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Time of pod set and seed position on the plant contribute to variation in quality of seeds within soybean seed lots

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Abstract

Time to pod set and seed position on the plant were studied as possible within-plant components contributing to variation in quality of seeds within seed lots of soybean (*Glycine max* (L.) Merrill) cultivar IAS-5. Plants were grown at 28/22 °C (NT = normal temperatures) or 33/27 °C from the beginning of pod set onwards (HT = high after normal temperatures). The heaviest seeds were produced on positions in the canopy where also the earliest pods were formed: main stems versus branches and upper versus lower main stem sections. The variation in weight between seeds within a seed lot was mainly determined by position in the canopy, contributing 23 and 30% under NT- and HT-conditions, respectively. Days to pod set contributed 2 and 27% respectively. Position fully accounted for variation resulting from days to pod set under NT- but not under HT-conditions. Seeds from earlier pods had a lower viability. For explaining differences in viability between seeds within a seed lot, days to pod set of individual seeds was more important than seed position. Components of variation between plants were largely additional to within-plant components and at least of equal importance.

Keywords: *Glycine max* (L.) Merrill, seed production, seed quality, image analysis, viability, seed weight, seed age, seed position, within-plant variation, between-plant variation.

Introduction

Considerable variation in seed quality is regularly observed within and between soybean seed lots (e.g. Hampton, 1995). The variation between seed lots can largely be ascribed to variations in crop and seed management, soil and weather conditions and the quality of the seed sown (e.g. Coolbear, 1995; Dornbos Jr., 1995). The variation within a seed lot has to arise mainly from variations between and within mother plants in the crop producing the seeds. Partly these again will be caused by variation in the above mentioned factors (e.g. differences in light, seed quality, local differences in soil conditions) within a crop.

Seed position on the plant is one of the components of within-plant variation that may account for part of the variation in physical (such as weight, shape) or physiological (such as viability, vigour) seed attributes. Soybean seeds from the upper plant strata are generally heavier than those from lower strata, but relations between position and physiological quality attributes are less clear. Moreira de Carvalho & Ferreira, quoted by Hampton *et al.* (1996) concluded that seeds from the lower third of the plant compared to seeds from the medium or upper third tended to have a lower 100-seed weight, inferior germination, and higher electrical conductivity after incubation in water, indicating a lower vigour. Ramseur *et al.* (1984) reported that top main stem seeds were heavier than seeds from bottom main stem or branches and performed better in conductivity and germination tests. Adam *et al.* (1989) found that seeds from the top of the plants were heavier than seeds from the bottom and showed faster seedling growth and a higher standard germination, but there were generally no differences in conductivity. Keigley & Mullen (1986) observed heavier seeds and more vigorous seedlings from the earliest main stem pods, but found no differences in germination.

It is likely that the position of the pod affects seed quality attributes both directly and through its association with time to pod set. Greater seed weights might result from a longer period of seed filling and/or higher rate of seed filling (e.g. Egli *et al.*, 1978; Egli & Wardlaw, 1980). Differences in seed maturity or in time left for seed ageing on the plant may explain differences in physiological seed attributes. Keigley & Mullen (1986) already suggested that seed maturity is an important factor explaining seed position effects but little is known about the relations between the duration of pod growth, development, maturation and ageing and physiological seed quality attributes for individual seeds. Also, external conditions around the pod (e.g. temperature, relative humidity, light) and internal characteristics (e.g. local sink-source relations) may contribute to position effects on physiological seed attributes.

We hypothesize that both the variation in duration of individual pod growth, development, maturation and ageing until harvest and the position of the pod on the plant significantly contribute to the total variation in physical and physiological quality attributes of seeds harvested from a soybean crop.

The main aims of this study were (1) to examine the relationships between time of pod set, seed position on the plants and physical (weight, projected area, shape) or physiological (viability and length of normal seedlings) quality attributes of individual seeds harvested from soybean crops grown in two production environments, and (2) to examine to what extent the components of within-plant variation accounted for the variation in physical or physiological seed quality attributes in the seed lots harvested.

Materials and methods

Crop growing conditions

Two crops were grown from the same seed lot of cv. IAS-5 (determinate growth habit; harvested and processed in the State of São Paulo, Brazil) from end of May until beginning of October 1993 in controlled glasshouses of the Department of Agronomy of the Wageningen Agricultural University, The Netherlands. The crops were grown under 13 hours at 28 °C with supplementary light (Philips SON-T 400W lamps) during the day, and 11 hours at 22 °C with total darkness during the night. Sixty days after sowing, when the first pod was set on more than 80% of the plants (stage R3 according to Fehr *et al.*, 1971) one of the glasshouses had the temperature adjusted to 33 °C during the day and 27 °C during the night. This sequence of normal and high temperatures will be called HT-conditions, whereas the continuously normal temperature will be indicated as NT-conditions.

Plant cultivation

Plastic pots with 30 cm diameter and 30 cm depth containing potting soil with pH 5.6 were arranged in 10 rows of 18 pots per glasshouse. The soil was previously sieved in order to provide optimal seed bed homogeneity. Seeds were planted at a depth of 4.5 cm and spaced 5 cm from each other in a row that followed the same direction as the row of pots. Distance between plant rows was an alternation of 45 cm and 75 cm, thus achieving 33.33 plants m^{-2}. Seeds were inoculated with *Bradyrhizobium japonicum* at sowing. Plants were watered with tap water when needed to provide adequate soil moisture. Liquid fertilizer was provided two weeks after sowing, based on 60–40–50 kg/ha of N, P and K, respectively, plus Ca, Mg and micronutrients. Plants were physically supported to avoid lodging. No visible symptoms of diseases were observed in either crop.

