Supporting Information

CRISPR/Cas13a-based single-nucleotide polymorphism detection for reliable determination of ABO blood group genotypes

Hongjuan Wei[1], Liyan Liu[1], Hanji Jiang, Hong Chen, Yunxiang Wang, Yongjun Han, Zhen Rong*, Shengqi Wang*

Bioinformatics Center of AMMS, Beijing 100850, P. R. China

[1] These authors contributed equally to this work

Corresponding Author

*(S.Q. Wang) E-mail: sqwang@bmi.ac.cn

*(Z. Rong) E-mail: rongzhen0525@sina.com

Table S1. The sequences of Amplicons, PCR primers and crRNAs

DNA	Detail of sequences 5`-3`
Amplicon of Exon 6-261	**GTCGCATTTGCCTCTGGTTG**GTTTCCCGGGGAAGGGCGGCTGCCTCTGG AAGGGTGGTCAGAGGAGGCAGAAGCTGAGTGGAGTTTCCAGGTGGGGGC GGCCGTGTGCCAGAGGCGCATGTGGGTGGCACCCTGCCAGCTCCATGTGA CCGCACGCCTCTCTCCATGTGCAGTAGGAAGGATGTCCTCGTGGTNACCC CTTGGCTGGCTCCCATTGTCTGGGAGGGCACATTCAACATCGACATCCTC AACGAGCAGTTCAGGCTCCAGAACACCACCATTGGGTTAACTGTGTTTGC CATCAAGAAGTAAGTCAGTGAGGTGGCCGAGGGTAGAGACCCAGGCAGT GGCGAGTGACTGTGGACATTGAGGTCTCTCCTTG**TGTTCAAGACAGAGT GGGGTG**
ABO-6-T7-F1	**GAAATTAATACGACTCACTATAGGGGTCGCATTTGCCTCTGGTTG**
ABO-6-T7-F2	GAAATTAATACGACTCACTATAGGGGGCAGAAGCTGAGTGGAGTT
ABO-6-T7-F3	GAAATTAATACGACTCACTATAGGGTCCAATGTTGAGGGAGGGCT
ABO-6-T7-F4	GAAATTAATACGACTCACTATAGGGGGGAATGATTTGCCCGGTTG
ABO-6-T7-F5	GAAATTAATACGACTCACTATAGGGGTTCCCGCAGGTCCAATGTT
ABO-6-R1	**CACCCCACTCTGTCTTGAACA**
ABO-6-R2	AATGTCCACAGTCACTCGCC
ABO-6-R3	CCCAATGGTGGTGTTCTGGA
ABO-6-R4	GAGCCTGAACTGCTCGTTGA
ABO-6-R5	ACTGCTCGTTGAGGATGTCG
6-261-G1	GTGACCCCTTGGCTGGCTCCCATTGTCT
6-261-G2	**ATGTCCTCGTGGTGACCCCTTGGCTGGC**
6-261-G3	GCAGTAGGAAGGATGTCCTCGTGGTGAC
6-261-D1	GT|ACCCCTTGGCTGGCTCCCATTGTCTG
6-261-D2	GATGTCCTCGTGGT|ACCCCTTGGCTGGC
6-261-D3	**TGCAGTAGGAAGGATGTCCTCGTGGT|AC**
Amplicon of Exon 7-796	**CCGTGTCCACTACTATGTCTTCACC**GACCAGCNGGCCGCGGTGCCCCGC GTGACGCTGGGGACCGGTCGGCAGCTGTCAGTGCTGGAGGTGNGCGCCT ACAAGCGCTGGCAGGACGTGTCCATGCGCCGCATGGAGATGATCAGTGA CTTCTGCGAGCGGCGCTTCCTCAGCGAGGTGGATTACCTGGTGTGCGTGG ACGTGGACATGGAGNTCCGCGACCACGTGGGCGTGGAGATCCTGACTCC GCTGTTCGGCACCCTGCACCCCGGCTTCTACGGAAGCAGCCGGGAGGCCT TCACCTACGAGCGCCGGCCCCAGTCCCAGGCCTACATCCCCAAGGACGAG GGCGATTTCTACTACNTGGGGGNGTTCTTCGGGGGGTCGGTGCAAGAGGT GCAGCGGCTCACCAGGGCCTGCCACCAGGCCATGATGGTCGACCAGGCC AACGGCATCGAGGCCGTGTGGCACGACGAGAGCCACCTGAACAAGTACC TGCTGCGCCACAAACCCACCAAGGTGCT**CTCCCCCGAGTACTTGTGG**
ABO-7-T7-F1	**GAAATTAATACGACTCACTATAGGGCCGTGTCCACTACTATGTCTTCACC**
ABO-7-T7-F2	GAAATTAATACGACTCACTATAGGGCCTGCCTTGCAGATACGTGG
ABO-7-T7-F3	GAAATTAATACGACTCACTATAGGGTCTAAGCCTTCCAATGGCCG
ABO-7-T7-F4	GAAATTAATACGACTCACTATAGGGGATGAAGTGAATCGCAGCCC
ABO-7-T7-F5	GAAATTAATACGACTCACTATAGGGCCTTCACCTACGAGCGCC
ABO-7-T7-F6	GAAATTAATACGACTCACTATAGGGTGCTGCTCTAAGCCTTCCAAT
ABO-7-T7-F7	GAAATTAATACGACTCACTATAGGGGGGCGTGGAGATCCTGACT
ABO-7-T7-F8	GAAATTAATACGACTCACTATAGGGATCCTGACTCCGCTGTTCG

