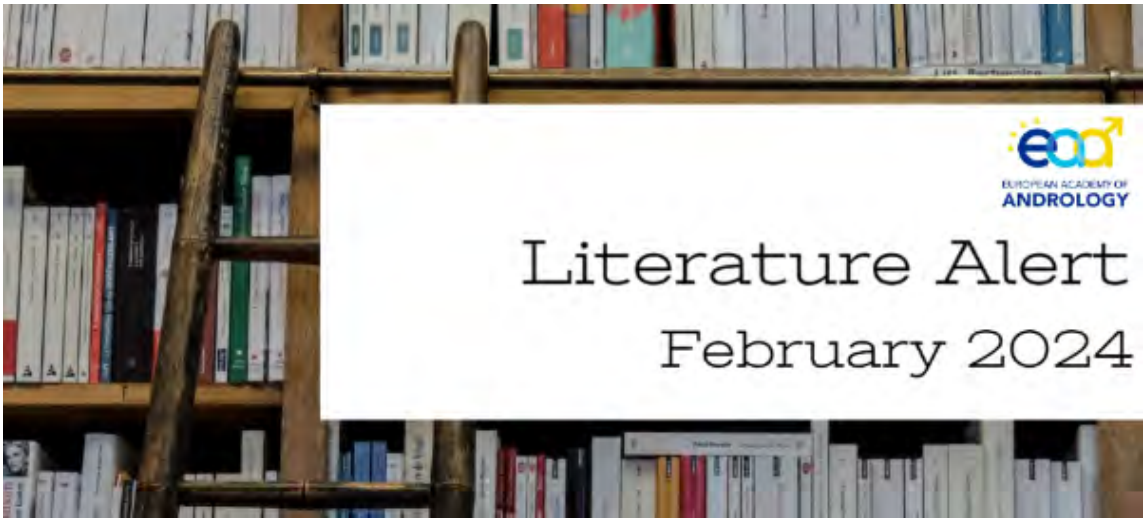


Dear EAA Members,

Exciting basic and clinical science in this issue: a new mouse model for reversible male contraception, transcriptome of human fetal gonads and reproductive tract, a missing link between piRNA and DNA methylation in germ cells, neurodevelopmental ZFX variant syndrome, sildenafil and Alzheimer, first autologous grafting of adult testis tissue, biopsy indications in testicular microlithiasis, anogenital distance and cryptorchidism, glucocorticoid exposure and sperm miRNA, several studies of endocrine disrupters/lifestyle and sperm quality, slice organ culture, machine learning for CT of testicular masses, and more. Check out also the latest issue of *Andrology* packed with interesting articles. Enjoy!

Clinical andrology and epidemiology

The third issue of *Andrology* in 2024 has already been released. It contains several very interesting articles (*a few previously highlighted in the EAA alerts*), including: the EAA/EMQN best practice guidelines for molecular diagnosis of Y-chromosomal microdeletions, a review on non-surgical treatment of Peyronie's disease, vit. D effect on hormone profiles of infertile men, a meta-analysis on anti-oxidants in OAT, the TEX genes in male infertility, the AZFc region duplications, several studies of erectile dysfunction and premature ejaculation, penile prosthesis, polarisation of testicular macrophages, and more.
Volume 12, Issue 3

“Viagra prevents Alzheimer!” – according to the popular press reacting to this interesting article. What this UK electronic-records-based study found was the association of multiple prescriptions of phosphodiesterase type 5 inhibitors (PDE5Is) in men with erectile dysfunction and a lower risk of Alzheimer disease.

Adesuyan M, Jani YH, Alsugeir D, Howard R, Ju C, Wei L, Brauer R. Phosphodiesterase Type 5 Inhibitors in Men With Erectile Dysfunction and the Risk of Alzheimer Disease: A Cohort Study. ***Neurology***. 2024 Feb 27; 102(4):e209131.
https://doi.org/10.1212/WNL.0000000000209131

The first autologous grafting of frozen-thawed testis tissue (2 years after mTESE) was performed in an adult man with NOA. Despite the graft survival and the presence of spermatocytes, no mature spermatozoa were detected; most likely due to the pre-existing spermatogenic failure.

