

## Modification in biochemical actions of wheat (*Triticum aestivum* L.) seed under variable storage situation

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### ABSTRACT

The biochemical action accompanying seed storability of wheat seed (cv. C306) was very much controlled through the approach of variable storing devices. The treatment outcome was prominent at later stages, accelerating a declining trend of biochemical reaction interrelated to seed quality or protecting storability. Considering diverse quality linking parameters, the inhibitory effect on waning nature provided the highest peak in PC (plastic container) followed by PP (40 micron plastic packet) where EP (earthen pot) retained uniformity mostly with the normal (brown paper packet), sometimes with PN (brown paper packet+ naphthalene ball @ 5 g kg<sup>-1</sup> seed). The 90 days storage was the turning point for lowering the biochemical activity in normal seed, representing in Electrical conductivity and in  $\alpha$ -amylase enzyme activity under variable imbibing stages though a significant discrepancy was observed in application of treatments. In action of peroxidase, a noticeable drop was started after 135 days of storage which was alike to total carbohydrate content. As a result, PC (plastic container) and PP (40 micron plastic packet) can be utilized for wheat seed storing to conserve the storability.

**Keywords** : Biochemical, storage container, storability, wheat seed

The seed production potentiality of any field crop mainly depends on nurturing aptitude and post-harvest handling. The seed at post-harvest state especially in stored situation controls the seed quality through alternation of internal integrity in biochemical action. In Indian subtropical condition, Wheat seeds are to pass a lot of discrepancy in environment particularly in eastern zone in which very high relative humidity of the rainy season at post-harvest stage was one of the main components for varying the biochemical action. Inherent potentiality of any genotype is vital for quality produce though their protection up to next season is similarly imperative for next crop. The seed of superior genotype cannot fully potential under poor maintenance in storage that must be linked to deviation of biochemical action of seed.

Deterioration of stored seed is influenced by physical (temperature, humidity), biological (micro-flora) (Dharam, 1986; Srivastava *et al.*, 2011) and technical (storage conditions, methods and duration) factors. Biochemical and physiological deterioration during seed ageing had been studied mostly under accelerate ageing conditions using high temperature and high seed water content (McDonald, 1999; Hsu *et al.*, 2003).

The plan of this study was to assess the nature of some biochemical activity in relation to seed storability of wheat under dissimilar micro-environment containing storing devices as well as to quantify the

deterioration pattern under a certain interval. The nature of these evaluations may specify the appropriate device for post-harvest handling.

### MATERIALS AND METHODS

The experiment was carried out in the Project Laboratory under Rashtriya Krishi Vikas Yojana, Department of Seed Science and Technology, Bidhan Chandra Krishi Viswavidyalaya, West Bengal in 2012-13. The pattern of biochemical action on seed obtained from a rainfed wheat cultivar C306 considering a few unlike containers like Plastic packet (40 micron, sealed) symbolized PP, Plastic container (hard plastic, air-tight) symbolized PC, Earthen pot (sealed) symbolized EP, Packet (brown paper) includes naphthalene ball (5g kg<sup>-1</sup> seed) symbolized PN and Control (brown paper packet) symbolized C. The initial seed moisture was 11.2 %.

The evaluation procedure was at every 45 days interval initiating from the day of storing and these were continued up to 180 days. The biochemical activity of seed was appraised through estimation of electrical conductivity (EC) (Martins *et al.*, 2004), total carbohydrate (Anthrone method), enzymes activity like peroxidase (Ambreen *et al.*, 2000), and  $\alpha$ -amylase (Filner and Varner, 1967) at variable extent of imbibition.

Completely Randomized design (CRD) was followed for analysis of facts associated to these activities. The outcomes were achieved at 5 % level of significant by using SPSS (version 10.0, 1990) in Excel worksheet of Microsoft 2007.

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## RESULTS AND DISCUSSION

The least value at Electrical conductivity ( $\mu\text{S cm}^{-1}$ ) of seed pointed out the most excellent result due to smallest amount leaches of internal substances from seed. A considerable demarcation was observed from 45 to 180 days (Table 1) definite storage time but declining rate was peak at 90 days (Fig. 1) for all treatments

signifying the highest in control. The EP and PN retained in an insignificant group which was significantly diverged from another insignificant group of PC and PP. Both groups kept a significant detachment with control. PC and control always maintained the highest and lowest value respectively in different stages of storage (Table 1).

