

Intrauterine administration of Lipiodol® enhances fertilization rates in mice inseminated with low sperm numbers

Edgar Del Llano¹, Marlene Rasschaert², Christophe Arnoult¹, Pierre F. Ray¹, Philippe Robert², Corinne Loeuillet¹.

¹ Team Genetics Epigenetics and Therapies of Infertility (GETI), Institute for Advanced Biosciences, INSERM U 1209, CNRS UMR 5309, Univ. Grenoble Alpes, 38000 Grenoble, France

² Research and Innovation Department, Guerbet Group, Roissy, France.

In the context of couple infertility, hysterosalpingography (HSG) is a common diagnosis used primarily to assess the patency of fallopian tubes in women. The technique consists in visualizing by radiography a contrast media injected into the uterus which fills the fallopian tubes before overflowing in the peritoneal cavity. Interestingly, several reports indicated a therapeutic side effect on fertility after HSG, with the observation of improved pregnancy rates, especially when using oil-based contrast media (Lipiodol®, ethyl esters of iodinated fatty acids of poppy seed oil) compared to water-based contrast media (1). However, the physiological mechanisms underlying this effect remain unknown.

In order to understand the beneficial effect of Lipiodol® administration on fertility, we developed a specific protocol using the mouse model. Lipiodol® was administrated into the mouse uterus, followed by intrauterine artificial insemination carried out two weeks later, corresponding to at least two mouse oestrus cycles (representing what is usually found in women, with pregnancy occurring usually two months after HSG). Mice were then sacrificed and all the oocytes/zygotes were collected and cultured in vitro to measure the fertilization rates.

In our first attempts, we observed comparable rates of fertilization ($67\pm 5.4\%$ and $60\pm 8\%$, $p>0.05$) from oocytes/zygotes originating from Lipiodol® treated and untreated control mice. This showed that, as practiced, uterine Lipiodol® injection has no deleterious effect but did not induce any improvement in fertilization. We postulated that a potential effect of the administration might be masked by the already optimal reproductive system of young wild type mice. We therefore decided to impair the reproductive efficiency of our model by mimicking oligozoospermia via inseminating the mice with a limiting number of spermatozoa. As expected, the number of fertilized oocytes was decreased but the overall fertilization rate was significantly improved in Lipiodol® administered mice compared to the controls ($29.9\pm 4.8\%$ and $12.6\pm 3\%$, $p<0.05$).

Overall, we established an amenable model to study the pro-fertility effect of Lipiodol® intrauterine administration and demonstrated that under a restrictive condition, Lipiodol® administration improved fertilization.

1- Dreyer, K. *et al.* Oil-Based or Water-Based Contrast for Hysterosalpingography in Infertile Women. *N Engl J Med* **376**, 2043–2052 (2017).