Evaluation of Sink and Source Relationship in Different Rice (*Oryza Sativa* L.) Cultivars

1Davood Eradatmand Asli, 2Anoosh Eghdami and 1Alireza Houshmandfar

1Department of Agronomy and Plant Breeding, Islamic Azad University, Saveh branch, Saveh, Iran
2Department of Biology, Islamic Azad University, Saveh branch, Saveh, Iran

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ABSTRACT

A field experiment was conducted to determine whether the grain yield of different rice (*Oryza sativa* L.) cultivars included beejar, khazar and binam is limited by the availability of substrates or by the capacity of the grains to assimilate and utilize the available substances. The sink and source ratio manipulation treatments included I. control, II. cutting flag leaf blade, III. thinning half of the plants, IV. thinning and cutting flag leaf blade. The potential grain weight was analyzed by a decrease of 50 percent of grains at anthesis. The distribution of dry matter accumulation between kernels was determined at harvest. Modification of sink and source ratio led to different patterns of allocation in dry matter accumulation between cultivars. The pattern of partitioning of dry matter accumulation observed in this study suggests sink and source limitation in binam and beejar cultivars, respectively. Furthermore, in khazar cultivar there was a relative balance between sink and source and none of the two factors were limited.

Key words: Source, sink, flag leaf, thinning, rice

Introduction

A substantial increase in yield potential is required to ensure food security in future decades. In striving to increase the yield potential, it is important to determine the physiological factors limiting yield. The first step towards this is to evaluate whether the growth of harvested organs is limited by the availability of substrates or by the capacity of the organ to assimilate and utilize the available substances for growth [29,35,45]. Attempts to identify physiological factors limiting yield must integrate sink and source interactions both spatially and temporally [29]. Results from the response of kernel weight and grain set to sink and source manipulations suggested yield limitation by both sink and source, depending on the seasons, genotypes, etc [12,1]. However, the time courses of sink and source control have not been well documented. Some authors [9,18,32] suggested that grain yield in rice and wheat may not be limited by the supply of carbon at any time during grain filling. However, there are data showing significant increase in mass per grain associated with reduction in grain number [13,22,21,19] implying source limitation at least on some occasions after anthesis. Analysis of sink and source interactions should also consider the role of alternative sinks in the plant [33]. In wheat, particularly, special attention should be given to the stems, since competition exists between the growing upper internodes and reproductive organs in the weeks before anthesis [42,4,7,8,37] the outcome of which depends on both genotype and environment. Furthermore, there is good evidence that temporary storage is very important under stress conditions [3,5,34]. Manipulation of sink and source ratios by control have not been well documented. Some authors [9,18,32] suggested that grain yield in rice and wheat may not be limited by the supply of carbon at any time during grain filling. However, there are data showing significant increase in mass per grain associated with reduction in grain number [13,22,21,19] implying source limitation at least on some occasions after anthesis. Analysis of sink and source interactions should also consider the role of alternative sinks in the plant [33]. In wheat, particularly, special attention should be given to the stems, since competition exists between the growing upper internodes and reproductive organs in the weeks before anthesis [42,4,7,8,37] the outcome of which depends on both genotype and environment. Furthermore, there is good evidence that temporary storage is very important under stress conditions [3,5,34]. Manipulation of sink and source ratios by control have not been well documented. Some authors [9,18,32] suggested that grain yield in rice and wheat may not be limited by the supply of carbon at any time during grain filling. However, there are data showing significant increase in mass per grain associated with reduction in grain number [13,22,21,19] implying source limitation at least on some occasions after anthesis. Analysis of sink and source interactions should also consider the role of alternative sinks in the plant [33]. In wheat, particularly, special attention should be given to the stems, since competition exists between the growing upper internodes and reproductive organs in the weeks before anthesis [42,4,7,8,37] the outcome of which depends on both genotype and environment. Furthermore, there is good evidence that temporary storage is very important under stress conditions [3,5,34]. Manipulation of sink and source ratios by
artificial reduction in grain number per inflorescence has been used in several cereal grain species to estimate potential kernel weight and study the grain filling process [6,38,10,28]. Actual kernel weight is less than potential kernel weight because of competition among kernels for available assimilate and interplant competition for light, water and nutrients [30]. Kernel weight in cereal spikes generally increases in response to reduced kernel number per spike [13,30,26], although kernel weight reductions have been reported [13] it is assumed that such kernel weight increase occurs because assimilate available to each remaining kernel increases [30,38]. Potential kernel weight is obtained when kernel number is reduced to a point at which competition among kernels for assimilate no longer exists [13,35,30]. Fischer and Laing [14] and Martinez-Carrasco and Thorne [24] have used thinning as a technique for increasing photosynthate supply for developing kernels and increasing kernel weight. Removal of the flag leaf [41] or a portion of it [43] has been used to reduce the amount of photosynthate available to developing kernels. It is possible that small-seeded cultivars are more sensitive to photosynthate supply. If this is true, treatments such as thinning, flag leaf removal, and spikelet removal should have different effects on small- and large-seeded cultivars. The aim of this study was to examine the performance of sink and source interactions after anthesis to evaluate the possible factor(s) limiting grain yield in rice cultivars. Artificial manipulations of the sink and source ratio and evaluation of the variation in dry matter partitioning in three rice cultivars was used to assess the existence of genotypic differences in the response of availability of photoassimilates.

