



# Immunohematology Case Studies 2016 - October Where Are My Antibodies?

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# Clinical History - 1



- This 63-y-o male patient presenting with low back pain and swelling pain of both legs for about 2 months came to our ER for assessment. He had undergone operation for thymoma 8 years ago
- Physical examination revealed a body temperature of 38.7 Celsius degree, and redness, swelling, and tenderness of both lower legs (R't > L't). Otherwise was unremarkable

# Clinical History - 2



- Complete blood count analysis revealed leukocytosis (WBC 13,420/uL) and microcytic anemia (Hb 8.3 g/dL, MCV 76.1 fL)
- Antimicrobial treatment with Oxacillin was given
- Blood transfusion was suggested as hemoglobin level dropped to 7.6 g/dL

# Laboratory Data



Test	April 27	May 02	May 06	May 09	Reference Range	Unit
WBC	<b>13.42</b>	<b>10.63</b>	9.27	9.83	3.8~10.4	$10^3/\mu\text{l}$
RBC	<b>3.35</b>	<b>3.1</b>	4.16	4.19	4.1~5.90	$10^6/\mu\text{l}$
HGB	<b>8.3</b>	<b>7.6</b>	<b>10.2</b>	<b>10.5</b>	13~17	g/dl

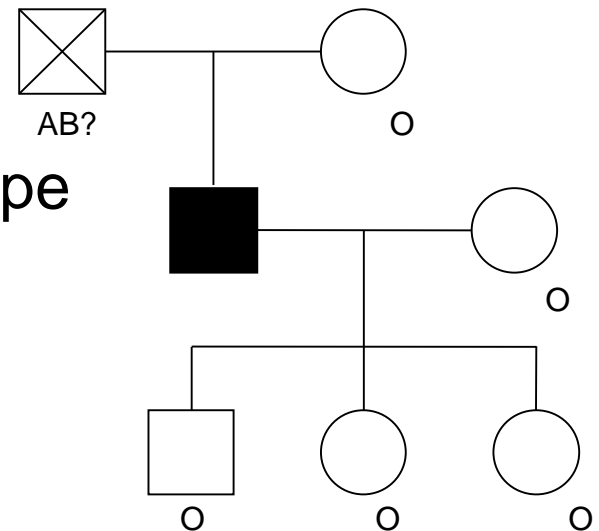
# Serologic History



## ABO Discrepancy

Anti-A	Anti-B	Anti-D	A1 cells	B cells	Ab Screening		
					I	II	III
—	—	4+	—	—	—	—	

- Ensured reagent/equipment QC, and rechecked the patient's sample again
- Underwent operation for thymoma and received 2U O-type packed red cells transfusion
- BMT : Denied



# The causes of ABO Discrepancy

(weakly reacting or missing antigen in forward grouping)



	Causes	Verification
1	Subgroups of A or B	Excluded
2	Leukemia	Not present
3	ABO incompatible HPC transplantation	Not present
4	Neutralization of anti-A and anti-B typing reagent by high concentration of A or B soluble substances in serum with serum or plasma suspended red cell	Rechecked after saline washing of red cells

Anti-H	Anti- A,B	Anti-A1	A2 cells	Adsorption and Elution		
				A Ag	B Ag	H Ag
4+	—	—	—	—	—	+

# The causes of ABO Discrepancy

(weakly reacting or missing antibodies in reverse grouping)



	Causes	Verification
1	Infants less than 4-6 month of age or elderly?	63 years old
2	ABO incompatible HPC transplantation	Not present
3	Severe hypogammaglobulinemia	?
4	Hematopoietic Chimerism	?

