

Evaluation of Some Plant Materials for Anti Microbial and Sprout Inhibition in Stored Tubers of Irish Potatoes (*Solanum tuberosum*. L)

L. Onyishi

Department of Botany, University of Nigeria, Nsukka

Abstract

Aqueous extracts and dust of *Mucuna pruriens*. Baker, *Bryophyllum pinnata*. Kurz, *Vernonia amygdalina*. Hook, *Azadirachta indica*. Juss; as well as ash from male and female inflorescences of oil palm and aqueous extract of potato peels were studied for their anti-sprout inhibition and anti-microbial properties on Irish potato tubers. In aqueous extract of potato peels 8.3% of tubers rotted as against 29.8% for control tubers after six months of storage. The use of dust rather than aqueous extract of plant material reduced tuber rots better. All the plant materials indicated anti-microbial and sprout inhibition properties compared to untreated tubers and at ($p = 0.05$), significant differences existed between one plant material and another in rot reduction. Aqueous extracts of potato peels reduced tuber sprouts to 3.2% as against 16.8% for control tubers. In all cases of storage Batata tubers sprouted and rotted more than Diamant tubers. *Rhizopus stolonifer*, *Phoma foveata*, *Fusarium oxysporium*, and *Pseudomonas solanacearum* were found to be associated with tuber deterioration in storage. Each of the isolated pathogen was able to reproduce rot symptoms characteristic of the organism in pathogenicity studies.

Introduction

The tuber forming species of *Solanum* (*S. tuberosum*) and (*S. andigena* L) are important vegetable crops grown in most parts of the world. Potato is a universal article of diet and it is reorganized as food with easily digestible starch, some essential amino acid such as lysine and useful minerals such as potassium, magnesium and phosphorus (Barry-Ryan and O-Beirne, 2003). Potato combines the typical characteristics of vegetables and staple crops, high mineral and vitamin content, an excellent protein quality and a good energy value.

Potato eaten within a month after harvest contains even more vitamins than apple (Finglas and Faulk, 1984). The biological value of its protein is higher than that of most vegetable protein and approaches that of chicken eggs (Gray and Hughes, 1978).

A consumer of potato gains mineral and vitamins as bonus in relation to other staple food crops such as wheat, rice, maize or yam. According to FAO (1989) statistics show that potato nutritional energy yield per hectare is higher than that of any other crop grown in developing countries. This is the main reason for its rapid expansion in cultivation.

A secondary advantage is its growth cycle. In many parts of world it has provided famine relief during adverse periods such as wars. For 4-5 days, per week potato products are consumed in Europe. It was a cheap source of food for the labouring classes and contributed substantially to Europe's industrial revolution (Salaman, 1937). In Nigeria potato is grown in most Northern states such as Kano, Yobe, Kaduna, Taraba, Adamawa, Jos and Bauchi as the most efficient tuber crop in Nigeria in terms of tuber yield and days to maturity. Tuber yields of up to 15-30 tones have been reported under Nigerian conditions, some obvious problems associated with rural agriculture notwithstanding (Okonkwo *et al*, 1986).

A major limiting factor in the adoption of potato as a popular family crop in different parts of

Nigeria is related to its storage problem. Microbial rotting and sprouting of tubers discourage potato users from bulk purchase since potato is properly accepted as a crop that does not generally store very well. Okonkwo *et al* (1995) noted that most potato tubers harvested in Jos are sold as soon they were harvested for fear of tuber rots.

In developed countries facilities are available for cold storage and damage to tubers as a result sprouting and microbial deterioration is reduced. Potatoes tubers are known to harbor varying degrees of latent rot pathogens that may initiate rotting process when tubers are kept in conditions favourable for their growth (Elphistone and Perombelon, 1986). Most tuber crops are stored in conditions that predispose them to post harvest pathogens (Udo *et al*, 2000).

For potato unlike some other crops, information on conventional storage methods that could reduce microbial deterioration as well as sprouting where controlled environmental conditions are not available is scarce. Inorganic anti-microbial agents are popular as tuber protectants in micro storage research works. The cost, availability, and health implication makes inorganic chemicals generally unacceptable for protecting edible food crops. On the contrary, recent research efforts have focused on the evaluation of safe biological materials for their suitability as antifungal and bacterial organisms. (Uzuegbu and Okoro, 1999; Udo *et al*, 2000; Maduegbu *et al*, 2000). Information is however scarce on the use of herb extract as sprout inhibitors in tuber crops especially in table potato tubers where sprouting poses serious storage problem.

