Fluorescence Anisotropy Titration
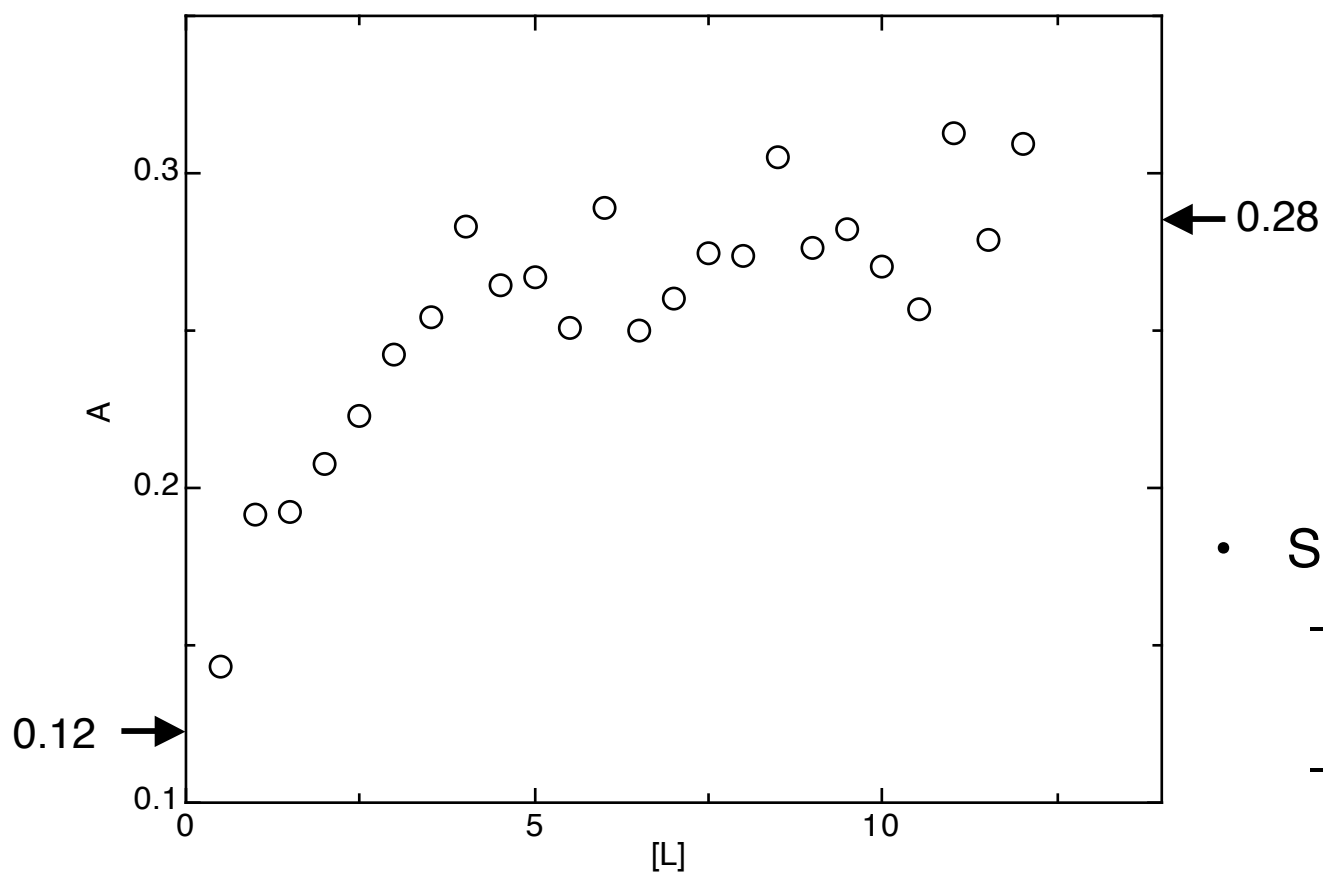
Scatchard Analysis