**Table 1: Electrical conductivity and total carbohydrate at different storage conditions**

Treatment	Electrical conductivity ( $\mu\text{S cm}^{-1}$ )					Carbohydrate ( $\text{mg g}^{-1}$ )				
	IMM	45 D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D
Control	81.50	90.81	140.08	145.53	144.79	108.26	107.56	107.65	107.44	106.82
PP	81.36	84.39	119.10	126.40	133.10	108.22	107.73	108.12	108.43	107.48
PC	80.83	79.20	110.60	123.20	132.68	108.27	107.60	107.89	108.44	107.96
EP	77.46	80.87	138.31	137.99	143.28	108.39	107.56	108.15	108.21	107.94
PN	77.81	80.05	136.50	136.13	142.83	108.43	107.72	107.75	108.03	107.88
Mean	79.79	83.06	128.92	133.85	139.33	108.31	107.64	107.91	108.11	107.62
<b>SEm (<math>\pm</math>)</b>	<b>4.09</b>	<b>3.09</b>	<b>2.58</b>	<b>3.28</b>	<b>2.37</b>	<b>0.29</b>	<b>0.55</b>	<b>0.49</b>	<b>0.41</b>	<b>0.53</b>
<b>LSD (0.05)</b>	<b>12.32</b>	<b>9.32</b>	<b>7.78</b>	<b>9.89</b>	<b>7.14</b>	<b>0.88</b>	<b>1.65</b>	<b>1.47</b>	<b>1.22</b>	<b>1.58</b>

*Note: PP- Paper packet, PC- Plastic Container, EP- Earthen pot, PN- Paper packet with naphthalene, IMM- Immediate, D- Days*

The total carbohydrate present in seed under different treatments and stages of storage maintained a very restricted percent of deviation towards losing from initial to 180 days of storage. The rate of deterioration was practically negligible though the rate was slightly higher in control under last stage of storage *i.e.*, 180 days (Table 1). Hence, the insignificant nature indicated no loss of total carbohydrates in sequence of storage while their internal modification/alteration may be there.

Peroxidase activity ( $\Delta\text{A min.}^{-1} \text{g}^{-1}$ ) was very much linked during the storage periods of seed. In general, the activity of iso-enzyme peroxidase will be increasing in adverse situation as well as with the progression of germination activity (duration) that clearly indicates in our observation. Therefore, the observations were continued up to day first count (96 hours) *i.e.* initiating stage of root and shoot. The 24, 48, 72 and 96 hours of water soaking were considered for variable stored seed. There was a little enhancing nature up to 90 days old seed except control that may be sustained up to subsequent examination *i.e.*, 135 days only in case of PP and PC. The reduced peroxidase activity was initiated at 135 days old seed in case of 48 and 96 hours only in EP and PN (Table 2) where control continued its maximum variation up to 180 days (Fig. 2). At 180 days, the rate of deviation was prominent indicating maximum percent of deterioration particularly in 48 hours of soaking (Fig. 2) where control was significantly maximum among all cases. The insignificant group EP and PN maintained a considerable gap with PP and PC.

The activity of  $\alpha$ -amylase ( $\text{mcg min.}^{-1} \text{g}^{-1}$ ) was very much correlated with germination procedure. In progression of the germination route, the enzyme activity was increasing while their negative tendency was marked during succession of storage duration.

The variable declining nature in action of  $\alpha$ -amylase was found in variable stored seed under different treatments though it was highest in control *i.e.*, without treatment. The control initiated the deterioration affinity from 90 days of storing (Table 3) irrespective of imbibing time (hours). But the treatments EP and PN showed the same nature at 135 days. The treatments PP and PC initiated the reduced activity of enzyme at last stages of stored seeds except 96 hrs. of seed imbibitions where maximum percent of deterioration was started from 135 days of seed storing (Fig. 3).

The minor percent of deterioration was found in between treatments and control up to 45 days but it was prominent from 90 days of storing (Fig.3). Among the different treatments, the significant distinction was initiated from 135 days. This nature was true for all variables during germinating activity up to the day of first count (96 hours).

