CHAPTER 1-24
AQUATIC AND WET MARCHANTIOPHYTA, CLASS MARCHANTIOPSIDA: MARCHANTIACEAE, PART 2

TABLE OF CONTENTS

CHAPTER 1-24
AQUATIC AND WET MARCHANTIOPHYTA, CLASS MARCHANTIOPSIDA: MARCHANTIACEAE, PART 2

Figure 1. ***Marchantia polymorpha*** in a typical waterside habitat. Photo by Michael Lüth, with permission.

Marchantiaceae, cont.

Marchantia polymorpha (Figure 1-Figure 12)

(syn. = *Marchantia alpestris*; *Marchantia aquatica*; *Marchantia coarctata*) – see Long (1995) for a discussion of the history of older naming of the species and subspecies.

Boisselier-Dubayle *et al.* (1995) elaborated on the genetic variability in the three subspecies of ***Marchantia polymorpha*** (Figure 1-Figure 12). The three subspecies [***montivagans*** (Figure 2-Figure 4), ***polymorpha*** (Figure 5), ***ruderalis*** (Figure 6-Figure 8)] had high genetic similarity within each subspecies over a wide geographic area, but the similarity was low between subspecies. These differences indicate that speciation included adaptation to different ecological niches, followed by reproductive isolation.

Figure 2. ***Marchantia polymorpha*** ssp. ***montivagans*** with antheridiophores. Photo by Jan-Peter Frahm, with permission.

Figure 3. ***Marchantia polymorpha*** ssp. ***montivagans*** with archegoniophores. Photo by Hugues Tinguy, with permission.

Figure 4. ***Marchantia polymorpha*** ssp. ***montivagans*** with gemmae cups. Photo by Hugues Tinguy, with permission.

Figure 5. ***Marchantia polymorpha*** ssp. ***polymorpha***. Photo by Jan-Peter Frahm, with permission.

Figure 6. ***Marchantia polymorpha*** ssp. ***ruderalis*** with gemmae cups. Photo by Jiří Kameníček, with permission.

Figure 7. ***Marchantia polymorpha*** ssp. ***ruderalis*** antheridial receptacles. Photo by Malcolm Storey, DiscoverLife, with online permission.

Figure 8. ***Marchantia polymorpha*** ssp ***ruderalis*** females with receptacles. Note smaller archegoniophores on the outer (younger) thalli. Photo by Malcolm Storey, DiscoverLife, with online permission.

The variety ***aquatica*** (Figure 9) (Campbell 1968; Hollensen & Taylor 1981; Kitagawa 1987) is not recognized by Söderström *et al*. (2016). Instead, it is considered within the species ***Marchantia polymorpha*** as ssp. ***polymorpha***. Nevertheless, it appears that those thalli that develop underwater can be distinguished by a black

midrib (Michael Lüth, pers. comm.). Recognition of this distinction can give us an ecological history of plants we find and deserves some experimental attention.

Figure 9. ***Marchantia polymorpha*** ssp. ***polymorpha*** with distinct black midribs, typical of aquatic forms, in Europe. Photo by Michael Lüth, with permission.

Google Scholar found 20,500 references in a search for ***Marchantia polymorpha*** (Figure 1-Figure 12). ***Marchantia polymorpha*** has been a subject of intensive study for nearly 200 years (Shimamura 2016). The species offers many benefits for research, including its short life cycle, ease of propagation and crossing, high frequency of transformation, haploidy, and small genome size (approximately 280 Mb).

Figure 10. ***Marchantia polymorpha*** with overlapping thalli. Photo by Sanja through Wikimedia Commons.

Berrie and Webster (1982) elaborated on the ultrastructure of the plastids and mitochondria of the ***Marchantia polymorpha*** gemmae (Figure 4). They found that division among plastids and maturing vegetative cells differed from that of developing oil body cells and rhizoid initials. Bopp and Vicktor (1988) developed methods for following protoplast development, determining that cell wall formation requires light.

Figure 11. ***Marchantia polymorpha*** thallus. Photo from Botany Website, UBC, with permission.

A Liverwort Model

genetics and sequencing

The species has served as a model organism in many biological studies (Chiyoda *et al*. 2008), including the discovery of sex in cryptogams and more recently the elucidation of the V chromosome (term for male chromosome in haploid organism), understanding the plant life cycle, and origins of polarity in development. The use of ***Marchantia polymorpha*** *s.l.* (Figure 1-Figure 12) as a plant model system continues (Durand 1908; Alam & Pandey 2016; Shimamura 2016), with its dominant haploid generation being a benefit for genetic studies and gene expression as well as details of evolutionary and developmental biology.

Marchantia polymorpha (Figure 1-Figure 12) has been used to demonstrate the presence of "X" and "Y" chromosomes in bryophytes, now referred to as U and V chromosomes, respectively (Renner *et al*. 2017). I shall continue this discussion using the designation of U for female and V for male. This early discovery of sex chromosomes was followed by the sequencing of the male and female genomes (Ohyama 2001). Ohyama found that some of the genes on the V chromosome were unique, whereas those on the U chromosome were also on somatic chromosomes or even on the V chromosome. Since these are haploid organisms, the males have a very small V chromosome with no U chromosome and females have one U chromosome with no second U or any V (Lorbeer 1934; Tanurdzic & Banks 2004). Sporophytes are UV.

Yamato *et al*. (2007) reported that these V chromosomes differ from other chromosomes in lacking recombination. They reported the gene organization of the V chromosome of ***Marchantia polymorpha*** (Figure 1-Figure 12) and identified 64 genes on the V chromosome. Of these, 14 are found only in the male genome and are expressed only in male reproductive organs. Another 40 are expressed in thalli and reproductive organs. Interestingly, at least 6 of these have U-linked counterparts that are expressed in both thalli and sex organs of females. Yamato and coworkers suggested that these sex chromosomes share ancestral autosomal genes, and they predict that essential genes on sex chromosomes of haploid organisms are more likely to persist than those in diploid organisms.

As in many other studies, ***Marchantia polymorpha*** (Figure 1-Figure 12) was the choice for studying the divergence of land plant chloroplast (Figure 12) genes (Morton 1994). Bischler (1986) analyzed the karyotype. In ***Marchantia polymorpha*** the genes with the highest codon adaptation index correspond to the ones that are expressed at the highest levels (Morton 1994). This relationship is weaker in *Nicotiana tabacum* (Figure 13).

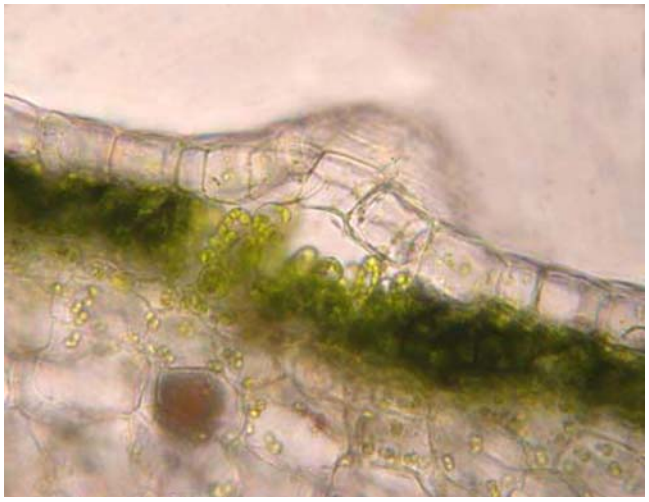

Figure 12. ***Marchantia polymorpha*** thallus section through pore, showing location of photosynthetic filaments with chloroplasts. Photo from Botany Website, UBC, with permission.

Figure 13. *Nicotiana tabacum*, a species that demonstrates the conservation of ***Marchantia*** genes in flowering plants. Photo through Creative Commons

One interesting finding is that chloroplast ribosomal protein rpl 21 in ***Marchantia polymorpha*** (Figure 1-Figure 12) is encoded by the plastid gene, but in tobacco and rice this is a nuclear gene (Smooker *et al*. 1990). Sone *et al*. (1999) reported for the first time a co-localization of repeat rDNA in land plants. These researchers suggested that the structural re-organization of rDNAs occurred after the evolutionary divergence of bryophytes from other plants.

Marchantia polymorpha (Figure 1-Figure 12) appeared to be one of the first organisms to have its chloroplast and mitochondrial DNA sequenced (Ohyama *et al*. 1986; Oda *et al*. 1992a; Kisiel *et al*. 2011; Lin *et al*. 2016). But unfortunately, the species used in early studies (Ohyama *et al*. 1986; Oda *et al*. 1992a,b,c,d) was a mis-identified ***Marchantia paleacea*** (Figure 14) (Kijak *et al*. 2013, 2016; Masaki Shimamura, pers. comm. 10 July 2022). Hence, this first sequencing cannot be attributed to ***Marchantia polymorpha***.

Figure 14. ***Marchantia paleacea*** with archegoniophores. Photo by Jan-Peter Frahm, with permission.

Posno *et al*. (1986) showed that there was "substantial" conservation in the chloroplast genome sequences between ***Marchantia polymorpha*** (Figure 1-Figure 12) and those of the aquatic flowering plant ***Landoltia punctata*** (Figure 15). Umesono and Ozeki (1987) found that the chloroplast genome sequence of ***Marchantia polymorpha*** differs little in gene makeup and function from that of flowering plants, despite flowering plants one that is 25%.

Figure 15. *Landoltia punctata*, a species with considerable conservation of the chloroplast genome sequences found in ***Marchantia polymorpha***. Photo from US Dept. Interior, through Creative Commons.

Kohchi *et al.* (1988) characterized parts of the chloroplast DNA in presumably ***Marchantia paleacea*** (Figure 14), under the name of ***M. polymorpha***. Raubeson and Jansen (1992) further explored what these DNA sequences had in common with later plants.

Takemura *et al.* (1992) elaborated on ribosomal proteins that are coded in the mitochondrial genome of ***Marchantia polymorpha*** (possibly ***M. paleacea***), and these differ substantially in size from their counterparts in the bacterium ***Escherichia coli*** (Figure 16). In the same year, Oda *et al*. (1992a,b,c) found that the mitochondrial DNA of ***Marchantia paleacea*** (Figure 14; misidentified as ***M. polymorpha***) is a single circular form that exhibits no incorporation of chloroplast DNA. Whichever species was actually used, Ohyama *et al*. (1982, 1983, 1986, 1988a,b,c; Ozeki *et al*. 1987; Los & Semenenko 1991) showed that many (most?) chloroplast genes in such liverworts as ***Marchantia*** have been conserved in tobacco, *Nicotiana tabacum* (Figure 13).

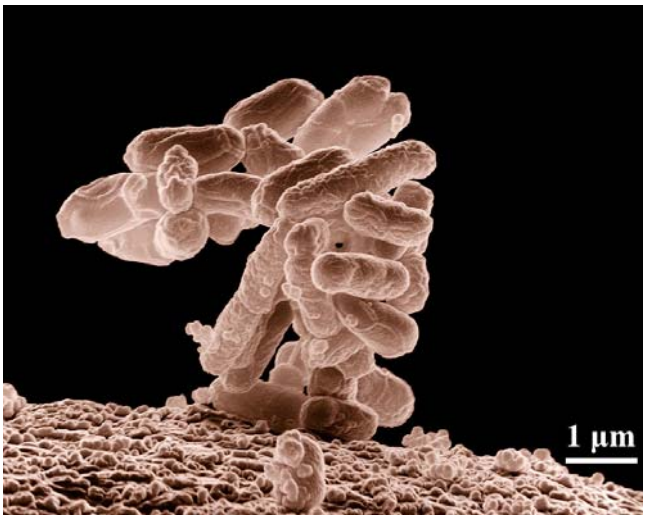

Figure 16. ***Escherichia coli***, a bacterium that exhibits very different ribosomal proteins from those of ***Marchantia polymorpha***/***paleacea***. Photo from USDA, through public domain.

Takemura *et al.* (1992) elaborated on ribosomal proteins that are coded in the mitochondrial genome of ***Marchantia polymorpha*** (possibly ***M. paleacea***), and these differ substantially in size from their counterparts in the bacterium ***Escherichia coli*** (Figure 16).

Downie *et al*. (1991) and Sibbald (1988) subsequently compared chloroplast DNA of dicotyledonous flowering plants with those attributed to ***Marchantia polymorpha***. Comparisons between mosses and liverworts with hornworts indicated that the two former groups were more closely related to each other than either was to hornworts (Katoh *et al*. 1983).

The entire gene sequence was not known until 2017, using ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 17-Figure 19) (Bowman *et al.* 2017). Since then, the subspecies have been sequenced and their evolutionary relationships elucidated (Linde *et al*. 2020). The results supported the hypothesis that ***M. polymorpha*** ssp. ***ruderalis*** is not a hybrid of the other two subspecies, as some researchers had thought.

Figure 17. ***Marchantia polymorpha*** ssp. ***ruderalis*** gemmae cups. Photo by David T. Holyoak, with permission.

Figure 18. ***Marchantia polymorpha*** ssp. ***ruderalis*** with archegoniophore. Photo by Jiří Kameníček, with permission.

Figure 19. ***Marchantia polymorpha*** ssp. ***ruderalis*** with archegonial head showing numerous scales. Photo by Jiří Kameníček, with permission.

Marchantia polymorpha (Figure 1-Figure 12) has likewise been used as a model organism in understanding development in thallose liverworts (Suzuki *et al*. 2020).

But the scientific study of this species, especially regarding development, has been occurring for at least 200 years (Shimamura 2016). Suzuki and coworkers described merophyte lineages and elaborated on the derivation of growth from stem cells in the apical notch (Figure 20).

Figure 20. ***Marchantia polymorpha*** showing apical notch with brown covering scale. Photo by Li Zhang, with permission.

Linde *et al*. (2021) compared rates of mutations among the major plant groups and found that the gymnosperms had the fewest synonymous mutations, flowering plants the most, and bryophytes were located between those two. In this regard, the silent site substitution rate (neutral evolution) is lower for the liverwort compared to flowering plants, but not as low as that of gymnosperms. They found the same selective constraints on the haploid-specific genes as those on the diploid-specific genes. However, in the haploid generation the new mutations experience immediate and direct selection, hence quickly being lost if they are maladaptive. The nonsynonymous to synonymous substitution rate ratio (dN/dS) represents selective evolution. This silent site substitution rate is lower for liverworts as compared to flowering plants, but again not as low as for gymnosperms. The selection pressure, measured as dN/dS, is not remarkably lower for bryophytes when compared to diploid dominant plants as we might expect based on the **masking hypothesis** (predicts more efficient selection in haploids than in diploids, because dominant alleles can mask deleterious effects of recessive alleles in diploids; however, gene expression breadth and noise can potentially counteract the effect of masking on the rate at which genes evolve), indicating that other factors are more important than ploidy.

Liu *et al.* (2019) noted that the dN/dS for nuclear genes was more than three times higher for bryophytes when compared to seed plants. However, the sets of genes compared were not the same genes in both groups. Linde and coworkers (2021) compared the same 42 genes in both groups and found that the dN/dS rates were much more similar. This raises the question of why bryophytes, given their long history, are less diverse and appear to have less morphological diversity than tracheophytes, especially flowering plants. But as I have argued elsewhere, they are confined to being small due to their lack of lignin. They furthermore, because of this small size, are at greater risk of disappearance due to herbivory and pathogens. If one considers their biochemical diversity, affording them protection from herbivores and pathogens, are they really less diverse?

Fang *et al*. (2014) reported for the first time the presence of oleosin genes in liverworts, using ***Marchantia polymorpha*** (Figure 1-Figure 12). The liverworts tested exhibited only M-oleosins, whereas three types are known from various plant lineages. This seems to be a precursor to the other two oleosins, suggesting another piece of the evolutionary story.

With all of our knowledge about molecular similarities and differences, molecular/genetic studies have become the driving force in plant systematics. Using the **Marchantiales**, Boisselier-Dubayle *et al*. (1997) were among the early researchers to point out that there can be an incongruence between morphological characters and molecular data, complicating our systematic efforts.

Understanding the genome, coupled with the greater ease of working with haploid organisms, has contributed to the use of ***Marchantia polymorpha*** (Figure 1-Figure 12) in transferring genes to determine their functions.

teaching

The genus ***Marchantia*** is notable in the pre-Renaissance literature, with illustrations of ***M. polymorpha*** (Figure 1-Figure 12) appearing as early as the mid-15th Century (Bowman 2016). Notable early treatments are those of Schmidel (1762) and Hedwig (1783). It has been used for centuries in nearly every textbook and classroom that teaches about bryophytes or life cycles (Cutting 1910; Inoue & Asakawa 1966; Register & West 1971; Une 1998; Bowman 2016).

Marchantia polymorpha (Figure 1-Figure 12) has also been used as a pedagogical model to teach the bioindicator concept to seventh graders (Pedroza-Manrique & Arévalo 2009). This was part of a study on comparison of learning strategies. Results demonstrated that "learning must be related to the environment of the students and must also represent a challenge for them, this allowed for significant learning."

Durand (1908), in preparing material for teaching a course in embryology, selected ***Marchantia polymorpha*** (Figure 1-Figure 12) to represent development in liverworts. He wrote that he "naturally selected ***Marchantia polymorpha***" because of its accessibility and ease of study. And it is illustrated in practically every textbook that treats liverworts.

Distribution

Marchantia polymorpha (Figure 1-Figure 12) has a worldwide distribution (Figure 21), especially in the Northern Hemisphere (Bischler-Causse 1989; Lu & Huang 2017). It is known from Afghanistan, Bhutan, China, India, Indonesia (Java, Sumatra), Iran, Iraq, Israel, Japan, Korea, Lebanon, Malaysia, Nepal, New Guinea, New Zealand, Pakistan, Philippines, Russia, Sri Lanka, Syria, Tadzhikistan, Taiwan, Tasmania, Thailand, Turkey, USSR, Uzbekistan, and Vietnam (Bischler-Causse 1989; Söderström *et al*. 2010; Ginting & Batubara 2019). As seen in Figure 21, it is also extensively reported throughout North America. ***Marchantia polymorpha*** has three subspecies. In South Africa, ***M. polymorpha*** ssp. ***ruderalis*** (Figure 6-Figure 8, Figure 17-Figure 19) only occurs in

nurseries (Figure 22-Figure 23), indicating that it has been introduced (Ginting & Batubara 2019).

Figure 21. ***Marchantia polymorpha*** distribution. Image from DiscoverLife, with online permission.

Figure 22. ***Marchantia polymorpha*** ssp. ***ruderalis*** in flower pot in nursery, the only place it occurs in South Africa. Photo courtesy of Javier Martínez-Abaigar.

Figure 23. ***Marchantia polymorpha*** ssp. ***ruderalis*** in flower pot intended for growing a tree. Photo courtesy of Javier Martínez-Abaigar.

Aquatic and Wet Habitats

Boisselier-Dubayle and Bischler (1989) described the electrophoretic biochemistry of ***Marchantia polymorpha*** (Figure 1-Figure 12) as having a good correlation with the habitats. They identified two ecotypes: domestic with a spread over a wide geographic area (possibly corresponding with ***M. polymorpha*** ssp. ***ruderalis*** – Figure 6-Figure 8, Figure 17-Figure 19), and a wet-habitat-restricted ecotype that exhibited two biotypes [corresponding with ***M. polymorpha*** ssp. ***polymorpha*** (Figure 5) and possibly ***M. polymorpha*** ssp. ***montivagans*** (Figure 2-Figure 4)].

In their recent publication on ***Marchantia*** in Japan, Zheng and Shimamura (2022) described the habitat of ***Marchantia polymorpha*** ssp. ***polymorpha*** (Figure 5) as growing on wet soil in marshland or stone near streams, seldom on wet concrete. They described ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 6-Figure 8, Figure 17-Figure 19) as often on soil, stone, gravel, or walls of drainage channels in anthropogenic regions, supporting the conclusions of Long (1995). But Zheng and Shimamura (2022) also found that when the latter subspecies grows in dripping wet habitats, "plants often have an erect and robust thallus with a distinct and continuous blackish median band on the thallus and are sometimes difficult to distinguish from ssp. ***polymorpha*** based on these characters." However, they consider that the discontinuous blackish median band and prostrate thallus of ssp. ***polymorpha*** will distinguish them. I would like to see common garden experiments on the effects of wet vs drier habitats on the expression of the black median band (midrib) in each of the subspecies.

Subspecies ***montivagans*** (Figure 2-Figure 4) in Britain and Ireland is a species occurring in springs, marshes, and flushes, in somewhat calcareous areas, and occurs mostly in subalpine or montane habitats (Long 1995). It can also be found on damp calcareous mossy banks and rocks beside streams and ravines. However, in their studies Boisselier-Dubayle and Bischler (1989) found it in some locations together with ***M. polymorpha*** ssp. ***polymorpha*** (Figure 5).

Frye (1928) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) was sometimes submersed in winter. This, presumably, was ssp. ***polymorpha*** (Figure 5).

Darigo (2004) reported the aquatic form of ***Marchantia polymorpha*** (as *Marchantia aquatica*; now ***M. polymorpha*** ssp. ***polymorpha***; Figure 5) as new to Missouri, USA, noting its distinctive black midrib. He found it in dense mats on shaded moist soil and limestone bedrock at the base of a small waterfall. It was associated with ***Fissidens grandifrons*** (Figure 24), ***Rhizomnium punctatum*** (Figure 25), and ***Leptodictyum riparium*** var. ***laxirete*** (Figure 26).

Marchantia polymorpha *s.l.* (Figure 1-Figure 12) occurs in wet places around lakes, especially in shade, and in bog water in Scotland (West 1910). In Denmark it can be found on terrestrial soil or submerged (Sørensen 1948). It is uncommon with the graminoid ***Eleocharis quinqueflora*** (Figure 27) and the moss ***Paludella squarrosa*** (Figure 28) Geissler & Selldorf 1986).

Figure 24. ***Fissidens grandifrons***, an associate of ***Marchantia polymorpha*** on limestone in wet habitats. Photo by Brad Von Blon, through Creative Commons.

Figure 25. ***Rhizomnium punctatum***, an associate of ***Marchantia polymorpha*** on limestone in wet habitats. Photo by Sharon Pilkington, with permission.

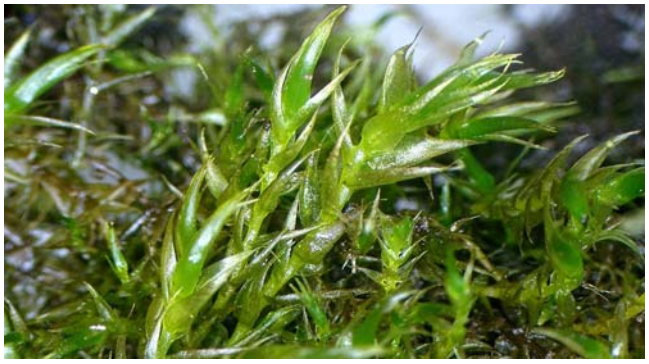

Figure 26. ***Leptodictyum riparium***; var. ***laxirete*** is an associate of ***Marchantia polymorpha*** on limestone in wet habitats. Photo by William Lampa, through Creative Commons.

Figure 27. ***Eleocharis quinqueflora***, an occasional associate of ***Marchantia polymorpha*** in Denmark. Photo by Kristian Peters, through Creative Commons.

Figure 28. ***Paludella squarrosa***, an occasional associate of ***Marchantia polymorpha*** in Denmark. Photo by Hermann Schachner, through Creative Commons.

Marchantia polymorpha (Figure 1-Figure 12) occurs in shady areas with high humidity and exhibits a low tolerance to desiccation (Lagos-López 2008). In such areas it can develop extensive cover (Linares 1986; Churchill & Linares 1995; Uribe & Aguirre 1995; Lagos-López 2008). But perhaps one of the most extensive covers develops in burned over areas (Durand 1908; Janet Marr, pers. obs. 2022), usually not an aquatic habitat.

streams and rivers

Watson (1919) described the habitat of ***Marchantia polymorpha*** (Figure 1-Figure 12) as often on boulders in streams, on banks with frequent submergence and slow water. In the Western Carpathians it occurs in the rock community in streams near Lacko (Mamczarz 1970). It

occurs in streams in Greece (Papp 1998) and in streams in northeastern Finland (Heino & Virtanen 2006). It is among the most common species in the River Tweed, UK (Holmes & Whitton 1975) and is among the commonest species in English and Welsh rivers (Scarlett & O'Hare 2006).

It can also be common in rivers (Ferreira *et al*. 2008), such as the Iskur River, Bulgaria, and its main tributaries (Papp *et al.* 2006), as a hygrophyte in Bulgarian rivers (Gecheva *et al*. 2010, 2013), and in the Iregua River of Spain (Martínez-Abaigar & Ederra 1992).

It is often a mountain species, occurring in alpine streams in the Swiss Alps (Geissler 1976) and in mountain streams of northwest Portugal (Vieira *et al.* 2005) as well as mountainous streams on Madeira Island (Luis *et al.* 2015). Frye (1928) reported it from western Washington, USA, in burned sites, but also in submersed sites. It is interesting that several ***Marchantia*** species occur in both aquatic habitats and as pioneers after fire.

stream and river banks

Perhaps the most common wet habitat for ***Marchantia polymorpha*** (Figure 1-Figure 12) is the damp banks of streams and rivers (Figure 29-Figure 32). In the Haute Ardenne rivers in Belgium, it occurs on earthy and gravelly substrates of river banks (Leclerq 1977) and in the UK it occurs on the riverbank of the River Tees (Holmes & Whitton 1977a). It can be found throughout the River Swale, Yorkshire, UK (Holmes & Whitton 1977b), and mostly in the mid to lower River Tyne, UK (Holmes & Whitton 1981b). It is similarly associated with the River Wear in England (Birch *et al*. 1988). In Germany it occurs in the middle and lower reaches in the Harz Mountains (Bley 1987). Ginting and Batubara (2019) similarly describe it from rocks of a creek wall in exposed places (Figure 33), at 1500 m altitude in Indonesia. In western Canada, it is restricted to terrestrial habitats of montane streambanks (Vitt *et al*. 1986; Glime & Vitt 1987).

Figure 29. ***Marchantia polymorpha*** ssp. ***polymorpha*** on the bank of a small stream, showing partially upright branches. Photo by Vladimir Bryukhov, through Creative Commons.

Figure 30. ***Marchantia polymorpha*** on steep stream bank. Photo by Olga Chernyagina, through Creative Commons.

Figure 31. ***Marchantia polymorpha*** ssp. ***polymorpha*** wet beside stream. Note the rhizoids hanging from the lower side of the thallus. Photo by Penny Anderson, with online permission.

Figure 32. ***Marchantia polymorpha*** with gemmae cups on moist soil by stream. Photo by Rudolf Macek, with permission.

In Denmark ***Marchantia polymorpha*** (Figure 1-Figure 12) can be found in ditches at *p*H 6.2-8.5 and in neutro-alkaline lakes, peat pits of spring bogs (Figure 34) at *p*H 6.2-8.0, on terrestrial soil or submerged (Sørensen 1948; Clausen 1952). Alfasane *et al*. (2013) recorded environmental conditions in Lake Rainkhyongkain, where it grew in masses in the shallow littoral zone of the lake. These included mean values (n = 4) of dissolved oxygen

content (7.93 ± 0.78 mg L^{-1}), alkalinity (1.70 ± 0.12 meq L^{-1}), soluble reactive phosphorus (17.25 ± 0.62 µg L^{-1}), soluble reactive silicate (10.44 ± 0.72 mg L^{-1}), and NO_3-N (34.00 ± 4.00 µg L^{-1}). The mean temperature on the sampling date was 33.5°C and *p*H was 7.39.

Figure 33. ***Marchantia polymorpha*** with ***Thuidium delicatulum*** growing over flagstone at Mountain Moss Enterprise, demonstrating the breadth of its habitat. Photo by Annie Martin, with permission.

In the high moor transition areas of Denmark ***Marchantia polymorpha*** (Figure 1-Figure 12) can be found at *p*H 5.0-6.09 (Sørensen 1948). O'Toole and Synnott (1971) found that ***Marchantia polymorpha***, along with ***Funaria hygrometrica*** (Figure 35), was an early indicator of increased calcium carbonate and phosphorus levels on blanket peat following fertilization of the peat. However, these two bryophytes are suited to other environmental conditions, with ***M. polymorpha*** preferring wet, unsheltered plots with no iron or copper, whereas ***F. hygrometrica*** prefers dry, sheltered locations with iron (Synnott 1987). Li Zhang (pers. comm. 4 August 2022) has found ***Marchantia polymorpha*** in peatlands (Figure 34) in China.

Figure 34. ***Marchantia polymorpha*** ssp. ***montivagans*** in a spring, Sierra Nevada near Merida, Venezuela. Photo courtesy of Javier Martínez-Abaigar.

Figure 35. ***Funaria hygrometrica***, an indicator, along with ***Marchantia poymorpha***, of increased calcium carbonate and phosphorus levels in blanket peat. Photo by Bonnie Nickel, through Creative Commons.

In Taiwan, Lu and Huang (2017) found ***Marchantia polymorpha*** (Figure 1-Figure 12) on damp soils from 300-2500 m asl, a habitat similar to that on Mt. Edith Cavell in Canada (Figure 36).

In high moors in Denmark it occurs in the transition areas at *p*H 5.0-6.0 (Sørensen 1948). In the Caucasas it is in watery and swampy lands (Alijev & Babajev 1976).

Figure 36. ***Marchantia polymorpha*** with gemmae, on damp soil with mosses on the mountainside of Mt. Edith Cavell, Jasper, Canada. Photo by Janice Glime.

Other damp soil habitats are also suitable, including open areas (Figure 37), moist slopes (Figure 38), marshy areas (Figure 39), and other wetlands (Figure 40). Thatcher (1949) found it in an artificially illuminated cave.

Figure 37. ***Marchantia polymorpha*** on soil, with gemmae cups, in Europe. Photo by Michael Lüth, with permission.

Figure 38. ***Marchantia polymorpha*** on a damp soil bank. Photo by Michael Lüth, with permission.

Figure 39. ***Marchantia polymorpha*** in a marshy habitat near a stream. Photo by Michael Lüth, with permission.

One of the more interesting habitats for ***Marchantia polymorpha*** (Figure 1-Figure 12) is on a **desalinating wadden-polder** (tract of low land reclaimed from the sea; Figure 41) (Joenje & During 1977). Although it was able to colonize, it was unable to compete after 2-3 years. It was especially common on mussel banks. Joenje and During suggested that its small spores contribute to its rapid arrival on newly available substrata.

Figure 40. ***Marchantia polymorpha*** ssp. ***montivagans*** with ***Calliergon cordifolium*** in a wetland. Photo by Des Callaghan, through Creative Commons.

Figure 41. Gradual transition from recently deposited salt marshes and the Wadden sea on a desalinating wadden-polder. This stage is too salty, due to manipulation, and the only bryophyte able to live here is ***Hennediella heimii***. Photo courtesy of Bart van Tooren.

Another unusual habitat where ***Marchantia polymorpha*** (Figure 1-Figure 12) can thrive is in geothermal areas (Figure 42). These can have some similarities to saline areas because of sulfur and other salts. In the Antarctic, they provide a warm haven for species from warmer climes, including ***M. polymorpha*** (Kennedy 1996). Takaki (1967) found it on an active volcano in Japan. Takaoki and Mitani (1986) used ***Marchantia polymorpha*** in experiments to develop a method for measuring the effects of SO_2 on photosynthesis in bryophytes and lichens. They found that illumination during the exposure to SO_2 caused the SO_2 to have greater inhibition of photosynthesis in ***M. polymorpha*** than did SO_2 alone. In this species, the photosynthetic system was more sensitive than the respiratory system. In concentrations less than 4 ppm, the thalli were able to partially recover.

Figure 42. Geothermal fissure with ***Sphagnum***, Geyser, Iceland, a potential habitat for ***Marchantia polymorpha***. Photo by Janice Glime.

Figure 43. Mt. Hood Riverside, Oregon, USA, post-fire, at a stage where ***Marchantia polymorpha*** can be a pioneer. Photo from U.S. Department of Agriculture, through public domain.

after fire

It is interesting that several species of ***Marchantia*** are post-fire colonizers (Figure 43). This is true for ***Marchantia polymorpha*** (Figure 1-Figure 12) (Benson & Blackwell 1926; Torrey 1932; Bradbury 2006). Frye (1928) and Hoffman (1966) both reported it on burned sites in Washington, USA, and Graff (1936) similarly found that the species invaded after forest fires. Adámek *et al*. (2016) studied post-fire vegetation in central Europe and again found that ***Marchantia polymorpha*** was among the first invaders, but it typically disappeared after a few years. Froment (1975) found it to be among the pioneer dominants in a Belgian high fen post-fire site. Heras-Ibáñez *et al.* (1991) recorded it in post burn sites in SE Spain, where it had diminished numbers after 6 years. They noted that some of the species, including ***M. polymorpha***, were known to fix nitrogen (through bacterial partners) on such sites. Duckett *et al*. (2008) noted that ***M. polymorpha*** is common after fires in Canada and explored post-fire invasion at Thursley Common, UK. Once again, ***M. polymorpha*** was a common invader. In fact, they noted that thalli had abundant reproductive structures in the first year following the fire. They attributed the rapid colonization to the tiny (10-16 μm) spores. Subspecies. ***ruderalis*** and ssp. ***polymorpha*** were present side by side. The species formed patches reaching up to 12 m in diameter, with individual colonies of 5-20 cm diameter (Duckett & Pressel 2009). Rather that naming tracheophyte competition, they attributed the later decline to nutrient leaching (Duckett *et al*. 2008). Its presence is so common after fire that Rees and Juday (2002), when reporting it from Alaskan burned sites, considered it to be a fire specialist.

The succession of species following fire is a fairly constant one, as noted already by Skutch (1929). Typically, the pioneer stage with ***M. polymorpha*** (Figure 1-Figure 12) is succeeded by ***Polytrichum*** (Figure 44).

These habitats would seem to be quite different from the wet ones already discussed. But one thing they have in common is reduced competition, giving the liverworts time to become established.

Figure 44. ***Polytrichum juniperinum***, a post-fire species that succeeds ***Marchantia polymorpha***. Photo by James K. Lindsey, with permission.

Having said that, how then do we explain that in East Anglia ***Marchantia polymorpha*** (Figure 1-Figure 12) is more common in town than in the countryside (Stevenson & Hill 2008)? Is the subspecies ***ruderalis*** more tolerant of competition, or do the spores manage to find spots with limited competition?

Physiology

It is not surprising that the anatomy and physiology of this well known species were among the early bryological studies (Mirbel 1835). Since that time, a long period of mostly taxonomic activity ignored the physiology of most bryophytes. However, ***Marchantia polymorpha*** (Figure 1-Figure 12) was frequently the subject of those studies that did address bryophyte physiology.

hormones - IAA

Among the many studies on ***Marchantia polymorpha*** (Figure 1-Figure 12), it often served as the model organism for understanding physiology of thallose liverworts, or bryophytes in general. Maravolo and Voth (1966) reported the actions of various concentrations of indoleacetic acid (IAA), naphthalene acetic acid (NAA), and maleic hydrazide on development of sterile gemmae (Figure 45) of ***Marchantia polymorpha***. They found that the two auxins

(IAA and NAA) promoted similar responses, often resembling those of tracheophytes. In ***M. polymorpha***, rhizoids elongated on the dorsal surface; the growing region became dormant, and cells became strongly elongated. When apical growth (Figure 20, Figure 46) was inhibited, the thallus became winged. The maleic hydrazide likewise elicited responses similar to those in tracheophytes, including inactivation of meristematic regions, deterioration of chloroplasts, inhibition of rhizoid production, and **hyperplasia** (enlargement of organ or tissue caused by increased reproduction rate of its cells).

Figure 45. ***Marchantia polymorpha*** with gemmae in gemmae cups. Photo by Holger Casselmann, through Creative Commons.

But at that time, the production of IAA by bryophytes was still unknown. Schneider *et al*. (1967) reported for the first time that IAA (indoleacetic acid) occurs in bryophytes, using ***Marchantia polymorpha*** (Figure 1-Figure 12) as one the test organisms. Furthermore, Sheldrake (1971) reported auxin in the substrata of bryophytes, so its effect on them is relevant. However, concentrations of the IAA in substrata with no bryophytes were the same in those that had them. Hence, it is questionable whether these substrate sources were of importance to the development of the bryophytes. Nevertheless, they could play a role in the induction of rhizoids as they do for roots. And they might have more effect on some bryophytes not included in the tests.

Following up on the possible functions, Otto and Halbsguth (1976) examined the effects of light and the auxin IAA on the formation of the primary rhizoids of gemmae in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that the number of rhizoid-forming gemmae depends on the wavelength of irradiation. Following a 1-hour exposure to 10^{-4} M IAA, rhizoids developed as they would if exposed to red irradiation for one hour. They concluded that this relationship suggests a role of the phytochrome system in membrane permeability for IAA.

Transport of the auxin 11C-indoleacetic acid occurs basipetally in the thallus, where it is localized in the midrib (Figure 46-Figure 47) (Maravolo 1976; Gaal *et al*. 1982).

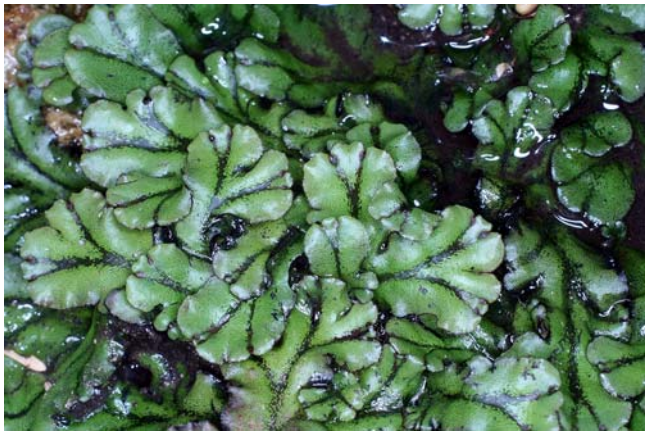

Figure 46. ***Marchantia polymorpha*** with strong midrib on older thalli; this character is common in aquatic forms. In several branches you can see apical dominance where a single branch is dominant and the other is shorter or has an unbranched midrib (*e.g.* **center left**). Photo from Dale A. Zimmerman Herbarium, Western New Mexico University, with permission.

Figure 47. ***Marchantia polymorpha*** with only hints of midrib on young thallus tissue, probably ssp. ***ruderalis***. Photo by Janice Glime.

Reynolds and Maravolo (1973) determined that extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12) contain substances that in the lab can enhance or inhibit IAA oxidation. These extracts contained four unknown phenolic compounds. Two of these enhanced the oxidase activity and two inhibited it. Although the inhibitors were present throughout the thallus, they were slightly more concentrated in basal and apical areas. In other words, there was an **acropetal** (movement of dissolved substances outward toward shoot and basal apices) gradient of increase in this cofactor concentration. In this case IAA moves away from the growing tips.

Gaal *et al*. (1982) used tracer experiments to determine that ^{14}C IAA transport is localized in the cells of the midrib of the thallus (Figure 46). It occurs both acropetally and basipetally, with a far greater intensity basipetally. In anaerobic conditions, basipetal transport was reduced by 40-50%. Centrifugation also disrupted it, reducing basipetal transport by 30-40%. Hence, in the parameters measured, transport of IAA in ***Marchantia polymorpha*** (Figure 1-Figure 12) is essentially the same as that in seed plants.

Davidonis and Munroe (1972) described apical dominance (Figure 46) in ***Marchantia polymorpha*** (Figure 1-Figure 12), a function known from tracheophytes. The species dichotomous branching and its apical dominance is exhibited by one of those branch lobes having greater growth than the other (Figure 46). They demonstrated that if the lobes are separated by a cut, the dominance is lost. IAA is needed to maintain this dominance. If it is applied to the smaller lobe after the cut, that lobe is again inhibited. It also exhibits greater inhibition of the smaller lobe when applied to the intact plant. Interestingly, the dominant lobe is neither inhibited nor enhanced by application of IAA to it. They suggested that this behavior indicates that the two lobes have a different sensitivity to the auxin. This is somewhat reminiscent of apical dominance in plants like snapdragons. IAA produced in the apex of the plant migrates downward and inhibits the development of side branches. If the apex is removed, a lower branch becomes dominant.

Hormones can both activate and suppress pathways in ***Marchantia polymorpha*** (Figure 1-Figure 12). Binns and Maravolo (1972) found that cytokinin suppressed the normal germination of gemmae (Figure 45), instead stimulating nodular, callus-like growths. Responses were time-dependent on the cytokinin-enriched medium. Auxin, on the other hand, had no effect on the regeneration of normal thallus growth. Auxin could, however, reverse the suppression caused by inhibiting compounds such as transcinnamic acid. Auxin exhibited a basipetal gradient and this gradient is essential to normal growth and regeneration. These two hormones interact, with high levels of cytokinin destroying the polarity by increasing the auxin-synthesizing capacity. Maintaining the right balance permits the liverwort to maintain its apical dominance (Figure 20, Figure 46).

Flores-Sandoval *et al*. (2015) characterized the functions of the auxin transcriptional response in ***Marchantia polymorpha*** (Figure 1-Figure 12). It acts to facilitate branching, differentiation, and growth, but it does not determine specific tissues.

Eklund *et al*. (2015) found that the auxin IAA regulates the dormancy of gemmae (Figure 48) on the ***Marchantia polymorpha*** (Figure 1-Figure 12) thallus. Kato *et al*. (2015) found that auxin repression could cause severe defects in the development of ***Marchantia polymorpha***, including gemmaling development, dorsiventrality, organogenesis, and tropic responses, noting the interactions of hormones. Billhardt (2021) further discussed the interactions, and using these was able to explain the long-known observation that gemmae typically do not germinate while still on the parent thallus (Figure 48). This dormancy mechanism is controlled by high levels of abscisic acid (ABA) in the cup (see also Tougane *et al*. 2010). But gemmae are not attached to the thallus while they reside in the cup. Therefore, it appears that the actual signal is a gas. Billhardt found that when the regulators of the ethylene signalling pathway were mutated, it affected the dormancy, suggesting that ethylene regulates dormancy through ABA. Müller (2021) notes that ABA and ethylene commonly act antagonistically, possibly explaining these observations.

Ishizaki *et al*. (2012) used transgenic plants to monitor the effects of auxin-mediated transcriptional activation in plants. This demonstrated that IAA had a role in the transcription of some genes. These genes were demonstrated at the bottom of the gemmae cups. Additional activity occurred at the gametophyte-sporophyte junction and in the developing sporophyte.

Figure 48. Gemmae cups with loose dormant gemmae on thallus of ***Marchantia polymorpha***, demonstrating their continued dormancy while associated with the parent tissue. Photo by Hermann Schachner, through Creative Commons.

hormones - gibberellins

Melstrom *et al*. (1974) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) exhibits gibberellin activity and was responsive to photoperiod. When they increased the photoperiod from 12 to 18 hours of light the activity of the gibberellins increased and thallus elongation and **orthogeotropic** (directly in line with gravitational pull; Figure 49) growth increased.

Figure 49. ***Marchantia polymorpha*** ssp. ***polymorpha*** growing upright in water. Photo by Oleg Kosterin, through Creative Commons.

Maravolo (1980) applied both auxins (IAA) and gibberellin to bryophytes, including ***Marchantia polymorpha*** (Figure 1-Figure 12). In this case, the applied auxin stimulated rhizoid growth, cell proliferation, and elongation. Gibberellin, on the other hand, promotes cell enlargement, chloroplast development, and starch degradation. Under the right photoperiod, it also influences the geotropic curvature and causes ultrastructural changes in starch granules and thylakoids.

Loomis and Maravolo (1985) found that exogenous gibberellin increases the amylolytic activity of two protein fractions from ***Marchantia polymorpha*** (Figure 1-Figure 12).

hormones - ethylene

Little information has been available on ethylene presence and physiology in bryophytes. Ethylene is a gaseous hormone and therefore can be used to communicate between plants. Katayose *et al*. (2021) noted that genes for ethylene have been conserved from the algae, but that the function and biosynthesis of this hormone remain unknown in the bryophytes. They found that ***Marchantia polymorpha*** (Figure 1-Figure 12) synthesizes ethylene. However, treatment with the precursor ACC only slightly promoted the production of ethylene. On the other hand, ACC "remarkably" suppressed thallus growth and rhizoids, contrasting with the slight promotion of thallus growth when external ethylene was applied. These experiments indicate that ethylene functions independently of ACC and that ACC is not essential to its production in ***Marchantia polymorpha***.

Li *et al*. (2020) similarly found that ACC (ethylene precursor) and ethylene can induce different responses in ***Marchantia polymorpha*** (Figure 1-Figure 12). Ethylene causes larger gemmae, induces more gemmae cups, and promotes the dormancy of the gemmae. ACC, on the other hand, inhibits gemma growth and development by suppressing cell division. This suggests that the pathway might be different from that tracheophytes.

In ***Marchantia polymorpha*** (Figure 1-Figure 12), more ethylene is produced in the light than after prolonged darkness (Fredericq *et al*. 1977; Rethy *et al*. 1977). Veroustraete *et al*. (1982) further elaborated on ethylene physiology in the species. They found involvement of the low energy red:far-red reversible type of phytochrome action for both the light-induced ethylene production and the control of **epinasty** (nastic movement in which plant part such as flower petal or thallus branch is bent outward and often downward; Figure 50) in the species. They found that CO_2 had no effect on the production of ethylene when the thallus was irradiated with terminal far-red light, but in controls without the light treatment, there was a clear CO_2 dependency. This behavior suggests the involvement of phytochrome. De Greef *et al*. (1979) studied environmental effects on ethylene production in ***M. polymorpha*** and concluded that ethylene production requires energy and depends on either cyclic photophorylation or oxidative phosphorylation.

Figure 50. ***Marchantia polymorpha*** showing an unusual margin formation that exhibits **epinasty**. Photo by Steve Trynoski, with permission.

hormones - cytokinins

Binns and Maravolo (1972) found that cytokinin suppressed germination of gemmae (Figure 45) in ***Marchantia polymorpha*** (Figure 1-Figure 12). Externally applied auxins had no effect on regeneration from thallus discs.

Aki *et al*. (2019a,b) noted that cytokinins regulate a variety of physiological events in plants. They found that the cytokinin signalling pathway in ***Marchantia polymorpha*** (Figure 1-Figure 12) controls the formation of both gemmae cups (Figure 45, Figure 48) and rhizoids (Figure 53-Figure 54) during the development of the thallus. It is further implicated in the distribution of air pores (Figure 51) and the shape of the thallus margin (compare Figure 49 and Figure 50), suggesting that cytokinins regulate cell division or differentiation of precursor cells, thereby coordinating development.

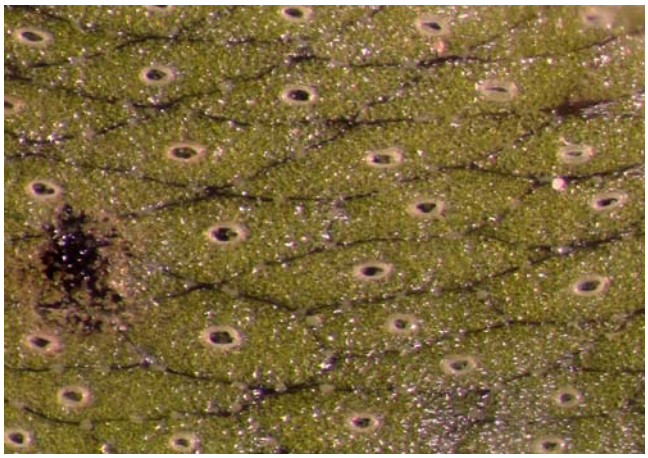

Figure 51. ***Marchantia polymorpha*** epidermis with air pores, showing green layer beneath. Photo by Walter Obermayer, with permission.

hormones – ABA and lunularic acid

In 1979, Weiler used a radioimmunoassay in an attempt identify the presence of ABA in ***Marchantia polymorpha*** (Figure 1-Figure 12). At that time, using that sensitive technique, ABA appeared to be absent. Fortunately, that result was not accepted by everyone. Li *et al*. (1994) announced, for the first time, the presence of abscisic acid (ABA) in liverworts, using ***Marchantia polymorpha*** as the model organism. In fact, the concentrations were similar to those of tracheophytes.

Akter *et al*. (2014) found that pretreatment with ABA and sucrose increases the survival rate after both freezing and desiccation of gemmalings in ***Marchantia polymorpha*** (Figure 1-Figure 12). ABA also increases the accumulation of soluble sugars. Furthermore, ABA induces the accumulation of transcripts for proteins that are similar to late embryogenesis abundant (LEA) proteins, proteins that accumulate in maturing seeds as they acquire desiccation tolerance. ABA also causes the vacuoles to fragment, causing an increase in the cytosolic volume and increasing the volume and density of chloroplast distribution.

Eklund *et al*. (2018) also found that ABA delays the germination of gemmae (Figure 48) in ***Marchantia polymorpha*** (Figure 1-Figure 12).

Ghosh *et al*. (2016) was the first to report on the regulation of ABA in the liverwort ***Marchantia polymorpha*** (Figure 1-Figure 12). Previously, a hormone with similar functions, **lunularic acid**, was known from some liverworts. Ghosh and co-workers found that the expression of ABA-induced β-glucoronidase (GUS) reporter gene was less in older, mature thalli than in young gemmalings of this species This change corresponded with reduction in the sensitivity to exogenous ABA. Lunularic acid, on the other hand, had no effect on GUS expression.

Nevertheless, it appears that ***Marchantia polymorpha*** (Figure 1-Figure 12) has **lunularic acid** (Gorham 1977; Abe & Ohta 1983), a hormone with functions similar to those of ABA. However, it appears that in ***M. polymorpha***, the prelunularic acid greatly exceeds the lunularic acid (Abe & Ohta 1984). Gorham found it in all parts of ***M. polymorpha***. In continuous light, both lunularic acid and fresh weight increased relative to that in interrupted light periods. It appears that the lunularic acid either was not inhibited by continuous light or that the photosynthetic products overrode the inhibition.

Imoto and Ohta (1985) found that lunularic acid compounds were equally distributed in vacuoles and cytoplasm, but they were absent in plastids, mitochondria, and peroxisomes.

hormones - brassinosteroids

Brassinosteroids occur in a wide range of organisms, including early land plants (Bajguz & Hayat 2009). These steroidal plant hormones affect the promotion of plant growth and development. Metabolism of these steroids is altered when plants respond to abiotic stresses as well as bacterial, fungal, and viral pathogens. Ko *et al*. (1995) characterized five 4-demethylsterols that seem to be potent biosynthetic precursors of brassinosteroids in suspension cell cultures of ***Marchantia polymorpha*** (Figure 1-Figure 12).

cell growth

Fries (1964) explored the effects of growth inhibitors on growth and elongation in ***Marchantia polymorpha*** (Figure 1-Figure 12).

Matsui *et al*. (1991) detected the activity of lipoxygenase in ***Marchantia polymorpha*** (Figure 1-Figure 12) in culture. Most of this activity occurred in the **cytosol** (aqueous component of cytoplasm of cell). The activity increases rapidly during the lag phase of cell growth (Matsui *et al.* 1996). This activity decreased in the logarithmic phase, then increased again in the stationary phase. This series of changes in lipogenase activity is caused by *de novo* synthesis and degradation of the same lipoxygenase. They also found enzyme activity that degrades fatty acid hydroperoxides – products of lipoxygenase.

circadian rhythm

Lagercrantz *et al.* (2020) examined the circadian clock in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that this clock coordinates the **nyctinastic** (periodic movement plant parts, especially flowers or leaves, caused by nightly changes in light intensity or temperature) thallus movement of the species and suggested it is controlled by auxin (IAA). The thalli "wave" up and down on a 24-hour cycle in 12 hours light: 12 hours dark. These movements in gemmalings are maintained in continuous light. The auxin, produced in the apical region (Figure 20), travels basipetally through the midrib region, creating a gradient. The circadian rhythm regulates the IAA levels. At low doses (10-100 nM) the angle of growth is reduced, creating a more flattened thallus. These experiments support the observations of Went and Thimann (1937) that the response to auxin depends on the time of day.

Marchantia polymorpha (Figure 1-Figure 12) has often been a model for evolution in land plants. Linde *et al*. (2017) found homologues of core clock genes in *Arabidopsis* (Figure 52), bryophytes, and charophytes, with fewer copies in the latter. The data supported the hypothesis that adaptation to terrestrial life occurred earlier than that supposed by current theory, particularly occurring in the charophytes. The bryophytes exhibit not only duplication and acquisition of new genes, but also loss of genes in development of their circadian clock.

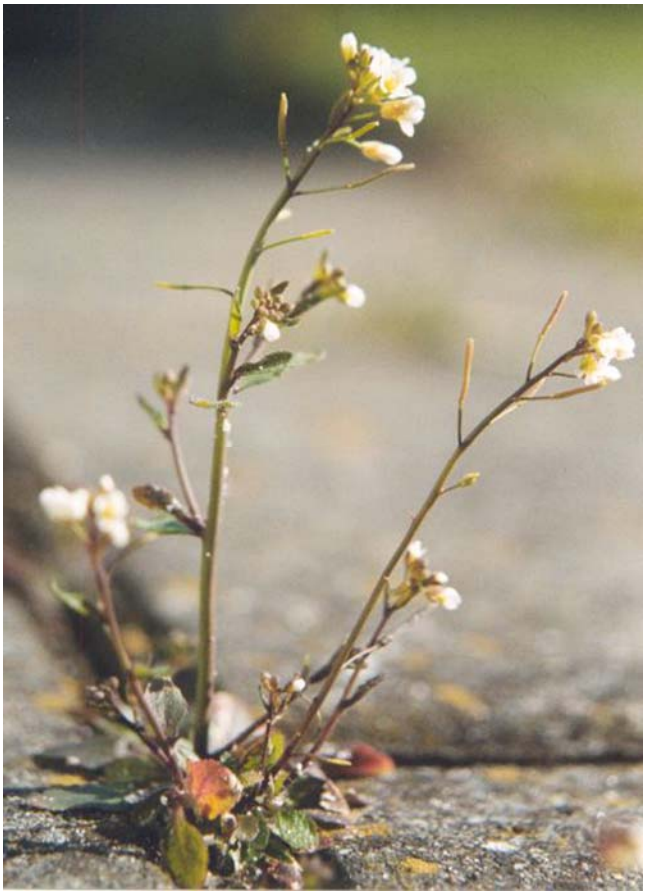

Figure 52. *Arabidopsis thaliana*, in a genus that has homolog genes with early land plants and is frequently used for evolutionary comparisons. Photo by Roepers, through Creative Commons.

