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Qualitative and quantitative analysis of the postnatal development of the ventroposterolateral nucleus of the thalamus in rat and rabbits

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The morphometric analysis of changes occurring in the rat and rabbit ventroposterolateral (VPL) nucleus of the thalamus during the postnatal development was performed using unbiased stereological methods. The materials used in the study included 30 Wistar rats and 32 New Zealand rabbits aged from P0 to P180 (P-postnatal day), which were divided into six and eight age groups, respectively. The following stereological parameters of VPL nucleus on the cresyl violet stained sections were determined: volume of the nucleus, numerical density and total number of neurons.

The total number of neurons indicated that the development of VPL nucleus in both species ended within the third week of postnatal life. The volume of VPL nucleus increased gradually (by about 2.2 and 5 times in rats and rabbits, respectively) in comparison with the volume of the cerebral hemisphere during the development from P0 to adulthood. The numerical density of VPL neurons decreased rapidly at the beginning of postnatal life and stabilized by the end of the third week. In both species, the gradual increase in the volume of VPL nucleus and the simultaneous decrease in the neuronal density in the first week of postnatal life were mainly caused by changes in the neuropil volume. The total number of cells did not change remarkably during the first postnatal week. However, it decreased significantly during the second week. This decrease was probably due to the naturaly occuring cell death.

These results show that the most prominent qualitative and quantitative changes in VPL nucleus and its neurons occur during the first two weeks of postnatal life of rats and rabbits. Also, because the thalamocortical relay neurons completely acquire their physiological features, this the most critical period of time for their morphological maturation.

key words: development, somatosensory thalamus, VPL, stereology, rat, rabbit

INTRODUCTION

The rat somatosensory system is a good model for studying sensory function, neural integration, development, response to injury and neural plasticity. Its advantages stem from an obvious correlation between structure and function. The rat somatosensory thalamus has been well examined [56]; contrary to this, we still know very little about the rabbit's

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thalamus. Generally, in mammals, ascending somatic sensory information is relayed to the primary somatosensory cortex through two major thalamic nuclei: ventral posterior (VP) and posterior (Po). VP consists of two subdivisions: ventroposterolateral (VPL) and ventroposteromedial (VPM) nuclei. VPL nucleus receives tactile inputs from the trunk, limbs and tail via the dorsal column nuclei and the spinal cord, which further transmits them to the somatosensory cortex in the topographically ordered manner [14, 26, 27, 38, 43, 64, 67].

VPL nucleus in mammals is composed of medium-sized multipolar neurons which are arranged in the laminar pattern parallel to the external medullary lamina [56]. But there are differences in the cellular constituents of the somatosensory thalamus between interspecies, e.g., cats and monkeys have numerous GABAergic neurons, which constitute about 20–25% of the total cellular population [49, 63, 69]. Probably, the population of interneurons in rabbits is on the similar level. In contrast, VP of an adult rat lacks local circuit neurons [8, 41] or they are less than 1% [29]. It contains only one major cell type, the thalamocortical relay cell, that processes all the somatosensory submodalities present in the nucleus [28, 30, 55].

The prenatal development of VPL nucleus in rats and rabbits is well known. Altman and Bayer [5, 6] showed in a series of autoradiographic studies that most cells of VPL nucleus in rats are generated on E15, whereas in rabbits this event takes place on E16 [15]. Before the day of birth, VPL nucleus is completely differentiated and shows a distinct boundary in comparison with the surrounding structures. On the basis of numerous studies on the prenatal development of thalamocortical connections [31, 37, 50–53, 70], it was concluded that the developing neurons send their axons to the subplate and create transitory connections with its cells waiting for the development of the cerebral cortex. For this reason, the developing neuron passes through a progressive series of morphological and metabolic alterations during maturation. It is forced through the flow and circulation of various sensory inputs in the thalamocortical loop during the postnatal period of life. The establishment of functional connections with other neurons or with end-organs in the periphery accelerates neuronal differentiation and is known to play an important role in neuronal maintenance [18, 19]. This period of time is different in various species and reliably depends on the degree of advanced intrinsic organization of VPL nucleus. Therefore, it seems interesting to compare the developmental events of VPL nucleus in rats and rabbits because of their differences in cytoarchitectonical structures.

In the present study, we investigated the qualitative and quantitative developmental changes in VPL nucleus neuronal population in rats and rabbits. In relation to this, we also analyzed the sequence of morphological developmental alterations in both species.

MATERIAL AND METHODS

Thirty Wistar rats and thirty two New Zealand rabbits of both sexes respectively divided into six and eight age groups (P0, P7, P14, P21, P45, P180; and P0, P7, P14, P21, P60, P90, P120, P180) were used in this study. The animals were cared for and treated in accordance with the guidelines for laboratory animals established by the National Institute of Health as well as by the local ethical committee. The animals were deeply anesthetized with Fentanyl (0.03 mg/kg i.p.) and a lethal dosage of Thiopental (80 mg/kg i.p.). Next, they were perfused transcardially with 50–250 ml (depending on the age of the animal) of cold 0.9% saline containing 10,000 units of heparin followed by 100–1000 ml 4% solution of paraformaldehyde in 0.1 M phosphate buffer (pH 7.4 and 4°C) and 50–300 ml of 10% solution of sucrose in 0.1 M phosphate buffer. Immediately after perfusion, the brains were removed from the skulls and refrigerated in 30% solution of sucrose in 0.1 M phosphate buffer overnight. Then, the brains were frozen and cut into 50 μm coronal sections with a cryostat Jung 1800 (Leica, Germany). Every sixth and tenth pair of sections (for rats and rabbits, respectively) were saved, mounted and stained using the cresyl violet method. After that, the sections were covered with DPX slips (Fluka, Germany) and studied under a light microscope (DMLC, Leica, Germany). The microscope was equipped with a camera (DP10, Olympus, Japan). To capture the photomicrographic collages of the thalamus, the image analysis system Q500 was used.

Stereological study

In the current study, the following stereological parameters of VPL nucleus were determined: volume, numerical density and total number of neurons. The volume of the cerebral hemisphere and the ratio of VPL nucleus volume to the volume of the cerebral hemisphere were also determined. The parameters were determined using the optical dissector and fractionator and the Cavalieri's principle. A microscope

equipped with a CAST grid system was used (microscope BX-51 (Olympus, Japan) [34, 66, 68].

The mean and standard deviations were calculated for each parameter. The statistical analysis was performed using a computer programme (Statistica v. 5.5, Statsoft, Poland). The analysis of variance ANOVA with posthoc LSD test (least significant difference test) was used to compare results for all age groups. The experiment-wise level of significance was 0.05. The data in the tables are given as mean ± standard deviation.

RESULTS

Qualitative analysis

General topography and morphology of VPL nucleus. VPL nucleus was clearly distinguishable from the surrounding structures on the day of birth in both species. On coronal sections, it was medially and rostrally near to the ventral lateral thalamic nuclei and medially and caudally near to the ventroposteromedial thalamic nuclei. Laterally, the boundaries of VPL nucleus are delimited by the external medullary lamina, which separates it from the reticular nucleus of the thalamus. The most medially and ventrally situated part of VPL nucleus is close to the ventral medial thalamic nuclei. On coronal sections, VPL nucleus has a peculiar appearance due to the presence of numerous bundles of myelinated fibres, which are going through it (Fig. 1A, B).

