





Figure S1. Endogenous hormone Salicylic acid (SA), jasmonic acid (JA), and ethylene (ET) contents in both *Alternanthera philoxiroides* and *A. sessilis* after *Rhizoctonia solani* inoculations. (a) SA, (b) JA and (c) ET. Four-week-old plants were inoculated with *R. solani* and un-inoculated leaves were used as a control (0h). Samples were harvested at the indicated time points for quantification using ELISA. Values are means \pm SE (n = 4). The different letters indicate significant difference in hormone levels compared with control un-inoculated plant leaves assessed by Duncan's test at *P* < 0.01. Abbreviations: AP - *A. philoxeroides*, AS - *A. sessilis*.

(a) *PAL* (SA)



(b) *JAR1* (JA)



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(c) *EIN3* (ET)



Figure S2. Comparative phylogenetic analysis of the selected hormone genes in both the invasive and native species, as well as other closely related species. A maximum likelihood analysis was performed for each gene: (a) *PAL* (Salicylic Acid), (b) *JAR1* (Jasmonic Acid), (c) *EIN3* (Ethylene). A maximum likelihood phylogenetic tree was constructed with 1000 bootstrap replicates. Bootstraps are shown on the nodes of the trees. Genetic distances are shown below each branch.

(a) *PAL* (SA)

1)											
1. PA	L_A.philoxeroides	5.90e-287 -									
2. PA	L_A.sessilis	1.67e-283 -									
3. XP	_010684079.1_PAL_B.vulgaris	1.93e-283 -									
4. XP	_021856773.1_PAL_S.oleracea	2.80e-285 -									
5. XP	_021730766.1_PAL_C.quinoa	2.57e-284 -									
6. AM	IQ23633.1_PAL_S.littorea	5.33e-285 -									
7. AA	R19393.1_PAL_S.longipes	2.22e-271 -				_					
8. NP	_001311946.1PAL_N.tabacum	7.89e-278 -									
9. XP	_009766905.1_PAL_N.sylvestris	7.89e-278 -			_	_					
10. XP	_019187025.1_PAL_I.nil	4.69e-275 -									6
2) 1.	≝2 	A EA	AAIME	I L DG	SYøKA	KAL	EmD	L Q K P K	(QDR Y	ALRISPOW	1.4e-389
2.		R¥	AT <mark>K</mark> sii	REIN			S RNKA		VFQ GT	<u>P GV SMDN</u>	3.2e-372
3.		ΚĻř	FAQFS	ĻŸŊŊ	YNNGL		TASRE	SLD	GFKG	A <mark>e I Amasy</mark>	1.5e-351
4.	RN	ĻQĢ	Y SG I R.	EILE	K	NNNI s s s s s s			I TAS <mark>g</mark>	DLVPLSYI	2.2e-346
5.	"	GSG	MAS _h V	F an			A¥FAE	YMNG	(PEFT	DILTIKLK	2.6e-335

(b) *JAR1* (JA)



- 4. **CSPDEVUEGEDE HQSLYCHLLCGLLERDEVQVUSSTEALS**UVAFRIEQ 1.5e-358
- 5. **TOCKEKFVPFNDELMESTMOLYKTSFAFRNBEFPUGNCKALOFIYSSKOF** 3.1e-342

(c) *EIN3* (ET)



Figure S3. Identification of conserved motifs in selected genes from both invasive and native species. (a) *PAL* (Salicylic Acid), (b) *JAR1* (Jasmonic Acid), (c) *EIN3* (Ethylene). For each gene the following are shown: 1) a conserved motif revealing five motifs from the full-length protein sequences of all analyzed species with greater e-value, and 2) conserved amino acid sequences and the conservation of each motif are represented as a logo in each gene with their bit score as height of amino acid.

