ZOO332H1S – Lecture 9b (AJE 2003)

Part 1: **Properties and Functions of Neuroglial Cells (Ch. 8)**

Part 2: **Spatial Organization of Sensory Inputs and Another Level of Complexity in the Well Modulated Cockroach**

1

Neuroglial cells in mammalian brain and PNS

- **Principal types**:
  - astrocytes – neuron-capillaries
  - oligodendrocytes
  - radial glial cells – development; and as adult (Bergmann and Muller cells)
  - microglia – controversial; wandering, resemble macrophages in the blood
  - ependymal cells – line ventricles
  - Schwann cells
- **"Satellite" Cells**

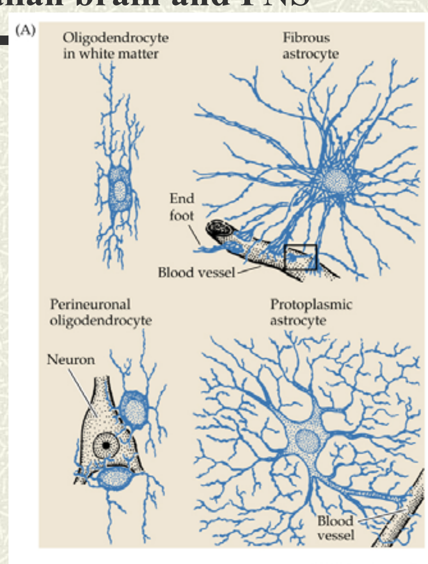
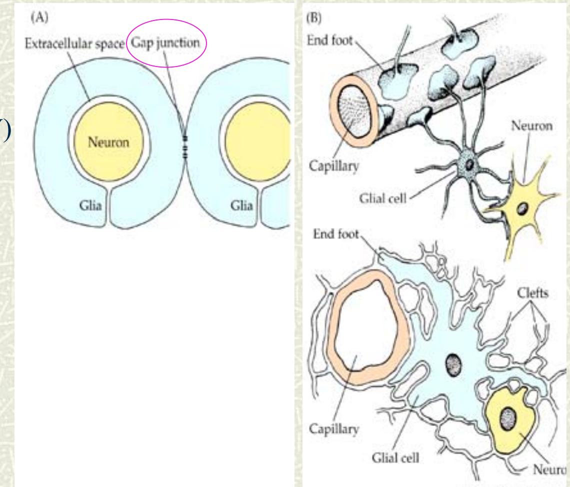


Fig. 8.1

2

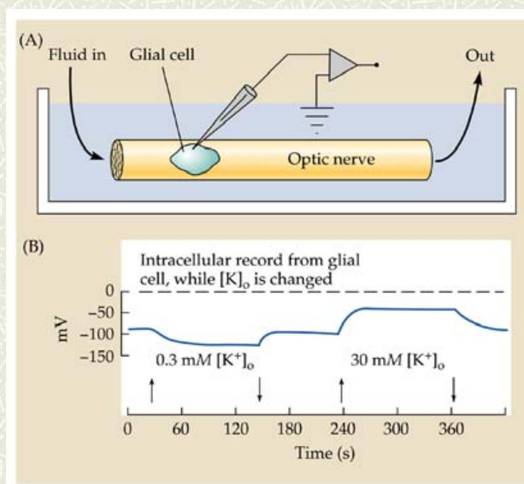
Connectivity amongst glia, neurons, and capillaries

- Ground work done in simple models first
- large resting potential (-90mV)
- connections
- channels for K^+ in glial cell membrane



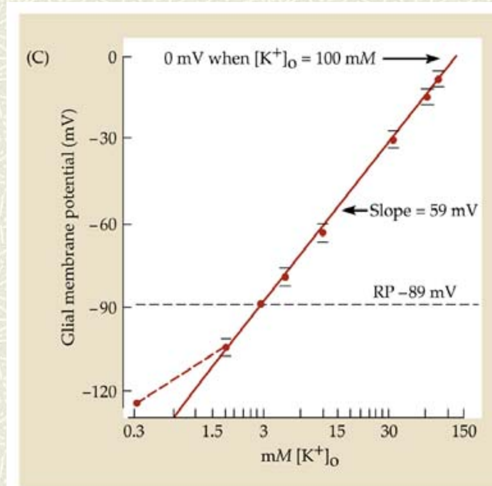
Recording from glial cells

- Recall Nernst equation and use
- permeability ratio of K^+: Na^+: Cl^- (1: 0.03: 0.1) in neurons
- Muller cells K^+: Na^+ (1: 0.01)



cont…Recording from glial cells

- Data plotted as change $[K^+]_o$
- Prediction by Nernst equation
- Note RP
- Assortment of channels, ion pumps, transporters (Glu, GABA, Gly), receptors



5

Functions of Neuroglial Cells

Myelination of axons (use of culture systems)

During development (and continues)

Example: PMP22 (peripheral myelin protein 22)

