1/

Nottingham. 2 March 73.

Our Approach to Developmental Biology

"What's going on in your lab?"

(Sorry, no slides)

Sydney Brenner: the change from molecular biology of ~~[illegible]~~ prokaryotes to development biology.

Plan of Talk: A General Remarks B Examples of our work in this area.

our interest in the nervous system:

how does it work, esp. in man? very difficult problem.

how is it built – one nearer our interests.

general problem of how cells ~~[illegible]~~ divide, move, interact, form tissues, organs and whole organisms.

how to change fields ~ general remarks.

danger of carryover inappropriate ideas.

Sir Cyril Hinshelwood, from physical chemistry to bacteriology.

~~[illegible]~~ excessive reliance on equations
not enough biochemistry, genetics. } → "by one bold mathematical assumption he managed to ignore the fundamental problem"

but not a stupid man

physicists to mol. biology – throw away most previous knowledge.

However this is likely to be less ~~[illegible]~~ useful for the move from mol-biol. → ~~[illegible]~~ cell biol.

What are the lessons of mol. biol.?

① firm theoretical substratum { physics, chemistry, ~~ph~~ physical chemistry

for cell biol, mol. biol. is the substratum.
only partly available for ~~a~~ higher organisms.

e.g. { v. genetic code – ok.
& chromosome structure – not yet
(nature of a gene

(at its own level)

② lucky to have a broad theoretical framework, fairly early
DNA ⇝ RNA ⇝ protein etc.

③ choice of simple systems eg. microorganisms
(in spite of obvious disadvantages)
concentration: eg. T even phages

④ use of modern (often simple) physico-chemical techniques
eg chromatography, ultracentrifuge, radioactive tracers,
electron microscopes, etc.

⑤ the use of genetics as a tool (see later)

⑥ the use of cell-free systems: partly for "purity" (though often not very pure)
partly for ease of addition.

⑦ use of antibiotics as research tools.

General approach S.B. & F.H.C.C.

- reformulate problem and, especially, divide into sub-problems.
  (prob. no easy & unique way to do this)
  for example: can be done in terms of function
  or of structures. (eg. components).
  omit enumeration: examples later.

- probably wise to avoid "classical" problems
  and perhaps "classical" organisms.
  (often chosen long ago, and for reasons which may no longer apply)

How to choose an organism

no general rule: typical questions: how big?
how easy to put electrodes or pipettes into it?
how short a life-cycle?
can it all be done in the lab.?
can one grow (or get) a lot of it?
radioactive labelling?
is biochemistry easy (acellular slime moulds)
how "simple" is it ~~[illegible]~~ (lower eukaryotes)
is its genetics easy?
worked out? haploid? diploid?

how useful is genetics ? not easy to guess.

may show (or hint at) the overall logic of the problem
eg. Mendel
one gene - one enzyme.

eventually likely to be a powerful tool when.
① the right general conceptual scheme.
② certain technical difficulties overcome
ⓐ "diploid"
ⓑ too many genes.

Overall requirements of the problem

Scientists are human - like to enjoy their work.
typically they would prefer - significance
- no very long investment
- techniques available or easily developed.
- interesting results "certain"
- no one else working ("a few friends")

impossible ! or if it exists, won't stay that way.

~~[illegible]~~ Examples of work in our laboratory

our interest in the nervous system..

sub-problem: how do cells know where they are in a tissue?

(colloquium this morning)

(1) insect ~~a~~ epidermis: ~~(1) cells know~~ 2D system. Peter Lawrence

move bits of tissue around & see change in pattern.

conclude: (1) cells "know" where they've come from

(2) " "forget" this ~~a~~ when they divide

(3) behave as if there was a gradient of some sort: nature of gradient not yet known

— also, bristles point down the gradient.

no biochemistry yet.

(2) "Spacing patterns" eg. bristles on the insect cuticle.

1.D work on ~~[illegible]~~ anabaena, one of the blue-green algae

Michael Wilcox & Graeme Mitchison. cylindrica catenula.

chains of cells: heterocysts — fix N_2 — never divide.

vegetative cells — always divide.

can be observed continuously & photographed at intervals.

∴ can construct long pedigrees.

~~[illegible]~~ can do "cut" chop but not "splice" experiments

three results (1) influence of neighboring heterocysts. new septum nearest older one.

why only one (2) ~~[illegible]~~ unequal division. — only smaller from hets.

(3) competition between proheta.

chemistry just starting.

nervous system : "new" experimental animal.

