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Autor(en): **Cailliau, Ariane / Price, Michelle J.**

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Hornworts in the agricultural fields of Geneva: new findings, the soil diaspore bank and ex situ soil cultures

Ariane Cailliau & Michelle J. Price

Abstract

CAILLIAU, A. & M. J. PRICE (2007). Hornworts in the agricultural fields of Geneva: new findings, the soil diaspore bank and ex situ soil cultures. *Candollea* 62: 165-172. In English, English and French abstracts.

The hornworts *Phaeoceros carolinianus* (Michx.) Prosk. and *Anthoceros agrestis* Paton are reported herein for the Canton of Geneva, Switzerland. The four new findings of *P. carolinianus* are the first records of this species from the canton since it was first collected by Jacques Rome from Meyrin in 1877. *Anthoceros agrestis*, not previously reported in the literature for Geneva, was recently discovered growing on cultured soil samples taken from three sites across the canton in the context of a bryophyte inventory of the canton. Its presence in the above-ground bryophyte flora was later confirmed by *in situ* populations found at seven localities. Culture studies of soil samples revealed that the diaspore bank plays a key role in the long-term maintenance of hornwort populations within Geneva. Hornworts are rare within the canton of Geneva.

Key-words

Anthoceros agrestis – *Phaeoceros carolinianus* – Geneva (Switzerland) – Ex situ cultivation – Diaspore bank – Bryophyte inventory

Résumé

CAILLIAU, A. & M. J. PRICE (2007). Anthocérotes dans les champs cultivés de Genève: nouvelles localités, banque de diaspores et mise en culture d'échantillons de sol. *Candollea* 62: 165-172. En anglais, résumés anglais et français.

Deux espèces d'anthocérotes sont répertoriées ici pour le canton de Genève, *Phaeoceros carolinianus* (Michx.) Prosk. et *Anthoceros agrestis* Paton. *Phaeoceros carolinianus* a été trouvé récemment dans quatre nouvelles localités, ce qui représente les premières données de cette espèce pour le canton depuis la récolte faite par Jacques Rome à Meyrin en 1877. *Anthoceros agrestis*, n'ayant jusqu'ici jamais été mentionnée dans la littérature pour Genève, a récemment germé dans des échantillons de sol récoltés sur trois sites du canton dans le cadre d'un inventaire des bryophytes. Des relevés de populations *in situ*, faits par la suite dans sept localités différentes du canton, ont confirmé la présence de cette espèce dans la bryoflore genevoise. Des études sur la mise en culture d'échantillons de sol ont révélé que la banque de diaspores joue un rôle clé dans le maintien à long terme des populations d'anthocérotes à Genève. Ces dernières sont rares dans le canton de Genève.

Address of the authors: Conservatoire et Jardin botaniques de la Ville de Genève, case postale 60, CH-1292 Chambésy.
Email: ariane.cailliau@ville-ge.ch

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Introduction

The project *Inventaire des hépatiques et des mousses de Genève* was started in 2004 with the aim of documenting the contemporary bryophyte flora of the canton of Geneva, Switzerland. Over the past century bryological collecting activity in Geneva markedly decreased, creating a gap in the understanding of the local bryophyte flora (BURGISSER & PRICE, 2005; CAILLIAU & PRICE, 2006). Recent collecting efforts resulted in a number of bryophyte species being newly recorded for the canton (PRICE, 2003; BURGISSER & al., 2004; BURGISSER, 2007). Newly reported herein is the hornwort *Anthoceros agrestis* Paton. Other species previously recorded from Geneva in the historical literature, such as the nationally rare hornwort *Phaeoceros carolinianus* (Michx.) Prosk., have recently been “re-found” in the canton.

Hornworts in Switzerland

Hornworts were first mentioned for Switzerland by HALLER (1768) under “*Anthoceros* [...] *levibus* Linn.” and “*Anthoceros* [...] *pennatifida*.” The former most likely refers to the taxon known in that epoch as *Anthoceros laevis* L. and the latter is of uncertain affinity. In his *Les hépatiques de la Suisse*, MEYLAN (1924) published five names for Switzerland corresponding to four currently recognised hornwort taxa, see Table 1. Today three species of hornworts are present in the country: *Anthoceros agrestis*, *A. punctatus* L. and *Phaeoceros carolinianus* (BISANG & URMI, 2006), two of which are now known to occur in the canton of Geneva (*A. agrestis* and *P. carolinianus*, see Fig. 1 and 2). These two species were previously known under *Anthoceros punctatus* var. *cavernosus* (Nees) Gottsche Lindenb. & Nees (= *A. agrestis*) (PATON, 1979) and *Phaeoceros laevis* subsp. *carolinianus* (Michx.) Prosk. (= *P. carolinianus*) (PATON, 1973). *Phaeoceros carolinianus* is considered of endangered status (EN) within Switzerland, *A. agrestis* falls into the category of least concern (LC) (SCHNYDER & al., 2004) while *A. punctatus* is in need of further evaluation to establish its status with respect to the Swiss bryophyte flora. In Switzerland hornworts are found on the plateau and to a lesser extent in the Jura mountains and in Tessin (BISANG, 1992). They grow most commonly in fields with oat, rye and wheat stubble from July through to November (BISANG, 1992, 1995a).

