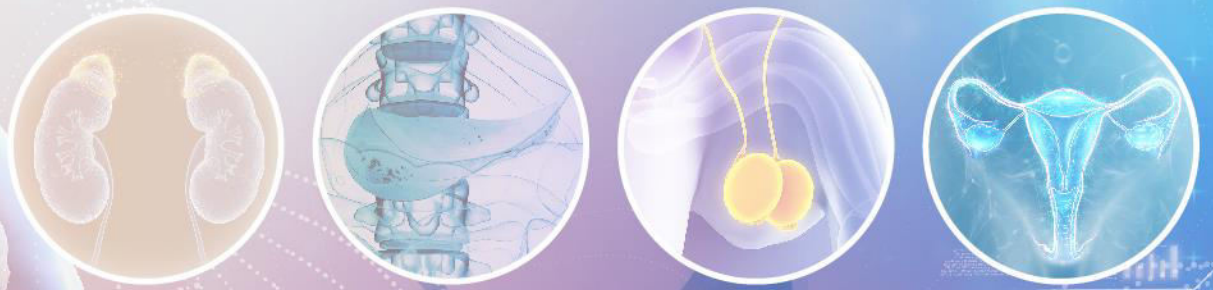
First Edition 2024

PRACTICAL GUIDE TO
ENDOCRINE DYNAMIC TESTS

KKM Endocrine Subspecialty Service



MEMS
Established 1981
Malaysia Endocrine & Metabolic Society

Ministry of Health Malaysia

Fertility Induction in hypogonadotropic hypogonadism males

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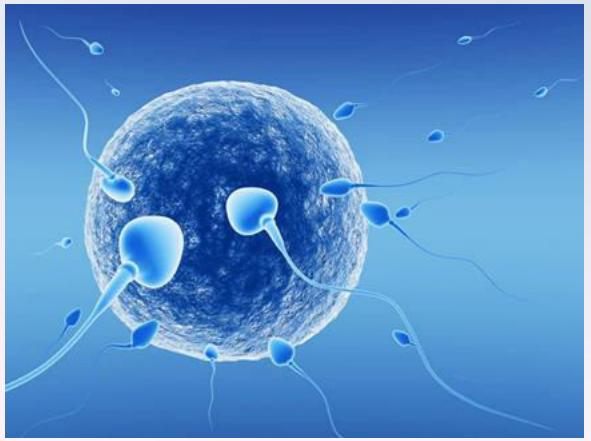
Alor Setar, Kedah

Overview

- Introduction and definition
- Embryology and physiology
- Causes of male subfertility
- Approach to patients
  - History/ PE
  - Investigations
- Case illustration
- Induction of spermatogenesis
- Summary



Abbreviations

- HH –hypogonadotropic hypogonadism
- T - testosterone
- TV – testicular volume
- Gn - gonadotropin
- SFA – seminal fluid analysis
- SA – semen analysis
- ART – assisted reproductive technology
- SSR – surgical sperm retrieval
- ICSI – intracytoplasmic sperm injection
- IVF – in vitro fertilisation
- HMG – human menopausal gonadotropin
- hCG – human chorionic gonadotropin (LH)
- hMG – human menopausal gonadotropin (LH + FSH)
- COS – controlled ovarian stimulation

Introduction

- 15% of couples have difficulty conceiving
- Infertility is classically defined as the inability for a couple to conceive after 12 months of regular coitus without the use of contraception
- Subfertility is a term used to describe reduced fertility that might require therapy for successful conception
- 30% - male reproductive dysfunction
- 20% - both partners have reproductive abnormality.
- Male reproductive dysfunction contributes to about half of all cases of subfertile couples.

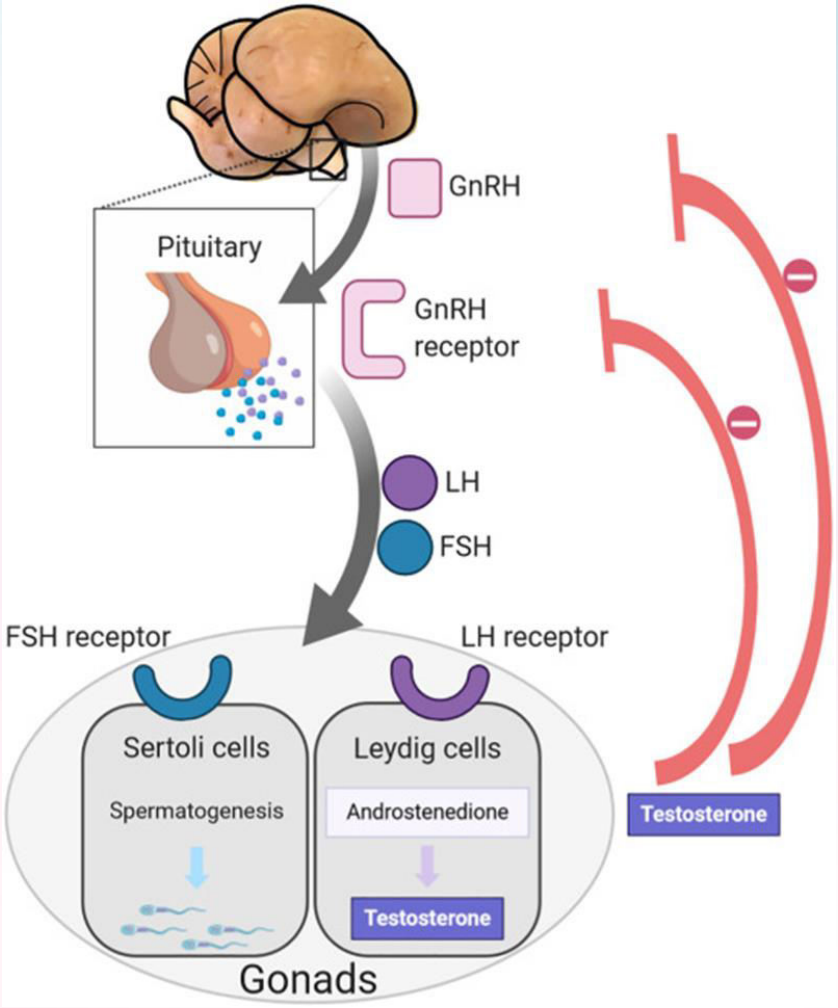
Embryology, physiology and causes of hypogonadism

Causes of male subfertility

- Sexual disorders
  - ED
- Primary testicular defect in sperm production
  - Chemotherapy/Orchidectomy/ Klinefelter's/ Pelvic irradiation/Cryptochidism/ Infection/Autoimmune/ Drugs /Tobacco/ Alcohol
- Endocrinopathies that affect spermatogenesis
  - **Hypothalamopituitary disease (hypogonadotropic hypogonadism)**
  - Thyroid dysfunction (RAI/ ablation)
  - Obesity / Cushing's syndrome/ CAH/ TART/ DM
- Defects in sperm transportation
  - Obstruction
  - Ejaculatory dysfunction

