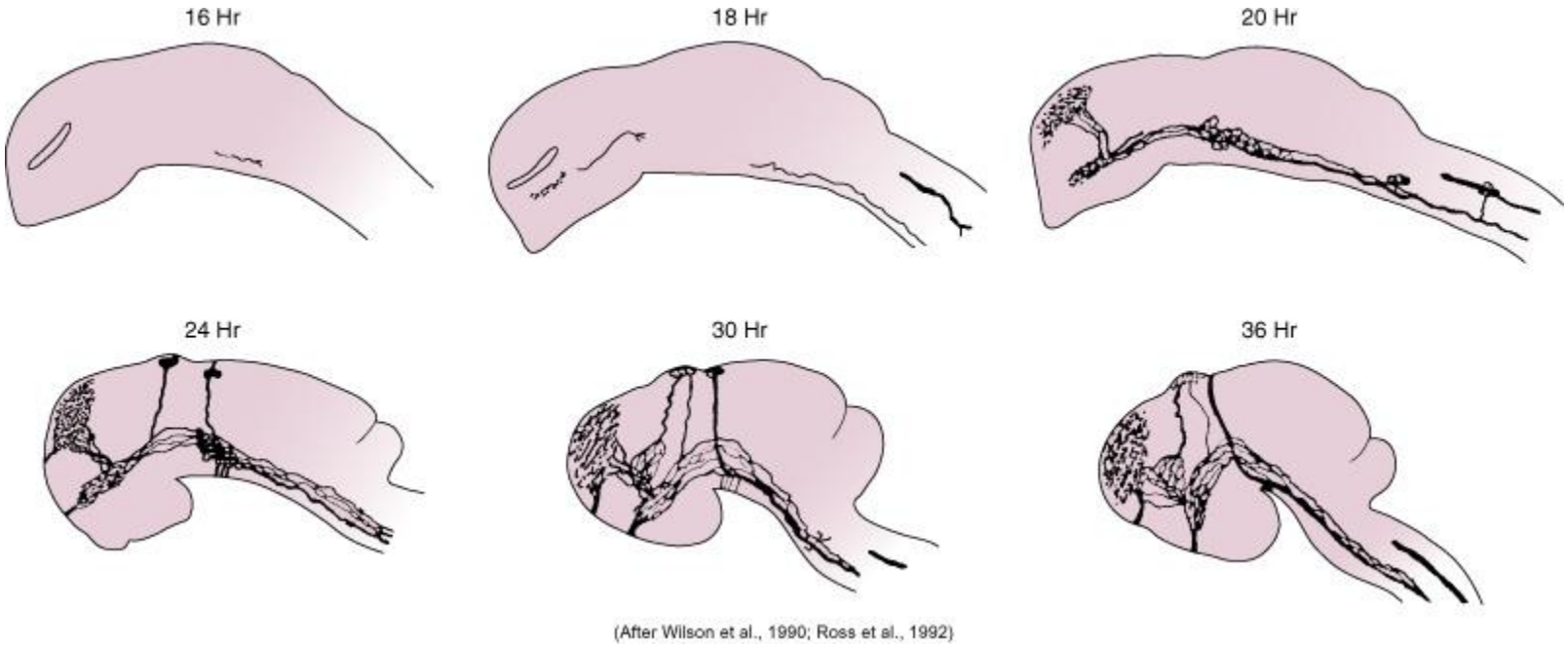
Development of the Nervous System HS2017

BIO 344/376-1305

Axon Growth and Survival

Esther Stoeckli
Institute of Molecular Life Sciences
University of Zurich

The complexity of axon tracts increases rapidly during early stages of development



(After Wilson et al., 1990; Ross et al., 1992)

Sanes, Reh, Harris, 2006

Axons have to:

survive

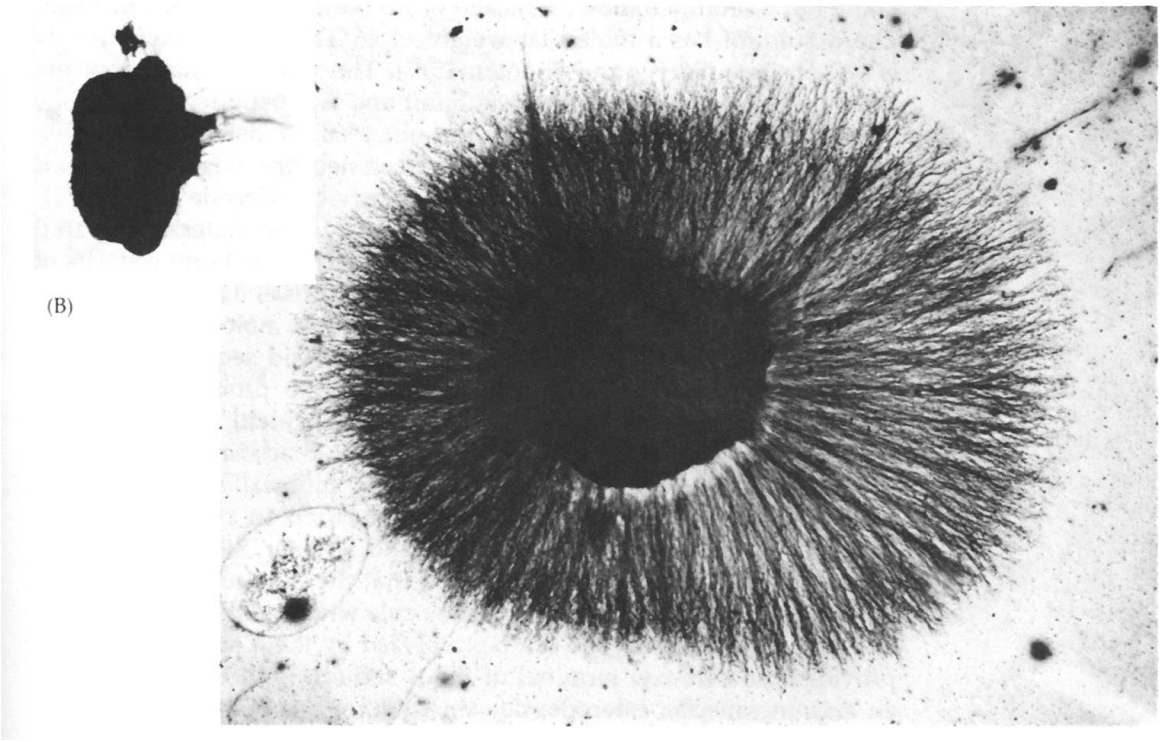
grow

find and get to target

recognize target

connect to target

Neurons need trophic factors for survival



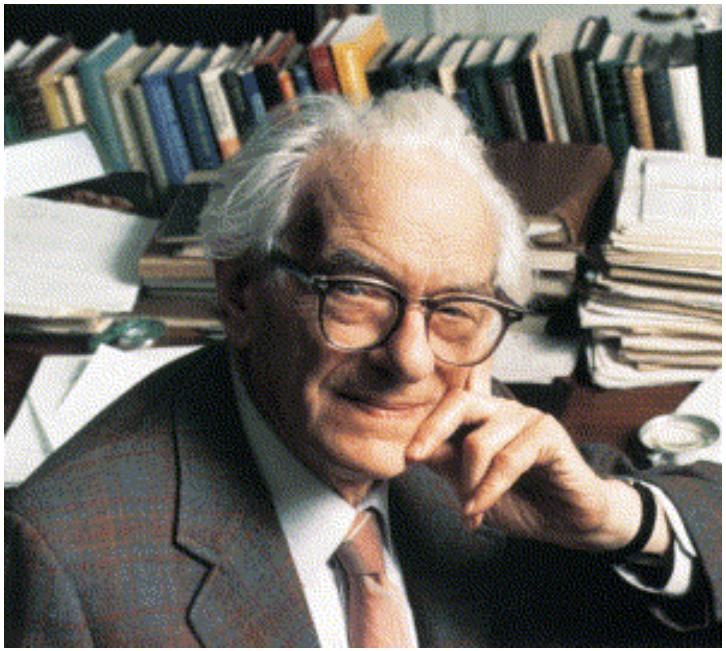
The discovery of Nerve Growth Factor (NGF)

Rita Levi-Montalcini
1909-2012

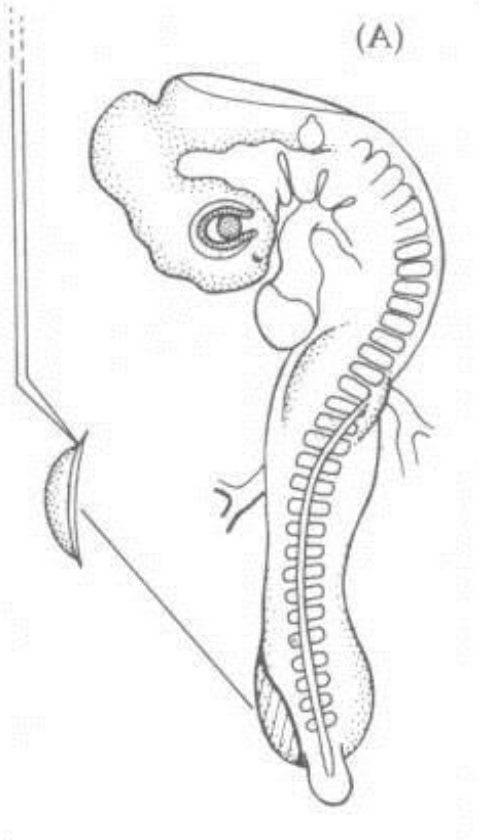
Nobel Prize 1986

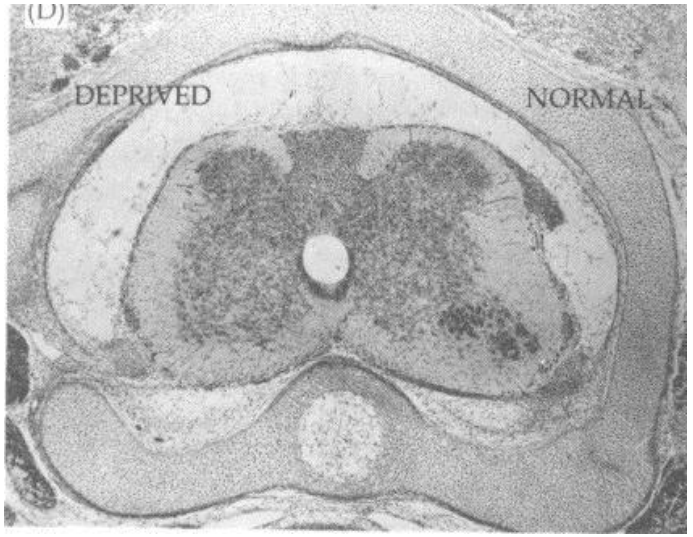
in 2009

Viktor Hamburger (1900 - 2001)

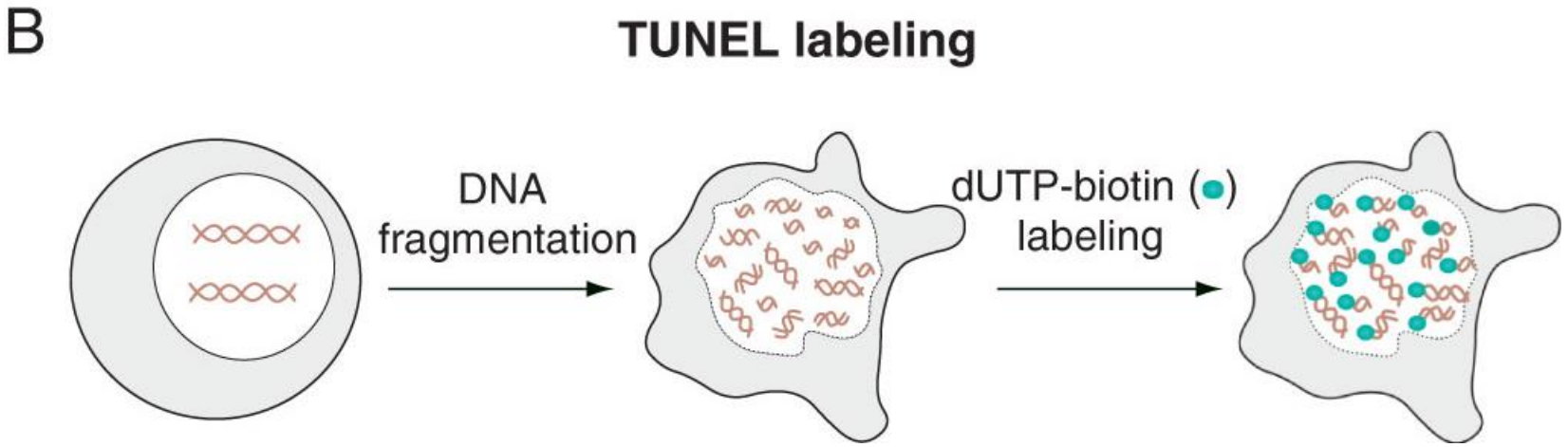


Reducing the target area enhances cell death





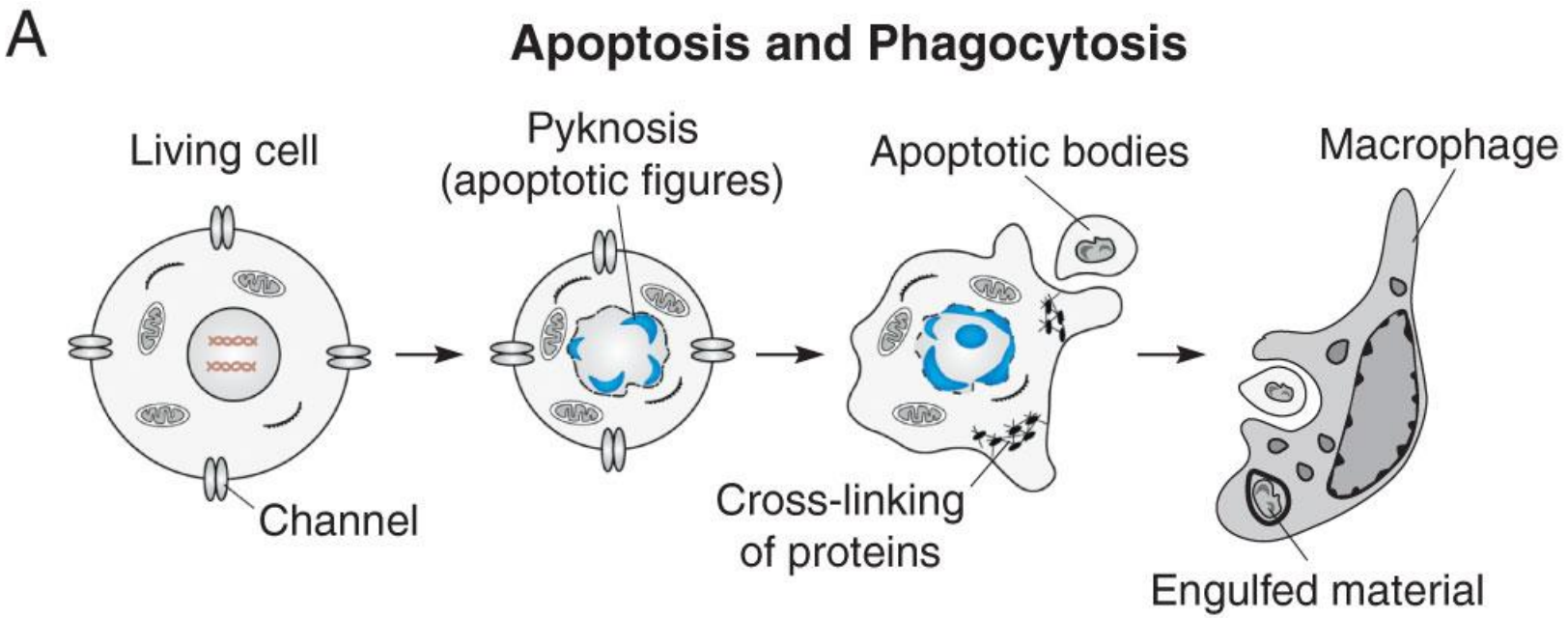
Cells that die by apoptosis can be recognized by specific features



pathway called apoptosis - you know it just too well (see cell biology summary)

Sanes, Harris, Reh 2012

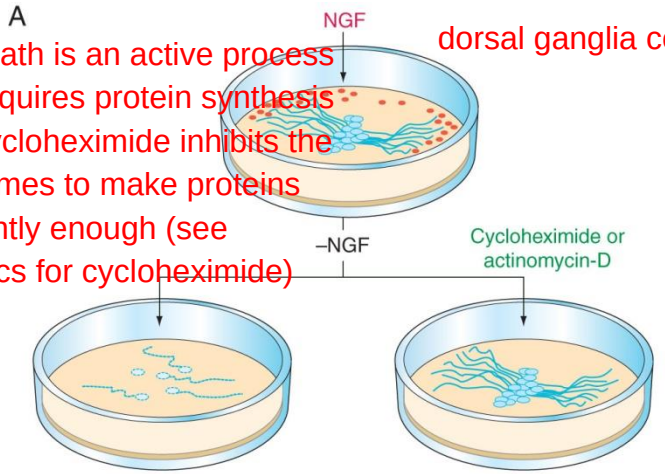
Cells that die by apoptosis can be recognized by specific features



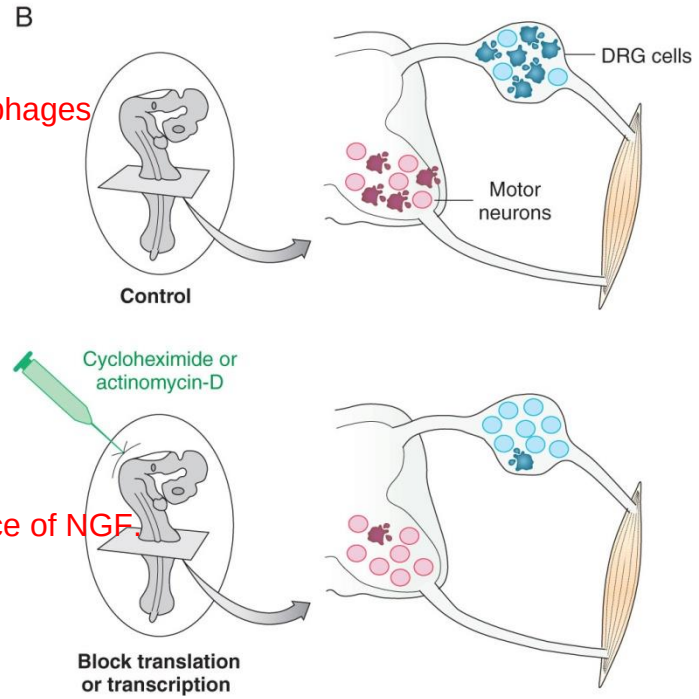
Sanes, Harris, Reh 2012

Cell death by apoptosis requires protein synthesis

cell death is an active process that requires protein synthesis and cycloheximide inhibits the ribosomes to make proteins efficiently enough (see genetics for cycloheximide)



protein synthesis can be blocked by cycloheximide.
it works both in vitro and in vivo, so less cells die actually, because they lack the signal on their surface for the macrophages



snake venom has proteases to cleave proteins

they used the venom to get at the cleavage site to make the enzyme site active. snake venom is a very potent source of NGF.

it was clear that NGF was a protein.

