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Research Report

Distinct developmental patterns in the expression of transient, persistent, and resurgent Na$^+$ currents in entorhinal cortex layer-II neurons

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ABSTRACT

Sub- and near-threshold voltage-dependent Na$^+$ currents (VDSCs) are of major importance in determining the electrical properties of medial entorhinal cortex (mEC) layer-II neurons. Developmental changes in the ability of mEC layer-II stellate cells (SCs) to generate Na$^+$-dependent, subthreshold electrical events have been reported between P14 and P18. In this study we examined the modifications occurring in the various components of VDSCs during postnatal development of mEC SCs. The transient, resurgent, and persistent Na$^+$ currents (I_{NaT}, I_{NaR}, and I_{NaP}, respectively) showed distinct patterns of developmental expression in the time window considered (P5 to P24–27). All three currents prominently and steeply increased in absolute amplitude and conductance from P5 to at least P16. However, capacitive charge accumulation, an index of membrane surface area, also markedly increased in the same time window, and in the case of I_{NaT} the specific conductance per unit of accumulated capacitive charge remained relatively constant. By contrast, specific I_{NaR} and I_{NaP} conductances showed a significant tendency to increase, especially from P5 to P18. Neither I_{NaR} nor I_{NaP} represented a constant fraction of the total Na$^+$ current at all developmental ages. Indeed, detectable levels of I_{NaR} and I_{NaP} were present in only ~20% and ~70%, respectively, of the cells on P5, and were observed in all cells only from P10 onwards. Moreover, the average I_{NaR}-to-I_{NaT} conductance ratio increased steadily from ~0.004 (P5) up to a plateau level of ~0.05 (P22+), whereas the I_{NaP}-to-I_{NaT} conductance ratio increased only from ~0.009 on P5 to ~0.02 on P22+. The relative increase in conductance ratio from P5 to P22 was significantly greater for I_{NaR} than for I_{NaP}, indicating that I_{NaR} expression starts later than that of I_{NaP}. These findings show that in mEC layer-II SCs the single functional components of the VDSC are regulated differentially from each other as far as their developmental expression is concerned.

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Abbreviations: AP, action potential; EC, entorhinal cortex; mEC, medial entorhinal cortex; I_{NaP}, persistent Na$^+$ current; I_{NaR}, resurgent Na$^+$ current; I_{NaT}, transient Na$^+$ current; MPOs, membrane-potential oscillations; SCs, stellate cells; VDSC(s), voltage-dependent sodium current(s); VGSC(s), voltage-gated sodium channel(s)

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1. Introduction

The functional behavior and electrophysiological properties of medial entorhinal cortex (mEC) layer-II neurons, and of stellate cells (SCs) in particular, have been recently the object of considerable interest, for various reasons: first, SCs are believed to represent the morphological correlate of mEC grid cells (Burgalossi et al., 2011; Schmidt-Hieber et al., 2010); second, these neurons exhibit an intrinsic, subthreshold, oscillatory activity in the theta frequency range (Alonso and Klink, 1993; Alonso and Llinás, 1989) that is likely related with their operative frequency preference and the emergence of theta-rhythm population activity in the EC, and the frequency of which correlates with grid field spacing (Giocomo et al., 2007); third, their excitable properties can undergo state- and activity-dependent modulatory processes able to promote emergent activity patterns in the form of sustained afterdischarges (Magistretti et al., 2004), which may represent a cellular correlate of the EC role in memory function. Voltage-dependent Na^+ currents (VDSCs) active in a sub- or near-threshold range of membrane potential are believed to be of special importance in the physiology of mEC layer-II neurons. The interplay of the persistent Na^+ current, I_{NaP} (Crill, 1996; Magistretti and Alonso, 1999), with the hyperpolarization-activated cation current, I_h, is known to underlie the generation of subthreshold, theta-like membrane-potential oscillations (MPOs) (Dickson et al., 2000; Fransén et al., 2004) that are a characteristic feature of mEC SCs. In turn, theta-like MPOs promote a near-threshold firing pattern, typical of SCs, characterized by spike clustering (Alonso and Klink, 1993; Dickson et al., 2000), and are believed to favor the emergence of theta-rhythm oscillatory activity in the EC network. In cortical and hippocampal neurons I_{NaP} is also implicated in the amplification of membrane-potential resonance (Hu et al., 2002; Hutcheon and Yarom, 2000), and in mEC SCs the channel activity underlying this current has been shown to importantly contribute to subthreshold membrane-potential noise (White et al., 1998) which can be the trigger of resonant and oscillatory behavior and significantly influence action-potential (AP) discharge organization (Dorval and White, 2005; White et al., 1998).

