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Postnatal maturation of the vascularisation of the suprasylvian gyrus of the cat

C. BEN HAMIDA*, J. C. BISCONTE† AND S. MARGULES†

**Laboratoire de Neuropathologie, Centre de Neurologie–Hôpital La Rabta, Tunis, Tunisie and † Laboratoire de Microscopie Quantitative, UER Biomédicale, 74 rue Marcel Cachin, 93 012 Bobigny, France*

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INTRODUCTION

In the course of a general study of the postnatal development of the cat suprasylvian gyrus, special attention has been paid to changes in its vascularisation during maturation. Among numerous earlier studies devoted to the maturation of the cerebral cortex, those concerned with its vascularisation (Dunning & Wolff, 1937; Caley & Maxwell, 1970; Bär & Wolff, 1973; Conradi, Eins & Wolff, 1979) have sought to establish whether or not this feature might be considered as an indicator of maturation.

The present study attempts to determine whether vascularisation could be a functional indicator, that is, one related closely to cell formation, to synaptogenesis and to myelination, as well as an indicator of structure. For example, it might be related to cytoarchitectonics, or be related to differences or similarities in the superficial and deep parts of the cortex.

The study of different vascular parameters, particularly the orientation of the vessels, is made possible by automatic image analysis, and may provide answers to some of the questions raised. Therefore, in this investigation, an evaluation has been made of mitoses, vascular densities and diameters, the ratio of vascular surface area to area of cerebral tissue (vascularisation coefficient), and the preferential orientations of vessels in the developing cat cerebral cortex.

MATERIAL AND METHODS

Two experimental series of cats were anaesthetised by an intraperitoneal injection of Nembutal (0·04 ml/10 g body weight) before tissue fixation.

Mitosis, vascular density and diameter

Seventeen kittens with postnatal ages of 2 hours to 42 days, and three adult cats, were given aortic perfusions of 2·5% glutaraldehyde in a phosphate buffer (Na_2HPO_4, KH_2PO_4) to which paraformaldehyde was added in various concentrations to give osmolarities of from 300 mosM (younger animals) to 600 mosM (older animals). The fixative was placed in a flask 1·2–1·5 metres above the animal, hence the pressure used, during perfusion, was approximately 150 cm water. This made possible fixation of cytological structures, and of vessels in particular, without distension or shrinkage artefacts. The same material was used in another study using the electron

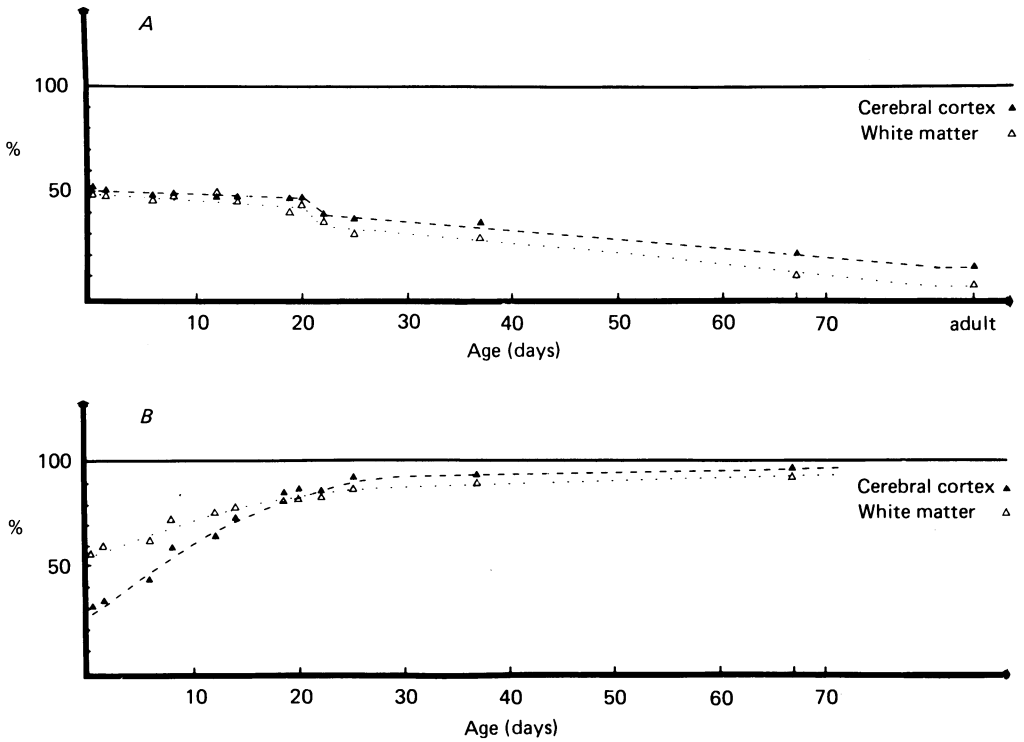

Fig. 1. Correction coefficients. (*A*) Shrinkage coefficient of brain sections, expressing shrinkage due to Pickworth technique as compared with Nissl sections for each animal. (*B*) Growth coefficient measured on frozen sections by comparing kitten brain sections with equivalent adult brain sections, expressed as percentage of adult value.

microscope. Thus the quality of fixation and perfusion was the result of the combined effects of osmolarity and pressure of the perfusing fluid.

Samples of cerebral cortex and of the underlying white matter were removed from the apex of the suprasylvian gyrus. They were post-fixed with osmium tetroxide (2 % in Palade's buffer) and embedded in Epon 812. The remainder of the brain was embedded in celloidin.

