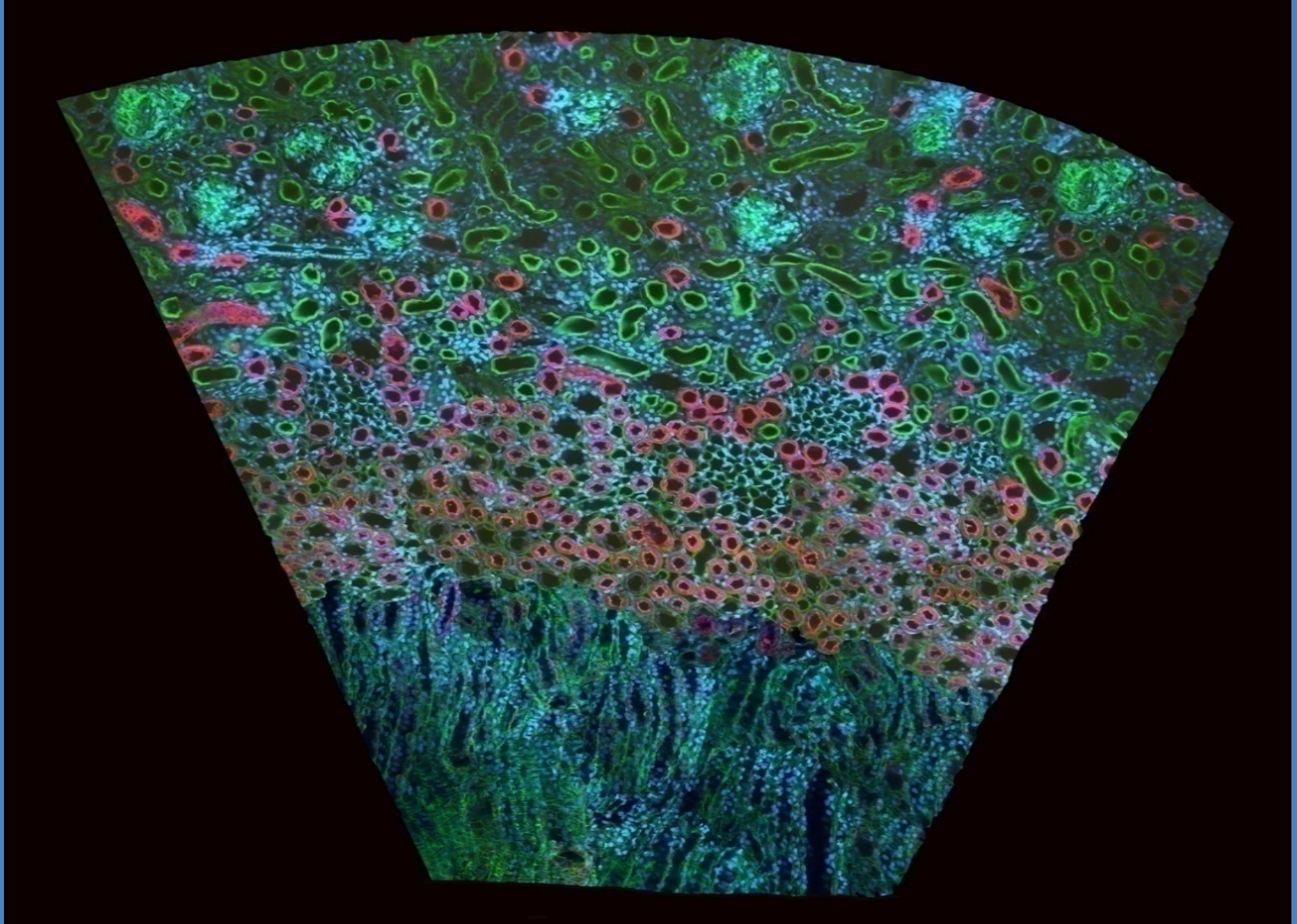


# New insights into sepsis-induced renal injury



# Takashi Hato

Rabih Kalakeche

Tarek El-Achkar

Ruben Sandoval

Kenneth Dunn

George Rhodes

Zoya Plotkin

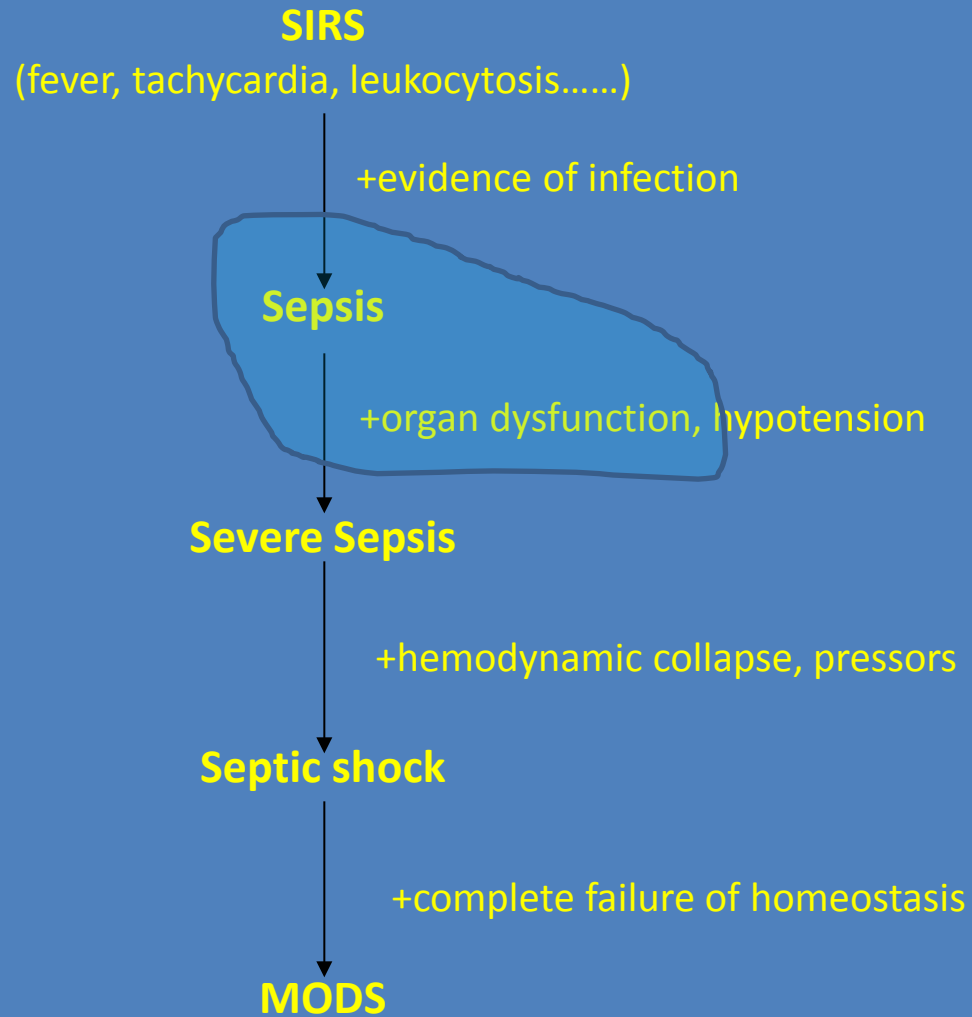
# Fact

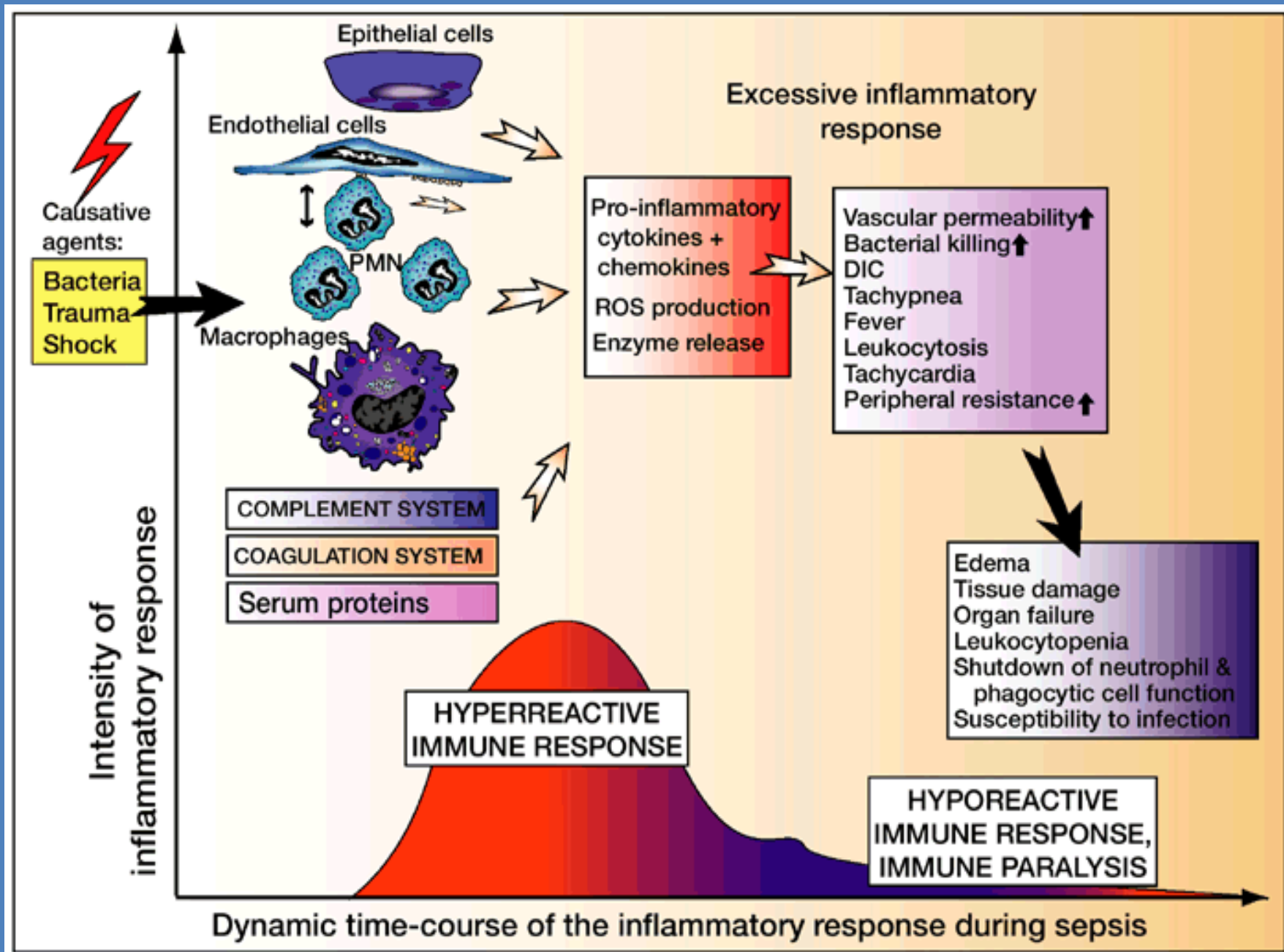
Sepsis and septic shock remain the most important cause of Acute Kidney Injury (AKI) in critically ill patients, and account for more than 50% of cases of AKI in the ICU

# What is Sepsis?

- Syndrome caused by the interaction of a pathogen with the host immune system
- Dynamic sets of events characterized by a maladaptive response of the immune system.

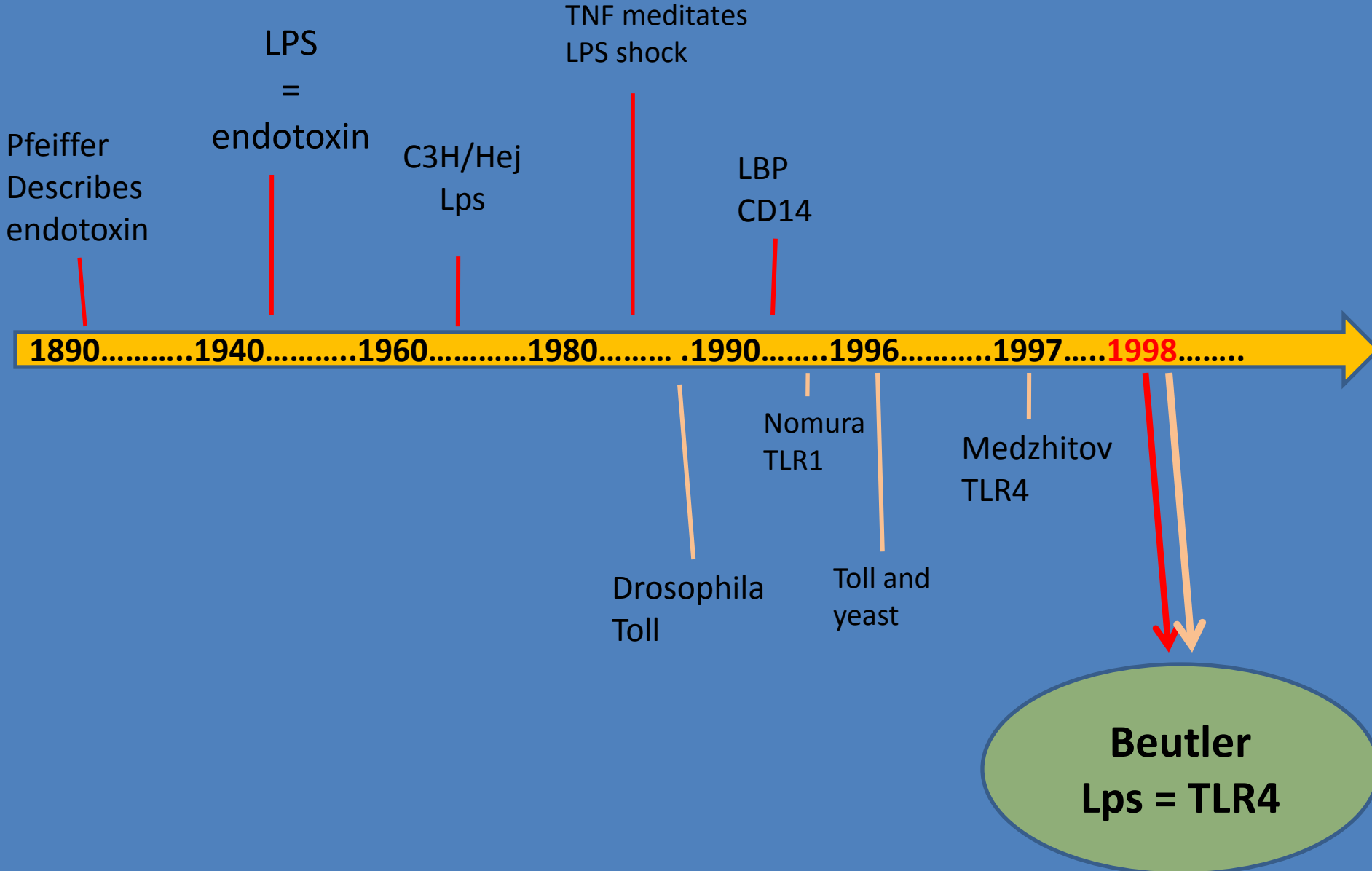
# Clinical Definition of Sepsis





# Complexity of sepsis literature

1. What animal sepsis model is being used
2. Studies in same or different species
3. Age of animals
4. Single bacteria vs Polymicrobial
5. Are the rates of bacterial release comparable
6. What bacterial strain/endotoxin is being used
7. Antibiotics
8. Are the doses of endotoxin comparable
9. What is the end-point: organ damage vs mortality
10. Systemic vs Local TLRs



Pfeiffer  
Describes  
endotoxin

LPS  
=  
endotoxin

C3H/Hej  
Lps

TNF meditates  
LPS shock

LBP  
CD14

1890.....1940.....1960.....1980.....1990.....1996.....1997.....1998.....

