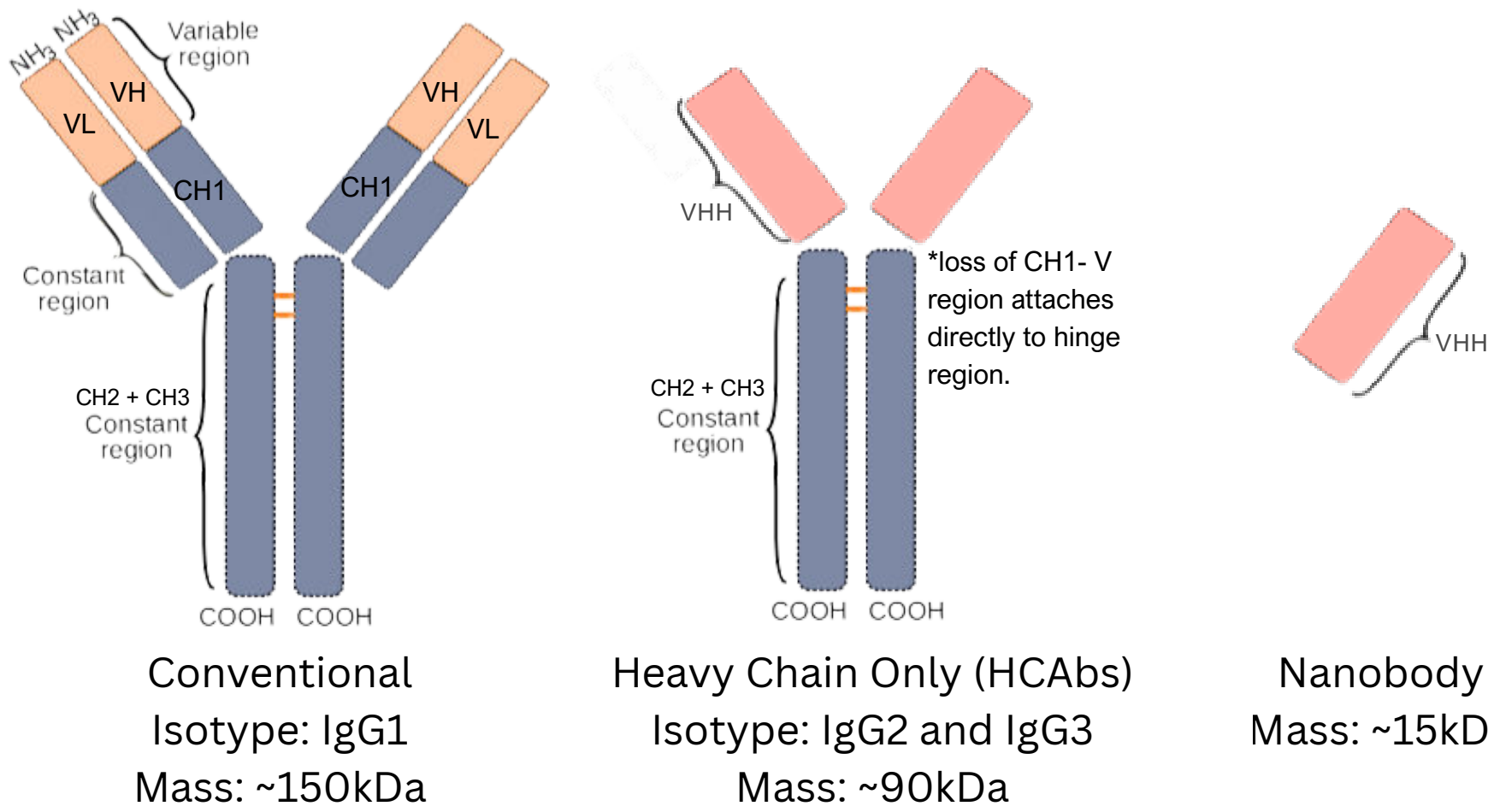


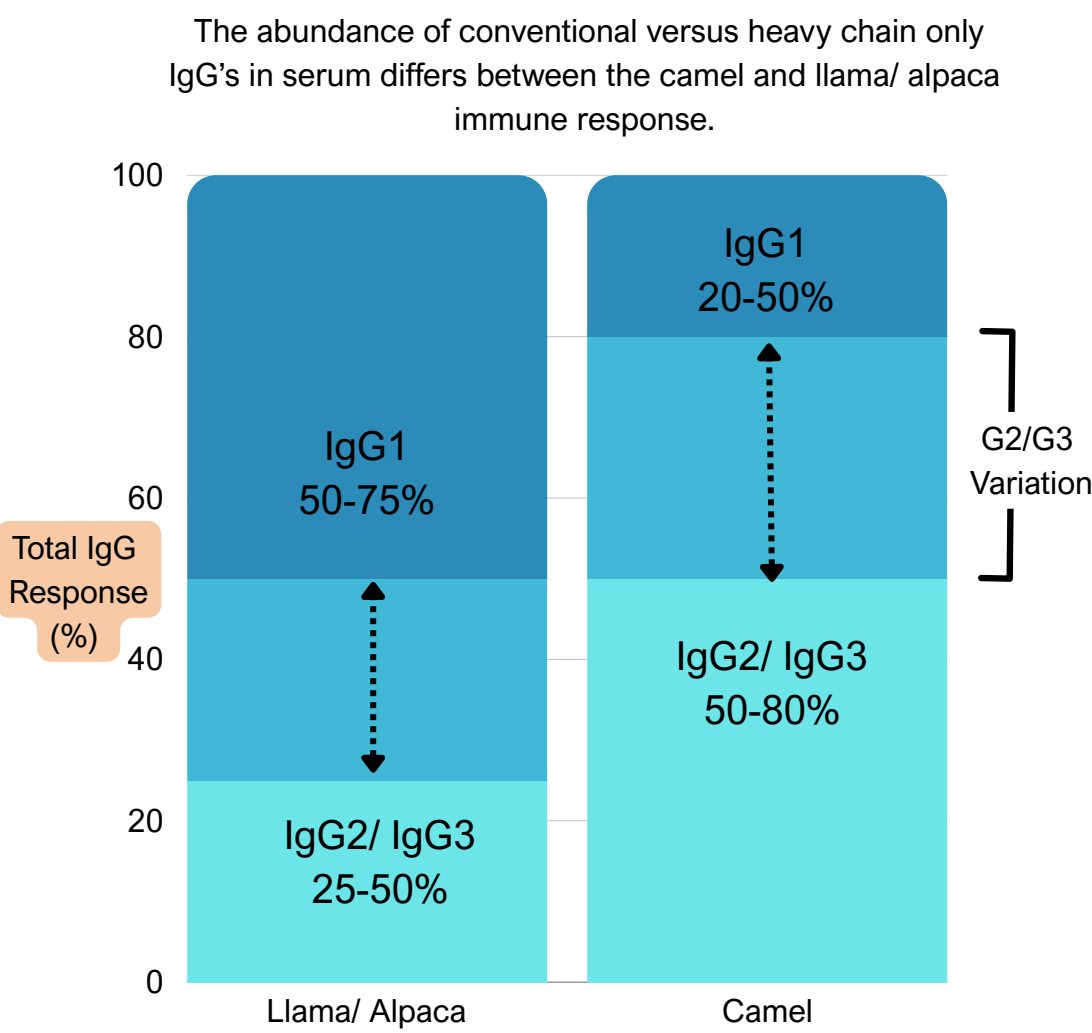
NANOBODY CHEAT SHEET



