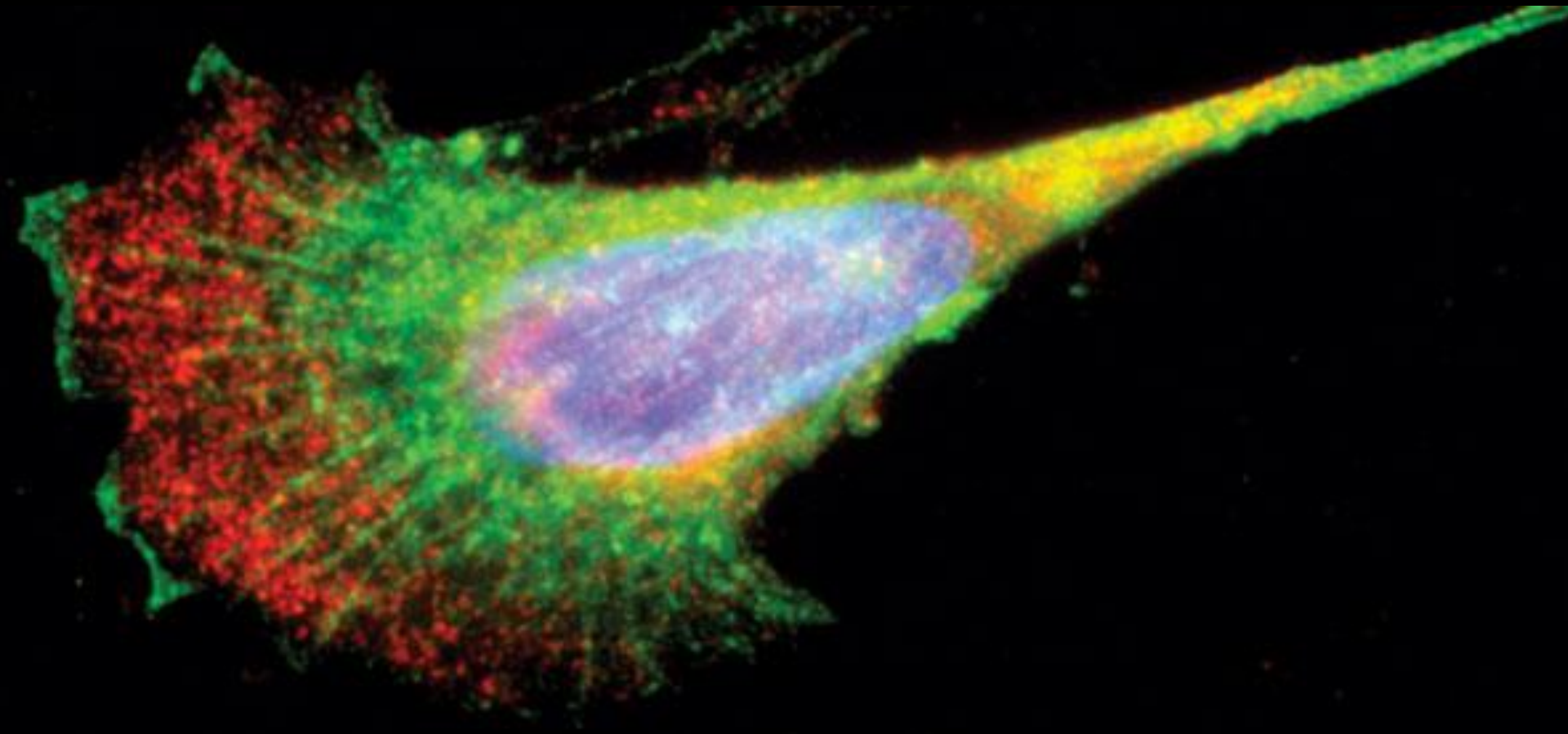
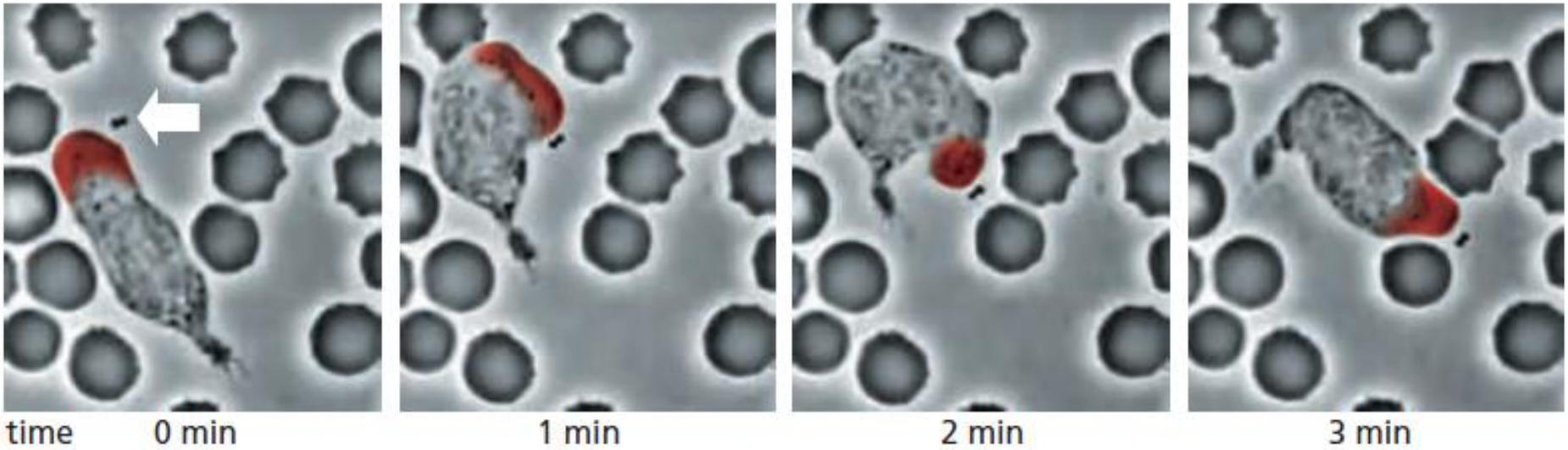
Motilidad celular



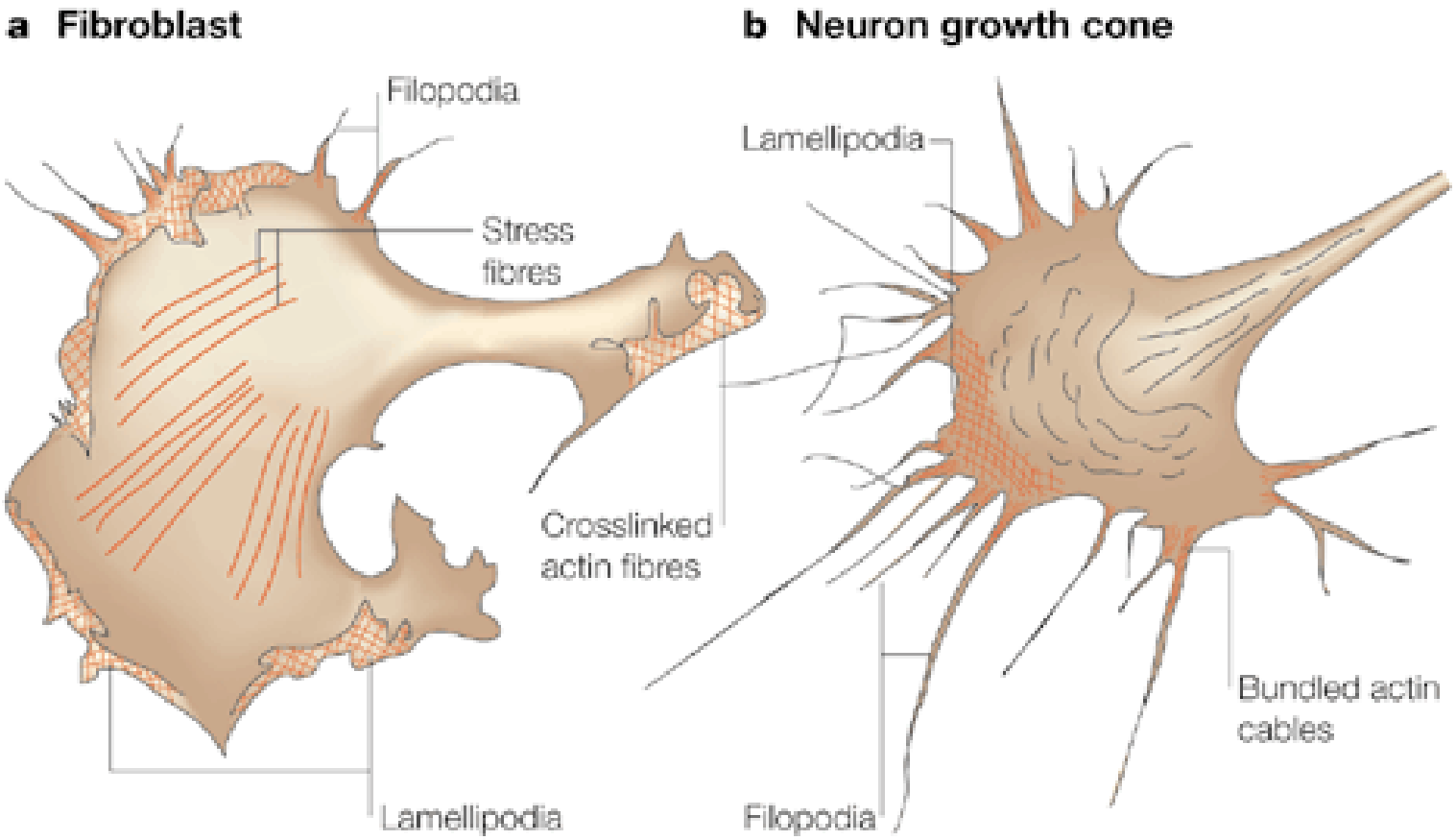
Uriel Koziol
ukoziol@fcien.edu.uy

https://www.youtube.com/watch?v=JnlULOjUhSQ

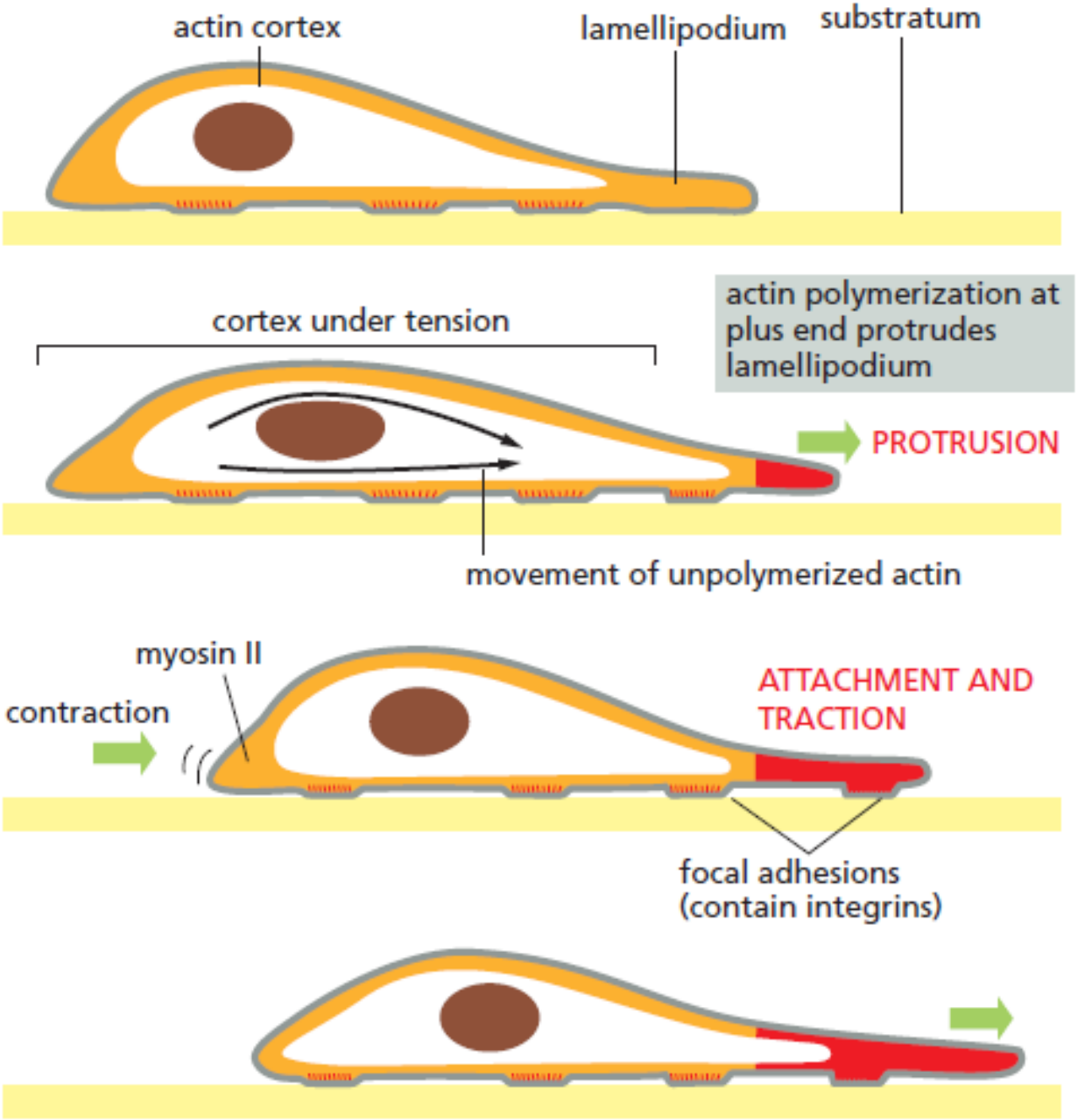

time
0 min
1 min
2 min
3 min

MIGRACIÓN CELULAR

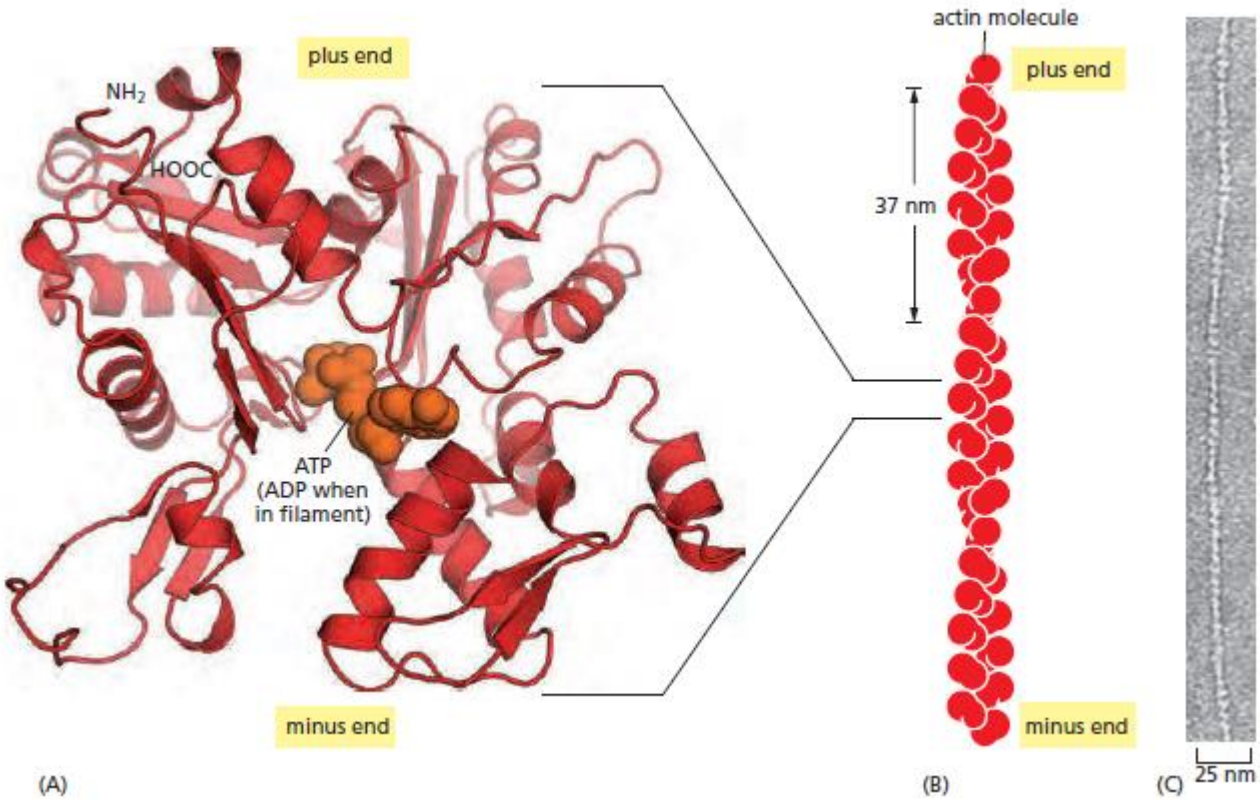
Proyecciones celulares

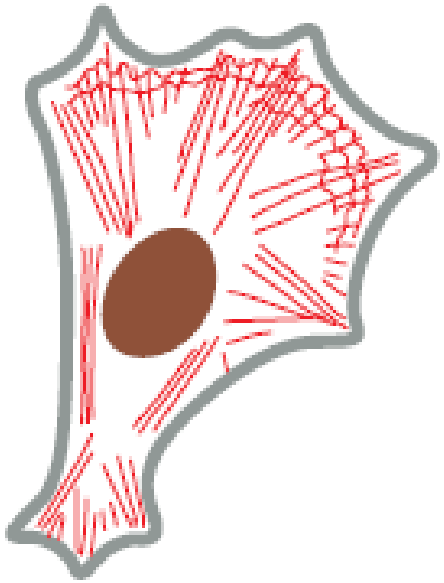


Nature Reviews | Neuroscience


actin cortex
lamellipodium
substratum
cortex under tension
actin polymerization at plus end protrudes lamellipodium
PROTRUSION
movement of unpolymerized actin
myosin II
contraction
ATTACHMENT AND TRACTION
focal adhesions (contain integrins)

