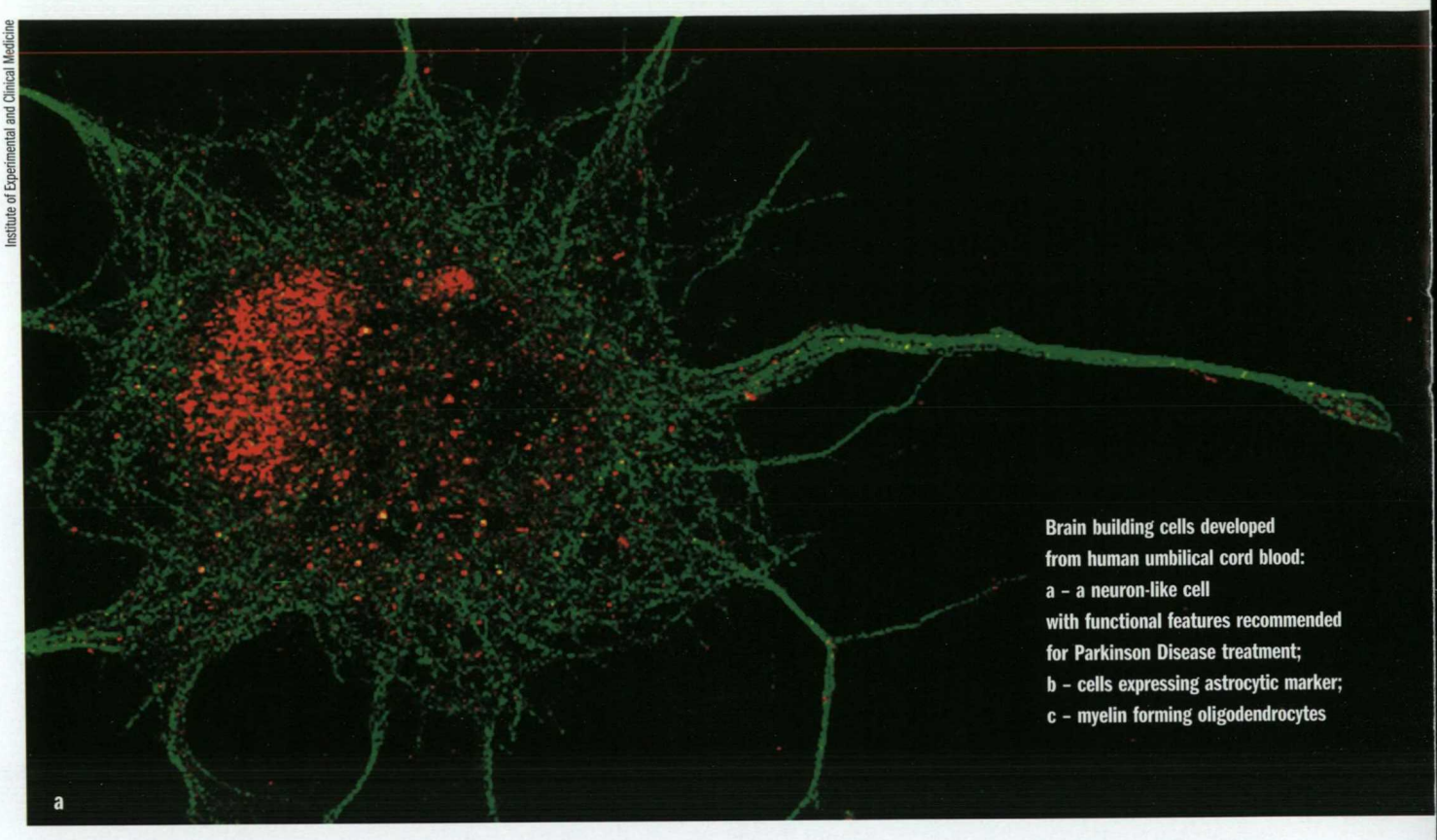

Brain building cells developed from human umbilical cord blood: a – a neuron-like cell with functional features recommended for Parkinson Disease treatment; b – cells expressing astrocytic marker; c – myelin forming oligodendrocytes

Cord Blood Cells for Brain Research

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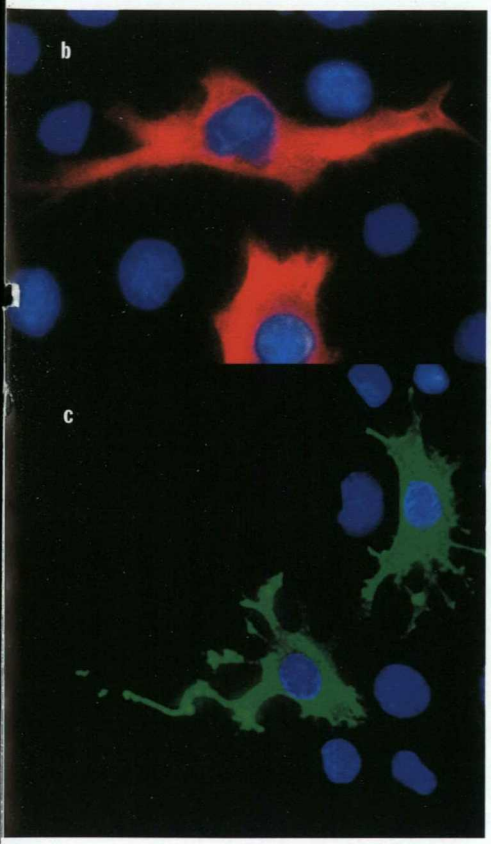
Umbilical cord blood doesn't become useless after a child is born. In the future it may serve as a source of "spare parts" for humans

