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Ambrosia day, 2014 - Ragweed allergy: 15 years of prevention April 4<sup>th</sup>, 2014, Rho (Milan), Italy

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### THIRD INTERNATIONAL RAGWEED CONFERENCE April 3<sup>rd</sup>-4<sup>th</sup>, 2014, Rho (Milan), Italy













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### THIRD INTERNATIONAL RAGWEED CONFERENCE AND AMBROSIA DAY 2014 April 3<sup>rd</sup>-4<sup>th</sup>, 2014, Rho (Milan), Italy

#### M. BONINI<sup>1</sup>, M. THIBAUDON<sup>2</sup>, T. KOMIVES<sup>3</sup>

<sup>1</sup>Head Organizer of the Conference - Email: maira.bonini@aslmi1.mi.it <sup>2</sup>General Secretary of the International Ragweed Society <sup>3</sup>President of the International Ragweed Society

This special issue contains papers presented at the "Third International Ragweed Conference" (3rd I.R.C.) held in Rho (Milan), Italy on April 3<sup>rd</sup>-4<sup>th</sup>, 2014. Conference Chairs are Maira Bonini (Local Health Authority Milan 1; Italian Association of Aerobiology - Italian Monitoring Network in Aerobiology  $[A.I.A.-R.I.M.A.^{\textcircled{B}}]$ and Tamas Komives (Plant Protection Institue, Hungary), Head Organizer of the 3rd I.R.C. and President of the International Ragweed Society (I.R.S.), respectively. The "Third International Ragweed Conference" is jointly organized to "Ambrosia day, 2014 - Ragweed allergy: 15 years of prevention" Conference (Local Health Authority Milan 1, Lombardy Region, Italy).

Both Conferences are also the scientific events organized for the "National Pollen Day" (G.N.P.<sup>®</sup>) of the Italian Association of Aerobiology (A.I.A.).

The meeting focuses on the theoretical and practical aspects of ragweed (Am-

*brosia artemisiifolia* L.) spread in Europe and other parts of the world giving special emphasis to the harmful effects of its pollen.

Basic research along with practical considerations in the fields of pollen allergen genetics, human (and animal) health, public health, aerobiology and meteorological aspects, ecology and management are emphasized. In addition laboratory, pilot, and modeling studies leading to innovative approaches are considered.

The conference is attended by many scientists from at least 20 countries and a total of 56 presentations are included in the final program. The presentations are arranged into six separate sessions according to their topics.

It is our pleasure to thank the invited speakers and the leading scientists actively involved in Ambrosia-related research who served as chairpersons of the scientific sessions. We are also grateful to the authors of papers for submitting the camera-ready copies, which made possible the timely publication of this issue.

Finally, special thanks to:

- the General Director of the Local Health Authority Milan 1, Dr. Giorgio Scivoletto, for supporting the organization of the Conference;
- the University of Milano Bicocca, the Italian Association of Aerobiology (A.I.A.) and the Park of Ticino for supporting the Conference;
- the Director, Dr. Edgardo Valerio, and all collaborators of the Department of Medical Prevention of the Local Health Authority Milan 1 for their daily activity permitting the realization of the conference;
- the President of the Italian Association of Aerobiology (A.I.A.), Dr. Roberto Albertini, and Prof. Sandra Citterio, University of Milano Bicocca, Department of Earth and Environmental Science, for their direct collaboration in the realization of the Conference.

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### THIRD INTERNATIONAL RAGWEED CONFERENCE AND AMBROSIA DAY 2014 April 3<sup>rd</sup>-4<sup>th</sup>, 2014, Rho (Milan), Italy

#### G. SCIVOLETTO

General Director - Local Health Authority Milan 1

As General Director of the Local Health Authority Milan 1, I'm honoured to hold the "Third International Ragweed Conference" (3<sup>rd</sup> I.R.C.).

The conference is jointly organized to "Ambrosia day, 2014 - Ragweed allergy: 15 years of prevention", Italian Convention.

The jurisdiction of the Local Health Authority Milan 1 represents one of the most polluted area by ragweed in Europe and the people who live in this area know well the health problems related to the widespread of this highly allergenic plant, therefore the location of the Conferences is well connected to the local context.

Since the beginning of the public health problems related to the ragweed presence, the Department of Medical Prevention of the Local Health Authority Milan 1 was very active in this issue, following closely the regional indications and went further, collaborating with the Lombardy Region in order to improve and perfect the preventive measures. A set of actions aimed at primary prevention of ragweed pollinosis were implemented: aerobiological monitoring; surveillance of grow over stands; epidemiological studies; assessment of direct health costs due to ragweed; experimental studies demonstrating the effectiveness of different methods to limit the ragweed spreading, currently at the root of the regional indications for the prevention of ragweed spreading; participation in the European projects and, last but not least, education of the population and of the public authority, also by organizing conventions like in the occasion of the 3<sup>rd</sup>-I.R.C.

The international collaboration is essential for development of research, education, information and laws, therefore the 3<sup>rd</sup>-I.R.C. is a good opportunity to share knowledge and experiences. Meanwhile, the "Ambrosia day" is a good point to outline of the situation after 15 years

from the first regional preventive measure against ragweed, taking into consideration the most recently clinical-diagnostic developments, the available preventive tools and the results and perspectives of the European projects. The presentations are arranged into four separate sessions and two roundtables according to their topics and a focus is dedicated to Ophraella communa, a beetle accidentally widespread in northern Italy and in particular in the area of the Local Health Authority Milan 1, which seems to have had a positive effect on the levels of the airborne ragweed pollen and might be a candidate for the biological control of ragweed. The convention is open to all health workers, local and public administrators and stakeholders. Hoping in a fruitful scientific work, a special thanks is dedicated to all the employees of the Local Health Authority who collaborated in the realization of these two important scientific events.

### VIII giornata nazionale del Polline Associazione Italiana di Aerobiologia

R. ALBERTINI

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L'Associazione Italiana di Aerobiologia (A.I.A.) è una società scientifica nata nel 1985 dalla collaborazione tra biologi, medici, agronomi ed altre figure professionali. La sua mission è promuovere e coordinare gli studi nel campo dell'aerobiologia e delle patologie legate ad agenti aerodispersi e divulgarne conoscenza e applicazione attraverso un approccio multidisciplinare. L'aerobiologia studia le particelle di origine biologica presenti in atmosfera, le fonti che le producono, le modalità del loro trasporto in aria e gli effetti che possono avere sull'ambiente (indoor e outdoor), sull'uomo, ma anche su animali, piante, manufatti, sulla conservazione dei beni culturali, ecc.. A.I.A., come ogni anno dal 2007, promuove e organizza una campagna di informazione, denominata "Giornata Nazionale del Polline" (GNP®), per sensibilizzare l'opinione pubblica su questi temi. Il padre del management P. Drucker diceva che bisogna "Misurare per conoscere e conoscere per gestire" e in aerobiologia il monitoraggio di pollini, spore fungine ed allergeni aerodispersi è un importante parametro di qualità dell'aria che, diffuso attraverso molteplici canali mediatici, consente ai Medici di Medicina Generale e Specialisti di migliorare le prestazioni diagnostiche e terapeutiche per i soggetti allergici; i pazienti possono riconoscere e gestire i loro sintomi anche lontano dalle sedi abituali di residenza, mentre Enti e Istituzioni preposti alla tutela della salute, dell'ambiente e della qualità dell'aria possono migliorare le attività di prevenzione sanitaria, ambientale e di gestione del territorio. La GNP® ha lo scopo di diffondere notizie sulle iniziative in campo aerobiologico, fornendo ai diversi soggetti, impegnati nello studio di questi fenomeni, l'occasione per un momento di incontro e collaborazione per sensibilizzare la platea più vasta possibile. La GNP® 2014, giunta quest'anno alla sua ottava edizione, è celebrata da A.I.A. con il patrocinio della 3ª Conferenza Internazionale sull'Ambrosia (3rd International Ragweed Conference) e del Convegno italiano "Ambrosia Day 2014 - Allergia all'Ambrosia: 15 anni di prevenzione", importanti convegni che affrontano i diversi aspetti sanitari e ambientali legati alla diffusione, in Europa e nel mondo, di questa pianta erbacea infestante che produce pollini in grande quantità e altamente allergenici. La Conferenza Internazionale è organizzata da International Ragweed Society (IRS) e ASL Milano1; il Convengo italiano è or-

ganizzato da ASL Milano1. Attraverso la partecipazione attiva ad eventi di questo genere, così importanti anche in ambito internazionale, A.I.A. testimonia il suo ruolo di Società scientifica la cui azione è indirizzata verso tutti gli attori che svolgono quotidianamente un compito importante in ambito aerobiologico. In ultima analisi, l'obiettivo di A.I.A. evidenziato in un contesto come quello della GNP®, a maggior ragione legato ad un importante evento internazionale, è anche quello di sensibilizzare l'opinione pubblica e le Istituzioni su questa attività e perseguire il riconoscimento normativo a livello regionale e nazionale del monitoraggio aerobiologico, così come è andato strutturandosi nel corso del tempo, per non disperdere conoscenze, competenze e informazioni. Le sinergie tra le discipline biologiche e mediche, Istituzioni, Associazioni scientifiche, di pazienti e Enti sono fondamentali per una lotta a trecentosessanta gradi nei confronti delle patologie allergiche respiratorie, per l'ottimizza-zione delle risorse utilizzate nelle attività di monitoraggio e destinate al servizio offerto al paziente che ne migliorino la qualità di vita e riducano i costi sanitari diretti ed indiretti per la collettività.





### Ecological and Management aspects of ragweed

Chair: Rodolfo Gentili, Tamas Komives

# Ecological and management aspects of common ragweed

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Ambrosia artemisiifolia L. (common ragweed) is spreading over the European territory, which is raising public-healthrelated issues. Common ragweed is an annual spring weed that colonizes perturbed areas such as roadsides (1). Road network, railways and waterways are known to be dispersal corridors for invasive species including common ragweed which seems, by its biological characteristics, particularly well adapted to these habitats.

In France, vegetation surveys showed that common ragweedhas a wide ecological tolerance and can found on habitats differing in terms of vegetation cover and typeof soil (2) (Table 1).

The long vegetative stage of common ragweed with a late seed production should support the effective management of this species. Furthermore, different management tools (chemical, soil tillage, biological, hand weeding) can be used. One of the problems is to find, according to the stage of development of the plants, the well-adapted method to the habitat concerned by the presence of the plants (road-sides, river banks, fields, urban areas, etc) in order the increase the success of the management (3). For example, a comparison between different herbicides indicated that common ragweed is an easy-to-wet species and that foliar active ingredients are quickly and completely absorbed by its leaves.

On a non-crop area, a 100% efficacy can be theoretically reached at vegetative and early flowering stages (3). However, after early treatments, new seedlings emergence led to the presence of vigorous plants that bore numerous flowers and produced high levels of pollen. After treatment at the flowering stage, flower production was not significantly affected, but achene weight was decreased by 60 to 70% and seed viability was very low (Table 2). Despite the high efficacy of herbicide, it is difficult to attain the two objectives of reducing seed production and pollen production by means of only one treatment (3).

In the case of management without herbicide, the competition is rarely enough strong to completely stop the flowering and the seed production and spontaneous vegetation offers a too weak competition to control common ragweed (4) (Table 3).

More generally, whatever the method chosen, the high adaptability of the plant and the difficulty of managing simultaneously pollens (pollinosis) and seeds (invasion) are largely responsible for the spread of the plant. Biological controls are not sufficient to control the dynamic of common ragweed. Predation rate by invertebrates was estimated on seed cards in cultivated fields: a mean preda-

Soil factor	Minimum	Median	Maximum	
Clay %	1.8	14.7	42.8	
Silt %	0.8	23.8	64.0	
Sand	11.5	62.4	97.4	
C/N	0.7	10.7	38.7	
pH (KCl)	4.1	7.7	8.6	
Organic matter (g/kg)	0.6	26.8	209.0	
CaCO <sub>3</sub> (g/kg)	0.2	94.1	761.0	

Tab. 1 - Chemical properties of soil in sites occupied by common ragweed.

	Control	N/2 Glu	N Glu	N/2 Gly	N Gly
Biomass	288	259	213	229	236
(mg per plant)	(49.4)	(16.5)	(21.6)	(18.0)	(18.7)
Seeds	27.4	16.9	15.1	19.3	24.9
per plant	(3.78)	(0.83)	(1.41)	(2.21)	(2.18)
Seed weight	6.4	1.2	2.2	3.3	3.3
per plant (g)	(0.69)	(0.10)	(0.32)	(0.51)	(0.11)
100-seedweight	445	134	135	163	180
(mg)	(5.0)	(3.5)	(3.3)	(8.8)	(8.7)

Tab. 2 - Influence of glufosinate (Glu) and glyphosate (Gly) on biomass, and seed production and weight of common ragweed plants treated at the flowering stage. N: 150 and 1080 g ha-1 glufosinate and glyphosate, respectively. (in parentheses SEMs).



Habitats	Surviving plants (%)	Meanbiomass (per plant)	Flowering ragweeds (%)
Alfafa	43,75% a	4,05 ns	6.3 a
Lawn	93,75% b	2,41 ns	13.2 b
Spontaneous vegetation	100% b	15,37 ns	25% с
Sown wildflowers	100% b	20,36 ns	24.2% с

Tab. 3 - Effects of different habitats on common ragweed development

tion of 24,61 % of *Ambrosia artemisiifolia* seeds was observed and was not different from that observed for other weed (mean = 28,44%) (6).

Resistance to herbicides or changes in defense against predators (7) have been already demonstrated. It appears that only a preventive management is effective to control its spread before the establishment of a seedbank. The pooling of successful of management actions and the exclusion of practices that favour the spread of the species are necessary to prevent the situation from getting worse. The curative management requires a long-term work in a context of fundings decrease where ragweed control is not always the priority.

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### WEEDINESS ASSESSMENT OF ANTHROPOGENIC PHYTOCENOSES ON THE BASIS OF SATELLITE REMOTE SENSINGE DATA (A CASE EXAMPLE OF COMMON RAGWEED)

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The use of remote sensing data to detect patches of common ragweed (Ambrosia artemisiifolia) is more complex than the traditional problem of the detection of the crops types and their yields. Since common ragweed grows irregularly within a field, its spectral characteristics are strongly dependent on the type of the main crop culture, and on the degree of field weediness. The purpose of this research is to assess the ambrosia weediness in the South of Russia. The results of the decoding of the medium resolution images from Landsat satellites, which allowed identifying pockets of common ragweed in agrophytocenoses of the South of Russia using the Normalized Difference Vegetation Index - (NDVI) are presented. Due to the fact that uniquely identifying of the ragweed on pictures is not possible at the current stage of our study, we use the term "risk of weediness". The approach is based on the use of remote sensing and geographic information systems, combined with field studies for

the construction of risk maps for the ragweed distribution. The characteristic feature of the vegetation and its condition is the spectral reflectivity. Agrophytocenoses infested with common ragweed in the Rostov region have been selected for the experiment. Calculating of vegetation indices has been performed in order to work with the spectral characteristics of common ragweed. The 4-channel spectrometer SkyeInstruments SpectroSense 2 + measuring radiation in the 3 channels in the visible and near infrared bands has been used to determine the spectral brightness of common ragweed in the field study. Also imagery from Landsat 4-5 Thematic Mapper (TM) has been created. The vegetation index on satellite images was calculated with the help of ArcGisDesktop software. Finally the NDVI index was calculated as the result of the image analysis and the values of the vegetation index for common ragweed were determined by comparisons with the field sampling data.

The most important information for building the risk maps is information on vegetation peaks of both the ragweed and related plant cultures. We can speak with a high degree of confidence only about the weediness of grain fields in late August early September, as the vegetation peaks of common ragweed and grain crops differ. The results obtained at this stage of our investigation indicate the absence of extensional areas heavily infested by the ragweed in the study area in the South of Russia at nowadays. Strong weediness is systematically observed only at the edges of fields and along the roadsides. This result confirms the conclusions of the field studies about the long-term efficiency of the introduction from the North America of the ragweed leaf beetle (Zygogramma suturalis) in the period 1978-1990. A retrospective analysis of archival satellite images taken during the second half of the 1980s, shows a reduction of the area covered by weeds in this period.

### A process-based approach to predicting the effect of climate change on *Ambrosia Artemisiifolia* in Europe

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The process-based model, Sirius 2010 (1), was used to predict the future potential for range expansion of *A. artemisiifolia* under climate change scenarios. Parameters driving the growth and development of *A. artemisiifolia*, including seed dormancy and germination, biomass partitioning and phenology were calibrated using data from literature and growth experiments. The model predicts plant fecundity and pollen production in response to environmental variations, e.g. daily temperature, rainfall and  $CO_2$ concentration. Future predictions were made across Europe based on projections from the HadCM3 global climate model for the near future (2010-2030) and long-term future (2050-2070) (2) with high emission scenario (A1B). The growth rate of the seed bank was calculated from the predicted plant fecundity and was presented as a measure of the



Fig. 1 - Logarithm of relative increase in pollen production, Pfuture/Pbs for the HadCM3(A1B) long term future scenario (2050-2070), where Pbs and Pfuture are pollen production for the baseline and future climate scenario.

potential for A. artemisiifolia population to establish (3). The future southern limit of the range of A. artemisiifolia was predicted to change very little as future rainfall patterns meant most of Spain, southern Italy and Greece remained too dry to maintain viable populations. However, the potential available habitat was predicted to extend further north and east under climate change as increasing summer temperatures resulted in higher probability of the plant completing its life cycle and producing mature seeds. Where A. artemisiifolia is currently well established, the model predicted smaller relative increase in pollen production. However, at the Northern range, larger increase in pollen productions were predicted which could potentially create health problems (Fig. 1).

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### Ambrosia artemisiifolia in italy: integration of classic biological control approach into a multidisciplinary program

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Common ragweed (Ambrosia artemisiifolia L.), an annual plant of the Asteraceae family native for North America, became an alien weed all around the world, including Asia, Australia and Europe. In Italy ragweed is recorded in the Northern and Central Regions, but it is very abundant mainly in the Po Valley. In the framework of the COST Action SMARTER, a sustainable program of management of this target weed is in progress to control the weed by using least toxic approaches. Among them, special emphasis will be attributed to classic biological control of weeds, evaluating the restricted host range of selected candidate agents through a series of filed observations and bioassays carried out in both no-choice and choice conditions. Candidate agents - generally arthropods

like insects and mites, have the same geographical origin of the target weed, with whom they are closely connected by a process of co-evolution. In this way the selected candidate agent(s) will feed, oviposit and develop only on one plant species, showing a clear monophagy or restricted oligophagy. Looking more in details at the potential biocontrol candidates under evaluation for ragweed, the most promising looks like Ophraella communa (Coleoptera: Chrysomelidae), a gregarious leaf beetle of North American origin, because its gregarious behavior, the strong fitness and the prolonged impact on its host plant. Moreover, during the summer 2013 large populations of Ophraella communa were recorded in several sites near Milan and in the Po Valley, confirming an accidental introduction in that area (1). In the frame work of a classic biological control program and in cooperation with other Italian and European teams, ENEA C.R. Casaccia will be involved in the evaluation of the host range of this leaf-beetle of North-American origin: field and laboratory trials will be carried out to evaluate the impact of the selected candidate agent alone and/or in combination with other methods of control.