Cuitun-Coronado *et al*. (2022) noted that photosynthesis is a circadian process in some flowering plants and Cyanobacteria. They reported the first record of circadian regulation of the photosynthetic pathway in a liverwort, ***Marchantia polymorpha*** (Figure 1-Figure 12). They determined that the light:dark cycle synchronized the 14-hour photosynthetic cycle, but that the phases of different thalli desynchronize under free-running

conditions. They suggested that chloroplast translation might be necessary for the clock to control the light-harvesting process in this plant.

Lagercrantz *et al*. (2021) used sequencing to identify the genes involved in circadian rhythms in ***Marchantia polymorpha*** (Figure 1-Figure 12). They identified a homolog of the *Arabidopsis* (Figure 52) gene DE-ETIOLATE1 as having a high amplitude and morning phase. The circadian rhythm resulting from Mp*DETl* expression is disrupted when core clock genes lose their function in mutants. In knock-down experiments with this gene, the circadian rhythm of nyctinastic thallus movement is altered. But the researchers were unable to detect any effect in response to light, leaving us with no explanation of the function of the Mp*DEP1* gene in ***M. polymorpha***.

water relations

Ghosh *et al*. (2021) explored the drought tolerance in this model organism. They desiccated gemmae in various desiccating solutions and found that these led to extreme growth inhibition, disruption of membrane stability, and reduction in chlorophyll content. At the same time, the accumulation of hydrogen peroxide and malondialdehyde increased and electrolyte leaked from the gemmalings, creating oxidative stress. Activities of antioxidant enzymes, including superoxide dismutase, catalase, ascorbate peroxidase, dehydroascorbate reductase, and glutathione S-transferase increased, while total antioxidant activity also increased in response to increased oxidative stress. When they applied exogenous ABA, it reduced drought-induced tissue damage and improved the activities of antioxidant enzymes and accumulation of proline.

Godinez-Vidal *et al*. (2020) noted that both water deficit and ABA cause osmotic adjustment in ***Marchantia polymorpha*** (Figure 1-Figure 12). This species increases its ABA levels under water deficit. Like Ghosh and coworkers, they found decreased growth and morphological changes in response to water deficits. Cell organelles changed locations, largely due to the volume change of the central vacuole, a consequence of the change in osmotic potential.

Hatanaka and Sugawara (2010) found that after exposure to drying below 0.1 g H_2O g^{-1} dry weight the desiccation tolerance level of ***Marchantia polymorpha*** (Figure 1-Figure 12) was very low, with a survival rate of less than 10%. When cells were pretreated in 0.5 M sucrose, the survival rate rose to 87%, even at lower water levels. This treatment caused cell alteration and the accumulation of a large amount of sucrose and newly made proteins.

Duckett and Ligrone (2003) found that rhizoids (Figure 53) in the bryophytes, particularly the **Marchantiales**, contribute to their water movement. They also contribute to movement of food. The smooth rhizoids (Figure 53-Figure 54) are living cells and often contain fungal hyphae. The pegged rhizoids (Figure 53-Figure 54) are dead, but they nevertheless contribute to the movement of water in the grooves of the archegoniophores. They also help to prevent the collapse when the thalli dry out and they facilitate recovery upon rehydration.

Figure 53. ***Marchantia polymorpha*** ventral surface of thallus showing pattern of horizontal pegged rhizoids that are close to the thallus and that facilitate water movement and uptake. The pinkish rhizoids toward the base are the perpendicular smooth rhizoids that adhere to the thallus. Photo by Larry Jensen, with permission.

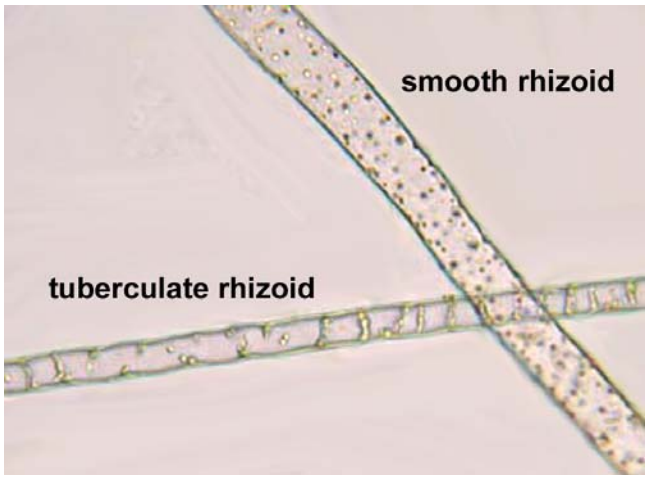

Figure 54. ***Marchantia polymorpha*** smooth and tuberculate (pegged) rhizoids. Photo from Botany Website, UBC, with permission.

Marchantia polymorpha ssp. ***ruderalis*** (Figure 6-Figure 8, Figure 17-Figure 19, Figure 56) was among the first plants to use extracellular ice formation (Figure 55) as a dehydrating agent, a mechanism to provide freezing avoidance (Schott *et al*. 2021). During exposure to freezing temperatures, ice crystals formed in the air chambers and grew out the pores. These hygroscopic crystals, along with other ice on the outside of the thallus, drew ice out of the cells and caused their dehydration. This ice removal can prevent the formation of crystals within the cells, where the crystals can cause membrane and structural damage. The thallose liverwort ***Conocephalum salebrosum*** (Figure 57) proved to be more resistant to frost than did ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 56).

Figure 55. Extracellular ice crystals on ***Marchantia polymorpha***, a mechanism for causing dehydration and preventing freezing damage to membranes. Photo by David Taylor, with permission.

Figure 56. ***Marchantia polymorpha*** ssp. ***ruderalis*** with gemmae. Photo by Malcolm Storey, DiscoverLife, with online permission.

Figure 57. ***Conocephalum salebrosum***, a species that is more frost resistant than is ***Marchantia polymorpha*** ssp. ***ruderalis***. Photo by Hermann Schachner, through Creative Commons.

translocation

Rota and Maravolo (1975) traced the translocation of ^{14}C sucrose in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that during regeneration, higher levels of ^{14}C-sucrose moved to the apical region than during normal growth. The auxin IAA inhibited the transport and thallus regeneration.

desiccation

Pence (1998) found that for successful cryopreservation, ***Marchantia polymorpha*** (Figure 1-Figure 12) required both ABA and encapsulation in alginate beads, differing from ***Riccia fluitans*** (Figure 58) and ***Helicodontium capillare*** (Figure 59) that only required one of these. Without ABA and encapsulation, ***M. polymorpha*** was killed upon drying in liquid nitrogen. I found that interesting because ***R. fluitans*** is more aquatic than is ***M. polymorpha***, but perhaps it is because it is structurally smaller.

Figure 58. ***Riccia fluitans***, a species requiring only one of the two cryopreservation treatments (ABA and encapsulation in alginate beads) required by ***Marchantia polymorpha***. Photo by Kerry Wixted, through Creative Commons.

Figure 59. ***Helicodontium capillare***, a species requiring only one of the two (ABA and encapsulation in alginate beads) cryopreservation treatments required by ***Marchantia polymorpha***. Photo by D. Peralta, MNHN, through Creative Commons.

nutrients

Voth and Hamner (1940) described some of the symptoms of nutrient deficiency as expressed in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that cultures that lacked Ca ions but contained ions of K and Mg were able to regenerate new thalli, primarily from adventitious buds that arose from ventral cells in the midrib region. When NO_3 and PO_4 were absent, the ventral layers of cells developed a red-violet color in the walls, reminiscent of the phosphate deficiency in seed plants. Nevertheless, phosphate was needed in very small quantities in ***Marchantia polymorpha***. Increasing levels of nitrate increased growth, provided all essential nutrients were present.

Rico-Reséndiz *et al*. (2020a) unravelled the responses to low phosphate in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that phosphate starvation elicited the induction of phosphatase activity, acidification of the media, reduction of the internal phosphate concentration, and developmental changes in the rhizoids. Lipid turnover enzymes led to the synthesis of **auronidins** (see below). Up-regulation of certain genes led to changes in organic acid biosynthesis and transport, favoring citric acid exudation. The genes involved in the synthesis of cytokinin are repressed and those involved in auxin and ethylene signalling are upregulated (Rico-Reséndiz *et al*. 2020b). Genes involved in jasmonate synthesis were highly upregulated, but those involved in jasmonate signalling did not change their expression. It appears that auxin and ethylene act as positive regulators in rhizoid development when phosphate is limited, possibly increasing surface area for potential phosphate absorption. Cytokinin, on the other hand, may act as a negative regulator. These observations revealed diverse strategies that contribute to the ability to cope with low phosphate levels.

Voth (1941, 1943) compared various nutrient solutions on ***Marchantia polymorpha*** (Figure 1-Figure 12) growth. It is interesting that vegetative growth was favored by different concentrations than were gemmae cups. Furthermore, gemmae cups are in greater number on male plants compared to female plants, with a ratio of 1.44 to 1.0. High salt concentrations caused the growing tips to die and the thalli to become translucent. In low salt concentrations, "anthocyanin" (probably auronidin; see Albert *et al*. 2018; Kubo *et al.* 2018) was produced, along with numerous rhizoids and a sturdy plant body with thicker cell walls. Presumably the assumption of anthocyanin was due to a red coloration.

Absence of K (potassium) in the medium causes development of tan-colored bases on the plants and narrower tips (Voth 1941). Absence of Ca causes nearly immediate death of the growing tips. Both nitrate and phosphate deficiency cause reddening of the scales (Figure 60-Figure 61), rhizoids, and lower epidermis.

Miller *et al*. (1962a) developed methods to obtain nutritionally deficient mutants of ***Marchantia polymorpha*** (Figure 1-Figure 12). At that time, even few mutants of tracheophytes had been created for that purpose. Nevertheless, little has been published on nutrient deficiency symptoms in bryophytes.

Takio (1987) reported nitrate reductase activities in extracts from cultured cells of ***Marchantia polymorpha*** (Figure 1-Figure 12) growing in a medium with only nitrate as a nitrogen source. They found that the liverworts differed from the mosses in using NADPH as the electron donor, whereas the mosses used NADH. The coenzyme requirement also differed from that known for the other green plants.

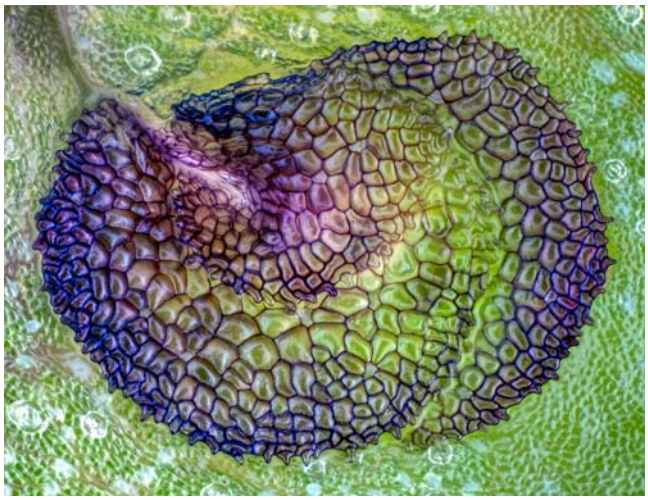

Figure 60. ***Marchantia polymorpha*** ssp. ***ruderalis*** scales showing purplish color that could develop as a deficiency symptom or possibly help to reflect the green light back to the chloroplasts. Photo by Des Callaghan, with permission.

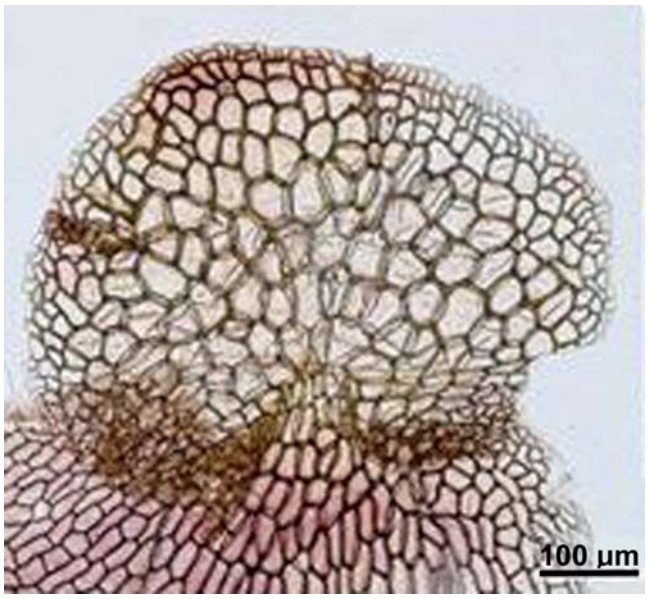

Figure 61. ***Marchantia polymorpha*** ventral scale and epidermis showing reddening that is typical of phosphate or nitrate deficiency. Photo by Masaki Shimamura, with permission.

Not all nutrients are inorganic compounds. Some of these are organic. Dunham and Bryan (1968) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) is able to use a number of different nitrogenous compounds. They tested 24 compounds and found that nine of these had any effect on normal development. Among these, l-isoleucine, l-leucine, l-methionine, and l-threonine caused a disruption of the apical regions at concentrations of 10^{-3} M. At even lower concentrations, l-arginine, l-histidine, l-hydroxyproline, l-lysine, and l-tryptophan each caused morphological irregularities that were highly specific to the

amino acid. The modifications ranged from lack of development of the air chambers to complete thallus disorganization. The effects were dependent on the concentrations of the amino acids. This negative interaction raises questions about the role of compounds released by the decomposition of leaf litter in the environment in interfering with the growth of at least some bryophytes.

Effects of nutrients can differ between males and females of ***Marchantia polymorpha*** (Figure 1-Figure 12). Males with abundant nitrates produce many gemmae cups, narrower thalli, and incurved margins Female plants in these conditions produce fewer cups, have broader thalli, and plant surfaces remain flat.

Bryophytes grow in strange places and on strange substrata. Walkinshaw *et al*. (1970) tested ***Marchantia polymorpha*** (Figure 1-Figure 12) among the plants used to see the effects of lunar rocks on plant growth. They found that this liverwort grew several times larger than normal and had enhanced pigmentation when grown on media enriched with lunar rocks. Hoffman (1974) tested the effects of finely ground ilmenite basalt and loam-textured C-horizon substrate that was rich in volcanic ash, mixtures that closely resemble lunar samples from the Apollo 11 mission, on the growth of ***Marchantia polymorpha***. The moon rock was already known to stimulate the growth of ***M. polymorpha***. Analyses of the two Earth volcanic soil types suggested that the growth stimulus resulted from additional nutrients present in the basalt or volcanic ash.

heavy metals and pollution

For most nutrients, high concentrations become lethal. This is particularly true for heavy metals (Wu & Bradshaw 1972; Ares *et al*. 2018). These are often needed in small quantities in enzymes, but soon become toxic at higher quantities.

For this reason, bryophytes, including ***Marchantia polymorpha*** (Figure 1-Figure 12Figure 23), are suitable organisms to indicate heavy metal pollution (Maschke 1981). Coombes and Lepp (1974) examined the effects of zinc and copper on gemmalings of ***Marchantia polymorpha***. Copper proved to be more toxic than zinc, inhibiting gemmaling growth at levels above 8 ppm. Zinc actually had little effect on the gemmalings.

Ares *et al*. (2018) examined the physiological responses of ***Marchantia polymorpha*** (Figure 1-Figure 12) to Cd, Cu, Pb, and Zn. Under high concentrations, there was a significant enrichment and translocation of Cu, Zn, and especially Cd, achieving a concentration of 1800 $\mu g\ g^{-1}$ in three weeks. On the other hand, Pb achieved the lowest concentration (50 $\mu g\ g^{-1}$), with 90% of the total concentration in the rhizoids. Ozkem *et al*. (2019) further found that when ***Marchantia polymorpha*** was exposed to elevated levels of $CuCl_2$, $ZnCl_2$, and $Pb(NO_3)_2$ it experienced a significant reduction in chlorophyll content.

Both zinc and copper are toxic to the gemmalings of ***Marchantia polymorpha*** (Figure 1-Figure 12), with copper being more effective (Coombes & Lepp 1974). At levels of copper above 8 ppm ***M. polymorpha*** gemmalings exhibited greatly reduced growth. Other morphological changes also occurred.

Lepp and Roberts (1977) found that cadmium at levels above 5 ppm had negative impacts on gemmaling growth of ***M. polymorpha*** (Figure 1-Figure 12). Furthermore, respiration rates diminished with increasing Cd levels. Gekeler *et al*. (1989) found that ***M. polymorpha*** produces two phytochelatins when exposed to cadmium.

Samecka-Cymerman *et al*. (1997) summarized previous studies on heavy metals, noting that the concentrations of elements in the liverworts they studied, including ***Marchantia polymorpha*** (Figure 1-Figure 12), correlate positively with the concentrations of elements in the soil. Some, such as cobalt, surpass the background values found in most bryophytes, indicating the ability of the bryophytes to accumulate them. Some elements, such as Fe, Co, Pb, and Cu, caused an ionic imbalance in this liverwort and others.

Iron is sometimes considered a micronutrient and sometimes a macro nutrient. ***Marchantia polymorpha*** (Figure 1-Figure 12) uses reduction-based iron acquisition. Under deficiency conditions, growth of this species is reduced. Activity of ferric chelate reductase is increased and proton ATPase becomes active (Lo *et al*. 2016).

Manganese is needed in photosynthesis where it catalyzes the water-splitting reaction, but excess Mn creates metal stress. In ***Marchantia polymorpha*** (Figure 1-Figure 12) excess Mn causes a strong accumulation of N-methylalanine, a response differing from that of tracheophytes (Messant *et al*. 2022). When the concentrations of Mn were not optimal, the ratio of photosystem I to PSII changed and the organization of the thylakoid membranes was altered. This is important in photoprotection. The deficiency of Mn favors cyclic electron flow around PSI, thus protecting PSII against photoinhibition.

As demonstrated by the **bryometer** (air bags holding bryophytes), pollutants such as SO_2, oxidants, NO, and NO_2 can cause severe toxicity to ***Marchantia polymorpha*** (Figure 1-Figure 12) on the leeward side of a pollution source, resulting in the lowest growth rate in the area (78%) (Yokobori 1978; Yokobori & Taoda 1980).

responses to abiotic stress

Fujita *et al*. (2006) pointed out that much of the research on molecular mechanisms that cope with stress in plants have been carried out independently. Hence, our understanding of the evolutionary relationships and convergence points between biotic and abiotic stress signaling pathways remains very incomplete. More recently, evidence is emerging that suggests that hormone signaling pathways regulated by abscisic acid, ethylene, jasmonic acid, and salicylic acid, in addition to ROS-signaling pathways, play important roles in the crosstalk between biotic and abiotic stress signaling.

Marchantia polymorpha (Figure 1-Figure 12) has a large repertoire of responses to environmental changes (Spinedi *et al*. 2021). In response to anthracene, they found an increase in the activity of main ROS-detoxifying enzymes of 34.09% of peroxidase and 692% of ascorbate peroxidase, supported at transcriptional level with the up-regulation of ROS-related detoxifying responses. The net result was the activation of antioxidant mechanisms and the accumulation of the anthracene pollutant within the plant tissues.

Hirata *et al*. (2000) used bornyl acetate as a chemical stressor of ***Marchantia polymorpha*** (Figure 1-Figure 12).

In response, the liverwort produced peroxidase. This is a glycoprotein that is stable at temperatures as high as 50ºC for up to one hour, suggesting that the liverwort might have protection against the increasing temperatures of climate change. Its optimum *p*H is 6.5, which does not bode well for the dangers of acid rain. The peroxidase appears to be unlike any of those known from tracheophytes.

Hydrogen peroxide (H_2O_2) often has a protective role in plants. It forms in ***Marchantia polymorpha*** (Figure 1-Figure 12) in the presence of $MnCl_2$. Its production is also stimulated by phenols such as 2,4-dichlorophenol (a 2,4-D precursor) or *p*-coumarate, both processes similar to the last step in lignification, suggesting that while bryophytes apparently lack lignin, they already had a large part of the process required for its production. Phenols are important substances in antiherbivory in bryophytes.

Bryophytes are often resistant to stresses that can kill tracheophytes. Merwin (2003) found that herbicides designed for long-term use on tracheophytes actually promoted the growth of ***Marchantia polymorpha*** (Figure 1-Figure 12). They furthermore were resistant to the human traffic in the orchard.

Measurements of impedance can be a tool to determine health of small plants like ***Marchantia polymorpha*** (Figure 1-Figure 12). Bulanda (1980) Researchers have used this species to develop and test the efficacy of a method for measuring the resistance and capacity of the thallus, based on previous methods for measuring these in cell suspensions (Bulanda 1980; Paszewski *et al*. 1982).

radiation damage

A need to understand radiation damage arose as we began to explore space and to use radiation for energy. Typically, the effects on the nucleus were used to assess such damage (Miller & Sparrow 1964). One generalization that arose indicated that cells with smaller nuclei had more resistance to the radiation than did those with large nuclei. Using ***Marchantia polymorpha*** (Figure 1-Figure 12) Miller and Sparrow found that a more accurate indicator was the nuclear volume (at interphase) divided by the chromosome number. Miller *et al*. (1965) found that larger nuclei in gemmae exhibited inhibition at lower levels of radiation than did smaller nuclei.

Sarosiek and Wozakowska-Natkaniec (1967) demonstrated that chronic gamma radiation caused inhibition of the development of sex organs in ***Marchantia polymorpha*** (Figure 1-Figure 12).

One of the tools used in assessing effects of such things as X-rays is to use mutants (Miller *et al*. 1962a, b). Bryophytes, particularly ***Marchantia polymorpha*** (Figure 1-Figure 12), are particularly suitable for this because of their haploid condition. Miller and coworkers used the method to obtain nutritionally deficient mutants, a condition that could affect the response to radiation.

CO_2

The current atmosphere has a CO_2 concentration of about 0.0415% (415 ppm) (Climate.gov, accessed 21 September 2022). But it seems that most of the studies on CO_2 effects on the physiology of ***Marchantia polymorpha*** have been done at much higher levels.

Katoh *et al*. (1979) found that in 1% CO_2 ***Marchantia polymorpha*** (Figure 1-Figure 12) had a dry-weight doubling time of 1.76 days. The increase rate of chlorophyll was 1.6 times that of the growth rate. In the exponential phase of growth, the photosynthetic activity was at least 60 µmol mg^{-1} chl h^{-1}. The highest chlorophyll content they recorded was 24 mg g^{-1} dry weight.

Bockers *et al*. (1997) compared responses of ***Marchantia polymorpha*** (Figure 1-Figure 12) to two levels of CO_2. At the higher concentration (2.0%), the chloroplast shape seemed modified and there were 70% more chloroplasts per cell than at 0.4%. However, the chlorophyll content per cell indicated a reduction in chlorophyll per chloroplast. Furthermore, the cell size was about 37% lower in the higher CO_2 concentration. The net result was that the photosynthetic oxygen evolution was about the same under both conditions.

Marchantia polymorpha (Figure 1-Figure 12) has a C_3 pathway (Hanson *et al*. 2002), as do all bryophytes. Its growth form is a thallus, similar to that of hornworts. Its **CO_2 compensation point** (CO_2 concentration at which photosynthetic rate = respiration rate) was 64 ppm (Hanson *et al*. 2002), whereas the hornwort ***Megaceros*** (Figure 62), a genus lacking pyrenoids (Villarreal & Renner 2012), had a compensation point of 31 ppm (Hanson *et al*. 2002). On the other hand, ***Notothylas*** (Figure 63) and ***Phaeoceros*** (Figure 64), both with pyrenoids, had compensation points of 11-13 ppm CO_2. Those species lacking **pyrenoids** had more **RuBisCo** content, permitting them to increase their carboxylation catalytic rate (***Marchantia***, 2.6 s^{-1}; ***Megaceros***, 3.3 s^{-1}; ***Phaeoceros***, 4.2 s^{-1}; ***Notothylas*** 4.3 s^{-1}). ***Marchantia polymorpha*** had the highest percentage of RuBisCo per soluble protein (8%), ***Megaceros*** followed (4%), and the pyrenoid-containing species had only 3%.

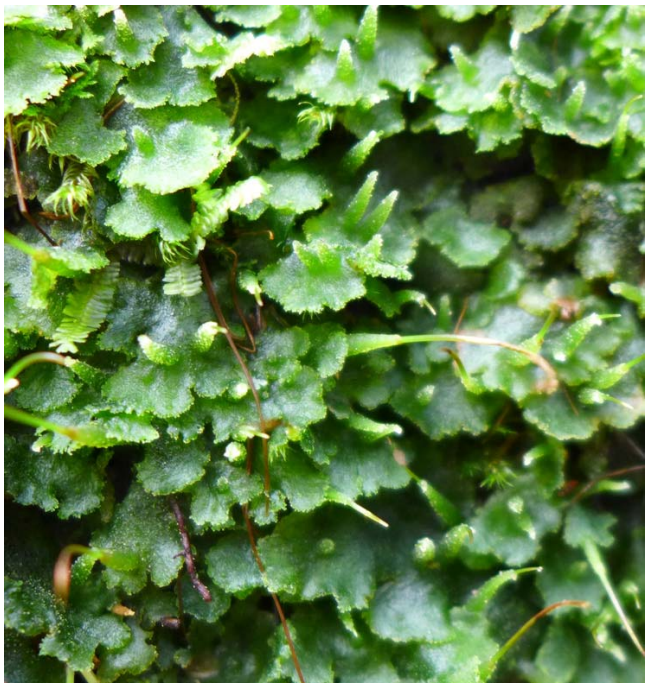

Figure 62. ***Megaceros flagellaris***, a hornwort species that lacks pyrenoids and has a lower CO_2 compensation point than does ***Marchantia polymorpha***. Photo by Scott Zona, through Creative Commons.

Figure 63. ***Notothylas orbicularis***, a hornwort species that has pyrenoids and has a CO_2 compensation point that is lower than that of ***Marchantia polymorpha*** (liverwort) or ***Megaceros*** (hornwort with no pyrenoids). Photo from One Thousand Plant Transcriptomes Initiative, through Creative Commons.

Figure 64. ***Phaeoceros carolinianus*** with capsules, a species that has pyrenoids and has a CO_2 compensation point that is lower than that of ***Marchantia polymorpha*** or ***Megaceros*** (with no pyrenoids). Photo by Hermann Schachner, through Creative Commons.

photosynthesis

Katoh (1983a) grew a cell line of ***Marchantia polymorpha*** (Figure 1-Figure 12) in suspension culture, using 1% CO_2. He found a growth rate in the exponential phase of 0.171 and a doubling time of 1.76 days. The rate of chlorophyll increase was 1.6 times higher than the growth rate. The cells reached their highest chlorophyll content at 24 mg g^{-1} dry weight in their exponential phase, with at least 60 µmol mg^{-1} chlorophyll h^{-1}.

Katoh (1983b) considered the inability of cells of ***Marchantia polymorpha*** (Figure 1-Figure 12) in suspension culture to grow in the dark to be the result of low respiration. In the light, the respiration increased to four times that in the dark. The compensation ratio (photosynthetic rate/respiration rate) was less than 1.0 during the growth period. Furthermore, these cells are unable to grow anaerobically in light in the absence of CO_2. Addition of 1% CO_2 permitted the liverwort to sustain growth. They found that at least one-third of the cellular carbon came from atmospheric CO_2.

light

Fredericq (1964) tested the influence of far-red light on thallus development in ***Marchantia polymorpha*** (Figure 1-Figure 12). Rethy *et al*. (1976) explored the effects of different light treatments on chlorophyll content in ***Marchantia polymorpha***.

Courtoy (1965-1966) experimented with light regimes on the germination and development of gemmae of ***Marchantia polymorpha*** (Figure 1-Figure 12). In artificial light of 4000 lux and 16-hour photoperiod, there were two distinct phases of growth. In the **juvenile phase**, requiring at least 15 days, light quality was unimportant. Adding sucrose in the juvenile phase reduces the phase to 5 days. In the **inductive phase**, when primordia appear, incandescent light permits development, suggesting the importance of red wavelengths.

Mache and Loiseaux (1973) found that the maximum growth rate of ***Marchantia polymorpha*** (Figure 1-Figure 12) in low light was at 2-3 x 10^3 lux, its saturation level. In optimal conditions, photosynthetic rates reach as high as 35 µM CO_2 h^{-1} mg^{-1}. High light inhibited the photosynthetic rate, with small grana in the chloroplasts and fret membranes being replaced by continuous grana.

Carter and Nickell (1967) experimented with the effects of wavelengths of light on both thallus growth and gemmae cup production. Using 16-hr light:8 hr dark at 21ºC day:13ºC night, they incubated 4 gemmae per Petri dish. After 11 weeks the controls with no colored acetate had produced a mean of 32.7 cm^2 of thalli and 83.3 gemmae cups per dish. Dishes with single-wrapped red acetate produced only 16.11 cm^2 of thalli (Figure 65) and 19.83 gemmae cups per dish (Figure 66). On the other hand, double-wrapped red dishes produced 17.5 cm^2 of thalli and 8.25 gemmae cups, suggesting that the lower light inhibited production of gemmae cups, putting more of the available resources into thalli. Those in single-layered green dishes produced 14.8 cm^2 of thalli and 8.16 gemmae cups per dish, a response consistent with the greater activity of photosynthesis in the red range. The double green, single blue, and double blue produced a mean of 10.5, 11.5, and 1.2 cm^2 of thalli respectively, but produced no gemmae cups, again supporting the importance of red light.

Aro (1982) used ***Marchantia polymorpha*** (Figure 1-Figure 12) to show that bryophytes had more chlorophyll associated with their light-harvesting protein complexes and less with reaction center complexes than did tracheophytes. Furthermore, the tracheophytes had a chlorophyll *a*:*b* ratio of 3, whereas it was only 2 in the bryophytes. These figures indicate that the bryophytes are shade plants, having proportionally more chlorophyll *b* when compared to that of tracheophyte sun plants.

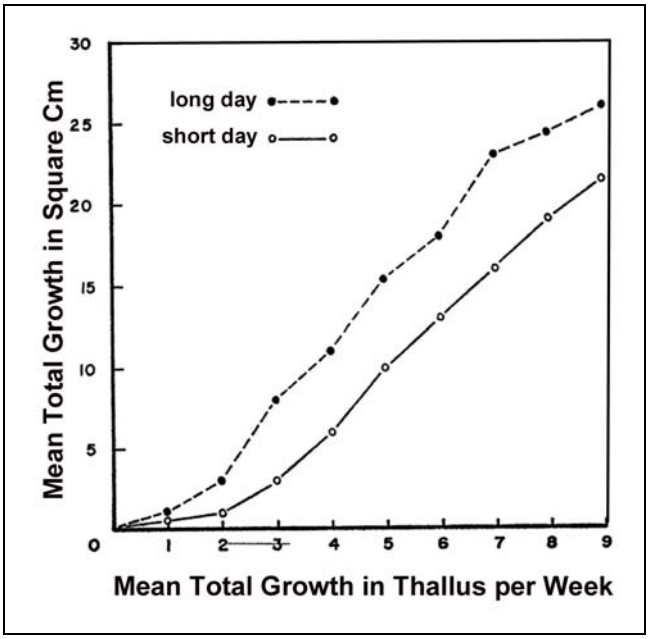

Figure 65. ***Marchantia polymorpha*** growth in long and short photoperiods. Image modified from Carter & Romine 1969.

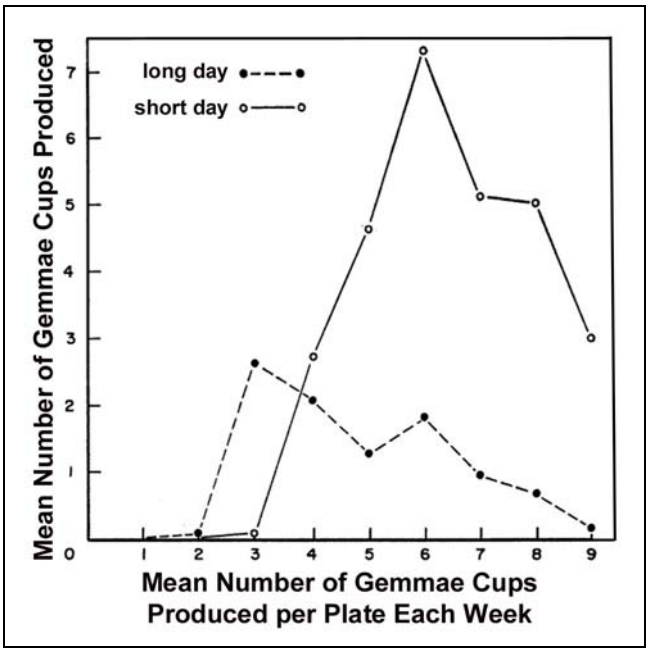

Figure 66. ***Marchantia polymorpha*** gemmae cups per week in long and short days. Modified from Carter & Romine 1969.

In further studies, Aro *et al*. (1981) determined that the plastid ultrastructure of the sun species ***Ceratodon purpureus*** (Figure 67) was characteristic of a sun plant, whereas in ***Marchantia polymorpha*** (Figure 1-Figure 12) it was characteristic of a shade plant. Nevertheless, both species exhibited photosynthetic kinetics typical of shade plants.

Figure 67. ***Ceratodon purpureus*** with young sporophytes, a sun species with plastid ultrastructure of a sun plant, but with photosynthetic kinetics of a shade plant. Photo by Claire Halpin, with permission.

Shinmen *et al*. (1991) determined that ***Marchantia polymorpha*** (Figure 1-Figure 12) produced arachidonic acid (ARA) and eicosapentaenoic acid (EPA) in cell culture. During high growth rate conditions the cells produced high quantities of both acids, with 98 mg L^{-1} of arachidonic acid and 48 mg L^{-1} of eicosapentaenoic acid. Kajikawa *et al*. (2004) isolated and characterized the genes behind the production of these acids in ***Marchantia polymorpha***. The role of arachidonic acids in cold weather has already been discussed. Eicosapentaenoic acids are known for their antifungal effects against plant pathogens (Bajpai *et al*. 2008). Both of these compounds seem to be essential in the wounding response forming volatiles in ***Marchantia polymorpha*** (Kihara *et al*. 2014). Eight-carbon volatiles form rapidly (within 40 minutes) of wounding.

Kajikawa *et al*. (2008) reported that ***Marchantia polymorpha*** (Figure 1-Figure 12) synthesizes arachidonic acid and eicosapentaenoic acid. By causing the overexpression of the involved genes, they produced 3-fold and 2-fold accumulation of these two acids, respectively. They were able to transplant these genes to tobacco and soybean, a feat that suggests that ***M. polymorpha*** can provide genes for transplantation to tracheophytes and provide them with desirable traits.

Later, Takemura *et al*. (2011) elucidated some of the physiological mechanisms involved in the observed effects of light quality and intensity on these acids. They noted that ***Marchantia polymorpha*** (Figure 1-Figure 12) synthesized both arachidonic acid (AA) and eicosapentaenoic acid (EPA), polyunsaturated fatty acids that are not known in tracheophytes. They found that the relative content of EPA to total fatty acid was highest under blue light, but that of AA did not vary. EPA content also increased under higher intensity white light. They found that 80 photon flux density μmol m^{-2} s^{-1} was the optimum intensity for both AA and EPA accumulation.

Harrer (2003) demonstrated that ***Marchantia polymorpha*** (Figure 1-Figure 12) has the same structure of the PS II-light-harvesting assembly as that of seed plants. They provided the first 3-d structure for such a large assembly by using this liverwort.

Marchantia polymorpha (Figure 1-Figure 12) was used in a study to describe the polyphasic rise of chlorophyll fluorescence at the onset of strong continuous light. Neubauer and Schreiber (1987) described the saturation characteristics and partial control by photosystem II.

In low light, bryophytes can exhibit etiolation. Ninnemann and Halbsguth (1965) elucidated the role of phytochrome in etiolation of ***Marchantia polymorpha*** (Figure 1-Figure 12). Ninnemann (1967) then described the growth substances, phytochrome, nucleic acid, and protein synthesis involved in the etiolation of the gemmae of ***Marchantia polymorpha***.

Rao *et al*. (1979) described the Hill reaction rates of three members of the Marchantiales, including ***Marchantia polymorpha*** (Figure 1-Figure 12). Using three different measures of the Hill reaction activity, they determined that the rate was lower in the three liverworts than in the seed plants tested. Furthermore, they also found lower total chlorophyll content and chlorophyll *a*:*b* ratio, all supporting the shade adaptation of these plants. They found that the greatest labelled ^{14}C occurred in the amino acids aspartate and alanine. ***Marchantia polymorpha*** exhibited higher photosynthetic rates than the other species in the test.

Maximum polyunsaturated fatty acids (PUFA) productivity is attained in ***Marchantia polymorpha*** (Figure 1-Figure 12) under low light intensity, with a photon flux density ca. 20 μmol m^{-2} s^{-1} (Chiou *et al*. 2001). Optimal inoculum size and glucose concentration for PUFA production are 8-12% and 2030 g L^{-1}, respectively. Ferrous ions, necessary components of many enzymes, promote PUFA productivity by increasing the intracellular lipid content. The highest productivities measured for PUFA were 35.0±2.1, for arachidonic acid 6.7±0.4, and for eicosapentaenoic acid 6.6±0.4 mg L^{-1} day^{-1}.

High light has different effects on ***Marchantia polymorpha*** (Figure 1-Figure 12) from those of low light (Volchenkova *et al*. 2001). It affects the area occupied by the lipid molecule. The digalactosyl diacylglycerol and phosphatidyl glycerol fractions increase significantly after high light stress, increasing from 0.50 to 0.80 nm^2 and from 0.47 to 0.63 nm^2 respectively.

red/far-red

Fredericq and de Greef (1966) examined the red:far-red control on growth and chlorophyll content in thalli of ***Marchantia polymorpha*** (Figure 1-Figure 12) grown in the light. Fredericq and de Greef (1968) followed up on these experiments by subjecting the plants to daily far-red (FR)-irradiations at the end of photoperiods of white fluorescent light. Following the first sequence, at the end of the night, the chlorophyll content of the tips of the thalli was significantly lower that that of the controls lacking the far-red treatment. Instead, upward growth (Figure 68) was beginning.

By the end of the night following the first FR-exposure, the chlorophyll content of the tips was already significantly lower than that of the controls, and there was a beginning of upward growth (Fredericq & de Greef 1968). After 8 hours in white light, those effects had become much more pronounced. After one week, a 5-minute far-red treatment at the end of the photoperiod cause a 20-30% lower chlorophyll content in the tips after the 8-hour photoperiod in white light. Following a 16-hour photoperiod, the chlorophyll was decreased by ±10%. Morphogenic effects also were less pronounced after 16 hours compared to 8 hours of white light. Basal parts of the thalli were less affected than the tips. On the other hand, a 1-hour photoperiod following far-red treatment caused much greater effects on the basal portions than on the tips. If the daily light period was diminished to 5 minutes, it caused drastic bleaching of 60-70% chlorophyll *a* loss compared to controls. Of relevance to the aquatic habitat, de Greef and Fredericq (1969) found that when the incandescent light was filtered through water for 10 minutes, it caused a somewhat weak effect. These responses are similar to those of tracheophytes and indicate that high levels of the PFR form of phytochrome are needed to maintain the horizontal growth and optimal chlorophyll content in these thalli.

Figure 68. ***Marchantia polymorpha*** exhibiting upward growth as it would occur under far-red light at the end of a period of white light. Photo by Vladimir Bryukhov, through Creative Commons.

Among the photoreceptors in ***Marchantia polymorpha*** (Figure 1-Figure 12) and other bryophytes are the phytochromes (Inoue *et al*. 2019). Phytochromes are the only known receptors for red light and far-red light and are therefore important in controlling various developmental processes. The phytochrome Mpphy regulates the formation of gametangiophores, similar to the far-red response of flowering plants. Inoue and coworkers identified the genes responsible for this regulation and demonstrated that the production of Mpphy increases when the gene is multiplied, while disappearing when the gene is deleted.

Another effect of far-red light is to accelerate senescence in ***Marchantia polymorpha*** (Figure 1-Figure 12) (de Greef & Fredericq 1972). Once again, the reversibility indicates the involvement of phytochrome. A daily exposure of only 5 minutes of red light will prevent this aging; photosynthesis plays no direct part in this response.

UV light

When photosynthetic organisms first invaded land, one of the new problems they had to deal with was their introduction to UV light (Jordan *et al*. 2016; Sancha 2017). The levels of UV reaching the Earth's surface at that time were higher than now due to the less-well developed ozone

layer. Therefore, it stands to reason that the surviving bryophytes, previously as early invaders of land, should have mechanisms to protect them from UV radiation. In seed plants, flavonoids are important in this role.

Khetwal (1985) demonstrated the presence of the flavonoid glucuronides apigenin, apigenin-7-O-glucuronide, luteolin, and luteolin-7-O-glucuronide in ***Marchantia polymorpha*** (Figure 1-Figure 12).

There has been much activity in recent years to determine the effects of increased UV light on bryophytes. Markham *et al*. (1998) found that as they increased UV-B levels, the growth rate of ***Marchantia polymorpha*** (Figure 1-Figure 12) decreased, the production of gemmae cups (Figure 69) decreased, and the proportion of dead thallus increased. Total flavonoid levels had no statistically significant change, but the ratio of luteolin to apigenin glycosides did increase (Figure 69-Figure 71). The researchers did not consider this to be a means of filtering and protecting the plants from the UV-B, but instead they experienced an improved level of antioxidant defense.

Figure 69. ***Marchantia polymorpha*** with red edges, perhaps in response to stress. Photo by Brenda Dobbs, through Creative Commons.

Figure 70. ***Marchantia polymorpha*** with red archegoniophores, perhaps responding to the stress of a cold climate in Laxarbakki, Myvatn, Iceland. Photo by Janice Glime.

Figure 71. ***Marchantia polymorpha*** with red archegoniophores, from Laxarbakki, Myvatn, Iceland, 26 July 1987. Photo by Janice Glime.

Clayton (2017) looked specifically at flavonoids in ***Marchantia polymorpha*** (Figure 1-Figure 12) exposed to UV-B radiation and determined that the flavonoids increased when UV-B radiation was enhanced. Flavones were the most predominant, with apigenin-based flavones in highest amounts and luteolin-based flavones second. The ratio shifted toward luteolin-based flavones at the higher UV-B levels. At these UV-B levels, reactive oxygen species (ROS) are produced, and the liverwort may require the luteolin flavones to scavenge these. Under low UV-B exposure, the flavone compounds accumulated in high concentrations in the epidermal layers, suggesting that they might participate in screening the UV-B. When flavone concentrations were lower, greater damage to the thallus occurred. Higher levels of flavones corresponded with greater protection and reduced thallus damage. When flavones were suppressed completely the plants became severely stunted under the UV-B treatment.

Sancha (2017) noted that bryophytes have "remarkable tolerance to UV radiation." Sancha subjected various bryophytes to enhanced UV radiation and found that all, including ***Marchantia polymorpha*** (Figure 1-Figure 12), showed increased levels of CARUVs (UV-radiation-absorbing compounds), with all being significant except for ***Anthoceros agrestis*** (Figure 72)**.**

Figure 72. ***Anthoceros agrestis***, a hornwort species with increasing, but not significant, UV-radiation-absorbing compounds with increasing of UV intensities. Photo by Hermann Schachner, through Creative Commons.

Soriano *et al*. (2019a) further examined UV damage in ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 56, Figure 60, Figure 73). They found that liverworts subjected to low photosynthetically active radiation (PAR), low PAR+ UV-A, low PAR + UV-B, low PAR + UV-A + UV-B, and high PAR exhibited no significant difference in the maximum quantum yield of PSII after 35 days. There were no changes in the chl *a*/*b* ratio and only slight changes in growth. But both chlorophylls and carotenoids decreased in content in the UV radiation treatments and even more strongly in the high-PAR treatment. The xanthophyll index (antheraxanthin + zeaxanthin) / (violaxanthin + antheraxanthin + zeaxanthin) increased only in the high-PAR (Figure 73). On the other hand, the **sclerophylly index** (ratio between thallus dry mass and surface area) increased in the UV-B-exposed treatments, suggesting a UV-induced structural protection. Only the UV-B treated liverworts exhibited DNA damage.

Figure 73. ***Marchantia polymorpha*** ssp. ***ruderalis*** showing red bases, perhaps in response to high UV levels. Photo by Malcolm Storey <www.DiscoverLife.com>, with online permission.

In further studies, Soriano *et al*. (2021) found that the developmental stage was important in determining the accumulation of UV-absorbing compounds in ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 56, Figure 60, Figure 73). They compared gemmae (Figure 69), one-month thalli, and two-month thalli after 38 days of exposure or non-exposure to UV radiation. They found that the UV responsiveness decreased with thallus age, with gemmae being the most responsive. Older thalli became progressively tougher in UV due to decreasing water content, possibly providing structural protection. Most phenolic compounds decreased with thallus age, but di-glucuronide derivatives were highest in the 1-month thalli.

Close and McArthur (2002) contend that phenolics have the primary function of protecting plants from photodamage, not from herbivores as originally thought.

Kondou *et al*. (2019) reported that MpUVR8 provides physiological benefits in UV-B resistance in ***Marchantia polymorpha*** (Figure 1-Figure 12). It is highly expressed in the apical notch (Figure 20) of the thalli and gametangiophores, including the antheridial and archegonial heads. In this species, citrine-fused MpUVR8 was translocated from the cytosol into the nucleus when exposed to increased UV-B radiation.

Ultraviolet light was a major stress to be overcome when plants first invaded land (Clayton *et al*. 2018). The ***Marchantia polymorpha*** (Figure 1-Figure 12) UVB response included many components already known from *Arabidopsis* (Figure 52), including production of UVB-absorbing flavonoids, the central activator role of ELONGATED HYPOCOTYL5 (HY5), and negative feedback regulation by REPRESSOR OF UV-B PHOTOMORPHOGENESIS1 (RUP1). Important differences included a greater importance for CHALCONE ISOMERASE-LIKE (CHIL). Mutants that disrupted the response pathway or flavonoid production were more easily damaged by UV-B than normal plants, whereas mutants that increased the flavonoid content exhibited increased UV-B tolerance.

Kondou *et al*. (2019) determined that UV-B resistance and the translocation of the UVR8 from the cytosol to the nucleus was operational in ***Marchantia polymorpha*** (Figure 1-Figure 12) in response to UV-B radiation. This series of events is highly expressed in the apical notch of the thalli and in the gametangiophores and receptacles.

fluorescence

It is widely known that chlorophyll fluoresces. The degree of fluorescence is a measure of the health of the plant. Shi *et al*. (1992) described two categories of fluorescence emission from bryophytes. ***Marchantia polymorpha*** (Figure 1-Figure 12) exhibits maximum emission around 725 nm. The fluorescence kinetics of primitive bryophyte photosynthesis, including ***M. polymorpha***, exhibited lower PS II activity, lower efficiency of primary photoconversion in PS II, and lower photosynthetic C assimilation and efficiency than did the advanced bryophyte species.

photoperiod

Photoperiod is known to control various events in the life of a plant. Differences in response to photoperiod can keep closely related species from interbreeding by bringing reproductive parts to maturity at different times (see Reproduction section below), while taking advantage of the climatic conditions at the optimum time for the event.

Marchantia polymorpha (Figure 1-Figure 12) grown in a long photoperiod (18 hours daylight) are larger and have greater dry weight than those grown in a short photoperiod (9 hours daylight) (Voth & Hamner 1940; Carter & Romine 1969). Short photoperiods favor production of gemmae (Figure 69, Figure 153-Figure 170), whereas long photoperiods favor the production of gametangiophores.

Benson-Evans (1961) found that photoperiod influences the number of gametophores, and like Carter and Romine, found that longer days (16 hrs light) result in greater thallus size and fewer gemmae cups, but a faster production of the cups.

tropisms

A little-studied area of bryophyte physiology is tropisms. Yet the ability to grow in response to the

direction of light and gravity is of considerable adaptive importance to most bryophytes.

There is a long history of studies on rhizoid tropisms in ***Marchantia polymorpha*** (Figure 1-Figure 12). Haberlandt (1889) noted **positive gravitropism** (originally known as geotropism; growing toward the gravitational pull) in the apical rhizoids of ***Marchantia polymorpha***. Weinert (1909) investigated rhizoid tropisms and growth in liverworts, including ***Marchantia***. Rawitscher (1932) reviewed the tropisms in this species. Douin (1936) reported that the thallus exhibited photogravitropism. But Miller and Voth (1962) experimented with various orientations of the thalli and found that the rhizoids would securely anchor the thalli no matter what position the plant held, contrasting with the view held by Haberlandt (1889). Perhaps this is explained by the behavior I observed in ***Fontinalis***. The rhizoids initially grow away from the plant and use a spiral growth pattern. However, once a rhizoid contacts a substrate, it branches and secures the plant to the substrate. I am not aware that this behavior has been observed in ***Marchantia polymorpha***, but such behavior has not been disproved either.

So what does a gemma cup do when its parent plant is attached to a vertical surface? Miller and Voth (1962) observed that initially the cup exhibits no tropism, growing in a perpendicular alignment with its thallus. But when it develops the achlorophyllous scale-like rim of the cup, this is negatively gravitropic, permitting the cup to become upright with respect to gravity.

Rethy *et al*. (1990) described the role of far-red illumination in tropisms of ***Marchantia polymorpha*** (Figure 1-Figure 12). It causes greater cell elongation on the ventral side of the thallus just below the apical notch, causing upward growth, whereas red light reverses the reaction.

Komatsu *et al*. (2019) demonstrated that under low light both sporelings and thalli of ***Marchantia polymorpha*** (Figure 1-Figure 12) develop narrow shapes and their apices grow toward the light source. These responses are blue-light dependent and respond to **phototropin** (photoreceptor protein; flavoproteins).

temperature

Somehow, this fleshy liverwort manages to survive winter, even when covered by snow (Figure 74). Exact responses to temperature at the cellular level have been a puzzle. ***Marchantia polymorpha*** (Figure 1-Figure 12) serves as a model system to unravel these responses (Hirano *et al*. 2022). Chloroplasts respond to cold by changing positions, optimizing photosynthesis. This response is triggered by the blue-light photoreceptor phototropin, the cold-sensing molecule. This sensor is present in the plasma membrane, cytosol, Golgi apparatus, and periphery of the chloroplast. By using genetic variants, Hirano and coworkers demonstrated that the cold response originates with the phototropin in the plasma membrane, at least in this liverwort.

Antropova (1974) included ***Marchantia*** in studies on temperature adaptations in bryophytes. He incubated the bryophytes at 10 and 20ºC for 72 hours. But this period of time does not influence either thermostability or cold resistance. A treatment of 3 hours at superoptimal temperatures does cause an increase in thermostability, but no change in cold resistance. The behavior of the bryophytes was similar to that of flowering plants but differed from the temperature acclimation of algae.

Figure 74. ***Marchantia polymorpha*** in snow; note how dry the thallus appears, a condition that reduces damage from interior ice crystals. Photo by Vladimir Teplouhov, through Creative Commons.

But experiments by Weis *et al*. (1986) differed. Using ***Marchantia polymorpha*** (Figure 1-Figure 12) and other thallose liverworts, they found that high temperature treatment elicited a reversible depression of photosynthesis. The time required to achieve complete recovery depended on the extent of the heat damage. With severe heat treatment, PS II was damaged and inactivation of photosynthesis was irreversible. Unlike Antropova, Weis and coworkers found that exposure of these thallose liverworts to high sublethal temperatures did not result in the significant increase in heat stability of the photosynthetic apparatus as had been seen in seed plants. They interpreted this to mean that the heat hardening capacity of water-loving liverworts was extremely low.

Fletcher (1982) found no frost damage to ***Marchantia polymorpha*** (Figure 1-Figure 12) in cultivation in New Zealand populations. Some of the other species of thallose liverworts became severely bleached or blackened in greenhouse cultivation down to -5.5ºC. The ***M. polymorpha***, on the other hand, remained a healthy green all winter.

Response to heat appears to be more complicated. It involves several subcellular compartments as well as multi-level regulatory networks (Marchetti *et al*. 2021). Studies on ***Marchantia polymorpha*** (Figure 1-Figure 12) indicate that the core components of the response are conserved from bryophytes to flowering plants.

Temperature affects the relative production of fatty acids in ***Marchantia polymorpha*** (Figure 1-Figure 12) (Saruwatari *et al*. 1999). At 25°C this liverwort contained approximately 18% linolenic acid (18:3ω3), 11% arachidonic acid (20:4ω6) and 3% eicosapentaenoic acid (20:5ω3) as percentages of total fatty acids. At 15ºC, the ratios of linolenic acid and arachidonic acid increased greatly, with less effect on the other acids. Arachidonic acid and eicosapentaenoic acid increased in the chloroplast but not elsewhere in the cell. Linolenic acid increased in

both fractions. Various galactolipids were present in one or the other or both compartments, but only monogalactosyldiacylglycerol and chloroplastic phosphatidylcholine increased in low temperatures. Gellerman *et al.* (1972) found that ***Marchantia polymorpha*** had arachidonic acid in all tissues, ranging 10-30% of the total fatty acids.

Takemura *et al*. (2012) likewise reported the production of arachidonic acid and eicosapentaenoic acid in ***Marchantia polymorpha*** (Figure 1-Figure 12), noting that neither is produced in tracheophytes. The accumulation of ω-3 polyunsaturated fatty acids increased significantly as the temperature decreased. At 5ºC the concentration was approximately 3x that at 15ºC. ω-6 polyunsaturated fatty acids, on the other hand, decreased at low temperatures.

