Students' Corner
Case Report

Warm Autoimmune Haemolytic Anaemia and autoimmune hepatitis in an asymptomatic carrier of hepatitis B virus

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Abstract

Warm antibody autoimmune haemolytic anaemia, a rare disease (0.2-1 per 100,000 populations), is due to the presence of warm agglutinins that react with protein antigens on the surface of red blood cells causing their premature destruction. Here, we present a case report of a 10 year old girl who came with features of haemolytic anaemia and history of blood transfusion since 3 years. On admission, laboratory test revealed that she had autoimmune hepatitis type 1 and was also an asymptomatic carrier of hepatitis B virus with positive HBs Ag. Steroid therapy resulted in clinical and laboratory remission. Direct antiglobulin test was negative after anaemia resolution, hepatitis B virus antigenemia persisted. To our knowledge, warm antibody autoimmune hemolytic anaemia has not previously been described in association with autoimmune hepatitis and asymptomatic carrier state of hepatitis B virus.

Keyword: Autoimmune haemolytic anaemia, Autoimmune hepatitis, Hepatitis B virus.

Introduction

Warm autoimmune haemolytic anaemia (WAIHA) results from the production of autoantibody against unspecified high frequency red cell antigen(s).[1] Usually, the

antibody is of the IgG isotype and causes the destruction of patient and donor red cells at body temperature. Primary and secondary forms of the disease exist.[1] The activation of the immune system in primary cases is poorly understood. In WAIHA, the antigens targeted by autoantibodies are either not known or react against red cell membrane protein(s).[2]

Autoimmune hepatitis (AIH) is a chronic necroinflammatory disease of the liver, occurring in the absence of a known etiologic agent, which causes chronic hepatitis and is characterized by fluctuating alanine aminotransferase (ALT) levels in serum, marked hypergammaglobulinaemia, and circulating autoantibodies.[3] These features, as well as the responsiveness to immunosuppressive therapy have led to the belief that this is an autoimmune disease in its own right, distinguishable from other liver diseases associated with hepatotropic viruses, metabolic disorders and drugs. Although the pathogenesis of AIH is not known, it is accepted as the loss of tolerance of an organism to its own liver tissue. On the other hand, in genetically susceptible persons, environmental factors such as virus, bacteria and chemical agents are thought to be responsible for inducing the autoimmune process.[3] In different studies it has been shown that asialoglycoprotein receptor, which is found on the outer surface of the liver hepatocyte membrane, causes immunological reactions in autoimmune chronic hepatitis.[3,4] A defect is suggested in specific suppressor T lymphocytes controlling the immunological response to asialoglycoprotein following long-lasting viral infection.[4]

Herpes simplex virus type I, Epstein-Barr virus, measles virus, and hepatitis A, B, C, and D viruses are thought to play a role in the etiology of AIH.[3] Viral proteins belonging to these viruses may be similar to the amino acid chain of different autoantigens in the liver. Therefore, cross immune reaction to viral proteins causes damage to liver tissue at the same time.[3,5] An example of this mechanism is the similarity between herpes simplex type I and cytochrome p450IID6, the major antigen of anti-liver kidney microsomal antibody-1 (anti LKM-1).[5] A few cases of AIH have been reported, linked to well-documented HBV infection,[6] HBV carriers who show autoimmunity as assessed by hypergammaglobulinaemia and the presence of autoantibodies are occasionally seen.

No coincidence of WAIHA and AIH associated with asymptomatic carrier state of HBV infection has been reported to date. Thus, we present our case with WAIHA and AIH with asymptomatic carrier state of HBV infection.

Case Report

A 10-year old non-vaccinated girl was admitted on 25th June 2010; complaining of progressive pallor and malaise. She was the second child to consanguineous parents. Her medical history before this episode revealed that, in February 2007, she had experienced similar complaints of extreme pallor and weakness, for which she was admitted to a local public sector hospital in Quetta, where she was worked up for anaemia. At that time, laboratory examination revealed deterioration of haemogram and positive Direct Coombs test (Table-1). The patient was diagnosed as autoimmune haemolytic anaemia and had recovered after transfusion of packed red cells. She then experienced similar attacks of pallor and malaise in March 2007 and January 2008, which were managed by supportive treatment. Prednisolone was prescribed in April 2007, which was continued. The patient presented with severe pallor, malaise and loss of weight since last two weeks.