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### INTENSIVE GRASSLAND SEEDING TO CONTROL AMBROSIA ARTEMISIIFOLIA L. SPREAD

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Ambrosia artemisiifolia L (common ragweed) has become a major problem across European countries due to its highly allergenic pollen and to its invasiveness, favoured by its ecological amplitude and phenotypic plasticity,. In this work we assumed that a strong competition for resources may inhibit ragweed growth, generating a negative feedback to its establishment and spreading. The specific goal was to determine by a field experiment the effect of mixtures of native species on ragweed growth and fitness, assessing the principal mechanisms of interaction. To this aim, we considered the following three different treatments within a quarry area (Botticino Basin, Brescia, Lombardy) extremely invaded by A. artemisiifolia: a) spontaneous succession (SS), starting from a bare ground; b) seeding of hayseed (H); c) seeding of a high density commercial seed mixture (CS). In these sites within random plots, we recorded vegetation percent cover, total number of common ragweed individuals and eight plant traits of the species such as plant height, lateral spread, inflorescence size and numbers, maximum leaf size and seed weight. Preliminary results highlighted strong differences in traits among the treatments. In general the CS, showed high vegetation cover (95%) and significant lower number of common ragweed individuals (2±1 individuals) with respect H (cover 75%; 15±5 individuals) and SS (cover 55%; 50±9 in-

dividuals). Moreover the CS treatment accounted the strongest reduction of common ragweed biomass in terms of plant height, lateral spread and leaf sizes; SS plots showed the highest values for all measured traits. However, for some parameters (for instance, lateral spread and inflorescence size), no significant difference was recorded between CS and H stands. These results suggested that enhancing interspecific competition establishing native plant communities (both from hayseed or seed mixture) could be a useful technique for restoring diversity on sites invaded by common ragweed. Density of seeding played a key role in reducing biomass and fitness of this extremely invasive species.

G.E.A.

# Controlling common ragweed (*Ambrosia Artemisiifolia*) in sunflower with high efficacy

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The main cause of allergy and pollen asthma in North America and Central Europe is pollen from ragweed (Ambrosia) a widespread genus in the Asteraceae. In Europe short or common ragweed (A. artemisiifolia) is prevalent. Common ragweed is the number one weed in Hungary: it covers ca 5% of the arable land. The competitive weed causes huge losses in row crops. In addition, because of the high allergenicity of its pollen, common ragweed is a huge burden on the health care system of the country. In Hungary, common ragweed infestation is heaviest in sunflower (Helianthus annuus), the third most important crop of the country (also an Asteraceae plant, thus, a botanical relative of common ragweed). In August-September common ragweed produces the overwhelming majority of allergenic pollen in the air ever in urban areas. Increasing importance of bioenergy production together with recent advances in improving the dietary value of sunflower oil will certainly increase the production area of sunflower in the future. Thus, there is an urgent need for new tools to improve the control of common ragweed in this crop. In the presentation we show the results of a seven-year (2007-2013) field study on the control of common ragweed by two acetolactate synthase inhibiting postemergence herbicides (imazamox and tribenuron methyl) in sunflower hybrids (NK Meldimi, NK Neoma and PR63E82) that carry the resistance gene against such herbicides. Common ragweed control by these herbicides was stable and excellent: they suppressed the growth of the weed plant until the canopy closure of the crop (8-leaf stage). Common ragweed plants germinating after this date were unable to compete

with the crop: although they survived, they remained small (ca 70% reduction in height), produced ca 90% less male inflorescences (source of the allergenic pollen), and caused no significant reduction in the crop yield. In areas where sunflower germination was poor, however, a second, mechanical common ragweed control measure was necessary to keep the weed density below damaging levels. In areas where sunflower germination was poor, however, a second, mechanical common ragweed control measure was necessary to keep the weed density below damaging levels.

#### Acknowledgement

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### SUSTAINABLE MANAGEMENT OF AMBROSIA ARTEMISIIFOLIA IN EUROPE (COST FA1203-SMARTER): OPPORTUNITIES AND CHALLENGES

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The EU-COST Action FA1203 on "Sustainable control of Ambrosia artemisiifolia L. (Asteraceae) in Europe (SMARTER)" was launched in February 2013 and will last for four years. Thirty-three countries have already signed the Memorandum of Understanding and over 180 researchers with specialists in weed research, invasive alien species management, ecology, aerobiology, allergology and economics are registered participants of SMARTER. COST Actions interlink nationally funded research projects and enable and finance conferences, working groups, training schools and research exchanges. SMARTER aims to initiate and develop long-term and sustainable control methods, to integrate these into existing mechanical and chemical control measures, and to quantify the success of these measures both for agriculture and health. The focus is on biological control methods with

insects and fungi (especially using alien species from the area of origin of Ambrosia) and vegetation management to achieve a competitive plant cover. The envisaged outcomes of SMARTER will allow the various stakeholders to select optimal habitat- and region-specific combinations of control methods. After an introduction and overview of the structure and the state of the Action, we briefly describe two planned activities typical for our Action, a study on the population dynamics of Ambrosia in different climates and habitats in Europe as a basis for estimating the efficiency of control measures, and an interdisciplinary study to clarify the impact the of North American native Ambrosia leaf beetle Ophraella communa (Coleoptera: Chrysomelidae) recently found in southern Switzerland and northern Italy 1, 2. Finally, we will briefly present the newly launched iPhone App "The SMARTER Ambrosia Reporter", which mainly serves to report the occurrence of Ragweed when you see it around. It also features a version for professionals, including more detailed information about the location and the vegetation.

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# Session II



### Aerobiology and meteorological aspects of ragweed pollen movement

Chair: Giuseppe Frenguelli, Roberto Albertini

### RAGWEED POLLINATION IN EUROPE

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Within the European Aeroallergen Network (EAN) are gathered all pollen data coming from almost 250 measurement stations located in 37 countries from Iceland to Turkey for instance. All these stations are provided with the same kind of Hirst-type pollen trap and analyses are carried out following the procedures enacted by the International Association for Aerobiology (IAA) and the European Aerobiology Society (EAS). In North America, there are also some measurement stations that follow the same protocol. 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. Characteristics of these pollen traps are subject of a normative description within the CEN 264, thanks to the new working group set up for the EAS. Analyses of the samples are made by optical microscopy following the approved procedures of the IAA which are included within the future technical sheet of CEN. If all pollens are identified and counted, ragweed pollens are subject to a particular attention. They are average-sized (20 µm), tricolporate and spherical-shaped with wide spines regularly distributed. Qualitative and quantitative data allow to obtain daily or bihourly concentrations by m<sup>3</sup> 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. To check the efficiency of fighting methods against ragweed, the set up in proximity position of passive pollen traps SLT (Sigma2 Like Trap) allows to evaluate quickly differences of exposure to ragweed pollens in infested or not infested areas. 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" (1) calculated







Fig. 2 - Ragweed pollens and score-symptom in France (2010-2012)



during all the season thanks to a network of doctors (Fig. 1).

At European level, EAN set up, thanks to the work of our Austrian colleagues (S. Jager and U. Berger), a system to collect directly the symptoms by the patients themselves. This system, named PHD (2) (for Pollen HayFever Diary), allow to calculate a "score-symptom" (Fig. 2).

These parameters (clinical index and score symptom) allow to establish individual levels of sensitivity of patients and averages of health impact of the exposure to ragweed pollens on an area.

Measures analysis of ragweed pollens reveals that the plant is present along the 45th parallel either in North America or Europe. The evolution of the average annual amounts increased from 2010 to 2012 but decrease in 2013. Daily data are used to make dispersion models of pollens showing a shift from West to East during all the season.

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. So the pollination in Europe was from 3 to 10 days late and, at the end of the season, some fields full of ragweed, almost flowering, dried without production of any pollen grains.

Ragweed pollen is a real public health problem increasing every year and invading more and more geographical areas.

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### Modelling of Ambrosia Airborne Pollen concentrations over Europe

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Because of the health effects of the invasive weed Ambrosia artemisiifolia L. (Common Ragweed), it is necessary to develop modelling tools to be able to forecast ambrosia air pollen concentrations and to inform allergic people when pollen threshold values are exceeded. This study is realised within the framework of the ATOPICA project (https://www.atopica.eu/) which is designed to provide first steps in tools and estimations of the fate of allergies in Europe due to changes in climate, land use and air quality. To calculate and predict airborne concentrations of ambrosia pollen, a chain of models has been built. The PMP phenological modelling platform is used to construct a model fitted with observed start and end dates of pollen season. The phenological model takes into account the daily air temperature, the daily soil moisture and photoperiod. The inter-annual pollen production is modelled using the vegetation

model ORCHIDEE. The release module based on Efsthatiou formulation (2011) (1) is implemented in the chemical transport model CHIMERE. Airborne pollens follow processes similar to air quality pollutants in CHIMERE with some adaptations. The detailed methodology, formulations and input data will be presented. A set of simulations has been performed to simulate airborne concentrations of pollens over long time periods on a large European domain. Hindcast simulations (2000 -2012) driven by ERA-Interim re-analyses are designed to best simulate past periods airborne pollens. The modelled pollen concentrations are calibrated with observations and validated against additional observations. Then, 20-year long historical simulations (1986 - 2005) are carried out using calibrated ambrosia density distribution and climate modeldriven weather in order to serve as a control simulation for future scenarios. By

comparison with multi-annual observed daily pollen counts we have shown that the model captures well the spatial distribution with correlation equal to 0.77, but the daily variability of pollen counts remains to be improved with correlations varying between 0.1 and 0.75. The model chain captures reasonably well the inter-annual variability of pollen yearly mean concentrations, correlations are positive for about 80% of sites. The main uncertainty in ambrosia pollen modelling is linked to the uncertainty in the plant density distribution. Preliminary results of the impact of environmental changes on pollen concentrations in the future will also be shown.

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### RAGWEED POLLEN FORECASTS BASED ON THE NUMERICAL DISPERSION MODEL COSMO-ART: CONCEPT, PROCESSES AND PERFORMANCE

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In Europe, more than 15% of the population suffer from pollinosis. Pollen forecasts are a helpful tool both for physicians and allergy sufferers. Traditionally, pollen forecasts are mostly linked to pollen observation sites. However, there is a strong need for regionally and temporally detailed pollen forecasts. Numerical pollen dispersion models such as COSMO-ART (COnsortium for SMall-scale Modeling - Aerosols and Reactive Trace gases) can provide the required temporal and spatial resolutions. The COSMO model is used in operational weather forecasts in a number of European weather services such as those of Germany, Italy and Switzerland. The pollen module of ART describes the pollen emission, dispersion and sedimentation processes. During the pollen season, MeteoSwiss operationally calculates hourly birch, grass and ragweed pollen concentrations with daily updates of the 72h-forecasts. The model domain covers Central and Southern Europe with a spatial resolution of 7 km. The distribution map of ragweed is based on plant inventories and pollen data. To describe the course of the pollen season, the climatological start and end dates in combination with Gaussian functions were used. The parameterization of the pollen emission is superposed on these factors and depends strongly on the local meteorological conditions. Both dry deposition processes and washout by precipitation is taken into consideration. The performance of COSMO-ART during the 2013 ragweed pollen season is presented. Time series comparison of measured and modeled concentrations shows mostly good agreement for French, Swiss and Austrian stations. The analyses include both daily as well as bihourly resolutions. Good agreement at stations remote to the main pollen sources highlights the ability of COSMO-ART to model the transport processes of pollen. However, the analysis of discrepancies indicate that the ragweed distribution is a key input to COSMO-ART. In addition, the parameterization of the emission processes is of paramount importance for the performance of the ragweed forecasts. To sum up, COSMO-ART is capable to provide spatially and temporally highly resolved pollen concentrations of good quality. Traditional pollen forecasts are largely station-oriented. The pollen concentration maps and time series of COS-MO-ART fill the gap between the stations and thus represent pollen information hitherto not available.

### Seasonal variations in the amount of airborne ragweed pollen in Milan in relation to environmental factors

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During the summer of 2013, the exotic oligophagous leaf beetle Ophraella commu-LeSage, 1986 (Coleoptera: па Chrysomelidae) was observed feeding on Ambrosia artemisiifolia L (common ragweed) plants in the northwest of the Province of Milan (NW Milan) in Northern Italy. At the same time, it was noticed that routine pollen-monitoring stations situated in three towns in the same area recorded less Ambrosia pollen than in the previous decade. A study has been conducted to determine whether this observed decrease in airborne Ambrosia pollen concentrations can be explained by environmental factors such as meteorology, or whether there is evidence to support the hypothesis that the decrease was related to the presence of large numbers of O. communa in the area. Analyses have shown that the observed decrease in the

amount of Ambrosia pollen recorded annually during August and September (Annual Ragweed Pollen - ARP) in NW Milan was significant (slope=-211.023, p=0.037). This study has also shown that the amount of airborne Ambrosia pollen is influenced by conditions before and during the main flowering period of Ambrosia. Rainfall during August seems to be particularly important (r=0.538, *p*=0.047), and appears to be related to the positive effect it has on water availability for pollen production, rather than the negative influence it has on the release and dispersal of pollen. The amount of precipitation recorded in 2013 was one of the lowest in the 2000-2013 dataset, and as a result the ARP in 2013 would be expected to be of a low magnitude. However, the regression models were, in generally, not able to explain the extreme decrease in airborne

Ambrosia pollen concentrations seen in 2013. The results of this study do not support the hypothesis that the observed decrease in airborne Ambrosia pollen was solely related to the presence of large numbers of O. communa in the area. However, the results of regression analysis suggest that the drastic decrease in ARP in 2013 cannot be explained by meteorology alone. The degree by which the infestation of the beetle influenced airborne Ambrosia pollen concentrations therefore remains unknown. More study of the longterm effects of O.communa on concentrations of airborne Ambrosia pollen is required. In addition, other factors that are expected to influence ragweed pollen counts should also be considered, such as changes in land use and management practices, implemented by local regulation since 1999.

# Evolution of ragweed pollination in France

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Native from North America, Ambrosia artemisiifolia was accidentally introduced in France in 1863, in a field of Allier, with seeds of red clover. Other introductions followed throughout the twentieth century in many places, especially during the First World War. Then ragweed spread to the roadsides and banks of rivers (Rhône, Loire ...). It is only around 1970 that its role was highlighted in pollinosis cases in Lyon (1). Currently, it is estimated that 6 to 12% of the French population is allergic to ragweed pollen, especially in Rhône-Alpes region where 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 like in Rhône-Alpes. With records covering more than 25 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... Pollen exposure measurements are based on data from Hirst-type spore traps and correspond to daily pollen concentration. The data are recorded on a 2hourly time step which allows to determine the circadian rhythm of the plant and to evaluate if the pollens are of local origin or transported over longer distance by the wind. In Lyon, one of the hot spot for the ragweed, the start date of the pollination tends to be earlier: the tendency shows that the first pollens of ragweed arrive now one week earlier than 25 years ago. Over the same period, the annual pollen index increased in most of the French stations in areas infested by ragweed. 2013 was however an exception to this rule due to a really late season. In some stations, circadian rhythm allows to see the evolution of the presence of the plant (2) in a same region: in Auvergne, the north of the region is already infested (pollens in the morning) while the south of the region is not yet infested (a few pollens in the afternoon). And now, ragweed pollens are also recorded on some places of the west of the country while it was not the case before. Twenty-five years of pollen data history offers an interesting overview of the evolution of ragweed pollination in France: season begins earlier, the amounts of pollen are increasing in most of the stations and it seems that more and more areas are infested and, consequently, more and more people become sensitive and symptomatic.

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### Update on Airborne ragweed pollen monitored in Istanbul

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The 15 million metropolis of Istanbul has comparably long been spared from ragweed. In 2007, Ambrosia pollens were observed for the first time in Buyukcekmece, in the western suburbs of the city. Due to the lack of reported local plant populations and low concentrations, long range transport was argued combined with a local source (1). In this paper, data sampled from the same trap location (TRISFU in EAN) during 2013 are presented. This paper also describes the changes in airborne concentrations of this highly invasive plant over the course of the last six years. Methodology: Pollen collected in 2007 (01.08 - 30.09) and 2013 (10.07 and 30.09) with a Hirst-Type trap and counted on three horizontal sweeps was analyzed. A Missing Data Analysis was performed with SPSS 15.0 for the ragweed season 2007. Their statistical difference was tested with a Two sample t-Test. Results and Discussion: For the data collected in 2007, a missing data treatment was necessary because 32.8% of the collected days were missing. Regression Estimated Statistics (REG) yielded a satisfactory outcome. The Seasonal Pollen Index (SPI) was 91 in 2007 and 454 in 2013, which indicates a fivefold increase of ragweed pollen over the last six years. The concentration level in western Istanbul during 2013 (which could be representative for the Turkish region Thrace) falls in the range of the annual PI of 200-500 reported for the Thessaloniki area in Greece (2). The mean daily concentration during the season increased from 1.49 grains/m<sup>3</sup> in 2007 to 7.44 grains/m<sup>3</sup> in 2013. In 2013, the concentration on the peak day (24.08.2013) was 51 grains/m<sup>3</sup>, while it was 20 grains/m<sup>3</sup> in 2007 (1). Between 11.08.2013-09.09.2013, fourteen days had concentrations greater than 10 grains/m<sup>3</sup>. The difference in airborne concentrations between 2007 and 2013 was highly significant (p<0.0001). Con*clusion:* In light of these results and the assumption that a Hirst-type trap represents mostly the flora in the range of 30 km from the receptor site (3), there is a strong indication of a local ragweed population. With regards to the implications for allergy sufferers, a forthcoming clinical panel study will shed light on the level of ragweed sensitization among the population of Istanbul.

