Effect of Cellulose (A Polysaccharide) and Tomato Extract on Growth, Folin Positive Protein Production and Sporulation by Blight Pathogens

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Abstract

The growth, folin positive, protein production and sporulation by blight pathogens i.e. Alternaria solani (virulent pathogen) and Alternaria triticina (Avirulent pathogen) on modified Richard's liquid culture medium with tomato fruit extract of Pusa Ruby (Resistant variety) and Prince Long (Susceptible variety) having different concentration (0.10 and 1.0%) of cellulose (a polysaccharide) have been studied. The stimulation of folin-positive, protein production and sporulation was recorded whereas the growth was inhibited by the pathogens on both varieties when the concentration of cellulose was increased from 0.1 to 1.0% in the culture medium. The stimulation and inhibition depend upon the carbon source of the host and added carbohydrate (like cellulose) consisting of cellobiose units in long chain along with the host relations with the pathogen.

Introduction

Tomato (*Lycopersicum esculentum*), an economically important vegetable crop is often infected by the blight pathogens in the urban and rural areas of the Ranchi District of Jharkhand state. The blight pathogens *Alternaria solani* (Jones and Grout) and *Alternaria triticina* (Prabhu and Prasad) produce leaf blight disease on tomato and wheat. Growth, sporulation and production of pectic and cellulolytic enzymes have been reported in liquid media containing pectic or cellulosic carbon source[1]. A wide variety of carbon sources help to grow cellulolytic fungi and cellulolytic enzyme produced only in the presence of cellulose[2,3]. The pectolytic and cellulolytic enzymes are responsible for both maceration and killing effects during the disease development and are secreted in a routine manner as a feature of host-parasite interaction[4,5,6].

The present investigation is an attempt to study the growth, folin-positive, protein production and sporulation by blight pathogens i.e. *A. solani* and *A. triticina* on the extract of tomato fruit of resistant and susceptible varieties with different concentrations (0.10 and 1.0%) of cellulose.

Material and Methods

The blight pathogens *A. solani* and *A. triticina* were isolated from the leaf blight of tomato and wheat used for further investigation. The pathogens were grown on modified Richard's liquid culture medium and pH 6.0 was adjusted with phosphate buffer (0.1 M).

Extract of tomato fruit was obtained from 125 gm of healthy tomato fruit of Pusa Ruby and Prince Long variety and thereafter squeezed through centrifuge. 180-185 ml of each supernatant and 70-65 ml phosphate buffer (0.1 M) was mixed to prepare 250 ml double strength liquid medium to protect the strength of nutrients and finally 500 ml solution was prepared. Each solution was divided into 5 breakers measuring 100 ml and to each 100 mg, 250 mg, 500 mg, 750 mg and 1.0 gm cellulose was added to get 0.10%, 0.25%; 0.50%; 0.75% and 1.0% concentration. Thereafter 25 ml of different concentrations of cellulose was dispensed into each 250 ml Erlenmeyer flask to get 4 vials, 2 vials for *A. solani* and 2 vials for *A. triticina* of each variety. The medium without cellulose served as control. The flasks were autoclaved at Ca 1.06 Kg/Sq. cm for 15 minutes prior to use.

The experiment with a minimum of 20 cultures of both Pusa Ruby and Prince Long variety were repeated thrice. The cultures were maintained in dark at 24±1°C for a fortnight. Standard culture methods were followed[7,8,9].

Results and Discussion

The present investigation revealed the effect of cellulose in different concentrations (0.10%-1.0%) on growth, folin positive, protein production and sporulation by blight pathogens *A. solani* and *A. triticina* grown on Richard's liquid culture medium containing tomato fruit extract of both varieties. The growth was decreased by 4.0 and 36.2%

Key words : Cellulose, Tomato, A. solani and A. triticina.

with the increase of 0.1 to 1.0% of cellulose concentrations in culture medium where as the production of folin-positive, protein and spore was increased by 4.9 to 31.6%, 6.0 to 20.0% and 4.3 to 24.1% when *A. solani* (Virulent pathogen) was grown with the extract of Pusa Ruby

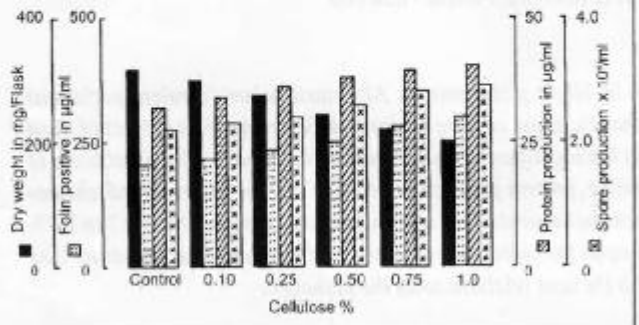

Fig. 1 : *A. solani* grwon on culture media containing extract of pusa ruby cultivar of tomato

(Resistant variety) (Fig. 1). When the same pathogen was grown with the extract of Prince Long (Susceptible variety) also the growth of the fungus was inhibited by 2.5 to 32.0% and the production of folin-positive, protein and spores were stimulated by 3.8 to 26.0%, 2.8 to 25.0%, 4.0 to 31.6% respectively (Fig. 2).

