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Control of Dry Rot Disease on Potato During Storage

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Potato dry rot disease caused by *Fusarium solani* during storage and marketing is investigated in the present work. Salts potassium phosphate dibasic K_2HPO_4, potassium phosphate mono basic KH_2PO_4, potassium carbonate K_2CO_3, potassium bi carbonate $KHCO_3$, sodium carbonate Na_2CO_3, and sodium bicarbonate $NaHCO_3$,, hot water and thiobendazole (Tecto) fungicide were tested for controlling of *F. solani* during cold storage at three-months (7^oC for one month then at room temperature 22 ± 2^oC for two months) and 90- 100% RH. Absolute efficacy (100%) of all tested treatments in naturally infected potato tubers for three months. The only exception was recorded for hot water treatment in that should 75.02 and 59.97% during 2011-2012, respectively. Potato tubers dipping in salts or hot water and Tecto treatments reduced the dry rot disease and maintained the proper quality of tubers during the cold storage at 7^oC for one month followed by shelf life up to 3 months. All post harvest treatments decreased the loss in fresh weight and Sprouting of Potato tubers compared with the untreated tubers during storage at 7^oC. On the other hand, all treatments increased the moisture content, and decreased polyglacturonase and cellulase activity. On the other hand, no PME activity was detected in naturally infected or artificially inoculated tubers with *F. solani* and the only exception was found in KH_2PO_4 and K_2CO_3 treatments. This study demonstrates that potato tubers can be safely stored without sprouting and control of dry rot for 3 months (7^oC for one month then storage of two month at room temp. 22 ± 2^oC) if dipped in KH_2PO_4, $KHCO_3$ and Na_2CO_3.

Keywords: Dry rot, *Fusarium solani*, hot water, post harvest, potato, salts, Tecto fungicide.

Dry rot is one of the most important post-harvest diseases of potato tubers (*Solanum tuberosum*), that causing significant economic losses worldwide (Stevenson *et al*., 2001 and Jensen1, *et al*., 2011). The predominant hosts for *Fusarium solani* are potato, pea, bean, and members of the cucurbit family such as melon, cucumber, and pumpkin. Some strains may cause infections in humans.

Potassium sorbate, sodium benzoate, sodium carbonate, sodium metabisulfite and trisodium phosphate were shown to reduce certain potato diseases as silver scurf caused by *Helminthosporiun solani* (Olivier *et al*., 1998; Hervieux *et al*., 2002; Mills *et al*., 2006) and dry rot caused by *Fusarium sambucinum* (Mecteau *et al*., 2002).

Many fruits and vegetables crops are tolerating the exposure to water temperatures of $50\text{-}60^oC$ for up to 10 min, but shorter exposure at this temperature can control many post harvest plant pathogens (Barkai-Golan and Phillips, 1991).

Sprouting of tubers due to invasion by bacterial and fungal pathogens are problems of potato storage that are usually dealt with by specialized chemicals. (Ranganna *et al*., 1998).

After harvest, normal seed tubers show dormancy for about 1-15 weeks, depending on cultivar, tuber size, and conditions before harvest and storage conditions. Small tubers, such as minitubers, even have longer periods of dormancy (Lommen, 1993) and are more sensitive to adverse conditions during storage (Struik and Wiersema, 1999). Changing conditions during storage, especially the storage temperature, can accelerate the progress of the physiological ageing. Cold shocks, heat shocks and warm temperatures are being concerned in breaking of dormancy (Struik and Wiersema, 1999).

The objective of this work is to study the effect of six salts solutions or hot water and the recommended benzimidazole (thiobendazole) fungicide Tecto, on protecting potato tubers from dry rot disease caused by *Fusarium solani* and maintaining quality of tubers during storage at 7^{o}C and 90- 100% RH for one month. The shelf life at 22$\pm$2^{o}C following the cold storage was also determined in addition to tubers quality characteristics.

Materials and Methods

A) Source of fungus pathogen:

Pathogenic isolate of *Fusarium solani* isolated from diseased potato tubers with dry rot was used for all control experiments.

