Objective: To evaluate the level of macrophage colony-stimulating factor (M-CSF) in serum in response to ovarian stimulation (group 1) in low-response (n = 26), moderate-response (n = 40), and high-response (n = 29) patients and to compare its changes (n = 23, group 2) throughout the menstrual cycle between pregnant and nonpregnant patients.

Design: Randomized controlled trial.

Setting: University IVF program.

Patient(s): Ninety-five women undergoing IVF.

Intervention(s): Serum and FF collection from 95 women.

Main Outcome Measure(s): The M-CSF concentration was determined by ELISA.

Result(s): The M-CSF levels in FF were higher than in serum. The M-CSF levels in serum increased from low-, through moderate-, to high-response patients; pregnancy rates were 11.5%, 22.5%, and 51.7%, respectively. Levels of M-CSF in serum increased throughout stimulation until the day of oocyte retrieval and decreased until ET. During the postretrieval days, from the day of ET, through implantation, to the day of confirmation of pregnancy, the M-CSF levels of those patients who became pregnant (n = 13) increased significantly and reached their highest level. After implantation the M-CSF level decreased slightly and reached a plateau during gestation.

Conclusion(s): Macrophage colony-stimulating factor is involved in follicle development and ovulation and could be an additional predictor for IVF outcome. (Fertil Steril 2010;93:116–23. ©2010 by American Society for Reproductive Medicine.)

Key Words: Macrophage colony-stimulating factor, serum, IVF, response, pregnancy
the absence of most individual cytokines, whereas leukemia inhibitory factor and interleukin (IL)-11 have indisputable roles in this process. In other cases, such as M-CSF, granulocyte macrophage colony-stimulating factor (GM-CSF), IL-1, and IL-6, the numbers of implantation sites or litter sizes are reduced when the cytokine is absent (21–26).

In the present study we describe the important role of the changes in serum M-CSF levels during the menstrual cycle, in the process of follicular maturation, ovulation, implantation, and pregnancy, and their response to ovarian stimulation with recombinant FSH (rFSH).

MATERIALS AND METHODS

Institutional review board approval was obtained at the beginning of this study.

Patients

From an original sample of 95 patients, serum and FF were collected on the day of follicular puncture (FP). The patients had tubal (excluding hydrosalpinges) or male factor infertility. Patients with endometriosis and polycystic ovary syndrome were excluded from this study. The age of the patients ranged from 20 to 42 years (median, 33 years), and the size of the lead follicle on the day of FP measured between 19 and 24 mm. These patients were divided into two groups, as follows.

Group 1 comprised 95 patients with tubal or male factor infertility, who were analyzed for [1] correlation between serum and FF with respect to M-CSF, and correlation between M-CSF and E2 in serum; and [2] comparison of M-CSF level in serum between pregnant and nonpregnant patients.

Group 2 comprised 23 of the 95 patients in group 1 with moderate response to ovarian stimulation. They were monitored throughout the menstrual cycle until 4 weeks after ET, as follows: at stimulation phase, including stimulation days (st.) 3–5, st. 6–8, st. 9–11, and day of hCG injection (Predalon; Organon, München, Germany); at oocyte retrieval (day of FP); on postretrieval days, including day of ET (2 to 3 days after FP) and 1 week after FP and embryo implantation (FP +1w); at 2 weeks after ET (day of β-hCG evaluation and confirmation of pregnancy) (ET +2w); and during gestation (3 and 4 weeks after ET) (ET +3w and ET +4w). One to three embryos at the four- to six-cell stage were transferred. Of the 23 patients, 13 became pregnant.

In this group, M-CSF levels in serum were analyzed throughout the different ovarian cycle phases and gestation (10 analyses for every pregnant patient and 8 analyses for every nonpregnant patient). The serum of all 23 patients was measured with the same ELISA M-CSF kit from the same lot number, to guarantee low intra- and interassay variances.

