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SHORT COMMUNICATION

ATPase activity in glial cells and in neuronal perikarya of rat cerebral cortex during early postnatal development[1]

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ATPase activities during development have been studied in rat brain (Samson and Quinn, 1967), in nerve-ending particle fractions from rat brain (Abdel-Latif, Brody and Ramahi, 1967) and in the cerebral cortex of the kitten (Huttenlocher and Rawson, 1968), as well as in preparations of synaptosomes and microsomes of rat brain (Abdel-Latif, Smith and Ellington, 1970). In a previous study, we compared the activities of Na–K ATPase and total ATPase of the cerebral cortical neuronal perikaryal fraction and glial cell fraction obtained as two separate populations by a bulk-isolation procedure (Medzihradsky, Nandhasri, Idoyaga-Vargas and Sellinger, 1971). In that study on 18–20-day-old rats, the glial cell fraction exhibited levels of both activities markedly higher than those of the neuronal fraction. In the present work, the activity of the Na–K ATPase and of the ouabain-insensitive ATPase were determined in the two cellular fractions during the postnatal development of rat cerebral cortex. The results of this study may contribute to our knowledge of the formation of cellular processes and synaptic junctions, elements rich in Na–K ATPase (Kurokawa, Sakamoto and Kato, 1965; Skou, 1965; Whittaker, 1965; Abdel-Latif *et al.*, 1967; Appel, Autilio, Festoff and Escueta, 1969), especially since Huttenlocher and Rawson (1968) found a close correlation between the activity of this enzyme and the spontaneous cortical activity during development in kitten cerebral cortex.

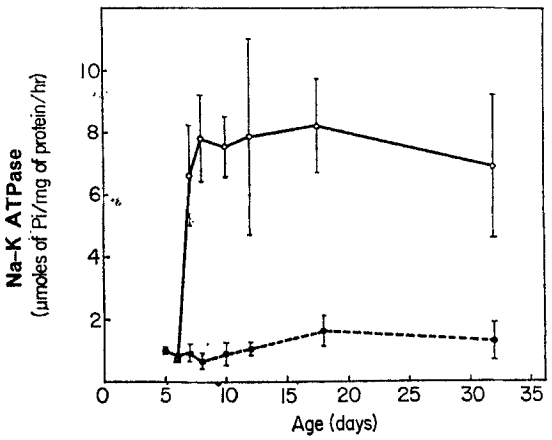

Fig. 1.—The specific activities of the Na–K ATPase in the neuronal perikaryal fraction (●– – – –●) and in the glial cell fraction (○———○) isolated from rat cerebral cortex during postnatal development. For each experiment, the fractions were isolated from a pool of at least 10 cerebral cortices by the procedure of Sellinger *et al.* (1971) and assayed as described by Medzihradsky *et al.* (1971). The Na–K ATPase activity was calculated by subtracting the ATPase activity determined in the presence of 1×10^{-4} M-ouabain from the enzyme activity obtained in the absence of this inhibitor. Each determination was run in duplicate and each time-point represents the results of three to five experiments. The mean values ± S.D. are given.

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The neuronal perikaryal fraction and the glial fraction were prepared from the cerebral cortices of male, Sprague–Dawley rats, as described previously (SELLINGER, AZCURRA, JOHNSON, OHLSSON and LODIN, 1971). The age of the animals ranged from 5 to 32 days. For each preparation the cortices of at least 10 rats were pooled and in each age group a minimum of three preparations was analysed. The sampling technique, assay of ATPase activity and determination of protein were as described previously (MEDZIHRADSKY *et al.*, 1971). For reasons indicated in that report, the cellular fractions used in the present study were also subjected to repeated freezing (−70°C) and thawing prior to enzyme assay. The specific activities of both the Na–K ATPase and of the ouabain-insensitive ATPase showed a similar pattern during postnatal development in each of the cellular fractions (Figs. 1 and 2). However, the developmental pattern of both enzymes in the glial cells was considerably different from that of the neuronal enzymes. In the glial fraction, a sharp increase of both enzymes was observed between days 6 and 12 (Fig. 1 for Na–K ATPase and Fig. 2 for the ouabain-insensitive ATPase). The abrupt 12-fold rise of the Na–K ATPase between the 6th and 8th postnatal day was especially striking (Fig. 1). The corresponding activities of the enzymes in the neuronal perikarya varied to a much lesser degree (Figs. 1 and 2).