**Materials and methods**

*Experimental Setup:*

A field experiment was conducted at the Rice Research Institute of Iran (RRII) in Rasht (37°16´ N, 49°36´ E; 7 m above mean sea level), located in Guilan state of northern Iran. This site has Mediterranean climate and a silty loamy soil. The experiment was arranged in a randomized complete block design (RCBD) with a split plot arrangement with four replicates. Plots were fertilized at sowing with 90, 20, 60, and 3.5 kg N, P, K, and Zn ha⁻¹, respectively. Entire amount of all the fertilizers except N was applied prior to transplanting while N was applied in three equal splits at 4ᵗʰ, 21ˢᵗ and 52ⁿᵈ day after transplanting. Plots were occasionally sprayed with fungicides to avoid disease and were hand weeded. Thirty-five days old rice seedlings of three cultivars included beejar, khazar and binam, were transplanted in plots consisted of seven rows, 7 meters long spaced 20 centimeter. The distance between plants was 20 centimeter. Treatments consisted of a factorial combination of three cultivars and four sink and source manipulations treatments. Main plots consisted of the three cultivars and the sub plots consisted of the four sink and source manipulation comprising I. control, II. flag leaf blade removed, III. thinning consisted of cutting rows 2, 4 and 6 to ground level and IV. thinning and cutting flag leaf blade. For determining potential grain weight 20 main shoots from the central rows of each plot were tagged and detillered to avoid the tillers becoming alternative sinks for mobilized carbohydrates [39], sink strength of main stems was decreased by half at anthesis by sterilizing 50 percent of fertile basal and lateral florets of the panicle. Total treatment was applied at anthesis stage, the day when anthers were extruded in 50 percent of the panicle in a plot [17].

**Plant Sampling and Analysis:**

At maturity, for determining the final grain and biological yields, one square meter from central lines in each plot after removing boundary harvested and hand threshed. For each plot ten plants randomly selected and component of yield and agronomic traits were measured. At maturity the potential grain weight were recorded by measuring the dry mass of grains in degraining panicle. Harvest index (HI) calculated from the following formula:

$$HI = \frac{\text{Economical yield}}{\text{Biological yield}} \times 100$$

At anthesis, the area of the main stem leaves and flag leaf was measured according to Yoshida [46] (Length × maximum width × 0.74). This formula represented the actual leaf area in the cultivars used in this experiment. Leaf area duration (LAD) was estimated according to Armas et al., [2] using the following equation:

$$LAD = \frac{A_2 - A_1}{\ln A_2 - \ln A_1} \times t_2 - t_1$$

Where $A_2$ and $A_1$ represent shoot leaf area at anthesis and physiological maturity respectively, and $t_2 - t_1$ represents the time between anthesis and physiological maturity. Data were analyzed statistically by analysis of variance (ANOVA) and means were compared by least significant difference (LSD) test [16]. The results of statistical analysis were considered significant when they were outside 95% confidence intervals.
Results and discussion

