

## Soil seed bank studies I-III

**Gerhard Karrer, Felicia Lener and Nina Waldhäuser**

Institute of Botany, Department of Integrative Biology and Biodiversity Research,  
University of Natural Resources and Life Sciences, Gregor Mendel Str. 33, 1180 Wien, Austria;  
e-mail: gerhard.karrer@boku.ac.at



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The soil seed bank of *Ambrosia artemisiifolia* (common ragweed) was analysed by Fumanal *et al.* (2007) for French populations and by Leitsch-Vitalos in Karrer (2009) for Austrian populations. No other studies focussed specifically on common ragweed soil seed banks. Further studies were performed during this project.

### I. Seed bank studies in different habitat types

#### Introduction

Studies on the soil seed bank of *Ambrosia artemisiifolia* can help to get better insights in the population dynamics of invading populations. *Ambrosia artemisiifolia* soil seed banks show considerable spatial variation (Fumanal *et al.* 2007, Vitalos *et al.* 2009). Any control options should be evaluated in view of the age of populations and duration of accumulation of seeds into the soil seed bank. Fumanal and Vitalos studied agricultural fields, pastures, roadsides and few abandoned fields. The soil seed bank of common ragweed in near natural habitat types (meadows on military training areas and ruderal sites along rivers) were studied in 2011 at the BOKU. The aim was twofold: a) to characterize the soil seed bank of subpopulations of different age and different environmental influence near to the river Danube, and b) to describe the soil seed bank of experimental plots in an intensively invaded military training area that were managed like meadows, i.e. cut at least once a year.

#### Methods

In case of the embankment plots in the northern part of Vienna, we selected two areas where soil sediments produced by Danube floods have been deposited nearby the river (Fig. 1). Furthermore we sampled a roadside on the way from the flooded area to the soil deposit (Fig. 2) and the former flooded area just few meters from the river (Fig. 3 and 4). The previous year there had been a heavy flooding by the Danube that left sandy sediments in the alluvial zone with ca. 30 cm depth. The sediment was taken by machines and transported to the deposit. Before the flooding a common ragweed population was already established for about 5 years on this place, whereas the roadside population was very young (2 years) and the 2 deposit subpopulations were 2 and 4 years old.

As seeds deposited on the soil surface can be embedded into the soil from undisturbed surfaces only slowly into deeper soil horizons we expected that the populations of different age differed with respect to the distribution of common ragweed seeds by soil depth. Unfortunately, the soil at plot beside the Danube (nr. 4) turned out to be very shallow. So from this plot we only could take one soil depth layer per core. From the very young roadside population (plot 3) we took 20 soil cores from 0 to 10 cm soil depth. From the deposit plot 1 we took only 5 soil cores because the area where common ragweed was known to grow was small, from plot 2 we took 16 cores; in both cases we sampled 0-10 cm and 10-20 cm separately. Generally the sampling strategy has to follow the arrangement of the plants in the field. Therefore many populations along rivers or roadsides are linear (narrow but long) whereas anthropogenous habitats like soil deposits tend to be more rounded and can be sampled rectangular or circular.

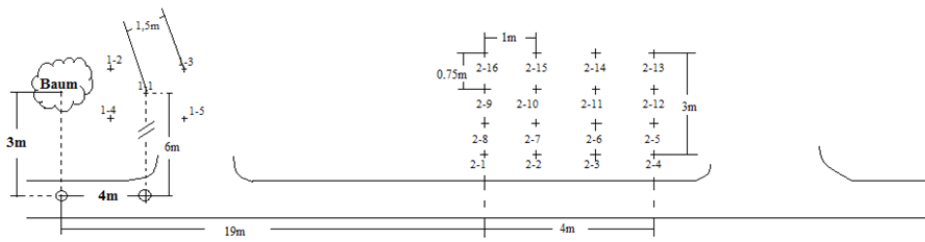


Fig. 1: Sampling design of plot 1 (left, circular sampling) and 2 (right, rectangular sampling) on the area of the artificial river sediment deposit

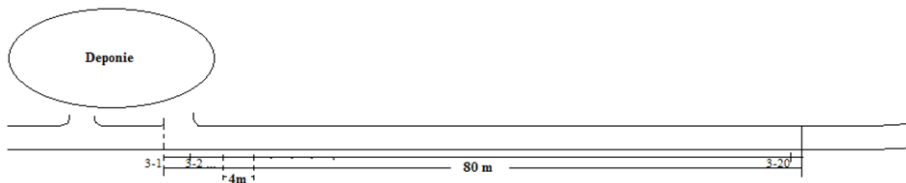


Fig. 2: Sampling design (linear transect) of plot 3 along the road between the river and the deposit

