# Use of Plasminogen Activator Inhibitor-2 in Hepatocellular Carcinoma and Metastatic Tumors of The Liver

**Esin ÖZYILKAN, Halis ŞİMŞEK, Oktay ÖZDEMİR, Bülent SİVRİ, Şerafettin KİRAZLI, Burhan KAYHAN, Hasan TELATAR**

***Summary:*** *Plasminogen activator inhibitor-2 (PAI-2) and alphafetoprotein (AFP) levels were studied in 5 patients with hepatocellular carcinoma, 11 patients with chronic liver disease, 14 patients with liver metastasis, and 10 healty volunteers. AFP was detectable in the sera of 19 of 40 samples. PAI-2 level was detectable only in one patient with postnecrotic liver cirrhosis. We found no correlation between PAI-2 and AFP.*

**Key Words:** Liver diseasis, plasminogen activator inhibitör-2, alphafetoprotein

Plasminogen activator inhibitor-2 (PAI-2) was routinely detected in plasma and placental extracts of pregnant women (1,2). In placenta, PAI-2 is fetal origin (3). PAI-2 antigen levels become detectable at about 8 weeks of gestation and increase continuously until term (4,5). Kruithof et al. (6) studied PAI-2 antigen levels in 295 patients with different pathological conditions and high values were only observed in pregnancy and one patient with hepatocellular carcinoma. In 1989 Tran-Thang et al. (7) showed elevated PAI-2 levels in two patients with liver cirrhosis and one patient with hepatocellular carcinoma, therefore, he raised the question that PAI-2 was probably an oncofetal marker.

Alphafetoprotein (AFP) is produced in the fetal liver, yolk sac, and gastrointestinal tract, and is the major serum protein of the fetus though its precise function is unknown. It consist of a single polypeptide chain, with a molecular weight of 70.000 and alpha-electrophoretic mobility (8). There is a physiologic elevation of AFP in pregnancy (9). Elevations of AFP are found in patients with benign liver diseases, hepatocellular carcinoma, testicular carcinoma, embryonel cell cancer, and metastatic disease of the liver (10). Using a radioimmunaossay, Waldmann and McIntire (8) reported AFP levels above 48 ng/mL in 72% of patients with hepatocellular carcinoma. AFP is elevated in 16% to 25% of patients with benign liver diseases. In four of the 50 patients with metastatic to liver, Wepsic (11) found elevated AFP levels.

Hacettepe University Medical Center, Department of Internal Medicine, Divisions of Gastroenterology and Hematology, Ankara, Turkey.

We aimed to investigate whether PAI-2 is associated with certain liver diseses as an oncofetal marker like AFP.

## PATIENTS and METHODS

Eleven patients with chronic liver disease (2 alcoholic and 9 postnecrotic), 5 patients with hepatocellular carcinoma, 14 patients with liver metastasis (6 adenocarcinoma, 3 squamose cell lung cancer, 3 small cell lung cancer, and 2 breast cancer), and 10 healthy volunteers were included in this study. The median age of these groups were 46, 61, 48, and 47 years, respectively. None of them was pregnant.

Blood samples were obtained by venipuncture,

**Table I:** AFP and PAI-2 levels

| Groups | undetectable AFP | >2 ng/mL | undetectable PAI-2 | >6 ng/mL |
|---|---|---|---|---|
| Hepatocellular carcinoma | | 5 | 5 | |
| Chronic liver disease | 6 | 5 | 10 | 1 |
| Liver metastasis | 7 | 7 | 14 | |
| Healthy volunteers | 8 | 2 | 10 | |

anticoagulated with sodium citrate, placed on ice, immediately processed, and frozen at -78°C until analysed. The plasma concentration of PAI-2 was determined by means of an enzyme-linked immunosorbent assay (Biopool TintElize, Sweden). It detects both the low molecular weight (46.6 kD) form found in placental tissue and the glycolyslated high molecular weigt (60 kD) form found in maternal blood. The sensitivity of the assay was 6ng PAI-2/mL. Serum levels of AFP activity was quantified using an immunoradiometric assay. The sensitivity of this assay was 2 ng AFP/mL.

## RESULTS

AFP was detectable in the sera of 19 of 40 samples (Table 1). AFP was 11.8, 14.5, 380.0, 487.0, and 3300.0 ng/mL in five patients with hepatocellular carcinoma. We found detectable AFP levels in 7 patients with tumors metastatic to the liver. AFP was found detectable in 5 patients with chronic liver disease and in 2 healthy volunteers. PAI-2 level was detectable only in one patient with postnecrotic liver cirrhosis. PAI-2 level was 20.7 ng/mL and AFP was 30.0 ng/mL in this patient. All other detectable AFP levels were less than 8.4 ng/mL. We found no correlation between PAI-2 and AFP.

