

# Estimating moss growth in arctic conditions: a comparison of three methods

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**ABSTRACT.** Except for *Sphagnum* mosses of peatland habitats, reliable methods to assess moss productivity in arctic or boreal biomes give usually highly variable results. Therefore, ecosystem processes are poorly understood in these biomes where mosses are an important component of the system. The aim of this study was to compare three methods to estimate moss growth in polygon patterned fens: cranked wires, natural markers and artificial white marks (an alternative to the spray method). Precision of estimates was significantly higher when natural markers were used (coefficients of variation, CV, between 17 and 27%), compared to cranked wires (CV=37%) or white marks (CV=56%). Natural markers also provided estimates for growth of moss stems 32 to 113% higher than the other methods. Although cranked wires were calibrated shortly after snowmelt, some moss growth is still missed and consequently moss growth is underestimated. Accuracy of cranked wires was poor, mainly caused by frost heaving or permafrost activities that can affect wire position. Thus, this method should be avoided in arctic ecosystems. Even if white marks were painted on moss stems at the end of the growing season prior to the sampling year, lower estimates of moss growth were still found. We suspect some interference with moss growth processes during the marking process, at least when used with brown mosses. The natural marker method, which provides increment for an entire growing season, appears to be the most accurate method of the three. Additionally, it is also the easiest and the least time consuming method to use. Its main drawback is that relatively few species have natural growth marks and these species may not always be present among the targeted species under study. Also, measurements of stem growth on the same sample did not differ between observers, even if the second measurement was done 12 years later. In conclusion, when species with natural markers are present, this method should be used to assess moss growth. For arctic/sub-arctic studies where such species are lacking, the artificial white marks method should be refined further.

**KEYWORDS.** Brown mosses, Amblystegiaceae, Meesiaceae, cranked wires, moss elongation, moss primary production, Natural markers, *Polytrichum*, White mark method.

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DOI: 10.1639/0007-2745-113.2.322



Bryophytes are an important component of arctic ecosystems in terms of primary production, phytomass and species diversity. They are often the dominant plant group, especially in wet habitats like polygon patterned fens (Vitt & Pakarinen 1977; Russell 1990; Ellis & Rochefort 2004). Hence, moss growth and productivity are key parameters when studying arctic ecosystem processes (Gauthier et al. 2004). Techniques used to measure bryophyte productivity can be classified into two main categories: biomass harvesting techniques and gas exchange techniques (Russell 1988). Gas exchange technique consists of taking instantaneous measures of photosynthesis and respiration. CO<sub>2</sub> exchange relationships with photosynthetically active radiation (PAR), air temperature, peat temperature and water table are used for modelling net ecosystem production over a season (Moore & Roulet 1991). However, it is an expensive technique requiring unwieldy electronic equipment that is not suitable for a remote arctic field location. This method needs complex calibration and many replicates during the growing season.

Direct techniques of biomass harvesting consist in taking biomass samples at different time periods. Weight difference between two samples corresponds to primary production for the given period. In fragile ecosystems, direct techniques are not desirable because of the associated disturbances and slow recovery. Indirect techniques of biomass harvesting consist of measuring increments of moss stems for an interval of time. These increments are associated with other parameters (e.g. weight, stem density, etc.) and are used to estimate productivity. Indirect methods are thus recommended and frequently used under such conditions. Three indirect methods, cranked wires (Clymo 1970), tied threads (Tallis 1959), and fluorescent spray (Russell 1984) are recommended by the International Tundra Experiment (ITEX) to estimate bryophyte stem growth in arctic ecosystems (Jónsdóttir et al. 1997), but their efficiency has not been evaluated. In peatlands of Southern Quebec (Canada), Poulin (1995) compared three methods to estimate bryophyte increments in *Sphagnum* carpets

(i.e., cranked wires, several fluorescent sprays, and plastic bands; Lindholm 1990) and found large differences among techniques. It is thus necessary to identify a reliable method to estimate moss stem growth and moss productivity under a range of field conditions. This is essential to allow comparisons among sites and studies, particularly for small brown mosses.

