



Weed seedbank dynamics in Mediterranean organic horticulture



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ABSTRACT

Managing weeds in organic cropping systems is notoriously difficult. We analyzed the seed bank dynamics in vegetable crop rotations carried out in both the typical Mediterranean scenarios of the open field and greenhouse. The lower seed bank detected in the greenhouse (about 53,000 seeds m^{-2}) showed a higher emergence rate than the seed bank found in the open field, which was almost twice the size. This higher emergence rate was due both to: i) the more favorable temperature in the greenhouse, and ii) the less depth-mediated soil-imposed dormancy, due to the seed bank being at a lower depth as a result of minimum tillage. A strong infestation of *Cyperus rotundus* was found in the greenhouse again, due to the tuber arrangement in the shallowest soil layer (0–10 cm). The ubiquitous and abundant *Amaranthus retroflexus*, taken as a model to investigate the dormancy-status of exhumed seeds showed that the lower emergence rate in the open field, was not due to dormancy but exclusively to depth-mediated soil inhibition. In addition, the calculation of various indexes of the weed seed bank botanical composition highlighted that the more quantitatively abundant greenhouse weed community was formed by more graminoid species (poaceae and cyperaceae), perennial species, C_4 photosynthetic pathway species and by lower biodiversity. Lastly agronomic strategies are proposed which could improve the weed control sustainability in organic cropping systems.

1. Introduction

Organic farming is likely to be majorly boosted in the European Union (Siderer et al., 2005) in view of its evident world-wide success. About 11.5 million hectares are grown with organic cropping systems in Europe, constituting 2.4% of the continent's agricultural land (Willer and Schaack, 2015). The large increase in organic farming stems from the need to: i) preserve the earning capacity of farmers in a world that requires fewer producers to feed the well-fed areas of the world's population; ii) preserve the rural countryside; iii) use cultivation methods that will conserve the soil and contribute to sustainability (Mayfield et al., 2001). Although the productivity of organic farming is sometimes less than conventional farming (Mäder et al., 2002), it shows benefits in terms of soil fertility sustainability and its higher biodiversity (Hole et al., 2005). It thus constitutes an effective ecosystem service (Sandhu et al., 2010) both for humans and the environment.

The main problem with organic cropping systems lies in the biotic adversity (Letourneau and Van Bruggen, 2006) especially in terms of the defense against weeds (Bond and Grundy, 2001).

The long-term sustainability of weed control strongly depends on the effectiveness and complementarity (Hatcher and Melander, 2003) of the methods available: mechanical (Van der Weide et al., 2008), physical (Ascard, 1995) agronomic (Stockdale et al., 2001) and

biological (Charudattan and Dinooor, 2000). Each type of weed control is not only aimed at eliminating the weeds that emerge, but also at limiting their seed production (Webster et al., 2003) and dispersal, thus minimizing the annual input of seeds in the soil (Kegode et al., 1999).

Periodical weed field emergence strongly depends on the amount, botanical composition and vertical arrangement of the seed bank. The vertical arrangement is key since only the shallowest seed bank, above all for the light-dependent small seeds (Milberg et al., 2000), is capable of germination and seedling emergence. In addition, high seed longevity is typical in many weeds (Burnside et al., 1996), and is a further feature which facilitates their long-term persistence in the soil. From a quantitative point of view, in spite of the extreme agronomic and environmental variability, the number of viable seeds in the soil ranges from about 2300 seeds m^{-2} (Sosnoskie et al., 2006), or even less (Cardina et al., 1991), in conventional cropping systems, to almost 10,000–20,000 seeds m^{-2} during the conversion towards organic management of both industrial (Albrecht, 2005) and vegetable crops (Sjursen, 2001). After long-term organic management this reaches 25,000 seeds m^{-2} (Teasdale et al., 2004) and sometimes exceeds even 50,000 seeds m^{-2} (Benvenuti et al., 2001a).

This seed bank accumulation occurs since most annually produced weed seeds undergo dormancy in the soil due to genetic, species-dependent (Allen and Meyer, 1998), and/or environmental soil-depth

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mediated (Benvenuti et al., 2001b) mechanisms. Consequently knowing the quantity, botanical composition and vertical distribution of buried seeds can play a crucial role in predicting the likely weed emergence dynamics. Basically, this information facilitates planning weed control strategies before weed emergence. In other words, a seed bank evaluation acts as a non-chemical weed management method, thus making it an useful agronomic tool.

Currently, despite the considerable agronomic importance of organic horticulture, there is scarce information on the weed seed bank evaluation and dynamics in this sustainable cropping system.

In the Mediterranean environment, horticultural products are typically used both in open fields and in greenhouses through short and complex crop rotations. Often greenhouse conditions are associated with heavy weed infestation, which sometimes it makes difficult to obtain a sustainable management. The main agro-ecological differences consist of both higher temperature dynamics, due to the well-known greenhouse effect, and the diversified soil management. Within polyethylene tunnels for example, where tractors cannot be used, a minimum tillage is usually performed, using a small rotary hoe (Bonanomi et al., 2014).

It is not clear whether, and in what way, these two different agro-ecological conditions, temperature and soil tillage, can determine a diversified weed seed bank and seedling emergence dynamics.

