SAPIENZA
UNIVERSITÀ DI ROMA

DIPARTIMENTO
DI MEDICINA SPERIMENTALE

CERTIFICATE FOR TRAINING CENTRE IN ANDROLOGY
E·A·A
EUROPEAN ACADEMY OF ANDROLOGY

LABORATORIO DI SEMINOLOGIA
BANCA DEL SEME
"LOREDANA GANDINI"

SAPIENZA
UNIVERSITÀ DI ROMA

I WEBINAR DEL MARTEDÌ

Appuntamenti d'informazione
medico-scientifica sulla fibrosi cistica

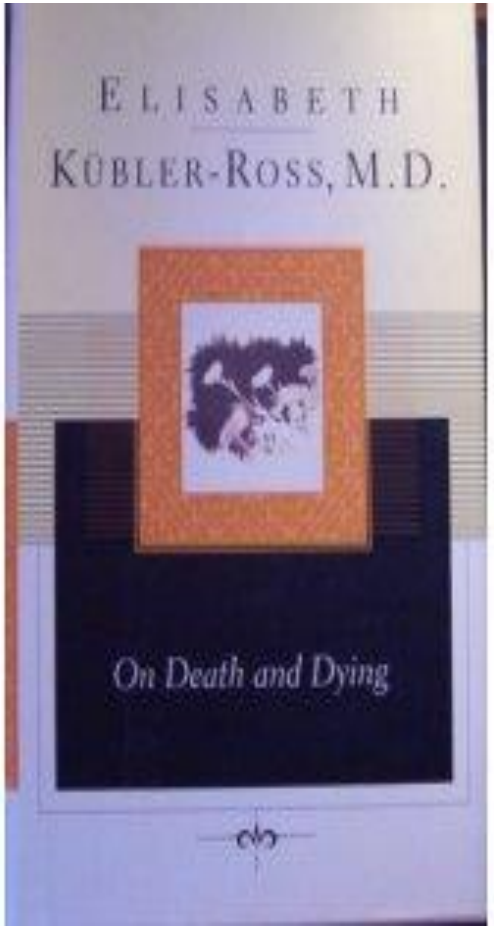
15 FEBBRAIO	1 MARZO	15 MARZO

ore 17.00 PARTECIPA → zoom

francesco.lombardo@uniroma1.it

Reazione alla notizia

- **Sorpresa**
- **Negazione**
- **Sofferenza**
- **Frustrazione**
- **Vergogna**
- **Depressione**
- **Dolore**
- **Risoluzione**
- **Stress**



INFERTILITÀ MASCHILE

ESAME DEL LIQUIDO SEMINALE

PATOLOGICO

OLIGOZOOSPERMIA
OAT

AZOOSPERMIA

DIAGNOSI MOLECOLARE DELL'AZOOSPERMIA

L'AZOOSPERMIA è la condizione più grave caratterizzata dall'assenza di spermatozoi nel liquido seminale. Talvolta sono presenti le cellule germinali immature, in particolare spermatociti e spermatidi, identificabili con colorazioni specifiche.

