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Sustainable Cellulases Production Using Solid Waste Feedstocks

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Abstract: Background*:* Cellulose is mainly dominating Agricultural waste and available naturally on earth as biopolymer. Being a renewable and abundant resource, it can be degraded by cellulase producing microbes. Generally, the raw material is considered as key factor in enzymatic production. Solid waste materials that are often reliable and abundant can be the best substitutes *per se*. Cellulase enzyme has been reported in numerous biotechnological applications including chemicals, single cell protein, food and agriculture. Thus, increasing potential of cellulase applications prompts screening for isolation of newer cellulase producing microorganisms, which can meet the industrial demand. Keeping in view the potential applications of cellulases, the present min-review was undertaken to summarize the role of microbes in sustainable cellulase productions using diverse solid waste feedstock to make the process economically viable. **Methods:** Microbial information regarding underutilized plants was acquired from a literature exploration of diverse databases for instance Scopus, Google Scholar and Pubmed. **Results:** Earlier studies revealed that traditionally only wheat and rice based sources were used. However, with the advent of time many other solid wastes like Coconut coir pith, Sweet sorghum silage, Sugar Cane, Round nut shell, Palm Kernel Cake, Rice bran, Filter Paper Cornmeal, Soybean hull, Soybean hull, Corn cob residue and wheat, bran ,Corn silage digestate, Soybean hulls and wheat bran, Soybean hulls and wheat bran, Paper, wood, litmus paper Freshly ripe tomato fruit, Olive processing residue, Apple pomace, Sawdust, Bagasse. **Conclusion:** The biotechnological facet of cellulase research using solid waste feedstock and their future prospects are also elaborated.

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Keywords: cellulose, cellulase, solid waste, feedstock, microbes

1. Introduction

Typically, cellulose which constitutes more than 50% of plant biomass, is the key predictable sustainable resource of fuels for human mankind. Therefore, agricultural based waste products like corn straw, wheat straw, rice straw etc are alternative cheap, renewable chiefly unexploited sources of biomass (Damato et al., 2010). Besides, industrial and agricultural processes produce multiple sources of lignocellulosic waste such as coconut biomass, citrus peel waste, paper pulp, sawdust, industrial waste, paper mill sludge and municipal cellulosic solid waste (Krishna, 1999). Cellulose structure is composed anhydro-D-glucose units, unbranched, linked by 1, 4-β-D-glucoside bonds, which can be hydrolyzed by microbial cellolytic enzymes (Acharya et al., 2008). Traditionally, hydrolysis of cellulose could be performed acids and enzymatic hydrolysis. But as reported by Mandels et al., (1974), enzyme based hydrolysis is typically favored as it yields smaller amount by-products and profits under milder condition. Also this conventional method is highly uneconomical because of low conversion efficiency, yellowing and bleaching of fibers during manufacturing and finishing. Therefore, microbial cellulases which cleave cellulose by a single step reaction, offer an attractive alternative. Cellulase, generally inducible and usually excreted in the medium, is designated as group of enzymes that are involved in cellulose degradation to glucose (Fig. 1) (Kawamori *et al*., 1985). As per Jing D et al., (2007) cellulose hydrolysis (Fig 1) can be performed by 3 components of cellulases indicated in Table 1. In the last decades cellulase enzyme has been reported in numerous environmental friendly technologies and biotechnological applications like single cell protein, pulp industries, fiber manufacturing and finishing (Howrad *et al*., 2003). Oinonen and Suominen (2002) cited that well-known application is the use of cellulases in biostoning and deinking of waste paper. Some other applications include: cotton processing, paper recycling, juice extraction, green-tea components extraction, malting, alcohol fermentation, ethanol production, are few practical citations promoting the usefulness of the enzyme. Keeping in mind the potential applications of cellulases, the present chapter summarizes the cellulase production

ability of microbial strains using cost effective solid agro wastes.

Table 1: Microbial cellulose system

Enzyme	E. C. number	Reaction	Other Names
i)Endo -1,4 β-D-glucan-	E. C. 3. 2. 1. 4	Cut at random at internal amorphous	Endoglucanse, Endo-1,4-
glucanohydrolase		sites of cellulose generating	β –glucanse,
		oligosaccharides of various lengths. It	Carboxymethyl cellulase,
		acts on Endo-1, 4-beta-D-glucosidic	β -1,4-endoglucon
		linkages in cellulose, lichenin and cereal	hydrolase, Endocellulase
		beta-D-glucans.	
ii)Exoglucanase or 1,4-β-D-	E.C.3.2.1.91	Hydrolysis of 1,4-beta-D-	Exoglucanase,
glucan cellobiohydrolases		glucosidiclinkages in cellulose and	Exocellobiohydrolase, 1,
(cellobiohydrolases)		cellotetraose, releasing cellobiose from	4- β-cellobiohydrolase.
		the non-reducing ends of the chains	
iii) Exoglucanases or 1,4-β-	EC 3.2.1.74	Removal of cellobiose from	Cellodextrinases
D-oligoglucan		cellooligosaccharide or from p-nitrphenyl-	
cellobiohydrolases		β-D-cellobioside	
iv) β - Glucosidases or β-D-	E.C.3.2.1.21	Hydrolysis of terminal non-reducing beta-	Gentobiase, Cellobiase,
glucoside gluco-hydrolases	.	D-glucose residues with release of beta-	Amygdalase.
		D-glucose.	
v) Cellobiose:	E.C. 2.4.1.49	It catalyzes the reversible phosphorolytic	Cellobiose
orthophosphate alfa–D-		cleavage of cellobiose	phosphorylase
glucosyl transferase			
vi) 1,4-β-D-	E.C. 2.4.1.20	It catalyzes the reversible phosphorolytic	Cellodextrin
oligoglucan:orthophosphate		cleavage of cellodextrins ranging from	phosphorylase
alfa –D-glucosyl transferase		cellotriose to cellohexoses.	
vii) Cellobiose 2- epimerase	EC 5.1.3.11	It catalyzes the cellobiose into 4-O- β-D-	Cellobiose 2- epimerase
		glucosylnannose.	
viii) Complete Cellulase	-	Catalyzes extensive hydrolysis of	Total cellulase
system		crystalline cellulose	

