Calcium-mediated transductive systems and functionally active gap junctions in astrocyte-like GL15 cells

ABSTRACT

In conclusion, results from this study support the use of the GL15 cell line as a suitable in vitro astrocyte model, which provides a valuable guide for studying glial physiological features at various differentiation phases.

INTRODUCTION

Background Astrocytes are the most abundant cell type of the central nervous system, where they are closely involved in the modulation of the activity of neuronal components. Astrocytes play a pivotal role in several physio-pathological brain events that involve the synthesis and secretion of neurotrophic growth factors. In addition, it has been shown that neurotrophin-mediated signalling may not be the only mechanism involved in astrocyte-neuron interactions. In fact, the presence of specific intercellular connections (gap junctions) between these two cell populations, which allow direct and selective cell-to-cell exchange of chemical signals (ions, small metabolites), may represent an additional, rapid and unique way for astrocytes to communicate with each other and to interact with adjacent neurons. In mammalian astrocytes, extracellular physiological agonists are able to increase the concentration of intracellular Ca2+ ([Ca2+]i) via voltage-dependent channels or controlled release from internal stores (via inositol triphosphate receptors and/or ryanodine receptors). This is one of the most utilised mechanisms for modulating astrocyte functions. However, Ca2+ waves, which are transmitted from cell to cell via gap junctions (gjs), are thought to be important for co-ordination of astroglial function. The genesis and propagation of Ca2+ waves were originally observed in brain-derived cell populations in culture and, more recently, this event has also been demonstrated in more integrated systems, such as brain slice preparations and living rat brain. In spite of the large number of contributions published in the last decade, the mechanism(s) involved in the genesis and propagation of Ca2+ waves are not yet clear. Moreover, there is insufficient data from in vivo experiments, especially those on human astrocytes. About ten years ago, the GL15 cell line was established from human glioblastoma multiforme. GL15 cells were characterised as an astroglial-like cell line by the study of the cell karyotype and immunohistochemical and cytogenetic demonstration of glial fibrillary acidic protein (GFAP) expression. Moreover, other biochemical properties peculiar to astroglials were found in the GL15 cellular population that confirmed their astroglial origin; for example, expression of glutamine synthetase, taurine transport, transforming growth factor receptor expression and interleukin-induced apoptosis. Although the data derived from the previous studies support the presence of an astroglial phenotype, as yet no determination has been made concerning the main physiological characteristics of the GL15 cells in relation to their differentiation. Therefore, we decided to focus our attention on one of the most important aspects of astrocyte physiology: the mechanism(s) of cell communication. Considering that in vivo astrocytes are capable of cellular communication both via membrane surface receptor-operated systems and/or gjs between two neighbouring cells, the investigation of the presence and activity of these mechanisms is fundamental in proposing GL15 cells to be an in vitro model of astrocytes. For these reasons, we define the characteristics of this model by analysing some morphological aspects, the mechanism of [Ca2+]i increase induced by different extracellular physiological agonists and the expression and functional capacity of the gjs system in relation to

the differentiative pathway.

CONCLUSION

Conclusions In conclusion, the data reported in this paper support the reliability of the GL15 cell line as a suitable in vitro model for astrocytes, which should aid in the investigation of their distinctive physiological properties, and subsequently contribute to clarifying the complex role of this cell type in the brain. It is important to remember that, by simply utilising the differentiated or undifferentiated phenotype of this cell line, it is possible to study the modality by which the cells communicate with each other, either via gjs and/or membrane receptors. The proposed model becomes even more fascinating when the human origin of this cell line is considered. This new astrocyte model provides a stepping-stone in the efficient analysis and interpretation of problems regarding the role of astrocytes during modulation and remodelling of the nervous system, their contribution to the electro-physiological activity of neurons and other relevant mechanisms.