Human ABO gene transcriptional regulation

Yoshihiko Kominato, Rie Sano, Yoichiro Takahashi, Akira Hayakawa, and Kenichi Ogasawara

The ABO blood group system, discovered by Karl Landsteiner in 1900, is fundamental to the safety of blood transfusion, which requires identification of weak phenotypes or subgroups. The system is composed of two carbohydrate antigens, A and B, and their antibodies. The International Society of Blood Transfusion designates four antigens—A; B; AB; and A1—based on the specificity of the antibodies. Biochemical and molecular genetic studies have clarified the molecular basis of the histo-blood group ABO system. The functional A and B alleles at the ABO genetic locus encode two glycosyltransferases, α-1-3-N-acetylgalactosaminyltransferase (A-transferase) and α-1-3-galactosyltransferase (B-transferase), respectively. Initial molecular genetic studies demonstrated that ABO is composed of seven exons spanning approximately 19.5 kb of genomic DNA (Fig. 1A) and that two critical single-base substitutions in the last coding exon result in amino acid substitutions responsible for the different donor nucleotide-sugar substrate specificity between the A- and B-transferases. A single-base deletion in Exon 6 is considered to shift the reading frame of codons and to abolish the transferase activity of A-transferase in most O alleles.

On the other hand, some aspects remain to be explored. The ABO antigens are expressed in a cell type-specific manner; the isoantigens A, B, and H of blood groups A, B, and O are not confined to red blood cells (RBCs) but are also found in most secretions and on some epithelial cells. However, they are absent in connective tissue, muscle, and the central nervous system. Moreover, ABH antigens are known to undergo drastic changes during the development, differentiation, and maturation of cells in the epithelial and erythroid lineages. For example, studies of A-antigen expression during the maturation of erythroid progenitors in a two-phase liquid culture system showed that A-positive cells gradually increased during erythroid maturation. Fluorescence-activated cell sorting analysis with monoclonal antibodies has demonstrated the expression of A-antigens on colony cells derived from blast-forming units–erythroid and colony-forming units–erythroid. In addition to these physiological processes, profound changes have also been documented in pathological conditions such as tumorigenesis. Reduction or complete deletion of A- or B-antigen expression in bladder and oral cancers has been documented. Moreover, the loss of ABH antigens has been correlated with the progression of various cancers, including lung and bladder carcinomas. Finally, since Yamamoto and colleagues clarified the molecular genetic basis of the ABO system, a number of weak phenotypes have been found to be attributable to single nucleotide polymorphisms in the coding exons and splicing sites and hybrid formation between common alleles, although other weak phenotypes for which no variant apparently exists in the coding exons and splicing sites have also been reported. Therefore, to understand the molecular mechanism responsible for the control of ABO gene expression in a cell type-specific manner, during normal cell differentiation, in cancer cells lacking A or B antigens, and in weak phenotypes, it is essential to grasp how ABO gene transcription is regulated.

The DNA sequences in and around specific genes provide the code that dictates when, where, and at what level specific genes are transcribed. This code comprises three parts: the core promoter where RNA production initiates and is directed toward Exon 2, the region proximal to the transcriptional start site. This is the fundamental code that dictates the efficiency and specificity of ABO gene transcription in a cell type as well as the physiological processes.

ABBREVIATIONS: ABOi = ABO-incompatible; AML = acute myeloid leukemia; CGIs = CpG islands; ChIP = chromatin immunoprecipitation; DHSs = DNase I-hypersensitive sites; EMSAs = electrophoretic mobility shift assays; HGVs = Human Genome Variation Society; MDS = myelodysplastic syndrome; SSP = single specific primer; TFs = transcription factors; vWF = von Willebrand factor.

From the 4Department of Legal Medicine, Gunma University Graduate School of Medicine, Maebashi, and the 5Japanese Red Cross Central Blood Institute, Tokyo, Japan.

Address reprint requests to: Yoshihiko Kominato, Department of Legal Medicine, Gunma University Graduate School of Medicine, Maebashi, 371-8511 Japan; e-mail: kominato@gunma-u.ac.jp.

