

## *Tuber aestivum* (syn. *T. uncinatum*) biotopes and their history on Gotland, Sweden

Christina WEDÉN<sup>1\*</sup>, Gérard CHEVALIER<sup>2</sup> and Eric DANELL<sup>3</sup>

<sup>1</sup>Department of Systematic Botany, Uppsala University, Norbyvägen 18D, SE-752 36 Uppsala, Sweden and Gotland University College, Visby, Sweden.

<sup>2</sup>Institut National de la Recherche Agronomique, 234 avenue du Brézat, 630 39 Clermont-Ferrand Cedex 2, France.

<sup>3</sup>Museum of Evolution, Botany Section, Uppsala University, Norbyvägen 16, SE-752 36 Uppsala, Sweden.

E-mail: Christina.Weden@ebc.uu.se

Received 21 April 2003; accepted 20 November 2003.

This study aimed at testing the hypothesis that the genetically distinct *Tuber aestivum* population on the island of Gotland, Sweden, is adapted to habitats different from French *T. aestivum* populations. The soil structure, soil chemistry, bedrock, climate, vegetation and host tree continuity of 18 *T. aestivum* sites on Gotland were analysed and compared with data from France. We conclude that *T. aestivum* can grow in soils with a broad soil structure range and that no striking differences in soil chemistry were found. No *T. aestivum* indicator plants other than the host trees were found, but the host tree continuity on the *T. aestivum* sites on Gotland was more than 300 yr. If the *T. aestivum* population on Gotland constitutes an ecotype it is rather an adaptation to the colder and drier climate on Gotland. Selecting local *T. aestivum* inoculum for truffle orchards in northern Europe could be important for successful truffle production.

### INTRODUCTION

*Tuber aestivum* (syn. *T. uncinatum*<sup>†</sup>), the Burgundy truffle, is an ectomycorrhizal hypogeous ascomycete found throughout Europe associated with, for example, *Quercus robur* and *Corylus avellana* (Chevalier & Frochot 1997). *T. aestivum* and *T. mesentericum* (Wedén, Ericsson & Danell 2001) are the only black truffles reported from Sweden, both being found only on Gotland, an island off the east coast of Sweden in the Baltic Sea isolated from the mainland since the last ice age 11 600 yr ago (Fredén 1998). Our preliminary studies on Gotland during the autumns 1998–2000 showed that *T. aestivum*, which until then only had been recorded three times from Gotland (Sunhede 1978, Bohus Jensen 1988, Kers 1992), was well distributed over the island and adapted to local climate and soil conditions (Wedén & Danell 1998, Wedén *et al.* 2001). Randomly amplified polymorphic DNA

(RAPD) data indicated that the Gotland *T. aestivum* was genetically distinct and separate from populations in central Europe and probably derived from one or two introductions (Wedén *et al.* 2004). It was also found that the soil preferences of the Gotland *T. aestivum* population seemed fundamentally different from what was previously known from French populations (Chevalier & Frochot 1997).

Understanding the climatic and edaphic preferences of the *T. aestivum* Gotland population could be of crucial importance when selecting inocula for planned truffle orchards on Gotland. The research presented here further investigated if the *T. aestivum* Gotland population occurs in areas with soil and climatic characteristics markedly different from French populations. The lack of soil profiles from forests or meadows on Gotland further motivated this study (Torbjörn Nilsson, pers. comm.).

### MATERIALS AND METHODS

*Tuber aestivum* sites on Gotland were found using trained truffle dogs with a site defined as a *Quercus robur* and/or *Corylus avellana* stand disconnected from other sites by, for example, roads or arable land.

\* Corresponding author.

† According to tradition, French truffle specialists name the Gotland black truffle *Tuber uncinatum* (M. Jalade, pers. comm.; D. Delage, pers. comm., *L'écomusée de la truffe*, Sorges). As was stated in Wedén *et al.* (2004), it is likely that *T. aestivum* and *T. uncinatum* are the same species. Since the oldest name has priority, we only use the name *T. aestivum* here.

