Supporting Information

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Forward and reverse blood typing principles

By cross-testing the RBCs and serum from different person's blood, Landsteiner found out that the blood of two people under contact agglutinates, which was due to contact of blood with blood serum. As a result he stated the law to describe the relationship between antigens on the red blood cells and antibodies in the serum for ABO blood system: 1. If an agglutinogen is present in the red cells of a blood, the corresponding agglutinin must be absent from the plasma; 2. If an agglutinogen is absent in the red cells of a blood, the corresponding agglutinin must be present in the plasma. This law and the determination a blood group is illustrated in Figure S1.

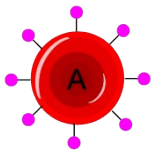
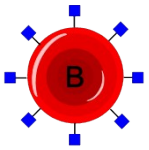
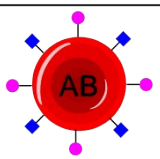
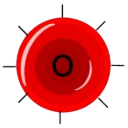
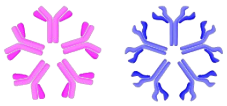
Blood groups	Group A	Group B	Group AB	Group O
Antigens on the RBCs	 Antigen A	 Antigen B	 Antigen A and B	 None
Antibodies in the plasma	 Anti-B	 Anti-A	None	 Anti-A and Anti-B

Figure S1. The determination of ABO blood groups followed by Landsteiner's law.

According to Landsteiner's law, the typing of ABO blood groups relies on the determination of the presence or absence of certain antigens on the surface of red blood cells (RBCs), as well as certain antibodies in the serum of the blood. The blood typing process which is based on antigen detection is referred to as the forward blood typing, whereas the blood typing through antibody detection is the method of reverse blood typing. The typing method for determining an individual's blood type is shown in Figure S2, and the result confirming principle can be found in Table 1, where the positive sign "+" means the observation of the agglutination while the negative sign "-" refers to the absence

of the agglutination reaction. It shows that the results determined by forward and reverse blood typing follow different principles.

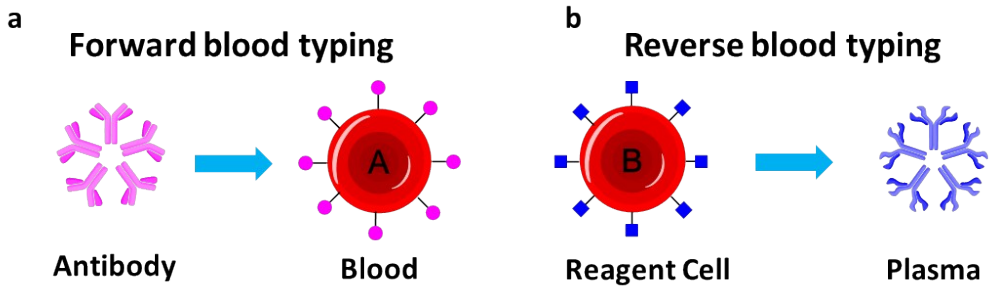
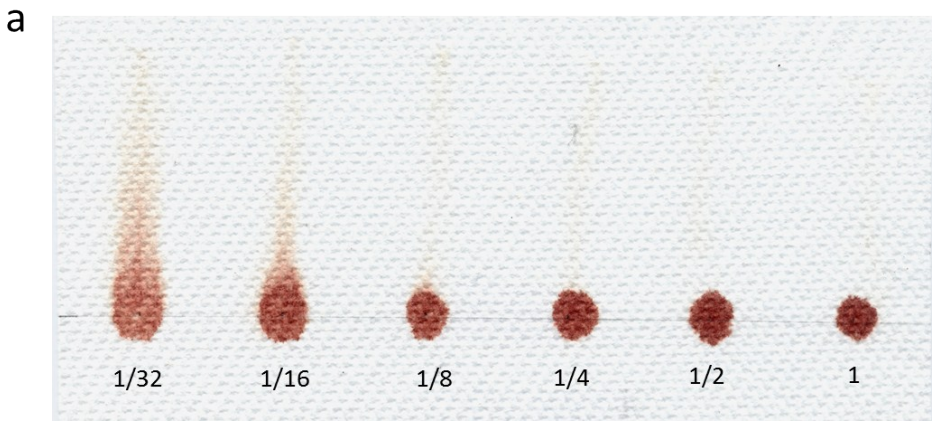
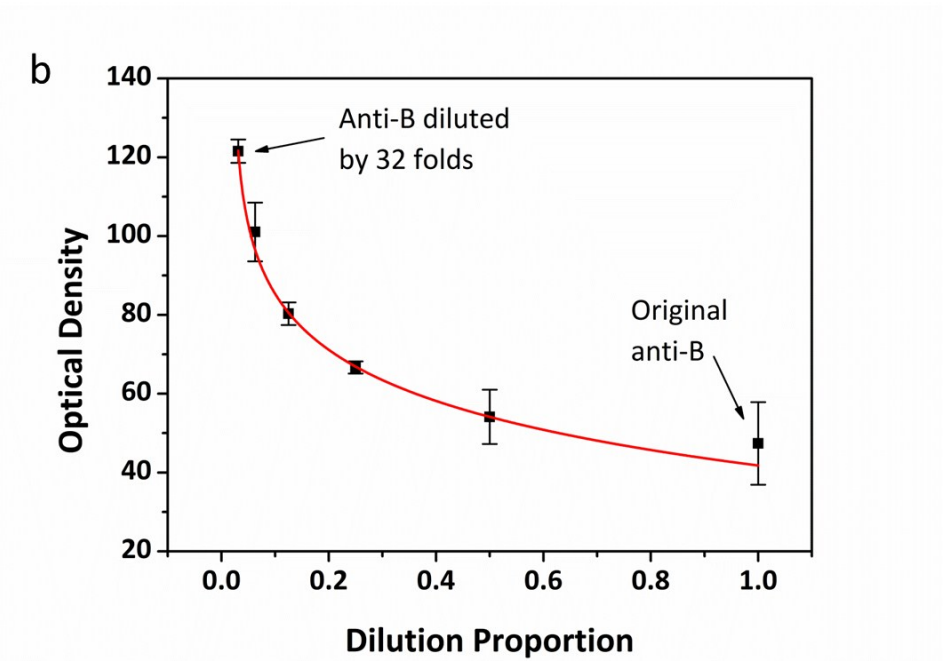