Assenza di spermatozoi nel liquido seminale

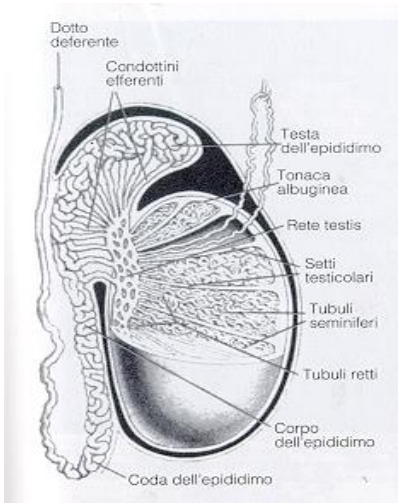
Esami Seminali **12.839**

Azoospermici **817**

6.3 %

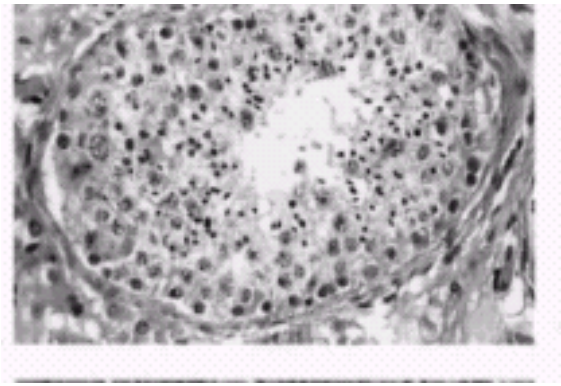
AZOOSPERMIA SECRETIVA → **87%**

AZOOSPERMIA OSTRUTTIVA → **13%**



AZOOSPERMIA

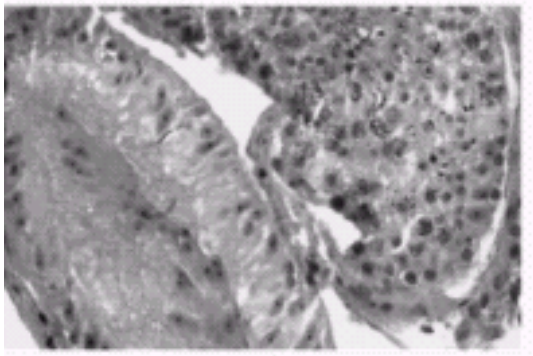
OSTRUTTIVA



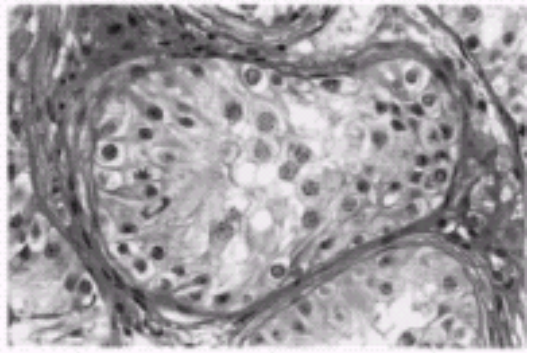
SECRETORIA

SCOS

MISTA

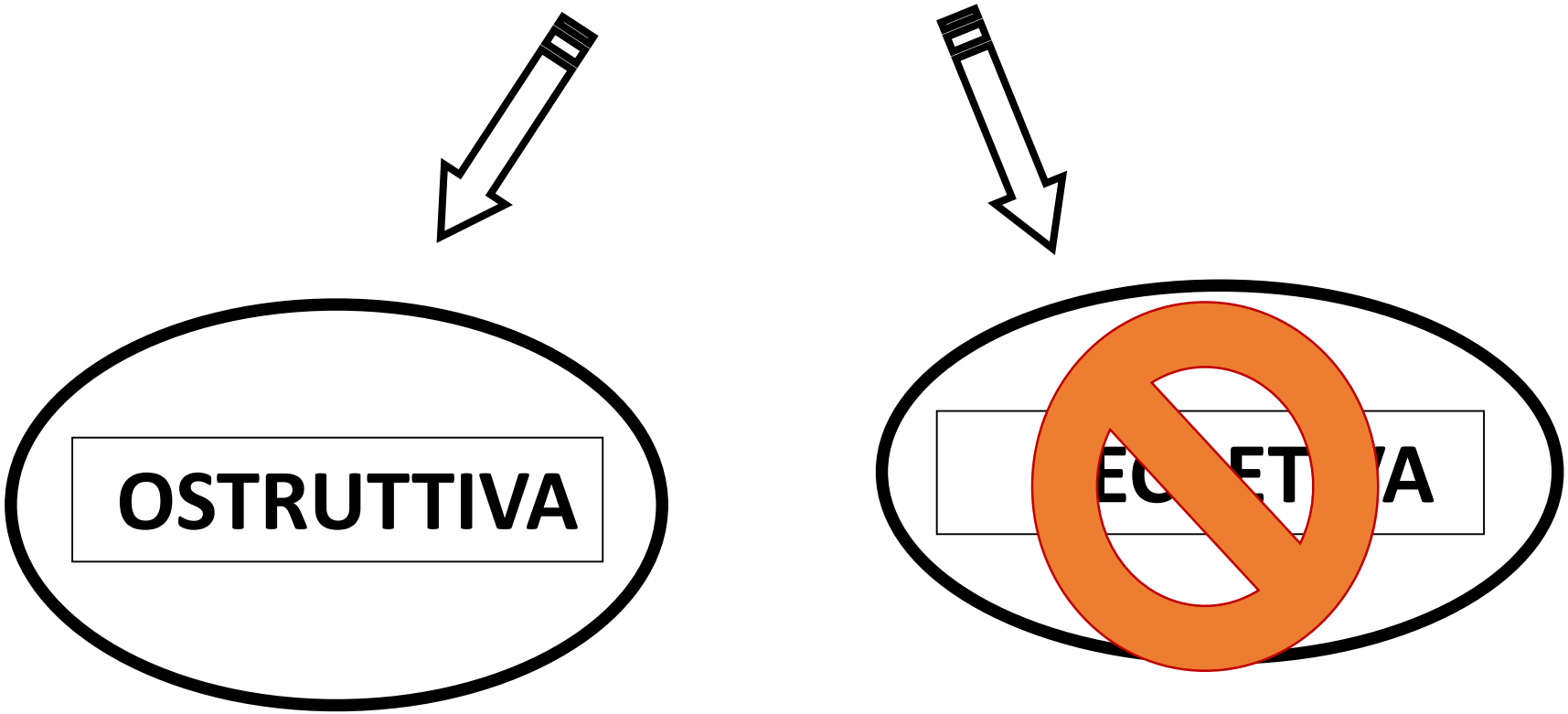


Ipospermatogenesi



arresto spermatogenico

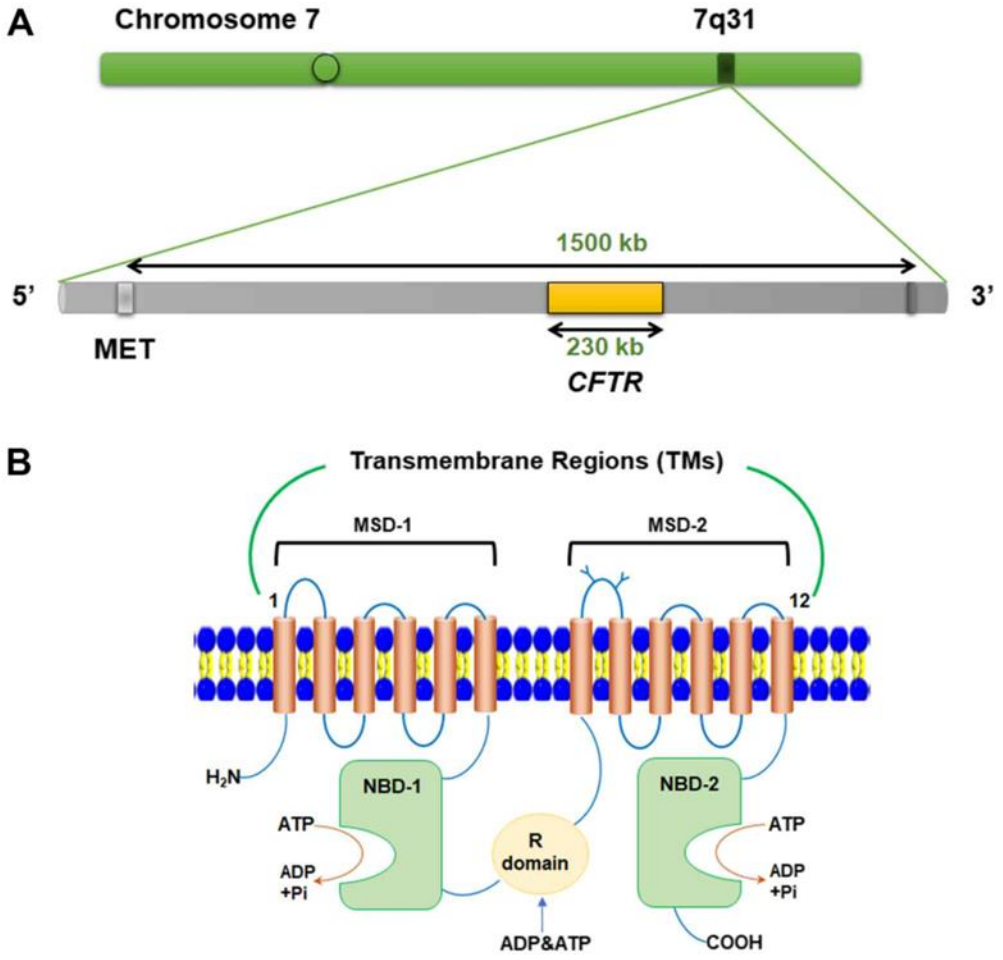
DIAGNOSI DIFFERENZIALE



AZOOSPERMIA OSTRUTTIVA

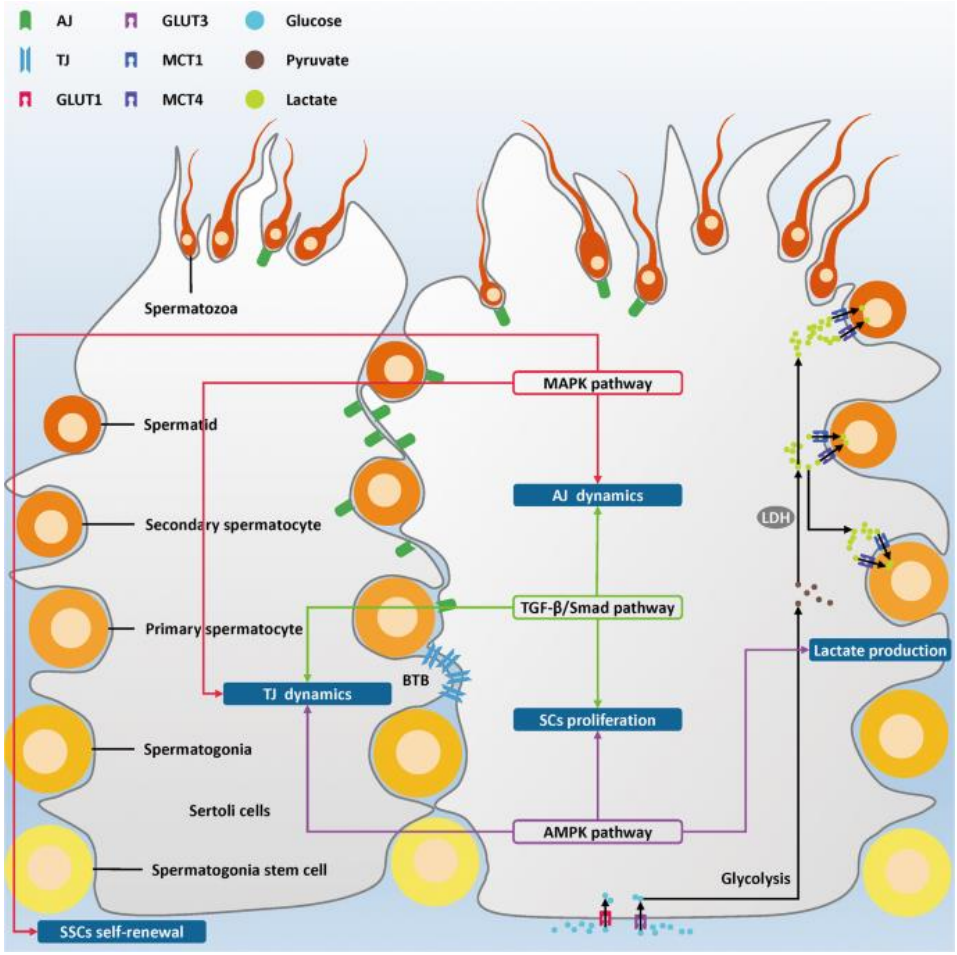
- Dati anamnestico – clinici
- Esame del Liquido Seminale
- Studio ormonale
- Studio Biochimica Seminale
- Ecografia Testicolare
- Ecografia prostatica
- Screening Fibrosi Cistica
- Citoaspirato testicolare
- Biopsia testicolare
- Crioconservazione del seme o del tessuto testicolare

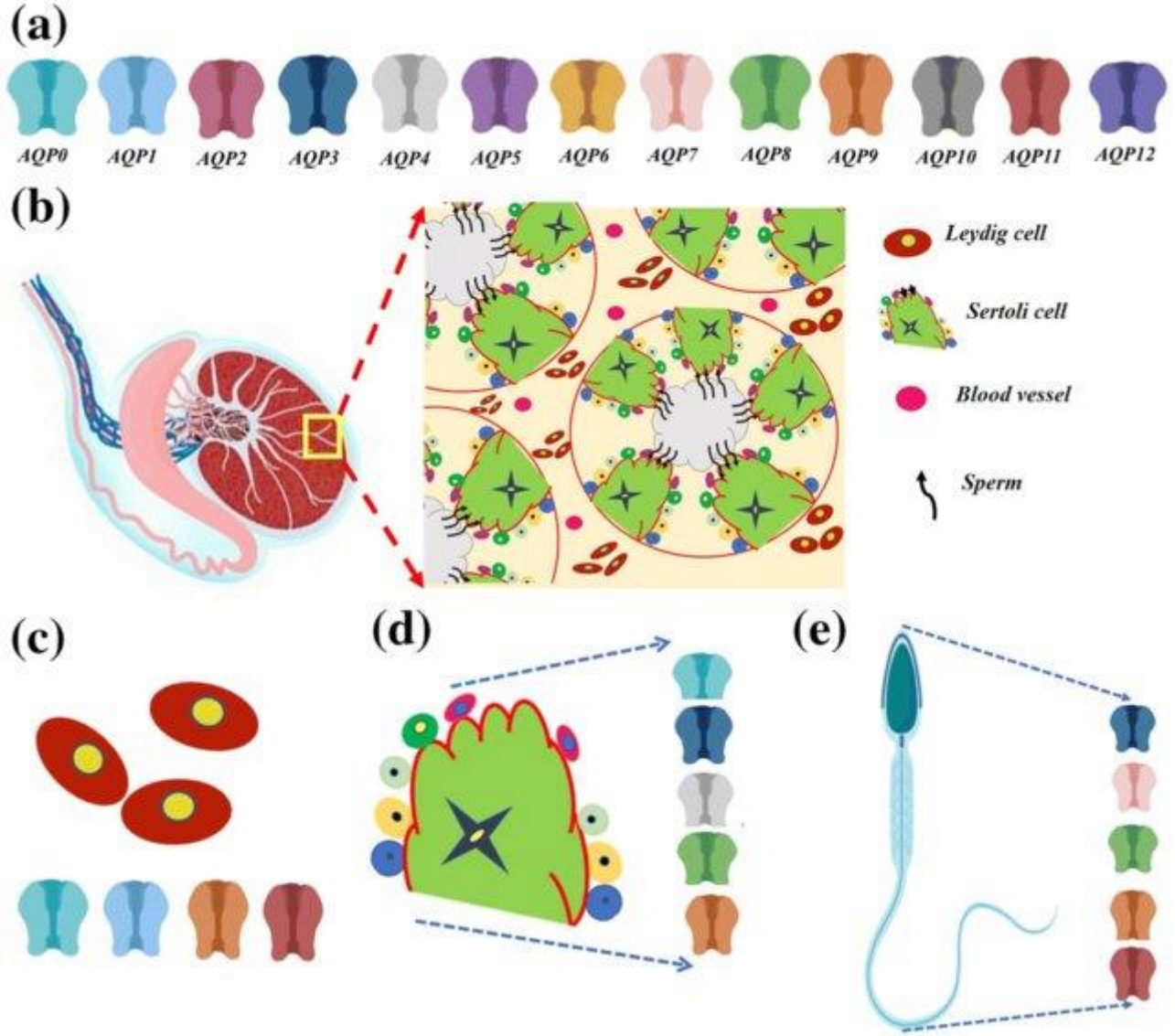
Effetti delle mutazioni del gene CFTR sulla spermatogenesi



