

Technical Study Plan: Comandra rust screening in Bulkley Valley lodgepole pine

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1.0 Introduction

1.1 Research question and objectives

There is evidence that genetic variation in resistance of lodgepole pine (*Pinus contorta* Dougl. var. *latifolia* Engelm.) to comandra rust (*Cronartium comandrae* Peck) exists (John 2001, 2003). The objective of this proposed research is to rank orchard parents so that custom seedlots with high rust resistance can be produced for deployment on high-risk sites. This project will focus on families in the seed orchard for the Bulkley Valley low-elevation seed planning unit [SPU 17 Pli BV low], at VSOC.

1.2 Background: literature review

Comandra blister rust is a disease that causes stem cankers on hard pines (Hiratsuka *et al.* 1995). In lodgepole pine this results in growth reduction, stem deformation and tree death (Johnson 1997, Geils and Jacobi, 1993). It has been described as causing significant losses in young lodgepole pine stands (van der Kamp and Spence 1987, Hiratsuka *et al.* 1988, Woods *et al.* 2000).

The rust has a complex life cycle and must parasitize a secondary host during one stage of its life cycle. The damaging effects of the fungus are limited by the distribution of its herbaceous hosts (Ziller, 1974). On dry open sites, the rust's alternate host is bastard toad-flax (*Comandra umbellata* (L.) Nutt. var. *pallida* (A. DC.) M.E. Jones). On moist, moderately open sites the alternate host is northern bastard toad-flax (*Geocaulon lividum* (Richards.) Fern. [syn. *Comandra livida*], family *Santalaceae* (MacKinnon *et al.* 1992)) which is a semi root parasite on other plants. In the Lakes TSA of BC, rust "hotspots" occur on the moister less open sites associated with northern bastard toad-flax. The rust basidiospores produced on these secondary hosts can only infect pines. These basidiospores have been described as "*the weakest link in the life cycle. They are small delicate, and unable to survive even a few hours of drought or high temperature ...*" (Hunt 1992). Comandra rust infections are thus only severe on sites where the alternate host (northern bastard toad-flax) is abundant and where environmental conditions are favorable around the time of sporulation from the alternate host. In the Lakes TSA, the alternate hosts are abundant, and humid and cool conditions occur frequently; this likely contributes to the severity of this disease in the Bulkley Valley SPU.

Previous analyses of comandra rust incidence in interior lodgepole pine [pli] progeny tests indicated that there is genetic variation in susceptibility to this pathogenic fungus with infection levels ranging from 0 to 69% by family on one site. A cluster analysis of infection rate by source revealed a geographic basis for susceptibility, with provenances from southeast BC showing high infection levels, and low infection levels from sources of intermediate latitude and longitude (John 2000, 2002, 2003). On the basis of these initial assessments, select rust resistant sources have been identified for use on high rust sites (Carlson, 2001).

1.3 Justification for study (*gap analysis*)

The Bulkley Valley (BV) progeny tests were designed to accurately estimate breeding values for growth, and testing for rust resistance was not an original trial objective; thus the trial design is not optimal for this purpose. In order to accurately rank the parents in the BV low SPU for rust resistance it is necessary to deploy adequate numbers of individuals per family on high rust incidence sites.

There appears to be considerable site-to-site variability in rust incidence. Only one of three BV progeny test sites (Chowsunket Lake) has shown a high level of rust infection. The confidence intervals on infection rate estimates on this trial are large, and the disease resistance ranking is uncertain. We can expect optimal expression of rust resistance on sites with reasonably high rust infection levels. We also expect smaller confidence intervals on resistance ranking with adequate numbers deployed under test, and appropriate field design.

Genetic selection for fusiform rust (*Cronartium quercum* (Berk.)Miyabe ex Shirai f.sp. *fusiforme*) resistance in slash pine (*Pinus elliotii* Engelm.) in the south eastern United States has lowered rust incidence from 50% to 24% on high rust hazard sites with a consequent increase in volume of 25.2 to 32.8% at ages 20 and 25 years. This translated as 40.2 to 89.8% value increases due to use of resistant stock (Brawner *et al.* 1999). It seems reasonable to expect that selection for resistance to the related comandra rust on lodgepole pine will also yield worthwhile gains.