Drosophila  
Toll

Nomura  
TLR1

Toll and  
yeast

Medzhitov  
TLR4

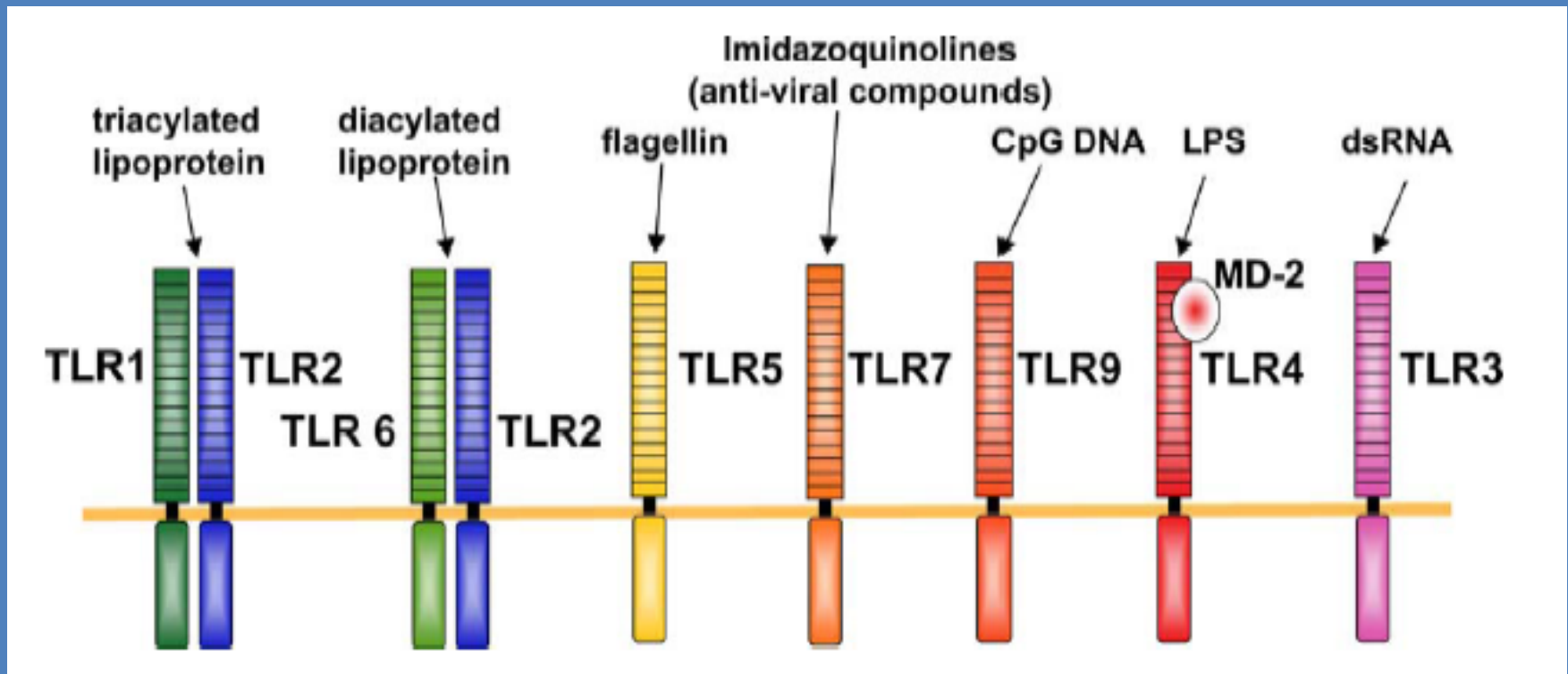
**Beutler  
Lps = TLR4**



# C3H/HeJ Mouse and LPS

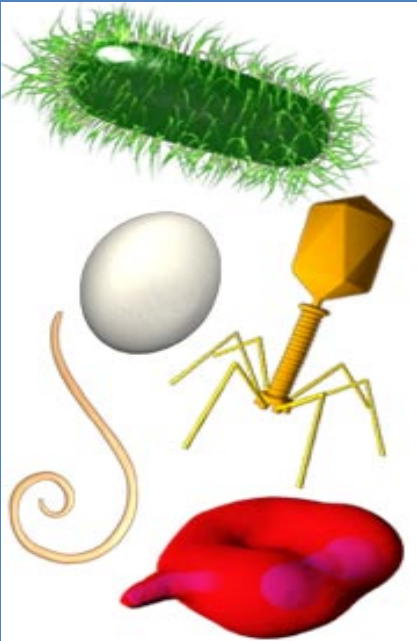
- C3H/HeJ mutant phenotype arose through spontaneous mutation between 1960 and 1968
- C3H/HeJ strain exhibits natural tolerance to otherwise lethal doses of LPS
- Hyporesponsive phenotype under the control of a single locus, *Lps* existing in two allelic forms, *Lps<sup>n</sup>* (responsive) and *Lps<sup>d</sup>* (hyporesponsive)
- *Lps* locus assigned to chromosome 4

# TLRs and their ligands

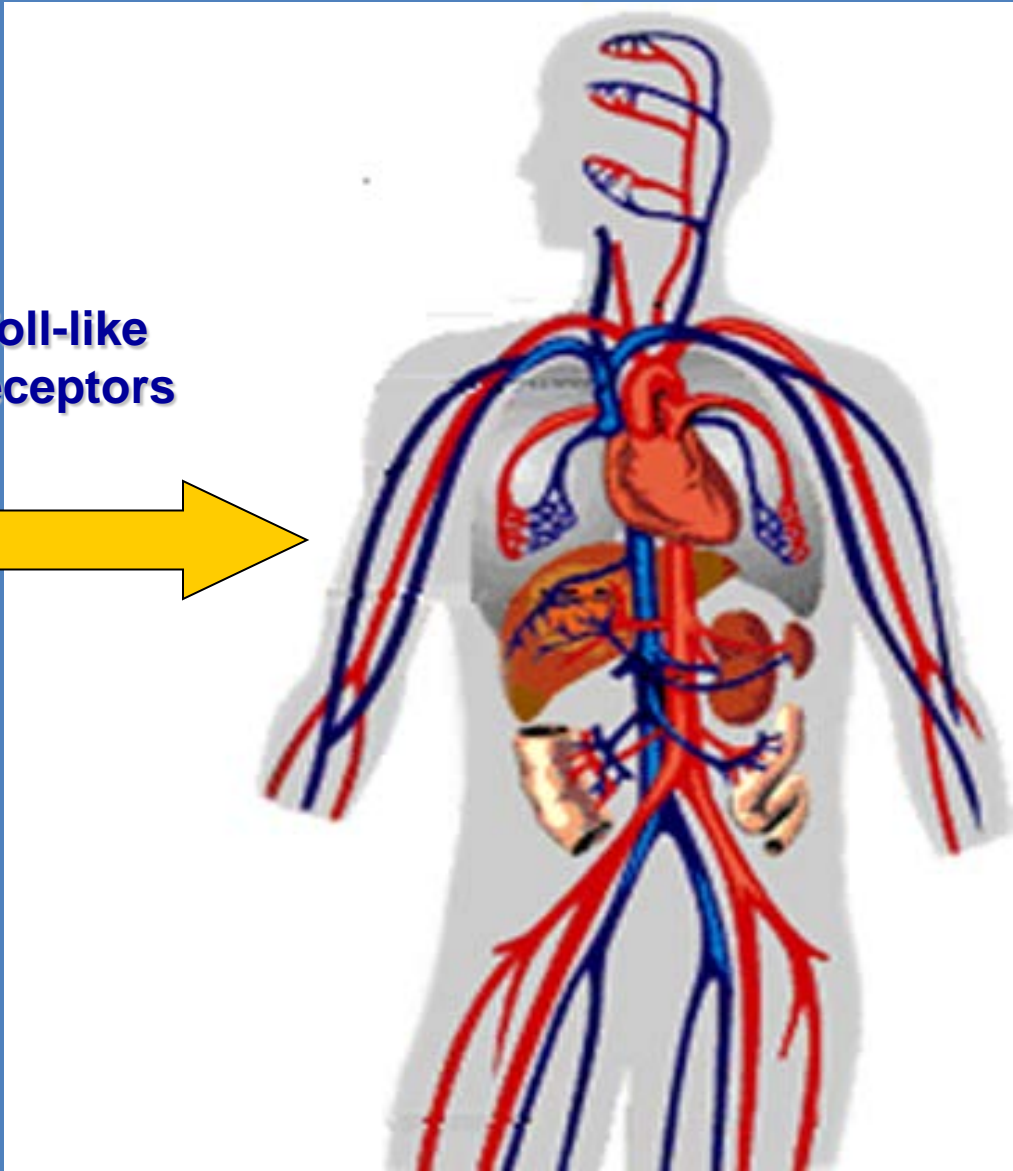
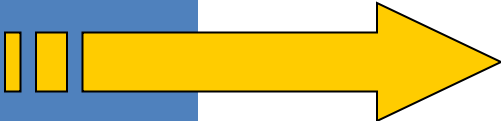


