

## Supporting Information

# **A Field-Deployable Diagnostic Assay for the Visual Detection of Misfolded Prions**

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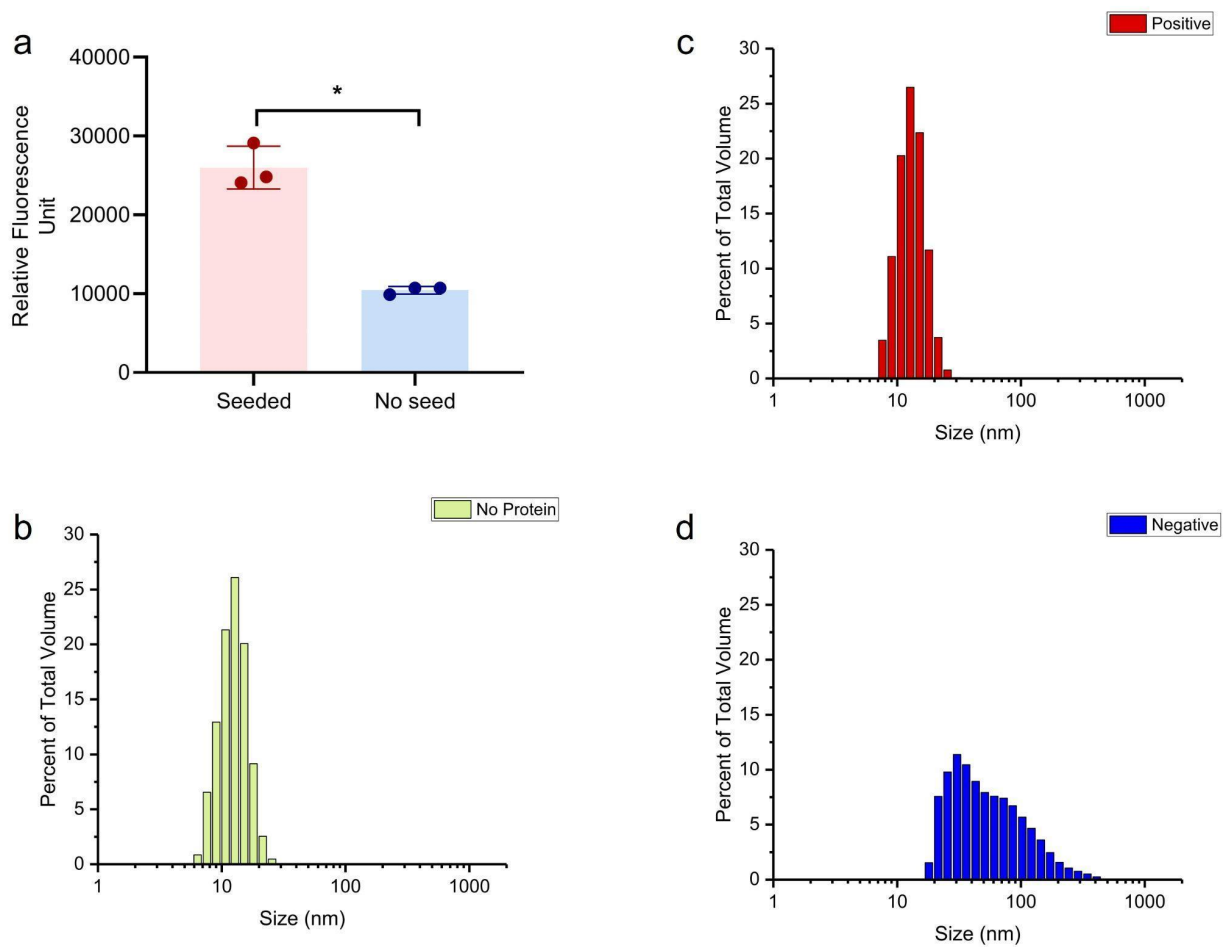
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## Supplemental Results



**Supplementary Figure 1.** ThT fluorescence, light absorbance, and average particle size measurements for native and misfolded recombinant hamster prion protein. **a.** The relative fluorescence units (ThT fluorescence) of post QuIC solutions containing misfolded protein seeds and solutions without misfolded protein seeds. **b.** Distribution of DLS readings of AuNP solution with no protein added. Reported as percent of total volume of AuNPs present. **c.** Distribution of DLS readings of AuNP solution with misfolded (positive) rHaPrP. **d.** Distribution of DLS readings of AuNP solution with native (negative) rHaPrP. \*, p-value < 0.05, error bars show standard deviation.

**Supplementary Table 1. Metadata of Examined Wild White-Tailed Deer RPLN.**

MNPRO = Minnesota Center for Prion Research and Outreach. RPLN = medial retropharyngeal lymph node. Official CWD test result indicates the RPLN was independently examined using ELISA and/or IHC testing at Colorado State University (see Schwabenlander et al. 2021, citation 26 in main text). ND=Not Detected.

