CHAPTER 8-4
NUTRIENT RELATIONS: UPTAKE AND LOCATION

TABLE OF CONTENTS

CHAPTER 8-4
NUTRIENT RELATIONS: UPTAKE AND LOCATION

Figure 1. Drops of "steam" from geothermal vents resting on the wire-like leaves of ***Campylopus holomitrius*** and providing a source of nutrients and a means of trapping airborne nutrients. Photo by Janice Glime.

Uptake

The **role** of bryophytes in nutrient uptake within ecosystems is generally ignored because of their small stature. Weetman and Timmer (1967) showed that the common feather moss ***Pleurozium schreberi*** (Figure 2) in a black spruce (*Picea mariana*; Figure 3) forest took up only 23-53% of N, P, K, and Mg taken up by trees. Nevertheless, that is a non-trivial figure. But bryophyte contributions to sequestering nutrients can be substantial. Yet we have little concept of whether their net contribution is beneficial or detrimental in those ecosystems where they abound.

Figure 2. ***Pleurozium schreberi***, a common feather moss in black spruce forests. Photo by Michael Lüth, with permission.

Figure 3. ***Picea mariana*** forest in Northern Alberta, Canada, with ***Pleurozium schreberi*** and ***Hylocomium splendens***. Photo by Richard Caners, with permission.

General Considerations

Sources

First we need to recall that the sources of nutrients for bryophytes include precipitation, dust, and to a limited extent, substrate. Brown (1982) explains that bryophytes absorb mineral nutrients over their entire surface (Figure 1). This ability is promoted by two characteristics of bryophytes: a large surface area to volume ratio, and a low surface resistance, relative to **tracheophytes** (lignified

vascular plants), due to the limited development of cuticle. This is further enhanced within the bryophyte by typically having leaves of only one cell layer in thickness, hence exposing every leaf cell directly to the nutrient supply immediately. Based on what we know thus far, actual entry into the cell is most likely similar to that of tracheophytes.

Site of Uptake

Their typical differences in site of uptake would seemingly remove the bryophytes from competition with tracheophytes for soil nutrients. Due to lack of vessels and tracheids, we have assumed that uptake of nutrients by bryophytes is primarily through their leaves. Even in the endohydric ***Polytrichum*** (Figure 4), water entry is gained primarily at the tips of the plants by water that has travelled upward through external capillary spaces (Trachtenberg & Zamski 1979). Hence, we can expect that most nutrient entry is not through rhizoids, but through leaves, and at least in some mosses may be greater at the tips than in lower parts of the plant. Brown and Wells (1990) remind us that despite their small size, the bryophytes have intricate surface areas that are effective at trapping both dust and moisture that can subsequently enter the bryophyte (Figure 1). The ratio of surface area to volume in bryophytes is enormous compared to that of trees and other tracheophytes.

Figure 4. ***Polytrichum commune***, a moss with internal conduction, but that transports nutrients externally through capillary spaces. Photo by Michael Lüth, with permission.

Rhizoids

Even if bryophytes were to use their rhizoids to gather some nutrients, the soil penetration by these structures is generally shallow and well above the zone occupied by most fine roots of tracheophytes, especially trees. Instead, we have assumed that bryophytes typically rely largely on dust on their surfaces and on nutrients dissolved in rainfall. In forests, these arrive primarily through leachates acquired in canopy throughfall. This is a quite different strategy from that of tracheophytes, although in ***Polytrichum commune*** (Figure 4) it does appear that some nutrients might enter through the rhizoids (Chapin *et al*. 1987). On the other hand, ***P. commune*** and other forest floor mosses in the black spruce forest (Figure 3) lose nutrients to the black spruce fine roots through **mycorrhizae** (fungal associates).

Growth Form

Growth form affects nutrient trapping and subsequent uptake. Taylor and Witherspoon (1972) found that ***Dicranum*** (Figure 5), which grows in a relatively tight clump, retains more particles than do open lichens such as *Cladonia*/*Cladina* (Figure 6), even though these lichens display considerable surface area. Hence, we should expect such tight cushions to be more effective at trapping than more open bryophytes like ***Brachythecium*** (Figure 7) or ***Mnium*** (Figure 8). On the other hand, Shacklette (1965) found that bryophytes were significantly contaminated with soil particles, including insoluble ones such as Al, Be, Fe, Si, and Zr. But, it would appear that even deeper soil is not immune to moss nutrient scavenging, perhaps through a combination of capillary action and concentration gradient.

Figure 5. ***Dicranum*** in its dry state, showing tight growth form that traps dust particles easily. Photo courtesy of Herschel Horton.

Figure 6. ***Cladina portentosa***, a highly branched lichen. Photo by Taka, through Creative Commons.

Figure 7. ***Brachythecium rutabulum***, showing open growth form that traps less dust than the more cushiony forms like ***Dicranum***. Photo by Janice Glime.

Figure 8. ***Mnium hornum***, an open bryophyte that may trap less dust than cushion forms. Photo by Tim Waters, through Creative Commons.

Age

Bryophyte uptake can relate to **age**. In studying the Alaskan black spruce (*Picea mariana*) forest (Figure 3), Chapin *et al*. (1987) found that in three of the moss taxa studied, the phosphate absorption capacity increases with age of green tissue, but decreases with age of brown tissue.

In the aquatic moss ***Warnstorfia fluitans*** (Figure 9), an **acidophile** (preferring acid habitats), iron (Fe) **accumulates** in the cell wall (Satake 2000). The highest concentrations are in the base, increasing toward the tip. In addition to the biological accumulation within the walls, iron is held on the mosses in crystal form.

Figure 9. ***Warnstorfia fluitans***, a species that can accumulate iron in its cell walls. Photo by Michael Luth,with permission.

Nutrient Concentration

Lou *et al*. (2013) found that the content of the heavy metals Pb, Cr, and Cu in the moss ***Haplocladium microphyllum*** (Figure 10) correlated with the concentrations in the medium. Iron (Fe), on the other hand, increased in a similar manner until the concentration in the medium reached 400 mg L^{-1} as Fe^{++}. Below that level the iron facilitated uptake of other nutrient ions. The absorption capacity for these metals follows the order Fe>Cr>Cu>Pb. Like most things in nutrient relationships, the amount matters. At low concentrations, both lead and copper ions, as with iron, promote the absorptive capacity of other nutrient elements. At high concentrations the same metals decrease uptake of other nutrient elements. Chromium is an exception, inhibiting absorption capacity of the nutrients P, K, Ca, S, Fe, and Cu even when the Cr concentrations are low. Lou and coworkers found that Pb and Cr are stored primarily in the peripheral cortex of the moss stem in ***Haplocladium microphyllum***. It is not clear how this affects uptake of other ions.

Figure 10. ***Haplocladium microphyllum***, a species in which uptake is dependent on concentration. Photo by Robin Bovey, with permission through Dale Vitt.

Water Source

The standing or flowing water habitat of ***Sphagnum*** fen (Figure 11) species contrasts sharply with the rainfall source of many other bryophytes. Although species occupying raised bogs with no ground water input may rely almost entirely on rainfall, those mosses in fen situations undoubtedly get nutrients from the ground water as well. In a study of 21 species of ***Sphagnum*** (Figure 21-Figure 29) in Poland, this genus demonstrated its ability to accumulate N, P, and K in the upper parts of the plant through active uptake, whereas Ca, Mg, and Na accumulated through passive **cation exchange** (Wojtun 1994; see below), suggesting an arrangement of nutrients within the plant similar to that of the tracheophytes.

Figure 11. Rich fen showing marl deposits (Ca^{++}) on plants. Photo by Janice Glime.

A number of researchers have concluded that **cryptogamic crusts** (soil crusts of algae, Cyanobacteria, fungi, lichens, and bryophytes) that live on the soil in areas with low rainfall increase the availability of essential elements, such as N, Cu, K, Mg, and Zn, thus benefitting seeds, seedlings, and mature tracheophyte plants (Harper & Pendleton 1993; Belnap & Harper 1995; Harper & Belnap 2001). This is most likely due to a combination of trapping airborne nutrients and preventing loss due to erosion and leaching from the soil. We are only beginning to understand the extent and role of bryophytes in nutrient trapping, sequestration, and release in various habitats.

Cation Exchange

Once we understand external transport, we must examine how the nutrients actually enter the moss. Are all nutrients equally capable of entry? Most likely not, but how is that controlled? And can these bryophyte leaves function as well as roots of tracheophytes in the absorption of nutrients?

Brown and Buck (1985) considered the **cation exchange capacities** (**CEC**; see below) of bryophyte cell walls to be important in their uptake and sequestering ability. Potassium (K) can be held on exchange sites, then remain in solution once it enters the plant. These researchers warned that it was important to know the locations of minerals within and on the bryophytes because ions such as those of Ca and Pb can remain on exterior exchange sites whereas Mg and Zn can be both internal and external.

Dainty and Richter (1993) identified two classes of weak-acid binding sites. One had a low *p*K (2-4) and the other a high one (>5). ***p*K** is the *p*H at which equal concentrations of acidic and basic forms of a substance are present; it is the negative $\log_{10}$ of the dissociation constant of the electrolyte. The binding sites are related to the uronic, amino, and phenolic acid contents of the cell walls. Dainty and Richter concluded that "valence-dependent reductions in cation activities in the wall phase are an important contributor to the differences in the *p*K estimates."

The ability of bryophytes to take up nutrients from weak solutions (Babb & Whitfield 1977) permits them to grow in situations that may be limiting to tracheophytes. We know that many (perhaps all) bryophytes sequester nutrients on exchange sites (Clymo 1964; Craigie & Maass 1966; Wells & Brown 1990; Bates 1997), but that the exchange capacity varies among species (Büscher *et al.* 1983).

Polyuronic Acids and CEC

In bryophytes, **cation exchange** is the process in which positively charged ions in the environment are able to replace H^+ ions at the surface of the cell walls, particularly those of leaves. **Cation exchange capacity** (CEC) is due to high concentrations of non-esterified **pectates**, mostly **polyuronic acids**, within the cell walls (Clymo, 1963; Craigie & Maass, 1966) and seems to be the first step in uptake of nutrient cations (Koedam & Büscher 1983). Fine roots of tracheophytes use this method as the first step in obtaining cationic nutrients from their surroundings. Koedam and Büscher (1983) demonstrated that CEC in mosses, typically much higher than in tracheophyte roots (Table 1; Knight *et al.* 1961), was related to soil preference and carbonate content of the bryophytes.

Table 1. Mean cation exchange capacity of cell walls of tracheophyte roots compared to that of bryophyte gametophores. Tracheophytes from Klein & Horst 2005; bryophytes from Bates 1982b.

	μg g^{-1} dry mass	
Calcicolous bryophytes		
Ctenidium molluscum	15,510	
Tortella tortuosa	15,160	
Schistidium apocarpum	12,940	
Homalothecium sericeum	12,460	
Orthotrichum cupulatum	12,250	
Syntrichia ruralis	10,160	
Calcifugous bryophytes		
Ptychomitrium polyphyllum	6,690	
Racomitrium fasciculare	3,330	
Dicranoweisia cirrata	3,200	
Andreaea rothii	2,660	
Grimmia donniana	2,610	
Racomitrium lanuginosum	2,330	
Tracheophytes	**0-5 mm**	**5-20 mm**
field bean	491.0	543.7
yellow lupine	422.0	527.4
barley	106.8	59.1
rye	63.1	65.5

The **uronic acids** are important in creating cation exchange sites. Popper and Fry (2003) have demonstrated that bryophytes (including hornworts, thalloid and leafy liverworts, and basal mosses) have higher concentrations of **glucuronic acid** in their primary cell walls than any of the other land plants. Basal mosses have higher concentrations than more advanced mosses, and the highest occurs in ***Sphagnum*** (Figure 21-Figure 29). ***Anthoceros*** (Figure 102-Figure 103) was unique in having a repeat-unit of glucuronic acid-α(1→3)-galactose, a substance nearly lacking in other kinds of plants in the study. **Galacturonic acid** is known as a subunit in some **xyloglucans**, a group of hemicellulose cell wall compounds (Peña *et al.* 2012). In particular, Peña *et al.* (2008) found that mosses and liverworts have **xyloglucans** that contain galacturonic acid, making them distinctly different from those xyloglucans demonstrated in both hornworts and tracheophytes. Popper and Fry (2003) considered that the cell wall xyloglucans may have been pre-adaptive substances that permitted early colonization of land, permitting rapid acquisition of nutrients during periods of short-lived surface water availability.

The role of cation exchange in nutrient uptake in poor nutrient habitats is further supported by the greater ability of ***Sphagnum*** (Figure 21-Figure 29) to exchange Ca^{++} and Mg^{++} ions for H^+ ions, providing them with a mechanism to obtain the very limited nutrients in their habitats. For example, Temple *et al.* (1981) reported the exchange capacity of ***Sphagnum*** to range 0.9 to 1.5 meq per gram dry biomass, whereas that of other mosses generally ranges 0.6-1.1. Figures in meq on tracheophytes were hard to

find; I was able to find that wheat (*Triticum vulgare*) has a low CEC of 0.02 meq per gram dry biomass of roots, with the highest in that study of 0.2 meq in cress (*Lepidium sativum*) (Wiersum & Bakuma 1959).

On the other hand, if the Ca^{++} content of the habitat is too high, ***Sphagnum*** will bind so much Ca^{++} to its leaf surfaces that it will eventually kill the moss (personal observation). Although this cation exchange process is beneficial in obtaining nutrients, it can also result in accumulation of high levels of heavy metal pollutants (Brown 1984) such as Cd because the moss lacks sufficient selectivity in either binding or uptake of these non-nutrients (Brown & Bates 1990).

The Mechanism

As early as 1961, Knight *et al*. found a correlation between **uronic acid** contents and cation exchange capacity. ***Sphagnum*** (Figure 21-Figure 29), in particular, has extensive binding sites through its use of the **polyuronic acid** known as **galacturonic acid** (Clymo 1963). Through this capability, ***Sphagnum*** is able to outcompete tracheophytes. By creating an "intense nutrient impoverishment" for other plants, ***Sphagnum*** gains a competitive edge (Van Breemen 1995). It can impede growth of peatland shrubs such as leatherleaf (*Chamaedaphne calyculata*; Figure 12) (Bartsch 1994) by sequestering nutrients the shrubs need for growth.

Figure 12. ***Chamaedaphne calyculata***, a species that must compete with ***Sphagnum*** for nutrients. Photo by Uleli, through Creative Commons.

Polyuronic acids such as galacturonic acid have a **carboxyl group** ($COOH^+$) protruding on the outer surface of the cell wall. This carboxyl group freely exchanges its H^+ for other cations in its surroundings (Figure 13). Hence, when cations such as K^+, Mg^{++}, and Ca^{++} filter through the bryophyte layer, these ions are often bound on these bryophyte cell wall exchange sites.

Seemingly all bryophytes have a large number of exposed exchange sites, compared to those even of roots of tracheophytes (Knight *et al*. 1961). These exchange sites are essential to the uptake of nutrients in non-***Sphagnum*** bryophyte taxa as well. For example, ***Pseudoscleropodium purum*** (Figure 14) ceased absorbing Mg^{++} and lost intracellular Mg when the exchange sites were saturated with $CaCl_2$, suggesting adherence to exchange sites may be a necessary prerequisite to Mg^{++} uptake (Bates 1989). Addition of both K^+ and Ca^{++} greatly increased their concentrations in the exchangeable fraction of the cell but significantly reduced the concentration of Mg^{++}. Malmer *et al*. (1992) found that the concentrations of Mg^{++} and Ca^{++} in Canadian mire species [three ***Sphagnum*** species and ***Tomentypnum nitens*** (Figure 15), all from hummocks] correlated with the surface water concentrations. It is interesting that when Ca^{++} is increased, the brown mosses are more competitive than are ***Sphagnum*** species. And there is evidence that brown mosses as well as ***Sphagnum*** can lower the *p*H, but that they typically do it at a higher level of *p*H (Figure 16) (Glime *et al*. 1982).

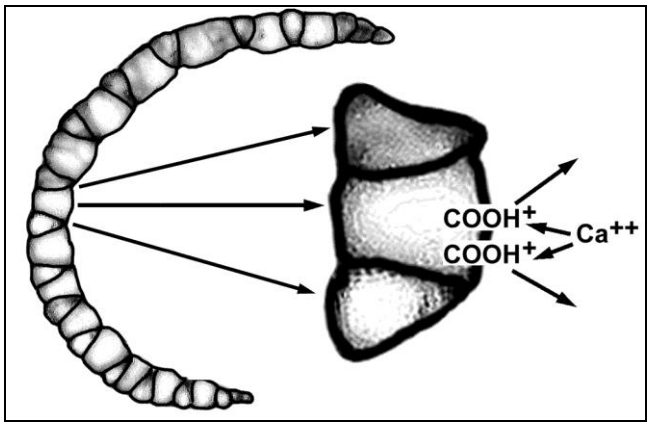


Figure 13. View of leaf cross section of ***Sphagnum*** (**left**) with two enlarged chlorophyllous cells and hyaline cell on **right**. Enlargement shows **carboxyl groups** ($COOH^+$) of the **polyuronic acid** and one Ca^{++} that will exchange for two H^+ ions in cation exchange. Drawing by Janice Glime.

Figure 14. ***Pseudoscleropodium purum***. Photo by Michael Lüth, with permission.

Figure 15. ***Tomentypnum nitens***, a species in which the Mg^{++} and Ca^{++} correlate with surface water concentrations. Photo by Michael Lüth, with permission.

