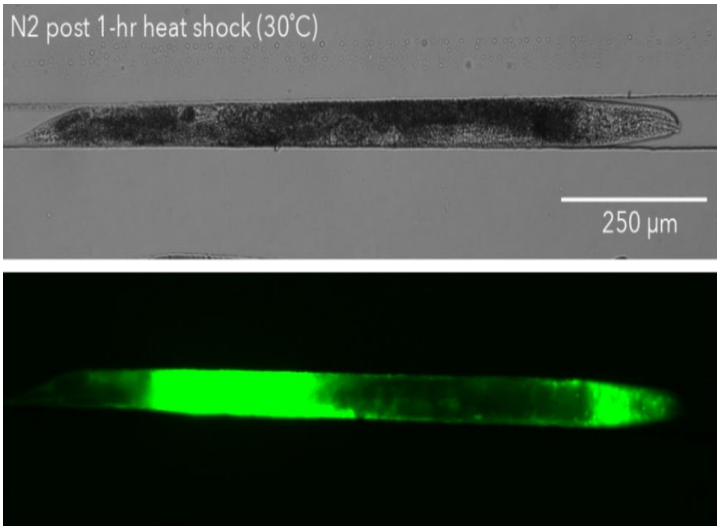
NemaMetrix

RediStain™ WormDye Live/Dead Green Reagent

SKU: DYE9438 SIZE: 500 UL (100 USES)

<-20°C		0.2mM
STORAGE UPON RECEIPT	**PROTECT FROM LIGHT**	**PACKAGED CONCENTRATION**
523nm	504nm	DMSO
EMISSION	**EXCITATION**	**SOLUTE**



Dead worm stained with RediStain™ WormDye Live/Dead Green reagent in the ScreenChip System.

Description

RediStain™ WormDye Live/Dead Green reagent is a dye that can indicate which worms in a population have died. RediStain™ WormDye Live/Dead Green reagent is chemically identical SYTOX™ Green nucleic acid stain (Molecular Probes).

Standard Worm Staining Protocol

1. PREPARE WORMS

1.1 Pipette 1 mL of M9 onto plate containing *C. elegans*. Swirl plate gently and transfer M9 and worms into EpiTube

1.2 Wash worms 3x.

1.2.1 Allow worms to settle or centrifuge at 6000 RPM for 2 minutes

1.2.2 Remove supernatant, leaving worms in pellet at bottom of the EpiTube

1.2.3 Add 1 mL M9

1.2.4 Repeat Steps 1.2.1-1.2.3 x 2

1.2.5 After the final rinse, remove as much liquid as possible, leaving only worm pellet

2. DILUTE

2.1 Dilute RediStain™ Live/Dead Green reagent immediately prior to use

2.1.1 Thaw at room temperature

2.1.2 Add 5 µL of RediStain™ Live/Dead Green reagent to 1 mL of M9

2.1.3 Pipette to mix

3. INCUBATE

3.1 Add diluted RediStain™ Live/Dead Green reagent to the freshly washed worms (step 1.3)

3.2 Incubate for 20-30 minutes at room temperature and away from light

4. WASH

4.1 Remove RediStain™ Live/Dead Green reagent, leaving worms in a pellet at the bottom of the EpiTube

4.2 Rinse in M9 buffer at least 3 times (see Step 1.2)*. Residual stain may obscure fluorescent signal.

**Alternatively, transfer worms to a fresh plate, and let crawl on a bacterial lawn for approximately 1 hour to destain. (If destaining worms on plate repeat steps 1-1.24, ending with rinsed worms in EpiTube.)*

5. USE

5.1 Image worms immediately.

Protocol for Simultaneous RediStain imaging and EPG in ScreenChip System

1. PREPARE WORMS

1.1 Pipette 1 mL of M9 onto plate containing *C. elegans*. Swirl plate gently and transfer M9 and worms into EpiTube

1.2 Wash worms 3x.

1.2.1 Allow worms to settle or centrifuge at 6000 RPM for 2 minutes

1.2.2 Remove supernatant, leaving worms in pellet at bottom of the EpiTube

1.2.3 Add 1 mL M9

1.2.4 Repeat Steps 1.2.1-1.2.3 x 2

1.2.5 After the final rinse, remove as much liquid as possible, leaving only worm pellet

2. PREPARE M9-5HT SOLUTION IMMEDIATELY PRIOR TO USE

2.1 Prepare 1000ul 10mM Serotonin in M9

2.2 Vortex or invert until solution is fully mixed

3. DILUTE

3.1 Add 5 µL of RediStain™ Live/Dead Green reagent to 1000uL of 10mM 5HT

3.2 Pipette to mix

4. INCUBATE

4.1 Add diluted RediStain™ Live/Dead Green reagent-10mM 5HT solution to the freshly washed worms and incubate for 20-30 minutes at room temperature in the dark

5. WASH

5.1 Remove RediStain™ Live/Dead Green reagent-10mM 5HT, leaving worms in a pellet at the bottom of the EpiTube

5.2 Rinse in freshly prepared 10mM 5HT in M9 buffer at least 3 times (see Step 1.2). Residual stain may obscure fluorescent signal.

6. USE

6.1 Load worms into ScreenChip, record EPG data, and image worms immediately

References

1. Sebastião Rodrigo Ferreira, Tiago Antônio Oliveira Mendes, Lilian Lacerda Bueno, Jackson Victor de Araújo, Daniella Castanheira Bartholomeu, and Ricardo Toshio Fujiwara, **"A New Methodology for Evaluation of Nematode Viability,"** *BioMed Research International*, vol. 2015, Article ID 879263, 7 pages, 2015. doi:10.1155/2015/879263
2. Gill MS, Olsen A, Sampayo JN, Lithgow GJ. (2003) **An automated high-throughput assay for survival of the nematode *Caenorhabditis elegans*.** *Free Radic Biol Med.* Sep 15;35(6):558-65.

About NemaMetrix

NemaMetrix Inc. specializes in developing and manufacturing devices, consumables, and software for automatic worm screening and phenotyping.

The company's mission is to enable scientists and researchers around the world to better understand human diseases and explore potential treatments for high-impact disorders such as Alzheimer's disease, and ALS (Lou Gehrig's Disease), and cardiac arrhythmias by offering a more affordable and rapid system that supplements the traditional mouse model. Please visit our website for the most up to date information.

Learn more at www.nemametrix.com/about-us

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