The diverse containers (treatments) effect on the variety, C306 proved an influencing role of storing devices through their biochemical activities like EC, carbohydrate, enzymes etc. allied to most responding outer expression like vigour and viability. The treatment effect was prominent at later stages due to maximum

Table 2: Peroxidase activity ( $\Delta A \text{ min.}^{-1} \text{ g}^{-1}$ ) of different imbibed seed (24, 48, 72 and 96 hrs.) at variable days of storage

Treatment	At 24 hours				At 48 hours				At 72 hours				At 96 hours							
	IMM	45 D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D
Control	0.29	0.32	0.29	0.30	0.24	0.38	0.39	0.36	0.32	0.22	0.44	0.46	0.41	0.42	0.27	0.63	0.64	0.61	0.43	0.29
PP	0.30	0.30	0.32	0.32	0.30	0.39	0.39	0.40	0.39	0.41	0.45	0.44	0.45	0.45	0.44	0.6	0.64	0.63	0.63	0.59
PC	0.30	0.30	0.32	0.31	0.29	0.39	0.38	0.39	0.39	0.40	0.43	0.46	0.46	0.45	0.44	0.62	0.62	0.64	0.64	0.60
EP	0.29	0.30	0.32	0.32	0.31	0.38	0.39	0.41	0.40	0.31	0.44	0.44	0.42	0.41	0.35	0.58	0.62	0.63	0.54	0.57
PN	0.31	0.30	0.31	0.32	0.30	0.37	0.40	0.41	0.41	0.31	0.42	0.45	0.42	0.42	0.34	0.61	0.63	0.62	0.53	0.55
Mean	0.30	0.30	0.31	0.31	0.29	0.38	0.39	0.39	0.38	0.33	0.44	0.45	0.43	0.43	0.37	0.61	0.63	0.63	0.55	0.52
SEm( $\pm$ )	0.01	0.02	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.01	0.01	0.01	0.01	0.01	0.02	0.01
LSD (0.05)	0.03	0.05	0.04	0.03	0.03	0.04	0.04	0.04	0.03	0.04	0.05	0.05	0.06	0.04	0.04	0.03	0.04	0.03	0.05	0.03

Table 3:  $\alpha$ -amylase activity ( $\mu\text{g min.}^{-1} \text{ g}^{-1}$ ) of various soaked seed (24, 48, 72 and 96 hours) at diverse stages of storage

Treatment	At 24 hours				At 48 hours				At 72 hours				At 96 hours							
	IMM	45 D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D	IMM	45D	90 D	135 D	180 D
Control	240.5	240.8	211.6	186.1	151.9	269.0	268.1	252.9	216.5	140.4	311.7	304.5	246.0	216.9	186.0	336.4	330.4	305.1	275.2	241.4
PP	243.7	242.2	244.2	242.4	230.1	269.4	274.0	271.4	262.6	256.4	311	312.7	284.1	299.3	236.2	335.9	334.8	336.4	299.4	287.2
PC	243.5	245.9	247.0	246.0	236.5	269.9	268.0	271.9	269.8	260.4	316.5	311.5	310.1	308.8	253.6	337.6	334.4	340.3	275.9	289.6
EP	245.8	241.2	240.6	224.3	207.9	271.0	268.3	269.4	242.5	210.4	314.4	314.3	318.8	299.6	236.9	340.0	333.6	335.4	283.4	274.2
PN	242.7	242.9	237.5	221.2	200.9	268.3	268.6	271.6	246.3	215.4	313.8	315.5	308.1	292.0	225.4	334.4	336.6	337.7	304.6	276.5
Mean	243.2	242.6	236.2	224.0	205.5	269.5	269.4	267.4	247.5	216.6	313.5	311.7	293.4	283.3	227.6	336.8	334.0	331.0	287.7	273.8
SEm( $\pm$ )	2.0	2.2	2.2	3.6	4.4	3.4	1.6	2.2	1.3	2.2	2.9	3.0	7.1	3.4	3.5	1.6	1.9	2.0	14.6	3.7
LSD (0.05)	5.9	6.6	6.7	10.9	13.3	10.1	4.7	6.5	4.0	6.5	8.8	9.0	21.2	10.2	10.5	4.9	5.8	5.9	44.1	11.3

Note : C- Control, PP- Paper packet, PC- Plastic Container, EP- Earthen pot, PN- Paper packet with Naphthalene, IMM- Immediate, D- Days