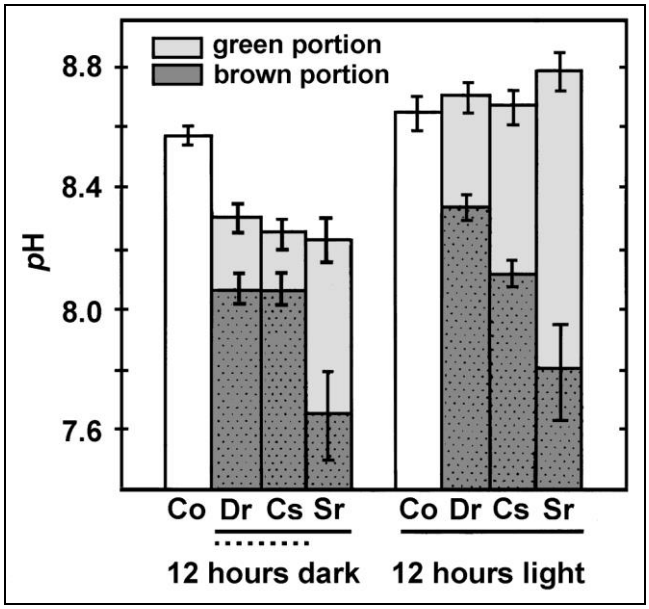


Figure 16. Comparison of *p*H-lowering ability of three mosses from an alkaline fen, Lawrence Lake, Barry County, Michigan, USA, following 48-hour incubation. Co = control lake water, Dr = ***Drepanocladus revolvens*** (=***Scorpidium revolvens***; Figure 17), Cs = ***Campylium stellatum*** (Figure 18), Sr = ***Sphagnum russowii*** (Figure 41). 12 hours dark and light indicate last cycle completed. Vertical lines represent 95% confidence intervals. Horizontal lines indicate no significant differences among green (active) (——) and brown (senescent) (• • •) moss species (distribution-free multiple comparisons test, $\alpha = 0.05$; n = 10). Starting *p*H = 8.25. From Glime *et al*. 1982.

Figure 17. ***Drepanocladus revolvens***, an alkaline fen moss that lowers the *p*H of its medium. Photo by Michael Lüth, with permission.

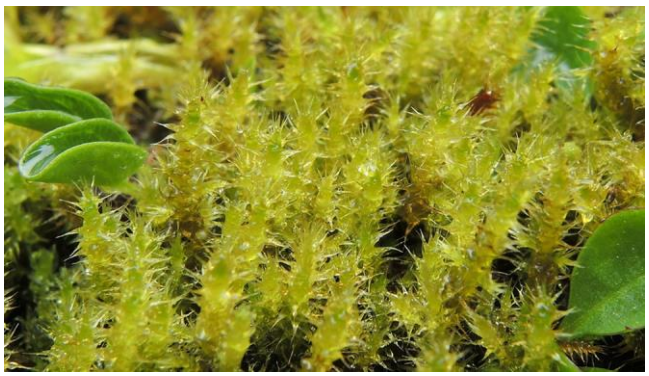


Figure 18. ***Campylium stellatum***, alkaline fen moss that lowers *p*H of surroundings. Photo by Michael Lüth, with permission.

Cation Competition

So how does competition between cations happen? Divalent cations such as Ca^{++} and Mg^{++} require two binding sites. When there are many of these ions in the environment, they compete for binding sites, occupying two of them in paired sites. Other ions that require two sites then have more difficulty finding the pair of sites they need to bind.

Because plants have a finite number of exchange sites, ions must compete with each other for those locations. Thus, if one cation is in excess, it can cause cellular deficiency of other cations that are unable to gain access to these exchange sites. Based on their experiments with ***Hylocomium splendens*** (Figure 19) and ***Sphagnum***, using artificial precipitation, Gjengedal and Steinnes (1990) considered that cations such as Na^{+} and Mg^{++} in the precipitation may occupy exchange sites and affect the uptake of other ions by this competition. They found that uptake of Zn and Cd were *p*H dependent and that increasing temperatures increased the uptake for all four of the metals tested (Ca, Cu, Pb, Zn).

Figure 19. ***Hylocomium splendens***, a species in which cations such as Na^{+} and Mg^{++} in the precipitation may occupy exchange sites and affect uptake of nutrient cations. Photo by Michael Lüth, with permission.

Figure 20. ***Calliergonella cuspidata***, a species whose growth is inhibited at high Ca concentrations. Photo by Michael Lüth, with permission.

Complexing reactions with anions such as Cl^{-} may also interfere with uptake. When Bates and Farmer (1990) applied $CaCl_2$ to three bryophytes, their responses varied by habitat. ***Pseudoscleropodium purum*** (Figure 14) and

Calliergonella cuspidatum (Figure 20) from chalk soil exhibited significantly reduced growth at high Ca concentrations (5 mol $CaCl_2$ m^{-3}), whereas ***P. purum*** and ***Pleurozium schreberi*** (Figure 2) from acidic clay were unaffected by the additions. The mosses from the chalk soil had lower initial tissue levels of K and Mg, suggesting that the additional $CaCl_2$ caused deficiencies in these nutrients through exchange site competition.

Ions in the external solution will first establish equilibrium with the exchange sites (Brown 1982). This physical process is completed very rapidly in the lab, but may require days in the field (Brown & Bates 1990). Once that is established, the remaining ions are available for uptake to the interior of cells (Pickering & Puia 1969). Hence, high concentrations of minerals will ultimately increase the uptake.

The number of exchange sites seems to be adaptive, at least in ***Sphagnum***. ***Sphagnum*** section ***Acutifolia*** (Figure 21), which inhabits drier locations, has more exchange sites per unit of biomass than do members of section ***Cuspidatum*** (Figure 22), which are wet hollow species (Brown 1982). Both Clymo (1963) and Spearing (1972) showed that the number of exchange sites correlated positively with height above water of the optimum habitat for ***Sphagnum*** species. This permits hummock species to hold nutrients on their cell surfaces until they are needed without having to wait for rainfall to provide a new source.

Figure 21. ***Sphagnum fuscum*** (Section **Acutifolia**) hummock, a ***Sphagnum*** species with a high number of cation exchange sites. Photo by Jutta Kapfer, with permission.

Figure 22. ***Sphagnum cuspidatum*** (Section **Cuspidatum**), a wet hollow species with a relatively low number of cation exchange sites. Photo by Jutta Kapfer, with permission.

Monovalent ions have little effect on CEC for divalent ions (Brehm 1968). But CEC of monovalent cations drops to 0.025 - 0.14 times capacity when in company of divalent cations, presumably due to double binding of divalent ions, much like doubling the strength of a magnet.

Brehm found that dead and living material have the same CEC on a dry weight basis. Nevertheless, living ***Sphagnum*** (Figure 21-Figure 29) cells contain most of the K^+ and Na^+, Ca^{++} is mostly on the external exchange sites, and Mg^{++} is on both locations. On the other hand, branches and stems of ***Sphagnum*** have very different CEC. The living ***Sphagnum*** is able to maintain a relatively constant cellular content of cations, even when the concentrations of the medium varies widely.

The ability of an exchange site to hold a given positively charged ion depends not only on the valence (charge) of the ion, but also on concentration. When there is a flood of H^+ ions, these will replace the other, more rare and higher mass cations. Again, this is like a magnet; it is harder for a magnet to hold something heavy than something light (like H^+). Hence, basic cations from the bryophyte surface are released into the soil (Foth & Ellis 1997). A striking example of this phenomenon is the case of acid rain making a ***Sphagnum*** (Figure 21-Figure 29) peatland alkaline and causing the ***Sphagnum*** to die! (Kilham 1982). The acid rain caused the release of alkaline positive ions from the surrounding hillside, which ultimately washed into the peatland. Although ***Sphagnum*** is equipped to bind such ions and make its surroundings more acid, it was not equipped to handle the large concentration that resulted from the uphill release. Instead, cations such as Mg^{++} and Ca^{++} accumulated on the surface of ***Sphagnum*** and eventually killed it. In forested ecosystems, cations released from soil exchange sites become available to roots, may be leached from the organic layer into deeper layers, or may be lost through runoff.

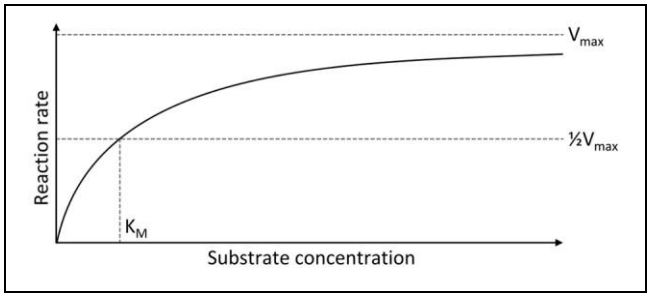
Heavy Metal Relationships

Several bits of information suggest that heavy metals like cadmium use cation exchange of low valence ions to aid their uptake. For example, in the thallose liverwort ***Dumortiera hirsuta*** (Figure 23), pretreatment with 80 mM KNO_3 causes higher Cd^{++} uptake, suggesting that potassium is able to strip exchange sites of competing cations, allowing higher valence cadmium to then strip some of those sites and enter cells (Mautsoe & Beckett 1996).

Figure 23. ***Dumortiera hirsuta***, a liverwort that uses cation exchange to take up heavy metals. Photo by Li Zhang, with permission.

The heavy metals, in turn, influence uptake of potassium and magnesium – two essential nutrients (Carballeira *et al*. 1999). The researchers suggested that loss of K, a very soluble nutrient, from the cells might be due to the effect of the heavy metals on the cell membranes, changing their permeability. The cation Mg^{++} was most likely displaced from the cation binding sites by the heavy metals.

In one experiment, three aquatic bryophytes were exposed for 60 minutes to solutions of 0, 1, 10, 50, 100, and 200 ppm each of Cd, Co, Cu, Ni, Pb, and Zn (Carballeira *et al*. 1999). Locations of these metals plus K and Mg were determined. Most metals remained on extracellular locations, rather than intracellular. On the other hand, only negligible amounts appeared in the particulate fraction. The relationship between water concentration and extracellular concentration could be modelled with a **Michaelis-Menten** equation:



$$v = \frac{d[P]}{dt} = \frac{V_{max}[S]}{K_M + [S]}$$

v = reaction rate
[S] = concentration of substrate S
P = product
t = time
V_{max} = max rate achieved at saturating substrate concentration
K_M = substrate concentration at which reaction rate is half of V_{max}
d [P]/dt = change in product per change in time

But the three aquatic species in this study by Carballeira *et al*. (1999) differed markedly. In ***Scapania undulata*** (Figure 24-Figure 25), the extracellular cation-binding sites demonstrated high metal affinity. On the other hand, ***Fissidens polyphyllus*** (Figure 26) has relatively low affinity. Nevertheless, ***F. polyphyllus*** had the highest internal concentrations of these metals at the end of the experiment. The uptake priorities were the same for all three species.

In these aquatic bryophytes, ***Fontinalis antipyretica*** (Figure 27), ***Scapania undulata*** (Figure 24-Figure 25), and ***Fissidens polyphyllus*** (Figure 26), the extracellular compartment held more metals than did the intracellular compartment (Vázquez Castro *et al*. 1999). The extracellular cation-binding sites of ***S. undulata*** had a high metal affinity, whereas it was relatively low in ***F. polyphyllus***. On the other hand, ***F. polyphyllus*** after the incubation in the metal solutions had the highest intracellular metal contents. All three species had the same ranking of metal uptake.

Uptake of heavy metals in these aquatic bryophytes led to considerable losses of intracellular K (probably due to effects on plasma membrane properties) (Carballeira *et al*. 1999). Similarly, Mg^{++} cellular contents decreased, but it was apparently due to competition by the metals on the binding sites, limiting uptake. Species differences were again interesting. ***Scapania undulata*** (Figure 24-Figure 25) exhibited the highest losses of K from internal cell sites, followed by ***Fontinalis antipyretica*** (Figure 27). On the other hand, ***S. undulata*** had the lowest losses of Mg from its extracellular exchange sites. These experiments help to explain competition among nutrients and locations in the short term, but long-term effects could be different, as seen in Chapter 8-3 on nitrogen.

Figure 24. ***Scapania undulata*** in its stream edge habitat. Photo by Michael Lüth, with permission.

Figure 25. ***Scapania undulata***, a species with high metal affinity on its cation exchange sites. Photo by Hermann Schachner, through Creative Commons.

Figure 26. ***Fissidens polyphyllus***, a species with low affinity for heavy metals. Photo by Janice Glime.

Figure 27. ***Fontinalis antipyretica***, a species that loses potassium and magnesium when exposed to heavy metals. Photo by Bernd Haynold, through Wikimedia Commons.

Much of what we know about uptake of minerals into plants comes from studies on these heavy metal pollutants. Cadmium, a common pollutant in areas with agricultural fertilizers and other human uses, moves from extracellular sites of the bryophytes to intracellular sites. In ***Rhytidiadelphus squarrosus*** (Figure 28) cadmium altered photosynthetic rates (Wells & Brown 1987). Its activity at the **plasmalemma** (cell membrane) may exercise control over other ions, affecting their accumulation within the cell, and *vice versa*.

Figure 28. ***Rhytidiadelphus squarrosus***, a species that has an altered photosynthetic rate in the presence of cadmium. Photo by Michael Lüth, with permission.

Differing Affinities

Breuer and Melzer (1990a) contributed to the explanation of ion competition using ***Sphagnum*** (Figure 21-Figure 29) from a high moor. They found that when two or more ions are present, there is an order to the binding success: $Pb^{++}>Cd^{++}\geq Ca^{++}>Mg^{++}>K^{+}>Na^{+}\geq NH_4^{+}$. Hence, those with higher binding affinities were able to suppress the binding of the lower affinity ions.

Breuer and Melzer (1990b) commented that ***Sphagnum*** (Figure 21-Figure 29) "shows behaviour of a relatively ideal ion exchanger." And, while species differ in their capacity, the coefficients of selectivity are independent of species. These bound cations can readily be displaced if another cation is present at a higher concentration, has a larger hydrated atomic radius, or has a higher valency (Bates 2000).

In ***Sphagnum*** (Figure 21-Figure 29) Hájek and Adamec (2009) found the exchangeable cation content decreased in the order of $Ca^{++} \geq K^{+}$, Na^{+}, $Mg^{++} > Al^{+++} > NH_4^{+}$, whereas the intracellular element content demonstrated the order of N > K > Na, Mg, P, Ca, Al. While Ca occurred primarily on exchange sites, Mg, Na, and especially K, Al, and N occurred inside the cells. Vertical position in the bog influenced the nutrient uptake and location. Hummock species have a higher cation exchange capacity (CEC) and accumulate more exchangeable Ca^{++}. By contrast, the hollow species have a lower CEC and accumulate more exchangeable Na^{+}, especially among the lower dead shoot segments. Intracellular N and P were consistently lower in the dead portions, indicating their translocation to growing upper portions. ***Sphagnum magellanicum*** (Figure 29) has about 40% lower N content in its cells compared to other species, suggesting its inability to compete for N. This can cause it to lose competition to other species (Hájek & Adamec 2009), but its drought tolerance aids it in occupying tops of hummocks (Li *et al*. 1992). This leaves us wondering why it has such a low N content.

Figure 29. ***Sphagnum magellanicum***, a species with low N content. Photo by Michael Lüth, with permission.

Rühling and Tyler (1970) demonstrated the order of binding affinity of several heavy metal cations using ***Hylocomium splendens*** (Figure 19): Cu ≅ Pb > Ni > Co > Zn ≅ Mn. In a two-hour experiment, these heavier cations preferentially bound to the exchange sites even when lighter cations of Ca^{++}, K^{+}, Mg^{++}, and Na^{+} were present in high concentrations. However, this sequence changed when the elements were supplied individually. Other researchers also demonstrated differences from this sequence in ***Rhytidiadelphus squarrosus*** (Figure 28) (Brown & Beckett 1985), ***Brachythecium rutabulum*** (Figure 7) (Brown & Buck 1978), and ***Pohlia nutans*** (Figure 30) (Webster 1985). These differences may relate to damage by some of the elements, such as cell membrane damage by mercury in some of the experiments,

concentration differences, and possible contamination from soil particles (Brown & Wells 1990).

Figure 30. ***Pohlia nutans***, a species of exposed, low-nutrient habitats. Photo by Michael Lüth, with permission.

In the moss ***Rhytidiadelphus squarrosus*** (Figure 28), competition occurs in the order of Cd ≅ H > Ca > Mg ≫ K on the extracellular sites (Wells & Brown 1990), suggesting that the low weight of H compensates for the higher valency of Cd. However, affinity of the intracellular Cd transport site occurred in the order of Ca > Cd > Mg ≫ K. Thus, internally calcium was a competitor for cadmium, but magnesium was not. Cadmium experienced maximum uptake at *p*H 5.6 and was very sensitive to *p*H. Potassium had no competitive ability against these ions. This presents another interesting question. How do internal differences in *p*H affect uptake, transport, and storage of ions? And what are the extent and locations of those *p*H differences?

Nieboer and Richardson (1980) found a divalent metal ion selectivity binding order of Pb > Cu > Cd > Co ≅ Fe > Ni > Zn > Mn, although Rühling and Tyler (1970) found a slightly different order for ***Hylocomium splendens*** (Figure 19): Cu ≅ Pb > Ni > Co > Zn ≅ Mn, an order that seems to be widespread in bryophytes (Bates 2000). However, once the sites are nearly fully occupied, this preferential binding is no longer the strongest force, possibly accounting for differences illustrated here. Isolated binding sites are only able to bind one position on the cation, hence eliminating the advantage for higher valency ions. In fact, at this stage, the isolated sites are more likely to bind univalent ions than divalent ones and more likely to bind divalent ones than trivalent ones (Richter & Dainty 1990). This is because divalent and trivalent ions require adjacent binding sites, whereas monovalent ions can utilize isolated sites. It is also likely that in systems with lower *p*H, more sites are occupied by H^+ ions, creating more isolated sites. This would favor the binding of lower valency ions such as K^+ and account for the high selectivity at a low *p*H.

