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Modification of Flag Leaf Senescence and Yield Characters in Barley (*Hordeum vulgare* L.) by Gibberellic Acid and Kinetin

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Abstract. Barley plants (*Hordeum vulgare* L.) received foliar applications of 10^{-4} M gibberellic acid (GA_3) and Kinetin (KN) individually and in combination at one or more of three growth stages: flag leaf appearance (I), ear emergence (II), and the first stage of senescence initiation in the flag leaf (III). Both plant growth regulators (PGR) hastened onset of senescence when sprayed at Stage I and/or Stage II. Treatment at Stage III, either alone or in combination with treatments at the other stages, tended to postpone senescence. Yield components also showed stage-dependent response: Stage I treatment increased the formation of total and bearing tillers, and Stage III treatment improved grain number and weight. However, while GA_3 proved more effective than KN, the two together acted antagonistically.

The flag leaf is the major source of photosynthate for developing grains in cereals. The contribution varies from 30 to 80% depending upon the method used and the crop in question (Thorne 1966, Yoshida 1972, Evans 1975, Ho 1978, Gifford and Evans 1981). In general, high yields are associated with a prolonged duration of leaf area available for photosynthesis following anthesis (Thorne 1961, Welbank et al. 1966, Mohiuddin and Croy 1980). A positive correlation between the flag leaf area index and the grain yield has been reported (Dhiman et al. 1980). Increased leaf area duration and high yields in modern hexaploid wheat cultivars are correlated (Evans 1975). The photosynthetic activity in leaves is highest in newly expanded leaves and declines thereafter during senescence (Wittenbach 1979, Patterson and Moss 1979, Woolhouse 1982). Leaf senescence in cereals occurs during the postanthesis period, causing a rapid decline in photosynthetic activity, at a time when grain dry weight is still increasing (Patterson et al. 1980). Such changes may limit the

ultimate grain yield; delaying the onset of leaf senescence may prove beneficial (Secor et al. 1983). Delay in onset of senescence could either be induced by chemical growth regulators or by genetic manipulations (Woolhouse 1982). Of late, plant growth regulators have proved useful in altering leaf senescence (Gifford and Evans 1981, Woolhouse 1982). However, there is insufficient information on the influence and interaction of plant growth regulators on yield components. The present study is aimed to examine the role of gibberellic acid (GA_3) and Kinetin (KN) in modifying barley flag leaf senescence and yield attributes.

Materials and Methods

Seeds of huskless barley (*Hordeum vulgare* L. cv 292) were surface-sterilized by dusting with 1% mercuric chloride powder. Ten seeds were sown in pots containing approximately 15 kg soil mixed with farmyard manure (2:1). These pots were kept in the open during the Rabi season (October through March) at a temperature range of 15–41°C (the mean ranging between 23°C and 34°C). Seedlings were thinned to three pot^{-1} after a fortnight (fourth leaf stage). A 15:15:15 NPK fertilizer (Suphala from Rashtriya Chemicals & Fertilizers, Bombay, India) was applied twice to the pots at the rate of 5 g pot^{-1} on the 15th and 45th day after sowing. There were approximately 100 plants per treatment at the time of foliar spraying of PGR. Whole plants were sprayed (to run-off) thrice on alternate days with aqueous solutions (containing 0.1% Tween-20) of GA_3 (90% purity, obtained from K&K Labs, Plainview, NY) and KN (AR grade, obtained from SISCO Research Labs, Bombay, India) at 10^{-4} M. For the GA_3 + KN treatments, plants were sprayed consecutively with each PGR with a 30-min delay between the two sprays. Treatment stages were: flag leaf (identifiable) appearance (Stage I), fully expanded flag leaf or ear emergence (Stage II), and the initiation of senescence (tip yellowing) in the flag leaf (Stage III). These stages correspond to the 41st, 60th, and 77th day from the date of sowing. Aqueous solutions (5 ml plant^{-1} spraying^{-1}) containing 0.1% Tween-20 but devoid of PGRs were used for spraying (to run-off) control plants. Treatments were also repeated stagewise; namely I + II, II + III, and I + II + III.