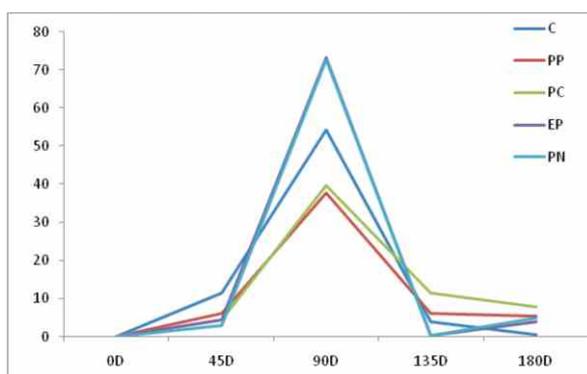


Fig. 1: Rate of deviation (%) at variable stages of storage considering electrical conductivity

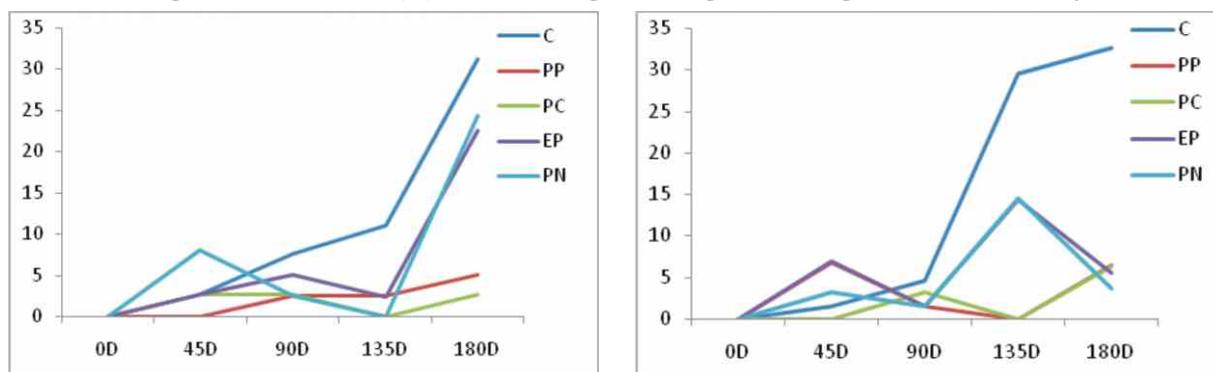


Fig. 2: Rate of deviation (%) at variable stages of storage considering the peroxidase activity at 48 and 96 hours of imbibition

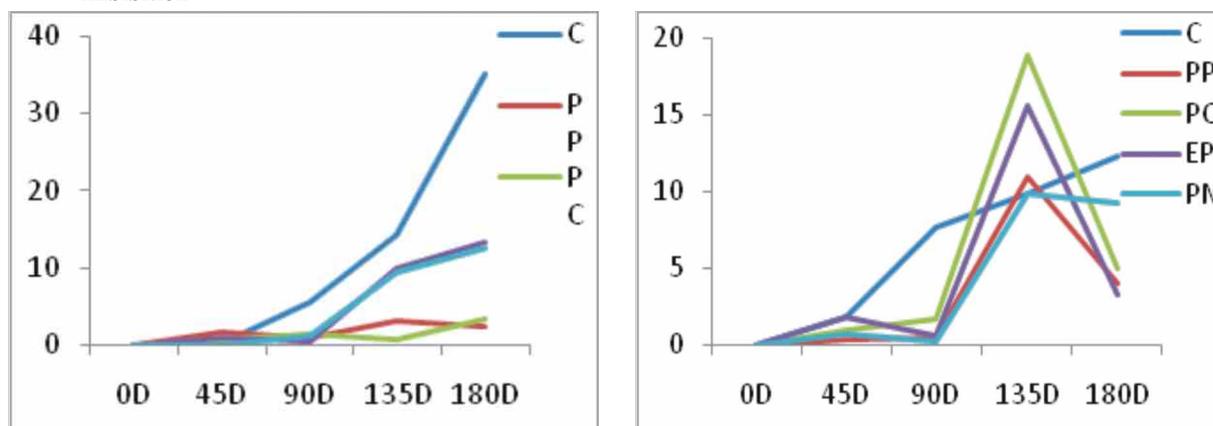


Fig. 3: Rate of deviation (%) at variable stages of storage considering  $\alpha$ -amylase activity at 48 and 96 hours of soaked seed.

deteriorating trend on biochemical action. In comparison to normal, the effect of PC provided the highest peak followed by PP where treatment EP retained uniformity mostly with the normal, sometimes with PN.