Repaso: Filamentos de Actina

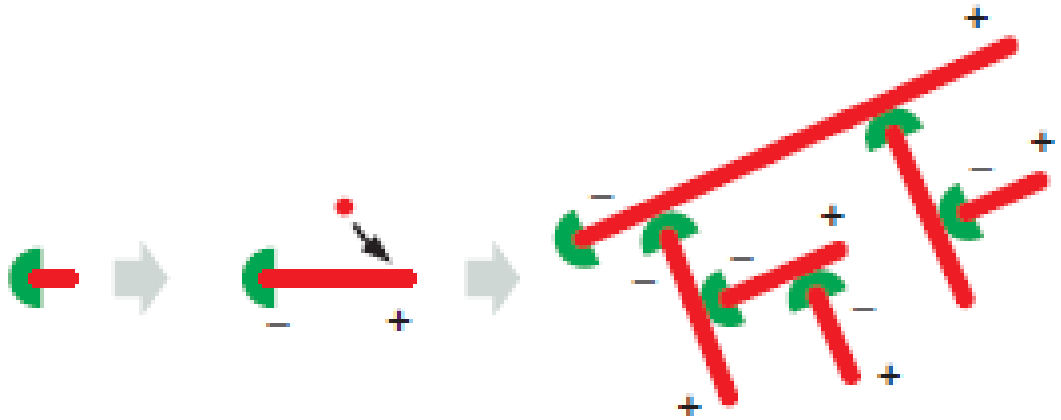




Repaso:
Polimerización y despolimerización de actina *in vivo*

cofilin

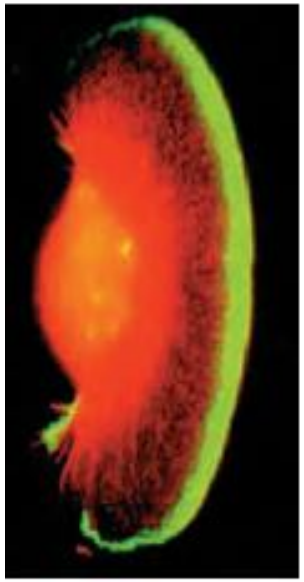
binds ADP-actin filaments, accelerates disassembly



