The Identification of Potato Spindle Tuber Viroid on Potato and Tomato Production in Libya

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***Abstract*—** This study aimed to reveal the presence of potato spindle tuber viroid (PSTVd) on potato in Libya. Samples Collected during autumn 2005 from the regions of the AL- Maamoura and Sabratha, the apparent symptoms on samples of potato plants were reduced leaf and size curling downword, while tubers were showing cracks and they were spindle shaped sprouts surrounded by swollen tissue like protruding eyebrows. Were the causative agents of these symptoms confirmed as potato Spindle tuber viroid (PSTVd) by using molecular hybridization of the nucleic acid and polymerase chain reaction (PCR) assays. The Libyan isolate of PSTVd was similar to the Egyptian and to the American isolates, because its nucleic acid contained 360 bp as well, and its movement appeared at the same level as the other two isolates in electrophoresis upon agar slices. And this is the first recording of the potato spindle tuber viroid of potatoes in Libya.

***Keywords*—** Viroid , Symptoms, Potato ,Tomato ,Libya.

I. Introduction

IT may be concerned that tomato plant Lycopersicon esculentum Mill which belong to Solanaceae , is very important as agriculture product used for food . and it contains food elements such as calcium, iron, carbohydrates, potassium, phosphorus, and some vitamins such as vitamin (A and G) . The percent of water in the fruit is 93-94% from the weight of fruit (Abugniah, 1998, Laroussi, 1993, Abd Aal, 1998, Abdel Moneim, 1988, Saleh Saeed, 1990, Alawi, Abdel Moneim, 1987), also the fruit contains the lycobin which is used as antioxidation and tumer disease control (John,Airdman, the global network of information research sites).

The tomato plant is an important crop in Libya , in addition the cultivated area in 2004 , was ten thousands of hectars and the production was 90 thausands tons (Yearbook of Agricultural Statistics, 2005).

The tomato plants infected with many pests like insects and diseases of, fungi, bacteria, viruses, viroids (Abuarqhob, 1996, Agrios and George, Plant Pathology, 1994, Robert, Daniel. 1986, Awad, 2005). The imported viroids which infect tomato plants were potato spindle tuber viroid (PSTVd), tomato planta macho viriod (TPMVd), tomato apical stunt viroid (TASVd), they differ in their severity, (Abuarqhob, 1996). Its known for many years that potato spindle tuber viroid (PSTVd), was caused severe loses in tomato plants, for this reason some tomato cultivars used as a test plants such as Rutegers, Sheyenne, and Rentia (Kryczynski and Stawiszynska 1980).

In Libya, there is no previous studies about the presence of potato spindle tuber viroid (PSTVd) on the potato, and its effect on the growth and production of tomato plant varieties inside the plastic greenhouses and in the fields, especially on the commercial tomato varieties which are imported to Libya and which are locally developed .

So, this study aimed to identify potato spindle tuber viroid (PSTVd) on potato crop in Libya and its influence on the growth and production of some commercial tomato varieties under the Libyan conditions and to find some resistance cultivars.

II. Materials And Methods

A. Samples collection and preservation

Four samples of potato tubers were collected from the area of Almaamoura, and a sample from the area of Sabratha, the symptoms were similar to symptoms of PSTVd on Potato, tubers were spindle shape and covered with a large number of prominent eyes, and some tubers were distorted and the cracks were deep (Figure 1), during the autumn planting season for 2005, note that the samples of the cultivar Spunta, and the source from the Netherlands, and placed in the refrigerator until tested.

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Fig. 1 Show spindle shaped and distortion and cracking of the Libyan potato tubers

B. plant response of Scopolia sinensis Hemsl for the Libyan isolate of PSTVd.

1. The source of seeds and planting:

The seeds of a plant Scopolia sinensis Hemssl were brought from the University of Ain Shams, laboratory of plant viruses, 12 pots were filled with sterile soil in the greenhouse and 3seeds in any pot were planted .

2. The preparation of inoculum of Libyan isolate of (PSTVd):

The inoculum was obtained and prepared after the cultivation of infected Libyan potato tubers planted in pots and get the shoots, 5-10 g of leaves of new growths were ground, in the mortar and in the presence of buffer solution phosphate 0.1 Muller and pH 7 using 5-10 ml, and juice was passed through a piece of double sterile gauze to get rid of plant tissue, and placed in the petri dish and adding to Article Alcarburandem (carborandum), and use the juice obtained in the process of mechanical inoculation.

