Developmental changes in the expression of low-voltage-activated Ca\(^{2+}\) channels in rat visual cortical neurones

A. N. Tarasenko, D. S. Isaev, A. V. Eremin and P. G. Kostyuk

Department of General Physiology of the Nervous System, Bogomoletz Institute of Physiology, National Academy of Sciences, Bogomoletz Street 4, Kiev-24, 252024 Ukraine

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1. The functional properties of low-voltage-activated (LVA) Ca\(^{2+}\) channels were studied in pyramidal neurones from different rat visual cortical layers in order to investigate changes in their properties during early postnatal development. Ca\(^{2+}\) currents were recorded in brain slices using the whole-cell patch-clamp technique in rats from three age groups: 2, 3 and 12 days old (postnatal day (P) 2, P3 and P12).

2. It was demonstrated that LVA Ca\(^{2+}\) currents are present in neurones from superficial (I—II) and deep (V—VI) visual cortex layers of P2 and P3 rats. No LVA Ca\(^{2+}\) currents were observed in neurones from the middle (III—IV) layers of these rats. The LVA Ca\(^{2+}\) currents observed in P2 and P3 neurones from both superficial and deep layers could be completely blocked by nifedipine (100 \(\mu\)M) and were insensitive to Ni\(^{2+}\) (25 \(\mu\)M).

3. The density of LVA Ca\(^{2+}\) currents decreased rapidly during the early stages of postnatal development, while the density of high-voltage-activated (HVA) Ca\(^{2+}\) currents progressively increased up to the twelfth postnatal day. No LVA Ca\(^{2+}\) currents were found in P12 neurones from any of the layers. Only HVA Ca\(^{2+}\) currents with high sensitivity to F\(^{−}\) applied through the patch pipette were observed.

4. The kinetics of LVA Ca\(^{2+}\) currents could be well approximated by the \(m^2h\) Hodgkin—Huxley equation with an inactivation time constant of 24 ± 6 ms. The steady-state inactivation curve fitted by a Boltzmann function had the following parameters: membrane potential at half-inactivation, −86·9 mV; steepness coefficient, 3·4 mV.

5. It is concluded that, in visual cortical neurones, LVA Ca\(^{2+}\) channels are expressed only in the neurones of deep and superficial layers over a short period during the earliest postnatal stages. These channels are nifedipine sensitive and similar in functional properties to those in the laterodorsal (LD) thalamic nucleus. However, the cortical neurones do not express another ('slow') type of LVA Ca\(^{2+}\) channel, which is permanently present in LD thalamic neurones after the second postnatal week, indicating that the developmental time course of cortical and thalamic cells is different.

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start of differentiation, and then decrease as other types of ion channel are expressed (Veselovsky & Fomina, 1986). In rat and mouse dorsal root ganglion neurones LVA (T-type) Ca\(^{2+}\) channels show a peak of expression just around the time of birth, while later on their density starts to decline until their complete disappearance in most neurones beyond the first few weeks (Fedulova, Kostyuk & Veselovsky, 1986; Fedulova et al. 1994). In cultured Xenopus sensory neurones they are expressed only during the first 20–40 h of culture (Barish, 1991). The same is true for neurones cultured from adult or embryonic rat and guinea-pig hippocampal pyramidal neurones (O’Dell & Alger, 1991; Thompson & Wong, 1991) and rat neostriatum (Bargas, Surmeier & Kitai, 1991). In contrast to this, however, LVA Ca\(^{2+}\) channels are the predominant type of VOCC in certain thalamic and hypothalamic neurones and are a constant feature of the excitability mechanism, playing an important role in the organization of slow rhythmic activity (Akaïke, Kostyuk & Osipchuk, 1989; Huguenard & Prince, 1992). The developmental appearance of such channels is quite complicated. Thus, the population of LVA Ca\(^{2+}\) channels in neurones of the rat laterodorsal (LD) thalamic nucleus was found to be homogeneous in kinetic and pharmacological properties only during the first postnatal week. The corresponding Ca\(^{2+}\) currents demonstrated fast inactivation kinetics with a monoexponential time course (~30 ms) and high sensitivity to nifedipine (\(K_a = 2.6 \, \mu\text{M}\)). However, from the second postnatal week onwards, a more slowly inactivating component appeared in LVA Ca\(^{2+}\) currents from the same neurones, which showed insensitivity to nifedipine but high sensitivity to Ni\(^{2+}\) (Tarasenko, Kostyuk, Eremin & Isaev, 1997). The two types of LVA Ca\(^{2+}\) channel are probably associated with different neuronal functions, as the appearance of the ‘slow’ channels (with an inactivation time constant of ~60 ms) coincided with development of the dendritic tree in the corresponding cells.

