

Genes, neurons and codes: remarks on biological communication

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Summary

I examine critically the application of information-theoretic ideas to biological communication during embryonic development and in the functioning central nervous system (CNS). I show that intercellular communication relies mostly on simple signals whose role is to effect a *selection* among predetermined cellular states. Hence, a crucial role is played by cellular memory, which stabilizes such states. Memory in cells is partly located in the nuclear DNA; no less important however is (phenotypic) memory lying in the cell's organelles and compartments. Because of combinatorial effects in gene expression patterns, cell memory is an enormously powerful mechanism, which also underlies *plasticity*, and thus constitutes the factor unifying genetic determination, plasticity and learning. Communication in the CNS is analyzed in some detail: here, cellular memory is embodied in anatomy (i.e., cell shape) and neurochemistry. These are the major, relatively "static" factors affecting the routing of neural impulses in the adult CNS. In addition, however, faster channeling is also required: action potentials must be directed to their targets along a few of exponentially many paths, and this dynamic routing is crucial for proper operation. I suggest that collective oscillatory modes may play a role in solving this addressing problem, in the same way that the clock signal gates the operation of man-made computers. *BioEssays* 25:699–708, 2003. © 2003 Wiley Periodicals, Inc.

Introduction

Much of contemporary research in biology deals with "information." DNA is life's information store; communication, i.e., the transmission of information, is crucial for the development and maintenance of organisms; it is the essence of a functioning nervous system. Current theoretical discussions in neurobiology or developmental genetics often appeal to information-theoretic concepts such as "bits," either informally or with the use of a mathematical apparatus. Since "information" is "processed" in the brain, there is, for example, much debate as to how the nervous system might "transmit

information" at a "maximal" or "optimal" rate with action potentials or other electrochemical means.⁽¹⁾ Another typical "problem" concerns whether genes have the "information-theoretic capacity" to "code for" development.

I will defend the view that, in biological processes, such as occur in the central nervous system or during development, very little information needs to be transmitted. Most of the required information is already where it should be, namely in the cells where it is used. I shall argue that it is fundamental to distinguish *information* itself from the way it is *encoded* and *decoded* by cells: a small fraction of the information that a cell harbors may sometimes be encoded by that cell into a signal, and that signal will be decoded by other cells, in ways relying critically on information that the receivers already possess by virtue of their genes and ontogenetic history.

I hope that drawing clear distinctions between these different aspects will help clarify some problems whose true impact has been obscured by persistent ambiguities between biologists and theoreticians. After some basic considerations regarding information in biology (see next section), I examine how information is stored and processed during the development of organisms, and in the functioning of the CNS. The approach leads to a number of simple experimental predictions regarding biological signaling systems.

Biological information

Consider embryonic induction: a molecule secreted by part of the embryo induces cells nearby to adopt a new fate. An example is mesoderm induction, which involves a major shift in cell specification. In the sense of information theory, the mere presence of a single molecular species (activin) is equivalent to just one bit of information. But is "transmission of a single bit of information" a fair description of mesoderm induction? Hardly so: most of the important phenomena take place in or on cells, during *secretion*, *propagation* and *transduction* of the activin signal. Classical information theory thus seems to have little to teach us about intercellular communication, because it neglects, as Weaver noted,⁽²⁾ the actual content of the message, which is, in this case, embodied in cell properties and determines when emission should take place and what a proper response should be. Reflecting on this failure leads to a possibly more fruitful framework.

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Information is in the eye of the beholder

In fact, the term “presence of the inducer” is itself a misleading phrase. The inducer should really be characterized by a space- and time-dependent concentration, representing a potentially *infinite* amount of information (depending on how it is generated)—but more on graded concentrations later. The point here is that a cell reads the inducer concentration *according to a given time schedule* and relative to some *cell-defined* threshold. As interpreted by cells then the induction signal is indeed reduced in some sense to reception of a single bit; but the signal is not really *activin*: the latter’s concentration becomes a signal only when interpreted in a certain way by a particular cell type.

“Signals” such as *activin* have no operational content unless emitted in a particular context and until they have been deciphered by a transduction pathway and interpreted in terms of response. Such encoding/decoding procedures are called *communication protocols*. Protocols are ways for emitter and receiver to agree on all aspects of transmission, such as, for example, timing, coding and error correction: thus, double-stranded DNA can be interpreted according to six different reading frames, or be noncoding. What sets the reading frame is a biochemical protocol dictating the initiation and ending of RNA polymerase processive runs. Similarly, cells come equipped to emit some signals and *transduce* some others impinging on them, crucially filtering out the rest.

