1.) Invasive pest: honey locust gall midge, (*Dasineura gleditchiae*) (Diptera: Cecidomyiidae)

The ornamental trees in urban public areas have been threatened by a growing number of invasive pests, spreading like epidemy, covering large areas in short time, resulting in conspicuous damages. In selecting target species for this study, we were focusing on pests of yet unknown chemical communication, preferably belonging to a taxon, which is only very poorly studied so-far in this respect so, although posing a challenge, offering remarkable new results. It was also considered whether the pest has recently been causing spectacular damage in our region, and therefore there is a demand / market for our expected results for practical application.

Taking these in account, we selected the honey locust gall midge, (*Dasineura gleditchiae*) (Diptera: Cecidomyiidae). Despite of many agricultural and horticultural pests in this group word-wide, only recently appeared the first pioneering publications of pheromone identifications in the family of Cecidomyiidae. Compounds representing unique chemical structures in insect pheromones, with importance of chirality in evoking behavioral activity, combined with the fact that females produce them only in ultra low amounts contributed to the difficulties which hampered break-though till the most recent times. Extremely short flight periods and relatively small body size may also be added to these difficulties.

Material and methods:

At first, basic observations were carried out to reveal that in which part of the day adults appear, female pose calling posture (emit pheromone), mate, and lay eggs. These observations were conducted in alleys of honey locust trees, *Gleditsia triacathos* cultivar. "Sunburst", at various places in Budapest (Palota-sétány, castle district, and Lágymányosi-híd).

Volatiles (pheromones emitted by calling females) were collected by a closed-loop stripping apparatus (CLSA, Brechbühler AG, Schlieren, Switzerland) on charcoal filter, in our laboratory. Several collections were completed, that one which resulted the break-through was made from 2300 calling females. The filter was rinsed in *n*-hexane. Activity of the extracts were checked by electroantennography (EAG) (Syntech), and behavioral tests.

Extracts were analysed by a gas chromatograph coupled to an electroantennographic detector (GC-EAD) (6890 N GC, Agilent Technologies Inc., Santa Clara, DB-wax capillary column, J&W Sci.), in our laboratory.

Chemical structure elucidation was performed using a GC-MS system, focusing on the the compound which had evoked antennal response in GC-EAD (based on RT and RI of the antennally active compound). This study was completed in an international, interdisciplinary cooperation with Prof. David Hall (NRI, Univ. Greenwich, Chatham Maritime, UK).). Enantiomer-specific synthetic route of the stereoisomers of the identified compound was also developed by this Group.

Field traping tests using synthetics were conducted in Budapest, using delta-shaped, sticky traps (Csalomon® RAG, PPI ARC HAS, Budapest).

Satistics: ANOVA followed by Games-Howell *post–hoc* test, SuperAnova®, Abacus Concept Inc.

Results:

The antennally active component was identified as (2R,8Z)-2-acetoxy-8-heptadecane. This is a new structure for insect pheromones. Even more surprising is that the *R*-enantiomer proved to be active, which is in contrast to the chirality of the so-far identified, related structures in gall midges (Molnár et al., J. Chem. Ecol., 2009, 35: 706-714). Spectacular trapping results were obtained already at the first field test: average daily captures of males per trap reached several hundreds at peak flight (Lágymányosi-híd, Budapest).