Recent data suggest the implication of CFTR protein in the regulation of tight junction assembly and differentiation of epithelial cell.

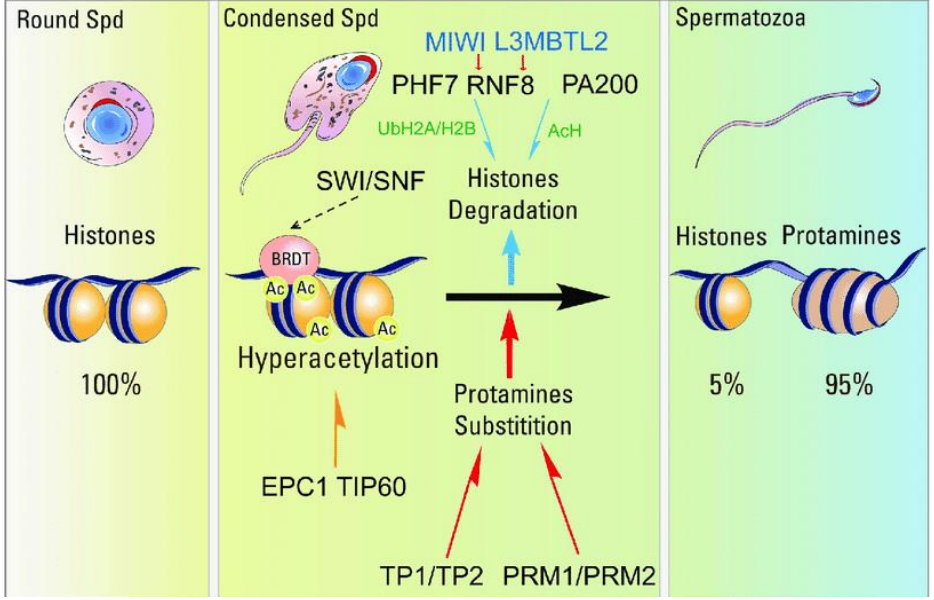
Moreover current evidence indicates the expression of defective CFTR has profounder effect on fatty acid, cholesterol and sphingolipid metabolism, as well as on the membrane phospholipid composition

