



## Epidemiology, Source of Resistance and Management of Blue Mould Rot Disease of Aonla (Indian Gooseberry) to Identify Research Priorities and Knowledge Gaps, a Review

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### Abstract

Earlier, the blue mould rot was a minor disease but now it becomes a major disease of Indian gooseberry. The effect of weather parameters i.e. temperature and relative humidity on disease development were studied, *in vitro*. The combination of 30 °C temperature with cent per cent relative humidity resulted in maximum disease incidence of 87 and 100% in both 5 and 10 days after inoculation (DAI), respectively. Minimum disease intensity (15 and 22%) was recorded at 10 °C temperature and 40 per cent relative humidity at 5 and 10 days after inoculation. However, with further increase in temperature and relative humidity, the per cent disease intensity was also increased and it also increased during the period of time (5 and 10 days after inoculation) clearly elucidated that low temperature (10 °C) with low relative humidity (40%) is ideal for the post harvest storage of Indian gooseberry. A minimum number of 50 spores/ml was necessary for disease initiation. The resistance and susceptibility against the blue mould rot (*P. islandicum*) disease can be varied among the varieties of *E. officinalis* suggesting that the resistant ones should be preferred for further plantation activities to avoid the frequent damage and losses caused by the disease. Relative efficacy of chemicals, plant extracts and bio-agents against blue mould rot tested *in vitro* and *in vivo* at different concentrations revealed that boric acid (chemical) at 1.0 M concentration, neem leaf extract (plant extract) at 20 per cent concentration and *Trichoderma harzianum* were exhibited the best to inhibit growth in pre- and post-inoculation after five and ten days of inoculation in both test conditions.

**Keywords:** Indian gooseberry, blue mould rot, epidemiology, source of resistance, disease management.

### Introduction

Aonla or Indian gooseberry (*Emblica officinalis* Goerth) is one of the most important indigenous fruit of Indian sub-continent (Baghel. *et al.*, 2007). It grows in tropical and subtropical parts of India, China, Indonesia and the Malay Peninsula (Golechha. *et al.*, 2012 and Srivasuki, 2012). During the last decade there has been unprecedented expansion in the area under aonla cultivation across the country utilizing the wasteland.

This has resulted in efficient utilization of resources leading to better income to farmers, nutritional security coupled with enhanced employment and rehabilitation of wastelands (Pathak, 2003). Among post-harvest diseases in aonla, fruit rot caused by *Penicillium islandicum* Sopp. (Radha and Mathew, 2007; Akhund. *et al.*, 2010) is the most important as it affects the fruit quality and quantity in relation to the market value. The open

wounds, created during harvesting, handling and packaging are the major sites of invasion by post-harvest wound pathogens, the protection of wounds by chemicals considerably decrease decay in storage (Yadav. et al., 2012). Akhund et al., (2010) observed 58.33 per cent incidence of *Penicillium islandicum* from aonla fruits from the samples of Hala (Pakistan).

Post-harvest fungi are responsible for changes in biochemical contents of fruits (Rajmane and Korekar, 2014). Plant enzymes are involved in defence reactions against pathogens by catalysing the formation of lignins and other oxidative phenols which contribute to the formation of defence barriers for reinforcing the cell structure (Liang. et al., 2011). Use of fungicides causes environmental hazards and development of resistance in pathogen. Yulin Cheng. et al., (2020) reviewed that green mould, major postharvest disease of citrus fruit, caused by *Penicillium digitatum* leads to huge economic losses worldwide and fungicides are still the main method currently used to control post-harvest green mould in citrus fruit. The molecular mechanism of plant-pathogen interactions is crucial for developing novel and safer strategies for effective diseases control.

Priyamedha Senguptaa. et al., (2020) reviewed different post-harvest fungal diseases like blue mould, anthracnose and fruit rot of Indian gooseberry along with its management strategies that covers physical, chemical and biological methods. The physical methods of management include osmo-drying, hot water treatment, UV-C treatment, application of high electric field current, efficient packaging in low and high density polythene pouches. Chemical methods involve treatment with different synthetic fungicides like mancozeb, gibberellic acid, carbendazim, calcium nitrate and calcium sulphate. Lastly, the biological control methods have been mentioned, which includes application of microbial antagonists and naturally occurring antifungal compounds. In recent years more emphasis has been given on the use of bio-agents and plant extracts (Maharishi. et al.,

2009; Yadav. et al., 2013; Jat. et al., 2013). Management of blue mould rot in aonla and other fruits through chemicals have been reported by several workers. (Rathod and Patel, 2005; Yadav et al., 2012).

