รายงานวิจัยฉบับสมบูรณ์

ภาคผนวก (การตีพิมพ์เผยแพร่ผลงาน)

โครงการกลไกทางสรีระเชิงนิเวศน์และพันธุกรรมที่ควบคุมการใช้ธาตุอาหารในพืช

(Ecophysiological processes and genetic controls relating to plant nutrition)

โดย เบญจวรรณ ฤกษ์เกษม และคณะ

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คณะผู้วิจัย	สังกัด
1. ศ.ดร. เบญจวรรณ ฤกษ์เกษม	มหาวิทยาลัยเชียงใหม่
2. รศ.ดร. สายสมร ลำยอง	มหาวิทยาลัยเชียงใหม่
3. รศ. ดร. ศันสนีย์ จำจด	มหาวิทยาลัยเชียงใหม่
4. ผศ. ดร.ศักดา จงแก้ววัฒนา	มหาวิทยาลัยเชียงใหม่
5. ดร. กนก ฤกษ์เกษม	มหาวิทยาลัยเชียงใหม่
6. ดร.จรรยา มณีโชติ	กรมวิชาการเกษตร
7. นายสิทธิชัย ลอดแก้ว	มหาวิทยาลัยเชียงใหม่
8. นายพิภพ ลำยอง	มหาวิทยาลัยเชียงใหม่
9. นายนริศ ยิ้มแย้ม	มหาวิทยาลัยเชียงใหม่
10. นางปณิดา บุญสิทธิ	มหาวิทยาลัยเชียงใหม่

สนับสนุนโดยสำนักงานกองทุนสนับสนุนการวิจัย

ชุดโครงการส่งเสริมกลุ่มวิจัย

ภาคผนวก

บทความวิชาการ

1. **Peer-reviewed papers published (ตีพิมพ์เผยแพร่แล้ว หรืออยู่ในระหว่างการตีพิมพ์)**

1.1 Rerkasem B, Jamjod S, Niruntrayagul, S. Boron *In press.* Increasing boron efficiency in many international bread wheat, durum wheat, triticale and barley germplasm will boost production on soils low in boron. Field Crop Research

1.2 Sansanee Jamjod, Sunisa Niruntrayagul & Benjavan Rerkasem. *In press.* Genetic control of boron efficiency in wheat (*Triticum aestivum* L.) Euphyitca

1.3 Wongmo, J., Jamjod, S. and Rerkasem, B. *In press.* Contrasting responses to boron deficiency in barley and wheat. Plant and Soil

1.4 Duangjai Nachiangmai, Bernie Dell, Richard Bell, Longbin Huang and Benjavan Rerkasem. *In press.* Genotypic variation in boron long distance transport into the reproductive organ of wheat. Plant and Soil

1.5 Yimyam, N. Rerkasem, K and Rerkasem, B. 2003. Fallow enrichment with pada (*Macaranga denticulata* (Bl.) Muell. Arg.) trees in rotational shifting cultivation in Northern Thailand Agroforestry Systems 57: 79-86.

1.6 M. Leesawatwong, S. Jamjod and B. Rerkasem. 2003 Determinants of a premium priced special quality rice. International Rice Research Notes. 28: 34

1.7 C. Prom-u-thai and B. Rerkasem. 2003. The effect of nitrogen on rice grain iron. International Rice Research Notes. December 2003.

1.8 Rerkasem K, Korsamphan C, Thong-ngam C, Yimyam N and Rerkasem B. 2002. Agrodiversity lessons in mountain land management. Mt. Res. Dev. 22: 4-9 (บทความได้รับรางวัลที่ 1 จาก United Nations University)

1.9 C. Prom-u-thai, B. Dell, G. Thomson, B. Rerkasem. *In press.* Easy and rapid detection of iron in rice grain. ScienceAsia

1.10 Rerkasem, B. and K. Rerkasem. 2002 Agrodiversity for *in situ* conservation of Thailand's native rice germplasm. CMU J. 1: 129-148.

1.11 Rerkasem B and Jamjod S. 2001. Overcoming wheat sterility problem with boron efficiency. Dev. Plant Soil Sci. 92: 82-83.

1.12 Prom-u-thai, C. and Rerkasem, B. 2001. Iron in Thai rice. Dev. Plant Soil Sci. 92: 350-351.

1.13 Benjavan Rerkasem. 2003. Biotechnology and Agriculture. An invited review, pp. 293-321, *in* Social Challenges for the Mekong Region, Mingsarn Kaosa-ard and John Dore (Eds.). White Lotus, Bangkok.

1.14 Kanok Rerkasem. 2003. Uplands Land Use. An invited review, pp. 323-346, *in* Social Challenges for the Mekong Region, Mingsarn Kaosa-ard and John Dore (Eds.). White Lotus, Bangkok.

1.15 Rerkasem, B. 2002. Crop responses to boron and genotypic variations. An invited review, pp. 269-280, *in* All Aspects of Plant and Animal Boron Nutrition, Eds: H. E. Goldbach, B. Rerkasem, M. A. Wimmer, P. H. Brown, M. Thellier and R.W. Bell. Kluwer and Plenum Academic Publishers

1.16 Ahmed M, Jaihiruddin M, Jamjod S and Rerkasem B. 2002. Boron efficiency in a wheat germplasm from Bangladesh. Pp. 299-303, *in* All Aspects of Plant and Animal Boron Nutrition, Eds: H. E. Goldbach, B. Rerkasem, M. A. Wimmer, P. H. Brown, M. Thellier and R.W. Bell. Kluwer and Plenum Academic Publishers.

1.17 NaChiangmai D, Dell B, Huang L, Bell R and Rerkasem B. 2002. The effect of boron on pollen development in two wheat cultivars (*Triticum aestivum* L.). Pp. 181-185, *in* All Aspects of Plant and Animal Boron Nutrition, Eds: H. E. Goldbach, B. Rerkasem, M. A. Wimmer, P. H. Brown, M. Thellier and R.W. Bell. Kluwer and Plenum Academic Publishers.

1.18 Rerkasem K, Thong-ngam C, Korsamphan C, Yimyam N and Rerkasem B. 2002. Pp. 200-232, *in*: Land Use Changes in the Highlands of Northern Thailand. An invited review paper in 'Cultivating Biodiversity' Eds. H Brookfield, C Padoch, H Parson and M Stocking. ITDG Publishers, London and United Nations University, Tokyo.

1.19 เนตรนภา อินสลุด Richard W. Bell และเบญจวรรณ ฤกษ์เกษม 2546 การตอบสนองของพันธุ์ข้าวไร่และข้าวนาสวนต่อสภาพดินขังน้ำและดินระบายน้ำดี วารสารเกษตร (มช) ACCEPTED

1.20 จำเนียร วงษ์โม้, ศันสนีย์ จำจด และ เบญจวรรณ ฤกษ์เกษม 2546 เปรียบเทียบการตอบสนองต่อการขาดธาตุโบรอนในข้าวบาร์เลย์และข้าวสาลี วารสารเกษตร (มช) ACCEPTED

1.21 ทินกร ศรีวิชัย ศันสนีย์ จำจด และ เบญจวรรรณ ฤกษ์เกษม 2546 การตอบสนองต่อโบรอนในถั่วพุ่ม วารสารเกษตร (มช) ACCEPTED

1.22 นริศ ยิ้มแย้ม สิทธิชัย ลอดแก้ว เบญจวรรณ ฤกษ์เกษม และ กนก ฤกษ์เกษม 2546 การจัดการความหลากหลายของดันปะดะในไร่หมุนเวียนของกะเหรี่ยงโปว์ ในภาคเหนือของประเทศไทย วารสารเกษตร (มช) ACCEPTED

1.23 รัตญา ยานะพันธุ์ และ เบญจวรรณ ฤกษ์เกษม 2546 การคัดเลือกพันธุ์ข้าวไทยภายใต้สภาพขาดธาตุเหล็กโดยวัดปริมาณคลอโรฟิลล์ในใบ วารสารเกษตร (มช) ACCEPTED

1.24 การเคลื่อนย้ายโบรอนในถั่วเขียว 2546 สาวิกา กอนแสง และเบญจวรรณ ฤกษ์เกษม วารสารเกษตร (มช) ACCEPTED

1.25 สุพรรณิการ์ พันชนะ ศันสนีย์ จำจด และเบญจวรรณ ฤกษ์เกษม 2546 การตอบสนองต่อความเป็นพิษของโบรอนในข้าวสาลีสามพันธุ์ที่มีระดับความทนทานต่อการขาดโบรอนแตกต่างกัน วารสารเกษตร (มช) ACCEPTED

1.26 อยุชย์ คงปั้น ศันสนีย์ จำจด และ เบญจวรรณ ฤกษ์เกษม 2546 อิทธิพลของโบรอนต่อคุณภาพเมล็ดในถั่วเขียวต่างพันธุ์ วารสารเกษตร (มช) ACCEPTED

2. **Papers submitted for publication in peer-reviewed journals**

2.1 Somchit Youpensuk, Benjavan Rerkasem, Bernie Dell and Saisamorm Lumyong. Arbuscular mycorrhizal fungi from the rhizosphere of a fallow enriching tree, *Macaranga denticulata* Muell. Arg. and their effect on the host plant. Submitted to Agroforestry Systems (January 2003).

2.2 C. Prom-u-thai, B. Dell, G. Thomson, B. Rerkasem. Distribution and structure of protein and phytin bodies in seed of four rice genotypes. Submitted to Canadian Journal of

3. **Papers in preparation, submission expected by end of 2003**

3.1 Ayut Kongpan, Sansanee Jamjod and Benjavan Rerkasem. Boron efficient germplasm identified in *Vigna mungo* (L.) Hepper and *Vigna radiation* (L.) Wilczek. For submission to Plant and Soil or Field Crop Research.

3.2 Supannika Punchana, Sansanee Jamjod and Benjavan Rerkasem. Are boron efficient wheat always susceptible to boron toxicity? For submission to Euphytica.

3.3 Chanakan Prom-u-thai and Benjavan Rerkasem. Iron in the Grain of High and Low Iron Density Rice Grown in Different Water Regimes.

3.4 Supawadee Ngorian, Sansanee Jamjod and Benjavan Rerkasem. Response of F_2 population derived from boron efficient (Fang 60) x boron inefficiency

(Bonza) wheat (*Triticum aestivum* L.) genotypes to boron levels. For submission to Euphytica or J. Plant Breeding.

3.5 Benjavan Rerkasem and Sansanee Jamjod. Boron Deficiency in Wheat: a Review Submitted to Field Crop Reserch

4. Book edited

4.1 Goldbach, H.E., Rerkasem, B., Wimmer, M.A., Brown, P.H., Thellier M. and Bell, R.W. 2002. All aspects of Plant and Animal Boron Nutrition, Kluwer and Plenum Academic Publishers

Increasing boron efficiency in many international bread wheat, durum wheat, triticale and barley germplasm will boost production on soils low in boron

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Abstract

Boron deficiency causes grain set failure and yield loss in many of the world's wheat growing countries. We suggest growing B efficient genotypes as a means to overcome the problem. This study evaluated an international germplasm of bread wheat (*Triticum aestivum* L.), durum wheat (*Triticum durum* Dest.), triticale (x*Triticosecale* Wittmack) and barley (*Hordeum vulgare* L.) for B efficiency. The first set of germplasm consisted of bread wheat, durum wheat and triticale from CIMMYT 1996/97 nurseries and a B efficient check wheat cv. 'Fang 60'. The lines were grown in the field on soil with 0.1 and 0.2 , and 0.3 mg HWS B kg^{-1} for durum wheat and triticale. The grain set index (GSI, percentage grain set in the first two florets of 10 central spikelets) measured B efficiency in wheat, durum and triticale genotypes without the need for a B sufficiency control. Three quarters of the lines tested

were B inefficient, which included all of the durum wheat, 84% of the triticale and 60% of the bread wheat lines. Six of the bread wheat lines evaluated were in the same B efficient class as Fang 60. The response to low B was confirmed in a sand culture without added B. Similarly high frequency of B inefficiency was found in a second set of germplasm which included bread and durum wheat, barley and triticale lines from CIMMYT and ICARDA 2000/01 international nurseries. Incorporating the B efficiency trait in germplasm such as these, would ensure their adaptation to low B soils, and so enable their genetic potential to be fully realized in some of the world's difficult production areas.

Keywords: Boron deficiency

Abbreviations: B, boron; HWS, hot water soluble; CIMMYT, the International Maize and Wheat Improvement Center; GSI, grain set index; ICARDA, International Center for Agricultural Research in the Dry Areas; 4HTWYT, 4th High Temperature Wheat Yield Trial; 17ESWYT, 17th Elite Selection Wheat Yield Trial; 18SAWSN, 18th Semi-Arid Areas Wheat Screening Nursery; 28IBON, 28th International Barley Observation Nursery; 28ITYN, 28th International Triticale Yield Nursery; 28IDYN, 28th International Durum Yield Nursery; 33IDYN, 33rd International Durum Yield Nursery; 33ITSN, 33rd International Triticale Screening Nursery; 33IBWSN, 33rd International Bread Wheat Screening Nursery.

1. Introduction

In many of the world's wheat growing areas, from Brazil's irrigated flood plains (da Silva and de Andrade, 1980) and Cerrados (da Silva and de Andrade, 1983), China (Li et al., 1978; Liu et al., 1981) to India's northwestern states and along the Indo-Nepal border (Tandon and

Naqvi,1992), Nepal (Misra et al., 1992) and Bangladesh (Reuter, 1987; Rerkasem, 1996), severe yield losses may be caused by B deficiency, through adverse effects on male fertility and grain set. These and later studies (Jamjod et al., 1992; Subedi et al., 1997) also reported large genotypic variation in the response to B, or B efficiency, in bread wheat. The term B efficiency is used here without inferring a mechanism, in the same way as defined for Zn efficiency (Graham, 1984), to designate the ability of a genotype to perform well in soils too deficient in B for other genotypes. Selecting for B efficiency has been suggested as one cost effective solution to the problem of yield loss due to boron deficiency in commercially grown wheat crops (Rerkasem and Jamjod, 1997).

The International Maize and Wheat Improvement Center (CIMMYT) is the world's single most important source of wheat germplasm. Each year thousands of lines and breeding populations from CIMMYT are introduced into countries throughout the world. In the last four years alone, more than 160 bread wheat, durum wheat, triticale, and barley varieties derived from CIMMYT germplasm have been released by more than 30 countries (www.cimmyt.org/Research/wheat). About 55 million hectares of spring bread wheat, nearly 80% of annual spring wheat area in the developing world excluding China, are now sown each year with varieties developed from the CIMMYT germplasm. Information about B efficiency of this major source of bread wheat and durum wheat, triticale and barley germplasm, especially the portion destined for areas with low B soils, could help to prevent crop losses due to B deficiency. This study aimed to evaluate the potential to perform in soils low in B of bread and durum wheat, triticale and barley germplasm.

2. Materials and methods

An international germplasm of bread and durum wheat and triticale from CIMMYT and barley from CIMMYT/ICARDA was grown in the field on a sandy loam Tropaqualf soil of San Sai series and in sand culture in Chiang Mai, Thailand. The first set of germplasm included 98 lines of bread wheat (the 4HTWYT and 17ESWYT) and 49 lines each of durum wheat (28IDYN) and triticale (28ITYN) and a B efficient check wheat, cv 'Fang 60' (Jamjod et al., 1992). The experiment was arranged as a split plot, with soil B levels in duplicated mainplots and lines of wheat and triticale (sown, at the seed rate of 3 g m^{-1}, in single rows 5 m long, 0.25 m between rows) as subplots. Borax at 0, 1 and 1.5 kg B ha^{-1} was applied to create the B levels, giving the soil 0.1 (B0.1), 0.2 (B0.2) and 0.3 (B0.3) mg HWS B kg^{-1}. Bread wheat are generally less sensitive to B deficiency than durum wheat and triticale (Rerkasem, unpublished), the 4HTWYT and 17ESWYT were grown at B0.1 and B0.2 only. At maturity, assessment was made of the number of grains per ear, number of spikelets per ear, and grain set index (GSI, percentage grain set in the first two florets of 10 central spikelets, Rerkasem and Loneragan, 1994) from 20 randomly selected ears, and the number of ears in one m length of row.

The 4HTWYT, 28ITYN and 28IDYN were also evaluated against Fang 60 in a sand culture to which no B was supplied. Entries were grown in duplicated earthenware pots (Ø 30 cm, 30 cm deep) containing washed river quartz sand. The pots were watered twice daily with 1 liter of nutrient solution containing 1000 μM $CaCl_2$, 250 μM $MgSO_4$, 500 μM KH_2PO_4, 10 μM Fe EDTA, 250 μM K_2SO_4, 1 μM $MnSO_4$, 0.5 μM $ZnSO_4$, 0.2 μM $CuSO_4$, 0.1 μM $CoSO_4$, 0.1 μM Na_2MoO_4 (Broughton and Dilworth, 1971) and 5 mM KNO_3. Grain set index was assessed at maturity on five ears from each replication for each line.

The second set of germplasm consisted of 1,108 lines from the 2000/01 international bread wheat (33IBWSN, 18SAWSN), durum wheat (33IDYN) and triticale (33ITSN) nurseries from CIMMYT and a barley nursery (28IBON) from CIMMYT/ICARDA. Entries were sown on soil with 0.1 mg HWS B kg^{-1} in single 5 m rows, with 0.25 m between rows and a row each of the B efficiency checks inserted after every 19 entries. The B efficiency checks were BRB9604 and BRB96-9 (Jamjod and Rerkasem, 1999) for the 28IBON, and Fang 60 and SW41 (Anantawiroon et al., 1997) for the other nurseries. At maturity the GSI was determined from 20 randomly selected ears for each row.

3. Results

3.1 Responses to boron in bread wheat, durum wheat and triticale

Bread wheat, triticale and durum wheat all exhibited responses to B, where number of grains per ear and GSI, that varied significantly among the genotypes (Table 1). Symptoms of B deficiency observable in bread wheat in B0.1 at anthesis included shrivelled anthers, poorly developed pollen that did not stain with iodine, and florets that failed to fertilize and remained open, giving the ears a translucent, "paper lamp" appearance. The paper lamp effect was also observed in triticale and durum in B0.1 and sometimes in B0.2. In these low B levels some triticale lines showed a "rat-tail" symptom of the ear tip, in which terminal spikelets were reduced in size and sometimes completely degenerated, leaving remnants of dead, papery white tissue. These symptoms did not occur in bread or durum wheat lines. There was a significant interaction between the genotype and B effect on the number of spikelets per ear in triticale, but not in bread wheat or durum. The number of ears m^{-1} was not affected by B in all three species .

At B0.1, the B efficient check Fang 60 set grain normally with GSI >85%, while genotypes evaluated ranged in GSI from 0% to 100%. Entries with grain set failure in half or more of their competent florets in B0.1 accounted for three-quarters of the 1996/97 germplasm (Table 2). These included all of the durum lines and all but eight of the triticale lines. Forty percent of the germplasm was most severely affected by B deficiency in B0.1, with grain set in only one in five of their competent florets or fewer. These included most of the durum (82%), half of the triticale, and 14% of the bread wheat lines. Six out of 98 entries of bread wheat, but none of durum and triticale, had GSI in B0.1 exceeding 85%, the same as B efficient Fang 60.

The species differences were also evident in the responses to B in means of GSI and the number of grains per ear. The whole nursery mean GSI in B0.1 was highest in bread wheat, followed by the 28ITYN and 28IDYN, in that order (Table 3). Increasing B level increased GSI in the B inefficient classes in all three species. In the moderately inefficient classes the GSI was approaching maximum when B was increased to B0.2, but in the more inefficient classes, which included all of the durum lines and almost all of the triticale lines, maximum GSI was not reached until B0.3. Responses to B in the number of grains per ear (Table 4) followed closely their GSI responses ($r^2 = 0.74$ for the 4HTWYT and 17ESWYT combined; 0.61 for 28ITYN and 0.62 for 28IDYN, all significant at $p < 0.001$).

In sand culture to which B had not been applied (B0) the 4HTWYT, 28ITYN and 28IDYN lines responded to B deficiency in the same way as in the field at B0.1. In all three species, the GSI means for each nursery and for each B efficiency class in B0 (Table 5) were almost identical to those in B0.1 (Table 4), r^2 for the 4HTWYT measured at 0.90. Some of the B

efficient and inefficient bread wheat lines are identified in Table 6. For these individual lines from the 4HTWYT, their GSI in the sand culture without added B also closely followed those in the low B soil.

In triticale, the number of spikelets per ear in about one third of the entries was depressed by B deficiency, with up to 50% depression in the most severe cases (Table 7). The ear size response to B in the 28ITYN, however, did not correlate with the GSI response ($r^2 = 0.002$, not significant at $p < 0.05$), neither did those in the 4HTWYT, 17ESWYT ($r^2 = 0.006$ and 28IDYN ($r^2 = 0.0004$).

