Editorial

# Expanding our understanding of clinical laboratory testing in male infertility patients

**Sandro C. Esteves[1], Ahmad Majzoub[2], Ashok Agarwal[3]**

[1]ANDROFERT, Andrology and Human Reproduction Clinic, Referral Center for Male Reproduction, Campinas, SP, Brazil; [2]Department of Urology, Hamad Medical Corporation, Doha, Qatar; [3]American Center for Reproductive Medicine, Cleveland Clinic, Cleveland, OH, USA

*Correspondence to:* Sandro C. Esteves. Director, ANDROFERT – Andrology & Human Reproduction Clinic, and Professor, Division of Urology, UNICAMP, Av. Dr. Heitor Penteado 1464, Campinas, SP, 13075-460, Brazil. Email: s.esteves@androfert.com.br.

*Response to:* Marmar JL. Is testing of sperm DNA fragmentation (SDF) ready for the basic work-up of male infertility? Transl Androl Urol 2017;6:S437-9.

Submitted Apr 15, 2017. Accepted for publication Apr 17, 2017.
doi: 10.21037/tau.2017.05.02
**View this article at:** http://dx.doi.org/10.21037/tau.2017.05.02

Dr. Marmar in his commentary contextualizing the article by Agarwal *et al.* (1) has provided constructive suggestions regarding the clinical use of SDF testing (2). First, Dr. Marmar illustrates the importance of the standardization process by calculating precision, accuracy, and coefficient of variation associated with the testing, and provides references for readers interested in deepening their understanding about clinical laboratory testing on male patients. Second, the author revisited the importance of varicocelectomy in the modern ICSI era given the newest Cochrane systematic review and meta-analysis conclusion that "treatment of infertile men with a clinically manifest varicocele and poor semen quality may be of benefit" (3). Third, Dr. Marmar provoke readers with two intriguing questions: (I) should ROS testing be considered for the infertility work-up, as well? and (II) will additional testing add significant costs to the patients? In our response, we aim to provide insights on his remarks.

Indeed, both accuracy, the degree to which the measurement reflects the real value, and precision, the reproducibility of the results, are critical for clinicians relying on the results provided by laboratory testing to guide counseling, further workup, and management of infertile couples (4,5). Notwithstanding, even semen analysis suffers from standardization issues despite the efforts of the World Health Organization to elaborate and update guidelines for the laboratory examination of human semen (6). Not surprising, the situation with SDF testing is not much different despite the genuine efforts to standardize the assays (7-11). Owing to the complex nature of semen, basic and advanced semen analysis, including SDF testing, should be carried out in laboratories equipped with proper instrumentation and skilled technicians. Moreover, internal and external quality control programs, including proficiency testing, should be an integral element of the services provided. Lastly, validation of test systems, quality assurance during all testing processes, and proper communication with clinicians and patients as regards the pre- and post-analytical assay elements are of utmost importance (5,12).

As far as the importance of varicocelectomy in the ICSI era is concerned, Dr. Marmar himself contributed remarkably to the refinements of varicocele surgery. In 1985, the first microsurgical varicocelectomy with an operating microscope and microsurgical instruments was reported by the author (13). Nowadays, the "Marmar technique" for varicocele repair has become the method of choice for the majority of urologists subspecialized in male infertility (14,15).

Lastly, in this section, we attempt to answer Dr. Marmar's questions mentioned above. Indeed, an increasing body of evidence indicates that oxidative stress represents a central element [reviewed by Cho *et al.* (15)]. In varicocele patients, reactive oxygen species (ROS) are released by the principal cells in the epididymis, endothelial cells of the dilated pampiniform plexus, and testicular cells (germ cells, Leydig cells, macrophages, and peritubular cells).

