

Impact of after-ripening in hot pepper seed development during post-anthesis physiological maturity

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ABSTRACT

Seed development during physiological maturity is critical for ensuring its quality hot pepper. Though seed attain its highest quality at this stage but certain quality attributes attain the optimum level only after-ripening. Present study highlights the affect of after-ripening on seed quality in hot pepper. The seed harvested at peak physiological maturity behaved differently under after-ripening period. The two-year pooled data showed seed harvested at 65 days after anthesis (DAA) with 2 days of after-ripening (DAR) and at 55 DAA with 5 DAR produce the highest seed germination (87 & 90%), speed of germination (12.4 & 12.8), viability (87 & 85.5%), usable transplants (83.5 & 88%), seedling vigour index (1446 & 1556) and 1000-seed weight (7.2 & 6.6 g).

Key words: Hot pepper, physiological maturity, post ripening, seed viability.

Seed development is critical for reproduction and dispersal in both gymnosperms and angiosperms, considering its indispensability for plant survival and perpetuation. In the case of fleshy-fruited species such as cucumber, seeds attain maximum germination and vigour at the end of the seed-filling period when physiological maturity is reached (Welbaum, 1). However, in some species such as hot pepper (Capsicum annuum L.), development of maximum seed dry matter and seed quality do not coincide (Demir and Ellis, 2; Oliveira et al., 3). In the case of tomato seeds, the highest germination percentage was obtained at 70 DAA (days after anthesis), while the maximum dry matter content was observed at 50 DAA. Similarly, in pepper seeds, maximum seed germination and dry matter were attained at 60 and 50 DAA, respectively (Harrington, 4). In general, degenerative changes occur in the seeds once full physiological maturity has been attained (Leubner-Metzger, 5). However, in the case of certain seeds like tobacco, dry storage of ripened seeds at ambient temperature helps overcome dormancy and encourages germination (Nichols et al., 6).

In the present study, influence of after-ripening on post-anthesis seed physiological maturity was studied in hot pepper at UAS, Raichur during 2012-14. For the study, two sets of five fruits in three replications were randomly selected from experimental block at 50, 55, 60, 65, 70 and 75 DAA and kept for after-ripening for 2 and 5 days. Seeds were extracted following the afterripening period, and washed with clean distilled water. Seeds were then subjected to various quality tests as per the standard methods, *i.e.* germination (%) Nichols *et al.* (6); speed of germination (Agrawal, 7), mean germination time (Ellis and Roberts, 8); emergence rate index (Adetimirin, 9); seedling vigour index (Abdul Baki and Anderson, 10); usable transplants (%) (Hartley *et al.*, 12), seed viability (%) using 2-3-5 triphenyl tetrazolium chloride (Perry, 11), and 1000-seed weight (Elliott *et al.*, 13).

The stained embryos in tetrazolium test were photographically documented during seed development at different post-anthesis stages microimage stereomicroscope (Fig. 1). The unstained sections of the embryo were considered unviable, dead or non-vigorous. The data collected was analyzed statistically using randomized block design (Panse and Sukhatme, 14). The seeds collected from fruits after different periods of anthesis with after-ripening effect, showed different quality behavior (Tables 1&2). The two-year pooled data on seeds collected at 65 DAA with 2 days after ripening (DAR) exhibited the highest germination (87%), speed of germination (12.4), mean germination time (44.01), emergence rate index (3.28), viability (87%), usable transplant (83.5%), seedling vigour index (1446) and 1000-seed weight (7.2 g). The viability test showed no living tissues when seeds were collected at 50 DAA with 2 DAR (Fig. 1-c). Further, seeds were under-developed with unfilled endosperm and non-living necrotic patches at 55 and 60 DAA with 2 DAR (Fig. 1-d & e). However, seeds collected at 65 DAA with 2 DAR possessed an intact embryo and a well-developed endosperm (Fig. 1-f). This stage was considered as the peak of physiological maturity period and seeds collected at this stage have the best quality. This was further proved by the stages in 70 and 75 DAA with 2