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the core promoter, and the more distant regulatory sequences that enhance RNA production. It has become obvious that enhancers usually work in groups (i.e., the locus control region and super-enhancers), each being bound by several transcription factors (TFs), forming a so-called enhanceosome. These enhanceosomes are nucleated

![Diagram of transcriptional regulation of ABO expression](image-url)
by pioneer TFs early during differentiation, and these TFs are subsequently replaced by other TFs that trigger recruitment of the preinitiation complex, involving RNA polymerase II, to the promoter. Enhancers also interact with each other through a multiloop structure. Thus, for elucidation of ABO regulation, it is important to reveal regulatory regions such as the core promoter, the region proximal to it, and the more distant enhancer, as well as the TFs that bind to those regions.

Although A- or B-antigen expression is dependent upon many steps including the structure of ABO, transcriptional regulation of ABO, translational regulation, modification or localization of A- or B-transferase, and H antigen expression, in this review we focus on the transcriptional regulation of the ABO gene through regulatory regions and TFs, and outline the molecular basis for weak phenotypes with variants in those regions.

REGULATORY REGIONS FOR ABO EXPRESSION

ABO gene regulatory regions have been identified by in vitro studies and genetic studies of weak phenotypes. The variants are described according to the HGVS nomenclature using the nucleotide sequences of accession numbers NG_006669.1 and NM_020469.1 as a reference in the genetic study sections. The relationship between the descriptions of variants in Intron 1 according to the Human Genome Variation Society (HGVS) nomenclature and those in the original reports is shown in Table 1. However, the positions that were used in the in vitro experiments described in the original papers remain unchanged. The positions reported in the original papers have been described according to the nucleotide sequences of accession number NT_035014.4 as a reference.

The proximal promoter of ABO

In vitro studies

Initially, Yamamoto et al. demonstrated two transcription initiation sites upstream of the ATG translation start site in ABO using human pancreatic cDNA. Consistently, similar transcription initiation sites were found upstream of the translation start site using erythroid cells differentiated in vitro from AC133 CD34+ cells and K562 cells by us and others. For demonstrating the proximal promoter of ABO, we carried out transfection experiments into gastric cancer KATOIII cells and erythroleukemia HEL cells using luciferase reporter plasmids prepared from a genomic clone of human ABO. Those experiments defined the proximal ABO promoter between −150 and −2 relative to the translation start site in those cells (Figs. 1A and 2), and the promoter showed constitutive activity regardless of the cell types examined. Electrophoretic mobility shift assays (EMSA) demonstrated that the GC box at −56 to −44 in the promoter bound a ubiquitous TF Sp1 or Sp1-like protein(s) (Fig. 1A), whereas mutations of the recognition motif that abrogated binding of those factors reduced the promoter activity in both cell types. Thus, Sp1 or Sp1-like protein(s) seemed to be important for proximal promoter activity. The nucleotide sequence in ABO reveals two CpG islands (CGIs) (Fig. 1A), one extending from the immediate 5’ flanking region through the first exon and into Intron 1, and the other extending from Intron 6 to Exon 7. Thus, the ABO proximal promoter is located within a CGI. Because a promoter within a CGI would include a few transcription initiation sites, the presence of several transcription initiation sites in ABO is relevant.

Genetic studies

Cai et al. reported a nucleotide deletion between −35 and −18, i.e., c.−35_−18del, in the proximal ABO promoter in the B3 phenotype (Fig. 2), which reduced the promoter activity in a plasmid-based reporter assay. Similarly, three single-nucleotide substitutions at −77, −76, or −68 in the ABO proximal promoter increased the promoter activity in the B3 phenotype (Fig. 2), which is consistent with the above findings.