Akter *et al*. (2011) acknowledged the role of ABA in both desiccation tolerance and freezing. This dual role is not surprising since one of the dangers of freezing is desiccation as ice crystals draw water out of the cell or make the water unavailable as ice. Akter and coworkers found that isolated gemmae (Figure 153-Figure 170) of ***Marchantia polymorpha*** (Figure 1-Figure 12) responded to increased ABA by increasing the sucrose concentration. These treated gemmae survived freezing, whereas most of the controls did not. The best survival occurred with 5% sucrose and 19 µM ABA.

Takeuchi *et al*. (1980) even found a successful method for freezing ***Marchantia polymorpha*** (Figure 1-Figure 12) for cryopreservation.

The movement of organelles in response to stress is seldom discussed. In response to a cold treatment, ***Marchantia polymorpha*** (Figure 1-Figure 12) sporelings and gemmalings nuclei and peroxisomes relocated from the **periclinal cell wall** (wall parallel to surface of meristem or surface of organ; Figure 75) to the **anticlinal cell wall** (wall arranged perpendicular to surface of plant body; anticlinal division results in formation of anticlinal walls between daughter cells, enabling tissue to increase circumference, thus keeping pace with any increase in girth of organ; Figure 75) (Ogasawara *et al*. 2013). Mitochondria, on the other hand, did not relocate.

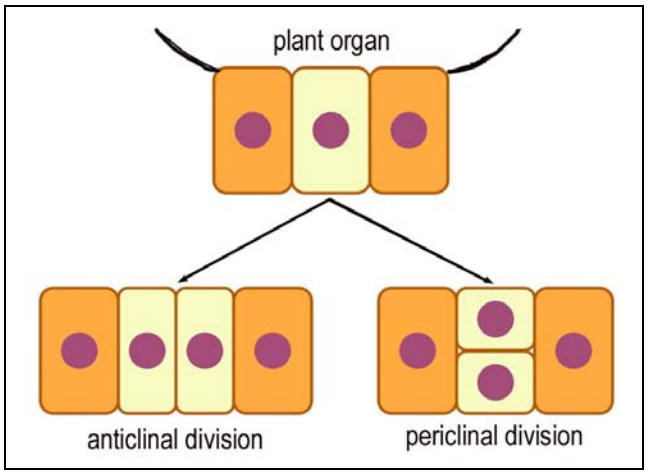

Figure 75. Comparison of anticlinal and periclinal divisions. Drawing modified from The Science of Plants, Meristem Morphology, Open Library <open.lib.umn.edu>.

senescence

Sometimes it seems like bryophytes never die. They just keep growing at the top while lower portions remain and may even decay. But **senescence** (process of aging) does indeed occur. Stanislaus and Maravolo (1994) examined cell factors that influenced senescence. They found light-induced senescence in young, middle-aged, and old tissues of ***Marchantia polymorpha*** (Figure 1-Figure 12) all exhibited suppressed senescence in treatments with spermine, spermidine, and putrescine. Not surprisingly, ethylene induced senescence in these tissue ages, an effect that could be retarded by putrescine, a compound they tentatively identified in ***M. polymorpha*** extracts.

LaBelle *et al*. (1997) identified two isophosphatases in senescing ***Marchantia polymorpha*** (Figure 1-Figure 12) thalli. They found that spermine reduced total phosphatase activity by 135-175%. Ethylene, on the other hand, reduced it by 133-155%. When ethylene and spermine were added together, they decreased the activity by 120-163% compared to the levels with the ethylene treatment alone. LaBelle and coworkers interpreted these findings to indicate that phosphatase and ethylene are associated with senescence. Spermine, on the other hand, functions in preventing senescence.

It may seem inconceivable to a bryologist, but ***Marchantia polymorpha*** (Figure 1-Figure 12) is not always a welcome visitor in a garden (Figure 76). Altland *et al*. (2008) found that quinoclamine can serve as both a PRE and POST activity herbicide at 4-6 mg L^{-1}. It could inhibit PRE germination in gemmae (Figure 153-Figure 170), but its ability to stay on the substrate made it increase the efficacy of other kinds of treatments.

Aging brings a variety of responses in plants. Maravolo (1976) found that transport, a function especially of the midrib, was inhibited by aging, cinnamic acid, and ethylene in ***Marchantia polymorpha*** (Figure 1-Figure 12).

Seaman *et al*. (2005) examined the role of spermine in programmed cell death of ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that **PKC** (protein kinase C) concentrations in untreated young thalli were higher than those of older tissues. **PKC** refers to a family of protein kinase enzymes involved in a variety of signal transduction pathways (Blumberg 1991). PKC enzymes play important roles in several signal transduction cascades. When Seaman and coworkers treated older tissues with spermine, they also had higher cytosolic putative PKC concentrations than even the young untreated thalli. The spermine also resulted in higher total protein levels than untreated tissues. Hence, it appears that the spermine causes both qualitative and quantitative decreases in senescence.

One of the consequences of spermine is to reduce DNA fragmentation, as demonstrated in ***Marchantia polymorpha*** (Figure 1-Figure 12) (Pagoria & Maravolo 2005). The fragmentation is localized near the lower epidermis of the apical meristem. From there it progresses into the mesophyll cells. However, when these plants were treated with 100 µM spermine, they exhibited significantly lower (p<0.001) levels of DNA fragmentation in aged (2-5 cm) tissues.

Figure 76. ***Marchantia polymorpha*** females in garden in Houghton, Michigan, USA. So far we have been unable to find any males. This illustrates the density it can reach. Fortunately, the owners find these plants fascinating, permitting their invasion. The yellow patches are ***Brachythecium*** cf. ***salebrosum***. Photo courtesy of Craig Waddell.

genetics

Marchantia polymorpha (Figure 1-Figure 12) is typically considered a model for the early terrestrial colonizing plants. With this in mind, Bowman *et al*. (2017) characterized the genome of this species as having low genetic redundancy in most of its regulatory pathways. This species differs from its purported charophycean ancestors by encoding novel biochemical pathways, new phytohormone signalling pathways (especially auxin), expanded repertoires of signalling pathways, and increased diversity of transcription factor families. It sheds light on the evolution of haploid sex chromosomes as they occur in a dioicous plant. The haploid condition makes gene transfer and subsequent study easier than in the diploid tracheophytes (Chiyoda *et al*. 2008).

Ikeuchi and Inoue (1988) used a computer-assisted homology search to identify the D1-D2-cytochrome b-559 complex protein region in the chloroplast genome of ***Marchantia polymorpha*** (Figure 1-Figure 12).

Takenaka *et al*. (2000) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) had at least 1-4 copies of the hpt gene, an example of gene redundancy in early land plants.

Chung *et al*. (2006a) compared two bryophytes and *Arabidopsis thaliana* (Figure 52) to determine the number of genes in common. They found 79% of the genes expressed by ***Marchantia polymorpha*** (Figure 1-Figure 12) were also expressed in the moss ***Physcomitrium patens*** (Figure 77). They found 763 genes expressed not only in both bryophytes, but also in *Arabidopsis thaliana*. Another 363 genes were found in the bryophytes, but not in *Arabidopsis*.

Figure 77. ***Physcomitrium patens***, a species that expresses 79% of the same genes as in ***Marchantia polymorpha***. Photo through public domain.

Chung *et al*. (2006b) contrasted gene expression in ***Marchantia polymorpha*** (Figure 1-Figure 12) with that of *Arabidopsis thaliana* (Figure 52 In the latter tracheophyte species, ~50% of the expressed genes exhibited cell-type-specific expression patterns. On the other hand, in ***M. polymorpha*** the expression in cultured cells did not differ from those of the thalli. Instead, 110 genes were expressed in cultured cells of ***M. polymorpha***, but not in those of *A. thaliana*, whereas in the 10 *A. thaliana* genes checked, they were expressed in whole plants of both species, but not in cultured cells of *A. thaliana*. Thus, ***Marchantia polymorpha*** with transplanted genes can be used more easily to determine the expression of tracheophyte genes.

Lin and Bowman (2018) identified micro RNAs in ***Marchantia polymorpha*** (Figure 1-Figure 12). Tsuboyama *et al*. (2018) elaborated on methods of using the model liverwort ***Marchantia polymorpha*** in AgarTrap transformation for studying genetic transformation, achieving a 97% transformation efficiency.

Schmid *et al*. (2018) found that methylation pattern of DNA changes in cytosines varies significantly during the life cycle. These coincide with four major epigenetic states, corresponding to the states of vegetative gametophytes, antherozoids, archegonia, and sporophytes. They concluded that epigenetic reprogramming occurs in at least two events during the life cycle, once in each generation. These events occur in parallel with the differences in the gene expression involved in DNA methylation.

Ishizaki *et al*. (2008) used ***Agrobacterium*** (Figure 20)-mediated transformations on immature thalli of ***Marchantia polymorpha*** (Figure 1-Figure 12) that had developed from spores. Plants grown from gemmae (Figure 153-Figure 170) of these plants all expressed the introduced gene GUS. Because of the haploid state of the

thallus, these plants offer a very useful system for such transformations.

Marchantia polymorpha (Figure 1-Figure 12) has become a model organism for using promoters in overexpression studies to determine gene functions (Althoff *et al*. 2014). The protocol developed has the potential to screen large numbers of transgenic plants, including the use of knock-down mutants.

These studies have shown the usefulness of ***Marchantia polymorpha*** to test the function of genes.

One of the applications of our genetic knowledge of this species is to determine its susceptibility to radiation damage. Using Co^{60} gamma rays, Miller and Sparrow (1965) determined that the ability of the two apical cells to reproduce was inhibited at doses less than the lethality dosage. The thallus exhibits different radiosensitivity from that of the gemmae (Figure 153-Figure 170) when based on energy absorption; the thallus is 4.3 times as sensitive as the gemmae.

Adaptations

Halbsguth (1953) explored the development of dorsiventrality in ***Marchantia polymorpha*** (Figure 1-Figure 12). We can assume that this body form has advantages, particularly in habitats that can at times be very wet and at other times can dry out. The overlapping thalli of the colony help to retain water in the soil (Figure 78). This same advantage can be accomplished by growing with mosses (Figure 79).

Figure 78. ***Marchantia polymorpha*** in Houghton, Michigan, showing overlapping thalli. Photo by Matt Tianen, with permission.

Bischler and Jovet-Ast (1981) commented that members of the **Marchantiales** seem to have some characters that are not essential for survival, reproduction, or dispersal. Others seem to be disadvantageous, but they have not prevented the continued existence of these traits. Instead, they concluded, the adaptations to their niches are linked primarily to biochemical and biophysical properties of the cell content rather than to morphological expressions.

Since wet habitat species typically experience dry seasons, among the most common adaptations are those that conserve water. McConaha (1941) described the ventral structures that affect water uptake and conservation. In the Marchantiales, these include smooth and tuberculate rhizoids (Figure 80-Figure 84) and ventral scales (Figure 83-Figure 86). Using several members of the order, including ***Marchantia polymorpha*** (Figure 1-Figure 12), McConaha reported that the **smooth rhizoids** (Figure 80) emerge from the scales and can make contact with the substrate (Figure 81-Figure 82). **Tuberculate (pegged) rhizoids** (Figure 53, Figure 83) serve a different purpose. They originate beneath the scales, forming numerous connected capillary strands that lie parallel to the thallus (Figure 53, Figure 83). This arrangement provides a rapid capillary distribution of water to all the absorptive areas of the thallus. The efficiency in water balance depends on the form and imbrication of the scales (Figure 84-Figure 86), as well as with the length and number of these rhizoids.

Figure 79. ***Marchantia polymorpha*** with gemmae cups, overgrowing mosses that can help to retain moisture in the soil and the liverwort thallus. Photo by Janice Glime.

Figure 80. ***Marchantia polymorpha*** ventral side showing smooth rhizoids along midrib. Photo from Botany Website, UBC, with permission.

Figure 81. ***Marchantia polymorpha*** archegoniophores and thallus showing brown, perpendicular rhizoids. Photo by Janice Glime.

Figure 82. ***Marchantia polymorpha*** ventral smooth rhizoids at midregion of thallus, along the midrib. Photo by Larry Jensen, with permission.

Figure 83. ***Marchantia polymorpha*** bundles of pegged rhizoids terminating in marginal scales. Photo by Larry Jensen, with permission.

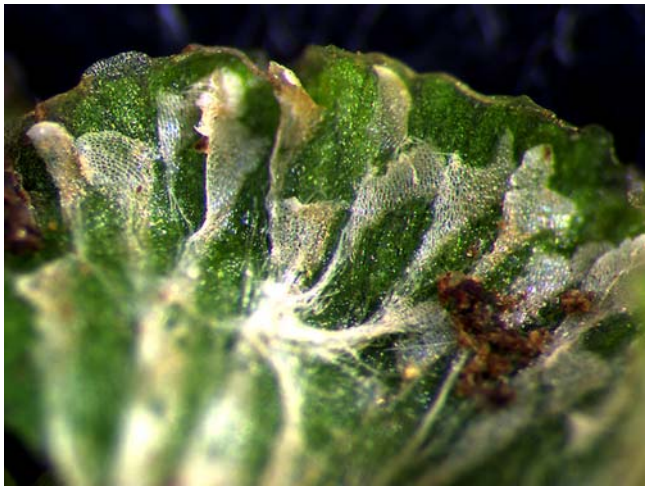

Figure 84. ***Marchantia polymorpha*** ventral growing tip of thallus with marginal scales. Photo by Larry Jensen, with permission.

Figure 85. ***Marchantia polymorpha*** ssp. ***ruderalis*** scales showing purple coloring. Photo by Des Callaghan, with permission.

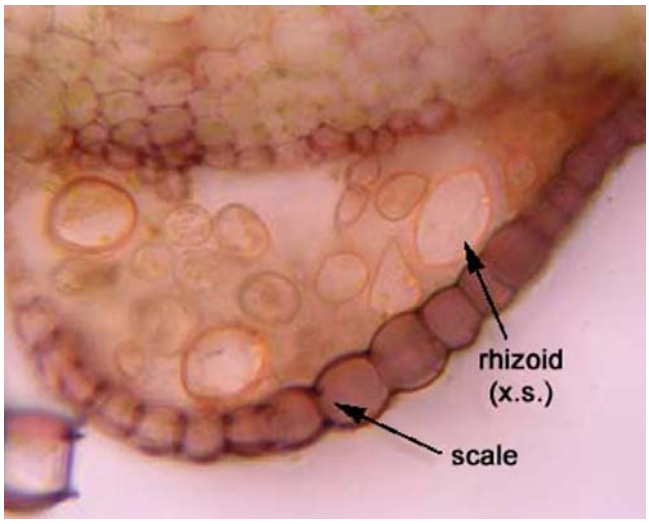

Figure 86. ***Marchantia polymorpha*** section showing scale and rhizoids on ventral surface. Photo from Botany Website, UBC, with permission.

Cao *et al*. (2014) further described the rhizoids (Figure 80-Figure 84) in ***Marchantia polymorpha*** (Figure 1-Figure 12). These researchers noted that the tuberculate (pegged) rhizoids converge toward the midrib (Figure 83-Figure 84). The smooth rhizoids occur in clusters in the free portions near the midrib at the thallus posterior (Figure 80, Figure 82). Unlike the tuberculate rhizoids, these lie perpendicular to the thallus, always growing toward the moist soil (Figure 81-Figure 82).

Many botany textbooks still claim that bryophytes lack cuticles. But we now know that this is not the case, at least for many bryophytes. Many species develop a thin cuticle that is not easily noticed. Brockington *et al*. (2013) and Xu *et al*. (2021) noted that even in flowering plants we lack understanding of the cuticle genetics and the role of the cuticle in evolution. Hence, these researchers have provided a detailed description of the cuticle and its genetic origins in ***Marchantia polymorpha*** (Figure 1-Figure 12). The cuticle is hydrophobic and is generally considered to be a barrier between the plant and the atmosphere wherein it helps to maintain internal moisture levels and to prevent entry of potentially pathogenic organisms. It is also a filter of UV radiation and barrier against mechanical damage. Because of these important roles in the terrestrial environment, it has been considered to be one of the key innovations needed for colonization of land (Corner 1964).

Xu *et al*. (2021) analyzed the role of the cuticle in preventing water loss in ***M. polymorpha*** (Figure 1-Figure 12). Using mutant plants, they found no change in morphology of the thallus for plants without a cuticle. However, they found more effects of desiccation in mutant plants with no cuticle. These plants frequently exhibited brownish tissues at the flank and tip of the thallus after five days with no cover and no added water. Most significantly, water content declined to about 70%, compared to 90% in non-mutant plants (Figure 87), the latter being only a 5% decrease in water content following the drying regime. Wu and coworkers were unable to detect any waxes in the cuticle of the lab-grown ***M. polymorpha***. Rather, the cuticle of ***M. polymorpha*** in these experiments consisted only of cutin, except for the waxy cuticle surrounding the pores (Figure 88). We know that environmental conditions affect the manufacture of cuticle waxes in flowering plants and could have been a cause for suppression of these waxes in the lab populations of ***M. polymorpha***.

While the plants with the addition of cuticles solved the problem of water loss, they had created another problem. An epidermis with a cuticle also interferes with gas exchange, impeding photosynthesis by the underlying photosynthetic tissue. In a thallose liverwort like ***Marchantia polymorpha*** (Figure 1-Figure 12) chloroplasts are buried (Figure 89, Figure 91-Figure 92) below the epidermis where light is reduced and it would be difficult to exchange CO_2 and O_2 if they did not have air pores (Figure 89-Figure 90). These openings permit the entry of air into the chambers (Figure 89, Figure 91-Figure 92) beneath them and the escape of the photosynthetic O_2. But these air pores present another problem. When the thallus becomes submersed or even when raindrops land in the pores, there is the danger that the water could enter the thallus and prevent the gas exchange. ***Marchantia polymorpha*** protects itself from this internal drowning by having cuticular ridges (Figure 90-Figure 94) around the opening of the pores (Schönherr & Ziegler 1975). The hydrophobic waxes, helped by the cohesive properties of water, repel the water and prevent its entry. Schönherr and Ziegler considered them to be "perfect structures."

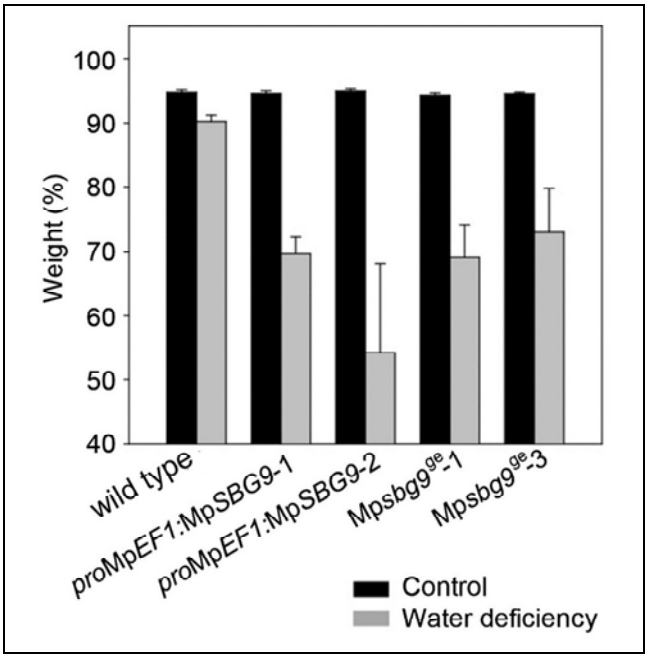

Figure 87. Cuticle water loss as percent weight in ***Marchantia polymorpha*** in wild type and four different mutants that reduce cuticle formation. Modified from Xu *et al*. 2021.

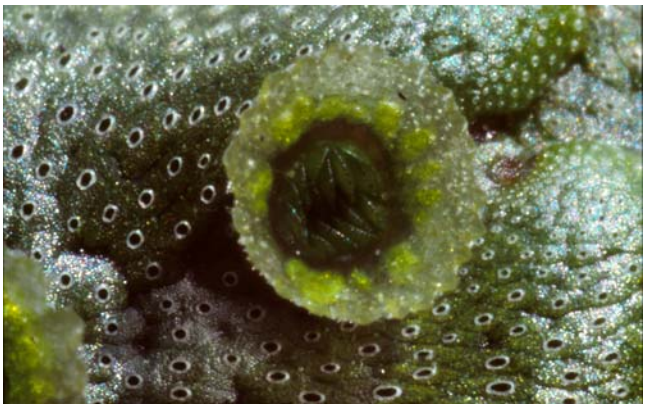

Figure 88. ***Marchantia polymorpha*** gemma cup and distinct air pores surrounded by white cuticle on thallus. Photo by John Forlonge, through Flickr.

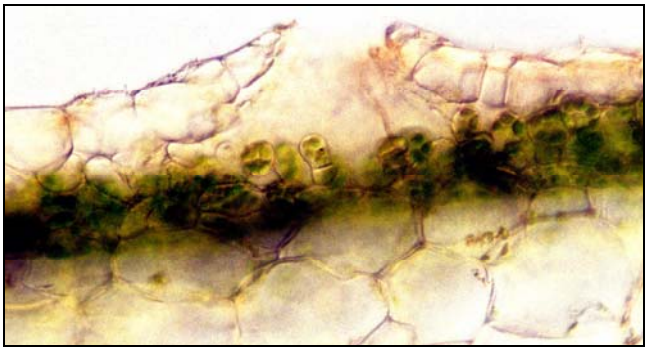

Figure 89. ***Marchantia*** thallus section showing pore opening and layer of chlorophyllous filaments. Photo by George Shepherd, with permission.

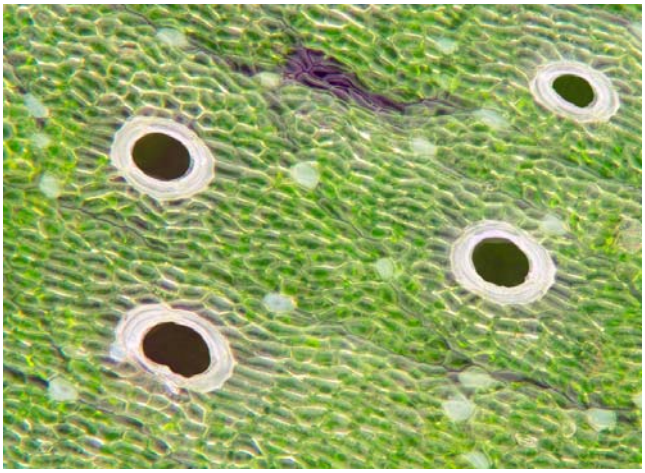

Figure 90. ***Marchantia polymorpha*** air pores and green layers showing through surface. Note the donut-shaped cuticular ridges. Photo by Des Callaghan, through Creative Commons.

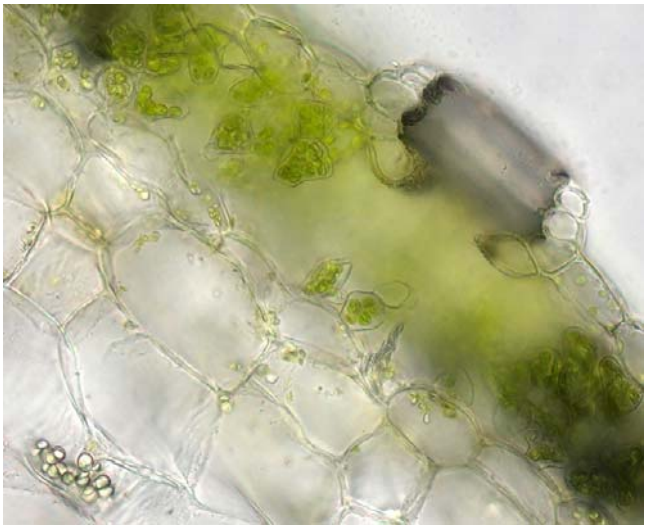

Figure 91. ***Marchantia polymorpha*** section through pore opening. Note the photosynthetic cells beneath the pore. Photo by Walter Obermayer, with permission.

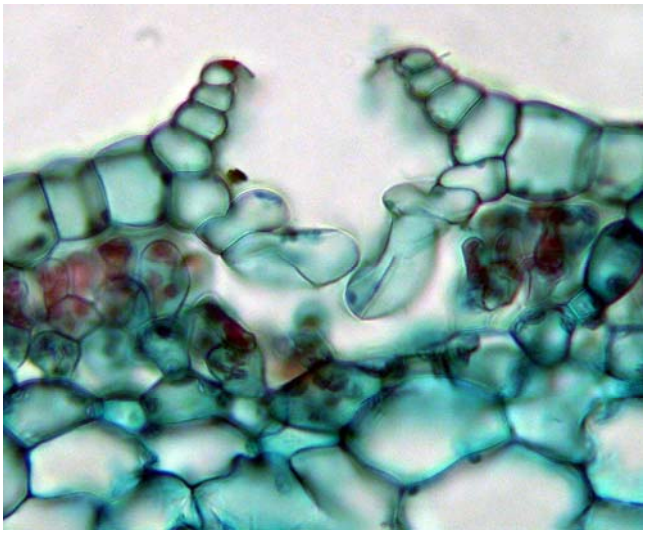

Figure 92. ***Marchantia*** thallus section showing pore opening. Photo by George Shepherd, with permission.

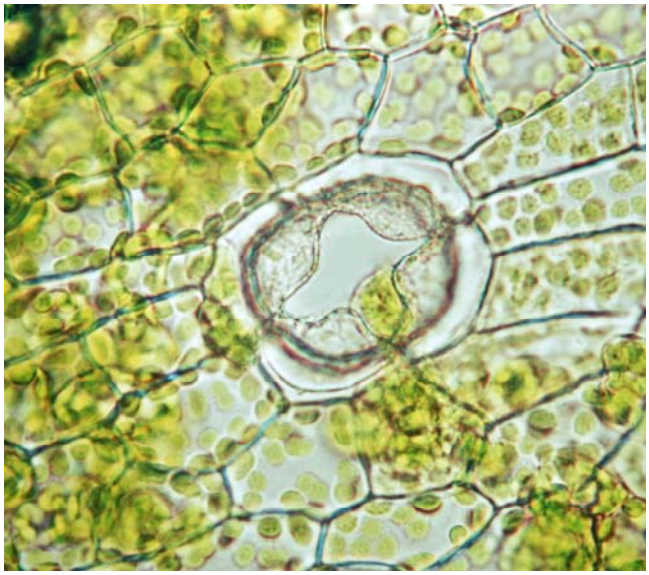

Figure 93. ***Marchantia polymorpha*** pore opening as seen from thallus surface, showing cuticular ridge surrounding the opening. Photo by Wilhelm Barthlott, with permission.

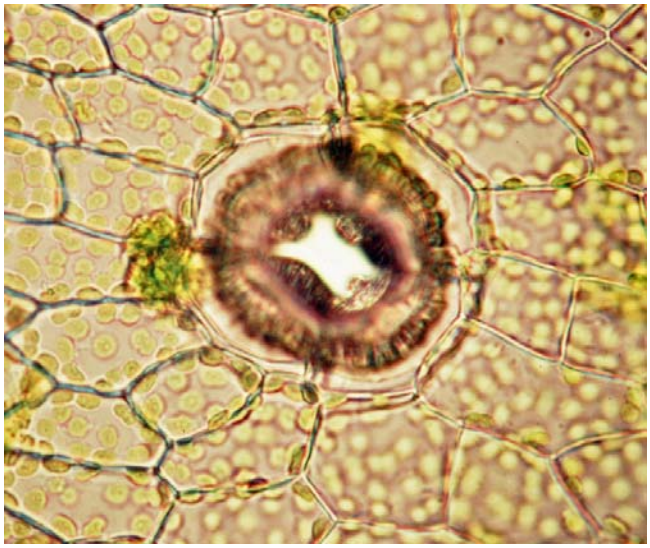

Figure 94. ***Marchantia polymorpha*** pore opening with cuticular ridge and small opening. Photo by Wilhelm Barthlott, with permission.

The adaptive value of oil bodies puzzled bryologists for a long time. Galatis and Apostolakos (1976) described the association of the microbodies with the cytoplasmic tubules of oil body cells in ***Marchantia***. They found that the tubules increase in number at the stage when the cells are actively synthesizing oil.

One of the protections exhibited by liverworts is the ability to store sesquiterpenes and bisbibenzyls in oil bodies (Figure 95). Suire *et al*. (2000) isolated a number of isoprenoid biosynthetic enzymes from oil bodies (Figure 95-Figure 96) in ***Marchantia polymorpha*** (Figure 1-Figure 12). In ***Marchantia polymorpha*** these oil bodies are localized in oil body cells (Tanaka *et al*. 2016). Tanaka *et al*. (2016) reported that oil bodies served as sites of accumulation of sesquiterpenoids and marchantin A. They also observed that the number of oil body cells increased in thalli grown in low-mineral conditions.

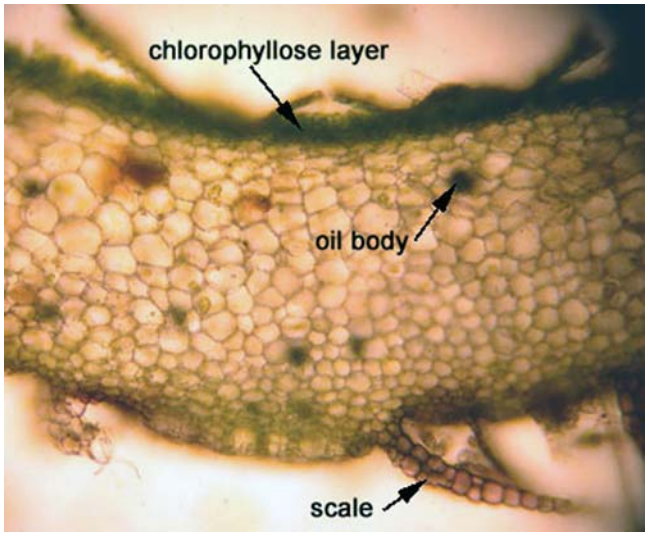

Figure 95. ***Marchantia polymorpha*** section showing scales and oil bodies. Photo from Botany Website, UBC, with permission.

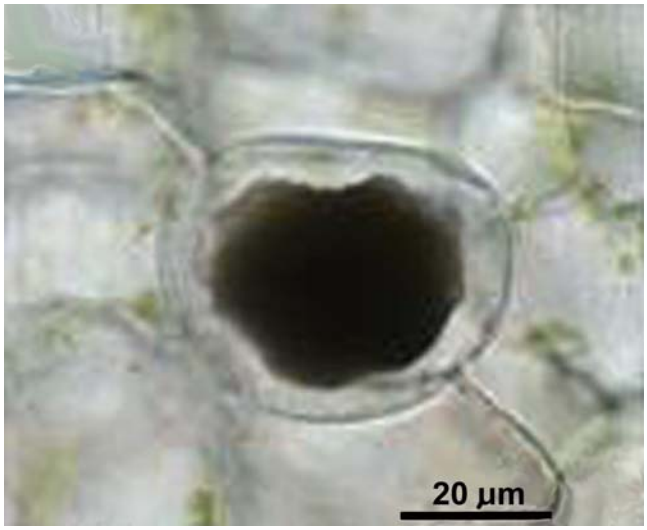

Figure 96. ***Marchantia polymorpha*** oil body in oil cell. Photo by Masaki Shimamura, with permission.

He *et al*. (2013) described the liverwort oil bodies (Figure 95-Figure 96) as "intracellular organelles bounded by a single unit membrane containing lipophilic globules suspended in a proteinaceous matrix." Oil bodies are unique to liverworts. In ***Marchantia polymorpha*** (Figure 1-Figure 12), we know that they contain protein complex that is immunologically related to the plastid and cytosolic enzymes of the isoprenoid synthesis. They are known as sites of essential oil accumulation and sequestration. Although they are known to contain compounds useful for medical purposes, their full function in liverworts remains unknown. What are the advantages of size and morphology – characters that differ sufficiently among species to be of taxonomic value?

By suppressing the genes controlling this sesquiterpenoid production, Romani *et al*. (2020) found that the terpenoid-rich oil bodies (Figure 95-Figure 96) are responsible for protecting ***Marchantia polymorpha*** (Figure 1-Figure 12) from arthropod herbivores, but that unlike those in tracheophytes, these oil bodies seem to have no role in abiotic stress response, including desiccation. Takizawa *et al*. (2021) found that when ***Marchantia polymorpha*** was exposed to non-axenic conditions the number of oil bodies increased, as did the amounts of sesquiterpenes. They likewise demonstrated that the bacterium ***Escherichia coli*** (Figure 16) elicited the same response.

Kihara *et al*. 2014) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) emitted C8 volatiles following mechanical wounding. Induction of these emissions occurred within 40 minutes of the wounding. When transgenic plants lacking arachidonic acid and eicosapentaenoic acid were wounded, only minimal C8 volatiles were detectable. Octan-3-one was produced only minimally when thalli were completely disrupted, but was the most abundant product in only partially disrupted thalli.

Yoshikawa *et al*. (2018) traced the wounding response in ***Marchantia polymorpha*** (Figure 1-Figure 12). Wounding of the thallus resulted in the synthesis of phenylpropanoids, including luteolin, apigenin, and isoriccardin C.

Watson (1919) suggests that reduced pores in ***Marchantia*** species might be an advantage in a wet habitat. Due to cohesion of the water molecules, water is unable to enter the smaller openings.

Among the advantages that bryophytes have are their plasticity and adaptability. Plasticity is exhibited by the various biochemical responses to different pathogens and environmental conditions. Adaptability is enhanced by the haploid condition. Selection on gametophyte plants is more rapid than in tracheophytes because there is only one set of chromosomes, permitting rapid removal of non-adapted genes in the population. This mechanism is evident in adaptations to heavy metals. Briggs (1972) demonstrated this in the response of ***Marchantia polymorpha*** (Figure 1-Figure 12) to lead contamination. When Briggs compared plants from areas with high levels of lead in the soil to those from an area with low levels of lead pollution, those from highly contaminated soil were highly tolerant of lead, whereas those from areas with less lead contamination are more sensitive. Krupinska (1976) found that lead tetraethyl causes distorted growth patterns in ***Marchantia polymorpha***. The thalli become "profusely" branched, a reversible phenomenon. The chloroplasts degenerate and growth of the spores and gemmae is inhibited.

Reproduction

sexual

Marchantia polymorpha has been used in a number of studies on sexual expression in plants, particularly to demonstrate that expression in early plants. Nagai *et al*. (1999) generated 970 expressed sequence tag (EST) clones from an immature female sexual organ (Figure 112-Figure 128) of the liverwort ***Marchantia polymorpha*** (Figure 1-Figure 12). In 376 ESTs they found 123 redundant groups, reducing the unique sequences to 717.

Marchantia polymorpha (Figure 1-Figure 12) is a **dioicous** species with easily recognizable differences between **antheridiophores** (Figure 98-Figure 110) and **archegoniophores** (Figure 110-Figure 128). Durand (1908) described these as well as the sporangium. The

stages of the sexual life cycle can be seen in Figure 98-Figure 146.

As noted earlier, sex in ***Marchantia*** is determined by a **small V chromosome** in males and no U chromosome, whereas the female has a **single U chromosome** and no V chromosome (Lorbeer 1934). But the designation of the gender is not perfectly genetic. Naidu (1973) reported abnormal receptacles that bore both archegonia and antheridia, based on specimens from a population in India. The V chromosome (Figure 97) has several chromosome-specific sequence elements (Okada *et al*. 2001; Ishizaki *et al*. 2002). Okada *et al*. (2000) identified 70 male-specific PAC clones and verified that the V chromosome exhibits unique sequences that are not present on the U chromosome or any non-sex chromosomes. These repeat sequences contribute 2-3 Mb on the V chromosome. Okada and coworkers introduced us to the first active V chromosome-specific gene known in plants. Fujisawa *et al*. (2001) isolated two female-specific and six male-specific DNA fragments that originated from these U and V chromosomes. Okada *et al*. (2000) suggested that this liverwort was a suitable model for identifying roles of sex genes in sexual differentiation. Bisang *et al*. (2010) noted that molecular sex markers have thus far only been described for a few bryophytes, one of which is ***Marchantia polymorpha***.

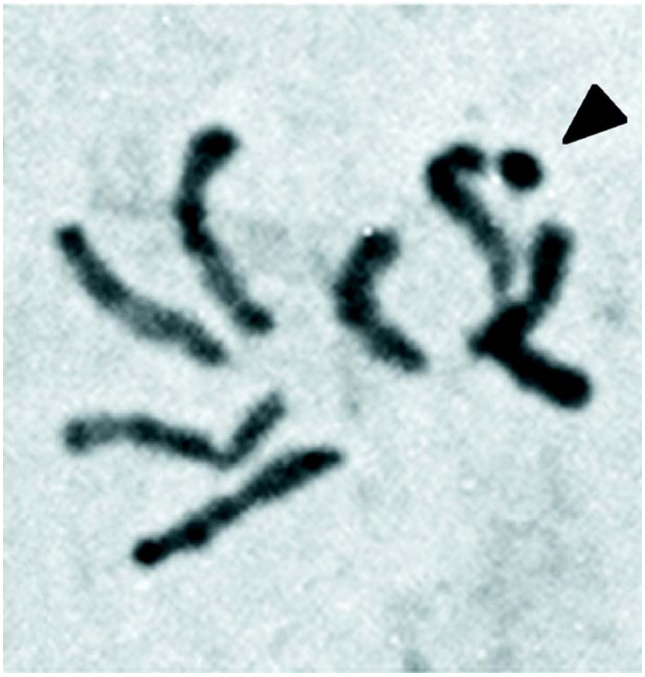

Figure 97. ***Marchantia polymorpha*** male chromosomes, with tiny V chromosome indicated by arrow. Photo from Okada *et al.* 2001, through Creative Commons.

Wann (1925) determined that photoperiod was important in initiating sexual branches in ***Marchantia polymorpha*** (Figure 1-Figure 12). In long days, males produce mature antheridiophores in 3-4 weeks. Females seem to respond to the same stimulus, but require 6-8 weeks for archegoniophores to reach maturity. The role of temperature remained unknown. High humidity hastens the sexual branches, but low humidity retards them and may completely inhibit their production, particularly for archegoniophores. And as in some of the algae, low nitrogen relative to carbohydrate may initiate sexual branches. These nutrient relationships in algae signal the end of the growing season and the beginning of the unfavorable conditions of winter. The same could be likely for the bryophytes as the tracheophytes use up the nitrogen during the growing season. Hence, forming a sexual reproductive structure that can remain dormant until favorable conditions return is adaptive.

Lloyd and Steinmetz (1937) found that high temperatures (above 30ºC) at least temporarily suppress development of archegoniophores and antheridiophores in ***Marchantia polymorpha*** (Figure 1-Figure 12). Low temperatures, on the other hand, promote development of these reproductive structures during short days. But during late autumn, plants moved to a warm greenhouse from the cool outdoors would also produce more archegoniophores and antheridiophores compared to plants that had remained in the greenhouse continuously. Furthermore, plants brought to the greenhouse in late autumn developed these sexual structures more quickly than those brought into the greenhouse earlier in the autumn. Greenhouse-grown plants could be induced to develop archegoniophores and antheridiophores during short days if they were exposed to natural autumn conditions found at Orono, Maine, USA. Nevertheless, a long photoperiod is important in inducing this sexual response.

Benson-Evans (1964) found that ***Marchantia polymorpha*** (Figure 1-Figure 12) produced gametangia at 21ºC in long days (18 hours), but lowering the temperature to 10ºC or the photoperiod to 6 hours resulted in no gametangia production.

Yamaoka *et al*. (2021) summarized that initiation of the development of gametangia depends on environmental factors such as light, but they considered that these factors are still elusive. They recognized recent studies that considered their development to use conserved regulatory modules that are involved in light signalling.

Maravolo *et al*. (1967) explored activity of 12 enzyme systems in various parts of ***Marchantia polymorpha*** (Figure 1-Figure 12). Of these, only phosphatases, esterases, and peroxidases were found in extracts of uninduced thalli, induced thalli, stalks, and antheridiophore and archegoniophore disks. They found an amplification of esterases in the antheridia (Figure 105-Figure 109). These esterases can hydrolyze particular esters into acids and alcohols or phenols. Gorska-Brylass (1970) reported increased esterase activity in the early stages of spermatogenesis in ***Marchantia polymorpha***, an activity that declines near the end of that cellular division. Could these be important in protecting the antheridia against stresses, especially desiccation?

Markham and Porter (1978) isolated an aurone (known for making flowers yellow) from ***Marchantia polymorpha*** (Figure 1-Figure 12) during its sexual phase. The aurone aureusidin 6-O-glucuronide is present only in the antheridiophores (Figure 98-Figure 110).

Figure 98. ***Marchantia polymorpha*** males with antheridial receptacles (antheridial heads). Photo by Li Zhang, with permission.

Figure 99. ***Marchantia polymorpha*** with expanding antheridiophores, showing development of the antheridial receptacle before elongation of the stalk. Photo by Des Callahan, with permission.

Figure 100. ***Marchantia polymorpha*** ssp. ***polymorpha*** male with antheridiophores that look healthy, despite the curling of the thallus. Photo by David Holyoak, with permission.

Figure 101. ***Marchantia polymorpha*** antheridiophores reaching full elongation. Photo by Walter Obermayer, with permission.

Figure 102. ***Marchantia polymorpha*** mature antheridial heads, showing how dense they can be. The presence of only one gender suggests that this is a clone. Photo by Steve Juntikka, with permission.

Figure 103, ***Marchantia polymorpha*** antheridial head in side view, with rhizoids and scales hanging from the head. Photo from Botany Website, UBC, with permission.

Figure 104. ***Marchantia polymorpha*** antheridial head that is not quite mature. Photo by Walter Obermayer, with permission.

Figure 105. ***Marchantia polymorpha*** mature antheridial receptacle showing yellow antheridia. Photo by Larry Jensen, with permission.

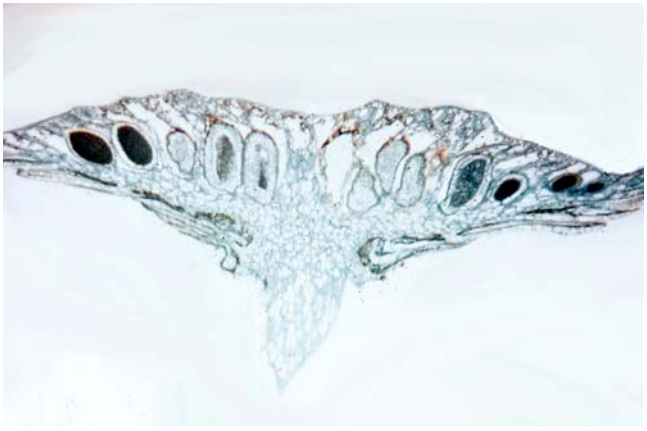

Figure 106. ***Marchantia polymorpha*** antheridial receptacle section showing arrangement of antheridia. Photo by Janice Glime.

Figure 107. ***Marchantia polymorpha*** antheridium section with developing sperm cells. Photo by Janice Glime.

Michelot-Gernez (1984) described the nuclear condensation during spermatogenesis in ***Marchantia polymorpha*** (Figure 1-Figure 12). Reynolds and Wolfe (1984) identified protamines in plant sperm, using ***Marchantia polymorpha*** as one of the representative organisms. These are small, arginine-rich proteins that replace histones near the end of the haploid phase of spermatogenesis; they are considered essential for sperm head condensation and DNA stabilization.

Carothers and Kreitner (1968) described the blepharoplast of the **spermatid** (developing spermatozoid) of ***Marchantia polymorpha*** (Figure 1-Figure 12). Bajon *et al*. (1995) described the nucleus of the **spermatozoid** (male gamete; sperm). They found that RNAs remain scattered in the spermatozoid throughout differentiation. They are closely associated with chromatin strands that fuse in the mature gamete. They found that mRNAs associated with the mature spermatozoid genome are stored mRNAs. They permit the transfer of paternal information to the zygote during fertilization. Using a high-speed video technique, Inouye and Hori (1991) described the movement of the sperm (spermatozoid) of ***Marchantia polymorpha*** as a breast stroke. Miyamura *et al*. (2002) further described the flagellar movement of the sperm, using high-speed video.

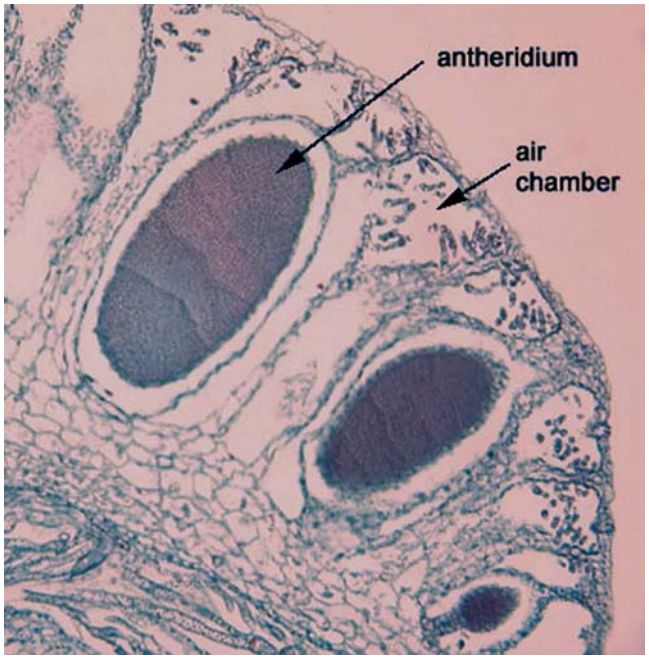

Figure 108. ***Marchantia polymorpha*** section of antheridial head. Photo from Botany website, UBC, with permission.

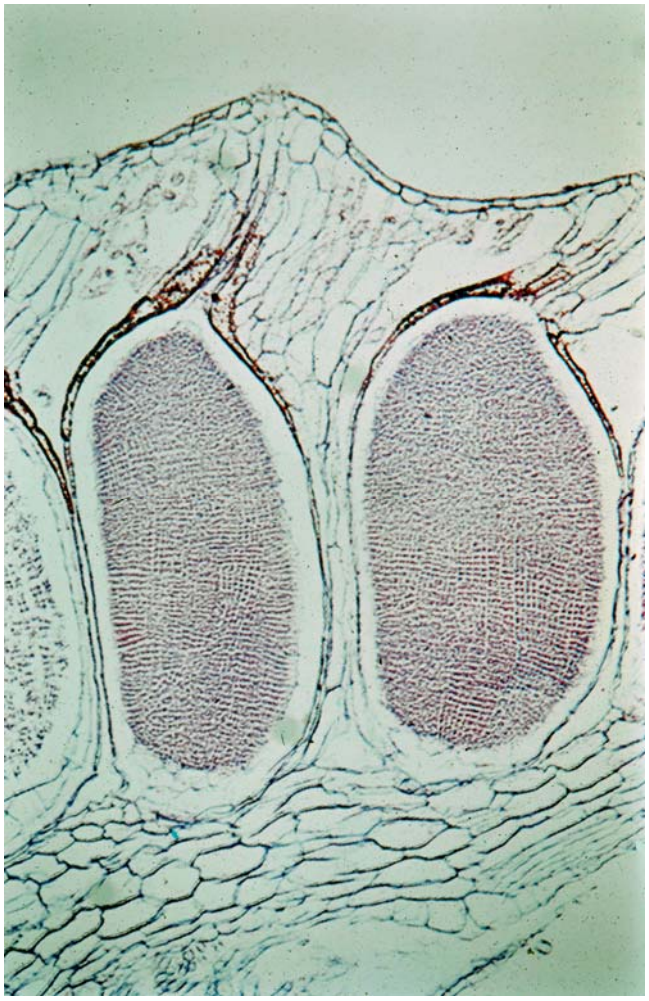

Figure 109. ***Marchantia polymorpha*** section of antheridia at maturity. Photo by Wilhelm Barthlott, with permission.

Figure 110. ***Marchantia polymorpha*** antheridiophores and archegoniophores, occurring on separate thalli. Photo by Robert Klips, with permission.

Une (1984) observed that female plants produce sexual branches more frequently than do male plants (Figure 111). Archegoniophores, in particular, may be inhibited by low humidity (Wann 1925). As in many algae, a low nitrogen:high carbohydrate ratio can stimulate the formation of sexual branches. This nutrient relationship often serves as a signal that growing conditions are declining and the formation of spores provides a mechanism for surviving until favorable conditions return or the spores land in a suitable habitat.

Figure 111. ***Marchantia polymorpha*** females in July 1982, in Rydhave, Denmark, showing how dense the population can become.

The archegoniophores (Figure 112-Figure 121) arise from a separate thallus from that of the antheridiophores. The archegoniophores are formed by an infolding and rolling of the thallus, trapping rhizoids and scales inside the stalk that is thus formed (Figure 118-Figure 121). The rhizoids, in particular, aid in the movement of water to the receptacle head at the top of the stalk.

Figure 112. ***Marchantia polymorpha*** archegonial heads before the elongation of the stalk. Photo by Rudolf Macek, with permission.

Figure 113. ***Marchantia polymorpha*** nearly mature archegoniophores before the arms of the receptacle spread. Photo from <www.aphotofauna.com>, with permission.

Figure 114. ***Marchantia polymorpha*** archegoniophores before the fingers spread. Photo from <www.aphotofauna.com>, with permission.

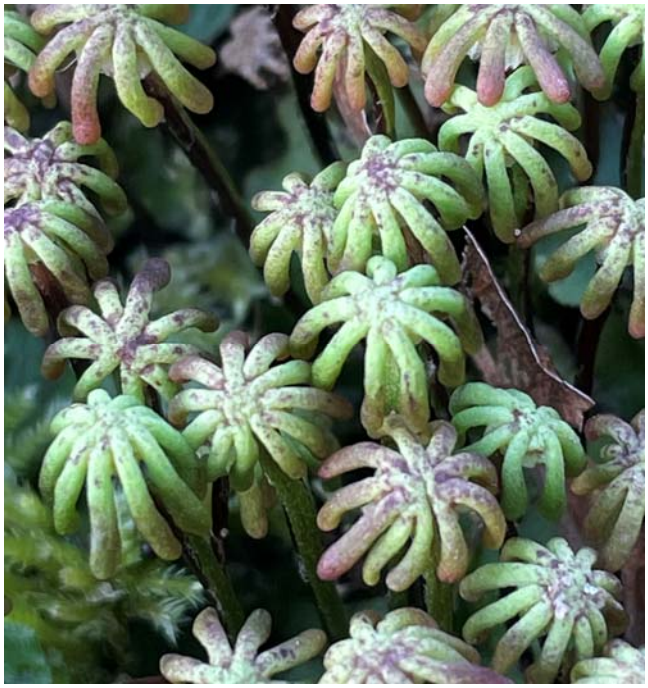

Figure 115. ***Marchantia polymorpha*** females before the fingers are uplifted. Photo by Craig Waddell, with permission.

Figure 116. ***Marchantia polymorpha*** archegoniophores with fully expanded fingers on the receptacle on 1 July 2009 in Michigan, USA. Photo by Janice Glime.

Figure 117. ***Marchantia polymorpha*** females in what appears to be a purely female clone in Houghton, Michigan, USA. Note the different stages of old and young archegoniophores. Photo courtesy of Craig Waddell.

The archegonial head, at maturity, is filled with scales that protrude from the fingers of the structure (Figure 118-Figure 122). These scales help to conserve water in the head and offer protection to the developing sporophyte.

Figure 118. Newly emerging and maturing archegoniophores of ***Marchantia polymorpha***. Note the rhizoids along the stalk and the scales protruding from under the receptacle head. Photo copyright Stuart Dunlop <www.donegal-wildlife.blogspot.com>, with permission.

Figure 119. ***Marchantia polymorpha*** mature archegoniophores 1 July 2009 in Michigan, USA. Photo by Janice Glime.

Figure 120. ***Marchantia polymorpha*** archegoniophores showing numerous scales hanging from the receptacle and groove in stalk where the rolled edges meet. Photo by Janice Glime.

Figure 121. ***Marchantia polymorpha*** archegoniophores nearing maturity. Note the rhizoids at the left. Photo by Rudolf Macek, with permission.

Figure 122. ***Marchantia polymorpha*** archegoniophore showing rhizoids along stalk, from Tahquamenon Falls, MI. Photo by Janice Glime.

The archegonia form on the fingers with the oldest near the stalk (Figure 123-Figure 129). Maintenance of dormancy by the egg (Figure 124-Figure 125, Figure 130) can prolong the period of time in which fertilization is possible. In ***Marchantia polymorpha*** (Figure 1-Figure 12), MpRKD regulates gametophyte development and keeps the egg cell dormant until fertilization occurs (Rövekamp *et al*. 2016). By doing this, it also prevents **parthenogenesis** (development of a zygote without fertilization).

In 1974, Zinsmeister and Carothers (1974) elucidated details of the fine structure changes involved in egg (Figure 124-Figure 125, Figure 130) formation. An amorphous substance surrounds the egg, perhaps preventing desiccation and protecting the egg.

Figure 123. ***Marchantia polymorpha*** archegoniophore with developing archegonia; showing thallus nature of the receptacle. Photo by George Shepherd, through Creative Commons.

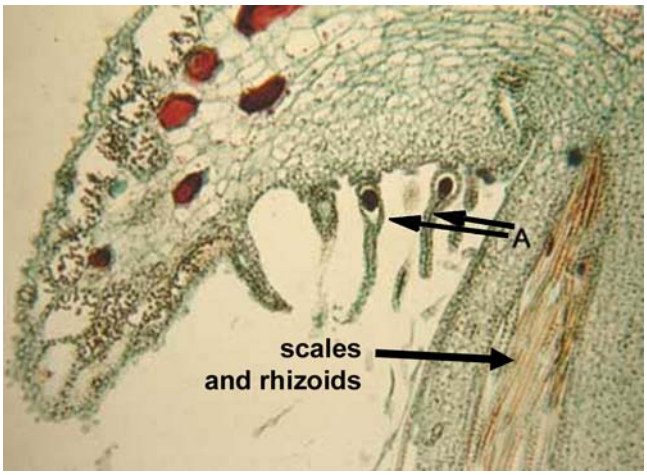

Figure 124. ***Marchantia polymorpha*** archegonial head longitudinal section showing archegonia (**A**) and scales and rhizoids in stalk. Image modified from Botany Website, UBC, with permission.

