



JAN 25, 2024

Thioflavin T Assay

In 1 collection

Michael X. Henderson¹

¹Van Andel Institute



michael.henderson

ABSTRACT

This protocol details Thioflavin T assay.

ATTACHMENTS

[932-2408.docx](#)

GUIDELINES

Adapted from Alex Crowe, Jing Guo, Dustin Covell 032012 protocol, Mian Horvath Updates

MATERIALS

Consumables:

- 1 mM Thioflavin T in MilliQ
- PBS
- Equipment needed: Spectrophotometer
- Black 96 well plate

OPEN ACCESS



DOI:

dx.doi.org/10.17504/protocols.io.x54v9p4kqg3e/v1

Protocol Citation: Michael X. Henderson 2024. Thioflavin T Assay. [protocols.io](https://dx.doi.org/10.17504/protocols.io.x54v9p4kqg3e/v1) <https://dx.doi.org/10.17504/protocols.io.x54v9p4kqg3e/v1>

License: This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

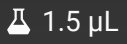
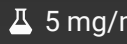

Protocol status: Working
We use this protocol and it's working

Created: Dec 20, 2023

Funders Acknowledgement:
Aligning Science Across
Parkinson's
Grant ID: ASAP-020616

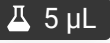
Thioflavin T Assay


1h

1 Resuspend fibril reaction. Fibrils will settle over time. Dilute  1.5 μ L of  5 mg/mL fibrillization reaction 1:50 with PBS (total  75 μ L).

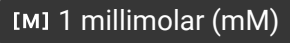



Note

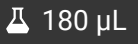
Ideally, the same dilution of monomer, PBS alone and a previous batch of PFFs should be run in parallel. Mouse PFFs have low ThT fluorescence. In this case  5 μ L PFFs can be diluted 1:10.

2 Assay each fibrillization in triplicate on the 96 well black assay plate. Dispense  20 μ L of diluted α -synuclein fibrils per well.



3 Dilute  1 millimolar (mM) Thioflavin T stock into PBS 1:1000 to obtain the required volume of Thioflavine T solution at a concentration of  1 micromolar (μ M) .



4 Dispense  180 μ L of  1 micromolar (μ M) Thioflavain T per well.



5 Maintain plate at  Room temperature for  01:00:00 in the dark.

1h



6 Read plate on a Spectrophotometer excitation 450 nm, emission 510 nm, cutoff 475 nm.