Using sections 1 μm thick, blood vessels were counted to obtain the vascular density per mm^2. If the objects had been spherical, the three dimensional nature of the sections would have made it necessary to use mathematical formulae to evaluate the real density of the objects present in the section. Since the objects studied were long tubes, the application of such formulae was considered to be unnecessary. These values were graphed. Shrinkage during tissue preparation was slight ($< 5\%$): the measured surface was considered to correspond to the actual surface.

These sections were then studied under a projection microscope. The smallest inner diameter – corresponding to the real diameter of an obliquely cut tube – of each of the 6134 vessels counted was measured. For each age group the results for vascular diameter of a given value were expressed in terms of the percentage of the total vessels measured.

The growth of the part of the brain under consideration as a function of age (growth coefficient, Fig. 1) was obtained by expressing the immature value as a percentage of the adult value. Thus a cerebral hemisphere at birth is smaller than an

adult hemisphere, and the surface area of a section of the brain at birth is potentially equivalent to, although smaller than, the surface area of the corresponding section in the adult brain. The expansion of the brain differs with the region of the brain. To estimate it in the region studied – the cingulate, the suprasplenial, the lateral, and the suprasylvian gyrus – 10 projections of sections from each brain were drawn on tracing paper of known weight. The areas of cerebral cortex and white matter were cut out and weighed, and for each brain the result was expressed as a percentage of the reference adult weight. Thus, at birth, the surface area of the cerebral cortex was only 30% of the equivalent surface area in the adult. For each brain, the growth coefficient was used to convert the surface measured into the equivalent adult surface, so as to obtain a corrected value of the vascular density. The values measured were compared before and after calculation using the growth coefficient. The application of this correction factor was particularly necessary to evaluate any density, since density was the element of analysis which was most affected by the spatial expansion of the cerebral tissue during maturation.

Endothelial mitoses were observed and counted in the celloidin-embedded median suprasylvian gyrus contralateral to that embedded in Epon. Shrinkage of tissues due to embedding in celloidin was identical, $\pm 5\%$, to that seen after Pickworth (1934) staining: the same correction was made in both cases (see below). The values obtained for mitoses/mm^2, as measured on sections 20 μm thick, were converted using the growth coefficient, and the results were graphed.

Vascular coefficient and orientation

Samples were taken from the brains of fifteen kittens aged between birth and 67 days old, and of one adult cat, and were fixed in hypertonic formol saline. Blocks were frozen, and sectioned frontally at a thickness of 200 μm. The sections were stained using the Pickworth (1934) technique, which localises the pseudo-peroxidase reaction of haemoglobin, using sodium nitroprusside and benzidine. The reaction was revealed with hydrogen peroxide. Section shrinkage with this technique was directly proportional to the immaturity of the brain (up to 50% at birth). The correction curve (shrinkage coefficient, Fig. 1) was drawn by comparing frozen sections (20 μm thick) mounted on slides, and stained (10% Unna blue differentiated with Gothard solution) to show Nissl bodies, with neighbouring sections treated using the Pickworth technique.

The sections obtained were analysed with a Leitz Texture Analysis System. This system comprises:

(i) a microscope (Orthoplan) equipped with a motorised stage (x and y axes) and with a revolving superstage;
(ii) a television camera (Plumbicon);
(iii) a densitometric module, making it possible to determine the threshold of the video signal;
(iv) an interactive module, making possible manual image changes such as erosion, dilation, subtraction, etc. and memorisation of the programme suited to the particular problem, which are then executed automatically by the system;
(v) an electronic cabinet containing the circuitry necessary for the execution of the different instructions.

Each section was positioned under the microscope in such a way that the horizontal

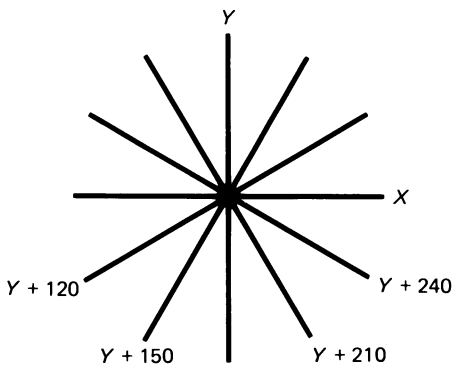

Fig. 2. 'Flower' diagram of electronic directions in the Leitz texture analysis system. The proportion of oriented vessels on each corresponding axis is measured and represented as a percentage of the sum of the values plotted on all the axes. See text for details and Fig. 7 for application.

spider line, the 'x' marker line, was tangential to the surface of the cerebral cortex. This made scanning comparable in the different sections. Analysis (Margules, Ben Hamida & Bisconte, 1981) was performed on contiguous fields, of side 175 μm, lying on a radial line extending from the cortical surface of the apex of the gyrus down to, and including, the white matter.

The programme was devised to study two parameters.

(*a*) The vascular coefficient, which was the ratio of the vascular surface area to the area of the cerebral tissue analysed. The measurements were used to draw three graphs for:

(i) the measured value;
(ii) the value corrected for shrinkage;
(iii) the value corrected both for shrinkage and growth.

(*b*) The preferential orientation of the vessels which was determined by their alignment in the three basic orthogonal projections of the texture analysis system (x, $y+120°$, $y+240°$), taken two by two. For example, a vessel was oriented in the direction 'x' if its projections on $y+120°$ and $y+240°$ were equal to each other and greater than its projection on 'y'. Using the same principle, it was possible to determine two other directions: $y+150°$, which was midway between y and $y+120°$, and $y+210°$, which was midway between y and $y+240°$ (Fig. 2).