Selection versus instruction

An emitter of information such as a cell or DNA must be able to assume, at a given moment, one of a set $\{E\}$ of possible, mutually exclusive internal states E_1 to E_n ; thus a neuron may be firing or not, a lymphocyte may express or not some antibody out of a large repertoire. As part of being in the state E_i , the emitter concocts signals S_i . Receiving cells, by virtue of being in their own initial state R_j (one of m such possible states in a set $\{R\}$) may detect the signal(s) and interpret them according to a definite protocol, perhaps switching to a new state R_j ; this is the *decoding* of S_i .

A crucial point is that there need not exist any relationship whatsoever between the richness of the transmitted signals $\{S\}$ and the complexity of either the $\{E\}$ states or the transitions mediated among $\{R\}$ states. Hence, a very “complicated” message can be transmitted with very “simple” means. This is possible because of the information that emitter and receiver share even before transmission begins, and which endows them with adequate $\{E\}$ and/or $\{R\}$ sets. In the case of embryonic cells, this consists of their genome, phylogenetic and ontogenetic history; in the case of nerve cells, their functional connectivity.

A signal may be effective in two different ways. An *instructive* signal would carry, spelled out in “rich” fashion, a complete description of what the receptors should do: think for example of a chemical whose distribution would delineate the

exact shape of a limb. A *selective* signal is a “poor” one which, by itself, does not explicitly convey any detailed description: the richness resides then in the sets $\{E\}$ and $\{R\}$, with properly timed and positioned signals selecting sophisticated responses from the set of possible ones. Think for instance of a signal whose gradient of concentration indicates proximo-distal polarity.

Clearly, signals in development are of the selective type. Robustness is presumably of evolutionary advantage and has favored *simple* signals. Indeed, even in an evolutionarily “privileged” environment such as the embryo, cells do not seem to emit or receive complicated signals: induction is a case in point. Among the most elementary signals in development are “semaphores,” useful for synchronisation and guidance.⁽³⁾ A semaphore’s message is: “I am here, in state x ”. The receiver—say a neuron growth cone passing by—will then know what to do. One of the most important set of signals in development are those subtending “community effects” whereby cells tend to adopt the fate of their neighbors^(4–6): “do what I do.” The converse—“Do not do what I do”—is implemented by lateral inhibition pathways, of which the Notch–Delta couple of receptor and ligand families is the paradigmatic example.⁽⁷⁾ (Note that to repeat or negate your neighbor’s fate is a relative determination; it does not say what your common or differing fates will actually be.)

The significance of biological information

Usefully correlated behavior of emitter and receiver is what makes a signal significant. The key word here is “useful:” in the natural context of a cell at a given moment, there is a behavior that it is *supposed* to adopt. There is, in other words, a developmental or behavioral *plan* that should be followed. Significance of a signal in biology lies in continued conformity with a “plan.” The responding cell should not just do “something,” but the “right” thing.

Significance therefore, has mostly to do with encoding and decoding. Whether the “bits” in a signal, however measured, are significant, depends principally on the context, i.e., on the genetic, anatomical and chemical background. Thus, a signal that is a member of a family of partly redundant cellular and/or intercellular pathways might be suppressed alone without much consequence, and would not appear very significant; yet, if several family members are suppressed, the single, “insignificant” member becomes highly significant. It is the family that is significant: in a way, we must know the answers about significance even before asking the question.

The significance of a biological signal is obviously not a simple concept. How it relates to individual cells is very indirect. We shall face this difficulty at its worst when we later consider plausible information codes of functioning neurons.

Gene Power in Development

Extracellular signals are fragile, so they are better kept simple. To maintain a limited signal variety, while improving adaptation, the only way is to increase the variety of responses generated reliably by the encoder/decoder (or transduction) systems, i.e., to refine ever more sharply cellular *identity* as defined operationally by the repertoire of states $\{E\}$ and $\{R\}$. More than signaling then, it is cellular and subcellular *memory* as embodied in the cells' regulation networks that should hold the key to biological communication.

Let us first consider the memory embodied in chromatin. Just how much of a memory potential is there in the genome and its associated transcription control machinery? Very simple reasoning shows that this is staggering; the demonstration is well worth spelling out—if only to avoid recurrent and muddled discussions about the so-called “necessity” of epigenetic phenomena. (see for example Refs. 8 and 9)

Not enough genes?

Although quite a few biologists know better, many are still impressed, for instance, by seemingly rigorous arguments to the effect that the orderly wiring of the CNS (say) lies beyond the power of the genes; they thus feel compelled to invoke various supplemental “correctives” such as DNA methylation, multiple transcripts from single genes, immunological-style hypervariability and recombination, “learning,” or even some form of cellular or synaptic “Darwinism.” This state of affairs makes it important to understand clearly why the mathematical arguments are only *mathematically* convincing.