Soils of arable fields are subject to regular disturbance events and provide a period with bare soil, which is essential to the establishment of arable-field bryophytes, including hornworts (PORLEY, 2001). Extensive studies on hornwort biology and population dynamics in Switzerland, mostly in the Canton of Bern, have been carried out by BISANG (1992, 1995b, 1998, 2001, 2003). Her work on the hornwort diaspore bank has revealed its importance as a reservoir for the survival of populations (BISANG, 1995b, 1996). For example *A. agrestis*

produces relatively high quantities of spores, that is to say 14 mio spores/m² (BISANG, 2001). Studies indicated that this species could remain dormant in the form of diaspores as long as the conditions of germination were unsuitable (BISANG, 1995b, 1996). As the above ground flora does not necessarily reflect the soil diaspore bank composition, knowledge of the history of a site is important to predict the possible diaspore bank content (DURING & TER HORST, 1983; DURING & al., 1987), as was found in Swiss hornworts populations (BISANG, 1995b, 1996). Using herbarium records and field data, since the 1900’s, a decline in the occurrence of *A. agrestis* and *P. carolinianus* within Switzerland has been noted (BISANG, 1992, 1995b) and this change was attributed to changes in agricultural practice. The crops planted, associated treatments (the use of herbicides and fertilisers, soil compaction by heavy harvesting machinery), immediate ploughing after crop harvesting, and the use of catch-crops to prevent nitrogen leaching over the winter can all have deleterious affects on hornwort population presence (BISANG, 1992, 1995b, 1998, 1999).

Hornworts of Geneva: historical records

The first report of a hornwort for Geneva, was published as “*Anthoceros laevis*,” by the Geneva based bryologist Henri Bernet in his *Catalogue des hépatiques du sud-ouest de la Suisse* (BERNET, 1888). It was based on duplicate collections made by Jacques Rome “dans un champ près Meyrin, 1 juin 1877” (Fig. 1A). Examination of these specimens revealed that they were in fact *P. carolinianus* (monoicous), a species now recognised as separate entity from *P. laevis* (dioicous) based on sexuality and spore characteristics (HÄSSEL DE MENÉNDEZ, 1987; HASEGAWA, 1991; BLACKSTOCK & BOSANQUET, 2004).

The Rome duplicate collections, in the Rome and Bernet herbaria deposited in G, were later cited in MEYLAN (1924).

Table 1. – Hornworts mentioned in *Les hépatiques de la Suisse* by MEYLAN (1924) with their modern usage. The taxonomic names used follow GROLLE & LONG (2000).

Names from Meylan	Actual usage
<i>Anthoceros crispulus</i> (Mont.) Douin	= <i>A. agrestis</i> Paton
<i>A. punctatus</i> L.	= <i>A. punctatus</i> L.
<i>A. husnotii</i> Steph.	= <i>A. punctatus</i> L.
<i>A. laevis</i> L.	= <i>Phaeoceros laevis</i> (L.) Prosk.*
<i>A. dichotomus</i> Raddi	= <i>P. bulbiculosus</i> (Brot.) Prosk.**

* This taxon is currently excluded from the Swiss flora as all specimens previously named under this taxon are now recognised as *Phaeoceros carolinianus* (Michx.) Prosk.

** *Phaeoceros bulbiculosus* was thought likely from the Tessin (MEYLAN, 1924) but has not been recorded from Switzerland.

Discovery of hornworts in Geneva: soil sample cultures

In the context of the bryophyte inventory project historical localities and previously unexplored areas were surveyed to confirm the presence of bryophyte species that had been recorded from Geneva. Extensive fieldwork to locate suitable hornwort habitats and *in situ* hornwort populations was carried out between 2004 and 2007. Soil samples from sites such as arable fields and gravel extraction sites were collected. Gravel extraction sites in Geneva largely consist of old agricultural fields. In them the top-soil is removed and put aside before gravel is extracted from deeper below ground. The re-distributed top-soils from these sites create conditions suitable for bryophyte diaspore germination as they are similar to those seen in actively managed agricultural fields.