# Organism

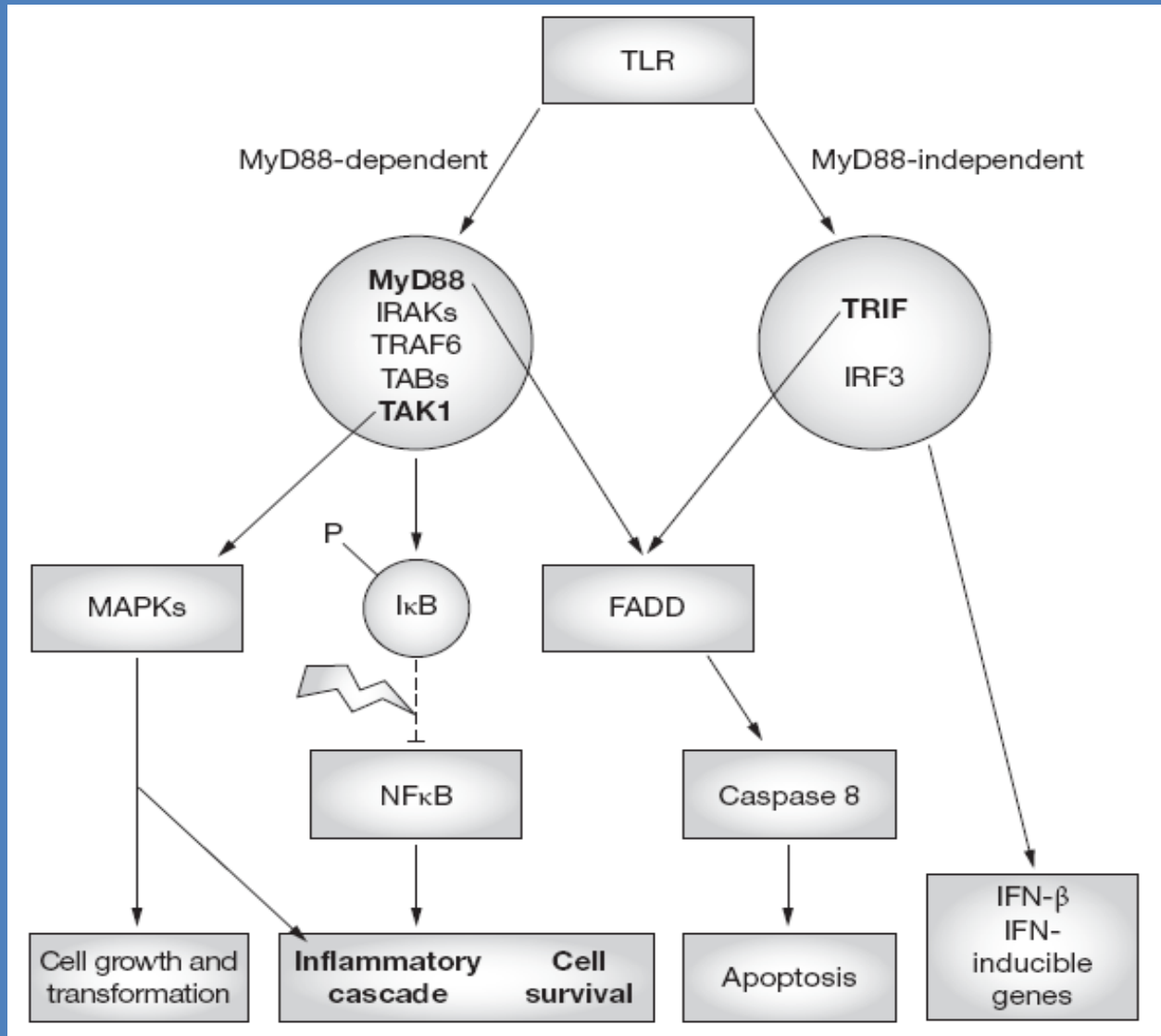
# Pathogens



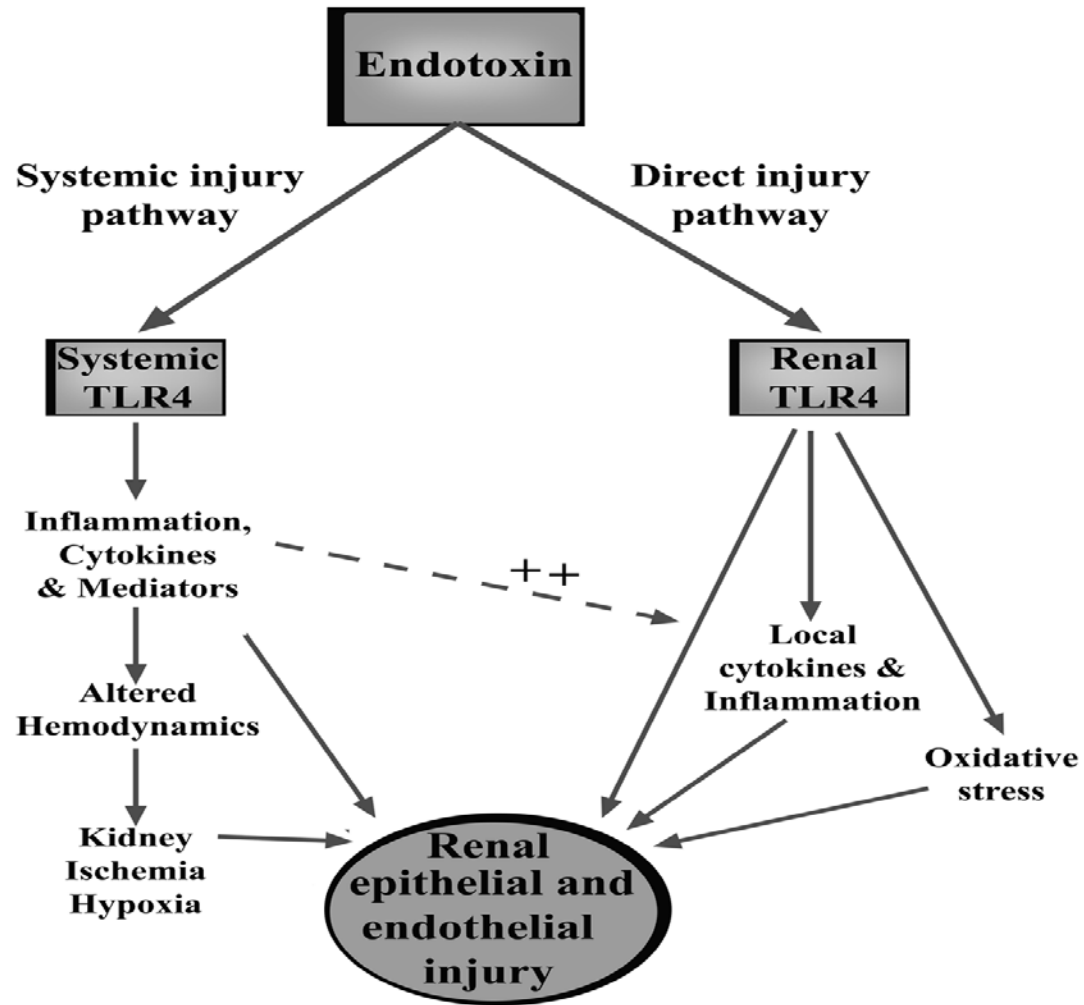
Toll-like  
Receptors

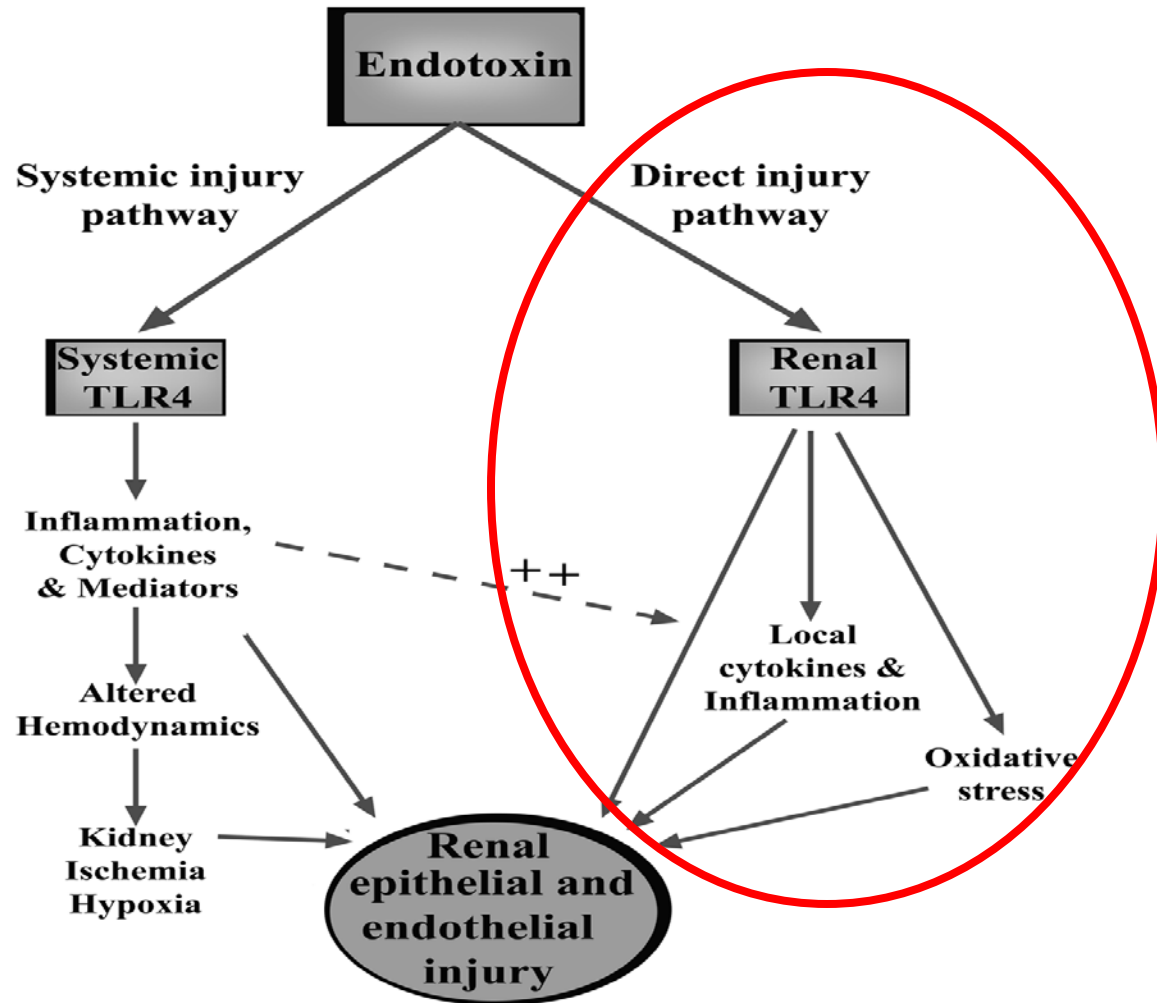


# Branching Nature of TLR Signaling

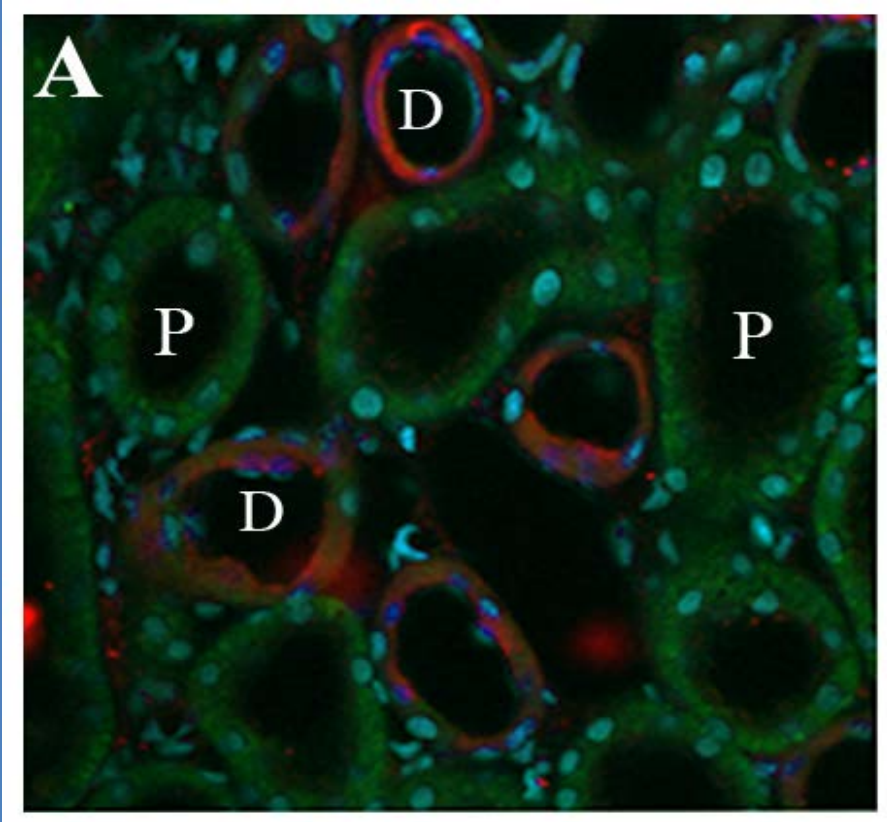


	<i>Advantages</i>	<i>Disadvantages</i>
•Endotoxin infusion or injection	<ul style="list-style-type: none"> <li>•Simple and inexpensive</li> <li>•Well standardized dose</li> </ul>	<ul style="list-style-type: none"> <li>•vasoconstriction</li> <li>•ARF requires high doses</li> <li>•high mortality, Short duration.</li> <li>•variable response</li> </ul>
•Bacterial infusion	<ul style="list-style-type: none"> <li>• Systemic hemodynamics like human.</li> <li>• Bacterial dose standardized</li> </ul>	<ul style="list-style-type: none"> <li>•No MODS.</li> <li>• Expensive and hard in large animals.</li> <li>• Standard supporting measures, often lacking.</li> </ul>
•Intraperitoneal infusion of bacteria	<ul style="list-style-type: none"> <li>•Simple and inexpensive.</li> <li>•Rapid onset</li> <li>•reproduces aspects of sepsis in humans</li> <li>•control over the dose of bacteria.</li> </ul>	<ul style="list-style-type: none"> <li>•Too severe in large animals.</li> <li>•ATN not produced clinically or pathologically.</li> </ul>
•Cecal ligation and perforation	<ul style="list-style-type: none"> <li>• Simple and inexpensive.</li> <li>• Septic shock with MODS</li> </ul>	<ul style="list-style-type: none"> <li>• response variable</li> <li>• human like ATN not reproduced.</li> </ul>