3. Mechanical Infection:

7 plants were inoculated mechanically by index finger on 30-12 - 2006, and 5 plants were left as a witness (control).

4. The process of irrigation and the use of pesticides:

The plants have been irrigated once every three days, and Alkonfodor pesticide was used to kill insect tunnels Makers.

5. Registration of symptoms:

Symproms recorded on the plants after a month of infection.

C. Study the sensitivity of tomato cultivars to the Libyan (PSTVd) isolate.

The same steps to the second stage were followed in the preparation of the greenhouse, planting seeds, and design of planting seedlings and preparation of the inoculum, and an infection, irrigation, and use of pesticides ,the mechanical inoculation of tomato varieties was carried on 31 - 12-2006 , Folkato Thuria Sankarh, Lebda, Hanaa , Jasmine,and Kenza, the number of treated plants was 10 from each cultivar and control 10, and symptoms recorded after 25-35 days of inoculation, the percentage of occurrence of the disease was calculated according the following equation:

The percentage of disease incidence =

$$\frac{\text{number of plants showed symptoms} \times 100}{\text{The total number of treated plants Detection of viroid (PSTVd)}}$$

The presence of infection with the viroid PSTVd was detected in potato tubers collected from Libya, as well as in tomato cultivars treated with Egyptian isolate such as (Lebda, Kenza, Jasmine, Super Halim, and variety 185) At the Agricultural Research Center, Department of viruses, Cairo - Egypt, the Nucleic Acid Hybridization, as well as Polymerase Chain Reaction (PCR) were used.

First – extraction of the nucleic acid from the leaves of tomato and potato tubers: -

The total nucleic acids was isolated from the leaves of tomato plants infected with PSTVd and infected potato tubers as described in the institution method of Bromeja, Madison, United States of America, and the way the world Chamol and others as follows:

1- 175 micro liter decomposition of a solution of SV RNA regulator with the beta - Mercapto ethanol added in a sterile tube and was weighed.

2 - Plant tissue was transferred to the tube and was mixed well by flipping and weighed, the ratio of tissue to solution decomposition acidity regulator is 30 mg / 175 ml.

3 - 350 ml of a solution of SV RNA was added to reduced acidity regulator and then mixing by flipping 3-4 times.

4 - The tube was put in a water bath (Figure 2) at a temperature of 70 ° C for 3 minutes, then the tube was put again in the slow speed centrifuge \ (Figure3) for 10 minutes at the speed of 16 thousand attractive, then the suspended water was transfer to the other clean tube by drag Pipette to get rid of sludge.

5 - 200 micro liters of 95% ethanol were added to the aqueous suspension, and mixing by pumping Pipette 3-4 times

6 - The mixture transferred to a rotating basket and was run at speed of centrifuge at 14 thousands attractive for one minute, to get rid of the water and keep the suspension.

7 – 600 micro liters solution of washing SV. RNA was added to (with added ethanol), and the tube was put in the centrifuge at the speed of 14 thousands attractive for one minute, and disposed of the pending water.

8 - A mixture consisting of incubator yellow concentrated solution of acidity regulator (40 micro liter) and manganese dichloride MnC12, 0.09 Molar (5 micro liter) and the enzyme DNase (5 micro liter) was added and has been mixing, these components were mixed by gently sucking.

9 - 50 micro liters of this solution were added directly to the membrane inside the basket, and the membrane was incubated at room temperature for 15 minutes, the solution of 200 micro liter of SV. DNase (with ethanol) was added to the membrane,

and placed the basket in the rotating centrifuge at speed of 14 thousands attractive for one minute.

10 - 60 micro liter of washing solution SV. RNA were added, and rotating basket was placed again in the centrifuge at the speed of 14 thousands attractive for one minute.

11 - Collection tube was empty and 250 micro liter of a solution of washing SV. RNA were added and placed in a centrifuge at the speed of 14 thousand attractive for two minutes, and then was transferred from the basket rotating tube to tube assembly endorsement.

12 - 100 micro liter of free water of Nuclease were added to the membrane, when it was placed in a centrifuge at speed of 14 thousands attractive to one minute for the endorsement acid RNA, then the purified RNA acid storage at a temperature of 70c.