<b>Animal MNPRO ID</b>	<b>Age</b>	<b>Sex</b>	<b>Official CWD Test Result</b>	<b>RT-QuIC Result</b>	<b>MN-QuIC Results</b>
166	Adult	Female	Positive	Positive	Positive
250	Adult	Male	Positive	Positive	Positive
360	Adult	Female	Positive	Positive	Positive
508	Adult	Male	Positive	Positive	Positive
922	Adult	Female	ND	ND	ND
924	Adult	Female	ND	ND	ND
925	Fawn	Female	ND	ND	ND
926	Fawn	Female	ND	ND	ND

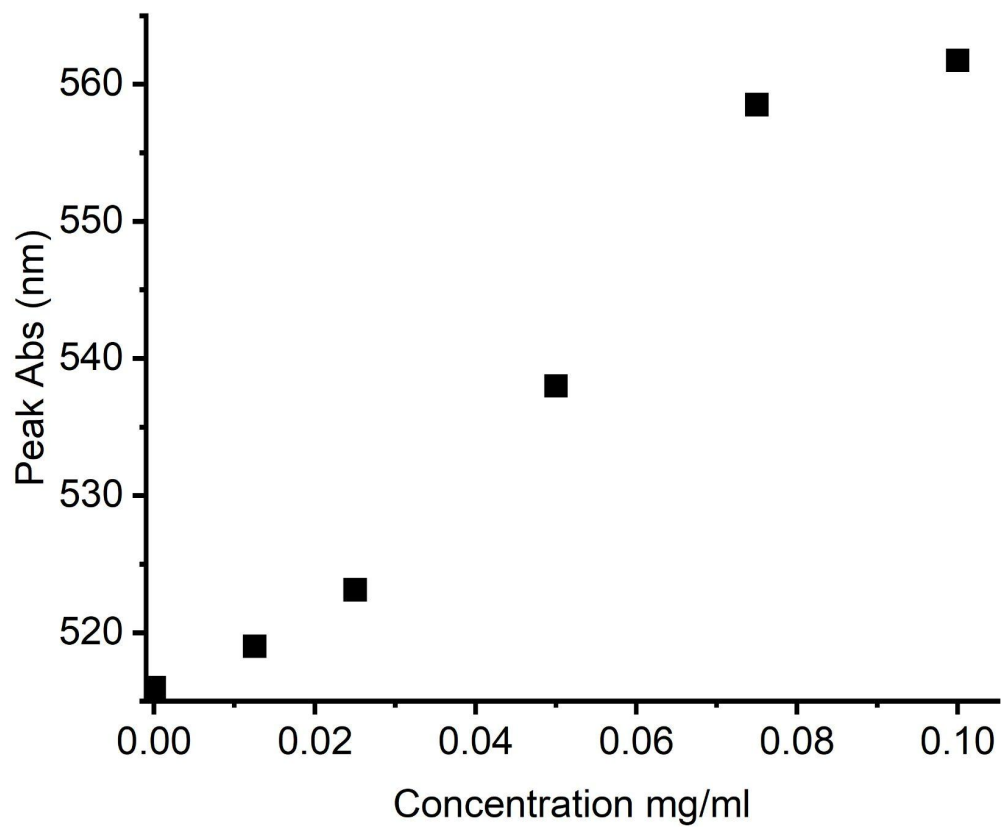
**Supplementary Table 2. Metadata of Examined Wild White-Tailed Deer in Field Test.** RPLN=medial retropharyngeal lymph node. Official CWD test result indicates the RPLN was independently examined using ELISA and/or IHC testing at Colorado State University (see Schwabenlander et al. 2021, citation 26 in main text). ND=Not Detected

<b>Animal ID</b>	<b>Age</b>	<b>Sex</b>	<b>Official CWD Test Results for RPLN</b>	<b>MN-QuIC Results</b>
197190	Female	Yearling	Positive	Positive
197270	Female	Fawn	ND	ND
197273	Female	Adult	ND	ND
197264	Female	Adult	ND	ND
197266	Female	Adult	ND	ND
197265	Female	Adult	ND	ND
197267	Male	Fawn	ND	ND
197268	Female	Fawn	ND	ND
197269	Female	Adult	ND	ND
197271	Female	Adult	ND	ND
197272	Male	Fawn	ND	ND
197225	Male	Yearling	Positive	Positive
197065	Male	Adult	Positive	Positive