Table 1 indicates the analysis of variance for the grain yield, components of yield, and potential grain weight of rice cultivars. The grain yield was significantly affected by different cultivar treatments. The maximum and minimum levels of grain yield were observed at bejeear and binam cultivars, respectively. Thinning at anthesis stage resulted in an average increase of 24 percent in grain yield (Table 2). The interaction effect of cultivars × thinning was also significant for grain yield. An average thinning treatment resulted in increase of grain yield by 16, 38 and 15 percent in khazar, bejeear and binam respectively (Table 2). Hence, if there is enough supply of assimilate, beeejar cultivar could utilize it more than other cultivars, and it is necessary to determine optimum density and other inputs for this cultivar in different environment. The extent of the source at the time of anthesis sets an upper limit to potential sink size in binam and khazar cultivars. Removal of the flag leaf blade at anthesis resulted in a decrease of 12 percent in grain yield (Table 2). This more considerable decrease in grain yield shows that flag leaf has an important role in grain filling. Supply more assimilates from other sources such as flag leaf sheath and the leaves below the flag leaf can somewhat compensate the lack of flag leaf, although contribution of flag leaf in grain filling is more than 12 percent [11]. The flag leaf blade is the principal source of photoassimilates imported by grains during grain filling [31]. However, removal of the flag leaf may lead in some circumstances to enhancement of the photosynthetic activity of other leaves and green parts of plant [20] and remobilization of stored carbohydrates [33]. These mechanisms avoid the restriction of grain filling in such a manner that often no source limitation occurs [32,36]. The comparison between control and defoliated plants indicates that cultivars differed in the ability to remobilize reserves from the stems to the grains. Such differences in the response to availability of photoassimilates seem to be the consequence of different patterns of photoassimilate partitioning between cultivars with varying source and sink ratios [12]. In khazar, beeejar and binam cultivars, removal of flag leaf resulted in a decrease of 10, 18 and 6.5 percent grain yield respectively (Table 2). These results represent the possibility of remobilization of assimilates from secondary sources to grains in khazar and binam are more than beeejar. One of the reasons for more decreased yield in beeejar related to other cultivars is the large flag leaf area in this cultivar as compared to others (data not shown) as one of the most important factors in photosynthesis rate and supply assimilate to ear is flag leaf area [25]. Removal of spikelet did not alter the pattern of senescence of photosynthetic tissues, and hence there were no significant differences in leaf area duration (LAD) between control and degraining plants. LAD in flag leaf removal treatment was reduced due to lack of the flag leaf blade (Figure 1). Table 1 demonstrates that there were significant effects between thinning treatments and cutting of flag leaf blade. Whenever these two treatments occurred synchronously, grain yield increased about 9 percent (Table 2). It is suggested that the role of increasing thinning is more than the role of decreasing removal of flag leaf in grain yield, because stored materials in stems and leaf sheath at removed flag leaf conditions partially compensate of low assimilate. The study of components of yield revealed that thinning and removal of flag leaf blade treatments had no significant effects on number of panicle per unit area, which is as a result of the time