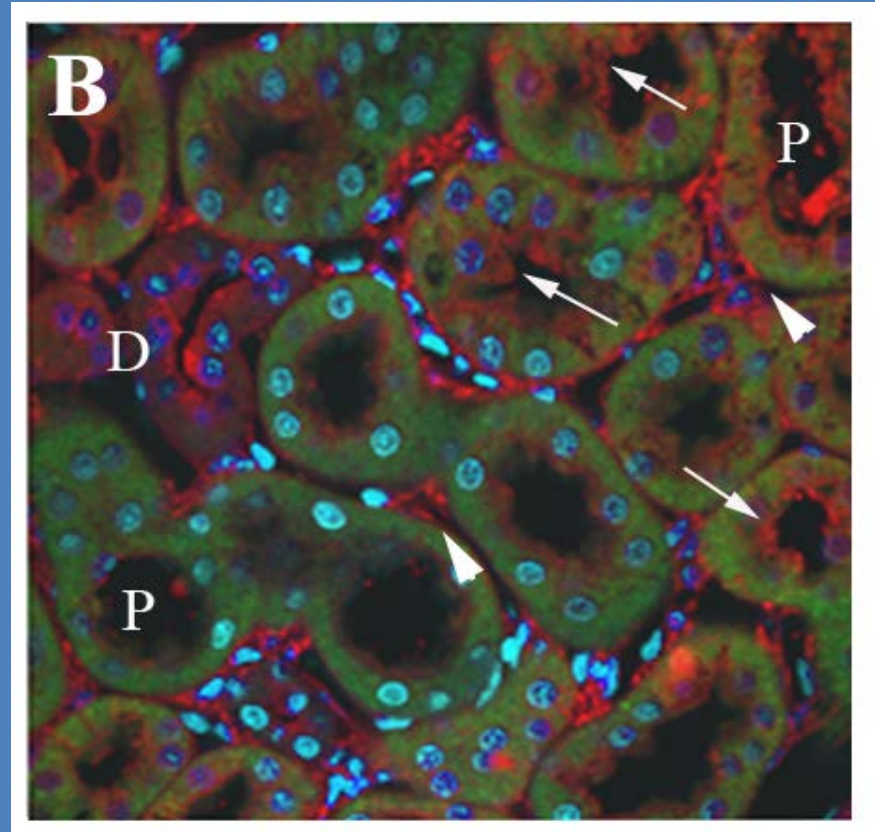




# Localization of TLR4



**Sham**

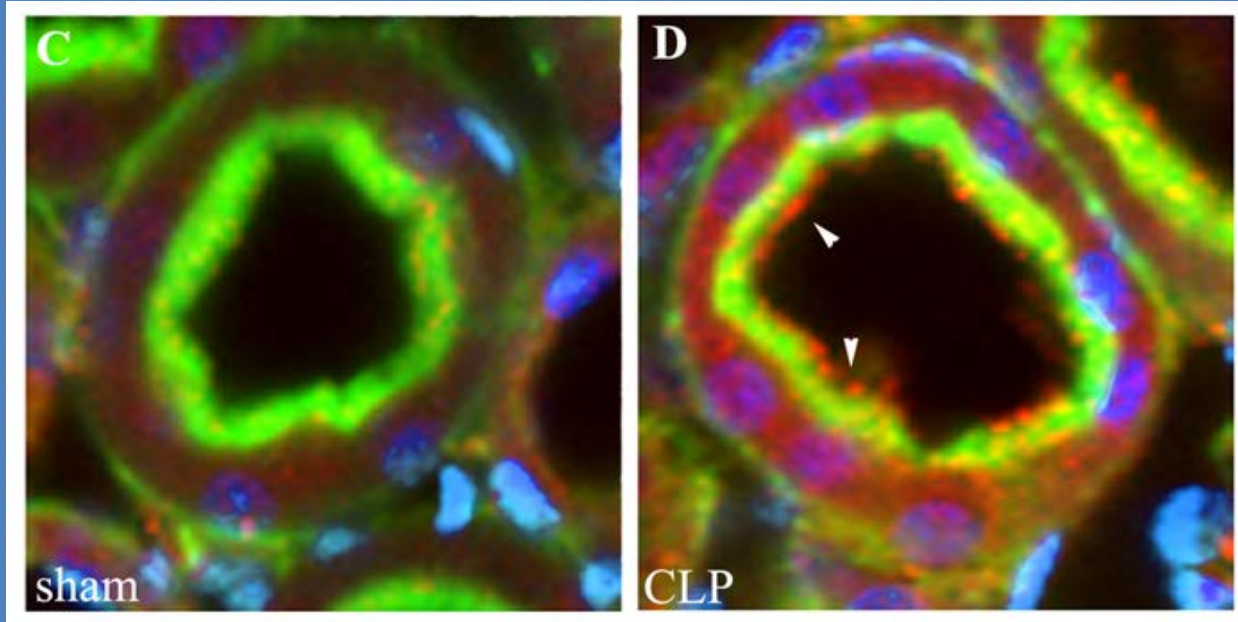


**CLP**

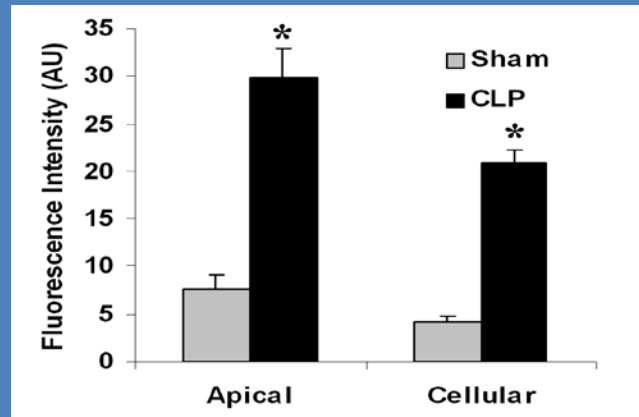


# TLR4 in Proximal Tubules

Sham

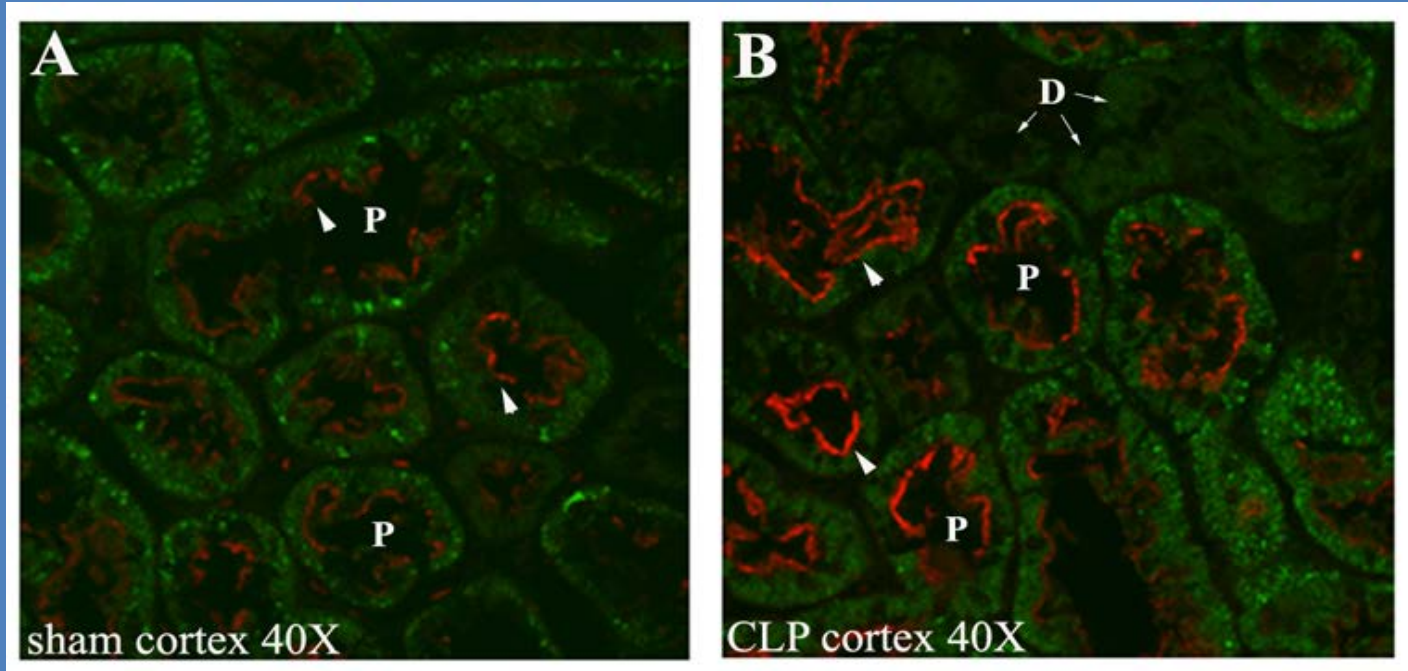


CLP

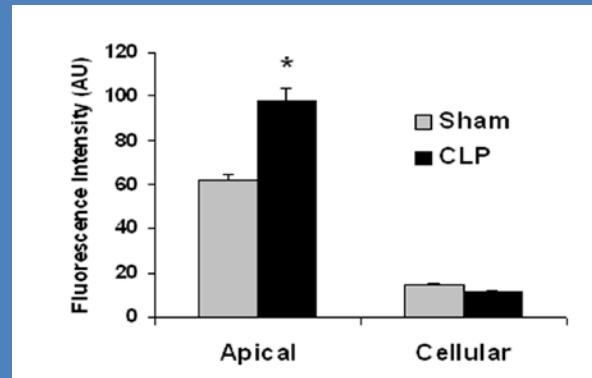


# Localization of CD14

Sham

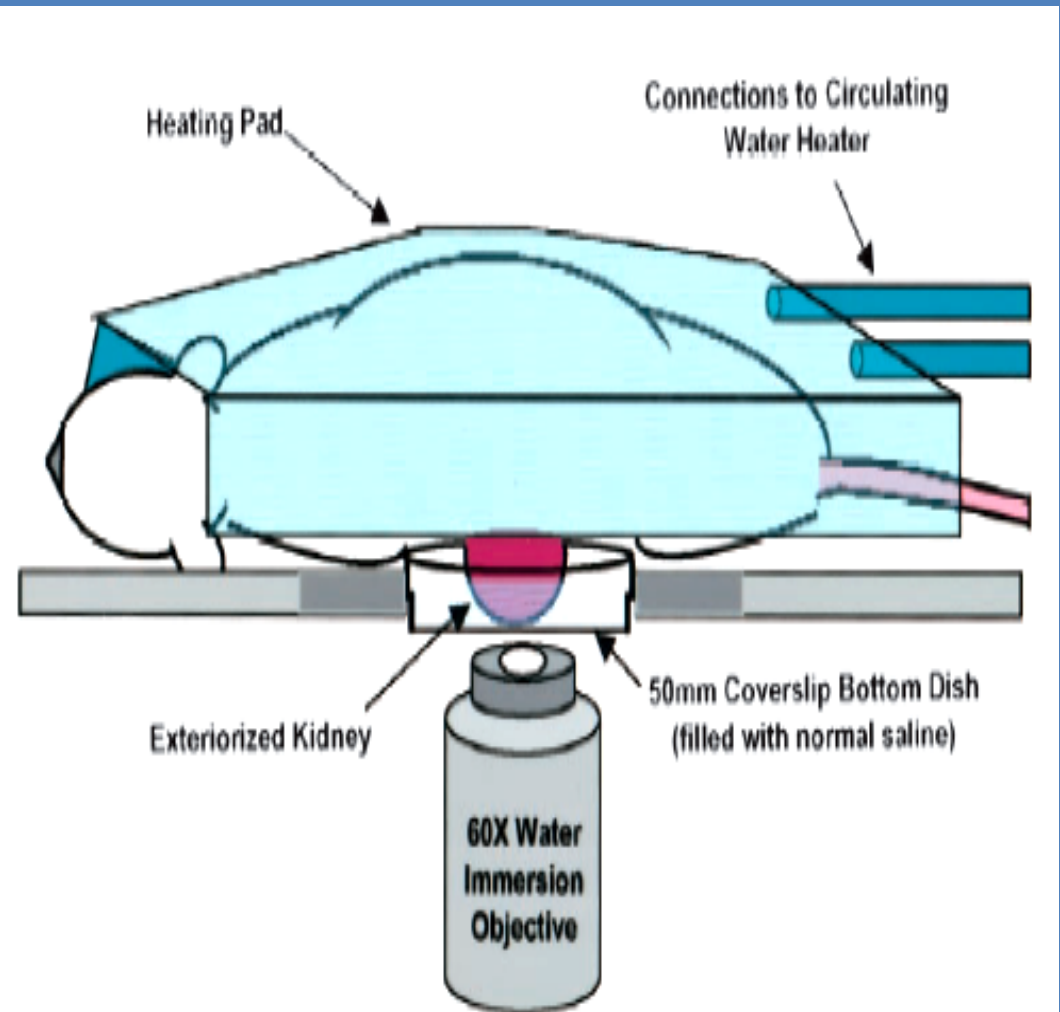


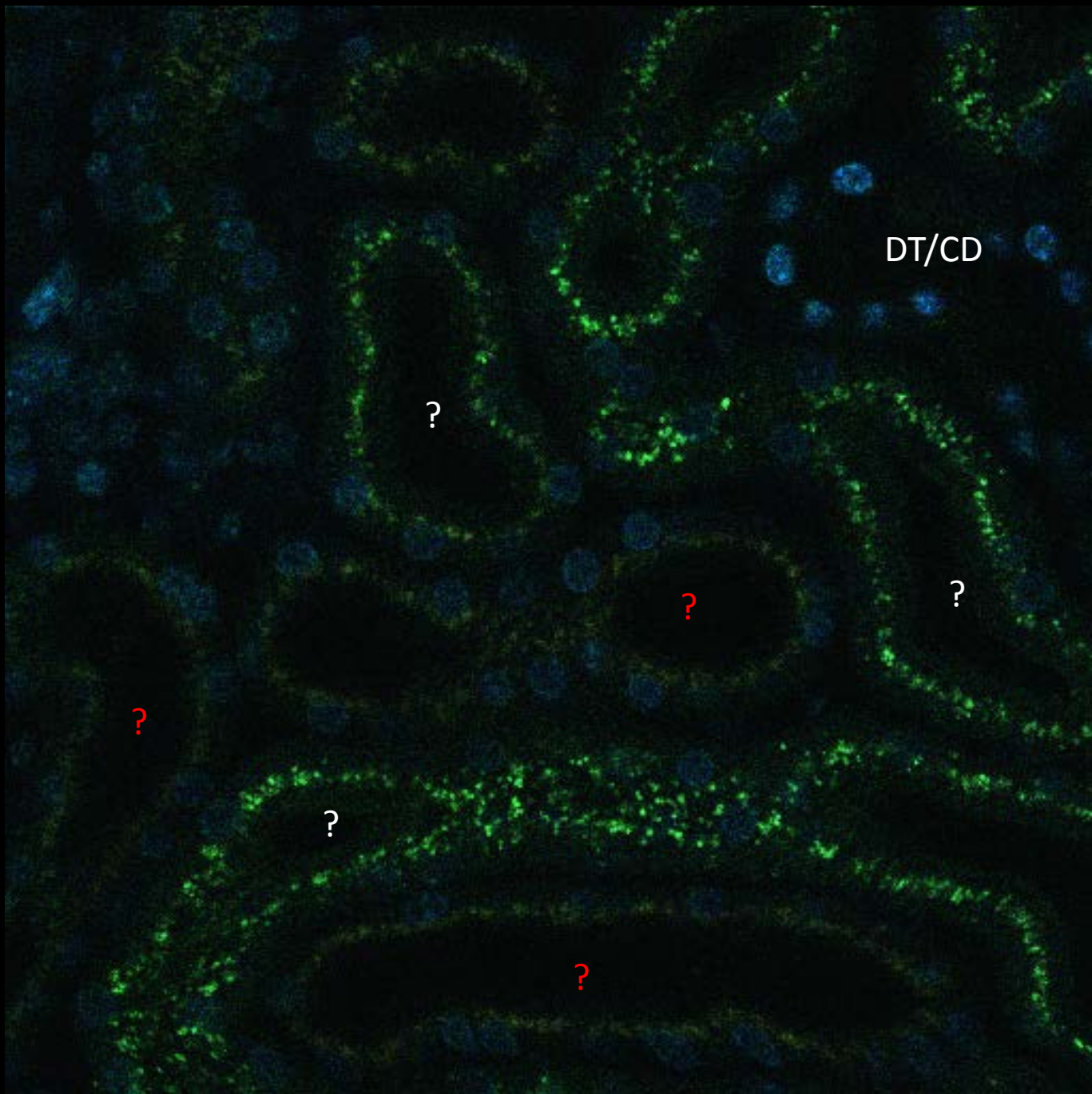
CLP



# Intra-vital 2 photon microscopy

Fluorescent LPS injected as a tracer of endogenous LPS followed by imaging





DT/CD

?

?

?

?

?

?

distal convoluted tubule

proximal convoluted tubule

S2 S1

S3

Bowman's capsule

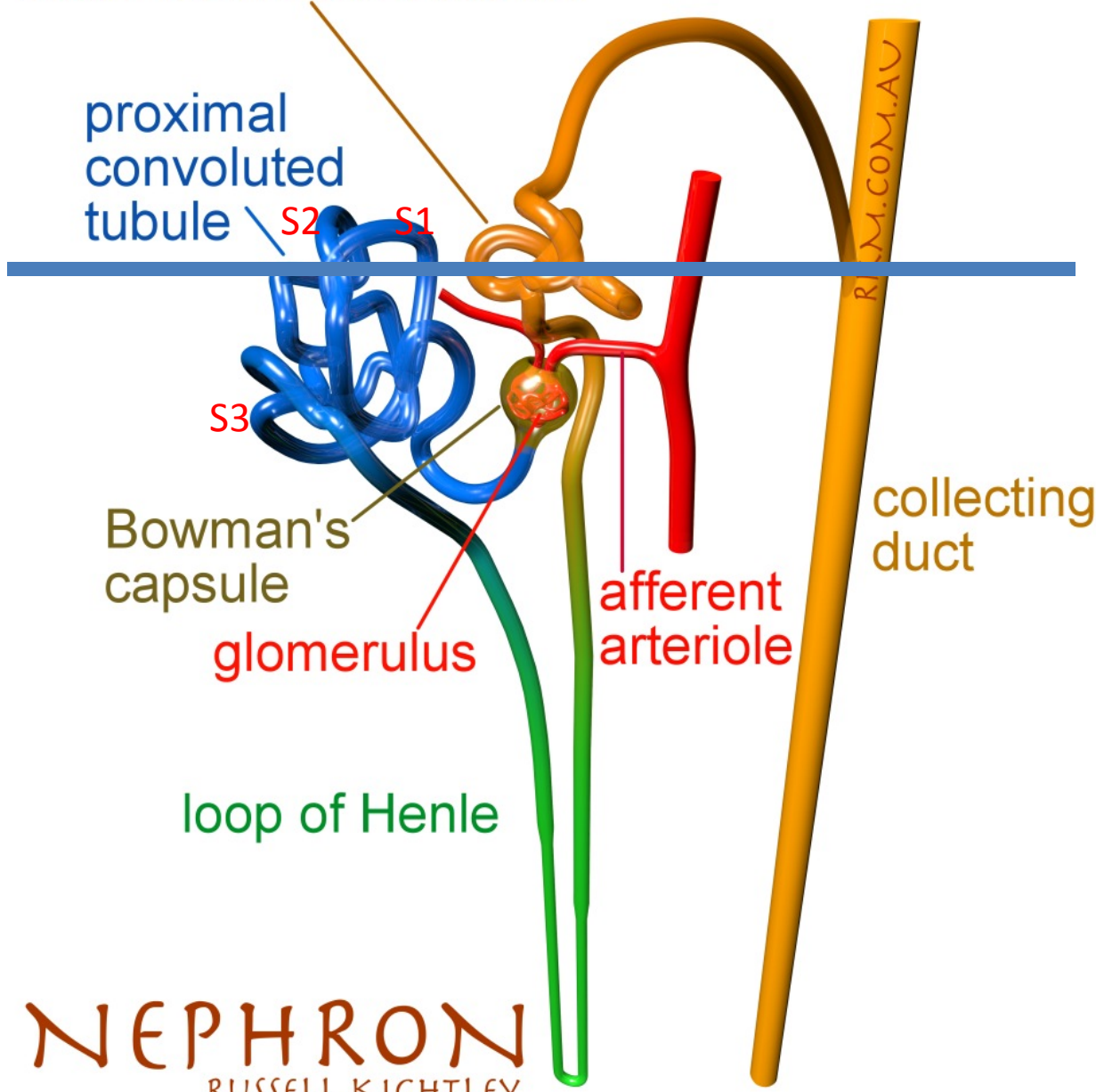
glomerulus

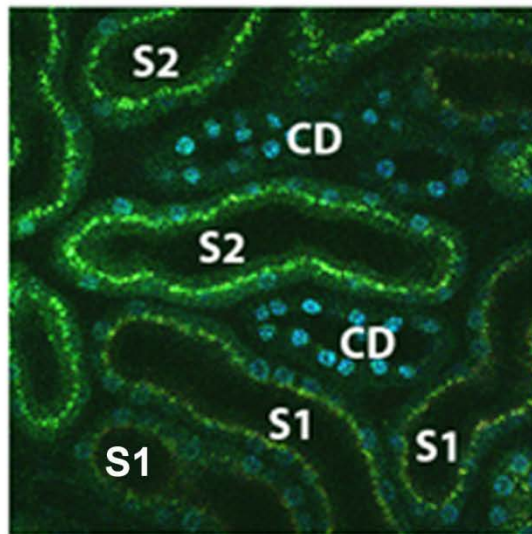
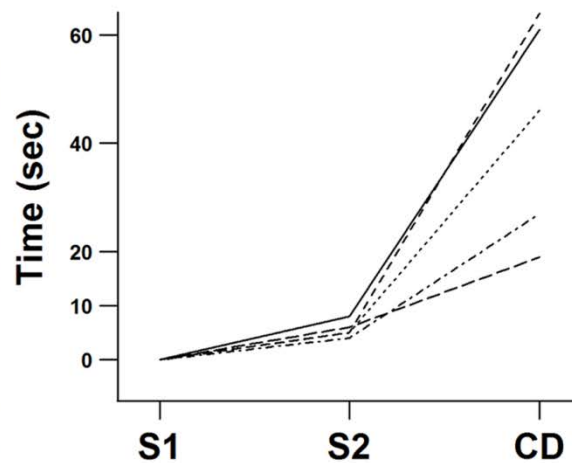
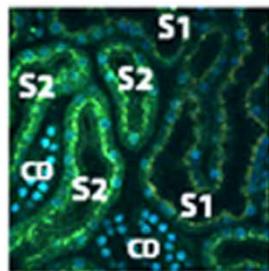
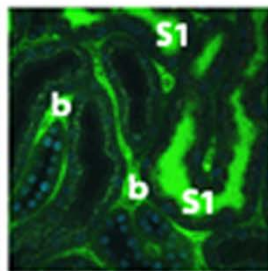
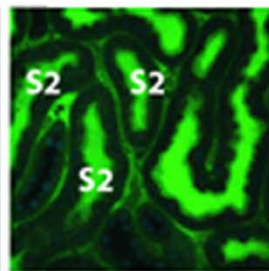
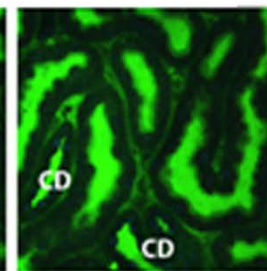
afferent arteriole

