Evaluation of Joint Linkage QTL Analyses for Partial Resistance to *Phytophthora sojae* Using Six Soybean Nested Inbred Populations

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Introduction

Joint linkage QTL analysis (JLA) is quantitative trait mapping strategy that uses multiple recombinant inbred line (RIL) populations, which are nested by one common parent. A possible issue in the application of JLA over multiple RIL populations is that individual experiments could be conducted with varying methods over time. So, this study aimed to evaluate the efficiency of JLA using six populations with heterogeneous experimental conditions. To test effect of heterogeneous assay conditions, JLA was conducted on RIL populations combined on the basis of the four scenarios outlined below. The QTL identified with JLA were compared to the results of linkage analysis (LA) in single populations.

❑ Four scenarios for combining populations

1. JLA limited to combinations of two RIL populations with the least confounding conditions
2. JLA with four RIL populations in which the generations of inbreeding differed and sets of SNP markers only partially overlapped
3. JLA with four RIL populations for which two different phenotypic assay methods were used to evaluate the resistance
4. JLA in six RIL populations with non-homogeneous phenotypic assays, differing inbreeding generations, and partially overlapping marker sets

Materials & Methods

❑ Heterogeneous conditions of 6 populations

Population				Methods		Scenario			
No.	Common parent	Resistance source	Generation (Pop. size)	SNP set	Phenotypic assay (traits)	*P. sojae* Isolates	1	2	3	4
OP1	OX20-8	PI 398841	F7:8 (305)	B	Tray test (Lesion length)	C2S1	+	+	+	+
OP2		PI 407861A	F7:8 (157)	B	Tray test (Lesion length)	OH25	+	+	+	+
OP3		PI 427106	F7:8 (367)	B	Layer test (Root dry weight)	1S11,OH30	+	-	+	+
OP4		PI 427105B	F7:8 (338)	B	Layer test (Root dry weight)	1S11,OH30	+	-	+	+
OP5		PI 398297	F4:6 (111)	A	Tray test (Lesion length)	OH7	+	+	-	+
OP6		PI 417178	F4:6 (128)	A	Tray test (Lesion length)	OH7	+	+	-	+

❑ Procedures

Phenotypic & Genotypic assays ⇨ BLUP estimation & Genetic map construction in a population

Standardization of the BLUP values & integration of genetic maps

Linkage analysis
Inclusive Composite Interval Mapping (ICIM)
w/ QTL IciMapping v3.2 (Li et al., 2007)

Joint linkage analysis
Joint Inclusive Composite Interval Mapping (JICIM)
w/ QTL IciMapping v3.2 (Li et al., 2011)