These findings stimulated us to analyse in more detail the kinetic, pharmacological and developmental properties of LVA Ca\(^{2+}\) channels in neocortical neurones closely connected functionally to the neuronal activity of associative thalamic nuclei. The presence of LVA Ca\(^{2+}\) currents has been reported in pyramidal neurones in slices from the rat visual cortex (Franz, Galvan & Constanti, 1986; Sutor & Ziegglansberger, 1987) and in pyramidal neurones acutely isolated from the rat sensorimotor cortex (Sayer, Schwindt & Crill, 1990). However, the properties of these channels have not as yet been fully characterized. The cellular heterogeneity of the cortex and the difficulty of identifying specific cell types has substantially limited the study of characteristics and developmental changes. This complexity is evident from a recent study of pyramidal neurones from guinea-pig medial frontal cortex, which found that only cells located lower than 500 \(\mu\text{m}\) from the pial surface (approximately layers V–VI) were able to generate LVA Ca\(^{2+}\) currents and low-threshold Ca\(^{2+}\) spikes (LTS) (De la Pena & Geijo-Barrientos, 1996).

In the present study we attempted to investigate this problem further by using neurones from different layers of the rat visual cortex during the first 12 days of postnatal development.

### METHODS

#### Slice preparation

Brain slices were obtained from Wistar rat of three age groups: 2, 3 and 12 days old (postnatal day (P) 2, P3 and P12, the day of birth being designated as P1). The procedure to obtain brain slices was similar to that previously described by Tarasenko et al. (1997). Animals were anaesthetized with ether and decapitated only after observation of deep sleep. Incubation time in the recovery solution depended on the age of the animal: P2 and P12 slices were incubated for 30 and 60 min, respectively. Slices were 200 \(\mu\text{m}\) thick.

#### Patch-clamp recordings

Whole-cell patch-clamp recordings were performed as reported previously (Tarasenko et al. 1997). Patch pipettes were pulled from thick-walled molybdenum glass capillaries, and had 3–6 M\(\Omega\) resistance for P2 and P3 cortical neurones and 2–4 M\(\Omega\) resistance for P12 neurones. The inner diameters of the pipettes varied from 1 to 3.5 \(\mu\text{m}\) depending on the site of the cell studied.

Taking into account variations in membrane surface properties and cell morphology in the different cortical layers and postnatal developmental stages, we controlled both the membrane capacitance and input resistance of the neurones. The membrane capacitance was calculated by subtracting the pipette electrode capacitance from the whole-cell capacitance, which in turn was estimated from the integral of the corresponding transient current induced from a holding potential of ~80 mV by 10 mV hyperpolarizing pulses after rupture of the cell membrane. Generally, these transients had fast monoexponential time decay and could therefore be treated as a single monoexponential unit (Kay & Wong, 1985) and accepted for analysis. In addition, only those cells that completely satisfied all the criteria of voltage-clamp control formulated in our previous work (Tarasenko et al. 1997) were included for further study. We could not compensate the series resistance (\(R_s\)) but determined it from the amplitude and time constant of the capacitive current after rupture of the membrane. As the amplitude of Ca\(^{2+}\) currents did not exceed 200 pA and \(R_s\) was no more than 15 M\(\Omega\), the error in voltage clamp was less than 3 mV.