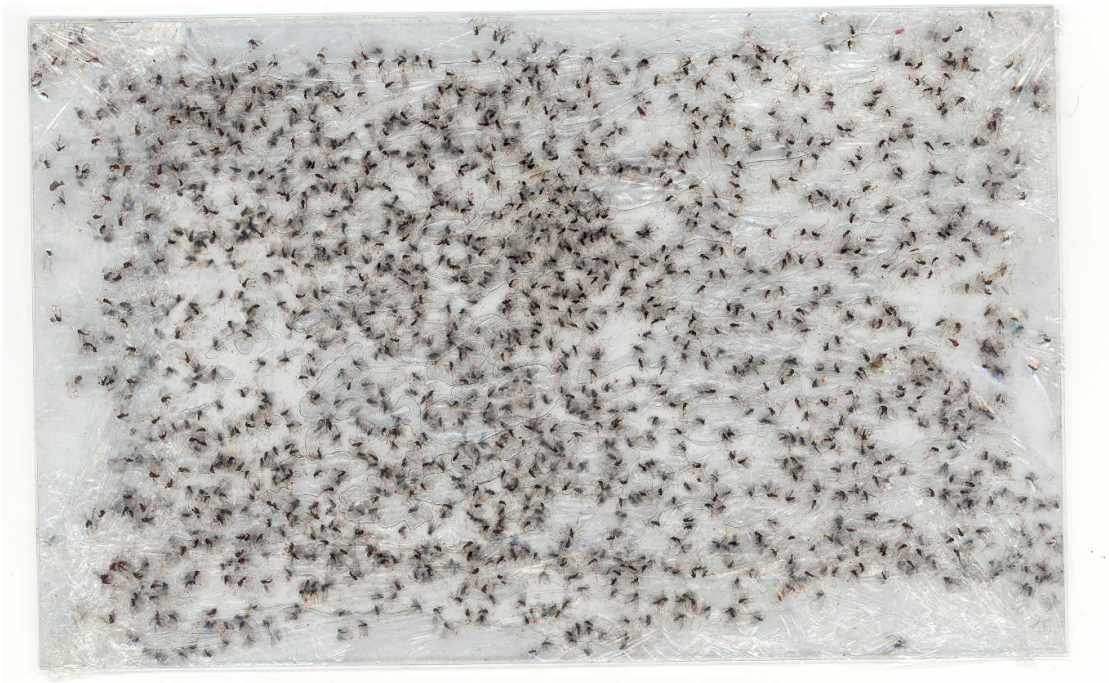

Photo: Daily captures of males of honey locust gall midge, on a 10 x 16 cm sticky layer of a delta trap (Csalomon® RAG), baited with 10ug of (2R,8Z)-2-acetoxy-8-heptadecane (767 males caught).

Our result has already been applied in practice: sticky pheromone traps for this pest have been commercially available in the frame of the extension service of the Department of Applied Chemical Ecology of our institute, as a new member of the Csalomon® trap family, from the subsequent year onwards of the appearence of our publication (2010).

With the help of these traps, we demonstrated the first occurrence of this expansive pest from Southern Sweden (Molnár et al., Entomol. Tidskrift 130: 113-120, 2009). A predatory bug, *Pinalithus (Orthops) cervinus* (Heteroptera: Miridae) (det. Előd Kondorosy, PhD) was found to prey upon *D. gleditchiae* in Sweden (Molnár et al., Plant Protection Forum, Keszthely, 19: 117-118, 2009). This predator is also present in Hungary.

Further details on racemic versus chiral composition of synthetic pheromone baits of traps for monitoring, as well as some practical intructions for conducting trappings for monitoring of this pest were also published (Molnár et al., Növényvédelem / Plant Protection, 46: 101-108, 2010, in Hungarian with English abstract).

Hungarian end-users were informed about our findings in the popular-science journal „Kertészet és Szőlészet" (Horticulture and viticulture) (Molnár and Szőcs, Kertészet és Szőlészet 60: 22-23, 2011).

These results constituted significant part of the PhD Theses of Béla Péter MOLNÁR (recently postdoc fellow, SLU, Alnarp, Sweden). In this connection, Béla Péter Molnár was awarded „Award for Young Scientists (Akadémiai Ifjúsági Díj), by the Hungarian Academy of Sciences, in 2011. Béla Péter Molnár obtained first a Hungarian grant (Magyar Állami Ötvös Ösztöndíj), then later a Swedish post-doctoral fellowship (Linné project) for two years to continue his studies on gall-midges in SLU, Alnarp, Sweden.

2.) Native pest: oak leaf-miner moth, *Tischeria ekebladella* (Lepidoptera: Tischeriidae)

This pest, native to our region, is recently causing growing problems in tree-nurseries, urban green areas, silviculture and also in chestnut orchards (*Castanea sativa*).

Material and methods were similar to that described before, with the exception of the techniques collecting the pheromone from calling females. Here ovipositor tip extracts were prepared from calling females (kept under reversed scotophase), pooling 10-100 ovipositors into an extract. The GC-EAD analyses were made on the same instrument (PPI ARC HAS, Budapest), while GC-MS measurement and synthesis were conducted by Prof. Wittko Francke (Inst. Org. Chem., Univ. Hamburg, Hamburg, Germany). Identification of mines (collected near to Budapest) and emerged adults, used in these studies, as well as of males captured during field tests (conducted near to Budapest) were verified by Dr. E. J. van Nieukerken (Naturalis Biodiversity Center, Leiden, NL) by external morphology, as well as of genitalia analysis.