Fig. 2 Shows the water bath used in the experiment

Fig. 3 Shows the centrifuge used in the experiment

Second - The creation of complementary DNA sequences (cDNA):

1 micrograms of TNA (Total Nucleic Acids) added to each sample and 1 microgram of the initial piece of viroid PSTVd (primer) (5ccctgaagcgctcctccgag3) and a piece of an initial debate (5atccccggggaaacctggagcgaac3), and 6 micro liter of a solution of Tris hydrochloric acid penta concentration (250 mM of Tris-HCL 8.3: 375 mM potassium chloride KCL, 15 mM solution of magnesium chloride MgCL2, and 3 micro liter of 0.1 Mole of the DI- Thaiotraathol (DDT), and the final volume reached to 30 micro liter water, deionized water, then heating the mixture for 5 minutes at a temperature of 100c, and the response directly in the snow two minutes, then lap at room temperature for 1-1.5 hours to allow Coalescence pieces to RNA template , and mixing 20micro liter of the reaction solution and 4 micro liter of product cDNA thread first structured acidity five concentration (institution bromeja, Madison, USA) and 5 micro liter from 0.3 molar 2 - beta-Mercapto ethanol, and 2.5 micro liter of 10 mM of each deoxy Nyukluotad triphosphate (dNTPs), and 1 micro liter of the enzyme RNase (40 units / micro liter, institution bromeja, Madison, USA) and 2 micro liter of 0.1 Molar DTT, and 4.5 micro liter of water deionized and 1 micro liter of enzyme transcrept cloned enzyme reverse viroid to murine leukemia (MMLV-RT) (GIBCO BRL techniques vital Jaydhirsberg, Maryland, USA) with the fusion reaction mixture and incubated at a temperature of 42 ° for 1-2 hours.

Third - polymerase chain reaction (PCR):

Amplification was carried out in thin-walled PCR tubes, each tube contained a mixture of the following reaction: 5 micro liter of PCR solution acidity regulator (1x = 10Mm Tris-HCL PH 8.3; 50Mm KCL and 0.001% gelatin), 3 of 25 micro liter of magnesium chlorid MgCL2 0.1 micro liter of 10 mM dNTPs, 1 micro liter of each of 0.1 of the piece the initial complementary and corresponding to the viroid PSTVd 2.5 units of Tag DNA polymer, and sterile water to a size 45 micro liter cover the mixture drop one of the mineral oil to prevent evaporation and was initially heated to a temperature 85c for 5 minutes in a thermal cycle programmable, 5 micro liter of a combination of cDNA was added to the reaction mixture and PCR amplified using the following measures of recycling:

A – Denaturation on the temperature of 94 c for 3 minutes.

B - Fusion of the initial segment on the temperature of 62 C for 30 seconds.

C - The elongation at temperature of 72 c for 45 seconds.

To amplify the PSTVd used 35-40 with the expansion of a final cycle at a temperature of 72 c for 10 minutes, and the tubes were stored at a temperature of 4c.

D. Analysis of amplified products of RT-PCR:

Separated DNA products amplified by PCR reaction of the electric displacement on the agarose gel was separated (gel-electrophorysis).

1 - 5-10 micro liter from PCR products on agarose gel were analyzed , 1% by the electric displacement through a vertical plate of the gel (11x 14x 0.15 cm) in a solution of (Tri-boric acid-EDTA) TBE) pH on the regulator voltage 100 Volt for one hour.

2 – Gel was dying with methyl Alathediom dye.

3 - The index to acid molecular weight DNA with length of 50 of pair bases to determine the size of cDNA products to

viroid PSTVd amplified by reverse polymerization reaction RT-PCR.

DNA probes was prepared by the index digoxegenen 11-UTP using the interaction of PCR.

A piece of DNA was amplified (a length of 360bp pair bases) by the reaction of PCR from viroid PSTVd isolate that infect the tomato plants using a piece of initial viroid PSTVd, which was used as a template for the synthesis of probes cDNA, and added digoxegenen - 11 - dUTP to synthesized newly DNA during 35 cycles of PCR. the following components were added to the centrifuge tube accurate sterile put on ice during the pumping components Pipette a 0.5 micro liter of a solution of PCR organizer , 0.3 micro liter of a solution of magnesium chloride MgCL2, 5 micro liter of a mixture marking DNTP, 100 ng / micro liter of initial piece of higher flow and down flow of viroid PSTVd, 32.5 micro liter of distilled water, 0.5 micro liter of Tag DNA polymerase 0.5 micro liter of template DNA, the size fuller to 50 micro liter by adding double distilled of water. The reagents mixed and placed in the centrifuge at the speed of 14 thousand attractive for two minutes to collect the sample (sludge) at the bottom of the tube, the sludge was covered with a layer of 100 micro liter of mineral oil to minimize evaporation of the mix, and inflate in a recycle heat, and the measures of rotation of the reaction as follows:

A –Denaturation on the temperature of 94 c for 3 minutes.