The method chosen to estimate moss stem increments in arctic ecosystems must take several abiotic and biotic factors into account. The biotic constraints specific to arctic mosses are their low annual net primary production and the fact that individual stems are small, fragile, and delicate (Haag 1974; Muc 1977). The short growing seasons (e.g., 50 days on Truelove Lowland, Devon Island, Nunavut, Muc 1977) create an abiotic constraint, meaning the set up timing for experiments is critical to avoid missing part of the moss annual growth. Furthermore, some bryophytes are able to photosynthesize under the snow (e.g. *Drepanocladus uncinatus* [Collins & Callaghan 1980] or *Polytrichum sexangulare* [Lösch et al. 1983]) or immediately after snow melt (*Kiaeria starkei*; Woolgrove & Woodin 1996). Thus, to be unbiased, methods should be able to capture whole seasonal growth increments, from snowmelt to the end of the growing season.

We combined data from four different studies carried out at an arctic site (Bylot Island) to assess the accuracy and precision of three methods to estimate stem growth of brown mosses. These three techniques were the cranked wires, the natural marker method, which involved moss species with visible annual growth segments, and an alternative to spray methods, which involved the painting of tiny white marks on individual stems (hereafter called the white mark technique). We further examined variation among researchers measuring moss stems with the natural marker technique.

## METHODS

**Study area.** Field work was carried out in a glacial valley (70 km<sup>2</sup>) of Bylot Island, Nunavut, Canada (73°08' N–80°00' W; Gauthier et al. 1996).

**Table 1.** Summary of data sets used to compare three methods of moss elongation measurements on Bylot Island. Numbers in method columns correspond to the mean number of measured stems [range of values] for each experimental unit.

Data set	Year of sampling	Number of experimental units	Dimension of experimental units	Method used			Reference
				Cranked wires	Natural markers	White marks	
1	1996	28	Diameter of 0.6 m	18 [18–18]	18 [4–27]	-	Pineau 1999
2	2004	84	2 m × 2 m	-	26 [3–49]	89 [43–140] <sup>1</sup>	Pouliot 2006
3	2007	168	1 m × 2 m	-	25 [4–60]	22 [3–89] <sup>2</sup>	Marchand-Roy 2009
4A	1995	17	4 m × 4 m	-	28 [8–44]	-	Gauthier et al. 2004
4B	1996	17	4 m × 4 m	-	20 [11–29]	-	Gauthier et al. 2004
4C	1997	17	4 m × 4 m	-	23 [17–31]	-	Gauthier et al. 2004
4D	1998	18	4 m × 4 m	-	22 [6–35]	-	Gauthier et al. 2004
4E	2004	35	2 m × 2 m	-	15 [5–26]	-	Gauthier et al. 2004

<sup>1</sup> Application of white marks was done in early summer, after spring run-off.

<sup>2</sup> Application of white marks was done the previous summer at the end of the growing season.

Lowlands are characterized by wet polygon patterned fens, typically ranging from 10 to 20 m in diameter. Most of them are concaves and form freshwater fens or shallow ponds. Fen and pond margins are covered by graminoids such as *Dupontia fisheri*, *Eriophorum scheuchzeri* and *Carex aquatilis* var *stans* growing through a dense and continuous carpet of mosses dominated by *Scorpidium cossonii*, *Campylium stellatum* var. *arcticum*, *Calliargon giganteum*, *Cinclidium arcticum*, *Bryum cyclophyllum*, *Aulacomnium palustre* and *Polytrichum swartzii*. Plant nomenclature follows USDA plant database (2009).

#### **Methods used to estimate moss stem growth.**

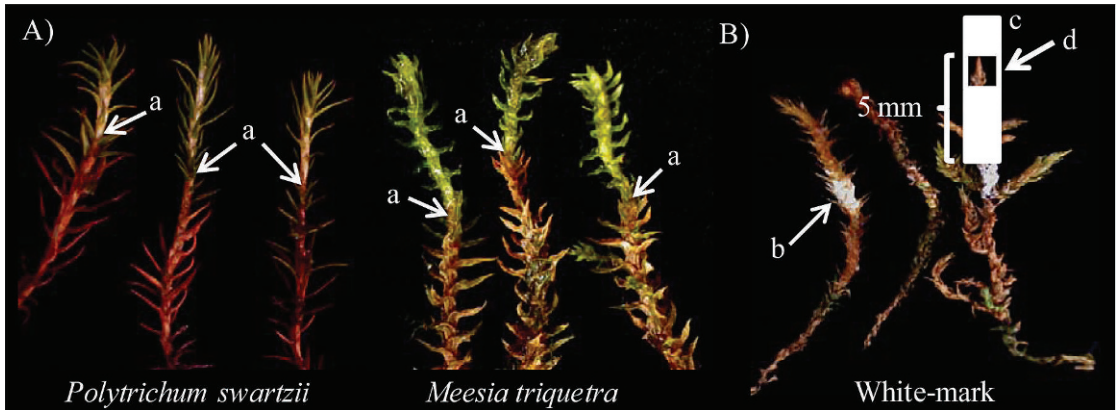
We used four different data sets collected on Bylot Island during as many studies aimed at evaluating the effect of nutrient addition or goose grazing on moss communities (Table 1). In all data sets, the natural marker method was used and in three of them one of the other two methods (cranked wire or white marks) was tested against the former. Only the natural marker method was used in the fourth study; this data set was solely used to evaluate variability among observers in stem measurement. Data set 4 was collected in permanent experimental units (4 × 4 m), but only a part of these units was sampled each year (not always the same).