Table 2: Some traditionally used microbial substrates

Strain name	Substrate	Max. Enzymatic Activity (IU)	Reference
T.koningii	Saw dust, wheat bran	na	Mandels et al., 1957
T.reesei	Wheat bran	1.0	Saddler *et al*., 1982
Sporotrichum pulverulentum , T.reesei	Wheat bran	na	Mackenzie *et al*., 1985
T.harzianum	Wheat straw, Wheat bran	198	Deschamps, F., *et al*., 1985
Trichoderma sp. Botritis sp.	Wheat bran, rice straw	na	Brown, *et al*., 1987
Neurospora crassa	Wheat straw	na	Macris *et al*.,1984
A.niger	Wheat bran	2100	Oguntinein, *et al*., 1992
A.niger	Coconut coir pith	na	Muniswaran, P., *et al*., 1994
Aspergillus sp.	Bagasse, Wheat bran, rice bran	na	Gupte, A., *et al*., 1994
Lentinus edodus	Wheat straw	na	Nigam, and singh 1994
T.reesei	Paddy straw	107	Han, *et al*., 1995
Gliocladium sp. Trichoderma sp. Penicillium sp.	Sweet sorghum silage	na	Smits *et al*., 1996
T.harzianum	Cassava waste	na	Onilude, A.A.,*et al*., 1996
A.fumigatus	Wheat straw	0.225	Umar Dahot, M., *et al*., 1996
Cerrena unicolor	Grape wine cutting waste	na	Elisashvili, V.I., *et al*., 1998
Bacillus subtilis	Banana wastes	9.6	Krishna 1999

2. Material and Methods

Microbial information regarding underutilized plants was acquired from a literature exploration of diverse databases for instance Scopus, Google Scholar and Pubmed up to 2020 from research publications. The foremost keywords used were “cellulases”, “solid waste feedstocks”, “agrowastes”.

1. Microbial Sources of Cellulases

Environmental pollution is majorly governed by agricultural and industrial based solid wastes. Majority of solid agro wastes which are also underutilized include wheat and rice husk, black gram husk, rice bran. Although these wastes worldwide mostly used as animal feeds. However, many of them left in fields which are decayed by cellulase producing microorganisms. The most common producer of cellulase is fungi, in addition to bacteria or actinomycetes. Traditional studies revealed that most commonly used substrate for cellulase productions was either wheat or rice based derivatives (Table 2). For instance: Mandels and Reese (1957) cited cellulase production by Trichoderma viride on wheat bran and metals. Saddler et al (1982) also utilized wheat bran as carbon source to produce cellulase. Although mostly used carbon sources were based on wheat and rice, however, some authors have documented role of some other carbon sources too like (Table 2). Aspergillus and Trichoderma are mostly used genera for commercial agricultural use as shown in table 1 and 2.

3.1 Bacteria

Zhuang et al.,(2007) studied for production of cellulase by using solid waste biomass. Table 3 illustrates studies based on consumption of lignocellulosic biomass for cellulase creation by bacterial. Odeniyi et al., (2009) also studied that agro wastes like pineapple are organic and is easily despoiled by microorganisms. Lah et al., (2012) utilized palm kernel cake as carbon solid waste courcee for *Bacillus pumilus EB3* for production of cellulase and reported substancial yield. Acharya and, Chaudhary, (2012) used filter sheet as carbon source for *Bacillus licheniformis* and documented cellulase activity. Annamalai et al. (2014) cited cellulase enzyme activities on rice bran in Bacillus carboniphilus CAS 3.

3.2 Fungus

Gordillo-Fuenzalida et al., (2019) reported cellulase by using food manufacturing wastes (FMWs), like solid from mills, tomato and grape waste, using Trichoderma reesei. Darabzadeh et al., (2019) used rice bran, rice husk, and rice straw for Trichoderma sp. It was observed that max activity was 1.37 U/ml. Singhania et al., (2006) using wheat bran, and Brijwani et al., (2011) used soybean hull reported significant cellulase activities. Using rice straw Dhillon et al. (2011) evaluated cellulase by Penicillium atrovenetum, Aspergillus flavus and Aspergillus oryzae (Omajasola *et al*., 2008). Wong Kok Mun *et al*., (2008) reported cellulase using palm oil effluent solid by Thermoascus aurantiacus and Aspergillus niger. For pineapple waste Omojasola et al., (2008) used Trichoderma longibrachiatum, Aspergillus niger and Saccharomyces cerevisiae. Muhannad *et al*., 2001 using *Trichoderma reesei* QM9414 and *Aspergillus terreus SUK-1* exploited sugarcane bagasse for cellulase production. Against the bio wastes, coir waste and saw dust Immanuel *et al*., 2007 documented the cellulase enzyme production by *Aspergillus niger* and *Aspergillus fumigates.* Jing *et al*., 2007 used mix culture of *Trichoderma koningii, Aspergillus niger, Lactobacillus* for cellulase production on waste pea shrub woody biomass . Ajayi *et al*., 2007 described cellulytic ability of *Aspergillus flavus Linn* from tomato fruits. Jhadhav et al. (2013) studied millet husk, banana peels, wheat bran Rice husk, coir waste for the productivity of cellulase enzyme using Trichoderma reesei and Apergillus phoenicis QM329. Similarly Waghmare et al. (2014) cited cellulase production by using aricultural wastes like paddy straw, corn straw, sugarcane bagasse/ barbojo, grass powder, sorghum husks, and for cellulolytic enzyme production by Klebsiella sp. PRW-1 (Table 1).