(a1) PAL (SA) - A. philoxeroides

	1	50 1 1 1 1 1	100		150	200		250	30 1 1 1 1		360
Superfamilies	7				PLN024	57 supei	rfamily				<u> </u>
seqsig_MYRRS_(Cdd:PLN02457	0c22b369ab57	78ae26407f01d4	29afe57 26 122	10 *	20 *	30 * TATRAAMLVRI SATRAAMLVRI	40 .* INTLLQGYSG INTLLQGYSG	50 .* IRFEILEAITS IRFEILEAITS	60 .* * SFLNNNVTPCLF KLLNANVTPCLF	70 *. LRGTITASGDLV	80 /PLS 105 /PLS 200
seqsig_MYRRS_(Cdd:PLN02457	0c22b369ab57	78ae26407f01d4	29afe57 106 201	90 *	100 *	110 .* AEQAFKTAHIS AAEAFKLAGIS	120 .* SSEFFELQPKI EGGFFELQPKI	130 .* EGLALVNGTAN EGLALVNGTAN	140 .* * /GSGMASIVLFE /GSALASTVLFE	150 * ANILAVLAEVIS ANVLAVLAEVIS	160 AIF 185 AVF 280
seqsig_MYRRS_ Cdd:PLN02457	0c22b369ab57	78ae26407f01d4	29afe57 186 281	170 *	180 *	190 .* QIEAAAIMEHI QIEAAAIMEHI	200 .*	210 .* AKALHELDPLO AKKLHETDPLO	220 * * QKPKQDRYALRT QKPKQDRYALRT	230 SPQWLGPQIEV SPQWLGPQIEV	240 IRWA 265 IRAA 360
seqsig_MYRRS_(Cdd:PLN02457	0c22b369ab57	78ae26407f01d4	29afe57 266 361	250 *	260 *	270 .* ALHGGNFQGTR ALHGGNFQGTR	280 .* PIGVSMDNARI PIGVSMDNTRI	290 .* LALAAIGKLLH LAIAAIGKLM	300 * * FAQFSELVNDYY FAQFSELVNDFY	310 MNGLPSNLTASF	320 NPS 345 NPS 440
seqsig_MYRRS_(Cdd:PLN02457	0c22b369ab57	78ae26407f01d4	29afe57 346 441	330 *	* IASY 360 IASY 455						
(a2) <i>PAL</i>	(SA) -	A. sessili	s	100	150		200		250	300	331
Query seq. Superfamilies				F	N02457	cuporfa	milu				
Superiumities				10	20		40	50	60	70	80
seqsig_FLNAG_ Cdd:PLN02457	b3e42e3fdc5	a706813829f01c	192962f 1 126	FLNAGVFGNGtE	TSHTLPHTATR	.*AAMLVRINTLI	.* LQGYSGIRFE	.*	.*	TTASGDLVPLS	/IAG 80 /IAG 204
seqsig_FLNAG_ Cdd:PLN02457	b3e42e3fdc5	a706813829f01c	192962f 81 205	90 *	100 *	110 .* FKTAHISSEFI FKLAGIEGGFI	120 .*	130 .* LVNGTAVGSGI LVNGTAVGSA	140 .* * MASIVLFEANIL LASTVLFDANVL	150 * AVLAEVISAIF/ AVLAEVLSAVF(160 AEVM 160 EVM 284
seqsig_FLNAG_ Cdd:PLN02457	b3e42e3fdc5	a706813829f01c	192962f 161 285	170 *	180 *	190 .*AAIMEHILDG AAIMEHILDG	200 .* SSYMKAAKALI SSYMKAAKKLI	210 .* HELDPLQKPK0 HETDPLQKPK0	220 ** QDRYALRTSPQN QDRYALRTSPQN	230 * ILGPQIEVIRMAT	240 KSI 240 KSI 364
seqsig_FLNAG_ Cdd:PLN02457	b3e42e3fdc5	a706813829f01c	192962f 241 365	250 *	260 *	270 .* ANFQGTSIGV GNFQGTPIGV	280 .* SMDNARLALA SMDNTRLAIA	290 .* AIGKLLFAQF AIGKLMFAQF	300 .** SELVNDFYNNGL	310 	320 .DYG 320 .DYG 444

seqsig_FLNAG_b3e42e3fdc5a706813829f01c192962f 321 FKGAEIAMASY 331 Cdd:PLN02457 445 FKGAEIAMASY 455

