



Phytoextracts as management tool against fungal diseases of vegetables

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Abstract

Vegetables are the basic source of food, energy and dietary fibers for mankind. But the production of vegetables is affected due to various biotic and abiotic factors. Fungal pathogens are responsible of vegetable diseases and cause high economic losses. Synthetic fungicides which are toxic and harmful to environment are used to control vegetables diseases caused by fungal pathogens and now a day's trend shifting towards healthy, safe and sound ecofriendly control of fungal pathogens. Phytoextracts of *Azadirachta indica*, *Allium sativum* and many others significantly inhibit the fungal growth and spore germination. Secondary metabolites of plants like polyphenols, glycosides, quinones, terpenoids, saponins, allicin, flavonoids and scopolamine cause fungal inhibition by disrupting mitochondria, cell membrane and cell wall of pathogen. Several technologies are being used for extraction of phytoextracts as these are natural bio-fungicides and there is further need to explore fungicidal activities in relation to human health.

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Introduction

Edible part of plant like root, stem, leaf, fruit, flower, bud, tubers, corms, bulbs that can be cooked and have variety of taste, aroma color, texture and nutritional value. More than 400 vegetable crops grown commercially in world (Kays., 2011). Vegetables source of vitamins, antioxidants, dietary fibers and minerals. Vegetables as balanced diet reduced health risks like cardiovascular disorders, gastrointestinal disorders and diabetes. Vegetables consumption assist to regulate blood glucose concentration, cholesterol while antioxidants quench the free radicals thus overall have health benefits (Dias. 2012). Fungi acts as natural decomposers that cause 20-25% lost vegetable and fruits (Massoud., 2013). Fungal pathogens have severe negative effect on crop production and cause qualitative and quantitative losses (Shuping and Eloff., 2017). Fungal pathogens may be biotrophs that depend on living plant cells absorb nutrient through appressoria and haustoria, necrotrophic fungi that actively kills host plant cells through cell wall degrading enzymes and toxins secretions while hemi-biotrophs initially biotrophic later become necrotrophic (Silva *et al.*, 2016). Synthetic fungicides being carcinogenic have undesirable effects so that plant extracts found to be effective and alternative ecofriendly fungicides (Alkhail, 2005). Plants rich in secondary metabolites such as tannins, terpenoids, alkaloids and flavonoids have antimicrobial properties since their use increasing rapidly for management diseases (Cowan, 1999). Phytoextracts (plant extracts) or plant derivatives are biological antifungal agents have potential to overcome fungal diseases without human health risks (Shuping and Eloff, 2017). Synthetic fungicides have severe effects on aquatic and terrestrial ecosystem while copper accumulation as result of severe use of copper fungicides have adverse effects on microorganisms and earthworms (Wightwick *et al.*, 2010). Fungicides applications extremely lowered the microbes present in soil. Extinction of microorganism effects nutrients holding capacity soil and said to be as dead soil. Fungicides against soil borne diseases are hazardous to environment and health (Ratna *et al.*, 2015). Fungal

pathogens adversely affect and cause biodiversity loss. Chemical fungicides remain in soil and possess serious ecological threat. So ecofriendly, less expensive and effective bio fungicides preferred over these chemicals (Deepa and Padmaja, 2013). On the other hands the plants containing secondary metabolites cause fungal inhibition of pathogen by dysfunction of mitochondria, suppression of cell division and disrupting cell membrane, cell wall, DNA, RNA and protein synthesis (Freiesleben and Jagar, 2014). Toxic and nonbiodegradable synthetic fungicides noxious to human and environment therefore plant extracts could be used as natural fungicides (Ekefan *et al.*, 2018). Phytoextracts proved better botanical pesticides as they environment friendly and cause significant reduction in fungal growth (Parajapati *et al.*, 2017).

Phytoextracts against fungal diseases

Preparation of plant extracts

Plant sample that may be root, stem, leaf, flower, fruit, bark taken as fresh and dried sample for extraction. Dried sample preferred over fresh sample because of their stability on the other hand fresh sample decay faster than dried sample. Moreover, smaller size particles of sample in powdered form have greater surface contact to solvent for efficient extraction than grinded sample (Azwanida., 2015).