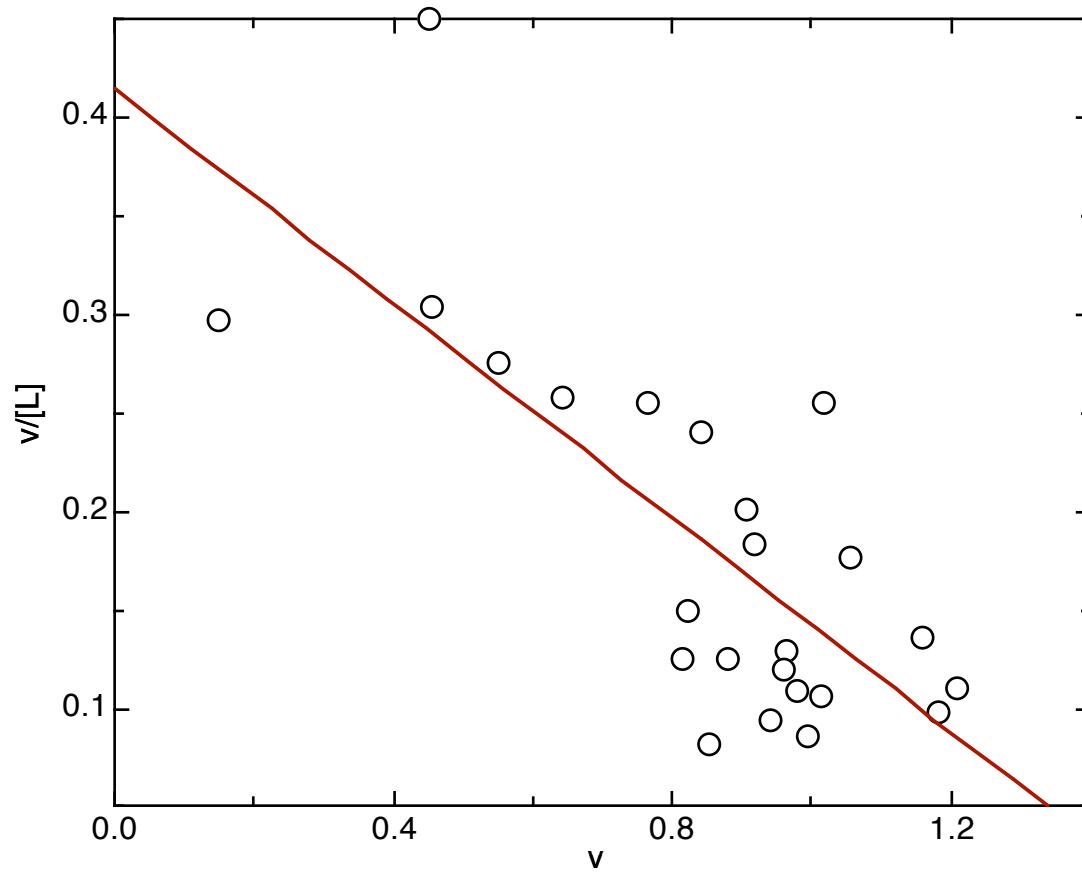
Slope = -0.27 ± 0.05
Intrcpt = 0.41 ± 0.04