Excessive ROS has been associated with SDF; the latter is postulated to be one of the mediators of poor sperm quality and resulting infertility in affected men (15). However, not all men with varicocele exhibit high SDF results. In one study involving 55 patients with clinical varicocele and infertility, increased DNA damage (defined as the mean of the control group plus 2SD) was noted in 49% patients with normal semen profile and 58% of patients with abnormal semen parameters (16). Added to this, intrinsic mechanisms may counteract the actions of ROS that might explain why many men retain their fertility in the presence of a varicocele. Measuring ROS in infertile men, including those with varicocele, may offer additional information, particularly for those with normal SDF results (17). In this scenario, protective antioxidant mechanisms may still be active providing a dynamic equilibrium. However, a deviation from homeostasis might lead to impairment in sperm function and consequent infertility. Also, ROS testing could be useful not only to monitor the results of interventions and to guide couples in the decision of pursuing ART but also suggest options for prevention (17). Although SDF testing, and also ROS testing, add costs that are not currently covered by many insurance companies, their significance should be weighed as a function of the likely better reproductive outcomes on an individual basis.

## Acknowledgements

None.

## Footnote

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

## References

1. Marmar JL. Is testing of sperm DNA fragmentation (SDF) ready for the basic work-up of male infertility? Transl Androl Urol 2017;6:S437-9.
2. Agarwal A, Majzoub A, Esteves SC, et al. Clinical utility of sperm DNA fragmentation testing: practice recommendations based on clinical scenarios. Transl Androl Urol 2016;5:935-50.
3. Kroese AC, de Lange NM, Collins J, et al. Surgery or embolization for varicoceles in subfertile men. Cochrane Database Syst Rev 2012;10:CD000479.
4. Esteves SC, Hamada A, Kondray V, et al. What every gynecologist should know about male infertility: an update. Arch Gynecol Obstet 2012;286:217-29.
5. Esteves SC. Clinical relevance of routine semen analysis and controversies surrounding the 2010 World Health Organization criteria for semen examination. Int Braz J Urol 2014;40:443-53.
6. Esteves SC, Zini A, Aziz N, et al. Critical appraisal of World Health Organization's new reference values for human semen characteristics and effect on diagnosis and treatment of subfertile men. Urology 2012;79:16-22.
7. Esteves SC, Sharma RK, Gosálvez J, et al. A translational medicine appraisal of specialized andrology testing in unexplained male infertility. Int Urol Nephrol 2014;46:1037-52.
8. Feijó CM, Esteves SC. Diagnostic accuracy of sperm chromatin dispersion test to evaluate sperm deoxyribonucleic acid damage in men with unexplained infertility. Fertil Steril 2014;101:58-63.e3.
9. Evenson DP. The Sperm Chromatin Structure Assay (SCSA(®)) and other sperm DNA fragmentation tests for evaluation of sperm nuclear DNA integrity as related to fertility. Anim Reprod Sci 2016;169:56-75.
10. Sharma R, Ahmad G, Esteves SC, et al. Terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL) assay using bench top flow cytometer for evaluation of sperm DNA fragmentation in fertility laboratories: protocol, reference values, and quality control. J Assist Reprod Genet 2016;33:291-300.
11. Ribeiro S, Sharma R, Gupta S, et al. Inter- and intra-laboratory standardization of TUNEL assay for assessment of sperm DNA fragmentation. Andrology 2017;5:477-85.
12. Esteves SC, Agarwal A. Ensuring that reproductive laboratories provide high-quality services. In: Bento FC, Esteves SC, Agarwal A. editors. Quality Management in ART Clinics: A Practical Guide. 1st ed. New York: Springer US, 2013: 129-46.
13. Marmar JL. The evolution and refinements of varicocele surgery. Asian J Androl 2016;18:171-8.
14. Esteves SC, Agarwal A. Afterword to varicocele and male infertility: current concepts and future perspectives. Asian J Androl 2016;18:319-22.
15. Cho CL, Esteves SC, Agarwal A. Novel insights into the pathophysiology of varicocele and its association with reactive oxygen species and sperm DNA fragmentation. Asian J Androl 2016;18:186-93.
16. Smit M, Romijn JC, Wildhagen MF, et al. Decreased

 

sperm DNA fragmentation after surgical varicocelectomy is associated with increased pregnancy rate. J Urol 2013;189:S146-50.

17. Majzoub A, Esteves SC, Gosálvez J, et al. Specialized sperm function tests in varicocele and the future of andrology laboratory. Asian J Androl 2016;18:205-12.

**Cite this article as:** Esteves SC, Majzoub A, Agarwal A. Expanding our understanding of clinical laboratory testing in male infertility patients. Transl Androl Urol 2017;6(Suppl 4):S440-S442. doi:10.21037/tau.2017.05.02

 