Table 3. Compilation of studies of cellulase production

Strain name	Substrate	Max. Enzymatic Activity (IU)	Reference
Bacteria			
Bacillus pumilus EB3	Pine apple waste	0.96	Odeniyi et al., 2009
Bacillus subtilisKO	wheat straw, and rice husk	0.08	Shabab et al., 2010
Bacillus sp	Sugar Cane	15	Patel et al., 2005
Bacillus sp	Round nut shell	0.4	Dey et al., 2002
Bacillus Cereus	Palm Kernel Cake	2.73	Lah et al., 2012
Bacillus carboniphilus CAS 3	Rice bran	4040	Annamalai et al., 2014
Bacillus licheniformis MVS1	Filter Paper	0.47	Acharya and, Chaudhary, 2012
Cellulomonas cellulans MTCC 23	Paddy straw	0.35	Mishra et al., 2007
Clostridium thermocellum	Cellulose and paper pulp	NA	Zhuang et al., 2007
Cytophaga hutchinsonii NCIM 2338	Paddy straw	0.33	Mishra et al., 2007
Fungus			
Trichoderma reesei RUT 30	Wheat bran	22.8	Sukumaran et al., 2009
Aspergillus terreus M11	Wheat straw		Gao et al. 2008
Aspergillus fumigatus Z5	Cornmeal	98.1	Liu et al., 2011
Trichoderma reesei	Soybean hull	na	Brijwani et al., 2011
Aspergillus oryzae	Soybean hull	5.4	Brijwani et al., 2011
Trichoderma reesei	Rice straw and wheat bran	5.4	Dhillon et al. 2011
Aspergillus oryzae	Rice straw and wheat bran	35.8	Dhillon et al., 2011
Trichoderma reesei ZU-02	Corn cob residue and wheat bran + mineral solution	40–158	Xia and Cen, 1999
Pleurotus ostreatus	Corn silage digestate	0.7	Santi et al., 2015
Trichoderma reesei (ATCC 26921)	Soybean hulls and wheat bran	10.78	Flodman and Noureddini, 2013
Aspergillus oryzae (ATCC 12892)	Soybean hulls and wheat bran	10.78	Flodman and Noureddini, 2013
Fusarium oxsporum	Paper, wood, litmus paper	2.0	Omojasola et al., 2008
Aspergillus flavus Linn	Freshly ripe tomato fruit	1.4	Damato et al., 2010
Trichoderma reesei	Olive processing residue	2.8	Sun et al., 2010
Trichoderma species	Apple pomace	13.8	Eldein et al., 2010
Aspergillus flavus	Sawdust, Bagasse		Singh et al., 2016
Aspergillus niger.	rice bran, wheat bran, black gram bran, coconut oil cake, groundnut oil cake, and gingely oil cake	5-30	Mahalakshmi N and S. Jayalakshmi, 2016
Aspergillus niger, Aspergillus terreus	Cassava waste	33.7	Siham et al., 2007
Aspergillus niger and *Trichoderma viride*	Waste paper	33.7	Pothiraj et al., 2006
A.niger	Radicle waste (malt manufacture residue)	74.4	Fadel, M.,2000
T.reesei QM9414, A.terreus SUK	Sugarcane bagasse	*na*	Muhannad *et al*., 2001
A.flaves Nigrospora sp.	Orange waste	na	Shahera *et al*. 2002
Phytophthora cinnamomi	mulch	na	Ben Faber *et al*. 230
A.flavus Linn NSPR 101	Saw dust bagasse and corn cob	0.07	Ojumu *et al*. 2003
A.niger KK2	Rice straw Wheat bran	na	Holker et al., 2004
T.harziamum	Wheat straw Wheat bran	na	Deschamps, F., *et al*., 2004
T. harzianum biomass	Oil palm biomass	10.1	Md.Zahangir Alam *et al*., 2005
A.niger MSK-7,T.viride MSK-10	Wheat bran	na	Ikram ul Haq *et al*., 2005
A.niger	Millet, guinea corn straw, rice husk and maize straw	102	Miala, M.A., *et al*.,2005

A.candidus	Rice husk, millet straw, guinea corn stalk and saw dust	7.5	Milala, M.A., *et al*., 2009
Thermoascus aureantiacus miche	Wheat bran, sugarcane bagasse, orange bagasse, corn cob, green grass, dried grass, saw dust and corn straw	3-60	Silva *et al*., 2005
A.niger, T.viride	Rice bran, wheat bran, cotton	na	Ikram ul Haq *et al*., 2005
T.reesei	Sugarcane bagasse, rice straw	44-68	Muthuvelayudham,R., *et al*., 2006
T.harzianum	Wheat bran,wheat straw, rice bran, rice husk and soybean	na	Ikram ul Haq *et al*., 2006
T.reesei LW1	Corn straw	996	Wang.J.Sh *et al*., 2006
Scopulariopsis	Rice bran	8.3	Bharathi Kodali *et al*., 2006
Penicillium echinalatum	Bagasse, wheat bran	na	Camassola, M., *et al*., 2007
T.koningiiAS3 4262	Wheat bran, vinegar waste	23.76	Jian Liu *et al*., 2007
T.koningii. A.niger, Lactobacillus	Pea shrub biomass	na	Debing Jing *et al*., 2007
A.niger Eb5, T.sp.EB6	Palm oil mill effluent	14.76	Wong Kok Mun *et al*., 2008
T.longibrachiatum, A.niger, Saccharomyces cerevisae	Orange waste	na	Omajasola, P.F., *et al*., 2008
T.longibrachiatum, A.niger, Saccharomyces cerevisae	Pine apple waste	na	Omajasola, P.F., *et al*., 2008
A.niger	Saw dust	na	Acharya, P.B., *et al*., 2008
A.niger	Green gram husk	na	Sharada.R et al., 2012
A.niger	Corn cob	0.027	Jahir Alam Khan et al., 2011
A.niger	Vigna mungo	na	Umbrin Ilyas et al., 2011,
T.reesei SEMCC	Water hyacinth	na	Shi hao Zhao et al., 2011
Rhizopus oryzae ME01	Palm kerne cake	na	Mohd.Firdaus Othman et al., 2013
Trichoderma reesei and Aspergillus niger.	municipal solid waste	26	Abdullah et al., 2016
T. reesei	water hyacinth	13.4	Zhao et al., 2013
	sugar cane bagasse	154	Singhania et al., 2006
	oil palm empty fruit bunches	8.2	Alam et al., 2009
	oil palm empty fruit bunches	1.1	Latifian et al., 2007
	Food manufacturing wastes (FMWs), such as olive mill solids, tomato pomace, and grape pomace,	10	Gordillo-Fuenzalida et al., 2019
	rice by-products	1.4	Darabzadeh et al., 2019
Aspergillus heteromorphus	distillery spent wash (ADSW) and rice straw	8	Bajar et al., 2020

Case specific study:

Lignocellulosics agricultural wastes key source in cellulase production by different microbial strains in submerged and solid state cultivations

Earlier studies showed that under submerged or solid-state fermentations, the main factor for the production of cellolytic enzymes is the choice of an appropriate inducing substrate [Kulkarni et al., 1999]. Therefore, keeping in mind the reduction in the production cost, use of inexpensive waste materials, such as rice straw, wheat straw, waste paper, sugarcane bagasse etc. are often reliable and abundant. In the present study, the effect of various lignocellulosics on extracellular cellulase enzyme production was studied for comparative purposes. The present study reports indigenously isolated Staphylococcus and Bacillus strains and two fungal Aspergillus strains (purchased from microbial bank) for their cellulase production ability on different lignocellulosics agricultural wastes under different fermentations conditions.