The measurement of the number of particles oriented in any given direction was expressed as a percentage of the total number of particles analysed. Round or very slightly ovoid particles were eliminated from the analysis. This gave a 'flower' diagram (by analogy to the gypsum flower, or 'desert rose', which it resembles) with 'petals' in six directions, where x referred to a plan parallel to the plane tangential to the cerebral surface (tangential orientation), and y to the plane perpendicular to the cerebral surface (radiate orientation). The directions, $y+150°$ and $y+210°$, were spatially closer to y, while the directions $y+120°$ and $y+240°$ were spatially closer to x (Fig. 2).

This study has been illustrated by 'flower' diagrams of preferential orientation, either by taking the average 'flower' diagram of the cerebral cortex and comparing it with the white matter diagram, or by reproducing the 'flower' diagrams in detail for successive horizontal fields through the grey and white matter. Each horizontal field was obtained by taking the average of four fields located at the same depth.

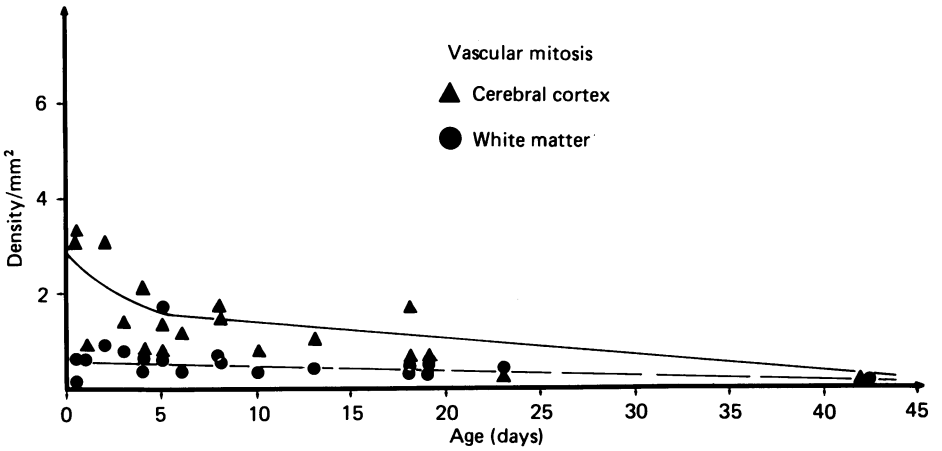

Fig. 3. Vascular mitoses in the cerebral cortex and the subjacent white matter.

The thickness of the cortical cell layers was measured on sections stained with Unna blue (1 % in distilled water), taken near the sections treated with the Pickworth reagent. After correction for shrinkage, the thickness of the layers in the Pickworth-stained sections was determined and represented (Fig. 7).

RESULTS

Vascular mitoses

Present from birth to 20 days of age, mitoses were almost absent beyond this age (Fig. 3). They were more numerous in the cerebral cortex than in the white matter.

Vascular density

The three graphs (Fig. 4) represent the vascular density (uncorrected), the vascular density compared to equivalent adult surfaces (corrected using the growth coefficient), and the vascular density by strata, comprising layer I, layers II–IV, and layers V and VI, corresponding to classical descriptions of stratification (infra- and supragranular) in the cerebral cortex. The vascular density increased with growth regardless of the mode of representation. Without correction, the increase was by a factor of from 1 to 2·5 between birth and adulthood, both in the cerebral cortex and the white matter, with scattered individual values. The slow growth of the first three weeks was followed by faster growth up to seven weeks. Growth slowed down greatly beyond seven weeks, when vascular density almost reached its adult value. After correction using the growth coefficient, the individual values were less scattered. Between birth and adulthood, the vascular density increased approximately sevenfold in the cerebral cortex and fivefold in the white matter, and was always greater in the cortex than in the white matter. In the adult, the vascular density of the white matter was approximately half that of the cortex. The three adult brains had a similar vascular density, a fact which supported its dependability as an indicator.

Analysis of the corrected vascular density of the cerebral cortex by strata showed that the vascular density of layer I increased slowly during the first week after birth. Thereafter it increased rapidly, and reached its adult value at five weeks. Layers II–IV had a vascular density which increased slowly during the first three weeks,