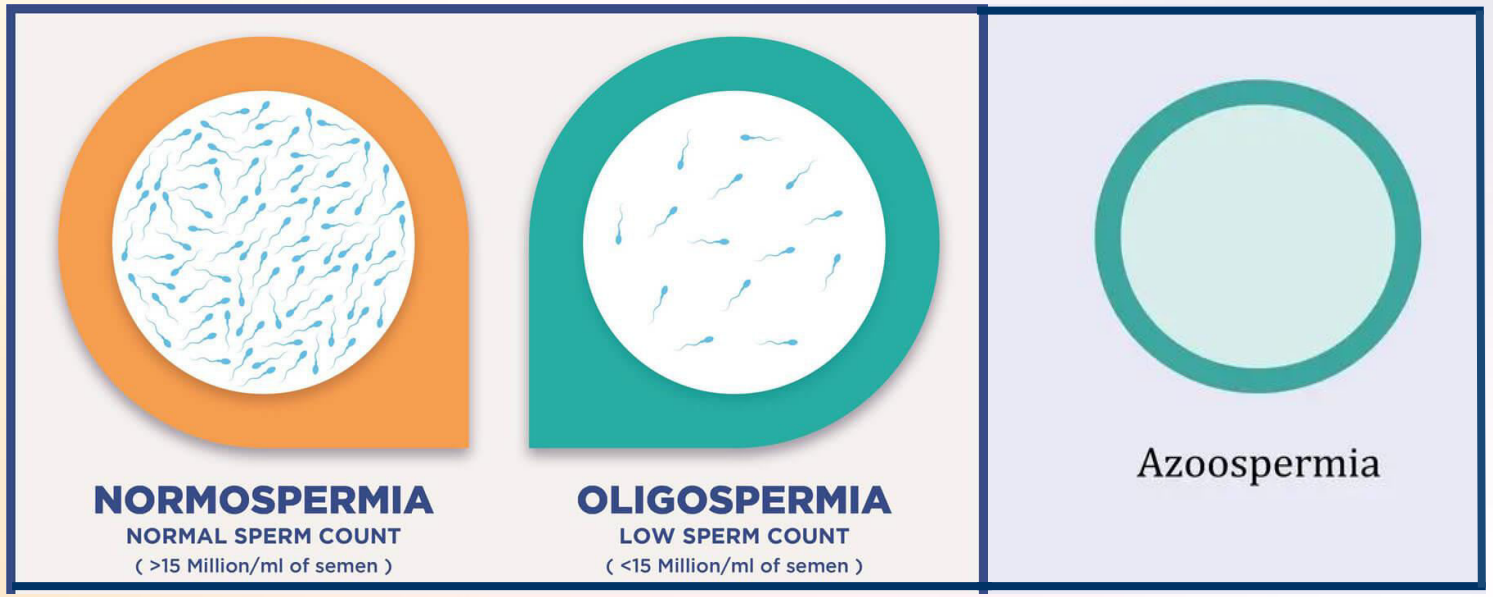
Definition of male hypogonadism

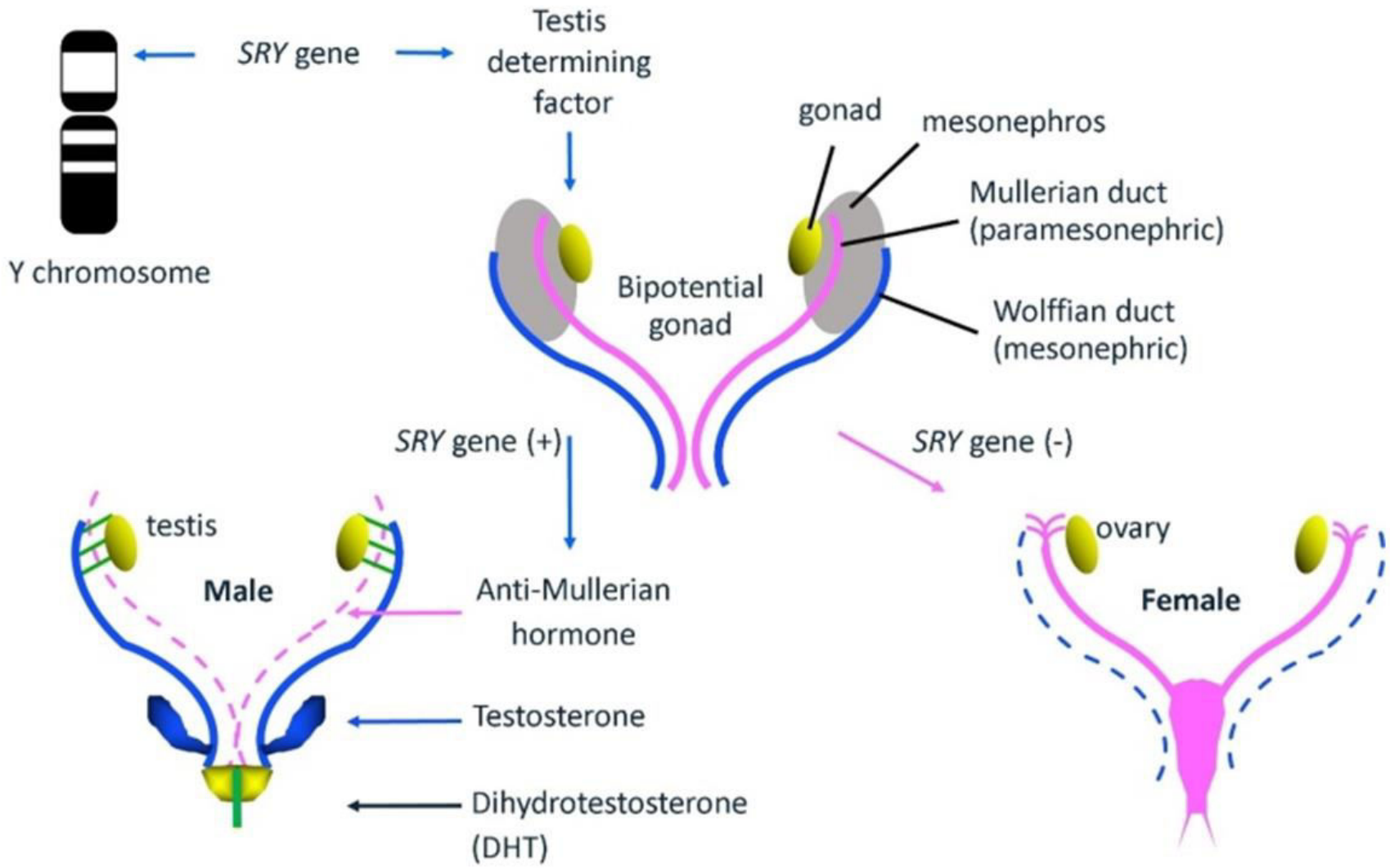
- A clinical syndrome
- failure of the testis to produce physiological
  - levels of testosterone
  - normal number of spermatozoa
- due to disruption of one or more levels of the hypothalamic-pituitary-testicular axis.