Cellulase Production on Different Substrates under Different Fermentation Conditions By Staphylococcus Strains

For determining the time course of cellulase production, *Staphylococcus* strains were cultivated in different fermentation conditions using 1% agricultural wastes as the carbon sources at 37°C. The data showed that there was a significant difference in the enzyme production when different carbon sources were used. Among different carbon sources, cellulase production was higher when strain ScLKC1 grew on wheat straw after four days of cultivation under stationary conditions, following by an eventual decrease (Figure 2A). However, with waste paper as a carbon source, two peaks of activity maxima (after 2 and 4 days of cultivation) were observed. As compared with other carbon sources used, ScLKC2 gave highest activity on wheat straw and eucalyptus bark after 4 days of cultivation under stationary conditions (Figure 2B). The depression in enzyme activity was observed between 2 and 4 days with saw dust and sugarcane molasses. Using ScLKC3, cellulase activity increased to the maximum at 4 days of stationary cultivation for all lignocellulosics materials used (Figure 2C). Among different carbon sources, maximum production was observed with saw sawdust and sugarcane molasses.

Under shaking conditions, ScLKC1 grown on sugarcane molasses gave the highest cellulase activity (0.610 IU/ml) after 3 days of cultivation, however, with eucalyptus bark the enzyme production was maximum after 2 days of cultivation and remained stable up to 3 days before the eventual decrease in the activity. Moreover, when saw dust was used as the carbon source, the maximum cellulase activity was observed after 1 day of cultivation. Further, as observed in stationary fermentation, the depression in enzyme activity was observed between 1 to 3 days, common to five substrates except eucalyptus bark where no depression in enzyme activity was observed. Similar results of enzyme activity have been reported by Ozumu *et al* (2003) in *Aspergillus flavus*. ScLKC 2 under shaking conditions, maximum activity was observed after 3 d of fermentation followed by a stable decrease. A depression in enzyme activity was observed at 2nd day of fermentation, common to five substrates except sugarcane molasses. In ScLKC 3, under shaking conditions, wheat straw supported maximum enzyme production after 3 days of fermentation, however, with all other carbon sources the enzyme production was maximum after 4 days of cultivation. A strong depression in enzyme activity was observed between 1 to 3 days with waste paper and eucalyptus bark.

Under stationary conditions, cellulase activity increased to maximum at 4 day of production with all the lignocellulosics materials used, however, with eucalyptus bark, another maxima of enzyme activity was observed after 2 days of cultivation (Figure 3). Under shaking conditions, the enzyme activity continued to increase up to 3 days of incubation for all the substrates used followed by a substantial decrease. Maximum activity was observed with sugarcane molasses. Since ScLKC4 strain gave higher activity (~0.6 IU/ml) using eucalyptus bark just after 2 day of cultivation under stationary conditions, hence, in addition of stationary and shaking fermentations, the strain ScLKC4 was also tested on solid-state fermentation condition. The strain depicted maximum activity with wheat straw, saw dust, eucalyptus bark and waste paper after 4 days of fermentation, however, with rice straw and sugarcane molasses the activity was maximum at 3rd day of cultivation.

Cellulase Production on Different Substrates Under Different Fermentation Conditions By Bacillus Strains

For determining the time course of cellulase production, two B*acillus* strains designated as: BaLKC1, and BaLKC2 were cultivated in different fermentation conditions using 1% agricultural wastes as the carbon sources at 37°C. Among all the substrates tested, BaLKC1 grown on waste paper gave the highest activity of 0.581 IU/ml after one day of stationary fermentation (Figure 4). However, with wheat and rice straw, the productivity was maximum after 3 and 4 days of cultivation, respectively. Under shaking conditions the maximum activity (0.811 IU/ml) was observed with sugarcane molasses after 3 days of incubation. Rest with all the substrates

maximum activity was observed after 4 days of fermentation.

The highest enzyme activity was observed with eucalyptus bark after 4 days of stationary conditions, however, with wheat straw the maximum enzyme activity was observed after 2 days of incubation (Figure 5). Under shaking conditions, the production rate was higher with wheat straw after one day, however, saw dust and sugarcane molasses gave maximum activity after 3 days. Moreover, a depression in enzyme activity was observed between 1 to 3 days, common to wheat straw, eucalyptus bark and rice straw which is in agreement with the earlier studies [Kotchoni et al., 2003) . Notably this strain depicted higher enzyme activity just after two days of cultivation using wheat straw in stationary and saw dust and molasses in shaking conditions, respectively. Therefore, further, when this strain was tested in solid-state fermentation, maximum enzyme production was supported by waste paper as a carbon source after 4 days of incubation. However, with eucalyptus bark, maximum activity was observed after 3 days of fermentation.

Cellulase Production 0n Different Substrates Under Different Fermentation Conditions By Aspergillus niger

During stationary conditions, *A. niger* grown on, saw dust, waste paper, sugarcane molasses and eucalyptus bark gave maximum enzyme production after one day followed by a sluggish decrease (Figure 6). However, maximum enzyme activity was observed with paper as a carbon source. The wheat straw and rice straw supported maximum enzyme production at 3 and 4 day of fermentation, respectively. Under shaking conditions, with all the substrates the enzyme activity maxima climaxed at one day followed by a dramatic decrease. The enzyme production was relatively same with all the different substrates tested. Under solid-state cultivation, the enzyme activity was maximum using wheat straw after 2 days of cultivation, however, using rice straw and eucalyptus bark the enzyme activity peaked at 2 and 3 day, respectively.

