

Supplemental Information

Defined enzyme cocktail from the anerobic fungus *Orpinomyces* sp. strain C1A effectively releases sugars from pretreated corn stover and switchgrass

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Supplementary File 1. Protein sequence of all enzymes cloned and expressed in this study.

>EG5

TLLGLAIIGTKAMRNISSELVKELTIGWSLGNTLDATCFETLNYAKDQTASET CWGNV
KTTQELYYKLNLDLGFNTFRIPPTWSGHFGDGPDYKISEAWMKRVHEIVDYALNADGYA
ILNIHETWNHAFQKNLES AKKILVAIWKQIAAEFADYDEHLIFEGMNEPRKVGDAAEW
NGGDYEGWNFVNEMNDL FVK TIRATGGNNALRHLMIPTYAACVNDGAINNFRFPSPGD
DKVIVSLHSYSPYNFALNNGAGAINNFNDGSEIDWVMNTINSSFISKGIPVIIIGFEGAMNR
NNEADREKWAEYYIKKATSLGIPCVIWDNGYFEGEGERFGLIDRKS LNVVYPRLVNGL
MKGLGDKKPKTTKVRTTTTTTIQVQPTNNNDCFSTKLG YDCCNGCEVFYTDNDGKWG
VENGNWCG

>Cel6A

PKSSGGSTTTPVNP GDVDATPGDQLT LSGNPFSGVEFYINPYYVAEVDGAIAQMTDSSL
KAKAEKMKTYSNAIWLDTIKNMNSWLESNLKGALAQQTGSKKVLTVFVVYDLPGRDC
HALASNGELLANSSDAQRYKTEYIDVIEKHLKTYKSQPVALIVEPDSL ANLVTNLESTPA
CRDSESYLDGHAYLIK KLGVLP HVAMYLDIGHAFWLGWDDNREKAGKVYAKVIQSG
SPGKVRGFTDNVSNYTPWEDPTLSRGP DTEWNP CPDEKRYLEAMHKDFKAAGISSVYF
VSDTSRNGHKTDRKH PGEWCNQTGVGIGAR PQASPISGMDYLDAFYWIKPLGESDGT S
DESAVRFDGYCGHETAMKPAPEAGQWFQKHFEQLENANPPL

>Cel48

MPSIRTALTLLGASAVIAAPT KRANDAYAQRVIDTYNAV TGNGSYRSDYFSPENVPYHS
VETMMVEAPDQGHESVSETYSFWWLEANGKMTGNYDGVNTAWSYLEKHIIPDSKN
QPGMSRYNSSSPATYAPEDDDITNYP SKLIFQDGLVGQDPIAAELQQA YGTWDIYAMH
WIIDGDNWYGYGQQGDGTTKPSFINTFQRGPSESAWKTVP HPCWEEFKWGGRNGFLDL
FTVDNSYAKQWRYTAAPDADARAIQAAYFAYIWAKEDGQNLSSLAGKAAKLG DYLR Y
AHYDKYFKKIGNCIGYQQCQAGSGKNSAHYLISWYFSWGGGLQGDWSWRIGSSHTHT
GYQNPLAAWVLSTESAFKPKGSTAVKDWATSLDRQLELFRWLQAAEGCIAGGATNSW
QGHYAQPSPDITSFYGMYYDWQPVYHDPSPN NWTGMQGWGMERVCSLYFLSGNEKA
GKVCQDWAKWVKNNVRVNGSEVVHATNFSWSGNPDEWK PANFNKSGLNASLHGTVS
TEGIDIGSLASIIKGLMWVSMKDG DQEGIDLA VQVMDAIEQYRDDHGYGVEEAREDYA
KFNEEVYIPSGWTGKNAQGANIRNGVKFIDIRPKYKQDPDWPQIENFLNGGEAPRFTYH
RFAWAQTEIAVANGLISYGLSDGSGVVP TPKPGSCAAA IKAQGYKCCSANCEVVYTDADG
DWGVENDEWCGCGGGEPECPAAITNQGYPCCSTCGTVYYT DGDGDWGVENGDWCG
MPTDC

>XYL11

GSGTSGTADFPYAKVYIGNQQNQPSYQPTTTRNNSQPTSPATGGSSGEIC SASGHSGEST
KVTSNKVGSINGIGYELWADSGNNSATFYSDGSFCTFQNAKDYL CRSGLSFDSTKTHS
QIGHLYADFKLVKQGISNV DYSYVGIYGWTRSPLVEFYIVDNWLSQYRPGDWVGNKKH
GDFTIDGAQYTVYENTRTGPSIDGNTQFKQYFSIRKTARDCGTIDITAHFQQWEKLGMT
MGKMHEAKVLGEAGSNGSGTSGTADFPYAKVYTGSSSNASSNNASSNNNNNNSSN
NSSNSNCAARWAQCGGNGFN GAKCCKEGTCVQMNQWYSQCQ

>BGL1

FATLLAASAACKCIVKPAADAAADVASDAEVSASDEENPLDNGEDSADDTPKGKLP
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YHKFDEDIKMMKIMNANHYRFSISWSRLFPDGQAKKVDGEWNVNEAGAQQYYDMVIDT
LLDNDIIPLATLYHWDLPYALHEKYGGWLDYHSQDFAKYAEFCFERYGDRVKGWITI
NEPWVNCVAGYKNGPGKAPYRCTGEAPRKLANDTTGLDLEGGCSYEIGPSQYTKDSEV
LPANRPPQKLEDVWCSHNILLGHAQAVKVYREKFKQKEQKGLIGITIDGEAQIPWEEPGM
SEEEYENNKKYANLAAEFRIGWYSDPVMFGDYPEVSVKQRNGKDMVFTDEEKKLLKGS
SSDFLGWNTYTSHWAAQVKNEDGSYVEPPTDEEASYDNSKRDMWDDNCKGRGDGWT
CIPPTLGSQAGSSWNTKYGPTIRVGLNWFSKRYEGIIKNGIITENGCAQPNYKVS RANDQ
VTLDYWKSIGKEEFADTYDEDIIEDESLEGLTIHDTYRIDWYKQYLENRLAYVEDGV
DVRGYMAWSLVDFEWENGYETFRGMTYIDFYNDKDLKRVPKDSL TFLGQWYLDNV
EQE

>BGL3

MKFSSVLSTVALLFVSKSLAITWEEADAKAREWCSDLTNEEKIALITGRENMTGVCVGS
DPIERKGFKGLCLQDGPAGVRFKGTATSWQASINTAATFDRTLLRKVGEAQGNEFYQ
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QENNRGASTSNVPEQALWEVYMEPFYRTVVEGEAGTVMASYNAINGTYSTQNKRLT
DYLKGGKLGFKGFVMSDWWAIFDVEKSFNGTDMNMPGGKYWGPDYVGDSFWGEHIQ
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NVLLKNADNILPIKSKKVKKIAVIGKDSMPPTLCEDMKCANGTLPLGWGSGTTDFKYV
VDPLSAITKRAKSKISVVSYGEDDAKKA AKVAKKADVAVFVQADS GEEYITVEGNA
GDRLNLDLWHGGNELIEAVAAVNKNTIVVIHAPGPVNPFLDKVKGIIFAGLPGQETGN
AITDVLFGDVNPSGHLPYTWAPREDYPTDVKYEPEYDPDGGEKMTVYDYNEGLFVGYR
WFDKKEIPTAFGYGLSYTTFEFSLEATMEDDGLYVSLTVTNTGKVAGAAVPMIFLSF
PESVKYPIRLFKGFDKVMLKAGESKETTIFIDAHGLSYFDVEKMEYVRPKDGEFTVYAG
SNARDLPLNVKVLANGDEIDSDNDNSDEEINEDIEDVEDSSDDEDEKDLKKRFYKLY

>Swol

MKSIQALGLSLLIAGTYAKCQTGYECCKTCDVSYTDDDGEWGIEDGQWCGIDTEKCNK
SGDDTPVTSECWSKELGFDCKSTTEVVAADASGK WGVENDQWCGIIESSATEDSCGA
YPCCKTCDSTYEDEDGKWAIENGDWCLLKKSCGDKPVDSTTTKATEPTTKSSGGVSRT
TLADGGILESGVFSTLPPSHTQPPYPHAENTGLASGAKWTLVDNVCVAMYCEDDLSE
NCDECGGVAGENGCVSVDPALCKSGIWPEVHDASDQPWKYSRSTHFGLTYGGACGFG
LYGLCTPKFNETGELCQRFCEHYPDLCADPEDISLRGNFCAPNGNYTQFWSSSLPGDYD
NYLSCGECYEVERTLPDGSSYPEGEKTDNIIQVLDSCPCTANAKWCCGSGTDHCNEIDF
KYGCPPEESFHVDLSDIAMSRLQTNDPY GEMREGVPIKYKRVPCPVKGNIHVWLRPGA
GPWYFALTVVNVANMGSLVAVEVQQADGK WVPLVRDPNYSSTRPQERYGSWTVKQG
TGFETPMNMRFDSAFNTLTAENAIKDWGDPDFDMDFLYIDTGVQFPMP

>Bgxl

MKSLSLFFAYVLLVTPYLLVKATNVLTVECNKLRRATHCANGSLYGITETTPRDYKSL
VDPLHPFVMRNPARGGNGNQHYPYGDAIKVARRLADTPGALVSVDLPDMLPGWPYRWP
GMQSWLNQVKS FINDKKASGLKNWYGLEIWNPDGTWNNNSNGSFEEMWKQTYQVIR
QADPNEKIIGPCYSWYTDDKLRNFLKYAKANNCLPDIISWHELSGIDGVSSHLRSYREIE

KSLGIPELPISINEYCDAEHELEGQPGSSARFIGKFERYKVDTAMITWWFVYPYPGRLGSL
ATDTQKGAGWYFYKWYGDMTGDMLYVKPPDNSKLV DGAACL

Table S1. Compositional analysis of pretreated corn stover and switchgrass.