A total of 30 soil samples were gathered from 16 localities in 13 communes. In each sample area (chosen at random within the field) the surface of the soil was scraped off to remove any surface vegetation and debris. Soil was dug from a depth of between 0 and 10 cm for 29 of the samples, and from 20 cm down the profile of a vertical soil bank of a gravel extraction site for one sample. Each soil sample was placed into a clean and labelled plastic bag using a cleaned trowel. In an experimental greenhouse the soils were transferred into individual plastic trays, to a depth of 4-5 cm, and moistened with distilled water before being covered with unsealed plastic bags. This was done to reduce excessive evaporation from the culture trays between watering events. The soils were kept moist throughout the experiment using distilled water.

Eighteen of the 30 samples contained bryophytes (mostly *Bryum* Hedw. species) and three of these (samples S00, S03, S07), taken from the communes of Avusy and Collex-Bossy, gave rise to populations of *A. agrestis* (Table 2, Fig. 2). Plants of *A. agrestis* varied from 0.5-1.5 cm in diameter and all were fertile, bearing up to 15 sporophytes each that measured up to 2 cm long. Between 15 and 20 first generation individuals and between 24 and 60 second generation individuals (developed from spores from first generation plants) were found growing on the soil samples. The duration of one generation of plants varied from three months (soil sample S00 - second generation) to one year (soil sample S03 - first generation) (Table 2). Samples S00 and S03 came from different soil depths at the same locality. Diaspores were present up to 20 cm down the soil profile at this site.

Riccia sorocarpa Bisch. grew on all three soil samples containing *A. agrestis*. *Riccia glauca* L. and *R. sorocarpa* have been observed associated with hornworts in arable fields (*pers. observ.*) so when they were present at a site it was surveyed for hornwort population presence. The presence of

small hyaline worms in the three samples was noted, although their effects on the *ex situ* hornwort populations (soil perturbation or other) were not studied. *Anthoceros agrestis* is thought to be a poor competitor (BISANG, 2003). It was noted that the growth of mosses and the accumulation of a covering of algae and/or fungi on the soil surface reduced the longevity of the cultured hornwort populations.

New populations of *Anthoceros agrestis* and *Phaeoceros carolinianus* in Geneva

Despite regular visits to arable fields and all previous sample sites in 2004, 2005 and early 2006 it was not until late in 2006 that *in situ* populations of *A. agrestis* were recorded within the canton in a stubble field in Meyrin and in a stubble field, a meadow and a harvested sunflower field in Collex-Bossy (Fig. 2). These finds represent the first *in situ* populations of hornworts recorded from the canton for over 100 years. In 2007, *A. agrestis* was recorded in two new sites: in a fallow field in Laconnex and from a gravel extraction site in Sézegnin.

During 2006-2007 observation of an area of disturbed soil in Park Barton, one of the lake-side parks near the Botanical Garden of Geneva, revealed germinating populations of two hornwort species: *A. agrestis* and *P. carolinianus*, the first record of this latter species from the canton since collections of it were made in 1877. An area of this park was excavated to install a heating-cooling system for a nearby newly constructed office-block. Once the excavation work was finished excavated soil was replaced and soil from another construction site in Vernier (Geneva) was imported to infill any remaining gaps (Orlatti SA, Karl Steiner SA, *pers. comm.*). Locally abundant populations of these two species grew on the bare soil at this site. However, no hornwort gametophytes were found on the soil remaining at the construction site in Vernier itself although many bryophytes, including *Riccia sorocarpa*, were observed growing there. The exact provenance of the diaspores of these two hornwort species in this case is difficult to establish (disturbed soil from Park Barton versus displaced soil from Vernier) but whichever the origin of the soil, buried diaspores were uncovered through the soil manipulations enabling them to germinate on the newly exposed soil surface. The presence of *P. carolinianus* in the canton of Geneva was confirmed in late 2007 when it was recorded from three new localities within the canton. In two of the sites (Laconnex and Sézegnin) it was growing with *A. agrestis* while in the third site a tree nursery in Bernex, it was growing alone. A total of seven sites in five different communes (*A. agrestis*) and four sites in four different communes (*P. carolinianus*) are now known to contain hornwort populations.



Fig. 1. – **A.** *Phaeoceros carolinianus* (Michx.) Prosk. specimen collected by Rome in 1877 (G - G00043046); **B.** *Anthoceros agrestis* Paton specimen collected by Cailliau & Price in 2006 (G - G00124062); **C.** *Phaeoceros carolinianus* from Park Barton, photographed *ex situ*; **D.** *Anthoceros agrestis* *in situ* in Collex-Bossy.