Physical examination revealed height 115 cm, weight 22 Kg, severe pallor, mild icterus and distended abdomen with hepatosplenomegaly.

Laboratory examination showed haemoglobin 2.7 g/dl, haematocrit 7.4%, total leukocyte count 39.4 /µl,

Table-1: Past laboratory findings before admission.

Date		2007/1/27	2007/3/27	2007/5/07	2008/1/23	2010/6/15	Normal range
RBC	$\chi 10^{12}$ / (L)						
0.57	1.57	2.42	0.93	0.90	(4.5-6.5)		
Hb	(g/dl)	2.8	4.0	8.9	4.4	4.10	(11.1-15.3)
Htc	(%)	8.1	18.0	26.6	12.2	12.80	(35-15)
MCV	(fl)	142.1	114.6	109.9	132	129	(76-96)
MCH	(pg)	49.1	36.9	36.8	47.7	41.4	(27-32)
MCHC	(g/dl)	34.6	32.2	33.5	36.1	32.0	(30-35)
LDH	(U/L)					1932	(240-480)
Reticulocyte	(%)	58	31	1.2	75	8.8	(0.2-2.5)
ESR	(mm 1st Hr)	170	120	18	135		(0-20)
ALT	(U/L)					65	(<31)
T.Bil	(mg/dl)		2.5			3.48	(0.1-1.0)
D.Coomb		+3	+3		+2	+2	(negative)

RBC: red blood cell, Hb: hemoglobin, Htc: hematocrit, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, LDH: lactate dehydrogenase, ESR: erythrocyte sedimentation rate, ALT: alanine transaminase, T.Bil: total bilirubin, D.Coomb: direct coombs test.

Table-2: Laboratory findings on admission.

CBC			normal range
WBC	39450	/µL	4000-11000
RBC	0.60	$\chi 10^{12}$ / (L)	3.7-5.1
Hb	2.7	g/dl	11.1-15.3
Htc	7.4	%	35-45
MCV	115.6	Fl	85.0-100.0
MCH	42.2	Pg	30.0-35.0
MCHC	36.5	g/dl	32.0-36.0
MCF	0.40	%NaCl	0.445
Platelet	15.6	$\chi 10^{4}$ / µl	15.0-40.0
Reticulocyte	41.35	%	0.5-2.0
ESR	140	mm 1st Hr	0-20
Biochemistry			
T. bilirubin	3.78	mg/dl	0.10-1.00
D. bilirubin	0.63	mg/dl	0.05-0.30
ALT	121	U/L	30-65
LDH	975	U/L	240-480
ALP	184	U/L	45-136
G6PD Quantitative	>19.5	U/gHb	>4.6
BUN	10	mg/dl	10-20
Cr	0.3	mg/dl	0.6-1.0
UA	3.5	mg/dl	2.6-6.0
Na	136	mmol/l	136-148
K	3.7	mmol/l	3.6-5.0
Cl	104	mmol/l	98-109
TP	8.3	g/dl	6.5-8.0
Albumin	3.8	g/dl	3.4-5.0
Globulin	4.5	g/dl	1.9-2.8
Ceruloplasmin	0.676	g/l	0.15-0.60
Hepatitis virus			
HBs Ag	+		negative
HBe Ag	-		negative
anti-HCV ab	-		negative
Immunology			
IgG	22.62	g/l	5.70-14.10
IgA	3.93	g/l	0.65-2.60
AMA	-		negative
ANA	-		negative
anti-ds DNA ab	+		negative
ASMA	+		negative
anti-LKM 1 ab	-		negative
CRP	3.4	mg/dl	0.0-0.9
C3	1.26	g/l	0.9-1.8
C4	0.25	g/l	0.1-0.4
D. Coomb	+3		negative
Coagulation			
APTT	27.5	Seconds	26
PT	11.0	Seconds	11.0
INR	1.00		
Urinalysis			
pH	7.5		5.0-7.0
SG	1.010		1.005-1.020
Proteins	Trace		nil
Ketone	-		nil
Sediment			
RBC	02/HPF		
WBC	03/HPF		
Epithel	02/HPF		
Cast	nil		
Urobilinogen	131	µmol/l	<16