Habitat Differences

Cation exchange sites can serve two conflicting purposes: bind the cations against further uptake, or concentrate them for absorption sites (Büscher *et al*. 1990). These roles have rarely been discussed for terrestrial bryophytes. This affects the bryophyte tolerance of various substrates. Using the **acidicline** (preferring soils with *p*H <5) species ***Atrichum undulatum*** (Figure 31), ***Leucobryum glaucum*** (Figure 32, ***Mnium hornum*** (Figure 33), and ***Polytrichum formosum*** (Figure 34, and the **neutrocline** (preferring *p*H close to neutral, *i.e.* >5) species ***Homalothecium sericeum*** (Figure 35) and ***Plagiomnium undulatum*** (Figure 36), Büscher *et al*. found that a**cidophilous** and **acidicline** taxa generally have lower CEC and are more able to tolerate the toxic aluminium (Al) levels, but not high levels of Ca. **Neutrocline** taxa instead avoid habitats with aluminium in the substrate and thrive on high calcium levels. (But what is the mechanism causing the avoidance?) They concluded that cation exchange properties do not protect mosses against potentially toxic ions, including aluminium, by sequestering them. But they did conclude that the exchange sites could increase the availability of cations. High CEC favored fixation of Al ions over Ca ions, indication that a low CEC is needed for taxa to tolerate acid soils.

Figure 31. ***Atrichum undulatum***, an **acidicline** species that has relatively low cation exchange capacity. Photo by Michael Lüth, with permission.

Figure 32. ***Leucobryum glaucum***, an **acidiphile** with lower numbers of cation exchange sites. Photo by James K Lindsey, with permission.

Figure 33. ***Mnium hornum***, an **acidiphile** with lower numbers of cation exchange sites. Photo by Tim Waters, through Creative Commons.

Figure 34. ***Polytrichum formosum***, an **acidiphile** with lower numbers of cation exchange sites. Photo by Michael Lüth, with permission.

Figure 35. ***Homalothecium sericeum***, a **neutricline** species with higher numbers of cation exchange sites. Photo by Michael Lüth, with permission.

Bates (1992) found that in epilithic and woodland soils the cation exchange capacity decreases with decreasing Ca content, and likewise with decreasing *p*H of the substrate. This change might help to protect the bryophytes against the toxic aluminium that increases in concentration in acidic solutions, *e.g.* soils polluted by acid rain.

Figure 36. ***Plagiomnium undulatum***, a **neutricline** species with higher numbers of cation exchange sites. Photo by Michael Lüth, with permission.

Calcareous rocks, inhabited by **calcicoles** (Ca-preferring species), typically have Ca^{++} concentrations 16-17 times that found in species from non-calcareous rocks (**calcifuges –** species avoiding Ca) (Bates 1982a). The calcicoles exhibit 3-4 times as many cation exchange sites as the calcifuges. Bates suggested that the calcicole mosses may require greater Ca^{++} concentrations to maintain cell membrane integrity.

Uptake Rate

Uptake of these nutrients is very rapid when concentrations are high. Half the maximum extracellular uptake can be achieved in 4.45±1.03 minutes in 100 µmol L^{-1} Cd (Brown & Beckett 1985). This rate is concentration dependent and at lower (more natural) concentration levels it can take several days to reach equilibrium (Mouvet 1987). Release of the cations from the exchange sites when the element is removed from the medium takes even longer, as shown in the aquatic liverwort ***Chiloscyphus polyanthos*** (Figure 37-Figure 38) (Maurel-Kermarrec *et al*. 1985). The uptake ability varies between clones that grow within meters of each other (Wells & Brown 1987; Wells 1988). This can result from differences in light/moisture availability in the open vs under shrubs, as demonstrated in ***Rhytidiadelphus squarrosus*** (Figure 28) (Wells & Brown 1987). This difference could have been caused by thicker cell walls in the higher light population of ***R. squarrosus***.

Figure 37. ***Chiloscyphus polyanthos*** in a typical habitat. Photo from <www.aphotofauna.com>, with permission.

Figure 38. ***Chiloscyphus polyanthos***, a leafy liverwort with cation exchange. Photo by Michael Lüth, with permission.

In summary, nutrient uptake into the moss is initially dependent on available exchange sites, but then it depends on affinity of a particular nutrient for appropriate transport sites of cell membranes, presence of competing elements, and turnover rate of the uptake site (Brown & Bates 1990), and perhaps cell wall thickness (Wells & Brown 1987).

Desiccation and Loss

Brown and Brumelis (1996) found that desiccation and duration of drought affected cellular location of elements in ***Hylocomium splendens*** (Figure 19), a boreal forest floor species. Rehydration partially reversed these effects.

When bryophytes become desiccated, nutrients leave the cells through leaky membranes (Bewley 1979). But Bates (1997) has shown that in ***Brachythecium rutabulum*** (Figure 7) and ***Pseudoscleropodium purum*** (Figure 14), leaked K^+ ions are able to remain on leaf surfaces (Figure 14), held there on exchange sites, and are re-absorbed upon hydration. Like tracheophyte roots, bryophytes utilize cation exchange sites to hold nutrients at their surfaces until those nutrients are moved into the plant.

Anion Uptake

Bryophytes also have exchange sites for **anions** (negatively charged ions), but these are far less abundant and likewise their role is less well understood (Clymo 1963). Even now, little is known about anion uptake. Wells and Richardson (1985) found that only living shoots of ***Hylocomium splendens*** (Figure 19) were able to accumulate arsenate and selenite, both **anions**. Arsenate uptake is inhibited by phosphate (anion) competition when both are supplied at the same time. On the other hand, if plants were incubated in phosphate before providing arsenate and selenite, it had no effect on their uptake. It appears that arsenate and selenite are accumulated by separate transport systems in this species and that these systems may be the ones responsible for phosphate uptake. *p*H was important, with arsenate uptake optima occurring between 3 and 5, whereas selenate was optimal at *p*H 3.

Polytrichum commune (Figure 4) has a well-developed conducting system and was the only bryophyte one study that had more uptake in brown portions than in green ones (Chapin *et al*. 1987). ***Sphagnum*** species were the only ones with significant P uptake in the current growth. But in seeming contradiction, the anionic form of N (nitrate) was preferred by ***Sphagnum*** over the cationic ammonium source of N (Rudolph *et al*. 1982). This preference likewise contradicts the results of Wanek and Pörtl (2008) who concluded that ***Sphagnum*** (Figure 21-Figure 29) prefers ammonium because of its numerous cation exchange sites. But it does coincide with the inhibition of nitrogen reductase by ammonium (Syrett & Morris 1963; Orebamjo & Stewart 1975). Furthermore, Wanek and Pörtl (2008) found that amino acids contributed a significant fraction of the N used by ***Sphagnum*** from the lowland rainforest in Puerto Rico

Brown (1982) suggested that anion adsorption is probably especially low in mosses because they have low iron and aluminium content and high cation exchange capacity Clymo 1963; Chapin *et al*. 1987). Phosphorus, as the phosphate anion, is taken up primarily from the mineral substrate (Bates 1992). Chapin *et al*. (1987) concluded that accumulation of the phosphate anion, as they observed in mosses of the Alaskan black spruce (*Picea mariana*, Figure 3) forest, was therefore by active absorption similar to that of higher plants. In these forests, mosses hold 17% of the phosphorus pool, despite accounting for 75% of the annual P accumulation. The mosses have a greater ability to absorb phosphate than do fine roots of the black spruce.

Proton Pumps

After ions have reached the surface of the cell, they require energy to enter the cell. In tracheophytes, the **proton pump** is well known in such activities as bringing nutrients into root hairs, opening and closing guard cells, closure of the Venus flytrap, and growth, to name only a few. In bryophytes, the proton pump has likewise been demonstrated, and like that of tracheophytes, it uses ATP to "pump" H^+ ions out of a cell (Figure 39). This leaves the cell with a negative charge that attracts cations into the cell (Raven *et al*. 1998). The resulting negative charge provides the force needed to bring in K^+, NH_4^+, Mg^{++}, Ca^{++}, sugars, and amino acids, and probably other cations that have not yet been confirmed experimentally.

Cotransport

As a positively charged ion enters the cell, it typically brings along an associated anion by **cotransport**. The pump, at the same time, regulates the *p*H within the cell to about 7.3-7.6. In bryophytes, the leaf cell surface and interstitial spaces between the cells provide sites where adhering cations are able to enter the cell through the proton pump mechanism.

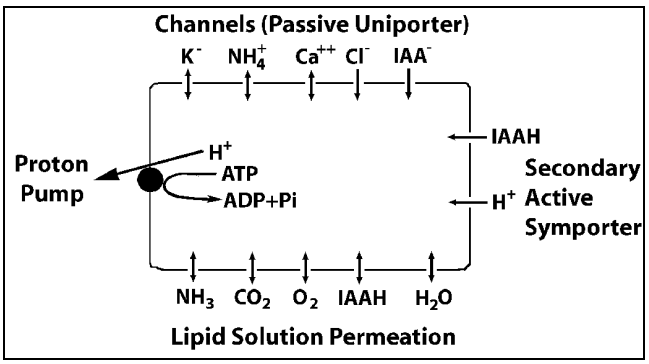


Figure 39. Known transport processes through the plasmalemma of a bryophyte cell. Diagram based on Raven *et al*. 1998.

As a result of ion movement through CEC and the proton pump, the bryophyte most likely has an influence on the **rhizoidosphere** (soil space immediately surrounding rhizoids) similar to that of tracheophytes on the rhizosphere (Raven *et al* 1998), although in the case of bryophytes, leaves may contribute to the alteration of conditions even more than the rhizoids. The rhizoidosphere is acidified in the process of cation exchange and proton pumping to bring nutrient cations into the cells, creating positive charges within the cells and accumulating organic anions in the cell vacuoles.

Pinocytosis

Pinocytosis results when a cell ingests a liquid by budding small vesicles inward from the cell membrane, thus containing the droplet. The droplet of liquid then is incorporated into the cell cytoplasm. Gullvåg *et al*. (1974) observed this mechanism in ***Rhytidiadelphus squarrosus*** (Figure 28) when it was treated with lead-rich particles in the lab. When they tested mosses that were exposed to lead pollution in the field, they found that the lead was bound within the nucleus. The importance of pinocytosis for incorporating nutrients into the cells of bryophytes seems to lack study.

Nanoparticles

The concept of nanoparticles is a relatively new idea in bryophyte ecology. Canivet *et al*. (2014) found, for the first time, that nanoparticles of iron in a mineral water suspension could penetrate the leaves of the moss ***Physcomitrella patens*** (Figure 40). In follow-up experiments Canivet *et al*. (2015) further demonstrated the penetration of iron nanoparticles into the moss ***Physcomitrella patens***. Using concentrations of 5 ng, 50 ng, 500 ng, 5 µg, and 50 µg per plant, they found no effect on ATP concentrations, reactive oxygen species, malondialdehyde, or glutathione, suggesting that the plants had not been physiologically harmed at any of these concentrations. The role of nanoparticles in providing essential nutrients or harming the plants seems to thus far lack exploration.

Figure 40. ***Physcomitrella patens***, a moss that can take up nanoparticles of iron. Photo by Michael Lüth, with permission.

Influence of Cellular Structures

Many studies have treated cellular influences as if the cells were homogeneous (Brown & Wells 1990). First, the nutrient must cross the cell membrane But in fact, once inside the cell, the nutrient may be held in solution, like K, or bound into amino acids and proteins, like some of the N and P. The chlorophyll molecule can take Mg out of play. When these binding compounds take the nutrient out of solution, they affect the concentration gradient from outside to inside the cell, affecting the concentration gradient used for the nutrient to cross the cell membrane and enter the cell. Others are bound to intracellular binding sites, again altering uptake rate.

Pickering and Puia (1969) described three phases of element uptake against time, an "unusual" process compared to that in algae and tracheophytes (Brown & Wells 1990). The first phase is the initial rapid uptake as the ions diffuse into interstitial spaces in the tissues (Pickering & Puia 1969). Then the uptake is controlled by equilibration with cell wall exchange sites. The final phase is a slow, linear increase of intracellular uptake. This third phase does not occur in dead material. As demonstrated in ***Rhytidiadelphus squarrosus*** (Figure 28), carriers can be used to transport the element across the membrane (Brown & Beckett 1985; Wells & Brown 1987). Specificity of these carriers determines how much inter-element competition there is for the intracellular uptake. This in turn affects the rate of uptake.

Location Is Important

As already noted, location of nutrients on and in the bryophytes is important (Brown and Buck (1985). The method used can present a bias that is misleading regarding normal nutrient concentrations. A nutrient adhering to the cell wall is not immediately available to the cells and may not be representative of the needs of the cells. Others may be held in the spaces within the walls. To fully understand the nutrient physiology, we must understand where these nutrients are located on and in the bryophyte plants (see Table 2).

Table 2. Element locations in bryophytes. Based on Brown 1982.

- in particles trapped by leaves
- in solution on exterior & in matrix of cell wall
- as ions bound to external exchange or chelating sites & on plasma membranes
- in solution in cytoplasm & vacuoles
- as insoluble substances in cytoplasm & vacuoles
- in leptome (especially Polytrichaceae)

New Growth

When new branches are formed and expand in the absence of additional nutrients, these nutrients must be obtained from existing tissues. In some cases, this is through **acropetal** (base to tip) transfer, as seen in ***Rhytidiadelphus squarrosus*** (Figure 28) (Wells 1988). Potassium, a very soluble nutrient, declines in lower portions as the apex grows. Calcium, on the other hand, is

not soluble and is taken from the initial apical segment, not transported from older tissues. Magnesium exhibited a somewhat similar response, but all segments lost Mg from intracellular sites as the apex grew.

Specificity

Some nutrients are taken up more easily than others. Leblond (2004) examined the uptake of heavy metals in the moss ***Pseudoscleropodium purum*** (Figure 14). The nutrient elements manganese and potassium had the highest retention. Non-nutrient ions of sodium, aluminium, and silica had the least retention. Youngest tissues accumulated the most nutrients, but internal redistribution occurred. Leblond found that soluble materials were taken in more easily than those deposited as particulates.

We know that cation exchange sites selectively bind higher valency cations (Richter & Dainty 1990). But at least in ***Sphagnum russowii*** (Figure 41-Figure 43) there are two classes of exchange sites. The well-known one is associated with polygalacturonic acids and accounts for more than 50% of the cation exchange capacity (Richter & Dainty 1989). In addition to that, **phenolic acids** account for about 25%, whereas **amino acid, sulfate ester,** and **silicate deposits** in the cell wall contribute to a lesser degree.

Figure 41. ***Sphagnum russowii***, a species with both polygalacturonic acid and phenolic acid exchange sites. Photo by Ralf Wagner <www.dr-ralf-wagner.de>, with permission.

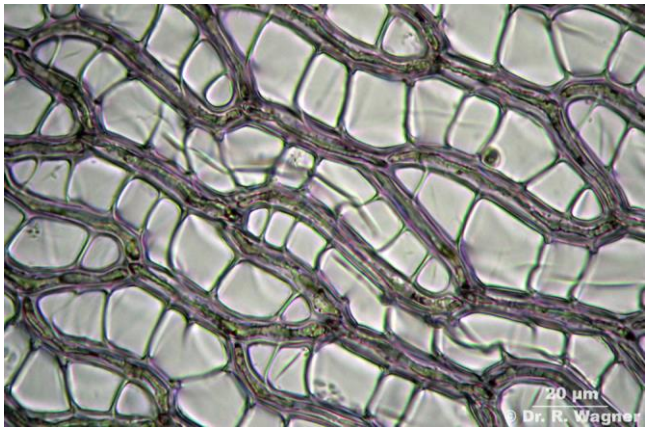


Figure 42. ***Sphagnum russowii*** leaf cells showing the exposed surface area of the hyaline cells (longer, wider cells with cross bars here). Photo by Ralf Wagner <www.dr-ralf-wagner.de>, with permission.

Figure 43. ***Sphagnum russowii*** leaf cells in cross section showing the exposed surface area of the much larger hyaline cells where cation exchange can occur on both inside and outside of the cell. Photo by Ralf Wagner <www.dr-ralf-wagner.de>, with permission.

When studying aquatic bryophytes, Burton and Peterson (1979) found that 33% of the cell-wall-bound Zn could be removed by the enzyme **pronase** (mix of enzymes that break down proteins), suggesting that a considerable portion of its binding might be due to protein binding. Richter and Dainty (1989) found a small number of binding sites that are more specific to small valency cations such as potassium. If these sites include phenolic compounds, one can presume that such binding sites might be widespread in bryophytes, wherein phenolic compounds are common (Mues & Zinmeister 1988; Liao 1993; Basile *et al*. 1999). Is this yet another use for these presumed "secondary" compounds? If so, what does it mean for cycling of potassium if it can be bound to the cell walls? Does this help the plant to retain its valuable potassium when cell membranes, damaged by desiccation, permit potassium to leak from the cell? Such a mechanism could contribute to the survival of bryophytes after desiccation and permit them to become a long-term sink for this and other ions.

We know that cation exchange is a somewhat selective process. Higher valency ions are bound preferentially over lower ones because they occupy more than one exchange site. Cations have binding preferences (Nieboer & Richardson 1980, 1981). **Class A** includes K, Ca, Mg, and S. These elements prefer oxygen-rich ligands, such as carboxylic groups. **Class B** elements such as Ag, Cu, H, Hg, Pb, and Au are toxic and prefer ligands that are rich in sulfur and nitrogen. The third group is a borderline class that includes Cu^{++}, Ni^{++}, Pb^{++}, and Zn^{++}. Hence, the quantity of any element bound to the cell wall will depend on concentration in the medium (precipitation, water, soil), its affinity for type of exchange site, and the total number of suitable exchange sites.