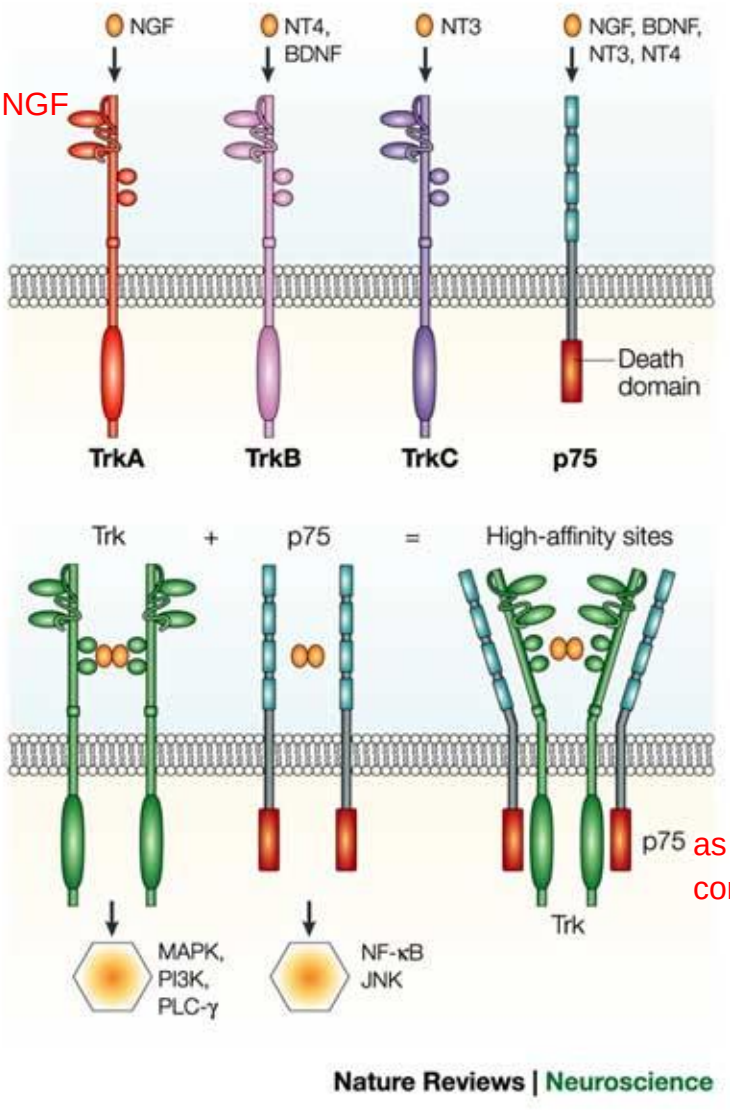
Sanes, Harris, Reh 2012

Neurotrophins support survival of sensory neuron subtypes

brain dervied growth factor

trkA: high affinity binding site for NGF
analogous for the other trks

(Trk:=track)

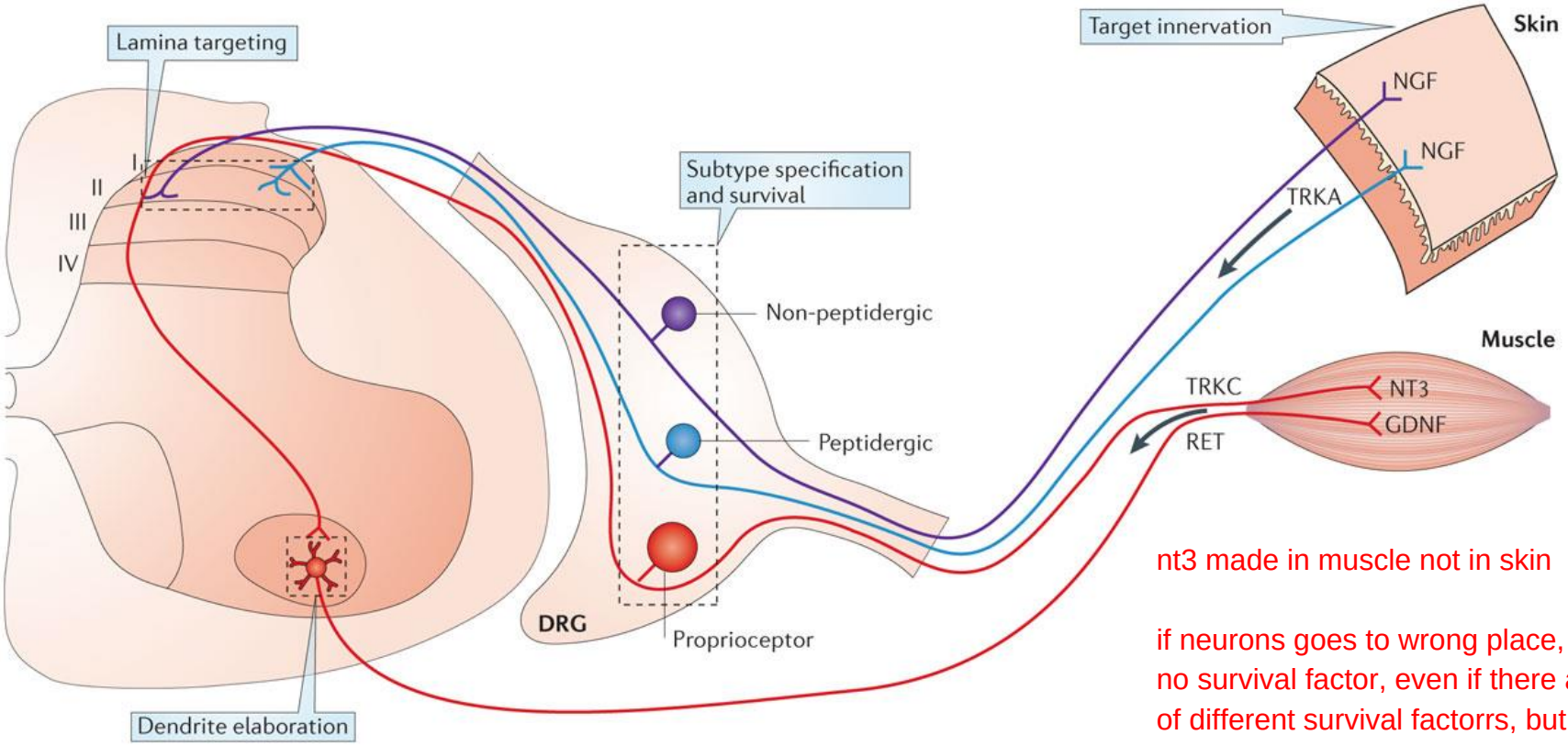


p75 alone triggers the cell deaths program actuaally

as a complex, it is a highly effective survival complex

Chao, 2003

Neurotrophins are released in a target-specific manner

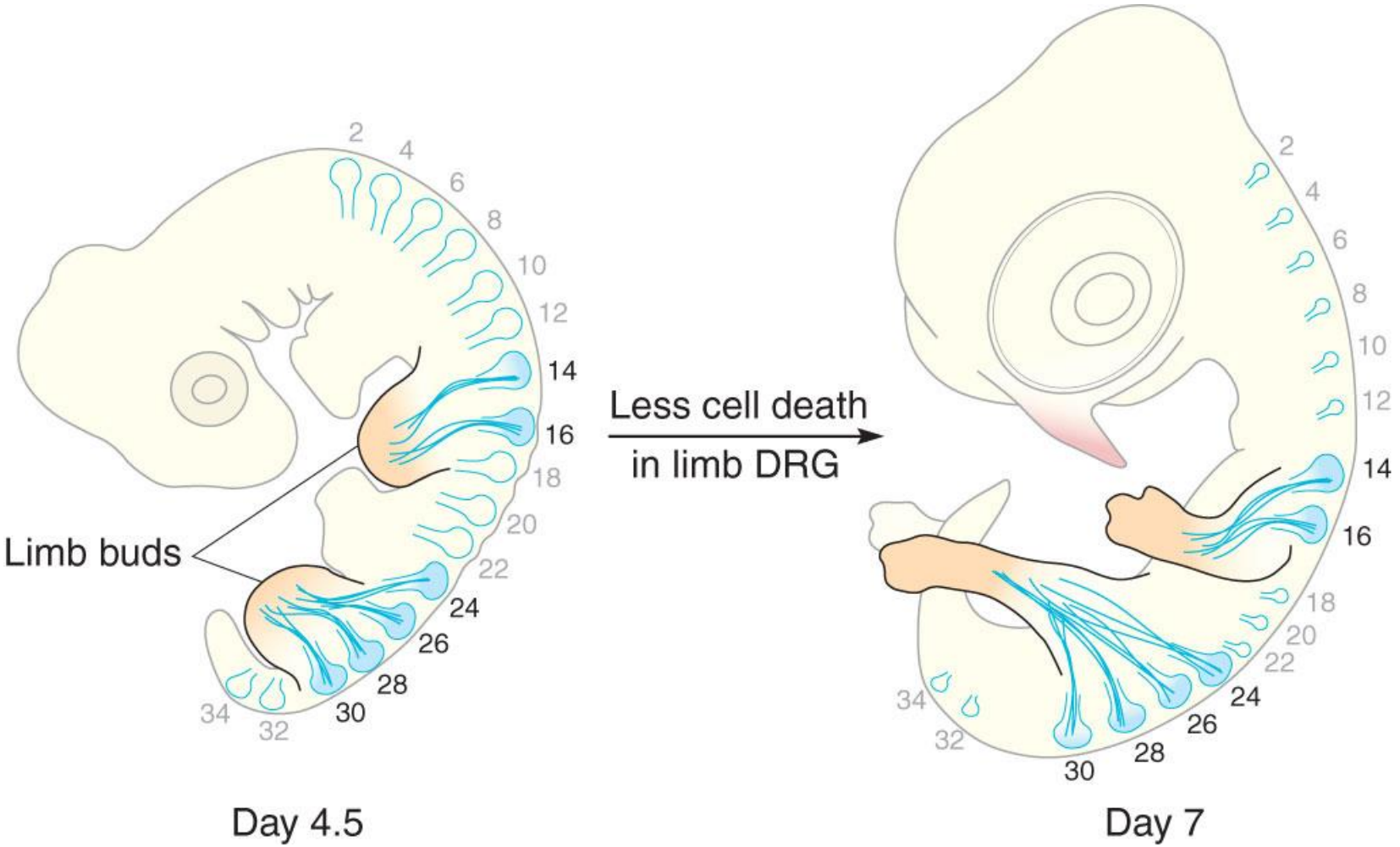


nt3 made in muscle not in skin

if neurons goes to wrong place, there is no survival factor, even if there are lots of different survival factorrs, but it will die because it lacks the right receptors

Harrington & Ginty, 2013

Neurotrophins provide a means to adapt innervation to tissue size



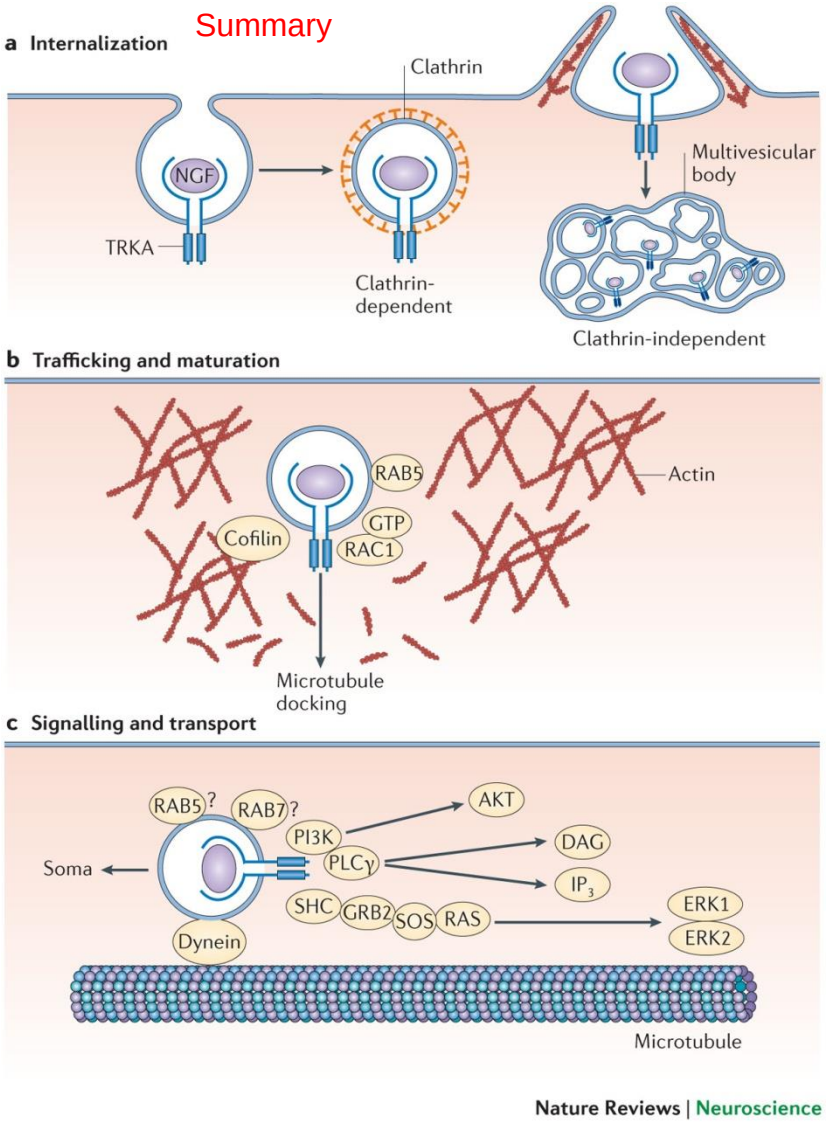
Sanes, Harris, Reh 2012

why are so many cells made if many die in the end?

