

Supplementary Material

Detection of *bar* transgenic sugarcane with a rapid and visual loop-mediated isothermal amplification assay

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1. Supplementary Figures and Tables

1.1 Supplementary Figures

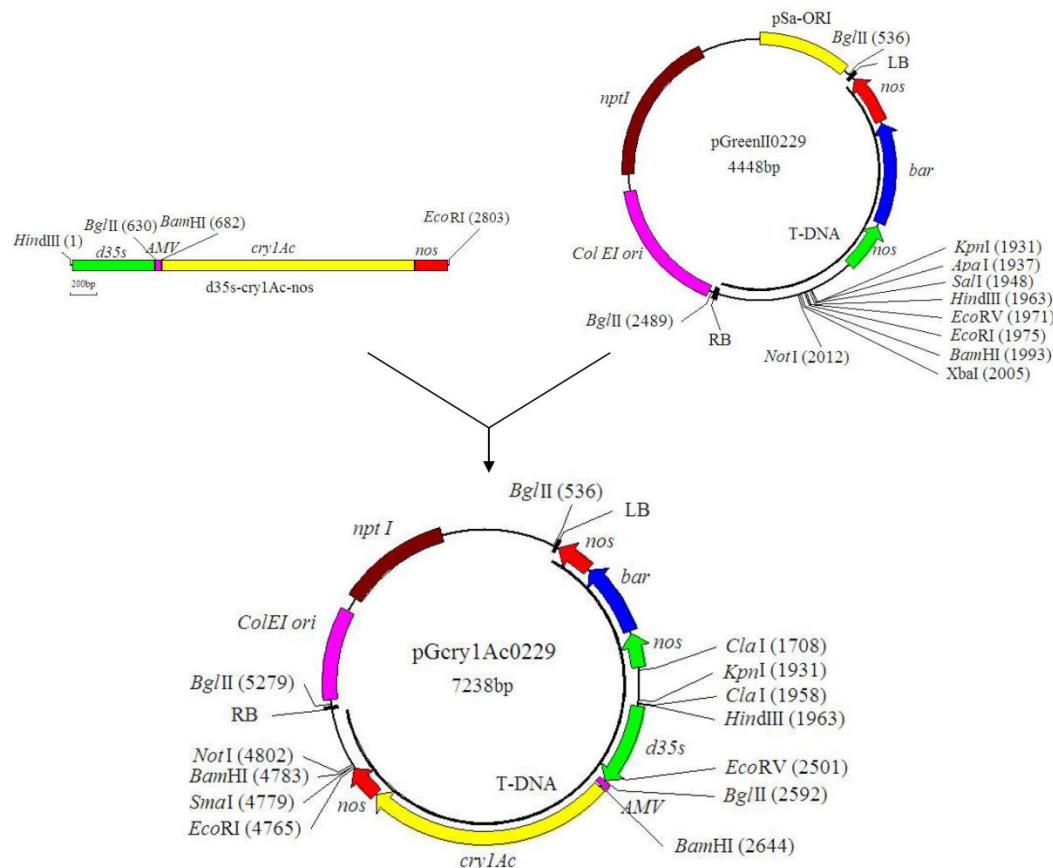


Figure S1. Construction of the plasmid pGcry1Ac0229 (1Ac0229).

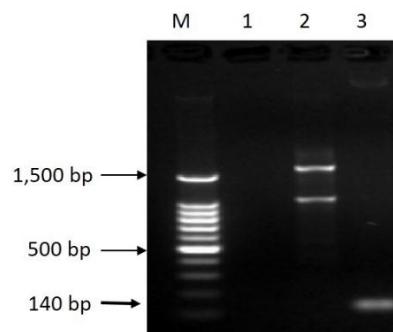


Figure S2 Agarose gel electrophoresis of the plasmid 1Ac0229 PCR products. Lane M: 100 bp DNA ladder. Lanes 1: ddH₂O (blank control). Lanes 2: FN15 PCR products (negative control). Lanes 3: the plasmid 1Ac0229 PCR products.