The purpose of this work is therefore to evaluate the suitability of different preparations of plant materials as anti sprout and anti microbial agent for Irish potato tubers in storage and also to identify the micro organisms that may be associated with potato tuber storage in Nsukka.

Materials and Methods

Preparation of herb extracts: This was adapted from the description given by Uzuegbu and Okoro (1999) and Owolade and Osinkanlu *et al* (1999). Fresh leaves of neem,

Azadirachta indica, *Vernonia amygdalina*, *Mucuna pruriens* and *Bryophyllum pinnata* were collected. For each plant materials 30g of leaves were washed with sterile water and ground separately in a mortar after which 1000mls of water was added to each sample. The mixture in each sample was pressed out using separate muslin cloth for each plant material. The extracts were concentrated by centrifuging at 1000rpm for 15 minutes in a rotary centrifuge. Diamant and Bateta tuber varieties were dipped in the extract to obtain as much coverage of tubers by plant extract as possible. Fifty tubers of each potato variety was used for each plant material. The tubers were left with the plant materials for 3 hours before they were removed to dry at room temperature for 6 hours. They were later kept in traditional baskets in a storage shack constructed in the Botany Garden, University of Nigeria Nsukka from July to December 2001.

Preparation of dust from plant material: This was obtained according to the methods described by Olufolaji (1999) in related studies involving other plant materials. For each plant materials, 30g of leaves was sun dried for seven days. Each dry leaf sample was ground in a mortar separately to obtain the dust for each plant material which was applied on potato tubers ensuring as much coverage of tubers by dust as possible. The tubers were also stored in baskets as previously described.

Obtaining ash from male and female inflorescence of oil palm: Bunches of male and female inflorescence of oil palm were burnt separately to obtain 30g of ash from each specimen. The ash samples were applied on tubers and stored just as in plant dust preparation above.

Producing water extracts of potato peel: Healthy potato tubers were collected, washed and rinsed in sterile water. A sterile scapel was used to peel the tubers to a depth of about 4mm to obtain 50g of potato peels. The peelings were soaked inside 2 liters of water in a bath for 12 hours. Muslin cloth was used to filter the peels. Potato tubers were soaked in the extracts as in the method used for herb water extract.

Isolation of pathogenic organism from diseased tubers: Diseased tubers portions adjacent to health ones were excised using a sterile scalpel. They were washed in domestic bleach for one minute and rinsed in sterile water at three changes. They were later plated on water agar. Any growth in water agar was later transferred into PDA in Petri dishes for growth, development and identification.

Plant extract-tuber-fungal inoculations: Cylindrical cores 15mm long were removed with a 4mm diameter sterile cork borer from the center

portion of tubers. About 4mm disk of a week-old culture of the micro organism was plugged in the holes to which plant water extract or dust had already been dropped from the potato tissue bored from tubers 2mm pieces of tissue was removed. This was replaced in the holes in the tuber. They were sealed with Vaseline and kept on laboratory benches for observation.

Results

Effect of plant material on percentage tuber rots: The lowest rate of tuber rots (8.3%) was recorded with tubers that were dipped in aqueous extract of potato peels before storage. This was followed by treatment with neem as dust (11.3%) and then *Bryophyllum* and *Vernonia* dust reduced tuber rots at a rate that did not differ significantly from one another ($P = 0.01$). This difference existed for one and all the other plant materials tested. Tuber rots progressed in all cases as tubers were kept through the months (Table 2 and 3).

Effect of plant materials on the sprouting of tubers: In all cases involving herb water extract and dust, sprouting of tubers was reduced when herbs were applied as dusts (Table 4). With water extract of potato peels only 3.2% of the tubers sprouted, while ash from male and female inflorescence of oil palm (6.8%) and (5.9%) respectively reduced tuber sprouts appreciably. Control tubers sprouted freely (16.8%) throughout the period of storage.

Organisms associated with tuber rots: *Rhizopus stolonifer*, *Phoma foveata*, *Fusarium oxysporium* and *Pseudomonas solanacearum* were isolated from different forms of tuber rots. All were able to reproduce rot symptoms characteristic of each one in pathogenicity tests. *Phoma foveata* induced the least tuber rots in terms of rot diameter for both Diamant (3.78cm) and Betata (4.45cm) compared to other organisms (Table, 5).