Neurotrophin signaling requires endocytosis and retrograde transport

basically, a summary of what we learned about neurotrphic signaling

Summary



you need clathrin coat for endocytosis, but also bulk endocytosis can occur.

although, the vesicles that are formed have different properties and possibilities.

those endocytosed structures need to pass through the actin skeleton somehow

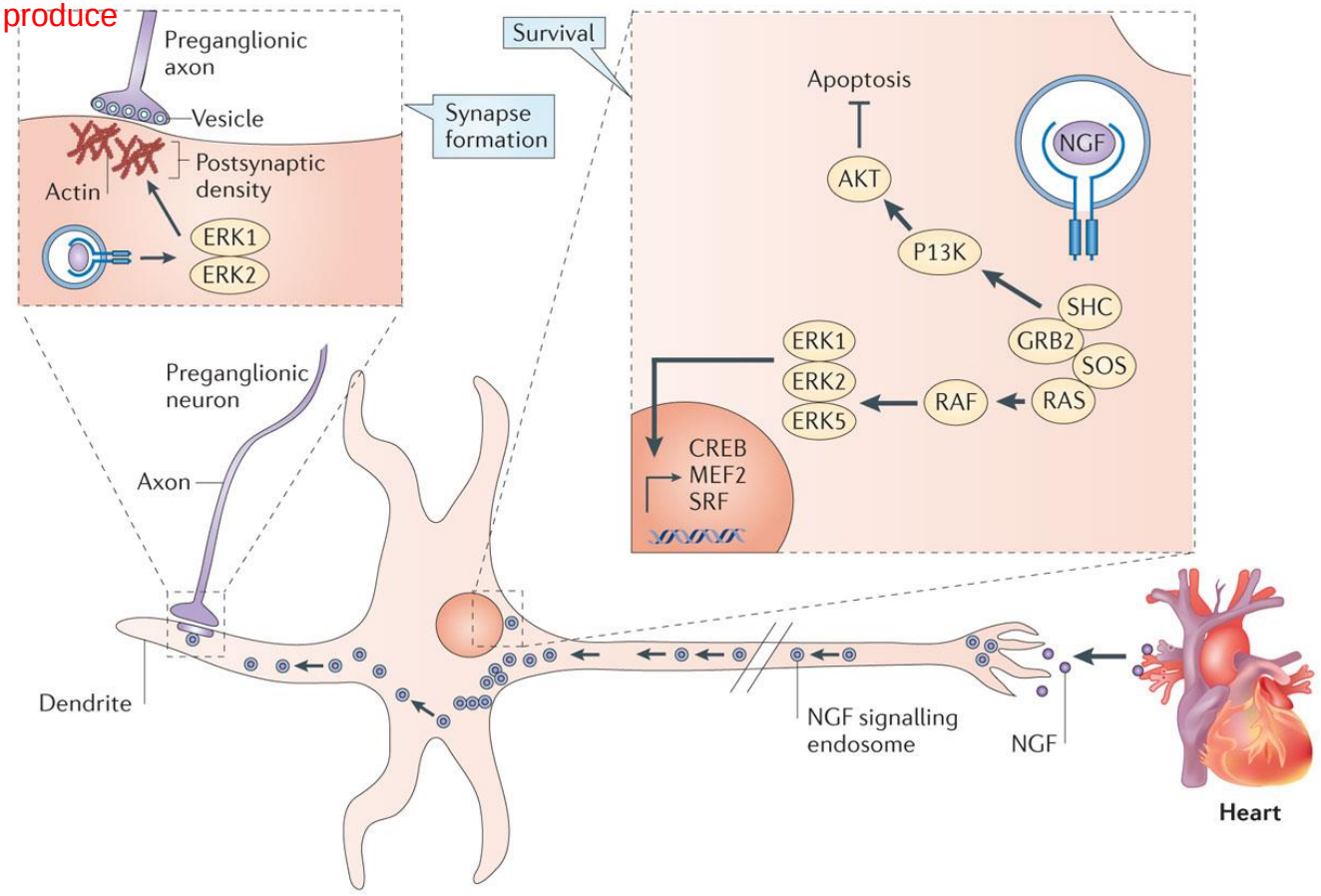
dynein is the "motor", a retrograde motor/transport, back to the cell body

(no need to learn all these factors, except for dynein) at the end, there is activation of ERK1/2 (kinase pathway)

Harrington & Ginty, 2013

Neurotrophins prevent apoptosis and support synaptogenesis

erk signaling helps to produce postsynaptic axons-



phosphorylation intitiates neuronal survival

Harrington & Ginty, 2013

Nature Reviews | Neuroscience

Axons have to:

survive

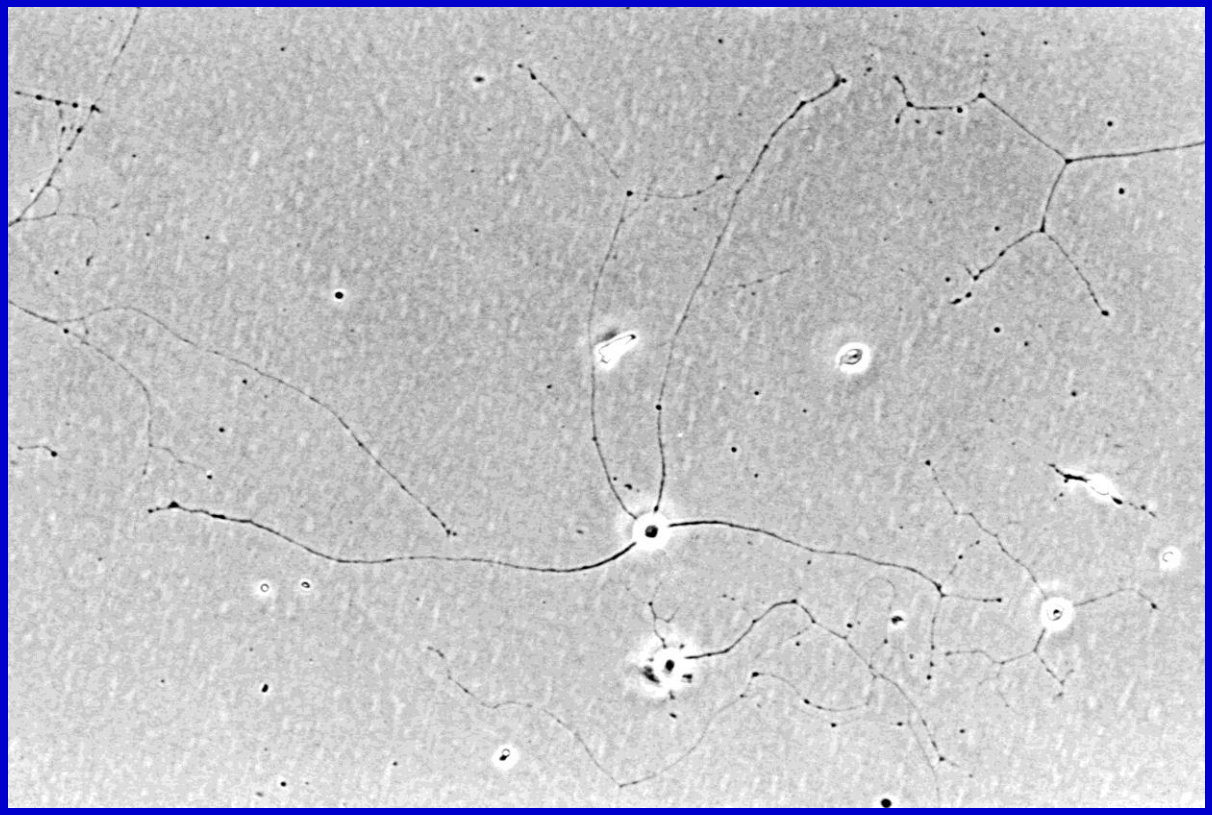
grow

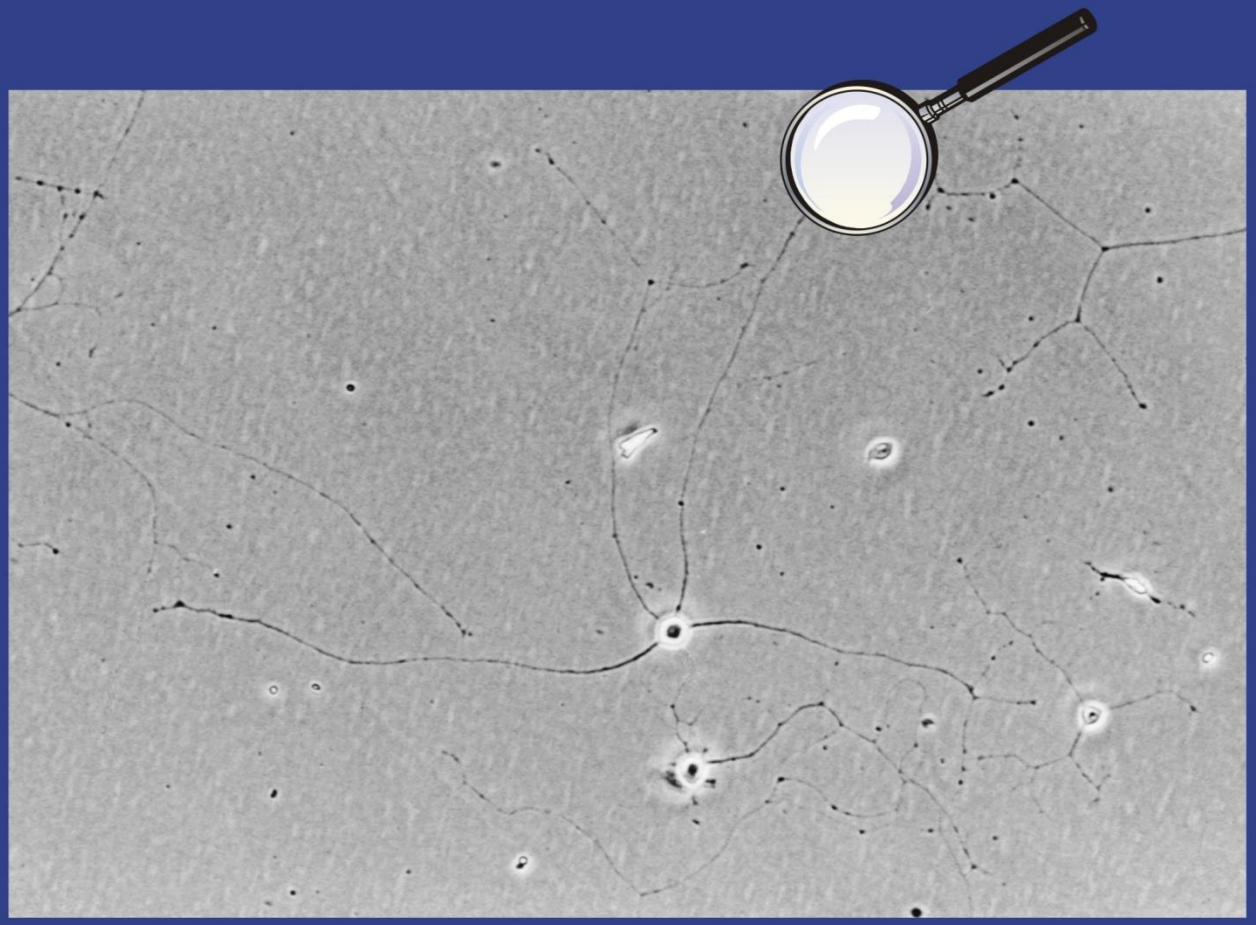
find and get to target

recognize target

connect to target

Axons extend long processes to connect to their targets

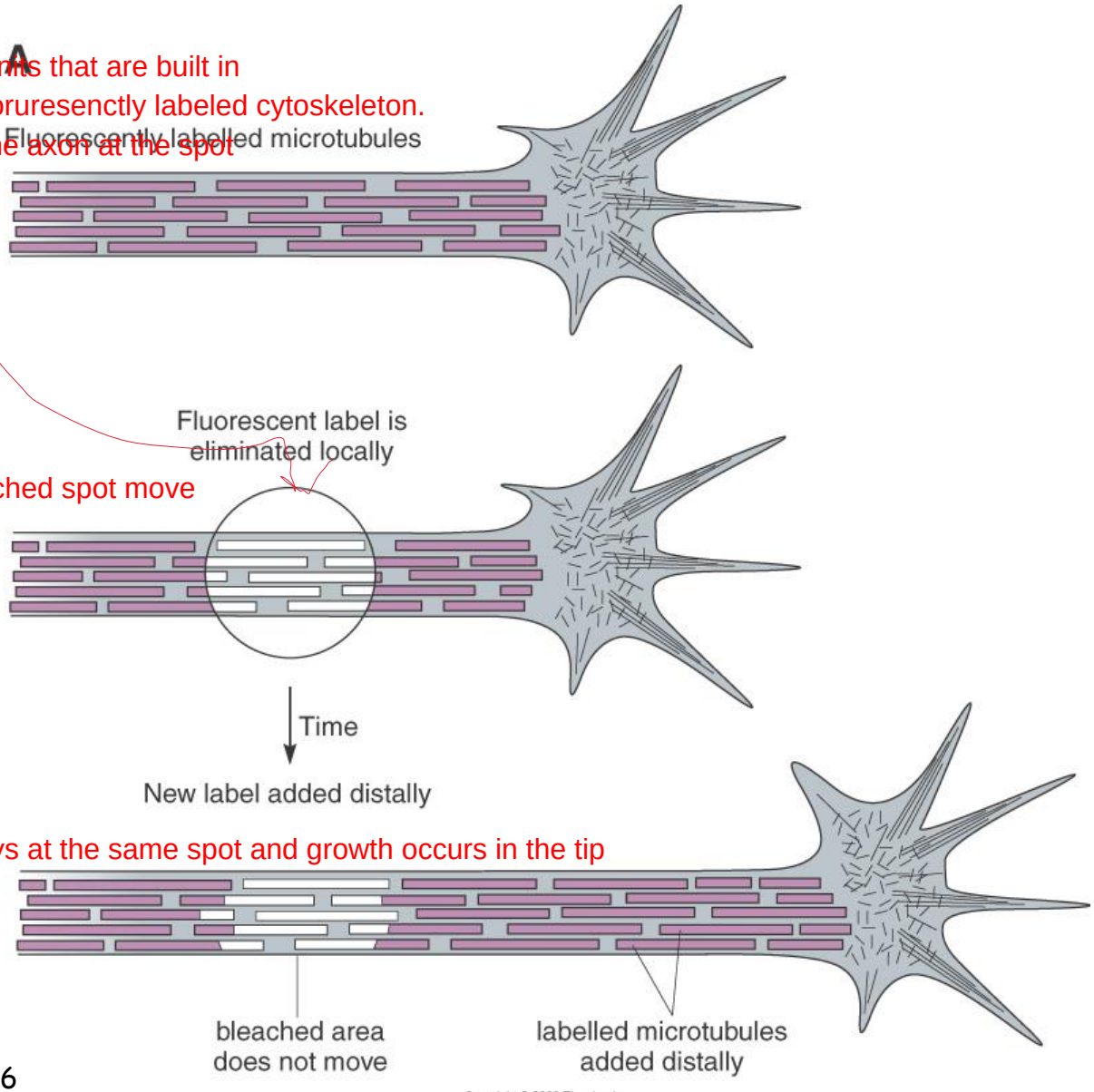




the tip of axon or dendrite has finger like structures, constantly growing with high motility, exploring their environment - called growth cone

Axons grow by adding new microtubules at the distal end

add flouresecIty labebeled units that are built in microtubules so you get a floruresenctly labeled cytoskeleton.
with laser, one can bleach the axon at the spot



