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Sustainable management of *Ambrosia artemisiifolia* in Europe

Final Conference

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Abstract Booklet

COST-SMARTER Conference - Tuesday 13 september 2016

Time	Speaker/Chair	WG/TF	Title
08:45	**Heinz Müller-Schärer**	Chair	Welcome
08:50	**Matt Smith**	WG4	**Temporal and spatial variations in airborne Ambrosia pollen as an indicator of changes in the distribution and abundance of ragweed plants on the ground**
09:10	**Ivana Milakovic**	WG2	**What is the most efficient efficacy measure of non-chemical control of common ragweed?**
09:30	**Heinz Müller-Schärer**	TF Ophraella	**Estimating the risk of non-target effects of *Ophraella communa* : from ecological to evolutionary perspectives**
09:50	**Suzanne Lommen**	TF Pop Dyn	**Variation in ragweed demography and modelling management impact in Europe**
10:10	**Sandro Steinbach**	WG4	**Evaluating the costs and benefits of common ragweed management – An application to the Rhône-Alpes region in France**
10:30	Coffee break		
11:00	**Michel Thibaudon**	WG4	**The French evolution of exposition and health impact since 10 years**
11:20	**Rodolfo Gentili**	WG2	**Enforcing competitive vegetation against ragweed**
11:40	**Florencia Yannelli**	WG3	**Biotic resistance to invasive alien plant: Using a multi-trait approach to design resistant communities**
12:00	**Yan Sun**	WG1	**Adaptive genetic divergence and increased competitive ability in invasive *Ambrosia artemisiifolia***
12:20	**Beate Alberternst**	WG2	**10 years action programme against ragweed in Bavaria – results of monitoring and scientific companionship**
12:40	**Alfons Oude Lansink**	WG4	**A multi-level and multi-actor approach to biosecurity management and risk governance: a conceptual framework to support policy development for Ambrosia weed control**
13:00	**Maira Bonini**	IRS	The International Ragweed Society
13:10	Lunch		
14:10	**Maira Bonini**	IRS	IRS General Assembly
14:40	**Lukasz Grewling**	WG4	**Non-native ragweed pollen in Western Poland – origins, behavior and impacts**
14:55	**Hana Skalova**	WG2	**Looking for competitors to ragweed: results from garden and growth chamber experiments**
15:10	**Friederike Trognitz**	WG1	**Plant associated bacteria to reduce the growth of *Ambrosia artemisiifolia***
15:25	**William Ortmans**	WG3	**Is *Ambrosia artemisiifolia* L. able to expand its invaded range northward in Western Europe?**
15:40	Coffee break		
16:10	**Per Kudsk**	COST SMARTER	Future ragweed management and success criteria for a successful management
16:40	**Heinz Müller-Schärer**	COST SMARTER	COST SMARTER - Achievements and the future
17:00	**Helen E. Roy**	COST ALIEN	COST ALIEN CHALLENGE - Achievements and the future
17:20	**Joint poster session**	SMARTER/ALIEN	
18:00	Conclusion		
20:00	Final SMARTER Dinner		

Abstracts' List

10 years action programme against ragweed in Bavaria – results of monitoring and scientific companionship

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In 2007 the Bavarian State Ministry started a comprehensive anti-ragweed programme in order to combat the species and stop further spread. In addition a scientific research and monitoring programm was installed to document the success of the programme, observe the development of the ragweed populations and to find new occurrences.
In Bavaria (and Germany) the combat of common ragweed is not compulsory and it depends on the willingness of landowners to start with control measures against the plants. In many cases landowners cooperate, especially when the infestation and the effort to remove the plants are small. But often the topic is not taken seriously and no or inconsistent measures occur. Up to 2015 in 78% of the stands which were combated the number of individuals decreased, but only 75 (21%) of the known 363 ragweed stands were removed completely. In some cases even after 10 years of ragweed control plants are still present.
Untill 2015 363 big ragweed stands (≥100 plants) became known in Bavaria. Although information on the plant is provided, only a few new ragweed stands are currently announced by citizens to the authorities. The number of announcements mainly depends on reportings by lokal press and decreases with declining coverages. A special search programme for new ragweed stands indicates that about twice as much occurences are still undiscovered. Thus special search programmes done by experts are necessary to find new ragweed stands.
Intensified measures are necessary to stop the invasion of common ragweed. Especially legal regulations including obligations to announce new stands, obligation to control the plant and implementation of penalties in case of lacking cooperation are necessary. A political signal indicating that ragweed control is of high priority is desirable.

Aerobiological monitoring and mapping of *Ambrosia* plants in the Province of Parma (Northern Italy, Southern Po Valley), a useful tool for targeted preventive measures

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Ambrosia (A.) includes some species of annual anemophilus weeds which produce highly allergenic pollen affecting public health in European countries. In Italy, the most infested region is Lombardy where, in some areas, it is the main cause of hay fever. In Parma, until 2007, these plants seemed to be very rare, despite an increase of Seasonal Pollen Index (SPI) and of asthma among sensitized patients to ragweed. We calculated the ragweed pollen season from 1996 to 2015 and verified the ragweed presence in the territory.
We analyzed the following parameters of pollination: start, end, duration, peak concentration date, peak values and SPI according to Jäger et al. During naturalistic activities we have identified sites with these plants.
As median, start, end, duration was, respectively, August 6, September 18, 46 days. The peak day was August 28th (36 p/m^3), SPI 259. We observed a significant increase of SPI and of peak value until 2011 (peak value 128 p/m^3, SPI 793 during 2010) and a reduction of SPI (-52.6%) and of peak value (-58.6%) 2012-2015 vs 2009-2011. In the territory we have identified numerous sites with ragweed plants: *A. artemisiifolia* (41), *A. coronopifolia* (37) and *A. trifida* (5), even within the city at the confluence of Parma river with the Baganza river. For the first time in 2014, in Vignale, we observed beetle *Ophraella communa* that eats ragweed leaves.
In the future, in our region, we could observe an increase of ragweed allergy related to plants spread over the territory. In our area the situation is very similar to what occurred in Lombardy at the end of the last century. No law and no enforcement initiative by institution and local authorities to prevent and to reduce *A*. plants spread could determine consequences on public health and on sanitary costs. At present, it remains to evaluate the role of *Ophraella* in reduction of pollen concentration observed in the last few years.