B - Fusion of the initial segment at 62 C for 30 seconds.

C - The elongation at temperature of 72 c for 45 seconds.

To amplify the PSTVd used 35-40 with the expansion of a final cycle at a temperature of 72 c for 10 minutes.

DNA hybridization:

E. Hybridization spot bullets:

The result of RNA extracted from viroid PSTVd isolates (using the system to isolate RNA) directly on the spot on the membrane of nylon and added to the size of one of the extracted plant for every three volumes of Saline Sodium Citrate (SSC) 10 concentrations containing 20% (w/v) formaldehyde, then the mixture blending in a mixer and waged at a temperature of 65c for 15 minutes and cooled in crushed ice, and the final extracts are put in form of spots on the, nylon membrane, and linked mutually by ultraviolet light for 45 minutes and then hybridized by PSTVd as follows:

A - The membranes were initially hybridized (for closure) in the hybridization tube at temperature of 50c for a period not less than one hour, and the membranes hybridized by 20 mM per 100 cm 2 from the membranes of the solution hybridization that contains 5-25 nano grams of probe cDNA index by DIG The newly denaturated and heated in a water bath at boiling point.

B - The membranes washed twice for 5 minutes at room temperature for at least 50 ml of SSC double concentration and 0.1% SDS (w/v) for each 100 cm 2 of membrane and twice for 15 minutes at a temperature of 68c by SSC concentration of 0.1, and 0.1% SDS (w/v).

C - The membranes modified in a solution of Genius buffer organizer for one minute at least 20 mL of a solution of pre-hybridization for each 100 cm 2 of membrane, then was eliminated from the solution organizer and 100 ml of a solution of Genius structured second acidity was added and the membranes were incubated for at least 30 minutes.

D- Alkaline phosphatase anti-digoxigenin was deluted to 1:5000 with Genius solution of the second acidity regulator, and lap for 30 minutes.

E- The membranes modified in a solution of the third Genius acidity regulator for two minutes and then revealed immunologically.

F- Membranes incubated for 16 h in 10 ml of the solution to prepare a modern color in a box in the dark (no fluctuation in the evolution of color) to get to the spot densities or tape.

F- The reaction stopped by washing the membranes for 5 minutes, with the amount of 50 ml of distilled water, the results were recorded photocopying for the Leachate or wet photography.

III. Results

Seven selected cultivars of local tomato resistance to tobacco Mosaic virus (TMV) grown in Libya are: Lebda, Kenza, Jasmine, Folkato, Hanaa, Thuria and Sankarh. The seeds of these varieties were planted on 31 -10 - 2006 in fast germinate dishes and packed in artificial soil (Boetmoss) and after a month the plants were grown in the land of greenhouse on 30/11/2006.

The inoculation process by viroid (PSTVd) has been made on 30/12/2006, and symptoms were recorded after 25-35 days of inoculation, also the symptoms on fruits were recorded after maturity.

Weights and lengths for a range of plants from wet and dry were recorded for treated plants and the control for comparison and statistical analysis to determine the impact of viroid on the growth and production of tomato varieties targeted in the study.

Among the objective of the present study is to identify the potato spindle tuber viroid in Libya on the potato crop.

Five diseased samples of potato tubers were collected from farm zones, "the AL- Maamoura and Sabratha during the autumn season of 2005, and the symptoms of virtual were spindle shaped of the tubers, the sprouts surrounded by fabric and shoots like a puffy eye brows prominent.

It has been confirmed that the causal agent of symptoms is a potato spindle tuber viroid by means of a test hybridization molecular of nucleic acid and polymerase chain reaction and was Libyan isolate, the viroid PSTVd similar to the isolation of Egypt and America isolates because its nucleic acid contained a 360bp like them and their movement was at the same level of movement isolates the segments of agar

The bioassay was conducted for the Libyan isolate on test plant of. Scopolia sinensis Hemsl. and the symptoms were appeared within one month of its inoculation.

