Assessment of Cellulolytic Activity from the Microorganisms Isolated From Lower Termite Soil

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***Abstract*- Termites play a pivotal role in the turnover and mineralization of complex biopolymers, such as wood and other cellulose and hemicelluloses containing materials. The current study focuses on the isolation of bacterial isolate from lower termite soil and the assessment of cellulolytic activity. It was found that LT8(lower termite) isolate showed maximum cellulolytic activity.**

***Index Terms*- Termites, Soil, Cellulose, Cellullolytic, Isolate**

INTRODUCTION

Termites are recognized as ecosystem engineer (Dangerfield et al., 1998),Both higher and lower termites have microbes and enzymes in their hindgut , the lower and higher termite soil also possess the aggrandized amount of microflora and this is therefore where the most symbiosis occurs(Sreeremya,2016). Soil on the other hand is a highly heterogenous environment (Rolf, 2004), that encompasses a high diversity of microorganisms (Liesack et al., 2002). Soil microorganisms are a valuable source of natural products providing important antibiotics for pharmaceuticals, enzymes and bioactive compounds for industries (Strohl, 2000).Since soil being a typically good habitat for the growth of many number of microorganisms, majorly observed microorganisms are bacteria: Bacillus sp, Klebsiellasp, Pseudomonas sp, Serratiasp, Xanthomonassp etc. Many fungal species are also obtained from higher termite soil, which include Aspergillussp, Phomasp, Neurosporasp, Trichodermasp, Penicillium sp. The major actinomycetes species observed areStreptomyces sp, Geosminsp, Nocardia sp. Cellulase consists of three different types of enzymes named as endoglucanases, exoglucanses and cellobiases. The study focuses on the isolation and characterization of bacterial species isolated from the lower termite soil.

MATERIAL AND METHODS

SAMPLE COLLECTION

The higher termite soil sample was collected from three different regions of Palakkad district (Nelliyampati, Kallepully, Chittur).The samples were collected,serially diluted and was spread plated.

4.2 ISOLATION OF BACTERIA FROM HIGHER TERMITE SOIL

The soil samples were isolated and was spread plated,from the three soil samples. The six Colonies with visually distinguishable morphologies were selected and restreaked on Nutrient agar to obtain pure cultures isolates were labeled as LT1, LT2, I3, I4, I51, I52.

4.3 IDENTIFICATION OF BACTERIA

Bacteria were identified and classified based on their physical and biochemical characteristics.Various biochemical test such as IMViC, TSI, Carbohydrate fermentation, Catalase, oxidase test, starch hydrolysis test were performed to identify the bacterial species.

SCREENING

The six isolates were inoculated by spot inoculation and incubated at 24-48 hrs.1% of congo red solution was added to the spot inoculated plates and then excess stains were removed by using 1MNaCl and zone of clearance were observed.

RESULT AND DISCUSSION

Lower termite soil samples were collected from three different regions (Nelliyampathy, Kuthanoor, Tholanoor).The three different soil samples were serially diluted, among the three different soil samples, the soil samples collected from Kuthanoor

showed unique colonies at 10^{-4} dilution. (Table:1).The selected colonies were labeled as LT1,LT2,LT3,LT4,LT5,,LT6,LT7.LT8.The specific screening and CMCase assays were carried out.In the previous studies lower termite soil was collected from North India and assessed for cellulose degrading ability(Saravanakumari et al.,2014), the colony which showed cellulose degrading ability whereshoing characteristics of irregular, round colonies(Sreeremya, 2016).

Then the isolates were subjected to physical and biochemical characterization, the results were tabulated (Table:1 to 4). After the preliminary screening the five isolates;LT2,LT4,LT5,LT7, LT8 isolate was found to be more efficient for the CMCase activity (Fig:1). Among which LT8 showed maximum cellulolyicactivity.

ENUMERATION OF BACTERIAL COLONIES

Table:1

SERIAL DILUTION	COLONIES OBTAINED (ONE QUDRANT)	COLONIES OBTAINED(FOUR QUADRANT)
10^{-2}	101	404
10^{-3}	96	384
10^{-4}	77	308
10^{-5}	66	264
10^{-6}	52	208
10^{-7}	15	60

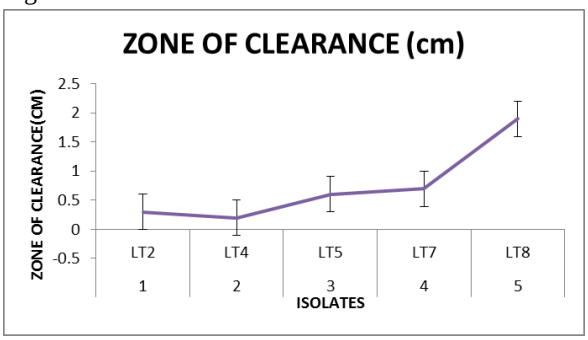
MORPHOLOGICAL EXAMINATION

Table:2

Sl no:	Name	Size	Shape	Color	Margin	Surface	Elevation	Transparency	Viscosity
1	LT1	Large	Irregular	Yellow	Entire	Glossy	Flat	Opaque	Moist
2	LT2	Small	Circular	White	Entire	Finely granular	Flat	Transparent	Dry
3	LT3	Large	Circular	Creamy	Entire	Glossy	Flat in growing	Opaque	Moist
4	LT4	Small	Circular	Creamy	Entire	Glossy	Flat	Opaque	Moist
5	LT5	Small	Circular	White	Entire	Glossy	Flat	Opaque	Mucoidal
6	LT6	Large	Irregular	White	Diffuse	Smooth	Flat in growing	Opaque	Moist
7	LT7	Small	Irregular	Creamy	Diffuse	Sticky	Flat	Opaque	Moist
8	LT8	Medium	Irregular	Yellow	Diffuse	Sticky	Flat	Opaque	Moist

CELLULOSE SCREENING

Fig:1



MICROSCOPIC EXAMINATION & BIOCHEMICAL CHARACTERIZATION

Table:3

SL NO:	Bacterial isolates	Microscopic features	Indole test	Methyl red	VP	Citrate utilization	Catalase	Urease	Starch hydrolysis
1	LT2	Gram negative, rod	-ve	-ve	-ve	+ve	+ve	-ve	-ve
2	LT4	Gram positive, rod	+ve	-ve	+ve	+ve	+ve	-ve	-ve
3	LT5	Gram negative, rod	-ve	-ve	-ve	-ve	+ve	-ve	-ve
4	LT7	Gram positive cocci	-ve	-ve	-ve	-ve	+ve	-ve	-ve
5	LT8	Gram positive, rod	-ve	-ve	+ve	+ve	-ve	-ve	-ve

BIOCHEMICAL CHARACTERIZATION

Table:4

SL NO:	Bacterial isolates	TSI	GLUCOSE	SUCROSE	LACTOSE
1	LT2	AS & AB	A & NG	A & G	G & NA
2	LT4	AS & AB	A & NG	A & G	G & NA
3	LT5	AS & NO AB	-VE	G & NA	G & NA
4	LT7	AS& NO AB	-VE	G & NA	NA & NG
5	LT8	AS & AB	A & NG	A &G	NA &NG

SL NO:	Bacterial isolates	TSI	GLUCOSE	SUCROSE	LACTOSE
1	LT2	AS & AB	A & NG	A & G	G & NA
2	LT4	AS & AB	A & NG	A & G	G & NA
3	LT5	AS & NO AB	-VE	G & NA	G & NA
4	LT7	AS& NO AB	-VE	G & NA	NA & NG
5	LT8	AS & AB	A & NG	A &G	NA &NG

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