VPL nucleus in both adult rabbits and rats was composed of scattered neurons, or located in groups of 2–3 neurons. In adult rats this structure was composed of an homogeneous population of medium-sized, multipolar or oval neurons with the large nucleus distinguishable from the surrounding darker cytoplasm. The neuronal nuclei usually contained few darkly stained nucleoli (Fig. 2D, H). The proximal parts of dendrites were visible. The neuronal population of VPL nucleus in rabbits consisted of an heterogeneous population of large cells and medium-sized cells. The large cells were oval and round, whereas the medium-sized cells were oval. They had large light nuclei with 1–2 centrally located nucleoli and darkly stained cytoplasm. The proximal parts of the dendrites were hard to distinguish (Fig. 3D, H).

The developmental morphology of VPL nucleus in rats. On the first day after the birth of rats (age P0), the neurons of VPL nucleus showed different types of morphological shapes. The heterogeneous population of neurons consisted of fusiform cells intermingled with multipolar and oval cells. The small-size neurons were very densely packed. They also had big nuclei with few nucleoli, which were surrounded by quite a wide ring of a dark cytoplasm (Fig. 2A, E). Apoptotic bodies were rarely found in the ventrolateral portions of this structure.

VPL nucleus at age P7 was characterized by the most homogeneous appearance. Fusiform and oval neurons predominated, whereas the remaining neurons were multipolar. Cells were less densely packed in comparison with the previous age group. Considerable increase in their size was obvious. Also, the cytoplasm occupied a large part of the neuron (Fig. 2B, F). The proximal portions of the dendrites were often visible. The light nucleus was still clearly distinct and big; it contained few nucleoli. In the neighbourhood of the external medullary lamina, apoptotic bodies were observed (Fig. 4A, B).

The neuronal population of VPL nucleus at age P14 was almost exclusively composed of multipolar cells. Their size and staining properties seemed not to differ significantly from that of the previous age group although they were less densely packed. The primary and secondary dendrites were often well visible (Fig. 2C, G). Apoptotic bodies were not noticed. For the remaining age groups (P21, P45 and P180), the morphology of neurons did not undergo further significant morphological changes.

The developmental morphology of VPL nucleus in rabbits. At age P0, the population of VPL neurons had densely packed and elongated cells with fusiform or multipolar shape. Due to similar staining properties, their nuclei were not often distinct from the surrounding cytoplasm. The proximal parts of the dendrites were usually visible, especially on the poles of the fusiform cells (Fig. 3A, E). In the neighbourhood of the external medullary lamina, apoptotic bodies were rarely observed.

VPL nucleus at age P7 had cells characterized by different types of shapes although the multipolar neurons predominated. The remaining cells were oval and round. We observed a significant increase in the size of cells and a decrease in their packing density (Fig. 3B, F). The neuronal nuclei were still often hard to distinguish from the surrounding cytoplasm. The proximal portions of the primary dendrites were clearly visible. Some apoptotic bodies were observed near the external medullary lamina (Fig. 4C, D).

At the later age (P14), the neuronal populations of VPL nucleus consisted of two types of neurons: (1) large and round, and (2) small and oval neurons. The former predominated over the small and oval

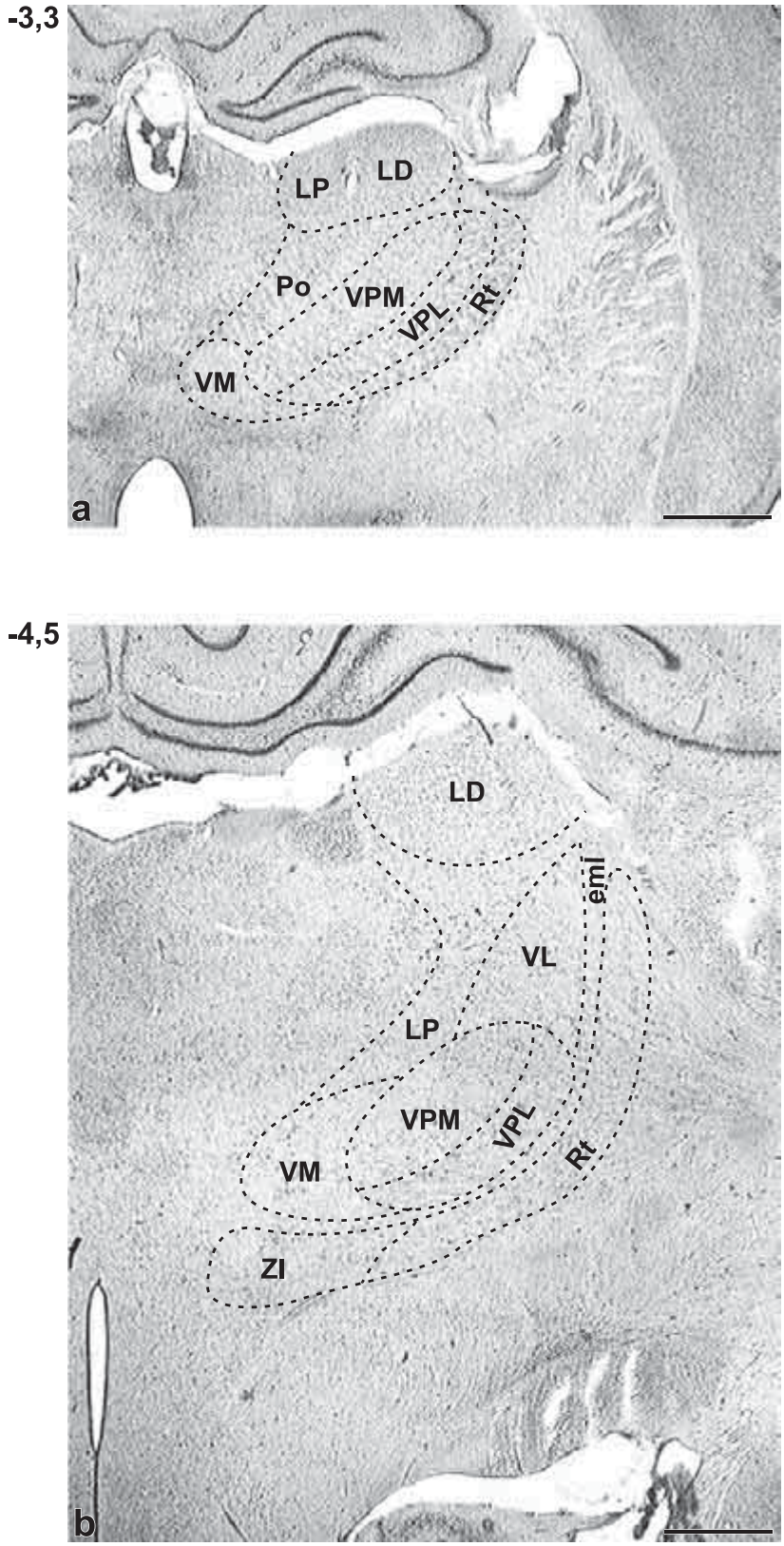

Figure 1. The selected coronal section through the middle part of the thalamus showing the general topography of the studied thalamic nuclei in the adult rat (**a**) and rabbit (**b**). Numbers in the upper left corners indicate the distance from bregma; eml — external medullary lamina; LD — lateral dorsal nucleus, LP — lateral posterior nucleus, Po — posterior nucleus, Rt — reticular nucleus, VL — ventrolateral nucleus, VM — ventromedial nucleus, VPL — ventroposterolateral nucleus, VPM — ventroposteromedial nucleus, ZI — zona incerta. Cresyl violet. Scale bar = 1 mm.