(b) JAR1 (JA) - A. philoxeroides

0							Ľ.
Specific hits			GH3				
Superfamilies		GH3	superfamily				
seqsig_MSKLL_faa286a75 dd:pfam03321	f505e9cee82a46bf57179c0 2:	10 *	20 30 * RKCSS1sKWYGLIPELF RECEKGWEGIIKRLW	40 ! .* * PNAKYIYGIMTGSMEI PNLKYVFCIGTGSME(50 60 . * * PYVKQLRRYAGWLPLVCGU QYIPKLRHYAGGLPLVSEC	70 80 * DYGSSEGWVAANVM GYGASEGFFGINLD	80 0 31
seqsig_MSKLL_faa286a75 Idd:pfam03321	f505e9cee82a46bf57179c0 3 3:	90 *	100 110 *	120 1: .* * SKTVGLHEVKVGQEYI PETLDLVEVEVGKEYI	30 140 . * * EIIVTTFAGLYRYRLGDAV ELVVTTFSGLYRYRIGDVV	150 160 * VKVMGFHNSTPELK VRVTGFYNQTPRLE) (16 5 39)
eqsig_MSKLL_faa286a75 Idd:pfam03321	f505e9cee82a46bf57179c0 10 39	170 *	180 190 * ITEKDLQVAVEEASKLLv TEEDLQKAVEEAAKEL-	200 2: .* * aEKIDLIDFTSNVNV -TGAELVDYTSYADT	10 220 . * * T-KEPGHYVIFWELSVEAD SgSIPGHYVLFWELK-DDD	230 240 * DENVMQECCNCLDK DPDVLEECCAELDE) (23) E 47(
eqsig_MSKLL_faa286a75 Idd:pfam03321	f505e9cee82a46bf57179c0 2 4	250 *	260 270 * MIGPLELRVLRKGTFHK SIGPLEIRVVKPGTFDK	280 29 .* *. ILLHSLSMGTTLNQFI	90 . *. (TPRFVGS 293 (VPRLVNS 526		
Query seq. Specific hits Superfamilies			EIN3				
			EIN3 superf	amily			
			EIN3 superf	amily			
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873	b08d591fe2277b5426e61c6	10 * * 15 DELEQRMWRDRMLLK 1 EDLEKRMWKDRMLLK	20 30 20*	40 	50 60 *	70 8 	0 N 91 K 80
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873	b08d591fe2277b5426e61c6 b08d591fe2277b5426e61c6	10 ** 15 DELEQRMWRDRMLLK 1 EDLEKRMWKDRMLLK 90 *	20 30 *	40 .* DAAKQRQSQEQARRKK GKSKPKQPSEQARRKK 120 12	50 60 . * *. MSRAQDRILKYMLKMMEV MSRAQDGILKYMLKMMEV 30 140 . * *. ASTPHTLQELQDTTLGSL CSGPHSLHELQDTTLGSL	70 8 * /CKAQGFVYGIIPK /CNAQGFVYGIIPE 150 16 * .LSALMQHCDPPQR .LSALMQHCDPPQR	00 N 91 K 80 R 17 R 15
<pre>seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873</pre>	b08d591fe2277b5426e61c6 b08d591fe2277b5426e61c6 b08d591fe2277b5426e61c6 1 1	10 ** 15 DELEGRMWRDRMLLK 1 EDLEKRMWKDRMLLK 90 ** 92 GKPVSGASDNLRAHW 81 GKPVSGSSDNLRAHW 170 ** 72 FPLEKGAPPPWMPIG 59 FPLEKGVPPPMWPTG	EIN3 superf 20 30 *	40 40 40 40 40 40 40 40 40 40	50 60 . * *. MSRAQDRILKYMLKMMEV MSRAQDGILKYMLKMMEV 30 140 . * *. ASTPHTLQELQDTTLGSL CSGPHSLHELQDTTLGSL 218 205	70 8 * CKAQGFVYGIIPK (CNAQGFVYGIIPE 150 16 * LSALMQHCDPPQR LSALMQHCDPPQR	00 N 91 K 80 R 17 R 15
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 (c2) EIN3 (ET	2608d591fe2277b5426e61c6 2608d591fe2277b5426e61c6 2608d591fe2277b5426e61c6 1 1 20 - A. sessilis	10 *	EIN3 superf 20 30 *	40 .*	50 60 . * *. MSRAQDRILKYMLKMMEV MSRAQDGILKYMLKMMEV 30 140 . * *. ASTPHTLQELQDTTLGSL CSGPHSLHELQDTTLGSL 218 205	70 8 * (CKAQGFVYGIIPK (CNAQGFVYGIIPE 150 16 *. LSALMQHCDPPQR LSALMQHCDPPQR	0 9: 8 8 1 1 7 1
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 (c2) EIN3 (ET Query seq.	2608d591fe2277b5426e61c6 2608d591fe2277b5426e61c6 2608d591fe2277b5426e61c6 1 1 2) - A. sessilis	10 *	EIN3 superf	40 40 40 40 40 40 40 40 40 40	50 60 . * *. MSRAQDRILKYMLKYMEV MSRAQDGILKYMLKYMEV 30 140 . * *. ASTPHTLGELQDTTLGSL CSGPHSLHELQDTTLGSL 218 205	70 8 * (CKAQGFVYGIIPK (CNAQGFVYGIIPE 150 16 * LSALMQHCDPPQR LSALMQHCDPPQR	488
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 (c2) EIN3 (ET Query seq.	bological statement in the second statement is a second statement in the second statement in the second statement is a second statement in the second statement in the second statement is a second statement in the second statement in the second statement is a second statement in the second sta	10 ** 15 DELEQRMWRDRMLLK 1 EDLEKRMWKDRMLLK 90 ** 92 GKPVSGASDNLRAHW 81 GKPVSGSSDNLRAHW 170 ** 72 FPLEKGAPPPWWPTG 59 FPLEKGVPPPWWPTG	EIN3 superf 20 30 *	40 40 40 40 40 40 40 40 40 40	50 60 . * *. MSRAQDRILKYMLKYMEV MSRAQDGILKYMLKYMEV 30 140 . * *. ASTPHTLQELQDTTLGSL CSGPHSLHELQDTTLGSL 218 205	70 8 	488
seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 seqsig_VGVEE_2094d2eb4 Cdd:pfam04873 (c2) EIN3 (ET Query seq. Specific hits Superfamilies	bo8d591fe2277b5426e61c6 bo8d591fe2277b5426e61c6 bb08d591fe2277b5426e61c6 1 1 2) - A. sessilis 5) - A. sessilis EIN3	10 ** 15 DELEGRMWRDRMLLK 1 EDLEKRMWKDRMLLK 90 ** 92 GKPVSGASDNLRAHM 81 GKPVSGSSDNLRAHM 170 ** 72 FPLEKGAPPPWWPIG 59 FPLEKGVPPPWWPIG 59 FPLEKGVPPPWWPIG	EIN3 superf	40 *	50 60 * MSRAQORILKYMLKYMEV MSRAQOGILKYMLKYMEV 30 140 * ASTPHTLQELQDTTLGSL CSGPHSLHELQDTTLGSL 218 205	70 8 	488

