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CAUSE OF REDUCED YIELD OF GINGER IN UDAIPUR (RAJASTHAN)

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ABSTRACT

Ginger is a major foreign exchange earning crop. This crop is highly susceptible to infection by rot causing fungi. 90% ginger in the Udaipur (Rajasthan) market comes from a place called Jhadol which is about 50 kms away from Udaipur. Samples of infected rhizomes of ginger collected from different agricultural fields in Jhadol and nearby villages, Udaipur district were screened for rot causing fungal pathogens and identification of the isolates. All isolates were found to belong either to *Pythium aphanidermatum*, *Pythium myriotylum*, or *Fusarium monilliformae*, and *Fusarium solani. P. aphanidermatum* and *P. myriotylum* were recovered from the samples which were infected from wet rot while *Fusarium solani* and *Fusarium monilliformae* from the ginger rhizome infected from dry rot. Pathogenicity testing was done according to Koch's postulates. Hence, this is the comprehensive report on identity of major fungal pathogens associated with ginger rot in Jhadol and nearby villages which are the cause of gingerrotin Udaipur.

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INTRODUCTION

Ginger (*Zingiber officinale* rose L.) is an important spice crop belonging to family *Zingiberaceae*. India stands first in the production (2, 73,333 tones) of ginger and second in the area (80,000 hectares) under ginger cultivation in the world,Ginger is a perennial herbaceous monocotyledon spice and also an important foreign exchange earning crop (Jhala 2010). Ginger is mainly exported in fresh, dry and powder forms. (indiaspice-2009) . Although ginger is a high return crop but ginger rot poses a persistent threat to the cultivation and storage of ginger. About 50-80% losses during storage have been reported due to this disease (Nirmal 1992). Rajasthan, the largest state of India by area, is the single largest producer of many spices like cumin, coriander, fenugreek, ajwan and sweet flag. Chilli, turmeric, ginger, fennel, dill seeds and garlic are the other spices grown in Rajasthan (agricommodityprices.com/2010). Around 90% of ginger produced in Rajasthan comes from Jhadol, a tribal-dominated block in Udaipur district However, during the last 10 years both the area under cultivation and average productivity of ginger have shown a declining trend due to severe rot attack Ginger is a high return but also a high risk crop. Traditionally, a large number of farmers cultivated ginger in the region, but many gave up its cultivation owing to the frequent ginger rot disease that destroyed the crops. (Access2008).

MATERIALS AND METHODS

SAMPLE COLLECTION:

Infected ginger rhizomes were collected in sterilized polybags from various ginger farms in Jhadol and near villages, Udaipur, in the month of July –August. The rhizomes were cleaned and surface sterilized by washing with running tap water and by 0.5% sodium hypochlorite for 3 minutes respectively. The rhizomes were cut into small blocks (1.5cm) and stored in a sterilized polybags for further analysis. Whole of the lab work was conducted at microbial research laboratory University College of science.

Isolation and Identification of fungal pathogens isolated from ginger:

Isolation and Identification of *Pythium species*:

For isolation of *Pythium spp*. 1.5 cm blocks of infected rhizome were washed in running tap water for about 2 hours, surface-sterilised in 0.5% sodium hypchlorite for 3

minutes, rinsed in sterile distilled water, blotted dry on sterile filter paper, and placed (3-5 mm in length) onto water agar (WA), PPP agar (0.10 g of pimaricin, 0.05 g of penicillin,and 0.05 g of polymyxin per liter in corn meal agar and PARP agar (0.005 g of pimaricin, 0.25 mg of ampicillin, 0.01 g of rifampicin, and 0.10 g of pentachloronitrobenzene per liter in corn meal agar (Jeffers and Martin 2010) for isolation of *Pythium* species. Plates were incubated in the dark at 25±1°C for 2 days and examined for the presence of *Pythium colonies*. *Pythium* species grow more rapidly than other fungi so actively growing hyphal tips from periphery were transferred to different medium (CMA, PCA, PDA, WA, V8A) and incubated at room temperature (28-30°C) to study the colony morphology (Van der Plaats-Niterink, 1981). Hyphae were successively transferred to the new medium and maintained.

