



A large-scale analysis of soil and bioclimatic factors affecting the infestation level of tobacco (*Nicotiana tabacum* L.) by *Phelipanche* species



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ARTICLE INFO

Article history:

Received 6 April 2015

Received in revised form

8 January 2016

Accepted 14 January 2016

Available online 5 February 2016

Keywords:

Broomrape

Climatic and soil factors

GIS

Multivariate analyses

ABSTRACT

Soil and bioclimatic factors constitute key factors that influence the interaction between host plants and broomrape. The identification and quantification of the relationship between environmental variables and the level of infestation with *Phelipanche aegyptiaca* and *Phelipanche ramosa* was examined for 144 tobacco fields in surveys during two years 2003–2004 in Greece. The distribution of *Phelipanche* species was mapped using a Geographical Information System to reveal variability in the infestation level between the species, the cultivated types and the sampling areas. To assess the effect of abiotic factors on the level of infestation with *Phelipanche* species, soil and bioclimatic parameters were taken into account: soil texture, pH, organic matter content (OM), total humidity index (THI) and growing degree days (GDD). Univariate and multivariate statistics were used to examine the influence of the above parameters on *Phelipanche* intensity. ANOVA showed that the infestation varied significantly between the sampling areas and *Phelipanche* species, while significant differences were also detected among the regions in terms of the studied abiotic parameters. Correlation analysis demonstrated that the level of *Phelipanche* infestation correlated negatively with pH, THI and positively with OM. Multiple regression analysis suggested that soil and climatic variables together explained 63% of the infestation variance. Discriminant functions accounted for 90% of the total variation in the dataset. All statistical analyses demonstrated that pH, THI and OM were the most decisive variables for the severity of infestation. The two *Phelipanche* species were clearly discriminated, with *P. ramosa* being found at higher and *P. aegyptiaca* at a lower level of infestation. A parasitism specification was also observed between the two *Phelipanche* species and tobacco types. The findings provide constructive information for modelling broomrape infestation based on abiotic factors and create a baseline for future monitoring of broomrape distribution.

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1. Introduction

Achlorophyllous root parasites such as broomrapes reduce yield by withdrawing water, nutrients and assimilates from the host vascular system. *Phelipanche ramosa* (L.) Pomel (branched broomrape) and *Phelipanche aegyptiaca* (Pers.) Pomel (Egyptian broomrape) have been registered as the most noxious root-parasitic weeds in Europe and neighbouring regions since they parasitize

crops from different botanical families (Parker, 2009). *Phelipanche* species are the branched types and closely related since they have similar host range and share many common phenological and physiological characteristics (Parker and Riches, 1993). Until recently, all broomrape species were treated under a single genus, *Orobanche*. Since 2009, however a new nomenclature of *Orobanche* and *Phelipanche* has been adopted, because major morphological, karyological and molecular differences were observed between the two taxa (Joel, 2009). *P. ramosa* and *P. aegyptiaca* were formerly known as *Orobanche ramosa* and *Orobanche aegyptiaca*.

Tobacco, tomato and canola are parasitized by *Phelipanche* spp. in different cultivation zones throughout Greece (Economou et al., 2007). Major problems have been encountered in areas where tobacco monoculture is common practice, with representatives being

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Oriental (sun-cured) and Virginia (flue-cured) types, the two major tobacco types cultivated at national level. Farmers very often have to deal with noteworthy yield losses due to the lack of effective control measures (Economou et al., 2005). Even though tobacco cultivation has decreased in Europe during the past decade due to EU Common Agricultural Practices (CAP), there is a clear trend for increasing tobacco production in Greece in view of the high qualitative and quantitative characteristics of the products (Hellenic Ministry of Rural Development and Food (2013)). Since control of broomrapes is very difficult by means of agronomic practices and application of herbicides, a greater understanding of the spatial distribution of broomrape in farmlands should provide insights into their ecological requirements and their potential management (Oveisi et al., 2010). Furthermore, the need to prevent the infestation of plant hosts by developing resistant crops has been stated by Rubiales (2003).

The potential spread of this holoparasite in broomrape-free areas where potential host plants are grown constitutes an important issue. New forms of broomrape species, such as *Orobancha foetida*, have evolved in Morocco and Tunisia, shifting from being wild parasitic plants to aggressive parasitic weeds (Vaz Patto et al., 2008; Abbes et al., 2007). Furthermore, different scenarios for the invasive potential of broomrape species have been analyzed thoroughly on a wide latitudinal basis in the light of global warming (Mohamed et al., 2006; Grenz and Sauerborn, 2007).

Broomrape occurrence in fields is the result of an interaction among various biotic and abiotic parameters. The host constitutes the most important biotic factor since via its root stimulants it induces the germination of broomrape seeds (Pieters and Aalders, 1986). Abiotic factors, such as edaphic and climatic characteristics, have been found to play a major role in the host–parasite interaction, since they can act directly on all phases of the parasite life cycle, and indirectly through the host plant (Moral et al., 2015; Ter Borg, 1986). Different broomrape species require different conditions for seed germination since their hosts have different geographical ranges (Kebreab and Murdoch, 1999a).

