Polycystic Ovary Syndrome and Ovarian Hyperstimulation Syndrome

(Polycystic ovarian syndrome, PCOS)

(0varian hyperstimulation syndrome, OHSS)　PCOS

cytochrome P450$_{c17}$　　　　　OHSS

(IL-6, IL-8, TNF-α)　vascular endothelial growth factor (VEGF)

OHSS

OHSS
PCOS

OHSS　　　OHSS

IL-6

OHSS

(IL-6, IL-8, TNF-α　VEGF)　　OHSS
PCOS

Abstract

Severe ovarian hyperstimulation syndrome (OHSS) is an important consideration when PCOS patients undergoing superovulation protocols. The risk of OHSS appears to be higher in these women undergoing ovulation induction. The exact mechanism for this increased sensitivity of the polycystic ovary to exogenous gonadotropins is not clearly understood, possibly as a result of cytochrome p-450c17 dysregulation. The pathogenesis of OHSS also remains poorly understood. The underlying mechanism response for the clinical manifestations of OHSS appears to be an increase in capillary permeability of mesothelial surface with acute fluid shift out of the intravascular space. The inflammatory cytokine (IL-6, IL-8, TNF-α) and vascular endothelial growth factor (VEGF) may be involved in accelerated metabolism in the growing follicles and angiogenesis occurred in the follicles of PCOS. If changes of these cytokines begin in the peri-ovulatory period, these may be used to predict more specifically which patients with PCOS will develop OHSS. The main purpose of this study is to examine these inflammatory cytokines in plasma and follicular fluid during peri-ovulatory period and correlated with serum steroid hormone in normal and PCOS patients undergoing in-vitro fertilization (IVF) and to assess the association between serum and follicular fluid cytokines concentrations and subsequently developing severe OHSS. This study suggests that elevated levels of IL-6 in the follicular fluid at the time of oocyte retrieval may predict early stage of OHSS. However, measurements of serum IL-6, IL-8, TNF-α and VEGF between the days of hCG administration and ET are not predictive for this syndrome. The mechanism for the increased sensitivity of the polycystic ovary to exogenous gonadotropins could not be explained by changes of inflammatory

cytokines in plasma and follicular fluid during peri-ovulatory period.

Keywords: Polycystic ovary syndrome, Ovarian hyperstimulation syndrome, Cytokine

(Polycystic ovarian syndrome, PCOS)

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　PCOS

MATERIALS AND METHODS

Subjects and Study Design

From September 1998 to March 1999, we recruited 156 consecutively IVF patients. The ovarian stimulation protocols were performed as previously described [1]. All patients were followed for the development of OHSS. According to the classification proposed by Galon et al. (14), 7 (4.5%) had moderate OHSS and 5 (3.2%) had severe OHSS. The study group comprised 12 patients who developed early-form moderate (n = 7) or severe (n = 5) OHSS required hospitalization. The two control groups comprised randomized selection of 12 high-risk women and 12 matched low-risk women in whom OHSS did not develop.In all patients, peripheral venous blood was drawn in the morning on the days of hCG administration, oocyte retrieval, and ET. During oocyte retrieval, clear follicular fluid from individual follicles of ≥ 18 mm was collected and pooled in each patient. The follicular fluid was centrifuged immediately (2,000 × *g*), and the supernatant was stored, together with the serum samples, at -70°C before assay for cytokines and steroids.

Cytokines and hormones assay

Levels of IL-6, IL-8 and TNF-α in serum and follicular fluid were measured with a solid-phase chemiluminescent enzyme immunoassay system (Immulite; Diagnostic Products Corporation, Los Angeles, CA). Serum and follicular VEGF concentrations were quantitated using an ELISA (Quantikine; R&D System, Inc., Minneapolis, MN). Serum and follicular fluid E$_2$ and progesterone concentrations were assayed using a chemiluminescent immunoassay (Immulite; Diagnostic Products Corporation).

RESULTS（　）

The clinical and laboratory data in the study and the two control groups are presented in Table 1. There are more patients with PCOS in study group than that in the two control groups.

Follicular fluid concentrations of IL-6 as well as P (Table 2) were significantly higher in the OHSS group than in the two control groups. Follicular fluid levels of IL-8, TNF-α, VEGF, and E$_2$ showed no statistically significant difference among the three groups.

Table 3 shows changes in serum levels of cytokine profile and steroids on the days of hCG administration, oocyte retrieval, and ET. Table 4 shows the correlations of cytokine

levels with steroid concentrations in serum and follicular fluid.