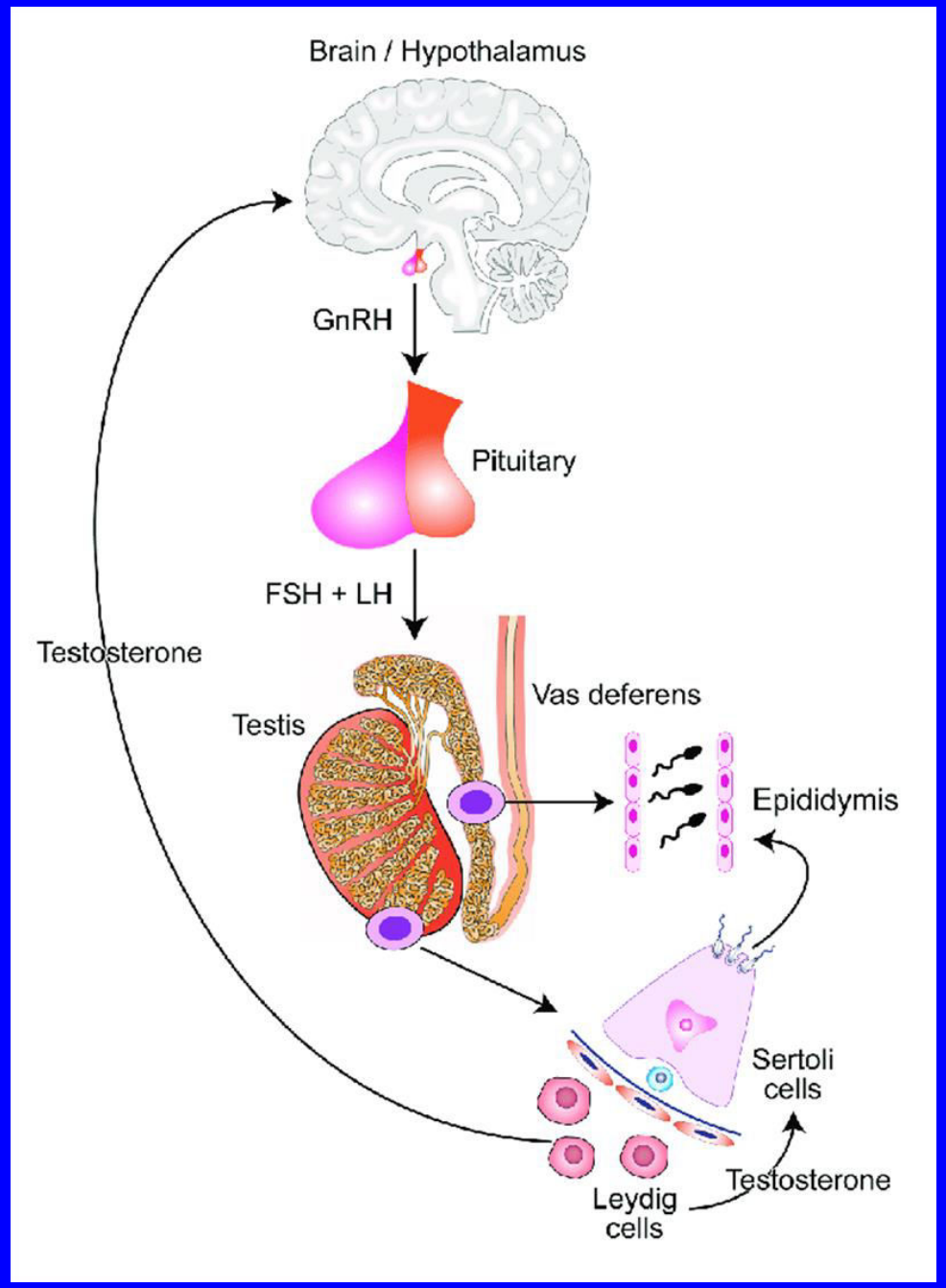


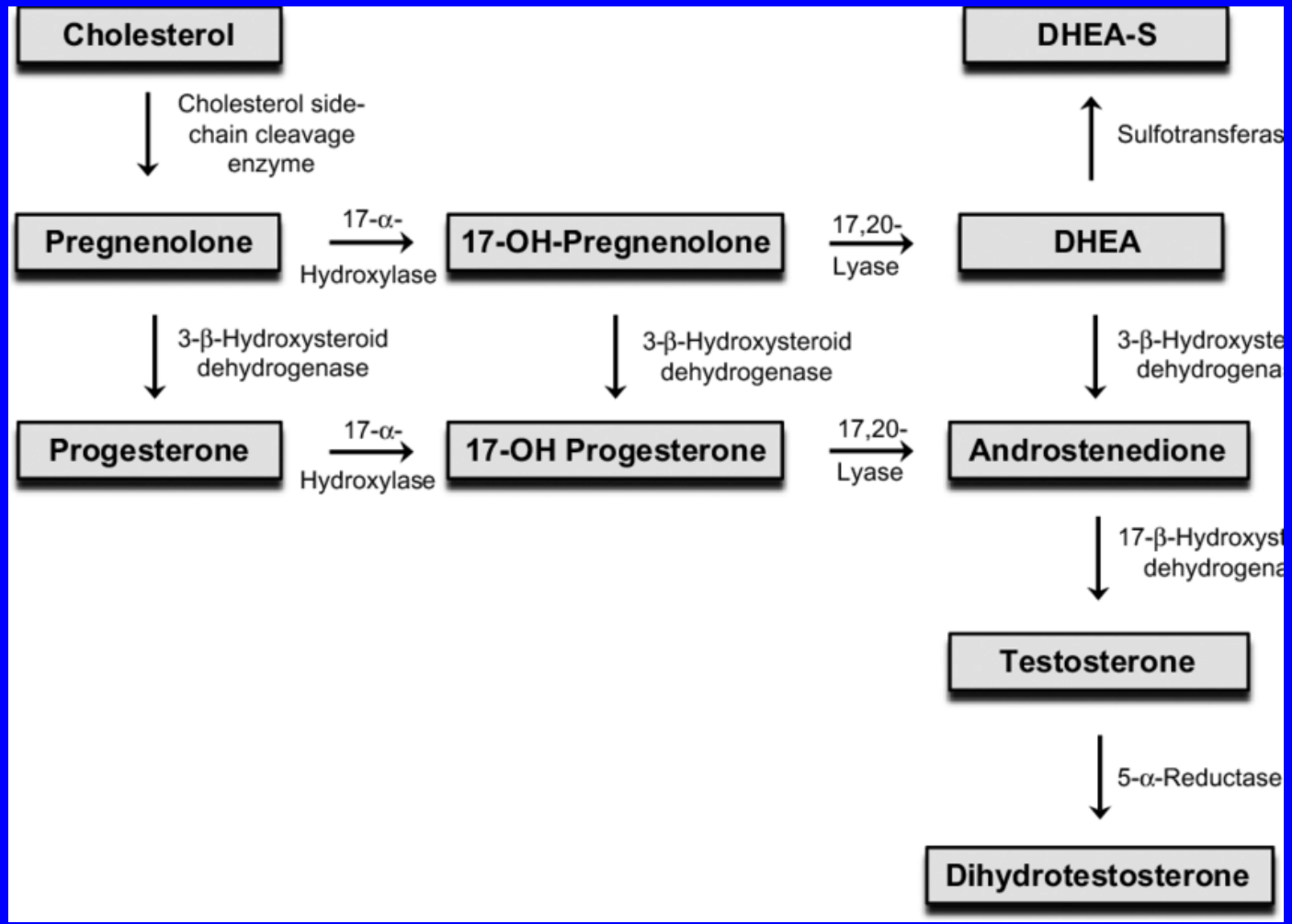
Definition

- Oligospermia : < 15million/ mL ejaculate
- Azoospermia : absence of sperm




SRY gene
Testis determining factor
Y chromosome
gonad
mesonephros
Mullerian duct (paramesonephric)
Wolffian duct (mesonephric)
Bipotential gonad
SRY gene (+)
SRY gene (-)
testis
Male
Anti-Mullerian hormone
Testosterone
Dihydrotestosterone (DHT)
ovary
Female


Brain / Hypothalamus
GnRH
Pituitary
FSH + LH
Testosterone
Testis
Vas deferens
Epididymis
Sertoli cells
Leydig cells
Testosterone


Cholesterol
Cholesterol side-chain cleavage enzyme
Pregnenolone
17-α-Hydroxylase
17-OH-Pregnenolone
17,20-Lyase
DHEA
DHEA-S
Sulfotransferas
3-β-Hydroxysteroid dehydrogenase
3-β-Hydroxysteroid dehydrogenase
3-β-Hydroxyste dehydrogena
Progesterone
17-α-Hydroxylase
17-OH Progesterone
17,20-Lyase
Androstenedione
17-β-Hydroxyst dehydrogena
Testosterone
5-α-Reductase
Dihydrotestosterone

9-10 weeks to complete stages of spermatogenesis

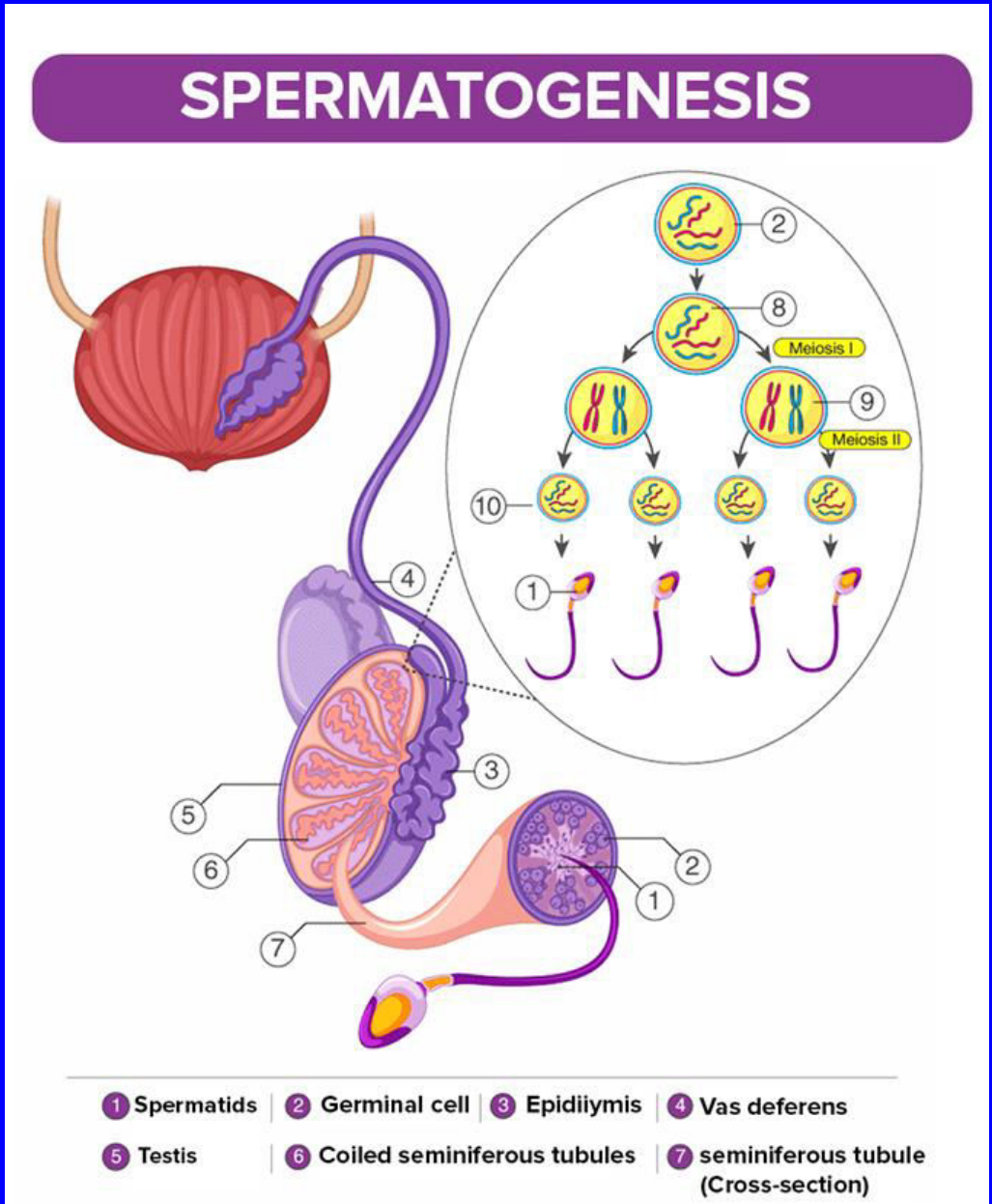
Spermatogonia → spermatocyte → spermatid → spermatozoa