Input resistance was evaluated by dividing the voltage step by the leakage current. After rupture of the cell membrane the leakage currents were compensated by using an additional electrical circuit throughout the recording session. The compensation procedure was considered as satisfactory if the current trace elicited at the onset of a 10 mV hyperpolarizing command potential from the holding potential coincided with the zero line.

For observation of the upper surface of the slice we used an upright microscope (Nikon, Japan) with a X3.2 Semiplan objective lens fitted with a calibrated eyepiece. Visual cortex (areas 17 and 18) was identified according to the rat brain stereotaxic atlas (Paxinos & Watson, 1982), a x 20 Leitz Wetzlar objective lens was used to identify the type of cortical cell and the particular cortical layer. The cell surface was thoroughly cleaned before seal formation by saline streamed from a neighbouring pipette. Ca\(^{2+}\) currents were isolated by blocking Na\(^{+}\) and K\(^{+}\) channels and by replacing intracellular K\(^{+}\) with Cs\(^{+}\) (see Solutions and Drugs, below). Currents were recorded at room temperature (18–22 °C) using an analogue of
the List EPC-5 patch-clamp amplifier at an output cut-off frequency of 1 kHz (−3 dB, 8-pole active Bessel filter). Ca²⁺ currents were elicited by a series of increasing depolarizing steps every 5–7 s, digitally sampled at 8 µs intervals, stored and analysed with an IBM PC-compatible computer. The current amplitude was measured as the difference between peak inward current and zero current. Other specific protocols are described in the text or figure legends. Graphing and curve fitting was performed with SigmaPlot 5.0 (Jandel Scientific GmbH, Erkath, Germany). Time constants and current kinetics were described by fitting with a Hodgkin–Huxley model, using methods identical to those described earlier (Coulter, Huguenard & Prince, 1989).

**Results**

**Passive properties of visual cortical neurones**

The results presented were obtained in neurons located in different layers below the external surface of the cortex. As described in Methods, the membrane capacitance was evaluated as the integral of the capacitive transient obtained in response to hyperpolarizing command pulses of 10 mV from a holding potential of ~80 mV. In P2 rat neurons from deep layers the membrane capacitance was 23 ± 4 pF (n = 8). However, in P3 and P12 rats the population of cortical neurons was observed to be heterogeneous. Two groups of cells were identified. The neurons of the first group appeared to be in the process of increasing their dendritic tree, as their cell capacitances were 36 ± 5 pF (n = 10) and 76 ± 12 pF (n = 12) for ages P3 and P12, respectively (Fig. 1). The neurons of the second group did not appear to be undergoing changes in their cellular membrane surfaces as their capacitances were 24 ± 4 pF (n = 9) and 24 ± 6 pF (n = 7) for P3 and P12 cells, respectively; these cells probably corresponded to non-pyramidal neurons and were not included in any further analysis.

The situation was different for the neurons of layers I–II. We did not find any substantial developmental changes in the membrane capacitance of these neurons. Membrane capacitance was 52 ± 11 pF (n = 15), regardless of postnatal stage.

The decay in the capacitive transients of the cells (measured after rupturing the cell membrane) could be fitted by a single exponential curve. The time constant of decay (τ) varied, depending on the age group, from 140 µs to 1.2 ms. The series resistance determined from the capacitive transients did not exceed 15 MΩ for those neurons tested.

**Figure 1.** Passive properties of visual cortical pyramidal neurons

Developmental changes in capacitance of pyramidal neurons from deep layers of the visual cortex. The transient capacitive current was evoked by a 10 mV hyperpolarizing command potential from a holding potential of ~80 mV. Constant leakage current for the neurons shown here was 5, 10 and 12 pA for P2, P3 and P12, respectively. The calculated values of membrane capacitance were 29, 46 and 68 pF for P2, P3 and P12, respectively. The decay of the capacitive current was fitted by a single exponential curve with time constants of 0.52, 0.64 and 0.41 ms for P2, P3 and P12 neurons, respectively.
No age-related differences in leakage currents were observed. As a rule, the leakage current was ~20 pA and no greater than 27 pA. The mean input resistance of tested cells was 500 MΩ.

