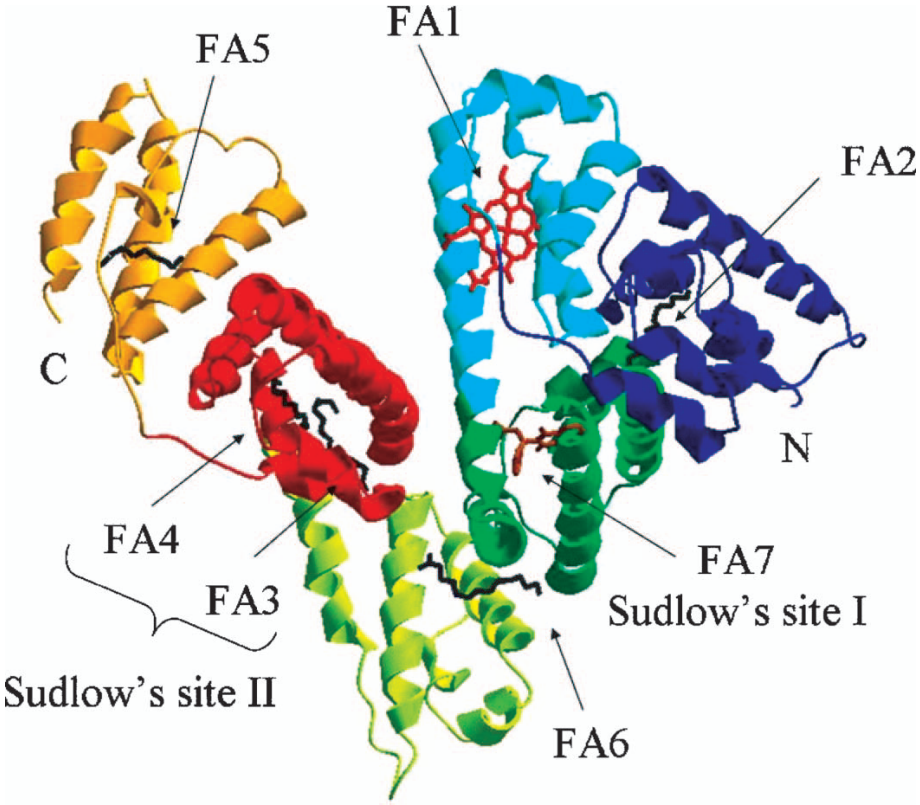
Human Serum Albumin (HSA)



The six subdomains of HSA are colored:
- subdomain IA: blue;
- subdomain IB: cyan;
- subdomain IIA: dark green;
- subdomain IIB: light green;
- subdomain IIIA: red;
- subdomain IIIB: orange.

Heme (red) fits the primary cleft in subdomain IB (FA1).

