

List of available fluorochromes widely used for flow cytometry and fluorescence microscopy

Fluorochrome	Excitation max. (nm)	Laser (nm)	Emission max. (nm)
Cascade Blue®	377	405	420
DyLight™ 405	400	405	420
Alexa Fluor® 405	402	405	421
Pacific Blue™	410	405	455
AmCyan	458	405	489
DyLight™ 488	493	488	518
Alexa Fluor® 488	495	488	519
FITC (Fluorescein isothiocyanate)	493	488	525
Cascade Yellow™	400	405	550
Pacific Orange™	400	405	551
Cy™3	514	408, 514	566
PE (R-phycoerythrin)	496, 565	488	575
TRITC (Tetramethyl rhodamine isothiocyanate)	557	488	576
DyLight™ 549	562	488	576
PE-Texas Red®	496, 565	488	613
Texas Red®	596	488	615
PE-DyLight™ 594	496, 565	488	618
DyLight™ 594	593	488	618
APC (Allophycocyanin)	645	633	660
Cy™5	649	633	666
Alexa Fluor® 647	650	633	668
PE-Cy™5	496, 565	488	670
PE-DyLight™ 649	496, 565	488	673
DyLight™ 649	654	633	673
PerCP (Peridinin-chlorophyll A protein)	482	488	675
PerCP-Cy™5.5	482	488	690
PE-Cy™5.5	496, 565	488	690
APC-Cy™5.5	650	633	690
Alexa Fluor® 700	696	633	719
PE-Cy™7	496, 565	488	774
APC-Cy™7	650	633	774
APC-Alexa Fluor® 750	650	633	774
PE-DyLight™ 750	496, 565	488	778
APC-DyLight™ 750	650	633	778
DyLight™ 750	752	Xenon Arc Lamp	778
DyLight™ 800	777	Xenon Arc Lamp	794

Please note that little variations in values of the indicated wavelengths occur in the literature and promotional brochures.

Cascade Blue®, Cascade Yellow™, Alexa Fluor®, Texas Red®, Pacific Blue™, Pacific Orange™ are trademarks of Molecular Probes, Inc. (a wholly owned subsidiary of Invitrogen Corporation); DyLight™ is a trademark of Thermo Fisher Scientific; Cy™ is a trademark of Amersham Biosciences Corp (a company owned by GE Healthcare).

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For twenty years EXBIO has been a primary manufacturer of high quality monoclonal antibodies. Our experienced staff is pleased to offer the resources available from EXBIO including our quality certified facilities, time saving expertise and proprietary techniques to researchers worldwide through our selection of

Custom Services

Monoclonal Antibody Production

Custom Conjugation Services

Hybridoma Development

Rabbit Polyclonal Antibody Production

Determination of Mouse Antibody Isotype

Detection of Mycoplasma Contamination

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Custom Conjugation Services

Catalog Nº	Service	Unit	Price
CC001	Conjugation with FITC	1–5 mg	€ 220
CC002	Conjugation with PE	1–5 mg	€ 440
CC003	Conjugation with APC	1–5 mg	€ 440
CC004	Conjugation with PerCP	1–5 mg	€ 550
CC005	Conjugation with biotin	1–5 mg	€ 220
CC006	Conjugation with HRP	1–5 mg	€ 440
CC007	Conjugation with small organic molecule*	1–5 mg	€ 220
CC008	Conjugation with a tandem dye*	1–5 mg	€ 550

* Customer may be required to supply commercially available small organic fluorochrome to prepare the requested conjugate.

Select one of the colors we offer and let EXBIO conjugate the protein or antibody of your choice. We offer conjugation of any antibody from our catalog with a broad range of fluorochromes or other molecules according to your request. We can also custom conjugate your own antibodies using our successful conjugation processes:

Conjugation with low-molecular weight fluorochromes, biotin or horseradish peroxidase (HRP):

- **Receipt** of antibody or other protein from a customer.
- **Dialysis** of the antibody or other protein.
- **Activation of HRP** and transfer to the conjugation buffer by gel filtration (only Cat. Nº CC006).
- **Conjugation.**
- **Removal** of unconjugated fluorochrome, biotin or HRP.
- **Stabilization** of the conjugate and **final adjustment.**
- **QC tests:** Spectrophotometric determination of antibody concentration and F/P ratio (F/P ratio not determined in case of biotin or HRP).
- **Delivery** of the conjugate to the customer.

Conjugation with high-molecular weight fluorochromes (phycobiliproteins or tandem dyes):

- **Receipt** of antibody or other protein from a customer.
- **Activation** of fluorochrome and the antibody or other protein.
- **Transfer to the conjugation buffer** by gel filtration.
- **Conjugation.**
- **Purification** of conjugate by size-exclusion chromatography (optimally conjugated fraction is isolated).
- **Stabilization** of the conjugate and **final adjustment.**
- **QC tests:** Spectrophotometric determination of antibody concentration.
- **Delivery** of the conjugate to the customer.