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# Session III



### Clinical aspects and genetic/environmental factors influencing ragweed pollen allergenicity

Chair: Claudio Ortolani, Sandra Citterio

### Weed pollen Allergy in Austria: crossand co-sensitization against ragweed and mugwort pollen

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In Austria, about 20% of pollen-allergic individuals suffer from weed pollen allergy. Before 1990, mugwort (Artemisia vulgaris) pollen was the main cause of seasonal allergy in late summer. In the 1990's ragweed (Ambrosia artemisifolia) plants have been brought in from Hungary, and since 2005 the number of reports on ragweed plants tremendously increased (1). With the presence of these plants, the ragweed pollen load and the prevalence of ragweed pollen allergy increased (2). To fight the threat of uncontrolled ragweed expansion and allergic sensitization, measures were initiated during the last 10 years and financed by national and federal agencies. Projects were started to investigate ragweed biology, geography and eradication strategies. Agricultural products potentially containing ragweed seeds became controlled, and the general awareness was increased by information campaigns. Studies revealed that from 1997 and 2007 neither the total pollen counts nor the prevalence of ragweed sensitization (i.e. presence of ragweedspecific serum IgE) increased to the extent that the plant reports had suggested. During the last 6 years the total ragweed pollen load remained relatively constant. Interestingly, the pollen season also became shorter. Thus, the relevance of ragweed pollen allergy in Austria for the moment may be rated "intermediate". A

major problem in the assessment of ragweed allergy arises from cross-reactions with mugwort pollen allergens. As the ragweed pollen season overlaps with the mugwort pollen season, diagnosis using pollen extracts is especially difficult in areas with exposure to both pollen species. In 2004, relevant mugwort and ragweedpollen allergens became available by molecular cloning and were useful tools to address the question of immunologic cross- versus co-sensitization in patients allergic to both allergen sources at the IgE- and T cell level. Mugwort- and ragweed pollen contain one major allergen each, Art v 1 and Amb a 1 respectively. We found that Art v 1 has a uniform T cell response to one single peptide within this protein. Art v 1-specific T cells generally did not cross-react with the corresponding peptide from the homolog protein in ragweed pollen (Amb a 4). In contrast, for Amb a 1 we found multiple T cell activating regions that can be widely presented in the population by diverse HLA-class II molecules. Amb a 1-induced T cells reacted weakly with Art v 6, the Amb a 1-homolog in mugwort pollen, whereas Art v 6-induced T cells responded stronger to Amb a 1. Art v 6 contains only a few T cell epitopes which cross-react with Amb a 1. At the humoral level, cross-inhibition experiments indicated that in patients sensitized to both

weeds, IgE is largely cross-reactive. However, Amb a 1 possesses more IgE-epitopes or alternatively binds IgE with higher affinity than Art v 6. Thus, Amb a 1 can elicit more diverse allergen-specific IgE- and T cell responses and dominates the cross-reactivity with its homolog. Nevertheless, in regions with high mugwort pollen exposure Art v 6 can sensitize by itself. Thus, it cannot be excluded that in Vienna Art v 6-sensitization might have facilitated sensitization to Amb a 1 by epitope cross-recognition of B and T cells at the time ragweed began to invade Austria. Together, the major allergens Art v 1 and Amb a 1 do not essentially contribute to the cross-reactivity of the two weed pollen species. By assessing IgE against the major allergens Art v 1 and Amb a 1, cross- vs. co-sensitization to both weeds can be discriminated and has implications for specific immunotherapy.

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## Towards the isolation of floral and reproduction related genes of the common ragweed (*Ambrosia Artemisiifolia* L.)

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The common ragweed (Ambrosia artemisiifolia L.) is the most widespread weed in Hungary and some other parts of Europe. In spite of strict legal regulations and intensive chemical and mechanical contrasting measures this invasive species is spreading (Kazinczi et al. 2009), and represents an example of aggressive invasion all over the world. Besides its economic impact on crop yield, it represents a major health problem because of its strongly allergenic pollen. While some members of the Amb a I pollen allergen gene family have already been isolated (Rafnar et al. 1991, Hiller et al. 1998, Ivanciuc et al. 2009a,b, Oezguen et al 2009), little is known about the floral genes of Ambrosia. Ambrosia artemisiifolia is a monoecious dicotyledonous bushy weed, with dense male flowers in terminal racemes on the branches, while female flowers are present in the axils of upper leaves. An average plant is able to produce 6 thousand million pollen grains, and thousands of seeds (Kazinczi et al. 2009). Considering that just in Hungary ragweed covers more than 5% of arable land (Novák et al. 2009)

and that it is acclimatizing and spreading rapidly, the allergy problem may turn more severe and it is unlikely that the plant will be controlled effectively with the presently applied methods. We believe that more detailed understanding of the plant at the molecular genetic level may help the elaboration of new protection technologies. To this end in the present study we aim to isolate genes which play a determining role in the development and identity of floral organs and in reproduction biology of ragweed. In this study, samples for analysis were taken from six plants which grew under natural conditions in our experimental garden. From each plant male and female flowers, as well as young leaves were biweekly sampled throughout the flowering season. For male flowers seven, while for female flowers nine developmental stages were determined. For each stage male and female flowers were collected separately, and at the same time young leaves were sampled,

From the samples RNA was extracted by RNAzol (MRC, USA), column purified

and treated with DNase. cDNA synthesis and subtractive hybridization was according to the recommendation of the PCR-Select cDNA subtraction kit (Clontech, USA). The subtractions were done in three different ways to get sequence collections which represent: 1) floral genes, 2) genes expressed only in male flowers, and 3) genes expressed only in female flowers. Since the subtracted sequences are short (<300 bp) regions of genes, for the isolation of the complete genes a male flower and a female flower cDNA library was constructed using the same RNA pools which have been used for subtraction. For cDNA library construction the In-Fusion Smarter Directional cDNA Library Construction Kit (Clontech, USA) was used. For the functional analysis of isolated genes we elaborated a genetic transformation and regeneration system of the common ragweed to silence genes with RNAi constructions and to study the changes on the regenerated plants. Our actual results on the analysis of floral and reproduction related genes will be presented here.

## Concomitant sensitization to ragweed and mugwort pollen: a re-evaluation of allergen cross-reactivity in milan area

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In the area of Milan a concomitant sensitization to ragweed and mugwort is common to many patients and given the overlapping flowering periods of the two plant, so far it has not been possible to diagnose the primary allergen sensitizing source and decide the right immunotherapy. The objective of this study was to elucidate whether double-sensitized patients are co-sensitized or crosssensitized and in the last case, to define the primary sensitizer. Patients underwent skin tests with ragweed and mugwort extracts. Sera were then collected and analysed for their IgE reactivity to rArt v 1 and Amb a 1 by ImmunoCAP assay and to Amb a 1, Art v 6, Art v 1 isoforms through a proteomic approach. Results showed that sera from patients monosensitized to ragweed contain a great variety of IgE directed against epitopes specific of all Amb a 1 isoforms. On the contrary, IgE antibodies of all the examined double-sensitized patients who were shown to be primarily sensitized to mugwort, reacted with mugwort Art v 1 and Art v 6 allergens and crossreacted with few isoforms of Amb a 1. Finally, sera of double-sensitized patients who resulted primarily sensitized to ragweed contained IgE reacting toall Amb a 1 isoforms and in part cross-reacting to Art v 6. No really co-sensitized patient was found. This study shows that Art v 6 plays an important role in mugwort allergy and that the cross-reactivity between Art v 6 and Amb a 1 is frequent in Milan area, clinically relevant, and bidirectional.

## Testing allergenicity of *Ambrosia Artemisiifolia* pollen in mice

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Ambrosia artemisiifolia is a highly invasive plant with pollen that causes severe allergy. Studies indicate that pollens respond biochemically to environmental changes such as air quality, extreme weather, and ground pollution, but a correlation between these changes and human health remains unknown. We sought to establish an experimental model of allergic disease to test the effect of environmental change on Ambrosia pollen. We administered commercially available pollen (Allergon, Sweden) and pollen collected from areas

near Vienna, Austria intranasally to BALB/c mice over a period of either 10 or 21 days. We then evaluated allergic responses locally in the lungs and systemically. Groups of mice receiving intranasal Ambrosia pollen had severe allergic lung inflammation, goblet cell hyperplasia and hypersecretion in 10 days, but no serum Ambrosia-specific antibodies were detected. After 21 days, mice developed severe allergic lung disease with concomitant high titres of serum Ambrosia-specific antibodies. Doses of Ambrosia were titrated to generate a dose-response curve. Commercially available processed Ambrosia pollen was compared with collected populations of pollens. The pollens were found to induce different responses in the mice, which lead to varying severity of allergic disease. Correlations between the environmental conditions and disease severity suggest and that environmental changes impact the allergenicity of pollen. This experimental mouse model provides novel opportunities to study pollen-induced allergic disease.

## BIOLOGY AND ALLERGENICITY OF AMBROSIA SPP. (RAGWEED) IN ISRAEL

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In the past three decades' Ambrosia spp. invaded vast areas in Israel. Five species of Ambrosia were identified over the years: (1) the annual native species, A. maritima typical of the Mediterranean basin, was last observed in 1981and declared an endangered species; (2) A. confertiflora, a perennial herb identified in Israel in 1990 -avery aggressive plant, forming large stands reaching a height of 3 m' with a very dense underground system of rhizomes and roots, and spreads very quickly; (3) A. tenuifolia, perennial, documented in Israel since 1984, grows only in isolated areas; (4) A. artemisiifolia, appears sometimes in Israel but is not established, perhaps due to the dry summer; (5) A. trifida, annual weed, last observed in Israel at 2009. Invasion paths of Ambrosia in Israel were by contaminated imported grains, spread along river banks, roads and railway tracks and by

themovement of contaminated soil. The species differ in their life cycle (annual or perennial), flowering season, flower structure and propagule shape. Particularly noticeable is the structure of A. confertiflora's achene that through hooked prickles can be caught in objects moving in the habitat (epizoochorous transport) thus contributing to the species dissemination. Germination and reproduction experiments conducted under controlled environment show that seed germination rate exceeding 50% for both perennial species (N>1000). Seeds germinate in light and emerge from upper soil level. Single plant of A. confertiflora creates an average of 45 sprouts within four months whereas A. tenuifolia forms an average of 245 new sprouts during the same period of time. Hence, attempts to control Ambrosia by mowing are inefficient. Our preliminary results show that

the number of A. confertiflora plants sprouting from rhizomes is increasing after removal of the plant apex indicating apex dominance, while A. tenuifoliarhizomes number does not increase. The Ambrosia is wind pollinated, producing large quantities of highly allergenic pollen. Allergy skin tests experiments were performed in two hospitals in Israel, using pollen extracts prepared from pollen collected in the field of the perennial species and commercial extracts for A. artemisiifolia. So far we tested 147 volunteers. Results shows that 20% of the tested people reacted at least to one of the species and that A. confertiflora is much more allergenic (15%) than A. tenuifolia and (5%) A. artemisiifolia (5%). The rapid growth and spread of Ambrosia spp. in Israel, is a threat to agricultural crops and public health.

### Symptom load index in European countries infested by ragweed: comparison of the situation of hay fever sufferers in Austria, Germany, Serbia and France

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The area of ragweed plant growth is not congruent with the area of ragweed pollen deposition. Moreover, duration and intensity of ragweed pollen seasons differ significantly across and within different countries and between biogeographical regions, thus suggesting also divergent levels of tolerance to airborne ragweed pollen loads. Spatial variation of health impact of airborne ragweed pollen was studied by using data from symptomatic people in France, the Pannonian Plain (i.e. Eastern Austria and Serbia) and Germany from the years 2009 until 2013. Data was collected by the Patients Hayfever Diary (PHD, www.pollendiary.com). PHD is a free online and app supported diary for pollen allergy sufferers amassing records for location of stay, overall feeling, organ-specific symptoms, symptom severity, and medicine intake. Acquired symptom scores together with corresponding pollen counts were used to calculate local symptom load indices (SLI) and hence estimate the allergenic power of ragweed pollen in various regions. It is evident that people in Serbia suffer most from ragweed pollen; significantly more than people in other countries under study here. France shows the second highest values of SLI, followed by Germany and Austria. It should be noted that although ragweed loads are lower in Germany, the German residents suffer from equally high daily ragweed pollen levels more than users in Austria. The results confirm spatial heterogeneity in allergological importance of ragweed pollen within regions under investigation. Burdens in core areas (i.e. Pannonian Plain and Rhône Valley) correspond to annually recorded pollen loads. At this stage problematical differences concerning high load - moderate response and low load - moderate response (Germany -Austria) are fairly explained by the SLI, but full clarification desires PHD exploitation also for other parts of Europe, where ragweed pollen is commonly recorded in the atmosphere (i.e. Hungary, Italy, Croatia, Slovenia, Slovakia, Czech Republic, Ukraine, Russia, Belarus, Poland and Romania).

## Risk for *de novo* sensitization to ragweed in Croatian children

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#### Introduction

FP7 project ATOPICA (*Atopic diseases in changing climate, land use and air qual-ity*) supported by EU Grant agreement NO: CP 282687 explores the combined pan-European impact of changes in climate, land use and air pollution on allergen pollen-induced diseases with an accent on atopy due to ragweed sensitization. Sensitization to ragweed pollen correlates with levels of airborne pollen concentration in environment, but can be enhanced by other environmental factors such as air pollution.

#### Material/Methods

Cohort of 3590 children, aged 4-10 years, was recruited from 3 regions of Croatia differing in airborne pollen concentrations (Slavonia, Zagreb and surrounding end Dalmatia). Each participant underwent skin prick test (SPT) to

the standard set of aeroallergens. For each region, pollen concentrations and air quality data were gathered from authorized institutions.

#### Results

A total of 369 children were sensitized to ragweed pollen. Prevalence of ragweed sensitization was 14.84 % in Zagreb area, 14.26 % in Slavonia and 1.52 % in Dalmatia. Comparing the highest pollen concentrations during ragweed pollinating period among 3 regions, Dalmatia has the lowest concentration of ragweed pollen of 30-40 grains/m3, while Zagreb measures 250-300 grains/m<sup>3</sup> and Slavonia 700-1000 grains/m3 per 24 hours. Analysis of sensitization in two age groups (4-6 and 7-10 years) reveals higher prevalence of ragweed, birch and D. pteronyssinus sensitization as well as double sensitization (birch and ragweed) in older age group for all 3 regions. Sensitization to above allergens was more prevalent in male participants. After two pollen seasons of followup, de novo sensitized children were recorded: 2.88% in Zagreb area, 2,73% in Slavonia and 0,00% in Dalmatia. Analysis of de novo sensitization in two age groups shows more de novo sensitized children in older age group for all regions. Pollen diary data (data on overall simptom score, eyes, nose, lungs symptoms/problems, usage of medicine and daily activity) also showed correlation between ragweed pollen concentrations and severity of symptoms.

#### Conclusions

Region of Slavonia measures highest ragweed pollen concentrations and also most de novo sensitized children, which leads to assumption that de novo sensitization rates are primary correlated to exposure levels to ragweed pollen.

## Session IV



## RAGWEED POLLEN ALLERGY: VETERINARY ASPECTS

Chair: Alessandro Grittini, Roberta Sacchi

## RAGWEED POLLEN ALLERGY: VETERINARY ASPECTS

#### N. FURIANI

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Hypersensitivity disorders are common in domestic animals. In particular, atopic diseases are frequently diagnosed in dogs, cats and to a lesser extent in horses. The major clinical manifestations can be atopic dermatitis (AD), recurrent urticaria (RU), rhino-conjunctivitis and respiratory disease. The first one is by far the most common in dogs. Atopic dogs can manifest conjunctivitis (more than 50% of cases) and quite rarely rhinitis or reverse sneezing. Although canine models of allergic rhinitis and asthma, including ragweed sensitized Beagles, exist, spontaneous allergic respiratory diseases are not expected to occur in dogs. Cats can manifest conjunctivitis and sneezing associated with cutaneous signs or can develop allergic respiratory syndromes, such as feline asthma and feline allergic bronchitis. Atopic horses can manifest cutaneous diseases, AD and RU rarely associated with rhino-conjunctivitis, or chronic lower airway diseases, the recurrent airway obstruction (RAO) and the inflammatory airway disease (IAD), or a combination of skin and respiratory signs. The specific role of ragweed pollen in triggering atopic diseases has been only marginally investigated in veterinary medicine. In dogs, the most important route of allergen exposure seems to be the epicutaneous route. In a canine model of atopic Beagles an early exposure to probiotics (Lactobacillus rhamnosus strain GG) do not protect from the epicutaneous ragweed sensitization but leads to a less severe clinical manifestations after allergen challenge (1). According to a recent

study, clinical manifestations in dogs spontaneously sensitized to short ragweed (Ambrosia artemisiifolia) do not differ from those usually reported in atopic dogs, 79% of that dogs were indoor living dogs and 66% showed unseasonal symptoms (2). The prevalence of ragweed sensitization in atopic dogs vary greatly from different countries and seems to correlate quite well with the plant's geographical distribution. Among studies from North America, where ragweed is native, two testing a "mixed ragweed" allergen reported a high prevalence of 35% and 59% and a third one using individual allergens of short ragweed, western ragweed (A. psilostachya) and prairie sage (A. tridentata) reported a prevalence of 34%, 37% and 35%, respectively (3,4). A study evaluating 1000 intradermal skin tests (IDT) from atopic dogs living in Southeastern Australia showed a prevalence of 13,4% for short ragweed and of 10,1% for western ragweed (5). Among European studies, short ragweed was the most usually tested. In two studies performed during the 1990s the prevalence reported was quite low, varying from 7,7% (Greece) to 9,2% (France) (3). Instead, recently published results from Northern Italy, especially the province of Milan, and Serbia, regions severely affected by ragweed colonization, revealed that A. artemisiifolia was the second most common allergen involved in canine AD with a prevalence of 21,6% and 66%, respectively (6, 2). Interestingly in a 2006 study from Hungary, another country where Ambrosia spp. is highly diffused,

atopic dogs reacted more frequently to mugwort (Artemisia vulgaris) (7,8%) than short ragweed (<4%) (8). As human patients, many atopic dogs sensitized to ragweed also react to mugwort, studies investigating if it is due to cross-reactivity or co-sensitization are lacking. Ognjenovic et al. have recently identified the major allergens of A. artemisiifolia involved in canine AD using an immunoproteomic approach (9). Similar to humans, these were represented by all five isoallergens of the Amb a 1 group. Some canine sera also react to the fragment of pollen allergen Amb a 1.0201, whereas none of them recognized lower mass allergens such as the fragment of Amb a 1 and Amb a 8 variants. The future use of these purified antigens in allergic testing as well as in allergen-specific immunotherapy could greatly help clinicians and investigators. To date, no data are available regarding the clinical response of atopic dogs to ragweed-specific immunotherapy and it would be difficult to perform studies with this purpose because cases of monosensitization are very rare (less than 0,5%, data unpublished). No substantial data are available about ragweed sensitization in cats where the use and the interpretation of allergic tests, both IDT and serum IgE testing, is still controversial. In equine medicine there are few studies evaluating positive reactions to aeroallergens in atopic horses by means of IDT or serum testing. In those reporting data about ragweed sensitization usually a "mixed ragweed" allergen was used. In two studies from North America horses affected



by AD, RU and RAO/IAD showed a prevalence of positive reactions of 57% (4/7), 60% (6/10) and 12,5% (2/16), respectively (10, 11). In an European study from Austria 43 IDT from atopic horses were evaluated but no positive reactions to ragweeds were recorded (12). Despite the limited data, ragweed doesn't seem to play an important role in inducing atopic diseases in horses.

