Vol. XXXIII, 2019 Vol. XXXIII, 2019

RESEARCH ON FOREST SPECIES FOR BLACK TRUFFLE (*TUBER AESTIVUM*) USING G.I.S. TECHNIQUES. CASE STUDY

Păcurar Ioan*, Roșca Sanda**, Păcurar Horea Mihai*, Bilașco Ștefan***, Negrușier Cornel*, Dirja Marcel*

*University of Agricultural Sciences and Veterinary Medicine, 3-5 Manastur St., 400372 Cluj-Napoca, Romania; ioanpacurarcj@yahoo.com, pacurar_horea@yahoo.com
**Babeş-Bolyai University, Faculty of Geography, Department of Physical and Technical Geography, Clinicilor Street, 400006 Cluj-Napoca, Romania; sanda.rosca@ubbcluj.ro
*** Romanian Academy, Cluj-Napoca Subsidiary Geography Section, 9 Republicii Street, 400015 Cluj-Napoca, Romania; stefan.bilasco@ubbcluj.ro

Abstract

Black truffle (Tuber aestivum Vitt.) is one of the most important species of ascomycete fungus in Europe due to its growing conditions and high price. As previous studies show, soil conditions are among the main factors which largely influence the spread and development of black truffle. Therefore, the main objectives of this study were to identify soil conditions, climate and forest vegetation which influence both directly and indirectly the distribution of black truffle (Tuber aestivum Vitt.) in Romania, and to identify the areas from the Subcarpathian Hills of Transylvania which support black truffle growth. Geo-informational techniques have been used for this purpose to integrate spatial databases and to identify through spatial analysis the suitability of different areas for the growth of black truffles. The results of land favorability analyses showed the highest favorability level for Quercus robur (91.9%), followed by Carpinus betulus (89.24%) and Fraxinus excelsior (88.4%). The lowest favorability percentage (5.83%) was recorded for Tilia platyphyllos due to climate conditions.

Key words: Tuber aestivum Vitt., soil humidity, G.I.S. modelling

INTRODUCTION

According to previous reports (Linderman 1988, Streiblová et al., 2010, Gryndler M. and Hršelová, 2012, Benucci et al., 2012) the spread and development of black truffle species are highly influenced by soil conditions as well as certain invertebrate animal species with direct and indirect influence on soil such as protozoa, worms, and earthworms or other insects whose influence can be felt up to 15 centimeters soil depth. These creatures contribute to the aeration of the soil and increase its fertility by participating directly in the decomposition of organic matter. Earthworms improve soil aggregation and porosity through the channels they leave, thus becoming indirect truffle indicators in the area (acidity level preferred by earthworms is similar to pH range of 5.5-8.5 considered optimal for truffles).

Black truffle (*Tuber aestivum*) grows in symbiosis with forest tree species belonging to the following genera: *Castanea, Citrus, Corylus, Fagus, Ostrya, Tilia, Picea, Carya, Pinus* and Quercus (Wang et al., 2008,

Wedén et al., 2009; Chevalier 2009, Benucci et al., 2012; Stobbe et al., 2013). In Europe 30 tree species were identified as potential hosts for truffle growth (Ceruti et al. 2003).

Further studies were carried out by Moser et al. (2017) regarding vegetation composition in favorable areas for black truffle in the southwestern part of Germany and in the western part of Switzerland, were the identified forest stands suitable for *Tuber aestivum* were consisted of *Fagus sylvatica, Carpinus betulus* and *Ostrya carpinifolia,* as well as a series of grass and shrub species indicating the existence of a warm and dry climate, and an alkaline soil developed on a limestone rock substrate.

Other studies suggest that in the vicinity of the colonized trees there is a low density of herbaceous species induced by the allelopathic compounds released by the mycelium of *Tuber aestivum* in the soil (Streiblova et al., 2012). The knowledge about these dependent relationships and reciprocity are indispensable to determine the existence of these valuable fungi as well as to improve their productivity (De Miguel et al., 2014). In this context, it was found that grassland vegetation was associated with forest tree species such as *Fagus sylvatica*, *Quercus robur* and *Corylus avellana* being dominant and a very good indicator for estimating the potential existence of *Tuber aestivum* species depending on the season and phenological characteristics of the fungi. The most common host tree species were found to be: *Fagus sylvatica*, *Carpinus betulus*, *Quercus sp.*, *Abies alba*, *Picea abies*, *Pinus sylvestris*, *Betula pendula*, *Ostrya carpinifolia* and *Tillia sp.* (Moser et al., 2017).