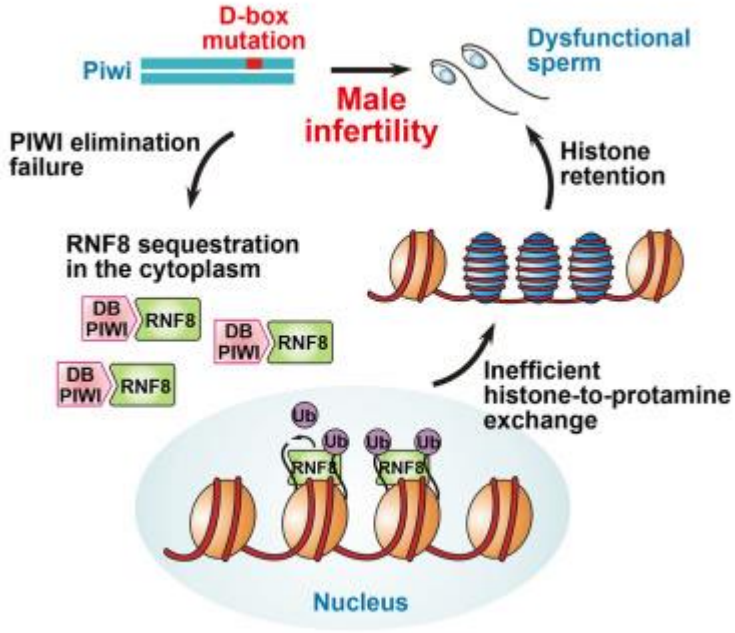


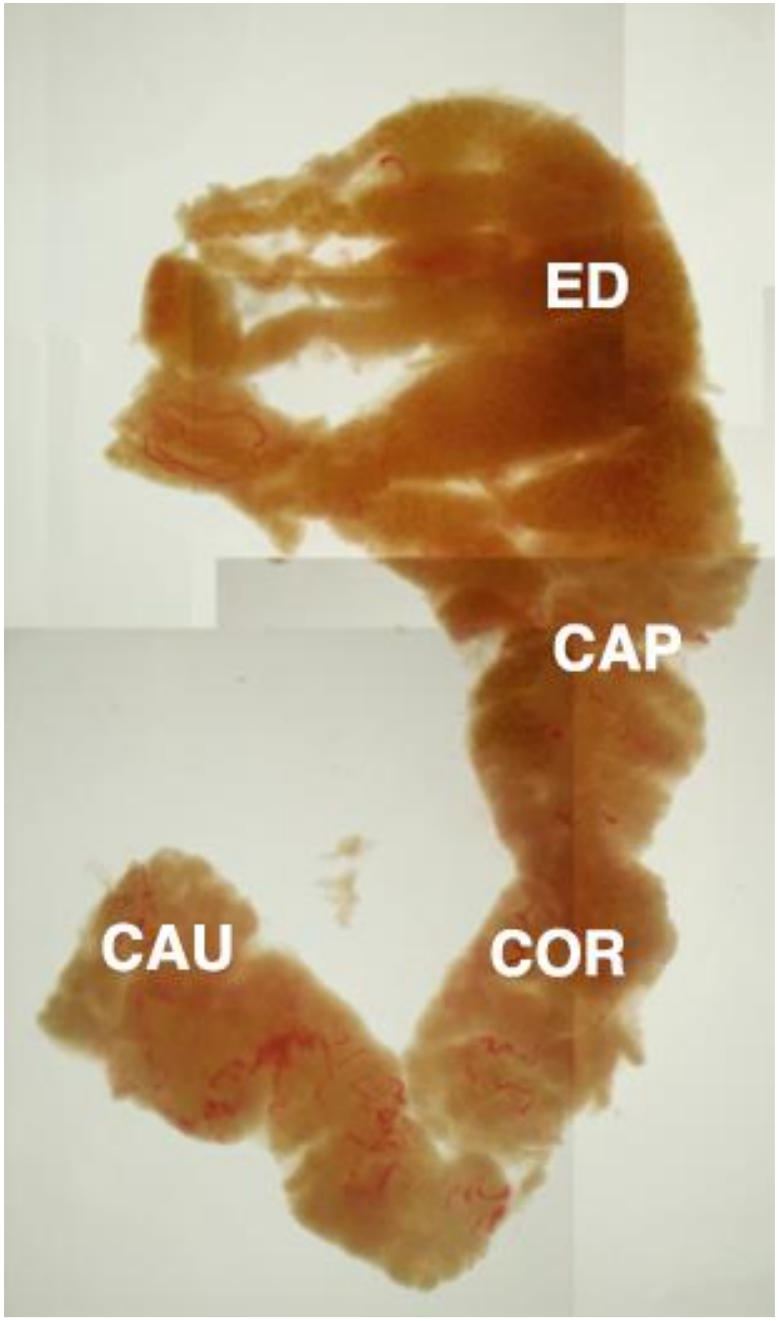


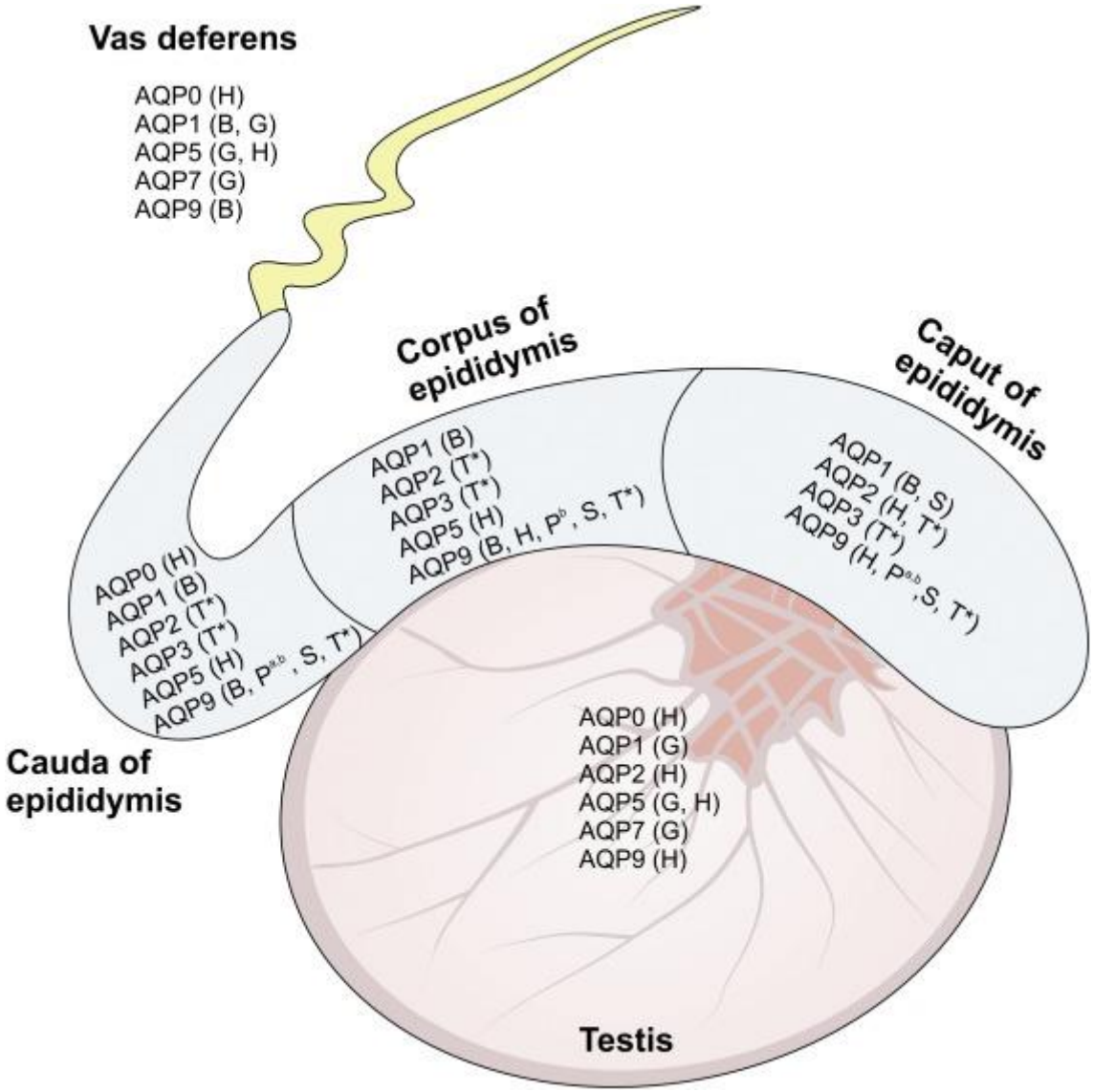
CFTR has been shown to maintain water homeostasis through its interaction with AQP9 or through direct diffusion of water

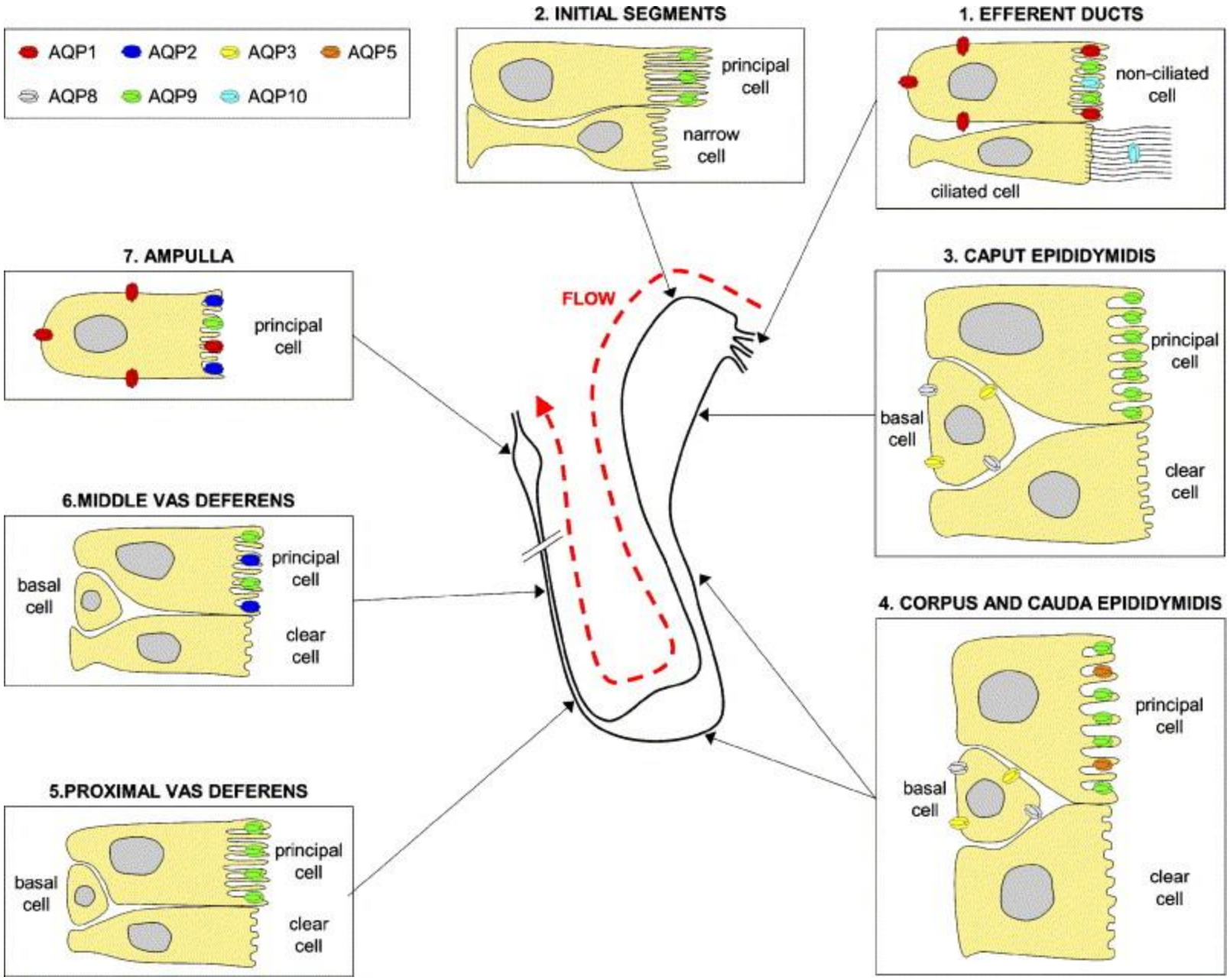
Inhibition of CFTR in somatic Sertoli cells from the testis or depletion of extra-cellular HCO^{3-} is thought to reduce FSH-stimulated, sAC-dependent cAMP production, and phosphorylation of CREB, the key transcription factor in spermatogenesis. Indeed, alteration of CFTR protein leads to a diminution of CREB and protamine 2 levels, suggesting a possible impact on sperm nuclear integrity at the post-meiotic stages of spermatogenesis, particularly at the step of histone-protamine exchange, which is essential for sperm head condensation and DNA stabilization.






ED
CAP
CAU
COR


Vas deferens
AQP0 (H)
AQP1 (B, G)
AQP5 (G, H)
AQP7 (G)
AQP9 (B)
Corpus of epididymis
AQP1 (B)
AQP2 (T*)
AQP3 (T*)
AQP5 (H)
AQP9 (B, H, P[b], S, T*)
Caput of epididymis
AQP1 (B, S)
AQP2 (H, T*)
AQP3 (T*)
AQP9 (H, P[a,b], S, T*)
AQP0 (H)
AQP1 (B)
AQP2 (T*)
AQP3 (T*)
AQP5 (H)
AQP9 (B, P[a,b], S, T*)
Cauda of epididymis
AQP0 (H)
AQP1 (G)
AQP2 (H)
AQP5 (G, H)
AQP7 (G)
AQP9 (H)
Testis


AQP1
AQP2
AQP3
AQP5
AQP8
AQP9
AQP10
2. INITIAL SEGMENTS
principal cell
narrow cell
1. EFFERENT DUCTS
non-ciliated cell
ciliated cell
7. AMPULLA
principal cell
FLOW
3. CAPUT EPIDIDYMIDIS
principal cell
basal cell
clear cell
6.MIDDLE VAS DEFERENS
basal cell
principal cell
clear cell
4. CORPUS AND CAUDA EPIDIDYMIDIS
principal cell
basal cell
clear cell
5.PROXIMAL VAS DEFERENS
basal cell
principal cell
clear cell