Developmental changes in Ca²⁺ conductance in cortical neurones from different layers

Typical records of Ca²⁺ currents in P2 and P3 neurones from deep layers are shown in Fig. 2. Generally, in P2 neurones, predominantly transient Ca²⁺ currents evoked by step depolarization to −55 mV from a holding potential of −95 mV could be observed. A sustained current may also have been revealed at depolarization potentials more positive than −45 mV but its mean value was significantly less than that of the transient current (Fig. 2A). The current–voltage (I–V) relationship had two maxima at about −50 and −15 mV, indicating the presence of both LVA and high-voltage-activated (HVA) Ca²⁺ currents (Fig. 2B). The amplitude of Ca²⁺ current induced by step depolarization to −50 mV was 38 ± 6 pA (n = 8). In P3 neurones under the same conditions, it was also possible to observe the LVA Ca²⁺ current (Fig. 2C). The I–V curve

![Figure 2](image_url)

**Figure 2. Changes in whole-cell Ca²⁺ current during the earliest stages of postnatal development**

A and C, depolarizing voltage steps from a holding potential of −95 mV for P2 (A) and P3 (C) neurones from layers V–VI evoked the superposition of transient LVA Ca²⁺ current and slowly inactivating HVA Ca²⁺ currents, the latter of which was predominant in P3 (C) neurones. The voltage protocol for each panel is shown at the top. B and D, I–V relationships of peak currents for neurones of the two age groups are presented beneath the current traces. The I–V relationships for neurones of both P2 and P3 age groups have 2 peaks, at about −50 and −15 mV, corresponding to LVA and HVA Ca²⁺ currents, respectively. For construction of the I–V curves data were averaged using 5 cells for P2 and 8 cells for P3 neurones.
again had two characteristic peaks around −50 and −15 mV (Fig. 2D). However, the amplitude of HVA Ca\(^{2+}\) current was 108 ± 12 pA, and it became predominant. There were no LVA Ca\(^{2+}\) currents in P12 neurones over a depolarization range between −75 and −45 mV (Fig. 3A) and the I−V curve had only one peak around −15 mV. The maximum amplitude of HVA Ca\(^{2+}\) current was 738 ± 64 pA (\(n = 7\)).

To verify that P12 neurones expressed only HVA Ca\(^{2+}\) channels, 5 μM F\(^−\) was added to the pipette solution to block HVA Ca\(^{2+}\) currents from the inside (Fig. 3B), a method previously used in isolated thalamic neurones (Bertollini, Biella, Wanke, Avanzini & De Curtis, 1994). As LVA Ca\(^{2+}\) current persisted in the presence of intracellular F\(^−\) (Carbone & Lux, 1987; Fraser & MacVicar, 1991), the I−V relationship could be expected to have two peaks if LVA Ca\(^{2+}\) currents were present. The blocking effect of F\(^−\) was 90 ± 5% (\(n = 12\)) and in the presence of this anion the I−V curve had only one peak at about −15 mV (Fig. 3C).

As it turned out, the expression of LVA Ca\(^{2+}\) channels was not a property common to all visual cortical neurones in the earliest stages of postnatal development. There were no LVA Ca\(^{2+}\) currents in neurones from the middle layer (III–IV) on either the second or twelfth postnatal days. However, the developmental changes in neurones from the superficial layer (I–II) had exactly the same time course as those of neurones in the deeper layer (V–VI). LVA Ca\(^{2+}\) currents were present in P2 and P3 neurones, but only HVA Ca\(^{2+}\) currents were recorded in P12 neurones. To clarify the question as to whether the neurones from deep
and superficial layers expressed the same LVA Ca$^{2+}$ channels, a special pharmacological and kinetic study of LVA Ca$^{2+}$ currents in neurones from different layers was undertaken.