Results revealed (3Z,6Z,9Z,19Z)-tricosa-3,6,9,-19-tetraene, as the sex pheromone of this species. This compound represents not only just a new structure in insect pheromones, but also a new natural product. Moreover, it is the first pheromone identification in the family of Tischriidae (Molnár et al., J. Chem. Ecol., 38: 1298-1305, 2012). In order to successfully complete the chemical identification, several structurally-realated tetraenes had to be synthetized and field tested, as the extremely rare situation was encountered that several tetraenes (isomers) had identical mass-spectra. Our result points also to interesting considerations in the evolution of pheromone systems in moths: polienic type of pheromones were probably independently evolved more than just one time in course of the evolution of Lepidoptera, appearing first in the ancient family of Tischriidae. As for practical relevance, the tetrane identified by us in this study can be used as trap bait for

monitoring of the pest. However, it has to be added that the complicated synthetic route hampers commercialization.

3.) Auxillary studies

Monitoring of Orthoptera *assablages in Budapest*
This survey had dual objectives. We wanted to see whether some „urban-desert" habitats may support build up of populations of some common locust pests, able to aggregate or even swarm-forming (posing a potencial risk for closely located agricultural areas) and, on the other hand, semi-natural reserves („natural islands") in Budapest are able to support survival of rare species, important in maintaining biodiversity. We found examples on both. *Calliptamus italicus* and *Dociostaurus brevicollis* were the two most dangerous pests found in several rural places in Budapest, while a new site of occurrence of *Acrida ungarica*, a rare, protected species of high value, was also found (Puskás et al., Plant Protection in Urban Area of Budapest, Budapest Meteropolitan Horticultural Corporation, Abstracts, 2009, in Hungarian).

Abundancy of the sycamore leafminer, Phyllonorycter platani *(Lepidoptera: Gracillariidae) and sycamore lace bug*, Corythuca ciliata *(Hemiptera: Tingidae) in cultivars*
Survey conducted in course of 2009-2011 in an ornamental tree nursery (Tahi) revealed that out of the five studied sycamore tree cultivars *Bloodgood* and *Columbia* cultivars, both resistant against *Gnomonia,* were less infected. However, the increasing level of infestation in course of the studied period is a warning sign that further studies are needed (Molnár et al., Növényvédelem / Plant Protection, 48: 147-152, 2012, in Hungarian with English abstract).

Timing of pesticide application against the horse chestnut leafminer, Cameraria ohridella *(Lepidoptera: Gracillariidae)*
Based on evaluation of our earlier-collected, however, yet unpublished sub-set of data, we corroborated our own earlier results that this urban pest can be controlled by a single control measure, timed in spring at the start of the mass-flight of moths, as monitored by large-capacity funnel type of pheromone traps (Szőcs et al., Növényvédelem / Plant Protection, 47: 248-250, 2011, in Hungarian with English abstract). Also, the second and third applications of pesticides, still widely practiced by others, can be spared so that the aesthetic value of tree can satisfactorily be protected for the entire season. By this way not only the pesticide load to the environment could be greatly reduced, but also the expenses of the unnecessary spraying spared. An article, popularizing our result, estimated that ca 50 milion HUF (ca 160-170 thousand EURO) can be spared yearly, only in the public domains of Budapest (Magyar Mezőgazdaság / Hungarian Agriculture, 65: 24, 2010 január 6, in Hungarian).

A shorter homologue, synergizing the sex pheromone of the horse chestnut leafminer, Cameraria ohridella *(Lepidoptera: Gracillariidae)*
Further data, collected during this project, corrobarated our own earlier results that a shorter homologue, designed by us earlier, synergizes the sex pheromone of *C.*

ohridella (Szőcs et al., Interntl. Soc. Chem. Ecol. 28th Ann. Meeting, Abstracts, p 239, 2012, Vilnius, Lithuania).

Beyond the primer aim of achieving new scientific results, we took emphasis also on disseminating of the relevance of our findings to the public. Our papers in public science journals, as well as lectures, given mostly on invitation (see e.g. meetings organized by the Budapest Metropolitan Horticultural Corporation) are indicating this.