

# New Phytologist Supporting Information

Article title: Deceptive *Ceropegia sandersonii* uses an arabinogalactan for trapping its fly pollinators Authors: Philipp Feichtlbauer, Mario Schubert, Caroline Mortier, Christof Regl, Peter Lackner, Peter Briza, Klaus Herburger, Ulrich Meve, John Dunlop, Michaela Eder, Stefan Dötterl, Raimund Tenhaken Article acceptance date: 26 March 2025

The following Supporting Information is available for this article:

**Supplementary Figures** 

Supplementary Tables

Supplementary Methods



**Fig. S1** <sup>1</sup>H 1D spectra of the hydrolysate (bottom) of the *Ceropegia sandersonii* droplets in comparison with spectra of reference monosaccharides. Matching signals of monosaccharides are color-coded.



Fig. S2 Symbol presentation of oligo- and polysaccharides used as reference for comparison of NMR data. The building blocks of relevance are highlighted by a red box. (Fugedi, 1987; Menestrina *et al.*, 1998; Delgobo *et al.*, 1999; Tan *et al.*, 2010; Endo *et al.*, 2013; Cartmell *et al.*, 2018; Tan *et al.*, 2023)



Other observed side chains:



Enzymatically released glycan:



**Fig. S3** Chemical structures of the *Ceropegia sandersonii* polysaccharide and the released glycan. The side chains are shown in magenta, their order is tentatively. The different building blocks are marked with letters a to g corresponding to the observed NMR spin systems.



Side chains:



Enzymatically released glycan:



**Fig. S4** Observed NOE cross peaks connecting the building blocks indicated with red arrows on the chemical structure of the *Ceropegia sandersonii* polysaccharide and the released glycan. The side chains are shown in magenta, their order is tentatively. The different building blocks are marked with letters a to g corresponding to the observed NMR spin systems.



Side chains:





Enzymatically released glycan:



**Fig. S5** Observed long range <sup>1</sup>H-<sup>13</sup>C correlations connecting the building blocks indicated with cyan arrows on the chemical structure of the *Ceropegia sandersonii* polysaccharide and the released glycan. The side chains are shown in magenta, their order is tentatively. The different building blocks are marked with letters a to g corresponding to the observed NMR spin systems

TRINITY\_DN520\_c0\_g2\_i1 4 (100% relative abundance, 10 peptides, 10 unique peptides) homolog to AtPLAT2 (Q9SIE7), identities: 55/160 (34%)

IRFSSFLLLL LATFTPSIVA QTSVEASQTL DGAKICMYTL YVKTGDVQNA GTNSTIR**MDF** TDSTGWTIRI SNIAAIGLMG AGHNYFQRGQ VDIFGFGIGC MRTPICRVAI SSDGTGKNPA WFLDTIQISY TGIGRRCHET VFGIYRWISD SNGLNFVLDN CGIPPFPPVR PFPPPPAPF PYHVKNEKVK ELVAEKIMEE EEVAPTSVEV ASVVLKPRMS FTEEN

TRINITY\_DN1987\_c0\_g1\_i2 2 (81% relative abundance, 7 peptides, 7 unique peptides) homolog to LTP2 (Q43194), identities: 52/98 (53%) NNMQRSPLPI STNKYNFNLQ SIYNTKNPIL TMESVRKMAG AAVIMIIINS VCMMMFSSSE AAITCSQVNS NIGPCIKYIV NKAILNPSDS CCKGVKTLNS AATSTPDLRI ACKCLKDIAR KFPGIDMKKA GAIPGKCGVK LPFQISLSTD CNKVQ

TRINITY\_DN6779\_c0\_g2\_i1 6 (63% relative abundance, 11 peptides, 11 unique peptides) homolog of endochitinase (P85084), identities: 137/244 (56%)

EIRNMIMKYY YMILIILSC FFTAFSSSSS ILADDQDYSN NNITISSVIN STIFNQMLGH SMDSRCPSNG FYTYDAFISA ATSFPAFGTT GDNDTQKREI AAFLGQTSHE TTGGWPNAPD GPYAWGYCII IKNKIQNHTF CNSRRVKCPP GKSYYMR**GPI QLIGSDNYYR** VGEEIGEDLL NNPDLVETNA TISFKTAIWF WMTDYFSTPS CHDVIVGKWK PSADDKRYGR LPGYGTITNI FNGESECGRG GVITPAENDR IQFYKRYCNM FGIYCGGPLD CRYQDPFGIR ILN

TRINITY\_DN1915\_c0\_g1\_i8 6 (62% relative abundance, 18 peptides, 18 unique peptides) homolog of aspartyl protease family protein (Q8S9J6), identities: 238/424 (56%)

IGPGRFGEGD	LNSYSSSSFL	IMANSNNYFF	FFFNFLHPIS	LLVFLLSFSN	LKGGGCAMLE
KTHEKVIAEA	HYHTVHVSSL	LHIQSSCKTS	S <mark>P</mark> HKGHNTKS	ASLEVIHKHS	ACKKDTIKEP
TLVEILAADE	SRVNSIHGRR	FLNSGLINNK	YRNSKANLPA	KPGVTIGSGN	YIVNVGLGTP
ARTLTLVFDT	GSDLTWTQCQ	PCARSCYDQQ	DPIFDPSKST	SYSNITCNST	QCSALPSATG
NKPGCIASTS	TCVYGIQYGD	QSFSVGYFSK	DKITLTQVDS	FDNFLFGCGQ	NNQGLFGKTA
GLLGLGRDPL	SIVSQTSQKY	GKFFSYCLPN	KSGGSKGHLT	FGKKNLPTNI	VYTPLKNTNQ
GTFYFVDIIG	INVGGQLLSI	NQSIFKNAGT	IVDSGTVITR	LPQTGYDALK	TAVKKQMSQY
PTAASVSILD	TCFDLSNYTS	VTVPKLVFIF	GGNSKVELPV	LNILVAAKQS	ILCLAFAGNG
DDSSVGIYGN	IQQQTFEVVY	DVAGGKLGFG	PSGCS		