3.2 Boron efficiency in the 2000/2001 international germplasm

Through out the experimental plot, the GSI in the B efficient check wheat, Fang 60, was approaching 100%, and was 84% in BCMU96-9 barley, while it ranged between 70% and 80% in the moderately inefficient wheat SW41 and 60% in BRB9604 (Table 8). Lines with GSI exceeding 90% included 10% of the 33IBSWYN, 8% of the 18SAWSN, and none of the 28IBON, 33IDYN and 33ITSN. Entries with GSI in the low B soil in same range as or lower than SW41, the moderately inefficient wheat check, accounted for 71% of the bread wheat, 96% of the triticale and all of the durum wheat and barley in the 2000/01 nurseries. The rat-tail symptom was observed on many of the barley as well as triticale entries.

4. Discussion

The predominance of B inefficiency in the bread and durum wheat, barley and triticale germplasm evaluated has a potential to cause serious problems in many of the world's

growing areas. Contrary to the often held view of low B requirement and insensitivity to B deficiency of wheat and related small grains (Lamb, 1967; Marten and Westermann, 1991), reports of yield loss due to B deficiency have come from many regions. Boron deficiency has been reported in the field on at least 132 crops in 80 countries in all continents (Shorrocks, 1997). Large areas of low B soils have been identified in the Americas, Europe, Africa and Asia, with the single largest contiguous area of B deficiency is in China (Liu et al., 1981).

Failure of commercial wheat crops due to B deficiency has been reported in China from Heilongjian in the northeast, (Li et al, 1978) to Yunnan in the southwest (Yang, 1992). The most extensive area of B deficient wheat so far identified is in one of the poorest corners of Asia, that extends from the northwest of India into Nepal and Bangladesh, where about one million ha of crop land is estimated to be adversely affected by B deficiency (Kataki et al., 2002). Low B soils and incidences of B deficiency are prevalent in the Indian northwestern states of Bihar, West Bengal, Orissa, Meghalaya and Assam (Sakal and Singh, 1995), where grain set failure and responses to B in wheat have been reported (e.g. Singh et al., 1976; Sarkar and Chakraborty, 1980; Mandal and Das, 1988; Dwivedi et al., 1990). A common local knowledge that the problem extends into neighboring areas of Bangladesh (Reuter, 1987) has been recently confirmed (Kataki et al. 2002). In Nepal B deficiency in wheat and other crops has been commonly reported from the Terai or flat plains, where half of the country's wheat crop is grown (Subedi et al., 1996). Reports of B deficiency-induced sterility have also begun to come from "less likely" regions. Incidences of B deficiency in wheat have been reported from Pakistan (Rashid et al., 2002), which is known for high levels of soil B (Sillanpaa, 1982) or even from areas prone to B toxicity problem. That pockets of B deficiency may occur among soils with toxic level B has been documented in the Anatolia in

Turkey (Gezgin et al. 2002). In one such pocket B application has been reported to increase grain yield of durum wheat (cv. Ç-1252) by 16% (Topal et al., 2002). Bread and durum wheat, barley and triticale germplasm with the same level of B efficiency as the majority of those evaluated in this study should be expected to do poorly in all such growing regions that are prone to B deficiency.

The international germplasm from CIMMYT, and ICARDA in the case of barley, consists of superior genetic materials that have incorporated desirable characteristics such as high yielding capacity and resistance to important diseases. Many developing countries, Nepal and Bangladesh included, depend on the CIMMYT germplasm as the source of their new cultivars. In areas prone to B deficiency, the genetic yield potential of this introduced germplasm cannot be fully utilized because of the B deficiency constraint. Just as we have found with the CIMMYT germplasm in general, very high proportions of the recommended and widely grown cultivars in Nepal (Subedi et al., 1997) and Bangladesh (Ahmed et al., 2002) have been rated as B inefficient. It is not surprising that grain set failure is still common in farmers' wheat fields in Bangladesh, especially on the very low B soils in the northwestern part of the country (Kataki et al, 2002), and Nepal (Pant, 1994; Subedi et al., 1996). One solution would be to apply B fertilizer, as is routinely done in Brazil. However, persistence of the problem in many countries indicates that this seemingly simple and inexpensive option is not always available to growers. Furthermore, new improved varieties selected on research stations where B fertilizer has been applied can be expected to fail commercially, unless advanced breeding lines are assessed for B efficiency before they reach on-farm trials.

Under the low B condition that completely depressed grain set in other genotypes, a few bread wheat lines have been found to set grain normally, along with the B efficient check Fang 60. This B efficiency trait confers a remarkable adaptation to low B soils, so offers a solution to B deficiency in wheat production. Grain set failure and yield losses may be prevented by ensuring that germplasm destined for areas prone to B deficiency is B efficient. A cross between B efficient bread wheat Fang 60 and very inefficient Bonza demonstrated that gene(s) for B efficiency may be easily transferred (Ngorian, 2001). Responses to B in the progenies suggested a few major genes controlling B efficiency. There is real scope for genetic improvement. The source of genes for B efficiency already exists in bread wheat, screening can be done in the field on soil with low B or in a simple sand culture, as we have demonstrated. Many of the B efficient genotypes identified were advanced breeding lines, i.e. ready to be released as cultivars, that were included in CIMMYT international yield nurseries such as the 4HTWYT and 17ESWYT. For triticale, durum wheat and barley, a transfer of relevant genes from B efficient bread wheat such as Fang 60 will probably be essential. As it is present in the more B efficient bread wheat but not in the less B efficient durum wheat, triticale and barley, the D genome is of great interest as a possible source of genes for increasing B efficiency.

The primary effect of B deficiency in bread and durum wheat, barley and triticale was grain set failure. Boron levels affected grain set in bread wheat lines in the same manner as previously reported for wheat (Li et al., 1978; da Silva and de Andrade, 1980) and barley (Ambak and Tadano, 1991). The present study has established that B deficiency has similar effects on grain set in durum wheat and triticale, and that the GSI may be used to assess B efficiency in these species as well as bread wheat (Rerkasem and Loneragan, 1994) and barley (Jamjod and Rerkasem, 1999). The effect of B deficiency on grain set was equally

well described using either the GSI or number of grains per ear. However, as the number of competent florets and the potential number of grains may vary with genotypes and environment, a B sufficiency control is required to measure the effect of B on the number of grains per ear in different genotypes and growing conditions. In contrast, the GSI is very useful to evaluate responses to low B in nurseries with large entry numbers without the need for a B sufficiency control. The extent of B deficiency in the screening environment may be indicated by inclusion of a set of check genotypes covering a range of responses to low B. The GSI is also useful to quantify the problem of grain set failure in wheat, triticale and barley under a wide range of environmental condition on-farm. The presence of common varieties with known susceptibility to B deficiency allows verification of B deficiency as the likely limiting factor.

In some cases, triticale also responded to low B with a depression the number of spikelets per ear, similar to a previous report on barley (Jamjod and Rerkasem, 1999). The poor correlation between the effect of B on the number of spikelets per ear and the GSI for individual genotypes and species suggests that a different process from ear size response may govern the grain set response. The rat-tail symptom provides a convenient measure for the effect of B deficiency on spikelet number in barley and triticale. A slight improvement in the precision of determination of the B effect on triticale may be expected from combining the GSI and ear size response, but this may not be worth the extra cost of a B sufficiency control.

It is as yet unclear what mechanisms are involved in B efficiency. The differential effects of B deficiency on B efficient and inefficient genotypes appear not to be reflected in the concentration of B in their flag leaf or developing ear (Rerkasem and Loneragan, 1994) or the way they partition B among the various organs (Subedi et al., 1999). Triticale has been

reported to have derived genes for Cu efficiency (Graham, 1979) and Zn efficiency (Cakmak et al., 1997) from the rye chromosomes. Since triticale is largely B inefficient, the mechanism for B efficiency is unlikely to be related to those involved in Cu and Zn efficiency. For tolerance to B toxicity, exclusion of B has been suggested as a primary mechanism (Nable et al., 1997). In areas where soils have both deficient and toxic levels B occurring in close proximity, it may be necessary to ensure that selection and breeding for B efficiency does not inadvertently increase susceptibility to B toxicity, especially in an international germplasm such as that generated and distributed by CIMMYT and ICARDA.

5. Conclusion

The predominance of B inefficiency in germplasm such as that generated and distributed by CIMMYT and ICARDA poses a constraint to the realization of its full genetic yield potential. However, the presence of B efficiency in the germplasm suggests a solution to the problem of B deficiency in wheat production. We have demonstrated that the B efficiency trait may confer adaptation to low B soils, and effectively prevent grain set failure and yield losses. We have further shown that screening for B efficiency can be done easily and recommend that B efficiency should be included in the breeding objectives for bread and durum wheat, barley and triticale germplasm destined for growing areas prone to B deficiency.

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References

Ahmed, M., Jaihiruddin, M., Jamjod, S. and Rerkasem, B. 2002. Boron efficiency in wheat germplasm from Bangladesh. In: Goldbach, H. E., Rerkasem, B., Wimmer, M. A., Brown, P. H., Thellier, M., and Bell, R.W. (Eds.), All Aspects of Plant and Animal Boron Nutrition, Kluwer and Plenum Academic Publishers, Dordrecht, The Netherlands, pp. 299-303.

Ambak, K. and T. Tadano. 1991. Effect of micronutrient application on the growth and occurrence of sterility in barley and rice in a Malaysian deep peat soil. Soil Sci. Plant Nutr. 37, 715-724.

Anantawiroon, P., Subedi, K. and Rerkasem, B. 1997. Screening wheat for boron efficiency. In Bell, R.W. and Rerkasem, B. (Eds.), Boron in Soils and Plants. Dordrecht, The Netherlands: Kluwer Academic Publishers, pp. 101-104.

Cakmak, I., Derici, R., Torun, B., Tolay, I., Braun, H.J., andSchelgel, R. 1997. Role of rye chromosomes in improvement of zinc efficiency in wheat and triticale. Plant Soil 196, 249-253.

da Silva, A.R. and de Andrade, J.M.V. 1980. A Cultura do Trigo nas Varzeas de Minas Gerais – possibilidades e dificuldades. Embrapa, Centro de Pesquisa Agropecuária dos Cerrados – CPAC. Circular Técnica no. 2. 69 p. (In Portuguese)

da Silva, A.R. and de Andrade, J.M.V. 1983 Efeito micronutrientes no chochamento do trigo de seqqueiro e nas culturas de soja e arroz, em latossolo vermelho-amarelo. [Influence of micronutrients on the male sterility, on upland wheat and on rice and soybean yields, in red-yellow latosol] Pesq. Agropec. Bras., Brasilia 18, 593-601. (In Portuguese with English Abstract)

Dwivedi, G.K., Dwivedi, M. and Pal, S.S.. 1990. Modes of application of micronutrients in acid soil in soybean-wheat crop sequence. J. Indian Soc. Soil Sci. 38, 458-463.

Dwivedi, G.K., Dwivedi, M. and Pal, S.S.. 1990. Modes of application of micronutrients in acid soil in soybean-wheat crop sequence. J. Indian Soc. Soil Sci. 38, 458-463.

Gezgin, S., Dursun, N., Hamurcu, M., Harmankaya, M., Önder, M., Sadea, B., Topal, A., Soylu, S., Akgün, N., Yorgancilar, M., Ceyhan, E., Çiftçi, N., Acar, B., Gültekin, I., Iş ik, Y., Şeker, C. and Babaoglu, M. 2002. Boron content of cultivated soils in central-southern Anatolia and its relationship with soil properties and irrigation water quality. In: Goldbach, H. E., Rerkasem, B., Wimmer, M. A., Brown, P. H., Thellier, M., and Bell, R.W. (Eds.), All Aspects of Plant and Animal Boron Nutrition. Kluwer and Plenum Academic Publishers, Dordrecht, The Netherlands, pp. 391-400.

Graham, R.D. 1979. The sensitivity of hexaploid and octaploid triticale and their parents to copper deficiency. Aust. J. Agric. Res. 30, 791-799.

Graham, R.D. 1984. Breeding for nutritional characteristics in cereals. Adv. Plant Nutr. 1, 57-102.

Jamjod, S., Mann, C.E. and Rerkasem, B. 1992. Screening for boron deficiency in wheat. In: Mann, C.E. and Rerkasem, B. (Eds.), Boron deficiency in wheat. CIMMYT Wheat Special Report No. 11, International Maize and Wheat Improvement Center, Mexico, D.F., pp. 79-82.

Jamjod, S. and Rerkasem, B. 1999. Genotypic variation in responses of barley to boron deficiency. Plant Soil 215, 65-72.

Kataki, P.K., Srivastava, S.P., Saifuzzaman, M. and Upreti, H.K. 2002. Sterility of wheat and response of field crops to applied boron in the Indo-Gangetic Plains. J. Crop Production 4, 133-165.

Lamb, C.A. 1967. Physiology. In: Quisenberry, K.S. and Reitz, L.P. (Eds.) Wheat and Wheat Improvement. Agronomy Monograph 13. ASA, Madison, WI., pp. 181-223.

Li, B.H., Li, W.H., Kui, M.C., Chao, W.S., Jern, H.P., Li, C.R., Chu, W.J. and Wang, C.L. 1978 Studies on cause of sterility of wheat. J. Northeastern Agric. Coll. 3, 1-19. (In Chinese)

Liu, Z., Zhu, Q. and Tang, L. 1981. Boron deficient soils and their distribution in China. Soil Research Report No. 5. Institute of Soil Science, Academia Sinica, Nanjing, China.

Mandal A B and Das A D 1988. Response of wheat (Triticum aestivum) to boron application. Indian J. Agric. Sci. 58:681-683.

Marten, J.M. and Westermann, D.T. 1991. Fertilizer applications for correcting micronutrient deficiencies. In: Mordtvedt, J.J. et al. (Eds.) Micronutrients in Agriculture. SSSA Book Series no. 4. SSSA, Madison, WI, pp. 549-592.

Misra, R., Munankarmi, R.C., Pandey S.P. and Hobbs P.R. 1992. Sterility work in wheat at Tarahara in the eastern Terai of Nepal. In: Mann, C.E. and Rerkasem, B. (Eds.), Boron deficiency in wheat. CIMMYT Wheat Special Report No. 11, International Maize and Wheat Improvement Center, Mexico, D.F., pp. 67-71.

Nable, R.O., G.S. Banuelos, and J.G. Paull. (1997) Boron toxicity. Plant Soil 193, 181-198.

Ngorian, S. 2001. Response of Segregating Wheat Populations to Boron Deficiency. M.Sc. Thesis (Agronomy). Graduate School, Chiang Mai University.

Pant, J. 1994. Genotypic response of wheat to boron application under irrigated and water stress conditions. M.Sc. Thesis (Agricultural Systems). Graduate School, Chiang Mai University.

Rashid, A., Muhammad, S. and Rafique, E. 2002. Gynotypic variation in boron uptake and utilization in rice and wheat. In: Goldbach, H. E., Rerkasem, B., Wimmer, M. A., Brown, P. H., Thellier, M., and Bell, R.W. (Eds.), All Aspects of Plant and Animal

Boron Nutrition, Kluwer and Plenum Academic Publishers, Dordrecht, The Netherlands, pp. 305-310

Rerkasem, B. 1996. A general survey of the incidence of wheat sterility. In: Rawson, H.M. and Subedi, K.D. (Eds.), Sterility in wheat in subtropical Asia: extent, causes and solution. ACIAR Proceedings No. 72., Australian Centre for International Agricultural Research, Canberra, ACT. pp. 8-12.

Rerkasem, B. and Jamjod. S. 1997. Boron deficiency induced male sterility in wheat (*Triticum aestivum* L.) and implications for plant breeding. Euphytica 96, 257-262.

Rerkasem, B. and Loneragan, J.F. 1994. Boron deficiency in two wheat genotypes in a warm, subtropical region. Agron. J. 86, 887-890.

Reuter, D.J. 1987. An Assessment of Wheat Nutrition Research in Bangladesh. A consultancy report for CIMMYT and CIDA. SAGRIC International. 45 p.

Sakal, R. and Singh, A.P. 1995. Boron research and agricultural production. *In* Tandon, H.L.S. (Ed.), Micronutrient research and agricultural production. Fertilizer Development and Consultation Organization, New Delhi, India, pp. 1-31.

Sarkar, A.K. and Chakraborty, S. 1980. Effect of micronutrient on yield of major field crops grown in the Gangetic delta region. Indian Agric. 24, 259-266.

Shorrocks, V.M. 1997. The occurrence and correction of boron deficiency. Plant Soil 193, 121-148.

Sillanpaa, M. 1982. Micronutrients and nutrient status of soils – a global study. FAO Soil Bulletin 48. FAO, Rome.

Singh, R.P., Sinha ,S.D. and Prasad, R.B.. 1976. Effect of boron on seed setting in wheat under North Bihar conditions. Indian J. Agron. 21, 100-101.

Subedi, K.D., Budhathoki, C.B. and Subedi, M. 1997. Variation in sterility among wheat (*Triticum aestivum*) cultivars in response to boron deficiency in Nepal. Euphytica 95, 21-26.

Subedi K.D., Budhathoki, C.B., Subedi, M. and Yubak, D.G.C.. 1996. Effect of sowing time and boron on sterility in four genotypes of wheat (*Triticum aestivum* L.) in the Western Hills of Nepal. In: Rawson, H.M. and Subedi, K.D. (Eds.), Sterility in wheat in subtropical Asia: extent, causes and solution. ACIAR Proceedings No. 72., Australian Centre for International Agricultural Research, Canberra, ACT., pp. 51-61.

Subedi, K.D., Greogory, P.J. and Gooding, M.J. 1999. Boron accumulation and partitioning in wheat cultivars with constrasting tolerance to boron deficiency. Plant Soil 214,141-152.

Tandon, J.P. and Naqvi, S.M.A. 1992. Wheat varietal screening for boron deficiency in India. In: Mann, C.E. and Rerkasem, B. (Eds.), Boron deficiency in wheat. CIMMYT Wheat Special Report No. 11, International Maize and Wheat Improvement Center, Mexico, D.F., pp. 76-78.

Topal, A., S. Gezgin, N. Akgün, N. Dursun and M. Babaoglu. 2002. Yield and yield attributes of durum wheat (*Triticum durum* Desf.) as affected by boron application. In: Goldbach, H. E., Rerkasem, B., Wimmer, M. A., Brown, P. H., Thellier, M., and Bell, R.W. (Eds.), All Aspects of Plant and Animal Boron Nutrition, Kluwer and Plenum Academic Publishers, Dordrecht, The Netherlands, pp. 401-406.

Yang, Y.H. 1992. Wheat boron deficiency in Yunnan, China. In: Mann, C.E. and Rerkasem, B. (Eds.), Boron deficiency in wheat. CIMMYT Wheat Special Report No. 11, International Maize and Wheat Improvement Center, Mexico, D.F., pp. 72-75.

www.cimmyt.org/Research/wheat Accessed February 1, 2002

Table 1. The effects of soil B levels (B) by genotypes (G) on the grain set index (GSI), the number of grain per ear, spikelets per ear and ears per m in bread wheat (4HTWYT, 17ESWYT), triticale (28ITYN) and durum (28IDYN).

	Bread wheat		Triticale	Durum
	4HTWYT	17ESWYT	28ITYN	28IDYN
df (B)	1	1	2	2
Error 1	1	1	2	2
df (G x B)	49	49	98	98
Error 2	98	98	152	152
GSI (%)				
B	***	*	***	***
G x B	*	*	*	*
Grains ear^{-1}				
B	***	*	***	**
G x B	*	*	*	*
Spikelets ear^{-1}				
B	NS	NS	NS	NS
G x B	NS	NS	*	NS
Ears m^{-1}				
B	NS	NS	NS	NS
G x B	NS	NS	NS	NS

*Significant at $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

NS not significant at $P < 0.05$.

Table 2. Frequency distribution of genotypes of bread wheat, triticale and durum by their grain set index (GSI) in soil in B0.1.†

Class by GSI in B0.1	Bread wheat		Triticale	Durum	Total
	4HTWYT	17ESWYT	28ITYN	28IDYN	
	Frequency (%)				
0-20%	14.3	14.3	51.0	81.6	40.3
21-50%	57.1	30.6	32.7	18.4	34.7
51-70%	12.2	30.6	16.3	0.0	14.8
71-85%	12.2	16.3	0.0	0.0	7.1
>85%	4.1	8.2	0.0	0.0	3.1
Total	49	49	49	49	196
†GSI (%±SE) of B efficient check, Fang 60					
	94.3±4.1	85.5±5.3	91.3±8.8	98.3±1.4	

Table 3. Responses to boron in the field in bread wheat, durum wheat and triticale, by B efficiency class.