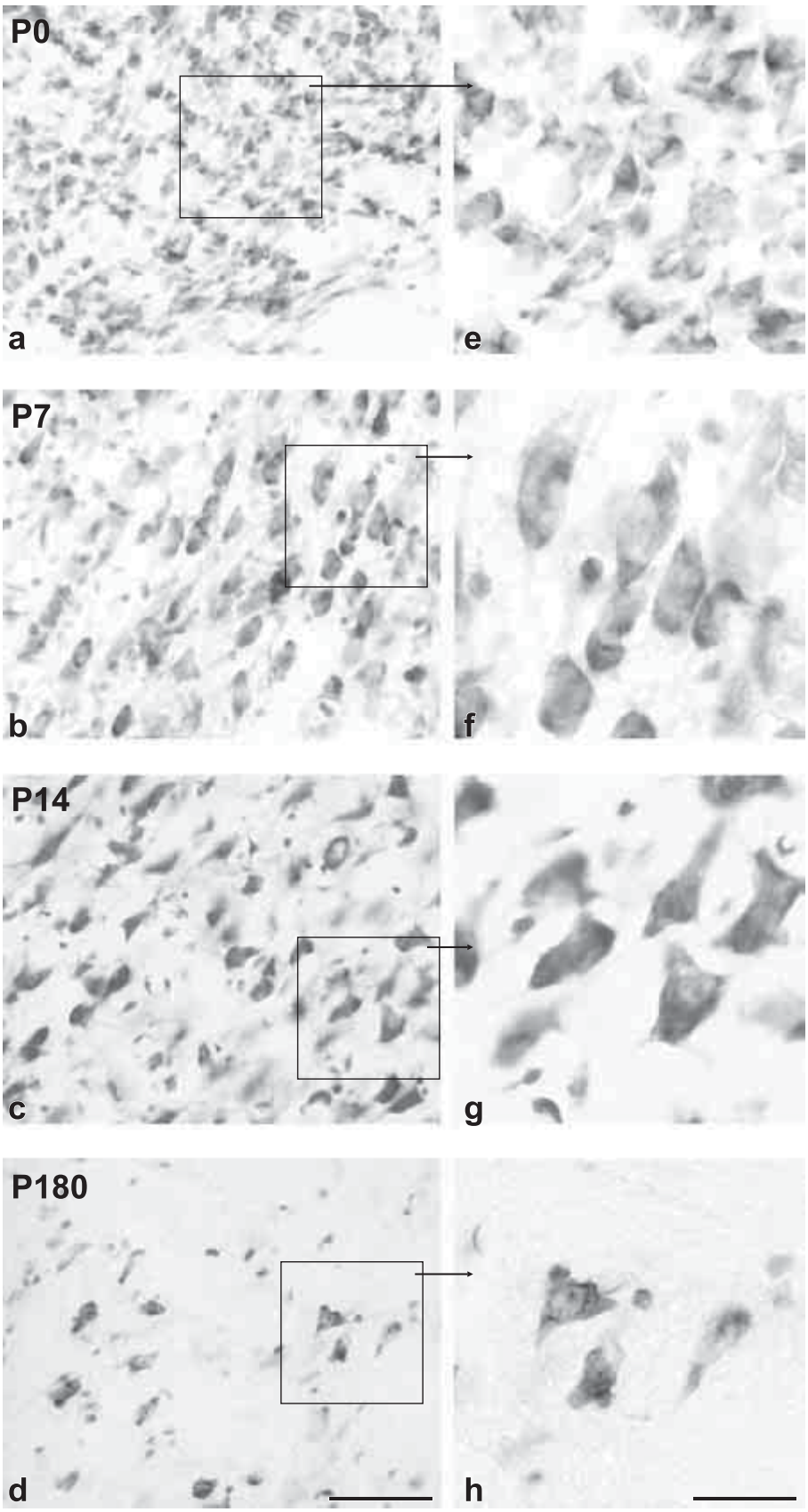

Figure 2. Postnatal changes in the distribution and morphology of neurons in the ventroposterolateral nucleus of the thalamus in the rat in selected age groups. Cresyl violet. Scale bars: a–d = 100 μm; e–h = 20 μm.