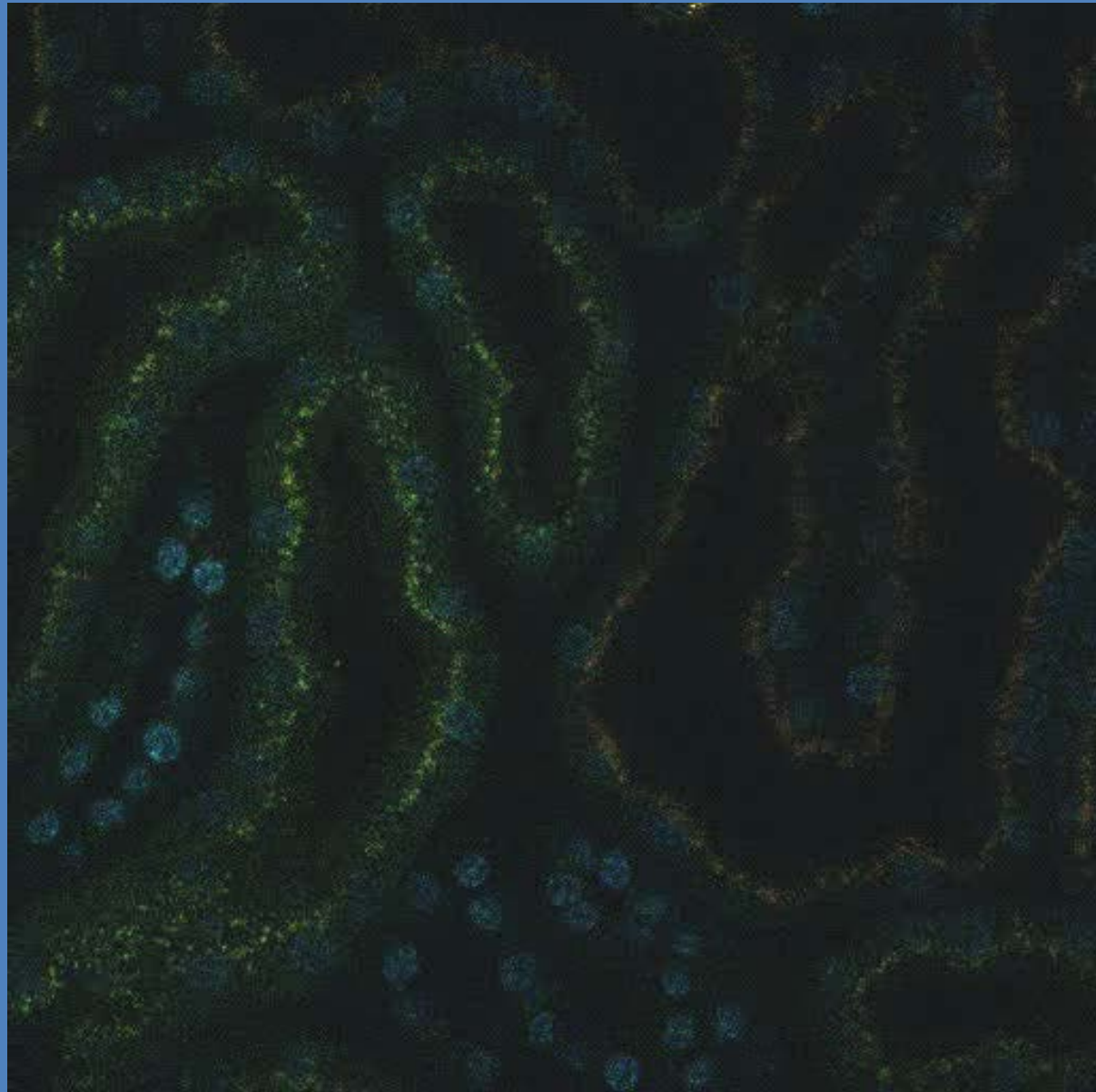
collecting duct

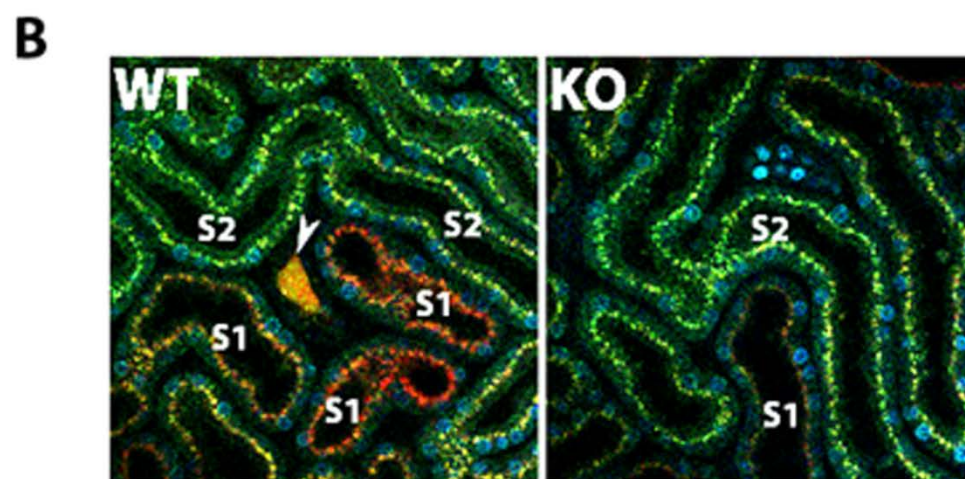
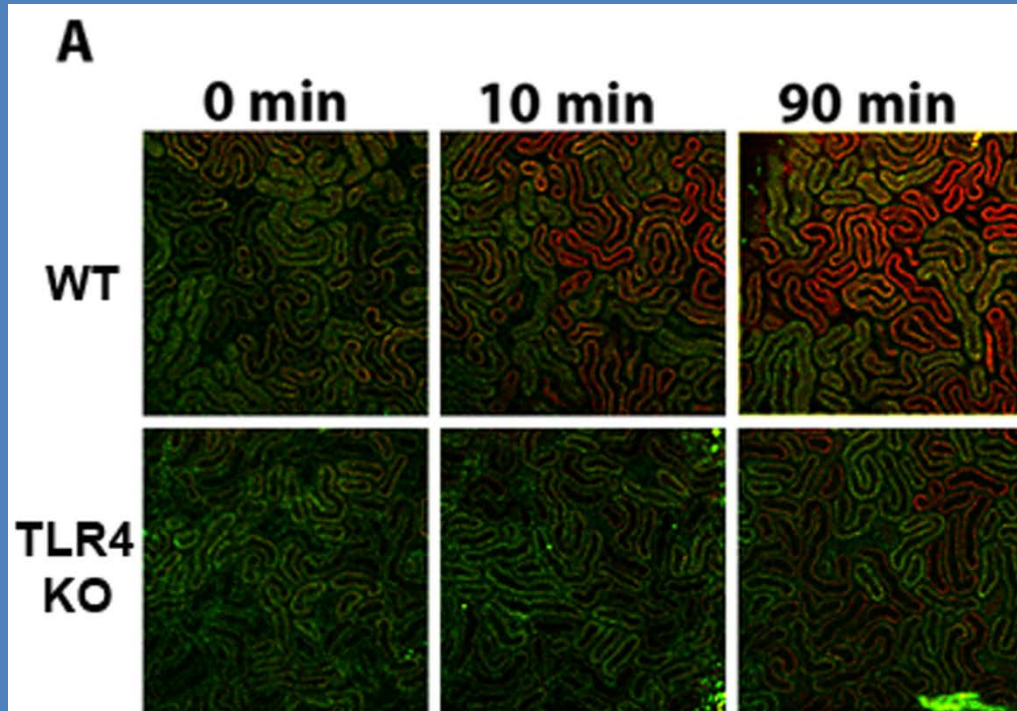
loop of Henle

NEPHRON  
RUSSELL KIGHTLEY

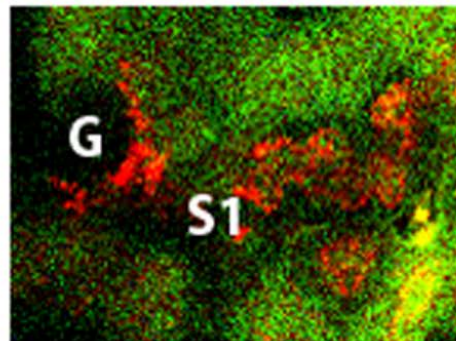
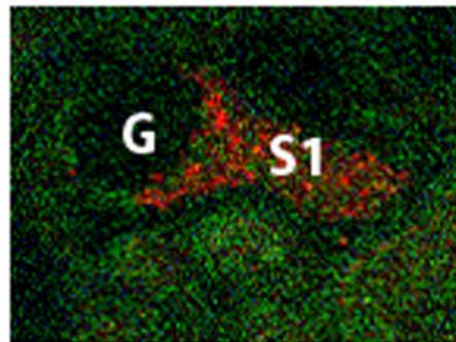
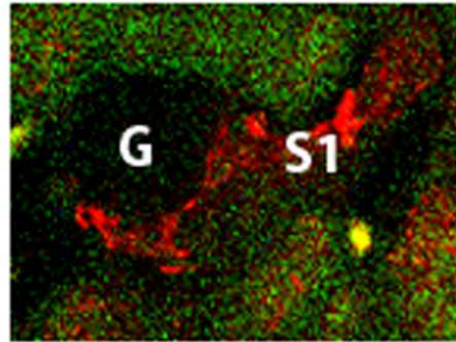
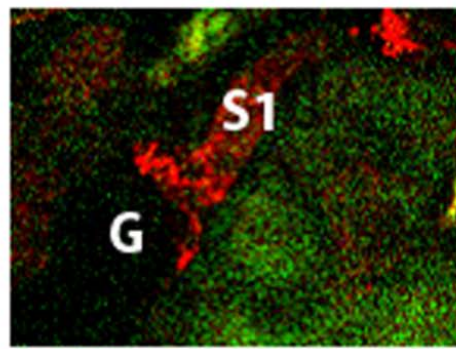


**A****C****B****0 sec****20 sec****25 sec****65 sec**

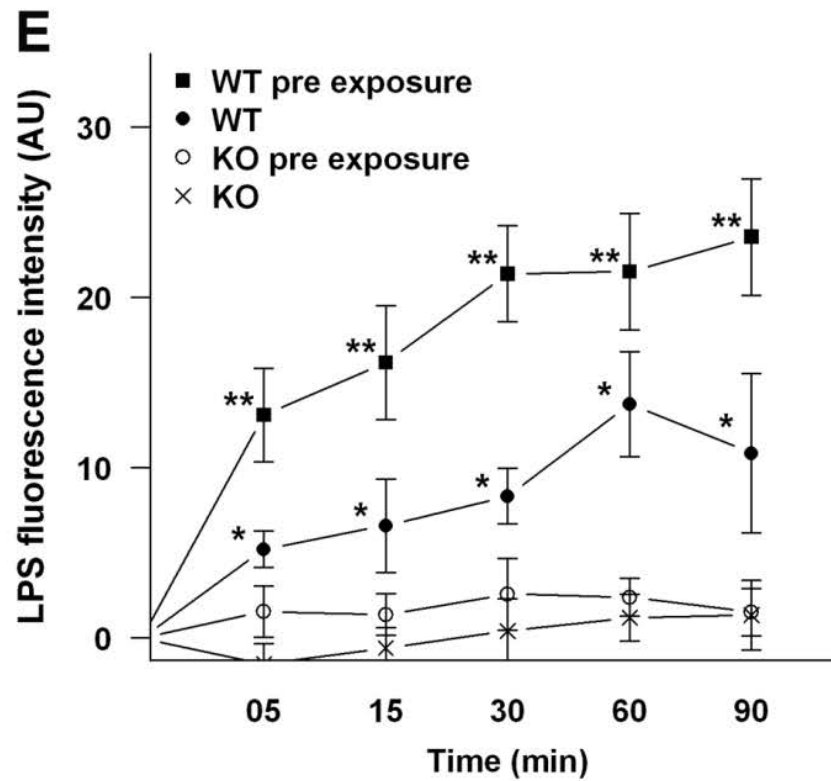
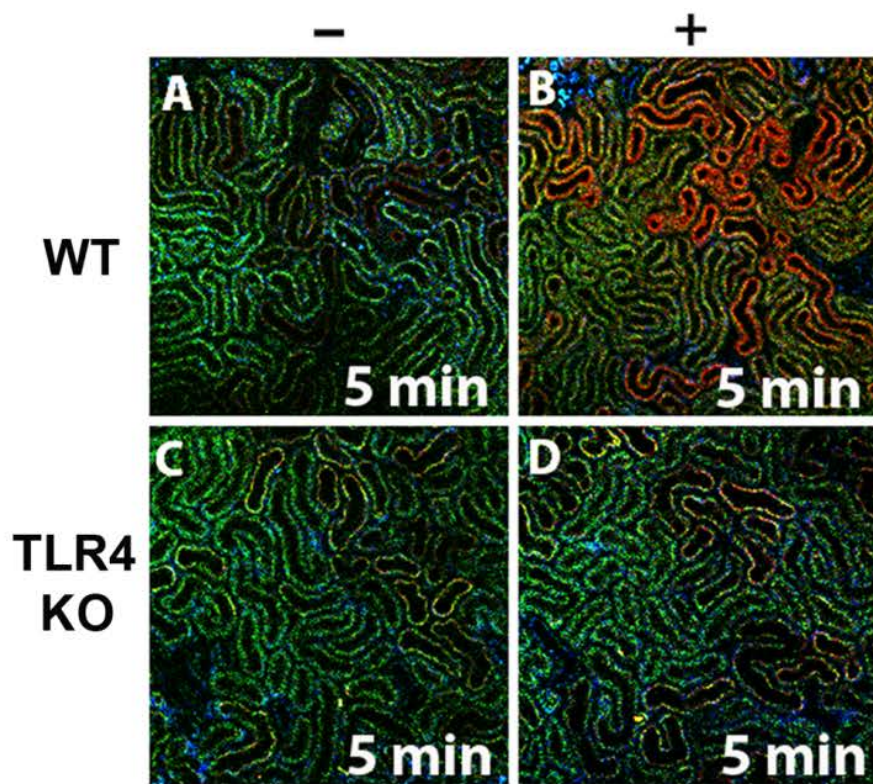




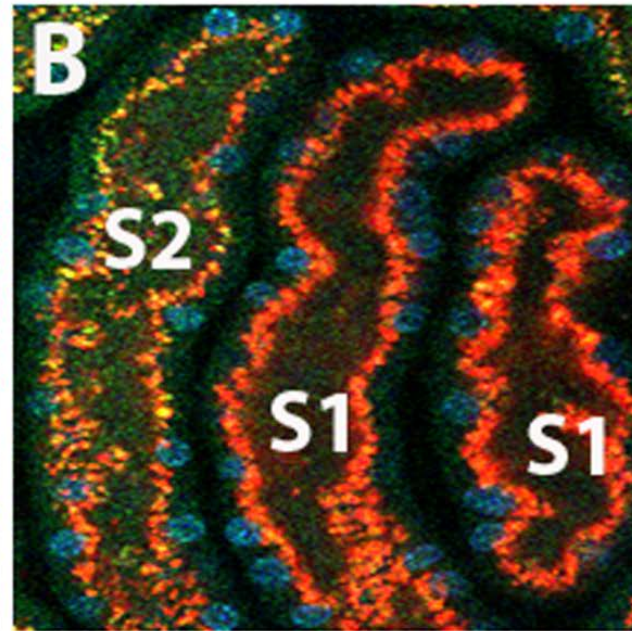
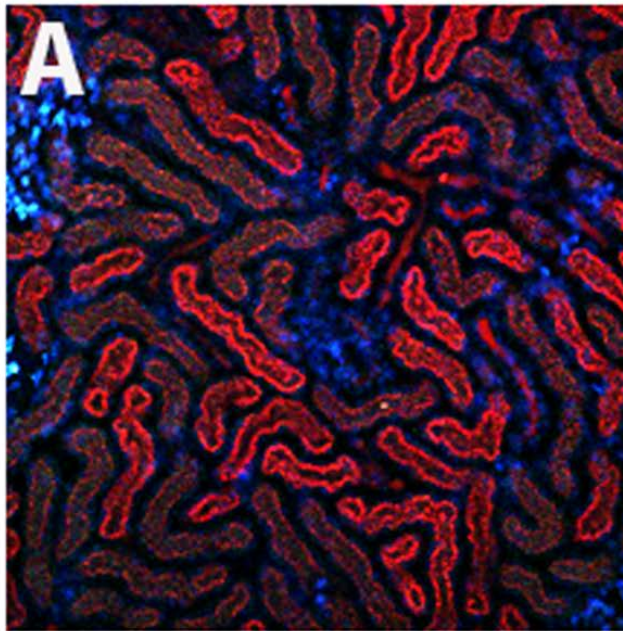




