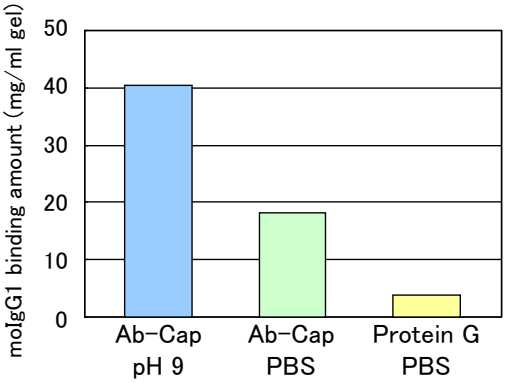
Ab-Capcher™
Purifying mouse monoclonal IgG1 (1)

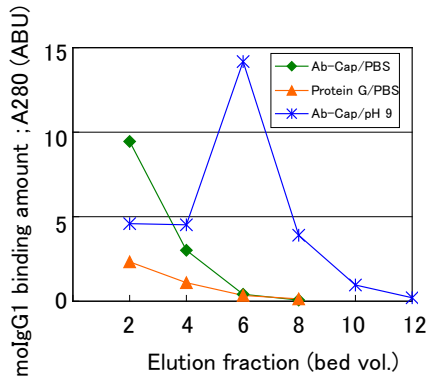
Purification flow chart

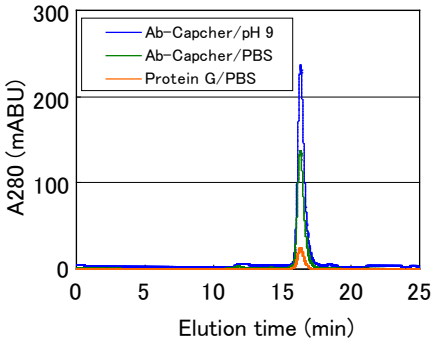
Mouse ascites
↓
50% ammonium sulfate precipitation fraction
↓
Dialysis (PBS)
↓
3-fold dilution with 1.5 M Glycine / 3 M NaCl (pH 9.0) or PBS
↓
Addition to affinity gel (spin column)
↓
Shake (2 hours)
↓
Wash (binding buffer)
↓
Elute with 0.1 M Glycine-HCl (pH 2.8)
(neutralize by adding 1 M Tris)

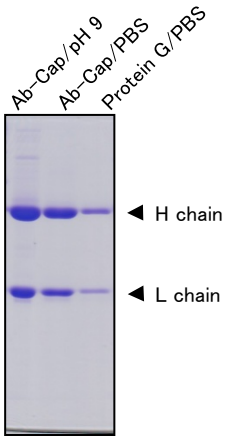
Mouse IgG1 binding amount under various conditions



Analysis of IgG1 fraction purified under various conditions







Elution profile
Elution fraction every 2 bed vols
IgG1 elution amount measurement (A280)

Gel filtration HPLC
Sample: IgG1 elution fraction 10 µl
Analytical column: G3000SWXL
buffer: 35 mM Na-Pi, 0.5 M NaCl, pH 7.0
Flow rate: 0.5 ml/min

SDS-PAGE
Sample: IgG1 elution fraction 1µl
Gel: 12.5 % (Tris-Glycine)
Stain: CBB

A 50% ammonium sulfate precipitate of mouse ascites was diluted with PBS or pH 9 buffer and added to Ab-Capcher™ or a commercial protein G gel. Ab-Capcher™ showed about 5 times the binding amount of mouse IgG1 than Protein G gel in PBS. When a pH 9 binding buffer was used, the Ab-Capcher™ yield increased twofold further compared to PBS. Thus, use of pH 9 buffer with Ab-Capcher™ can greatly increase the yield of mouse monoclonal antibody, but caution is required because the elution of mouse IgG1 is delayed.

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