TRINITY\_DN520\_c0\_g1\_i3 5 (40% relative abundance, 5 peptides, 5 unique peptides)homolog to AtPLAT2 (Q9SIE7), identities: 60/164 (37%)REAKKREKRK KMAIIRFSSF LLLLATFTT SIVAQNSVAA LKVDGAKICL YTIYVKTGDIEMAGTDSKIS MNFTDYTGSKLIVENLTSIG LMGEGHDYFK RGKVDIFGVG TQCLLTPICSVEVALSSNGN GILPDWYLETIQISFTGIDG GCRQTVFIIN EWLTTSDILS YEANECSIRPFPSPFHGSIN EEGHEELIVA KKLMLKEKVA QPSVVAAAGE LKPLRSVNHN SFSQ

TRINITY\_DN4435\_c33\_g1\_i1 4 (38% relative abundance, 1 peptide, 1 unique peptide) homolog to LPT2 (P82353), identities: 29/57 (51%) ITAFQSGAPQ SSGCCVSLKS QQPCLFQYAK NPALR**QYITN PNAK**KVAQSC MVPLPKC

**Fig. S6** Sequences of most abundant proteins identified by proteomics using all potential reading frames of the transcriptome of *C. sandersonii*. Prolines in sequences predicted to be hydroxylated are indicated in magenta, otherwise grey. Clusters of such hydroxyprolines are typically found in arabinogalactan proteins, typically in unstructured regions or tails. Sequence regions covered by a peptide found in the MS analysis are underlined. The most abundant peptide is marked in bold letters. The corresponding relative abundance as well as the number of matched peptides and uniquely matched peptides is denoted in the parentheses after the Trinity accession code.

TRINITY\_DN3751\_c6\_g1\_i3 5 (36% relative abundance, 6 peptides, 6 unique peptides) homolog to LTP (Q39950), identities: 73/122 (60%) KKKRKKKKKE KKIKKMEMMG KKFVLFMVVA IMVALIKPGV EATITCGQVS GSLAPCIPYL TKNAPMGECC DGVQKLNSQA QTTPDRQTAC GCLKSAYK**SM TGINPALAAG LPAK**CGVSIP YKISPDTDCT KVR

TRINITY\_DN520\_c0\_g1\_i18 5 (35% relative abundance, 7 peptides, 0 unique peptides) homolog to AtPLAT1 (Q65660), identities: 54/130 (42%) KEKMAIIHFS SLLLLLATF TTSIVAQTSV EAPKVDGAKI CLYSVYVKTG DIEKAGTDSK ISMNFTDYTG RTLVVQNLR**S IGLMGNDHDY FER**GQVDLFS TGSHCVRTPI CSVSILSDGT GILPAWYLET IQISFTGIGR NCYKTVFNIN KWLSSSGSLS YVANNCRFRP FPPFSFLG SNEEEQELAA KKIMLEEEVA KPSVVAAGEL KPLRSVNHNS FSEQ

TRINITY\_DN827\_c3\_g1\_i1 1 (30% relative abundance, 1 peptide, 1 unique peptide) homolog to LTP (Q43681), identities: 37/80 (46%) LVMMIIIVLL AGDEVKVSKE QNCNPTELSS CLPAFQYGAP TSSGCCGCLK **SQQPCLCQYA K**HPGFRQFIT NPNANIVAQS CKVPFPKC

TRINITY\_DN1483\_c0\_g1\_i3 6 (26% relative abundance, 1 peptide, 1 unique peptide) homolog to E3 ubiquitin ligase UPL6 (Q8RWB8), identities: 489/691 (71%)

FFHGTŠGLHQAVDFATVATPLLGALPSITPŠNEGNKQNGIVEDDEMVYGDEHGEEVLHGDIQLQIYNAIDSRFLLQLTNVLLGGVSLPDKSCNGVPSSKDVAAVGAACAFLHVTFNILPLERIMTVLAYRTGLVPVLWNFMKRCHVNNIWSSLSRQSAYLPEGAFGWLLPLAVFCPVYKHMLMIVDNEEFYDQEKPLPLDDIRFLIVILKQALWQLMWLNPVVPPNFSKSSSSINAMKQQPLEFLQHKVCVTTSELLSQLQDWNNRQFTPPSDFHADGVDENFISQAWTENTKANATLKLAPFLLPFTSRAKIFHSQLAAVKERHAHHDAFSRIRFRVRRDHILEDAFSQLNALSEEDIRGQIRITFINEFGVEEAGIDGGGIFKDFMESVTRAAFDVQYGLFKETADHLLYPNPGSGLVHEMHHQLFYFLGTVLAKAMFEGILVDIPFATFFLSKLKQKYNYLNDLPSLDPELYRHLIFLKHYEGDISDLELYFVIVNNEYGEQTEEELLPGGKNVRVTNENVITFIHLVANHRLNAQIRQQSSHFSRGFQQLIQKEWIDVFNEHELQLLISGSVDGFDIDDLRANANYGGGYSAEHYVIQMFWEVVKHLSLENQRKFLKFVTGCSRGPLLGFKNLEPLFCIQRAAGRASEEALDRLPTAATCMNLLKLPPYKSKEQMEQKLLYAINAAAGFDLS

TRINITY\_DN3376\_c1\_g1\_i2 3 (25% relative abundance, 8 peptides, 8 unique peptides) homolog to pathogen related protein 1 (P11670), identities: 100/159 (63%)

KKKKKKKKKK KKKKEIMGST RLMAAVMITI TTTLFSLTWL PSGATAQNSN QDYLNAHNAA RAQVGVGPMI WDNQLAAYAL NYANSQKGRC PNLSHSSGPY GENLAAGTGD FTGRQAVNLW VNEKQFYDYG SNSCAAGRVC GHYTQVVWRN SVRLGCARVR <u>CSNNTWWYVI CSYDPRGNYV</u> GQRPY

# TRINITY\_DN7514\_c0\_g1\_i1 3 (25% relative abundance, 25 peptides, 25 unique peptides) homolog to subtilisin-like protease (O65351), identities: 543/761 (71%)

