Available online at www.sciencedirect.com
SCIENCE DIRECT®
JNMU
Journal of Nanjing Medical University, 2007, 21(2): 112-115
www.elsevier.com/locate/jnmu
Research Paper

The value of PAPP–A in the diagnosis and prognosis of GTD

Jing Fang[a,*], Shu Wang[a], Wen-li Gou[a]
[a]*Department of Obstetrics and Gynecology, the First Hospital of Xi'an Jiaotong University, Xi'an 710061, China*
Received 18 December 2006

Abstract

Objective: To investigate the value of PAPP-A (pregnancy assouated plasma protein-A) in the diagnosis and prognosis of gestational trophoblastic disease (GTD). **Methods**: The serum β-HCG(β-human chorionic gonadotropin) and PAPP-A levels of 25 normal pregnant women, 28 patients with complete hydatidiform mole and 38 patients with invasive mole were measured by enzyme linked immunosorbent assay (ELISA) during the periods of diagnosis, treatment and follow-up. **Results**: Compared with control group, patients with complete mole and invasive mole had higher levels of β-HCG ($P < 0.01$). But there was no significant difference between the complete and invasive mole group ($P > 0.05$). The PAPP-A level of complete mole group was significantly higher than that of control group ($P < 0.01$). The PAPP-A level of invasive mole group was significantly higher than that of complete mole group and control group($P < 0.05$). In complete mole group, serum β-HCG and PAPP-A levels of the patients with malignant sequelae were significantly higher than those with benign sequelae ($P < 0.05$). The β-HCG level had no relationship with the clinical stage of invasive mole. However, the PAPP-A level increased with clinical advancement of invasive mole. The levels of β-HCG and PAPP-A gradually decreased after evacuation in patients with complete moles, but maintained positive or even increased in patients with subsequent malignancy. **Conclusion**: The PAPP-A level can give us some help not only in early diagnosis of hydatidiform mole and invasive mole, but also in the prognosis of malignant sequelae.

Keywords: pregnancy assouated plasma protein-A; β-human chorionic gonadotropin; hydatidiform mole; invasive mole

INTRODUCTION

Originated from placenta and villous trophoblastic cell, gestational trophoblastic disease (GTD) includes hydatidiform mole, invasive mole and choriocarcinoma. Although β-HCG has been extensively used for diagnosis and follow-up of GTD, We have no way to differ malignant diseases from benign ones only by it. Even though continuously monitoring the β-HCG level can be advantageous in informing us the condition of trophoblast proliferation in some degree, However, it takes at least 4 weeks to have the result [1,2]. And, individual variation and other factors can cause some problems in the clinical practice of β-HCG [3]. Therefore, other associated markers may be necessary to use in diagnosing GTD and screening the high-risks.

Pregnancy-associated plasma protein A (PAPP-A) is a protein enzyme associated with insulin growth factor bound protein 4 (IGFBP4)[4,5]. Previous studies proved that PAPP-A was specifically secreted by syncytiotrophoblast and could reflect the physiological secretion function of this cell in large degree. Accordingly, we supposed that PAPP-A might reflect the differentiation and growth of trophoblast in normal placenta, as well as in GTD. This study measured the serum levels of β-HCG and PAPP-A in patients with hydatidiform mole and invasive mole pretherapy, before diagnosis, during treatment and follow-up. Compared with normal pregnant women, we aimed at investigating whether PAPP-A could be taken as a reliable marker used for high-risk group screening, early diagnosis of malignantcy, treatment effectiveness monitoring, and prognosis evaluation.

MATERIALS AND METHODS

Cases

*Corresponding author.
E-mail address: fangjing88993@126.com.

Sixty-six GTD patients treated in the first Affiliated Hospital of Medical College of Xi'an Jiaotong University from 2002 to 2005 were involved in the study. 25 normal pregnant women examined regularly in out-patient department from August to November 2005 were taken as the control group. Group of hydatidiform mole: 28 patients were treated with uterine evacuation. Before the evacuation, no metastasis was found and B ultrasound showed the sonogram of hydatidiform mole. Pathological diagnosis of all cases after operation was complete hydatidiform mole. The age of patients ranged from 18 to 32 years old with a mean of 28.2 years old. The average period of amenorrhea was 10.5 weeks. According to the sequelae of disease, the cases were divided into malignant sequelae group and benign sequelae group. Group of invasive mole: among 38 patients, 10 were in stage I, 5 in stage II, 21 in stage III and 2 in stage IV. They aged from 23 to 46 years old (mean 28.3 years). The average period of amenorrhea was 11.2 weeks before first therapy. Control group: twenty-five normal pregnant women with similar age and pregnant time were taken as control group.