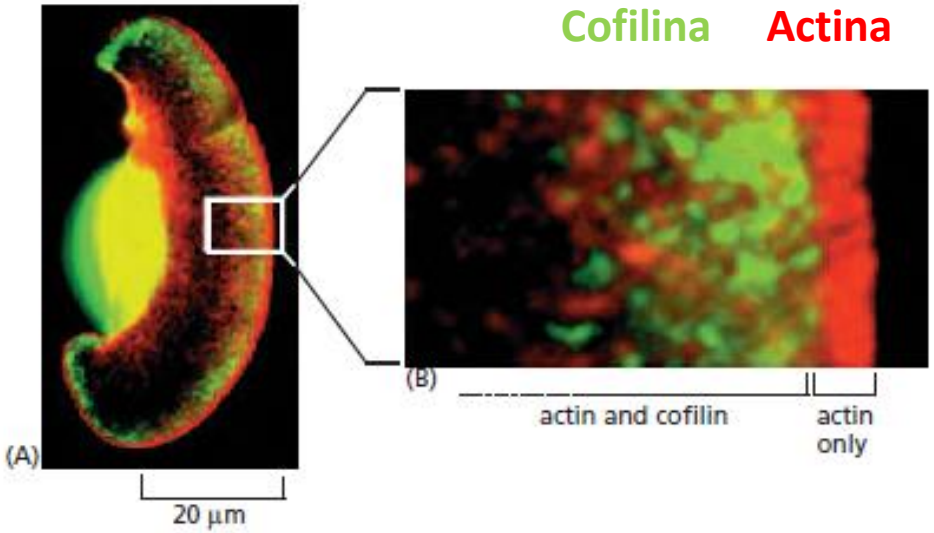
Arp2/3 complex

nucleates assembly to form a web and remains ··sociated with the minus end

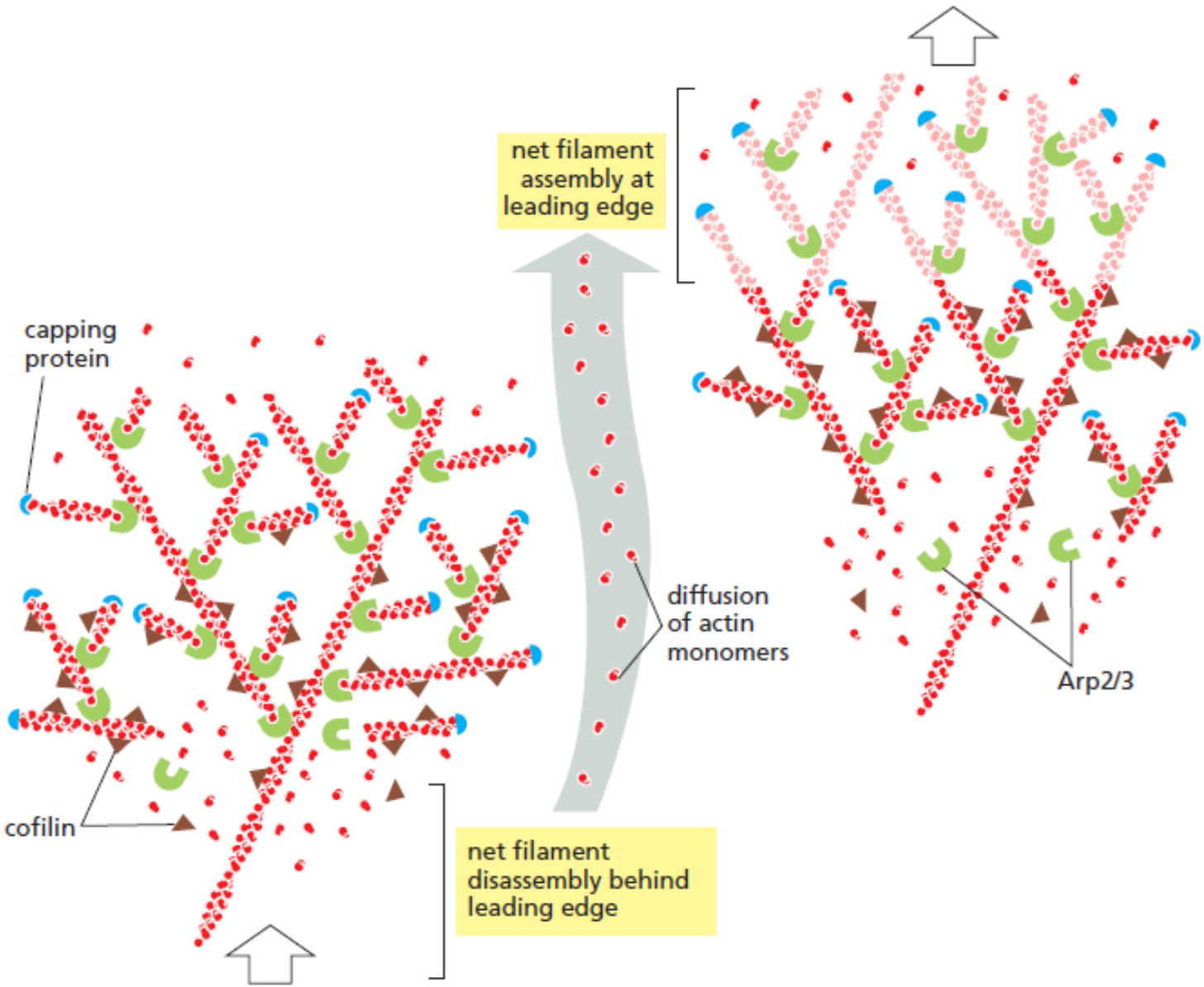
Protrusión del lamellipodio



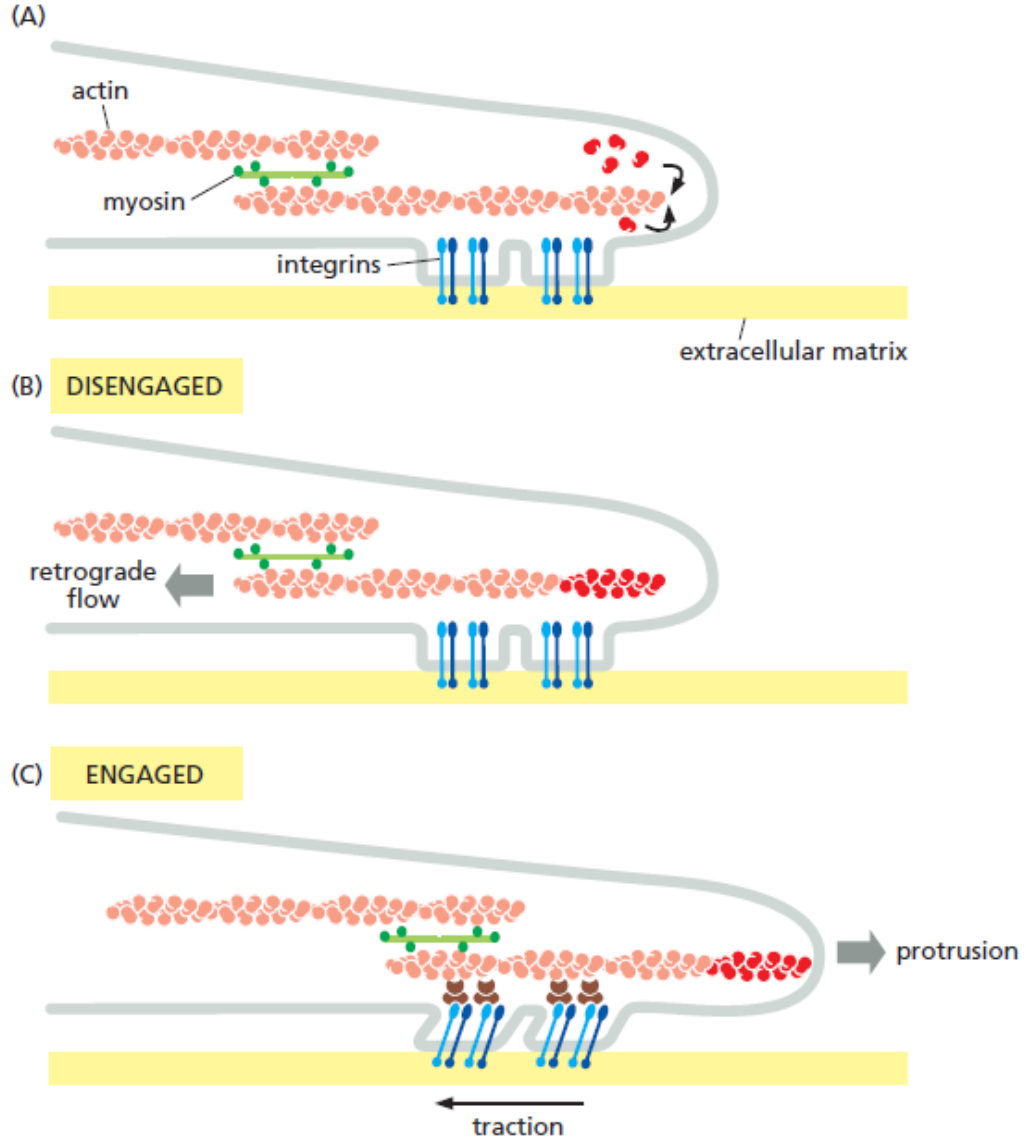
Arp2/3 en borde líder
Actina



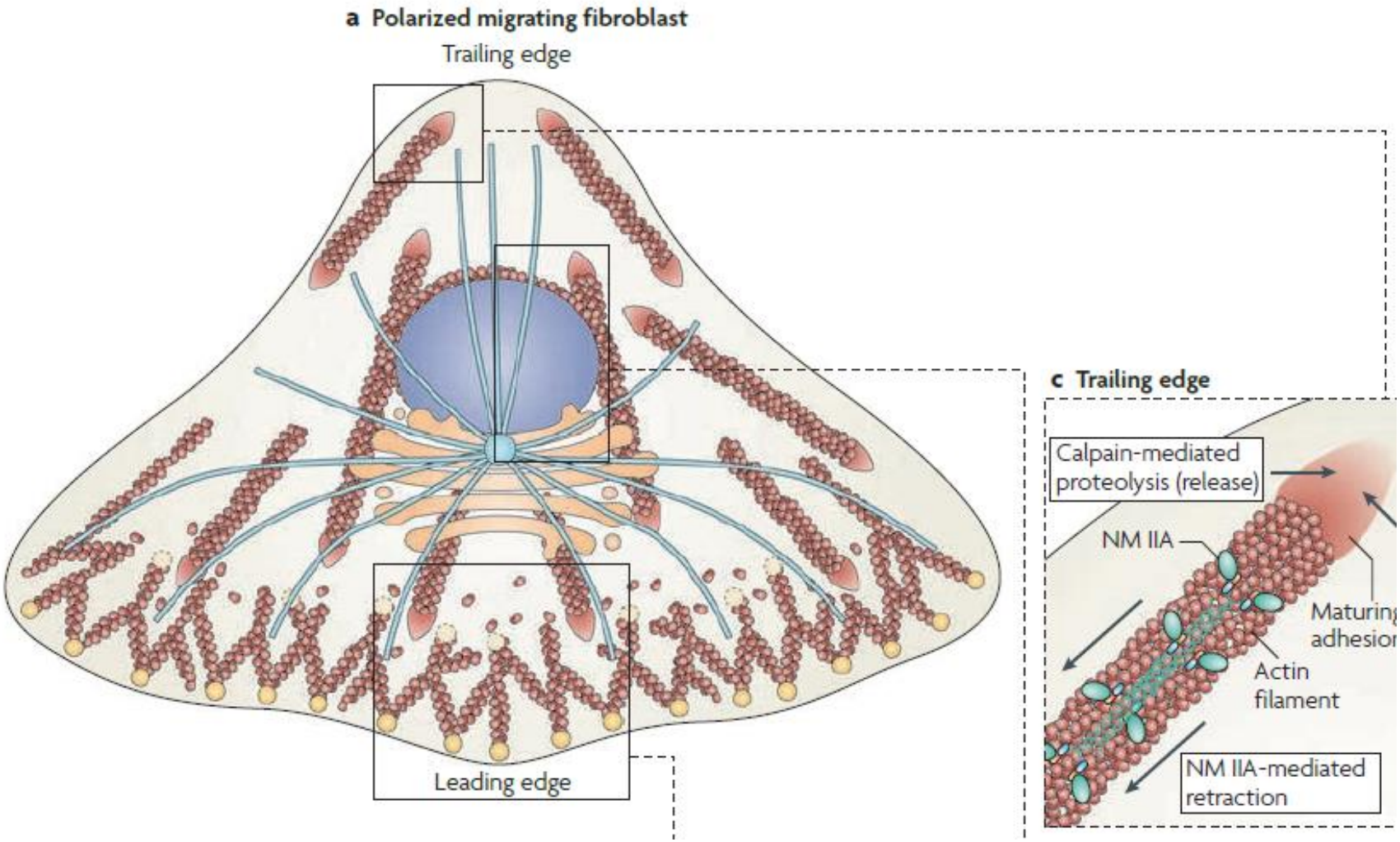
Protrusión del lamellipodio



Unión a sustrato – adhesión a la matriz extracelular



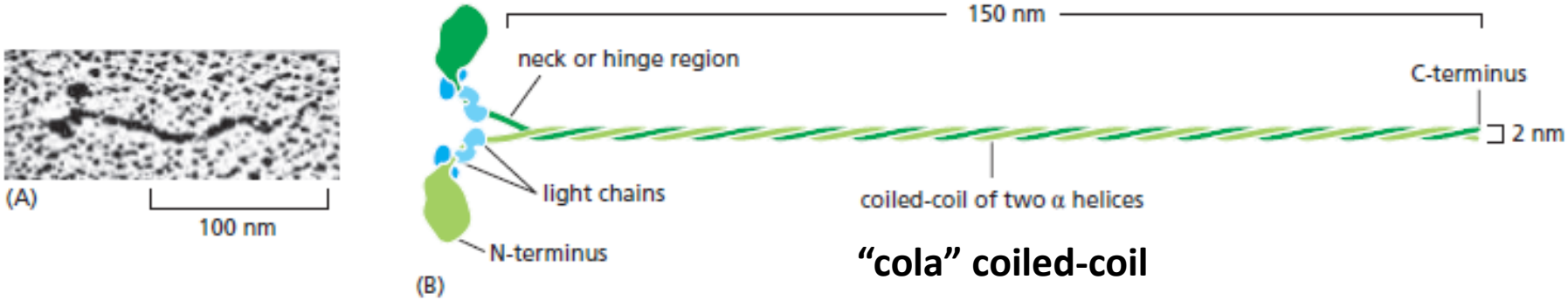
Contracción por fibras de tensión de actina y miosina llevan a la retracción del borde posterior



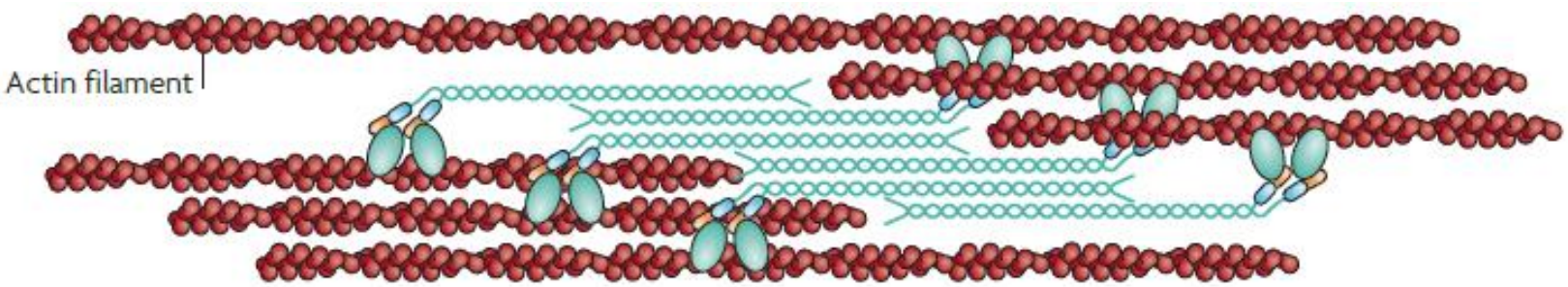
Desensamblaje de adhesiones posteriores

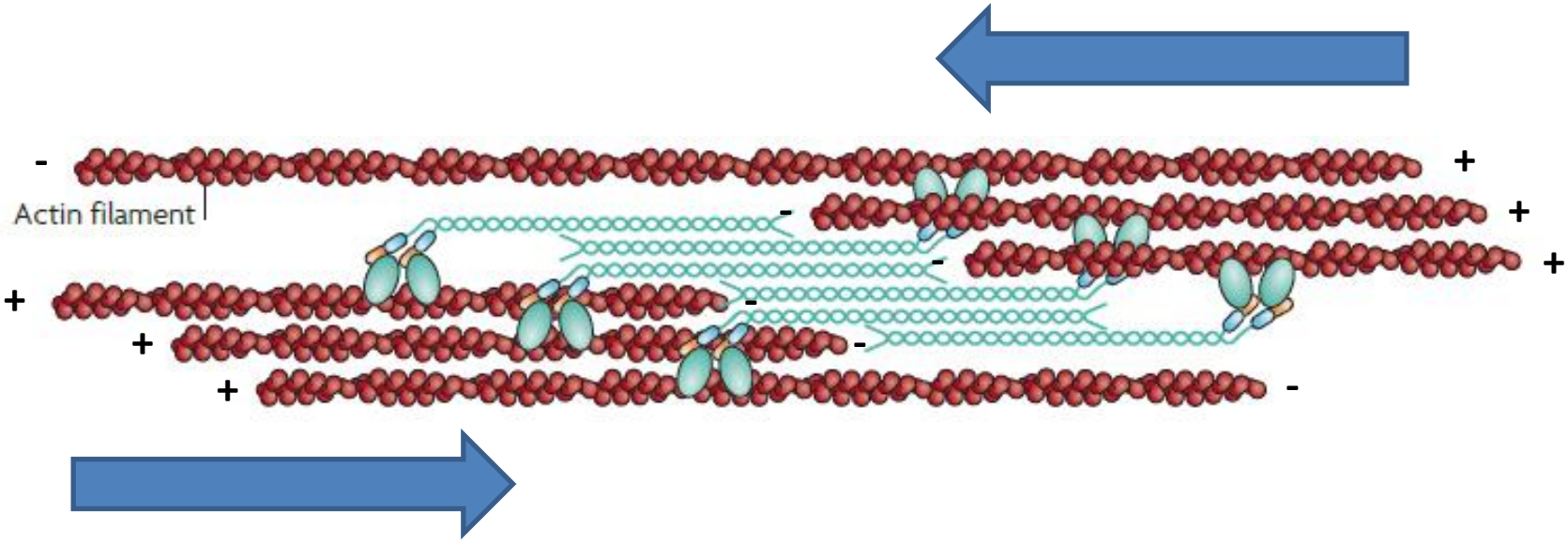
Miosina II forma haces que se intercalan con los filamentos de actina en fibras de tensión (y en el músculo)