### TABLE 1. Relationship between the descriptions of variants in Intron 1 according to the HGVS nomenclature and those in the original reports

| Variants according to HGVS nomenclature | Variants in the original reports |
|----------------------------------------|---------------------------------|
| c.28 + 4077_7107del                    | +4105_ + 7136del†               |
| c.28 + 5110_10898del                   | +5137_ + 10914del†              |
| c.28 + 5443_11354del                   | c.28 + 5443_29−1655del†         |
| c.28 + 5859G > C                       | c.28 + 5830G > C†               |
| c.28 + 5861 T > G                     | +5890 T > G†                    |
| c.28 + 5864G > A                       | +5893G > A†                     |
| c.28 + 5865_5887del                    | +5892_ + 5914del†               |
| c.28 + 5875C > T                       | +5904C > T†                     |
| c.28 + 5880A > G                       | +5909A > G†                     |
| c.28 + 5885C > T                       | c.28 + 5885C > T†               |

* The original reports are listed in the Reference section. The specific reference numbers are shown in the Reference column.
† Variants were described according to the reference sequence of NG_006669.1. It is likely that c.28 + 5443_11354del is easier to indicate the deletion size than c.28 + 5443_29−1655del.
‡ Variant was described according to the reference sequence of NT_035014.4.
§ Variant was described according to the reference sequence of NG_006669.1.
promoter, i.e., c.−77C > G, c.−76G > C, or c.−68G > T, respectively, have been reported in A3 and B3, each substitution reducing the promoter activity in luciferase reporter assays. Recently, another single-nucleotide substitution at −72 in the ABO promoter, i.e., c.−72G > A, was reported in B3. These genetic variants confirm the functional significance of the proximal ABO promoter in vivo.

Conclusion
ABO transcription is likely regulated by the proximal promoter and cell-specific regulatory regions (Fig. 1B), because the proximal promoter shows constitutive activity regardless of cell type.

Regulatory region for erythroid cell–specific expression of ABO
For delineation of distal regulatory regions involved in ABO regulation in erythroid cells, we used both in vitro experiments and genetic approaches.

In vitro studies
First, because DNase I–hypersensitive sites (DHSs) are associated with transcriptional regulatory regions including the promoter and distal enhancer, we prepared luciferase reporter plasmids on the basis of DHSs within a 15-kb region of genomic DNA in and around ABO in erythroleukemia K562 cells with publicly available data from DNase-Seq and FAIRE-seq on the University of California, Santa Cruz (UCSC) Genome Browser. Subsequent plasmid-based reporter assays demonstrated a distal regulatory region between +5653 and +6154 relative to the translation start site, named the +5.8-kb site (Fig. 1A), in K562 cells, and the regulatory activity of this region was specific to erythroid cells. According to the HGVS nomenclature, the +5.8-kb site corresponds to c.28 + 5624_6125. The same site was referred to the GATA binding site by the others. EMSAs and chromatin immunoprecipitation (ChiP) assays demonstrated that the region bound hematopoietic TF GATA-1 or -2, and RUNX1 (Fig. 1A), whereas mutations of the recognition motifs that abrogated binding of those factors reduced the regulatory activities of the +5.8-kb site in K562 cells. Thus, binding of GATA-1 or -2, and RUNX1 to the +5.8-kb site seemed to be crucial for the erythroid cell–specific activity of the region (Fig. 1B).

Genetic and serologic studies
Comparison of the genomic DNA in the ABO gene between humans and these primate species demonstrated high conservation between the ATG translation start codon and the stop codon of the ABO gene, except for a few regions. The +5.8-kb site is conserved among humans, chimpanzees, and orangutans, showing similar expression of the A and B antigens on RBCs. However, it is intriguing to note that the site is not conserved in rhesus monkeys and marmosets, in which the A and B antigens are expressed only slightly on RBCs. Therefore, a comparative approach could indicate involvement of the site in ABO expression in human erythroid lineage cells.