Plant	Pretreatment	Glucan (%)	Hemicellulose (%)	Xylan (%)	Lignin (%)
Corn stover	Untreated	45	36	30	18
	Alkali	55	34	28	9
	Acid	42	25	22	33
	Ionic Liquid	31	26	17	43
Switchgrass	Untreated	40	38	30	23
	Alkali	48	34	30	18
	Acid	56	13	13	31
	Ionic Liquid	33	33	28	35

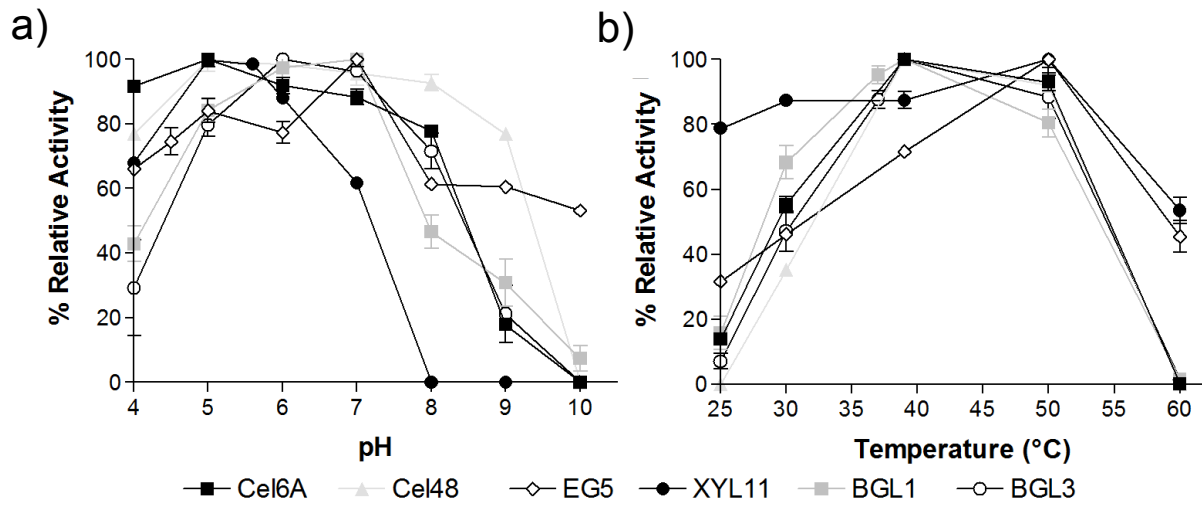


Figure S1. Optimal enzyme conditions. a) Optimal pH and b) Optimal Temperature. For all graphs, all values presented as relative specific activities. Error bars represent standard deviation of triplicate (n=3) samples.

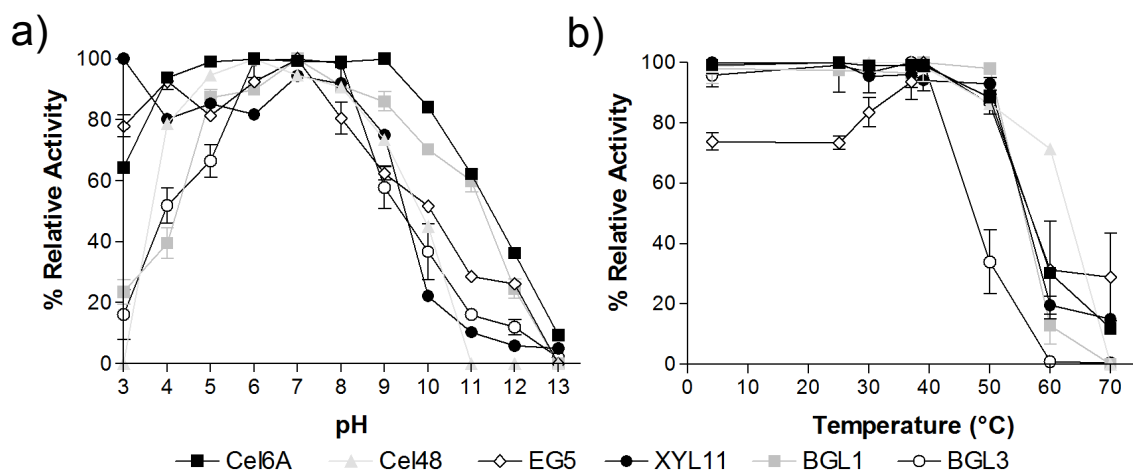


Figure S2. Enzyme stability. a) pH stability and b) thermal stability. For all graphs, all values presented as relative specific activities. Error bars represent standard deviation of triplicate (n=3) samples.

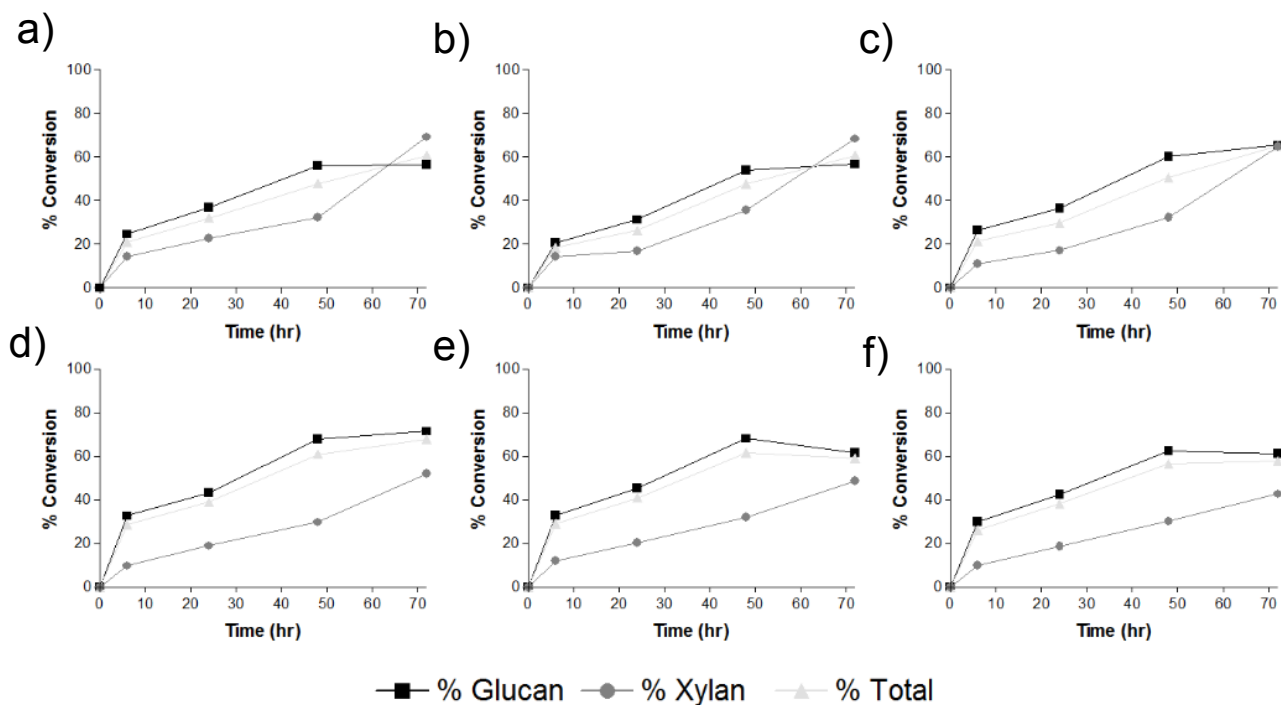


Figure S3. Enzymatic cocktail design examining ratios of cellulose-degrading cocktail and xylan-degrading cocktail on acid-pretreated corn stover (a-c) and switchgrass (d-f).

Cel6A:EG5:Bgxg1:XYL11 ratios are as follows: **a** and **d**: 30:10:25:35; **b** and **e**: 40:13:23:24; and **c** and **f**: 20:7:27:46.

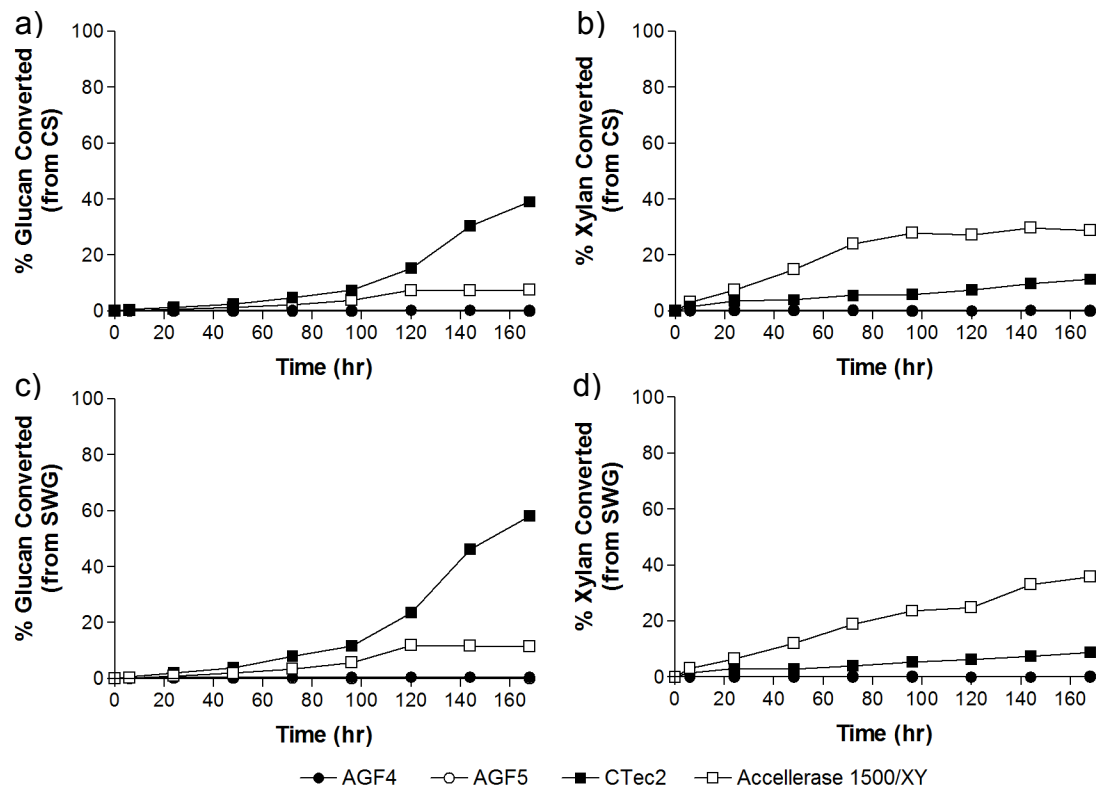


Figure S4. Enzymatic cocktails performance against untreated corn stover (a - glucan, b - xylan), and switchgrass (c - glucan, d - xylan).