Sidney Brenner +

small ~~s~~ soil nematode : caenorhabditus elegans. — nerves, muscle, epithelial cells etc.

~~[illegible]~~ small egg, several moults, adults ~ 1 mm.

3½ day life cycle burst size 100-200 (∴ ½ day doubling time).

live on E.coli dish. liquid culture. → diploid (n=6) self-fertilising hermaphrodites — complications.

~~[illegible]~~ occasional males.

making mutants : point mutagen.

1 week later, ~~[illegible]~~ for recessive behavioral mutants,

~~[illegible]~~ show behaviour.

~~[illegible]~~ morphological mutants

mapping (6 linkage groups). complementation groups

So far - over 1000 separately arising mutants.

- over 100 complementation groups ~~[illegible]~~ (= genes)

Thus genetics: very favourable - will eventually prob. be better than drosophila.

paper in preparation.

biochemistry easy to grow say a kilogram

... label. (autoradiograph)

not E.coli

studies just beginning : muscle proteins being studied.

eg. DNA kinetic complexity ~ ×20 E.coli.

7

Anatomy & esp. neuro-anatomy

~1000 cells ? 300 nerve cells ~ 96 body muscle cells

no. of cells invariant, also axon branches & connections etc.

has to be done by thin e/m sections.

very hard work. only part of animal ~~too~~ studied intensively so far.

∴ need for a computer. or 35mm film

assistant analyses data ∴ feeds in (eventually feed out)

expect to have ~~an~~ complete nervous system of adult wild type done with about 2 years.

mutants: some c mers.

some large changes — no dorsal, but two ventral

= to nerve cords

some with no changes in that region.

some with small changes esp. branching pattern of c few cells.

diff. indiv. always same cells, but abnormal branching may be different in diff. individuals.

only a beginning.

no cell division after both, except for genitalia.

embryology : need for study : special nature. cell-lineage up to 60 cell stage with light microscope.

e/m : difficulties ("skills") : fixing ; now ok.

time ~~to~~ to finish: within c year or so. (then) study of mutants.

8/

Neurophysiology : electrodes look impossible.
(v difficult even for Ascaris)
but can prob. stain for neurotransmitters.

Chemotaxis : ~~to~~ Sam Ward . cyclic AMP head nutrients "spiral."
Thus ~~has~~ has advantages + disadvantages - likely to become a
standard animal.

~~Chemo~~

Other lines of work.

① Fibrillar elements Denis Bray (+ Dick Fine)

cytoplasmic ~~[illegible]~~ actin in embryonic nerve cells. (of chick) 20%
also in many other cells other than muscle.
(work of Wessels)
probably also ~~is~~ a myosin (slime moulds)
sliding? cytoplasmic flow.
extension of cell processes?
ruffling membrane? nerve growth cone.

② other fibrillar elements : tubulin - microtubules.
~~to~~ neurofilaments.

which slide over which?
how do they get to correct position in cell?

9

membranes

recent advances (review by John Singer)

lipid bilayer : not denaturated protein outside

but globular ones stuck in it + through it.

~~Ab~~ dynamic picture.

Mark Bretscher : shows that ~~the~~ two major proteins in a

red cell go right through

: believes lipid distribution asymmetrical.

quarts outside, serine + ethanolamine, inside

(controversial)

making ghosts.

Bray study other proteins in membranes in cells.

Brenner goes to study by protein chem. new surface antigens on surface

of cells ~~[illegible]~~ transformed by chemical carcinogens

— large variety, etc.

~~[illegible]~~

~~Theory~~ ~~Dr~~ (other lines)

x-ray studies of chromatin Aaron Klug + Roger Kornberg.

: biochemistry : Marcus Noll. Sept.

Theory of nervous system : David Marr.

Cerebellum.

~~[illegible] only a small frac~~

General Remarks

can only tackle a small part of the field.
advantage of having several lines of attack.

- one new organism nematodes.
- one "new" problem positional information.
- ~~th~~ well populated problems eg. fibrillar elements
  cell membranes.
  chromosome structure.
- one "difficult" technique : John Gurdon : injection into counter.
- theory : David Mann (René Thom)

We find it interesting & ~~[illegible]~~ but a long way to go

Classical mol. biol. — tRNA [illegible] Brian Clark
— [illegible] John Smith
— RNA polymer. Brian Clark.

John Gurdon + his group:

Nematode

Positional Information. — spermatogenesis.

Fibrillar elements; cell movement.

Cell Membranes:

Chromosome structure: — X-rays
— colinearity problem.

Theory: David Marr: see theory. — elementary computation