reticulocytes count 41.35%, direct coombs IgG positivity (3+), ESR 140 mm in the 1st hour and bone marrow, dyserythropoietic features like bi and tri-nucleated erythroblasts with erythroid hyperplasia. Glucose 6 phosphate dehydrogenase enzyme activity was normal. Dimorphic picture, macrocytosis, anisocytosis, polychromasia, red cell clumps, spherocytosis, nucleated RBC's, occasional myelocytes and metamyelocytes were detected in blood smear. Urinalysis was normal except for increased urobilinogen. Serum LDH (975 U/l), ALT (121 U/l), total bilirubin (3.78 mg/dl), indirect bilirubin (3.15 mg/dl), total protein (8.3 g/dl), globulin (4.5 g/dl), serum immunoglobulin lgG (22.62 g/l) and lgA (3.93 g/l), C-reactive protein (3.4mg/dl), and ceruloplasmin (0.67 g/l) were raised (Table-2). RBC osmotic fragility, Hb electrophoresis, serum electrolytes, total lipid, cholesterol, glucose, urea and creatinine levels were normal. Blood culture was positive for Pseudomonas aerogenosa for which patient received combination of injection amikacin and syrup ciprofloxacin for a total of 10 days. Antinuclear antibody (ANA), antimitochondrial antibody (AMA) and anti-liver kidney microsomal antibody-1 (anti LKM-1) were found negative, while anti-smooth muscle antibody (ASMA) and anti-ds DNA antibody were found positive. Abdominal ultrasonography revealed spleen massively enlarged measuring approx. 22.2 cm, and mild hepatomegaly with regular contours and normal parenchymal echogenecity. HBs-antigen was positive, while HBe-antigen and anti-HBs antibody were negative in serum. Liver biopsy was not performed. Serum anti-HCV antibody was negative. Echocardiography showed dilated left ventricle with mild generalized left ventricular dysfunction and aortic regurgitation.

On the basis of the above findings a diagnosis of warm antibody autoimmune haemolytic anaemia and autoimmune hepatitis type with concomitant asymptomatic carrier of hepatitis B was made. She was transfused 150ml of packed RBC initially on admission. When the diagnosis of AIHA was confirmed, she was put on a regular dose of prednisolone adjusted according to her body weight. The haemolytic anaemia improved after a hospital stay of 37 days; the patient was therefore discharged from hospital with haemoglobin 10.6 g/dl, haematocrit 34.6 %, reticulocytes 13.5%, direct coombs lgG negative and persistently positive HBs-antigen and negative anti-HBs antibody. Patient was advised for regular follow up every 6 weeks, but she did not comply with the instruction.

Discussion

Haemolytic anaemia is caused by premature destruction of circulating red blood cells. It can be intravascular, as in infections, transfusion reactions, and

paroxysmal cold haemoglobinuria, or extravascular, as in enzyme deficiencies, membranopathies, haemoglobinopathies, drug reactions or autoimmune.

In this case, the elevated LDH and indirect bilirubin, and raised reticulocytes count all were consistent with haemolysis. No malarial or other haemoparasite were seen in peripheral blood smear. Hb electrophoresis revealed no haemoglobinopathy. A normal G6PD level, obtained after the acute phase of the illness, eliminated G6PD deficiency as a possibility. Direct Coombs test revealed an anti-IgG antibody on the RBC's, consistent with warm autoimmune haemolytic anaemia (WAIHA). The leading diagnosis was therefore WAIHA. WAIHA results from production of an IgG isotype autoantibody against unclassified red cell antigens or red cell membrane proteins.[1,2] In most cases of WAIHA, the nature of the antigens is unknown.[2] WAIHA has been reported in patients with several liver diseases including chronic hepatitis C virus infection,[7] hepatitis A viral infection,[8] asymptomatic carrier of hepatitis B viral infection,[9] and non-alcoholic steatohepatitis.[1]

Autoimmune hepatitis (AIH) usually presents as an acute disease with poor prognosis. It can affect any age group but tends to predominate in women.[3] In AIH there are several autoantibodies formed against antigens specific or nonspecific to the liver. AIH is classified as type I where ANA and/or ASMA is positive and type II where antiLKM-1 and/or anti-liver cytosol type 1 antibody is positive.[3] Pathogenetic mechanisms of AIH are not totally clear, but in patients with symptomatic AIH the inducing factors should be investigated.