FSH → Sertoli cells → sperm

LH → Leydig cells → T

Seminiferous tubules - 90% of TV → size of the testes critical indicator of fertility potential.

Both FSH and T are necessary for both quantitatively and qualitatively normal spermatogenesis



Approach to patient

Approach depends on…

- Which clinic we sit in
  - General medical clinic
  - Endocrinology clinic
  - O&G clinic

- Timing of insult
  - Pre pubertal
  - Attained puberty but arrested before completion
  - Post pubertal-completion

- Determines what investigations done initially and how to further manage

Assessment

History

- What is actual problem
- Any child from previous union
- Delayed puberty
- Hypoandrogenism
  - low libido
  - morning erections
  - shaving frequency
- Medical conditions
  - DMT2
  - Hormone-related
  - Systemic illness
  - Mumps orchitis
- Trauma/ surgery
- Chemotherapy/ pelvic irradion
- Drugs

Physical examination

- Stature
  - Height
  - Body habitus
- Dysmorphism
- Cushingoid/acromegaloid/dysthyroid appearance
- Sexual characteristic
  - Voice
  - Muscle bulk
  - Body hair distribution
  - Phallus / adrenarche
- Testicular
  - present / absent
  - consistency / tortuosity
  - size – Prader orchidometer
- Neurological
  - Reflexes
  - Peripheral neuropathy


measured in mL
Childhood
Adulthood
Early Puberty
Late Puberty
1
2
3
4
5
6
8
10
12
15
20
25

Investigations

- LH (ref range 1.6-8.0 IU/ L)
- FSH (ref range 1.3 – 8.4 IU/L)
- Serum T (ref range 9.3-37.1 nmol/L) – follow own lab
- TFT
- Prolactin
- RP/ LFT
- SFA
- +/- scrotal ultrasound
- Other investigation accordingly- MRI/ bloods

K Sikaris et al. J Clin Endocrinol Metab 9:5928–5936, 2005

SFA - how collected

- At least 2 samples
- After 2-7 days of abstinence from ejaculation
  - Cooper TG et al. Hum Reprod Update.2010; 16:231–45.
  - Sample reaches lab within 30 minutes

Seminal fluid analysis (SFA)

Table 2. WHO 2010 (5th Edition) and WHO 2021 (6th Edition) lower fifth percentile (with 95% confidence interval) of semen parameters from men in couples starting a pregnancy within one year of unprotected sexual intercourse leading to a natural conception.

	WHO 2010	WHO 2021
Semen volume (mL)	1.5 (1.4–1.7)	1.4 (1.3–1.5)
Total sperm number (10^6 per ejaculate)	39 (33–46)	39 (35–40)
Total motility (%)	40 (38–42)	42 (40–43)
Progressive motility (%)	32 (31–34)	30 (29–31)
Non progressive motility (%)	1	1 (1–1)
Immotile sperm (%)	22	20 (19–20)
Vitality (%)	58 (55–63)	54 (50–56)
Normal forms (%)	4 (3–4)	4 (3.9–4)