Dead cells may actually have more exchange sites than live ones due to shrinkage of cell membranes that cover them on the inside. On the other hand, Wells (1988) found that when the cells are killed by a strong acid, the exchange capacity decreases, a phenomenon he attributed to loss of cytoplasm.

Hence, the quantity of any element bound to the cell wall will depend on concentration in the medium (precipitation, water, soil), its affinity for type of exchange site, and the total number of suitable exchange sites. Dead cells may actually have more exchange sites than live ones due to shrinkage of cell membranes that cover them on the inside. On the other hand, Wells (1988) found that when

the cells are killed by a strong acid, the exchange capacity decreases, a phenomenon he attributed to loss of cytoplasm.

Fortunately, bryophytes seem to have uptake specificity for things they need over things they do not. For example, the thallose liverwort ***Dumortiera hirsuta*** (Figure 23) preferentially took up Ca, Mg, and Zn over Cd (Mautsoe & Beckett 1996). When KNO_3 was used to pretreat the plants, Cd uptake occurred, suggesting that the high concentration of K^+ removed the competing ions from the exchange sites and they were subsequently replaced by Cd. Light and increased temperatures also stimulated Cd uptake. Even ***Sphagnum*** (Figure 21-Figure 29, Figure 41-Figure 44), the champion of cation exchangers, distinguishes among ions in ways that do not seem to depend strictly on valence. It accumulates Al and Mn, but excludes Cu and Zn, accumulating much less of these than the concentrations in the surrounding fen water (Li & Glime 1990).

Shimwell and Laurie (1972) found that ectohydric and mixohydric mosses differ in their absorption, retention, and excretion of heavy metals. During droughts, **ectohydric** (having external conduction) mosses excrete such heavy metals as Zn and Pb, forming surface crusts containing up to 6% Pb and 1-5% Zn. In **mixohydric** (having both external and internal conduction) mosses, on the other hand, the metals generally are located at the base of the moss carpet in the older growth, suggesting their accumulation in older tissues and lack of internal transport.

Seasons

Since most bryophytes gain most of their nutrients from precipitation, we might assume that most nutrient uptake therefore occurs when it rains. Yet the relationship is most likely not so simple. Francez and Loiseau (1999) found that ***Sphagnum fallax*** (Figure 44) was more efficient at intercepting applied N (as NH_4NO_3) in August than in June, even though August had the lowest rainfall. Dust accumulation can benefit bryophytes that are able to absorb nutrients in early morning dew and even on humid nights when there is no benefit for tracheophytes.

Figure 44. ***Sphagnum fallax***, a species that takes up more N in August than in June. Photo by Michael Lüth, with permission.

Bates (1992) considered that in rapidly growing species such as ***Brachythecium rutabulum*** (Figure 7) the seasonal deposition of tracheophyte litter is especially important. In the slower-growing species such as ***Pseudoscleropodium purum*** (Figure 14), wet deposition may be the most important.

Turner and coworkers (2003) found that rates of acid phosphatase activity in moss apices differed markedly among species, but most taxa had the most activity in winter and least in summer. Nevertheless, tissues maintained relatively constant N and P concentrations throughout the year. A negative correlation between phosphatase activity and P concentration in the tissues suggests that the enzyme may become active in response to phosphorus needs and serves to indicate nutrient stress.

Núñez-Olivera *et al*. (2001) found that seasonal differences in several aquatic bryophytes [***Fontinalis antipyretica*** (Figure 27), ***F. squamosa*** (Figure 45, ***Jungermannia eucordifolia*** (Figure 46), and ***Pellia endiviifolia*** (Figure 47)] did not mimic the seasonal differences in their native streams. Rather, the concentrations depended on the interactions of internal and external factors. The elements that had the most persistent annual cycle were mostly essential nutrients: N, P, and Fe, plus the non-essential Na. The lowest concentrations occurred in spring and the highest in autumn. Concentrations were lowest during periods of growth.

Figure 45. ***Fontinalis squamosa*** in alpine water. Photo from <www.aphotofauna.com>, with permission.

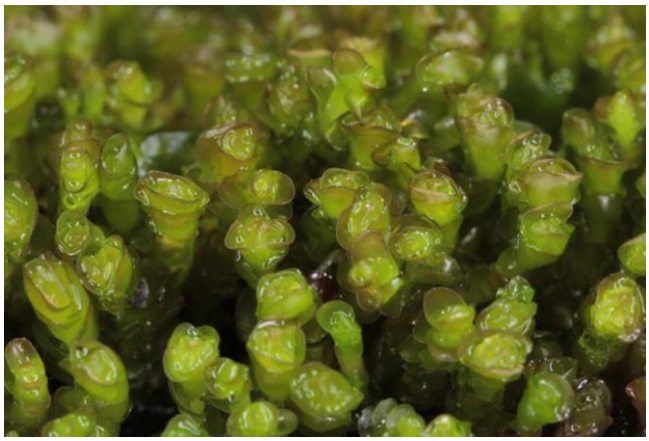


Figure 46. ***Jungermannia eucordifolia***, a species for which internal nutrient concentrations do not mimic those of its stream habitat. Photo by Jan-Peter Frahm, with permission.

Figure 47. ***Pellia endiviifolia***, a species for which internal nutrient concentrations do not mimic the seasonal changes of its habitat. Photo by Janice Glime.

Glucose Uptake

External glucose can enhance growth of at least some bryophytes (Jennings 1918). ***Ceratodon purpureus*** (Figure 48) grew 4-5 times as much when provided with glucose on nutrient agar compared to nutrient agar without glucose. This implies that organic sources of carbon that may be available in the substrate are suitable carbon sources for at least some mosses. Vujičić *et al*. (2009) found that the best conditions for axenic culture of the moss ***Dicranum scoparium*** (Figure 49) was in MS medium enriched with sucrose at 1.5% at 18-20°C.

Figure 48. ***Ceratodon purpureus***, a species that grows faster when external glucose is supplied. Photo by Michael Lüth, with permission.

Bryophytes can store their carbohydrates as sucrose and fructan, as exhibited in ***Porella platyphylla*** (Figure 50) and ***Sphagnum flexuosum*** (Figure 51-Figure 52) (Marschall 2010). Galloway and Black (1989) demonstrated that the bryophytes they tested have the necessary enzymes for sucrose to enter cellular metabolism by the sucrose synthase pathway. Adding glucose, fructose, and sucrose to the medium causes these bryophytes to down-regulate photosynthesis when the bryophytes are kept either in the dark or in the light (Marschall 2010). On the other hand, when no carbohydrates were added, darkness had little influence on total carbohydrates, suggesting that they maintain a well-regulated carbohydrate pool.

Figure 49. ***Dicranum scoparium*** on forest floor, a species that seems to benefit from added sucrose in culture. Photo by Janice Glime.

Figure 50. ***Porella platyphylla***, a species that stores carbohydrates as sucrose and fructan, growing better when these and other sugars are added to the growth medium. Photo by Janice Glime.

Figure 51. ***Sphagnum flexuosum*** in its habitat on the forest floor. Photo by Michael Lüth, with permission.

Figure 52. ***Sphagnum flexuosum***, a species that stores sucrose and fructan and down-regulates photosynthesis when sugars are available in the medium. Photo by Michael Lüth, with permission.

Sugars differ in their effects on bryophyte development (Sabovljevic *et al*. 2005). In ***Bryum argenteum*** (Figure 53), added sugars have a positive effect on development of the protonema and multiplication of the shoot. On the other hand, all tested sugars had a negative effect on both of these developmental stages in ***Atrichum undulatum*** (Figure 31).

Figure 53. ***Bryum argenteum***, a species for which added sugars have a positive effect on development. Photo from India Biodiversity Images, through Creative Commons.

Not only do bryophytes store sugars, but as we might expect as a consequence, they also release them (Coxson *et al*. 1992). In the tropical montane rainforest of Guadeloupe, frequent wet-dry cycles cause the epiphytic bryophytes to accumulate 950 kg ha^{-1} of sugars and polyols. These are released during rewetting, contributing to sugars available to other organisms in the canopy. The canopy leafy liverwort ***Frullania atrata*** (Figure 54) stored 17% of its dry biomass as sugar and polyol reserves, whereas the lower canopy species ***Phyllogonium fulgens*** (Figure 55) stored less than 6%. On the other hand, it was the lower canopy bryophytes that released the most sugars and polyols (0.9 g m^{-2}) during rewetting, compared to 0.3 g m^{-2} for the upper canopy. This release yielded an estimated 122 kg ha^{-1} from the upper canopy. These sugars contribute significantly to nutrient cycling by providing an energy source for the decomposer organisms.

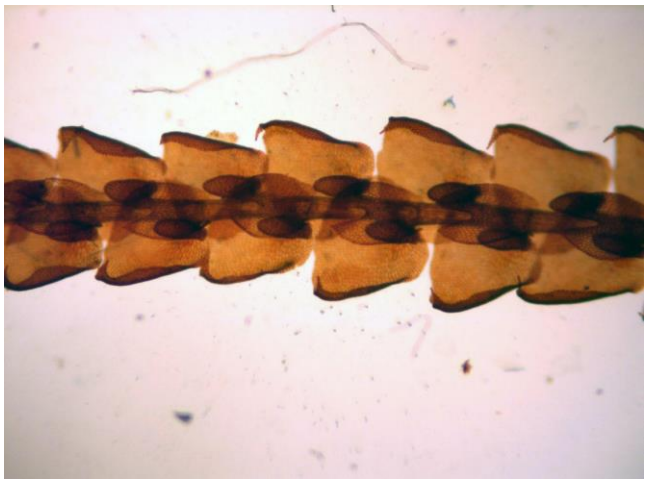


Figure 54. ***Frullania atrata***, a species that has 17% of its dry biomass as sugars in the canopy of a tropical cloud forest. Photo by Juan Larrain, with permission.

Figure 55. ***Phyllogonium fulgens***, a Neotropical species that lives in the lower canopy and stores less than 6% of its dry biomass as sugars. Photo by Michael Lüth, with permission.

Fungal Connections – Mycorrhizae?

One mode of uptake by bryophytes has largely been ignored by ecologists until recently, potentially causing researchers to be looking in the wrong places or not all important places for bryophyte effects on ecosystem nutrient budgets. That mode is by means of **mycorrhizae** (fungal associations that function in transfer of nutrients to roots or rhizoids) or similar partnerships with **fungi**.

In 1976, Kottke and coworkers recognized that the ability of mosses to compete was affected by differential growth stimulation of the mosses by fungi. Still, little attention was paid to moss-fungal interactions from an ecosystem perspective, but bryologists began noticing that many mosses seemed to have fungal hyphae associated with their underground parts. Meanwhile, the tree physiologists were recognizing that fungal partners were critical to the nutrient and water uptake of trees. And orchid growers recognized that the native fungi must be kept with the orchids for successful growth. Now, fungi are recognized as essential to the nutrient uptake of tree

roots, and stories about their partnerships with roots are replacing the traditional teaching emphasis on root hair mechanisms of uptake.

Ecologists estimate that 95% of all plant species are in genera that form mycorrhizal associations (Sylvia *et al*. 2004). In temperate and boreal forests, up to 95% of the short roots of trees form **ectomycorrhizae** [form of symbiotic relationship that occurs between a fungal symbiont and the roots (or rhizoids) of various plant species]. Mycorrhizae are critically important to most forest trees, which depend on them to increase surface area and contact nutrients in a much greater volume of soil than the tree is able to reach. Bryophytes, likewise, are able to take advantage of this partnership to reach sources otherwise unavailable to them. Even in the Antarctic, such fungal relationships can be important, as in the leafy liverwort ***Cephaloziella exiliflora*** (Figure 56) (Williams *et al*. 1994; Chambers *et al*. 1999). There are also indications that nutrients are transferred from the moss mat to the tree roots through mycorrhizae (Chapin *et al*. 1987). But we know little of the extent of these relationships.

Figure 56. ***Cephaloziella exiliflora***, a leafy liverwort with fungal associations in the Antarctic. Photo by Tom Thekathyil, with permission.

Although Boros reported a unique parasitic fungus on mosses in 1926, most botanists considered the bryophytes to be almost immune from fungal attack; even less attention was paid to the possibility of any sort of fungal partnership. In 1970, Kamal and Singh reported on the **rhizoidosphere** fungal flora of bryophytes. In 1975, Pirozynski and Malloch offered the theory that mycorrhizae were an essential part of the invasion of land by the original bryophyte-like plants, helping them to survive in an environment that was poor in nutrients and sustained frequent periods of desiccation. But actual proof of a mycorrhizal partnership, extant or extinct, was not forthcoming.

Finally, in the 1980's, reports of bryophyte mycorrhizal (shouldn't it be mycorrhizoidal?) associations began to appear in the literature (Parke & Linderman 1980; Rabatin 1980; Pocock & Duckett 1985a; Iqbal *et al*. 1988a, b; Ligrone 1988). These have included associations with ***Funaria hygrometrica*** (Figure 57) (Parke & Linderman 1980; Iqbal *et al*. 1988a), ***Sphagnum palustre*** (***cymbifolium***) (Figure 58-Figure 59), ***Polytrichum commune*** (Figure 4) (Iqbal *et al*. 1988a), and in ***Marchantia emarginata*** (=***M. palmata***, Figure 60) both rhizoids and the ventral thallus (Iqbal *et al*. 1988b).

Figure 57. ***Funaria hygrometrica*** protonemata and buds, as well mature plants with capsules – a mycorrhizal species. Photo by Janice Glime.

Figure 58. ***Sphagnum palustre*** habitat. Photo by Michael Lüth, with permission.

Figure 59. ***Sphagnum palustre***, a species with mycorrhizal associations. Photo by Michael Lüth, with permission.

Figure 60. ***Marchantia emarginata***, a species with mycorrhizal associations. Photo from Taiwan Mosses, through Creative Commons.

Ligrone and Lopes (1989) demonstrated **vesicles** and **arbuscules** ("little trees"; branched structures formed by fungi within plant cells; Figure 61) in both rhizoids and parenchyma cells of the thallose liverwort ***Conocephalum conicum*** (Figure 62), suggesting a true mycorrhizal association. The arbuscules are thought to be the site of nutrient exchange (Harrison 1999), at least in roots. Even ***Phaeoceros laevis*** (Figure 63), a member of the **Anthocerotophyta** and host of a ***Nostoc*** (**Cyanobacteria**; Figure 64-Figure 65) symbiont, has a fungal associate that appears to be mycorrhizal (Ligrone 1988). When ***P. laevis*** is infected, the plastid forms a networking structure, the vacuole mass decreases, and the organelle density increases, all modifications suggestive of a partnership.

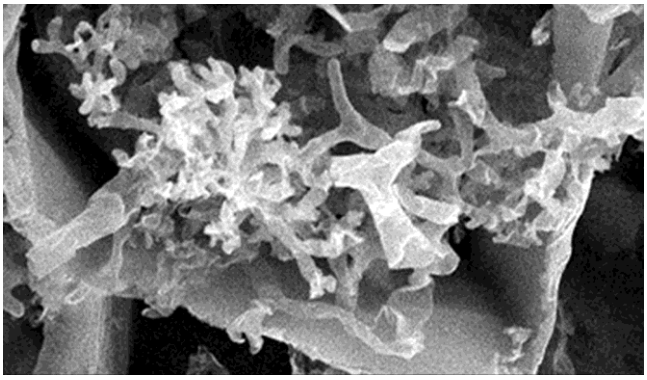


Figure 61. Arbuscules typical of those formed in roots by VAM fungi, but in this case within the thallus of the liverwort ***Marchantia foliacea***. Photo by Julia Russell, with permission.

Figure 62. Thallus of ***Conocephalum conicum***. Photo by Janice Glime.

Figure 63. ***Phaeoceros laevis*** sporophytes, a species with both **Cyanobacteria** (***Nostoc***) and a fungal associate. Photo by Robert Klips, with permission.

Figure 64. Colonies of **Cyanobacteria** (***Nostoc*** or ***Aphanothece***) with mosses. Photo by Janice Glime.

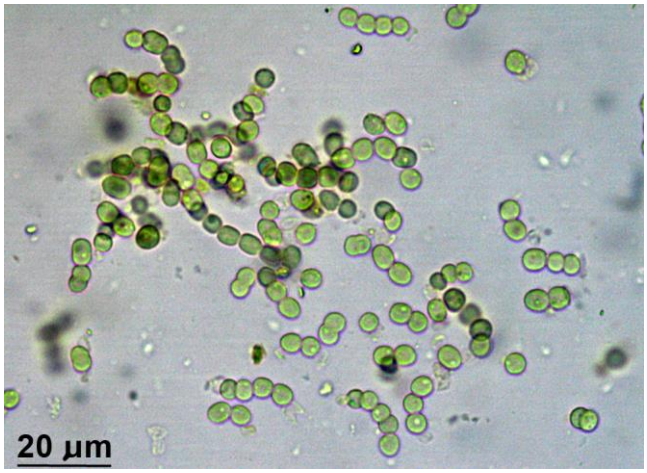


Figure 65. ***Nostoc*** colonies from the hornwort ***Anthoceros agrestis***. Photo by Ralf Wagner <www.dr-ralf-wagner.de>, with permission.