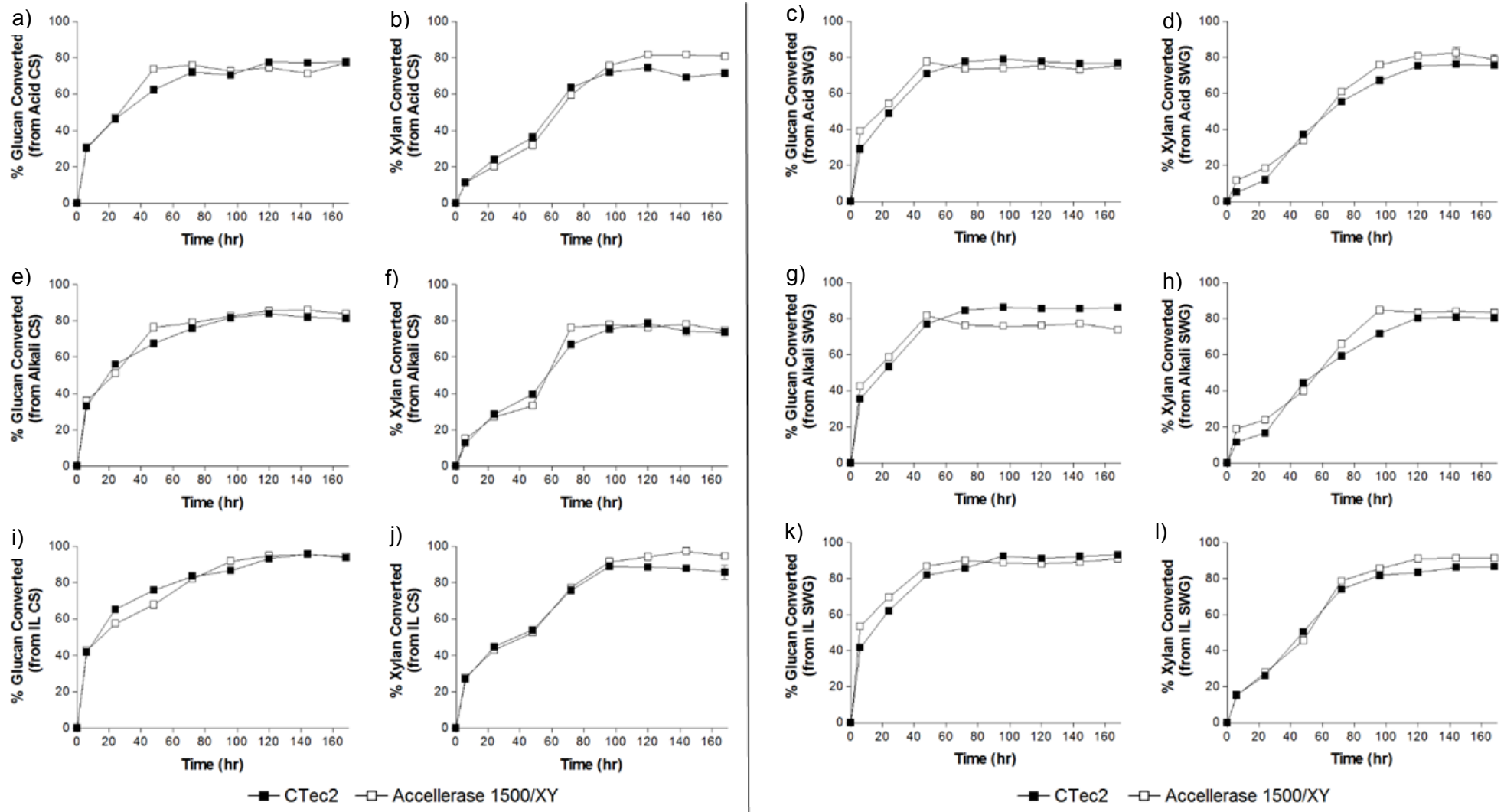


Figure S5. Commercial enzyme preparations (CTec2 and Accellerase 1500/XY) tested on various pretreatments of corn stover (CS) and switchgrass (SWG). a-b) Acid-treated corn stover, c-d) Acid-treated switchgrass, e-f) Alkali-treated corn stover, g-h) Alkali-treated switchgrass, i-j) Ionic-liquid-treated corn stover, k-l) Ionic-liquid-treated switchgrass. For a, c, e, g, i, k, the percentage of glucan converted is shown. For b, d, f, h, j, l, the percentage of xylan converted is shown.

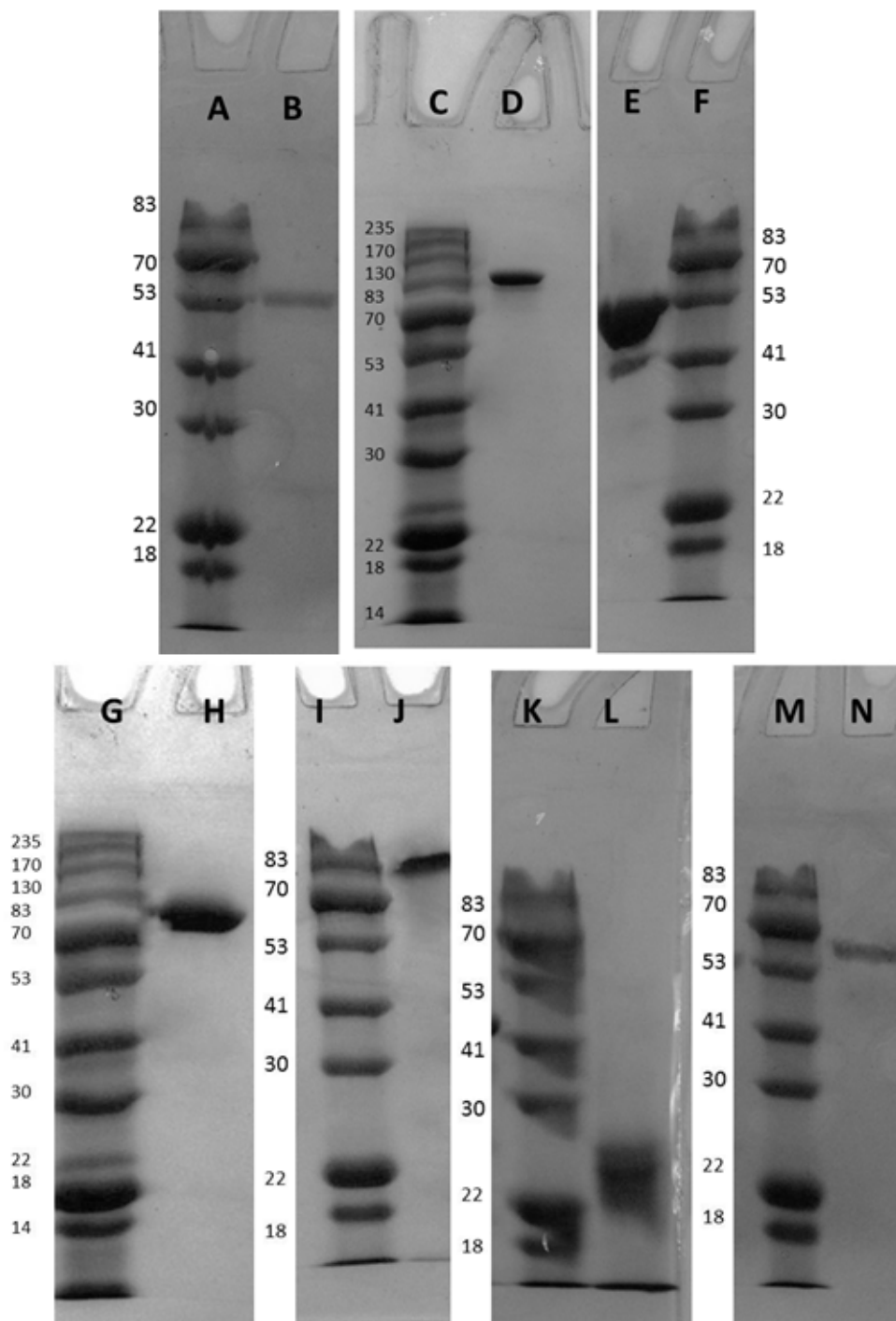


Figure S6. SDS-PAGE analysis of proteins. 12.5% SDS-PAGE analysis of recombinant proteins. *Lane A, C, F, G, I, K, and M:* Pre-stained Protein Ladder (Caisson Labs, Smithfield, Utah). *Lane B:* Cel6A, ~55 kDa; *Lane D:* Cel48, ~86 kDa; *Lane E:* EG5, ~49 kDa; *Lane H:* BGL1, ~75 kDa; *Lane J:* BGL3, ~80 kDa; *Lane L:* XYL11, ~25 kDa; *Lane N:* Swol, ~67 kDa. Protein molecular weights shown are consistent with the predicted protein size with linker and double-histidine tag.

Table S2. Properties of enzymes with exoglucanase activity All values listed are from enzymatic activities against Avicel. “-“, not reported in the study. Rows of anaerobic fungi are highlighted.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mg/mL)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	Cel6A	2.03	0.00476	73.5	5	4-10	39	4-50	Cellobiohydrolase	This Study
	Cel48	0.20	1.257	0.218	5	4-9	39	4-60	Cellobiohydrolase Endoglucanase	This Study
<i>Aspergillus nidulans</i>	Exo-I	0.348	50	0.8	5.5	-	50	-	None	1
	Exo-II	0.785	38.4	1.3	-	-	-	-	None	1
	Exo-III	0.296	37.7	0.2	-	-	-	-	None	1
<i>Chrysosporium lucknowense</i>	EG25	0.05	-	-	4.5-6	5-8.5	60-70	60-75	Endoglucanase	2
	EG28	0.02	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase	2
	EG44	0.06	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase Xylanase	2
	EG47	0.07	-	-	4.5-6	5-8.5	60-70	60-75	Endoglucanase Xylanase	2
	EG51	0.19	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase Xylanase	2
	EG60	0.08	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase β-glucosidase	2
	CBHI	0.19	-	-	4.5-6	5-7	60-70	60-75	Cellobiohydrolase Endoglucanase β-glucosidase	2
	CBHII	0.08	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase Cellobiohydrolase	2
<i>Fusarium oxysporum</i>	Xylanase	0.02	-	-	7.4	6-8	50	to 50	Endoglucanase Xylanase	3
<i>Fusarium solani</i>	Cellulase	1.34	-	-	5	5-7	65	to 65	β-glucosidase	4
<i>Gloeophyllum trabeum</i>	Cel5A	0.0045	-	-	-	-	-	-	Endoglucanase	5
	Xyn10A	0.0018	-	-	-	-	-	-	Xylanase	5
<i>Humicola grisea</i>	Exoglucanase I	0.00128	-	-	6-8	4-14	55-70	30-75	β-glucosidase Endoglucanase Cellobiohydrolase	6
<i>Neocallimastix patricianum</i>	CelA	1.1	-	-	6	5.2-11.3	50	to 70	Endoglucanase	7
	CelA	26.8	-	-	-	-	-	-	Endoglucanase	8
	CelB	1.4	-	-	-	-	-	-	Endoglucanase Cellobiohydrolase	8
	CelC	0.9	-	-	-	-	-	-	Endoglucanase	8
	CelD	0.18	-	-	-	-	-	-	Endoglucanase	9