Previous reports also show that the presence of termophyte species such as Cornus sp., Corvlus avellana, Crataegus sp., Prunus spinosa, etc. is a good indicator for the existence of Tuber aestivum in any areas (Hall, 1988; Paolocci et al., 1997, Willner, 2002; Baciarelli et al., 2006; Reyna et al., 2009; Murat et al., 2008; Reyna et al., 2014; Berch and Bonito 2016; Bonet et al., 2006; García-Montero et al., 2009; Willner et al., 2017). The occurrence of Fagus sylvatica together with termophyte species were also considered reliable indicators of the existence of Tuber aestivum species in Italy (Bencivenga et al., 1995b), Southern Germany (Stobbe et al., 2012) and Hungary (Hilszczańska et al., 2014) as well. Tuber aestivum can also develop symbiotic relationships with *Picea abies or Abies alba*, indicating thus a high phenotypic plasticity of this fungi. However, it was observed that these territories were previously covered by beech forests with cold climate and limestone soils (Stobbe et al., 2013). According to Gryndler et al. (2014) Tuber aestivum can also establish symbiotic relationships with herbaceous species as well such as Geum urbanum or Hedera helix as nonhost species for Tuber aestivum.

Taking into consideration black truffle requirements against environmental conditions, the main aim of this research was to identify land favorability induced by forest vegetation for black truffle growth across the Subcarpathian Hills of Transylvania using spatial analysis and quantitative modeling G.I.S techniques.

MATERIAL AND METHOD

Among forest tree species, the most widespread across the studied area were: *Tilia platyphyllos*, *Carpinus betulus*, *Quercus robur*, *Acer pseudoplatanus*, *Fraxinus excelsior*. To determine the favorability of these areas for black truffle growth, land favorability maps were generated based on climatic parameters including annual average temperature, the amount of annual average precipitation, the length of the bioactive period, relative atmospheric humidity and wind regime.

Relief features were also analyzed such as altitude, slope orientation, and soil characteristics: the degree of base saturation (%), soil acidity (pH), type of humus, edaphic volume (m^3/m^2) , the degree of soil compaction and soil texture, according to the ecological records (Stănescu, 1987) to determine land favorability levels for *Tuber aestivum* growth.

Regarding climate conditions and their influence on forest tree species favorability for black truffle growth. the annual average temperature of the study area ranged from 5.09 to 10.07 °C, providing favorable conditions for all the investigated tree species, being divided in three different favorability classes such as low, medium and high (Table 1).

The average amount of rainfall ranged between 640 and 1049 mm /year recorded for the analyzed territories, which offers highly favorable conditions for *Carpinus betulus*, *Quercus robur* and *Fraxinus excelsior* and average favorability for *Tillia platyphyllos* and *Acer pseudoplatanus* found in the most part of the territory (Gilman and Watson, 1993; Taheri et al., 2013; Clinovschi, 2015). Beside temperature and precipitation, soil acidity is a very important growth indicator of truffle species. Across the investigated territories, the pH of the soil varied between 5.48-7.7 in most parts of the Subcarpathian Hills of Transylvania providing thus a high favorability level for the trees analyzed from this point of view.

Table 1

	Favorability classes	Tilia platyphyllos	Carpinus betulus	Quercus robur	Acer pseudoplatanus	Fraxinus excelsior
Annual average temperature	High	9-11	8-10	8-10	5-8	>8
	Medium	8-9, >11	6-8, >11	7-8,>11	4-5, 8-10	6-8
	Low	<8	<6	<7	<5,>10	<6
Annual average rainfall	High	500-600	600-800	600-800	800-1100	500-700
	Medium	400-500, 600-700	500-600, 800-900	500-600 800-100	700-800, 1100-1300	700-900, 400-500
	Low	<400	<500,>900	<500, >1000	<700, >1300	>900, <400
Length of bioactive period	High	>7	>6	>8	5-7	>7
	Medium	6-7	5-6	5-8	4-5,>7	5-7
	Low	<6	<5	<5	<3	<5

Favorability classes of forest tree species based on climatic conditions across the Subcarpathian Hills of Transylvania

The degree of soil compaction divides soil classes as lightly, moderately and heavily compacted. The Subcarpathian Hills of Transylvania are moderately compacted, while lightly compacted soils have been identified in the valley areas of Bistrița and Homoroadelor hills, which provide a high favorability level for the investigated forest tree species. Moderately compacted soils provide an average favorability especially for lime and ash trees.

