

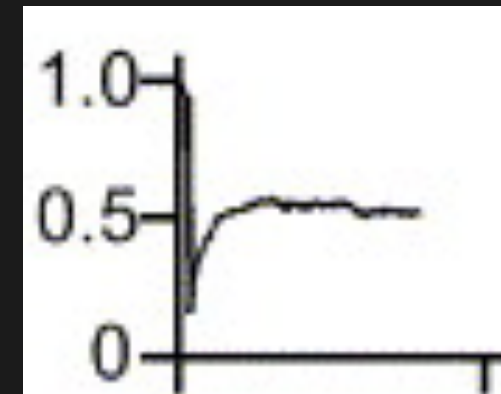
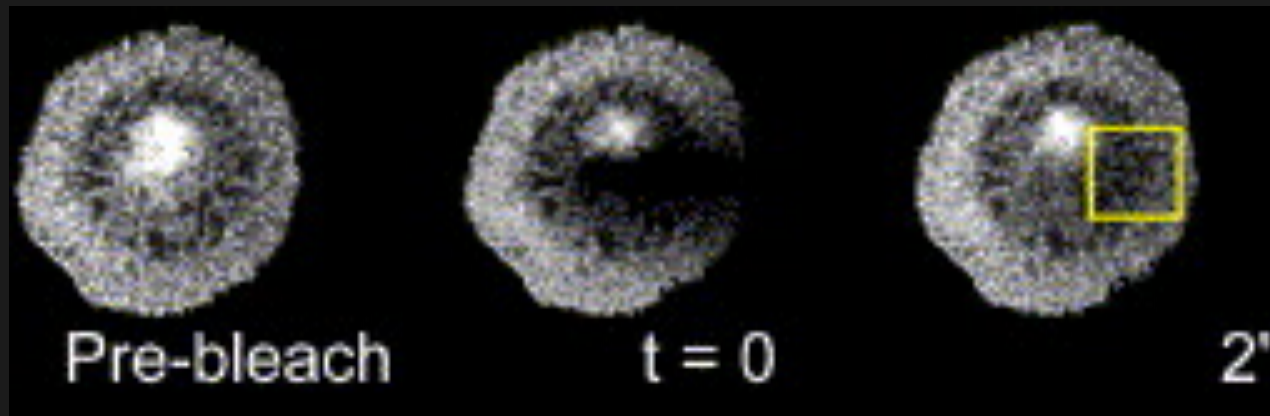
Plasma membrane microdomains
organize biochemical reactions to
transduce signals

Yoshihisa Kaizuka

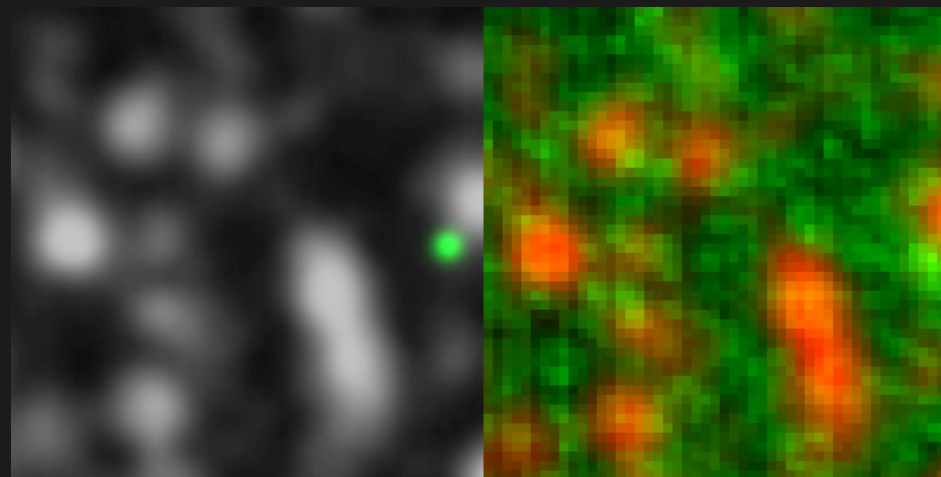
National Institute for Materials Science
University of California, San Francisco

T cell membrane microdomains: dynamic and fluid structures

Mobility of proteins: FRAP



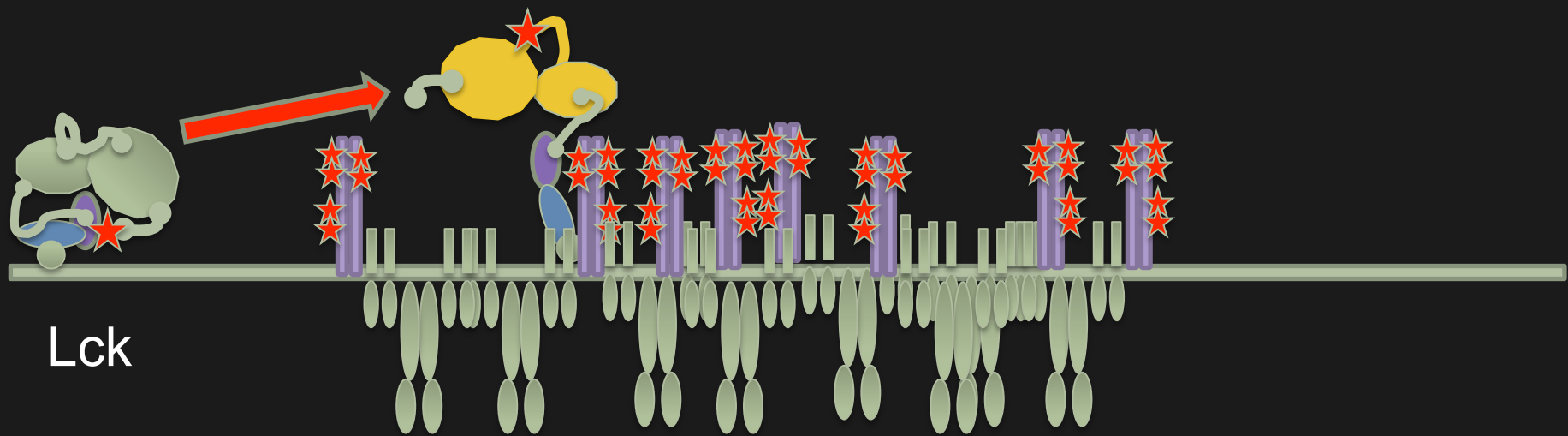
Single molecule
tracking of
fluorescent
proteins