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Table 1. Effect of after-rik	oening or	n germir	nation and	l seed p	hysiolog	ical matu	ırity.								
Treatment	Gern	nination	(%)	Speed	of germ	ination	Mean g	erminati	on time	Emerge	snce rate	index	ΪŻ	ability (%	(9
I	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled
T-1-50 DAA* + 2 DAR*	64	60	62	8.9	8.1	8.5	34.36	33.12	33.74	2.5	2.4	2.43	61.0	59.0	60.0
T-2-55 DAA + 2 DAR	73	71	72	10.2	9.9	10.1	39.21	36.43	37.82	2.8	2.8	2.80	73.0	74.0	73.5
T-3-60 DAA + 2 DAR	76	78	77	11.1	10.4	10.8	38.93	39.01	38.97	2.8	2.8	2.79	70.0	68.0	69.0
T-4-65 DAA + 2 DAR	89	84	87	13.0	11.9	12.4	45.57	42.44	44.01	3.3	3.3	3.28	89.0	85.0	87.0
T-5-70 DAA + 2 DAR	81	83	82	11.3	12.8	12.0	43.86	43.20	43.53	3.1	3.1	3.12	78.0	86.0	82.0
T-6-75 DAA + 2 DAR	78	81	80	10.9	11.2	11.0	42.00	39.26	40.63	3.0	3.0	3.00	75.0	72.0	73.5
T-7-50 DAA + 5 DAR	74	71	73	11.1	11.0	11.0	36.79	35.44	36.12	2.6	2.6	2.61	70.0	69.0	69.5
T-8-55 DAA + 5 DAR	91	89	06	13.3	12.3	12.8	46.50	42.42	44.46	3.3	3.3	3.31	85.0	86.0	85.5
T-9-60 DAA + 5 DAR	88	89	89	12.4	11.9	12.1	47.21	44.26	45.74	3.4	3.4	3.39	80.0	79.0	79.5
T-10-65 DAA + 5 DAR	67	66	67	9.7	10.5	10.1	34.64	34.54	34.59	2.5	2.5	2.49	64.0	65.0	64.5
T-11-70 DAA + 5 DAR	71	70	71	10.3	9.4	9.9	36.43	37.21	36.82	2.6	2.6	2.60	68.0	67.0	67.5
T-12-75 DAA + 5 DAR	63	65	64	9.4	10.0	9.7	31.50	33.40	32.45	2.3	2.3	2.28	60.0	60.0	60.0
CD at 5%	1.79	5.68	3.7	0.39	0.25	0.47	0.19	1.64	0.85	0.18	0.38	0.36	4.03	4.32	4.77
*Days after anthesis; #Days o Table 2. Effect of after-rip	f after-ripe sening or	ning 1 seed v	/igour and	d seed n	naturity p	oaramete	ß.								
Treatment	Usable	transpl	ant (%)	Roo	t length	(cm)	Shoc	xt length	(cm)	Seedlir	ng vigour	· index	1000-s	eed weig	ght (g)
	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled
T-1-50 DAA* + 2 DAR*	60.0	62.0	61.0	9.4	9.6	9.5	4.1	4.0	4.1	867	816	842	6.1	6.11	6.1
T-2-55 DAA + 2 DAR	73.0	69.0	71.0	9.8	9.4	9.6	4.5	4.6	4.5	1040	994	1017	6.0	6.12	6.0
T-3-60 DAA + 2 DAR	71.0	70.0	70.5	10.3	10.4	10.4	4.7	4.6	4.6	1141	1170	1155	6.3	6.42	6.4
T-4-65 DAA + 2 DAR	85.0	82.0	83.5	11.3	11.2	11.3	5.4	5.5	5.5	1490	1403	1446	7.4	7.00	7.2
T-5-70 DAA + 2 DAR	76.0	80.0	78.0	9.5	10.2	9.9	4.5	4.4	4.5	1140	1212	1176	6.3	6.82	6.5
T-6-75 DAA + 2 DAR	75.0	76.0	75.5	11.1	11.3	11.2	5.4	5.5	5.4	1282	1361	1322	6.3	6.70	6.5
T-7-50 DAA + 5 DAR	71.0	73.0	72.0	10.6	10.9	10.8	5.2	5.3	5.2	1168	1150	1159	6.2	6.24	6.2
T-8-55 DAA + 5 DAR	87.0	89.0	88.0	11.9	10.7	11.3	5.9	6.1	6.0	1617	1495	1556	6.6	6.61	9.9
T-9-60 DAA + 5 DAR	84.0	88.0	86.0	10.8	11.0	10.9	5.1	6.2	5.6	1393	1531	1462	6.2	6.23	6.2
T-10-65 DAA + 5 DAR	65.0	61.0	63.0	10.4	9.9	10.1	5.0	5.2	5.1	1029	697	1013	6.4	6.12	6.2
T-11-70 DAA + 5 DAR	69.0	60.09	64.5	9.8	9.2	9.5	4.1	4.9	4.5	988	987	988	6.1	6.18	6.1
T-12-75 DAA + 5 DAR	59.0	62.0	60.5	9.6	9.6	9.6	4.3	4.9	4.6	875	943	606	6.0	6.21	6.1
CD at 5%	3.83	3.1	3.48	0.16	0.41	0.72	0.22	0.35	0.50	20.38	26.9	45.36	0.21	0.24	0.25

