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Opinion

Refining Intracytoplasmic Sperm Injection Technique: How to Avoid Oocyte Degeneration

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Abstract

Clinical ICSI is the most frequently used procedure in modern human assisted reproduction. Despite the technique was firstly applied almost three decades ago, the quest for the highest fertilisation and pregnancy rates after ICSI still continues. The present article focuses on the event of oocyte degeneration post-ICSI and analyses how the use of ultra-thin injection needles and Laser-Assisted hatching can salvage fragile oocyte improving post-ICSI ova survival and clinical pregnancy rates.

Keywords: *Intracytoplasmic Sperm Injection; ICSI; Laser-Assisted ICSI; Oocyte Degeneration; Fertilisation*

In 1988, Lanzendorf., *et al.* [1] reported the first normal fertilisation in human after the injection of a single spermatozoon into an ovum. Four years later, in 1992, Palermo., *et al.* [2] reported the first pregnancies after microsurgically passing a single spermatozoon into the oolemma of a human ovum during in vitro fertilisation (IVF). The technique is known as intracytoplasmic sperm injection (ICSI).

Today, ICSI is being performed in every IVF clinic worldwide and ensures high fertilisation and pregnancy rates, especially when male or severe male factor infertility is being suspected. ICSI is also used in specialized techniques such as Preimplantation Genetic Screening/Diagnosis (PGS/D) or in pronuclear transfer techniques (PNT) in order to eliminate the possibility of either polyspermy or cross-contamination of samples by multiple spermatozoa bound on the zona pellucida (ZP) during conventional IVF.

The technique of ICSI is quite demanding for the performing clinical embryologist, requiring excellent hand-eye coordination and high micromanipulation skills. The gold aim is to avoid destroying the oolemma of an injected oocyte, allowing the development of the latter into a zygote 16 - 18hrs

post-injection. The phenomenon of unwanted oolemma breakage leads to ovum apoptosis, known as oocyte degeneration.

Oocyte degeneration can be predicted by the pattern of oolemma breakage during injection.

There are three distinctive oolemma breakage patterns during ICSI:

- A: The oolemma breaks suddenly during injection without aspiration.
- B: The oolemma breaks very difficult and there is a need for severe suction resulting in the formation of a large cytoplasmic funnel during ICSI and
- C: The oolemma breaks smoothly after a light suction resulting in the formation of a small cytoplasmic funnel during ICSI.

It has been recorded by ICSI practitioners that pattern A results in the highest degeneration rates after ICSI, patterns A and B result in the lowest fertilisation rates, while pattern C results in the highest fertilisation and survival rates after ICSI.

Fragility of oocyte membrane remains the most important factor resulting in high degeneration rates, even in patients with high oocyte number retrieved. When the zona pellucida is quite thick and hard to penetrate by the ICSI needle, the oocyte gets severely compressed and deformed in order the operating embryologist to be able to penetrate it and deliver the sperm in the middle of the oolemma. This compression though, results again in elevated oocyte degeneration and frequently in poor ICSI outcome.

There are two ways that excessive oocyte degeneration can be avoided during ICSI:

1. By using a very thin and spiked injection needle and
2. By Laser-Assisted ICSI (LA-ICSI).

ICSI injection needles are nowadays commercially available in different inner diameters and in a variety of tip morphology. It has been established though that the thinner and sharpest (spiked) an ICSI needle is, the survival of the oocytes and the ICSI outcome in general, dramatically improves. The ideal ultra-thin ICSI injection needle has an inner diameter of a maximum 4.5 μm and has a quite sharp and spiked tip. It is known by the laws of physics that the larger the surface of an object, the highest the pressure it exerts. Hence, a sharpest and more spiked injection needle tip due to its decreased surface, exerts lower pressure on the ZP and on the oolemma, resulting in improved oocyte survival rates.

LA-ICSI is a method that has been specifically developed to improve oocyte survival in clinical cases where oocyte fragility is known to exist. Elevated oocyte degeneration in a previous cycle in conventional ICSI, thick ZP, discoloured ZP and thawed oocytes, are some of the factors predisposing the oocytes to higher fragility and may result in poor outcome and survival after ICSI. In these cases, it is important to create a small incision at the "3 o'clock" position of the oocyte employing a 1.48-μm diode laser pulse. The opening should be around 5μm, just enough to allow a smooth passing of the injection needle through the ZP. In this way there is no increased pressure on the ZP that is being transferred to the oolemma resulting in its sudden breakage and oocyte degeneration. LA-ICSI is being used in many clinics nowadays with excellent results in oocyte survival, especially in cases where oolemma fragility is already known [3,4].

It is within the duties of the modern clinical embryologist to employ the abovementioned techniques in order to salvage fragile oocytes and to increase fertilisation and pregnancy rates in these vulnerable clinical cases. It is our duty to offer the patients the best treatment keeping in mind that each ovum is unique for them and could potentially be "The One" that will produce the so much wanted live birth.

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