

AMB Express

Supplemental information for

**Characterization of FsXEG12A from the cellulose-degrading ectosymbiotic fungus
Fusarium spp. strain EI cultured by the ambrosia beetle**

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Table S1. Identification of the cellulolytic enzyme produced by *Fusarium* spp. strain EI

Annotation name	Gene ID ^a	PMF search		MS/MS ion search	
		Score	MW ^b	Cov. ^c	Identified peptide sequence
Hypothetical protein	EMT67806.1	127	20.9	54	GSYTVSGLGQR, TYDAANFGLFK

^aProtein name and accession numbers were obtained from the NCBI protein database.

^bTheoretical mass.

^cSequence coverage (%) in peptide mass fingerprinting (PMF).

Table S2. Substrate specificity of the purified GH12 enzyme

Substrate	Specific activity (U/mL)	Relative activity (%)
CMC	158.9 ± 10.7	59.2
MCC	57.3 ± 9.4	21.3
Xyloglucan	268.6 ± 50.1	100
Lichenan	263.7 ± 37.5	98.2
Glucomannan	121.3 ± 14.4	45.2

Data are means ± standard deviation of three experiments. Recombinant GH12 enzyme (1.0 μM) was incubated with substrates in 50 mM acetate buffer (pH 3.0) at 37°C.

Figure S1. Nucleotide sequence alignment of *18S rRNA*.

18S rRNA sequences of *Fusarium* spp. were aligned using ClustalW. Identical nucleotides are highlighted. Dashes indicate gaps. *18S rRNA* sequences of LC534254 from *Fusarium* spp. strain EI, LT841222.1 from *F. oxysporum* f. sp. *dianthi* strain Fod001, LT841236.1 from *F. oxysporum* f. sp. *dianthi* strain Fod008, LT841208.1 from *F. oxysporum* f. sp. *cumini* strain F11, LT841250.1 from *F. proliferatum* strain ITEM2287, LT841264.1 from *F. proliferatum* strain ITEM2400, KC866343.1 from *F. sporotrichioides* isolate CHILEFUS1, KC866344.1 from *F. sporotrichioides* isolate CHILEFUS2, KP310124.1 from *F. solani* strain XXG-2, EF397944.1 from *F. solani* strain 421502, KX132128.1 from *F. graminearum* strain CBE330.6, and KX132126.1 from *F. graminearum* strain CBE330.5 are shown.

<i>F. oxysporum</i> f. sp. <i>dianthi</i> strain Fod008	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	279
<i>F. oxysporum</i> f. sp. <i>dianthi</i> strain Fod001	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	279
<i>Fusarium</i> spp. strain E1	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	258
<i>F. oxysporum</i> f. sp. <i>cuminii</i> strain F11	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	279
<i>F. proliferatum</i> strain ITEM2287	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	279
<i>F. proliferatum</i> strain ITEM2400	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	279
<i>F. sporotrichioides</i> isolate CHLEFUS1	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	276
<i>F. sporotrichioides</i> isolate CHLEFUS2	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	276
<i>F. solani</i> _strain_XG-2	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	276
<i>F. solani</i> _strain_4215032	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	276
<i>F. graminearum</i> _strain_CBE330.6	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	266
<i>F. graminearum</i> _strain_CBE330.5	T	G	G	T	T	C	A	T	T	C	A	A	A	T	T	T	T	C	T	T	C	G	C	T	A	T	C	A	A	C	266

Figure S2. Nucleotide sequence alignment of *EF-1α*.

The nucleotide sequence alignment was performed using partial *EF-1α* sequences of *Fusarium* spp. strain EI, *Fusarium* species registered in the JGI database (<https://mycocosm.jgi.doe.gov/Fusarium/Fusarium.info.html>) and the *Fusarium* sp. AF-3 sequence. The *EF-1α* sequences of *Fusarium* spp. were aligned using ClustalW. Identical nucleotides are highlighted. Dashes indicate gaps. *EF-1α* sequences of KC691533.1 from *Fusarium* sp. AF-3, KC691536.1 from *Fusarium* sp. AF-3, 599354 from *F. commune* MPI-SDFR-AT-0072, 8475 from *F. fujikuroi* IMI 58289, 9400 from *F. graminearum* FGSG_08811T0, 17733 from *F. oxysporum* f. sp. *lycopersici* 4287, 554906 from *F. oxysporum* MPI-SDFR-AT-0094, 4767 from *F. pseudograminearum* CS3096, 485664 from *F. solani* FSSC 5, 10379 from *F. verticillioides* 7600, and LC534255 from *Fusarium* spp. strain EI are shown.

[illegible]

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Accession	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120	121	122	123	124	125	126	127	128	129	130	131	132	133	134	135	136	137	138	139	140	141	142	143	144	145	146	147	148	149	150	151	152	153	154	155	156	157	158	159	160	161	162	163	164	165	166	167	168	169	170	171	172	173	174	175	176	177	178	179	180	181	182	183	184	185	186	187	188	189	190	191	192	193	194	195	196	197	198	199	200	201	202	203	204	205	206	207	208	209	210	211	212	213	214	215	216	217	218	219	220	221	222	223	224	225	226	227	228	229	230	231	232	233	234	235	236	237	238	239	240	241	242	243	244	245	246	247	248	249	250	251	252	253	254	255	256	257	258	259	260	261	262	263	264	265	266	267	268	269	270	271	272	273	274	275	276	277	278	279	280	281	282	283	284	285	286	287	288	289	290	291	292	293	294	295	296	297	298	299	300	301	302	303	304	305	306	307	308	309	310	311	312	313	314	315	316	317	318	319	320	321	322	323	324	325	326	327	328	329	330	331	332	333	334	335	336	337	338	339	340	341	342	343	344	345	346	347	348	349	350	351	352	353	354	355	356	357	358	359	360	361	362	363	364	365	366	367	368	369	370	371	372	373	374	375	376	377	378	379	380	381	382	383	384	385	386	387	388	389	390	391	392	393	394	395	396	397	398	399	400	401	402	403	404	405	406	407	408	409	410	411	412	413	414	415	416	417	418	419	420	421	422	423	424	425	426	427	428	429	430	431	432	433	434	435	436	437	438	439	440	441	442	443	444	445	446	447	448	449	450	451	452	453	454	455	456	457	458	459	460	461	462	463	464	465	466	467	468	469	470	471	472	473	474	475	476	477	478	479	480	481	482	483	484	485	486	487	488	489	490	491	492	493	494	495	496	497	498	499	500	501	502	503	504	505	506	507	508	509	510	511	512	513	514	515	516	517	518	519	520	521	522	523
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Figure S3. Amino acid sequence alignment of GH12 enzymes.

Amino acid sequences of GH12 members were aligned using ClustalW. Identical amino acids are highlighted. Dots and colons indicate conserved amino acids with substitutions. Dashes indicate gaps. The 25-kDa protein (LC634256) from *Fusarium* spp. strain EI, CVL13720.1 from *F. proliferatum*, XP_018760412.1 from *F. verticillioides* 7600, XP_011325323.1 from *F. graminearum* PH-1, OXN20159.1 from *Aspergillus fumigatus*, and GAM41656.1 from *Talaromyces cellulolyticus* are shown. The putative active site is highlighted in yellow.

