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Research Article

Is Ingestion of Potyvirus (PRSV-P) infected Papaya Fruit and bottle gourd safe for Human Health: A Case Study

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Abstract

In recent decades infectivity of viruses across the kingdoms of living beings viz. animals to humans, birds to humans, and plants to humans has raised questions on the host specificity of the viral pathogen within their kingdom. A plant virus particularly Pepper-mild mottle virus (PMMov) causes fever, abdominal pain, and pruritus in patients ingesting pepper infected with PMMoV. Certain plant viruses-infected fruits, vegetables, and food products that are ingested by humans, in the absence of the proper knowledge of plant viral infection, such plant viruses are detected in human cells and excretion. Not all such plant viral ingestion are researched for their effect on human health and for clinical symptoms. A Potyvirus (PRSV-P) infecting Papaya and Cucurbitaceae family plants and exhibiting typical symptoms of ring spots and blisters on the infected fruits are consumed and ingested by humans, however; their ingestion effect has not yet been studied and reported. The present case study was therefore undertaken to assess the effect of ingestion of PRSV-infected fruits on human health. It is observed that PRSV-P-infected papaya and bottle gourd were safe for human consumption without having any clinical symptoms on human health.

Keywords: clinical symptoms; potyvirus; infected papaya; infected-bottle gourd; human health

Introduction

It was believed that plant viruses only infect plants and can not cause disease in other living beings was a myth because plant viruses in three families viz. Bunyaviridae, Rhabdoviridae, and Reoviridae are known to infect plants, animals, and humans. Colson et.al. (2010) indicated that pepper mild mottle virus (PMMOV), a plant virus might infect human beings which triggers other researchers to conduct further studies to re-evaluate the dogmatic concept that plant viruses are safe for human health as numerous plant viruses infected fresh fruits, vegetables, and food products are ingested by humans. Plant viruses are highly prevalent in wild and cultivated plants including fruits and vegetables. For example, it was found that approximately 60 % of plants in a geographical area of Costa Rica (encompassing a total of 7000 plant species) harbored plant viruses (Wren et. al,2006). A classical example is of Tobacco Mosaic Virus (TMV) which infects over 150 plants including tomatoes, peppers, and cucumbers (Hu et.al, 2011). Tomato bushy stunt virus (TBSV), a tombuvirus, is an important pathogen of tomato and other plants and was observed to reach a concentration of approximately 200 mg/kg of infected leaves in the experimental host such as Nicotiana clevelandii (Tomlinson et. al,1982). Moreover, this virus can remain infectious despite being frozen for several years and its thermal inactivation point is 80-90 0C (Hollings and Stone,1965). Some of these virus-infected plant's produce with masked virus symptoms are consumed raw as fruits or in the form of salad. Some plant viruses are also present in food products as detected for pepper mild mottle virus (PMMoV) whose RNA was in 57 % of 28 pepper-based food items and demonstrated that PMMoV in these food items was still able to induce plant infection (Colson et.al, 2010). Several researchers (Rebolledo-Mendez et. al,2013., Mondal and Jain, 2010., Balique et.al, 2015 and Parrish et.al, 2008) raise the question of whether plant viruses can cross the kingdom barrier to infect humans. The clinical symptoms induced by such plant viruses are not fully studied.

Plant viruses are vibrant pathogens infecting several crops including fruits and vegetables. These viral-infected fruits and vegetables are sold in the market at lower prices and many people who are unaware of the symptoms of virus infection, tend to purchase such fruits and vegetables at a lower price. Whether such viral-infected fruits and vegetables have any clinical significance on human health is not much studied. In the present case study, a Papaya ring spot virus (PRSV-P) infected papaya fruits and bottle gourds were assessed for its adverse effect or clinical symptoms in humans.

Material and Methods

1.Collection and analysis of data of PRSV-P infected Papaya fruits and Bottle gourds from fruit/vegetable vendors and case study participants.