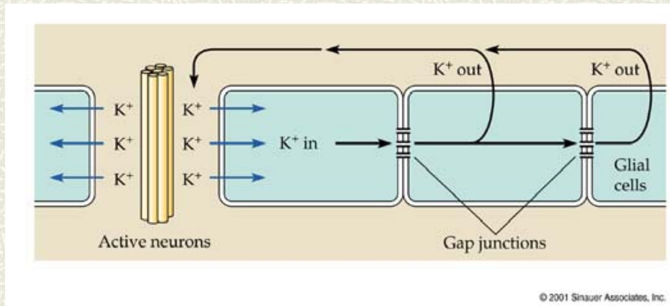
Glial cells influence positioning of Na^+ channels in nodal region

Microglial cells – respond to damage

Schwann cells – can guide axons and promote outgrowth

6

Role of glial cells in K+ homeostasis



cont…Functions of Neuroglial Cells

During development

- Groupings of neurons into nuclei – these nuclei are first outlined by glial cells (neurons arriving later)
- Radial glial cells used to guide migrating neurons (in cerebral cortex, hippocampus, cerebellum) (next slide)

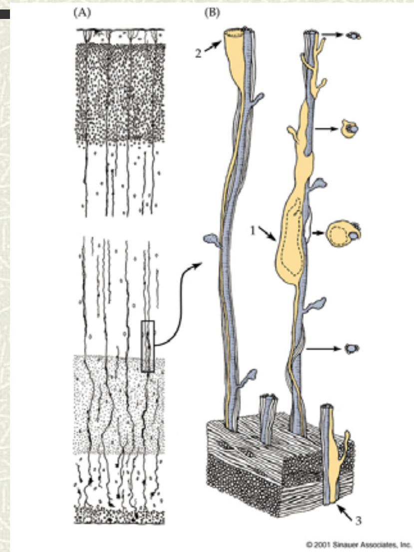
Neurons migrating along radial glia

(A) Camera lucida drawing of migrating neurons and radial glia in cortex

(B) Schematic of the process

Neuron follows chemical cues along glial surface membrane to arrive at cortical layer

Axons sent out and synapses formed



Schwann cells as paths for outgrowth in PNS

Interesting example although special case (?)

Adult rat soleus muscle

Partially denervated

Schwann cell locates endplate on adjacent muscle fibre

Induces sprouting and migration of axon

Functional innervation

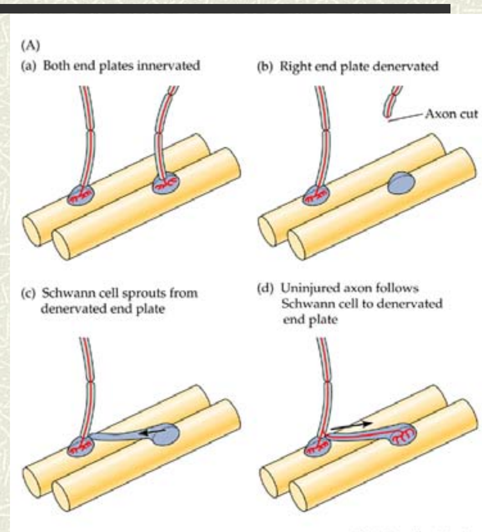
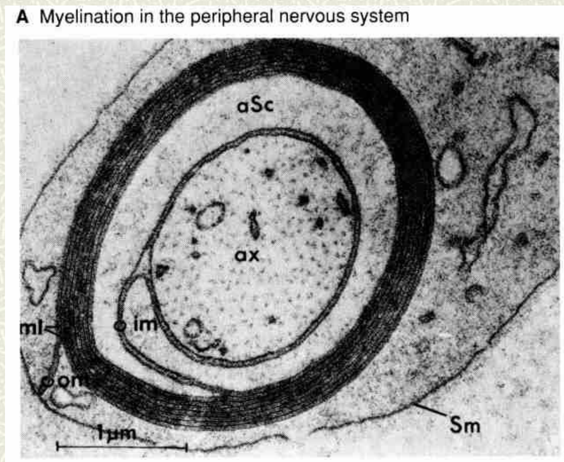


Fig. 8.12

Schwann cells and interaction with peripheral axons

- **Myelin sheath (ml)**
- **Inner mesaxon (im)**
- **Surface membrane of Schwann cell (Sm)**
- **Outer mesaxon**
- **Schwann cell cytoplasm (aSc)**
- **Axon (ax)**