**Supplementary Table 3. Metadata of Examined Wild White-Tailed Deer Tonsils.**

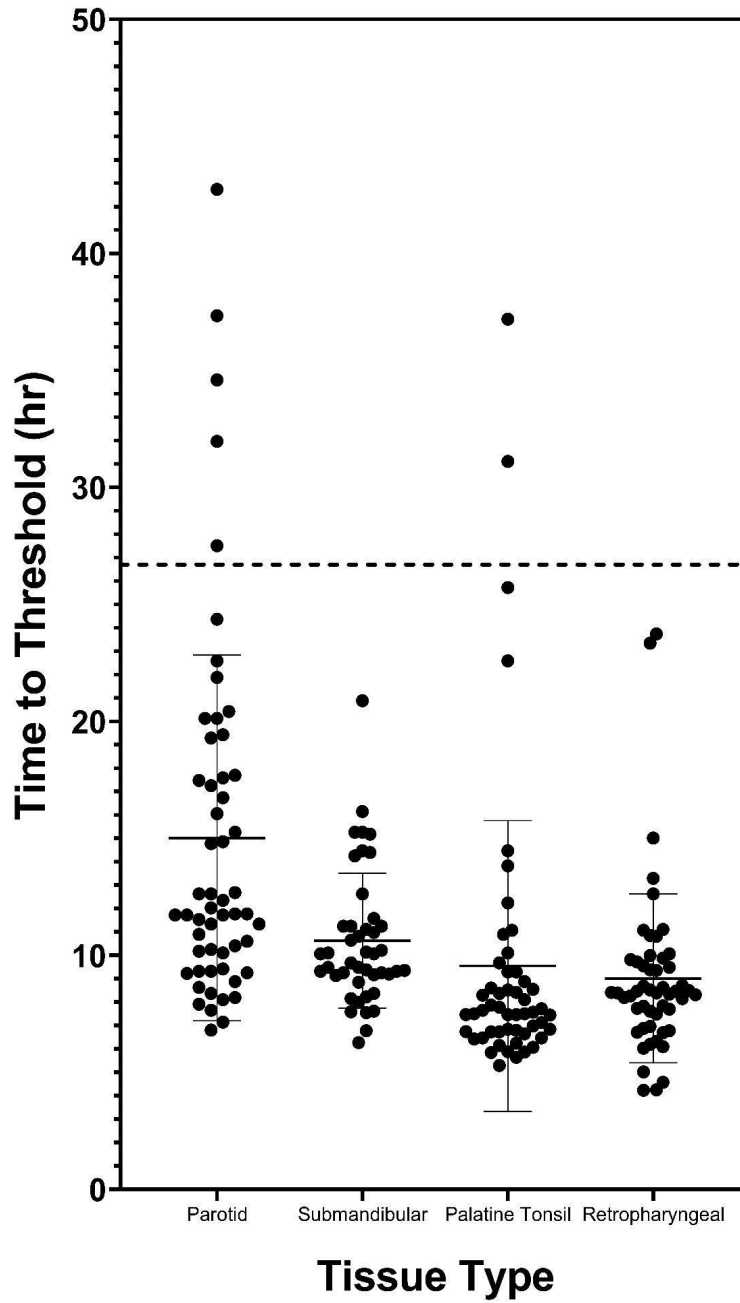
MNPRO = Minnesota Center for Prion Research and Outreach. RPLN=medial retropharyngeal lymph node. Official CWD test result indicates the RPLN was independently examined using ELISA and/or IHC testing at Colorado State University (see Schwabenlander et al. 2021, citation 26 in main text). ND=Not Detected

<b>Process ID</b>	<b>Animal MNPRO ID</b>	<b>Age</b>	<b>Sex</b>	<b>Official CWD Test Results for RPLN</b>	<b>RT-QuIC Results for Tonsil</b>	<b>MN-QuIC Results</b>
000734	000166	Adult	Female	Positive	Positive	Positive
000735	000197	Yearling	Male	Positive	Positive	Positive
000736	000250	Adult	Male	Positive	Positive	Positive
000737	000333	Adult	Female	Positive	Positive	Positive
000738	000360	Adult	Female	Positive	Positive	Positive
000739	000363	Adult	Female	Positive	Positive	Positive
000740	000376	Adult	Female	Positive	Positive	Positive
000741	000384	Adult	Female	Positive	Positive	Positive
000742	000498	Yearling	Male	ND	ND	ND
000743	000508	Adult	Male	Positive	Positive	Positive
000744	000735	Adult	Female	Positive	Positive	Positive
000745	000065	Adult	Female	ND	ND	ND
000746	000067	Adult	Female	ND	ND	ND
000747	000091	Adult	Female	ND	ND	ND
000748	000093	Adult	Female	ND	ND	ND
000749	000230	Fawn	Female	ND	ND	ND
000750	000232	Adult	Female	ND	ND	ND
000751	000283	Fawn	Male	ND	ND	ND
000752	000415	Fawn	Female	ND	ND	ND
000753	000431	Adult	Male	ND	ND	ND



**Supplementary Figure 2.** Wavelength of peak absorbance of AuNPs spiked with various concentrations of native HaPrP substrate in standard mastermix buffer.

### Time to Threshold for 4 Types of Lymph Nodes



**Supplementary Figure 3:** Prion seeding activity as shown by time (in hours) to Thioflavin-T fluorescence threshold via RT-QuIC reactions for various CWD positive white-tailed deer lymph tissues. Dashed line identifies a 24hr threshold used for MN-QuIC analyses of retropharyngeal lymph nodes.