Figure S2. a) Commercial antibodies are using to test the presence of the corresponding antigens on RBCs for forward blood typing; b) Reagent RBCs with known antigens are determining the antibodies in the serum for reverse blood typing.

Antibody dissolution from the plastic slides

Following the procedure for studying the antibody A dissolution behaviour from the plastic substrate(Fig. 3 and Fig. 5), the dissolution behaviour of antibodies B and D have also been quantified. The calibration curve of anti-B dilution showed that anti-B lost its activity after a dilution of 1/16, indicating a weaker activity compared with anti-A (Fig. S3a). The calibration curve was fitted with formula (2) (Fig. S3b), which quantitatively showed that the concentration of anti-B in the first saline wash was 23.1% of its original concentration CB (Table 2), The more efficient dissolution of anti-B by saline solution than of anti-A confirms that anti-B can be dissolved more easily from the plastic slide, therefore anti-B weakened more rapidly than anti-A with the number of washes by saline (Fig. S3c). As shown in Figure S3c, anti-B could sustain three washes and the residual anti-B on the plastic substrate still had sufficient activities for unambiguous blood typing.




b
Optical Density
Dilution Proportion
Anti-B diluted
by 32 folds
Original
anti-B
140
120
100
80
60
40
20
0.0
0.2
0.4
0.6
0.8
1.0