Fig. 2. Nucleotide sequence of the 5′-flanking region in ABO. The sequence is given in full, from position −260 to +40, relative to the translation start ATG site of ABO. The upper case letters denote the coding sequence of Exon 1, and the lower case letters the noncoding genomic sequence. High arrows above the sequence indicate the transcription initiation sites that were determined by 5′-RACE using human pancreas cDNA by Yamamoto et al., and low arrows denote the transcription initiation sites that were determined by in vitro erythroid culture of AC133−CD34+ cells. The proximal ABO promoter is located between −150 and −2 relative to the ATG translation start site. The recognition motif for TF Sp1 is indicated by an overbar. Nucleotide substitutions at −77, −76, −72, and −68 are indicated in red, and the deletion between −35 and −18 is denoted by an underbar. [Color figure can be viewed at wileyonlinelibrary.com]
phenotype, termed $B^{\text{m}}5.8$ and $B^{\text{m}}3.0$, respectively (Fig. 1C). These deletions involved the +5.8-kb site. The $B^{\text{m}}$ phenotype is characterized by the discrepancy of $B$ antigen expression between RBCs and secretions: $B^{\text{m}}$ RBCs are not agglutinated by anti-$B$ or anti-$A,B$ antibody, whereas the saliva of $B^{\text{m}}$ secretors contains about as much $B$ substance as that of a normal $B$ secretor. However, the $B$ antigens on RBCs can only be detected by sensitive techniques such as adsorption and elution of anti-$B$. $B^{\text{m}}$ erythrocytes contain abundant $H$ sites, which can be converted into $B$ sites by in vitro treatment with $B$-transferase derived from normal $B$ individuals. $B$-transferase activity was detected in serum of $B^{\text{m}}$ individuals, although the activity was distinctly reduced in all cases. The $B^{\text{m}}$ trait is inherited as a rare allele at the $ABO$ locus, although a few non-hereditary cases have also been reported. Thus, deletion of the erythroid cell–specific regulatory region or the +5.8-kb site on the $B^{\text{m}}$ allele could explain the discrepancy of $B$-antigen expression between RBCs and secretions in $B^{\text{m}}$. Further genetic studies found variants in the GATA motif of c.28 + 5861 $T > G$ or c.28 + 5859$G > C$ in $B^{\text{m}}$ or $A_{\text{mo}}$, respectively (Fig. 3). The former was termed $B^{\text{m}}GAGA$. $A_{\text{mo}}$ is analogous to $B^{\text{m}}$, in blood group A. Moreover, deletion of the RUNX1 binding motif of c.28 + 5865–5887del was revealed in $A_{\text{mo}}$ (Fig. 3). A similar discrepancy of blood antigen expression between RBCs and secretions is also observed in $A_{3}$ and $B_{3}$ phenotypes, where single nucleotide substitutions of c.28 + 5864$G > A$ and c.28 + 5880$A > G$ were found around the GATA and RUNX1 motifs in the +5.8-kb site (Fig. 3). Similarly, single-nucleotide substitution of c.28 + 5885$C > T$ was reported around the RUNX1 motif in an individual with $B_{3}$. These genetic studies confirmed the regulatory significance of the +5.8-kb site for erythroid-cell-specific expression of $ABO$ in vivo.

Conclusion
It appears that the proximal promoter and the +5.8-kb site are required for $ABO$ expression in an erythroid cell–specific manner (Fig. 1B), although it remains to be explored whether regions other than the +5.8-kb site might affect $ABO$ expression in erythroid cells.

Regulatory region for epithelial cell–specific expression of $ABO$

In vitro studies
For delineation of $ABO$ regulation in epithelial cells, we prepared luciferase reporter plasmids on the estimation of enhancers within a 50-kb region of genomic DNA in and around $ABO$ in epithelial cells by publicly available data from DNase-Seq and chromatin state segmentation (ChromHMM) on the UCSC Genome Browser. Subsequent plasmid-based reporter assays indicated a distal regulatory region between +22563 and +22781 relative to the translation start site of $ABO$, termed the +22.6-kb site, in KATOIII cells (Fig. 1A), and the regulatory activity of the region was specific to epithelial cells. Subsequently, we validated the significance of the +22.6-kb site with use of KATOIII cells with homozygote deletion of the site constructed by the CRISPR/Cas9 system. EMSAs and ChIP assays demonstrated that the region bound an epithelial cell–specific TF, Elf5 (Fig. 1A), whereas variant of the recognition motif that abrogated binding of the factor reduced the regulatory activity of the site in KATOIII cells. Thus, binding of Elf5 seemed to be crucial for the epithelial cell–specific activity of the region.