Among meteorological parameters, temperature and moisture play a key role in germination, the first critical step in the life cycle of these holoparasites (Mohamed et al., 2006; Kebreab and Murdoch, 1999b). Simple weather variables constitute static measurements that are not related to plant growth (Valencia-Barrera et al., 2002). On the other hand, bioclimatic indices take account of more than one measure at the same time, or their temporal evolution, or the plant response (Chamayou, 1994). Broomrape distribution and magnitude of infestation constitute climate-sensitive processes which could be better interpreted through bioclimatic indices instead of separating the individual effects of different meteorological parameters. Moreover, the host–parasite interaction is a dynamic and complex phenomenon which cannot be attributed to a single weather variable (Parker and Riches, 1993).

Growing degree days (GDD), and humidity and aridity indices are climatically derived variables which have a major direct ecophysiological impact on plant species (Randin et al., 2009). Eizenberg et al. (2004) utilized temperature dependency to develop a day-degree model for predicting the parasitism of *Orobancha minor* in red clover. Ephrath and Eizenberg (2010) also quantified the dynamics of the host–parasite relationship between sunflower and two broomrape species as related to thermal time as measured by GDD.

Humidity and drought indices, which characterize water resources over a large geographical area, incorporate potential evapotranspiration and precipitation variables (Tsakiris and Vangelis, 2005; Tiggas et al., 2013). Similar indices have been used for climatic classification of areas globally and contribute to elucidating mechanisms determining species' distributions

(Papadakis, 1975; Villers-Ruiz and Castañeda-Aguado, 2013). The knowledge of bioclimatic conditions that prevail in a region has great significance for management of parasitic weeds, since their further expansion beyond their present geographical ranges could be prevented and broomrape-free areas maintained (Grenz and Sauerborn, 2007). In addition, water availability in broomrape-infested regions affects the conditioning phase, a key factor that renders water-imbibed broomrape seeds responsive to germination stimulants exuded by host plants (Kebreab and Murdoch, 2001).

The soils on which *Phelipanche* species occur in the Mediterranean region are mainly alkaline (Parker and Riches, 1993). pH constitutes a soil parameter involved in the stabilization of stimulant molecules exuded from host roots which are able to trigger germination of *Orobancha* seeds. Both absorption and degradation of such molecules seem to be influenced by soil structure and properties (Bailey and White, 1970; Babiker et al., 1988).

Only a few recent studies have explored the environmental determinants of the geographic distribution of broomrapes. Cantamutto et al. (2012) and Miladinovic et al. (2012) in exploratory studies showed the effect of geographic and micro-abiotic parameters on *Orobancha cumana* distribution. Grenz and Sauerborn (2006, 2007), through simulation models, also combined abiotic parameters to predict *Orobancha crenata* expansion on a climate region basis. All the aforementioned abiotic parameters change in space and time and determine the spatial heterogeneity. If these parameters are imported into a Geographical Information System (GIS) database, maps can be produced for locating and exploring high-intensity broomrape infestations. This approach gives insight into the landscape conditions that favour the acclimatization of broomrape populations and which increase the magnitude of infestation.

Several studies have explored the environmental determinants of the geographic distribution of *O. cumana* (Cantamutto et al., 2012; Miladinovic et al., 2012) and *O. crenata* (Grenz and Sauerborn, 2007). As far as we are aware, no information exists on the influence of abiotic parameters on the level of infestation with *Phelipanche* species, particularly on the specific contribution of each of these parameters in terms of the variation in infestation level.

In order to understand the multiple interactions among hosts, broomrapes and the environment, ecogeographic surveys were conducted in the main tobacco zones in Greece, measuring the level of infestation with broomrapes on tobacco plants and its relation to specific abiotic parameters that defined each site. The objectives of the research were to: a) map the observed level of infestation with the aid of spatial technologies, b) identify the most important abiotic parameter(s) that influenced the extent of parasitism in the infected areas after initial broomrape emergence, c) develop a statistical model to predict the level of broomrape infestation via multivariate statistical techniques, and d) detect differences in broomrape resistance between Oriental (Basmas) and Virginia tobacco types.

2. Materials and methods

2.1. Study areas

Seven tobacco zones of Greece, namely Katerini, Kozani, Voio, Domokos, Lamia, Agrinio and Nafplio, were thoroughly screened for natural infestation by *P. ramosa* and *P. aegyptiaca* during consecutive years, 2003 and 2004. The location of each collection site is depicted in Fig. 1. Detailed profiles of the study areas are presented in Table 1. Agrinio was in the lowest (85 m) and Voio in the highest (691 m) land. Domokos was characterized by the lowest

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