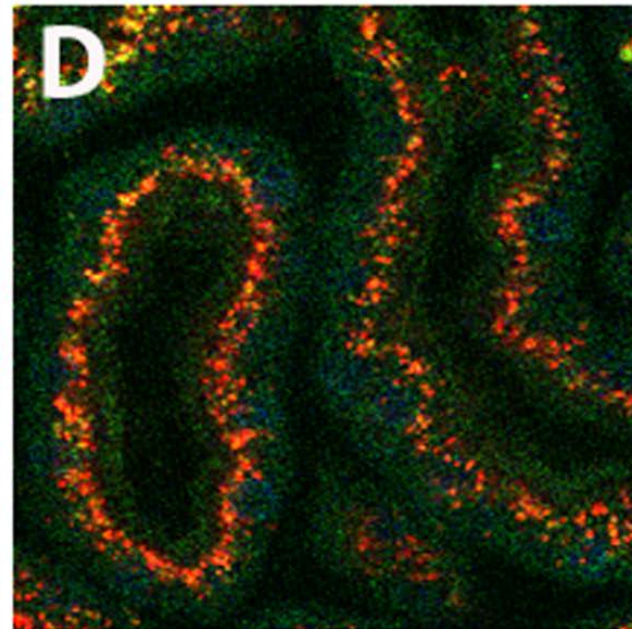
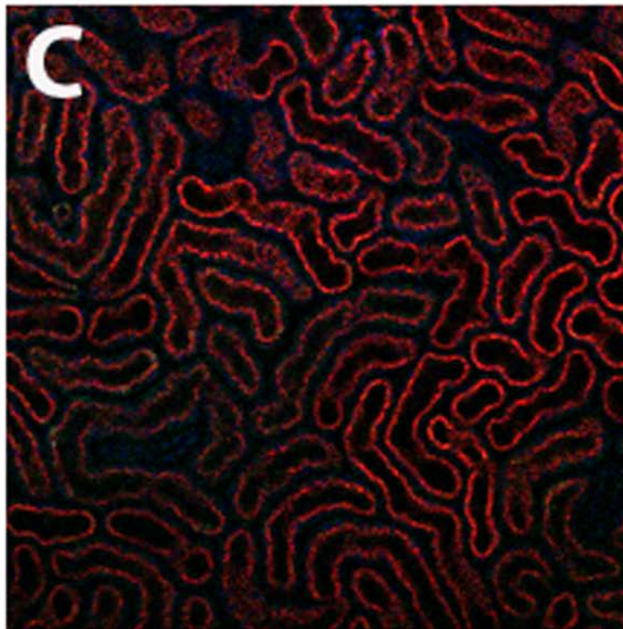
## LPS pre exposure

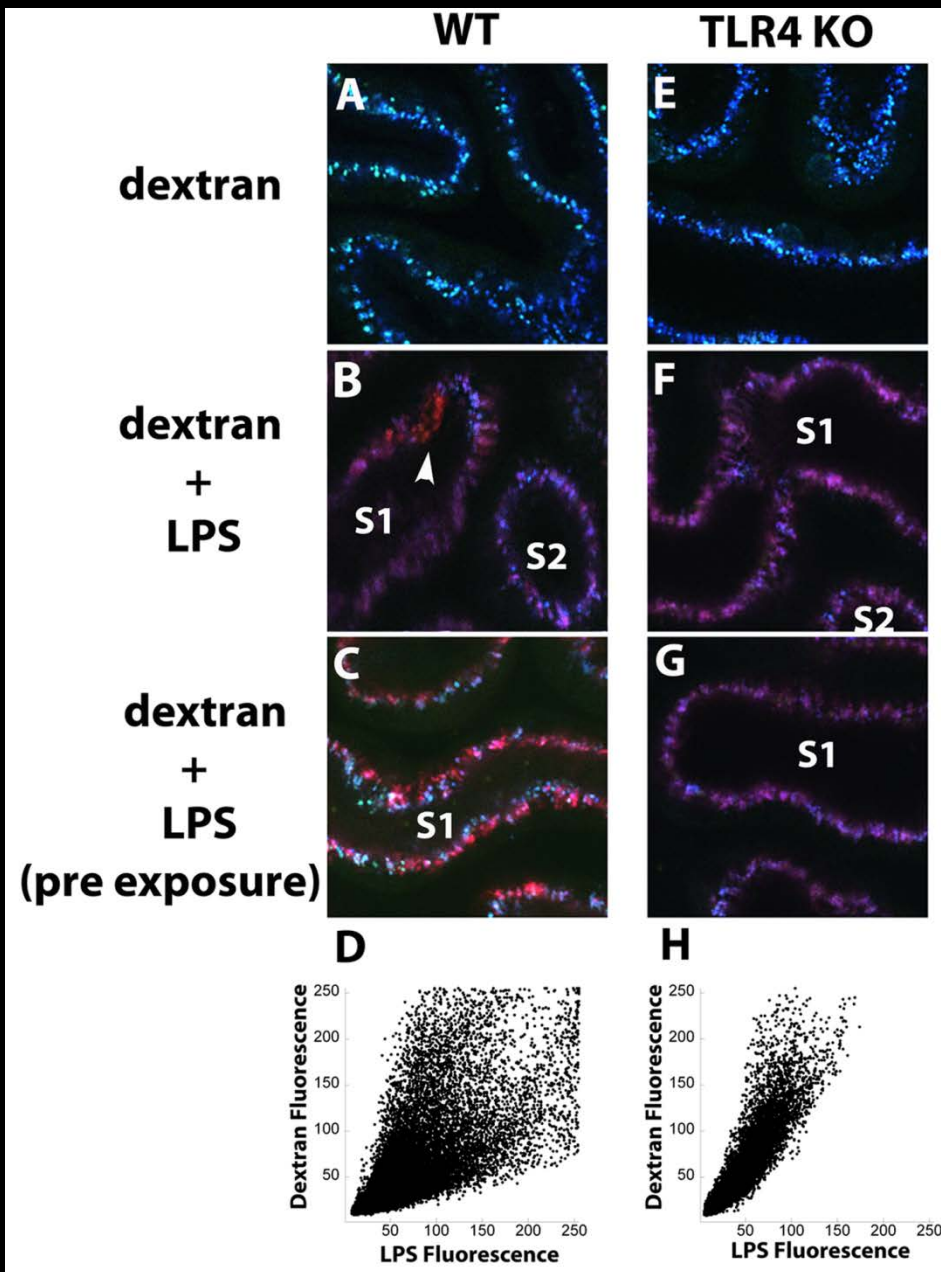


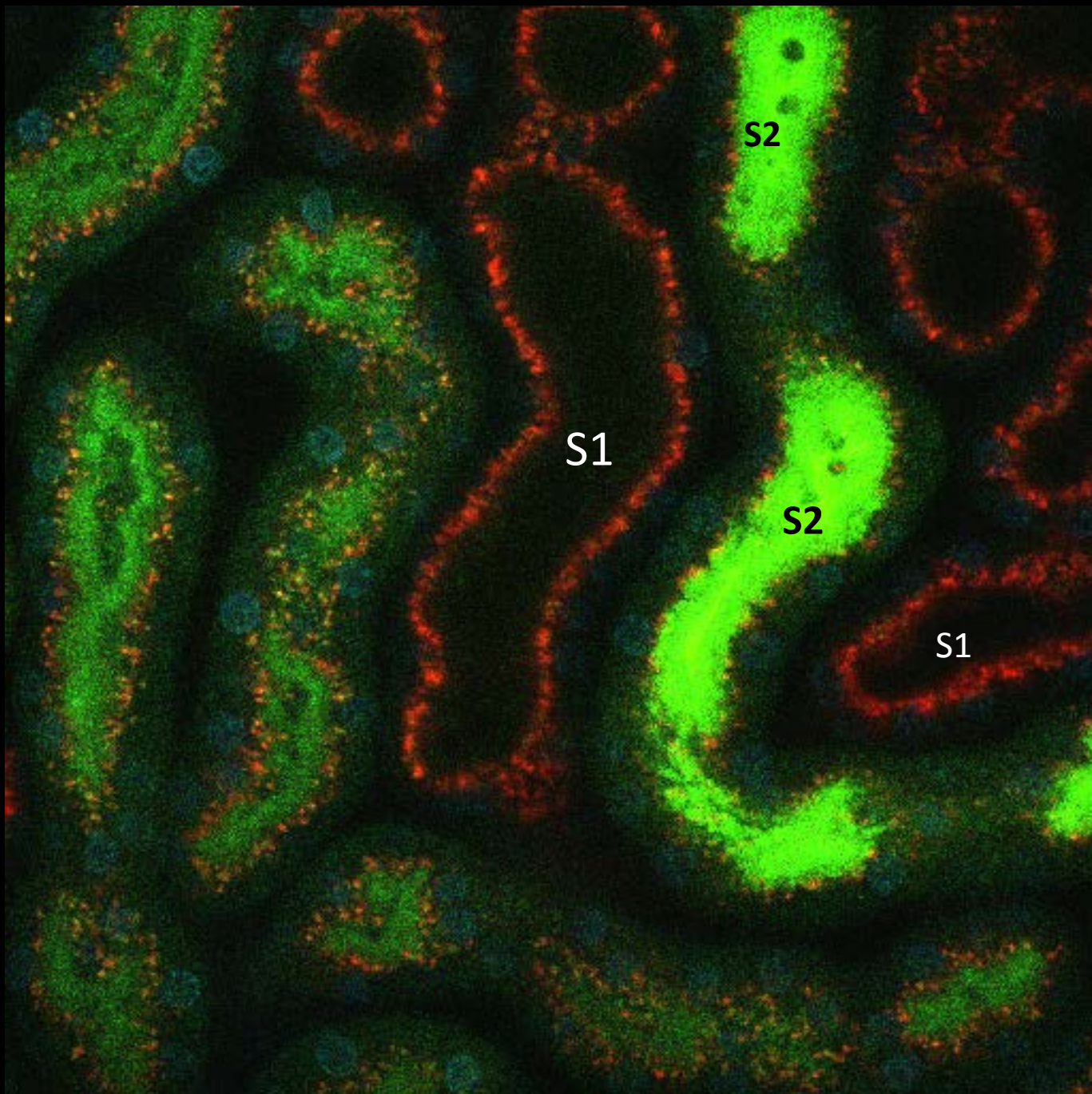
**WT**

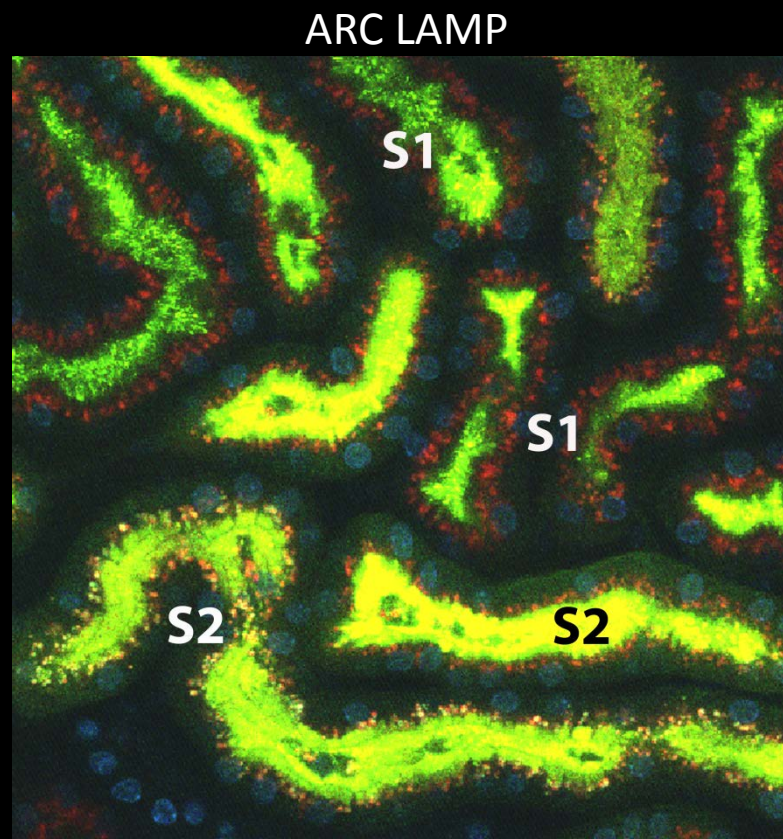
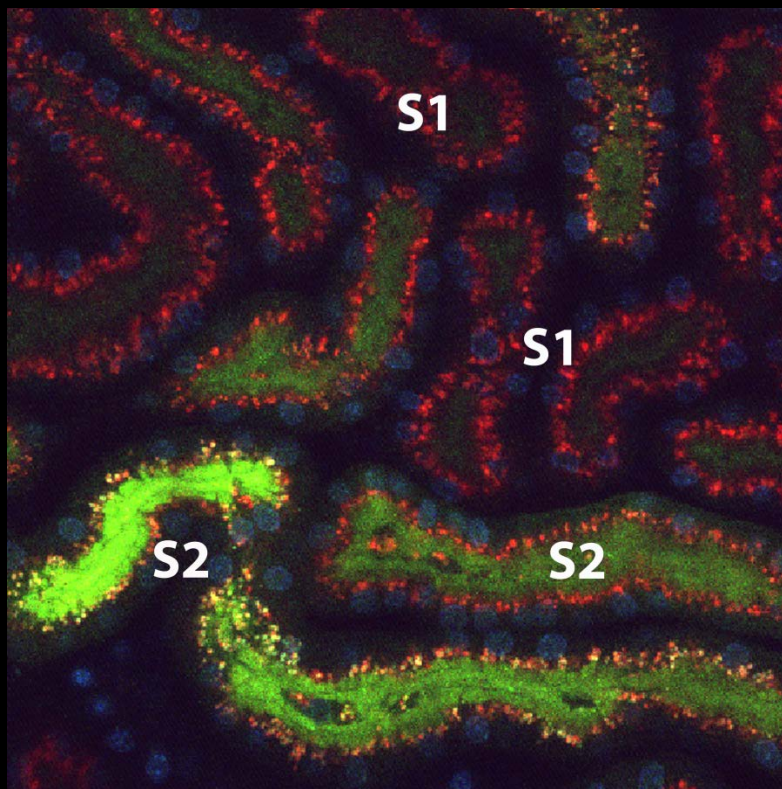