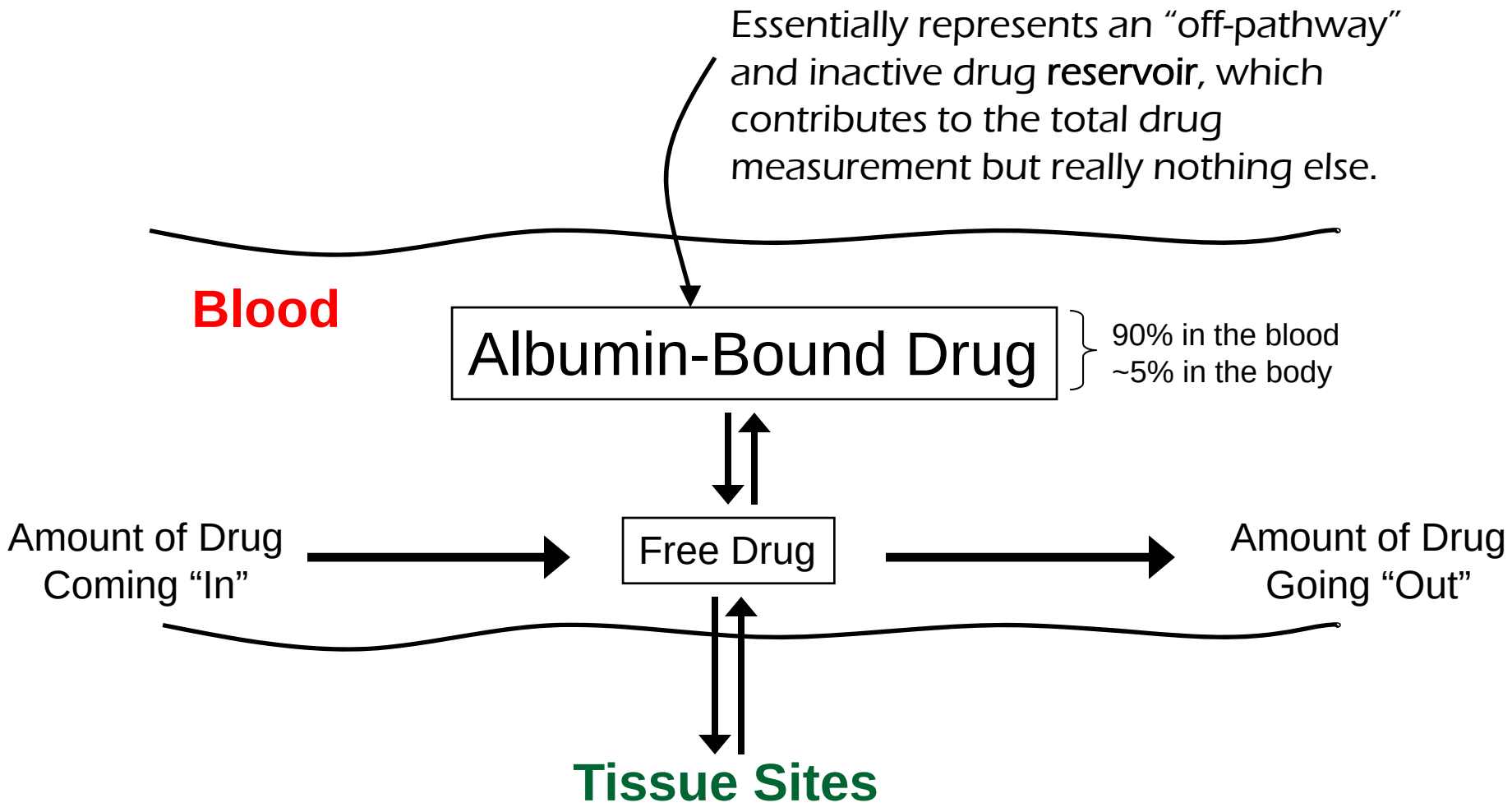
Sudlow's site I (in subdomain IIA) is occupied by warfarin (brown).

Sudlow's site II (in subdomain IIIA) is occupied by two myristate anions.

Except for FA1 and FA7, all other FA sites are occupied by myristate.

Atomic coordinates were taken from PDB entries 1H9Z and 1O9X (26, 29).

