Objective: Elevated levels of tumor necrosis factor (TNF)-α have been reported in the peritoneal fluid of endometriosis patients, however its effect on oocyte and embryo is unclear. Therefore, the objective of this study was to examine the effect of exogenous TNF-α on oocyte spindle quality (microtubule morphology and chromosomal alignment) as well as on embryo development. Design: Prospective in vitro study Materials and Methods: Mouse TNF-α was diluted with human tubal fluid (HTF) to give concentrations ranging from 100 to 600 ng/mL. Immunohistochemical staining was used to evaluate the effect on oocyte microtubule morphology and chromosomal alignment. Fixed oocytes were incubated with anti-α-tubulin monoclonal antibody for microtubule staining, followed by incubation with FITC labeled anti-mouse IgG antibody. For chromosome staining, oocytes were incubated with propidium iodide. Stained oocytes were scored for alterations in microtubule morphology and chromosomal alignment under a Fluorescent (Leica, Germany) and scanning Confocal microscope (Leica Lasertechnik GmbH, Heidelberg, Germany). Scores of 1-2 were considered as being normal for oocyte microtubule morphology and chromosomal alignment, and 3-4 as abnormal (modified from Saunders and Parks, 1999). To evaluate embryotoxic effects, embryos were incubated with various concentrations of TNF-α and controls were incubated with an equal volume of HTF. Blastocyst development rate (% BDR) was examined after 72 hours of incubation. Results: A dose dependent increase in the percentage of oocytes with abnormal microtubule architecture and chromosomal alignment were seen with increasing concentration of TNF-α. Compared with controls, the rate of abnormal microtubule morphology (52% vs. 15.2%) and chromosomal alignment (60% vs. 20%) were significantly increased at 200 ng/mL (Fig. 1). A significant increase in embryo toxicity was seen only at concentrations >400 ng/mL compared with controls (%BDR: 51.1% vs. 98%) (Fig. 1).
Fig. 1. Effect of various concentrations of TNF-α on alterations in microtubule (MT) and chromosomal alignment (CH) in the oocyte and percent blastocyst development rate (%BDR) in the embryo.

Conclusion: TNF-α can exert deleterious effect on oocyte cytoskeleton that may compromise subsequent embryo development. Higher levels of TNF-α in peritoneal fluid of endometriosis patients may be responsible for the poor oocyte quality and embryo development in many endometriosis patients. Support: None

Author Disclosure Block: W. Choi, None; J. Banerjee, None; X. Zhang, None; R.K. Sharma, None; A. Agarwal, None; T. Falcone, None.

Category (Complete): Reproductive Biology: Animal and Experimental Models (RBPG)

Keyword (Complete): tumor necrosis factor-alpha (TNF-a) ; oocyte ; embryo ; endometriosis

Additional (Complete):
- Presenting Author Fellow: No
- In-Training Award: True
- ACCME Disclosure: I will not be discussing non-FDA approved products
- I Agree: True

Status: Complete