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Isolation of Satellite Glial Cells for High-Quality RNA purification

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Highlights

- An isolation protocol for highly pure Satellite Glial Cells (SGC) is described.
- The protocol relies on recognized intracellular SGC markers.
- High-quality RNA is isolated from these cells for down-stream applications.

Abstract

Background: Satellite glial cells (SGCs) envelope the neuronal somas in the dorsal root ganglia (DRG) and are believed to provide important neuronal support. Animal models of peripheral nerve injury, diabetes or chemotherapy all demonstrate activation of SGCs, suggesting important physiological roles for SGCs in various states of peripheral neuropathy. However, the biology of these glial cells is only poorly characterized under normal as well as pathological conditions due to suboptimal isolation methods.
New Method: The method presented here allows complete dissociation and isolation of highly pure SGCs from rat DRGs by fluorescence-activated cell sorting (FACS) using SGC-specific antibodies. The method further allows purification of high-quality RNA from the fixed and permeabilized cells.
Results: The purified RNA shows very little degradation, demonstrated by RNA integrity number (RIN) analysis with an average value of 8. The purified RNA, therefore, lends itself very well to downstream applications such as qPCR and transcriptome analysis.
Comparison with existing methods: Primary SGC cultures have previously been established for *in vitro* studies. Unfortunately, SGCs quickly change morphology and gene expression *in vitro,* complicating biologically meaningful interpretation of the obtained results. In contrast, this method allows the investigation of SGC gene regulation *in vivo* by isolation of high-quality RNA.
Conclusions: This method enables investigation of SGC transcriptional response *in vivo* by isolation and analysis of mRNA expression, allowing a more detailed investigation of SGC biology under normal as well as pathological conditions.

Keywords: Satellite Glial cells, cell purification, RNA isolation, FACS, peripheral neuropathy

1. Introduction

Satellite glial cells (SGCs) are flattened glial cells in the peripheral nervous system, enveloping each neuronal soma in the sensory ganglia and thereby supporting and protecting the sensory neurons (Pannese, 1981; Hanani, 2005). In the DRGs, the neuron and its enveloping SGCs create functional