

## TALON® Metal Affinity Resins Protocol-at-a-Glance

(PT1320-2)

The following summary table for purifying his-tagged proteins with TALON Spin Columns and TALON®, TALON Superflow, and TALON CellThru Resins is provided for your convenience and is not intended for first-time users. For detailed protocols, see the TALON Spin Columns Protocol-at-a-Glance (PT5171-2) and the indicated sections of the TALON Metal Affinity Resins User Manual (PT1320-1), which are available at [www.clontech.com/manuals](http://www.clontech.com/manuals).

Method	Spin Column (PT5171-2)	Batch (PT1320-1, Sec. V.B)	Batch/Gravity-Flow (PT1320-1, Sec. V.A)
<b>Binding Conditions</b>			
Resin bed-volume <sup>a</sup>	0.5 ml	1.0 ml	1.0 ml
Polyhistidine-protein binding capacity	2–3 mg	~5 mg	~5 mg
Sample volume	1 ml (each loading)	~13 ml	~13 ml
Binding time	5 min	20 min	20 min
<b>Washing Conditions<sup>b</sup></b>			
pH gradient	2 x 1 ml (pH 7)	2 x 10 ml (pH 7)	2 x 10 ml (pH 7)
	1 x 1 ml (pH 7) (optional)	1 x 10 ml (pH 7)	2 x 3 ml (pH 7)
Imidazole	2 x 1 ml (pH 7) + 5–10 mM imid.	2 x 5 ml (pH 7) + 5–10 mM imid.	2 x 3 ml (pH 7) + 5–10 mM imid. (in column)
Washing time	5 min/wash	10 min/wash	10 min/wash
<b>Elution Conditions</b>			
pH gradient	2 x 0.5 ml (pH 5) (1 ml fractions)	4 x 1 ml pH (5) (0.5 ml fractions)	4 x 1 ml (pH 5)
Imidazole	2 x 0.5 ml (pH 7) + 100 mM imid. (1 ml fractions)	4 x 1 ml (pH 7) + 100 mM imid. (0.5 ml fractions)	4 x 1 ml (pH 7) + 100 mM imid.
Elution time	5 min/elution	10 min/elution	10 min/elution

<sup>a</sup> Volume of TALON Resin that was prepared as described in the TALON Metal Affinity Resins User Manual (PT1320-1, Section V.A.1).

<sup>b</sup> "2 x 1 ml (pH 7.0)" means wash two times with 1 ml of buffer at pH 7.0 each time.

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(030812)



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