Table 2

Forest tree favorability classes based on relief conditions of the Subcarpathian Hills of Transylvania

	or riunsjivania							
	Favorability classes	Tilia platyphyllos	Carpinus betulus	Quercus robur	Acer pseudoplatanus	Fraxinus excelsior		
6	High	100-500	300-700	200-600	800-1200	<1100		
ltitud	Medium	<100, 500-600	700-1000	100-200 600-800	600-800, 1200-1600	1100-1300		
A	Low	>600	<200 >1000	<100 >800	<600, >1600	>1300		
ope Exposition	High	Sunny Partly sunny	Partly shady Shady	Sunny Partly sunny Depression areas	Partly shady Shady	Partly shady Shady Depression areas		
	Medium	Partly shady	Partly sunny Depression areas	Partly shady Shady	Sunny Partly sunny Depression areas	Partly sunny		
S	Low	Depression areas Shady	Sunny	-	-	Sunny		

Due to the clayey texture of the investigated area which represents 22.5% of the territory, these areas proved to be suitable for the growth of ash trees (*Fraxinus excelsior*). For *Carpinus betulus, Acer pseudoplatanus*,

Quercus robur and *Tillia platyphylos* medium favorability levels were determined due to the clay texture of the soil (Table 3).

Table 3

Transylvana								
	Favorabil	Tilia	Carpinus	Quercus	Acer	Fraxinus		
	ity classes	platyphyllos	betulus	robur	pseudoplatanus	excelsior		
	High	>70	>60	>55	>55	>65		
Base saturat on	Medium	50-70	40-60	35-55	35-55	45-65		
	Low	<50	<40	<35	<35	<45		
Acidi ty	High	6.6-7	>6.3	6.4-6.8	5.8-7	6.6-7		
	Medium	4.6-6.6	5.6-6.3	5.2-6.4;>6.8	4.8-5.8;>7	6-6.6;>7		
	Low	<4.6	<5.6	<5.2	<4.8	<6		
Edaphi c volume	High	>0.45	>0.45	>0.60	>0.30	>0.45		
	Medium	0.30-0.45	0.15-0.45	0.45-0.60	0.15-0.30	0.15-0.45		
	Low	<0.30	<0.15	<0.45	<0.15	<0.15		
Compaction	High	Loamy	Sandy-loam	Sandy-loam Loamy	Sandy-loam Loamy	Loamy, Loamy-clay		
	Medium	Loose	Loose Moderately compact	Loose Moderately compact compact	Very loose Loosen	Loose, Moderately compact		
	Low	Very loose Moderately compact	Very loose	Very loose	Moderately compact	Very loose, compact		
Texture	High	Compact, Very compact	Compact, Very compact	Very compact	Very compact, compact	Very compact		
	Medium	Loamy	Sandy-loam	Sandy-loam Loamy	Sandy-loam Loamy	Loamy, Loamy-clay		
	Low	Sandy-loam, Loamy-sand, Loamy-clay	Loamy- sand, Loamy-clay	Clay-loam Loamy-Clay	Clay-loam	Loamy-clay, Sandy-loam		
		Sandy, Clay-loam, Clay	Sandy Loamy-clay Clay	Clay Sandy Loamy-sand	Sandy, Loamy-sand Loamy-clay Clay	Sandy, Loamy-sand Clay		

Forest tree favorability classes based on soil conditions of the Subcarpathian Hills of

Carpinus betulus L. is considered a forest tree species with low drought resistance (Hamerlynch et al. 2002; Pallardy 2008; Clinovschi 2015). The length of the bioactive period between 3-6 months in these regions represents a limiting factor which ranks the investigated territories in a medium favorability class for *Acer pseudoplatanus* and a low favorability for the rest of the investigated forest tree species.

RESULTS AND DISCUSSION

Our results show that the area of low and medium hills characterized by an altitude of 250-500 m, having a 46% spread across the investigated area offer a high favorability level for the growth of lime, hornbeam and oak, while the areas of high and very high hills (501-1000 m) offer high favorability conditions for the development of sycamore tree and an average favorability for hornbeam trees. In Romania, natural symbiotic relationships were observed between black truffle and hazelnut (*Corylus avellana*) which led to the establishments of new truffle plantations which success is still uncertain at national level, due to the lack of scientific information regarding the economic efficiency of this type of plantations especially due to the long period until the first harvest (5 to 10 years). High favorability for lime is induced by soils with a clay texture for 7.4% of the territory and for *Carpinus betulus*, *Quercus robur* and *Acer pseudoplatanus* on a clay-sandy texture (for 11.9% of the territory). The results of land favorability analyses revealed a very good favorability level for *Quercus robur* (91.9% of the area), followed by *Carpinus betulus* (89.24%) and *Fraxinus excelsior* (88.4%) as shown in Fig. 1.



