



## Research paper

Earthworm functional groups respond to the perennial energy cropping system of the cup plant (*Silphium perfoliatum* L.)

Quentin Schorpp\*, Stefan Schrader

Johann Heinrich von Thünen-Institute (TI), Federal Institute for Rural Areas, Forestry and Fisheries, Institute of Biodiversity, Bundesallee 50, D-38116 Braunschweig, Germany

## ARTICLE INFO

## Article history:

Received 21 September 2015

Received in revised form

8 February 2016

Accepted 10 February 2016

Available online xxx

## Keywords:

Biodiversity

Perennial energy crops

Earthworms

*Silphium perfoliatum*

Soil ecology

Long-term development

## ABSTRACT

Biodiversity is a major issue in the sustainable production of bioenergy from biomass, since it is threatened by land use change and intensification. Functional biodiversity in soils is a major contributor to ecosystem services, essential for human well-being. We assessed the long term development of earthworm communities in a chronosequence of the perennial bioenergy plant *Silphium perfoliatum* L. and compared them to traditional biomass production from maize. The cultivation period covered by the chronosequence was 1–9 years.

Abundances of anecic and endogeic earthworms increased with the age of the fields. Endogeic earthworms exhibited significant differences to younger stages later than anecic earthworms. A successive increase in biodiversity was observed with endogeic species of the genus *Octolasion* and epigeic species. They occurred in cup plant fields after 3 and 5 years, respectively. Overall, maize fields had decreased earthworm biomass and population densities as well as less functional diversity. Cropping systems of *S. perfoliatum* have positive ecological implications for soil biodiversity and according ecosystem services. Due to the facilitation of functional diversity, soil processes, such as water infiltration, erosion resistance and nutrient cycling, are expected to increase. Hence we suggest a potential for remediation of formerly intensively managed soils. *S. perfoliatum* is considered to be well suited for the diversification of bioenergy farming landscapes.

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## 1. Introduction

Recent biomass production is based on intensively cultivated traditional crops, especially maize. However, the large scale cultivation of maize and tight crop rotations stresses soil functions and impairs the sustainability of bioenergy generation. Diversification of agroecosystems using perennial energy crops is a promising strategy to counteract the adverse developments of recent biomass production [1–3]. Among these alternative energy crops is the cup plant (*Silphium perfoliatum* L.). Besides its competitive properties in biogas production, this tall perennial, flowering plant is considered to be of high ecological value, since its reduced management preserves the soil and allows for minimal pesticide application [4,5]. The absence of tillage and cultivation of at least 10 y implies almost no disturbance of the soil habitat. The resultant permanent rooting and ground cover could counteract soil erosion [6].

The seventh European Environmental Action Program [7] has recognized, that unsustainable land use fosters soil degradation and claims that biodiversity conservation and the protection of soil should be fully taken into account in decisions relating to renewable energy. However, studies on the link between biomass production, biodiversity and ecosystem services are scarce or concentrate mainly on *Miscanthus*, which has been shown to be of weak palatability and of intermediate value for soil organisms [8–10].

Soil organisms play an essential role in maintaining soil functions such as the degradation of organic matter, cycling of nutrients, sequestration of carbon and degradation of pollutants [11]. Therefore, soil biodiversity plays an inevitable part in restoring and securing soil ecosystem services. In this context, earthworms are among the most important soil organisms with significant influence on soil structure (e.g., formation of aggregates and pores) and the breakdown of organic matter (e.g., fragmentation, burial, and mixing of plant residues) [12].

Based on morphology, burrowing behavior, feeding ecology and microhabitat, earthworms are assigned to three functional groups:

\* Corresponding author.

E-mail address: [quentin.schorpp@ti.bund.de](mailto:quentin.schorpp@ti.bund.de) (Q. Schorpp).

(i) anecic species feed on surface litter and live in deep vertical and permanent burrow systems, (ii) endogeic species inhabit the mineral soil layer and feed on soil and associated organic matter, their burrows are non-permanent and network-like, and (iii) epigeic species inhabit the soil litter interface and utilize fresh surface litter and carbon rich upper layers of mineral soil, their burrow system is oriented horizontally [13]. Depending on their functional group, many species are able to accelerate pollution remediation or to reduce erosion and surface water run-off by the formation of casts and burrow systems [14].

Earthworm densities are affected by land-use change, intensification is negative or may completely eliminate them, whereas the conversion of degraded soils to conservation management (e.g., no-tillage) is positive [15,16]. The intensity of impacts depends on functional group membership. Anecic earthworms are at higher risk of mechanical injuries from tillage due to larger body sizes [17]. In contrast, endogeic species, especially *Aporrectodea caliginosa*, tend to benefit from ploughing when the incorporation of organic residues increases soil organic carbon (SOC) stocks [18,19]. However this might be restricted to short periods until the nutrient supply is exhausted [20]. Epigeic species seem to be unaffected by tillage in temperate agro-ecosystems [21].

Emmerling [22] documented an increase in earthworm abundance and biomass relative to maize in several bioenergy plants including the cup plant after 3 years of cultivation. However, endogeic and epigeic species were dominant and anecic earthworms showed rather low abundances. Regarding species number and abundance of endogeic and epigeic earthworms, the perennial energy plant *Miscanthus* adopted an intermediate position between permanent pasture and maize after 15 years of cultivation [9]. However, anecic earthworms did not show a significant increase compared to other crops.