## DISCUSSION

PAI-2 was first identified in extracts of human placenta (1,2) and is present in and secreted by monocytes and macrophages (12,13). PAI-2 antigen levels increase throughout pregnancy (4,5). Placenta is the key source of PAI-2 and patients with preeclampsia were found to have decreased PAI-2 (14). Tran-Thang et al. (7) found PAI 2 levels of approximately 45 ng/mL in two patients with alcohol induced cirrhosis and 66 ng/ml in one patient with hepatocellular carcinoma. Kruithof et al. (6) reported PAI-2 level of 75 ng/mL in a patient with hepatocellular carcinoma. AFP was not determined in these two studies. In another study no correlation was found between AFP and PAI-2 (Grimaudo and Kruithof, unpublished observation).

Heyward et al. (15) performed a screening study of AFP in 1394 hepatitis B surface antigen-positive Alaskan natives. Patients with levels over 25 ng/mL were observed with montly AFP determinations. Patients with rapidly raising levels over 400 ng/mL were carefully investigated for hepatocellular carcinoma. Of the 126 patients with AFP levels over 25 ng/mL, 100 were attributable to pregnancy and 9 of remaining 26 patients had hepatocellular carcinoma. A correlation between the incidence of AFP elevation and tumor burden has been reported by some authors (16,17). Alpert et al. (17) reported that AFP producing tumors were larger and less differentiated than AFP negative tumors. Following surgical resection of tumor, there is a dramatic decline in AFP levels (10). A persistent elevation reflects residual disease (18,8). AFP also correlates with response to chemotherapy and slow and incomplete declines accompany an incomplete response (18). The serum half-life of AFP is 3.5 to 6 days and from this, we can predict the time of normalization of serum levels after treatment (10).

In conclusion, this preliminary study does not support the idea that PAI-2 is a useful marker in the diagnosis of hepatocellular carcinoma and liver metastasis like AFP. To conduct more precise studies it will therefore be necessary to develop more sensitive assays for PAI-2.

## REFERENCES


1. Lecander I, Astedt B. Isolation of a new specific plasminogen activator inhibitor from pregnancy plasma. Br J Heamatol 1986; 62: 221-8.
2. Astedt B, Lecander I, Ny T. The placental type plasminogen activator inhibitor, PAI-2. Fibrinolysis 1987; 1: 203-8.
3. Astedt B, Hagerstrand I, Lecander I. Cellular localisation in placenta of placental type plasminogen activator inhibitor. Thromb Haemostas 1986; 56: 63-5.
4. Lecander I, Nilsson I, Astedt M. Depression of plasminogen activator activity during pregnancy by placental inhibitor PAI-2. Fibrinolysis 1988; 2: 165-7.
5. Kruithof EKO, Tran-Thang C, Gudinchet A, et al. Fibrinolysis in pregnancy: A study of tissue plasminogen activator inhibitors. Blood 1987; 69: 460-6.
6. Kruithof EKO, Gudinchet A, Bachmann F. Plasminogen activator inhibitor 1 and plasminogen activator 2 in various disease states. Thromb Haemostas 1988; 59: 7-12.
7. Tran-Thang C, Fasel-Felley J, Pralong G, et al. Plasminogen activators and plasminogen activator inhibitors in liver deficiencies caused by chronic alcoholism and infectious hepatitis. Thromb Haemostas 1989; 62: 651-3.
8. Waldmann TA, McIntire KR. The use of radioimmunoassay for alpha-fetoprotein in the diagnosis of malignancy. Cancer 1974; 34: 1518-15.
9. Egan ML, Engvall E, Ruoslaht, EI, et al. Detection of circulating tumor antigens. Cancer 1977; 40: 458-66.
10. Bates SE, Longo DL. Use of serum tumor markers in cancer diagnosis and management. Semin Oncol 1987; 14: 182-38.
11. Wepsic HT. Alpha-fetoprotein: Its quantitation and relationship to neoplastic disease. In: Alphafetoprotein. Laboratory Procedures and Clinical Applications. Kirkpatrick AM, Nakamura RM (eds). Masson, New York 1981; pp 115-29.
12. Kopitar M. Isolation and some characteristics of urokinase inhibitors isolated from pig leukocytes. Haemostasis 1981; 10: 215-32.
13. Saksela O, Hovi T, Vaheri A. Urokinase-type plasminogen activator and its inhibitor secreted by cultured human monocytes-macrophages. J Cell Physiol 1985; 122: 123-32.
14. Amparo E, Gilabert J, Andreu C, et al. Plasminogen activator inhibitors type 1 and type 2 and plasminogen activators in amniotic fluid during pregnancy. Thromb Haemostas 1990; 64: 281-5.
15. Heyward WL, Lanier AP, McMahon BJ, et al. Early detection of primary hepatocellular carcinoma. Screening for primary hepatocellular carcinoma among persons infected with hepatitis B virus. JAMA 1985; 254: 3052-4.
16. Masseyeff R. Human alpha-feto-protein. Pathol Biol 1972; 20: 703-5.
17. Alpert E, Hershberg R, Schur PH, et al. Alphafetoprotein in human hepatoma: Improved detection in serum, and quantitative studies using a new sensitive technique. Gastroenterol 1971; 61: 137-43.
18. Matsumoto Y, Suzuki T, Ono H, et al. Response of alphafetprotein to chemotherapy in patients with hepatomas. Cancer 1974; 34: 1602-6.