Reproductive Ecology of Willows along the Bowron River

Research Report 2006

Cecilia Alstrom-Rapaport

INTRODUCTION

Willows are an important component of many plant communities in the northern hemisphere and provide food and shelter to mammals, birds and insects. Many willows are pioneering species, establishing at primary succession on virgin substrates, following forest fires and on river floodplains. Genetic analysis based on molecular markers of pioneering plant populations, revealing their spatial and temporal structure, is important in the understanding of primary succession in general and forest regeneration in particular. In spite of their prevalence and ecological importance, only a few studies have explored the genetic population structure of willow species (Rottenberg et al. 1999, Lian et al. 2003). Although the reproductive biology has been examined for several willow species (e.g. Argus 1974, Kaul and Kaul 1984, Alliende and Harper 1989, Dawson and Bliss 1989, Douglas 1989, Fox 1992, Ottenbreit and Staniforth 1990, Peters and Totland 1999) few studies have applied a molecular ecology approach, combining information from life-history and reproductive characteristics with that of genetic molecular markers from the same natural populations.

My research goals are to assess the genetic diversity and genetic structure of willow populations (population genetic analyses of willows) and, in parallel, examine the pollination mode, sex ratio and frequency of clonal growth of the same populations (reproductive ecology of willows). Subsequently, as I combine these two approaches I will be able to make inferences on how these reproductive traits influence the genetic diversity and genetic structure (molecular ecology of willows).

As a first step I initiated a study to examine the reproductive ecology in lowlands of British Columbia, focusing on the pollination mode (insect and/or wind pollination), asexual reproduction and sex ratio. Six riparian study sites were established, two of these are located at Aleza Lake Research Forest (ALRF). This report summarizes results from these populations obtained in 2005. However, as this study now is part of a larger 5-year NSERC discovery grant funded project (2005-2010), I intend to continue to monitor and do research at these sites for several years.

MATERIALS AND METHODS

Study area and species

In 2005 six riparian study sites were chosen (Figure 1). In three of these locations the willow population numbers were so large that plots of 5 m x 5 m were established along the river banks approximately 50 meters apart. At the Bowron River study site within

ALRF, 10 plots were established. All individuals within each plot were labeled with individually numbered plastic tags. Species identity of individuals was determined based on reproductive, bark and leaf morphology. In three populations (Island Park, Camp Creek and Penny Access Road) (Figure 1) all individuals were included in the study. In all populations three leaves were collected from each individual for future population genetic structure analysis.



Figure 1. Locations of study sites of riparian willows. Sites studied in 2005 are indicated by blue triangles while additional planned study sites are shown by red squares.

Sex ratio

At the time of flowering (April-May) the sex of each tagged individual was determined and recorded. Deviations from 1:1 sex ratio for each species and study site were examined with Chi-square test.

Pollination mode

Insect excluders, made of plastic wire and 1x1 mm mesh cloth, were attached to branches of females just before flowering. This allows pollen carried by wind to reach the female catkins. Three weeks later the branches carrying the insect excluder as well as a branch from the same individual carrying catkins which had been allowed to pollinate by both wind and insects, was cut down and brought back to the lab. Catkins were dried and seeds were counted and compared for each pollination treatment. In addition, the viability of

seeds from each treatment was examined in a green house germination experiment. Seeds were placed in petridishes on moist filter paper and the number of seeds germinated was recorded after 24, 48 and 72 hours.

RESULTS

Species identification

Five different willow species (*S. scouleriana*, *S. lucida*, *S. lucida*, *S. prolixa* and *S. maccaliana*) were found to occur at Camp Creek. At this location the species identity of 202 individuals could be determined while the heavy browsing of beavers and ungulates made the identification of the remaining 31 individuals impossible. Population density was estimated to be approximately 0.02 individuals per m².

In addition to the five *Salix* species found at Camp Creek, a sixth species, *Salix sitchensis* was found along Bowron River at ALRF. At this location 152 individuals of these six species were labeled for 10 plots. However, in July 2005, 38 of these individuals had been so heavily browsed by beavers that either their tags could not be found (the tagged branch had been removed so that the identity of the individual was uncertain) or no remaining branches could be used for species identification. Estimates of population density at this site range from 0.16-1.04 individuals m².



Figure 2. Number of individuals and sex ratios of five willow species at Camp Creek.



Figure 3. Number of individuals and sex ratio of six willow species at Bowron River in ALRF.

Sex ratio

At Camp Creek the females outnumbered males for four out of five species (Figure 2) although only *S. scouleriana* had enough individuals permitting statistical testing (p<0.01). Along the Bowron river site at ALRF (Figure 3), no significant deviation from an even sex ratio for *S. scouleriana* was found while for *S. sitchensis* the sex ratio was skewed with 13 females to only one male (number too small to statistically test).

Pollination mode

Seeds from catkins within insect excluders and from open-pollinated catkins were obtained from all six study populations. As expected, fewer seeds per capsule were found on catkins within insect-excluders than on open-pollinated catkins (data now shown). However, only viable seeds were obtained from the Island Park study site. Surprisingly, seeds from the other five populations failed to germinate. Hence, no meaningful comparison can be made with respect to insect and wind pollination.

THOUGHTS AND FUTURE STUDIES

I expect that data from two additional field seasons must be gathered before publication. The species identification which is tricky in willows needs to be confirmed as the species did not completely correspond to those previously reported in the area. The lack of viable seeds within these populations must also be examined further. The poor germination may be a result of the early spring in 2005, willows flowered 2-3 weeks earlier than normal. This in turn may have had an effect on the synchronizing of the male and female flowering of the same willow species. The unviable seeds that were produced may be the result of crosses between two different species.

While conducting the above described study during the summer of 2005 and spring of 2006 I have come to realize that one important factor influencing the genetic structure of willow populations as well as their regeneration was omitted from my original proposal. This factor is beavers which cut down a majority of the willow individuals on a regular basis promoting clonal expansion through sprouting. However, more importantly, some of the branches they cut down are used to improve lodges or repair dams. I have seen that these branches can establish as clonal representatives at new locations (on top of the beaver lodge for example). Furthermore, I have also found evidence that show that some of the branches cut down by beavers may be dragged for a few meters and then abandoned with a subsequent establishment at the new location. Several branches tagged in the summer of 2005 were found in the spring of 2006 at new locations. Along smaller creeks these branches may not travel far, however, at sites along Fraser and Bowron Rivers the branches cut by beavers could potentially travel tens to hundreds of kilometers establishing new individuals with identical genotypes to those upstream. Hence, the continuous beaver cutting may not only influence the genetic structure at the intrapopulation level but also be an important source of downstream migration of willows.

Earlier studies have addressed the impact beavers have on the succession, abundance and dispersal of willows as well as poplars and aspen (Bailey et al. 2004, Baker et al. 2005). The beavers modify their environment by building dams that raise the water table and ponds that trap sediment. These alterations benefit willows as they create moist virgin substrates for seed germination and moisture regimes which increase seedling survival. This in combination with the sprouting of willows and the tendency of beavers to shift their activity upstream and downstream if necessary as the willows recovers from foraging suggest that the willow-beaver system could persist indefinitely within the same stream reach (Baker and Hill 2003). Baker et al. (2005) go so far as to suggest that the beaver and willow are mutualists.

I will address the questions regarding beaver-willow interaction focusing on the genetic structure of willows as well as the beaver impact on the sexual and asexual willow reproduction in future field seasons.

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