DISCUSSION(   )

The unique feature of our study is that we assessed multiple relevant cytokines in serum and follicular fluid simultaneously in patients who subsequently developed OHSS and matched controls who did not. We also examine the possible role of serial measurement of these mediators between the day of hCG administration and the day of ET in the prediction of OHSS. We found that only follicular fluid IL-6 levels at the time of oocyte retrieval can serve as an early predictor for the development of OHSS. We further expanded on the assay of circulating cytokines by showing that serum levels of IL-6 between the day of hCG administration and the day of ET are not predictive for this syndrome. This suggests the ovary may be the main resource of mediators that work as the initiators of the changes and lead to the full appearance of OHSS.

Our findings are concomitant with those of Krasnow et al. and Geve et al., who reported no difference in follicular fluid VEGF levels was detected between OHSS and controls. Based on our work and previous reports, circulating VEGF levels between the day of hCG administration and the day of ET do not appear to be a good marker or predictor of OHSS.

Our data illustrated the minimal cytokine variability in serum between the day of hCG administration and the day of ET. Therefore, it is unlikely that serum concentrations of these cytokines during this period could serve as early predictors for the development of OHSS. Moreover, the mechanism for the increased sensitivity of the polycystic ovary to exogenous gonadotropins could not be explained by changes of inflammatory cytokines in plasma and follicular fluid during peri-ovulatory period.

1. Chen CD, Wu MY, Chen HF, Chen SU, Ho HN, Yang YS. Prognostic importance of serial cytokine changes in ascites and pleural effusion in women with severe ovarian hyperstimulation syndrome. Fertil Steril 1999:72:286-92.

IL-6
OHSS
(IL-6, IL-8,
TNF-α   VEGF)   OHSS

PCOS

Table 1 Clinical and laboratory data in the study and control groups

| | Study group | Control group | |
| | OHSS | High risk | Low risk |
Variable	(n = 12)	(n = 12)	(n = 12)
No. of patients with PCOS (%)	7 (58.3)	3 (25.0)	1 (8.3)*
Age (y)	30.9 ± 4.8	33.7 ± 2.8	34.0 ± 6.2
No. of hMG ampules	23.5 ± 9.6	18.3 ± 5.7	19.0 ± 5.1
E_2 level on day of hCG administration (pg/mL)	3,607 ± 2,121	2,705 ± 757	1,117 ± 375*
No. of oocytes retrieved	23.4 ± 9.1	16.5 ± 8.8	7.1 ± 3.9*
No. of embryos transferred	5.3 ± 1.1	4.8 ± 1.1	3.7 ± 1.7*
Clinical pregnancy (%)†	8 (75)	4 (25)	4 (25)

Note: Values are means ± SD unless otherwise indicated. OHSS = ovarian hyperstimulation syndrome; PCOS = polycystic ovary syndrome.

* $P < .05$ (versus study group).

† No of patients who became pregnant (%).

Table 2 Follicular fluid concentrations of various cytokines and steroid hormones in the study and control groups

| | Study group | Control group | | |
| | OHSS | High risk | Low risk | |
Variable	(n = 12)	(n = 12)	(n = 12)	$P*$
Cytokine (pg/mL)				
IL-6	42.7 (0 to 106.6)	6.1 (3.7 to 8.6)	9.5 (3.6 to 15.4)	.026
IL-8	357.0 (0 to 716.2)	160.5 (91.5 to 229.6)	191.4 (127.5 to 255.4)	NS
TNF-α	21.7 (10.8 to 32.6)	10.4 (7.8 to 12.9)	14.1 (6.6 to 21.6)	NS
VEGF	2,989 (2,094 to 3,884)	2,492 (1,288 to 3,696)	2,945 (1,801 to 4,088)	NS
E_2 (ng/mL)	245.3 (139.7 to 350.9)	198.7 (70.4 to 327.0)	247.2 (171.5 to 289.3)	NS
Progesterone (ng/mL)	6,962 (4,585 to 9,339)	4,275 (3,140 to 5,409)	3,771 (2,437 to 5,105)	.043

Note: Values are means (95% confidence intervals) unless otherwise indicated.

* Determined by Kruskal-Wallis test.

Table 3 Changes in serum levels of various cytokines and steroids on the days of hCG administration, oocyte retrieval, and embryo transfer.