Correlation Coefficient = -0.74

leads to

K = 0.27 ± 0.05 μM
n = 1.5

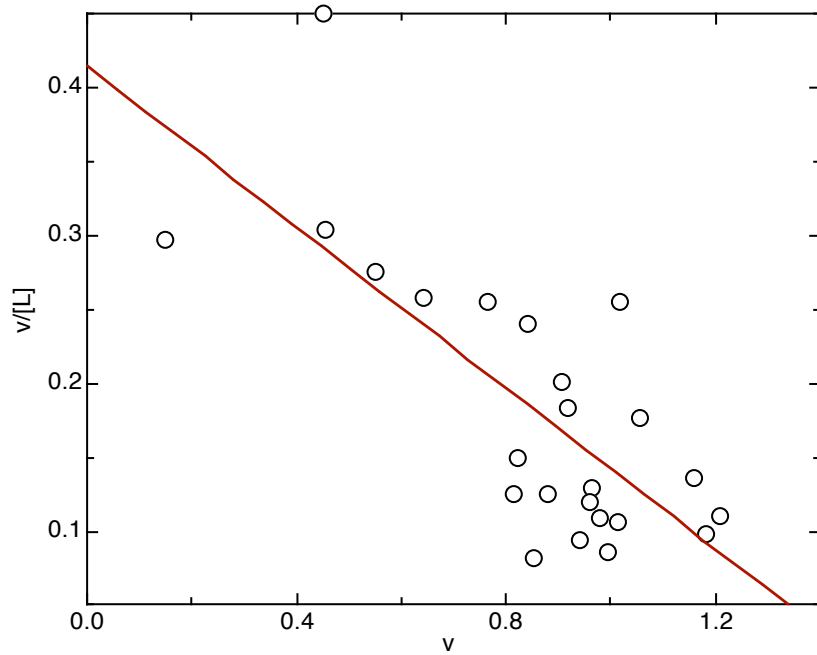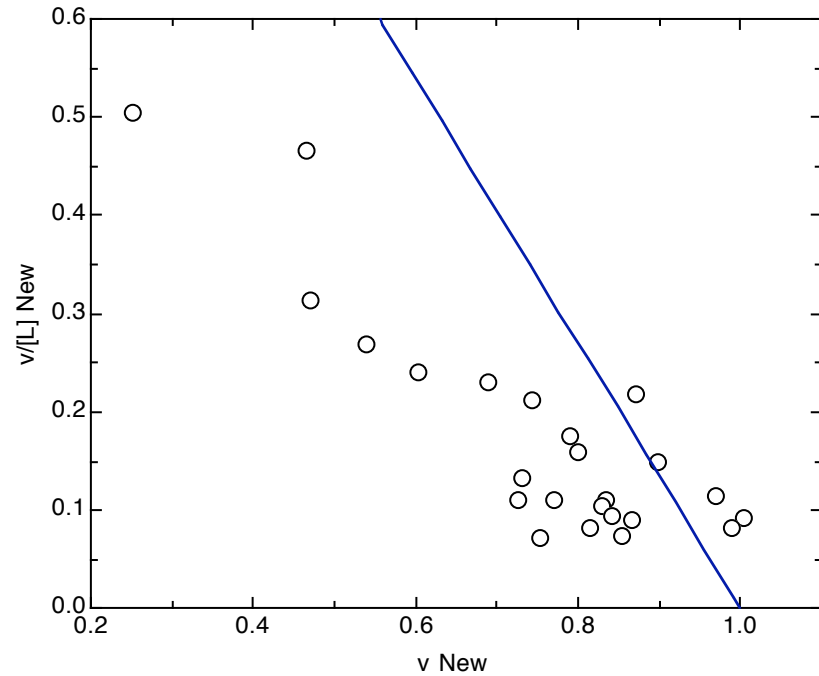How many binding sites?