11

Schwann Cells - the basics

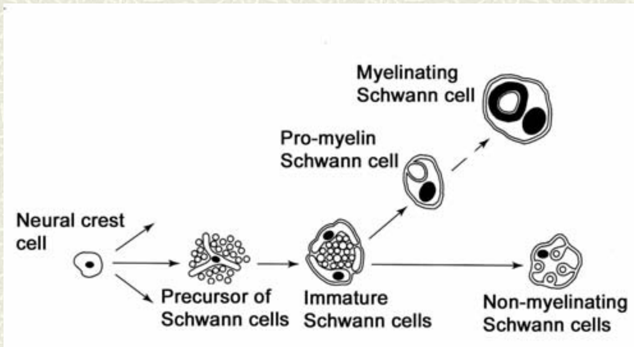
- peripheral glial cells - myelinating and non-myelinating
- reliance on signalling from axons
- neuron-derived signals during development and when mature
- new evidence supports glial-derived signalling as critical for neuronal survival during specific periods of development
- regulate molecular and f'al specialization's of axons; maturation of perineurial sheath

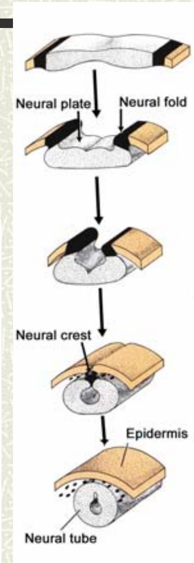
Jessen and Mirsky, 1999

12

Schwann cell lineage

1. Formation of Schwann cell precursors
2. Formation of immature Schwann cells
3. Commitment: myelinate or non-myelinating



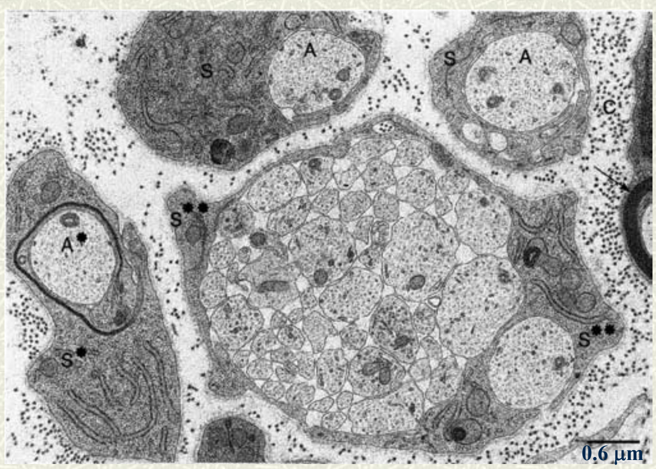


13

Jessen & Mirsky, 1999; Gilbert, 1997

Schwann cell - Sciatic nerve newborn mouse

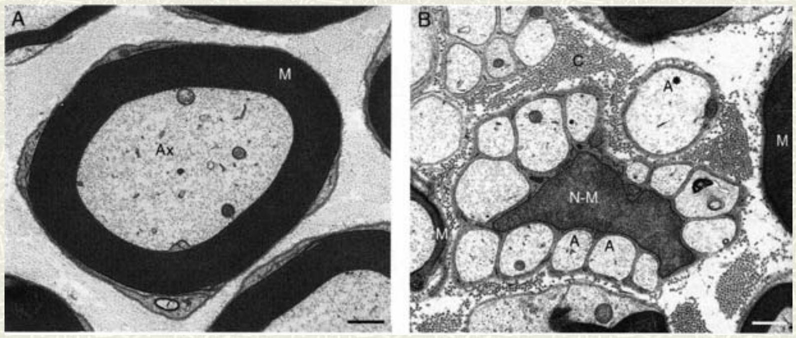
- **S**** - immature Schwann cells ("communal corral")
- **S** - pro-myelination stage (1:1 with **A**xon)
- **S*** - sheath beginning around **A***
- **C** - collagen



Jessen and Mirsky, 1999

14

Mature Schwann cells - Transverse section sciatic nerve



(A) classic profile of myelinated axon (and portions of others) **0.5 μm**

(B) nucleus of non-myelinating Schwann cell **(N-M);** 13 axons ensheathed, each inter-digitated and surrounded by the Schwann cell. Also, non-myelinating SC around single axon profile (**A***).

Jessen and Mirsky, 1999 15

Summary points on Schwann cells

- glial lineage arises from neural crest (NC) cells
- major peripheral myelin protein (Po) found to be earliest glial cell marker (found in migrating NC cells)**
- Po expression modulated by axons (up and down…)
- narrow window for transition from precursors to Schwann cell (E14/E15--E17 rat (mouse E15)) ("no" precursors in mature nerves)
- β-neuregulins (axonal) bias NC cells to differentiate to glial cell, although some controversy needs to be resolved

****Bhattacharyya *et al*. 1991; Lee *et al*. 1997; *etc*.** 16

cont. summary points…1

• dependence on signalling from axons for survival (β-neuregulin)

• Evidence: *in vitro* cultures and KO's, β-neuregulin essential for precursor cell survival and the change from precursor to glial cell

• period from about birth to 3 weeks get final differentiation step

• membrane synthesis, up and down regulation of genes

• transection of nerve leads to changes which revert glial phenotype to immature state

• environment formed which would promote axonal re-growth

...and new evidence from new technology

17

cont. summary points…2

Knock-out of *Erbb3* gene

• a major receptor for β-neuregulin in crest cells and early glia

• initially number of DRG and motor neurons normal during embryogenesis (*ca*. E12)

• these mice **<u>lack</u>** Schwann-cell precursors and Schwann cells

• by E14, 80% of DRG neurons lost; by E18, 80% motorneurons were lost (as late as E16 all OK)

• chimeric experiments (Erbb3 in neurons but not 'glia')

18

cont. summary points…2

• **DRG** - **RIP** - too early to receive trophic signals from targets

• **motorneurons** – last until E18 then die – why?

