

Long term cell survival and protein secretion from microencapsulated cells

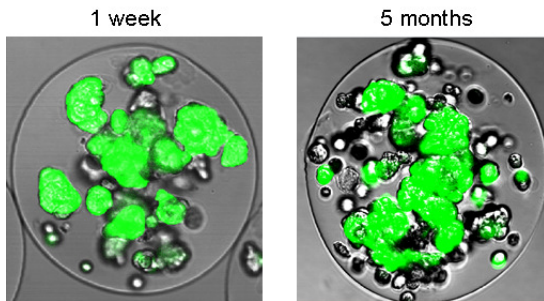
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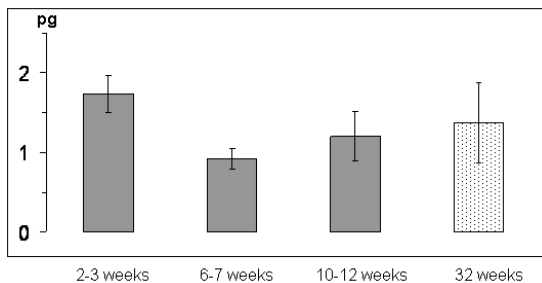
INTRODUCTION: One of the most important parameters for the use of encapsulated cells for medical treatment is their long term survival and production/secretion of the therapeutic peptide/protein of interest. Here we address cell survival *in vitro* and *in vivo* and secretion from alginate cell capsules. Capsule integrity and cell survival was further investigated *in vivo* after implantation into mouse brain.

Method: The producer cells (C2C12 or BHK) were harvested and mixed into a 2% sodium alginate (ultrapure, low viscosity, high guluronic acid content; PRONOVA (TM) UP LVG) saline solution (0.9% NaCl, 10mM MOPS, pH 7.4) to give a concentration of 50×10^6 cells/ml alginate solution. Beads were generated with an electrostatic bead generator. The gelling bath was composed of 0.1M CaCl₂ in 0.9% NaCl with 10mM MOPS, pH 7.4.

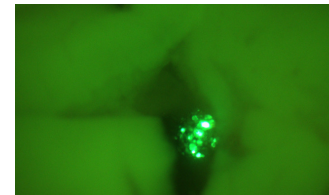
Abbreviations: NRP – Neuropilin, LRIG - leucine-rich repeats and immunoglobulin-like domains, CNTF – Ciliary Neurotrophic Factor.



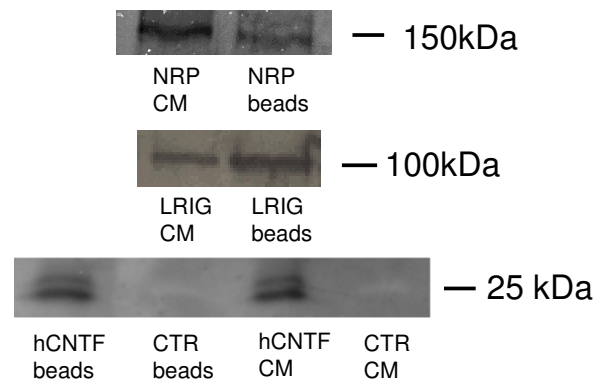
1) Examples of alginate encapsulated GFP expressing C2C12 cells after 1 week and 5 months in vitro. Living cells have been observed up to more than 1 year after encapsulation.



2) Measurement of hCNTF secretion over time in pg (Mean±SEM) per 24 hours per alginate bead using ELISA assay.



3) Viable BHK encapsulated cells expressing GFP 4 weeks after implantation into the ventricle of the mouse brain (2 week old beads were implanted)



4) Western blot of conditioned medium from encapsulated cells and cell cultures. Proteins/peptides with a wide range of molecular weight were secreted from the present alginate beads.

CONCLUSIONS: We have been able to show that cells can survive encapsulated for several months and even more than 1 year. Furthermore, we have shown that a large proteins and small peptide can be secreted, and that the secretion is stable for at least 8 months for the latter. We have also shown that encapsulated cells can survive for at least 4 weeks after implantation into the brain ventricles.