The soil was sampled by first removing the litter and vegetation and then collecting 1 kg of soil from the soil layer where the fruit body was found, i.e. down to 10–20 cm depth, depending on how rocky the soil was. Six sites were sampled in Sept. 1999 (samples A-T2, B-T5, C-T10, D-T9, E-T8 and P-T15) and the remaining 12 in Sept. 2001. All the sites, where soil was sampled and analysed, were also represented in our previous RAPD study (Wedén *et al.* 2004). The vegetation was also recorded to determine if there was any *T. aestivum* indicator species. We defined an indicator species as a vascular plant which biotope preferences coincide with those of *T. aestivum*. At one *T. aestivum* site, the occurrence of mature *T. aestivum* fruit bodies was recorded throughout 1998–2002 to estimate the duration of the *T. aestivum* fruit body season on Gotland. Representative specimens are preserved in UPS.

The soil analyses were carried out by the Laboratoire d'Analyses des Sols, Institut National de la Recherche Agronomique (INRA; Arras, France). This laboratory had previously carried out the analyses of the only published study of soils from natural *T. aestivum*\* sites from Burgundy, Lorraine, Franche-Comté and Auvergne regions of eastern France where the Burgundy truffle is commonly found (Chevalier & Frochot 1997). Some soil data from other European *T. aestivum* populations also exist (Bratek *et al.* 1991, 2001, Belloli *et al.* 2001). These data are congruent with the French soil data, but mainly consist of pH data, data from a limited number of sites or summarised soil data where the range is unknown, which obstruct more thorough comparisons. The parameters measured in our study were: particle size in five fractions: <2 µm (clay), 2–20 µm (fine silt), 20–50 µm (coarse silt), 50–200 µm (fine sand), and 200–2000 µm (coarse sand) (Norme Internationale ISO 11277); pH in water (NF ISO 13878); total CaCO<sub>3</sub> content (NF ISO 10693); assimilable P<sub>2</sub>O<sub>5</sub> (Joret & Hebert 1955, Duval 1963); ammonium acetate exchangeable Ca, Mg and K (Thomas 1982, Ciesielski & Sterckeman 1997); organic carbon and organic matter (NF ISO 10694); organic nitrogen (NF ISO 13878) and C/N ratio.

Detailed 1:50 000 soil type maps (Munthe *et al.* 1927–40) were also studied in order to define the general soil types and bedrock at the *T. aestivum* sites on Gotland. Weather data (temperature and precipitation) was derived from the Swedish Meteorological and Hydrological Institute (SMHI) and Meteo France.

A cadastral, digitised 1:10 000 map of Gotland from 1700, showing land use such as arable land, meadowland and forest (MAPINFO 6.5), was superimposed on modern 1:50 000 vegetation maps (MAPINFO 6.5),

to determine the management history of the Gotland *T. aestivum* sites. We defined meadow as anything from thickly wooded meadows with a more or less closed canopy cover to more open, sparsely wooded grasslands (Croneborg 2001). To estimate the continuity of the sites after year 1700, we also studied a 1:10 000 map of Gotland based on aerial photographs made in 1930 (Gotland County Administration, Visby).

## RESULTS

Soil characteristics from Gotland are presented in Table 1. In Fig. 1 and Table 2, soil analysis data from Swedish and French *Tuber aestivum* sites are summarised.

According to the detailed soil type maps (Munthe *et al.* 1927–40), the soil at the *T. aestivum* sites is characterised by either moraine marlstone or different kinds of gravel and sand mixtures, or a mix of the two. The bedrock belongs to five different groups which from youngest to oldest are: bedrock belonging to the Hemse group – stratified, predominantly crystalline, partly fine oolitic limestone, reef shaped limestone, reef limestone, marly limestone and marlstone (nine sites); Mulde marlstone – marlstone and marly limestone (one site); Halla limestone – stratified, more or less marly limestone and reef limestone (one site); Slite group – stratified, crystalline limestone and reef limestone, marlstone and marly limestone, sand limestone and lime sandstone (six sites) and on Högkling limestone – stratified, more or less marly limestone and marlstone, and reef limestone (one site) (Munthe *et al.* 1927–40).