IVF Stimulation

Patients undergoing IVF-ET were stimulated with rFSH (Merck, Serono, Munich, Germany) after down-regulation with the GnRH agonist Synarel (Pharmacia, Erlangen, Germany). Monitoring of follicle development by real-time ultrasound scans and serum E2 levels was performed from day 5 until the day of FP. Once the leading follicle measured >17 mm in diameter and the 17β-E2 level was adequately increased but still <3,000 pg/mL in serum, 5,000–10,000 IU of hCG were administered SC. To avoid overstimulation, only 5,000 IU of hCG were injected in patients with E2 values exceeding 2,500 pg/mL or with >10 follicles. Progesterone levels were measured parallel to E2 and LH. Every increased level of P or LH was included into timing of hCG. Follicles were aspirated 36 hours after administration of hCG. After ET the patients were treated with P (Utrogest, 600 mg daily; Dr. Kade/Besins, Berlin, Germany) for luteal support until confirmation of pregnancy by β-hCG determination, 16 days after FP.

In response to ovarian stimulation, the patients were grouped as low (n = 26), moderate (n = 40), and high responders (n = 29), according to a scoring system based on the number of oocytes, total injected dose of rFSH (± SD) up to the day of hCG injection, and the increase in E2 levels.

Biochemical Analyses

Blood and FF were taken from 95 patients undergoing IVF-ET on the day of FP, processed by centrifuge for 10 minutes at 350 × g at 5°C, shock-frozen, and kept at −80°C. After pick-up of the oocytes the FF underwent the same treatment as the blood.

Macrophage colony-stimulating factor levels in serum and FF were measured in duplicate by a solid-phase ELISA using Quantikine M-CSF kit (R&D Systems, Wiesbaden, Germany). A fivefold dilution for serum and FF was performed with the Calibrator Diluent RD6F (R&D Systems). This assay uses the quantitative sandwich enzyme immunoassay technique. The M-CSF levels ranged from 31 to 2,000 pg/mL, with a sensitivity of 9 pg/mL. The M-CSF precision was <3% for intra-assay and <6.5% for interassay.

Only those cases in which both FF and serum could be simultaneously collected on the day of oocyte retrieval were included in this study.

Estradiol and P levels were measured by a solid-phase, competitive chemiluminescent enzyme immunoassay with the Immulite 2000 auto-system (DPC-Bierrmann, Siemens, Bad Nauheim, Germany) within the range of 0–2,000 pg/mL for E2 (sensitivity, 15 pg/mL) and 0.2–40 ng/mL for P.

Statistical Analysis

Statistical analysis was performed using SPSS (Chicago, IL). Pearson’s correlation coefficient (r) was applied to investigate the correlation between serum and FF with respect to M-CSF and between M-CSF and E2 in serum. To differentiate between groups, we used nonparametric procedures.
We performed a Kruskal-Wallis test to analyze differences in M-CSF levels among more than two groups (patients with low, moderate, and high response to ovarian stimulation). A Friedman test was applied for samples with repeated measurements and more than two groups, such as between M-CSF levels in serum throughout different ovarian cycle phases and gestation.

Paired comparisons were analyzed by the Mann-Whitney U test for unpaired and by the Wilcoxon signed rank test for paired samples. The differences in pregnancy rates among low-, moderate-, and high-response patients were analyzed according to a \( \chi^2 \) test. A \( P \) value of < .05 was considered statistically significant.

RESULTS

M-CSF Level in Serum and FF on the Day of Oocyte Retrieval

On the basis of normally distributed values of M-CSF and E\(_2\) levels in serum, we found a significant and positive weak correlation on the day of oocyte retrieval (Pearson \( r = 0.445, P = .001 \), determination coefficient \( r^2 = 0.196 \); Fig. 1). The mean (±SD) M-CSF level on the day of oocyte retrieval in FF (2,612 ± 72.59 pg/mL) was significantly higher than that in serum (282.25 ± 101.79 pg/mL) (\( P < .01 \), Wilcoxon signed rank test). There was also a significant and positive correlation between the M-CSF levels in serum and FF (Pearson \( r = 0.518, r^2 = 0.268, P < .001 \)).

There were no significant differences in M-CSF concentration in FF between follicles with fertilized oocytes and follicles with unfertilized oocytes. On the other hand, the level of M-CSF in follicular aspirates with oocytes (\( n = 78; 2,771.2 \pm 274.3 \) pg/mL) was higher than in those without (\( n = 17; 2,639.06 \pm 270 \) pg/mL), but the difference was not significant.