Discussion

The use of various formulations of *Azadirachta indica*, *Bryophyllum pinnata*, *Mucuna pruriens* and *Vernonia amygdalina*, *Ixora divaricata*, *Citrus aurantifolia* as anti microbial agents has previously been reported on other crops (Olufolaji, 1999; Maduegbu *et al*, 2000 and Udo *et al*, 2000). Injury to tuber crops in storage is traditionally protected from getting diseased by wood ash. (Eze and Maduegesi, 1989; Uzuegbu and Okoro, 1998). The use of biological materials as anti microbial agents and as sprout inhibitors have good promises as against synthetic chemicals protectants from the point of view of health and environmental implications.

Potato tuber rot and sprouting reported in this work were reduced with the application of water extract of potato peels probably due to the explanation given by Bennet-Clark and Kefford (1953). They noted that a water soluble anti-sprout "inhibitor B-complex" exists in potato peels as in the leaves.

Table 1: Effect of plant materials (PM), potato variety (PV) and PM x PV interactions on the percentage rot of Bateta and Diamant tubers stored after treatment with different preparation of plant materials

Methods of storage	Diamant	Bateta	Means
Neem extract (A)	15.3	18.7	17.00
Neem dust (B)	10.5	12.2	11.35
Vernonia extract (C)	21.3	29.5	25.40
Vernonia dust (D)	14.2	15.1	14.65
Bryophyllum extract (E)	22.1	27.7	24.90
Bryophyllum dust (F)	11.3	18.3	14.80
Mucuna extract (G)	21.9	28	24.95
Mucuna dust (H)	15.9	18.9	17.40
Ash of male inflorescence of oil palm tree (I)	17.1	20.3	18.70
Ash of female inflorescence of oil palm tree (J)	16.8	23.7	20.25
Water extract of potato peels (K)	7.2	9.3	8.25
Control (L)	26.3	33.5	29.90
Means	16.66	21.27	18.96

LSD (0.05) comparing: Plant materials (PM) = 0.107, Potato variety (PV) = 0.044, PM x PV interaction = 0.151

Table 2: Effect of months of storage (MS), potato variety (PV) and MS x PV interactions on the percentage rot of Bateta and Diamant tubers stored after treatment with different preparations of plant materials

Months of Storage	Variety		Mean of Months
	Bateta	Diamant	
1	0.004	0.004	0.004
2	5.8	2.7	4.1
3	18.9	15.7	17.2
4	34.9	29.4	34.2
5	43.9	36.9	40.5
6	48.5	40.9	44.7
Mean of variety	20.7	16.2	18.3

LSD (0.01) comparison, MS = 0.107, Variety = 0.044, MS x PV = 0.371

Table 3: The interaction effects of plant material (PM) by months of storage (MS) by potato variety (PV) on the percentage rot of Bateta and Dimant potatoes stored after treatment with different preparations of plant materials

Variety	Months	Plant Materials*											
		A	B	C	D	E	F	G	H	I	J	K	L
Diamant	1	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	2	5.70	1.60	2.20	2.00	5.30	0.50	2.20	3.90	4.60	2.00	0.50	4.60
	3	17.30	5.90	24.00	20.70	19.90	9.90	24.00	13.90	13.90	16.90	3.80	28.00
	4	25.30	19.90	37.90	23.30	44.70	20.60	43.30	29.90	31.30	30.30	9.70	49.30
	5	30.60	26.60	50.00	28.70	48.60	29.30	50.10	35.30	34.70	39.90	21.90	56.70
	6	33.30	29.30	53.90	33.30	50.80	31.30	54.70	37.90	45.30	43.90	24.00	63.90
Bateta	1	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	2	7.90	3.80	9.30	7.30	7.30	4.60	7.30	3.90	3.80	8.70	1.20	14.60
	3	24.60	8.70	29.90	26.00	26.00	17.30	27.30	17.90	22.60	17.20	8.80	30.60
	4	29.90	22.60	56.70	53.90	53.90	35.90	54.70	38.60	38.50	47.30	15.90	39.40
	5	33.40	26.60	60.60	60.00	60.00	38.60	60.00	39.90	43.30	50.80	21.30	68.10
	6	38.60	30.60	65.30	63.90	63.90	41.40	63.90	43.90	46.70	55.30	25.30	79.20

Plant materials: See Table 1 for interpretation, LSD (0.05) comparing: Method of storage x Potato, variety x Months interaction = 0.371

This "inhibitor B-complex" is described as an amylase inhibitor and it brings about its inhibitory effect by uncoupling phosphorylation from the electron transfer systems and by this means deprive the tissue of energy (ATP) necessary for the performance of synthetic reactions associated with growth. Its specific effect on rot pathogens was however not indicated.

