

Original Research Article

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**Serious Leaf Spot Disease Problem of *Calotropis procera* (Aiton) W.T.Aiton.  
by *Alternaria alternata* in Gurgaon (Haryana), India**

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*Calotropis procera* (Aiton) W.T. Aiton. (Asclepiadaceae) commonly known as Madar or 'Aak' is a medicinally important wild shrub native to India useful in treatment of a number of illnesses including dysentery, intermittent fevers, cold, cough, and asthma. We observed a serious leaf spot disease of madar growing on wasteland sites near about AUH in the Panchgaon, Manesar- Gurgaon (Haryana) in year onward 2014. In January and February 2017, we observed this leaf spot to be very prevalent or on almost all plants in the vicinity. Disease incidence was greater than 90% and caused extensive abscission of leaves. Symptoms on infected leaves were first observed in end of October to November of each year as small, circular spots with large yellow to dark brown halos on both sides of the lower and upper leaves. The spots gradually enlarged in size. Soon they became irregular in shape or remained circular with concentric rings or zones. This is a new record from Aravali Hills near AUH at Panchgaon, Gurgaon Haryana.

**Introduction**

'Aark' plants (*Calotropis procera* (Aiton) W. T. Aiton.) of family Asclepiadaceae has been in use for sun worship since Vedic times. This plant is also considered very special to Lord Ganesha. The leaves and the flowers are offered to the deity, especially during Ganesh Chaturthi. The plant is also sacred to Lord Shiva and also to some of the village deities. One of these is Arkamma, named after the plant.

*Calotropis procera* (Aiton) is a spreading shrub of up to 2.5 m of height after attaining age of 3-5 years. It is a multipurpose shrub commonly known as Madar in Hindi. Plant

forms a deep taproot system up to 3-4 m in depth. The stems are crooked and covered with a fissured corky bark and yield a fiber useful for making ropes, bags, nets and paper (Orwa *et al.*, 2011).

This plant is under use since ancient time in Indian traditional system of medicine mainly for treatment of leprosy, ulcers, tumors, piles and diseases of spleen, liver and abdomen. The traditional folk healers use the milky sap or latex of *C. procera* for preparation of different drugs and ailments mainly for treatment of boils, infected wounds, eczema and rashed skin (Khare, 2007). In Nigerian

traditional medicine, *C. procera* latex is either used alone or with other herbs to treat common diseases such as fever, leprosy, dropsy rheumatism, indigestion, cold, eczema and diarrhoea. (Herbal monograph). The processed *C. procera* latex is used in treating vertigo, baldness, hair fall, tooth aches, intermittent fevers, rheumatoid/joints swellings and paralysis (Kew, 1985). Its leaf latex is applied on fresh cuts to stop bleeding immediately and is also used to treat boils (Vohra, 2004) and to remove thorn from body (Rai *et al.*, 2000). The dried aerial parts and roots are consumed as expectorant (Agharkar, 1991) purgative, improve digestion (Oudhia and Tripathi, 1997) and are also used to prepare an effective laxative. Dry root powder is used to good tonic to cure bronchitis, asthma, leprosy, eczema and elephantiasis, hepatic and splenic enlargement (Agharkar, 1991).

The *A. alternata* infection on Madar (*Calotropis procera*) has been reported from the Punjab state of India (Saini *et al.*, 1989) and on *Calotropis gigantea* (L.) R. Br. ex. Ait from Rajasthan, India (Sain *et al.*, 2009).

Since no work has been done regarding *A. alternata* infection on *Calotropis procera* plants in Aravali Hills near Panchgaon, Gurgaon region, Haryana a survey was conducted in Gurgaon region and infected samples were brought to the laboratory for mycological analysis.

### **Materials and Methods**

Naturally infected parts of *Calotropis procera* (Aiton) W.T. Aiton. (Asclepiadaceae) commonly known as Madar (living leaves, flower buds, stem, fruits) were collected in sterilized polyethylene bags brought to Laboratory. They were examined visually and and symptoms recorded. The scrapings were taken from different infected parts since March 2014 till recently.

Locations with the infection were recorded and field notes made regarding incidence of infection, nature of colonies, pathogenicity, locality, etc.

### **Microscopic investigations**

For identification of the fungus anatomical-morphological characteristics of fungus were used. They were observed by taking thin cross sections from naturally infected leaves. Symptoms on leaves with different stages of the disease were recorded and incitant examined under microscope.

The growth stage at which the leaf becomes susceptible to the infection was also recorded in Aravali Hills Panchgaon area. Infection was noticed young buds to mature leaves. The collected specimens of *A. alternata* were deposited in the Mycological Herbarium of the Institute, Amity University Haryana.