Methods

Blood samples (5 ml) were obtained from veins, and the sp ecimeat were separated by centrifugation and stored at -20℃ until later analysis. The serum β-HCG and PAPP-A levels were measured by ELISA method, according to the manufacture's instructions. The test kits were purchased from American DSL Company. The levels of β-HCG and PAPP-A were determined according to the standard curves.

Statistical analysis

Statistical software package SPSS 13.0 was used. All data were presented as mean±SD and analyzed with *t* test. $P < 0.05$ was considered statistically significant.

RESULTS

β-HCG and PAPP-A levels of normal group, hydatidiform mole group and invasive mole group

The β-HCG levels of hydatidiform mole group and invasive mole group were obviously higher than that of control group, with significant statistical difference ($P < 0.01$). However, the β-HCG level was of no significant difference between the hydatidiform mole and invasive mole group ($P > 0.05$). The PAPP-A level of hydatidiform mole group was significantly higher than that of control group ($P < 0.01$). The PAPP-A level of invasive mole group was also significantly higher than that of control group and hydatidiform mole group($P < 0.05$) (***Tab 1***).

Relationship between the sequelae of hydatidiform mole patients and the levels of β-HCG and PAPP-A before uterine evacuation

The β-HCG level and PAPP-A level of the patients with malignant sequelae were significantly higher than those with benign sequelae($P < 0.05$, ***Tab*** 2).

Tab 1 **β-HCG and PAPP-A levels of control group, hydatidiform mole group and invasive mole group** ($\bar{x} \pm s$)

Group	Patients	β-HCG (KIU/L)	*P* value	PAPP-A (IU/L)	*P* value
Control group	25	31.40 ± 13.75	–	1.89 ± 1.48	–
Hydatidiform mole group	28	165.30 ± 38.20	< 0.01	5.21 ± 2.11	< 0.01
Invasive mole group	38	170.20 ± 40.18*	< 0.01	9.49 ± 3.43**	< 0.01

P value: Compared with control group; Compared with Hydatidiform mole group, *$P > 0.05$; **$P < 0.05$.

Tab 2 **β-HCG and PAPP-A levels of benign and malignant sequelae group** ($\bar{x} \pm s$)

Group	Patients	β-HCG (KIU/L)	*P* value	PAPP-A (IU/L)	*P* value
Benign sequelae	20	145.01 ± 36.02	< 0.05	2.29 ± 1.02	< 0.01
Malignant sequelae	8	216.03 ± 50.58		6.38 ± 2.55	

Relationship between β-HCG and PAPP-A levels and clinical stages in invasive mole group

The β-HCG levels showed no relation with clinical stage of invasive mole ($P > 0.05$). With the progress of clinical stage, serum PAPP-A level elevated. The PAPP-A levels of stage III and stage IV were much higher than those of stage I and stage II with significant difference ($P < 0.05$, ***Tab 3***).

Tab 3 **Relationship between β-HCG and PAPP-A level of invasive mole group and clinical stages** ($\bar{x} \pm s$)

Clinical stage	Patients	β-HCG (KIU/L)	*P*	PAPP-A (IU/L)	*P*
Stage I-II	15	169.37 ± 44.74	< 0.05	7.12 ± 2.76	< 0.05
StageⅢ-Ⅳ	23	162.65 ± 40.51		11.04 ± 3.87	

Change of β-HCG and PAPP-A levels of hydatidiform mole and invasive mole during the treatment and follow-up

Among 28 patients with hydatidiform mole, the β-HCG and PAPP-A levels of 20 cases with benign sequelae descended quickly after uterine evacuation. The β-HCG level dropped down fast during the first three weeks and reached normal level (<5IU/L) in the fourth week after evacuation. While the PAPP-A level fell sharply during the first two weeks and could not be detected at the end of the third week. The β-HCG levels of the 8 patients with malignant sequelae elevated in the second and third week after operation. The PAPP-A levels dropped quickly at first, then elevated again. At the same time, 2 cases of uterus myometrium infiltration and 1 case of vaginal metastasis were found. Chest X-ray films of 3 cases showed pulmonary metastasis.