In 1985, Pocock and Duckett (1985b) investigated the rhizoids of 206 of the 284 British liverworts. They found that swollen rhizoids occurred in 33 species of the Jungermanniales and were always associated with fungal hyphae. Swollen and branched rhizoids were particularly well developed in the **Lepidoziineae** (Figure 66) and **Cephaloziineae** (Figure 94) and often occurred on flagelliform shoots, but were better developed on the

underground axes. Duckett *et al*. (1991) later described the highly specialized associations between ascomycetous fungi, known for their ectomycorrhizal partnerships, and 46 species of British liverworts. They found the majority of these ascomycetous fungi to occur with the leafy liverwort suborders **Lepidoziineae** and **Cephaloziineae**. [Ascomycetous associations are found in a relatively small number of families of leafy liverworts (Read *et al*. 2000)]. Strikingly, 33 of these 46 British liverwort taxa form flagelliform axes (Duckett *et al*. 1991). These axes have elongate parenchyma cells with abundant plasmodesmata in their transverse end walls. Their apices are mucilaginous and the subapical amyloplasts appear to act in detecting gravity, much as they do in protonemata. In addition to serving as perennating structures, these axes appear to be major organs of assimilation. Is this facilitated through a mycorrhizoidal partnership?

In all these leafy liverwort cases, the fungi infect the individual rhizoids independently, but most of these 46 taxa nevertheless have abundant fungi-infected rhizoids that extend 20-30 cm into the peaty substrate (Duckett *et al*. 1991). What an extension for a tiny bryophyte! In the liverworts ***Lepidozia*** (Figure 66), ***Kurzia*** (Figure 67), and ***Telaranea*** (Figure 68), but known in no others, the rhizoids swell prior to fungal infection. In ***Cladopodiella*** (Figure 69), the fungi form a pseudoparenchymatous sheath around the swollen rhizoidal tips.

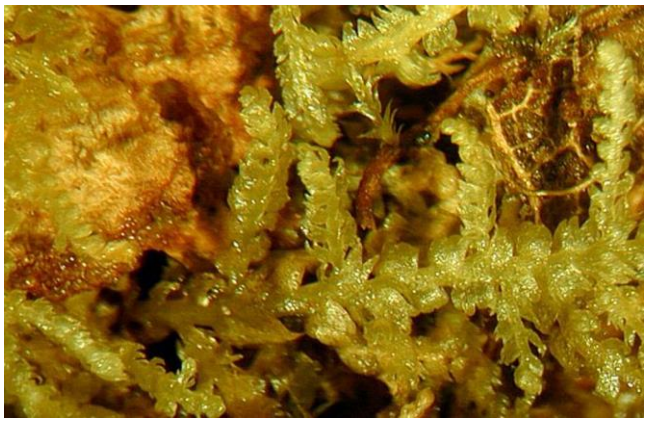


Figure 66. ***Lepidozia reptans***, a species in which rhizoid tips swell prior to fungal association. Photo by Michael Lüth, with permission.

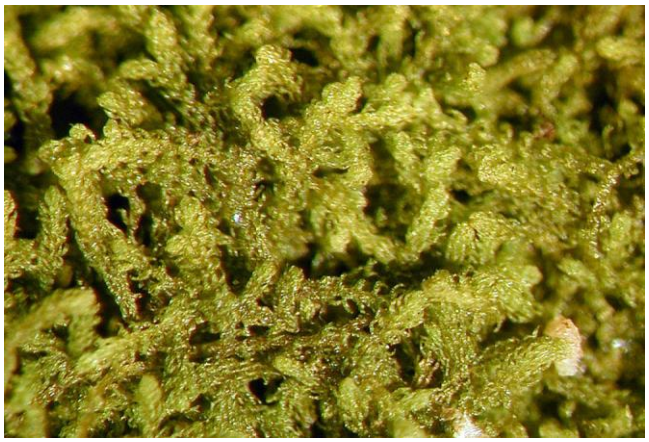


Figure 67. ***Kurzia cf trichoclados***, a species in which rhizoid tips swell prior to fungal association. Photo by Michael Lüth, with permission.

Figure 68. ***Telaranea nematodes***, a species in which rhizoid tips swell prior to fungal association. Photo by Michael Lüth, with permission.

Figure 69. ***Cladopodiella fluitans***, a species in which fungi form a sheath around rhizoid tips. Photo by Kristian Peters, with permission.

By 1988, Boullard had presented evidence that the fungal symbiotic relationship with the liverworts was evolutionarily very old. Yet, in 1990, During and van Tooren pointed out that "in only very few cases have these interactions been analysed functionally."

Other associations have been documented in the field. Although not truly mycorrhizoidal because they lack the composite structure definitive of this relationship, bryophytes now are known to enter into partnerships. Even buried wood, inoculated with ^{32}P, was able to provide P for the living tips of ***Hypnum cupressiforme*** (Figure 70-Figure 71) through a saprotrophic fungus, ***Phanerochaete velutina*** (Figure 72), that connected to the older parts of the moss (Wells & Boddy 1995).

Figure 70. ***Hypnum cupressiforme*** growing on a log. Photo by Michael Lüth, with permission.

Figure 71. ***Hypnum cupressiforme***, a species that is able to derive phosphorus from buried wood. Photo by David Holyoak, with permission.

Figure 72. ***Phanerochaete velutina***, a fungal associate of ***Hypnum cupressiforme*** on wood. Photo by James K. Lindsey, with permission.

The fungal association may in some small way benefit the neighboring plants, and they in turn the bryophyte (Duckett & Read 1995). As noted earlier, Chapin *et al*. (1987) have found an association that may indeed benefit the trees. In an Alaskan forest they found that the mycorrhizal fungi of the black spruce (*Picea mariana*, Figure 3) stimulated the moss carpet above to release phosphorus to the tree roots! When the mycorrhizae were inhibited, more P remained with the mosses and less escaped from the plots, where it presumably went to tree roots.

Rhizoids of at least some leafy liverworts in the **Lepidoziaceae** (Figure 66), **Calypogeiaceae** (Figure 73), **Cephaloziaceae** (Figure 94), and **Cephaloziellaceae** (Figure 56) can be infected by the same fungus, ***Hymenoscyphus ericae*** (Figure 74), an ascomycetous fungus, that infects members of the Ericaceae such as *Calluna* (Figure 75), *Erica* (Figure 76), *Rhododendron* (Figure 77), and *Vaccinium* (Figure 78-Figure 79) (Duckett & Read 1995). So far, there appears to be no evidence of a transport pathway from moss to fungus to ericaceous plant or vice versa, but the presence of one of these host plants would enhance the opportunities for the fungus to grow there and thus provide greater opportunities for the fungus to join with the other host. This is similar to the partnership between *Monotropastrum humile* (an achlorophyllous flowering plant; Figure 80), a fungus, and a beech tree (*Fagus crenata*, Figure 81) (Kasuya *et al*. 1995). The fungus in the *F. crenata* appears to be the same as that in the *M. humile*, and evidence implies that the fungus joins the two tracheophytes. In this way, the *M. humile* could take advantage of the sunlight reaching the canopy of *Fagus crenata* by receiving carbohydrates from the canopy transferred through the fungus to the *M. humile*. The fungus appears to be a member of the Russulaceae (Figure 82) (Yamada *et al*. 2008; Matsuda *et al*. 2011).

Figure 73. ***Calypogeia azurea***, a leafy liverwort that can be infected with ***Hymenoscyphus ericae***. Photo by Hermann Schachner, through Creative Commons.

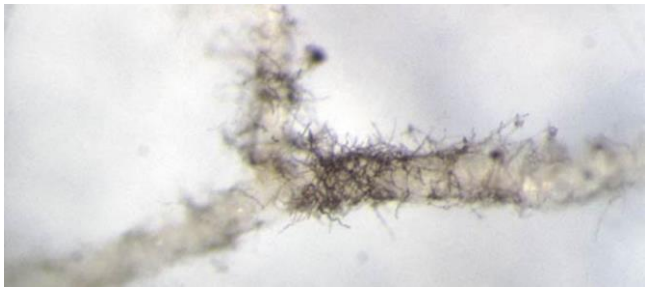


Figure 74. ***Hymenoscyphus ericae*** on ***Rhododendron*** root, a species that can infect leafy liverworts in **Lepidoziaceae** (Figure 66), **Calypogeiaceae** (Figure 73), **Cephaloziaceae** (Figure 94), and **Cephaloziellaceae** (Figure 56). Photo by Mark C. Starrett, David A. Heleba, and Adam R. Wheeler, through Creative Commons.

Figure 75. ***Calluna vulgaris***, a host for the fungus ***Hymenoscyphus ericae***. Photo by Janice Glime.

Figure 76. ***Erica*** sp., a host for the fungus ***Hymenoscyphus ericae***. Photo by Janice Glime.

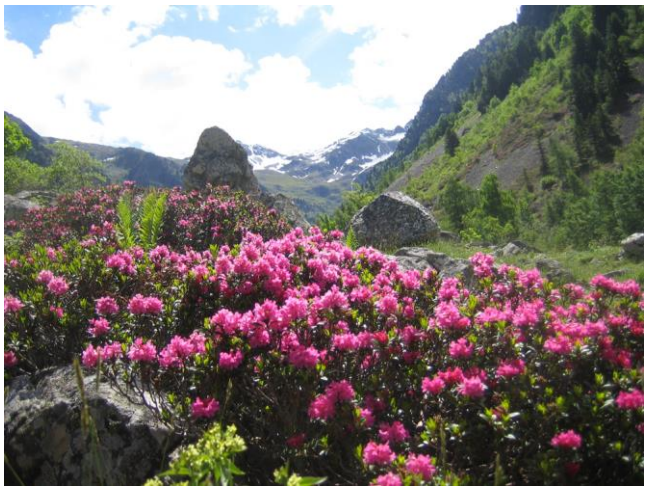


Figure 77. ***Rhododendron ferrugineum***, a host for the fungus ***Hymenoscyphus ericae***. Photo by Albert Kok, through Creative Commons.

Figure 78. ***Vaccinium angustifolium*** in ***Pinus banksiana*** forest. ***Vaccinium*** is a host for the fungus ***Hymenoscyphus ericae***. Photo by Photo by Mricon, through Creative Commons.

Figure 79. ***Vaccinium angustifolium***, a host for the fungus ***Hymenoscyphus ericae***. Photo by Mricon, through Creative Commons.

Figure 80. ***Monotropastrum humile***, an achlorophyllous flowering plant that partners with a beech tree through a fungal partner. Photo by Qwert, through Creative Commons.

Figure 81. ***Fagus crenata***, host tree for ***Monotropastrum humile*** and its fungal partner. Photo by Alpsdake, through Creative Commons.

Figure 82. *Russula.cavipes* with mosses, an ectomycorrhizal fungus in the family Russulaceae that is associated with *Monotropastrum humile* and *Fagus crenata*. Photo by James K. Lindsey, with permission.

Cryptothallus mirabilis

It appears that the fungi may be to some liverworts what the mycorrhizae are to the grape fern *Botrychium* and to many of the saprophytic forest floor flowering plants – a means of getting sufficient energy when the canopy is blocking an extensive portion of the light. Such a relationship is essential to the thallose liverwort ***Cryptothallus mirabilis*** (Figure 83), a European species known as ghostwort. It occurs nestled in mires and lacks chlorophyll. Certainly for it, a partnership is essential. But this liverwort has a **Basidiomycota** fungus as its ectomycorrhizal partner (Ligrone *et al*. 1993). They concluded that this liverwort is a **parasite**! It was thought that its fungal partner joined it to a species of *Betula* (birch), from which it ultimately obtained its carbohydrate energy source (Wiehle 1988; Pocock & Duckett 1984; Frey & Kürschner 1991; Read *et al*. 2000), much like the parasitic flowering plant *Monotropa uniflora* (Figure 84), the Indian pipe. However, Ligrone *et al*. (1993) disagree. They found that the fungi in *Betula* roots had a different morphology from those in the associated ***C. mirabilis***. It appears that the association of ***C. mirabilis*** is more like that of the goblin fern *Botrychium mormo*, wherein the fungus derives carbon from decomposing litter and transfers some of it to the fern, permitting it to live in low light (Gundale 2002). But could it also be that the form of the fungus depends on the host, thus differing between that of the ***C. mirabilis*** and that of the *Betula*?

Bidartondo *et al*. (2003) determined that ***Cryptothallus mirabilis*** (**Basidiomycota**; Figure 83-Figure 86) is an **epiparasite**, depending on a species of the fungus ***Tulasnella*** (Figure 85-Figure 86). This fungus forms **ectomycorrhizal** (symbiotic relationship between fungal symbiont and roots of plant species) associations with surrounding trees. It is able to transfer labelled ^{14}C from birch (*Betula*) seedlings in the lab, and presumably from tree roots in the field. Species of this same genus are also associated with ***Aneura pinguis*** (Figure 87) (Kottke *et al*. 2003) and some orchids (Clements & Ellyard 1979; Roche *et al*. 2010).

Figure 83. ***Cryptothallus mirabilis***, an achlorophyllous thallose liverwort in the **Aneuraceae**. This parasitic liverwort depends on a basidiomycete fungus to provide it with nutrients and energy. Photo by Michael Lüth, with permission.

Figure 84. ***Monotropa uniflora***, an achlorophyllous flowering plant that gets its carbon through its fungal partner. Photo by Magellan, through Creative Commons.

Figure 85. ***Tulasnella*** sp. ectomycorrhizae from a ***Betula pendula*** association. Photo courtesy of Martin Bidartondo.

Figure 86. ***Cryptothallus mirabilis*** and its symbiotic partner ***Tulasnella*** sp. Photo courtesy of Martin Bidartondo.

Figure 87. ***Aneura pinguis***, a species with chlorophyll and that is closely related to ***Cryptothallus mirabilis***. Photo by Michael Lüth, with permission.

When it develops, the ***Cryptothallus mirabilis*** (Figure 83) fungus (Figure 88-Figure 91) forms large, intracellular coils in the liverwort (Ligrone *et al* 1993). Then the liverwort cytoplasm proliferates and the starch content of its plastids decreases. As the hyphae die back and aggregate into large masses, the liverwort cells senesce. In ***C. mirabilis***, the fungal hyphae contain abundant glycogen and occasionally amyloid deposits. It is interesting that the fungal partner in ***C. mirabilis*** is identical to the one in ***Aneura pinguis*** (closely related but photosynthetic; Figure 87) from alpine sites but different from the fungus in ***A. pinguis*** from a chalk pit and sand dunes. In ***C. mirabilis***, net carbon transfer is to the liverwort, and it is likely that there is transfer from the fungus to the liverwort in ***A. pinguis*** as well. In addition to the morphological similarities, further support for this hypothesis in ***A. pinguis*** is that spores of both liverwort species fail to develop beyond a few cells in **axenic** (sterile) culture.

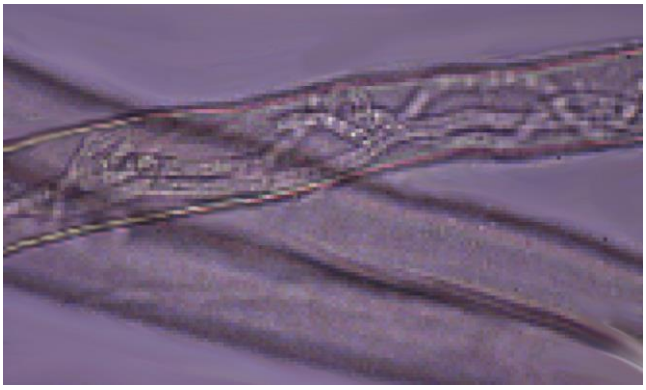


Figure 88. ***Cryptothallus mirabilis*** rhizoid with ***Tulasnella*** sp. Photo by Martin Bidartondo, with permission.

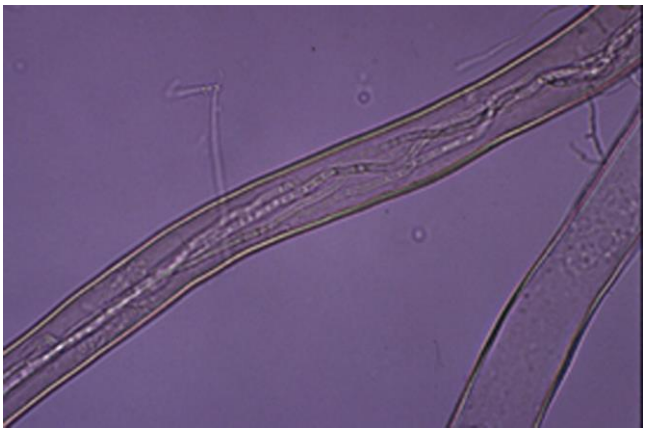


Figure 89. ***Cryptothallus mirabilis*** rhizoid with its fungal partner ***Tulasnella*** sp. Photo courtesy of Martin Bidartondo.

Figure 90. ***Cryptothallus mirabilis*** with ***Tulasnella*** sp. Photo by Martin Bidartondo, with permission.

Figure 91. ***Cryptothallus mirabilis*** with fungal partner ***Tulasnella.*** Photo courtesy of Martin Bidartondo.

Bidartondo and Duckett (2010) concluded that most of the thalloid liverworts contain **Glomeromycota** (Figure 92) that form arbuscular mycorrhizae with them. Many leafy liverwort species and members of the thallose **Aneuraceae** have a relationship with **Basidiomycota**. Whereas the **Aneuraceae** associate almost exclusively with species of ***Tulasnella***, eight leafy liverwort genera predominately associate with members of ***Sebacina vermifera*** (**Basidiomycota**; see Figure 93). ***Sebacina*** species have a habit of surrounding plants, so some of them may envelop the plants and prevent photosynthesis. It is interesting to note that when multiple species of bryophytes occur together, they rarely share the same fungal species. Furthermore, the bryophyte symbioses are not like those of the tracheophytes.

Figure 92. ***Claroideoglomus claroideum***, a member of **Glomeromycota**, common on bryophytes. Photo from Biomesfirst09, through Creative Commons.

