

STRYCHNINE NEURONOGRAPHY

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One of the most important contributions McCulloch and his immediate colleagues made to the empirics of nervous science was the development of the technique of strychnine neurography. By use of this technique it was possible to show in a few animals the enormous complexity of cortico-cortical connections in mammals, and particularly in primates. In spite of its enormous value the method has lapsed into desuetude in recent years for no obvious reason. I will try to review both the technique and the usefulness of its results.

Cortex of man and the other primates is a complicated arrangement of neurons. The surface of the brain can be divided into areas anatomically distinct one from the other by the patterns of organization in the depth of the gray matter. Anatomical neuronography, although done crudely by the staining of cell bodies and to some extent the fibers, is one of the modes of distinguishing functional parts of the cortex one from the other. Briefly, if one looks at a cross-section of the cortex in depth, the layering of the cells and of the fibers' connections to them is quite ordered, but the pattern of distribution of the cells and of the fibers vary strongly from one region to another, sometimes with sharp boundaries. Taking a particular pattern, one goes over the cortex as far as this pattern extends, delineates it and then continues to look at the bounding areas in the same way. Brodmann and the Vogts spent a great deal of time -- in fact most of their scientific lives -- in laying out the map of the cortex in terms of such distinctive areas. The number of areas into which the brain can be divided by neuronographic technique of this sort is somewhere in the neighborhood of 50 or 60. These areas were later given a definite functional significance in terms of their connections to

different sensory or motor modalities or in terms of their operation with respect to higher functions. Most of the information about the functions of these areas came from deletion experiments, that is, the kind of trouble that one gets with stroke or from stimulation experiments -- the kind of trouble that one gets from epilepsy. In addition, for the primary sensory areas as well as the primary motor areas, there was enough anatomical evidence from the major tracts going in and out to delineate very specific areas, for example, areas 17, 18 and 19 for the visual system, area 4 for the motor system, etc. Both cell type and cell distribution entered into the description of a particular area. So, for example, the large pyramidal cells exist principally in area 4, which is somehow or other connected to the motor function. But the connections of the cortex are not simply in and out of the brain. The cortex is completely connected to itself, not only locally within an area but between areas. Since some of the functional significance of the different areas was already known from clinical neurology, it seemed to neurologists to be a good idea to lay out a map not only of the areas but of their interconnections. While these connections could not be more than grossly established, they seemed to be important. Any detail about connectivity, namely, which fibers go to which layer in an area, would have to be left to the fine anatomy done by conventional methods.

With the many areas of the brain available and the enormous density of connections through the white matter, it appeared to be almost an endless task to pursue such a system anatomically; that is, the number of connections of an area to other areas would require a sacrifice of a single animal per area to be able to read the connectivity. The amount of work necessary to be able to lay out a complete map for a single area would be absolutely enormous even from the point of view of conventional mapping. Clinical neurology had already given enough hint that the connectivity of cortex to itself was incredibly important in the higher functions. The work of Hugo Liepmann as well as of Goldstein, of Dejerine and of a variety of other neurologists had already established a kind of nosology for higher functions in terms of the strokes or tumors that invaded the cortex. Let's not suppose a

simple phrenological interpretation of the Brodmann and Vogt areas; on the contrary, these were specialized regions in which specialized processing was devoted to different kinds of the higher functions for different areas. For empirical neurology, knowledge about connectivity between these regions was of the greatest importance for diagnostics.

In an earlier comment, Norman Geschwind says that McCulloch and his colleagues were so overborne by Lashley's insistence on the uniformity of the grey matter that the cortico/cortical connections were considered simply as an exercise. In spite of that comment, I do not believe that this is entirely true. Certainly from the clinical material available at that time which was, for example, from the demyelinating diseases invading the cortex, or from small tumors producing large effects, it was clear that unless one knew something about cortico/ cortical connections one would not be able to diagnose. We must admit that the sophistication of Kurt Goldstein, of Dejerine, of Liepmann had not yet invaded English and American neurology. But the material was there and already translated, as Geschwind has already pointed out.

The technique devised for revealing cortico/cortical connections is a somewhat strange one; that is, its operation depends upon the choice of a recording technique that accidentally excludes the kinds of electrical activity that one doesn't want. It is because of that apparent crudeness of the method that I think it has lapsed. Nobody, to my knowledge, has shown the sophistication of the way by which McCulloch and his colleagues came to that method.