The papaya fruits and bottle gourd vegetables showing typical symptoms of papaya ring spot virus on papaya (Fig.1) and bottle gourd (fig 2) were procured (25 each in number) from a concerned vendor and were asked to give these to the customers free of cost, who were desirous to buy these. These vendors were unaware of the cause of the symptoms on the infected fruits and thought it may be due to environmental effects or some other unknown factors and sold such infected fruits at lower rates as a usual

practice. These vendors were asked to participate in the case study to furnish the effect of the sold infected fruits to customers if they report back any effect on their health after consumption of the sold fruits.

Twenty-five persons/participants were analyzed for the effect of or clinical symptoms of PRSV-infected papaya fruit ingestion while the same number of persons were analyzed for the effect of or clinical symptoms of PRSV-infected bottle gourd consumption as a case study. Here again, the participating individuals were unaware that they were provided with the PRSV-infected fruits for their consumption even though the viral infection symptoms were apparent on the purchased fruits. This is because these persons were regular purchasers of such fruits without any hesitation due to unawareness of the viral infection of the fruits. These participants in the case studies were asked to report back to the vendor for collecting their token gift (of fruits) within 48 hours of consumption of the supplied fruits. The participants reporting back for the token gift were asked the questions viz. how was the test of the fruit, whether these had any unpleasant smell or test, and whether they noticed any clinical symptoms after the ingestion of the fresh fruit or cooked vegetables. After getting the requisite information from the participants, they were informed about the aspect of the study and were made aware of the symptoms of the PRSV on the infected papaya fruit and bottle gourd vegetable (that were being purchased by the consumers due to their unawareness of PRSV-P infections on the papaya fruits and bottle gourd vegetables).

a. Symptoms of papaya ring spot virus (PRSV-P) on papaya fruits:

Papaya ringspot is a destructive disease of papaya plants and fruits characterized by a yellowing and stunting of the crown of papaya trees, a mottling of foliage, shoe-stringing of younger leaves, water-soaked streaking of the petiole (stalks), and small darkened rings on the surface of the fruits (fig.1.a). The viral-infected fruits exhibit bumps and the classic 'ring spot" on the fruit's surface. However, the infected fruits did not show any unpleasant symptoms in the fruit flesh when cut open (fig.1.b) and further, there is no difference in the taste of ripened flesh when compared to non-infected fruits.