seqsig_MGDEA_ae4829c9a4cc6f4c9a81d0141b7e1964 Cdd:pfam04873	104 81	90 * GKPVSGASDNLRAWW GKPVSGSSDNLRAWW	100 EKVRFDR EKVRFDR	110 .* NGPAAISKYQAI NGPAAIAKYQAI	120 * DHSIPGTNED DNLLAGMSDE	130 * CNtvASTPH SGCSGPH	140 .* TLQELQDTTLG SLHELQDTTLG	150 * SLLSALMQHC SLLSALMQHC	160 * DPPQRR : DPPQRR :	183 158
seqsig_MGDEA_ae4829c9a4cc6f4c9a81d0141b7e1964 Cdd:pfam04873	184 159	170 * FPLEKGVPPPWWPIGI FPLEKGVPPPWWPIGI	180 IEEWWPQL (ELWWGQL)	190 .* GLPKDQGPPPYI GLPKDQGPPPYI	200 * KKPHDLKKAW RKPHDLKKAW	210 * KVSVLTAVII KVSVLTAVII	220 .* KHMSPDIAKIR KHMSPDISKIR	230 * KLVRQSKCLQ KLVRQSKCLQ	240 * DKMTAK DKMTAK	263 238
seqsig_MGDEA_ae4829c9a4cc6f4c9a81d0141b7e1964 Cdd:pfam04873	264 239	250 *	5							

Figure S4. Identification and alignment of the conserved domain in each of the selected genes from both invasive and native species. We identified conserved domains for each gene: (a) *PAL* (Salicylic Acid), (b) *JAR1* (Jasmonic Acid), (c) *EIN3* (Ethylene). The upper figure represents the conserved domain, while the lower figure shows the respective domain peptide sequence aligned with related species using the NCBI domain database. We could not predict the conserved domain for *JAR1* due to the short number of amino acids from open reading frame.



(c) EIN3 (ET)



Figure S5. Multiple sequence alignments of predicted amino acid sequences of both *Alternanthera philoxeroides* and *A. sessilis* along with other closely related plants. (a) *PAL* (Salicylic Acid), (b) *JAR1* (Jasmonic Acid), (c) *EIN3* (Ethylene). The superfamily domain of each gene is highlighted with a red line. Star and colon symbols represent the conservation of amino acids and the dots and white space represent low conservation or mismatches respectively. Hyphens in the aligned sequences represent gaps in the sequence.



Figure S6. Endogenous hormone Salicylic acid (SA), jasmonic acid (JA), and ethylene (ET) contents in *Alternanthera philoxiroides* after infected with *Rhizoctonia solani*. (a) SA, (b) JA and (c) ET. Four-week-old plants were used for inoculations. Un-inoculated leaves were used as a control (0h). Samples were collected at different time intervals for quantification using ELISA. Values are means \pm SE (n = 4). Data bars with different letters indicate they are significantly different compared with the un-inoculated leaves (using Duncan's test at *P* < 0.05).



Figure S7. Sampling site of invasive *Alternanthera philoxeroides* and native *A. sessilis*. Samples were collected from the Fuzhou National Forest Park, Fuzhou, Fujian, China.

Table S1. Fold-change ratio of defense hormones and their responsive genes under different treatment conditions in invasive *Alternanthera philoxeroides* compared to native *A. sessilis*. Fold-change ratios are based on the relative amounts of signal molecules produced in *A. philoxiroides* and *A. sessilis* at the different time intervals. Fold-change ratios are calculated based on the comparative $2^{-\Delta\Delta CT}$ method normalized with the *Actin* gene at indicated time intervals after infection. Values highlighted in green show positive (higher) expression and red indicate negative (reduced) expression, compared with the control. Abbreviations: JA – Jasmonic acid, SA – Salicylic acid, ET – Ethylene, AP - *A. philoxeroides*, AS - *A. sessilis*.