Douglass and Vale

Modulation of molecular conformations- functions by lateral organizations

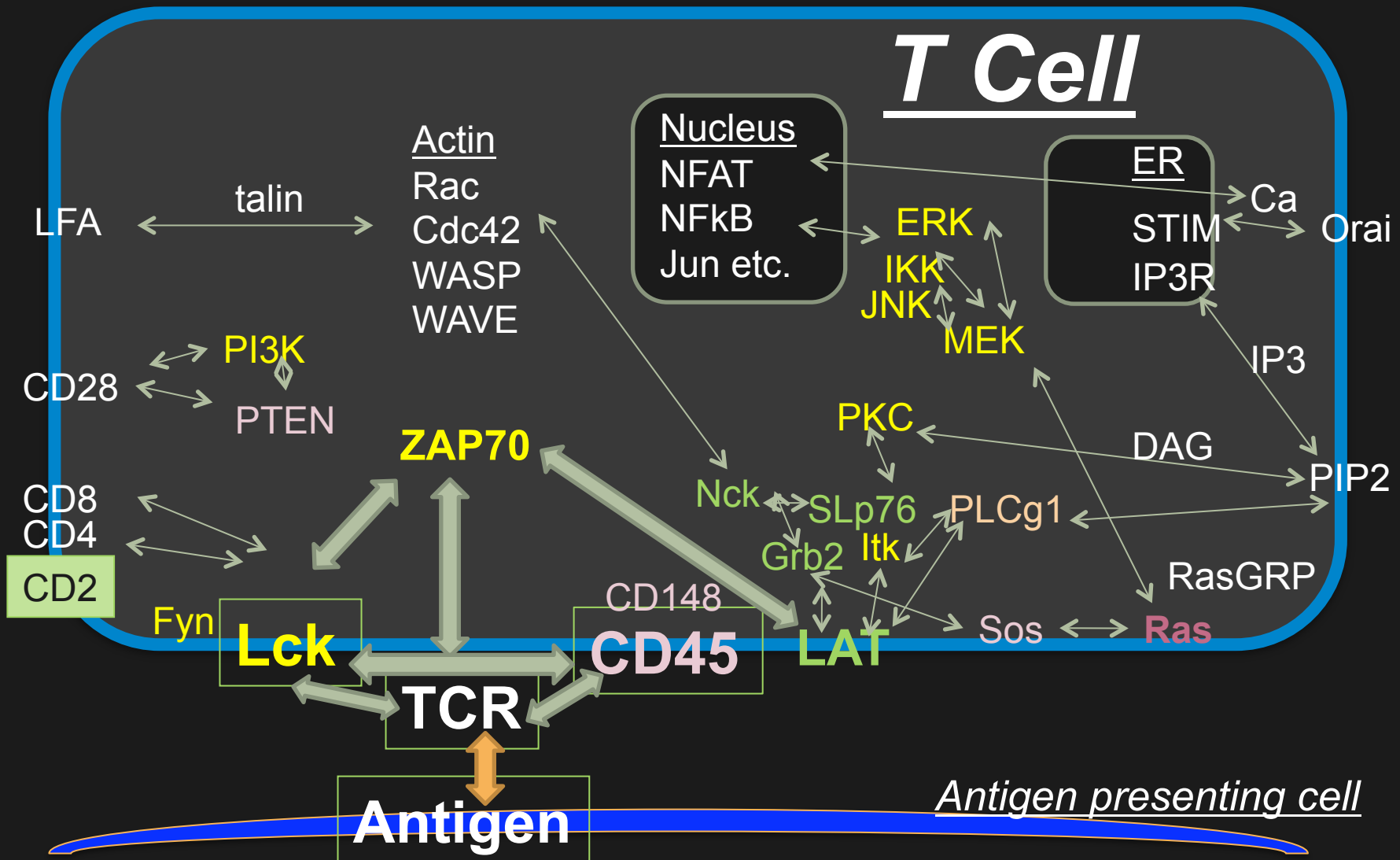
T cell



Lck

Receptor enriched membrane microdomain

Signal transduction in T cell activation