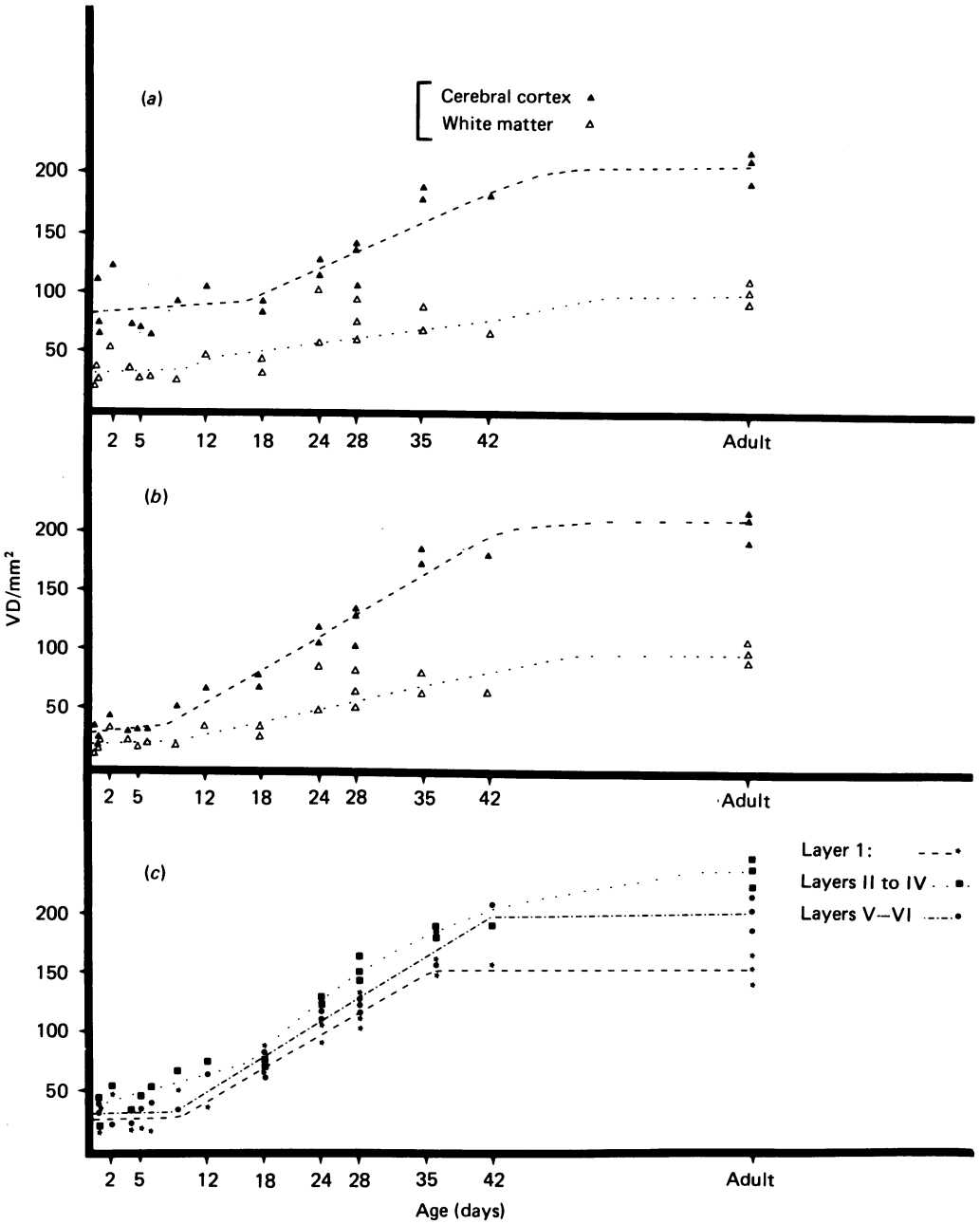

Fig. 4. Vascular density of the cerebral cortex and of the subjacent white matter: (*a*) uncorrected; (*b*) corrected using the growth coefficient; (*c*) cerebral cortex subdivided into layers, corrected using the growth coefficient. The shrinkage coefficient was not used, as tissue fixed for electron microscopy shrank only slightly.

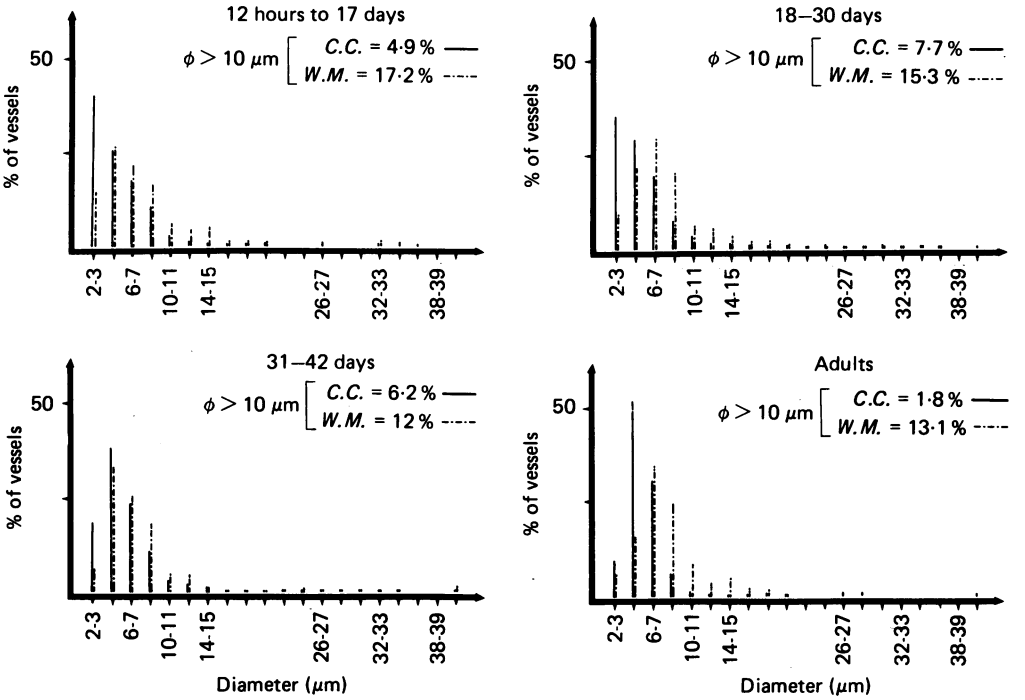

Fig. 5. Vessel diameters as a function of age. The animals whose diagrams have the same profile are grouped together; four groups of individuals were identified in correlation with other parameters. ϕ, diameter; *C.C.*, cerebral cortex; *W.M.*, white matter.

and then rapidly from the fourth to the sixth week. After the sixth week, the density continued to increase slowly to the adult value. In layers V and VI, the increase in vascular density was slow during the first week, but, commencing with the second week, was rapid until it reached its adult value at six weeks.

At birth the vascular densities were very similar in these three strata, although layers II, III and IV were slightly richer in vessels. During maturation, and in the adult, the vascular density remained slight in layer I. It was greatest in layers II, III and IV, and moderate in layers V and VI.