DISCUSSION

HCG had higher titer in GTD, due to hyperplasia of tropholastic cells[6]. As the distinctive structure of HCG, β-HCG was con sidered as a specific marker for gestation and associated disorders [7,8]. β-HCG could be used to identify hydatidiform mole from normal one. In previous literatures and our study, there was no significant difference of serum β-HCG between hydatidiform mole and invasive mole. And it also failed to distinguish benign mole from malignant mole. PAPP-A is a kind of macroglobulin produced by placental syncytiotrophoblast and decidual cell. It is detectable as early as the 6th gestational week, increasing gradually as pregnancy continues and reaching a peak at the last trimester of pregnancy [5,9]. According to other researches [10], we presumed that PAPP-A could reflect the differentiation and growth of trophoblastic cells either in normal placenta, or in GTD tissue. Jaurliaux *et al* [11] found that serum PAPP-A levels in complete hydatidiform mole were much higher than those in normal gravida at the same gestational week. The study investigated the serum PAPP-A level of 25 normal pregnant women, 28 hydatidiform mole patients and 38 invasive mole patients. We found that pre-operation level of PAPP-A of hydatidiform mole was much higher than those of normal pregnant women. The level of invasive mole group was much higher than that of normal pregnant women and hydatidiform mole group. The results indicated that serum PAPP-A level could not only be used as a reference index for diagnosing hydatidiform mole, but also probably be applied to detect malignant diseases at early stage and therefore, was helpful in early treatment.

It is difficult in GTD treatment to predict the malignant change of hydatiform mole at early stage, especially before uterine evacuation. Studies showed that HCG played a role in tumor genesis and devel opment[12,13]. It could inhibit immune response through stimulating the integration of nucleotide and DNA and depressing the T lymphoblastic transformation caused by T dependent antigen [14]. HCG and its heteroplasmon could also affect the tumor autocrine accommodation by combining with other glycoprotein receptors (e.g., thyrotropin receptor). Hotakainen[15] found that β-HCG level of bladder transitional cell carcinoma elevated and it was positively related with clinical stage and pathological grade. It further proved that β-HCG acted as a stimulating factor in tumor growth and it could promote the genesis and development of tumor[16-18]. We found that the β-HCG level before evacuation in hydatidiform mole patients with malignant sequelae was obviously higher than that of those with benign sequelae, which suggested that β-HCG and its heteroplasmon might promote the genesis and development of GTD. PAPP-A is a kind of protein enzyme associated with insulin growth factor bound protein 4 (IGFBP4)[10]. It acts on mother-fetus interface to modulate the proliferation, differentiation and infiltration of trophoblastic cells in coordination with insulin-like growth factor (IGFs)and insulin-like growth factor bound proteins (IGFBPs)[19]. PAPP-A produced through autocrine or paracrine by tissue flanking cells can increase the biological activity of IGF and stimulate cell proliferation [20]. This study indicated that high PAPP-A level of hydatidiform mole before operation was related to later malignant change, meanwhile, the serum PAPP-A level was positively related to clinical stage of invasive mole, which implied that PAPP-A probably had some correlation to trophoblast proliferation and infiltration in GTD. It inferred that measuring PAPP-A levels in hydatidiform mole patients could and early diagnosing malignant change, and help to

distinguish patients with high risk. This study indicated that it was more sensitive and reliable to combine serum β-HCG level and PAPP-A level for screening high-risk GTD patients. Therefore, it was potentially beneficial in selecting chemoprophylaxis and in applying combined chemotherapy for high-risk patients.

The follow-up of patients with hydatidiform mole in our study showed that post-operation levels of β-HCG and PAPP-A fell quickly in patients with benign sequelae. Four weeks after operation, β-HCG level became normal, but PAPP-A could not be detected at the end of the third week. These results proved that PAPP-A could also be used as a reliable marker for hydatidiform mole following-up. Combined with β-HCG level, PAPP-A could be help ful in diagnosing malignant change and guiding treatment.

In conclusion, β-HCG level might be a reliable marker for diagnosis and follow-up of GTD. PAPP-A might be useful in early diagnosis, prognosis evaluation and follow-up. Especially, it was probably useful in distinguishing malignant diseases from benign ones at early stage. Therefore, it would be more reliable to combine β-HCG and PAPP-A in diagnosis and follow-up of GTD. Moreover, the detecting methods are convenient and easy to apply in practice.

References

[1] FIGO Oncology Committee. FIGO staging for gestational trophoblastic neoplasia 2000. *Int J Gynecol Obstet* 2002; 77: 285-7.

[2] Soper JT, Mutch DG, Schink JC, American College of Obstetricians and Gynecologists. Diagnosis and treatment of gestational trophoblastic disease: *ACOG Practice Bulletin NO*. 53. *Gynecol Oncol* 2004; 93: 575-85.