Antibody Structures



IgG1 vs. IgG2/IgG3

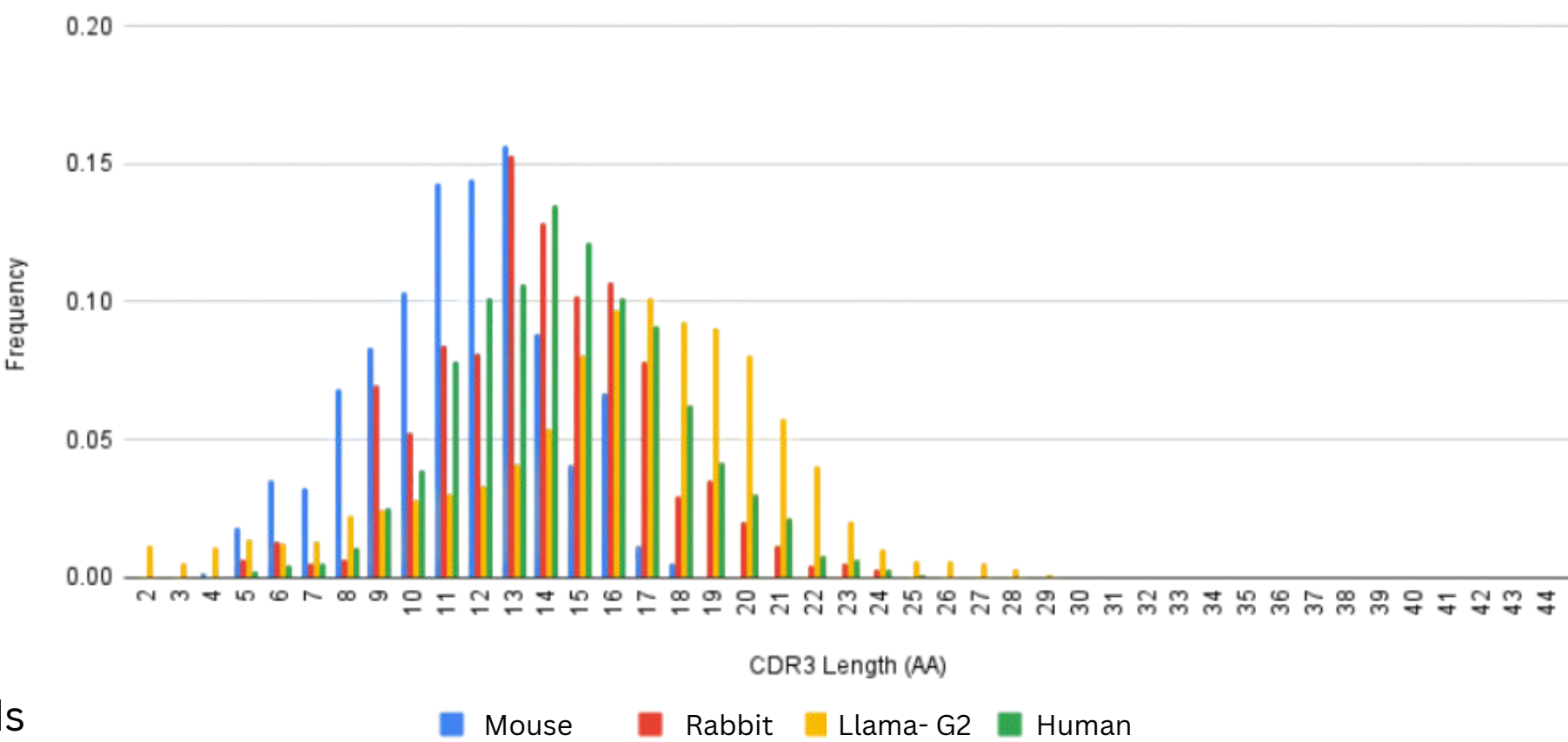


Trait	Conventional	VHH
Solubility	moderate	high
Tissue Access	surface access	higher penetration
Stability	moderate	high
Half Life	days	minutes to hours