Free vs. Total Drug Levels



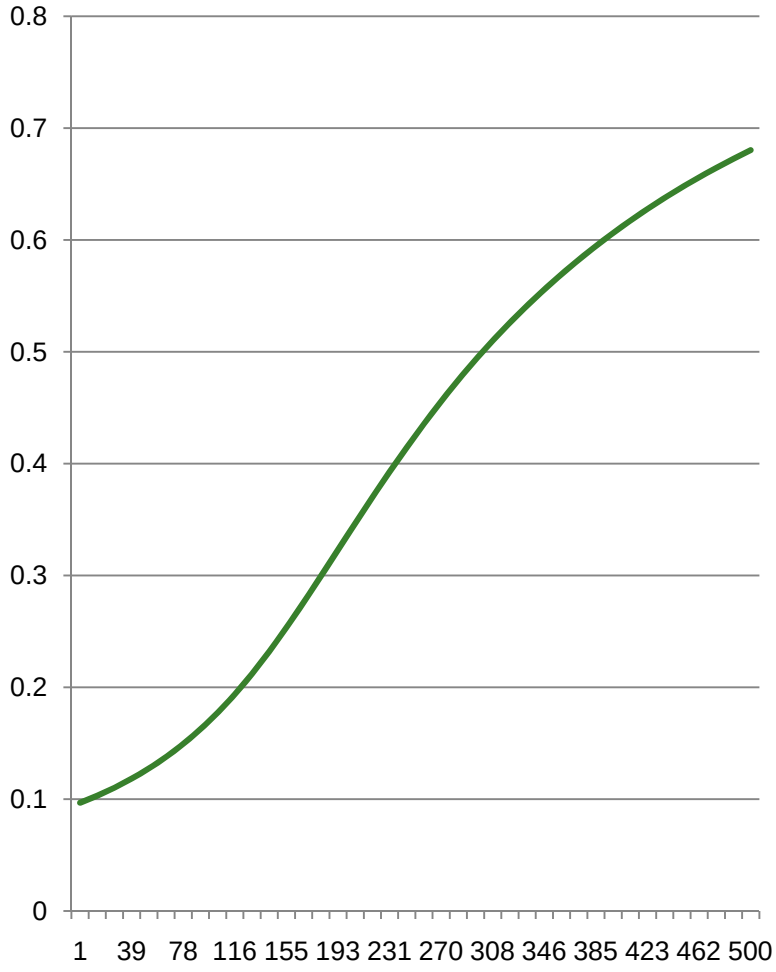
Free vs. Total Drug Levels

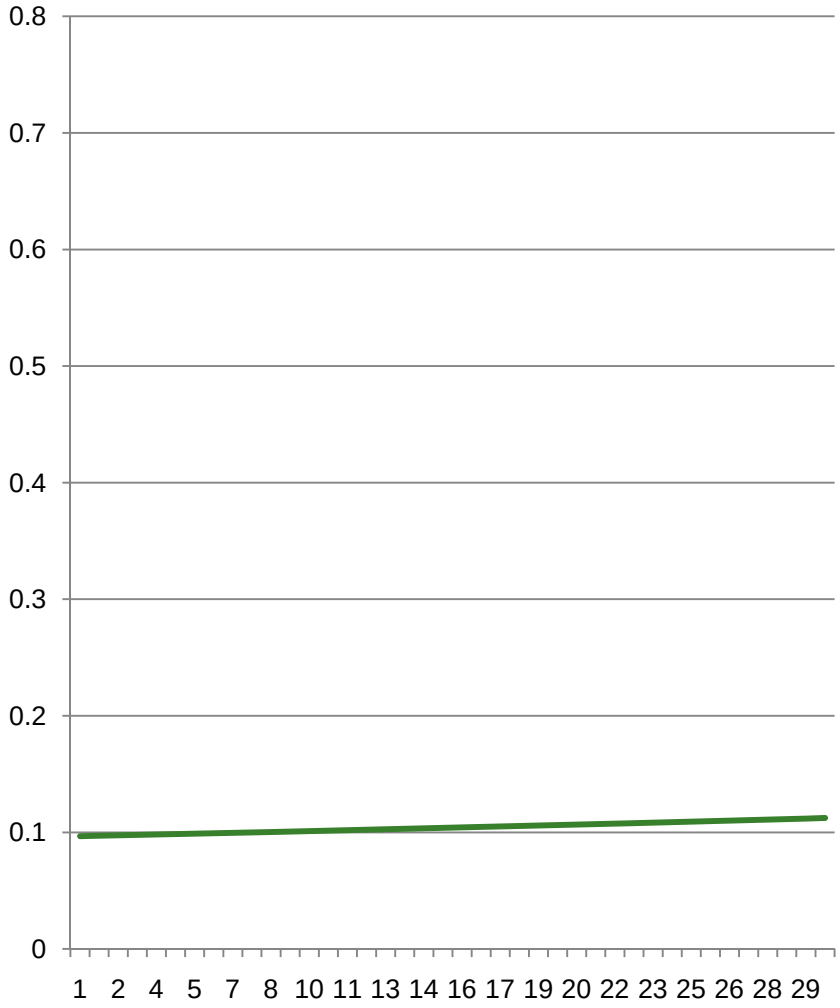
- With fixed albumin, free phenytoin is ~10% of total phenytoin across a relatively wide range of total phenytoin concentrations.

- In contrast, the fraction of total phenytoin that is free varies greatly with albumin concentration.
  - Or, more equivalently, the concentration of available albumin binding sites. Other compounds that compete with phenytoin for albumin binding, lower the effective albumin concentration (or binding affinity).

