

Biophysical studies suggest a new structural arrangement of crotoxin and provide insights into its toxic mechanism

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Supplementary material

Supplementary Fig. 1. SDS-PAGE (8-25% Phast gels) of CB after cross-linking. Glutaraldehyde concentrations are indicated at the top and molecular markers (kDa) on the left side of the figure. A and B show different preparations of CB and C shows CTX.

Supplementary Fig. 2. Autocorrelation function and regularization fit based on CONTIN algorithm of (A) CTX; (B) CA; (C) CB and (D) reconstituted CTX by mixing isolated CA and CB subunits at a 1:1 molecular ratio.

Supplementary Fig. 3. Linear regression curves of PLA₂ activity of native (white circles) and reconstituted (by mixing CA and CB isolated subunits) CTX (black circles). The regression curves were $3.895 \cdot X + 1.651$ for native CTX with R squared of 0.9843 and $3.887 \cdot X + 1.783$ for reconstituted CTX with R squared of 0.9146. A pooled slope of 3.8905 was generated when both activity curves were analyzed together.

Supplementary Fig. 4. Guinier plot and radius of gyration (R_g) calculated by Guinier analysis from small angle x-ray scattering data on (A) CA; (B) CB and; (C) CTX.

Supplementary Fig. 5. Far-UV circular dichroism spectra of CTX (blue line), CA (black line) and CB (red line).

Supplementary Table 1. Mean lifetimes (ns) of total tryptophan fluorescence emission at different wavelengths of CA, CB and CTX obtained by time-resolved spectroscopy fluorescence.

Glutaraldehyde [mM]