The use of cranked wires was a method developed by Clymo (1970) for mosses that grow upright. This method was judged suitable for arctic mosses because their shoots usually grow upright in

dense carpets. We used smaller cranked wires than the original design (5 cm instead 10 cm long under the moss surface) to ensure that wires were always above permafrost during the growing season.

Eighteen cranked wires were systematically put in each experimental unit and calibrated shortly after snowmelt (end of June, Table 1). Calibration consisted in measuring the distance from the moss surface to the cranked wire's top (M1). The same distance was measured at the end of the growing season (M2; mid-August) and increase in moss length was the difference between the two measures (M2-M1).

In highly seasonal environments, such as the Arctic, several moss species show visible annual growth segments (Clarke et al. 1971; Vitt & Pakarinen 1977), which can be used as a natural marker. On Bylot Island, *Polytrichum swartzii* and *Meesia triquetra* have clear seasonal differences in leaf size and spacing (Fig. 1A). As the growing season cools down, the growth of these mosses slows down, resulting in smaller leaves closer to each other on the stem. In contrast, at the peak of the growing season, leaves are longer and show a wider spacing between insertion points as the stem grows faster (Vitt & Pakarinen 1977). Annual growth of *P. swartzii* or *M. triquetra* was thus estimated by measuring the distance between the top of a stem and the separation zone between widely and tightly spaced leaves. Our objective was to harvest at least 15 stems of *P. swartzii* or *M. triquetra* in each experimental unit in mid-



**Figure 1.** Illustrations of two of the methods used to measure moss stem growth. **A.** Natural markers. **B.** White marks. a = starting point of the annual growth segment, b = white mark, c = plastic straw, and d = window located at 5 mm from the bottom of a plastic straw.

August (end of the growing season at our study site) to measure increases in length to an instrument precision of  $\pm 0.01$  mm using the graded scale of a stereomicroscope (data set 1) or a digital caliper under the stereomicroscope (data sets 2 and 3; **Table 1**) at  $6.4\times$ . For data sets 4A to 4E, stems were measured at each sampling by a different observer each year (referred to as Observer A in **Table 1**). Samples were then pressed, dried and stored, and in 2007 all stems were re-measured by the same observer in our laboratory at Laval University (Observer B). Increments were originally measured with a ruler (data sets 4A to 4D) or a digital caliper (data set 4E; Observer A in **Table 1**) but Observer B used a digital caliper under a stereomicroscope for all re-measurements in 2007.

The white mark technique (used successfully on hollow *Sphagnum* species and floating mosses, Ilomets 1982) consisted in marking brown moss stems in each experimental unit using white insoluble oil-based paint (Painty® of ZIG®, manufactured by Kuretaka Co., Ltd.; **Table 1**). For data set 2, paint was applied after spring run off resulting from snowmelt (end of June, same year of sampling). For data set 3, paint was applied the year before sampling (mid-August 2006, end of growing season) to ensure a complete measurement of the growth increment during the next season. In order to facilitate the finding of marks at the end of the summer, marks were concentrated in four quadrats ( $\sim 10 \times 10$  cm) randomly placed in each experimental unit. Moss