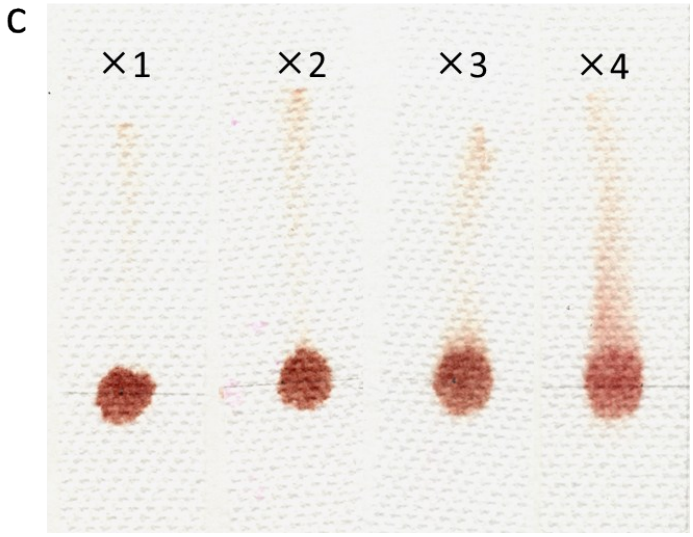
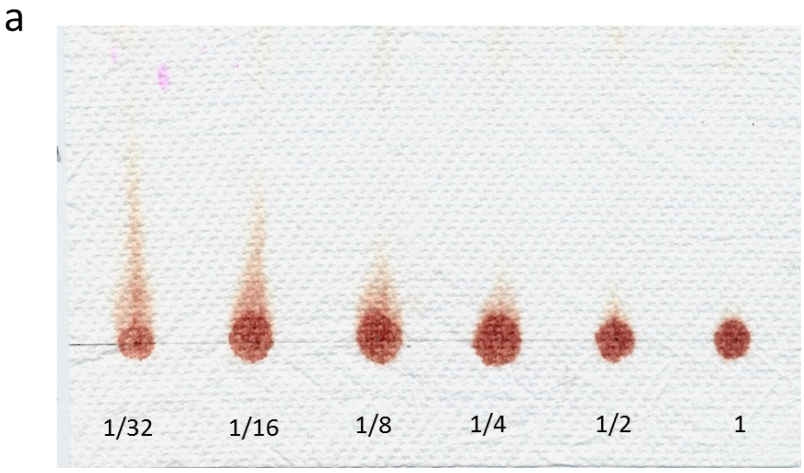
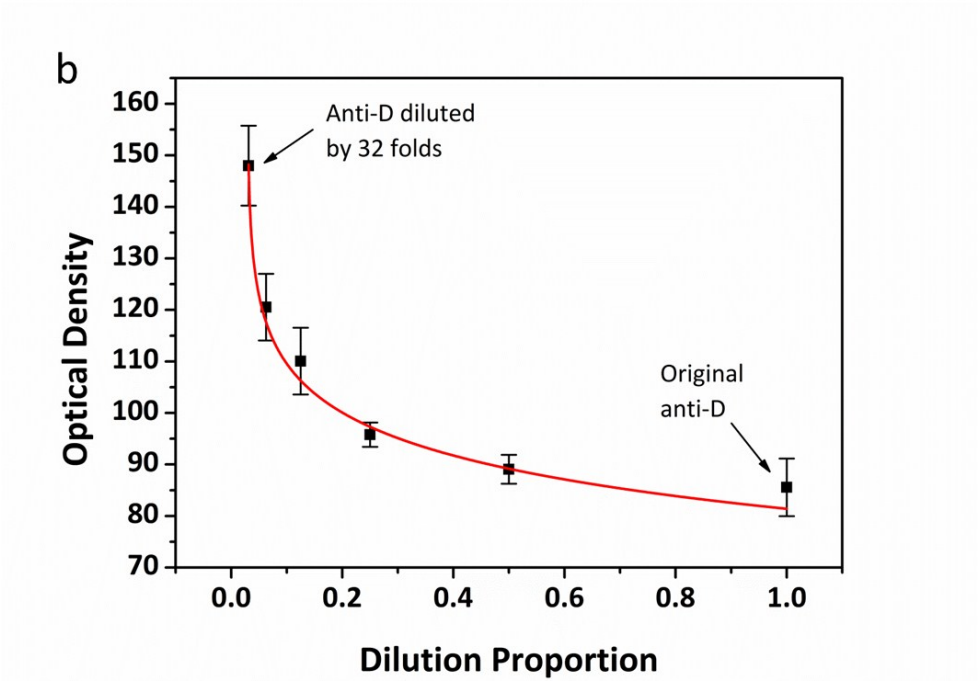


Fig. S3. Antibody dissolution behaviour study for anti-B: (a) anti-B activity as a function of series of dilution, tested by the agglutination colour density of the B reagent red cell (concentrated from the commercial reagent to a hematocrit level of 45%); (b) the standard curve of the B cell colour density as a function of anti-B dilution factor; (c) anti-B dissolution behaviour of five consecutive PBS dissoluiton rinses of the plastic device.

However, as for anti-D, the dissolution was even more efficient (Fig. S4a and S4b), our measurement showed that 92.0% of its original concentration was dissolved (Formula (3) and Table 2) and removed from the plastic substrate in the first saline wash; anti-D lost its activity at 1/8 dilution (Fig. S4c).





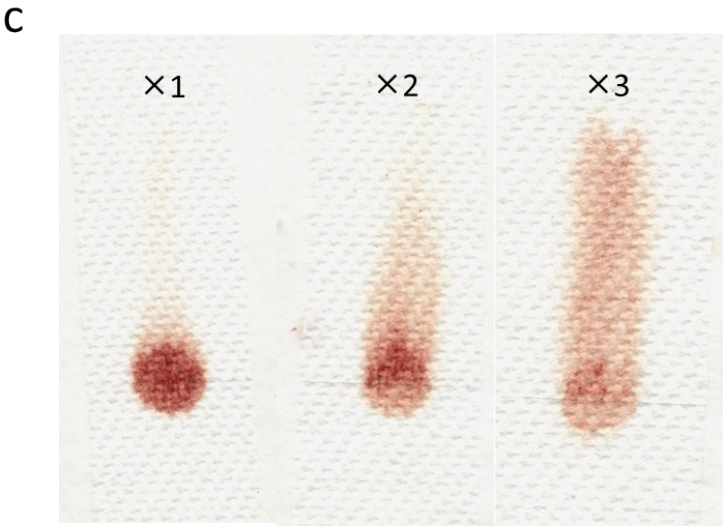


Fig. S4. Antibody dissolution behaviour study for anti-D: (a) anti-D activity as a function of series of dilution, tested by the agglutination colour density of the C reagent red cell (concentrated from the commercial reagent to a hematocrit level of 45%); (b) the standard curve of the C cell colour density as a function of anti-D dilution factor; (c) anti-D dissolution behaviour of five consecutive PBS dissoluiton rinses of the plastic device.