SFA done 24/3/22

Appearance normal

Liquefaction abnormal <60min

Consistency abnormal <2cm long

Volume <0.5mL

pH 7.7

Concentration 0

Motility (A+B) 0

A Progressive 0

B Non progressive 0

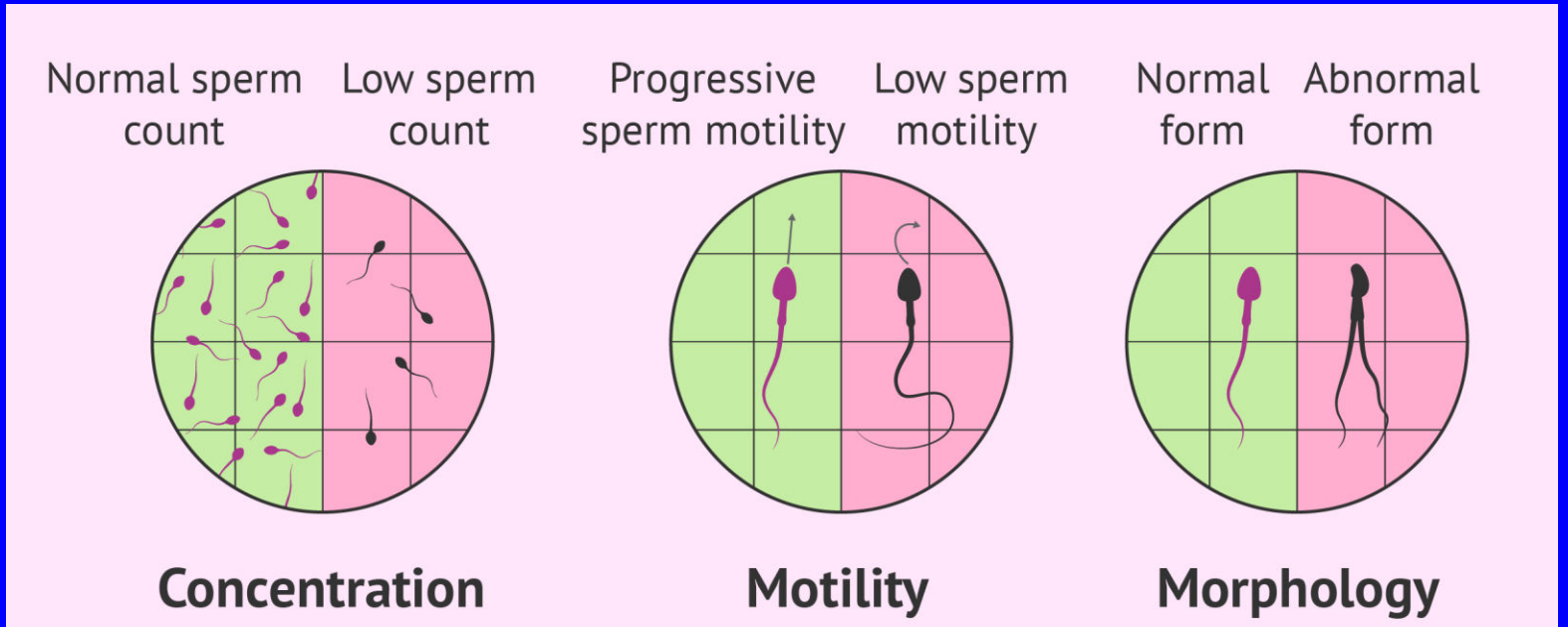
C Immotile 0

Total sperm/ejaculate 0

Morphology 0

WBC 1X10^6

Δ Azoospermia



SFA

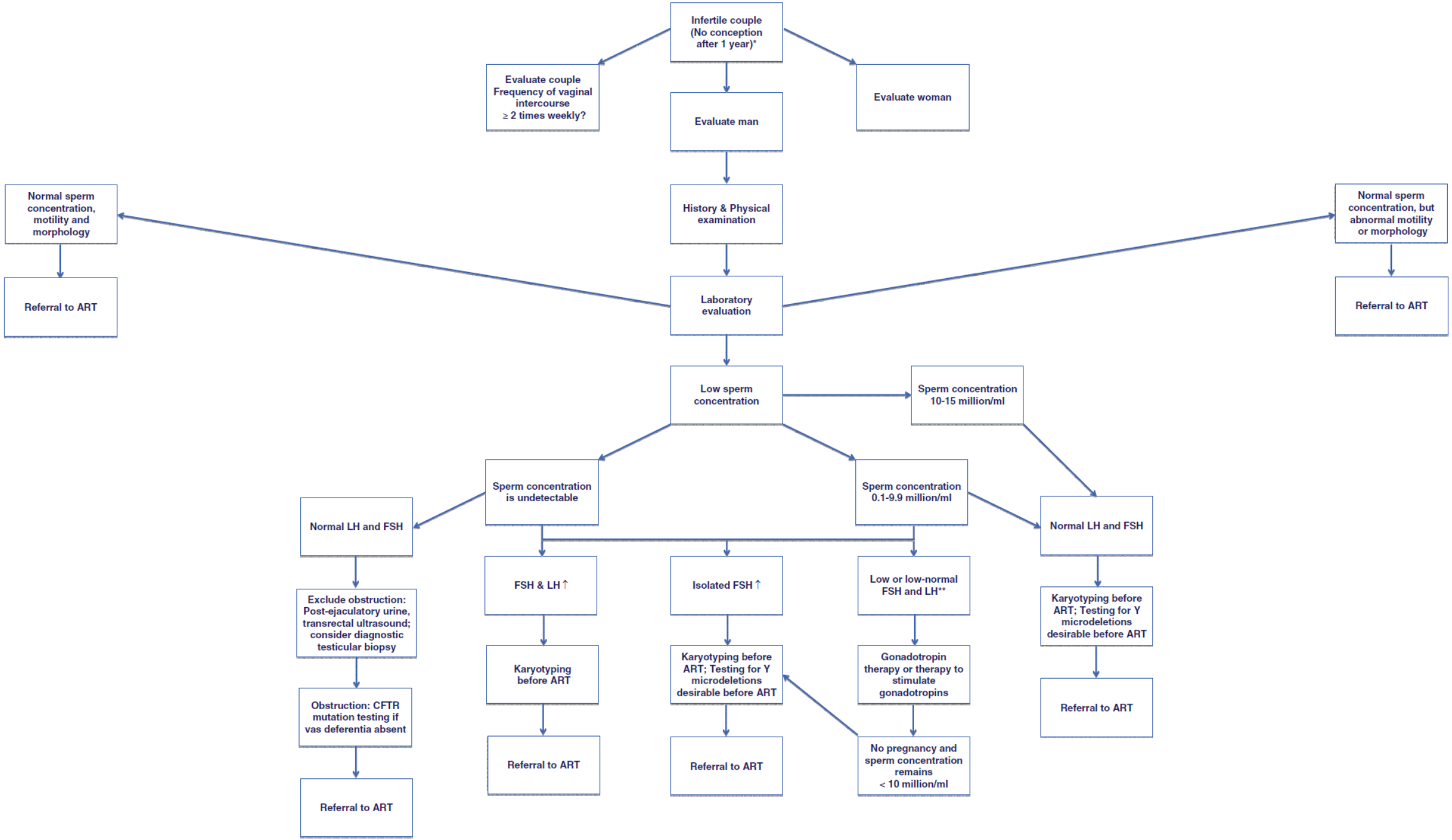
Table 2. World Health Organization Reference Values for Sperm Parameters[a]

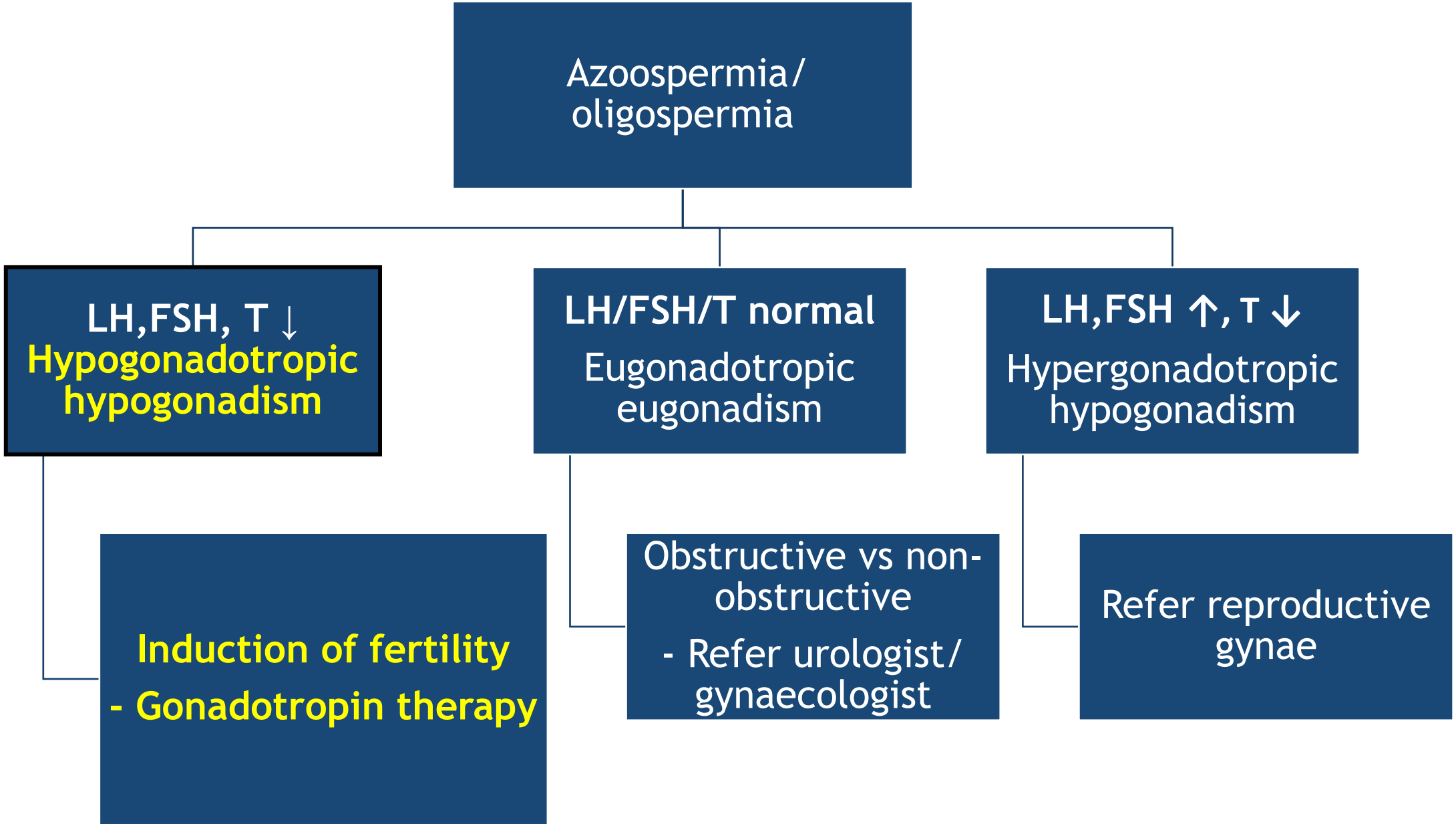
Sperm concentration	≥15 million/mL
Total sperm	≥39 million
Total motility (% of total)	≥40%
Normal morphology (% of total)	≥4%

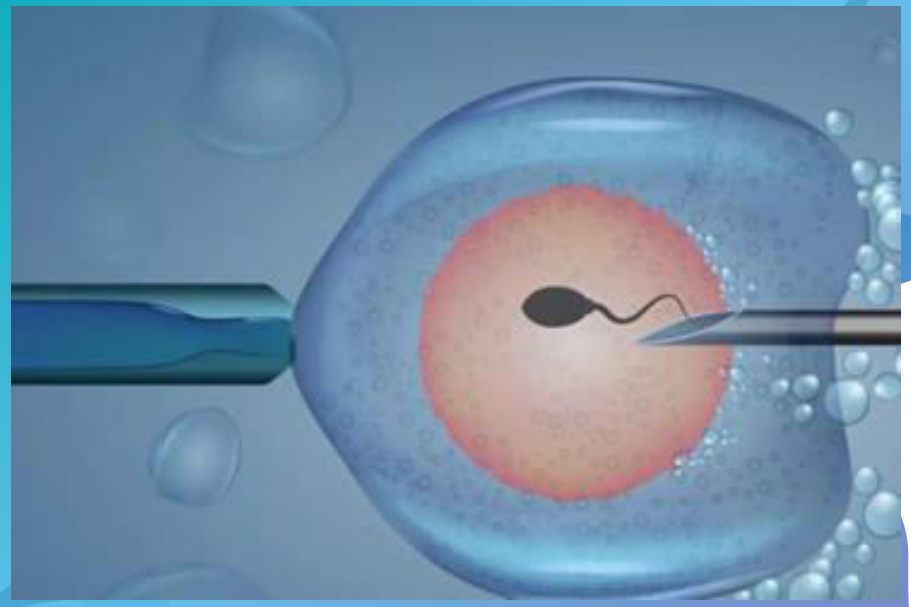
[a] Based on the 5th percentile of recent fathers.