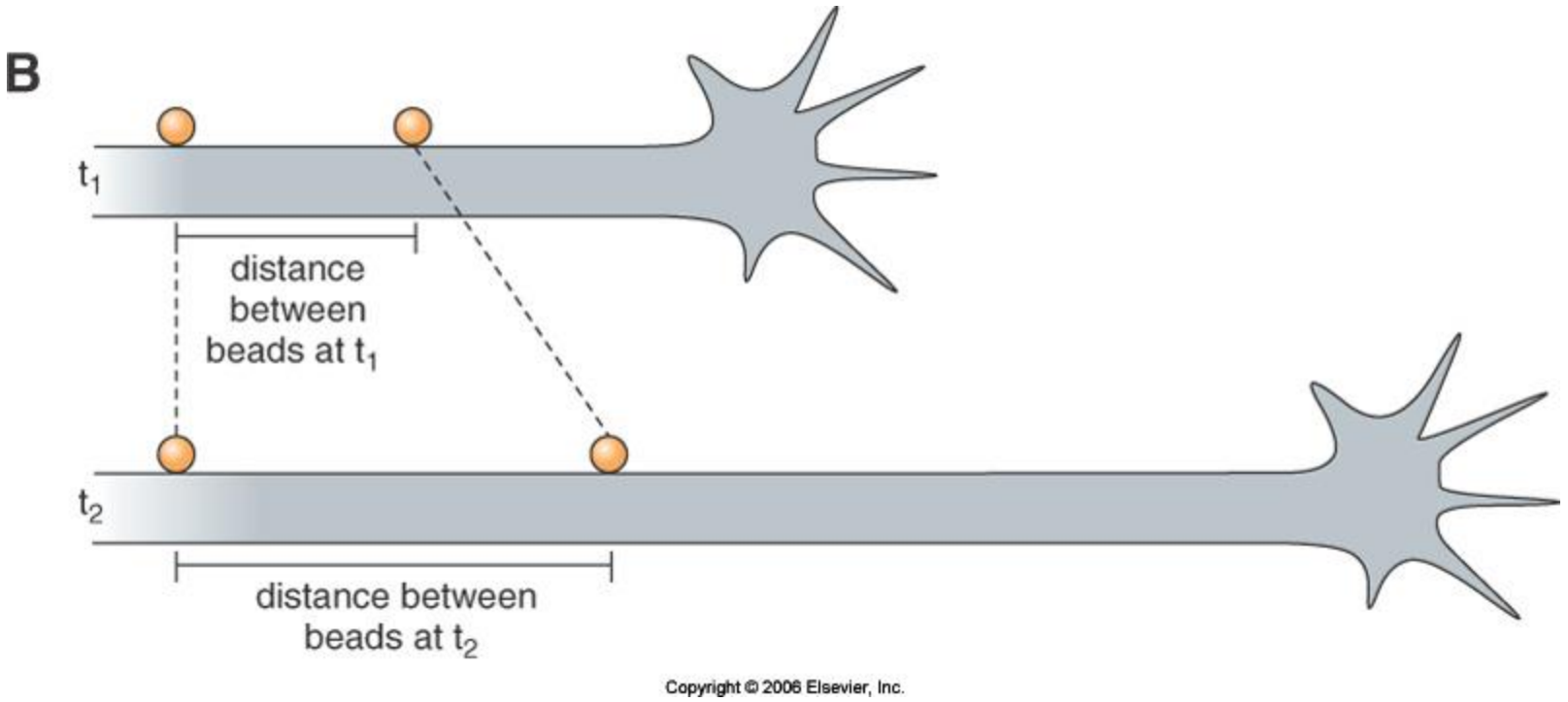
we can now watch the bleached spot move

what was shown: axon stays at the same spot and growth occurs in the tip

Sanes, Reh, Harris, 2006

axonal growth not only occurs on the tip, but also all along the axon

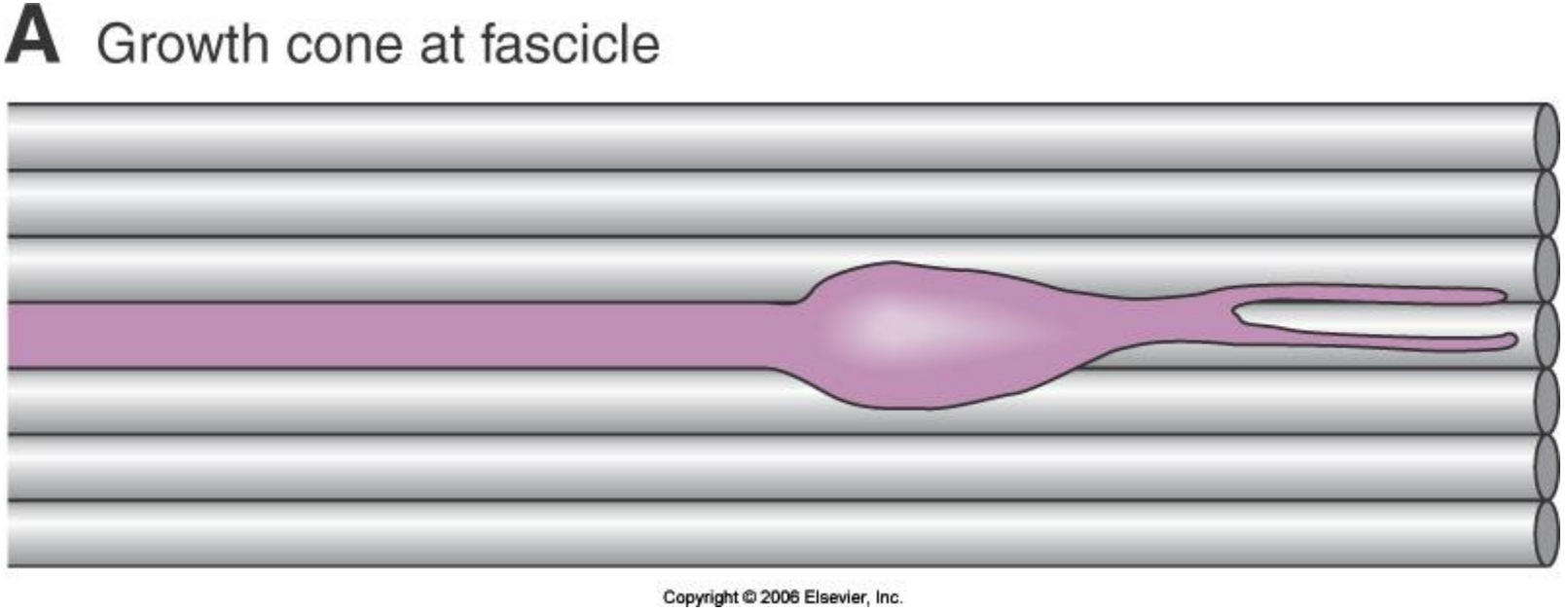
Axons grow by stretching



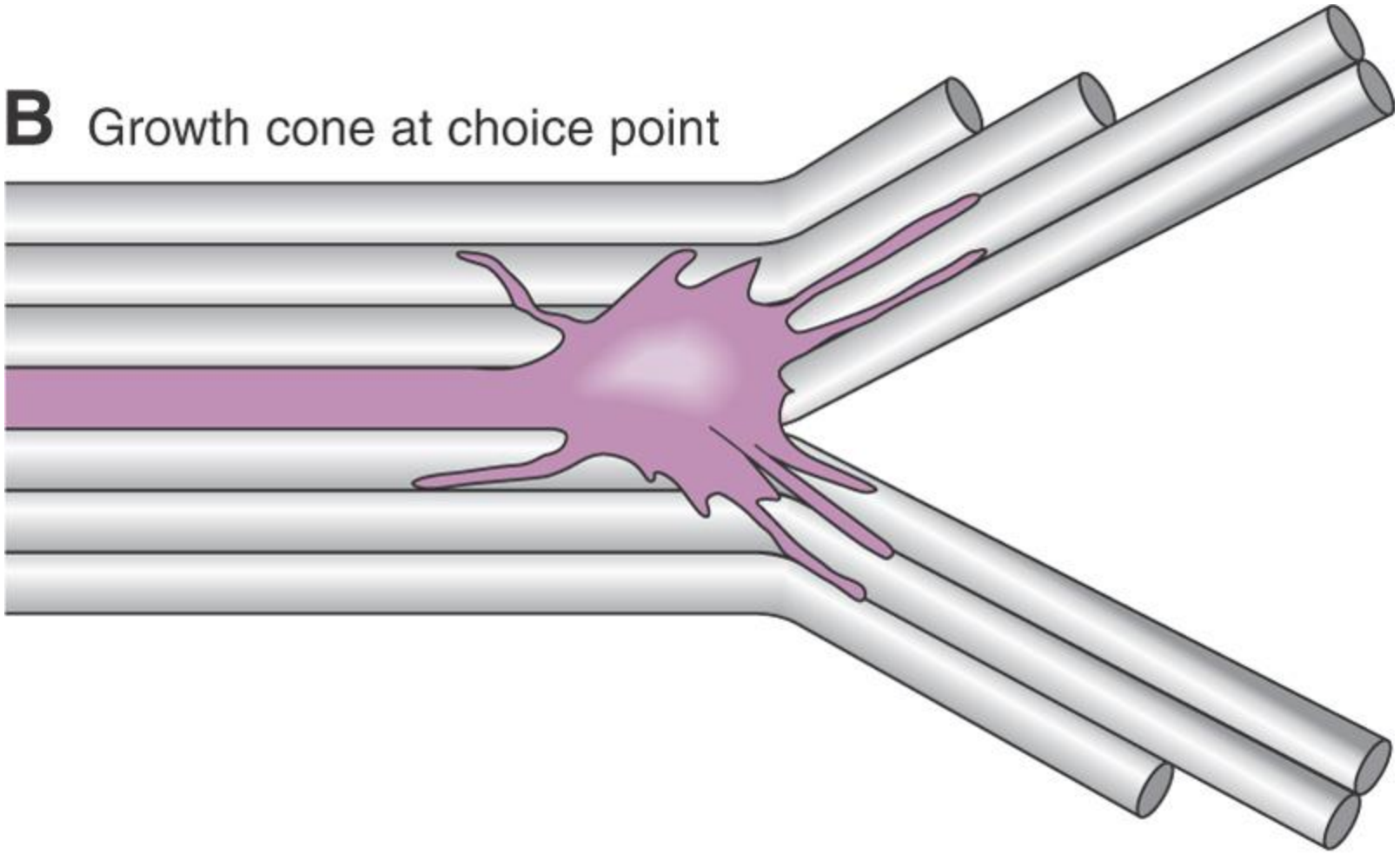
resolution of dilemma: depends on the stage of life: when it grows for the first time, it is the trip that grows.
after contact establishment, the tip no longer grows, because it is connected, but now stretching occurs.

Sanes, Reh, Harris, 2006

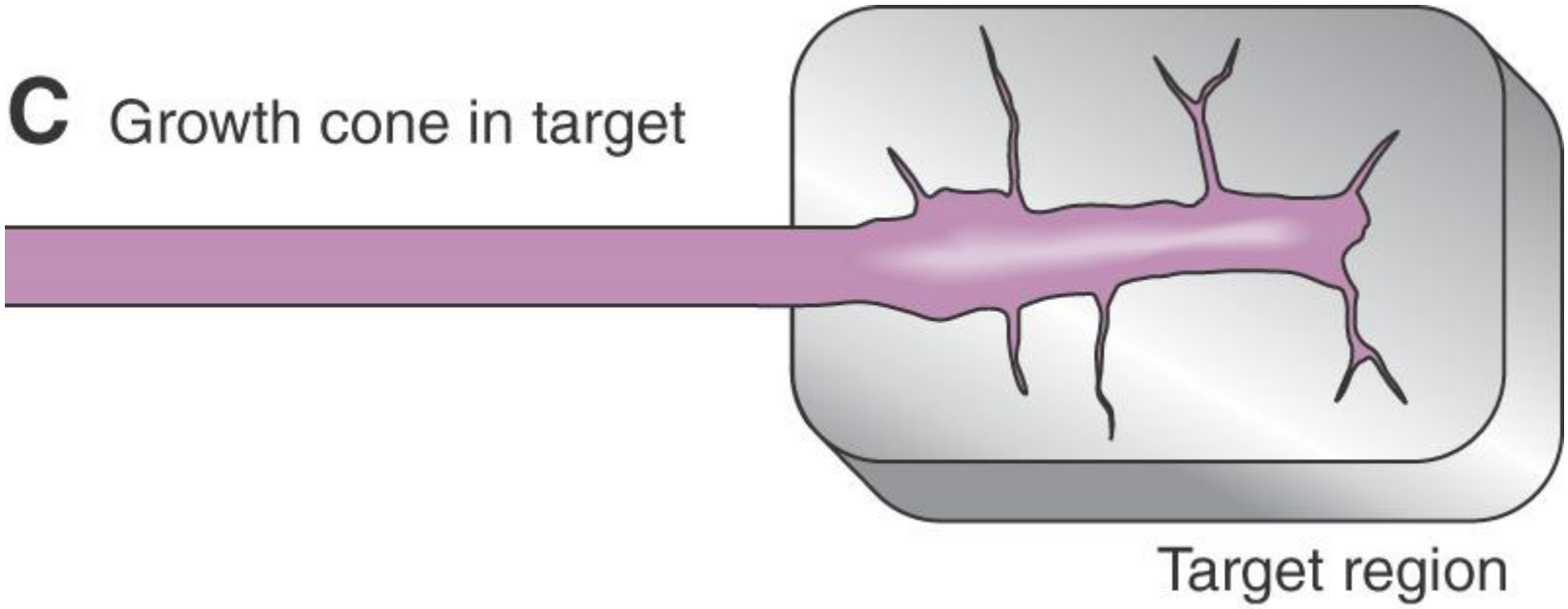
The shape of the growth cone differs depending on the environment or the „growth phase" of the axon



Sanes, Reh, Harris, 2006

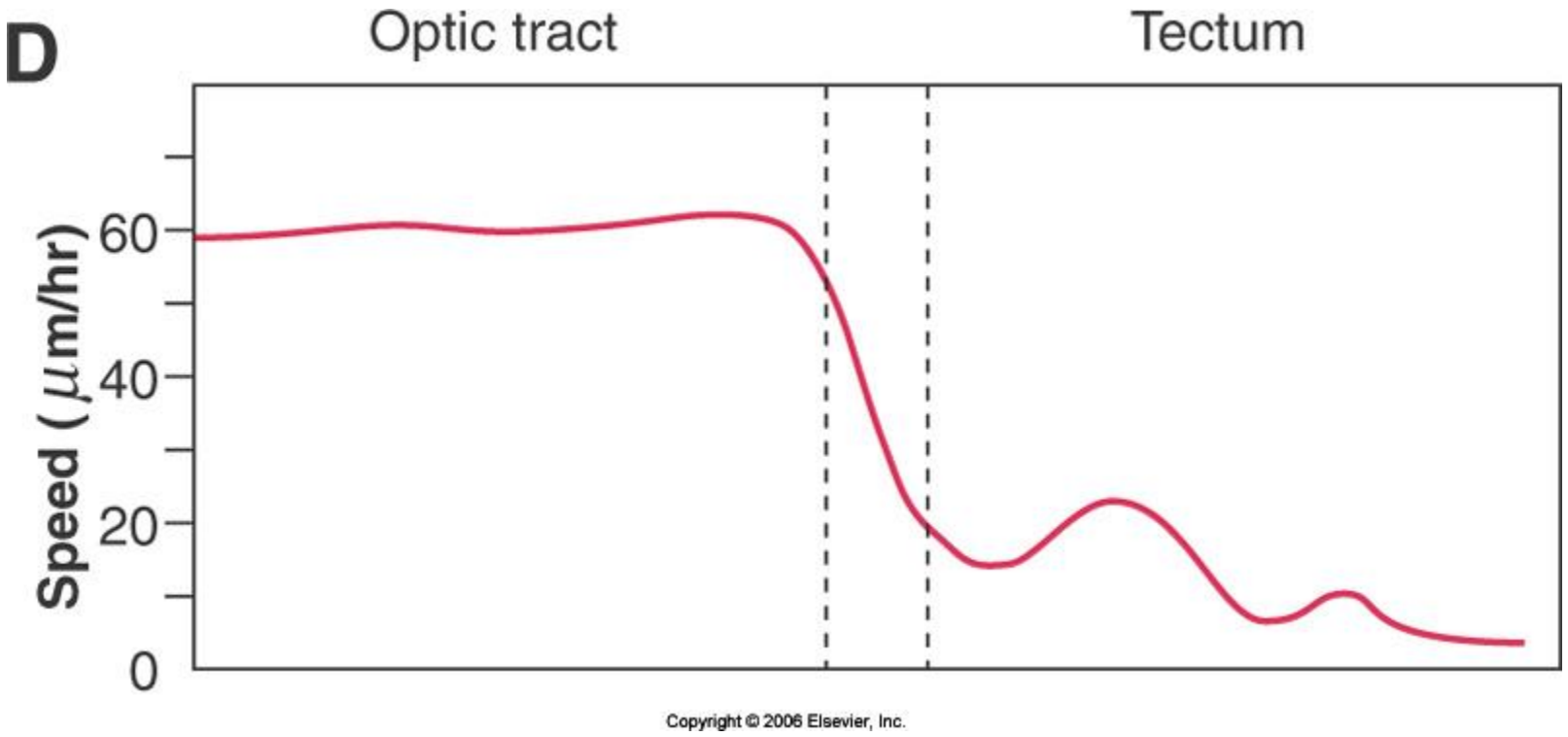


Sanes, Reh, Harris, 2006



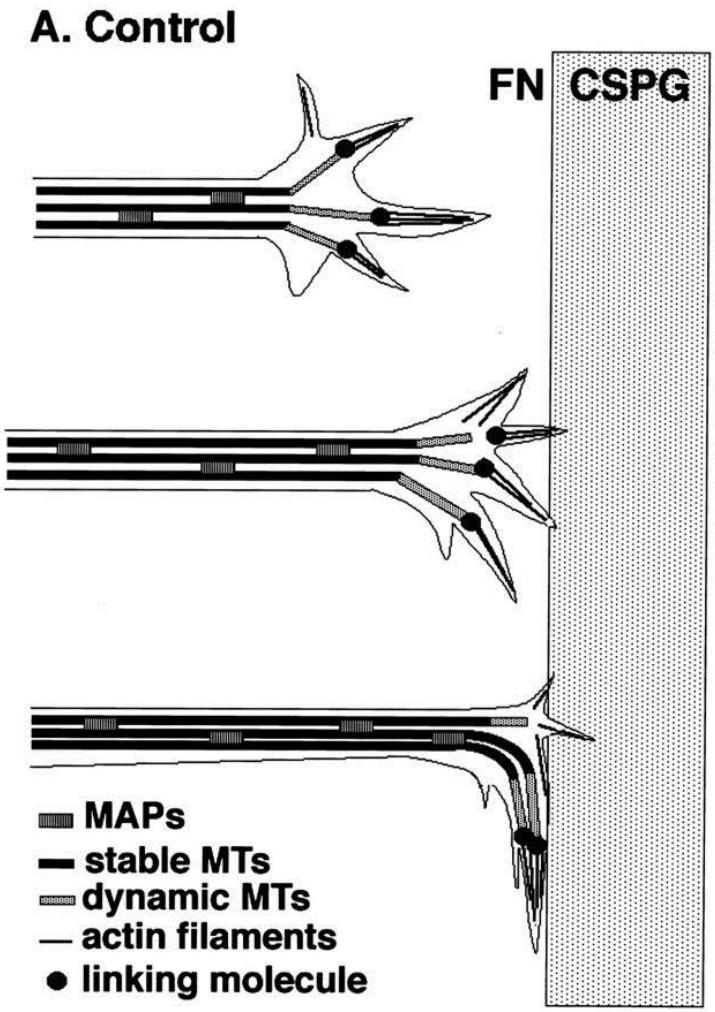
Sanes, Reh, Harris, 2006

The speed of the growth cone depends on the location

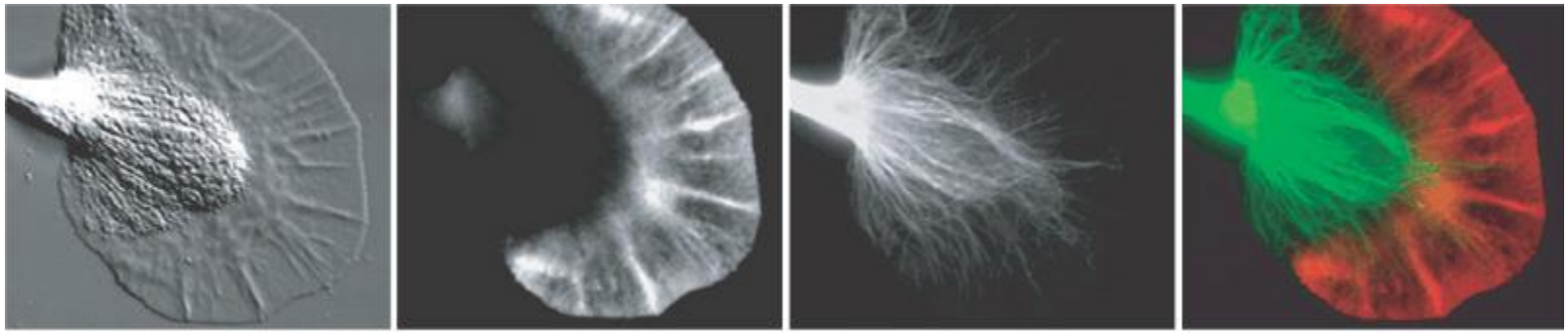


Sanes, Reh, Harris, 2006

Non-permissive substrates induce turns of growth cones at the substratum boundary