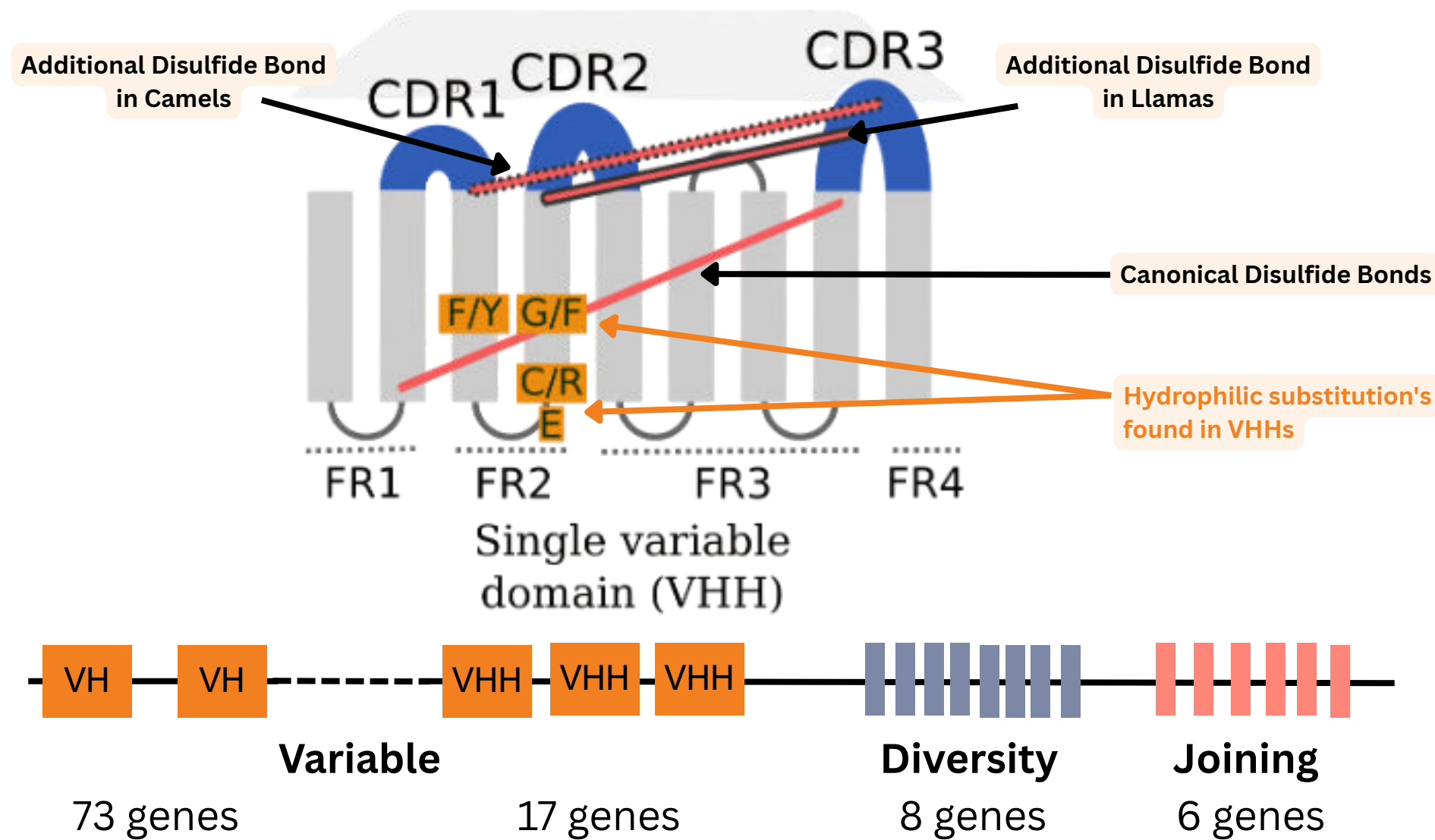
CDR3 Length

Median CDR3 length:
Mouse: 11 amino acids
Rabbit: 13 amino acids
Human: 14 amino acids
Llama: 16-17 amino acids

