Synervia™

Human Total Tau ELISA Kit

Cat. No. SN-TTAU-ELISA-96 • 96-well (12 × 8 strips) • RUO

At-a-glance

Parameter	Value
Validated matrices	Serum, EDTA plasma, Li-heparin plasma, CSF
Assay format	Sandwich ELISA (capture + biotinylated detection + Streptavidin-HRP)
Readout	450 nm with 620–650 nm reference
Total assay time	≈ 4–5 hours (excluding plate prep)
Sample volume	100 µL per well (25 µL sample + 75 µL Sample Diluent option)

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Quick Start (Bench Card)

1. Bring all reagents and plate to RT (18–25 °C). Mix gently; avoid foaming.
2. Prepare standards (CAL A serially diluted) and controls in Sample Diluent (no sodium azide).
3. Optional: pre-wash plate 1× with 1× Wash Buffer.
4. Load 100 µL/well Standards, Controls, Samples (or 25 µL sample + 75 µL Sample Diluent).
5. Incubate 2 h at RT, 400–600 rpm (e.g., 500 rpm).
6. Wash 4× with ≥300 µL/well; tap-dry each time.
7. Add 100 µL/well biotinylated Detection Antibody; incubate 1 h at RT with shaking.
8. Wash 4×.
9. Add 100 µL/well Streptavidin-HRP (freshly diluted; no azide). Incubate 30 min at RT with shaking.
10. Final wash 5× with 30–60 s soak each wash; tap-dry.
11. Add 100 µL/well TMB; develop 10–20 min in dark (no shaking).
12. Add 100 µL/well Stop Solution (1 N H_2SO_4).
13. Read A450 with 620–650 nm reference within 15 min.

WARNING: Do NOT use sodium azide in any solution—HRP inhibition will cause signal loss. Use Sample Diluent (not Wash Buffer) for sample/standard/control dilutions.

Intended Use & Introduction

The Synervia™ Human Total Tau ELISA Kit is intended for the quantitative measurement of total Tau (t-Tau) in human serum, EDTA plasma, lithium-heparin plasma, and cerebrospinal fluid (CSF). Tau is a microtubule-associated protein involved in neuronal structure and stability; levels in biofluids may change with neuroaxonal injury and tauopathy. This kit is for Research Use Only (RUO); not for use in diagnostic procedures.

Principle of the Test

This sandwich ELISA uses a plate pre-coated with a monoclonal capture antibody specific for human Tau. Standards, controls, and diluted specimens are added; Tau binds to the immobilized antibody. After washing, an HRP-conjugated biotinylated detection antibody binds a distinct non-overlapping epitope, forming a capture–antigen–detection complex. Following washes, TMB substrate is added; HRP catalyzes color development proportional to bound Tau. The reaction is stopped with acid and read at 450 nm with 620–650 nm reference. Concentrations are interpolated from a 4-parameter logistic (4PL) standard curve ($1/y^2$ weighting recommended).

Kit Contents

Component	Quantity	Storage	Part #
Pre-coated 96-well microplate (12 × 8 strips)	1 plate	2–8 °C	TTAU-PLT
Tau Standard, CAL A (lyophilized)	1 vial	2–8 °C	TTAU-CAL-A
Calibrators CAL B–G (lyophilized or to prepare from CAL A)	6 vials/recipe	2–8 °C	TTAU-CAL-SET
Low/High Controls (lyophilized)	2 vials	2–8 °C	TTAU-CTRL-SET
Biotinylated Detection Antibody (concentrate)	1 vial	2–8 °C	TTAU-DET-BIO
Streptavidin-HRP (concentrate)	1 vial	2–8 °C	HRP-STREP
Sample Diluent (no azide)	1 bottle	2–8 °C	DIL-SAMP
Antibody/HRP Diluent (no azide)	1 bottle	2–8 °C	DIL-AB-HRP
Wash Buffer (×20 concentrate; no azide)	1 bottle	RT/2–8 °C	WASH-20X
TMB Substrate	1 bottle	2–8 °C, dark	TMB-SUB
Stop Solution (1 N H_2SO_4)	1 bottle	RT	STOP-SOL
Plate sealers, IFU	—	—	—

Materials Required But Not Provided

- Calibrated micropipettes and filtered tips; reservoirs; polypropylene tubes
- Plate washer (or multichannel pipette) and plate reader (450 nm with 620–650 nm reference)
- Timer; plate shaker (400–600 rpm); absorbent paper
- Deionized/distilled water (for Wash Buffer 1×)
- Personal protective equipment (PPE)

Storage & Stability

Store the kit at 2–8 °C unless otherwise stated. Protect TMB from light. Do not freeze HRP or TMB. Reconstituted standards/controls are stable per lot-specific instructions; aliquot and store at −20 °C or −80 °C as validated. Avoid repeated freeze–thaw cycles.