We know, says the mathematician, that each neuron in our CNS makes about 10^4 synaptic connections; each connection must choose one of the system's 10^{11} other neurons as its target, making $(10^{11})^{10^4}$ choices just for those connections to which a given cell is presynaptic; since there are 10^{11} cells, specifying the destinations of all connections entails a choice among roughly $[(10^{11})^{10^4}]^{10^{11}}$ possibilities, a number one might want to call “astrological.”

Neuroastrologically, brain development is indeed a “deep” mystery. The epigenetic factors often mentioned as remedies increase the possibilities by mere factors of thousands or billions, i.e., they are utterly negligible in view of the apparent gap. But could one not argue comparably that genes do not have the power to code for the position of each atom in the simplest bacterium? Of course, they are not required to do that: the bulk of the work is done by the universal laws of chemistry. In like manner, genes may “use” the laws of molecular cell biology which more or less universally regulate cell growth, shape, function, and death.

Thus, Sperry⁽¹⁰⁾ proposed concentration *gradients* as way to provide an essentially infinite amount of information from a single chemical substance, i.e., as the solution needed to “multiply the bits”. Consider Sperry's problem of retinotectal mapping. There are about 10^6 ganglion cells in the frog retina,

and the same number of target cells for them in the tectum. Sperry considered that there had to exist a million chemical “locks” on the targets and as many “keys” on the incoming axons. His solution of this perceived quandary was the gradient idea. Thus if there exists a gradient of ligand $L(x)$ on incoming axons, and it is met by an opposite gradient of membrane receptor $R(x)$ on target cells,^(11,12) the simple assumption that axons continue searching for their target until a uniform, preset signaling level $C = R(x)L(x)$ obtains everywhere, suffices to build a smooth mapping.⁽¹³⁾

Sperry's chemoaffinity gradient is an *analogue* (continuous) signal, able to assume a continuous infinity of spatially coordinated values, and which is clearly able to direct the formation of maps. Mappings are obviously an important large and medium-scale component of brain ontogenesis. But is it likely that cortical microcircuitry is built exclusively, down to the minutest level, from simple mappings?.

Plenty of bits: the combinatorial labeling model

In reality, transcriptional specification and orderly, fine-scale regulation of a “million locks and keys” is not a preposterous proposition.^(14,15) Let us start with specification. Assume a family of molecules, each of which may either be effectively synthesized in a given cell, and presented on its outer membrane, or not. How many such molecules would be necessary in order to compose a unique molecular label characterising each individual ganglion or tectum cell? The answer is $M = \log_2 10^6$, i.e., $M \approx 18$ molecules. This is the famous and almost always underestimated “power of combinatorial coding”: if each label molecule is either present or not, so that there are, with M molecules, 2^M possible combined labels, this grows exponentially to billions with increasing M . How Sperry's lock and key mechanism might be fully implemented from there is still far from evident; but the capability is clearly on hand. Labeling all nerve cells uniquely in the brain would not tax the genes much more heavily: on the order of $\log_2 10^{11} \approx 37$ labels would be sufficient.⁽¹⁶⁾ Each synapse could be individualized at the cost of roughly 50 molecular labels used combinatorially.

But can the genetic system control the patterned expression of such labels, so as to provide from them, e.g., positional information?⁽¹⁷⁾ Gierer⁽¹⁸⁾ has sketched hypothetical mechanisms which would allow cells to keep a complete memory of their lineage in precisely the space-time dependent manner required; but the modular structure of eukaryotic cis-regulatory sequences, which is now becoming understood,⁽¹⁹⁾ opens here a further combinatorial treasure trove.

Plenty of bits: combinatorial cis-regulation

Cis-regulatory proteins bind indeed to dedicated response elements within gene promoters, and form modular blocks that interact with other blocks (or “co-regulators”).^(20–22) In principle, interactions among regulatory modules may be

additive and/or multiplicative and are subject to thresholds. Additive effects lead to, for example, presence of any one of several factors being sufficient to reach the threshold for downstream transcription (a logical “OR”);⁽¹⁹⁾ multiplicative effects engender the need for a joint presence of two or more factors (logical “AND”). It is not difficult to see that the complexity of the *cis*-regulatory functionality that can be assembled from a given number of modules grows exponentially (see Ref. 23 for example), thus easily keeping pace with the number of label gene configurations that they should be able to generate and stabilize.

The neuroastrological formula describes a brain where a “developmental daemon” could connect each neuron precisely with any other, arbitrarily chosen neuron; Sperry’s gradient model introduced simple rules with extremely few molecular components, leading to restricted wiring possibilities. In the present chemical labelling model, in contrast, the neurons are endowed with combinatorial properties, acquire individual identity, and may thus participate in the setup of very (but *not* arbitrarily) complex circuits.

