

Biotin free media

Medium	Manufacturer	Biotin content [µg/L]	Required amount of Biotin blocking solution per Liter medium [ml]
Mammalia			
DMEM ⁹	Multiple Suppliers	-	-
Leibovitz's L-15 ¹⁰	Multiple Suppliers	-	-
ProCHO™ 5**	Lonza (Cat. No.12-766Q)	-	-
ExCell 293 HEK	Sigma (Cat. No.14571C)	-	-
insect cells			
Graces Insect Medium**	Gibco (Cat. No.11605-045)	-	-
Schneider's Medium ¹¹	Multiple Suppliers	-	-

⁹Schneider, I. (1964), Exp. Zool. 156, 1, 91; ¹⁰Leibovitz, A. (1963) Am. J. Hyg. 78, 173; ¹¹Dulbecco, R. Freeman, G. (1959) Virology 8, 396. Smith, J.D., Vogt, M. and Dulbecco, R. (1960) Virology 12, 185

* Manufacturer data

** IBA internal measurement

Further methods to remove biotin:

- to precipitate the recombinant protein in a first step by ammonium sulfate precipitation, then to remove the biotin containing supernatant and finally to dissolve the precipitated protein prior to Strep-tag® chromatography with buffer W (100 mM Tris-Cl pH 8.0; 150 mM NaCl; 1 mM EDTA (EDTA can be omitted in case of metalloproteins)).

- to make a crude ion exchange step with elution at slightly alkaline pH (>7.5) for direct application on a Strep-Tactin column.

- to concentrate the protein by cross flow ultrafiltration. Please use buffer W for exchange so that the protein concentrate can be applied directly to a Strep-Tactin column.

Although more labor-intensive than adding avidin, these procedures have the advantage that the recombinant protein will be concentrated which contributes to stability of the recombinant protein and which enables higher efficiency of Strep-tag® affinity chromatography.

pH > 7.5 is necessary for efficient Strep-tag chromatography and has in every case to be respected. Precipitates may form during masking with avidin or during concentration steps and have to be removed prior to Strep-tag affinity chromatography.

Note: In case Biotin has already bound, BioLock enables the affordable removal of biotin from surfaces/matrices with reversible biotin binding affinity (regeneration) through an appropriate incubation and subsequent washing step. Equilibrium will be shifted to the avidin-biotin complex driven by its extraordinary affinity in the femtomolar range. A sufficient off-rate of the biotin binding interaction on the surface/matrix in correlation with the incubation time of BioLock is nevertheless a prerequisite for efficient regeneration.