



Effects of Inbreeding on Whitebark Pine

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INTRODUCTION

Whitebark pine (*Pinus albicaulis*) is the only North American stone pine (genus *Pinus*, section *Strobus*, subsection *Cembrae*). It has a mutualistic relationship with the Clark's nutcracker (*Nucifraga columbiana*), which is its primary seed disperser. The foraging and caching behavior of the nutcracker results in clumps of related individual trees randomly distributed across the landscape. This clumpy growth habit leads to higher levels of inbreeding (both selfing and bi-parental) compared with other wind-pollinated conifers (Krakowski *et al.*, in press). The effects of this inbreeding on quantitative traits (inbreeding depression) as well as on genetic diversity are unknown. A previous study of whitebark pine observed an increase fixation index ($1-H_o/H_e$) with increasing latitude and decreasing longitude throughout British Columbia that corresponds to levels of white pine blister rust infection (cause by the fungus *Cronartium ribicola*). It has been hypothesized that blister rust may be the main selective force driving the observed trends in fixation index.

OBJECTIVES

Components of this study will:

- 1) quantify inbreeding depression in quantitative traits, and
- 2) evaluate the role of inbreeding in response to white pine blister rust infection

1) INBREEDING DEPRESSION

INTRODUCTION

A mating system study by Krakowski *et al.* (in press) revealed that whitebark pine has a mixed mating system, and the distribution of outcrossing rates across individual seed parents is bimodal (see Figure 1). A comparison of parental outcrossing rate, family inbreeding coefficient and progeny performance will be used to determine the magnitude and effects of inbreeding depression. This relationship may reveal whether or not whitebark pine has undergone purging of strongly deleterious alleles and evolved to withstand inbreeding without severe negative effects on fitness.

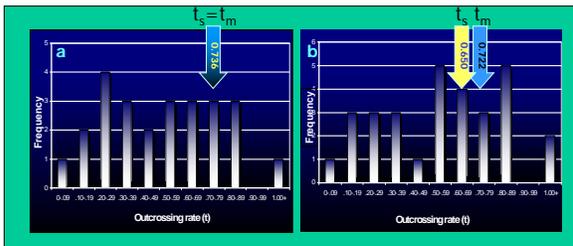


Figure 1. Distribution of family outcrossing rates from two sites; a) Manning Park and b) Mt. Baldy. Most inbreeding at Manning is due to selfing ($t_s = t_m$) while at Mt. Baldy, bi-parental inbreeding is occurring ($t_s < t_m$) (Krakowski *et al.*, in press)

MATERIALS AND METHODS

Progeny from ~100 different families, from three regions pine will be genotyped using isozyme analysis of diploid embryo and haploid megagametophyte tissue. The MLTR program (Ritland 1990) will be used to determine the mean outcrossing rate for each family from this data.

Quantitative traits will be measured on open-pollinated seedlings from the same seed parents growing in a common garden study. Regression analysis will be used to determine the relationship between parental outcrossing rate (independent variable) and individual quantitative traits (dependent variable).

Inbreeding depression (δ) for each trait will be calculated as:

$$\delta = 1 - e^{-Bx^F}$$

where F = family inbreeding coefficient ($1-H_o/H_e$)

B = slope of the regression of family trait mean on F



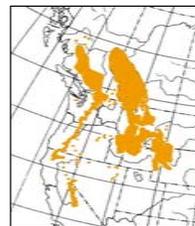
Clark's nutcracker



Clump of whitebark pine stems



Common garden bed



Species distribution of whitebark pine



Large mature whitebark pine

2) GENETIC DIVERSITY: INBREEDING AND DISEASE EFFECTS

INTRODUCTION

Many conifers that have an excess of heterozygotes at maturity are found to have an excess of homozygotes at the seed stage due to inbreeding. A previous study of the genetic diversity of whitebark pine observed a slight heterozygote excess in southeast B.C. populations ($F < 0$), but populations in northwest B.C. showed a heterozygote deficiency ($F > 0$) (see Figure 2). Trends have also been observed in the level of white pine blister rust in B.C., with infection levels highest in the southeast of the province (Stuart-Smith, 1998 and Zeglen, 2002) (see Figure 2). These trends may be indicating that blister rust is exerting an increased selective pressure against the more homozygous, inbred individuals.

MATERIALS AND METHODS

Isozymes will be used to determine genetic diversity of three age cohorts (6-month-old seedlings, young (5-30 year old) saplings, and mature trees) from 11 sites throughout B.C., WA and ID. A survey will be done on each site to assess the level of blister rust infection.

ANOVA will be used to examine differences in mean fixation index among cohorts and sites.

To investigate the effects of blister rust on genetic structure, regression will be used to examine association between the percent of trees infected on a site and fixation index within each cohort.

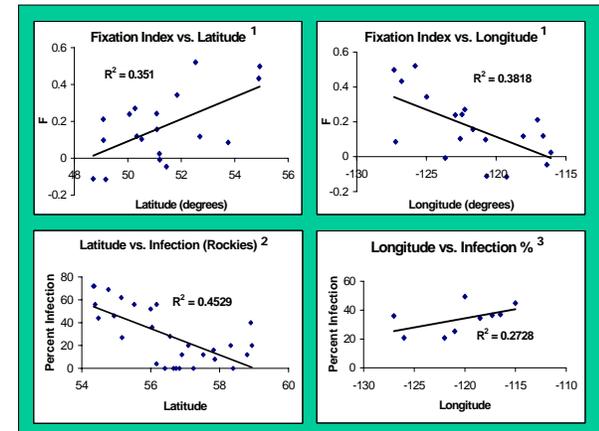


Figure 2: Geographic trends of fixation index and rust infection
^{1/} From Krakowski 2001 ^{2/} From Stuart-Smith 1998 ^{3/} From Zeglen 2002

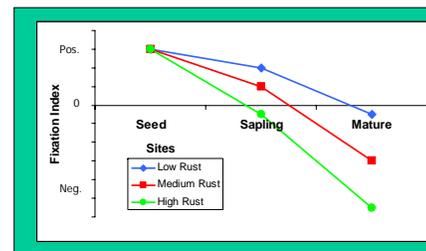


Figure 3: Predicted change in fixation index with age cohort and site mean level of rust infection

HYPOTHESES

Figure 3 illustrates the hypothesized results from the comparison of fixation index (F) among cohorts and sites. Within a site, F is expected to decrease with age, as less fit inbred individuals are removed from the population. Among sites, the difference between age cohorts is expected to be greatest for the mature cohort due to the length of exposure to rust. This would indicate that blister rust is exerting an additional selection pressure against more homozygous, inbred individuals.

REFERENCES

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