Measurements

Twenty neighbouring plants in the centre of each crop were observed, but three plants in the crop under NT-conditions appeared not to have produced any seed. Neighbouring plants were chosen to reduce variation in external conditions between plants and because individual plants are difficult to distinguish and handle within a crop. The date of pod set was assessed by tagging daily all pods that were 5 mm long. This criterion proved more functional than date of flower set because of the natural overproduction of flowers in soybean (Gai *et al.*, 1984). More than 70% of the pods were coded. For part of the analysis, seeds from these pods were later classified into five categories of time to pod set. Crop harvest maturity (HM) (Fehr *et al.*, 1971) was achieved 3283 °C days (130 days) after sowing for the crop under NT-conditions (base temperature 0 °C), and 3935 °C days (140 days) for the crop under HT-conditions. All plants, pods and seeds were individually harvested by hand 101 °C days after HM under NT-conditions and 61 °C days after HM under HT-conditions. From all harvested seeds the position on plants was noted. The seeds were later classified in four ways according to their position (Figure 1).

Harvested seeds were kept in a chamber with 40–45% relative humidity for a few days in order to adjust their moisture content to approximately 10%. All seeds were individually weighed and measured by image analysis for the following features as

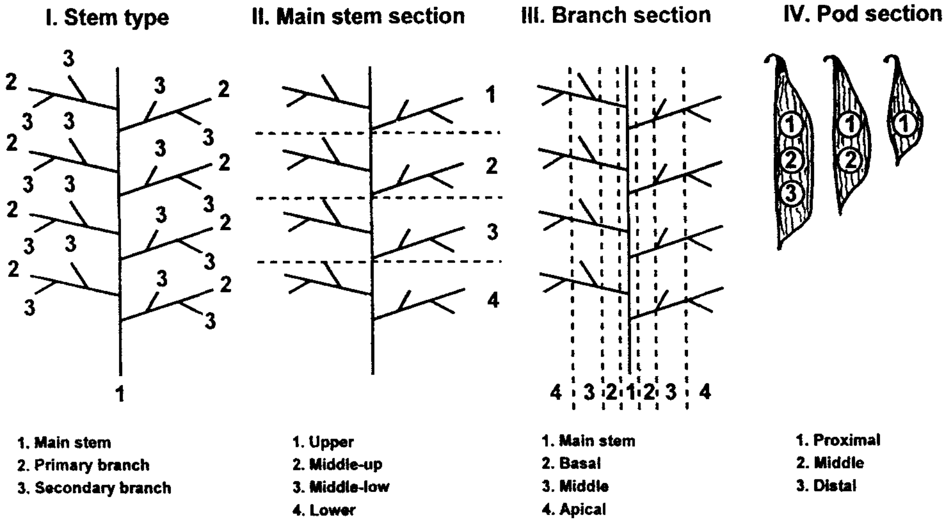

Figure 1. Seed positions on the plant used for seed classifications. For main stem section: Upper = uppermost node – node 11; Middle-up = nodes 10–8 ; Middle-low = nodes 7–5; Lower = nodes 4–1. For branch section: Basal = node 2; Middle = node 3–5 under NT-conditions and 3–4 under HT-conditions; Apical = node 6–10 under NT-conditions and 5–6 under HT-conditions. Position of seeds from pods containing a single seed was considered proximal, and position from pods containing two seeds was proximal or middle.

described by Illipronti Jr. *et al.* (1997):
Area: Derived from the total number of pixels of the projected area of the seed.
Eccentricity: The length (major axis of an ellipse fitted to the contour of the seed) divided by the width (minor axis of an ellipse fitted to the contour of the seed). This was calculated because selecting seeds by image analysis using shape attributes was suggested by Misra *et al.* (1989) to be an easy way of improving the quality of seed lots.
Thereafter the seeds were stored at 5 °C until assessment of viability and seedling growth.

To evaluate the viability and seedling growth, all seeds were individually placed between folds of pleated filter paper sheets code 3014 (Schleicher and Schuell, Dassel, Germany). Twenty-five seeds were placed per sheet 2 cm from the top edge, and orientated with the embryonic axis away from the paper and the micropyle towards the top of the sheet. After placing the seeds, the pleated filter paper was covered with an additional sheet of filter paper and placed in a plastic germination box of 21 x 15 x 3 cm (l x w x h). Fifty ml of tap water was sprayed per box. Piles of 6 boxes with seeds plus an empty one on the top to avoid moisture loss were incubated at an angle of about 45° in a seed germinator at 25 °C, approximately 96% relative humidity, and 12 hours of day length. The seedling length (total length of hypocotyl + root) was recorded after 96 hours of incubation. After 125 hours of incubation, the seeds were classified into three viability classes ('normal seedlings', 'abnormal seedlings' and 'ungerminated seeds'), according to the rules of the International

Seed Testing Association (Anonymus, 1996). Only the length of seedlings classified as 'normal seedlings' was further considered.