### **Culture**

In order to culture this fungus, Diseased leaf were surface sterilized in 0.1% sodium hypochlorite and washed with double distilled water. Freshly collected infected leaves were cut into 5mm pieces and placed on potato dextrose agar (PDA) of pH 7 in petridishes for growth of the fungus.

### **Pathogenicity**

**Method -1:** Pathogenicity was tried by spraying infected leaf (prepared by dissolving one 5mm diameter of dark brown bloom infection portion in 100ml double distilled water) on 5- 10 healthy *C. procera* seedlings. Plants were kept under observation up to 30 days.

**Method-2:** Koch's Postulates were proved by dusting conidia from the infected leaves on 5- 10 healthy young leaves of 30 *C. procera*

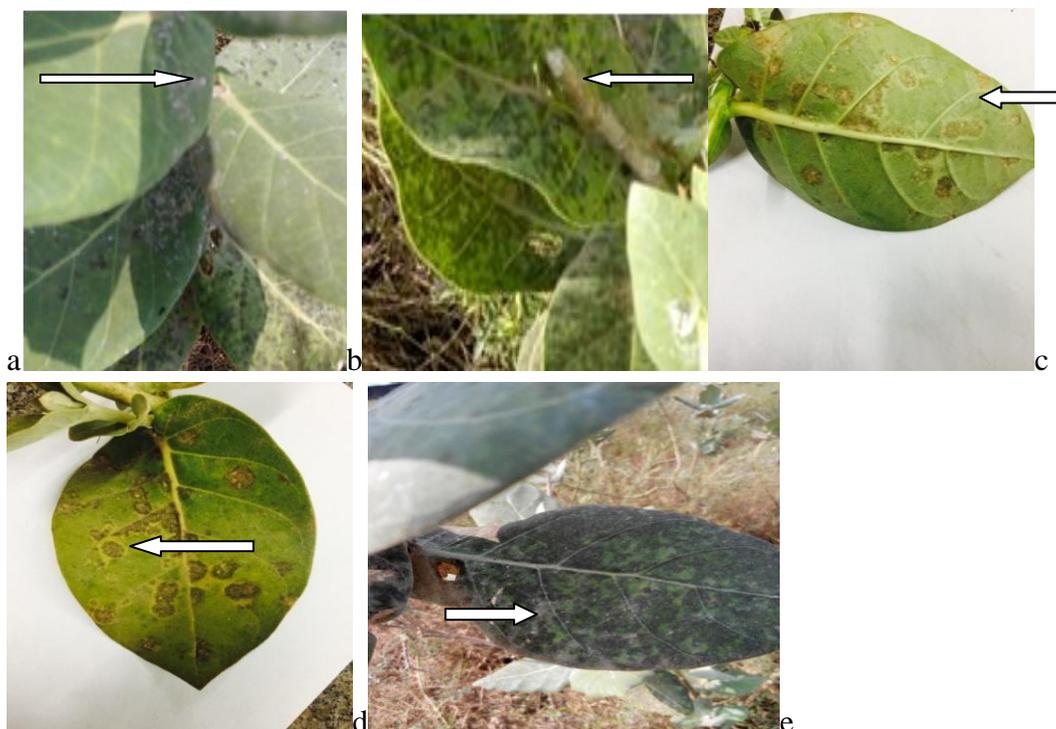
plants. A sterile brush was used to transfer conidia from the infected leaves to fully expanded leaves of healthy plants. For control, sterile brush was touched on healthy plant leaves. A plastic bag was used to cover each plant for 10-15 days.

