

How to refer to the cultures prepared by the PCN-CERVO:

Acknowledgments:

Any publication made pursuant to the use of the cultures prepared by the PCN-CERVO Platform shall acknowledge the “Neuronal Culture Platform of the CERVO Brain Research Centre”.

Neuronal culture method for rat HIPPOCAMPAL mixed cultures

Rat hippocampal neuronal cultures were performed as previously described [1]. Briefly, hippocampi were dissected out of postnatal rats (P0-P1) and cells were dissociated both enzymatically using 12 U/ml papain (Worthington Biochemical Corporation) and mechanically by trituration with a Pasteur pipette. After dissociation, cells were centrifuged and plated on poly-d-lysine coated coverslips (Neuvitro Corporation), and maintained in Neurobasal medium (Gibco) supplemented with B-27 (50:1), 50 U/ml penicillin/ 50 ug/ml streptomycin mixtures, 0.5 mM L-glutamax and 2% FBS-HI (Hyclone). To reduce the number of glial cells, 5 µM of cytosine arabinofuranoside (ARA-C; Sigma) was added on day 5 of the culture and cells were kept in culture until day 9 in the presence of ARA-C and serum. As of day 9, half of the growth medium was replaced with medium without ARA-C or serum twice a week. Neurons were cultured at 37°C in a humidified atmosphere of 95% air and 5% CO₂.

Neuronal culture method for rat CORTEX mixed cultures

Rat cortex neuronal cultures were performed as previously described [1]. Briefly, cortex were dissected out of postnatal rats (P0-P1) and cells were dissociated both enzymatically using 12 U/ml papain (Worthington Biochemical Corporation) and mechanically by trituration with a Pasteur pipette. After dissociation, cells were centrifuged and plated on poly-d-lysine coated coverslips (Neuvitro Corporation), and maintained in Neurobasal medium (Gibco) supplemented with B-27 (50:1), 50 U/ml penicillin/ 50 ug/ml streptomycin mixtures, 0.5 mM L-glutamax and 5% FBS-HI (Hyclone). To reduce the number of glial cells, 5 µM of cytosine arabinofuranoside (ARA-C; Sigma) was added on day 5 of the culture and cells were kept in culture until day 9 in the presence of ARA-C and serum. As of day 9, half of the growth medium was replaced with medium without ARA-C or serum twice a week. Neurons were cultured at 37°C in a humidified atmosphere of 95% air and 5% CO₂.

References:

1. Nault F., De Koninck P. (2009) Dissociated Hippocampal Cultures. In: Doering L. (eds) *Protocols for Neural Cell Culture*. Springer Protocols Handbooks. Humana Press, p. 137-159.