Indications for intracytoplasmic sperm injection

L.Hamberger¹, K.Lundin, A.Sjögren and B.Söderlund

Department of Obstetrics and Gynecology, University of Gothenburg, S-413 45 Gothenburg, Sweden

¹To whom correspondence should be addressed

Intracytoplasmic sperm injection (ICSI) is the latest of several microfertilization techniques that have been utilized predominantly to overcome severe male subfertility, giving fertilization and term pregnancy rates similar to conventional in-vitro fertilization (IVF) (but for other indications). Even though available data on children born after ICSI are very encouraging, the procedure must still be considered as novel and the safety aspect to a great extent unexplored. In our opinion, therefore, ICSI should only be used for specific indications, and in this communication the non-existent, relative and absolute indications for performing ICSI are outlined and discussed. With an apparently normal sperm sample, ICSI should not be used in a first cycle even if only few oocytes are obtained. When there is reason to suspect poor fertilization, ICSI can be used in combination with conventional IVF in a split cycle. This includes cases of ‘subnormal’ sperm samples, high titres of antisperm antibodies, or following a single cycle of poor fertilization using conventional IVF. Absolute indications for ICSI include two previous fertilization failures with conventional IVF, use of epididymal or testicular sperm samples, or when only acrosomeless or immotile spermatozoa are available. The fertilization of oocytes prior to preimplantation genetic diagnosis is another absolute indication. It is, however, important to keep in mind that for this novel technique, indications should not be rigid, but remain variable with respect to new findings.

Key words: ICSI/IVF/indications/safety

Introduction

Microfertilization techniques were introduced clinically a decade ago, initially with the sole aim to overcome severe male subfertility. However, over the years other indications have also been included. Zona drilling, partial zonal dissection and the subzonal sperm injection technique were all demonstrated to work in principle, but their clinical success rate was disappointingly low (Cohen et al., 1991; Fishel et al., 1992; Vanderzwalmen et al., 1992; Wolf et al., 1992). This situation changed with the introduction of intracytoplasmic sperm injection (ICSI) since similar pregnancy rates could be obtained with this new technique as with conventional in-vitro fertilization (IVF) (Van Steirteghem et al., 1993, 1996). It should however be emphasized that comparisons between ICSI and conventional IVF in terms of success rates are probably not relevant, since they are performed on different classes of infertile couple, and the proposition that all IVF cases would have a better chance of success if treated with ICSI is not presently proven or valid. On the contrary, Petersen et al. (1996) showed that transferring couples with unexplained infertility and poor fertilization in conventional IVF to ICSI did not enhance their success rate. Taking this fact into consideration, it is not certain that the success rates of IVF and ICSI should be expected to be equal, and the only way to study this properly would be to perform a prospective randomized trial among couples for whom both IVF and ICSI is applied for fertilization. It has recently been suggested that, when fertilization is performed in natural cycles or if only two or three oocytes are obtained in ‘poor responders’, ICSI should be
Indications for ICSI preferred to conventional IVF (Norman et al., 1995). In our opinion the chances of fertilization of a very low number of oocytes are in most cases as good with conventional IVF as with ICSI. In this article, the optimal choice of fertilization technique will be discussed in relation to specific patient groups.

**IVF as a first choice**

In our programme when the husband presents with an ejaculated sperm sample which after preparation (swim-up or gradient centrifugation), has a total motile sperm count (TMC) exceeding \( \sim 0.8 \times 10^6 \) and a morphology (strict criteria) of >5% normal forms, then the couple will undergo conventional IVF in their first cycle, irrespective of the number of oocytes (Lundin et al., 1997). In cases of unexpected total fertilization failure reinsemination by ICSI can be performed on day 2 (Sjögren et al., 1995). However, even though term pregnancies have been reported with this strategy the success rate is very low (Lundin et al., 1996). Also, it cannot be excluded that risks of e.g. polyspermy (if fertilization has already occurred) and/or chromosomal damages are increased.

