THE EFFECT OF FRUIT LOAD ON POD AND SEED CHARACTERISTICS OF OKRA (Abelmoschus esculentus L.)

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Abstract. A study to determine the effects of fruit load on the pod and seed characteristics of okra was carried out. Four cultivars of okra ('Boyiatiou', 'Veloudo', 'Clemson' and 'Pylaias'), very common in Greece and the Mediterranean Basin, were cultivated under field conditions and three levels of fruit loading $[L_1, L_2]$ and L_3 (10, 15 and all fruits plant⁻¹ respectively)] were applied on plants. Leaving more fruit on the plant (L₃) caused lower seed germination in cv. 'Boyiatiou' and 'Veloudo', where in cv. 'Pylaias' and 'Clemson' the opposite effect was observed. Highest germination was recorded for plants with the lowest fruit load (L1). The most seedhardness and least seed germination was observed in cv. 'Boyiatiou', but storage of seeds for 18 months increased germination by up to 16.2%. Flower induction, fruit set and fruit dimensions were unaffected by fruit load, irrespective of cultivar. Seed number per fruit was not affected by fruit load, but seed size (mean 100 seed weight) tended to decrease with fruit load in cv. 'Boyiatiou' and 'Pylaias'. There was also no consistent effect of fruit load on plant height. In conclussion, pod load is not considered to affect okra seed quality and especially for crops intended for seed production growers should not apply pod thinning since no effect in seed quality was observed.

Keywords: Abelmoschus esculentus L.; fruit load; okra; pod set; seedhardness.

INTRODUCTION

It has long been known that in okra plants, flowers and fruits develop singly at each node after about the 5th to 8th leaf. Additionally, the presence of a developing fruit at one node tends to inhibit flowering and fruit growth at next node [3]. This means that for the fresh commodity, if fruits are not harvested regularly at an immature stage (e.g. every 3-4 days), production will occur in waves and yield will be reduced. However, so far no experiments have been carried out in order to evaluate the effect of fruit load on seed quality, especially seed germination and occurrence of hardseedness.

In the seed crop, fruits grow to maturity on the plant and thus the effect of fruit load on seed fruit production is greater than in the fresh fruit crop. The developing fruits act as sinks that influence photoassimilate distribution and plant growth. The manipulation of these sinks by harvesting immature fruits for fresh consumption constantly alters the balance between supply and demand for assimilates within the plant and in turn influences physiological processes such as photosynthesis, growth, and development [3]. Regular harvesting seems to increase the activity of the photoassimilate source (leaves), delays leaf senescence and increases photassimilation. In parallel, the total number of pods formed increases [3, 16, 18], although the magnitude of this increase varies between cultivars [4] and plant species [2, 8, 9, 17, 20, 22, 23]. Picking of green fruits encourages vegetative growth, fruit characteristics and ultimately seed quality in okra [11, 21, 24]. Moniruzzaman and Quamruzzaman [12] cultivated okra cv. BARI Dheros-1 was cultivated for seed and fresh pod production simultaneously on the same plant and found that seed yield was highest when 4-6 green fruit were harvested from the plants prior to leaving pods to mature for seed production.

So far no experiments have been carried out in order to evaluate the effect of fruit load on seed quality, especially seed germination and occurrence of hardseedness, in okra. Therefore, in the present experiment, the effect of three levels of fruit loading on okra plants in relation to pod and seed quality characteristics was studied in four genotypes in order to evaluate its effect when plants are cultivated for seed production.

MATERIAL AND METHODS

Plant material and experimental conditions

The experiment was carried out at the experimental farm of the University of Thessaly throughout the growing season of 2011. The sand, silt and clay content of the experimental site were 48, 29 and 23%, respectively. The pH was between 7.7-8.1 and the percent organic matter was 1.3. Seeds of four okra cultivars (Abelmoschus esculentus [L.] Moench) were sown in seeding trays on 20 May and retained under controlled temperature (20°C) within the nursery until field transplantation. Plants were transplanted at the stage of 3-4 true leaves, on 10 June.

The experimental treatments were as follows:

a) Four cultivars of okra, cv. 'Clemson', 'Boyatiou', 'Veloudo' and 'Pylaias'.

b) Three loading levels: L_1 = Leaving 10 fruit on the plant and removing all successive fruits; L₂= leaving 15 fruits on the plant and removing all successive fruits; L_3 = control (leaving all the fruits on the plant).