The aims of this work were: i) to quantify the seed bank of a typical organic crop rotation of vegetable species conducted in the open field or in the greenhouse, ii) to test their emergence rate, iii) to evaluate the effects of cropping systems in terms of botanical structure, biodiversity and agronomic sustainability.

2. Materials and methods

2.1. Agricultural environment and cropping systems

Trials were carried out during 2014 at Ortonovo (Sarzana SP, Italy; 44.09° N, 10.04° E) in an organic Farm specialized in vegetable crop production for twenty years. Planting of the several vegetable crops (transplanting except for Spinach and Faba bean) were conducted in the open field and in a greenhouse (randomized complete block design for each cropping systems) in both cases using organic cropping systems (Table 1). Weed control was achieved exclusively by mechanical means and soil fertilization was carried out using only organic products. Note that in the case of the greenhouse (polyethylene tunnels) cultivation, soil tillage was carried by rotary hoeing since the tunnel height was limiting for the tractor (2.9 m in the middle but much lower at the sides). Soil tillage in the open field cultivation was managed by plowing

Table 1
Agronomic information about the organic cropping system adopted in greenhouse and open field conditions.

Agrotechniques	Cropping system	
	Greenhouse	Open field
Locality	Ortonovo (SP) Italy	
Geographic coordinates	44.09 N, 10.04 E	
Cultivars	International and local germplasm certified as “organic seed”	
Planting techniques	Transplanting except Spinach, Beet and Faba bean (seed sowing)	
Irrigation	Drip irrigation	
Soil tillage	Rototilling (10 cm)	Plowing (30 cm) + rototilling (10 cm)
Soil texture	Sandy loam	
Fertilization	Organic (commercial pelleted manure)	
Weed control	Mechanical inter-row hoeing	
Greenhouse type	Polyethylene tunnels (4.20 m width x 40 m length)	

(30 cm).

Table 2 shows the various crop rotations (3–4 crops per year) carried out in both the greenhouse and open field (three each) over the last ten years. In each rotation (repeated every 2 years), both microtherm (i.e. spinach, strawberry, fennel, etc.) and macrotherm (e.g. tomato, eggplant, squash) were cropped.

2.2. Measurement of soil temperature

During the experiment, temperature probes (HOBO[®] Pedant temperature data logger, Campbell Scientific, UK) were placed into the soil (3 cm depth) of both cropping systems: open field and greenhouse (three replicates each). The probes (one read per hour) were used throughout 2014.

2.3. Weed seedbank analysis

A seedbank evaluation was carried out at the end of March 2014 (after the seedbed preparation of each spring crop) in both the open field and greenhouse of each crop rotations. The preceding crops were the summer crops of the second year indicated in Table 1 (with the only exception of rotation 3 carried out in open field in this case preceded by the autumn crop fennel). For this analysis, large experimental plots measuring 300 m² were set up, divided into four sub-plots of 25 m² (5 × 5 m). The four selected fields were adjacent and analogous in terms of physical soil characteristics, fertility and drainage. Soil samples were taken using a metal probe which extracted 10 cm long and 4 cm Ø soil cores. Sampling was done at the end of winter (last ten days of February). For each sub-plot, 30 cores were taken at each of the following depths: 0–10, 10–20 and 20–30 cm (total of 90 soil cores per sub-plot). Seeds were extracted from the cores by pre-treating the soil for roughly 10 h in 5 g of sodium hexametaphosphate solution. This resulted in the dispersal of the soil colloid matrix, facilitating the subsequent washing phases. Washing was performed according to a previously adopted methodology (Benvenuti and Macchia, 2006) using a pressure adjustable hydrojet (20–120 bar) in order to regulate the spray force according to the need, thereby averting possible damage to the seeds. Soil samples were washed inside metal cylinders (5 cm diameter and 50 cm long) closed on one side by a removable stopper equipped with a fine metallic mesh (250 µm). The material extracted (seeds, sand, plant residues, etc.) was separated manually by a backlit magnifying glass (8×). Seeds were then identified using an optical microscope (45×, Model Optech Biostar 5, Optech Scientific Instruments, Thame, Oxfordshire, UK). Seed weight was determined by weighing extracted seeds, chosen randomly, according to ISTA rules for seed testing (ISTA, 1999).

2.4. Seedbank germination and dormancy evaluation

Extracted seeds of the most abundant weed species (*A. retroflexus*) were subjected to germination tests in order to investigate their dormancy and viability characteristics. Seeds were thus placed in Petri dishes equipped with filter paper (Whatmann no. 1) moistened with distilled water, and incubated in climatic cabinets with a photoperiod of 12/12 h (alternating dark-light). The incubation temperature was 30 °C according to the temperature requirements for the germination of this species (Ghorbani et al., 1999). A light source of about 200 µmol m⁻² s⁻¹ was obtained from fluorescent tubes (PHILIPS THL 20W/33). The germinated seed count was completed four weeks when germination had essentially finished. Ungerminated seeds were categorized as not dormant, and underwent chilling (4° C in the dark, on moistened filter paper for one month) to induce dormancy-breaking and were then re-incubated under the same, previously described, conditions. Seeds that germinated following this treatment were categorized as light dormant. Any seeds that remained ungerminated despite this treatment, were subjected to the seed-crushing test (Sawma

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