Vessel diameter

The diameters of the vessels also changed with age (Fig. 5). In the cerebral cortex up to 30 days after birth, the diameters most frequently encountered, 2–3 μm, were in the lowest part of the range whereas after 30 days it was the diameters from 4 to 7 μm which were most numerous. This was similar to the adult profile, which was characterised by an increase in the percentage of vessels of 4–7 μm and a decrease in that of vessels of 2–3 μm diameter.

The percentage of vessels greater than 10 μm in diameter increased up to 30 days, and then decreased; in adult specimens, it was below the percentage found in newborn kittens. This shift in the spectrum of large diameter vessels reflected the increasing capillary population, of 4–7 μm diameter.

At birth, larger diameter vessels (4–9 μm) were more frequent in the white matter than in the cortex. Commencing at birth, however, the percentage of vessels with diameters larger than 10 μm decreased, but remained higher in the white matter than

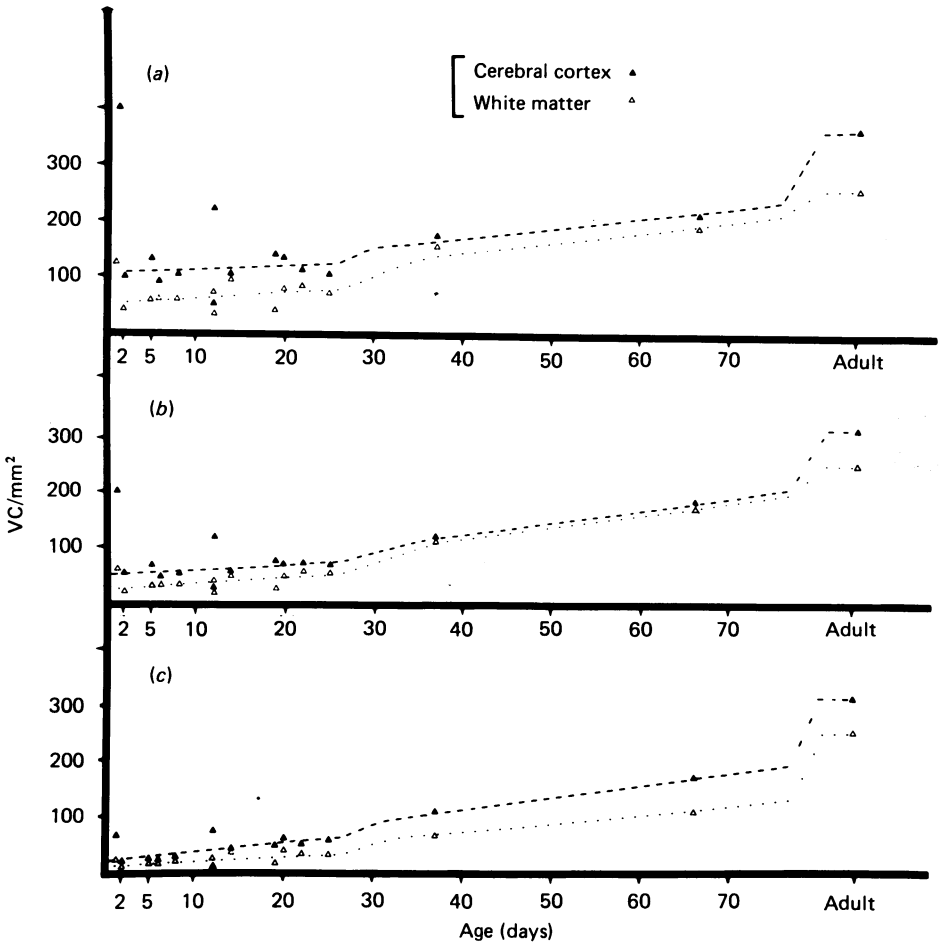

Fig. 6. Vascular coefficient of the cerebral cortex and of the subjacent white matter: (*a*) without correction; (*b*) corrected for shrinkage; (*c*) with the two coefficients of correction.

in the cortex. As in the cerebral cortex, the number of large diameter vessels in the white matter decreased as a function of age.

Vascularisation coefficient

Without correction, the individual values were very scattered (Fig. 6). With the exception of one animal which had a high vascular coefficient at birth (due to a predominance of large vessels), the vascular coefficient increased between the age of two days and adulthood by a factor of 3·5. The increase was slow, reaching only three fifths of its adult value at 67 days. After correction using the shrinkage coefficient, the vascular coefficients of the cerebral cortex and of the white matter, which were greater at birth than at two days, still increased slowly and reached only three fifths of the adult value at 67 days.

The adult specimen had a vascular coefficient six times greater than that of the animal two days old, but the vascular coefficient of the white matter remained 20% lower than that of the cortex.

After double correction for both shrinkage and growth, the vascular coefficients of