The crop was irrigated regularly on a weekly basis with a drip irrigation system, whereas for weed control Fusilade herbicide was applied by spraying when the plants were 20 cm in high (fluazifop-P-butil, 800 $ml/1000 m^2$).

First anthesis was recorded 33 days after transplantation (DAT) and from then on each flower was recorded and tagged on a daily basis, noting the date of flowering, position on the plant etc. The

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distance between two rows was 2 m and the distance between two plants within each row was 30 cm. The total area was 200 m^2 .

Measurements

Seed germination was measured using 400 seeds per treatment, with four replications of 100 seeds each [10]. Seeds were surface sterilized with mancozeb fungicide prior to the test, then placed on a double layer of Whatman No.1 filter paper moistened with distilled water in sterilized Petri dishes. The Petri dishes were placed in an incubator at 25°C for 21 days and every 2 days germinated seeds were counted and removed; in case of moisture deficiency distilled water was added. The criterion for germination was the emergence of the radicle outside seed coat [25, 1]. Hard seeds were considered to be those s that had not germinated and remained rigid and free of visible contamination at the end of 21 days, according to the ISTA description of hard seed characteristics (2014). Throughout the experiment the following data were recorded:

- Seed germination
- Total flower induction per plant
- Number of pod set per plant
- Pod length
- Pod diameter
- Average number of seeds per pod
- 100 seed weight
- Plant height

Statistical analysis

The experiment was laid out in a Randomized Complete Block design with factorial arrangement using three replications (n=3), with each block being consisting of 12 plots of 9 m² (3×3 m), in total 36 plots. Statistical analysis was carried out with the aid of the S.A.S. statistical package (SAS Institute Inc., USA) and means were compared by Duncan's Multiple Range Test (DMRT) and the Least Significant Difference (LSD) test at p<0.05. Graphs were generated using Microsoft Excel Office software (Microsoft Corporation, USA).

RESULTS

Climatic conditions during the cultivation period

The climatic conditions of the experimental site are given in Fig. 1. The temperature gradually increased from May to July and then decreased till October. There was a difference of about 12-16 °C between minimum and maximum temperatures (in mean overall differences) throughout the experimental period. There was no rainfall during July, but during the other months varied from 23 to 43 mm. Relative humidity thus decreased during July, but afterwards increased progressively. This means that as the number of days from anthesis increased (from 30-50 DAA) pods were increasingly exposed to lower temperatures and higher humidity.



Figure 1. Climatic conditions of the experimental site during the cultivation period (vertical arrows indicate the start and end point of harvest period). (The black horizontal arrow shows the harvest time of L_1 and L_2 and the red horizontal arrow shows the harvest time of L_3)

Flower induction, pod set and the number of pods per plant retained until maturity

Flower induction was significantly higher in 'Pylaias' 'Clemson', and 'Veloudo' than in 'Boyiatiou'. In all cultivars the percent pod set was high (approximately 80.0-90.0%), but tended to be lower in 'Boyiatiou' than in the other cultivars. This meant that the total number of pods set per plant was significantly lower in 'Boyiatiou' (mean value 9.9 pods plant¹) than in the other cultivars (mean values ranging from 16.7-22.3 pods plant⁻¹). Because of this, the number of pods in treatment L_2 of 'Boyiatiou'(6 pods plant⁻¹⁾ failed to reach the target number (15) and was less even than in treatment L_1 (10 pods). Differences in pod set also led to variation in the number of pods plant⁻¹ in treatment L₃ (all pods retained), and was significantly higher than L₂ in 'Clemson' marginally higher in 'Veloudo', but virtually the same in 'Pylaias' (Table 1).

Effect of fruit load on mean pod length and diameter

Mean pod length was highest in cv. 'Veloudo' (21.6 cm) and lowest in cv. 'Clemson' (16.8 cm). There was no effect of fruit load on mean pod length in any cultivar, except 'Veloudo' where pod length was higher in treatment L_1 (10 fruit per plant) than in L_2 (15 fruit per plant), but not in relation to L_3 (all fruits) (Table 1). Similarly, pod diameter was not affected by fruit load, except in 'Veloudo' where pod diameter was highest in L_1 and lowest in L_2 (Table 2).