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## RAGWEED POLLEN ALLERGY: PUBLIC HEALTH ISSUES

Chair: Maira Bonini, Michel Thibaudon

## RAGWEED POLLEN ALLERGY AND PUBLIC HEALTH ISSUES: INSIGHTS FROM RECENT RESEARCH

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In the province of Québec (Canada), wherever ragweed (Ambrosia artemisiifolia) is abundant, close to 18% of the population suffers from allergic rhinitis linked to pollen dispersal. In the context of climate change and global warming, the dispersal of common ragweed, associated to a longer pollen season (1), further increases public health concerns. Therefore, there is a need to provide conclusive data on the impact of the control of ragweed on health and on quality of life. These will support decisions on interventions to prioritize. Hence, an epidemiological study including a 4-year followup of 400 allergic volunteers was previously carried out in the Montérégie region (province of Quebec) between 2007 and 2010. Initial results showed that, when applied twic eat specific periods (mid-July and mid-August), the concerted control of ragweed involving a limited number of key actors (i.e., the municipality, the ministry of transport and managers of large lands) induced a significant reduction in pollen concentrations. It also decreased the symptoms of allergic

rhinitis and improved the quality of life. Following these results, further research was conducted in order to include a spatial analysis which attempted at establishing the relationship between the land use, the spatial dispersion of ragweed and the pollen concentrations. Its main purpose was to attribute an individual level of pollen exposure to each allergic participant enrolled in the epidemiologic study in relation with health symptoms. This approach is innovative and promising in the ragweed field, however additional validations are needed to provide reliable.

#### Conclusions

Furthermore, a cost-utility analysis was performed in order to assess the economic impact of a concerted intervention for the control of ragweed. Clinical data from the epidemiological study were converted into utility values using the method proposed by Keiding and Jorgenson (2). The costs of all components of the intervention totalized CDN\$60,603. The con-

certed intervention was associated to a gain on health of 10.15 Quality Adjusted Life Years (QALYs). The incremental cost-utility ratio derived from these parameters, CDN\$5,744/QALY, demonstrated that concerted management of ragweed is highly cost-effective. Therefore, the findings emerging from this research help to identify major components (e.g., municipal regulation, localisation of priority areas for intervention or choice of the optimal intervention period related to the biology of the plant) of importance to maximize the efficacy of the control of ragweed. Overall, these recent analyses reinforce advocacy in favor of a concerted management for the control of ragweed.

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## Public and private roles in management of ambrosia

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The protection of plant health and biodiversity is an important issue in many countries over the world. The protection of plant health covers a wide range of areas such as the 'legislative and regulatory measures [..] that include eradication and containment campaigns, surveys, risk assessments, [..] certification and marketing schemes' (Ebbels, 2003). Plant health issues play an important role in the WTO negotiations as individual countries are allowed to introduce trade barriers in order to protect their domestic agriculture against the introduction of plant pests. Although the probability of a catastrophe due to introduction and spread of new plant pests is generally very low, the economic and environmental impacts can be extremely large if a pest does occur. This holds in particular also for the consequences of common Ragweed as the presence of this species has consequences for agriculture (yield losses, extra costs for management), trade in plant products (as seeds may be spread through e.g. cereals) and human health. The consequences for human health may be particularly important, as not only the well-being of people living in infested areas is at stake, but also revenues from tourism and recreation. National plant health services have been assigned the task of safeguarding the national plant health status. Under EU legislation, national plant health services are required to inhibit the spread and introduction of harmful organisms (European Commission, 2000). In EU directive 2000/29 and its Annexes (European

Commission, 2000). In addition, in the case of Ambrosia pollen also human health authorities are involved in monitoring and managing exposure to Ambrosia pollen. This paper analyses the institutional framework associated with management of the Ambrosia. First, a conceptual model of management of Ambrosia depicting the relations between various stakeholders involved in management of Ambrosia such as farmers, citizens, plant health authorities and human health authorities. Second, this paper discusses the public and private good characteristics of Ambrosia and the consequences for the choice of the regulatory and institutional arrangements for managing Ambrosia. The roles of the public and private sectors in managing Ambrosia are clarified.

## Results of the EU project halt ambrosia

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HALT AMBROSIA is a research project funded by the European Commission, DG Environment (07.0322/2010/58340/SUB/B2) running from Feb. 2011 till Feb. 2014.

The overall aim of the project is to contribute to the reduction of the prevalence of the invasive alien plant *Ambrosia artemisiifolia* in European countries in order to reduce the burden on public health, agriculture and biodiversity.

Partners in Germany, Hungary, Slovenia, Austria and Denmark contributed to research in the following fields:

- Biological fundamentals.
- Non-chemical and integrated control strategies.
- Best use of herbicides.
- Impact on non-target species and biodiversity.
- On the basis of these studies we developed recommendation for best the ways to
- prevent the spread of Ambrosia to new habitats;
- reduce the risks of establishment of

new populations;

- control existing populations;
- dispose of plant material that may contain viable seeds.

With these recommendations we propose elements of strategies against common ragweed that take into specific scenarios into account, e.g., the level of infestation of a given country or region, legal opportunities to use herbicides, etc.

The out come of the project will be used by the EU Commission as a pilot study for the possibilities to control invasive species.

## Health impact of exposure to ragweed POLLEN IN FRANCE

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Ecological studies show a temporal association between the presence of ragweed, the consumption of anti-allergy drugs and hospitalizations for asthma. However, there are few panel studies, showing the relationship between the concentration of ragweed pollen in the air and the intensity of symptoms among patients with hay fever on a significant number of subjects.

Two studies were carried out in France, based on questionnaires filled in by patients themselves:

- for the first study POLPAT (1), 37 patients suffering from hay fever during ragweed pollination period, completed a daily questionnaire scored from 0 to 3 on the intensity of nasal and ocular symptoms in July, August and September 2009 and 2010 (2,960 daily data). RNSA provided the rate of pollen, Météo-France meteorological parameters, ADEME PM<sub>10</sub>, NO<sub>2</sub>, and  $O_3$ .
- for the second study, information on the user's location (biogeographical

regions), pollen data were also provided by RNSA pollen traps. Health impact was assessed based on the entries of Patient's Hay fever Diary (2) (PHD) users, who fill in their overall feeling, organ specific symptoms and medication use on a web-based platform. All data were included to calculate a symptom score.

In Polpat study, the rate of patients with various types of symptoms increased in a linear, significant way with exposure to ragweed. For an increase of 10 grains/m<sup>3</sup>, odd-ratio of ocular symptoms was 1.324 in 2009 and 1.049 in 2010. For nasal symptoms, the effect of ragweed pollen was more important on weekends (OR = 1.426 in 2009) than on weekdays (OR = 1.184).

The PHD results show that there is a dose-response relationship between the ragweed pollen exposure and the symptom severity: the onset threshold of symptoms is about four pollens/m<sup>3</sup> of air. Biogeographical variation in symptom severity was observed. In conclusion,

clinical response among sensitized patients shows a linear and significant relationship, without any threshold, between the increase in ragweed pollen and nasal and ocular symptoms in POLPAT study. PHD study gives insight into the relation between health condition and pollen exposure, and confirm that only few ragweed pollen grains are enough to trigger symptoms.

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### Assessment of the healthcare costs related to ragweed in Rhone-Alpes region

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Rhône-Alpes region is the French region which is the most affected by ragweed pollen pollination. The aim of this study is to estimate the healthcare cost of this allergy, thanks to different indicators from the database of repayment health insurance, like purchase of anti-allergic drugs, doctor consultation or short work stoppage. These indicators were selected comparing similarities between ragweed pollen peaks and date of drug purchase for instance.

The analysis concerns policyholders from 6 to 64 of the general social security system who had at least one repayment for anti-allergic drugs during the pollination period of ragweeds and on the other care consumptions like allergens, consultations, search for specific IgE, desensitization, skin tests, treatment of asthma, and short work stoppage. In parallel, data about the ragweed pollination are provided by the RNSA in order to analyze and corroborate mapping of pollen peaks (exposure) with care consumptions. Two hypotheses were considered based on the drug consumption: one named "low hypothesis" corresponding to allergy sufferers who are certainly sensitive to ragweed pollens (buying antihistaminic in August and September) and one named "high hypothesis" corresponding to the same sufferers plus the ones who could be sensitive (buying drugs during the same period and also in October and November). In 2012, between 154914 (low hypothesis, +36% in comparison with 2008) and 197870 (high hypothesis, +23% in comparison with 2008) people used cares in relation with allergy to ragweed. Health costs, estimated from repayment of expenditures of ambulatory cares and work stoppages, are evaluated between 11.3 and 15.7 millions of euros. So the cost for each recipient is estimated between 72.9 and 79.3 euros. This study shows a real increase in the number of allergy sufferers to ragweed pollens in Rhône-Alpes region in the last years, maybe more important than the increase of the yearly amounts of ragweed pollens.



#### Fig. 1 - Number of antihistamines boxes for 1000 recipients of the general social security system

### Common ragweed invasion in sweden and human health: preliminary results of an impact assessment

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Common ragweed (Ambrosia artemisiifolia) is a neophyte from North America with a wide ecological tolerance and adaptation capacity that has allowed its rapid spread throughout Europe. The plant has harmful impacts on many sectors, particularly agriculture and human health. In addition to increasing weed management costs for farmers, the pollen of common ragweed is known to cause severe allergic reactions. The allergic reactions associated with an exposure to common ragweed pollen are of two kinds: skin reactions (atopic dermatitis) and illnesses of the respiratory system (allergic rhinitis, hayfever and asthma). Because its pollen is highly allergic, many countries have adopted containment and mitigation measures against common ragweed. Most of the published contributions on the health impacts of common

ragweed focus on countries with well-established populations of common ragweed. In contrast, little is know about the impacts of common ragweed in countries where the common ragweed populations is still in its lag phase. During this phase, a population adapts itself to the habitat's conditions and the population size is small. Our study aims at filling this gap and we find evidence that an invasive species still in its lag phase can have adverse impacts on human health. We chose Sweden as our case study because the ragweed population there is newly established, with the first flowering plant having been documented in the early 1990s. Our identification strategy relies on spatial and temporal variation in common ragweed population and health data. Data for the distribution of common ragweed populations are taken from the Swedish Species Observation System, while health data come from the Swedish Health and Welfare Statistical Database. We study the effect of common ragweed on in-patient care indicators for allergic rhinitis, asthma, and allergic contact dermatitis for the period 1998 to 2011. We develop a reduced form regression model that we estimate with panel data estimation techniques, controlling for much of the unobserved heterogeneity in botanical and health data. To ensure the robustness of our findings, we test and correct for heteroskedasticity and autocorrelation. We find small but significant deleterious effects of ragweed presence on human health. The estimates are robust to a battery of robustness checks. Our estimates show that an invasive species still in its lag phase can yet have adverse impacts on human health.





## Poster

## Chair: Uwe Berger, Victoria Rodinkova

# The assessment of communities ragweed fights by the soil pollen dust flux method

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Since a few years, AFEDA have been asked by some municipalities a method for an assessment of their fight (Déchamp and Méon, 2010, 2011, 2013). The pollen dust flux method, set by Cour in 1974 in a lot of countries, was used in this aim. Filters are the same as those of the AFEDA traps (Cour model) set at about 5 km. In the past, this method had not compared the pollination period of a plant as its aim was only to identify vegetation (often in the desert). Actually, the greater difficulty of the new aim for using this method is to find a not concreted track in a town! For that reason it is difficult to realize many studies.

#### Methods

3 different tracks have been studied each year in 2010, 2011, 2012, 2013 (in Saint-Priest town) and 2 (in Mions town) in 2012 and 2013: at the start of the *Ambrosia (A)* pollination period, week 30, and at the end of the *A* pollination period, week 38. So results of 29 measures are studied. Pollens were collected on the same forbidden to circulation, with the same type of cars, dirty tracks,. No precipitations for three days. This pollen dust flux method measures *A* pollen numbers/dust-gram and/km, their %

versus total pollen. Control of A pollen at the start of the season is necessary to know the principal remain of the last season.

#### Results

At the start of the pollination period, A pollen/dust gram and/km are much lower than at the end, A % are always < 2% of the total flora pollen and are closed to the airborne A pollen. At the end of the pollination period, A pollen/dust-gram and/km, A % have always considerably increased and in most cases this increase is coupled to airborne A pollen trap count. During all these years, at the end of the season, these communities fights have not always produced a decrease of the A ground pollen but they have decreased or they are stable in town centre; rural area is the more difficult one to clean.

#### Discussion

Pollen vegetation distribution is different at any time of the year and varies each year, so the more reliable criterion is pollen number/km, because pollen dust/gram depends on the type of the ground and for instance of its humidity being different in each area, nevertheless A % could be a positive facility to compare these data to the *Ambrosia*-type airborne pollen but it must be controlled by the other criterions. Our study is not totally new but the larger measures (29) versus former measures (9) are a significant novelty.

#### Conclusion

at the start of the A pollination period, A pollen has almost disappeared from the ground and, even if the pollens grains are caught with two different systems, their % seems to be related to the airborne Apollen trap count; at the end of the Apollination period, A pollen can decreased after a local fight. This method can help us to understand the spread of invasive plants or weeds. In the case of anemophilous pollens at the stage of research using the soil pollen flux method, coupled with a pollen trap might help to understand the spread of a plant. The main difficult is in comparable tracks in homogeneous environments: it is easier to have good results in the centre town than in a technologic park and moreover in a rural area. Another difficulty is to find in towns not concreted tracks, so it is possible that we will not can obtain a security index for A pollen that was our first aim.

## Germination of common ragweed seeds – *Ambrosia Artemisiifolia* L.

#### B. KONSTANTINOVIČ, B. KONSTANTINOVIČ, M. POPOV, N. SAMARDŽIČ

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Wide distribution of *Ambrosia artemisiifolia* L. on the territory of AP Vojvodina, as well as its high reproductive potential influenced the formation of seed reserves in the soil, so-called "seed banks". It has been established that ragweed seeds maintain germination capability for over 40 years, and that seed bank formation is an additional factor enabling this species to be one of the most competitive among weed-ruderal plants. Common ragweed (*A. artemisiifolia* L.) is nowadays considered an invasive weed, although it has been present for a long time as a weed species, at first as a resident of ruderal sites, and later occurring as a frequent weed in row crops. Today this weed species is primarily recognised as a plant whose pollen is a strong allergen. This study examined germination rate of the seeds of A. *artemisiifolia* L. in relation to the temperature of their storage. The seeds were collected in October and November 2012 and 2013 from a large number of plants in various microsites within each of the 10 studied locations in the area of AP Vojvodina. The collected seeds were placed in different environmental conditions with different temperature fluctuations. On the basis of these conditions, the seeds were divided into three groups – the seeds vernalised at a temperature of -8°C, the seeds vernalised at 4°C and the seeds stored at room temperature. Germination rate was determined after 9, 14, 21 and 28 days after placing the seeds in a cold chamber. The results indicate that there is a statistical significance in the germination rate, that is, *A. artemisiifolia* L. seeds vernalised at -8 °C showed the highest germination rate in comparison to the seeds vernalised at 4°C and the ones stored at room temperature.

## Impact of campaigns to control common ragweed on the pollen Production

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Common ragweed pollen (Ambrosia artemisiifolia) is known for its allergenicity causing hay fever, rhinoconjunctivitis and asthma (1) to patients living in infested areas. The dispersion of ragweed is anthropogenic, and because it is an invasive plant, a number of control procedures have been implemented whose effectiveness was measured. Measures on pollen emissions were compared between areas with different level of management. In addition to Hirst volumetric pollens traps of the RNSA network (2), it was decided to use passive traps SLT [SIGMA2 Like-Trap (3)] positioned in proximity near a controlled or non-controlled area. SLT samples were collected weekly during the months of August and September.

Two areas with different levels of infestation were selected in France:

Infested areas: 12 SLT traps were set up:
8 traps in the department of Isère, in the town of Estrablin with 2 trapsin non-controlled areas and 6 in wellcontrolled areas.

- 4 traps in the department of Drôme, in the metropolitan area of Valence with one trap in non-controlled area and 3 in well-controlled areas.
- A few infested area in the department of Côte d'Or with four traps located in lightly infested areas (4) of which one in an area without ragweed plants and three in areas with presence of small amounts of ragweed plants.

Ragweed pollen amounts collected from controlled areas and non-controlled areas are different, with a maximum of a few hundred of pollen grains for controlled areas against a few to tens of thousands for the non-treated areas. On the area not really infested, the quantities of pollen are limited to low tens of ragweed pollen grains. Periods of heavy pollination of ragweed pollens are the same for SLT traps and for Hirst pollen traps. Even if the amounts of pollen collected from the well-controlled areas infestations are still high, they are significantly less important than the amounts present on non-controlled areas. The set-up of management practices enable to locally reduce the pollen pressure but only a generalized and integrated control will actually enable to relieve people aware.

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## Health impact of exposure to ragweed pollen in France

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The cultivation of Ambrosia artemisiifolia in Piedmont is documented in the Botanical Garden of the University of Torino in 1772, and this is probably one of the most ancient cultivation of the species in Europe. First data on its naturalization are indicated by specimens of the Herbarium of the University of Torino (HP-TO) dated 1902 and 1910 respectively in Alba and Torino. The present distribution of the species has been recorded in a monitoring campaign carried out from 2006 to 2009 and is related to 10 x 10  $km^2$ quadrats based on the UTM (Universal Transverse Mercator coordinate system) grid (Fig. 1), published in a recent book on invasive alien species in Piedmont (1). Ragweed is actually occurring in 160 quadrats (60%) out of 268. Its frequence is highest in the quadrants of the Plain, in the Provinces of Alessandria, Novara, Vercelli, Biella, Asti and Torino and is lower in the Southern and in the Northern part of the region (Provinces of Cuneo and Verbano-Cusio- Ossola). Moreover, the species is absent in almost all the alpine valleys. In the last years the species increased strongly its distribution, invading not only cities, streets and railways but also agricultural areas, with a very high frequency.

The increase in the distribution and abundance of the species correspond to a notable increase in the airborne pollen concentration which is well represented by pollen counts monitored in the city of Torino from 1984 (3 days with concentrations from 0.5 or 1 pollen/m<sup>3</sup>) to 2013 (62 days with from 0.5 to 54 pollen/ $m^3$ ). The pollen concentration monitored in the center of the city often reached values of 20-30 pollen/m<sup>3</sup> and rarely 50-60 pollen/m<sup>3</sup>. Concentrations are lower than those monitored in other cities of Europe and Northern Italy but the length of ragweed pollen season significatively increased. Mono and polysensitization of the allergic patients monitored at "Azien-

da Ospedaliera dell'Ordine Mauriziano" of Torino increased from 3-4% during the years 1999-2000 to 20-21% in the last 5 years. The rapid spread of this invasive alien species, the consequent increase of its pollen concentration in the city of Torino and of the percentage of allergic patients, highlights that its control becomes mandatory to reduce the impacts of this pest on economy and human health. A regional law on control and eradication of the most important invasive alien species, similar to the recent regional laws approved in the nearby Lombardy and Aosta Valley, is urgently needed.

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## Drought impact on short ragweed pollen density: A field perspective

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ALK-Abelló is in the initial phase of setting up a field trial experiment to investigate the link between Short Ragweed pollen density in the fields and specific allergenicity of the pollen with abiotic stress factors such as late summer heat and drought stress at specific time intervals in the vegetative and reproductive growth season. The aim is to investigate if the allergen protein content of pollen and abiotic stress are linked, as well as other changes in pollen that could potentially affect the allergy (1). The field-based approach will aim to provide a direct link be-

tween drought conditions on the field and aerobiological models for airborne pollen density. The project is enabled by ALK-Abelló's pollen production facility in Illinois, USA (on a latitude similar to Napoli). There detailed field observation of reproductive traits and pollen density combined with high resolution multi-layer soil moisture monitoring combined with direct measurements of plant stress levels on the individual fields and a state of the art determination of the allergy potential of the pollen will provide unique insights into the causes of increased pollen allergenicity. This insight is of particular interest when viewed thought the lens of climate change, where a hotter drier climate could cause a significant increase in both the pollen allergenicity and airborne distribution of the pollen (2).