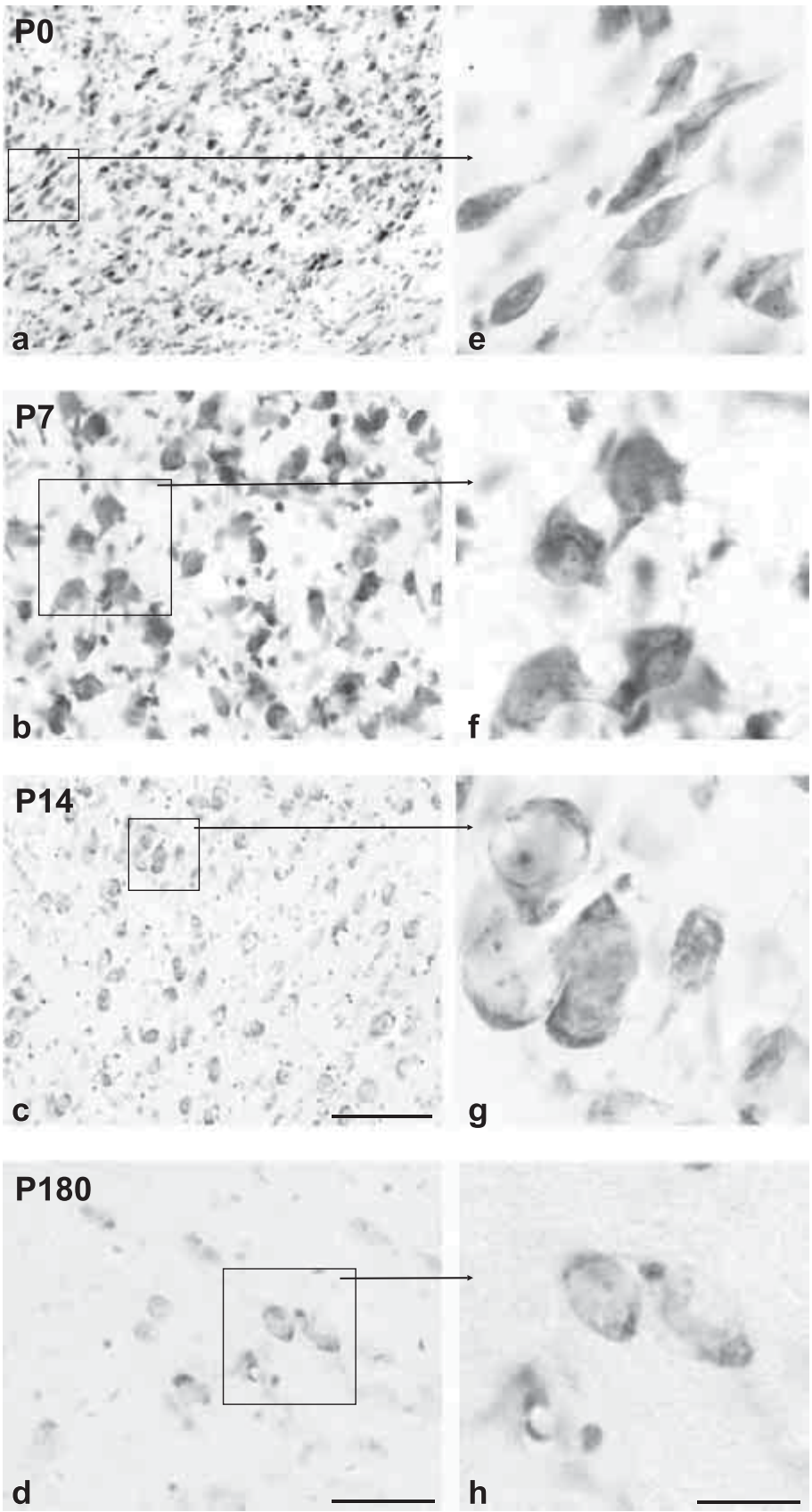

Figure 3. Postnatal changes in the distribution and morphology of neurons in the ventroposterolateral nucleus of the thalamus in the rabbits in selected age groups. Cresyl violet. Scale bars: a, c = 100 μm; b, d = 50 μm; e–h = 20 μm.

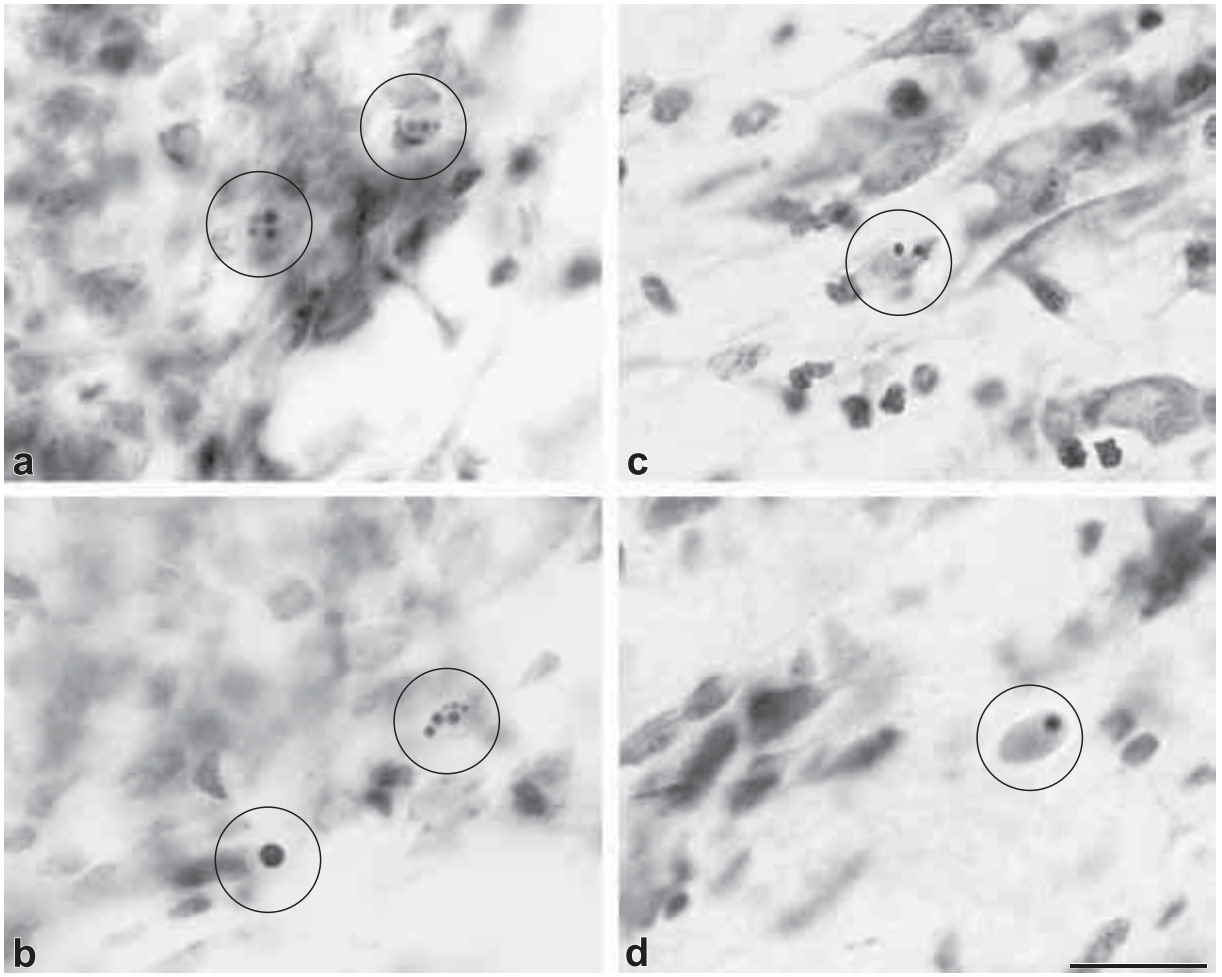

Figure 4. Coronal sections through the middle part of the ventroposterolateral nucleus of the thalamus during the first week of postnatal life in the rat (**a**, **b**) and rabbit (**c**, **d**). Encircled apoptotic bodies indicate that they are located near the external medullary lamina. Cresyl violet. Scale bar = 20 μm.

neurons. Besides their shape and size, these populations of neurons were also characterized by different staining properties. Large cells usually contained only a thin rim of cytoplasm and clearly distinguishable light nuclei. The remaining population was composed of darker cells with a larger amount of cytoplasm (Fig. 3C, G). The decrease in the neuronal density was observed in comparison with the previous age group.

The morphology of VPL neurons in rabbits of the remaining age groups (P21, P60, P120 and P180) was not significantly different from that observed at the preceding age (P14).

Quantitative analysis

The data describing stereological parameters of the cerebral hemisphere and VPL nucleus in rats and rabbits are given in Tables 1 and 2, respectively.

In rats the volume of the cerebral hemisphere underwent significant changes until the end of the third postnatal week. It increased by 2.6 times. The highest increase was observed during the first postnatal week of life (1.6 times). It did not change significantly from the third week of postnatal life; the increase in the volume was only about 10%.

