INTERPOPGER Project
Interactions between wild and artificial poplar stands and selection pressures associated with their management in French landscapes

V. Jorge, A. Dowkiw, M. Juteau, V. Guérin, P. Poursat, M. Villar, C. Bastien [1]
P. Frey, J. Pinon, S. Duplessis, A. Kohler, B. Barres, C. Rinaldi, F. Martin [2]
P. Faivre-Rampant, A. Bresson [3] C. Plomion, C. Lalanne [4] E. Klein, D. Allard [5]

[1] Unité Amélioration, Génétique et Physiologie Forestières, INRA Orléans, BP20619, Ardon 45166 Olivet Cedex France.
[2] UMR INRA-UHP1136 Interactions Arbres/Micro-organismes, INRA Nancy, 54280 Champenoux, France
[3] Unité de Recherches en Génomique végétale, INRA Evry, 91057 Evry Cedex, France, [4] UMR BIOGECO 1202 Biodiversité, gènes et écosystèmes, INRA Pierroton, 33612 Cestas cedex, France, [5] UR Biométrie Domaine St-Paul, INRA Avignon, site Agroparc, 84 914 Avignon cedex 9, France

Introduction

In French landscapes, **black poplar** (*Populus nigra* L.) can be found in three contrasting, and presumably interconnected, ecosystems . Originally, *P. nigra* is a major species of the **riparian forest**. It is also widely used in **monoclonal plantations**, mostly through *P.deltoides* x *P. nigra* interspecific hybrids. Last but not least, *P.nigra* is widely represented in **rural and ornamental landscapes** by Lombardy poplar, undoubtedly the most famous black poplar clone for its columnar growth.

INTERPOPGER project aims at evaluating the impact of cultivated or ornamental *P.nigra* genetic pools on the genetic diversity of wild **black poplars**, but also on the genetic diversity of the associated pathogen ***Melampsora larici-populina* leaf rust (*Mlp*)**.

Objectives of the project are: **(i)** To identify molecular determinants of the poplar/*Mlp* interaction and developing pertinent markers ; **(ii)** To quantify gene flow from cultivated to wild poplars using neutral and adaptive markers; **(iii)** To identify biological and physical factors conditioning gene flow success through modelling gene flow using spatial information; **(iv)** To evaluate the evolutionary impact of gene flow on poplar/*Mlp* interactions.

Genomic tools

P. nigra

Regarding **neutral markers**, a large amount of SSR markers are available and localised on the Populus genome sequence and on genetic maps. Some of these markers have already been used to screen the national *P. nigra* French collection, but also the Lombardy poplar and some of the most cultivated hybrids (Fossati *et al.*, 2004). Other markers are available that were developed by the GEVES Institute for cultivar identification. Regarding **adaptive markers**, we will focus on SNPs in candidate genes involved in qualitative or quantitative resistance to *Mlp* (Dowkiw *et al.*, 2004; Jorge *et al.*, 2005; Rinaldi *et al.*, submitted). These genes will be identified both by expressional analysis of contrasted material, and by positional cloning. A first evaluation of polymorphism of candidate genes is being realised with the Ecotilling method (Comai *et al.*, 2004; Figure 2)

Mlp

SSR markers have been developed through genomic library enrichment (Barres et al., *in preparation*). The *Mlp* genome sequence will be soon available (2006, US-DoE), which will give direct access to **SSR and SNP markers**. Eight virulences are offering **phenotypic markers** that are under strong selection pressures by the cultivated hybrids and their qualitative resistance genes. EST sequencing and expression studies will give access to additional markers and candidates genes controlling virulence in *Mlp*.

Quantification of gene flow

P. nigra

Among the elements that contribute to gene flow, only **pollen flow** will be measured, through *in situ* paternity analysis. The study site (11.5 Ha, 1225 m x 180 m) is located along the Loire River at Saint Ay (France), and hosts 510 wild *P.nigra* trees, and 13 Lombardy poplars (Figure 3). Flowering phenology data collected in spring 2006 show that Lombardy poplar is among the earliest flowering trees, together with 25 *P. nigra* females and 64 males. Seeds have been collected on 99 females distributed over the site. Leaves of all studied females and all potential males have been collected, and will be genotyped to identify the set of SSR markers suitable for paternity analysis.

Mlp

The **spatio-temporal dynamics of epidemics** will be studied in the corridor valley of the Durance river. Our work hypothesis is that rust epidemics start in the upstream part of the valley, where larch (on which sexual reproduction of *Mlp* occurs) is also present, before spreading downstream through the continuous riparian *P. nigra* forest. Some cultivated stands containing different cultivars with qualitative resistance genes have been localized that could have an impact on this scenario. *Mlp* populations will be intensively collected along the valley in the wild and cultivated stand, and will be characterised using SSR markers and in terms of virulence composition.