samples within quadrats were gently removed from the moss carpet, marks were applied in the field and samples were put back to their initial position at the same level than surrounding mosses. This manipulation was not visible a few weeks later. Around 160 (data set 2) or 40 (data set 3) marks were applied on stems at 5 mm below moss tip in each experimental unit, i.e. below the apical bud, to hopefully avoid disturbing the growth process (**Fig. 1B**). Species were marked proportionally to their abundance in the data set 2 (70.7% of marks were made on *Scorpidium* sp., other marks were made on nine different species; Pouliot 2006) and mainly *Scorpidium* sp. was marked in the data set 3 (also a small amount of *C. stellatum* var. *arcticum* and *C. giganteum* was marked; Marchand-Roy 2009). To paint marks, individual bryophyte stems were inserted in a plastic straw that was pushed down until a window located at the 5 mm reference level was in line with the moss tip. Marks were applied just below the straw with a fine hair brush, (**Fig. 1B**). In mid-August, marked mosses were collected and distance between the mark top and the moss tip was measured with a digital caliper (instrument precision of  $\pm 0.01$  mm), and subsequently subtracting the initial 5 mm. To be sure that the initial 5 mm was accurate, we marked an additional 50 stems and we measured the distance between the mark and the moss tip immediately after marking.

**Statistical analyses.** Coefficients of variation (CV) of mean stem growth measurements obtained

**Table 2.** Estimates of moss stem elongation (in mm; mean  $\pm$  SD) and coefficients of variation (CV, calculated within each experimental unit, in %; mean  $\pm$  SD) for different methods of measurement or different observers. Data sets are resumed in Table 1. Sample size corresponds to the number of experimental units in Table 1.

Data set	Method								
	Cranked wires		Natural markers				White marks		
	Elongation	CV	Observer A <sup>1</sup>		Observer B <sup>2</sup>		Elongation	CV	
		Elongation	CV	Elongation	CV	Elongation	CV	Elongation	CV
1	12.3 $\pm$ 6.6	36.6 $\pm$ 18.7	16.3 $\pm$ 5.1	17.2 $\pm$ 5.8	-	-	-	-	-
2	-	-	10.4 $\pm$ 2.4	21.2 $\pm$ 0.4	-	-	4.9 $\pm$ 3.4	55.9 $\pm$ 13.3	
3	-	-	7.1 $\pm$ 1.7	27.1 $\pm$ 0.6	-	-	4.6 $\pm$ 1.7	56.2 $\pm$ 17.2	
4A	-	-	5.6 $\pm$ 1.5	30.8 $\pm$ 7.4	6.4 $\pm$ 1.7	25.2 $\pm$ 6.2	-	-	
4B	-	-	8.0 $\pm$ 3.4	23.9 $\pm$ 6.9	9.2 $\pm$ 3.2	18.1 $\pm$ 6.7	-	-	
4C	-	-	11.0 $\pm$ 4.2	23.5 $\pm$ 8.4	10.4 $\pm$ 3.6	21.0 $\pm$ 7.3	-	-	
4D	-	-	13.3 $\pm$ 6.9	20.6 $\pm$ 11.8	14.2 $\pm$ 6.4	19.4 $\pm$ 9.8	-	-	
4E	-	-	9.0 $\pm$ 2.5	27.1 $\pm$ 7.2	8.9 $\pm$ 2.0	21.8 $\pm$ 7.4	-	-	

<sup>1</sup> For data sets 4A to 4E: a different observer made the measurements each year

<sup>2</sup> For data sets 4A to 4E: the same observer remeasured all stems in 2007

