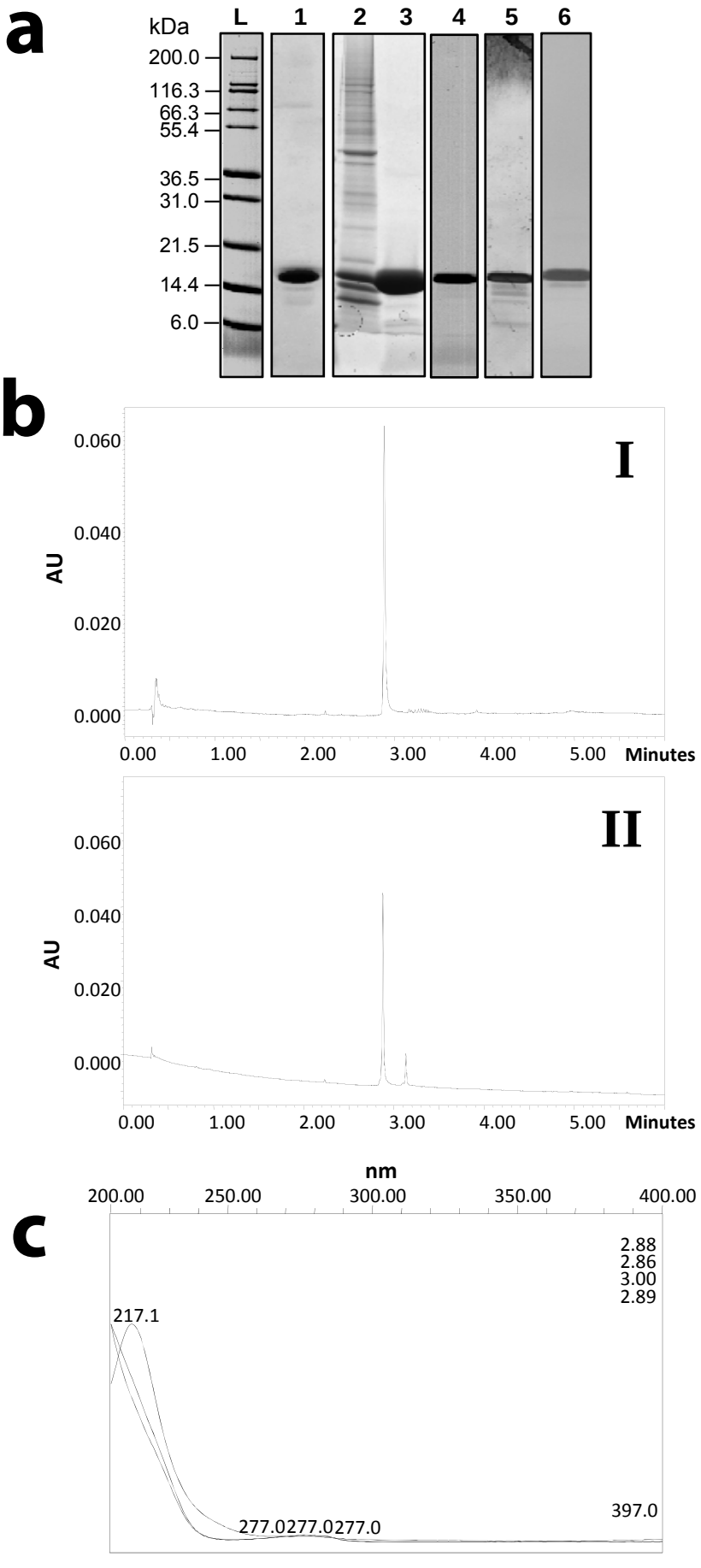
Figure S1


a
kDa
L
1
2
3
4
5
6
200.0
116.3
66.3
55.4
36.5
31.0
21.5
14.4
6.0
b
I
II
AU
0.060
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1.00
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Minutes
c
nm
200.00
250.00
300.00
350.00
400.00
217.1
2.88
2.86
3.00
2.89
277.0277.0277.0
397.0

Figure S1. SDS-PAGE, Immunoblot, and RP-UPLC Analysis of purified recombinant AnAPN1 produced in *E. coli.* (a) Solubilized inclusion bodies (Lane 1), Ni-IMAC flow through (Lane 2), and eluate using 500 mM Imidazole (Lane 3) were run on two non-reduced 14% Tris-Glycine gels (Invitrogen). L, Mark 12 protein ladder, kDa (Invitrogen), is shown on the left. The final purified material after IEX purification and dialysis (2 µg) was analyzed by SDS-PAGE followed by Coomassie blue staining (lane 4), silver staining (lane 5) and also detected by rabbit antibodies against AnAPN1 (Lane 6). (b) Reverse phase chromatography analysis of AnAPN1. Final purified recombinant protein (Panel I), in addition to an in-process sample during purification (Panel II) were analyzed. (c) Peak purity using 3 wavelength passes is shown for the main peak (2.878 minutes). Note: Yield was determined during in-process analysis by analyzing protein bands from SDS-PAGE with a GS-800 self-calibrating densitometer. The AnAPN1 eluate from the Ni column and BSA, a known standard, were run on a reduced 14% Tris-Glycine gel (Invitrogen) and stained with Coomassie Brilliant Blue. Final material was analyzed by A280nm and calculated using the extinction coefficient for yield. The LAL endotoxin test (0.25 EU/mL Gel Lot) of the research grade AnAPN1 in 15% sucrose, 10 mM Tris, 0.2% Tween-80, pH 7.8 (95.2% purity) was < 0.25 EU/mL.