Tu((Time (hpi) —		LOX	JA-J	AR1	JA-J	PR6	SA	-PAL	SA-	PR3	ET-l	EIN3
	Time (npi)	AP	AS	AP	AS	AP	AS	AP	AS	AP	AS	AP	AS
	6	<mark>2.71</mark>	<mark>2.90</mark>	<mark>1.28</mark>	<mark>1.12</mark>	<mark>1.09</mark>	<mark>1.64</mark>	0.53	<mark>1.13</mark>	0.06	<mark>0.13</mark>	<mark>0.92</mark>	<mark>0.68</mark>
	12	<mark>0.43</mark>	1.09	<mark>0.60</mark>	<mark>0.92</mark>	<mark>0.69</mark>	<mark>0.83</mark>	<mark>1.30</mark>	<mark>2.21</mark>	0.04	<mark>0.16</mark>	<mark>1.25</mark>	<mark>0.79</mark>
	24	<mark>1.94</mark>	<mark>0.45</mark>	<mark>0.64</mark>	<mark>1.20</mark>	<mark>0.87</mark>	<mark>2.00</mark>	<mark>3.24</mark>	<mark>1.74</mark>	<mark>3.30</mark>	<mark>11.36</mark>	<mark>1.02</mark>	<mark>0.60</mark>
Phizotonia coloni	48	<mark>0.79</mark>	<mark>1.89</mark>	<mark>0.47</mark>	<mark>1.59</mark>	<mark>0.60</mark>	<mark>2.32</mark>	<mark>8.81</mark>	<mark>3.02</mark>	10.24	<mark>10.62</mark>	<mark>1.67</mark>	<mark>0.98</mark>
Kni2ocioniu Soluni	72	1.04	<mark>0.61</mark>	<mark>0.55</mark>	<mark>1.16</mark>	<mark>0.71</mark>	<mark>1.92</mark>	<mark>6.86</mark>	<mark>6.59</mark>	<mark>7.56</mark>	<mark>13.99</mark>	<mark>1.20</mark>	<mark>0.61</mark>
	96	<mark>1.75</mark>	0.32	<mark>0.62</mark>	<mark>0.66</mark>	<mark>0.97</mark>	<mark>0.93</mark>	<mark>4.96</mark>	<mark>7.58</mark>	<mark>7.72</mark>	<mark>4.84</mark>	<mark>2.66</mark>	<mark>0.83</mark>
	6	<mark>0.27</mark>	<mark>4.15</mark>	<mark>0.32</mark>	<mark>1.90</mark>	<mark>0.27</mark>	<mark>0.80</mark>	0.21	<mark>5.11</mark>	<mark>4.01</mark>	<mark>1.91</mark>	<mark>0.49</mark>	<mark>9.88</mark>
	12	<mark>0.32</mark>	<mark>4.78</mark>	<mark>0.24</mark>	<mark>1.09</mark>	<mark>0.56</mark>	<mark>1.20</mark>	<mark>0.89</mark>	<mark>6.76</mark>	<mark>2.45</mark>	<mark>2.08</mark>	<mark>0.37</mark>	<mark>4.19</mark>
	24	<mark>0.40</mark>	1.88	<mark>0.64</mark>	<mark>1.97</mark>	<mark>0.57</mark>	<mark>1.32</mark>	<mark>0.37</mark>	<mark>3.21</mark>	<mark>6.95</mark>	10.54	<mark>0.66</mark>	<mark>1.93</mark>
MeJA pretreated before R. solani inoculations	48	<mark>0.05</mark>	<mark>2.04</mark>	<mark>0.23</mark>	<mark>0.91</mark>	<mark>0.90</mark>	<mark>1.82</mark>	1.97	10.65	<mark>75.56</mark>	<mark>2.04</mark>	<mark>0.83</mark>	<mark>3.35</mark>
	72	<mark>0.10</mark>	1.44	<mark>0.07</mark>	<mark>0.67</mark>	<mark>0.39</mark>	<mark>1.04</mark>	<mark>3.23</mark>	<mark>8.73</mark>	<mark>73.16</mark>	<mark>1.41</mark>	<mark>0.62</mark>	<mark>2.46</mark>
	96	<mark>0.08</mark>	1.47	<mark>0.17</mark>	<mark>0.61</mark>	<mark>0.66</mark>	<mark>0.92</mark>	1.03	<mark>6.88</mark>	<mark>56.92</mark>	<mark>4.25</mark>	<mark>0.47</mark>	<mark>2.52</mark>
	12	<mark>0.06</mark>	<mark>0.11</mark>	<mark>1.29</mark>	<mark>0.54</mark>	<mark>0.76</mark>	<mark>1.47</mark>	<mark>1.41</mark>	<mark>1.04</mark>	<mark>38.89</mark>	<mark>2.57</mark>	<mark>2.11</mark>	<mark>0.90</mark>
SA pretreated before R. solani inoculations	24	<mark>1.70</mark>	<mark>0.80</mark>	<mark>1.18</mark>	<mark>0.73</mark>	<mark>2.13</mark>	<mark>1.36</mark>	<mark>1.62</mark>	<mark>1.70</mark>	<mark>56.04</mark>	<mark>1.20</mark>	<mark>1.53</mark>	<mark>1.46</mark>
	48	<mark>0.63</mark>	0.02	<mark>1.13</mark>	<mark>0.71</mark>	<mark>2.53</mark>	<mark>0.49</mark>	<mark>1.25</mark>	<mark>3.44</mark>	<mark>98.83</mark>	<mark>1.88</mark>	<mark>1.44</mark>	<mark>1.62</mark>
	12	<mark>0.39</mark>	0.22	<mark>1.93</mark>	<mark>2.44</mark>	<mark>2.22</mark>	<mark>1.63</mark>	<mark>0.63</mark>	<mark>1.61</mark>	<mark>7.46</mark>	<mark>5.92</mark>	<mark>1.86</mark>	<mark>2.48</mark>
ET pretreated before R. solani inoculations	24	<mark>2.01</mark>	1.39	<mark>1.78</mark>	<mark>1.91</mark>	<mark>1.41</mark>	<mark>2.88</mark>	<mark>0.87</mark>	1.50	1.33	<mark>2.31</mark>	<mark>1.01</mark>	<mark>2.71</mark>
	48	<mark>3.01</mark>	1.37	<mark>1.63</mark>	<mark>1.50</mark>	<mark>1.37</mark>	<mark>1.48</mark>	<mark>1.80</mark>	<mark>0.61</mark>	<mark>13.32</mark>	<mark>0.25</mark>	<mark>1.24</mark>	<mark>2.26</mark>
	6	<mark>1.46</mark>	<mark>2.10</mark>	<mark>1.04</mark>	<mark>1.23</mark>	<mark>1.10</mark>	<mark>1.32</mark>	<mark>0.81</mark>	<mark>1.28</mark>	0.14	<mark>0.17</mark>	<mark>0.68</mark>	<mark>0.58</mark>
	12	<mark>0.78</mark>	1.11	<mark>0.60</mark>	<mark>0.68</mark>	<mark>0.80</mark>	<mark>0.49</mark>	<mark>0.83</mark>	1.46	0.02	<mark>0.04</mark>	<mark>0.76</mark>	0.37
	24	<mark>1.68</mark>	<mark>6.07</mark>	<mark>0.60</mark>	<mark>1.60</mark>	1.05	<mark>3.13</mark>	0 <mark>.77</mark>	<mark>2.87</mark>	0.03	0 <mark>.39</mark>	0 <mark>.54</mark>	1.15
Systemic logyos	48	<mark>1.56</mark>	<mark>3.37</mark>	<mark>0.86</mark>	<mark>1.49</mark>	<mark>1.33</mark>	1.65	<mark>1.33</mark>	<mark>1.28</mark>	0.18	<mark>0.24</mark>	0.58	<mark>1.06</mark>
Systemic leaves	72	<mark>2.03</mark>	<mark>3.01</mark>	<mark>0.95</mark>	<mark>1.64</mark>	<mark>1.08</mark>	<mark>1.44</mark>	1.45	<mark>1.69</mark>	0.38	<mark>0.32</mark>	<mark>0.54</mark>	1.02
	96	<mark>1.81</mark>	<mark>3.80</mark>	0.74	<mark>1.85</mark>	1.03	<mark>1.86</mark>	1.31	1.46	0.05	0.21	0.47	0.70