Data on flag leaf senescence and several vegetative and reproductive characters were recorded periodically. The number of plants having completely yellow flag leaves on the main culm were recorded daily. The final data are expressed as percent of control. In general, data were recorded in terms of total plant height, total bearing tiller plant^{-1}, and the grain holding capacity of the ears, etc. Reproductive characters on pot^{-1} basis were noted in terms of grain yield (g), developed grain numbers, and 1,000 grain weight (g), etc.

The experiment was designed in a completely randomized block system. Each of the four blocks (replications) contained nine pots per treatment/control, there being three plants per pot^{-1}. Comparisons of control with individual treatment have been done using the *t* test of significance.

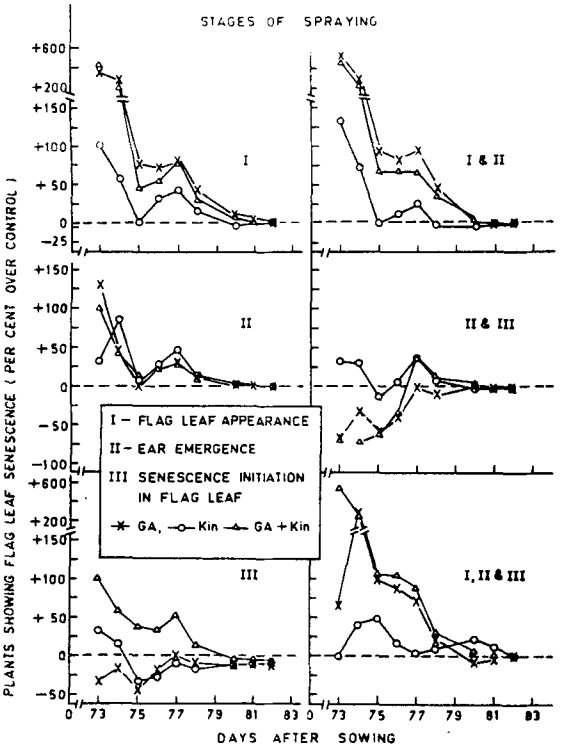

Fig. 1. Effect of foliar sprays with GA_3, KN, and GA + KN solutions given at one or more of three specified stages of growth on flag leaf senescence in barley.

Results

Changes in the senescence behavior of flag leaves due to PGR treatment with GA_3 and KN are given in Fig. 1. In general, at Stage I these compounds hastened the visible senescence. Such an effect was less marked at Stage II, and at Stage III there was delaying of senescence except in the GA_3 + KN treatment. Whenever treatments were repeated in combination with Stage I, the hastening was increased considerably as may be seen from I + II and I + II + III stage combinations. On the contrary, whenever Stage III treatment was combined with Stage II it was characterized by delaying the senescence. GA_3 and GA_3 + KN showed maximum hastening of senescence, especially whenever given at Stage I or its combination with other stages. In contrast, there was the maximum senescence delay by GA_3 and KN at Stage III.

Observations on vegetative characters are presented in Fig. 2. As expected, plant height was affected most by GA_3 treatment either given alone or in combination as GA_3 + KN. KN alone did not affect this character, even proving inhibitory at times, especially whenever given at Stage I. Such an inhibitory effect was annulled in the presence of GA_3, but only at Stage I. Even the GA_3 + KN combination at Stages II, III, or II + III affected plant height adversely. Stagewise response to GA_3 treatment either alone or in combination with KN on bearing tillers was similar to plant height, except that KN treatment at all stages and at various stage combinations was significantly promotive. Total

