Subcutaneously Administered Granulocyte Ceolony-Sstimulating Ffactor (G-CSF) Mmodulates Ovevarian Ffollicular Ffluid Cytokines During ~~an In Vitro Fertilization (IVF) cycle~~Ovarian Stimulation

N Mansouri-Attia[1], DR Ritsick[1], K Arnold[2], C Bommer[1], B Leader[2], J Braverman[1]
[1]Braverman Reproductive Immunology PC, Woodbury, New York, USA; [2]ReproSource, Inc., Woburn, Massachusetts, USA

Objective: During oocyte and follicle development, the intra-follicular microenvironment undergoes various regulations modulations resulting from the somatic-germ cells interactions and thatwhich may directly impacts the oocyte quality. Our study aims to investigated if recombinant granulocyte colony- stimulating factor (G-CSF) administration could was associated with changes in affect follicular fluid cytokine levels inin follicles retrieved from women undergoing ovarian hyper-stimulationIVF cycle.

Design: retrospective analysis

Methods and methods: Follicular fluid cytokines were measured from approximately 20 follicles in discarded samples from eEight randomly selected consenting women -including 4 women who received GCSF (mean age 38 years old) and 4 (mean age 38.5 years old) who did not. For each woman, 4 to 5 specimens were obtained each of which represented pooled aspirated fluid from 4 to 6 independent follicles.failed to become pregnant after repeated embryo transfers were selected and assigned to the control group (n=4, with 5 follicular fluid samples for each, mean age of 38 years old) or the G-CSF group (n=4 with 5 follicular fluid samples for each, mean age of 38.5 years old).

In addition to the luteal support, continuous Filgastrim, the recombinant G-CSF (Neupogen) was administerstrated by subcutaneous.c injection at 1µg (100,000 UIIU)//kg/day) through the IVF cycle, starting right aftersimultaneously with gonadotropin administration through oocyte retrieval. the birth control pill was stopped and until a pregnancy testβhCG testing was done. Follicular fluid samples weraes collected at the time of oocyte retrievafrozen at -80C within 1hr hour of collection. Levels of and levels of
2938 different soluble serum cytokines factors were screened formeasuredin the follicular fluid collected at the time of oocyte retrieval. Cytokine levels were measured in plasma by using the (Milliplex Map Human Cytokine Panel (Millipore Corp, St Charles, Mo) run on a the Luminex LX200 cytometer (Luminex, Austin, TX). Mean cytokine levels between control and GCSF treated patients were compared Comparisons between groups were made using an unpaired t-test.

Results: GCSF-treated women Results showed a significant regulation difference of in follicular fluid levels of six factors cytokines. Significant increases were observed in in follicular fluid by s.c. G-CSF: levels of G-CSF (3-fold, p vpalue <0.01), -macrophage-

Formatted: Font: Bold

Formatted: Font: Bold

Formatted: Space After: 0 pt, Line spacing: single, Don't adjust space between Latin and Asian text, Don't adjust space between Asian text and numbers

Formatted: Font: Bold

Formatted: Space After: 0 pt, Line spacing: single

Formatted: Space After: 0 pt, Line spacing: single, Don't adjust space between Latin and Asian text, Don't adjust space between Asian text and numbers

Formatted: Font: Bold

Formatted: Space After: 0 pt, Line spacing: single

derived chemokine (MDC, 1.3-fold, p ~~value~~< 0.05), and interferon-γ inducible protein 10 (IP-10, 1.79-fold, p ~~value~~< 0.001). Decreases were observed in ~~were significantly increased in G-CSF treated patients compared to controls while the anti-angiogenic factor~~ growth regulated oncogene (GRO, 1.~~-~~75-fold, p ~~value~~<0.05),~~,~~ interleukin 8 (IL-8, 1.76-fold, p ~~value~~ < 0.05) and monocyte chemoattractant protein-1 (MCP-1, 1.44-fold, p ~~value~~ <0.001). ~~were down regulated in G-CSF treated individuals.~~

Conclusions: ~~——~~Our data indicate that administration of G-CSF is associated with significant changes in follicular fluid cytokines including increased GCSF and IP-10 levels. As previous studies have demonstrated that increased follicular fluid GCSF and/or IP-10 are associated with increased oocyte yield, improved ~~showed that G-CSF administration during an IVF cycle induces a 3-fold increase in G-CSF follicular fluid levels that has been correlated with~~ embryo quality and higher successful pregnancy ~~outcom~~rates, it is possible that ~~e in previous published studies. Furthermore, we see an increase in IP-10 level in G-CSF treated patients, which has been previously positively associated with a successful pregnancy. In addition, GRO, IL-8 and MCP-1, three potent chemokines recruiting neutrophils and monocyte/macrophages and that are involved in a timely follicle rupture, were down-regulated by G-CSF administration. This could reflect the higher rate of ovulation in G-CSF treated patient.~~

Formatted: Font: Bold

Formatted: Space After: 0 pt, Line spacing: single