Growth cones depend on a dynamic cytoskeleton



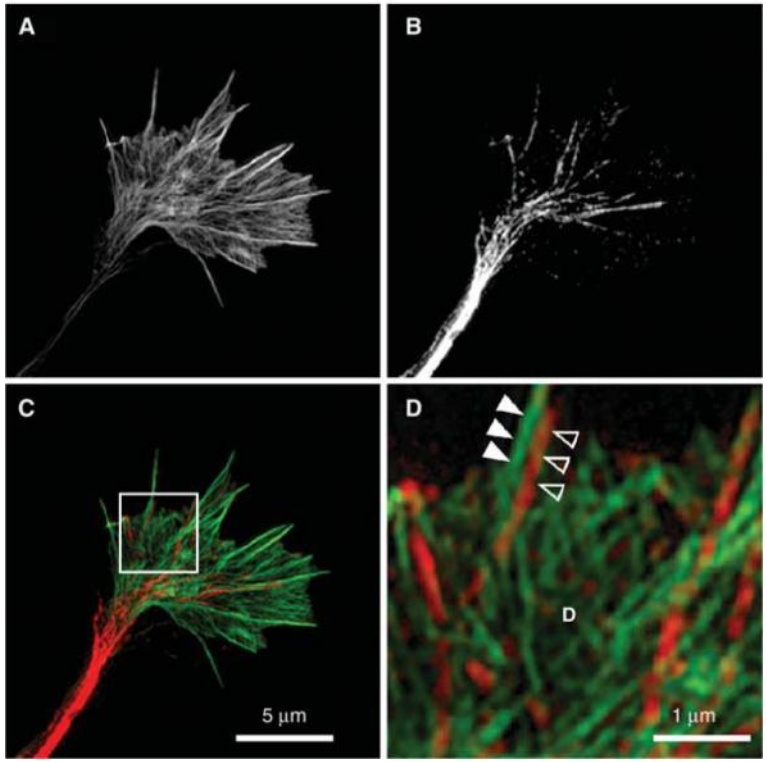
(From Paul Forscher)

actin filaments microtubules

Sanes, Reh, Harris, 2006

In vertebrate growth cones actin filaments and microtubules meet in the peripheral zone

actin filaments



microtubules

Dent et al., Cold Spring Harb Perspect Biol 2011

Actin and microtubulin form elements of the growth cone's cytoskeleton

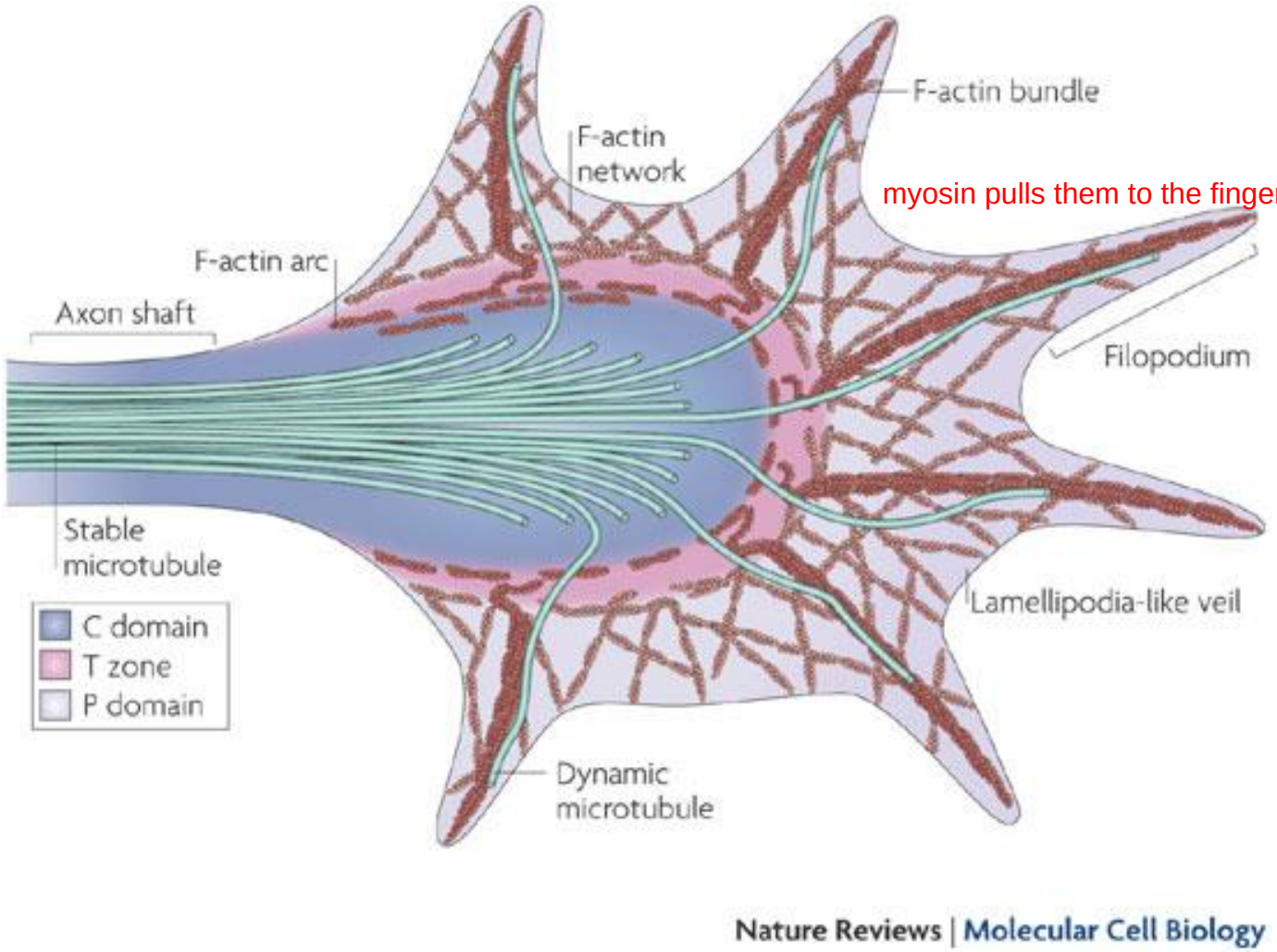
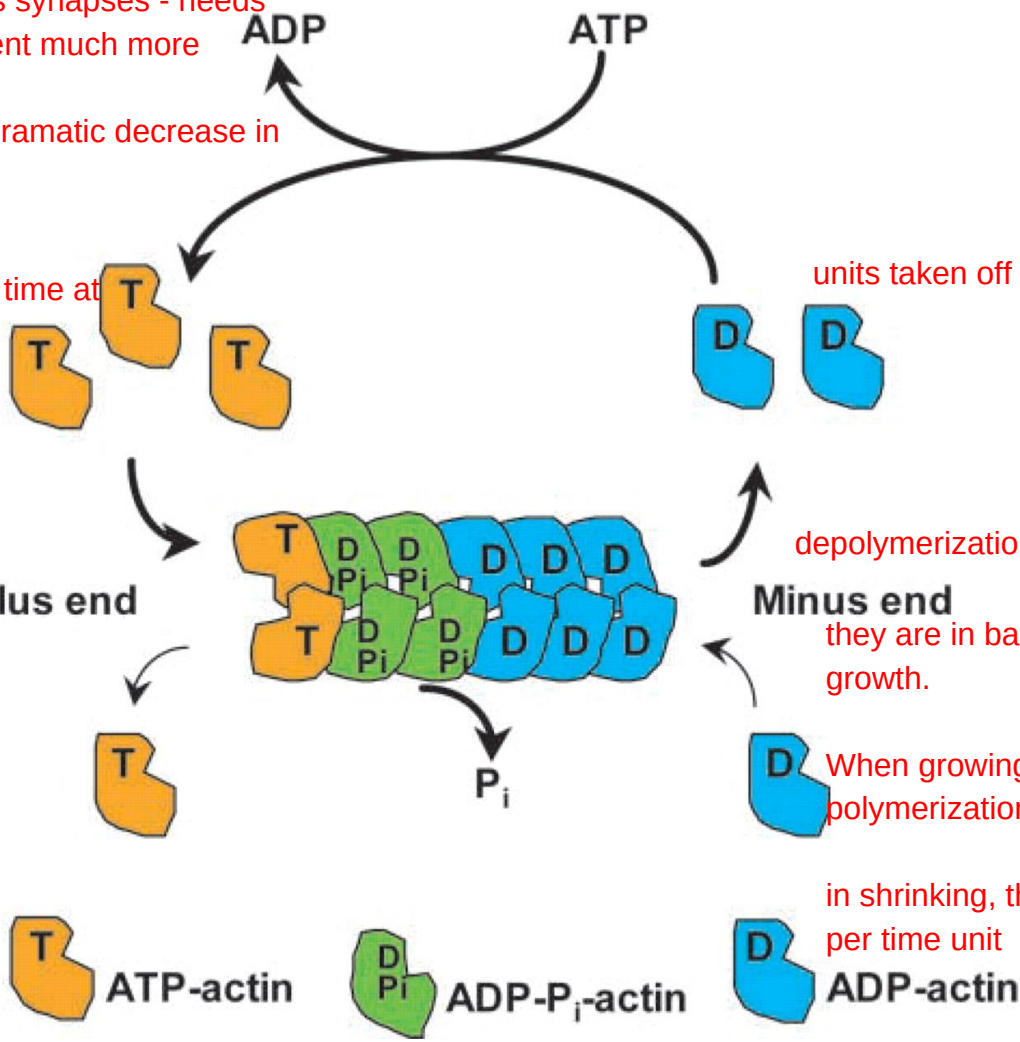


Figure 1 Treadmilling cycle of actin filaments at steady state.

and also for microtubules

place where it makes synapses - needs to explore environment much more

therefore there is a dramatic decrease in growth speed



actin units added all the time at plus end

units taken off

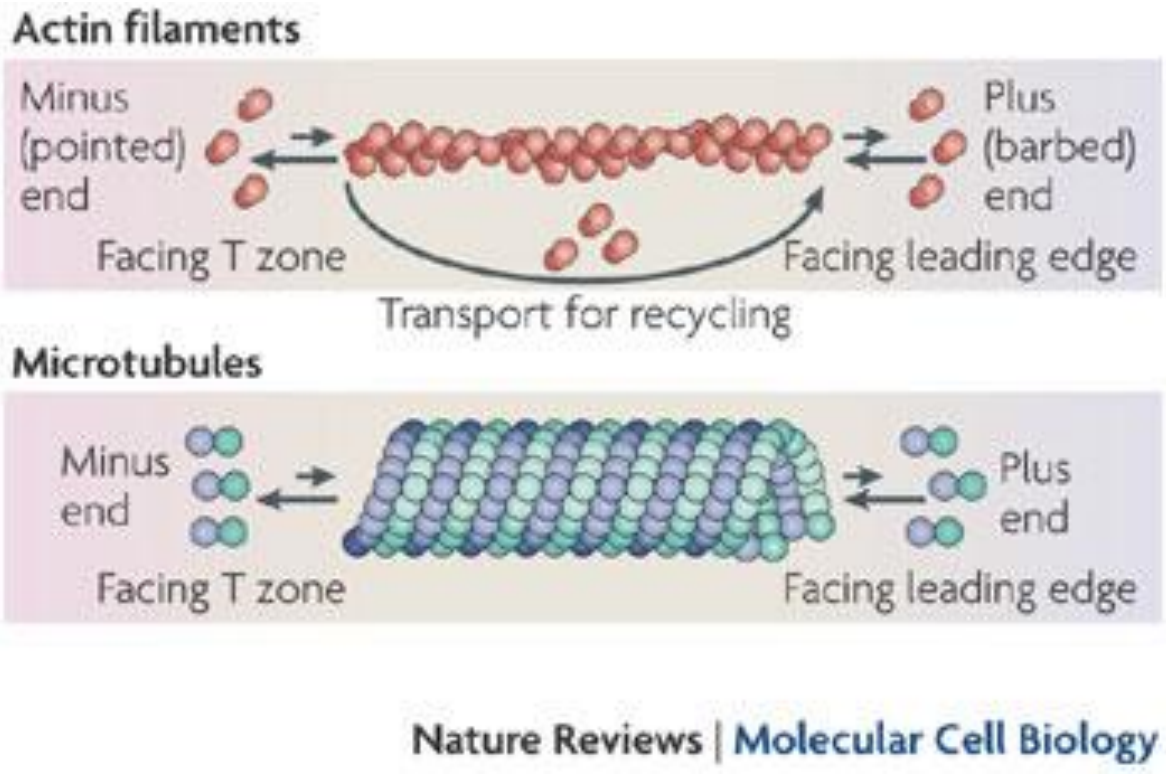
they are in balance, so no net growth.

When growing: then more polymerization

in shrinking, there is less adding per time unit

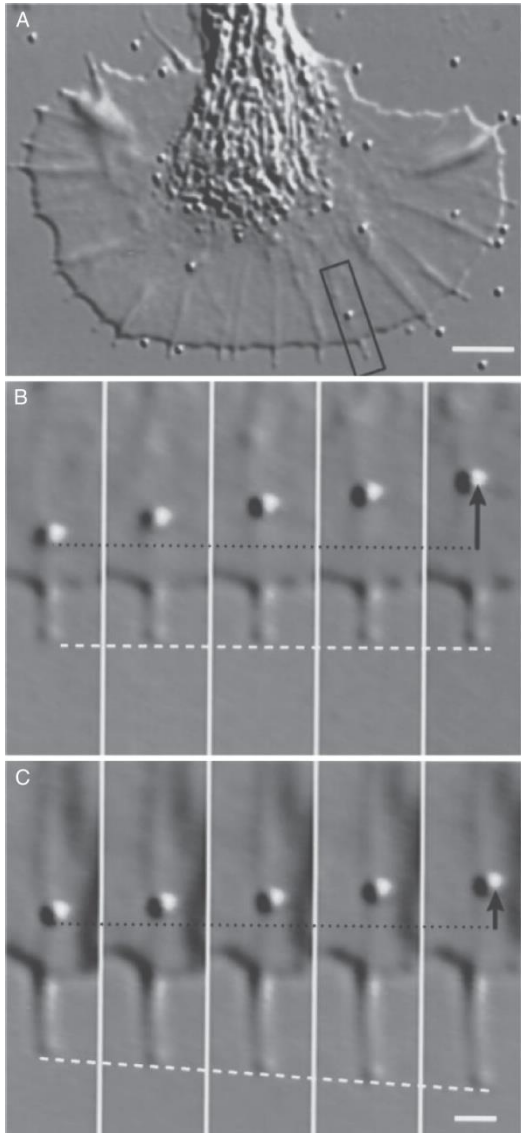
Gungabissoon R A , Bamburg J R J Histochem Cytochem 2003;51:411-420

Actin filaments and microtubules are in a ‚dymamic steady-state'

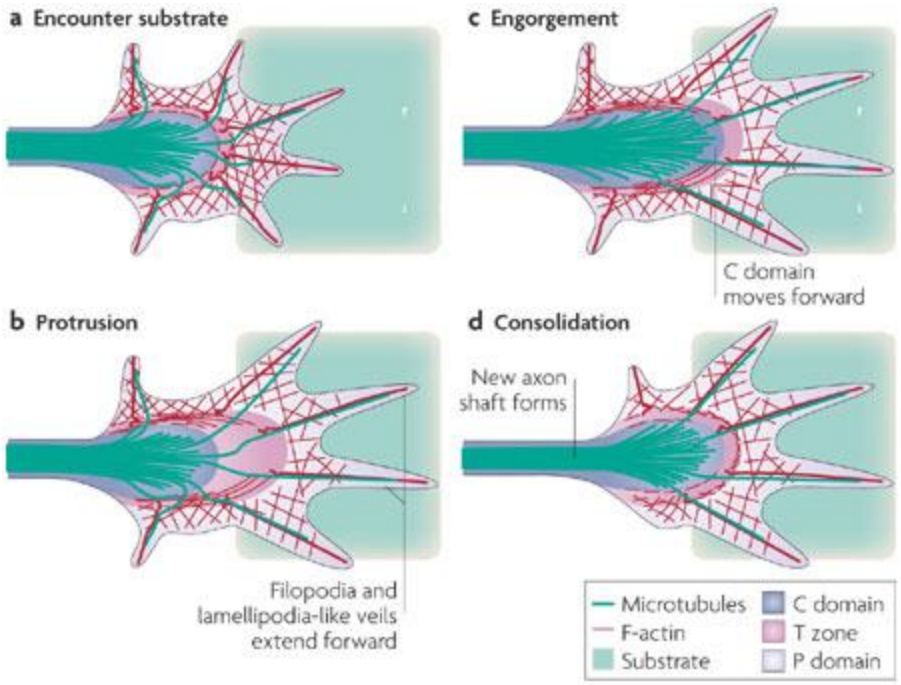


so basically they are rebuilt all the time.

