NRP Feb 9, 1998

A. Questions

Molecular Rules ~~for~~ ~~[illegible]~~ ~~Intercellular~~ Communication

1) Kinds of molecular signals?

2) " " " receivers?

3) " " signal-receiver pairs?

4) " " ~~translation~~?

5) Mechanisms for modifying translation?

6) Rules for cell assembly?

~~7) Kinds of~~

7) Kinds of cells?

B. Expt Approaches

1) In Vivo • ~~[illegible]~~ Genetics as a tool to dissect function.

1) Mutation
2) Clones - Nematodes - Free living
3) ~~[illegible]~~
4) ~~Fixed~~ cell # ~800 / [illegible]

~~2) In Vitro~~

5) Constant cell position ~170 Neurons in CNS
6) Generation time = ~100 hrs
7) Techniques
   1) Reducing
   2) Behavioral
      1) Normal Response
      2) No
      3) Negative

2) In Vitro

1) Obtained "pure" populations of each kind of cell. (&/or synaptomes
2) Dissociate cells & fractionate cell A population
   B
   C
3) Others

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~~Questions~~

1) Kinds of signals

2) Kinds of receptors

3) Kinds of signal-receptor combinations

4) Kinds of cells

5) Kinds of signal translation by cell

6) Mech. for modifying "

7) Rules for cell-cell assembly

Approaches

1) In Vitro.

1) Methods for separating populations of cells A B C

a) Separating kinds of receptors

2) Others

2) In vivo

Genetics or tod - correlate function & molecule.

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Prelim NRP Sch

Systems

1) Adler – E. coli motility
2) Delbrück – Phycomycetes – Light
2) Brenner (Myself) Nematodes

3) Benzer – Drosophila

4) Myself – Tissue Culture
1) Cell segment
2) Tumors of nervous system
Oncogenic Viruses
3) Biochemical Markers

5) Soto – Glisttumor

6) Lippman – Protein Syn.
7) Magasanian – Effect analogs of morphine on E. coli
Hela cells.

8) Receptor Proteins – Changeum.
1) Affinity labeling

Seminar NRP
2/4/69 Simple
Strategies

1. Strategy

2. Objective
General
1. Rule at molecular level.
1. Molecular logic
2. *Possible evolution of [illegible] specific
1. Construction
a) [illegible] program
2. Operation

One [illegible] specific problem in order [illegible] general phenomenon
Predicting a specific [illegible] the brain is encoded in the genes
What are the rules for expressing genetic info. [illegible]
Unique features of the Program

3. Information for construction
1. Template
a. Selective device
b. Biological doct.
2. Amt of info.
a. Ecoli < 3000 kinds of proteins
b) Drosophila < 75,000 " "
c) Chicken < 750,000
d) Human < 2,000,000
3. Duplication (~ 10^5-10^6 kinds of proteins)
4. What genes are expressed in [illegible] of expression
5. Number of [illegible] proteins in 1 Ecoli cell
1 mammalian cell
6. Mammal 10^8 - 10^9 cells/gm tissue
Not enough info to specify each cell

4
5. If one is interested the problem of diff. study the development of a [illegible] in Ecoli.

6. Basic problem – Discover the rules for processing info by complex communication of molecules

1. How simple? How low on evolutionary scale?
2. Probability of Universal rules?
   Universal logic, neural code
3. Two guidelines
   1. Biochemistry – General Pathway
   2. Genetic code
   1. Pressing code evolved. Prob. frozen
   2. Predict Langley universal logic
      " " mechanism
      " " specific code
      a) Kauffman logical schemes.
      b) Palmer phot universal
      c) Acquisition of new mechanisms (Memory)
   3. Age of neural code. Code vs genetic code.

6) Need simple system.

1. Degree of Evolutionary complexity?

1) Evolution of Logic

2) Molecular mechanisms

3) Temporal DNA (Prewired pattern)

a) Wigglesworth

2. Selective

gene amplification 1) Select a set of genes that are expressed. Enzyme

1) genes 2) cells

repression - derepression

2) Select cell population. Immunology

synapse amplification 3) Select a set of synapses.