Soil boron level	Class of genotypes, by GSI in B0.1					Nursery mean	Fang 60
	0-20%	21-50%	51-70%	71-85%	>85%		
	Grain set index (%)						
Bread wheat (4HTWYT)							
0.1	10.2a	35.8a	59.1a	79.3a	89.2a	42.5a	94.3a
0.2	71.8b	82.4b	84.4b	89.5b	95.4a	82.5b	98.5a
Bread wheat (17ESWYT)							
0.1	13.5a	35.3a	60.9a	77.5a	87.6a	51.6a	85.5a
0.2	71.5b	79.5b	85.7b	91.7b	95.5a	83.7b	99.0b
Triticale (28ITYN)							
0.1	9.9a	29.3a	57.6a	ne[†]	ne	24.7a	91.3a
0.2	62.5b	80.6b	84.5b	ne	ne	72.3b	98.2a
0.3	95.2c	95.1c	97.9c	ne	ne	95.6c	96.3a
Durum wheat (28IDYN)							
0.1	6.5a	31.4a	ne	ne	ne	12.8a	98.3a
0.2	54.7b	72.3b	ne	ne	ne	58.5b	100.0a
0.3	88.9c	95.8c	ne	ne	ne	90.3c	99.5a

Significant effect of B in each class of genotypes is denoted by different letters by means of LSD ($P < 0.05$)

[†]ne = no entry in this class

Table 4. Number of grains per ear in B0.1 relative to B sufficiency (± standard error) in different classes of genotypes of bread wheat, triticale and durum wheat grown in the field.

Class of genotypes, by GSI in B0.1	Bread wheat		Durum	Triticale
	4HTWYT	17ESWYT	28ITYN	28IDYN
	Number of grains ear^{-1} in B0.1 relative to B sufficiency†			
0-20%	0.20±0.11	0.33±0.08	0.10±0.01	0.06±0.08
21-50%	0.39±0.18	0.53±0.20	0.25±0.01	0.14±0.03
51-70%	0.69±0.15	0.57±0.18	0.45±0.07	ne
71-85%	0.80±0.18	0.60±0.11	ne‡	ne
>85%	0.97±0.19	0.75±0.14	ne	ne
Fang 60	0.94±0.26	1.01±0.04	1.05±0.27	1.14±0.05
Mean	0.49±0.29	0.56±0.29	0.22±0.17	0.06±0.10

† B sufficiency was B0.2 for bread wheat and B0.3 for durum wheat and triticale.

‡ne = no entry in this class

Table 5. Mean grain set index (GSI) of bread wheat, durum wheat and triticale lines in different boron efficiency classes in sand culture without added boron (B0), compared with a boron-efficient check, Fang 60.

Class by GSI in B0.1	Bread wheat 4HTWYT	Triticale 28ITYN	Durum wheat 28IDYN
	Grain set index in B0 (%±SD)		
0-20	28.4±20.2	10.4±16.7	8.3±11.5
21-50	58.6±18.6	17.0±18.2	10.3±10.0
51-70	63.5±20.0	39.1±26.4	ne†
71-85	67.3±19.8	ne	ne
>85	92.0±11.4	ne	ne
Nursery mean	57.2±22.7	17.9±23.9	10.6±16.6
Fang 60	95.0±7.1	96.4±3.2	97.2±4.0

†No entry in this class

Table 6. Grain set index (GSI) of various boron efficient and inefficient bread wheat lines in a soil low in B (0.1 mg HWS B kg^{-1}, B0.1) and sand culture without added B (B0). (Each GSI number is mean of two replicates, ±standard deviation)

Boron condition for screening		Soil (B0.1)	Sand (B0)
Nursery, entry no., variety or cross		Grain set index (%±SD)	
4HTWYT			
1	Fang 60	94.3±1.7	95.0±7.1
4	MOCHIS T 88	29.3±8.8	38.8±14.5
5	FASAN	3.6±12.0	8.3±18.0
12	PAT10/ALD//PAT72300/3/PVN/4/BOW	92.7±2.4	89.4±1.9
13	PAT10/ALD//PAT72300/3/PVN/4/BOW	85.8±0.4	91.6±3.4
15	TRAP#1/BOW	83.5±15.6	85.3±2.1
20	PRINIA	0±0.0	15.8±0.0
30	TIA.1	85.0±4.2	82.4±7.1
17ESWYT			
1	Fang 60	85.5±14.1	nd[†]
6	WEAVER	2.8±3.2	nd
7	CHIL/2*STAR	89.0±0.6	nd
9	TURACO/CHIL	17.8±1.8	nd
28	CHEN/AEGILOPS SQUARROSA (TAUS)//BCN	5.3±0.4	nd
31	STAR//KAUZ/PVN	87.0±1.4	nd
43	KAUZ*2//SAP/MON/3/KAUZ	88.3±11.0	nd
47	CPAN 3004	86.0±16.3	nd

[†]nd = no data

Table 7. Effect of B on ear size (number of spikelets per ear) in triticale grown in the field at different levels of B, with ranges in parentheses.

B response class†	Number of entries	B0.1	B0.2	B0.3
		Spikelets per ear		
Responsive	19	14.66 (12.7-15.4)	19.44 (14.8-25.5)	22.81 (19.4-27.9)
Non-responsive	30	18.04 (12.8-24.8)	20.73 (14.1-24.8)	20.29 (13.4-25.6)
Fang 60		12.75	14.10	13.40
LSD ($P < 0.05$)			5.84	

†Separation between responsive and non-responsive classes by means of the difference for each genotype between B0.1 and B0.3 in number of spikelets per ear that is either greater or less than the LSD.

Table 8. Frequency distribution (%) of boron efficiency classes in selected sets of CIMMYT 2000/01 international germplasm grown in soil with 0.1 mg HWS B kg^{-1}.

Grain set index class (%)	Bread wheat 33IBS-WYN	Bread wheat 18SA-WSN	Barley 28IBON†	Durum wheat 33IDYN	Triticale 33ITSN	Whole germ-plasm
	Frequency (%)					
< 20	36.4	23.0	37.0	34.7	42.8	35.4
20-49	25.0	23.0	39.0	51.0	25.5	29.2
50-74	8.0	28.7	24.0	14.3	27.5	19.5
75-90	20.6	17.2	0.0	0.0	4.1	11.0
> 90	10.1	8.0	0.0	0.0	0.0	4.9
Total	388	190	265	49	216	1108
GSI of B efficiency checks (%, with standard deviation in brackets)						
SW41	71.1±7.4	71.6±5.5	60.5±6.6‡	82.8±1.0	70.1±4.3	
Fang 60	97.2±0.8	96.8±1.2	84.3±2.3§	99.3±0.1	96.5±0.8	

† From CIMMYT/ICARDA

‡ BCMU96-9 and § BRB9604

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Genetic control of boron efficiency in wheat (*Triticum aestivum* L.)

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Key words: boron, deficiency, efficiency, wheat

Abstract

The genetic control of boron (B) efficiency in wheat (*Triticum aestivum* L.) was studied for three genotypes representing B inefficient (I, Bonza), moderately B inefficient (MI, SW 41) and B efficient (E, Fang 60) categories. Boron efficiency was expressed as a partially dominant character but the phenotypes of F_1 hybrids, relative to parents, indicated genetic control varied from recessive to additive to completely dominant with different cross combinations and B levels. Major genes were identified from the evaluation of F_2-derived F_3 populations derived from intercrosses between the three parents. Monogenic segregation was found in Bonza x SW 41 and SW 41 x Fang 60 crosses and digenic segregation resulted between Bonza x Fang 60 cross. Among the three wheat genotypes with widely different B efficiency, genetic variation for response to B could be accounted for by two genes, *Bo$_d$1* and *Bo$_d$2*.

Abbreviations: B, boron; E, efficient; GSI, grain set index; HWS, hot water soluble; I, inefficient; MI, moderately inefficient

Introduction

Low boron (B) soils are widespread in many subtropical wheat growing areas (Sillanpaa, 1982; Shorrocks, 1997). These include the northern region of Thailand, where wheat is being promoted (Rerkasem & Jamjod, 1989) and established wheat growing areas of China (Li et al., 1978), Bangladesh (Kataki et al., 2002), northwestern India (Sakal & Singh, 1995) and the Terai and mid-hills of Nepal (Subedi et al., 1996). Boron deficiency causes yield reduction by inducing male sterility, resulting in grain set failure (Rerkasem & Loneragan, 1994). A wide range of genotypic variation for response to low B has been identified and genotypes were classified into distinct B efficiency classes, namely, efficient, moderately efficient, moderately inefficient and inefficient (Rerkasem & Jamjod, 1997). Below a critical B level, inefficient genotypes were completely sterile and set no or just a few grains, while efficient genotypes set grain normally.

Genotypic variation offers a solution to sterility of wheat in low B soils. Boron efficient genotypes have been found to avoid sterility in the field in low B soils in Bangladesh, Nepal and northern Thailand where inefficient genotypes sustained serious yield losses (Rerkasem & Jamjod, 1989; Subedi et al., 1996; Kataki et al., 2002). Avoidance of B deficiency through selection and breeding for B efficient cultivars appears to be a promising approach for wheat. Furthermore, the response to low B in wheat is unique in that at certain levels of B deficiency male fertility, easily quantified as grain set, is the only adverse effect observed (Rerkasem et al., 1997). Wheat therefore offers a model plant on which the effect of B deficiency on male fertility may be studied without the confounding effects from other physiological

processes. However, the lack of understanding of genetic control of B efficiency traits involved in grain set hampers selection and breeding for B efficiency as well as genotypic variation studies. In this study, we evaluated responses to B of F_1 hybrids and F_2-derived F_3 populations in comparison to those of their B efficient, moderately inefficient and inefficient parents, to determine their genetic control.

Materials and Methods

Genetic materials

Fang 60 (B efficient; E, Jamjod et al., 1992), SW 41 (moderately B inefficient; MI, Rerkasem & Loneragan, 1994) and Bonza (B inefficient; I, Rerkasem & Jamjod, 1997) were used as parents. F_1 hybrid plants of three crosses, Bonza x SW 41, SW 41 x Fang 60 and Bonza x Fang 60, including reciprocal crosses were tested in Experiment 1 and sown in B sufficient soil to produce F_2 and F_3 generations. F_3 populations were tested in sand culture in Experiment 2. The first backcross population (BC_1) was made for the Bonza x SW 41 cross using Bonza as a recurrent parent, i.e. Bonza x (Bonza x SW 41), and used in Experiment 3.

Experiment 1: Evaluation of F_1 hybrids to B levels

Parents and F_1 populations were sown in a low B soil (0.1 mg hot water soluble B kg^{-1}) in the field at Chiang Mai University. The experiment was arranged as a split plot design with 3 replications. Three B levels were arranged in main plots, including nil (B0), limed at the rate 2 t ha^{-1} (BL), to accentuate B deficiency (Rerkasem & Jamjod,

1989) and boron at the rate 10 kg borax ha^{-1} (B+) applied to the soil. Three parents (Fang 60, SW 41 and Bonza) and their F_1 hybrids were arranged in sub plots. In each plot, 5 plants of each parent and F_1 were sown in single rows with 0.1 m between plants and 0.25 m between rows. At boot stage, the first two ears of each plant were bagged to prevent outcrossing. At maturity, the bagged ears were harvested and the effect of B deficiency quantified as the grain set index (GSI, percentage of the 20 basal florets from 10 central spikelets with grain; Rerkasem & Loneragan, 1994). Responses to B of parents and F_1 hybrids were compared by Duncan's Multiple Range Test (DMRT).

Experiment 2: Evaluation of F_2-derived F_3 families

Three populations of F_2-derived F_3 families examined included (Bonza x SW 41), (SW 41 x Fang 60) and (Bonza x Fang 60). Families were sown in freely drained earthenware pots (0.3 m diameter and 0.3 m deep) containing washed river quartz sand with no detectable available B. The sand was watered twice daily with an otherwise complete nutrient solution without added B (B0). The nutrient solution was adapted from Broughton & Dilworth (1971) and consisted of (μM): $CaCl_2$, 1000; $MgSO_4$, 250; KH_2PO_4, 500; FeEDTA, 10; K_2SO_4, 250; $MnSO_4$, 2; $ZnSO_4$, 0.5; $CuSO_4$, 0.2; $CoSO_4$, 0.1, Na_2MoO_2, 0.1 and KNO_3, 5000. This sand culture system has been successfully used in screening for B efficiency in wheat (Rerkasem & Jamjod, 1997) and barley (Jamjod & Rerkasem, 1999). In each pot, eleven plants per family or parental line were sown with one plant of the B inefficient genotype, Bonza, at the center, i.e. 12 plants per pot. The number of families for each combination were 85 for (Bonza x SW 41)F_3, 92 for (SW 41 x Fang 60)F_3 and 114 for (Bonza x Fang 60)F_3. Twelve pots of each parental line were included. At boot stage, the first

two ears from each plant were bagged. At maturity, the bagged ears were harvested and assessed for grain set with the GSI.

To classify families into each type of response to B, mean GSI and variance within family were calculated and related to those of the parents. With respect to B, the families were classified as homozygous efficient or homozygous inefficient when their mean and variance were in the range of either efficient or inefficient parents. Families with means outside the range of the parents but having a variance within the range of the parents were classified as homozygous intermediate. Families with a variance higher than those of parents were classified as segregating. Chi-square analysis was used for testing goodness of fit of the observed segregation ratio to the value expected for each of two models. Families for each population were tested for the monogenic ratio of 1 homozygous efficient : 2 segregating : 1 homozygous inefficient and for the digenic ratio of 1 homozygous efficient : 14 (homozygous intermediate + segregating) : 1 homozygous inefficient.

Experiment 3 Evaluation of backcross population

The backcross population (BC_1) of Bonza x (Bonza x SW 41), B inefficient recurrent parent; Bonza and donor parent, SW 41 were tested for response to B in sand culture without added B as described in Experiment 2. One hundred of BC_1F_1 plants and 50 plants of each parent were sown. At boot stage, the first two ears from each plant were bagged. At maturity, plants were harvested and grain set determined based on the GSI of bagged ears. Chi-square tests were conducted to examine the segregation ratio of the backcross population with the expected ratio of 1 Bonza type to1 heterozygote.

Results

Response of F_1 hybrids

There was a significant difference in the response to B among the three wheat genotypes and their F_1 hybrids (Table 1). The GSI in all genotypes exceeded 80% when B was applied (B+). The parents differed in GSI in response to the lower B treatments, BL and B0. Fang 60 was the most efficient and did not show an adverse effect of B deficiency. SW 41 showed an intermediate B efficiency, with GSI of 58.3% in B0 and 44.2% in BL. With GSI of 32.9% in B0 and 0.2% in BL, Bonza was the most inefficient.

The GSI of F_1 hybrids relative to their parents varied with the parental combinations and B treatments. There was no significant difference between reciprocal crosses at each B level.

With the BL and B0 treatments the GSI of the F_1 hybrids from SW 41 x Fang 60 combinations were not significantly different from the efficient parent, Fang 60. Those from Bonza x Fang 60 combinations were intermediate between the two parents but closer to Fang 60 at BL and not different from Fang 60 at B0. In contrast, F_1 hybrids of Bonza x SW 41 were not significantly different from the inefficient parent (Bonza) when grown at BL and were similar to the two parents at B0.

Response of F_2-derived F_3 populations

As in low B soil, the three parental lines and their F_3 populations tested in this study showed a large range of responses to low B in sand culture (Table 2). Mean GSI of Bonza, SW 41 and Fang 60 parental lines were between 0-3, 33-69 and 87-99%, respectively. Mean GSI of F_2 derived families were within the range of their respective parents. Within line variances of the inefficient Bonza and efficient Fang 60 parents were between 0-189 while those of the intermediate, moderately inefficient SW 41 parents were between 657-1328. Within family variances of the F_3 varied from 0 to 2508 (Table 2).

Means and variances of F_3 families for each cross were compared to parents and classified into homozygous efficient, segregating, homozygous intermediate and homozygous inefficient types. Families from Bonza x SW 41 and SW 41 x Fang 60 could not be clearly classified into homozygous SW 41 and segregating classes due to high phenotypic variance within the SW 41 parent (Figure 1a and 1b). Therefore, families were classified into homozygous inefficient (I) and segregating plus homozygous moderately inefficient (Seg + MI) for the Bonza x SW 41 cross and segregating plus homozygous moderately inefficient (Seg + MI) and homozygous efficient (E) for the SW 41 x Fang 60 cross (Figure 1c). Chi-square analysis demonstrated that segregation of F_3 families from the crosses Bonza x SW 41 and SW 41 x Fang 60 was consistent with the single gene model (Table 3). Segregation of F_3 families from the cross Bonza x Fang 60 deviated from the monogenic ratio and was consistent with a two gene model (Table 3).

Response of backcross population

When tested in B0, the inefficient recurrent parent, Bonza set no grain (Figure 2). The GSI of the donor SW 41 parent ranged between 25% and >75%. Nearly 50% of the BC_1 population expressed the same GSI as the recurrent parent. Chi-square analysis was consistent with 1:1 ratio of Bonza type: heterozygote ($\chi 2 = 1.17$, $p = 0.5945$).

Discussion

The segregation patterns of F_2-derived F_3 families and the BC_1 population between parents with contrasting levels of B efficiency were consistent with B efficiency being under the control of major genes. The full response range of the three parents was shown to be controlled by two major independent loci, with the efficient Fang 60 having efficiency alleles at both loci and the moderately inefficient SW 41 having efficiency alleles at one locus. The B inefficient Bonza is expected to have alleles for inefficiency at both loci. If the gene symbol *Bo$_d$* is assigned to the loci controlling B efficiency, the proposed genotypes of these parental lines are as follows: Fang 60 *Bo$_d$1Bo$_d$1 Bo$_d$2Bo$_d$2*, SW 41 *bo$_d$1bo$_d$1 Bo$_d$2Bo$_d$2* and Bonza *bo$_d$1bo$_d$1 bo$_d$2bo$_d$2*. This study presents the first clear evidence of genes controlling B efficiency in wheat, although there have been reports in other crops. For example, Xu et al. (2001) suggested one major gene controlled B efficiency in oil seed rape, but that possibly another three minor genes were also associated with B efficiency. Major gene control of B efficiency has also been reported for celery (Pope & Munger, 1953), tomato (Wall & Andrus, 1962) and red beet (Tehrani et al., 1971).

The results of this study suggested that the responses of F_1 hybrids compared to parents varied with B treatments and parent combinations. Responses of the F_1 from the Bonza x SW 41 were close to the B inefficient Bonza at BL and intermediate to the two parents at B0, suggesting recessive and additive control of B efficiency, respectively. In contrast, for the crosses SW 41 x Fang 60 and Bonza x Fang 60 at BL and B0, the GSI of the F_1 hybrids were close to the B efficient Fang 60, indicating completely dominant control of B efficiency. The changing response of F_1 hybrids according to their parental combinations and treatments was also found in the study of tolerance to high concentrations of B (Paull et al., 1991). These responses were consistent with the hypothesis of Knight (1973) that for a quantitative trait the response of an F_1 hybrid relative to its parents, will vary according to the environmental conditions.

The nature of the expression of B efficiency will influence the level of the treatments selected for screening segregating populations in a breeding programme. For example, B0 in sand culture was able to identify homozygous inefficient genotype but could not differ heterozygote from homozygous moderately inefficient genotypes from an I x MI cross (Figure 2) whereas BL in soil suppressed grain set of both genotypes (Table 1). However, low levels of B could not be used to discriminate heterozygotes and homozygous efficient (E) genotypes when screening an F_2 and other segregating generations due to the dominant gene action (Table 1, Ngorien, 1999). Progeny testing, as shown in the study of F_3 (Figure 1), is suggested for screening segregating populations involving B efficient (E) parents.

As B efficiency in wheat is controlled by major genes, the backcross method is the most efficient way for transferring B efficiency into locally adapted, inefficient varieties. Backcross-derived lines between SW 41 (Bo_d2) as the donor parent and Bonza as the recurrent parent are now being developed at Chiang Mai University and will be evaluated for yield advantage in the next growing season. The Bonza derivatives carrying Bo_d2 will be selected and following further crosses with Fang 60 as a donor parent, lines carrying Bo_d1 and Bo_d2 will be developed for evaluation under low B conditions.

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References

Broughton, W.J. & M.J. Dilworth, 1971. Control of leghaemoglobin synthesis in snake bean. Biochem. J. 125: 1075-1080.

Jamjod, S., & B. Rerkasem, 1999. Genotypic variation in response of barley to boron deficiency. Plant and Soil 215: 65-72.

Jamjod, S., C.E. Mann & B. Rerkasem, 1992. Screening for boron deficiency in wheat. In: C.E. Mann & B. Rerkasem (Eds.), Boron Deficiency in Wheat, pp. 79-82. CIMMYT What Special Report No. 11, CIMMYT, Mexico.

Kataki, P.K., S.P. Srivastava, M. Saifuzzaman & H.K. Upreti, 2002. Sterility of wheat and response of field crops to applied boron in the Indo-Gangetic Plains. J. Crop Prod 4: 133-165.