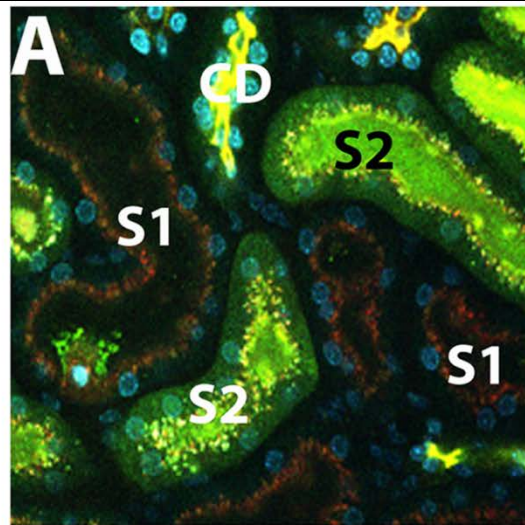
**TLR4  
KO**



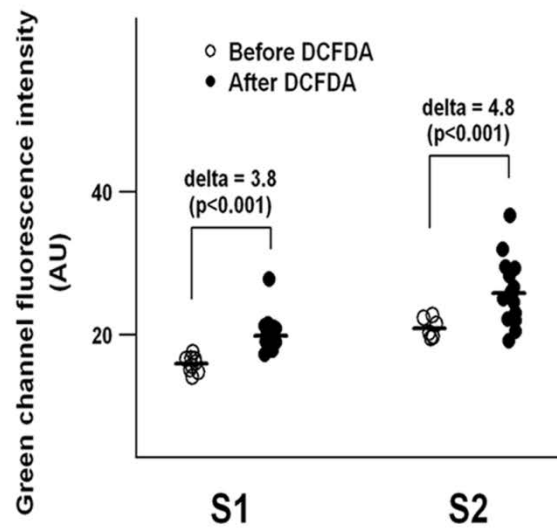




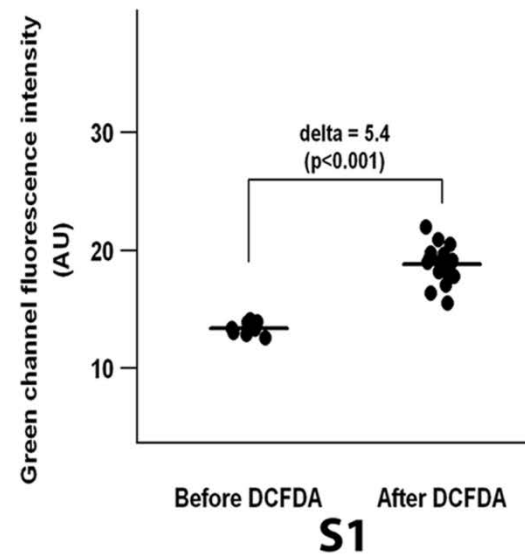


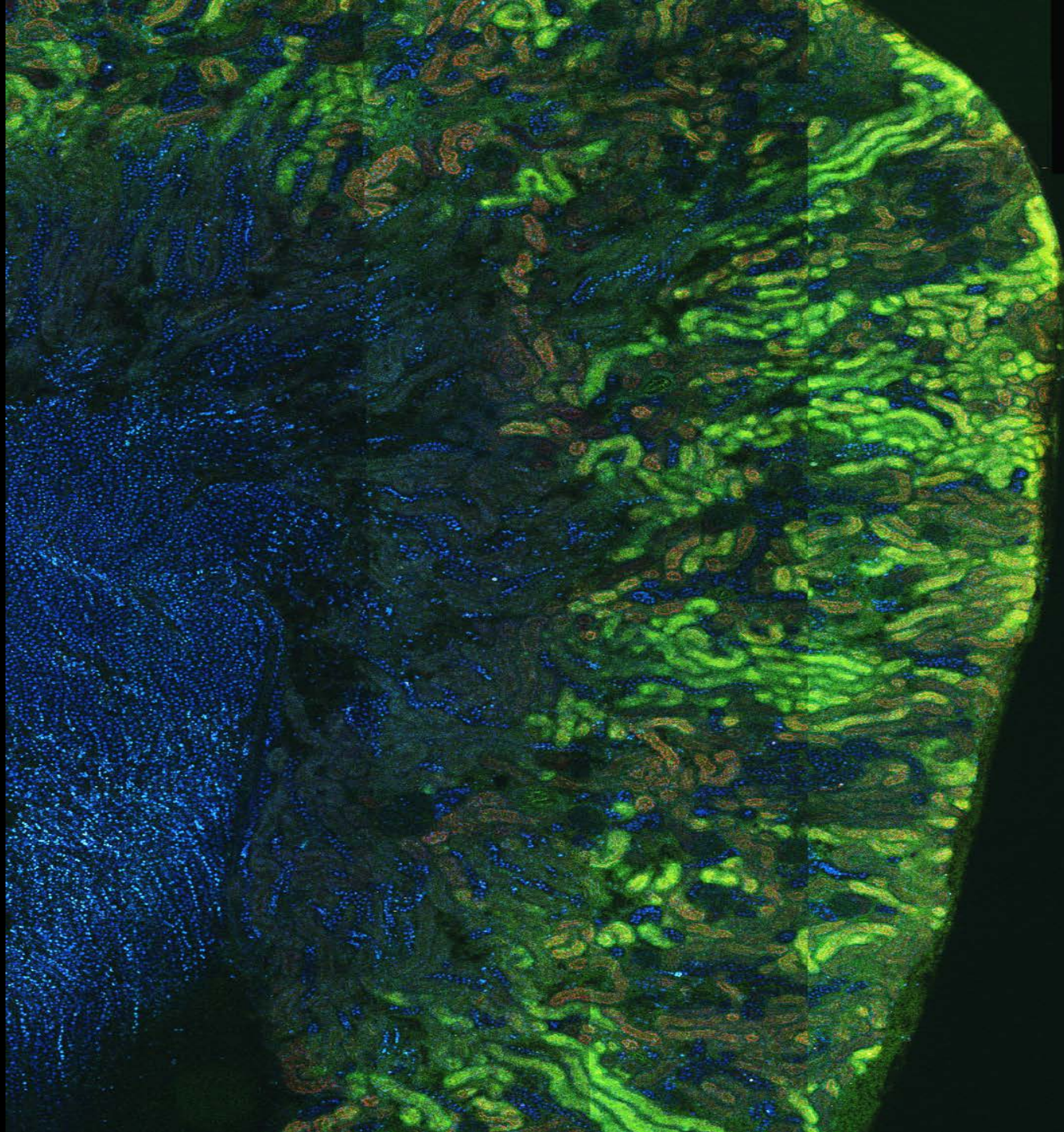


**B**



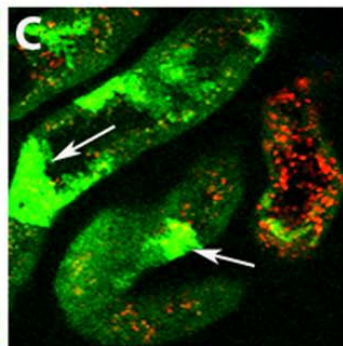
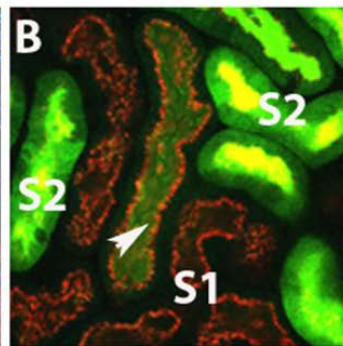
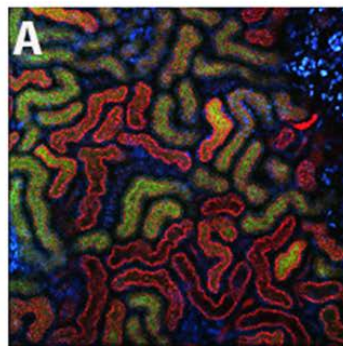
**C**



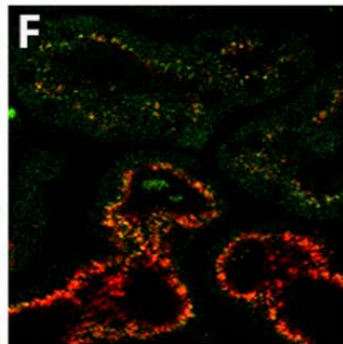
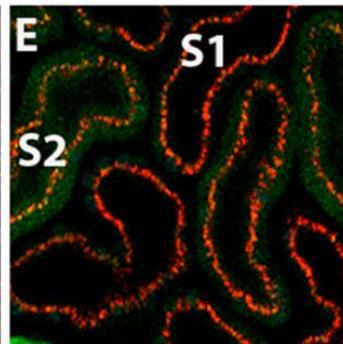
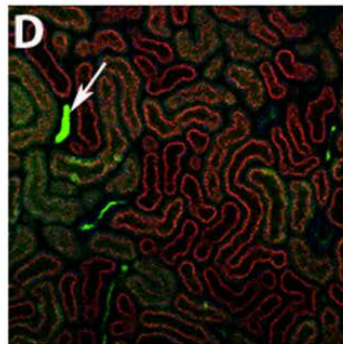




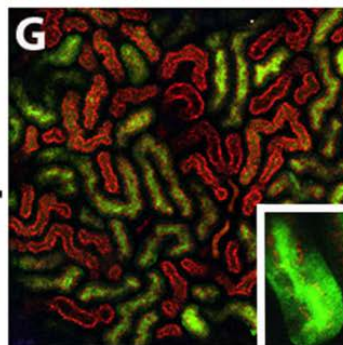
WT



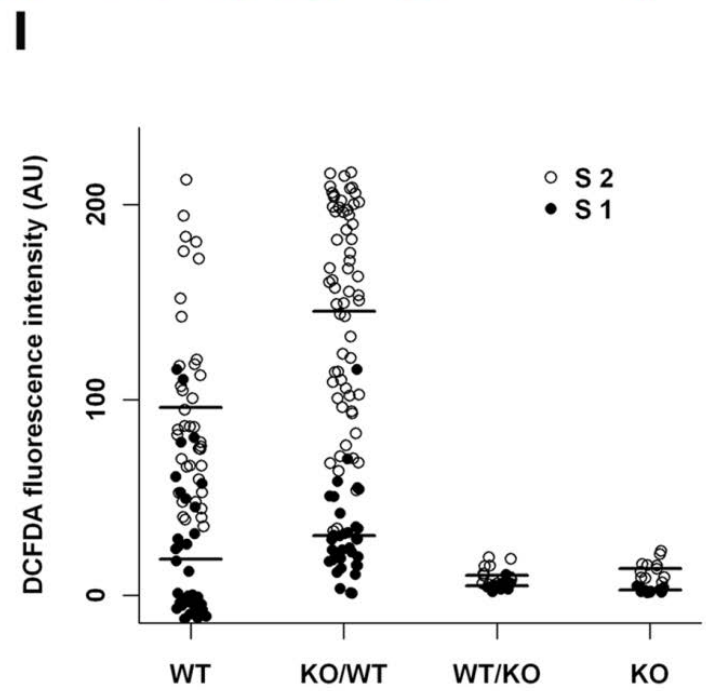
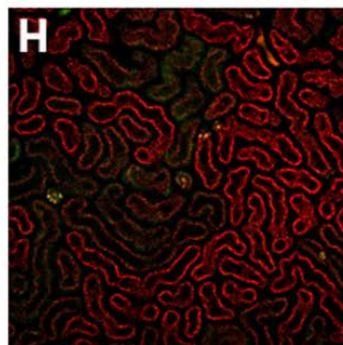
TLR4  
KO

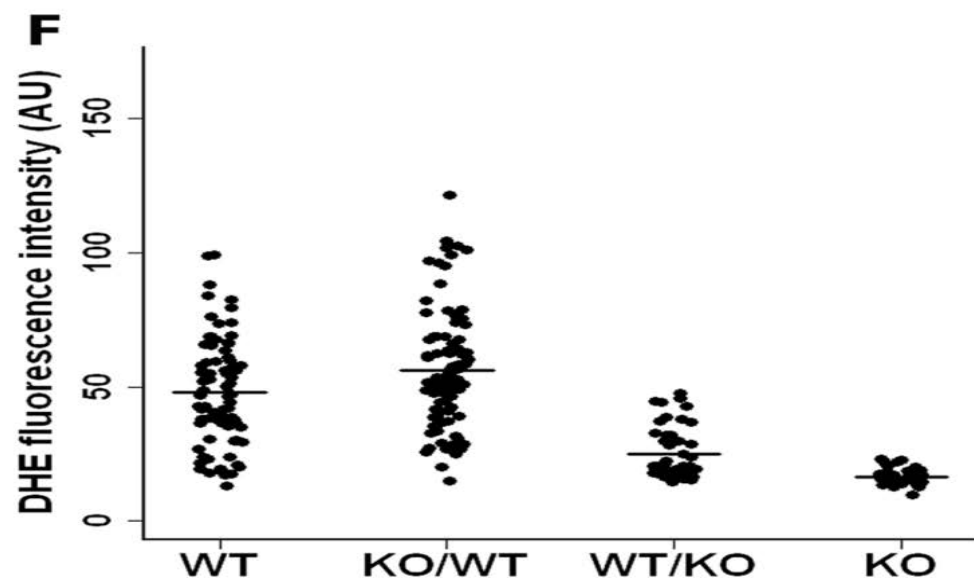
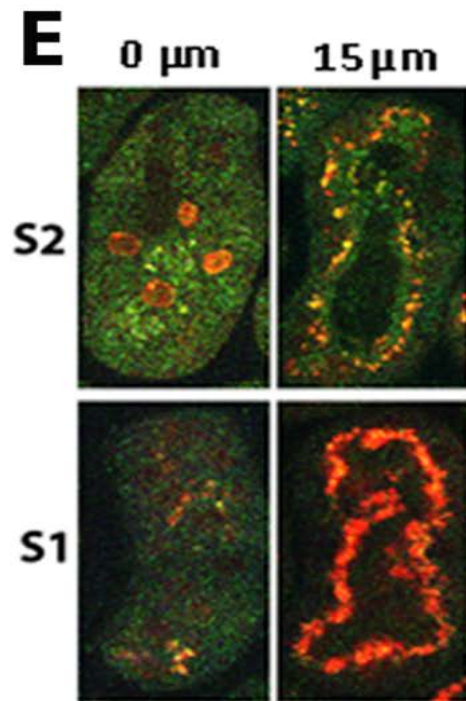
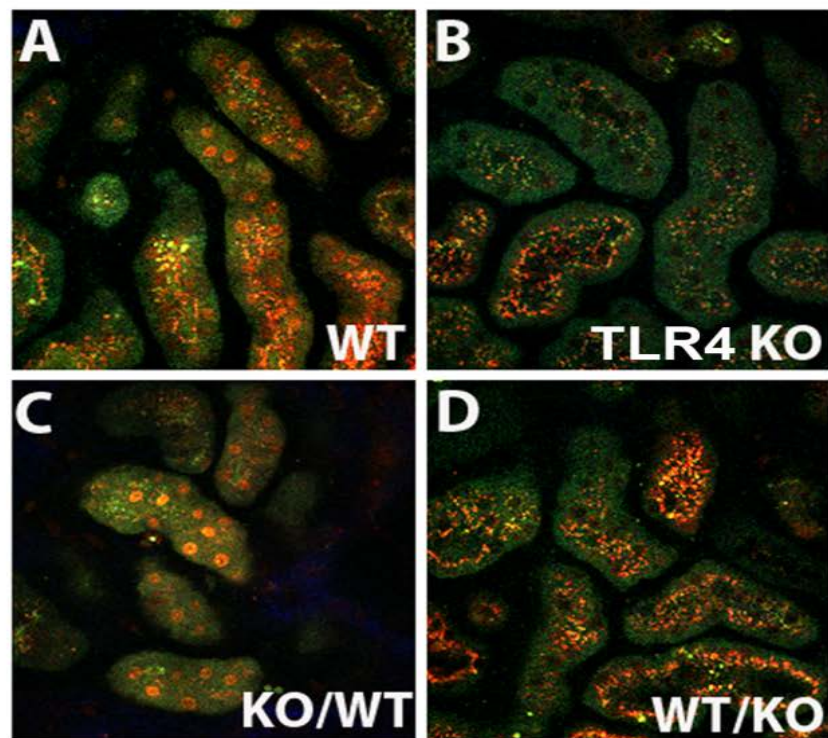