M

0

0.5

0

0.2

0.5

0

0.5

1.0

97.0

66.0

45.0

30.0

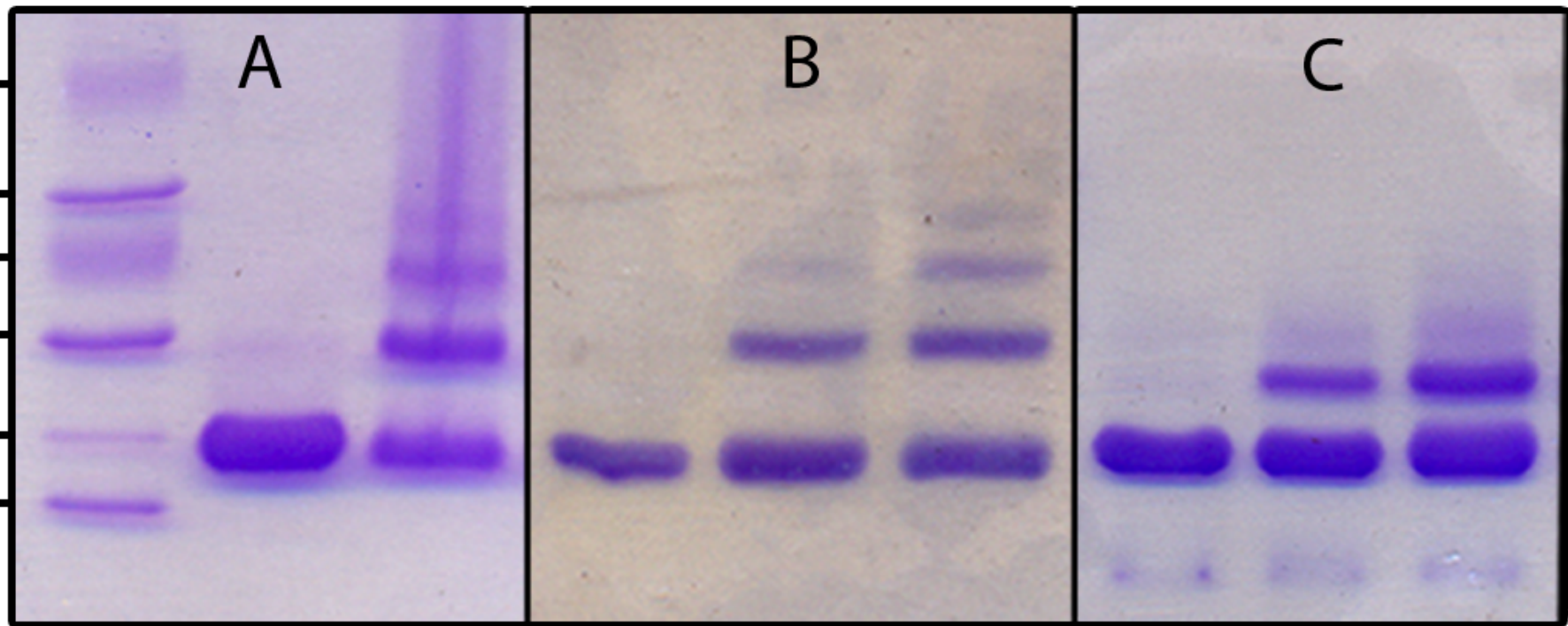
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