# Further Work



Test	05-06	06-09	Reference	Unit
Creatinine	1.05	1.18	0.7~1.20	mg/dl
eGFR	>60	>60	>60	
Test	05-06	06-18	Ref.	Unit
Total Protein	<b>4.4</b>	<b>5.72</b>	6.6~8.7	g/dl
Albumin	<b>3.1</b>	4.33	3.5~5.0	g/dl
Globulin	<b>1.3</b>	<b>1.4</b>	2.1~3.5	g/dl

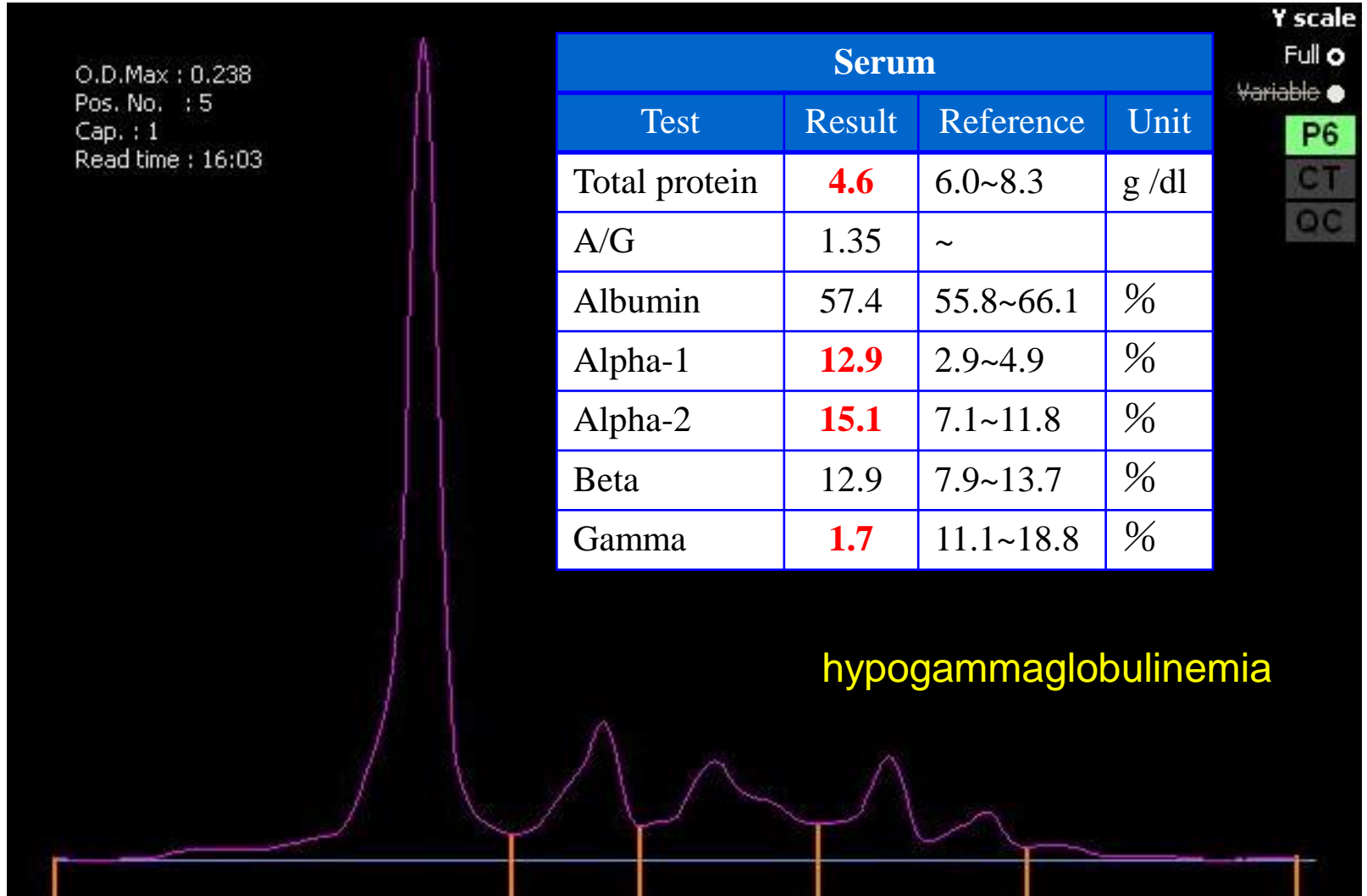


# Further Work (Immunoglobulin)



Test	May-06	June-18	Reference	Unit
IgA	< 24.5	< 24.5	70~400	mg/dl
IgG	< 6.5	13.7	700~1600	mg/dl
IgM	< 15.6	< 15.6	40~230	mg/dl
IgE	< 4.1	< 4.1	0~200	IU/ml
C3	139	140	90~180	mg/dl
C4	38.1	37.5	10~40	mg/dl