2.0-2.5 ml ejaculate volume as lower limit normal

Cooper TG et al. Hum Reprod Update.2010; 16:231-45


Infertile couple (No conception after 1 year)*
Evaluate couple Frequency of vaginal intercourse ≥ 2 times weekly?
Evaluate man
Evaluate woman
History & Physical examination
Normal sperm concentration, motility and morphology
Referral to ART
Laboratory evaluation
Normal sperm concentration, but abnormal motility or morphology
Referral to ART
Low sperm concentration
Sperm concentration 10-15 million/ml
Sperm concentration is undetectable
Sperm concentration 0.1-9.9 million/ml
Normal LH and FSH
Normal LH and FSH
Exclude obstruction: Post-ejaculatory urine, transrectal ultrasound; consider diagnostic testicular biopsy
Obstruction: CFTR mutation testing if vas deferentia absent
Referral to ART
FSH & LH ↑
Isolated FSH ↑
Low or low-normal FSH and LH**
Karyotyping before ART
Karyotyping before ART; Testing for Y microdeletions desirable before ART
Gonadotropin therapy or therapy to stimulate gonadotropins
Karyotyping before ART; Testing for Y microdeletions desirable before ART
Referral to ART
Referral to ART
No pregnancy and sperm concentration remains < 10 million/ml
Referral to ART


Azoospermia/ oligospermia
LH,FSH, T ↓
Hypogonadotropic hypogonadism
LH/FSH/T normal
Eugonadotropic eugonadism
LH,FSH ↑, T ↓
Hypergonadotropic hypogonadism
Induction of fertility
- Gonadotropin therapy
Obstructive vs non-obstructive
- Refer urologist/ gynaecologist
Refer reproductive gynae



Induction of fertility in hypogonadotropic hypogonad males

Fertility induction begins with production of sperm

- Protocols using Gn aim to replicate the natural endocrine control of spermatogenesis.

- Previously virilised men with adult-onset HH and normal testicular volume respond well to monotherapy in which human chorionic gonadotrophin (hCG) acts as a long-acting LH-analogue stimulating spermatogenesis.

- Congenital HH (CHH) (e.g. Kallmann syndrome)
  - combined gonadotrophin therapy (hCG + FSH)

- Key baseline predictors of successful spermatogenesis-induction include
  - prior spontaneous testicular volume [TV] >4ml
  - serum inhibin B (IB) concentration >60pg/ml
  - no history of maldescended testes
  - Previous use of exogenous T

Fertility induction

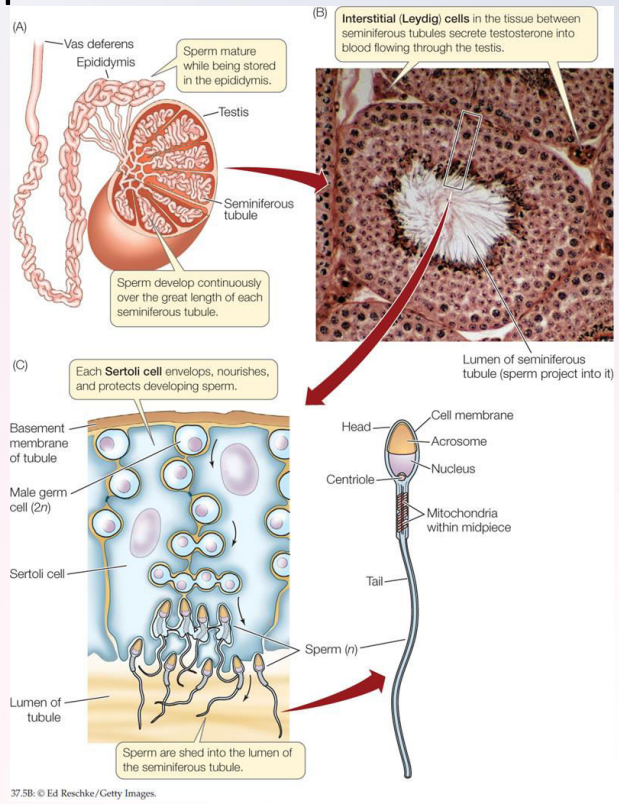
- Initial phase of hCG monotherapy to normalise serum T concentrations prior to the introduction of FSH.
  - Boyar RM et al. J Clin Invest. 1974;54(3):609–18.

- Seminiferous tubules → 90% of TV → size of the testis critical indicator of fertility potential.
- Both FSH and T are necessary for both quantitatively and qualitatively normal spermatogenesis
  - Schaison G et al. J Clin Endocrinol Metab. 1993;77(6):1545–9

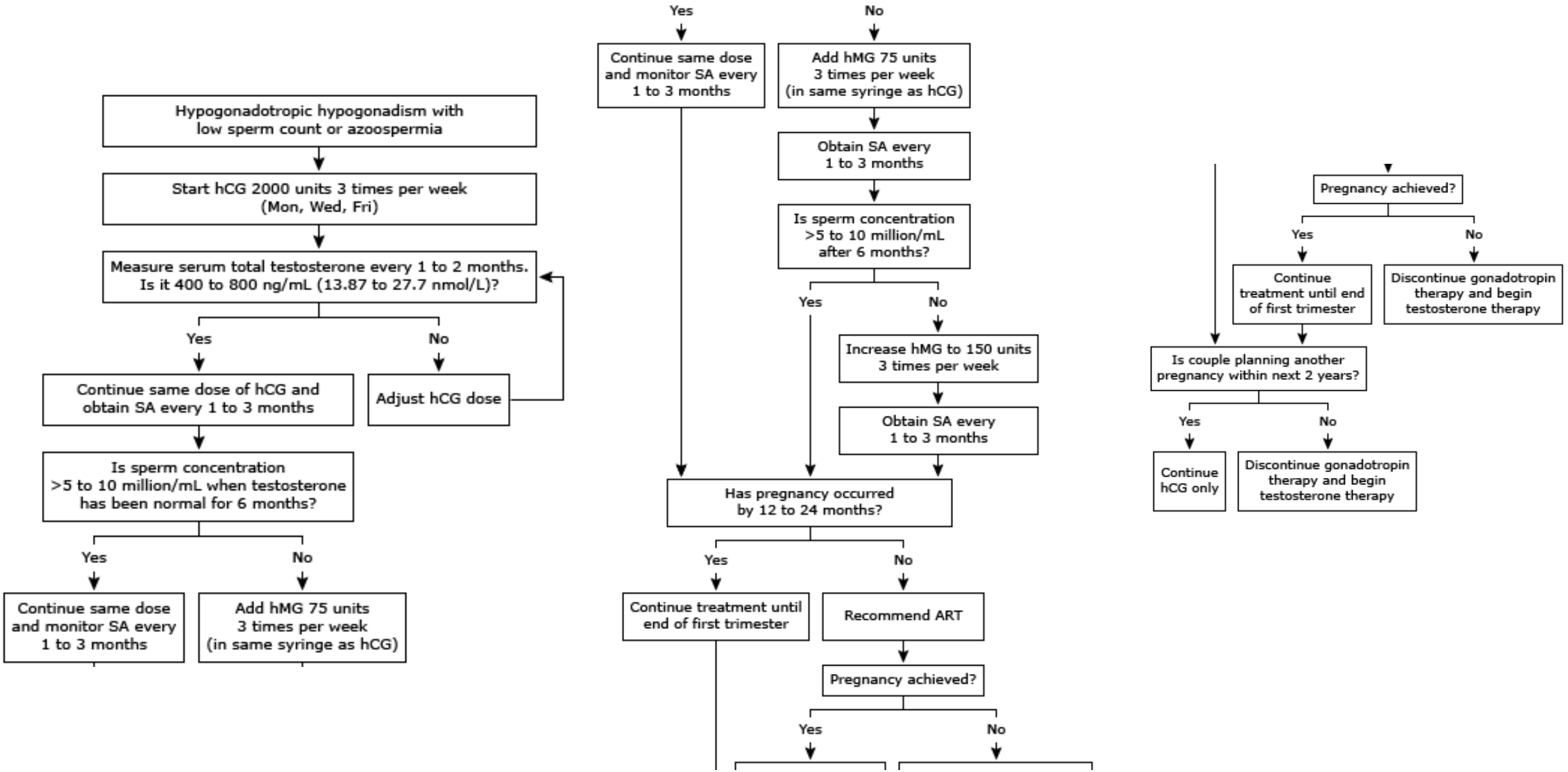
- Intratesticular T concentration higher with hCG (100x) than in peripheral circulation
  - Coviello AD et al. J Clin Endocrinol Metab 90:2595–2602

Dose & timeline

- Maturation of testis requires between 6 months to 3 years
- The dosage of hCG (HuCoG) is adjusted based on trough serum T
- FSH (HuMoG) dosage is titrated based on serum FSH levels and sperm counts.