of treatments application. However, thinning treatment at anthesis time increased tillering but most of them were infertile and thinning merely only increased biomass (Figure 2). Cock and Yoshida [9] have similar conclusions. Table 1 also indicates the number of grains per ear as influenced by cultivars. Highest and lowest numbers of grains per ear were observed at khazar and binam cultivars, respectively. According to Xu and Vergara [44] variability of total grain number in cultivars has a genetic basis which depends on growth length and plant height. Thus binam cultivar with a taller height and a weaker stem has lower number of grains per ear (Figure 3). The number of grains per ear was determined before panicle initiation, thus after determining primary grains continued growth and filling grain depends on supply assimilate from different parts of plant [40]. By manipulate of source potential ratio such as removal of flag leaf or thinning, some of physiological indices as sink and source capacity, carbohydrates stored and potential translocation of assimilates during stress conditions can be determined [25]. This present study has shown that in control plants of binam cultivar, nearly 91 percent of grains fully filled and matured (Figure 4). These results are consistent with findings of Matsushima [25] and Murty and Muty [27] which concluded that if matured grain ratio is more than 80 percent, capacity of sink is limiting factor, which revealed that the limiting factor of yield in this cultivar is sink capacity. In this direction total of filled grains per plant on khazar cultivar was about 80 percent (Figure 4) and showed that none of the factors i.e. capacity of sink and assimilate content are not limiting in this cultivar. On the other hand, in khazar cultivar there is a balance between sink and source. In this study, in beeejar cultivar about 71 percent of grains in control plant matured (Figure 4). According to the definitions of Matsushima [25] and Murty and Muty [27] since number of filled grains in this cultivar are less than 80 percent, therefore, supply of assimilate
Table 1: Analysis of variance of grain yield, components of yield and potential grain weight of three rice cultivars and four sink and source manipulations in rice.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>df</th>
<th>Grain yield (g m⁻²)</th>
<th>Biomass (g m⁻²)</th>
<th>HI (%)</th>
<th>No. of panicles m⁻²</th>
<th>No. of grain per panicle</th>
<th>1000-Grain weight (g)</th>
<th>Potential grain weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replication</td>
<td>3</td>
<td>176.25**</td>
<td>840.92**</td>
<td>0.64ns</td>
<td>11.55**</td>
<td>11.04**</td>
<td>0.04**</td>
<td>0.02**</td>
</tr>
<tr>
<td>Cultivars (V)</td>
<td>2</td>
<td>733551.39**</td>
<td>461602.08**</td>
<td>1013.66**</td>
<td>44984.31**</td>
<td>28340.74**</td>
<td>72.17**</td>
<td>55.04**</td>
</tr>
<tr>
<td>Thinning (T)</td>
<td>1</td>
<td>488440.33**</td>
<td>1573252.08**</td>
<td>17.40**</td>
<td>30.08**</td>
<td>1534.54**</td>
<td>84.00**</td>
<td>0.04**</td>
</tr>
<tr>
<td>Remove flag leaf (F)</td>
<td>1</td>
<td>179585.33**</td>
<td>315252.08**</td>
<td>51.46**</td>
<td>14.08ns</td>
<td>424.83**</td>
<td>41.25**</td>
<td>0.42*</td>
</tr>
<tr>
<td>V×T</td>
<td>2</td>
<td>34739.52**</td>
<td>45033.33**</td>
<td>51.46**</td>
<td>14.08ns</td>
<td>424.83**</td>
<td>41.25**</td>
<td>0.42*</td>
</tr>
<tr>
<td>V×F</td>
<td>2</td>
<td>34739.52**</td>
<td>45033.33**</td>
<td>51.46**</td>
<td>14.08ns</td>
<td>424.83**</td>
<td>41.25**</td>
<td>0.42*</td>
</tr>
<tr>
<td>V×T×F</td>
<td>2</td>
<td>34739.52**</td>
<td>45033.33**</td>
<td>51.46**</td>
<td>14.08ns</td>
<td>424.83**</td>
<td>41.25**</td>
<td>0.42*</td>
</tr>
</tbody>
</table>