Effect of fruit load on mean number of seeds/pod and mean 100-seed weight

No significant difference was observed between fruit load and the number of seeds per pod in any of the

cultivars. Overall, the number of seeds per pod in 'Clemson' and 'Pylaias' was higher than in 'Boyiatiou' and 'Veloudo' but statistically significant differences were observed only between plants with 10 or 15 pods $(L_1 \text{ and } L_2)$ (Table 3).

Seed size (mean 100-seed weight) was significantly higher in plants with only 10 pods (L_1) than in the control (L_3) in 'Boyiatiou' and 'Pylaias' as well as in plants with 15 pods ('Boyiatiou' and 'Veloudo'). No effect of pod number per plant on seed size was observed in cv. 'Clemson' (Table 3).

Cultivar (C)	Fruit load level (L)	Total flower induction (flowers/plant)	Pod set (pods/plant)	Percent pod set (%)	Pods retained until maturity/plant
'Boyiatiou'	L ₁	13.6 ^a	11.3ª	83.1	10.0
	L ₂	8.6 ^a	6.6 ^a	76.7	6.0
	L ₃	15.0 ^a	12.0 ^a	80.0	12.0
Mean		12.4	9.9	79.9	9.3
LSD		7.3	6.5	-	-
'Veloudo'	L ₁	15.6 ^b	14.0 ^a	89.7	10.0
	L ₂	21.0 ^a	18.3 ^a	87.1	15.0
	L ₃	20.3 ^{ab}	18.0 ^a	88.7	18.0
Mean		18.9	16.7	88.5	14.3
LSD		4.9	6.2	-	-
'Pylaias'	L ₁	22.6 ^a	21.3ª	94.2	10
	L ₂	23.6ª	19.6 ^{ab}	83.1	15
	L ₃	16.6 ^a	14.0 ^b	84.3	14
Mean		20.9	18.3	87.2	13.0
LSD		8.5	6.9	-	-
'Clemson'	L ₁	23.0 ^a	21.0 ^a	91.3	10
	L ₂	20.0 ^a	17.6 ^a	88.0	15
	L ₃	31.3 ^a	28.3 ^a	93.6	28
Mean		24.7	22.3	90.6	17.6
LSD		11.4	11.5	-	-
C×L					
(C×	L ₁)	*	*	-	-
$(C \times L_2)$		*	*	-	-
$(C \times L_3)$		*	*	-	-

Table 1. The effect of fruit load on total flower induction and pod set, percentage pod set and the number of pods per plant retained until maturity

Mean separation by LSD test. Mean values in the same column followed by different letters differ significantly at P = 0.05; *: statistically significant (p<0.05); ns: not significant; L_1 = Leaving 10 fruit on the plant and remove all other successive fruits; L_2 = leaving 15 fruits on the plant and remove all other successive fruits; L_3 = control (leave all the fruit on the plant).

Table 2. The effect of fruit load on pod dimensions at see	d maturity stage
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Cultivar	Fruit load level	Mean pod length	Mean pod diameter
(C)	(L)	(cm)	(cm)
	L_1	19.6 ^a	2.1 ^a
'Boyiatiou'	L ₂	18.8 ^a	1.8 ^a
	L ₃	19.4 ^a	2.0 ^a
Mean		19.2	1.9
LSD		2.2	0.3
	L ₁	23.0ª	2.0ª
'Veloudo'	L_2	20.0 ^b	1.7 ^c
	L ₃	21.9 ^{ab}	1.9 ^b
Mean		21.6	1.8
LSD		2.3	0.09
	L ₁	18.6 ^a	1.8ª
'Pylaias'	L ₂	21.5 ^a	1.8 ^a
	L ₃	21.7 ^a	1.7 ^a
Mean		20.6	1.7
LSD		4.4	0.2
	L ₁	15.7 ^a	1.1 ^a
'Clemson'	L ₂	17.2 ^a	1.2 ^a
	L_3	17.5 ^a	1.2ª
Mean		16.8	1.1
LSD		2.5	0.1
C×L			
(C>	<l<sub>1)</l<sub>	*	*
(C>	(L ₂)	*	*
(C>	<l<sub>3)</l<sub>	*	*

Mean separation by LSD test. Mean values for each cultivar separately in the same column followed by different letters differ significantly at P = 0.05; *: statistically significant (p<0.05); ns: not significant; L_1 = Leaving 10 fruit on the plant and removing all other successive fruits; L_2 = leaving 15 fruits on the plant and removing all other successive fruits; L_3 = control (leaving all the fruit on the plant).