Despite its relative northern location (latitudes 57–58° N, longitudes 18–19° E), Gotland belongs to the warmest Swedish climate zone, together with the very south of Sweden (latitudes 55.5° N, longitudes 13° E). This is due to the warming effect of the large body of water surrounding the island of Gotland, making the climate milder than that of the mainland at the same latitude. Gotland's location east of the mainland also results in lower precipitation and a greater number of hours of sunshine than on the mainland. The annual mean precipitation in Cruzy, Burgundy, France (1971–2000) was 884.6 mm, while on Gotland (Sanda weather station, 1971–2000) it was 527.5 mm. The annual mean temperature (1971–2000) in Cruzy was 10.4 °C and on Gotland (Visby Airport, 1961–90) it was 6.8 °. The duration of the *T. aestivum* fruit body season is presented in Table 3.

The ectomycorrhizal host species *Quercus robur* and *Corylus avellana* grew at all the 18 *T. aestivum* sites on Gotland. The relative occurrence of the two species differed between the sites, some being dominated by *Q. robur* and others by *C. avellana*. In the vicinity of where the *T. aestivum* fruit bodies were found, *Fraxinus excelsior* occurred on at least half of the sites, while

\* When French *Tuber aestivum* data is referred to, the data regards truffles that in France are called *T. uncinatum*. There is yet no convincing evidence that *T. aestivum* and *T. uncinatum* are different species, why we only use the name *T. aestivum* in this article (see p. 304 footnote).

Table 1. Overview of the soil composition at the 18 *Tuber aestivum* sites on Gotland, Sweden.