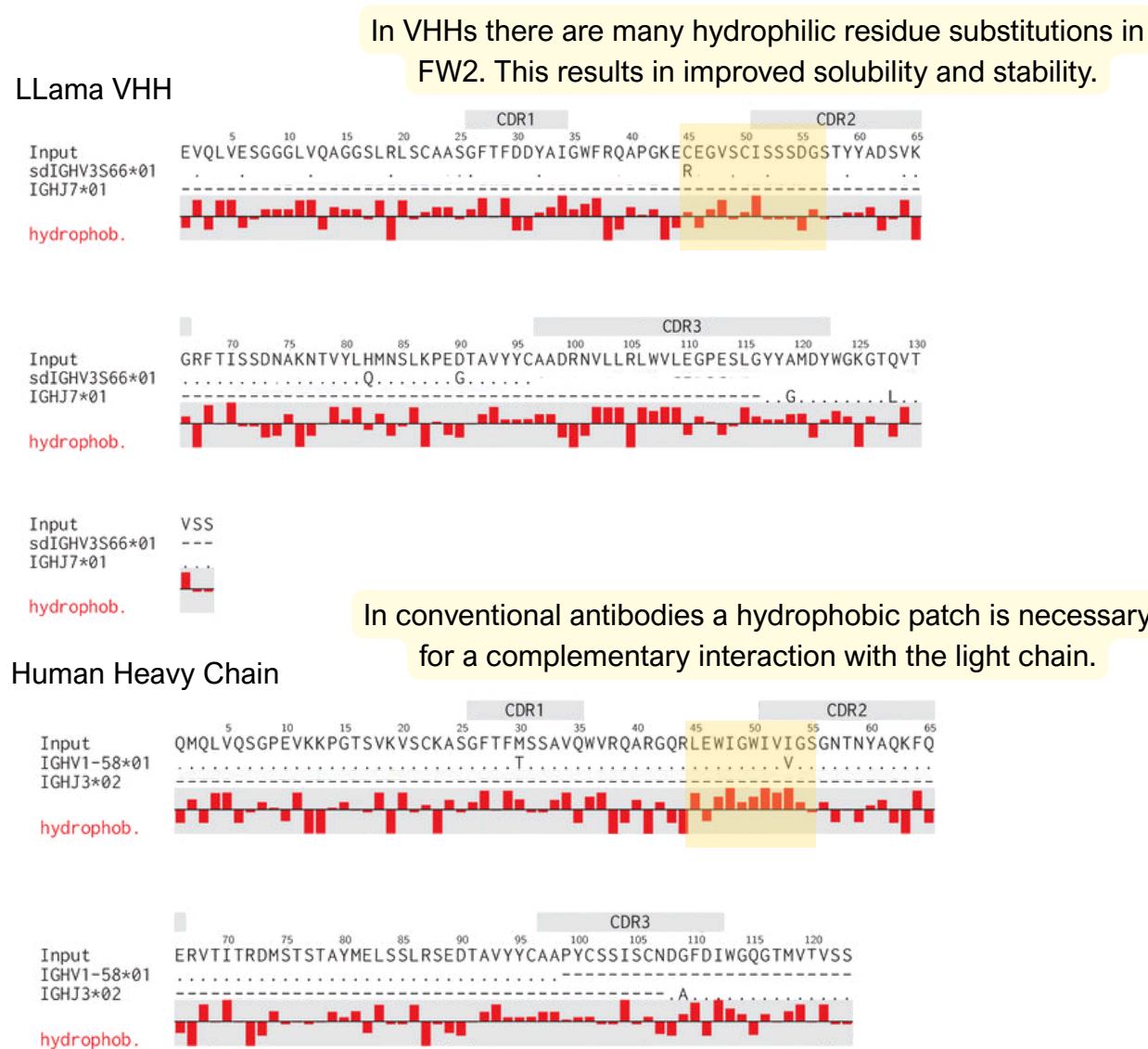
The VHH repertoire has **longer CDR3s** than other species