Cellulase Production On Different Substrates Under Different Fermentation Conditions By Aspergillus oryzae

Among different carbon sources utilized, the maximum enzyme activity was observed with waste paper after one day of fermentation (Figure 7). However, with wheat straw and rice straw the activity maxima peaked at 3 and 4 day, respectively, followed by a sharp decrease. On the contrary, under shaking conditions the strain depicted maximum activity after 1 day of cultivation and decreased thereafter. Under solid-state cultivation the highest activity was observed with rice straw after 3 days, however, with waste paper, sugarcane molasses and saw dust, the maximum activity was observed 2 days of fermentation.

To conclude, a look at the present and previous studies clearly reveals that irrespective of the carbon source and cultivation conditions used, the strains reported in this study produced higher activity than the earlier reported strains e.g. *Bacillus* sp. (0.25 IU/ml) (Kotchoni et al., 2003); *Caldibacillus cellulovorans* (0.005 IU/ml) (Huanag and Monk, 2004) and *Aspergillus* sps (0.056-0.076 IU/ml)(Ojumu et al., 2003). Therefore, microbial species isolated in the present study could be another promising organisms for various industrial applications. The possibility of producing cellulase from agricultural wastes has also been demonstrated in the study and it may be an effort in improving the economy of enzyme production in biotechnological industries. A further study on characterization, purification and immobilization of this enzyme is underway for exploiting its commercial potential.

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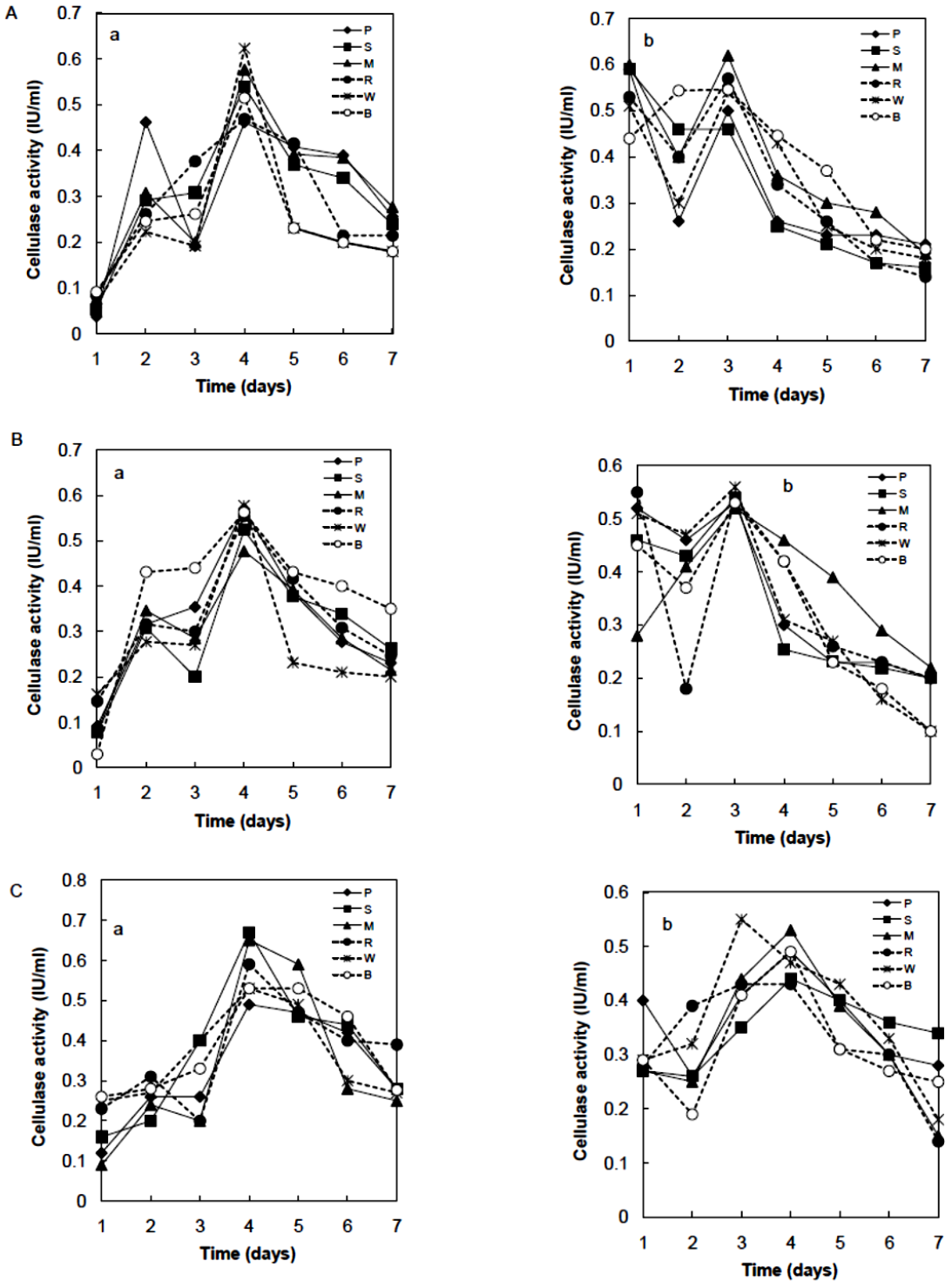

Figure 2: Cellulase production by different *Staphylococcus* strains viz; ScLKC1 (A), ScLKC2 (B), ScLKC3 (C), under stationary (a) and shaking (b) conditions. Symbols used: P, waste paper; S, saw dust; M, sugarcane molasses; R, rice straw; W, wheat straw; B, eucalyptus bark.