Experimental analysis

Statistical tests were applied as described by Snedecor & Cochran (1989). For comparisons between class means, analysis of variance was used. Because of the different numbers of observations between classes, Least Significant Differences were calculated for every comparison between two means. The dependence between seed position on the plant or pod set class and seed viability was evaluated by chi-square goodness of fit test on numbers. For presentation purposes, the figures show percentages (instead of numbers). Regression models were used to evaluate the contribution of multiple components of within- and between-plant variation to the total variance in physical seed attributes and length of normal seedlings. The variance accounted for was the adjusted R^2 (1-(residual m.s.)/(total m.s.)), expressed as percentage. Seed position categories and mother plants were included in the regression models as qualitative explanatory variates, days to pod set as a quantitative explanatory variate.

The number of observations was different for different analyses because (i) seed position was recorded for all seeds, (ii) pod set was only recorded for the seeds from 70% of the pods, (iii) not all seeds produced normal seedlings, and (iv) the number of seeds differed between glasshouses.

Results

Crop structure

More seeds were harvested from primary branches than from the main stem (Figure 2). Secondary branches contributed little to seed production, especially under HT-conditions. Middle main stem sections were more important for seed production than upper and lower main stem sections (Figure 2), especially under HT-conditions. Primary branches from the lowest two nodes only contributed substantially to seed production under NT-conditions. Higher nodes contained more main stem seeds, lower nodes more seeds from branches (Figure 2), and more seeds were produced on the basal branch sections than on those further away from the main stem ($P < 0.05$; Tables 1, 2). The number of seeds per pod varied between one and three, but under both conditions more than 70% of the pods contained two seeds. The distribution of pods over pods with one, two or three seeds did not differ between the two conditions (not shown) and also the resulting distribution of seeds over pod sections did not differ (Table 2). Proximal seeds were most abundant (53%). Distal seeds only contributed 5%.

Relations between seed position on the plant and time to pod set

Pod set occurred between 51 and 84 days after sowing (Figure 3). Pod set was earlier for seeds produced on the main stem than for seeds produced on the branches (Fig-

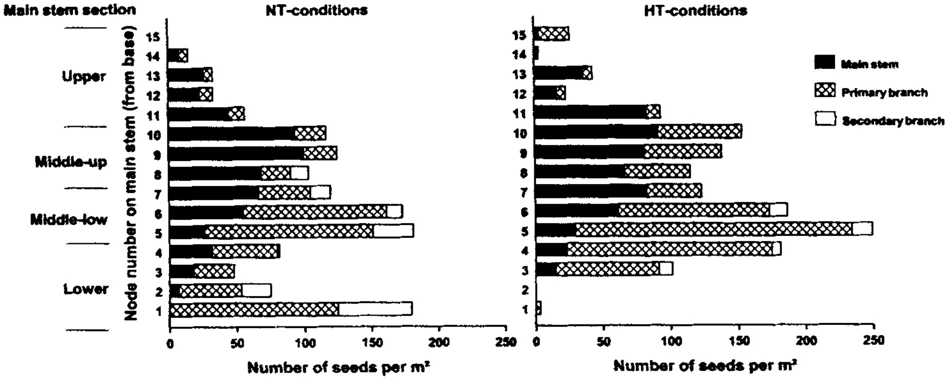

Figure 2. Number of seeds per m^2 classified according to their position on main stem nodes and stem types, for seeds produced under NT- and HT-conditions. NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing. Chi-square analysis showed that seeds at different main stem sections were not equally distributed over stem types (NT: $\chi^2 = 393.07^{**}$, HT: $\chi^2 = 192.76^{**}$).

ure 3, Table 1) and also earlier for seeds produced in the upper main stem sections than for seeds produced in the lower (Table 1). These differences were more pronounced under HT- than under NT-conditions. Seeds from the basal branch sections

Table 1. Average number of days from sowing to pod set for seeds produced at different positions on plants grown under NT[1]- and HT-conditions.

Seed position classification	NT-conditions		HT-conditions	
	No of seeds	Days from sowing to pod set	No of seeds	Days from sowing to pod set
I. Stem type				
Main stem	221	60.4 b[2]	236	61.9 b
Primary branch	293	63.7 a	356	66.4 a
Secondary branch	59	64.2 a	18	68.3 a
II. Main stem section				
Upper	138	59.9 b	97	58.7 c
Middle-up	191	59.3 b	127	59.5 c
Middle-low	136	64.6 a	251	67.2 b
Lower	108	64.7 a	135	69.3 a
III. Branch section				
Main stem	221	60.4 b	236	61.9 c
Basal	154	63.4 a	208	65.1 b
Middle	147	64.1 a	126	68.6 a
Apical	51	64.3 a	40	66.8 ab
Total seed lot	573	62.5	610	64.7

[1] NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing.

[2] Means followed by the same letter within a column, separately for the three classifications, are not statistically different ($P \geq 0.05$) according to the LSD test.

had earlier pod set than seeds from the middle branch section under HT-conditions (Table 1). Time to pod set did not differ significantly for seeds classified into different pod sections (data not shown).