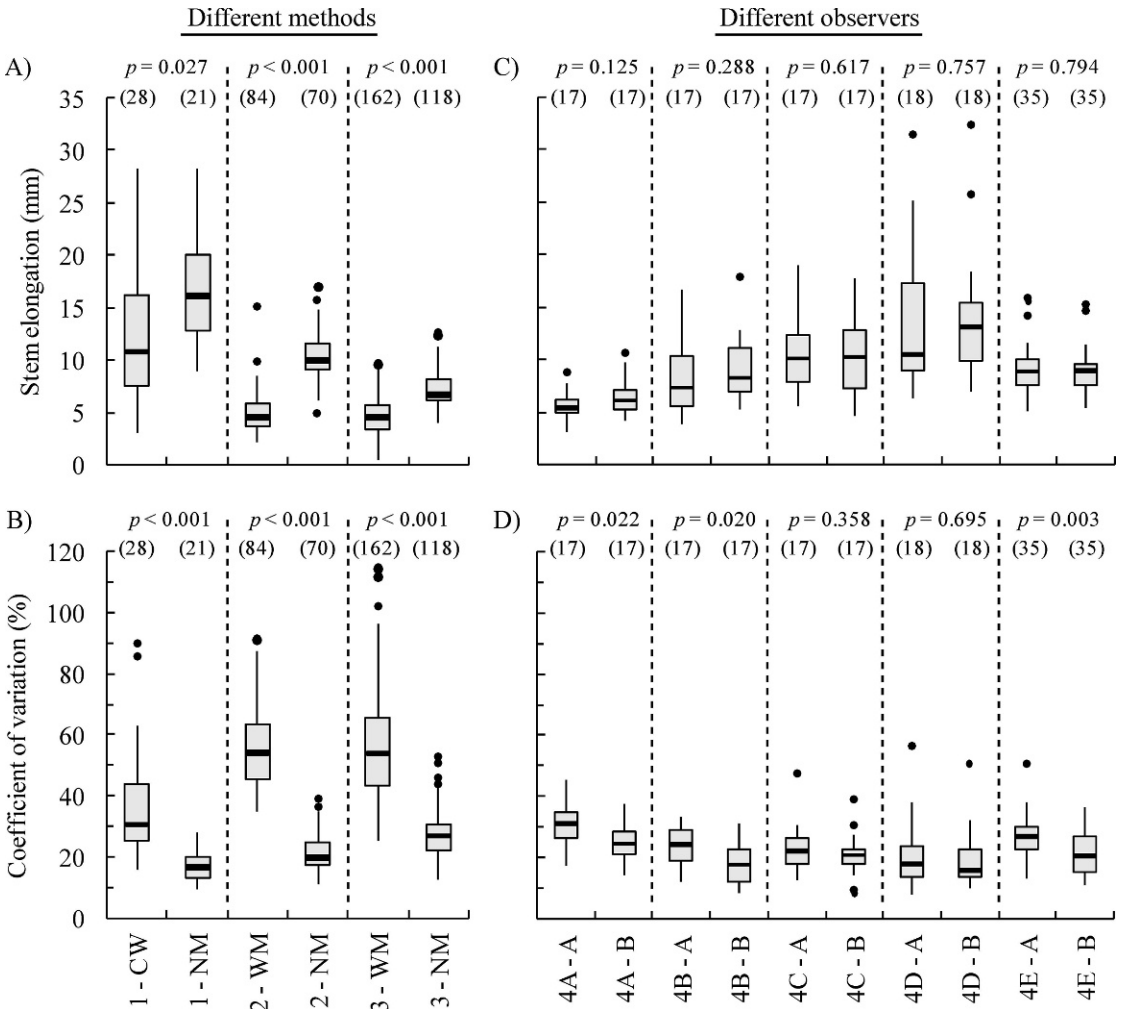
in each experimental unit were calculated to estimate precision. Student's t-tests were performed to compare moss growth and coefficients of variation estimated by different methods used in the same year on the same data set (cranked wires vs natural markers and white mark technique vs natural markers) or for different observers (sample size here was the number of experimental units; **Table 1**). Homogeneity and normality of variance were respected in all cases. All analyses were conducted using SAS Software (SAS Institute 2003).

## RESULTS

**Potential species bias.** In 2004 (data set 2), there was no significant difference between the moss stem growth of *P. swartzii* and *M. triquetra*, the two species used as innate markers ( $P = 0.91$ ). Data were thus pooled before subsequent analyses involving innate markers. We considered all species with white marks together in subsequent analyses relating to white mark method for the following reasons. Firstly, for the white mark method, we compared moss stem growth of the less frequent species individually with *Scorpidium* sp. values (73% dominance), and we found a significant difference for 4 species out of 10 (*Aulacomnium turgidum*, *Brachythecium turgidum*, *Bryum cyclophyllum* and *Cinclidium arcticum*;  $P \leq 0.04$ ). These 4 species accounted for only 9.1% of the

moss carpet composition (Pouliot 2006). Secondly, no significant difference was found for the moss stem growth between *Scorpidium* sp. and species known to have innate markers but painted by natural occurrence with white marks for this direct comparison (*Polytrichum swartzii* and *M. triquetra*;  $P = 0.21$ ). Then, when we compared *Scorpidium* sp. stem growth against the other species (pooled) with white marks, no significant difference was found ( $P = 0.59$ ). Also, we did not do these comparisons in 2007 (data set 3) as only a small amount of two other species was marked whereas no significant difference with *Scorpidium* sp. was detected in 2004 (*C. stellatum* var. *arcticum*,  $P = 0.33$ ; *C. giganteum*,  $P = 0.09$ ). Finally, for a general estimate of moss growth or production, one will generally consider all species together or the growth of the dominant species (or group of dominant species).

