International Journal of Plant Pathology and Microbiology

E-ISSN: 2789-3073
P-ISSN: 2789-3065
IJPPM 2022; 2(2): 51-54
Received: 13-09-2022
Accepted: 15-10-2022

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Seed borne nematode of small millets: A review

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Abstract
Among seven small millets, only foxtail millet (*Setaria italica*) and proso millet (*Panicum miliaceum*) were found infested with a seed borne nematode, *Aphelenchoides besseyi*. Basically, the nematode is a serious pest of rice and causes whip tip of paddy, but significant infestation was also recorded in foxtail millet and proso millet. Green foxtail millet (*Setaria viridis*), crab grass (*Panicun sanguinale*) and guinea grass (*Panicum maximum*) were also found infested with *A. besseyi*. It causes Ear blight or Light ear disease in foxtail millet. Characteristic symptoms of nematode infestation were distortion and spreading of panicles, production of chaffy grains and delayed maturity. Maximum nematode population was recorded in the seeds having black seed coat and abnormal seeds of both the millets. Variation in morphology and morphometric attributes of nematodes was recorded among rice, foxtail millet and proso millet isolates. Amphimictic mode of reproduction was reported in the populations of *A. besseyi* in foxtail and proso millet. More survival of nematode was found in proso millet as compared to rice. Auxenic culture of *Alternaria alternata* and *Curvularia lunaat* supports the growth and reproduction of *A. besseyi* in proso millet. Number of females over one male was maximum in foxtail millet (5.25) followed by rice (3.41) and proso millet (3.16). Lowest population of nematode was recorded in foxtail millet cultivars RFM 21, 23, 51, 68, 74, 76, 84, 99, SiA 3156 and SiA 3190. and proso millet cultivars DHPM 50-1-1, TNAU 137, TNAU 149, TNAU 151, TNAU 183 and TNAU 191. Adverse effect of herbicides *viz*. Propaquizafop and Bispyribac sodium on survival and mortality of *A. besseyi* was observed.

Keywords: Foxtail millet, proso millet, seed borne nematode, *Aphelenchoides besseyi*

Introduction

Small millets is a collective term referring to a number of annual small seeded cereal crops usually cultivated as grain and fodder crops in a variety of agro-ecological conditions in diverse soils and varying rainfall. The most important small millets are finger millet (*Eleusine coracana*), little millet (*Panicum sumatrense*), barnyard millet (*Echinochloa frumentacea*), foxtail millet (*Setaria italica*), kodo millet (*Paspalum scrobiculatum*), proso millet (*Panicum miliaceum*) and brown top millet (*Bracharia ramosa*). These crops are nutritionally superior to other cereals in terms of protein, fibers, minerals and vitamins, now referred as *nutri-cereals*. They conserve the soil water level and can produce significant yield at marginal soils. Among biotic stresses, nematode infection is one of the factors causing severe damage to crop and seed quality (Esser, 1991, Tiwari, 2014, Sanyal *et al*. 2020) [7, 30, 24]. Plant parasitic nematodes associated with small millet crops, their distribution, biology, interrelationship with other plant pathogens and management have been reviewed (Jain, 2009) [11]. Nematode problems in millets and their management were discussed by Nayak (2010) [14]. Most of the plant parasitic nematodes are soil inhabiting and few are seed borne pathogenic nematodes exhibiting special adaptive mechanisms. Seeds are regarded as highly effective means for transporting plant pathogens for long distance. Veerbhadra (1992) [25] studied the nematode parasites of finger millet and foxtail millet. Youssef (2014) [28] reviewed the study of leaf and bud nematode, *Aphelenchoides besseyi* in relation to its identification, economic importance and control measures. Shanmuga Priya *et al*. (2019) [16] conducted survey to explore the existence of plant parasitic nematodes and investigated the community structure of the phytonematodes associated with millet crops in Tiruvannamalai district of Tamil Nadu. Foliar nematode, *Aphelenchoides besseyi* is one of the important seed borne plant parasitic nematode of rice and millets. Proso millet, foxtail millet, green foxtail millet (*Setaria viridis*) and guinea grass (*Panicum maximum*) were reported to be infested with *A. besseyi*. Proso millet commonly known as *Cheena* is gluten free, low glycemic index, high protein and fibers. The crop is highly adapted to dryland cropping systems with high water use efficiency and short growing season. It can be grown in marginal lands and can give a significant amount of yield compared to other major cereal crops.

The present review is concentrated on *A. besseyi*, the seed borne nematode of foxtail millet and proso millet in relation to its distribution, symptoms, morphology, reproduction and management.