Jensen C, Mamsen L, Wang D, Fode M, Giwercman A, Jørgensen N, Ohl DA, Fedder J, Hoffmann ER, Andersen CY, Sønksen J. Results from the first autologous grafting of adult human testis tissue: a case report. ***Hum Reprod*** 2024; Feb, 39:303–309.
https://doi.org/10.1093/humrep/dead243
Commentary by R. Mitchell & J. Ives: ***Hum Reprod***. Feb. 2024,
https://doi.org/10.1093/humrep/dead258

Men with testicular microlithiasis are at an increased risk of germ cell neoplasia in situ (GCNIS), which this study found in 7.8% [95% CI: 4.3, 13.2] of men in a series of infertile patients. The authors suggest that only the presence of testicular atrophy should be an indication for a biopsy in men with microlithiasis.

Frandsen RH, Durukan E, von Rohden E, Jensen CFS, Thamsborg AKM, Azawi N, Fode M. Testicular biopsies in men with testicular microlithiasis and additional risk factors for cancer: A case series. ***Andrology***. 2024 Feb 20.
https://doi.org/10.1111/andr.13610

This study of boys with cryptorchidism revealed that anogenital distance (AGD) was reduced in the whole cohort, with the shortest AGDs in boys with vanished testis. Interestingly, boys with bilateral cryptorchidism had more often hypogonadotropic hypogonadism but had longer AGDs than those with unilateral cryptorchidism.

Cortes D, Fischer MB, Hildorf AE, Clasen-Linde E, Hildorf S, Juul A, Main KM, Thorup J. Anogenital distance in a cohort of 169 infant boys with uni- or bilateral cryptorchidism including 18 boys with vanishing testes. ***Hum Reprod.*** 2024 Feb 19:deae025.
https://doi.org/10.1093/humrep/deae025

The reproductive outcomes of patients after unilateral vs. bilateral orchidopexy were compared. The latter had worse sperm parameters and a higher demand for ICSI. Interestingly, both groups exhibited similar rates of fertilization, clinical pregnancy and live births.

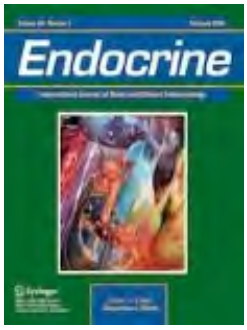
Fan L, Shi L, Liu S, Zhang Z, Shi J. Bilateral versus unilateral orchidopexy: IVF/ICSI-ET outcomes. ***Front Endocrinol.*** 2024 Feb 1;15:1294884.
https://doi.org/10.3389/fendo.2024.1294884

Repeated measurements of biochemical and anthropometric variables and serum testosterone (T), DHT, estradiol and estrone were done in healthy men (>40 y.) using validated LCMS. The study provided estimates of variability in serum T and androgen biomarkers for future studies of androgen action in male ageing.

Handelsman DJ, Sartorius G, Desai R, Idan A, Turner L, Savkovic S, Ly LP, Forbes E, Allan CA, McLachlan R, Conway AJ. Sex steroids and androgen biomarkers in the healthy man study: within-person variability and impact of fasting. ***Eur J Endocrinol*.** 2024; 190(1):54-61.
https://doi.org/10.1093/ejendo/lvad178

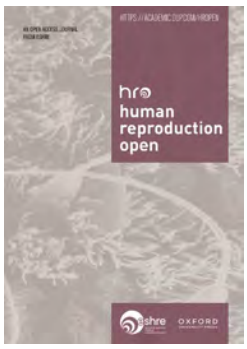
This study confirmed that idiopathic infertile men have widely variable serum testosterone (T) levels, and around a quarter of them present some sort of functional hypogonadism. The authors call for better classification of idiopathic male infertility, taking T levels into account.

Spaggiari G, Costantino F, Dalla Valentina L, Romeo M, Furini C, Roli L, De Santis MC, Canu G, Trenti T, Granata ARM, Simoni M, Santi D. Are they functional hypogonadal men? Testosterone serum levels unravel male idiopathic infertility subgroups. ***Endocrine***. 2024 Feb 19.
https://doi.org/10.1007/s12020-024-03717-3



Consumption of ultra-processed food was inversely associated with total sperm count, sperm concentration and total motility in men of reproductive age. If these results are replicated, diet programs could be included in the management of poor semen quality in men.

Valle-Hita C, Salas-Huetos A, Fernández de la Puente M, Martínez MÁ, Canudas S, Palau-Galindo A, Mestres C, Manzanares JM, Murphy MM, Marquès M, Salas-Salvadó J, Babio N. Ultra-processed food consumption and semen quality parameters in the Led-Fertyl study. ***Hum Reprod Open***. 2024 Jan 17;2024(1):hoae001.
https://doi.org/10.1093/hropen/hoae001



This study assessed the sperm epigenome in relation to p,p'-DDE serum levels in two different populations. Differences in DNA-me and H3K4me3 enrichment were identified at transposable elements and regulatory regions involved in fertility, disease, development, and neurofunction, some predicted to persist in the preimplantation embryo.