U	1				
SVQTIASSLS	LSLSLSLYIY	IYITAEHFQN	IS <mark>P</mark> TPFPSFS	FTSLGLNMGN	TNFHGALIAL
VLLLFVCHAS	SQAAMLKKNT	YIVHMAKSEM	PESFDDHTSW	YDSSLKSVSD	SSQMIYTYND
VIHGFAARLT	PEEARLLRKR	SGIVSVLPEM	RYELHTTRTP	SFLGLERSAS	LFPESDSASD
VVIGVLDTGV	WPESKSYDDT	GMGPIPASWK	GVCQTGTKFT	TANCNRKLIG	ARYFSDGYEA
TLGPIDVSKE	SKS <mark>P</mark> RDDDGH	GTHTSSTAGG	SLVTGASLFG	YANGTARGMA	<mark>P</mark> HARLAIYKV
CWIGGCFSSD	ILMALEMAIQ	DNVNILSLSL	GGGMSDYYRD	SVAIGAFAAM	QKGILVSCSA
GNAGPS <mark>P</mark> YSL	SNVA <mark>P</mark> WITTV	GAGTLDRDFP	AYVSLGNGKK	FAGVSLYRGP	DLTTKMFPFI
YAANASNVTN	GNLCMTGTLI	PEKVKGKIVL	CDRGVNARVQ	KGSVVK <b>AAGG</b>	AGMVLSNTAA
NGEELVADAH	<b>LLPATAVGEK</b>	SGDAIK <u>DYLI</u>	ADANPTVTIL	FEGTKVGIEP	S <mark>P</mark> VVAAFSSR
GPNSITPQIL	KPDVIA <mark>P</mark> GVN	IIAAWTGAVG	PTGLAEDGRR	VGFNIISGTS	MSCPHVSGLA
ALLKGAHPTW	S <mark>P</mark> AAIRSALM	TTAYSAYKNG	KALEDLATGQ	SSTPFDLGAG	HVDPVSALNP
GLVYDLEATD	YLNFLCALNF	TSLQIYSLAK	SNFSCDPGKK	YSIGDLNYPS	FAVLLETQTE
GGGNGGSKTG	SSVVKHSRTL	TNVGA <mark>P</mark> ATYK	VTTTVDDPSV	KISVVPETLT	FTAMNEKKSY
TVTFSTSSKP	ANTNVFGRIE	WSDGKQHVVG	S <mark>P</mark> VAVSWT		

Fig. S6 continued.

TRINITY\_DN1706\_c0\_g2\_i1 5 (14.1% relative abundance, 8 peptides, 8 unique peptides) homolog to AtPLAT2 (Q9SIE7), identities: 66/171 (39%)

LIIQKKMSER TRFFAYLLLF LTLSSVAVTA HK<u>SDCAYTIV IK</u>TADEAKAG TDAKIKITLG DFKGKSVHVV DLEK**WGIMGS DYDYYER**GST DLFSGKDNCL RQPVCRLKLE SNAKGES<mark>P</mark>GW KIDYVDVTTV IPEVGCKQAK FKVDQWLGVE GKNELSIVHE ECIDYLKNNK KKQQQSTTRS LVKGKSSNHN MV

TRINITY\_DN8840\_c0\_g1\_i1 1 (12.2% relative abundance, 6 peptides, 6 unique peptides) homolog to AtPLAT1 (Q65660), identities: 60/140 (43%) LIKEMENYRC FSLLVLLLLI AFSSATTQAY VHKCVYTFFV KTGDILSAGT DSTIGLTLSD AGGGSIDIPD LKKWGIMEWR HDYFERGNLD IFSGR**DTCLE PFICNLK**LVS NGVGTHPAWN VINVFVGISR PDGPCTEINF PVNGWLGAFG HPFVLTVNLP GCPKRSTLKN AVESSTKALD Y

TRINITY\_DN791\_c0\_g1\_i2 3 (11% relative abundance, 4 peptides, 4 unique peptides) homolog to Sneakin-2 (Q93X17), identities: 56/107 (52%) LNPTQPNPTC HHHHYYFLCL YNKDSVLIIS FISLNNKYKQ TMAFLKAAIA ALFLCLLFLH STIALQGVNI PSSSSSPSPA PLPQIKK<u>IDC GSACGVR</u>CSK TKRPNLCKR<u>A CGSCCVKCNC</u> VPEGTYGHYE TCPCYFNLTT HNNTRKCP

TRINITY\_DN2808\_c0\_g1\_i1 2 (11% relative abundance, 11 peptides, 5 unique peptides) homolog to Ervatimin-C (A8DS38), identities: 159/365 (44%)

RTELTKMNSS SNLFIIIIFL SISQTINSIN NNGESVSWQR TDNELMALFE EWMMQHQKFY SSSSSVLGEK IDRFEIFKEN LRYIDEHNNL PNTTFQLGLN QFSDLTYEEF ESMYLSRISM NKMSPINQEE LELESDDNYG GDQFNYSTLT LPNYVDWRK<u>E GAVNPIK</u>NQG YCGSCWSFTT VASVEAINKI KNGKLVSLSE QMLLDCVTSN GGCNGGTAAN SFNYMKTYGV ALNEDYPYVG FKGVCQNKPI AVKIDGFNLV LPRLELK**LLP AVANQVLTIA IK**AGSEDFRH YKSGVFSGQC GNKFSHGMNV VGYSFKNGKL YWILRNSWGE TWGEKGYMRI AGLVSGYERG YCGIAEEATY PIMSTSVNQE LLSVV

TRINITY\_DN4498\_c1\_g1\_i2 1 (10% relative abundance, 5 peptides, 5 unique peptides) homolog to PsLTP1 (A0A161AT60), identities: 61/113 (54%) TMYTFKNMMI MIMSVMMVLC MVMFSSEYSG AEAAVTCGLV SSSVAPCMSY LQNK**AATIPS** TGCCSGIKSL SSAASSTPDR QAACKCLKVL AGQIKGIDMN KAAGLPGKCS VNVPFKISLS TNCDTVR

TRINITY\_DN3849\_c2\_g1\_i2 2 (7% relative abundance, 8 peptides, 6 unique peptides)homolog to AtPLAT2 (Q9SIE7), identities: 61/184 (33%)RAKMVRSFFS TLLLFTLLV ATVVAHKEAI SSLSVDRQCA YSLFVRTGDR LNSGTNAKINVTLVDSHGES FVIHNLVKFG LMKTGHNYLE NGNTDFFSVK GKCLTNPICK LILKSDGSGYRPSWFVVYLKVVQNEANVPC FESTFNIGKW IGSDYHNDGG YGVVKGGGIK HAPFELVVTLDECVKRARHSKRIDHSH