Modeling of possible distribution of common ragweed (*Ambrosia artemisiifolia* L.) under climate change in Armenia

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Understanding the spread of an alien invasive species opens the chance to design and implement adequate mitigation measures in early phases of spread. In case of ragweed it is highly indicated to prevent or at least slow down the spread because of the disastrous allergic reaction caused by its pollen.

In 2013-2015 was carried out the present distribution of *Ambrosia artemisiifolia* on the territory of Armenia for monitoring of population dynamics and for assessment of main threats. Only during these 2 years were found 12 new populations, based on which we can assume that common ragweed can occupy new habitats and extend its spread. Our hypothesis was that for next 100 years orographical condtions and urban areas will not undergo significant changes, but climate change will be determining factor for its further spread. For forecasting future distribution in territory of Armenia we've used Species Distribution models, which are internationally widely used tools for such issues. Here we've used 4 models: RF, MARS, GBM, MAXENT. As input data were used: anthropogenic roads and railways (with buffers of 3 km), urban areas (with buffers of 10 km), orographical, edaphic and climatic variables. For modeling of distribution under climate change was used the worst RCP 8.5 climate change scenario for different climatic models CSM4, GISS-E2-R, HadGEM2-AO and GFDL-CM3. All bioclimatic models show trends of further spread of common ragweed in Armenia. According to results the best models are GISS-E2-R and HadGEM2-AO, the worst - GFDL-CM3. According to GISS-E2-R ragweed in Armenia has big potential of spread and till 2080 will occupy all Central and North-Eastern Armenia and some territories in south of the country. According to GFDL-CM3 potential of further distribution is low and current occupied territories will not be expanded. But *in fact, Ambrosia artemisiifolia will continue its spread and will become even more dangerous for both agricultural and natural ecosystems and for biodiversity of Armenia.*

Case study prevention and measuring management success in Italy

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The North-Western Milan area has been colonized by *Ambrosia artemisiifolia* since the 1940s. Clinical manifestations of ragweed allergy were observed in allergy clinics of this area only starting from the middle of the 1980s. Currently, the zone is the most infested by ragweed in Italy, together the nearby Southern Varese area (both in Lombardy) and one of those most infested in Europe. Seeing the situation, in 1999, the Lombardy Region promulgated the first measures to prevent the spread of the plant and the production of its pollen. These measures also included the functions of the Local Health Authorities in the management of the problem. Therefore, we have developed and improved our management strategy, that is a set of primary prevention actions: epidemiological studies; assessment of direct health cost; studies on methods to limit the ragweed spreading (also in collaboration with the COST-SMARTER team); information and education of the Public Authority and the population; control of the territory (aerobiological monitoring, surveillance and monitoring of the infested area and town planning). Several epidemiological studies demonstrated the increase in prevalence of ragweed allergy, confirming the aggressive allergic behavior of ragweed pollen (about 40% of the ragweed allergic patients suffered from asthma). Thus, these studies are tools to quantify the impact on health due to ragweed, to point out the necessity to adopt a management strategy, to monitor the efficiency of this strategy and to improve it. Starting from 2012, we observed a decline of the infested areas. Finally, we detected an impressive decrease of ragweed airborne pollen levels mainly due to the *Ophraella communa* spreading starting from 2013. In conclusion, some tools we used (epidemiological studies, pollen levels, control of the infested areas) have given us clear indications of the efficiency of our prevention strategy and when it was necessary to improve it.

Long distance transport of *Ambrosia* sp. allergens

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Ambrosia sp. (ragweed) is one of the most important invasive plant in the world and pollen grains is important for the allergic individuals. *Ambrosia* spp. pollen grains can be dispersed by air masses far from their source. Air temperature, humidity and solar radiation on pollen grains in the atmosphere could impact on the ability of long distance transported (LDT) pollen to maintain allergenic potency. The aims of the study to detect of the major allergen of *Ambrosia artemisifolia* (Amb a 1) in the atmosphere of uninfected areas for a potential trigger of allergic reaction and to compare daily *Ambrosia* spp. pollen grains/Amb a 1 amount in per m^3.

Samples collected by Chemvol sampler for allergen detection and by hirst trap for calculation of pollen concentration. Chemvol collects particles at 800 l/min and it contains 2 impaction stages PM>10 micron and 10 > PM>2.5 micron. Calculation of Amb a 1 in the air samples was performed by antibody-based two-site immune enzymatic assay (ELISA). Evaluation of these methods for collecting and quantifying airborne pollen allergens has been performed during the HIALINE project (www.hialine.com). Samples were collected during the year 2010 and 2014.

Samples were collected for 44 days during the investigated period in 2010. Amb a 1 was recorded on 37 days and pollen grains on 35 days. Daily average Amb a 1 levels varied from 0.13 to 101.8 pg m^3 and were significantly correlated with daily average *Ambrosia* sp. pollen concentrations. The mean seasonal *Ambrosia* pollen potency was 3.07 Amb a 1/pollen grain.

Samples were collected for 63 days during the investigated period in 2014. Amb a 1 was recorded on 54 days and pollen grains on 43 days. Daily average Amb a 1 levels varied from 0.29 to 263.3 pg m^3 and were significantly correlated with daily average *Ambrosia* sp. pollen concentrations (adjusted R^2 value is 0.78; spearman correlation coefficient = 0.81; p = 0.006). The mean seasonal *Ambrosia* pollen potency was 3.44 Amb a 1/pollen grain.

The findings suggest that that Amb a 1 have a potential trigger of allergic reaction even in areas where the Ambrosia is not widely distributed.

This study was supported by TUBITAK project no109S032 and 114Z698 and I would like to thank Buse Vatansever for technical assistance.

Potential sources of *Ambrosia* pollen in Bursa, Turkey

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Ambrosia pollen is an important allergen in North America originally and, as last decade reported, in most European countries. It was reported that the affected area was the northeast and Thrace region of Turkey. Ragweed has not been yet recorded in other parts of the country. The study investigates *Ambrosia* pollen episodes during the peak days of 11 yearly periods, between the years 2003-2015. The HYSPLIT back trajectory model was used to identify a potential source of atmospheric *Ambrosia* pollen at Bursa, Turkey. Yearly pollen level was changed between 99-1620 pollen/m^3. Back-trajectory analyses showed that the air masses arriving at the sampling station predominantly came from the North or Northeast, and were in Russia and Ukraine where the place around the Azov Sea. Results indicate that these countries are potential source areas for *Ambrosia* pollen infection for the Bursa. Note that atmospheric concentrations of *Ambrosia* pollen exceeded the clinical threshold during some years in investigated region. Taking into consideration the high allergenicity of *Ambrosia* pollen, the present findings suggest that the number of sensitized individuals might increase in the near future.