In another experiment to study the sensitivity of a number

of varieties of tomatoes cultivated heavily in Libya in order to find resistant varieties with the viroid, the study showed clear symptoms in all varieties that have been inoculated.

A. Detection of viroid PSTVd in Libyan potato tubers

Molecular hybridization test:

The prepared extracts from tissues of Libyan potato tubers gave positive signals for hybridization in the samples (Hassan, Ahmed Abdel Moneim, 1988. John,Airdman , the global network of information research site Saleh and Saeed, 1990), the samples of (Abd Aal, 1998; Daniel. 1986) showed Non-hybridization, note that the two samples (Abdel Moneim, 1988. Airdman, the global network of information research sits) from the area of the AL- Maamoura, and the sample number (Saleh and Saeed, 1990) of Sabratha area (Figure 4).

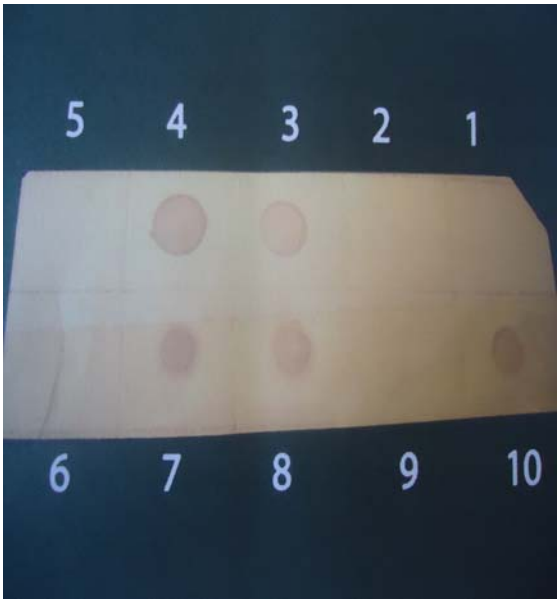

Fig. 4 shows the results of molecular hybridization of nucleic acid, samples of 1-5 leaves of tomato infected with Egyptian isolate varieties of Lebda, Kenza, 185, Soberhalim, Jasmine, respectively, and samples of 6-10 of potato Libyan tubers

The test of polymerase chain reaction PCR

After the removal of the nucleic acid through gel, it was found that the viroid nucleic acid extracted from potato tubers infected with potato spindle, has reached the same level reached by the viroid nucleic acid of potato defined previously in America and Egypt, which contains a 360bp (figure 5) and this shows that viroid nucleic acid which was extracted from samples of potato and tomato was Potato spindle tuber viroid (PSTVd).

A- The response of Scopolia Sainensis to infection for Libyan isolate of PSTVd.

The symptoms showed stunting of inoculated plants (Figure 6).

B-The Response of tomato cultivars to infection by the Libyan isolate of PSTVd

All varieties reacted to the viroid, and showed symptoms on the leaves varied from deformation and yellowing and death of leaf tissue , the small size and dark brown spots , yellow, white, and also varied in the incidence of disease among the tested varieties between 20 to 95% (Table I, plate 1).

Fig. 5 shows the electrical migration of the nucleic acid for potato spindle tuber viroid in the gel using the polymerase chain reaction (PCR), where the M marker, (1) tomato leaves infected with Egyptian isolate, (2) potato tubers infected with Libyan isolate, and (3) non infect tobacco leaf**.**

Fig. 6 shows the symptoms of the plant stunting of *Scopolia sinensis Hemsl*. (Right health, left infected).