“cabezas” globulares

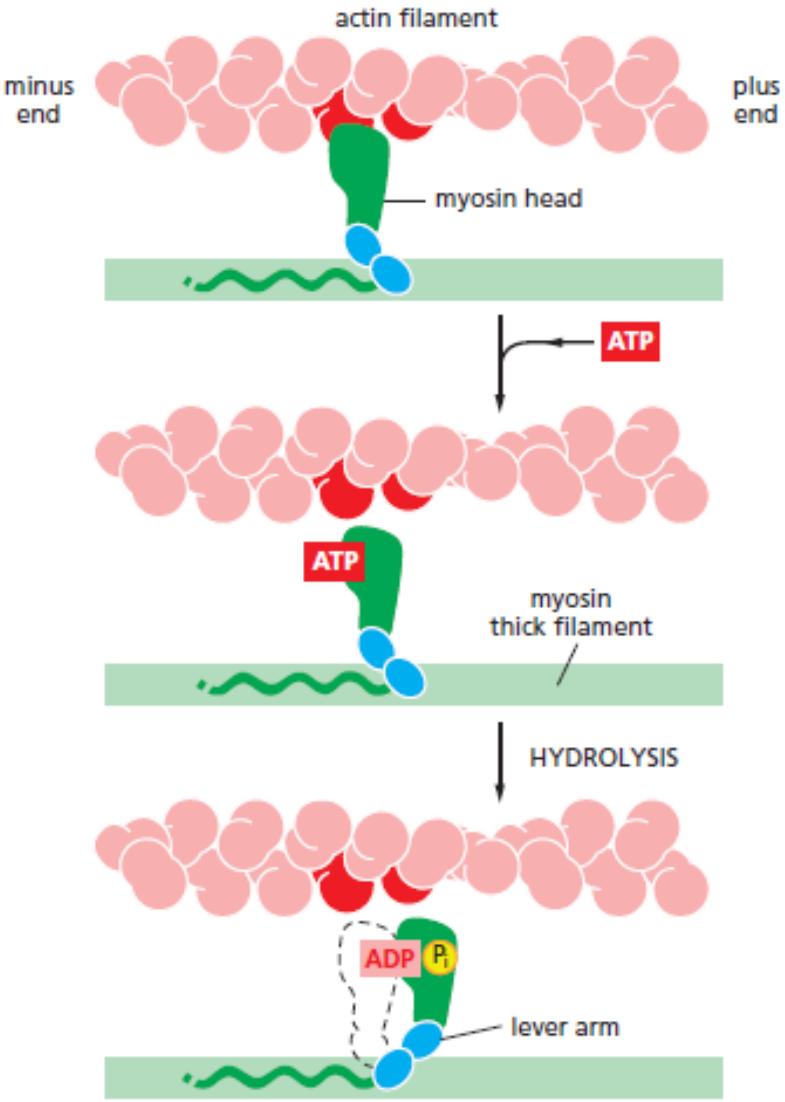


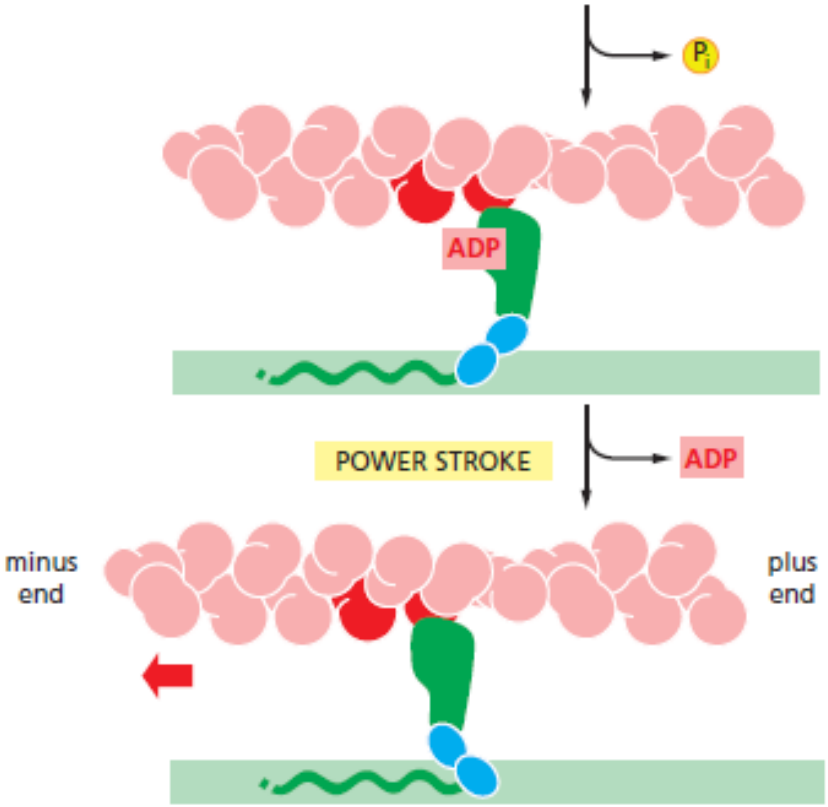
“cola” coiled-coil

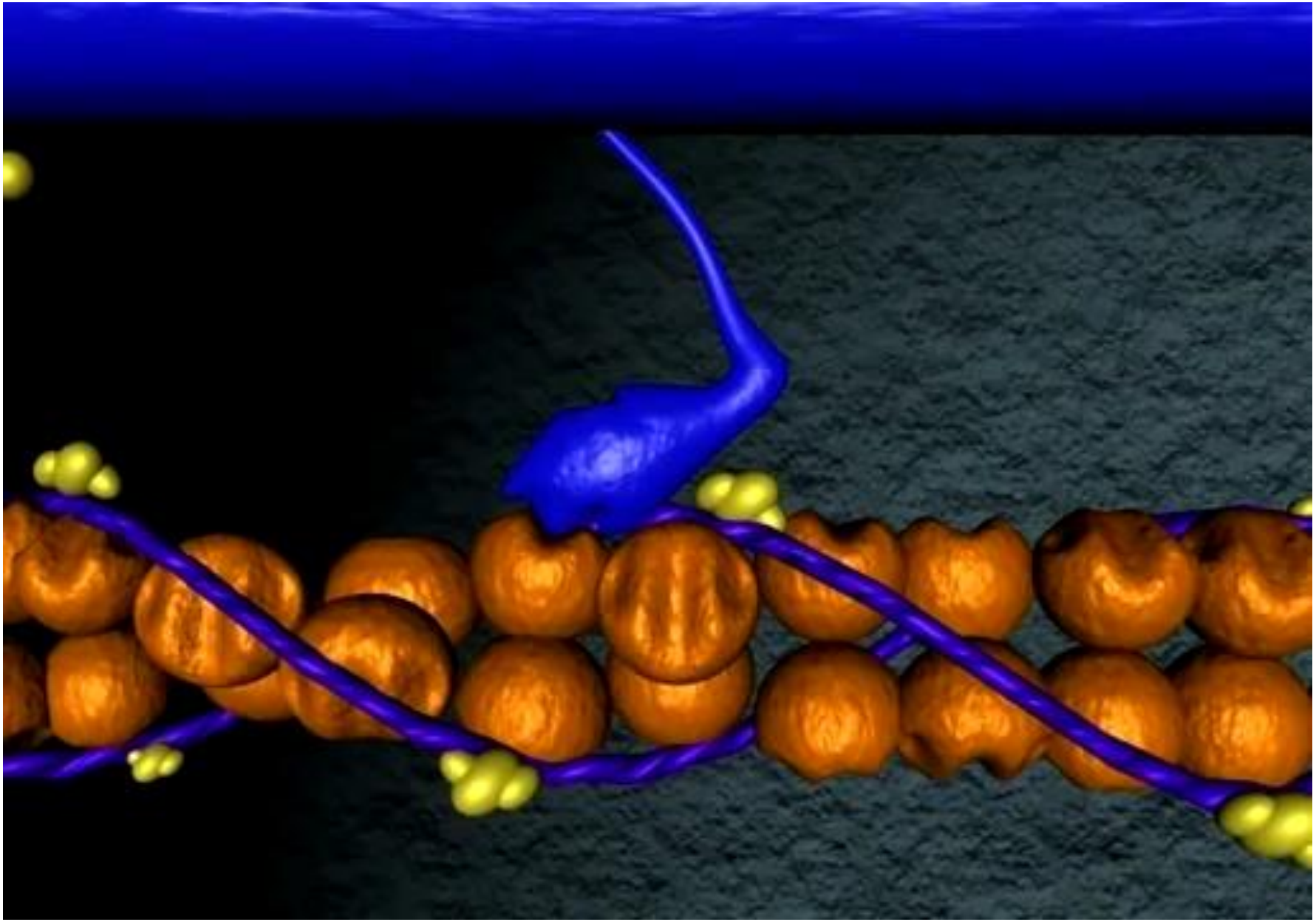



Actin filament
-
+
+
+
+
+
+
-
-
-
-
-

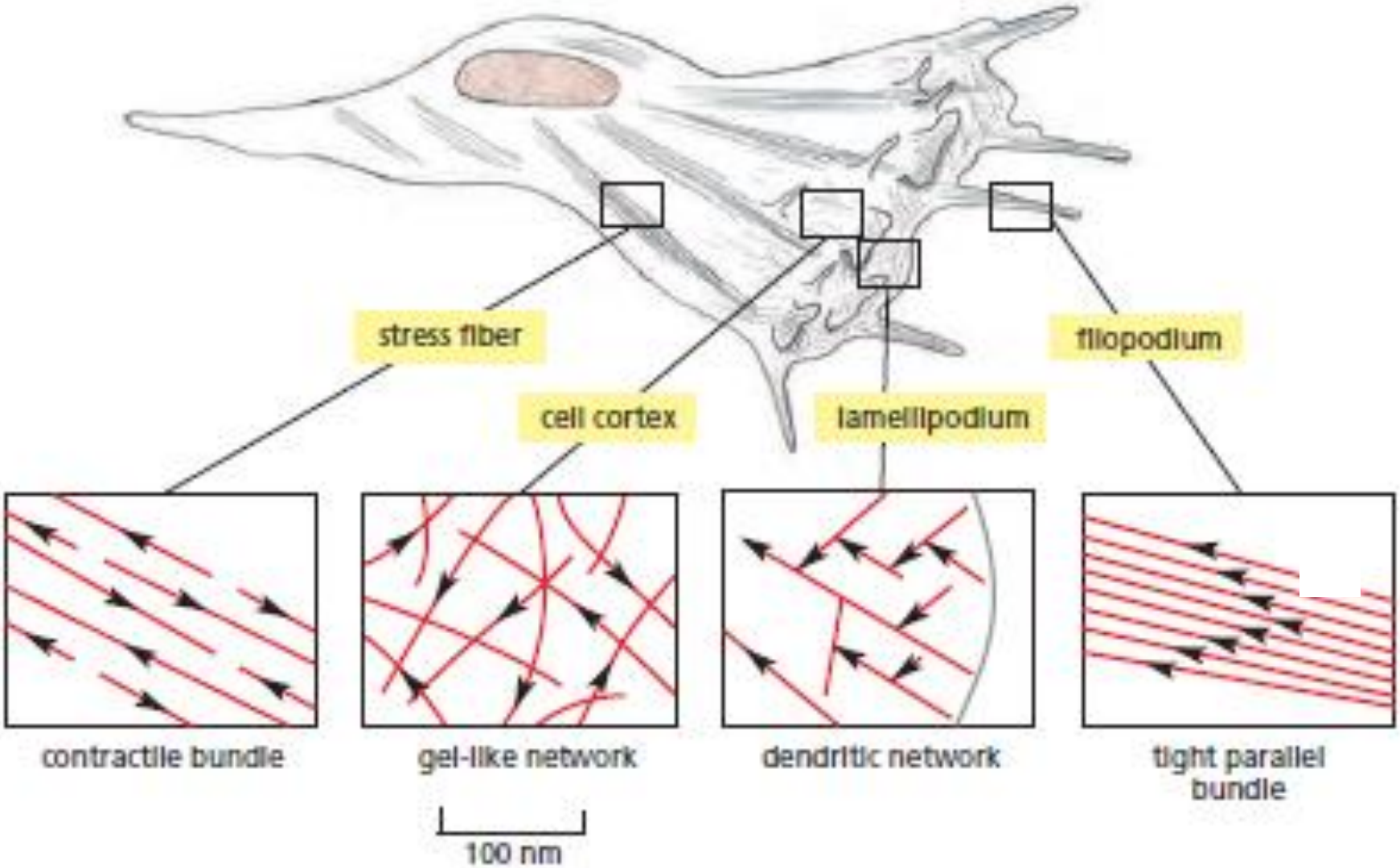
Generación de fuerza por miosina II sobre filamentos de actina







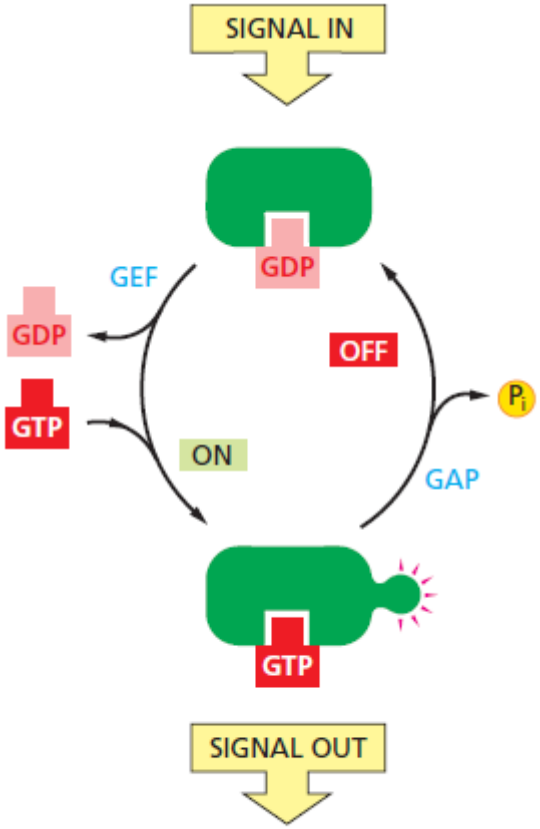
https://www.youtube.com/watch?v=gJ309LfHQ3M&feature=youtu.be


stress fiber
cell cortex
lamellipodium
filopodium
contractile bundle
gel-like network
dendritic network
tight parallel bundle
100 nm

¿Cómo regula la célula todas estas actividades durante la migración?

¿Cómo se mantiene la polaridad de la célula?

GTPasas como interruptores moleculares

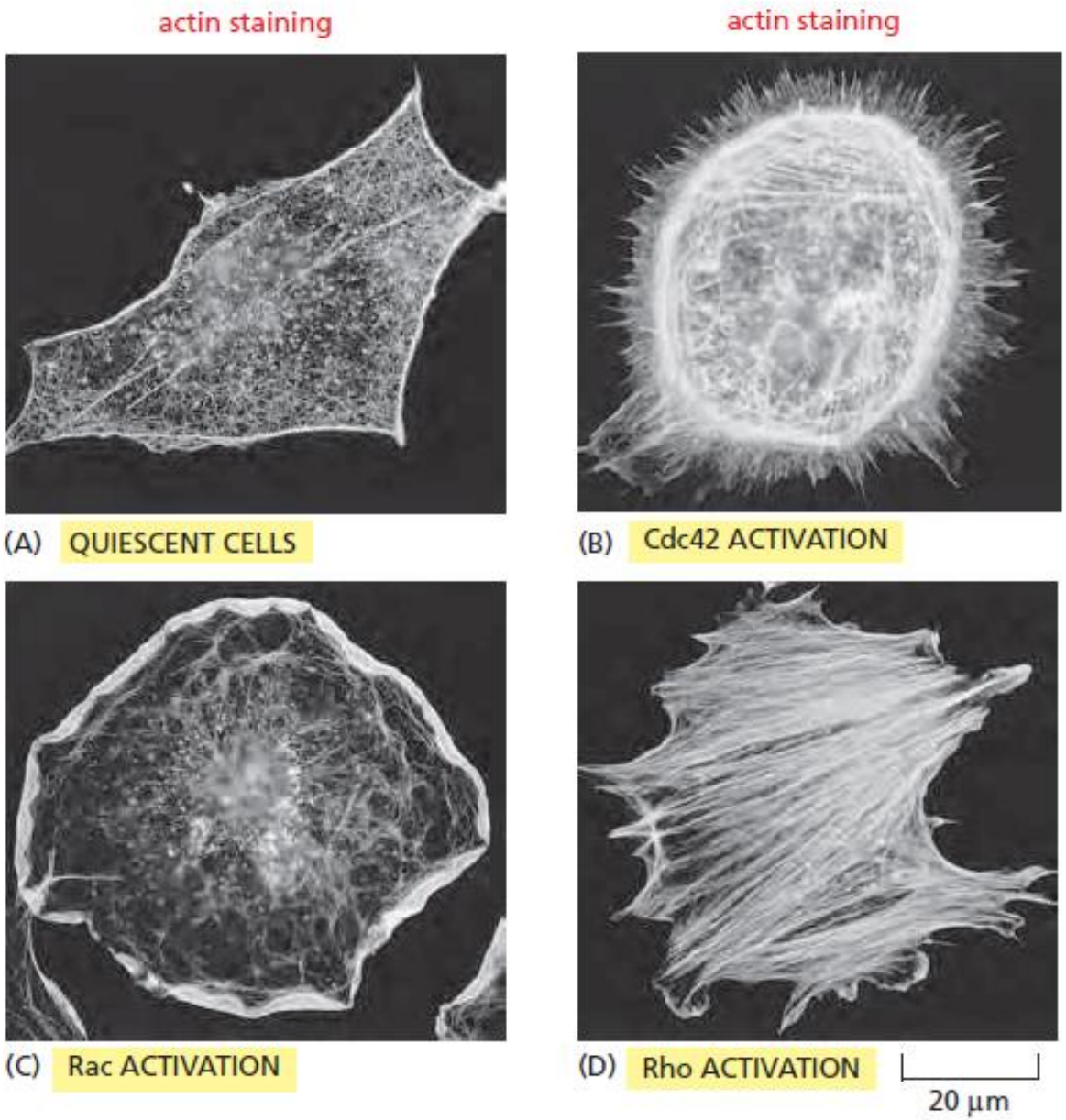


SIGNALING BY GTP-BINDING PROTEIN

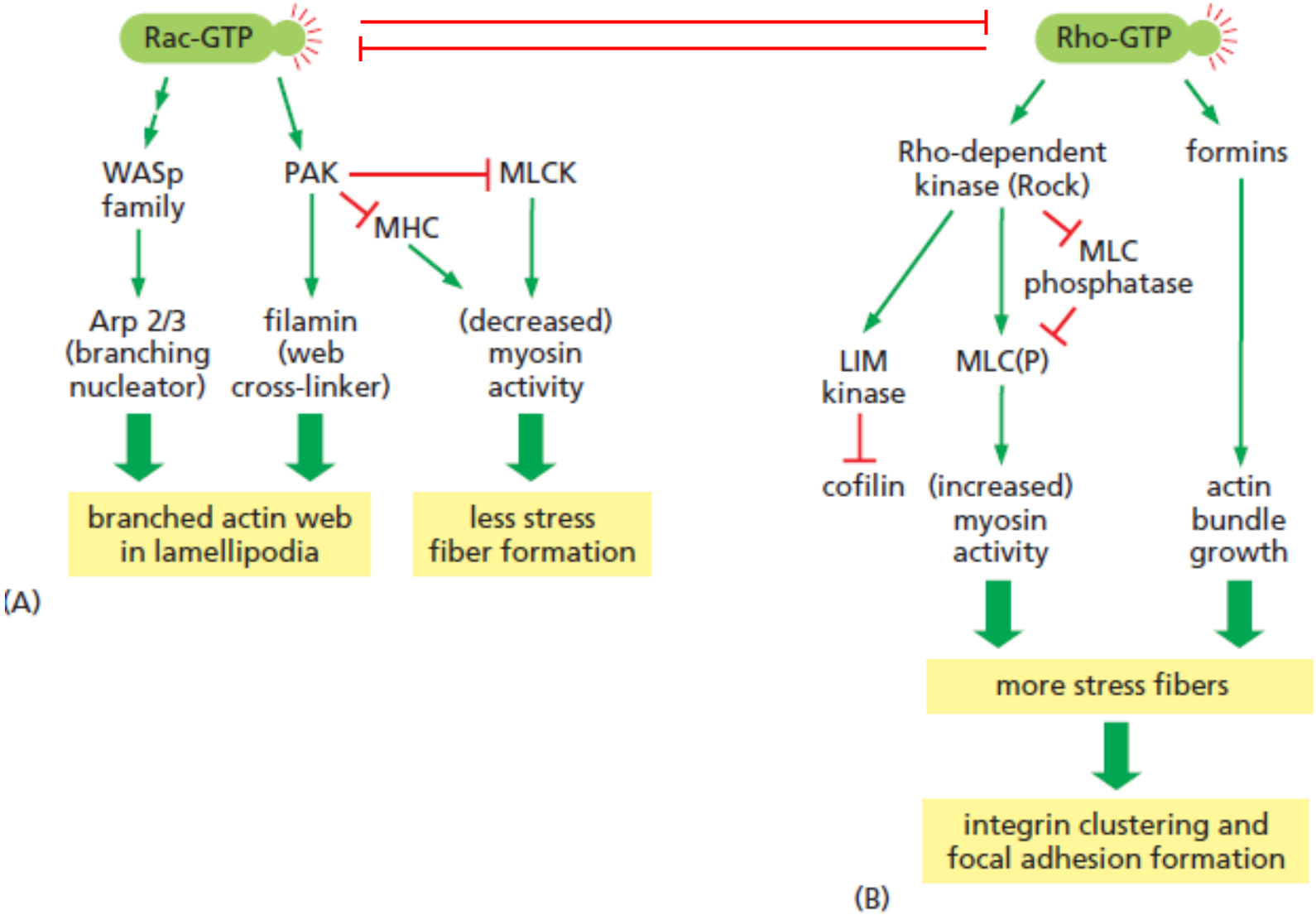
TABLE 15–5 The Ras Superfamily of Monomeric GTPases

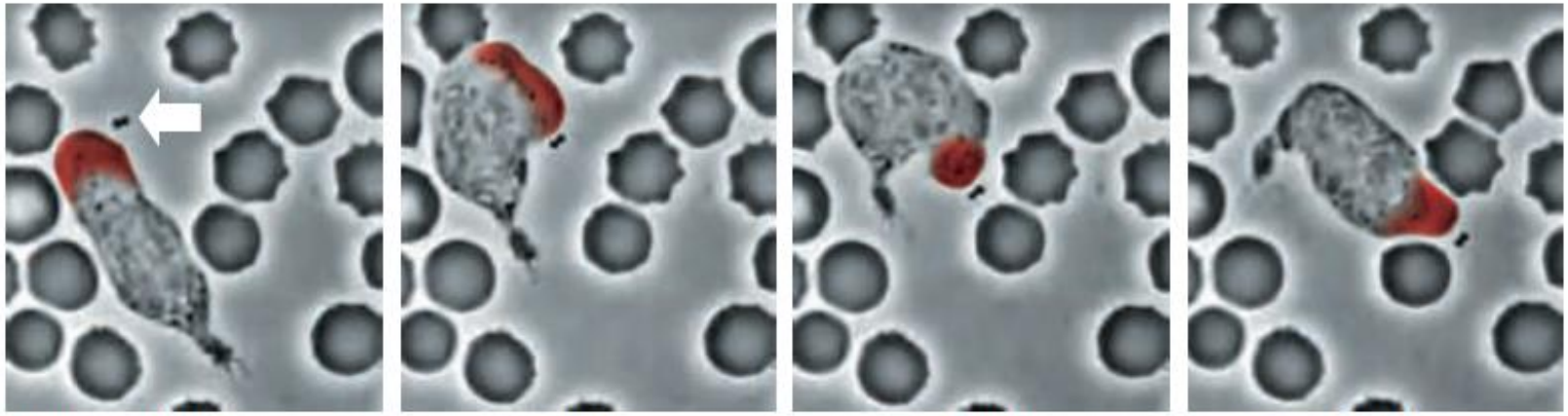
Family	Some family members	Some functions
Ras	H-Ras, K-Ras, N-Ras	Relay signals from RTKs
	Rheb	Activates mTOR to stimulate cell growth
	Rap1	Activated by a cyclic-AMP-dependent GEF; influences cell adhesion by activating integrins
Rho*	Rho, Rac, Cdc42	Relay signals from surface receptors to the cytoskeleton and elsewhere
ARF*	ARF1–ARF6	Regulate assembly of protein coats on intracellular vesicles
Rab*	Rab1–60	Regulate intracellular vesicle traffic
Ran*	Ran	Regulates mitotic spindle assembly and nuclear transport of RNAs and proteins

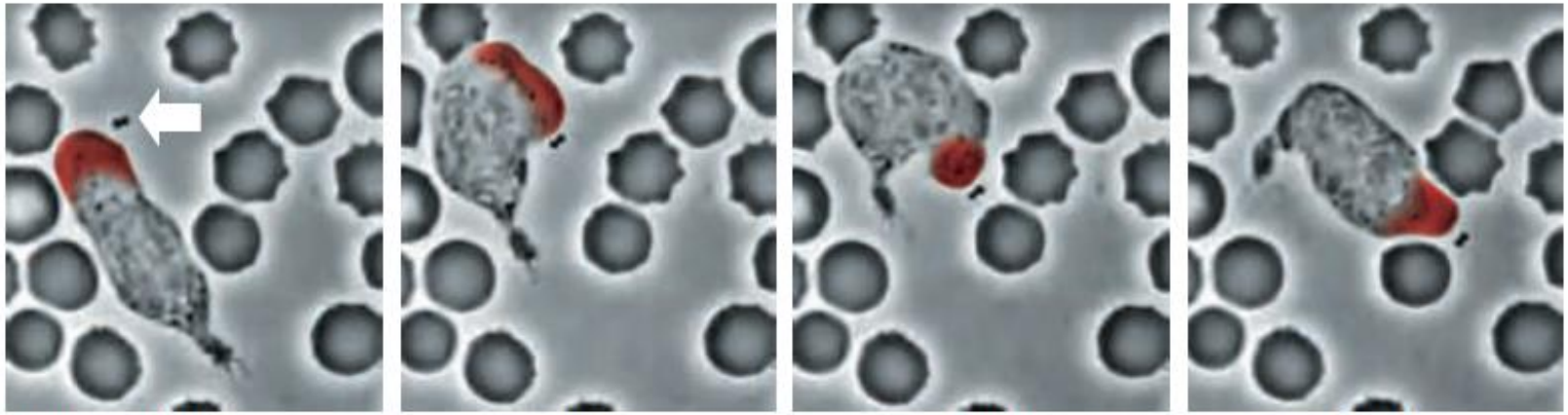
*The Rho family is discussed in Chapter 16, the ARF and Rab proteins in Chapter 13, and Ran in Chapters 12 and 17. The three-dimensional structure of Ras is shown in Figure 3–67.