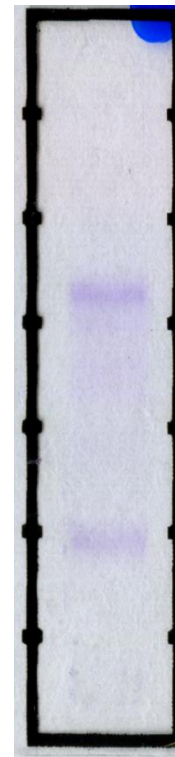
# Further Work (Serum Protein Electrophoresis)



# Further Work (Urine Protein Electrophoresis)



Urine		
Test	Result	Unit
Total protein	8	g /dl
A/G	0.3	
Albumin	22.7	%
Alpha-1	10.3	%
Alpha-2	22.6	%
Beta	37.2	%
Gamma	7.2	%



← Gamma globulin

← Beta globulin

← Alpha-2 globulin

← Alpha-1 globulin

← Albumin

Physiological proteinuria pattern

# Hypogammaglobulinemia



**TABLE 2**

## Laboratory evaluation of hypogammaglobulinemia

DISORDER	TOTAL IgG	IgA	IgM	IgG SUBCLASS
<b>Common variable immune deficiency</b>	Low	Variable	Variable	Low
Selective IgA deficiency	Normal	Low	Normal	Variable*
IgG subclass deficiency	Normal	Normal	Normal	Low*
Selective antibody deficiency	Normal	Normal	Normal	Variable
<b>Secondary hypogammaglobulinemia</b>	Low	Variable	Variable	Low

\*Most commonly IgG<sub>2</sub> subclass

Rose ME, Lang DM. Evaluating and managing hypogammaglobulinemia. Cleve Clin J Med 2006; 73:40,43–4,133–7. 7028

# Common Variable Immunodeficiency (CVID)



- A heterogeneous immune disorder, is characterized by frequent and **recurrent infections** and decreased concentrations of **multiple classes of immunoglobulins**
  - Normal numbers of B lymphocytes
  - Fail to differentiate into antibody-secreting plasma cells.
  - May have increased apoptosis of helper T cells and decreased T-cell function and signaling
- The estimated prevalence of CVID is **1 in 20,000 to 100,000**. More often presents in **adults**
- Treatment with high doses of intravenous immunoglobulins (**IVIG**) leads to improved outcomes

# Secondary Hypogammaglobulinemia



- **Diseases of immunoglobulin loss**
  - Protein-losing enteropathies
  - Chronic renal disorders
- **Diseases of immunoglobulin production**
  - A number of malignancies, including chronic lymphocytic leukemia (CLL), lymphoma, and multiple myeloma, are associated with secondary hypogammaglobulinemia
- **Drug induced states**
  - Disease-modifying antirheumatic drugs, Systemic steroids, Phenytoin, Carbamazepine, Androgen replacement therapy
- **High-stress states**
  - Extreme physical activity
  - Low calorie intake
  - Sleep deprivation

# Reference Review (Good's Syndrome)



- The presence of **thymoma** with **hypogammaglobulinemia** confirmed the diagnosis of Good's syndrome
- Good's syndrome is rare and comprises **less than 5%** of thymoma patients
- This case had a history of thymoma with surgery 8 years ago