Autoimmunity, as assessed by hypergammaglobulinaemia and the presence of autoantibodies, is well recognized in chronic hepatitis C.[3] Although it remains unclear whether HCV induces AIH or not, recent studies have shown a high incidence of seropositivity for anti-HCV antibodies in patients with AIH. The existence of anti-GOR antibody, which cross-reacts with both cell-nuclear protein and nucleocapsid protein of HCV, or molecular mimicry between the nucleocapsid protein of HCV and the liver-kidney microsome would support the possibility of HCV infection being involved in the pathogenesis of autoimmune hepatitis.[3,5]

Only a few studies have evaluated the autoimmune response in patients with chronic hepatitis B. In 1996, Murakami et al. reported a 43-year old Japanese woman who developed AIH ten years after being diagnosed as asymptomatic HBV carrier.[10] The autoimmune response has been reported to be more marked in patients with chronic hepatitis C than in those with chronic hepatitis B.[3] This case suggests that an autoimmune mechanism may play an important role not only in patients with chronic hepatitis C but also in HBV carriers.

Conclusion

In conclusion, HBV carrier can induce AIHA and AIH that may progress to liver insufficiency and cirrhosis or other autoimmune diseases in patients genetically susceptible to autoimmune diseases. Therefore, a diagnosis of AIHA should alert paediatricians to the possibility of an associated systemic disease. Thus, faced with a patient with AIHA, clinical and laboratory investigations are indicated with the objective of identifying subjacent pathologies such as infectious diseases, autoimmune diseases and neoplasms as early as possible. Also, the optimal treatment strategy for viral hepatitis associated with autoimmunity remains to be established. More strict diagnostic methods that differentiate AIH from viral hepatitis are required in order to determine appropriate treatment.

Consent:

Written informed consent was obtained from the patient's parents for publication of this case report and any accompanying images.

Competing Interests:

The authors declare that they have no competing interests.

References

1. Boctor FN, Gorak E, Prichard JW, Firouzi M, Difilippo W. Unusual warm autoimmune hemolytic anemia in non-alcoholic steatohepatitis. Ann Clin Lab Sci 2008; 38: 273-6.
2. Kamesaki T, Kajii E. [Detection and characterization of anti-red cell autoantibodies in autoimmune hemolytic anemias]. Nippon Rinsho 1996; 54: 2430-5.
3. Malik TA, Saeed S. Autoimmune hepatitis: a review. J Pak Med Assoc 2010; 60: 381-7.
4. Yoshioka M, Mizuno M, Morisue Y, Shimada M, Hirai M, Nasu J, et al. Anti-asialoglycoprotein receptor autoantibodies, detected by a capture-immunoassay, are associated with autoimmune liver diseases. Acta Med Okayama 2002; 56: 99-105.
5. Manns MP, Obermayer-Straub P. Viral induction of autoimmunity: mechanisms and examples in hepatology. J Viral Hepat 1997; 4 (Suppl 2): 42-7.
6. Laskus T, Slusarczyk J. Autoimmune chronic active hepatitis developing after acute type B hepatitis. Dig Dis Sci 1989; 34: 1294-7.
7. Ohsawa I, Uehara Y, Hashimoto S, Endo M, Fujita T, Ohi H. Autoimmune hemolytic anemia occurred prior to evident nephropathy in a patient with chronic hepatitis C virus infection: case report. BMC Nephrol 2003; 4-7.
8. Urganci N, Akyildiz B, Yildirmak Y, Ozbay G. A case of autoimmune hepatitis and autoimmune hemolytic anemia following hepatitis A infection. Turk J Gastroenterol 2003; 14: 204-7.
9. Yoshioka K, Miyata H. Autoimmune haemolytic anemia in an asymptomatic carrier of hepatitis B virus. Arch Dis Child 1980; 55: 233-4.
10. Murakami C, Hino K, Okazaki M, Fujii K, Okuda M, Hanada H, et al. Hepatitis B virus carrier status linked to autoimmune hepatitis. Intern Med 1996; 35: 468-71.