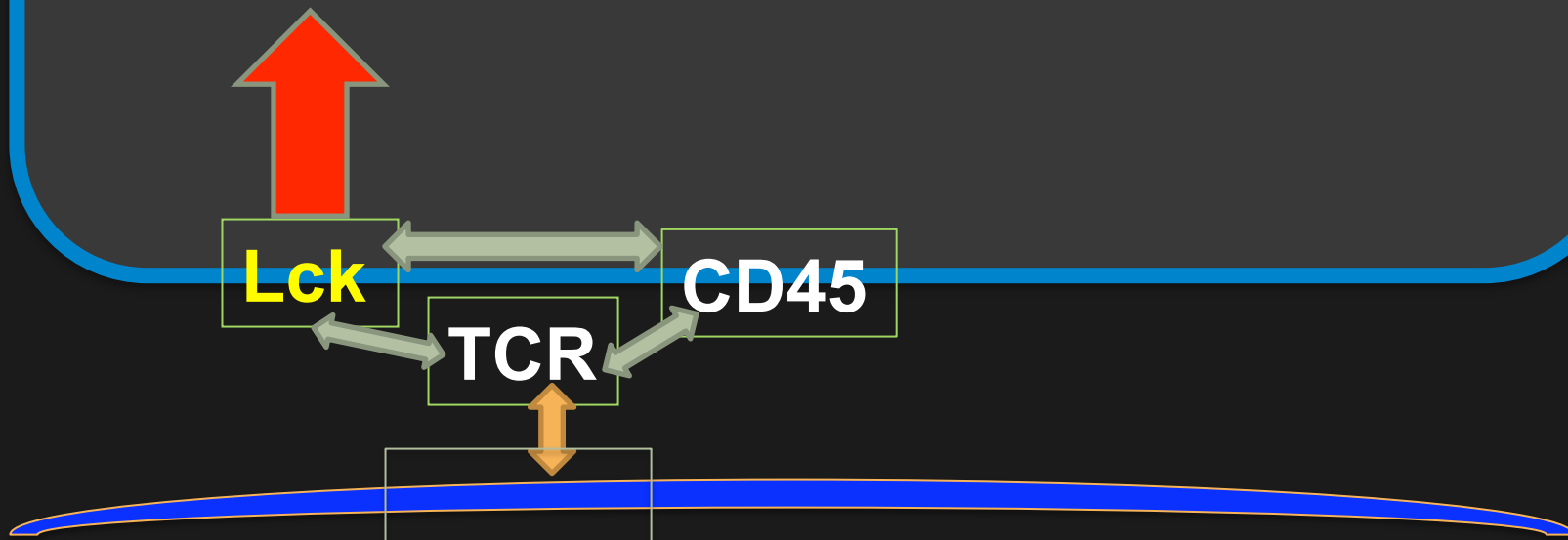


Critical kinase Lck is activated on plasma membrane

T Cell

How is Lck activated?

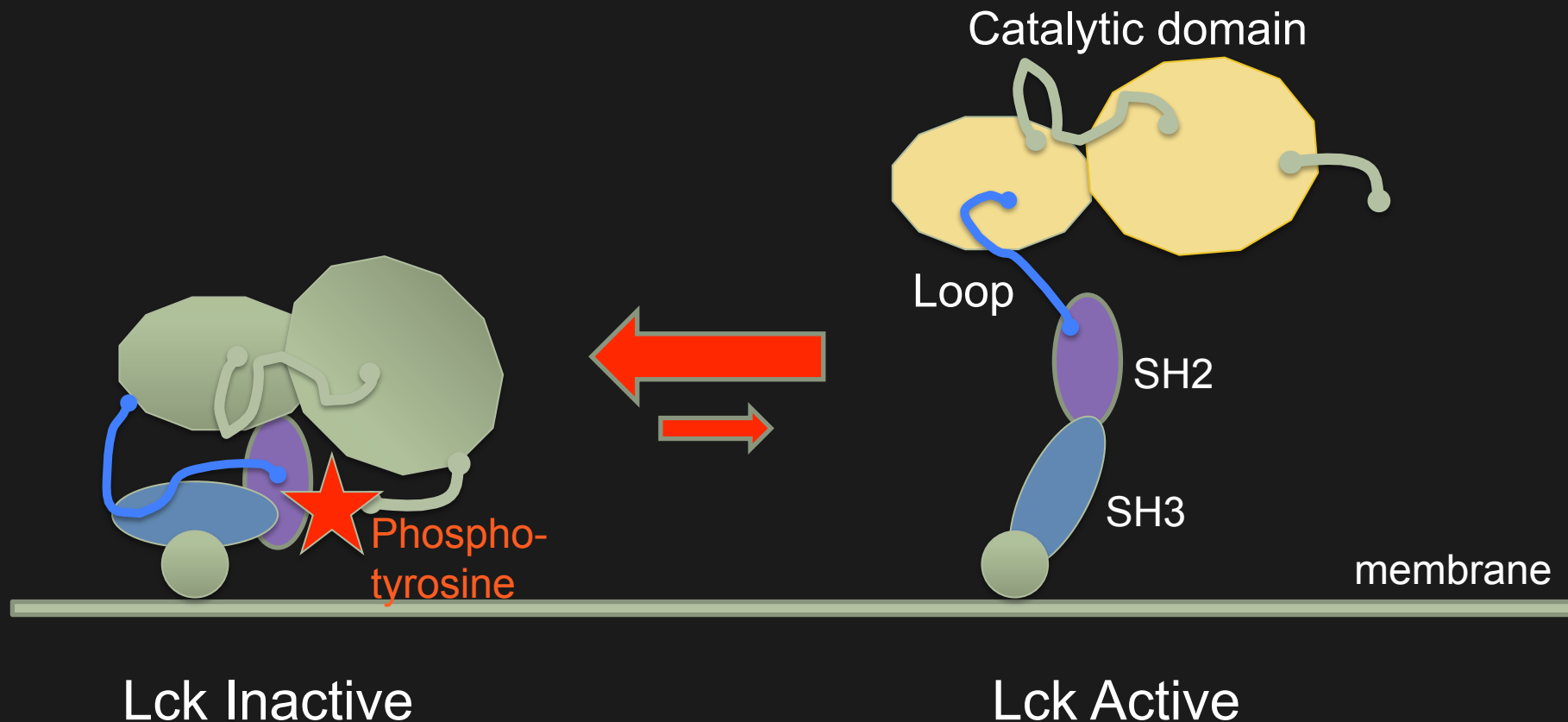
Lck (src kinase) – CD45 (phosphatase) – Receptor



How is Lck activated in T cell?