1. Foliar nematode (*Aphelenchoides besseyi* Christie, 1942)

Distribution and economic importance

Aphelenchoides besseyi is an economically important widely distributed seed borne nematode of rice and millets. It can survive in stored seeds for several years under dry conditions. *A. besseyi* was reported first time on foxtail millet (*Setaria italica*) from Japan (Yoshi and Yamamota, 1950) [27], India (Dave *et al*, 1979, Lal and Mathur, 1988) [6, 12] and China (Cui *et al*., 1989, Chen *et al*, 2022) [5, 2]. Lal and Rajan (1995) [13] collected 1138 accessions of foxtail millet from diverse geographical regions of India and examined for seed borne nematodes by soaking the seeds in water for 24-48 h at 30 ^{0}C. About 8% (91) accessions were found infected with A. besseyi with inoculum level varying from 2 to 36 per seed. Foxtail millet is a major cereal crop in Northern China especially in the semi-arid areas and is an important domestic quarantine nematode in Henan province of China. The average number of nematode per earhead of foxtail millet was up to 1738.75±107.72 (Chen *et al*, 2022) [2]. The disease caused by *A. besseyi* in foxtail millet was referred as Ear blight of italian/foxtail millet (Yoshi and Yamamota, 1950) [27] and Light ear disease of foxtail millet (Dave *et al*, 1979, Shukla *et al*, 1997) [6, 23]. About 10% seed samples of foxtail millet were found infected with bud and foliar nematode (*A. besseyi*) during survey (1980 to 1985) in Madhya Pradesh, India (Shukla *et al*, 1997) [23]. Chouhan (2014) [3] studied the population dynamics of *A. besseyi* in germplasm of foxtail millet and proso millet. Maximum nematode population per 250 seeds was recorded in the seeds of proso millet having black seed coat (284) followed by off white (198), mixed (147) and slight red seed coat (146.5). Similar observations were recorded in foxtail millet showing black seed coat (185.6), off white (87.1), slight red (42.1) and mixed (41.3). Population of nematode was maximum in apparent bold seeds (237.5) as compared to apparent small sized seeds (168) in proso millet, where as in foxtail millet highest population was recorded in small sized seeds (91.5) as compared to bold sized seeds (58.5). Abnormal seeds exhibited more number of nematodes as compare to apparent healthy seeds in proso and foxtail millet. Wang *et al*. (2015) [26] studied morphological characteristics and measurements of the nematode isolated from infected ears of foxtail millet in Hebei province of China for identification of pathogenic nematode. They amplified its 28S rRNA-D_2/D_3 and ITS regions of ribosomal RNA, sequenced, aligned and the phylogenetic trees were constructed by Neighbor-joining method using MEGA 6.0 software. The results showed that the population was 100% similar to *A. besseyi* and the similarity of ITS was 98 to 99%. Sharma *et al*. (2017c) [22] reported that loss in proso millet due to *A. besseyi* is correlated with nematode population in the seeds.

Host

The foliar nematode is known to occur on a wide range of hosts. Among different small millets, foxtail millet (*Setaria italica*) and common millet or proso millet (*Panicum miliaceum*) were reported prone to *A. besseyi* infection. Yoshi and Yamamota (1950) [27] reported that *Setaria viridis* (Green foxtail) and *Panicum sanguinale* (crab grass) may be infected with the nematode. Guinea grass (*Panicum maximum*) were also found infested with *A. besseyi* (Garcia *et al*, 2000) [8].

Symptoms

The concomitant infection of *Aphelenchoides besseyi* and *Phoma* sp. was reported in the panicles of foxtail millet under field conditions showing discolouration in irregular patches. In the discoloured grains, few nematodes were found and grain formation in such panicles was not observed. (Dave *et al*,. 1979) [6]. Chouhan (2014) [3] observed distortion in panicles, sterile grain in the posterior region of the panicle and late maturation of earhead in foxtail millet. Sometimes completely sterile glumes were also observed. Chen *et al*. (2022) [2] reported that some upper leaves showed chlorosis without or with necrotic tips. Flag leaves presented crinkling and distortion, coloured stalks, vertical ear heads, open brown to light black glumes and thin grains due to infestation of *A. besseyi* in foxtail millet. In proso millet, distortion and scattering of earhead, chaffy grains and poor grain set in the earheads were observed due to infestation of *A. besseyi* (Chouhan, 2014) [3]. Sharma *et al*. (2017b) [21] reported that panicles of infected proso millet plant showed distortion, delayed maturity and often failure of panicle emergence. If panicle emerged, sterile and chaffy seeds were observed. It was also observed that floral primordial formation is necessary for the successful transmission of nematodes to the seeds.

Morphology

Sharma *et al*. (2017a) [20] studied the morphometric attributes of *A. besseyi* in rice, foxtail millet and proso millet. Greater body length and width of *A. besseyi* was recorded in rice (681.97 µm and 18.43 µm) followed by foxtail millet (665.62 µm and 17.59 µm) and proso millet (541.86 µm and 18.22 µm). Length of metacarpus and posterior glands in female was maximum in rice isolate followed by foxtail millet and proso millet. Similar trend was observed in tail length of female and gonads of *A. besseyi*. Sharma (2018) [18] also reported variation in morphology and morphometric attributes of *A. besseyi* populations. Larger body length, greatest body width, oesophageal length, tail length of female and male, vulva of female, spear and spicules of male were greater in rice isolates than the foxtail millet and proso millet isolates. Chen *et al*. (2022) [2] characterized the females isolated from foxtail millet were slender with a short stylet, an oval metacorpus with a distinct valve, a labial region slightly wider than the first body annules and a conoid tail with a terminus bearing a star shaped mucro with four pointed processes. The males had three pairs of ventro-submedian papillae with the first one adanal, spicula curved with a slight basal process, terminus bearing four mucrons arranged variably and the whole worm was in 'J' shape.