1 TCAGATTCG GTGACGGGCA GGACCGGAGC GGGCGGCACC GGCAGGCTGA AGTCCAGCTG
 61 F3 → CCAGAAACCC ACGTCATGCC AGTTCCCGTG CTTGAAGCCG GCCGCCCGCA GCATGCCACG
 121 ← GGGGGCATAT CCGAGCGCCT CGTGCATGCG CACGCTCGGG TCGTTGGCA GCCCCGATGAC
 181 AGC_GACCACG CTCTTGAAGC CCTGTGCCTC CAGGGACTTC AGCAGGTGGG TGTAGAGCGT
 241 ← LB B2 B3
 241 ← GGAGCCCAGT CCCGTCCGCT GGTGGCGGGG GGAGACGTAC ACGGTTGACT CGGCCGTCCA
 301 GTCGTAGGCG TTGCGTGCCT TCCAGGGACC CGCGTAGGCG ATGCCGGCGA CCTCGCCGTC
 361 CACCTCGCGC ACGAGGCCAGG GATAGCGCTC CCGCAGACGG ACGAGGTCGT CGGTCCACTC
 421 CTGCGGTTCC TGCGGCTCGG TACGGAAGTT GACCGTGCTT GTCTGGATGT AGTGGTTGAC
 481 GATGGTGCAG ACCGCCGGCA TGTCCGCCTC GGTGGCACGG CGGATGTCGG CGGGCGTCG
 541 TTCTGGGCTC AT

Figure S3. LAMP primers and their positions in *bar* gene (GenBank: EU048867.1) sequence.

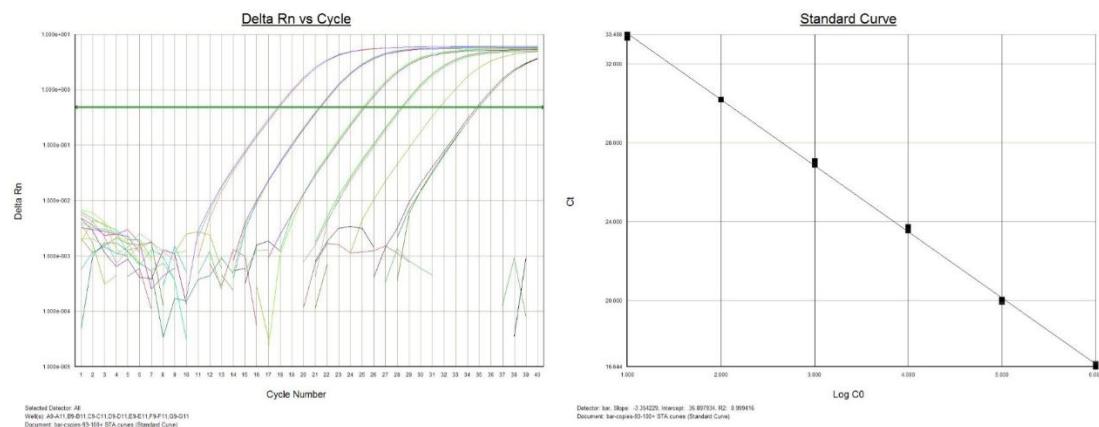


Figure S4. The amplification curves and standard curves obtained by quantitative SYBR Green real-time PCR with the primer pair of *bar* gene. In the performed quantitative SYBR Green real-time PCR assays, the standard curve formula is $y = -3.354x + 36.898$, coefficient of determination ($R^2 = 0.999$) and amplification efficiency ($E = 0.99$).

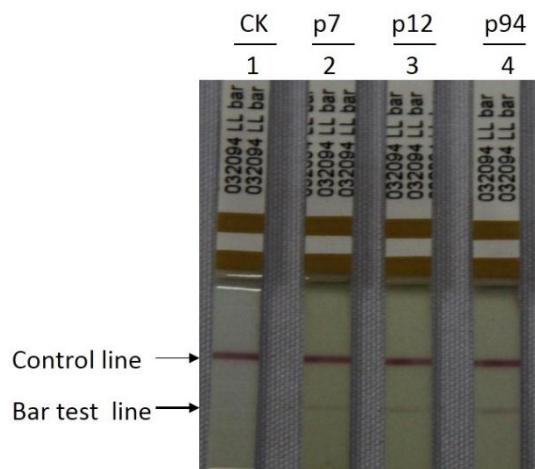


Figure S5. Bar protein expression in putative *bar* transgenic sugarcane by quick test strip kit detection.