Extraction means separation of soluble plant metabolites by using suitable solvents. Maceration, Decoction and infusions are conventional methods used for extraction. Choice of solvent depends upon the type of compound to be extracted (Handa, 2008). Water used as solvent for extraction of phenolic acids, alkaloids, flavonoids, lectins, glycosides and phenylpropanoids contents (Chikezie *et al.*, 2015). Tannins, polyphenols, flavanol, terpenoids and alkaloids extracted in Ethanol. On the other way saponins, tannins, polyphenols, terpenoids, flavones and anthocyanin extracts obtained by Methanol solvent (Azmir *et al.*, 2013). Acetone used to extract alkaloids, sugars, tannins and quinones from plant material (Neelamathi and Kannan, 2016). Chloroform could also be used for extraction of the bioactive

compounds like anthocyanin, flavonoids, flavones and polyphenols (Selvamuthukumar *et al.*, 2017).

Other than conventional methods there are some modern techniques like ultrasonic assisted extraction and microwave assisted extraction are also used for preparation of phytoextracts.

Fungal diseases

Fungal pathogens cause quantitative and qualitative loss in yield of nutritionally and economically important vegetable crops of family Solanaceae, Brassicaceae, Cucurbitaceae (Salau *et al.*, 2015). Irish famine 1845-1846 due to potato blight caused by *Phytophthora infestans* adversely malign the crop globally. Microbial pathogens cause 16% loss in agriculture production and fungal pathogens responsible for 70-80% of these losses (Moore *et al.*, 2011). Variety of fungal pathogens belongs to genera *Aspergillus*, *Alternaria*, *Colletotrichum*, *Phomopsis*, *Fusarium*, *Phytophthora*, *Penicillium*, *Fusarium*, spoiled the fresh vegetables and cause serious economic losses (Tournas, 2005).

Diseases of tomato (Lycopersicon esculentum)

Early blight of tomato

Peculiar leaf spots, fruit rot and stem lesions formed that caused 32%-57% crop losses (Mehta and Sharm, 2016). Early Blight caused by *Alternaria solani* that form small circular brown or dark brown concentric rings that coalesce form patches in results occur defoliation (Mamgain *et al.*, 2013). *A.solani* foliage infection known as Early Blight, infection on stem seedling collar rot and on fruit called as fruit rot that cause infected fruit drop before maturity. (Joshi *et al.*, 2018). Leaf extracts of *Datura stramonium*, *Allium sativum*, *Azadirachta indica* with 5% concentration in invitro exhibit maximum mycelial growth reduction while in green house experiment extracts of *Datura stramonium*, *Allium sativum*, with 5% concentration significantly enhance the fruit yield of tomato (Nashwa and kamal, 2013). Garlic bulb (*Allium sativum*) extract, *Eucalyptus camaldulensis* and *Azadirachta indica* leaf extracts proved effective against early blight (Chourasiya *et al.*, 2013). Aqueous extracts of neem, lemon grass leaves and garlic bulb

reduce mycelial growth of *A.solani* and disease severity (Afifi *et al.*, 2009). Extracts of *Azadirachta indica* (Neem), *Eucalyptus camaldulensis* (sufeda), *Allium sativum* (garlic), *Parthenium hysterphoresis*, and *Datura stramonium* significantly inhibited mycelial growth. *Azadirachta indica* most effective botanical that inhibited the mycelial growth invitro due phenolics compounds and tetra terpenoids. Antifungal Allicin present in *A.sativa* while specific phytochemical also reported in *Datura stramonium* (Raza *et al.*, 2016). Aqueous and ethanolic extracts of *Calatropis procera*, *Acacia nilotica*, *Achillea fragrantissima* suppressed mycelial growth and germination due to bioactive constituents. Ethanolic extracts of *C.procera* most effective against disease severity under field conditions (Baka and Rashad, 2016). Application of leaf extract of *Azadirachta indica*, *Carica papaya*, *Euphorbia heterophylla* and *Tithonia diversifolia* significantly influenced the early blight of tomato and fruit yield. *Carica papaya* proved most effective against disease severity thus could be used in management of early blight (Joseph *et al.*, 2015).

Anthracnose of tomato

Anthracnose characterized as dark brown small sunken lesions forms on fruits caused by *Colletotrichum* species as result infected fruits have short shelf life (Saini *et al.*, 2017). *Colletotrichum* has a wide range of hosts like vegetables, cereals, legumes and perennial crops. Initial infection involves attachment of conidia to plant surface, germination of conidia, formation of appressoria, penetration to epidermis, colonization, Production of acervuli and sporulation (Than *et al.*, 2008). Ethanolic extracts of *Azadirachta indica* and *Nicotiana tabacum* show antifungal activity against *Colletotrichum*. Mycelial inhibition increase with concentration. Plants extracts of Neem and tobacco are ecofriendly and effective replacement of chemical fungicides (Opeyemi *et al.*, 2018). Fruit extracts of *Rhus coriaria* lowered disease severity, reduced the mycelial growth and increased the fruit yield. Aqueous extracts of *R.coriaria* used as better ecofriendly, cost effective natural fungicide in management of Anthracnose (Rashid *et al.*, 2018).

