Guide to Antigen Preparation

If you are providing us with your own protein antigen for immunisation then you may find the following guide useful.

In general, most buffers and additives are suitable for antibody production apart from detergents, protein solubilising and refolding agents (such as Urea and Guanidinium Hydrochloride) and organic solvents such as Dimethylsulphoxide (DMSO). There are certain limitations in buffers and additives suitable for antibody production and the table below shows the buffer component limitations for antigens:



CHAPS 0.1-0.2M DMSO Not Acceptable DTT 1M EDTA 10mM Other chelates 10mM Ethanol <5% Glycerol 20% Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCI, KCI, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable Methanol Not Acceptable	Chemical	Suitable for Immunisations
DTT 1M EDTA 10mM Other chelates 10mM Ethanol <5%	CHAPS	0.1-0.2M
EDTA 10mM Other chelates 10mM Ethanol <5% Glycerol 20% Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	DMSO	Not Acceptable
Other chelates 10mM Ethanol <5% Glycerol 20% Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCI, KCI, MgCI2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	DTT	1M
Ethanol <5% Glycerol 20% Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	EDTA	10mM
Glycerol 20% Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	Other chelates	10mM
Glutathione Must dialyze against PBS before it can be used Imidazole 1M Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	Ethanol	<5%
Imidazole 1M Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	Glycerol	20%
Mercapto-ethanol 1mM NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8)	Glutathione	Must dialyze against PBS before it can be used
NaCl, KCl, MgCl2 1M Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8)	Imidazole	1M
Octylgluside 1% PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	Mercapto-ethanol	1mM
PMSF Not Acceptable Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2% Tris 0.1M Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	NaCl, KCl, MgCl2	1M
Primary Amines <1M (pH 6 to 8) Salts 1M SDS <0.2%	Octylgluside	1%
Salts 1M SDS <0.2%	PMSF	Not Acceptable
SDS <0.2%	Primary Amines	<1M (pH 6 to 8)
Tris 0.1M Triton X-100 <0.5%	Salts	1M
Triton X-100 <0.5% Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	SDS	<0.2%
Tween 20 <0.1% Urea 6M Guanidine Not Acceptable	Tris	0.1M
Urea 6M Guanidine Not Acceptable	Triton X-100	<0.5%
Guanidine Not Acceptable	Tween 20	<0.1%
	Urea	6M
Methanol Not Acceptable	Guanidine	Not Acceptable
•	Methanol	Not Acceptable