ABO-7-R1	**CCACAAGTACTCGGGGGAG**
ABO-7-R2	TTGTTCAGGTGGCTCTCGTC
ABO-7-R3	AGCACCTTGGTGGGTTTGTG
ABO-7-R4	ACAACAGGACGGACAAAGGAAACAG
ABO-7-R5	ACTTGTTCAGGTGGCTCTCG
ABO-7-R6	CACAAGTACTCGGGGGAGAG
ABO-7-R7	CACAACAGGACGGACAAAGGA
ABO-7-R8	TTCTGCTAAAACCAAGGGCG
7-796-C1	**CAAGGACGAGGGCGATTTCTACTACCTG**
7-796-C2	CAAGGACGAGGGCGATTTCTACTTCCTG
7-796-C3	AGGACGAGGGCGATTTCTACTACCTGGG
7-796-A1	CAAGGACGAGGGCGATTTCTACTACATG
7-796-A2	CAAGGACGAGGGCGATTTCTACTTCATG
7-796-A3	**AGGACGAGGGCGATTTCTACTACATGGG**
FAM-Reporter	FAM-UUUUU-BHQ1

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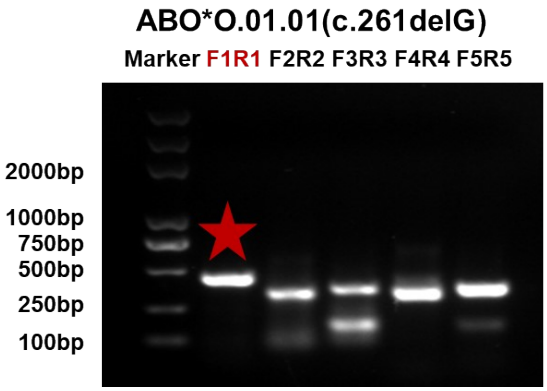

Figure S1. Five pairs of PCR primers were screened for ABO*O.01.01(c.261delG) locus, the best of which was F1R1.