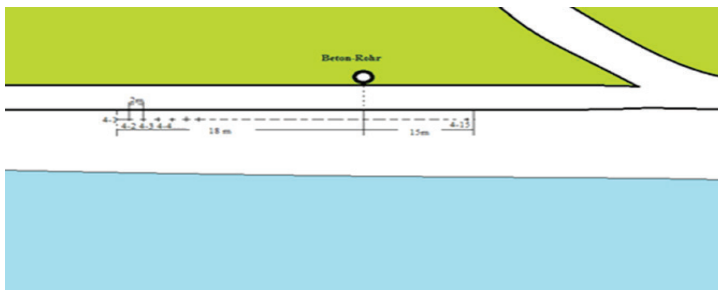


Fig. 3: Sampling design (linear transect) of plot 4 near the river (in blue)



Fig. 4: Overview (left) and detail (right) of plot 4 along the river



Fig. 5: Overview (left) and details (right) from sampling the military training area near Bruckneudorf

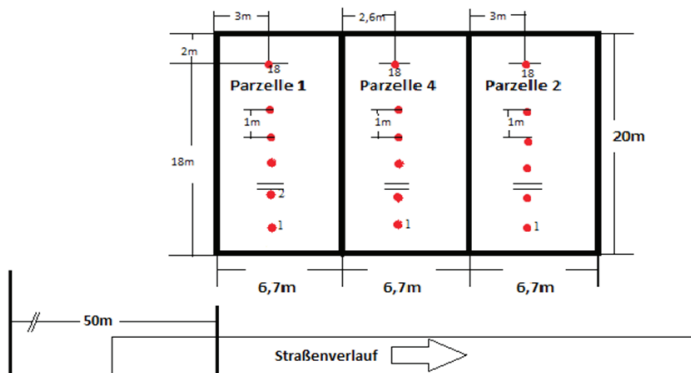


Fig. 6: Sampling design for one plot (out of 3 replicates) on the military training area near Bruckneudorf

In case of the military training area (near Bruckneudorf, Burgenland, Austria) sampling served to describe the soil seed bank of common ragweed on the experimental plots where different treatments for seed bank depletion will be tested. Therefore the design is more extensive. 3 different treatments were replicated 3 times along a meadow (Fig 5 and 6). On each treatment-subplot we sampled (linearly arranged) 20 soil cores with 6.6 cm diameter divided into layer 0-10 and 10-20 cm. The soil cores were stored for further analyses in darkness at 4°C. Common ragweed seeds were then sorted out by automated floating sieves and put into petri dishes in climate chambers (8 h light, 30°C, 16 h dark, 15°C). During the first germination turn (4 weeks) many seeds germinated and discarded. Afterwards, the remaining seeds were kept dry and cool again for 8 weeks to start a second germination turn aiming at breaking secondary dormancy of remaining seeds. Seeds that still remained dormant after the second germination round were tested with Tetrazolium dye for vitality.

## Results

The two soil deposit samples had common ragweed seeds only in the upper soil layer (0-10 cm) with 467.9 and 146.22 seeds per m<sup>2</sup> (plot 1 and plot 2, resp.). The roadside plot 3 showed 59.76 seeds per m<sup>2</sup> and the river bank plot 4 226.66 seeds per m<sup>2</sup>. On the military training area (meadows) we found on average 188 seeds per m<sup>2</sup>, in 0-10 cm depth a mean of 129 and in 10-20 cm a mean of 59 seeds per m<sup>2</sup>.

The germination rates of seeds from the Danube plots ranged from 100% to 88% for deposit plot 1 and 2, resp.; the seeds from the roadside plot 3 germinated by 100% and those of the river bank plot 4 by 88.23%. The seeds from the military training area gave germination rates of 55 (0-10 cm) and 68% (10-20 cm) on average. The second germination round as well as the viability test (TCC) showed that all seeds left were not dormant but dead.

The low age of the common ragweed populations from plot 1 to 3 can be deduced from the fact that there no seeds had penetrated into deeper soil layers. However, the considerable high number of seeds in the deeper soil layer of the military training area indicates that the population was established already for many years at this site.