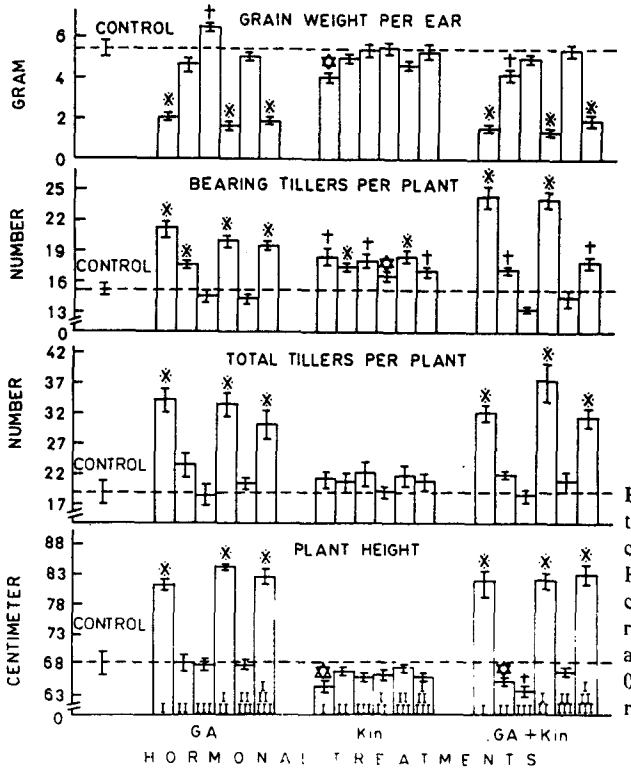

Fig. 2. Effect of hormonal treatments on the vegetative characters of the barley plants. Horizontal dotted lines give control level and vertical bars represent variation as SE, *, †, and ☆ signs show significance at 0.01, 0.05, and 0.10 *P* levels, respectively.

tillers plant^{-1} was significantly affected by Stage I treatments as well as its combination with other stages. Such an effect was more pronounced in GA_3 and also in its combination with KN but not in KN alone. Surprisingly, grain weight ear^{-1} was drastically curtailed by all treatment at all the stages, except that of GA_3 treatment given at Stage III. GA_3 and GA_3 + KN at Stages I, I + II, and I + II + III proved inhibitory. KN treatment given alone at Stage I adversely affected this character.

Yield components are shown in Fig. 3. GA_3 and KN, when sprayed at Stage III, caused a significant increase in grain yield. There was a drastic reduction in yield by GA_3 alone or in combination with KN at Stage I and also its combination with other stages. Such a deleterious effect seems to be due to a decrease in the number of developed grain with a concurrent increase in underdeveloped grain populations. Such was the case whenever plants were treated with GA_3 + KN at Stages I, I + II, and I + II + III. The reverse was true in the case of GA_3 and KN treatment at Stage III. KN treatment produced more developed grains at Stages II and III. The 1,000 grain weight was increased most by GA_3. KN alone had no effect, or decreased 1,000 grain weight as Stages I and II + III.

Correlative data on grain development in terms of number and size and its relation with the stage of treatment are presented in Fig. 4. The linear regressions were calculated by the least-squares technique. There is a linear increase both in grain size (1,000 grain weight) and number with the advancing growth