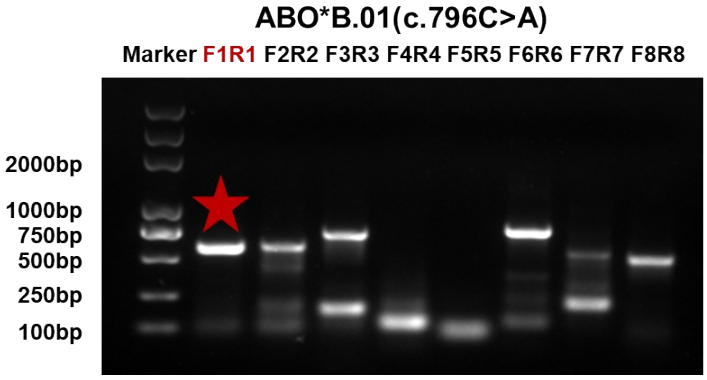

Figure S2. Eight pairs of PCR primers were screened for ABO*B.01(c.796C>A) locus, the best of which was F1R1.

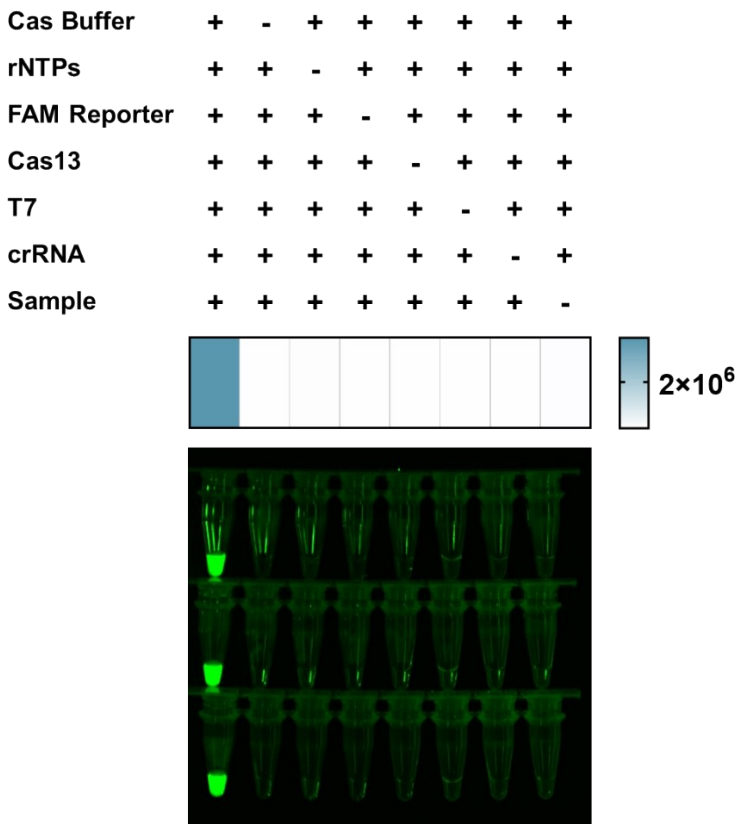

Figure S3. Verification of the effect of each component of the Cas13 trans-cleavage activity showed that each component is indispensable.

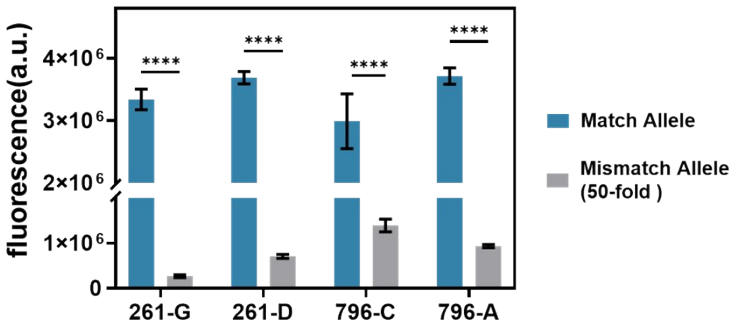

Figure S4. The discrimination capacity validation experiment. The four screened crRNAs demonstrated good discrimination capacity in samples with matched sequences and 50-fold concentrations of mismatched sequences with SNP.