The mean M-CSF level in FF and in serum of patients who underwent intracytoplasmic sperm injection (ICSI) (\( n = 57; 2,632.76 \pm 809 \) pg/mL and 296.19 ± 111.69 pg/mL) was higher than in those who underwent IVF (\( n = 38; 2,580.6 \pm 597 \) pg/mL and 260.98 ± 81.3 pg/mL), but this difference was also not significant.

Additionally, no significant statistical differences were found between FF or serum M-CSF concentrations and patient age and body mass index in relation to the level of injected hCG dose for ovulation induction or the number of transferred embryos.

Relationship Between M-CSF Levels in Serum and Response to Stimulation and Pregnancy Rate

The M-CSF levels in serum (\( n = 95, \) group 1) express the response to ovarian stimulation with rFSH (Fig. 2). The patients were grouped as low, moderate, and high responders according to a scoring system based on the number of oocytes, total injected dose of rFSH (± SD) up to the day of hCG injection, and the increase in E\(_2\) levels. Table 1 shows the E\(_2\) levels and number of oocytes identified on the day of FP. The differences in mean rFSH and mean E\(_2\) levels among patients with low, moderate, and high response to ovarian stimulation were significant according to the Kruskal-Wallis test (\( P = .001 \)). Paired-comparison Mann-Whitney U test: high/moderate, \( P = .02 \); high/low, \( P = .001 \); moderate/low, \( P = .04 \).
were statistically significant according to the Kruskal-Wallis test (P = .001 and P = .003).

As seen in Table 1, patients with a low response had the highest injected dose of rFSH but the lowest mean level of E2 and the lowest number of oocytes (fewer than five). In these patients the lowest level of M-CSF (227.8 ± 54.1 pg/mL; n = 26) was determined. Patients with a moderate response had a medium injected dose of rFSH, a medium level of E2, and the number of retrieved oocytes ranged between 6 and 10. The level of M-CSF was determined at 278.6 ± 86 pg/mL (n = 40). Patients with a good response had the lowest injected dose of rFSH, the highest level of E2, and >11 oocytes. The level of M-CSF in these patients was the highest at 339.7 ± 112.8 pg/mL (n = 29). The differences in M-CSF levels among patients with low, moderate, and high response were statistically significant according to the Kruskal-Wallis test (P = .001).

Patients with a good response showed the highest pregnancy rate of 51.7% (Table 1). The pregnancy rate among patients with a moderate response was 22.5%, and an 11.5% pregnancy rate was seen in patients with a low response.

The differences in pregnancy rates among low, moderate, and high responders were found to be significant at P = .05, according to the χ² test. The 95 patients showed a total pregnancy rate of 28.5%.

**TABLE 1**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Low response</th>
<th>Moderate response</th>
<th>High response</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>26</td>
<td>40</td>
<td>29</td>
<td></td>
</tr>
<tr>
<td>Total rFSH dose (pg/mL)</td>
<td>3,860.9 ± 1,052.4</td>
<td>2,903.1 ± 1,214.5</td>
<td>2,541.8 ± 1,138.4</td>
<td>.001⁹</td>
</tr>
<tr>
<td>E2 (pg/mL)</td>
<td>733.72 ± 421.8</td>
<td>883.7 ± 411.5</td>
<td>1,230.3 ± 459.9</td>
<td>.003⁹</td>
</tr>
<tr>
<td>Oocytes (n)</td>
<td>&lt;5</td>
<td>6–10</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>M-CSF (pg/mL)</td>
<td>227.8 ± 54</td>
<td>278.6 ± 86</td>
<td>339.7 ± 112</td>
<td>.001⁹</td>
</tr>
<tr>
<td>Nonpregnant (n)</td>
<td>23</td>
<td>31</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>Pregnant (n)</td>
<td>3</td>
<td>9</td>
<td>15</td>
<td></td>
</tr>
<tr>
<td>Pregnancy rate (%)</td>
<td>11.5</td>
<td>22.5</td>
<td>51.7</td>
<td>.002⁹</td>
</tr>
</tbody>
</table>

*Note: Values are mean ± SD unless otherwise noted.*

⁹ Kruskal-Wallis test.
⁹χ² test.