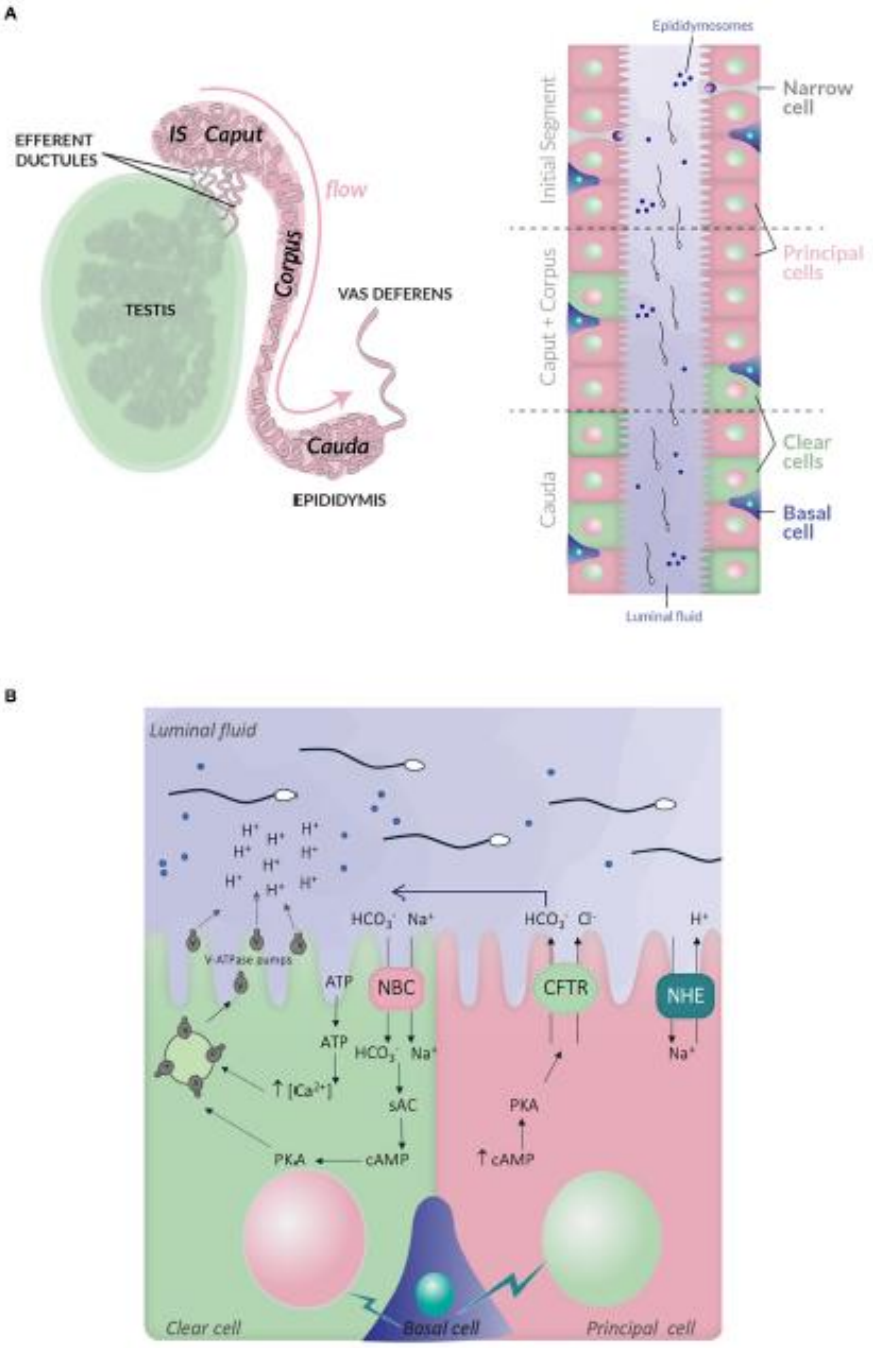
Principal Cells (PCs), Clear Cells (CCs), Narrow Cells (NCs), and basal cells; the luminal fluid shows epididymosomes, which are small vesicles transferring material from epithelia cells to the sperm cells.

CCs expressed the V-ATPase pumps, which expression at the plasma membrane is induced by $HCO3^-$ and c-AMP dependent pathway.

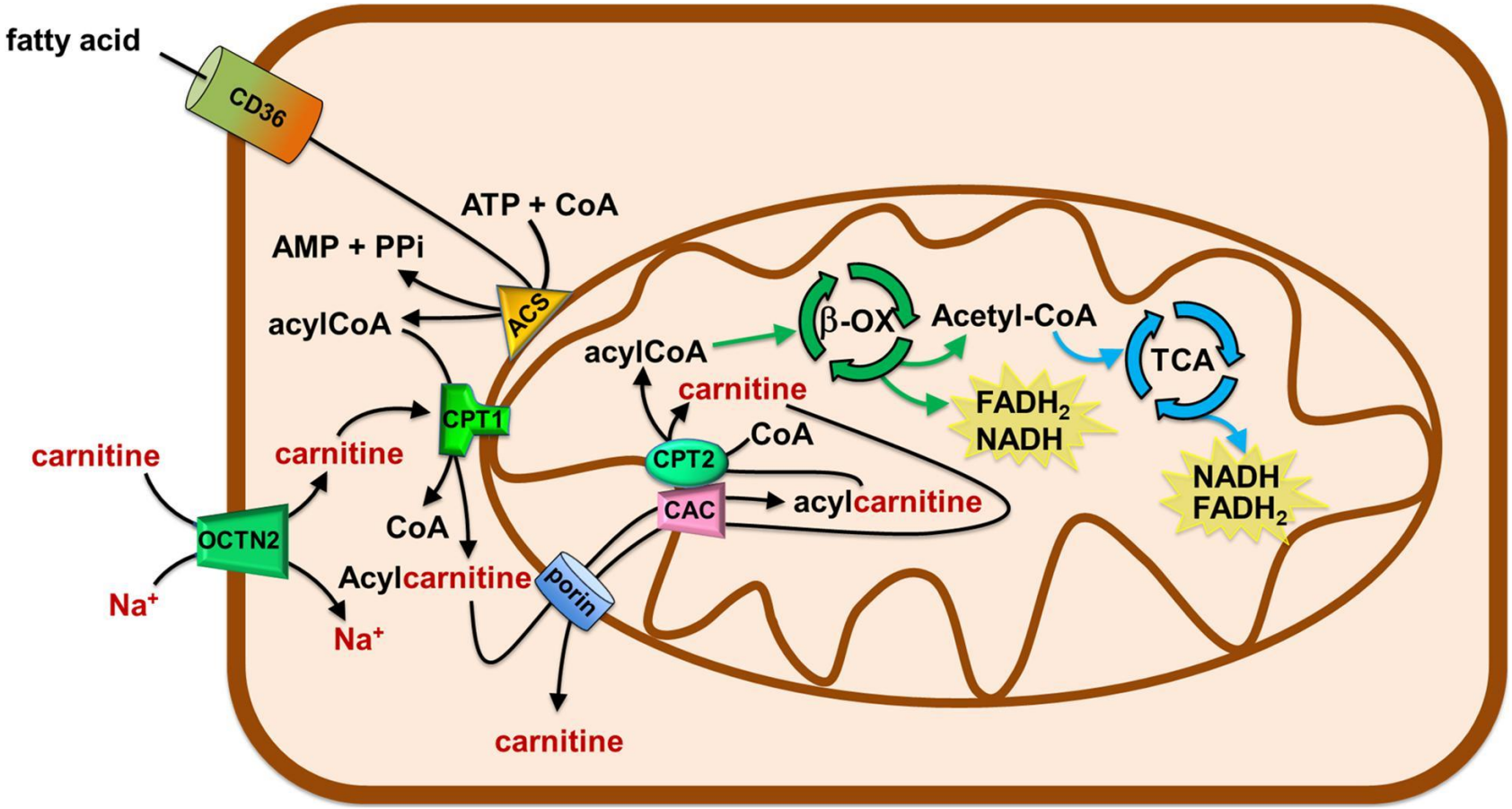
The $HCO3^-$ influx in CCs is mediated by the NBC sodium- $HCO3^-$ transporter. ATP also induces intracellular rise of Ca2+, which increase V-ATPase translocation at the plasma membrane and proton secretion.