Leaf and rhizome extracts of *Curcuma longa*, leaf extract of *Tagetes erecta* and rhizome extract of *Zingiber officinales* most effectively inhibited conidial germination of *Colletotrichum* (Imtiaj *et al.*, 2005). *Lantana camara* superiorly inhibited mycelial growth and spore germination as compared to other botanicals. Plant extracts are safe and healthy for mammals and as well as for environment (Ademe *et al.*, 2013). In vitro and in vivo Aqueous, ethanolic and acetonetic leaf extracts of *Thevetia peruviana* showed fungicidal activities by mycelial reduction and spore germination suppression due to presence of bioactive compounds like alkaloids, sugars, terpenes, sterols, Flavonoids and coumarins (Dooh *et al.*, 2014). Strong fungitoxic activity against *Colletotrichum* shown by stem and root bark of *Azadirachta indica* and *Vernonia amygdalina* (Nduagu., 2008). *Cestrum nocturnum* and *Annona cherimola* leaf extracts gave better antifungal effect against conidia germination of *Colletotrichum* (Hernandez *et al.*, 2007).

Late blight of tomato

Late blight is a destructive disease of tomato characterized by blackening of leaves, rotting of fruits with chocolate brown or marbled brown greasy spots caused by *Phytophthora infestans* (Griffith *et al.*, 1995). First LB lesions appeared at top of the stem or at node then further spread through plant body lead to defoliation and rendered green fruits. Significant economic annual crop losses happened due to ability of *P.infestans* to overcome resistant genes of plant and have greater genetic variations due to reproduction by rapid sexual mating (Nowicki., 2013).

Extracts of *Tephrosia vogelli* and *Entandrophragma angolense* significantly cause inhibition of sporangial germination and reduced disease severity (Goufo *et al.*, 2012). Neem and garlic extract in In vivo situation proved affective against late blight (Yadav *et al.*, 2017). Antifungal potential of *Azadirachta indica*, *Moringa olifera*, *Zingiber officinales*, *Citrus calosynthis* and *Calotropis gignentia* extracts evaluated against late blight of tomato and significant reduction being noted in disease severity (Yousaf *et*

al., 2015). Leaf extracts of *Hedera helix*, and *Paeonia suffruticosa* exhibited high degree of disease suppression (Rohner *et al.*, 2003). *Inula viscosa* leaf extracts strongly inhibited *P.infestans* due to presence of terpenoid, sesquiterpene, sesquiterpene lactones, flavonoids and other compounds (Wang *et al.*, 2004).

Diseases of potato (*Solanum tuberosum*)

Late blight of potato

Pale brown to purplish black spots at leaves caused by *Phytophthora infestans* that lead to formation of white spores under leaves around necrotic edges which fall and germinated in soil to infect tubers with reddish brown purple areas. Crop loss highest in Sub Saharan Africa highest about 44% due to late blight of potato (Arora *et al.*, 2014). Aqueous Extracts of nine medicinal tested against *P.infestans* in vitro and in vivo conditions and the extracts of these plants successfully suppressed mycelial growth, germination of spores and showed notable reduction in diseases severity specifically major reduction exhibited by leaf extracts of Lemon grass(*Cymbopogon citratus*), basil leaves (*Ocimum bacilicum*) and Chilies fruits(*Capsicum frutescens*) (Khair *et al.*, 2007). Aqueous Extract of sundried leaves of *Podophyllum hexandrum* at 25%W/V effective against disease incidence and gave higher yield of potato as compared to extracts of *Withania somnifera* and *Xanthium strumarium* under field conditions (Majeed *et al.*, 2011). Extracts of *Rheum rhabarbrum* (rhizome) and *Sophora flavescens* with 2% concentration showed 91.67% and 75% efficacy against disease on seedling (Wang *et al.*, 2007). *Acorus calamus*, *Terminalia bellerica* extracts in acetone, hexane and methanol solvents provide highest efficacy against late blight of potato under both in vitro, in vivo experiments. Applications of phytoextracts are ecofriendly and non-toxic to animals and environment (Anju *et al.*, 2019).