**IVF and ICSI in combination**

Following one IVF cycle with unexpected poor (<15%) or failed fertilization it may not be necessary to switch completely to ICSI in the following cycle. In a recent publication from our laboratory (Hamberger et al., 1995) the fertilization rate in a second cycle after initial poor fertilization was evaluated by dividing the oocytes, and inseminating half by ICSI and half by conventional IVF. It was found that 2/3 of the couples with poor fertilization in the first IVF cycle obtained good fertilization and pregnancy rates with routine IVF in the following cycle, while the remaining 1/3 still had no or poor fertilization with IVF, but good results with ICSI. This shows that the majority of IVF cycles with poor fertilization are due to transient factors, possibly non-optimal stimulation protocols or temporarily poor sperm samples. In Figure 1 a flow schedule is given of how the techniques can be selected and shifted dependent upon sperm sample quality and fertilization rate.

Other reasons for performing these ‘split’ or ‘50/50’ cycles are when the male partner presents with a sperm sample either with \(<0.8 \times 10^6 \) TMC after preparation, or < 5% normal forms. In these situations many clinics turn directly to the ICSI method. However, we think that, both for diagnostic purposes and in order not to use this very invasive method unnecessarily, the ‘oocyte split’ method is very useful.

In our laboratory we also use this split method for men with a high titre of antisperm antibodies (ASA), and have found that many of these cases achieve good fertilization and pregnancy rates using conventional IVF (Lundin and Hamberger, 1995), although certain subgroups of men with high ASA may need ICSI to obtain fertilization. It should be emphasized that the split technique, in order to be properly evaluated, is used only when the number of oocytes exceeds 10, otherwise ICSI will be performed.

The use of microdrops or insemination with an increased concentration of spermatozoa instead of ICSI has sometimes been advocated in cases of sperm samples with low numbers or poor morphology. Incubation with a higher number of spermatozoa has been shown to result in a higher fertilization rate, although the pregnancy rate did not increase (Grow et al., 1994; Ombelet et al., 1994).

**Absolute indications for ICSI**

In some cases ICSI should definitely be used as a first choice (Table I). This includes those couples described in the previous section, who had fertilization failure also in their second IVF cycle but good fertilization results with ICSI. In our opinion absolute indications also include all groups where non-ejaculated samples (i.e. spermatozoa aspirated from vas deferens, epididymis or the testis) are used. Even when aspirates with very good sperm concentration and motility are obtained from the epididymis, low fertilization and pregnancy rates are generally achieved using routine IVF (Silber et al., 1994). In a case of acrosome-deficient (round headed) spermatozoa, we were the first group to report successful pregnancy and healthy offspring following fertilization with ICSI (Lundin et al., 1994). In this case subzonal sperm injection had failed to achieve fertilization. Several similar
patients with immotile cilia syndrome frequently have 100% immotile spermatozoa or only few motile spermatozoa and thus constitute another absolute indication for ICSI. In this case a vital stain should be performed in advance to evaluate the chances of choosing a viable spermatozoon when performing the injection. The ICSI method can be combined with a hypo-osmotic swelling test (HOS) which will help to select viable sperm that can be used directly for ICSI (Chida, 1995; Esteves et al., 1996). This procedure can also be performed for testicular samples where often only immotile spermatozoa are found. Other subgroups where ICSI should be chosen are those in which fertilization is to be combined with preimplantation genetic diagnosis (PGD) since the use of conventional IVF leaves numerous spermatozoa stuck in the zona pellucida or in the perivitelline space. When blastomere biopsy is performed, such spermatozoa may ‘contaminate’ the biopsy and thus give incorrect diagnosis.

Utilization of frozen–thawed spermatozoa
Cryopreservation of ejaculated sperm causes a decrease in sperm viability and motility, but the thawed spermatozoa can still in most cases be used

Table I. The non-existent, relative, and absolute indications for intracytoplasmic sperm injection (ICSI)