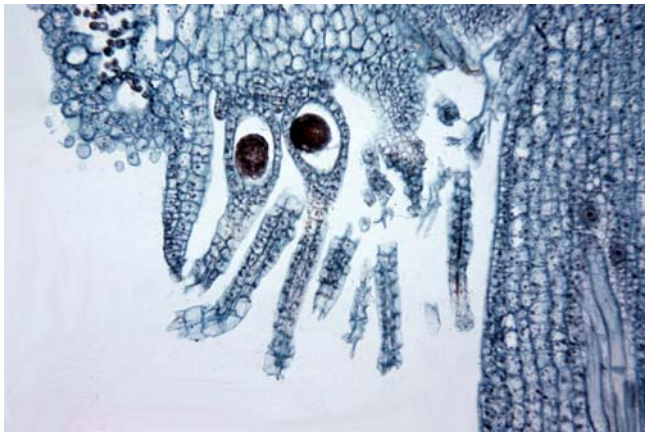

Figure 125. ***Marchantia polymorpha*** archegonia with what appear to be zygotes. Note the rhizoids within the stalk. Photo by Janice Glime.

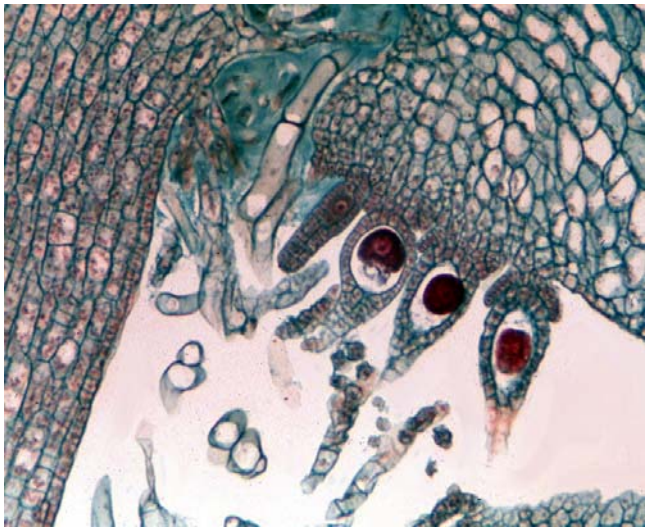

Figure 126. ***Marchantia polymorpha*** archegonia in various stages of development with least mature being closest to the stalk; the one furthest away has a zygote. Photo by Janice Glime.

Figure 127. ***Marchantia polymorpha*** with very young archegoniophores before stalk has expanded fully, mixed with mature archegoniophores showing yellow sporangia and elaters. Photo by Janice Glime.

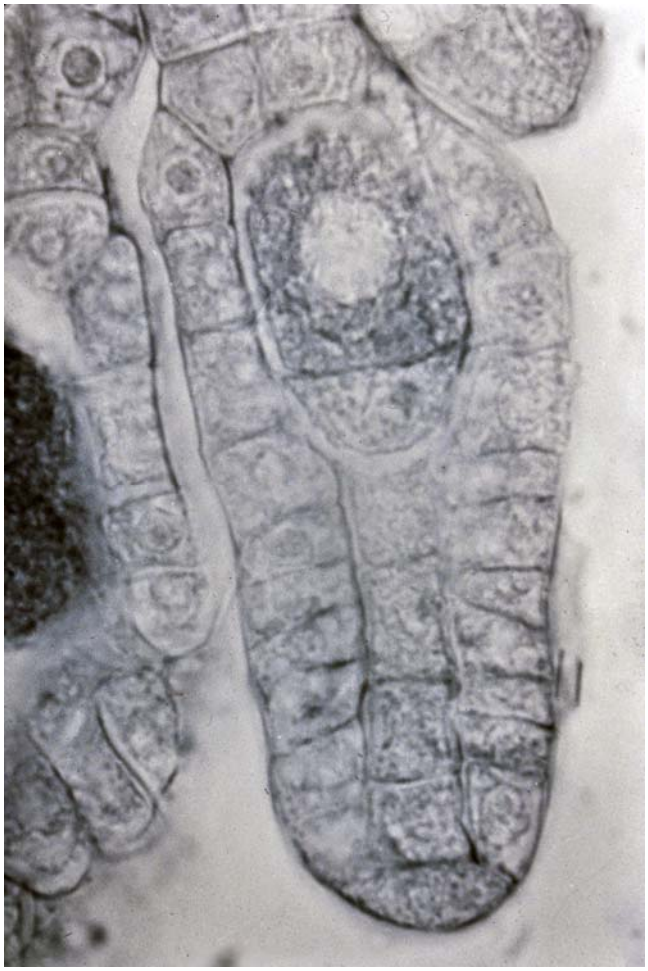

Figure 128. ***Marchantia polymorpha*** unfertilized archegonium before neck canal cells disintegrate. Photo by Wilhelm Barthlott, with permission.

Figure 129. ***Marchantia polymorpha*** archegoniophores in a female clone on 6 July 2018 in Houghton, Michigan, USA; males were nearby in a separate clone. Photo by Janice Glime.

Part of this curiosity was to understand how the sperm could reach the egg in these dioicous plants. Kitagawa (1985) noted that due to the dioicous nature of ***Marchantia polymorpha*** (Figure 1-Figure 12), the male and female must be near each other (Figure 110) for fertilization to occur (Figure 130). But just how near is near?

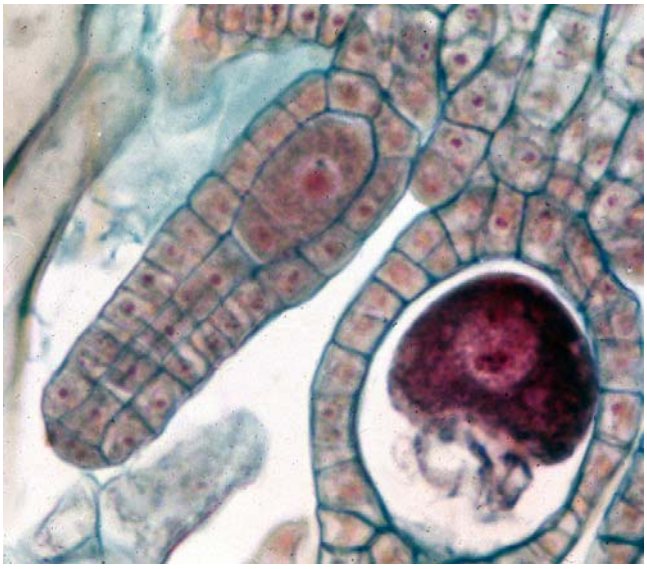

Figure 130. ***Marchantia polymorpha*** archegonia; one on left is immature with neck canal cells still in place; one on right has neck canal cells disintegrated (not shown) and the sperm is uniting with the egg. Photo by Janice Glime.

Strasburger (1869, in Parihar 1961) described the splashing of **sperm** (=male gamete, antherozoid, or spermatozoid; Figure 131-Figure 132) from the surface of the antheridial receptacle to the surface of the archegonial receptacle at a time when the antheridiophore was taller than the archegoniophore, thus permitting the water droplets to travel downward to the archegonial receptacles. He considered the splashing to extend to about 65 cm. By this time archegonia would have developed on the lower surface and the water would flow over the edge of the archegonial receptacle to reach them.

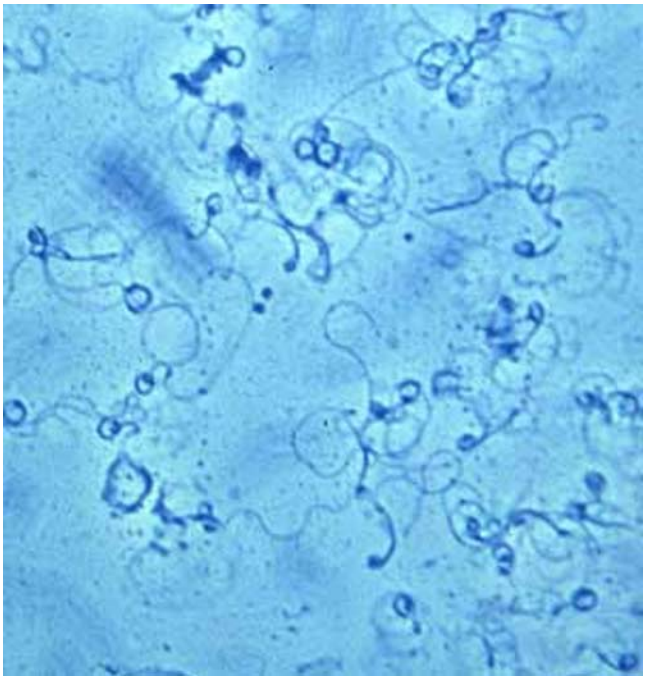

Figure 131. ***Marchantia polymorpha*** sperm. Photo from Botany Website, UBC, with permission.

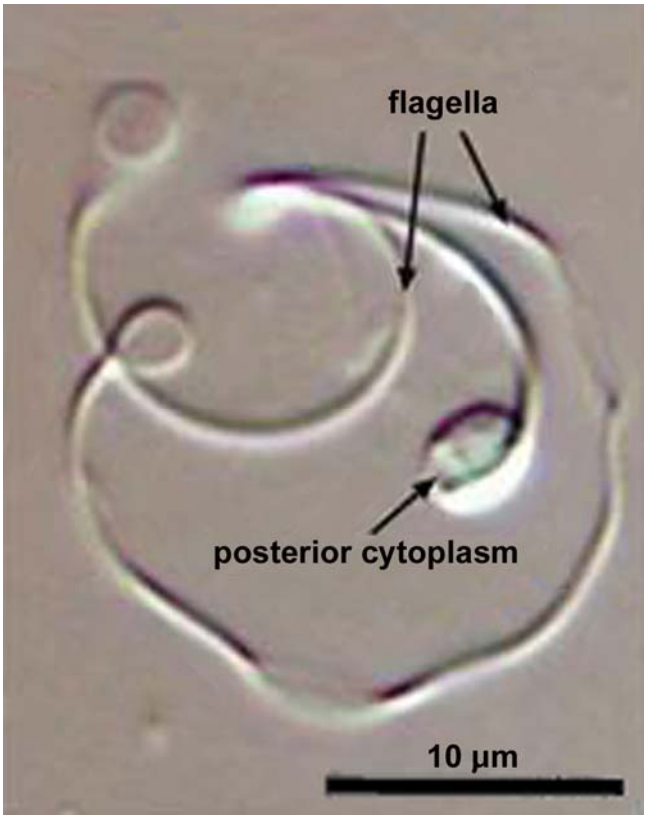

Figure 132. ***Marchantia polymorpha*** sperm. Photo by Masaki Shimamura, with permission.

One of the puzzles in fertilization of plants is how the sperm find the eggs, or at least the archegonium. Lidforss (1904) was among the early researchers on this question. Again using ***Marchantia polymorpha*** (Figure 1-Figure 12), Lidforss found the proteins albumin, hemoglobin, and diastase could each attract the sperm into the capillary tube that held them.

Åkerman (1910) experimented with various salts as attractants or repellants of sperm (Figure 131-Figure 132) in ***Marchantia***, possibly ***M. polymorpha*** (Figure 1-Figure 12). Some salts had positive effects on chemotaxis, whereas others, especially heavy metals, had negative effects.

Furuichi and Matsuura (2016) found time-dependent changes in sperm motility (Figure 131-Figure 132) from high to low motility states in ***Marchantia polymorpha*** (Figure 1-Figure 12). Based on the average lifetime of the high motility state and the speed of movement, they estimated that these sperm would travel less than 3 cm. Hence, they concluded that other factors were needed to explain apparent travel distances greater than this, and that the motility of the sperm itself most likely only was important in the final fertilization step.

Alvarez (2017) described the movement of the sperm (Figure 131-Figure 132) in ***Marchantia polymorpha*** (Figure 1-Figure 12), based on the publication of Myamura *et al*. (2002). The movement caused by these biflagellated sperm is waveform and differs between the two flagella. This permits the sperm to adjust both steering and propulsion. The beat of the posterior flagellum is more 3-d than that of the anterior flagellum. When the sperm collides with an obstacle, it does not exhibit backward swimming.

Shimamura (2016) reported that water droplets could splash sperm cells (Figure 131-Figure 132) 30 cm or farther from the male plants of ***Marchantia polymorpha*** (Figure 1-Figure 12), as previously demonstrated experimentally (Burgeff 1943; Brodie 1951; Duckett & Pressel 2009). Drops of dye-containing water dropped onto the antheridia did limited splashing and most of the dye (>90%) was absorbed by the ventral side of the antheridial receptacles. But the dye also quickly moved to the ground level and managed to spread throughout the entire colony within an hour. This was facilitated by the bundles of rhizoids in the archegoniophore. When encountering a female plant, the dye moved up the archegoniophore stalk to the archegonial receptacle through the bundles of rhizoids enclosed by the stalk. This movement upward required 30-60 minutes. Furthermore, Duckett and Pressel (2009) observed that the youngest sporophytes are located near the stalk and the older ones are located near the periphery, indicating that fertilization continues after stalk elongation.

But Pressel and Duckett (2019) also measured the distances travelled by the sperm (Figure 131-Figure 132) of ***Marchantia polymorpha*** (Figure 1-Figure 12) to achieve fertilization. They followed more than 80,000 males and females for two years after a major fire and recorded the number of sporophytes. While these numbers seem high, they found the astounding number of more than 200,000 sperm in individual antheridia of ***Marchantia polymorpha*** (Figure 1-Figure 12). This is a greater number than in most bryophytes, but it is coupled with very effective sperm dispersal. They found that distances could exceed 20 m and that dispersal resulted in 100% fertilization of the female plants. The dehiscing antheridia release lipids that help to move the sperm in the surface water films both along the antheridiophores and across the surface water films to the archegoniophores. In a single flooding event, a male thallus with 10-12 antheridiophores can release more than 50 million sperm. This high fertilization success, coupled with the numerous tiny spores, can account for the ease with which the species seems to arrive after disturbances such as fire.

sporangia

Fertilization occurs in the archegonia, making possible a number of sporangia on the same archegonial head. The embryos (Figure 133-Figure 134) remain in the arms of the archegonial head where they are protected by many scales. The maturing embryo forms a foot, seta, and capsule inside the archegonium (Figure 135-Figure 136). The scales protrude more as the embryo matures (Figure 137-Figure 146), thus helping to maintain moisture.

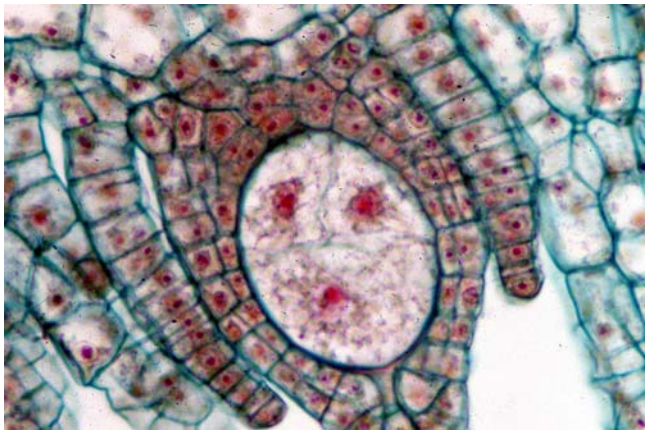

Figure 133. ***Marchantia polymorpha*** archegonium with young sporophyte embryo. Photo by Janice Glime.

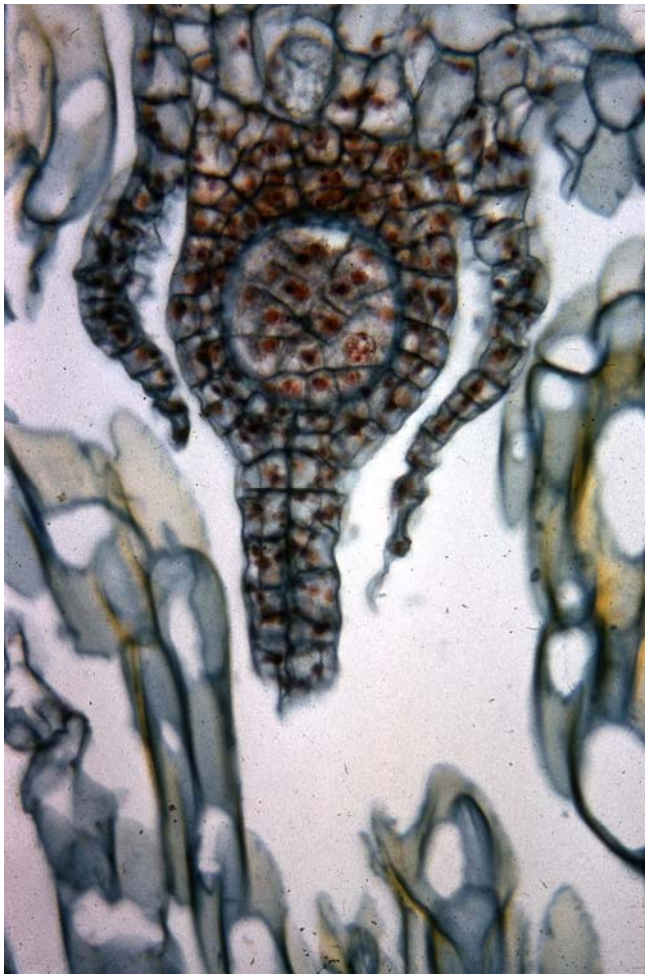

Figure 134. ***Marchantia polymorpha*** archegonium with young embryo. Photo by Janice Glime.

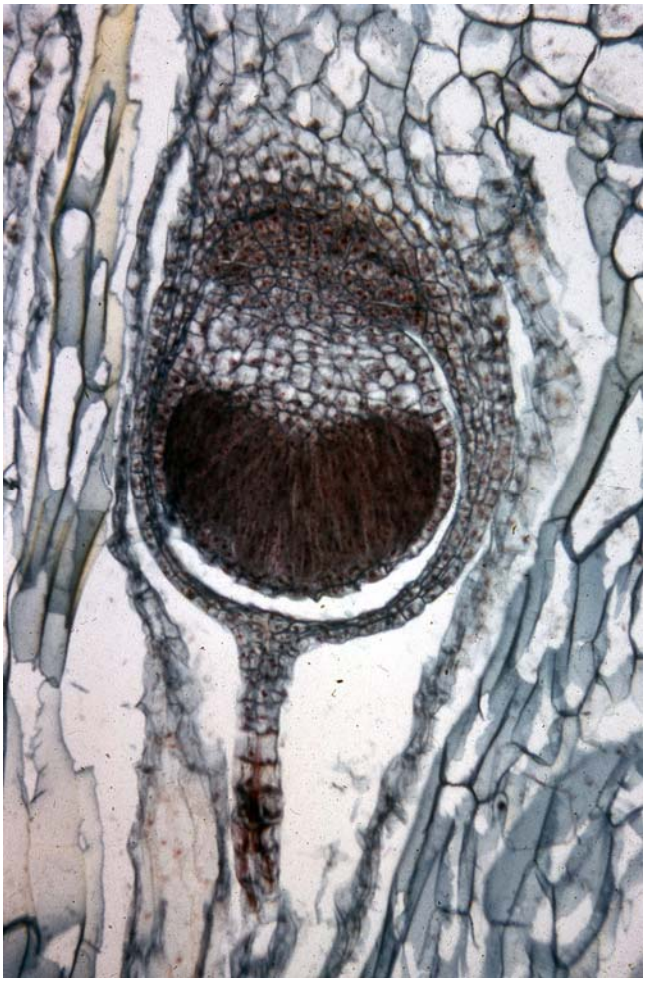

Figure 135. ***Marchantia polymorpha*** archegonium with young sporophyte showing foot, seta, and capsule (sporangium). Photo by Janice Glime.

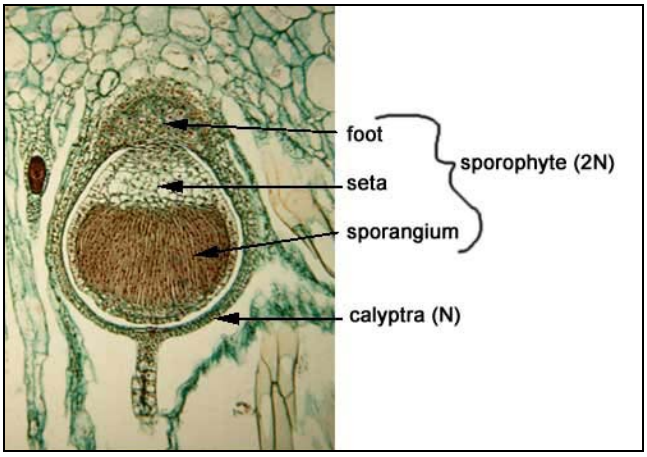

Figure 136. ***Marchantia polymorpha*** capsule longitudinal section. Image modified from Botany Website, UBC, with permission.

Figure 137. ***Marchantia polymorpha*** females with sporophytes, forming a female clone in Myvatn, Iceland, 8 August 1987. Photo by Janice Glime.

Figure 138. ***Marchantia polymorpha*** mature archegoniophores, Keweenaw Peninsula, Michigan, USA. Photo by Janice Glime.

Wann (1925) found that sporophytes (Figure 139-Figure 145) became mature in 10-12 weeks. This rate can be increased by high humidity and retarded by relatively low humidity.

Figure 139. ***Marchantia polymorpha*** archegonial heads showing a bluish green variant. Sporangia are just beginning to emerge from the scales. Photo by Felipe Osorio-Zúñiga, with permission.

Figure 140. ***Marchantia polymorpha*** archegoniophores showing population with bluish-green coloring and purple scales with capsules emerging. Photo from BlueRidgeKitties, through Creative Commons.

Figure 141. ***Marchantia polymorpha*** ripe, unopened sporangia. Photo by Felix Riegel, through Creative Commons.

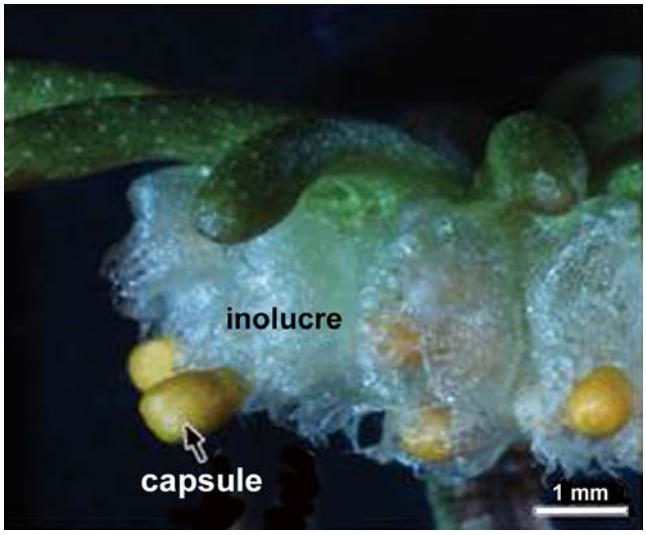

Figure 142. ***Marchantia polymorpha*** sporangia (capsules) emerging from the involucre. Photo by Masaki Shimamura, with permission.

Figure 143. ***Marchantia polymorpha*** archegonial head with elaters emerging on left, an empty capsule in center, and a capsule ready to open on right. Note the protective scales from which they are emerging. Photo by George Shepherd, through Creative Commons.

Figure 144. ***Marchantia polymorpha*** with mature sporangia dispersing spores, Laxarbakki, Myvatn, Iceland, 8 August 1987. Photo by Janice Glime.

Figure 145. ***Marchantia polymorpha*** archegoniophore with several unopened capsules on left and numerous elaters extended elsewhere. Note the purplish fringes on the scales of this specimen. Photo by Janice Glime.

Dörken (2012) described the sporophytes (Figure 139-Figure 145) of ***Marchantia polymorpha*** as short-lived, dying back after releasing the spores (Figure 146). A capsule typically contains several hundred thousand spores of similar size and shape, helping to account for the ability of this species to colonize newly disturbed areas such as those after fire.

Figure 146. ***Marchantia polymorpha*** archegonial head with empty sporangia among the scales and with sporangia beginning to die back. Photo by Janice Glime.

spores

O'Hanlon (1926) reported that in the Midwest of the USA, spores of ***Marchantia polymorpha*** (Figure 1-Figure 12) were "available" from early July to the middle of September. The sporophyte (Figure 147) produces an elater to spore (Figure 148-Figure 149) ratio of 1:128. A single capsule (Figure 141-Figure 142) of ***Marchantia polymorpha*** holds about 300,000 spores. Based on the typical number of capsules per archegonial head (~24), this would yield >7,000,000 spores per receptacle. But under favorable conditions, ~100 sporophytes are produced on one receptacle during a single growing season, suggesting that the total number of spores per individual archegonial receptacle is probably much greater (Duckett & Pressel 2009). The spores can be available for dispersal from early July to mid September, depending on latitude and altitude (O'Hanlon 1926). The spores remain viable for about one year.

Figure 147. ***Marchantia polymorpha*** archegonial head with dispersing sporangia having exserted elaters (yellow). Note the rhizoids on the rolled stalk. Photo by George Shepherd, through Creative Commons.

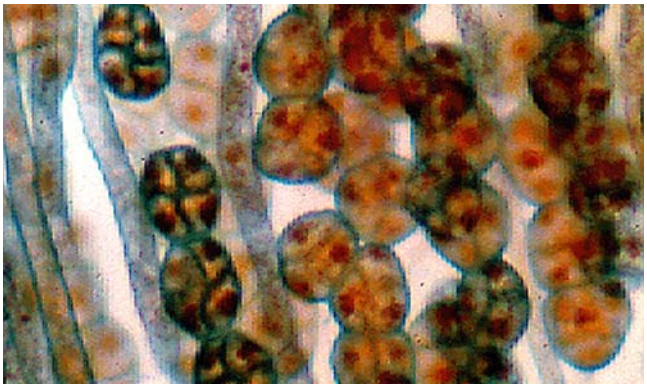

Figure 148. ***Marchantia polymorpha*** spore tetrads and immature elaters in capsule. Photo by Janice Glime.

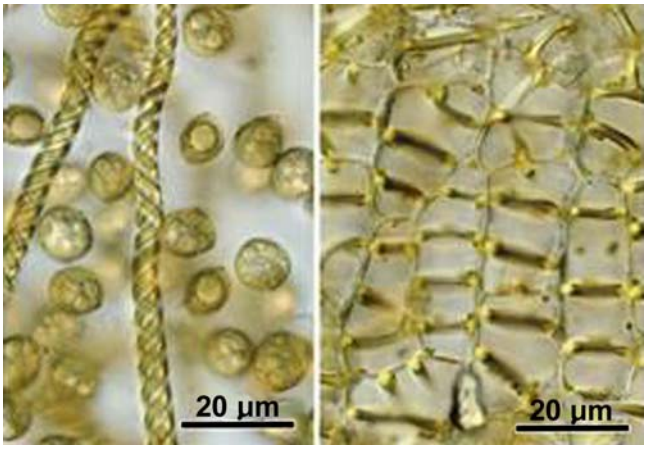

Figure 149. ***Marchantia polymorpha*** spores and elaters on **left** with closer view of thickenings of elaters on **right**. Photo by Masaki Shimamura, with permission.

Young and Kläy (1971) found ***Marchantia polymorpha*** (Figure 1-Figure 12) on the crater of a volcano on Deception Island, Antarctica, following the 1969 eruption. This rapid colonization of newly exposed substrata is itself remarkable, but the closest known source of propagules is 1,000 km away in South America! They assumed that numerous propagules must have arrived on just a small area to produce the colony pattern observed. Hence, the dispersal potential of this species is great, a factor that relates to the small size of its spores.

O'Hanlon (1925) detailed the germination of spores (Figure 150) and the early gametophyte stages in ***Marchantia polymorpha*** (Figure 1-Figure 12). Inoue (1960) studied the spore germination and early gametophyte development in the Marchantiales. Bischler (1984) examined spore morphology and germination in ***Marchantia***.

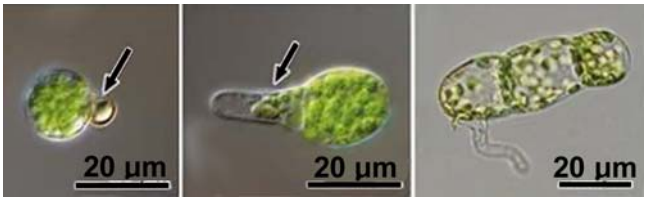

Figure 150. ***Marchantia polymorpha*** spore germination stages to a 3-celled protonema. Photo by Masaki Shimamura, with permission.

When the spore germinates (Figure 150), it typically produces a single primary rhizoid (Figure 151) after the spore has expanded and produced chlorophyll (O'Hanlon 1926). The spore first produces a very short filament. This is followed by division in a second cell, resulting in the thalloid structure that describes the mature protonema. Branching of the young thallus is common and can begin at an early stage. Rather than having a single apical cell dividing, it produces a marginal row of meristematic cells that continue to produce the mature thallus. At a stage of 30-40 cells, a notch develops in the apical region (Figure 151). Rhizoids develop behind this notch, anchoring the thallus and establishing its dorsiventrality. Mucilage cells arise on the lower side of the apex. In early stages, diminished light seems favorable, but by this stage 13-15 hours of light per day is optimum, with an optimum temperature of 18-22ºC. However, the optimum for "fruiting" is lower at 10-15ºC. They also germinate and grow better on a solid substrate than in a liquid medium.

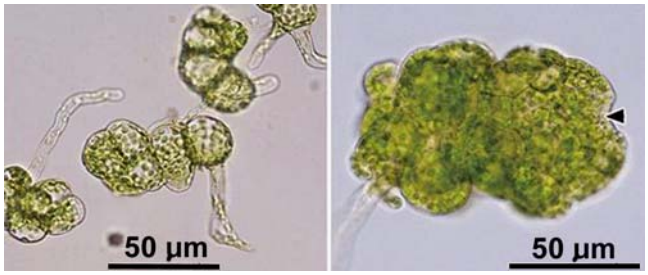

Figure 151. ***Marchantia polymorpha*** protonema development. Apical notch is shown at arrow on right. Photo by Masaki Shimamura, with permission.

Nakazato *et al*. (1999) were able to induce spore germination in ***Marchantia polymorpha*** (Figure 1-Figure 12) with intermittent irradiation with 15-min red light pulses given every 1 or 2 h for 24 h. Germination could also be induced by the addition of glucose to spores in total darkness. Their experiments indicate that photosynthesis is involved in the photoinduction of spore germination in this species, supporting the conclusion of Inoue (1960).

In the right conditions, the spores swell and gain chlorophyll (Figure 150) (Shimamura 2016). Once swollen they shed the primary spore walls and germinate within a few days. This germination is light-dependent (Heald 1898), suggesting a need for additional energy resources. The light requirement is 10 hours or longer (Nakazato *et al*. 1999). It seems to require a brighter light for protonema and thallus development than that required by other liverworts (Inoue 1960).

Red and far-red light affect both cell division and elongation in ***Marchantia polymorpha*** (Figure 1-Figure 12) sporelings (Figure 150) (Nishihama *et al*. 2015). Thus, it is likely that phytochromes are involved in development of sporelings.

Gemmrich (1976) found that both Fe and $Ca(NO_3)_2$ induce germination of the spores (Figure 150) of ***Marchantia polymorpha*** (Figure 1-Figure 12). Optimal germination also requires KNO_3 and $MgSO_4$. Gibberellic acid had no effect on induction of spore germination in dark cultures.

Initial spore germination (Figure 150) is dependent on light (Hartmann & Weber 1990). As shown in ***Marchantia***

polymorpha (Figure 1-Figure 12), following the initial series of reactions that ultimately result in the swelling of the spore, polarity develops (Figure 150). This becomes obvious when the protonema protrudes from the spore as a filamentous germ tube (Figure 150).

Spore germination (Figure 150) of ***Marchantia polymorpha*** (Figure 1-Figure 12) requires 10 hours or longer (Nakazato *et al*. 1999). The entire light spectrum is effective for germination, but red light is the most effective. The effect of red light is not reversed by subsequent far-red light.

Shibaya *et al*. (2005) demonstrated that AGPs (arabinogalactan proteins) differed before and after protonema development, suggesting that they are involved in differentiation and development. Furthermore, binding of the AGPs inhibits protonema development in ***Marchantia polymorpha*** (Figure 1-Figure 12), causing disturbances at the cell surface and inhibiting cell-wall synthesis.

Upon germination, one primary rhizoid appears (Figure 150) following the growth of the spore and chlorophyll development (O'Hanlon 1926). At a stage of about 30-40 cells a notch appears in the apical region of the young gametophyte (Figure 151-Figure 152). Although germination seems to benefit from more moderate light, growth is best at 12-15 hours of "good intensity light," with the best temperatures in the range of 18º-22ºC. However optimum temperatures for sporophyte development are 10º-15ºC. It is interesting that chlorophyll can form in the spores as they imbibe water in the dark. Furthermore, with glucose in the medium spore germination can occur in the dark, further supporting the conclusion that photosynthesis, hence the production of sugar, is necessary for germination.

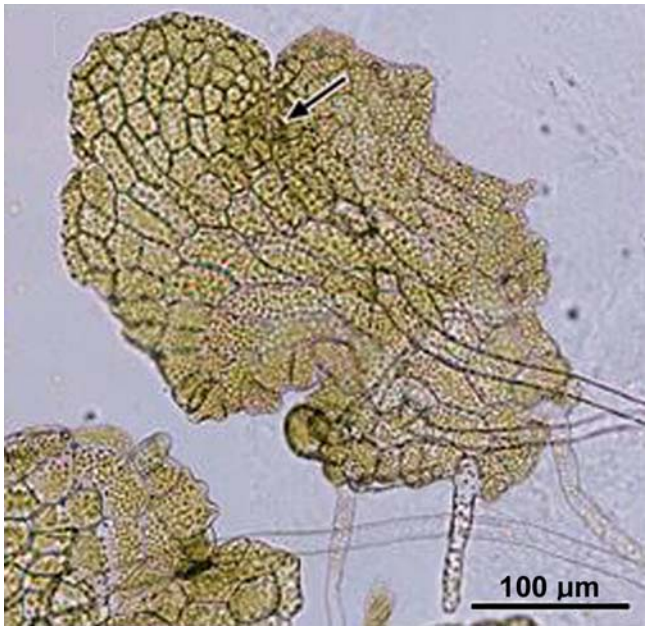

Figure 152. ***Marchantia polymorpha*** mature thalloid protonema with rhizoids. **Arrow** indicates apical notch. Photo by Masaki Shimamura, with permission.

gemmae (Figure 153-Figure 170)

Hallier (1966) experimented with germination of gemmae in ***Marchantia polymorpha*** (Figure 1-Figure 12). He examined the effect of 2,4-mononitrophenol on respiration and induction of germination.

Terui (1981) reported that archegoniophore production in ***Marchantia polymorpha*** (Figure 1-Figure 12) occurs under long-day conditions, conditions that at the same time suppress the formation of gemmae cups. The gemmae cup suppression occurs about 20 days before the archegoniophore protrudes. High sucrose, on the other hand, induces gemmae cup development. When low light was provided for a prolonged time, it stimulated gemmae cup formation.

Une (1984) confirmed the negative correlation between gemmae cup (Figure 153-Figure 170) production and initiation of sexual structures. The gemmae cups occur more frequently on the margins of the colony, decreasing in number as sexual branches arise toward the inner part of that colony. This seems to relate to the age of the thalli and consequent change in the nutrient condition of the soil beneath the colony, with younger thalli occurring at the margins. Une also found that female plants produce sexual structures more frequently than do males.

Benson-Evans (1964) cultured ***Marchantia polymorpha*** (Figure 1-Figure 12) that produced gemmae (Figure 153-Figure 170) at 10ºC in short days (6 hrs), the opposite conditions of those that resulted in archegoniophore production. On the other hand, Hedger *et al*. (1972) found that long days were needed to maintain the development of gemmalings of this species on an inorganic medium. Carbon additions did not affect the growth rate under long-day or short-day photoperiods.

Figure 153. ***Marchantia polymorpha***; note the arrangement of the gemmae cups along the midrib in these older thalli. Photo by Jan-Peter Frahm, with permission.

Figure 154. ***Marchantia polymorpha*** gemmae cups arranged on midrib. Photo by Robert Klips, with permission.

Figure 155. ***Marchantia polymorpha*** showing gemmae cups along the midrib. Photo by Walter Obermayer, with permission.

Figure 156. ***Marchantia polymorpha*** with red edges; note the rim within the gemmae cup, holding young gemmae within it. Photo by Brenda Dobbs, through Creative Commons.

Figure 157. ***Marchantia polymorpha*** gemmae cup with lenticular gemmae and thallus showing pores. Photo by Walter Obermayer, with permission.

Figure 158. ***Marchantia polymorpha*** gemmae cup. Photo by Bernard de Cuyper, with permission.

Figure 159. ***Marchantia polymorpha*** gemmae in red cups that may indicate high light or other stress. Photo by Dick Haaksma, with permission.

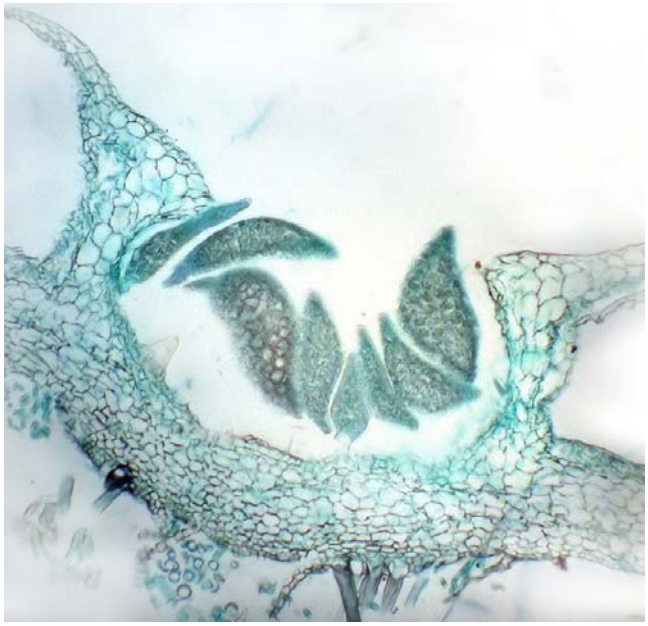

Figure 160. ***Marchantia polymorpha*** section of gemmae cup; note stalk on middle gemma. Photo by George Shepherd, with permission.

Figure 161. ***Marchantia polymorpha*** gemmae cup section. Note the inner rim that confines the young gemmae in the cup. Photo by Ralf Wagner <www.dr-ralf-wagner.de>, with permission.

Figure 162. ***Marchantia polymorpha*** gemmae cups; note that some of the cups have disintegrated, permitting gemmae to escape easily, but mostly onto the thallus. Photo by Andrew Spink, with permission.

Figure 163. ***Marchantia polymorpha*** gemmae cups as the mature gemmae begin to leave the cup. Photo by Walter Obermayer, with permission.

Figure 164. ***Marchantia polymorpha*** gemmae cups with a fringe; some gemmae are on the thallus, ungerminated. Photo from BlueRidgeKitties, through Creative Commons.

Figure 165. ***Marchantia polymorpha*** gemma. Photo by Des Callaghan, through Creative Commons.

Figure 166. ***Marchantia polymorpha*** females showing gemmae cups on younger thalli and mature archegoniophores on older parts. Photo courtesy of Craig Waddell.

Tarén (1958) described the gemmae (Figure 165) as growing on stalks in the cup (Figure 160, Figure 167),

where they are surrounded by hairs that excrete slime (Figure 161). They have two growing regions and branches develop from both.

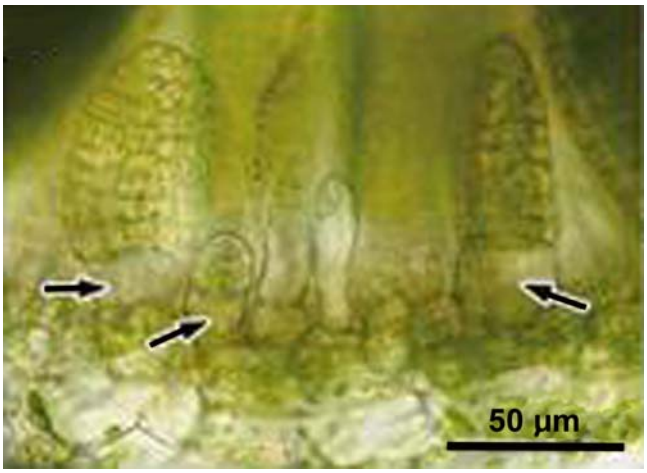

Figure 167. ***Marchantia polymorpha*** gemmae with arrows indicating stalks. Photo by Masaki Shimamura, with permission.

Hiwatashi *et al*. (2019) and Yasui *et al*. (2019) explored the genetic encoding for the genes that are involved in the regulation and formation of gemmae (Figure 153-Figure 165).

Gemmae cups (Figure 153-Figure 164) need nutrients to develop. Plants lacking nitrate become light green and have few gemmae cups and infrequent forking (Voth 1941). By contrast, those lacking phosphate become dark green and produce gemmae cups. They also have frequent dichotomies that cause the plants to form rosettes.

The mechanism of dispersal of the gemmae (Figure 153-Figure 165) has been a favorite example for textbooks. In fact, Laplaud *et al.* (2022) even presented their work at a meeting of the American Physical Society.

The splash cup (Figure 168) a dispersal mechanism served as the topic for an entire book (Brodie 1951). Brodie noted that splash cups commonly form 60-70º angles with their horizontal surface. These cups have a broad basal attachment and the propagules are **lenticular** (lens-shaped). He suggested that such cups could facilitate splashing of their contents for about 60 cm in ***Marchantia polymorpha*** (Figure 1-Figure 12). Equihua (1987) conducted further experiments, finding that raindrops could splash the gemmae up to 120 cm from the parent cup.

Figure 168. ***Marchantia polymorpha*** with gemmae cups. Note the bird's nest fungus (*Nidularia*) beside the thallus, a larger splash cup. Photo by Martin Hutten, with permission.

The ease of dispersal of the gemmae (Figure 153-Figure 165) has been a point of consternation for nursery growers, inspiring research on the mechanism. England and Jeger (2005) experimented with an overhead sprinkler system to determine various nozzle differences and their effects on dispersal. They demonstrated the effectiveness of the dispersal by using red dye in the cups in place of gemmae. At extreme water pressures of 1.5 and 3 bars, fewer gemmae were dispersed at all nozzle sizes. When the flow rate was adjusted to 160 L h^{-1} dispersal number increased with height of the nozzle. At the other flow rates tested the nozzle height lacked any clear effect on number dispersed. The maximum distance travelled was 1.6 m.

The gemmae cup (Figure 153-Figure 164) has a decorated border and is cone-shaped (Figure 169). The cup-shaped container permits a raindrop to splash the gemmae to some distance from the cups (Laplaud *et al*. 2022). When a raindrop is non-centered when it lands, it creates a jet of water splash that carries a few gemmae with it. Laplaud and coworkers found that this propulsion can carry gemmae up to a meter from the cup. They are continuing their research to determine the effects of cone angle and the presence of decorations on its border (Figure 169).

Figure 169. ***Marchantia polymorpha*** gemma cup showing decorated border. Photo by John Forlonge through Flickr.

The climatic conditions can be right for germination, but the gemmae (Figure 153-Figure 164) could be in the wrong place. To determine if a substrate surface is present, ethylene is the most likely hormone to carry out this function. Because it is a gas, it is able to accumulate between a gemma and its substrate. Duarte (2020) used constant ethylene-signalling mutants to search for such a response. She found that the hormone ethylene could be part of the process of dormancy establishment, maintenance, and release of gemmae (Figure 153-Figure 165) in ***Marchantia polymorpha*** (Figure 1-Figure 12).

But experiments by Thullen (1965) suggest that the substrate is not important in the orientation of the rhizoids (Figure 82-Figure 54) of ***Marchantia polymorpha*** (Figure 1-Figure 12) gemmae (Figure 153-Figure 165). When the light source is above, surface gemmae produced 92% of their rhizoids on the side away from the light; those gemmae submersed in the agar produced only 85% on the side away from light. When the light source is beneath the gemmae, only 50 and 45% of the rhizoids, respectively,

appeared on the side opposite the light. When light was provided from beneath the gemmae it changed the percentage of rhizoids arising from the upper surface, but did not change the total number of rhizoids. Gemmae grown in total darkness produced rhizoids only on the lower side. Thullen concluded that both gravity and direction of light are important in rhizoid production. Light intensities seemed to have no effect. Temperature appears to be an important determinant in gemma germination of ***Marchantia polymorpha*** gemmae. Thullen found a sharp decrease in the number of rhizoids produced by gemmae at temperatures of 26ºC and above.

Otto (1976) also demonstrated that the orientation of the gemma (Figure 153-Figure 165) determines where rhizoids form, with the gravitational force assuring their development on the lower side of the gemma. If that side is in contact with the substrate, more rhizoids are produced than if that surface is exposed to air. In the dark, only ~20% of the gemmae produce rhizoids. If the gravitational direction is alternated and no illumination is provided, no rhizoids form. The direction of light source influences the location of rhizoids only when the gravitational direction is not constant, with more rhizoids formed on the darker side.

As already noted, Otto and Halbsguth (1976) found that 350 nm light was the most effective wavelength to induce rhizoids in the gemmae of ***Marchantia polymorpha*** (Figure 1-Figure 12). Wavelengths below 550 or above 670 nm failed to stimulate rhizoid formation. The responses exhibited red far-red reversibility, suggesting that phytochrome was involved. IAA at 10^{-4} M causes the same effect as 1 hour of red radiation. They suggested that the wavelength of light might affect the influence of the phytochrome system on permeability of the membrane to IAA.

Rousseau (1952, 1953, 1954a) explored the influence of heteroauxins (IAA) on the growth of gemmae cups (Figure 169) in ***Marchantia polymorpha*** (Figure 1-Figure 12). Rousseau (1954b) further showed that coumarin inhibited the growth of the gemmae.

Prior and Brown (1970) attempted to identify the hormone(s) involved in initiation of rhizoids. They found no influence on germination or initial intercalary growth of gemmae (Figure 153-Figure 165) by 2,4-D, maleic acid hydrazide, gibberellic acid, or 2-furanacrylic acid (β-2-furylacrylic acid) in a range of concentrations Gibberellic acid delayed development. But none of them caused a difference in number of rhizoids. They did, however, find that apical cell activity and cell elongation were suppressed. Both 2,4-D and maleic hydrazide suppressed internal differentiation They did find that increasing age of the thallus caused greater sensitivity to both type and concentration of the regulator.

Dunham and Bryan (1968) explored the effects of amino acids on the development of the gemmalings in ***Marchantia polymorpha*** (Figure 1-Figure 12). At concentrations of 10^{-3}, l-isoleucine, l-leucine, l-methionine, or l-threonine resulted in a disruption of the apical regions. At lower concentrations, l-arginine, l-histidine, l-hydroxyproline, l-lysine, or l-tryptophan caused morphological irregularities. The irregularities were amino acid specific.

Gemmalings can reach reproductive maturity relatively quickly. Miller and Colaiace (1969) found that within 3-6 weeks the gemmalings of ***Marchantia polymorpha*** (Figure 1-Figure 12) responded to a 1% agar medium in a 24-hour photoperiod at 23ºC by producing antheridiophores and archegoniophores.

During (2001) hypothesized that the tradeoff between dispersability and longevity in soil diaspore banks could result in the scarcity of weedy species such as ***Marchantia polymorpha*** (Figure 1-Figure 12) in the soil bank. But in fact, the opposite appears to be the case. Species with large spores (*i.e.* limited dispersal distances) tend to be more persistent in the soil diaspore bank. Presumably, this larger diaspore would include the large gemmae of ***M. polymorpha***. This is also in sharp contrast to seeds, wherein small seeds predominate in the diaspore bank. During suggests that there is more predation on larger seeds. Furthermore, bryophytes have much more representation of asexual diaspores than do seed plants. Such asexual diaspores as gemmae are generally produced through a greater part of the growing season than the very seasonal seeds or most kinds of bryophyte spores. The spores of ***M. polymorpha*** might be an exception to that spore seasonality, however. And certainly, its spores greatly exceed gemmae in number.

Miller and Alvarez (1965) emphasized that in gemmae both notches with apical cells are capable of growing. In their experiments with ^{60}Co they found that both cells had to be damaged to eliminate the survival of the gemma.

Miller (1966) described the gemmae of ***Marchantia polymorpha*** (Figure 1-Figure 12) as discoidal with two apical notches 180º from each other. Each notch has two apical cells. These apical cells, however, are not the only locations where growth, as cell proliferation, can occur. In irradiated gemmae, the nuclear volume is important in the cell survival, with larger nuclei having greater survival.

Nehira (1973, 1977) explored the development of the rhizoids of the gemmae of ***Marchantia polymorpha*** (Figure 1-Figure 12) and the adsorption of Ca on the rhizoids of the gemmae and its role in their differentiation.

One surprising effect on germination of the gemmae is that of nickel (Ni). Lepp and Hockenhull (1983) found that $NiSO_4$ served as a significant growth stimulus at 0.15 ppm Ni, but at concentrations above 0.25 ppm the gemmalings exhibited a toxic response. On the other hand, $NiCl_2$ stimulated growth of the gemmalings at 0.25 ppm Ni, and they could tolerate $Ni(NO_3)_2$ up to 0.5 ppm. Furthermore, they observed different toxicity depending on the environment of the gemmalings, with those from an urban area being more tolerant than those grown in the glasshouse.

regeneration and growth

Perhaps the earliest form of reproduction is **regeneration**. This ability to grow new plants from fragments is known from the **Cyanobacteria** and algae and permits them to survive from parts when most of the whole has died. It is particularly useful in aquatic organisms that can be moved rather easily to new locations by the water.

Vöchting (1885) noted the regeneration capability of the **Marchantiales** and in 1887 (Vöchting 1887) reported that that every living cell in ***Marchantia polymorpha*** (Figure 1-Figure 12) was capable of regenerating an entire plant. Frye (1928) determined the age of a population of ***Marchantia polymorpha*** in western Washington, USA, to

be four years old. Only three of these yearly growth segments was alive. The growth habit of ***Marchantia polymorpha*** to die at the postical end while growing at the bifurcating apical end permits the population to expand.

Giles (1971) discussed the mechanism that governed the stability of differentiation in ***Marchantia***. These included a possible mechanism intrinsic to the to the cell, presumably in more highly differentiated species. However, in other bryophytes this control might be more affected by factors in the environment. But the factors affecting cellular dedifferentiation of an isolated fragment remained unknown. Giles suggested that these factors must be biochemical, probably involving RNA metabolism, and should be investigated.

Barner (1990) found that only the rhizoids from subcultured explants were able to regenerate thalli. The cultures required a directional light source.

Light is critical in regeneration of bryophytes (Nishihama *et al*. 2015). These researchers demonstrated that ***Marchantia polymorpha*** (Figure 1-Figure 12) has a single phytochrome gene and that phytochrome regulates re-entry into the cell cycle and control of cell shape in newly regenerating tissues. Nevertheless, light is not essential for regeneration, but it exhibits considerable control over the process. But, sugar can cause normal regeneration in the dark, suggesting the importance of photosynthesis to supply the energy.

Li (1990) described the difficulties of culturing gemmae and gametophytes of ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that in their cultures it required dedifferentiation and redifferentiation, with rather specific cultural conditions and media. They used 2,4D and 3% sucrose to encourage the tissue development. This process could require as long as 10 months. Nevertheless, they found the process to be easier than in tracheophytes.

Bryophytes are known for their "extraordinary competency of regeneration" (Nishihama *et al*. 2015). This is possible due to their high level of developmental plasticity, permitting them to regenerate from cells, tissue fragments, branches, and even reproductive organs. Gardeners in Japan and elsewhere take advantage of this ability to propagate many plants from just a few by drying and fragmenting them (see Horticulture chapter in Volume 5).

Yoshikawa *et al*. (2018) found that the stress caused by wounding (Figure 170) induces phenylalanine ammonia lyases. These lyases initiate the accumulation of phenylpropanoids in ***Marchantia polymorpha*** (Figure 1-Figure 12). Wounding induces the biosynthesis of luteolin, apigenin, and isoriccardin C, all of which are biosynthesized through the phenylpropanoid pathway.

Ishida *et al*. (2022) found that diminished auxin signalling triggers the cellular reprogramming needed for regeneration in ***Marchantia polymorpha*** (Figure 1-Figure 12). Auxin is produced in apical cells, and removal of the apex enhances regeneration. Addition of auxin inhibits regeneration. They were able to identify the gene responsible for the cell proliferation needed for regeneration.

Mechanisms that control regeneration are also at play in controlling apical dominance (Figure 20) in ***Marchantia polymorpha*** (Figure 1-Figure 12). Davidonis and Munroe (1972) found that the larger lobe in this dichotomously branching plant is always the one closest to the midrib. If these two lobes are separated by a cut while still in an early stage of lobe growth, the smaller lobe is no longer inhibited and is able to grow to equal size. Adding the auxin IAA to the smaller lobe after cutting will re-establish the dominance of the other lobe. The researchers suggested "that the type of neighbor lobe dominance in ***Marchantia*** resulting in its typical fan-shaped growth habit is maintained by auxin through a differential sensitivity of the two neighbors to auxin inhibition."

Figure 170. ***Marchantia polymorpha*** gemmae cups and antheridiophores in Europe; note the dead thalli and the red gemmae cups that indicate stress. At the same time, the apical portions continue to grow. Photo by Michael Lüth, with permission.

Maravolo *et al*. (1975) traced the transport of labelled IAA in the thallus of ***Marchantia polymorpha*** (Figure 1-Figure 12). There was a marked dominance of the hormone in the primary lobe. Movement to the secondary lobe could be enhanced by disruption of the conductive tissue or by removing the primary apex. Gibberellin and cytokinin also increased the activity of the IAA in the subdominant lobe, indicating that gradients of these three hormones might serve as growth regulators and these could be established independently at each apex.