This study was founded by a grant from TUBITAK (project number 109S032) and supported by Scientific Research Unit of Uludag University (project number UAP(F)-2011/76).

Potential sources of *Ambrosia* pollen in Istanbul and Kocaeli, Turkey

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Ambrosia pollen is an important allergen in North America originally and, as last decade reported, in most European countries. It was reported that the affected area was the northeast and Thrace region of Turkey, and ragweed has not been yet recorded in other parts of the country. The study investigates *Ambrosia* pollen episodes between the years 2010-2012. The HYSPLIT back trajectory model was used to identify a potential source of atmospheric *Ambrosia* pollen during the 2 or 3 yearly periods. Back-trajectories were computed for three sampling stations which placed European and Asian side of Istanbul and in the center of Kocaeli, Turkey. Yearly pollen level was changed between 529-4875 pollen/m^3. Back-trajectory analyses showed that the air masses arriving at the sampling station predominantly came from the Northeast and North, and were in Russia and Ukraine where the place around the Azov Sea. Also another region for potential pollen sources was identified as Black Sea region of Turkey. These findings indicate that these regions are potential source areas for *Ambrosia* pollen infection for the cities of Istanbul and Kocaeli.

This study was founded by a grant from TUBITAK (project number 109S032) and supported by Scientific Research Unit of Uludag University (project number UAP(F)-2011/76).

Enforcing competitive vegetation against ragweed

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Following ecosystems degradation due to human activity, many non-native species colonize open disturbed habitats and fill vacant niches, preventing native flora establishment. *Ambrosia artemisiifolia* is known to successfully colonize open disturbed habitats during the early stages of succession occupying vacant niches. With the increase of vegetation cover, the species is replaced by perennial plants that may inhibit its growth through competition mechanisms. Therefore, we envisaged that creating a dense vegetation cover of native species negatively influences *A. artemisiifolia* growth and spreading.

During a three-years field experiment, we assessed the effect of vegetation cover on *A. artemisiifolia* abundance, growth and fitness, measuring the species traits. In different open habitats invaded by *A. artemisiifolia* (Lombardy, Italy), we tested the following treatments, within 3x3 m plots: a) spontaneous succession (C); b) seeding of hayseed (HS) mixture from natural grasslands; c) seeding of commercial seed mixture (CS).

After the first growing season, the results highlighted that, compared with spontaneous succession, creating a dense vegetation cover both from commercial seed and hayseed, significantly reduced *A. artemisiifolia* abundance and inhibited its vegetative growth and reproductive fitness. After the second growing season, common ragweed disappeared in the commercial seed stands and after the third growing season it also disappeared in the hayseed and spontaneous succession stands. This study indicated that restoring a vegetation cover from spontaneous succession or by seeding grassland mixtures fills vacant niches of open disturbed habitats and then determines a competitive exclusion of *A. artemisiifolia*. The restoration of a vegetation cover could be used in combination with other methods (mowing and biological control) to suppress common ragweed from invaded habitats.

Ambrosia in Greece: First citing of *A. psilostachya*

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Ambrosia psilostachya originates from North America. Nowadays, it has expanded in Asia, Africa, Australia and many European countries. The species is reported to be highly competitive, allelopathic, invasive, capable of forming large populations within a short time and with very allergenic pollen. Recently, we found a small population *of Ambrosia psilostachya* DC. var*. coronopifolia* (Torr. et Gray) Farwell ex Fernald (= *Ambrosia coronopifolia* Torr. & A. Gray) in the periurban forest of Thessaloniki, south of the village Exochi. This is the first citing of this species in Greece. It is noteworthy that it has not been reported so far from the neighbouring countries (Albania, FYROM, Bulgaria, Turkey). This finding could explain, at least to a certain degree, the systematic presence of ragweed pollen in the air of the city. However, when *Ambrosia* pollen started to be detected in the air of Thessaloniki this year (2nd week of August), a visit to the population showed that it was not yet in bloom. This makes the existence of other *Ambrosia* populations in and around the city quite possible. We will increase our efforts in surveying the area and make sure that measures will be taken against these obnoxious xenophytes.

Non-native ragweed pollen in Western Poland – origins, behavior and impacts

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The distribution of *Ambrosia artemisiifolia* L. (common ragweed) in Poland is mainly limited to South-Eastern regions. Western Poland is considered to be a ragweed-free area, i.e. *Ambrosia* has only occurred there ephemerally, but airborne ragweed pollen grains are observed over this region during August and September each year. It is believed that the majority of the pollen grains recorded are transported by air masses from distant sources (over 500km away). Ragweed pollen seasons in Western Poland are intermittent, and exposure of ragweed pollen depends on meteorological conditions facilitating long-distance transport (LDT). It is not, however, uncommon to record airborne ragweed pollen in concentrations that can potentially induce allergy symptoms in sensitized individuals. The role of non-native *Ambrosia* pollen in inducing new sensitization is still under debate, as the action of unfavourable weather conditions, solar radiation, and air pollution might affect the allergenic properties of transported pollen grains. This study will present the current state of knowledge regarding the atmospheric transport of ragweed pollen to Western Poland, with special attention to the mechanism and routes of transport. This is the first study that assesses the allergenic properties of ragweed pollen grains after LDT by air masses and thus it evaluates the impact of this well-known phenomena on human population.