Conclusion
It is likely that the proximal promoter and the +22.6-kb site are required for $ABO$ expression in epithelial cells (Fig. 1B), although regions other than the +22.6-kb site might also have some influence.

Other regulatory regions for $ABO$ expression

CCAAT-binding factor/NF-Y enhancer region
The initial luciferase reporter assays with KATOIII cells demonstrated that a positive element was located between −3931 and −3650 from the translation start site where four tandem copies of 43-bp repeat units bound a positive TF, CCAAT-binding factor/NF-Y, through the CCAAT motif (Fig. 1A). However, similar regulatory activity was not
observed in K562 cells and HEL cells of erythroid origin.\textsuperscript{18,23} Thus, it was likely that the minisatellite was not involved in transcriptional regulation of ABO in erythroid cells. Genetic population studies revealed that both the B and O alleles are linked via four tandem copies of a 43-bp repeat unit, and that the A\textsubscript{1} allele is linked in the absence of this tandemly repetitive element.\textsuperscript{49} Seltsam et al.\textsuperscript{52} observed unexpected variations in the CCAAT-binding factor/NF-Y enhancer region including the repeat units in four individuals with weak B phenotypes, suggesting that those weak phenotypes might be caused by sequence variations in the enhancer region. On the other hand, Thuresson et al.\textsuperscript{53} reported a hybrid allele between O\textsuperscript{2} and B which lacked three repeat units, although the B transcript level was similar to that in fresh peripheral blood samples from normal controls. Thus, it remains controversial whether ABO transcription is influenced by the CCAAT-binding factor/NF-Y enhancer region in erythroid cells.\textsuperscript{36,52,53}

**Distal promoter of ABO**

At the 5’ end of the CGI involving the ABO proximal promoter in cultured cells expressing ABO, an alternative starting Exon 1a comprising 27 base pairs was found by 5’-RACE (Fig. 1A). The level of transcription from Exon 1a was much lower than that from Exon 1.\textsuperscript{35} The luciferase reporter assays demonstrated that the sequence located between −864 and −699 was responsible for transcription from Exon 1a in both erythroid and epithelial cell lineages. However, significance of Exon 1a remained elusive.

**The region proximal to the proximal ABO promoter**

The plasmid-based reporter assays demonstrated a negative regulatory element just upstream from the proximal ABO promoter in KATOIII cells and HEL cells,\textsuperscript{54} suggesting that ABO transcription is regulated by negative elements in the −307 to −150 region from the translation start site. EMSAs indicated that this region bound to a nuclear factor from KATOIII cells. However, we have not identified this factor.

**INFERENCES REGARDING ABO REGULATION**

**Cell type–specific expression**

ABO transcription is regulated by a constitutive proximal promoter and a cell-specific regulatory region such as the +5.8-kb site or the +22.6-kb site (Fig. 1B). Luciferase reporter assays showed that the erythroid cell-specific regulatory activity of the +5.8-kb site was dependent upon binding of the epithelial cell–specific TF Elf5.\textsuperscript{48} In fibroblasts not expressing GATA-1 or -2, or Elf5, it is plausible that abrogation of the site’s cell-specific regulatory activity contributes to lack of ABO expression.\textsuperscript{48} Therefore, it is likely that the cell type–specific expression of ABO is dependent upon expression of cell-specific TFs binding to those cell-specific regulatory elements.