The distribution of favorability classes and favorability maps for each tree species are presented in Figures 3-6. The lowest favorability percentage (5.83%) was recorded for *Tilia platyphyllos* due to climate conditions. The importance and usefulness of GIS technologies in the identification of favorability classes and limitations for diverse forest tree species had already been demonstrated in previous studies carried out across the Transylvanian Valley as described by Rosca et al. (2017).

Acknowledgments

The present work has received financial support through the project: Entrepreneurship for innovation through doctoral and postdoctoral research, POCU/360/6/13/123886 co-financed by the European Social Fund, through the Operational Program for Human Capital 2014- 2020. All authors have equal contribution to this paper.

REFERENCES

- 1. Baciarelli-Falini, L., Rubini, A., Riccioni, C., & Paolocci, F., 2006, Morphological and molecular analyses of ectomycorrhizal diversity in a man-made T. melanosporum plantation: description of novel truffle-like morphotypes. Mycorrhiza, 16(7), pp. 475-484.
- Bencivenga, M., Di Massimo, G., Donnini, D., Tanfulli, M., 1995a. Confronto tra la vegetazione delle tartufaie di *Tuber aestivum Vitt.*, T. magnatum Pico e T. melanosporum Vitt. nell'Italia centrale. Micol. Ital. 24, pp. 87-95.
- Benucci Gmn, Bonito G, Baciarelli Lf, Bencivenga M, 2012, Mycorrhization of Pecan trees (*Carya illinoinensis*) Fith commercial truffle species: *Tuber aestivum Vittad. and Tuber borchii Vittad.* Mycorrhiza 22, pp. 383-392.
- 4. Berch, S. M., & Bonito, G., 2016, Truffle diversity (Tuber, Tuberaceae) in British Columbia. Mycorrhiza, 26(6), pp. 587-594.
- Bonet, J. A., Fischer, C. R., & Colinas, C., 2006, Cultivation of black truffle to promote reforestation and land-use stability. Agronomy for sustainable development, 26(1), pp. 69-76.
- Ceruti A., Fontana A., Nosenzo C., 2003, The specie europee del genere Tuber. Una revision storica. – 467 p. Torino.
- 7. Chevalier, G., 2010, La truffe d'Europe (*Tuber aestivum*): limites geographiques, ecologie et culture. Osterr. Z. Pilzk. 19, pp. 249-259.
- 8. Clinovschi F., 2015, Dendrologie. Suceava: Editura Universitatii din Suceava; p. 296.
- 9. De Miguel, A.M., Agueda, B., Sanchez, S., Parlade, J., 2014, Ectomycorrhizal fungus diversity and community structure with natural and cultivated truffle hosts: applying lessons learned to future truffle culture. Mycorrhiza 24, S5eS18.
- Garcia-Montero, L., Moreno, D., Monleon, V., Arredondo-Ruiz, F., 2014, Natural production of *Tuber aestivum* in central Spain: Pinus spp. versus *Quercus spp.* Brûles, Forest Systems, 23(2), pp. 394-399.
- 11. Gilman Ef, Watson Dg., 1993, Carpinus betulus, Fact Sheet ST-118, a series of the Environmental Horticulture Department, Florida Cooperative Extension Service. Florida: Institute of Food and Agricultural Sciences, University of Florida Cooperative Extension Service.
- 12. Gryndler M., Hršelová, H., 2012, Isolation of bacteria from ectomycorrhizae of *Tuber* aestivum Vittad., ACTA MYCOLOGICA, 47 (2), pp. 155–160.
- Gryndler M. Černá L. Bukovská P. Hrš-Elová H. Jansa J., 2014, *Tuber aestivum* association with non-host roots, Mycorrhiza, DOI 10.1007/s00572-014-0580-9
- 14. Hall, I. R., 1988, The black truffle: its history, uses and cultivation. Ministry of Agriculture and Fisheries, Invermay Agricultural Centre.
- Hamerlynck Ep, Mcauliff E, Mcdonald Ev, Smith Sd., 2002, Ecological responses of two Mojave Desert shrubs to soil horizon development and soil water dynamics. Ecology 83:768-779.
- Hilszczanska, D., Rosa-Gruszecka, A., Szmidla, H., 2014, Characteristic of *Tuber spp*. localities in natural stands with emphasis on plant species composition. Acta Mycol. 49, pp. 267-277.
- 17. Linderman R.G., 1988, Mycorrhizal interactions with the rhizosphere microflora: the mycorrhizosphere effect. Phytopathology 78: pp. 366–371.
- Moser, B., Büntgen, U., Molinier, V., Peter, M., Sproll, L., Stobbe, U., Tegel, W., Egli, S., 2017, Ecological indicators of *Tuber aestivum* habitats in temperate European beech forests, Fungal Ecology 29, pp. 59-66.