In rabbits the volume of the cerebral hemisphere changed significantly by the end of the third week (by approximately 7.5 times). The highest increase in the volume occurred during the first week of life. It did not change significantly from the third week of postnatal life. Although the significant increase was only during the first week of postnatal life (1.4 times), the volume of VPL nucleus in rats increased correspondingly with changes in the hemispheric volume. The volume of VPL nucleus in rabbits increased gradually; significant changes occurred be-

Table 1. Mean values (±SD) of parameters of particular age groups in the rats

Age group	Volume of hemisphere [mm^3]	Volume of VPL [mm^3]	Ratio of the volume of VPL to the volume of hemisphere (%)	Numerical density of VPL neurons [mm^{-3}]	Total number of VPL neurons [1/1]
P 0	47 ± 5.7	0.18 ± 0.021	3.97 ± 0.33	133208 ± 12763	23577 ± 1298
P 7	76 ± 3.7	0.26 ± 0.027	3.51 ± 0.19	102294 ± 7318	26337 ± 2125
P 14	98 ± 1	0.32 ± 0.021	3.64 ± 0.6	56182 ± 4557	17367 ± 71
P 21	124 ± 8.4	0.34 ± 0.047	2.78 ± 0.24	52228 ± 3077	17338 ± 2620
P 45	131 ± 12.3	0.36 ± 0.07	2.79 ± 0.48	42390 ± 1970	14827 ± 2624
P 180	134 ± 8.3	0.40 ± 0.076	3.6 ± 0.34	34883 ± 4130	13352 ± 1082

Table 2. Mean values (±SD) of parameters of particular age groups in the rabbits

Age group	Volume of hemisphere [mm^3]	Volume of VPL [mm^3]	Ratio of the volume of VPL to the volume of hemisphere (%)	Numerical density of VPL neurons [mm^{-3}]	Total number of VPL neurons [1/1]
P 0	62 ± 17	0.17 ± 0.04	2.86 ± 0.16	162849 ± 36816	26760 ± 2380
P 7	196 ± 21	0.33 ± 0.08	1.71 ± 0.23	93747 ± 9073	30156 ± 4350
P 14	246 ± 24	0.44 ± 0.03	1.81 ± 0.05	45284 ± 4619	19993 ± 832
P 21	463 ± 74	0.62 ± 0.06	1.35 ± 0.2	27371 ± 5806	14909 ± 1342
P 60	500 ± 33	0.7 ± 0.1	1.4 ± 0.12	20448 ± 1613	14181 ± 1326
P 90	593 ± 96	0.74 ± 0.16	1.24 ± 0.1	21141 ± 2933	15195 ± 3060
P 120	550 ± 100	0.72 ± 0.11	1.33 ± 0.29	20962 ± 2148	15034 ± 3772
P 180	590 ± 98	0.85 ± 0.22	1.42 ± 0.19	15807 ± 1327	12997 ± 2952

tween longer time intervals (two weeks). During the first two weeks, VPL nucleus in rabbits increased by 2.6 times. At ages between P7 and P21, the increase in the volume increased by about 1.9 times. Similarly, the volume of VPL nucleus stabilized at age P21. The relative volume of VPL nucleus (ratio of VPL nucleus volume to the cerebral hemisphere volume) decreased significantly in both species during the first three weeks due to a larger increase in the volume of the cerebral hemisphere.

The changes in the numerical density of VPL nucleus in rats and rabbits proceeded in the similar manner. In both species, significant decreases in the numerical density during the first and the second week of postnatal life were observed. During the first week, the decrease was equal to 23% and 42% in rats and rabbits, respectively. But both species were characterized by similar decreases in the numerical density during the second week. From age P0 to P7, the total number of neurons in VPL nucleus showed no significant changes. A significant decrease in the total number of neurons was observed during the second week of postnatal life; no significant changes were noticed after this time.

DISCUSSION

VPL nucleus was clearly distinguished from the surrounding structures at age P0 in both rats and rabbits. According to the data reported by Altman and Bayer [6], VPL neurons in rats are generated between the 14th and 16th day of prenatal life. Then, during the next two days of migration, they get localized in the target region and axonogenesis takes place [11]. With reference to the duration of pregnancy (which is longer in rabbits by about one week), VPL neurons in rabbits appear relatively earlier (since E16; [36]) than in rats.

Taking into account the shape of neurons, the neuronal population of VPL nucleus at age P0 in rats was heterogeneous, whereas that that in rabbits was homogeneous. This remarkable difference in the morphology of neuronal population shapes at P0 was

possibly due to the shorter period of the prenatal development in rats.

The neurons of VPL nucleus in both species underwent significant qualitative changes during the first two weeks of postnatal life. During the first week of postnatal life of rabbits, the morphological changes in VPL neurons were similar to changes occurring in rats during the second week of postnatal life. The above mentioned changes concern the shape as well as the staining properties of perikarya and neuronal nuclei. The intense staining of neurons in rabbits and rats, which was probably caused by the high level of basophilic compounds, indicates transcriptional and translational activities that occurred postnatally in the former species (at ages P0 and P7), whereas in the latter they occurred till at age P14. The morphological changes described above can also be related to the differences in the periods of the prenatal development.

The sizes of cells in both species increased considerably during the first week of postnatal life. Although there is no data with regard to this parameter, there are some studies on other developmental structures of either rats or rabbits. According to Zantua et al. [71], the volume of perikarya of VPM nucleus in mice increases significantly during the first two weeks of postnatal life. The number and length of the original primary dendrites does not change, but the number and length of the secondary dendrites increases. Similar data concern the maturation processes in rats. On the basis of studies on Golgi bodies, Schiebel et al. [61] showed that the neuropil in VPL nucleus reaches its mature appearance during the first two weeks of postnatal life. They observed an increase in the dendritic field, prolongation of the secondary dendrites, loss of the typical dendritic nodules and primitive spines and gradual occurring of specialized structures on the dendrite surface. On the other hand, the same authors showed that the dendritic organization of VPL nucleus in cats required two more weeks than that in rats to reach the morphology of an adult animal.

On the basis of studies on EM, De Biasi et al. [17] proved the achievement of well developed cellular organella by VP neurons was in the first two weeks of the life of rats. This process occurs at the time as the functional development. The expression of Thy-1 immunoglobulin [9], which is responsible for regulating the distribution of cell-surface signaling proteins related to synaptic signaling transmission, revealed considerable changes during the first week of postnatal life. This indicates that immature neuropil and neurons at age P0 are probably able to function after the first week of animal life.

While comparing the morphology of VPL nucleus in adult animals of various species, we should respect the remarkable differences in cytoarchitectonics and intrinsic organization of the nucleus. Numerous studies [8, 28, 41, 55] prove that VPL nucleus in rats is composed of homogeneous population of the relay neurons. However, some authors found interneurons; but their number was very small (0.3%) [29]. On the contrary, an heterogeneous neuronal population characterizes VPL nucleus in cats. In this species two types of thalamocortical relay neurons and probably interneurons can be observed [42, 63, 69].

