosci. Biotech. Biochem.* **39**: 158–160.

Tremblay, S. L. 2010. Evidence of oil bodies in Paleozoic liverwort fossils. 25th New Phytologist Symposium: Colonization of the Terrestrial Environment, Bristol, UK (poster).

Vieler, A., Brubaker, S. B., Vick, B., and Benning, C. 2012. A Lipid Droplet Protein of Nannochloropsis with Functions Partially Analogous to Plant Oleosins. *Pl. Physiol.* **158**: 1562–1569.

von Küster, W. 1894. Die ölkörper der leber moose und ihr verhältnis zu den elaioplasten. *Dissert. Basel*. 41 pp.

Wakker, J. H. 1888. Studien über die Inhaltskörper der Pflanzenzelle. *Jahrb. wiss. Bot.* **19**: 482–487.

Wellman, C. H., Osterloff, P. L., and Mohluddin, U. 2003. Fragments of the earliest land plants. *Nature* **425**: 282–284.

Wilson, S., Gradstein, R. S., Schneider, H., and Heinrichs, J. 2007. Unravelling the phylogeny of Lejeuneaceae (Jungermanniopsida): Evidence for four main lineages. *Mol. Phyl. Evol.* **43**: 270–282.

Wurzel, G. and Becker, H. 1989. Sesquiterpene lactones in vitro cultures of the liverwort *Ricciocarpos natans*. *J. Hattori Bot. Lab.* **67**: 373–375.

Xue, X., Sun, D. F., Sun, C. C., Liu, H. P., Yue, B., Zhao, C. R., Lou, H. X., and Qu, X. J. 2012. Inhibitory effect of riccardin D on growth of human non-small cell lung cancer: in vitro and in vivo studies. *Lung Cancer* **76**: 300–308.

Zhu, R. L. and So, M. L. 2001. Epiphyllous liverworts of China. Berlin: J. Cramer. 418 pp.

Zirkle, C. 1932. Vacuoles in primary meristems. *Z. Zellforschg* **16**: 26–47.

Zwickel, W. 1932. Studien über die ocellen der Lebermoose. *Beih. Bot. Centralbl.* **49**: 569–648.