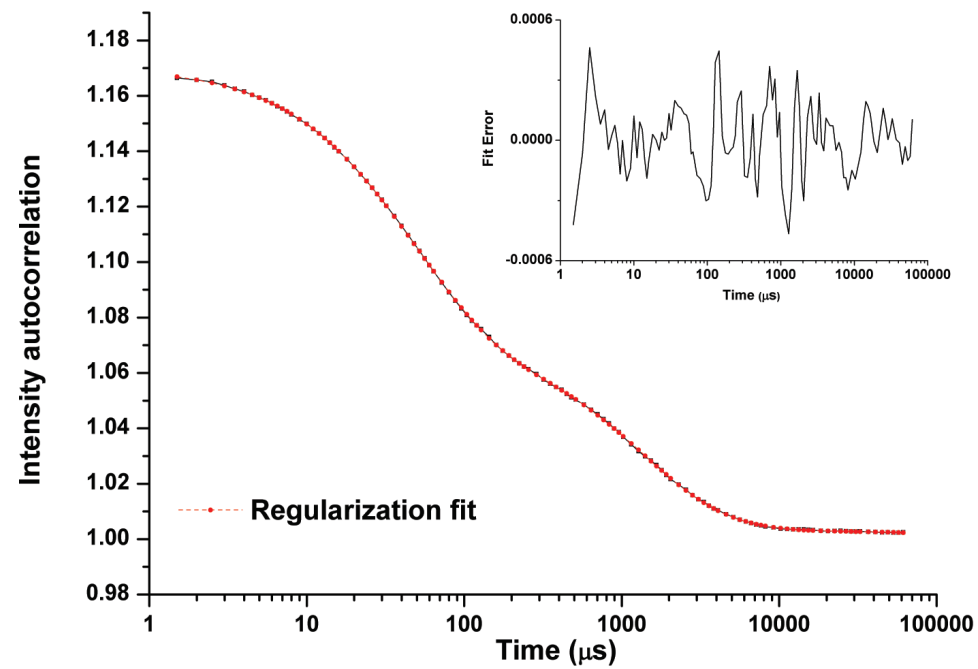
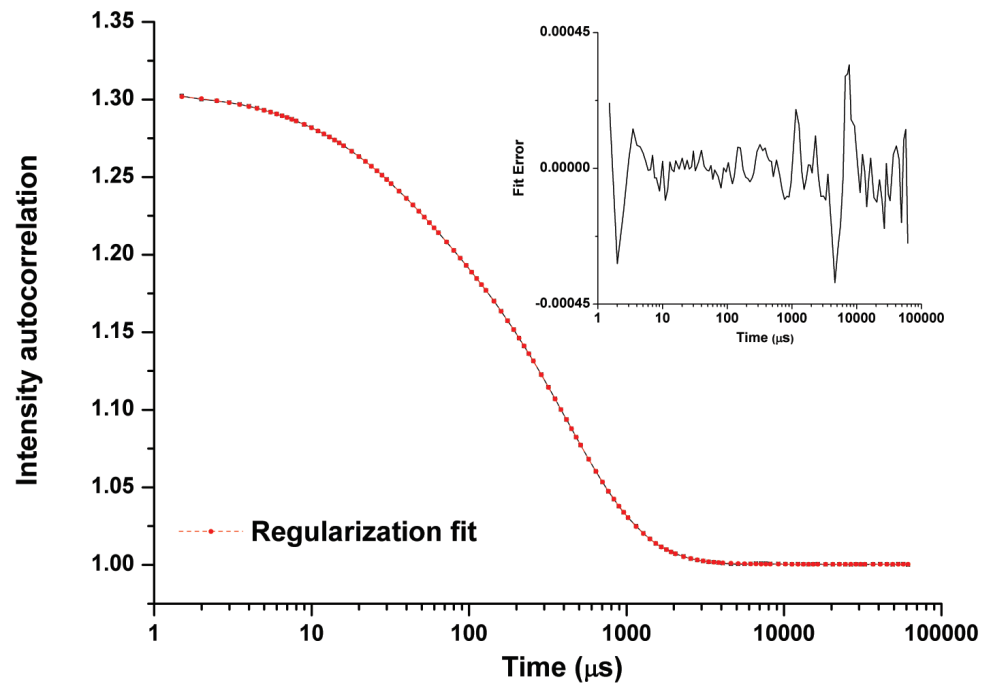
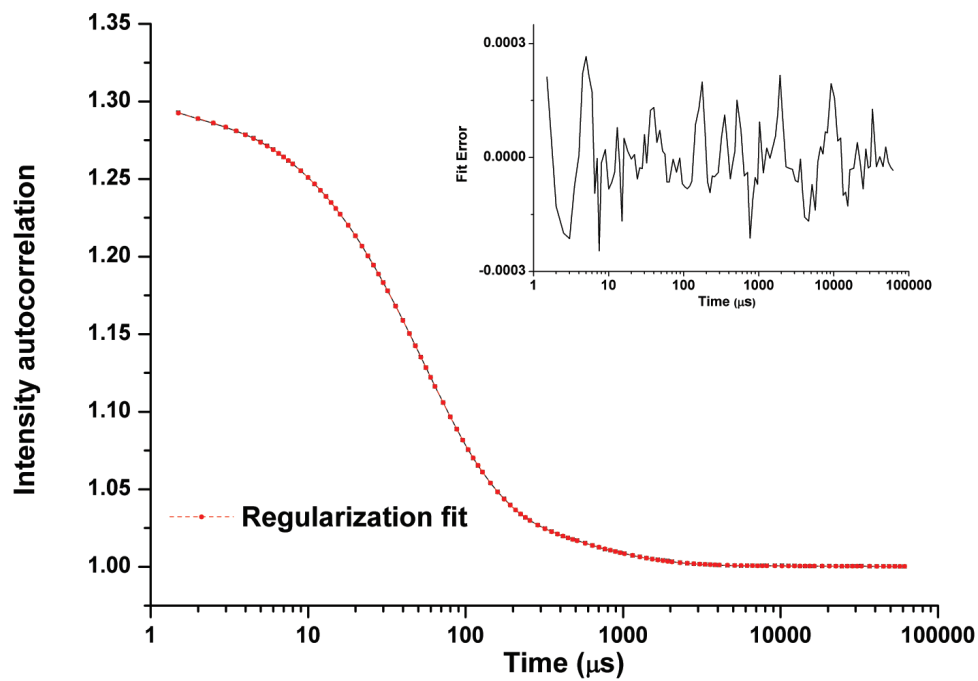