Bhargava and Chauhan (1978) reported a dichotomously branched vegetative thallus at the tip of a gametophore stalk. I have to wonder if this is a germinated gemma, such as that observed by Li Zhang for ***Marchantia emarginata*** (Figure 171) rather than a branch.

Figure 171. ***Marchantia emarginata*** ssp. ***tosana***, in Guangdong, S. China, with germinated gemmae and young thalli with gemmae cups growing on male receptacle arms. Photo courtesy of Li Zhang.

Kubota *et al*. (2013) were successful in achieving ***Agrobacterium***-mediated (Figure 172) transformation in regenerating thalli of ***Marchantia polymorpha*** (Figure 1-Figure 12). Developmental timing is important to the success of this transformation. Previously, efficient ***Agrobacterium***-mediated transformation had only been accomplished with sporelings. Iwakawa *et al*. (2021) further described a protocol useful for using this method in ***M. polymorpha***. GUS (β-glucuronidase) activity was detected 2 days after infection and became saturated after 3 days.

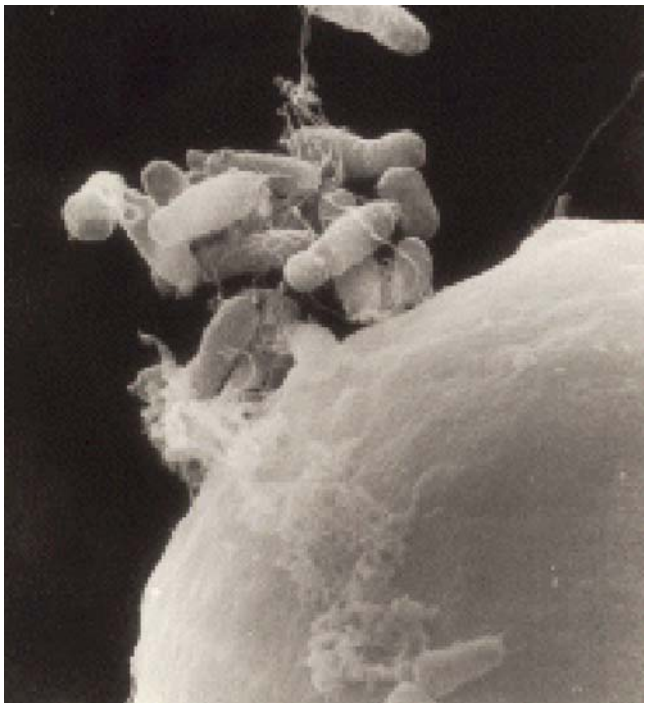

Figure 172. SEM of ***Agrobacterium tumefaciens***; the genus ***Agrobacterium*** is used for genetic transformation and has been developed as a tool in ***Marchantia polymorpha***. Photo from CDC, through public domain.

Shibaya and Sugawara (2007) found that the protoplasts of ***Marchantia polymorpha*** (Figure 1-Figure 12) could regenerate new cell walls in the initial culture, but the survival rate then decreased rapidly. β-glucosyl Yariv reagent (βglcY) could suppress this reduction in the survival rate. This substance binds to arabinogalactan proteins (AGPs) and does not increase survival except when added during the incipient cell wall regeneration. The researchers suggested that AGPs were involved in cell wall regeneration. Adding activated charcoal to the medium also permitted the cells to divide vigorously. It appears that AGPs and β-1,3-glucan are important in the survival and subsequent cell division of regenerated cells of ***M. polymorpha*** protoplast cultures.

Harashima and Ono (1991) tested the long-term culturing of ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that after years of suspended culture they regained regeneration potential. The loss of morphogenetic potential in some was correlated with chromosome aberrations. They were able to maintain gemmae cultures for 213 months and spore cultures for 64 months.

Takenaka *et al*. (2000) used ***Marchantia polymorpha*** (Figure 1-Figure 12) for direct particle bombardment with plasmid pMT. They produced hygromycin-resistant cell masses that developed into hygromycin-resistant thalli. These modified thalli transmitted the genetic modification to their gemmae for three generations. Hence, this could be a valuable tool for molecular analysis of this species and others.

Role

The role of aquatic bryophytes in accumulating pollutants is well known. Even ***Marchantia polymorpha*** (Figure 1-Figure 12) has been tested for its ability to purify water (Baltazar Pereda & Rebaza 2021). The researchers were concerned that there is no treatment for the wastewater for a sewage system in La Libertad. The water is used to irrigate stem crops, but it causes bad odors and endangers the health of those living around the canal. Both the water hyacinth (***Eichhornia crassipes*** – Figure 173) and duckweed (***Lemna minor*** – Figure 174) showed an excellent removal efficiency, improving oxygen and diminishing coliforms. However, the ***Marchantia polymorpha*** failed to make significant changes in water quality.

Figure 173. ***Eichhornia crassipes*** (water hyacinth), a species that is much more efficient at removing pollutants than is ***Marchantia polymorpha***. Photo by Wouter Hagens, through public domain.

Figure 174. ***Lemna minor*** (duckweed), a species that is much more efficient at removing pollutants than is ***Marchantia polymorpha***. Photo by Barbarossa, through Creative Commons.

Briggs (1972) reported that this species contains higher levels of lead than bryophytes that had been used in previous studies. Large quantities of plants could be grown quickly from gemmae, then exposed in a lead-polluted atmosphere to determine levels in the plants, and by extrapolation, the environment.

Accumulation of heavy metals characterizes many bryophytes (Cahuana & Aduvire 2019). Sharma (2007) placed ***Marchantia polymorpha*** (Figure 1-Figure 12) in moss bags to monitor pollution in several areas to compare pollutants. Plants accumulated the highest levels (2276 µg g^{-1} dry weight) of lead in summer. But ***Marchantia*** lacks the high level of surface area seen in most mosses, making Sharma conclude that it a less useful accumulator than we might find in mosses and leafy liverworts.

Perhaps the most useful role of ***Marchantia polymorpha*** (Figure 1-Figure 12) is its use in the laboratory. The gemmae (Figure 153-Figure 165) of liverworts such as ***M. polymorpha*** represent isogenic progeny that can be used to experiment with gene expression (Kubota *et al*. 2013). The most common system used is to supply ***Agrobacterium*** (Figure 20) to regenerating thalli produced from these gemmae. These bacteria are able to transfer genes into the liverwort.

Interactions

Seed plants have an array of structural defenses as well as biochemical defenses against herbivores. Bryophytes, on the other hand, generally lack structural defenses, at least the elaborate ones such as spines, thick cuticle, and dense, lignified tissues. But the bryophytes, instead, are endowed with an extremely varied array of biochemical defenses.

Despite knowing about the wide diversity of secondary compounds in bryophytes, especially in liverworts, the research on their functions in the ecosystem and their sources, particularly in cooperation with microorganisms, has been rather neglected until recently (Stelmasiewicz *et al*. (2021). Noting that bryophytes produce many compounds unique to bryophytes, Stelmasiewicz and coworkers used a volatile extract to isolate the volatile compounds produced by the ***Marchantia polymorpha*** (Figure 1-Figure 12)-microorganism symbiosis. They isolated cuparane-, chamigrane-, acorane-, and thujopsane-type sesquiterpenoids from ***Marchantia polymorpha***. These compounds proved to be active against some types of human cancer. But what do they do for the liverwort?

Poveda (2020a) touted the use of ***Marchantia polymorpha*** (Figure 1-Figure 12) as a model organism in studies of plant-microorganism interactions. He reviewed the published literature on these interactions.

Bacterial Interactions

As new studies are emerging, we are learning of the great dependence of bryophytes on other organisms. Bacteria are among these partners. Kutschera and Koopmann (2005) found that epiphytic methylobacteria promote the growth of ***Marchantia polymorpha*** (Figure 1-Figure 12). While living on the surfaces of plants these bacteria secrete cytokinins. The bacterial extracts had no effect on seeds of maize or sunflower, but did promote the growth of isolated ***M. polymorpha*** gemmae (Figure 153-Figure 165) on agar plates.

Alcaraz *et al*. (2018) identified ***Bryobacter, Lysobacter*** (Figure 175), ***Methylobacterium*** (Figure 176), ***Paenibacillus*** (Figure 177), ***Pirellula***, ***Rhizobium*** (Figure 178-Figure 179), and ***Steroidobacter*** from ***Marchantia polymorpha*** (Figure 1-Figure 12) as well as from ***M. paleacea*** (Figure 14; see part 1 of this chapter). These plant symbionts are known for plant-growth promotion, complex exudate degradation, nitrogen fixation, methylotrophs, are disease-suppressive bacteria, and are hosted within the plant thallus.

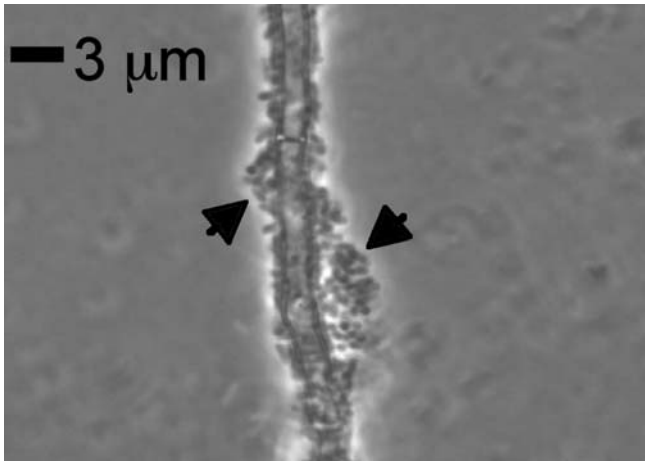

Figure 175. ***Lysobacter*** hyphae, a genus that is sometimes associated with ***Marchantia polymorpha***. Photo by Don Kobayashi, through Creative Commons.

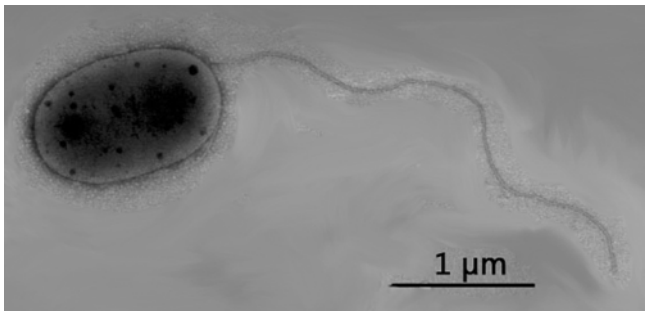

Figure 176. ***Methylobacterium jeotgali***; ***Methylobacterium*** is a genus that is sometimes associated with ***Marchantia polymorpha***. Photo from Aslam *et al*. 2007, through Creative Commons.

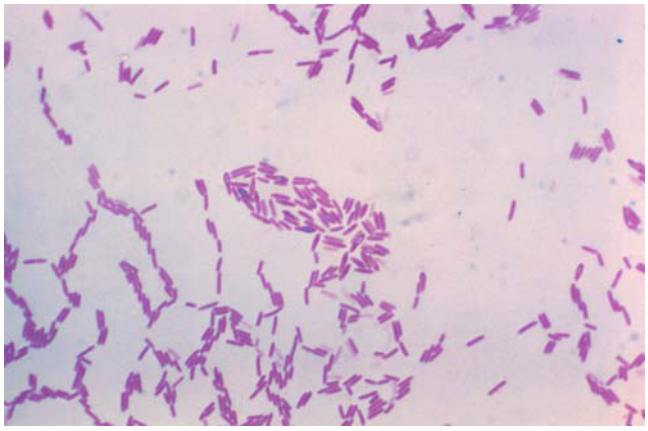

Figure 177. ***Paenibacillus polymyxa***; ***Paenibacillus*** is a genus that is sometimes associated with ***Marchantia polymorpha***. Photo from CDC, through public domain.

Figure 178. ***Rhizobium*** root nodule on *Vicia faba* (broad bean) roots; ***Rhizobium*** is a nitrogen fixer and known associate of ***Marchantia polymorpha***. Photo by Whitney Cranshaw, through Creative Commons.

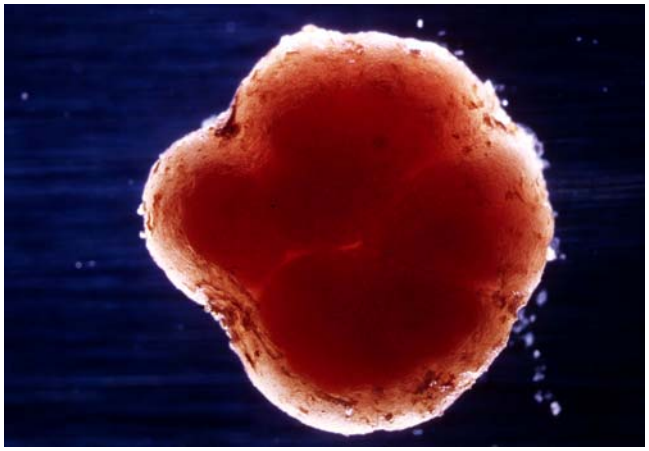

Figure 179. ***Rhizobium*** nodule, showing typical red color. Photo from CSIRO, through Creative Commons.

Bryophytes are used in some cultures as medicinal plants. Some groups of Indonesian people use ***Marchantia polymorpha*** (Figure 1-Figure 12) as a traditional medicine to treat skin infections (Ramadhan & Agustien 2019). The species has flavones and flavone glycosides as well as simple terpenoids that are able to inhibit the multiplication of bacteria. These researchers chose to isolate endophytic bacteria from ***Marchantia polymorpha.*** Six species of endophytic bacteria that have the potential to produce antibiotics were successfully isolated from the thallus; these bacteria were successful against ***Staphylococcus aureus*** (Figure 180).

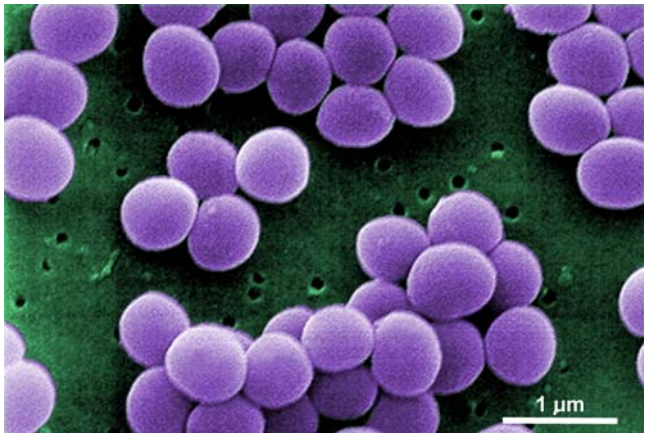

Figure 180. Colorized SEM of bacterium ***Staphylococcus aureus***, a species that is inhibited by antibiotics produced by other bacteria endophytic in ***Marchantia polymorpha***. Photo by Janice Haney Carr, CDC, through public domain.

Himanshu *et al*. (2007) found that acetone-soluble extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12) had antibiotic activity against the Gram negative bacteria ***Escherichia coli*** (Figure 16) and ***Salmonella typhi*** (Figure 181) and two fungi ***Aspergillus niger*** (Figure 182) and ***Candida albicans*** (Figure 183-Figure 184), all human pathogens. Such antibiotic activity makes this species of interest for finding applications to replace the ever-growing number of antibiotics that are helping to create "super bugs" with antibiotic resistance. But do they help the bryophyte? One indication that they might not, at least as antibacterial agents, is that the water soluble extracts did not show any inhibitory effects on the pathogens tested.

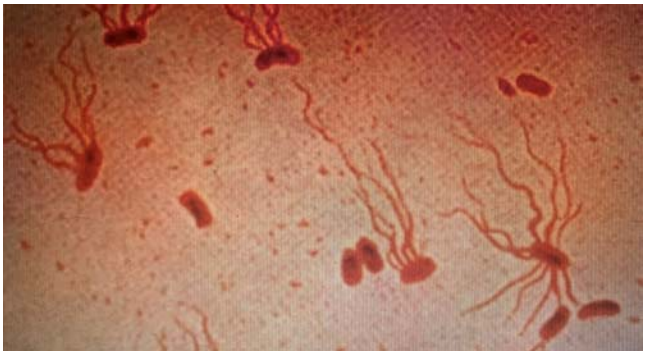

Figure 181. ***Salmonella typhi*** with flagellar stain, a species of bacteria that experiences antibiotic activity from acetone-soluble extracts of ***Marchantia polymorpha***. Photo by Microbewriter, through Creative Commons.

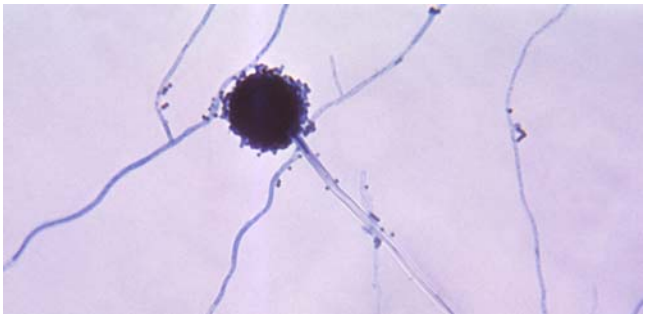

Figure 182. ***Aspergillus niger***, a species of fungus that experiences antibiotic activity from acetone-soluble extracts of ***Marchantia polymorpha***. Photo from CDC, through Creative Commons.

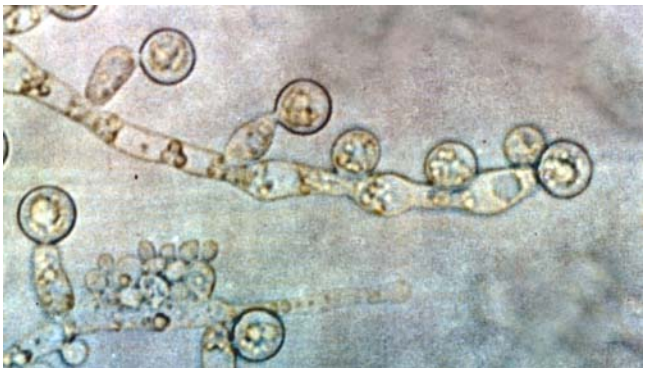

Figure 183. ***Candida albicans***, fungus that experiences antibiotic activity from acetone-soluble extracts of ***Marchantia polymorpha***. Photo by Graham Colm, through Creative Commons.

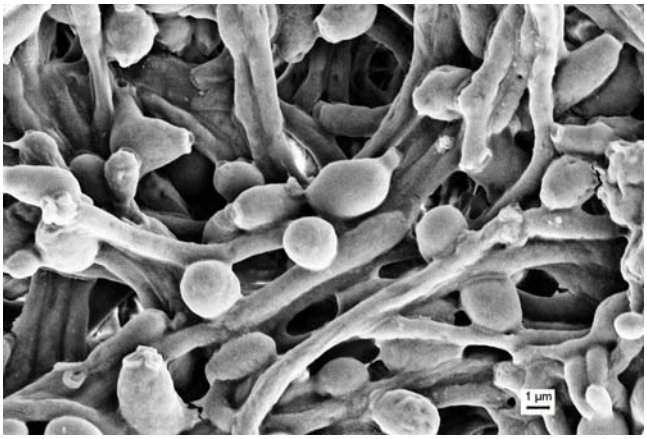

Figure 184. SEM of ***Candida albicans***. Photo by Vader1941, through Creative Commons.

Using specimens from Vietnam, Son *et al*. (2020) isolated lunularin, marchantin A, isoriccardin C, luteolin, and apigenin from ***Marchantia polymorpha*** (Figure 1-Figure 12). Isoriccardin C had "remarkable" antibacterial activity against ***Staphylococcus epidermidis*** (Figure 185). Several extracts exhibited anticancer activity.

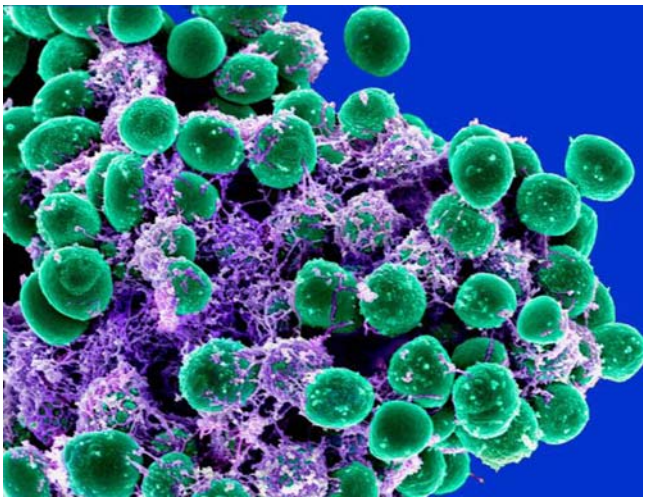

Figure 185. ***Staphylococcus epidermidis***, a species that experiences strong negative effects from isoriccardin C from ***Marchantia polymorpha***. Photo from NIAID, through Creative Commons.

Ivković *et al*. (2021) identified terpenes, oils, sugars, and bis-bibenzyls in methanol extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12). These extracts were effective in inhibiting Gram-positive bacteria but had no effect on Gram-negative bacteria.

Mewari and Kumar (2008) tested antibacterial activity using crude methanol and flavonoid extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12) against three strains of bacteria [***Escherichia coli*** (Figure 16), ***Proteus mirabilis*** (Gram negative; Figure 186), and ***Staphylococcus aureus*** (Figure 180) (Gram positive)] and four of fungi [***Aspergillus flavus*** (Figure 187), ***A. niger*** (Figure 182), ***Candida albicans*** (Figure 183-Figure 184), and ***Trychophyton mentagrophytes*** (Figure 188)]. All the microorganisms proved to be sensitive to all of the extracts, suggesting that ***M. polymorpha*** had a good potential as a source of antimicrobial drugs.

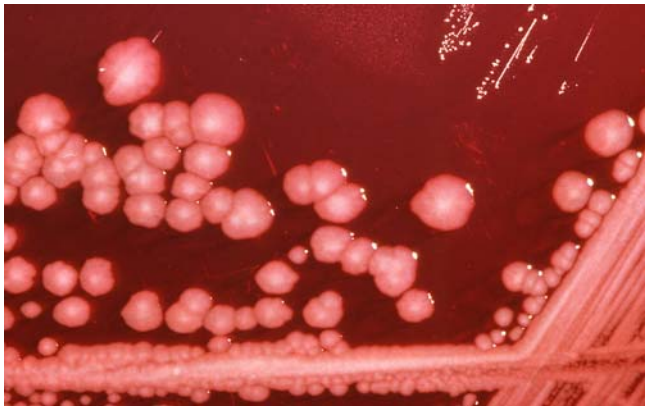

Figure 186. ***Proteus mirabilis***, a bacterial species that is sensitive to crude methanol and flavonoid extracts of ***Marchantia polymorpha***. Photo from CDC, through public domain.

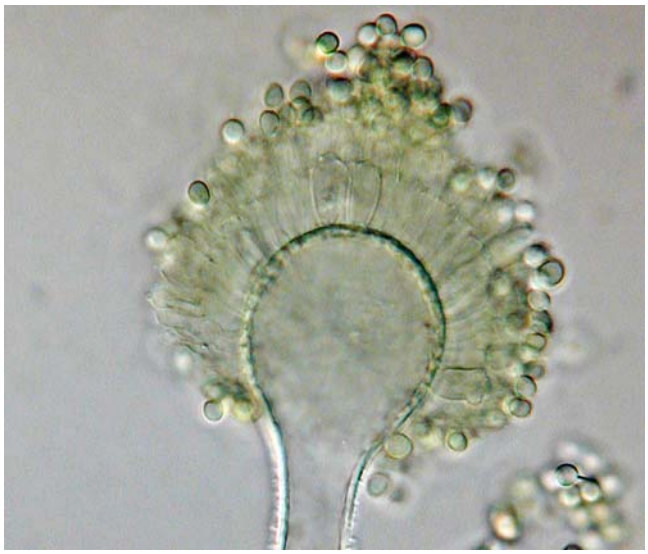

Figure 187. ***Aspergillus flavus***, fungal species that is sensitive to crude methanol and flavonoid extracts of ***Marchantia polymorpha***. Photo from Medmyco, through Creative Commons.

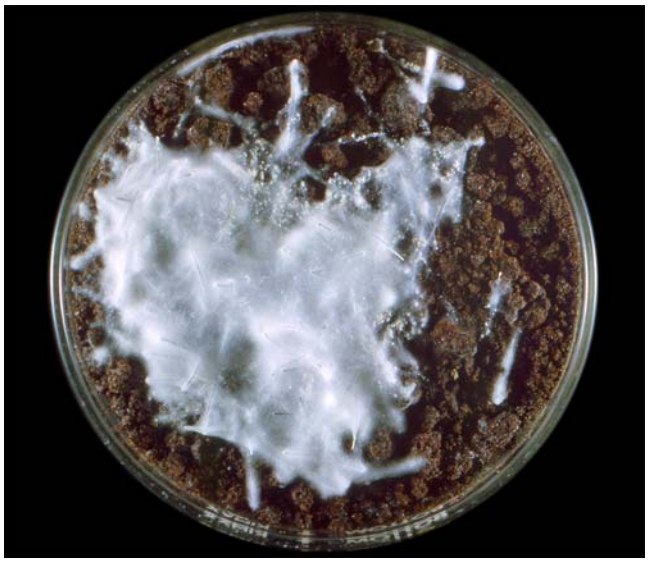

Figure 188. ***Trichophyton mentagrophytes***, fungal species that is sensitive to crude methanol and flavonoid extracts of ***Marchantia polymorpha***. Photo by Dr. Libero Ajello, CDC, through public domain.

Wang *et al*. (2016) found that the total flavonoid content of the archegoniophore was ten times that of the thallose gametophyte. This correlated with greater bio-activity in the archegoniophore, potentially protecting the reproductive structure from bacteria, fungi, and perhaps even herbivory.

Most of the work on antibacterial activity by secondary compounds in bryophytes has been done on human pathogens. The question remains, what can they do for bryophytes? De *et al*. (2015) noted that bryophytes lack the mechanical protections available to tracheophytes and that instead use secondary metabolites as protectants. If such is the case, then we should see greater selection for higher concentrations in bryophytes from habitats where there are more herbivores or pathogens present. However, few studies have attempted to address this hypothesis. De and coworkers compared secondary compounds from ***Marchantia polymorpha*** (Figure 1-Figure 12) from five different altitudes in Darjeeling Himalayas. To my surprise, they found higher antibacterial activity at higher altitudes. This suggests to me that their antibacterial activity is not the most important factor operating in selection for these secondary compounds.

Protozoa Interactions

Protozoa are also inhibited by extracts from ***Marchantia polymorpha*** (Figure 1-Figure 12). Jensen *et al*. (2012) found that marchantin A extracted from this liverwort inhibited the proliferation of the protozoan ***Plasmodium falciparum*** (Figure 189). ***Trypanosoma brucei rhodesiense*** (Figure 190), ***T. cruzi*** (Figure 191), and ***Leishmania donovani*** (Figure 192), all human pathogens, likewise experienced inhibition by marchantin A.

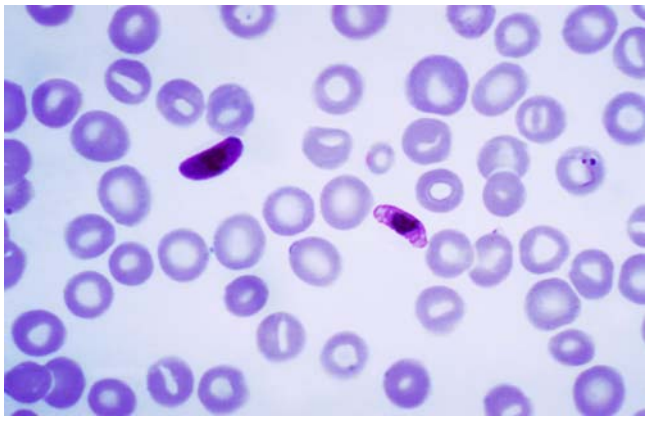

Figure 189. ***Plasmodium falciparum***, protozoan inhibited by marchantin A extracted from ***Marchantia polymorpha***. Photo from CDC, through public domain.

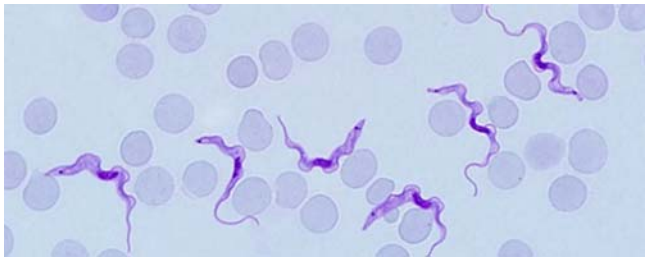

Figure 190. ***Trypanosoma brucei***, protozoan inhibited by marchantin A extracted from ***Marchantia polymorpha***. Photo from CDC, through public domain.

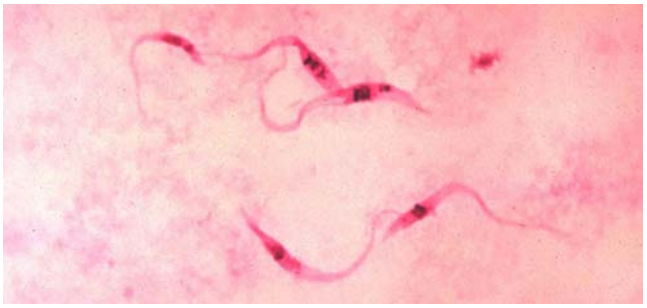

Figure 191. ***Trypanosoma cruzi***, protozoan inhibited by marchantin A extracted from ***Marchantia polymorpha***. Photo by Dr. Myron G. Schultz, CDC, through public domain.

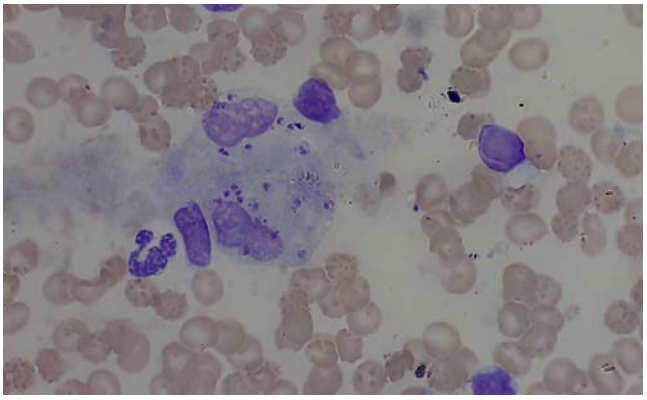

Figure 192. ***Leishmania donovani***, protozoan inhibited by marchantin A extracted from ***Marchantia polymorpha***. Photo by Ajay Kumar Chaurasiya, through Creative Commons.

Fungal Interactions

The associations of fungi with bryophytes was a long-neglected topic in bryology. In fact, many seemed to assume that fungi could not live with bryophytes, or that the bryophytes had too low a nutrient concentration to be of value to the fungi. But this view has now been disproved.

endophytes

Döbbeler (1979) found fungal infections in the rhizoids of ***Marchantia polymorpha*** (Figure 1-Figure 12). Duckett and Ligrone (2003) further reported that the living smooth rhizoids of **Marchantiales** contained the hyphae of fungal endophytes (Figure 193). Benkert (1998) reported a fungus (***Octospora inthacaensis***) growing on ***M. polymorpha***. But what does this relationship mean for the bryophyte?

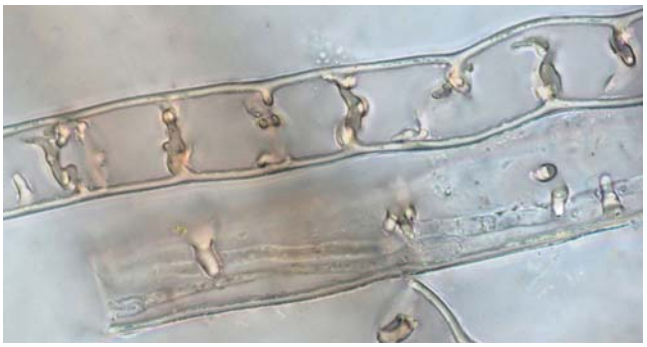

Figure 193. Rhizoids of ***Marchantia polymorpha*** with fungus in lower rhizoid. Photo by Walter Obermayer, with permission.

Döbbeler (2002) reported that the Ascomycetes (fungi) growing on the gametophytes of bryophytes typically did not form ascomata on general locations on their bryophyte hosts, but instead occupied distinct locations that were specific for the host species. These microsites usually offered protection against rapid water loss by the fungus and permitted enough exposure to permit unhindered spore dispersal of the fungus by air currents. Among the thallose liverworts, the ascomata were typically immersed in the spongy thalli. But what do they do to or for the liverwort?

Fungal actions may be subtle, and certainly fungi can inhabit ***Marchantia polymorpha*** (Figure 1-Figure 12). Guminska and Mierzenska (1992) reported that the fungus ***Loreleia marchantiae*** (=*Gerronema marchantiae*; Figure 194) was associated with this species and with ***Nostoc*** sp. (**Cyanobacteria**).

Figure 194. ***Loreleia marchantiae*** growing with ***Marchantia polymorpha***. Photo by Alexey Sergeev, with permission.

Nelson and Shaw (2019) found a very diverse fungal community that was distinct between patches of ***Marchantia polymorpha*** (Figure 1-Figure 12). Only a few core fungi were the same across widely separated populations across the USA. However, they found that the two methods used detected different species.

symbiosis?

Trees are usually dependent on fungal partners in their roots as a means of scavenging nutrients from a much larger soil volume than that available to roots. Hanke and Rensing (2010) considered that ***Marchantia polymorpha*** (Figure 1-Figure 12), with what is now a sequenced genome, provided many genetic tools by which to establish the association of fungi such as ***Glomus intraradices*** with gametophyte plants, including culturing, infection strategies, and staining procedures.

Marchantia species, including ***M. polymorpha*** ssp. ***montivagans*** (Figure 34) (Ligrone *et al*. 2007), often form mycorrhizal associations (Bowman *et al*. 2016). However, such associations are not known in ***M. polymorpha*** ssp. ***polymorpha*** or ssp. ***ruderalis*** (Figure 6-Figure 8, Figure 17-Figure 19). Bowman and coworkers (Bowman *et al*. 2017) attribute this independence to increased transport capacity in the subspecies, permitting them to be weedy colonizers of newly disturbed habitats.

pathogens

Certainly not all fungi have a friendly relationship with the bryophytes. Verkley *et al*. (1997) reported the ascomycete ***Bryoscyphus atromarginatus*** (Figure 195) as a new species parasitizing the thallus of ***Marchantia polymorpha*** (Figure 1-Figure 12) in the Netherlands. The type specimen of the species named as ***Bryoscyphus marchantiae*** (Figure 196) was actually collected from ***Reboulia haemisphaerica*** (Figure 197). The description given by Naumov (1964) of the collection he reported as ***Hymenoscyphus marchantiae*** (Figure 198) on ***M. polymorpha*** agrees well with the new species described by Verkley *et al*. Naumov commented that it appeared that this could be a new species.

Figure 195. ***Bryoscyphus atromarginatus*** growing on thallus of ***Marchantia***. Photo ©Michel Hairaud, through Creative Commons.

Figure 196. ***Bryoscyphus marchantiae*** on ***Marchantia***. Photo ©Iain Munro, through Creative Commons.

Figure 197. ***Reboulia hemisphaerica***, apparently also another host substrate for ***Bryoscyphus atromarginatus***. Photo by Dale A. Zimmerman Herbarium, Western New Mexico University, with permission.

Figure 198. ***Hymenoscyphus kathiae*** on submerged twig of *Alnus glutinosa*; ***Hymenoscyphus marchantiae*** can inhabit ***Marchantia polymorpha***. Photo ©Nick Aplin, through Creative Commons.

Fraiture and Ertz (2007) reported that the fungus ***Didymosphaeria marchantiae*** (Figure 199) was a parasite on ***Marchantia polymorpha*** (Figure 1-Figure 12).

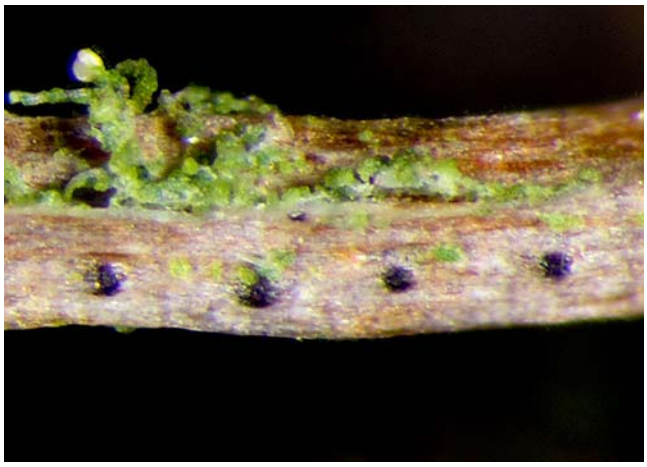

Figure 199. ***Didymosphaeria marchantiae*** infecting leafy liverwort, but also known to be a parasite on ***Marchantia polymorpha***. Photo by Dragiša Savić, with permission.

Nelson (2017; Nelson *et al*. 2018) noted the lack of studies on fungal endophyte interactions with bryophytes while at the same time realizing that the reactions of the plants to these organisms is quite varied. They found that such fungi in ***Marchantia polymorpha*** (Figure 1-Figure 12) ranged from "aggressively pathogenic to strongly growth-promoting." Nevertheless, most of them seemed to cause no change in host growth. Furthermore, those that promoted growth were dependent on nutrient concentrations and their effects on growth were inhibited by inoculation of the liverwort with multiple fungi. Some of the fungi that are known as pathogens in tracheophytes were actually beneficial to the liverworts.

With the large arsenal of secondary compounds, we should consider it to be likely that some of these are effective against fungal pathogens. Takikawa *et al*. (2014) inoculated several powdery mildews on the thallus and gemmae (Figure 153-Figure 165) of ***Marchantia polymorpha*** (Figure 1-Figure 12). The conidia and germ tubes of ***Erysiphe trifoliorum*** (Figure 200) were destroyed. The germ tube tip was destroyed in four hours when it reached the gemma leaf surface. After six hours the conidial bodies were destroyed as well. On the other hand, ***Oidium neolycopersici*** (see Figure 201) continued growth with no destruction of conidia and produced normal appressoria on the surface of the gemmae.

Figure 200. ***Erysiphe trifoliorum*** on ***Trifolium pratense*** leaves; this fungus is destroyed when inoculated on thallus or gemmae of ***Marchantia polymorpha***. Photo by John Plischke, through Creative Commons.

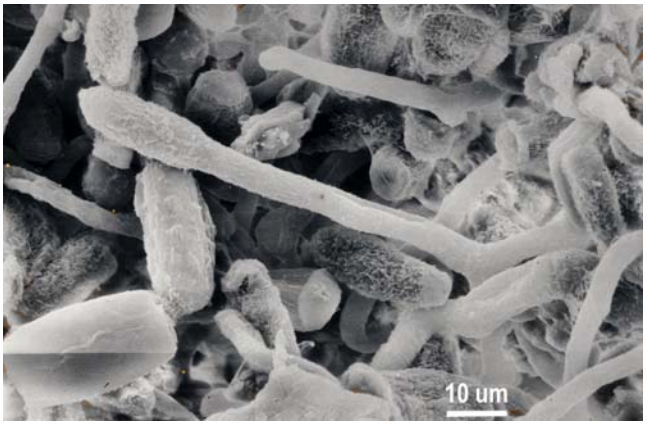

Figure 201. SEM of ***Oidium*** sp; ***Oidium neolycopersici*** continued growth with no destruction of conidia and produced normal appressoria on surface of gemmae of ***Marchantia polymorpha***. Photo by Gerald Holmes, through Creative Commons.

Peumans *et al*. (2007) found the surprising result that a **lectin** (protein that binds to carbohydrates – features that lectins use to defend plants in nature may cause problems during human digestion; lectins resist being broken down in gut and are stable in acidic environments, features that protect lectin-containing plants in nature) identified in ***Marchantia polymorpha*** (Figure 1-Figure 12) had never been found in any plant, but that instead it closely resembles one from the common edible mushroom ***Agaricus bisporus*** (Figure 202), *i.e.*, it is a lectin that was hitherto considered to be exclusive to fungi. It was also confirmed in the moss ***Syntrichia ruralis*** (Figure 203).

Figure 202. ***Agaricus bisporus***, common edible mushroom that has lectin similar to one found in ***Marchantia polymorpha***. Photo by Мыць Денис, through Creative Commons.

Figure 203. ***Syntrichia ruralis***, species that shares same lectin as that found in ***Marchantia polymorpha***. Photo by Alexis Orion, through Creative Commons.

Carella *et al*. (2019) explored the molecular response of ***Marchantia polymorpha*** (Figure 1-Figure 12) to oomycete infection. These early land plants necessarily had to defend against pathogens with their new exposure to land. We know of many pathogens and defenses in tracheophytes, but we know little about the plant-pathogen interactions in these early land plants. Whereas we have explored their medical use and even uses as fungicides for tracheophytes, we know relatively little about their functions in the bryophytes that produce them. Carella and coworkers traced the response of this liverwort to the oomycete pathogen ***Phytophthora palmivora*** (Figure 204). They found that the liverwort shared a set of orthologous microbe-responsive genes with tracheophytes. These include members of the phenylpropanoid metabolic pathway. Using both knockout and induction techniques, they determined that MpMyb14 leads to the accumulation of anthocyanin-like pigments (**auronidin**) while greatly enhancing the resistance of ***Marchantia polymorpha*** to ***Phytophthora palmivora*** infection. The auroidin prevented the penetration of the fungal hyphae into the pigmented portions of the liverwort.

Figure 204. ***Phytophthora palmivora*** infecting papaya; ***Marchantia polymorpha*** was used to identify the mechanism of resistance to this plant pathogen. Photo by Scot Nelson, through Creative Commons.

Gahtori and Chaturvedi (2011) used methanol and chloroform extracts to test the activity of ***Marchantia polymorpha*** (Figure 1-Figure 12) against three bacterial and four fungal species. The extracts exhibited antimicrobial activity with potency that differed among organisms that were pathogenic to both plants and animals. Some were inhibitory toward multiple organisms, and others showed potential.

In other experiments, Mewari and Kumar (2011) made similar tests and found that ***Marchantia polymorpha*** (Figure 1-Figure 12) completely inhibited the mycelial growth of the fungal pathogen ***Rhizoctonia solani*** (Figure 205). Furthermore, most extracts also caused 100% inhibition of spore germination of fungal pathogens ***Alternaria solani*** (Figure 206) and ***Fusarium oxysporum*** (Figure 207).

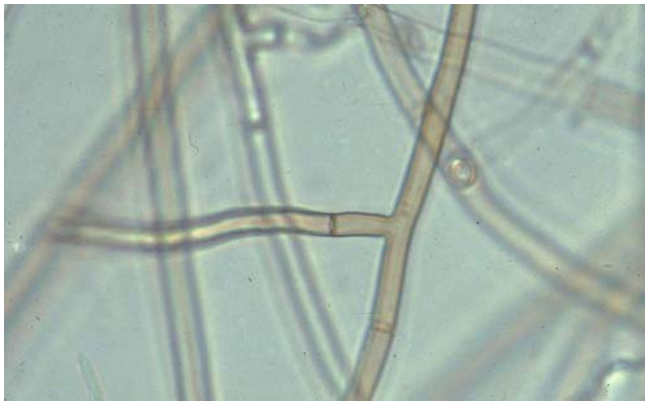

Figure 205. ***Rhizoctonia solani***, fungal species that is completely inhibited by extracts of ***Marchantia polymorpha***. Photo by Gerald Holmes, through Creative Commons.

Figure 206. ***Alternaria solani*** on tomato leaf. Extracts of ***Marchantia polymorpha*** completely inhibit spore germination of this fungal pathogen. Photo from USDA Cooperative Extension, through Creative Commons.

Figure 207. ***Fusarium oxysporum*** on *Cucumis sativa*; extracts of ***Marchantia polymorpha*** completely inhibit spore germination of this fungal pathogen. Photo by Jerzy Opioła, through Creative Commons.

In China, Niu *et al*. (2006) isolated seven bis[bibenzyl]-type macrocycles, including three new ones. They assessed their antifungal activities against ***Candida albicans*** (Figure 183-Figure 184), using TLC bioautography. This fungal species is an opportunistic pathogenic yeast that is the most prevalent cause of fungal infections in humans. Several of the compounds proved to be active against this fungus.

Purkon *et al.* (2022) reported the medicinal use of ***Marchantia*** in China, North America, Ancient Greece, and Indonesia for treatment of open wounds, burns, hepatotoxicity (damage to liver caused by medicine, chemical, or herbal or dietary supplement), and infection prevention.

Matsui *et al*. (2020) found antagonism between salicylic acid and jasmonate in the fungal pathogen interaction with ***Marchantia polymorpha*** (Figure 1-Figure 12). They isolated ***Bjerkandera adusta*** (Figure 208), ***Irpex lacteus*** (Figure 209), and ***Phaeophlebiopsis peniophoroides*** (Figure 210) from diseased ***M. polymorpha***. They found that salicylic acid promotes infection by ***I. lacteus***, but this action is suppressed when jasmonate is treated at the same time.

Figure 208. ***Bjerkandera adusta***, species that has been isolated from diseased ***Marchantia polymorpha***. Photo by James K. Lindsey, with permission.

Figure 209. ***Irpex lacteus***, species that has been isolated from diseased ***Marchantia polymorpha***. Jasmonate can stop the infection. Photo by Otto Miettinen, through Creative Commons.

Figure 210. *Phaeophlebiopsis ravenelii*; ***Phaeophlebiopsis peniophoroides*** is a species that has been isolated from diseased ***Marchantia polymorpha***. Photo by James K. Lindsey, with permission.

Hipol and Broñola-Hipol (2016) screened 22 fungal associates of ***Marchantia polymorpha*** (Figure 1-Figure 12) for their carboxyl esterase activity. Half were endophytes and half were epiphytes on the liverwort. All of the isolates produced this enzyme, with the fungus ***Colletotrichum boninense*** (see Figure 211) producing the lowest levels of the enzyme and ***Nodulisporium*** sp. (Figure 212) produced the highest levels.

Figure 211. *Colletotrichum lindemuthianum*; ***Colletotrichum boninense*** produced lowest levels of carboxyl esterase activity among 22 fungi isolated from association with ***Marchantia polymorpha***. Photo by David B. Langston, through Creative Commons.

Figure 212. ***Nodulisporium cecidiogenes*** on rotten wood; species of ***Nodulisporium*** produced highest levels of carboxyl esterase activity among 22 fungi isolated from association with ***Marchantia polymorpha***. Photo by Alexey Sergeev, with permission.

Poveda (2020b) asked if the arbuscular-mycorrhizal fungal (AMF) association with ***Marchantia polymorpha*** ssp. ***ruderalis*** (Figure 6-Figure 8) was beneficial or harmful. Despite all the studies on this species, this is one of the many questions remaining unanswered. They found that *in vitro*, the interaction is detrimental, causing reduced growth and tissue viability, with only those elements involved in plant defenses increasing in nutritional content. These changes were coupled with increases in reactive oxygen species (ROS) content. One such fungus is ***Rhizophagus fasciculatus*** (Figure 213) – a species present only when there is evidence of thallus damage. Hence, this fungus appears to be a pathogen to ***Marchantia polymorpha*** ssp. ***ruderalis***. Further examination of fungi with bryophytes could reveal interesting symbiotic relationships, modes of infection, and defenses against fungi. ***Marchantia polymorpha*** (Figure 1-Figure 12) is large enough, cultures easily, and is known to respond in a variety of ways to fungal invaders. Hence, it would be a good initial test organism to help us understand these relationships and their effects in their ecosystem.

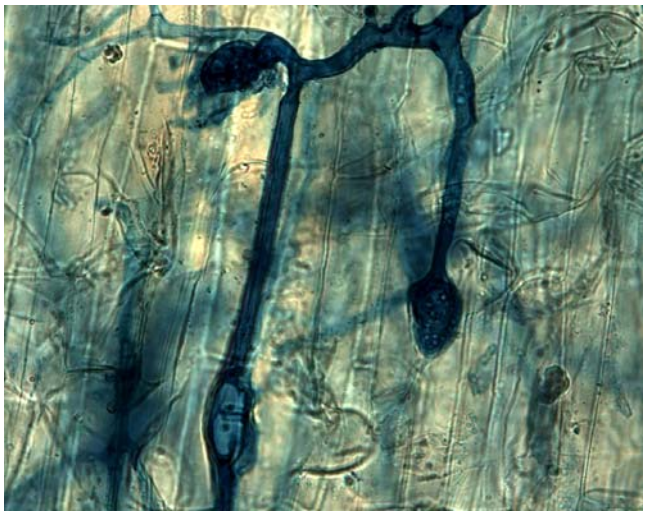

Figure 213. *Rhizophagus irregularis*; ***Rhizophagus fasciculatus*** is fungal pathogen on ***Marchantia polymorpha*** ssp. ***ruderalis***. Photo by Mylène Durant, through Creative Commons.

Invertebrate Interactions

Herbivory (Figure 214) on liverworts seemed to be a blind spot in older bryological studies, but recent reports tell us it is real. Sawangproh *et al*. (2016) reported the feeding of ***Scatopsciara cunicularius*** (**Diptera**: **Sciaridae**) larvae on ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that larvae fed at 12ºC fed more slowly than those at 22ºC, but those at the lower temperature fed over a longer period of time, ultimately causing more damage to the liverwort. The researchers suggested using these larvae to control the invasion of the liverwort in plant nurseries and greenhouses. I wonder what other plants they eat?

Figure 214. ***Marchantia polymorpha*** female eaten on one of the receptacle fingers. Photo by Li Zhang, with permission.

Koeduka *et al*. (2022) found that allene oxide synthase (AOS) from ***Marchantia polymorpha*** (Figure 1-Figure 12) is important in inhibiting the survival rate and oviposition of the spider mite ***Tetranychus urticae*** (see Figure 215). When mutants lacking the AOS gene were subjected to these spider mites, survival of the mites was greater than in the wild type. Their research indicated that defense system signalling pathways respond to spider mite presence.

Figure 215. ***Marchantia polymorpha*** gemmae cups with mite, possibly spider mite. Photo By Bernard de Cuyper, with permission.

Some invertebrates commonly live among bryophytes, ***Marchantia polymorpha*** (Figure 1-Figure 12) included. A common bryophyte inhabitant is the isopod (Figure 216). In some cases these might actually eat the bryophytes. In other cases, there is no evidence of herbivory. Spiders also build webs (Figure 217), perhaps catching some of the other inhabitants of the thallus community.

Figure 216. ***Marchantia polymorpha*** and isopod; there does not seem to be any evidence of herbivory – yet. Photo by Walter Obermayer, with permission.

Figure 217. ***Marchantia polymorpha*** males with gemmae cups and spider web. Photo by Nancy Leonard, with permission.

The importance of bryophyte herbivory in the ecosystem is virtually unknown. This seems to be particularly true for ***Marchantia polymorpha*** (Figure 1-Figure 12). As the climate shifts and herbaceous plant communities change, it is possible some herbivores could shift to consumption of bryophytes. Few studies have addressed the effects of temperature or precipitation patterns on the interaction of invertebrates with bryophytes.

Tracheophyte Interactions

Little has been written about the interaction between bryophytes and tracheophytes. Bryophytes are not good competitors with tracheophytes because of the small size of

the bryophytes. How, then, do the tiny bryophytes survive in habitats dominated by tracheophytes?

Whittemore (1991) suggested that bryophytes such as those in the **Marchantiales** might be toxic not only to herbivores and pathogens, but also to competing plants such as tracheophytes. On the other hand, Asakawa *et al*. (1982) found that compounds from the tracheophyte family Apiaceae could inhibit the growth of ***Marchantia polymorpha*** (Figure 1-Figure 12).

Nakayama *et al*. (1996) tested the inhibitory effects of lunularic acid and its analogs on the growth of ***Marchantia polymorpha*** (Figure 1-Figure 12), ***Rorippa nasturtium-aquaticum*** (watercress; Figure 218), and ***Phleum pratense*** (timothy grass; Figure 219). The analogs proved to be more inhibitory than lunularic acid, suggesting that the liverworts that produce this hormone might have less growth inhibitory activity than tracheophytes, or they are somehow protected from its inhibitory effects.

Figure 218. ***Rorippa nasturtium-aquaticum***, plant that has less inhibition by lunularic acid than more advanced analogs. Photo by Matt Lavin, through Creative Commons.

Figure 219. ***Phleum pratense***, species that has less inhibition by lunularic acid than more advanced analogs. Photo through Creative Commons.

Sharma *et al*. (2009) used ***Marchantia polymorpha*** (Figure 1-Figure 12) water extracts as one of the bryophyte species they tested for allelopathic effects on the flowering plant ***Bidens biternata*** (Figure 220). Although there was 100% inhibition of seed germination in the lipophilic extract, germination was not delayed significantly in water extracts. Sharma and coworkers reasoned that hydrophilic allelochemicals would be released more easily into the environment, but that these would also leach from the soil more quickly, thus favoring hydrophobic allelopathic compounds as a better defense.

Figure 220. ***Bidens biternata***, species for which seed germination is 100% inhibited by lipophilic extracts of ***Marchantia polymorpha***, but not by water extracts. Photo by J. M. Barg, through Creative Commons.

Kaihara and Takimoto (1990) found that a water extract of ***Marchantia polymorpha*** (Figure 1-Figure 12) could inhibit the flower-inducing activity of L-pipecolic acid. This L-pipecolic behaved synergistically with water extracts of ***Lemna aequinoctialis*** (duckweed; Figure 221) and ***Ipomoea hederacea*** (ivy-leaved morning glory; Figure 222) to enhance flowering, but all other tested plants suppressed it. This flowering inhibition is a sneaky way for the liverwort to compete with tracheophytes, but we have no field assessment of its effectiveness.

