Relation of Crop Maturity and Physiology to Air Pollution Incited Bronzing of Phaseolus vulgaris

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ABSTRACT

Bronzing symptoms of white beans (*Phaseolus vulgaris*) are necrotic stippling of the upper leaf surface followed, in 2-3 days, by chlorosis and then abscission of leaves. Natural fumigation does not always result in necrosis. The disease occurs late in the growing season. Within a relatively uniform crop, damage is often not uniform. Crop maturity (stage of development) regulates the time of symptom expression, and crop vigor its severity. Leaf area per plant begins decreasing at "full bloom", and bronzing occurs about 10 days later, or 59 days after seedling emergence in most plot areas and about 1 week later in others. Dry matter production, before symptoms, is negatively correlated with disease severity. It is suggested that carbohydrate depletion of leaves predisposes them to airborne-oxidant-incited necrosis, chlorosis, and defoliation. Phytopathology 60:407-410.

Bronzing of white beans (*Phaseolus vulgaris* L.) is incited by an atmospheric oxidant, probably ozone (14). The symptoms are typical of ozone injury on other crops (10); reddish-brown necrotic stippling of the upper leaf surface, general chlorosis, and premature abscission of leaves. In southwestern Ontario, the disease always occurs late in the growing season, about 10-14 days before normal rapid defoliation is expected.

During surveys of white bean crops with bronzing, large variations in disease severity were often found in small (3 ha) fields. Often, of two adjacent crops of the same cultivar, one was bronzed, the other not. The fumigation dose presumably was uniform within and between fields. They were located in various parts of southwestern Ontario, and were generally more than 100 km from the major industrial sources of airborne-oxidants.

The sensitivity of plants to air pollution damage is affected by many environmental and edaphic factors (7), but there were no obvious differences in the crops surveyed. A growth analysis experiment was used to establish the extent of variation in injury in a small, relatively uniform crop. Also, it was used to show the effect of crop physiology on the predisposition of *P. vulgaris* to bronzing.

Growth analysis was developed primarily to study the determinants of crop yield, and has been a valuable technique for elucidating the physiology of crops as opposed to individual plants (12). Samples of a growing crop are taken two or more times. Leaf area is measured and used as an estimate of potential photosynthetic assimilation. Dry wt of various plant parts (roots, stems, leaves, and pods) and the sum of these weights is measured to obtain estimates of actual assimilation. The analysis of leaf area, dry wt, the absolute and relative rate at which they increased or decreased, and the relationships between them, constitute growth analysis.

Wallace & Munger (11) explained yield differences between different types and varieties of *P. vulgaris*. They found that white or pea beans, the type used in the investigation reported here, had a high leaf area ratio (leaf area per unit stem dry wt) and relative leaf growth rate. These contributed to white beans having mature plant dry wt and seed yield higher than any other bean type.

Growth analysis has been used to study the effect of disease on growth and yield (13). The investigation reported here revealed its utility in examining the effect of crop physiology on predisposition of plants to a disease.

MATERIALS AND METHODS.—An apparently uniform portion of a field of Brady sandy loam at the Harrow Research Station was selected. *P. vulgaris* 'Sanilac', a type of dry bean, was planted in June 1967. There were 25 6×6 m plots in a square design consisting of five "rows" and five "columns" of plots. No "treatments" were applied, as the occurrence of bronzing in an apparently uniform crop was being studied.

Random samples of five plants were removed from each plot every 2 weeks. Leaf laminae were removed from the plants and leaf area (one side) determined with an optical planimeter (2). The entire plant above the cotyledon node was dried at 85-90 C for 2-3 days, and weighed.

For each sampling time, an analysis of variance of dry wt and leaf area between rows and columns of the square design was performed. Growth analysis was done as suggested by Radford (9). Using least square estimates of the parameters, two equations were obtained which gave a good fit with the actual data for the increase and decrease of dry wt (W) and leaf area (A) with time (t). The rate of dry matter accumulation has been called crop growth rate, CGRWB (weight basis), and the rate of leaf area increase and decrease CGRAB (area basis). CGR was obtained by evaluating the first derivative of the weight and area equations, i.e., $W = f(t)$; $CGRWB = dW/dt$; at any instant of time. Another quantity, net assimilation rate (NAR) was used as a particularly sensitive indicator of oxidant injury because it measured the rate of dry matter accumulation per unit leaf area per day. It was calculated by dividing CGRWB by A. In all cases, $t = 0$ was the date, 7 June, when 75% of the cotyledons had emerged from the soil.