**Method comparisons.** Estimates of moss stem growth obtained with the natural marker method were significantly higher than other methods used in all comparisons (**Table 2**; **Fig. 2A**). Mean stem growth measured with natural markers was 1.3 times higher than that measured with cranked wires and 1.5 to 2.1 times higher than that measured with white marks. Also, moss stem growth for natural marker species with white marks was lower than the growth estimated with the annual growth segment (in data



**Figure 2.** Variation in moss stem growth and precision (coefficients of variation). **A** and **B.** Comparison of different measurement methods used in the same year (CW = cranked wires, WM = white mark technique, NM = natural markers). **C** and **D.** Comparison of two different observers measuring the same samples. Number before method or observer on horizontal axis corresponds to data set as described in **Table 1**. The thick line in the box-plot represents the median, ends of box-plots represent 1<sup>st</sup> and 3<sup>rd</sup> quartiles and black circles represent extreme values. Number in parentheses refers to *n* for the box-plot just below.

set 2;  $P < 0.01$ ). Increment values were quite variable (large box-plots, **Fig. 2**), but this is due in part to various fertilization treatments applied to different experimental units. However, fertilization treatments affected moss growth in the same way regardless of the measurement method (Pouliot 2006). Natural markers also showed a precision significantly higher than other methods as shown by the coefficients of variation, which were 2.1 to 2.6 times lower (**Table 2**; **Fig. 2B**). Accuracy of the initial 5 mm in the white mark method was good (mean  $\pm$  SD:  $5.3 \pm 0.4$  and coefficient of variation: 8.2%).

**Observer’s effect.** No significant difference was found in estimates of moss stem growth measured by different observers for all comparisons (**Table 2**; **Fig. 2C**). However, there were significant differences in precision between observers for three of the five data sets (4A, 4B and 4E). Coefficients of variation were about 1.3 times lower when re-measured with a digital caliper under stereomicroscope (Observer B) compared to the original measures taken during the sampling year (Observer A) with a ruler or a digital caliper without stereomicroscope (**Table 2**; **Fig. 2D**).

## DISCUSSION

**Comparisons between methods.** Natural markers have often been used to measure growth increments of mosses (e.g., Clarke et al. 1971; Potter et al. 1995; Camill et al. 2001; Gauthier et al. 2004) or to assess the accuracy of increment method (innate time marker, Rochefort et al. 1990). Seasonal demarcations are easy to detect in species that show such growth patterns and thus this method is likely to be relatively free of measurement errors. This may explain why the coefficient of variation was lowest with this method. This technique is a good choice when species with seasonal demarcation in growth are present, as in the Arctic. The natural marker method is also quick, simple, requires few manipulations and is relatively non-destructive because only few stems need to be collected. It can be carried out repeatedly on the same experimental unit over several years (Gauthier et al. 2004) without disturbing the natural environment. In some instances, several yearly marks can be visible, thereby allowing productivity estimation for a number of prior growing seasons.

Could the higher stem increment found with natural marker be due to the fact that measurements were made on only two moss species compared to several species for the other methods? We believe not, because in polygon patterned fens with dense moss carpets, growth is relatively homogenous among species. By growing at the same level, moss stems protect each other against high evapotranspiration rates by reducing air resistance, increasing boundary layer thickness and providing capillary spaces for water retention (Glime 2007). So, we assume that moss canopies of mixed species extend upward at an even rate and species with natural markers can be used to extrapolate growth of species without these markers (Vitt 2007). We suggest that the higher estimates of moss growth with natural markers are because cranked wires or white marks (only for data set 2) did not estimate the whole annual stem growth. In addition, species with natural markers (i.e., *M. triquetra* and *P. swartzii*) are relatively scarce and not ubiquitous at our study site (Pouliot 2006). In fact, 25% (data set 1), 17% (data set 2) and 30% (data set 3) of our experimental units did not contain species with natural markers. Moreover, because

moss primary production can vary according to microtopography (Johnson & Damman 1993; Rydin 1993), stems with natural markers have to be harvested close to moss samples used to calculate other parameters involved in production estimate. But when present, species with natural marks of growth are likely to be the best method to estimate moss growth and should be favoured.