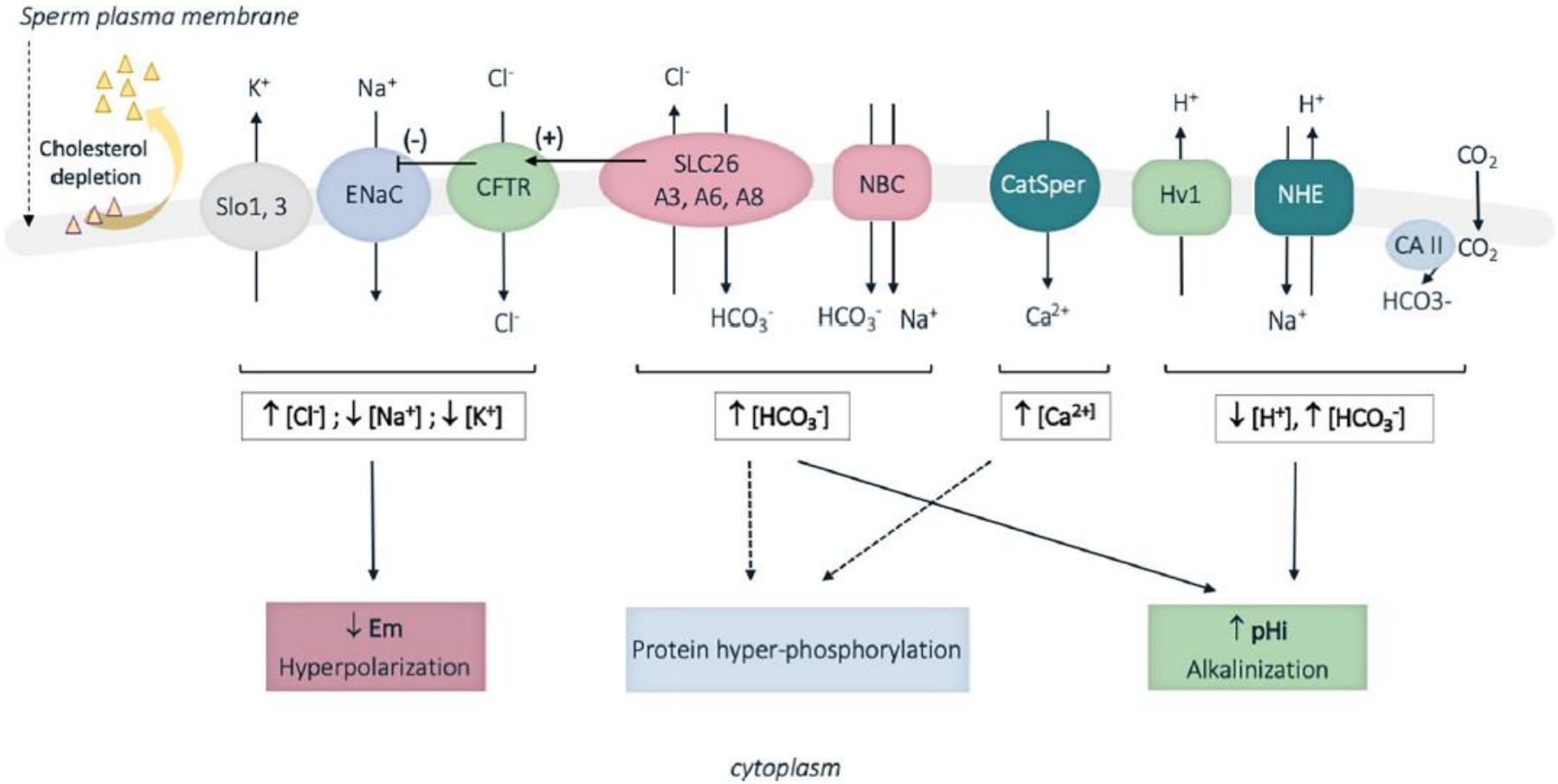
PCs express the NHE3 sodium-proton antiporter, which contributes to proton secretion and luminal acidification. They also secrete $HCO3^-$ through the CFTR channel.

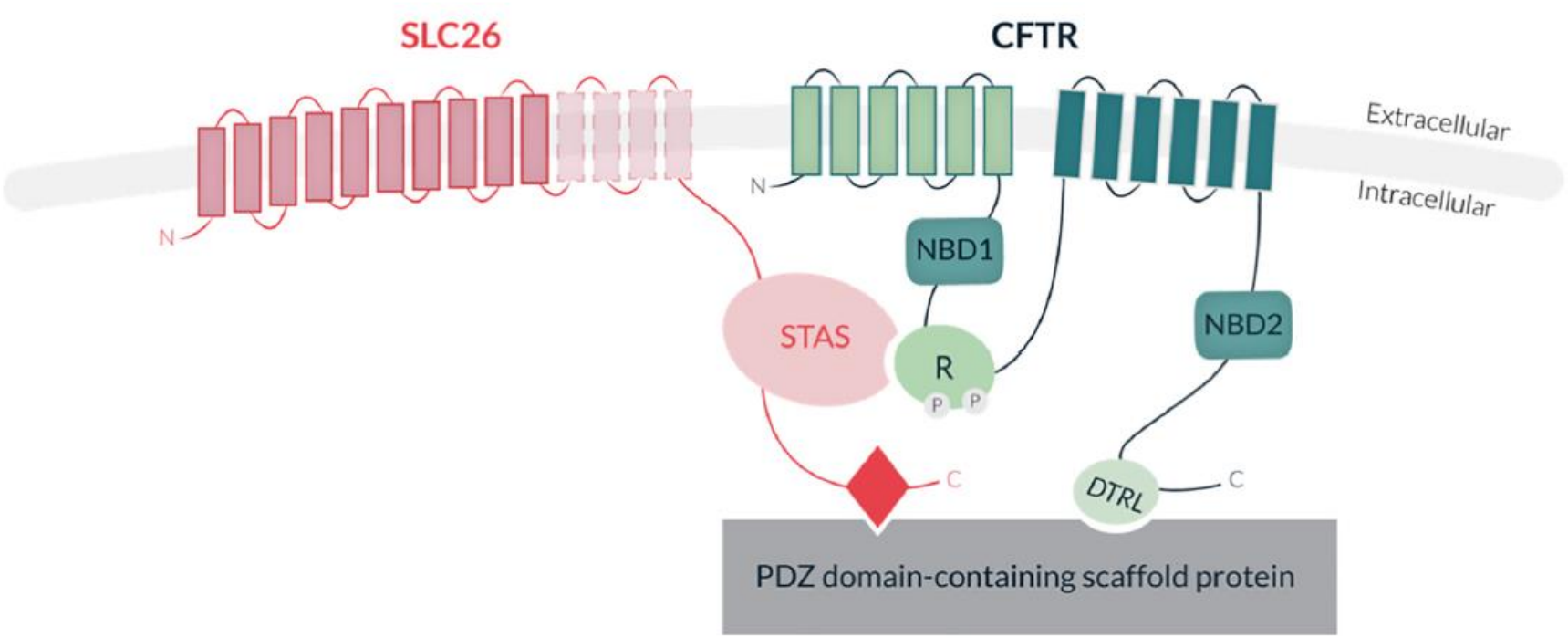
Lastly, basal cells transmit physiological cues, in particular during sexual arousal, which regulate the activity of principal and CCs.



CF mutation is associated with lipid profile abnormalities: possible relationship with sperm parameters




Sperm plasma membrane
Cholesterol depletion
K+
Na+
Cl-
(-)
(+)
Cl-
H+
H+
CO2
Slo1, 3
ENaC
CFTR
SLC26 A3, A6, A8
NBC
CatSper
Hv1
NHE
CA II
CO2
HCO3-
Cl-
HCO3- HCO3- Na+
Ca2+
Na+
↑ [Cl-] ; ↓ [Na+] ; ↓ [K+]
↑ [HCO3-]
↑ [Ca2+]
↓ [H+], ↑ [HCO3-]
↓ Em
Hyperpolarization
Protein hyper-phosphorylation
↑ pHi
Alkalinization
cytoplasm



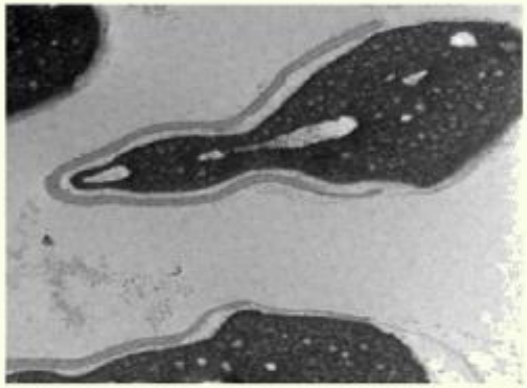
Schematic representation of SLC26 protein structure and interaction with the CFTR

SLC26 proteins share a conserved transmembrane region of 10–14 hydrohobic spans, associated with their anion transport activity, and a cytoplasmic STAS domain (Sulfate Transporter and Anti-Sigma factor antagonist), involved in protein-protein interaction and regulation. Some members also contain a PDZ binding motif at their carboxy-terminal extremity.

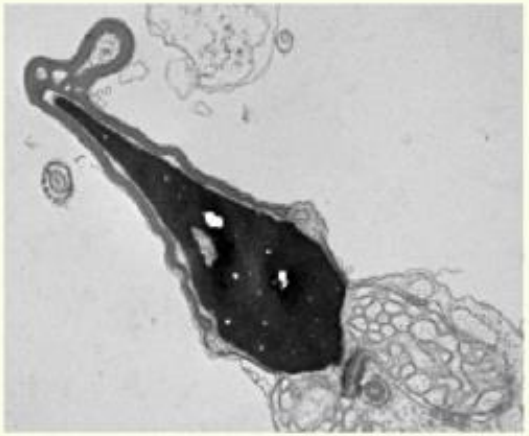
The CFTR protein consists of two transmembrane domains (TMD) (each containing six spans of alpha helices), two nucleotide-binding domains (NBD1 and NBD2) and a central regulatory domain (R-domain). CFTR activity is regulated by PKA-phosphorylation of the R-domain and ATP binding and hydrolysis at the two NBDs.

Direct interaction of SLC26 with CFTR is mediated by the STAS domain and the regulatory (R) domain of CFTR. Indirect interaction of the proteins occurs through binding of both SLC26s and CFTR to common PDZ motif-containing scaffold proteins.