INDUCTION OF SPERMATOGENESIS



Case study: Patient A

Mr A / history

- Mr A, 33 years old , married 6 years
- 1st seen Dec 2020
- No shaving, erection or ejaculation
- No drugs/ medicine
- Sought fertility treatment
- Non-smoker
- Not anosmic

- Wife – no medical/gynae issues

Mr A / physical examination

- Not dysmorphic
- Thin, BMI 17 kg/m2
- Normotensive
- High- pitched voice
- No gynaecomastia
- Micropenis
- Testicular size : 2 mls each side

Mr A / investigations

- LH - 0.07 IU/L
- FSH – 0.32 IU/L
- T - 0.91 nmol/L
- SFA – unable to ejaculate
- Chromosomal study - XY
- MRI brain - normal

- Imp: Hypogonadotropic hypogonadism 2° to non-anosmic Kallman Syndrome

Mr A / progress

- Aug 2021 → underwent induction of fertility
- Progress until Feb 2023
- Pubic hair Tanner 4
- Right testis 8 ml
- Left testis 6 ml
- Penile length : 5.5 → 7cm

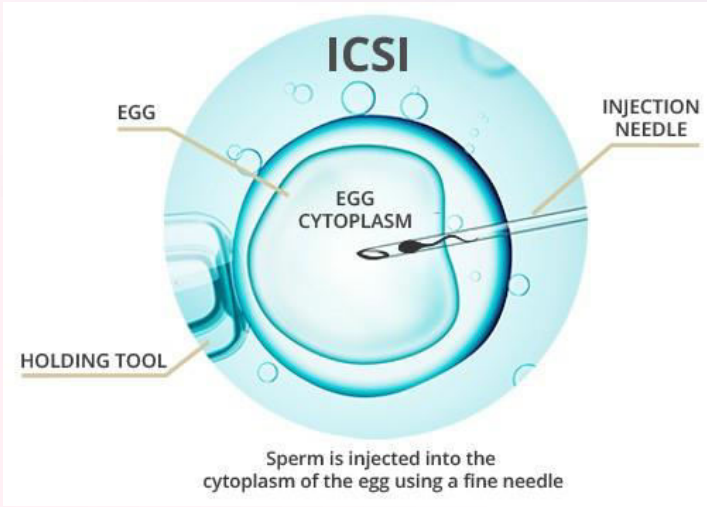
- Given hCG monotherapy for 19 months (until March 23)
- Still azoospermia although T and TV improved
- hMG added for 3/12 – immotile sperm , still severe oligospermia

Date	Duration of Rx (months)	T (nmol/mL)	Medication	Testicular size	SFA
Aug 21	0	2.98	hCG1500 iu 3x/week	2ml B/L	Unable to ejaculate
Dec 21	4	3.54	hCG 2500 iu 3x/week		
March 22	7	7.18	hCG 2500 iu 3x/week		Volume: 1.6 mL Azoospermia
Sept 22	13	8.03	hCG 2500 iu 4x/week		
Jan 23	17	10.41	hCG 2500 iu 5x/week	6 ml B/L	
March 23	19	10.88	hCG 2500 iu 5x/week + HMG 75 iu 3x/week		Volume: 1.5 mL Count: $2x10^6$ (all immotile)
April 23	20	12.48	hCG 2500 iu 5x/week + HMG 75 iu 3x/week		Volume : 2 mL 3-5 motile sperm outside grid

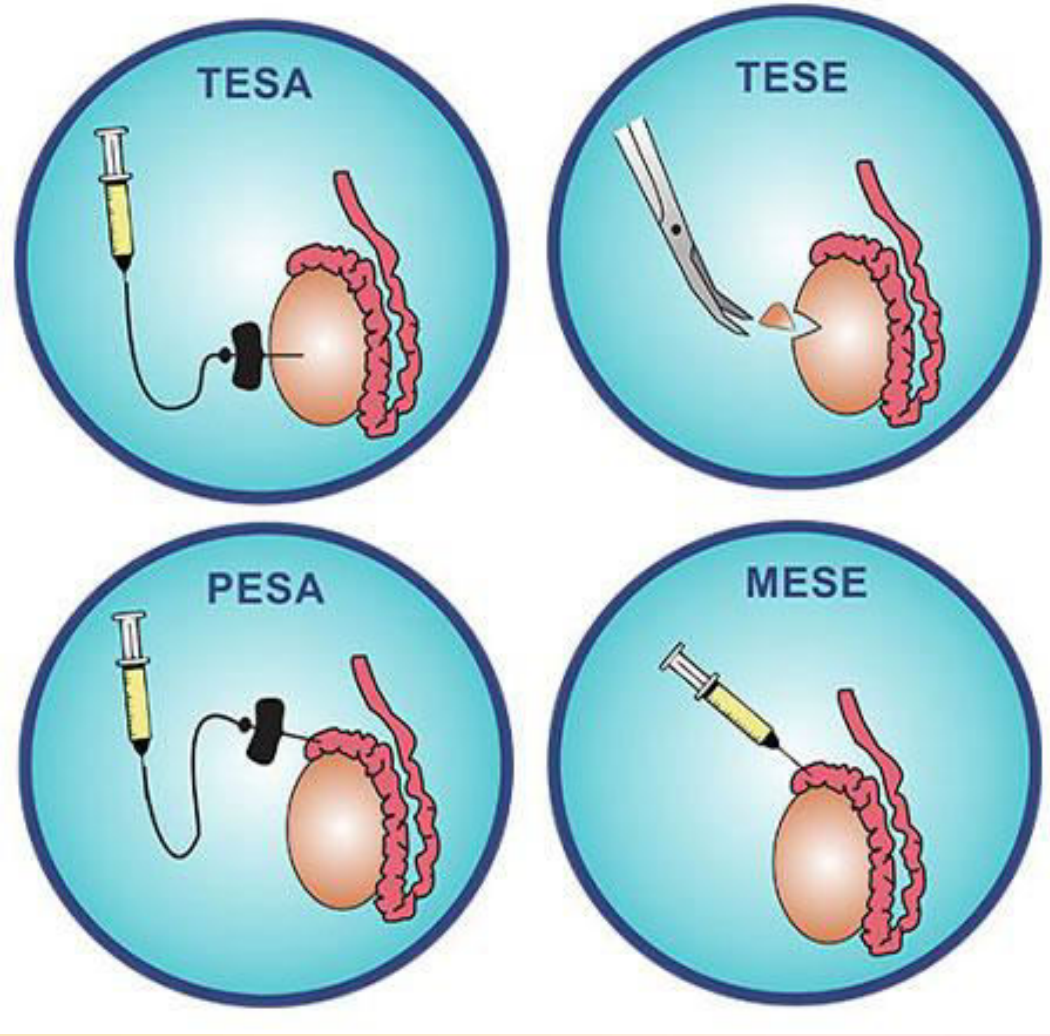
Patient A / IVF + ICSI

- Patient able to produce sperm but many immotile
- Not suitable for cryopreservation
- Proceeded with IVF
- KIV SSR if unable to produce sperm for fertilization on day of OPU day
- Able to produce sperm – normal morphology and motile → ICSI

- Successful pregnancy
- Wife delivered a pair of twins



SSR



TESA – TEsticular Sperm Aspiration

PESA - Percutaneous Epididymal Sperm Aspiration

TESE – TEsticular Sperm Extraction

MESA - Micro Epididymal Sperm Aspiration

**** Usually not used in hypogonadotropic hypogonadism**

Case study: Patient B

Patient B

- 33/ man , referred from PGH
- Non-smoker
- Acromegaly, underwent TSS in Oct 2023 – panhypopituitarism + DI
- on replacement therapy post op, no T
- Married 1 year before diagnosis
- Post op – loss of libido and erection
- Serum T (nmol/L) - 10 (June 23) → 0.5 (Dec 23) → 0.6 (Jan 24)
- FSH (IU/L) - 8.2 (June 230 → 3.7 (Dec 23)→ 3.5 (Jan 24)
- LH (IU/L) - 3.7 (June 23) → 2.6 (Dec 23) → 2.3 (Jan 24)
- Estradiol (pg/mL) - 18.6
- Prolactin (IU/L) - 52