Using the excess bits: dynamical systems and canalization

Interactions in *trans* introduce yet another degree of sophistication in the use of information during development. Transcription factors regulate their own transcription in complex fashion. Formal analysis of the resulting networks has been initiated by Thomas⁽²⁴⁾ and by Kauffman.⁽²⁵⁾ Such networks are instances of dynamical systems, which have been extensively studied in classical mechanics (see Box 1).

For biological purposes, the lesson of dynamical systems theory is that all systems converge to a limited number of *attractors*. This can be interpreted as a type of *canalization*,⁽²⁷⁾ or “error correction.” The fact that certain genes can be perturbed (e.g., knocked out) without noticeable phenotypic effect is testimony to the existence of such canalization: genes do not leave the organism underdetermined, they actually overdetermine it. From the point of view of the present labelling model, canalization is bought at the cost of reducing somewhat the possible diversity resulting from combinatorial labelling and combinatorial control of transcription; but note that this cost is actually negligible, since a fraction of an exponential is still an exponential. Many factors contribute to canalization; let me simply mention here that many such factors are somewhat neglected today because they have no simple, direct genetic counterpart. Thus a variety of memory effects, such as inertia of cell shape, delays in transcription, translation and transport can be expected to improve the stability (but not the responsiveness) of organisms. The importance of these factors for plasticity will be pointed out shortly (see “Comparison with Experiment II”).

Genes have been selected for efficiency, and this may actually on occasion convey a misleading impression of

Box 1. Genetics and Dynamical Systems⁽²⁶⁾

Dynamical systems are defined as sets of interacting variables. For instance if several genes code for transcription factors, the genetic activities in this network form a dynamical system. Starting from some initial condition, such a system undergoes a transient regime before settling in a permanent, so-called “attractor”. The fundamental mathematical result is that there are only three possible attractor types:

- a steady state
- an oscillatory regime
- an endless (but organized) chaotic wandering.

Importantly, the number of attractor states is small compared to the number of possible configurations. This is due to the interactions, which introduce constraints: a simple illustration is a single (directly or indirectly) autocatalytically controlled gene such as, say, *engrailed*. Although the activity level of many autocatalytic genes could presumably be tuned in continuous fashion, because of their strong positive feedback such genes often turn out to be either on or off: a potentially infinite continuum of states of activity has been replaced by just two, due to a simple interaction. In addition to such direct effects, many indirect, network effects must of course be expected in development, where, say, one factor regulates a second which in turn regulates the first. When genes involved in cell–cell signaling (coding, for example, for receptors and ligands) are included, the dynamical system comes to encompass the genetic and molecular state of groups of cells rather than that of single cells.

genetic thrift. Thus, it may sometimes be possible to isolate subsets of genes or “modules” that assume identical states in two different parts of the organism, or at different times during development, and whose function is only slightly altered by their differing contexts: such is the case, for example, for the genes building up the legs or antennae of insects;⁽²⁸⁾ modular organization is also apparent along the rostrocaudal axis of insects and vertebrates.⁽²⁹⁾ I hope it is clear, by now, that this economical reuse of genes is the result of selection, not of genetic paucity.

Phenotypic information storage

Plasticity is the ability for a cell to alter its response durably, according to external circumstances. Plasticity depends on cellular memory, which is therefore a fundamental tenet not only of differentiation, but of developmental and behavioral variability as well. Plasticity and genetic determination are

usually presented as two opposite poles on a scale: plasticity is supposedly “modern” and “dynamic”, denoting flexibility and learning; while genetic determination is “old-fashioned” and “stodgy”, being the “primitive” way characteristic of, say, insects. In view of the preceding argument, however, I propose that they are but two faces of the same, omnipresent coin.

In cellular memory, a pivotal role is played by gene transcription, which underlies cell differentiation. Memory at the level of organelles such as synapses is often considered separately, since it does not necessarily or directly affect the nuclear DNA, but rather cellular transport pathways such as the Golgi apparatus and submembranar localisation systems.⁽³⁰⁾ Yet, to maintain this “non-genetic” memory, dedicated genetic and epigenetic mechanisms are crucial: to stabilize memory beyond molecular turnover times—in the form, for example, of neurite shapes or synaptic structures—the required materials must be properly synthesized and translocated. This alone can ensure that some of the adaptations of cellular subsystems, whether plasticity-dependent or not, are conserved over the lifetime of the cell itself, or even over that of the organism.⁽³¹⁾

Cell and tissue maintenance mechanisms thus form a necessary but relatively neglected feedback system from phenotypic plasticity to cellular differentiation.⁽³²⁾ This “Lamarckian” feedback may have arisen partly through genetic assimilation whereby genetic systems subtending, say, faithful synapse maintenance and “reproduction,” were selected due to faster learning and longer-lasting adaptation by their lucky owners.^(27,33)

Why depend on external stimulation?