Lismer A, Shao X, Dumargne MC, Lafleur C, Lambrot R, Chan D, Toft G, Bonde JP, MacFarlane AJ, Bornman R, Aneck-Hahn N, Patrick S, Bailey JM, de Jager C, Dumeaux V, Trasler JM, Kimmins S. The Association between Long-Term DDT or DDE Exposures and an Altered Sperm Epigenome-a Cross-Sectional Study of Greenlandic Inuit and South African VhaVenda Men. ***Environ Health Perspect*.** 2024 Jan;132(1):17008.
https://doi.org/10.1289/ehp12013

Two studies demonstrated a widespread contamination of human semen with persistent endocrine disruptors polychlorinated biphenyls (PCBs), and found some associations between the exposure and human sperm quality parameters:

Ješeta M, Kalina J, Franzova K, Fialkova S, Hosek J, Mekinova L, Crha I, Kempisty B, Ventruba P, Navratilova J. Cross sectional study on exposure to BPA and its analogues and semen parameters in Czech men. ***Environ Pollut***. 345, 2024, 123445, ISSN 0269-7491
https://doi.org/10.1016/j.envpol.2024.123445

Donato F, Rota M, Ceretti E, Viola GCV, Marullo M, Zani D, Amoresano A, Fontanarosa C, Spinelli M, Lorenzetti S, Montano L; FASt Study Group. Polychlorinated Biphenyls and Semen Quality in Healthy Young Men Living in a Contaminated Area. ***Toxics***. 2023 Dec 20;12(1):6.
https://doi.org/10.3390/toxics12010006

Androgenetics

ZFX (Xp22.11) encodes a transcription factor, germline mutations of which can cause X-linked intellectual disability, more prevalent in men than women. This study presented clinical and molecular characterization of 18 individuals with germline ZFX variants, who often have similar behavioural and facial features.

Shepherdson JL, Hutchison K, Don DW, et al et Shinawi M. Variants in ZFX are associated with an X-linked neurodevelopmental disorder with recurrent facial gestalt. ***Am J Hum Genet (AJHG)***. 2024 Feb 5:S0002-9297(24)00007-7.
https://doi.org/10.1016/j.ajhg.2024.01.007

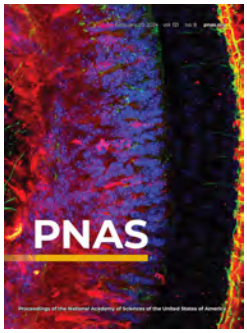
This case-control GWAS study of men with severe spermatogenic failure found novel association signals displaying interaction with age-groups. The authors found links between these loci and diverse non-genetic factors and proposed that variable exposome could influence genetic predisposition to complex diseases.

Cerván-Martín M, González-Muñoz S, Guzmán-Jiménez A, Higueras-Serrano I, Castilla JA, Garrido N, Luján S, Bassas L, Seixas S, Gonçalves J, Lopes AM, Larriba S, Palomino-Morales RJ, Bossini-Castillo L, Carmona FD. Changes in environmental exposures over decades may influence the genetic architecture of severe spermatogenic failure. ***Hum Reprod.*** 2024 Feb 1:deae007.
https://doi.org/10.1093/humrep/deae007

Novel bi-allelic deleterious variants of ADGB (androglobin) were identified in 3/105 Chinese men with OAT. The men had multiple sperm malformations but were successfully treated with ICSI.

Gao Y, Liu L, et al et He X. Whole-exome sequencing identifies ADGB as a novel causative gene for male infertility in humans: from motility to fertilization. ***Andrology***. 2024 Feb 22.
https://doi.org/10.1111/andr.13605

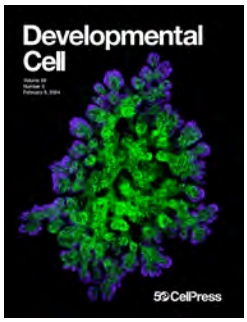
Translational and basic andrology



This excellent study reports the development of a novel contraceptive approach. The authors showed that inhibiting the action of the silencing mediator of retinoid and thyroid hormone receptors (SMRT) complex with chronic, low-dose oral administration of a histone deacetylase inhibitor reversibly blocks spermatogenesis and fertility without affecting libido. This demonstration validates pharmacologic targeting of the SMRT repressor complex for non-hormonal male contraception.

