Supporting information------

Versatile antibody-sensing Boolean logic for simultaneous detection of multiple bacterial toxins

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Content list

Experimental Section

B. cereus toxin production

The reference and mutant strains of *B. cereus* used in the current study are listed in Table S1. The standard medium for *B. cereus* growth was casein hydrolysat-yeast medium plus 1% glucose (CGY) according to the previous publication (1).

Monoclonal antibodies

Nine monoclonal antibodies (mAbs) specifically recognizing different components of the toxins or protein marker from *B. cereus* are involved in the present work. mAbs 1A8 for NheA, 1C2, 1E11 and 2B11 for NheB, and 3D6 for NheC; 8B12 for L2, 1E9 for L1 and 1E8 for B in the Hbl complex; 3C6 for a protein marker of cereulide producing by *B. cereus* strains (Table S2).

ELISA and single tube assay

Single antibody and sandwich type immunoassays in microtiter plates were performed as described earlier (1). For the single tube assay, 0.2 mL thin walled PCR tubes with flat cap were used. For the antibody-based OR gates, all primary mAbs were mixed together and applied simultaneously, the secondary antibody-horseradish peroxidase (HRP) conjugate was added subsequently after a washing step.

Tables

Toxins		Nhe complex			Cer	Hbl complex			References
		A	B	C		L2	L1	B	
B. cereus (MHI No.)	1491	+	+	+					*1, 2*
	165	+	+	+	+				*3*
	1505	+	+	+		+	+	+	*1*
	3016	+	+	+	+	+	+	+	*4*
	1489	+	+	+					*This study*
	2970	+	+	+					*This study*
	3038	+	+	+					*This study*
	3086	+	+	+					*This study*

Table S1 *B. cereus* strains used in the current study. The NheB components of MHI 1489, 2970, 3038 and 3086 contain point mutations in the range of amino acid residues 122 to 150.

		Nhe complex			Cer	Hbl complex			References
		A	B	C		L2	L1	B	
Antibodies	1A8	+							*5*
	1E11		+						*5*
	2B11		+						*5*
	1C2		+						*5*
	3D6			+					*6*
	3C6				+				*This study*
	8B12					+			*7*
	1E9						+		*8*
	1B8							+	*8*

Table S2 Monoclonal antibodies used for the detection of different components from *B. cereus* strains.

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Figures

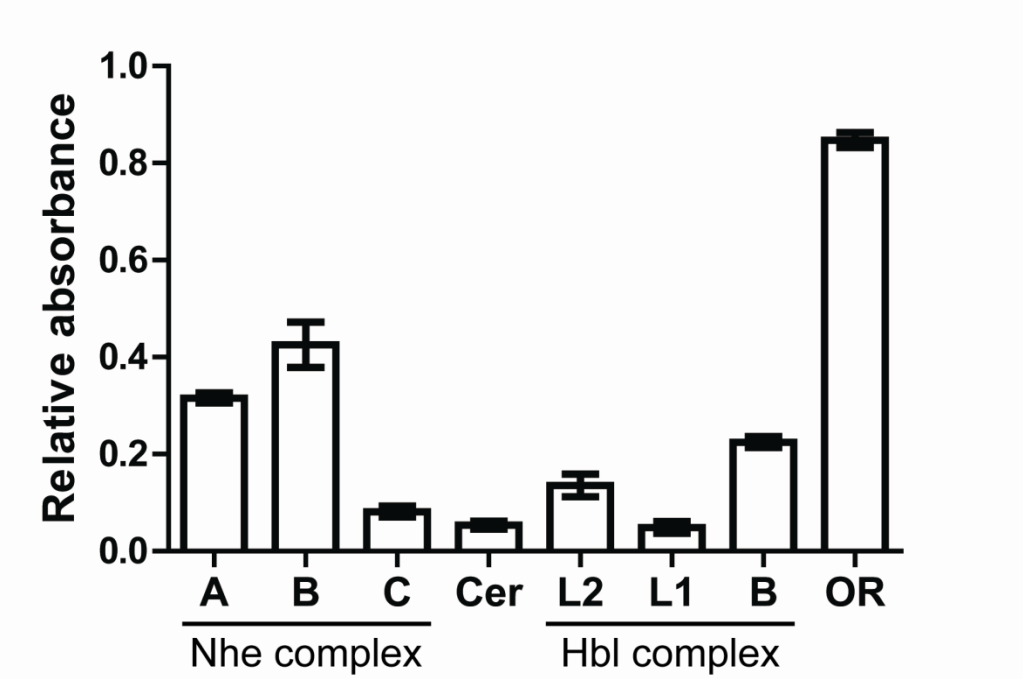

Figure S1 Sensitivity of the antibody-based OR gate in an ELISA microtiter plate.