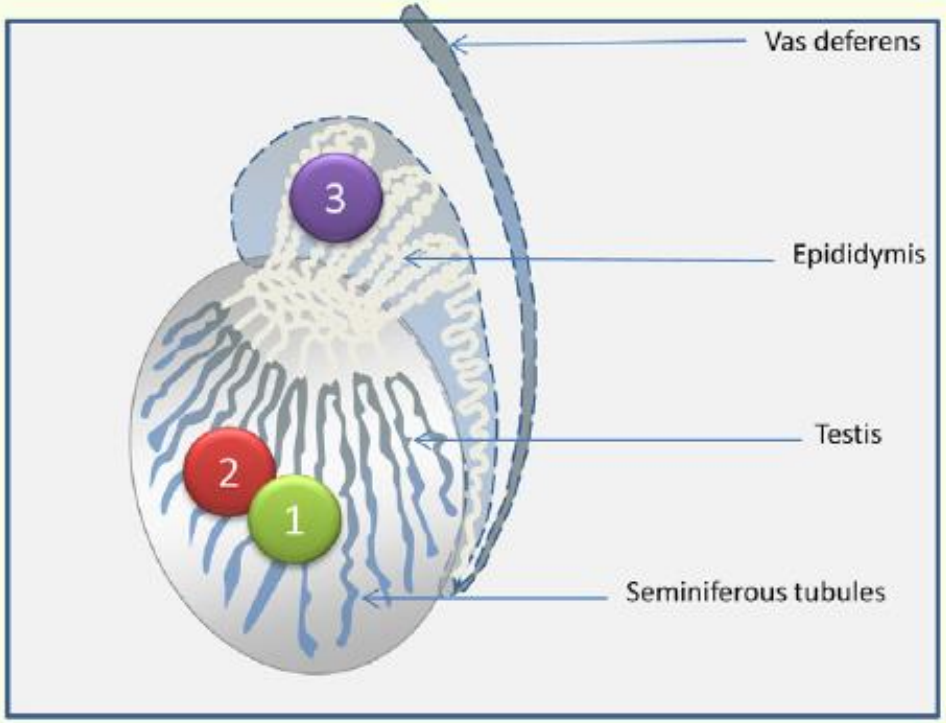
Acrosomal membrane of spermatozoa after its maturation in normal and CF microenvironment



Normal microenvironment



CF microenvironment



Spermatogenesis

Spermiogenesis

Epididymal maturation

Spermatozoon

Tight junctions
Electrolyte and fluid transport
Fatty acid metabolism

CREB
Protamine 2
Electrolyte and fluid transport
Fatty acid metabolism

Tight junctions,
Electrolyte and fluid transport
Fatty acid metabolism

↘ HCO_3^- *transport*
↘ *cAMP production*

Pre-meiotic and meiotic steps

Post-meiotic steps

Spermatozoa production and maturation

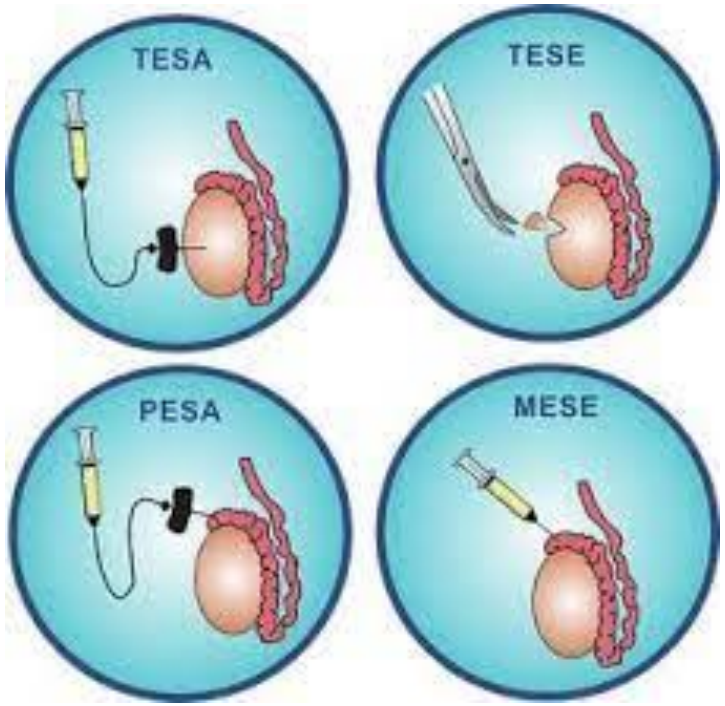
Recupero degli spermatozoi

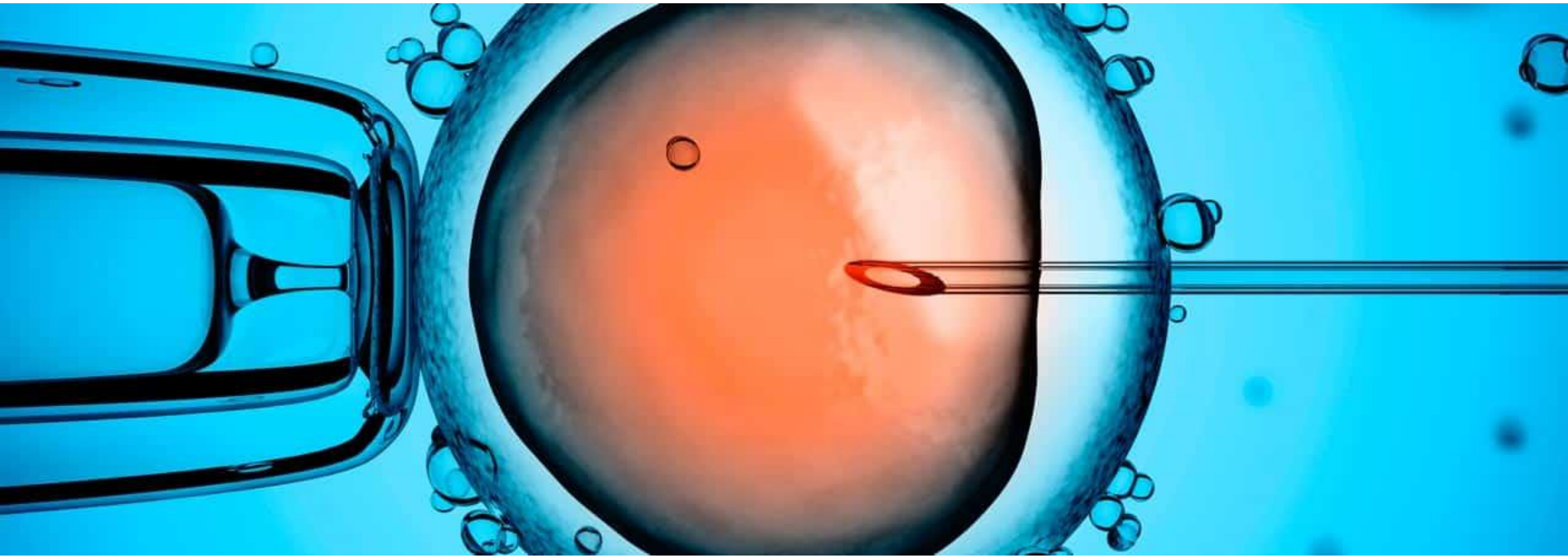
OA

- **MESA:** Microsurgical Epididymal Sperm Aspiration
- **PESA:** Percutaneous Epididymal Sperm Aspiration
- **TESE:** Testicular Sperm Extraction
- **TESA:** Testicular Sperm Aspiration
- **ESE:** Epididymal Sperm Extraction
- **MVSA:** Microsurgical Vasal Sperm Aspiration
- **DISTA:** Distal Seminal Tract Aspiration

NOA

- **TESE:** TEsticular Sperm Extraction
- **MicroTESE:** Microsurgical TEsticular Sperm Extraction
- **(TESA):** TEsticular Sperm Aspiration





Fertility and Sterility® Vol. 101, No. 5, May 2014

Association of cystic fibrosis transmembrane-conductance regulator gene mutation with negative outcome of intracytoplasmic sperm injection pregnancy in cases of congenital bilateral absence of vas deferens

Shaoming Lu, M.D.,[a] Yanyi Cui, M.Sc.,[b] Xiao Li, M.B.,[a] Haobo Zhang, M.D.,[a] Jiaolong Liu, M.B.,[a] Bin Kong, M.B.,[a] Feifei Cai, M.B.,[a] and Zi-Jiang Chen, M.D., Ph.D.[a]