Site identification	A-T2	B-T5	C-T10	D-T9	E-T8	F-T11	G-T1	H-T6	I-T7	J-T20	K-T19	L-T16	M-T17	N-T18	O-T13	P-T15	Q-T21	R-T22
Soil particle size fractions (%)																		
<2 µm (clay)	21.4	15.0	22.4	No data	10.4	32.6	23.1	14.2	16.4	18.3	15.8	22.0	22.9	18.1	18.1	21.4	16.8	18.5
2–20 µm (fine silt)	14.7	16.2	49.0	No data	6.9	23.1	18.4	7.1	12.5	13.1	16.6	20.4	18.5	19.5	16.9	16.6	15.3	13.2
20–50 µm (coarse silt)	5.4	10.8	15.7	No data	2.9	9.5	8.8	4.6	6.4	7.2	8.2	10.4	7.2	5.4	11.4	4.3	6.8	4.2
50–200 µm (fine sand)	11.6	29.6	6.2	No data	9.3	16.9	22.8	20.6	43.1	17.7	28.9	26.7	19.4	13.3	25.0	10.9	20.4	8.1
200–2000 µm (coarse sand)	46.9	28.4	6.7	No data	70.5	17.9	26.9	53.5	21.6	43.7	30.5	20.5	32.0	43.7	28.6	46.8	40.7	56.0
Water pH	7.0	6.8	7.1	7.4	7.6	7.1	7.2	7.6	7.9	7.8	7.6	7.9	7.7	7.6	7.4	7.6	7.5	7.7
Total lime stone (CaCO <sub>3</sub> ) (%)	0.2	0.1	0.2	0.2	1.7	0.6	0.1	0.9	8.2	8.0	1.7	2.0	10.5	6.3	0.3	4.1	0.6	7.5
Assimilable phosphorus (P <sub>2</sub> O <sub>5</sub> ) (%)	0.0161	0.0228	0.0100	0.0044	0.0064	0.0028	0.0039	0.0053	0.0068	0.0062	0.0048	0.0324	0.1174	0.0142	0.0038	0.0844	0.0016	0.0054
Exchangeable calcium (Ca) (%)	0.356	0.382	0.443	0.422	0.563	0.604	0.588	0.641	0.991	0.848	0.832	0.612	0.818	0.927	0.519	0.829	0.664	1.071
Exchangeable magnesium (Mg) (%)	0.0286	0.0233	0.0166	0.0091	0.0142	0.0186	0.0330	0.0130	0.0233	0.0206	0.0140	0.0129	0.0142	0.0137	0.0180	0.0451	0.0100	0.0178
Exchangeable potassium (K) (%)	0.0627	0.0276	0.0127	0.0112	0.0127	0.0331	0.0109	0.0102	0.0348	0.0086	0.0076	0.0309	0.0565	0.0145	0.0077	0.0187	0.0081	0.0160
Ca/Mg	12.4	16.4	26.7	46.4	39.6	32.5	17.8	49.3	42.5	41.2	59.4	47.4	57.6	67.7	28.8	18.4	66.4	60.2
K/Mg	2.2	1.2	0.8	1.2	0.9	1.8	0.3	0.8	1.5	0.4	0.5	2.4	4.0	1.1	0.4	0.4	0.8	0.9
Carbon and nitrogen																		
Organic carbon (%)	5.520	5.460	4.690	3.470	8.690	5.797	6.303	7.358	8.956	6.217	9.215	3.465	5.256	12.338	4.165	9.110	7.053	11.674
Organic matter (%)	9.49	9.39	8.07	5.97	14.95	9.97	10.84	12.66	15.40	10.69	15.85	5.96	9.04	21.22	7.16	15.67	12.13	20.08
Organic nitrogen (%)	0.483	0.422	0.371	0.264	0.551	0.488	0.549	0.756	0.789	0.497	0.766	0.270	0.434	1.060	0.357	0.500	0.405	0.800
C/N	11.43	12.94	12.64	13.14	15.77	11.88	11.48	9.73	11.35	12.51	12.03	12.83	12.11	11.61	11.67	18.22	17.41	14.59

*Crataegus* spp. grew at 13 of the 18 sites. Ground vegetation differed with the habitat type and no common ground vegetation could be distinguished to indicate the presence of *T. aestivum*, other than its host trees. In more open habitats, the ground was sparsely vegetated with different grasses and the earth could be more or less covered by mosses such as *Plagiomnium affine* and *Brachythecium rutabulum*. In the *T. aestivum* forest or meadow habitats, the ground was more or less bare and the very sparse ground vegetation was composed of, for example, *Hepatica nobilis* and *Fraxinus excelsior* shoots. Seven sites were grazed by cattle or sheep, on three sites the grass was cut and four sites were unmanaged but shaded, thus restraining ground vegetation. On the remaining four sites, *T. aestivum* fruit bodies were found both in a mown lawn and in two of these sites also in an adjacent vegetable bed. Some of the *T. aestivum* producing trees showed moderate truffle burns (brûlés), which appeared as sparse vegetation around the trees (Chevalier & Frochot 1997).

Of the 18 sites studied, 15 were found on land which, according to the 1700 cadastral map, was defined as meadow. The remaining three sites were found on land within a 100 m. of what had been meadow in 1700. In the first of these three sites, *T. aestivum* was located on what was built-up area in 1700 adjacent to a meadow. The second site was a field adjacent to a built up area and meadow and the third site was located just outside the border of a meadow. While great care was exercised, it was possible that the different scales of the 1700 cadastral map (1:10 000) and the modern map (1:50 000) could have led to slight translation errors.

