DOES THE NEO-DARWINIAN PRINCIPLE OF HOMOLOGY WORK AT THE GENOME LEVEL?

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Abstract

*Brain tissues from wild forest mice **(Clethrionomys)** of two similar species were used for isolation and purification of highly homologous polyribosomal poly(A)$^+$ mRNA sequences by molecular-hybridization with depleted bacterial plasmid DNAs. The isolated highly homologous populations of mRNAs from both organisms were translated **in vitro** using cell-free protein synthesis systems. Resulting polypeptides chains were analyzed by slab-gel electrophoresis to test the extent of homology between proteins encoded by the homologous mRNAs. Results indicate a lack of correlation between mRNA homology and protein homology.*

Introduction

Homology among sequences of so-called "informational biopolymers" (DNA and mRNA) are treated as ideal criteria for an estimation of phylogenetic relationship in most recent biochemical and biophysical studies. These studies have included a large scale of living systems from viruses (Davison and Taylor, 1987) to primates including man (Walker and Gedamu, 1990). But interpretation of these data, including the efficiency of DNA-DNA and DNA-RNA hybridization methodology, involve difficult theoretical and technical problems which lead to many misunderstandings (Britten, 1989; Marks et al., 1989; Hasegawa et al., 1989). Therefore, questions about the scientific significance of DNA-DNA and DNA-RNA homology as background for evolutionary conclusions must be investigated both theoretically and empirically. For these reasons we have investigated correlations between the extent of eukaryotic mRNA homology and the extent of homology between polypeptides chains encoded by these mRNA sequences.

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Procedures

The poly(A)$^+$mRNA was isolated and purified from the brain tissue of wild forest mice, *Clethrionomys frater* and *Clethrionomgy gapperi* (referred to hereafter by upper case numbers 1, 2), using standard procedures of phenol-chloroform deproteinization of polyribosomes followed by chromatography on Poly-(U)Sepharose (Kouznetsov and Richter, 1987). For isolation of homologous mRNA sequences, we used a semi-preparative version of hybridization according to De Groot et al. (1987). The pMC9, PGX12 and PFIPVB12 purified depleted recombinant plasmids were donated by Dr. Solomon Torin, Department of Virology, School of Medicine, UCLA. Hydroxyapatite chromatography was used for the separation of stable hybrids from non-complimentary sequences (Anonymous, 1984). Then homologous mRNA^1 and mRNA^2 sequences were extracted from hybrids with 1.7 M guanidine thiocyanate, and the ethanol-precipitated pellets washed with 2xSSC solution using ultrafiltration through Diaflo YM30 membranes. The mRNA samples were treated with RNase-free DNase. Integrity and purity of homologous mRNAs isolated from *C. frater* and *C. gapperi* were controlled by electrophoresis in 2.6% agarose gel containing 7 M urea, 1 mM EDTA, and 0.035 M tris-borate buffer (pH 8.30). Purified homologous sequences of mRNA^1 and mRNA^2 were translated in rabbit reticulocyte lysate cell-free systems containing [^{35}S]-L-methionine as a protein precursor.

Procedures with the cell-free translation system were carried out according to De Groot et al. (1987). Selective inhibition of protein synthesis initiation reactions were arrested by addition of 85 umol/ml of Pactamycin. The polypeptides chains synthesized *in vitro* were analyzed using slab electrophoresis in linear 7.5-20% polyacrylamide gel containing 1% SDS, 0.1% iodoacetamide, 0.025 M tris (pH 8.0), at 150 v per each 0.2 x 120 x 120 mm gel. After fixation in 7.5% acetic acid, gels were dried under vacuum and then scanned for [^{35}S]-scintillation in a DBM-3 Autoradioanalyser.

Results

As seen in Figure 1, the polypeptides chains which had been programmed by highly-homologous messenger RNAs do not share electrophoretic likeness. The heterogeneities of polypeptides chains synthesized by homologous mRNA^1 and mRNA^2 templates (both complementary to one and the same DNA strands) indicate a very low extent of homology of resulting proteins. Significant *differences* were observed in all six experiments using three different plasmid DNA strands.

At least in the case of biological templates tested, we find no strong correlation between nucleic acid homology in two similar species of wild animals, and homology of proteins encoded by these isolated homologous nucleic acid sequences (mRNA, DNA).