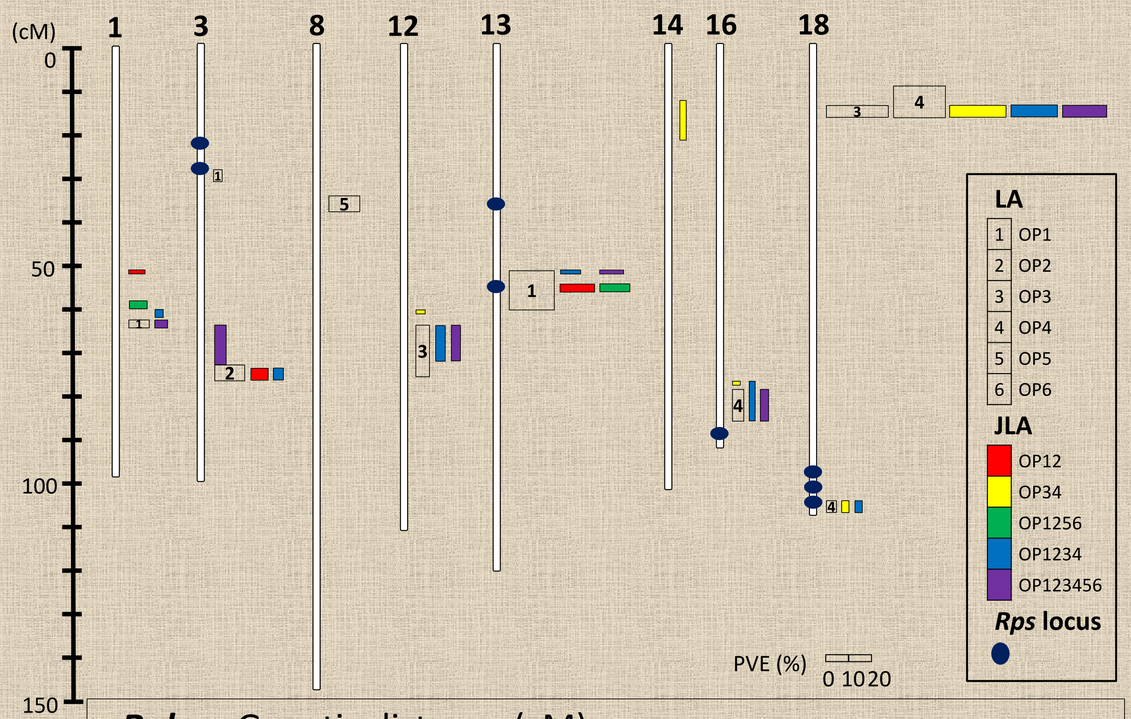
Results & Discussion

❑ Minimal effect of standardization

1. Unequal variance of phenotypic data by different assays requires standardization of phenotypes.
2. Methods of standardization: by variance of population or variance of checks in raw data or BLUP values.
3. Methods of standardization evaluated for equality of variance among populations by Levene's test (Levene, 1960)
4. No false positive or negative QTL detected by ICIM using the Z scores
   - Standardized by population on the equation, $Z = (BLUP - \mu_{BLUP}) / \sigma_{BLUP}$

Standardization was considered to have no effect on subsequent JLA.

❑ Identification of QTL by LA and JLA



- ***Ruler*** : Genetic distance (cM)
- ***White vertical long bars*** **:** Chromosome
- ***Bar height*** : Interval of log of odds (LOD) peak for a QTL
- ***Bar width*** **:** Phenotypic variance (%) explained (PVE) by a QTL
- ***Navy circles*** : Known resistance genes (*Rps*) to *P. sojae*

❑ Evaluation of Joint linkage analysis

Generally, JLA resulted in similar QTL that were mostly in accordance with the those detected by LA in single populations with only a few missed or additional QTL. The present study utilized only up to 6 populations. Thus, there was no dramatic increase in the number of QTL identified by JLA, as reported in Buckler et al. (2009) which used 25 populations. Instead, this study agreed with other studies which applied JLA with fewer populations (Chandler et al., 2013; Li et al., 2011; Yang et al., 2013).

- **Possible benefits:**

1. Additional QTL could be identified when heterogeneous conditions were minimal among combined populations. In JLA of OP34 (OP3 and OP4 combined), one additional QTL was detected on chromosome 14. This QTL was insignificant in both LA of OP3 and OP4.
2. Once data was standardized, differing phenotypic assay methods negligibly affected the identification of QTL in JLA (scenario 3 and 4). Consequently, it is possible to combine populations screened by different phenotypic assay methods after standardization.

- **Possible drawbacks:**

1. For rare QTL, which segregate in only one population and have marginal significance, JLA hindered QTL detection. This was also reported in experiments conducted under the homogeneous conditions in genotypic and phenotypic assays (Chandler et al., 2013; Li et al., 2011; Yang et al., 2013).
2. Many non-overlapping markers among populations, may result in significant changes in the integrated genetic map and, thus, changes in QTL detection.

❑ Novel QTL and a major effect QTL

Sixteen QTL conferring partial resistance to *P. sojae* were identified, 4 of which were first reported in the present study (chromosomes 4, 9, 12, and 16). A major QTL on chromosome 18 explained up to 45% of the phenotypic variance and the resistance alleles of the QTL were provided by the parental lines PI 427106 and PI 427105B.

References cited

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