* initial survival and migration to target independent of signals from immature glial cells

* BUT: target-derived and glial signals required for survival

* Note timing: link to transformation of glial precursors to immature glial cells usually occurs just prior to E18

19

Myelination - Essential for efficient, rapid conduction of APs

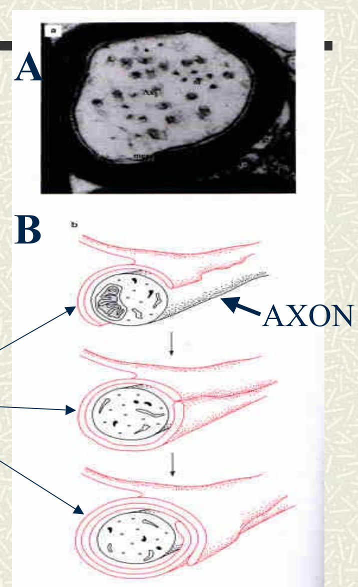
(A) electron micrograph

(B) formation of myelin sheath

Oligodendrocyte (CNS)

Schwann Cell (peripheral NS)

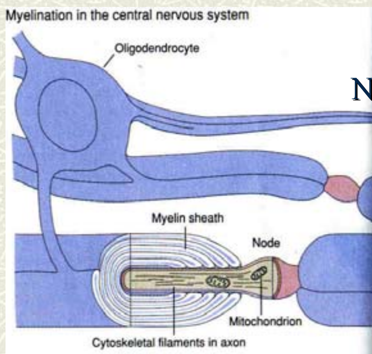
- mature myelin sheath: only most inner and outer layers contain significant amount cytoplasm

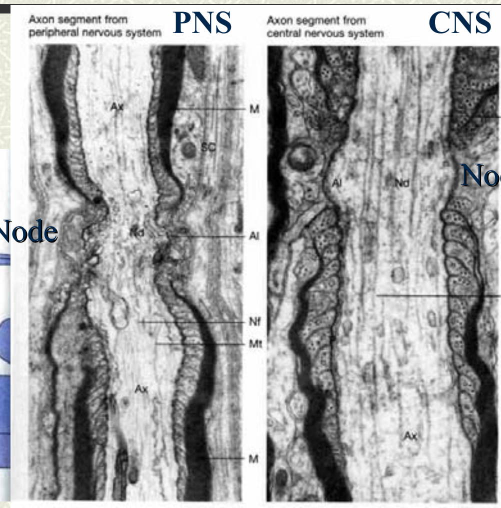


20

Myelination - electron micrographs

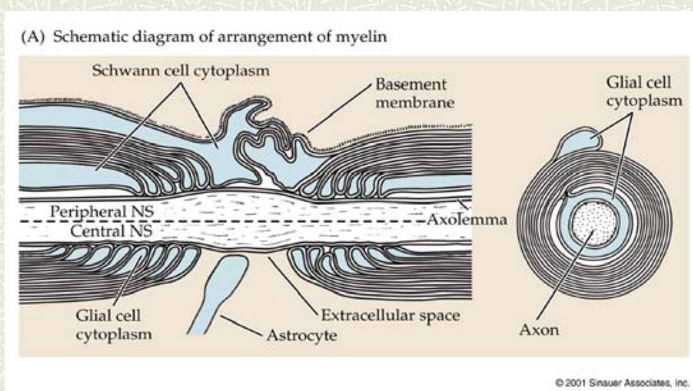
Diagrammatic





21

cont…Myelination



22

cont. Myelination

- Myelin interrupted at nodes of Ranvier (1 - 1.5mm spacing)
- Measurements made indicate CV for fibres > 11µm is 6 times axon diameter; fibres <11µm about 4.5 X
- Balance: thickness of myelin (increases R) and cross-sectional area of axon (decreases - causes increase in internal longitudinal R) - compromise: axon diameter 0.7 x overall fibre diameter
- Distance between nodes optimized

23

cont. Myelination

- Single Schwann cell makes myelin in one internode region (*ca*. 500 needed for single peripheral axon); oligodendrocyte can do several axons
- Formation of myelin by Schwann cells appears to be axon dependent-signaling; oligodendrocytes rely on astrocytes for signaling
- Myelin Basic Proteins - found in both; group of 7 related proteins (alternative splicing variants)

24

cont. Myelination

• Classic experiments done by Ritchie and co-workers (mostly on rabbit nerves)