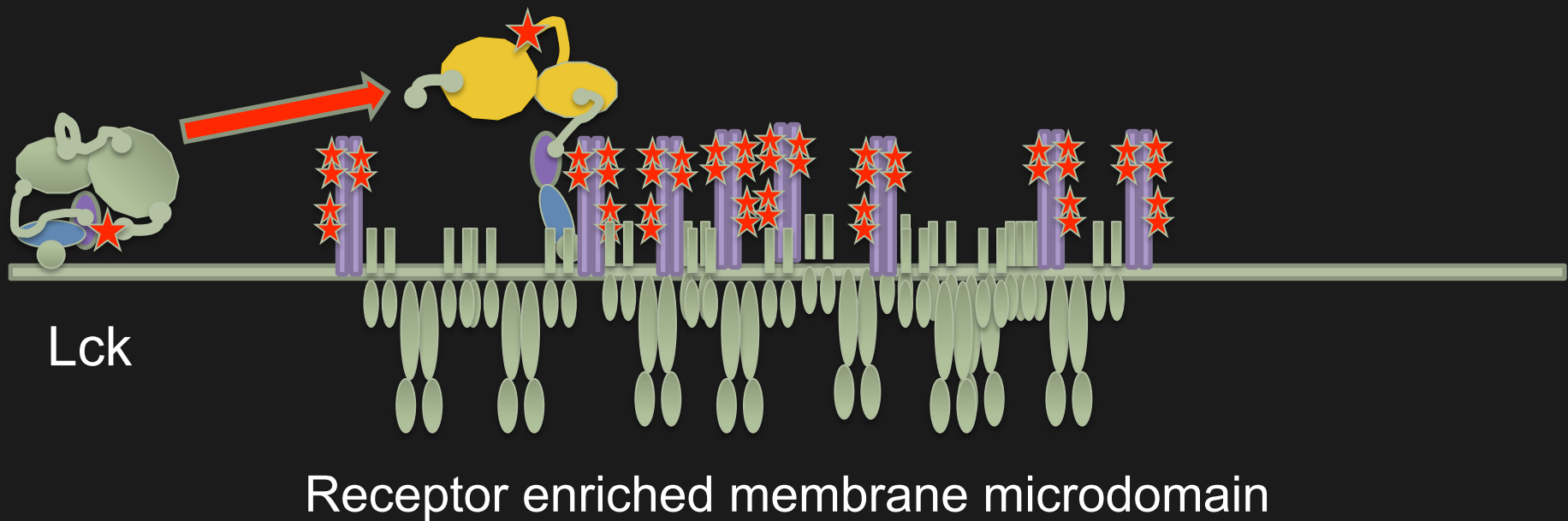


How is Lck opened in T cell?
(release of autoinhibitory interactions)

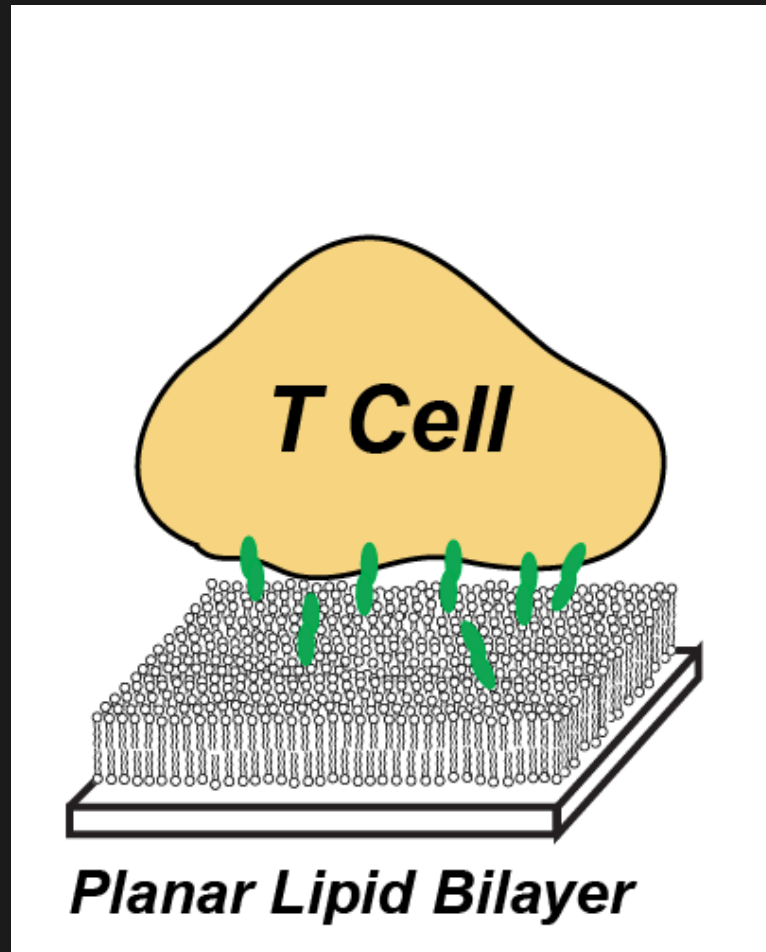


Model: Locally Concentrated Receptors Activate Lck by Releasing Autoinhibitory Interactions