The organisms isolated from diseased potato tubers are regular tuber pathogens. *Rhizopus stolonifer* is associated with rot of cocoyam corms in Nsukka (Eze and Maduwesi 1990). *Pseudomonas solanacearum* is an early disease of potato causing soft rot of tubers in the field as well as in storage or transit (Smith and

Ramsey 1947; Okonkwo, *et al*, 1995). *Fusarium* cause dry rots on potato tubers. Dry rots are usually masked among tubers because of absence of wetness as in *Rhizopus* and bacterial soft rots. *Phoma foveata*, and *Fusarium oxysporium* have previously been reported on potato and other crops (Croke and Logan. 1982; Tindall, 1993).

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Table 4: Effect of plant materials (PM), potato variety (PV) and PM x PV interactions on the percentage sprouting of Bateta and Diamant tubers

Plant Materials	Variety		Mean of plant materials
	Diamant	Bateta	
A	8.3	11.7	9.93
B	7.2	10.9	8.9
C	9.4	12.1	10.6
D	7.4	8.1	7.7
E	10.4	11.6	10.9
F	7.8	8.9	8.4
G	10.3	11.7	10.9
H	9.2	8.6	8.9
I	6.0	7.7	6.8
J	4.6	7.4	5.9
K	2.3	4.3	3.2
L	14.7	19.1	16.8
Means	7.8	9.9	8.9

LSD (0.5) Comparing: Plant Materials (PM) = 0.128; Potato Variety (PV) = 0.052; PM x PV interactions = 0.181

Table 5: Effects of organisms (OG), potato variety (PV) and OG x PV interaction on rot severity in (cm) of Bateta and Diamant tubers.

Organisms	Diamant	Bateta	Means
Fusarium	4.14	4.94	4.54
Phoma	3.78	4.55	4.17
Pseudomonas	3.84	4.71	4.28
Rhizopus	4.07	5.08	4.58
Means	3.96	4.82	4.39

LSD (0.05) comparing: organisms (OG) = 0.053; Potato variety (PV) = 0.038; and OG x PV interaction = 0.151

Table 6: Effect of plant extract (PE), potato variety (PV) and organism (OG) and PE x PV x OG interactions on rot severity in (cm) of Bateta and Diamant tubers.

Variety	Plant leaf extracts	Organisms				Mean
		Fusa	Phoma	Psuedo	Rhizo	
Bateta	<i>Bryophyllum</i> extracts	4.87	5.2	5.2	5	5.07
	<i>Bryophyllum</i> powder	4.87	5.37	5.0	4.47	4.85
	<i>Mucuna</i> extract	5.57	5.2	5.0	5.07	5.21
	<i>Mucuna</i> powder	5.0	4.6	4.83	5.43	4.97
	Neem extract	3.55	2.37	2.73	3.4	3.01
	Neem powder	3.17	2.0	2.57	3.03	2.69
	<i>Vernonia</i> extract	3.4	2.93	2.83	3.13	3.07
	<i>Vernonia</i> powder	3.0	2.4	2.57	3	2.74
	Means	4.12	3.78	3.84	4.07	3.95
Diamant	<i>Bryophyllum</i> extract	5.1	6.27	6.53	5.57	5.87
	<i>Bryophyllum</i> powder	5.0	6.0	6.0	5.57	5.64
	<i>Mucuna</i> extract	6.77	6.7	6.6	6.7	6.69
	<i>Mucuna</i> powder	5.87	6.0	6.17	6.0	6.01
	Neem extract	4.2	3.0	3.17	4.3	3.67
	Neem powder	3.8	2.8	2.6	3.6	3.20
	<i>Vernonia</i> extract	4.8	3.1	3.6	4.7	4.05
	<i>Vernonia</i> powder	4.0	2.53	3.0	4.2	3.43
	Means	4.94	4.55	4.71	5.08	4.82

LSD (0.05) comparing: Plant extract (PE) = 0.063; Potato variety (PV) = 0.043; Organism (OG) = 0.083, and PE x PV x OG interactions = 0.055

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