

DISCUSSION

All blast samples were collected from rice and weed in the farmer fields such as TDK1, TSN1, RD10, CR203, unknown rice and weed in April, 2004. The samples of blast symptoms were sampling from rice field in 5 districts that covering rice production area of Savanakheth Province, Central part Laos. Finally, diverse groups of blast fungi identified by pathotyping were represented as Lao *Pyricularia grisea* population to Laos.

Among seventy monospore of *Pyricularia grisea* isolates were obtained from infected rice leaves and panicles collected in Central plain. The collections were collected from 5 districts: Khanthabuly (SKH), Champhone (SCH), Xonbuly (SXN), Xayphuthong (SXT) and Xaybuly (SXB) of Savanakheth Province. Sporulation was induced on specimens by overnight incubation in petri dishes at room temperature and 100 % relative humidity.

By using pathotypic grouping, the 50 *Pyricularia* isolates clustered into 4 groups indicating genetic divergence within these pathotypic groups where all weed blast isolates clustered on 1 group separately from rice blast isolates. 22 representative rice blast isolates from 50 *Pyricularia* clustered into 2 predominant and 1 minor group, First group, most isolates were collected from CR203 cultivar, Champhone district and the second one was derived from different rice varieties and 5 districts. The similarity and clustering analysis with 5 selected isolates from Northern, North-eastern and Southern Thailand showed that 5 Lao isolates belong to THL318 and 16 Lao isolates were similar to THL557, THL899, TH16 and THL191 indicating genetic divergence within these pathotypic group, suggesting that despite of the small number of isolates tested coming from only five districts of Savanakheth, the population of *P. grisea* is highly diverse and virulent, it's probably that blast pathogen in Laos are similar or the same race as to Thailand.

Under natural condition, JHN has long been resistant to blast pathogen. Results of leaf blast scoring of parental lines and 587 RILs population screening have

supported prior observation that JHN was broadly resistant to all Lao blast isolates. This was shown with high RI value of 1 for JHN while RI value for KDML105 was only 0.38 which is incomparable with those of JHN for leaf blast screening. Thus, progenies with alleles contributed by JHN would be potentially more durable than those with alleles from KDML105 leading to further investigation. Virulence analysis showed that three rice varieties LaiDaw, KDML105 and CR203 are highly susceptible to leaf blast disease. Only two isolates infected IR64 and IR62266. None of the Lao blast isolates were virulent to JHN and CT9993.

The use of 3 isolates for leaf blast screening resulted to 3 sets of data for blast reaction. Data from two out of three sets had fitted a ratio of 3:1 meaning the presence of 2 QTLs while a ratio of 1:1 meant the possibility of 1 QTL being detected. For leaf blast screening, LPDR16 and LPDR47 caused the same phenotypic reaction of resistant to susceptible ratio on the RILs population. The same trend for each screening following the inoculation of LPDR16 and LPDR47 indicated that these two isolates had triggered corresponding virulent genes/QTL possibly at the same region on chromosome1 and 11.

The RILs population that responded to LPDR47 infection is slightly different from those of LPDR16 and LPDR41. The resistant to susceptible ratio for leaf blast screening using LPDR47 was equivalent to 3: 1. In comparison to the response given by LPDR16 and LPDR47, it seemed that LPDR47 also triggered the activation process of genes/QTL at the same region on the chromosome as it occurred with the first two isolates following the inoculation at vegetative stage. Therefore, LPDR47 appeared to be ideal for leaf blast screening.

Inoculation with 3 selected isolates revealed QTLs for leaf blast on chromosome1 and 11 of which JHN was responsible for all resistant alleles. However, the presence of QTLs for leaf blast on chromosome12 when different isolates were used indicated different donors were detected that contributed to the resistant alleles. QTL for leaf blast on chromosome12 had resistant alleles from KDML105. Interestingly, KDML105 was the sole contributor of resistant allele on this QTL .

In this study, QTL with JHN as sole contributor of resistant alleles were found on chromosome1 with high LOD for leaf blast resistance. The peak of QTL on chromosome1 was found near RM319 marker which was close to RZ19 – RG331 flanking marker reported by Prashanth *et al.* (2002) who also found QTL on this chromosome. On chromosome11, LOD values for leaf blast resistance were rather high ranging from 24.24 – 86.02. These indicated the strong presence of polygenes of which the alleles were again contributed by JHN. One of the possible major genes could be *Pi-7(t)* since it was reportedly linked to RG103 and Npb186 flanking marker (Wang *et al.*, 1994) which was mapped close to RM139 marker used in this study. The other possible qualitative gene on chromosome11 was *Pi 1* linking to the flanking marker RZ536 (Hittalmani *et al.*, 2000) which was mapped and only 3.5 cM far from RM139 (Chen *et al.*, 1997). Inukai *et al.* (1996) have reported that *Pi lm2* was allelic or closely linked to *Pi 1* on chromosome11. Tabien *et al.* (2000) reported that *Pi lm2* was linked to RZ536 which was close to RM139. Therefore, *Pi lm2* could be the third major gene detected on chromosome11 in this study. Unlike those of chromosome1 and 11, QTLs on chromosome12 had resistant alleles contributed by both KDML105 and JHN. The presence of a resistant gene *Pi ta* linked to RZ397 at a distance of 3.3 cM on chromosome12 was reported by Hittalmani *et al.* (2000). Thus, *Pi ta* could be one possible resistant gene found in this study since RZ397 was 12.1 cM far from RM179. Since Kiyosawa (1984) had found alleles between *Pi ta* and *Pi 4a(t)* and Inukai *et al.* (1994) reported that *Pi 4a(t)* was allelic to *Pi 4b(t)*, therefore, *Pi 4a(t)* and *Pi 4b(t)* could be the other two possible major genes on chromosome12. In addition to this, Mew *et al.* (1994) reported that there could be a cluster of QTLs on chromosome12 with *Pi 4(t)* being closely linked to RG869 and RZ397. Thus, *Pi 4(t)* was highly likely to be the fourth major gene detected on chromosome12.