TRINITY\_DN11309\_c0\_g2\_i3 4 (7% relative abundance, 18 peptides, 17 unique peptides)homolog to isoflavonoid 7-O-beta-apiosyl-glycosidase (A3RF67), identities: 230/469 (49%)FGFGSSAYQT EGAWNVDGKG PSQWDNFTHT YPERIEDKSN GDNATNAYYL YQEDIRQLEIMNADAYWFSISWSRVLPNGRINGGRGINYKGIEYYNNLIDVLISKGLKPCVTMYFFDLPQALQDEYGGLLNGPKFREDFVQYADLLFRTFGDRVKQWVTINDPLRIVTLAYDLGVFPENRCSSWVNKDCFGGDSATEPYIVAHNLLLAHADVVQLYRSEYKEKQEGIIGIILQGTWPRPYNYSNLADHKAAGRYIDFTVGLFLSPITFGYYPQTVRDYVGERLPEFTEEQSLLLRGSIDFLGFNYFSARFVYELNIPREPRSYFDDIHVGVNYTNVNGTLVGEKLGVSWQYSYPKGIWRYLRYIKRRYNSPIIYITGNGASEVNKPHISLRAALRDNFRMKFFYLHLSYLKKAIDLEGANVKGYFAWSLTDNFEWNYGYTIRFGIIYVDHNNFARYKKLSAVWFKRFLK

Fig. S6 continued.

TRINITY DN1383 c0 g1 i2 5 (6% relative abundance, 15 peptides, 15 unique peptides) homolog to LRR receptor-like serine/threonine protein kinase GSO1 (C0LGQ5), identities: 105/324 (32%) FFFPISKFTM TQPRPSFFFF FLFPFILHLI FLFFASSVQS LTLYSDIEAL KSFRASIKTT SIPSYSCLGS WNFTAADPCA NPRVTYFTCG LMCNGNRVTH ITLDPAGYVG TLTPLISKLT QLIALDLSTN QFRGPIPVIS SLTNLQTLVL RSNSFSGGIP <mark>P</mark>SLTALK**SLE TLDFSHNYLS** GALPKSMNSL VSLIRLDLSF NRLAGPLPKL PPNIIELAIK ANSISGLLYK ASFDGLSRLE VVELSENRLT GTIQPWFFLL PSLQQVDLAN NSFTGIEIWK SSGLNSDLVA VDLGFNKIEG KLPVNFVSGG YPLLSSLSLR YNQFRGPIPV EYSKKETLKR LYLDGNYLNG WP<mark>P</mark>SGFFTGD TSFSGSLGDN CLRNCPTSSQ LCLKSQKPAS ICQLAYGGKP RS TRINITY DN4175 c2 g1 i11 1 (6% relative abundance, 7 peptides, 7 unique peptides) homolog to FLA1, fasciclin-like arabinogalactan protein 1 (Q9FM65), identities: 240/387 (62%) RTFKSIYSLL LHSSLLILSF HSLPQISSSQ VTQPISKGKE SFPKNLCNHL LFHFHAFIMQ LRTATVAAAV AFSFAFFLLP SATRAHNITH ILAGFPEFST FNHYLTTHL AAEINSRETI TVCAVDNAGM ADLLSKHLSI YALKNVLSLH VLLDYFGAKK LHQITNGTAL AATMYQATGS A<mark>P</mark>GSSGFVNI TDLKGGK<u>VGF GAVDSGAIDA TFVK</u>SIKEIP YNISIIQISK ILPS<mark>P</mark>DAEA<mark>P</mark> TPGPSQMNLT SIMSAHGCK**V FAETLLTSPA EK**TYEDNVDG GLSVFCPGDD AMKNFLPKFK NLTADGKQSL LEYHATPVYM SMPMLKSNNG PMNTLATDGP NKFAFVIQND GQEVTLKSKL

VTTK<u>IISTLI DEQPLAIYSI NK</u>VLLPKELF KGALA<mark>P</mark>TPAP A<mark>P</mark>SPDAADA<mark>P</mark> S<mark>P</mark>KKSKKHKS

PPAPAAPADS PADGPAADSA DQTADGNFAV RFSGGRILAG ALSFWFAIFI L

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Fig. S6 continued.
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**Table S1** The two gradients used for HPAEC-PAD analyses. Buffer A was 200 mM NaOH, buffer B was 15 mM NaOH and buffer C was 50 mM NaOH with 0.5 M NaAc. The time is given in minutes.

	Grad	ient 1		Gradient 2							
Time	%A	%B	%C	Time	%A	%B	%С				
-13	100	0	0	-13	100	0	0				
-8	100	0	0	-8	100	0	0				
-7	0	100	0	-7		100	0				
12	0	100	0	9	0	100	0				
12.1	0	60	40	9.1	5	85	10				
22	0	60	40	23	0	60	40				
22.1	100	0	0	25	0	60	40				
24	100	0	0	26	0	100	0				
25	0	100	0	28	0	100	0				

**Table S2** Binding of monoclonal antibodies to two samples of the *Ceropegia* 

 sandersonii polysaccharide. The *C. sandersonii* polymer was spotted along with a few

 commercial polymers to estimate the amount and strength of binding to monoclonal antibodies.

 Out of 46 selected antibodies only two showed a sufficiently strong binding.

Legend: signal strength is reported; very strong (++); strong (+): weak (+/-) or no signal (-)