Effect of fruit load on mean plant height

The effect of fruit load on plant height was variable and showed no consistent trend. For example, in 'Veloudo' restricting the number of pods per plant (L_1 and L_2) appeared to reduce plant height in comparison with the control (L_3). In 'Clemson' and 'Pylaias' plant height was lower in treatment L_2 (15 pods per plant) than in either L_1 (10 pods per plant) or L_3 (28 pods plant⁻¹ in 'Clemson', but only 14 pods plant⁻¹ in 'Pylaias'). In contrast, in 'Boyiatiou' no difference in plant height occurred between any of the treatments (Table 3).

Cultivar	Fruit load level	Mean number of	Mean 100 seed	Plant height
(C)	(L)	seeds/pod	weight (g)	(cm)
	L ₁	74.3 ^a	5.4 ^a	118.0 ^a
'Boyiatiou'	L ₂	63.3 ^a	4.7 ^b	117.0 ^a
	L ₃	67.3 ^a	4.8 ^b	128.0 ^a
Mean		68.3	4.9	121.0
LSD		20.9	0.3	16.3
	L ₁	65.0 ^a	6.0 ^a	132.0 ^b
'Veloudo'	L ₂	89.3 ^a	5.4 ^b	120.0 ^b
	L ₃	63.3 ^a	6.0 ^a	148.0 ^a
Mean		72.5	5.7	133.3
LSD		36.7	0.9	13.9
	L ₁	79.6 ^a	6.1ª	134.0 ^a
'Pylaias'	L ₂	83.6 ^a	5.7 ^{ab}	103.0 ^b
	L ₃	90.3 ^a	5.0 ^b	122.0 ^a
Mean		84.5	5.6	119.6
LSD		33.1	1.4	15.5
	L ₁	107.6 ^a	5.3 ^a	88.0 ^a
'Clemson'	L ₂	95.3 ^a	5.2ª	72.0 ^b
	L_3	91.6 ^a	5.5 ^a	90.0 ^a
Mean		98.1	5.3	83.3
LSD		36.1	0.5	13.2
C×L				
$(C \times L_1)$		*	*	*
(C×L ₂)		*	*	*
(C×L ₃)		ns	*	*

Mean separation by LSD test. Mean values for each cultivar separately Mean values for each cultivar separately in the same column followed by different letters differ significantly at P = 0.05; *: statistically significant (p<0.05); ns: not significant; L_1 = Leaving 10 fruit on the plant and removing all other successive fruits; L_2 = leaving 15 fruits on the plant and removing all other successive fruits; L_3 = control (leaving all the fruit on the plant).

Table 4. The effect of fruit load on seed	moisture content and seed germination
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Cultivar (C)	Fruit load level (L)	Mean moisture content at harvest (%)	Mean germination (%)	Seed hardness (%)
	L1	13.7 ^a	73.6 ^a	16.3
'Boyiatiou'	L ₂	11.4 ^a	69.0 ^b	23.6
	L ₃	11.0 ^a	61.0 ^b	27.3
Mean		12.0	67.8	22.4
LSD		3.9	2.4	
	L1	14.5 ^a	82.6 ^a	9.6
'Veloudo'	L ₂	15.3 ^a	75.6 ^b	13.0
	L ₃	14.0 ^a	71.0 ^c	16.3
Mean		14.6	76.4	12.9
LSD		4.6	2.7	
	L1	16.2 ^a	88.6 ^b	6.6
'Pylaias'	L ₂	16.3ª	94.6 ^a	3.0
	L ₃	15.4 ^a	89.6 ^b	5.3
Mean		15.9	90.9	4.9
LSD		3.7	3.0	
	L1	15.5 ^a	92.6 ^b	3.3
'Clemson'	L ₂	15.3 ^a	96.6 ^a	2.0
	L ₃	15.5 ^a	95.0 ^{ab}	3.0
Mean		15.4	94.7	2.7
LSD		2.8	2.7	-
C×L				
$(C \times L_1)$		ns	*	-
$(C \times L_2)$		*	*	-
(C×L ₃)		*	*	-

Mean separation by LSD test. Mean values for each cultivar separately in the same column followed by different letters differ significantly at P = 0.05; *: statistically significant (p<0.05); ns: not significant; L_1 = Leaving 10 fruit on the plant and removing all other successive fruits; L_2 = leaving 15 fruits on the plant and removing all other successive fruits; L_3 = control (leaving all the fruit on the plant).