## DISCUSSION

### Soil

The French and Swedish truffle populations both occupy land with high pH (7–8) soils and low phosphorus concentrations (0.002–0.080%). The only exception was the Swedish site B-T5 where the pH was 6.8, and M-T17 where there was a high phosphorus concentration of 0.12% (Table 1). The texture of the soil samples from the Swedish *Tuber aestivum* sites was silty to sandy, while French soils were more clayey (Fig. 1). Due to the broad range in soil particle size in both Swedish and French sites, we conclude that *T. aestivum* has a wide soil texture tolerance and the differences in the sites simply reflected regional geology.

Unlike French *T. aestivum* soils, which always respond to the hydrochloric acid (HCl) test indicating free CaCO<sub>3</sub> (Chevalier & Frochot 1997), some *T. aestivum* soils from Gotland failed to react indicating a lack of free CaCO<sub>3</sub>, a finding supported by our analyses (Table 2). However, the fraction of assimilable calcium was often much higher in the Swedish than in the French soils (Table 2). The Ca/Mg ratios were also of interest, since calcium and magnesium compete for the same positions on soil colloids. This means that sites



**Table 2.** A soil composition comparison between Swedish (Table 1) and French *T. aestivum* sites, presenting the mean, SD and range. French data originate from Chevalier & Frochot (1997).

Measured parameter	Mean ± SD		Range	
	Sweden	France	Sweden	France
Clay <2 µm %	19.3 ± 4.9	34.1 ± 11.1	10.4–32.6	13.6–52.8
Silt 2–50 µm %	25.1 ± 11.9	48.3 ± 11.6	9.8–64.7	17.3–67.4
Sand 50–2000 µm %	55.6 ± 14.9	17.5 ± 16.1	12.9–79.8	2.8–69.1
Water pH	7.5 ± 0.3	7.6 ± 0.2	6.8–7.9	7.1–8.0
CaCO <sub>3</sub> (total) %	3.0 ± 3.5	16.7 ± 17.2	0.1–10.5	0.4–52.0
Exchangeable calcium <sup>a</sup> (Ca) %	0.67 ± 0.21	0.51 ± 0.14	0.36–1.07	0.28–0.79
Assimilable phosphorus (P <sub>2</sub> O <sub>5</sub> ) %	0.020 ± 0.032	0.009 ± 0.017	0.002–0.120	0.002–0.082
Exchangeable magnesium <sup>a</sup> (Mg) %	0.019 ± 0.009	0.017 ± 0.011	0.009–0.045	0.005–0.041
Exchangeable potassium <sup>a</sup> (K) %	0.023 ± 0.017	0.059 ± 0.022	0.008–0.063	0.025–0.104
Ca/Mg	40.6 ± 17.3	58.5 ± 31.6	12.4–67.7	19.5–116.5
K/Mg	1.2 ± 0.9	4.5 ± 2.4	0.3–4.0	1.3–8.1
Organic matter %	11.9 ± 4.5	9.7 ± 4.2	6.0–21.2	4.4–21.1
Organic carbon %	6.9 ± 2.6	5.6 ± 2.4	3.5–12.3	2.6–12.3
Organic nitrogen %	0.54 ± 0.21	0.46 ± 0.14	0.3–1.1	0.3–0.8
C/N ratio	13.0 ± 2.2	11.9 ± 2.6	9.7–18.2	8.9–20.4

<sup>a</sup> French values are recalculated from m.e./100 g to %.

**Table 3.** Monthly mean precipitation and temperature, and the duration of the *Tuber aestivum* fruiting body season, on Gotland, Sweden and in Burgundy, France. The Swedish precipitation data is derived from the weather station Sanda at the East Coast of Gotland (years 1971–2000) and the temperature data is derived from the weather station at Visby Airport, Gotland (years 1961–90). The reason for using separate weather stations for the precipitation and temperature data was due to data availability. The French precipitation and temperature data is derived from the weather station in Cruzy, Burgundy, France (years 1971–2000). The duration of the season for finding mature fruiting bodies of *T. aestivum* in Burgundy refer to Chevalier & Frochot (1997).

