

Fig. S1. Phenotypes and genetic information of site-mutants and knockout lines of OsCESA4,7,9 isoforms. (a) Rice phenotypes at the mature stage. (b) Genetic information of mutation sites in coding sequence and corresponding protein.

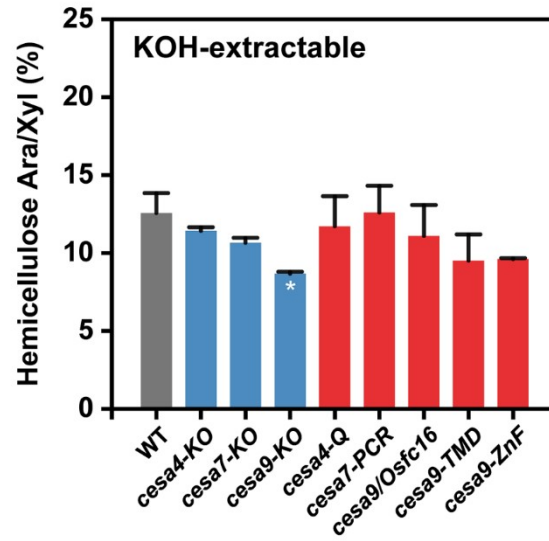


Fig. S2. The ratio of arabinose to xylose of KOH-extractable hemicelluloses of site-mutants and knockout lines of OsCESA4,7,9 isoforms. Data as means \pm SD ($n = 3$); Significant differences between the WT and mutant were determined using two-tailed Student's t-test: $**P < 0.01$, $*P < 0.05$.

Table S1. Primers used for CRISPR-Cas9 editing of OsCESA4, 7, 9 isoforms.

Mutant	Vector	sgRNA oligonucleotides	Mutation screening
<i>cesa4-KO</i>	pCSGAPO1	TGTGTGTGCGCGTCGTCCCCGCACG AAACCGTGCGGGGACGACGCGCACA	GTTCCATGTCCGCTCAACCTT TGCTTACCACTCCCAACTCC
<i>cesa7-KO</i>	pCSGAPO1	TGTGTGCCACCTCCTCGCCGCACACC AAACGGTGTGCGGCGAGGAGGTGGCA	CCTGCATCTGATCATCAACCAGC CTTGGTGGGGCTCTTGATCTG
<i>cesa9-KO</i>	pRGEB32	GGCATGAACTTCGAGAAGCGGTTTC AAACGAACCGCTTCTCGAAGTTCA	CGCTGTACGGATAACAACCCAC GACCCGTAGATCCACCCAG
<i>cesa4-Q</i>	pCSGAPO1	TGTGTGAGCCCACCGGAGAACCTGG AAACCCAGGTTCTCCGGTGGGCTCA	TCCGCAAACCCATCTACT CACCGATGACCCAGAACTGC
<i>cesa7-PCR</i>	pRGEB32	GGCATGCATCACCCACCCTTCCTC AAACGAGGAAGGGTGGGTGATGCA	TTGGAGCATCTCGTGATCAGT ATCGTTAGCTCACGTCACGG
<i>cesa9-TMD</i>	pRGEB32	TAGGTCTCCAGCCGTTGTTGAGTTTTAGAGCTAGAA (target 1) ATGGTCTCAGGCTCCGAGGCGTGACCAGCCGGGAA (target 1) TAGGTCTCCCATCAAGGCCAGTTTTAGAGCTAGAA (target 2) ATGGTCTCAATGGTGAAGGGATGCACCAGCCGGGAA (target 2)	CGGGATCGACACCAACTTCA ATGGCCATCTGTCCATTCCC
<i>cesa9-ZnF</i>	pCSGAPO1	TGTGTGGCGCTCGTACTCGTAGCAG AAACCTGCTACGAGTACGAGCGCCA	CCATATAGCCCCTGCCACAA CCTCCCGTAGCTCATCTTGC

Table S2. Primers used for detection of off-target mutagenesis in *CESA*-edited plants.