Patient B

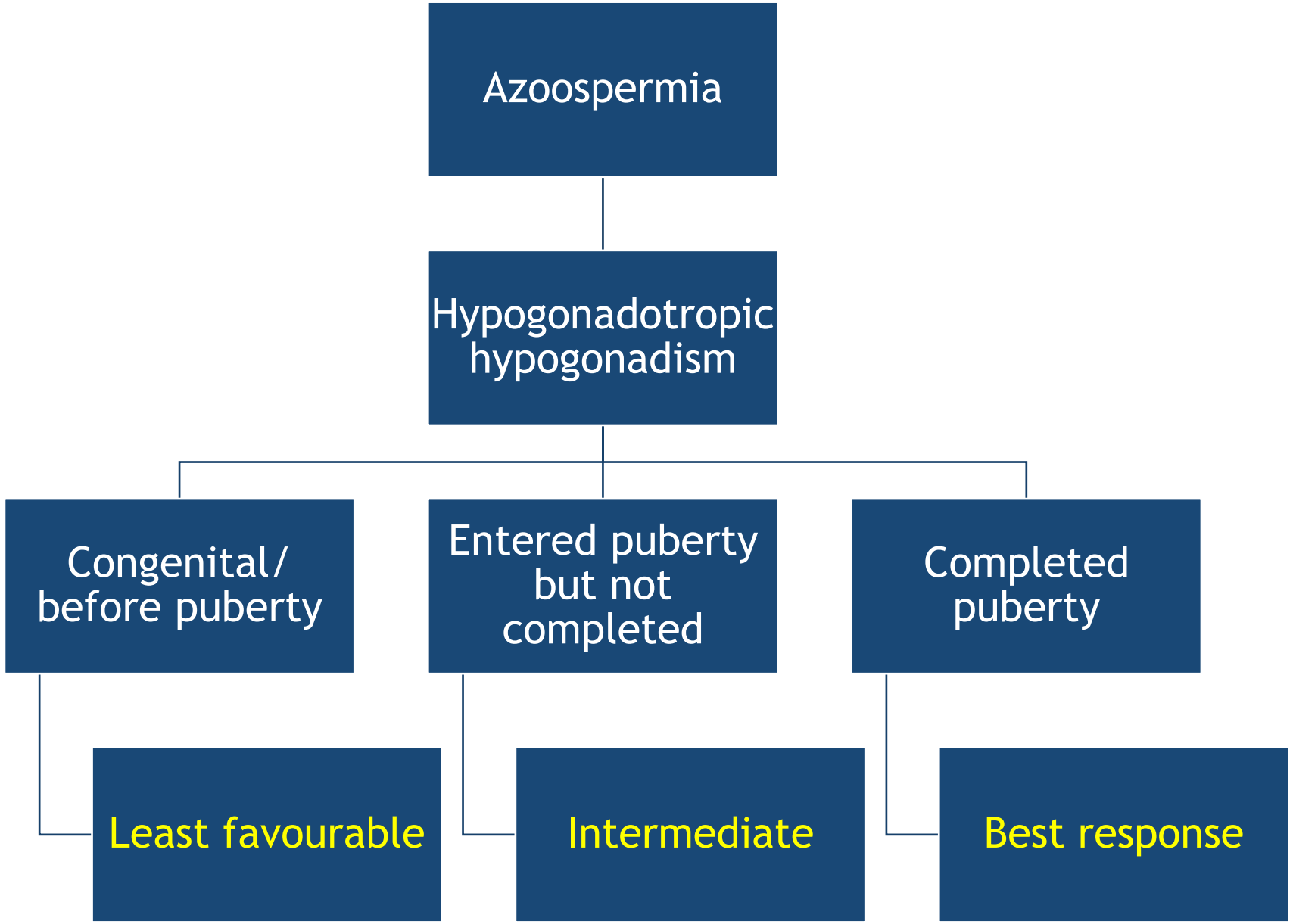
- Imp: Hypogonadotropic hypogonadism 2° **to** TSS for acromegaly
- Started on Letrozole 2.5 mg od , after 2/12 not keen to continue
- Still anejaculation (March 24)
- Not keen until July 2024
- Agreed for induction of fertility and IVF

- O/E
- 64 kg , height 165 cm, BMI 23.5 kg/m2
- Penis – normal size
- Testes 10 ml B/L

Date	Duration of Rx (months)	T (nmol/L)	Medication	Testicular size	SFA
June 23	-	10		10 ml B/L	Unable to ejaculate
Oct 23	TSS				
Dec 23		0.5			
Jan 24		0.6	Letrozole 2.5 mg od		Still ED anejaculation
March		1.83			
July 24	0	0.7	HCG 1500 iu 3x/week		
Oct 24	3	41.8			Volume : 2.5 mL Count 92 x 10^6/ml Motility (a+b): 60% Morphology: 4%

Patient B

- Has EM erection , maintain erection and ejaculate
- Shaves weekly
- T (nmol/L) 41.8 (5.5 -29.0)
- Estradiol (pmol/L) – 258.0 (high)
- Sperm cryopreservation
- COS + ICSI



Response to fertility treatment

SUMMARY

- Hypogonadotropic hypogonadism male subfertility is treatable using gonadotropin induction
- Induction requires long-acting LH analog (hCG) for T production to initiate spermatogenesis at doses between 1500 -2500 iu 3x/week
- Monotherapy with hCG might be sufficient, some require FSH in form of hMG 75-150 iu/3x week especially if insult is prepubertal.
- Success of fertility induction in hypogonadotropic hypogonadism males depends on
  - previous testicular volume
  - Biochemical assessment to guide choice of therapy

References

SPECIAL FEATURE

Approach to the Patient

Approach to Male Infertility and Induct of Spermatogenesis

Bradley D. Anawalt

University of Washington Medical Center, Department of Medicine, Seattle, Washington 98195

Accreditation and Credit Designation Statements
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Learning Objectives
Upon completion of this educational activity, participants should be able to:

Medicine®

OBSERVATIONAL STUDY

OPEN

Efficacy and Outcome Predictors of Gonadotropin Treatment for Male Congenital Hypogonadotropic Hypogonadism

A Retrospective Study of 223 Patients

Zhaoxiang Liu, MD, Jangfeng Mao, MD, Xueyan Wu, MD, Hongli Xu, MS, Xi Wang, MD, Bingkun Huang, MD, Junjie Zheng, MD, Min Nie, MD, and Hongbing Zhang, PhD

REVIEW

Clinical Management of Congenital Hypogonadotropic Hypogonadism

Review > Clin Endocrinol (Oxf). 2018 Dec;89(6):712-718. doi: 10.1111/cen.13850. Epub 2018 Oct 9.

Fertility induction in hypogonadotropic hypogonadal men

Matthew Prior [1], Jane Stewart [1], Kevin McEleny [1], Andrew A Dwyer [2], Richard Quinton [3 4]

Affiliations + expand

PMID: 30194850 DOI: 10.1111/cen.13850

life

MDPI

Review

The Sixth Edition of the WHO Manual for Human Semen Analysis: A Critical Review and SWOT Analysis

Florence Boitrelle [1,2], Rupin Shah [3], Ramadan Saleh [4,5], Ralf Henkel [6,7,8,9], Hussein Kandil [10], Eric Chung [11,12], Paraskevi Vogiatzi [13], Armand Zini [14], Mohamed Arafa [8,15,16] and Ashok Agarwal [8,*]

0021-972X/05/$15.00/0
Printed in U.S.A.

The Journal of Clinical Endocrinology & Metabolism 90(11):5928–5936

doi: 10.1210/jc.2005-0962

Reproductive Hormone Reference Intervals for Healthy Fertile Young Men: Evaluation of Automated Platform Assays

Ken Sikaris, Robert I. McLachlan, Rymantas Kazlauskas, David de Kretser, Carol A. Holden, and David J. Handelsman

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- JM Fairuz – Reproductive Medicine Unit, O&G Department, HSBAS
- All endocrinology fellows – past and present in HSBAS
- MOs in O&G Department HSBAS
- SN Magenderan, SN Suhaili – DE, DRC HSBAS
- Patients in Reproductive Medicine Unit O&G Department, HSBAS

THANK YOU VERY MUCH FOR YOUR KIND ATTENTION