Specimen Collection & Handling

Validated matrices: serum, EDTA plasma, lithium-heparin plasma, CSF. Collect using standard procedures. For serum, allow clot 30–60 min. Centrifuge within 2 h at 1,300–2,000 × g for 10–15 min. Transfer supernatant to polypropylene tubes; aliquot immediately to avoid freeze–thaw. Short-term 2–8 °C ≤72 h; long-term −80 °C preferred. Limit freeze–thaw to ≤2 cycles. Avoid hemolyzed/lipemic/icteric samples where possible; clarify by high-speed spin if needed. Do not use tubes/buffers containing sodium azide (HRP inhibitor).

Reagent Preparation

- Wash Buffer (1×): dilute the 20× concentrate with deionized water; mix gently (no azide).
- Standards (CAL series): prepare serial dilutions in Sample Diluent using CAL A as the highest point; do not use Wash Buffer for dilution.
- Controls: reconstitute/dilute in Sample Diluent.
- Biotinylated Detection Antibody: dilute in Antibody Diluent per vial instructions.
- Streptavidin-HRP: dilute freshly in HRP Diluent immediately before use (discard unused).
- TMB and Stop Solution: ready-to-use; protect TMB from light.

Assay Procedure

14. Bring reagents/plate to RT (18–25 °C). Optional: pre-wash plate 1× with 1× Wash Buffer.
15. Add 100 µL/well Standards, Controls, Samples (or 25 µL sample + 75 µL Sample Diluent).
16. Incubate 2 h at RT on plate shaker 400–600 rpm (e.g., 500 rpm).
17. Wash 4× with ≥300 µL/well 1× Wash Buffer; tap-dry after each wash.
18. Add 100 µL/well biotinylated Detection Antibody; incubate 1 h at RT with shaking.
19. Wash 4×.
20. Add 100 µL/well Streptavidin-HRP (freshly diluted). Incubate 30 min at RT with shaking.
21. Final wash 5× with 30–60 s soak each wash; tap-dry.

22. Add 100 µL/well TMB substrate; incubate 10–20 min in the dark (no shaking).
23. Add 100 µL/well Stop Solution (1 N H_2SO_4).
24. Read A450 with 620–650 nm reference within 15 min of stopping.

Critical: Do NOT use sodium azide in any solution. Use Sample Diluent for all dilutions.

Calculations & Reporting

- Subtract blank (0 standard) from all readings.
- Fit a 4-parameter logistic (4PL) standard curve ($1/y^2$ weighting recommended).
- Interpolate concentrations of unknowns from the standard curve; multiply by dilution factor.
- Report results within the validated reportable range; flag out-of-range values for re-testing.

Quality Control

Criterion	Acceptance
Curve fit	4PL, $R^2 \geq 0.990$; back-cal %RE ±15% (±20% near LoQ)
Precision	Duplicate CV ≤10–12% for controls/samples
Controls	Low and High levels within established ranges
Background	Blank (0) ≤ specified OD; verify washer residuals

Performance Characteristics (to be established)

- Sensitivity: LoB/LoD/LoQ per CLSI EP17-A2.
- Precision: within-run, between-run, and total precision per CLSI EP05-A3.
- Linearity/Parallelism: serial dilution and spike-recovery studies.
- Specificity: cross-reactivity with related proteins; interference (HIL, biotin).
- Hook Effect: evaluate high-dose samples.
- Carryover: automated/manual wash evaluation.

Limitations & Warnings

- For Research Use Only. Not for use in diagnostic procedures.
- Use Sample Diluent for all dilutions; do not use Wash Buffer for dilution.
- Do not use any reagents containing sodium azide (HRP inhibitor).
- Matrix effects vary; verify recovery/parallelism in your lab.
- Results must be interpreted with appropriate controls and clinical context.

Troubleshooting

Issue	Recommended Action

High background	Extend final wash soaks; ensure no azide contamination; prepare fresh HRP; verify washer residuals.
Low signal	Confirm TMB and HRP are active; read within 15 min after Stop; check standard prep in Sample Diluent.
Poor precision	Use duplicates/triplicates; verify pipette calibration; ensure uniform shaking and timing.
Nonlinear curve	Check standard dilutions; mix thoroughly; avoid edge effects; confirm 4PL fit with $1/y^2$ weighting.

References

- Lachno DR, et al. J Alzheimers Dis. 2011;26(3):531–541.
- Olsson B, et al. Lancet Neurol. 2016;15(7):673–684.
- Blennow K, Zetterberg H. Nat Rev Neurol. 2010;6(3):131–144.
- Skillbäck T, et al. Brain. 2015;138(9):2716–2731.

Manufacturer & Document Control

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Revision History: Rev A — Initial release.