Knight, R., 1973. The relation between hybrid vigour and genotype-environment interactions. Theo. Appl. Genet. 43: 311-318.

Li, B.H., W.H. Li, M.C. Kui, W.S. Chao, H.P. Jern, C.R. Li, W.J. Chu & C.L.Wang, 1978. Studies on cause of sterility of wheat. J. Northeastern Coll. 3: 1-19 (*in Chinese with English translation*).

Ngorien, S. 1999. Response of segregating wheat populations to boron deficiency. MS. Thesis, Chiang Mai University, Chiang Mai, Thailand. 68 p.

Paull, J.G., A.J. Rathjen & B. Cartwright, 1991. Major genes control of tolerance of bread wheat (*Triticum aestivum* L.) to high concentrations of boron. Euphytica 55: 217-228.

Pope, D.T. & H.M. Munger, 1953. The inheritance of susceptibility to boron deficiency in celery. Proc. Am. Soc. Hortic. Sci. 61: 481-486.

Rerkasem, B. & J. F. Loneragan, 1994. Boron deficiency in two wheat genotypes in a warm subtropical region. Agron. J. 85: 887-890.

Rerkasem, B. & S. Jamjod, 1997. Boron deficiency induced male sterility in wheat (*Triticum aestivum* L.) and implications for plant breeding. Euphytica 96: 257-262.

Rerkasem, B. & S. Jamjod, 1989. Correcting boron deficiency induced ear sterility in wheat and barley. Thai J. Soils and Fertilizers 11: 200-209. (*in Thai with English summary*).

Rerkasem, B. Lordkaew, S. & Dell, B. 1997. Boron requirement for reproductive development in wheat. Soil Sci. Plant Nutr.43: 953-957.

Sakal, R. & A.P. Singh, 1995.Boron research and agricultural production. In:H.L.S. Tandon (ed.), Micronutrient Research and Agricultural Production, pp. 1-31. Fertilizer Development and Consultation Organization, New Delhi, India.

Shorrocks, V.M. 1997. The occurrence and correction of boron deficiency. Plant and Soil 193: 121-148.

Sillanpaa, M. 1982. Micronutrients and nutrient status of soils. FAO Bull. 48, FAO, Rome.

Subedi, K.D., C.B. Budhathoki, M. Subedi & D.G.C. Yubak, 1996. Effect of sowing time and boron on sterility in four genotypes of wheat (*Triticum aestivum* L.) in the Western hills of Nepal. In: H.M. Rawson & K.D. Subedi (eds.), Sterility in Wheat in Subtropical Asia: Extent, Causes and Solution, ACIAR Proceedings No. 72, pp. 51-61. ACIAR, Canberra.

Tehrani, G., H.M. Munger, R.W. Robinson & S. Shannon, 1971. Inheritance and physiology of response to low boron in red beet (*Beta vulgaris* L.). J. Am. Soc. Hortic. Sci. 96: 226-230.

Wall, J.R. & C.F. Andrus, 1962. The inheritance and physiology of boron response in the tomato. Am. J. Bot. 49: 758-762.

Xu, F.S., Y.H. Wang & J. Meng, 2001. Mapping boron efficiency gene(s) in *Brassica napus* using RFLP and AFLP markers. Plant Breeding 120: 319-324.

Table 1. GSI (%) of three parents, F_1 hybrids and reciprocal crosses in response to B levels. BL = limed soil with no added B; B0 = unlimed soil with no added B; B+ = 10 kg Borax ha^{-1} added to unlimed soil.

Parents/F_1 hybrids	B treatments[a]		
	BL	B0	B+
Bonza	0.2 d	32.9 b	86.0 a
SW 41	44.2 c	58.3 b	90.8 a
Fang 60	98.1 a	99.1 a	97.9 a
Bonza x SW 41	3.3 d	46.0 b	80.6 a
SW 41 x Bonza	9.7 d	38.7 b	94.2 a
Bonza x Fang 60	65.8 bc	91.5 a	96.9 a
Fang 60 x Bonza	87.5 ab	97.2 a	97.8 a
SW 41 x Fang 60	93.2 a	97.0 a	93.6 a
Fang 60 x SW 41	93.1 a	98.6 a	94.7 a
F-test	B^{**}, G^{***}, BxG^{***}		

** and *** Significant at 0.01 and 0.001 probability levels, respectively.

[a] Means within a column with the same letter do not differ significantly at 5% level with Duncan's Multiple Range Test.

Table 2. Range of mean GSI (%) and variance within family/line of 3 parents and F_2-derived families, from 3 crosses grown in sand culture without added B (B0). Values are based on 11 plants per F_2-derived family.

Line/Family		Mean GSI (%)			Variance		
	n	Min	Mean	Max	Min	Mean	Max
Parents							
Bonza	12	0	1	3	0	12	51
SW 41	10	33	52	69	657	953	1328
Fang 60	12	87	94	99	4	75	189
F_3 families							
(Bonza x SW 41) F_3	85	0	17	71	0	466	1907
(SW 41 x Fang 60) F_3	92	30	69	98	10	625	1586
(Bonza x Fang 60) F_3	114	0	41	100	0	759	2508

n = number of parental lines or F_3 families

Table 3. Chi-square analysis of the responses to B deficiency of F_2-derived F_3 families from three crosses grown in sand culture without added B.

Cross	Model[a]		Number of families[b]			$\chi 2$	P
			I	Seg+MI			
Bonza x SW 41	1:3	Exp.	21.25	63.75		0.18	0.6611
	1:15	Exp.	5.31	79.69		18.65	<0.001
		Obs.	23	62			
				Seg+MI	E		
SW 41 x Fang 60	3:1	Exp.		69	23	1.06	0.3355
	15:1	Exp.		86.25	5.75	11.65	<0.001
		Obs.		73	19		
			I	Seg+Int.	E		
Bonza x Fang 60	1:2:1	Exp.	28.5	57	28.5	61.34	<0.001
	1:14:1	Exp.	7.125	99.75	7.125	5.26	0.078
		Obs.	12	92	10		

[a] Exp. – expected ratio; Obs. – observed ratio.

[b] E = homozygous efficient, MI = homozygous moderately inefficient, I = homozygous inefficient, Seg = segregating, Int. = homozygous intermediate.

Figure 1. Mean GSI (%) and variance within families of F_2-derived F_3 families and parents grown in sand culture without added B. Note: Mean GSI and variance of Bonza parent were both 0 and obscured by F_3 families' data.

a) Bonza x SW 41

b) SW 41 x Fang 60

c) Bonza x Fang 60

Figure 2. Response of GSI (%) of backcross population (BC_1), recurrent (Bonza) and donor (SW 41) parents grown in sand culture without added B.

Figure 1.

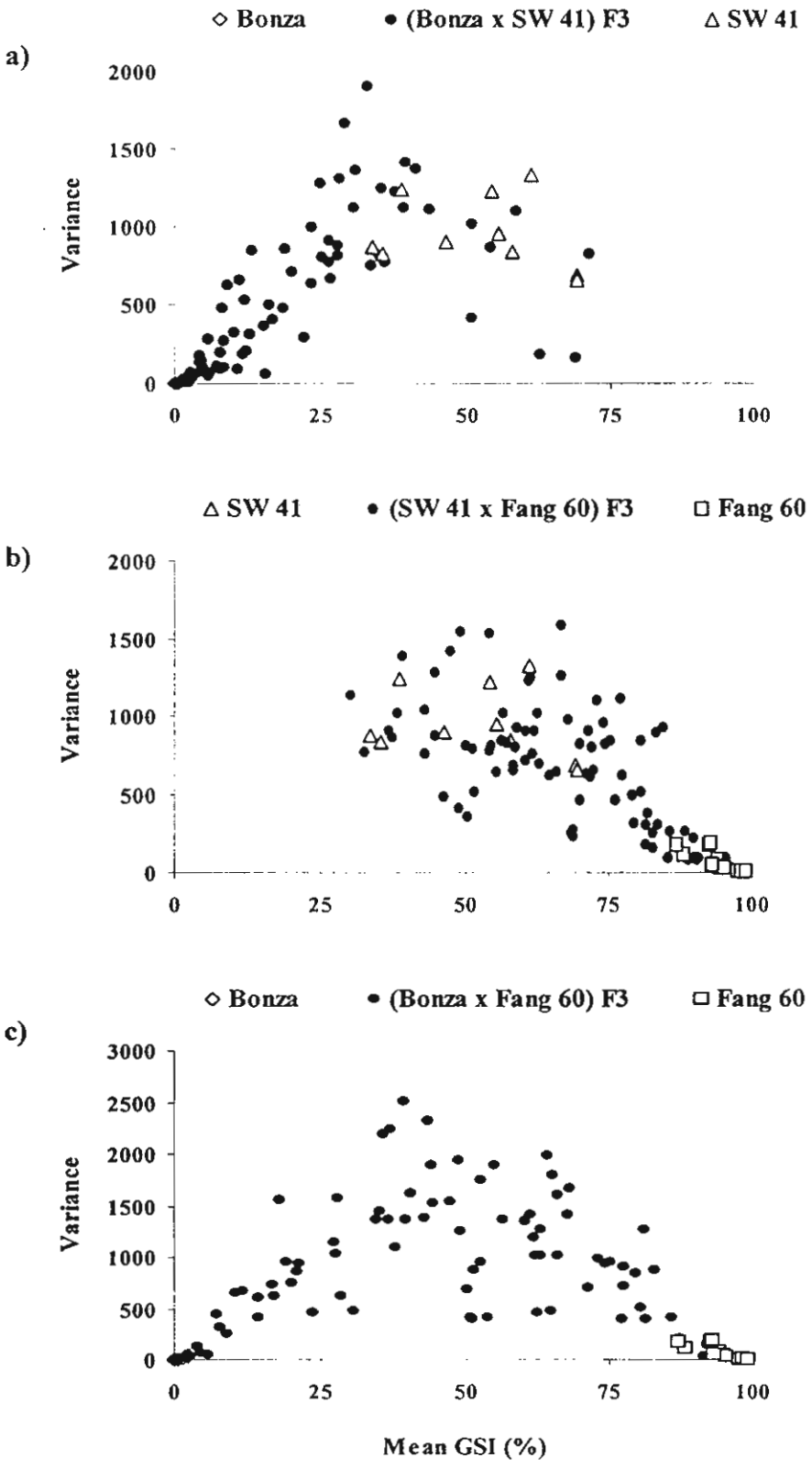
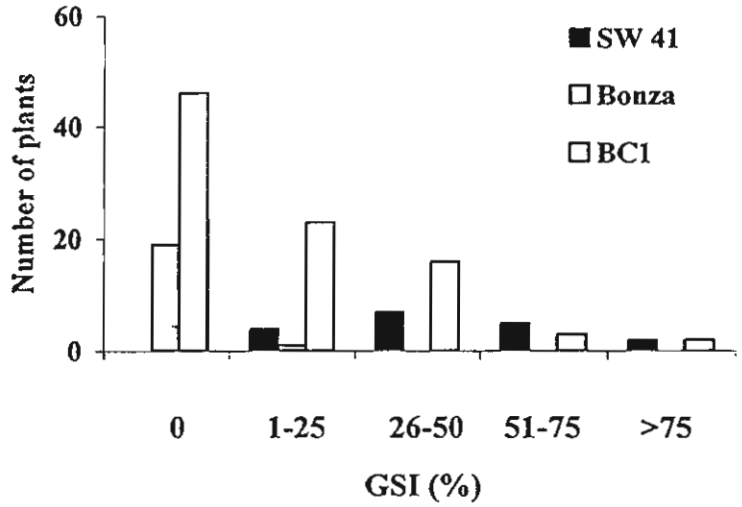

◇ Bonza
• (Bonza x SW 41) F3
△ SW 41
a)
Variance
2000
1500
1000
500
0
0
25
50
75
100
△ SW 41
• (SW 41 x Fang 60) F3
□ Fang 60
b)
Variance
2000
1500
1000
500
0
0
25
50
75
100
◇ Bonza
• (Bonza x Fang 60) F3
□ Fang 60
c)
Variance
3000
2500
2000
1500
1000
500
0
0
25
50
75
100
Mean GSI (%)

Figure 2.


60
40
20
0
Number of plants
SW 41
Bonza
BC1
0
1-25
26-50
51-75
>75
GSI (%)

Contrasting responses to boron deficiency in barley and wheat

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Keywords: Barley, Boron deficiency, Boron efficiency, Genotypic variation, Wheat

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Contrasting responses to boron deficiency in barley and wheat

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Abstract

To determine if boron (B) deficiency, commonly reported to depress grain set in wheat, has the same effect in barley, a set of experiments compared five wheat and seven barley genotypes at various B levels in sand culture and in the field. In sand culture, plants were grown with levels of added B, from 0 to 10 μM. In the field, they were sown in a low B soil [0.15 mg hot water soluble (HWS) B kg^{-1}] with three B treatments (nil, 2 t lime ha^{-1}, 1 kg B ha^{-1}). In sand culture without added B, the genotypes ranged in grain set index (GSI) from 0 to 93 % for wheat and 0 to 67 % for barley. Boron concentration of the spike and flag leaf at booting in wheat and barley correlated ($r = 0.8 - 0.9$, $p < 0.01$) with the effect of B on GSI. Grain set was the only response, measurable in decreased number of grain $spike^{-1}$ and grains $spikelet^{-1}$, to low B in wheat. In barley, low B also depressed the number of spikelet $spike^{-1}$ by 23 to 75 % and induced a "rat-tail" symptom of terminal spikelet degeneration. There was a weak correlation between spike and flag leaf B and the effect of B on spike size in barley ($r = 0.47$ and 0.37, respectively, $p < 0.1$). In some barley genotypes, the low B level that depressed grain set sometimes also delayed spike emergence and depressed the number of spikes $plant^{-1}$ but sometimes increased tillering and dry weight of straw. These results demonstrate that the phenotype of plant response to low B is more complex in barley than wheat and may require different strategies for managing B nutrition of barley including different approaches for selecting B efficient genotypes.

Introduction

Although it is often reported that cereals have low sensitivity to B deficiency (Marten and Westerman, 1991; Shorrocks, 1997), wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.) can be adversely affected by B deficiency in the field. In both species, B deficiency depresses male fertility, resulting in grain set failure (e.g. see Li et al., 1978 and da Silva and de Andrade, 1983 for wheat and Ambak and Tadano, 1991 for barley). Genotypic variation in the response to low B supply has been reported in wheat (Rerkasem and Jamjod, 1997) and barley (Jamjod and Rerkasem, 1999). Those genotypes that are able to grow and yield well in soils in which other genotypes are adversely affected by B deficiency have been called B-efficient genotypes (Rerkasem and Jamjod, 1997). In wheat, genotypic variation to B has only been expressed in reproductive growth (Rerkasem and Loneragan, 1994), whereas in barley, it has been reported in both reproductive and vegetative growth. In barley, however, the B response is somewhat confusing as B deficiency that depresses male fertility and grain set has been reported to either enhance (Ambak and Tadano, 1991) or depress (Jamjod and Rerkasem, 1999) vegetative growth. This study aims to determine how differently barley and wheat phenotypes respond to B deficiency by comparing a range of wheat and barley genotypes under defined conditions of B supply. This information has relevance for managing B nutrition in the field and also for selecting B-efficient genotypes in the future.

Materials and Methods

Barley and wheat genotypes covering a wide range of B efficiency (Jamjod and Rerkasem, 1999, Anantawiroon et al., 1997) were compared at various levels of B supply in two sand culture experiments, and one experiment in the field and sand culture in Chiang Mai, Thailand. These genotypes have been identified from extensive screening of germplasm from the International Maize and Wheat Improvement Center (CIMMYT). In experiment 1, three genotypes of two-row barley (BRB 9, BCMU 96-9 and CMBL 92029) and wheat (Fang 60, SW 41 and Tatiara) were grown in sand culture with three levels of added B, 0, 0.1, 0.3 and 5 μM B. Experiment 2, also in sand culture, compared one genotype of barley (BRB 9) with one wheat (SW 41) at 0 and 10 μM B. Experiment 3 evaluated seven

genotypes of two row (BRB 9604, BRB 9, BCMU 96-9, and Stirling) and six row (BRB 2, FNBL 8309 and LARTC 9408) barley and five genotypes of wheat (Fang 60, Flycatcher, SW 41, Bonza and Tatiara) in sand culture with two levels of added B, 0 and 10 μM. All of the sand culture experiments were arranged in three replicates. The same genotypes were also grown in the field as subplots in main plots of three B levels: BL (soil limed at 2 t ha^{-1} to accentuate B deficiency), B0 (nil) and B10 (10 kg borax ha^{-1}), arranged in four replicated blocks.

In the sand culture, plants were grown in earthenware pots (30 cm in diameter, 30 cm deep) containing washed river quartz sand. Each pot was watered twice daily with 1 liter of nutrient solution containing varying levels of added B as described above, and other nutrients including 1000 μM $CaCl_2$, 500 μM KH_2PO_4, 250 μM $MgSO_4$, 250 μM K_2SO_4, 10 μM Fe EDTA, 1μM $MnSO_4$, 0.5 μM $ZnSO_4$, 0.2 μM $CuSO_4$, 0.1 μM Na_2MoO_4 (Broughton and Dilworth, 1971) and 5 mM KNO_3. This nutrient solution has been successfully used to screen barley (Jamjod and Rerkasem, 1999) and wheat germplasm (Rerkasem and Jamjod, 1997). The field experiment was on a Tropaqualf sandy loam soil of San Sai series with 0.15 mg hot water soluble (HWS) B kg^{-1}. The subplot containing each genotype consisted of 4 x 1 m rows, with 20 cm between rows. For experiments 1 and 2, there was one complete set of pots for each of two harvests. The first harvest, at boot stage (GS 45, Zadok et al., 1974), was for determination of spike and flag leaf B. Plant samples were dried, dry ashed and B concentration determined by the azomethine-H method (Lohse, 1982). Grain set and the number of spikelets $spike^{-1}$ were determined, at maturity in all experiments, on all plants and tillers in each pot in sand culture, from 20 randomly selected spikes from the 0.6 m middle section of two central rows in the field experiment. Grain set in wheat was assessed by the grain set index (GSI), percentage grain set, in the first two florets of 10 central spikelets (Rerkasem and Loneragan, 1994). The GSI in barley was assessed in the two side florets of the central spikelets for six-row barley, and the single fertile median florets in two-row barley (Jamjod and Rerkasem, 1999). Other effects of B that were recorded included number of tillers $plant^{-1}$, spikes $plant^{-1}$, spikelets $spike^{-1}$, days to floral initiation and spike emergence, shoot dry weight at boot stage and grain yield.

Results and discussion

There were both similarities and major differences in the response to low B of barley and wheat. Although B deficiency was moderate in the field and extreme in the pot trials, for the most part, trends were similar across experiments. In the sand culture without added B, grain set measured with GSI ranged from 0 to >90% in wheat and from 0 to 67% in barley genotypes (Tables 1, 2). The two most tolerant genotypes were wheat, Fang 60 and Flycatcher (Table 2). They already had GSI that exceeded 90% even without added B, while increasing B increased grain set in the other wheat and all barley genotypes. Most sensitive to B deficiency were Bonza, Tatiara (wheat) and Stirling (barley), which consistently had the lowest grain set in the lower B levels in pots and in the field (Tables 1 and 2). None of the barley genotypes was as tolerant as Fang 60, the most tolerant wheat genotype. In addition to its adverse effect on grain set, B deficiency depressed the number of spikelets spike^{-1} in barley by 21 to 75 % but not in wheat (Tables 3, 4, 5). Barley spikes that were B deficient exhibited the "rat-tail" symptom (Figure 1e) in which terminal spikelets had degenerated into wisps of white papery tissues similar to copper deficiency (Snowball and Robson, 1983). The lack of response to low B of the spikelet number in wheat is consistent with previous reports where a large number of genotypes have been evaluated under a wide range of conditions (e.g. Subedi et al. 1997; Anantawiroon et al. 1997).