**Cell differentiation–specific regulation**

In vitro erythroid differentiation of CD34\textsuperscript{+} cells and AC133\textsuperscript{+}CD34\textsuperscript{+} cells from peripheral blood mononuclear cells indicated that ABO was expressed at an early stage and disappeared later,\textsuperscript{35,42} and that the period when ABO was

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**Fig. 4. Schematic illustration of the expression of cell surface antigens and genes during the course of in vitro erythroid cell differentiation.** The diagrams were constructed with the expression of genes such as FUT1, ABO, GATA-1, GATA-2, and RUNX1 as well as the cell surface expression of B antigen and H antigen 7 days, 11 days, and 15 days after in vitro erythroid cell differentiation from CD34\textsuperscript{+} cells reported previously.\textsuperscript{52} The time course of the relative expression of individual genes or antigens is indicated. The maximum gene expression or cell population showing antigen expression during erythroid differentiation was expressed as 1.0 on the vertical axis, whereas the relative expression ratios at other time points were calculated for individual genes and antigens. Also, changes in the proportions of cells expressing CD71 antigen and CD235 antigen that were not included in that report were added to the diagrams.
expressed at a higher level preceded that of FUT1 expression (Fig. 4). Expression of RUNX1 and GATA-2 characteristically decreases during erythroid differentiation of CD34+ cells. Thus, it seems likely that down regulation of ABO expression might be ascribed to reduction of RUNX1 in the later phase of erythroid differentiation. However, the mechanism of ABO expression at an early stage of erythroid differentiation remains to be explored.

Loss of ABO antigens in malignancy

ABH antigens are often absent from glycoproteins and glycolipids of malignant tissue in the gastrointestinal tract, oral cavity, uterine cervix, lung, prostate, breast, and bladder. ABO antigen expression decreases as a result of down regulated transcription of ABO, which is ascribed to at least two different mechanisms: allelic loss and hypermethylation of the ABO promoter region in a CGI.55–58

There is abundant evidence that methylated CGIs at transcription start sites are associated with some silent genes. We and others have also demonstrated that hypermethylation of the ABO promoter could be responsible for absence of the ABO transcript and A-antigen in gastric and colon cancer cell lines.56,57 Using clinical samples of oral squamous cell cancer, Gao et al.58 showed that loss of A/B antigens was responsible for molecular events such as loss of the A/B allele or ABO promoter hypermethylation in two-thirds of tissue samples they examined. Thus, an additional mechanism for loss of A/B antigens other than allelic loss or promoter hypermethylation remained to be clarified in one-third of such cases. The origin of DNA hypermethylation is currently being clarified, and mutations in such genes as Ten-eleven-translocation 2, DNA methyltransferase 3A, and isocitrate dehydrogenase 1 and 2 encoding epigenetic regulators are known to be involved in DNA methylation.59 Besides mutated epigenetic regulators, mutations in splicing factors may also dysregulate epigenetics through splicing of epigenetic enzymes, and down regulation of transcription is associated with DNA hypermethylation of the promoter. Therefore, the causes and consequences of DNA hypermethylation need to be carefully distinguished.59

The association of weak A expression with acute myeloid leukemia (AML) is well documented.19 Bianco-Miotto et al.50,61 have reported that loss of ABH antigens from RBCs of patients with myeloid malignancies is a frequent phenomenon and that DNA methylation of the ABO promoter underlies the loss of ABO allelic expression in a significant proportion of leukemia patients. Such myeloid malignancies include AML, myelodysplastic syndrome (MDS) and myeloproliferative disorders including chronic myeloid leukemia. Recently, we have reported a patient with MDS in whom blood typing demonstrated mixed-field agglutination, prompting us to investigate the mechanism underlying blood group A-antigen reduction on RBCs.62 Screening of somatic mutations using bone marrow cells demonstrated mutations in ASXL1, EZH2, RUNX1, and WT1. Experiments involving transient transfection into K562 cells showed that the expression of ABO was decreased by expression of the mutated RUNX1. Since the frame-shift mutation of RUNX1 could encode an abnormally elongated protein without a transactivation domain that might act as a dominant negative inhibitor, it was plausible that this RUNX1 mutation might be a genetic factor contributing to A-antigen loss on RBCs. This is another example of a factor other than DNA methylation that could be responsible for ABO antigen reduction in patients with leukemia. Therefore, there is a need for further investigation of patients with leukemia and ABO antigen reduction on RBCs to clarify which events are attributable to ABO-antigen loss on RBCs in leukemia.

