Zhurnal Nevropatologii i Psikhiatrii imeni S. S. Korsakova, vol. 72, no. 7, pp. 1024-1029, 1972.

ABSOLUTE NUMBER OF NEURONS AND THICKNESS OF THE CEREBRAL CORTEX DURING AGING, SENILE AND VASCULAR DEMENTIA, AND PICK'S AND ALZHEIMER'S DISEASES

V. F. SHEFER

Department of Experimental Diagnosis and
Laboratory of Brain Pathomorphology,
Bekhterev Research Institute,
Leningrad

The absolute number of neurons and thickness of the cortex were studied in areas 6, 10, 18, 21, 21/38 and 40 and in the subiculum of mentally healthy persons of different ages and in patients with senile and vascular dementia or with Pick's or Alzheimer's disease. In old age the mean absolute number of neurons in mentally healthy persons is reduced by 20%, while the thickness of the cortex on the free surface of the gyri remains unchanged. In persons with senile and vascular dementia the number of nerve cells is reduced by 35-38%, but there is no decrease in the thickness of the cortex on its free surface. In Alzheimer's disease the number of nerve cells is reduced by half and the thickness of the cortex by 6%. Pick's disease is characterized by mass death of nerve cells in the affected areas, leading to a reduction of 14-30 in their number, and to a decrease in thickness of the cortex by half. The subicular cortex thickness in old age is reduced by 28% and in diseases leading to dementia by 47-71%.

It is generally considered that in old age and, in particular, in diseases leading to dementia some of the nerve cells in the cortex die, as a result of which atrophy of the brain develops and its cortex becomes thinner. However, it is not known which portion of the total number of nerve cells dies or whether there is a relationship between the severity of diseases leading to dementia and the number of nerve cells found in the cortex. I found no reference to any investigation in which the absolute numbers of nerve cells in the cerebral cortex were compared in young and old or in patients with various types of dementia. Methods of determining the absolute number of nerve cells in the cerebral cortex are also undeveloped; I know of three papers describing studies of the thickness of the cortex and its layers during normal aging of the brain, but of no similar study of the dementias.

Dalakashvili (1) concluded that in all areas (1, 4, 9 and 19) the cortex thickness in the middle-aged is reduced by 1-4% and in old persons by 2.7-10.7% compared with the brain of a man aged 35 years used as the control (one case). Sheinina (2) who measured the thickness of area 45 in persons 7 to 105 years old found that the cortex becomes thinner in old age. However, her value for the thickness of the cortex in a person 105 years old (2.34 mm) lies within normal limits (2.5-2.3 mm) as given in (3). Haug (4) states that there is no difference in the thickness of the cortex and its layers in areas 17 in persons 15 to 75 years old. Unfortunately these workers do not describe the cortical surface where the measurements were made.

The objective of my investigation was to calculate the ratios between the absolute numbers of nerve cells in layer III of certain cortical areas of the brain and to measure the thickness of the cortex and its layers in the same areas in the following groups: 1) young persons from 19 to 28 years old with no mental disease and dying from accidental causes (5 persons). This group will subsequently be named the control; 2) ten mentally healthy old persons (mean age 77 years); 3) six patients (mean age 79 years) who suffered from senile dementia; 4) six persons (mean age 71 years) in whom a vascular dementia had been diagnosed clinically; 5) six patients (mean age 67 years) with Alzheimer's disease; 6) six patients (mean age 55 years) with Pick's disease. In two patients with Pick's disease only the temporal lobes were affected; in the other four patients the frontal and temporal lobes were affected.

The brains were fixed in formalin, embedded in celloidin, and sections were stained by Nissl's method. The ratios between the absolute numbers of nerve cells in layer III of the free cortical surface of the left hemisphere were determined in areas 6, 10, 18, 21, 21/38 and 40 and also in the subiculum (the archicortex in the depth of the hippocampal fissure).