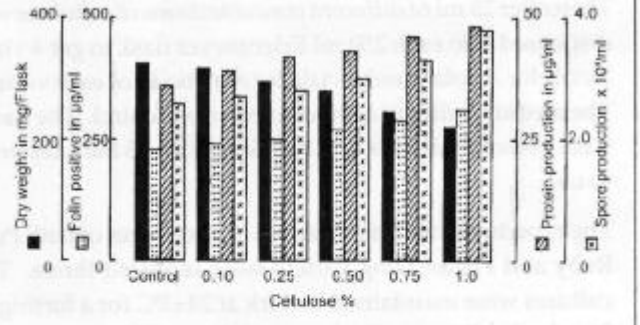

Fig. 2 : *A. solani* grown on culture media containing extract of prince long cultivar of tomato.

When the *A. triticina* (virulent pathogen) was grown with the extract of Pusa Ruby the growth was inhibited by 5.6 to 44.1% with the increase of cellulose concentrations from 0.1 to 1.0% whereas the production of folin-positive, protein and spore were increased by 8.5 to 24.5%, 5.0 to 32.1% and 4.2 to 18.0% (Fig. 3) respectively. The same pathogen when grown on the extract of Prince Long the growth was inhibited by 2.6% at 0.1% and 31.0% at 1.0% of cellulose. The folin positive was stimulated by 5.0 to 16.0%. Protein

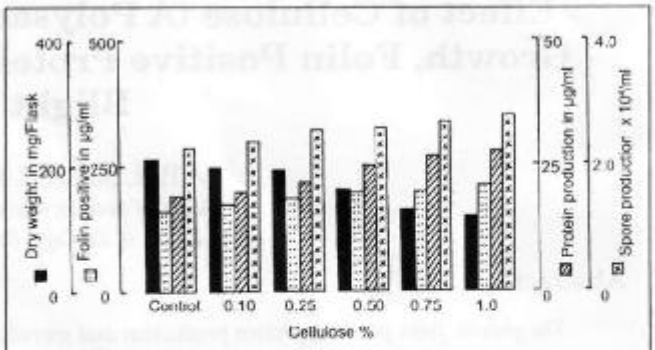

Fig. 3 : *A. triticina* grown on culture media containing extract of pusa ruby cultivar of tomato.

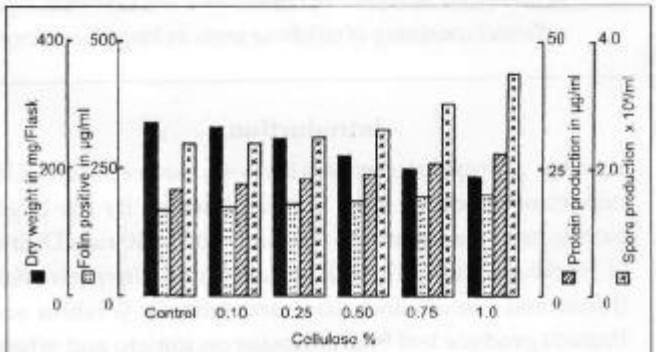

Fig. 4 : *A. triticina* grown on culture media containing extract of prince long cultivar of tomato.

production increased by 4.5 to 25.0% and spore formation increased by 0.0 to 31.0% with the increase of cellulose concentration from 0.1% to 1.0% in the culture media (Fig. 4).

The three strains of *A. solani* from tomato, potato and other plants have been reported to produce a large number of spores under green region of visible spectrum[10]. High growth and sporulation have been reported by *A. solani* in culture with yeast extract and citric acid[11].

Induction of sporulation in *A. porri* and *A. solani* by inhibition of its vegetative development under the interruption of dark, light and moisture has also been reported[12], which is in accordance with present observation. Bhowmic[13] in his detailed study observed that wheat seed infection by *A. triticina* and *A. alternata* form dense mycelial mat between the epidermal surface and cross layer cells. Direct sunlight exposure to three days old culture of *A. solani* for 10 minutes has been found to increase sporulation whereas a marked reduction due to high temperature[14].

The investigation revealed the role of cellulose, consisting cellobiose units in long chain in the inhibition of growth

and stimulation in folin-positive, protein production and sporulation by the virulent (*A. solani*) and avirulent (*A. triticina*) pathogens. The growth was inhibited with resistant (Pusa Ruby) and susceptible (Prince Long) variety, while the folin-positive, protein and spore production was stimulated with different concentration of cellulose. The extent of inhibition as well as stimulation was more in the inoculum of *A. triticina* in comparison to *A. solani*.

Sporulation, protein production and folin positive by virulent and avirulent pathogen in cultures depend not only on carbon source in the medium but also on the host parasite interaction and virulency of the pathogen.

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