Our observations (the neuronal population of VPL nucleus in adult rats) are in agreement with the data mentioned above. But homogeneity of the neuronal population in rats becomes characteristic at age P14. The morphological types of neurons in adult rabbits are similar to those described in cats. This neuronal heterogeneity also appears at age P14 (like in rats). Although the intrinsic connections in rabbits are far more complicated (probably requiring a longer time for forming and maturation) than in rats, the duration of these processes is similar in both species. The above described multidirectional process of maturation (involving general morphology, ultrastructure and biochemistry) seems to be related to the functional development, which is remarkably influenced by the external environment conditions after the birth of the species.

On the basis of our data, the volume of VPL nucleus in rats increased significantly during the first and second weeks of postnatal life. In rabbits, statistically significant changes in volume took place after a longer time; they stabilized during the second month. Following the initial decrease in the relative volume of VPL nucleus (the volume of VPL nucleus to the volume of the cerebral hemisphere), the ratio stabilized after the third week of postnatal life of both species. The observed decrease in the ratio during the first three weeks of postnatal life indicates that the increase in the volume of the cerebral hemisphere was faster than that of VPL nucleus.

No data describing the developmental changes of VPL nucleus volume were found in the literature. However, some data on the volumetric changes of VPM nucleus in mice were reported by Zantua et al. [71]. According to their observations, the volume of VPM nucleus increases by three times. Since VPM and VPL nuclei constitute the ontogenetically inseparable complex, we may predict that the dynamism

of their development is comparable. In our study, the volume of VPL nucleus in rats increased by 2.2 times from P0 to adulthood, whereas in rabbits this parameter increased by five times. Zantua et al. [71] reported that the increase in VPM nucleus volume in mice is by about three times. Hence, their results are in agreement with our results for VPL nucleus in rats. Briefly, the volumetric changes in both parts of VPL nucleus in rats and mice are analogous. Small discrepancies may result from the methodological methods used, and in rabbits discrepancies may also be connected with the brain volume (which is bigger than that of either rats or mice). During development from P0 to P180, the volume of the brain of rats increased about three times, whereas the volume of the brain of rabbits increased by approximately ten times in the same period. In summary, the data show that the increase in the volume of VPL nucleus (as a phylogenetically older structure) progresses proportionally to the increase in the volume of the brain.

The increase in volume was observed in the development of various structures in the central nervous system of rodents, e.g., dorsal lateral geniculate nucleus [1, 33], principal sensory nucleus of trigeminal nerve [2], dentate gyrus [10, 12], and claustrum [45, 68].

According to our study, the density of VPL neurons decreased significantly in both species during the first and second week of postnatal life (23% and 45% in the rat, 42% and 52% in the rabbit, respectively). These changes were possibly due to the increase in the neuropil volume and, to a less degree, to the increase in the cell volume. According to De Biasi et al. [17], the neuropil of VP lacks the myelinated fibers on the day of birth; only a few synaptic terminals on VP neurons are present in the wide extracellular space. During the second week of postnatal life, progressing maturational changes resulted in the development of myelinated fibers and the appearance of numerous synaptic endings, which resembled those that exist in adult animals. Between ages P14 and P18, the development of spino- and trigeminothalamic terminals was almost finished and, at the end of the third week of postnatal life, the neuropil appearance and synaptic organization of VP were not different from that observed in adult animals. Similar observations were reported by Mattews et al. [47, 48] in their studies concerning synaptogenesis in VP. The three-fold increase in the number of synaptic contacts and their differentiation that enhanced the morphological complexity occurred in the second week. Moreover, the ontogenetical studies by Schiebel et al. [61] showed that there are differences in the course and the time of extrinsic maturation (originating from both periphery and cortex) and the intrinsic systems within the thalamic somatosensory area. The extrinsic fibres provide the frame within which the processes of maturity can occur. According to Schiebel et al. [61], this process takes place postnatally. But depending on the species it occurs at different periods of time.

In both species, the total number of neurons did not undergo significant changes till the 7th day of postnatal life. Possibly, this can be explained by the gradual increase in the volume of the nucleus. The volume increased correspondingly with the decrease in the neuronal density. Both of these processes were mainly caused by changes in the neuropil volume. A statistically significant decrease in the total number of neurons was observed in both species on 14th day of postnatal life. Then, it did not seem to change.

The 34% loss of VPL neurons was probably caused by apoptosis taking place in the normally developing nervous system. This is a common phenomenon that occurs in differentiating and maturing nervous systems in mammals controlled by numerous genetic [13, 46] and epigenetic [3, 25, 60] mechanisms. The former is regarded as a structural process, whereas the latter as a functional one [25, 54, 59]. Structural apoptosis concerns neurogenesis, whereas the functional one is closely related with synaptogenesis, especially with the processes of forming and establishment of neuronal circuitry.

Apoptosis is described for various structures of the central nervous system (spinal cord [54], trigeminal mesencephalic nucleus [4], colliculus superior of lamina tecti [7, 16, 23], retina [57, 58], some thalamic nuclei [33, 62], basal nuclei [35, 44], claustrum [45, 68] and cerebral cortex [20–22, 24, 32, 40, 62]). Its extent is different and depends not only on a structure, but also on a specimen. In some structures only few apoptotic cells are found (thalamic nuclei in the rat [62]), whereas in others as much as 50% of the neuronal population are apoptotic cells (lumbo-sacral motoneurons of the chick embryo [54]). Even in the same species the extent of apoptosis is different among structures [62].

Natural death of cells occurring during the development of the rat VP nucleus is described by Waite et al. [65]. According to their data, apoptotic cells are visible from E19 till P8, however their highest

number occurs at birth. The estimation of the total number of VP neurons indicates the postnatal loss of about 27% of neurons. Our estimation of the neuronal loss of VPL nucleus is in agreement with data given above. However, different results were reported by Spreafico et al. [62]. Although their results were only semi quantitative, they showed that during the development of the rat thalamus (from E16 till adulthood), only few single apoptotic cells can be found. The highest number of apoptotic cells was found after about 5days of postnatal life. The other qualitative data were reported by Leamley et al. [39], who studied the phenomenon of apoptosis in marsupials. Although the authors observed a peak of apoptosis in VPM nucleus from P76 till P108, this period of marsupials' life (taking into account the appearance of barreloids) is related to perinatal or early postnatal period of higher mammals because the barreloids were clearly visible in the rat VPL nucleus at the end of the first postnatal week.

Even after considering the neurogenetic gradient [6] and the characteristic spatial distribution of apoptotic bodies (observed by us in the ventro-lateral portions of VPL nucleus), the confirmation of the presence of the partial structural component in apoptotic process can be excluded due to its early (prenatal) appearance.

Taking the above data into account, it appears that the sensory system is "morphologically ready" to transmit impulses on the day of birth of the animal. In spite of substantial differences between interspecies, the present results indicate that the intrinsic organization of VPL nucleus and the appreciable similarity during the processes of development and maturation of VPL nucleus in rats and rabbits exist. The total number of VPL neurons in both species studied stabilized in the third week of postnatal life. The gradual increase in the volume of the nucleus corresponded with the decrease in the neuronal density in the first week of postnatal life. Both of these processes were mainly caused by changes in the neuropil volume because the total number of neurons did not change significantly until P7. The second week was considered as the critical period for the achievement of mature morphology of the nucleus. During this time, both density and total number of neurons decreased. With appearance of striking impulses from the external environment, the complicated sensory system undergoes remodeling resulting in the physiological cell death and development of neuropil.