Relations between seed position on the plant and physical seed attributes

Weight per seed ranged between 45 and 312 mg and was higher under NT-conditions than under HT-conditions (Table 2). Seeds from the main stem were heavier and larger than seeds on branches (Table 2). Primary branches in turn yielded heavier and larger seeds than secondary branches. Seed eccentricity values ranged between 1.01 and 1.76 and were slightly higher under HT-conditions than under NT-conditions. Seed eccentricity was not different for the three stem types under NT-conditions, but under HT-conditions seeds on main stems and primary branches were more eccentric than those on secondary branches.

From the upper to the lower stem sections, seed weight and size decreased (Table 2). Seeds were found least eccentric in the middle parts of the plants under NT-conditions and in the upper parts of the plants under HT-conditions.

Table 2. Average physical attributes of seeds from different positions on the plants grown under NT[1]- and HT-conditions.

Seed position classification	NT-conditions				HT-conditions			
	No of seeds	Weight (mg)	Area (mm^2)	Eccentricity	No of seeds	Weight (mg)	Area (mm^2)	Eccentricity
I. Stem type								
Main stem	342	216.6 a[2]	40.0 a	1.22 a	358	188.7 a	37.4 a	1.29 a
Primary branch	375	181.0 b	35.7 b	1.21 a	481	161.3 b	33.8 b	1.28 a
Secondary branch	89	159.8 c	32.9 c	1.22 a	27	144.1 c	30.6 c	1.24 b
II. Main stem section								
Upper	153	224.5 a	40.8 a	1.23 a	114	204.8 a	39.2 a	1.25 b
Middle-up	209	203.1 b	38.1 b	1.21 b	244	185.7 b	36.9 b	1.30 a
Middle-low	262	180.2 c	35.7 c	1.21 b	336	162.7 c	34.0 c	1.28 a
Lower	182	176.9 c	35.5 c	1.23 a	172	149.4 d	32.4 d	1.29 a
III. Branch section								
Main stem	342	216.6 a	40.0 a	1.22 b	358	188.7 a	37.4 a	1.29 a
Basal	195	178.0 bc	35.0 c	1.21 b	295	167.7 b	34.7 b	1.28 a
Middle	201	171.7 c	34.4 c	1.20 b	157	149.4 c	32.0 c	1.28 a
Apical	68	189.5 b	37.9 b	1.25 a	56	152.8 c	32.5 c	1.24 b
IV. Pod section								
Proximal	435	187.2 b	36.3 b	1.22 a	450	168.6 b	34.7 bc	1.30 a
Middle	340	200.2 a	38.2 a	1.21 a	371	175.0 a	35.7 a	1.27 b
Distal	31	216.0 a	39.9 a	1.19 a	45	182.4 a	35.7 ac	1.22 c
Total seed lot	806	193.8	37.2	1.22	866	172.1	35.2	1.29

[1] NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing.

[2] Means followed by the same letter within a column, separately for stem type, main stem, branch and pod sections, are not statistically different ($P < 0.05$) according to the LSD test.

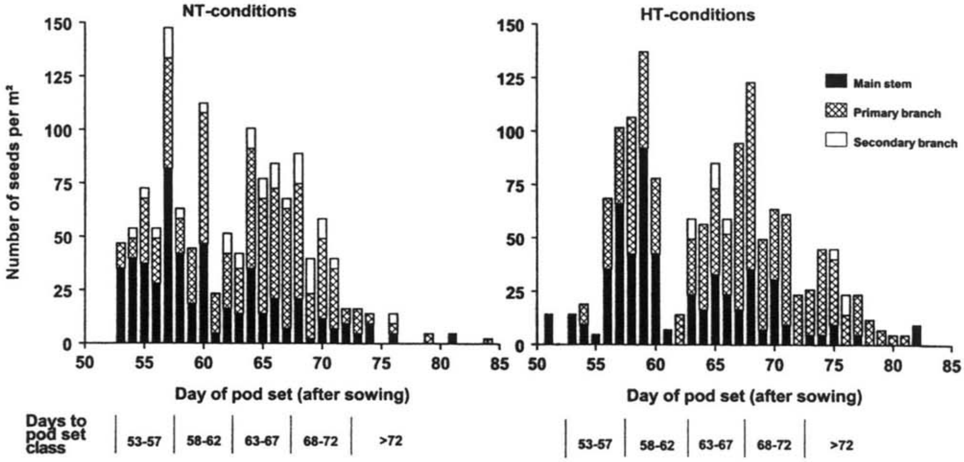

Figure 3. Number of seeds per m^2 classified according to their day of pod set and stem type, for seeds produced under NT- and HT-conditions. NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing. Chi-square analysis showed that for each stem type, pods from seeds produced under different conditions were set at different days (χ^2 = 90.89**, 107.58** and 37.11** for seeds on main stems, primary and secondary branches respectively).

Under NT-conditions, seeds in the apical section of the branches were heavier and larger than seeds from basal and middle sections (Table 2). Under HT-conditions, seeds were heaviest in the basal sections. In the NT-crop the seeds from apical branch sections were most eccentric, in the HT-crop those from basal and middle branch sections.

Seeds produced in the proximal section of the pods were lighter and smaller than seeds produced in the middle or distal sections (Table 2). This was also found when positions were compared within pods containing two or three seeds (results not shown). Eccentricity values decreased from the proximal to the distal sections of the pods only for plants grown under HT-conditions.