In many of the barley genotypes and one of the wheat (Tatiara), tillering was increased at the level of B that depressed grain set (Tables 4, 5). This stimulating effect of low B was observed even when grain set was completely suppressed. That reproductive growth can be limited by B deficiency, at a level that is not limiting to vegetative growth, has been previously reported for both barley (Ambak and Tadano, 1991; Jamjod and Rerkasem, 1999) and wheat (Rerkasem et al., 1997). The increased tillering in low B was reflected in a significant increase in plant dry weight. However, these extra tillers were largely barren. The number of spikes plant^{-1} was actually depressed by B deficiency in some of the barley genotypes, by up to 90%. The extra tillers developed into extra spikes only occasionally, in BRB 9 barley in experiment 2 and Tatiara wheat in experiment 3. Boron deficiency delayed spike emergence in some barley but in none of the wheat genotypes (Tables 4, 5). Boron deficiency delaying spike emergence in barley has been previously reported (Phasook, 2000). The author also showed that there was no effect of B deficiency on the rate of barley development to the double ridge stage of floral initiation. Apparently unrelated to the

induction of the reproductive primordia, the delay in spike emergence in barley is likely to be associated with arrested subsequent ontogeny of the primordia, leading to abnormal morphology (Figure 1b-d). The relationship between various primary and secondary reproductive and vegetative responses to low B in barley may not be direct and straightforward. Responses to B in terms of grain set did not correlate with relative responses to B in the number of tillers plant^{-1}, spikes plant^{-1} and spikelets plant^{-1} ($p < 0.05$)

Boron concentration in the flag leaf and spike of wheat and barley at boot stage (Tables 6, 7 and 8) correlated well with the effect of B on their GSI ($r = 0.8 - 0.9$, $p < 0.01$). However, the grain set response to B could be predicted by tissue B only in some genotypes. The more severe effect of low B on grain set in Tatiara was clearly associated with lower B concentrations in its flag leaf and spike. The difference was less clear-cut between Fang 60 and SW 41, and among the barley genotypes covering a relatively narrow range of sensitivity to B deficiency. Previous reports also found SW 41 to be indistinguishable from Sonora 64, another B efficient wheat genotype (Rerkasem and Loneragan, 1994) as well as from Fang 60 (Subedi et al., 1999) by their tissue B concentration. Boron deficiency depresses grain set in wheat by adversely affecting pollen development and stigma function (Rerkasem et al., 1993), and the same possibly applies in barley. The grain set response to B in wheat correlated more closely with B concentration in the anther and carpel than in the secondary reproductive parts of the spike such as lemmas and palea (Rerkasem et al., 1997). Thus anthers and carpel B may better predict grain set response in different genotypes than in leaf and ear.

Wheat and barley had the same inflorescence type and are similar in the enclosure of developing spikes within leaf sheaths. The differential effects of B on spikelet number in wheat and barley were not reflected in their tissue B concentration. The species had about the same range of B concentrations in the spike and flag leaf (Tables 6, 7, 8) at the booting stage, while B deficiency depressed spikelet and spike number only in barley and not in wheat. Among the barley genotypes, the effect of B on spikelet number in barley correlated weakly ($p < 0.1$) with spike ($r = 0.47$) and flag leaf B ($r = 0.37$). The response to low B in spikelet number may be related to differences in B requirement for spikelet development or the ability to supply B to the growing spike inside the leaf sheath. Wheat could be more effective in transporting B into developing spikes than barley. However, for wheat genotypes most sensitive to low B in term of grain set such as Bonza and Tatiara this would have to happen in spite of a very poor ability to supply B for development and function of

the primary reproductive organs, anthers and carpel. There is a possibility that differential demand or supply for B may be related to specific parts of the developing spike and spikelets, at critical stages of their development.

While mechanisms behind the difference between wheat and barley remain to be explained, agronomic implications of the findings are clear. In both wheat and barley, the role of B is critical for grain yield and consideration needs to be given to the time taken for fertilizer B to enter critical sites in the spike at critical times. Wheat grain yield is depressed by B deficiency primarily through grain set. The effect may be simply predicted by the GSI, which may be used to evaluate large numbers of wheat lines for B efficiency (Anantawiroon et al., 1997; Rerkasem and Jamjod, 2001). In barley, in contrast, the effect of B on grain set may be attenuated by other responses, depending on the genotype. The combined effect of low B on grain set and spikelet number on the number of grain spike^{-1} and grain yield in BRB 9 barley (Table 5) is likely to be found in genotypes that behave like BRB 9604 and BRB 2 (Table 4). For other genotypes the grain yield may be depressed even further through the adverse effect of low B on the number of spike plant^{-1}, as in BCMU 96-9, FNBL 8309 and LARTC 9408. However, a particularly severe effect of B deficiency (e.g. grain set in BRB 2 and Stirling and the number of spike plant^{-1} in BCMU 96-9), may override all other responses. The GSI is used to evaluate for B efficiency without the need for a B sufficiency control, the degree of severity in B deficiency is indicated by a set of B efficiency (Anantawiroon et al., 1997). The index could also be used in a first round identification of those barley genotypes that are most sensitive to B deficiency in grain set. However, genotypes in which grain set is tolerant to low B will need to be assessed for other responses such as spikelet number and spike number, which can be done only in comparison with a B sufficiency control. Considerable savings may be achieved by such two-step screening especially with germplasms with a high proportion of genotypes in which grain set is sensitive to B deficiency. The rat-tail symptom provides a useful visual marker for the more severe effect of B deficiency on spikelet number.

This paper has shown that the phenotypic response to B deficiency of barley is more complex than that in wheat. Different strategies for managing B nutrition are required, including different approaches to select for B efficient genotypes.

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References

Ambak K and Tadano T 1991 Effect of micronutrient application on the growth and occurrence of sterility in barley and rice in a Malaysian deep peat soil. Soil Sci. Plant Nutr. 37, 715-724.

Anantawiroon P, Subedi K D and Rerkasem B 1997 Screening wheat for boron efficiency. *In* Boron in Soils and Plants. Dds R W Bell and B Rerkasem. Kluwer Academic Publishers, The Netherlands. pp 101-104.

Broughton W J and Dilworth M J 1971 Control of leghaemoglobin synthesis is snake beans. Biochem. J. 125, 1075-1080.

da Silva A R and de Andrade J M V 1983 Influence of micronutrients on the male steriltiy, on upland wheat and on rice and soybean yields, in red-yellow latosol. Pesq. agropec. bras., Brasilia 18, 593-601. (In Portuguese with English Abstract)

Gupta U C 1993 Introduction in boron and its role in crop production. Ed U C Gupta. P1. CRC Press, Boca Raton, FfL, USA.

Jamjod S and Rerkasem B 1999 Genotypic variation in response of barley to boron deficiency. Plant Soil. 215, 65-72.

Li B H, Li W H, Kui M C, Chao W S, Jern H P, Li C R, Chu W J and Wang C L 1978 Studites on cause of sterility of wheat. Journal of Northeastern Agricultural College. 3, 1-19.

Lohse G 1982 Microanalytical azomethine-H method for boron determination in plant tissues. Commun. Soil Sci. Plant Anal. 13,127134.

Marten J M and Westermann D T 1991 Fertilizer applications for correcting micronutrient deficiencies. *In* Micronutrients in Agriculture. 2nd ed. Eds. J J Mordtvedt, F R Cox, L M Shuman and R M Welch. pp. 549-592. SSSA Book Series no. 4. SSSA, Madison, WI.

Phasook T 2000 Boron efficiency in two barley genotypes. M.Sc. (Agronomy) Thesis. Graduate School, Chiang Mai University, Thailand.

Rerkasem B and Jamjod S 1989 Correcting boron deficiency induced spike sterility I wheat and barley. Thai J. Soils and Fertilizers 11, 200-209 (in Thai with English summary)

Rerkasem B and Jamjod S 1997 Boron deficiency induced male sterility in wheat (*Triticum aestivum* L.) and implications for plant breeding. Euphytica 96, 257-262.

Rerkasem B and Jamjod S 2001 Overcoming wheat sterility problem with boron efficiency.

In Plant nutrition-Food security and sustainability of agro-ecosystems. Eds WJ Horst, M K Schenk, A Bürkert, N Claassen, H Flessa, W B Frommer, H Goldbach, H-W Olfs, V Römheld, B Sattelmacher, U Schmidhalter, Shubert, N v Wirén and L wittenmayer. Kluwer Academic Publishers, The Netherlands. pp 82-83.

Rerkasem B and Loneragan J F 1994 Boron deficiency in two wheat genotypes in a warm, subtropical region. Agron. J. 86, 887-890.

Rerkasem B, Netsangtip R, Lordkaew S and Cheng CH 1993 Grain set failure in boron deficiency wheat. Plant Soil. 155/156, 309-312.

Rerkasem B, Lordkaew S and Dell B 1997 Boron requirement for reproductive in wheat. Soil Sci. Plant Nutr. 43, 953-957.

Shorrocks V M 1997 The occurrence and correction of boron deficiency. Plant Soil. 193, 121-148.

Snowball K and Robson A D 1983 Symptoms of Nutrient Deficiencies. Department of Soil Science and Plant Nutrition. Institute of Agriculture, University of Western Australia.

Subedi K D, C B Budhathoki and M Subedi 1997 Variation in sterility among wheat (*Triticum aestivum* L.) genotypes in response to boron deficiency in Nepal. Euphytica. 95, 21-26.

Subedi K D, P J Gregory and M J Gooding 1999 Boron accumulation and partitioning in wheat cultivars with contrasting tolerance to boron deficiency. Plant Soil. 214 (1-2), 141-152.

Zadok J C, Cheng T T and Konzak C F 1974 A decimal code for the growth stages of cereals. Weed Res. 14, 415-420.

Table 1. Response in Grain Set Index (%) in two-row barley and wheat to boron supply in sand culture (Experiment 1).

Species/genotype	Added B (μM)			
	0	0.1	0.3	5.0
Barley				
BRB 9	32.2 aD	57.5 bD	67.6 cC	76.7 dC
BCMU 96-9	23.4 aC	24.8 aC	28.4 aB	54.6 bB
CMBL 92029	12.5 aB	13.5 aB	22.0 bB	41.6 cA
Wheat				
Fang 60	94.8 aF	94.4 aF	98.7 aE	98.5 aD
SW 41	67.3 aE	74.6 bE	77.4 bcD	81.9 cC
Tatiara	0.2 aA	1.9 aA	13.0 bA	61.2 cB
Effects	Boron (B)	Genotype (G)	B x G	
F-test	**	**	**	

Differences (by LSD $p < 0.05$) in the same row are indicated by different lowercase letters and in the same column by different uppercase letters. ** significant at $p < 0.01$

Table 2. Response in Grain Set Index (%) in barley and wheat grown in sand culture with and without added boron; and in the field with limed soil (BL), no added boron (B0) and boron treated soil (B+) (Experiment 3).

Species/	Sand culture (μM B)		Field soil treatment		
genotype	0	10	BL	B0	B+
Two row barley					
BRB 9604	66.9 aC	94.5 bC	86.7 aDE	93.4 aF	93.6 aBC
BRB 9	57.3 aC	94.4 bC	61.7 aC	69.1 aDE	83.0 bAB
BCMU 96-9	25.9 aB	76.1 bA	74.5 aCD	92.4 bF	87.5 abBC
Stirling	0 aA	92.2 bBC	37.7 aB	52.2 bC	89.6 cBC
Six row barley					
FNBL 8309	56.9 aC	79.0 bAB	80.7 aDE	83.4 aF	90.5 aBC
LARTC 9408	29.8 aB	86.7 bABC	69.0 aCD	79.4 abEF	91.1 bBC
BRB 2	8.7 aA	83.5 bBC	63.9 aCD	65.4 aCD	73.6 aA
Wheat					
Fang 60	93.3 aD	100 aC	98.2 aE	97.8 aF	98.6 aC
Flycatcher	92.7 aD	95.0 aC	72.0 aCD	92.5 bF	95.2 abBC
SW 41	60.8 aC	91.7 bBC	72.6 aCD	93.4 bF	95.4 bBC
Bonza	1.3 aA	97.4 bC	26.0 aB	31.3 aB	87.8 bBC
Tatiara	0 aA	75.3 bA	1.8 aA	13.6 aA	71.5 bA
	Effects	Boron (B)	Genotype (G)	B x G	
	F-test	**	**	**	

Differences (by LSD $p < 0.05$) in the same row are indicated by different lowercase letters and in the same column by different uppercase letters. ** significant at $p < 0.01$

Table 3. Response in the number of spikelets spike^{-1} in two-row barley and wheat to boron supply in sand culture (Experiment 1).

Species/genotype	Added B (μM)			
	0	0.1	0.3	5.0
Barley				
BRB 9	8.9 aA	9.9 aA	10.0 aA	9.2 aA
BCMU 96-9	20.2 aE	25.6 cE	24.0 bE	24.2 bE
CMBL 92029	13.0 aB	15.0 bCD	15.9 bD	17.6 cD
Wheat				
Fang 60	14.3 aC	14.2 aBC	14.6 aBC	14.4 aBC
SW 41	13.0 aB	13.7 aB	13.9 aB	14.0 aB
Tatiara	15.7 aD	15.8 aD	15.4 aCD	15.6 aC
Effects	Boron (B)	Genotype (G)	G x B	
F-test	**	**	**	

Differences (by LSD $p < 0.05$) in the same row are indicated by different lowercase letters and in the same column by different uppercase letters. ** significant at $p < 0.01$

Table 4. Relative boron responses (B0 as % of B10) for tillers, spikes, spikelets, Grain Set Index (GSI) and time of spike emergence in barley and wheat grown in sand culture (Experiment 3).

Species/ genotype	Tillers plant^{-1}	Spikes plant^{-1}	Spikelets spike^{-1}	GSI (%)	Days to spike emergence
Two-row barley					
BRB 9604	103.0 ab	84.7 b	45.1 b	70.8 c	112.0 b
BRB 9	204.0 cd	94.5 b	27.9 a	60.7 c	131.9 d
CMU 96-9	260.7 d	10.8 a	24.9 a	34.0 b	125.4 cd
Stirling	110.9 ab	11.6 a	35.2 ab	0.0 a	118.6 bc
Six-row barley					
FNBL 8309	119.7 ab	55.6 ab	76.9 c	72.0 c	125.4 cd
LARTC 9408	237.7 d	55.6 ab	45.6 b	34.4 b	95.0 a
BRB 2	186.7 cd	74.2 b	36.8 ab	10.4 a	118.8 bc
Wheat					
Fang 60	103.0 ab	98.0 b	95.0 d	93.3 d	100.6 a
Flycatcher	82.4 a	67.6 b	87.8 cd	97.6 d	100.0 a
SW 41	109.4 ab	98.0 b	88.1 cd	66.3 c	101.7 a
Bonza	95.8 ab	79.7 b	90.6 cd	1.3 a	99.0 a
Tatiara	151.9 bc	154.9 c	97.8 d	0.0 a	102.3 a
Analysis of variance					
Effects	F-test				
Boron (B)	**	**	**	**	**
Genotype (G)	**	**	**	**	**
B x G	**	**	**	**	**

Differences (by LSD $p < 0.05$) in the same column are indicated by different letters.

** Significant at $p < 0.01$

Table 5. Response of vegetative and reproductive growth in wheat (SW 41) and barley (BRB 9) to boron supply in sand culture (Experiment 2).

Plant response	Added B (μM B)				F-test
	0	10	0	10	Genotype x Boron
	Barley (BRB 9)		**Wheat (SW 41)**		
Shoot dry weight (g pot^{-1})	40.4 b	29.6 a	65.4 c	67.2 c	*
Tillers plant^{-1}	32.4 b	15.5 a	12.0 a	9.6 a	**
Day of spike emergence	49.1 b	45.8 a	56.9 c	57.3 c	*
Spikes plant^{-1}	18.8 c	13.1 b	7.2 a	6.4 a	*
Spikelets spike^{-1}	8.9 a	16.9 b	17.6 bc	19.0 c	*
Grains spike^{-1}	2.7 b	16.7 c	0.1 a	44.5 d	*
Grain yield (g pot^{-1})	5.2 a	67.2 b	1.3 a	82.6 c	*
Grain Set Index (%)	24.6 b	98.0 c	0.3 a	97.7 c	*

F-test of genotype by boron interaction, significant level: * $p < 0.05$, ** $p < 0.01$. Differences between B levels in each species are indicated by different lowercase letters (by LSD $p < 0.05$).

Table 6. Boron concentration (mg B kg^{-1}) in the flag leaf at booting of barley and wheat grown at four levels of boron in sand culture (Experiment 1).

Species/genotype	Added B (μM)			
	0	0.1	0.3	5.0
Barley				
BRB 9	13.2 aD	13.4 abD	14.6 bcE	15.6 cBC
BCMU 96-9	4.6 aA	5.4 abA	6.5 bB	8.7 cA
CMBL 92029	6.9 aB	7.0 aB	7.7 aB	7.7 aA
Wheat				
Fang 60	9.3 aC	10.9 bC	12.1 cD	16.2 dC
SW 41	7.5 aB	7.7 aB	9.3 bC	14.3 cB
Tatiara	3.8 aA	4.1 aA	4.5 aA	8.1 bA
Effects	Boron (B)	Genotype (G)	B x G	
F-test	**	**	**	

Differences (by LSD $p < 0.05$) in the same row are indicated by different lowercase letters and in the same column by different uppercase letters. ** significant at $p < 0.01$

Table 7. Boron concentration (mg B kg^{-1}) in the spike at booting of barley and wheat genotypes grown in sand culture at four levels of boron (Experiment 1).

Species/genotype	Added B (μM)			
	0	0.1	0.3	5.0
Barley				
BRB 9	6.6 aB	9.5 bC	12.3 cD	12.8 cE
BCMU 96-9	9.3 bD	8.5 abBC	8.2 aB	9.3 bC
CMBL 92029	4.1 aA	5.0 abA	5.7 bcA	6.1 cA
Wheat				
Fang 60	8.5 aCD	8.8 aBC	9.9 bC	10.4 bD
SW 41	7.8 aC	8.2 aB	10.9 bC	10.9 bD
Tatiara	4.9 aA	5.4 aA	6.4 bcA	7.3 bB
Effects	Boron (B)	Genotype (G)	B x G	
F-test	**	NS	**	

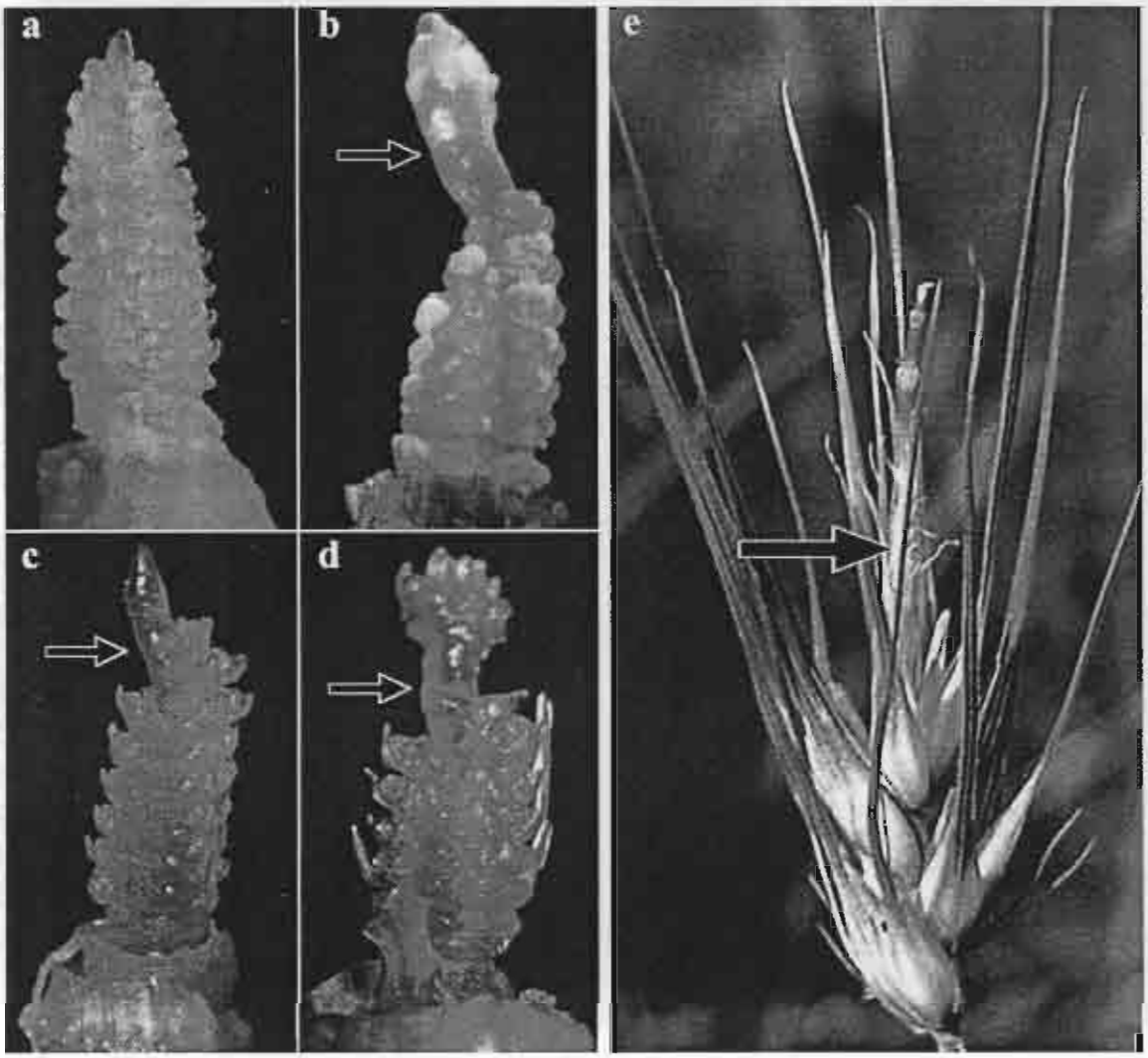
Differences (by LSD $p < 0.05$) in the same row are indicated by different lowercase letters and in the same column by different uppercase letters. ** significant at $p < 0.01$

Table 8. Boron concentration in the whole spike and flag leaf of the main stem at booting in wheat and barley grown with (B10) or without (B0) boron in sand culture (Experiment 2).