Month	Monthly mean precipitation (mm)		Monthly mean temperature (°C)		Main season for finding mature <i>Tuber aestivum</i> <sup>a</sup> fruiting bodies (exceptions exist)	
	Gotland	Burgundy	Gotland	Burgundy	Gotland	Burgundy
January	43.3	73.6	−1.1	2.7		//////////
February	32.6	69.3	−1.8	3.6		
March	34.6	69.3	0.1	6.5		
April	29.3	60.4	4.0	9.0		
May	28.0	81.9	9.6	13.2		
June	39.5	75.9	14.3	16.0		
July	51.6	65.4	16.3	18.7		
August	48.5	66.4	15.9	18.6	////////	
September	56.8	72.0	12.1	15.1	//////////	////////
October	50.5	80.0	8.1	10.9	//////////	//////////
November	57.5	83.2	3.9	5.9	//////////	//////////
December	55.5	87.1	0.7	3.8		//////////

<sup>a</sup> Data from Burgundy refer to the data of *T. uncinatum* from Chevalier & Frochot (1997).

CaCO<sub>3</sub> give the soils a good structure, which compensates for the otherwise heavy structured soils and give them good drainage characteristics (Chevalier & Frochot 1997, Chevalier *et al.* 2002). On Gotland, the *T. aestivum* soils are lighter in structure (Fig. 1), but the organic matter may still play an important role in aerating the soil, which would benefit *T. aestivum*. Although *Pinus sylvestris* constitutes up to 90% of the forest on Gotland (Kloth & Lovén 1987) it does not seem to form mycorrhizae with *T. aestivum*. However, *P. sylvestris* is generally planted on poor land where the soil layer can be very thin and the calcareous bedrock exposed. This is quite unlike the organic rich soils at *T. aestivum* sites (Table 1) (Chevalier & Frochot

1997) which may be the reason why *T. aestivum* is not associated with *P. sylvestris*.

The Swedish and French C/N ratios are also similar. For agricultural land, a C/N ratio below 10 is known to be an indication that the soil has been N-fertilised (Eriksson *et al.* 1997). Only one Swedish soil sample (Table 1, site H-T6, C/N=9.73) and two out of the 25 French soil samples (Chevalier & Frochot 1997) had a C/N ratio below ten, indicating that *T. aestivum* prefers soils which are poor in readily degradable nitrogen. This is well in accordance with the findings of Wallander (1992), that a high nitrogen content in the soil can act harmfully on mycorrhizas. *T. aestivum* does not seem restricted to any particular kind of calcareous

bedrock, but was found on five different kinds of limestone.

### Climate

Our comparison of the climatic data shows Gotland to be drier and cooler than Burgundy, which is probably why the season for mature *Tuber aestivum* fruit bodies starts and ends earlier on Gotland than in France (Table 3). It is also possible that the cold Swedish winters are a strong selective force for suitable *T. aestivum* strains. From the results of their study of *T. melanosporum* orchards in Italy, Bencivenga & Granetti (1988) suggested the importance of using mycorrhized seedlings produced from seeds and spores of local ecotypes. A test of the hypothesis that the *T. aestivum* population on Gotland is an ecotype resulting from a climatic adaptation, would be to study its survival in other areas such as Burgundy in France, and *vice versa*.

### Vegetation

*Quercus robur* and *Corylus avellana* are by far the most common *T. aestivum* host species growing on Gotland. Other known *T. aestivum* host species, such as *Fagus sylvatica*, *Carpinus betulus* and *Tilia cordata*, although common on the mainland of southern Sweden, only occur very sparsely on Gotland. That both *Q. robur* and *C. avellana* grow on all the investigated *T. aestivum* sites on Gotland, is probably only due to the two species sharing the same biotope preferences. It is not uncommon to find *T. aestivum* on Gotland under *F. excelsior* surrounded by *C. avellana*. Although *F. excelsior* is not an ectomycorrhizal species, the canopy cover it provides and its influence on soil temperature and soil conditions may have a positive effect on *T. aestivum* fruiting. Similarly, the roots of *Crateagus* spp. may have an aerating effect on the soil, which could also aid *T. aestivum* development. Both of these possibilities need further investigation. No common ground vegetation could be distinguished to indicate the presence of *T. aestivum*, other than its host trees.