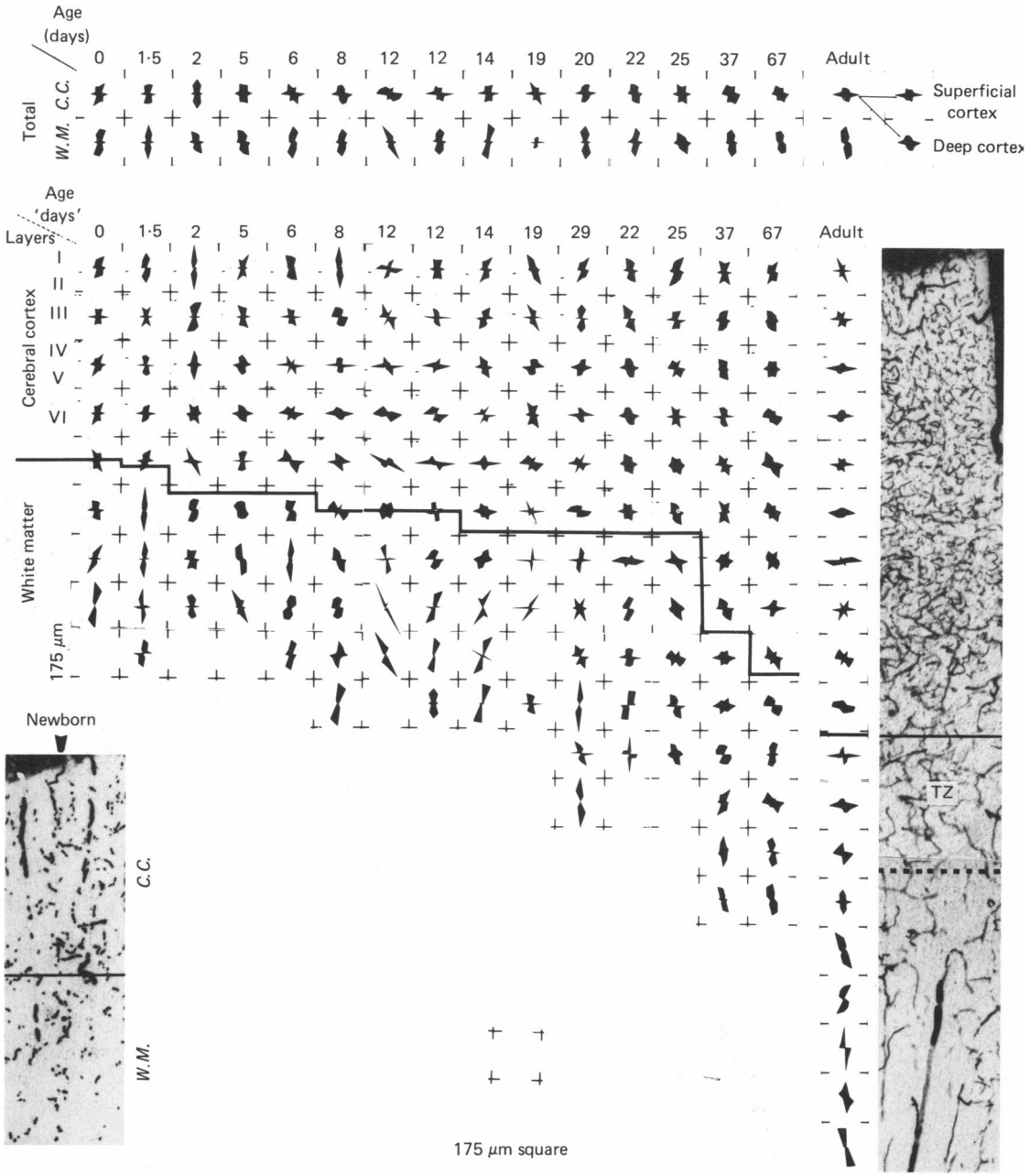

Fig. 7. 'Flower' diagrams of directions of vessels in the cerebral cortex and in the subjacent white matter. Upper diagram: total cerebral cortex and white matter. Lower diagram: analysed field by field (175 μm square). Note the appearance of the diagrams in the transitional zone (TZ) area and of the corresponding vessels. The cell layers are separated for each kitten by an arrow. *W.M.*, white matter; *C.C.*, cerebral cortex.

the cortex and of the white matter appeared to remain constant from 2 to 12 days, then increased slowly until at 67 days they reached three fifths of the adult values, which were approximately ten times those observed at birth.

Preferential orientation in the cerebral cortex

Vascular orientation was analysed in all layers of the cortex and in the subjacent white matter (Fig. 7). In the cortex as a whole, the vessels of the radiate type predominated from birth to 2 days, giving the cortex a resemblance to the white matter. From 5 days to 19–20 days, the development of tangential vessels changed the appearance of the 'flower' diagram considerably (compare 2 and 12 days, Fig. 7, upper diagram). From 22 to 67 days, there was a restoration of the multidirectional balance between vessels. In the adult, the tangential vessels predominated, although other orientations were also well represented. From 67 days to adulthood, the changes in preferential orientation produced a tangential predominance in the superficial cortex although vessels were multidirectional in the deep cortex.

When analysed layer by layer, vascular orientation in layer I of the cortex was predominantly radiate, regardless of age (Fig. 7, lower diagram). The same was true of layer II, up to 22 days after birth. From 25 days until adulthood, there was an equivalent distribution between tangential and radiate vessels. In layers III–VI, radiate vessels predominated until 2 days after birth; from 5 to 20 days there was growth of the tangential vasculature, so that between 22 and 67 days, the vessels were multidirectional. In the adult, the tangential component distinctly predominated in all layers, evidently after a relative directional reorganisation.

If horizontal orientation of the vessels were to be taken as a criterion of vascular maturity, then layer II was the most mature layer at birth; at 5 days, layers V and VI were the most mature; starting from 6 days, all layers showed a vascular maturity, and maturation progressed outwards toward the surface (Fig. 7, 'flower' diagrams of preferential orientation at 2 and at 5 days).

Preferential orientation in the white matter

Regardless of age, vascularisation in the white matter as a whole was predominantly radiate (Fig. 7). When analysed field by field, the white matter was still predominantly radiate in the youngest animals. From 12 days, the boundary between cortex and white matter became difficult to define using the criterion of vessel orientation, since the 'flower' diagram became multidirectional. For this reason, it was necessary to try to define the boundaries of the layers using sections stained with Unna blue. This made it possible to detect a transitional zone between cortex and white matter, containing both radiate and tangential vessels. This transition zone was twice as thick in the adult as at 12 days of age, and its vascular bed was less dense than that of the cerebral cortex. In addition, on approaching the white matter, the predominance of vessels of the radiate type was confirmed.