As compared to a number of other cell populations of the medial temporal-lobe memory system, mEC layer-II neurons also show an unusually high expression level of the resurgent sodium current (I_{NaR}) (Castelli et al., 2007b). I_{NaR} is a Na^+-current component activated upon repolarization following large, transient depolarizations such as the AP (Raman and Bean, 1997), and has been shown to depend on a Na^+-channel blocking mechanism promoted by channel activation and in competition with the process of classical inactivation (Raman and Bean, 2001). I_{NaR}, and the resurgence mechanism underlying this current, are potentially able to accelerate return of membrane potential to the threshold level after discharge of an action potential, enhance repetitive firing (Castelli et al., 2007a; Khaliq et al., 2003; Magistretti et al., 2006), induce depolarizing after potentials (Magistretti et al., 2006), and promote burst firing (Enomoto et al., 2006; Raman and Bean,

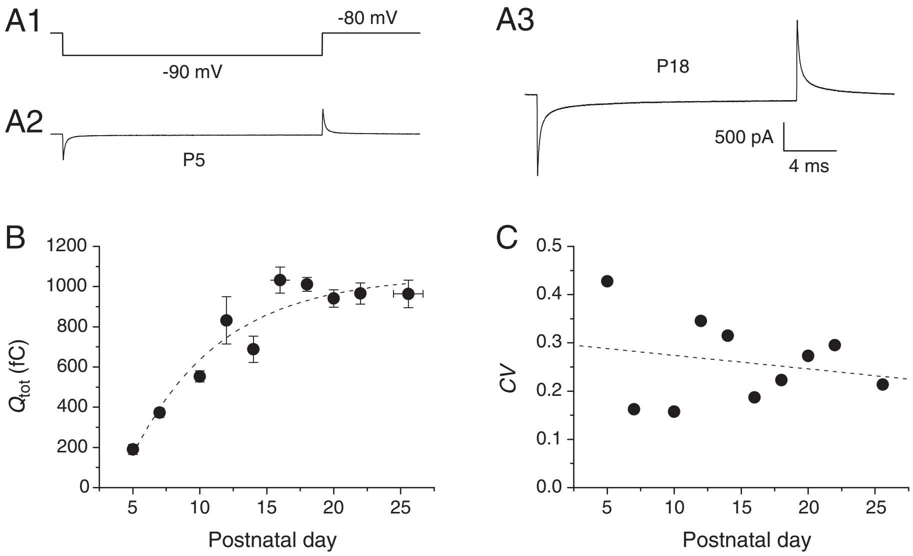

Fig. 1 – Electrophysiological evaluation of postnatal increase in membrane surface in mEC SCs. (A) Voltage-clamp protocol applied to elicit capacitive transients (A1) and currents recorded in response thereof in two representative neurons on P5 (A2; cell C9331) and P18 (A3; cell B7702). The negative-going transient in each recording was used, after zeroing the steady-state current observed at the end of the –10 mV pulse, to calculate the integral of current over time, thereby obtaining the total charge (Q_{tot}) transferred during the capacitive transient. (B) Plot of average Q_{tot} as a function of postnatal age (in days). The dashed line is the exponential best fitting to data points, obtained by applying Eq. (1) (see the Experimental procedures) as the fitting function (fitting parameters are specified in Supplemental Table 1). Numbers of observations were: 11 (P5), 7 (P7), 10 (P10), 6 (P12), 11 (P14), 9 (P16), 43 (P18), 35 (P20), 29 (P22), 9 (P24+). (C) Plot of the coefficient of variation (CV) of average Q_{tot} as a function of postnatal age. The straight, dashed line is the linear regression to data points: slope coefficient was –0.0027/day; the two variables were not significantly correlated ($p=0.57$).