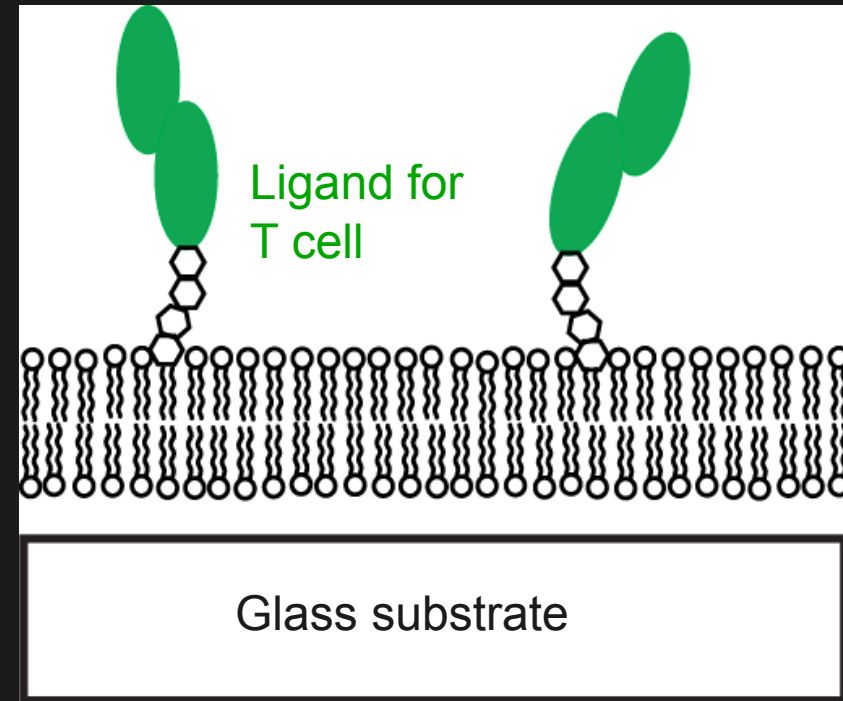
T cell



Imaging T Cell membrane microdomains

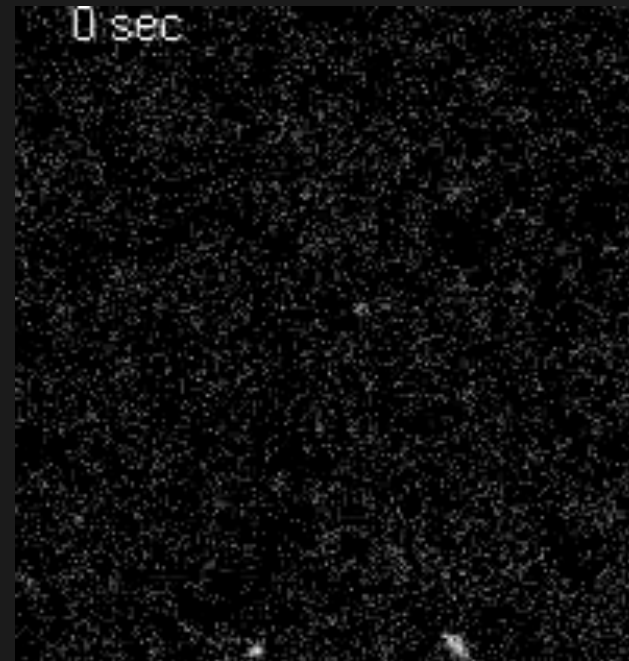
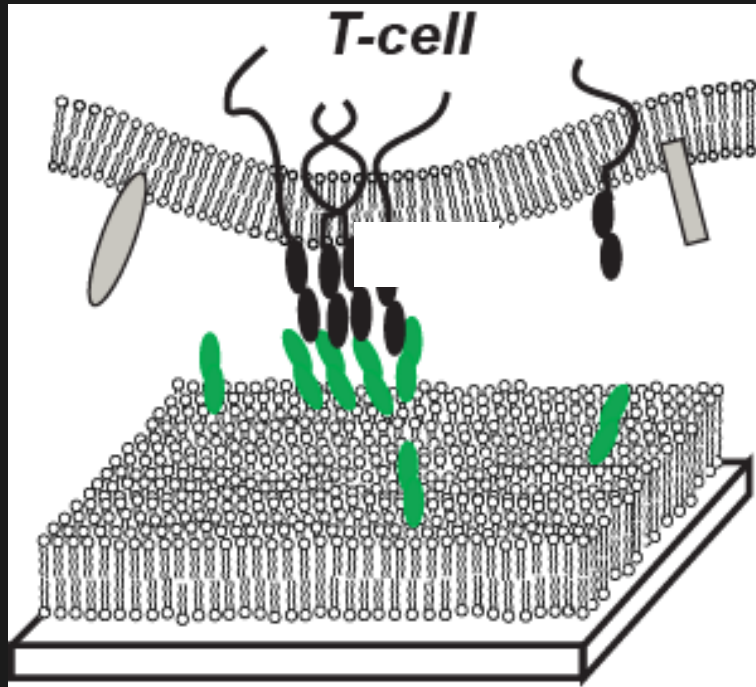


Side View



- Mimic cell-cell interaction
- Biochemical control
- Surface-based imaging (TIRF, confocal)

Microdomain formation in activated T Cell membranes

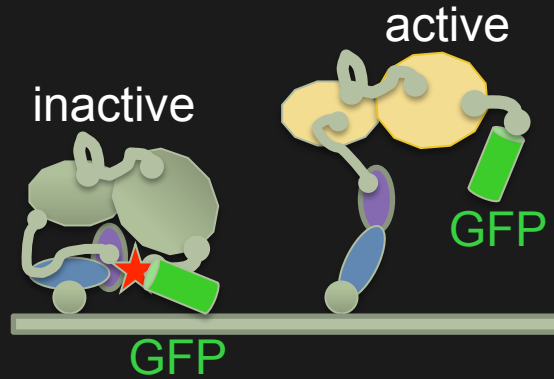


5 μm

- Functional assays show T cell is activated upon microdomain formations, suggesting Lck activation.
- How does Lck activation occur?