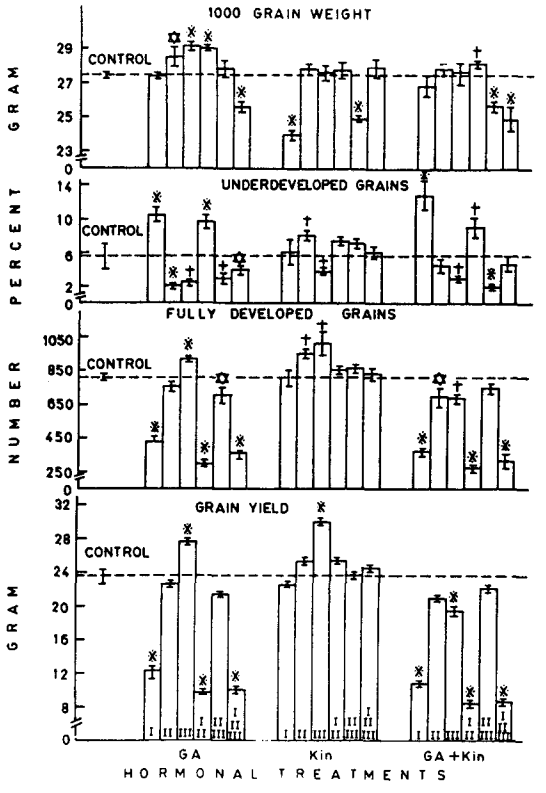

Fig. 3. Effect of hormonal treatments (as in Fig. 1) on reproductive attributes of barley plants. Other details are as in Fig. 2.

stages. Linearity was most distinct in GA_3 alone or in combination with KN compared to KN alone. There were two discernible zones of influence in both GA_3 and GA_3 + KN treated lots: Stage III or its combination stages being higher than Stage I or its combination stages. In the case of KN treatment, while there was linearity in grain number, the grain size showed minimal linearity in grain number, the grain size showed minimal linearity or near horizontality without marked formation of stage sensitive zones. It is evident that there is a direct correlation between grain size and numbers, the stage of hormonal treatment, and also the PGR in question.

Discussion

Leaf senescence in cereals occurs during the postanthesis period. It causes the canopy photosynthetic rate to decline rapidly from the maximum at anthesis (Wittenbach 1979, Patterson et al. 1980). This drop, at a time when the grain dry weight is still increasing, may present a limitation to the yield. If leaf senescence could be delayed at this juncture, then the photosynthetic activity of the canopy would be maintained for a longer duration. A PGR-induced delay in the initiation of flag leaf senescence at Stage III (Fig. 1) and the subsequent yield improvements conform to the above postulations. Stagewise differences

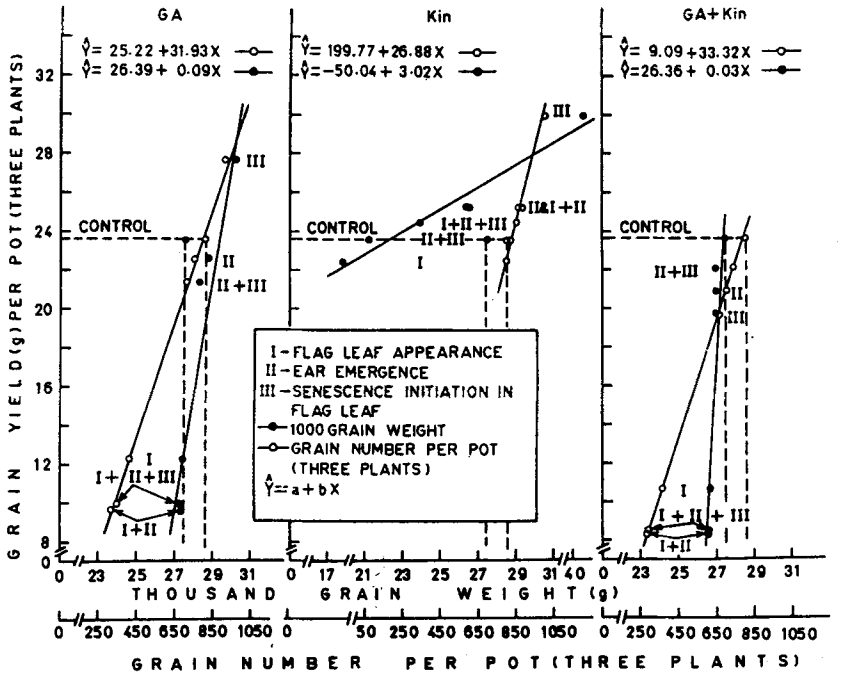

Fig. 4. Regression lines and equations giving degree and direction of dependence of grain yield on grain development (1,000 grain weight and grain number per plant). Treatments and treatment stages are as in Figs. 1, 2 and 3.

are well known due to changes in hormonal levels *in situ* (Corcooran and Phinney 1962, Tang et al. 1974) and the response to externally applied PGRs (Mishra and Gaur 1980, Mishra et al. 1981, 1982, 1983). Knowledge of the exact role of endogenously produced PGRs in regulating various yield components in cereals is very rudimentary (Evans 1975). The tillering habit in cereals is hormone-controlled and the endogenous hormone supply could become limiting (Langer et al. 1973); this can be supplemented exogenously, as in barley (Ruckenbauer and Kirby 1973) and wheat (Herzog 1975). Our findings are similar to theirs wherein PGR application at Stage I (Fig. 2) promoted tillering.