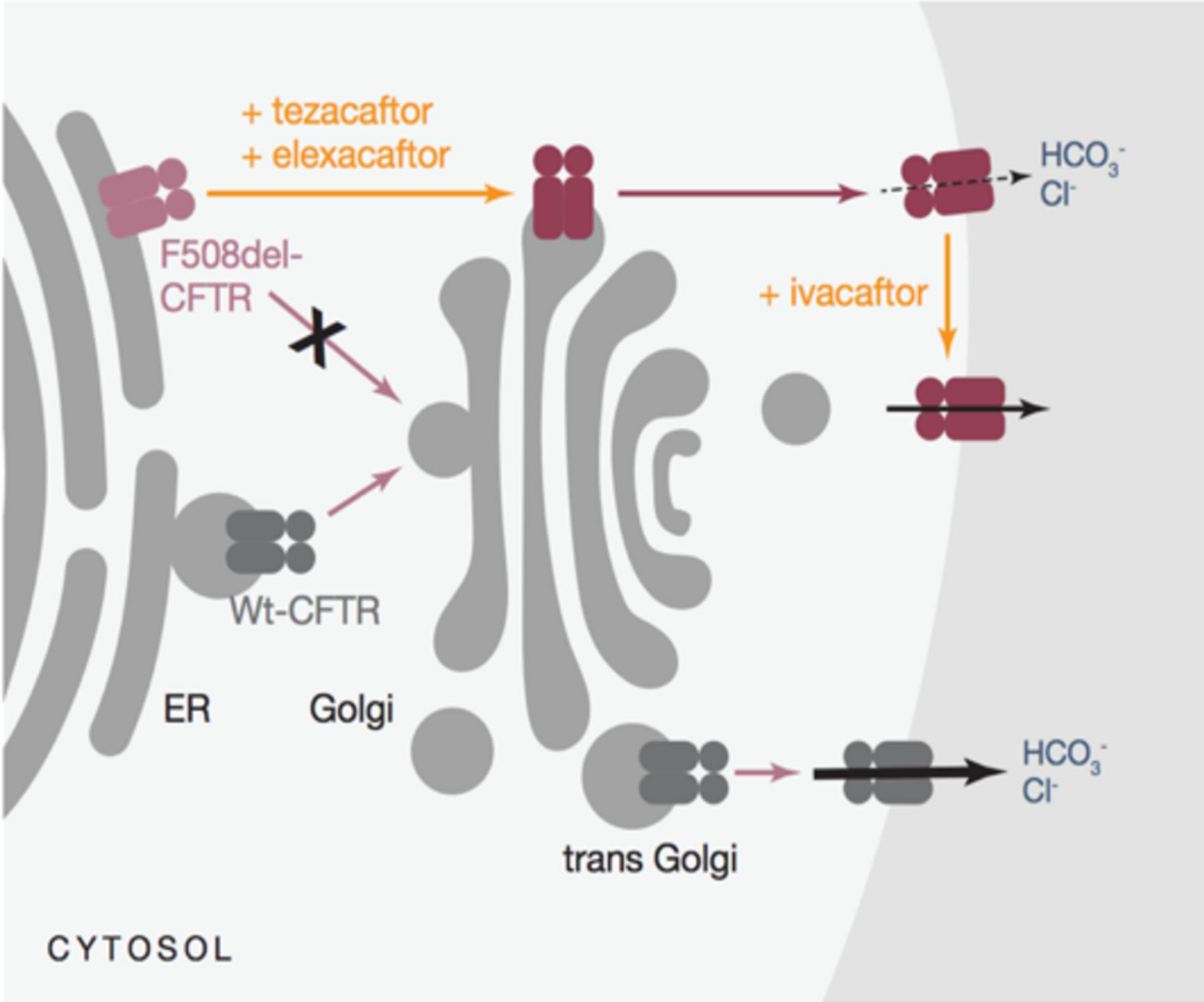
TABLE 1

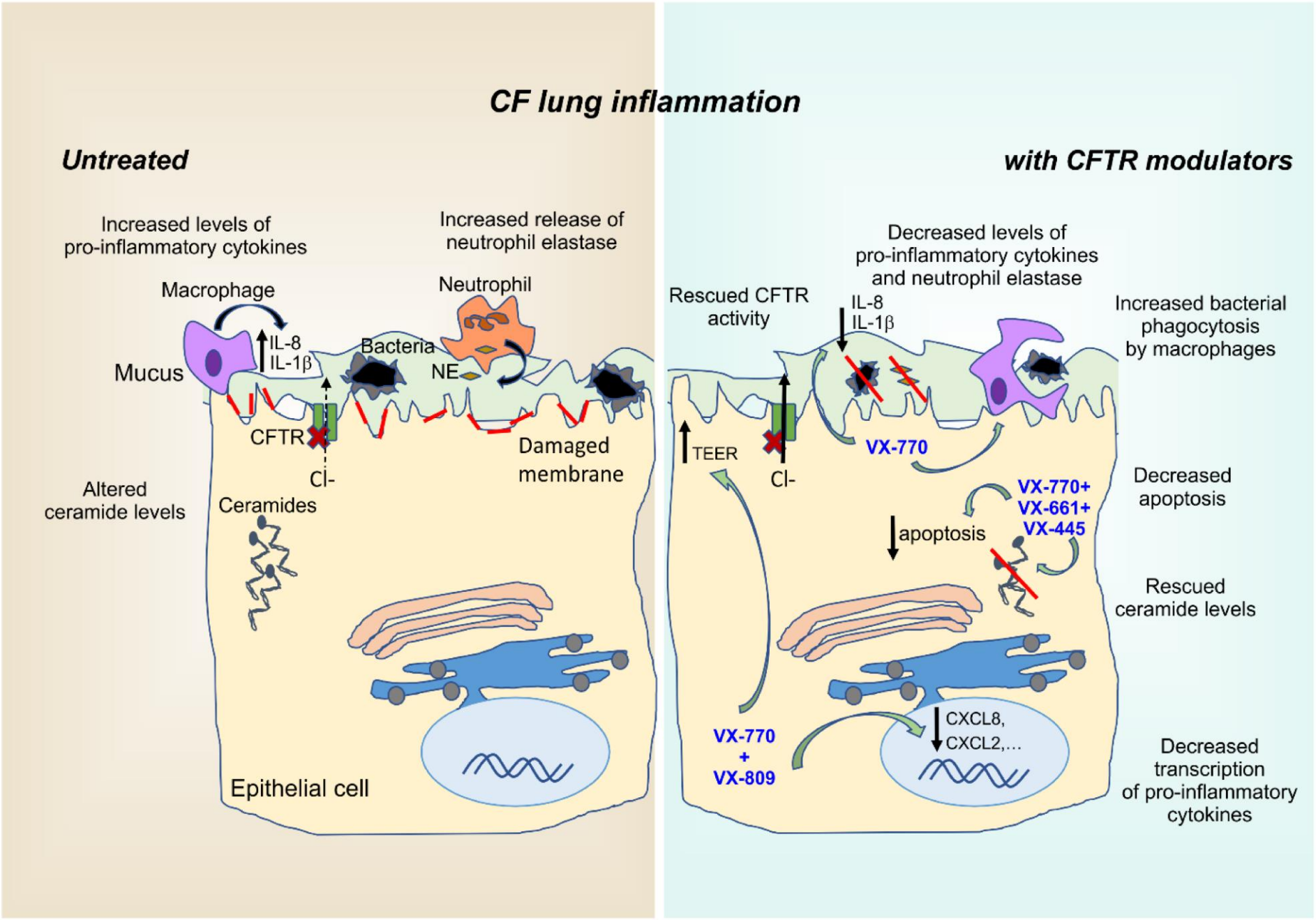
Comparison of ICSI outcomes in men with CBAVD and in men with other types of OA.

	CBAVD	Acquired OA	*P* value
No. of patients	531	883	
Age of women (mean ± SD)	28.80 ± 4.86	29.43 ± 5.02	.123
Age of men (mean ± SD)	29.96 ± 4.84	29.42 ± 5.02	.187
No. of MII oocytes (mean ± SD)	12.90 ± 7.02	12.76 ± 6.79	.705
No. of 2-pronuclei oocytes (mean ± SD)	8.87 ± 5.15	8.56 ± 4.96	.246
No. of good embryos (mean ± SD)	5.11 ± 4.04	4.54 ± 3.55	.056
No. of transferred embryos (mean ± SD)	2.04 ± 0.86	2.02 ± 0.80	.584
Rate of fertilization, %	70.1	68.2	.066
Rate of good embryos, %	51.1	52.1	.485
Rate of implantation, %	40.2	36.3	.195
Rate of clinical pregnancy per embryo transferred, %	49.7 (264/531)	48.8 (431/883)	.783
Rate of live birth per embryo transferred, %	35.0 (186/531)	41.4 (366/883)	.019*
Rate of miscarriage/stillbirth per embryo transferred, %	11.9 (63/531)	6.1 (54/883)	.000*
Rate of ectopic pregnancy per embryo transferred, %	2.8 (15/531)	1.2 (11/883)	.053

* $P < .05$, statistically significant.

Lu. CFTR mutation in pregnancy with CBAVD. Fertil Steril 2014.


+ tezacaftor
+ elexacaftor
F508del-
CFTR
Wt-CFTR
ER
Golgi
trans Golgi
+ ivacaftor
HCO_3^-
Cl^-
HCO_3^-
Cl^-
CYTOSOL


CF lung inflammation
Untreated
with CFTR modulators
Increased levels of pro-inflammatory cytokines
Increased release of neutrophil elastase
Macrophage
Neutrophil
IL-8
IL-1β
Mucus
Bacteria
NE
CFTR
Cl-
Damaged membrane
Altered ceramide levels
Ceramides
Epithelial cell
Decreased levels of pro-inflammatory cytokines and neutrophil elastase
Rescued CFTR activity
IL-8
IL-1β
Increased bacterial phagocytosis by macrophages
TEER
Cl-
VX-770
VX-770+
VX-661+
VX-445
apoptosis
Decreased apoptosis
Rescued ceramide levels
VX-770
+
VX-809
CXCL8,
CXCL2,...
Decreased transcription of pro-inflammatory cytokines