	#TB Unknown polyme	#TC Unknown polyme	Polygal. Acid	Pectin esterified	RG I	Pectic galactan	Larch arabinogalactan	Linear arabinan	Probe name	Epitope
ľ		$\sim$	++	+	-27	+/-	-	-	2F4	Ca2+ cross linked HG
Γ		-	++	+	-		-	-	лм5	Homogalactauronan with a low DE
ſ	-	121	++	++	+/-	-	-	-	JIM7	Homogalactauronan with a high DE
Γ	945		++	+	( <b>2</b> 3)	++	- 2	-	LM19	Methylesterified homogalactauronan (low DE)
		(-1)	++	++		-	-	-	LM20	Methylesterified homogalactauronan (high DE)
	+	+	+	+/-	++	+	-	-	INRA-RU2	Backbone of rhamnogalacturonan I (4 units)
	-	-	+/-	+/-	++	+	-	-	INRA-RU1	Backbone of rhamnogalacturonan I (12 units)
L	-	-	+/-	+/-	+	+	-	-	CCRC-M1	Rhamnogalacturonan I/Me BSA complex
	-	-	+/-	+/-	+	+	-	-	CCRC-M13	Rhamnogalacturonan I/Me BSA complex
	$\sim$	1	3	-	-	++	++	-	LM5	(1→4)-β-D-galactan
		3			-	-	+	++	LM6	(1→5)-α-L-arabinan
Γ	$\sim$	3	0	~	-	-	-	++	LM13	Linearised (1→5)-α-L-arabinan
Γ		6	-	-	+/-	-		+	LM16	(1→5)-α-L-arabinan, RG backbone
Γ	-22	ŝ		0	100	100	2	-	LM8	Xylogalacturonan
Γ	-23	ŝ	3	÷.	-23	127	2	2	LM15	Xyloglucan (XXXG motif)
ſ		121	-	-		-	-	-	LM24	Xyloglucan (XLLG motiv, weak to XXLG)
		147	-	Ξ.	-	340	2	-	LM25	Xyloglucan (XXXG, XXLG, XLLG motiv; also GGGGGG)
Γ	-	-	-	-	-	-	-	-	LM10	(1→4)-β-D-xylan (low-substituted)
	-	140		-	94	-	-	-	LM11	(1→4)-β-D-xylan (low-substituted)/arabinoxylan)
Γ	-	-	-	-	1-0	-	-	-	LM23	(1→4)-β-D-xylan/xylogalacturonan
	-	-		-	140		=	-	BS-400-2	(1→3)-β-D-glucan
ſ	1-2	-	-	-		-	-	-	BS-400-3	(1→3) (1→4)-β-D-glucan
Γ		-	-	-	-	-	-	-	BS-400-4	(1→4)-β-D-mannan
Γ	-	-	-	~	-	-	-	~	LM21	(1→4)-β-D-mannan/galactomannan/glucomannan
Γ	-	-	-	-	-	-	-	-	LM22	(1→4)-β-D-mannan/glucomannan
Ī		-				-	-	-	JIM8	AGP (Gal-rich)
	+	+	-	-	+/-	+/-	+/-	-	JIM13	AGP; $\beta$ -GlcA-(1 $\rightarrow$ 3)- $\alpha$ -GalA-(1 $\rightarrow$ 2)- $\alpha$ -Rha
ſ		-		~	-	-	-	-	LM2	AGP; $(1\rightarrow 6)$ - $\beta$ -Gal with terminal $\beta$ -GlcA
T	~	-	-	-			-	-	LM14	AGP and/or pectic type II arabinogalactan
T	-23	-22	12	- 2	-22	12	2	2	MAC207	AGP; $\beta$ -GlcA-(1 $\rightarrow$ 3)- $\alpha$ -GalA-(1 $\rightarrow$ 2)- $\alpha$ -Rha
ſ	121	120	-	-	-	-	-	-	LM1	Extensin (hydroxyproline-rich motif THRGP)
ſ		-		-	123		- 21	-	LM3	Extensin
ſ	-		-	=			-	-	JIM11	Extensin
ſ	-	120	+/-	+/-		-	-	-	JIM12	Extensin
Ī		-	-	-	-	-	-	-	JIM19	Extensin
T	-	-	+/-	+/-	-	-		-	JIM20	Extensin
Ī	100	-	-	-	-	-	-	-	B-1005	Concanavalin A; α-linked Man
f		-	-	-	-	-	-	-	B-1015	Soybean agglutinin; α- or β-linked GalNAc
t		-	-	~	-	-	-	-	B-1025	Wheat germ agglutinin; dimers and trimers of GlcNAc
Ī		-		-	-	-	-	-	B-1065	Ulex Europaeus Agglutinin Ι; α-linked Fuc
t	-	-	-		-	-	-	-	B-1085	Ricinus communis agglutinin; Gal or GalNAc residues
ſ	-		-	-	-	-	-	-	B-1285	Bauhinia purpurea lectin; Gal-α-(1→3) GalNAc, terminal GalNAc
T	-	-	-	-	-	-	-	-	B-1305	Sambucus nigra lectin; sialic acid attached to term. Gal, GalNAc
Ī	+/-	+/-	-	-		-	-	-	B-1335	Euonymus europaeus lectin; Gal- $\alpha$ -(1 $\rightarrow$ 3)-Fuc- $\alpha$ -(1 $\rightarrow$ 2)-Gal
ſ	-	100	-		-	-	-	-	B-1365	Psophocarpus tetragonolobus lectin; GalNAc
ſ	- 1	-	-	-		-	-	-	B-1405	Psophocarpus tetragonolobus lectin II; Gal and GalNAc
-	-	-								· · · · ·

**Table S3** Observed chemical shifts of the disaccharide GlcA $\beta$ 1,6Gal at 298 K, referenced to DSS in comparison with chemical shift predictions with the software CASPER (Furevi *et al.*, 2022) and the NMR values of Menestresina et al. 1998 (Menestrina *et al.*, 1998).

Moiety	H1	H2	H3	H4	H5	H6	H6'	C1	C2	C3	C4	C5	C6	
	4.545 (α)ª 4.552 (β)ª	3.36	3.54	3.55	3.85 <sup>b</sup>			105.2	75.6	78.1	74.3	78.1 <sup>₅</sup>	177.1 <sup>₅</sup>	this work
GICAD 1,0-	4.52 (α)ª 4.54 (β)ª	3.36	3.53	3.52	3.73 (α)ª 3.72 (β)ª			105.2	75.6	78.2	74.4	78.7	178.2	CASPER⁰
	4.48 (α)ª 4.49 (β)ª							105.2					177.8	Menestresinad
R1 6C alax	5.28	3.81	3.87	4.02	4.28	3.84	4.06	95.0	71.0	71.7	72.0	71.9	72.1	this work
-p1,00al0	5.22	3.77	3.82	3.98	4.22	3.82	3.98	95.0	71.0	71.7	72.0	71.8	71.5	CASPER⁰
	5.23							95.0						Menestresinad
81 6C al 8	4.60	3.51	3.66	3.96	3.90	3.88	4.06	99.1	74.5	75.3	71.5	77.8	72.0	this work
-рт,обар	4.55	3.46	3.60	3.93	3.84	3.79	4.00	99.0	74.6	75.4	71.5	76.4	71.4	CASPER⁰
	4.55							99.1						Menestresinad

<sup>a</sup> the chemical shifts depend on the anomeric form at the Gal at the reducing end. ( $\alpha$ ) stands for Gal $\alpha$  at the reducing end and ( $\beta$ ) stands for Gal $\beta$  at the reducing end.