Estimation of seed longevity in artificial soil seed banks of common ragweed

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Two experiments, started more than 100 years ago in the US, have shown that seeds of common ragweed (*Ambrosia artemisiifolia* L.) can survive up to 39 years in soil seed banks. Since no data are available for alien European ragweed populations, the HALT-Ambrosia team set up a long-term experiment to test ragweed seed longevity from different origins buried at 2 depths at 5 locations in Europe. Seed lots from Hungary (Kaposvar 2010) were buried in each country; additionally, a second seed lot from other origins was buried at each site: Germany used seeds from Domsdorf (Germany 2010); Austria and Hungary used seeds from Hagenbrunn (Austria 2011); Denmark and Slovenia used seeds from Unterpurkla (Austria 2010). In the winter 2011/12 seeds were buried at 5-8 cm and 25 cm depth respectively in fields or lawns in the surrounding of Vienna (Austria), Braunschweig (Germany), Kaposvar (Hungary), Flakkebjerg (Denmark) and Ljubljana (Slovenia). Fifty seeds were put in fine mesh polyethylene nets and buried at the two soil depths. Every year in early spring until 2022 a randomly chosen subset of five polyethylene nets per depth and origin were/will be dug out. Intact seeds are counted and tested for viability by germination test (wetted in petri dishes, 12h light at 25°C, 12h darkness at 15°C) for 4 weeks; the remaining not germinated seeds are subsequently tested for viability with the TTC-test. Germinability was expressed as percentage of initial number of seeds.

In the first year (spring 2013), the excavated seeds from Kaposvar still showed relatively high viability rates of 40-97% (mean of 88% at soil depth 5-8 cm) and 48-98% (mean of 89 % at soil depth of 25 cm). The seed lots from Hagenbrunn lost viability (35-82% at 5-8 cm depth and 38-89% at 25 cm), similarly as the seeds from Unterpurkla (23-42% at 5-8 cm depth and 19-43% at 25 cm). Mean survival rates of the Domsdorf seeds were 91% at 5-8 cm and 98% at 25 cm depth. The laboratories tried to follow the manual as strictly as possible; nevertheless variation in the estimation of viability between the labs was relatively high.

Data from subsequent years (2014, 2015) showed less variation between laboratories. The trend observed in the first year that Kaposvar seeds survived at higher rates than the other seed lots continued. The differences in ragweed seed survival with respect to burial depth tends to increase with duration of burial, but without significant differences up to now.

What is the most efficient efficacy measure of non-chemical control of common ragweed?

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The North American Ambrosia artemisiifolia L. is one of the worst IAS to Europe. Agricultural yield losses, costs for control as well as health costs exceed financial losses caused by other vascular plant pests. In many situations, herbicides cannot be applied, thus non-chemical control measures have to be established. Cutting and outcompeting ragweed are favored tools in sensible environments. I.e. on roadsides, ragweed was favored to spread due to inappropriate cutting regimes and prevention of competitive exclusion by native plants.
As series of experiments were set up to find optimal cutting regimes and optimal competitive seed mixtures in infested habitat types that are under non-chemical management. Cutting experiments under controlled conditions indicated the high regenerative power after cut – measured as number of regrown shoots/biomass or inflorences (Bohren et al. 2004, Milakovic et al. 2014a). Long-term field experiments testing various cutting regimes made evident that ragweed is able to regrow from lower buds and produces seeds after cut (Karrer 2014). Milakovic et al. (2014b) found that the number of produced seeds could be minimized by adequate mowing regimes. The authors also could show that the number of cuts must not be higher than regulary but they have to be set more sophisticated at dates that consider the phenological development of ragweed and its regrowth potential. In all this cutting experiments the efficacy was measured by reproductive trait like number of male inflorescences or number of female flowers/fruits.
The success of control measures against annual weeds like common ragweed depends very much on the number of seeds produced. Seed survival and seed longevity in the soil is crucial for long-term efficacy of control. Milakovic & Karrer (2016) showed that the above mentioned optimized cutting regime also was very efficient in reducing the resulting soil seed bank with respect to both absolute number and viability of ragweed seeds.
The competitive effect of different added seed mixtures to ragweed-infested soil was evaluated also by counting the number and viability of ragweed seeds under different cutting regimes – in greenhouse experiments and under field conditions. Final soil seed bank analysis after a long-term field experiment indicated that applying the fitted mowing regime is more efficient that the competitive performance of the tested seed mixtures.
In ecological terms, soil seed bank is the most relevant efficacy measure of control activities against annual plants like common ragweed.

Activities of Romanian group in the COST Project SMARTER FA-1203 during 2013-2016

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Romania was invited for the first time to take part in a global project dedicated to ragweed, due to clear expansion of this invasive weed to Eastern European countries and need for global evaluation of the danger. The SMARTER group consisted in nine persons from six centers, representing four different geographical regions, including the capital Bucharest.The specialists involved are biologists, agricultural specialists and medical doctors, already interested in the field of invasive weeds, environment and health impact of allergenic pollens.The main activities were focused on establishment of national aerobiology network, dissemination of main SMARTER objectives and achievements, searching opportunities for financed projects. The activity of SMARTER members consisted in publication of papers on ragweed and participation to many national and international conferences and symposia. Romania has no national aerobiology network and pollen measurements have been done between 2000-2012 in the West region only. No pollen measurement data are available for other regions. In the frame of SMARTER we established a three years collaboration with Reseau National de Surveillance Aerobiologique (RNSA) from France and started to perform pollen monitoring on regular basis in Bucharest.Monthly results during three pollen seasons, between 2013-2016, have been sent for validation to laboratory of RNSA and then to European Aeroallergen Network (EAN) . Our attention was focused on grasses and *Ambrosia artemisiifolia* pollens, which were measured for the first time in the South-East part of the country. The results of these first pollen monitoring seasons in Bucharest show significant amounts of allergenic pollens , mainly *Ambrosia artemisiifolia*, which was previously considered to be more prevalent in West regions and rural areas. Further efforts to recuperate the gap comparing to other European countries are required .There is an urgent need to establish a national aerobiology network and to develop financed research projects with health and environment authorities.