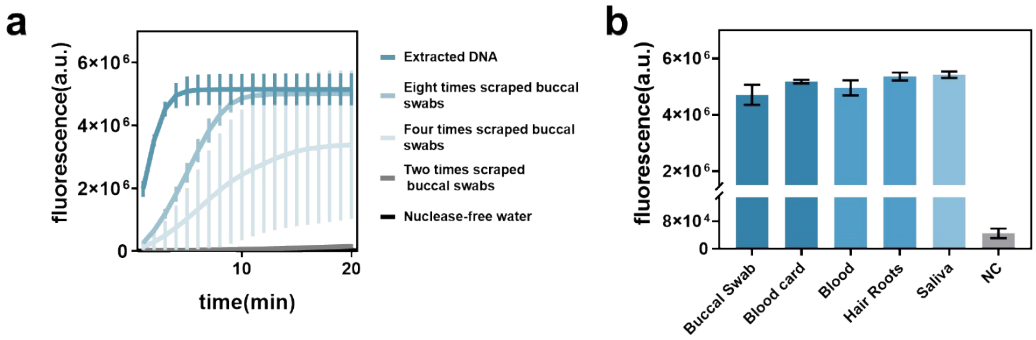

Figure S5. Extraction-free sample pretreatment was tested. (a) There is no significant difference between positive control (extracted DNA) and directly treated buccal swabs when scraped times outnumber eight. (b) Different samples including buccal swabs, blood cards, peripheral blood, hair roots, and saliva tested show the adaptivity of the method.

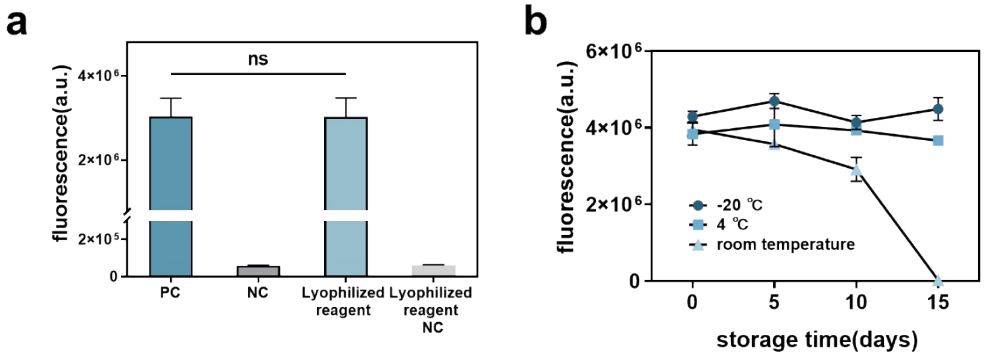

Figure S6. (a) Lyophilization reagent techniques were tested, which is also compatible with this assay. (b) The end-point fluorescence intensity at 20 minutes using the lyophilized reagents stored at －20 °C, 4 °C, and room temperature for 0, 5, 10, and 15 days.

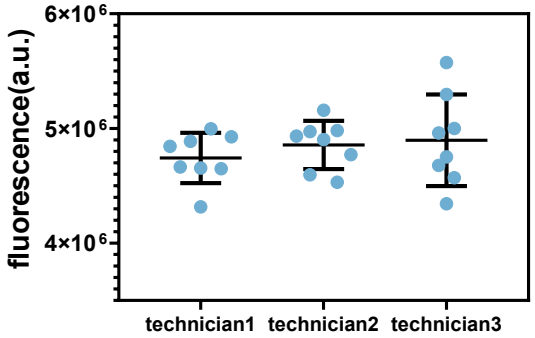

Figure S7. The reproducibility of our method was validated by three different lab technicians conducting eight experiments.