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<th>IVF as first choice</th>
<th>IVF and ICSI in combination</th>
<th>ICSI as first choice</th>
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<tr>
<td>Normal sperm sample</td>
<td>No or poor fertilization in first cycle</td>
<td>No or poor fertilization in two IVF cycles</td>
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<td>Less than (0.8 \times 10^6) spermatozoa after preparation</td>
<td>Epididymal/testicular spermatozoa</td>
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<td>Sperm morphology &lt;5% normal</td>
<td>Globozoospermia</td>
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<td>Antisperm antibodies</td>
<td>Immotile spermatozoa</td>
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<td>Frozen-thawed spermatozoa with poor survival</td>
<td>Frozen–thawed spermatozoa with poor survival</td>
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<td>Preimplantation genetic diagnosis</td>
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IVF = in-vitro fertilization.
for conventional IVF after thawing. In cases with extreme impairment of motility after freezing–thawing and in cases of initial poor sperm quality, ICSI is of course preferred. We, along with other groups (Devroey et al., 1995; Hovatta et al., 1996; Holden et al., 1997), have found that spermatozoa obtained from the epididymis and from the testis usually survive well in the freezer and retain good fertilizing potential after thawing. It is to be recommended that in all cases where spermatozoa have been isolated from the epididymis or from the testis, excess spermatozoa should be frozen for use in later cycles. In such cases ICSI should of course always be applied to achieve fertilization. In our programme we have found exceptionally high fertilization and pregnancy rates using frozen–thawed epididymal spermatozoa. This may possibly be due to changes in the membrane structure of the spermatozoon, which may facilitate its decondensation within the oocyte. This is currently under investigation in our laboratory.

A ‘good’ epididymal sample can be frozen using a protocol similar to that of an ejaculated sample, i.e. with glycerol or egg yolk. Cohen et al. (1997) have developed a method of freezing a very low number spermatozoa inside an empty zona pellucida, an elaborate but promising technique for extreme cases. Testicular biopsies can be frozen either as tissue pieces, or after dissection and preparation.

Future indications

It is important to remember that the indications for ICSI should at present not be rigid. As more experience is gathered, new indications will appear, and old indications may well change. For example, Bertrand et al. (1995) found that the zona pellucida of fertilized oocytes in conventional IVF was significantly thinner than those of unfertilized oocytes. A thick zona pellucida may thus subsequently become an indication for ICSI (possibly in combination with assisted hatching). Another indication may be the acrosomal index of the sperm sample, i.e. the specific morphology of the acrosome (Menkveld et al., 1994).

Concluding remarks

In this short communication interest has been focused predominantly on various male factors

Indications for ICSI

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<td>oocyte penetration tests</td>
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Figure 2. Cross-checking of fertility within the couple by use of both donor oocytes and donor sperm. Possible sperm factors can also be studied using e.g. zona binding tests on salt-stored unfertilized oocytes, and oocyte penetration tests using zona-free hamster oocytes. Where ICSI may be the method of choice. The main advantage of this method is that the outcome is not directly related to any of the three basic semen parameters: the number of spermatozoa, their motility or their morphology (Nagy et al., 1995). The maturation stage, the acrosome reaction, the binding to the zona pellucida and the fusion with the oolemma are all bypassed. What is still required is the decondensation of the spermatozoon inside the oocyte and the activation of the oocyte. Failure of these events to occur are presumably due not only to sperm factors, but also to oocyte factors. Oocyte factors responsible for fertilization failure with conventional IVF are presumably not infrequent but are more difficult to demonstrate without the use of some type of cross-fertilization (Figure 2). In many countries legal restrictions make such cross-fertilization impossible. It may still be possible to study sperm binding to salt-stored unfertilized oocytes (Liu et al., 1989), which can give an indication as to whether an oocyte factor or sperm factors are responsible for problems in the binding process.

Complete fertilization failure when using ICSI is very unusual, and in most cases is presumably due to either failed oocyte activation or incomplete decondensation of the spermatozoon (Sousa et al., 1994; Flaherty et al., 1995). Again, this may be due to either an oocyte factor or a sperm factor. The approach of splitting oocytes for IVF and
ICSI in the same cycle may both improve the clinical success rate with minimal increase in workload, and also increase understanding of the reason for unsuccessful fertilization. Laboratories where the experience of the ICSI technique is limited may well produce better results with conventional IVF in cases of e.g. moderate teratozoospermia, where the indication to use ICSI is only relative. It therefore follows that the same guidelines cannot be applied in all laboratories. Also the couples’ own opinion and economic aspects must be taken into account when advice is given concerning the optimal technique for treatment. It is our impression that the enthusiasm raised by the good results achieved with ICSI have presently led to an uncritical overuse of this new technique. Until it is proven that ICSI does not introduce any increased risks for malformations and/or genetic defects, it is our opinion that the indications for its use should be strict and that the technique should be used only when properly indicated (Table I). From the above presentation it is obvious that indications for using ICSI may either be nonexistent, relative or absolute.

Acknowledgements

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References


activity, morphology (strict criteria) and fertilization in vitro. **Hum. Reprod.**, **9** (Suppl. 4), 99.