<sup>b</sup> the chemical shift values are pH dependent due to the equilibrium between COOH and COO<sup>-</sup> at C6

<sup>c</sup> the calculated <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.7 ppm was added to the <sup>13</sup>C values

<sup>d</sup> the calculated <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +2.2 ppm was added to the <sup>13</sup>C values

**Table S4** Observed chemical shifts of the intact polysaccharide of *Ceropegia sandersonii* referenced to DSS at 25°C in comparison with previously reported NMR data (Tan *et al.*, 2010; Cartmell *et al.*, 2018; Tan *et al.*, 2023). In addition very small signals of short Araf $\alpha$ 1,3Gal $\beta$ 1,6-and Gal $\beta$ 1,6-side chains are also visible in the spectra but omitted here. Their chemical shifts are reported in Table S5. The name of the species in the references is indicated in brackets.

Comment	Moiety	H1	H2	H3	H4	H5	H6/ H5'	H6'	C1	C2	C3	C4	C5	C6	
	Rha (b)	4.747	3.952	3.781	3.436	4.043	1.262		103.4	72.9	72.8	74.6	71.7	19.2	this work
		4.742	3.926	3.746	3.432	4.013	1.245		103.6ª	72.9ª	72.8ª	74.6ª	71.8ª	19.1ª	Cartmell
		4.90	3.98	3.75	3.47	3.96	1.26		103.6 <sup>b</sup>	73.2 <sup>b</sup>	73.1 <sup>b</sup>	74.8 <sup>b</sup>	71.9 <sup>b</sup>	19.2 <sup>b</sup>	CASPER
linear side chain	GlcA (d)	4.525	3.384	3.584	3.59	3.763	_		105.3	76.0	76.9	81.8	78.8	177.9	this work
Rhaα1,4 GlcAβ1.6		4.558	3.369	3.605	3.639	3.955	_		105.4ª	75.9ª	76.8ª	81.6ª	77.1ª	176.0ª	Cartmell
Galβ1,6- bbGal		4.55	3.38	3.65	3.66	3.80	_		105.2 <sup>₅</sup>	76.0 <sup>b</sup>	77. <b>1</b> <sup>b</sup>	82.4 <sup>b</sup>	78.1 <sup>b</sup>	178.0 <sup>b</sup>	CASPER
	Gal (f)	4.459	3.548	3.669	3.965	3.93	4.076	3.917	106.3	73.4	75.3	71.3	76.2	72.3	this work
		4.433	3.529	3.651	3.934	3.894	4.016	3.897	105.8ª	73.4ª	75.2ª	71.3ª	76.5ª	72.2ª	Cartmell
		4.44	3.52	3.61	3.91	3.84	4.02	3.81	105.9⊳	73.7 <sup>b</sup>	75.6 <sup>b</sup>	71.7♭	76.6 <sup>b</sup>	71.7 <sup>₅</sup>	CASPER
	Rha (b)	4.747	3.952	3.781	3.436	4.043	1.262		103.4	72.9	72.8	74.6	71.7	19.2	this work
		4.81	3.95	3.78	3.42	4.02	1.25		103.3ª	73.0 <sup>d</sup>	73.0 <sup>d</sup>	74.8 <sup>d</sup>	71.7ª	19.2 <sup>d</sup>	Tan 2010 (R)
branched	GlcA	4.525	3.384	3.584	3.59	3.763	_		105.3	76.0	76.9	81.8	78.8	177.9	this work
Rhaα1,4	(d)	4.51	3.45	3.56°	3.63	3.73⁰	—		105.3ª	76.0 <sup>d</sup>	77.1 <sup>d,e</sup>	81.8 <sup>d</sup>	79.0 <sup>d,e</sup>		Tan 2010 (UA)
GlcA $\beta$ 1,6 [Araf $\alpha$ 1,3]	Araf	5.25	4.232	3.948	4.138	3.841	3.715		111.9	84.0	79.3	86.5	64.0		this work
Galβ1,6- bbGal ∘	(a)	5.25	4.21	3.95	4.13	3.82	3.71		111.9ª	83.9 <sup>d</sup>	79.3 <sup>d</sup>	86.7 <sup>d</sup>	64.0 <sup>d</sup>	_	Tan 2010 (A)
	Gal	4.505	3.658	3.732	4.152	3.914	4.076	3.917	106.1	72.5	82.9	71.1	76.3	72.3	this work
	(e)	4.47	3.65	3.71	4.11	3.91	4.04	3.94	106.1ª	73.3 <sup>d</sup>	83.3 <sup>d</sup>	71.5 <sup>d</sup>	76.3 <sup>d</sup>	72.1 <sup>d</sup>	Tan 2010 (Ga)
short linear		4.522	3.358	3.531	3.536	3.742	—	_	105.3	75.6	78.2	74.5	78.6	n.d.	this work
short linear side chain GlcAβ1,6 Galβ1,6- bbGal	GlcA (d′)	4.51	3.34	3.52e	3.54	3.74 <sup>e</sup>			105.8 <sup>r</sup>	75.7 <sup>f</sup>	78.0 <sup>e,f</sup>	73.0 <sup>r</sup>	78.2 <sup>e,f</sup>	n.r.	Tan 2023 (E)
		4.54	3.36	3.54	3.52	3.72			105.4 <sup>b</sup>	75.8 <sup>b</sup>	78.5 <sup>b</sup>	74.6 <sup>b</sup>	78.9 <sup>b</sup>	178.4 <sup>b</sup>	CASPER
	Gal (f)	4.459	3.548	3.669	3.965	3.93	4.076	3.917	106.3	73.4	75.3	71.3	76.2	72.3	this work