Relations between seed position on the plant and seed viability or length of normal seedlings

For the whole seed lot, the distribution of seeds classified into the three viability classes was similar for both temperature conditions. Significant chi-square values indicated that under NT-conditions, viability was higher for seeds produced on the branches than for seeds produced on the main stem (Figure 4A). Under these conditions, seed viability also increased from the basal to the apical branch sections (Figure 4E), whereas it was not different for seeds from different main stem sections (Figure 4C) or different pod sections (not shown). For the seeds produced under HT-conditions, viability was not significantly related to any of the seed position categories (Figure 4B, D, F).

The length of normal seedlings ranged between 25 and 132 mm and was consider-

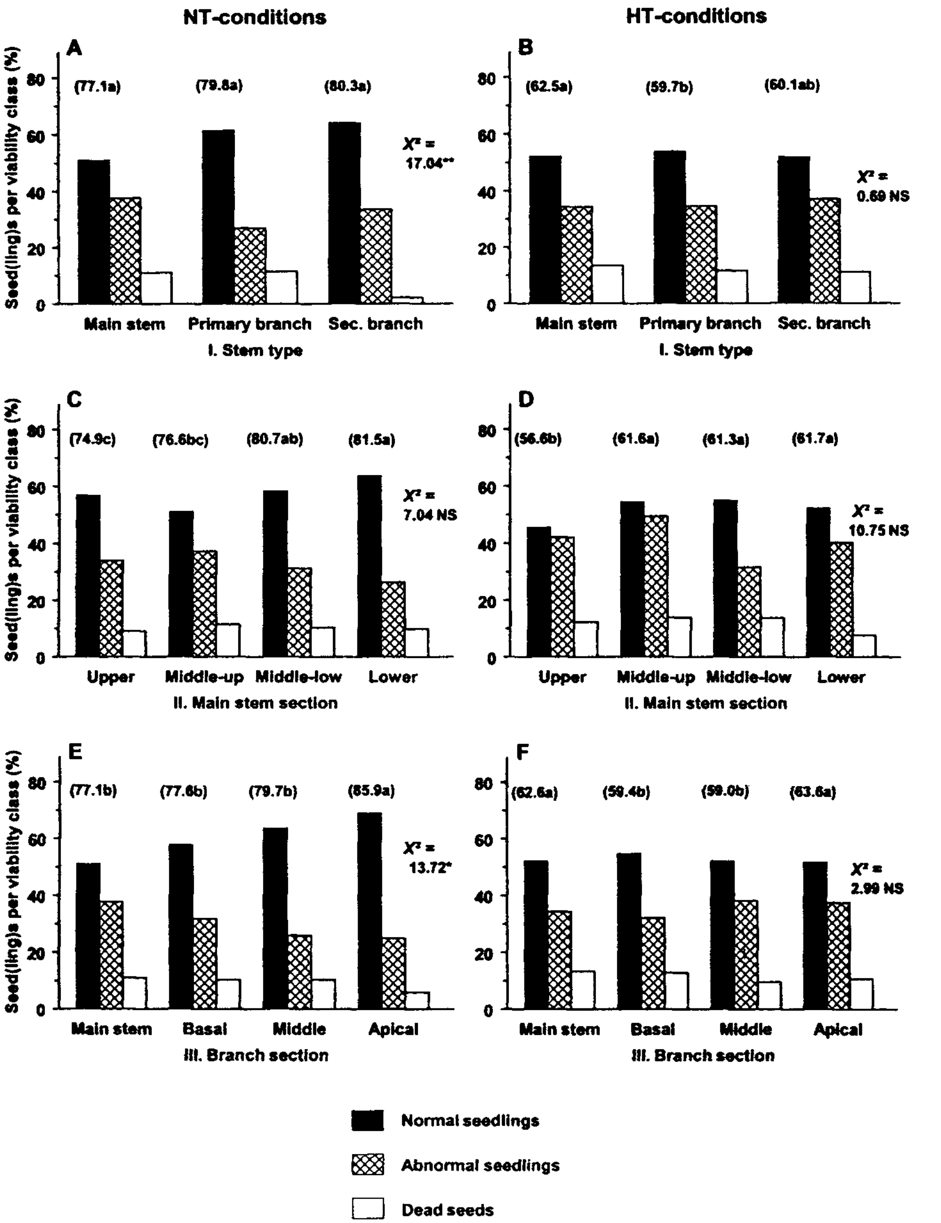

Figure 4. Distribution of seed(ling)s over viability classes and length (mm) of normal seedlings (between brackets), for seeds produced at different positions on plants grown under NT- and HT conditions. NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing. χ^2 = chi-square; NS, * and ** indicate non-significant and significantly different distibutions at $0.01 \leq P < 0.05$ and $P < 0.01$, respectively. Lengths of normal seedlings marked by the same letter do not differ significantly ($P \geq 0.05$) according to the LSD test. For seed numbers see Table 2.

Table 3. Average physical attributes of seeds produced under NT[1]- or HT-conditions classified according to time to pod set.