VHH Structure



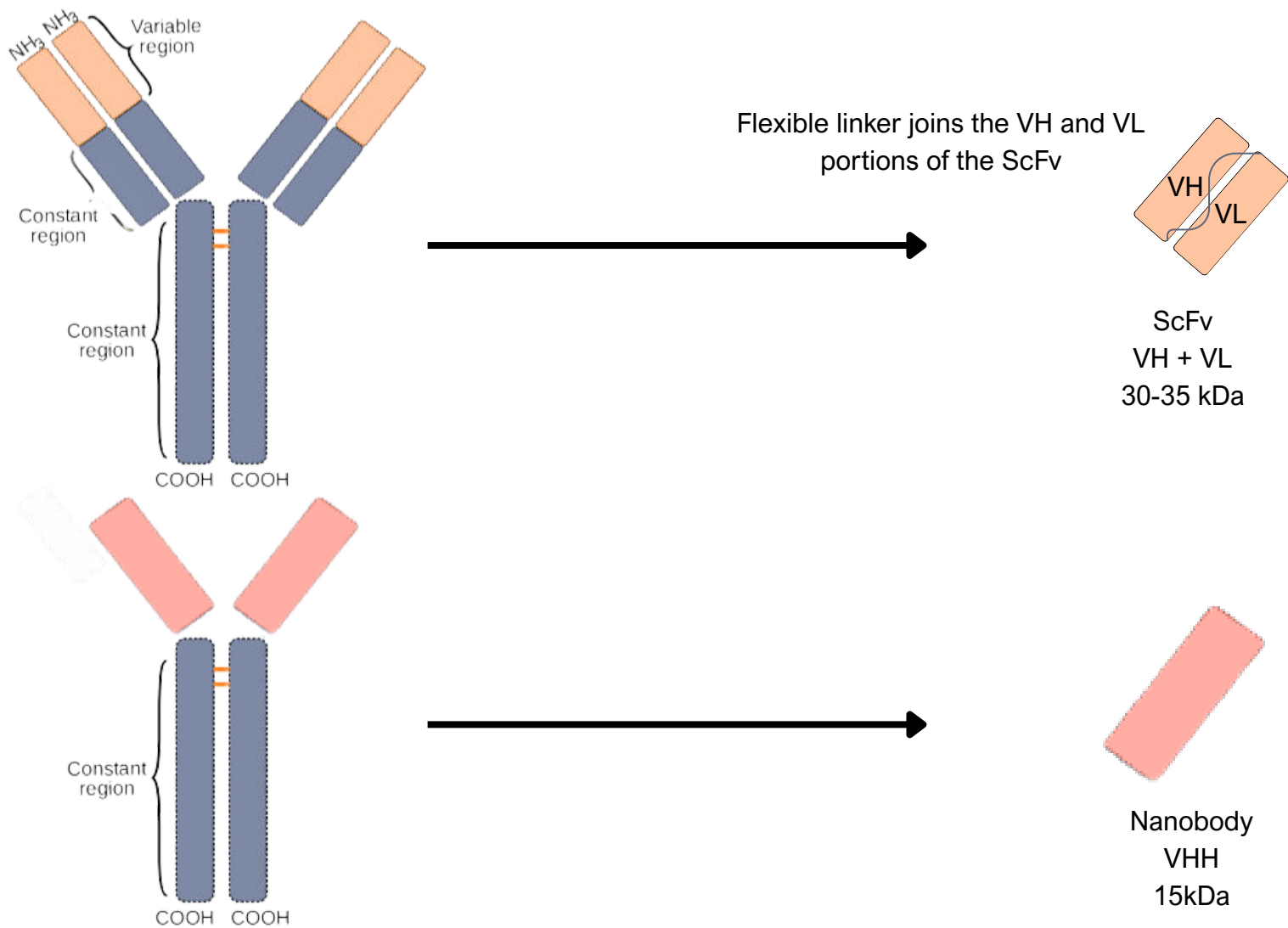
Hydrophobicity



NANOBODY CHEAT SHEET



Antibody Fragments



Protein A/G Affinity

Species	Immunoglobulin	Protein A	Protein G
Alpaca/ Llama	IgG1	+	+
	IgG2	+	-
	IgG3	+	+

Protein G lacks affinity to the G2 subclass of HCAbs due to differences in the Fc region of the antibody. The Hinge region of the IgG3 subclass is 11 amino acids while the hinge region of the IgG2 subclass is 35 amino acids.