If one accepts the foregoing, an interesting question arises whenever some behavior appears to be sculpted by external stimuli: why did stimulus-independent “genetic programming” leave a hole there—and not elsewhere? Thus, many birds are, from the time they hatch, sensitive to subtle global features in the visual profile of potential predators.⁽³⁴⁾ This certainly implies complex neural wiring, acquired maybe through genetic assimilation. However, young birds must *learn* to sing,⁽³⁵⁾ yet singing would not appear at first sight to require circuitry any more complex than predator identification.

Some very basic behavioral patterns of mammals, such as care for the offspring, often depend critically on olfactory imprinting: thus the ewe needs to be imprinted by the scent of her lamb in order that her maternal behavior be “released.”⁽³⁶⁾ Why is such a critical task left to the potential accidents and vagaries of experience-dependent neural plasticity? The reason is certainly *not* that the task is impossible in a “genetically programmed” framework.

Comparison with experiment I

“[...] any bird people would tell you that all feathers are different.” -Lewis Wolpert

I have argued above that genetics is rich enough, that each feather and, indeed, even each cell in each feather can be different. There is growing molecular evidence in favor of this view. By tracing gene expression in enhancer-trap *Drosophila* lines, Kaiser’s group has made an amazing discovery: the tissue of the fly’s entire Malpighian renal tubule is differentiated, in terms of gene activity patterns, down to the *single-cell level*.⁽³⁷⁾ This certainly adduces strong support for the cell-labelling model. Less thorough, but extremely suggestive, evidence comes from other experiments: many data have thus been gathered recently about the way cells and even synapses in the CNS might be determined according to simple codes.⁽³⁸⁾

From Kaiser’s group again, using the same enhancer-trap technique, come precise indications that, in the mushroom bodies of the fly, which play a great role in olfactory learning, the so-called Kenyon cells, are much more differentiated genetically than previously suspected.⁽³⁹⁾ Olfaction is a modality where cell variety seems the rule: thus, olfactory receptor expression^(40,41) seems to single out cells in combinatorial fashion, and regulates their complex, highly stereotyped cortical connectivity.⁽⁴²⁾ Cortical neurons too, have been shown to be surprisingly diverse⁽⁴³⁾ and combinatorially marked: the regional cadherin code⁽⁴⁴⁾ is particularly significant in the present context; enough cadherins are known by now to potentially label each synapse. But other differential cortical layer markers such as *Otx*,⁽⁴⁵⁾ *SCIP*,⁽⁴⁶⁾ *Emx* and *Dlx*,⁽⁴⁷⁾ phylogenetically conserved markers of somatosensory barrels,⁽⁴⁸⁾ or regionally graded markers such as ephrins⁽⁴⁹⁾ have been discovered as well. Any attempt to display several of these markers on a single diagram reveals a mind-boggling parcellation. Complementary to this comprehensive cell marking, the extent to which neural development can proceed without any contribution from activity-dependent plasticity has become glaringly evident; thus, in the olfactory system, programmed genetic specificity seems sufficient to drive the establishment of extremely precise neural connectivity (namely convergence of the axons of widespread neurons onto single glomeruli), in the complete absence of electrical activity.⁽⁵⁰⁾ Knockout mice totally lacking neurotransmitter release develop an apparently normal brain all the way to birth, when massive apoptosis finally sets in.⁽⁵¹⁾

Whether all of the encoding power revealed by this diversity and resilience is actually put to use in terms of phenotype is however a question for experiment to resolve.^(43,52)

Comparison with experiment II: further suggestions and predictions

The proposed model asserts that, more than signaling itself, it is the coordinated course of events among signal emitters and receptors which is important. A prediction immediately follows: provided that a signaling pathway is not excessively pleiotropic (i.e., it does not affect too many responses simultaneously)

ligand and receptor molecules may be able to evolve coherently, somewhat faster than the molecules involved in the upstream and downstream transduction cascades; they might therefore be expected to exhibit rather high polymorphism. In this respect, the experiments recently initiated on the *Ras* pathway of *Drosophila*, measuring the selection pressure to which its various components are subjected, may prove enlightening.⁽⁵³⁾

I have suggested that phenotypic plasticity is a facet of cell memory, and emphasized the processes that occur at their interface. These processes deserve much closer theoretical and experimental scrutiny than they have hitherto received,⁽⁵⁴⁾ in the context of development, of evolution, and most obviously, in studies of the evolution of development itself (evo/devo).