7) Strategy - function of tools. What is doable.

1. Structure & function

What molecules are required (directly) for info processing

2. What is the function of each molecule. The steps in info

8

1) In Vivo
2) In Vitro

1) Mutants - Genetics
2) Embryology
3) Tissue Culture
4) Biochemistry

8) Experimental approaches:

Biochemical Genetics
Tissue culture
Embryology

1. Genetic mutations to dissect physiology.
a) Look for differences. Create biology.
1. Can form, ~~fast generation~~
2. Ease of genetic manipulation
2. Generation time

b) Nematodes

c) Drosophila

2. Embryology.
1. Useful. Developmental abnormality
2. Restrictions -

3. ~~Tissue~~ Cell Culture
1. Cell types - Differentiation. Nutrition
2. Tumor cell built differentiated functions.
3. Our work - ~~[illegible]~~
fdo

4. ~~Biochemistry~~
Cell free systems
Identify ~~[illegible]~~ Unique molecular components.

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1) ~~Tell~~ Molecular ~~[illegible]~~ Biology ~~[illegible]~~ DNA-RNA-Ribosomes Molecular Communication

1) Question ~~[illegible]~~ Processing ~~[illegible]~~ Molecular Communication

1) ~~Molecular~~ Behavioral Rules for chemical nervous system. What is the Program

a) Construction ~~[illegible]~~

b) Operation

2) Genetics ~~[illegible]~~ Single Rules - Universal! Few mechanisms

2) a) ~~[illegible]~~ Kinds of units

b) Number of units of each kind

c) Relation of unit to another

Units

1) Cells

2) Synapses

A. 1. Kinds

2) Numbers of each kind

3. Unit-Unit Relations

3) Genetic Program for constructing machine.

~~1) [illegible]~~

~~[illegible]~~ ~~[illegible]~~ Wiring Prewired

4) ~~Chemotaxis~~ phenomenon. Enzyme ~~[illegible]~~

1) Memory.

2) Dependance.

3) Tolerance.

5)

Strategies Phoenix

2. Genetics [illegible] for discovery complex events ~~[illegible]~~.

1. Mutants. Conditional lethal ts 240K

30 mutant.

2.

Strategy (ie)

1. Systems

1. Requirements

1) M[illegible] complex (ie) Determine effect of varying only

1 [illegible] at a time.

a) E. coli vs mammals.

b) [illegible]

c) Evolution of rules.

memory

2. Tools of Molecular Biology - Quantitative vs Descriptive

1) Genetics [illegible]

1) Ease of growing

2) Generation time

3) Ease of genetic analysis

4) Create biology.

1) Biochemistry

2. Homogeneity

1. [illegible] tissue

3) Cell Markers

2) Embryonic Development.

a) Like mutants. Age degenerant

b) Amt of time

c) Homogeneity

d) Generation time

e) Degree of synchronizing.

4) Tissue culture

1) In Vitro synapse formation

I'd like to describe the problems which we are studying currently. Virtually all the questions relate to the mechanisms of genetic transcription & translation particularly the genetic code & the regulation of protein synthesis.

NRP Tabb 2/8/67

Problems in lab — ~~5 fellows~~

1) Regulation — Differentiation

2) Differentiation of Nervous system.

~~a) Single system —~~

~~1) Nematodes~~

~~2) Artemia (?)~~

~~3) Drosophila~~

3) Problems in lab — ~~5-6~~ 7 fellows

~~1) Code — Deciphering~~ largely completed.

~~1) Universality~~

1) Largely universal

~~2) Synonyms.~~

~~3) Synonyms.~~

2) SRMA ~~fractionation~~ — Most diff between metazoa & Ecl. ~~transfer RNA~~ codon ~~response~~ between amphibian & transfer RNA

1) Quick pigs SRMA vs Ecl SRMA

1) B. megatherium spore-veg.