Hinge (35 AA)

Hinge (11 AA)

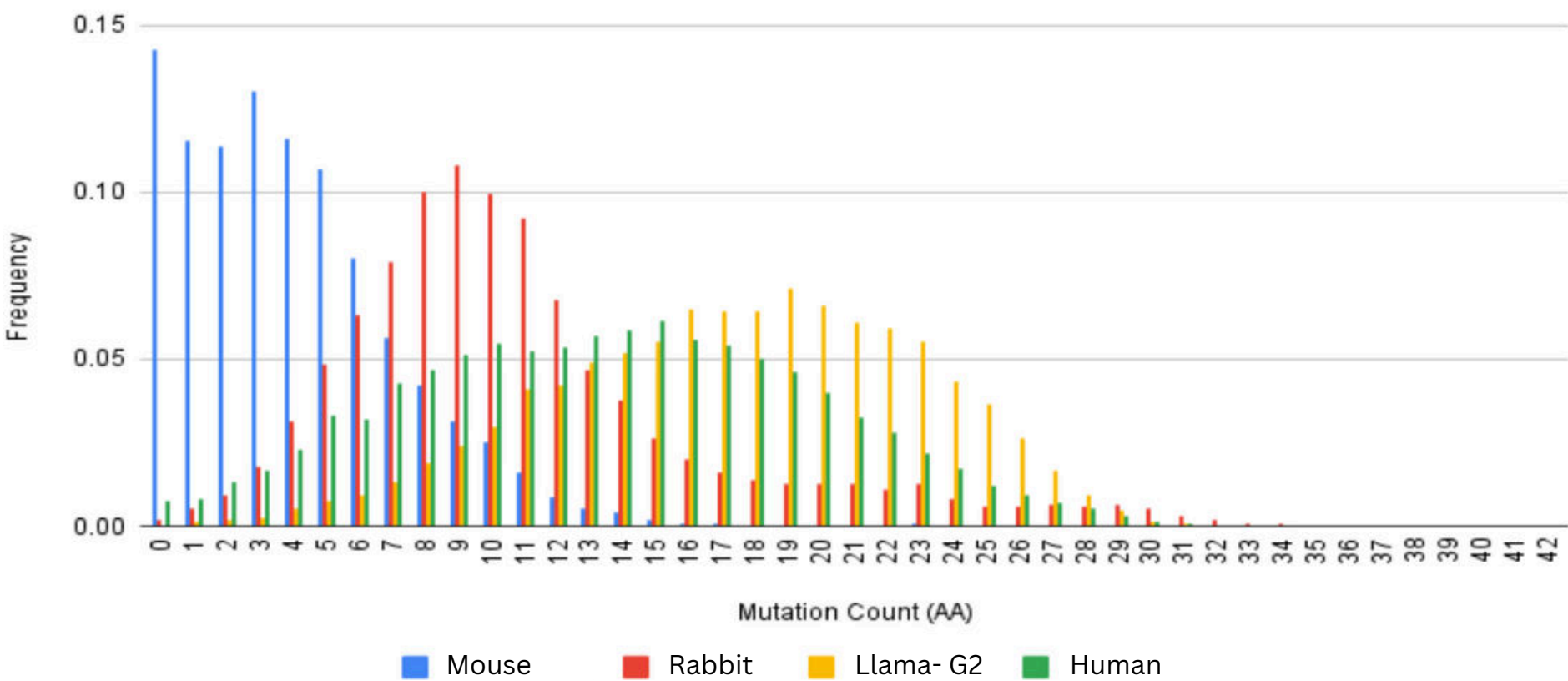
IgG2

IgG3

Trait	ScFV	VHH
Flexibility	High Due to flexible linker	Low due to canonical S-S bond linking FW2 and FW4.
Solubility	Low. Maintains the Hydrophobic patch which encourages protein coagulation.	High. Mutated FW2 region eliminates hydrophobic patch.
Stability	Low to moderate. Highly affected by temp, pH and solvent contents.	High
Half Life	Engineered from the conventional Abs. Requires precise accurately paired heavy and light chains.	Naturally produced as a part of the camelids immune response.

Mutation Rate

VHH's tend to be more mutated than other species



Common Mutations in VHHs