Bronzing was measured by visually comparing the

TABLE 1. F values derived from analysis of variance in white bean growth and oxidant-incited bronzing among rows and columns of a square field plot design

Variable	Source	Days after emergence					
		19	35	49	62	76	92
Dry wt	Rows	1.67	<1	2.39	1.01	3.55[a]	1.08
	Columns	2.08	1.20	<1	<1	<1	<1
Leaf area	Rows	1.01	2.44	3.45[a]	3.50[a]	18.59[b]	
	Columns	2.50	1.43	<1	<1	1.22	
Bronzing rating	Rows				37.22[b]		
	Columns				<1		

[a] Probability of a larger $F < .05$; 16 and 4 degrees of freedom.
[b] Probability of a larger $F < .01$; 16 and 4 degrees of freedom.

plants in the 25 plots and rating them on a 1 (minimum) to 9 (maximum) scale (14). The ratings were made 4 days after bronzing symptoms were first noted.

RESULTS.—*Extent of variation in bronzing.*—Relatively high levels of airborne oxidants were recorded for short durations during most of the growing season (14), but typical bronzing symptoms were first observed on 5 August, 59 days after emergence. The upper leaf surfaces were flecked or stippled. There was considerable variation in the severity of the symptoms among the 25 plot areas. The growth of plants also varied before bronzing occurred. The bronzing rating, leaf area, and dry wt differed between rows of the field plots (Table 1). The mean bronzing rating for the rows, as they occurred from east to west in the field, was 3.7, 1.1, 7.3, 3.1, and 4.1 ± .37. They were ordered 1-5 in increasing rank of bronzing severity, but the rating of rows 2-4 was not significantly different. As will be shown below, the growth of these plants was also equal, and the response of plants in rows 2 and 4 are omitted from some of the data reported.

Plant leaves in row 5 (most bronzing) had many very small necrotic spots on the upper surface. Chlorosis and rapid defoliation began 2 to 3 days after necrosis. There was some bronzing on the pods, but they did not drop. Plants in rows 2, 3, and 4 had a lower bronzing rating, as their leaves were only moderately stippled. These leaves, however, also became chlorotic and dropped prematurely. In row 1, the least affected row, the bronzing symptoms did not appear until almost 1 week after injury was found in the other plants. When the symptoms appeared, there was relatively little necrotic stippling, and the leaves were already becoming chlorotic as a consequence of normal senescence.

Relation between bronzing and crop physiology.—On the 62nd day after emergence, the dry wt and leaf area of the plants ranked in reverse order of the bronzing rating (Fig. 1-A, B).

To interpolate between sampling dates, and to determine CGR and NAR, it was necessary to find an equation which described the dry wt and leaf area growth. The equation, fitted by least squares, was $\log_e W$ (or A) $= b_0 + b_1 t + b_2 t^2$ where b_i are the regression coefficients. Using these coefficients, the calculated values for dry wt and leaf area were obtained (Fig. 2-A, B). The multiple correlation coefficient for the regression was always high, and proved that the equations used to describe the plant growth were suitable (Table 2).

The rate of dry matter accumulation, CGRWB, differed between rows during the entire growing season (Fig. 3-B). Again, the highest growth rate and the lowest amount of bronzing occurred in row 1. Row 3 had a lower CGRWB and row 5 a still smaller one.

CGRAB was an excellent measure of crop maturity or stage of development. The initiation of flowering coincided with the maximum CGRAB and full bloom with CGRAB = 0. Most plants began flowering be-