									Cellobiohydrolase	
<i>Orpinomyces joyonii</i>	CelA	0.12	-	-	4	3-7	40	to 50	Endoglucanase	10
<i>Orpinomyces</i> sp. PC-2	CelA	0.062	-	-	4.8	4.3-6.8	50	to 40	Endoglucanase	11
	CelC	0.113	-	-	5.6-6.2	4.6-7	40	to 40	Endoglucanase	11
<i>Orpinomyces</i> sp. Y102	CelC7	20.27	-	-	6.5	4-11	40	30-50	Endoglucanase	12
<i>Penicillium brasilianum</i>	EGa	0.04	-	-	-	-	-	-	Endoglucanase	13
	EGb1	0.02	-	-	-	-	-	-	Endoglucanase	13
	EGb2	0.06	-	-	-	-	-	-	Endoglucanase	13
	CBHa	0.11	-	-	-	-	-	-	Cellobiohydrolase	13
	CBHb	0.07	-	-	-	-	-	-	Cellobiohydrolase	13
<i>Penicillium funiculosum</i>	Cellulase	-	172	-	5	-	50	to 60	β -glucosidase	14
									Cellobiohydrolase	
<i>Pichia pastoris</i>	EXG1	1.07	3.5	192	-	-	-	-	β -glucosidase	15
									β -xylosidase	
<i>Piromyces rhizinflatus</i>	CbhYW23-2	6.2	-	-	6	-	46.4	to 50	Endoglucanase	16
<i>Piromyces</i> sp. E2	Cel6A	0.0053	-	-	-	-	-	-	Cellobiohydrolase	17
<i>Termitomyces</i>	Cellulase IF	24.6	-	-	4.4	3-5.5	55	to 42	Endoglucanase	18
<i>Trichoderma harzianum</i>	EXG	49.2	-	-	5.5	-	28	-	β -glucosidase	19
									Endoglucanase	
	Cellulase	130	-	-	4-5	-	37-40	to 40	None	20
<i>Trichoderma neapolitana</i>	CelB	0.71	-	-	6-6.6	-	106	to 106	Cellobiohydrolase	21
<i>Trichoderma reesei</i>	CBHII	0.001	-	-	-	-	-	-	Cellobiohydrolase	22
	EGI	0.4	-	-	-	-	-	-	Endoglucanase	22
	XYL9	0.00005	-	-	-	-	-	-	Xylanase	22
	XYL5.5	0.00005	-	-	-	-	-	-	Xylanase	22
	CBHI	0.0105	-	-	4	2-10	60	to 50	Cellobiohydrolase	23
	CBHII	0.0205	-	-	5	3-8	55	to 50	Cellobiohydrolase	23
	EGI	0.014	-	-	4	2-8	60	to 45	Endoglucanase	23
									Xylanase	
									Cellobiohydrolase	
		EGIII	0.0116	-	-	4	2-6	70	to 55	Endoglucanase
	CBH I	0.014	-	-	-	-	-	-	Cellobiohydrolase	24
	CBH II	0.027	-	-	-	-	-	-	Cellobiohydrolase	24
<i>Trichoderma viride</i>	Cel7B (EG I)	0.8	-	-	-	-	-	-	Cellobiohydrolase	25
									Endoglucanase	
	Cel2A (EG III)	0.4	-	-	-	-	-	-	Cellobiohydrolase	25
									Endoglucanase	
	Cel61A (EG IV)	0.3	-	-	-	-	-	-	Cellobiohydrolase	25
								Endoglucanase		
	Cel7A (CBH I)	0.3	-	-	-	-	-	-	Cellobiohydrolase	25
								Endoglucanase		
	Cel6A (CBH II)	0.7	-	-	-	-	-	-	Cellobiohydrolase	25
								Endoglucanase		

	Cel6B (CBH IIb)	0.2	-	-	-	-	-	-	Cellobiohydrolase Endoglucanase	25
<i>Anaerocellum thermophilum</i>	CelA	0.055	-	-	5-6	-	85-95	-	Endoglucanase	26
<i>Bacillus amyoliquefaciens</i>	Cellulase	14	-	-	7	4-9	50	40-80	Endoglucanase Cellobiohydrolase Xylanase	27
<i>Bacillus</i> sp.	CH43	0.001	-	-	5-6.5	6-10	65	to 50	Endoglucanase	28
	HR68	0.0063	-	-	5-6.5	6-8	70	to 50	Endoglucanase	28
	E-H	1.1	-	-	-	-	-	-	Endoglucanase	29
	E-L	1.1	-	-	-	-	-	-	Endoglucanase	29
	Cel	0.014	-	-	-	-	-	-	Cellobiohydrolase	30
<i>Bacillus sphaericus</i>	Cellulase	5.76	-	-	8	7-10.5	60	to 70	Endoglucanase	31
<i>Bacillus subtilis</i>	Cellulase	2380	-	-	6	6-7.5	50-60	to 50	Endoglucanase	32
<i>Caldibacillus cellulovorans</i>	CMCase	1.7	-	-	6.5-7	-	80	to 70	Endoglucanase	33
<i>Cellulomonas fimi</i>	Cex	0.00016	-	-	-	6-9	-	-	Endoglucanase	34-36
	CenA	0.00218	-	-	-	-	-	-	Endoglucanase	36
	CenB	0.00222	-	-	-	-	-	-	Endoglucanase	36
	CenC	0.00099	-	-	-	-	-	-	Endoglucanase	36
	CenD	0.00242	-	-	-	-	-	-	Endoglucanase	36
<i>Clostridium cellulolyticum</i>	CelF	0.0134	2.4	-	5.5-6	-	55	20-45	Endoglucanase	37
<i>Clostridium stercorarium</i>	Avicelase II	0.041	-	-	5-6	4.5-7	75	65-80	None	38
<i>Clostridium thermocellum</i>	CelJ	0.0098	-	-	6	5-10	60	to 70	None	39
	CelS	0.019	-	-	5-6	-	70	-	None	40
	Endoglucanase	0.17	-	-	5.2	4.5-7	62	to 70	Endoglucanase	41
<i>Pseudomonas fluorescens</i>	Cellulase	6.9	5.3	10	6.5-7	5.8	35	to 50	None	42
<i>Ruminococcus flavefaciens</i>	Exo A	0.049	-	-	5	5-5.5	-	-	None	43
<i>Streptomyces flavogriseus</i>	Exoglucanase	0.019	-	-	-	-	-	-	None	44
<i>Macrotermes mulleri</i>	Cellulase IT	21	-	-	4.4	3-5.5	55	to 42	Endoglucanase	18
	Cellulase II	36.2	-	-	4.3-4.5	3-5.5	37	to 42	Endoglucanase	18

Table S3. Properties of enzymes with cellobiohydrolase activity. Table is organized by origin, fungal, then bacterial, then plant. All values listed are from enzymatic activities against *p*-nitrophenyl- β -D-cellobioside, PNPC. “-”, not reported in the study. Rows of anaerobic fungi are highlighted.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mM)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	Cel6A	2.82	0.228	3.27	5	4-10	39	4-50	Exoglucanase	This Study
	Cel48	0.36	8.98	0.357	5	4-9	39	4-60	Exoglucanase Endoglucanase	This Study
<i>Acremonium persicinum</i>	β -glucosidase	0.00075	0.91	-	5.5	-	-	-	β -glucosidase	45
<i>Candida peltata</i>	β -glucosidase	36.7	66	75	5	3.5-6	50	25-45	β -glucosidase	46
<i>Chaetomium cellulolyticum</i>	Xylanase 3	0.1	-	-	6-7	-	50	-	Xylanase Arabinosidase	47
<i>Gloeophyllum trabeum</i>	Cel5A	0.065	-	-	6-7	-	50	-	Endoglucanase Exoglucanase Xylanase	5
	Xyn10A	12	-	-	-	-	-	-	Endoglucanase Exoglucanase Xylanase	5
	Cel12A	0.069	-	-	-	-	-	-	Endoglucanase Exoglucanase Xylanase	5
<i>Humicola grisea</i>	Endoglucanase I	0.69	-	-	6-7	4-12	40-60	30-65	Exoglucanase Endoglucanase	6
	Endoglucanase II	0.8	-	-	5-6	6-10	55	30-55	Exoglucanase Endoglucanase	6
	Exoglucanase I	8.09	-	-	6-8	4-14	55-70	30-75	β -glucosidase Endoglucanase Exoglucanase	6
	β -glucosidase 1	0.91	-	-	4-5	4-12	60-65	30-60	β -glucosidase β -galactosidase β -xylosidase	6
	β -glucosidase 2	2.97	-	-	6-8	4-12	60-65	30-50	β -glucosidase β -galactosidase β -xylosidase	6
	β -glucosidase 3	3.19	-	-	6-8	4-12	60-65	30-50	β -glucosidase β -galactosidase β -xylosidase	6
	β -glucosidase 4	1.8	-	-	6-7	4-12	50-55	30-50	β -glucosidase β -galactosidase β -xylosidase	6
	β -glucosidase 5	3.45	-	-	6	4-12	60	30-55	β -glucosidase	6

	β -glucosidase 6	3.72	-	-	6-7	4-12	65-70	30-65	β -galactosidase β -xylosidase β -glucosidase β -galactosidase β -xylosidase	6
	BGL 4	1.8	-	-	6	6-11	55	to 50	β -glucosidase β -galactosidase	48
<i>Neocallimastix patricianum</i>	CelB	1.31	-	-	-	-	-	-	Endoglucanase	8
	CelD	0.169	-	-	-	-	-	-	Exoglucanase Endoglucanase Exoglucanase	9
<i>Orpinomyces joyonii</i>	CelB2	4.9	-	-	5.5	5-7.5	40	to 50	Endoglucanase	49
<i>Orpinomyces</i> sp. PC-2	CelE	0.2	-	-	-	-	-	-	Endoglucanase	50
<i>Paecilomyces thermophile</i>	β -glucosidase	49.1	0.65	272	6.2	5-8.5	75	65-70	β -glucosidase Exoglucanase	51
<i>Penicillium brasilianum</i>	EGa	0.34	-	-	-	-	-	-	Exoglucanase	13
	EGb1	0.38	-	-	-	-	-	-	Endoglucanase Exoglucanase	13
	EGb2	1.16	-	-	-	-	-	-	Endoglucanase Exoglucanase	13
	CBHa	2	-	-	-	-	-	-	Endoglucanase Exoglucanase	13
	CBHb	0.98	-	-	-	-	-	-	Endoglucanase Exoglucanase Endoglucanase	13
<i>Penicillium funiculosum</i>	Cellobiohydrolase	0.56	-	-	4-5.5	2.5-5.5	60	to 70	Exoglucanase	52
	Cellulase	-	10	-	5	-	50	to 60	β -glucosidase Exoglucanase	14
<i>Phanerochaete chrysosporium</i>	β -glucosidase	33	-	-	4-5.2	4-8	25	to 45	β -glucosidase	53
<i>Piromyces equi</i>	Cel5A	0.1	-	-	5.1	5.1-6.4	45	to 50	Endoglucanase	54
<i>Sporotrichum thermophile</i>	β -glucosidase I	1	0.83	-	5.4	4-6.5	65	to 65	β -glucosidase	55
<i>Stachybotrys</i> sp.	β -glucosidase p21	-	2.22	59	5	5-7	50	to 60	β -glucosidase	56
<i>Trichoderma koningii</i>	β -glucosidase	6.6	1.18	-	4-5	4-6	45	to 65	β -glucosidase	57
	β -glucosidase II	9.4	0.86	-	4-5	4-6	45	to 65	β -glucosidase	57
<i>Trichoderma reesei</i>	BGL 2	0.89	-	-	6	6-11	45	to 45	β -glucosidase	48
	CBH I	0.355	-	-	4	2-10	60	to 50	β -galactosidase Endoglucanase	23
	EG I	9.19	-	-	4	2-8	60	to 45	β -glucosidase β -glucosidase	23
	BGL I	0.894	-	-	4	5-9	70	to 55	Endoglucanase β -glucosidase	23
	CBH I	0.00104	-	-	-	-	-	-	β -glucosidase Endoglucanase	58
	CBH I	0.0103	-	-	-	-	-	-	Endoglucanase	59

