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DNA Content of Neurons in the Cat Hippocampus

Abstract. *The DNA content of individual cell nuclei of cat hippocampus has been measured by means of a scanning cytophotometric technique. The pyramidal cells are tetraploid, whereas the remaining cell types, which include glial cells, interneurons, and the granule cells of the dentate gyrus, are diploid. Nuclei of tetraploid pyramidal cells are significantly larger than those of diploid granule cells.*

The finding of tetraploid amounts of DNA (*1*) in normal human (*2*) and rat (*3*) cerebellar Purkinje cells and in autonomic ganglion cells (*4*) has extended knowledge of normal polyploid states in mammals to include the nervous system. The extent of this phenomenon in the nervous system is unknown.

Three cats (2.5 to 3.7 kg) were anesthetized with pentobarbital sodium (25 mg per kilogram of body weight) injected intraperitoneally and perfused through the left ventricle with Ringer solution. The animals were then perfused with a mixture of glacial acetic acid and 95 percent ethanol (1:3). Brain and spinal cord were removed, and blocks were processed in the usual manner for paraffin embedding.

Sections were cut at 22 μ for measurement of hippocampal pyramidal cell nuclei and at 13 μ for measurement of dentate granule cell nuclei. They were stained by the Feulgen reaction (*5*). Absorption of monochromatic light (550 ± 15 nm) by single nuclei was measured with a Barr and Stroud integrating microdensitometer (*6*). Results in arbitrary units represent total nuclear DNA content.

Two cell populations within the hippocampus are easily identifiable morphologically and functionally. The dentate gyrus, consisting of tightly packed granule cells, forms the primary receptive area (*7*). The pyramidal cell layer is composed of large, loosely packed cells and forms the main projection pathway from the hippocampus (*8*). In addition, there are various interneurons and glial cells (*9*).

Figure 1 shows the DNA content in the various cell types of the hippocampus. Glial cells and interneurons form a single population having diploid amounts of DNA (Fig. 1A). In order to establish the tetraploid content of DNA, two diploid glial or interneuron nuclei were measured within the same field, the same conditions of measurement applied to pyramidal cell nuclei being used (Fig. 1A).

The DNA values for granule cells of the dentate gyrus are diploid (Fig. 1, A and B). The pyramidal cell nuclei (Fig. 1B) have a total DNA content significantly higher ($P < .005$) than that for the other cell types measured, comparable to that amount from two glial or interneuron nuclei in the same field (Fig. 1, A and B).

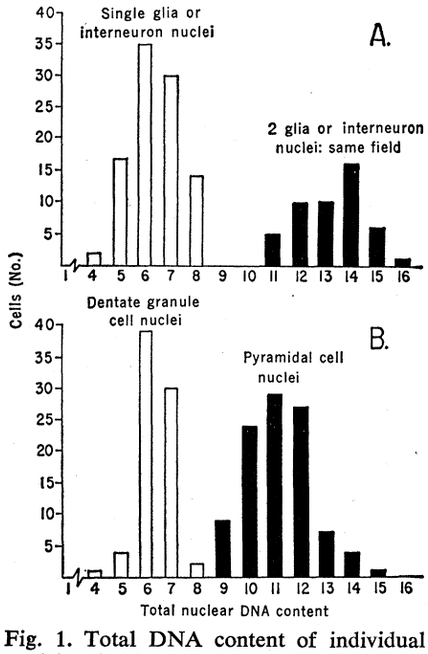

Fig. 1. Total DNA content of individual nuclei of hippocampal cells. Ordinates represent number of cells; abscissas represent the total nuclear DNA content in arbitrary units. (A) Glial and interneuron nuclei (□); 98 nuclei; DNA content, 6.4 ± 0.10 (average ± standard error). Two glial or interneuron nuclei in the same field, measured together (■); 48 nuclei; DNA content, 13.2 ± 0.20. (B) Dentate granule cells (□); 76 nuclei; DNA content, 6.4 ± 0.06. Pyramidal cell nuclei (■); 101 nuclei; DNA content, 11.2 ± 0.10. Data represent pooled values from three animals.

Sections (22 μ) from one of the three animals were treated for 23 hours with deoxyribonuclease before Feulgen staining. Following digestion, measurements of eight pyramidal cell nuclei averaged less than 1 unit, an indication that only a minimum contribution of nonspecific absorption is due to material other than DNA.

Associated with their higher DNA content, 75 pyramidal cell nuclei measured 14.8 ± 1.5 μ (mean ± S.D.) in diameter. Seventy-five dentate granule cell nuclei measured 10.3 ± 2.6 μ in diameter. This difference in size is highly significant ($P<.001$).

Of the cell types in the hippocampus, the pyramidal cells are unique in having larger nuclei with a higher nuclear DNA content. In light of the measurements on two small nuclei in the same field, the most logical interpretation of these data is that the pyramidal neurons are tetraploid. Thus, tetraploidy within the higher integrative centers of the nervous system is not an isolated phenomenon of Purkinje cells. Exploration of the extent of the phenomenon and its correlation with histochemical and physiological data on the cell types involved may give some clue to its functional significance in the nervous system.

CHESTER J. HERMAN
LOWELL W. LAPHAM
Department of Pathology, University of Rochester, Rochester, New York

References and Notes

1. "Polyploidy," "tetraploidy," and so forth, as used in this paper refer only to the amount of DNA in the cell nucleus, without implying a doubling of chromosomes rather than chromatids.
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