Product Data Sheet 590

BioMag® anti-Human CD71

9025 Technology Dr. • Fishers, IN 46038-2886
800.387.0672 • 317.570.7020 • Fax 317.570.7034
info@bangslabs.com • www.bangslabs.com

Bangs Laboratories, Inc.

B E A D S ● A B O V E T H E R E S T ™

DESCRIPTION

The CD71 antigen is also known as the transferrin receptor. CD71 is expressed by activated T and B lymphocytes, macrophages, and erythroblasts. CD71 expression is upregulated on all proliferating cell types and is absent from resting blood leukocytes. A type II membrane glycoprotein, it is a disulfide-bound dimer of two identical subunits of 95 kDa. The N-terminal, intracytoplasmic domain of CD71, mediates rapid endocytosis and recycling.

BioMag® anti-Human CD71 particles recognize the 95 kDa transferrin receptor molecule present on proliferating cells including neoplastic cells, hematopoietic precursor cells, and activated lymphocytes. Research applications include the identification of proliferating neoplastic cells of the hematopoietic and other organ systems.

BioMag® anti-Human CD71 is a suspension of magnetic particles approximately 1.5µm in size. The suspension is supplied in a phosphate buffered saline (pH 7.5) containing EDTA, 1.0% BSA, and 0.1% sodium azide.

CHARACTERISTICS

Mean Diameter:	~1.5µm
Particle Concentration:	4 mg/mL
Particle Count:	1 x 10^8 BioMag® particles per mg

PROCEDURE

Researchers are advised to optimize the use of particles in any application.

Depending on antigen availability and the size of the target cell population, cell sorting applications may require up to 50-60 magnetic particles per cell. Magnetic particles and cells should be incubated at room temperature for 30-60 minutes in media containing 5-10% protein (to reduce nonspecific binding) for successful separation. Gentle end-over-end mixing or rocking during incubation is required for optimal results. (*Note*: Increasing the incubation time beyond one hour may be necessary to achieve the desired depletion.)

Some applications require the detachment of BioMag® antibody particles from cells after separation. One approach would involve culturing cells after positive selection. Cultures can be maintained for about 48 hours during which magnetic particles fall away from cells due to cell surface changeover. The magnetic particles are then easily removed via a magnetic separation. Another approach is the use of a protease, such as chymopapain, to break the antigen-antibody bond and remove the particles magnetically. Depending upon the application, it may not be necessary to remove the cells from the BioMag® particles. BioMag® particles are only 1-2µm in size and have been successfully used in FACS equipment. They will not jam the machine and are distinguishable from cells. Alternatively, negative selection approaches can be very effective in producing specific cell populations.

Cell sorting results using BioMag® anti-Human CD71 leukocyte particles for positive selection. *Typically, whole blood or purified leukocytes and particles are incubated for 30 minutes at room temperature and then magnetically separated. The supernatant is collected, incubated with the appropriate two-color antibody cocktail, and then analyzed by flow cytometry. Figure A-1 depicts the lymphocyte cell population prior to positive selection. Figure A-2 depicts the lymphocyte cell population after positive selection. Figures B-1 and B-2 depict the results for monocytes. Figures C-1 and C-2 depict the results for neutrophils. The bead:cell ratios reported above are based on experiments where cells were exposed to the particles once.*

** The values under "General Recommendation" should be used as a starting point in optimizing experimental protocols. Due to differences in the distribution of cell types in samples and other variables, the researcher is strongly encouraged to determine the optimal particle to cell ratios for their experiments.*

Figure A-1

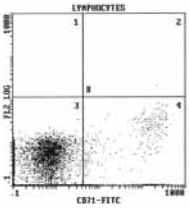
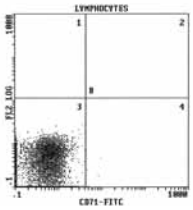


Figure A-2



General Recommendation:

Conc. #	4.0 x 10^8 particles/mL
Volume Used	0.025mL
# Particles	1.00 x 10^7 per test
# Target Cells	4.69 x 10^5 per test
Particle:Target Cell Ratio	21.3
% Depletion	97.4%

Figure B-1

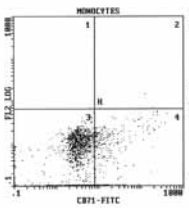
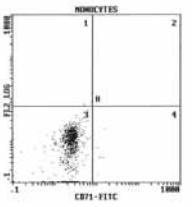


Figure B-2



General Recommendation:

Conc. #	4.0 x 10^8 particles/mL
Volume Used	0.025mL
# Particles	1.00 x 10^7 per test
# Target Cells	1.87 x 10^5 per test
Particle:Target Cell Ratio	53.4
% Depletion	97.27%

Figure C-1

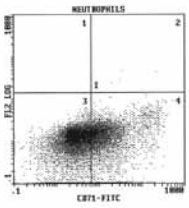
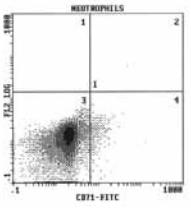


Figure C-2



General Recommendation:

Conc. #	4.0 x 10^8 particles/mL
Volume Used	0.050mL
# Particles	2.00 x 10^7 per test
# Target Cells	2.92 x 10^6 per test
Particle:Target Cell Ratio	6.8
% Depletion	96.52%

STORAGE AND STABILITY

Store at 2-8˚C. Freezing, drying, or centrifuging BioMag® may result in irreversible aggregation and loss of binding activity. Washing BioMag® anti-Human CD71 particles in sterile media to remove preservative prior to use is recommended. Using a magnetic separation unit for washing instead of centrifugation is also strongly recommended.

SAFETY

This particle suspension contains sodium azide. Sodium azide may react with lead and copper plumbing to form explosive metal azides. Upon disposal of

material, flush with a large volume of water to prevent azide accumulation. Please consult the Material Safety Data Sheet for more information.

These products are for research use only and are not intended for use in humans or for *in vitro* diagnostic use.

ORDERING INFORMATION

Cat. Code	Description	Size
BM590	BioMag® anti-Human CD71	5mL

Order online anytime at **www.bangslabs.com.**