Filopodia growth and retrograde actin flow are inversely correlated

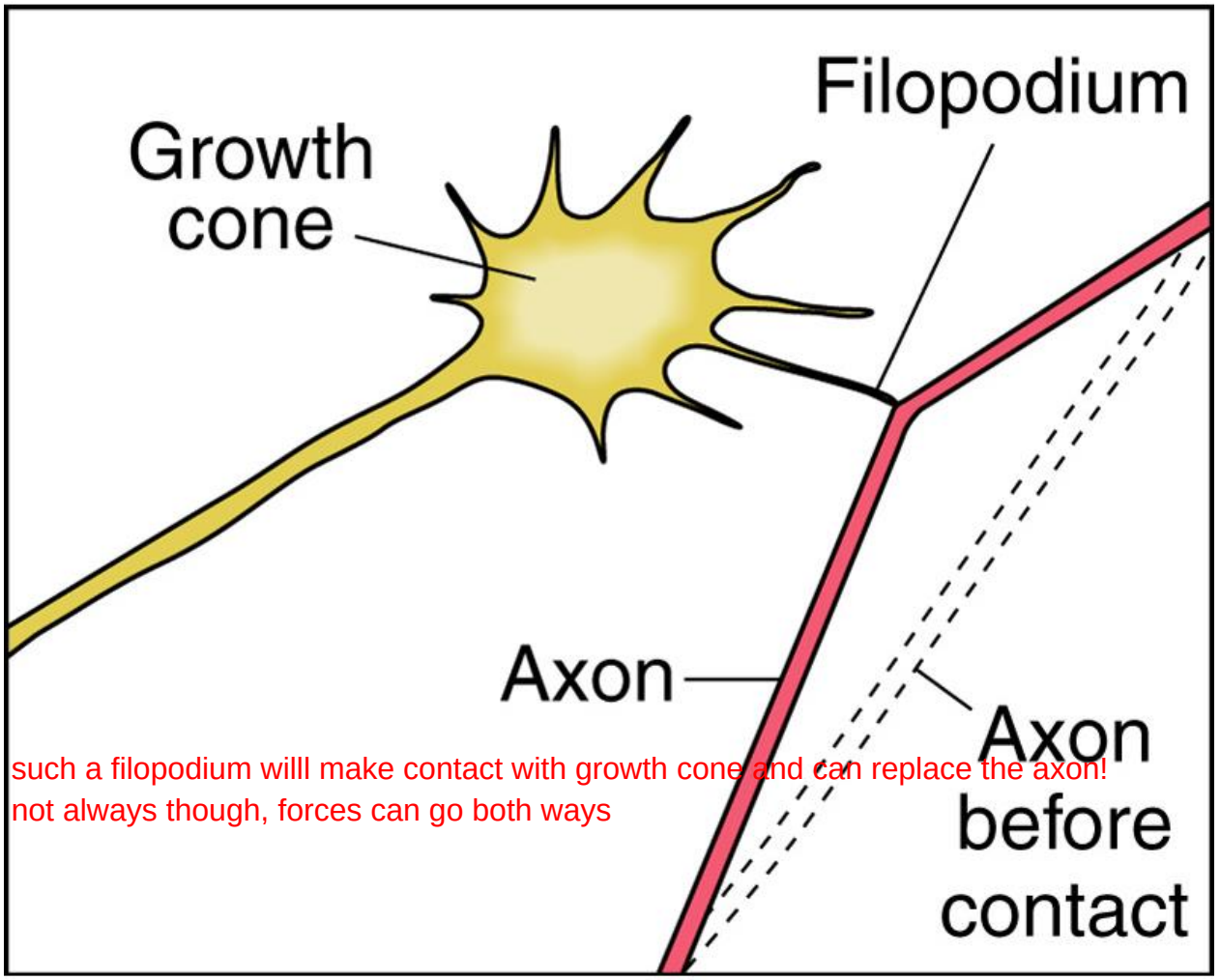


Sanes, Harris, Reh 2012



Nature Reviews | Molecular Cell Biology

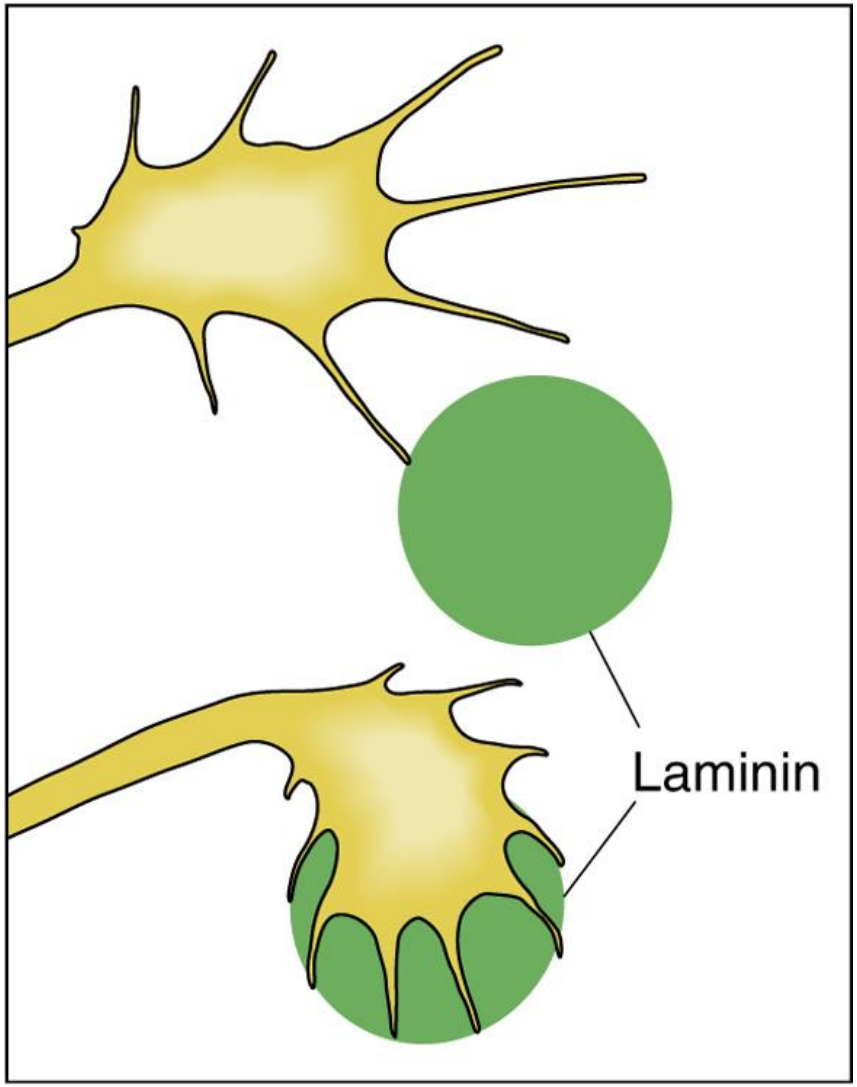
Filopodia can exert force



such a filopodium willl make contact with growth cone and can replace the axon!
not always though, forces can go both ways

Sanes, Reh, Harris, 2006

Filopodia induce growth cone turning



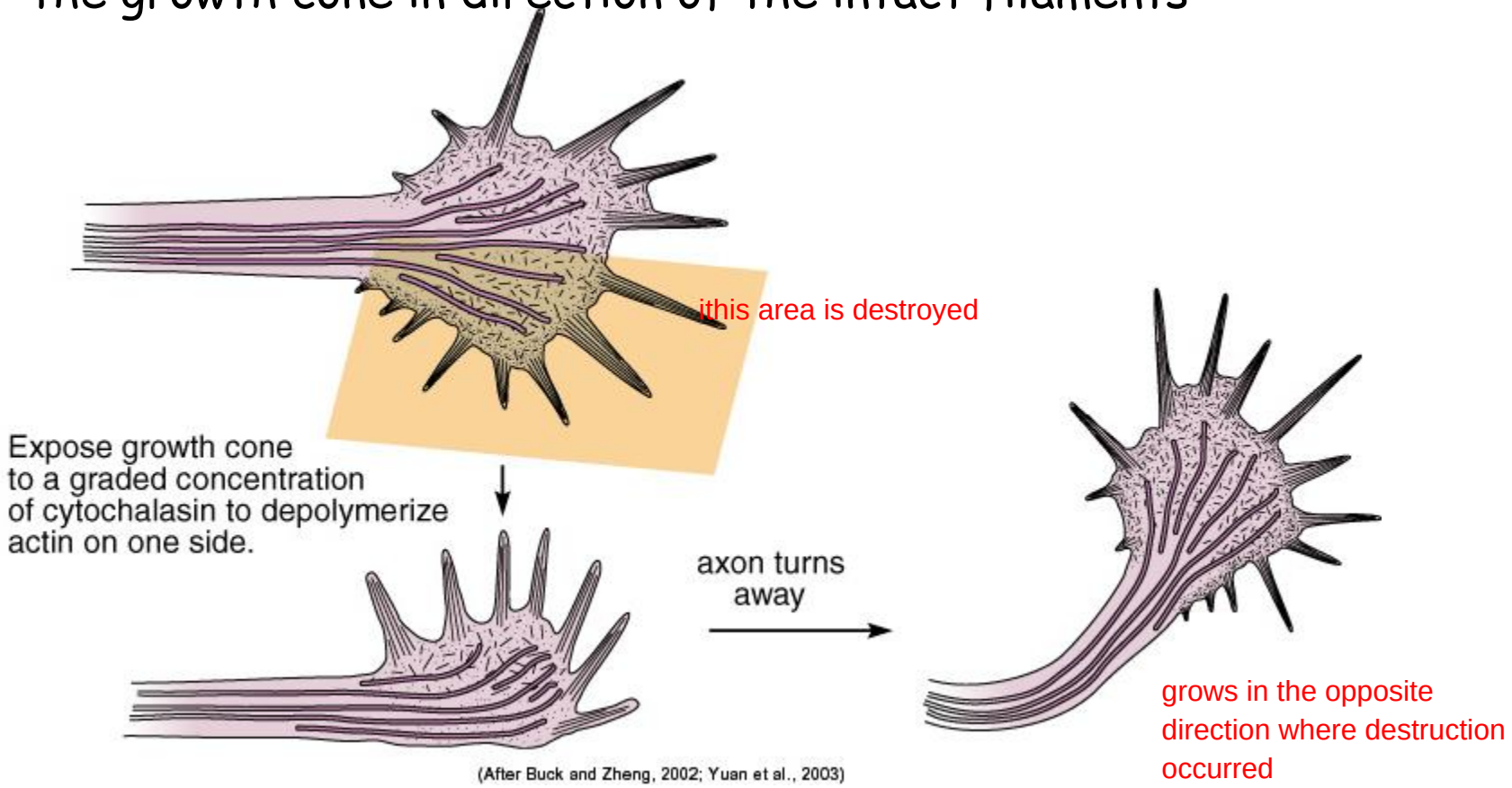
(After Mitchison and Kirschner, 1988)

Sanes, Reh, Harris, 2006

The cytoskeleton steers the growth cone

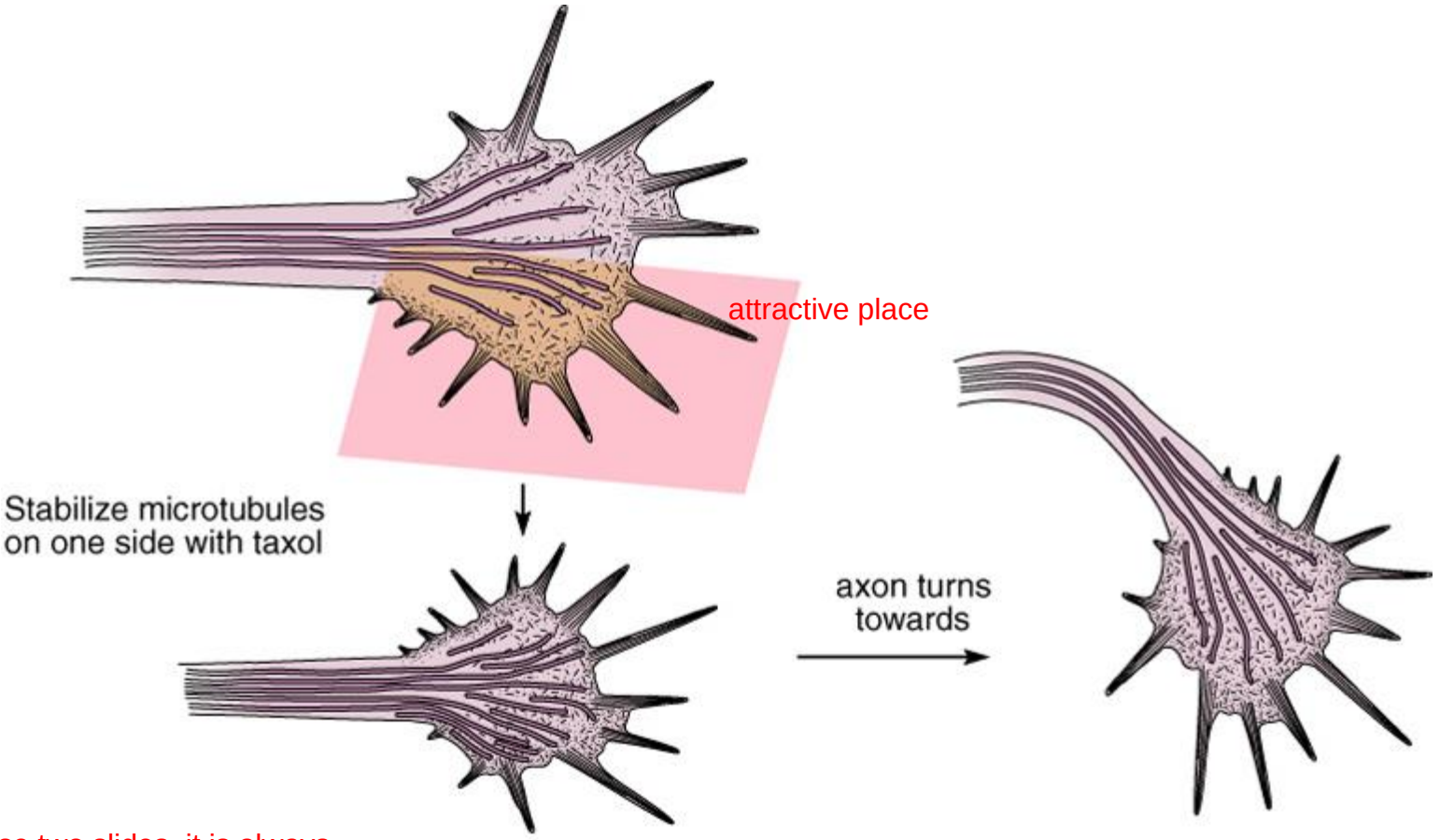
in vitro

Depolymerization of actin filaments induces a turn of the growth cone in direction of the intact filaments



Sanes, Reh, Harris, 2006

Stabilization of microtubules induces a turn of the growth cone in direction of the stabilized microtubules

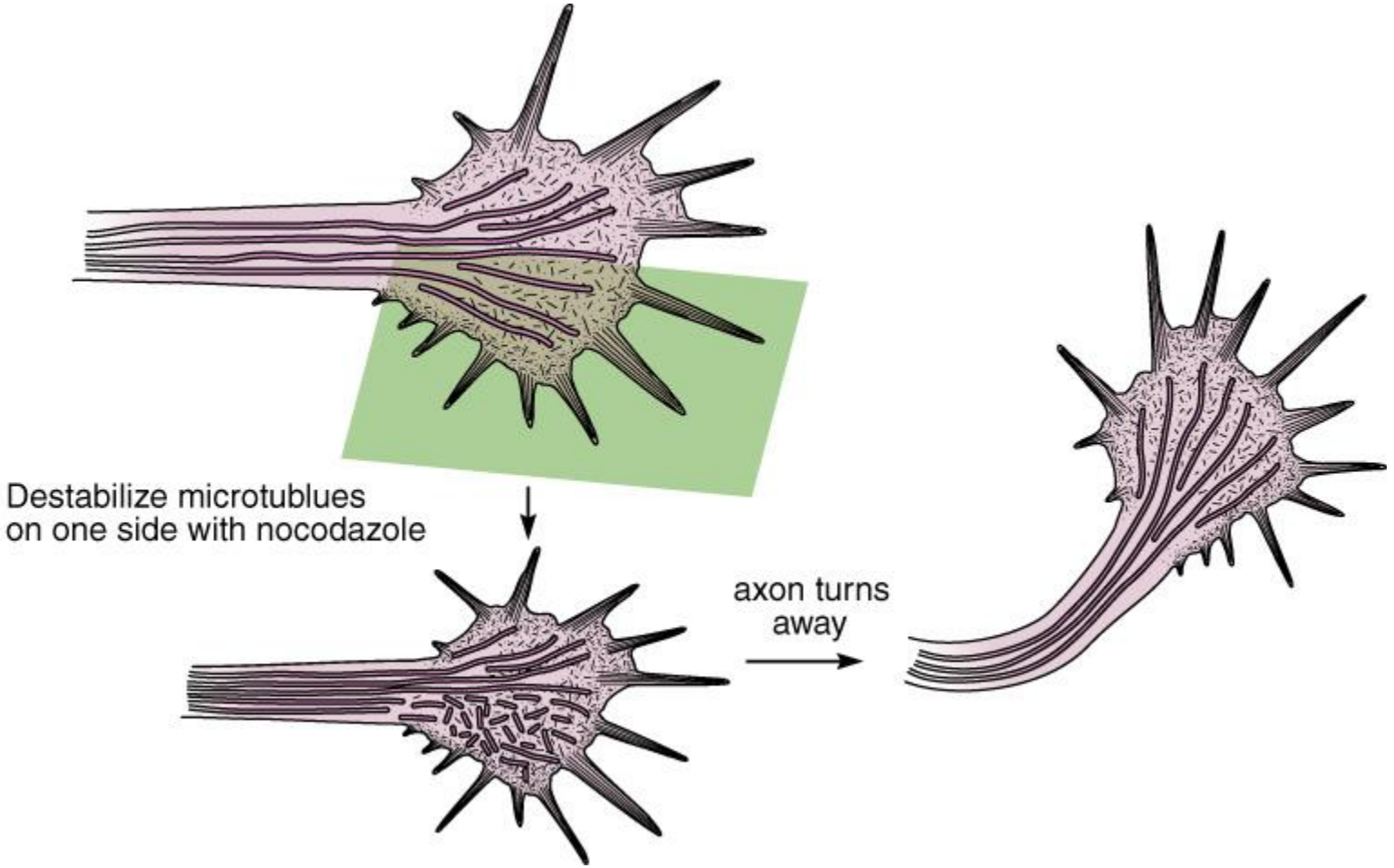


(After Buck and Zheng, 2002; Yuan et al., 2003)

in these two slides, it is always
the microstubules or actin filaments that are involved in the direction of growth