Sample	Target site	Potential off-target site	Location	PCR primer
<i>cesa4-KO</i>	GTGCGCGTCGTCCCCGCACGCGG	CTGGGCGTGGTGCCCCGCACGAGG	Chr6:+22001840	CGCGGATGGAGATGATGTGA TCCGAGGTAGGCCATTCTGT
		GTGCGCGCCGTCCCAGCACACGG	Chr6:-1573708	TGATGATGGACGGACAGACA GACTACGACTACGGCTG
		GCGCGCGACGTGCCGGCACGCGG	Chr9:-22046877	GAACACGCGGAAAATGCTGT CACTCATCTGTGCTGGGTCC
		GGTGGCGTCGTCCGCGCACGGGG	Chr1:-37789455	ATGAAAGGCTCACCAGGTCTG CCCATGTCCATCACGGTCAA
		CAGCGCGTCGTCGCCGTACGGGG	Chr5:+29485019	AAGAAGGCAATGGAGCGGAA GCCACCAGGGTCGAATTGAT
		GGGCGGCTCGTCCCCGCAGGAGG	Chr7:+7757629	CGGCGTGGCTTCACCTTC CGCTTCGCTCCTTCTCT
		GTGCGCCTCGTCCTCGCACTAGG	Chr11:+12133769	ACAAGCTGAAGTACACGT AGGCAATCACAGACAATC
<i>cesa7-KO</i>	CCACCTCCTCGCCGCACACCCGG	CTGCGTCCTCACCGCACACCTGG	Chr8:-23134438	GCCGATACCAAGGAGGTACG AAAAGTCGTGGCCTCATGCT
		CGGCCTCCTCGCCGCCACCCGG	Chr2:-27804497	GAGCCACCTGCAGAACATCT AGTTCAAAAATGTCCGCGCC

		CGGCCTCCTCGCCGCCACCCGG	Chr4:-29195964	CTTCTTCCCGGACAACCTCC GCGACATTACTCCCCTGGAC
		CCGCATCCTCGCCGCGCACCCGG	Chr4:-7256358	TGATGACGCACATCTACGGG GGATCTCGCCACGATGAACT
		CCGACGGCTCGCCGCACACCAGG	Chr9:-21819639	CGTTCCTCCAGGTGTTCCCTC AACTTGTCCGGCGACTTCAT
		CCACCTCGGCGCCGTACACCAGG	Chr1:-16285405	GGTGAACGCTTGAAACTC TCATTACGGACAACGGCA
<i>cesa9-KO</i>	TGAACTTCGAGAAGCGGTTCCGG	TGAAATTGGAGAAACGGTTCAGG	Chr11:+28132545	ACGCTCCTAGCTACCACGTA CTGGGACAGGGCAGAGAAAG
		TGAGCTTCGCGCAGCGGTTTCGG	Chr3:+3655081	GGTGCCTGTAGTCGGAGATG CCGTGTCTCCTGTGGTCAA
		TGGACTGGGAGAAGCGGTACCGG	Chr1:+38116186	TGTGGCCGTGAAGCAACTAT CATCCGCGAGATGGTTGAGA
		AGAACTTCGAGCAGCGGTGGAGG	Chr2:+1064032	ATGTACACGCAGCACCAGAA CAGAGTCCACTGACCGGAAC
		AGAACTTCGACGAGCGGCTCCGG	Chr8:+25494609	GCTCAACTGCATCCTCGACT GACATACCGACGAGAGGCAC
		AGAACGGCGAGAAGCGGCTCCGG	Chr2:+13369089	GCGGGTGGTGATACCAAGAT TAGTTGGACAGCTTGCGGAG