• Location of V-gated channels - **not** what you might expect!

or
  • Na^+ channels conc'd in nodes of Ranvier; none paranodal

  • K^+ channels conc'd under sheath (between nodes)

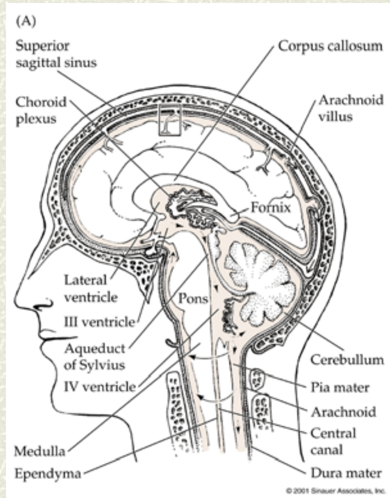
• V/C showed nodes displayed only inward currents and repol'n **NOT** by an increase of G_K+ - then what?

• Chronic demyelination by **diphtheria toxin** - Na^+ channels eventually populate demyelinated region and then get continuous conduction through the area, but poor substitute

25

The blood brain barrier (BBB)

- 3 main compartments
  - Blood in capillaries
  - CSF surrounds bulk of NS, contained in ventricles
  - Intercellular clefts

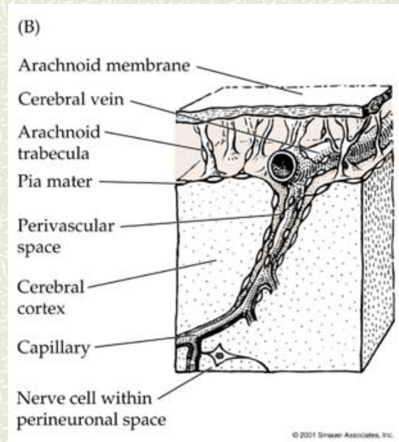


•Endothelial cells of capillaries specialized to be less permeable
•Most substances blocked; not lipophilic or gases (dissolved)
•Choroid plexus: specialized epithelial cells surround cp capillaries. These cells produce and secrete CSF.
•Intercellular clefts (20 nm): gateway to neurons

26

cont…The blood brain barrier (BBB)

- Fluid movement thru intercellular spaces, not thru glia (experiment: inject HRP into, product from peroxidase rx electron dense, look at distribution)



Parting shots at glial cells

Glial cells act to separate individual or groups of neurons

Help regulate $[K^+]$ in extracellular environment

Transmitters can act on glial membranes – role ?

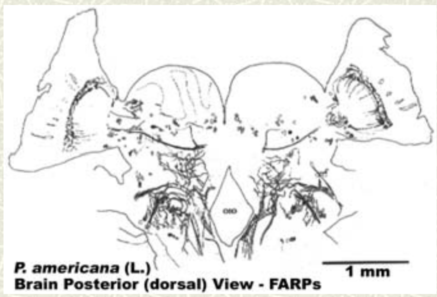
Glutamate transporter in glial cells

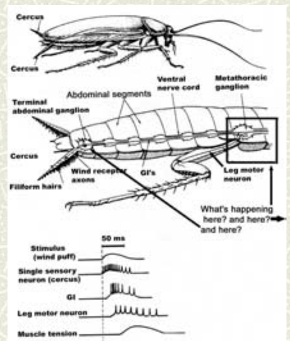
What if persistent high $[Glu]_o$? (mice that lack gene for astrocytic glu-transporter (GLT-1) develop epilepsy and increased susceptibility to convulsants)

Spatial Organization of Sensory Inputs and Another Level of Complexity in the Well Modulated Cockroach

(Further Exploration of "Simple" Nervous Systems)

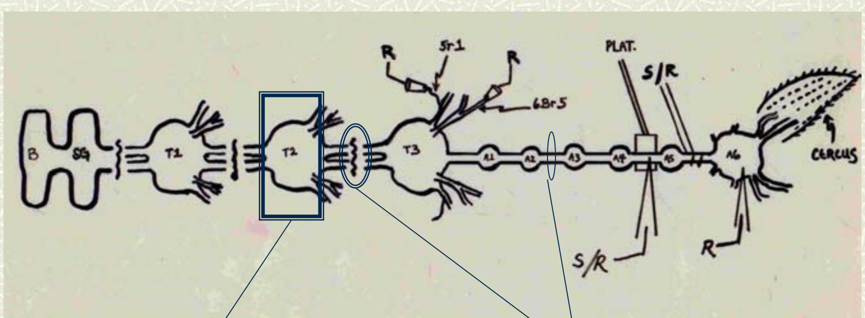
AJE





29

Reminder of Insect (Cockroach) CNS

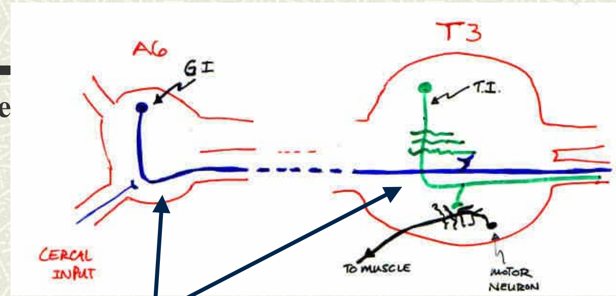


Ganglion - synapses; cell bodies; dendrites; tracts course thru some giving branches

Connectives - bilaterally symmetrical; carry axon tracts ("highways"); no cell bodies or synapses

30

GI's indirectly drive leg motor neurons via thoracic interneurons ("TI's")



…potential for neuromodulation...