Figure 221. ***Lemna aequinoctialis***, plant for which its water extracts work synergistically with L-pipecolic to enhance flowering. Photo by Kevin Thiele, through Creative Commons.

Figure 222. ***Ipomoea hederacea***, plant for which its water extracts work synergistically with L-pipecolic to enhance flowering. Photo by Bobby Hattaway <www.discoverlife.org>, with online permission.

Marchantia polymorpha (Figure 1-Figure 12) can even become a pest in domestic gardens. The need to control weeds can provide new spaces where this liverwort can easily invade through gemmae or spores brought to the surface by the disturbance or as companions when new plants are introduced. Callaghan (2009) assessed domestic gardens in Britain and found that more than 80% of them included six moss species, but that ***Marchantia polymorpha*** was present in only 30% of the gardens. Nevertheless, Caron (1972) emphasized the need to fight such bryophytes as ***Marchantia polymorpha*** in arboriculture.

At least some herbicides tend to have different effects on different groups of plants. Bryophytes are no exception. Balcerkiewicz and Rusinska (1987) found that ***Marchantia*** actually expanded its populations in areas treated with herbicides. This may be due to the elimination of the competing tracheophytes, but the possibility existed that the herbicides could actually promote the growth of the liverworts. Iwata *et al*. (1992) subsequently suggested that ***Marchantia polymorpha*** (Figure 1-Figure 12) suspension cultures would be an excellent system for herbicide assays because of the rich chlorophyll content of the liverwort and its ability to grow in both **mixotrophic** (deriving nourishment from both autotrophic and heterotrophic mechanisms) and **autotrophic** (producing complex organic compounds using carbon from simple substances such as carbon dioxide) conditions.

Biochemistry

In a species that has been fundamental in so many studies to represent bryophytes, it is not surprising that there have been many biochemical studies as well. Those included here are only representative of the many publications.

Konno *et al*. (1987) isolated three classes of pectic polysaccharides from ***Marchantia polymorpha*** (Figure 1-Figure 12): rhamnogalacturonan polymer class, glucose rich polymer class, and galacturonan core. The cell walls are low in arabinosyl residues, unlike those of flowering plants.

Boisselíer-Dubayle and Bishler (1989) reported on the presence of esterases, peroxidases, acid phosphatases, and glutamate-oxaloacetate transaminase as revealed by their electrophoretic studies on ***Marchantia polymorpha***. Izumi *et al*. (1995) identified esterase secreted from suspension cell culture of ***Marchantia polymorpha***.

flavonoids

Singh *et al*. (1987) confirmed the presence of saponins, tannins, and flavonoids in ***Marchantia polymorpha*** (Figure 1-Figure 12). However all tests for alkaloids were negative, thus eliminating one of the chemical groups used in antiherbivory in some tracheophytes.

Markham and Porter (1974) identified major flavonoids of ***Marchantia polymorpha*** ssp. ***polymorpha***. Flavonoids exhibit properties of antiherbivory and UV filters (Johnson 1983; Treutter 2006). Treutter (2006) reviewed the literature on their roles in plants and found that they act as stress protecting agents, attractants, or feeding deterrents, and have a significant role in plant resistance.

Flavonoids are important compounds that enabled the first land plants to interact with their environment (Davies *et al*. 2020). Hence, it is hypothesized that the flavonoid pathway must have evolved during the colonization of land by early plants, about 450 million years ago, providing essential protection against abiotic stress (Albert *et al*. 2018). The flavonoids are important reactants that permitted early plants to tolerate both abiotic and biotic stresses. Their production in plants can be induced by cold, UV-B light, strong white light, nutrient deficiency, desiccation, salinity, metal toxicity, senescence, and attack by pests and pathogens (Agati & Tattini 2010; Cheynier *et al*. 2013; Landi *et al*. 2015; Davies *et al*. 2018, 2020). They provide signals to microbes, serve as **allelochemicals** (chemical produced by living organism, exerting detrimental physiological effect on individuals of another species when released into environment), and can be important **nutraceuticals** (any food substance that provides medical or health benefits, including prevention and treatment of disease) in the animal diet (Taylor & Grotewold 2005).

Kubo *et al*. (2018) demonstrated that the regulation of gene expression as a stress response was already present in ***Marchantia polymorpha*** (Figure 1-Figure 12). They found that overexpression of one regulatory gene greatly increased the amount of riccionidins, a flavonoid. The gene was up-regulated by UV-B irradiation, nitrogen deficiency, and NaCl treatment.

Clayton *et al*. (2018) monitored the biosynthesis of flavonoids in ***Marchantia polymorpha*** (Figure 1-Figure 12), using three different UV-B regimes and mutant cultures. They found that the **chalcone** isomerase-like compound was one of greater importance. Mutants with a disrupted pathway for this enzyme were more easily damaged by UV-B. Those mutants with increased flavonoid content demonstrated greater UV-B tolerance.

The flavonoid pathway starts with **chalcones** as the first flavonoids (Davies *et al*. 2020) Since their origin, more than 8,000 different flavonoid structures have been reported (Andersen & Markham 2006). The major flavonoid classes are the flavones, flavonols, isoflavonoids, aurones, 3-deoxyanthocyanins, anthocyanins, proanthocyanidins (condensed tannins), and auronidins (Davies *et al*. 2020). Most flavonoids go to the vacuoles as water-soluble glycosides, but in some species they are

transported to the cell wall or are released to the environment.

Many flavonoids can absorb UV light, but the colored anthocyanins and auronidins can screen visible light (Lee & Gould 2002; Landi *et al.* 2015; Berland *et al.* 2019). Thus far, flavonoids have not been found in hornworts. They seem to have no role in development in bryophytes; a mutant of ***Marchantia polymorpha*** (Figure 1-Figure 12) lacking flavonoids has normal developmental patterns (Clayton *et al*. 2018).

In ***Marchantia polymorpha*** (Figure 1-Figure 12), nitrogen deprivation and increased white light exposure both induce the accumulation of auronidin (Albert *et al*. 2018; Kubo *et al.* 2018), a phenomenon similar to that of seed plants for anthocyanin accumulation in *Arabidopsis* and apples (Rubin *et al.* 2009; Wang *et al.* 2018). But in ***M. polymorpha*** auronidin also greatly increases the resistance of the plants to infections by ***Phytophthora palmivora*** (Figure 204), with hyphae apparently unable to penetrate into highly pigmented regions of the plant (Carella *et al*. 2019). Thus their roles are widespread.

Berland *et al*. (2019) reported **auronidins** for the first time. These flavonoid pigments seem to be important in protecting the plants from such environmental stresses as high light, drought, and nutrient deprivation. We initially thought that the red pigments bound in the cell walls of the early land plants were anthocyanins, but recent studies have revealed that they are in fact a group of phenylpropanoids that Berland and coworkers named **auronidins**. Their colors are similar to those of anthocyanins, but they are synthesized differently and have different optical properties. It appears that they contribute to the ability of ***Marchantia polymorpha*** (Figure 1-Figure 12) to survive extreme environments.

Excess light of any quality enhances the biosynthesis of flavonoids in plants, performing multiple functions at the expense of the antioxidant flavonoids and hydroxycinnamates (Agati & Tattini 2010). Several research groups have provided indications that common oxidative signal components may up-regulate flavonoid biosynthesis, regardless of their origins (Taylor & Grotewold 2005; Fujita *et al.* 2006; Quattrocchio *et al.* 2006) and may link the REDOX potential of the cell to the control of flavonoid accumulation (Taylor & Grotewold 2005). It appears that the main purpose of the flavonols is their involvement in responses to abiotic and biotic stresses (Roberts & Paul 2006; Kilian *et al*. 2007; Mellway *et al*. 2009). More research is needed on their involvement in reducing oxidative stress.

Using knockout genes, Albert *et al*. (2018) determined that all pigmentation was lost from the flavonoid riccionidin A in ***Marchantia polymorpha*** (Figure 1-Figure 12), but when overexpression was used these plants produced large amounts of flavones and riccionidin A and exhibited red pigmentation. Light- and nutrient-deprivation stress induced flavonoid pigmentation in the thallus, as these stresses do for the anthocyanins in flowering plants.

In evolutionary theory, red leaves represent a signal of the health status of a tree, providing a signal to insects to migrate when trees change color in autumn (Cheynier *et al* 2013). Could similar signals be active in bryophytes, with red plants signalling nutrient deficiency or other poor health condition? Red can also signal toxicity and is known as **warning coloration**.

phenolics

From the beginning of my interest in antiherbivory, I understood that phenolics were the important antiherbivore compounds in plants. But Close and McArthur (2002) challenged that thinking. They contend that it is not antiherbivory, but photoprotection, that makes these compounds so important for plants. They suggest that phenolics accomplish this role by acting as antioxidants and that their levels may vary with environmental conditions to provide this protection. Thus, their level of phenolics in the plants is dependent on the risk of photodamage and not on resources in the environment.

Cheynier *et al*. (2013) noted that for the successful colonization of land plants needed UV light screens. These were apparently achieved by phenolic compounds. These compounds play no role in the developmental and growth processes, but they are vital for survival in the interaction of the plants with their environment, for their reproductive strategy, and reputedly, for their defense. These survival mechanisms are controlled by plant phenolics that respond to potentially overlapping regulatory signals. Some of these effects are likewise associated with the growth hormone auxin.

Soriano *et al*. (2019b) found that bryophytes rarely exhibited a quick response to UV radiation in their production of UV-absorbing compounds. They experimented with ***Marchantia polymorpha*** (Figure 1-Figure 12) and measured the phenolic content under three realistic UV levels on day 1 and day 22 (Figure 223). The levels of UV-absorbing compounds mostly responded with linear or hyperbolic relationships with the UV level (Figure 224). They identified thirteen flavones (apigenin and luteolin derivatives) and two hydroxycinnamic acids (p-coumaric and ferulic acids) in the soluble and insoluble fractions, respectively. The speed of response depended on the compound, but those identified in ***M. polymorpha*** were slow responders.

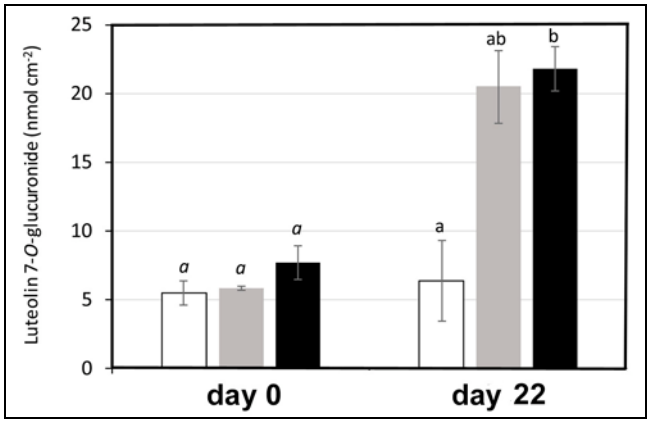

Figure 223. UV-absorbing compounds and time. White bars = UVBE low; grey bars = medium; black bars = high. Letters that are same represent means that are not significantly different from each other. Image modified from Soriano *et al*. 2019b.

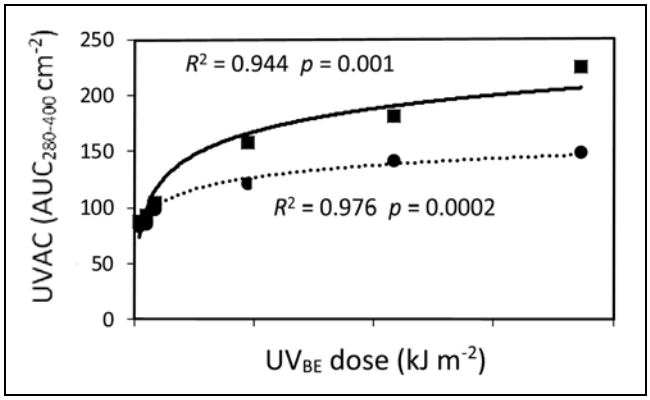

Figure 224. ***Marchantia polymorpha*** phenolics produced with increasing levels of UV-B. Photo modified from Soriano *et al*. 2019b.

light effects

Roberts and Paul (2006) introduced the concept that light serves to modulate plant defenses, in some cases being essential for the development of that resistance. They suggest that this interaction is multifaceted, extending across both temporal and biological scales. This needs further exploration in bryophytes and might explain our (false?) assumption that phenolic compounds have antiherbivore properties. Perhaps they accomplish both light protection and antiherbivory.

phenolics - phrenanthenes

Adam and Becker (1993a) reported phenanthrenes and other phenolics from cultured ***Marchantia polymorpha*** (Figure 1-Figure 12). Phenolics are secondary compounds that were widely considered to be antiherbivory compounds, but now the their importance in antiherbivory is questionable. Phenathrenes, on the other hand, have been used in traditional medicine, including usefulness in cytotoxicity, antimicrobial, spasmolytic, anti-inflammatory, antiplatelet aggregation, antiallergic activities, and phytotoxicity (Kovács *et al*. 2008), suggesting that they could serve as antiherbivore compounds as well as antibiotics. But perhaps their role in allelopathy toward tracheophytes is also feasible.

Anton *et al*. (1997) also found phenanthrene in the leafy liverwort ***Plagiochila*** (Figure 225), suggesting that it might be somewhat widespread in liverworts. Phenanthrene seems to have an interesting relationship with plants. It is one of the PAHs – phenols that occur naturally in coal, crude oil, and gasoline. PAHs are released from burning coal, oil, gas, wood, garbage, and tobacco and are toxic to both plants and animals (Wei *et al*. 2014). For example, phenanthrene inhibits seed germination and affects growth and chlorophyll levels of wheat seedlings. They also decrease the effects of antioxidants. Dupuy *et al*. (2016) also found that phenanthrene exposure causes developmental perturbation in maize roots. In the environment, they are degraded by bacteria (Anton *et al*. 1997). Corgié *et al*. (2003) demonstrated that root exudates are able to stimulate the degradation of PAHs in soil. In fact, it appears that the presence of the phenanthrene stimulates the release of more of these exudates, thus increasing the bacterial populations that accomplish the breakdown of the phenanthrene (Muratova *et al*. 2009). Thomas *et al*. (2019) also noted that plants can stimulate microbial degradation of PAHs, using phenanthrene as the test compound. The soil bacteria **Proteobacteria** (**Pseudomonadota**; Figure 16), **Actinobacteria** (Figure 226), and **Firmicutes** (bacterial groups that are also common on bryophytes; Figure 227) are phenanthrene degraders. Plant root exudates enhanced the development of these phenanthrene-degrading bacteria.

Figure 225. ***Plagiochila strombifolia***; phenanthrene occurs in at least some members of this genus. Photo by John Walter, through Creative Commons.

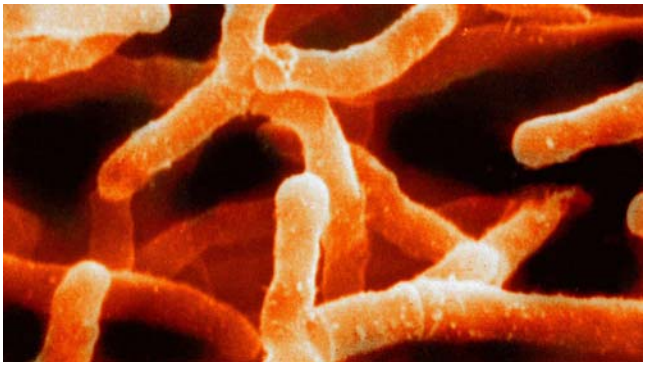

Figure 226. ***Actinomyces israelii***, member of **Actinobacteria**, group often associated with bryophytes and soil; some are phenanthrene degraders. Photo by Graham Beards, through Creative Commons.

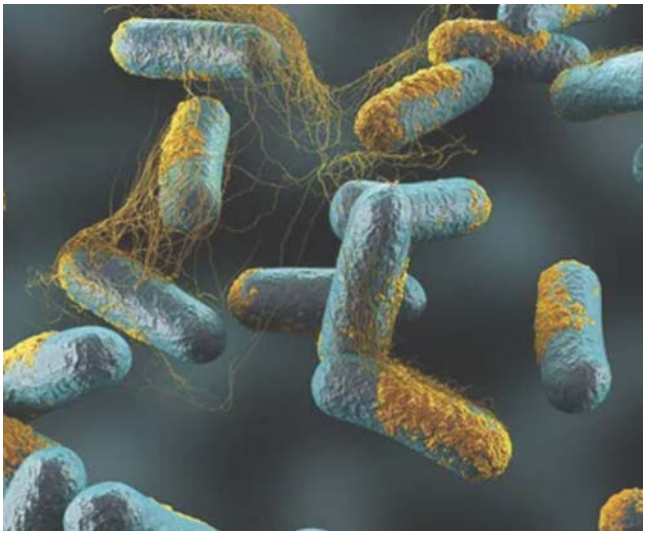

Figure 227. A member of **Firmicutes**, group often associated with bryophytes and soil; some are phenanthrene degraders. Photo by Argonne National Laboratories, through Creative Commons.

This raises the question of the presence of phenanthrenes in liverworts. Does it help, harm, or have a neutral action on them? Or like many of these compounds, might it have multiple roles? Perhaps the work with grassland plants might suggest a possible interaction. Chiapusio *et al*. (2007) found that in grasslands the phenanthrene did not generally affect plant biomass. In fact, red clover biomass was enhanced by it. This apparently resulted by a stimulation of its *Rhizobium* partner, a nitrogen-fixing bacterium. On the other hand, phenanthrene had a drastic negative effect on the mycorrhizal colonization of both ryegrass and red clover. As in other studies, the phenanthrene stimulated the PAH degraders in the soil.

This raises a possibility for ***Marchantia polymorpha*** (Figure 1-Figure 12) and its production of phenanthrene. It could stimulate the bacterial partners that are so common among the bryophytes (see Volume 2 chapter on bacterial interactions). In this role, it could play a critical role in their development. This won't be discovered in a sterile lab culture.

sesquiterpenoids and terpenes

Matsuo *et al*. (1985) isolated a series of ent-sesquiterpenoids that were stereoisomers of those compounds known in the tracheophytes, supporting the ancient origin of many of the defense compounds.

In 1990, Asakawa *et al*. described three new ent-sesquiterpenoids from German populations of ***Marchantia polymorpha*** (Figure 1-Figure 12). The chemistry of this species, including other isolates in this study, suggest that the German populations are close to the Japanese ***M. polymorpha*** and ***M. paleacea*** ssp. ***diptera*** (Figure 228).

Figure 228. ***Marchantia paleacea*** ssp. ***diptera*** females with capsules, Arima, Japan, 7 August 1988 – subspecies that has chemistry similar to that of ***M. polymorpha*** from Germany. Photo by Janice Glime.

Terpenes are volatile unsaturated hydrocarbons that constitute the essential oils and are aromatic compounds found in plants. Kumar *et al*. (2016) noted that despite their ability to accumulate structurally diverse terpenes that are "believed" to serve in deterring disease and herbivory, the genes and enzymes responsible for this chemical diversity of terpenes in ***Marchantia polymorpha*** have never been described. They were able to identify four diterpene synthase genes by function that were related phylogenetically to those in diverged plants. However, there were also nine "rather unusual" monoterpene and sesquiterpene synthase-like genes.

lectins

Adam and Becker (1993b) tested ***Marchantia polymorpha*** (Figure 1-Figure 12) for **lectins**. **Lectins** are proteins that bind to carbohydrates. They are defense compounds in plants, but can cause problems in human digestion because they are able to resist being broken down in the gut and remain stable in acidic environments. In some cases they interfere with the absorption of other nutrients. These capabilities make them good antiherbivore compounds. They are in many human foods, especially dried beans, and those extracted from ***Marchantia polymorpha*** agglutinate the erythrocytes of various mammals and exhibit carbohydrate specificity against complex carbohydrate structures. This was the first report of lectins in liverworts. On the other hand, they are important in attracting specific ***Rhizobium*** species toward roots of host plants, suggesting they could possibly have a similar role in bryophytes.

bibenzyls

Asakawa *et al*. (1987) isolated two new cyclic bis[bibenzyls] from Indian populations of ***Marchantia polymorpha*** (Figure 1-Figure 12). Bis[bibenzyls] are rare products of plants, but more than 125 types have been discovered among liverworts (Asakawa *et al*. 2021). They are biosynthesized from lunularic acid, perhaps explaining why this compound has remained in even those liverworts such as ***Marchantia*** that also have ABA. The known biological activities of cyclic bis[bibenzyls] include antimicrobial, antifungal, cytotoxic, muscle relaxation, antioxidant, tubulin polymerization inhibitory, and antitrypanosomal activities.

Niu *et al*. (2006) isolated and identified seven bis[bibenzyl]-type macrocycles from Chinese populations of ***Marchantia polymorpha*** (Figure 1-Figure 12). Several of these compounds exhibited antifungal activities against ***Candida albicans*** (Figure 183).

The **marchantins** are bis[bibenzyls]. As far as we know, these are unique to bryophytes and are cytotoxic, having cancer treatment applications (Kodama *et al*. 1988). Kámory *et al*. (1995) isolated marchantin A from ***Marchantia polymorpha*** (Figure 1-Figure 12). This was followed later by isolation of a number of different marchantins. At least some of these have proven antibacterial activity.

Fang *et al*. (2007) described three new bibenzyl (=1,1′-(ethane-1,2-diyl)bisbenzene) derivatives from Chinese populations of ***Marchantia polymorpha*** (Figure 1-Figure 12). Its polymorphin A was a new type of bis[bibenzyl] and one compound was described as the first discovery of a bibenzyl that is oxidatively coupled to a phenylmethanol.

Friederich *et al*. (1999) elaborated on the pathway from lunularic acid to formation of marchantin C and CO_2 and the hydroxylation of marchantin C to marchantin A. Both of these reactions depend on the presence of O_2 and NADPH. Both are also inhibited by CO in the dark.

Marchantins are another example in which Kubo *et al*. (2018) demonstrated that the regulation of gene expression as a stress response was already present in ***Marchantia polymorpha***. They found that overexpression of one

regulatory gene greatly increased the amount of several marchantins. The gene was down-regulated by NaCl.

antibacterial

Zehr (1990) found that ether extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12) inhibited bacteria at 84.4%, whereas the ethanol extract lacks inhibitory ability. Those most affected were ***Bacillus subtilis*** (Figure 229) and ***Escherichia coli*** (Figure 16), whereas ***Enterococcus faecalis*** (Figure 230) was least inhibited. Zhu *et al*. (2006) noted that the antibacterial activity of ***Marchantia*** was "particularly prominent." As in many antibacterial studies with bryophytes ***Staphylococcus aureus*** (Figure 180) was the most resistant of the seven bacterial species tested. ***Bacillus subtilis*** was the most sensitive species to liverwort extracts.

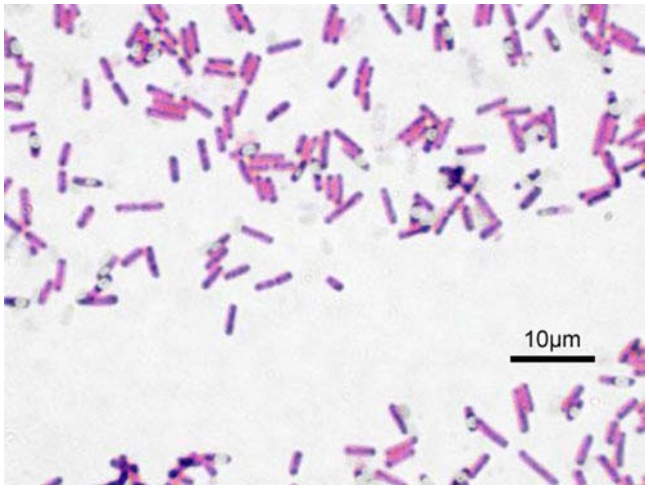

Figure 229. ***Bacillus subtilis*** Gram stained, one of bacteria most affected by extracts of ***Marchantia polymorpha***. Photo by Y. Tambe, through Creative Commons.

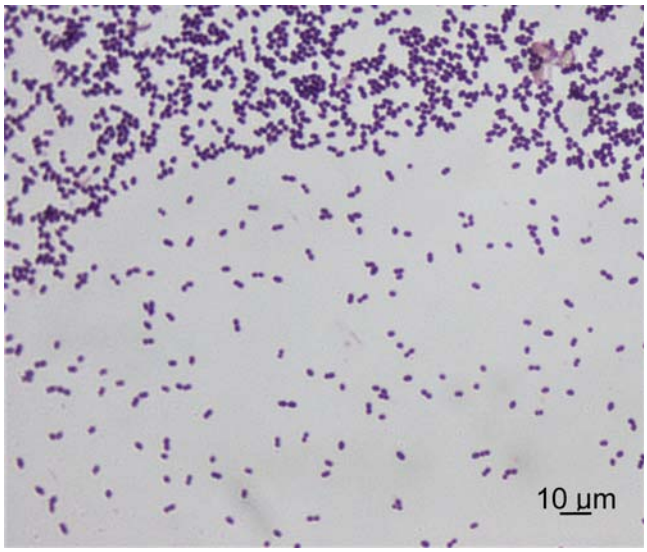

Figure 230. ***Enterococcus faecalis***, one of bacteria least affected by extracts of ***Marchantia polymorpha***. Photo by Dr. Sahay, through Creative Commons.

Gahtori and Chaturvedi (2011) likewise touted the usefulness of extracts of ***Marchantia polymorpha*** (Figure 1-Figure 12) as antimicrobial agents, some killing the organisms and others simply arresting growth. Among those affected were the Gram-negative bacterial strains ***Pasteurella multocida*** (Figure 231), ***Salmonella enterica*** (Figure 232), and ***Xanthomonas oryzae*** pv. ***oryzae*** (Figure 233), and the four fungal strains ***Fusarium oxysporum*** f. sp. ***lini*** (Figure 207), ***Rhizoctonia solani*** (Figure 205), ***Sclerotium rolfsii*** (Figure 234), and ***Tilletia indica*** (Figure 235). They found a unique activity against ***X. oryzae*** and ***P. multocida***. They also acted against the fungi ***S. rolfsii*** and ***F. oxysporum***. ***Marchantia polymorpha*** showed different potencies against micro-organisms that are pathogenic to both plants and animals.

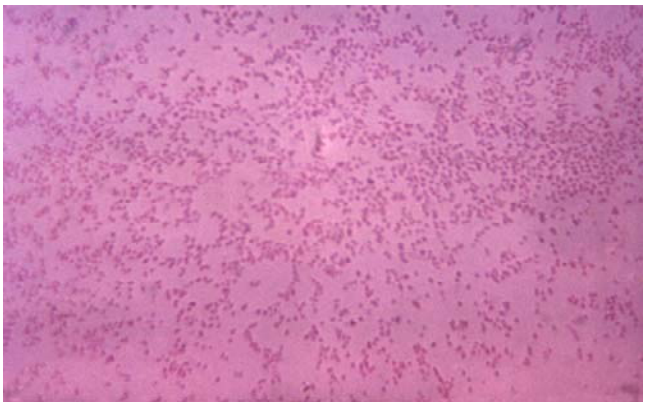

Figure 231. ***Pasteurella multocida*** bacteria, causative agent of fowl cholera, and species that is negatively affected by extracts of ***Marchantia polymorpha***. Photo by Dr. R. Weaver, CDC, through public domain.

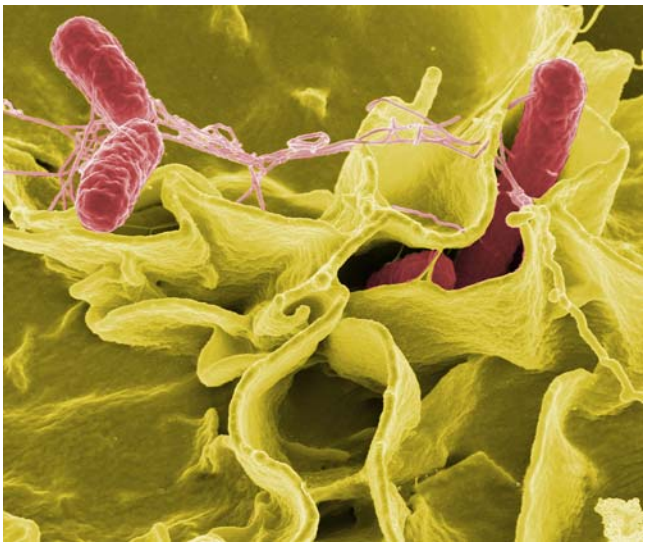

Figure 232. ***Salmonella*** in human tissue, and species that is negatively affected by extracts of ***Marchantia polymorpha***. Photo by NIH, HHS, through public domain.

Figure 233. ***Xanthomonas oryzae***, species that is negatively affected by extracts of ***Marchantia polymorpha***. Photo by Rui map Zheng at <Bugwood.org>, through Creative Commons.

Figure 234. ***Sclerotium rolfsii***, fungal species that is negatively affected by extracts of ***Marchantia polymorpha***. Photo by Bridget Lassiter, NCDA&CS <Bugwood.org>, with online permission.

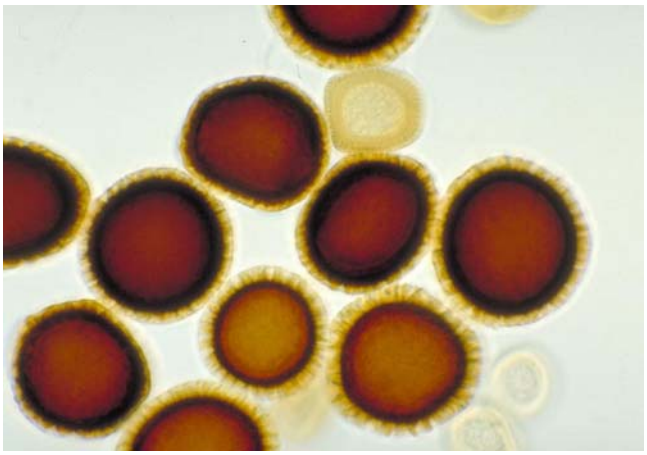

Figure 235. ***Tilletia indica***, fungal species that is negatively affected by extracts of ***Marchantia polymorpha***. Photo by Ruben Durán, through public domain.

antifungal

Hamashima *et al*. (2019) noted the development of many techniques using ***Marchantia polymorpha*** (Figure 1-Figure 12). They used S-Agar Trap to genetically transform the species, using spores. They were able to insert a T-DNA mutant and isolate and characterize a gain-of-function mutant. This mutant hyper-accumulates the flavonoid riccionidin A, verifying that this technique is a powerful tool in the genetic tool library, including production of antibiotics.

In a comprehensive study on published antifungal activities of bryophytes, Miranda *et al*. (2022) found that in the period of 2000 to 2019 ***Marchantia polymorpha*** (Figure 1-Figure 12) was included in the most publications.

glycosides

Qu *et al*. (2007) isolated four new glycosides and three known aromatic glycosides from ***Marchantia polymorpha*** (Figure 1-Figure 12) and described their structures. Many drugs and poisons derived from plants are glycosides, such as the several cyanogenic glucosides used by the Heliconius butterfly that incorporate these plant compounds in their tissues as a defense against predators (Nahrstedt & Davis 1983). I wonder if anything eats bryophytes to gain their secondary compounds for their own defense.

medicinal uses

Dioscorides (De Materia Medica, 50-70 AD) extolled the value of ***Marchantia polymorpha*** (Figure 1-Figure 12) for treating liver ailments, based on its liver form (Doctrine of Signatures; in Schuster 1966). In Yunnan and Sichuan this liverwort is available in street markets (Wu & Yu 2003). It is used to cover the skin for curing jaundice and as an antipyretic. Asakawa (1981, 1982) reported that this species contains sesquiterpenoids that have anti-tumor activity. Hartwell (1982) referred to its use against cancer, referring to Pliny the Elder.

Fischer *et al*. (1995) verified the presence of chalcone synthase activity in ***Marchantia polymorpha*** (Figure 1-Figure 12), and the subsequent presence of naringenin. Chalcones are used medicinally in treatment of viral disorders, cardiovascular diseases, parasitic infections, pain, gastritis, and stomach cancer. **Naringenin** has strong anti-inflammatory and antioxidant activities and seems to be beneficial in treating obesity, diabetes, hypertension, and metabolic syndrome in humans.

Kumar *et al*. (2007) reported that ***Marchantia polymorpha*** (Figure 1-Figure 12) was among those liverworts useful for treating tumors and that it was among the traditional herbal medicines in India.

Culturing and Cultivating

With so many studies being performed on the species ***Marchantia polymorpha*** (Figure 1-Figure 12), the methods of culturing have been worked out well (Figure 236). Voth and Hamner (1940) grew ***Marchantia polymorpha*** on glass cloth in an open moist chamber. They tested 56 nutrient solutions to develop suitable conditions for culturing the species in the lab. Voth (1941) indicated that the omission of K, Ca, NO_3, or PO_4 ions caused differences in the gross appearance of the species. Mg and SO_4 are not indicated by any characteristic symptoms. Effects of calcium appear to be dose dependent. Almost immediate death occurs in its absence, whereas in 0.3 mM of calcium per liter of solution the thallus apices die and degenerate.

Figure 236. ***Marchantia polymorpha*** ssp. ***ruderalis*** in peat disc culture. Photo courtesy of Javier Martínez-Abaigar.

Molar concentrations that are adequate for good growth, based on Voth (1941) are:

K 0.0012 M L^{-1}
Ca 0.0007 M L^{-1}
Mg 0.0014 M L^{-1}
NO_3 0.0034 M L^{-1}
PO_4 0.0004 M L^{-1}
SO_4 0.0008 M L^{-1}

These can be provided in a 1 L, 0.5 M solution of the following forms:

KNO_3 1.6 cc
$Ca(NO_3)_2$ 1.4 cc
$Mg(NO_3)_2$ 1.2 cc
KH_2PO_4 0.8 cc
$MgSO_4$ 1.6 cc

Minor variations of the concentrations do not cause growth problems.

Voth (1943) contributed additional experiments on nutrient concentrations, finding that some were better for production of gemmae cups than others. High salt concentrations often killed the growing tips and thalli became translucent.

Schneider *et al*. (1967) further developed culture methods that attempted to standardize them and address some of the inconveniences of past methods. They found that five substrates worked well, including vermiculite, perlite, glass cloth, nutrient agar, and nutrient solution. These are compared for their maintenance, yields, and usefulness for particular experimentation.

Miller (1964) contributed to the culturing protocol by defining procedures for harvesting, surface sterilizing, and culturing of gemmae. He also described conditions for a high production of gemmae and large numbers of gemmae cups in stock plants. He described methods for culturing the gemmae axenically, noting that they did not fare well at temperatures above 25ºC, but that they were tolerant of high light. Miller *et al*. (1962b) used five different photoperiods to determine the best photoperiod for developing gemmalings. The greatest size and weight were achieved in an 18-hour photoperiod. They experimented with X-rays, light intensity, various nutrients, amino acids, vitamins, and other supplements to determine their effects and ability to prevent damage to the plants.

Miller and Colaiace (1969) cultured gemmae to ultimately produce antheridiophores and archegoniophores, structures that developed after 3-6 weeks on 1% agar medium in 24-hour photoperiods at 23ºC.

In his attempts to grow gemmae of ***Marchantia polymorpha*** (Figure 1-Figure 12), Gemmrich (1976) found that Fe and $Ca(NO_3)_2$ induced germination, but optimal germination occurred on $Ca(NO_3)_2$, KNO_3, and $MgSO_4$. Gibberellic acid failed to induce germination in the dark.

Sugawara *et al*. (1983) found that activated charcoal in the culture medium increases cell wall regeneration and subsequent cell division, suggesting that something in the medium is too strong for he plant, or that the plant's own by-product(s) become inhibitory. Charcoal is usually used to bid things, thus removing them from availability to plants.

Pedroza-Manrique and Caballero Arévalo (2009) recognized that bryophytes typically require lower nutrients than do tracheophytes and algae. They successfully grew ***Marchantia polymorpha*** (Figure 1-Figure 12) propagules in 25% Murashige and Skoog (1962) mineral salt concentration, incubated at 25°C ± 1°C. They warned that when transplanting such cultures to their natural environment or other conditions, one should provide gradual adjustment to new humidity, temperature, and substrate conditions.

Katoh *et al*. (1980) used a modified Murashige and Skoog's medium to culture ***Marchantia polymorpha*** (Figure 1-Figure 12). They improved the medium for use with the liverwort, including only eight of the 24 original micro-organic constituents. This new medium resulted in richer chlorophyll and a higher growth rate in the exponential phase.

Xu *et al*. (2021) found that they could induce reproductive organs in ***Marchantia polymorpha*** (Figure 1-Figure 12) on agar plates. Cultures from gemmae were transplanted after 10 days onto soil at 22ºC with a 16h:8h photoperiod using white light. After 14 days, they supplemented the cultures with far red light.

Gradstein (2006) reported the successful cultivation of ***Marchantia*** in the bryophyte garden of the Cibodas Botanical Garden, Java, Indonesia. Supplementary spray is needed during the dry season.

Control

Marchantia polymorpha (Figure 1-Figure 12) can be a serious weed in some types of gardens, especially in nurseries, and in greenhouses. Jin and Pyon (2007) noted the need to control it in ginseng gardens. Uva *et al*. (1997) even listed it in their publication on the weeds of the northeastern USA. Schofield (1997) listed the species as one obviously spread by human activity in British Columbia, Canada.

Sato *et al*. (1991), working in Japan, used Cyclohexanedione derivatives on ***Marchantia***, causing photosynthetic inhibition in cultured cells. They suggest that indicated the usefulness of the liverwort in herbicide assays.

Fausey (2003) considered ***Marchantia polymorpha*** (Figure 1-Figure 12) to be highly invasive and difficult to control, becoming a concerning pest in ornamental containers. They compared pre-emergence and postemergence herbicides, using chlorothalonil, captan, ammonium chlorides, hydrogen dioxide, flumioxazin, oxyfluorfen, pelargonic acid, acetic acid (vinegar), Cu sulfate, cinnamaldehyde, prodiamine, and oxadiazon. Of these, only flumioxazin, oxyfluorfen, pelargonic acid, acetic acid, and oxadiazon elicited acceptable control. Sprayable preparations were more effective than were granular ones.

Newby *et al*. (2007) experimented with various herbicides as a means of controlling ***Marchantia polymorpha*** (Figure 1-Figure 12) in nursery containers. The effectiveness differed by location, with flumioxazin and oxadiazon being the most effective for control in Alabama, whereas flumioxazin and oxyfluorfen + oryzalin were the most effective for control in Oregon.

Since ***Marchantia polymorpha*** (Figure 1-Figure 12) benefits from the same high humidity and shading as that needed in most nurseries for growing native tree seedlings, it quickly becomes a pest species there (Navas *et al*. 2014). Navas and coworkers used a variety of treatments,

including sterilizing the soil, using three concentrations of acetic acid, using two concentrations of oxygenated water, and using the herbicides glyphosate and fomesafen. They also used pre-emergence application of diuron and trifluralina. Only the two pre-emergence applications caused a 100% control of ***Marchantia polymorpha***.

Särkkä and Tahvonen (2020) suggested several means of control of ***Marchantia polymorpha*** (Figure 1-Figure 12) in nurseries where it appeared as a weed. In pots with growing horticultural plants it can reduce access to water and nutrients (Figure 237). Growth of the liverwort can be minimized with mulches. Särkkä and Tahvonen used ***Sphagnum*** (Figure 238) and 1-cm blackcurrant stem pieces. Highbush blueberry and blackcurrant controlled the liverwort for two years, whereas rhododendron controlled it for only one year. Blueberry and rhododendron require an acid medium that is beneficial for the liverwort. Blackcurrant mulch was nearly 100% effective, whereas other treatments ranged 78-99%.

Figure 237. ***Marchantia polymorpha*** with splash cups, in nursery flower pot. Photo by Janice Glime.

Figure 238. ***Sphagnum capillifolium***; ***Sphagnum*** can be used in layers to discourage growth of ***Marchantia polymorpha*** in pots. Photo by Bernd Haynold, through Creative Commons.

Khamarea *et al*. (2021) used a method of substrate stratification and fertilizer to control ***Marchantia polymorpha*** (Figure 1-Figure 12) growth. They used pine bark and other layers with different physical properties to manipulate the soil moisture dynamics and improve irrigation and fertilizer efficiency. This proved to also work as a tool of weed management. Each of the stratified techniques reduced the liverwort coverage by nearly 100%.

Summary

Marchantia polymorpha is perhaps the most studied bryophyte on the planet. It is a teaching model for liverworts, has had its genome sequenced, and is common in both wet habitats and as an invader after fires. It has been the subject of many physiological studies, including hormone effects and capillary movement of water among rhizoids. Growth hormones such as IAA elicit responses like those in tracheophytes. The thalli exhibit circadian rhythms that regulate the IAA levels. Lipoxygenase also contributes to control of cell growth. Gibberellins respond to photoperiod. Ethylene causes larger gemmae, induces more gemmae cups, and promotes the dormancy of the gemmae, whereas the precursor ACC inhibits gemma growth and development by suppressing cell division. Cytokinins control formation of both gemmae cups and rhizoids during thallus development and seem to influence distribution of air pores and shape of thallus margin, implicating control of cell division. Desiccation causes oxidative stress. ***Marchantia polymorpha*** has both lunularic acid and ABA, important hormones in desiccation tolerance. Light affects senescence, but spermine, spermidine, and putrescine reduce it. Ethylene induces senescence. Nitrate and phosphate deficiency cause the ventral layers of cells to develop a red-violet color. Thalli can grow on vertical surfaces, but gemmae cups exhibit gravitropisms.

Adaptations are a coordinated set of resistance to effects of drying. These include rhizoids, scales, thick thallus, thin cuticle, air pores with a waxy ring, and oil bodies, as well as a suite of biochemical adaptations.

It is dioicous and prolifically produces sexual structures. However, gemmae are usually the most important means of reproduction and enable it to colonize rapidly after a disturbance. Short photoperiods favor production of gemmae, whereas long photoperiods favor the production of gametangiophores and greater growth. Developmental stage is important in determining the accumulation of UV-absorbing compounds in ***Marchantia polymorpha*** ssp. ***ruderalis***. High temperatures can change the ratios of fatty acids and cause a reversible depression of photosynthesis; there seems to be limited thermostability. They are much more cold tolerant and don't suffer frost damage. The ω-3 polyunsaturated fatty acids increase as the temperature decreases and ABA seems to play a role in cold survival.

Marchantia polymorpha produces sesquiterpenoids that are active against some fungi and bacteria. Some methylobacteria promote the growth of gemmae. Other bacteria promote thallus growth, fix nitrogen, and inhibit pathogens. Extracts of the liverwort have antibiotic properties. The

archegoniophore is particularly well protected, with a flavonoid content ten times that of the thallose gametophyte. Antibacterial properties increase with altitude. The antibacterial compounds also inhibit protozoa. Fungi can occur as endophytes and pathogens, but others might contribute to symbiosis. The subspecies ***montivagans*** develops mycorrhizal associations with some fungi, but this relationship is not known in subspecies ***polymorpha***. The liverwort protects the fungi from water loss and can have a number of species living in association with it.

We have a meager understanding of the invertebrates that depend on ***Marchantia polymorpha***. At least one fly (***Scatopsciara cunicularius***) feeds on it as larvae. Extracts of allene oxide synthase inhibits oviposition and survival rate of the spider mite ***Tetranychus urticae***. Spiders and isopods inhabit them, but herbivory needs to be studied.

Some members of the **Apiaceae** can inhibit growth of ***Marchantia polymorpha***, whereas the liverwort can inhibit seed germination in ***Bidens biternata***.

It appears that the species has evolved numerous biochemical adaptations. Anthocyanins and auronidins can screen visible light, protecting it from strong sunlight. Auronidins seem to be important in protecting the liverwort in the extremes of its environmental conditions. Flavonoids protect against UV light and herbivory. They may provide antibiotics benefitting animals in nature. Their phenolic compounds are important in UV protection and might not have the importance in antiherbivory we once thought.

Large increases in CO_2 cause a number of changes in the ***Marchantia polymorpha*** photosynthetic system, but the net result is that there is little change in photosynthetic rate. High light intensity inhibits photosynthesis. Light quality can affect growth and gemma production. The red:far red ratio affects chlorophyll concentration and senescence and is mediated by phytochromes.

The species accumulates heavy metals, but not in large quantities like mosses do, due to lower surface area. Its best role seems to be in the laboratory. It is easy to culture, so easy that in greenhouses it is necessary to find ways to discourage its growth in pots for trees and other plants. Chronic gamma radiation causes inhibition of the development of sex organs in ***Marchantia polymorpha*** and its responses can be used to monitor radiation effects. Rapid and easy growth and haploid condition make the species useful to test the function of plant genes.

Acknowledgments

Lars Söderström helped me find the current acceptable names for a number of older taxa. Thank you to Rui-Liang Zhu, Brent Mishler, and Yoan Coudert for providing me with the publications on gene sequencing in ***Marchantia polymorpha***. Nils Cronberg alerted me to important papers on dN/dS and problems with gene matching. Masaki Shimamura alerted me to nomenclature problems in ***Marchantia paleacea*** *vs.* ***M. polymorpha***. Thank you to Brent Mishler and Anders Hagborg for providing me with access to the article by Åkerman (1910). Marc Favreau, Marc-Frédéric Indorf, Jean Gagnon, Geert Raeymaekers, Michael Haeusler, Yoan Coudert, and Stephen Rae helped me interpret "lumiere rouge-fonce." Bart van Tooren took pictures for me to illustrate a wadden polder, with the help of Heinjo During in coordination.

Literature Cited

Abe, S. and Ohta, Y. 1983. Lunularic acid in cell suspension culture of *Marchantia polymorpha*. Phytochemistry 22: 1917-1920.

Abe, S. and Ohta, Y. 1984. The concentrations of lunularic acid and prelunularic acid in liverworts. Phytochemistry 23: 1379-1381.

Adam, K.-P. and Becker, H. 1993a. Phenanthrenes and other phenolics from *in vitro* cultures of *Marchantia polymorpha*. Phytochemistry 35: 139-143.

Adam, K. P. and Becker, H. 1993b. A lectin from the liverwort *Marchantia polymorpha* L. Experientia 49: 1098-1100.

Adámek, M., Hadincová, V., and Wild, J. 2016. Long-term effect of wildfires on temperate *Pinus sylvestris* forests: Vegetation dynamics and ecosystem resilience. Forest Ecol. Mgmt. 380: 285-295.

Agati, G. and Tattini, M. 2010. Multiple functional roles of flavonoids in photoprotection. New Phytol. 186: 786-793.

Åkerman, Å. 1910. Über die Chemotaxis der *Marchantia*-Spermatozoiden. Z. Bot. 2: 94-103.

Aki, S. S., Nishihama, R., Kohchi, T., and Umeda, M. 2019a. Cytokinin signaling coordinates development of diverse organs in *Marchantia polymorpha*. Plant Signal. Behav. 14(11): 1668232.

Aki, S. S., Mikami, T., Naramoto, S., Nishihama, R., Ishizaki, K., Kojima, M., Takebayashi, Y., Sakakibara, H., Kyozuka, J., and Umeda, M. 2019b. Cytokinin signaling is essential for organ formation in *Marchantia polymorpha*. Plant Cell Physiol. 60: 1842-1854.

Akter, K., Tougane, K., Kaneko, M., and Takezawa, D. 2011. Effect of sugar and abscisic acid on freezing and desiccation tolerance in the liverwort *Marchantia polymorpha*. Cryobiol. Cryotech. 57(1): 83-86.

Akter, K., Kato, M., Sato, Y., Kaneko, Y., and Takezawa, D. 2014. Abscisic acid-induced rearrangement of intracellular structures associated with freezing and desiccation stress tolerance in the liverwort *Marchantia polymorpha*. J. Plant Physiol. 171: 1334-1343.

Alam, A. and Pandey, S. 2016. *Marchantia polymorpha* L.: An emerging model plant system to study contemporary plant biology–a review. Plant Sci. Today 3(2): 88-99.

Albert, N. W., Thirmawithana, A. H., Mcghie, T. K., Clayton, W., Deroles, S. C., Schwinn, K. E., Bowma, J. L., Jordan, R. R., and Davies, K. M. 2018. Genetic analysis of the liverwort *Marchantia polymorpha* reveals that R2R3MYB activation of flavonoid production in response to abiotic stress is an ancient character in land plants. New Phytol. 218: 554-566.

Alcaraz, L. D., Peimbert, M., Barajas, H. R., Dorantes-Acosta, A. E., Bowman, J. L., and Arteaga-Vázquez, M. A. 2018. *Marchantia* liverworts as a proxy to plants' basal microbiomes. Sci. Rept. 8(1): 1-12.

Alfasane, M. A., Ullah, M. S., and Khondker, M. 2013. Limnology of Lake Rainkhyongkain of Bangladesh with a new record of *Marchantia polymorpha* L. var. *aquatica* Nees. Bangladesh J. Bot. 42: 223-229.

Alijev, D. A. and Babajev, F. A. 1976. Contribution to the flora of mosses of watery and swampy lands of smaller Caucasas (within Aserbaijan SSR). Bot. Zurn. SSSR 61: 470-472.

Althoff, F., Kopischke, S., Zobell, O., Ide, K., Ishizaki, K., Kohchi, T., and Zachgo, S. 2014. Comparison of the MpEF1α and CaMV35 promoters for application in *Marchantia polymorpha* overexpression studies. Transgen. Res. 23: 235-244.

Altland, J. E., Wehtje, G., Mckee, M. L., and Gilliam, C. H. 2008. Liverwort (*Marchantia polymorpha*) response to Quinoclamine in a pine bark substrate. Weed Sci. 56: 762-766.

Alvarez, L. 2017. The tailored sperm cell. J. Plant Res. 130: 455-464.

Andersen, O. M. and Markham, K. R. (eds.). 2006. The Flavonoids: Chemistry, Biochemistry and Applications. CRC Press, Boca Raton, USA.

Anton, H., Kraut, L., Mues, R., and Maria, I. M. 1997. Phenanthrenes and bibenzyls from a *Plagiochila* species. Phytochemistry 46: 1069-1075.

Antropova, T. A. 1974. Temperature adaptation studies on the cells of some bryophyte species. Tsitologiya 16(1): 38-42.

Ares, Á., Itouga, M., Kato, Y., and Sakakibara, H. 2018. Differential metal tolerance and accumulation patterns of Cd, Cu, Pb and Zn in the liverwort *Marchantia polymorpha* L. Bull. Environ. Contam. Toxicol. 100: 444-450.

Aro, E.-M. 1982. A comparison of the chlorophyll-protein composition and chloroplast ultrastructure in two bryophytes and two higher plants. Zeits. Pflanzenphysiol. 108: 97-105.

Aro, E.-M., Niemi, H., and Valanne, N. 1981. Photosynthetic studies on two ecologically different bryophytes. In: Akoyunoglou, G. (ed.). Photosynthesis III. Structure and Molecular Organization of The Photosynthetic Apparatus. Balaban International Science Services, Philadelphia, pp. 327-335.

Asakawa, Y. 1981. Biologically active substances obtained from bryophytes. J. Hattori Bot. Lab. 50: 123-142.

Asakawa, Y. 1982. Chemical constituents of Hepaticae. In: Herz, W., Grisebach, H., and Kirby, G. W. (eds.). Progress in the Chemistry of Organic Natural Products. vol. 42. Springer, Wien, pp. 1-285.

Asakawa, Y., Matsuda, R., and Takemoto, T. 1982. Mono- and sesquiterpenoids from *Hydrocotyle* and *Centella* species. Phytochemistry 21: 2590-2592.

Asakawa, Y., Tori, M., Takikawa, K., Krishnamurty, H. G., and Kar, S. K. 1987. Cyclic bis(bibenzyls) and related compounds from the liverworts *Marchantia polymorpha* and *Marchantia palmata*. Phytochemistry 26: 1811-1816.

Asakawa, Y., Tori, M., Masuya, T., and Frahm, J. P. 1990. Ent-sesquiterpenoids and cyclic bis(bibenzyls) from the German liverwort *Marchantia polymorpha*. Phytochemistry 29: 1577-1584.

Asakawa, Y., Ludwiczuk, A., Novakovic, M., Bukvicki, D., and Anchang, K. Y. 2021. Bis-bibenzyls, bibenzyls, and terpenoids in 33 genera of the Marchantiophyta (liverworts): Structures, synthesis, and bioactivity. J. Nat. Prod. 85: 729-762.

Aslam, Z., Lee, C. S., Kim, K.-H., Im, W.-T., Ten, L. N., and Lee, S.-T. 2007. *Methylobacterium jeotgali sp. nov*., a nonpigmented, facultatively methylotrophic bacterium isolated from jeotgal, a traditional Korean fermented seafood. Intertnat. J. Syst. Evol. Microbiol. 57: 566-571.

Bajguz, A. and Hayat, S. 2009. Effects of brassinosteroids on the plant responses to environmental stresses. Plant Physiol. Biochem. 47(1): 1-8.

Bajon, C., Blaize-Sauvanet, A., Robert, D., and Roland, F. 1995. *In situ* detection of mRNAs in the nucleus of the motile spermatozoid in a bryophyte (*Marchantia polymorpha*). Cryptog. Bryol. 16(1): 1-10.

Bajpai, V. K., Shin, S. Y., Kim, H. R., and Kang, S. C. 2008. Anti-fungal action of bioconverted eicosapentaenoic acid (bEPA) against plant pathogens. Indus. Crops Prod. 27(1): 136-141.

Balcerkiewicz, S. and Rusinska, A. 1987. Expansion of bryophytes on areas treated with herbicides. Symp. Biol. Hung. 35: 285-293.

Baltazar Pereda, L. G. and Pérez Rebaza, C. A. 2021. Evaluación de tres plantas nativas en la depuración de aguas residuales municipales mediante humedales en Santiago de Chuco, La Libertad. [Evaluation of three native plants in the purification of municipal wastewater through wetlands in Santiago de Chuco, La Libertad.]. <http://dspace.unitru.edu.pe/handle/UNITRU/17695>.

Barner, J. 1990. Uber die okologische Vorbedingungen zur Entstehung von Thalli aus Rhizoiden von *Marchantia polymorpha*: Eine regenerationsokologische Studie. [Ecological prerequisites to the development of thalli from rhizoids of *Marchantia polymorpha*: A regeneration-ecological investigation.]. Phyton (Horn) 30: 201-207.