[3] Berkowitz RS, Goldstein DP. *Novak 's Gynecology*. 13th Edition. Philadelphia, Lippincott Williams &Wilkins, 2002: 1353-73.

[4] Bunn RC, Green LD, Overgaard MT, Oxvig C, Fowlkes JL. IGFBP-4 degradation by pregnancy-associated plasma protein-A in MC3T3 osteoblasts. *Biochem Biophys Res Commun* 2004; 325: 698-706.

[5] Handschuh K, Guibourdenche J, Guesnon M, Laurendeau I, Evain-Brion D, Fournier T. Modulation of PAPP-A expression by PPARgamma in human first trimester trophoblast. *Placenta* 2006; 27 Suppl A: S127-34.

[6] Kohorn EI. Persistent low-level "real" human chorionic gonadotropin: a clinical challenge and a therapeutic dilemma. *Gynecol Oncol* 2002; 85: 315-6.

[7] Cole LA, Sutton JM. HCG test in the management of gestational trophoblastic diseases. *Clin Obstet and Gynecol* 2003; 46: 523-40.

[8] Song HZ, Yang XY, Xiang Y. *Diagnosis and treatment of gestational trophoblastic tumor*, 2nd edition. Beijing, People's Medical Publishing House, 2004: 90-6.

[9] Cuckle HS, Vanlith JM. Appropriate biochemical parameters in first-trimester screening for Down syndrome. *Prenat Diagn* 1999; 19:505-12.

[10] Guibourdenche J, Frendo JL, Pidoux G, Bertin G, Luton D, Muller F, Porquet D, Evain-Brion D. Expression of pregnancy-associated plasma protein-A (PAPP-A) during human villous trophoblast differentiation *in vitro*. *Placenta* 2003; 24: 532-9.

[11] Jauniaux E, Bersinger NA, Gulbis B, Meuris S. The contribution of maternal serum markers in the early prenatal diagnosis of molar pregnancies. *Hum Reprod* 1999; 14: 842-6.

[12] Hayashi T, Arai G, Hyochi N, Suzuki M, Masuda H, Kawakami S, Okuno T, Ishizaka K, Kageyama Y, Kihara K. Suppression of spermatogenesis in ipsilateral and contralateral testicular tissues in patients with seminoma by human chorionic gonadotropin beta subunit. *Urology* 2001; 58: 251-7.

[13] Acevedo HF. Human chorionic gonadotropin (hCG), the hormone of life and death. *J Exp Ther Oncol* 2002; 2: 133-45.

[14] Kayisli UA, Selam B, Guzeloglu KO, Demir R, Arici A. Human chorionic gonadotropin contributes to maternal immunotolerance and endometrial apoptosis by regulating Fas-Fas ligand system. *J Immunol* 2003; 171: 2305-13.

[15] Hotakainen K, Haglund C, Paju A, Nordling S, Alfthan H, Rintala E, Stenman UH. Chorionic gonadotropin beta-subunit and core fragment in bladder cancer : mRNA and protein expression in urine , serum and tissue. *Eur Urol* 2002; 41: 677-85.

[16] Janssens JP, Verlinden I, Gungor N, Raus J, Michiels L. Protein biomarkers for breast cancer prevention. *Eur J Cancer Prev* 2004, 13: 307-17.

[17] Minamino K, Adachi Y, Okamura A, Kushida T, Sugi M, Watanabe M, Muguruma K, Sugao H, Suzuki Y, Iwasaki M, Nakano K, Koike Y, Wang J, Mukaide H, Zhang Y, Matsuda T, Matsumura M, Ikehara S. Autopsy case of primary choriocarcinoma of the urinary bladder. *Pathol Int* 2005; 55: 216-22.

[18] Kuroda H, Mandai M, Konishi I, Tsuruta Y, Kusakari T, Kariya M, Fujii S. Human ovarian surface epithelial (OSE) cells express LH/hCG receptors, and hCG inhibits apoptosis of OSE cells via up-regulation of insulin-like growth factor-1. *Int J Cancer* 2001; 91:309-15.

[19] Sun IY, Overgaard MT, Oxvig C, Giudice LC. Pregnancy-associated plasma protein A proteolytic activity is associated with the human placental trophoblast cell membrane. *J Clin Endocrinol Metab* 2002; 87:5235-40.

[20] Kong H, Wu ER, Ma X. Advanced studies of pregnancy associated plasma protein A. *J Reprod Med* 2003; 12: 176-80.