To calculate the absolute number of nerve cells in cortical layer III in a given area it is necessary to know the volume of this layer (V) and the mean number of cells (a) in a known volume of this layer (v). Clearly the absolute number of nerve cells (A) in layer III of the area studied can be expressed by the equation:

$$A = \frac{V}{v} \times a$$

V can be calculated by the equation:

$$V = L \times M \times H$$

where L is the length of the layer; M its width; H its height. The volume of brain substance in one field of vision of the section under constant magnification was taken as v. Its value is found by the equation:

$$v = S \times h$$

where S is the area of a field of vision of the microscope under constant magnification; h the thickness of the section in the same field of vision. The number of nerve cells (a) is then counted. By substituting the values of V and v in the equation for calculating the absolute number of nerve cells it assumes the following form:

$$A = \frac{V}{v} \times a = \frac{L \times M \times H}{S \times h} \times a$$

However, to compare absolute numbers of nerve cells in the various groups of subjects, an equation can accordingly be deduced. Assume that the absolute number of nerve cells in one particular group of subjects is A_1 (all terms in the equation are marked by the corresponding subscript 1), and that the absolute number

in the group taken for comparison is A_2 (all other terms are given the corresponding subscript). The equation for determining the ratio between the absolute number of nerve cells (K) will then be:

$$K = A_1 : A_2 = \left(\frac{V_1}{v_1} \times a_1\right) : \left(\frac{V_2}{v_2} \times a_2\right) = \frac{V_1 \times a_1 \times v_2}{V_2 \times a_2 \times v_1}.$$

By substituting the values of V and v in the equation it assumes the following form:

$$K = \frac{(L_1 \times M_1 \times H_1) \times a_1 \times (S_2 \times h_2)}{(L_2 \times M_2 \times H_2) \times a_2 \times (S_1 \times h_1)}.$$

Since S is a constant, the final equation is:

$$K = \frac{L_1 \times M_1 \times H_1 \times a_1 \times h_2}{L_2 \times M_2 \times H_2 \times a_2 \times h_1}.$$

To calculate the ratios between the numbers of nerve cells in the groups of subjects it is essential to know the length, width and thickness of layer III of the cortex area investigated, the mean number of nerve cells in the microscopic field, and the mean thickness of the section where the nerve cells were counted. Evidently the width and length of the free surface of layer III in a particular gyrus are directly proportional to its width and length. The ratios between the length and width of the gyri of a given area of cortex will thus be equal to the same ratios between the width and length of the free surface of layer III. When the value of K is determined for the cortex area and not for an individual gyrus, the number of gyri in that area must be taken into account. However, since the number of gyri in the brain is on the average constant, and evidently unchanged with age, it was decided that this value could be disregarded. Simple calculation shows that the length of a wavy line (the gyri have approximately this form) varies directly proportionally to the distances between the extreme points of the wavy line perpendicularly to the axis. The ratios will thus be equal to the ratios between the lengths of the corresponding wavy line. To judge the length of the gyri, three basic measurements were used: 1) the shortest distance from the frontal pole (the most anterior point) to the precentral gyrus at the level of the first frontal fissure. This distance is a sufficiently objective criterion of the change in length of the frontal gyri; 2) the shortest distance from the frontal pole to the occipital (after deduction of the preceding value from it). This was used as the criterion of a change in length of the occipital and parietal gyri; 3) the shortest distance from the temporal to the occipital pole. This value was used to estimate changes in length of the temporal gyri. To determine the mean width of a gyrus or of the gyri of a particular lobe (M) the width of all gyri of the frontal lobe and temporal lobe and of the supramarginal and precentral gyri was measured. Up to 400 such measurements were made on the left hemisphere of each brain. The thickness of layer III (H) on its free surface was measured in histological sections under the microscope by an ocular micrometer. Under a magnification (90×7×1.5) the nerve cells were counted in 20 fields of vision of layer III_3 of the free cortical surface. The mean number of nerve cells (a) in the field of vision was determined. The fields of vision counted formed a continuous series. Only those nerve cells in which a nucleus and nucleolus could be distinguished were counted. In each field of vision in which the number of nerve cells was counted thickness of the section was measured. The mean thickness of the section was then calculated (h). To determine the thickness of the section the upper and lower points of the section were focussed and the distance measured by the vernier of the micrometer screw. As a result of these measurements values expressed in different units of measurement were obtained. It is unnecessary to reduce them to the same metric unit, however,

for in accordance with the equation derived it is the ratio between these values which matters, and this would be unaffected by the introduction of the same correction factor into both numerator and denominator. All the data were thus obtained for calculating the ratios between the absolute numbers of nerve cells in different groups in the cortex.

