User Guide



Quick-Neuron™ Sensory - Maintenance Medium

Catalog Number: SS-MM

Introduction

Quick-Neuron™ Sensory - Maintenance Medium may be used for the long-term maintenance of human pluripotent stem cell-derived sensory neurons following differentiation as outlined in the Quick-Neuron™ Sensory - mRNA Kit and Human iPSC-derived Neurons user guides. Quick-Neuron™ Sensory differentiated cell cultures display typical neurite outgrowth and express a variety of neuronal markers, such as tubulin beta 3 class III (TUBB3) and variety of sensory neuron markers such as peripherin (PRPH), islet-1 (ISL1), and brain-specific homeobox/POU domain protein 3A (BRN3A/POU4F1). When handled and maintained according to the instructions in this user guide, sensory neurons are viable long-term and are suitable for a variety of characterization and neurotoxicity assays.

Scale: The Quick-Neuron™ Sensory - Maintenance Medium provides sufficient medium for 4 wells of a

24-well plate, 1 well of a 6-well plate, or 16 wells of a 96-well plate for up to 2 weeks.

Related Products: Quick-Neuron™ Sensory - mRNA Kit, Catalog Number: SS-mRNA

Quick-Neuron™ Sensory - Human iPSC-derived Neurons, Catalog Number: SS-mRNA-CW

Contents

Upon receipt, store the reagents at the temperatures indicated in the table below. All reagents are shipped on dry ice.

Contents	Volume	Storage	Thaw
Component N	840 µl	-20°C or -80°C	On ice or 4°C
Component S1	22 µl	-20°C or -80°C	On ice or 4°C
Component P	50 μl	-20°C or -80°C	Room temperature

Condition of Use

This product is for research use only. It is not approved for use in humans or for therapeutic or diagnostic use.

Technical Support

For technical support please refer to the FAQ on our website.

You may also contact us at <u>cs@elixirgensci.com</u> or call +1 (443) 869-5420 (M-F 9am-5pm EST).

Required Consumables

Item	Vendor	Catalog Number
DMEM/F12	ThermoFisher	21331020
Neurobasal Medium	ThermoFisher	21103049
GlutaMAX	ThermoFisher	35050061
Penicillin-Streptomycin	ThermoFisher	15140122

Preparation

Medium N(S1)

- 1. Prepare Medium N(S1) using the reagents listed in the table below.
 - o Thaw each Component for 20-30 minutes at the temperature indicated in the "Contents" table on page 1.
 - Warm all other reagents at room temperature for 20-30 minutes.
 - o Tap each Component tube 3 times and then briefly spin all tubes down before use.
 - Keep Medium N(S1), and any subsequent media made with it, protected from light.
 - Store Medium N(S1) for up to 2 weeks at 4°C.
 - Leftover Components N and S1 can be discarded or saved at 4°C for up to two weeks.

Reagents	Volume
DMEM/F12	6 ml
Neurobasal Medium	6 ml
GlutaMAX	63 µl
Penicillin-Streptomycin (10000 units/ml; 100x)	126 µl
Component N	391 µl
Component S1	12.6 µl

First Week

- 1. Prepare Medium N(S1P) using the reagents listed in the table below.
 - o Thaw Component P for 20-30 minutes at the temperature indicated in the "Contents" table on page 1.
 - Warm all other reagents at room temperature for 20-30 minutes.
 - o Tap the Component P tube 3 times and then briefly spin it down before use.
 - Store Medium N(S1P) for up to 2 weeks at 4°C.
 - Leftover Component P can be saved at 4°C.

Reagents	Volume	
Medium N(S1)	5.2 ml	
Component P	5.2 µl	

2. Pipet out half of the old medium from each well and very slowly along the wall of the well, add room temperature Medium N(S1P) according to the following table.

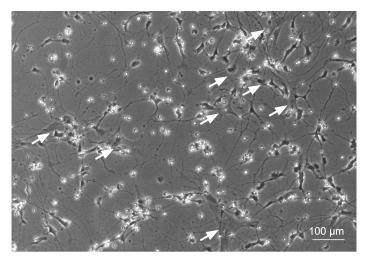
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	Rec	Required volume per well			
Reagents	6-well plate	24-well plate	96-well plate		
Medium N(S1P)	1 ml	400 µl	75 µl		

- 3. Incubate the cultures at 37°C, 5% CO₂ for 2 days.
- 4. Repeat Steps 2-3 every 2-3 days such as on Monday, Wednesday, and Friday for 1 week.

Second Week

Warm Medium N(S1) at room temperature for 20-30 minutes until it no longer feels cold.
Note: If there is an outgrowth of non-neuronal flat cells in the culture (as seen marked by arrows in the sample image below) users should continue using Medium N(S1P) in the second week, following the instructions to prepare Medium N(S1P) provided in the "First Week".



2. Pipet out most of the old medium, but not completely (i.e., just enough to cover the surface of the well), from each well and very slowly along the wall of the well, add Medium N(S1) according to the following table.

	Required volume per well		
Reagents	6-well plate	24-well plate	96-well plate
Medium N(S1)	2 ml	800 µl	150 µl

- 3. Incubate the cultures at 37°C, 5% CO₂ for 2 days.
- 4. For subsequent medium changes, pipet out half (see volumes in the table above) of the old medium from each well and replace with an equal volume of room temperature Medium N(S1).
- 5. Repeat Step 4 every 2-3 days such as on Monday, Wednesday, and Friday for 1 week.

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