Gibberellins influence many processes in the growth and development of cereal crops. Maximum GA levels are found in young leaves and under high levels of nutrition (Evans 1975). The hastening at Stage I and delaying of senescence at Stage III by GA_3 (Fig. 1) are in keeping with the above observations. GAs are also known to be involved in the filament growth and anther dehiscence. In developing grain, gibberellin content reaches a peak during rapid grain filling period (Wheeler 1972). Leaf orientation affecting yield is well known (Duncan et al. 1967, Lendent and Renard 1982). This observation conforms with the Stage III response to GA_3 (Fig. 3). The advantageous effect of GA_3 at this stage might be due to an increase in leaf erectness (Kumar and Wareing 1972, Ziv et al. 1976). This is a trait closely associated with modern high yielding cultivars of many cereals (Evans 1975). Similarly, GA_3 and KN-induced delay in the onset of senescence at Stage III (Fig. 1) supports the

postulations of Tollenaar and Daynard (1978) regarding greater leaf longevity and yield ameliorations.

Very little is known about the role of cytokinins in cereals, compounds which are at a peak during anthesis and show a rapid decline thereafter (Wheeler 1972). An appreciable grain number response to KN at Stage II and III (Fig. 3) in our studies may be of relevance in this context. The beneficial effect of KN at Stage III might be due to promotion of the marginal tillers to give at least a few more ears bearing developed grains, resulting into more grain number and higher yield (Fig. 3). Besides, increased tiller number due to KN (Fig. 2) may modulate the plant to have reduced inclination of the leaves, a favorable character conducive of higher yield (Evans 1975). In contrast to our findings, Williams and Cartwright (1980) observed that benzylademine (BA) application at the preheading stage in barley increased yield by 57% due to enhanced growth of smaller shoots. They also reported that the increased grain yield of these smaller shoots was attributable to the increased weight of individual kernels, especially from the basal and distal part of the ear, and also maintained a greater uniformity of the kernel size within the ear. Increase in grain number by KN may be a reflection of more, otherwise unfilled, seeds countable due to increased transport of photosynthate to the ear during maturation (Evans 1975, Williams and Cartwright 1980, Gifford and Evans, 1981).

The antagonistic effect of GA_3 + KN combination is rather surprising and difficult to explain. This is indicative of a common site and for constituent processes involved in the action of these two hormones. It is worth noting that this suggestion is specific to growth stage at the time of treatment. However, this is opposed to the earlier observations made by Sabater et al (1981) in terms of differential effect of KN and GA_3 on the senescing barley leaf segments. This could be the reason for our observations in terms of dissimilar effects of combined treatment of GA_3 and KN compared to their individual treatment effect. It is known that both GA_3 and KN contents are higher at anthesis (Wheeler 1972), and hence may change into supraoptimal levels at Stage I due to GA_3 + KN application, and might have given deleterious effects. It is also possible that such an effect might have persisted even to the later stages. However, the antagonistic effect at Stage III seems to be different from this. Such effects might be caused through altered physiology of late-formed tillers, whose otherwise contributory role to the developed seed population is well supported from the present work. The antagonistic effect of GA_3 and KN to each other is known from the work on barley and other crops (Williams and Cartwright 1980, Sabater et al. 1981).

Hence, it is concluded that the stage-dependent beneficial effect of PGRs on the yield components may be attributed to the improved photosynthate availability as a result of postponement of flag leaf senescence.

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