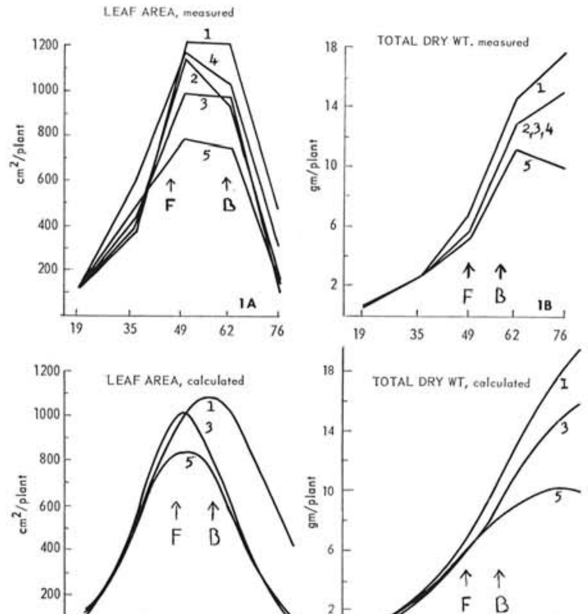

Fig. 1, 2. Growth of white beans before and after occurrence of air pollution incited bronzing. Symptom severity rating: 1 (slight)-5 (severe). F = full bloom; B = first bronzing observed. **1-A)** Measured leaf area (one side) on days indicated. (S.E. on successive days = 8.09, 50.2, 95.0, 89.6, 34.0). **1-B)** Measured dry wt (excluding roots) on days indicated. (S.E. on successive days = 0.0600, 0.259, 0.492, 1.28, 1.50). **2-A)** Leaf area (one side) calculated from the growth function $\log_e Y = b_0 + b_1 t + b_2 t^2$; t = days from emergence. **2-B)** Dry wt (excluding roots) calculated from the same growth function.

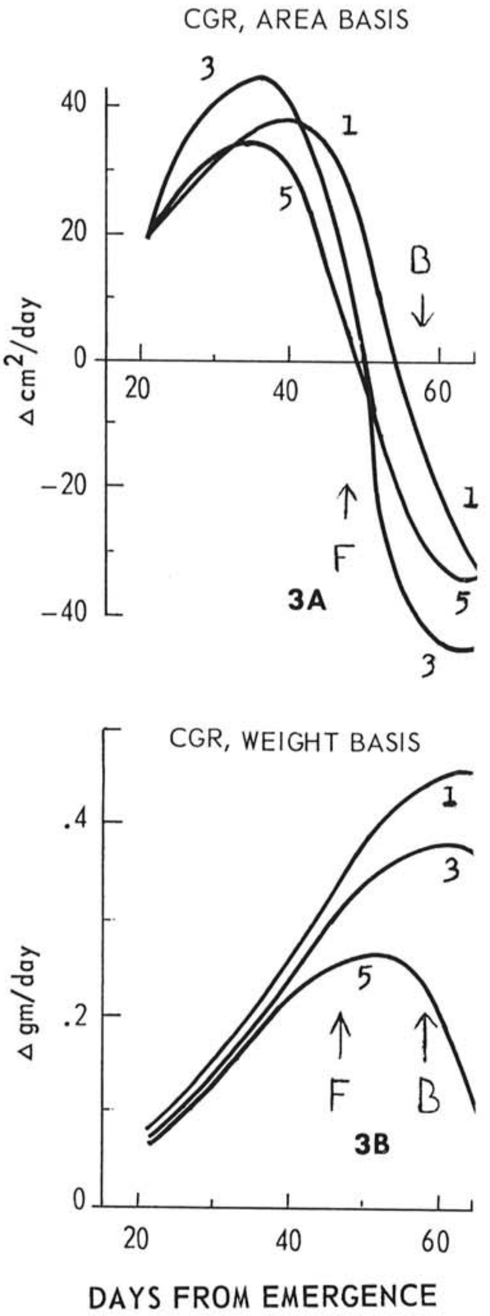

Fig. 3. Crop growth rate (CGR) of white beans before and after occurrence of air pollution incited bronzing. Symptom severity rating: 1 (slight)-5 (severe). F = full bloom; B = first bronzing observed. **A)** Leaf area basis. **B)** Dry wt basis.

tween day 40 and 45, just when the maximum CGRAB was attained. They reached full bloom about day 48-49, when CGRAB equaled zero. That was the time when leaf area loss began to exceed the production of new leaf area. The plants in row 1 were an exception because, although they flowered and reached their maximum CGRAB at the same time as those in rows 3 and 5, full bloom occurred 5 or 6 days later (Fig. 3-A). However the CGRAB again equaled zero at full bloom.

The plants in row 1 not only had the least bronzing and attained the highest leaf area and dry wt, but their increase in dry wt per unit area of leaf surface was highest during the 4 weeks prior to day 50 (Fig. 4). The NAR of all plants decreased during the first weeks of the growing season, but those that exhibited the necrotic and chlorotic symptoms had a larger decrease in NAR.