		4.46	3.66	3.72	4.10	3.88	4.03	3.92	106.0 <sup>r</sup>	72.2 <sup>f</sup>	82.9 <sup>r</sup>	71.1 <sup>r</sup>	76.2 <sup>r</sup>	72.0 <sup>r</sup>	Tan 2023 (F)
		4.44	3.52	3.61	3.91	3.84	4.02	3.81	105.9	73.7 <sup>b</sup>	75.6 <sup>b</sup>	71.7 <sup>b</sup>	76.6 <sup>b</sup>	71.7 <sup>₅</sup>	CASPER
		4.713	3.785	3.895	4.248	3.894	4.052	3.984	106.4	72.8	84.3	71.2	76.3	72.0	this work
backbone	Gal (c)	4.64 <sup>g</sup>	3.65 <sup>g</sup>	3.85	4.23	3.77	4.04	3.94	106.6 <sup>d</sup>	73.2 <sup>d</sup>	84.6 <sup>d</sup>	71.3ª	76.4 <sup>d</sup>	72.1ª	Tan 2010 (G2)
Galβ1,3-		4.70	3.79	3.88	4.21	3.79	4.03	3.92	106.3 <sup>r</sup>	72.2 <sup>f</sup>	84.8 <sup>r</sup>	71.2 <sup>r</sup>	75.9 <sup>r</sup>	72.0 <sup>r</sup>	Tan 2023 (C)
		4.62	3.73	3.78	4.19	3.86	3.99	3.85	106.8 <sup>b</sup>	72.8 <sup>b</sup>	84.8 <sup>b</sup>	71.1 <sup>b</sup>	76.2 <sup>b</sup>	71.4 <sup>b</sup>	CASPER

<sup>a</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.9 ppm was added to the <sup>13</sup>C values

<sup>b</sup> the calculated <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.9 ppm was added to the <sup>13</sup>C values

<sup>c</sup> CASPER does not include arabinofuranose and therefore the chemical shifts of oligosaccharides containing Araf cannot be predicted

<sup>d</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.6 ppm was added to the <sup>13</sup>C values

<sup>e</sup> the reported values seemed to have interchanged C3/H3 and C5/H5 assignments. Here we swapped the assignments

<sup>f</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +2.0 ppm was added to the <sup>13</sup>C values

<sup>g</sup> values deviate, but values of Tan et al. 2023 fit

**Table S5** Observed chemical shifts of the polysaccharide of *Ceropegia sandersonii* after enzymatic cleavage referenced to DSS at 25°C in comparison with previously reported NMR data (Delgobo *et al.*, 1999; Endo *et al.*, 2013) and predictions with the software CASPER (Furevi *et al.*, 2022). The compound number of the reference is indicated in brackets.

Comment	Moiety	H1	H2	H3	H4	H5	H6/ H5'	H6'	C1	C2	C3	C4	C5	C6	
lin e en eide	Araf (a)	5.253	4.228	3.953	4.135	3.84	3.721		111.9	84.1	79.3	86.6	64.0	—	this work
		5.26	4.20	3.93	4.07	3.82	n.r.	_	111.7 <sup>₅</sup>	84.0 <sup>b</sup>	79.3 <sup>b</sup>	86.8 <sup>b</sup>	63.9 <sup>b</sup>	_	Delgobo (4)
chain		5.25°	4.22 <sup>c</sup>	3.96 <sup>c</sup>	n.r.	3.85 <sup>c</sup>	3.76°		112.0ª	84.0 <sup>d</sup>	79.3 <sup>d</sup>	86.6 <sup>d</sup>	64.0 <sup>d</sup>	—	Endo
Arafα1,3 Galβ1,6-		4.499	3.653	3.724	4.111	3.713	3.774		106.1	72.7	83.0	71.3	77.7	63.7	this work
DDGal ª	Gal (e')	4.48	3.58	3.78	4.01	n.r.	n.r.		105.7 <sup>₅</sup>	73.7 <sup>b</sup>	75.8 <sup>b,e</sup>	77.7 <sup>b,f</sup>	n.r.	64.0 <sup>b</sup>	Delgobo (4)
		4.48°	n.r.	n.r.	n.r.	n.r.	3.80°		105.9ª	n.r.	82.9 <sup>d</sup>	70.6 <sup>d</sup>	n.r.	n.r.	Endo
single saccharide	Gal	4.449	3.548	3.653	3.928	3.706	3.776		106.3	73.5	75.5	71.4	77.8	63.7	this work
side chain Galβ1,6- bbGal backbone Galβ1,3-	(f′)	4.42	3.51	3.60	3.87	3.65	3.74	3.66	106.0 <sup>g</sup>	73.7 <sup>g</sup>	75.7 <sup>g</sup>	71.6 <sup>g</sup>	77.9 <sup>g</sup>	63.8 <sup>g</sup>	CASPER
	Gal	4.713	3.786	3.882	4.241	3.931	4.057	3.92	106.5	73.0	84.5	71.3	76.2	72.0	this work
	(c)	4.62	3.73	3.78	4.19	3.86	3.99	3.85	106.8 <sup>g</sup>	72.8 <sup>g</sup>	84.8 <sup>g</sup>	71.1º	76.2 <sup>g</sup>	71.4 <sup>g</sup>	CASPER

<sup>a</sup> CASPER does not include arabinofuranose and therefore the chemical shifts of oligosaccharides containing Araf cannot be predicted

<sup>b</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +2.3 ppm was added to the <sup>13</sup>C values

<sup>c</sup> the reported <sup>1</sup>H values showed an offset due to different referencing. For better comparison a correction of +0.06 ppm was added to the <sup>1</sup>H values

<sup>d</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +2.6 ppm was added to the <sup>13</sup>C values

<sup>e</sup> the reported value for C3 is likely incorrectly assigned

<sup>f</sup> the reported value for C4 is likely incorrectly assigned, it would fit to C5

<sup>g</sup> the calculated <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.9 ppm was added to the <sup>13</sup>C values

**Table S6** Observed chemical shifts of the cleaved disaccharide Rhaα1,4GlcA at 298 K, referenced to DSS in comparison with chemical shift values of Fügedi 1987 (Fugedi, 1987) and predictions with the software CASPER (Furevi *et al.*, 2022).

