

**COST Action IPLANTA
WG1 Meeting**

**‘Advance delivery strategies for dsRNA
into cells’**

**Ghent University
Ghent, Belgium**

5th Sept 2019

The major target species for RNAi-based pest control are insects, plants, viruses and fungi. Previous research has demonstrated that several barriers related to cellular uptake and endosomal release could impact their sensitivity to RNAi. Hence, understanding the cellular uptake of dsRNA is mandatory and crucial to the development of RNAi-based technologies. Additionally, this WG1 meeting will coordinate the collection and generation of a database, containing information about dsRNA sequences used for RNAi studies in plants, viruses, fungi and insects.

Thursday 5th Sept 08.30 -18.00

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| 08.30 – 08.50 | Registration/ tea/ coffee |
| 08.55 – 09.00 | Welcome and introduction by Guy Smagghe (Ghent University) |
| 09.00 – 09.40 | Nanocarriers for improved delivery of dsRNA into insects. Olivier Christiaens (Ghent University, Belgium) |
| 09.40 – 10.10 | Extra- and intracellular barriers for the delivery of nucleic acids in mammalian cells. Stefaan De Smedt (Ghent University, Belgium) |
| 10.10 – 10.35 | Tea/Coffee break |
| 10.35 – 11.05 | Repurposing cationic amphiphiles for improved cytosolic delivery of RNA therapeutics. Koen Raemdonck (Ghent University, Belgium) |
| 11.05 – 11.35 | Sucking of siRNA by <i>Macrolophus</i> bugs. Michel Ravelonandro (French National Institute for Agricultural Research, France) |
| 11.35 – 12.05 | RNAi for pathogen resistance in major woody fruit species. Silvia Sabbadini (Marche Polytechnic University, Italy) and Elena Baraldi (University of Bologna, Italy) |
| 12.05 – 13.05 | Lunch break |
| 13.05 – 14.05 | Generation of a dsRNA sequence database. Guy Smagghe (Ghent University) |
| 14.05 – 18.00 | Visit to Syngenta innovation center, Ghent |
| 19.00 | Dinner |



iPlanta



3rd COST IPLANTA TRAINING SCHOOL

‘Microscopy and studying cellular uptake of dsRNA for improved RNAi efficiency’

Ghent University
Ghent, Belgium

6th Sept 2019

The aim of the 3rd iPlanta Training School is to provide training and best-practice in visualising cellular uptake of fluorescently labelled dsRNA using confocal microscopy. One of the major target species for RNAi-based pest control are insects. Previous research has demonstrated that several barriers related to cellular uptake and endosomal release could impact insect sensitivity to RNAi. Further research investigating these fundamental mechanisms in various insect species is important to further elucidate these mechanisms. One of the most promising tools is using confocal microscopy to follow the fate of fluorescently labelled dsRNA. In this training school, the participants will receive the training, both theoretical and practical, in using this technology to investigate cellular uptake mechanisms and cytoplasmic release in cells. Moreover, recently developed polymer and liposome-

based nanoparticles will be used to demonstrate how these can improve cellular uptake of dsRNA. High qualified trainers from iPLANTA network will be invited.

All scientists/researchers/experts of the biotechnology area are eligible to submit an application for the iPLANTA grants. In the selection, priority will be given to Early Career Investigators (ECIs) and to the qualification of the curriculum. Grant applications should be made using the appropriate application forms in the Training School area (at: <http://iplanta.univpm.it/node/6>) before 9th August 2019. Unselected trainees can attend the school at their own expense.

Friday 6th Sept 08.30 -18.00

08.30 – 09.00 Registration/ tea/ coffee

Microscopy and advanced delivery of dsRNA into cells

09.00 – 09.40 Five minutes presentations from trainees about their research topics/interests

09.40 – 10.15 Confocal laser scanning microscopy: historical perspectives and recent developments. Andre Skirtach (Ghent University, Belgium)

10.15 – 10.30 UGent centre of expertise on advanced light microscopy. Herlinde Dekeersmaecker (Ghent University, Belgium)

10.30 – 11.00 Tea/Coffee break

11.00 – 11.35 Lectins and CPP-based dsRNA carriers. Kristof De Schutter (Ghent University, Belgium)

11.35 – 12.10 Tracking cellular uptake of dsRNA by fluorescent microscopy. Zarel Martinez (Ghent University, Belgium)

12.10– 13.30 Lunch

Laboratory practical session: Fluorescent microscopy for studying cellular uptake of dsRNA

13.30 – 18.00 Practical session: The use of fluorescent dyes/probes for labelling, detection and tracking of naked dsRNA, nanocarriers of dsRNA and cellular components, will be demonstrated. Furthermore, inhibitors of known cellular uptake mechanisms will be used to further illustrate what happens at the cellular level when dsRNA (either naked or complexed) is exposed to cells (extracellular to intracellular)