Potato dry rot

Potato dry rot caused different *Fusarium sp.* That cause 25% crop loss and 60% storage tubers infection. Dark depressions with crinkled concentric rings form

brownish blackish necrotic regions as result tuber rot out (Sandipan *et al.*, 2016). *Fusarium solani*, *Fusarium oxysporum* and *Fusarium sambucinum* are most common and destructive species than other *Fusarium* spp. that caused potato dry rot (Stefanczyk and Sobkowiak, 2016). Dry rot is a postharvest disease of potato caused serious economic losses in world. Eleven potato cultivars susceptibility to *Fusarium* species studies found that *F.sambucinum* most assertive as compared to other species (Trabelsi *et al.*, 2002).

Foliar applications of Saponins contents of *Chenopodium quinoa* extract minimized the Dry rot severity (Almaghrabi *et al.*, 2010). Methanolic extracts of Eucalyptus and Artemisia at 5%, 10%, and 15% concentrations deceased mycelial growth in invitro and in vivo and reduced dry rot development in tubers (Zakir, 2014). Aqueous and organic extract of *Sargassum vulgare* showed efficacy against Dry rot severity (Ammar *et al.*, 2017). Methanolic extracts of *Punica granatum* could be used as natural fungicide against fusarium as it reduced the 75.5% fungal mycelial growth (Elsesherbiny *et al.*, 2015). Fungitoxic abilities of *Nicotiana tabacum*, *Aloe vera* and neem extracts evaluated against dry rot causing fungal pathogens in vivo. Tobacco extracts cause maximum decrease due to presence of tannins while all three extracts have Alkaloids and flavonoids contents (Taiga and Friday 2009).

Black scurf disease of potato

Brown black sclerotia developed on tubers surface by soil born pathogen *Rhizoctonia solani* that limits the growth of underground tubers cause serious economic loss to potato production. *R.solani* remains symptomless during colonizing phase until infection to be establish in form of short swollen cells (Kumar *et al.*, 2017). Irregular, superficial, flat dark brown to black masses of sclerotia appeared at tuber surface as result of *R.solani* infection (Malik *et al.*, 2014). Plant extracts of *Azadirachta indica*, *Allium sativum*, *Eucalyptus camaldulensis*, *Allium cepa* and *Peganum harmala* cause 71%, 56%, 48%, 41% and 34% growth inhibition of *R.solani* in lab while under

green-house conditions *Azadirachta indica* give 62% eyes germination of potato (Atiq *et al.*, 2014).

Extracts of Neem, garlic, shetodron (*Leucas aspera*), *Swietenia mahagoni* and bishkatali (*Polygonum hydropiper*) highly effective to suppress radial growth of *R.solani* due to their unique antifungal capability (Sifat and Monjil, 2017).

Six plant extracts tested against *R.solani* and *Cannabis sativa*, *Peganum harmala*, *Datura stramonium* majorly reduced fungal spread as they possessed antifungal compounds. Flavonoids, glycosides, aldehydes, ketones esters and lactones constituent of *C.sativa*. Tannins, scopolamine, atropine are partof *D.stramonium*, similarly tetrahydroharmine, tetrahydroharmol are integrant of *P.harmala* that have antifungal potential (Hussain *et al.*, 2014).

Diseases of eggplant (Solanum melongena)

Early blight of eggplant

Early Blight caused by a Fungus called *Alternaria solani* usually Small circular concentric brown necrotic spots formed that extend and cause leaf senescence (Nasehi *et al.*, 2012). Collar rot on stem or stem lesions, fruit rot on fruit, concentric rings with yellowish zone leads to premature defoliation and heavy fruit drop caused by *Alternaria solani*. It required favorable High temperature, humidity and heavy rainfall for reproduction (Chaerani and vorrips,

2006). Aqueous extracts of *Arica papaya L.* and *Azadirachta indica* contained antifungal compounds that quench mycelial growth of *A.solani*. (Suleiman, 2010). *Dodonaea viscosa* have inhibitory effect on mycelial growth of *Alternaria*. *Dodonaea viscosa* exhibited (56.96%) maximum inhibition of *Alternaria solani*. Plant extract could be used for management of fungal disease caused by *A.solani* (Aslam *et al.*, 2010). Extract of *Allium sativum* cause 100% inhibition of mycelial growth *A.solani* at both 10% and 20% concentrations meanwhile *Datura stramonium* leaf extract cause 65.17% and 76.43% reduction respectively (Kumar *et al.*, 2018). Foliar

extracts of *Lauris nobilis*, *Salvia officinalis*, *Humus lupulus* and *Cirsium arvense* limited the radial mycelial growth of *Alternaria solani* (Yanar *et al.*, 2011).