Days to pod set class	NT-conditions				HT-conditions			
	No of seeds	Weight (mg)	Area (mm^2)	Eccentricity	No of seeds	Weight (mg)	Area (mm^2)	Eccentricity
53–57	160	198.3 a[2]	37.2 a	1.22 a	94	204.4 a	38.6 a	1.23 c
58–62	126	198.4 a	37.8 a	1.23 a	145	190.0 b	37.9 a	1.30 a
63–67	159	193.7 a	37.3 a	1.22 a	150	171.9 c	35.1 b	1.26 bc
68–72	104	176.1 b	35.4 b	1.21 a	136	151.8 d	32.5 c	1.28 ab
< 73	24	171.3 b	34.3 b	1.19 a	85	144.7 d	31.9 c	1.31 a

[1] NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing.
[2] Means followed by the same letter within a column are not statistically different ($P \geq 0.05$) according to the LSD test.

ably higher for seeds produced under NT-conditions than for those produced under HT-conditions. For NT-conditions, the length of normal seedlings was not different for different stem types (Figure 4A), but under HT-conditions main stem seeds produced longer normal seedlings than seeds from primary branches (Figure 4B). Normal seedlings from seeds harvested in the lower main stem sections of the plants were longer than normal seedlings from seeds harvested in the upper section under both conditions (Figure 4C, D). Longer normal seedlings arose from seeds produced in the apical section of the branches than from those in lower branch sections (Figure 4E, F). Seed position within the pods did not significantly affect the length of normal seedlings (data not shown).

Relations between time to pod set and physical or physiological seed attributes

Under both temperature conditions, seeds from earlier pods were heavier and larger than those from later pods, with the effect being strongest under HT-conditions (Table 3). Differences in eccentricity were not significant between seeds from distinct pod set classes under NT-conditions, but seeds from late pods were more eccentric than seeds from the earliest pods under HT-conditions.

Viability increased with later date of pod set under both conditions (Figure 5). The longest normal seedlings originated from pods of intermediate age under NT-conditions and seeds from the latest pods under HT-conditions (Figure 5).

Contribution of components of variation within and between plants to the variance in physical and physiological seed quality attributes in the seed lots produced

Days to pod set accounted for 2 and 27% of the variance in seed weight in the seed lots produced under NT- and HT-conditions respectively (Table 4). When the individual within-plant components were compared for their contribution to the variance in weight and size in the seed lot produced, stem type and branch section were the most

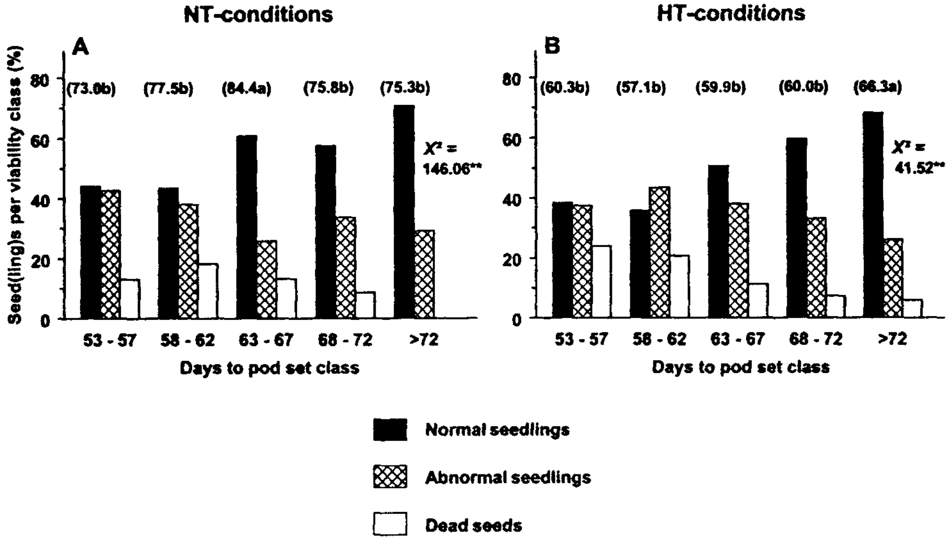

Figure 5. Distribution of seed(ling)s over viability classes and length (mm) of normal seedlings (between brackets), for seeds from pods grown under NT- and HT conditions and classified according to the day of pod set. NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing. X^2 = chi-square; ** indicates significantly different distibutions at $P < 0.01$. Lengths of normal seedlings marked by the same letter do not differ significantly ($P \geq 0.05$) according to the LSD test. For seed numbers see Table 3.

contributing components under NT-conditions, days to pod set and main stem section under HT-conditions (Table 4). Pod section had the lowest relevance under both conditions.

All seed position characteristics combined were able to account for 23 and 30% of the variance in weight under NT- and HT-conditions respectively (Table 4). For projected area, these values were somewhat lower. When days to pod set was combined with this model, the percentage of variance in size and weight accounted for was significantly improved under HT-conditions, but not under NT-conditions. This shows that position fully accounted for the variance resulting from days to pod set under NT-conditions. Also under HT-conditions, effects were not completely additional.

To get an impression of the variation due to the different plants within the crop, the plant number of the mother plant the seeds originated from was added as a qualitative variate in the regression analysis (Between-plant components; Table 4). Between-plant components often were as important in accounting for the variance in size or weight attributes as the best within-plant component, especially under HT-conditions. Fairly large percentages of variance in seed size and weight attributes were accounted for when within- and between-plant components were combined. For instance, 65.4% of the variance in seed weight under HT-conditions was accounted for by this model.