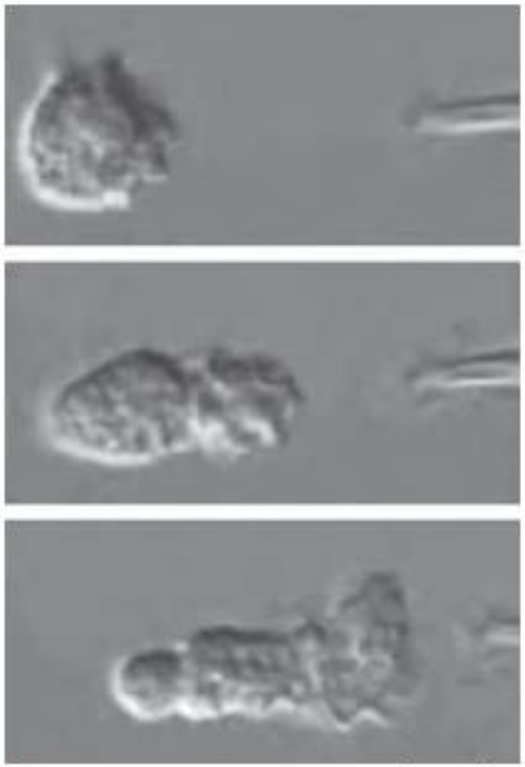


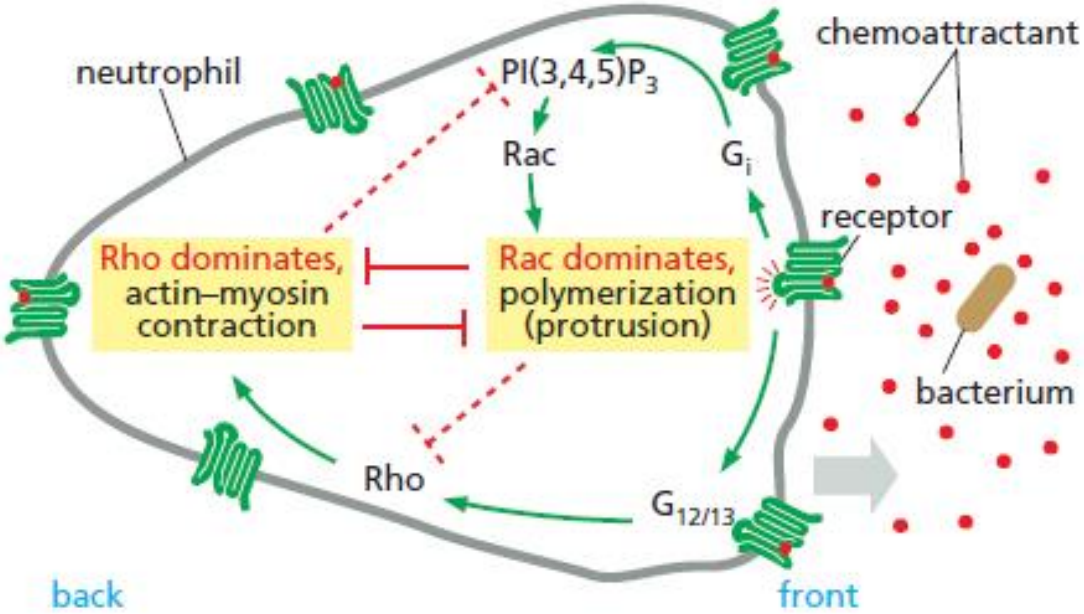
¿Qué tipo de estructuras son promovidas por cada GTPasa?


Rac-GTP
Rho-GTP
WASp family
PAK
MLCK
MHC
Arp 2/3 (branching nucleator)
filamin (web cross-linker)
(decreased) myosin activity
branched actin web in lamellipodia
less stress fiber formation
(A)
Rho-dependent kinase (Rock)
formins
MLC phosphatase
LIM kinase
MLC(P)
cofilin
(increased) myosin activity
actin bundle growth
more stress fibers
integrin clustering and focal adhesion formation
(B)

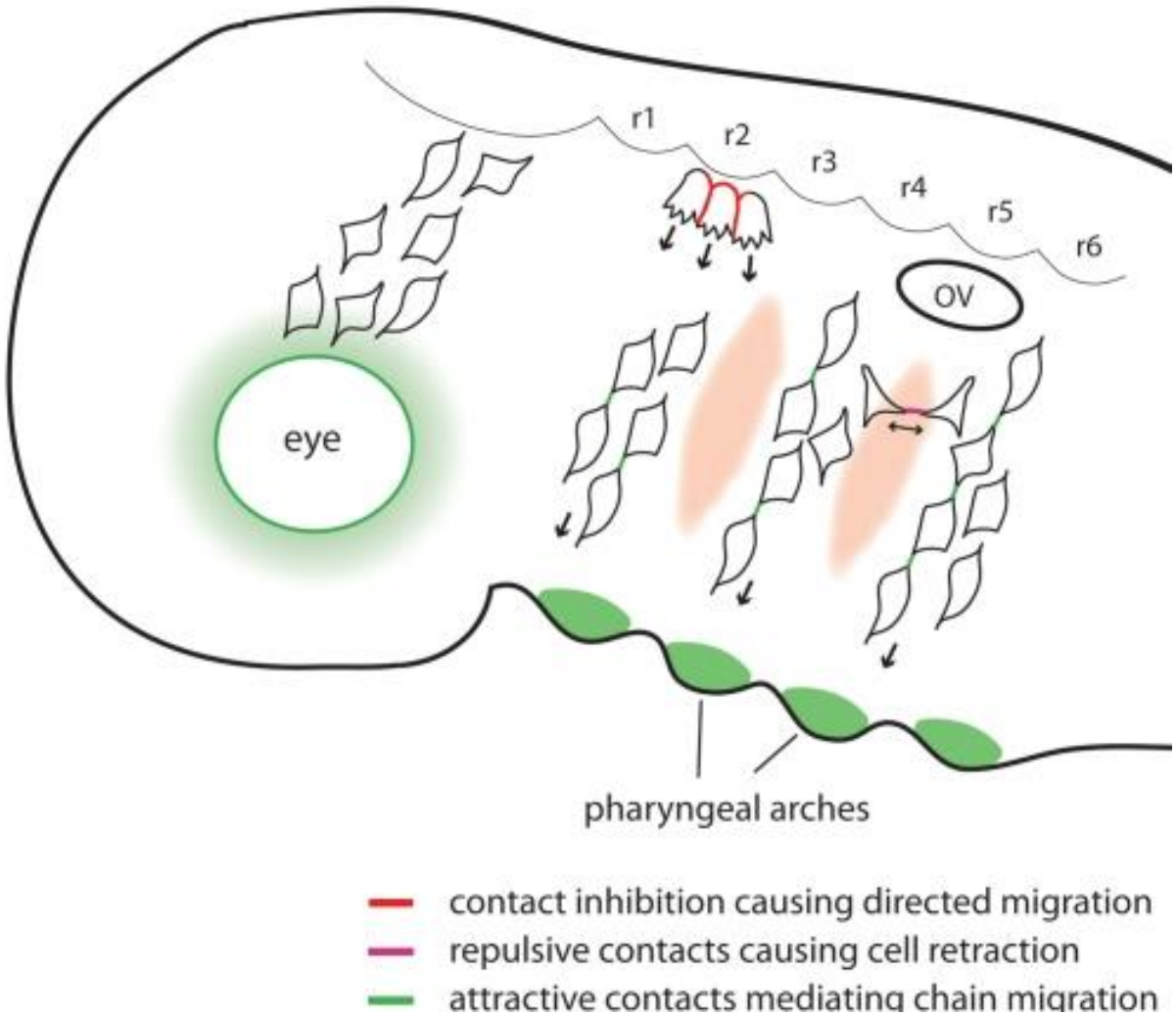

time
0 min
1 min
2 min
3 min


time
0 min
1 min
2 min
3 min


(A)
5 μm


neutrophil
chemoattractant
PI(3,4,5)P3
Rac
Gi
receptor
Rho dominates, actin–myosin contraction
Rac dominates, polymerization (protrusion)
bacterium
Rho
G12/13
back
front
(B)

Combinaciones de señales atractivas y repulsivas durante el desarrollo

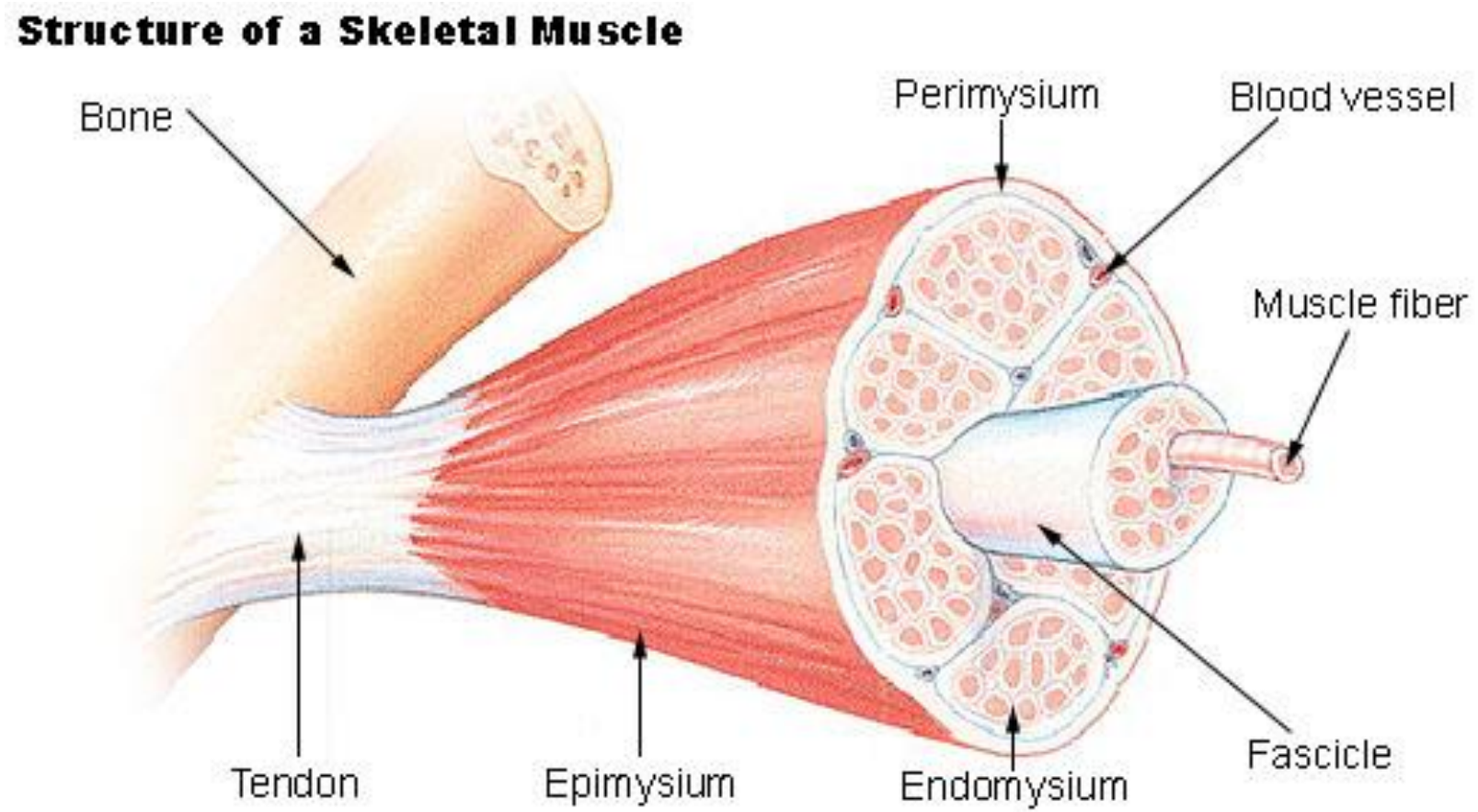


CÉLULAS MUSCULARES

Células especializadas para la contracción

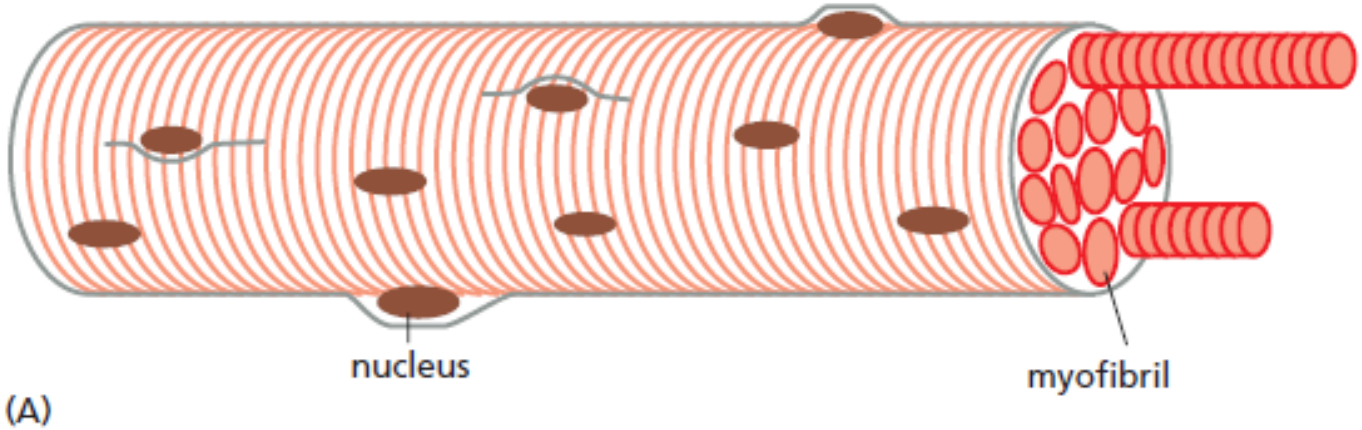
Estructuras altamente organizadas de filamentos de actina + motores tipo miosina