Barring few reports about its occurrence, epidemiology and source of resistance, the comprehensive studies have not been taken up on this pathogen in aonla fruits, therefore, the information available on fruit rots of aonla and other fruit crops caused by *Penicillium* spp. in particular and other fungi in general has been reviewed as under.

### **Epidemiological Studies**

Susceptibility of crops and relative importance of fungi in causing post-harvest rotting is reported to vary according to site, season, damage and harvesting conditions as well as post-harvest treatments and storage conditions (Derbyshire and Shipway, 1978, Denis, 1983).

### **Effect of Temperature and Relative Humidity on Disease Development**

*Alternaria* rot, *Penicillium* mould rot and *Aspergillus* black mould rot of citrus caused by *Alternaria alternata*, *Penicillium digitatum*, *Penicillium italicum* and *Aspergillus niger*, respectively have been reported to develop more rapidly at temperature, 15-35°C, 20-25°C and 30-35°C (Fawcett and Barger, 1927, Agrawal and Hasija, 1967 and Bhargava, 1972) respectively. Temperature, ranging from 20-35°C is reported to be optimum for the rapid development in case of fruit rot of citrus. Severity of *Penicillium* and *Botryodiplodia* rot of sweet orange caused by *Penicillium italicum* and *Botryodiplodia theobromae*, respectively have been reported to be highest at 100 per cent relative humidity and lowest at 40 per cent relative humidity (Tandon. et al., 1975 and Godara, 1994). Gupta and Chauhan (1998) reported effect of temperature on the development of soft rot of *Emblica officinalis* caused by *Penicillium citrinum* strains I to VI. They observed that strain VI produced maximum rot (49.5 per cent) after 24 days at 25°C, while at temperatures 15°C, 20°C, 30°C, 35°C, percentage rot were 17.3, 30.2, 18.3 and 4.8, respectively. Plant diseases including fruit

rots were to be more common in humid to wet regions with cool and warm or tropical temperature (Agrios, 2005). The best temperature for growth of *Penicillium* spp. was found to be 23°C, and by decreasing the temperature fungal growth decreased (Agostini. et al., 2006). Baghel et al., (2008) studied the effect of temperature and relative humidity on fruit rot of aonla caused by *Penicillium citrinum*. They observed that temperature of 10°C was most effective for preservation of aonla as it showed minimum disease severity. However, temperature of 25 and 20°C showed maximum disease severity of 87.7 and 71.8% respectively. They also reported that highest disease severity (100%) was observed in the fruits stored at 100 per cent RH. Minimum rotting (47.1%) was recorded when fruits were stored at 50 per cent RH.

#### **Effect of Inoculum Load on Disease Initiation, Threshold Point and Population Build Up**

In a study, it was found that 108 conidia ml<sup>-1</sup> of *A. alternata* in tomato resulted in 93.4% infection recorded after 7 days of incubation meaning there by that 108 conidia ml<sup>-1</sup> could serve as the inoculum potential of *A. alternata* (Verma, 2004). Yousefi and Hagian Shahri, (2009) conducted pathogenicity tests by inoculating slightly wounded plant tissue with conidial suspension adjusted to 1.5×10<sup>4</sup> conidia ml<sup>-1</sup> of *Alternaria alternata* using a hemocytometer that was capable of causing infection within 2-4 days. *Alternaria* isolates was capable of causing infection on citrus leaves from suspension of conidia (105 conidia /ml) prepared from 5-7 days old PDA cultivated isolates (Kakvan. et al., 2012).

#### **Source of Resistance**

Saini et al., (2019) reported that resistance and susceptibility against the blue mould rot (*Penicillium islandicum*) disease can be observed among the varieties of *E. officinalis* suggesting that the resistant ones should be preferred for further plantation activities to avoid the frequent damage and losses caused by the disease. Among nine commercial varieties (Desi, Hathizool, Kanchan, Krishna, Chakaiya, Banarsi, NA-7, NA-9, NA-10), least disease intensity was recorded in Desi (2.67%)

and Kanchan (3.33%) while maximum disease intensity was observed in Chakaiya (50.00%) and Banarsi (49.33%).