The cell labeling model exhibits the close relationship between hierarchical genetic organization and mastery of the combinatorial “explosion” in development; the example of *Drosophila* renal tubules shows however that this hierarchical principle, though possibly at work in many situations, might not always be apparent unless expressly sought after, as the recent discovery of fibroblast diversity indicates.⁽⁵⁵⁾ Thus, a remarkable example of very fine-grained “positional memory” is afforded by limb regeneration in the amphibians: expression of the *Prod1* surface molecule is graded along the newt's proximodistal axis, and *Prod1* levels are clearly implicated in the control of cellular positional properties.⁽⁵⁶⁾ It is not easy to propose plausible biochemical mechanisms for the robustness exhibited by a smooth distribution such as that of *Prod1*. The present model would predict that a more “all-or-none” or “digital” genetic mechanism possibly underlies such a distribution.

The Central Nervous System: neural codes

In the CNS, cellular memory is usually seen as residing essentially in synapses. Yet, the shape of nerve cells, and the distribution of cytoplasmic and membrane molecules, bear a relatively permanent trace of their development and plasticity. This anatomical factor, underlying as it does the local and global wiring of the system, may form an even more crucial part of memory than do synaptic strengths.

Finding out how and what neurons communicate to each other is usually referred to as the “neural code” problem.^(57–59) Neurons emit, receive and integrate numerous signals;⁽⁶⁰⁾ the $\{E\}$ and $\{R\}$ sets of emitter and receptor states of neurons are complicated neurophysiological states, and much is known about the laws linking e.g., action potential reception and generation at the cellular level,⁽⁶⁰⁾ leading to the detection and amplification of input correlations. But on the scale of the whole CNS, complex signaling is compounded with complex wiring: how then does a central neuron sort out, among the thousands of signals it receives, the ones it should attend to at a given moment?

The signal routing problem: a look at computers

Why was this question of signal sorting not mentioned in the context of development? Because there, the situation is fairly clear: molecular signals are presented or secreted by embryonic territories, bind to their receptors in the same or in nearby territories, and are transduced. Territories thus communicate by virtue of relatively *static* or slow-varying neighborhood relationships and/or fixed chemical affinities between ligand and receptor families. In a multifunctional brain, multiple, intersecting pathways must remain potentially open at any time, while only some are effectively operating. Routing must therefore be *dynamic*.

Just like the CNS, man-made computers face the dynamic protocol and signal routing problem; protocols and codes devised for computers may serve as a surprisingly good starting point for reflection. Since they “understand” only bits, digital computers use ASCII or other codes to group bits by 7 or 8 into characters of text. But in a continuous flow of bits, which are to be grouped to form characters is a matter of communication protocol, of which *synchronization* is a key aspect. Even the fundamental binary system calls for a protocol, for in digital machines, voltages may range from, typically, 0 to 3 volts, and as processing elements (gates) switch from minimal to maximal voltage (i.e., from “0” to “1”), they will unavoidably cross through intermediate, “meaningless” values. A *clock* is indispensable so that each gate is instructed when to read the appropriate inputs, and when again to display its computed output to the proper wires. The signal of this central clock has to be distributed all over the system (this is a major design constraint); its frequency is what is meant when referring to a “300 MHz chip.”

Observing the currents in such a chip, one would notice immediately strong oscillatory, correlated currents at 300 MHz. This massive synchronized activity, however, does not reflect much about actual computation, other than the prosaic need for processing to occur in a globally coherent fashion. The information's important features reside in the system wiring and in the functionality of the individual gates being connected. This is where the encoding resides, and not in the oscillation, which merely serves to lock select gates in phase.

Canalization of neural signals: routing in the brain

In order for widespread, remote groups of neurons to compute together, they must solve the same problems as computer elements: they must fall in step somehow, and information must be distributed correctly. As to synchronization, the time scales of interest (synaptic transmission, postsynaptic integration, action potential generation) all overlap, and timing control must be sharp for correct performance of critical tasks;⁽⁶¹⁾ myelination of long axons, which speeds transmission, is testimonial to the importance of the question. As to

signal routing, the number of paths to be eliminated grows exponentially with their length.

However, a relatively simple system for synchronization and routing on large or even global scales in the CNS does in fact exist: it consists in taking advantage of the collective, space-time dependent dynamic modes of activity that occur inevitably in any complex nonlinear network. These states are the attractors of the neural network, in complete formal analogy to the attractor states of genetic networks (see Box 1 and section "Using the excess bits..."). Neural attractors corresponding to spontaneous oscillatory regimes, and involving topographically distinct (though possibly overlapping) groups of neurons are indeed, on mathematical grounds, bound to arise in the brain. These medium to large scale dynamic structures are easy to stabilize selectively by Hebb-type mechanisms.⁽⁶²⁾ Oscillatory regimes are of course readily observed in the CNS, e.g., in epileptic foci, and significant variations of basic oscillatory regime are seen during various phases of the circadian cycle;⁽⁶³⁾ oscillatory attractors constitute, I suggest, a ready-made (but, without reinforcement, rather imprecise) tool for dynamic functional wiring.