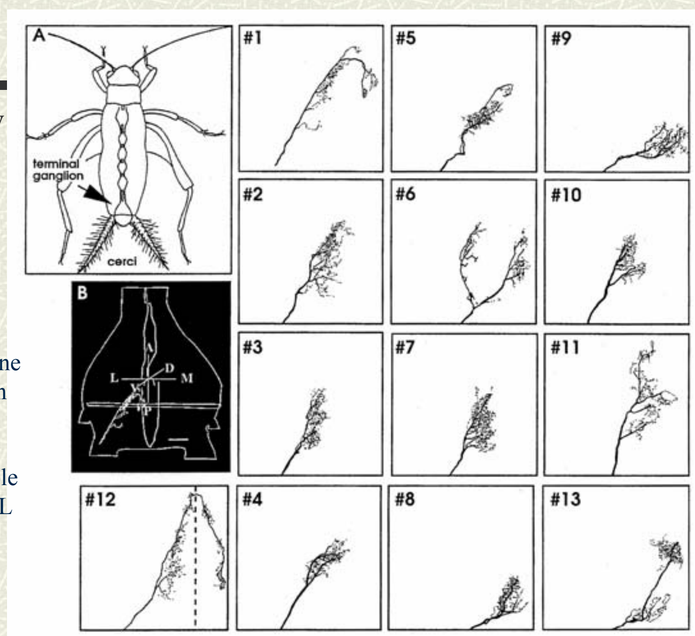
What's happening (can happen) here?

Could the <u>brain</u> be involved in such a simple organism?

31

Cercal sensory afferents in cricket terminal abdominal ganglion

- Dorsal view, midline on right side of each diagram
- M afferents shown
- uniquely identifiable and corresponds to L afferent

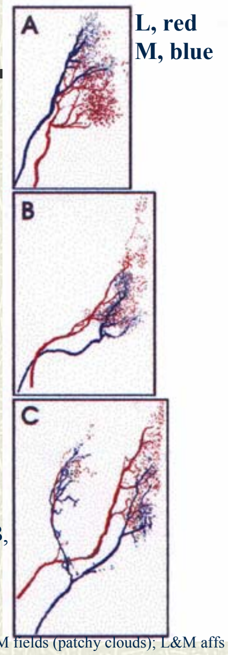


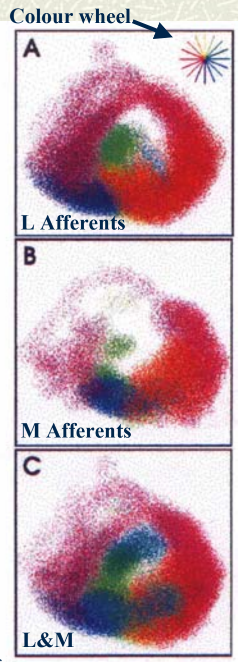
Paydar et al. 1999

Spatial relationships between L and M afferents (left); and f'al representation of direction in the terminal ganglion (right)

- **A,B,C (left)**: same directional tuning of L and M, 3 different directions
- terminal fields of M afferents smaller and overlay L affs fields
- some segregation between each pair

RIGHT: Direction represented by colour. **A,B,C** formed by L & M afferents terminal arborization. **A**, ***Colour clouds*** represent probability distribution of L afferents tuned to a particular direction. **B**, M afferents; **C**, overlay of A & B.

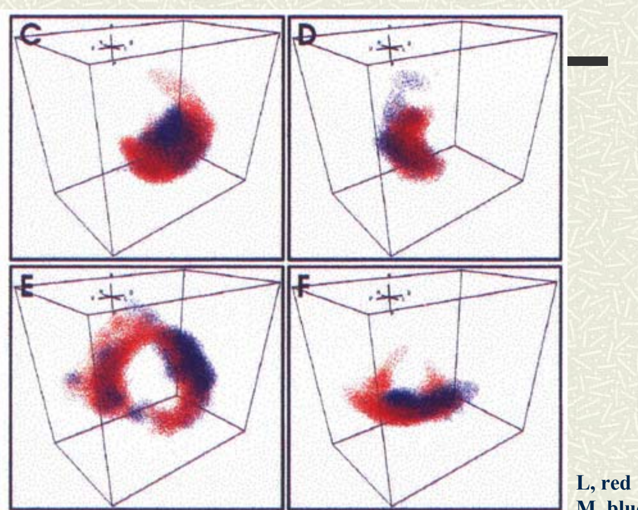




Colour wheel shows direction wrt animals body; Less overlap of M fields (patchy clouds); L&M affs of same direction in register; direction as a whole represented continuously in the ganglion.