		TGCACTTGGAGCAGCTGTTCAGG	Chr5:+13625425	GAGGACCCCTGTGGACAATG AGGCCTTTACCCATCGCATC
<i>cesa4_Q</i>	GAGCCCACCGGAGAACCTGGTGG	GACCCCCTGGGAGAACCTGGGGG	Chr10:-4568506	TTCCACACCGAGAACGAC AGCAAAACCCAAAAACCT
		TCGCCCACCGGAGCACCTGGTGG	Chr12:+22604011	AGGGTGATGACGAGGAGGAA GGCGTGATCTCCTGCTTCTA
		GAGACCACCGGAGAACTTCGAGG	Chr12:-8036153	GAGCTGAACGTTCTCCACC CAAGAGGCAGAAAACACGGC
		TGGCCCATCGGAGGACCTGGTGG	Chr10:-23065051	TGAGGAAGGTGACGTTGAGC CGAGCTTCTGATCGACTCC
		CCGCCCAGCGGAGCACCTGGTGG	Chr10:+10181413	TTCCGCCACCACTCACCC CGCCGTTCTAGCCTCCG
		TAGCCATCCGGACAACCTGGAGG	Chr11:-17513749	TGAAAGGTACAAGCCCTCACA TGCTTTGTGCAGGCAACGAT
<i>cesa7_{P-CR}</i>	TGCATCACCCACCCTTCCTCCGG	TGCATAATCCAGCCTTCCTCAGG	Chr1:-31425473	TCTCTACTCCAAGTGGCAACC TCACTTGCGGAAACAGCAGA
		TCCTGGACCCACCCTTCCTCCGG	Chr3:+3549793	CCACATCATTCAACCGGGGA CCGCCGAATGAGATCAAGA
		AGCAGCACCGGCCCTTCCTCCGG	Chr1:+14745236	CCCAGAGGTTGAGGAAGTCG AGGTTCACTTCTGGATTTCTGA

		AGCTTCATCCACCCCTCCTCCGG	Chr6:-28488169	TCAGGTCCTCCTCCGACTG AACCTTGCCCTATTGGTGGG
		CTCATCCCCACCCCTTCCACCGG	Chr3:-4475835	AGCCGTTTCGATTAACGTGGA CTCGCCGTCCATACGATCAA
		AGCTTCATCCACCCCTCCTCCGG	Chr1:+3392297	CGATCTCCCTGCACAGTCTC GAGGAACAAGTCCGGGGAAG
		TTCATGATCCACCCCTCCGCCGG	Chr9:-15283289	AGATGAAGGATGTGGTGGCG AGAAGTTCAGCATCGAGCCC
		TGCATGAATCTCCCTTCTCCGG	Chr5:+22744868	GAACCGACCCATGAACCCAT GGGTGGGGCTCAACAAGTA
<i>cesa9_{TMD}</i>	CGCCTCGGAGCCGTTGTTGATGG (target 1)	CAGCTCGAACCCGTTGTTGATGG	Chr3:+117633	GTGCCTGATCGTCTTGGACA GCTCATCCTCAACAGGGTCC
		TGCCTCGGTGTCGTTGTTGACGG	Chr6:+13955149	TTGTTAGAGTTTGTGTGCG TAATCCTAGTGCTAGTGG
		AGCCCCCGAGCTGTTGTTGAGGG	Chr4:-29228645	GTGACCCACAGATCCCTT TTCCGAACTAGTCGCAAA
		AGCCCCCGAGCTGTTGTTGAGGG	Chr12:-3048984	TGCATTTTAGGAGTACCC TATCTCGAGAAGTCAGG
		AGCCCCCGAGCTGTTGTTGAGGG	Chr3:-27750400	CGCATTTTAGGAGTACCC TATCTCGAGAAGTCAGG

