'Present-day research' is not 'fundamental research', and it moves further and further away from true 'basic research'. The state of present-day biological research can be seen, for example, in the program of the important 10th International Gerontological Congress in June 1975. The different contributions are already published in 2 volumes[1]. There are 660 lectures on social and clinical gerontological studies and about 110 reports on theoretical research in more than 20 different aspects of the functions of different organs in ageing.

One has the impression of a growing and endless diversion from what had started as a few certain facts, and one wonders whether this is the way to a fundamental understanding of the subject? More and more single problems are differentiated, and the distance from the central core of the question becomes rapidly greater.

The large subject of the behaviour of atoms, molecules, ions, protons, neutrons and radiation, as it is known to us in the discipline of cosmic and terrestial physics, up to the mystery of gravitation which dominates our whole environment, is hardly touched – unless in the publications of HINES[2], the astro-physicist, which of course are not in the field of biological problems.

We have come to the stage as if a plant had grown out from the seed and produced twigs and branches; the continually changing effects of the developing structure can no longer give us any information as to the basic *forces* which are at work.

Examples of the path of real research are (amongst many others, ofcourse) two deeply thought out studies: one by BERNAL and SYNGE[3] on the development of the animal species, worked out with the greatest knowledge and logic; and the other by WOOLHOUSE[4] which describes in a similarly admirable way the development of plant organisms. Both these authors are unconcerned by the fact that they must work with periods of time in the order of 1,000 million years, and with no certain data in space or time. The development and progress of animals not less than plants spreads into endless variation of form and function.

In this mass of reactions, there is not a comprehensible order. Unexpected new reactions lead to new products, the mutations', without us finding any other cause than 'chance'. New forms appear: many are instable and are immediately changed. Others retain their new characteristics, and retain them so long as they are not again changed in a new 'chance' situation. A particular situation may lead to a very stable combination and to one which will repeat itself in a long series of reproductions.

In this way, perhaps, that new combination arose which had the characteristic 'Life'.

We know some of the conditions for the appearance of life, which would require relatively low temperatures (below 60–90 °C), water and the presence of certain inorganic and possibly organic substances to react with one another.

For the phenomenon 'Life' there does not seem to be one specific physico-chemical characteristic. But the factor 'Life' can under some circumstances survive, and then it forms 'new life'.

When and where does Life occur? No one will doubt that great changes in dispersion of energy may have nothing to do with Life: the fall of a rock in the Alps represents a change of energy, but not life.

Life appears first when the concept of 'non-living' or 'death' appears. The concept of 'Life' is bound to the notion of something arizing in an environment which is non-living, which is dead.

Conditions for the production of life have been sought. For instance, oxygen was believed to be one of the essential conditions of life, as observations on vertebrates with their blood circulation and tissue respiration indicated. Now we know, however, that in the depths of the sea, where living organisms exist, the oxygen pressure would not be sufficient to provide the cells with oxygen and support oxydation.

Some years ago, it was found that in the interstellar space there are simple amino acids, which could by a chemical synthesis form proteins.

Such the *conditions* of life are known, but when and where they first arose is unknown. Of all the different systems known to us, there is none which is identical with life. Must we consider life to be a unique form of energy?

'Life' is only to be conceived where there is an environment of 'death'. The concept of 'death' is the negative of the concept of 'Life'. Life represents a particular and unique form of energy, which is a concept in the field of philosophy.

The problem *Death–Life* remains a totally unsolved mystery within our astro- and geo-physical universe. Basic research must be recognized as the path towards its ultimate understanding.

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PRO EXPERIMENTIS

An 'Ultra' Rapid Golgi Method for Vertebrate Neuroanatomy

MILENA KEMALI[1]

Laboratorio di Cibernetica del C.N.R., Via Toiano 2, I–80072 Arco Felice, Napoli (Italy), 30 December 1975.

Summary. A modification of the hardening solution of the rapid Golgi method permits constant successful impregnations of the brain of several vertebrate representatives in only 24 hours.

The Golgi method is a descriptive anatomical technique which, when compared to other histological methods, provides the image that most distinctively characterizes the nervous system: the shape and the spatial arrangement of isolated nerve units. Its greatest advantage is selectiveness as only 1 to 5% of the elements in a field appear impregnated, often with startling completeness and clarity on a nearly colourless background.

There are three main methods of GOLGI[2] himself and multiple modifications of the procedure used have been developed for many years[3], but the rapid method of Golgi is the one most frequently used on fresh animal

tissue. It consists essentially of immersing fresh pieces of nervous tissue, a few millimeters thick, first in a solution containing potassium dichromate and osmic acid and subsequently in a solution of silver nitrate. Then follows a wait of some days, sometimes weeks. A black deposit of a reduced silver salt will be formed in the processes and cell bodies of many neurons.