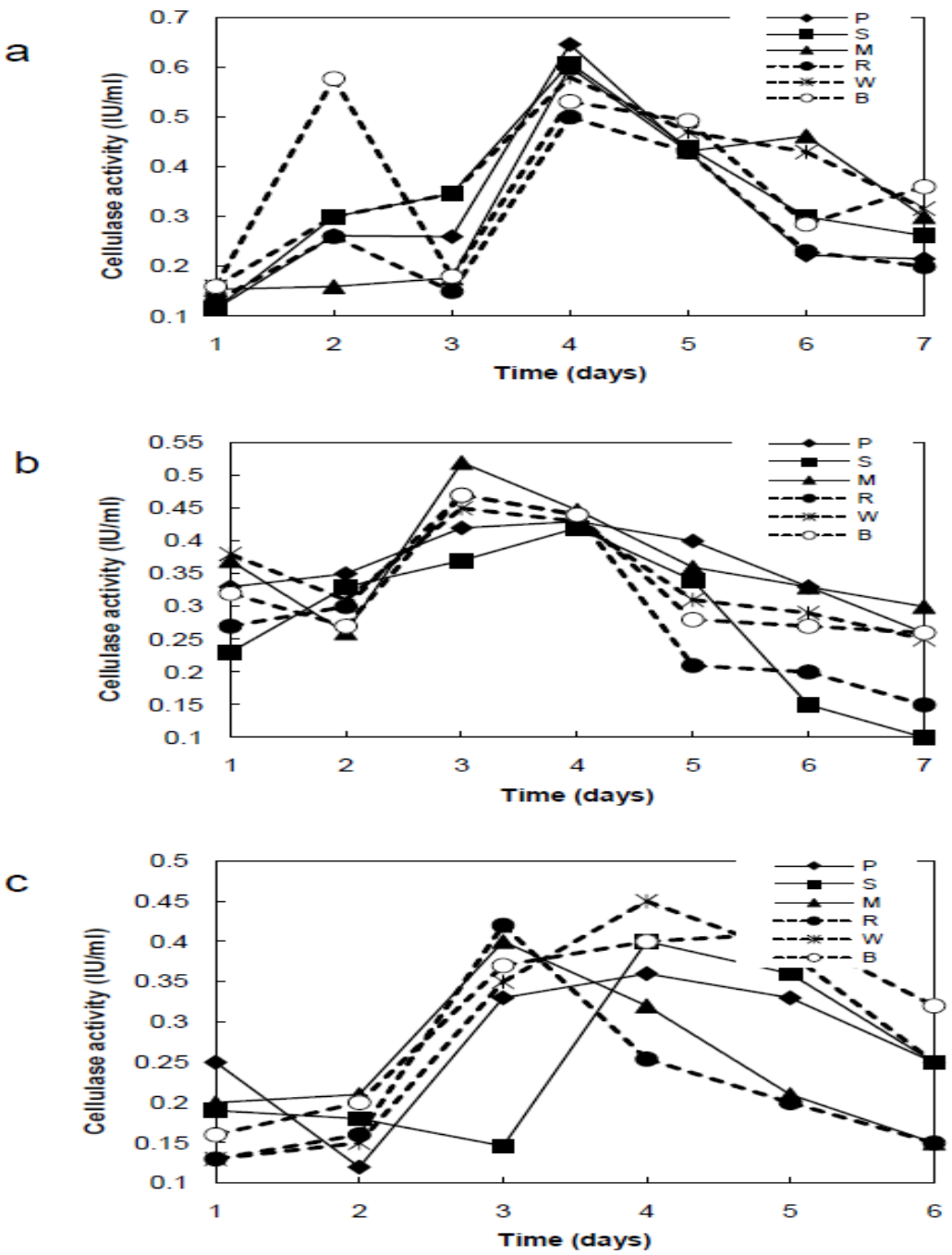

Figure 3: Cellulase production by Staphylococcus strains viz; ScLKC4 under stationary (a), shaking (b) and solid state (c) conditions.

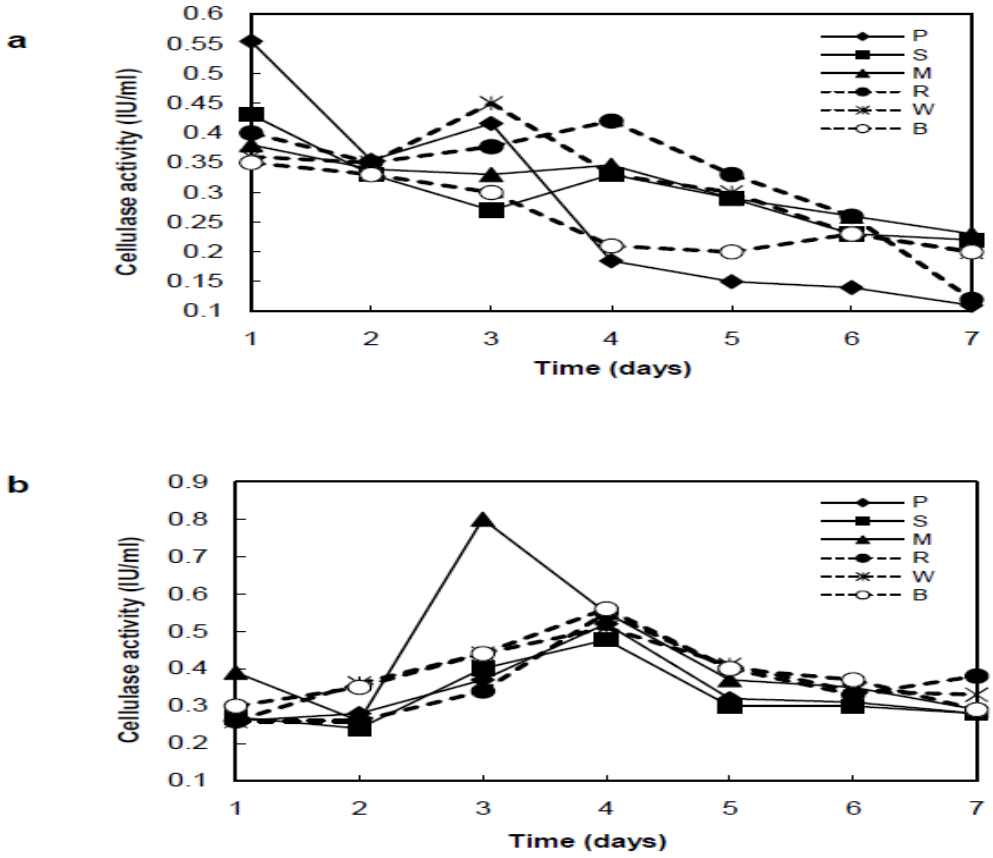

Figure 4: Cellulase production by B*acillus* sp. LKC1 under stationary (a) and shaking (b) conditions..