DISCUSSION

The anatomical analysis of a vascular territory by a technique using the natural content (blood) or artificially perfused content (colloidal carbon, or a dye, as a tracer) is probably not always adequate. As with other organs, such as glands (Craigie, 1920), it must be supposed that certain vascular territories remain closed

temporarily. Thus, it is possible that the results obtained with the Pickworth technique on non-perfused brains, i.e. the measurements of vascular coefficient and of preferential vascular orientation, may correspond more to the physiological reality than to the strictly anatomical reality. The results obtained on perfused brains, for evaluation of vascular diameter and density, may correspond more to the anatomical than to the physiological reality.

The shrinkage coefficient was used in this study to correct the experimental data and give them greater real precision, and is a method proposed by Eins & Wilhems (1976). The application of a correction using the growth coefficient to the curves obtained from our results has made it possible to compare potentially and functionally similar parts of the brain, regardless of the age of the animal.

Stages of development

The postnatal cerebral vascular maturation of the cat is slower than that of the rat and takes twice as long to attain the vascular profile of the adult. Using five indicators of vascular maturation (vascular mitosis, density, diameter, vascular coefficient, and the preferential vascular orientation), different consecutive periods are observed.

The first week of immaturity, exhibiting active mitoses, corresponds to the period of sprouting of Bär & Wolff (1973), with constant vascular diameter and vascular coefficient, an immature diameter (predominantly 2–3 μm), and a primarily radiate preferential orientation comparable to that of the white matter. This period is almost identical to that in the rat, which has a low and constant vascular density (Craigie, 1955; Singh & Nathaniel, 1975; Bär, 1978). This is succeeded by the period of vascular growth, as established by the indicator of vascular density, which reaches almost its maximum value. This period covers the second to the seventh weeks after birth and corresponds to the second and third weeks in the rat. It is characterised by a rapidly increasing vascular density and by an increase in the number of capillary ramifications (Bär & Wolff, 1973). Nevertheless, taking into consideration other indicators such as vascular diameter, vascular coefficient, and preferential orientation, this period can be subdivided into two stages in the cat.

The first stage, from the second week to 30 days after birth (premature period) is characterised by vascularisation of an immature type (maximum percentage of diameters, 2–3 μm), an increasing percentage of diameters greater than 10 μm, and a predominantly tangential orientation of vessels.

The second stage, from 30 to 49 days (pre-adult period), is characterised by a maximum but decreasing percentage of vascular diameters greater than 10 μm, and by both radiate and tangential orientation of vessels. The adult period resembles the pre-adult period, but with preferentially tangential orientation of vessels; some large vessels disappear in both the cortex and the white matter.

Gradients in the cerebral cortex

Parallel to a gradient of maturation of cortical neurons from the deep strata towards the superficial strata (Berry, Rogers & Eayrs, 1964; Rabinovitch, 1964; Caley & Maxwell, 1968*a*, *b*), it has been noted by Bär (1978) in the newborn rat, that there is a greater capillary density in the deep cerebral cortex than in the superficial cerebral cortex. Up to 3 weeks of age, he observes a more rapid development in the superficial and deep cortex, with a delay of 4 days in layer IV.

In the newborn kitten, the vascular densities in the superficial and the deep layers

of the suprasylvian gyrus are approximately equal. Consideration of the postnatal changes of vascular density in the cortical layers (Fig. 4) suggests that extrapolation into the prenatal period would give a vascular density of layers II–IV which is less than the vascular density of layers V–VI before birth, in agreement with observations made in the rat. This indicates that deep vascular maturation precedes superficial vascular maturation. Thereafter, superficial vascularisation develops to a greater extent than deep vascularisation until adulthood. This is in agreement with the description of a more highly developed vascularisation of the suprasylvian gyrus of the cat in layers III and IV (Campbell, 1939) than in other layers.

Vessels greater than 10 μm in diameter continue to form until 30 days, completing the population of transitional vessels existing at birth. It should be noted that at 42 days almost all indicators have reached their adult value, apart from the vascular coefficient, which remains at 37 % of the adult value. Consequently, the vessels increase essentially in length, growing neither in number nor in diameter. This is in agreement with Sakla (1965), who attributes the decrease in the number of vessels larger than 10 μm diameter to longitudinal vessel growth, a consequence of the increase in cerebral volume. The vessels which form after birth are mainly capillaries, i.e. their diameters are less than or equal to 8 μm (Bär & Wolff, 1973; Hunziker, Prey & Schulz, 1974; Mato & Ookawara, 1979; Conradi, Engvall & Wolff, 1980). Indeed between birth and adulthood it is these vessels (4–8 μm diameter) which increase in number, while those of other diameters decrease.

Myelination

The formation of the vascular bed and the commencement of myelination have been compared chronologically (Kennedy, Grove, Jehle & Sokoloff, 1970, 1972; Bär, 1978), but analysis of the present observations does not seem to reveal any direct correlation between myelination and vascular growth. While vascular density increases greatly in the cerebral cortex starting from the seventh day, myelination does not reach the cortex until 19 days, that is, 12 days after the beginning of vascular growth. Two studies have been devoted to the vascularisation of the white matter. Following anatomical studies of vasculogenesis (Craigie, 1955; De Reuck, 1972; Conradi & Sourander, 1980), Kennedy *et al.* (1972), studying the vascular flow, note a transitory increase in flow in relation to myelination.