The success of the Golgi method depends on a number of invoked variables: time of permanence of the pieces in the hardening and reducing solutions; thickness of the pieces; temperature at which the process takes place; the age of the individual; the region of the brain. The fixation by heart perfusion is often considered a necessary step in the success of the impregnation[4].

The modification of the rapid Golgi procedure which we have set up has permitted us 1. to have always successfull impregnations; 2. to impregnate, in the usual selective manner, all the zones of the central nervous system; 3. to shorten the period of the complete procedure to only 24 h.

The procedure, described in detail elsewhere[5], is based mainly on the use of a 2% osmium tetroxide solution buffered with sodium barbital at pH 7.2 planned originally for electron microscopy fixation for the frog and consequently adjusted to match the osmolarity of amphibia[6]. To 1.5 parts of this buffered solution, 8.5 parts of 3% potassium dichromate was added. No perfusion was used and the brains in toto of several amphibia representatives (*Rana esculenta*, *Xenopus laevis*, *Triturus cristatus*) were immersed in the hardening and successively in the reducing solutions for a period of 12 h in each.

Surprisingly, the same solutions gave also substantial results (Figures 1 and 2) when pieces of brains of rabbits and mice more than 1 cm thick and brain in toto of bats were immersed in it for the same period of time as for amphibia.

The constant success of the impregnation, the fitness of the method to various vertebrate representatives, the fact that cardiac perfusion is not necessary, the 'ultra' rapidity of the procedure and the economical aspect, that is the possibility of keeping a stock of buffered osmium tetroxide solution at 4 °C for long periods of time, are advantages which cannot be disregarded.

The reasons why the adding of a buffer transforms the rapid Golgi solution in a suitable vehicle of impregnation for any brain and for any cerebral zone is unknown. The chemistry of the rapid Golgi method is beyond the scope of this paper. The aim of the present publication is to underline the fact that for the first time it is possible to give the rules for obtaining successful Golgi impregnations, rules as to both the composition of the hardening solution and the duration time of the immersion of the brains in it.

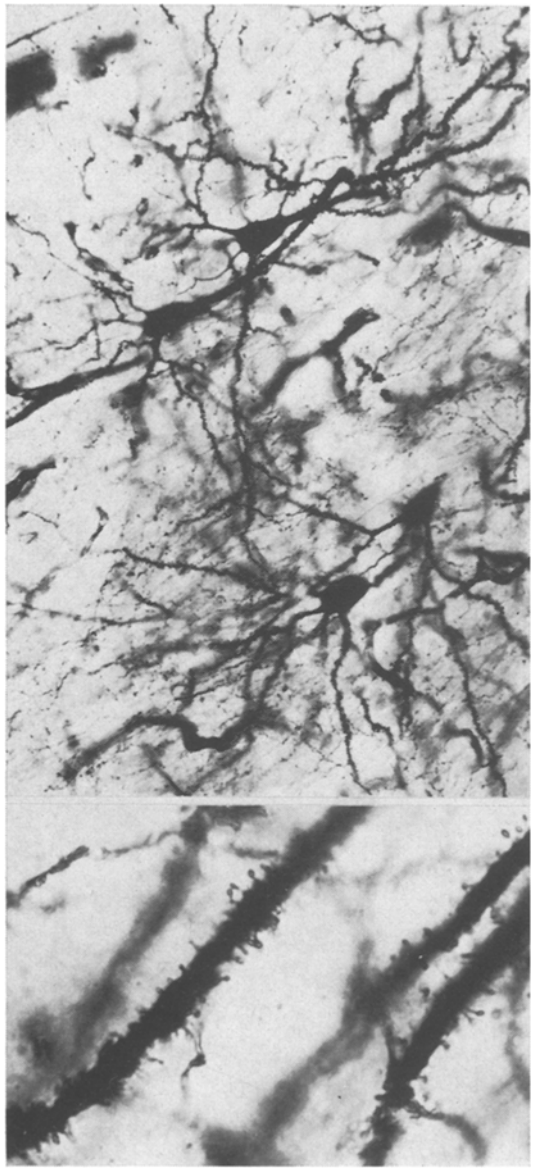

Fig. 1. Cerebral cortex of the rabbit. × 260. Geometric arrangement of pyramidal cells in 'columns' oriented vertically to the cortical surface. The apical dendrites directed vertically and the basilar dendrites directed horizontally show numerous spines. The axons of 2 neurons emerging from the base of the cell body are visible. In the background, the intricacy of an axonal net is apparent.

Fig. 2. Cerebral cortex of the rabbit. × 1,200. Detail from a 'bundle' of dendrites showing the numerous spines which greatly increase the receptive surface area of the neuron.

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