Discussion.—Correlating the occurrence of bronzing with the growth analysis of the white bean crop shows that the rate of growth influences the severity of the disorder, and the stage of growth regulates the time or dose at which bronzing can be incited. In addition, the experiment confirmed observations made during farm surveys; major variation in bronzing severity occurs in small, apparently uniform fields.

Maturity was based upon the time of first flowers and full bloom. Maximum CGRAB coincided with the beginning of flowering; at full bloom, CGRAB equaled zero. Bronzing always appeared about 10-11 days after CGRAB = 0.

Crop physiology was related to the severity of injury. Comparisons were made between plants maturing uniformly. The dry wt, leaf area, CGRAB, and CGRWB were all lowest for the injured plants. This observation does not support the thesis that "conditions . . . optimum for plant growth, generally will also be optimum for maximum sensitivity of a plant" (3). Heggestad (8) noted that lower soil moisture tensions will usually be associated with increased sensitivity to air pollution damage. The Brady sandy loam on which the plants were grown is only moderately well drained. Possibly the most severely injured plants were subjected to the lowest average moisture tensions. Such tensions may not have been conducive for optimum growth of white beans. *P. vulgaris* is extremely sensitive to nonoptimum soil moisture conditions (1).

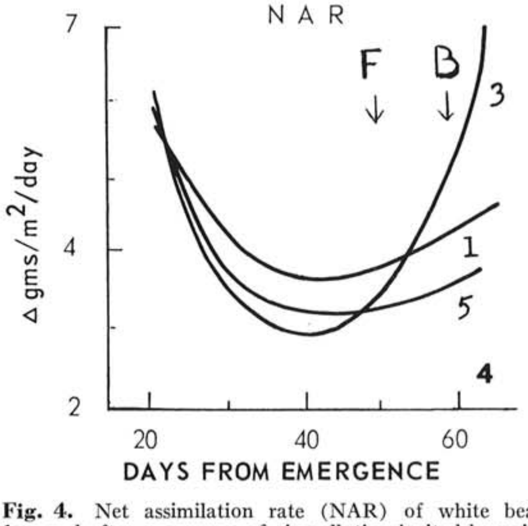

Fig. 4. Net assimilation rate (NAR) of white beans before and after occurrence of air pollution incited bronzing. Symptom severity rating: 1 (slight)-5 (severe). F = full bloom; B = first bronzing observed.

TABLE 2. Multiple correlation coefficients for the regression[a] on growth of white beans through five time periods

Variable	Row in square field plot design				
	1	2	3	4	5
Dry wt	0.985	0.986	0.973	0.977	0.989
Leaf area	0.920	0.944	0.936	0.942	0.969

[a] Dependent variable: $\log_e$ (dry wt) or $\log_e$ (leaf area). Independent variables: days after emergence and (days after emergency)^2.

The observed effects of crop maturity and physiology on the sensitivity of the bean crop may reflect the influence of carbohydrates on the predisposition of *P. vulgaris* to ozone-incited injury (4). When leaves are kept in darkness more than 48 hr, they become very sensitive. The carbohydrate content of the leaves decreases from 11 to below 3.5 mg/g fresh wt. Low carbohydrate levels in leaves of a field-grown plant would be expected shortly after full bloom when pods are developing. Flower and fruit development initiates a redistribution of carbohydrates from the vegetative to the reproductive organs (5, 6). Bronzing occurred shortly before normal senescence would be observed. Chlorosis followed the necrotic symptom so rapidly that senescence of the leaves probably was proceeding before bronzing was seen.

The relative values of NAR suggest that there was little or no airborne-oxidant damage prior to necrosis. Although NAR decreased between emergence and CGRAB = 0, the decrease was not larger than expected (11). Bronzing occurred when CGRAB < 0. Then, unfortunately, NAR becomes a complicated function of growth and leaf abscission, and is not an index of photosynthetic efficiency.

There are some implications from the results which should be considered when a disease control program is established. Control of air pollution injury has focused on chemicals that will remove oxidants from the air around the plants, and on breeding for varieties that have stomatal apparatus sensitive to increased pollutant concentrations. Field testing for control will be extremely difficult with white beans, because major variations in disease incidence can occur in an apparently uniform area. The variability between chemicals or varieties being tested is likely to be small when compared with the variability within chemicals or varieties. Field tests will be especially difficult in a breeding program, because within-line variability is already high.

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