Treatment	Usable	transpla	ant (%)	Root	length ((cm)	Shoo	t length	(cm)	Seedlin	ig vigour	index	1000-s	eed we
	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013	Pooled	2012	2013
T-1-50 DAA* + 2 DAR#	60.0	62.0	61.0	9.4	9.6	9.5	4.1	4.0	4.1	867	816	842	6.1	6.11
T-2-55 DAA + 2 DAR	73.0	69.0	71.0	9.8	9.4	9.6	4.5	4.6	4.5	1040	994	1017	6.0	6.12
T-3-60 DAA + 2 DAR	71.0	70.0	70.5	10.3	10.4	10.4	4.7	4.6	4.6	1141	1170	1155	6.3	6.42
T-4-65 DAA + 2 DAR	85.0	82.0	83.5	11.3	11.2	11.3	5.4	5.5	5.5	1490	1403	1446	7.4	7.00
T-5-70 DAA + 2 DAR	76.0	80.0	78.0	9.5	10.2	9.9	4.5	4.4	4.5	1140	1212	1176	6.3	6.82
T-6-75 DAA + 2 DAR	75.0	76.0	75.5	11.1	11.3	11.2	5.4	5.5	5.4	1282	1361	1322	6.3	6.70
T-7-50 DAA + 5 DAR	71.0	73.0	72.0	10.6	10.9	10.8	5.2	5.3	5.2	1168	1150	1159	6.2	6.24
T-8-55 DAA + 5 DAR	87.0	89.0	88.0	11.9	10.7	11.3	5.9	6.1	6.0	1617	1495	1556	6.6	6.61
T-9-60 DAA + 5 DAR	84.0	88.0	86.0	10.8	11.0	10.9	5.1	6.2	5.6	1393	1531	1462	6.2	6.23
T-10-65 DAA + 5 DAR	65.0	61.0	63.0	10.4	9.9	10.1	5.0	5.2	5.1	1029	697	1013	6.4	6.12
T-11-70 DAA + 5 DAR	69.0	60.0	64.5	9.8	9.2	9.5	4.1	4.9	4.5	988	987	988	6.1	6.18
T-12-75 DAA + 5 DAR	59.0	62.0	60.5	9.6	9.6	9.6	4.3	4.9	4.6	875	943	606	6.0	6.21
CD at 5%	3.83	3.1	3.48	0.16	0.41	0.72	0.22	0.35	0.50	20.38	26.9	45.36	0.21	0.24
*Days after anthesis; *Days o	f after-ripe	ening												

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DAR, where the embryo has developed a dead root tip and was thus prone to mechanical damage like harvesting injury (Fig. 1g & h).