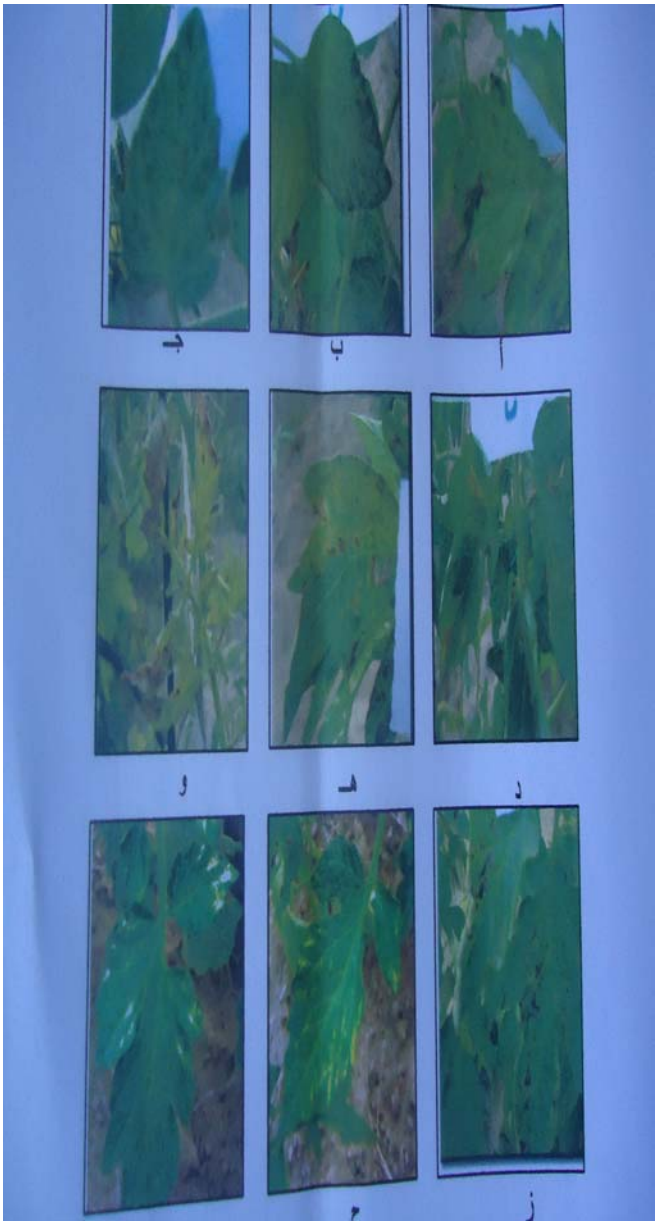

plate (1) shows the effect of the Libyan PSTVd isolate on tomato varieties

TABLE I
SHOWS THE SYMPTOMS OF THE LIBYAN PSTVD ISOLATE ON TOMATOES VARIETIES

Symptoms	% To the occurrence of disease	variety
Leaf distortion and death of tissue (plate a)	95	Felkato
Small size of the leaves and the appearance of dark brown spots (plate b)	80	Thoria
The small size of leaves and yellowing (plate C)	80	Hana
Deformation and death of leaf tissue and wrap up (plate-D)	90	Lebda
Yellowing of leaves and death of tissue (plate E)	95	Sankarh
Death of leaf tissue and yellowing (plate F)	20	Kenza
Distortion of the leaves and the appearance of dark brown spots Note with yellow spots on some plants and then turned to the white spots (plate g, h, j)	90	Jasmin

IV. DISCUSSION

Galindo et al. 1982; Galindo; 1987), (Watler et al. 1980; Watler, 1987) described tomato planta macho viroid (TPMVd) and tomato apical stunt viroid (TASVd) that they separate (two types) and noted that they infect specific areas.

Similarly, a description of the same idea (McClean, 1948) in South Africa on tomato curling top viroid (TCTVd), as well as (Diener et al. 1972) in (PSTVd)

Symptoms for each viroids of the three previously mentioned very similar to the symptoms of potato spindle tuber viroid (PSTVd) from Libya, regardless of some of minor difference not to exceed only the difference in symptoms resulting from the type strain moderate or severe strain (PSTVd), (Kryczynski,. 1986; 1988 and Paduch- Cichal,, 1985).

Sequences of nucleotide to Tomato apical stunt viroid (TASVd) and Tomato planta macho viroid (TPMVd) may be the normal and showed similarity by 73% and 83%, respectively, with the sequence of (PSTVd-RNA) (Kiefer.; 1983).

Identification and classification of all these pathogens may be the subject of controversy and possible to be considered separately (types) or separate strains of the same viroid.

The same situation has stated on potato spindle tuber viroid (PSTVd) and chrysanthemum stunt viroid (CSVd) and cucumbers pale fruit viroid(CPFVd) that cause the same symptoms on tomato and chrysanthemums and some other hosts , in addition to the possibility to use the resistance between these plants (NIblett et al. ; 1978; Paduch-Cichal,, 1985; Kryczynski and Paduch-Cichal,, 1986, 1987), although it is separate viroids (types) on the basis of different installation sequences of nucleotide (Kryczynski and Paduch-Cichal, 1990), and tomato plants may infected with viroids and get all the pathological pattern of one in every case of injury.