Lck is activated in microdomains

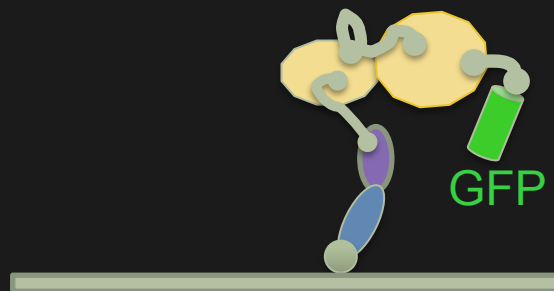
Wildtype Lck



microdomains



Open form mutant Lck
(Y505F)

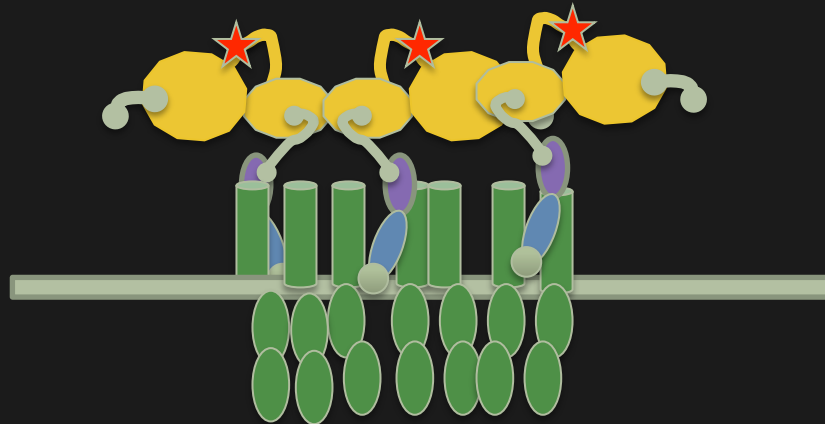


microdomains

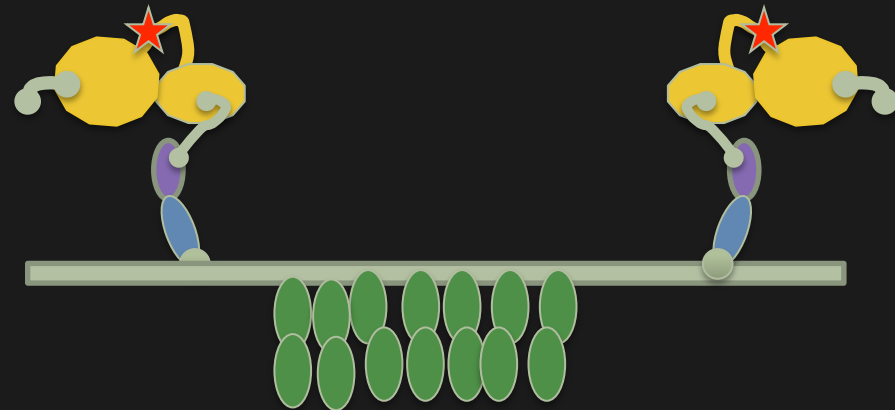


2 μ m

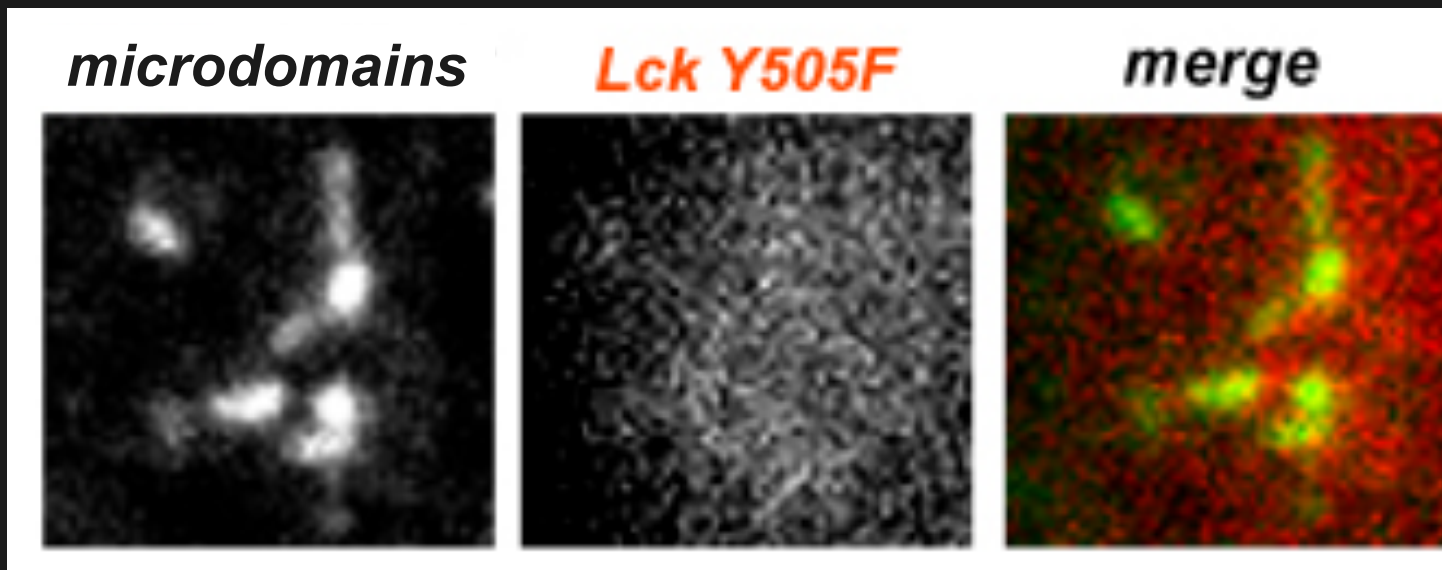
Mechanisms for Active Lck Localization (1)