Benkert, D. 1998. Beiträge zur Kenntnis bryophiler Pezizales-Arten. 7. *Octospora inthacaensis*. Zeits. Mykol. 64: 41-44.

Benson, M. and Blackwell, E. 1926. Observations on a lumbered area in Surrey from 1917 to 1925. J. Ecol. 14: 120-137.

Benson-Evans, K. 1961. Environmental factors and bryophytes. Nature 191: 255-260.

Benson-Evans, K. 1964. Physiology of the reproduction of bryophytes. Bryologist 67: 431-445.

Berland, H., Albert, N. W., Stavland, A., Jordheim, M., McGhie, T. M., Zhou, Y., Zhang, H., Deroles, S. C., Schwinn, K. E., Jordan, B. R., Davies, K. M., and Andersen, Ø. M. 2019. Auronidins are a previously unreported class of flavonoid pigments that challenges when anthocyanin biosynthesis evolved in plants. Proc. Natl. Acad. Sci. U. S. A. 116: 20232-20239.

Berrie, G. K. and Webster, P. M. 1982. Ultrastructure of plastids and mitochondria in gemmae of *Marchantia polymorpha* L. Ann. Bot. (London) 50: 199-206.

Bhargava, A. K. and Chauhan, T. S. 1978. Report on the presence of gametophore in *Marchantia polymorpha*. Acta Bot. Indica 6: 211-212.

Billhardt, A. 2021. Functional analyses of growth and development in the liverwort *Marchantia polymorpha*. Ph.D. dissertation, Acta Universitatis Upsaliensis, 99 pp.

Binns, A. N. and Maravolo, N. C. 1972. Apical dominance, polarity, and adventitious growth in *Marchantia polymorpha*. Amer. J. Bot. 59: 691-696.

Birch, S. P., Kelly, M. G., and Whitton, B. A. 1988. Macrophytes of the River Wear: 1966-1976, 1986. Trans. Bot. Soc. Edinb. 45: 203-212.

Bisang, I., Korpelainen, H., and Hedenäs, L. 2010. Can the sex-specific molecular marker of *Drepanocladus trifarius* uncover gender in related species?. J. Bryol. 32: 305-308.

Bischler, H. 1984. Spore morphology and spore germination in *Marchantia* L. In: Vána, J. (ed.). Proceedings of the Third Meeting of the Bryologists from Central and East Europe. Praha, Univerzita, Karlova, pp. 77-86.

Bischler, H. 1986. *Marchantia polymorpha* L. *s. lat.* karyotype analysis. J. Hattori Bot. Lab. 60: 105-117.

Bischler, H. and Jovet-Ast, S. 1981. The biological significance of morphological characters in Marchantiales (Hepaticae). Bryologist 84: 208-215.

Bischler-Causse, H. 1989. *Marchantia* L. The Asiatic and Oceanic taxa. Bryophyt. Biblio. 38: 1-317.

Bley, K. A. 1987. Moosfloristische und -oekologische Untersuchungen in Fliessgewässern des Harzes. Herzogia 7: 623-647.

Blumberg, P. M. 1991. Complexities of the protein kinase C pathway. Molec. Carcin. 4: 339-344.

Bockers, M., Capková, V., Tichá, I., and Schäfer, C. 1997. Growth at high CO_2 affects the chloroplast number but not the photosynthetic efficiency of photoautotrophic *Marchantia polymorpha* culture cells. Plant Cell Tiss. Organ Cult. 48(2): 103-110.

Boisselier-Dubayle, M. C. and Bischler, H. 1989. Electrophoretic studies in *Marchantia polymorpha* L. J. Hattori Bot. Lab. 67: 297-311.

Boisselier-Dubayle, M. C., Jubier, M. F., Lejeune, B., and Bischler, H. 1995. Genetic variability in the three subspecies of *Marchantia polymorpha* (Hepaticae): isozymes, RFLP and RAPD markers. Taxon 44: 363-376.

Boisselier-Dubayle, M.-C., Lambourdiere, J., Leclerk, M., and Bischler, H. 1997. Phylogenetic relationships in the Marchantiales (Hepaticae). Apparent incongruence between morphological and molecular data. Compt. Rend. Acad. Sci. 3 Sci. Vie 320: 1013-1020.

Bopp, M. and Vicktor, R. 1988. Protoplasts of *Marchantia polymorpha* and its development. Plant Cell Physiol. 29: 497-502.

Bowman, J. L. 2016. A brief history of *Marchantia* from Greece to genomics. Plant Cell Physiol. 57: 210-229.

Bowman, J. L., Araki, T., Arteaga-Vazuez, M A., Berger, F., Dolan, L., Haseloff, J., Ishizaki, K., Kyozuka, J., Lin, S.-S., Nagasaki, H., Nakagami, H., Nakajima, K., Nakamura, Y., Ohashi-Ito, K, Sawa, S., Shimamura, M., Solano, R., Tsukaya, H., Ueda, T., Watanabe, Y., Yamato, K. T., Zachgo, S., and Kohchi, T. 2016. The naming of names: Guidelines for gene nomenclature in *Marchantia*. Plant Cell Physiol. 57: 257-261.

Bowman, J. L., Kohchi, T., Yamato, K. T., Jenkins, J., Shu, S., Ishizaki, K., Yamaoka, S., Nishihama, R., Nakamura, Y., Berger, F., Adam, C., Sugamata Aki, S., Althoff, F., Araki, T., Artega-Vazquez, M. A., Balasubrmanian, S., Barry, K., Bauer, D., ... and Schmutz, J. 2017. Insights into land plant evolution garnered from the *Marchantia polymorpha* genome. Cell 171: 287-304.

Bradbury, S. M. 2006. Response of the post-fire bryophyte community to salvage logging in boreal mixedwood forests of northeastern Alberta, Canada. Forest Ecol. Mgmt. 234: 313-322.

Briggs, D. 1972. Population differentiation in *Marchantia polymorpha* L. in various lead pollution levels. Nature 238: 166-167.

Brockington, S., Glover, B., Duckett, J. G., and Pressel, S. 2013. The cuticle in *Marchantia*: An overlooked innovation in land plants. Conference of the International Association of Bryologists, 15-19 July 2013 at Natural History Museum, London, UK.

Brodie, H. J. 1951. The splash-cup dispersal mechanism in plants. Can. J. Bot. 29: 224-230.

Bulanda, W. 1980. Calculation methods for resistance and capacity of *Marchantia polymorpha* thallus. Folia Soc. Sci. Lublin. Biol. 2. 22: 63-70.

Burgeff, H. E. N. 1943. Genetische studien an *Marchantia*. G. Fischer.

Cahuana, L. and Aduvire, O. 2019. Bioacumulación de metales pesados en tejidos de vegetación acuática y terrestre evaluados en áreas donde existen pasivos ambientales mineros en el Perú. [Bioaccumulation of heavy metals in aquatic and terrestrial vegetation tissues evaluated in areas where there are mining environmental liabilities in Peru.]. Rev. Medio Ambien. Mineria 4(2): 19-36.

Callaghan, D. 2009. Bryophytes of domestic gardens in Britain. Field Bryol. 98: 23-27.

Campbell, E. O. 1968. *Marchantia polymorpha* var. *aquatica* in New Zealand. Tuatara 16: 179-184.

Cao, J. G., Dai, X. L., Zou, H. M., and Wang, Q. X. 2014. Formation and development of rhizoids of the liverwort *Marchantia polymorpha*. J. Torrey Bot. Soc. 126-134.

Carella, P., Gogleva, A., Hoey, D. J., Bridgen, A. J., Stolze, S. C., Nakagami, H., and Schornack, S. 2019. Conserved biochemical defenses underpin host responses to oomycete infection in an early-divergent land plant lineage. Curr. Biol. 29: 2282-2294.

Caron, J. E. A. 1972. Mosbestrijding in de Boomkwekerij. [Fighting moss in arboriculture.]. Bedrijfsontwikkeling 3: 621-623.

Carothers, Z. B. and Kreitner, G. L. 1968. Studies of spermatogenesis in the Hepaticae: II. Blepharoplast structure in the spermatid of *Marchantia*. J. Cell Biol. 36: 603-616.

Carter, J. L. and Nickell, G. L. 1967. The effects of various wavelengths of visible light on thallus growth and gemmae cup production in *Marchantia polymorpha* L. (Marchantiaceae). Southwest. Nat. 12: 113-118.

Carter, J. L. and Romine, K. G. 1969. The effects of long and short photoperiods on the rate of growth and gemmae cup production in *Marchantia polymorpha* L. Trans. Kans. Acad. Sci. 72(1): 98-107.

Cheynier, V., Comte, G., Davies, K. M., Lattanzio, V., and Martens, S. 2013. Plant phenolics: Recent advances on their biosynthesis, genetics, and ecophysiology. Plant Physiol. Biochem. 72: 1-20.

Chiapusio, G., Pujol, S., Toussaint, M. L., Badot, P. M., and Binet, P. 2007. Phenanthrene toxicity and dissipation in rhizosphere of grassland plants (*Lolium perenne* L. and *Trifolium pratense* L.) in three spiked soils. Plant Soil 294: 103-112.

Chiou, S. Y., Su, W. W., and Su, Y. C. 2001. Optimizing production of polyunsaturated fatty acids in *Marchantia polymorpha* cell suspension culture. J. Biotechnol. 85: 247-257.

Chiyoda, S., Ishizaki, K., Kataoka, H., Yamato, K. T., and Kohchi, T. 2008. Direct transformation of the liverwort *Marchantia polymorpha* L. by particle bombardment using immature thalli developing from spores. Plant Cell Rept. 27: 1467-1473.

Chung, S. J., Takechi, K., Sakai, A., Ono, K., and Takano, H. 2006a. Profiling of bryophyte gene expression by hybridization of an *Arabidopsis* cDNA array with bryophyte cDNA. J. Hattori Bot. Lab. 99: 233-244.

Chung, S. J., Takechi, K., Sakai, A., Ono, K., and Takano, H. 2006b. Detection of genes differentially expressed in cultured cells of *Marchantia polymorpha*, but not in

Arabidopsis thaliana, using an *Arabidopsis* cDNA macroarray. J. Hattori Bot. Lab. 99: 245-257.

Churchill, S. and Linares, E. 1995. Prodromus Bryoloqiae Novo-Granatensis. Introducción a la flora de musgos de Colombia. Parte 1 y 2. Editora. Guadalupe Ltda. Bogotá.

Clausen, E. 1952. Hepatics and humidity: A study on the occurrence of hepatics in a Danish tract and the influence of relative humidity on their distribution. Dansk Bot. Ark. 15(1): 1-80.

Clayton, W. 2017. The effect of UV-B radiation on flavonoid accumulation and gene expression in *Marchantia polymorpha*. Ph.D. dissertation, Lincoln University.

Clayton, W. A., Albert, N. W., Thrimawithana, A. H., Mcghie, T. K., Deroles, S. C., Schwinn, K. E., Warren, B. A., McLachlan, A. R. G., Bowman, J. L., Jordan, B. R., and Davies, K. M. 2018. UVR8-mediated induction of flavonoid biosynthesis for UVB tolerance is conserved between the liverwort *Marchantia polymorpha* and flowering plants. Plant J. 96: 503-517.

Close, D. C. and McArthur, C. 2002. Rethinking the role of many plant phenolics – protection from photodamage, not herbivores? Oikos 99: 166-172.

Coombes, A. J. and Lepp, N. W. 1974. The effect of Cu and Zn on the growth of *Marchantia polymorpha* and *Funaria hygrometrica*. Bryologist 77: 447-452.

Corgié, S. C., Joner, E. J., and Leyval, C. 2003. Rhizospheric degradation of phenanthrene is a function of proximity to roots. Plant Soil 257: 143-150.

Corner, E. J. H. 1964. The Life of Plants. University of Chicago Press, Chicago, IL, USA, 376 pp.

Courtoy, R. 1965-1966. Contribution a l'etude du role de la lumiere dans la sexualisation du gametophyte de *Marchantia polymorpha* L. Arch. Inst. Bot. Stat. Ecol. Veget. Hautes-Fagnes Lab. Phytotron 32: 441-447.

Cuitun-Coronado, D., Rees, H., Colmer, J., Hall, A., Dantas, L. L. D. B., and Dodd, A. N. 2022. Circadian and diel regulation of photosynthesis in the bryophyte *Marchantia polymorpha*. Plant Cell Environ. 2022: 1-14.

Cutting, E. M. 1910. On androgynous receptacles in *Marchantia*. Ann. Bot. 24: 349-357.

Darigo, C. E. 2004. *Aneura maxima* (Hepaticae: Aneuraceae) and *Marchantia aquatica* Hepaticae: Marchantiaceae) New to Missouri and interior highlands of North America. Evansia 21: 76-78.

Davidonis, G. H. and Munroe, M. H. 1972. Apical dominance in *Marchantia*: Correlative inhibition of neighbor lobe growth. Bot. Gaz. 133: 177-184.

Davies, K., Albert, N., Zhou, Y., and Schwinn, K. 2018. Functions of flavonoid and betalain pigments in abiotic stress tolerance in plants. Ann. Plant Rev. Online 1: 1-41.

Davies, K. M., Jibran, R., Zhou, Y., Albert, N. W., Brummell, D. A., Jordan, B. R., Bowman, J L, and Schwinn, K. E. 2020. The evolution of flavonoid biosynthesis: A bryophyte perspective. Front. Plant Sci. 1: 7.

De, A., Mukherjee, S., and Dey, A. 2015. Altitudinal variation of anti-human-pathogenic-bacterial activity and antioxidative properties of Darjeeling Himalayan *Marchantia polymorpha* L. J. Biol. Active Prods. Nat. 5(1): 33-42.

Döbbeler, P. 1979. Untersuchungen an Moosparasitischen Pezizales aus der Verwandtschaft von Octospora. Nova Hedw. 31: 817-864.

Döbbeler, P. 2002. Microniches occupied by bryophilous Ascomycetes. Nova Hedw. 75: 275-306.

Dörken, V. M. 2012. *Marchantia polymorpha*: Brunnenlebermoos (Marchantiaceae). Jahrb. Bochumer Bot. Ver. 3: 236-245.

Douin, R. 1936. Sur le photogeotropisme des thalles pedonculaires et capitulaires des Marchantiacees. Compt. Rend. Acad. Sci. 201: 154-156.

Downie, S. R., Olmstead, R. G., Zurawski, G., and Soltis, D. E. 1991. Six independent losses of chloroplast DNA rp/2 intron in dicotyledons: Molecular and phylogenic implications. Evolution 45: 1245-1259.

Duarte, S. 2020. The functions of ethylene-signaling in the regulation of gemma dormancy and germination in the liverwort *Marchantia polymorpha*. M.S. thesis, Uppsala University, Uppsala, Sweden, 37 pp.

Duckett, J. G. and Ligrone, R. 2003. What we couldn't have done if we'd stayed in Europe: Selection and serendipity in the Southern Hemisphere! Bull. Brit. Bryol. Soc. 80: 19-21.

Duckett, J. G. and Pressel, S. 2009. Extraordinary features of the reproductive biology of *Marchantia* at Thursley NNR. Field Bryol. 97: 2-11.

Duckett, J., Matcham, H., and Pressel, S. 2008. Thursley common NNR: Bryophyte recolonization one year after the great fire of July 2006. Field Bryol. 94: 3-11.

Dunham, V. L. and Bryan, J. K. 1968. Effects of exogenous amino acids on the development of *Marchantia polymorpha* gemmalings. Amer. J. Bot. 55: 745-752.

Dupuy, J., Leglize, P., Vincent, Q., Zelko, I., Mustin, C., Ouvrard, S., and Sterckeman, T. 2016. Effect and localization of phenanthrene in maize roots. Chemosphere 149: 130-136.

Durand, E. J. 1908. The development of the sexual organs and sporogonium of *Marchantia polymorpha*. Bull. Torrey Bot. Club 35: 321-335.

During, H. J. 2001. Invited essay – new frontiers in bryology and lichenology. Diaspore banks. Bryologist 104: 92-97.

Eklund, D. M., Ishizaki, K., Flores-Sandoval, E., Kikuchi, S., Takebayashi, Y., Tsukamoto, S., Hirakawa, Y., Nonomura, M., Kato, H., Kouno, M., Bhalerao, R., Lagercrantz, U., Kasahara, H., Kohchi, T., and Bowman, J. L. 2015. Auxin produced by the indole-3-pyruvic acid pathway regulates development and gemmae dormancy in the liverwort *Marchantia polymorpha*. Plant Cell 27: 1650-1669.

Eklund, D. M., Kanei, M., Flores-Sandoval, E., Ishizaki, K., Nishihama, R., Kohchi, T., Lagercrantz, U., Bhalerao, R. P., Sakata, Y., and Bowman, J. L. 2018. An evolutionarily conserved abscisic acid signaling pathway regulates dormancy in the liverwort *Marchantia polymorpha*. Curr. Biol. 28: 3691-3699.

England, J. and Jeger, M. 2005. The effect of nozzle size, water pressure and nozzle height on dispersal of *Marchantia polymorpha* gemmae, using an overhead sprinkler system. In: Proceedings of the 13th EWRS Symposium, Bari, Italy, 19-23 June 2005. European Weed Research Society.

Equihua Z., C. A. 1987. Diseminación de yemas en *Marchantia polymorpha* L. (Hepaticae). Cryptog. Bryol. Lichénol. 8: 199-217.

Fang, L., Guo, H. F., and Lou, H. X. 2007. Three new bibenzyl derivatives from the Chinese liverwort *Marchantia polymorpha* L. Helv. Chim. Acta 90: 748-752.

Fang, Y., Zhu, R.-L., and Mishler, B. D. 2014. Evolution of oleosin in land plants. PLoS ONE 9(8): e103806. doi: 10.1371/journal.pone.0103806.

Fausey, J. C. 2003. Controlling liverwort and moss now and in the future. HortTechnol. 13(1): 35-38.

Ferreira, M. T., Aguiar, F. C. F., Rodríguez-González, P., Albuquerque, A., and Sérgio, C. 2008. Manual Para a

Avaliação Biológica da Qualidade da Água Em Sistemas Fluviais Segundo a Directiva Quadro da Água. Protocolo de amostragem e análise para os macrófitos. [Manual for Biological Assessment of Water Quality in River Systems under the Water Framework Directive. Sampling and Analysis Protocol for Macrophytes.]. Ministério Do Ambiente, Do Ordenamento Do Território e Do Desenvolvimento Regional, 18 pp.

Fischer, S., Böttcher, U., Reuber, S., Anhalt, S., and Weissenböck, G. 1995. Chalcone synthase in the liverwort *Marchantia polymorpha*. Phytochemistry 39: 1007-1012.

Fletcher, M. 1982. Frost damage to bryophytes in cultivation. Bryol. Times 15: 3.

Flores-Sandoval, E., Eklund, D. M., and Bowman, J. L. 2015. A simple auxin transcriptional response system regulates multiple morphogenetic processes in the liverwort *Marchantia polymorpha*. PLoS Genetics 11(5): e1005207.

Fraiture, A. and Ertz, D. 2007. *Didymosphaeria marchantiae*, un champignon parasite d'hépatiques, nouveau pour la flore Belge. Nat. Mosana 60(1): 21-29.

Fredericq, H. 1964. Influence formatrice de la lumiere rouge-fonce sur le developpement des thalles de *Marchantia polymorpha* L. [Formative influence of deep-red light on the development of thalli of *Marchantia polymorpha* L.] Bull. Soc. Roy. Bot. Belg. 98(1): 67-76.

Fredericq, H. and Greef, J. de. 1966. Red (R), far-red (FR) photoreversible control of growth and chlorophyll content in light-grown thalli of *Marchantia polymorpha* L. Naturwissenschaften 53: 337-338.

Fredericq, H. and De Greef, J. 1968. Photomorphogenic and chlorophyll studies in the bryophyte *Marchantia polymorpha*. 1. Effect of red, far-red irradiations in short and long-term experiments. Physiol. Plant. 21: 346-359.

Fredericq, H., Veroustraete, F., Greef, J. D., and Rethy, R. 1977. Light-enhanced ethylene production in *Marchantia polymorpha* L. Arch. Internat. Physiol. Biochim. 85: 977-978.

Friederich, S., Rueffer, M., Asakawa, Y., and Zenk, M. H. 1999. Cytochromes P-450 catalyze the formation of marchantins A and C in *Marchantia polymorpha*. Phytochemistry 52: 1195-1202.

Fries, K. 1964. Über ienen genuinen Keimungs- und Streckungswachstumsaktiven Hemmstoff bei *Marchantia polymorpha* L. [On a genuine germination and elongation growth inhibitor in *Marchantia polymorpha* L.]. Beitr. Biol. Pflanzen 40: 177-235.

Froment, A. 1975. Les premiers stades de la succession vegetale apres incendie de tourbe dans la reserve naturelle des hautes Fagnes. Vegetatio 29: 209-214.

Frye, T. C. 1928. Observations on the age of a few bryophytes. Bryologist 31: 25-27.

Fujisawa, M., Hayashi, K., Nishio, T., Bando, T., Okada, S., Yamato, K. T., Fukuzawa, H., and Ohyama, K. 2001. Isolation of X and Y chromosome-specific DNA markers from a liverwort, *Marchantia polymorpha*, by representational difference analysis. Genetics 159: 981-985.

Fujita, M., Fujita, Y., Noutoshi, Y., Takahashi, F., Narusaka, Y., Yamaguchi-Shinozaki, K., and Shinozaki, K. 2006. Crosstalk between abiotic and biotic stress responses: A current view from the points of convergence in the stress signaling networks. Curr. Opinion Plant Biol. 9: 436-442.

Furuichi, T. and Matsuura, K. 2016. Kinetic analysis on the motility of liverwort sperms using a microscopic computer-assisted sperm analyzing system. Environ. Control Biol. 54(1): 45-49.

Gaal, D. J., Dufresne, S. J., and Maravolo, N. C. 1982. Transport of ^{14}C-indoleacetic acid in the hepatic *Marchantia polymorpha*. Bryologist 85: 410-418.

Gahtori, D. and Chaturvedi, P. 2011. Antifungal and antibacterial potential of methanol and chloroform extracts of *Marchantia polymorpha* L. Arch. Phytopath. Plant Protect. 44: 726-731.

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.

Gecheva, G., Yurukova, L., Cheshmedjiev, S., and Ganeva, A. 2010. Distribution and bioindication role of aquatic bryophytes in Bulgarian rivers. Biotech. Biotech. Equip. 24(supp1.): 164-170.

Gecheva, G., Yurukova, L., and Cheshmedjiev, S. 2013. Patterns of aquatic macrophyte species composition and distribution in Bulgarian rivers. Turkish J. Bot. 37: 99-110.

Geissler, P. 1976. Zur Vegetation alpiner Fliessgewässer. Beitr. Kryptogamenfl. Schweiz 14(2): 1-52

Geissler, P. and Selldorf, P. 1986. Vegetationskartierung und Transektenanalyse im subalpinen Moor von Cadagno di fuori (Val Piora, Ticino). Saussurea 1986: 35-70.

Gekeler, W., Grill, E., Winnacker, E.-L., and Zenk, M. H. 1989. Survey of the plant kingdom for the ability to bind heavy metals through phytochelatins. Zeits. Naturforsch. C Biosci. 44: 361-369.

Gellerman, J. L., Anderson, W. H., and Schlenk, H. 1972. Highly unsaturated lipids of *Mnium*, *Polytrichum*, *Marchantia*, and *Matteuccia*. Bryologist 75: 550-557.

Gemmrich, A. R. 1976. Keimungsinduktion durch Salzionen, Licht und Gibberellinsäurebei Sporen von *Marchantia polymorpha* L. [Induction of germination by salt ions, light and gibberellic acid in spores of *Marchantia polymorpha* L.]. Flora 165: 479-480.

Ghosh, T. K., Kaneko, M., Akter, K., Murai, S., Komatsu, K., Ishizaki, K., Yamato, K. T., Kohchi, T., and Takezawa, D. 2016. Abscisic acid-induced gene expression in the liverwort *Marchantia polymorpha* is mediated by evolutionarily conserved promoter elements. Physiol. Plant. 156: 407-420.

Ghosh, T. K., Tompa, N. H., Rahman, M. M., Mohi-Ud-Din, M., Al-Meraj, S. Z., Biswas, M. S., and Mostofa, M. G. 2021. Acclimation of liverwort *Marchantia polymorpha* to physiological drought reveals important roles of antioxidant enzymes, proline and abscisic acid in land plant adaptation to osmotic stress. PeerJ 9: e12419.

Giles, K. L. 1971. Dedifferentiation and regeneration in bryophytes: A selective review. N. Z. J. Bot. 9: 689-694.

Ginting, N. and Batubara, M. S. 2019. The diversity of liverworts in the Dolok Sordang Sub-District of Sipirok, South Tapanuli, Indonesia. Budapest Internat. Res. Exact Sci. 1(2): 55-64.

Glime, J. M. and Vitt, D. H. 1987. A comparison of bryophyte species diversity and niche structure of montane streams and stream banks. Can. J. Bot. 65: 1824-1837.

Godinez-Vidal, D., López-Leal, G., Covarrubias, A. A., and Reyes, J. L. 2020. Early events leading to water deficit responses in the liverwort *Marchantia polymorpha*. Environ. Exper. Bot. 178: 104172, 13 pp.

Gorham, J. 1977. Recent research on lunularic acid. British Bryological Society, AGM & Symposium Meeting 1977, Leicester, 1-2 October 1977. Accessed on 23 April 2006 at <http://rbg-web2.rbge.org.uk/bbs/meetings/mtgs77.htm>.

Gorska-Brylass, A. 1970. Hydrolytic enzymes in spermatogenesis in *Marchantia polymorpha*. Acta Soc. Bot. Poloniae 39: 185-189.

Gradstein, R. 2006. Bryophyte Garden inaugurated in Cibodas Botanical Garden, Java, Indonesia. Bryol. Times 120: 11.

Graff, P. W. 1936. Invasion by *Marchantia polymorpha* following forest fires. Bull. Torrey Bot. Club 63: 67-74.

Greef, J. de and Fredericq, H. 1969. Photomorphogenic and chlorophyll studies in the bryophyte *Marchantia polymorpha*. 2. Photobiological responses to terminal irradiations with different red/far-red ratios. Physiol. Plant. 22: 462-468.

Greef, J. A. de and Fredericq, H. 1972. Enhancement of senescence by far-red light. Planta 104: 272-274.

Greef, J. de, Veroustraete, F., Fredericq, H., and Wiemeersch, L. van. 1979. Study on the interaction of light and limiting physiological factors on the ethylene production by green *Marchantia polymorpha* thalli. In: De Greef, J. (ed.). Photoreceptors and Plant Development. Antwerpen University Press, Antwerpen, pp. 423-429.

Guminska, B. and Mierzenska, M. 1992. *Gerronema marchantiae* Sing. et Clem. – a fungus associating with *Marchantia polymorpha* L. and *Nostoc* sp. Zeszyty Naukowe Uniwersytetu Jagiellońskiego. Prace Bot. 24: 171-177.

Haberlandt, G. 1889. Ueber das Längenwachstum und den Geotropismus der Rhizoiden von *Marchantia* and *Lunularia*. Öster. Bot. Zeit. 39: 93-98.

Halbsguth, W. 1953. Über die Entwicklung der Dorsiventralität bei *Marchantia polymorpha* L. Biol. Zentralbl. 72(1-2): 52-104.

Hallier, U. 1966. Die Wirkung des 2,4-Einitrophenols auf die Atmung und die Induktion der Dorsiventralitat keimender Brutkorper von *Marchantia polymorpha* L. [The effect of 2,4-mononitrophenol on respiration and induction of dorsiventrality in germinating brood bodies of *Marchantia polymorpha* L.]. Biol. Zentralbl. 85(1): 47-86.

Hamashima, N., Xie, X., Hikawa, M., Suzuki, T., and Kodama, Y. 2019. A gain-of-function T-DNA insertion mutant of *Marchantia polymorpha* hyper-accumulates flavonoid riccionidin A. Plant Biotechnol. 36: 201-204.

Hanke, S. T. and Rensing, S. A. 2010. In vitro association of non-seed plant gametophytes with arbuscular mycorrhiza fungi. J Endocytobiosis Cell Res. 20: 95-101.

Hanson, D., Andrews, T. J., and Badger, M. R. 2002. Variability of the pyrenoid-based CO_2 concentrating mechanism in hornworts (Anthocerotophyta). Funct. Plant Biol. 29: 407-416.

Harashima, S. and Ono, K. 1991. Physiological characteristics and morphogenetic potential of long-term cultured cells in bryophytes. J. Hattori Bot. Lab. 69: 171-184.

Harrer, R. 2003. Associations between light-harvesting complexes and Photosystem II from *Marchantia polymorpha* L. determined by two- and three-dimensional electron microscopy. Photosyn. Res. 75(3): 249-258.

Hartmann, E. and Weber, M. 1990. Photomodulation of protonema development. In: Chopra, R. N. and Bhatla, S. C. (eds.). Bryophyte Development: Physiology and Biochemistry. CRC Press, Ann Arbor, pp. 33-54.

Hartwell, J. L. 1982. Plants used against cancer: A survey. Quarterman Publications, Lawrence, MA, 710 pp.

Hatanaka, R. and Sugawara, Y. 2010. Development of desiccation tolerance and vitrification by preculture treatment in suspension-cultured cells of the liverwort *Marchantia polymorpha*. Planta 231: 965-976.

He, X.-L., Sun, Y., and Zhu, R.-L. 2013. The oil bodies of liverworts: Unique and important organelles in land plants. Crit. Rev. Plant Sci. 32: 293-302.

Heald, F. de. 1898. Conditions for the germination of the spores of bryophytes and pteridophytes. Bot. Gaz. 26(1): 25-45.

Hedger, D. K., Taylor, J., Montague, J. J., and Schindler, B. V. 1972. The relationship between some environmental parameters and the growth of *Marchantia* gemmalings. Bryologist 75: 81-83.

Heino, J. and Virtanen, R. 2006. Relationships between distribution and abundance vary with spatial scale and ecological group in stream bryophytes. Freshwat. Biol. 51: 1879-1889.

Heras-Ibáñez, J. de las, Guerra Montes, J., and Herranz, J. M. 1991. Changes in floristic diversity and fugacity of bryophytes in burnt sites of SE Spain. Lindbergia 17: 11-16.

Himanshu, V., Dubey, R. C., and Pandey, N. 2007. Antimicrobial activity of three bryophytes against human pathogens. Curr. Trends Bryol. 2007: 47-59.

Hipol, R. M. and Broñola-Hipol, R. L. 2016. Carboxylesterase activity of fungi isolated from *Marchantia polymorpha*. World 5(2): 6-8.

Hirano, S., Sasaki, K., Osaki, Y., Tahara, K., Takahashi, H., Takemiya, A., and Kodama, Y. 2022. The localization of phototropin to the plasma membrane defines a cold-sensing compartment in *Marchantia polymorpha*. PNAS Nexus 1(2): pgac030.

Hirata, T., Ashida, Y., Mori, H., Yoshinaga, D., and Goad, L. J. 2000. A 37-kDa peroxidase secreted from liverworts in response to chemical stress. Phytochemistry 55: 197-202.

Hiwatashi, T., Goh, H., Yasui, Y., Koh, L. Q., Takami, H., Kajikawa, M., Kirita, H., Kanazawa, T., Minamino, N., Togawa, T., Sato, M., Wakazaki, M., Yamaguchi, K., Shigenobu, S., Fukaki, H., Mimura, T., Toyooka, K., Sawa, S., Yamato, K. T., Ueda, T., Urano, D., Kohchi, T., and Ishizaki, K. 2019. The RopGEF KARAPPO is essential for the initiation of vegetative reproduction in *Marchantia polymorpha*. Curr. Biol. 29: 3525-3531.

Hoffman, G. R. 1966. Ecological studies of *Funaria hygrometrica* Hedw. in eastern Washington and northern Idaho. Ecol. Monogr. 36: 157-180.

Hoffman, G. R. 1974. Growth stimulation of *Marchantia polymorpha* from ilmenite basalt and volcanic ash. Bryologist 77: 632-636.

Hollensen, R. H. and Taylor, J. 1981. A gemmiparous population of *Marchantia polymorpha* var. *aquatica* in Cheboygan County, Michigan. Mich. Bot. 20: 189-191.

Holmes, N. T. H. and Whitton, B. A. 1975. Submerged bryophytes and angiosperms of the River Tweed and its tributaries. Trans. Bot. Soc. Edinburgh 42: 383-395.

Holmes, N. T. H. and Whitton, B. A. 1977a. Macrophytic vegetation of River Tees in 1975 – Observed and predicted changes. Freshwat. Biol. 7: 43-60.

Holmes, N. T. H. and Whitton, B. A. 1977b. Macrophytic vegetation of River Swale, Yorkshire. Freshwat. Biol. 7: 545-558.

Holmes, N. T. H. and Whitton, B. A. 1981. Plants of the River Tyne system before the Kielder water scheme. Naturalist 106: 97-107.

Ikeuchi, M. and Inoue, H. 1988. A new photosystem II reaction centre component (4.8-kDa protein) encoded by chloroplast genome. Fed. Eur. Biochem. Soc. Lett. 241: 99-104.

Imoto, S. A. and Ohta, Y. 1985. Intracellular localization of lunularic acid in suspension cultured cells of *Marchantia polymorpha*. Plant Physiol. 79: 751-755.

Inoue, H. 1960. Studies in spore germination and the earlier stages of gametophyte development in the Marchantiales. J. Hattori Bot. Lab. 23: 148-191.

Inoue, H. and Asakawa, T. 1966. Making a motion picture of the life history of *Marchantia polymorpha*. Bryologist 69: 369-373.

Inoue, K., Nishihama, R., Araki, T., and Kohchi, T. 2019. Reproductive induction is a far-red high irradiance response that is mediated by phytochrome and PHYTOCHROME INTERACTING FACTOR in *Marchantia polymorpha*. Plant Cell Physiol. 60: 1136-1145.

Inouye, I. and Hori, T. 1991. High-speed video analysis of the flagellar beat and swimming patterns of algae: Possible evolutionary trends in green algae. Protoplasma 164: 54-69.

Ishida, S., Suzuki, H., Iwaki, A., Kawamura, S., Yamaoka, S., Kojima, M., Takebayashi, Y, Yamaguchi, K., Shigenobu, S., Sakakibara, H., Kohchi, T., and Nishihama, R. 2022. Diminished auxin signaling triggers cellular reprogramming by inducing a regeneration factor in the liverwort *Marchantia polymorpha*. Plant Cell Physiol. 63: 384-400.

Ishizaki, K., Shimizu-Ueda, Y., Okada, S., Yamamoto, M., Fujisawa, M., Yamato, K. T., Fujisawa, M., Yamato, K. T., Fukuzawa, H., and Ohyama, K. 2002. Multicopy genes uniquely amplified in the Y chromosome-specific repeats of the liverwort *Marchantia polymorpha*. Nucleic Acids Res. 30: 4675-4681.

Ishizaki, K., Chiyoda, S., Yamato, K. T., and Kohchi, T. 2008. *Agrobacterium*-mediated transformation of the haploid liverwort *Marchantia polymorpha* L., an emerging model for plant biology. Plant Cell Physiol. 49: 1084-1091.

Ishizaki, K., Nonomura, M., Kato, H., Yamato, K. T., and Kohchi, T. 2012. Visualization of auxin-mediated transcriptional activation using a common auxin-responsive reporter system in the liverwort *Marchantia polymorpha*. J. Plant. Res. 125: 643-651.

Ivković, I., Bukvički, D., Novaković, M., Ivanović, S., Stanojević, O., Nikolić, I., and Veljić, M. 2021. Antibacterial properties of thalloid liverworts *Marchantia polymorpha* L., *Conocephalum conicum* (L.) Dum. and *Pellia endiviifolia* (Dicks.) Dumort. J. Serb. Chem. Soc. 12: 1249-1258.

Iwakawa, H., Melkonian, K., Schlüter, T., Jeon, H. W., Nishihama, R., Motose, H., and Nakagami, H. 2021. *Agrobacterium*-mediated transient transformation of *Marchantia* liverworts. Plant Cell Physiol. 62: 1718-1727.

Iwata, S., Nakayama, N., Nakagawara, S., and Ohta, Y. 1992. Response of liverwort cells to peroxidizing herbicides. Zeits. Naturforsch. C Biochem. Biophysik Biol. Virol. 47: 394-399.

Izumi, S., Yamamoto, Y., and Hirata, T. 1995. Secretion of an esterase from the cultured suspension cells of *Marchantia polymorpha*. Phytochemistry 38: 831-833.

Jensen, S., Omarsdottir, S., Bwalya, A. G., Nielsen, M. A., Tasdemir, D., and Olafsdottir, E. S. 2012. Marchantin A, a macrocyclic bisbibenzyl ether, isolated from the liverwort *Marchantia polymorpha*, inhibits protozoal growth in vitro. Phytomedicine 19: 1191-1195.

Jin, C. H. and Pyon, J. Y. 2007. Distribution, growth and control of *Marchantia polymorpha* L. in ginseng gardens. Korean J. Weed Sci. 27: 202-208.

Joenje, W. and During, H. J. 1977. Colonisation of a desalinating wadden-polder by bryophytes. Vegetatio 35: 177-185.

Johnson, N. D. 1983. Flavonoid aglycones from *Eriodictyon californicum* resin and their implications for herbivory and UV screening. Biochem. Syst. Ecol. 11: 211-215.

Jordan, B. R., Albert, N. W., Clayton, W., Deroles, S. C., Schwinn, K. E., and Davies, K. M. 2016. Regulation of UV-induced flavonoid production in *Marchantia polymorpha*: A role in the evolution of plants for land colonisation? UV4 Plants Bull. 1: 17-19.

Kaihara, S. and Takimoto, A. 1990. Interaction between L-pipecolic acid and water extracts of various plant species in floral induction of *Lemna paucicostata*. Plant Cell Physiol. 31: 1059-1061.

Kajikawa, M., Yamato, K. T., Kohzu, Y., Nojiri, M., Sakuradani, E., Shimizu, S., Sakai, Y., Fukuzawa, H., and Ohyama, K. 2004. Isolation and characterization of Δ6-desaturase, an ELO-like enzyme and Δ5-desaturase from the liverwort *Marchantia polymorpha* and production of arachidonic and eicosapentaenoic acids in the methylotrophic yeast *Pichia pastoris*. Plant Molec. Biol. 54: 335-352.

Kajikawa, M., Matsui, K., Ochiai, M., Tanaka, Y., Kita, Y., Ishimoto, M., Kohzu, Y., Shoji, S. -I., Yamato, K. T., Ohyama, K., Fukuzawa, H., and Kohchi, T. 2008. Production of arachidonic and eicosapentaenoic acids in plants using bryophyte fatty acid delta6-desaturase, delta6-elongase, and delta5-desaturase genes. Biosci. Biotech. Biochem. 72: 435-444.

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

Katayose, A., Kanda, A., Kubo, Y., Takahashi, T., and Motose, H. 2021. Distinct functions of ethylene and ACC in the basal land plant *Marchantia polymorpha*. Plant Cell Physiol. 62: 858-871.

Kato, H., Ishizaki, K., Kouno, M., Shirakawa, M., Bowman, J. L., Nishihama, R., and Kohchi, T. 2015. Auxin-mediated transcriptional system with a minimal set of components is critical for morphogenesis through the life cycle in *Marchantia polymorpha*. PLoS Genetics 11(5): e1005084.

Katoh, K. 1983a. Kinetic photoautotrophic growth of *Marchantia polymorpha* cells in suspension culture. Physiol. Plant. (Copenhagen) 59: 242-248.

Katoh, K. 1983b. Photosynthesis and photoassimilation of glucose during photomixotrophic growth of *Marchantia polymorpha* cells in suspension culture. Physiol. Plant. (Copenhagen) 57: 67-74.

Katoh, K., Ohta, Y., Hirose, Y., and Iwamura, T. 1979. Photoautotrophic growth of *Marchantia polymorpha* L. cells in suspension culture. Planta 144: 509-510.

Katoh, K., Ishikawa, M., Miyake, K., Ohta, Y., Hirose, Y., and Imamura, T. 1980. Nutrient utilization and requirement under photoheterotrophic growth of *Marchantia polymorpha*: Improvement of the culture medium. Physiol. Plant. 49: 241-247.

Katoh, K., Hori, H., and Osawa, S. 1983. The nucleotide sequences of 5s ribosomal RNAs from four Bryophyta-species. Nucleic Acids Res. 11: 5671-5674.

Kennedy, A. D. 1996. Antarctic fellfield response to climate change: A tripartite synthesis of experimental data. Oecologia 107: 141-150.

Khamarea, Y., Marble, S. C., Altland, J. E., and Chandler, A. 2021. Growth of liverwort (*Marchantia polymorpha*) and spotted spurge (*Euphorbia maculata*) decreases with substrate stratification and strategic fertilizer placement. Third Place – Charlie Parkerson Graduate Student Research Paper Competition, 13 pp. Available at <https://sna.ipps.org/uploads/docs/5bstudent3yuvrajkhamare2021.pdf>

Khetwal, K. S. 1985. Flavonoid-glucuronides of *Marchantia polymorpha* L. Herba Hung. 24: 23-26.

Kihara, H., Tanaka, M., Yamato, K. T., Horibata, A., Yamada, A., Kita, S., Ishizaki, K., Kajikawa, M., Fukizawa, H., Kohchi, T., Akakbe, Y., and Matsui, K. 2014. Arachidonic acid-dependent carbon-eight volatile synthesis from wounded liverwort (*Marchantia polymorpha*). Phytochemistry 107: 42-49.

Kijak, H., Kisiel, K., Miwa, H., Cieślewicz, A., and Odrzykoski, I. J. 2013. What has been really sequenced? Organellar genome sequences of *Marchantia polymorpha* from GeneBank belong to *M. paleacea* ssp. *diptera* not to *M. polymorpha sensu lato*. Abstract, conference paper, International Association of Bryologists, London. <https://www.researchgate.net/publication/282567855_What_has_been_really_sequenced_Organellar_genome_sequences_of_Marchantia_polymorpha_from_GeneBank_belong_to_M_paleacea_subsp_diptera_not_to_M_polymorpha_sensu_lato#fullTextFileContent>.

Kijak, H., Rurek, M., Nowak, W., Dabert, M., Odrzykoski, I. J., Berger, F., and Dolan, L. 2016. Resequencing the 'classic' *Marchantia polymorpha* chloroplast genome. Conference poster. <https://www.researchgate.net/profile/Hania-Kijak/publication/308699734_Resequencing_the_'classic'_Marchantia_polymorpha_chloroplast_genome/links/57ebf43d08ae93b7fa9578b5/Resequencing-the-classic-Marchantia-polymorpha-chloroplast-genome.pdf>.

Kilian, J., Whitehead, D., Horak, J., Wanke, D., Weinl, S., Batistic, O., D'Angelo, C., Bornberg-Bauer, E., Kudla, J., and Harter, K. 2007. The AtGenExpress global stress expression data set: Protocols, evaluation and model data analysis of UV-B light, drought and cold stress responses. Plant J. 50: 347-363.

Kisiel, K., Miwa, H., and Odrzykoski, I. J. 2011. Taxonomic identification of chloroplast genome of *Marchantia polymorpha* using DNA barcode sequences. In: Fourth International Barcode of Life Conference, p. A43.

Kitagawa, N. 1985. Observations on *Marchantia polymorpha* – on some problems concerning fertilization. Proc. Bryol. Soc. Japan 4: 15-20.

Kitagawa, N. 1987. *Marchantia polymorpha* var. *aquatica*. Proc. Bryol. Soc. Japan 4: 133-134.

Ko, K.-W., Han, K.-S., Nam, S.-J., Kwak, S.-S., Shim J.-K., and Kim, S.-K. 1995. Identification of 4-demethylsterols from suspension cultured cells of *Marchantia polymorpha* L. J. Plant Biol. 38: 219-225.

Kodama, M., Shiobara, Y., Sumitomo, H., Matsumura, K., Tsukamoto, M., and Harada, C. 1988. Total syntheses of marchantin A and riccardin B, cytotoxic bis(bibenzyls) from liverworts. J. Org. Chem. 53: 72-77.

Koeduka, T., Takaishi, M., Suzuki, M., Nishihama, R., Kohchi, T., Uefune, M., and Matsui, K. 2022. CRISPR/Cas9-mediated disruption of ALLENE OXIDE SYNTHASE results in defective 12-oxo-phytodienoic acid accumulation and reduced defense against spider mite (*Tetranychus urticae*) in liverwort (*Marchantia polymorpha*). Plant Biotechnol. 22-0328.

Kohchi, T., Shirai, H., Fukuzawa, H., Sano, T., Komano, T., Umesono, K., Inokuchi, H., Ozaki, H., and Ohyama, K. 1988. Structure and organization of *Marchantia polymorpha* chloroplast genome, IV. Inverted, repeat and small single copy regions. J. Molec. Biol. 203: 353-372.

Komatsu, A., Nishihama, R., and Kohchi, T. 2019. Observation of phototropic responses in the liverwort *Marchantia polymorpha*. In: Yamamoto, K. T. (ed.). Phototropism. Methods in Molecular Biology, vol 1924. Humana Press, New York, NY, pp. 53-61.

Kondou, Y., Miyagi, Y., Morito, T., Fujihira, K., Miyauchi, W., Moriyama, A., Terasawa, T., Ishida, S., Iwabuchi, K, Kubo, H., Nishihama, R., Ishizaki, K., and Kohchi, T. 2019. Physiological function of photoreceptor UVR8 in UV-B tolerance in the liverwort *Marchantia polymorpha*. Planta 249: 1349-1364.

Konno, H., Yamasaki, Y., and Katoh, K. 1987. Fractionation and partial characterization of pectic polysaccharides in cell walls from liverwort (*Marchantia polymorpha*) cell cultures. J. Exper. Bot. 38: 711-722.

Kovács, A., Vasas, A., and Hohmann, J. 2008. Natural phenanthrenes and their biological activity. Phytochemistry 69: 1084-1110.

Krupinska, I. 1976. Influence of lead tetraethyl on the growth of *Funaria hygrometrica* L. and *Marchantia polymorpha* L. Acta Soc. Bot. Poloniae 45: 421-428.

Kubo, H., Nozawa, S., Hiwatashi, T., Kondou, Y., Nakabayashi, R., Mori, T., Saito, K., Takanashi, K., Kohchi, T., and Ishizaki, K. 2018. Biosynthesis of riccionidins and marchantins is regulated by R2R3-MYB transcription factors in *Marchantia polymorpha*. J. Plant Res. 131: 849-864.

Kubota, A., Ishizaki, K., Hosaka, M., and Kohchi, T. 2013. Efficient *Agrobacterium*-mediated transformation of the liverwort *Marchantia polymorpha* using regenerating thalli. Biosci. Biotechnol. Biochem. 77(1): 167-172.

Kumar, K., Nath, V., and Asthana, A. K. 2007. Concept of bryophytes in classical text of Indian ethnobotanical prospective. In: Nath, V. and Asthana, A. K. (eds.). Current Trends in Bryology. Bishen Singh Mahendra Pal Singh. Dehra Dun, India, pp. 215-220.

Kumar, S., Kempinski, C., Zhuang, X., Norris, A., Mafu, S., Zi, J., Bell, S. A, Nybo, S. E., Kinison, S. E., Jiang, Z., Goklany, S., Linscott, K. B., Chen, X., Jia, Q., Brown, S. D., Bowman, J. L., Babbitt, P. L., Peters, R. J., Chen, F., and Chappell, J. 2016. Molecular diversity of terpene synthases in the liverwort *Marchantia polymorpha*. Plant Cell 28: 2632-2650.

Kutschera, U. and Koopmann, V. 2005. Growth in liverworts of the Marchantiales is promoted by epiphytic methylobacteria. Naturwissenschaften 92: 347-349.

LaBelle, J. L., Dutton, K. G., and Maravolo, N. C. 1997. The influence of ethylene and spermine on phosphatase activity during senescence in *Marchantia polymorpha*. Internat. J. Plant Sci. 158: 552-555.

Lagercrantz, U., Billhardt, A., Rousku, S. N., Ljung, K., and Eklund, D. M. 2020. Nyctinastic thallus movement in the liverwort *Marchantia polymorpha* is regulated by a circadian clock. Sci. Repts. 10(1): 1-9.

Lagercrantz, U., Billhardt, A., Rousku, S. N., Leso, M., Reza, S. H., and Eklun, D. M. 2021. DE-ETIOLATED1 has a role in the circadian clock of the liverwort *Marchantia polymorpha*. New Phytol. 232: 595-609.

Lagos-López, M. I., Sáenz-Jimenez, F. A., and Morales-Puentes, M. E. 2008. Briófitos reófilos de Tres Quebradas del Páramo de Mamapacha, Chinavita (Boyacá-Colombia). [Rheophilic bryophytes of Three River of the Páramo de Mamapacha, Chinavita (Boyacá-Colombia).]. Acta Biol. Colomb. 13(1): 143-160.

Landi, M., Tattini, M., and Gould, K. S. 2015. Multiple functional roles of anthocyanins in plant-environment interactions. Environ. Exper. Bot. 119: 4-17.

Laplaud, V., Josserand, C., Duprat, C., and Boudaoud, A. 2022. Splash-cups and rain dispersal of *Marchantia polymorpha* gemmae. APS March Meeting 2022 poster. Bull. Amer. Phys. Soc. 67(3).

Leclercq, L. 1977. Vegetation et caracteristiques physicochimiques de deux rivieres de Haute Ardenne (Belgique): La Helle et la Roer superieure. [Vegetation and

physicochemical characteristics of two rivers in the Haute Ardenne (Belgium): La Helle and la Roer superieure.]. Lejeunia 88: 1-42.

Lee, D. W. and Gould, K. S. 2002. Anthocyanins in leaves and other vegetative organs: An introduction. Adv. Bot. Res. 37: 1-16.

Lepp, N. W. and Hockenhull, Y. 1983. Growth responses of *Marchantia polymorpha* gemmalings in relation to concentration and chemical form of applied nickel. Bryologist 86: 342-346.

Lepp., N. W. and Roberts, M. J. 1977. Some effects of cadmium on growth of bryophytes. Bryologist 80: 533-536.

Li, D., Flores-Sandoval, E., Ahtesham, U., Coleman, A., Clay, J. M., Bowman, J. L., and Chang, C. 2020. Ethylene-independent functions of the ethylene precursor ACC in *Marchantia polymorpha*. Nat. Plants 6: 1335-1344.

Li, W.-A. 1990. In vitro propagation, dedifferentiation and redifferentiation of *Marchantia polymorpha* L. Acta Bot. Sinica 32: 852-856.

Li, X., Wurtele, E. S., and LaMotte, C. E. 1994. Abscisic acid is present in liverworts. Phytochemistry 37: 625-627.

Lidforss, B. 1904. Über die Reizbewegungen der *Marchantia*-Spermatozoiden. Jahrb. Wiss. Bot. 41: 65-87.

Ligrone, R., Carafa, A., Lumini, E., Bianciotto, V., Bonfante, P., and Duckett, J. G. 2007. Glomeromycotean associations in liverworts: a molecular, cellular, and taxonomic analysis. Amer. J. Bot. 94: 1756-1777.

Lin, P. C., Lu, C. W., Shen, B. N., Lee, G. Z., Bowman, J. L., Arteaga-Vazquez, M. A., Liu, L. Y., Hong, S. F., Lo, C. F., Su, G. M., Kohchi, T., Ishizaki, K., Zachgo, S., Althoff, F, Takenaka, M., Yamato, K. T., and Lin, S.-S. 2016. Identification of miRNAs and their targets in the liverwort *Marchantia polymorpha* by Integrating RNA-Seq and degradome analyses. Plant Cell Physiol. 57: 339-358.

Lin, S. S. and Bowman, J. L. 2018. Micro RNAs in *Marchantia polymorpha*. New Phytol. 220: 409-416.

Linares, E. 1986. Estudios taxonómicos y ecológicos de la brioflora en la franja alto-andina de el Tablazo, Cundinamarca [trabajo de grado]. Bogotá, Departamento deBiología. Facultad de Ciencias. Universidad Nacional de Colombia.

Linde, A.-M., Eklund, D. M., Kubota, A., Pederson, E. R. A., Holm, K., Gyllenstrand, N., Nishihama, R., Cronberg, N., Muranaka, T., Oyama, T., Kohchi, T., and Lagercrantz, U. 2017. Early evolution of the land plant circadian clock. New Phytol. 216: 576-590.

Linde, A.-M., Sawangproh, W., Cronberg, N., Szövény, P., and Lagercrantz, U. 2020. Evolutionary history of the *Marchantia polymorpha* complex. Front. Plant Sci. 11: 829.

Linde, A.-M., Eklund, D. M., Cronberg, N., Bowman, J. L., and Lagercrantz, U. 2021. Rates and patterns of molecular evolution in bryophyte genomes, with focus on complex thalloid liverworts, Marchantiopsida. Molec. Phylogen. Evol. 165: 107295.

Liu, Y., Johnson, M. G., Cox, C. J., Medina, R., Devos, N., Vanderpoorten, A., Hedenäs, L., Bell, N. E., Shevock, J. R., Aguero, B., Quandt, D., Wicket, N. J., Shaw, A. J., and Goffinet, B. 2019. Resolution of the ordinal phylogeny of mosses using targeted exons from organellar and nuclear genomes. Nat. Comm. 10: 1485. Available at <https://doi.org/10.1038/s41467-019-09454-w>.

Lloyd, C. E. and Steinmetz, F. H. 1937. Temperature as a factor influencing the sexual response of *Marchantia*. Amer. J. Bot. 24: 423-425.

Lo, J.-C., Tsednee, M., Lo, Y. C., Yang, S.-C., Hu, J.-M., Ishizaki, K., Kohchi, T., Lee, D.-C., and Yeh, K. C. 2016. Evolutionary analysis of iron (Fe) acquisition system in *Marchantia polymorpha*. New Phytol. 211: 569-583.

Long, D. G. 1995. *Marchantia polymorpha* in Britain and Ireland. Bull. Brit. Bryol. Soc. 66: 29-37.

