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Ümmügülsüm Can*, Ersin Sayar, Muhittin A. Serdar

Macro-aspartate aminotransferase (AST): a case report

Makro aspartat aminotransferaz (AST): Bir olgu takdimi

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Abstract: Aspartate aminotransferase (AST) macroenzyme leads to an increase in AST without the presence of any disease. In present report, an isolated increase of AST was detected in a 6-year old girl following the investigations prior to tonsillectomy. As to her history, physical examination and other laboratory test results, no abnormal findings were detected. AST levels were measured as 591 IU/L and 585 IU/L after repeated tests. While found as <5.4 IU/L (recovery <0.8%, reference interval 42.0–82.2%) after polyethylene glycol (PEG) precipitation, AST was determined as 561 IU/L (recovery 95.4%) as a result of the assessment performed with non-specific human anti-mouse antibodies (HAMA). No diseases were encountered in the patient, and reason for the increase of AST was considered to be macroAST. Because continuously increased enzyme value may lead to various invasive and expensive diagnostic tests, macroAST should be taken into account in the evaluation process.

Keywords: Macro-aspartate aminotransferase (AST)

Özet: Aspartat aminotransferaz (AST) makroenzimi herhangi bir hastalık olmadan AST'de yükselmeye neden olur. Bu çalışmada 6 yaşında bir kız operasyon öncesi yapılan incelemeler sonrası izole AST yüksekliği saptanmıştır. Anamnez fizik muayene ve diğer laboratuvar testlerinde anormal bir veri saptanmamıştır. Yapılan tekrar ölçümler sonucunda 591 ve 585 U/L saptanmıştır. PEG çöktürme sonrası AST <5.4 IU/L (geri kazanım <0.8%, referans aralık 42.0–82.2%) saptanmıştır. HAMA bloke edici tüplerle yapılan çalışma sonucunda 561 (geri kazanım 95.4%) geri kazanım U/L saptanmıştır. AST artış nedeni olarak makro AST düşünmüş ve zemininde patoloji tespit edilmemiştir. Devamlı artmış enzim değeri çeşitli invaziv veya pahalı teşhis testlerinin yapılmasına yol açabileceğinden değerlendirme sırsında göz önünde bulundurulmalıdır.

Anahtar Kelimeler: Makro aspartat aminotransferaz (AST)

Introduction

A 6-year-old girl was admitted to the hospital with complaints, and tonsillectomy was decided as a result of physical examination and laboratory test results. Within pre-op period, an isolated increase of aspartate aminotransferase (AST) was determined in the case. On her history and physical examination, no pathological findings, use of drugs and history of any diseases were detected. Scleras were found to be anicteric, abdomen to be soft, and no palpable mass or edema was observed.

Laboratory test results were found as follows: AST, 585 IU/L (reference interval, 13–35); alanine aminotransferase (ALT), 13 IU/L (12–40); alkaline phosphatase (ALP), 216 IU/L (80–325); lactate dehydrogenase (LDH), 267 IU/L (112–295); albumin, 4.5 g/dL (3.5–5.2); gamma-glutamyltransferase (GGT), 14 IU/L (0–38); hemoglobin, 12.5 g/dL (11.5–14.5); total bilirubin, 1.0 mg/dL (0.3–1.2); direct bilirubin, 0.1 mg/dL (0.0–0.2); creatine kinase (CK), 58 IU/L (0–145);

***Corresponding author: Ümmügülsüm Can:** Konya Training and Research Hospital, Department of Biochemistry, Konya, Turkey, e-mail: cangulsum@yahoo.com
Ersin Sayar: Konya Training and Research Hospital, Department of Pediatric Gastroenterology, Konya, Turkey, e-mail: ersinsayar@hotmail.com
Muhittin A. Serdar: Acıbadem University, Clinical Laboratory of Department of Medical Biochemistry, İstanbul, Turkey, e-mail: maserdar@hotmail.com

Table 1: Different measurements for macroAST.