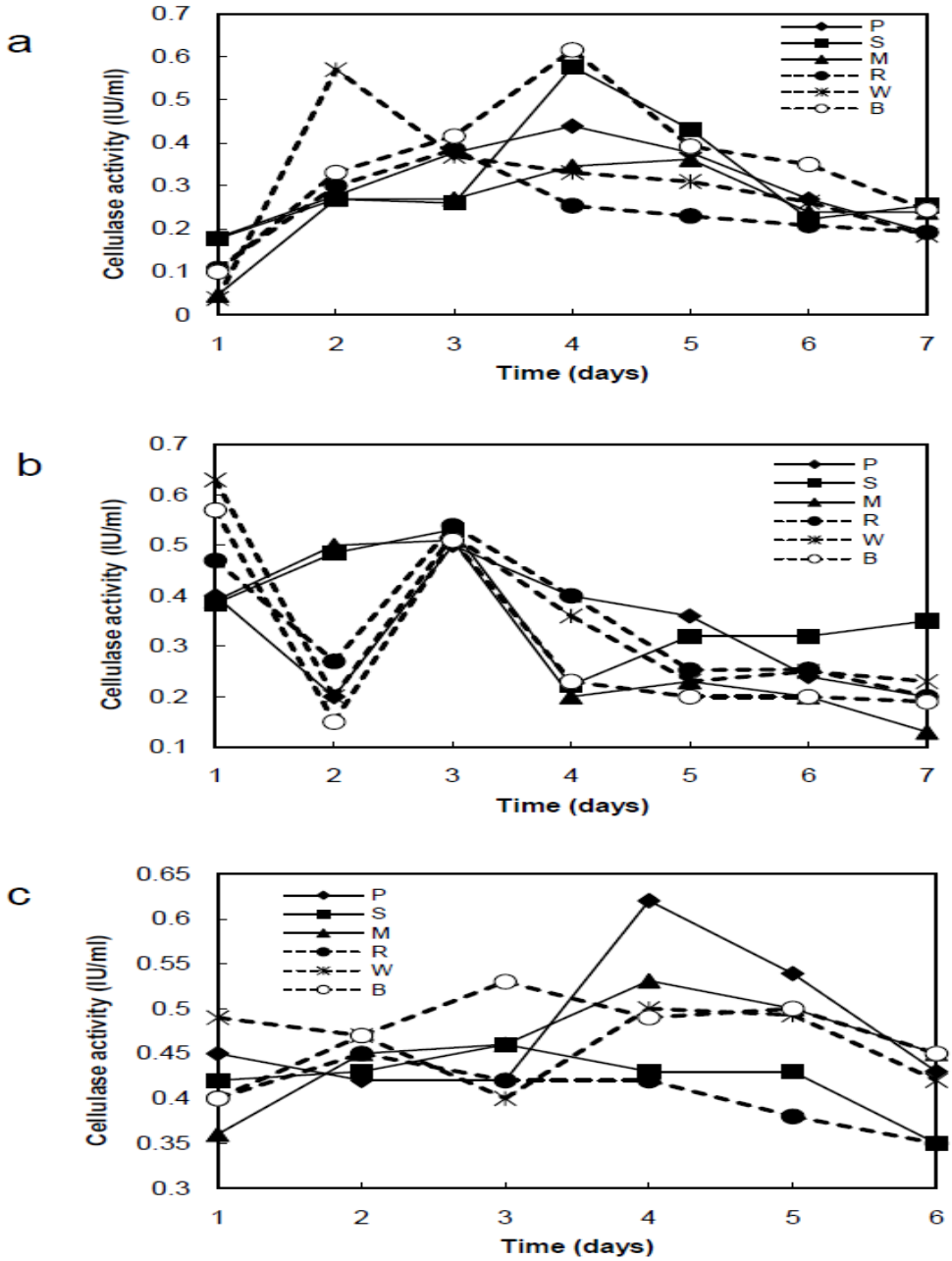

Figure 5: Cellulase production by B*acillus* sp. LKC2 under stationary (a), shaking (b) and solid state (c) conditions.