Gene	*Source	Primers (5' to 3')	Product size (bp)	Reference
PAL	A. philoxeroides &	F: 5'-TCTATGGATAATGCAAGGTTGG-3'	10(This starder
	A. sessilis	R: 5'-GGTTTGAGGGAAGACCATTG-3'	106	This study
PR3	A. philoxeroides &	F: 5'-ATCTCATGGAACTACAACTACGG-3'	110	This study
	A. sessilis	R: 5'-AGAAGCCTGTCTTGAAGGATATAA-3'	112	This study
LOX	A. philoxeroides	F: 5'-GGTCGCTGAAACAATCAATAAC-3'	96	This study
		R: 5'-TCTTGATAACAGCTGGGACATC-3'	00	This study
LOX	A annailia	F: 5'-GGTCGCTGAAACGATCAATAAC-3'	96	This study
	A. sessuis	R: 5'-TCTTGATAACAGCTGGGACATC-3'	00	This study
JAR1	A. philoxeroides &	F: 5'-CTACTCGAGCTGCTATGTCAAA-3'	06	This study
	A. sessilis	R: 5'-AGCCATACCATTTGCTTAAACTG-3'	90	This study
PR6	A. philoxeroides &	F: 5'-TGAAGCAAAGGTATGGGTGAA-3'	101	This study
	A. sessilis	R: 5'-CACCAAGGTCTGAAGGAGTAATAG-3'	101	This study
EIN3	A. philoxeroides &	F: 5'-TGCTGAAGATGATGGAGGTTT-3'	106	This study
	A. sessilis	R: 5'-CACCAGGCTCGAAGATTGT-3'	100	This study
β-Tubulin	A. philoxeroides & A.	F: 5'-CTACCTCTTTAGTGCTCATCTTACC-3'	106	This study
	sessilis	R: 5'-CAAATGTGGGATGCCAAGAAC-3'	100	This study
Elongation factor 1-	A. philoxeroides & A.	F: 5'-TTGCTTCTGACTCCAAGAATGA-3'	100	This study
alpha	sessilis	R: 5'-GCATAACCATTTCCAATCTGACC-3'	100	This study
Actin	A. philoxeroides, A.			This study &
Actin	sessilis &		104	Li & Smigocki,
	Beta vulgaris	N. 9 -GIGACIAACACCAICACCAGAG-3		2016

Table S2. Primers used for quantitative RT-qPCR. *A species-specific primer was used for the *LOX* gene due to difficulty in finding common sequences between species. For other genes, a single common primer to both species was used for expression analysis.

Plant	Hormone	Gene	Function	Size (bp)	GenBank Accession
A. philoxeroides	Colignitie A aid	DAI	CA biographosis	440	MK790145
A. sessilis	Sancyne Aciu	PAL	SA biosynthesis	436	MK790146
A. philoxeroides			A si li s shiting an	304	MK790153
A. sessilis	Sancync Acid	PK3	Acidic chitinase	303	MK790154
A. philoxeroides		LOV	IA bigging the sig	578	MK790155
A. sessilis	Jasmonic Acid	LOX	JA biosyntnesis	613	MK790156
A. philoxeroides	Learnin Asid	IAD1	Cataluzas the sumthasis of several IA amide conjugates	305	MK790147
A. sessilis	Jasmonic Acia	JAKI	Catalyzes the synthesis of several JA-amido conjugates	307	MK790148
A. philoxeroides		חחר	D 111.		MK790149
A. sessilis	Jasmonic Acid	PKb	Proteinase inhibitor	172	MK790150
A. philoxeroides	Ethylopo		A transprintion factor and positive regulator of ET regranding gang compaging	525	MK790151
A. sessilis	Ethylene	EIN3	A transcription factor and positive regulator of E1-responsive gene expression	527	MK790152

Table S3. List of putative defense hormones and responsive genes isolated in both Alternanthera philoxeroides and A. sessilis.

Table S4. Sequence information of six defense hormones and responsive genes isolated from *Alternanthera philoxeroides* and *A. sessilis*. See Supplementary Table S2 for additional information regarding each isolated gene. Selected ORF (Open Reading Frame) indicates six-reading frames of each isolated gene sequence. Abbreviations: AP - *A. philoxeroides*, AS - *A. sessilis*, CDS - Coding DNA Sequence.