Most studies about the influence of perennial cropping systems on earthworm communities represent snap-shots of an actual state or a short time span, although conducted after a long period of cultivation. By sampling a chronosequence of *S. perfoliatum* (1–9 years), the recent study takes the consecutive development over a long period of time into account. We mapped the development of earthworm communities during a common cultivation period on an artificial timeline. We hypothesized that continuous cultivation of *S. perfoliatum* affects earthworm functional groups as follows: (i) anecic earthworms increase in abundance over time, (ii) endogeics decrease, (iii) epigeics are unaffected and (iv) *S. perfoliatum* shows more diverse earthworm species-communities than maize.

## 2. Materials and methods

### 2.1. Field locations and study design

The fields under investigation were located in a large region, covering Thuringia and the south of Lower Saxony, Germany. All fields belong to commercial farms and business-related research farms. The particular locations were Erfurt (50.98132°N, 11.00591°E, 215 m a.s.l.), Niederdorla (51.13602°N, 10.40000°E, 289 m a.s.l.), Dornburg (51.00365°N, 11.65741°E, 242 m a.s.l.), Pahren (50.65273°N, 11.89898°E, 417 m a.s.l.) and Hessberg (50.42461°N, 10.78183°E, 380 m a.s.l.) in Thuringia; and Burgstemmen (52.14602°N, 9.78059°E, 88 m a.s.l.) and Gehrden (52.29513°N, 9.60931°E, 90 m a.s.l.; WGS84) in Lower Saxony. All fields have been in the vicinity of conventionally managed agricultural fields. They differed in size from 120 m<sup>2</sup> to 9000 m<sup>2</sup>. The management of cup plant (*S. perfoliatum*) fields included mechanical weed management during the first and second year of cultivation. Fertilization took place at the beginning of the vegetation period and pesticides were not applied. Harvest of the whole

above-ground biomass took place early in September, from the second year of cultivation on. Dry matter yields were in the range between 13 and 18 t ha<sup>-1</sup>. To focus on cultivation duration as an essential factor in perennial cropping systems, cup plant fields were chosen from four different years of establishment (2011, 2009, 2007 and 2004/05), n = 3 replicates per year. The number of cup plant fields of a suitable age was limited, and field selection aimed for as similar soil textures as possible (Fig. S1, Table 1). Sampling campaigns took place during autumn (2012, 2013) and spring (2013), when earthworm activity is greatest [13]. Conventional maize (*Zea mays*) fields located near the cup plant fields served as comparison group. All maize fields had similar management regimes (mineral fertilizer, ploughing, pesticide application during seed-bed preparation) and cultivation histories (i.e., grain as previous crop). Since maize is an annual crop, we switched between the years, to have maize fields again in the second year of sampling, giving a total number of six maize fields.

### 2.2. Earthworm sampling

Earthworms were sampled according to ISO guidelines [24] by a combination of hand sorting and chemical extraction using allyl isothiocyanate (AITC) as chemical expellant [25]. The procedure was replicated 4 times on each field. Samples were taken randomly in the middle of the fields at a minimum distance of 20 m from field edges. Hand sorting was conducted with soil from monoliths that covered 50 × 50 cm at a depth of 10 cm. Dilution of AITC was carried out according to Felten and Emmerling [9]: 800 mm<sup>3</sup> AITC were emulsified with 16 cm<sup>3</sup> methyl alcohol, the dilution was given to 10 × 10<sup>3</sup> cm<sup>3</sup> of water and finally poured into the sampling pits. Until identification in the lab, all earthworms were stored alive in cooled boxes (V = 10<sup>3</sup> cm<sup>3</sup>; T = 4 °C) filled with soil watered with 50 cm<sup>3</sup> tap water. Before weighing, living individuals were transferred to a water bath for 1 h at room temperature until guts were voided.

Identification was conducted with live earthworms according to keys from Graff [26], Sims and Gerard [27] and Herr and Bauchhenß [28]. Sexual development was assessed according to the scheme of Van Gestel et al. [29]. Clitellate individuals were recorded as adult, those with a full tubercle pubertatis but no clitellum as sub-adult, and individuals without either of these structures as juveniles. Adult and sub-adult individuals were identified at species level. All others were assigned either to *Lumbricus* juveniles, if their prostomium was tanylobous, or endogeic juveniles if it was epilobous. If a field was inhabited only by one single endogeic or *Lumbricus* species, juvenile individuals were allocated respectively. All species were assigned to a functional group according to Bouché [13]. *Aporrectodea longa* was considered to be anecic, although some authors mentioned a rather endo-anecic lifestyle [30].

### 2.3. Environmental parameters

Recorded parameters were soil moisture, pH, C:N mass ratio and grain size fractions and as well as climatic parameters. Water mass fraction (WMF) was measured from soil cores (n = 5, d = 4 cm, h = 10 cm) by weighing before and after drying in the oven at 105 °C for at least 24 h. Measuring soil pH was performed with a pH electrode and a pH meter using 10 g of air dried soil samples, cleaned for organic matter, in a CaCl<sub>2</sub> solution (ratio 1:1.25). C and N contents were determined in a TruMac CN Carbon/Nitrogen determinator (Leco, Mönchengladbach, Germany) with 3 g of air dried soil, ball milled to fine powder. Grain size fractions were analyzed by LUFA Nord-West (Oldenburg, Germany). Climate parameters were taken from weather stations close to the sampling locations and were provided by Agricultural Institute of the State

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