Cytokine Levels in a Patient with Severe Ovarian Hyperstimulation Syndrome Before and After the Ultrafiltration and Reinfusion of Ascitic Fluid

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Purpose: To report serum concentrations of several cytokines (interleukin-6, interleukin-8, tumor necrosis factor- α , and vascular endothelial growth factor) before and after the reinfusion of ultrafiltrated ascitic fluid.

Methods: A case report of a woman hospitalized for the treatment of severe OHSS at the Department of Obstetrics and Gynecology, Tottori University Hospital. The serum concentrations of cytokines were analyzed by ELISA.

Results: Cytokine concentrations declined in parallel with the improvement of clinical conditions and resolution of OHSS.

Conclusions: Measurement of serum cytokine concentrations may be useful in evaluating the severity of OHSS.

KEY WORDS: ovarian hyperstimulation syndrome; ultrafiltration and reinfusion; cytokine; growth factor.

INTRODUCTION

The pathogenesis of ovarian hyperstimulation syndrome (OHSS) has revealed that an increase in capillary permeability causes a fluid shift from the intravascular space into the abdominal cavity. In consequence, hypovolemia, hemoconcentration, and hyperosmolarity occur. Although the biochemical factors inducing capillary permeability in this syndrome have not been fully elucidated, several factors, such as LH, histamine, prostaglandins, and ovary derived-prorenin, seem to play a role in the pathogenesis of this syndrome. The development of severe OHSS is dependent on either exogenous administration of hCG or endoge-

nous, pregnancy-derived hCG stimulation. A recent study showed that a significant correlation was found between cytokine concentrations, such as interleukin-1 (IL-1), interleukin-6 (IL-6) and tumor necrosis factor- α (TNF- α) and certain biological characteristics of this syndrome (1). Vascular endothelial growth factor (VEGF), which is produced by luteinized granulosa cells and associated with follicular development and luteinization, is known as one promoting factor in OHSS. We present a case of severe OHSS treated with ultrafiltration and reinfusion of ascitic fluid and illustrate one role of cytokines in the development of OHSS.

CASE REPORT

The patient was a 32 year-old woman, gravida 0, para 0, with a 4 year history of infertility. She had been diagnosed with polycystic ovary syndrome by raised serum LH and the presence of polycystic ovaries depicted by ultrasound. Her husband's semen analysis revealed a volume of 2.4 ml, a sperm density of 2.3×10^6 ml, and 0% motility. She underwent ovulation induction using a GnRH agonist, FSH, and hMG for intracytoplasmic sperm injection in order to alleviate severe oligospermia. Briefly, Buserelin acetate (Suprecur, Hoechst Marion Roussel, Tokyo, Japan) was given intranasally at a daily dose of 900 μ g, starting during the midluteal phase. After the suppression of ovarian activity, controlled ovarian hyperstimulation was begun using FSH (Fertinome, Serono Japan, Tokyo, Japan) and hMG (Humegon, Organon Japan, Tokyo, Japan). On the 11th day after the commencement of ovulation stimulation, her serum estradiol concentration was ≥ 3882 pg/ml and an ultrasound examination showed a total of 43 follicles. After

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obtaining informed consent about the risk of hyperstimulation, a dose of 5000 IU of hCG (hCG Mochida, Mochida, Tokyo, Japan) was administered on that day and oocyte retrieval was performed 36 hr later. After the oocyte retrieval, the patient was immediately hospitalized from severe OHSS based on the classification proposed by Daniel *et al.* (1992), including oliguria, massive ascites, and pleural effusion. Three days after the oocyte retrieval, severe abdominal distension due to massive ascites, a decrease in urine output, and weight gain were observed. Embryo transfer was not performed to prevent her deteriorating condition. Intravenous liquid infusion and administration of osmotic diuretic were begun, but hypoproteinemia and oliguria continued to worsen. Therefore daily albumin supplementation (25 g/day) and low-dose-dopamine therapy (5 µg/kg/min) were added. Despite these therapies, both ascites and pleural effusion distinctly increased and she complained of respiratory distress. Ten days after the oocyte retrieval, we decided to perform abdominal paracentesis and reinfusion of ultrafiltrated ascites fluids. We used Plascit 01 (Asahi Medical, Tokyo, Japan) as the ultrafiltration and reinfusion system. Before ultrafiltration, 3600 ml of ascitic fluid was withdrawn, and the fluid was then ultrafiltered and concentrated to 400 ml. The concentrated ascitic fluid was intravenously reinfused for 2 hr. After the treatment, her general condition and laboratory data remarkably improved (Table I). One day after the treatment, hemoconcentration was improved and urine output dramatically increased. She was discharged 4 days after the reinfusion therapy. Before and after the procedure, we measured the serum concentration of several cytokines (Table II). The serum cytokine concentrations such as IL-6, TNF-α, IL-8, and VEGF were reduced after the treatment.