White matter and transitional zone

In the kitten, the low vascular density and the high vascular coefficient of the white matter in comparison with the cerebral cortex indicate that the vessels in the white matter are of greater diameter. The vascular bed of the white matter is composed preferentially of radiate vessels, i.e. transitional vessels formed at an early stage (Wolff, 1978; Conradi *et al.* 1980), and a well represented group of capillaries, which is approximately constant up to 42 days after birth (70 to 73 % of the vascular population), reducing slightly in the adult (66 %), and probably belonging to the transitional zone. The transitional zone appears at the age of 12 days, and contains a vascular network with larger mesh than that of the cerebral cortex. The richness of this vascular network is expressed in its relatively high percentage of capillaries and in a preferential tangential orientation of its vessels (Fig. 7), recalling the appearance of the cortex. The transitional zone, supplied by corticomedullary vessels (De Reuck, 1972), seems to correspond to a zone rich in basal dendrites of pyramidal cells, in apical dendrites of abnormally oriented pyramidal cells (Van der Loos, 1965; Globus

& Scheibel, 1967), and in dendrites of other cells. It is not possible, either in the white matter or in the cerebral cortex, to detect different stages of development, for all the indicators increase slowly and regularly. This is the case even between 13 and 19 days, which is the period of progression of myelination between the white matter of the suprasylvian gyrus and layer VI of the cortex.

The vessels of the white matter are the first to form; proliferation occurs essentially in the fetal period, and their growth pre- and postnatally.

Synaptogenesis

Concerning the relationship with synaptogenesis observed in the cerebral cortex (Bär, 1978; Conradi *et al.* 1979, 1980), work in progress suggests the existence of a close correlation between synaptogenesis and vascular maturation. Synaptic growth and neuropil development commence from the second postnatal week in the suprasylvian gyrus of the cat, producing maximum synaptic density of the superficial cortex where the zone of maximum vascular density (layers II–IV) is located.

Structural significance

With respect to capillaries 2–3 μm in diameter it appears from the work of Mato & Ookawara (1979) that they may be functional, since rheology experiments demonstrate the extreme ease of distortion of erythrocytes. This provides no information, of course, concerning possible exchange through the vascular wall. The population of small vessels – potential capillaries, arterioles and venules – is very large at birth (40%) and up to 30 days (36%). In the immature state, all these vessels may play a role in metabolic exchange. With the maturation of the vascular system, each type of vessel seems to acquire its adult characteristics, i.e. selectivity and a preponderant role in metabolic exchange as compared with that of the intercellular space.

According to Johanson (1980), the permeability of the cerebral and cerebellar cortices to certain substances decreases *pari passu* with increase in vascularisation, and the formation of the blood–brain barrier is related to a decrease in the extracellular space. The permeability of small, immature vessels, such as capillaries, arterioles, venules, may therefore be temporary. Selective exchange comes with maturity; it concerns primarily the capillaries, whose endothelial membranes gradually become enriched in proteins which play a role in selectivity. The impermeability of the walls of arteriole and venule seems to be directly related to their thickness.

The extracellular spaces seem to regress in parallel with the development of the vessels (Caley & Maxwell, 1970) and of the neuropil. Until recently, there have been no definitive arguments making it possible to establish whether extracellular spaces larger than 20 nm are artifactual (Pappas & Purpura, 1964) or not (Luse, 1960; De Robertis & Gerschenfeld, 1961; Caley & Maxwell, 1970). However, the detection of anions in extracellular spaces (Derer & Nakanishi, 1980) seems to indicate that these spaces are indeed preferential routes of passage of metabolites during the perinatal period. As the vascular density is very low in the newborn as compared with the adult (even after correction using the growth coefficient), and the vascular diameter is small (40% of the vessels, 2–3 μm), the passage of metabolites would naturally be via the extracellular spaces.

The assumption that extracellular space accounts for 17–20% of the adult mammalian brain (Cragg, 1979) suggests that it continues to play a role in the passage of metabolites in the adult. The capillaries may then selectively complement the extra-

cellular space, necessary but not sufficient for the normal functioning of the cerebral tissue.

SUMMARY

Vascular growth in the median suprasylvian gyrus of the cat has been analysed quantitatively with respect to mitoses, vascular density, vascular diameters, vascular coefficient and preferential vascular orientation. After correction for shrinkage and growth, four maturation periods were identified:

(i) Immature period (first postnatal week), when the tissue exhibited numerous vascular mitoses, a low but constant vascular density and vascular coefficient, preferentially radiate vessels and immature (small) vascular diameters.

(ii) Premature period (second to fourth week), with few mitoses, a rapidly increasing vascular density, immature (small) vascular diameters, an increase in the number of vessels more than 10 μm in diameter, and a preferential orientation of tangential vessels.

(iii) Pre-adult period (fifth to sixth week), without mitoses. The vascular density increased greatly, the vascular diameters reached adult profile, the number of vessels over 10 μm in diameter decreased, the vascular orientations were both tangential and radiate and the vascular coefficient remained slight.

(iv) Adult period, when the vascular density and the vascular coefficient were maximal, the vessel diameters were of adult type, and the orientation was tangential.

From six weeks, the vessels increased only in length. Vascular maturation proceeded from the depth toward the surface of the cerebral cortex. Layers II–IV were those most highly vascularised, regardless of age. This is discussed in relation to synaptic growth. There was no direct relation between vasculogenesis and myelination.

The white matter had a typically radiate vascularisation. A transitional zone between cortex and white matter was identified. It had a loose mesh vascular network and corresponded to the area in which dendrites of inverted pyramidal cells were found.

The role of immature vascularisation in the nutrition of the neuropil is discussed.

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