### Results and Discussion

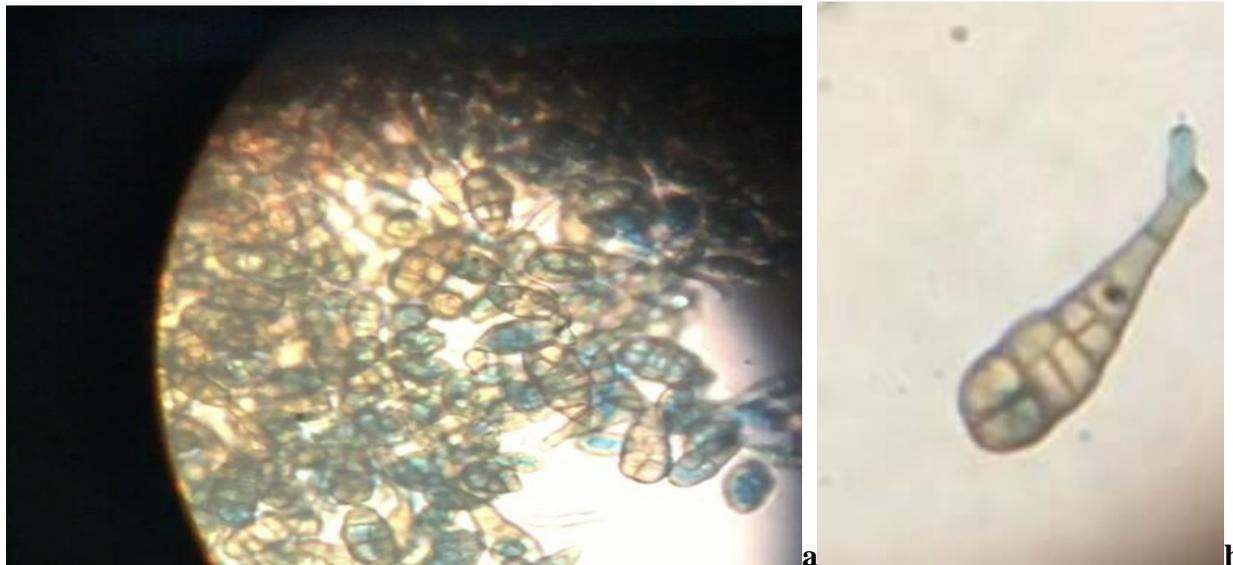
A survey of natural or wild *C. procera* plantations at various places of Panchgaon, Gurgaon from March 2014 to March 2017, revealed that Symptoms on infected leaves were first observed in end of October to November of each year as small, circular spots. The infection was seen on 30-60% plants in 2014 to 2016 but it was on almost every plant in the vicinity in 2017.

The symptoms of the disease was initially a few white small, circular (3-5mm diam) spots and these soon enlarge to yellow to dark brown spots with halo on upper sides of the leaf in November each year. This became visible in the month of December on the lower side of the leaf. The spots gradually enlarged further and became irregular in shape or remained circular with concentric rings or zones. Diseased spots were prominent on older leaves and lower stems and covering up to 90% area of upper and lower surface of leaf, highly reducing the photosynthetic area of the plant. Finally there was development of a dark brown bloom on infected leaves as the infection progresses (Fig.1 a,b,c,d,e).

**Fig.1** Symptoms of *Alternaria alternata* on the (a) *Calotropis procera* plant in Month of October 2016 having inoculum in white dusty colour (b) Initial stage of infection (c) Infection on upper leaf (d) Infection on lower surface (e) Infection covering 90% leaf surface



**Fig.2** Conidia of *Alternaria alternata* collected from naturally diseased leaf (a,b)



The infection was also found to be present on young flower buds and they failed to bloom. In field after 2-3 months of infection the leaves got distorted or curled, turned yellow and eventually dropped off.

Conidia on plants were pale to golden brown, obclavate, obpyriform, ovoid, or ellipsoidal with cylindrical beak having 5 to 8 transverse septa and a few longitudinal septa (Fig.2a,b). Incubated infected leaves showed development of Grayish black colonies with fluffy margins of *A.alternata*. Conidiophores arising singly or in small groups were 79.8 to 171.1  $\mu\text{m}$  in length and 3.1 to 6.2  $\mu\text{m}$  thick, simple or branched straight or flexuous, golden brown and smooth. The fungus produced dark brown conidia with a short, 2 to 5- $\mu\text{m}$  thick, pale beak. Conidia were 20-62  $\times$  9-17  $\mu\text{m}$  with 3 to 7 transverse septa. On the basis of the morphological investigations, the pathogen was identified as of *Alternaria alternata*. (Fr.) Keisseler.

Culture of the incitant *Alternaria alternata*. (Fr.) Keisseler from *Calotropis* leaf spot was easily cultured on PDA medium.

Leaf spots initially appeared on the upper surface of leaves within 6-7 days of spray inoculation and by 13 to 16 days leaf spots developed on every inoculated on lower leaves. *A. alternata* was consistently re-isolated from the inoculated plants. Method-2 pathogenicity test also produced similar Leaf spots and fungal colony morphology on both upper and lower surfaces of older leaves of inoculated plants in about 8-10 days. No symptoms developed in any of the healthy control plants.

A leaf spot of Madar (*C.procera*) caused by *A. alternata* has previously been reported from the Punjab (Saini *et al.*, 1989) and from Bahraich, UP (Kumar and Mall, 2012). This has also been reported on *Calotropis gigantea* (L.) R. Br. ex. Ait from Rajasthan, India (Sain *et al.*, 2009) and on *Stevia rebaudiana* from Bengal (Maiti *et al.*, 2006). This is the first record of the disease from the Aravali hills in Gurgaon district (Haryana) India.

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