B) Post harvest treatments:

Six salts, *i.e*. potassium phosphate dibasic K_2HPO_4, potassium phosphate mono basic KH_2PO_4, potassium carbonate K_2CO_3, potassium bi carbonate $KHCO_3$, sodium carbonate Na_2CO_3, and sodium bicarbonate $NaHCO_3$, were applied at 600 ppm concentration (0.6g/l). Also, hot water (45^{o}C/5min) as well as the fungicide Thiobendazole (Tecto 45%) at concentration of 1.5ml/l, were tested.

Fresh potato tubers (cv. Spunta) apparently free of mechanical damage and visual disease symptoms were used in this experiment. Tubers were surface disinfected with 70% ethyl alcohol. The tubers were dipped in post-harvest treatments for 1 minute then transferred into carton bags and stored at 7^{o}C for one month followed by two months at room temp (22$\pm$2^{o}C). Each treatment contained three replicates each of 9 tubers. Three bags each contained 9 untreated tubers were used as control. The percentage of infection and efficacy with *Fusarium solani* was determined as follows:

$$\text{Infection (\%)} = \frac{\text{Infection}}{\text{Total tubers}} \times 100$$

$$\text{Efficacy (\%)} = \frac{\text{Control} - \text{treatment}}{\text{Control}} \times 100$$

Other group of potato tubers inoculated by *Fusarium solani* growth disks (3mm), inserted onto potato tubers injured. After 24 hours, the inoculated potato tubers were dipped in the given treatments.

All treated tubers as well as the untreated (non inoculated and inoculated) were stored for one month at 7°C and 90-100% RH conditions. The stored potato tubers were examined weekly for detection of disease progress Post harvest treated, as well as untreated potato tubers were stored after packing in a cold room at 7°C for one month. After the cold storage, the Diseases severity was calculated using the scale from 0-4 (0 = healthy, 1 = 1-25% infection, 2 = 26-50% infection, 3 = 51-75% infection and 4 = 76-100% infection) and formula adopted by Hanounik (1986) as follows:

$$\text{Disease severity (\%)} = \frac{\Sigma\,(\text{NCC} \times \text{CR})}{\text{NTC} \times \text{MSC}} \times 100$$

Whereas: NCC= No. of potato tubers in each class rate.
NTC= No. of total potato tubers.
CR = Class rate.
MSC= Maximum disease severity class rate.

Shelf-life:

After cold storage for one month, the shelf life was estimated for potato tubers either treated or untreated with post harvest treatments by removing the tubers from the refrigerator and keeping them at 22±2°C for shelf life investigation. Shelf life was determined, as the 50% of tubers remained healthy without decay symptoms.

C) Potato tubers quality parameters:

Tuber quality parameters as shown by Loss in weight (LW), moisture content (MC), and Sprouting were determined 4 month after inoculation for inoculated and non-inoculated tubers.

LW:

Losses in potato tubers fresh weight percentage (grams fresh weight %) were estimated in the inoculated and non-inoculated potato tubers for all treatments (average weight of 27 tubers for each treatment) according to the following formula:

$$\text{LW (\%)} = \frac{\text{Initial weight - weight of potato tubers at sampling date}}{\text{Initial weight of potato tubers.}} \times 100$$

MC:

Moisture content of potato tubers was estimated according to the following equation:

$$\text{MC (\%)} = \frac{\text{Fresh weight of potato tubers - dry weight of potato tubers}}{\text{fresh weight of potato tubers}} \times 100$$

Sprouting:

$$\text{Sprouting (\%)} = \frac{\text{No of potato tubers sprouting}}{\text{Total potato tubers}} \times 100$$

Pectolytic enzymes:

Potato samples (100 g) taken from particular treatment were blended for 15 min. in a warning blender containing the same amount of distilled water (100 ml), then filtered through muslin and centrifuged at 3000 rpm for 20 minutes. The clear supernatants were used to estimate the activity of pectin methyl esterase (PME), polyglacturonase (PG) and Cellulase (Cx) enzymes. The measurement of enzyme activity was carried out in the filtrates according to the method of (Aneja, 2001).