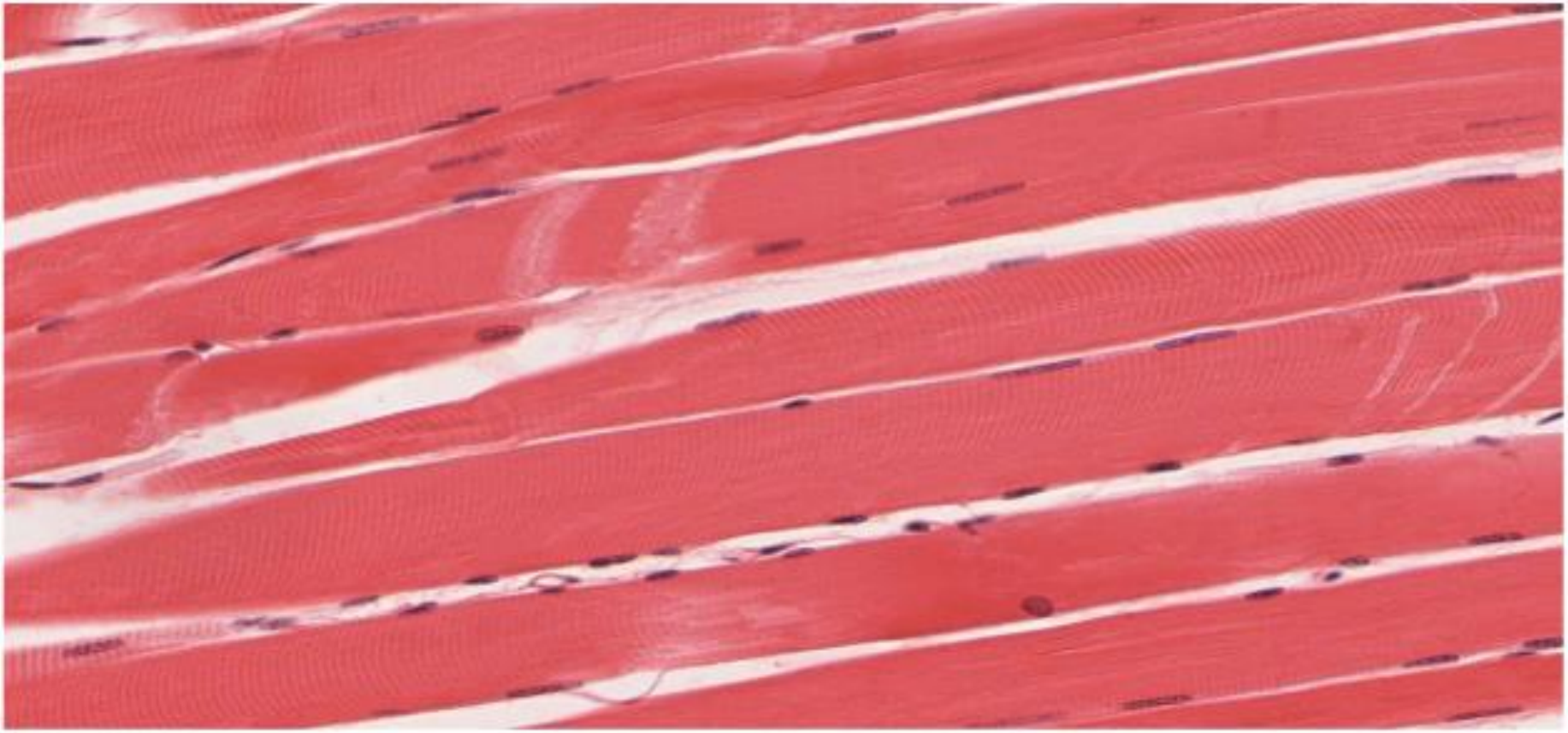
Músculo esquelético – fibras sincitiales

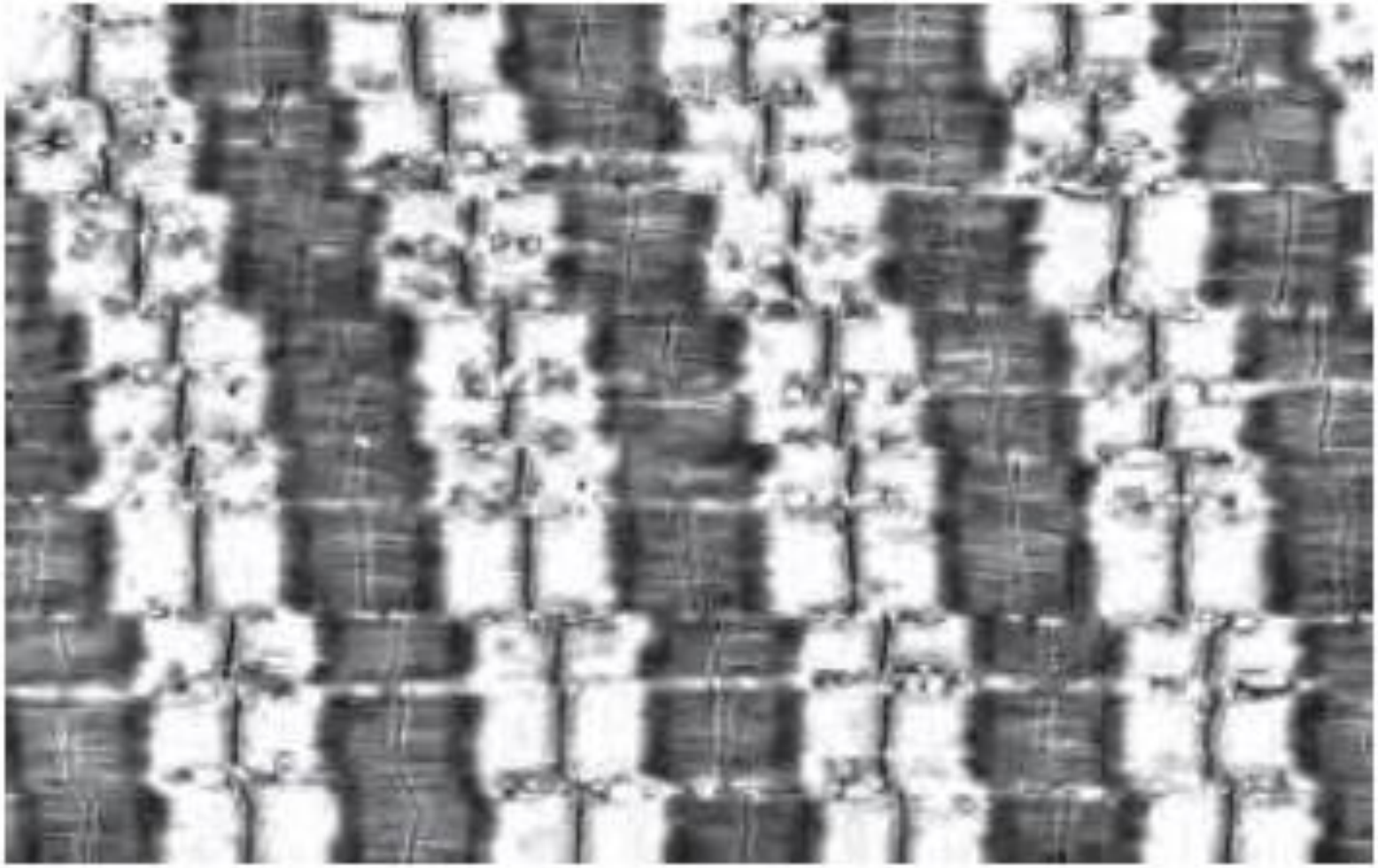


Músculo esquelético – fibras sincitiales

Figure 16–31 **Skeletal muscle cells (also called muscle fibers).** (A) These huge multinucleated cells form by the fusion of many muscle cell precursors, called myoblasts. Here, a single muscle cell is depicted. In an adult human, a muscle cell is typically 50 μm in diameter and can be up to several centimeters long. (B) Fluorescence micrograph of rat muscle, showing the peripherally located nuclei *(blue)* in these giant cells. Myofibrils are stained *red*. (B, courtesy of Nancy L. Kedersha.)







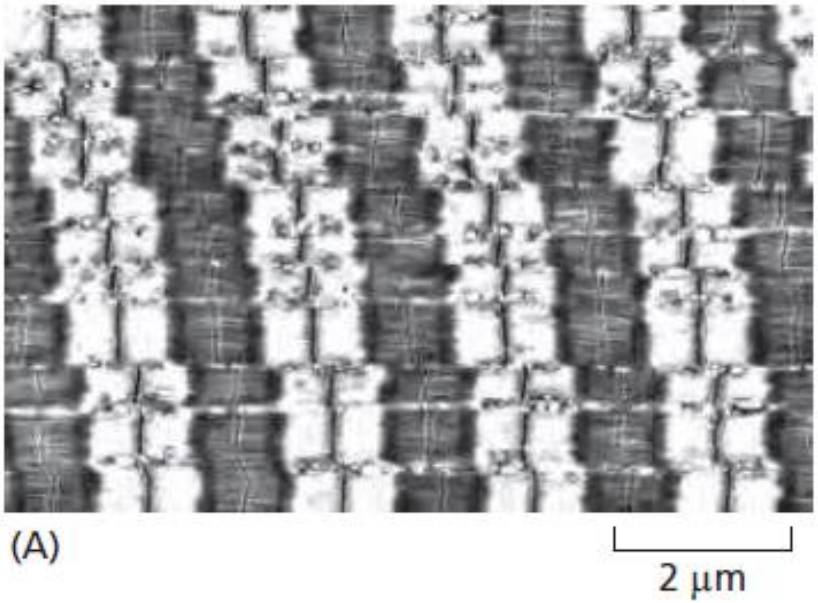
(A)

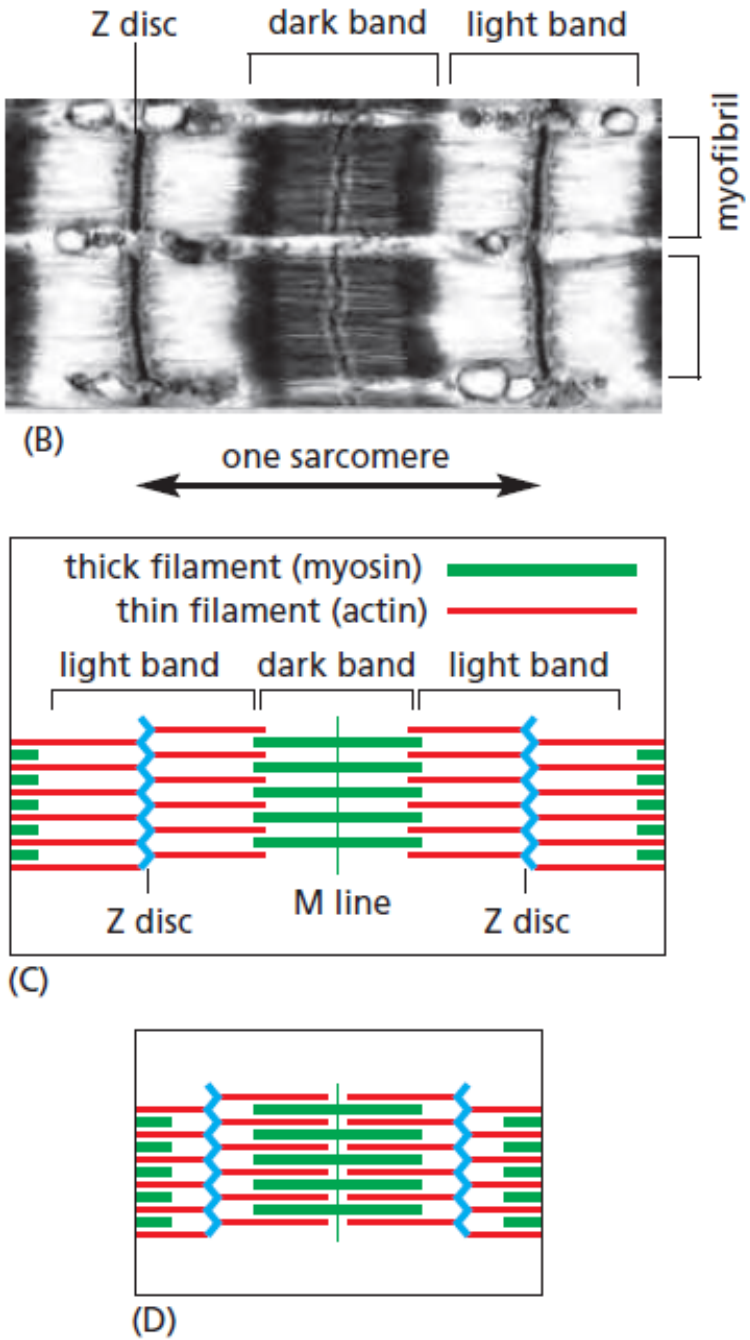
Banda I (lúcida)

Banda A (densa)

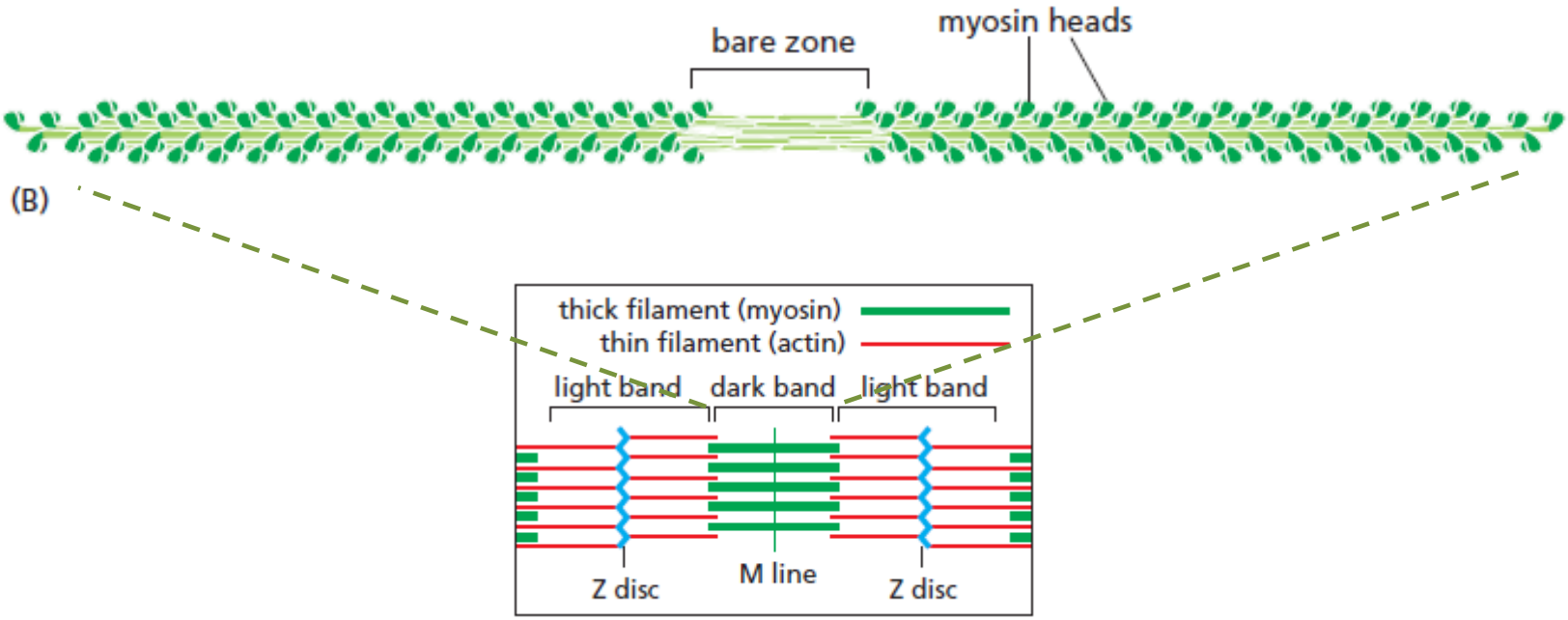
2 μm

Organización en <u>miofibrillas</u> = repeticiones de <u>sarcómeros</u> (unidades contráctiles)