Tiwari et al., (2008) reported field screening of aonla varieties against *Deudorix Isocrates* (Fabr.). The maximum 43.70% fruit damage was observed in NA-7 (Narendra-7) and the minimum 33.60% was observed in Chakaiya. The order of susceptibility of different varieties was NA-7 (43.70%), Kanchan (41.25%), NA-6 (40.80%), NA-10 (38.40%) and Chakaiya (33.60%). Similer observations were also reported by Padmavati. et al., (2002). Meshram and Soni, (2011) screened certain varieties for resistance to insect pests and diseases in central India. They reported that ten varieties of *Embllica officinalis* Gaertn. including Kanchan, Chakaiya, Francis, NA-7, NA-10 (Narendra 10), Anand-1, Anand-2, Krishna, Hatizola (Local) and Local-wild were screened against insect pests {gall forming insect (*Betousa stylophora* Swinhoe), leaf roller (*Garcillaria acidula* Forster), bark eating caterpillar (*Indrabela quadrinotata* walker)} and diseases that is vascular wilt (*Fusarium oxysporum* f.sp., *albedinis* Killian and Maire), fruit disease (*Alternaria* sp.). The results revealed that variety NA-10 followed by Kanchan was found to be least preferred by *B. stylophora*, *G. acidula*, *I. quadrinotata* and *Alternaria* sps. in clonal seed orchards. Whereas, variety Hatizola (Local) followed by Francis showed less incidence caused by *Fusarium oxysporum* in nursery stage.

#### **Biochemical Basis of Resistance**

The fungal infection not only blemishes and disfigures the fruits, but also bring about changes in nutritional contents, physiological and biochemical properties (Sarkar et. al., 2010). The post-harvest fungi are responsible for changes in biochemical contents of fruits (Pawar 2012). Rajmane and Korekar, (2014) studied the biochemical changes in mango and papaya fruits due to post harvest fungi and found maximum decrease in reducing sugar in local varieties of mango.

#### **Disease Management**

Literature scanned revealed that a very meagre research work has been carried out on

blue mould rot (*Penicillium islandicum*) of aonla and their management. The open wounds, created during harvesting, handling and packaging are the major sites of invasion by postharvest wound pathogens, the protection of wounds by chemicals considerably decrease decay in storage (Yadav. et al., 2012) Many chemical compounds have been used as part of post-harvest treatment of fruits for the retardation of microbial infection (Mecteau. et al., 2002, Hervieux. et al., 2002, Mills. et al., 2004).

### Chemical Control

Fungicides are often combined as co-formulations because they are capable of improved disease control, and resistance management. Mixture of calcium chloride, bavistin, and bayleton, when applied to leaves, was found to enhance the shelf-life of Indian gooseberry (Gupta and Singh, 2016). For controlling the development of white specks in aonla (*Emblica officinalis*) fruits (Varieties Chakaiya and Desi) during storage period of 90 days in solution at ambient temperature (11.33°C), a chemical solution containing 10% salt and 0.04% KMS (Potassium metabisulphite) was found effective (Premi. et al., 1999). The uniformly harvested mature fruits of aonla var. Chakaiya were dipped for 10 minutes in an aqueous solution of Gibberallic acid (GA<sub>3</sub> 10 or 25 ppm) and Kinetin (100 or 150 ppm), the minimum decay loss due to (*Aspergillus* spp; *Penicillium* spp. and *Colletotrichum* spp.) was observed in fruits treated with GA<sub>3</sub> at 25ppm, followed by GA<sub>3</sub> at 10 ppm (Singh and Kumar, 2000). Post-harvest application of thiabendazole (0.5%), benomyl (0.05%) and carbendazim (0.05%) were found effective against the various fungal rots caused by *Aspergillus niger*, *Penicillium digitatum* and *Penicillium italicum* in citrus fruits (Verma and Tikoo, 2003). Sodium metabisulfite, a compound with antimicrobial activity has been shown to completely inhibit *in vitro* mycelial growth and sporulation of *H. solani* *Fusarium sambucinum* a wide range of potato postharvest pathogens pathogens (Hervieux. et al., 2002, Mecteau. et al., 2002, Mills. et al., 2004) at a concentration of 0.2M.