1.4 Species and/or traits of interest

Species: Lodgepole pine from Bulkley Valley low elevation Seed Planning Unit.

Trait of interest: Resistance to comandra rust.

1.5 Overview: activities and timelines

Sow seed	Spring 2003
Select test sites (4-6)	Summer 2003
Confirm test sites (3-4)	Fall 2003
Site preparation	Fall/Winter 2003/ 2004
Site layout	Fall 2003 – Spring 2004
Trial establishment	Spring 2004
First assessment	Spring 2005
Second assessment	Spring 2006
Third assessment	Spring 2007
Fourth assessment / Analysis and ranking	Spring 2010

2.0 Materials

2.1 Test material

131 seedlots will be tested. Seedlots include orchard single-family collections, wild collections from original ortets, controlled crosses between putatively resistant parents, putatively susceptible seedlots, and operational (control) seedlots (see Appendix 1 for seedlot listing by category).

2.2 Physical test environment

Field tests will be established on 3-4 test sites. Initially, 4 to 6 sites will be chosen for maximum likelihood of rust infection. This will be determined by high infection levels in adjacent stands and by the presence of the alternate host, bastard toadflax (*Geocaulon lividum*). The site choice will be finalised on-site in the fall of 2003.

3.0 Methods

3.1 Experimental design

Rust susceptibility is assessed as a presence/absence trait, and can be expected to display a binomial distribution. In order to obtain a 95% confidence limit within 5% at low infection levels (approx. 5% infection) and within 10% at high infection levels (50% infection) a sample size of roughly 100 individuals is required. In order to end up with 100 well exposed plot trees, we propose to establish 3 test sites each with 50 replicates of single tree plots for all 131 seedlots under test for a total of 150 test trees per seedlot. Each test site will include 6,300 trees. The trials will be established at 1 x 1 m square spacing for a total of 0.63 ha per site. If seedling numbers allow, a fourth site may be established.

An alpha design, most likely designed with 12 sub-blocks of 11 plots (modified to 11 sub-blocks of 11 and one sub-block of 10, so seedlots total 132) seedlots, overlain with an interlocking design of single tree plots, will be used. Design will be finalised once seedling numbers are known.

3.2 Proposed data collection methodology

The trials will be fully assessed for presence of comandra stem galls in 2005, 2006, 2007 and 2010. It is likely that a simple presence/absence scoring will be used, but this may be modified, should variation in symptom presentation occur. Timing of assessment will coincide with maximum visibility (around the time of sporulation, in May).

3.3 Data storage/archiving

All data will be stored, at a minimum, at VSOC, Babine Forest Products, and IPF Ltd; data will also be made available to MoF staff.

3.4 Analysis procedures

Analysis procedures will be developed in consultation with MoF staff and academics.

4.0 Results and reporting

4.1 Expected benefits

- Users of seed from VSOC orchard 219 may be able to design custom lots for high rust sites.
- Users of lodgepole pine generally will benefit from a better understanding of genetic resistance

to rust.

- Users of improved seed in BC will benefit from better understanding of selection for disease resistance.

4.2 Applicability

Results will be:

- applicable to Pli breeding population for SPU Pli BV low and VSOC orchard 219 specifically;
- applicable to Pli breeding province-wide (methodology);
- applicable to conifer disease resistance breeding province-wide (methodology);
- applicable to rust resistance breeding in conifers generally.

4.3 Performance measures and indicators

- Test site short-list: 4-6 sites, # of sites on short list
- Test site verification and selection, # of sites selected
- Test sites prepared for planting, # of test sites prepared
- Tests established, # planted
- Test sites measured, # measured
- Results analysed, interpreted and reported, analysis reports, BV ranking for rust susceptibility, analysis report with family rankings.

4.2 Reporting schedule and recipients

Project plan (this document), Fall 2003; Establishment report, Fall 2004
Measurement reports, Fall 2005, Fall 2006, Fall 2007, Fall 2010
Analysis reports, Fall 2005, Fall 2006, Fall 2007, Fall 2010

All plans and reports will be distributed to Babine Forest Products, VSOC, and Ministry of Forests Tree improvement and Research branches.