Variation in ragweed demography and modelling management impact

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To eventually better manage the invasive ragweed *Ambrosia artemisiifolia*, and reduce pollen and seed production, it is vital to understand i) natural variation in the performance of ragweed plants and populations, and ii) the long-term impact of different management strategies on ragweed population dynamics. We here present the first results of a 3-year demographic survey of over 40 ragweed populations across the European continent, covering a wide range of climates and habitats. For each population we assessed the density and performance of *A. artemisiifolia within permanent subplots, and collected data on* environmental variables likely affecting these properties. We confirm that seed and pollen production in the field is mostly determined by ragweed size, with larger plants producing more. In 2014, individual ragweed plants produced most seeds and pollen under the combination of warm and wet conditions. In grasslands, individual plants produced least seeds and pollen, while the production of seeds and pollen per square meter was highest on (ex)-arable fields, as compared to ruderal habitats and those along roads and railways. In the absence of ragweed management, we observed a general decrease in ragweed density over the years, most likely due to succession of the vegetation. We constructed a demographic model of ragweed and parameterised this with the field observations. Integrating experimental data on the impact of the ragweed leaf beetle *Ophraella communa* into the models revealed that this potential biological control agent can reduce ragweed population growth by more than 10-fold, and can reduce the soil seedbank faster in the long term. We similarly present results on the long-term effect of mowing regimes. Altogether, this work shows how the combination of observations, experiments and modelling can help designing effective management strategies for ragweed. We then present demographic models of *A. artemisiifolia* parameterized by the field data, and integrate experimental data on new management strategies (vegetation management and biological control) to project the long-term effects of these strategies for different populations.

Estimating the risk of non-target effects of *Ophraella communa*: from ecological to evolutionary perspectives

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Common ragweed, *Ambrosia artemisiifolia* (L.), is a worldwide invasive weed originating from North America. This annual plant is causing great damage to our society due its highly allergenic pollen, and because it grows as a weed in many crops in Europe, where it is hard to control. The leaf beetle *Ophraella communa*, also native to North America, was accidently introduced to Europe, where it was first recorded in 2013 in the Milan area of Northern Italy and Southern Switzerland, and from where it spread to more than 300 km both west- and eastwards up to 2015. Where it occurs, it significantly reduced pollen concentrations in the air and can cause complete defoliation and death of ragweed before flowering. Before the beetle can be actively spread to control ragweed across Europe, detailed target effects (efficiency) and non-target effects (host specificity) have to be conducted in order to balance benefits with costs. During 2014 and 2015, we conducted extensive host specificity tests both under controlled quarantine conditions and in open field tests in Northern Italy, using various designs for oviposition, and egg and larval transfer tests and both under no-choice and choice conditions. Test plants included other *Ambrosia* species occurring in Europe, taxonomically closely related crop plants (*Helianthus annuus* and *H. tuberosus*) and other native and exotic plant species. Our results achieved so far are greatly in favor for using this candidate for further spread, but additional region-specific plants need to be tested. I will also report on presently ongoing studies to elucidate the potential of *O. communa* to develop a “sunflower strain” using selection experiments and NGS tools. This will render long-term risk assessment and thus biological control more predictive.

Is *Ambrosia artemisiifolia* L. able to expand its invaded range northward in Western Europe?

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Ambrosia artemisiifolia L. (common ragweed, Asteraceae) is an invasive weed causing a health crisis in Europe, due to its highly allergenic pollen. In Western Europe the invaded range covers most of central and southern France, and northern Italy. Northwards beyond the edge of this range, occurrence of casual population have been described for years, but these populations do not appear to become invasive, and the species does not seem to spread. This situation raises the following question: Has the invaded range reached a limit or will the species continue its invasion northwards?

To answer this question, we followed two complementary approaches. First we set up an experimental garden in Belgium, 250 km north to the current invaded range, to see if the local climate allows the completion of the species reproduction cycle. Second, we performed an in situ measurement campaign in 12 population located beyond the edge, within the range but near the margin, and in the center of the invaded range. The aim of this campaign was to test whether the species had reduced plant performance towards range margins.

The results showed that the species is able to establish populations with high growth rates in Belgium. Furthermore, the species expressed similar performance across the considered areas, even beyond the current invasion front. No evidence of processes constraining the invasion was found, which suggests a great potential for invasion north to the current invaded range. In this uncertain situation, awareness actions should be considered in the northern countries.

A multi-level and multi-actor approach to biosecurity management and risk governance: a conceptual framework to support policy development for Ambrosia weed control

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Invasive species such as Ambrosia (an annual weed) pose a biosecurity risk whose management depends on the knowledge, attitudes and practices of many stakeholders. It can therefore be considered a complex policy and risk governance problem. Complex policy problems are characterised by high uncertainty, multiple dimensions, interactions across different spatial and policy levels and the involvement of a multitude of actors and organisations. This paper provides a conceptual framework for analysing the multi-level and multi-stakeholder dimensions of Ambrosia management. Potential and existing public, private and public-private management strategies are identified to address the interests and needs for different stakeholder groups across different levels. We conclude that policies that promote a mix of public and private Ambrosia management strategies may better respond to the needs and interests of different actor groups across different levels as compared to an “one-size-fits-all” approach. However, multiple policy strategies need to be aligned in order to lead to synergies and spreading coherent messages to the public. Collaboration may enhance the likelihood that biosecurity management and risk governance of Ambrosia is adequately implemented and enforced, but is most likely challenging to realize.

Ambrosia in Lithuania: coincidence or permanent phenomenon

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Common ragweed (*Ambrosia artemisiifolia* L.) frequently found in Lithuania among the most prevalent 3 species of the genus *Ambrosia*. For the first time in Lithuania this plant has been found in 1884. Since 1961 botanical research of genus *Ambrosia* has started in the country. Until 1980 original location of common ragweed were established in 5 areas of Lithuania, however during the next 10-year period the number of plants of this species increases dramatically (88 areas of habitat). After 1990 the spread of *Ambrosia artemisiifolia* L. plants started significantly decline. Common ragweed was found in 45 areas over the period of 1991-2000 and over the period of 2001-2010 was found only in 7 areas. In Lithuania since 2004 performed systematic aerobiological research has revealed the dynamics of *Ambrosia* pollen distribution in the air.

Airborne ragweed pollen has been studied during the 2004-2015 period. Aerobiological research stations are mounted in 3 Lithuanian cities (Vilnius, Klaipeda and Siauliai), which equipped by Hirst type volumetric spore traps. *Ambrosia* pollen was identified by microscope using twelve vertical strips method. The amount of pollen during the day was calculated in 1 m^3 of air (pollen concentration) and seasonal pollen index (SPI) indicated the amount of pollen during the pollen season.