Patient group (Time point)	n	Cytokine level (pg/mL)				E_2§ (ng/mL)	Progesterone‖ (ng/mL)
		IL-6	IL-8	TNF-α	VEGF		
OHSS							
Day 0 (Day of hCG)	12	4.0 (1.9 to 6.1)†	34.5 (3.1 to 65.9)	7.9 (6.6 to 9.2)	153.3 (111.7 to 194.9)	3.61 (2.26 to 4.95)	1.8 (1.3 to 2.2)
Day 2 (Ovum pick-up)	12	2.2 (1.6 to 2.8)	6.1 (3.7 to 8.5)	7.6 (6.0 to 9.2)	110.3 (81.6 to 138.9)	2.27 (1.13 to 3.41)	15.6 (13.4 to 17.8)
Day 4 (Day of ET)	12	2.9 (1.9 to 3.8)	9.9 (4.2 to 15.5)‡	8.7 (6.9 to 10.5)	166.2 (94.6 to 237.8)	2.23 (1.38 to 3.08)	105.2 (57.8 to 152.7)
P value*		.095	.135	.590	.140	097	< .001
No OHSS, high risk							
Day 0	12	6.3 (2.3 to 10.3)†	39.3 (3.8 to 74.7)	7.5 (6.2 to 8.9)	204.4 (125.7 to 283.1)	2.71 (2.22 to 3.18)	1.4 (1.1 to 1.7)
Day 2	12	2.0 (2.0 to 2.0)	5.0 (5.0 to 5.0)	7.4 (6.1 to 8.8)	160.6 (84.2 to 237.0)	1.51 (1.12 to 1.92)	11.9 (6.6 to 17.2)
Day 4	12	2.1 (1.9 to 2.3)	5.1 (4.8 to 5.4)‡	10.3 (7.1 to 13.5)	184.2 (90.1 to 278.3)	1.57 (10.3 to 2.11)	142.0 (86.1 to 198.0)
P value*		.007	.043	.352	.674	.001	< .001
No OHSS, low risk							
Day 0	12	2.0 (2.0 to 2.0)†	5.1 (4.9 to 5.2)	9.7 (7.1 to 12.2)	183.8 (62.3 to 302.4)	1.12 (0.88 to 1.36)	1.0 (0.8 to 1.3)
Day 2	12	2.0 (1.9 to 2.1)	7.7 (4.1 to 11.3)	9.8 (7.8 to 11.8)	187.6 (68.4 to 306.7)	0.87 (1.13 to 1.97)	5.2 (3.8 to 6.6)
Day 4	12	2.1 (1.8 to 2.5)	5.1 (4.8 to 5.5)‡	9.8 (7.5 to 12.1)	187.3 (69.2 to 305.4)	0.64 (0.40 to 0.87)	33.1 (13.6 to 52.5)
P value*		.597	.101	.933	.922	.025	< .001

Note: Values are means (95% confidence intervals) unless otherwise indicated. IL = Interleukin; TNF = tumor necrosis factor; VEGF = vascular endothelial growth factor; OHSS = ovarian hyperstimulation syndrome.

* Determined by Kruskal-Wallis test.

† *P* = .015 by Kruskal-Wallis test.

‡ *P* = .017 by Kruskal-Wallis test.

§ *P* < .05 among the groups at each time-point, Kruskal-Wallis test.

‖ *P* < .05 among the groups at each time-point, Kruskal-Wallis test.

Table 4 Correlations of cytokine levels in serum and follicular fluid with steroid levels.

Spearman's correlation coefficient	Cytokine							
	IL-6		IL-8		TNF-α		VEGF	
	FF	Serum	FF	Serum	FF	Serum	FF	Serum
	(n = 36)	(n = 108)	(n = 36)	(n = 108)	(n = 36)	(n = 108)	(n = 36)	(n = 108)
IL-6	1.000	1.000	.082	.548†	.296	.058	−.069	.236*
IL-8	.182	.548†	1.000	1.000	.437†	.080	.397*	.186
TNF-α	.296	.058	.437†	.080	1.000	1.000	.180	.223*
VEGF	−.069	.236*	.397*	.186	.180	.223*	1.000	1.000
E_2	−.009	.379†	.114	.280†	−.201	−.281†	.047	−.046
Progesterone	−.008	.324†	.176	−.013	−.054	.067	.490†	.035

* Correlation was significant at the 0.05 level (2-tailed). FF = follicular fluid; IL = Interleukin; TNF = tumor necrosis factor; VEGF = vascular endothelial growth factor.

† Correlation was significant at the 0.01 level (2-tailed).