

Tagungsbeitrag zu: Workshop Kommission III „Bodenökologie und Bodenbiologie“ der Deutschen Bodenkundlichen Gesellschaft  
Titel der Tagung: Experimenting with Earthworms  
Veranstalter: Kom. III der DBG  
20.-21. März 2009 in Trier  
Berichte der DBG (nicht begutachtete online Publikation) <http://www.dbges.de>

## **Experimenting with earthworms in the field: The method and impacts of earthworms on the diversity-ecosystem functioning relationship**

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### **Abstract**

Plant diversity is one major factor driving plant productivity in temperate grasslands. Although decomposers are known to affect plant productivity, interacting effects of plant diversity and earthworms on plant productivity have been neglected in previous field studies. We investigated the effects of earthworms, plant species richness and the interaction between earthworms and plant species richness on several ecosystem functions.

**Keywords** Earthworm, decomposers; plant species richness, the Jena Experiment

### **The method**

In the framework of the Jena Experiment (ROSCHEr et al. 2004) we manipulated earthworm densities in a plant diversity gradient. Earthworm subplots were established on the 1 (16 plots), 4 (16 plots) and 16 plant species richness levels (14 plots) in September 2003. In each plot two randomly selected subplots of 1 × 1 m were

used to establish the following treatments: earthworm addition and earthworm reduction. Subplots were enclosed with PVC shields aboveground (20 cm) and belowground (15 cm) to prevent the escape or colonization of earthworms. Earthworm addition subplots (+ew) received 25 adult individuals of *Lumbricus terrestris* L. (average fresh weight with gut content  $4.10 \pm 0.61$  g) per year (15 individuals in spring and 10 in autumn). Further, two earthworm extraction campaigns were performed per year (spring and autumn) on the adjacent earthworm reduction subplots (-ew) by electro-shocking. A combination of four octet devices (DEKA 4000, Deka Gerätebau, Marsberg, Germany) was used (THIELEMANN 1986).

In each subplot earthworm extraction was performed for 35 minutes, increasing the voltage from 250 V (10 min) to 300 V (5 min), 400 V (5 min), 500 V (5 min), and 600 V (10 min).

The success of earthworm density manipulations was measured via the soil surface activity of *L. terrestris* which is known to bury plant seeds irrespective of seed size and shape (MILCU et al. 2006; EISENHAUER et al. 2008a; EISENHAUER & SCHEU 2008). In May 2006, we performed a seed dummy experiment to determine earthworm soil surface activity (EISENHAUER et al. 2008b). While earthworm soil surface activity in earthworm addition subplots did not differ from that in control subplots, it was decreased considerably in earthworm reduction subplots (-38%; EISENHAUER et al. 2008b).

### **Earthworms affect the diversity-ecosystem functioning relationship**

We measured the following ecosystem functions on earthworm subplots: primary productivity, invasibility (EISENHAUER et al. 2008b), stability (EISENHAUER et al. 2008b), litter decomposition (MILCU et al. 2008), plant seed burial (EISENHAUER et al. 2008b) and plant regrowth. Most of these variables were affected by plant diversity underlining common knowledge on diversity impacts;

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however, earthworms affected all ecosystem functions (Table 1).

**Table 1:** Ecosystem functions as affected by plant species richness (SR) and earthworms (EW).

Variable	SR	EW	SR × EW
Productivity	***↑	*↑	ns
Invasibility	**↓	*↑	ns
Stability	**↑	*↓	*
Decomposition	ns	**↓	ns
Seed burial	*↓	***↑	ns
Plant regrowth	ns	**↑	*

\*\*\*,  $P < 0.001$ ; \*\*,  $P < 0.01$ ; \*,  $P < 0.05$ ; ns, not significant; ↑, increase with higher diversity or higher earthworm densities; ↓, decrease with higher diversity or higher earthworm densities

Earthworms enhanced plant primary productivity, plant invasibility, litter decomposition, plant seed burial and plant regrowth but decreased the stability of the plant community. Interestingly, in some cases earthworms modified the diversity-ecosystem functioning relationship (stability and plant regrowth), i.e. earthworm effects depended on the diversity of the plant community (EISENHAUER et al. 2008b). These results show that soil animals should be considered in plant diversity experiments and in the diversity-ecosystem functioning debate.

## Literature

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