Effect of fruit load on mean pod moisture content at harvest, percent germination and the incidence of hard seeds

The number of pods per plant did not affect the seed moisture content at harvest, irrespective of cultivar (Table 4). Seed moisture content tended to be higher in 'Pylaias' and 'Clemson' (mean = >15%) than in 'Boyiatiou' (12.0%) and 'Veloudo' (14.6%), but statistically significant differences between cultivars were recorded only in treatments L_2 and L_3 .

Seed germination was also higher in Pylaias' and 'Clemson' (mean = >90.0%) than in 'Boyiatiou' (68.0%) and 'Veloudo' (76.0%) (Table 4). The lower germination of 'Boyiatiou' and 'Veloudo' resulted from the higher incidence of hard seeds (22% and 13%, respectively) compared to that in the other cultivars (<5.0%). The percent seed germination of cv. 'Boyiatiou' was statistically lower than that of 'Clemson' and 'Pylaias' in all treatments (Table 4), indicating that both germination and the incidence of hard seeds is determined primarily by the genotype and not by the number of pods per plant. This conclusion is supported by the correlation coefficients presented in Table 5. Similarly, the rate of germination (mean germination time) was lower in Boyiatiou than in the other cultivars (not shown).

 Table 5. Correlations between cultivar, fruit load and seed germination

	Germination	Fruit Load	Cultivar
Germination	-	0.1848	0.9179
-	-	(36)	(36)
-	-	0.2806	0.0000
Fruit Load	-0.1848	-	0.0000
-	(36)	-	(36)
-	0.2806 ^{ns}	-	1.0000
Cultivar	0.9179*	0.0000	-
-	(36)	(36)	-
-	0.0000	1.0000	-

ns: not significant, *: Significant at P<0.05.

Effect of seed storage on seed germination

Seed storage for 18 months at room temperature improved germination in all cultivars, but the percent improvement differed, e.g. in 'Veloudo' seed germination increased by 20.9 % after storage but in 'Clemson' by only 4.6 % (Fig. 2).



Figure 2. Improvement of okra seed germination by storing the seeds for 18 months

DISCUSSION

Although the percent pod set was high (approximately 80.0-90.0%) in all four cultivars, flower induction was significantly higher in 'Clemson', 'Pylaias' and 'Veloudo' than in 'Boyiatiou'. Consequently, the number of pods per plant was low in 'Boviatiou' and did not correspond to the programmed fruit load in L_2 (6 pods instead of 15) and was low in L₃. Differences in flower induction also led to variation in the number of pods plant⁻¹ in treatment L₃ (all pods retained), which was significantly higher than L₂ in 'Clemson', only marginally higher in 'Veloudo', and virtually the same in 'Pylaias' (Table 1). These deviations from the programmed fruit loads therefore have to be taken into account when interpreting the results.

In the crop for fresh consumption, it is well known that frequent harvesting increases pod formation and yield [16, 3, 18, 19]. However, in the seed crop, retention of the pod on the plant until maturation reduces pod set, as was the case especially in 'Boyiatiou'. It might be expected, therefore, that pod size would be affected by the number of pods maturing on the plant since each pod acts as a sink for nutrients [6]. In reality, however, pod length and diameter, are genetically determined traits [3]; they were therefore not affected by fruit load. An apparent exception to this was the higher pod length of cv. 'Veloudo' in treatment L_1 (10 pods plant⁻¹) compared with L_2 (15 pods plant⁻¹), but not in relation to L_3 (18 pods plant⁻¹). Pod length in okra has been reported to decrease with higher pod load, due to an increase in water stress [5, 6]. In addition, in another experiment it was reported that fruit load affected yield which was highest when 12 fruits per plant were reatained, whereas pod weight and pod dimensions wer highest when six fruits per plant were retained [15]. In the present experiment there was no indication of water stress during cultivation, which may explain the absence of an effect of pod load on pod length. Similarly, pod diameter was highest in L_1 . but lowest in L_2 and not L_3 (Table 2). It can therefore be surmised that even in this instance, the differences in pod dimensions were unlikely to have resulted from the fruit load on the plant, but were due to a possible lack of homogeneity in the plant population or perhaps to a difference in sampling time. Finally, although pods were permitted to set after the desired number for each treatment had been attained (these pods were immediately removed once set), the number of these additional pods per plant (Table 1) depended on the cultivar and was not related to fruit load.