	CBH II	0.0064	-	-	-	-	-	-	-	β -glucosidase Endoglucanase	59
	EG I	0.133	-	-	-	-	-	-	-	β -glucosidase Endoglucanase	60
	EG II	0.0075	-	-	-	-	-	-	-	Exoglucanase Endoglucanase	60
	CBH I	0.46	-	-	-	-	-	-	-	Exoglucanase	60
	CBH II	0.048	-	-	-	-	-	-	-	Exoglucanase	60
<i>Trichoderma viride</i>	Cel7A (CBH I)	2.5	-	-	-	-	-	-	-	Exoglucanase	25
	Cel6A (CBH II)	5.7	-	-	-	-	-	-	-	Exoglucanase	25
	Cel6B (CBH IIb)	1.1	-	-	-	-	-	-	-	Exoglucanase	25
<i>Bacillus amyoliquefaciens</i>	Cellulase	24.6	-	-	7	4-9	50	40-80		Endoglucanase Exoglucanase Xylanase β -glucosidase	27
<i>Bacillus</i> sp.	Cel	2.28	-	-	-	-	-	-	-	Exoglucanase	30
	E-H	0.3	-	-	-	-	-	-	-	Exoglucanase	29
	E-L	0.6	-	-	-	-	-	-	-	Exoglucanase	29
<i>Cellulomonas fimi</i>	Exg	3	0.641	-	6	5-9	60	60-75		Exoglucanase	61
<i>Clostridium stercorarium</i>	β -glucosidase	10.1	33	-	5.5	5-7	65	to 75		β -glucosidase Exoglucanase	62
<i>Clostridium thermocellum</i>	β -glucosidase	33.3	-	-	6-6.5	5.5-7.5	60	to 68.5		β -glucosidase	63
	CelJ	0.0041	-	-	6	5-10	60	to 70		Exoglucanase	39
<i>Ruminococcus flavefaciens</i>	Exo A	0.298	-	-	5	4.5-5.5	-	-		Exoglucanase	43
<i>Thermomonospora fusca</i>	E1	0.04	-	-	-	-	-	-		Endoglucanase	58
	E5	0.0148	-	-	-	-	-	-		Endoglucanase	58
	E5cd	0.0144	-	-	-	-	-	-		Endoglucanase	58
	E6	0.00004	-	-	-	-	-	-		Endoglucanase	58
<i>Thermatoga maritima</i>	Cellulase I	0.035	-	-	6-7.5	-	95	to 80		Exoglucanase	64
	Cellulase II	0.015	-	-	6-7.5	-	95	to 80		Exoglucanase	64
	CelA	7.4	-	-	5	4-7	85-90	to 95		Exoglucanase	65
	CelB	0.05	-	-	7	5-9	85-90	to 95		Exoglucanase	65
<i>Thermatoga neapolitana</i>	CelA	69.2	0.97	69	6	-	95	to 95		Exoglucanase	21
	CelB	18.4	0.3	18	6-6.6	-	106	to 106		Exoglucanase	21

Table S4. Properties of enzymes with endoglucanase activity. Table is organized by origin, fungal, then bacterial, then plant. All values listed are from enzymatic activities carboxymethyl cellulose (CMC), unless otherwise noted. “-“, not reported in the study. Rows of anaerobic fungi are highlighted. “*” value calculated with CellG3 as substrate.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mg/mL)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	EG5	9.96 *209	0.00027 *0.000004	622 *775	7	3-8	50	4-50	β-glucosidase Xylanase	This Study
	Cel48	*0.045	*30.1	*0.05	5	4-9	39	4-60	Exoglucanase Cellobiohydrolase	This Study
<i>Aspergillus aculeatus</i>	XEG	-	3.6	260	3.4	3-3.8	30	to 50	None	66
<i>Aspergillus nidulans</i>	Endo-I	9.15	5	15	6	-	65	-	None	1
	Endo-II	8.26	4	12	5	-	50	-	None	1
<i>Aspergillus niger</i>	Cellulase	194	-	-	6-7	5-10	70	to 60	None	67
	Cellulase	45.2	14.5	85	3.8-4	1-9	45	to 25	None	68
	Cellulase	116.8	0.086	-	4	5-8	45-50	to 70	None	69
<i>Chaetomonium cellulolyticum</i>	Xylanase 1	0.06	-	-	6-7	-	50	-	Exoglucanase Xylanase	47
	Xylanase 2	0.05	-	-	6-7	-	50	-	Exoglucanase Xylanase	47
	Xylanase 3	0.03	-	-	6-7	-	50	-	Exoglucanase Xylanase	47
<i>Chrysosporium lucknowense</i>	EG25	17	-	-	4.5-6	5-8.5	60-70	60-75	Exoglucanase	2
	EG28	11	-	-	4.5-6	5-7	60-70	60-75	Xylanase	2
	EG44	59	-	-	4.5-6	5-7	60-70	60-75	Exoglucanase	2
									Xylanase	
	EG47	14	-	-	4.5-6	5-8.5	60-70	60-75	Exoglucanase	2
									Xylanase	
	EG51	52	-	-	4.5-6	5-7	60-70	60-75	Exoglucanase	2
									Xylanase	
EG60	12	-	-	4.5-6	5-7	60-70	60-75	Exoglucanase	2	
CBH I	0.2	-	-	4.5-6	5-7	60-70	60-75	Cellobiohydrolase	2	
CBH II	1.1	-	-	4.5-6	5-7	60-70	60-75	Cellobiohydrolase	2	
								Exoglucanase Xylanase		
<i>Cryptococcus</i> sp. S-2	CSCMCase	248.7	-	-	3.5	5.5-7.5	40-50	to 90	None	70
<i>Fusarium oxysporum</i>	Xylanase	5	13.1	-	7.4	5.8-8.2	50	to 50	Xylanase	3
									Exoglucanase	
<i>Fusarium solani</i>	Cellulase	4.75	-	-	5	5-7	65	to 65	β-glucosidase Cellobiohydrolase Exoglucanase	4

<i>Gloeophyllum trabeum</i>	Cel5A	120	-	-	-	-	-	-	Xylanase Exoglucanase	5
	Xyn10A	0.95	-	-	-	-	-	-	Xylanase Exoglucanase	5
	Cel12A	42	-	-	-	-	-	-	Xylanase Exoglucanase	5
<i>Humicola grisea</i>	Endoglucanase 1	9.06	-	-	6-7	4-12	40-60	30-65	None	6
	Endoglucanase 2	24.1	-	-	5-6	6-10	55	30-55	None	6
<i>Neocallimastix patricianum</i>	CelA	108	-	-	6	5.2-11.3	50	to 70	Exoglucanase	7
	CelA	15.1	-	-	-	-	-	-	Exoglucanase	8
	CelB	53.6	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	8
	CelC	20.6	-	-	-	-	-	-	Exoglucanase	8
	CelD	4.9	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	9
<i>Orpinomyces jayonii</i>	Cellulase	203	0.5	-	5.5	5-7.5	40	to 50	Exoglucanase Cellobiohydrolase Xylanase	49
	CelB2	203	-	-	5.5	5-7.5	40	to 50	Cellobiohydrolase	49
	CelA	13.2	-	-	4	3-7	40	to 50	Exoglucanase	10
<i>Orpinomyces</i> sp. PC-2	CelE	3.2	-	-	-	-	-	-	Cellobiohydrolase	50
	CelB	2.9	-	-	-	-	-	-	None	50
	CelE	72.5	-	-	6	5.5-7.5	45	40-55	None	71
	CelA	1.2	-	-	4.8	4.3-6.8	50	to 40	Exoglucanase	11
	CelC	1.1	-	-	5.6-6.2	4.6-7	40	to 40	Exoglucanase	11
<i>Orpinomyces</i> sp. Y102	CelC7	25.96	-	-	6.5	4-11	40	30-50	Exoglucanase	12
<i>Penicillium brasilianum</i>	EGa	39.8	-	-	-	-	-	-	Exoglucanase	13
	EGb1	54	-	-	-	-	-	-	Exoglucanase	13
	EGb2	36.3	-	-	-	-	-	-	Exoglucanase	13
<i>Penicillium funiculosum</i>	Cellobiohydrolase	0.05	-	-	4-5.5	2.5-5.5	60	to 70	Cellobiohydrolase	52
<i>Piromyces equi</i>	Cel5A	3.2	-	-	5.1	5.1-6.4	45	to 50	Cellobiohydrolase	54
	Cel45A	42.5	-	-	6.5	5.2-7.9	60	to 65	None	54
<i>Piromyces rhizinflatus</i>	CbhYW23-2	6.0	-	-	6	-	46.4	to 50	Endoglucanase	16
<i>Termitomyces</i> sp.	Cellulase IF	378	-	-	4.4	2.8-5.5	55	to 42	Exoglucanase	18
<i>Thermoascus aurantiacus</i>	Cellulase I	0.525	3.9	6.3	5	6-8	75	to 65	None	72
	Cellulase II	0.0031	1.2	1.1	5	2-12	60	to 65	None	72
	Cellulase III	0.24	1.9	33	5	6-8	65	to 65	None	72
<i>Trichoderma harzianum</i>	Endoglucanase	312.5	-	-	5-5.5	-	40	to 35	None	20
	EG	0.63	-	-	5.5	-	-	-	None	19
<i>Trichoderma reesei</i>	CBHI	0.0021	-	-	-	-	-	-	Cellobiohydrolase Exoglucanase β -glucosidase Xylanase	58