1- Pectin methyl esterase (PME):

Five ml of crude enzyme preparation of each post harvest treatment and five ml 1.2% aqueous highmeoxyl pectin buffered at PH 5.6 were allowed to react for 4 hours at 30^{o}C after that titrated back to PH 5.6 with 0.01 N (NaOH) solution. The reaction was continued for 20 hours after which the PH was readjusted to PH 5.6 and the total volume of 0.01 N (NaOH) required over the 24 hours was calculated. A check was carried out in which 5 ml of boiled crude enzyme was added to 5 ml buffer substrate. The difference between the volume in ml of 0.01 N (NaOH) required for the crude enzyme and that required for the boiled check, gave an approximate indicator of the relative activities of the comparable preparation. Contamination with microorganisms was prevented by the addition of two drops of chloroform to each reaction mixture tube.

2- Cellulase (Cx) and Polyglacturonase (PG) activities:

Samples of 100 grams of potato tubers of each treatment were blended in 200 ml distilled water for 2 minutes. The mixture was squeezed through several layers of cheesecloth and centrifuged at 3000 rpm for 20 minutes. Supernatants were kept at 5^{o}C until assaying. Loss in viscosity was measured according to the following formula:

$$\text{Activity (\%)} = \frac{(T_0 - T_t)}{T_0 - T_w} \times 100$$

Whereas: T_0 = Time of flow of the reacted mixture at zero time; T_t = Time of flow at a given time intervals of the sample and T_w = Time of flow of the distilled water

Statistical analysis:

All data obtained were subjected to the proper statistical analysis using the MSTAT statistical software and comparison was made following Fishers L.S.D. (0.05).

Results

Data in Table (1) indicated that KH_2PO_4, K_2CO_3, $KHCO_3$, Na_2CO_3, $NaHCO_3$, concentration (0.6g/l), resulted in complete inhibition (100% efficacy) after one month storage season 2011 compared to the control but the diseases severity increased after three months storage with the salts in the range (6.48 to 20.37) season 2011. On the other hand, hot water after one, two and three months gave the highest disease severity (18.52-29.63-61.11) respectively. Generally, all treatments

Table 1. Effect of pre storage tuber dip in salts or hot water on severity of infection and efficacy of *Fusarium solani* season 2011 of potato tubers after storage for one month at 7°C followed by two, three months at room temp (22±2°C)

Treatment	Disease severity (%)			Efficacy (%)		
	One	Two	Three	One	Two	Three
K_2HPO_4	2.77	8.33	15.74	94.5	90.0	83.2
KH_2PO_4	0.0	0.0	10.19	100	100	89.1
K_2CO_3	0.0	10.18	20.37	100	69.9	78.2
$KHCO_3$	0.0	0.0	6.48	100	100	93.0
Na_2CO_3	0.0	0.0	8.33	100	100	91.0
$NaHCO_3$	0.0	4.62	9.26	100	94.5	90.1
Hot water	18.52	29.63	61.11	62.96	64.4	34.7
Tecto	0.0	7.41	12.96	100	91.1	86.1
Control	50.0	83.3	93.52	0.0	0.0	0.0
L.S.D. at 0.05%	0.97	0.61	1.28			

significantly decreased diseases severity after storage for one, two and three months (50.0 - 83.3 - 93.52), respectively, compared to the control. The efficacy of $KHCO_3$and Na_2CO_3 to control *Fusarium solani* reached 93.0% and 91.0% followed by KH_2PO_4 (88.9%) and $NaHCO_3$ (83.0%) in the season 2011 for three months.

Data in Table (2) show that KH_2PO_4, K_2CO_3, $KHCO_3$, Na_2CO_3, $NaHCO_3$ and the fungicide Tecto gave the best control of disease severity (0.0) followed by K_2HPO_4 and hot water (3.77, 12.30), respectively, to control tuber infection with *Fusarium solani* after one month cold 7°C storage at the season 2012.

Table 2. Effect of pre storage tuber dip in salts or hot water on severity of infection with *Fusarium solani* season 2012 of potato tubers after storage at 7°C for one month followed by two, three months at room temp (22±2°C)

Treatment	Disease severity (%)			Efficacy (%)		
	One	Two	Three	One	Two	Three
K_2HPO_4	3.77	9.26	16.67	92.60	89.01	82.85
KH_2PO_4	0.0	0.93	11.11	100	98.90	88.57
K_2CO_3	0.0	13.80	22.22	100	83.62	77.14
$KHCO_3$	0.0	2.77	7.41	100	96.71	92.38
Na_2CO_3	0.0	3.70	9.30	100	95.61	90.43
$NaHCO_3$	0.0	5.56	10.19	100	93.40	89.52
Hot water	21.30	30.56	61.81	58.17	63.73	33.34
Tecto	0.0	8.33	16.67	100	90.11	82.85
Control	50.92	84.26	97.22	0.0	0.0	0.0
L.S.D. at 0.05%	1.04	0.52	0.73			

Prolonged tuber storage at room temperature (22±2°C) for three months resulted in high disease severity that reached 61.81% during season 2012 (Table 2).