The Potato spindle tuber viroid (PSTVd) was identified at 1986 in Poland (Gabriel and Kaczmarek, 1986).

Some tomato varieties are routinely used to detect viroid (PSTVd) (Raymer and O'Brien, 1962a; Singh et al., 1964; Fernow, 1967; Kryczynski and Stawiszynska, 1980).

It is common and well known that the viroids are plant pathogens found in subtropical areas, or in an atmosphere of continental climate (Singh, 1983), and in the tropics are serious pathogens on plants that grow under the covers (Kryczynski ,1992a).

Better interaction between the host plant and viroid during inoculation is when a temperature of not less than 28-30c in the daytime Whitney and Peterson, 1963;, (Raymer and O'Brien, 1962a, Kryczynski, , 1980) .

Under the high light intensity of -least 6000 _ 10.000 lux, as well as high temperature viroids replicated in infected plants (Harris and Browning, 1980; Muhlbach and Sanger, 1977), while high light-intensity help the development of symptoms and the appearance more clearly (Harris and Browning, 1980).

Since viroids active in tropical and semi-warm it for sure that infect plants cultivated in Libya, and in one of the field visits to the regions of the AL- Maamoura and Sabratha during the symptoms of potato spindle tuber viroid, were collected, five samples of potato tubers symptoms represented in the virtual form season of autumn 2005, symptoms on potato tubers look - like the symptoms of PSTVd, they were fusiform tubers, and shoots ,and the buds fabric surrounded by eye brows.

Several experiments were conducted to confirm the presence of viroid (PSTVd) in Libya ,the results proved the following:

- Its confirmed that the cause of these symptoms is the Potato spindle tuber viroid by means of a molecular hybridization test and polymerase chain reaction PCR.
- Test conducted on viroid vital for test plant is Scopolia sinensis Hemsl. , and showed symptoms of stunting, one month after inoculation with Libyan isolate.
- Another study, showed the effect of (PSTVd) on tomato growth by PSTVd – Egyptian isolate, of five tomato varieties mostly cultivated in Libya in the greenhouses.

The results show that the molecular hybridization of nucleic acid (Figure 4) and PSTVd in potato tubers collected from the regions of the AL- Maamoura and Sabratha and the polymerase chain reaction (Figure 5).

The isolation of PSTVd similar to the isolate of Egypt and the USA because the nucleic acid contained a 360bp and the level of migration of nucleic acid in the gel columns at the same level of migration of these isolates.

While tubers were showing cracks and they were spindle shaped, surrounded by swollen tissue like protruding eyebrow(Figure 1), and attributed symptoms form fusiform and cracked tubers that caused by PSTVd due to imbalance in the hormonal system in the infected potato plants, where it was found a decrease in the level of gibrellins and oxins and an increase in cytokynins (EL Dougdug,1989).

After confirming the presence of PSTVd, an other test was carried on the Libyan isolate by bioassay and infection was happed by mechanical inoculation of the Scopolia sinensis Hemsl, The interaction of this plant by PSTVd showed symptoms of stunting (Figure 6), and this is contrary to what is mentioned that it produces local lesions on the leaves (Singh, 1971).

Another experiment has been carried out to search for resistance tomato varieties or immune or tolerant for Libyan isolate of PSTVd and varieties are: (Felkato,thuria, Hanaa, Lebda, Sankarh, Kenza, Jasmine), all varieties interacted and gave varying symptoms Table 1 plate 1.

A difference in symptoms has been found when comparing the symptoms of the disease for the Libyan isolate and symptoms of the disease for the Egyptian isolate, they sharing in three varieties: the thuria, Felkato, Lebda, has been attributed (EL Doudoug, 1989and Singh ,1987), this difference due to the different in pathogenesis ability of two isolates and perhaps to different relay nucleotide sequences , in addition to that potato spindle tuber viroid has two strains , one very moderate, and other one severe strain , and there are some difference between moderate strain and severe strain in three variable nucleotides only, in addition to the difference in the flow rate of migration in a gel way electrophoresis.

V. Conclusion

From the results obtained during the study we recommended the following

1- Further studies for survey to include other areas in Libya

2-Conduct extensive studies to assess tomatoes and potatoes planted in Libya and induce their susceptibility to PSTVd.

3-To prevent mechanically transmission must disinfect pruning buds and planting resistant varieties.

4-Develop standardized programs of quarantine for detection of viroid.

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