Figure 93. ***Sebacina incrustans***, a jelly fungus, on moss, surrounding it an ultimately able to kill it. Photo © Slavko Serod, with online permission for non-commercial use.

Underground and Other Partnerships

It appears that ***Cryptothallus*** (Figure 83) is not the only liverwort capable of living below ground with an **Ascomycota** fungal partner (Duckett *et al*. 1989). In bog communities, the leafy liverworts (**Jungermanniales**) ***Cephalozia*** (Figure 94), ***Cladopodiella*** (Figure 69), ***Kurzia*** (Figure 67), ***Lepidozia*** (Figure 95), ***Odontoschisma*** (Figure 96), and ***Telaranea nematodes*** (Figure 68) can all develop extensive underground stem systems with numerous rhizoids that have swollen, fungus-containing tips. These liverworts can produce new shoots down to 24-30 cm in peat and to 10 cm in rotten logs (***Lepidozia reptans***, Figure 95).

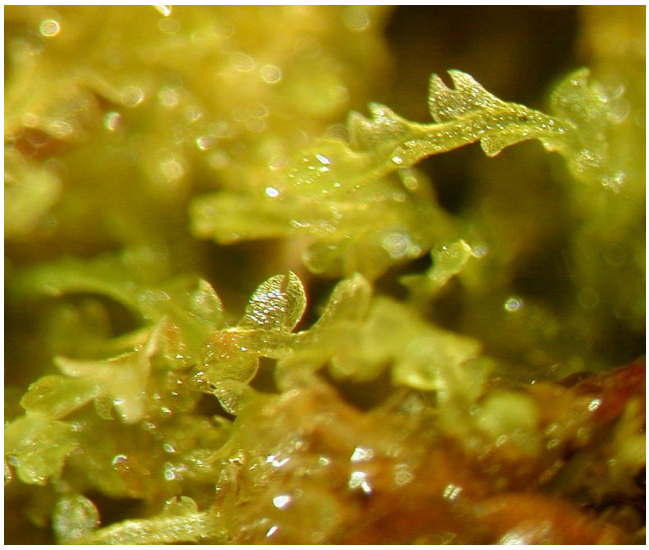


Figure 94. ***Cephalozia macrostachya***, member of a genus that houses fungi in swollen rhizoid tips in bogs. Photo by Michael Lüth, with permission.

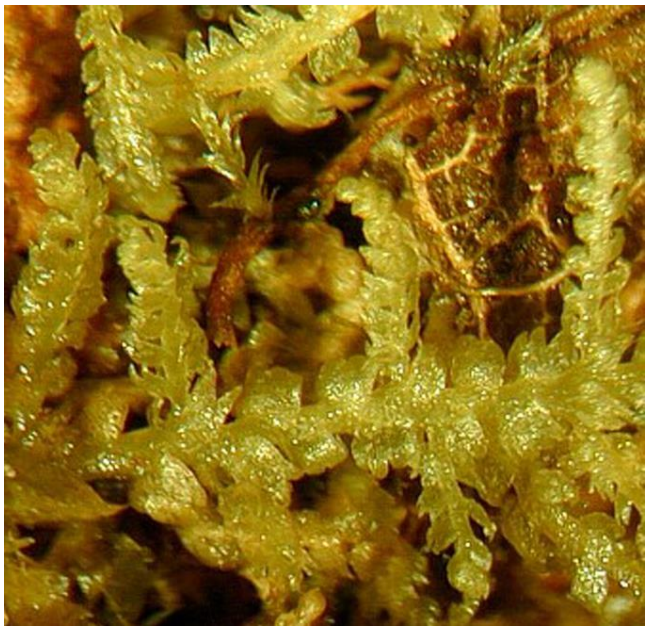


Figure 95. ***Lepidozia reptans*** growing on rotten wood that its rhizoids can penetrate down to 10 cm. Photo by Michael Lüth, with permission.

Figure 96. ***Odontoschisma macounii***, a species that houses fungi in swollen rhizoid tips in bogs. Photo by Michael Lüth, with permission.

In Malaysia, members of the leafy liverwort family **Lepidoziaceae** (Figure 95) can produce such axes down to 1.5 m in the peaty soil of the upper montane rainforest (Duckett *et al*. 1989). When these develop in the dark, they retain their partnership morphology, but when the shoots are exposed to light they regenerate into leafy shoots and lose their gravitropic response. This loss of fungal partnership morphology appears to be related to the disappearance of subapical amyloplasts, known to have a gravimetric response. Duckett and coworkers suggest that these liverworts may be acting as alternative hosts to ericaceous mycorrhizae, particularly in places like Malaysia. In Great Britain, less than 20% of the **Jungermanniales** (Figure 94-Figure 95) have rhizoidal fungi, whereas in the montane forests of Malaysia, where ericaceous shrubs are extensive, the percentage may be as high as 80-90%.

As the search continues, more and more fungal taxa are being described in bryophyte associations, but not all are mycorrhizal (Khan *et al*. 1997; Döbbeler 1997; Brouwer 1999). In fact, a number appear to be parasitic; others are just coexisting, perhaps benefitting from the modulated temperature and moisture. Nevertheless, approximately 300 species of **Ascomycota** appear to grow obligately on bryophytes (Döbbeler 1997). More than 40 species of **Ascomycota** in six orders occur on the **Polytrichaceae** alone, primarily on ***Polytrichum*** *s.l.* (Figure 4, Figure 34) and ***Dawsonia*** (Figure 97) (Felix 1988). Some fungi, for example ***Lemprospora*** (Figure 98) and ***Octospora*** (Figure 99), are known only from bryophytes (Döbbeler 1997; Brouwer 1999); in other cases, the bryophyte has never been found without its fungal associate (Döbbeler 1997). ***Octospora*** and other genera infect the subterranean rhizoids of **Polytrichaceae** (Figure 4, Figure 34, Figure 97), while others occupy the spaces between the vertical leaf lamellae (Felix 1988). In fact, 20 different **Ascomycota** species are known to occupy that unusual habitat without apparently having any effect on the moss.

Figure 97. ***Dawsonia superba***, a genus that has **Ascomycota** associates. Photo by Jan-Peter Frahm, with permission.

Figure 98. ***Lamprospora seaveri***, a fungus that only occurs on bryophytes. Photo by G. Moyne, through Creative Commons.

Figure 99. ***Octospora excipulata***, a fungus that lives exclusively on bryophytes. Photo by Malcolm Storey, through Creative Commons.

Raspe and De Sloover (1998) suggested that the discomycetous fungus ***Mniaecia jungermanniae*** (Figure 100-Figure 101), which lives exclusively on leafy liverworts in the **Jungermanniales** (Figure 100), might have achieved the first step toward mutualism. This destructive parasite grows inside the bryophyte rhizoids but

does not seem to afford any direct benefit to the liverwort. It appears it has a long way to go to reach mutualism.

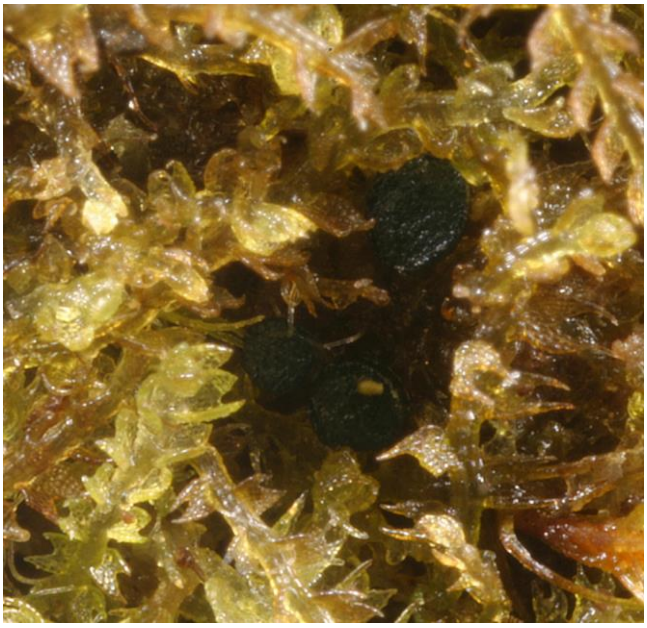


Figure 100. ***Mniaecia jungermanniae*** (fungus in center) on leafy liverworts. Photo by Malcolm Storey (DiscoverLife), with online permission.

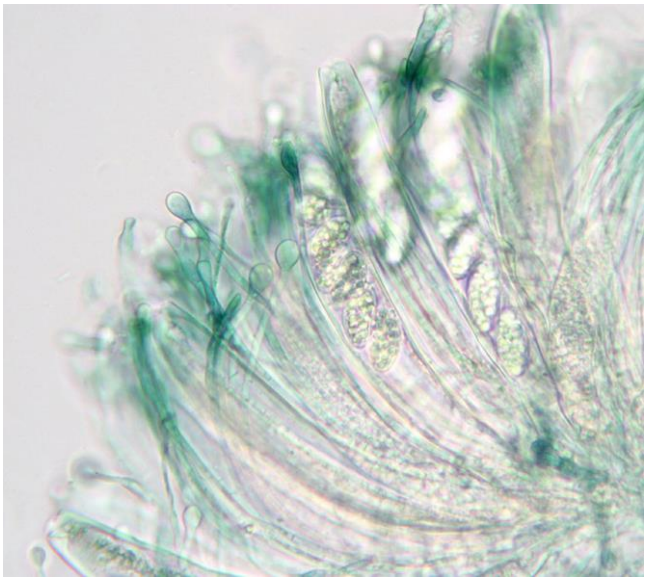


Figure 101. ***Mniaecia jungermanniae*** asci and ascospores. Photo by Malcolm Storey (DiscoverLife), with online permission.

We have noted several times that bryophytes obtain phosphate, and possibly other nutrients, from the bedrock. It is likely that at least in some cases fungi contribute to this nutrient source. Calling them "**rock-eating mycorrhizae**," Schöll *et al*. (2008) demonstrated that tunnels occur in mineral grains. They attributed these to hyphae from ectomycorrhizae that can dissolve mineral grains. Whether these fungi are directly associated with bryophytes, or they use litter or other plants for their carbon source, these fungi permit phosphates and other nutrients to enter nutrient cycling, potentially making some of them available to the bryophytes.

Arbuscular Mycorrhizae

Harrison (1999) reported that arbuscular mycorrhizae, restricted to the fungal order **Glomales** (**Zygomycota**, more recently named **Glomeromycota**; Figure 92), infected some bryophytes. Schüßler (2000) reported that a member of this order, ***Claroideoglomus claroideum*** (Figure 92), formed a mycorrhiza-like symbiosis with the hornwort ***Anthoceros punctatus*** (Figure 102-Figure 103). Following inoculation with spores, Schüßler found branched hyphae within the thallus within 20 days. This was the first definite experimental establishment of an arbuscular mycorrhiza-like association between a member of the **Glomales** and a bryophyte, although Felix (1988) had reported mycorrhiza-like associations in a number of taxa (Table 3). In 2003, Jakucs *et al*. found vesicles of a glomalean fungus in the moss ***Hypopterygium*** (Figure 104), suggesting that there might indeed be a mutualistic relationship in which the fungus also benefits, but that hypothesis still awaits verification.

Figure 102. ***Anthoceros punctatus*** with young sporophytes; ***Claroideoglomus claroideum*** forms a mycorrhizal association with this species. Photo by Des Callaghan, with permission.

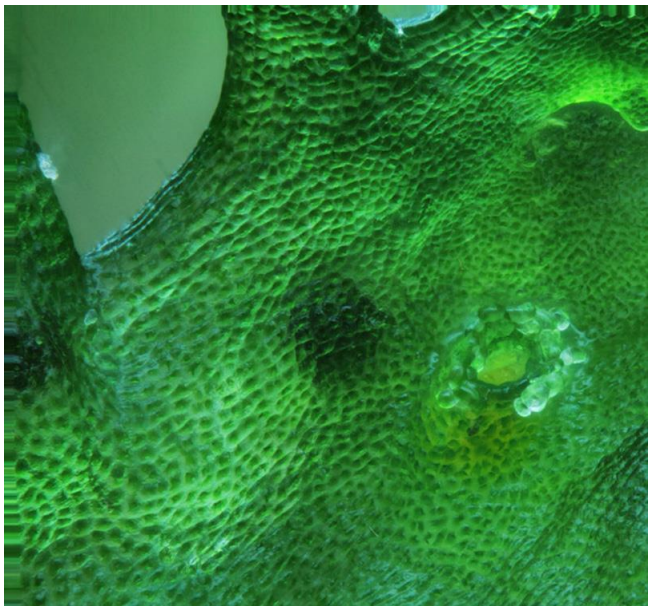


Figure 103. ***Anthoceros punctatus*** antheridial pit and ***Nostoc*** colony (dark area). Photo by Des Callaghan, with permission.

Figure 104. ***Hypopterygium didictyon***, a species that associates with a glomalean fungus (Figure 92). Photo by Juan Larrain, through Creative Commons.

Table 3. Mycorrhiza-like fungus-bryophyte associations. From Felix (1988) and Russell & Bulman (2004).

Fungus	Bryophyte	Reference
various spp	*Anthoceros*	Kamal & Singh 1970, Singh 1974
	Riccia	"
	Funaria	"
	Polytrichum commune	Grasso & Scheirer 1983
	Haplomitrium	Carafa *et al*. 2003
phycomycetous mycorrhizae	*Marchantia berteroana*	Baylis 1970
swollen rhizoids	liverworts	Pocock & Duckett 1985b
Endogone	bryophytes	Gerdemann 1968
Glomus tenuis	*Pogonatum*	Rabatin 1980
Glomus mosseae group	*Marchantia foliacea*	Russell & Bulman 2004
Claroideoglomus claroideum	*Anthoceros punctatus*	Schüßler 2000
Mycena cinerella	*Atrichum undulatum*	Hildebrand *et al*. 1978
	Brachythecium rutabulum	"
	Funaria hygrometrica	"

Figure 105. ***Mycena*** sp.; ***M. cinerella*** forms mycorrhizae with ***Atrichum undulatum***, ***Brachythecium rutabulum***, and ***Funaria hygrometrica***. Photo by James K. Lindsey, with permission.

Figure 106. ***Endogone pisiformis***, a genus known to form mycorrhizae with bryophytes. Photo by Adolf and O. Ceska, with permission.

There is a certain degree of specificity among the bryophyte species that have fungal associations. Russell and Bulman (2004) found that ***Marchantia foliacea*** (Figure 107-Figure 109) from two locations in New Zealand supported ***Glomus*** (Figure 109) (*n.b.*, many species of ***Glomus*** are now placed in ***Claroideoglomus***; Figure 92) arbuscular fungi internally (Figure 109), but that ***M. polymorpha*** (Figure 110) did not. Every ***M. foliacea*** thallus they examined contained this ***Glomus*** species in the parenchyma tissue around the midrib. The fungus invaded the thallus through the smooth rhizoids and grew upward through the thallus, forming arbuscules only in the upper portion of the thallus. The hyphae crossed directly through the cell walls of the liverwort. This same fungus forms mycorrhizal associations with the conifer, *Podocarpus* (Figure 111), and it may be that this fungus is shared by both plants. Unfortunately, we still have no evidence if this relationship between the fungus and the liverwort is truly symbiotic.

Figure 107. ***Marchantia foliacea*** thallus, a species that houses arbuscular growth of the mycorrhizal fungus ***Glomus*** (Figure 92) around the midrib. Photo courtesy of Julia Russell.

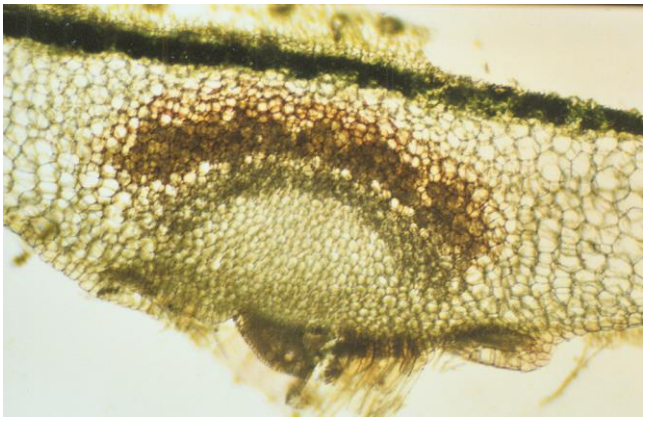


Figure 108. ***Marchantia foliacea*** thallus with arbuscular growth of the mycorrhizal fungus ***Glomus*** (Figure 92) around the midrib. Photo courtesy of Julia Russell.

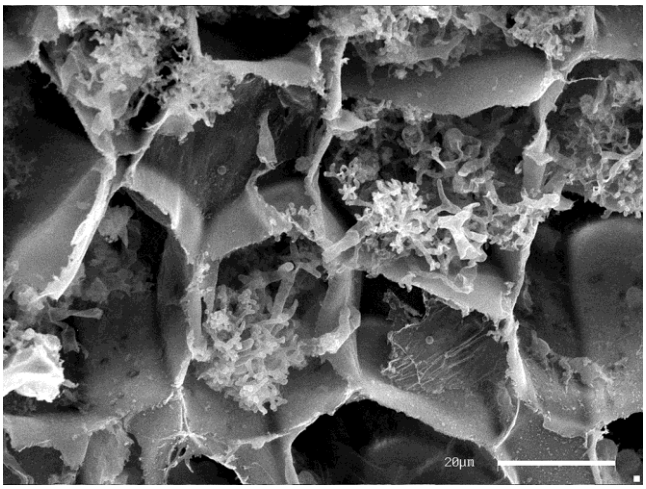


Figure 109. SEM ***Marchantia foliacea*** thallus with arbuscular growth of the mycorrhizal fungus ***Glomus*** in cells around the midrib. Photo courtesy of Julia Russell.