Morphological identification: The main and traditional methods for identifying *Pythium* species are based on morphological and physiological studies (Taechowisan et al., 2008). Identification of *Pythium aphanidermatum* and *Pythium myriotylum* was based on standard keys suggested by Van der Plaats-Niterink (1981) and Waterhouse (1967,1968) whereas identification of *Fusarium solani* and *Fusarium monilliforme* was done according to keys suggested by Wang et. al. (1996) and Alexopolous and Mims (1993) respectively.

Morphological studies were carried out microscopically (Olympus CX41RF, Olympus Optical, Philippines) for preliminary identification. The studied microscopic characteristics include asexual and sexual structures, appressoria and hyphal swellings, oospores, colony characters etc. Slides were prepared from these cultures and stained with lacto-phenol cotton blue according to to Parija and Prabhakar (1995) and examined under the microscope.

Pythium was identified to the species level on the basis of sexual and asexual structures, The examined morphological characteristics were compared to the documented one in Van der Plaats-Niterink 1981,Waterhouse 1967, 1968, Dick 1990.for both Pythium species. Microscopic examinations were made on living material under the low power (X 10) on undisturbed water culture and under high powers (X20, X45) stained in cotton blue with lectophenol.

To differentiate *Pythium aphanidermatum* and Pythium myriotylum from other fungal species growing with in the same petriplate, microscopic examination of asexual and sexual structure was under the microscope. Sporangial formation was induced by placing young leaf blade (Dick 1990) segments (1 cm) of petal of Rosa indica and Poa annua plants colonized by *Pythium* species in Petri-dishes containing10 ml of sterilized distilled water and incubated at, 25°C, (Abdelzaher et al. 1997). Sexual reproduction is abundant in such water cultures. Identification was based on studies of both water cultures and solid agar media i.e. CMA and potato carrot agar (PCA), each supplemented with 500 mg/ml wheat germ oil. Since, Antheridia and sporangia form rapidly, cultures were observed 12 hours after inoculation and periodically at every four hour interval until all possible characters were observed.

Study of colony growth rate at different temperatures: A 5 mm diameter mycelial plugs taken from the edge of actively growing 3-day-old cultures of each isolates were inoculated centrally on a Petri dish (90 mm diameter) containing 20 ml PDA and incubated at different temperatures such as 5, 10, 15, 20, 25, 30, 35 and 40°C. Five replicates were used at each temperature. The colony diameters of the cultures were recorded at 24 and 48 hours in two directions perpendicular to each other. The colony growth in millimeters over a 24 h period was calculated using the mean colony diameters at 24 and 48 hours, Sporangia were produced by cutting a 4mm diameter, disks from the advancing margins of colony growing on V8 juice agar and floating these disks on 10 ml of sterile soil solution (100 gms of soil to 900 ml of distilled water, shake and leave to settle 24 hours. Take 50 ml of supernatant to 950 ml of distilled water and autoclave it). The dimensions of oogonia, sporangia and oospores were measured. For all characteristics studied, 25 measurements were made for each isolate, and average value was calculated.

Isolation and Identification of *Fusarium species*: For isolation of *Fusarium* species rhizomes of ginger were surface sterilized with 1% sodium hypochlorite for 3 min and rinsed in several changes of sterile distilled water. All the sterilized samples were placed onto general medium, water agar (Burgess et al., 1994) and Pentachloronitrobenzene agar (PPA) plates, a selective medium for *Fusarium* (Nash and Snyder, 1962). The plates were incubated at 25°C for 24 h. The resulting single-spore Fusarium colonies were transferred to fresh Potato dextrose agar plates for further studies. To study the growth rates and pigment production of *Fusarium* spp, all strains were transferred onto potato dextrose agar (PDA) plates and incubated at 25°C. Ten replications were maintained for each Fusarium sp. for microscopic observations, colony of *Fusarium* were transferred to Potassium chloride agar (KClA) (Fisher et al., 1983) medium, Nash medium and Potato Dextrose Agar medium. The species were identified on the basis of macroscopic as well as microscopic observation. Identification of species was based on species description of Gerlach and Nirenberg (1982).