Carbendazim at 1000 ppm gave 100 per cent control of *Penicillium italicum* in Kinnow fruits upto 60 days in storage (Singh and Thakur, 2005). Rathod and Patel, (2005) observed carbendazim (500 and 1000 µg/l) and mancozeb, (2000 and 4000 µg/l) most effective against the *colletotrichum*, *Penicillium* and *Alternaria* rots in aonla fruits both in pre and post-inoculation treatments. Application of bavistin (0.05%) and Kavach (0.2%) found effective in minimizing the *Penicillium* rot (*Penicillium fellutanum*) severity in aonla fruits. Verma, (2008) reported that post-inoculation treatments with thiabendazole, benomyl and carbendazim (@ 0.05%) were most effective against the green mould (*Penicillium digitatum*) and blue mould (*Penicillium italicum*) rots of mandarin oranges. Latifa et al., (2011) reported the effect of organic acids and salts on the development of *Penicillium italicum*, the causal agent of citrus blue mould. Tested at 0.2 M concentration 14 out of 28 studied compounds completely inhibited the mycelial growth and sporulation of *Penicillium italicum*. Among these chemicals, ammonium carbonate, ammonium molybdate, Sodium phosphate, sodium sulfite, EDTA, sodium metabisulfite and sodium salicylate were also fungicidal. Montesinos- Herrero. et al., (2011) reported that postharvest green mold and blue mold, caused by *Penicillium digitatum* and *Penicillium italicum* respectively, were effectively controlled by fumigation of lemons and oranges with ammonia gas. Yadav. et al., (2012) recorded lowest *Penicillium* rot severity (*P. funiculosum* Thom.) in aonla fruits treated with carbendazim at 1000 ppm both in pre (12.49%) and post-inoculation (12.83%) followed by benomyl 1000 ppm both in pre (13.49%) and post-inoculation (15.03%) treatment at 4 days after inoculation.

### Plant Extracts

Extract of the indigenous plant parts have shown success in plant disease control and are proved to be harmless and non-phytoxic unlike chemical fungicides (Spencer. et al., 1957). The extract of the plant also exhibited marked effect on germination of fungal spores as well (Shekhawat and Prasad 1971 and

Singh. et al., 1983). Sonawane et al., (2012) studied the effects of leaf extracts of some medicinal plant against *Aspergillus niger*, a causal agent of storage disease of amla. Among the 10 plant extracts tested against *A. niger*, *Tinospora cordifolia* (78.10%) were significantly superior over all other plant extracts followed by *Boerhavia diffusa* (72.45%), *Ocimum santum* (71.00%), *Tribulus terrestris* (62.18%) and *Adathoda vasica* showed less inhibitory effect (6.60%). Goswami and Sumbali, (2010) evaluated the plant bulb extracts of *Allium sativum* (garlic) and *A. cepa* (onion) and rhizome extract of *Zingiber officinale* for the management of *Penicillium* rot of *Phyllanthus emblica*. They reported that the extract of onion bulb was comparatively less effective than garlic extract. Aqueous extract of onion bulb gave 8.8 to 36.5% control as a pre-infection dip and 26.4 to 46.3% control as a post-infection dip treatment whereas acetone extracts gave 19.3 to 41.9% control as a pre-infection dip treatment and 16.7-36.8% control as a post-infection dip treatment. Efficacy of onion bulb extract may be due to the presence of protocatechuic acid and catechol in the bulb, which were responsible for the bursting of young fungal hyphae. In addition, onion bulb was reported to contain certain active components like cycloallin, Ace-AMP I and allicepin, which might be responsible for its antifungal activity (Wang and Ng, 2003). Bhujbal et al., (2011) reported that maximum per cent disease control due to *Aspergillus niger* was observed in treatment carbendazim (89.88%) followed by chlorothalonil (84.91%), *Trichoderma viride* (83.29) and *Trichoderma viride* + *Bacillus subtilis* treatment (76.68%) while tulsii extract (74.61%), *Bacillus subtilis* (62.46%) and onion extract (60.15%) were found to be effective in reducing the infection of *A. niger*. Among all treatments, the neem extract was found to be least effective. Bio-efficacy of nine phytoextracts at 10 per cent were tested against the mycelial growth and sporulation of *Penicillium funiculosum* *in vitro*, among them significantly lowest mycelial growth was recorded in neem leaf extract (7.75 mm) showing 89.49% growth inhibition. Further, it also proved most effective in reducing the

*Penicillium* rot severity both in pre (20.44%) and post-inoculation (20.87%) treatments at 7 day after inoculation, respectively (Yadav. et al., 2013). In three years studies, Jat. et al., (2013) found maximum blue mold rot of aonla control with *Azadirachta indica* leaf extract (5%) followed by carbendazim (0.1%), *Curcuma longa* rhizome extract (5%) in both pre- and post-inoculation treatments.