Similarly, the after-ripening effect could be deduced further in another experiment where seeds were collected 5 DAR. Here, the seeds collected at 55 DAA with 5 DAR showed a well-developed



Fig. 1. Seed development with viability staining with tetrazolium solution. a&b-Sampling and afterripening for two and five days; c to h-Seed development from 50-75 Days After Anthesis (DAA) with 2-Days of After-Ripening (DAR); c-at 50 DAA+ 2 DAR Embryo development showing immature embryo completely unstained and dead; d-at 55 DAA+2 DAR seed with under developed and unfilled endosperm; e-at 60 DAA+2 DAR good seed filling but embryo show some necrotic patches: f- at 65 DAA+2 DAR seed with highest physiological maturity showing intact and vigorous embryo; g-at 70 DAA+2 DAR seed over ripen showing dead root tip; h-75 DAA+2 DAR seed fully surpassed physiological maturity prone for mechanical damage; i to n-Seed development from 50-75 DAA with 5-DAR; i-at 50 DAA+5 DAR seeds showing little viability; j-at 55 DAA+5 DAR seed is well developed with intact embryo; k-at 60 DAA+5 DAR over ripen or decaying seed; I-at 65 DAA+5 DAR seeds with dead tissue-half of embryo dead; m-at 70 DAA+5 DAR seeds with low vigorous embyo; n-at 75 DAA+5DAR seeds with radical protrusion which may germinate inside the fruit.

embryo and endosperm (Fig.1-j) with the highest germination (90%), speed of germination (12.8), viability (85.5%), usable transplant (88%), seedling vigour index (1556) and 1000-seed weight (6.6 g). However, higher mean germination time (44.01) and emergence rate index (3.39) were recorded when seeds were collected at 60 DAA with 5 DAR. The embryo and endosperm were not well-developed and displayed little staining at 50 DAA with 5 DAR (Fig. 1-i). Although the seeds displayed remarkable development within 10 days after that, rapid decaying was observed after 60 DAA with 5 DAR (Fig. 1-k to m). At 75 DAA with 5 DAR, protrusion of radical indicating the start of germination inside the fruit could be observed (Fig. 1-n). Seeds collected at this stage if kept for a longer period after ripening, are likely to undergo pre-germination. The development of intact embryo with healthy endosperm was observed at 65 and 55 DAA with 2 and 5 days of after-ripening; this could be mainly attributed to appropriate physiological maturity and higher sensitivity of seed to increased gibberellic acid (GA) and decreased abscissic acid (ABA) levels (Leubner-Metzger, 4). Significantly, lack of after-ripening can interfere with germination at early stages even under favorable environmental conditions (Finch-Savage and Leubner-Metzger, 15). Afterripening is a must for triggering uniform germination. The seeds harvested at correct physiological maturity with proper after-ripening will have a well-developed embryo, hypocotyl, endosperm and cotyledons, which result in uniform and vigorous plants (Penfield, 16). Seeds collected at early stages tend to possess an abnormal embryo and other structures due to low dry matter accumulation, which reduce expression of such essential structures (Demir and Ellis, 2). Similarly, seeds harvested at delayed physiological maturity with or without after-ripening show ageing, dead root tips or pre-germination. This may be due to quick hydration of micropylar endosperm cap with hypocotyls, which result in imbibition of radical and its protrusion (Terskikh, 17). The protrusion of radical is the onset of seed germination. Hence, the correct stage for harvest in hot pepper has been deciphered.

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