**Pharmacological properties of LVA Ca$^{2+}$ current**

As we have shown previously (Tarasenko et al. 1997), LD thalamic neurones can express two types of LVA Ca$^{2+}$ channels with different sensitivities to nifedipine and Ni$^{2+}$. In the present study we found that neurones from both deep and superficial layers had LVA Ca$^{2+}$ currents with high sensitivity to nifedipine (Fig. 4A). The nifedipine-sensitive component was obtained as the difference between the control current and the current obtained after blocking by nifedipine (100 μM). The amplitude of nifedipine-sensitive current in P2 deep neurones (Fig. 4B) was larger (22 ± 4 pA, n = 6) than that in P3 neurones (12 ± 4, n = 8). The same postnatal changes in amplitude of LVA Ca$^{2+}$ currents were observed in neurones from the superficial layer. To determine whether any other component was present in the LVA Ca$^{2+}$ current, Ni$^{2+}$ at 25 μM was applied to the neurones tested. No changes were observed in the control Ca$^{2+}$ current in either deep (n = 5) or superficial neurones (n = 7).

**Steady-state inactivation of the LVA Ca$^{2+}$ current**

Taking into account the specificity of steady-state inactivation of the slow LVA Ca$^{2+}$ current component in LD neurones (half-inactivation membrane potential ($V_{0.5}$), −100 mV; Tarasenko et al. 1997), in the next part of our study a special protocol for recording Ca$^{2+}$ currents was used. A membrane depolarization to −50 mV followed a hyperpolarization pulse in the range −90 to −120 mV of 500 ms duration. There was no difference in the amplitude of LVA Ca$^{2+}$ currents between neurones from the two (superficial and deep) layers, demonstrating the presence of the same type of LVA channel. The recordings of LVA Ca$^{2+}$ currents made using this protocol are shown in Fig. 5A. The inactivation time constant for LVA Ca$^{2+}$ current was the same using this protocol as when using the normal protocol (see above), at 23 ± 4 ms (n = 4). This fact led us to conclude that visual cortical neurones do not express slowly inactivating LVA Ca$^{2+}$ channels after the second postnatal week, although they are permanently present in LD thalamic neurones (Tarasenko et al. 1997). The normalized peak amplitudes of the evoked currents could be fitted by a Boltzmann function (Fig. 5B). The $V_{0.5}$ of −86.9 mV and slope factor (k) of 3.4 mV were similar to those described in LD thalamic neurones for the ‘fast’ component ($I_{f}$) of the LVA Ca$^{2+}$ current.

**Kinetic properties of the LVA Ca$^{2+}$ current**

The LVA Ca$^{2+}$ current induced by membrane depolarization to −60 mV from a prepulse potential of −95 mV showed monoeXponential decay and declined completely in about 100 ms in both P2 and P3 neurones. Analysis of the activation and inactivation time courses of this current indicated that it could be well fitted in terms of the $m^2 h$ Hodgkin–Huxley equation, as has already been shown for the kinetics of other types of Ca$^{2+}$ currents (Kostyuk, Krishtal, Pidoplichko & Shakhovalov, 1979; Kay & Wong, 1982).

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**Figure 4. Blocking effect of nifedipine on LVA Ca$^{2+}$ current in P2 visual cortical neurones**