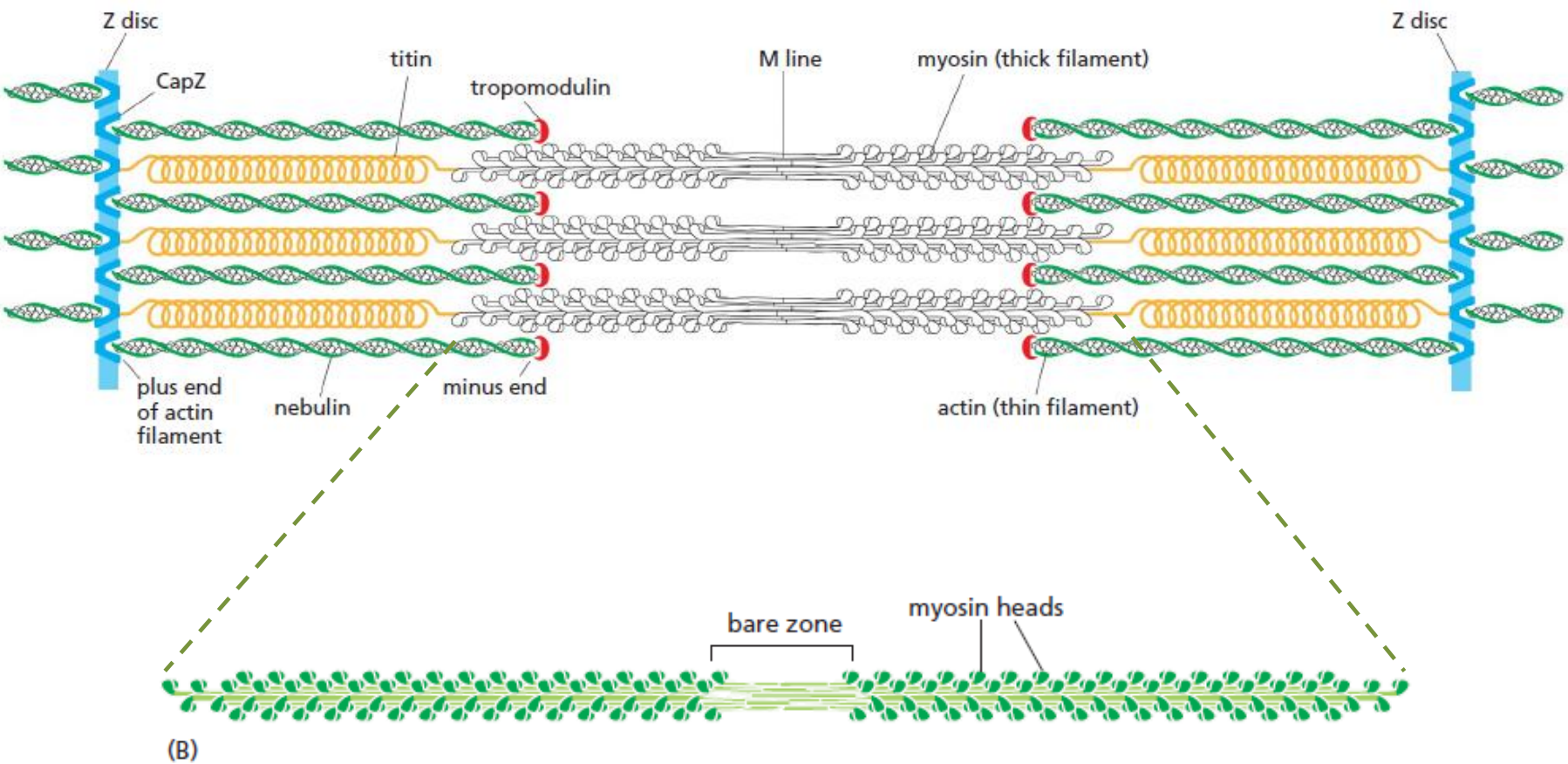


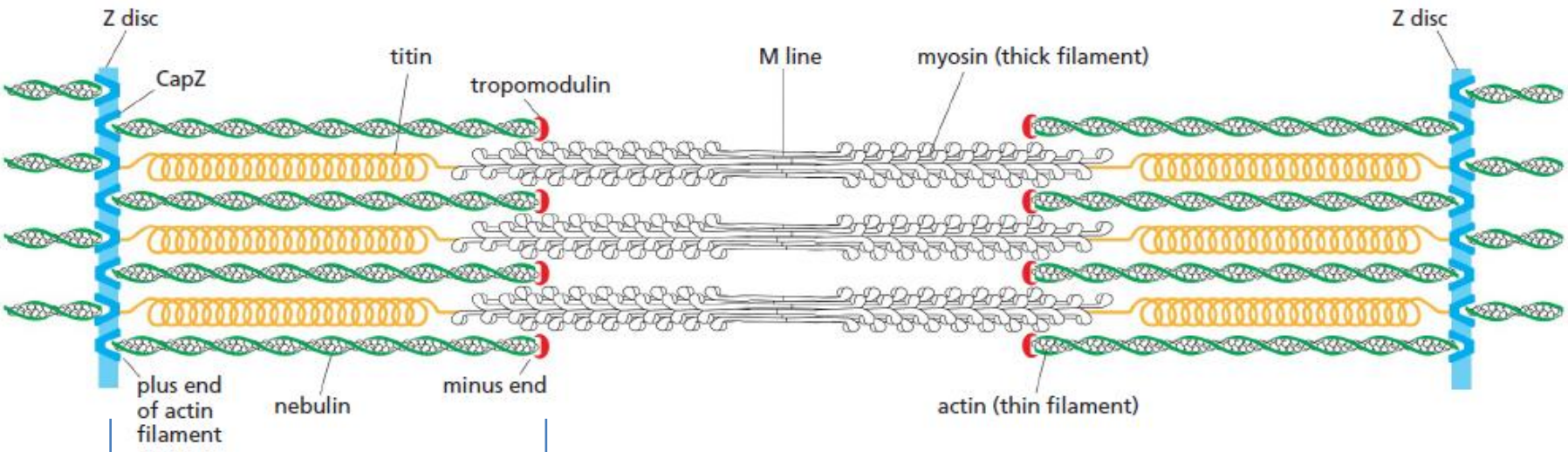


Miosina II en músculo esquelético



Organización del sarcomero – filamentos de actina, miosina, y proteínas accesorias

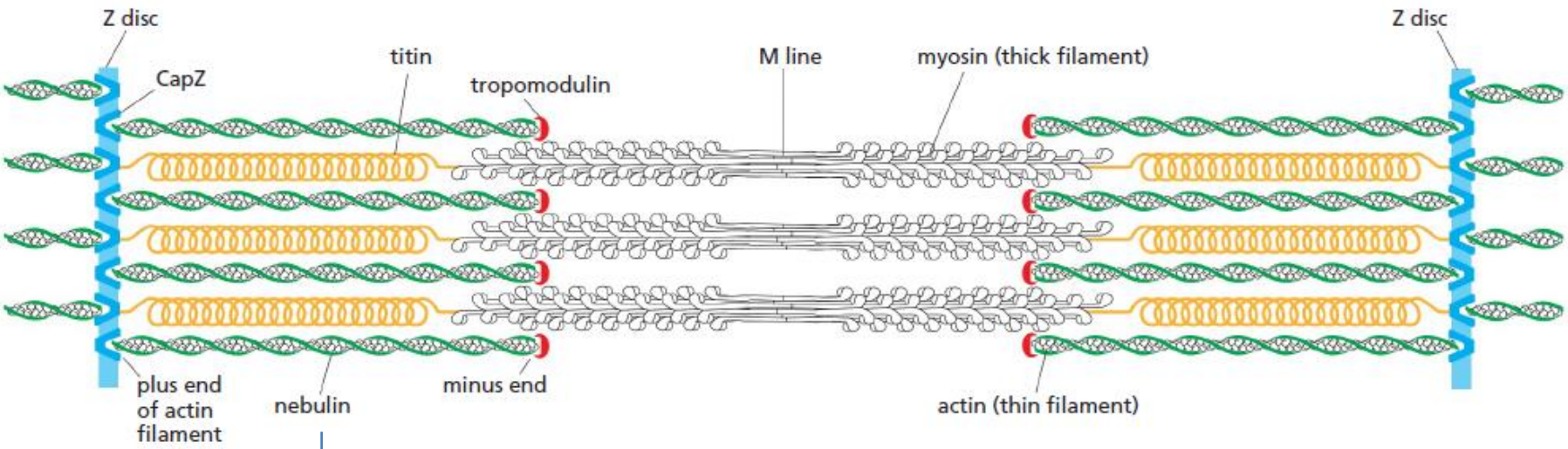




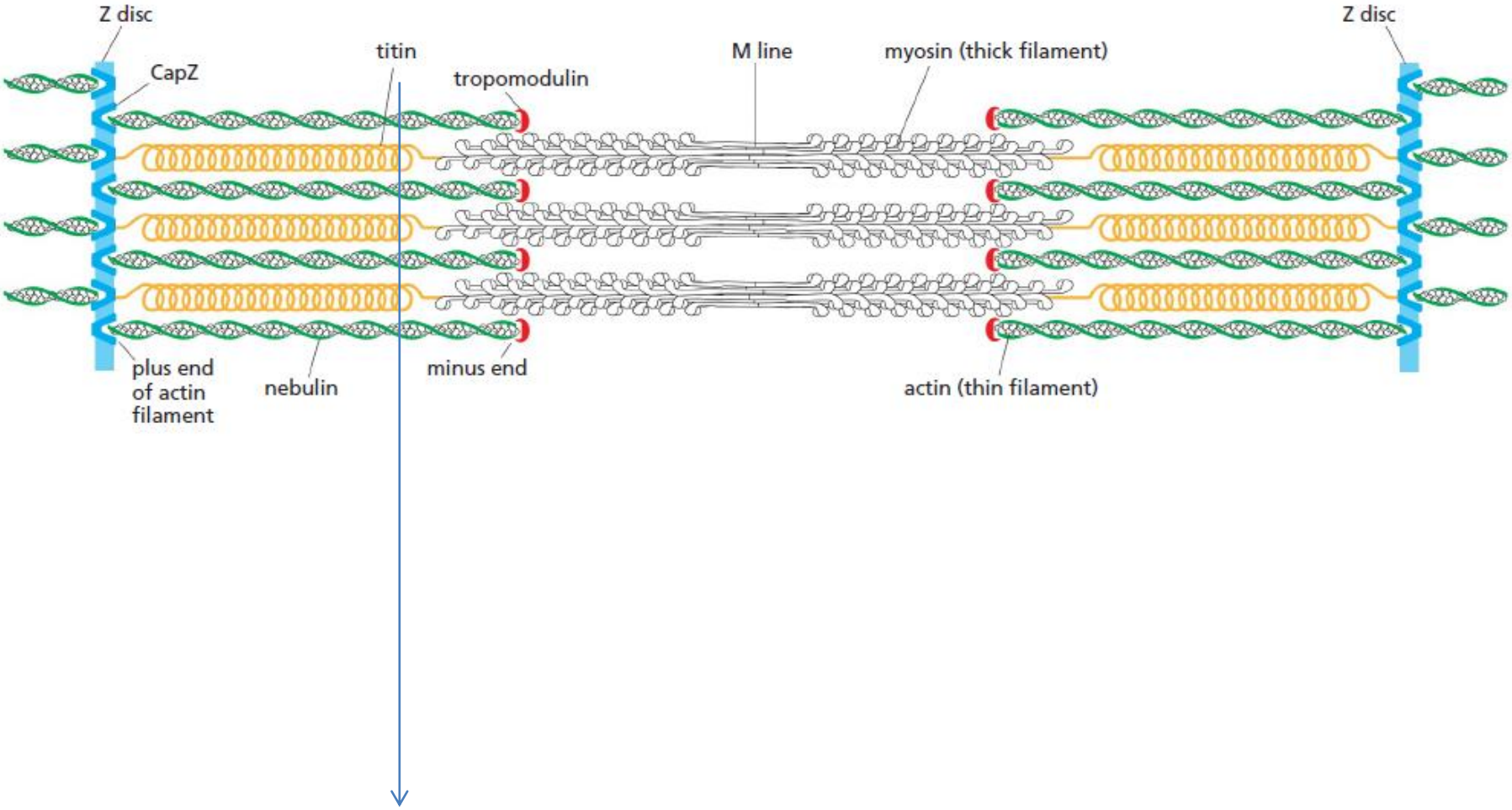
Discos Z: CapZ unido a extremo (+) de filamento de actina, impide despolimerización
α-actinina da separación regular

Tropomodulina unida a extremo (-) impide despolimerización

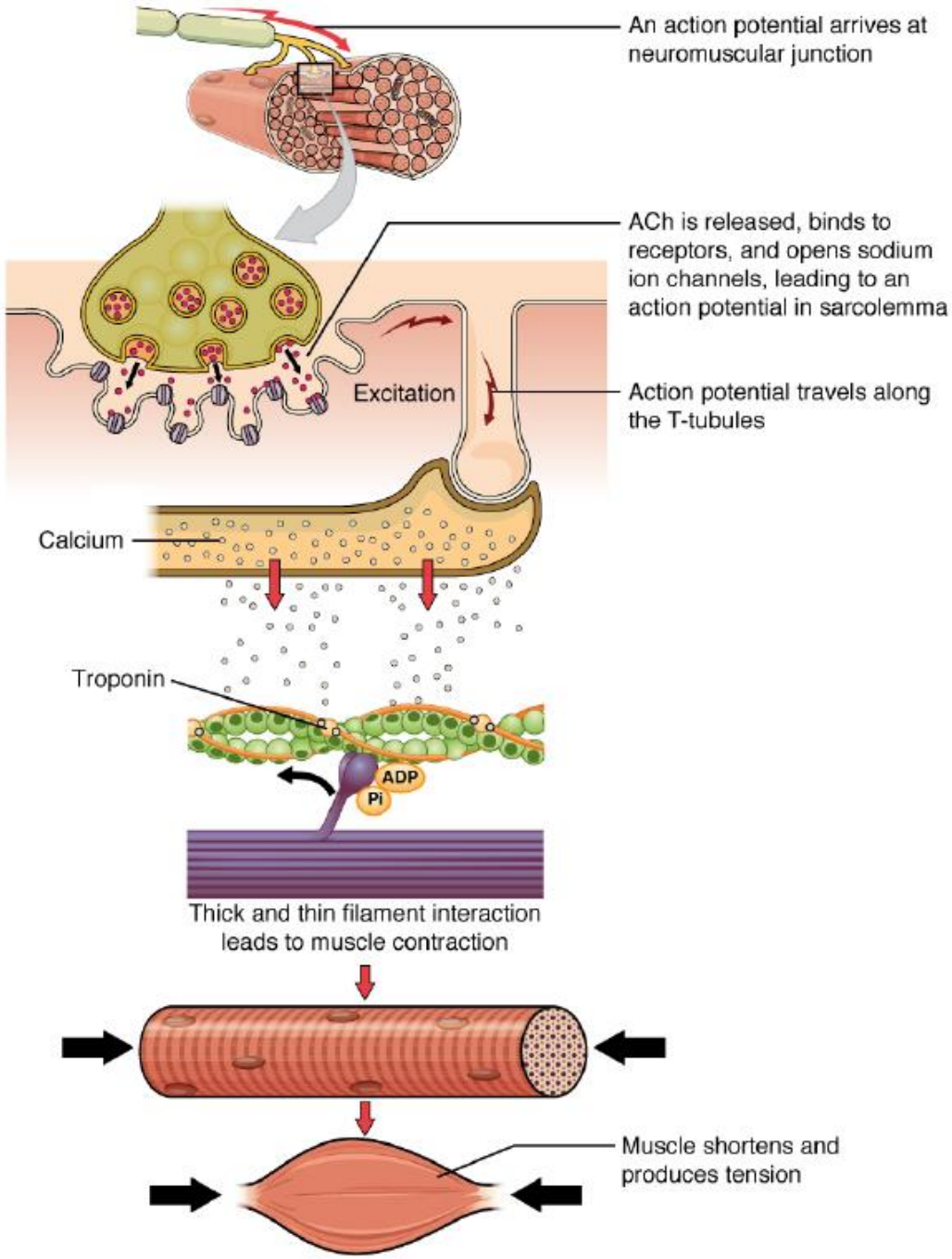
Si bien hay recambio de subunidades de actina, tiempos de recambio son **muy** largos (días!) comparados con microfilamentos en otras células