Recent aerobiological research showed that *Ambrosia* pollen occurs in the air sporadically and the start, duration and intensity of pollen season vary year to year. According systematized research data were observed that in Lithuania ragweed pollen appears in July – August and occurs in air samples until the end of September. During the period of 2004-2015 in Klaipeda (seaside station) was estimated the lowest seasonal pollen index (SPI = 3), while the highest seasonal pollen index (SPI = 294) was determined in Vilnius (the most continental station). In 2011 the highest peak value (144 pollen/m^3) was recorded in the most continental aerobiological station of Lithuania (Vilnius). Furthermore evaluated bi-hourly ragweed pollen data showed that pollen is abundant in the air all the day, thus enhancing the assumption of the long-range transport.

State of art on *Ambrosia artemisiifolia* L. research in Croatia

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Common ragweed is an important weed species in Croatia and is commonly found in corn which is grown on around 300,000 ha as well as in other row crops. Under favourable field and weather conditions, ragweed emergence was usually observed as early as mid-March but peak germination and emergence of ragweed in continental Croatia most likely occur during April and May when mean monthly temperatures are above 10 ^{0}C. For better understanding of the population dymanics of common ragweed in continental part of Croatia, Under SMARTER COST Action FA1203, Task Force Population Dynamics, in three vegetation years (2014-2016) data (seedling establishment, seedling survival and growth, reproduction and seed survival) have been collected in the field from three different populations (MSHRZG777; MSHRSK776; MSHRDJ778).The monitoring of populations is ongoing and the first results are expected in the end of this year.

Croatia has high abundance of common ragweed and therefore causes major health problems to allergic people. It is extremely important to inform them on the daily movement of allergens to be able to adjust their daily activities. Public health institutes at 17 monitoring stations in Croatia follow the concentration of pollen in the air. Teaching Institute of Public Health Dr Andrija Stampar is a coordinator of pollen forecast for Croatia. Data are collected from monitoring stations and reproduced daily (or twice weekly) as a unique information on web site and mobile application. Institute participates in the education of citizens, preschool and school children through the activities of "European Mobility Week" on identifying and importance of removing ragweed. Each pollen season is followed by numerous articles, TV reports, talk shows and radio advertising on the allergenic plants. In cooperation with City Office for Health issued an educational brochure on Ambrosia, Institute each year organizes actions of removing this allergenic plant at various locations in the city.

Temporal and spatial variations in airborne Ambrosia pollen as an indicator of changes in the distribution and abundance of ragweed plants on the ground

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COST SMARTER aims to develop new innovative solutions for the the sustainable management of *Ambrosia artemisiifolia* L. (ragweed) in Europe and to assess their cost-

effectiveness. Several recent studies undertaken by members of COST SMARTER have examined temporal and spatial variations in airborne *Ambrosia* pollen at different scales, which can be used to determine the distribution and abundance of ragweed plants on the ground. Trends in airborne *Ambrosia* pollen recorded during a 10-year period (2004-2013) at 242 monitoring sites situated across Europe, showed that any changes varied locally and reflect changes in sources of the pollen. The accidental introduction of the oligophagous leaf beetle *Ophraella communa* LeSage 1986 (Coleoptera: Chrysomelidae) in Northern Italy coincided with a drastic decrease in atmospheric ragweed pollen that could not be explained by meteorology. Top-down *Ambrosia* pollen source inventories developed for all major regions infested with ragweed within the EU27 zone (e.g. France, Austria, Pannonian Plain, and Italy) that combine airborne pollen data, land cover data and habitat management models, can be used for forecasting atmospheric pollen concentrations and evaluating the integrated management of ragweed plants. Such studies show the importance of pollen monitoring in providing an early warning of the spread of ragweed, and represent a method for monitoring the management of the expansion of this invasive and noxious plant.

Looking for competitors to ragweed: results from garden and growth chamber experiments

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Competition by native plant species is considered among tools for ragweed (*Ambrosia artemisiifolia*) suppression. To identify successful competitors to supress *A. artemisiifolia*, its growth was compared with that of 10 native herbs and grasses under different temperature regimes in climatic chambers. The results indicate that the natives have different temperature optima that were always lower than that of *A. artemisiifolia.* This fact should be reflected if choosing the competing species and setting time of competition establishment. Effect of competitors were tested in two pot experiments in an experimental garden. In the first one, *A. artemisiifolia* was grown alone, in pairs with individual species and within species mixture. *Bromus erectus* was revealed a strong competitor causing decrease in number of *A. artemisiifolia* individuals and their size with positive effect of *A. artemisiifolia* on number of *B. erectus* individuals and without any significant effect on its size. In the second (still running) experiment, effect of two competitive mixtures and *Agropyron repens* is tested in soils differing in pH, nutrient availability and texture. Parallel to this effect of increased temperature on plant performance is tested.

Evaluating the costs and benefits of common ragweed management – An application to the Rhône-Alpes region in France

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Common ragweed has become one of the primary causes of seasonal allergies in Europe. Each ragweed plant produces up to one billion pollen grains, which are released after midsummer, when warmth, low humidity, and active breezes after sunrise create an ideal flowering environment. The pollen of common ragweed can travel over long distances. Therefore, ragweed pollen is a public health concern both at the release site and in more distant areas, where the pollen is transported by high winds. A sensitized person that is exposed to ragweed allergens can suffer from severe allergic symptoms. These symptoms include eye irritation, sneezing, runny nose, itchy throat, and more severe symptoms such as asthma, but also including chronic cough, wheezing, and difficulty breathing. Although the epidemiological literature has provided strong evidence for a significant relationship between ragweed pollen exposure and human health, concerns about the validity of these findings remain because endogeneity issues are often insufficiently addressed. This study contributes to the literature by quantifying the dose-response relationship between ragweed pollen exposure and health outcomes using historical data from the Rhône-Alpes region in France. Because the region is heavily infested by common ragweed, the local population is exposed to significant amounts of ragweed pollen, making the study area a suitable environment to investigate both the consequences of ragweed pollen exposure and the benefits of ragweed management. First, I identify the health consequences of ragweed exposure using a fixed effects approach allowing me to account for selection of people into geographical locations. I pay particular attention to the construction of geographically explicit exposure measures. My results indicate that ragweed pollen exposure is positively associated with various health outcomes. Second, I develop a framework to measure the health benefits associated with management activities. I illustrate this method by calculating the expectable health benefits from the arrival of the ragweed leaf beetle in the region. This beetle feeds on ragweed plants, and therefore diminishes the plant's potential to release pollen. By exploiting this link, I find that the beetle's arrival will have a significant and positive effect on health outcomes in the Rhône-Alpes region. The proposed framework can be used to evaluate other management activities, and compare their costs and benefits.