5.0 References

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- Woods, A.J., A. Nussbaum, and B. Golding. 2000.** Predicted impacts of hard pine stem rusts on lodgepole pine dominated stands in central British Columbia. *Can. J. For. Res.* 30: 476-481.

Appendix 1. Seedlot listing: Vernon Seed Orchard Company Comandra Rust Family Screening Trial

- 131 seedlots sown
- 2 blocks of each family (112 cavities/block 410A)
- proposed: 3 (possibly 4) sites with up to 50 trees per seedlot per site (subject to seedling recovery)

Orchard o.p.seed

	Family	# seed (225 if blank)		# Blocks	Code
1	223			2	301
2	233			2	302
3	236			2	303
4	250			2	304
5	258			2	305
6	266			2	306
7	275			2	430
8	276			2	307
9	284			2	308
10	291			2	309
11	299			2	310
12	346			2	311
13	375			2	312
14	385			2	313
15	392			2	314
16	402			2	315
17	406			2	316
18	422			2	317
19	428	95	6	1	318
20	430			2	319
21	431			2	320
22	445			2	321
23	472			2	322
24	479			2	323
25	480			2	324
26	483			2	325
27	484			2	326
28	488			2	327
29	490			2	328
30	491			2	329
31	495			2	330
32	499			2	331
33	501			2	332
34	502			2	333
35	1630			2	334
36	1666			2	335
37	1730			2	336
38	1732			2	431
39	1733			2	337
40	1739			2	338
41	1740			2	339
42	1741			2	340
43	1745			2	341
44	1770			2	342

	Family	# seed (225 if blank)		# Blocks	Code
45	1773			2	343
46	1775			2	344
47	1779			2	345
48	1795			2	346
49	1799			2	347
50	1820	90		1	348
51	1822			2	349
52	F2001219			2	350
OP Seed from wild collections (no orchard seed available)					
1	224	225		2	351
2	275	225		2	352
3	327	225		2	353
4	428	225		2	354
5	478	225		2	355
6	1616	225		2	356
7	1618	225		2	357
8	1629	225		2	358
9	1679	225		2	359
10	1732	225		2	360
11	1736	225		2	361
12	1740	225		2	362
13	1772	225		2	363
14	1791	225		2	364
15	1792	225		2	365
16	1804	225		2	366
17	1809	225		2	367
18	1820	225		2	368
OP Seed (family also represented by orchard seed)					
1	223	225		2	369
2	233	225		2	370
3	250	225		2	371
4	258	225		2	372
5	276	225		2	373
6	299	225		2	374
7	346	225		2	375
8	375	225		2	376
9	385	225		2	377
10	392	225		2	378
11	402	225		2	379
12	406	225		2	380
13	422	225		2	381
14	430	225		2	382
15	431	225		2	383
16	472	225		2	384
17	479	225		2	385
18	480	225		2	386
19	483	225		2	387
20	484	225		2	388

	Family	# seed (225 if blank)		# Blocks	Code
21	488	225		2	389
22	490	225		2	390
23	491	225		2	391
24	495	225		2	392
25	1630	225		2	393
26	1730	225		2	394
27	1733	225		2	395
28	1739	225		2	396
29	1741	225		2	397
30	1745	225		2	398
31	1770	225		2	399
32	1773	225		2	400
33	1775	225		2	401
34	1779	225		2	402
35	1795	225		2	403
36	1799	225		2	404
37	1822	225		2	405
Operational Seedlots					
1	33201			2	406
2	33209			2	407
3	33216			2	408
4	39411			2	409
5	39544			2	410
6	42843			2	411
7	47310			2	412
8	47373			2	413
9	47391			2	414
10	47935			2	415
11	61033			2	416
12	61039			2	417
13	61041			2	418
Susceptible Seedlots					
1	44201			2	419
2	45786			2	420
Controlled Crosses with putative comandra resistance					
	PARENT_1	PARENT_2			
1	1809	1617		2	421
2	1809	1834		2	422
3	1610	1619		2	423
4	433	1617		2	424
5	433	1619		2	425
6	1618	1611		2	426
7	1618	1616		2	427
8	1809	1616		2	428
9	1829	1611		2	429
			count	260	131