Today there is no known cure for neurodegenerative diseases, such as Alzheimer's or Parkinson's. Severely damaged organs can only replaced by transplantation, but the number of donors will never be sufficient to satisfy the demand. Will this situation ever change? Stem cell research offers some hope. Stem cells have the ability to perpetuate themselves through self-renewal and to generate mature cells of a particular type of tissue through differentiation. In adult animals and humans, stem cells are present in various tissues including the bone marrow, skeletal muscle, liver, epidermis and retina. In these tissues stem cells are likely to replenish cells that are lost to physiological turnover, as well as to pathological conditions including injury and degenerative diseases. However, mature nervous tissue has long been considered incapable of cell renewal, especially in mammals.

Studies carried out in the last decade have revealed unexpected neurogenesis in adults and a surprising presence of stem cells in the mature mammalian central nervous system. These neural stem cells that persist into adulthood offer potential applications for the treatment of brain pathologies: from neurodegenerative disorders to post-traumatic lesions. However, the availability of human neural stem cells is limited because adult neural stem cells do not appear as an easily harvestable population, unless collected during necropsy. Nevertheless, there is another source for obtaining neural stem cells.

Somatic stem cells obtained from bone marrow and umbilical cord blood have recently been shown to differentiate into neural cells upon exposure to adequate stimuli. Human



umbilical cord blood represents a better source of naive, immature cells than bone marrow. Such blood is still of primitive ontogeny, little exposed to immunologic challenges, and can be noncontroversially and noninvasively obtained.

The far-off objective

Unfortunately, neural progenitor cells spontaneously differentiate in laboratory culture, and enter an irreversible growth arrest after a finite number of cell divisions. Such limited life-span is also a typical phenomenon for neural progenitors isolated from fetal or adult human brain tissue. One important reason for such difficulty may lie in the inherent asymmetric kinetics of how stem cells isolated from somatic tissues divide. A single stem cell which enters into asymmetric division gives rise to one stem cell and one proliferating progenitor cell, which further expands in response to intrinsic and environmental cues. Since divisions of progenitor cells lead to their differentiation, the expected fate of a cell culture with an asymmetric kinetics is senescence (aging). Thus the as-yet unresolved challenge is to suppress the asymmetric cell division typical of somatic stem cells, thereby enabling them to grow indefinitely (like embryonic stem cells) in the laboratory as well established and defined lines. Such lines are critically needed for further basic research as well as for expected therapeutic use.

Not 3 months, but 3 years

In vitro studies carried out by our research group have documented a previously unexpected plasticity of progenitor cells from human umbilical cord blood. These cells revealed characteristics similar to neural stem cells. After exposure to retinoic acid (RA) and brain derived nerve factor (BDNF), these progenitors differentiated into various classes of cells with molecular characteristics typical of neurons, astrocytes and oligodendrocytes.

In laboratory culture, progenitor cells obtained from umbilical cord blood form floating cell aggregates. They do differentiate toward cells activating genes and producing proteins typical of neural cells, but hardly expand and survive for longer than 3 months.

However, from the same neural progenitors, by employing prolonged exposure to serum, agents that induce mitosis and the further culture of progeny, we have succeeded in establishing a stable line of human neural stem cells derived from umbilical cord blood. These cells have been maintained as an undifferentiated proliferating cell line for about 3 years in continuous culture (more than 50 passages). The cells retain their normal chromosomal pattern and an unchanged capacity to proliferate and self-renew. In laboratory culture with no serum added, they form free-floating, undifferentiated spheres, resembling the spheres obtained from the human central nervous system. Further directed differentiation using substances inducing the "specialization" of neurons (RA, BDNF and dBcAMP) led to the production of more advanced neuronal markers: eventually even proteins typical of functional neurons but still displaying immature type of electrophysiologic features.

"Stemness" in genes

The molecular mechanisms underlying stem cells' developmental decisions, as to whether to proliferate or to differentiate into neurons, are not yet clear despite much recent attention. However, the essential role of at least three signaling pathways for stem cells' self-renewal capacity have been documented for brain and embryo derived neural stem cells.

In our study, we tested the activity of approximately 33,000 human genes of neural stem cells obtained form human umbilical cord blood and its reference population. More than 90% of stem- and neural-related genes were shown to be activated, as compared to the control cells, including many genes involved in "stemness"-related signaling pathways.

The results of our group indicate that the expansion of our neural stem cell line may be due to the activation of the so-called LIF, WNT and NOTCH tranduction pathways. It still remains to be investigated whether the molecules involved in these signaling pathways suppress the asymmetric divisions and thereby promote self-renewal and long term maintains of neural stem cells from umbilical cord blood. According to data on brain derived neural stem cells, which are in agreement with the above results, such a possibility may be seriously considered. ■

Further reading:

Bużańska L., Machaj E. K., Zablocka B., Pojda Z., Domańska-Janik K. (2002). Human cord blood - derived cells attain neuronal and glial features in vitro. *J Cell Sci.* 115, 2131-2138.

Corti S., Locatelli F., Strazzer S., Guglieri M., Comi G. P. (2003). Neuronal generation from somatic stem cells: current knowledge and perspectives on the treatment of acquired and degenerative central nervous system disorders. *Curr Gene Ther.* 3, 247-72.