Direct Fit Gives a Better Result

[P] = 0.1 μM

← 0.31
← 0.28

K = 1.35 ± 0.66 μM

A_{bound} = 0.312 ± 0.012
$A_{unbound}$ = 0.087 ±!0.040

0.12 →
0.09 →

K = 0.27 ± 0.05 μM

A_{bound} = 0.28
$A_{unbound}$ = 0.12
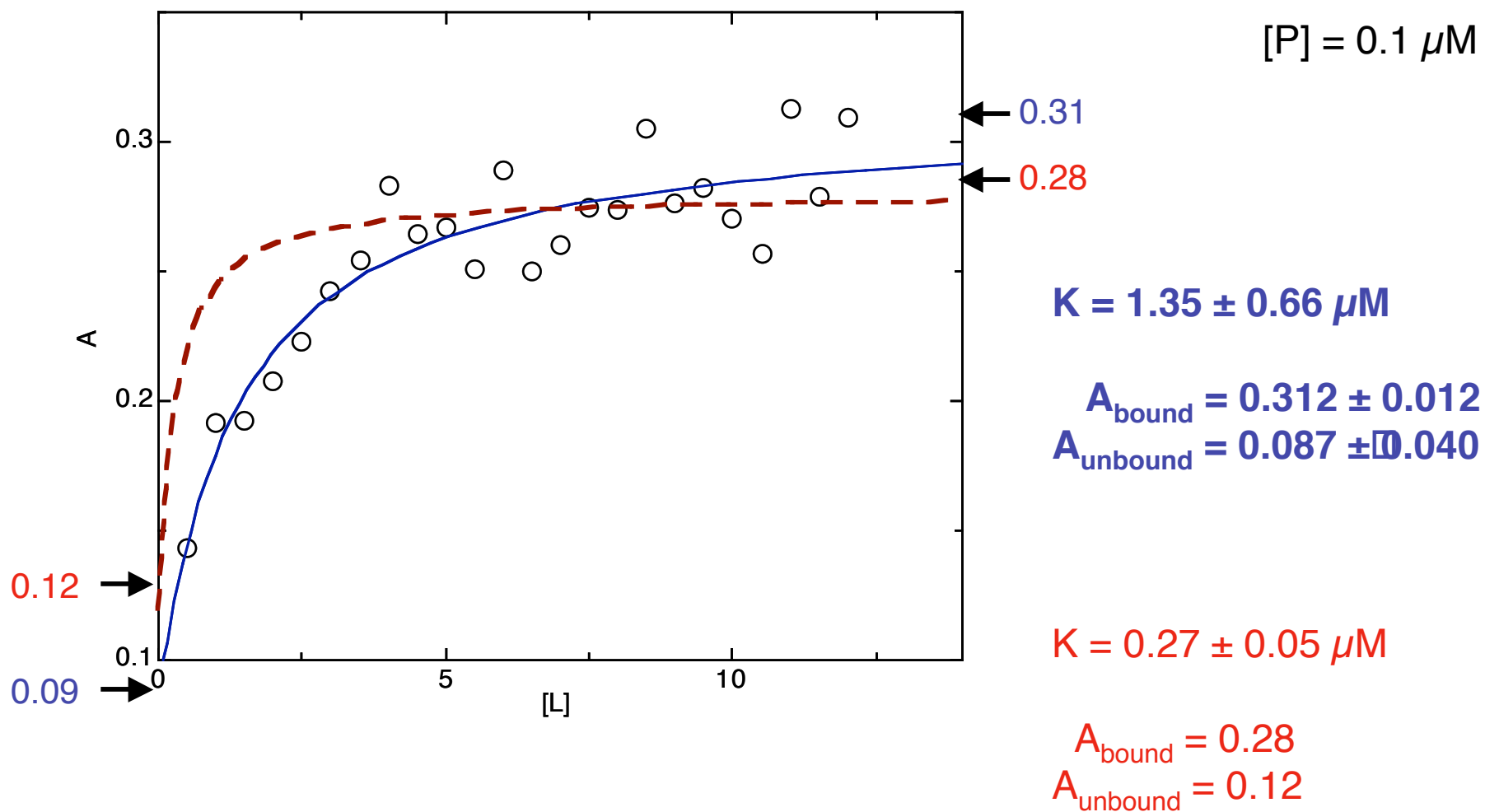
Back to Scatchard
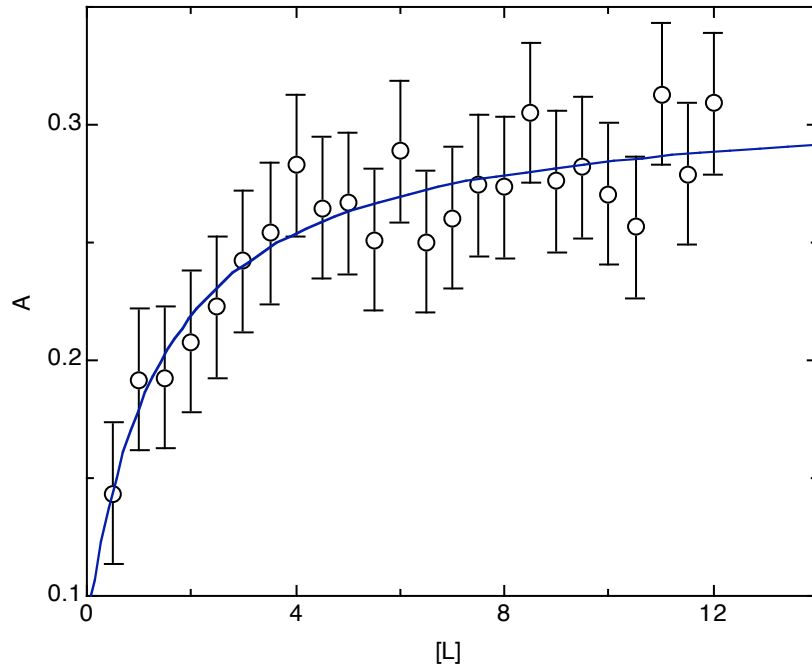