	_	Salicylic Aci			Jasmonio	c Acid			Ethylene			
Predictions	PA	4L	P.	R3	LC	OX	JAR1		PR6		EIN3	
	AP	AS	AP	AS	AP	AS	AP	AS	AP	AS	AP	AS
Nucleotide (bp)	451	448	305	306	307	622	310	311	175	174	535	537
Selected ORF	+2	+3	+1	+3	+1	+2	-3	+1	-2	-1	+3	+2
CDS length (bp)	418	414	165	303	282	270	132	123	147	147	447	480
Peptide length (aa)	139	138	55	101	94	90	44	41	49	49	149	160
Similarity (Gene	Phenylalanir	ne ammonia-	EP3	-like	Lipoxyger	nase 6 gene	JAR1-like		No significant		Ethylene insensitive 3-like	
family)	lya	lyase protein		fan	family			simi	larity	protein		

Table S5. Functional analysis of three hormone genes in both *Alternanthera philoxeroides* and *A. sessilis*. Assembled contigs and annotated gene sequences were used for predicting protein isoelectric point (<u>http://bioinformatics.org/sms2/protein_iep.html</u>) and protein molecular weight (<u>http://bioinformatics.org/sms2/protein_mw.html</u>). We could not predict the gene from the assembled contigs of *JAR1* from *A. sessilis*. Abbreviations: AP - *A. philoxeroides*, AS - *A. sessilis*.

Destistions	PAL	(SA)	JAR1 (JA)		EIN3	6 (ET)
Fredictions	AP	AS	AP	AS	AP	AS
Contig length (bp)	1411	1286	1284	1206	675	1698
Number of Exons	2	1	1	-	1	1
mRNA length (bp)	1080	993	945	-	654	1464
No. of amino acids	360	331	314	-	218	488
Isoelectric Point (pH)	7.37	6.68	8.04	-	9.34	6.34
Protein molecular weight (kDa)	39.01	35.67	35.29	-	24.98	54.95
No. of motifs	5	5	3	-	2	3
	PLN02457 superfamily	PLN02457 superfamily	GH3 superfamily		EIN3	EIN3
Representing domain	(PAL)	(PAL)	domain	-	superfamily	superfamily
	(26 to 360 aa)	(1 to 331 aa)	(1 to 293 aa)		(15 to 218 aa)	(27 to 275 aa)

Table S6. Screening and isolation of defense hormones and responsive gene sequences from both invasive *Alternanthera philoxeroides* and native *A. sessilis*. The highlighted green color in the table are the sequences of genes that were successfully isolated (see Supplementary Table S3 above).

S. No	Gene name and function	Primer Name	Primers (5' to 3')	Primer Length (bp)	Tm (°C)	GC (%)	Product size (bp)
	Salicylic Acid regula	ted signaling pat	hway genes				
1	NDR1 NDR1 (Non race-specific Disease Resistance 1); a plasma membrane protein, for CC-NBS-LRR subset of R genes.	AmNDR1-F AmNDR1-R	F: 5'-TGCTAGAGGGGTAAAATGGGA-3' R: 5'- ACCCAAAACACCCAACAACATAA-3'	21 23	58.15 59.22	47.62 39.13	250
2	EDS1 PAD4 (PhytoAlexin Deficient 4); lipase-like. Act upstream of SA in basal resistance to biotrophic pathogens & ETI initiated by TIR-NBS-LRR subset of R genes.	AmEDS1-F AmEDS1-R	F: 5'-GTGGAATGAGGTAGCCCAGA-3' R: 5'-CGATCCAGTGTCCTCGTTCT-3'	20 20	58.80 59.19	55 55	577
3	NPR1 NPR1 (Nonexpresser of PR genes 1); transcription cofactor. Positive regulator of SA and negative regulator of JA.	AmNPR1-F AmNPR1-R	F: 5'-ACACAGAGCTTTGGAATCCG-3' R: 5'-ATGCACAACCGATCCTTTGG-3'	20 20	58.19 58.82	50 50	375
4	PR1 Pathogenesis related protein 1	AmPR1-F AmPR1-R	F: 5'-TCATCCTTGCATCAACCCCT-3' R: 5'-AGCCTGTCGTCCCAAGTAAA-3'	20 20	59.00 58.95	50 50	328
5	<i>PAL</i> (see Table S2) Phenylalanine Ammonia Lyase.	AmPAL-F AmPAL-R	F: 5'-TCACCCAGGACAGATTGAGG-3' R: 5'-GCTCGGAACAGTAAGAAGCC-3'	20 20	58.72 58.64	55 55	<mark>476</mark>
6	PR2 PR-2 (endo-1,3-beta-glucanase)	AmPR2-F AmPR2-R	F: 5'-CGGCCATGCAAAATGTCCAA-3' R: 5'-GCCGCATACACAGTGTCTAC-3'	20 20	60.04 58.72	50 55	331
7	<i>PR3</i> (see Table S2) PR3-Acidic chitinase.	AmPR3-F AmPR3-R	F: 5'-TCCATGCAACCCAAGTAAGC-3' R: 5'-GGTTATTCCCTGGCGAAACG-3'	<mark>20</mark> 20	58.46 59.27	50 55	<mark>332</mark>
8	PR5 PR-5 (thaumatin-like)	AmPR5-F AmPR5-R	F: 5'-CCATGGTTTTCAACGCTCCA-3' R: 5'-GTTAAACCCGTCGACAAGGC-3'	20 20	59.04 59.49	50 55	212
	Jasmonic Acid regula	ted signaling pat	hway genes				
1	JAR1 (see Table S2) (JAsmonate Resistant 1); jasmonate-amino synthetase; a member of the GH3 family. JAR1 enzyme that produces the JA-Ile conjugate.	AmJAR1-F AmJAR1-R	F: 5'-TGCAGTCCTGATGAAGTGGT-3' R: 5'-GCATTCGGGAAAAGCTCAGG-3'	20 20	58.94 59.55	50 55	335
2	CO11 (COronatine Insensitive 1); ubiquitin-protein ligase, required for jasmonate-regulated defense.	AmCOI1-F AmCOI1-R	F: 5'-AGGTTAAAGCGGCTCAGGAT-3' R: 5'-GCCCGACATAACTGAGACCT-3'	20 20	59.09 59.18	50 55	352
3	MYC2 JIN1 (Jasmonate INsensitive 1); MYC2; TF & a positive regulator of JA-responsive genes such as VSP2 and LOX2, whereas it acts as a negative regulator of JA/ET-responsive genes such as PDF1.2.	AmMYC2-F AmMYC2-R	F: 5'-CGAGTTCGCCTTGTAATCGG-3' R: 5'-ATATCTCCCCTGATTCCGGC-3'	20 20	59.08 58.72	55 55	513
<mark>4</mark>	LOX (see Table S2)	AmLOX-F	F: 5'-TTCCGCGTGATGAAGCTTTT-3'	<mark>20</mark>	58.77	<mark>45</mark>	<mark>470</mark>