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**M-CSF Expression During the Menstrual Cycle**

The evaluation of M-CSF levels in the serum of 23 patients (group 2: 13 pregnant and 10 nonpregnant) throughout the ovarian stimulation cycle and up to 3 to 4 weeks later is shown in Figure 3. A gradual increase of M-CSF from st. 3–5 through st. 6–8, st. 9–11, to the day of hCG injection is demonstrated, reaching a peak on the day of oocyte retrieval. The results are summarized in Table 2.

The differences within these groups, as analyzed by the Friedman test, were significant (P < .006). The levels of M-CSF in pregnant and nonpregnant patients on the day of oocyte retrieval were significantly higher than during st. 3–5, st. 6–8, and st. 9–11 (P < .001, Wilcoxon signed rank test). However, the increase in M-CSF level in pregnant patients from the day of hCG injection to the day of FP was significantly higher than in nonpregnant patients.

Up to the day of hCG injection there were no significant differences in the M-CSF levels of patients, whether they became pregnant or not.
After the day of hCG injection the M-CSF level in those patients who became pregnant increased significantly, whereas the level in the other patients increased only marginally. From the day of FP the M-CSF level decreased significantly in the pregnant and nonpregnant patients up to the day of ET ($P=0.034$, $P=0.038$). On the day of ET the M-CSF level in pregnant patients was significantly higher than in nonpregnant patients.

On the postretrieval days, from the day of ET through implantation (FP+1w) to the day of confirmation of pregnancy (ET+2w), the M-CSF levels of those patients who became pregnant (n=13) increased significantly and reached their highest level (Table 3). After implantation the M-CSF level decreased slightly ($P>0.05$) and reached a plateau during gestation (3 to 4 weeks after ET).

Those patients who did not become pregnant (n=10) showed no changes in M-CSF level from ET to implantation (FP+1w), respectively to ET+2w.

The differences in M-CSF levels among pregnant patients were significant according to the Friedman test ($P<0.006$). NS = nonsignificant (paired comparison by Wilcoxon signed rank test).

### TABLE 2
Comparison of M-CSF values in nonpregnant (n=10) and pregnant (n=13) patients during stimulation phase and on day of FP.

<table>
<thead>
<tr>
<th>Cycle phase</th>
<th>Pregnant total M-CSF (pg/mL)</th>
<th>$P$ value</th>
<th>Nonpregnant total M-CSF (pg/mL)</th>
<th>$P$ value</th>
</tr>
</thead>
<tbody>
<tr>
<td>st. 3–5</td>
<td>100.32 ± 14.1</td>
<td></td>
<td>94.35 ± 21.1</td>
<td></td>
</tr>
<tr>
<td>st. 6–8</td>
<td>110.77 ± 40.58</td>
<td></td>
<td>105.16 ± 21.7</td>
<td></td>
</tr>
<tr>
<td>st. 9–11</td>
<td>141.87 ± 35.8</td>
<td>0.02 b</td>
<td>163.82 ± 61.7</td>
<td>0.03 b</td>
</tr>
<tr>
<td>hCG injection</td>
<td>187.17 ± 32.4</td>
<td>&lt; 0.04 c, d</td>
<td>188.78 ± 39.6</td>
<td>0.012 c, NS d</td>
</tr>
<tr>
<td>FP</td>
<td>265.49 ± 88.4</td>
<td>&lt; 0.025 e, f</td>
<td>193.98 ± 76.7</td>
<td>0.008 e, NS f</td>
</tr>
<tr>
<td>$P$ value</td>
<td>.006</td>
<td></td>
<td>.003</td>
<td></td>
</tr>
</tbody>
</table>

Note: Values are mean ± SD. The differences within these cycle phases, as analyzed by the Friedman test, were significant ($P<0.006$). NS = nonsignificant (paired comparison by Wilcoxon signed rank test).

