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Characterization of Cellulolytic Enzymes of *Fusarium* Soil Isolates**Sayan deb Dutta^a, Mrinmay Tarafder^a, Roushan Islam^b and Bejoysekhar Datta^{a1}**^a Mycology and Plant Pathology Research Laboratory, Department of Botany, University of Kalyani^b Department of Botany, Sripat Singh College, Mushidabad, West Bengal¹ Correspondence: bejoy.datta@gmail.com; Tel.: +91-033-2582-8282

ABSTRACT: Cellulase has tremendous industrial application. *Fusarium* spp. should be exploited as commercial source of cellulase. Three *F. solani*, three *F. oxysporum* and one *F. chlamydosporum* isolates have been recovered from soil, identified through rDNA sequence analysis and their cellulolytic activities have been characterized based on carboxymethylcellulose assay, filter paper assay and cotton assay. All the isolates grew profusely in CMC containing potato dextrose broth and produced sufficient protein including cellulase. Highest CMCase activity (0.445 IU ml⁻¹) by *F. oxysporum* SF0801, FPase activity (9.25 IFPU ml⁻¹) by *F. oxysporum* SF1905 and cotton degradation activity (0.053 U ml⁻¹) by *F. solani* SF1303 have been observed. Optimum temperature and pH of the enzymes were found to be 50°C and acidic condition in most cases. Enzymes from all the isolates showed stability and increased activity when pre-incubated in temperature (40-80°C), pH (4-9), metal salts (CaCl₂, CdCl₂, CuSO₄, FeCl₃, HgCl₂, KCl, MgCl₂, NaCl, NiCl₂, ZnSO₄), inhibitors (β-mercaptoethanol, EDTA, urea, IAA), surfactants (Tween 80, SDS) and oxidizing agent (H₂O₂). Enzyme kinetic analysis revealed low Km value in *F. oxysporum* SF0801 (0.121 mg ml⁻¹) indicating its strongest affinity with the substrate (CMC). Thus the isolates might be utilized as the potential candidates for bioconversion cellulose into fuel and other industrial process.

Keywords: *Fusarium*, cellulase, optimum condition, stability, kinetic analysis**1. INTRODUCTION**

Cellulose is the most abundant biopolymer on earth. Land plants and algae have been reported to synthesize cellulose at the rate of 0.85 X 10¹¹ tonnes per annum (Dar et al., 2013). About 50% of the photosynthetically fixed CO₂ is being accumulated in the form of cellulose (Singh, 2016). It is also produced by some animals e.g., tunicates and a few bacteria. The individual cellulose chain is a linear, unbranched homopolysaccharide consisting of glucose subunit joined together via β, 1-4 glycosidic linkages. These chains vary widely in length and are usually arranged in bundles which aggregate to form microfibrils of 5-15 nm diameters. In the microfibril cellulose chains are arranged in a highly ordered fashion to create the crystalline structures, while less ordered arrangement results the paracrystalline (amorphous) region.

Utilization of cellulose as a nutrient source by the microorganisms requires an enzyme-complex designated as cellulase. Three types of enzymes viz., endo-1,4-β-D-glucanase (EC3.2.1.4), cellobiohydrolase (EC3.2.1.91) and β-D-glucosidase (EC3.2.1.21) act synergistically to depolymerize cellulose. Cellulase is also functioned as a chemical weapon for a number of plant pathogens. Any process that could efficiently and economically convert cellulosic material to glucose would be of immense industrial significance. This glucose could be further used as substrate for subsequent fermentation or other processes which could yield valuable end products such as ethanol, butanol, methane, amino acid, single-cell protein and so on. But most important application of cellulase is for production of fuel ethanol through bioconversion of lignocellulosic biomass. Additional uses include in various

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