The basis for the method was the discovery in the 19th century that strychnine was a convulsant compound. Strychnine was brought into the United States and to England early on in the 19th century, and by 1820 Magendie had already done a thesis on its action which is as modern as one should want. In every respect strychnine seems to imitate that dread disease, lockjaw, or tetanus, in which a patient exhausts himself through convulsions that go on to death. The convulsions are interesting in that they

are entirely motor, consisting of profound synchronous jerks all over the body without loss of consciousness. The jerks are extremely painful and, in fact, can be large enough in severe intoxication to actually break the bones of the patient. The compound was in use as a rat poison throughout the 19th and early 20th centuries in both England and the United States, as well as on the continent, and it was occasionally used as a poison by criminals. Possibly the most interesting case of this is that of Palmer who was hanged for murder on the basis of a superb forensic distinction of the effects of strychnine from the effects of tetanus. The evidence in that case, done in the 1960's, is probably a model for all forensic handlings of toxicology since then.

Dusser de Barenne had found that when strychnine is applied locally on the nervous system (that is, in the vertebrate nervous system -- in invertebrates the case is quite different), there is a volley, a synchronous discharge of the fibers that leave that region and go to other regions. This synchronicity of discharge is notable. It is larger in fact than one can get by almost any kind of electrical means, and it is clean in the sense that it is not distorted by the electrical artefacts from stimulation through an electric shock. In fact, what happens with strychninization of a region is that not only do the outgoing fibers fire in synchrony, but also the entering fibers fire backward in synchrony if they are not already occupied by their own discharges. Following such a volley coming out of the strychninized area, there is a period during which the neurons are far less excitable, the threshold has gone up enormously, and then the threshold drops over a period lasting several seconds, a new synchronous volley occurs. In a word, the tissue, instead of going through its ordinary operations, now behaves all-or-none. This fact, observed by Dusser de Barenne with his collaborator McCulloch, was then taken as a tool to exhibit the connectivity of the nervous system to itself. But two problems afflicted this tool. One was this: if the fibers which enter the region strychninized are fired as well as the fibers which leave that region, how could one distinguish one from the other and use the tool to investigate anatomical connections? To any one who has worked with nervous tissue, the problem is not a very difficult one. When one

fires axons synchronously in a forward direction, the impulses invading the terminal arbors of the axon set up large local sources and sinks whose recording as surface voltage signals is also quite large. That is to say, the signals set up by the forward-firing into their terminal pools of axons is an easily recorded event. Retrograde firing up axons in the reverse direction sometimes invades cell bodies, frequently does not, but in any case does not set up a very large signal at all. furthermore, the forward going signal, that is, the impulses proceeding synchronously down the fibers issuing from the region, not only set up a very large voltage in the tissue to which they feed, but one that endures for some time. That is, the transient evoked has a time constant of at least 30 milliseconds and generally more. *Per contra*, the retrograde impulse proceeding backward along the fibers that enter the region sets up a fast transient signal lasting no more than a few milliseconds and certainly not of the magnitude that is observed in the forward going case. Thus, if one set up simple filters for low-pass (such that only relatively long transients would come through), one could easily distinguish between backward-going and forward-going firing of the nerve fibers. In this way, one would be able to consider only the forward-going fibers to be the things recorded. Now the pen writer of an electroencephalographic machine of the kind that was in use in the 1930's and 1940's (for example, that built by Albert Grass or Frank Offner), was capable at best of frequency response to about 30 cycles per second at its upper limit. Thus, however synchronous a nerve volley might be, if its transient were quite short, just a matter of a few milliseconds, it would barely appear in the pen record. On the other hand, a volley that set up a transient which lasted well over 30 milliseconds would appear as a significant signal in the pen record.

A second feature was available in the setting up of the technique. Almost anywhere in the nervous system, if one set up a synchronous volley, say by a sharp electrical pulse, the consequences of that volley would be an excitement of cells or neurons in the regions where the axons ended. But one's relayed volley would only have a few components, so-called monosynaptic ones, related to the entry volley and most of the discharge would occur

in a very scattered fashion over a much longer period of time. The result is that secondary or relayed representation of the initial volley would have neither the magnitude nor the synchronicity of the initial one and thereby be rather easily distinguishable from the direct volley. This feature, tested out in several regions of cortex where the connectivity was known, could be established with a high degree of reliability. There was no case in which a secondary or relayed spike had the characteristics of the primary or direct spike conveyed from the strychninized region to the region to which the fibers directly projected.