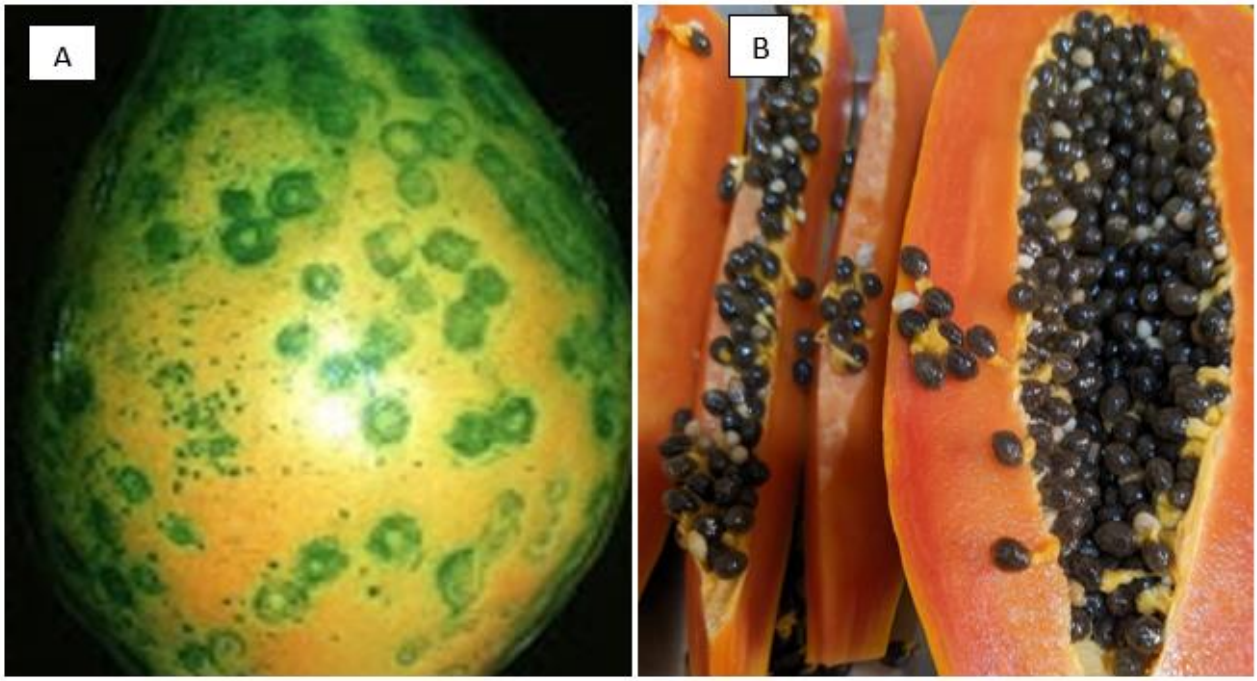

Figure 1: Papaya ring spot virus infected papaya fruits.

A= Ring spot symptoms on papaya fruits, B = internal ripped flesh

b. Symptoms of papaya ring spot virus (PRSV-P) on bottle gourd vegetable fruits:

The virus isolates produced characteristic symptoms of mosaic, mottling, interveinal chlorotic bands, and leaf distortion in the virus-infected bottle gourd plants while malformation of bottle gourd fruits, and reduction in fruit size are observed in such infected plants. In severe infections, blisters on the fruit surface commonly occur (fig 2.a). The said virus produced systemic mosaic mottling symptoms in 11 cucurbitaceous plants and the virus isolate reacted strongly to Papaya ringspot virus (P and W strains) antisera indicating it as Papaya ring spot virus (Mantri et.al, 2005). Though the severe infection of the PRSV has mottling symptoms and blisters on the bottle gourd fruit surface, the internal portion of the infected bottle gourd did not show any deformities (fig 2 b). Further, the internal flesh has normal tests when tested as raw or cooked as vegetables.

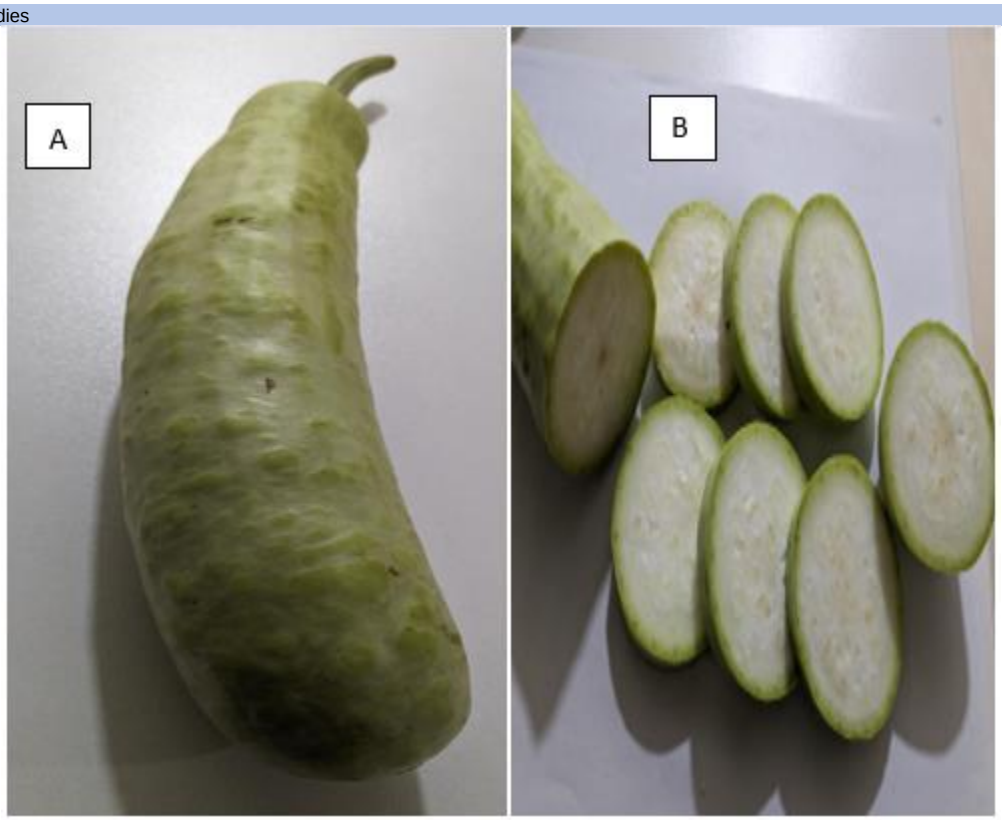

Figure 2: Symptoms of RSPV on bottle gourd: A=blisters on fruit surface; B= internal portion

