



Instruction Card LabTIE OP50 V.2.1

for NGM agar and Liquid culture **Batch# 002**



 NGM Agar	How to use OP50 <i>C. elegans</i> nutrition for (NGM) agar Petri dish experiments. <i>Follow the instructions below by using a sterile technique.</i>	
Amount to dissolve	1)	Dissolve the content of 1 vial containing 0.90 gram in 25 mL Sterile dH ₂ O by vortexing the suspension thoroughly for at least 30 seconds. (equals 250 small and 100 medium Petri dishes to be seeded with $\pm 6 \times 10^{10}$ cells/mL).
Amount to seed on a Petri dish	2)	Apply 100 μ L on a small Petri dish (60 mm) Apply 250 μ L on a medium Petri dish (90 mm) <i>Tip: <u>Slightly</u> circle the plate to spread the OP50 <u>a little</u> to speed up the drying process. When spread to much, the lawn can become thin and single OP50 colonies can become visible.</i>
Evaporation of dH ₂ O	3)	Dry the Petri dish for 15-60 minutes in a laminar flow hood.
Seeding time	4)	<i>C. elegans</i> can be inoculated directly after the Sterile dH ₂ O has been evaporated Another option is to place the seeded Petri dish in an incubator for 12 hours at 37°C and inoculate afterwards.
 Liquid Culture	How to use OP50 <i>C. elegans</i> nutrition for 250 mL liquid culture experiments (S-medium). <i>Follow the instructions below by using a sterile technique.</i>	
Amount to dissolve	1)	Standard use: Dissolve 0.90 gram (1 Vial) in 250 mL S-medium (1 liquid culture) using a magnetic stirrer. This will result in a concentration of $\pm 6 \times 10^9$ cells/mL. For advised concentrations for <i>C. elegans</i> assays see other side: Optimal OP50 Concentration Chart V.2.1
Inoculation time	2)	Once the OP50 has been dissolved, <i>C. elegans</i> can be inoculated.
We recommend that you also view the instruction documentation & videos: www.labtie.com/op50 Always check our website www.labtie.com/op50 to see if this Instruction Card is the latest version or that it has been updated.		

OP50 Concentration Chart V.2.¹

for Liquid culture **Batch# 002**

Additional information on OP50 *C. elegans* nutrition for Liquid Culture experiments (S-medium)

Type of assay	Optimal OP50 CFU/mL		Amount of LabTIE OP50 to dissolve in 250 mL
Dietary restriction ^{[1][2]}	5x10 ¹¹ 5x10 ⁸	Ad libitium (Not advised to create a 250 mL liquid culture) Dietary Restriction	75 Gram 0.075 Gram
Micro Fluidics ^[3]	2.4x10 ⁹	Pharyngeal pumping	0.38 Gram
Life Cycle & population growth rate ^{[4][5]}	5x10 ⁹ <5x10 ⁹ >5x10 ¹⁰	Optimal concentration Larval mortality increases + decrease of fertility Lower life expectancy	0.75 Gram <0.75 Gram 7.5 Gram
Behavior assay ^[6]	1.5x10 ¹⁰		1.50 Gram
Survival assay ^[7]	1 - 4x10 ⁹		0.75 Gram

1 E.L Greer, *et al.* An AMPK-FOXO Pathway Mediates Longevity Induced by a Novel Method of Dietary Restriction in *C. elegans*, Current Biology, 2007, Volume 17, Issue 19, Pages 1646–1656

2 C. Heintz, *et al.* Splicing Factor 1 Modulates Dietary Restriction and TORC1 Pathway Longevity in *C. elegans*, Nature. 2017 Jan 5; 541(7635): 102–106.

3 K. Conery, E.coli induced pharyngeal pumping in *C. elegans*, NemaMetrix, May 31, 2016

4 T.E Johnson, *et al.* Genetic variants and mutations of *Caenorhabditis elegans* provide tools for dissecting the aging processes. Genetic effects on aging II. Edited by: Harrison DE. 1990, Caldwell, NJ: Telford, 101-127

5 D. Muschli, *et al.* Life cycle and population growth rate of *Caenorhabditis elegans* studied by a new method, *BMC Ecology*, 2009, 9:14

6 University of Tübingen, Worm Methods, Animal Evolutionary Ecology, Version 29/01/06

7 G.M Solis, M. Petrascheck. Measuring *Caenorhabditis elegans* life span in 96 well microtiter plates. J Vis Exp. 2011, 49:2496

Do you want to add your optimal OP50 concentration findings to this list? Send the publication to: info@labtie.com