Only small or non-significant percentages of variance in eccentricity were accounted for by the components of variation within plants (Table 4). Between-plant

Table 4. Percentage of variance in physical attributes of seeds and in length of normal seedlings produced from them accounted for by components of plant variation in crops grown under NT[1]- and HT-conditions during pod growth development, maturation and ageing. Data from 300 seeds under NT-conditions and 305 seeds under HT-conditions for seedling length and from 573 and 610 seeds respectively for other attributes.

Components of plant variation	NT-conditions				HT-conditions			
	Weight	Area	Eccentricity	Length of normal seedlings	Weight	Area	Eccentricity	Length of normal seedlings
1. Days to pod set	2.3 ***[2]	0.8 *	– NS	3.8 **[3]	27.2 ***	22.1 ***	1.0 **	4.8 ***[3]
2. Stem type	18.1 ***	14.4 ***	– NS	0.7 NS	16.8 ***	14.4 ***	– NS	– NS
3. Main stem section	13.3 ***	9.0 ***	0.3 NS	2.7 *	25.5 ***	20.9 ***	0.8 NS	2.7 *
4. Branch section	17.2 ***	14.3 ***	1.2 *	3.8 **	19.4 ***	16.9 ***	0.7 NS	– NS
5. Pod section	2.3 ***	2.6 ***	0.4 NS	– NS	0.4 NS	0.1 NS	4.4 ***	0.1 NS
6. Seed position on the plants 2+3+4+5	23.4***	19.7 ***	1.7 *	3.4 *	30.0 ***	25.2 ***	5.9 ***	2.5 NS
7. Within-plant components 1+2+3+4+5	23.3 ***	19.7 ***	1.7 *	4.9 **[3]	35.7 ***	29.6 ***	6.2 ***	6.4 **[3]
8. Between-plant components	16.2 ***	15.1 ***	8.6 ***	9.7 ***	36.5 ***	34.0 ***	9.2 ***	25.8 ***
9. Within- and between-plant components 1+2+3+4+5+8	41.5 ***	37.2 ***	9.5 ***	12.5 ***[3]	65.4 ***	56.4 ***	12.6 ***	28.4 ***[3]

[1] NT = normal temperatures (28/22 °C), HT = high temperatures (33/27 °C) during pod growth, development, maturation and ageing.

[2] NS, *, ** and *** indicate statistically non-significant, significant at $0.01 \leq P < 0.05$, $0.001 \leq P < 0.01$ and $P < 0.001$, respectively; – indicates that the residual variance exceeded the variance of the variate.

[3] For accounting for variance in length of normal seedlings, days to pod set was included in the model as a linear plus a quadratic term.

components accounted for greater percentages of variance in eccentricity than within-plant components. Combined they could account for about 10%.

Also for length of normal seedlings, the percentages of variance accounted for by components of within-plant variation were lower than those for seed weight and size (Table 4). Days to pod set was the within-plant factor accounting most. Again, between-plant components were more important, especially under HT-conditions.

Discussion

Weight and size attributes

The observations that the heavier and larger seeds in a seed lot originate from main stems, upper main stem sections and more distal positions in a pod (Table 2) are in accordance with results from Ramseur *et al.* (1984) and Smiciklas *et al.* (1992) on stem type, Ramseur *et al.* (1984) on main stem section and Egli *et al.* (1978) on position in the pod. Because of mutual associations among the four seed position categories (e.g. relatively more seeds from branches on lower main stem sections, Figure 2) the contribution of the individual position categories to the total variation in seed weight and size was not completely additional (Table 4). Combined they accounted for 23–30% of the variance in seed weight.

The heaviest and largest seeds were generally found on those positions on the plants which also produced the earliest pods, i.e. main stems versus branches and upper versus lower main stem sections (Tables 1 and 2). Multiple regression (Table 4) showed that under NT-conditions, seed position was more important than days to pod set and even fully accounted for the variation resulting from time of pod set.

From our data it is not possible to distinguish between seed growth rate and seed filling duration as causes for differences in seed weight within plants. The larger weight and size of seeds in the upper main stem or apical branch sections (Table 2) might be partly attributable to a higher light interception by the upper leaves and pods (e.g. Sakamoto & Shaw, 1967). The large weight and size of main stem seeds might be associated directly with significant amounts of assimilates translocated downwards in the plants during the pod filling period (Blomquist & Kust, 1971) and indirectly with main stem seeds being more frequent in higher main stem sections (Figure 2). However, also for individual main stem sections, seeds on the main stem generally were heavier than seeds on branches (results not shown). Also the seed filling period could have been shorter for seeds from later pods (cf. Gbikpi & Crookston, 1981; Spaeth & Sinclair, 1984). Seeds being lighter in the proximal section of pods than in the middle or distal section (Table 2), were shown by Egli *et al.* (1978) to result from a lower seed growth rate.

However, under HT-conditions both seed position on the plants and time of pod set appeared to contribute considerably to the variance in seed weight and size, although effects were not completely additional (Table 4). This implies that later pod set reduced seed weight on all positions. Especially seeds from later pods produced under HT-conditions were lighter and smaller than seeds produced under NT-conditions (Table 5). Since the HT-crop took longer to reach harvest maturity, this probably resulted from a lower individual seed growth rate under HT-conditions rather than a shorter duration of growth. Although seed filling periods generally are shorter when temperatures are increased to the HT-conditions of our experiment (Egli & Wardlaw, 1980), there was a delay of 10 days in harvest maturity of the HT-crop compared to the NT-crop while pod set on average was only 2 days later (Table 1). This is in accordance with data from Hesketh *et al.* (1973), but it remains unknown if this resulted from a longer seed filling period under HT-conditions or from an extremely prolonged period of maturation drying. Also, under HT-conditions, flowering, pod and seed set for the later pods took place under warmer external conditions than for the earlier pods. A higher temperature during flowering was shown by Egli & Wardlaw (1980) to reduce seed growth rate. The different external conditions probably could have changed the number of cotyledon cells initiated in the seeds (cf. Gbikpi & Crookston, 1981) and consequently their growth rate or ultimate size.