The weak phenotype Bm with deletion or mutation of the +5.8-kb site

The prevalence of ABO subgroups was 0.048% among transfusion donors in Tokyo, Japan, in 2010–2011.26 The total occurrence of the Bm and A1Bm subgroups was 0.024% at the Kanto-Koshinetsu Blood Center, indicating that both are the most frequent ABO blood group variants in the Japanese population. A recent study demonstrated Bm5.8 in 1300 individuals, BmGAGA in two, and Bm3.0 in one among 1303 Japanese individuals with Bm and A1Bm with sequence-specific polymerase chain reaction (SSP) targeting Bm5.8 (Fig. 1A).63 In practice, Bm is determined by serologic procedures and SSP targeting Bm5.8 at blood centers in Japan.26 Genetic diagnosis of Bm5.8 is beneficial because Bm accounts for almost one-half of all weak phenotypes in the Japanese population. Based on the above observations, it seems likely that Bm5.8 might have been inherited over a long time period and spread throughout the Japanese population. Since the variant has not been reported in Korea and China, from where ancient people migrated to Japan, Bm5.8 could be specific to the Japanese population.64

Recently, the mutated GATA site in BmGAGA has been reported in individuals with Ael and Bel.31 Moreover, a 5.9-kb deletion of Intron 1 including the +5.8-kb site has been found in an individual with Ael.28 It seems intriguing to explore the reason that the same mutation or a similar deletion of the +5.8-kb site could result in different phenotypes.

PROSPECT

As shown above, ABO transcription is regulated by the proximal promoter and cell-specific regulatory regions. However, ABO transcription was not completely lost in the cells with biallelic deletions of the +22.6-kb site.48 Also, reporter assays demonstrated transcriptional activity in the DHS region +36.0 (Fig. 1A),48 which was suggested to interact with the ABO transcription start site on the basis of publicly available data from GeneHancer Regulatory Elements and
Gene Interactions on the UCSC Genome Browser. In addition, recent studies have suggested coordinated activity of multiple enhancers to control a single gene, regulation of multiple genes by the same enhancer, and competition or coordination between neighboring promoters. Therefore, it would be informative to investigate elements other than the promoter and cell-specific regulatory regions to clarify the regulatory mechanism of ABO transcription, and delineation of the regulatory mechanism of ABO transcription regulation would yield new insight into the regulatory network involving ABO and the genes associated with the ABO regulatory regions.

Histone deacetylase inhibitors are reported to reduce ABO expression in cultured cells, suggesting that ABO transcription may be regulated epigenetically. This also appears to be an intriguing phenomenon in the context of cardiovascular disorders because it has been reported that the levels of von Willebrand factor (vWF) are approximately 25% higher in individuals with blood group types other than O, and this seems to be the main reason for the higher risk of venous thromboembolism and coronary heart disease in non-group O individuals. Because it has been argued that addition of A/B-antigens to vWF in endothelial cells might influence circulating levels of vWF, ABO suppression in cells might reduce the risk of developing these diseases. A similar argument could apply to organ transplantation. In cases of ABO-incompatible (ABOi) liver transplantation, acute humoral rejection directed against donor-oriented A/B antigens on endothelial cells of liver arteries or bile ducts is the most serious form of rejection, and this seems to be the main reason for the higher risk of venous thromboembolism and coronary heart disease in non-group O individuals. Thus, a decrease in the amount of antigen on endothelial cells might ameliorate any adverse effects resulting from ABOi liver transplantation. Further studies may shed further light on ABO transcriptional regulation and provide clues for clinical applications.

CONCLUSION

Clarification of the mechanism of ABO transcriptional regulation has contributed to practical transfusion medicine, and delineation of the regulatory mechanism of ABO transcription regulation would yield new insight into the regulatory network involving ABO and the genes associated with the ABO regulatory regions on Chromosome 9.

CONFLICT OF INTEREST

The authors have disclosed no conflicts of interest.

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