Although overall the number of seeds per pod in 'Clemson' and 'Pylaias' (mean value 98 and 84, respectively) was higher than in 'Boyiatiou' and 'Veloudo' (mean value 68 and 72, respectively), this characteristic was not affected by the pod load. This result differed from those reported from Moniruzzaman and Quamruzzaman [12] who found that when the first-formed fruit were harvested from the plant in a crop grown for both fresh production and seed, the

number of seeds formed per fruit subsequently was higher, wheras Nabi et al. [13] reported that the number of seeds per fruit and seed yield in total was higher when the 12 fruit per plant were retained. Similarly, differences in mean 100 seed weight in each cultivar could not generally be related to pod load. For example, mean seed weight of 'Boyiatiou' was higher in L_1 (10 pods plant⁻¹) than in L_3 (12 pods plant⁻¹), but also higher than in L_2 (only 6 pods plant⁻¹). In 'Veloudo' mean 100 seed weight was higher in L_1 and L_3 (10 and 18 pods plant⁻¹, respectively) than in L_2 (15 pods plant⁻¹). In 'Pylaias' mean 1000 seed weight was higher in L_1 (10 pods plant⁻¹) than in L_3 , but not L_2 (15 and 14 pods plant⁻¹), whereas in 'Clemson' no differences were observed in 100 seed weight even though in L_1 there was only a third of the number of pods per plant in relation to L_3 (10 and 28 pods plant⁻¹, respectively). However, Nabi et al. [13] reported that 100 seed was higher when six fruit per plant were retained.

The effect of fruit load on plant height was also variable and showed no consistent trend (Table 3). In contrast, Nabi et al. [15] reported that plant height was highest when 12 fruits per plant were retained. Moreover, the number of pods per plant did not affect the seed moisture content at harvest, irrespective of cultivar (Table 4). Seed moisture content tended to be higher in 'Pylaias' and 'Clemson' (mean = >15.0%) than in 'Boyiatiou' (12.0%) and 'Veloudo' (14.6%), indicating that perhaps seed drying occurred at a more rapid rate in the latter, thus contributing to the higher incidence of hard seeds in these cultivars [7].

Seed germination was higher in Pylaias' and 'Clemson' (mean = >90.0%) than in 'Boyiatiou' (68%) and 'Veloudo' (76.0%) (Table 4) due to the higher incidence of hard seeds in the latter (22.0% and 13.0%, respectively) compared with <5.0% in the other cultivars. Although seed germination is known to correlate with the position of the pod on the plant [6], it did not correlate with the number of pods per plant. For example, in 'Boyiatiou' germination was highest in seed from plants with 10 pods (rather than 6 or 12), while in 'Pylaias' and 'Clemson' germination was highest in seed from plants with 15 pods (rather than from plants with a higher or lower number of pods). Only in the case of 'Veloudo' was a progressive reduction in seed germination recorded with increasing pod load (Table 4). Nabi et al. [13, 14] reported that seed germination was highest when the lower number of fruits per plant was retained (six fruit per plant). Overall, therefore, it may be concluded that both germination and the incidence of hard seeds is determined primarily by the genotype and not by the number of pods per plant.

An important observation here was that seed storage for 18 months at room temperature improved germination in all cultivars, especially in 'Veloudo' and 'Boyiatiou', indicating that storage reduced the effect of seed hardness.

In conclusion, the effects of pod load on pod and seed characteristics were slight in comparison with the

effect of genotype. Genotype strongly influenced pod size, seed number, germination and seed hardness, whereas differences within cultivars showed no consistency with the number of pods maturing on the plant. Therefore, pod load is not considered to affect okra seed quality and especially for crops intended for seed production growers should not apply pod thinning since no effect in seed quality was observed.

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