Moiety	H1	H2	H3	H4	H5	H6	C1	C2	C3	C4	C5	C6	
Rhaα1,4 (b)	4.727 (α)ª 4.734 (β)ª	3.953 (α)ª 3.980 (β)ª	3.784 (α)ª 3.765 (β)ª	3.441 (α)ª 3.435 (β)ª	4.052 (α)ª 4.042 (β)ª	1.260	103.5	73.0	72.8	74.7	71.6	19.20	this work
	n.r. <sup>b</sup>	n.r. <sup>b</sup>	103.6°	72.8°	72.8°	74.5°	71.8°	19.1°	Fügedi				
	4.90	3.98	3.75	3.47	3.96	1.26	103.6 <sup>d</sup>	73.2 <sup>d</sup>	73.1ª	74.8 <sup>d</sup>	71.9 <sup>d</sup>	19.2 <sup>d</sup>	CASPER
	5.239	3.611	3.743	3.548	4.137	—	94.7	74.4	74.2	82.4 <sup>e</sup>	74.6 <sup>e</sup>	n.d.	this work
GlcAα (gα)	n.r. <sup>b</sup>	_	94.8°	73.9°	73.9°	81.4°	72.3°	n.r. <sup>b</sup>	Fügedi				
	5.25	3.61	3.87	3.67	4.16	_	94.7 <sup>d</sup>	74.5 <sup>d</sup>	74.1ª	82.6 <sup>d</sup>	73.5 <sup>d</sup>	178.9ª	CASPER
	4.633	3.311	3.559	3.576	3.759	—	98.6	77.1	77.1	82.0e	79.2 <sup>e</sup>	n.d.	this work
GlcAβ (gβ)	n.r. <sup>b</sup>	_	98.7°	76.7°	76.7°	<b>81.7</b> ℃	76.2 <sup>c</sup>	n.r. <sup>b</sup>	Fügedi				
	4.66	3.32	3.64	3.68	3.80	_	98.5 <sup>d</sup>	77.2 <sup>d</sup>	77.1 <sup>d</sup>	82.4 <sup>d</sup>	78.0 <sup>d</sup>	177.9 <sup>d</sup>	CASPER

<sup>a</sup> the chemical shifts depend on the anomeric form at the Gal at the reducing end. ( $\alpha$ ) stands for GlcA $\alpha$  at the reducing end, ( $\beta$ ) stands for GlcA $\beta$  at the reducing end

<sup>b</sup> n.r. stands for not reported, few <sup>1</sup>H data were reported for a different solvent

<sup>c</sup> the reported <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.9 ppm was added to the <sup>13</sup>C values

<sup>d</sup> the predicted <sup>13</sup>C values showed an offset due to different referencing. For better comparison a correction of +1.9 ppm was added to the <sup>13</sup>C values

<sup>e</sup> the atoms are close to the carboxyl group which is in equilibrium between the neutral and charged form depending on the pH. Chemical shifts in the environment can change depending on the pH value.

#### Methods S1

### Efforts to identify the protein component of the Ceropegia polymer by LC-HRMS

One 110  $\mu$ L aliquot of the gliding zone wash-off was diluted 1/2 (v/v) in 100 mmol/L triethylammonium bicarbonate buffer (TEAB, pH 8.50, Sigma-Aldrich), containing 10 % (w/w) sodium dodecyl sulfate (SDS, Sigma-Aldrich) and 1× protease inhibitor cocktail (Roche), and heated for 5 min at 95°C for denaturation. In order to reduce possible disulfides, the sample was treated with 5 mmol/L tris-(2-carboxyethyl)-phosphine-hydrochloride (TCEP, Sigma-Aldrich) at 55 °C for 15 min. This was followed by alkylation of the cysteine residues by addition of iodoacetamide (Sigma-Aldrich) to a final concentration 40 mmol/L and incubation at 22 °C in the dark for 10 min. After this, the sample was acidified to  $pH \le 1$  with 12 % (v/v) orthophosphoric acid (Merck) followed by protein precipitation by adding 7:1 (v/v) of 100 mM TEAB (pH 7.55) in 90 % methanol (v/v). Next the proteins were purified by suspension trapping employing S-Trap mini columns (Protifi) according to the manufacturer's instructions, and digested to peptides by addition of 10 µg trypsin (sequencing grade modified, porcine, Promega, Madison, WI, USA) and incubation at 37 °C for 16 h. The purified peptides were dried at 50 °C in a vacuum centrifuge and resuspended in 10  $\mu$ L 1.0% aqueous acetonitrile (ACN; VWR International) with 0.010% formic acid (FA; Sigma-Aldrich). Next, 0.330 µL of the sample were injected and chromatographically separated by reversed phase HPLC on an UltiMate<sup>™</sup> 3000 RSLCnano System (Thermo Fisher Scientific), employing a DNV PepMap<sup>™</sup> Neo column (150 x 0.075 mm i.d.) (Thermo Fisher Scientific). For the separation, 0.10 % aqueous FA (solvent A) and 0.10 % FA in ACN (solvent B) were pumped at a flow rate of 300.0 nL/min in the following order: 1.0 % B for 2.0 min, a linear gradient from 1.0-10.0 % B in 3.0 min, a second linear gradient from 10.0-35.0 % B in 45.0 min, and a third linear gradient from 35.0-45.0 % B in 10.0 min. This was followed by flushing with 99.0 % B for 5 min and column re-equilibration with 1.0 % B for 30 min. The column temperature was kept constant at 50 °C, the autosampler was kept at 5 °C. The nanoHPLC system was hyphenated to a Q Exactive™ Hybrid Quadrupole-Orbitrap<sup>™</sup> mass spectrometer via a Nanospray Flex<sup>™</sup> ion source (both from Thermo Fisher Scientific). The source was equipped with a SilicaTip emitter with 360 µm o.d., 20 µm i.d. and a tip i.d. of 10 µm (CoAnn Technologies Inc). The spray voltage was set to 1.5 kV, S-lens RF level to 60.0 and capillary temperature to 250 °C. Each scan cycle consisted of a full scan at a scan range of m/z 350-2,000 and a resolution setting of 70,000 at m/z 200, followed by 15 datadependent higher-energy collisional dissociation (HCD) scans in a 2.0 m/z isolation window at 28 % normalized collision energy at a resolution setting of 17,500 at m/z 200. For the full scan the automatic gain control (AGC) target was set to 3e6 charges with a maximum injection time of 100 ms, for the HCD scans the AGC target was 1e5 charges with a maximum injection time of 150 ms. Already fragmented precursor ions were excluded for 30 seconds. Data acquisition was conducted using Thermo Scientific<sup>™</sup> Chromeleon<sup>™</sup> 7.2 CDS (Thermo Fisher Scientific). For raw data evaluation, MaxQuant 2.0.1.0 (Cox & Mann, 2008) was used in the default settings. For protein identification a custom protein sequence database was created by translating all Ceropegia transcriptome sequences in all six reading frames using transeq (Madeira et al., 2022). Subsequently, the longest continuous protein sequence fragment appearing within the respective six reading frames was extracted and added to the custom database. This custom-built database was used for protein identification, applying a 1 % false discovery rate. The mass spectrometry proteomics data have been deposited to the ProteomeXchange Consortium via the PRIDE (Perez-Riverol et al., 2025) partner repository with the dataset identifier PXD062347.

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