

Cat. No. MF-PRA-3000

Qbeads-Protein A

Product description

Qbeads-Protein A is designed as a rapid and simple tool or immunoprecipitation, purification/ depletion assays, and other magnetic separation applications. Antibody can easily bind to the Qbeads due to its' high affinity with protein A. Via the antibody specific binding ability, the target protein along with Qbeads-Protein A could be temporarily immobilized at tube wall, so the other parts in the supernatant can be removed easily and efficiently under magnetic attraction.

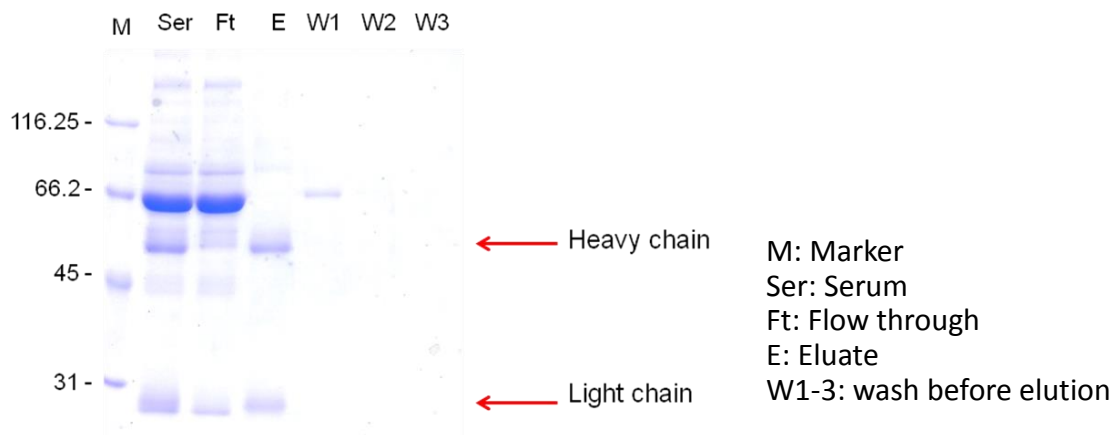
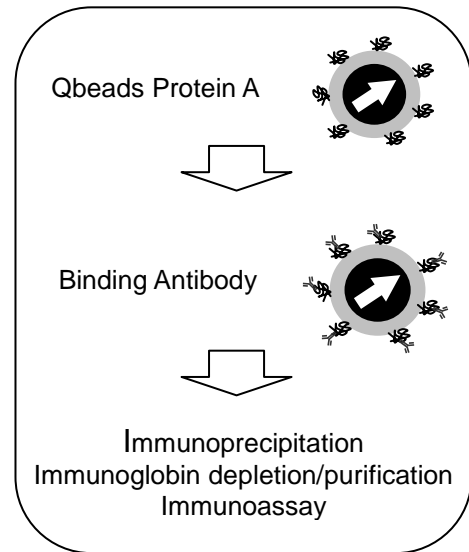


Figure 1. Purification of human IgG (indicated by arrow) from serum sample. (SDS-PAGE stained with Coomassie blue)

Binding characteristics

The binding capacity of Qbeads-Protein A is more than 26 μg human IgG per 100 μL . The binding strength of Qbeads-Protein A to different immunoglobulins is listed as below:

Table 1. Binding strength of Qbeads-Protein A

Species	Binding strength
Human IgG (normal)	++++
Mouse IgG1	+
Rat IgG1	-
Goat IgG	+/-
Rabbit IgG	++++
Chicken IgG	-

Material supplied

Qbeads-Protein A provides Fe₃O₄ beads coated with dextran of an average ~1 µm in diameter. Protein A, about 42 kDa, is coupled covalently to dextran. Qbeads are supplied in phosphate buffered saline, pH 7.4, containing 0.02% Tween 20 and 0.09% sodium azide.

Additional material required

- Washing buffer: PBS buffer (pH 7.4) with 0.02% Tween 20
- Elution buffer if necessary
- Neutralization buffer if necessary
- Desired antibody
- Magnetic stand: **Magdorf** (MDF-08) for the best performance
- Tilt rotation device or vortexer
- Eppendorf tubes & pipett

Protocol

Preparation of Qbeads-Protein A for use

1. Resuspend the Qbeads-Protein A thoroughly by pipetting or vortexing the vial for 15 secs.
2. Transfer adequate amount of Qbeads-Protein A into a clean tube.

NOTE: Take appropriate amount of beads according to the binding capacity mentioned in “Binding Characteristics” above. For example, if 2~3 µg of antibody is used in immunoprecipitation test, 5~10 µl of Qbeads-Protein G is enough for one test. Exceed amount of beads may cause high background in some cases.

3. Place the tube on the magnetic stand for 30-60 seconds to immobilize the beads at tube wall.
4. Discard the supernatant by aspiration with a pipette.
5. Remove the tube from the magnetic stand.
6. Add 200 uL Washing buffer and resuspend the beads by pipetting.
7. Place the tube on the magnetic stand for 30-60 seconds to immobilize the beads at tube wall.
8. Discard the supernatant, and then remove the tube from the magnetic stand.
9. Repeat steps 6-8 twice.

NOTE: Qbeads-Protein A contains 0.09% NaN₃, so we strongly recommend that wash the beads at least three times before use.