### Biological Control

Bagwan, (2003) reported the potentiality and viability of *Trichoderma* spp. and *Candida* spp. to control green mould (*Penicillium digitatum*) and blue mould (*Penicillium italicum*) of citrus (*Citrus sinensis* L.) respectively. The biocontrol agent *Trichoderma viride* was also found effective in controlling the green mould and blue mould fruit rots of citrus (Bagwan, 2003). Krol, (2004) observed *Trichoderma* spp. as best antagonist to control *Phomopsis viticola* on grape vines. Rathod, (2010) found *Trichoderma harzianum* as most efficient antagonist in controlling the rots caused by *Penicillium islandicum*, *Colletotrichum gloeosporioides* and *Alternaria alternata* in aonla fruits. Etebarian et al., (2005) observed biological control of apple blue mould (*Penicillium expansum* or *Penicillium solitum*) with *Pseudomonas fluorescens*. However, *Trichoderma viride* was superior over *Pseudomonas fluorescens*. Choubey and Patil, (2009) evaluated the antagonists namely *Trichoderma viride*, *T. harzianum* and *T. virens* against *Phomopsis* fruit rot of aonla caused by *Phomopsis phyllanthi* both in pre-inoculation and post-inoculation methods. In pre and post-inoculation methods, *Trichoderma harzianum* was found significantly most effective in reducing the *Phomopsis* fruit rot severity (2.07 and 18.59%) on 4<sup>th</sup> and 7<sup>th</sup> day of inoculation followed by *Trichoderma viride* (2.13 and 19.82%) over control. Management of blue mould rot (*Penicillium italicum*) of Kinnow fruits was explored using biocontrol agents viz. *Trichoderma*, *Gliocladium*, *Bacillus*, *Pseudomonas*, *Debaryomyces*, and *Sporidiobolus* spp. revealed variable efficacy *in vitro*. On further testing as pre and post-inoculation treatment, the yeasts showed an edge over other fungal and bacterial spp. with *D.*

*hansenii* and *S. pararoseus* exhibiting 89.12% and 84.12% as pre-inoculation, and 86.37% and 78.26% as post-inoculation treatments, respectively (Maharishi. et al., 2009). Jat et al., (2013) found maximum fruit rots (*Aspergillus* rot and *Penicillium* blue mould rot) control with *Azadirachta indica* leaf extract (5%) followed by carbendazim (0.1%), *Curcuma longa* rhizome extract (5%), *Trichoderma viride* and *Pseudomonas fluorescens* in both pre and post-inoculation treatment. Wakade et al., (2015) evaluated the effect of microbial antagonists against the fruit rot complex of Indian gooseberry *in vitro* and reported that *P. fluorescens* recorded the maximum growth inhibition against *A. niger* and *P. funiculosum*.

### Conclusion

The blue mould rot aonla caused by *Penicillium islandicum* adversely affects the fruit quality, quantity and ultimately reduces the marketable value. The epidemiological studies revealed that the injury of the aonla fruits was found to be a pre-requisite for infection. A minimum number of 50 spores/ml was necessary for disease initiation whereas maximum blue mould rot incidence was noted at 30°C and 100 per cent relative humidity. Varietal screening revealed that least blue mould rot intensity was found in Desi and Kanchan, while maximum rot was observed in Chakaiya and Banarsi of aonla varieties. In biochemical studies, it was observed that total soluble solids, total phenol, peroxidase (PO) and polyphenol oxidase (PPO) activity increased in diseased fruits in comparison to healthy fruits, while ascorbic acid and acidity decreased over time in both healthy and diseased fruits. Relative efficacy of chemicals, plant extracts and bio-agents against blue mould rot tested *in vitro* and *in vivo* at different concentrations revealed that boric acid (chemical) at 1.0 M concentration, neem leaf extract (plant extract) at 20 per cent concentration and *Trichoderma harzianum* were exhibited the best to inhibit growth in pre- and post-inoculation after five and ten days of inoculation in both test conditions.

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