Monoclonal Antibody Production

Catalog No.	Service	Price (€/mg of antibody)				
		10–20 mg	21–50 mg	51–100 mg	101–200 mg	200 mg +
CM-IgG	Production of IgG monoclonal antibody	€ 40	€ 30	€ 25	€ 20	Inquiry
CM-IgM	Production of IgM monoclonal antibody	€ 50	€ 40	€ 30	€ 25	Inquiry

Reliable and cost effective, EXBIO is ready to maintain your hybridoma and produce on demand your mouse, rat, or hamster monoclonal antibody in bulk quantities.

You supply the hybridoma and we will produce the antibodies following a proven process that yields the highest quality results:

- **Receipt** of hybridoma cells from a customer.
- **Propagation** of the hybridoma and **testing** for the presence of **mycoplasma**, determination of **antibody concentration** in a culture supernatant.
- **Cryopreservation** of 2 vials of the hybridoma (back-up storage).
- **Production** of hybridoma culture supernatant (concentrated by tangential flow ultrafiltration).

• **Antibody purification:**

- IgG by affinity chromatography (protein A or protein G) or by HCIC (Hydrophobic Charge Induction Chromatography)
- IgM by precipitation methods and hydroxyapatite ion-exchange chromatography.
- **Final adjustment** (dialysis, concentration and filter-sterilization).
- **QC tests:**
 - antibody concentration by spectrophotometric analysis (A280 nm)
 - purity of antibody by SDS-PAGE analysis
- **Delivery** of the antibody to the customer.

Rabbit Polyclonal Antibody Production

Catalog Nº	Service	Unit	Price
RB001	Production of Rb PAb – crude antiserum	1 rabbit	€ 280
RB002	Production of Rb PAb – purified IgG	1 rabbit	€ 375

Cost effective, high quality service – if you do not require monoclonals, we will prepare a polyclonal antibody raised in rabbits. We use following process:

- **Receipt** of the antigen from a customer.
- Collection of **pre-immune serum.**
- **3 immunizations** of the rabbit; collection of **serum samples** (2–5 ml) after 2nd and 3rd immunization dose.
- Application of **booster dose.**
- Collection of **antiserum** (60–80 ml).

• **Delivery** of the antiserum to the customer (Cat. Nº RB001).

or

- **Purification** of IgG fraction by protein A – affinity chromatography.
- **Final adjustment** (dialysis, filter-sterilization)
- **QC tests:**
 - antibody concentration by spectrophotometric analysis (A280 nm)
 - purity of antibody by SDS-PAGE analysis
- **Delivery** of the antibody to the customer (Cat. Nº RB002).

Hybridoma Development

Catalog Nº	Service	Unit	Price
HY001	Hybridoma development		Inquiry

Ask the experts at EXBIO to develop and produce novel monoclonal antibodies to meet your specific needs.

With our effective immunization techniques, proprietary one-step hybridoma cloning process, and the use of semi-solid growth media we will generate a new monoclonal antibody for you. We utilize the following time-saving process:

- **Receipt** of the antigen from a customer.
- **Immunization** of 4 balb/c mice by 2 different immunization protocols.
- **Monitoring and evaluation** of the immune response, **selection** of appropriate mouse.

- **Fusion** of splenocytes with Sp2/0 myeloma cells.
- **Seeding** of the fusion mixture into semi-solid hybridoma cloning and growth media.
- **Harvesting** of up to 576 hybridoma colonies (6×96 well plates), **propagation** and **screening** by antigen-coated ELISA.
- **Selection and expansion** of up to 10 ELISA-positive clones; delivery of 2 ml of **tissue culture supernatant** corresponding to selected clones to the customer for further testing.
- **Delivery** of the chosen hybridomas to the customer.

Determination of Mouse Antibody Isotype

Catalog Nº	Service	Unit	Price
AD001	Determination of mouse immunoglobulin isotype	1 sample	€ 30

We determine mouse immunoglobulin isotypes by multiplexed particle-based flow cytometric assay. This procedure employs a series of spectrally discrete beads, each coated with particular goat antibodies specific to the respective mouse antibody isotype. Captured mouse antibody is revealed with fluorochrome-labeled secondary antibodies against mouse κ or λ light chain. Mixture of beads is analysed by flow cytometry.

Therefore, both heavy and light chains of tested monoclonal antibody are identified in a single analysis using following process:

- **Receipt** of an antibody (tissue culture supernatant) from a customer (alternatively, antibodies resulting from Hybridoma Development project are analysed).
- **Analysis** of the sample by multiplexed flow cytometry bead assay.
- **Report** of the results to a customer.

Detection of Mycoplasma Contamination in Cell Culture

Catalog Nº	Service	Unit	Price
AD002	Mycoplasma testing	1 sample	€ 20

We use a highly sensitive PCR procedure for detection of traces of Mycoplasma contamination in cell cultures. We utilize primers complementary to conserved regions of 16S rRNA that were carefully selected for *M. hyorhinis*, *M. arginini*, *M. orale*, *M. fermentas* and *M. laidlawii*, species proved to be responsible for 98% of Mycoplasma contamination. The genus-specific primers also react with members of the genera *Acholeplasma*, *Ureaplasma* and *Spiroplasma*.

Our well established procedure has been validated against commercial PCR kits and fluorescent microscopy-based methods. Get your results reliably and fast with the following process:

- **Receipt** of a cell line from a customer.
- **Sample Analysis**
- **Report** of the results to a customer.