Table II. Cytokine Concentrations in Serum Before and After the Reinfusion of Ultrafiltrated Ascitic Fluid

	Before	After
IL-6 (pg/ml)	1050	19.4
TNF-α (pg/ml)	27.5	5.05
IL-8 (pg/ml)	10,400	640
VEGF (pg/ml)	442	308

DISCUSSION

Recent studies have shown that cytokines such as IL-1, IL-2, IL-6, IL-8, and TNF-α may be involved in the pathogenesis of OHSS. IL-6 and TNF-α have been implicated as mediators of the acute response occurring in sepsis, injury, and immunological change, a response characterized by capillary leakage, very similar to that seen in OHSS. IL-6 is a pleiotropic cytokine produced by various types of cells. IL-6 is found in the follicular fluid of patients undergoing ovarian stimulation and IL-6 mRNA is produced during ovarian follicular neovascularization (2). IL-6 has been immunolocalized to the human corpora lutea and is markedly elevated in the ascites and serum of patients with OHSS (3). These findings suggest that this cytokine may play a role in the increased capillary permeability and neovascularization in OHSS.

TNF-α has been known to stimulate ovarian cell growth and to participate in the regulation of follicular development. TNF-α originating in the ovary may exert its effects, interacting with specific granulosa cell surface receptors (TNF-α receptor type 2) to modulate gonadotrophin hormonal action. A significant increase in TNF-α activity was found in severe OHSS followed by a rapid decrease accompanying clinical resolution (4). Similarly, a marked decrease in the serum TNF-

Table I. Laboratory Findings

Test	Normal value	Day of admission	Ultrafiltration infusion		
			Before	One day after	Two days after
WBC	3300–8800	12700	12400	8400	7800
Hemoglobin (g/dl)	11.0–15.0	14.0	12.6	10.9	11.2
Hematocrit (%)	33.0–44.0	41.7	37.9	31.5	32.8
Total protein (g/dl)	6.0–8.0	5.8	5.0	5.2	5.9
Albumin (g/dl)	3.5–5.0	3.3	3.4	3.5	3.8
Blood urea nitrogen (mg/dl)	8–20	17	22	15	
Creatinin (mg/dl)	0.6–1.6	0.7	0.8	0.7	
Urine output (ml/day)		1350	1620	2990	4350
Abdominal circumference (cm)		67	84.6	76.5	74.2

α level was obtained after the reinfusion therapy in the present case.

IL-8 was found to mediate the transendothelial migration and activation of neutrophils at the site of inflammation. One study, similar to the present case, showed that high serum IL-8 concentrations were recorded in three of seven OHSS patients and decreased with clinical improvement. Extremely high IL-8 concentrations were also found in the ascitic fluid. These results suggest that IL-8 plays a role in the intraperitoneal acute inflammatory process in OHSS by inducing the transendothelial migration of neutrophils into the peritoneal cavity.

VEGF, known as vascular permeability factor (VPF), is thought to promote angiogenesis in normal tissues as well as tumors. It has long been known that the permeability enhancing capacity of this factor is more than 1000 times that of histamine. VEGF concentrations, in the serum of patients who developed OHSS, are elevated. Follicular fluid levels of VEGF after hCG administration is found to be 100-fold greater than serum or peritoneal fluid (5). Another study showed that the mRNA expression of VEGF in human luteinized granulosa cells is dose- and time-dependently enhanced by hCG in vitro (6). Considering these findings, hCG administration causes the production and secretion of VEGF in the granulosa cells which in turn may cause clinical symptoms of OHSS. Additionally, endogenous pregnancy-derived hCG may enhance expression of VEGF, making the clinical symptoms more severe.

Several reports have documented that decreasing levels of serum cytokine concentrations were found accompanying clinical resolution of OHSS. This is the first report showing that serum cytokine concentrations

are analyzed before and after ultrafiltration and reinfusion of ascitic fluid in severe OHSS. The ultrafiltration and reinfusion system can remove cells and bacteria without losing the protein. The advantage of this technique is that the patient's own protein and electrolytes can be used. In this case, serum cytokine concentrations dramatically fell along with the improvement of clinical symptoms. Although we measured the serum concentration of cytokines in only one case for the present, this result suggests that the measurement of serum cytokine concentrations may be useful in evaluating the severity of OHSS. Many questions are still unanswered and further study is necessary to completely understand the role of cytokines and growth factors in OHSS.

REFERENCES

1. Abramov Y, Schenker JG, Lewin A, *et al.*: Plasma inflammatory cytokines correlate to the ovarian hyperstimulation syndrome. *Hum Reprod* 1996; 11:1381–1386
2. Motro B, Itin A, Sachs L, *et al.*: Pattern of interleukin 6 gene expression in vivo suggests a role for this cytokine in angiogenesis. *Cell Biol* 1990; 87:3092–3096
3. Friedlander MA, Loret de Mola JR, Goldfarb JM: Elevated levels of interleukin-6 in ascites and serum from women with ovarian hyperstimulation syndrome. *Fertil Steril* 1993; 60:826–833
4. Balasch J, Arroyo V, Carmona F: Severe ovarian hyperstimulation syndrome: role of peripheral vasodilatation. *Fertil Steril* 1991; 56: 1077–1083
5. Krasnow JS, Berga SL, Guzick DS, *et al.*: Vascular permeability factor and vascular endothelial growth factor in ovarian hyperstimulation syndrome: a preliminary report. *Fertil Steril* 1996; 65:552–555
6. Joseph N, Zhaoping Y, Sonja R, *et al.*: Human chorionic gonadotropin-dependent expression of VEGF/VPF in human granulosa cells: Importance in ovarian hyperstimulation syndrome. *J Clin Endocrinol Metab* 1997;80: 1967–1971