With these features in mind, it was possible to use a relatively poor instrument, such as the EEG machine, for doing the best possible quick work in demonstrating cortico-cortical connections. It is interesting that the limited bandwidth of the recording was the one aspect of the instrumentation that rescued the whole procedure from ambiguity. This is not the first time that such a corruption has been of immense value. It is useful here to tell the anecdote about what happened with EKG machines when high fidelity was introduced. The EKG machines were invented at the turn of the century in Holland by Einthoven. They consisted of string galvanometers with a reasonably low bandwidth, certainly no more than about 30 or 40 cycles. The EKG recorded by this method was collected over tens of thousands of patients before a kind of norm could be established whence one could read disease processes. The establishment of this norm was far more important than the fidelity of the record, so that when in the 1940's and 1950's high fidelity electronic instrumentation with fast-writing pens was introduced, few clinicians could recognize disease in the new kind of record. The importance of convention and of filtering in the establishment of records that are readable is not something that can be decided *a priori*, but is given strictly by the practice itself. The notion that one must always have high fidelity available is nonsense, as anyone who has ever used a stethoscope can testify. With the criteria for reading off the strychnine spike available, McCulloch, Garol, Bailey, French and a variety of other colleagues decided to look at the cortico/cortical connections of macaque and chimpanzee. Using principally the Brodmann maps

as their guides, they set out the roadmap of principal cortico/cortical connections as carefully as they could by long-term use of a single animal. The experiments were tedious, lasting three to four days. The results, however, were prolific compared with the time of experiment of any other method of finding such connections. It is true that the connections were not specified in terms of the nature of the terminal arbor or any of the other details that many anatomists require. But there was no way of getting around the fact that the white matter connections, that is, the axonal connections from one region to another, were amply given by the method. None of the material revealed by the strychnine neuronography, as described here, has been controverted by the later and more tedious studies made by conventional anatomy. While connections could be discovered that were not revealed by strychnine neuronography, none of the connections described by strychnine neuronography were ever found not to exist.

The existence of these maps made in the 1940's by McCulloch and his colleagues could have served as the basis for arguing the disconnection syndromes later brought to light by Norman Geschwind, particularly in the case of the parietal lobe where the connectivity is most important and distinct between the two sides. For strychnine neuronography not only revealed cortico/cortical connections of one side but also revealed the callosal connections between the two hemispheres. Now that the callosal connections have become of such immense importance in the discussion of higher functions, it is a little bit surprising that the connectivity revealed by the strychnine neuronography has not been employed as much in argument as it should be; as far as I know it has not been employed at all.

The same feature that allows one to do strychnine neurography at the surface of the cortex between one area and another also permits neuronography to be done between cortical structures and subcortical structures, so that one can investigate the relationship between basal ganglia and cortex, thalamus and cortex, brain stem and cortex, by the appropriate use of local injection of strychnine into the regions whose fibers to the cortex

ought to be known, or in regions where local recording can tell whether a cortically-generated spike has arrived. Indeed, some studies were made between the caudate nucleus and various other subcortical nuclei and cortex by means of this technique. What has militated against the reliance on such a method has been a set of what can only be called superstitions of excellence. For example, it is commonly supposed by neurophysiologists that strychnine inhibits glycine which is the inhibitory transmitter supposedly in the spinal cord. If one reviews the evidence for glycine being such a transmitter, it is relatively paltry. Its only value seems to lie in the fact that it was difficult to get. Certainly glycine is not a compound that appears in the cortex whatsoever, and yet strychnine acts on cortex in the same way as it does on spinal cord.

Other experiments have suggested that strychnine acts somewhat like local anesthetics, such as xylocaine or novocaine. This is material which arises from the voltage clamp on axons. Yet, it is notorious that neither xylocaine nor the other local anesthetics produced the same convulsive activity when given either systemically or logically. The neuropharmacological questions raised are somewhat beside the point from the view of empirics, because if the method can be shown to work for whatever reason, suggestions as to mechanism are somewhat beside the point. Similarly, anatomists have raised the question of whether all the connections are revealed by strychnine neuronography. This again is not to the point, because I do not think that McCulloch or his colleagues ever proposed that this was the case. All that they proposed was that the connections revealed by strychnine neuronography were connections that could be verified by anatomy, and that certainly is true.

The snobbery associated with high technology has done more to wreck a simple and extremely useful procedure capable of revealing a great deal of material easily and in a short time, than to supplant it with procedures of greater resolution as easily done. It seems to me that with the present attention being paid to the disconnection syndrome, as voiced by Geschwind, the use of strychnine neuronography becomes more important than ever,

and that no tool developed since then would yield the material needed for clinical study as well as this tool did.