*, significant at P<0.05
**, significant at P<0.01
ns, non significant

Table 2: Comparison of mean triple interaction between cultivar, thinning and flag leaf cutting on yield and components of yield in rice cultivars.

<table>
<thead>
<tr>
<th>Cultivar</th>
<th>Thinning</th>
<th>Remove flag leaf</th>
<th>Grain yield (g m⁻²)</th>
<th>Biomass (g m⁻²)</th>
<th>HI (%)</th>
<th>No. of panicles m⁻²</th>
<th>No. of grains per panicle</th>
<th>1000-Grain weight (g)</th>
<th>Potential grain weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Khazar</td>
<td>T₀</td>
<td>F₀</td>
<td>896.2</td>
<td>1750.8</td>
<td>51.1</td>
<td>314.3</td>
<td>162.8</td>
<td>24.2</td>
<td>25.4</td>
</tr>
<tr>
<td></td>
<td>F₁</td>
<td>850.5</td>
<td>1694.1</td>
<td>50.2</td>
<td>313.3</td>
<td>160.2</td>
<td>23.5</td>
<td>25.4</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>T₁</td>
<td>F₀</td>
<td>1114.8</td>
<td>2044.2</td>
<td>54.2</td>
<td>325.0</td>
<td>142.1</td>
<td>24.0</td>
<td>28.8</td>
</tr>
<tr>
<td></td>
<td>F₁</td>
<td>1005.9</td>
<td>1902.0</td>
<td>52.4</td>
<td>326.7</td>
<td>136.5</td>
<td>22.4</td>
<td>28.6</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>T₁</td>
<td>F₀</td>
<td>1298.9</td>
<td>2310.9</td>
<td>56.2</td>
<td>327.3</td>
<td>148.0</td>
<td>26.8</td>
<td>28.8</td>
</tr>
<tr>
<td></td>
<td>F₁</td>
<td>1189.5</td>
<td>2168.7</td>
<td>54.4</td>
<td>326.3</td>
<td>143.1</td>
<td>25.2</td>
<td>28.6</td>
<td>ns</td>
</tr>
<tr>
<td>Beejar</td>
<td>T₀</td>
<td>F₀</td>
<td>714.0</td>
<td>1801.3</td>
<td>39.6</td>
<td>328.7</td>
<td>80.0</td>
<td>28.2</td>
<td>28.3</td>
</tr>
<tr>
<td></td>
<td>F₁</td>
<td>690.6</td>
<td>1757.7</td>
<td>39.2</td>
<td>329.3</td>
<td>81.2</td>
<td>27.6</td>
<td>28.2</td>
<td>ns</td>
</tr>
<tr>
<td></td>
<td>T₁</td>
<td>F₀</td>
<td>764.4</td>
<td>1959.5</td>
<td>39.0</td>
<td>320.0</td>
<td>85.5</td>
<td>28.5</td>
<td>28.4</td>
</tr>
<tr>
<td></td>
<td>F₁</td>
<td>740.1</td>
<td>1915.4</td>
<td>38.7</td>
<td>229.0</td>
<td>84.0</td>
<td>28.0</td>
<td>28.3</td>
<td>ns</td>
</tr>
</tbody>
</table>

LSD (5%) 16.1 33.0 0.8 3.0 3.5 0.3 0.3

*, T₀ and T₁, represent unthinned and thinning treatments, respectively
**, F₀ and F₁, represent no cutting and flag leaf cutting of flag leaf treatments, respectively

Fig. 1: Effect of sink and source modification on leaf area duration (cm² d⁻¹); T₀ and T₁, represent unthinned and thinning treatments, respectively; F₀ and F₁, represent no cutting and flag leaf cutting of flag leaf treatments, respectively.