Figure 110. ***Marchantia polymorpha*** developing archegoniophores, a species that does not form an association with the fungus ***Glomus*** (Figure 92). Photo by Bob Klips, with permission.

The specificity of some of the groups for specific plant phyla is fascinating. For example, **Zygomycota** (Figure 112) colonize members of **Anthocerotophyta** (Figure 102) and **Marchantiophyta** (Figure 110, Figure 113), but not **Bryophyta** (Figure 104) (Read *et al*. 2000). On the other hand, members of the **Glomales** (Figure 92) isolated from the flowering plant *Plantago lanceolata* were able to colonize the thallose liverwort ***Pellia epiphylla*** (Figure 113) and produce arbuscules and vesicles.

Figure 111. ***Podocarpus***, a genus whose roots serve as host for ***Glomus*** (Figure 92) and may share it with bryophytes. Photo by Koppchen, through Creative Commons.

Figure 112. **Zygomycota** sporangia, a phylum that colonizes hornworts and liverworts, but not mosses. Photo by Kristi Yim, through Creative Commons.

Figure 113. ***Pellia epiphylla***, a species that can be colonized by the same member of **Glomales** as those found on the flowering plant *Plantago lanceolata*. Photo by Michael Lüth, with permission.

Figure 114. ***Plantago lanceolata***, a species that has the same fungal partner as ***Pellia epiphylla***. Photo by Forest & Kim Starr, through Creative Commons.

These fungal-bryophyte associations form structural associations similar to those of vesicular-arbuscular mycorrhizae of tracheophytes. Despite the large number of associations recognized between bryophytes and fungi, Read and coworkers (2000) still stressed the "need for analysis of the functional attributes of these symbioses." They presented further evidence that these fungal associations were ancient, being important to the first plants to colonize land. This contention is supported by fossil evidence of glomalean fungal structures associated with early bryophytes in Ordovician sediments that are 460 and 400 million years old (Remy *et al*. 1994; Redecker *et al*. 2000).

Beneficial or Harmful?

The fungal associates are not always beneficial to the bryophytes. Zobel *et al*. (1999) treated a sub-Arctic forest community with fungicide and found that the bryophytes and dwarf shrubs increased in biomass relative to the control. Could it be that the fungi are frequently stealing from the bryophytes and making nutrients available to trees?

Summary

Unlike tracheophytes, bryophytes take up nutrients over their entire surface. With leaves only one cell thick in most taxa every leaf cell is thus exposed to environmental sources of nutrients. The three most limiting nutrients (N, P, K) accumulate in the upper parts of the plants through active uptake, whereas Ca, Mg, and Na accumulate through passive **cation exchange**. Bryophytes have high **cation exchange capacity** (CEC) due to **polyuronic acids** in their cell walls. Once ions are bound on exchange sites, a **proton pump** removes H^+ ions from the cell, creating a **charge gradient** that brings in positive ions. These bring along negative ions by **cotransport**. It appears that bryophytes have two, perhaps more, types of exchange sites, permitting differential binding of ions. They also seem to have specificity for things they need over things they do not. Anion exchange sites can contribute to phosphate uptake. Abundance of cation sites compared to anion sites can account for the preference of ammonium (cation) over nitrate (anion).

Further active processes are able to distinguish ions formed by N, P, and K from more exchangeable cations such as those of Ca^{++} or Mg^{++}, and they are generally able to maintain relatively constant levels of these essential nutrients despite changes in environmental concentrations. Increasing temperatures increase the uptake, which is also pH-dependent. Some uptake occurs through **pinocytosis** and entry of **nanoparticles**.

Fungi are often associated with the rhizoids of bryophytes. It may be that a large number of bryophytes are afforded the advantages of fungal partner relationships, providing them with considerably more surface area for acquiring nutrients. The thallose liverwort ***Cryptothallus mirabilis*** has a fungal partner (***Tulasnella***) that provides carbohydrates for this non-chlorophyllous plant.

Many bryophyte-fungal associations have been discovered, but the types of interaction lack our understanding. We know that glomalean fungi are frequently associated with bryophytes, but the association has not been clearly described. This could be a very fruitful area for further research.

Acknowledgments

I appreciate the contributions of undergraduate Phil Gaudette and M. S. student Jennifer Jermalowicz Jones for their critical reading of the manuscript from the perspectives of students interested in nutrient relationships of bryophytes. Dana Richter made many helpful suggestions on the fungal section. Simon Bulman helped me to locate Julia Russell to obtain her mycorrhizae pictures. Jean Faubert made suggestions to improve the chapter. Many photographers have contributed their images through Creative Commons or have given me permission.

Literature Cited

Babb, T. A., and Whitfield, D. W. S. 1977. Mineral nutrient cycling and limitation of plant growth in the Truelove Lowland ecosystem. In: Bliss, L. E. (ed.). Truelove Lowland, Devon Island, Canada: A High Arctic Ecosystem. University of Alberta Press, Edmonton, pp. 589-606.

Bartsch, I. 1994. Effects of fertilization on growth and nutrient use by *Chamaedaphne calyculata* in a raised bog. Can. J. Bot. 72: 323-329.

Basile, A., Giordano, S., López-Sáez, J. A., and Cobianchi, R. C. 1999. Antibacterial activity of pure flavonoids isolated from mosses. Phytochemistry 52: 1479-1482.

Bates, J. W. 1982a. The role of exchangeable calcium in saxicolous calcicole and calcifuge mosses. New Phytol. 90: 239-252.

Bates, J. W. 1989. Retention of added K, Ca and P by *Pseudoscleropodium purum* growing under an oak canopy. J. Bryol. 15: 589-605.

Bates, J. W. 1992. Mineral nutrient acquisition and retention by bryophytes. J. Bryol. 17: 223-240.

Bates, J. W. 1997. Effects of intermittent desiccation on nutrient economy and growth of two ecologically contrasted mosses. Ann. Bot. 79: 299-309.

Bates, J. W. 2000. Mineral nutrition, substratum ecology, and pollution. In: Shaw, A. J. and Goffinet, B. Bryophyte Biology. Cambridge University Press, Cambridge, UK, pp. 248-311.

Bates, J. W. and Farmer, A. M. 1990. An experimental study of calcium acquisition and its effects on the calcifuge moss *Pleurozium schreberi*. Ann. Bot. 65: 87-96.

Baylis, G. T. S. 1970. Root hairs and phycomycetous mycorrhizas in phosphorus-deficient soil. Plant Soil 33: 713-716.

Belnap, J. and Harper, K. T. 1995. Influence of cryptobiotic soil crusts on elemental content of tissue of two desert seed plants. Arid Soil Res. Rehab. 9: 107-115.

Bewley, J. D. 1979. Physiological aspects of desiccation tolerance. Ann. Rev. Plant Physiol. 30: 195-238.

Bidartondo, M. I. and Duckett, J. G. 2010. Conservative ecological and evolutionary patterns in liverwort–fungal symbioses. Proc. Royal Bot. Soc. London 277: 485-492.

Bidartondo, M. I., Bruns, T. D., Weiß, M., Sérgio, C., and Read, D. J. 2003. Specialized cheating of the ectomycorrhizal symbiosis by an epiparasitic liverwort. Proc. Royal Soc. London B Biol. Sci. 270: 835-842.

Boros, A. 1926. A new parasitic fungus on mosses. Bryologist 29: 2-3.

Boullard, B. 1988. Observations on the coevolution of fungi with hepatics. In: Pirozynski, K. A. and Hawksworth, D. L. (eds.). Coevolution of Fungi with Plants and Animals. Academic Press, London, pp. 107-124.

Breemen, N. van. 1995. Nutrient cycling strategies. CEC/IUFRO Symposium on Nutrient Uptake and Cycling in Forest Ecosystems, Halmstad, Sweden, 7-10 Jun 1993. Plant Soil 168-169: 321-326.

Brehm, K. 1968. Die Bedeutung des Kationenaustausches fiir den Kationengehalt lebender Sphagnen. Planta 79: 324-345.

Breuer, K. and Melzer, A. 1990a. Heavy metal accumulation (lead and cadmium) and ion exchange in three species of Sphagnaceae. Oecologia 82: 461-467.

Breuer, K. and Melzer, A. 1990b. Heavy metal accumulation (lead and cadmium) and ion exchange in three species of Sphagnaceae. II. Chemical equilibrium of ion exchange and the selectivity of single ions. Oecologia 82: 468-473.

Brouwer, E. 1999. Mosschijfjes (*Lamprospora* en *Octospora*): Voorkomen en verspreiding in Nederland. Coolia 42: 2-20.

Brown, D. H. 1982. Mineral nutrition. In: Smith, A. J. E. (ed.). Bryophyte Ecology, Chapman & Hall, London, pp. 383-444.

Brown, D. H. 1984. Uptake of mineral elements and their use in pollution monitoring. In: Dyer, A. F. and Duckett, J. G. (eds.). The Experimental Biology of Bryophytes, Academic Press, New York, London, pp. 229-256.

Brown, D. H. and Bates, J. W. 1990. Bryophytes and nutrient cycling. J. Linn. Soc. Bot. 104: 129-147.

Brown, D. H. and Beckett, R. P. 1985. Intracellular and extracellular uptake of cadmium by the moss *Rhytidiadelphus squarrosus*. Ann. Bot. 55: 179-188.

Brown, D. H. and Brumelis, G. 1996. A biomonitoring method using the cellular distribution of metals in moss. Sci. Total Environ. 187: 153-161.

Brown, D. H. and Buck, G. W. 1978. Cation contents of acrocarpous and pleurocarpous mosses growing in a strontium rich substratum. J. Bryol. 10: 199-209.

Brown, D. H. and Buck, G. W. 1985. The cellular location of metals in two bryophytes and a lichen. Cryptog. Bryol. Lichénol. 6: 279-286.

Brown, D. H. and Wells, J. M. 1990. The extracellular and intracellular uptake of inorganic chemicals by bryophytes. In: Zinsmeister, H. D. and Mues, R. (eds.). Bryophytes, Their Chemistry and Chemical Taxonomy. Proceedings of the Phytochemical Society of Europe 29, Oxford University Press, Oxford, pp. 299-318.

Burton, M. A. S. and Peterson, P. J. 1979. Metal accumulation by aquatic bryophytes from polluted mine streams. Environ. Pollut. 19(1): 39-46.

Büscher, P., Koedam, N., and Neirinck, L. 1983. Cation-exchange capacity of mosses in relation to soil preference. In: Cran, W. J., Janácek, K., Rybová, R., and Sigler, K. (eds.). Membrane Transport in Plants. Proc. Symp. held in Prague, Czechoslovakia Aug. 15-21, 1983. John Wiley & Sons, New York, pp. 477-478.

Büscher, P., Koedam, N., and Speybroeck, Van D. 1990. Cation-exchange properties and adaptation to soil acidity in bryophytes. New Phytol. 115: 177-186.

Canivet, L., Dubot, P., and Denayer, F.-O. 2014. Uptake of iron nanoparticles by *Aphanorrhegma patens* (Hedw.) Lindb. J. Bryol. 36: 104-109.

Canivet, L., Dubot, P., Garçon, G., and Denayer, F. O. 2015. Effects of engineered iron nanoparticles on the bryophyte, *Physcomitrella patens* (Hedw.) Bruch & Schimp, after foliar exposure. Ecotoxicol. Environ. Safety 113: 499-505.

Carafa, A., Duckett, J. G., Ligrone, R. 2003. Subterranean gametophytic axes in the primitive liverwort *Haplomitrium* harbour a unique type of endophytic association with aseptate fungi. New Phytol. 160: 185–197.

Carballeira, A., López, J., and Vázquez, M. D. 1999. Uptake of heavy metals to the extracellular and intracellular components in three species of aquatic bryophytes. Ecotox. Environ. Safety Environ. Res. 44: 12-24.

Chambers, S. M., Williams, P. G., Seppelt, R. D., and Cairney, J. W. G. 1999. Molecular identification of *Hymenoscyphus* from rhizoids of the leafy liverwort *Cephaloziella exiliflora* in Australia and Antarctica. Mycol. Res. 103: 286-288.

Chapin, F. S. III, Oechel, W. C., Cleve, K. van, and Lawrence, W. 1987. The role of mosses in the phosphorus cycling of an Alaskan black spruce forest. Oecologia 74: 310-315.

Clements, M. A. and Ellyard, R. K. 1979. The symbiotic germination of Australian terrestrial orchids [*Pterostylis*, *Diuris*, *Thelymitra* inocultates with mycorrhizal fungi *Tulasnella* and *Ceratobandium*]. Amer. Orchid Soc. Bull.

Clymo, R. S. 1963. Ion exchange in *Sphagnum* and its relation to bog ecology. Ann. Bot. N. S. 27: 309-324.

Clymo, R. S. 1964. The origin of acidity in *Sphagnum* bogs. Bryologist 67: 427-431.

Coxson, D. S., McIntyre, D. D., and Vogel, H. J. 1992. Pulse release of sugars and polyols from canopy bryophytes in tropical montane rain forest (Guadeloupe, French West Indies). Biotropica, 121-133.

Craigie, J. S. and Maass, W. S. G. 1966. The cation-exchanger in *Sphagnum* spp. Ann. Bot. 30: 153-154.

Dainty, J. and Richter, C. 1993. Ion behavior in *Sphagnum* cell walls. Adv. Bryol. 5:107-128.

Döbbeler, P. 1997. Biodiversity of bryophilous Ascomycetes. Symposium on Mycology: Past, Present and Future, at British Mycological Society Symposium Sheffield, UK, Apr 1996. Biodiv. Conserv. 6: 721-738.

Duckett, J. G. and Read, D. J. 1995. Ericoid mycorrhizas and rhizoid-ascomycete associations in liverworts share the same

mycobiont: Isolation of the partners and resynthesis of the associations in vitro. New Phytol. 129: 439-447.

Duckett, J. G., Renzaglia, K. S., Pell, K., and Russell, A. 1989. The biology of underground organs of Jungermanniales. Bull. Brit. Bryol. Soc. 53: 19-21.

Duckett, J. G., Renzaglia, K. S., and Pell, K. 1991. A light and electron microscope study of rhizoid-Ascomycete associations and flagelliform axes in British hepatics with observations on the effects of the fungi on host morphology. New Phytol. 118: 233-257.

During, H. J. and Tooren, B. F. van. 1990. Bryophyte interactions with other plants. International Symposium on Bryophyte Ecology Edinburgh, UK, 19-22 July 1988. J. Linn. Soc. Bot. 104: 79-98.

Felix, H. 1988. Fungi on bryophytes, a review. Bot. Helv. 98: 239-269.

Foth, H. D. and Ellis, B. G. 1997. Soil Fertility, 2nd Ed. CRC Press, Boca Raton, Florida, 290 pp.

Francez, A. J. and Loiseau, P. 1999. Devenir de l'azote mineral dans une tourbiere a *Sphagnum fallax* Klinggr. et *Carex rostrata* Stokes du Massif central (France). [The fate of mineral nitrogen from peat (*Sphagnum fallax* Klinggr. and *Carex rostrata* Stokes) of the Massif central (France).]. Can. J. Bot. 77: 1136-1143.

Frey, W. and Kürschner, H. 1991. Das *Fossombronio-Gigaspermetum mouretii* in der Judäischen Wüste. 2. [The *Fossombronio-Gigaspermetum mouretii* in the Judean deserts. 2. Ecosociology and life strategy.]. Ökosoziologie und Lebensstrategien. Cryptog. Bot. 1/3: 73-84.

Galloway, C. M. and Black, C. C. 1989. Enzymes of sucrose metabolism in bryophytes. Bryologist 92: 95-97.

Gerdemann, J. W. 1968. Vesicular-arbuscular mycorrhiza and plant growth. Ann. Rev. Phytopathol. 6: 397-418.

Gjengedal, E. and Steinnes, E. 1990. Uptake of metal ions in moss from artificial precipitation. Environ. Monitor. Assess. 14: 77-87.

Glime, J. M., Wetzel, R. G., and Kennedy, B. J. 1982. The effects of bryophytes on succession from alkaline marsh to *Sphagnum* bog. Amer. Midl. Nat. 108: 209-223.

Grasso, S. M. and Scheirer, D. C. 1981. Scanning electron microscopic observations of a moss-fungus association. Bryologist 84: 348-350.

Gullvåg, B. M., Skaar, H., Ophus, E. M. 1974. An ultrastructural study of lead accumulation within leaves of *Rhytidiadelphus squarrosus* (Hedw.). Warnst. A comparison between experimental and environmental poisoning. J. Bryol. 8: 117-122.

Gundale, M. J. 2002. Influence of exotic earthworms on the soil organic horizon and the rare fern *Botrychium mormo*. Conserv. Biol. 16: 1555-1561.

Hájek, T. and Adamec, L. 2009. Mineral nutrient economy in competing species of *Sphagnum* mosses. Ecol. Res. 24: 291-302.

Harper, K. T. and Belnap, J. 2001. The influence of biological soil crusts on mineral uptake by associated vascular plants. J. Arid Environ. 47: 347-357.

Harper, K. T. and Pendleton, R. L. 1993. Cyanobacteria and cyanolichens: Can they enhance availability of essential minerals for higher plants? Great Basin Nat. 53: 59-72.

Harrison, M. J. 1999. Molecular and cellular aspects of the arbuscular mycorrhizal symbiosis. Ann. Rev. Plant Physiol. Plant Molec. Biol. 50: 361-389.

Hildebrand, R., Kottke, I., and Winkler, S. 1978. Untersuchungen über den Einfluβ von Pilzen auf die *p*H-Abhängigkeit von Laubmoosen. Beitr. Biol. Pflanzen 54: 1-12.