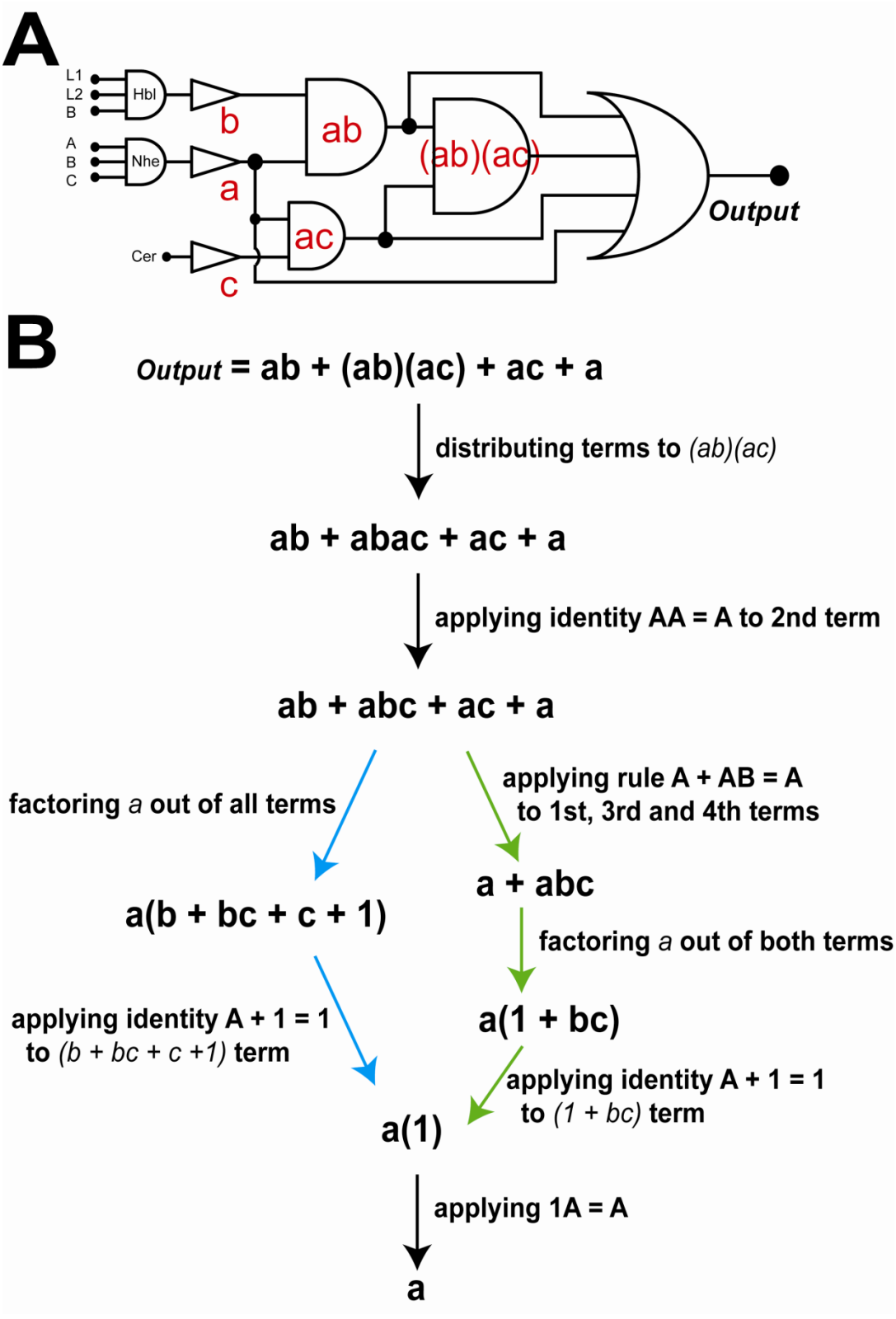

Figure S2 Proofs of the Boolean rule of *simplification*. (A) The Boolean expressions for the toxin profiles of the *B. cereus* strains; (B) The proofs of *simplification* using two methods.

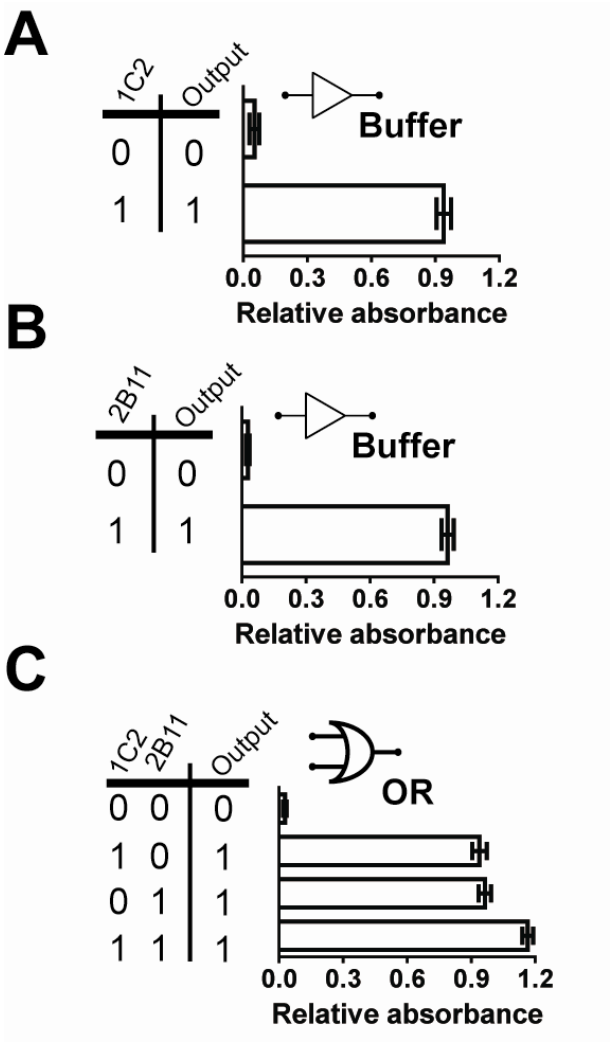

Figure S3 Single antibody model for constructing different logic gates. Two Buffer gates using mAbs 1C2 (A) and 2B11 (B), and an OR gate using mAbs 1C2 and 2B11 in the same assay(C).

References

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