A comparison of the 1700 and 1930s cadastral maps shows that it is very likely that the *T. aestivum* sites have existed for at least 300 yr. The old tradition of meadowland management allowed deciduous trees to grow in groups surrounded by grassland and has been in practise on Gotland since the iron age, 1500 yr ago (Kloth & Lovén 1987). The old management practise involved cutting, removing and burning the grass in mid-summer, and cutting the young branches of deciduous trees for fodder for cattle and sheep. Of the estimated 82 000 ha meadows in 1700, only 2300 ha remain (Länsstyrelsen i Gotlands län 2002) of which only a small fraction are still traditionally managed. The decrease in meadows has also decreased the total area of potential *T. aestivum* habitats. Since *T. aestivum* was never found on sites with tall grass, it seems to have only survived in either managed habitats where grazing

by domestic animals or cutting the grass have contributed to keeping down the ground vegetation, or where the ground vegetation has been sparse due to other factors. For example, in some of the former meadowlands, the canopy cover has closed leaving the forest floor very sparsely vegetated. Despite the conspicuous decline of meadowlands on Gotland, there are still enough suitable *T. aestivum* habitats left to find inoculum to produce seedlings aimed for the establishment of new truffle orchards on Gotland. This could be of great importance, since a climatically adapted *T. aestivum* ecotype may be required on this northern latitude.

### ACKNOWLEDGEMENTS

We are grateful to Olle Persson, Anders Wedén, Michel Jalade (France), Pierre Poinot and Marcelle Poinot (France), Hjalmar Croneborg, Torbjörn Nilsson, Karin Wågström, Stellan Hedgren, Leif Karleby, and landowners on Gotland for their valuable contributions to this study. This study was financed by the Municipality of Gotland, the European Agriculture Guidance and Guarantee Fund under the Objective 5b Gotland Programme, the European Regional Development Fund under the Objective 2 Islands Programme, the Gotland County Administration, and further supported by the KK Foundation (Kunskaps- och kompetensutvecklingsstiftelsen), Carl the XVI Gustafs 50th Anniversary Fund and Carl Tryggers Foundation.

### REFERENCES

- Belloli, S., Bologna, F., Gregori, G. & Zambonelli, A. (2001) Il tartufo nero di Fragno (*Tuber uncinatum* Chatin): ecologia e coltivazione. In *Actes du V<sup>ème</sup> Congrès International Science et Culture de la Truffe*: 367–371. Federation Française des Trufficulteurs, Paris.
- Bencivenga, M. & Granetti, B. (1988) Risultati produttivi di tartufoaie coltivate di *Tuber melanosporum* Vitt. in Umbria. In *Atti del Secondo Congresso Internazionale sul Tartufo* (M. Bencivenga & B. Granetti, eds): 313–321. Comunità montana dei monti martani e del serano, Spoleto.
- Bohus Jensen, E. (1988) Om tryfflar på Gotland. *Rindi* 8: 15–18.
- Bratek, Z., Bagi, I., Parádi, I. & Vikor, J. (2001) Differentiation among truffle species based on habitat preferences. In *Actes du V<sup>ème</sup> Congrès International Science et Culture de la Truffe*: 193–195. Federation Française des Trufficulteurs, Paris.
- Bratek, Z., Király, I. & Láng, F. (1991) R-spectra of some hypogeous mushrooms. *Micologia e vegetazione mediterranea* 7: 95–102.
- Chevalier, G. & Frochot, H. (1997) *La Truffe de Bourgogne*. Pétrarque, Levallois-Perret.
- Chevalier, G., Gregoire, G., Frochot, H. & Zambonelli, A. (2002) The cultivation of the Burgundy truffle. In *Edible Mycorrhizal Mushrooms and their Cultivation, Proceedings of the Second International Conference on Edible Mycorrhizal Mushrooms* (I. R. Hall, Y. Wang, A. Zambonelli & E. Danell, eds): CD-ROM. New Zealand Institute for Crop and Food Research Limited, Christchurch.
- Ciesielski, H. & Sterckeman, T. (1997) A comparison between three methods for the determination of cation exchange capacity and exchangeable cations in soils. *Agronomie* 17: 9–15.
- Croneborg, H. (2001) *Gotländska Ängar. En katalog över slåttermarker i hävd på Gotland år 2001*. [Livsmiljöenheten Rapport No. 4.] Gotland County Administration, Visby.
- Duval, L. (1963) Etude des conditions de validité du dosage céruléomolybdique de l'acide phosphorique. Conséquences pratiques. *Chimie Analytique* 45: 237–250.