Fruit rot of brinjal

Brownish halo, greyish sunken spots that enlarged to form yellow and brown necrotic regions of concentric rings. *Phomopsis vexans* cause fruit rot of brinjal in results serious economic crop production loss occurs (Mahadevakumar *et al.*, 2016). *P.vexans* lessened the yield and market value qualitatively and quantitatively up to 50% while 12%-25% yield losses occurred due to flower and fruit rot. Extracts of neem, Datura, garlic, and *Trichoderma sp.* reduced germination of spores, fungal growth and disease severity. Zeatin, citric acid and gluconic acid bioagents of *Trichoderma sp.* have potential to promote the yield of eggplants (Reddy *et al.*, 2018).

Aqueous Bulb extract *Allium sativum* bark extract of *Terminalia arjuna* and root leaf and seed extracts of *Ricinus communis* reduced fungal mycelial growth. Aqueous extracts with 15% concentrations of *Allium sativum* (100%), *Rccinus communis* seed, root and leaf extracts caused 77.1%, 55.5%,53.8% inhibition respectively while bark extract showed only 27.5%inhibition (Das *et al.*,2018). *In vitro* evaluation of garlic (*Allium sativum*) clove, kokum (*Garcinia indica*) fruit, turmeric (*Curcuma longa*) rhizome and onion (*Allium cepa*) bulb extracts at 5% and 10% represented intensive *P.vexans* suppression as compared to other to to clerodendron (*Clerodendron inermis*), lantana (*Lantana camera*) leaf extracts (Jakatimath *et al.*, 2017).

Damping off in Brinjal

Pre and post-emergence damping off in brinjal caused by *Pythium spp.*, *P.aphanidermatum*, *P.irregulare* and *P.ultimum* in result softening, water soaking, rots of collar portion of the seedling occurs for ultimate death of seedling. *Pythium sp.* is parasitic, nonspecific to host have wide range of host including species of family *Solanaceae*, *Amaranthaceae*, *Rosaceae*, *Leguminosae*, *Cucurbitaceae* and

Malvaceae (West *et al.*, 2003). White cottony mass developed by rapid germination for aggressive prevalence and acts as limiting factor for eggplant crop (Parveen and Sharma, 2014). Neem leaf extract 75%, Garlic clove extract 67%, Turmeric extract 59%, Datura leaf extract 63%, Onion bulb extract 62%, Ginger extract 56%, Tulsi leaf extracts cause 54% mycelial inhibition @10% concentrations of all extracts. Percentage of seed germination of brinjal was high with neem leave and garlic clove extract (Choudhary *et al.*, 2017). Efficacy of 10 phytoextracts tested *in vitro* against *Pythium* pathogen at three different concentrations.

Garlic (*Allium sativum*) most effective against radial mycelial growth followed by Adulsa (*Justicia adhatoda*), Datura (*Datura stramonium*) and mehndi (*Lawsonia inermis*). Phyto-extracts could be better choice as fungitoxic material as compared to harmful synthetic fungicides (Gholve *et al.*, 2014).

Aqueous, n-butanol and methanolic extracts of *Lawsonia inermis*, *Tephrosia pupurea*, *Vinca rosea*, *Phyllanthus niruri* and *Mimosa pudica* applied to *Pythium debaryanum*. Methanolic extracts showed high antifungal abilities as compared to aqueous and n-butanol which showed no antifungal activity (Ambikpathy *et al.*, 2011).

Conclusion

Application of chemical fungicides against diseases fungal diseases of vegetables lead to toxicity and bioaccumulation of heavy metals that are hazardous to human health. Plant extracts are natural organic Bio-degradable, ecofriendly fungicides. Natural extracts proved beneficial for pre-harvest and post-harvest management of fungal diseases. Plant extracts need specific application attentions to lower the deleterious effects of chemical fungicides on human and other life exists on biosphere. It could be better choice to replace synthetic fungicides and pesticides with respect to future perspective. Phytoextracts could be easily accessed and cheapest source to maintain plant health as compared to synthetic fungicides that damaged natural ecosystem.

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