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## Ragweed areas and preventive measures in Ukraine

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#### Background

Ambrosia spp. was introduced to Ukraine by a few routes and in different years. In accordance for some sources, a German pharmacist Krikker grew ragweed in the Dnipropetrovsk region as a medicinal plant (the substitute for quinine and as an anthelmintic remedy) in 1914. Ragweed was found in the Kyiv region in 1925. The army of General Denikin brought Ambrosia with seeds of alfalfa to Eastern Ukraine, so this weed was spread in Zaporozhye, Donetsk and Lugansk regions. The next ragweed intervention to Ukraine was registered in 1946 when the first wheat consignment was shipped to USSR from USA.

In accordance with the data of Quarantine State Inspection of Ukraine the area contaminated by ragweed in 2013 is 31,5 times increased from 1973 (3 523 138,442 ha versus 107 600 ha accordingly. Presence of this allergenic weed, *Ambrosia artemisiifolia* mostly, is currently noted in every from 25 Ukrainian regions. The Eastern region of Ukraine is more contaminated with this weed. Government reports the largest area of *A. artemisiifolia* in Donetsk (1016796,04 ha), Zaporizhzya (838835,22 ha), Mykolayv (813406,3183 ha), Kherson

(288763,88 ha), Kyrovograd (276334,67 ha) and Dnipropetrovsk (193721,79 ha) oblasts. Ragweed is usually spread from southern and Eastern parts of Ukraine toward North-West. The seeds distribution occurs by cars, railway, with sunflower seeds contaminated by ragweed due to their transportation from steppe to forest-steppe zone of Ukraine for planting. Sensitivity of compromised children to Ambrosia pollen was 3% in 2000 and it was 10% already in 2009 in the Western part of Ukraine. However, the Ukrainian west is still characterized by the minor ragweed areas. While the Central region of Ukraine is contaminated by ragweed particularly, the aim of our study was to estimate the changes in ragweed area in Central region of Ukraine for the recent years.

#### Method

To establish the area contaminated by *Ambrosia* spp. the data of Ukrainian Phytosanitary inspection was used. Pollen collection was done by gravimetric sampling for years 1999 and 2000. From 2009 to 2013 research used volumetric methods employing a Burkard trap placed at a height of 25 meters above the ground on the roof of a Vinnitsa Medical University building. Samples were taken from March 1 until October 31.

#### Results

Ambrosia pollination had been static in abundance from 1999 to 2009 in Vinnitsa, Ukraine, but has increased from 2010 to 2011 and now. The greatest ragweed pollen count is seen during the third ten day period of August (up to 760 pollen grains/m<sup>3</sup> for the year 2011) and the first and second ten day period of September. The eradicative measures to prevent the spread of ragweed included the cut of Ambrosia plants before the flowering season mostly. Penalty for the householdings and farms for ragweed plants found on their territory is established as well. Control of ragweed area includes check of sunflower seeds contamination with seeds of ragweed before planting. Intensive eradication of Ambrosia in Vinnitsa region in summer, 2012 led to significant (4-5 folds in comparison with year 2011) decrease of pollen count of this type in ambient air of Vinnitsa city. This tendency remained for the year 2013 after proper prevention of ragweed flowering in Vinnitsa region.

## Morphology and vitality of the ragweed pollen grains from agricultural, industrial and ruderal areas

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Ragweed (*Ambrosia artemisiifolia*) possesses all 'weapons' of aggressive weeds such as high ecological tolerance and ability of rapid regeneration and colonisation which enable them to become the pioneer plant species of secondary succession. As the majority of pollutants coming from traffic, industrial and agricultural activities gets into our environment, we compared the morphology and vitality of the ragweed pollen grains coming from different agricultural, industrial areas and ruderalia.

In areas exposed to strong pollution morphological changes of ragweed pollen were observed. In pollen samples from ruderalia the average number of deformed pollen was 40 %. Values were significantly higher in samples coming from industrial and agricultural areas. The quantity of deformed pollen was correlated to the pollen viability. The prolin concentration of pollen grains from ruderalia was the lowest. Our results suggest an environmental impact on pollen morphology, affecting the vitality of pollen grains, which can modify allergic symptoms.

## RAGWEED PLANTS AND POLLEN SPREADING IN PARMA, NORTHERN ITALY

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Ragweed is an annual, anemophilous weed producing pollen extremely allergenic. The genus Ambrosia (A.) includes some species of ragweed. Pollen of ragweed represents a question of public health in several European countries where plants are abundant. In France and Hungary 80% of the allergic population is sensitized to ragweed. In Italy most affected region by ragweed is Lombardy where the plant is widely distributed and where its pollen represents a major problem for human health; in this area it now represents the first cause of pollinosis. In the Parma area, until a few years ago, the plant had not been identified as anything other than sporadic. In our previous report, we described the situation up to 2008, with a significant increase in SPI (Seasonal Pollen Index) and in patients with positive SPT (Skin Prick Test) for ragweed pollen, and among these a significant increase of asthma. The aim of our study was to confirm pollen results for subsequent

years until today and detect the presence of ragweed plants sources on the territory. Parma lies in the Po valley, to the South of the Po River. The aerobiological surveillance was carried out according to the methods of the Italian Association of Aerobiology. We checked aerobiological data regarding start dates, end dates, duration of pollination, peak dates, peak values and SPI for Ambrosia pollen according to Jäger et al. 1996. In Parma we had season start on Aug 7th, season end on Sep 21th, season duration 46 days, day of peak Aug 30th, peak (p/m<sup>3</sup>) 36, SPI 245 grains, median calculated during last 18 years. We found an increased, not significant, trend for SPI (y=29.741x+9.961, R<sup>2</sup>=0.495), peak value (y= 3.205x+12.549, R<sup>2</sup>=0.286) and a decreased trend for duration (y=-0.632x + 57.621, R<sup>2</sup>= 0.028). In the last two years, we found a decrease in ragweed pollen (-57.75% 2012 vs 2011, -53.91% 2013 vs 2011). In Parma, although it has been observed in the neighboring

Province of Piacenza, there is currently no evidence of the presence of Ophraella communa, which feeds on ragweed leaves and which has been probably associated to a decrease in the concentration of ragweed pollen in Lombardy during 2013. The survey work carried out on our territory has identified numerous sites spreading A. artemisiifolia L. (21), A. coronopifolia Torr. & A. Gray (15) and A. trifida L. (3). On the basis of these results and considering our previous results we expect a rapid increase of allergy to ragweed related to the Ambrosia spread on our territory. The case of Lombardy is cautionary: almost nothing was done despite numerous warnings to fight ragweed at an early stage of the invasion. In our area the situation is tracing what happened in Lombardy during the early years of the spread of the plant. The consequence of the lack of action taken by the authorities may be very important in terms of public health and health costs in our territory.

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## Diffusion and trend of ragweed pollen in Italy

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Five species of ragweed are present in Italy: Ambrosia maritima L., A.artemisiifolia L., A.tenuifolia Sprengel, A. psilostachya D.C. (A. coronopifolia Torr. & A. Gray), A. trifida L.

In the 80's, ragweed pollen was recorded only in northern regions such as Lombardy, Piedmont and Emilia Romagna. The last ragweed pollen map, dated 2011, showed that this pollen was also detected in Central and Southern Italy. Nevertheless, values recorded in these regions were significantly lower and sporadically detected than those recorded in the northern regions. Moreover, within the Po Valley, some areas of Lombardy (North-Western Milan and Southern Varese areas) represented the area most polluted by ragweed in Italy and one of the most in Europe, reaching a Seasonal Pollen Index (SPI) of 6,915 and a peak

of 1,001 p/m<sup>3</sup>, meanwhile the rest of the Po Valley area presented lower values (average SPI of 3,209, varying from a minimum of 150). A first assessment of the ragweed pollen trend (Cox-Stuart test; p<0.05 significant), showed that from 2001 to 2012, an evident, but not statistical significant increase in the SPI has been observed in the Po Valley, whilst no trend has been observed in the Alps.

The aim of our survey is to update the diffusion and trend of ragweed pollen in Italy.

The following table shows the pollen season parameters of 2013 for each area, calculated according to Jäger, the SPI and the peak values.

With regards to the distribution of zones presenting different levels of ragweed pollen concentration along the peninsula, the situation of 2013 was similar to 2011. Meanwhile the results showed an overall decrease in the SPI, more prominent in Northern Italy, even if this finding doesn't influence the long-period trends from 2001, which seems to be increasing in the Po Valley (though non statistical significant; p=0.344) and absent in the Alps. Nevertheless, a shorttime analysis showed a wide reduction in the SPI of 2013 compared to 2012. As well as the different meteorological parameters to explain this decrease, could be due to the presence of Ophraella communa (an oligophagous leaf beetle), detected during the summer of 2013 in Northern Italy. But, further investigation is needed to evaluate the beetle's impact on ragweed pollen production and other possible causes, such as meteorological and environmental factors

Year 2013	Season start	Season end	Season duration (days)	Day of peak	Peak (p/m <sup>3</sup> )	SPI
Alps and Pre-Alps	22-Aug	21-Sept	31	4-Sept	32	369
Po valley	12-Aug	23-Sept	43	3-Sept	43	643
North Tyrrhenian*	10-Aug	18-Sept	39	2-Sept	7	49
North Adriatic	13-Aug	24-Sept	43	15-Aug	12	115
Central Tyrrhenian *	1-Aug	28-Sept	58	2-Sept	3	26
Central Adriatic *	21-July	25-Sept	66	21-Aug	2	18
South Adriatic-Puglia *	10-Aug	19-Sept	40	1-Sept	16	128
Sicily-South Tyrrhenian*	24-Sept	25-Sept	2	24-Sept	1	1

## The impact of meteorological variables on ragweed: data analysis and proposal of a forecasting model for the area with high pollinic concentrations in piedmont (Italy)

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Extreme high and low temperatures and precipitation totals may have important effect on daily and annual pollen concentrations. The aim of this study was to analyze the pollen seasons of ragweed (Ambrosia) in relationship with the meteorological data and to propose a statistical approach to forecasting the cumulative concentrations of Ambrosia, in order to provide early information to allergists and allergic people. For this study the station of Novara (Italy) was selected because the highest concentrations of the whole region are recorded in this area. We used the airborne pollen data from 2002 to 2012 and meteorological data for the same period (average, maximal and minimal air temperature, cumulative rainfall, difference between maximal and minimal temperature, growing degree-day). The results showed that the concentrations of ragweed are strongly increasing in the monitoring station of Novara (Figure 1). Also the meteorological variables suggested a certain correlation with the concentration of pollen. In particular, in the forecasting model tested, the meteorological factors

registered in the period before the start of the season seem to influence the amount of Ambrosia of imminent pollen season.

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### The problem of ragweed pollen in krakow against a background of other polish regions and some allergological aspects

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The problem of the Ambrosia artemisiifolia occurrence in Poland is still open. The permanent pollen monitoring has been performed over 20 years and the potential impact of A. artemisiifolia on crops was assessed, but the predicted direction of plant migration into Poland and the problem of allergy to ragweed is not fully confirmed. This report gives the present state of knowledge of the ragweed problem in Krakow in relation to the aerobiological data obtained in PAN centres, and to some medical aspects. Nowadays, on several sites in Poland A. artemisiifolia has been settled for a dozen or so years generally not demonstrating the tendency to spread beyond these sites (the surroundings of Przemy1 and Lublin, Kraków, Czarna Tarnowska), however in a few regions with the distinct tendency of colonizing of new sites (e.g. the Silesia region). In spite of not favourable climatic conditions, considering the tendency of temperature rising, the risk of fully settlement of A. artemisiifolia in the region of the Silesia Upland

and Lubelszczyzna is possible. In spite of not scattered distribution of Ambrosia plants, its pollen is observed from year to year in many regions of Poland. The results of the regional ragweed pollen monitoring in Poland in 1991-2012 pointed out that the highest pollen concentrations were noted in Lublin (daily counts exceeded 300 PG/m3, annual total exceeded 1000 PG/m<sup>3</sup>), while the lowest in Kraków (daily counts up to 80 PG/m<sup>3</sup>, annual total up to 200 PG/m<sup>3</sup>). The distinct rising trend of annual total pollen concentration was not observed in the studied years, while the weak trend towards earlier pollen seasons was observed in Sosnowiec. It was indicated that ragweed pollen registered in Poland originated partially from the Pannonian Plain (southern path) or from Ukraine (eastern path). The direction of the pollen influx from Ukraine, Slovakia and the Czech Republic needs to be confirmed including data from these regions. The potential threat of ragweed allergens to human health is hard to estimate. Regarding the

information from other allergological centres in Poland, the percentage of patients sensitive to Ambrosia allergens ranges from several to about 50%. The results obtained in the 90ties of the 20th century in Krakow pointed out the readiness of population to the immunological response to ragweed allergens. Nowadays, in a group of 90 patients suffering from allergic rhinitis caused by pollen allergens, the sensitization to ragweed was confirmed in 16 patients on the basis of the positive SPT. Only in 5 of the ragweed sensitized patients, the oral allergy symptoms were manifested. The relevant knowledge on the threat of ragweed in Poland and the pathomechanism of human population sensitivity needs to be improved to make the evident contribution in the integrated activities on A. artemisiifolia control in Europe.

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### Ambrosia pollen at high altitude (Campo Imperatore-Gran Sasso d'Italia)

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In order to explore the aerobiological particles present at high altitude five years of measurements were carried out at the monitoring station housed in the Alpine Botanical Garden of Campo Imperatore. The Botanical Garden is located in the Gran Sasso massif (in the Gran Sasso Monti della Laga National Park) at 2117 m a.s.l. and it accommodates the main peculiar species of the regional mountainous flora. At present there are some over 300 autochthonous plant species cultivated in the Garden, with particular attention being paid to those that are rare and vulnerable including numerous endemic species and glacial relicts (1). The vascular flora of the Gran Sasso has 2364 species accounting for 72.3% of the flora of Abruzzo region (2). The aerobiological monitoring was conducted only in summer months due to severe weather condition that avoid the access to the garden during the rest of the year. The natural vegetation of the Gran Sasso area is composed of various types of habitats such as meadows, cliffs, scree and rocks. The analysis of the percentage of airborne pollens shows that most of the grains belong to the families of Urticaceae (44% in the year 2012), Asteraceae (23% in the year 2009), Gramineae (28% in the year 2012) were also identified pollen of Euphorbiaceae, Polygonaceae, Pinaceae, Umbelliferae, Corylaceae and Fagaceae (Castanea sativa). As far as the fungal spores was detected the presence of Alternaria, Cladosporium, Epicoccum, Helminthosporium, Pleospora, Polythrincium, Stemphylium and Torula, the most prevalent are Alternaria and Cladosporium. In Abruzzo region the presence of Ambrosia psilostachya is certain in the sandy beaches of Martinsicuro (Te) while in Marche region is present Ambrosia artemisiifolia at various location (between Pesaro and Falconara), along the river Candigliano (at Furlo) and along the river Metauro (at Fossombrone) (3). During the summer season of the years 2010-2013 were captured and identified pollen compatible with Ambrosia. In particular, were identified respectively: 6 pollens (August 2010), 36 pollens (August / September

2011); 37 pollens (August / September 2012); 36 pollens (August / September 2013). The study presented here, carried out in collaboration with the Center of Excellence CETEMPS, aims to identify the origin of the pollens detected at the Botanical Garden of Campo Imperatore using the back-trajectories analysis. These observations and analysis allow us to identify the possibility of transport at high altitude of pollens produced in Central European area and the Balkan Peninsula.

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### Comparison of Ambrosia pollen seasons in the Air of Lublin (Poland) and Lviv (Ukraine) in 2011-2013

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A. artemisiifolia L. and A. psilostachya DC are the most common species of the genus Ambrosia in Poland. In turn, A. artemisiifolia L., A. trifida L., and A. aptera DC are the most prevalent Ambrosia species in Ukraine. The aim of the study was to compare the concentration of Ambrosia pollen in the air of Lublin and Lviv and to assess the length of the pollen seasons. The investigations in Lublin were carried out using the volumetric method. In Lviv, the gravimetric method was employed and, in order to compare the data obtained with the different methods, the gravimetric method results were converted in accordance with the recommendations of Basset et al. (1978). The distance between Lublin and Lviv is ca. 200 km. In the years 2011 and 2012, the Ambrosia pollen season determined with the 98% method started in Lviv in the first week of July and in 2013 - on July 18. In Lublin, the onset of the pollen season was recorded a month later during the first two years and in 2013 - 6 days later (July 24) than in Lviv. The end of the Ambrosia pollen season in 2011 and 2012 was noted in the second half of October in both cities, although in Lublin 6 days later than in Lviv. In turn, the pollen season in 2013 was considerably shorter in both cities and persisted until the third decade of September. In 2011 and 2012, the sum of daily values of pollen grains during the seasons were higher in Lublin (216 and 125 pollen grains) than in Lviv (103 and 111 pollen grains); in contrast, in 2013 a reverse relationship was observed: substantially higher quantities of pollen were recorded in Lviv (64 pollen grains) than in Lublin (26 pollen grains). Maximum pollen concentrations in Lublin and Lviv were achieved in the same periods: 27.08 (2011), 12.09 (2012), and 20.08 (2013). In 2011 and 2012, the values of maximum concentrations were 3-fold higher in Lublin (38 and 23 P/m<sup>3</sup>) than in Lviv (10 and 7 P/m<sup>3</sup>), whereas in 2013 the values reported from Lviv were nearly two-fold higher (9 P/m<sup>3</sup>) than in Lublin (5 P/m<sup>3</sup>). Importantly, no Ambrosia localities were found in Lublin and its surroundings, whereas the plant has numerous localities in Lviv.

#### Conclusions

More considerable amounts of pollen grains were recorded in Lublin than in Lviv. It may be connected with using of different research methods.

The annual sums of *Ambrosia* pollen grains noted in Lublin during the 3 years exhibited high variation (216, 125, 26 pollen grains), which may be related to the longdistance pollen transport dependent on the changing meteorological conditions.

The annual sums of *Ambrosia* pollen grains recorded in Lviv (103, 111, 64 pollen grains) were more similar than in Lublin, which may be associated with the occurrence of plants of this taxon in the native vegetation.

Considerable differences in the length of the *Ambrosia* pollen seasons between the 3 investigation years were noted in both Lublin and Lviv. This trend was similar in both cities.