	CBHII	0.0014	-	-	-	-	-	-	-	Cellobiohydrolase Exoglucanase β -glucosidase Xylanase	58
	CBHI	0.2	-	-	-	-	-	-	-	Cellobiohydrolase Exoglucanase β -glucosidase Xylanase	59
	CBHII	21.5	-	-	-	-	-	-	-	Cellobiohydrolase Exoglucanase β -glucosidase Xylanase	59
	EG I	59.8	-	-	4	2-8	60	to 45		Xylanase Exoglucanase β -glucosidase β -xylosidase	23
	EG III	30.7	-	-	4	2-6	70	to 55		None	23
	EG V	9.22	-	-	4	2-6	50-65	to 80		None	23
<i>Trichoderma viride</i>	Cel7B (EG I)	7.6	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	25
	Cel2A (EG III)	2.8	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	25
	Cel61A (EG IV)	1.9	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	25
	Cel6A (CBH II)	0.1	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	25
	CMCase	0.258	0.88	3.5	4	3-7	50	30-70		None	73
	Cellulase	105	68	148	6.5	6.5-8	55	45-55		None	74
<i>Anaerocellum thermophilum</i>	CelA	1.758	-	-	5-6	-	85-95	-	-	Exoglucanase Xylanase	26
<i>Bacillus</i> sp.	CH43	0.0062	1.5	0.93	5-6.5	6-10	65	to 50		Exoglucanase	28
	HR68	0.0159	1.7	1.7	5-6.5	6-8	70	to 50		Exoglucanase	28
	Cellulase	58.9	0.48	-	9	6-12	40	to 60		None	75
	Cellulase	49.4	11.4	135	8.6-9	5-11	45	to 60		β -glucosidase Cellobiohydrolase Exoglucanase Xylanase	76
	E-H	23.5	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	29
	E-L	43.8	-	-	-	-	-	-	-	Exoglucanase Cellobiohydrolase	29
	Cellulase	71	-	-	10	6-10	50	to 60		None	77
	Cel	128	-	-	-	-	-	-	-	Cellobiohydrolase	30
	E-I	0.12	-	-	6	5-8	60	to 80		None	78

	E-II	60	-	-	7	7-10	50	to 60	None	78
	E-III	59	-	-	7	6-10	50	to 60	None	78
	Cellulase	246	-	-	6	5-7	50	to 50	None	79
<i>Bacillus amyoliquefaciens</i>	Cellulase	100	-	-	7	4-9	50	40-80	Exoglucanase Cellobiohydrolase	27
<i>Bacillus pumilus</i>	Cellulase	0.079	-	-	6	5-9	60	30-70	Exoglucanase β -glucosidase	80
<i>Bacillus sphaericus</i>	Cellulase	38.4	-	-	8	7-10.5	60	to 70	β -glucosidase	31
<i>Bacillus subtilis</i>	CelDR	0.3	-	-	6.5	-	50	to 75	None	81
	Cellulase	7001	-	-	6	6-7	50-60	to 50	Exoglucanase	32
<i>Caldibacillus cellulovorans</i>	CMCase	30.6	3.4	44.7	6.5-7	-	80	to 70	None	33
<i>Cellulomonas fimi</i>	Cex	0.01	-	-	-	6-9	-	-	Exoglucanase	34-36
	CenA	0.760	-	-	-	-	-	-	β -glucosidase Cellobiohydrolase	36
	CenB	0.928	-	-	-	-	-	-	Exoglucanase β -glucosidase Cellobiohydrolase	36
	CenC	1.016	-	-	-	-	-	-	Exoglucanase	36
	CenD	0.01	-	-	-	-	-	-	Exoglucanase	36
	Exg	20	3.18	-	6	5-9	-	60-75	Exoglucanase Cellobiohydrolase	61
<i>Cellulomonas sp.</i>	Cellulase I	162	-	-	-	-	-	-	None	82
	Cellulase CA	136	-	-	-	-	-	-	None	82
	Cellulase CB	9.7	-	-	-	-	-	-	None	82
<i>Clostridium cellulolyticum</i>	CelF	0.0014	-	-	5.5-6	-	55	20-45	Exoglucanase	37
<i>Clostridium thermocellum</i>	CelJ	13.1	4.06	164	6	5-10	60	to 70	Exoglucanase Cellobiohydrolase	39
	Endoglucanase	65.1	-	-	5.2	4.5-7	62	to 70	Exoglucanase	41
<i>Marinobacter sp.</i>	CMCase	2548.75	-	-	9	4-12	27-35	-	None	83
<i>Salinivibrio sp.</i>	Cellulase	32.4	3.03	143	7.5	6.5-8.5	35	10-40	None	84
<i>Sinorhizobium fredii</i>	CMCase	3.822	-	-	7	6-9	35	30-45	None	85
<i>Thermomonospora curvata</i>	Cellulase	0.3	3.5	213	6	5-8	65	to 65	None	86
<i>Thermomonospora fusca</i>	E1	5.41	-	-	-	-	-	-	None	58
	E2	0.369	-	-	-	-	-	-	None	58
	E2cd	0.344	-	-	-	-	-	-	None	58
	E3	0.0013	-	-	-	-	-	-	None	58
	E4	0.122	-	-	-	-	-	-	None	58
	E5	2.840	-	-	-	-	-	-	None	58
	E5cd	2.480	-	-	-	-	-	-	None	58
	E6	0.0642	-	-	-	-	-	-	None	58
<i>Thermatoga maritima</i>	Cellulase I	1.2	-	-	6-7.5	-	95	to 80	Exoglucanase Cellobiohydrolase	64

	Cellulase II	1.5	-	-	6-7.5	-	95	to 80	Exoglucanase	64
	CelA	302	-	-	5	4-7	85-90	to 95	Cellobiohydrolase	65
	CelB	0.89	-	-	7	5-9	95	to 95	Exoglucanase	65
									Cellobiohydrolase	
<i>Thermatoga neapolitana</i>	CelA	1219	0.25	1219	6	-	95	to 95	β -glucosidase	21
	CelB	1536	0.22	1536	6-6.6	-	106	to 106	Cellobiohydrolase	21
									β -glucosidase	
									Cellobiohydrolase	
<i>Ergates faber</i>	Cellulase A	30	20	-	4-4.7	-	37	to 55	None	87
	Cellulase B	56	28	-	5.2	-	37	to 45	None	87
	Cellulase C	157	6	-	5.2	-	37	to 60	None	87
<i>Macrotermes mulleri</i>	Cellulase IT	360	-	-	4.4	2.8-5.5	55	to 42	Exoglucanase	18
	Cellulase II	274	-	-	4.3-4.5	2.8-5.5	37	to 42	Exoglucanase	18
<i>Panesthia cribrata</i>	EG1	22.2	9.4	1850	-	-	-	-	None	88
	EG2	88.3	6.8	3440	-	-	-	-	None	88
<i>Panesthia hilaris</i>	Cellulase	150	-	-	5.5	4-7	-	-	None	89
<i>Phaseolus vulgaris</i>	Cellulase 4.5	17	-	-	5.7-6.2	6-9	35-43	to 30	None	90
	Cellulase 4.8	814	-	-	4.8-5.6	6.5-8	45-60	to 36	None	90

Table S5. Properties of enzymes with β -glucosidase activity. Table is organized by origin, fungal, then bacterial, then plant. All values listed are from enzymatic activities against *p*-nitrophenyl- β -D-glucopyranoside, PNPG. “-”, not reported in the study. Rows of anaerobic fungi are highlighted.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mM)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	BGL1	1.063	0.164	5.4	7	5-9	39	4-50	None	This Study
	BGL3	0.8	0.275	1.6	6	6-8	39	4-39	β -xylosidase	This study
	EG5	0.29	84	0.45	7	3-8	50	4-50	Endoglucanase	This Study
	XYL11	0.22	0.216	0.28	5	3-9	50	4-60	Xylanase	This Study ⁹¹
	Bgxl	73.4	0.000013	769	6	4-12	39	4-70	β -xylosidase β -galactosidase Xylanase	
<i>Acremonium persicinum</i>	β -glucosidase	0.0183	0.3	-	5.5	-	-	-	Cellobiohydrolase	45
<i>Aspergillus fumigatus</i>	β -xylosidase	0.76	-	-	4.5	2-8	75	to 65	β -xylosidase	92
	β -Glucosidase	1.03	-	-	4.5	2-8	65	to 60	β -xylosidase	92
<i>Aspergillus nidulans</i>	B-Gluco-I	4.52	0.24	8	5	-	65	-	None	1
	B-Gluco-II	0.55	0.12	0.7	6	-	35	-	None	1
<i>Aspergillus niger</i>	β -xylosidase	0.15	-	-	6.7-7	-	42	to 46	β -xylosidase	93
	β -xylosidase 1	0.2	-	-	-	-	-	-	β -xylosidase	94
	β -xylosidase 2	0.3	-	-	-	-	-	-	Arabinosidase β -xylosidase Arabinosidase	94
<i>Aureobasidium</i> sp.	β -xylosidase	15.6	-	-	3.5	3.5-9	80	to 80	β -xylosidase	95
<i>Candida peltata</i>	β -glucosidase	108	2.3	221	5	3.5-6	50	25-45	Cellobiohydrolase	46
<i>Chrysosporium lucknowense</i>	EG60	0.12	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase	2
	CBH1	0.02	-	-	4.5-6	5-7	60-70	60-75	Cellobiohydrolase	2
<i>Fusarium oxysporum</i>	Xylanase	0.9	-	-	7.4	5.8-8.2	50	to 50	Xylanase Endoglucanase Exoglucanase	3
<i>Fusarium solani</i>	Cellulase	4.65	-	-	5	5.7	65	to 65	Cellobiohydrolase Endoglucanase Exoglucanase	4
<i>Humicola grisea</i>	Exoglucanase 1	1.14	-	-	6-8	4-14	55-70	30-75	Cellobiohydrolase Endoglucanase Exoglucanase	6
	β -glucosidase 1	85.7	0.16	64	4.5	4-12	60-65	30-60	Cellobiohydrolase β -galactosidase β -xylosidase	6
	β -glucosidase 2	12.5	0.25	21	6-8	4-12	60-65	30-50	Cellobiohydrolase β -galactosidase β -xylosidase	6