Data in Table (3) showed the effect of post harvest treatments on natural infection of potato tubers after three months storage at two successive seasons 2011and 2012. High efficacy (100% control) on potato tubers was resulted when tubers were stored after certain post harvest treatments with all salts and Tecto fungicide in the two seasons. Hot water, however, had the least effect on the disease infection after three months (3.70 and 7.41) efficacy (75.02 and 59.97%) at the two seasons2011 and 2012, respectively, compared with the control.

Table 3. Effect of pre storage tuber dip in salts or hot water on natural infection in 2011 and 2012 after storage at 7°C for one month followed by two months at room temp (22±2°C)

Treatment	2011		2012	
	Infection (%)	Efficacy (%)	Infection (%)	Efficacy (%)
K_2HPO_4	0.0	100	0.0	100
KH_2PO_4	0.0	100	0.0	100
K_2CO_3	0.0	100	0.0	100
$KHCO_3$	0.0	100	0.0	100
Na_2CO_3	0.0	100	0.0	100
$NaHCO_3$	0.0	100	0.0	100
Hot water	3.70	75.02	7.41	59.97
Tecto	0.0	100	0.0	100
Control	14.81	0.0	18.51	0.0
L.S.D. at 0.05%	0.25		0.41	

Data in Table (4) show that post harvest treatment highly reduced the loss of fresh weight of potato tubers during (the cold storage at 7°C for one month and two months at room temp.). The treatments with KH_2PO_4, $KHCO_3$ and Na_2CO_3 were the most effective in decreasing the weight loss of the artificially inoculated potato tubers with *F. solani*. Generally, pre storage dip of potato tubers in post harvest treatment decreased the development of dry rot disease and maintained the tubers quality and reduced the weight loss during storage for one month at 7°C and two months at room temp.

The shelf life of potato tubers increased with the post harvest treatments in concern (Table 5). Found to be regarding the shelf life of the artificially inoculated potato tubers, it was extended by K_2CO_3 treatment to 100 days, but hot water treatment to 60 days when tubers were cold stored for one month. On the other hand, the shelf life of naturally infected potato tubers was extended to 120 days by all post harvest treatments when the tubers were cold stored for one month.

Table 4. Effect of pre storage tuber dip in salt treatments and hot water treatment on weight loss of potato tubers after storage at 7°C for one month at 7°C followed by two, three months at room temp. (22±2°C)

Treatment	*Fusarium solani*			Natural infection		
	One	Two	Three	One	Two	Three
K_2HPO_4	0.77	1.90	3.82	0.0	0.31	0.70
KH_2PO_4	0.03	0.95	1.32	0.0	0.88	1.10
K_2CO_3	0.01	0.99	1.77	0.0	0.32	0.75
$KHCO_3$	0.05	0.88	1.34	0.0	0.37	0.79
Na_2CO_3	0.08	0.97	1.40	0.0	1.57	3.24
$NaHCO_3$	2.15	4.33	8.18	0.0	0.41	1.22
Hot water	5.64	8.66	12.80	0.0	1.06	2.25
Tecto	0.66	1.30	3.62	0.0	0.85	1.31
Control	15.21	20.3	30.0	0.50	2.21	4.2
L.S.D.at0.05%	0.35	0.58	0.77	0.04	0.35	0.49

Table 5. Shelf life of potato tubers in days at 22±2°C after storage treatments at 7°C and 90-95% RH for one month

Treatment *	*F. solani*
K_2CO_3	100
Hot water	60
Control	30

* K_2HPO_4, KH_2PO_4, $KHCO_3$, Na_2CO_3, $NaHCO_3$, and Tecto artificial inoculation of *F. solani* increase of 120 days and also shelf life in all treatments to the natural infection increase of 120 days.