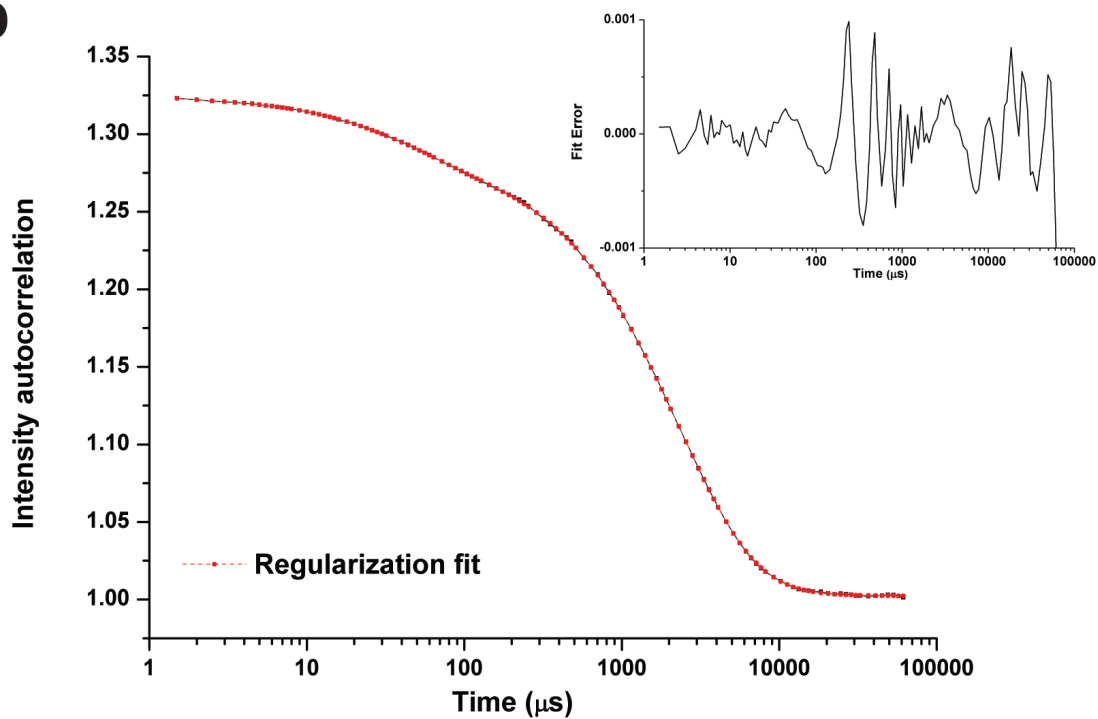
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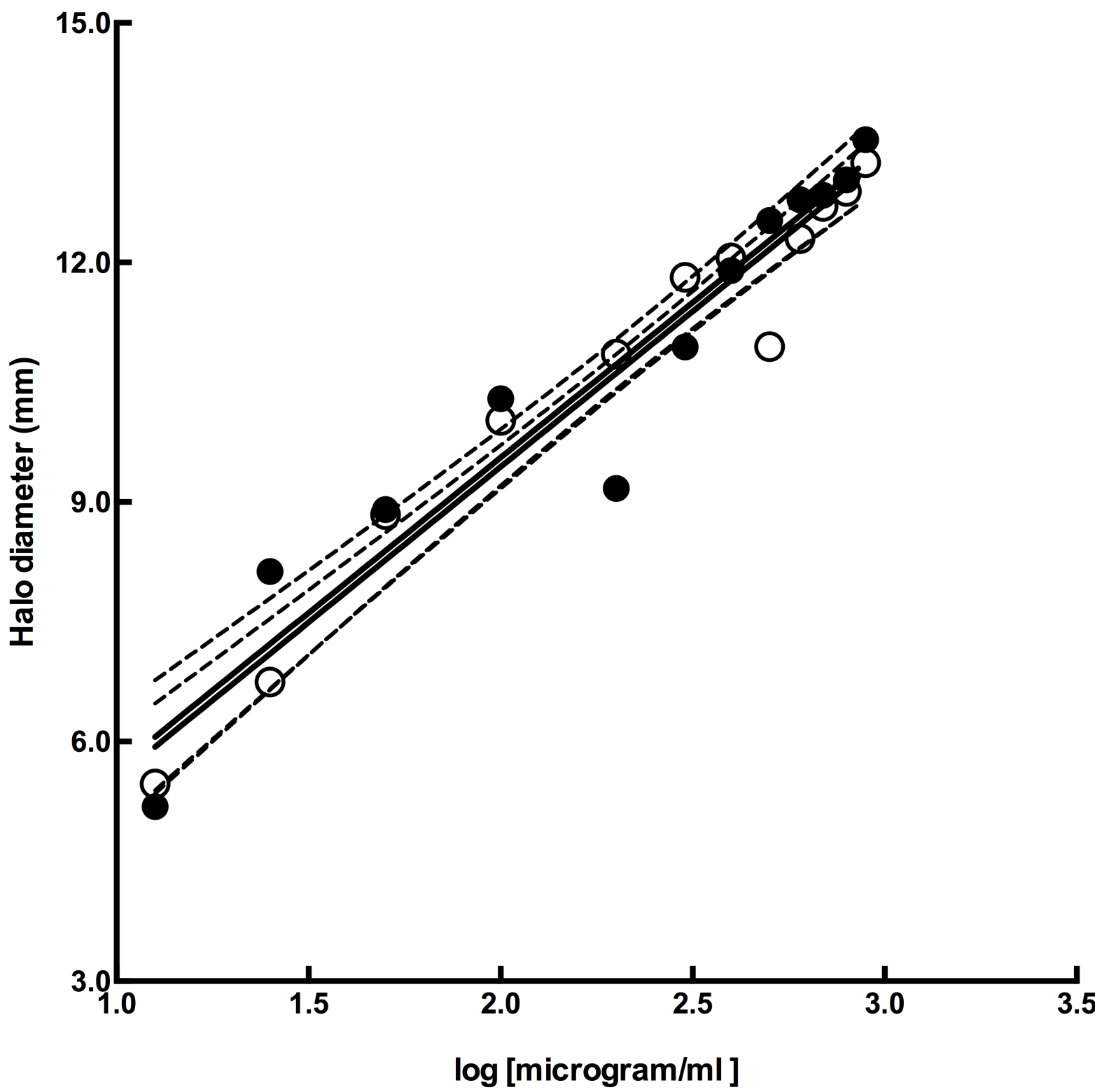
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