is a limiting factor of yield. Grain weight has more pronounced effects on grain yield, as we considered grain weight is different within the cultivars and binam and khazar with 28 and 24.2 mg had higher and lower grain weight respectively (Table 2). Thinning increased grain weight approximately 11 percent (Table 2). This increase was mainly due to supply of more assimilates to grains and a decrease of competition between plants. Thus after thinning the remaining plants may have more ability of using current photosynthates. Zia [48] in rice had similar conclusion. In three cultivars of khazar, beejar and binam thinning treatment increased grain weight by 7, 25 and 3 percent respectively (Table 2). Beejar and binam cultivars with 5.5 and 0.7 mg had highest and lowest increases respectively. Thus in beejar cultivar adjustment of yield with decreased composition between plant can occur through grain weight variation. If grain weight increased in responses to more supply of assimilates, it can be said that grains are under source limited [47]. In general, thinning treatment showed that beejar related to other cultivars is source limited and if this cultivar had more assimilate it can produce heavier grains.
Fig. 2: Effect of sink and source modification on number of fertile tillers per hill; $T_0$ and $T_1$, represent unthinned and thinning treatments, respectively; $F_0$ and $F_1$, represent no cutting and flag leaf cutting treatments, respectively.

Fig. 3: Effect of sink and source modification on stem height (cm); $T_0$ and $T_1$, represent unthinned and thinning treatments, respectively; $F_0$ and $F_1$, represent no cutting and flag leaf cutting treatments, respectively.

Fig. 4: Effect of sink and source modification on grain filling percentage; $T_0$ and $T_1$, represent unthinned and thinning treatments, respectively; $F_0$ and $F_1$, represent no cutting and flag leaf cutting treatments, respectively.

Cutting flag leaf resulted in decrease of grain weight by 7 percent and represents the importance of flag leaf supply material for grain growth. Of course more amount of decrease was compensated by flag leaf sheath, internodes and other leaves and even spikes photosynthesis [46]. If kernel weight was limited in some cultivars due to sink capacity and not in others, reductions of photosynthate availability
would cause greater decrease in non limited cultivars than in limited cultivars. An interaction between cultivar and flag leaf removal would be further evidence of limitations in sink size. In three cultivars of khazar, beejar and binam with removed flag leaf grain weight decreased by 4, 12 and 4 percent respectively (Table 2). These results show that, may be, remobilization from secondary sources in khazar and binam is more than beejar cultivar and higher photosynthetic activity of ear in these two cultivars as compared to beejar cultivar. Source limitation in beejar cultivar could be the cause of the significant decrease in specific mass of grain in cutting flag leaf treatment as compared to other cultivars. In general grains weight more than number of grain is under treatment as compared to other cultivars. In general decrease in specific mass of grain in cutting flag leaf beejar cultivar could be the cause of the significant as compared to beejar cultivar. Source limitation in photosynthetic activity of ear in these two cultivars with removed flag leaf were not the same for all cultivars, as indicated highly significant difference between these cultivars in analyses of variance. Spikelet removal at anthesis resulted in a large increase in some cultivars (Table 2). Sink reduction not only decrease the competition for assimilates among growing grains, but eliminated, if existing, physical size constraints affecting the development of florets. The effects of spikelet removal were not the same for all cultivars, as indicated highly significant difference between these cultivars in analyses of variance. Spikelet removal at anthesis resulted in significant increase in potential grain weight for beejar and khazar cultivars but a small increase in average potential grain weight in binam 4.8. The significant increase in potential grain weight of beejar (4.8 mg) and khazar (1.2 mg) suggested that small-seeded cultivars may have greater grain weight response to spikelet removal than large-seeded cultivars, as insignificant increased grain weight for large-seeded binam cultivar (0.1 mg) support such a thesis. However, there are data in the literature showing a significant increase in mass of grains associated with reductions in grain number after anthesis, implying source limitation, at least some time during grain filling 15. The small-seeded cultivars included beejar and khazar tended to show the greatest response to spikelet removal. These results agree with those of Ma et al. [23] and Blum et al., [5].

Conclusion:

The pattern of partitioning of dry matter accumulation observed in this study suggests a diverse sink and source relationship in different rice cultivars. The sink limitation could explain the lack of growth of the remaining grains in half-panicle plants in binam cultivar. Whereas grain yield in beejar cultivar was limited by source activity rather than sink size. Furthermore, there is a relative balance between sink and source in khazar cultivar and none of the two factors were limited grain growth of this genotype.

References