Table 2. – The three out of 30 soil samples taken from across the canton of Geneva containing *A. agrestis* diaspores.

Sample code	S00 Avusy	S03 Avusy	S07 Collex-Bossy
Site	Gravel extraction site of Sézegnin	Gravel extraction site along "Creux de Boisset"	Stubble field between "le Seuchat" and "le Bracasset"
Vegetation	Fallow with pioneer ruderal species	Fallow with harvested sunflowers	<i>Phacelia tanacetifolia</i>
Sample depth	0.5 cm	20 cm	0-10 cm
Bryophytes present at the site	<i>Riccia sorocarpa</i> <i>Bryum argenteum</i>	None	<i>Riccia sorocarpa</i>
Start date	05.2004	11.2004	11.2004
1st generation	09.2004 - 03.2005	06.2005 - 06.2006	05.2005 - 01.2006
No. plants 1st generation	20 plants	15 plants	15 plants
2nd generation	05.2005 - 08.2005	09.2006 - still growing	dead
No. plants 2nd generation	60 plants	24 plants	0

Discussion

Over the past century the canton of Geneva has undergone extensive change as a result of increased urbanisation (DIA, 1981). Label information for the *Phaeoceros* specimens collected in Geneva in 1877 is vague. The ancient locality for *P. carolinianus*, given as "Meyrin," may have been built over as this site has developed from a small village into a large conurbation with a trading estate, retail estate and an industrial zone. It is also now home to the International airport of Geneva and to CERN.

In parallel with urban expansion and development, agricultural and land management practices have changed over time. Certain areas of the canton, such as around Collex-Bossy, were drained and the land reclaimed for cultivation (DIA, 1981). More generally cultivation practices have become more mechanised and intensified over time. Nowadays, farmers are encouraged to plant winter catch crops to protect the soil or for the prevention of nitrate leaching and thus fields are quickly ploughed after harvesting. The fields where *A. agrestis* was found growing in both Meyrin and Collex-Bossy in 2006 were ploughed four weeks after these populations were first seen and no populations were found in these sites the following year. Ploughing time seems to be the main factor influencing above-ground hornwort population presence (BISANG, 1992). If fields are ploughed soon after the harvest, then no time is available for hornworts to establish themselves and to complete their life-cycle (BISANG, 1992, 1998; PORLEY, 2001). In addition to mechanical damage, the use of herbicides and fertilisers may be responsible for hornwort gametophyte absence (BISANG, 1995b), although the effect of such chemical treatments on the diaspore bank is poorly known (BROWN, 1992; PORLEY,

2001). Additional factors that can interfere with hornwort presence and/or population development are drought and local soil disturbance caused by heavy rain or by animals (BISANG, 2003).

Hornworts were found in Geneva during the autumn of 2006 and 2007. Both these years had a wet and/or mild summer, conditions that may have encouraged the germination of diaspores. In the fields of the canton of Bern, central Switzerland, populations of both *A. agrestis* and *P. carolinianus* were also more numerous than in previous years (*pers. observ.*; BISANG, *pers. comm.*).

Since bryophyte collecting and recording waned between the early-1900's and the beginning of the current inventory project, the lack of reports and herbarium collections of hornworts from Geneva directly reflects collecting activity rather than the "absence" of them from the flora. Based on our current field observations, *A. agrestis* and *P. carolinianus* are both rare within the canton of Geneva. The emergence of *A. agrestis*, first from soil cultures and later in *in situ* populations, shows how a diaspore bank can contain elements of the flora that are absent from the soil surface. Populations of *A. agrestis* were found growing in the two localities where soil samples that contained diaspores of *A. agrestis* had been taken: namely in fields of Collex-Bossy and in the gravel extraction site of Avusy. The absence of populations of *A. agrestis* in the above-ground flora and its presence as diaspores in the soil has previously been noted in Switzerland (BISANG, 1995b, 1996, 1999) and in Great Britain (PORLEY, 2001).