Russell (1988) showed that artificial fluorescent markers can successfully estimate growth of patches of small moss species. However, Poulin (1995) demonstrated that several types of dyes or fluorescent brighteners are partially leached out when used on aquatic mosses or in wet habitat. The marks also spread along the stem, they do not remain fixed as the white paint used in our trials and their use is definitively discouraged. Even if we recovered only around 50% of marked stems at the end of the summer, our oil-based marker did not leach and marks had withstood water immersion. In our opinion, the failure to find all the marked stems is due to the dense moss carpet rather than to the painted marks leaching because marks were as visible after several months under water as when they were applied. Marks also persisted well when mosses were dried, transported to the laboratory, and then re-wetted to measure increments. In contrast to natural markers, the white mark method allows estimation of stem growth directly on dominant moss species of wet habitat (*Scorpidium* ssp. in our case), which have no annual growth segment.

Nevertheless, estimates of stem growth were lower and more variable with the white mark technique compared to natural markers. In the first year that we tested the white mark method (data set 2), marks were made only after spring runoff for logistic reasons. Thus, part of the estimate of stem growth was lost because mosses are able to grow as soon as the snow melts and, for some species, even before (Collins & Callaghan 1980; Löscher et al. 1983; Woolgrove & Woodin 1996). However, estimates of moss growth were still lower than those obtained with natural markers when marks were applied at the end of the previous summer (data set 3). Other studies using similar procedures to evaluate moss increments (with fluorescent markers or dyes, for example) generally spread the marking substance on

the moss surface, potentially interfering with photosynthetic processes (Russell 1988). We tried to avoid interference with the activity of the meristematic apical cell of mosses by marking 5 mm below the tip of the stem. However, we suspect that the paint may have negatively affected moss growth. Mark application could create a mechanical injury, which could cause death of individual stems. This could explain why we did not find all the marked stems at the end of summer. Moreover, since mosses can absorb nutrients from the environment directly through their cell walls (Brown & Bates 1990), applying a waterproof mark could disrupt absorption processes and then, slow down growth. Also, *Scorpidium* species often have some ramifications. By marking one of those, it is possible that the plant concentrates growth energy on another ramification that is not affected by the paint mark. These points may explain why the coefficients of variation were relatively high for the method since some stems may have had a vertical growth close to zero because of injuries or ramification's growth. The 5 mm starting point of the white mark method was precise (CV = 8.17%), hence the lack of accuracy found with this method cannot be explained by the nature of the marker used. One should, however, also take into account that accurate marking of numerous stems at a standard level below the apical moss tip is time consuming, as well as the time to measure them again at the end of the summer. We conclude that in this present form, the white mark method is not suitable for estimating moss growth in arctic wet environments but deserves further investigation because of its versatility. This method has proved to be excellent for hollow *Sphagnum* species and floating mosses in pools (Ilomets 1982). Consequently, further studies should be made to test the effect of mark widths on moss growth to see if marks will interfere with absorption or photosynthetic processes.

Marking with cranked wires, as developed by Clymo (1970) or with modified wires (Gunnarsson & Rydin 2000), is the most frequently used method to measure moss stem growth in peatlands (e.g. Rochefort et al. 1990; Moore et al. 2002; Chapin et al. 2004; Vitt et al. 2003). It is recommended when the substrate is stable and flat and when moss stems are

erect. Modified wires are better anchored into moss carpets than their original version because of the additional horizontal part at the base of each wire. As the white mark method, cranked wires showed an important variability (coefficients of variation more than twice as high as for natural markers) and underestimated stem growth. Because of their intrinsic variability, indirect measurement methods of moss growth, like cranked wires, are not well adapted to species with small annual increments (Russell 1988) such as arctic brown mosses. Furthermore, cranked wires were originally designed for *Sphagnum* mosses, which have faster growth rates than arctic brown mosses (Clymo 1970).

Performance of methods requiring foreign material, like cranked wires inserted into the moss carpet, may be negatively affected by the presence of permafrost or frequent surface disturbances caused by flooding or ground subsidence due to permafrost melting. In our case, cranked wires had to be inserted in the moss carpet when experimental units were still partly flooded in spring. At that time of the season, cranked wires cannot be inserted very deep because the permafrost front is still close to the surface. Water movement or freezing and thawing cycles may cause upward moves of cranked wire or the wires may tilt. This may explain why the moss growth was underestimated. Lindholm and Vasander (1990) suggested that, for moss species with small annual increments, indirect measurement markers should be set the year before sampling to avoid interference with moss growth. However, cranked wires would then be subjected to several freeze/thaw cycles, leading to considerable movements of wires and thus even larger biases. For these reasons, we recommend to avoid the cranked wire method in arctic wet habitats like polygon patterned fens.