In the equation for determining the absolute number of nerve cells in area 6 the index (M) reflected the mean width of the precentral gyrus, while the term L was disregarded for no sufficiently objective criterion for it was available. In area 10 the index M was taken as the mean width of all the frontal gyri, and the index L as the distance from the frontal pole to the precentral gyrus. In area 18 the index M reflected the mean width of all the occipital gyri, while the index L reflected the distance from the precentral gyrus to the occipital pole. In area 21, M represented the mean width of the middle temporal gyrus and L the distance from the temporal to the occipital pole. In area 21/38, M was taken as the mean width of the temporal gyri and L as the distance from the temporal to the occipital pole. In area 40 the factor M reflected the mean width of the supramarginal gyrus and L the distance from the precentral gyrus to the occipital pole. When the number of nerve cells in the subiculum was determined the factors M and L were disregarded and factor H was taken as the thickness of the cortical lamina.

The method used is not perfect but it can provide approximate data for the absolute number of nerve cells in different parts of the cerebral cortex.

The results obtained for the absolute numbers of nerve cells in the control and other groups are given in Table 1. The corresponding data are presented as ratios between the number of cells in the control group and the number of cells in each of the other groups.

Comparison of the number of cells in the cortex of mentally healthy persons dying in old age with the number of cells in the brain of the control subjects i.e., persons dying at the age of 19-28 years, reveals a decrease in the absolute number of nerve cells in old age in all regions of the cortex. This decrease was particularly marked in areas 6 (by 22%), 10 (by 28%) and 21 (by 23%) and the subiculum (29%), with an average decrease of 20% for all regions (the index for the control group was always 100%).

In senile dementia the decrease in the number of nerve cells was greater than in the previous group (on the average by 38%). It was particularly marked in areas 10 (35%), 21 (by 47%) and 21/38 (by 37%) and in the subiculum (by 60%). This suggests that in this disease more nerve cells die in the frontal and temporal lobes and in the subiculum than in the precentral gyrus (area 6) and the parietal and occipital lobes (areas 40 and 18).

In vascular dementia the mean decrease in the number of nerve cells was by 35%. The considerable decrease (by 36 %) in the number of nerve cells in the motor cortex (area 6) in this disease is noteworthy. On the whole, the character and degree of the changes have much in common in senile and vascular dementia.

In Alzheimer's disease the decrease in the absolute number of nerve cells in all regions except the subiculum was greater than in the groups of patients with senile and vascular dementia. The mean decrease in the number of nerve cells was about 50%, falling to approximately 48% of their number in the brain of the control group. By contrast with the groups with senile and vascular dementia, in this disease there is a well-marked deficiency of cells (a decrease of 46%) in area 40 (the supramarginal gyrus). In area 18 the absolute number of nerve cells was reduced by 39% and in area 10 by 43%. In all regions of the cortex the number of nerve cells was reduced approximately by half.

In Pick's disease the changes observed were different and consistent. In this group the mean index across brain areas was calculated (8.05), but is relatively useless because it obscures the local character of the changes in this disease.

TABLE 1. Ratios between Absolute Numbers of Nerve Cells in Groups Studied

Groups compared with control	Ratios between numbers of nerve cells in control group and all other groups							
	Cortical regions investigated							
	6	10	18	21	21/38	40	subiculum	for all cortical regions
Old, mentally healthy persons,	1.29	1.4	1.16	1.3	1.14	1.18	1.41	1.27
Senile dementia	1.2	1.53	1.34	1.87	1.59	1.3	2.46	1.61
Vascular dementia	1.58	1.45	1.26	1.75	1.35	1.44	1.94	1.54
Alzheimer's disease	2.04	1.74	1.65	1.96	2.02	1.86	2.16	1.92
Pick's disease	1.32	5.17	1.1	14.0	29.8	1.7	3.26	8.05

TABLE 2. Mean Thickness of Cortex (in conventional units)

