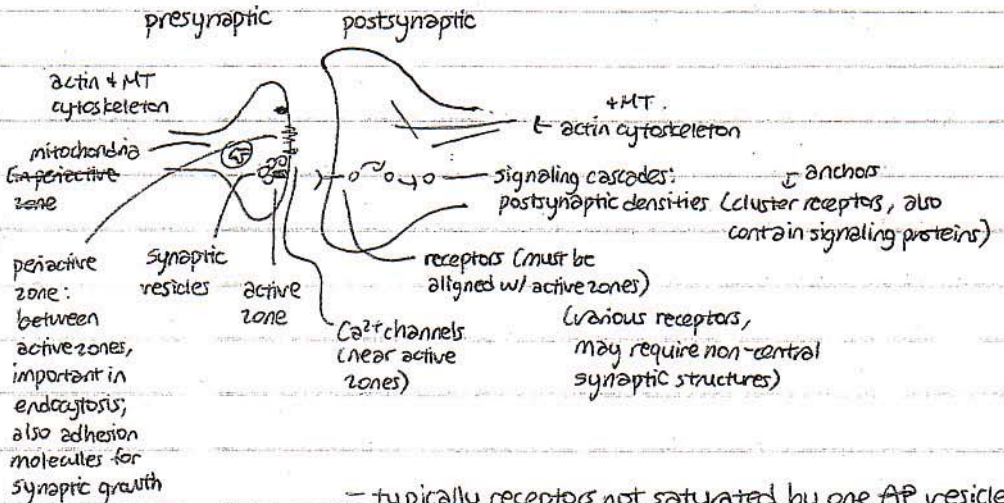
4/12/04

- glia also require trophic factors to survive (from neurons) : need axons to myelinate
- neurons compete for trophic factors

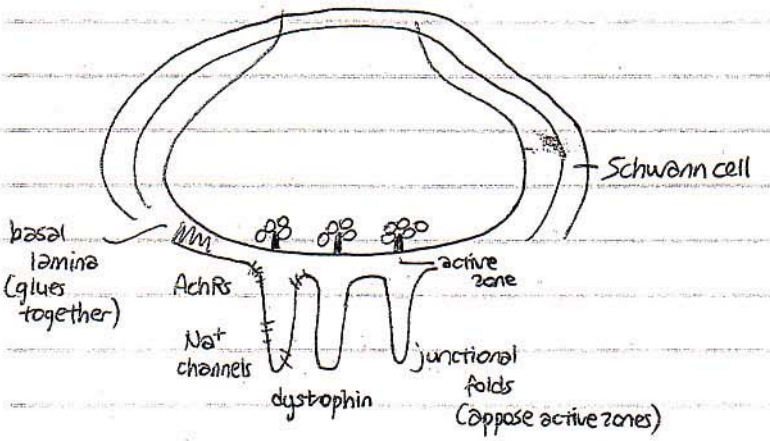
synapse formation:

1. in PNS, especially at NMJ (large, good model system)
2. in CNS (slightly different mechanisms)



- typically receptors not saturated by one AP vesicle (10-300 GluR)
- in NMJ, ~1000 receptors, single vesicle can activate all

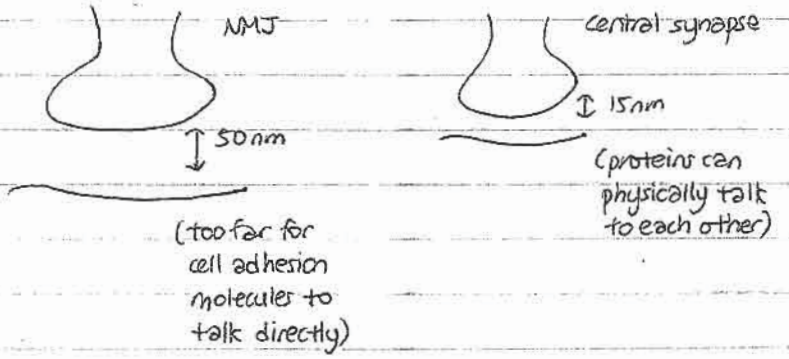
- NMJ useful b/c can culture in vitro



- if grow neuron w/o muscle cell, has active zones & vesicles, can release NT (10/μm² in nonsynaptic regions)
- if have outside-out patch w/ AChRs, can sense this NT release
- motor neuron can have all synaptic components already w/o muscle at mature synapse ↓
- if grow muscle, has AChRs (but no junctional folds, not same AChR density : 1000/μm² vs. 10,000/μm²)

- synapse formation is organization rather than instruction of synaptic elements

- synapse is 0.1% of muscle surface (muscles multinucleated; fused)
  - how to cluster clusters AChRs at synapse?
    - α-bungarotoxin binds nAChRs
      - ↳ fluorescently labeled; looked for neuron-secreted factors that would cluster AChRs (only low number labeled)
    - 3 discoveries from this assay:
      1. clustering factor exists (about 10 molecules found: FGF, laminins, agrin, etc) ← this is right molecule (based on knockout + motor neuron expression)
      2. from in situs, AChR mRNAs much higher from nuclei right under synapse (so clustering could be from high transcription rates of synaptic nuclei): neuregulin
      3. low AChR transcription from nonsynaptic nuclei
  - when nerve contacts muscle, agrin secreted, ~~binds~~ localized to basal lamina, gives clustering