The observations on biochemical action pointed out an understandable pattern of seed deterioration which was indicated by earlier workers (Shelar, 2008; Tatipata, 2010). Mandal *et al.*, (2013) observed the variable action of bio-molecules in existence of external factors that may conveyed its effect in seedling behavior. The 90

days of storage was the turning point of normal seed deterioration where a significant discrepancy was observed in biochemical activity under different treatments. The PC and PP created a separate insignificant better group in comparison to control and others where initiation of declining comparatively later and in reduced rate.

The storage effects on seed relating to containers depend on internal microenvironment like temperature, CO<sub>2</sub>/O<sub>2</sub> pressure, seed moisture etc. that can induce the biochemical activity (Vasudevan *et al.*, 2012; Tatipata,

2010). Variable micro-environment influenced the activity of enzymes, metabolic pathways, fat acidity, membrane permeability that can expressed in seedling behavior like variation of seed germination and vigour.

Shelar (2008) described the mitochondrial action in seeds stored at 75 % RH, where it amplified the maximum after 90 days of storage and then rapidly declined. The rapid mitochondrial respiration reached an uninhibited situation which accompanied by the consumption of Co A and its acetyl derivatives. Rehman *et al.* (1999) reported that amylase activity, total soluble sugars decreased up to 37% with the progress of 180 days storage. The enhanced activity of  $\alpha$ -amylase during imbibition and germination was indispensable to expresses the seed vigour in ultimate.

The loss of germination rate varied with the specification of containers. The stored soybean seed had higher germination percentage in polylined gunny bags than the normal gunny bags irrespective of varieties, threshing and processing methods (Shelar, 2002). Vanangamudi and Ramaswamy (1984) also demonstrated the superiority of moisture impervious containers over the ordinary containers for successful carryover of seed. Reducing quantity of oxygen around the seed might also decrease the initiation of free radicals (McDonald 1999) that can partially prohibited by the effect of containers. Agarwal *et al.* (1990) observed that germination loss during storage was due to oxidation of Glucose by EMP pathway as well as pentose phosphate pathway where container played a crucial role.

Controlled microenvironment of container reduced the accelerated ageing which enhanced the germinating activity of seeds. The physiological activity like peroxidase activity, which probably eliminated chromosome-damaging peroxy radicals, developed at the time of storing. A light increase in peroxidase activity during imbibition and germination (Essemine *et al.* 2007; Mitchell *et al.* 2001) was also comparable to our observation. The peroxidase activity in endosperm and seed coat increased by 1.5 and 1.8 times respectively in 24 hrs. swelling of wheat seed (Rogozhin *et al.*, 2001). After measuring, it was clear that storing methods influenced the activity of peroxidase. Peroxidase and other assumed defense enzymes *viz.* endochitinase, exochitinase, protease, lectins *etc.* were probably worked together as a multi-component defense system to protect seeds during imbibition and germination (Mitchell *et al.*, 2001).

Lastly, the estimation of Electrical conductivity (E.C.) of stored seed was the easiest method for

evaluation of seed quality. Lower vigour seed leached out greater quantity of solutes and it would show higher value of E.C. The increasing E.C. due to ageing showed significant negative correlation with Vigour Index (V.I.) and germination (Thakur *et al.*, 2004).

Biochemical action allied to germination process was very much associated to quality of seed. Poor quality may deteriorate this activity through damage of cellular membranes and other essential organelles by oxidation. The modification of storage techniques specifically in micro-environment level increases the potentiality of seed which ultimately enhances the seed quality as well as their storability. To amplify the seed potential particularly in wheat cultivars, the present observation is very important.

The potentiality of seed can be retained up to next season through modification of macro and micro environment of various storing devices. In cereals, mainly Wheat was most approachable to prevent deterioration in presence of PC (plastic container) and PP (40 micron plastic packet). Therefore, care should be taken in post-harvest stage through application of proper storing devices.

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