Climatic suitability ranking of biological control candidates: a biogeographic approach for ragweed management in Europe

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The selection of candidates is a critical step in a biocontrol program before more elaborate experiments are conducted. Here we propose a biogeographic approach to identify candidates and their combinations to potentially cover a large invasive range. We studied *Ambrosia artemisiifolia* (common ragweed), native to North America (NA) and invasive worldwide, and six NA biocontrol candidates for the introduced European (EU) range of ragweed. Specifically, we ask: 1) what percentage of the suitable ragweed range is also suitable for candidates; 2) does this relative overlap in NA correlate with their projected overlap in EU; and, 3) which combination of agents would cover the most area, and which particular biotypes would be needed to fill in the yet uncovered part of the suitable ragweed range in EU? For the first time, we constructed species distribution models simultaneously for a plant invader and its biocontrol candidates. Ordination techniques were used to explore climatic constraints of each species and to perform niche overlap with ragweed. We show a large overlap in climatic space between candidates and ragweed, but a considerable discrepancy in geographic overlap between EU (31.4%) and NA (83.3%). This might be due to niche unfilling and expansion of ragweed in EU and the fact that habitats with high ragweed occurrences in EU are rare in NA and predicted to be unsuitable for the candidates. Total geographical overlap is expected to decrease under climate change, but each candidate will respond differently. The relative geographical coverage of ragweed by candidates at home is largely transferable to the introduced range, even when plant invader shifts its niche. Our analyses also provide a guide for which candidates' combinations are expected to cover the most area and for which abiotic conditions to select for in developing climatically adapted strains for particular regions where ragweed is currently unlikely to be controlled.

Adaptive genetic divergence and increased competitive ability in invasive *Ambrosia artemisiifolia*

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Invasive alien plant species face different abiotic and biotic conditions in their native and introduced range; furthermore, biological control programs will create an additional mosaic of species interaction in the introduced range. Together, these make them ideal study systems to address basic question on rapid evolutionary changes of plant population. Differences in selection and genetic variation in relevant traits can make adaptation difficult to predict. We used a common-garden experiment to assess patterns of adaptation in the invasive plant, *Ambrosia artemisiifolia*, sampled across 11 populations in its native (USA) and 10 invasive (China) ranges, with the 10 Chinese populations having a different history of biological control management. We tested for adaptive trait evolution by comparing differentiation at neutral genetic loci (F_{ST}) with differentiation at quantitative traits (Q_{ST}). We estimated multivariate Q_{ST}-F_{ST} tests to determine if phenotypic differences between countries are due to adaptation. Between countries, we found that two growth traits are distinguishable from genetic drift ($Q_{ST}>F_{ST}$), and interestingly those two traits are correlated with climatic variation in the USA, but not in China. Within countries, we found more traits under selection in the USA than that in China. Moreover, we found increased adaptive potential in the Chinese populations without biological control history ($Q_{ST}>>F_{ST}$) than with biological control history. Additionally, a pairwise greenhouse competition experiment using 10 invasive Chinese populations in competition with a native USA grass showed a reduced competitive ability for populations with than without biological control history, which might be due to a tradeoff between herbivore resistance and plant growth. Our results indicate that adaptation in invasive Chinese populations does not parallel patterns in native US populations due to differences in abiotic and biotic selection, genetic constraints, or both.

The French evolution of exposition and health impact since 10 years

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Ambrosia artemisiifolia L. (common ragweed) was accidentally introduced into France and into different West-European countries in the 1860s. Its single first vector of introduction in Europe was red clover seeds (*Trifolium pratense* L.), probably coming from Pennsylvania, United States. Other introductions followed throughout the twentieth century in many places, especially during the First World War. *Ambrosia artemisiifolia* was later on introduced into France in many places and at different times but it's only around 1970 that its role was highlighted in pollinosis cases in Lyon. Currently, it is estimated that 10 to 20% of the Rhône-Alpes population is allergic to ragweed pollen. In Rhône-Alpes region the plant is particularly well established in every department since many years in flatlands and hills. Ragweed is also present in other French regions, going from only a few plants to sometimes larger amounts but do not seem to cover wide spaces as in Rhône-Alpes. In France, all the stations are provided with the same kind of Hirst-type pollen trap and analyses are carried out following the procedures establish by the new technical sheet of CEN 264 approved in December 2015 CEN/TS 16868. The main characteristics of Hirst type pollen traps (Lanzoni or Burkard) are to work continuously, to inhale a constant air volume and to impact continuously particles present in the air on a transparent support. Analyses of the samples are made by optical microscopy following a normative key of determination. If all pollens are identified and counted, ragweed pollens are subject to a particular attention. They are average-sized (20µm), tricolporate and spherical-shaped with wide spines regularly distributed. If all pollens are identified and counted, ragweed pollens are subject to a particular attention. Analysis of ragweed pollens reveals that the plant is present along the 45th parallel either in North America or Europe. With records covering more than 10 years pollen data, RNSA follows the evolution of ragweed pollination: annual pollen index, start and length of the season, new areas with ragweed pollen, etc. Qualitative and quantitative data allow to obtain daily or bi-hourly concentrations by m3 of air. Daily concentrations allow the evaluation of allergy sufferers exposure to this pollen, while the bi-hourly concentrations, when they exist, allow to complete the distribution maps of the plant at a local scale. The measurement of health impact related to the exposure to pollen could be done in different ways, according the protocols used in each country. Some countries like France use "a clinical index" calculated during all the season thanks to a network of doctors. In France, the annual pollen index and clinical index increased in most of the French stations in areas infested by ragweed. 2013 was a specific year which was favourable towards allergy sufferers: the germination began two or three weeks later than usual and the development of the plant kept this lateness. Ragweed pollen is a real public health problem increasing every year and invading more and more geographical areas.