Viability and length of normal seedlings

Seed performance in seed quality tests is not consistently related to seed position categories on the plant (e.g. Ramseur *et al.*, 1984; Keigley & Mullen, 1986; Adam *et* *al.*, 1989; Smiciklas *et al.*, 1992). This research shows that for explaining differences in viability and length of normal seedlings in the seeds produced, time of pod set was more important than seed position (Figures 4, 5; Table 4). Because time of pod set was different for seeds produced on different positions (Table 1), apparent effects of positions act partly through time of pod set. Effects of position on seed viability were only detected under normal conditions for stem type and branch section (Figure 4A, E) and in those cases they were consistent with pod set sequence on these positions (Table 1). There also was an increase in the average length of normal seedlings from seeds produced from the upper to the lower main stem sections (Figure 4C, D), which also was the sequence followed by the set of individual pods (Table 1). The higher viability of seeds from later pods under both conditions (Figure 5) suggests that seeds from earlier pods were already deteriorating before harvest. This deterioration is commonly observed on a crop level when harvest is delayed longer after seeds have reached physiological maturity (e.g. TeKrony *et al.*, 1980; Dornbos Jr., 1995). Because in our experiment most seeds tested were produced from pods set at intermediate dates (Figure 3), an earlier harvest might have improved the viability of the total seed lot harvested.

The viability of seeds produced under NT-conditions mostly was higher than that of seeds produced under HT-conditions (Figure 5; Gibson & Mullen, 1996), especially for seeds from early pods which were exposed to ageing conditions longer. Because more seeds were produced relatively late under HT-conditions compared to NT-conditions (Figure 3) and later seeds had a higher viability (Figure 5), the overall viability of the seed lots produced did not differ for the two conditions. The lengths of normal seedlings were shorter for seeds produced under HT-conditions than for seeds produced under NT-conditions, suggesting a lower vigour even at comparable viability levels.

Relationship between physical and physiological seed attributes

The literature about relationships between seed size or weight and physiological quality of soybean seeds is inconsistent (e.g. Hoy & Gamble, 1985; Adam *et al.*, 1989; Smiciklas *et al.*, 1992; Dornbos Jr. & Mullen, 1991; Sung, 1992; Illipronti Jr. *et al.*, 1999). The present results show that – within seed lots – relations between seed size and viability partly act through seed age, and are likely to be affected by the harvest moment and production condition. In our experiment, the heaviest seeds were produced in the earliest pods (Table 3). These had the lowest viability (Figure 5). Earlier harvesting might have lead to a relatively better performance of the earlier produced, larger sized seeds. The relation between seed size and viability will also differ between production conditions, because the importance of pod set date in explaining the variance in seed weight differed between production conditions (Table 4). Earlier research (Illipronti *et al.*, 1997, 1999) already showed that relationships between seed size and performance were masked by associations between seed size and other undesirable traits like cracking and wrinkling.

Selecting only properly shaped seeds has been considered as an easy way to improve the quality of seed lots (Misra *et al.*, 1989). There were no indications in our research that categories with lower eccentricity (Tables 2, 3) were associated with a lower viability (Figures 4, 5).

Contribution of within- and between-plant variation in explaining variance in quality attributes

When individual seeds are analysed, components of variation can only be studied in a multiple regression analysis when the quality attributes of the seeds studied are of a quantitative nature – like size or weight. Viability, which is of more relevance to seed production, is a qualitative variate. Using quantitative physiological seed attributes, like electrical conductivity per area of seed, probably could improve the knowledge in this field.

All components of within-plant variation together (combined positions plus days to pod set) explained 23 and 36% of the variance in size and weight attributes in the seed lots produced under NT- and HT-conditions respectively (Table 4). Between-plant variation accounted for similar parts of the variance. Effects of within- and between-plant variation were additional or at least additional to a major extent (Table 4). This implies that seed size was regulated by day of pod set and seed position in comparable ways in the individual plants, but that plants varied in the average level. Which components of variation between plants determine this level is yet unknown. Because neighbouring plants were analysed to minimize variation in environmental conditions within the crop, variation between plants must have mainly arisen from the differences in quality, growth and development of the seeds sown and the resulting variation in competition within the crop.

Within- and between-plant variation together accounted for > 40% and > 60% of the variance in individual seed weight under NT- and HT-conditions respectively, and for lower percentages in eccentricity and length of normal seedlings. This leaves a part of the variance still unexplained. A better division of within plant-components and interaction between within- and between-plant components may improve the results further. Also inclusion of the moment of physiological maturity of the individual seeds to mark the period of seed filling may increase the understanding of the sources of differences in weight and size. This moment will also be important for explaining differences in seed viability or vigour, because ageing starts from this moment onwards.

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