



NeuroKine Workshop – Miltenyi Biotec

Cell Separation

March 12-13, 2015



VENUE:

Miltenyi Biotec GmbH
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PROGRAM:

Thursday: 12 March, 2015

9:30	Transfer Haus Thal - Miltenyi Biotec
10:00	Welcome and Introduction
10:15-11:00	Talk: Introduction: Cell Separation and Cell Analysis (Martin Pyka, Miltenyi Biotec)
11:00-11:15	Coffee Break
11:15-12:00	Talk: Neuroscience at Miltenyi Biotec (Melanie Jungblut, Miltenyi Biotec)
12:00-13:00	Lunch Break
13:00-17:00	Practical Course

Isolation of Microglia from neonatal mouse brain tissue

Neonatal mouse brain tissue is dissociated into a single cell suspension by combining mechanical dissociation using the gentleMACS Octo Dissociator with an optimized enzymatic treatment using the Neural Tissue Dissociation Kit (P).

Then, the CD11b+ microglia are magnetically labeled with CD11b (Microglia) MicroBeads and isolated by the magnetic cell sorting (MACS) technique. Subsequently, purity and efficiency of the magnetic cell separation are analysed by immunofluorescent staining and flow cytometric analysis using CD11b specific antibodies and the MACSQuant Analyser.

19:00 Dinner Cologne

Friday: 13 March, 2015

9:00 Transfer Haus Thal - Miltenyi Biotec

9:30 Introduction

9:45-12:30 First Part: Practical Course

Isolation of Astrocytes from adult mouse brain tissue

In contrast to neonatal brain dissociation, adult brain dissociation is very demanding and requires sophisticated mechanical and enzymatic treatment to degrade the extracellular matrix and successfully disaggregate the tightly connected neural cells.

We have recently developed a new automated method for gentle dissociation of adult rodent brain tissue by combining mechanical dissociation using the gentleMACS Octo Dissociator (Miltenyi Biotec) with an optimized enzymatic treatment. The dissociation is followed by a novel protocol for removal of debris and erythrocytes, which is crucial for subsequent successful cell isolation.

12:30-13:30 Lunch Break

13:00-16:00 Second Part: Practical Course

Isolation of Astrocytes from adult mouse brain tissue

After tissue dissociation, astrocytes are isolated using the astrocyte specific Anti-ACSA-2 (astrocyte cell surface antigen-2) MicroBeads. Purity and efficiency of the magnetic cell separation are analysed

by immunofluorescent staining and flow cytometric analysis using Anti-ACSA-2 specific antibodies and the MACSQuant Analyser.

16:00-16:30 Wrap-up and departure