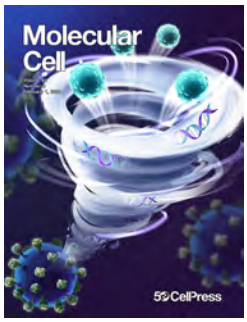
Hong SH, Castro G, Wang D, Nofsinger R, Kane M, Folias A, Atkins AR, Yu RT, Napoli JL, Sassone-Corsi P, de Rooij DG, Liddle C, Downes M, Evans RM. Targeting nuclear receptor corepressors for reversible male contraception. Proc Natl Acad Sci USA. 2024 Feb, https://doi.org/10.1073/pnas.2320129121

Using single-cell transcriptomics and validation by immunofluorescence, this study examined the development of fetal human gonads and the adjacent reproductive tract of both sexes. The authors found pronounced sex differences already in the first trimester, identified some key genes in the early reproductive tract associated with urogenital diseases, and stromal and epithelial signatures diverging in gonads and reproductive tract.



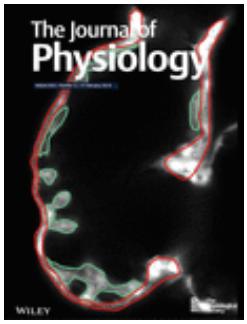
Taelman J, Czukiewska SM, Moustakas I, Chang YW, Hillenius S, van der Helm T, van der Meeren LE, Mei H, Fan X, Chuva de Sousa Lopes SM. Characterization of the human fetal gonad and reproductive tract by single-cell transcriptomics. ***Dev Cell.*** 2024 Jan 29:S1534-5807(24)00006-6. https://doi.org/10.1016/j.devcel.2024.01.006

PIWI proteins and PIWI-interacting RNAs (piRNAs) guide silencing of transposons by DNA methyltransferase DNMT3C to protect male germline. This study identified loss-of-function variants in human SPOCD1 that cause defective transposon silencing and male infertility. The team also discovered that an uncharacterized protein C19ORF84 is essential for orchestrating piRNA-directed DNA methylation.



Zoch A, Konieczny G, Auchynnikava T, Stallmeyer B, Rotte N, Heep M, Berrens RV, Schito M, Kabayama Y, Schöpp T, Kliesch S, Houston B, Nagirnaja L, O'Bryan MK, Aston KI, Conrad DF, Rappsilber J, Allshire RC, Cook AG, Tüttelmann F, O'Carroll D. C19ORF84 connects piRNA and DNA methylation machineries to defend the mammalian germ line. ***Mol Cell.*** 2024 Feb 7.
https://doi.org/10.1016/j.molcel.2024.01.014

Studies in an animal model found that maternal exposure to betamethasone altered miRNA levels in epididymal sperm (but not in germ cells in the testes) of the offspring that were apparent for at least two generations.



Hamada H, Casciaro C, Moisiadis VG, Constantinof A, Kostaki A, Matthews SG. Prenatal maternal glucocorticoid exposure modifies sperm miRNA profiles across multiple generations in the guinea-pig. ***J Physiol.*** 2024 Jan 29. https://doi.org/10.1113/jp284942

This study identified transposable elements that likely operate as regulatory elements for sex-specific genes during mouse embryonic gonadal development.

frontiers in Cell and Developmental Biology

Stévant I, Gonen N, Poulat F. Transposable elements acquire time- and sex-specific transcriptional and epigenetic signatures along mouse fetal gonad development. ***Front Cell Dev Biol***. 2024 Jan 12;11:1327410.
https://doi.org/10.3389/fcell.2023.1327410

Methodology

Machine-learning classifiers and a logistics regression model were used to differentiate testicular masses using contrast-enhanced computed tomography (CT) and CT texture analysis in a series of patients with histopathologically confirmed testicular tumours and benign masses.

frontiers in Oncology

Hu C, Qiao X, Xu Z, Zhang Z, Zhang X. Machine learning-based CT texture analysis in the differentiation of testicular masses. Front Oncol. 2024 Jan 16;13:1284040.
https://doi.org/10.3389/fonc.2023.1284040

Slice culture is a useful and easy-to implement technique for live imaging of reproductive events at cellular resolution. This article provides a step-by-step protocol to facilitate setting up this technique.

Amato CM, Yao HH. New uses for an old technique: live imaging on the slice organ culture to study reproductive processes. ***Biol Reprod.*** 2024 Feb 5:ioae023.
https://doi.org/10.1093/biolre/ioae023