Sanes, Reh, Harris, 2006

Destabilization of microtubules induces a turn of the growth cone in direction of the stable microtubules

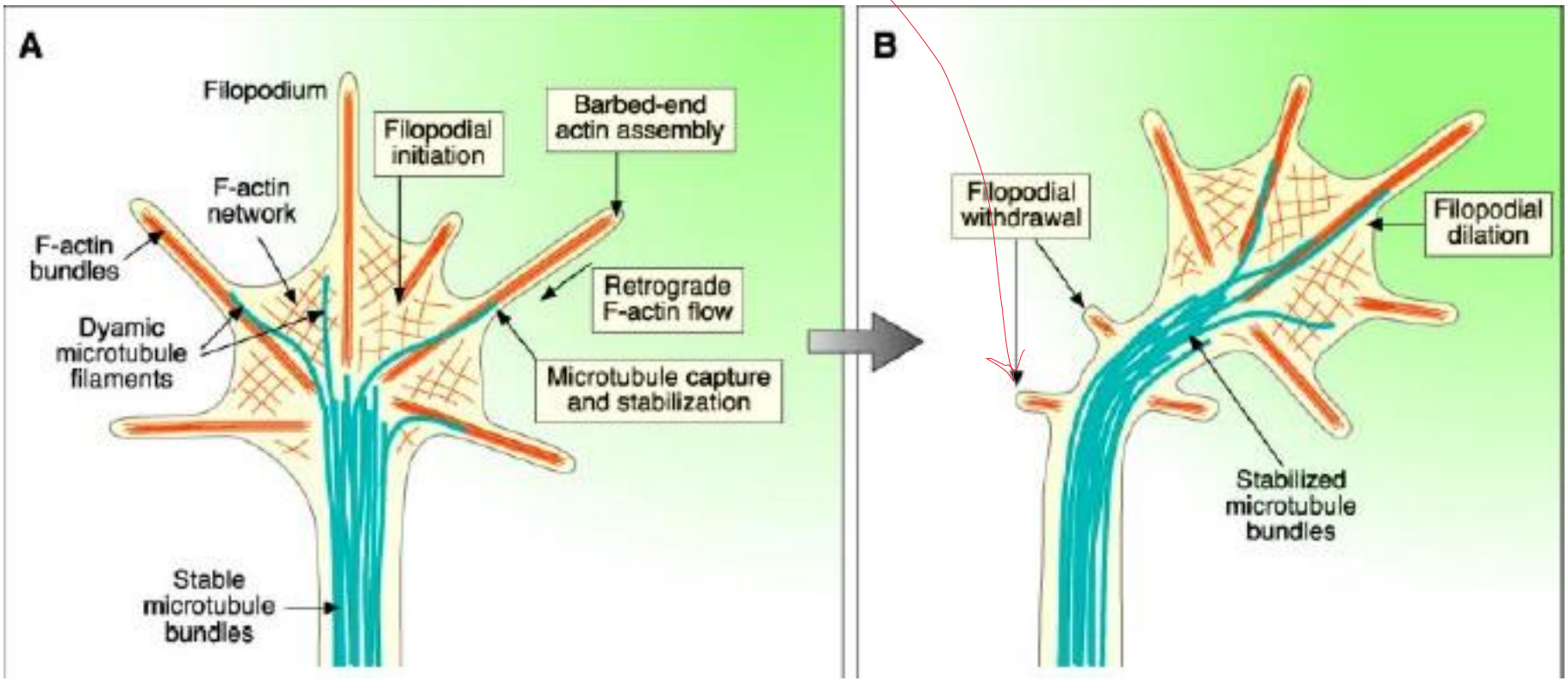


slide left out

Sanes, Reh, Harris, 2006

and in vivo

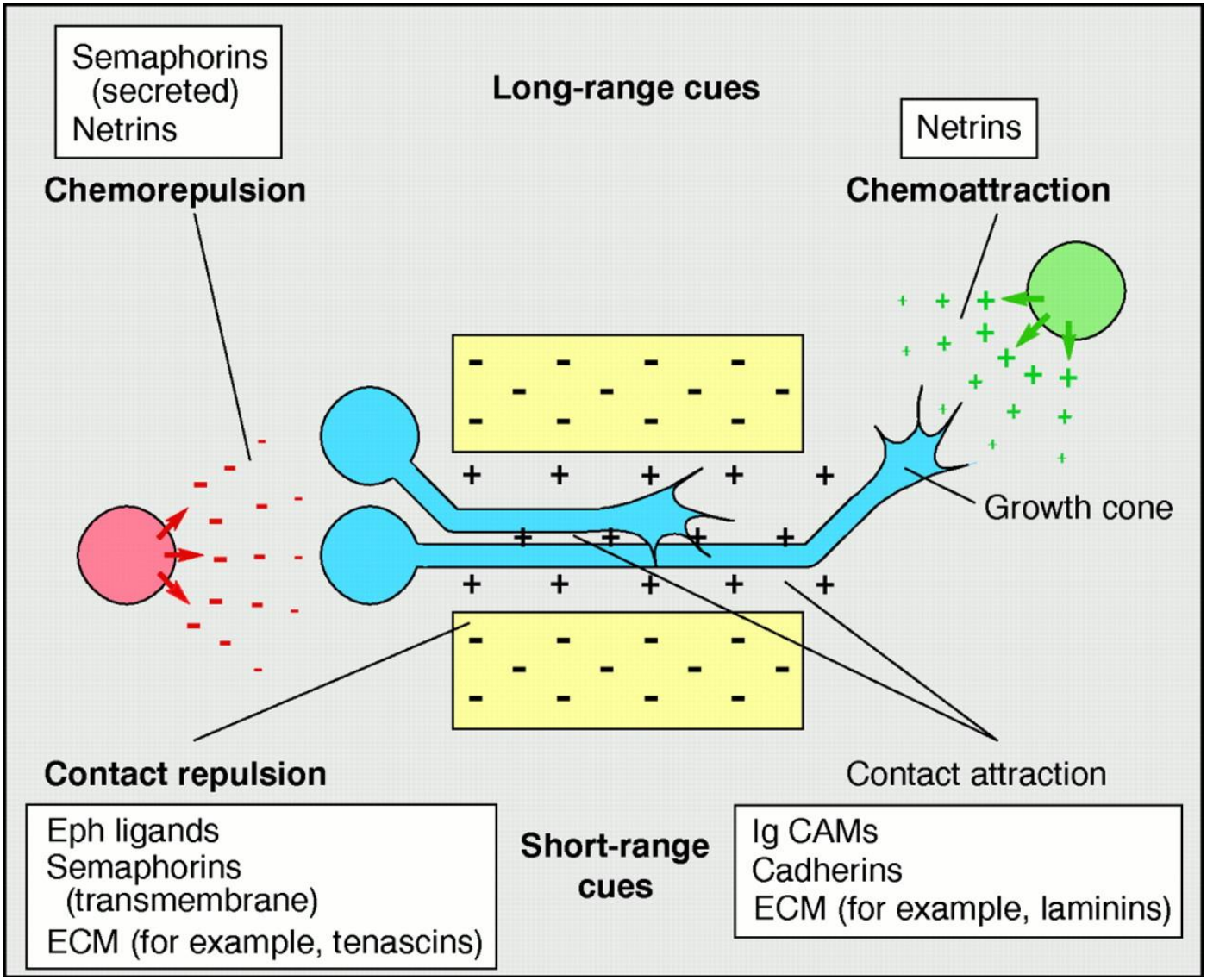
shrinkage or destruction where the growth cone is not supposed to be



Dickson, 2002

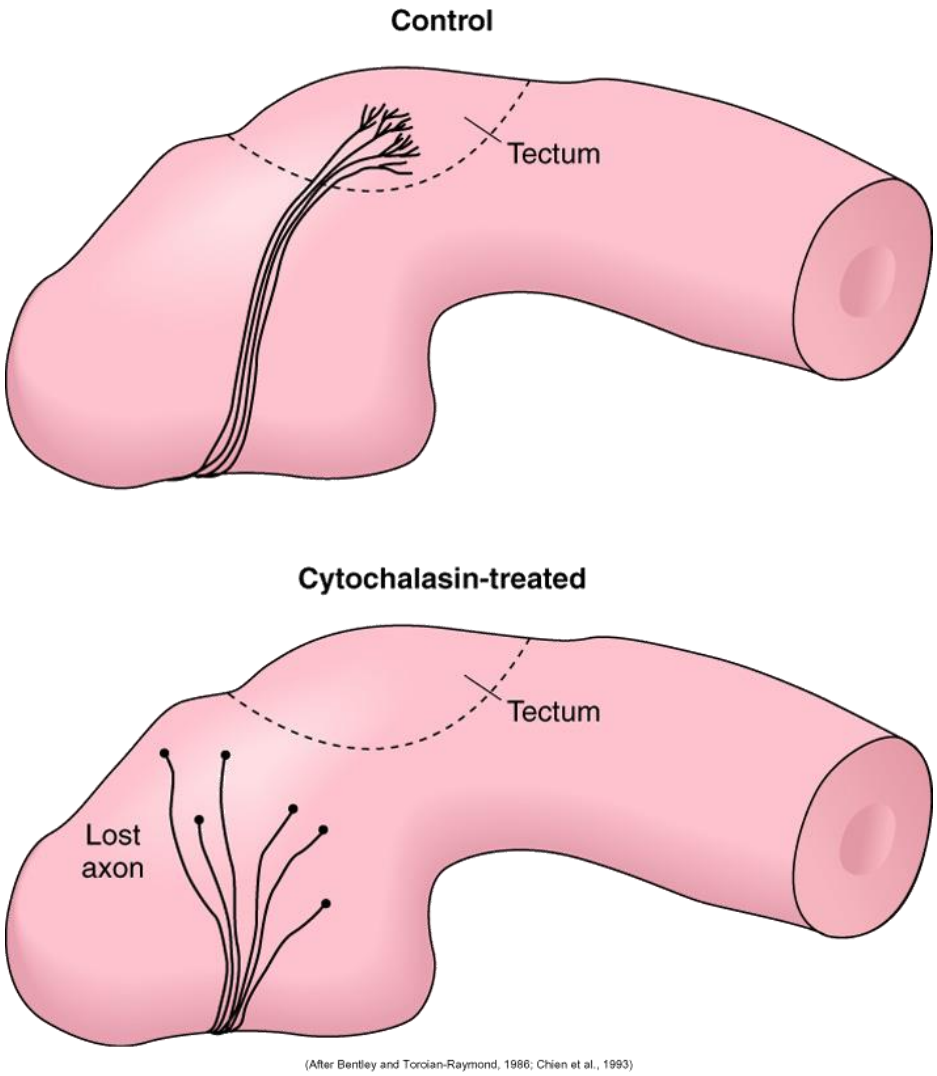
Information derived from the interaction of surface receptors with guidance cues is transmitted to the cytoskeleton

Four mechanisms cooperate to guide axons



Tessier-Lavigne and Goodman, Science, 1996

Actin filaments are required for axon guidance

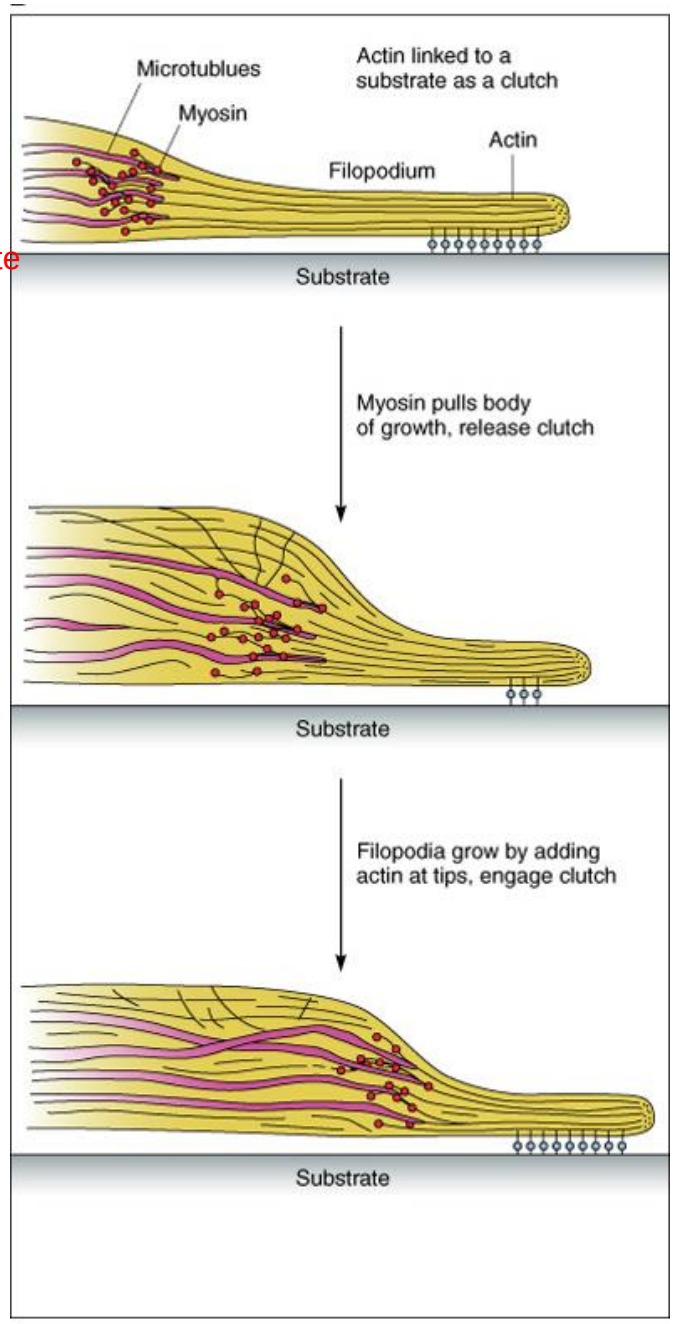


Sanes, Reh, Harris, 2006

information exchange:

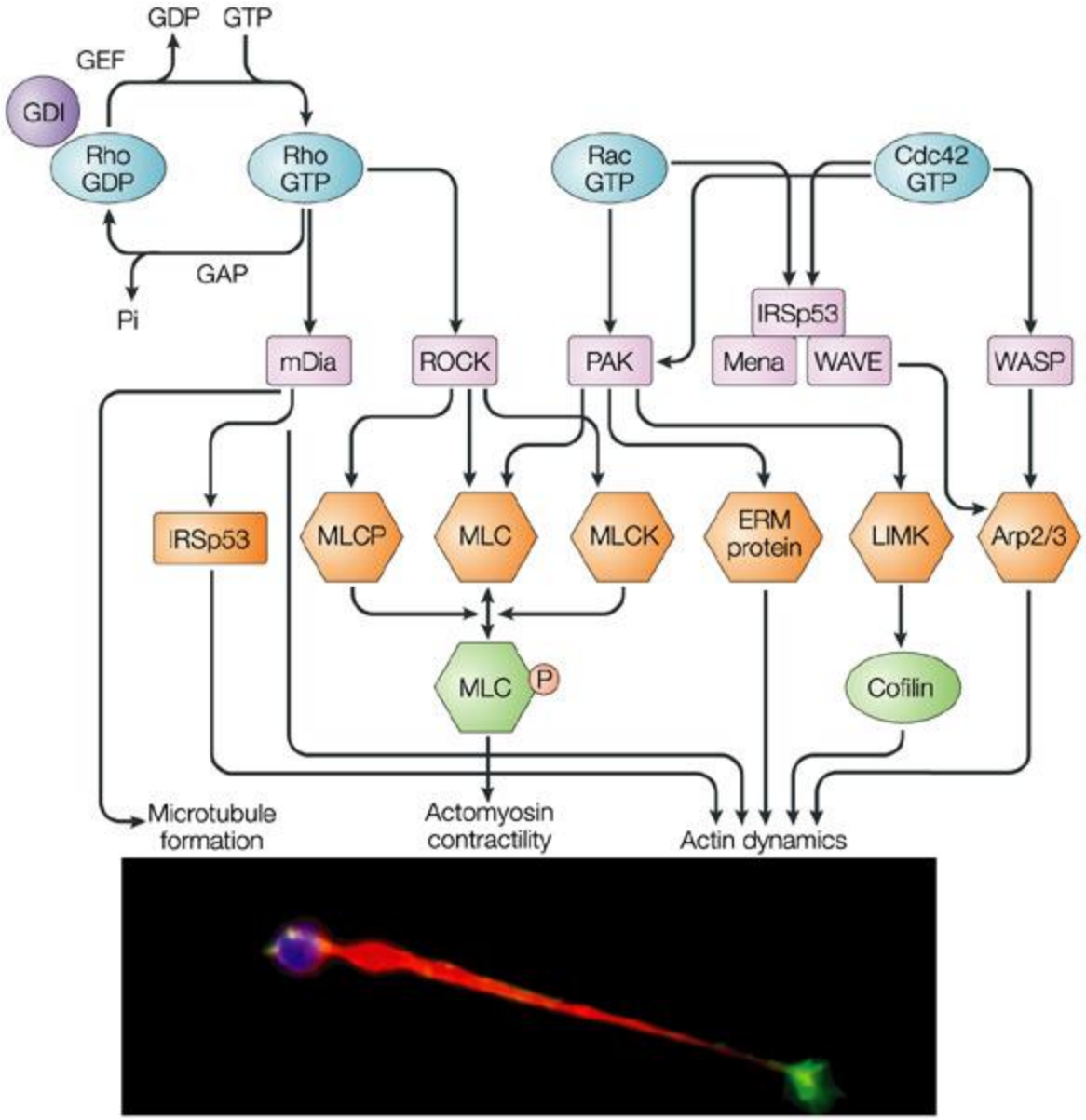
The clutch mechanism

myosin pulls the actin filaments, but filemants are glued to substrate
so the central part of the growth cone has no choice but to grow.