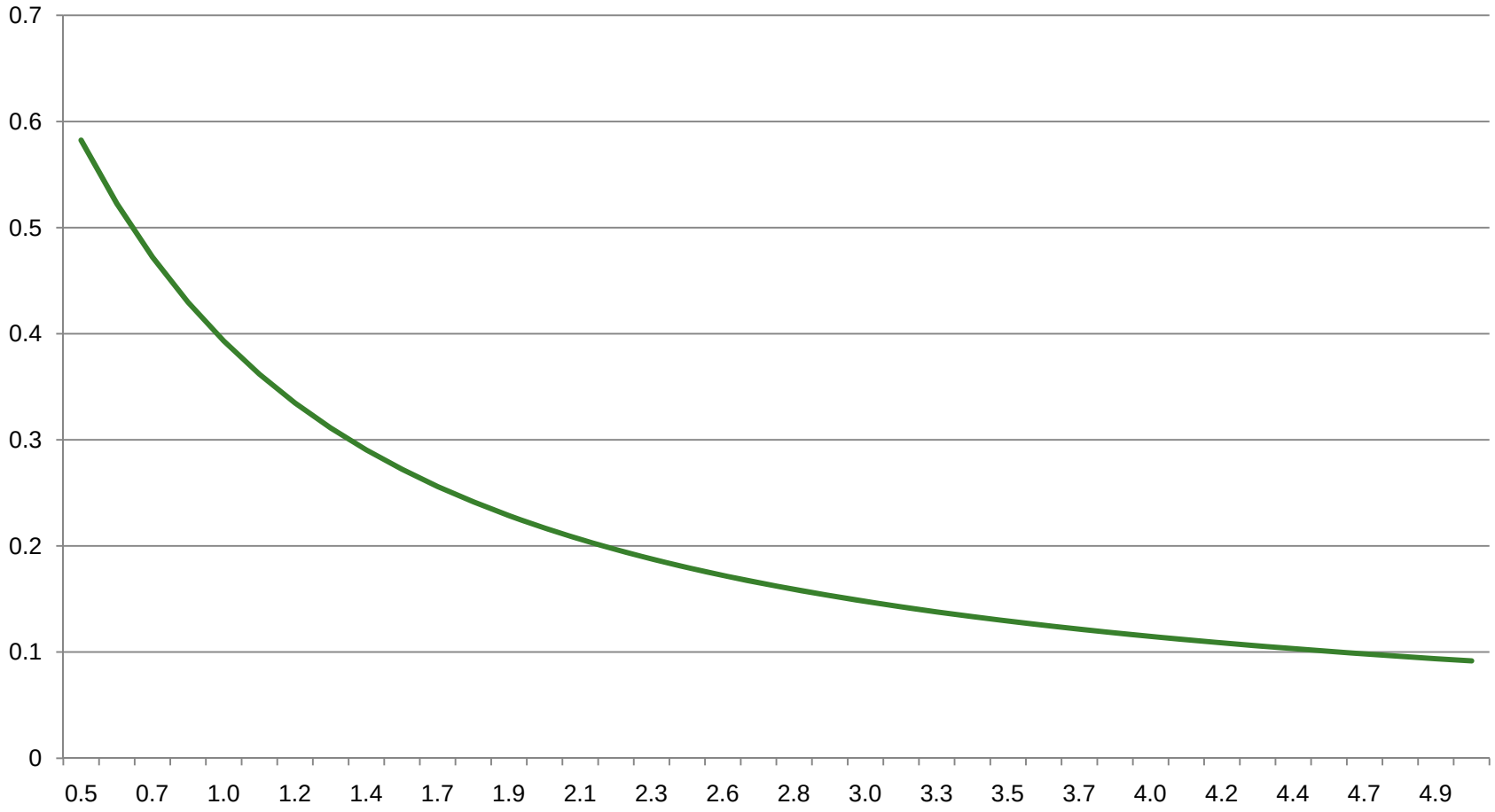
- Always remember that drug efficacy, toxicity, and clearance depend ONLY on the free drug concentration.