**Comparisons between observers.** In long-term studies using the natural marker method, the need to measure annual growth increments by different observers frequently arises. We showed that this should not be a problem because measurement of stem growth on the same sample did not differ between observers. This was true even for samples that were re-measured several years later. For this purpose, however, mosses with natural markers need to be well pressed, dried and kept under ideal



conditions such as in a herbarium. In this study, we were able to correctly remeasure moss stems 12 years after sampling. Even though the coefficient of variation was slightly lower for the observer that made the second set of measurements, we believe that this discrepancy is likely due to a difference in the technique used for measurements. Indeed, Observer B used a digital caliper under a stereomicroscope, unlike the previous observers, and this resulted in the lowest coefficient of variation (from 18 to 25%).

**Alternative methods not considered in this study.** Some common techniques used to estimate moss stem growth or primary production were not considered here because they were judged inappropriate for arctic polygon patterned fens. Other indirect methods include tags or velcro wrap around moss stems (Raeymaekers & Glime 1986; Rochefort & Vitt 1988; Li & Glime 1990) or stems cut to a reference level (Clymo 1970; van der Heijden et al. 2000; Pearce et al. 2003) but they were considered too invasive for arctic environments dominated by brown mosses with small and fragile stems. Furthermore, the disturbance created by isolating an individual stem to apply the markers can interfere with water conduction and growth potential (Russell 1988). Another method, autoradiography of tissues with  $^{14}\text{C}$  or  $^{14}\text{CO}_2$  (e.g., Wallén et al. 1988; Aerts et al. 2001), could be a good alternative to estimate moss stem growth in polygon fens. It is a relatively inexpensive method (assuming an easy access to radioactivity reading equipment) and can be easily adapted to any type of field sites (Russell & Botha 1988). However, releasing radioactive material in the environment, even in small amounts, may not be desirable or even possible in protected areas such our study site in Similirk National Park.

**Conclusion.** The natural marker method proved to be the most accurate of the three methods tested and thus should be preferred to other methods at arctic study sites when the target genera (e.g. *Polytrichum* or *Meesia* genus) are relatively frequent in the community and homogeneously dispersed. In general, natural markers within mixed moss carpets grow at the same rate than other species and can be used as surrogate for the growth of the others. Also, natural markers are appropriate to estimate seasonal growth under a wide range of climatic conditions and

a single visit in the experimental site at the end of the growing season is sufficient to obtain all the measurements needed. However, when species with natural markers are absent or too sparse, none of the other method tested will provide an unbiased estimate of moss stem growth. Although the white mark method could still be used under certain circumstances (e.g. when one is interested in the relative response of moss species to specific treatments such as fertilization), it still cannot provide an unbiased measure of moss productivity at the ecosystem level. Further tests are required to determine the source of the biases associated with this method before it can be more widely used. For example, the effect of the paint on up and down movement of water into mosses or potential of injuries could be studied.

#### ACKNOWLEDGMENTS

These studies were funded by the Fonds québécois de recherches sur la nature et les technologies, Ducks Unlimited, the Natural Science and Engineering Research Council of Canada, the Arctic Goose Joint Venture of the Canadian Wildlife Service, the Department of Indian Affairs and Northern Development, ArcticNet, and the Centre d'Études Nordiques of Université Laval. We also received generous logistical support from the Polar Continental Shelf Project (project 643-04) and we thank the Pond Inlet Hunter and Trapper Association and Parks Canada for allowing us to work on Bylot Island. We are grateful to Julien Beaulieu, Joël Bêty, Jean-Nicolas Jasmin, Kathy Jones, Heidi Kristensson, Jean-Bastien Lambert, Diane Leclerc, Stéphane Menu, Joanne Pelletier, Nathalie Piedboeuf, Monique Poulin, Catherine Poussart, Mathilde Renaud, Claudia St-Arnaud and Benoît Tremblay for their assistance on the field. A special thank to Mati Ilomets (Tallinn University, Estonia) for initiating us to the white mark technique. Finally, thanks to Martha Graf, Edgar Karofeld, Isabel Thibault and Peatland Ecology Research Group members for comments on earlier versions of the manuscript.

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ms. received May 7, 2009; accepted January 19, 2010.