	β -glucosidase 3	12.6	0.22	23	6-8	4-12	60-65	30-50	Cellobiohydrolase β -galactosidase β -xylosidase	6
	β -glucosidase 4	8.34	0.34	8.7	6-7	4-12	50-55	30-50	Cellobiohydrolase β -galactosidase β -xylosidase	6
	β -glucosidase 5	17.9	0.56	18	6	4-12	60	30-55	Cellobiohydrolase β -galactosidase β -xylosidase	6
	β -glucosidase 6	33.4	0.12	40	6-7	4-12	65-70	30-65	Cellobiohydrolase β -galactosidase β -xylosidase	6
	BGL I	26.1	0.34	25	6	6-11	55	to 50	Cellobiohydrolase β -galactosidase	48
<i>Neocallimastix frontalis</i>	β -xylosidase	0.2	2.98	0.27	6.4	-	37	to 40	β -xylosidase	96
	β -glucosidase	9.0	0.031	-	-	-	-	-	Arabinosidase β -galactosidase	97
	β -glucosidase	15	0.55	-	6	5-8	50	to 40	Cellobiohydrolase None	98
<i>Orpinomyces</i> sp. strain PC-2	β -glucosidase	33.5	0.39	47.5	6.2	5-8	50	to 65	Cellobiohydrolase	99
<i>Paecilomyces thermophila</i>	β -glucosidase	97.2	0.27	780	6.2	5-8.5	75	65-70	Exoglucanase	51
<i>Penicillium funiculosum</i>	Cellulase	-	0.77	-	5	-	50	to 60	Cellobiohydrolase Exoglucanase	14
<i>Phanerochaete chrysosporium</i>	β -glucosidase	-	0.096	-	4-5.2	4-8	25	to 45	None	53
<i>Pichia pastoris</i> X-33	EXG1	37.5	-	-	-	-	-	-	β -xylosidase Exoglucanase	15
<i>Piromyces communis</i>	β -glucosidase	20	0.52	-	6	5-8	50	to 40	None	98
<i>Piromyces</i> sp. E2	β -glucosidase	0.25	1.0	66.5	5.7-6.3	-	47	to 37	Cellobiohydrolase	100
<i>Sphaeromonas communis</i>	β -glucosidase	46	0.51	-	6	5-8	50	to 40	None	98
<i>Sporotrichum thermophila</i>	β -glucosidase I	89	0.29	-	5.4	4-6.5	65	to 65	None	55
	β -glucosidase A	0.595	0.5	-	5.6	-	50	to 40	None	101
	β -glucosidase B	1.295	0.18	-	6.3	-	50	to 40	None	101
<i>Stachybotrys</i> sp.	β -glucosidase p21	0.33	0.27	78	5	5-7	50	to 60	None	56
<i>Thermoascus aurantiacus</i>	β -glucosidase	232	0.52	6500	5	6-8	70	to 75	None	72
	β -glucosidase	0.3	0.11	-	4.5	4.4-5.2	80	70-90	Cellobiohydrolase	102
<i>Trichoderma harzianum</i>	BGL	0.35	-	-	-	-	28	-	None	19
	β -xylosidase	0.28	-	-	4-4.5	-	60	-	β -xylosidase β -galactosidase Arabinosidase	103
<i>Trichoderma koningii</i>	β -glucosidase	16	0.37	-	4-5	4-6	45	to 65	Cellobiohydrolase	57
	β -glucosidase II	14	0.85	-	4-5	4-6	45	to 65	Cellobiohydrolase	57
<i>Trichoderma reesei</i>	BGL2	23.9	2.22	40	6	6-11	45	to 45	Cellobiohydrolase β -galactosidase	48

	BGLI	767.7	-	-	4	5-9	70	to 55	Cellobiohydrlase	23
	CBH I	5.5	-	-	-	-	-	-	Cellobiohydrolase	59
									Endoglucanase	
	CBH II	33	-	-	-	-	-	-	Cellobiohydrolase	59
									Endoglucanase	
	CBH III	0.00001	-	-	-	-	-	-	Cellobiohydrolase	22
									Xylanase	
									Mannanase	
									β -xylosidase	
									Exoglucanase	
	EG I	0.00005	-	-	-	-	-	-	Cellobiohydrolase	22
									Xylanase	
									Mannanase	
									β -xylosidase	
									Exoglucanase	
	XYL 9	0.00001	-	-	-	-	-	-	Xylanase	22
									Mannanase	
									β -xylosidase	
	XYL 5.5	0.000001	-	-	-	-	-	-	Xylanase	22
									Mannanase	
									β -xylosidase	
<i>Trichoderma viride</i> T100-14	β -glucosidase	5.6	-	-	-	-	-	-	Cellobiohydrolase	25
<i>Bacillus</i> sp. KSM-S237	Cellulase	5.4	-	-	8.6-9	5-11	45	to 60	Endoglucanase	76
									Cellobiohydrolase	
									Exoglucanase	
									Xylanase	
<i>Bacillus pumilus</i> EB3	Cellulase	0.038	-	-	6	5-9	60	30-70	Exoglucanase	80
									Endoglucanase	
<i>Bacillus sphaericus</i> JS1	Cellulase	3.07	-	-	8	7-10.5	60	to 70	Endoglucanase	31
<i>Cellulomonas fimi</i>	CenA	1.6	-	-	-	-	-	-	Exoglucanase	36
									Endoglucanase	
									Cellobiohydrolase	
	Exg	0.9	-	-	6	5-9	-	60-75	Xylanase	61
									Endoglucanase	
									Exoglucanase	
									Cellobiohydrolase	
<i>Clostridium stercorarium</i>	β -glucosidase	31.6	0.8	-	5.5	5-7	65	to 75	Cellobiohydrolase	62
<i>Clostridium thermocellum</i>	β -glucosidase	127	-	-	6-6.5	5.5-7.5	60	to 68.5	Cellobiohydrolase	63
<i>Thermotoga neapolitana</i>	CelA	0.01	-	-	6	-	95	to 95	Endoglucanase	21
									Cellobiohydrolase	
	CelB	0.18	-	-	6-6.6	-	106	to 106	Endoglucanase	21
									Cellobiohydrolase	
<i>Panesthia cribrata</i>	GD1	9	10.6	-	-	-	-	-	Endoglucanase	88
	GD2	3	13.8	-	-	-	-	-	Endoglucanase	88

Table S6. Properties of enzymes with β -xylosidase activity. Table is organized by origin, fungal, then bacterial, then plant. All values listed are from enzymatic activities against *p*-nitrophenyl- β -D-xylopyranoside, PNPX. “-”, not reported in the study. Rows of anaerobic fungi are highlighted.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mM)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	BGL3	0.06	50.6	0.068	6	6-8	39	4-39	β -xylosidase	This study 91
	Bgxgl	11.5	0.00485	127	6	4-12	39	4-70	β -glucosidase β -galactosidase Xylanase	
<i>Acromonium persicinum</i>	β -glucosidase	0.002	-	-	5.5	-	-	-	β -glucosidase	45
<i>Aspergillus awamori</i>	β -xylosidase	34.1	0.003	476	6.5	-	70	to 70	Xylanase	104
<i>Aspergillus carbonarius</i>	β -xylosidase	3.29	0.198	3.64	4	3.5-6.5	60	to 50	Arabinosidase	105
<i>Aspergillus fumigatus</i>	β -xylosidase	27.5	2	-	4.5	2-8	75	to 65	β -glucosidase	92
	β -Glucosidase	1.03	-	-	4.5	2-8	65	to 60	β -glucosidase	92
<i>Aspergillus nidulans</i>	β -xylosidase	1.2	1.1	25.6	5	4-6	50	to 45	None	106
<i>Aspergillus niger</i>	β -xylosidase	5.2	0.22	-	6.7-7	-	42	to 46	β -glucosidase	93
	β -xylosidase 1	60.2	-	-	-	-	-	-	β -glucosidase Arabinosidase	94
	β -xylosidase 2	60.9	-	-	-	-	-	-	β -glucosidase Arabinosidase	94
<i>Aspergillus phoenicis</i>	β -xylosidase	821	2.36	-	4-4.5	4-6	75	to 60	None	107
<i>Aspergillus pulverulentus</i>	β -xyII	32	-	-	2.5-3.5	1.5-6.5	60	to 50	None	108
	β -xyIII	12	-	-	4-5	1.5-6.5	60	to 50	None	108
<i>Aureobasidium</i> sp.	β -xylosidase	626	2	940	3.5	3.5-9	80	to 80	β -glucosidase	95
<i>Aureobasidium pullulans</i>	β -xylosidase	7.3	-	-	4.5	2-9.5	80	to 70	None	109
<i>Chaetomonium cellulolyticum</i>	Xylanase 3	0.03	-	-	6-7	-	50	-	Xylanase	47
									Arabinosidase Endoglucanase	
									Xylanase Endoglucanase Exoglucanase	3
<i>Fusarium oxysporum</i>	Xylanase	0.5	-	-	7.4	5.8-8.2	50	to 50	Xylanase Endoglucanase Exoglucanase	
<i>Fusarium proliferatum</i>	β -xylosidase	53	0.77	-	4.5	-	60	-	None	110
<i>Fusarium verticillioides</i>	β -xylosidase	57	0.85	-	4.5	-	65	-	None	111
<i>Humicola grisea</i>	β -glucosidase 1	4.38	-	-	4.5	4-12	60-65	30-60	β -glucosidase β -galactosidase Cellobiohydrolase	6
	β -glucosidase 2	0.07	-	-	6-8	4-12	60-65	30-50	β -glucosidase β -galactosidase Cellobiohydrolase	6
	β -glucosidase 3	0.08	-	-	6-8	4-12	60-65	30-50	β -glucosidase β -galactosidase Cellobiohydrolase	6

	β -glucosidase 4	0.21	-	-	6-7	4-12	50-55	30-50	β -glucosidase β -galactosidase Cellobiohydrolase	6
	β -glucosidase 5	0.36	-	-	6	4-12	60	30-55	β -glucosidase β -galactosidase Cellobiohydrolase	6
	β -glucosidase 6	0.17	-	-	6-7	4-12	65-70	30-65	β -glucosidase β -galactosidase Cellobiohydrolase	6
	β -xylosidase	19.6	1.37	13	6.5	4-9	55	to 50	None	112
<i>Neocallimastix frontalis</i>	β -xylosidase	4.3	0.33	-	6.5	-	35	-	Xylanase	113
	β -xylosidase	0.9	2.98	0.27	6.4	-	37	to 40	β -glucosidase Arabinosidase	96
	β -xylosidase	16	0.34	-	6.5	5-8	35	to 40	None	98
<i>Neocallimastix patriciarum</i>	β -xylosidase I	30.4	0.59	38	6	5-8	50	45-60	Arabinosidase	114
	β -xylosidase II	8.7	0.13	8.9	6	5-7	40	25-45	None	114
<i>Neurospora crassa</i>	β -xylosidase	0.26	0.047	-	4.5-5	-	55	-	Cellobiohydrolase	115
<i>Penicillium wortmanni</i>	Xylosidase 1	22	4.2	42.6	3-4.5	-	55-65	-	None	116
	Xylosidase 2	40	2.3	73.2	3-4.5	-	55-65	-	None	116
	Xylosidase 3	61.5	1.0	88.2	3-4.5	-	55-65	-	None	116
	Xylosidase 4	30.5	3.3	106.6	3-4.5	-	55-65	-	None	116
<i>Pichia pastoris X-33</i>	EXG1	11.6	-	-	-	-	-	-	β -glucosidase β -galactosidase	15
<i>Piromyces communis</i>	β -xylosidase	28	0.4	-	6.0	5-8	39	to 40	None	98
<i>Sphaeromonas communis</i>	β -xylosidase	27	0.36	-	6.5	5-8	39	to 40	None	98
<i>Talaomyces emersonii</i>	Xyl I	3.0	0.13	-	2.5	-	60	-	None	117
	Xyl II	92.9	32.9	-	4.2	-	78	-	None	117
	Xyl III	0.2	1.4	-	3.5	-	67	-	None	117
<i>Trichoderma harzianum</i>	β -xylosidase	3.42	0.103	-	4-4.5	-	60	-	β -glucosidase β -galactosidase Arabinosidase	103
<i>Trichoderma reesei</i>	CBHII	0.00001	-	-	-	-	-	-	β -glucosidase Mannanase Xylanase Cellobiohydrolase	22
	EGI	0.00001	-	-	-	-	-	-	Exoglucanase β -glucosidase Mannanase Xylanase Cellobiohydrolase	22
	XYL 9	0.00002	-	-	-	-	-	-	Exoglucanase β -glucosidase Mannanase Xylanase	22