Species/genotype	Spike B (mg B kg^{-1})		Flag leaf B (mg B kg^{-1})	
	B0	B10	B0	B10
Barley, BRB 9	3.1	4.2	4.3	11.2
Wheat, SW 41	5.2	5.8	3.4	11.5
Effects	F-test			
Boron (B)	**		**	
Genotype (G)	**		NS	
B x G	NS		NS	

** significant at $p < 0.01$, NS = not significant, $p < 0.05$

Figure 1. The primordia of barley (cv. BRB 9) at lemma - awn primordium stage (Zadok et al., 1974) showing complete spikelet terminal in sufficient B supply (a) compared with abnormal primordia (b-d) in B deficiency (arrows showing arrested development of terminal spikelets). B oron deficient barley spike exhibiting the rat-tail symptom, in which terminal spikelets had degenerated in to wisps of white papery tissue (e).


a
b
c
d
e

Genotypic Variation in Boron Long Distance Transport into the Reproductive Organ of Wheat

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ABSTRACT

Genotypic variation in Boron (B) efficiency in wheat (*Triticum aestivum* L.) is expressed as large differences in grain set and pollen sterility under low soil B, but the mechanisms responsible for such differences are unknown. This paper aims to determine the mechanism for cultivar difference in B efficiency by comparing B-efficient (Fang 60) and B-inefficient (SW41) cultivars. Plants were grown with adequate ^{11}B (10 μM), until the premeiotic interphase stage in anther development, then transferred into ^{10}B at 0.1 or 10 μM. After five days, ending at the young microspore stage, all plants were transferred into adequate ^{11}B. Plants were sequentially harvested at 0, 1 and 5 days after transferring into ^{10}B, and at anthesis, anthers were collected and fresh pollen examined for viability. After 5 days in 0.1 μM B, during the critical stage of pollen development, pollen viability in SW41 was depressed by 47 %, but Fang 60 was not affected. Fang 60 maintained total B concentration in the ear at 6.8 mg kg^{-1} DW, in contrast to 3.8 mg kg^{-1} DW in SW 41. Insensitivity to B deficiency of Fang 60 was associated with its greater capacity to supply adequate ^{10}B to the ear directly from the root, but not from the previously taken up ^{11}B when external supply was restricted.

INTRODUCTION

There are numerous reports of genetic variation in B efficiency among wheat genotypes (Li et al, 1978; Tandon and Naqvi, 1992; Subedi et al, 1993; Rerkasem et al, 1993; Rerkasem and Jamjod, 1997) and other cereals such as barley (Rerkasem and Jamjod, 1989). Differences in B efficiency in wheat are generally associated with differences in the degree of pollen sterility or the ability to set grain in low B soil (Subedi et al, 1993; Rerkasem and Loneragan, 1994; Rerkasem and Jamjod, 1997). Pollen development in susceptible cultivars is impaired by B limitation resulting in pollen grains that are small and misshapen and do not accumulate starch (Rerkasem et al, 1989; Subedi et al, 1997; Anantawiroon et al, 1997). It has been suggested that the critical phase of anther development is surrounding pollen meiosis (Rawson, 1996), especially the period from premeiotic interphase through meiosis to late tetrad development (Huang et al, 2000; Dell and Huang, 2002). However, the mechanisms underlying cultivar differences in B efficiency in wheat remain unknown, but clearly they are associated with B supply to the ear during critical stages of microsporogenesis.

In broccoli (*Brassica oleracea* var. *italica* Plenck ~~cv. Commander~~), greater remobilization of B into the inflorescence was responsible for the avoidance of anatomical disorders in the inflorescence due to B deficiency in some cultivars (Shelp et al, 1992). Similarly, Stangoulis (2001) recently reported that greater B efficiency in ~~the~~ oilseed rape (*Brassica napus* L.) cv.~~ultivar~~ Huashuang) was associated with retranslocation of ^{10}B from old leaves whereas B-inefficient cultivars lacked this capacity. However, both of these studies were carried out with *Brassica* species which exhibit a degree of phloem mobility of B in contrast to many other species

which do not (Brown and Shelp, 1997). Huang et al (2001) showed that some ^{10}B, incorporated into vegetative plant parts following absorption from the external solution, was capable of retranslocation into the ear of wheat. However, the authors concluded that the amount of ^{10}B retranslocated was insignificant relative to the B demand of the ear. Since only a B-inefficient cultivar, Wilgoyne, was used in this study (Rerkasem unpublished), the possibility remains that the capacity to retranslocate B into the ear may differ across cultivars, and that this difference may allow the~~the~~ mechanism of B efficiency in wheat to be identified. The avoidance of male infertility in B-efficient cultivars may involve the ability to supply B adequately into the nontranspiring anthers or the developing ear. This experiment set out to examine B responses of efficient and inefficient wheat genotypes during the most critical stage in pollen formation and to compare the routes of B mobilization into the ear.

RESULTS AND DISCUSSION

Huang et al. (2000) suggested that the period of pollen development from the premeiotic interphase to the late tetrad was the main phase sensitive to B deficiency. They also postulated that B deficiency during meiosis might impair the formation of pollen cell walls and cell expansion, leading to a reduction in pollen viability in wheat. Pollen viability in B-inefficient SW 41 was nearly halved when the external B supply was interrupted during this critical stage (Figure 1). The same treatment had no effect on pollen viability of Fang 60. This is consistent with known differences in the sensitivity to B deficiency of the genotypes as represented by their male fertility and grain set in low B (Anantawiroon et al, 1997; Subedi et al, 1999).

The differential response to short-term B deficiency in SW 41 and Fang 60 was associated with significantly lower concentration of B in the ear of SW 41 (Table 2).

Thus, the B-efficient and inefficient genotypes in this study are distinguishable by the B concentration in their ears. This difference was not detected by others who have reported genotypic variation in responses to low B for male fertility and grain set (Rerkasem and Lordkaew, 1996) including a comparison between SW 41 and Fang 60 (Subedi et al., 1999). In these previous studies, determination of B in the ear was not done until booting. By this time, the B deficient wheat ear may have continued to accumulate B in the palea, lemma and other secondary sexual parts of the ear although damage to the pollen had already occurred. The present results suggest that it may be feasible to distinguish between the B-efficient and inefficient genotypes by early B analysis of the ear. However, timing of the analysis will be critical. Sampling for the ear has to be done immediately upon the completion of the critical meiotic stage of pollen development.

Boron concentrations in upper canopy leaves of Fang 60 were also greater than those in SW 41 after 5 days of low B supply (Table 2). This suggests that cultivar difference in sensitivity to B deficiency may be associated with the pattern of B distribution within the plant when B was limited. The greater ability of Fang 60 to distribute B into the developing ear may contribute to its tolerance to low external B. The reproductive ~~plant~~ parts of wheat, the anthers and carpels, were found to require B at greater concentration for their normal development than leaves (Rerkasem et al, 1997; Huang et al, 2000). The greater amount of B distributed into the apical regions of the plant may increase the opportunity for reproductive success.

A suggestion has been made by some that previously accumulated B might be effectively remobilized to supply the ear of B-efficient genotypes, such Fang 60, when external supply becomes limited (Rawson, 1996; Subedi et al, 1999). We found this to be unsupportable by three sets of results, which instead suggested that the

primary mechanism for B efficiency in Fang 60 to be associated with its long distance transport directly from the root. Firstly, accumulation of ^{11}B in the ear of both SW 41 and Fang 60 seemed to have stopped after ^{11}B in the nutrient solution was replaced by ^{10}B. The content of ^{11}B in the ear of either SW 41 or Fang 60 did not increase after the ^{10}B treatments were imposed (Table 3). Data on the content of ^{10}B in the ear provided the second piece of supporting evidence that the mechanism for B efficiency is related to direct supply from the root and not recycled B from some plant parts. In the low B treatment, the ^{10}B content in the ear of B-efficient Fang 60 was three times that of the B-inefficient SW 41 (Table 4). Examination of the ^{10}B:^{11}B ratio provides the third piece of evidence. The ratio of ^{10}B:^{11}B increased significantly with time during the 5 days that the B^{11} supply was replaced by ^{10}B (Table 5) but at different rates in the two genotypes. The increase in the ratio of B^{10}:B^{11} with time after the transfer from ^{11}B to ^{10}B was much stronger in Fang 60 than SW 41, especially with lower external B. During the 5 days in which external B was lowered to 0.1 μM, the ^{10}B:^{11}B ratio in Fang 60 was increased from 0.17 to 1.15 and less so in SW 41, from 0.11 to 0.38.

In conclusion, pollen viability of the B-efficient Fang 60 was not affected by withholding B during the critical stage of microsporogenesis while pollen viability in inefficient SW 41 was nearly halved. The genotypic difference in B efficiency is related to the greater ability of Fang 60 to accumulate and distribute B into the developing ear than SW41. By using ^{11}B and ^{10}B, we were able to demonstrate that net B movement into the ear, when external supply was restricted, did not come from the ^{11}B previously taken up by the plant. The greater amount of ^{10}B accumulated by Fang 60 in low B further confirmed that the primary mechanism for B efficiency in

Fang 60 is its greater capacity to supply adequate B to the ear directly from the root, enabling Fang 60 to avoid pollen sterility caused by B deficiency.

MATERIALS AND METHODS

Plant material and culture

This experiment selected two spring wheat cultivars from the efficient (cv. Fang 60) and inefficient (cv. SW 41) classes of B efficiency determined by Rerkasem and Jamjod (1997). Seeds were imbibed in aerated 2 mM $CaSO_4$ solution for 24 hours and germinated on paper towels moistened with 2 mM $CaSO_4$ for 48 hours in the dark at 25 °C. Seeds of SW41 were germinated 1 day before seeds of Fang 60 in order to synchronize the stage of ear development at microsporogenesis. Seedlings were transferred into trays containing 8 L of 1/3 strength nutrient solution (Huang et al, 1996) with a concentration of 0.1 μM ^{11}B and 5 mM 2-[N-Morpholino]ethanesulfonic acid. The pH was adjusted daily to 6.0 ± 0.2 with 1 M KOH or 10 % H_2SO_4. Four days after germination, the plants were grown in a 5 L full-strength basal nutrient solution that contained 10 μM ^{11}B and aliquots of all nutrients were added to each pot during the experiment by the programmed nutrient addition as described in Huang et al. (1996). Tillers were restricted to a maximum of 4 tillers plant^{-1}, by removing extra tillers as they emerged from the 5th onward. The stage of growth was determined by dissection of spare plants at the 6, 7 and 8 leaf stages and the stage of pollen development was determined on extra B–adequate plants ~~with~~ using DAPI (4'-6-Diamidino-2-phenylindole 2HCl, Sigma Lot 104F-0542) ~~test~~ fluorescence (Vergne, Delvallee and Dumas,is this style of 3 authors OK? 1987). Ten μM ^{11}B was supplied continuously up to ~~the~~ late premeiotic interphase /early meiosis in the main stem (Bennett et al, 1973). The plants were then treated with either 0.1 or 10 μM of 99.43 % ^{10}B-enriched boric acid continuously up to the

late tetrad, after which the plants were returned to solutions with 10 μM ^{11}B up to anthesis (Table 1). ^{10}B was used as a tracer for B distribution to the ear during critical stage of microsporogenesis, while ^{11}B was used as a tracer for B remobilization to the ear.

The pH was adjusted to 6.0 -± 0.2 with 1 M KOH or 10 % H_2SO_4. The pots were randomly distributed in temperature-controlled water baths (18-22 °C) and repositioned daily within the baths and shifted between baths every 3 days. The growth conditions in the glasshouse were: mean air temperature 27.5 °C (range:20-35); mean PAR (photosynthetic active radiation) 1165 μmol m^{-2} s^{-1} (range: 960-1900) . Nutrient solutions wereas aerated with filtered air and the dry weight increments of extra plants were used to calculate the amount of nutrients required to maintain nutrient supply by programmed nutrient addition (Asher and Blamey, 1987). Analytical grade chemicals were used to make up the nutrient solutions and water, purified by passing through a column packed with B-specific resin (IRA-743, Sigma Chemical Co.), was used for making up all nutrient solutions. Between the treatments, the roots were washed with DI water, then rinsed three times in 5 mM $CaSO_4$ solution, in order to remove unbound B from root free space.

Sampling and plant analysis

The first harvest was taken before the ^{10}B treatment at the beginning of the critical stage of pollen development in the main stem (Huang et al, 2000) (Table 1). The second and third harvests were taken 1 day and 5 days (late tetrad stage) after the ^{10}B treatment began. At each harvest, three pots (replicates) of plants (two plants each) were taken from each cultivar. The plants were subdivided into flag leaf, ear, penultimate leaf, stem between flag leaf and penultimate leaf of the main stem. All samples were analysed for ^{11}B and ^{10}B- content. The fourth harvest was taken at

anthesis. From three replicate pots (~~replicates~~two plants per pots) ~~of plant (two plants each) from~~ of each cultivar, pollen was taken from the central 4 spikelets of the main shoot ear. Pollen at anthesis was tested for viability with the fluorochromatic (FCR) method (Heslop-Harrison and Shivanna, 1984). The plant samples were oven-dried (70 °C). Dry samples were ground in a stainless steel mill and dry-ashed in 1 % nitric acid as described in Huang et al, (2001) and B concentration was determined using~~by~~ an inductively coupled plasma automatic emission spectrometer (ICP-AES) (Zarcinas et al, 1987).

Statistical analysis

Data were analyzed statistically by analysis of variance. Significantly different means were separated at the 0.05 probability level.

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LITERATURE CITED

Anantawiroon P, Subedi KD and Rerkasem B (1997) Screening wheat for boron deficiency . In Boron in Soil and Plant. Eds RW Bell and B Rerkasem, Kluwer Academic Publishers Dordrecht, New York, 101-104; ?

Asher CJ and Blamey FPC (1987) Experimental control of plant nutrient status using programmed nutrient addition. J. Plant Nutr 10: 1371-1380? Check others

Bennet MD, Rao MK, Smith JB and Bayliss MW (1973) Cell development in the anther, the ovule and the young seed of *Triticum aestivum* L. var. Chinese Spring. Phil. Trans. Royal Soc. London Ser. B 266: 39-81.

Brown PH,?and Shelp BJ (1997) Boron mobility in plants. Plant and Soil 193: 85-101

Cheng C and Rerkasem B (1993) Effect of boron in the pollen viability of wheat. Plant Soil 155/156: 313-315

Dell B, Huang L and Bell RW (2002) Boron in Plant Reproduction. In HE Goldbach, B Rerkasem, MA Wimmer, PH Brown, M Thellier and RW Bell eds, Boron in Plant and Animal Nutrition, Kluwer Academic/Plenum, New York, 103-117

Heslop-Harrison J, Heslop-Harrison Y, Shivahna KR (1984) The evaluation of pollen quality, and a further appraisal of the fluorochromatic (FCR) test procedure. Theor Appl Genet 67: 367-375.

Huang L, Plant J, Bell RW, Dell B and Deane K (1996) Effects of boron deficiency and low temperature on wheat sterility. In: Rawson HM and Subedi KD eds. Sterility in Wheat I Sub-Tropical Asia: Extent, Causes and Solutions. ACIAR Proceedings No. 72, ACIR, Camberra, 90-101

Huang L, Plant J, Dell B, and Bell WR (2000) Effects of boron deficiency on anther development and floret fertility in wheat (Triticum aestivum L. Wilgoyne). Ann. Bot. 85: 493-500

Huang L, Plant J, Dell B, and Bell WR (2001) Boron supply into wheat (*Triticum aestivum* L. cv. Wilgoyne) ear whilst still enclosed within leaf sheaths. J. Exp. Botany 52: 1731-1738

Li BH, Li WH, Kui MC, Chaows, Jern HP, Li CR, Chu WJ and Wang CL (1978) Studies on cause of sterility of wheat. J. Northeast Agric. College 3: 1-19 (In Chinese, with English translation)

Rawson HM and Hofstra G (1969) Translocation of remobilization of 14C assimilated at different stages by each leaf of the wheat plant. Aust. J. Biol. Sci 22: 321-331

Rawson HM.? (1996) The developmental stage during which boron limitation causes sterility in wheat genotypes and the recovery of ferility. Aust. J. Plant Physiole 23: 709-717

Rerkasem, B and Jamjod, S (1989) Correcting boron deficiency induced ear sterility in wheat and barley. Thai J. Soils Fert 11: 200-209 (in Thai with English summary)

Rerkasem B and Loneragan J F (1994) Boron deficiency in two wheat genotypes in a warm, subtropical region. Agron. J. 86: 887-890

Rerkasem B and Lordkaew S (1996) Tissue Boron. In Sterility in Wheat in Sub-Tropical Asia Extent, Causes and Solutions. Eds HM Rawson and KD Subedi, ACIAR Proc. No. 72, 36-38.

Rerkasem B, Lordkaew S, Dell B (1997) Boron requirement for reproductive development in wheat. In: T Ando, K Fujita, T Mae, H Matsumoto, S Mori, J Sekiya, eds. Plant nutrition for sustainable food production and environment. Dordrencht Kluwer Academic Publishers, New York, 69-73

Rerkasem B, Netsangtip R, Lordkaew S and Cheng C (1993) Grain set failure in boron deficient wheat. Plant Soil 155/156: 309-312

Rerkasem B and Jamjod S (1997) Genetypic variation in plant response to low boron and implications for plant breeding. Plant Soil. 193: 169-180

Bens refs are not in the correct order

Rerkasem B, Saunders DA and Dell B (1989) Grain set failure and boron deficiency in wheat in Thailand. J. Agric. (CMU) 5:1-10 Font?

Shelp BJ, Penner R and Zhu Z (1992) Broccoli (*Brassica oleracea* var. *italica*) cultivar response to boron deficiency. Can. J. Plant Sci. 72: 883-888

Stangoulis JCR, Brown PH, Bellaloui N, Reid RJ and Graham RD (2001) The

efficiency of boron utilisation in canola. Aust. J. Plant Physiol. 28:1109-1114

Subedi KD, Budhathoki CB, Subedi M and Tuladhar JK (1993) Survey and research report on wheat sterility problem (1992/93). LARC working Paper No. 93/49. Nepal, Lumle Agricultural Research Centre.

Subedi KD, Budhathoki CB, Subedi M and GC Yubak D (1997) Response of wheat genotypes to sowing date and boron fertilization aimed at controlling sterility in a rice-wheat rotation in Nepal. Plant Soil 188: 249-256

Subedi KD, Gregory PJ and Gooding MJ (1999) Boron accumulation and partitioning in wheat cultivars with contrasting tolerance to boron deficiency. Plant Soil 214: 141-152

Tandon JP and Naqvi SMA (1992) Wheat varietal screening for boron deficiency in india. In: CE Mann and B Rerkasem, eds. Boron Deficiency in wheat, Wheat Special Report No.11. Mexico DF, CIMMYT, 76-78

Vergne P, Delvallee I, Dmas C (1987) Rapid assessment of microspore and pollen development stage in wheat and maize using DAPI and membrane permeabilization. Stain Technology 62: 299-304

Zarcinas BA, Cartwright B, Spouncer LR (1987) Nitric acid digestion and multielement analysis of plant material by inductively coupled plasma spectrometry. Comm. Soil Sci. Plant Anal. 18: 131-146

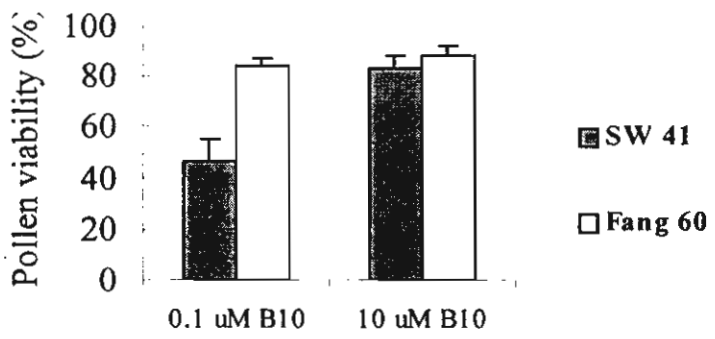

Figure 1. Effect of short term B deficiency during microsporogenesis (days 0-5: see Table 1) on pollen viability in two

Table 1. *Boron treatments, harvest times and stage of pollen development at each harvest of the main stem.*

Days*	Harvest	B treatment	Pollen development stage during treatment
		10 μM ^{11}B	Seedling to white anther (premeiotic stage)
0	1	10 μM ^{11}B	Late premeiotic stage/ early meiosis
1	2	0.1, 10 μM ^{10}B	After late premeiotic stage by 24 hr. still unclear
5	3	0.1, 10 μM ^{10}B	Late tetrad to young microspore stage
	4	10 μM ^{11}B	Anthesis

*= duration of low and adequate ^{10}B treatment.