	JA-mediated defense pathway marker gene.	AmLOX-R	R: 5'-AATGGCCTCTTCGACACTCA-3'	<mark>20</mark>	<mark>59.02</mark>	<mark>50</mark>	
5	AOS Allene Oxide Synthase 1, chloroplastic.	AmAOS-F AmAOS-R	F: 5'-GTTCTCTCCTACCTCGACCC-3' R: 5'-TTCGTCACGTGAGATCCCAA-3'	20 20	58.61 59.04	60 55	471
6	PR6 (see Table S2) PR6 proteinase inhibitor.	AmPR6-F AmPR6-R	F: 5'-CTGGAGTTTGCAAGGGTTGT-3' R: 5'-AGCACCAAGGTCTGAAGGAG-3'	20 20	58.60 59.31	50 55	204
7	PR9 Peroxidase	AmPR9-F AmPR9-R	F: 5'-TTGTCAGGGGTTGTGATGGA-3' R: 5'-AATTGCTTGAGAAACCGGCA-3'	20 20	58.86 58.67	50 45	302
	Ethylene regulated	l signaling pathw	vay genes				
1	ACC synthase Aminocyclopropane-1-carboxylic acid synthase is an enzyme that catalyses the synthesis of 1- Aminocyclopropane-1-carboxylic acid (ACC), a precursor for ethylene, from S-Adenosyl methionine.	AmACC-F AmACC-R	F: 5′-AGAACAGGGGTGGAGCTTTT-3′ R: 5′-CCAAACACTGTGGCTCCATA-3′	20 20	59.15 57.80	50 50	254
2	EIN3 (see Table S2) EIN3 (Ethylene INsensitive 3); transcription factor. A positive regulator of ET-responsive gene expression.	AmEIN3-F AmEIN3-R	F: 5'-GCGAGTGGTGGAAAGACAAG-3' R: 5'-GGACACTCACTTTCCATGCC-3'	20 20	59.13 58.83	55 55	351
3	ERF1 AP2/ERF and B3 domain-containing transcription factor. A TF & positive regulator of JA and ET signaling.	AmERF1-F AmERF1-R	F: 5'-AAATGGTCATTGGGGTGCAC-3' R: 5'-GTCAGTGGCATAGTGGGCTT-3'	20 20	59.02 60.04	50 55	205
4	<i>ERF2</i> Ethylene-responsive transcription factor ERF024. A TF & positive regulator of JA and ET signaling.	AmERF2-F AmERF2-R	F: 5'-GGGAAGTGGGTCTCGGAAAT-3' R: 5'-AGCTGCCTGAATGTCCCTT-3'	20 19	59.38 58.92	55 52.63	204
5	<i>PDF1.2-A</i> Defensin-like protein AX1. ET/JA-mediated defense pathway marker gene.	AmPDF1.2A- F AmPDF1.2A- R	F: 5'-TGAGATGAACATGGTGGTTGAG- 3' R: 5'-ACGGGCACACAGATTCCAAT-3'	22 20	58.32 59.96	45.45 50.00	150
6	<i>PDF1.2-B</i> Defensin Ec-AMP-D2-like. ET/JA-mediated defense pathway marker gene.	AmPDF1.2B- F AmPDF1.2B- R	F: 5'-CAACTGCAGAGGTAGGACCA-3' R: 5'-TGTTTAGTGCAGAAGCATCGAC- 3'	20 22	59.02 59.26	55.00 45.45	172