Stable, topographically differentiated oscillatory attractors, then, might constitute one at least of the synchronization and signal routing protocols, over which the actual signal circulates. A computer model which uses this possibility has been proposed to account for pheromone detection in Insects.⁽⁶⁴⁾ A quite detailed formal model of short term memory has also been described,⁽⁶⁵⁾ which uses the phase of the theta wave in the mammalian brain for time multiplexing (i.e., "time-slicing" as done, for example, in transmitting multiple conversations on a single cellular telephone channel).

Whatever these models' fate, experimental evidence shows that oscillations and/or synchronous activity are pervasive in the CNS. Their role in perception or even consciousness^(66,67) should however be placed in proper perspective for, if my present claim is correct, oscillations have little to do with the actual neural code: synchrony and accurate timing are simply a "carrier wave" used for transmission in even the most primitive nervous systems. Synchronization as such certainly cannot be considered as mediating a code, not even in the mild sense of binding together separate perceptual features to form a perceived object.⁽⁶⁸⁾ For what is synchronized need indeed not be meaningfully bound, as epilepsy or sleep EEG amply demonstrate. Synchrony *may* sometimes be necessary for what in psychology is known as "Gestalt" (or holistic) perception, but it is certainly not sufficient.^(63,69)

Neural "meaning:" encoding and decoding

To appreciate the *significance* of what a neuron does in terms of information processing, we should know something of the "plan" of the brain's operations, as explained in the section on "The significance of biological information". Since Hubel and Wiesel's work, we know that extracting *features* from sensory

data is part of the plan; and if we notice that a particular neuron seems to extract a particular feature (judging this from how much its behavior depends on the presence of the feature in a stimulus), we can assess this neuron's efficiency at this task.^(70,71) Beyond feature extraction, however, little has transpired experimentally as to the more global representations (if any) the brain may build from the extracted features, in order to generate adaptive behavior.

What is a particular neuron in an integrative area "coding for"? Unfortunately, it does not really do to use the *ex post facto* discovery that neurons in some area appear to code for "something" in the given experimental context (mirror neurons of the premotor cortex⁽⁷²⁾ or delayed-response neurons in the dorsolateral prefrontal cortex⁽⁷³⁾ are examples). Such knowledge is of course highly valuable, but that some cells seem to detect certain features of an experimental procedure says little in itself as to why or how they accomplish that feat, or as to what other feats, related or not to that particular one, they might additionally perform. And the role of the vast majority of the area's cells, many of which must contribute somehow to the system's performance, remains completely unclear: these cells' activity is usually treated as nothing more than background noise.

Even in the "simple" circuits of invertebrates, the precise "role" of neurons in behavior is complicated, corresponding to no clear "information pathways": thus in the lobster's stomatogastric ganglion, who is to say what each neuron codes for?⁽⁷⁴⁾ These neurons do not seem to function in terms of task-related informational features which could be easily discerned.⁽⁷⁵⁾ Here, the question for information theory would not be about numbers of bits, but more simply: are there bits at all? Consider also stroke recovery in humans: information purported to be flowing along damaged pathways is able to reemerge along different ones, showing that it must have been potentially distributable there even before the lesion, but was somehow left unexploited.

Neural "simplicity"

It is worth considering briefly a notable exception to this state of affairs, that of the Mauthner cells of teleost fishes.⁽⁷⁶⁾ These two identified central neurons (one on each side of the animal) mediate, all by themselves, the escape reflex in these fish. Anatomically and functionally, the input and output pathways of these cells are well charted. This would seem to be a situation where reasoning in terms of information processing can be fruitful, because Mauthner cells confer "meaning" to impinging signals and to their own output by virtue of their functional connectivity properties: they *decode* into an exquisitely modulated adaptive response (stay put, or escape from the right side, or escape from the left) information (presence and motion of various threatening stimuli) *encoded* by the senses.

The lesson from the Mauthner cells is that encoded information must indeed be decoded: there has to be a consistent,

context-dependent mapping between input and output. In fact much of the message resides *in* the map itself. The necessity of encoding sensory information is usually recognized, but the corresponding need for decoding is taken rather lightly: yet, in order to elaborate adapted behavior from stimulus or from stored memory, decoding (e.g., in motor language) is clearly indispensable.

