

From bench to clinic

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Dr. Ward in his commentary summarized the currently available sperm DNA fragmentation (SDF) tests. He classified the eight SDF assays into four groups based on the different target sites and illustrated the grouping in a diagram to simplify understanding of the readers (1). We would like to supplement, in the following sessions, by (I) further solidifying the current clinical evidence of SDF, and (II) discussing means to move SDF from laboratories to clinics.

It is uncertain if one assay is superior over another since the exact nature of DNA fragmentation detected and their implications are not fully understood. In fact, the different type of SDF detected by each assay may be complementary to each other in different clinical settings which warrant further research. Even in absence of a more complete understanding about the nature of SDF, these tests have shown its promise in the workup of infertile men. There is a clear association between high SDF and decreased pregnancy rates in natural conception and intrauterine insemination (IUI) (2). Emerging evidence also suggests a negative impact of high SDF on pregnancy outcomes in *in vitro* fertilization (IVF) and intracytoplasmic sperm injection (ICSI) cycles (2). Sperm Chromatin Structure Assay (SCSA) is the first assay introduced clinically in the detection of damaged sperm DNA and altered proteins in sperm nuclei via flow cytometry of acridine orange stained sperm (3). The test results are stable over time in healthy individual (4). The correlation between sperm DNA integrity measured by SCSA and natural pregnancy outcome has been demonstrated in the Danish first pregnancy planners study. The study showed that the time-to-pregnancy increased with sperm DNA fragmentation index (DFI) in men who became infertile

with DFI exceeding 30% (5). A meta-analysis involving 616 couples showed that high DFI measured by SCSA was associated with a failure to achieve natural pregnancy with an odds ratio of 7.01 (6). The currently established clinical threshold of DFI is 25% for placing men into a statistical probability of longer time to natural pregnancy, low odds of IUI pregnancy, more miscarriages or no pregnancy (7). Furthermore, TUNEL assay has been shown to have high correlation with SCSA ($r=0.87$) by using the same flow cytometer in the same laboratory (8). The correlation between TUNEL result and assisted reproductive technique (ART) outcomes has also been reported. SDF measured by TUNEL was shown to predict IUI outcomes and no pregnancy was achieved with >12% SDF in the sample (9). The precision and reproducibility of TUNEL assay in a recent study showed that a high correlation in result ($r=0.94$) could be achieved between laboratories by using standardized protocol (10). SDF assays will firmly establish its role in the management of infertile couples with further clinical data.

To enhance the utilization of SDF as a new test of sperm function requires that, in addition to the available laboratory and clinical data, effort should be spent in popularizing the test among clinicians. First of all, deficiency in the current practice and a need for new advancement must be recognized. The limitation of semen analysis as the cornerstone of the male fertility assessment tool should be noticed as there is a significant overlap in semen parameters between fertile and infertile men (11). In fact, semen analysis failed to detect any abnormality in up to 30% of infertile men based on the current workup algorithm (12). Despite all the

advances in ART, the use of ICSI as the treatment of male infertility resulted in a live birth of no more than 30% (13). Therefore, the potential of SDF test in providing additional information for male infertility patients should be conveyed to the clinicians. The practice recommendations by Agarwal *et al.* (14) attempted to bridge the gap between laboratory and clinic by suggesting wider clinical application of SDF tests in specific clinical scenarios based on the best current evidence. We believe it is an important first step to move the test from bench to clinic and benefit the infertile couples. Furthermore, the cost of SDF testing will probably be justified in face of the high cost of ART. A more targeted selection of ART technique based on SDF testing result will be a more economical choice than the trial-and-error approach of repeated ART cycles. In one study, Esteves *et al.* evaluated the use of testicular in preference over ejaculated sperm for ICSI in men with high SDF in semen. The authors showed that the number needed to treat by testicular compared to ejaculated sperm to obtain an additional live birth per fresh transfer cycles was 4.9 (95% CI: 2.8–16.8) (15). Added to this, the risk of ART cycles and multiple gestations on both female partner and fetus can be reduced with appropriate selection of ART which will offer the highest success rate. We argue that the added information provided by the non-invasive SDF tests should lead to a better counselling of infertile couples in the busy clinics. Familiarity of the SDF tests amongst the clinicians is the missing link. Confidence in incorporating the test result in treatment decisions will certainly increase with time and more extensive use of the test.

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Footnote

Conflicts of Interest: The authors have no conflicts of interest to declare.

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