The site Bruckneudorf (military training area) gave mean seed numbers of 626.33 seeds/m<sup>2</sup> (SD = 864) for plot 1, 583.69 seeds/m<sup>2</sup> (SD = 800.37) for plot 2 and 12.9 seeds/m<sup>2</sup> for plot 3. These figures correspond to the seed densities given by Fumanal *et al.* (2007a, b) with 536 +/- 194 to 4477 +/- 717 seeds/m<sup>2</sup>. Fumanal also found that the upper soil layer (0-5 cm) of arable fields gave lower common ragweed seed numbers than in lower soil depths (5-20 cm). On fallows and unmanaged abandoned fields the seed numbers in deeper soil was lower (cf. Fumanal *et al.* 2007b, p. 101)

## References

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## II. Seed bank studies: Quantification of soil seed bank losses of common ragweed between autumn and spring sampling

### Introduction

The soil seed bank of *Ambrosia artemisiifolia* is commonly sampled between seed dissemination and natural germination in spring. To our knowledge nobody ever tested for differences of the natural seed banks of common ragweed between autumn and early spring. That is why we started an experiment to test the seed bank of common ragweed from 7 different experimental plots along roadsides.

### Methods

We used the plots from the mowing experiment to test for the differences in seed bank composition between autumn 2011 and spring 2012 (Schöberl & Lebernegg, 2013). Thus we sampled 19 soil cores each from every treatment plot of all sites of the field cutting experiment. The soil cores were taken from the upper soil layer assuming that the intrusion of seeds to the soil at undisturbed habitats takes rather long time and differences would be assured by using the upper soil layer between 0 and 7 cm.

The soil cores from the autumn sample were stratified for 6 weeks in darkness at 4°C, the spring samples were directly analysed.

Both sets of soil cores were washed out by use of a wet sieving machine (Retsch). All obviously viable seeds were put into petri dishes and treated for 4 weeks in climate chambers at 8 h light at 30°C and 16 h darkness at 15°C. All seeds that germinated were taken out from the dishes. The remaining seeds that obviously stayed dormant were left for drying and afterwards stored again in darkness at 4°C for further stratification. After four weeks, a second germination test under the same conditions as before was performed in the climate chambers. Again all seeds that germinated were counted and deleted.

Still remaining seeds were subsequently tested for dormancy/viability via the TTC-test.

## Results

We compared the total number of viable seeds (germinated + TTC-fully stained) per site between the two sampling periods and found on average an expected decrease of seeds from autumn to spring, except for one site (see Fig. 1). The unpredicted difference at the Halbenrain site might be caused by some local effect of seed introduction by very late mowers or snowplough. All other sites showed generally losses of viable seeds by 5 to 30%.

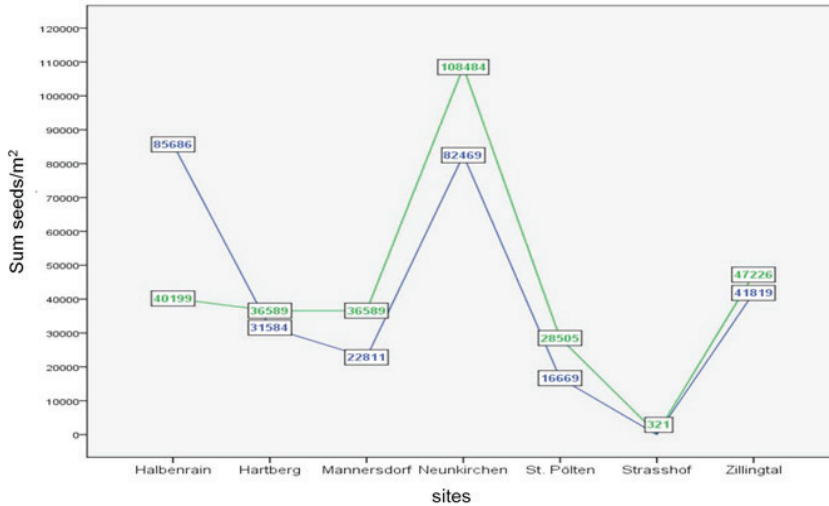


Fig. 7: Total number of viable seeds/m<sup>2</sup> in autumn 2011 (green) and spring 2012 (blue) at different experimental plots

In consequence comparisons of soil seed bank data as an efficacy measure should be designed in that way that the same season must be sampled. It is well known that the soil seed bank decreases in summer crop weeds from late winter/early spring to summer significantly (Fumanal *et al.* 2007) but it was not known until now that the difference between autumn and spring sampling can also be serious.