Generally, pre storage dip of potato tubers in post harvest treatments decreased the development of dry rot disease and maintained the tubers quality and decreased the weight loss during storage for three months and prolonged the shelf life.

All post harvest treatments increased moisture content and decreased Sprouting of potato tubers compared to the control in naturally infection and artificially inoculated with *F. solani* (Table 6). The moisture content of potato tubers reached its maximum (77% and 69%) when K_2HPO_4 after harvest in natural and artificially inoculated tubers, respectively. On the other hand, sprouting of potato tubers reached in hot water treatment in both natural infection and artificial inoculation 66.67 - 70.37, compared to the control (81.48 –100), respectively.

Activities of CX and PG enzymes were determined in extracts of natural infection and artificially inoculated potato tubers with *F. solani*. The obtained results in Table (7) indicate that post harvest treatments both natural infection and artificially inoculated in potato tubers decrease activities of both PG and CX enzymes compared with the control It was clear also from the obtained results that the activities of PG and CX enzymes were higher in artificial inculcation of *F. solani* than natural infection. Also, increasing the reaction time from 15-30 min. raised the activities of PG and CX enzymes gradually to be higher at 30 min., than at 15 min.

Table 6. Effect of certain post harvest treatments on Moisture content and Sprouting of potato tubers during 3months storage at (7°C/1 month and 2 months at room temp.) under natural infection and artificial inoculated of *F. solani*

Treatment	Moisture content (%)		Sprouting	
	F. solani	Natural infection	*F. solani*	Natural infection
K_2HPO_4	69	77	40.74	37.04
KH_2PO_4	67	70	7.41	3.70
K_2CO_3	68	71	14.81	11.11
$KHCO_3$	65	69	11.11	7.41
Na_2CO_3	64	66	29.63	22.22
$NaHCO_3$	66	72	33.33	18.52
Hot water	62	65	70.37	66.67
Tecto	60	67	48.15	25.93
Control	54	60	100	81.48

Table 7. Cellulase and Polyglacturonase activity on natural or artificially infected potato tubers for 2 months storage

Treatment *	Reduction in viscosity (%)				
	Time (Min)	Cx		PG	
		F. solani	Natural infection	*F. solani*	Natural infection
K_2HPO_4	15	22.22	0.0	26.19	1.02
	30	26.19	17.10	35.13	18.3
KH_2PO_4	15	20.10	0.0	29.03	3.3
	30	23.14	8.88	29.23	10.4
K_2CO_3	15	24.44	17.07	25.04	18.06
	30	25.16	20.83	25.31	21.06
$KHCO_3$	15	22.13	17.33	24.04	19.2
	30	24.17	20.04	24.33	21.33
Na_2CO_3	15	21.14	17.05	25.15	19.22
	30	22.75	18.9	25.31	20.3
$NaHCO_3$	15	22.09	16.44	23.21	17.3
	30	23.29	19.33	23.93	20.4
Hot water	15	22.17	14.22	23.04	15.33
	30	23.17	20.13	23.21	21.19
Tecto	15	20.10	18.55	22.13	18.93
	30	22.05	20.22	24.05	21.32
Control	15	38.88	22.60	40.5	20.83
	30	48.30	31.00	55.55	30.83

* PME recorded zero in all treatments, except in case of KH_2PO_4 and K_2CO_3 in artificial inoculation (recorded 0.1 and 0.8, respectively).

Discussion

Potato (Solanum tuberosum L.) is one of the world's most popular vegetables. Its importance as a crop is reflected in its large- scale cultivation throughout the world. Potato plants are vulnerable to infect by post harvest fungi such as Fusarium and Helminthosporium.

Different treatments in potato tubers, *i.e.* K_2HPO_4, KH_2PO_4, K_2CO_3, $KHCO_3$, Na_2CO_3, $NaHCO_3$, at concentration of (0.6g/l), as well as hot water (45°C/5min) or fungicide (Tecto 45%), recorded decrements in the diseases severity after storage for one, two and three months, compared to the control. Mélanie *et al.*(2002) stated that sodium metabisulfite, sodium carbonate and sodium bicarbonate in preventive application significantly reduced the development of dry rot in potato tuber. Obtained results from this study demonstrate that selected salts can be used to control potato dry rot.