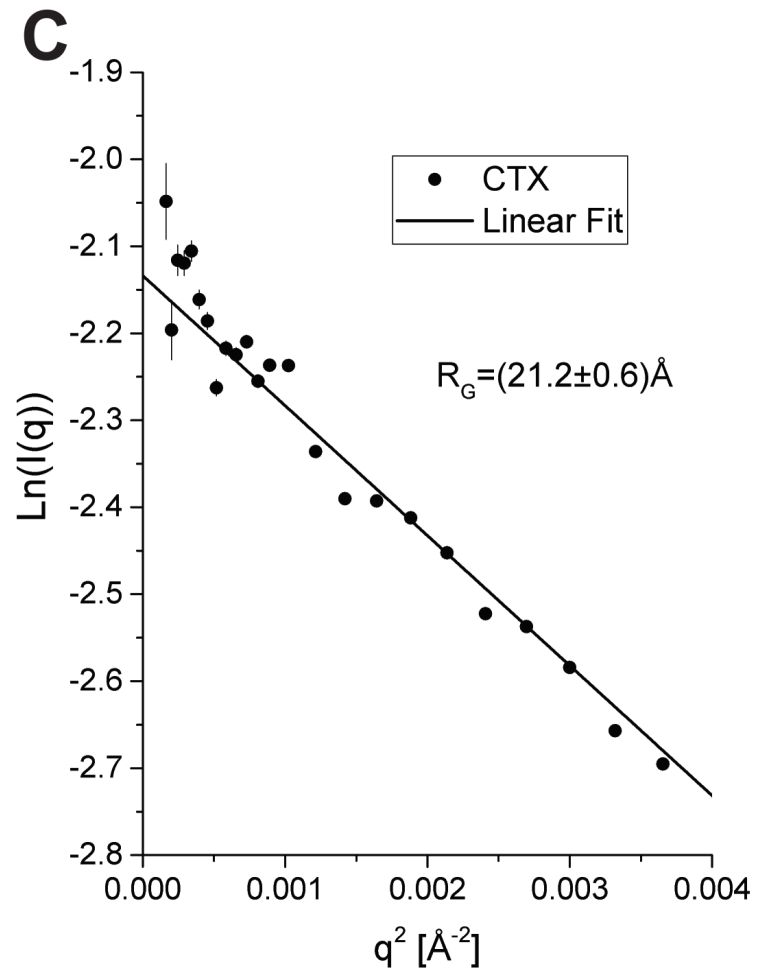
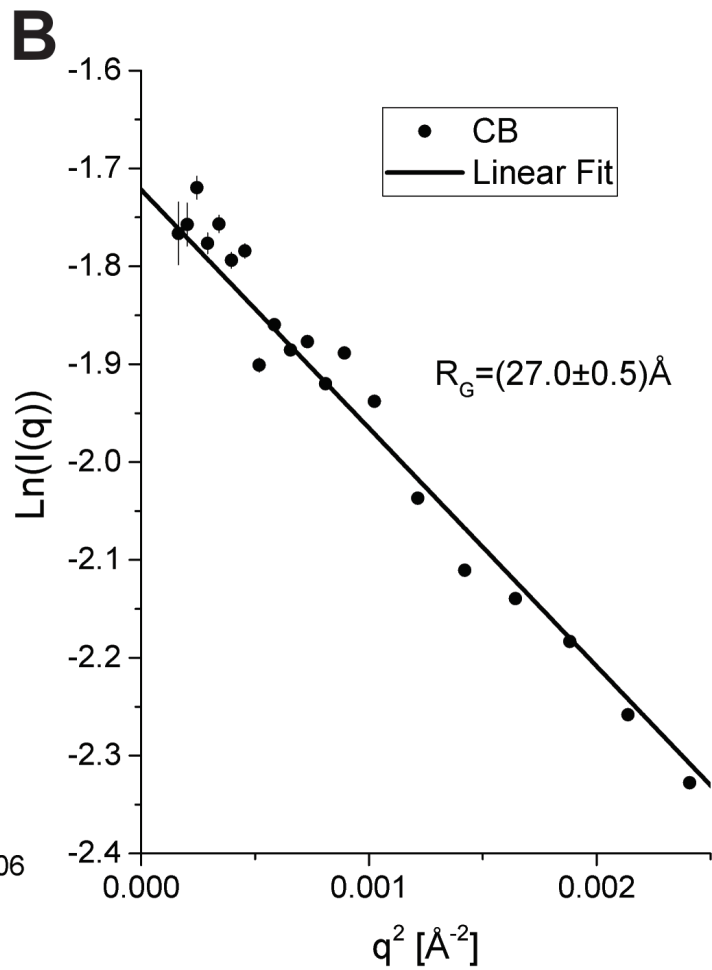
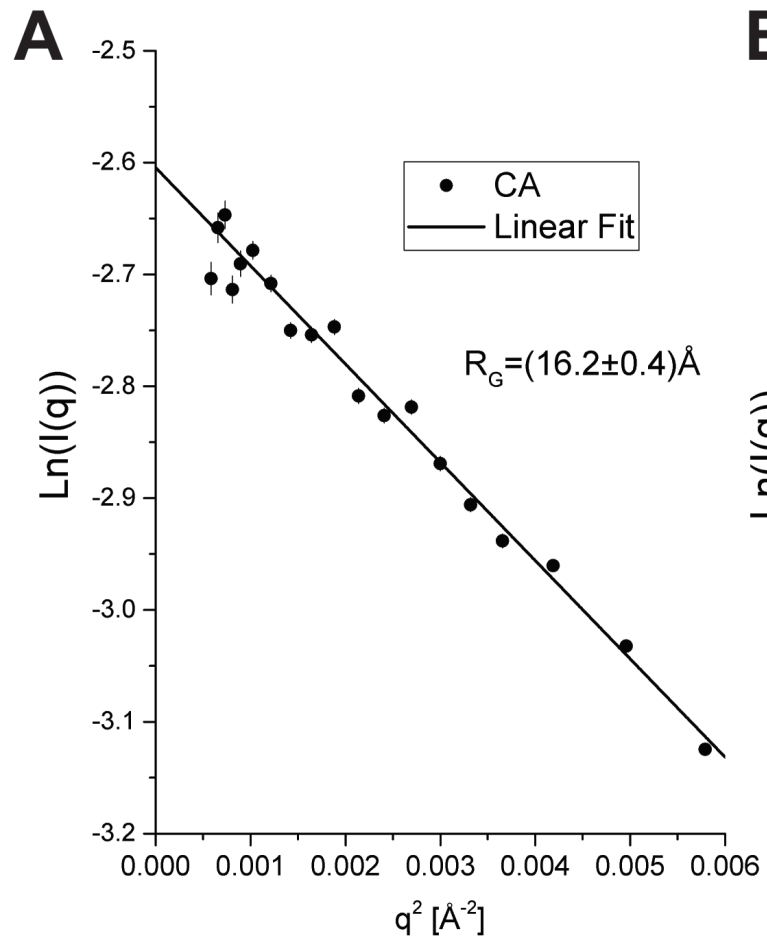