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REFERENCES

1. Ahmad A, Spear PD (1993) Effects of aging on the size, density, and number of rhesus monkey lateral geniculate neurons. J Comp Neurol, 334: 631–643.
2. Al-Ghoul WM, Miller MW (1993) Development of the principal sensory nucleus of the trigeminal nerve of the rat and evidence for a transient synaptic field in the trigeminal sensory tract. J Comp Neurol, 330: 476–490.
3. Allen RT, Hunter WJ, III, Agrawal DK (1997) Morphological and biochemical characterization and analysis of apoptosis. J Pharmacol Toxicol Meth, 37: 215–228.
4. Alley KE (1974) Morphogenesis of the trigeminal mesencephalic nucleus in the hamster: cytogenesis and neuronal death. J Embryol Exp Morphol, 31: 99–121.
5. Altman J, Bayer SA (1988) Development of the rat thalamus: I. Mosaic organization of the thalamic neuroepithelium. J Comp Neurol, 275: 346–377.
6. Altman J, Bayer SA (1989) Development of the rat thalamus: IV. The Intermediate lobule of the thalamic neuroepithelium, and the time and site of origin and settling pattern of neurons of the ventral nuclear complex. J Comp Neurol, 284: 534–566.
7. Arees EA, Astrom K-E (1977) Cell death in the optic tectum of the developing rat. Anat Embryol (Berl), 151: 29–34.
8. Barbaresi P, Spreafico R, Frassoni C, Rustioni A (1986) GABAergic neurons are present in the dorsal column nuclei but not in the ventroposterior complex of rats. Brain Res, 382: 305–326.
9. Barlow JZ, Huntley GW (2000) Developmentally regulated expression of Thy-1 in structures of the mouse sensory-motor system. J Comp Neurol, 421: 215–233.
10. Bayer SA (1982) Changes in the total number of dentate granule cells in juvenile and adult rats: A correlated volumetric and 3H-Thymidine autoradiographic study. Exp Brain Res, 46: 315–323.
11. Bayer SA, Altman J, Russo RJ, Zhang X (1993) Timetables of neurogenesis in the human brain based on experimentally determined patterns in the rat. Neurotoxicology, 14: 83–144.
12. Bayer SA, Yackel JW, Puri PP (1982) Neurons in the rat dentate gyrus granula layer substantially increase during juvenile and adult life. Science, 216: 890–892.
13. Bright J, Khar A (1994) Apoptosis: Programmed cell death in health and disease. Biosci Rep, 14: 67–82.
14. Chmielowska J, Carvell GE, Simons DJ (1989) Spatial organization of thalamocortical and corticothalamic projection systems in the rat SmI barrel cortex. J Comp Neurol, 285: 325–338.
15. Clancy B, Darlington RB, Finlay BL (2001) Translating developmental time across mammalian species. Neuroscience, 105: 7–17.

16. Cunningham TJ (1982) Naturally-occurring neuron death and its regulation by developing neural pathways. Intl Rev Cytol, 74: 163–186.
17. De Biasi S, Amadeo A, Arcelli P, Frassoni C, Meroni A, Spreafico R (1996) Ultrastructural characterization of the postnatal development of the thalamic ventrobasal and reticular nuclei in the rat. Anat Embryol, 193: 341–353.
18. Erzurumlu RS, Jhaveri S (1990) Thalamic axons confer a blueprint of the sensory periphery onto the developing rat somatosensory cortex. Dev Brain Res, 56: 229–234.
19. Erzurumlu RS, Kind PC (2001) Neural activity: sculptor of «barrels» in the neocortex. Trends Neurosci, 24: 589–595.
20. Ferrer I, Serrano T, Soriano E (1990) Naturally occurring cell death in the subicular complex and hippocampus in the rat during development. Neurosci Res, 8: 60–66.
21. Ferrer I, Soriano E, Del Río JA, Alcántara S, Auladell C (1992) Cell death and removal in the cerebral cortex during development. Prog Neurobiol, 39: 1–43.
22. Ferrer I, Tortosa A, Blanco R, Martín F, Serrano T, Planas A, Macaya A (1994) Naturally occurring cell death in the developing cerebral cortex of the rat. Evidence of apoptosis-associated internucleosomal DNA fragmentation. Neurosci Lett, 182: 77–79.
23. Finlay B, Berg AT, Sengelaub DR (1982) Cell death in mammalian visual systems during normal development. II. Superior colliculus. J Comp Neurol, 204: 318–324.
24. Finlay B, Slattery M (1983) Local differences in the amount of early cell death in neocortex predict local specializations. Science, 219: 1339–1351.
25. Gordon N (1995) Apoptosis (programmed cell death) and other reasons for elimination of neurons and axons. Brain Dev, 17: 73–77.
26. Haidarliu S, Ahissar E (2001) Size gradients of barreloids in the rat thalamus. J Comp Neurol, 429: 372–387.
27. Haidarliu S, Sosnik R, Ahissar E (1999) Simultaneous multi-site recordings and iontophoretic drug and dye applications along the trigeminal system of anesthetized rats. J Neurosci Meth, 94: 27–40.
28. Harris RM (1986) Morphology of physiologically identified thalamocortical relay neurons in the rat ventrobasal thalamus. J Comp Neurol, 251: 491–505.
29. Harris RM, Hendrickson AE (1987) Local circuit neurons in the rat ventrobasal thalamus — GABA immunocytochemical study. Neuroscience, 21: 229–236.
30. Havton LA, Ohara PT (1993) Quantitative analyses of intracellularly characterized and labeled thalamocortical projection neurons in the ventrobasal complex of primates. J Comp Neurol, 336: 135–150.
31. Herrmann K, Antonini A, Shatz CJ (1994) Ultrastructural evidence for synaptic interactions between thalamocortical axons and subplate neurons. Eur J Neurosci, 6: 1729–1742.
32. Heumann D, Leuba G, Rabinowicz T (1978) Postnatal development of the mouse cerebral neocortex. IV. Evolution of the total cortical volume of the population of neurons and glial cells. J Hirnforsch, 19: 385–393.
33. Heumann D, Rabinowicz Th (1980) Postnatal development of the dorsal lateral geniculate nucleus in the normal and enucleated albino mouse. Exp Brain Res 38: 75–85.
34. Howard CV, Reed MG (1998) Unbiased stereology. Three-dimensional measurement in microscopy. BIOS Scientific Publishers.
35. Itoh K, Suzuki K, Bise K, Itoh H, Mehraein P, Weis S (2001) Apoptosis in the basal ganglia of the developing human nervous system. Acta Neuropathol, 101: 92–100.
36. Jones EG (1985) Development of the thalamus. In: Jones EG (ed.). The Thalamus. 1 edn. Plenum Press, New York, pp. 260–319.
37. Kostovic I, Rakic P (1990) Developmental history of the transient subplate zone in the visual and somatosensory cortex of the macaque monkey and human brain. J Comp Neurol, 297: 441–470.
38. Land PW, Buffer SA, Jr, Yaskosky JD (1995) Barreloids in adult rat thalamus: three-dimensional architecture and relationship to somatosensory cortical barrels. J Comp Neurol, 355: 573–588.
39. Leamey CA, Marotte LR, Waite PME (1996) Time course of development of the wallaby trigeminal pathway. II. Brainstem to thalamus and the emergence of cellular aggregations. J Comp Neurol, 364: 494–514.
40. Leuba G, Rabinowicz T (1979) Long term effects of postnatal under nutrition and maternal malnutrition on mouse cerebral cortex. I. Cellular densities, cortical volume and total number of cells. Exp Brain Res, 37: 283–298.
41. Liu XB (1997) Subcellular distribution of AMPA and NMDA receptor subunit immunoreactivity in ventral posterior and reticular nuclei of rat and cat thalamus. J Comp Neurol, 388: 587–602.
42. Loopuijt LD, Villablanca JR, Hovda DA (1995) Morphological changes in the thalamus and neocortex of the cat brain after a restricted unilateral fetal neocortical lesion. Dev Brain Res, 85: 259–272.
43. Ma PM (1993) Barrelettes — architectonic vibrissal representations in the brainstem trigeminal complex of the mouse. II. Normal post-natal development. J Comp Neurol, 327: 376–397.
44. Maciejewska B, Lipowska M, Kowianski P, Pytel-Domaradzka B, Morys J (1998) Postnatal development of the rat striatum — a study using *in situ* DNA end labelling techniques. Acta Neurobiol Exp, 58: 23–28.
45. Maciejewska B, Morys J, Berdel B, Dziewiatkowski J, Narkiewicz O (1997) The development of the rat claustrum — a study using morphometric and *in situ* DNA end labelling techniques. Eur J Anat, 1: 137–143.
46. Maslinska D, Muzylak M (1995) Apoptosis, programmed cell death in the central nervous system. Folia Neuropathol, 33: 69–76.
47. Matthews MA, Faciane CL (1977) Electron microscopy of the development of synaptic patterns in the ventrobasal complex of the rat. Brain Res, 135: 197–215.
48. Matthews MA, Narayanan CH, Narayanan Y, Onge MF (1977) Neuronal maturation and synaptogenesis in the rat ventrobasal complex: alignment with developmental changes in rate and severity of axon reaction. J Comp Neurol, 173: 745–772.
49. Meng XW, Ohara PT, Ralston HJ, III (1996) Nitric oxide synthase immunoreactivity distinguishes a sub-population of GABA-immunoreactive neurons in the ventrobasal complex of the cat. Brain Res, 728: 111–115.