c.The Papaya ringspot Virus

Papaya ringspot virus (PRSV) is a positive-sense single-stranded RNA virus in the genus Potyvirus, family Potyviridae. Based on the host range, PRSV is grouped into two serologically indistinguishable biotypes: PRSV type-P (PRSV-P) and PRSV type-W (PRSV-W). While PRSV-P isolates can infect species in the families Caricaceae, Cucurbitacea, and Chenopodiacea, isolates of the W type infect only species in the Cucurbitacea and Chenopodiacea (Tripathi et.al, 2008). PRSV is a filamentous flexuous rod virus (760-800 x 12 nm) with a single-stranded positive-sense RNA as its genome. Virus particles contain 94.5 % protein and 5.5 % nucleic acid. The protein component consists of the virus coat protein (CP) which has a molecular weight of about 36 kDa as estimated by Western Blot analysis. The density of the sedimenting component in purified PRSV preparation is 1.32 g/cm3 in CsCl. The PRSV genome consists of a unipartite linear single-stranded positive-sense RNA of 10326 nucleotides with a 5' terminus genome-linked protein, VPg.

Results and Discussion

Humans are exposed to plant viruses through the ingestion of viral-infected fruits, vegetables, and herbal medicines. In some countries in Asia and sub-Saharan Africa, about 80 % of the population uses traditional medicine for primary health care (WHO,2003). TMV is reported to be present and stable in smoked tobacco (Bothwell, 1960; Wahyuni et.al, 2008; Wetter, 1975; Chyle et.al, 1971), resistant to the manufacturing process, and was further detected in smokers' silva (Balique et.al, 2012). Similarly, the ingested plant viruses can be detected in human cells and excreta (Zhang et.al, 2006). Colson et.al, (2010) tested stool samples of 304 adults and 137 children and 21 various food products (sauce, spicy powder, etc) for the presence of PMMOV using real-time PCR, sequencing, and electron microscopy. PMMOV was detected in 57 % of food products, 7.2 % of stool samples of adults, and 0.7 % of children.

Some of the plant viruses detected in human cells and excretion are summarized in Table 1.

Virus Nucleic acid	Virus Family	Genus	species	Detected in human	
ssRNA+	Bromoviridae	Ilarvirus	Prunus necrotic ring-spot virus		Stool (Zhang et.al, 2006)
	Closteroviridae	Closterovirus	Citrus tristeza virus		Stool (Nakamura et.al,2009)
	Secoviridae	comovirus	Cowpea mosaic virus	Hela cell , huvec cell, , and KB cell	
		sobemovirus	Cocksfoot mottle virus		
	Tombusviridae	Tombusvirus	Tomato bushy stunt virus		
		Panicovirus	Panicum mosaic virus		
		Carmovirus	Melon necrotic spot virus		
		Necrovirus	Tobacco necrosis virus		
		Necrovirus	Olive latent virus 1		
		Arenavirus	Oat chlorotic stunt virus		
		Machlomovirus	Maize chlorotic mottle virus		
	Tymoviridae	Marafivirus	Grapevine asteroid mosaic-associated virus		Stool (Zhang et.al, 2006)
			Maize rayado fino virus		