## References

- Fumanal, B., Chauvel, B., Bretagnolle, F. (2007): Estimation of pollen and seed production of common ragweed in France. *Ann Agric Environ Med* 14: 233-236.
- Schöberl, S., Lebernegg, M. 2013: Die Bodensamenbank eines invasiven Beikrauts als Bewertungsmöglichkeit für den Erfolg von Bekämpfungsmaßnahmen (*Ambrosia artemisiifolia*). Bachelor thesis. University of Natural Resources and Life Sciences Vienna. 77 pp.

### III. Seed bank studies: Efficacy of various mowing regimes used for ragweed control along roadsides measured by soil seed bank

#### Introduction

Sampling of soil seed bank is an important efficacy measure for the different control options of common ragweed. We will use this measure to test the efficacy of different cutting regimes on those experimental plots along road shoulders of Eastern Austria that were established for the HALT Ambrosia project.

#### Methods

In total, 7 experimental sites in Lower Austria, Styria and Burgenland have been sampled in spring 2012 for evaluating the effects of cutting experiments. 20 soil cores (depth 7cm) per treatment and locality were taken by end of March and analysed for common ragweed seed content using a wet sieving machine (Retsch). We counted all obviously intact seeds and put them into petri dishes. In order to detect the proportion of viable seeds, first germination was induced by wetting the dishes and putting them into climate chambers at following conditions: daylight for 8 hours at 30°C and darkness for 16 hours at 15°C. After 4 weeks the first germination trial was stopped, the dishes left for drying out and stored for 4 weeks at +4°C in darkness. A second germination phase was started afterwards (mid July 2012) at the same conditions like in the first session. All seeds that did not germinate within the next 4 weeks were tested afterwards by the standard TTC-test described in B1.2 for any seeds still alive.

The results were compared to soil seed bank data of the sampling of the same experimental sites in spring 2009, before the start of the experiment (see Karrer et al. 2011). That way, it is possible to conclude on the effect of the tested mowing regimes on the soil seed bank after 3 years of application.

Statistical treatment: GLLM (in Statistica 10.0)

#### Results

In 2012, soil seed bank at different sites varied from 0 to 1061 seeds per m<sup>2</sup>, for a depth of 0 to 7cm. The germination rates were generally very high (91% in average). No seeds germinated in the second germination test and no living seeds were detected by the subsequent TTC test.

After 3 years of applying different mowing regimes, the ragweed soil seed bank of treatment 1 (control, unmown) increased almost threefold, the one of treatment 2 (first cut in late June, second cut in 2<sup>nd</sup> week of September) did not change significantly, whereas it decreased by ca. 80% under treatment 3 (first cut in 3<sup>rd</sup> week of August, second cut in 2<sup>nd</sup> week of September), ca. 60% under treatment 4 (first cut in late June, second cut in 2<sup>nd</sup> week of September) and ca. 45% under treatment 5 (cut 3 times: first cut in late June, second cut in the 3<sup>rd</sup> week of August, third cut in in 2<sup>nd</sup> week of September).

#### Conclusion on all soil seed bank studies I-III

Because most management options act superficially, the most problematic aspect of common ragweed control is the elimination of the persistent seeds from the soil. The results of this long term experiment show that the soil seed bank can strongly be diminished by a carefully thought and adapted mowing management. The mowing management consisting of a first cut in August just about the start of appearance of female flowers and a second cut in early September, the results suggest that this management can be evaluated as very sustainable and environmentally friendly control option, as it progressively empties the soil seed bank. This way the common ragweed populations decline and can be managed easier. The most effective measure of hand-pulling of the remaining plants might become feasible after seed bank depletion.

## Final comment

Based on the comparison of the soil seed bank of all experimental plots between spring 2012 and autumn 2011 (sampling done in October 2011) we found a mean loss of seeds during winter by up to 20% (Schöberl & Lebernegg 2013). The number of counted seeds showed considerable high variation. That is why we decided to do a final scientific test of the trial effects in early spring 2014 independent from the HALT-Ambrosia project. For the analysis of the experiments within the HALT-Ambrosia project the data from spring 2012 are valuable and fit better to the experimental design (comparison of spring data 2009 with spring data 2014 is better than comparison of spring samples with autumn samples using correction functions to estimate seed numbers that would be counted in spring 2014).

## References

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