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Immunochemical Studies on Blood Group H and B Substances from Human Hair

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Summary. Blood group H and B substances were extracted from urea-treated human hair of group O and B individuals, respectively, with methanol-ethyl ether (1:1,v/v) and chloroform-methanol (1:1,v/v). The blood group activities of H and B substances were destroyed by H-decomposing enzyme (α-L-fucosidase) from *Bacillus fulminans* and B-decomposing enzyme (α-D-galactosidase) from *Clostridium sporogenes* Maebashi, respectively. It is concluded therefore that the extract from the hair of group O contained blood group H-active glycolipid with α-L-fucose as the non-reducing sugar and the one from group B contained blood group B-active glycolipid with α-D-galactose as the non-reducing sugar.

Key words: Blood groups, H and B substances of hair – H- and B-decomposing enzymes.

Zusammenfassung. Das H-blutgruppenaktive Glycolipoid wurde mit Methanol-Äther (1:1,v/v) und Chloroform-Methanol (1:1,v/v) aus Harnstoff-behandelten Haaren extrahiert. Wenn nach Behandlung der H-Substanz von Haaren mit H-zerstörenden Enzym (α-L-Fucosidase) aus *Bacillus fulminans* H-Aktivität spurlos verschwindet, so kann es sein, daß α-L-Fucose für die H-Aktivität von Haarglycolipoid benötigt wird. Das B-blutgruppenaktive Glycolipoid wurde aus Menschenhaaren extrahiert. Durch Einwirkung von B-zerstörendem Enzym (α-D-Galaktosidase) aus *Clostridium sporogenes* Maebashi auf das B-Glycolipoid aus Haaren, kommt nunmehr die H-Eigenschaft nach Inaktivierung von B zum Vorschein. Darum kann es sein, daß α-D-Glaktose für die B-Aktivität vom Haarglycolipoid benötigt wird.

Schlüsselwörter: Blutgruppen, H und B-Substanz in Haaren – Haare, H und B-Substanzen.

It has been reported that blood group A-active glycolipid was extracted from human hair and that its serological determinant was *N*-acetylgalactosamine [1]. The present paper concerns immunochemical properties of blood group H- and B-active glycolipids obtained from human hair.

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Table 1. Haemagglutination inhibition tests of glycolipids extracted from group O and B hair

Blood group of hair	Antisera		
	Anti-H (chicken)	Anti-A (human)	Anti-B (human)
O	125[a]	> 2000	> 2000
B	500	> 2000	125

[a] Minimum concentration of each glycolipid giving complete inhibition (μg/0.1 ml)

Materials and Methods

Hair. Hair was collected from the head of adult men of group O or B. The methods for preparation of the hair, isolation of glycolipids, incubation conditions with blood group substance-decomposing enzymes, and serological procedures were those described previously [1].

Results

1. Yields of Hair Glycolipids. Twenty g of group O human hair gave 10 mg of acetone precipitate after extraction with methanol-ethyl ether (1:1,v/v) and then chloroform-methanol (1:1,v/v). Twenty g of group B human hair gave 20 mg of acetone precipitate by the same procedure. Each precipitate contained crude glycolipids.

2. Blood Group Activities of the Hair Glycolipids. The glycolipids from human hair were assayed for blood group A, B and H activities by haemagglutination inhibition tests. Glycolipid from the hair of a group O individual exhibited strong inhibition in an O red cells–anti-H system but no inhibition in an A red cells–anti-A or a B red cells–anti-B system. Glycolipid from the hair of a group B individual exhibited strong inhibition in a B red cells–anti-B system, weak inhibition in an O red cells–anti-H system but no inhibition in an A red cells–anti-A system (Table 1).

3. Action of Blood Group Substance-Decomposing Enzymes on the Hair Glycolipids. In order to examine the serological activity and chemical structure of the hair glycolipids, these were treated with blood group substance-decomposing enzymes, viz., H-enzyme from *Bacillus fulminans* [2], A-enzyme from *Clostridium tertium* [3] and B-enzyme from *Clostridium sporogenes* Maebashi [4]. H-enzyme acted on the H-active glycolipid from the hair to give a loss of H activity. However, A- and B-enzymes exerted no effect on the serological activity of H-active glycolipid (Table 2). B-enzyme acted on the B-active glycolipid from the hair to give a loss of B activity and development of H activity; however, no serological change occurred after treatment with H- and A-enzymes (Table 3).

Discussion

Glycolipids were extracted with methanol-ethyl ether (1:1,v/v) and chloroform-methanol (1:1,v/v) from urea-treated hair of group O and B individuals. Blood group H and B activities could be demonstrated in these glycolipids, respectively, by haemagglutination inhibition tests.

Important information of the immunochemical structures associated with serological activity has been obtained using microbial blood group-specific glycosidases [5].

Table 2. Serological changes in the glycolipid extracted from group O hair with blood group-decomposing enzymes

Enzymatic treatment	Antisera		
	Anti-H (chicken)	Anti-A (human)	Anti-B (human)
Before	125[a]	> 2000	> 2000
After			
H-enzyme	1000	> 2000	> 2000
A-enzyme	125	> 2000	> 2000
B-enzyme	125	> 2000	> 2000

[a] See footnote of Table 1

Table 3. Serological changes in the glycolipid extracted from group B hair with blood group-decomposing enzymes

Enzymatic treatment	Antisera		
	Anti-H (chicken)	Anti-A (human)	Anti-B (human)
Before	500[a]	> 2000	125
After			
H-enzyme	1000	> 2000	125
A-enzyme	500	> 2000	125
B-enzyme	125	> 2000	2000

[a] See footnote of Table 1

H substance can be degraded by an H-enzyme from *Bac. fulminans*: with the liberation of the H-specific determinant sugar, α-L-fucose, it is converted to a substance of O_h phenotype [5]. The serological activity of the H substance in human hair was destroyed by this enzyme. Based on the results of this investigation, it seems reasonable to conclude therefore that the blood group H determinant of the hair contained α-L-fucose as the non-reducing terminal sugar. Treatment of B substance with B-enzyme from *Cl. sporogenes* Maebashi resulted in a release of galactose, the determinant sugar of B specificity, so giving rise to the production of a substance with high H activity comparable to that of group O [5]. B-enzyme from this strain destroyed B activity in the B-active glycolipid from human hair with enhancement of H activity. It is apparent therefore that the blood group B determinant of the hair contained α-D-galactose as the non-reducing terminal sugar.

References

1. Kishi, K., Iseki, S.: Studies on blood group A substance from human hair. Z. Rechtsmedizin **80**, 191–195 (1977)
2. Iseki, S., Tsunoda, S.: On a bacterial enzyme which specifically decomposes O substance. Proc. Japan Acad. **28**, 370–373 (1952)
3. Iseki, S., Masaki, S.: Transformation of blood group substance by bacterial enzyme. Proc. Japan Acad. **29**, 460–465 (1953)
4. Iseki, S., Furukawa, K., Yamamoto, S.: B substance-decomposing enzyme produced by an anaerobic bacterium. 1. Serological action on the B-decomposing enzyme. Proc. Japan Acad. **35**, 507–512 (1959)
5. Iseki, S.: Glycosidases and serological changes in blood group substances. In: Blood and Tissue Antigens, Aminoff, D. (ed.), pp. 379–394. New York: Academic Press, Inc. 1970

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