(After Mitchison and Kirschner, 1988)

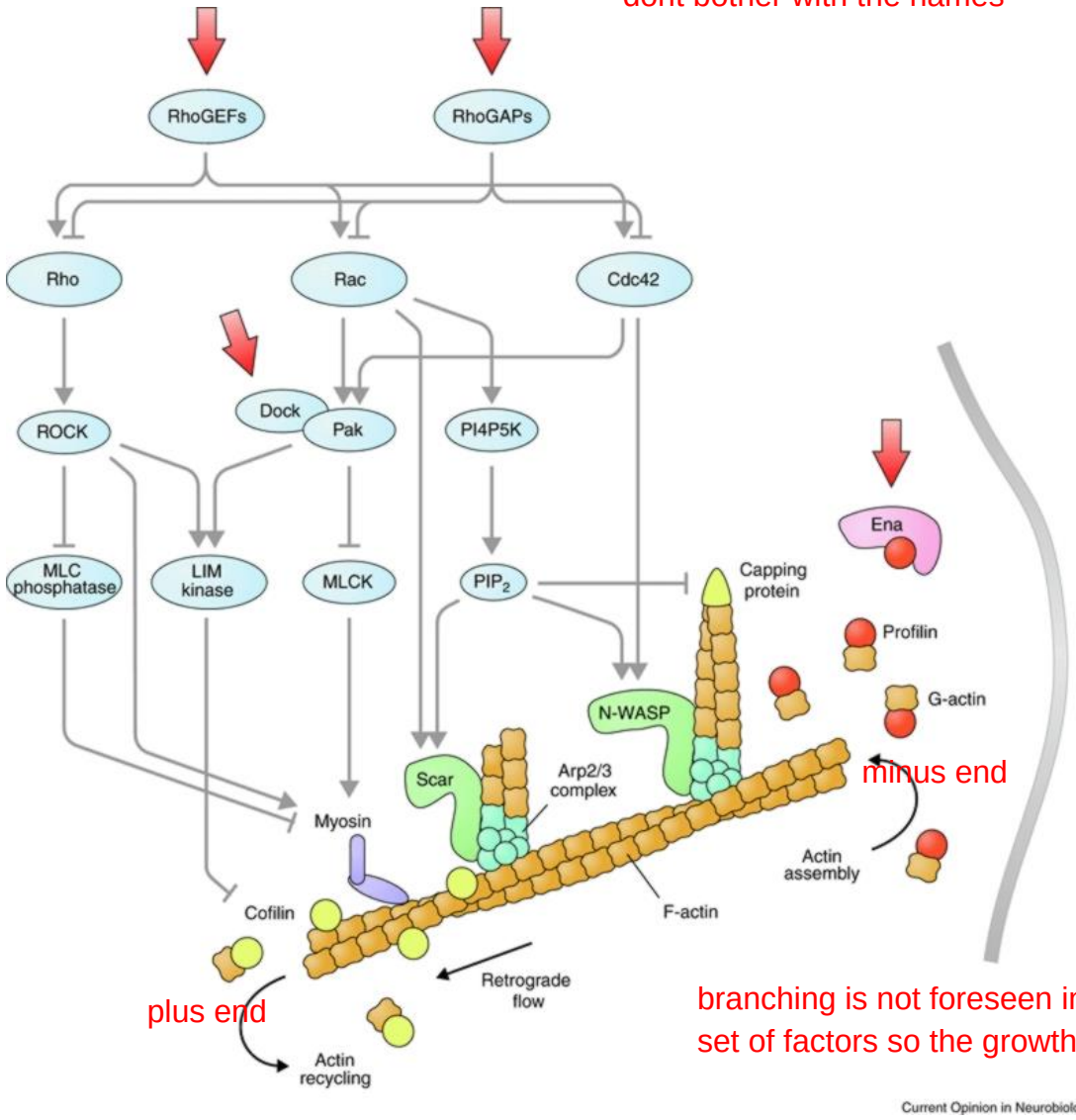
Sanes, Reh, Harris, 2006



Nature Reviews | Neuroscience

Signal transduction pathway linking Rho GTPases to the cytoskeleton

dont bother with the names

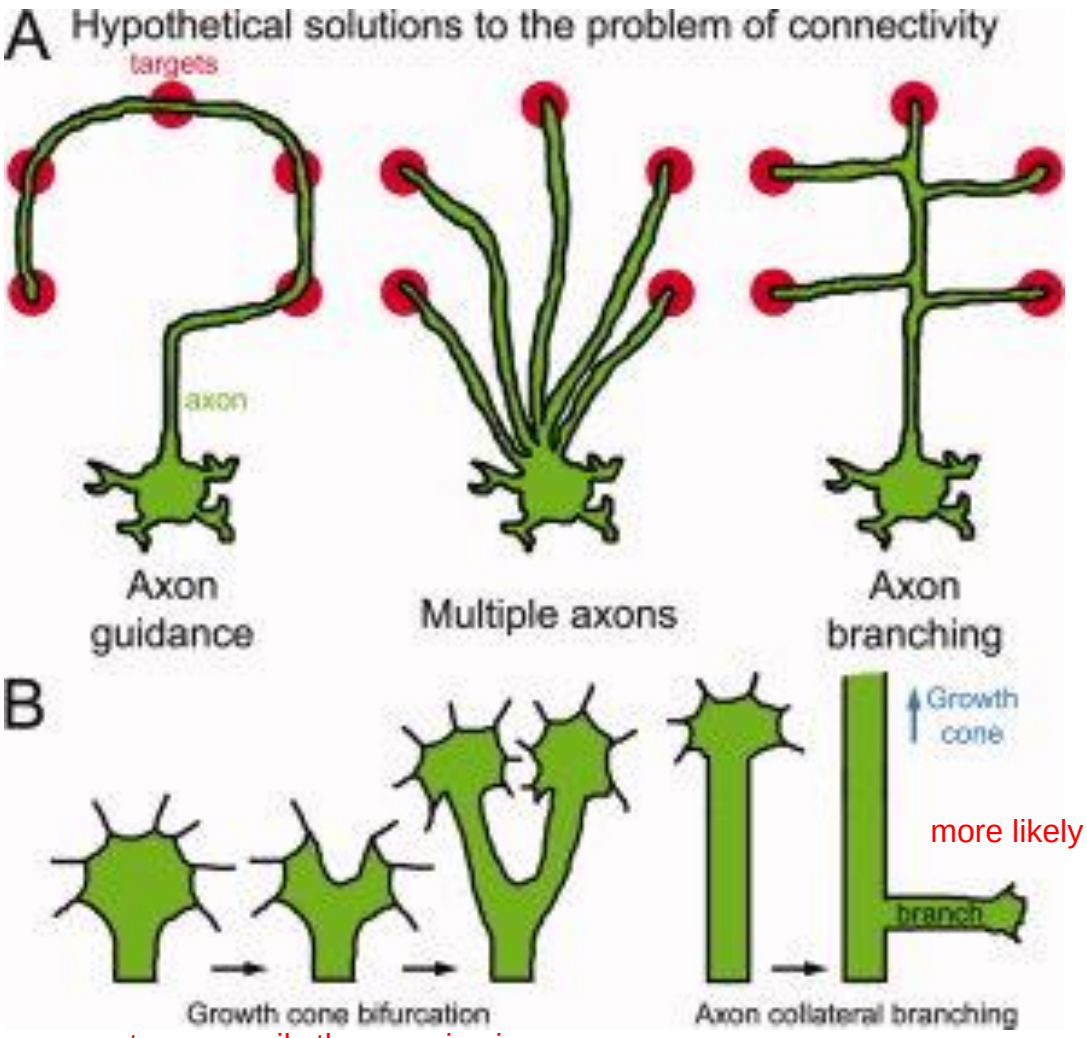


minus end

plus end

branching is not foreseen in this model - needs a whole set of factors so the growth cone can be built

Axon branching is important for neuronal connectivity



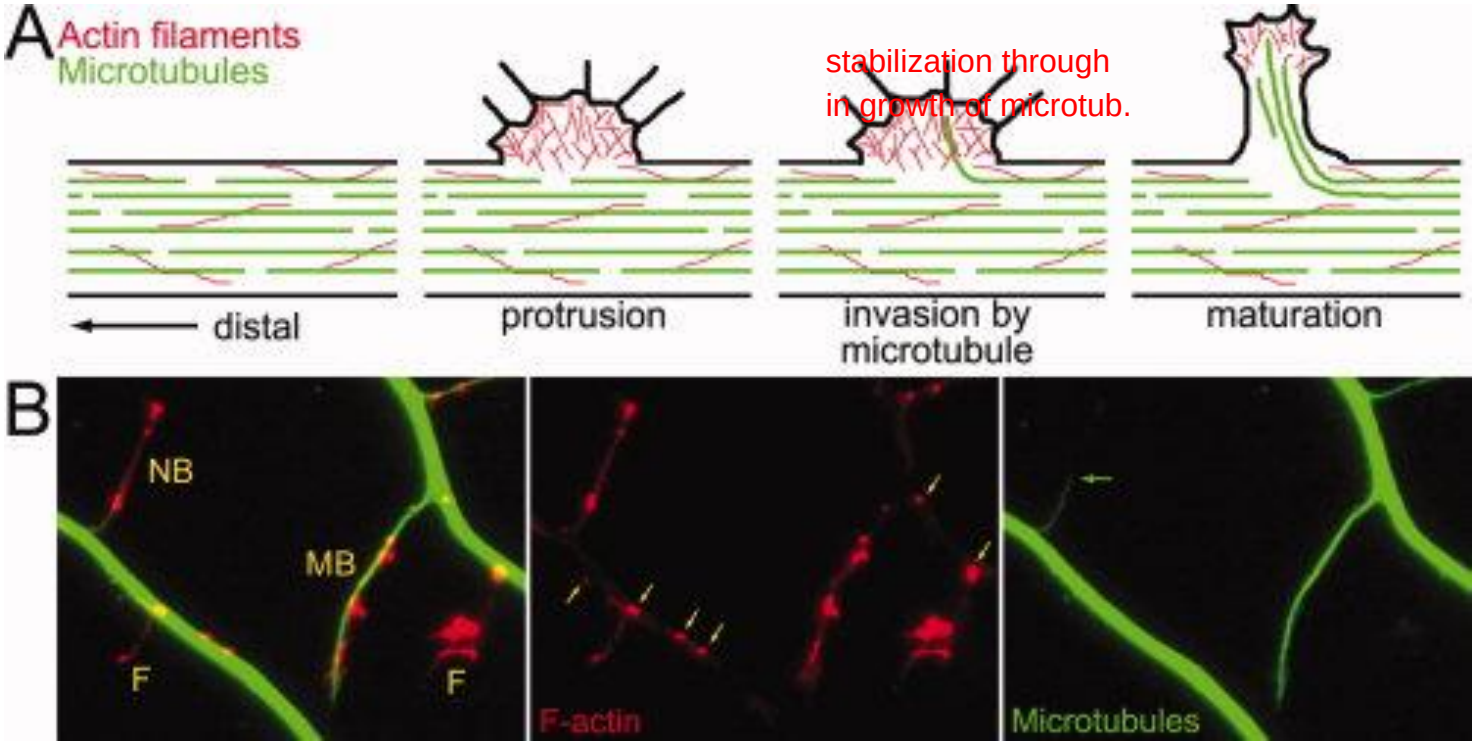
not necessarily the case in vivo

Gallo, Dev. Neurobiol. 71(2011)201-220

Axon collaterals form by a well-orchestrated sequence of cytoskeletal changes

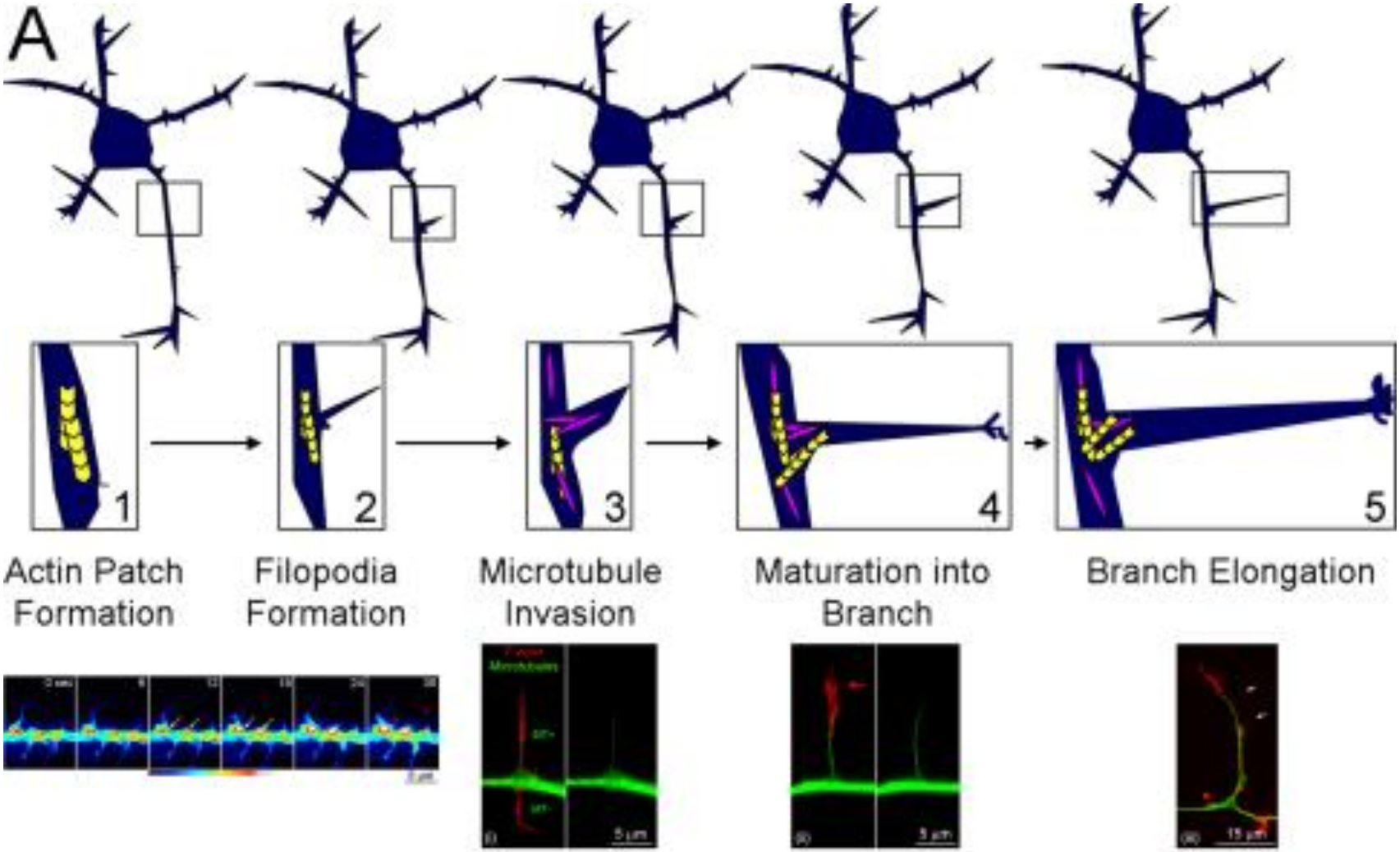
this is the decsion point
where a side branch will
either mature or vanish again

stabilization through
in growth of microtub.



Gallo, Dev. Neurobiol. 71(2011)201-220

Branch formation requires actin filaments and microtubules



Winkle et al., Dev. Neurobiol. 76(2016)1293

next time lecture contents:

Which molecules act as guidance cues (or their receptors)?

How many are there?

Do all axons listen to the same guidance cues?