Table II. *Total B concentration in plant parts (mg kg^{-1} DW) after 5 days of varied B supply treatment. Values are means of three replicates ± SE.*

Plant part	B treatment	Genotype	
	(μM ^{10}B)	Fang 60	SW 41
Ear	0.1	6.8 ± 0.7	3.8 ± 0.3
	10	12 ± 1.4	7.8 ± 0.5
Flag leaf	0.1	13 ± 2.3	9.3 ± 0.6
	10	21 ± 1.4	20 ± 0.5

Penultimate leaf	0.1	17 ± 0.4	13 ± 0.7
	10	19 ± 0.5	20 ± 0.8
Stem segment between flag	0.1	4.0 ± 0.4	4.7 ± 0.3
~~leaf~~ and penultimate	10	6.2 ± 0.9	5.9 ± 0.6
leaves~~f~~			

Table III. *The content of ^{11}B in the ear ($\mu g\ ear^{-1}$) at day 0 (D0), day 1 (D1) and day 5 (D5) of B treatment.*

B^{10} treatment	Fang 60			SW 41		
($\mu M\ B^{10}$)	D0	D1	D5	D0	D1	D5
0.1	0.117	0.098	0.108	0.130	0.100	0.104
10	0.117	0.079	0.070	0.130	0.086	0.140

$LSD_{(0.05)} = 0.038$

Table IV. *The content of ^{10}B in the ear ($\mu g\ ear^{-1}$) at day 1 (D1) and day 5 (D5) of B treatment.*

B treatment	Fang 60		SW 41	
($\mu M\ ^{10}B$)	D1	D5	D1	D5
0.1	0.016	0.122	0.011	0.039
10	0.039	0.170	0.043	0.193

$LSD_{(0.05)} = 0.028$

Table V. *The ratio of ^{10}B : ^{11}B in ear at day 1 (D1) and day 5 (D5) of B treatment. Values are means of three replicates.*

B treatment	Fang 60		SW 41	
(μM B^{10})	D1	D5	D1	D5
0.1	0.17	1.15	0.11	0.38
10	0.49	2.62	0.50	1.38

$LSD_{(0.05)} = 0.021$

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Fallow enrichment with pada (*Macaranga denticulata* (Bl.) Muell. Arg.) trees in rotational shifting cultivation in northern Thailand

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Key words: Improved fallow, *Macaranga denticulata* (Bl.) Muell. Arg.), Slash and burn, Upland rice,

Abstract

Shifting cultivators in Thailand widely attribute the maintainance of crop productivity to pada (*Macaranga denticulata* (Bl.) Muell. Arg.), rotation cycles having become much shorter than the customary 10–20 years. This paper examines the use of pada in a 7-year rotation on an acid soil with low available soil P (2–4 mg kg^{-1} by Bray II). Dense pada patches in 7-year-old fallow averaged 43 tons ha^{-1} of above ground biomass, 20% more than sparse patches. The biomass in dense pada contained disproportionately more P, K, Ca and Mg (34%, 92%, 80% and 107% more, respectively) than in sparse pada patches. Slashing and burning 7-year-old fallow with dense pada produced a subsequent rice yield that was three time that with sparse pada. Rice grown after dense pada had been slashed and burned after three years yielded less than one third of that after a full 7-year rotation. It is, as yet, unclear how rice yield in dense pada patches is enhanced in the full 7-year rotation. Nutrient concentrations in the mature rice were generally either the same or higher in the sparse than dense pada patches. In dense pada patches rice accumulated twice to four times as much nutrients as in sparse pada patches, and a much larger fraction of the nutrients was stored in the fallow. Uptake of nutrients in the sparse pada patches may have been limited by some factor that either governs availability of the nutrients released by burning or depressing rice growth and so its nutrient demand.

Introduction

Rotational shifting cultivation has been shown to be a productive and sustainable form of land use in mountainous areas where land is sufficiently plentiful to allow a fallow period of 10–20 years (Kunstadter 1978). Population pressure combined with an increasing demand for conservation (from watersheds, erosion control, carbon sequestration to biodiversity) by society at large, however, have made certain that this luxury of very long fallows is now no longer an option for most shifting cultivators in Southeast Asia (Rerkasem and Rerkasem 1995) and elsewhere. Considerable interest, therefore, has arisen in approaches that might maintain crop productivity with shorter fallow. Numerous efforts, especially in Africa, have gone into identification of trees and other plants as fallow–enriching species (e.g., Tarawali (1991) and Kwesiga and Coe (1994), Mafongoya and Nair (1997), Kaya and Nair (2001)). This paper reports on the use of a local tree called *pada* for fallow improvement that has helped to maintain upland rice yield at reasonable levels of 2 to 4 Mg ha^{-1} on a seven years rotation.

Pada (*Macaranga denticulata* (Bl.) Muell. Arg.) is well known for its fallow-enriching property amongst the various ethnic groups who make a living on rotational shifting cultivation in northern Thailand. It is known as *Teen Tao* amongst the *Khamu* and *H'tin* who populate the northeastern mountains, on the border with Laos. *Pada* is the name in *Skaw Karen* (Thailand's largest minority group, now concentrated along

the western border with Myanmar), while the *Pwo Karen* call it *Letha*. The *Lua* (who are believed to have been the dominant group in the region until about a thousand years ago) call it *Tong Coab*. The *Akha* (a group not known to practice rotational shifting cultivation in Thailand) call the tree *Loom Piah*. Amongst lowland *Thai* it is variously called *Tong Taeb, Tong Tao, Tao Maew, Por Khee Haed* or *Bai Hoo Chang*. Pada is a small evergreen tree of the Euphorbiaceae family, that can reach 19 m in height and up to 40 cm in diameter at breast height. *Macaranga* is a relatively large genus of pioneer species (Whitmore 1982). Some 80 species have been identified in Africa and 200 in the Eastern Tropics, although not all are pioneer species. *M. gigantea* and *M. kurzii* also occur in the study village of Huai Tee Cha, but according to farmers pada is the only species with a fallow-improving property. The presence of pada in the fallow is believed to be responsible for maintaining productivity of upland rice in the shorter rotation. This study set out to measure the effect of pada on nutrient accumulation and upland rice yield in farmers' fields in a village where rotational shifting cultivation is still the dominant cropping system.

Materials and methods

The study was conducted in the rotational shifting cultivation fields of the village of Huai Tee Cha (19°78′ N, 93°84′ E), Sob Moei District, Mae Hong Son Province, about 250 km southwest of Chiang Mai. The soil is reddish clay loam. Before commencing plant and soil sampling, we took 30 days to become acquainted with the system by interviewing farmers and extensive field walks. The communally managed shifting cultivation area was divided into fields that are cropped in different years, designated by the year in which rice was last grown (Figure 1). According to the villagers this rotation has taken place for about 200 years, ever since Tee Cha was settled from a neighbouring village. A preliminary study of vegetation composition of the fallow forests was carried out in February and March (the village's normal slashing and burning season) of 2000. Trees were recognized as pada or 'others'. From areas with high and low density pada (defined in collaboration with farmers and designated dense and sparse pada), density of pada and other species was determined in three replicates of 10 × 10 m quadrats. Dominant 'other' species were *Microcos peniculata, Lithocarpus sp., Phoebe lanceolata* and *Glochidion sphaerogynum* (Further information about vegetation composition as well as tree density, biomass and nutrient contents of fallow areas with different ages will be reported elsewhere).

Mature fallow before slashing and burning

As field 1994/2000 was to be slashed and burned it was sampled for biomass and nutrient contents. Dry weight (sub-samples dried to constant weight at 80 °C) of the above ground live biomass, divided into pada and others, and litter were determined. The samples, whole plants in case of pada and others, were analyzed for N, P, K, Ca and Mg. Before burning soil samples were taken from the same area at 0–30 and 30–60 cm for determination of pH (water, 1:1), organic matter content (Walkley-Black), available P (Bray II, Wanatabe and Olsen (1962)), K, Ca and Mg (1 N NH_4OAc pH7).

The upland rice crop

A detailed study of the upland rice was carried out on the crop belonging to one farmer, Nopporn. At 30 days from sowing, samples (whole tops) of ten rice plants each were taken from the sparse and dense pada area for determination of N, P, K, Ca and Mg. At the same time soil samples were taken for determination of fertility characteristics at 0–30 and 30–60 cm depth. At maturity, samples of the rice crop were taken in 2 × 5 m quadrats from the sparse and dense pada areas. In addition, 1 × 1 m samples of rice at maturity were also taken from sparse and dense pada areas from five other farmers in the same village. The rice samples were threshed in the field and separated into grain and straw. Grain yield was determined after 3 days of sun drying (to water content of about 12%), straw yield was measured after drying for 48 hours at 80 °C. Sub-samples from Nopporn's field were also evaluated for the yield components, i.e. number of hills m^{-2}, plants $hill^{-1}$, tillers $hill^{-1}$, panicles $plant^{-1}$ and 1,000 seed weight, and analyzed for N, P, K, Ca and Mg in the grain and straw.

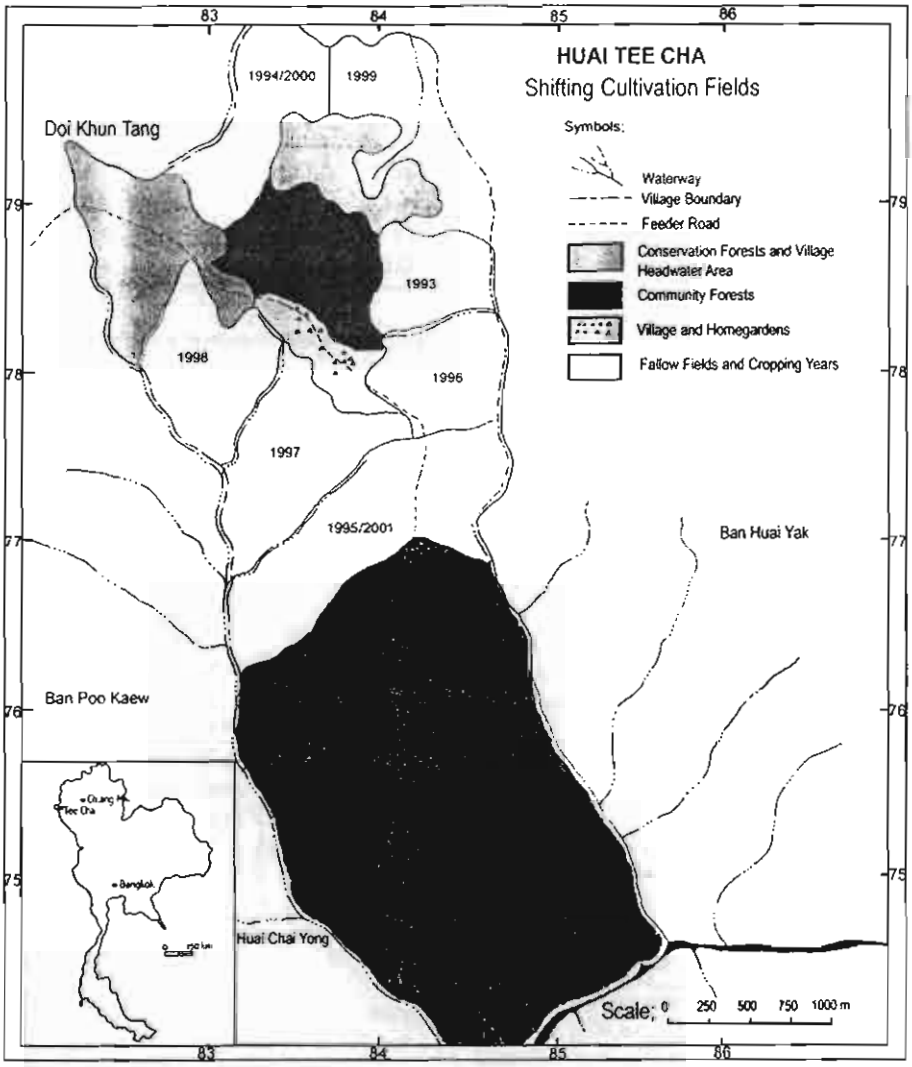

Figure 1. Rotational shifting cultivation fields of Huai Tee Cha village in northern Thailand indicating year of cropping.

Results and discussion

Fallow biomass and nutrient content

After seven years of regrowth (the rice cropping year is considered year 1 of regrowth, as fallow regeneration begins with upland rice emergence) above ground biomass averaged 43 Mg ha^{-1} in dense pada patches, about 20% more than that in sparse patches (Table 1). Pada contributed more than half of the biomass in the dense patches, and only 39% in sparse patches. There was also considerably more litter in dense pada patches. The above ground biomass in dense pada patches contained 536 kg N, 38 kg P, 253 kg K, 132 kg Ca and 46 kg Mg ha^{-1}. This was 10% more N, 34% more P, 92% more K, 80% more Ca and 107% more Mg than that in the sparse pada patches (Table 2). In the mature fallow of 8 years rotation in the village of Pa Pae (18°15′ N 98°3′ E) in the same vicinity where no pada was found (Sabhasri 1978) 143 kg N, 16 kg P and 176 kg K ha^{-1} were reported to have accumulated in the 63 t of above ground biomass (Zinke et al. 1978). The slight difference in total biomass N between sparse and dense pada patches in this study is accounted for by other species, which in sparse pada contributed as much N as pada in the dense pada patches. Pada was clearly crucial to the accumulation of extra P, K, Ca and Mg, though not so much for N. The higher amount of nutrients accumulated in dense pada came largely from

Table 1. Total aboveground biomass in the fallow vegetation after seven years of regeneration of a rotational shifting cultivation system in northern Thailand.

	Sparse pada	Dense pada	Significant difference by t-test
	Mg ha^{-1}		
Pada	9.4	22.2	$p < 0.01$
Other species	21.6	14.2	$p < 0.01$
Litter	4.7	6.4	$p < 0.01$
Total	35.7	42.8	$p < 0.05$

Table 2. Above ground nutrient contents in the fallow vegetation after seven years of regeneration of a rotational shifting cultivation system in northern Thailand.

Vegetation type	Nutrient element	Dense pada	Sparse pada	Significant difference
		Nutrient content (kg ha^{-1})		
Pada	N	289	131	$p < 0.01$
	P	21	6	$p < 0.01$
	K	97	32	$p < 0.01$
	Ca	72	21	$p < 0.01$
	Mg	10	6	$p < 0.05$
Other	N	191	297	$p < 0.01$
	P	12	18	$p < 0.05$
	K	118	85	$p < 0.01$
	Ca	28	36	NS
	Mg	23	6	$p < 0.01$
Litter	N	56	59	NS
	P	5	4	NS
	K	38	14	$p < 0.01$
	Ca	32	16	$p < 0.05$
	Mg	13	10	NS
Total	N	536	488	$p < 0.01$
	P	39	29	$p < 0.05$
	K	253	132	$p < 0.01$
	Ca	132	73	$p < 0.01$
	Mg	46	22	$p < 0.01$

pada in the case of P, pada and litter in the case of Ca, and from other species as well as pada and litter in the case of K and Mg. Pada roots were found to be associated with a diversity (genera and species) and abundance of arbuscular mycorrhizal fungi, both in the number of spores found in the rhizosphere and in root colonization (Youpensook et al. (in press)).

Soil fertility characteristics before slashing and burning and under rice

The study site was on a very acid soil, with pH 4.3 (1:1 water) before burning (Table 3). As others have previously shown (e.g., Nye and Greenland (1964) and Sanchez (1976)), burning clearly had a strong liming effect. Thirty days after rice sowing, pH of the surface soil measured 4.8, and slightly less deeper in the soil profile. Soil organic matter content declined slightly from around 4% before burning to 3.6% by the time rice was 30 days old. The first rainfalls of the season that germinated the rice may have contributed to this decline by stimulating microbial activity. Available soil P was generally very low at 2–4 mg kg^{-1}. There were significant but very small effects of pada density and burning on available P. The highest P value (Bray II) was 4.3 mg kg^{-1}, in dense pada patches after burning, which is very low indeed compared with about 12 mg kg^{-1} considered to be suffi-

Table 3. Fertility characteristics of the soil of the study area, in a rotational shifting cultivation system in northern Thailand, before burning and 30 days after rice sowing.

Pada density	Time	pH (1:1 water)	Organic matter (%)	P	K	Ca	Mg
				(mg kg^{-1})		(meq 100 g^{-1})	
Depth, 0–30 cm							
Sparse	Before[a]	4.30	3.96	2.23	133	0.53	0.27
	After[b]	4.79	3.60	3.34	181	0.64	0.48
Dense	Before	4.26	4.19	2.62	148	0.78	0.65
	After	4.83	3.84	3.70	198	0.75	0.67
Depth, 30–60 cm							
Sparse	Before	4.29	3.87	1.53	107	0.38	0.13
	After	4.41	3.43	3.00	156	0.47	0.49
Dense	Before	4.24	3.97	2.02	110	0.56	0.37
	After	4.58	3.73	4.29	178	0.59	0.50
Significant effects by analysis of variance							
Effects	Pada	NS	NS	*	NS	*	*
	Time	*	*	*	*	NS	NS
	Depth	NS	NS	NS	*	*	*
PxT, TxD, PxT, PxTxD		NS	NS	NS	NS	NS	NS

a) Before burning; b) After burning and 30 days from rice sowing

cient (Sanchez 1976). There was no significant difference in soil pH, organic matter and extractable K content between dense and sparse pada patches (Table 3). On the other hand, dense pada was associated with 47% more extractable Ca and twice to three times as much extractable Mg than in the sparse pada patches.

Pada establishment

As upland rice germinated after the first rains, pada emerged in thick pink carpets among the rice. Farmers do not treat pada as weeds, they are not routinely removed when the rice is weeded by hand. Thinning may be done where the density is considered too high and some attempts are sometimes made to transplant seedlings to low density areas. At rice maturity, about six months after sowing, and when pada had reached almost 1.5 m height, the number of pada averaged about 7 plants m^{-2} in dense patches, twice that in sparse patches (Table 4). In the fields cropped in different years, there was a trend of pada density decrease with fallow age (data not shown). In the 1995 field (slashed and burned in 2001), dense pada patches averaged 0.4 pada-tree m^{-2}, and 0.1 pada-tree m^{-2} in sparse patches. Since these are a fraction of the numbers of pada seedlings in the first year, the problem of low pada density must have been associated with survival of the seedlings rather than recruitment.

Table 4. Density of trees, pada and other species, after 6 months and 7 years in a rotational shifting cultivation system in northern Thailand.

Species	Pada density	Trees/m^2	
		6 months[a]	7 years[b]
Pada	Sparse	3.27 ± 0.32	0.10 ± 0.01
	Dense	6.60 ± 0.12	0.42 ± 0.02
Others†	Sparse	ND	0.12 ± 0.01
	Dense	ND	0.21 ± 0.01

a) At rice harvest ND = not determined, very few other plants at this time; b) Before slashing and burning for the next crop, the rice year is year 1 of regrowth; † Major species included *Microcos peniculata, Lithocarpus sp., Phoebe lanceolata* and *Glochidion sphaerogynum*. Values are mean ± standard errors of three replicates.

Upland rice nutrition and yield

Rice plants at one month in dense pada contained N and K at significantly higher concentrations than those in sparse pada patches (Table 5). No significant difference was found in the concentration of P, Ca and Mg. Based on published data (Reuter et al. 1997), the rice crop at 30 days was deficient in N and K in sparse pada area, and deficient in P in both sparse and dense pada area.

Table 5. Nutrient concentration in the upland rice (whole tops) at 30 days from sowing, in areas following pada at low and high densities in a rotational shifting cultivation system in northern Thailand.

Concentration (%)	Sparse pada	Dense pada
N	2.76[a]	4.06[b]
P	0.29	0.32
K	3.60[a]	4.99[b]
Ca	0.19	0.26
Mg	0.22	0.24

a) For same nutrient element, different letters designate significant difference by LSD ($p < 0.05$).

In 7-year rotation, rice grain yield in the dense pada area determined from detailed measurements in a field belonging to one farmer, averaged 2.57 Mg ha^{-1}, three times that in the sparse pada area (Table 6). Similar effects of pada density on the rice grain and straw yield were observed in 7-year rotation fields belonging to five other farmers (Table 7). The higher yield in dense pada patches was associated largely with a higher number of panicles and percentage of fertile tillers, and to a less extent higher plant density and number of tillers $hill^{-1}$ (Table 8). The effect was clearly cumulative, requiring more than three years of fallow.