- Eriksson, J., Andersson, A. & Andersson, R. (1997) *Tillståndet i Svensk Åkermark*. [Naturvårdsverket Rapport No. 4778.] Naturvårdsverket Förlag, Stockholm.
- Fredén, C. (1998) *Berg och Jord – Sveriges Nationalatlas*. Sveriges geologiska undersökning, Uppsala.
- Joret, G. & Hebert, J. (1955) Contribution à la détermination du besoin des sols en acide phosphorique. *Annales Agronomiques* **2**: 233–299.
- Kers, L. E. (1992) The truffle *Fischerula macrospora* Matt. found in Sweden. *Svensk Botanisk Tidskrift* **86**: 295–298.
- Kloth, J.-H. & Lovén, U. (1987) *Gotlands Natur*. Bonnier Fakta Bokförlag, Stockholm and Gotland County Administration, Visby.
- Länsstyrelsen i Gotlands län (2002) *Nyckelbiotopsbasen*. Gotland County Administration, Visby.
- Munthe, H., Hede, J. E. & Lundqvist, G. (1927–1940) *Beskrivning till Kartbladet Ronehamn (1925); Beskrivning till Kartbladet Klintehamn (1927); Beskrivning till Kartbladet Hemse (1927); Beskrivning till Kartbladet Slite (1928); Beskrivning till Kartbladet Katthammarsvik (1929); Beskrivning till Kartbladet Kappelshamn (1933)*. Sveriges Geologiska Undersökning, Stockholm.
- Sunhede, S. (1978) *Geastrum pseudolimbatum*, *Perenniporia fraxinea* and *Tuber aestivum* found in Sweden. *Svensk Botanisk Tidskrift* **72**: 263–269.
- Thomas, G. W. (1982) Exchangeable cations. In *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties* (A. L. Page *et al.*, eds): 154–157. [Agronomy Monograph No. 9 (2nd edn).] American Society of Agronomy & Soil Science Society of America, Madison, WI.
- Wallander, H. (1992) *Regulation of ectomycorrhizal symbiosis in Pinus sylvestris L. seedlings – influence of mineral nutrition*. PhD thesis, Swedish University of Agricultural Sciences, Uppsala.
- Wedén, C. & Danell, E. (1998) *Tuber aestivum* and other truffles in Sweden. *Svensk Botanisk Tidskrift* **92**: 65–80.
- Wedén, C., Danell, E., Camacho, F. & Backlund, A. (2004) The population of the hypogeous fungus *Tuber aestivum* syn. *T. uncinatum* on the island of Gotland. *Mycorrhiza*: in press.
- Wedén, C., Ericsson, L. & Danell, E. (2001) Research on *Tuber aestivum* syn. *T. uncinatum*, and *T. mesentericum* reported from Sweden for the first time. *Svensk Botanisk Tidskrift* **95**: 205–211.

Corresponding Editor: J. I. Lelley