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TRANSFUSION

ONE-PAGE REPORT OF NEW ALLELES OR ANTIGENS

Blood Group Genomics

Identification of a novel c.506A>C variant on the *ABO*A1.02* allele in a Chinese individual

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1 | BACKGROUND

Identification of the ABO blood group is crucial for ensuring the safety of blood transfusion and transplantation. Discrepancies in serologic forward and reverse typing are often indicative of ABO subtypes. In 1990, Yamamoto et al. made a significant contribution by cloning and elucidating the *ABO* gene,[1] enabling genotype analysis of the ABO blood group. Combined serologic and genotypic testing can lead to a deeper understanding of how ABO subtypes are formed at the molecular level. This report presents a case of A_w subtype, in which the genotype was identified using third-generation long-read sequencing. This subtype is characterized by a new allele with c.506A>C (rs number not available in dbSNP) variant on the *ABO*A1.02* for one haplotype and a hybrid allele of *ABO*O.01.02-ABO*A1.01* for the other haplotype.

2 | BRIEF METHODS

The proband in this study was a 29-year-old female who visited a general outpatient clinic. Her informed consent was obtained to collect 3–5 mL of EDTA-anticoagulated whole blood. The study was approved by the Ethics Committee of the First Affiliated Hospital of Zhengzhou University (2023-KY-0870-003). Serologic tests were performed using the tube method, and the following reagents were used: anti-A, anti-B (Beijing Jinhao), anti-A_1, anti-H (Sanquin), A_1c, Bc, and Oc (Changchun Bioxun), and A_2c (Immucor). DNA extraction was carried out using the Blood Genome Kit (Beijing TIANGEN) to obtain the genomic DNA of the proband. The extracted sample was then sent to Xi'an Haorui Gene Technologies Ltd for third-generation long-read sequencing, and the protocol used for sequencing was based on the method established by Wang et al.[2] The International Society of Blood Transfusion (ISBT)-recommended NG_006669 was used as the reference genomic sequence for comparison. To understand the effect of amino acid substitutions on glycosyltransferase A (GTA) structure and function, PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2/) was also queried.

Abbreviations: GTA, glycosyltransferase A; ISBT, International Society of Blood Transfusion.

3 | RESULTS AND DISCUSSION

Serological analysis (Table 1) revealed weak expression of A antigen (1+) in forward typing, with weak anti-A_1 antibody (1+) detected in reverse typing, suggesting that the proband has an A_w subtype. Third-generation long-read sequencing results demonstrated that one haplotype of the proband exhibited c.467C>T, c.506A>C compared to *A1.01*. These variants formed a new *ABO* allele, which has been assigned GenBank accession number OR538568. The other haplotype of the proband displayed c.106G>T, c.188G>A, c.189C>T, c.220C>T, c.261delG, and c.297A>G. Based on the presence of the c.261delG variant, it is inferred that the haplotype belongs to the

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TABLE 1 Results of serologic grouping and *ABO* gene analysis.

ABO blood group typing								*ABO* genotype and variant	
Anti-A	Anti-A_1	Anti-B	Anti-H	A_1c	A_2c	Bc	Oc	Haplotype 1	Haplotype 2
1+	0	0	4+	1+	0	4+	0	*ABO*AW.new* (c.467C>T; c.506A>C)	*ABO*O.01.02-ABO*A1.01* (c.106G>T; c.188G>A; c.189C>T; c.220C>T; c.261delG; c.297A>G)

O.01.XX group. The allelic variations observed in exons 1–6 corresponded to the *O.01.02* allele, whereas exon 7 matched either the *A1.01* or *O.01.01* allele, indicating a recombination event between exons 6 and 7. Third-generation long-read sequencing pinpointed the potential recombination breakpoint to a region extending from nucleotide 1014 in intron 6 to nucleotide 645 in exon 7, spanning a sequence length of 310 base pairs. This hybrid allele was initially reported by Suzuki and colleagues,[3] who provided sequencing details for exons 6 and 7, as well as intron 6, however, sequence data for exons 1–5 were not included in their report. Additionally, this allele was not found in Genbank, Erythrogene, and ISBT *ABO* allele database. The c.467C>T variant (p.Pro156Leu) is a common polymorphic site in the Chinese population, and it does not significantly affect the structure and activity of the GTA enzyme.[4] However, the p.Gln169Pro caused by c.506A>C is located in the Rossmann fold region, which is close to the N-terminal domain and primarily associated with sugar donor binding.[5] As predicted by PolyPhen-2 ("probably damaging" with a score of 0.997 by HumDiv and 0.984 by HumVar), the substitution is probably to impair protein function due to its impact on the local structure of the Rossmann fold region, which likely reduces the recognition of the UDP-GalNAc donor substrate by the GTA enzyme. As a result, it is hypothesized that p.Gln169Pro ultimately leads to a decrease in GTA enzyme activity.

4 | BRIEF SUMMARY

Our results suggest that c.467C>T and c.506A>C form a novel *ABO* allele, and that the c.506A>C variant results in an alteration of p.Gln169Pro in the Rossmann fold region, which may be responsible for the reduced activity of GTA synthesized by the *AW.new* allele.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

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REFERENCES

1. Yamamoto F, Clausen H, White T, Marken J, Hakomori SI. Molecular genetic basis of the histo-blood group ABO system. Nature. 1990;345:229–33.
2. Wang C, Tang Y, Zhang P, Xiong L, Chen W, Lv X. Detection and phenotype analysis of a novel Ael blood group allele. Vox Sang. 2024;119:74–8.
3. Suzuki K, Iwata M, Tsuji H, Takagi T, Tamura A, Ishimoto G, et al. A de novo recombination in the ABO blood group gene and evidence for the occurence of recombination products. Hum Genet. 1997;99:454–61.
4. Yamamoto F, McNeill PD, Hakomori S. Human histo-blood group A2 transferase coded by A2 allele, one of the A subtypes, is characterized by a single base deletion in the coding sequence, which results in an additional domain at the carboxyl terminal. Biochem Biophys Res Commun. 1992;187:366–74.
5. Patenaude SI, Seto NO, Borisova SN, Szpacenko A, Marcus SL, Palcic MM, et al. The structural basis for specificity in human ABO(H) blood group biosynthesis. Nat Struct Biol. 2002;9:685–90.

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