Paydar et al. 1999

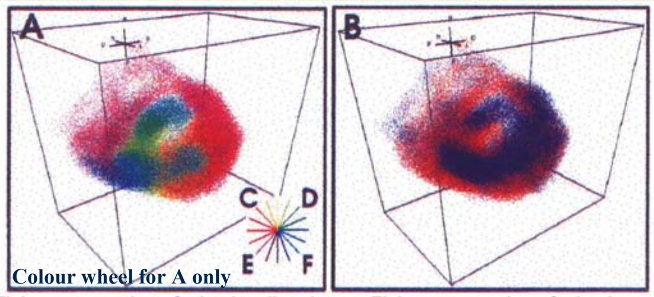
Frequency response mapping of L & M afferents



Arborization patterns of subsets of L and M afferents tuned to different air current directions. In C, 3 L and 3 M afferents, peak directional sensitivity near -45^{o} In D, +45^{o} . E,F: 3 L & 3M afferents -135^{o} and +135^{o}.

34

Functional Representations: Stimulus direction vs. stimulus frequency



F'al representation of stimulus direction by L and M afferents, colour coded according to peak directional tuning wrt body coordinates.

F'al representation of stimulus frequency; L (red), M (blue) arborizations

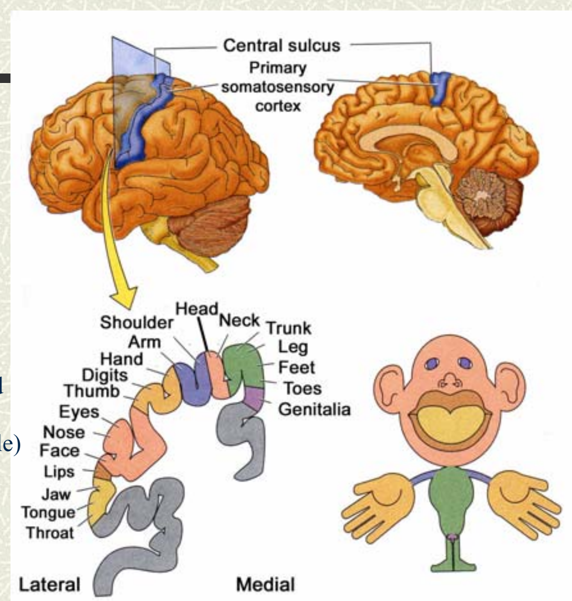
Paydar et al. (1999)

Can the degree of spatial representation be indicative of a "sensory reliance" of the organism (on a particular modality)? Does the amount of area in the neural map occupied by particular sensory field correlate with sensitivity of the area mapped?

The Cricket and the Sensory homunculus - somatotopic organization in the human primary somatic sensory cortex

• More complex than represented here
• Plasticity in the map (modifiable)

(also see KSJ fig.18-4...-8)



Purves et al.1997 (Fig8-8)

Continuing saga of the "well modulated" cockroach

Recall,

Octopamine (OA), dopamine (DA), and serotonin (5-HT) as putative neuromodulators

- effects on thoracic interneurons that drive motor neurons

Role of FMRFamide-like peptides

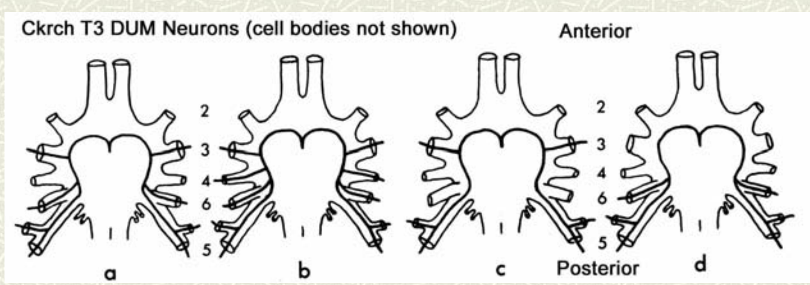
- peripheral innervation of skeletal muscles
- central release sites
- release into haemolymph (blood)

Identifying specific neurons involved in modulation of activity in T3 and in skeletal muscle