Loomis, P. D. and Maravolo, N. C. 1985. Modulation of amylolytic activity by gibberellin in the Hepatic, *Marchantia polymorpha*. Bryologist 88: 65-68.

Lorbeer, G. 1934. Die Zytologie der Lebermoose mit besonderer Berücksichtigung allgemeiner Chromosomenfragen. I. Jahrb. Wiss. Bot. 80: 567-818.

Los, D. A. and Semenenko, V. E. 1991. A computer search and analysis of chloroplast DNA sequences homologous to the light responsive elements of photosynthetic nuclear genes. Fiziol. Rast. 38(2): 213-221.

Lu, Y. W. and Huang, S. F. 2017. *Marchantia* L. (Marchantiaceae-Marchantiophyta) in Taiwan. Taiwania 62(1): 55-62.

Luís, L., Bergamini, A., and Sim-Sim, M. 2015. Which environmental factors best explain variation of species richness and composition of stream bryophytes? A case study from mountainous streams in Madeira Island. Aquat. Bot. 123: 37-46.

Mache, R. and Loiseaux, S. 1973. Light saturation of growth and photosynthesis of the shade plant *Marchantia polymorpha*. J. Cell Sci. 12: 391-401.

Mamczarz, H. 1970. The bryophyte communities in streams near Lacko in the Sadecki Beskid. Ann. Univ. Mariae Curie-Sklodowska, Sec. C Biol. 3. Biol. 25: 105-136.

Maravolo, N. C. 1976. Polarity and localization of auxin movement in the hepatic, *Marchantia polymorpha*. Amer. J. Bot. 63: 526-531.

Maravolo, N. C. 1980. Control of development in hepatics. Bull. Torrey Bot. Club 107: 308-324.

Maravolo, N. C. and Voth, P. D. 1966. Morphogenic effects of three growth substances on *Marchantia* gemmalings. Bot. Gaz. 127: 79-86.

Maravolo, N. C., Garber, E. D., and Voth, P. D. 1967. Biochemical changes during sexual development in *Marchantia polymorpha*: I. Esterases. Amer. J. Bot. 54: 1113-1117.

Maravolo, N. C., Otto, W. J., and Frueger, W. C. 1975. The influence of cytokinin and gibberellin on auxin mobilization; its role in regeneration and apical dominance in *Marchantia polymorpha*. Bot. Gaz. (Crawfordsville) 137: 231-236.

Marchetti, F., Cainzos, M., Cascallares, M., Distéfano, A. M., Setzes, N., López, G. A., Zabaleta, E., and Pagnussat, G. C. 2021. Heat stress in *Marchantia polymorpha*: Sensing and mechanisms underlying a dynamic response. Plant Cell Environ. 44: 2134-2149.

Markham, K. R. and Porter, I. J. 1974. Flavonoids of the liverwort *Marchantia polymorpha*. Phytochemistry 13: 1937-1942.

Markham, K. R. and Porter, L. J. 1978. Production of an aurone by bryophytes in the reproductive phase. Phytochemistry 17: 159-160.

Markham, K. R., Ryan, K. G., Bloor, S. J., and Mitchell, K. A. 1998. An increase in the luteolin: apigenin ratio in *Marchantia polymorpha* on UV-B enhancement. Phytochemistry 48: 791-794.

Martinez-Abaigar, J. and Ederra, A. 1992. Brioflora del río Iregua (La Rioja, España). Cryptog. Bryol. Lichenol. 13: 47-69.

Maschke, J. 1981. Moose als Bioindikatoren von Schwermetall-Immissionen. Eine Übersicht der bereits Untersuchten Lokalen und Regionalen Gebiete. Bryophyt. Biblioth. 22: 1-492.

Matsui, H., Iwakawa, H., Hyon, G. S., Yotsui, I., Katou, S., Monte, I., Nishihama, R., Franzen, R., Solano, R., and Nakagami, H. 2020. Isolation of natural fungal pathogens from *Marchantia polymorpha* reveals antagonism between salicylic acid and jasmonate during liverwort-fungus interactions. Plant Cell Physiol. 61: 265-275.

Matsui, K., Narahara, H., Kaijwara, T., and Hatinaka, A. 1991. Purification and properties of lipoxygenase in *Marchantia polymorpha* culture cells. Phytochemistry 30: 1499-1502.

Matsui, K., Kaji, Y., Kajiwara, T., and Hatanaka, A. 1996. Developmental changes of lipoxygenase and fatty acid hydroperoxide lyase activities in cultured cells of *Marchantia polymorpha*. Phytochemistry 41: 177-182.

Matsuo, A., Nakayama, N., and Nakayama, M. 1985. Enantiomeric type sesquiterpenoids of the liverwort *Marchantia polymorpha*. Phytochemistry 24: 777-781.

McConaha, M. 1941. Ventral structures effecting capillarity in the Marchantiales. Amer. J. Bot. 28: 301-306.

Mellway, R. D., Tran, L. T., Prouse, M. B., Campbell, M. M., and Constabel, C. P. 2009. The wound-, pathogen-, and ultraviolet B-responsive MYB134 gene encodes a R2R3 transcription factor that regulates proanthocyanidin synthesis in poplar. Plant Physiol. 150: 924-941.

Melstrom, C. E., Maravolo, N. C., and Stroemer, J. R. 1974. Endogenous gibberellins in *Marchantia polymorpha* and their possible physiological role in thallus elongation and orthogeotropic growth. Bryologist 77: 33-40.

Merwin, I. A. 2003. Orchard-floor management systems. In: Ferree, D. C., and Warrington, I. J. (eds.). Apples - Botany, Production and Uses. CABI Publ., Wallingford, England, pp. 303-318.

Messant, M., Hennebelle, T., Guérard, F., Gakière, B., Gall, A., Thomine, S., and Krieger-Liszkay, A. 2022. Manganese excess and deficiency affects photosynthesis and metabolism in *Marchantia polymorpha*. bioRxiv. <https://doi.org/10.1101/2022.01.24.477552>.

Mewari, N. and Kumar, P. 2008. Antimicrobial activity of extracts of *Marchantia polymorpha*. Pharmaceut. Biol. 46: 819-822.

Mewari, N. and Kumar, P. 2011. Evaluation of antifungal potential of *Marchantia polymorpha* L., *Dryopteris filix-mas* (L.) Schott and *Ephedra foliata* Boiss. against phyto fungal pathogens. Arch. Phytopath. Plant Protect. 44: 804-812.

Michelot-Gernez, M.-E. 1984. Evolution nucleaire au cours de la spermatogenese chez bryophytes. Ann. Sci. Nat. Bot. 13: 201-202.

Miller, M. W. 1964. A technique for isolating and culturing gemmae of *Marchantia polymorpha* L. under aseptic conditions. Bryologist 67: 317-320.

Miller, M. W. 1966. Relation between extrapolation number and apical cell number in gemmae of *Marchantia polymorpha* L. Nature (London) 210: 748-749.

Miller, M. W. and Alvarez, M. R. 1965. A relationship between extrapolation number and cellular kinetics in apical cells of gemmae of *Marchantia polymorpha* L. Bryologist 68: 184-192.

Miller, M. W. and Colaiace, J. 1969. The induction of sexual reproductive structures of *Marchantia polymorpha* grown under aseptic culture conditions. Bryologist 72: 45-48.

Miller, M. W. and Sparrow, A. H. 1964. Relationship between nuclear volume and radiosensitivity of different cell types in gemmae of *Marchantia polymorpha* L. Nature (London) 204: 596-597.

Miller, M. W. and Sparrow, A. H. 1965. The radiosensitivity of thalli of *Marchantia polymorpha* L. to acute gamma irradiation. Rad. Bot. 5: 567-580.

Miller, M. W. and Voth, P. D. 1962. Geotropic responses of *Marchantia*. Bryologist 65: 146-154.

Miller, M. W., Garber, E. D., and Voth, P. D. 1962a. Biosynthetic pathways in nutritionally deficient mutants of *Marchantia polymorpha* L. Nature (London) 195: 1220-1221.

Miller, M. W., Garber, E. D., and Voth, P. D. 1962b. Nutritionally deficient mutants of *Marchantia polymorpha* induced by X-rays. Bot. Gaz. 124: 94-102.

Miller, M. W., Sparrow, A. H., and Rogers, A. F. 1965. The radiosensitivity of gemmae of *Marchantia polymorpha* L. to acute gamma irradiation. Bryologist 68: 31-47.

Miranda, T. G., Alves, R. J. M., Assis, D. M. S. de, Sarah, A. T., Júnior, A. D. S. M., and Tavares-Martins, A. C. C. 2022. Atividade antifúngica de briófitas: Um estudo cienciométrico. [Antifungal activity of bryophytes: A scientometric study.]. Res. Soc. Dev. 11(4): e10111427127-e10111427127.

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.

Miyamura, S., Matsunaga, S., and Hori, T. 2002. High-speed video microscopical analysis of the flagellar movement of *Marchantia polymorpha* sperm. Bryol. Soc. Japan 8: 79-83.

Morton, B. R. 1994. Codon use and the rate of divergence of land plant chloroplast genes. Molec. Biol. Evol. 11: 231-238.

Müller, M. 2021. Foes or friends: ABA and ethylene interaction under abiotic stress. Plants 10(3): 448, 7 pp.

Murashige, T. and Skoog, F. 1962. A revised medium for rapid growth and bioassays with tobacco tissue cultures. Physiol. Plant. 15: 473-497.

Muratova, A., Golubev, S., Wittenmayer, L., Dmitrieva, T., Bondarenkova, A., Hirche, F., Mercbach, W., and Turkovskaya, O. 2009. Effect of the polycyclic aromatic hydrocarbon phenanthrene on root exudation of *Sorghum bicolor* (L.) Moench. Environ. Exper. Bot. 66: 514-521.

Miyamura, S., Matsunaga, S., and Hori, T. 2002. High-speed video microscopical analysis of the flagellar movement of *Marchantia polymorpha* sperm. Bryol. Soc. Japan 8: 79-83.

Nagai, J., Yamato, K. T., Sakaida, M., Yoda, H., Fukuzawa, H., and Ohyama, K. 1999. Expressed sequence tags from immature female sexual organ of a liverwort, *Marchantia polymorpha*. DNA Res. 6: 1-11.

Nahrstedt, A. and Davis, R. H. 1983. Occurrence, variation and biosynthesis of the cyanogenic glucosides linamarin and lotaustralin in species of the Heliconiini (Insecta: Lepidoptera). Compar. Biochem. Physiol. B Compar. Biochem. 75: 65-73.

Naidu, T. R. B. 1973. Occurrence of androgynous receptacles in *Marchantia polymorpha*. Bryologist 76: 427-429.

Nakayama, T., Fukushi, Y., Mizutani, J., and Tahara, S. 1996. Inhibiting effects of lunularic acid analogs on the growth of liverwort, watercress, and timothy grass. Biosci. Biotech. Biochem. 60: 862-865.

Nakazato, T., Kadota, A., and Wada, M. 1999. Photoinduction of spore germination in *Marchantia polymorpha* L. is mediated by photosynthesis. Plant Cell Physiol. 40: 1014-1020.

Naumov, N.A. 1964. Flora gribow Leningradskoj oblasti. Vol. II. Leningrad.

Navas, R., Pereira, M. R. R., Souza, G. S. F. de, and Martins, D. 2014. Physical and chemical methods for the control of *Marchantia polymorpha*. Científica (Jaboticabal) 42: 198-202.

Nehira, K. 1973. Adsorption of Ca in the differentiation of rhizoids in gemmae of *Marchantia polymorpha* L. Mem. Fac. Gen. Ed. Hiroshima Univ. 3(7): 1-6.

Nehira, K. 1977. A developmental study of gemmae in *Marchantia polymorpha* L. Hikobia 8: 104-109.

Nelson, J. M. 2017. Diversity and effects of the fungal endophytes of the liverwort *Marchantia polymorpha*. Ph.D. dissertation, Duke University, Durham, NC, USA.

Nelson, J. and Shaw, A. J. 2019. Exploring the natural microbiome of the model liverwort: Fungal endophyte diversity in *Marchantia polymorpha* L. Symbiosis 78(1): 45-59.

Nelson, J. M., Hauser, D. A., Hinson, R., and Shaw, A. J. 2018. A novel experimental system using the liverwort *Marchantia polymorpha* and its fungal endophytes reveals diverse and context-dependent effects. New Phytol. 218: 1217-1232.

Neubauer, C. and Schreiber, U. 1987. The polyphasic rise of chlorophyll fluorescence upon onset of strong continuous illumination: I. Saturation characteristics and partial control by the photosystem II acceptor side. Zeits. Naturforsch. C Biosci. 42: 1246-1254.

Newby, A., Altland, J. E., Gilliam, C. H., and Wehtje, G. 2007. Pre-emergence liverwort control in nursery containers. HortTechnology 17: 496-500.

Ninnemann, H. 1967. Über die Beteiligung von Wuchsstoffen, Phytochrom, Nucleinsäure- und Protein-synthese beim Etiolement der Brutkörper von *Marchantia polymorpha*. [On the involvement of growth substances, phytochrome, nucleic acid and protein synthesis in the etiolation of the brood bodies of *Marchantia polymorpha*.]. Biol. Zentralbl. 86: 303-364.

Ninnemann, H. and Halbsguth, W. 1965. Rolle des Phytochroms beim Etiolement von *Marchantia polymorpha*. [Role of phytochrome in the etiolement of *Marchantia polymorpha*.]. Naturwissenschaften 52(5): 110-111.

Nishihama, R., Ishizaki, K., Hosaka, M., Matsuda, Y., Kubota, A., and Kohchi, T. 2015. Phytochrome-mediated regulation of cell division and growth during regeneration and sporeling development in the liverwort *Marchantia polymorpha*. J. Plant Res. 128: 407-421.

Niu, C., Qu, J. B., and Lou, H. X. 2006. Antifungal bis [bibenzyls] from the Chinese liverwort *Marchantia polymorpha* L. Chem. Biodiv. 3(1): 34-40.

Oda, K., Kohchi, T., and Ohyama, K. 1992a. Mitochondrial DNA of *Marchantia polymorpha* as a single circular form with no incorporation of foreign DNA. Biosci. Biotechnol. Biochem. 56: 132-135.

Oda, K., Yamato, K., Ohta, E., Nakamura, Y., Takemura, M., Nozato, N., Akashi, K., Kanegae, T., Ogura, Y., Kohchi, T., and Ohyama, K. 1992b. Complete nucleotide sequence of the mitochondrial DNA from a liverwort, *Marchantia polymorpha*. Plant Molec. Biol. Reporter 10(2): 105-163.

Oda, K., Yamato, K., Ohta, E., Nakamura, Y., Takemura, M., Nozato, N., Akashi, K., and Ohyama, K. 1992c. Transfer RNA genes in the mitochondrial genome from a liverwort, *Marchantia polymorpha*; the absence of chloroplast-like TRNAs. Nucleic Acids Res. 20: 3773-3777.

Oda, K., Yamato, K., Ohta, E., Nakamura, Y., Takemura, M., Nozato, N., Akasi, K., Kanegae, T., Ogura, Y., Kohchi, T., and Ohyama, K. 1992d. Gene organization deduced from the complete sequence of liverwort *Marchantia polymorpha* mitochondrial DNA. A primitive form of plant mitochondrial genome. J. Molec. Biol. 223: 1-7.

Ogasawara, Y., Ishizaki, K., Kohchi, T., and Kodama, Y. 2013. Cold-induced organelle relocation in the liverwort *Marchantia polymorpha* L. Plant Cell Environ. 36: 1520-1528.

O'Hanlon, M. E. 1925. Germination of the spores and early stages in the development of the gametophyte of *Marchantia polymorpha*. Bot. Soc. Amer., Meeting Kansas City, Dec. 29-31.

O'Hanlon, M. E. 1926. Germination of the spores and early stages in the development of the gametophyte of *Marchantia polymorpha*. Bot. Gaz. 82: 215-222.

Ohyama, K. 2001. Molecular biology of the liverwort, *Marchantia polymorpha* L.: Construction of male and female genomic and EST libraries, sequencing of sex chromosomes, transformation system and gene-tagging for morphological mutants, ABSTRACT. Moss 2001: A meeting dedicated to moss biology, 27-29 May, Okazaki, Japan, schedule and abstracts.

Ohyama, K., Wetter, L. R., Yamano, Y., Fukuzawa, H., and Komano, T. 1982. A simple method for isolation of chloroplast DNA from *Marchantia polymorpha* L. cell suspension cultures. Agric. Biol. Chem. 46: 237-242.

Ohyama, K., Yamano, Y., Fukuzawa, H., Komano, T., Yamagishi, H., Fujimoto, S., and Sugiura, M. 1983. Physical mappings of chloroplast DNA from liverwort *Marchantia polymorpha* L. cell suspension cultures. Molec. Gen. Genet. 189: 1-9.

Ohyama, K., Fukuzawa, H., Kohchi, T., Shirai, H., Sano, T., Sano, S., Umesono, K., Shiki, Y., Takeuchi, M., Chang, Z., Aota, S.-I., Inokuchi, H., and Ozeki, H. 1986. Chloroplast gene organization deduced from complete sequence of liverwort *Marchantia polymorpha* chloroplast DNA. Nature 322: 572-574.

Ohyama, K., Kohchi, T., Fukuzawa, H., Sano, T., Umesono, K., and Ozeki, H. 1988a. Gene organization and newly identified groups of genes of the chloroplast genome from a liverwort, *Marchantia polymorpha*. Photosyn. Res. 16: 7-22.

Ohyama, K., Kohchi, T., Sano, T., and Yamada, Y. 1988b. Newly identified groups of genes in chloroplasts. Trends Biochem. Sci. 13: 19-22.

Ohyama, K., Fukuzawa, H., Kohchi, T., Sano, T., Sano, S., Shirai, H., Umesono, K., Shiki, Y., and Takeuchi, M. 1988c. Structure and organization of *Marchantia polymorpha* chloroplast genome, I. Cloning and gene identification. J. Molec. Biol. 203: 281-298.

Okada, S., Fujisawa, M., Sone, T., Nakayama, S., Nishiyama, R., Takenaka, M., Yamaoka, S., Sakaida, M., Kono, K., Takahama, M., Yamato, K. T., Fukuzawa, H., Brennicke, A., and Ohyama, K. 2000. Construction of male and female PAC genomic libraries suitable for identification of Y-chromosome-specific clones from the liverwort, *Marchantia polymorpha*. Plant J. 24: 421-428.

Okada, S., Sone, T., Fujisawa, M., Nakayama, S., Takenaka, M., Ishizaki, K., Kono, K., Shimizu-Ueda, Y., Hanajiri, T., Yamato, K T., Fukuzawa, H., Brennicke, A., and Ohyama, K. 2001. The Y chromosome in the liverwort *Marchantia polymorpha* has accumulated unique repeat sequences harboring a male-specific gene. Proc. Natl. Acad. Sci. 98: 9454-9459.

O'Toole, M. A. and Synnott, D. M. 1971. The bryophyte succession on blanket peat following calcium carbonate,

nitrogen, phosphorus and potassium fertilizers. J. Ecol. 59: 121-126.

Otto, K. R. 1976. Der Einfluß von äußeren Faktoren auf die Bildung von Primärrhizoiden bei Brutkörpern von *Marchantia polymorpha* L. [Influence of external factors on the formation of primary rhizoids on gemmae of *Marchantia polymorpha* L.] Zeit. Pflanzenphysiol. 80(3): 189-196.

Otto, K. R. and Halbsguth, W. 1976. Die Förderung der Bildung von Primärrhizoiden an Brutkörpern von *Marchantia polymorpha* L. durch Licht und IES. [The promotion of primary rhizoid formation on brood bodies of *Marchantia polymorpha* L. by light and IES.]. Zeit. Pflanzenphysiol. 80(3): 197-205.

Ozeki, H., Ohyama, K., Inokuchi, H., Fukuzawa, H., Kohchi, T., Sano, T., Nakahigashi, K., and Umesono, K. 1987. Genetic system of chloroplasts. Cold Spring Harbor Symp. Quant. Biol. 52: 791-804.

Ozkem, T. Y., Latife, C. I., and Selehattin, Y. 2019. The effect of heavy and alkaline metal accumulation on the chlorophyll content of *Fontinalis antipyretica* Hedw. and *Marchantia polymorpha* L. from Ida mountain. Fresenius Environ. Bull. 28: 8053-8061.

Pagoria, D. A. and Maravolo, N. C. 2005. DNA fragmentation in *Marchantia polymorpha* thalli in response to spermine treatment. Internat. J. Plant Sci. 166: 589-594.

Papp, B. 1998. Investigation of the bryoflora of some streams in Greece. Stud. Bot. Hung. 29: 59-67.

Papp, B., Erzberger, P., and Sabovljević, M. 2006. Contribution to the bryophyte flora of the Djerdap National Park (E Serbia). Stud. Bot. Hung. 37: 131-144.

Parihar, N. S. 1961. An Introduction to Embryophyta. Volume 1. Bryophyta. 4th ed. Central Book Depot, Allahabad, p. 57.

Paszewski, A., Bulanda, W., Dziubinska, H., and Trebacz, K. 1982. Resistance and capacity in the thallus of *Marchantia polymorpha*. Physiol. Plant. 54: 213-220.

Pedroza-Manrique, J. A. and Caballero Arévalo, M. 2009. Evaluating the effect of MS medium and temperature in developing *Marchantia polymorpha* L. (Marchantiaceae) propagules in *in vitro* and *ex vitro* conditions. Rev. Colomb. Biotecnol. 11(2): 85-104.

Pence, V. C. 1998. Cryopreservation of bryophytes: The effects of abscisic acid and encapsulation dehydration. Bryologist 101: 278-281.

Peumans, W. J., Fouquaert, E., Jauneau, A., Rougé, P., Lannoo, N., Hamada, H., Alvarez, R., Devreese, B., and Van Damme, E. J. van. 2007. The liverwort *Marchantia polymorpha* expresses orthologs of the fungal *Agaricus bisporus* agglutinin family. Plant Physiol. 144: 637-647.

Posno, M., Vliet, A. van, and Groot, G. S. P. 1986. Localization of chloroplast ribosomal protein genes on *Spirodela oligorhiza* chloroplast DNA. Curr. Genet. 10: 923-930.

Poveda, J. 2020a. *Marchantia polymorpha* as a model plant in the evolutionary study of plant-microorganism interactions. Curr. Plant Biol. 23: 100152.

Poveda, J. 2020b. *Marchantia polymorpha* ssp. *ruderalis* (Bischl. & Boissel.-Dub.) – arbuscular mycorrhizal fungi interaction: Beneficial or harmful? Symbiosis 82(3): 165-174.

Pressel, S. and Duckett, J. G. 2019. Do motile spermatozoids limit the effectiveness of sexual reproduction in bryophytes? Not in the liverwort *Marchantia polymorpha*. J. Syst. Evol. 57: 371-381.

Prior, P. V. and Brown, P. R. 1970. Germination and early growth responses of *Marchantia* gemmae to some growth regulators. Bryologist 73: 687-691.

Purkon, D. B., Fadhlillah, F. M., Maigoda, T. C., Iwo, M. I., Soemardji, A. A., Nadhifah, A., and Sudaryat, Y. 2022. Phytochemical, use in ethnomedicine, and therapeutic activities of *Marchantia* genus. J. Vocational Health Stud. 5(3): 174-185.

Qu, J. B., Xie, C. F., Ji, M., Shi, Y. Q., and Lou, H. X. 2007. Water-soluble constituents from the liverwort *Marchantia polymorpha*. Helv. Chim. Acta 90: 2109-2115.

Quattrocchio, F., Baudry, A., Lepiniec, L., and Grotewold, E. 2006. The regulation of flavonoid biosynthesis. In: Grotewold, E. (ed.). The Science of Flavonoids. Springer Science, New York, NY, USA, pp. 97-122.

Ramadhan, Z. A. and Agustien, A. 2019. Potential of endophytic bacteria from liverwort (*Marchantia polymorpha* L.) which produces antibiotics in *Staphylococcus aureus* L. World J. Pharmaceut. Res. 8(9): 171-178.

Rao, K. R., Kumar, N. R., and Reddy, A. N. 1979. Studies of photosynthesis in some liverworts. Bryologist 82: 286-289.

Raubeson, L. A. and Jansen, R. K. 1992. Chloroplast DNA evidence on the ancient evolutionary split in vascular land plants. Science 255: 1697-1699.

Rawitscher, F. 1932. Der Geotropismus der Pflanzen. Gustav Fisher, Jena.

Rees, D. C. and Juday, G. P. 2002. Plant species diversity on logged versus burned sites in central Alaska. Forest Ecol. Mgmt. 155: 291-302.

Register, T. E. and West, W. R. 1971 *Marchantia* life cycle. Carol. Biol. Supply Co. 34: 17-20.

Renner, S. S., Heinrichs, J., and Sousa, A. 2017. The sex chromosomes of bryophytes: Recent insights, open questions, and reinvestigations of *Frullania dilatata* and *Plagiochila asplenioides*. J. Syst. Evol. 55: 333-339.

Rethy, R., Fredericq, H., Maton, J., and Greef, J. de. 1976. The effect of different light treatments on the chlorophyll content of *Marchantia polymorpha* thalli. Biol. Jaarb. 44: 269-79.

Rethy, R., Fredericq, H., Greef, J. de, Maton, J., and Cappelle, M. 1977. Role of photosynthesis in the specific photoregulation of ethylene production in *Marchantia polymorpha* (L.). Arch. Internat. Biochim. 85: 1013-1014.

Rethy, R., Fredericq, H., Greef, J. de, and Maton, J. 1990. Light and the diagravitropic growth of *Marchantia polymorpha* L. thalli: Phytochrome-controlled epinasty. Mém. Soc. Roy. Bot. Belg. 12: 77-88.

Reynolds, A. C. and Maravolo, N. C. 1973. Phenolic compounds associated with development in the liverwort *Marchantia polymorpha*. Amer. J. Bot. 60: 406-413.

Reynolds, W. F. and Wolfe, S. L. 1984. Protamines in plant sperm [*Chara corallina*, *Marchantia polymorpha*, *Marsilea vestitia*]. Exper. Cell Res. 152: 443-448.

Rico-Reséndiz, F., Cervantes-Pérez, S. A., Espinal-Centeno, A., Dipp-Álvarez, M., Oropeza-Aburto, A., Hurtado-Bautista, E., Cruz-Hernández, A., Bowman, J. L., Ishizaki, K., Arteaga-Vázquez, M. A., Herrera-Estrella, L., and Cruz-Ramírez, A. 2020a. Transcriptional and morpho-physiological responses of *Marchantia polymorpha* upon phosphate starvation. Internat. J. Molec. Sci. 21(21): 8354.

Rico-Resendiz, F., Diaz-Santana, Z. H. U., Dipp-Alvarez, M., Cruz-Hernandez, A., Bowman, J. L., Herrera-Estrella, L., Ishizaki, K., Arteaga-Vazquez, M. A., and Cruz-Ramirez, A. 2020b, December. Phosphate Starvation Triggers Transcriptional Changes in the Biosynthesis and Signaling Pathways of Phytohormones in *Marchantia polymorpha*. In:

Biology and Life Sciences Forum 4(1): 89 <10.3390/IECPS2020-08729> MDPI.

Roberts, M. R. and Paul, N. D. 2006. Seduced by the dark side: Integrating molecular and ecological perspectives on the influence of light on plant defence against pests and pathogens. New Phytol. 170: 677-699.

Romani, F., Banić, E., Florent, S. N., Kanazawa, T., Goodger, J. Q., Mentink, R. A., Kierschke, T., Zachgo, S., Ueda, T., Bowman, J. L., Tsiantis, M., and Moreno, J. E. 2020. Oil body formation in *Marchantia polymorpha* is controlled by MpC1HDZ and serves as a defense against arthropod herbivores. Curr. Biol. 30: 2815-2828.

Rota, J. A. and Maravolo, N. C. 1975. Transport and mobilization of ^{14}C-Sucrose during regeneration in the hepatic, *Marchanti* (*sic*) *polymorpha*. Bot. Gaz. 136: 184-188.

Rousseau, J. 1952. Influence des heteroauxines sur la croissance des corbeilles a propagules de *Marchantia polymorpha* L. et de *Lunularia cruciata*. [Influence of heteroauxins on the growth of gemmae cups with propagules of *Marchantia polymorpha* L. and *Lunularia cruciata*.]. Rev. Bryol. Lichénol. 21: 239-241.

Rousseau, J. 1953. Action des heteroauxines sur les thalles de *Lunularia cruciata* Adams et de *Marchantia polymorpha* L. Rev. Bryol. Lichénol. 22: 22-25.

Rousseau, J. 1954a. Action des heteroauxines a l'obscurite sur les propagules de *Marchantia polymorpha* L. Compt. Rend. Acad. Sci. (Paris) 238: 2111-2112.

Rousseau, J. 1954b. Inhibition de la croissance des propagules de *Marchantia polymorpha* L. par la coumarine. [Inhibition of the growth of *Marchantia polymorpha* L. propagules by coumarin.]. Compt. Rend. Acad. Sci. (Paris) 239: 1420-1422.

Rövekamp, M., Bowman, J. L., and Grossniklaus, U. 2016. *Marchantia* MpRKD regulates the gametophyte-sporophyte transition by keeping egg cells quiescent in the absence of fertilization. Curr. Biol. 26: 1782-1789.

Rubin, G., Tohge, T., Matsuda, F., Saito, K., and Scheible, W. R. 2009. Members of the LBD family of transcription factors repress anthocyanin synthesis and affect additional nitrogen responses in *Arabidopsis.* Plant Cell 21: 3567-3584.

Samecka-Cymerman, A., Marczonek, A., and Kempers, A. J. 1997. Bioindication of heavy metals in soil by liverworts. Arch. Environ. Contam. Toxicol. 33: 162-171.

Sancha, G. S. 2017. Effects of ultraviolet radiation on Bryophytes. Ph. D. dissertation, Universidad de La Rioja.

Särkkä, L. and Tahvonen, R. 2020. Control of liverwort (*Marchantia polymorpha* L.) growth in nursery plants with mulches of *Sphagnum* moss and blackcurrant stem pieces. Agric. Food Sci. 29: 250-256.

Sarosiek, J. and Wozakowska-Natkaniec, H. 1967. Response of *Marchantia polymorpha* L. to chronic gamma radiation under natural conditions. Acta Soc. Bot. Poloniae 36: 187-197.

Saruwatari, M., Takio, S., and Ono, K. 1999. Low temperature-induced accumulation of eicosapentaenoic acids in *Marchantia polymorpha* cells. Phytochemistry 52: 367-372.

Sato, F., Yamada, Y., Kwak, S. S., Inchinose, K., Kishda, M., Takaahasi, N., and Yoshida, S. 1991. Photoautotrophic cultured cells: A novel system to survey new photosynthetic electron transport inhibitors. Zeit. Naturforsch., C. Biochem., Biophys., Biol., Virol. 46: 563-568.

Sawangproh, W., Ekroos, J., and Cronberg, N. 2016. The effect of ambient temperature on larvae of *Scatopsciara cunicularius* (Diptera: Sciaridae) feeding on the thallose liverwort *Marchantia polymorpha*. Eur. J. Entomol. 113: 259-264.

Scarlett, P. and O'Hare, M. 2006. Community structure of in-stream bryophytes in English and Welsh rivers. Hydrobiologia 553: 143-152.

Schmid, M. W., Giraldo-Fonseca, A., Rövekamp, M., Smetanin, D., Bowman, J. L., and Grossniklaus, U. 2018. Extensive epigenetic reprogramming during the life cycle of *Marchantia polymorpha*. Genome Biol. 19(1): 1-17.

Schneider, M. J., Voth, P. D., and Troxler, R. F. 1967. Methods of propagating bryophyte plants, tissues, and propagules. Bot. Gaz. 128: 169-174.

Schofield, W. 1997. Bryophytes unintentionally introduced to British Columbia. Botanical Electronic News - BEN #162, Accessed on 10 December 2006 at <http://www.ou.edu/cas/botany-micro/ben/ben162.html>.

Schönherr, J. and Ziegler, H. 1975. Hydrophobic cuticular ledges prevent water entering the air pores of liverwort thalli. Planta 124: 51-60.

Schott, R. T., Nebel, M., and Roth-Nebelsick, A. 2021. Comparison of the freezing behavior of two liverwort species–*Conocephalum salebrosum* and *Marchantia polymorpha* ssp. *ruderalis*. Lindbergia 44: 1-10.

Schuster, R. M. 1966. The Hepaticae and Anthocerotae of North America East of the Hundredth Meridian, Vol. 1. Columbia University Press, NY, 802 pp.

Seaman, J. J., Orth, T. L., Mueller, R. E., and Maravolo, N. C. 2005. The influence of spermine on the activity of a protein kinase C-like enzyme in *Marchantia polymorpha* thalli during programmed cell death. Bryologist 108: 110-117.

Sharma, S. 2007. *Marchantia polymorpha* L.: A bioaccumulator. Aerobiologia 23(3): 181-187.

Sharma, A., Bargali, K., and Pande, N. 2009. The allelopathic potential of bryophyte extract on seed germination and seedling growth of *Bidens biternata*. Nat. Sci. 7: 30-38.

Sheldrake, A. R. 1971. The occurrence and significance of auxin in the substrata of bryophytes. New Phytol. 70: 519-526.

Shi, D.-J., Wu, P.-C., Qiu, Y.-Y., and Wang, M.-Z. 1992. Comparative studies on photosynthetic fluorescence spectra and fluorescence kinetics of bryophytes. Acta Phytotax. Sinica 30: 320-330.

Shibaya, T. and Sugawara, Y. 2007. Involvement of arabinogalactan proteins in the regeneration process of cultured protoplasts of *Marchantia polymorpha*. Physiol. Plant. 130: 271-279.

Shibaya, T., Kaneko, Y., and Sugawara, Y. 2005. Involvement of arabinogalactan proteins in protonemata development from cultured cells of *Marchantia polymorpha*. Physiol. Plant. 124: 504-514.

Shimamura, M. 2016. *Marchantia polymorpha*: Taxonomy, phylogeny and morphology of a model system. Plant Cell Physiol. 57: 230-256.

Shinmen, Y., Katoh, K., Shimizu, S., Jareonkitmongkol, S., and Yamada, H. 1991. Production of arachidonic acid and eicosapentaenoic acids by *Marchantia polymorpha* in cell culture. Phytochemistry 30: 3255-3260.

Sibbald, P. R. 1988. Patterns of base usage, nearest neighbour analysis and identification of genes in two completely sequenced chloroplast genomes. Curr. Genet. 13: 523-530.

Singh, V. B., Dixit, B. S., and Srivastava, S. N. 1987. Chemical examination of *Marchantia polymorpha* with reference to its structure, habit and distribution. J. Econ. Tax. Bot. 10: 421-423.

Skutch, A. F. 1929. Early stages of plant succession following forest fires. Ecology 10: 177-190.

Smooker, P. M., Kruft, V., and Subramanian, A. R. 1990. A ribosomal protein is encoded in the chloroplast DNA in a lower plant but in the nucleus in angiosperms. Isolation of the spinach L21 protein and cDNA clone with transit and a unusual repeat sequence. J. Biol. Chem. 265: 16699-16703.

Söderström, L., Gradstein, S. R., and Hagborg, A. 2010. Checklist of the hornworts and liverworts of Java. Phytotaxa 9: 53-149.

Söderström, L., Hagborg, A., Konrat, M. von, Bartholomew-Began, S., Bell, D, Briscoe, L., Brown, E., Cargill, D. C., Costa, D. P., Crandall-Stotler, B. J., Cooper, E. D., Dauphin, G., Engel, J. J., Feldberg, K., Glenny, D., Gradstein, S. R., He, X., Heinrichs, J., Hentschel, J., Ilkiu-Borges, A. L., Katagiri, T., Konstantinova, N. A., Larraín, J., Long, D. G., Nebel, M., Pócs, T., Puche, F., Reiner-Drehwald, E., Renner, M. A. M., Sass-Gyarmati, A., Schäfer-Verwimp, A., Moragues, J. G. S., Stotler, R. E., Sukkharak, P., Thiers, B. M., Uribe, J., Váňa, J., Villarreal, J. C., Wigginton, M., Zhang, L., and Zhu, R.-L. 2016. World checklist of hornworts and liverworts. PhytoKeys 59: 1-828.

Son, N. C. T., Tan, T. Q., Lien, D. T. M., Huong, N. T. M., Tuyen, P. N. K., Phung, N. K. P., Phung, Q. N. D., and Thu, N. T. H. 2020. Five phenolic compounds from *Marchantia polymorpha* L. and their *in vitro* antibacterial, antioxidant and cytotoxic activities. Vietnam J. Chem. 58: 810-814.

Sone, T., Fujisawa, M., Takenaka, M., Nakagawa, S., Yamaoka, S., Sakaida, M., Nishiyama, R., Yamato, K. T., Ohmido, N., Fukui, K., Fukuzawa, H., and Ohyama, K. 1999. Bryophyte 5S rDNA was inserted into 45S rDNA repeat units after the divergence from higher land plants. Plant Molec. Biol. 41: 679-685.

Sørensen, H. 1948. Studies on the ecology of Danish water- and bog mosses. Dansk Bot. Ark. 12(10): 1-46.

Soriano, G., Del-Castillo-Alonso, M. Á., Monforte, L., Tomás-Las-Heras, R., Martínez-Abaigar, J., and Núñez-Olivera, E. 2019a. Photosynthetically-active radiation, UV-A and UV-B, causes both common and specific damage and photoprotective responses in the model liverwort *Marchantia polymorpha* ssp. *ruderalis*. Photochem. Photobiol. Sci. 18: 400-412.

Soriano, G., Del-Castillo-Alonso, M. Á., Monforte, L., Núñez-Olivera, E., and Martínez-Abaigar, J. 2019b. Phenolic compounds from different bryophyte species and cell compartments respond specifically to ultraviolet radiation, but not particularly quickly. Plant Physiol. Biochem. 134: 137-144.

Soriano, G., Del-Castillo-Alonso, M. Á., Monforte, L., Tomás-Las-Heras, R., Martínez-Abaigar, J., and Núñez-Olivera, E. 2021. Developmental stage determines the accumulation pattern of UV-absorbing compounds in the model liverwort *Marchantia polymorpha* ssp. *ruderalis* under controlled conditions. Plants 10(3): 473.

Spinedi, N., Storb, R., Aranda, E., Romani, F., Svriz, M., Varela, S. A., Moreno, J. E., Fracchia, S., Cabrera, J., Batista-Garcia, R. A., Ponce de León, I., and Scervino, J. M. 2021. ROS-scavenging enzymes as an antioxidant response to high concentration of anthracene in the liverwort *Marchantia polymorpha* L. Plants 10(7): 1478, 18 pp.

Stanislaus, R. C. and Maravolo, N. C. 1994. The influence of polyamines on senescence in *Marchantia polymorpha*. Bryologist 97: 162-165.

Stelmasiewicz, M., Świątek, Ł., and Ludwiczuk, A. 2021. Phytochemical profile and anticancer potential of endophytic microorganisms from liverwort species, *Marchantia polymorpha* L. Molecules 27(1): 153, 11 pp.

Stevenson, C. R. and Hill, M. O. 2008. Urban myths exploded: Results of a bryological survey of King's Lynn. J. Bryol. 30: 12-22.

Strasburger, E. 1869. Die Geschlechtsorgane und die Befruchtung bei *Marchantia polymorpha* L. Jahrb. Wiss. Bot. 7: 409-422.

Sugawara, Y., Mori, K., Matsushima, H., and Takeuchi, M. 1983. Enhancement of cell division in *Marchantia* protoplast culture by activated charcoal. Zeits. Pflanzenphysiol. 109: 275-278.

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. Plant Physiol. 124: 971-978.

Suzuki, H., Harrison, C. J., Shimamura, M., Kohchi, T., and Nishihama, R. 2020. Positional cues regulate dorsal organ formation in the liverwort *Marchantia polymorpha*. J. Plant Res. 133: 311-321.

Synnott, D. M. 1987. The effects of drainage, shelter and fertilisers on bryophyte colonisation and succession on blanket peat in Western Ireland. Glasra 10: 83-99.

Takaki, N. 1967. Bryophytes of Mt. Showashinzan, an active volcano formed by the 1943-'45 eruption. J. Jap. Bot. 42: 23-27.

Takaoki, T. and Mitani, K. 1986. A new fumigation method for measuring the effects of sulphur dioxide on photosynthesis of bryophytes and lichens. Lindbergia 12: 60-66.

Takemura, M., Oda, K., Yamato, K., Ohta, E., Nakamura, Y., Nozato, N., Akashi, K., and Ohyama, K. 1992. Gene clusters for ribosomal proteins in the mitochondrial genome of a liverwort, *Marchantia polymorpha*. Nucleic Acids Res. 20: 3199-3205.

Takemura, M., Okimura, Y., Kida, H., Hamada, T., and Ohyama, K. 2011. Blue light enhances the accumulation of eicosapentaenoic acid in a liverwort, *Marchantia polymorpha* L. Plant Biotech. 28: 489-492.

Takemura, M., Hamada, T., Kida, H., and Ohyama, K. 2012. Cold-induced accumulation of ω-3 polyunsaturated fatty acid in a liverwort, *Marchantia polymorpha* L. Biosci. Biotech. Biochem. 76: 785-790.

Takenaka, M., Yamaoka, S., Hanajiri, T., Shimizu-Ueda, Y., Yamato, K. T., Fukuzawa, H., and Ohyama, K. 2000. Direct transformation and plant regeneration of the haploid liverwort *Marchantia polymorpha* L. Transgen. Res. 9: 179-185.

Takeuchi, M., Matsushima, H., and Sugawara, Y. 1980. Long-term freeze-preservation of protoplasts of carrot and *Marchantia*. Cryo-letters 1: 519-524.

Takikawa, Y., Senga, Y., Nonomura, T., Matsuda, Y., Kakutani, K., and Toyoda, H. 2014. Targeted destruction of fungal structures of *Erysiphe trifoliorum* on flat leaf surfaces of *Marchantia polymorpha*. Plant Biol. 16(1): 291-295.

Takio, S. 1987. Coenzyme requirements of nitrate reductase in extracts from suspension cultured cells of four bryophyte species. J. Hattori Bot. Lab. 62: 269-280.

Takizawa, R., Hatada, M., Moriwaki, Y., Abe, S., Yamashita, Y., Arimitsu, R., Yamato, K. T, Nishihama, R., Kohchi, T., Koeduka, T., Chen, F., and Matsui, K. 2021. Fungal-type terpene synthases in *Marchantia polymorpha* are involved in sesquiterpene biosynthesis in oil body cells. Plant Cell Physiol. 62: 528-537.

Tanaka, M., Esaki, T., Kenmoku, H., Koeduka, T., Kiyoyama, Y., Masujima, T., Asakawa, Y., and Matsui, K. 2016. Direct evidence of specific localization of sesquiterpenes and

marchantin A in oil body cells of *Marchantia polymorpha* L. Phytochemistry 130: 77-84.

Tanurdzic, M. and Banks, J. 2004. Sex-determining mechanisms in land plants. Plant Cell 16: S61-S71.

Tarén, N. 1958. Factors regulating the initial development of gemmae in *Marchantia polymorpha*. Bryologist 61: 191-204.

Taylor, L. P. and Grotewold, E. 2005. Flavonoids as developmental regulators. Curr. Opinion Plant Biol. 8: 317-323.

Terui, K. 1981. Growth and gemma-cup formation in relation to archegoniophore protrusion in *Marchantia polymorpha* L. Ann. Rept. Fac. Ed. Iwate Univ. 40: 19-28.

Thatcher, E. P. 1949. Bryophytes of an artificially illuminated cave. Bryologist 52: 212-214.

Thomas, F., Corre, E., and Cébron, A. 2019. Stable isotope probing and metagenomics highlight the effect of plants on uncultured phenanthrene-degrading bacterial consortium in polluted soil. ISME J. 13: 1814-1830.

Thullen, R. J. 1965. The response of *Marchantia polymorpha* L. gemmae in aseptic culture to temperature light and substrate. M.S. thesis, University of Montana, Bozeman, 26 pp.

Torrey, R. H. 1932. Another report of *Marchantia polymorpha* after forest fires. Torreya 32: 128-129.

Tougane, K., Komatsu, K., Bhyan, S. B., Sakata, Y., Ishizaki, K., Yamato, K. T., Kohchi, T., and Takezawa, D. 2010. Evolutionarily conserved regulatory mechanisms of abscisic acid signaling in land plants: Characterization of ABSCISIC ACID INSENSITIVE1-like type 2C protein phosphatase in the liverwort *Marchantia polymorpha*. Plant Physiol. 152: 1529-1543.

Treutter, D. 2006. Significance of flavonoids in plant resistance: a review. Environ. Chem. Lett. 4(3): 147-157.

Tsuboyama, S., Nonaka, S., Ezura, H., and Kodama, Y. 2018. Improved G-AgarTrap: A highly efficient transformation method for intact gemmalings of the liverwort *Marchantia polymorpha*. Sci. Rept. 8(1): 1-10.

Umesono, K. and Ozeki, H. 1987. Chloroplast gene organization in plants. Trends Genet. 3: 281-287.

Une, K. 1984. A field observation on the reproductive mode in *Marchantia polymorpha* L. Hikobia 9: 15-18.

Une, K. 1998. Bryophytes as teaching materials. Hikobia 12: 373-379.

Uribe, J. and Aguirre, J. 1995. Las especies Colombianas del género *Symphyogyna* (Hepaticae: Pallaviciniaceae). Caldasia 17: 429-458.

Uva, R. H., Neal, J. C., and DiTomaso, J. M. 1997. Weeds of the Northeast. Comstock Publishing Associates, Ithaca, N. Y., viii + 397 pp.

Verkley, G., Aa, H. A. van der, and Cock, G. W. 1997. *Bryoscyphus atromarginatus spec. nov*. (Leotiaceae), a new ascomycete parasitizing the thallus of *Marchantia polymorpha*. Persoonia 16: 383-387.

Veroustraete, F., Fredericq, H., Wiemeersch, L. V., and Greef, J. D. 1982. Specific photoregulation by phytochrome of epinasty and light - induced ethylene production in *Marchantia polymorpha*. Photochem. Photobiol. 35: 261-264.

Vieira, C., Sérgio, C., and Séneca, A. 2005. Threatened bryophytes occurrence in Portuguese stream habitat. Bol. Soc. Española Bríol. 26: 103-118.

Villarreal, J. C. and Renner, S. S. 2012. Hornwort pyrenoids, carbon-concentrating structures, evolved and were lost at least five times during the last 100 million years. Proc. Natl. Acad. Sci. 109: 18873-18878.

Vitt, D. H., Glime, J. M., and LaFarge-England, C. 1986. Bryophyte vegetation and habitat gradients of montane streams in western Canada. Hikobia 9: 367-385.

Vöchting, H. 1885. Über die Regeneration der Marchantieen. Pringsh. Jahrb. 16: 297.

Vöchting, H. 1887. Uber Theilbareit im Pflanzen reich und die Wirkung innerer und ausserer Krafte auf Organbildung an Pflanzentheilen. Pflugers Arch. Ges. Physiol. 15: 123-190.

Volchenkova, T. A., Kalabina, N. A., Schaefer, C., Zubov, V. P., and Zaitsev, S. Y. 2001. A study of moss (*Marchantia polymorpha*) thylakoid membrane lipids in monolayers. Membr. Cell Biol. 14: 579-585.

Voth, P. D. 1941. Gemmae-cup production in *Marchantia polymorpha* and its response to calcium deficiency and supply of other nutrients. Bot. Gaz. 103: 310-325.

Voth, P. D. 1943. Effects of nutrient-solution concentration on the growth of *Marchantia polymorpha*. Bot. Gaz. 104: 591-601.

Voth, P. D. and Hamner, K. C. 1940. Responses of *Marchantia polymorpha* to nutrient supply and photoperiod. Bot. Gaz. 102: 169-205.

Walkinshaw, C. H., Sweet, H. C., Venketeswaran, S., and Horne, W. H. 1970. Results of Apollo 11 and 12 quarantine studies on plants. BioScience 20: 1297-1302.

Wang, N., Liu, W., Zhang, T., Jiang, S., Xu, H., Wang, Y., Zhang, Z., Wang, C., and Chen, X. 2018. Transcriptomic analysis of red-fleshed apples reveals the novel role of MdWRKY11 in flavonoid and anthocyanin biosynthesis. J. Agric. Food Chem. 66: 7076-7086.

Wang, X., Cao, J., Wu, Y., Wang, Q., and Xiao, J. 2016. Flavonoids, antioxidant potential, and acetylcholinesterase inhibition activity of the extracts from the gametophyte and archegoniophore of *Marchantia polymorpha* L. Molecules 21(3), 360.

Wann, F. B. 1925. Some of the factors involved in the sexual reproduction of *Marchantia polymorpha*. Amer. J. Bot. 12: 307-318.

Watson, W. 1919. The bryophytes and lichens of fresh water. J. Ecol. 7: 71-83.

Wei, H., Song, S., Tian, H., and Liu, T. 2014. Effects of phenanthrene on seed germination and some physiological activities of wheat seedling. Compt. Rend. Biol: 337(2): 95-100.

Weiler, E. W. 1979. Radioimmunoassay for the detection of free and conjugated abscisic acid. Planta 144: 255-263.

Weinert, H. 1909. Untersuchungen über Wachstum und tropistische Bewegungserscheinungen der Rhizoiden thalloser Lebermoose. Bot. Zeit. 12: 29, 202-230.

Weis, E., Wamper, D., and Santarius, K. A. 1986. Heat sensitivity and thermal adaptation of photosynthesis in liverwort thalli. Oecologia 69: 134-139.

Went, F. W. and Thimann, K. V. 1937. Phytohormones. Macmillan Company, New York.

West, G. 1910. An epitome of a comparative study of the dominant phanerogamic and higher cryptogamic flora of aquatic habit, in seven lake areas of Scotland. In: Murray, J. Bathymetrical Survey of the Scottish Fresh-water Locks, Conducted Under The Direction of Sir John Murray… and Laurence Pullar… During The Years 1897 to 1909. Report on the Scientific Results. Edinburgh, Challenger Office, pp. 156-260.

Whittemore, A. T. 1991. The secondary chemistry of the Marchantiales. Adv. Bryol. 4: 75-102.

Wu, L. and Bradshaw, A. 1972. Aerial pollution and the rapid evolution of copper tolerance. Nature 238: 167-169.

Wu, P.-C. and Yu, J. 2003. The medicinal uses of bryophytes. Acta Bot. Yunnan (Suppl.) 14: 51-55.

Xu, B., Taylor, L., Pucker, B., Feng, T., Glover, B. J., and Brockington, S. F. 2021. The land plant – specific MIXTA-MYB lineage is implicated in the early evolution of the plant cuticle and the colonization of land. New Phytol. 229: 2324-2338.

Yamaoka, S., Inoue, K., and Araki, T. 2021. Regulation of gametangia and gametangiophore initiation in the liverwort *Marchantia polymorpha*. Plant Repro. 34: 297-306.

Yamato, K. T., Ishizaki, K., Fujisawa, M., Okada, S., Nakayama, S., Fujishita, M., Bando, H., Yodoya, K., Hayashi, K., Bando, T., Hasumi, A., Nishio, T., Sakata, R., Yamamoto, M., Yamaki, A., Kajikawa, M., Yamano, T., Nishide, T., Choi, S.-H., Shimizu-Ueda, Y., Hanajiri, T., Sakaida, M., Kono, K., Takenaka, M., Yamaoka, S., Kuriyama, C., Kohzu, Y., Nishida, H., Brennicke, A., Shin-i, T., Kohara, Y., Kohchi, T., Fukuzawa, H., and Ohyama, K. 2007. Gene organization of the liverwort Y chromosome reveals distinct sex chromosome evolution in a haploid system. Proc. Natl. Acad. Sci. USA 104: 6472-6477.

Yasui, Y., Tsukamoto, S., Sugaya, T., Nishihama, R., Wang, Q., Kato, H., Yamato, K. T., Fukaki, H., Mimura, T., Kubo, H., Theres, K., Kohchi, T., and Ishizaki, K. 2019. GEMMA CUP-ASSOCIATED MYB1, an ortholog of axillary meristem regulators, is essential in vegetative reproduction in *Marchantia polymorpha*. Curr. Biol. 29: 3987-3995.

Yokobori, M. 1978. Measuring of phytotoxic air pollution based upon response of bryophytes using filtered-air growth chamber. Jap. J. Ecol. 28: 17-23.

Yokobori, M. and Taoda, H. 1980. Nachweis der phytotoxischen Wirkung von Luftverunreinigungen durch Messung der Reaktion von Bryophyten mit dem "Bryometer." [Proof of the phytotoxic effect of air pollution by measuring the reaction of bryophytes with the "Bryometer."]. Staub-Reinhalt. 40: 490-496.

Yoshikawa, M., Luo, W., Tanaka, G., Konishi, Y., Matsuura, H., and Takahashi, K. 2018. Wounding stress induces phenylalanine ammonia lyases, leading to the accumulation of phenylpropanoids in the model liverwort *Marchantia polymorpha*. Phytochemistry 155: 30-36.

Young, S. B. and Kläy, J.-R. 1971. Bryophytes in the 1969 crater of Deception Island, Antarctica: An apparent case of rapid long-distance dispersal. Ohio J. Sci. 71: 358-362.

Zehr, D. R. 1990. A simplified technique for assaying the production of bacteriostatic compounds by hepatics. Lindbergia 16: 128-132.

Zheng, T. X. and Shimamura, M. 2022. Taxonomic revision of the genus *Marchantia* (Marchantiaceae) in Japan and the redefinition of the genus. Hattoria 13: 33-77.

Zhu, R. L., Wang, D., Xu, L., Shi, R. P., Wang, J., and Zheng, M. 2006. Antibacterial activity in extracts of some bryophytes from China and Mongolia. J. Hattori Bot. Lab. 100: 603-615.

Zinsmeister, D. D. and Carothers, Z. B. 1974. The fine structure of oogenesis in *Marchantia polymorpha*. Amer. J. Bot. 61: 499-512.