	Serum direct	1/10 Serum dilution	1/2 PEG precipitation	1/1 PEG precipitation	HAMA with tube
AST U/L	591.9	61.8	77.1 (the real value 231)	<5.4	561
ALT U/L	15.1			<5.4	15.5
GGT U/L	12.4				14.5
ALP U/L	260.1				264.8
CPK U/L	153				

prothrombin time, 8 sec (0–12); international normalized ratio (INR), 0.9 (0.8–1.2); thyroid stimulating hormone (TSH), 3.0 μIU/mL (0.5–6.0); antinuclear antibodies (ANA) negative (-); hepatitis B surface antigen (HBsAg), negative (-); and, hepatitis C virus (HCV), negative (-).

Methods

For polyethylene glycol (PEG) 8000 precipitation (250 g/L in phophate buffer) (Sigma Aldrich, St.Louis, MO), serum samples were mixed with equal volumes of PEG 1:1 and 1:2 PEG solutions, vortexed for 30 sec, incubated in room temperature for 10 min and by centrifuging at 10.000xg for 5 min, enzyme measurements were performed in the supernatant and serum. In our study, enzymatic measurements were done using kits of Cobas Integra 400 plus (Roche Diagnostics, Mannheim, Germany).

Recovery formulation is % recovery=(ASTPEG activity/ASTPBS activity)×100 (reference interval 42.0–82.2%) [1]. Precision values for AST were 2.33–2.64% and uncertainty for AST was 6.46% (These were mentioned in the article). For detecting heterophile antibodies, 500 ul sample was added into non-specific human anti-mouse antibodies (HAMA) tubes (Scantibodies Laboratory, Inc, Santee, CA) and mixed slightly for 5 minutes and then incubated in the room temperature for 1 hour. Enzymes would be measured after this procedure.

Based on abovementioned findings, in the patient in whom isolated AST was considered, an isolated increase of AST values were found as 77.1 U/L as a result of precipitation test performed with 1:2 PEG and as <5.4 U/L (recovery <0.8%) as a result of precipitation test performed with 1:1 serum: PEG. However, in measurements performed with HAMA tubes, AST was found to be 561 U/L. No significant changes were observed in ALT, GGT, ALP and CPK measurements performed via HAMA tubes (Table 1).

For 10 different concentrations, a recovery study of AST was performed, and the recommended recovery reference intervals of AST were detected to be between %60–140 (Figure 1).

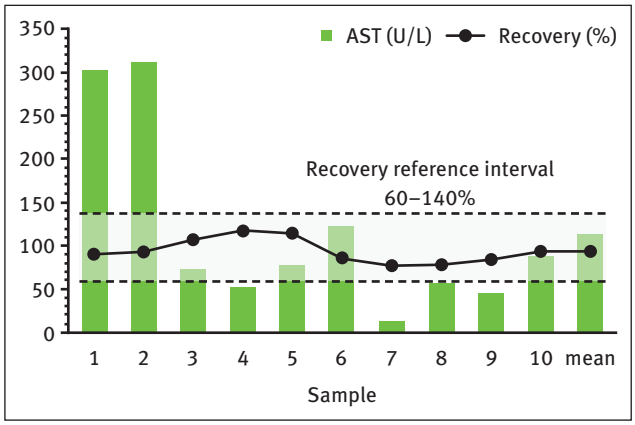

Figure 1: Recovery rates of AST measurements in 10 different concentrations via PEG method.

In light of these findings, an isolated increase of AST was strongly considered to be macroAST, and other medical staff and the patient's relatives were informed in terms of the existence of macroAST.