Identification of factors driving gene flow and modelisation

Effective gene flow depends on several physical and biological factors that will be measured *in situ* and/or *ex situ*. Importance of the following **biological factors** will be evaluated: flowering synchronism, quantity and viability of pollen, pollen competition, quantity and viability of seeds, competition between seedlings, stress resistance. The main **physical factors** that we intend to study is the distance between males and females and the impact of wind. All the studied trees were consequently georeferenced and a GIS was developed. A **statistical approach** using spatial and genetic data will allow to characterise the long distance dispersion curve (Oddou-Muratorio *et al.*, 2005). Anisotropic models will be used to evaluate the impact of dominant winds.

Evolutionary and functional impacts in cultivated - wild interactions

Selection pressures exerted by hybrid cultivars on *Mlp* populations: thanks to a unique collection of more than 1000 strains collected since 1982 in cultivated and wild poplar stands, we will check the hypothesis that past deployments or breakdowns of qualitative resistances carried by commercial cultivars had an impact on the evolution of aggressiveness in *Mlp*. In a forward approach, we will evaluate the frequency of *Mlp* strains able to overcome some major resistance factors identified in our breeding material but not yet deployed in the field (Dowkiw *et al.*, 2004).

Impacts of cultivated poplars on wild poplar stands: Gene flow consequences will be assessed by studying the potential adaptive advantage of introgressed *P. nigra* genotypes. This will be measured through the relative rust resistance levels of (*P. deltoides* x *P.nigra*) x *P.nigra* versus *P.nigra* x *P.nigra* controlled crosses involving common *P.nigra* parents. Regarding *Mlp* flow consequences, aggressiveness of *Mlp* strains collected in wild versus cultivated poplar stands will be evaluated on a representative sample of the French wild *P. nigra* populations.

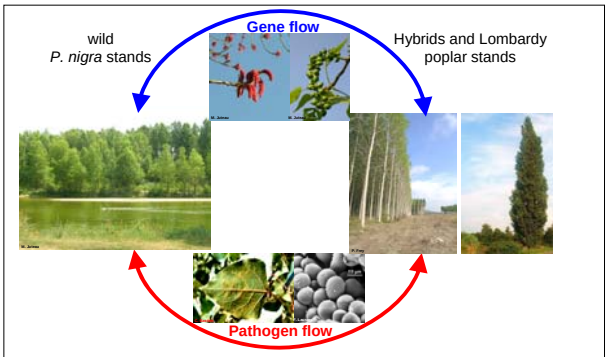

Figure 1: Poplar/*Mlp* interactions studied at the landscape scale in INTERPOPGER project

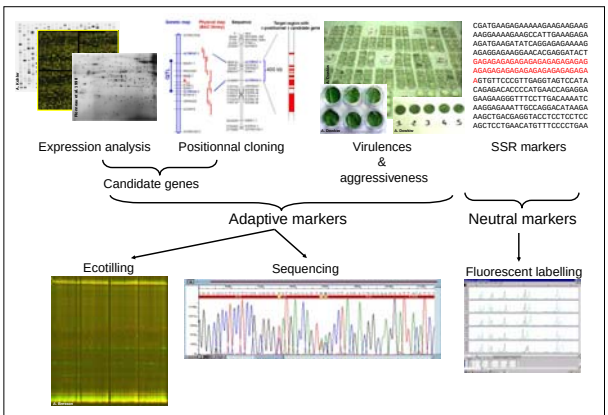

Figure 2: Integration of genomic tools

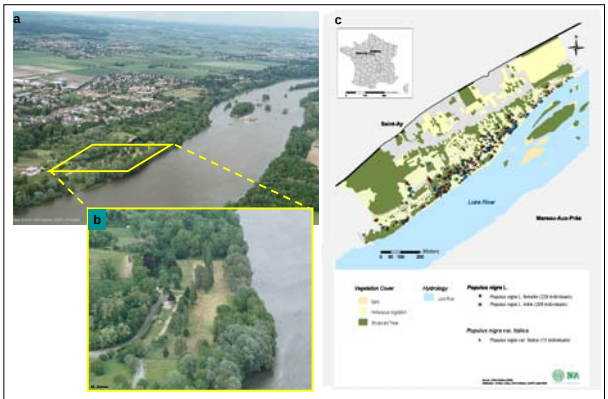

Figure 3: Saint ay study site: **a.** Aerial photography. **b.** Detailed view showing Lombardy *P. nigra* alignment. **c.** GPS localisation of wild *P. nigra* trees.

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