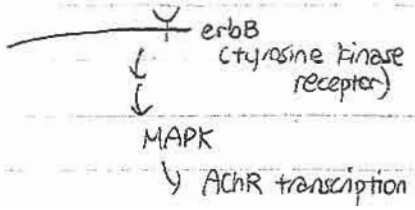


  - agrin binds postsynaptic MuSK (tyrosine kinase): phosphorylation of rapsyn, which binds + clusters AChRs (? →)
    - ↳ cytoplasmic clustering protein (binds AChRs + cytoskeleton)
    - AChRs normally diffuse: w/ agrin KO, no clustering (some spontaneous MuSK-activation normally, small clusters)
    - MuSK KO: no clustering
    - rapsyn KO: no clustering

    w/ rapsyn KO, still get MuSK centrally located
  - increased transcription in synaptic nuclei from neuregulin release from motor neuron

    neuregulin → 0

    ↳ also works in migration of neural crest cells, differentiation into glia



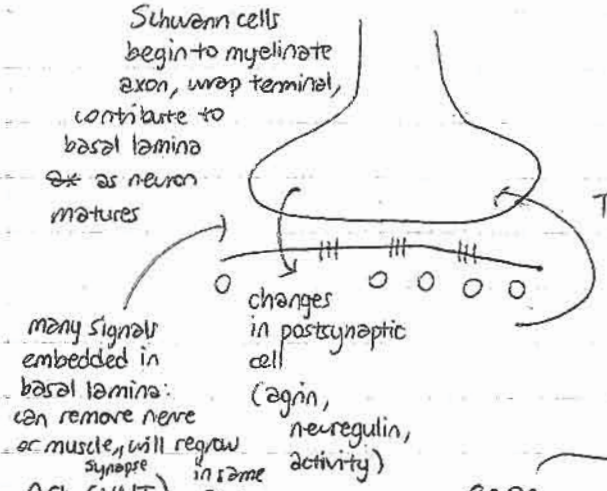
- decreased AChR transcription in nonsynaptic nuclei
  - if denervate ~~nerve, no~~ muscle, get higher nonsynaptic AChR transcription
  - provide electrical activity, repress again: activity itself represses transcription

activity → AChRs → Ca^{2+} ↑ → PKC ⊣ transcription

- junctional receptors, nonjunctional receptors

	junctional	nonjunctional
1. distribution	10,000/μm^2	1000/μm^2
2. different subunits	$\alpha\beta\underline{\varepsilon}\delta$	$\alpha\beta\underline{\gamma}\delta$
3. half-life ($T_{1/2}$)	14 days (stable)	< 1 day (unstable)
4. motility	nonmotile (b/c of rapsyn)	motile
5. function	open for 1 ms	open for 4 ms

- attractive model for plasticity (changes in receptors over time)

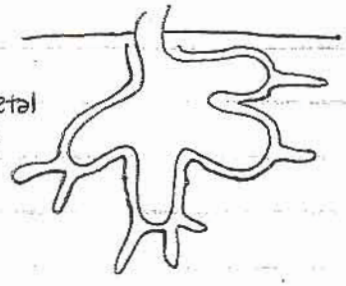


initially, very few vesicles fuse: after retrograde signals, increase in vesicles, active zones, release properties (mT)

central synapses

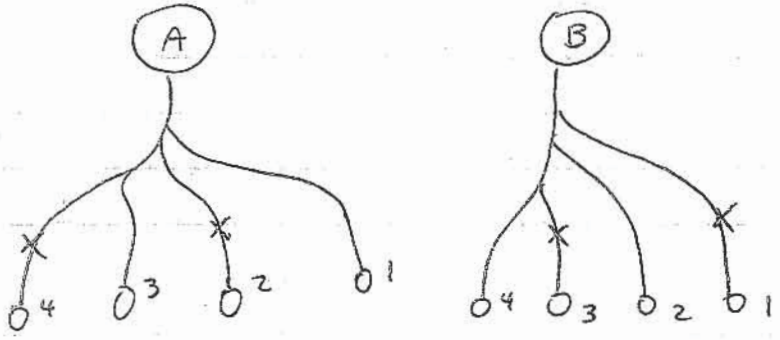
ACh (NMJ)	GABA	Glutamate	receptor anchors
rapsyn	gephyrin	PSD-95, GRIP, PICK (PDZ proteins)	receptor anchors
10,000/μm^2 (1000) 4 per active zone	1250/μm^2 (15-500)	? (10-300)	#Rs/active zones
yes	?	no (implications for plasticity)	single vesicle saturation?
~~junctia~~ junctional folds	no PSD (electron-dense scaffold)	PSD (electron-dense)	postsyn. specializ.
Schwann cell	astrocyte (ensheath synapse, contains transporters to get rid of NTs from cleft)	astrocyte (use EM to see if excitatory or inhibitory)	synaptic cleft
50nm	15nm	15nm	glia
		AMPAR, NMDAR bind different PDZ proteins for clustering	

- multiple redundant mechanisms in synapse formation in CNS



synaptic elimination:

- muscles initially polyinnervated; motor neurons compete, all but one withdraw
  - if inhibit muscle activity, eg w/ α-bungarotoxin, no competition: more than one stay
  - if block synaptic activity of one axon, it will lose (eg by dom. neg.)
  - if increase activity of one axon, it will live (eg by altering electrical properties)
- assures potent connections (strengthen surviving connections)



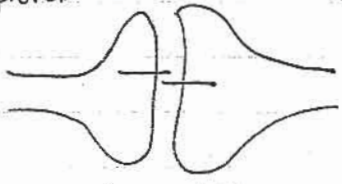
neurexin + ~~neuroglian~~ neuroligin: ligand/receptor pair

neurexin

presynaptic ECM protein

cluster synaptic vesicles, Ca^{2+} channels

(molecular scaffold, binds PDZ proteins)



(many different classes of cell adhesion molecules) so can synapse on different partners

~~neuroligan~~ neuroligin

~~neuroglian~~

receptor w/ AChE-like domain, postsynaptic (binds PSD-95, which clusters GluRs)

molecular scaffolds assembled

7.29J / 9.09J Cellular Neurobiology

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