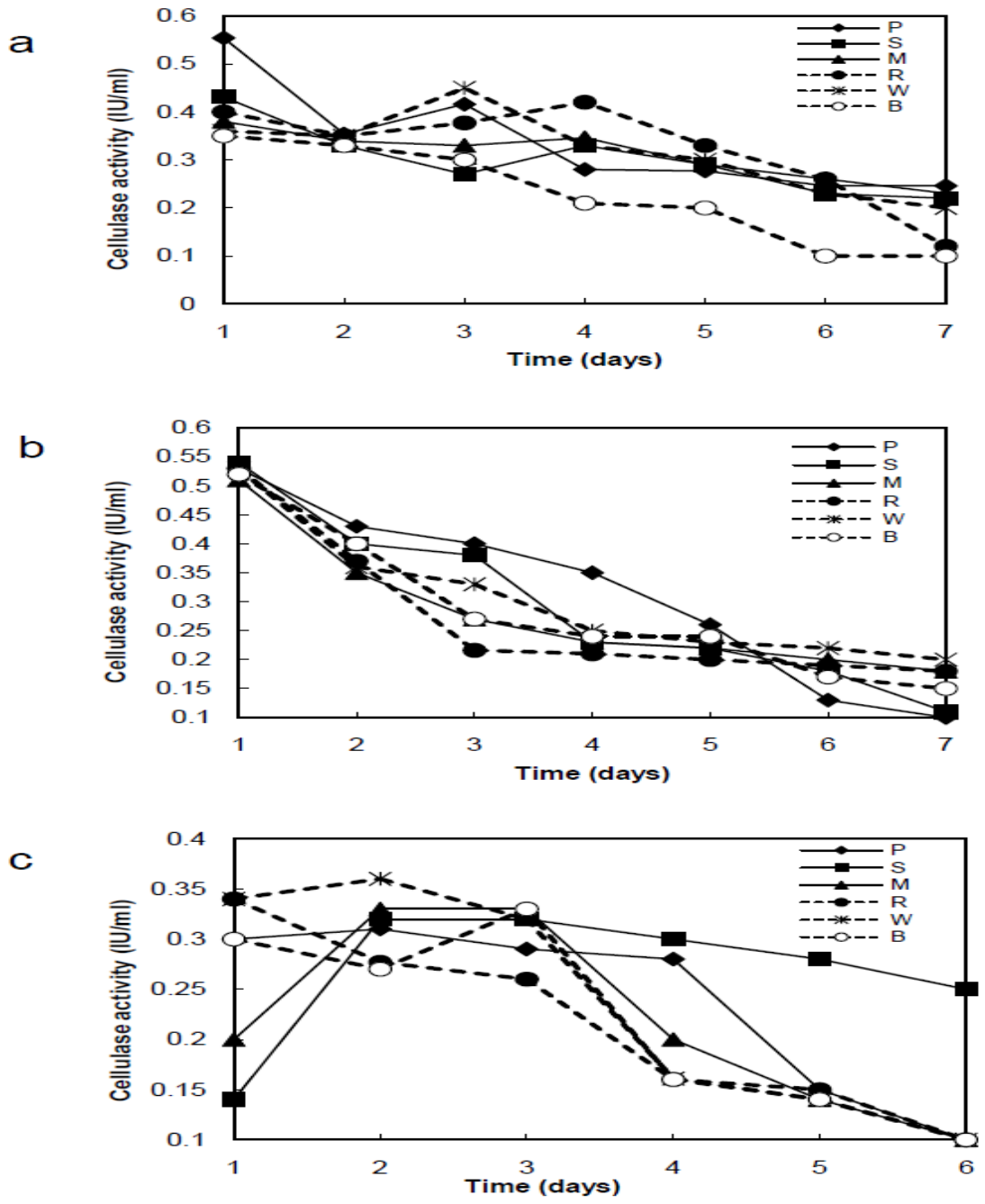

Figure 6: Cellulase production by *Aspergillus niger* under stationary (a), shaking (b) and solid state (c) conditions

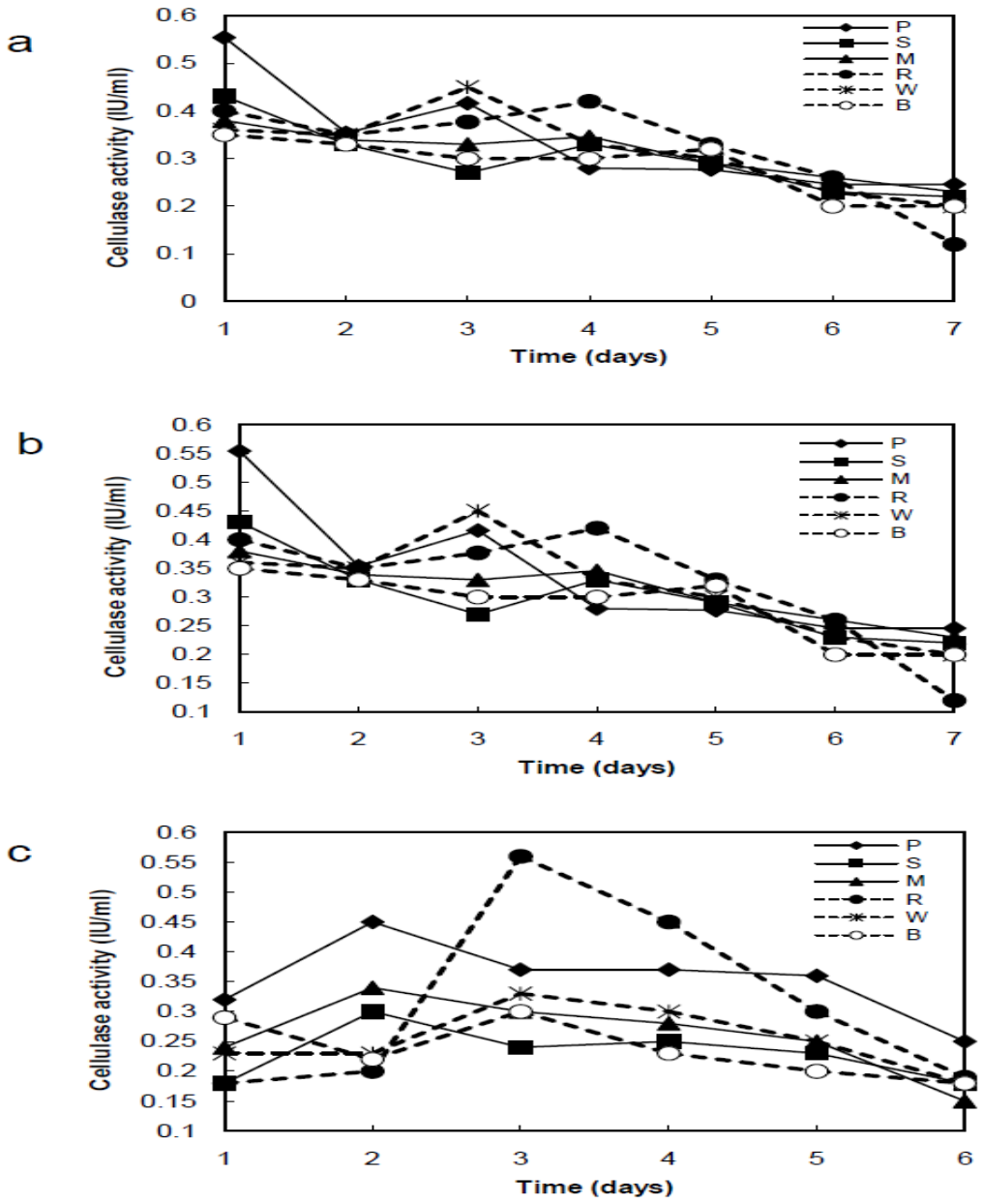

Figure 7: Cellulase production by *Aspergillus oryzae* under stationary (a), shaking (b) and solid state (c) conditions

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