2) Xenopus neural — adult

Xenopus adult Muscle vs liver

3) SRMA / AASRMA ~~Ratio~~ 3) ~~SRMA / AASRMA~~ 3) Chick embryos vs adult

~~Ecl SRMA — T2 Phage Sucker~~

4) Sucker T2 — Ecl SRMA 4)

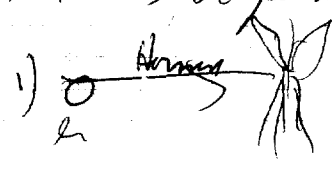
1) Turns off host prot syn.

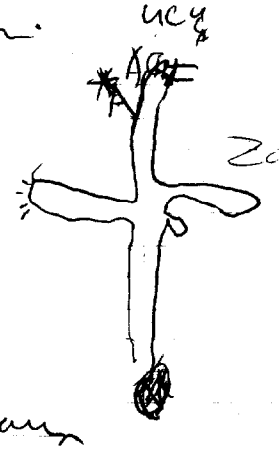
5) ← 5) Ser SRMA isoperiodyl A

1) Horma Skoog

UCY

Zachau

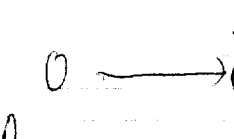




SRMA oxidized iodide

I_2 KI Carbon

3) Coupled Repression →



O → Brown

RNA phosphate ~~Zinder~~ Tumor — Sinsheimer — Spiegelman

6) Behavioral + Developmental nematode mutants

4) Sum up.

1) ~~Different~~ ~~of~~ ~~neurons~~ ~~sys~~ ?!
2) ~~Estimate~~ ~~[illegible]~~ ~~[illegible]~~ ~~[illegible]~~ ~~[illegible]~~ - competent

1) Single systems
*1) Nematodes
(2) Artemia
*3) Insect
*4) Tissue Culture

*2) ~~[illegible]~~ - Mutants

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NRP Associates Talk. (Final)
Strategies of Simple Systems
2/5/69

①

1. Strategy

2. Objectives: Focus on the basis of exact data, the molecular rules, the logic which [illegible] for information processing; then prove [illegible] For operation of the brain

a) Rules, logic at molecular level

b) Sequential questions

1. Yes
2. No.
Maybe

2. Rules for construction of brain.

~20 years [illegible] adequate

1. Genetic Program.

a) Differentiation. Eclipse

b) Are there any new genetic principles

Evolution of new genetic principles

Constructed for Brain.

Limitations: [illegible] the rules

Restrictions: [illegible]

1. Nature of questions
2. Order
3. Length of time of [illegible]

2. Amount of genetic information [illegible]

a) 1) Eclipse 3000

2) Drosophila 75,000

3. Chick 750,000

4. Man 2,000,000

b) Duplication

c) ~ 500,000,000 cells / generation

d) Simple rules seem to predominate

3. Is it likely that the logic will be numerical?

1. Genetic code.
2. Cellular Metabolism — Computing breakdown
3. Predict largely numerical logic
Genetic Code

4. Age of genetic code vs neural code.
   1. 1 event vs population of events
5. Predict great variability in surviving diagrammatic (sequence of neurons)
   a) Basic units combined in endless ways to [illegible] machines evolve.
   b) additional kinds of units may evolve. Addition
6. Problem is to ~~[illegible]~~ an experimental one. What is the Devise simple approaches which will ~~[illegible]~~ (for more) ~~[illegible]~~ them ~~yield~~ which will enable one to deduce the molecular [illegible] & then to prove that the deductions are correct in the simplest, most direct way. ~~with the minimum amount of work~~

1. Biochemistry ~~[illegible]~~ identify kinds of molecules & the reactions that are directly involved ~~[illegible]~~ cell free systems essential.
   a) Handles
      1. Genetics - powerful tool. Create Biology
         a) Hour-long on evolutionary scale
            1. ~~[illegible]~~
            2. ~~[illegible]~~
            3. [illegible]
            4. Drosophila
            5. Mammal.
      2) Tissue Culture
         1. Cell types - Differentiation Problem
         2. Tumors
         3. Our Work
            Gato
      3) Embryology