$K = 0.27 \pm 0.05 \ \mu M$

$A_{bound} = 0.28$
$A_{unbound} = 0.12$

$K = 1.35 \pm 0.66 \ \mu M$

$A_{bound} = 0.312 \pm 0.012$
$A_{unbound} = 0.087 \pm !0.040$

- Note that the "data" are different

Direct fit
K = 1.35 ± 0.66 μM

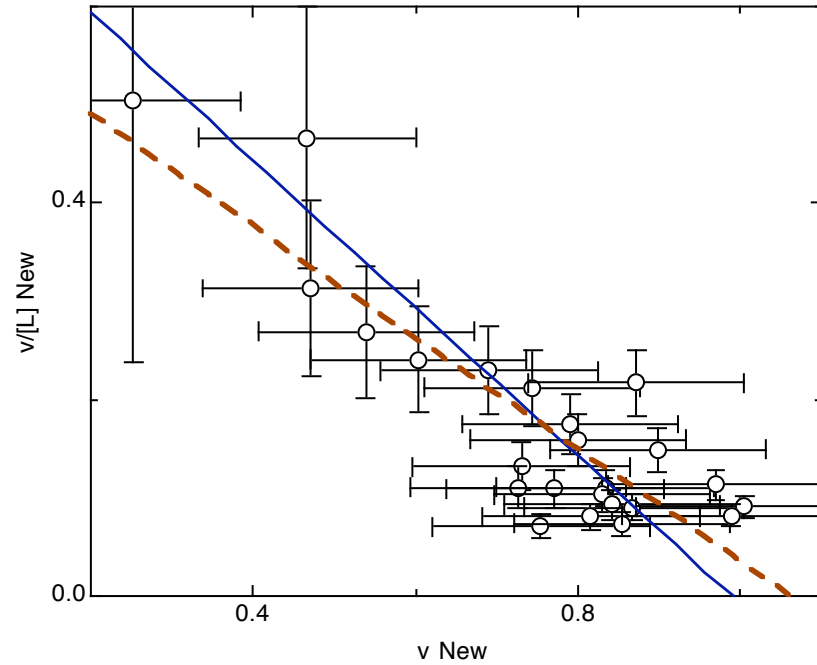A_{bound} = 0.312 ± 0.012
$A_{unbound}$ = 0.087 ±!0.040

Weighted fit
K = 0.75 ± 0.20 μM
n = 0.99

Unweighted fit
K = 0.57 ± 0.07 μM
n = 1.06

Fixed
A_{bound} = 0.312
$A_{unbound}$ = 0.087

K = 1.0

A_{bound} = 0.3
$A_{unbound}$ = 0.1