PATHOGENECITY:

Pathogenicity of the isolates of *Pythium* spp. and *Fusarium* spp. were evaluated on hypocotyl tissue of 30 days' old sprouts grown in 12 x 15 cm size polythene bags on a standard pot mixture. Four hours before inoculation, bags were watered to field capacity in order to distribute the inoculum uniformly throughout the soil. Isolates of *Pythium* spp. *Fusarium* spp. were grown separately on potato dextrose agar and in petriplates. Seven days old cultures of *Pythium* spp. and *Fusarium* spp. were harvested by washing mycelial growth and then suspended in 100 ml of tap water and maintained by a magnetic stirrer. The mycelial/spore suspension was added to the bags around the hypocotyl @ 50 ml per bag. Separate plants were independently inoculated with the isolates of *Pythium aphanidermatum, Pythium myriotylum, Fusarium solani* and *Fusarium monilliformae* and suitable controls were also maintained. When the symptoms became apparent, the same were recorded from the infected plants and re-isolations made from them. Fungi in the genus *Pythium* were identified with the use of standard keys by Van der Plaats-Niterink 1981and Waterhouse's Monograph (1968) and *Fusarium* with (Gerlach and Nirenberg 1982, Alexopolous and Mims,1993).

RESULTS:

In this study, the recovered isolates from diseased ginger rhizomes were identified on their morphological characteristics as well as colny growth and morphology on different mediums these isolates were identified as, *Pythium aphanidermatum, Pythium myriotylum, Fusarium monilliformae* and *F. solani*.

Pythium aphanidermatum The hyphae are hyaline and the mycelium has no cross walls. Main hyphae up to 7.5 µm wide and oospores aplerotic, spherical, smooth, average 41 µm in diameter. Sporangia are lobate consisting of terminal complexes of swollen hyphal branches of various length and germinated by extension of long exit tube and vesicle formation and zoospore discharge. Vesicles average 41 µm in diameter. The presence of apluerotic oogonium, Oogonia globose, terminal, smooth, average 28 µm in diameter, with straight oogonial stalks. Antheridia typically intercalary, usually diclinous and average 14 µm long and 11 µm wide, commonly 1 per oogonium . It produces white colored aerial mycelium on different mediums.

Pythium myriotylum In case of *Pythium myriotylum* the growth on PDA and cornmeal agar is without a special pattern, colony show white colour. Main hyphae up to 8 µm wide. Appressoria clavate, knob-like or sickle-shaped, usually formed in clusters. Sporangia terminal or intercalary, filamentous, consisting of undifferentiated and inflated lobulate or digitate elements of variable length. Oogonia terminal or intercalary, globose, Oospores are aplerotic

Fusarium solani, The colonies are woolly to cottony with cream to white aerial mycelium on PDA medium. pigmentation of reverse colony is red, macroconidia mostly 5 septate and thick walled. Apical cell is tapered and basal cell foot shape. Mycelium were abundant, Macroconidia have three to four septa on average, were slightly curved, were wide and thick walled, and they were showing a slightly blunted apical end. Microconidia were abundant, oval to kidney shaped, and formed in false heads.

F. monilliforme, The colonies of *F. monilliforme* were slimy, cottony , pink in colour on PDA the hyphae were slender , 3 to 5 microns broad, closely septate and much branched. Microconidia are formed in chain. Each microconidia is 1 to 2 celled elliptical to oval and unicellular comma shaped. Macroconidia are 2-5 celled and falcate narrow at both ends.

On the basis of above studies the isolates G1,J G2,J G3,J and G4,J were found to be *Pythium aphanidermatum , Pythium myriotylum, Fusarium solani* and *Fusarium monilliformae* respectively.

DISCUSSION:

The two main fungal pathogens associated with rot of ginger in Jhadol and near villages are Pythium spp. (Pythium myriotylum and Pythium aphanidermatum) and other is Fusarium spp (Fusarium solani and Fusarium monilliformae).

Pythium is one of the most important seed and soil borne fungi cause diseases in numerous types of vegetable crops that reduce the germination and cause pre- and post-emergence damping-off of seedling, resulting in poor plant stand (Nunez, 2001; Gotlieb, 2002). Generally, Pythium species produces sporangia on leaves and these sporangia may be carried on the surface of irrigation water especially under flooding and furrow irrigation (Duniway, 1974; Zentmayer and Erwin, 1970) and may infect other plants. Pythium ahanidermatum (Edson) Fitz. is a major constraint for the production of healthy rhizome all over the world, sometimes causing total failure of crop (Fagaria et al. 2006). Fusarium comprises a wide and heterogeneous group of fungi it is a parasitic member of family deuteromycetes have a wide host range causes wilting as well as rotting , Fusarium solani is a phytopathogenic fungus and is an important causal agent of several crop diseases (Luginbuhl 2010), Fusarium monilliformae is also an important plant pathogenic fungus that causes serious diseases in many crops. (Rangaswami,1975)