50. Miller B, Chou L, Finlay BL (1993) The early development of thalamocortical and corticothalamic projections. J Comp Neurol, 335: 16–41.
51. Molnár Z (1998) Development of thalamocortical connections. Springer-Verlag.
52. Molnár Z, Adams R, Blakemore C (1998) Mechanisms underlying the early establishment of thalamocortical connections in the rat. J Neurosci, 18: 5723––5745.
53. Molnár Z, Cordery P (1999) Connections between cells of the internal capsule, thalamus, and cerebral cortex in embryonic rat. J Comp Neurol, 413: 1–25.
54. Oppenheim RW (1985) Naturally occurring cell death during neural development. Trends Neurosci, 11: 487–493.
55. Peschanski M, Lee CL, Ralston HJ, III (1984) The structural organization of the ventrobasal complex of the rat as revealed by the analysis of physiologically characterized neurons injected intracellularly with horseradish peroxidase. Brain Res, 297: 63–74.
56. Price JL (1995) Thalamus. In: Paxinos G (ed.). The rat nervous system. 2nd ed. Academic Press, San Diego, pp. 629–648
57. Provis JM (1987) Patterns of cell death in the ganglion cell layer of the human fetal retina. J Comp Neurol, 259: 237–246.
58. Provis JM, Van Driel D, Billson FA, Russell P (1985) Development of the human retina: Patterns of cell distribution and redistribution in the ganglion cell layer. J Comp Neurol, 233: 429–451.
59. Rakic S, Zecevic N (2000) Programmed cell death in the developing human telencephalon. Eur J Neurosci, 12: 2721–2734.
60. Rose CD, Henneberry RC (1993) Mechanisms of programmed cell death and their implications for the brain. Neurodegeneration, 2: 287–297.
61. Scheibel ME, Davies TL, Scheibel AB (1976) Ontogenetic development of somatosensory thalamus. I. Morphogenesis. Exp Neurol, 51: 392–406.
62. Spreafico R, Frassoni C, Arcelli P, Selvaggio M, De Biasi S (1995) In situ labeling of apoptotic cell death in the cerebral cortex and thalamus of rats during development. J Comp Neurol, 363: 281–295.
63. Spreafico R, Schmechel DE, Ellis LC, Jr, Rustioni A (1983) Cortical relay neurons and interneurons in the N. ventralis posterolateralis of cats: a horseradish peroxidase, electron-microscopic, Golgi and immunocytochemical study. Neuroscience, 9: 491–509.
64. Sugitani M, Yano J, Sugai T, Ooyama H (1990) Somatotopic organization and columnar structure of vibrissae representation in the rat ventrobasal complex. Exp Brain Res, 81: 346–352.
65. Waite PME, Lixin L, Ashwell KWS (1992) Development and lesion induced cell death in the rat ventrobasal complex. Neuroreport, 3: 485–488.
66. West MJ, Slomianka L, Gundersen HJG (1991) Unbiased stereological estimation of the total number of neurons in the subdivisions of the rat hippocampus using the optical fractionator. Anat Rec, 231: 482–497.
67. Wilson P, Kitchener PD, Snow PJ (1999) Cutaneous receptive field organization in the ventral posterior nucleus of the thalamus in the common marmoset. J Neurophysiol, 82: 1865–1875.
68. Wojcik S, Dziewiatkowski J, Kowianski P, Ludkiewicz B, Jagalska-Majewska H, Morys J (2002) Qualitative and quantitative study of the postnatal development of the rabbit insular claustrum. Int J Dev Neurosci, 20: 113–123.
69. Yen CT, Conley M, Jones EG (1985) Morphological and functional types of neurons in cat ventral posterior thalamic nucleus. J Neurosci, 5: 1316–1338.
70. Yuasa S, Kitoh J, Kawamura K (1994) Interactions between growing thalamocortical afferent axons and the neocortical primordium in normal and reeler mutant mice. Anat Embryol, 190: 137–154.
71. Zantua JB, Wasserstrom SP, Arends JJ, Jacquin MF, Woolsey TA (1996) Postnatal development of mouse "whisker" thalamus: ventroposterior medial nucleus (VPM), barreloids, and their thalamocortical relay neurons. Somatosens Mot Res, 13: 307–322.