Iqbal, S. H., Nasim, G., and Shahjahan. 1988a. Vesicular-arbuscular mycorrhizal fungi associated with three mosses (*Sphagnum cymbifolium*, *Polytrichum commune*, and *Funaria hygrometrica*). Biologia (Lahore) 34: 269-273.

Iqbal, S. H., Nasim, G., and Jahan, S. 1988b. II. Vesicular-arbuscular mycorrhizal fungi associated with a bryophyte: *Marchantia palmata*. Biologia (Lahore) 34: 275-278.

Jakucs, E., Naár, Z., Szedlay, G., and Orbán, S. 2003. Glomalean and septate endophytic fungi in *Hypopterygium* mosses (Bryopsida). Cryptog. Mycol. 24(1): 27-37.

Jennings, O. E. 1918. Certain organic substances assimilated by *Ceratodon purpureus*. Bryologist 21: 86.

Kamal and Singh, C. S. 1970. Rhizosphere mycoflora of some bryophytes. Ann. Inst. Pasteur (Paris) 119: 752-755.

Kasuya, M. C. M., Masaka, K., and Igarashi, T. 1995. Mycorrhizae of *Monotropastrum globosum* growing in a *Fagus crenata* forest. Mycoscience 36: 461-464.

Khan, M. R., Imamual Huq, S. M., and Hasanuzzaman, M. 1997. Moss rhizosphere and its microflora. Bangladesh J. Bot. 26(2): 163-168.

Kilham, P. 1982. Acid precipitation: Its role in the alkalization of a lake in Michigan. Limnol. Oceanogr. 27: 856-867.

Klein, M. and Horst, W. J. 2005. Cation specific exchange capacity of cell wall material isolated from roots of plant species differing in Al resistance. Institute of Plant Nutrition, Department of Horticulture, University of Hannover, Germany. Accessed 6 June 2005 at <http://www.ipe.uni-hannover.de/publication/klein_poster1.pdf>.

Knight, A. H., Crooke, W. M., and Inkson, R. H. E. 1961. Cation-exchange capacities of tissues of higher and lower plants and their related uronic acid contents. Nature 192: 142-143.

Koedam, N. and Büscher, P. 1983. Studies on the possible role of cation exchange capacity in the soil preference of mosses. Plant Soil 70: 77-93.

Kottke, J., Krisch, T., and Winkler, S. 1976. Untersuchungen über den Einfluss von Pilzen auf die Konkurrenzfahigkeit von Moosen. Beitr. Biol. Pflanzen 51: 407-415.

Kottke, I., Beiter, A., Weiss, M., Haug, I., Oberwinkler, F., and Nebel, M. 2003. Heterobasidiomycetes form symbiotic associations with hepatics: Jungermanniales have sebacinoid mycobionts while *Aneura pinguis* (Metzgeriales) is associated with a *Tulasnella* species. Mycol. Res. 107: 957-968.

Leblond, S. 2004. Étude pluridisciplinaire du transfert des métaux de l'atmosphère vers les mousses (*Scleropodium purum* (Hedw.) Limpr.): Suivi sur un site rural (Vouzon, France). Thèse de Doctorat, Université Paris 7 – Denis Diderot, Paris, France, 214 pp.

Li, Y. and Glime, J. M. 1990. Growth and nutrient ecology of two *Sphagnum* species. Hikobia 10: 445-451.

Li, Y., Glime, J. M., and Liao, C.-L.. 1992. Responses of two interacting *Sphagnum* species to water level. J. Bryol. 17: 59-70.

Liao, C.-L. 1993 Chemical defence in bryophytes with high apparency. Bryol. Times 75: 1-4.

Ligrone, R. 1988. Ultrastructure of a fungal endophyte in *Phaeoceros laevis* (L.) Prosk. (Anthocerotophyta). Bot. Gaz. (Crawfordsville) 149: 92-100.

Ligrone, R. and Lopes, C. 1989. Cytology and development of a mycorrhiza-like infection in the gametophyte of

Conocephalum conicum (L.) Dum., Marchantiales, Hepatophyta. New Phytol. 111: 423-434.

Ligrone, R., Pocock, K., and Duckett, J. G. 1993. A comparative ultrastructural study of endophytic Basidiomycetes in the parasitic achlorophyllous hepatic *Cryptothallus mirabilis* and the closely allied photosynthetic species *Aneura pinguis* (Metzgeriales). Can. J. Bot. 71: 666-679.

Lou, Y., Cao, T., Zhang, Y., Zeng, Y., Cao, Y. 2013. A study of the characteristics of elements accumulation in tissue culture plants of the moss *Haplocladium microphyllum* under heavy metal stress by the SRXRF technique. Conference of the International Association of Bryologists, 15-19 July 2013 at Natural History Museum, London, UK.

Malmer, N., Horton, D. G., and Vitt, D. H. 1992. Element concentrations in mosses and surface waters of western Canadian mires relative to precipitation chemistry and hydrology. Ecography 15: 114-128.

Marschall, M. 2010. Photosynthetic responses, carbohydrate composition and invertase activity in fructan accumulating bryophytes (*Porella platyphylla* and *Sphagnum flexuosum*) under different environmental conditions (carbohydrate treatments, dark starvation, low temperature, desiccation). Acta Biol. Hung. 61(Supplement 1): 120-129.

Matsuda, Y., Okochi, S., Katayama, T., Yamada, A., and Ito, S. I. 2011. Mycorrhizal fungi associated with *Monotropastrum humile* (Ericaceae) in central Japan. Mycorrhiza 21: 569-576.

Maurel-Kermarrec, A., Pally, M., Fouiquier, L., and Hébrard, J. P. 1985. Cinétique de la fixation et de la désorption d'un mélange de cobalt 60, de chrome 51, de césium 137, de manganese 54 et de sodium 22 par *Chiloscyphus polyanthos* (L.) Corda in opiz. Cryptog. Bryol. Lichénol. 6: 359-377.

Mautsoe, P. J. and Beckett, R. P. 1996. A preliminary study of the factors affecting the kinetics of cadmium uptake by the liverwort *Dumortiera hirsuta*. S. Afr. J. Bot. 62: 332-336.

Mouvet, C. 1987. Accumulation et relargage de plomb, zinc, cadmium, chrome et cuivre par des mousses aquariques en mileu naturel et au laboratoire. Laboratoire d'Ecologie, Université de Metz, Metz.

Mues, R. and Zinsmeister, H. D. 1988. Chemotaxonomy of phenolic compounds in bryophytes. J. Hattori Bot. Lab. 64: 109-141.

Nieboer, E. and Richardson, D. H. S. 1980. The replacement of the nondescript term 'heavy metals' by a biologically and chemically significant classification of metal ions. Environ. Pollut. (Ser. B) 1: 3-26.

Nieboer, E. and Richardson, D. H. S. 1981. Lichens as Monitors of Atmospheric Deposition. Atmospheric Pollutants in Natural Waters, Ann Arbor Science Publishers, Inc., Ann Arbor, MI, pp. 339-388.

Núñez-Olivera, E., García-Álvaro, M., Beaucourt, N., and Martínez-Abaigar, J. 2001. Changes in element concentrations in aquatic bryophytes over an annual cycle. Arch. Hydrobiol. 152: 253-277.

Orebamjo, T. O. and Stewart, G. R. 1975. Ammonium repression of nitrate reductase formation in *Lemna minor* L. Planta 122: 27-36.

Parke, J. L. and Linderman, R. G. 1980. Association of vesicular-arbuscular mycorrhizal fungi with the moss *Funaria hygrometrica*. Can. J. Bot. 58: 1898-1904.

Peña, M. J., Darvill, A. G., Eberhard, S., York, W. S., and O'neill, M. A. 2008. Moss and liverwort xyloglucans contain galacturonic acid and are structurally distinct from the xyloglucans synthesized by hornworts and vascular plants. Glycobiology 18: 891-904.

Peña, M. J., Kong, Y., York, W. S., and O'Neill, M. A. 2012. A galacturonic acid-containing xyloglucan is involved in *Arabidopsis* root hair tip growth. Plant Cell 24: 4511-4524.

Pickering, D. C. and Puia, I. L. 1969. Mechanism for the uptake of zinc by *Fontinalis antipyretica*. Physiol. Plant. 22: 653-661.

Pirozynski, K. A. and Malloch, D. W. 1975. The origin of land plants: A matter of mycotrophism. Biosystems 6: 153-164.

Pocock, K. and Duckett, J. G. 1984. A comparative ultrastructural analysis of the fungal endophytes in *Cryptothallus mirabilis* Malm. and other British thalloid hepatics. J. Bryol. 13: 227-233.

Pocock, K. and Duckett, J. G. 1985a. The alternative mycorrhizas: Fungi and hepatics. Bull. Brit. Bryol. Soc. 45: 10-11.

Pocock, K. and Duckett, J. G. 1985b. On the occurrence of branched and swollen rhizoids in British hepatics: Their relationships with the substratum and associations with fungi. New Phytol. 99: 281-304.

Popper, Z. A. and Fry, S. C. 2003. Primary cell wall composition of bryophytes and charophytes. Ann. Bot. 91: 1-12.

Rabatin, S. C. 1980. The occurrence of the vesicular-arbuscular-mycorrhizal fungus *Glomus tenuis* with moss. Mycologia 72: 191-195.

Raspe, O. and De Sloover, J. R. 1998. Morphology, ecology and chorology of *Mniaecia jungermanniae* (Ascomycota) in Belgium and the significance of its association to leafy liverworts (Jungermanniales). Belgian J. Bot. 131:251-259.

Raven, J. A., Griffiths, H., Smith, E. C., and Vaughn, K. C. 1998. New perspectives in the biophysics and physiology of bryophytes. In: Bates, J. W., Ashton, N. W., and Duckett, J. G. (eds.). Bryology in the Twenty-first Century. Maney Publishing and the British Bryological Society, UK, pp. 261-275.

Read, D. J., Duckett, J. G., Francis, R., Ligrone, R., Russell, A., Newton, A. E., and Kenrick, P. 2000. Symbiotic fungal associations in 'lower' land plants. Philosoph. Trans. Roy. Soc. London B 355: 815-831.

Redecker, D., Kodner, R., and Graham, L. E. 2000. Glomalean fungi from the Ordovician. Science 289: 1920-1921.

Remy, W., Taylor, T. N., Hass, H., and Kerp, H. 1994. Four hundred-million-year-old vesicular arbuscular mycorrhizae. Proc. Natl. Acad. Sci. USA 91: 11841-11843.

Richter, C. and Dainty, J. 1989. Ion behavior in plant cell walls. I. Characterization of the *Sphagnum russowii* cell wall ion exchanger. Can. J. Bot. 67: 451-459.

Richter, C. and Dainty, J. 1990. Ion behaviour in plant cell walls. IV. Selective cation binding by *Sphagnum russowii* cell walls. Can. J. Bot. 68: 773-781.

Roche, S. A., Carter, R. J., Peakall, R., Smith, L. M., Whitehead, M. R., and Linde, C. C. 2010. A narrow group of monophyletic *Tulasnella* (Tulasnellaceae) symbiont lineages are associated with multiple species of *Chiloglottis* (Orchidaceae): Implications for orchid diversity. Amer. J. Bot. 97: 1313-1327.

Rühling, A. and Tyler, G. 1970. Sorption and retention of heavy metals in the woodland moss *Hylocomium splendens* (Hedw.) Br. et Sch. Oikos 21: 92-97.

Russell, J. and Bulman, S. 2004. The liverwort *Marchantia foliacea* forms a specialized symbiosis with arbuscular mycorrhizal fungi in the genus *Glomus*. New Phytol. 2004: 1-13.

Sabovljevic, A., Sabovljevic, M., Grubisic, D., and Konjevic, R. 2005. The effect of sugars on development of two moss

species (*Bryum argenteum* and *Atrichum undulatum*) during in vitro culture. Belgian J. Bot. 138: 79-84.

Satake, K. 2000. Iron accumulation on the cell wall of the aquatic moss *Drepanocladus fluitans* in an acid lake at *p*H 3.4-3.8. Hydrobiologia 433: 25-30.

Schöll, L. van, Kuyper, T. W., Smits, M. M., Landeweert, R., Hoffland, E., and Breemen, N. Van. 2008. Rock-eating mycorrhizas: Their role in plant nutrition and biogeochemical cycles. Plant Soil 303: 35-47.

Schüßler, A. 2000. *Glomus claroideum* forms an arbuscular mycorrhiza-like symbiosis with the hornwort *Anthoceros punctatus*. Mycorrhiza 10(1): 15-21.

Shacklette, H. T. 1965. Element content of bryophytes. Geol. Surv. Bull. 1198-D., U. S. Govt. Print. Off., Washington, D. C.

Shimwell, D. W. and Laurie, A. E. 1972. Lead and zinc contamination of vegetation in the southern Pennines. Environ. Pollut. 3: 291-301.

Singh, H. B. 1974. Rhizosphere fungal flora of bryophytes. Botanique 7: 131-136.

Spearing, A. M. 1972. Cation-exchange capacity and galacturonic acid content of several species of *Sphagnum* in Sandy Ridge Bog, central New York state. Bryologist 75: 154-158.

Sylvia, D. M., Furhmann, J. J., Hartel, P. G., and Zuberer, D. A. 2004. Principles and Applications of Soil Microbiology, 2nd ed. Prentice Hall, Englewood Cliffs, N. J.

Syrett, P. J. and Morris, I. 1963. The inhibition of nitrate assimilation by ammonium in *Chlorella*. Biochim. Biophys. Acta – Specialized Section on Enzymological Subjects 67: 566-575.

Taylor, F. G. and Witherspoon, J. P. 1972. Retention of simulated fallout particles by lichens and mosses. Health Phys. 23: 867-869.

Temple, P. J., McLaughlin, D. L., Linzon, S. N., and Wills, R. 1981. Moss bags as monitors of atmospheric deposition. J. Air Pollut. Control Assoc. 31: 668-670.

Trachtenberg, S. and Zamski, E. 1979. The apoplastic conduction of water in *Polytrichum juniperinum* Willd. gametophytes. New Phytol. 83: 49-52.

Turner, B. L., Baxter, R., Ellwood, N. T. W., and Whitton, B. A. 2003. Seasonal phosphatase activities of mosses from Upper Teesdale, northern England. J. Bryol. 25: 189-200.

Varma, A. and Hock, B. 1999. Mycorrhiza. Springer Verlag, Berlin.

Vázquez Castro, M. D., López, J., and Carballeira, A. 1999. Uptake of heavy metals to the extracellular and intracellular compartments in three species of aquatic bryophytes. Ecotoxicol. Environ. Safety Environ. Res. B 44: 12-24.

Vujičić, M., Sabovljević, A., and Sabovljević, M. 2009. Axenically culturing the bryophytes: a case study of the moss *Dicranum scoparium* Hedw. (Dicranaceae, Bryophyta). Bot. Serbica 33: 137-140.

Wanek, W. and Pörtl, K. 2008. Short-term ^{15}N uptake kinetics and nitrogen nutrition of bryophytes in a lowland rainforest, Costa Rica. Funct. Plant Biol.: 35: 51-62.

Webster, H. J. 1985. Elemental analyses of *Pohlia nutans* growing on coal seeps in Pennsylvania. J. Hattori Bot. Lab. 58: 207-224.

Weetman, G. F. and Timmer, V. 1967. Feather moss growth and nutrient content under upland black spruce. Woodlands Research Index, Pulp and Paper Research Institute of Canada, Pointe Claire, P. Q., Canada 138: 1-38.

Wells, J. M. 1988. The role of the cell wall in metal uptake, redistribution and tolerance in the moss *Rhytidiadelphus squarrosus.* Ph. D. Thesis, University of Bristol, Bristol.

Wells, J. M. and Boddy, L. 1995. Phosphorus translocation by saprotrophic basidiomycete mycelial cord systems on the floor of a mixed deciduous woodland. Mycol. Res. 99: 977-980.

Wells, J. M. and Brown, D. H. 1987. Factors affecting the kinetics of intra- and extracellular cadmium uptake by the moss *Rhytidiadelphus squarrosus*. New Phytol. 105: 123-137.

Wells, J. M. and Brown, D. H. 1990. Ionic control of intracellular and extracellular Cd uptake by the moss *Rhytidiadelphus squarrosus* (Hedw.) Warnst. New Phytol. 116: 541-553.

Wells, J. M. and Richardson, D. H. S. 1985. Anion accumulation by the moss *Hylocomium splendens*: Uptake and competition studies involving arsenate, selenate, selenite, phosphate, sulphate and sulphite. New Phytol. 101: 571-583.

Wiehle, W. 1988. *Cryptothallus mirabilis* – ein mykotrophes Lebermoos. Boletus 12: 15-22.

Wiersum, L. K. and Bakema, K. 1959. Competitive adaptation of the cation exchange capacity of roots. Plant Soil 11: 287-292.

Williams, P. G., Roser, D. J., and Seppelt, R. D. 1994. Mycorrhizas of hepatics in continental Antarctica. Mycol. Res. 98: 34-36.

Wojtun, B. 1994. Element contents of *Sphagnum* mosses of peat bogs of Lower Silesia (Poland). Bryologist 97: 284-295.

Yamada, A., Kitamura, D., Setoguchi, M., Matsuda, Y., Hashimoto, Y., Matsushita, N., and Fukuda, M. 2008. *Monotropastrum humile* var. *humile* is associated with diverse ectomycorrhizal Russulaceae fungi in Japanese forests. Ecol. Res. 23: 983-993.

Zobel, M., Pilt, I., Moora, M., Partel, M., and Liira, J. 1999. Small-scale dynamics of plant communities in an experimentally polluted and fungicide-treated subarctic birch-pine forest. Acta Oecol. 20(1): 29-37.