1.2 Supplementary Tables

Table S1. The sequence information of LAMP primers used in this experiment.

Primer	Position	Primer sequences (5'- 3')
F3	61-79	CCAGAAACCCACGTCTATGC
B3	224-241	CACGCTCTACACCCACCT
FIP (F1c+F2)	127-145 86-101	GCACGAGGCCTCGGATAT- CCGTGCTTGAAGCCGG
BIP (B1c+B2)	162-180 205-222	CGTTGGCAGCCGATGAC- CTGAAGTCCTGGAGGCA
LF	106-120	CGTGGCATGCTGCGG
LB	183-200	CGACCACGCTTTGAAGC

Table S2. Estimation of copy number of *bar* gene in transgenic sugarcane by quantitative SYBR Green real-time PCR.

Lines	Copy number of <i>bar</i> in single cell (Mean ±St)	Lines	Copy number of <i>bar</i> in single cell (Mean ±St)	Lines	Copy number of <i>bar</i> in single cell (Mean ±St)
p1	1.82±0.04	p35	7.39±0.54	p69	6.99±0.25
p2	1.15±0.02	p36	7.25±0.57	p70	6.49±0.15
p3	1.94±0.02	p37	4.82±0.12	p71	7.47±0.17
p4	3.41±0.11	p38	7.13±0.09	p72	5.99±0.11
p5	4.16±0.09	p39	5.86±0.09	p73	8.23±0.17
p6	2.82±0.07	p40	8.25±0.19	p74	7.33±0.33
p7	0.35±0.00	p41	7.20±0.10	p75	7.98±0.03
p8	1.83±0.06	p42	8.47±0.77	p76	8.04±0.20
p9	1.57±0.04	p43	7.37±0.51	p77	9.07±0.12
p10	1.49±0.01	p44	8.94±0.75	p78	5.50±0.09
p11	0.56±0.00	p45	7.77±0.16	p79	14.15±0.17
p12	0.20±0.00	p46	8.90±0.11	p80	11.81±0.15
p13	5.89±0.08	p47	6.63±0.20	p81	13.18±0.04
p14	1.29±0.12	p48	6.06±0.02	p82	18.54±0.11
p15	1.34±0.02	p49	8.34±0.17	p83	13.67±0.50
p16	1.90±0.01	p50	9.24±0.17	p84	16.72±0.55
p17	0.71±0.03	p51	6.98±0.16	p85	11.83±0.11
p18	0.83±0.06	p52	6.76±0.25	p86	11.17±0.05
p19	8.75±0.36	p53	10.08±0.22	p87	13.08±0.25
p20	0.65±0.01	p54	9.01±0.12	p88	11.62±0.10
p21	4.30±0.08	p55	5.49±0.11	p89	8.58±0.42
p22	3.60±0.05	p56	6.22±0.28	p90	11.44±0.28
p23	4.09±0.08	p57	7.57±0.23	p91	13.84±0.50
p24	3.91±0.07	p58	7.74±0.40	p92	16.90±0.39
p25	7.88±0.25	p59	9.78±0.29	p93	17.31±0.81
p26	4.72±0.36	p60	8.89±0.51	p94	0.53±0.01
p27	4.40±0.13	p61	9.41±0.85	p95	17.01±0.75
p28	3.52±0.13	p62	9.94±0.17	p96	17.91±0.28
p29	3.35±0.03	p63	7.17±0.12	p97	16.36±0.73
p30	2.74±0.07	p64	9.51±0.34	p98	12.38±0.20
p31	4.41±0.17	p65	10.29±0.33	p99	14.27±0.64
p32	10.33±0.13	p66	8.45±0.48	p100	13.70±0.12
p33	11.52±0.18	p67	7.10±0.46	FN15(CK)	0.02±0.00
p34	9.68±0.40	p68	5.51±0.17		