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Title	Enhanced selectivity of hydrogel-based molecularly imprinted polymers (HydroMIPs) following buffer conditioning.
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Creators	El-Sharif, HF, Phan, QT and Reddy, Subrayal M

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Hydrogels	polyAA	polyNHMA	polyNiPAm
Bovine Haemoglobin (BHb)	8:1	8:1	6:1
Equine Myoglobin (EMb)	4:1	3:1	4:1
Bovine Catalase (BCat)	3:1	4.5:1	5:1

Table 1 – Calculated MIP:NIP selectivity ratios (α) for a variety of protein imprinted hydrogels under MilliQ water conditioning. All values are means of triplicate experiments.

No.	MIP Prepared in	Conditioning series	BHb Loaded in	Selectivity ratio (α)
1	Water	PBS pH 7.4	PBS pH 7.4	27:1
2	Water	Water	PBS pH 7.4	25:1
3	PBS pH 7.4	PBS pH 7.4	PBS pH 7.4	25:1
4	PBS pH 7.4	PBS pH 4.7	PBS pH 4.7	8:1
5	Water	Water	Water	8:1
6	Water	PBS pH 7.4	Water	7:1
7	Water	PBS* pH 7.4	PBS* pH 7.4	7:1
8	PBS pH 7.4	PBS* pH 7.4	PBS*pH 7.4	6:1
9	PBS pH 7.4	PBS pH 7.4	Water	4:1
10	PBS pH 7.4	Water	Water	1:1
11	PBS pH 7.4	Tris pH 7.4	Tris pH 7.4	30:1
12	Water	Tris pH 7.4	Tris pH 7.4	128:1
13	Water	Tris pH 5.4	Tris pH 5.4	60:1
14	Water	Tris pH 9.4	Tris pH 9.4	60:1
15	PBS pH 7.4	Succinate pH 7.4	Succinate pH 7.4	10:1
16	Water	Succinate pH 7.4	Succinate pH 7.4	14:1
17	PBS pH 7.4	Succinate pH 2.9	Succinate pH 2.9	9:1
18	Water	Succinate pH 2.9	Succinate pH 2.9	1:1

Table 2 – Calculated selectivity ratios (α) for BHb-MIP_{polyAA} hydrogels in different preparing, conditioning and protein loading media, * denotes a ½ dilution of PBS. All values are means of triplicate experiments.

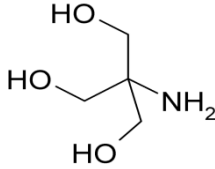
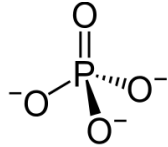
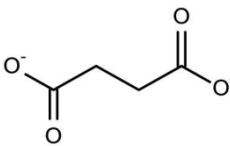
Buffer	pKa	Structure
Tris buffer	8.1	
PBS buffer	7.2	
Succinate buffer	4.2	

Table 3 – Buffer structures and pKa values for Tris, PBS and succinate [15].

Protein	pI	net charge in		Selectivity ratios (α)		
		Water (pH5.4)	Tris (pH7.4)	polyAA	polyNHMA	polyNiPAm
Bovine Haemoglobin (BHb)	6.8	+ve	-ve	128:1	44:1	33:1
Equine Myoglobin (EMb)	7.2	+ve	-ve	35:1	43:1	34:1
Bovine Catalase (BCat)	5.5	-ve/+ve	-ve	26:1	24:1	29:1

Table 4 - Protein isoelectric points (pI) and net charge variations in Tris buffer (pH 7.4) and MilliQ water [15, 17- 22]; calculated MIP:NIP selectivity ratios (α) for hydrogels under Tris buffer (pH 7.4) conditions. All values are means of triplicate experiments.

Conditioning series	Phase	Selectivity ratio (α)
Tris Series	Tris Load	128:1
	Water Load	0.8:1
Water Series	Water Load	8:1
	Tris Load	46:1

Table 5 - Calculated MIP:NIP selectivity ratios (α) for BHb-MIP_{polyAA} hydrogels under two different water and Tris buffer (pH 7.4) conditioning series with interchanging protein Load phase conditions.