ADULT ASTROGLIA IS COMPETENT FOR NA^+/CA^{2+} EXCHANGER-OPERATED EXOCYTOTIC GLUTAMATE RELEASE TRIGGERED BY MILD DEPOLARIZATION

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Glial sub-cellular particles (gliosomes) have been purified from rat cerebral cortex and investigated for their ability to release glutamate when subjected to depolarizing agents. Previous work has shown that gliosomes represent a viable astrocytary preparation poorly contaminated by synaptosomes, which exhibits several ~30 nm non-clustered cytoplasmic vesicles and contains most of the proteins of the exocytotic machinery. Moreover, the Ca^{2+} ionophore ionomycin efficiently stimulated the release of glutamate by a mechanism resembling neuronal exocytosis (1).
We show here that KCl (15, 35 mM), 4-aminopyridine (0.1, 1 mM) or veratrine (1, 10 μM) induced Ca^{2+}-dependent glutamate release from gliosomes. KCl increased membrane potential, cytosolic [Ca^{2+}] and cytoplasmic vesicle fusion rate in gliosomes; Ca^{2+} entry and glutamate release were independent from VSCC opening; they were instead abolished by KB-R7943, suggesting a role for the Na^+/Ca^{2+} exchanger working in reverse mode. KCl induced a sustained increase of intracellular [Ca^{2+}] and Ca^{2+}-dependent glutamate release also in cultured astrocytes prepared from adult but not from neonatal rats. Glutamate release was even more marked in *in-vitro* neuron-conditioned adult astrocytes. As for gliosomes, K^+-induced Ca^{2+} influx into cultured adult astrocytes and glutamate release were abolished by KB-R7943. Interestingly, proteins involved in the release machinery, such as VAMP, syntaxin, GLAST or GLT1, were much more concentrated in gliosomes than cultured astrocytes, suggesting their origin from specialized areas of the astrocyte. These data indicate that *in-vivo* maturation allows cortical astrocytes to release glutamate by a process involving depolarization-triggered exocytosis. (Supported by grants from Italian Ministry of University)

(1) Stigliani S. Zappettini S., Raiteri L., Passalacqua M., Melloni E., Venturi C., Tacchetti C, Diaspro A., Usai C., Bonanno G., J. Neurochem. (2006) 96: 656-668.