A, LVA Ca$^{2+}$ current recorded in P2 neurones before (Control) and after blocking by 100 μM nifedipine. The current was obtained by 35 mV step depolarization from a holding potential of −95 mV (top). B, the nifedipine-sensitive component was obtained by subtracting LVA Ca$^{2+}$ current after blocking by 100 μM nifedipine from the control LVA Ca$^{2+}$ current. It was fitted in terms of the $m^2 h$ model (continuous line) with the inactivation time constant ($\tau_{i,n}$) indicated.
The equation used for fitting the LVA Ca\(^{2+}\) current \((I_{Ca})\) was:

\[
I_{Ca} = A[1 - \exp(-t/\tau_m)]\left[\exp(-t/\tau_h) + I_\infty/A\right], \tag{1}
\]

where \(t\) is time, \(\tau_m\) and \(\tau_h\) are the activation and inactivation time constants, respectively, \(I_\infty\) is a sustained \(Ca^{2+}\) current, and \(A\) is an amplitude scaling factor. The fitting parameters for the LVA \(Ca^{2+}\) current in P2 neurones (Fig. 6A) were \(\tau_m = 4.2 \pm 0.6\) ms and \(\tau_h = 26 \pm 5\) ms (\(n = 14\)) for activation and inactivation, respectively (Fig. 6B). Since the pharmacological sensitivity and kinetics of the LVA current in cells from both age groups did not show significant differences \((P > 0.1)\), we concluded that these neurones express nifedipine-sensitive LVA \(Ca^{2+}\) channels similar to 'fast' channels expressed in P12 LD neurones.

Postnatal changes in densities of \(Ca^{2+}\) current

According to our estimation, the current density for LVA currents in P2 neurones was \(1.65 \pm 0.17\ pA\ \mu F^{-1}\ (n = 6)\) and for HVA currents it was \(1.08 \pm 0.13\ pA\ \mu F^{-1}\ (n = 8); current density did not depend upon whether the neurone was situated in the deep or the superficial layer. However, already by the third postnatal day (P3), the relative proportion of LVA to HVA \(Ca^{2+}\) currents had substantially changed. The density of LVA \(Ca^{2+}\) current \((1.34 \pm 0.12\ pA\ \mu F^{-1}\ (n = 6))\) in P3 neurones became smaller compared with that of neurones of the first age group, P1 \((P < 0.01)\). On the other hand, the density of slowly inactivating HVA \(Ca^{2+}\) current in P3 neurones became significantly greater than in P2 neurones and reached \(3.08 \pm 0.31\ pA\ \mu F^{-1}\ (n = 6)\). By the twelfth postnatal day the density of HVA \(Ca^{2+}\) current had increased considerably compared with P2 and P3 neurones, reaching \(9.7 \pm 0.7\ pA\ \mu F^{-1}\ (n = 12)\).

Figure 5. Steady-state inactivation of LVA \(Ca^{2+}\) current in P2 neurones

\(A\), voltage dependence of steady-state inactivation of the nifedipine-sensitive LVA \(Ca^{2+}\) current. The voltage protocol is shown at the top. The membrane was depolarized to \(-50\) mV from different holding potentials between \(-105\) and \(-75\) mV. The duration of the prepulse impulse was \(500\) ms. \(B\), the inactivation curve fitted by a Boltzmann equation \((I/I_{\text{max}} = [1 + \exp(V - V_{1/2})/k])^{-3}\), where \(I\) is the current amplitude, \(I_{\text{max}}\) is the maximum current amplitude, \(V\) is the voltage, \(V_{1/2}\) is the half-inactivation voltage and \(k\) is the slope factor) with \(V_{1/2} = -86.9\) mV and \(k = 3.4\) mV \((n = 7)\).
DISCUSSION

Using the whole-cell patch-clamp technique combined with the direct visualization of neurones during recording in brain slice preparations, we studied the early postnatal development of the electrophysiological properties of pyramidal and non-pyramidal neurones of rat visual cortex. Special attention has been paid to LVA Ca^{2+} currents because of the multiple roles these currents play in the functioning of different neurones (short-lasting currents that trigger neuronal morphogenesis and continuous currents that are responsible for special forms of neuronal activity) as well as the recently established existence of several kinetically and pharmacologically different subtypes of the corresponding LVA Ca^{2+} channels (Tarasenko et al. 1997).