Natural enemies of *Ambrosia artemisiifolia* in invaded areas

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Common ragweed (*Ambrosia artemisiifolia*) is an invasive alien species indigenous to North America. Ragweed is a big threat to agriculture and has a serious impact on human health. The species is an excellent target for biological control. However, impact of native natural enemies (insects, mites, pathogens) as well exotic ones on ragweed in invaded areas is poorly known. The regional and habitat differences in ragweed's natural enemies were investigated in Slovakia, Italy, and Switzerland during 2013-2016. Hereto, the identical sampling protocols were used to survey natural herbivores across various populations. In particular, the level of attack by specialist versus generalist natural enemies among populations was assessed. The only eriophyid mite, *Aceria artemisiifoliae* Vidović & Petanović (Acari: Eriophyidae), was found as a specialist from among all natural enemies during the study. In addition to the native herbivores, one alien insect species was in the spotlight. The ragweed beetle, *Ophraella communa* LeSage (Coleoptera: Chrysomelidae) is a North American species which was first time noted in North Italy (Lombardia) and South Switzerland (Ticino). Most of the common ragweed populations in this area are heavily attacked by *O. communa*. This species is regarded as a successful biological control agent against *A. artemisiifolia* in China. Nevertheless the threat to closely related wild or crop species and the level of their damage under field conditions remain unclear. Here we investigated the potential of *O. communa* to feed on non-target host plants in the field. The field surveys were complemented with open field simulating herbivory experiments in the garden. These field experiments helped to detect impact on ragweed growth, especially number of female flowers and aboveground and belowground biomass. All of the research topics will be discussed and potential of herbivores and pathogens for biological control highlighted.

Plant associated bacteria to reduce the growth of *Ambrosia artemisiifolia*

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The use of herbicides against *Ambrosia artemisiifolia* is limited or in some cases also not allowed. The use of bioherbicides in weed management is until now unexploited. We evaluated plant associated bacteria for their bioherbicidal activities against *A. artemisiifolia*. The vast majority of microbes in the environment is uncharacterized and represents a major reservoir of new bioactivities including herbicidal effects. The use of microbes with herbicidal activity is a novel approach, which likely involves novel mechanisms and is less prone to cause environmental damage.
The interaction between microorganisms and plant can be mutualistic, neutral or deleterious. Microorganisms may show dose-dependent effects such as some weed-associated microorganisms, which may exert pathogenic effects on their host when present in high cell numbers. Along these lines we isolated plant-associated bacteria from *Ambrosia artemisiifolia* L. and tested their effects on their host and sunflower. From three different locations in Austria (Burgenland, Lake Neusiedl) bacteria were isolated from the rhizosphere as well as from the root and stem endosphere. A total of 296 unique isolates was tested for different functional activities as well as their effects on seeds of ragweed, sunflower and lettuce. Seeds were inoculated with the bacterial strains and the germination rate was compared to untreated seeds. Twenty-five strains reduced germination up to 32% of ragweed. From these 25 strains two strains also inhibited the germination of lettuce seeds. Additionally, *Pseudomonas* strains from different locations in Austria were tested on *in vitro* and greenhouse plants. We observed that different *Pseudomonas* strains induced different effects on ragweed plants, like growth inhibition, germination reduction and inhibition of re-sprouting after wounding.

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Biotic resistance to invasive alien plant: Using a multi-trait approach to design resistant communities

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The invasion of alien species (IAS) contributes to ecosystem degradation, complicating efforts to restore degraded systems. Therefore, a key question arising is whether we can prevent the successful establishment of IAS during restoration by designing resistant plant communities. The theory of limiting similarity has been proposed as a tool for predicting biotic resistance of a target community to potential IAS. It states that IAS will be unlikely to establish if native species with similar traits are present in the resident community. However, most studies base trait similarity on single traits or broad classification of life forms. In this study we investigate whether we can predict biotic resistance by using using a system of linear equations and a multiple-trait approach based on more specific functional traits related to competitive strength obtained from trait databases (see Laughlin 2014, Ecol. Lett. 17: 771-784). For doing so, a specific community was designed based on the trait similarity of grassland species native to central Europe and against each of two common IAS (*Ambrosia artemisiifolia* and *Solidago gigantea*). In a greenhouse experiment both specifically designed communities were separately tested against both IAS along with a control consisting on a monoculture of each IAS. Our results show that the community designed to repel *S. gigantea* was successful at suppressing both IAS, supported by the lower values of aboveground biomass found compared to the one designed to repel A. *artemisiifolia* and the control. A negative correlation between native and IAS aboveground biomass was only found for A. *artemisiifolia*. While these findings would suggest a partial support to the limiting similarity theory for *S. gigantea*, the suppression success of the same mixture on *A. artemisiifolia* indicates that other factors might better explain the suppression effect. That is, resistance seems to be related by either a biomass-driven suppression or the presence of particular highly competitive species, i.e. *Achillea millefolium*. Overall, our results indicate that species-specific effects can influence early stages of such artificial communities rather than limiting similarity. The lack of support could also be a consequence of failing to include really meaningful traits with respect to the most wanted functions of early development competitive suppression. Such can lead to the discussion if we have the right traits available in current trait databases.

Phytotoxic metabolites produced by *Colletotrichum gloeosporioides* as potential natural herbicides for the control of *Ambrosia artemisiifolia*

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Ambrosia artemisiifolia L. (common ragweed) is a plant native to North America that has become a widespread and troublesome invasive weed of cultivated and uncultivated areas all over the World. Besides the difficulties of its management as a weed, the major concern is its pollen grain that is carrier of noxious allergens which cause sensitization of the allergenic human population, generating huge medical costs. Searching for alternatives to the common methods used for controlling this weed, a strain of the fungal pathogen *Colletotrichum gloeosporioides*, responsible for necrotic spots on *A. artemisiifolia* leaves and stems, was studied for the possible production of toxic metabolites that might be used as natural herbicides against the weed. When grown in a defined liquid medium the fungus produced phytotoxic filtrates that were exhaustively extracted with EtOAc. The organic extract, retaining the phytotoxicity, was purified by column chromatography affording four phytotoxic fractions. The residues of these four fractions were purified by several TLC steps yielding some pure metabolites. These were assayed in different biological systems, i.e.: on punctured detached *A. artemisiifolia* leaves, on excised plantlets, as inhibitors of germination of parasitic weed seeds, for degradation of chlorophyll on aquatic plants, and for their antibiotic and zootoxic activities. The results of the chemical and biological characterization of the compounds will be presented.