KO/WT

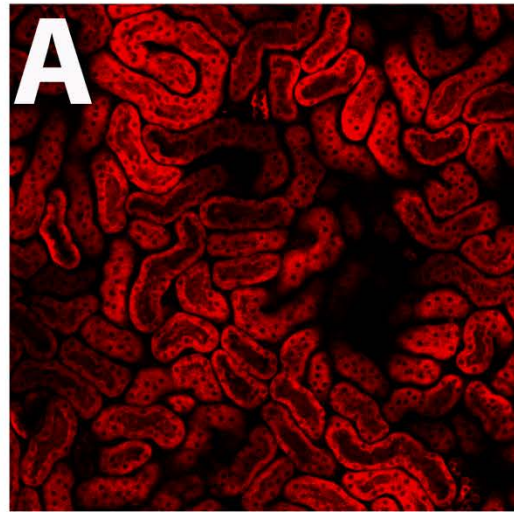


WT/KO

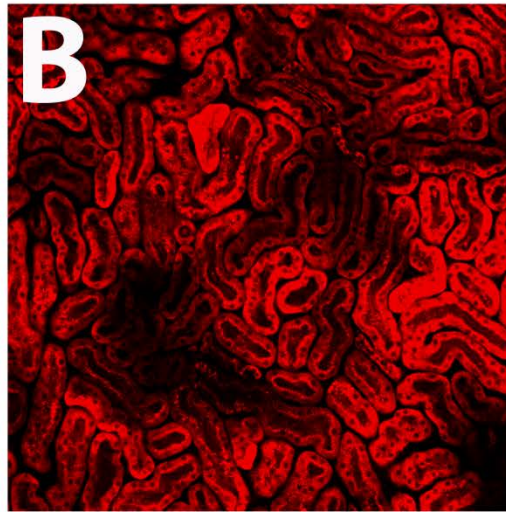




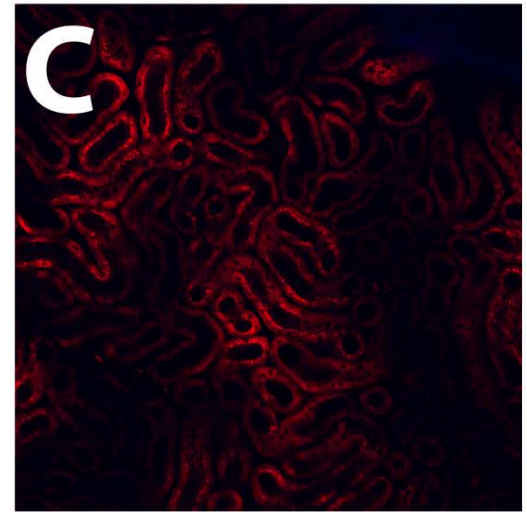
**control**



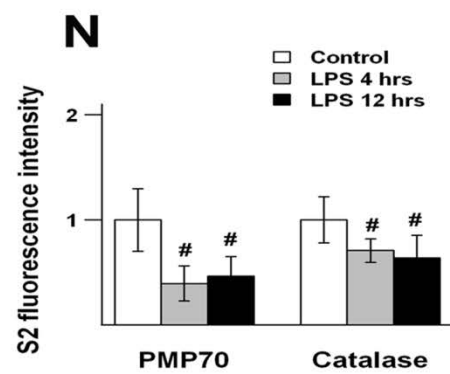
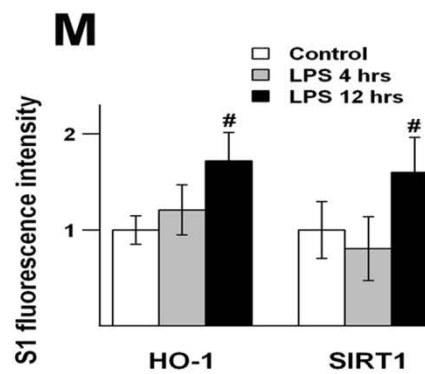
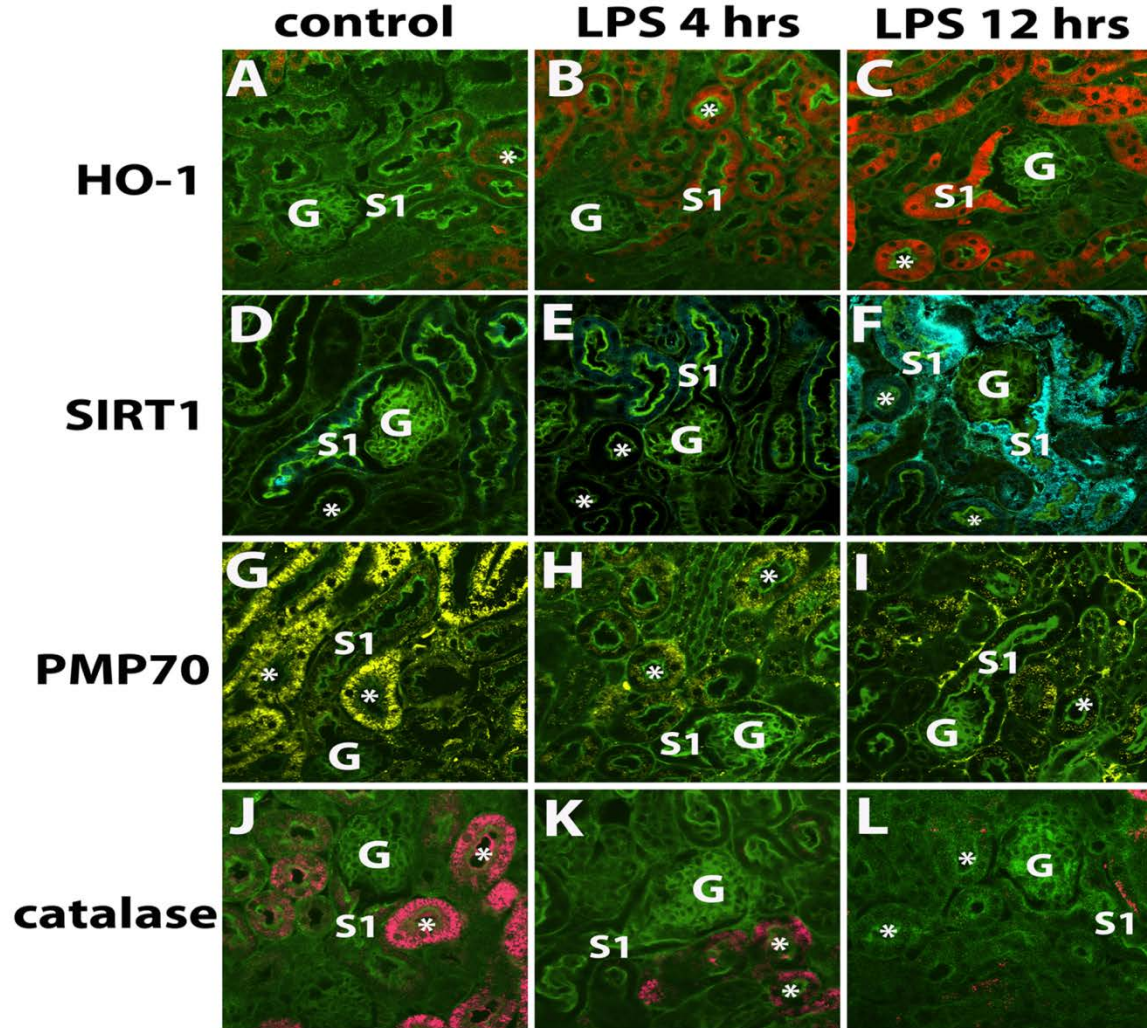
**LPS 4 hrs**



**LPS 12 hrs**

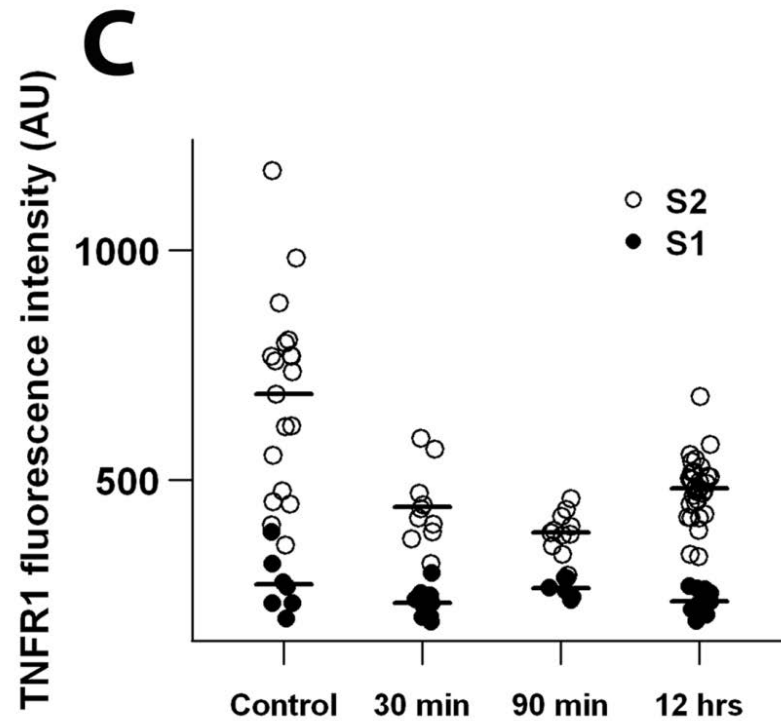
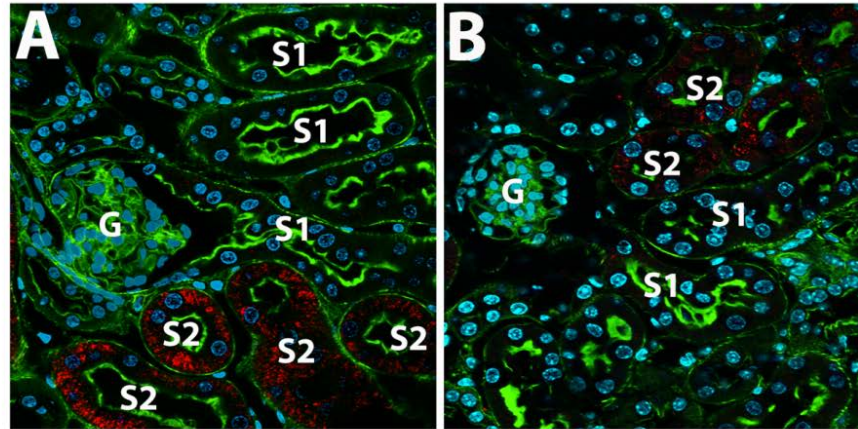


TMRM



**control**

**LPS 90 min**



No CD14

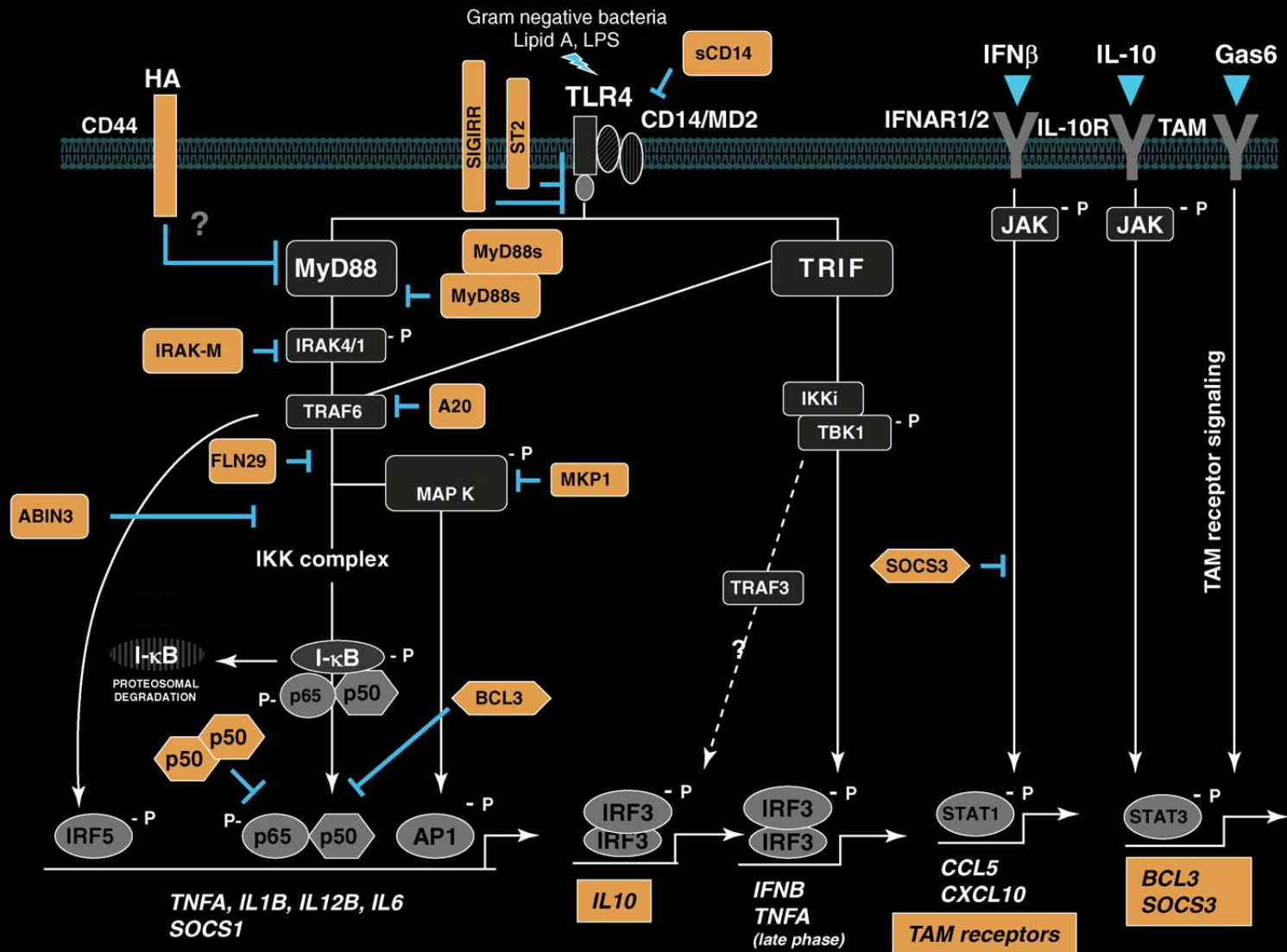
With CD14

Rough LPS



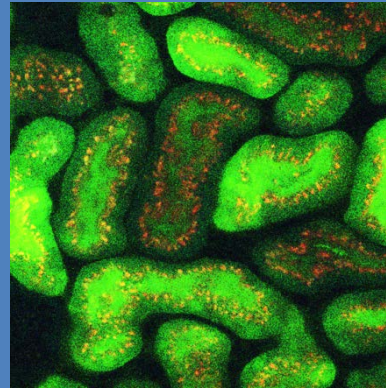
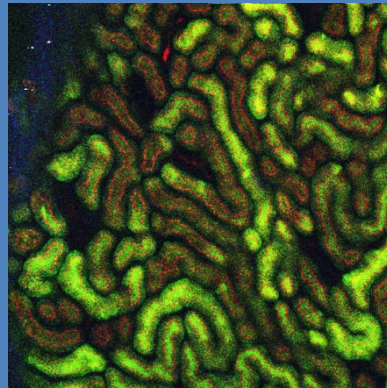
Smooth LPS



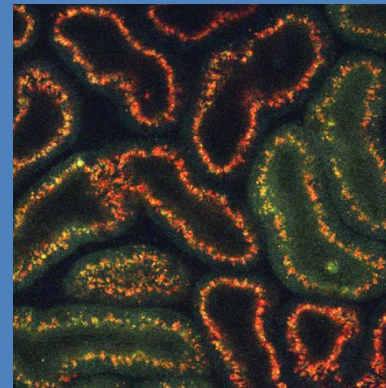
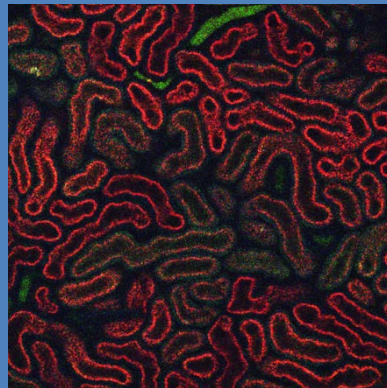


WT

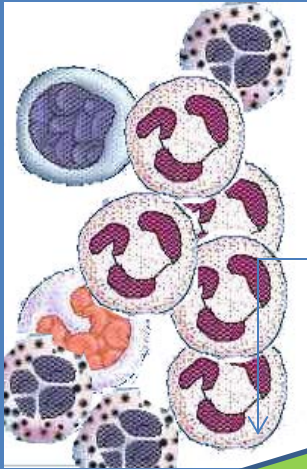
Sal 100



E 5  
Sal 100







Tolerance



LPS

Toxicity



Tolerance