The present study has demonstrated that LVA Ca^{2+} channels are expressed in the visual cortex only in pyramidal neurones of deep (V—VI) and superficial (I—II) layers for a very short period during the earliest stages of postnatal development. HVA Ca^{2+} channels are expressed in these neurones at this developmental time at a much lower density. Obviously, the LVA channels immediately after birth are the main gates for intracellular Ca^{2+} entry. These channels are, by their kinetic and pharmacological properties, analogous to those previously described by us as ‘fast’ LVA Ca^{2+} channels.

![Inactivation kinetics of LVA Ca^{2+} currents in P2 and P3 visual cortical neurones](image)

Figure 6. Inactivation kinetics of LVA Ca^{2+} currents in P2 and P3 visual cortical neurones

A and B (left), a transient LVA Ca^{2+} current in P2 (A) and P3 (B) deep layer neurones activated by depolarization to −60 mV from a prepulse potential of −95 mV. A and B (right), the current traces were fitted by computer simulation using the \( m^2 h \) equation with the inactivation time constants (\( \tau_i \)) indicated. The residue (Res) current was obtained by subtraction of the simulated current from the LVA Ca^{2+} current (control).
Ca\(^{2+}\) channels in the LD thalamic neurones, indicating that in both cases the function of these channels may be similar, i.e. triggering neuronal differentiation and establishing neuronal connections. However, the time characteristics of the expression of these channels in neocortical and thalamic neurones are substantially different. In cortical neurones, by as early as the third postnatal day the density of LVA Ca\(^{2+}\) channels disappeared simultaneously in the superficial and deep cortical layer neurones; during neocortical development the interaction between neurones of layer I–II (Cajal-Retzius cells) and layer V–VI is extremely important for the correct positioning of migrating neurones and their maturation (Ogawa et al. 1995).

In the middle layer (II–III) neurones, only HVA Ca\(^{2+}\) currents were found even on days P2 and P3. One day later, the pyramidal neurones from both the deep and superficial layers also started to express a substantial number of HVA Ca\(^{2+}\) channels. It has been shown previously that in guineapig medial frontal cortex only pyramidal neurones are able to generate low-threshold Ca\(^{2+}\) spikes; they were rarely observed in layers II–III and frequently in layers V–VI (De la Pena & Geijo-Barrientos, 1996). This may indicate that neurones in the middle layers differentiate very early on and achieve maturity even before birth, while those in the superficial and deep layers still have immature membrane properties during the postnatal period and continue to develop over the first two postnatal weeks (Zhou & Hablitz, 1996).

In contrast to neocortical neurones, neurones of certain thalamic and hypothalamic structures maintain a high density of LVA Ca\(^{2+}\) channels in the adult state (Akaike et al. 1988; Huguenard & Prince, 1992). Obviously, besides contributing in some way to neuronal differentiation and the establishment of synaptic contacts, these channels are important for generating specific types of neuronal activity in the form of membrane potential oscillations and repetitive firing, enabling thalamic neurones to act as pacemaker elements in thalamocortical circuits (Dossi, Nuñez & Steriade, 1992; Huguenard & Prince, 1992; Kang & Kwei, 1993; Hutschison, Miura, Yarom & Puil, 1994). The observed differences clearly indicate a leading role for subcortical structures in the generation and maintenance of the corresponding rhythms. The Ca\(^{2+}\) channels responsible for such activity evidently form a special subtype of LVA channel, differing from the transient subtype in inactivation kinetics, location and pharmacology. As has already been stated, in LD thalamic neurones these channels seem to be located mainly on dendrites; in accord with this location, the thalamocortical rhythmic activity in rats starts just after maturation of the dendritic tree of the thalamic neurones (Muller, Miegjeld & Swandulla, 1992; Karst, Joels & Wadman, 1993).

The pharmacological specificity of the permanent LVA Ca\(^{2+}\) channels in thalamic neurones merits special attention. A more extensive evaluation of possible specific antagonists to this type of channel may produce drugs that could attenuate paroxysmal thalamocortical activity without affecting the normal course of postnatal development of the corresponding structures.