Fraction Free vs. [Total Phenytoin]





Free Phenytoin vs. [Albumin]



Free vs. Total Drug Levels: PK Parameters

- Pharmacokinetic parameters such as V_d, Cl, and the reference range are typically defined for the total drug concentration (since that is what we usually measure) based on the assumption that the free is constant fraction of the total.
  - The reference range for total phenytoin is 10 – 20 mg/L, with a corresponding range of 1 – 2 mg/L for free drug.
  - Volume of distribution (V_d) is 0.7 L/kg in adults for total phenytoin. This corresponds to 7.0 L/kg for free drug.
  - A similar relationship exists for clearance (Cl), although phenytoin is eliminated by non-linear pharmacokinetics. The clearance for total phenytoin is always ~10% the clearance for free phenytoin.
  - However, notice that the half-life ($t_{1/2}$) is the same (use equation #3).
  - Note, that some drugs display “non-restrictive clearance” where both bound and free drug is available for metabolism (receptor-mediated), in which case none of this stuff applies (just an FYI).

Free vs. Total Drug Levels: Simple Case

- What happens when albumin is much lower than normal (i.e. 2 g/dL instead of 4 g/dL)?
- In terms of phenytoin dosages, free phenytoin levels and elimination rates, not much actually.
  - Note that the amount of phenytoin bound to albumin in the bloodstream is small relative to the overall amount of phenytoin in the body.
    - At 20 mg/L, there is 20 mg/L*0.65L/kg*70kg = 910 mg of phenytoin in the entire body.
    - And 18 mg/L * ~3 L (plasma volume) = 54 mg bound to albumin in the bloodstream.

Free vs. Total Drug Levels: Simple Case

- Imagine a patient on a stable dose of phenytoin with a total drug level of 15 mg/L and a serum albumin of 4 g/dL.
  - free phenytoin = 1.5 mg/L
  - bound phenytoin = 13.5 mg/L
- The patient develops mild nephrosis and their serum albumin slowly declines from 4 to 2 g/dL with no significant change in liver function.
  - What will happen to their free and total phenytoin concentrations?
  - Does their rate of elimination change?
  - Do they need a change in dose?

Free vs. Total Drug Levels: Simple Case

- The only thing that happens to this patient is that the amount of drug bound to albumin is cut in half (from ~13.5 to ~6.75 mg/L).
- Note that the amount of drug going in (i.e. the dose), the free phenytoin level and the amount of drug being eliminated are NOT changed.
- The measured free phenytoin will remain at 1.5 mg/L but the total will decline to ~8 mg/L.
  - At this point, the patient's ratio of free:total drug will have increased to ~0.2 (from 0.1 originally).
  - Hence, another way of adjusting the expected ***total*** phenytoin is using the factor "[albumin]/4 g/dL".
- There is no need for a change in dosage!

Free Phenytoin Consult

- An ICU patient has an unexpectedly low total [phenytoin] = 6 mg/L, despite an appropriately calculated loading dose.
- The ICU resident calls and asks if there could be a problem with our assay.
- We notice that the patient has a very low albumin of 1.5 g/L.
- How do we advise them on interpreting this low total phenytoin level?

Free Phenytoin Consult

- There are actually multiple approaches, but they all rely on initially calculating an appropriate ***correction factor (f)*** for the low albumin:
  - Simple method: f = 1.5 / 4 = 0.375
  - Better equation: f = (0.2 * 1.5) + 0.1 = 0.4 (*Sheiner-Tozer*)
- Method 1: Calculate a corrected reference range for total phenytoin.
  - 0.4 * (“standard ref range”) = “corrected ref range”
  - 0.4 * (10 – 20 mg/L) = 4 – 8 mg/L
- Method 2: Calculate a corrected total phenytoin.
  - $[\text{phenytoin}]_{\text{total,corr}}$ = (6 mg/L) / 0.4 = 15 mg/L
- Method 3: Calculate a predicted free phenytoin.
  - $[\text{phenytoin}]_{\text{free,predicted}} = 0.1/0.4 * [\text{phenytoin}]_{\text{total,measured}}$
  - $[\text{phenytoin}]_{\text{free,predicted}}$ = 0.1/0.4 * 6 mg/L = 1.5 mg/L
- Note that all three of the above methods demonstrate that the patient is precisely in the middle of the standard reference range.