### TABLE 3
Comparison of the M-CSF and P levels of patients during postretrieval days and gestation.

<table>
<thead>
<tr>
<th>Postretrieval days and gestation</th>
<th>Pregnant total M-CSF (pg/mL)</th>
<th>$P$ value</th>
<th>Nonpregnant total M-CSF (pg/mL)</th>
<th>$P$ value</th>
<th>Pregnant P (ng/mL)</th>
<th>Nonpregnant P (ng/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ET</td>
<td>230.06 ± 109</td>
<td>.03</td>
<td>144.7 ± 31.99</td>
<td>.018</td>
<td>63.8 ± 32.8</td>
<td>62.6 ± 29.7</td>
</tr>
<tr>
<td>FP +1w</td>
<td>194.18 ± 69</td>
<td>NS c</td>
<td>150.98 ± 39.4</td>
<td>.05</td>
<td>18.5 ± 3.9</td>
<td>23.2 ± 11.1</td>
</tr>
<tr>
<td>ET +2w</td>
<td>338.1 ± 135.5</td>
<td>.006 d</td>
<td>146.61 ± 41.1</td>
<td>.016</td>
<td>203.6 ± 93</td>
<td>10.6 ± 7</td>
</tr>
<tr>
<td>ET +3w</td>
<td>278.08 ± 96.3</td>
<td>.07 d</td>
<td>181.9 ± 86</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ET +4w</td>
<td>235.56 ± 66.1</td>
<td>.04 d</td>
<td>147.8 ± 62</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$P$ value</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are mean ± SD. NS = non significant.

a Friedman test.
b Pregnant vs. nonpregnant patients, Mann-Whitney $U$ test.
c Wilcoxon signed rank test, vs. day of FP.
d Wilcoxon signed rank test, vs. FP +1w.
DISCUSSION

In the present study we measured the M-CSF concentration in FF in comparison with serum on the day of oocyte retrieval in patients with tubal or male factor infertility. Our results showed that M-CSF concentration in FF is significantly higher than in serum. Higher levels of M-CSF in FF compared with serum have also been reported by others (12, 13, 27). This implies an intrafollicular production and a potential autocrine or paracrine role of M-CSF within the follicular environment.

The mean M-CSF level in serum and in FF of patients who underwent ICSI was higher than in those who underwent IVF ($P = .08$). The reason for higher levels of M-CSF in ICSI patients may be that ICSI patients (mainly women with healthy ovaries, men with pathologic spermogram) produce more M-CSF in FF than do IVF patients (female cause of infertility). This is in accordance with our earlier finding relating to granulocyte (G)-CSF (28).

With regard to the response to ovarian stimulation with rFSH, M-CSF levels in serum on the day of FP increased from low-, through moderate-, to high-response patients ($P = .001$), and pregnancy rates rose from 11.5% to 22.5% to 51.7%, respectively. Similar results (9, 19) showed that ovarian stimulation with hMG leads to a gradual increase in M-CSF levels in patients with $>20$ follicles (good response) but not in those with $\leq 2$ follicles (poor response). Thus, M-CSF levels in serum may reflect successful stimulation and ample follicle maturation. Although the cause of poor response to gonadotropins is complicated and may consist of several dysfunctions of cytokines or growth factor networks, the defect in the mechanism of local (intrafollicular) M-CSF production could be one of the causes or results of poor ovarian response to gonadotropins. In this regard, the role of M-CSF in reproduction has recently been studied by a number of investigators (29–31). These findings were mainly from studies with osteoporotic (csfmop/csfmop) mutant mice. Female op/op mice show a relative paucity of ovarian macrophages. The M-CSF–deficient mice have extended estrous cycles, significantly lower ovulation rates, substantially lower numbers of both antral and mature follicles in the proestrus ovary, and a reduced proliferation capacity of granulosa cells in antral follicles. After administration of M-CSF all of these deficiencies could be compensated. These data provide evidence that M-CSF is implicated in the process of folliculogenesis and ovulation.

Our study is the first to measure the M-CSF level in serum in all cycle phases until 4 weeks after ET, to the time of gestation or new menstruation. In our results M-CSF levels in serum increased gradually throughout the ovarian stimulation cycle from st. 3–5, through st. 6–8, st. 9–11, and the day of hCG injection and reached a peak on the day of FP, indicating that gonadotropins influence M-CSF release. These results demonstrate that M-CSF is produced in the human follicular phase, immediately before the ovulatory phase, and plays an important role in folliculogenesis and in the mechanism of ovulation. These results correspond with the results of Nishimura et al. (9), Fukumatsu et al. (10), and Nishimura et al. (11), who showed that M-CSF and ovarian macrophages have a stimulatory effect on folliculogenesis, promoting ovulation in gonadotropin–primed immature rats.