Open Lck enrichment in microdomain

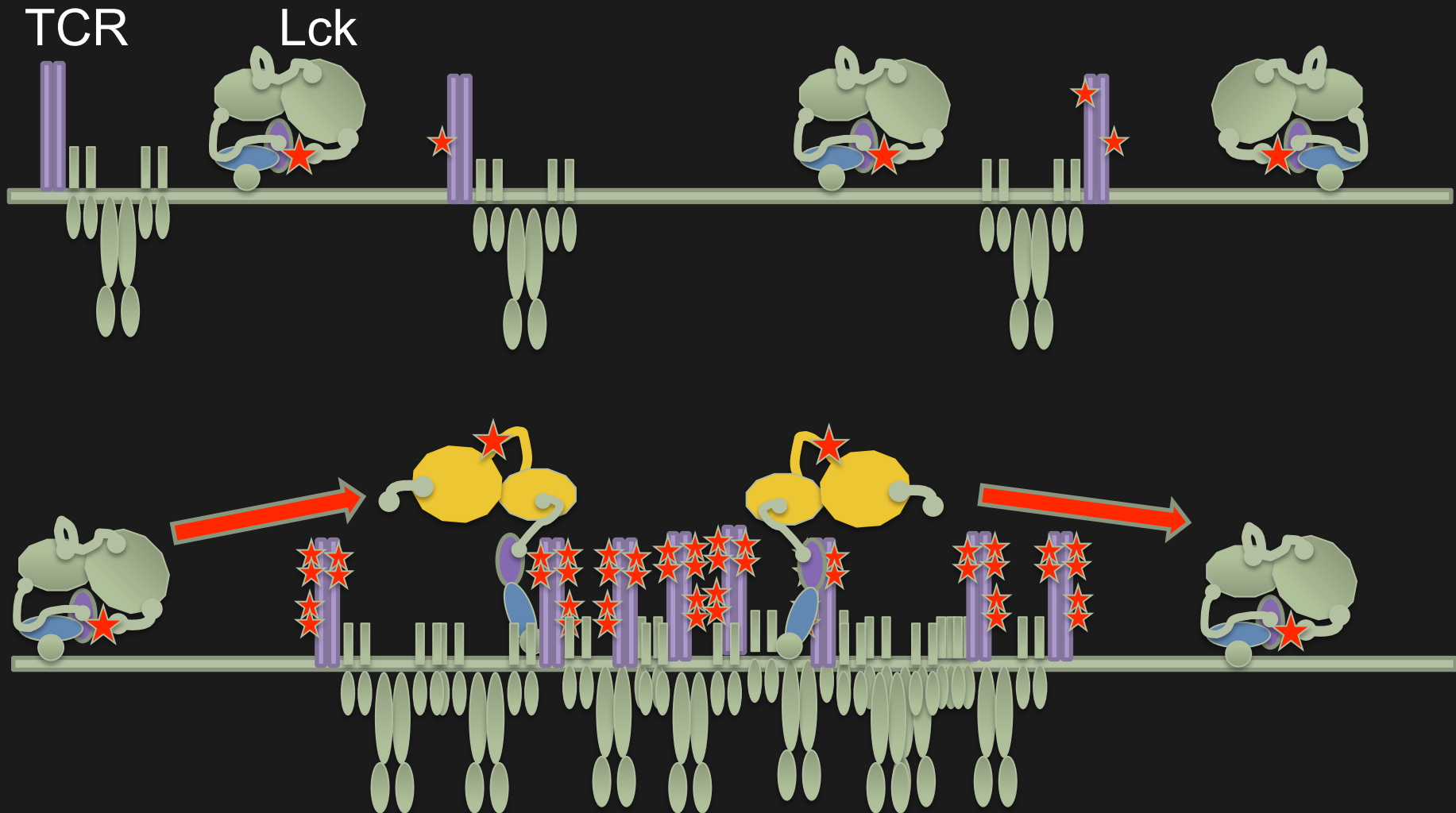


Cytoplasmic domain deletion

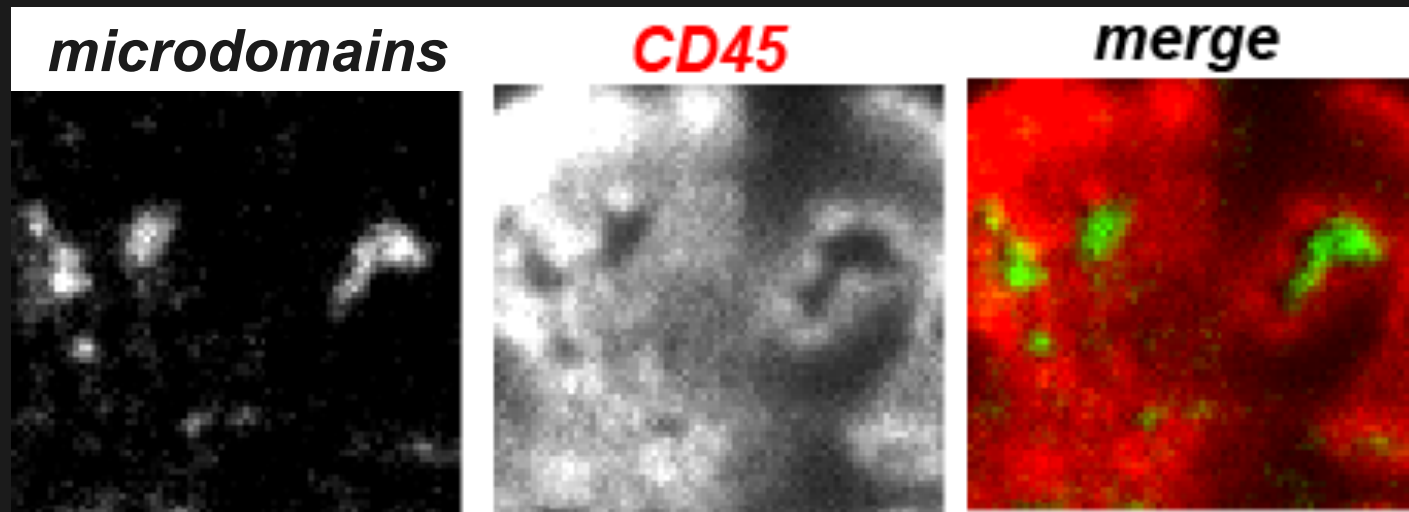
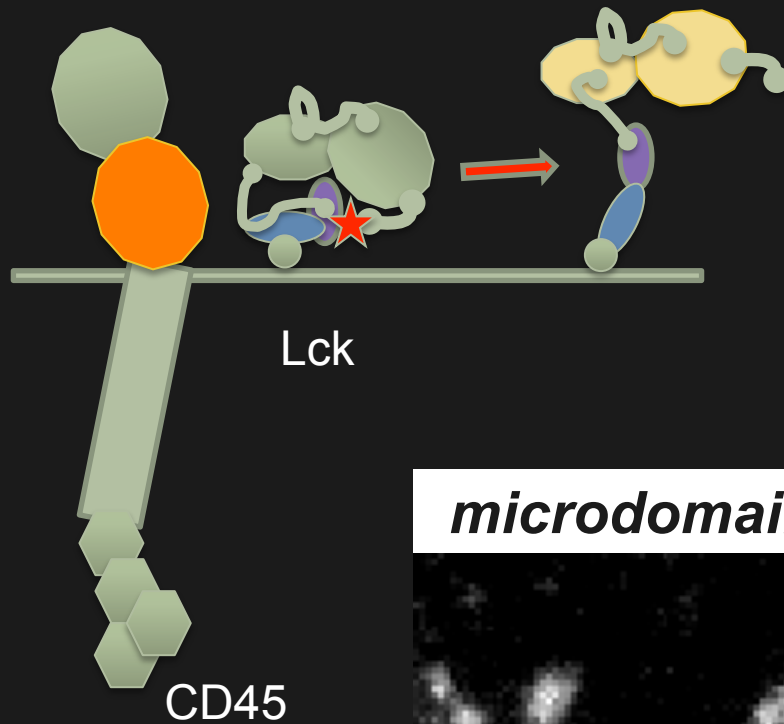