Binding of antibody

10. Mix appropriate amount of antibody (you may start from 2µg of antibody for the preliminary test) in 200 uL Washing buffer and transfer to the tube from step 9. Then vortex for 10 secs.
11. Incubate with tilt rotation for 30 minutes at room temperature.
12. Place the tube on the magnetic stand for 30-60 seconds to immobilize the beads at tube wall.
13. Discard the supernatant, and then remove the tube from the magnetic stand.
14. Add 200 uL Washing buffer and resuspend the beads by pipetting.
15. Place the tube on the magnetic stand for 30-60 seconds to immobilize the beads at tube wall.
16. Discard the supernatant, and then remove the tube from the magnetic stand.

17. Repeat steps 14-16 two more times to remove unbound antibody.

Immunoprecipitation for target antigen

18. Add at least 100 μ L cell lysate sample containing target antigen to the tube from step 17.

Then vortex for 10 secs.

19. Incubate with tilt rotation for 30 minutes at room temperature or at 4°C overnight.

20. Repeat steps 14-16 for three times.

21. If necessary, proceed to “SDS-PAGE analysis and Western Blot analysis” or “Elution of target antigen”.

SDS-PAGE and Western Blot analysis

1. Mix appropriate SDS-PAGE loading buffer to the beads from step.20, and heat to 95°C for 5 min.

2. Direct load the mix of Qbeads and buffer solution to the well of SDS-PAGE, then proceed to the standard SDS-PAGE and Western Blot analysis.

Elution of antibody / target antigen

1. Add 20 μ L elution buffer (e.g. 0.1 M Glycine-HCl, pH 2.0) after step.20 and gently mix with the Qbeads-antibody-antigen complex by pipetting.

2. Incubate with tilt rotation for 2 minutes at room temperature.

3. Place the tube on the magnet stand for 30-60 seconds.

4. **Collect the supernatant** to a clean tube, and then adjust the pH by adding 2 μ L neutralization buffer (e.g. 1M Tris-HCl, pH 8.5).

Trouble Shooting

Troubles	Solutions
Immunoglobulin binding is low.	<ol style="list-style-type: none">1. Make sure the Qbeads are suspended thoroughly before use.2. Mix Qbeads and sample thoroughly and continuously with either a tilt rotation device or a vortexer.3. Refer to Table 1 to match the binding preference of protein A with various Immunoglobins.4. Incubation time and temperature can be optimized depending on the sample column and affinity of antibody for target antigen.
Non-specific and background binding is high.	<ol style="list-style-type: none">1. Reduce the usage amount of Qbeads per test according to the binding capacity mentioned in “Binding Characteristics”2. After incubate Qbeads-antibody complex with antigen (step 19), increase wash procedures (step 20) to 5 times.3. Increase the concentration of Tween 20 to 0.1% in Washing buffer prior to elute the sample.4. Ensure remove washing buffer completely.

Qbeads do not collect on the magnet.	<ol style="list-style-type: none">1. Make sure the tube is directly contact with the magnet.2. Use Magdorf magnetic stand for best performance.
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Storage

Please keep the reagent at 2-8 °C . The validity is warranted for 12 months.

Contact Information

Please contact us when you have any question or comments via e-mail: info@magqu.com, or phone: +886-2-8667-1897.

Remarkable Notes

1. Please keep the reagent away from magnets during storage.
2. Do not freeze.
3. Qbeads-Protein A is for research use only.



MF-PRA-3000-04.02.2015



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Product Information

Magnetic Qbeads Series

Products	Cat. No.
Qbeads-Protein A	MF-PRA-3000
Qbeads-Protein G	MF-PRG-3000
Qbeads-NTA-Ni	MF-HIS-3000
Qbeads-Streptavidin	MF-STA-3000
Qbeads-Silica	MF-SIL-5010 MF-SIL-5024
Qbeads-Hydroxyl	MF-DEX-3000
Qbeads-Carboxyl	MF-COO-3000
Qbeads-Amine	MF-NHH-3000
Qbeads-Carboxyl Labeling Kit	KT-COO-3000-5SE

Accessory

Products	Description	Cat. No.
Magdorf	for 1.5 ml eppendorf tube	MDF-08
	for magnetic separating column	MSD-01
Magstand	for 15 ml falcon tube	MSD-15
	for 50 ml falcon tube	MSD-50
Magtractor	for 96-well culture plates	MTR-96
	for 24-well culture plates	MTR-24
	for 6-well culture plates	MTR-06

Magnetic NanoParticle Series

Products	Particle size	Cat. No.
Magnetic Fluid- Hydroxyl	30 nm	MF-DEX-0030
	60 nm	MF-DEX-0060
	90 nm	MF-DEX-0090
Magnetic Fluid- Carboxyl	30 nm	MF-COO-0030
	60 nm	MF-COO-0060
	90 nm	MF-COO-0090
Magnetic Fluid- Amine	30 nm	MF-NHH-0030
	60 nm	MF-NHH-0060
	90 nm	MF-NHH-0090
NanoQ-Carboxyl Labeling Kit	60 nm	KT-COO-0060-1SE

Fluorescent Magnetic Nanoparticles

Products	Particle size	Cat. No.
Blue FluoroNanoQ	60 nm	MF-FBL-0060
Green FluoroNanoQ	60 nm	MF-FGR-0060
Red FluoroNanoQ	60 nm	MF-FRE-0060

Customized Conjugation Service

Products	Particle size	Cat. No.
Customized conjugated magnetic beads	3 μ m	MF-CCS-3000
	30 nm	MF-CCS-0030
Antibody or peptide provided by customers (100 ug)	60 nm	MF-CCS-0060
	90 nm	MF-CCS-0090



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