How did high pada density increase the rice yield? Rice grown in dense pada patches that were slashed and burned after three years of fallow yielded only 0.74 Mg ha^{-1} of grain (Table 6). Clearly, the higher rice yield was not due to some pre-existing condition in the dense pada patches. As discussed above, the growth of rice at 30 days in sparse pada patches may have been limited by N and K deficiency, and more severely by P deficiency. By maturity, the concentration of N and Mg in the rice grain and P in both grain and straw were higher in the sparse than in the dense pada area (Table 9). The concentration of straw N and Mg and grain K were not different between rice in sparse and dense pada area. Straw K was the only case of nutrient concentration in rice in dense pada exceeding that in the sparse area. Critical K deficiency concentrations have been reported at 0.4% in the grain and 1% in the straw (Reuter et al. 1997). Compared with these, K deficiency is not indicated by the the grain K concentration at 0.45% and straw K at 1.91% in the rice in the sparse pada patches.

At maturity, the rice crop accumulated twice as much N, P, Ca and Mg and four times K in the dense as in the sparse pada patches (Table 9). However, the amount of N, P and K taken up by the rice crop accounted for a much smaller fraction of the nutrients accumulated above ground in 7-year old-fallow in the sparse (4%, 15% and 16%, for N, P and K, respectively) than in the dense pada patches (9%, 25% and 30%, respectively). The rice Ca and Mg uptake relative to the nutrient in fallow biomass were somewhat closer between sparse (13% for Ca; 5% for Mg) and dense (15% and 6%, respectively) pada patches. Although much of the above ground N in the fallow would have been lost with burning, most of the P, K, Ca and Mg could be assumed to remain in the ash. Much more nutrients would have been present in the root zone of rice than was taken up by the rice crop. The much smaller fraction of fallow accumulated P and K taken up by rice in sparse compared with dense pada patches suggested that the uptake of these nutrients was limited by some factor(s). Such a factor may limit uptake through demand by depressing growth and yield of the rice or through availability of the nutrients released by burning. Further explanation is not yet possible from data obtained so far.

This paper has shown that upland rice in a 7-year rotation yielded three times as much grain with pada at 0.42 trees m^{-2} as with 0.10 trees m^{-2} in the fallow, and that the effect was not due to some pre-existing condition in the high pada patches. The factor most immediately relevant to the farmers of Huai Tee Cha, and others who similarly depend on rotational shifting cultivation for their living in the mountainous region of mainland Southeast Asia, is if and how this effect of dense pada may be transferred to areas with sparse or no pada. The answer may be dependent on identification of (a) factors that are limiting to the survival of pada seedlings, and (b) how pada affects rice yield. Both of these are currently under investigation.

Table 6. The yield of upland rice in seven and four years rotation, in areas following pada at low and high densities during the fallow period, in a rotational shifting cultivation system in northern Thailand.

Pada density	Dense	Sparse	Dense	Rotation effect
Rotation	7 years	7 years	4 years	
	Yield (Mg ha^{-1})			
Grain	2.57[a]	0.83[b]	0.74[b]	$p < 0.01$
Straw	2.35[a]	0.97[b]	0.72[c]	$p < 0.01$
Total	4.92[a]	1.79[b]	1.47[c]	$p < 0.01$
Harvest Index	52.3	46.1	50.7	

a) Numbers in same row followed by different letters are significantly different by LSD ($p < 0.05$).

Table 7. Range and variation of rice yield in seven year rotation, in areas following pada at low and high densities during the fallow period, in a system of rotational shifting cultivation in northern Thailand.

Yield	Grain		Straw	
Pada density	Dense	Sparse	Dense	Sparse
	Mg ha^{-1}			
Maximum	4.53	1.56	3.80	1.75
Minimum	2.48	0.71	2.21	0.86
Mean (of 8 fields, 6 farms)	3.04	1.15	2.74	1.19
Standard deviation	0.71	0.33	0.59	0.30

Table 8. Yield components of upland rice in a rotational shifting cultivation system in northern Thailand with dense and sparse pada densities during the fallow period.

Yield component	Pada density		Significant Difference[a]
	Dense	Sparse	
Hills m^{-2}	6.2	5.5	**
Tillers hill^{-1}	13.9	10.3	*
Panicles hill^{-1}	12.3	7.1	**
Panicles m^{-2}	76.7	39.0	**
Fertile panicles (%)	90.1	66.6	**
1000 seed weight (g)	29.1	29.1	NS

a) NS = not significant ($p < 0.05$); ** = significant ($p < 0.01$)

Table 9. Nutrient concentration and contents at maturity of the rice after fallow with dense and sparse pada in a rotational shifting cultivation system in northern Thailand.

	Grain[a]		Straw[a]	
Pada density	Dense	Sparse	Dense	Sparse
(a) Nutrient concentration (%)				
N	1.13[a]	1.42[b]	0.48	0.63
P	0.23[a]	0.31[b]	0.04[a]	0.10[b]
K	0.42	0.45	2.47[b]	1.91[a]
Ca	0.11	0.10	0.33	0.36
Mg	0.11[a]	0.13[b]	0.13	0.13
(b) Nutrient content (kg/ha)				
N	34.1[b]	13.9[a]	13.6[b]	7.3[a]
P	7.6[b]	3.3[a]	2.1[b]	1.0[a]
K	13.1[b]	5.4[a]	62.9[b]	16.1[a]
Ca	3.1[b]	1.1[a]	8.1[b]	3.5[a]
Mg	3.4[b]	1.4[a]	3.3[b]	1.5[a]

a) Different letters designate significant difference (by LSD, $p < 0.05$) between dense and sparse pada patches for grain or straw yield.

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References

Kaya B. and Nair P.K.R. 2001. Soil fertility and crop yields under improved-fallow systems in southern Mali. Agrof Syst 52: 1–11.

Kwesiga F. and Coe R. 1994. The effect of short rotation *Sesbania sesban* planted fallows on maize yield. For Ecol Mgt 64: 199–208.

Kunstadter P. 1978. Subsistence agricultural economics of Lua' and Karen Hill Farmers, Mae Sariang District, Northwestern Thailand. In: Kunstadter P., Chapman E.C. and Sabhasri S. (eds), Farmers in the Forest. The University Press of Hawaii, Honolulu, pp. 71–133.

Mafongoya P.L. and Nair P.K.R. 1997. Multipurpose tree prunings as a source of nitrogen to maize under semiarid conditions in Zimbabwe. Part 1. Nitrogen-recovery rates as influenced by pruning quality and methods of application. Agrof Syst 35: 31–46.

Nye P.H. and Greenland D.J. 1964. Changes in the soil after clearing a tropical forest. Plant Soil 21: 101–112.

Rerkasem K. and Rerkasem B. 1995. Montane mainland South-east Asia: agroecosystems in transition. Global Environmental Change 5: 313–322.

Reuter D.J., Edwards D.G. and Wilhelm N.S. 1997. Temperate and tropical crops. Plant Analysis, an Interpretation Mannual. : 81–285.

Sanchez P.A. 1976. Properties and Management of Soils in the Tropics. John Wiley & Sons New York, New York, 618 pp.

Sabhasri S. 1978. Effects of forest fallow cultivation on forest production and soil. In: Kunstadter P., Chapman E.C. and Sabhasri S. (eds), Farmers in the Forest. The University Press of Hawaii, Honolulu, pp. 160–184.

Tarawali G. 1991. Effect of Stylosanthes fodder banks on grain yield of maize. Trop Grass 25: 26–31.

Wanatabe F.S. and Olsen S.R. 1962. Colorimetric determination of phosphorus in water extracts of soil. Soil Sci 93: 183–188.

Whitmore T.C. 1982. On pattern and process in forests. In: Newman E.I. (ed.), The Plant Community as Working Mechanism: Special Publication of the British Ecological Society Produced as a Tribute to A.S. Watt. Blackwell Scientific Publication, Oxford, pp. 45–59.

Youpensook S., Lumyong S., Yimyam N. and Dell B. Biodiversity of Arbuscular mycorrhizal fungi from rhizosphere of *Macaranga denticulata* (Euphorbiaceae) on the highland rotational shifting cultivation area in Thailand. Aust. J. Botany (in press).

Zinke P., Sabhasri S. and Kunstadter P. 1978. Soil fertility aspects of the Lua forest fallow system of shifting cultivation. In: Kunstadter P., Chapman E.C. and Sabhasri S. (eds), Farmers in the Forest. University Press of Hawaii, Honolulu, pp. 134–159.

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Contents

MINI REVIEW

Cover photo: Gene Hettel

Determinants of a premium-priced, special-quality rice

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Thai Jasmine or Thai Hom Mali is Thailand's special-quality rice for which local consumers and export markets are willing to pay a premium price. Thai Jasmine rice does not always receive a premium price from buyers. The price varies, depending on where the crop is grown. For example, milled rice (special grade, 100% head rice) from Prao is sold at a retail price of 22 baht kg^{-1} (US$1 = 43 baht), that from Sanpatong costs 20 baht kg^{-1}, whereas Mae Chan rice fetches 16 baht kg^{-1}. Milled rice from Prao with 5% broken grain is priced at 17 baht kg^{-1}. In this paper, we examined the relationship between quality parameters used by commercial rice buyers and the price they pay for KDML 105.

Twenty-seven 500-g samples of unhusked KDML 105 were collected from farmers' fields in Chiang Mai in the upper part of northern Thailand, as well as 20 samples from Nakornsawan in the lower north. The samples were priced and rated for quality characteristics by a commercial rice buyer (Chiangmai Chaiwiwat Ricemill Co., Ltd.). The quality characteristics evaluated were grain moisture, percentage head rice yield, aroma, vitreousness, and translucency. Grain moisture was measured by a Riceter series L grain moisture tester (Kett Electric Laboratory). A small sample (20–30 g) of rice was unhusked on a board with a roller. An experienced rice buyer rated percentage head recovery, vitreousness, translucency, and aroma, using a 0–3 scale.

All 27 samples from Chiang Mai were priced above 7,000 baht t^{-1} (US$162). In contrast, samples from Nakornsawan were more varied and received prices ranging from less than 5,500 to more than 7,000 baht t^{-1}. Only 10% of the samples from Nakornsawan were judged to be of premium quality and priced at more than 7,000 baht t^{-1}; 40% were priced at less than 5,500 baht t^{-1}. Grain moisture did not exceed the standard 14% in any of the samples—those from Chiang Mai ranged from 13.3% to 14.0%, while the Nakornsawan samples had 11.2–12.0%. The prices of Nakornsawan samples were determined primarily by their aroma and vitreousness scores, with percentage head rice and translucency having relatively minor effects (equation 1).

$$P = 3{,}259.7 + 28.8X_1^{*} + 403.5X_2^{*} + 357.1X_3^{*} + 71.9X_4^{ns} \quad r^2 = 0.82^{***} \quad (1)$$

where P = sample price, X_1 = percentage head rice, X_2 = aroma score, X_3 = vitreousness, score, and X_4 = translucency score.

All 27 samples from Chiang Mai received the full score of 3 for vitreousness, translucency, and aroma. Price was determined by only one quality characteristic, percentage head rice (equation 2).

$$P = 6{,}969.4 + 11.3X_1^{***} \quad (2)$$
$$r^2 = 0.92^{***}$$

These results illustrate the effect of geographic differences on special-quality rice. The Chiang Mai samples represent areas that produce rice with high-quality characteristics in terms of aroma, grain translucency, and vitreousness. In such a situation, attention to factors that influence percentage head rice would ensure that farmers are paid premium prices for their harvest. Grain moisture at harvest and during application of N fertilizer has been shown to strongly affect head rice percentage (Nangju and De Datta 1970, Jongkaewattana et al 1993). The problem is much more complicated for farmers in areas where important quality parameters, such as aroma and vitreousness, are highly variable as we found in Nakornsawan. In such a situation, production of Thai Jasmine rice that will fetch premium prices will not be possible until environmental factors governing these quality parameters are identified and controlled.

References

Jongkaewattana S, Geng S, Brandon DM, Hill JE. 1993. Effect of nitrogen and harvest grain moisture on head rice yield. Agron. J. 85:1143-1146.

Nangju D, De Datta SK. 1970. Effect of time of harvest and nitrogen level on yield and grain breakage in transplanted rice. Agron. J. 62:468-474.

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The effect of nitrogen on rice grain iron

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Previous studies have shown that iron (Fe) content of rice grain may vary widely among rice genotypes (Senadhira et al., 1998; Prom-u-thai and Rerkasem, 2001). In addition, grain Fe may also be affected by environmental and management conditions. This experiment measured grain Fe concentration in five rice genotypes (KDML105, IR68144, UBON2, BASMATI370 and RD6) grown under three levels of N (0, 60, 120 kg N ha^{-1}). The field experiment was in a split plot design with three replications. Basal fertilizer consisted of 15 kg P_2O_5 and 15 kg K_2O ha^{-1}. The basal fertilizer and half of the N were applied at transplanting, and the other half of N was applied after four weeks. Fe concentration was determined in mature grain, as unhusked (whole grain with palea and lemma intact) and brown rice (palea and lemma removed), husk (palea and lemma) and polished grain (30 second), by dry-ashing and atomic absorption spectrometry (Emmanuel *et al.*, 1984).

In all five genotypes, grain yield was increased slightly by the application of 60 kg N ha^{-1}, but there was no further effect of increasing N to 120 kg N ha^{-1} ($p < 0.05$) (Table 1). Nitrogen fertilizer generally increased N content of the rice grain (Table 2). On the other hand, grain Fe concentrations in the five genotypes were affected by N rate differently (Table 3). The Fe concentration in unhusked rice of KDML105, IR68144, Basmati370 and RD6 increased with N to the rate 120 kg N ha^{-1}. Nitrogen had no effect on Fe in unhusked grain of UBON2. Much higher concentrations of Fe were found in the rice husk compared with the rest of the grain. Nitrogen levels had no effect on Fe concentration in brown rice and husk. Basmati370 was an exception, the Fe in brow rice

of this genotype was increased with N to the rate of 120 kg N ha^{-1}. To produce white rice, the mill normally polishes brown rice for about 30 seconds. In this study, we found that grain Fe concentration generally declined after polishing, indicating that a high proportion of Fe is contained in the bran or polishing. In UBON2 and RD2, grain Fe in white rice in N60 and N120 was about twice as much that in N0. There was similar, although slightly less, effect of N in Basmati370. In IR68144 the grain Fe in white rice at N120 was slightly less than these at N0 and N60, but in KDML105, the grain Fe in white rice showed no response to nitrogen. However, the grain Fe in unhusked, brown and white rice were not correlated with grain N in rice grain and grain yield at three levels of N application.

Different parts of the rice grain: the husk, the bran and the endosperm, appeared to contain Fe at different concentrations and their Fe contents responded differently to nitrogen fertilizer. The effect of nitrogen fertilizer on grain Fe appeared to be mostly in the husk. The Fe concentration in unhusked rice was only weakly correlated with that in brown rice ($r = 0.66$) and white rice ($r = 0.42$).

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Refferences

Emmanuel D, Dell B, Kirk G, Loneragan J, Nable R, Plaskett D, Webb M. 1984. Manual of Research Procedures. First edition. Plant nutrition research group school of environmental and life science. Murdoch University, Australia.

Prom-u-thai C, Rerkasem B. 2001. Grain iron concentration in Thai rice germplasm. *Dev. Plant Soil Sci.* 92, 350-351.

Senahira D, Gregorio G, Graham RD. 1998. Breeding iron and zinc – dense rice. Paper presented at the International Workshop on Micronutrient Enhancement of Rice for Developing Countries, September 3, 1998. At Rice Research and Extension Center, Stuttgard, Arkansas.

Table 1 Grain yield (t ha^{-1}) of five genotypes grown at three levels of nitrogen (0, 60, 120 kg N ha^{-1})

Genotypes	Grain yield (t ha^{-1})		
	N0	N60	N120
KDML105	3.6	4.0	3.8
IR68144	2.7	4.7	3.9
UBON2	4.1	4.0	3.7
RD6	3.1	3.4	3.3
BASMATI370	3.7	3.3	3.4
	3.2 a	3.8 b	3.6 b
Analysis of variance			
p(Genotypes)	ns		
p(Nitrogen)	< 0.05		
p(G X N)	ns		
LSD (nitrogen, 0.05)	0.4		

Table 2. The N concentration (%N) in unhusked rice of five genotypes grown under three levels of N (0, 60, 120 kg N ha^{-1})

Genotypes	Grain N concentration (%N)		
	N0	N60	N120
KDML105	1.3 aA	1.6 aB	1.8 bB
IR68144	1.5 aA	1.6 aA	1.9 bcB
UBON2	1.5 aA	1.6 aA	2.0 cB
RD6	1.3 aA	1.6 aB	1.8 bB
BASMATI370	1.3 aA	1.5 aA	1.5 aA
Analysis of variance			
p(Genotypes)	< 0.01		
p(Nitrogen)	< 0.01		
p(G X N)	< 0.01		
LSD (nitrogen, 0.05)	0.30		
LSD (genotypes, 0.05)	0.20		

Lower case for comparison of the genotype effect

Upper case for comparison of the N effect

Table 3. The Fe concentration in unhusked, brown rice husk and polished grain (white grain) at 30 and 60 seconds of five genotypes grown under three levels of N (0, 60, 120 kg N ha^{-1}

Genotypes	N	Fe concentration (mg Fe kg^{-1})			
		Unhusked	Brown rice	White (30s)[§]	Husk
KDML105	N0	13.0 a	7.8 a	7.9 a	36.0 a
	N60	14.6 ab	8.2 a	7.2 a	32.6 a
	N120	16.1 b	8.8 a	7.3 a	37.4 a
IR68144	N0	15.8 a	13.5 a	12.3 b	37.9 a
	N60	17.2 a	13.0 a	13.6 b	38.3 a
	N120	21.1 b	13.1 a	10.0 a	48.6 a
UBON2	N0	13.8 a	9.3 a	4.6 a	42.7 a
	N60	13.6 a	8.1 a	8.8 b	44.8 a
	N120	13.5 a	8.3 a	6.5 ab	48.5 a
RD6	N0	15.2 a	8.2 a	5.1 a	39.8 a
	N60	15.8 b	8.3 a	10.3 b	51.3 a
	N120	18.3 b	9.0 a	8.2 b	44.2 a
BASMATI	N0	15.6 a	10.8 a	7.6 a	35.8 a
	N60	16.3 a	12.1 ab	6.5 a	37.5 a
	N120	18.3 b	12.4 b	10.2 b	47.3 b
Analysis of variance					
p(Genotypes)		< 0.01	< 0.01	< 0.01	< 0.01
p(Nitrogen)		< 0.01	ns	< 0.05	< 0.05
p(G X N)		< 0.05	< 0.05	< 0.01	< 0.05
LSD (0.05)		2.6	1.5	2.3	11.5

*LSD used to comparison the different at the same genotypes at different treatment
[§] Polished for 30 seconds

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Agrodiversity Lessons in Mountain Land Management

Intensification of crop production in the mountains has long been perceived as unsustainable. However, since the late 1980s it has become increasingly evident that decline and collapse are not always inevitable. The present article provides examples from the highlands of northern Thailand to show that local food security can be improved while impacts on the resource base and the environment are minimized. This was achieved with the help of cropping systems developed and adapted by farmers themselves (Figure 1). Studying farmers' management techniques will allow this success to be repeated elsewhere, but only if it is based on the idea of dynamic variation in cropping system management that occurs within and between mountain agroecosystems, defined as agrodiversity.

FIGURE 1 A mosaic of mountain land uses: vicinity of Mae Cham village, Chiang Mai, Thailand. (Photo by Kanok Rerkasem)

A mountain agroecosystem on the verge of collapse

The following account of the Hmong village of Pah Poo Chom (see following description) found in the files of the Tribal Research Center in Chiang Mai aptly portrays the dire situation of the highlands of northern Thailand in the 1970s and 1980s:

> After seven years most of the surrounding forests had been cleared and cropped with rice, opium and maize. There was increasing competition for land from the neighboring villages of Thai, Lahu and Lisu. In 1970 the village grew 35 *rai* (5.6 ha) of opium, but the yield was very low. The village was extremely poor, and more than 55% of the total adult population was addicted to opium. Many had to make a living from employment outside the village.

The highlands of northern Thailand are populated by several ethnic groups. The people originally made a living from slash-and-burn systems of land use broadly classified as rotational and pioneer types of shifting cultivation. Rotational shifting cultivators (Karen, H'tin, Lua, and Khamu) typically settled in one place to grow rice and associated crops in a system of rotation involving 1 year of cropping and 9–14 years of fallow. Pioneer shifting cultivators (Hmong, Lahu, Lisu, Yao, Akha, and Haw) were migratory.

It was a harsh way of life with a constant risk of crop failure. The system was