## Dynamics of ragweed (ambrosia spp.) pollen seasons in the conditions of central-eastern Poland

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Three species from the genus Ambrosia have been reported from Poland, i.e. A. artemisiifolia L., A. psilostachya DC., and A. trifida L. The first species mentioned has a status of an invasive plant in Poland. A. artemisiifolia occurs in many localities dispersed in different parts of the country, with a majority thereof in the south-western part of Poland. Monitoring of Ambrosia pollen grains was performed in Lublin (central-eastern Poland) with the volumetric method in 2001-2013. A Lanzoni VPPS 2000 spore trap was located on a building roof (18 m) in the city centre. Ambrosia pollen seasons were recorded; they began in the first decade of August and ended in the third decade of October. The maximum concentrations of

pollen grains were recorded at various dates between August 20 and September 12. The values of the highest diurnal pollen concentrations oscillated between 5 and 320 P/m<sup>3</sup>/24 h. The annual pollen index (API) ranged from 27 to 1200 grains. Trend line analysis performed based on the investigations conducted in Lublin revealed a downward tendency in both the maximum diurnal concentration of pollen of this taxon and the annual sums of pollen grains.

In the present study, we show the number of days characterised by an *Ambrosia* pollen concentration exceeding the threshold value, at which allergy symptoms appear in susceptible individuals (5 P/m<sup>3</sup>/24 h). In 2001-2013, there were 1-16 days on which the threshold value for the concentration of pollen of the taxon was exceeded. Spearman's correlation coefficients between the concentration of Ambrosia pollen and meteorological conditions (temperature, precipitation, humidity, wind speed) revealed a significant correlation solely with temperature. In turn, intradiurnal assessment of pollen concentrations shows that the largest quantities of Ambrosia pollen grains persisted in the atmosphere of Lublin in different hours of the day, which may imply long-distance transport of pollen of this taxon. This corresponds with the results of phytosociological investigations, according to which there are no localities of Ambrosia in the Lublin region (up to 100 km).

## EFFECTS OF WEATHER FACTORS ON CONCENTRATION OF RAGWEED POLLEN IN BUSTO ARSIZIO AREA (LOMBARDIA)

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In this research we wanted to investigate the effects of weather factors on concentration of ragweed pollen in the air during its pollination season.

#### Method

Pollen was recorded in Busto Arsizio with a volumetric pollen trap (VPPS 2000 Lanzoni) and pollen counts were obtained according to the Italian Association of Aerobiology (AIA) criteria. Metereological data was provided from both Centro Geofisico Prealpino and both Malpensa airport archive.

#### Results

The research has been carried out on the

data collected over ten years of aerobiological sampling 2003-2012 by the Allergology Department of Busto Arsizio Hospital (Varese - Italia). The weather parameters taken into account were average, minimum and maximum temperature, relative humidity, precipitation, storm and strong wind. The ragweed flowering in the area starts in the first decade of August and continued until the first decade of October. The pollination peak is in our Country between de August last days from the first September 10 days. The highest pollen counts have been recorded in the years 2009, 2011 and 2003, when the temperature in the month of August was medium-high, together with long periods of good weather. The flowering started or increased after rainy period associated with temperature increase. The lowest

pollen counts occurred in 2005 and coincide with the lowest average temperature in August during the period considered (21°C). The relative humidity in the flowering ragweed time varies from 20-100% (from last July days to the first October 15 days).

#### Conclusion

Using a multi-linear regression over 10 years of pollen data we have obtained a model statically significant ( $R^2=74\%$ , F<0,05) which shows that the pollination depend to mean air temperature and millimeters of rain. The weather factors have consequences on the ragweed flowering and on total annual Pollen Index ragweed pollen.
### Observations on Ambrosia pollen Morphology

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In Italy, Ambrosia artemisiifolia L. is listed among the plant species of allergenic importance. Five species from the genus Ambrosia are reported for Italy, variously distributed in the territory (1, 2). The aim of aerobiological monitoring is to identify with a reasonable certainty the majority of pollen and spores captured by a volumetric sampler. The identification is carried on by optical microscopy (OM), and studying the morphological characteristics of the grains. The pollen grains of Ambrosia are typically tricolporate, spheroidal or slightly oblate. All species have short echines, conical in shape; the shots are very short, almost circular (3). The purpose of this preliminary study is to verify the distinctive characteristics in the OM of the pollen grains of the five different species of Ambrosia genus present in Italy. Pollen of the different species was collected from plants in open fields or from exiccata preserved in the herbarium of the Museum

of Natural History, University of Florence. The slides were prepared according to standardized methodologies (4, 5); each of them was photographed and 50 grains of each species were measured by using the OM model Zeiss LAB A1 equipped with a camera Axio Cam ICc5. The pictures were taken at 400x magnification. The pictures were analyzed with the program Rovellometro, that allows digital measurement of objects; the following parameters were measured: polar diameter, equatorial diameter, width of the echines, height of the echines, width of the colpi, height of the colpi. The average of the considered parameters and standard deviations for each of the 5 species were: polar diameter: 19.74±1.62 μm, equatorial diameter: 19.85 $\pm$ 1.82 µm, width of the echine: 2.14±0.26 µm, height of the echine:  $0.73\pm0.07$  µm, width of the colpi: 3.46±0.33 µm, height of the colpi: 2.26±0.16 µm. Moreover, for the parameters polar and equatorial diameter, width and height of the echine, one-way ANOVA test was applied. No significant differences, with p >0.05 were observed. All these different species contribute to airborne pollen concentrations in Italy and their distinction is very difficult to OM.

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## Study of intra and inter population genetic variability of common ragweed in relation to amb a 1 isoforms and their allergenicity

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The alien plant Ambrosia artemisiifolia L. is one of the main allergenic species in Europe, particularly in Northern Italywhere it has become the second cause of respiratory allergy in the last 2 decades (1). Rgweed causes great concern because it is one of the most invasive species in Europe and produces large amounts of pollen containing noxious allergens. The major allergen of ragweed pollen is Amb a 1, a protein belonging to the family of pectate lyase and showing many genetic isoforms (Amb a 1.1, Amb a 1.2, Amb a 1.3, Amb a 1.4, Amb a 1.5) (2). The expression and the allergenic potential of these different Amb a 1 isoforms in ragweed population and in single plants are currently unknown.

The aims of this research are:

to determine the inter and intra-population expression variability of Amb a 1 isoforms and their immunological reactivity

• to identify genetic and environmental factors responsible for the possible different expression of the isoforms.

To this end, seeds from 15 different populations (5 from Canada, 5 from France and 5 from Italy) were collected and used to obtain ragweed plants.

First, plants were characterized by applying molecular markers (SSR), in order to assess the intra and inter population genetic variability. The analysis of 20 plants per population confirmed the previous results obtained in French and American populations (3) showing high and low intra- and inter-ragweed population variability, respectively. On the basis of our results, it is likely that the origin of Italian ragweed populations is the Canadian area of Quebec the same area from which French ragweed originated. In order to collect pollen grains, at least five plants per population (in total 90 plants) were left to flower in controlled condition inside a greenhouse. Pollen samples were collected from each individual and are currently used to study the expression and the immunological reactivity of the Amb a 1 isoforms. The preliminary results will be reported and discussed during the meeting.

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# Common ragweed (Ambrosia artemisiifolia): systems biology to understand the reaction of the allergenic pollen to air pollutions and climate change

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Ambrosia artemisiifolia (common ragweed) is an invasive neophyte from North America, which is now spreading throughout Europe. This annual herb produces huge amounts of pollen grains that have a very high allergenic potential and are known as the prime cause of allergic diseases in the US. Climate change and air pollution will affect the allergenic potential of pollen, either by changes of the pollen season or the pollen amount (1-3), by a changed surface exine or by directly increasing the transcripts and the expression of allergenic proteins, and interactions with biologically important ligands, e.g., flavonoids. We carried out a systems biology approach of ragweed pollen upon elevated O<sub>3</sub>/CO<sub>2</sub>/NO<sub>2</sub> and drought stress towards a better understanding of molecular mechanisms in ragweed pollen under

global change scenarios. Fourier transform infrared spectroscopy (FTIR) indicated changes of pollen cell wall components upon  $O_3$ -fumigation (4). Upon elevated CO<sub>2</sub> plus drought stress conditions it has been shown by HPLC that individual flavonoid metabolites were increased. Transcriptomic analyses showed changes in allergen-encoding transcripts upon elevated  $O_3/CO_2$  and drought stress. NO<sub>2</sub>treatment resulted in increased amounts of allergenic proteins. In addition, nitrosylation of Amb a isoforms was observed that may also influence the allergenic potential. Treatment of mice B cells with pollen-extracts from CO<sub>2</sub>-fumigated and/or drought stressed plants augmented the IgE enhancing effect, indicating an impact on allergic diseases. The data highlight an influence of climate change on the

transcript and protein level of pollen allergens and support the idea of a direct influence of air pollution on allergens. In addition, the large-scale sequencing contributes to the identification of stress-related transcripts in mature pollen (4).

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# Ragweed in Busto Arsizio: since 1986 to date

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A study on patients referred to Allergology Department of Busto Arsizio Hospital between 1986 and 1990 showed that in 1986, 22,8% of skin positive patients had sensitivity to Ragweed, in 1987 28,1 %, in 1988 28,6% in 1989 27,5% and in 1990 39,9%. A further study (1995) showed that in 1994 the percentage of patients sensitive to Ragweed had reached 47,9%. In the same way, since 1988, year of the beginning of the daily pollen sampling, we observed a concentration increase of Ragweed pollen in atmosphere from August to September. We started from a pick of 125 pollens/m<sup>3</sup> air in 1988 and we arrived, with annual alternations related to climatic conditions, to 672 pollens/m<sup>3</sup> air in 1998 and 780 in 2009. A further review of the case study has been made between 1998 and 2002 on approximately 2000 Allergology patients. 71% of skin positive patients turned out Ragweed sensitive, in most cases in combination with others

pollens. Finally we considered 2151 patients who went to Busto Arsizio Allergology, between 2008 and 2012, to undergo SPT. 1558 of them (72%) resulted positive to inhalant allergens and 1189 (76%) had been positive for Ragweed. 801 patients (in this last 1189 group we are now considering) are symptomatic in the flowering season and 68 (8,5%) of them result monosensitive. Regarding symptoms, patients mainly show rhinitis (64,3%). Asthma associated with rhinitis is present in 31,8% of cases. The asthma as a single symptom in 3,1% and conjunctivitis (as a single symptom) in 0,6%. Conclusions: comparison of works carried out throughout a period of almost 30 years in Busto Arsizio Allergology Department showed a significant and growing sensitization of Ragweed pollen moving from 22,8% in 1986 to 71% in the period 1998-2002. In the last decade we have seen a much slower growth of sensitization (71-76%), even though the pollen concentration in the considered years had a similar trend as previous years. This evolution can be explained considering that at the beginning of 2000's we reached a stabilization of local people sensitization. The slight increase in ragweed-positive patients in these recent years can be explained by the sensitization of people migrant from country without Ragweed.

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## Common ragweed in the romanian plain: HISTORY OF PLANT IDENTIFICATION AND ACTUAL SENSITIZATION PREVALENCE IN ALLERGIC RHINOCONJUNCTIVITIS

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Common ragweed (Ambrosia artemisiifolia var. elatior) has two common Romanian names: floarea pustei and iarba parloagelor. The Romanian Plain is located in south part of the country and has a temperate continental climate, with local submediteranean, semi-arid and humid subtropical influences. In Southern Romania, ragweed was first identified in 1908 at Orsova (1, 2), a Danube river port city with shipyard since 1890. Years later this invasive weed was reported in Oltenia (3), Muntenia (4) and Dobrogea (5). The presence of ragweed is indicated recently in all regions and it seems more extensively spreading towards south and centre of the country, the last reports coming from Baragan and Southern Moldova (6). Long-distance transport of ragweed pollen from the Pannonian Plain may be discussed as a source of this weed pollen in the Balkan Peninsula (7, 8), but this seems not to induce new sensitizations in the short term (9) and the Carpathian Mountains separate this re-

gion of Hungary from the Romanian Plain. Recent local studies (10, 11) suggest that sensitization to common ragweed pollen is significant in patients from the Romanian Plain presenting allergic rhinoconjunctivitis with or without asthma symptoms. Sensitization to weeds pollen is important in almost a quarter of patients with respiratory allergy, the rate of sensitization to ragweed pollen being similar compared with mugwort pollen (15%). The vast majority of patients with allergic rhinononjuctivitis symptoms from end of July-August to September-mid October have positive skin prick tests to Ambrosia and/or Artemisia pollen extract, sometimes raising the issue of cross-reactivity or cosensitization (12, 13).

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## RAGWEED ALLERGY IN THE LEGNANO AREA: OVER 20 YEARS OF STUDY

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### Introduction

Ragweed (*Ambrosia artemisifolia*) has been expanding in many European countries during the second half of the 20<sup>th</sup> century. The aim of this study is to analyse the development of the Ambrosia sensitization and its clinical symptoms in more than 20 years in Legnano.

### Materials and methods

The study population consisted in an average of 1100 adult patients per year in the period between 1989 and 2012. The sensitization of patients to Ambrosia and other aeroallergens was tested by skin prick test. The patients have been interviewed about clinical symptoms and the time of their appearance.

### Results

The sensitization rate to Ambrosia increased from 24% to over 70% among the patients who resulted positive to the skin prick test. The percentage of positive patients with respiratory symptoms in late summer markedly increased in the period between 1989 and 2004, and reached a plateau from 2004 to 2012. The appearance of the Ambrosia sensitization has increased in patients older than 50 years.

### Conclusions

Ambrosia allergy is very frequent in the area of Legnano, with a constant increase of the sensitization rates in the last 15 years, and a higher number of people with asthma symptoms. Furthermore, elderly people too can develop allergic sensitization to Ambrosia.

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## RAGWEED POLLEN SENSITIVITY AMONG CHILDREN OF CENTRAL UKRAINE

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### Background

Ragweed pollen count is raised year-byyear in the central area of Ukraine. Its provoke ragweed pollen sensitization among the population of Vinnitsa region for recent years. In the regions with old and well-known contamination by Ambrosia pollen like Dnepropetrovska oblast of Ukraine sensitization of this type was rated as 7-21% among children in years 2005-2007. It was often associated with the sensitivity to mugwort (Artemisia), sunflower (Helianthus annuus) and giant sumpweed (Cyclachaena xanthiifolia) pollen. Children' rate sensitive to both Ambrosia and Cyclachena pollen grains ranged from 10,38 to 27,52%. Sensitization to ragweed and mugwort pollen were seen for 10,85% to 27,52% of children tested there [1]. The aim of our study was to determine the character of sensitivity of children to ragweed pollen grains in the Vinnitsa region.

### Method

Symptoms of seasonal allergy were analyzed by reviewing medical records from an allergy specialty clinic at Vinnitsa Re-

gional Clinical Children's Hospital, Vinnitsa, Ukraine. 38 patients aged from 3 to 16 years were reviewed with 20 selected for further analysis among children admitted from 2004 to 2013. Prick tests for inhalant pollens using extracts made in Ukraine were done.

### Results

17 or 85% of patients were males. Only 3 were from urban Vinnitsa, others being from rural areas. Children aged 6 to 10 years old typically had the first appearance of allergy symptoms. 13 or 65% of children were tested at this age for the first time. Sensitization to pollens prevailed over other sensitizations in children aged 6 to 16. Ragweed and sunflower allergens were leading pollen causal agents for allergy symptoms for children aged from 3 to 5 years, often with very high sensitivity. At the next age group ragweed sensitivity was less prominent. Poaceae pollen was the leading causal agent for allergy symptoms of the children aged 6 to 10 years. Ragweed-sensitized patients strongly reacted to the mugwort and sunflower pollen in this group as well. Sensitivity to tree pollen was revealed in the group of children aged 11 to 16. Ambrosia sensitization from moderate to very high level was determined just for 2 patients from 9 children tested in this group. Both of them were sensitized to mugwort and dandelion pollen. Allergy to grass pollen predominated among children of this group too.

### Conclusion

Ragweed pollen is the leading causal agent for allergy symptoms of the children from early childhood in central region of Ukraine. Sensitivity to Ambrosia pollen is replaced by Poacea and tree pollen allergy respectively with the age increase. Combined sensitization to different pollen types including Asteracea family members is seen for children tested.

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### Features of short-ragweed allergy in North Western Milan area: preliminary results

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The North-Western Milan (NWM) and South Varese areas (Lombardy, Northern Italy), are the short-ragweed most infested areas in Italy and one of the most polluted in Europe. A specific epidemiological study, showed that in the first area at least 14% of the sample population was allergic to ragweed, with an increase of prevalence from 9.2% (1996) to 14% (2005) and high percentage of asthma (more than 40%) in ragweed allergic patients. Ragweed is the main cause of pollinosis in the NWM area: a study (2008-2010) in an Allergy Service showed that 71% of new patients suffering from pollinosis were allergic to ragweed. A more recent study (2012) was conducted in order to evaluate the epidemiology and the prevalence of ragweed allergy in a sample population of the NWM area. During July, resident population of Sedriano (10,744 inhabitants) received a questionnaire about some personal features (name, age, sex, education, occupation) and some clinical data about the allergic symptoms and previous diag-

nosis of ragweed allergy. Only complete questionnaires were considered. A total of 1,790 valid questionnaires were collected: 888 males (49.6%) and 902 females (50.4%). The first data processing, already presented, showed that 230 people need further medical investigation, whilst 1560 people have diagnosis. Of them, 1304 were nonallergic (83.6%) and 256 allergic (16.4%): in the studied population ragweed allergy involves at least 16.4% of the sample, suggesting an increased trend, since previous study in the same area indicated a lower prevalence (14%). A higher prevalence was hypothesized, since 230 people waiting for a complete diagnosis are symptomatic and in the previous study diagnosis was confirmed in a large part of symptomatic people. The aggressive allergic behavior of ragweed pollen was confirmed: around 40% of ragweed allergic people show asthma, 88.3% rhinitis and 60.6% conjunctivitis. The aim of this work is to update the data processing of the 1,560 confirmed diagnosis people. No association was found between sexes and ragweed allergy. A relation was found between age groups and allergy. The age groups associated to ragweed allergy are: 46 to 65, 66 to 80 and over 80 years. The first group present 1.48 times the risk of allergy (Odds Ratio-OR-=1.48; age is a risk factor); the 2<sup>nd</sup> and 3<sup>rd</sup> group present 0.43 and 0.069 times the risk of allergy respectively (OR=0.43 and 0.069; age is a protective factor). An association was found ( $\chi^2$ =10.973) between education and ragweed allergy ( $\chi^2$ >3.84 significant) and the higher educated people show a 1.67 times the risk to allergy (OR=1.67). We also found a significant difference ( $\chi^2$ =18.99) between allergic and non-allergic people with regards to their work ( $\chi^2$ >16 significant); employees present a risk 1.57 times to develop ragweed allergy meanwhile retired people show 0.53 times the risk of developing ragweed allergy. This is a partial of results obtained from our study, which demonstrate some relations between ragweed allergy and some social demographic parameters.

# Smartphone application for ragweed description in Rhône-Alps

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The 20<sup>th</sup> of June, 2013, Valence Agglo launched its "ragweed application" in the framework of the International Ragweed Day. This free application is available on all smartphone types and aims:

- to alert people to be able to identify the plant via a specific menu;
- to locate the plant and inform Valence Agglo department with GPS coordinates;
- to inform people about the level of allergy risk for the days to come.