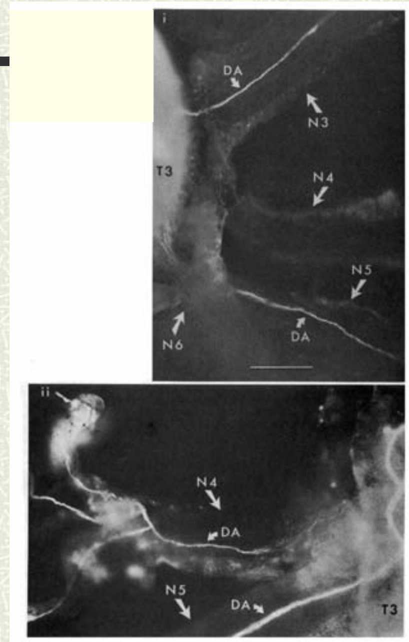
- **(**Dorsal Unpaired Median (DUM) Neurons (peripheral and central (?) connections)

Patterns of innervation by DUM neurons in T3

- DUM3,5,6; DUM3,4,5,6; DUM3,5; DUM5,6
- Where do they go and what do they do?
- Central vs. peripheral roles



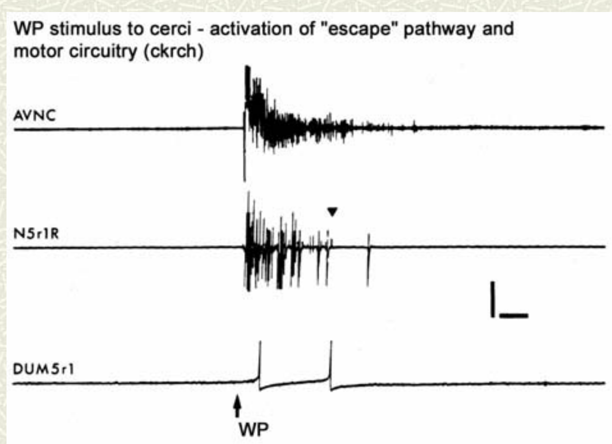
Lucifer Yellow fill of two T3 DUM neurons



39

Recruitment of DUM Neuron during WP-evoked escape response

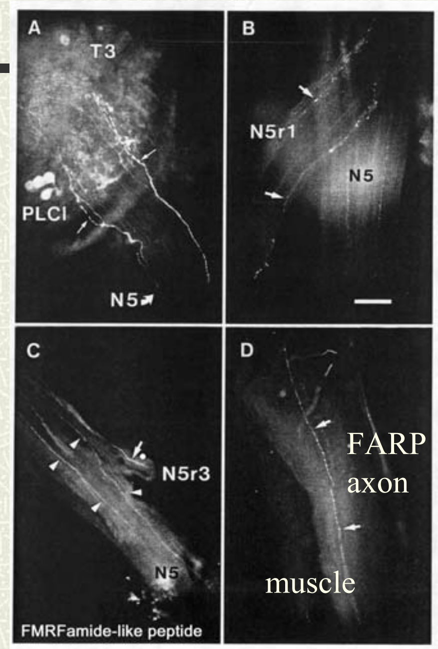
- WP, wind puff directed at the cerci from behind
- animal secure but able to move legs



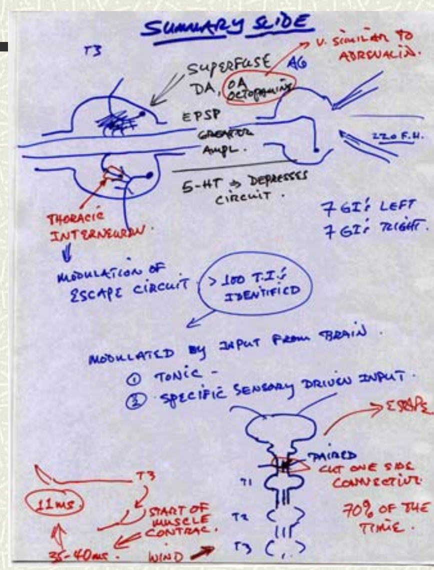
40

FMRFamide-like peptides in CNS and PNS

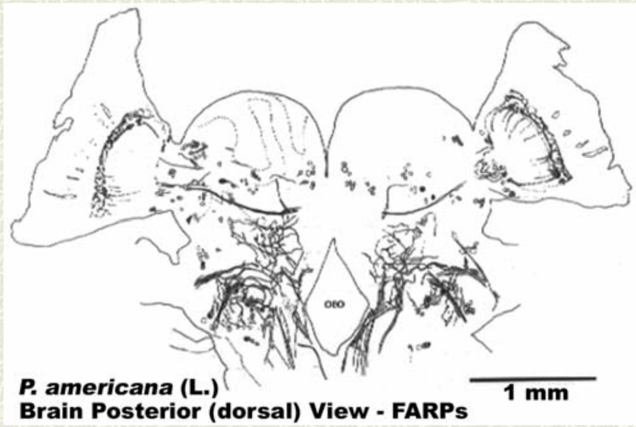
- Roach T3 (again) and metathoracic leg muscles
- immunohistochemistry
- fluorescence microscopy
- central and peripheral components



Summary of Neuromodulation and inputs in T3 (not complete)



42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1 mm
P. americana (L.)
Brain Posterior (dorsal) View - FARPs