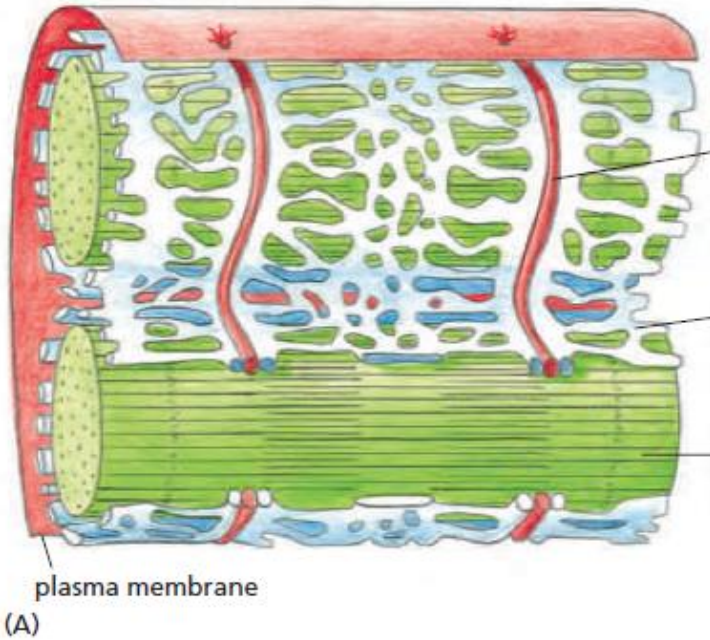
Nebulina: proteína gigante (900 KDa)
Repetición de dominio de unos 35 aminoácidos, genera “regla” que da largo de microfilamentos en sarcómero



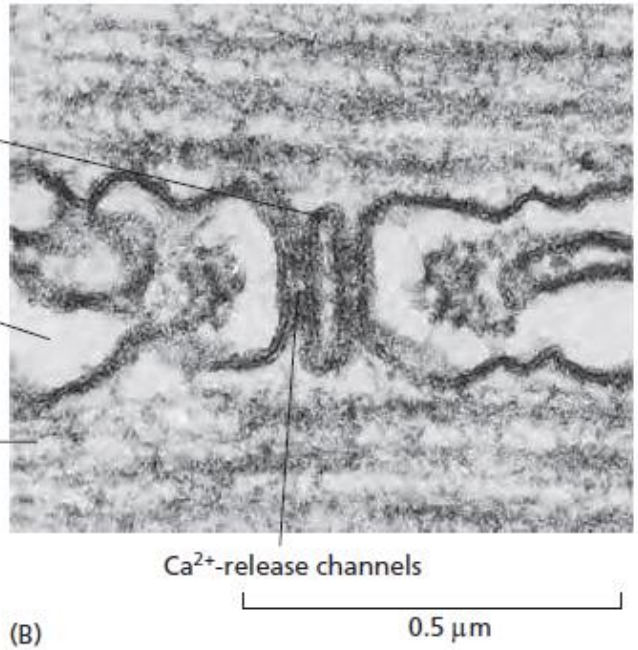
Titina: proteína gigante, “resorte molecular” con repetidos que se pliegan y despliegan según tensión. Une filamentos gruesos de miosina y línea Z, manteniéndolos centrados, también posiblemente “regla” que da largo total del sarcomero

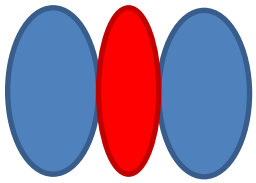

An action potential arrives at neuromuscular junction
ACh is released, binds to receptors, and opens sodium ion channels, leading to an action potential in sarcolemma
Excitation
Action potential travels along the T-tubules
Calcium
Troponin
ADP
Pi
Thick and thin filament interaction leads to muscle contraction
Muscle shortens and produces tension

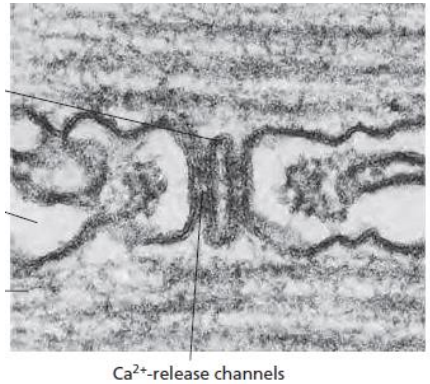
Regulación de la contracción – rol del Ca^{2+}

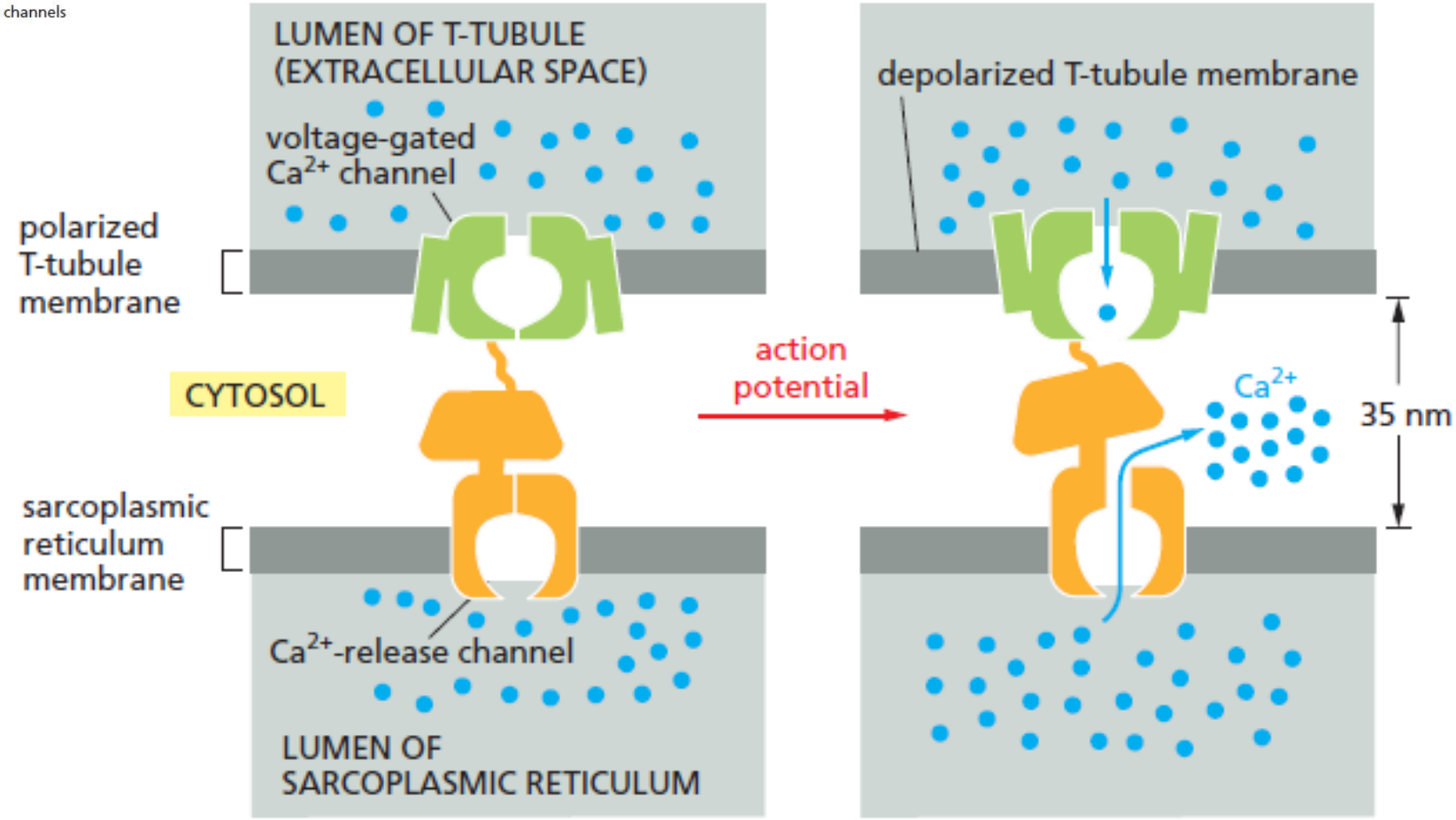


“tríadas”

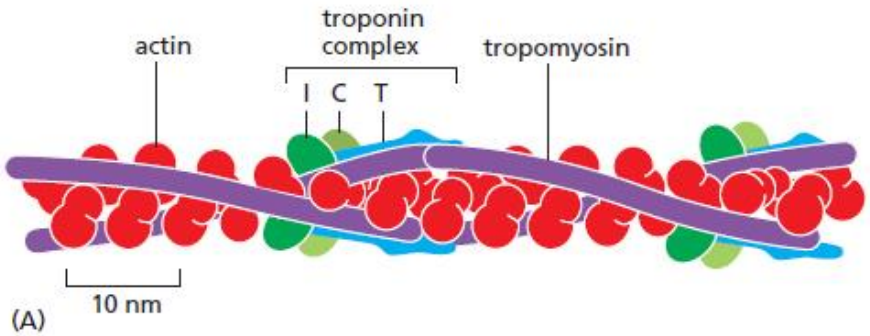


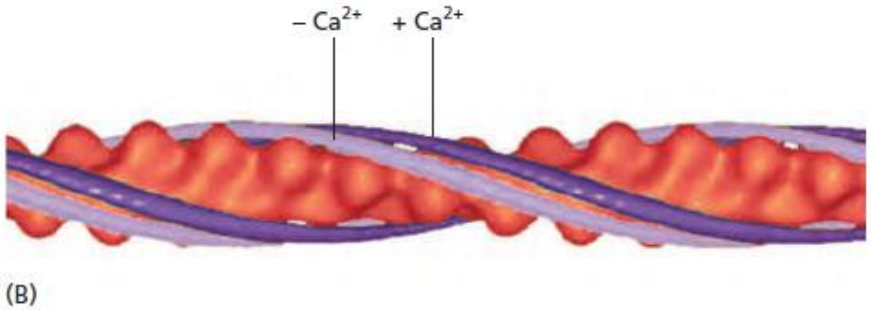


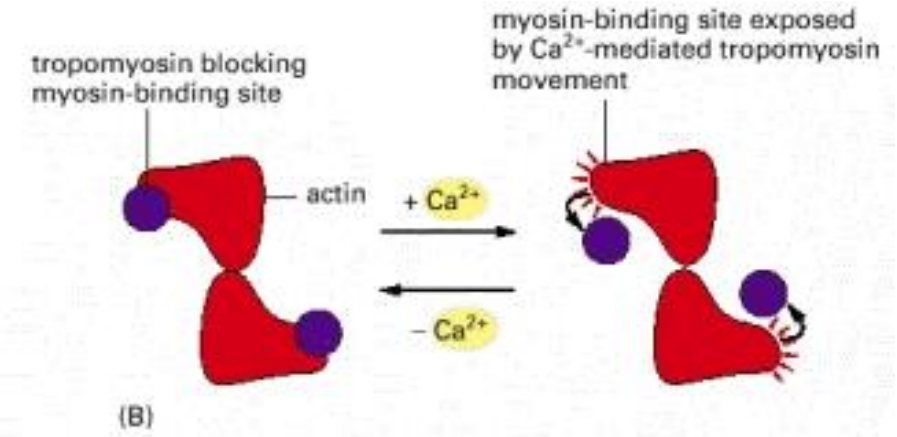

Ca^{2+}-release channels


LUMEN OF T-TUBULE
(EXTRACELLULAR SPACE)
voltage-gated
Ca^{2+} channel
polarized
T-tubule
membrane
CYTOSOL
sarcoplasmic
reticulum
membrane
Ca^{2+}-release channel
LUMEN OF
SARCOPLASMIC RETICULUM
action
potential
depolarized T-tubule membrane
Ca^{2+}
35 nm

La entrada de calcio lleva a que se descubran los sitios de las subunidades de actina que interaccionan con miosina







Sinapsis neuromuscular (acetilcolina en vertebrados)

↓

Potencial de acción en membrana de célula muscular (fibra muscular) que se extiende a túbulos-T

↓

Activación de canal de calcio voltaje dependiente en túbulos-T

↓

Activación de canal de calcio en retículo sarcoplásmico

↓

Aumenta Ca^{++} citosólico

↓

Únión de Ca^{++} a troponina C lleva a cambio conformacional en complejo de troponinas

↓

Desplazamiento de tropomiosina destapa sitios de interacción con miosina en filamentos de actina

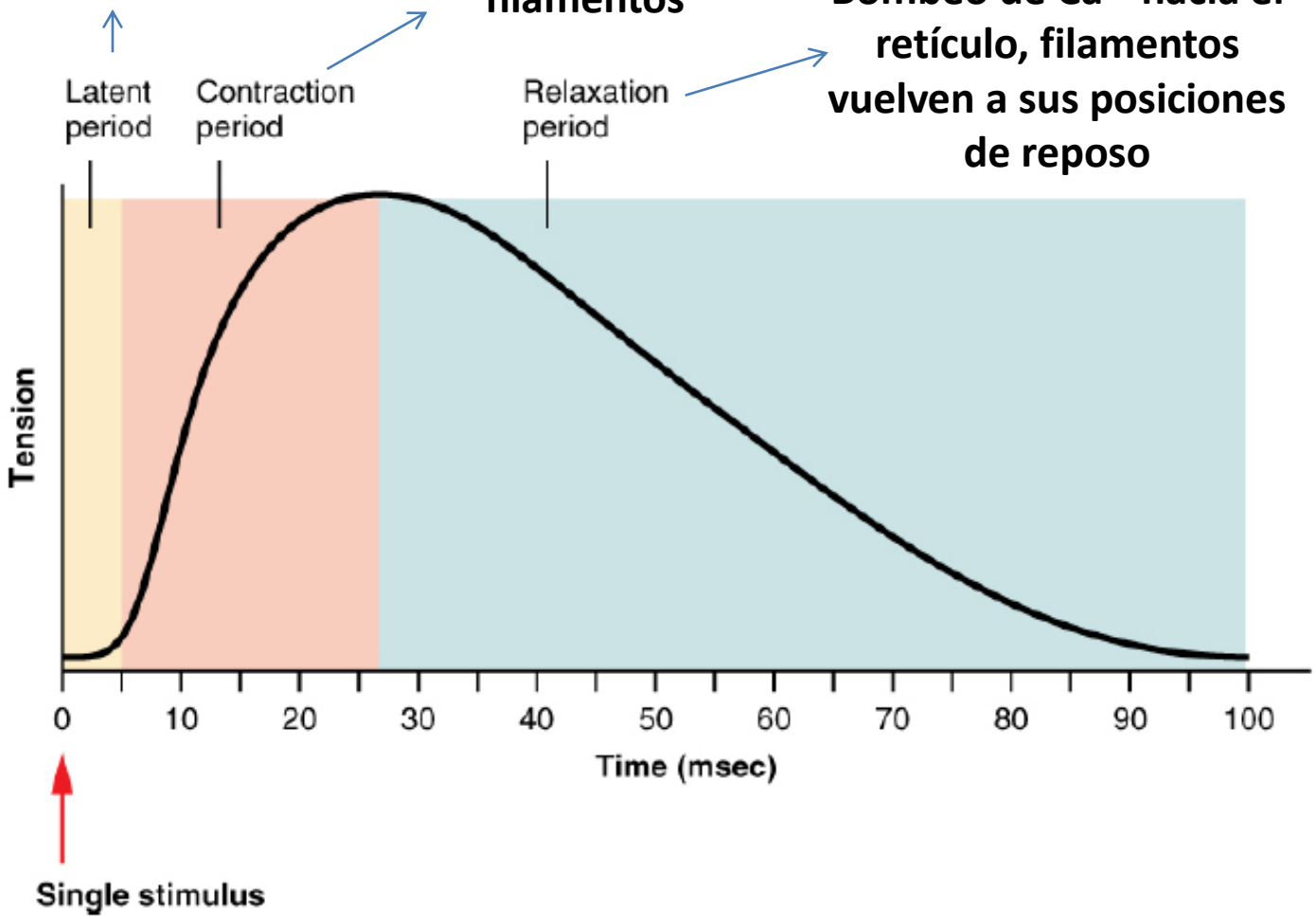
↓

Deslizamiento de filamentos gruesos y delgados (ATP)

Propagación del potencial de acción

Liberación de Ca^{++} deslizamiento de filamentos

Bombeo de Ca^{++} hacia el retículo, filamentos vuelven a sus posiciones de reposo



En práctico verán:

- Músculo esquelético (visto hoy)
- Músculo cardíaco
- Músculo liso

Pregunta de examen

Describa el rol de las fibras de tensión de actina y miosina en la migración celular.