The pathogens identified in this study have ability to grow on a wide range of substrates and have efficient mechanisms for dispersal as well as they can survive in the soil and in plants for many years as chlamydospores and sporangiospores (Schulz et al., 2000). Therefore, it is very important to develop proper management practices to control this type of fungi. The results of this study revealed that *Pythium* and *Fusarium* species which are pathogenic to ginger are found adundant in farms of Jhadol and near villages. In this investigation we observed that the severity of the disease is high due to high temperature condition in Rajasthan which is favourable for the growth of specially both *Pythium aphanidermatum* and *Pythium myriotylum*.

The identification of fungi causing the decay of ginger rhizomes is the first step towards further studies to develop an integrated crop management program to prevent and control ginger rot in the fields.

CONCLUSION:

Species of *Pythium aphanidermatum, Pythium myriotylum, Fusarium solani* and *Fusarium monilliformae* were confirmed by morphology under microscopic observation and it is confirmed that these four fungal pathogens are found in Jhadol and near villages region causes ginger rot and are the cause of yield reduction .

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REFERENCES:

Abdelzaher HMA (1999). The genus Pythium in Egypt. Proceedings of the second international conference on Fungi: Hopes & Challenges. Cairo, 29 Sept 1 Oct.vol. 11 – 31.

Alexopolous CJ and Mims CS (1993). Introductory Mycology (3rd ed.), Wiley Eastern Limited, New Delhi, 24-25.

Burgess LW, Summerell BA, Bullock P and Backhouse D (1994). Laboratory manual for Fusarium research, Department of crop science. University of Sydney, Sydney, Australia. 3rd ed. p. 133.

Dake GN (1995). Diseases of ginger (Zingiber officinale Rosc.) and their management. Journal of Spices and Aromatic Crops 4:40–48.

Duniway JM (1974). Formation of sporangia by Phytophthora drechsleri in soil at high matric potentials. Can. J. Bot. 53: 1270-1275.

Fageria MS, Choudhary, BR and Dhaka RS (2006). Vegetable crops production technology, Vol. II, Kalyani Publisher, New Delhi, pp. 223-227.

Fisher NL, Marasas WFO and Toussoun TA (1983). Taxonomic importance of microconidial chains in Fusarium section Liseola and effects of water potential on their formation. Mycologia. 75: 693-698.

Gotlieb, A. (2002). Seedling Damping-off. The University of Vermont Extension.http://www.uvm.edu/extension/publications/gardendisease/gd9.htm. Accessed on 20 February 2003.

Gerlach W and Nirenberg H (1982). The genus Fusarium - a pictorial atlas. Mitt. Biol. Bundesanst. Land Forstwirtsch. Berlin Dahlem 209:1-406.Ho HH (1986).

Palmer LT and Kommedahl T (1969). Root-infecting Fusarium species in relation to rootworm infestations in corn. Phytopathol. 59: 1613-1617.

Jeffers, S N and Martin SB (1986). Comparison of two media selective for Phytophthora and Pythium species. Plant Dis.70:1038-1043

Kamoun S, Huitema E and Vleeshouwers VGAA (1999). Resistance to Oomycetes: a general role for the hypersensitive response. Trends in Plant Science 4:196–200.

Lawrence BM (1984). Major tropical spices - Ginger (Zingiber officinale Rosc.). Perfumer & Flavorist 9:1–40.

Mounira S B N, Abad C P, Alv arez L A and Sierra APG (2010). First report of Pythium indigoferae and P. irragulare Asssociated to Applle Tres decline in Tunisia. Journal of Phytopat hology,159:352-357.

Nirmal K, Samsuddin K and Ratnambal MJ (1992). Plant, Cell tissue and Organ culture. 29:71-74.

Nunez A (2001). Seedling diseases of vegetables. Vegetables crops. Plant Pathology http // www .co . kern . ca . us / farm/NUNPUBO7.htm.

Plaats-Niterink, AJ (1981). Monograph of the genus Pythium. The Netherlands. Centraalbureau Voor Schimmelcultures 21: 1-242.