Discussion

In childhood period, higher rates of aminotransferases should be investigated as to the existence of liver and other systemic diseases. Twenty to 30% of liver diseases in childhood are composed of genetical and metabolic disorders. Despite large number of studies, increases of enzymes cannot be elucidated in various cases. AST are found in cardiac and skeletal muscles, kidneys, brain, pancreas, liver, lung and erythrocytes. The increase of muscle-caused transaminases is seen and accompanied by an increase in CK. Serum ALT is more beneficial in non-alcoholic fatty liver disease. Ultrasonography and liver function tests are used as primary care diagnostic procedures [2]. Another reason for AST increase in childhood diseases is myositis developing secondary to mycoplasmic pneumonia or viral infections. Myositis is generally seen following upper tract respiratory disease [3,4]; however, no symptoms or findings were observed in our case. ALT

and AST are mainly found in the cytoplasmic fraction of cells. As different from ALT, AST has also a mytochondrial form. The reason why ALT increases higher than AST depends on its half-life and cytoplasmic distribution (half-life of AST: nearly 16 h, half-life of ALT: nearly 50 h) [5,6]. Biochemical function of aminotransferases transfers the amino group of amino acides to keto acides by utilizing pyridoxal phophate (vitamin B6) as a co-factor. Such a reaction is vital for metabolismic maintenance. ALT is more specific for liver, compared to AST. Cardiac and skeletal injuries lead to more increase in AST, compared to that in ALT [7]. The reasons for non-liver increase of AST are cardiac, skeletal muscle and intestinal injuries, hemolitical anemia and hemolysis. Hepatitis is mainly caused by such reasons as viruses (e.g. hepatitis A, B, C), toxins like acetaminophen, use of alcohol, ischemia, Reye syndrome and autoimmunal diseases [2,5,7]. Aminotransferases may increase fifty times more than the referred values in viral, ischemic and toxic hepatitis cases [5,7].

Macroenzymes are non-specific increases of the enzymes in serum and generally known not to be clinically serious because some antibodies and molecules are bound to enzyme comlexes, and so mass of enzymes is increased, and the clearence of enzymes is decreased [8]. Macroenzymes of amylase, CK, ALP, AST, GGT, LDH and lypase have been reported [9,10]. Although the rate of macroamylasemia and frequency of macroLDH were reported as 0.98% and <1:10000 respectively, no rates related to the increases in macroAST have yet to be reported [10]. In two different studies performed with children, the increases of macroAST were reported [4,8].

Serum AST makes up macroenzymes in general by binding to immunuglobuline G and A [4]. Macroenzymes have been reported less in children and adolescents. MacroAST cannot be filtrated through renal glomeruli due to molecular expansion and remains in plasma [2]. Macroenzymes often lead to the interference of the results of serum enzymes, and thus giving rise to wrong diagnosis and treatment [2]. MacroAST, the macroenzyme form of AST, is considered to cause an isolated increase of AST [3]. In a study performed by Fortunato et al. in 10 children with asymptomatic high AST, four children were reported as macroAST [6]. MacroAST was suggested as a continuous and benign, but not a congenital entity [6,8]. In general, macroenzymes are not evaluated to exhibit pathological features [7]. Continuously increased values of enzymes have caused various invasive and expensive tests to be performed [7]. The determination of macroenzymes will enable monitoring tests and treatments to be avoided. Current methods used to determine mocroenzymes are electropheresis, precipitation with PEG or amonium sulphate and gel filtration chromatography [2,7,8]. As a limitation of our study, however, gel filtration could not be performed due to lack of technical equipment and opportunities. Still, in protein precipication assays performed with PEG or Cepharos to diagnose macroAST, it was reported that a decrease of >%95 in plasma AST levels may be assessed as macroAST [7,8].

Conclusion

Although rarely witnessed, macroenzymes cause asymptomatic clients to be exposed to unnecessary and troublesome procedures. Especially in children, the condition should be evaluated to be more dramatic. In cases with persistant and asmyptomatic high enzymes, macroenzymes should certainly be taken into account.

Conflict of Interest: The authors have no conflict of interest.

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