Upon reflection, it can but become clear that decoding entails the existence, maybe not exactly of homunculi, but of their conceptual equivalents, which alone, as far as one can see, can decode body- and mind related decisions from the encoded message delivered by the senses and by centers for action planning. Functional convergences, appropriately sorted, must take place, even if not necessarily in what are usually recognized as “areas of convergence.” The Mauthner cells are in effect functional *homunculi*, “control rooms” watching all relevant sensory modalities for potential danger and interpreting the overall result in terms of a simple output connectivity and adaptive behavior. While *homunculi* are usually disparaged nowadays as utterly ridiculous, I believe something like homuncular-style neural systems, able to make sense in bodily and mental terms of neurally encoded messages, will have to be sought if one is to understand brain pathways.

I thus propose that, in general, the situation in the CNS could be similar to that in the Mauthner system, information being embodied in anato-chemical channels, *independently* of whether these channels are activated or not at a given moment by, say, synchronous activity. The “meaning” (i.e., the useful link between signal and response) is defined (by phylogeny and ontogeny) before the channels are used, and is conveyed in full as soon as they are activated. Intermediate representations may be scrambled, but they are unscrambled by functional “homunculi.” Alternative views have been proposed by von der Malsburg,⁽⁷⁷⁾ who emphasizes “real-time” flexibility of neural connections in the elaboration of responses, and by Edelman who, if I understand correctly, posits that the brain does not use representations at all in its processing of information.⁽⁷⁸⁾

... or “complexity”?

It should be clear by now that application of the mathematical notion of information in biology requires much biological understanding. One of the latest avatars of information theory is “complexity theory”;⁽⁷⁹⁾ is it likely to fare better, as far as biology is concerned? This is doubtful, for the ideas behind “complexity” are essentially those behind “information:” thus, a “complex” system is usually construed as one requiring a “maximal” number of “information bits” for its description. We now know that this is a criterion which must be manipulated with great care, because such information bits, whether numerous or not, will not tell us anything biologically relevant

unless we also have something to say about their encoding and decoding.

Difficulties in this respect arise as much from our *description* of the problem as from the problem itself. The motion of 10^{23} or so particles in a cm^3 of gas is certainly “complex”, “chaotic” etc.; however, does such a statement reflect a “deep truth” about gases? On one side, underlying the “complex” motion are the laws of mechanics, which are simple; on the other, resulting from molecular movements are the laws of thermodynamics, which are simple too. “Complexity,” just like information itself, is in the eyes of the beholder (see above); it is a question of point of view, and science is precisely about finding the right point of view, not about wallowing in “complexity.”

Neural information: experimental and theoretical predictions

The present model predicts that sensory and motor representations linked directly to sense and effector organs—sensory and motor homunculi—might exhibit relatively slow evolutionary change, and mild ontogenetic variation, relative to the wider variety to be expected at the level of the deep “internal” representations instrumental to the enactment of subtle behavioral patterns, which representations also correspond grossly to evolutionarily more recent circuitry. This prediction entails that it might be difficult to observe these higher representations by methods such as functional magnetic resonance imaging⁽⁸⁰⁾ (fMRI), which often necessitate an averaging of measurements over several subjects and/or trials.

By its emphasis on cell memory, the model points to the necessity of studying in greater detail the molecular turnover and maintenance mechanisms that underlie the permanence of sensory, motor and other memories over time scales extending from weeks to years (see section “Comparison with experiment II: ...”). The crucial maintenance systems are likely to reside in neurons themselves.

The finely parcellated genetic tagging of brain tissue, down to the cellular level, and its hierarchical, modular organization, have been offered here as simple openings toward constructive theoretical thinking in neural development and functioning. While we cannot seriously expect that brain circuits and their function are determined by genes in complete detail, the model would nevertheless predict that a vast amount of the brain’s macro- and microcircuitry *is* established in a highly specific, invariant way. Experimental checks should become possible soon, as the mapping of brain circuits is now progressing rapidly.^(81,82)

Conclusions

The main idea of this paper is that most biological “information” resides in the biological objects—principally, the cells—which have to use it. Signals are simple, relative to their effect,

which is mainly to trigger responses based on cell-memorized information. In development, the current state of differentiation of a cell dictates how it will react to a subsequent stimulus; in the central nervous system, the significance of a cell's firing depends, not so much on the details of the impinging synaptic signals, as on their organization, which is the result of cellular memory embodied in neuronal shapes and synaptic structure. Oscillations, in this view, would primarily serve a supplementary, "on the fly" signal routing purpose.

Since the important factors in transmission are 1) the conditions under which the signal is emitted and 2) the action it triggers in the receiver, I suggest that the "conditions" in (1) and the "action" in (2) should, in general, be less variable in phylogenesis and ontogenesis than the signals and receptors that effect the transmission proper. These may be highly variable as they evolve in concert with the sole restriction of maintaining mutual coherence.

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