Parker CK (2010). Pythium aphanidermatum PP728 soilborne plant pathogens class project. Available on tp://www.cals.ncsu.edu/course/pp728/Pythium/P_aphanidermatum. Html.

Parija W and PK Prabhakar, (1995). Evaluation of lacto-phenol cotton blue for wet mount preparation of feces. J. Clin. Microbiol., 33: 1019-1021.

Plaats-Niterink, (1981). A.J. van der Plaats-Niterink, Monograph of the genus Pythium, Stud. Mycol. 21 (1981), pp. 1–244.

Rangaswami G (1975). Disease of Cereals. In: Disease of Crop Plants in India, (2nd ed.), Hall of India Private Ltd., New Delhi.

Selvan MT, Thomas KG and Manojkumar K (2002). Ginger (Zingiber officinale Rosc.). In: HP Singh, K Sivaraman and MT Selvan (editors). Indian Spices - Production and Utilization. Coconut Development Board, India. pp. 110–131.

Schulz D, Lambrecht S and Wolf G (2000). Biological control of Pythium ultimum in Sugar

Beet. Department of Plant Pathology and Plant Protection, Germany.http://www.gwdg.de/~dschulz2/leereseite.1.htm. Accessed on 5 February 2000.

Sarah Luginbuhl (2010). A class project for PP728 Soilborne Plant Pathogens, Fall 2010.

Taechowisan T, Wanbanjob A, Tuntiwachwuttikul P, Shen Y and Lumyong S (2008). Synergistic activities of 4-arylcoumarins against phytopathogenic fungi. Res. J. Microbiol., 3: 237-245.

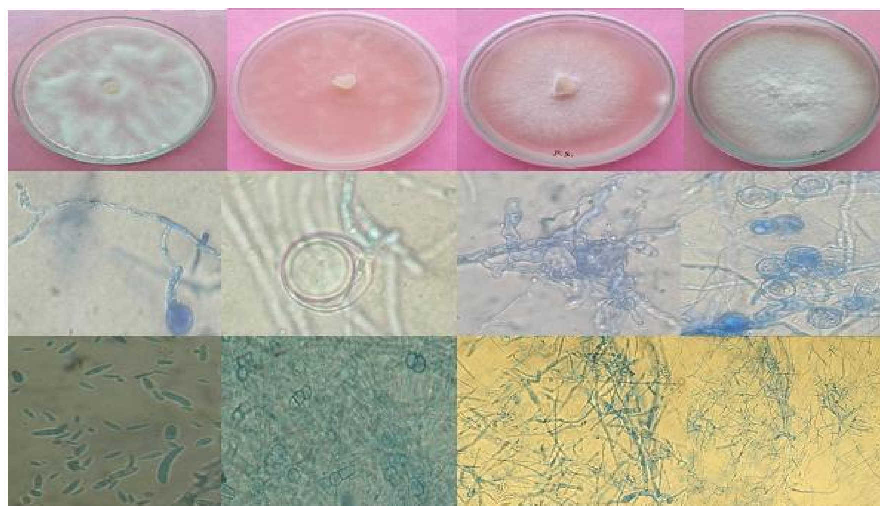
Wang G C, Zheng Z., Ye Q M, Zhang CL (1996). Guide to identification of common Fusarium species (Chinese). Beijing, China: China Agricultural Science and Technology Press.

Waterhouse GM (1967). Key to Pythium Pringsheim. Mycol Pap 109:1 – 15.

Jhala YC (2009). Ginger cultivation: Capital intensive but profitable Published on: Sep 2009 www.indianspices.com/admin/contentFiles/.../Review 2008-09Diseases.

Zentmayer GA, Erwin DC (1970). Development and reproduction of Phytophthora spp. Phytopathol. 60: 1120-1127

Zheng XB (1997). Phytophthora and Methods in Phytophthora. Beijing, China: Agricultural Press



Figures from left to right Ist Row: *P. aphanidermatum, P.myriotylum, F.solani, F.monilliforme,* Colony on PDA.
IInd Row: P.a. (Aseptate hyphae, Sporangium, Oospore). P.m. (Aseptate hyphae, Appresorium, Developing Oospores).
IIIrd Row: F.s.(Macroconidia, Microconidia, Chlamydospores). F.m. (Septate hyphae, Macroconidia, Microconidia).