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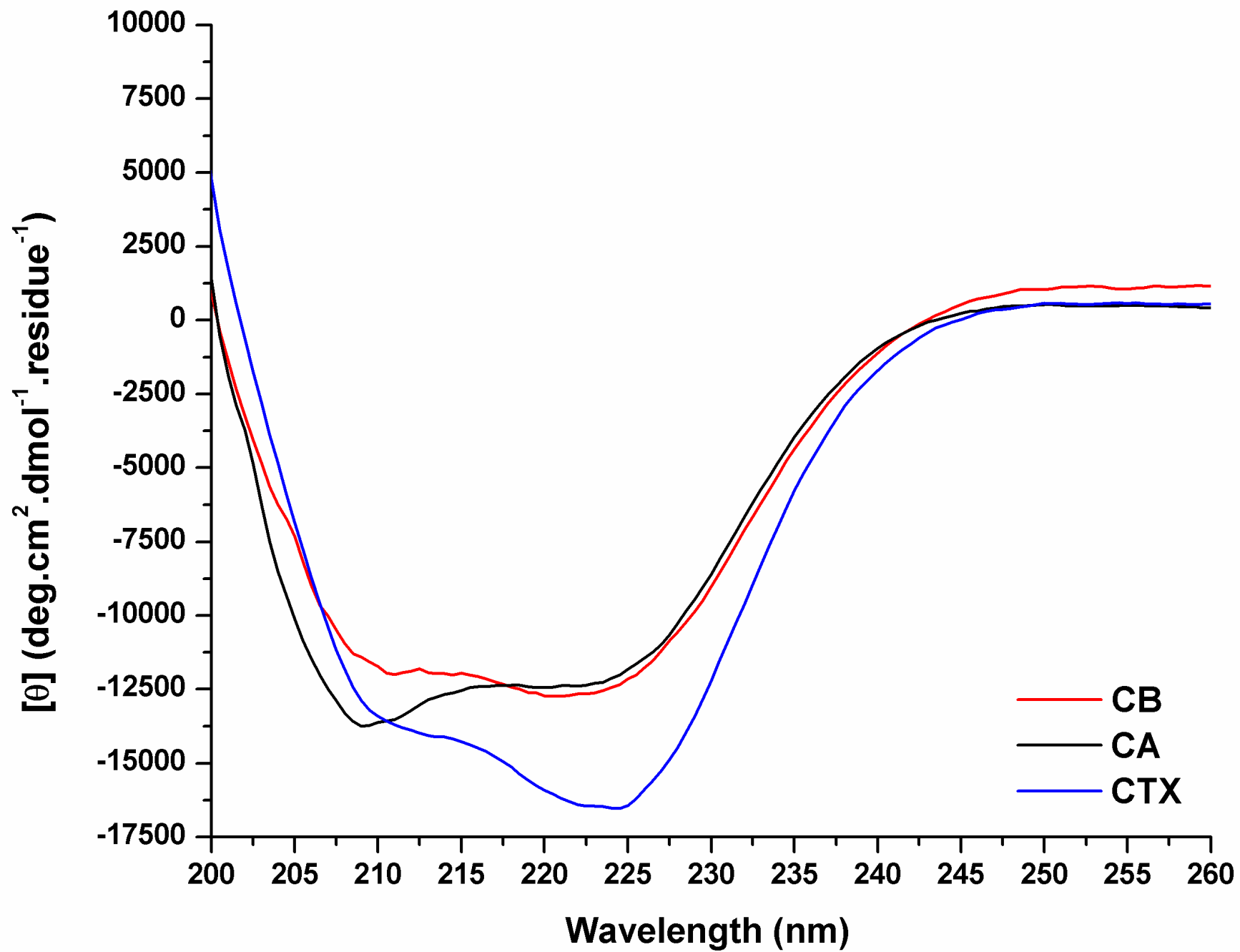
C



A**B****C****D**







Supplementary Table 1

λ_{em}	CA	CB	CTX
335	2.70	3.33	1.73
340	2.74	3.35	1.85
345	2.75	3.45	1.90
350	2.77	3.46	1.99
355	2.82	3.54	2.03
360	2.85	3.56	2.13
365	2.88	3.58	2.13