Receptor cytoplasmic domain is required for Lck activation

Dynamic and transient activation of Lck in microdomains facilitates T cell activation



Mechanisms for Active Lck Localization (2)



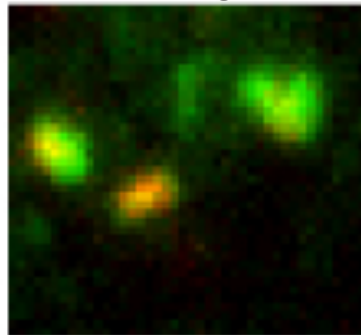
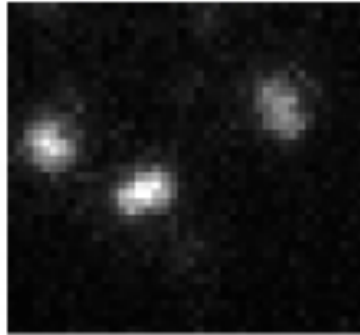
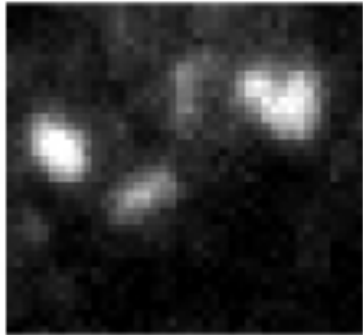
Phosphatase CD45 is excluded from microdomain
→ CD45 as a negative regulator

Localized Active Lck Facilitates Downstream Signaling

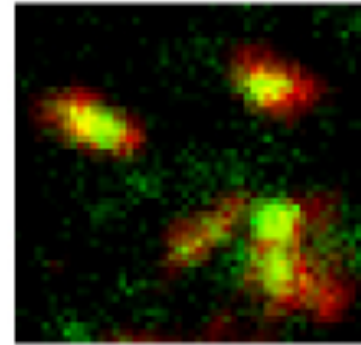
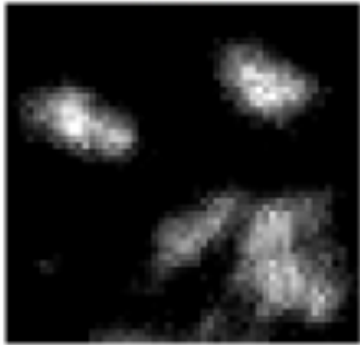
microdomains

pTCR ζ

merge



LAT



Proliferation, differentiation

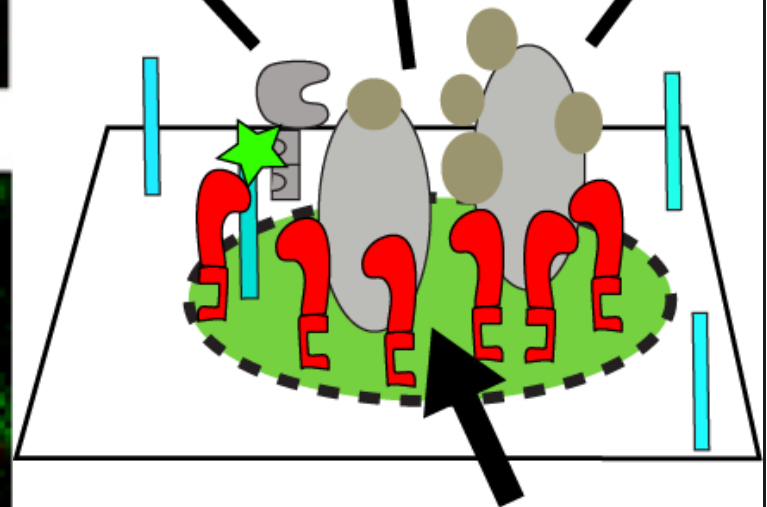


Transcription

Ras

Calcium

Kinase



Microdomain enriched with active Lck

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