After stopping ovarian stimulation with rFSH and administration of hCG, the level of M-CSF increased from the day of hCG to the day of FP. It seems that hCG also influences M-CSF release. Some investigators describe LH/hCG as a proinflammatory hormone that may well be involved in ovulation, an inflammatory-like process with macrophages migrating into the follicle and producing M-CSF; this suggests that intraovarian M-CSF, possibly induced by LH/hCG, plays an important role during ovulation and luteinization (10, 32–34).

Our results show clearly that in those patients who became pregnant the M-CSF level increased significantly from the day of ET through implantation and reached its highest level on the day of confirmation of pregnancy. After implantation the M-CSF level decreased and reached a plateau during gestation (3 to 4 weeks after ET). In contrast, those patients who did not become pregnant showed no changes in M-CSF level from the day of ET to the day of confirmation of pregnancy. In this regard, we observed the same distribution pattern in the values for P as for M-CSF in pregnant and nonpregnant patients. This points to a connection between these two factors. In accordance with our findings, Hatayama et al. (35) and Kanakzaki et al. (36) observed that M-CSF is produced in the human endometrial stromal cells and that it plays an important role in the reproductive process. Their findings indicate that human decidua cells express M-CSF and suggest that they secrete M-CSF in a P-dependent manner during the process of decidualization.

The characteristic expression profile of the M-CSF cytokine in the postretrieval days suggests that M-CSF plays an important role in the implantation process and in the maintenance of pregnancy. If implantation does not occur or fails, the M-CSF levels do not change. Because reduced M-CSF levels re-upregulated significantly at ET +2w, this may correspond to an invasion phase of the attached embryo(s) into the decidua. This acute rise of M-CSF may reflect an inflammatory process accompanied by invasion. We found a positive and significant correlation between P and M-CSF level at this time ($P = .02$, Pearson $r = 0.67$). This up-regulation could be caused by P (35–37).

Interestingly, the M-CSF level in pregnant patients was significantly higher on the day of oocyte retrieval and during the postretrieval phase (FP +1w, embryo implantation) than in nonpregnant patients. In this connection, it has been described that some cytokines, such as leukemia inhibitory factor, IL-11, G-CSF, M-CSF (i.e., CSF1), GM-CSF, IL-1, and IL-6 may have a more important function in achieving or maintaining pregnancy and may be essential members of the “implantation window” (21, 24, 25, 28, 38–43). The same cytokines that are implicated in implantation in mice
are generally maximally expressed in human endometrium, with maximal production in the secretory phase, particularly during the “implantation window.” Therefore, the high level of M-CSF during the luteal phase (implantation, day of confirmation of pregnancy, gestation) indicates the important role of this cytokine during the implantation process. The increase in M-CSF and its plateauing at a high level during the early pregnancy phase could serve as a pregnancy biomarker. Similar results (44) showed that M-CSF–treated embryos have significantly more trophoblast cells than control embryos. Furthermore, Guleria and Pollard (45) reported that trophoblast cells can take over M-CSF–regulated functions from the macrophages for the modulation of immune responses to invading pathogens at the maternal–fetal interface.

Moreover, the characteristic expression profile of the M-CSF cytokine during the menstrual cycle suggests that this cytokine is under the control of steroid hormones. In fact, stimulation of cytokine messenger RNA in endometrial cells by steroid hormones has been reported for M-CSF (46, 47), transforming growth factor β1 (48), and vascular endothelial growth (49). The amount of FSH is necessary to trigger follicular development. Estradiol, together with M-CSF, gives a better recognition of the beginning of pregnancy. Kligman and Rosenwaks (50) show that markers of ovarian reserve (day-3 FSH, inhibin, and E2) are particularly predictive and useful in guiding the choice for an optimal protocol for assisted reproductive treatment.

In conclusion, our data show that M-CSF is involved in follicle development and ovulation and plays an important role in the maintenance of pregnancy. It could also be a predictor of embryo implantation for IVF outcome.

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