Group	Thickness of cortex					
	in layers					Over-all
	I	II	III	IV	V-VII	
Control	3.1	2.5	11.7	3.2	19.3	39.8
Old, mentally healthy persons	3.2	2.5	12.2	3.4	18.6	40.0
Vascular dementia	3.7	2.8	11.8	3.5	18.7	40.5
Senile dementia	3.1	2.7	11.0	3.8	18.6	39.2
Alzheimer's disease	3.2	2.5	10.8	3.3	17.4	37.2
Pick's disease[1]	3.0	2.4	10.2	3.5	15.3	34.4
	2.0	1.8	7.4	2.4	9.3	22.9

[1]Top row of numbers — measurements in cortical areas 6, 18, and 40; bottom row — measurements in areas 10, 21 and 21/38.

TABLE 3. Thickness of Cortex in Subiculum (in conventional units)

Group	Thickness of cortex	Change compared with control group, in %
Control	28.5	
Old, mentally healthy persons	20.6	72
Vascular dementia	15.1	53
Senile dementia	12.8	45
Alzheimer's disease	12.4	43
Pick's disease	8.3	29

Pick's disease is characterized by mass death of the nerve cells in areas 10, 21, and 21/38, with a resulting decrease in the absolute number of nerve cells in these regions by 5, 14, and 30 times respectively. The figures obtained for the frontal lobe (area 10) do not reflect the true picture, for of six cases of Pick's disease studied a characteristic lesion in the frontal lobes was observed in only four. If one considers, however, only the cases with frontal lobe involvement, the decrease in the absolute number of nerve cells in area 10 is approximately the same as in area 21/38. In Pick's disease there was the greatest (threefold) decrease in the number of nerve cells in the subiculum. In area 40 it was greater than in senile and vascular dementia and was approximately the same as that observed in Alzheimer's disease (a decrease of 41 %). The number of nerve cells in areas 6 and 18 in Pick's disease was the same as in the group of mentally healthy old persons, i.e., it was evidently within the normal limits for that age group.

Changes in the thickness of the free surface of cortex and its layers are presented in Table 2. The mean thickness of the cortical layers on its free surface are shown. The results indicate that the thickness of the cortex and its layers in young and old mentally healthy persons and in persons with senile and vascular dementia are practically identical. In Alzheimer's disease the thickness of the cortex is reduced on the average by 6%. In persons with Pick's disease, on the other hand, in the less severely affected regions (areas 6, 18 and 40) the thickness of the cortex is reduced by 14%, while in the most severely affected regions (areas 40, 21 and 21/38) it is reduced by almost half. When some of the nerve cells die and there is no increase in the absolute number of glial cells (these also were counted), there is no decrease in the thickness of the cortex on its free surface. A possible explanation of this fact is that the volume of cortex on the free surface is reduced mainly through narrowing of the gyri (Fig. 1). In Alzheimer's and Pick's diseases, however, this evidently does not compensate for the reduction in volume of the cortical substance, so that the cortex becomes thinner.

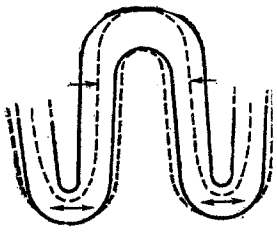

Fig. 1. Schematic representation of changes in transverse section through gyrus.

Measurements of cortical thickness in the subiculum (given in Table 3) indicate 1) a decrease in the thickness of this layer in old, mentally healthy subjects by 28%, in vascular dementia by 47%, in senile dementia by 55%, in Alzheimer's disease by 57%, and in Pick's disease by 71% and 2) a marked decrease is characteristic of these diseases.

The results described in this paper reflect changes in the nerve cells in layer III of the cortex and in the subiculum only and without verification they cannot be applied to other layers of the cortex or to other regions of the brain.

REFERENCES

1. DALAKASHVILI, S.M. The State of the Structure of some Cortical Areas of the Brain during Aging. Tbilisi, 1967.
2. SHEININA, L.I. Soobshch. Akad. Nauk Gruz. SSR. 35:413, 1954.
3. Atlas of the Cytoarchitectonics of the Human Cerebral Cortex. Moscow, 1955.
4. HAUG, H. Quantitative Untersuchungen an der Sehrinde. Stuttgart, 1958.