For each report, Valence Agglo department looks for the owner of the concerned field and asks him to destroy the plant. Some control rounds are made by Valence Agglo and reminder letters are sent. A mapping is carried out at the end of the season. In 2013, the application was downloaded 700 times in 2 months, and used more than 100 times: 104 reports were received and followed by treatments for public area or by information sent to the owner of the land. Among this reports, 92 of them led to a complete treatment of the infested area. For 2014, on the basis of the application, the ARS Rhône-Alpes contributes to the fundings of the development of a ragweed regional platform for reporting, which aims are to collect, manage and view the reports:

- collect via smartphones, emails or manual updates;
- manage, allow to experts to validate the reports and follow them up;
- view according to its land (town, department, region, etc...).

In the long term, the aim of this application is, on one hand, to evaluate the good management of the treatment of infested area and, in the other hand, to release and use the application on all the country and, maybe, across Europe.

## TREND OF PRESCRIPTION OF RAGWEED IMMUNOTHERAPY OVER TEN YEARS AS INDIRECT INDEX OF RAGWEED ALLERGY IN TREVIGLIO'S AREA

### G. MANZOTTI, A. BESOZZI, S. PANZA, M. DESTRO

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### Introduction

Ambrosia is spreading invasively in many European countries (1). Prevalence of this allergy is extremely variable with a peak of 40% in some areas even in Europe (2). The characteristics of ragweed pollinosis are: severity, long lasting duration and the presence of nasal/ocular involvement and asthma and/or tracheitis in many cases (3). In Treviglio's General Hospital, allergy clinic provide diagnosis and allergen immunotherapy (AIT) for the major allergic disease including Ragweed pollinosis, according to Word Allergy Organization (WAO) guideline (4). In Treviglio's area, Bergamo's airport and the realization of the new motorway "BREBEMI" connecting Brescia, Bergamo and Milan passing across Treviglio's suburbs (started around 2007 and just about being completed) could have played a role in Ambrosia diffusion.

### Objective

Aim of this paper is to analyse the trend of Ragweed Immunotherapy (IT) prescriptions over ten years follow up in Treviglio's area, located near Bergamo, Northen Italy.

#### Methods

Since 2004 a database of Treviglio's AIT prescription is constantly upgraded. Going through our files we scored total number of AIT prescriptions and number of Ragweed IT, either Subcutaneous or Sublingual. We designed the Ragweed IT trend over ten years.

### Results

The percentage of patients receiving Ragweed IT increased from 3.5% to over 6.7% among 10 years: The increasing rate seems to be constant except for a peak in 2008 (5.3%) and a second one in 2013 (6.7%).

#### Conclusion

In Treviglio's area, Ambrosia ITS prescriptions have almost doubled! Increasing prescriptions for Ambrosia IT over the time could be considered an indirect index of increasing number of patient not only sensitized, but also symptomatic to Ragweed, even in this area considered at low Ambrosia diffusion up to now. Despite application also in this area of regional low on Ambrosia environmental control (5), Ragweed allergy is becoming here a public health trouble, involving important economical aspects too. Our next aim is to collect data about Ragweed sensitization trend and clinical manifestation of respiratory allergy in our area, underlining the importance to compare clinical data with aerobiologic records, not available at the moment.

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### Assessment of health costs due to ragweed allergy

### M. BONINI, C. TESTONI, E. VALERIO

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Currently, the Northwest of the Province of Milan (Lombardy, Northern Italy) is one of the most European polluted areas by ragweed. Indeed, Ambrosia has enormously spread due to changes in the ecological balance since the early '90s, leading to the birth of a vast public health problem. Ragweed pollen causes the onset of allergic diseases, so the assessment of health costs related to it is an important tool to estimate annual Ambrosia impact on public health (1). Aim of this work is to assess the direct health care costs related to ragweed allergy in the Local Health Authority Milan 1 jurisdiction (924.417 inhabitants), from 2001 to 2011. Our hypothesis is an inference based on the annual percentage of new ragweed allergic patients of the total number of new patients suffering of respiratory allergies and pollinosis, the number of patients in SIT (specific immunotherapy), the data of hospital admissions due to pollinosis, the data of drug and allergenic extracts consumption as well as clinical and therapeutic experience of the allergists.

Data is furnished by hospital allergy departments (a total of 15 departments: 13 in public hospitals and 2 in private), Epidemiological Office and Pharmacy Service of the Local Health Authority Milan 1 and the two public Hospital Organizations (each is a group of several hospitals with the same direction). The following table shows direct costs related to ragweed allergy: data on costs for first examination testing, control examination in SIT patients, allergenic extracts, other drugs consumption and clinical admissions due to ragweed pollinosis.

This assessment of direct health care costs is however not exhaustive since it does not include the drugs sold without prescription, the indirect costs (loss of productivity by patients) and intangible costs (related to impairment in the quality of life) (2). Data shown could be useful to evaluate the effectiveness of activities to control ragweed allergy in Local Health Authority Milan 1 jurisdiction. This data would be more accurate if a ragweed dedicated data bank was available.

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Year	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011
First examination (€)	83.013	82.810	94.019	104.398	129.123	128.639	121.920	147.206	146.483	152.939	180.599
Control examination SIT patients (€)	114.435	68.216	86.464	73.884	95.361	112.459	112.876	81.663	91.783	84.819	76.403
Allergenic extracts (€)	191.319	141.264	194.064	177.521	223.636	263.734	274.456	275.470	335.256	378.369	333.731
Drugs (€)	1.251.182	1.084.631	639.527	981.656	1.145.266	1.426.646	1.143.661	1.389.622	1.115.623	1.105.310	1.216.590
Clinical admission (€)	32.065	13.496	18.709	11.278	17.498	27.282	13.671	24.321	16.748	20.849	24.128
Total costs (€)	1.672.014	1.390.417	1.032.783	1.348.737	1.610.884	1.958.760	1.666.585	1.918.283	1.705.893	1.742.285	1.831.450

Tab. 1 -

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### RAGWEED ACTION IN RHONE DEPARTMENT, France

### J. DA PASSANO<sup>1</sup>, M. NUEZ<sup>1</sup>, M. THIBAUDON<sup>2</sup>

<sup>1</sup>Conseil Général du Rhône, Lyon, France - Email: rnsa@rnsa.fr <sup>2</sup>Réseau National de Surveillance Aérobiologique, Brussieu, France

The Rhône General Council, under the aegis of Mr Jean-Luc Da Passano, elected at the highway department, pharmacist and Mayor of Irigny, has been committed for over 15 years in the fight against ragweed. This problem was difficult to position in public actions and had gone through many services, and has created (then aggregated) different types of action to make a complete package:

- from 2000 to 2003, public health service: establishment of information documents to pharmacies and MSA (agricultural insurance);
- from 2004 to 2008, in agriculture and environment department: development of a new communication media (itinerant exhibitions, letters to all authorities of the Rhone, ragweed toll free number, press articles, ...);
- from 2008 to nowadays, road services: recovery of the global coordination of existing packages, integration to the steering committee of Health authorities, design and implementation of revegetation techniques to take preventive actions in the fight against ragweed.
  During all these years, Jean-Luc Da Passano motivated and supported innovation of actions on the political level, giving necessary funds for:
- documents for communication: 5000 € /year;
- markets mowing insertion: 110 000 € / year;
- revegetation techniques: 400 000 € / year in the frame of cross-actions;
- personal along the roads: 2-3 mechanized mowing/year.
- Since 6 years, the revegetation tech-

niques in road sectors or wasteland have shown its efficiency. In 2013, the experimental site of Communay, infested by ragweed on all the 3.5 hectares of the field has been treated hydraulic grassing. Being a biological control, the aim is to compete with ragweed by planting other plants. This competition takes place by occupying the place where ragweed grows depriving it of sunlight and water. With an optimum price-quality ratio, 1.5 to 2  $\notin/m^2$ , this biological control avoids the use of herbicides but needs to be well-reasoned on two years, by following accurate procedures. After the first shoot of seedlings produced in 2013, this procedure has shown 100% of efficiency but has invited to be vigilant because ragweed seeds are viable in the soil during almost twenty years.

# The effects of ragweed on the public health: analysis of the entrances to the emergency room in the city of turin (Italy)

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The monitoring of allergenic pollen is becoming more important in relation to the increase of the prevalence of allergics in the last few years in Piedmont and in Italy. Like other species of pollen, the ragweed (Ambrosia) concentrations also are increasing in Piedmont and in the city of Turin. The scientific literature claims that ragweed pollen is a very strong allergen and causes various diseases in allergy sufferers. Symptoms include sneezing, inflammation of nasal and conjunctival membranes. Severe complications include asthma, chronic obstructive pulmonary disease, and anaphylaxis. The aim of this study was to analyze the stream of entrances in the emergency rooms of Turin for some typical pathologies of who are allergic to pollen. We used the airborne pollen data from 2007 to 2012 and health data for the same period (allergic rhinitis, conjunctivitis, pollinosis and asthma - ICD IX: 472.0, 477, 493). Examining the entrances to the emergency room, we have observed a correlation between increments of the ragweed concentration and the trend of access to the emergency room. In particular, the increments of average weekly of Ambrosia is matched by an increments of the number of weekly entrances into the emergency room, from the following weeks (Figure 1). Other aspects about the effect of the health will be deepened.



# RAGWEED POLLEN HYPERSENSITIVITY IN ATOPIC DOGS: EVALUATION OF 357 ALLERGIC TESTS FROM A REFERRAL PRACTICE IN MILAN (2003-2013)

### N. FURIANI<sup>1</sup>, L. ORDEIX<sup>2</sup>, C. SARETTO<sup>3</sup>, G. ZANNA<sup>3</sup>, F. SCARAMPELLA<sup>3</sup>

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During the last decades ragweed (Ambrosia artemisiifolia) has progressively colonized Northern Italy, as other parts of Europe, becoming the first cause of respiratory allergy in humans, especially in the area of Milan (1,2). The aim of this study is to describe the patterns of ragweed pollen hypersensitivity in atopic dogs from Northern Italy. The results of 357 allergic tests (319 intradermal tests and 38 serologic tests) performed between 2003 and 2013 were evaluated. Tests with ragweed positivity were checked for dogs' gender, age, breed and geographical origin, presence of concurrent reactions with other allergens and temporal distribution. Ninety tests, 82 intradermal (25,7%) and eight serologic (21%), were positive to ragweed for an overall prevalence of 25,2%, which was the highest among pollens. Among dogs there was no significant difference in gender distribution, the average age was 4,18 years and the most common breeds

were cross-breed (13,3%) and boxer (9%). Fifty dogs (55,5%) came from the province of Milan and the majority lived in urban areas. Ragweed presented copositivity with at least one other weed pollen in 82,2% of cases. The association with mugwort (Artemisia vulgaris) was the most frequent (50%). Positivity to ragweed showed high variability over the years, ranging between 9,1% (2008 and 2010) and 37,5% (2011). Similar results were observed when considering the tests from the province of Milan, with prevalence ranged from 4,1% (2007) to 33,3% (2011). This study confirms ragweed pollen as the plant allergen more involved in canine atopic dermatitis in Northern Italy as reported by our previous study, with an increased prevalence from 21,6% to 25,2% (3). Such as reported in humans, high prevalence of co-positivity between ragweed and mugwort was observed (4). Nevertheless, further investigations are needed to establish

whether this is due to cross-reactivity or co-sensitization. Changes in environmental factors could be partly responsible for the variability observed in the prevalence of ragweed positive reactions between different years.

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# Ambrosia day, 2014 Ragweed Allergy: 15 years of prevention April 4<sup>th</sup>, 2014, Rho (Milan), Italy





Regione Lombardia

### AMBROSIA DAY 2014 "ALLERGIA ALL'AMBROSIA: 15 ANNI DI PREVENZIONE" 4 Aprile 2014

### PROGRAMMA DEL CONVEGNO

15 Anni di Prevenzione

**Registrazione partecipanti** 13.00-13.30 13.30-13.45 Saluti Autorità M. Mantovani - Assessore alla Salute Reg.Lombardia, G. Scivoletto - Direttore Generale ASL Milano 1 13.45-13.55 Presentazione della Giornata Nazionale del Polline R. Albertini Sessione 1: La pianta e il polline - Moderatori: G. Frenguelli, S. Citterio 13.55-14.10 Introduction spread of common ragweed in Europe: B. Chauvel ecological aspects 14.10-14.25 Pollen grains H. Méon Sessione 2: La malattia nell'uomo e negli animali - Moderatori: G. Moscato, A. Flores D'Arcais Allergy: the human health impact of ragweed -Ambrosia-14.25 - 14.40 C. Déchamp pollen 14.40 - 14.55Allergia al polline dell'Ambrosia in medicina veterinaria N. Furiani 14.55 - 15.30 Tavola rotonda: "Come interpretare la doppia positività C.Ortolani, P. Bottero, A. Tosi, R.Asero, B. Jahn-Schmid Ambrosia - Artemisia" Sessione 3: Gli interventi di Sanità Pubblica - Moderatori: A.Tosi, P. Bottero 15 anni di prevenzione nell'ASL Milano 1 15.30 - 15.40M. Bonini 15.40 - 16.00Public health interventions against ragweed in Quebec N. Noisel Pausa caffè Sessione 4: La prevenzione del futuro - Moderatori: G. Frenguelli, M. Bonini, C. Ortolani 16.15 - 16.30Come controllare la pianta infestante Ambrosia M. Cristofaro artemisiifolia in Italia mediante un approccio biologicointegrato 16.30 - 16.45Sustainable management of Ambrosia artemisiifolia in H. Müller Schärer Europe (COST FA1203-SMARTER): objectives, recent achievements and future opportunities 16.45 - 17.00A population dynamics modelling approach for the S. Lommen management of ragweed - a case study of a potential biocontrol agent in Norther Italy 17.00 - 17.45 Tavola Rotonda: "E' possibile un nuovo modello di F. Giovanazzi, Dir. Gen. Agricoltura Reg. Lombardia - M. prevenzione?" Gramegna, N. Cornaggia, Dir. Gen. Salute Reg. Lombardia - M.Thibaudon, Direttore RNSA, Segretario IRC, Francia - T. Komives, Presidente IRC Ungheria - F. Caronni, V. Parco, Parco del Ticino - R.Grittini, Comune di Corbetta - G.Bogliani, Univ. Pavia 17.45 - 18.00Conclusioni ≤ DEGLI STUD cino Con il patrocinio di BICDCC International Ragweed Society

Rho (MI) - C.so Europa 228 - Collegio dei Padri Oblati – Sala Mantovani Furioli

### Comitato Scientifico ed Organizzatore

Responsabile scientifico: Maira Bonini Segreteria scientifica: Maira Bonini, Paolo Bottero, Sandra Citterio Segreteria Organizzativa: Segreteria del Dipartimento di Prevenzione Medica dell'ASL Milano 1

# Sessione I



# La pianta e il polline

# Moderatori: Giuseppe Frenguelli, Sandra Citterio

# INTRODUCTION SPREAD OF COMMON RAGWEED IN EUROPE: ECOLOGICAL ASPECTS

### B. CHAUVEL<sup>1,2</sup>

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Common ragweed (Ambrosia artemisiifolia) is an annual invasive species that colonizes perturbed and open habitats. The presence along the roadsides (1) of a large amount of this allergenic plant species during its flowering period is a major problem in terms of public health. Historical studies indicated that this species has been introduced at various independent geographical points and at various times in Europe. Commercial trade and American troops have contributed to its spread during the XXth century (2). At first present in cultivated habitats, its recent spread in different new areas is explained by the co-occurrence of different ecological and agronomical factors. Different surveys in Europe showed that ragweed infestations increase along roadside, which appears to be not only favourable habitats but also corridors for its spread in new areas. Different reasons can explain the success of common ragweed along roadside: i) a late germination in spring, ii) soil disturbance is favourable for the development of this annual species seeds, iii) winter salting limits the growth of the other species and gives to ragweed an advantage explained by its salt tolerance (3), iv) the seeds are disseminated by mowing machines (4).

The high ability of common ragweed to be invasive is due to its huge plasticity in seed mass which may help it to cope with a wide range of conditions and to establish in disturbed habitats.. But, if common ragweed appears to be a successful pioneer in early successional habitats with a high degree of disturbance (erosion area, burning places) (5), the species does not seem to be able in France to compete in habitats with a level of competition (meadow, forest). The efficacy of mowing is limited by the high resprouting capacity of common ragweed (4). The timing and the frequency of mown can modify its reproductive traits and this species seems always able to produce enough seeds that can maintain the density of the population. In France, a study of common ragweed spread on the 'Autoroutes Paris Rhin Rhône' network showed that its density increased between 2010 and 2012 despite a strict plant management. Due the high variability of this species, its ability to tolerate various stress (mowing, herbicide, eg.,), it seems that only a preventive management appears to be effective to control its spread before the establishment of a seedbank.

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# Pollen grains

### H. MÉON<sup>1,2</sup>

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Palynology, etymologically study of the dust, is the science that concerns specifically plant dust i.e. the spores and pollen grains. This discipline provides a wide scale of scientific activities or practices including geology, general biology, agronomy...



Fig. 1 - Ambrosia artemisiifolia L.



Fig. 2 - Ambrosia tenuifolia



Fig. 3 - Ambrosia artemisiifolia L.



Fig. 4 - Ambrosia tenuifolia

**Sexual Reproduction.** The fusion between 2 haploid cells, the gametes, gives one diploid cell: the zygote or egg. Gamete fusion occurs in two steps, always in the same order: fusion of cytoplasm (plasmogamy) and nuclear fusion (karyogamy). Karyogamy may be delayed. Two haploid nuclei (dikaryon) then coexist in the cell after the plasmogamy.

**Pollination.** Pollination is the process by which pollen grains are transported from one flower to another, allowing the approximation of gametes and fertilization. Pollination mechanisms differ between plant species. The main agents of pollination are wind and animals, especially insects.

Anther opening and pollen dispersal. Anther opening may be considered to occur in two simple steps: first, locular fluid disappears and second, the anther opens and the pollen grains are shed. Explosive dehiscence of pollen is a characteristic of anemophilous species.

"Ambrosia pollen is initially shed from the staminate flowers in large pollen clumps containing hundred of grains. These pollen grains are quickly broken, so at regional scales *A* pollen is released as a largely homogeneous plume" (1).

The pollen grains. Following the first haploid mitosis, pollen begins to store

starch, the amount of which varies according to season in ripe pollen grains.

The exine of spores and pollen grains present ornamentation varied and specific to each taxon. This ornamentation is clearly visible on when the material is acetolysised in which the cytoplasm has disappeared. Mature pollen grains are characterised by a thin, ornamented exine and an intine. Germination is through apertures (openings) that are thinnings of the outer wall (exine), circular (pores) or elongated (= colpus) shapes. In Ambrosia there are 3 colpus and 3 pores and a spiny exine. The specificity of the exine ornamentation is a remarkable property of the walls of spores and pollen grains, the chemical nature of this wall is different. The substance which constitutes it, sporopollenin is with cutin cuticles and chitin arthropods, one of the most resistant chemicals the living world.