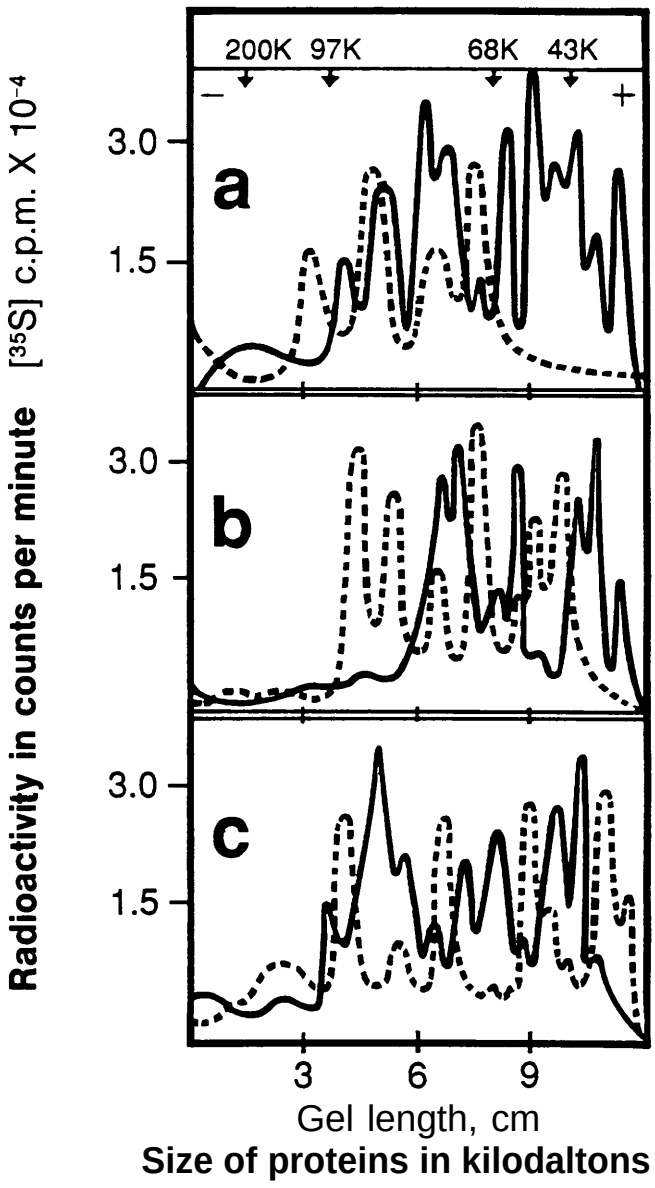

Figure 1. These autoradiographs were obtained from the sulfur-labeled amino acid, methionine, which had been incorporated into polypeptides chains in cell-free translation systems. Highly homologous mRNA templates had been isolated from *Clethrionomys frater* and *C. gapperi* brain cells.

— Polypeptides chains programmed by mRNA *(C. frater).*

------ Polypeptides chains programmed by mRNA *(C. gapperi).*

Polypeptides chains programmed by mRNAs extracted from hybrid with:

a. pGX12 plasmid DNA.
b. pMC9 plasmid DNA.
c. pFIPVB12 plasmid DNA.

Discussion

In summarizing these data we emphasize that proteins constitute the molecular basis for more gross phenotypes, such as tissues and organs. However, our research utilizing three types of highly homologous fractions of purified mRNA from each of two similar species of wild forest mice, shows that in all three cases the proteins translated by the highly homologous pairs of mRNA differ considerably, as determined by electrophoresis. Sequence likenesses between mRNAs from two similar organisms are not reflected in sequence likenesses between the translated proteins. We consider that our data indicate caution in accepting any evolutionary conclusions based on the data of DNA-RNA homology among similar organisms. Similarity is not enough to indicate relationship.

As for causes of observed phenomena, our opinion is that they occur because of the limits of precision in

modern DNA-DNA and DNA-RNA hybridization research. Often results of this research are interpreted to fit a broad evolutionary pattern, but our observations indicate that more restraint and caution should be exercised in evaluating results of these types of studies.

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PHILOSOPHICAL ESSAY

HISTORICAL VARIATION IN THE HUMAN CREATURE

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Abstract

The human body has varied little in basic structure but our historical artifacts have changed dramatically. Intelligence, seeking to conquer "chance" by force, speed, efficiency and control, is largely responsible. Evolutionary science does not care about quality but rather quantity under mathematical control. Rejecting supernatural intelligence by fiat in the very definition of the scientific method, leaves reason without a true foundation for the existence of anything. More important, the final source of all variation is ignored. This source is Gods love of the beauty and complexity of the design he himself created. As we are made in His image, we should likewise enjoy His work of beauty and complexity. Evolution has stolen this from the life of millions.

Introduction

The world is full of millions of things: plants that fix sunlight energy for food, the ceaseless ocean tides, high mountain timber, tiny amoebas, massive rhinoceros and delicate humming birds. Heavenly bodies and earthly creatures, often of startling and strange array, are everywhere. We are not their source, nor did we create ourselves, yet we can use our bodies in incredibly varied ways, generating strikingly diverse cultures. This variety is based in intelligence, not random processes or mutations. Each person and each culture has a unique bounding line. These bounding lines define the field of action and the dwelling place of each human. This boundary line is both physical and spiritual—our dearest friends in another state are spiritually "closer" than a passerby a few feet away.

The Evolutionary Model and Human Variation

According to evolution, all variation in nature and man have chance, natural law, and time as their source. By chance and chaos, all life evolved from an original explosion of dead matter. Evolution maintains that there is no unique bounding line for each creature and culture. Transitional forms must abound. Our intelligence, grounded in chance, cannot rise above its chance "cause." Thus any thought is as "good" as another. Even Darwin's thoughts are true only as by majority vote, not objective knowledge based in absolute truth.

In the evolutionary model, men are a mass of self conscious beings, reaching more and more closely to the goal of perfection as proposed by the leading evolutionary scientists. Evolutionary men hope to develop an autonomous utopia of their own design, subject to no god. Man can do anything. He is on his way to control of the universe, the final frontier. Soon every being on the earth or in the skies will be numbered and graphed and tracked with the exception of God, who has been declared dead and need not be counted!

Force, speed, efficiency, and control are the values of the coming world government utopia, with mathematics as the operational language. Quality, beauty, truth, justice and love are no longer factors. Quantity is the sole guide. Variation among all things is to be crushed. The devices and gadgets do man's work. Man controls all their actions through his intelligence. There is a downgrading of quality for the sake of monotonous, efficient repetitious action as in fast-food chains and educational mills.

Nature is the "raw" material for man's gigantic refabrication schemes to turn her into designs of every human wish. Nature has no independent meaning and is self-generated by chance. Chance provides the many

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