# Hematopoietic Chimerism



## Definition :

- The persisting presence of allogeneic donor blood cells in a recipient

## Classification:

- Macrochimerism: The goal of hematopoietic peripheral blood progenitor cell transplantation
- Microchimerism (less than 1-5% donor cells):  
Organ transplantation, Pregnancy, Transfusion

## Detection (real-time PCR) :

- Screening of all non-HLA-identical donor-recipient combinations for the presence of disparate HLA antigens
- Screening for insertion/deletion polymorphisms (markers)

- Bob a. van Dijk, Dorre I. Boomsma, and Achile J.M. de Man. Blood Group Chimerism in Human Multiple Births Is Not Rare. *Am J Med Genet.* 1996 Jan 22;61(3):264-8.
- Fast L. Microchimerism: a lasting legacy of transfusion? *Transfusion* 2006;46:1856-8.
- Lee T, Paglieroni T, Ohto H, et al. Survival of donor leukocyte subpopulations in immunocompetent transfusion recipients: frequent long-term chimerism in severe trauma patients. *BLOOD* 1999;93:3127-39

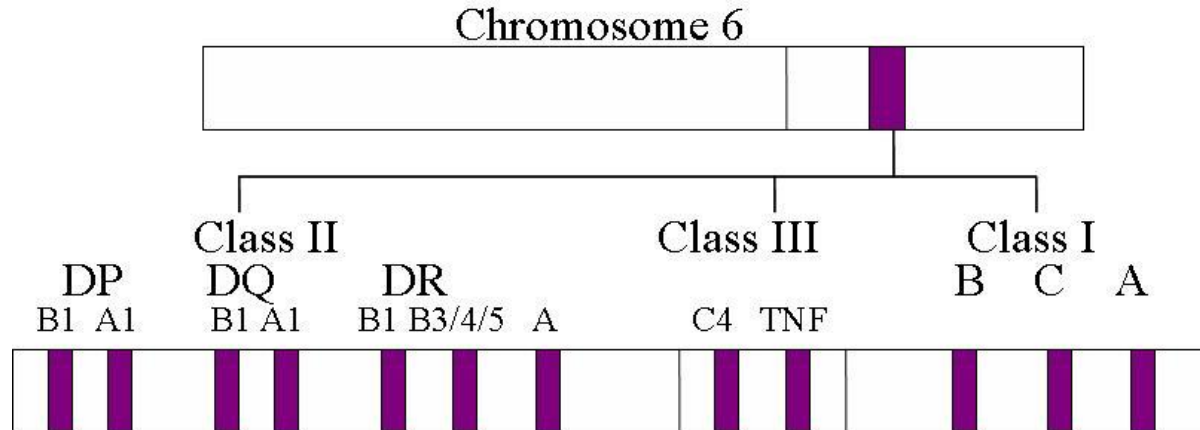


# Microchimerism (Blood Transfusion)



- The factors that may be associated with establishment of chimerism are storage time of blood units of **less than 14 days** and administration of the transfusion within **48 hours** of a major physiologic insult
- After transfusion in **trauma patients** is that the patient's response to the trauma facilitates the establishment of chimerism
- The degree of HLA disparity between donor(s) and recipient. The data indicate that **chimeric cells often express one or two disparate HLA-DR antigens**, which would mean that chimeric cells would often be haploidentical at the **DR locus**

# Further Testing



➤ HLA :

- A2 A24
- B38 B54
- Bw4 Bw6
- Cw1 Cw7
- DR4 DR9**
- DQ8 DQ9

# Conclusions



- The most probable explanation for the ABO discrepancy **was hypogammaglobulinemia**, according to the previous investigation
- Type O blood component is recommended if the patient needs transfusion

# Lessons Learned by the Case



- Diminished titer of expected ABO blood group antibodies was not uncommon
- Stepwise serological investigation incorporating pertinent medical history could not be overemphasized

# References



- Bob a. van Dijk, Dorre I. Boomsma, and Achile J.M. de Man. Blood Group Chimerism in Human Multiple Births Is Not Rare. *Am J Med Genet.* 1996 Jan 22;61(3):264-8.
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