		AGCCCCCGAGCTGTTGTTGAGGG	Chr3:+13208577	AGTCTTTGAGGGCGGTTG CTGGGCGTGACATTATCC
		AGCCCCCGAGCTGTTGTTGAGGG	Chr5:+24405896	GGCCAGAAGAACCCTTAT TAGCCCCGAGTTATCCAA
		AGCCCCCGAGCTGTTGTTGAGGG	Chr8:+21369606	GTCTTTGAGGGCGGTTGG CTCGGAGTCAGGTGGCTG
<i>cesa9_{TMD}</i>	TCCCTTCACCATCAAGGCCAGGG (target 2)	ACCCCTCAACATCAAGGCCGGG	Chr3:+2406173	GCCTCCATACATGCCCTTCA CGTGGAGCTGTTTGCTCCTA
		TCCATTCACCATGAAGGCAAAGG	Chr7:-17290256	ACCTCGCTCTCCTCCTCTTT TCTCCGGTGATCGATCTGGT
		TTGCTTCACAATCACGGCCAAGG	Chr5:+20203894	CCGATTTTCAACTGCGGACC TGTGTTCTTGGAGACCGTCG
		TCCCTTCACTAAAAAGGCAAGGG	Chr1:+38228757	ACGCTAGCCATTCCAAGACC GCAGCATTGTGAGCTTTGCT
		TCCCGTCACCATGATGGCAACGG	Chr8:+15382715	ATGGTGAATGAGGTGGGCAG TCGTCGACAGAAGCTCCCTA
		TGCCATCACCATCTAGTCCAAGG	Chr6:+9292546	GGTGTGAATACTGAATACATCTTGC GTTCAGCACTTGGACCGGA
		TCCATTCACCAGGAAGGCAAAGG	Chr1:+15180274	GAACCTGCCAGTGTCAAAGC TAGTTGGTCGGTTGGTGTGC

<i>cesa9</i> _{ZnF}	GGCGCTCGTACTCGTAGCAGGGG	TGCGCTCGTACTCGTAGCAGGGG	Chr7:+5857909	GCAAAGCCTTAGCATGACTGT GGTGAGCTGAGAGCTGGTTT
		TGCGCTCGTACTCGTAGCAAGGG	Chr5:+4570390	GGCGGCAAAACAATCACAGT CTGATGACGCAGAGGCTGAA
		CGCTCTCGTACTCGTAGTAGCGG	Chr11:+3686778	ATGTCATGTCGCTCGTCCTC AACGACCTTGTGGTGTGAA
		GGAGCTCGTTTTCGAAGCAGGGG	Chr6:+18109646	CGCTGTTTCCTGTTTGTC GGTCCTTCTCTCGCTGAT
		GGAGCTCGTTTTCGAAGCAGGGG	Chr7:+15104834	AGGATATGTTGAGAGCCT CCTACTGAACTTTGTAGAA
		GGAGCTCGTTTTCGAAGCAGGGG	Chr4:+10039708	ATGTGCTTGACTTTGGTG GGTCCTTATCTCGTTGGT

Table S3. LC-MS/MS identification of *T. reesei*-secreted enzymes after incubation with nanofibers substrate.

Protein Name	Accession No.	MW [kDa]	WT-CNF	<i>cesa9-TMD</i>-CNF
Cellobiohydrolase I	A0A024RXP8.1	54.1	√	√
Cellobiohydrolase II	A0A024SH76.1	49.6	√	√
Endo- β -1,4-glucanase I	A0A024SNB7.1	48.2	√	√
Endo- β -1,4-glucanase II	A0A024SH20.1	44.1	√	√
Endo- β -1,4-glucanase IV	ETS06300.1	35.5	√	√
Endo- β -1,4-glucanase VII	A0A024SFJ2	26.8	√	√
Endo- β -1,4-glucanase	A0A024SNE0	79.8	√	√
Endo-1,3- α -glucosidase	A0A024SDT8	46.6	√	√
β -glucosidase	A0A024SB94	84.7	√	√
β -glucosidase	A0A024SD46	93.7	√	√
Endo-1,4- β -xylanase I	P36218.1	24.6	√	√
Endo-1,4- β -xylanase II	P36217	24.1	√	√
Endo-1,4- β -xylanase III	A0A024SIB3	38.1	√	√