Some pollen grains of different species of *Ambrosia*.

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# Sessione II



# La malattia nell'uomo e negli animali

Moderatori: Gianna Moscato, Alberto Flores D'Arcais

# Allergy: the human health impact of ragweed -Ambrosia- pollen

### C. DÉCHAMP

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Morrill Wyman, teacher at Harvard university, identified Ambrosia (A) rhinitis in 1872. He was ill in Boston and he was not in the White Mountains set in the "Rocky Mountains". To prove this fact he brought a short ragweed flower in the White Mountains and when he smelt it, he presented a rhinitis, it was the same observation for his son and his father. Phanerogam, dicotyledon, annual plant, monoic: Ambrosia artemisiifolia L. (Aa) were introduced from North-America to Europe at the beginning of the twentieth century. In Europe, four naturalized species and one native could be considered as source of Ambrosia-type airborne pollen. In France, fields are covered with Aa (common/short ragweed) in the mid Rhône Valley, Ambrosia trifida L.(At) is identified in South-West crops, A. psilostachya DC (Ap) is now identified in Camargue, A. tenuifolia Sprengel (Ate) introduced in Sète in 1839 is in Porquerolles. A. maritima (Am), the native species, is found on the marine sands of the Mediterranean region. A pollen is the main trigger of rhinitis (90%), conjunctivitis (75%), asthma (50%) and sometimes of dermatitis and of the oral syndrome (Kennel thesis 1987). The first symptoms generally occur simultaneously. Pollinosis, now called seasonal allergic rhinitis by the international terminology, includes other symptoms, so we use the term A. pollinosis. The characteristics of

A pollinosis are severity, duration -from August to September (recently from the end of July to the start of October)- and the presence of asthma and/or tracheitis in about 50% of cases. Bronchial symptoms generally appeared a few weeks after nasal symptoms. The impact of A pollen differs among patients: sometimes they are mono-sensitized, the disease starts late in life, no personal or family history of atopy is found, the titration of serum total immunoglobulin levels (total IgE) is within the normal range, sometimes they are poly-sensitized, the disease starts earlier in life, personal or family history of atopy is found, total serum IgE are higher than in the other group. In both groups, the clinical reactivity is similar; skin tests are positive in 93% of cases and a positive correlation between skin test and IgE specific titration (Aa) are observed in 85% of cases (Kennel). The severity of symptoms and the appearance of asthma are in both groups correlated with A pollen counts values. Moreover, in Lyon, about 65% of sensitized patients to A are sensitized to Artemisia. During the A pollination there is no other allergenic pollen except Artemisia (only at the start and the end of the A pollination): so diagnosis is easy. It must be confirmed after the A pollination season by a skin test and an IgE specific titration: it is dangerous to do a skin test during the symptoms period. The French Foundation For Ragweed Study (AFEDA) applies a predictive model of A pollination for improving care for allergic patients, thanks to a sensitive pollen monitoring trap always set at 3m high. This Cour pollen trap is important because for certain patients threshold values for clinical symptoms development is of some pollen grains/m<sup>3</sup>, about 5 (Déchamp et al 1995). In contrast to other pollen types, A pollinosis tends to be more prevalent in suburbs than in towns (Harf and Déchamp 2001). In Europe, from 1987 to 2010, different kinds of publications may be distinguished: studies where authors tell about the % of Apollinosis treated in their unit, official evaluations without epidemiological study, clinical/biological hypersensitivity studies, real epidemiological studies (in the Milano area and in the mid Rhône valley). No works seems published about the allergy impact of every *A* identified in Europe except Aa, but numerous allergens have been identified for some years into Aa, At, Ate. Some of them presents a cross-reactivity. These works bring a proof of the allergenicity of these pollen that is mentioned in non scientific informations in Internet. The other Ambrosia species seem have not been studied. In conclusion, as, now, European Union is interested in ragweed subject, it would be important to realize prevalence studies in each polluted country with a common protocol.

# Allergia al polline dell'Ambrosia in MEDICINA VETERINARIA

### N. FURIANI

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Le malattie allergiche sono comunemente riportate negli animali domestici. Tra queste, le forme atopiche sono ad oggi ben riconosciute nel cane, nel gatto e nel cavallo. Le loro manifestazioni cliniche variano da specie a specie: nel cane la più comune è la dermatite atopica (DA), spesso associata a congiuntivite, mentre nel gatto come nel cavallo possono osservarsi sia forme cutanee che respiratorie, in alcuni casi associate tra loro. Il ruolo dell'ambrosia nella patogenesi di queste malattie è stato solo marginalmente studiato. I dati più consistenti si hanno nel cane, mentre nessuna informazione specifica esiste ad oggi per il gatto. Riguardo al cavallo, i pochi studi disponibili in letteratura sembrano indicare che l'ambrosia abbia un ruolo limitato sia nelle forme cutanee, come la DA e l'orticaria ricorrente, che in quelle croniche respiratorie. La prevalenza della sensibilizzazione all'ambrosia nei cani atopici sembra essere correlata con il grado di diffusione della pianta nelle varie aree geografiche. Studi

eseguiti nel Nord America, in cui l'ambrosia è nativa, riportano una prevalenza elevata, variabile dal 35% al 59%, sia verso allergeni testati singolarmente che in gruppo (2). Studi europei degli anni '90 mostrano valori contenuti tra 7,7% (Grecia) e 9,2% (Francia) (2). Mentre studi recenti riguardanti il Nord Italia, soprattutto la provincia di Milano, e la Serbia, aree in cui l'ambrosia è ad oggi molto diffusa, rivelano che A. artemisiifolia è il secondo allergene maggiormente coinvolto nella DA canina con prevalenze del 21,6% e 66%, rispettivamente (3,1) In uno studio del 2006 sulla DA canina in Ungheria, Artemisia vulgaris è risultato l'allergene pollinico con più risposte positive (7,8%) diversamente dall'ambrosia (<4%). Come in umana, così anche nei cani atopici spesso si osserva positività concomitante tra ambrosia e artemisia nei test allergologici ma se ciò è dovuto a cosensibilizzazione o cross-reattività non è stato indagato. I maggiori allergeni di A. artemisiifolia interessati nella DA canina sono stati da poco identificati da Ognjenovic e colleghi e corrispondono agli isoallergeni del gruppo Amb a 1, gli stessi riconosciuti nell'uomo.

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## Weed pollen-allergy in Austria: crossand co-sensitization against ragweedand mugwort pollen

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In Austria, about 20% of pollen-allergic individuals suffer from weed pollen allergy. Before 1990, mugwort (Artemisia vulgaris) pollen was the main cause of seasonal allergy in late summer. In the 1990's ragweed (Ambrosia artemisifolia) plants have been brought in from Hungary, and since 2005 the number of reports on ragweed plants tremendously increased (1). With the presence of these plants, the ragweed pollen load and the prevalence of ragweed pollen allergy increased (2). To fight the threat of uncontrolled ragweed expansion and allergic sensitization, measures were initiated during the last 10 years and financed by national and federal agencies. Projects were started to investigate ragweed biology, geography and eradication strategies. Agricultural products potentially containing ragweed seeds became controlled, and the general awareness was increased by information campaigns. Studies revealed that from 1997 and 2007 neither the total pollen counts nor the prevalence of ragweed sensitization increased to the extent that the plant reports had suggested. During the last 6 years the total ragweed pollen load remained relatively constant. Interestingly, the pollen season also became shorter. Thus, the relevance of ragweed pollen allergy in Austria for the moment may be rated "intermediate". A major problem in the assessment of ragweed allergy arises from cross-reactions with mugwort pollen allergens. As the ragweed pollen season overlaps with the mugwort pollen season, diagnosis using pollen extracts is especially difficult in areas with exposure to both pollen species. In 2004, relevant mugwort and ragweed-pollen allergens became available by molecular cloning and were useful tools to address the question of cross- versus co-sensitization in patients

allergic to both allergen sources at the IgE- and T cell level. We found that allergy to mugwort- resp. ragweed pollen is characterized by the major allergens Art v 1 and Amb a 1. Neither allergen essentially contributes to the cross-reactivity of the two pollen species. Hence, by assessment of IgE against Art v 1 and Amb a 1 crossvs. co-sensitization to both weeds can be discriminated and has implications for specific immunotherapy.

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# Sessione III



# Gli interventi di sanità pubblica

# Moderatori: Anna Tosi, Paolo Bottero

# Allergia all'ambrosia: 15 anni di prevenzione nell'ASL Milano 1

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Nel nord-ovest della Provincia di Milano Ambrosia artemisiifolia è conosciuta dai botanici fin dal 1940 come pianta esotica naturalizzata. Agli inizi degli anni '80 era presente solo sporadicamente e sostanzialmente sconosciuta dal punto di vista allergologico. Dagli anni '90, a seguito di alterazioni dell'equilibrio ecologico, si è diffusa enormemente ed oggi questa zona è una delle più inquinate da polline di Ambrosia in Europa. La conseguenza in termini di salute della popolazione è l'elevata quota di soggetti ad essa allergici, con conseguenti elevati costi diretti ed indiretti associati. I dati più recenti evidenziano che Ambrosia è la principale causa di pollinosi nella zona e che ogni anno a circa 3.250 persone viene diagnosticata questa allergia, pari al 54% delle pollinosi ed al 38% di tutte le allergie respiratorie diagnosticate. Nel 2013 almeno il 16,4% della popolazione residente era allergica all'Ambrosia (risultati preliminari), con un trend in aumento ed una elevata percentuale di asmatici (circa il 40%). La diffusione della pianta ha avuto quindi un grande impatto sulla salute della popolazione della zona, configurandosi come un vero e proprio proble-

ma di salute pubblica ed appare evidente come si siano resi necessari interventi di prevenzione primaria a tutela della salute dei cittadini. Con questa finalità, la Regione Lombardia nel 1999 ha emanato un primo provvedimento che prevedeva l'esecuzione di tre sfalci consecutivi antecedenti la fioritura, ma la cui applicazione si è rivelata problematica (per la possibilità di utilizzare un solo metodo di contenimento e per la numerosità degli interventi da eseguire secondo la periodicità prevista). L'ASL Milano 1, in collaborazione con Regione Lombardia e Provincia di Milano, ha quindi condotto uno studio sperimentale quadriennale, dimostrando l'efficienza e la fattibilità di diversi metodi di contenimento dell'infestante (uno o due tagli antecedenti la fioritura, in relazione al tipo di area; pacciamatura; aratura; diserbo), alla base delle attuali indicazioni regionali. Le altre misure preventive implementate dall'ASL prevedono il controllo del territorio, attraverso il monitoraggio aerobiologico e la vigilanza sulle aree infestate, la collaborazione e consulenza ai Comuni e gli interventi di informazione e di educazione sanitaria alla popolazione, questi ultimi fondamentali per rendere accettabili gli interventi di contenimento proposti. Il risultato di questa strategia di prevenzione primaria, improntata alla flessibilità e all'integrazione degli strumenti a disposizione, è stato positivo, in quanto si è tradotta in un trend in diminuzione delle concentrazione di polline aerodiffuso di Ambrosia, specialmente nelle zone inizialmente meno infestate. Trend che nel 2013 è stato favorito anche dalla diffusione accidentale di un coleottero, Ophraella communa, che si ciba preferibilmente proprio di questa pianta. Gli effetti di questo insetto saranno studiati approfonditamente nell'ambito della COST ACTION EU "SMARTER" (Sustainable Management of Ambrosia artemisiifolia in Europe), cui l'ASL partecipa attraverso un proprio rappresentante. La collaborazione internazionale, e nazionale, è infatti essenziale per lo sviluppo della ricerca, lo scambio delle informazioni, la definizione di una strategia e di una legislazione comunitaria, ma anche utile per risolvere le questioni aperte a livello locale (ad es. la necessità di una diversa forma di provvedimento regionale o nazionale).

## Public health interventions against ragweed in quebec: examples from the monteregie region

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In the province of Québec (Canada), wherever ragweed (Ambrosia artemisiifolia) is abundant, close to 18% of the population suffers from allergic rhinitis linked to pollen dispersal. To address this situation, more than 15 years of public health interventions have been implemented in the province of Quebec. Federating these actions becomes more important considering the context of climate change, as health effects of ragweed are expected to increase in prevalence, severity and duration. In particular, the Table québécoise surl'herbe à poux (TQHP), a ministerial Task force initiative gathering experts and stake holders from different fields (public health, environment, transportation and agricultural ministries) endorses the promotion of a collaborative action at various territorial scales. In this context, the Montérégie Department of Public Health conducted an epidemiological study that included a

4-year follow-up of 400 allergic volunteers in order to support evidence-based decisions regarding the current management of ragweed. Results showed that a concerted control of ragweed induced a significant reduction in plant density, pollen concentrations and allergic rhinitis symptoms. A cost-utility analysis performed on these data estimated the costs of concerted control of ragweed at CDN\$60,603 with an associated gain of Quality Adjusted Life Year 10.15 (QALYs). This leads to an incremental cost-utility ratio estimated at CDN\$5,744/QALY. Compared to the threshold generally accepted in Canada for health intervention (CDN\$20,000 -100,000 /QALY), the concerted control of ragweed is a sound cost-effective strategy. In addition, a cartographic project has been developed in three different regions in Québec. It aimed at predicting the presence of ragweed depending on

the land use. Some tendencies arising from this project provided insights for municipalities with limited resources to identify priority areas for intervention. This is particularly relevant since municipalities play an increasing role in the implementation of healthy living environment. These promising data have to be complemented by a structured framework of knowledge translation. To adapt promising and evidence-based practices to specific territorial context, involvement of the different levels of government (provincial and local) is essential to support all regions in Québec. Hence, numerous activities (scientific webinars, technical webinars, day of continuing training, etc.) have been offered and unfolded to public health practitioners and municipal officers in order to share tools leading to an integrative strategy of ragweed management.

# Sessione IV



# La prevenzione del futuro

# Moderatori: Giuseppe Frenguelli, Maira Bonini, Claudio Ortolani

### Come controllare la pianta infestante *Ambrosia artemisiifolia* in Italia mediante un approccio biologico-integrato

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Ambrosia artemisiifolia è una pianta infestante di origine americana che è stata introdotta accidentalmente in Europa nella prima metà del secolo scorso. L'impatto di questa infestante è legato a due tipi di danno, sia medico (allergie) che agronomico. L'infestante è presente sul territorio nei campi coltivati ma predilige habitat disturbati come zone rurali e campi incol-Un controllo di tipo agronomiti. co/chimico (erbicidi), sostenibile nel caso dell'infestazione di campi coltivati, risulta improponibile quando la pianta infestante è presente nelle aree incolte vicine ai centri abitati. Pertanto, un programma di controllo territoriale di Ambrosia artemisiifolia necessita dell'attuazione di una strategia multidisciplinare a basso impatto ambientale, che preveda l'integrazione

di interventi di lotta biologica associati ad altre strategie di lotta integrata. La lotta biologica prevede lo studio e il successivo utilizzo di organismi (generalmente insetti o acari) specifici, ovvero in grado di alimentarsi e riprodursi unicamente sulla specie vegetale da combattere. Tali organismi specifici hanno ovviamente la stessa origine geografica della pianta infestante (in questo caso Nord America): pertanto i primi studi di lotta biologica di tipo classico si basano sul censimento delle specie fitofaghe specifiche presenti sulla specie da combattere nelle sue zone di origine. Per quanto riguarda Ambrosia artemisiifolia, nella lista de gli organismi fitofagi associati alla pianta, il coleottero Ophraella communa è ai primi posti sia per la sua biologia che per il tipo di danno che

procura alla infestante. Pertanto, in un contesto di lotta biologica di tipo classico, la presenza accidentale sul territorio della Lombardia del coleottero *Ophraella com-muna* (1) permette di valutare con studi di pieno campo l'efficacia di questo specie fitofaga su *Ambrosia* (e sulla sua produzione di polline), sia da sola che in combinazione con altre strategie di lotta a basso impatto ambientale.

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## SUSTAINABLE MANAGEMENT OF AMBROSIA ARTEMISIIFOLIA IN EUROPE (COST FA1203-SMARTER): OBJECTIVES, RECENT ACHIEVEMENTS AND FUTURE OPPORTUNITIES

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The EU-COST Action FA1203 on "Sustainable control of Ambrosia artemisiifolia L. (Asteraceae) in Europe (SMARTER)" was launched in February 2013 and will last for four years. Thirtythree countries have already signed the Memorandum of Understanding and over 180 researchers with specialists in weed research, invasive alien species management, ecology, aerobiology, allergology and economics are registered participants of SMARTER. COST Actions interlink nationally funded research projects and enable and finance conferences, working groups, training schools and research exchanges. SMARTER aims to initiate and develop long-term and sustainable control methods, to integrate these into existing mechanical and chemical control measures, and to quantify the success of these measures both for agriculture and health. The focus is on biological control methods with insects and fungi (especially using alien species from the area of origin of Ambrosia) and vege-

tation management to achieve a competitive plant cover. The envisaged outcomes of SMARTER will allow the various stakeholders to select optimal habitatand region-specific combinations of control methods. After an introduction and overview of the structure and the state of the Action, we briefly describe two planned activities typical for our Action, a study on the population dynamics of Ambrosia in different climates and habitats in Europe as a basis for estimating the efficiency of control measures, and an interdisciplinary study to clarify the impact the of North American native Ambrosia leaf beetle Ophraella communa (Coleoptera: Chrysomelidae) recently found in southern Switzerland and northern Italy (1, 2). SMARTER is an ideal framework to respond quickly to the recent establishments of O. communa in Europe and to collect the data that will help decide whether this event should be considered as a troublesome introduction of an alien invertebrate that causes damage to crops or native plant species, or whether it is likely to become

the first case of a successful biological control of an invasive weed in Europe. Finally, we will briefly present the newly launched iPhone App "The SMARTER Ambrosia Reporter", which mainly serves to report the occurrence of Ragweed when you see it around. It also features a version for professionals, including more detailed information about the location and the vegetation.

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## A population dynamics modelling approach for the management of ragweed – a case study of a potential biocontrol agent in northern Italy

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Classical biological control is one of the management strategies of ragweed investigated within the EU-COST Action SMARTER "Sustainable management of Ambrosia artemisiifolia in Europe". This method constitutes the introduction of highly specific natural antagonists from the native range of the weed. For such introduction to be safe and efficient, both the potential risks and impact, respectively, need to be assessed beforehand. To prospectively evaluate the potential impact of biological control agents on the long-term dynamics of ragweed populations, and in the wide range of environments throughout Europe, a modelling approach could be useful (1). Our research

focusses on the development of a population dynamics model of ragweed that allows such evaluation. We first outline how we collect data on ragweed populations to construct the model. Since the ragweed leaf beetle, *Ophraella communa*, was recently found to have established south of the Alps in Northern Italy and Southern Switzerland (2, 3), we then focus on this case study to exemplify how the impact of this potential biocontrol agent will be assessed using the model. We finally show that the same method can be used to evaluate other management strategies, such as vegetation management.

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