- Murat, C., Zampieri, E., Vizzini, A., & Bonfante, P., 2008, Is the Perigord black truffle threatened by an invasive species? We dreaded it and it has happened!. New Phytologist, 178(4), pp. 699-702.
- 20. Pallardy SG., 2008, Pallardy physiology of woody plants. 3rd ed. San Diego, CA: AcademicPress.
- Paolocci, F., Rubini, A., Granetti, B., & Arcioni, S., 1997, Typing Tuber melanosporum and Chinese black truffle species by molecular markers. FEMS Microbiology Letters, 153(2), pp. 255-260.
- 22. Păcurar Horea, Marcel Dîrja, Mihai Buta, Ioan Păcurar, Sanda Roşca, Ștefan Bilaşco, 2018, Identification of Soils Factors Influence in the Distributions of *Tuber aestivum* in Transylvanian Subcarpathian Hill, Romania, Not Bot Horti Agrobo, 2019, 47(1), DOI:10.15835/nbha47111378.
- 23. Păcurar H., Dîrja, M., Păcurar, I., Roşca, S., Bilaşco, Şt., Negruşier, C., 2019, Ecological Influences On *Tuber Aestivum* Distribution In The Subcarpathian Region Of Transylvania, Bulletin of University of Agricultural Sciences and Veterinary Medicine. Horticulture.
- Reyna, S., & Garcia-Barreda, S., 2014, Black truffle cultivation: a global reality. Forest Systems, 23(2), pp. 317-328.
- 25. Reyna-Domenech, S., & García-Barreda, S., 2009, European black truffle: its potential role in agroforestry development in the marginal lands of Mediterranean calcareous mountains. In Agroforestry in Europe pp. 295-317, Springer, Dordrecht.
- 26. Roşca S., Bilaşco, Şt., Păcurar, I., Colniță, D., Fodorean, I., Vescan, I., Petrea D., Păcurar, H., 2017, Quantitative evaluation of forest favourability using GIS database in a hill area in the Transylvania Depression, Romania, Geomatics, Natural Hazards And Risk, pp. 1-22, https://doi.org/10.1080/19475705.2017.1401012
- Sestraş, P., Bilaşco, Ş., Roşca, S., Naş, S., Bondrea, M. V., Gâlgău, R., & Cîmpeanu, S. M., 2019, Landslides Susceptibility Assessment Based on GIS Statistical Bivariate Analysis in the Hills Surrounding a Metropolitan Area. Sustainability, 11(5), 1362.
- 28. Stănescu, V., 1979, Dendrologie, Editura Didactică și Pedagogică, București.
- Stobbe U, Egli S, Tegel W, Peter M, Sproll L, Büntgen U, 2013, Potential and limitations of Burgundy truffle cultivation. Mini-Review. Appl Microbiol Biotechnol 97:5215-5224.
- Stobbe, U., Büntgen, U., Sproll, L., Tegel, W., Egli, S., Fink, S., 2012, Spatial distribution and ecological variation of re-discovered German truffle habitats. Fungal Ecol. 5, 591-599.
- 31. Stobbe, U., Stobbe, A., Sproll, L., Tegel, W., Peter, M., Büntgen, U., Egli, S., 2013b. New evidence for the symbiosis between *Tuber aestivum* and *Picea abies*. Mycorrhiza 23, pp. 669-673.
- Streiblová, E., Gryndlerová, H., Valda, S., Gryndler, M., 2010, *Tuber aestivum* hypogeous fungus neglected in the Czech Republic. A review., CZECH MYCOL. 61(2), pp.163–17383.
- Streiblova, E., Gryndlerova, H., Gryndler, M., 2012, Truffle brûle: an efficient fungal life strategy. Fems Microbiol. Ecol. 80, 1e8.
- Taheri Abkenar K, Chafjiri Fs, Sardabrud Sm., 2013, Study of Carpinus betulus die back distribution using topographic factors. Int J Environ Resour Res. 1(2), pp.181–189.
- Wang, Q., Adiku, S., Tenhunen, J., & Granier, A. 2005, On the relationship of NDVI with leaf area index in a deciduous forest site. Remote sensing of environment, 94(2), pp. 244-255.
- 36. Willner, W., 2002, Syntaxonomische Revision der südmitteleuropaischen Buchenwalder. Phytocoenologia 32, pp. 337-453.