Post harvest treatments lowered the loss in fresh weight of potato tubers during storage with naturally and artificially inoculated with *F. solani*. Frances *et al*. (1988) showed that hot water dip treatments in the range 40–100°C for 2–240 s were investigated for the control of pathological decay in sweet potatoes. It was found that dipping roots at 90°C for 2 s, 80°C for 2, 4 or 10 s, 70°C for 10 s or 40°C for 120 s substantially delayed the time to initial rot without affecting respiration rate or weight loss. Also, Naffa *et al*. (2003) reported that hot water treatment at 45^0C for 5 min reduced the weight loss of green onion plants either naturally infected or artificially inoculated with *B. allii* during the cold storage for 4 weeks. Also, Naffa and Rabie (2006) found that Na_2HPO_4 and $KHCO_3$ with packaging reduced the loss in fresh weight of cucumber fruits compared to the untreated fruits.

Salts, hot water and thiobendazole (Tecto) fungicide treatments increased the moisture content in naturally infection and artificially inoculation with *F. solani*. Naffa and Rabie (2006) show that $CaCl_2$, Na_2HPO_4 and $KHCO_3$ increased in naturally and artificially inoculation of *Botrytis cinerea* compared to the untreated (control).

The activities of PG and CX enzymes were higher in artificial inculcated potato tubers with *F. solan*i than the natural infection. Also, increasing the reaction time from 15-30 min., raised the activities of PG and CX enzymes gradually to be higher at 30 min., than at15 min. All post harvest treatments on potato tubers both artificial and natural infection decreased the activities of PG and CX enzymes compared with the control. Similar results were obtained also by (Nigro *et al.,* 2006) suggested that pH and inhibition of polyglacturonase activity of *B. cinerea* seem to play a role in the mode of action of sodium bicarbonate, sodium carbonate, potassium carbonate and calcium chloride. While, Ahmed (2010) found that the loss% in viscosity was increased by increasing the reaction time from 15 to 30 min.

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مكافحة العفن الجاف على البطاطس أثناء التخزين

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تصاب درنات البطاطس بمرض العفن الجاف المتسبب عن الفطر *Fusarium solani* أثناء التخزين والتسويق ويهدف هدا البحث الى اختبار أملاح فوسفات البوتاسيوم الأحادى والثنائى- كربونات وبيكربونات الصوديوم - كربونات وبيكربونات البوتاسيوم والماء الساخن والمبيد الفطرى التكتو فى مكافحة مرض العفن الجاف المتسبب عن الفطر *Fusarium solani* أثناء التخزين المبرد لمدة ثلاثة أشهر(على درجة ٧ °م لمدة شهر ثم التخزين على درجة حرارة الغرفة لمدة شهرين عند رطوبة نسبية ٩٠-١٠٠%) .واتضح من النتائج أن جميع المعاملات بالاملاح للدرنات غير المحقونة بالفطر أدت الى كفاءة ١٠٠% عند التخزين لمدة ثلاثة أشهر فيما عدا المعاملة بالماء الساخن كانت ٧٥.٠٢ ،٥٩.٩٧% لموسمى ٢٠١١ و ٢٠١٢ على التوالى. وحافظت كل المعاملات على جودة الدرنات من حيث اطالة عمر الدرنات بتخزينها لمدة أكثر من ثلاثة أشهر على درجة حرارة الغرفة وأيضا انخفض الفقد فى الوزن وحدث زيادة المحتوى الرطوبى لدرنات البطاطس وقلة التزريع مقارنة بالكنترول الغير معامل وأيضا وقللت نشاط الانزيمات المحللة البولى جلاكتورنيز والسيلوليز سواء للدرنات الطبيعية (بدون عدوى) أو الدرنات المحقونة بالفطر *Fusarium solani* واتضح من النتائج أيضا انة لا يوجد نشاط لانزيم بكتين ميثيل استريز للدرنات الطبيعية أو الدرنات المعدية المعاملة بالاملاح فيما عدا ملحى فوسفات البوتاسيوم الثنائى- كربونات البوتاسيوم كانت (٠.٨،٠.١٠) على الترتيب.