Reproduction and growth

Gotke *et al*. (2001) [10] evaluated 16 populations of *A besseyi* from different hosts and geographical areas for mode of reproduction and host range. Amphimictic mode of reproduction was found in the populations of *A besseyi* from *S. italica* and *P. miliaceum* collected from Bangalore, India. Chouhan (2014) [3] studied the ratio of male and female nematodes in the seeds of proso millet and foxtail millet. Female nematodes were found continuously greater in number as compared to male nematodes. During diapauses period females of *A. besseyi* were encountered tightly coiled compared to males which were loose coiled below the hull (Sharma *et al*. 2017a) [20]. Axenic culture of *Alternaria alternata* and *Curvularia lunata* favours the growth and reproduction of *A. besseyi* in proso millet, whereas *Fusarium* sp., *Penicillium* sp. and *Bipolaris* sp. fail to reproduce nematode population (Sharma *et al*, 2017b) [21]. Sharma (2018) [17] recorded highest (5.25) females over one male in foxtail millet followed by 3.41 female in rice and 3.16 female over one male in proso millet. Mode of seed infestation of *A. besseyi* in proso millet was studied by Sharma and Tiwari (2017) [24] and suggested more survival of *A. besseyi* in proso millet as compare to rice. Zhenjum *et al* (2019) [29] examined 99 foxtail millet seed samples collected from different millet production regions of China. A PCR based detection method was established to detect *A. besseyi* in foxtail millet seeds with a good specificity and high sensitivity, which could be used to detect the foxtail millet seeds carrying *A. besseyi*. Out of 99 seed samples, 33 samples were detected to be contaminated, in which contamination rates ranged from 33.3 to 100.0% and they were mainly distributed in spring foxtail millet areas. The foxtail millet nematode is a major disease in summer millet areas, but with the millet exchange between spring and summer millet area, it has become a disease in spring millet area in China.

Management

Meager information in available regarding management of *A. besseyi* in small millet crops. However, few studies on identification of resistant sources and non-target effect of herbicides on nematode population were carried out.

Cui *et al*. (1989) [5] identified 1919 foxtail materials resistant to *A. besseyi* during 1984-1988. Twenty two resistant materials were rescreened. Among them five were poly resistant and five were amphi-resistant sources. Peroxidase isoenzymes were markedly different between resistant varieties and infected varieties. the disease resistance was interrelated with the millet growth period, the density of spikelets and the length of seta.

Chouhan (2014) [3] evaluated 19 proso millet germplasm and none was free from *A. besseyi* infestation. The population of nematode was between 50 to 500 nematodes per 250 seeds. Lowest population was found in DHPM 50-1-1 (50) followed by TNAU 151 and TNAU 183 (51-100 nematodes). Chouhan (2014) [3] evaluated 46 foxtail millet germplasm and 16 germplasm were free from nematode infestation. The population of nematode was between 0 to 528 nematodes per 250 seeds. Least population (up to 50 nematodes per 250 seeds) was recorded in RFM 21, 23, 51, 68, 74, 76, 84, 99, SiA 3156 and SiA 3190. Sharma (2015) [18] screened 9 proso millet germplasm against *A. besseyi* and nematodes per seed ranging from 3 to 8 were recorded. None of the germplasm was found free from nematodes. However, lowest population of nematode in 250 seeds was recorded in TNAU 191 (90) followed by TNAU 183 (115), TNAU 137 (118) and TNAU 149 (121). Whereas highest population was in TNAU 194 (256) followed by TNAU 155 (249), TNAU 145 (238) and TNAU 164 (237).

Gotke and Mathur (1993) [9] reported that pre-soaking of seeds in 1% H_2O for three hours followed by hot water treatment at 48 ^{0}C for 15 minutes is best treatment for eradication of *A. besseyi* from seeds of foxtail millet and proso millet. Without pre-soaking, complete kill was achieved by exposing the seeds to 50 ^{0}C for 15 minutes. Bhag mal *et al*. (1999) [1] reported the elimination of *A. besseyi* from seeds of proso millet and foxtail millet by pre-soaking the seeds in 1% hydrogen peroxide. *In vitro*, efficacy of thirteen herbicides was tested against mortality of *A besseyi*. Positive correlation between adverse effect of herbicides on the survival and death of *A. besseyi* was encountered. All 13 herbicides were found significant. However, Propaquizafop and Bispyribac sodium affected the nematode population in terms of maximum mortality. Whereas, Flufenacet exhibited lowest effect on nematode mortality.

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