			Grapevine rupestris vein feathering virus		
			Oat blue dwarf virus		
		Maculavirus	Grapevine fleck virus		
			Grapevine red globe virus		
		Tymovirus	Eggplant mosaic virus		
			Onion yellow mosaic virus		Stool (Zhang et.al,2006)
	Virgaviridae	Tobamovirus	Tobacco mosaic virus	Hela cell	
			Pepper mild mottle virus		Sliva (Balique et.al,2012)
			Cucumber green mottle mosaic virus		
			Tomato mosaic virus		
			Turnip vein-clearing virus		
			Tobacco mild green mosaic virus		Stool (Zhang et.al,2006)
			Paprika mild mottle virus		
			Crucifer tobamo virus		
			Nigerian tobacco latent virus		
ds DNA	phycodnaviridae	Chlorovirus	Acanthocystis turfacea chlorella virus		Oropharynx Sample (Yolken et.al, 2014)

Table 1: Plant Viruses Detected in Human

Li et.al (2012) reported that invasion of TMV RNA induces endoplasmic reticulum stress-related outophagy in HeLa cells. The majority of plant viruses are PNS viruses that often harbor multipartite genomes and have either spherical or rod shapes (Hogenhout et.al, 2008). In contrast, animal viruses more evenly harbor RNA and DNA genomes, which are mainly monopartite, and a majority of virions have a spherical shape. Some of the wildlife viruses have crossed their host range barrier to infect humans (Table 2).

Virus (es)	Original host	New Host	Mechanism and/or time
Measles virus	Possibly cattle	Humans	Host switching and adaptation. Time not known
Smallpox virus	Other primates or camels (?)	Humans	Host switching and adaptation >10,000 years ago
Influenza virus	Water birds	Humans, pigs, horses	1910-1916, 1957-1968
HIV-1	Old World primates, chimpanzees	Humans	1930, 1970
SARS CoV	Bats	Himalayan palm civets or related carnivores; humans	2003-2004
Dengue virus	Old World primates	Humans	< 500 yr before
Nipah virus	Fruit bats	Humans (via pig, or bat-to-human contact	
Marburg virus and Ebola viruses	Reservoir host not proven (bats?)	Chimpanzees and humans	
Hendra virus	Fruit bats	Horses and humans	

Table 2: Wildlife viruses that infected humans

Source: Parrish et.al. 2008. Microbiology and Molecular Biology Reviews. 72(3): 457-470.

Plant viruses were found to trigger immune responses in invertebrates, vertebrates, and humans, which might only reflect a history of exposure to foreign protein and does not necessarily imply a role of the plant virus in pathogenesis. CPMV has been shown to induce humoral and persistent systemic and local immune responses in mice as detected by ELISA following oral administration (Florindo et.al, 2002). Mice inoculated with potato virus Y developed antibodies to the virus (Friedland et.al, 2008). AntiPMMoV antibodies in humans were significantly more frequent in serum samples from patients with PMMoV than in a serum sample from control (Colson et. al,2010), and anti-TMV IgG was detected at a higher level in smokers than in non-smokers (Liu et.al, 2013). Production of immunoglobulin (antibodies) IgG and IGM is reflected by fever in the immunized host animal/human (Borkar et.al, 1985). We assume that the clinical symptoms expression may be the result of the initiation of antibody production against the given virus, and therefore only those viruses that show such clinical symptoms may induce immunoglobulin production against the given virus in the host or in other words activate the defense mechanism of the body against the known pathogen. Generally during the vaccination/immunization, the injection of antigen in the body reacts with the fever in the injected host. In the case study of pepper-mild mottle virus

(PMMov), fever, abdominal pain, and pruritus were found significantly common in patients detected with PMMov. Anti-PMMoV IgM antibodies were detected in all PMMoV-positive patients indicating a specific immune response to PMMoV. Based on these findings Colson et.al (2010) concluded that PMMoV, a plant virus, might infect humans and cause clinical symptoms.

In the present case study, the PRSV-P does not induce any clinical symptoms in the volunteers/ participants involved in the case study. It is important to note that the general public is not aware of the symptoms of PRSV-P infection on papaya fruits or bottle gourds and therefore tends to purchase these. Although no clinical symptoms of PRSV-P are reported at present, the awareness of the symptoms of PRSV-P in the papaya and bottle gourd is necessary for the general public to avoid the inbuilt virus inoculum in the

human body. Turri et.al (2020) also reported that cauliflower mosaic virus (CaMV) is not infectious for the human being.

Conflict of Interest: There is no conflict of interest

Source of Funding for Studies: There was no source of funding for these studies

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