	XYL 5.5	0.00005	-	-	-	-	-	-	-	β -glucosidase Mannanase Xylanase Arabinosidase	22 118
	β -xylosidase	470	0.08	-	4	3-7	60	to 55			119
<i>Trichoderma viride</i>	β -xylosidase	10.8	-	-	3.5	4-5	55	to 65	None		120
<i>Bacillus halodurans</i> C-125	BH1068	-	30.1	-	6.5	5-7	47	to 45	None		121
<i>Bacillus stearothermophilus</i>	β -xylosidase	34.2	0.0012	-	6	6-8	70	to 60	None		122
<i>Bacillus thermantarcticus</i>	β -xylosidase	160	0.5	-	6.0	-	70	to 60	None		123
<i>Butyrivibrio fibrisolvens</i>	xylB	8.9	-	-	-	-	-	-	Arabinosidase		124
<i>Caulobacter crescentus</i>	CcXynB2	215	9.3	402	6	4.5-7.5	55	to 50	None		62
<i>Clostridium stercorarium</i>	β -glucosidase	5.7	-	-	5.5	5-7	65	to 75	β -glucosidase Cellobiohydrolase Exoglucanase Arabinosidase		125
	xylA	3.5	2.5	5.9	7	5-10	65	-			126,127
<i>Geobacillus stearothermophilus</i>	β -xylosidase	133	2.38	147	6.5	5.5-7	70	50-70	None		43
<i>Ruminococcus flavefaciens</i>	Exo A	0.01	-	-	5	4.5-5.5	-	-	Cellobiohydrolase Exoglucanase		128
<i>Thermoanaerobacterium saccharolyticum</i>	β -xylosidase	53.8	28	276	6	6-6.5	65	to 67	None		129
<i>Thermoanaerobacter</i> sp.	β -xylosidase	66	0.038	183	5.9	5-8.5	93	to 86	Arabinosidase		130
<i>Hordeum vulgare</i>	β -xylosidase	6.06	-	-	-	-	-	-	Arabinosidase		

Table S7. Properties of enzymes with xylanase activity. Table is organized by origin, fungal, then bacterial, then plant. All values listed are from enzymatic activities against xylan. “-”, not reported in the study. Rows of anaerobic fungi are highlighted.

Origin	Protein Name (Annotation)	Specific Activity (U/mg)	Kinetics Km (mg/mL)	Kinetics Vmax (U/mg)	Optimal pH	pH Stability	Optimal Temp. (°C)	Temp. Stability (°C)	Other Activities	Ref.
<i>Orpinomyces</i> sp. strain C1A	XYL11	73.5	0.024	370	5	3-9	50	4-60	β-glucosidase	This Study
	EG5	9.37	0.035	36.5	7	3-8	50	4-50	Endoglucanase	This Study
	Bgxl	10.8	0.038	25.6	6	4-12	39	4-70	β-glucosidase β-glucosidase β-xylosidase β-galactosidase	91
<i>Acrophialophora nainiana</i>	Xyn III	31.25	4.37	0.24	6.5	-	50	to 55	None	131
	Xyn II	33.2	40.9	-	7	-	55	to 55	None	132
	Xyn I	13.6	0.731	-	6	-	50-60	to 50	None	133
<i>Aspergillus awamori</i>	β-xylosidase	1.7	-	-	6.5	-	70	to 70	β-xylosidase	104
<i>Aspergillus niger</i>	Xylanase I	18.9	-	-	6-6.5	-	65-80	to 80	Exoglucanase	93
	Xylanase IA	35.2	-	-	5.5-6	-	65-80	to 80	Exoglucanase	93
	Xylanase II	24.5	-	-	4-4.5	-	65-80	to 80	None	93
	Xylanase IIA	48.0	-	-	4	-	65-80	to 80	None	93
<i>Chaetomonium cellulolyticum</i>	Xylanase 1	22	-	-	6-7	-	50	-	Endoglucanase	47
	Xylanase 2	18	-	-	6-7	-	50	-	Exoglucanase	47
	Xylanase 3	11	-	-	6-7	-	50	-	Endoglucanase Arabinosidase	47
<i>Chrysosporium lucknowense</i>	EG28	0.2	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase	2
	EG44	0.07	-	-	4.5-6	5-7	60-70	60-75	Exoglucanase	2
	EG47	0.08	-	-	4.5-6	5-8.5	60-70	60-75	Endoglucanase	2
	EG51	0.18	-	-	4.5-6	5-7	60-70	60-75	Exoglucanase	2
	CBHII	1.4	-	-	4.5-6	5-7	60-70	60-75	Endoglucanase Cellobiohydrolase	2
<i>Fusarium oxysporum</i>	Xylanase	5.2	3.8	-	7.4	5.8-8.2	50	to 50	Endoglucanase	3
<i>Gloephyllum trabeum</i>	Cel5A	0.77	-	-	-	-	-	-	Exoglucanase	5
	Xyn10A	390	-	-	-	-	-	-	Endoglucanase Exoglucanase	5

	Cell2A	0.88	-	-	-	-	-	-	Endoglucanase Exoglucanase	5
<i>Humicola grisea</i>	X1	-	7.233	-	5	-	50	to 50	None	134
	X2	10.23	10.87	-	4.5-6.5	-	55-60	to 60	None	134
	Xylanase	169.4	3.3	229	5.5	-	70	to 50	None	135
<i>Neocallimastix frontalis</i>	Xylanase	0.36	1.13	-	6	3-8	50	to 40	None	136
<i>Orpinomyces joyonii</i>	Cellulase	5.2	-	-	5.5	5-7.5	40	to 50	Exoglucanase Endoglucanase Cellobiohydrolase	49
<i>Paecilomyces thermophila</i>	Xylanase	2063	2	2344	7	6-10	75-80	to 75	None	137
<i>Penicillium brasilianum</i>	XYL	279	-	-	-	-	-	-	None	13
<i>Penicillium citrinum</i>	Xylanase	360.7	-	-	8.5	4-10	50	0-60	None	138
<i>Piromyces communis</i>	Xylanase	0.37	1	-	5.5	-	50	-	None	136
<i>Sphaeromonas communis</i>	Xylanase	0.43	0.98	-	6	-	50	-	None	136
<i>Trichoderma harzianum</i>	XYL2	-	13.66	-	5	3.5-6.5	45	to 50	None	139
	Xylanase	2400	-	-	-	-	-	-	None	140
<i>Trichoderma reesei</i>	EG I	0.55	-	-	-	-	-	-	Endoglucanase Cellobiohydrolase	60
	EG II	0.003	-	-	-	-	-	-	β -glucosidase Endoglucanase Cellobiohydrolase	60
	CBH I	0.0003	-	-	-	-	-	-	β -glucosidase Endoglucanase Cellobiohydrolase	60
	CBH II	0.002	-	-	-	-	-	-	β -glucosidase Endoglucanase Cellobiohydrolase	60
	CBH III	0.0002	-	-	-	-	-	-	β -glucosidase Endoglucanase β -xylosidase	22
	EG I	0.29	-	-	-	-	-	-	Endoglucanase β -glucosidase β -xylosidase	22
	XYL 9	11.33	-	-	-	-	-	-	Endoglucanase β -glucosidase β -xylosidase	22
	XYL 5.5	3.855	-	-	-	-	-	-	Endoglucanase β -glucosidase β -xylosidase	22
	EG I	46.2	-	-	4	2-8	60	to 45	Endoglucanase Exoglucanase	23

<i>Anaerocellum thermophilum</i>	CelA	0.372	-	-	5-6	-	85-95	-	Endoglucanase Exoglucanase Cellobiohydrolase	26
<i>Bacillus amyoliquefaciens</i>	Cellulase	22.5	-	-	7	4-9	50	40-80	Endoglucanase Exoglucanase Cellobiohydrolase β -glucosidase	27
<i>Bacillus circulans</i>	XylA	2039.5	4	2667	6-6.5	4-8	75-80	to 65	None	141
	XylB	6423	25	200000	6-6.5	4-8	65-70	to 65	None	141
<i>Bacillus licheniformis</i>	Xylanase	28.7	3.33	111	6-7.5	5-8	60	to 60	None	142
<i>Bacillus stearothermophilus</i>	Xylanase	122	3.8	-	7	5-11	60	to 60	None	121
<i>Bacillus subtilis</i>	Cellulase	980	-	-	6	6-7.5	50-60	to 50	Endoglucanase	32
<i>Bacillus thermantarcticus</i>	Xylanase	141	1.6	-	5.6	-	80	to 60	None	122
<i>Caldibacillus cellulovorans</i>	CMCase	4.56	-	-	6.5-7	-	80	to 70	Endoglucanase Exoglucanase	33
<i>Cellulomonas fimi</i>	Exg	37	-	-	6	5-9	-	60-75	Endoglucanase Cellobiohydrolase	61
<i>Clostridium stercorarium</i>	Avicelase II	0.340	-	-	5-6	4.5-7	75	65-80	Exoglucanase	38
<i>Streptomyces lividans</i>	Xylanase	364	0.78	850	6	5.5-7	60	0-37	None	143
<i>Streptomyces rameus</i>	Xylanase	4326	5.8	-	5.3	4.3-6.7	70	40-65	None	144
<i>Streptomyces thermoviolaceus</i>	STX-I	1460	-	-	7	5-9	70	to 50	None	145
	STX-II	1405	-	-	7	5-9	60	to 60	None	145
<i>Thermatoga maritima</i>	CelA	2.5	-	-	5	4-9	85-90	to 95	Endoglucanase Exoglucanase Cellobiohydrolase	65
	CelB	0.008	-	-	7	5-9	85-90	to 95	Endoglucanase Exoglucanase Cellobiohydrolase	65
<i>Thermatoga neapolitana</i>	CelB	37	-	-	6-6.6	-	106	to 106	Endoglucanase Exoglucanase Cellobiohydrolase	21

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