Intracytoplasmic sperm injection (ICSI) is the direct introduction of a spermatozoon into an oocyte to achieve fertilization and pregnancy when the number of spermatozoa in the ejaculate is very low or absent. In the latter case, ICSI can be performed using spermatozoa obtained from the epididymis (Liu et al., 1994; Tournaye et al., 1994) or directly extracted from testicular tissue by biopsy or fine needle aspiration (Devroey et al., 1994; Silber et al., 1995; Kahraman et al., 1996; Lewin et al., 1996; Salzbrunn et al., 1996). Furthermore, techniques of spermatid injection into oocyte and first term pregnancies have been achieved in human (Tesarik et al., 1995, 1996).

Despite the worldwide diffusion of this procedure in the last years, only very recently have the possible risks that might ensue from its indiscriminate use been considered. This involves the whole scientific community and raises controversial opinions in scientific journals which are not specifically dedicated to the problems of human reproduction (Foresta et al., 1996a,b; Morris and Gleicher, 1996; Reijo et al., 1996).

ICSI arouses more fears of the transmission of genetic abnormalities to offspring than other forms of assisted reproduction because it bypasses all the physiological mechanisms related to fertilization. Physiological fertilization needs a spermatozoon able to active motility, to undergo a normal capacitation and acrosome reaction and to start all mechanisms required to penetrate the oocyte. By bypassing these steps, ICSI allows an altered spermatozoon to fertilize an oocyte, thus increasing the risk of genetic defects in the offspring.

Many reports describe a high frequency of chromosomal abnormalities, especially of the sex chromosomes, after ICSI, even when peripheral chromosome studies in the parents are normal (Bonduelle et al., 1995; Liebaers et al., 1995; In’t Veld et al., 1995; Tournaye et al., 1995). Persson et al. (1996) suggests that the higher chromosomal aneuploidy may depend on a mosaicism or Klinefelter syndrome (46XY/47XXY) in the father. Persson et al. postulated that these subjects could present a mosaicism with an aneuploid cell line confined to the germ tissue not detectable by peripheral karyotype. This hypothesis is in agreement with the increased incidence of chromosome abnormalities in infertile males with a trend that is inversely proportional to the sperm concentration (Kjessler, 1974) (20.3% in non-obstructive azoospermic subjects) (Yoshida et al., 1995). On the other hand, it has been observed that 47XXY germ cells can initiate meiosis producing 24XY spermatozoa (Cozzi et al., 1994). In accordance with this study which used fluorescence in-situ hybridization (FISH), we (Foresta et al., 1996a) have recently detected the presence of 24XY spermatozoa in 30 and 40% of spermatozoa ejaculated from two patients with Klinefelter syndrome (47XXY detected in 100 metaphases) showing a sperm count of <1 X 10⁶/ml. However, in our study, sex chromosome diploidy was also found in the spermatozoa of 15 severe oligozoospermic men with a normal 46XY karyotype (~15% of spermatozoa examined in each subject). In these subjects the pathogenesis of the testiculopathy was due to previous orchitis, testicular trauma or antiublastic therapy. In these cases, it is unlikely that the sperm aneuploidy depends on mosaicism but it is possible that testicular tubular alteration may determine abnormalities in the meiotic process. Interestingly, if this is true, other forms of chromosome diploidy beyond sex chromosomes should be expected, as reported by Pang et al. (1995) and Moosani et al. (1995). On the basis of these considerations it is not possible to divide infertile men into euploid 'low risk' and aneuploid 'high risk' groups by means of peripheral karyotyping, as advocated by Persson et al. (1996). According to our hypothesis of the meiotic derangement in the presence of severe testicular damage, such division can be made only by direct sperm chromosome analysis.

Another important aspect is that among men attending fertility clinics, >30% are diagnosed as severe oligozoospermic or azoospermic with no known aetiological explanation. Deletions of specific sequences on the long arm of the Y chromosome (Yq) are now known to characterize a fraction of idiopathic infertile men (Chandley and Cooke, 1994). Recently in Yq interval 6, two different genes have been identified as candidates for the azoospermia factor (AZF), defined as 'Y-chromosome RNA recognition motif' (YRRM; Ma et al., 1993) and 'deleted in azoosperma' (DAZ; Reijo et al., 1995) respectively. Deletions of both these genes have been detected in somatic cells and in mature spermatozoa of azoospermic and severe oligozoospermic subjects, supporting their role in controlling spermatogenesis (Ma et al., 1993; Reijo et al., 1995, 1996; Najmabadi et al., 1996; Stuppia et al., 1996).

Using polymerase chain reaction (PCR) analysis, we detected interstitial de novo microdeletions in the AZF region in six out of 23 idiopathic severe oligozoospermic (sperm count <5 X 10⁶/ml) men and in five of 16 idiopathic non-obstructive azoospermic men. Our studies demonstrated a high frequency (28.2%) of Yq microdeletions in idiopathic severe spermatogenic impairment and that genes other than YRRM and DAZ regulating spermatogenesis are localized within interval 6.
Furthermore, according to Reijo et al. (1996), no correlation could be identified between the severity of the spermatogenic defect (Sertoli Cell-only syndrome, hypospermatogenesis, partial or complete maturation arrest) as evaluated by testicular fine needle aspiration cytology (Foresta et al., 1995) and the localization and extent of the Y chromosome deletion.

ICSI performed using spermatozoa of these Y-deleted patients will invariably pass this defect on to all male children, whose phenotype of infertility will be identical to the father. Screening for deletion within AZF or at least an informed consent should therefore be obtained in idiopathic infertile males undergoing an ICSI programme.

The primary rationale of medically-assisted reproduction is the opportunity for parents to have a healthy baby. A mandatory condition for achieving this goal is that the genetic characteristics of the gametes should be normal. The important improvements in micromanipulation-assisted fertilization techniques have dramatically changed ideas regarding male infertility and make the involvement of an andrologist important. Before the introduction of ICSI, the andrological approach to male infertility included clinical examination, diagnostic evaluation and, even if often empirically, therapeutic attempts to obtain an improvement of ejaculated spermatozoa and of functional sperm activity. ICSI, by circumventing these aspects, may make the role of the clinical andrologist apparently pointless. Hence, the role of the andrologist is of fundamental importance in evaluating male candidates for ICSI.

References