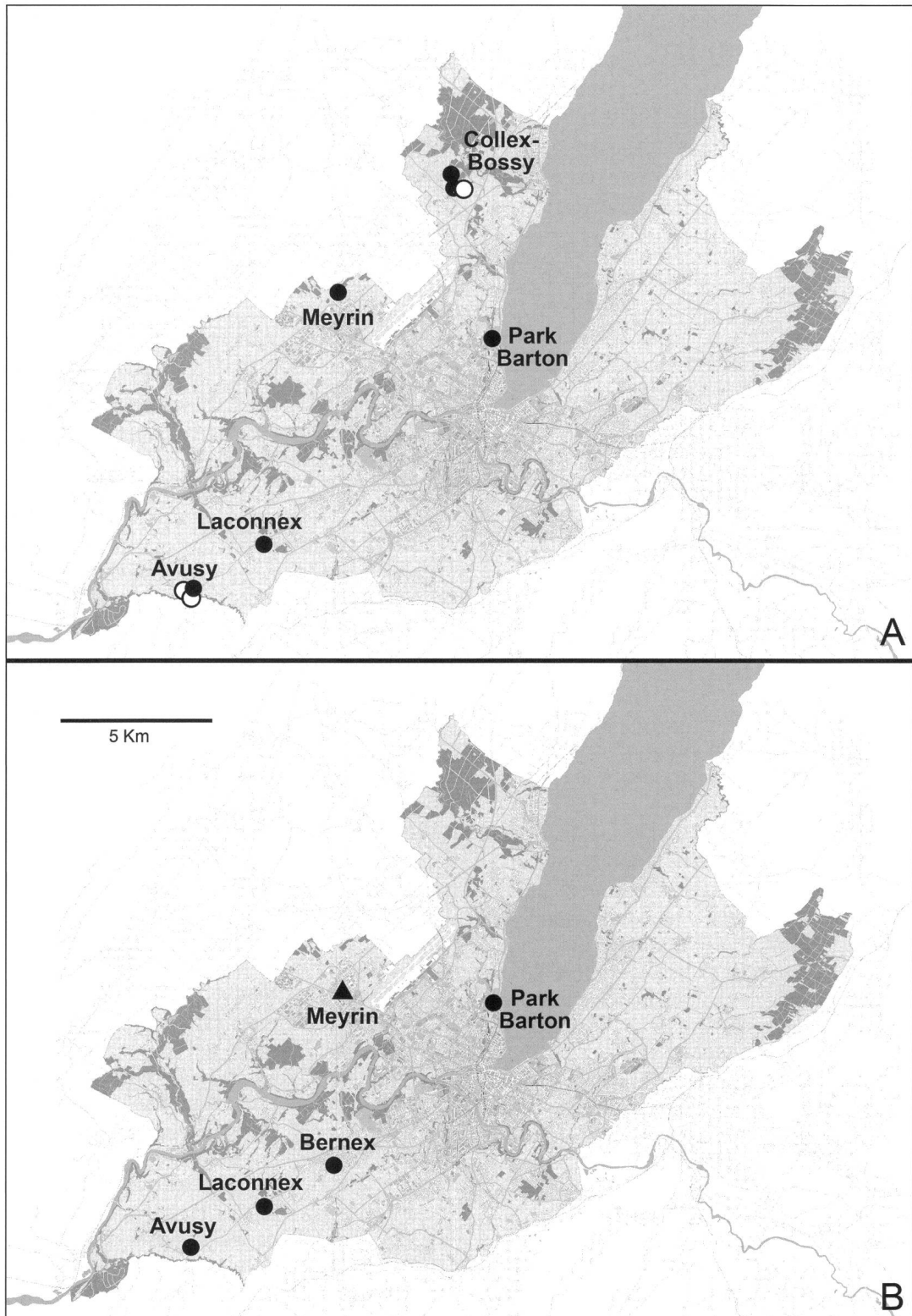


Fig. 2. – Localities of the two hornwort species in Geneva. **A.** *Anthoceros agrestis* Paton: sites where viable diaspores have been found O; sites of *in situ* populations ●; **B.** *Phaeoceros carolinianus* (Michx.) Prosk.: sites of *in situ* populations ●; location of the Rome collections of 1877 ▲.

Conclusion

The two species *A. agrestis* and *P. carolinianus*, found growing *in situ* in Geneva in 2006, represent the first published report of hornworts from the canton for 120 years. The continuous presence of hornworts in the canton during this period cannot be confirmed using herbarium records although it is likely that they have been present in low densities during this period as diaspores are present in the soil bank across the canton. Our soil cultures and the appearance of hornworts on recently disturbed soils within the city limits shows that the diaspore bank plays an important role in the maintenance of hornwort populations. Both the hornwort species now known for Geneva are rare within the canton. As agricultural land accounts for nearly 50% of the canton's surface, potential habitats for hornworts are well represented. Further long-term investigations are required to follow the dynamics of hornwort populations in the canton, to assess the abundance / frequency of the apparition of populations, and to assess exactly which factors influence hornwort presence and population densities on a site by site basis. Farming regime, crop type, soil structure, surface treatments, and soil or soil surface humidity as well as weather conditions should all be taken into account in any future assessments.

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