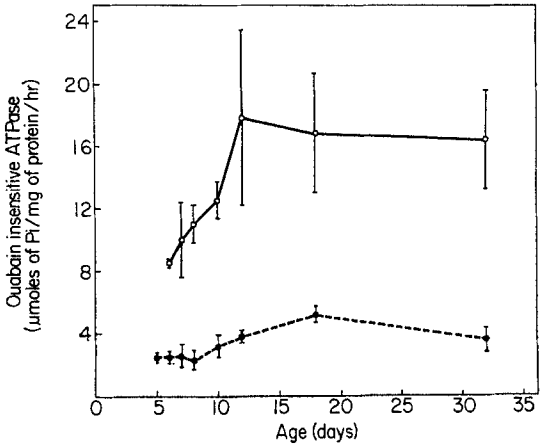

FIG. 2.—The specific activities of the ouabain-insensitive ATPase in the neuronal perikaryal fraction (●----●) and in the glial cell fraction (○———○) isolated from rat cerebral cortex during postnatal development. Experimental details were as described in the legend to Fig. 1.

In a study of the correlation of spontaneous cortical activity and ATPase activities, HUTTENLOCHER and RAWSON (1968) reported a 12-fold rise of the Na–K ATPase activity between 7 days and 6 weeks postnatally. During the same time, the ouabain-insensitive ATPase increased four-fold. The specific activity of Na–K ATPase assayed in rat brain homogenates increased 5-fold during the first 20 postnatal days, the sharpest rise occurring between days 5 and 10 (SAMSON and QUINN, 1967). Again, the increase of the ouabain-insensitive ATPase component was considerably smaller. ABDEL-LATIF *et al.* (1967) reported a dramatic increase in the specific activity of the Na–K ATPase and of the ouabain-insensitive ATPase in the nerve-ending fraction of the developing rat brain just prior to birth and in the 1-day-old animal. The rise in Na–K ATPase continued up to 15 days of age. Finally, an abrupt rise of the Na–K ATPase in preparations of synaptosomes and microsomes of rat brain was noted during the first 10 days of life after which time the enzyme activity levelled off (ABDEL-LATIF *et al.*, 1970). Whereas in the present study the general pattern of appearance of both enzyme activities in the two cellular preparations correlates well with that obtained by other workers for the enzymes in brain homogenates and subcellular fractions, the abrupt increase of the Na–K ATPase between days 6 and 8 was observed only in the glial cells (Fig. 1). In confirmation of the previous observations made on 18–20-day-old rat cerebral cortices (MEDZIHRADSKY *et al.*, 1971), both ATPase activities were considerably higher in the glial cells than in the neuronal fraction during the entire period of postnatal development examined in this study.

In a recent communication, HENN, BLOMSTRAND and HAMBERGER (1971) underlined the functional importance of the Na–K ATPase in glial cells as a regulator of the extracellular K^+ concentration in the neighbourhood of synaptic junctions. On the other hand, EMBREE, HESS and SHEIN (1971) reported levels of Na–K ATPase activity in astroglial cells grown subcutaneously in newborn hamsters to be

only 1 per cent as high as those of the average intact cell of the upper layer of rat cerebral cortex in which neurons abound. Finally, we again emphasize that the measurements of ATPase made on neuronal perikaryal preparations exclude ATPase associated with the neuronal processes and with the synaptic membranes shown by JOHNSON and SELLINGER (1971) to be virtually absent from these preparations.

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