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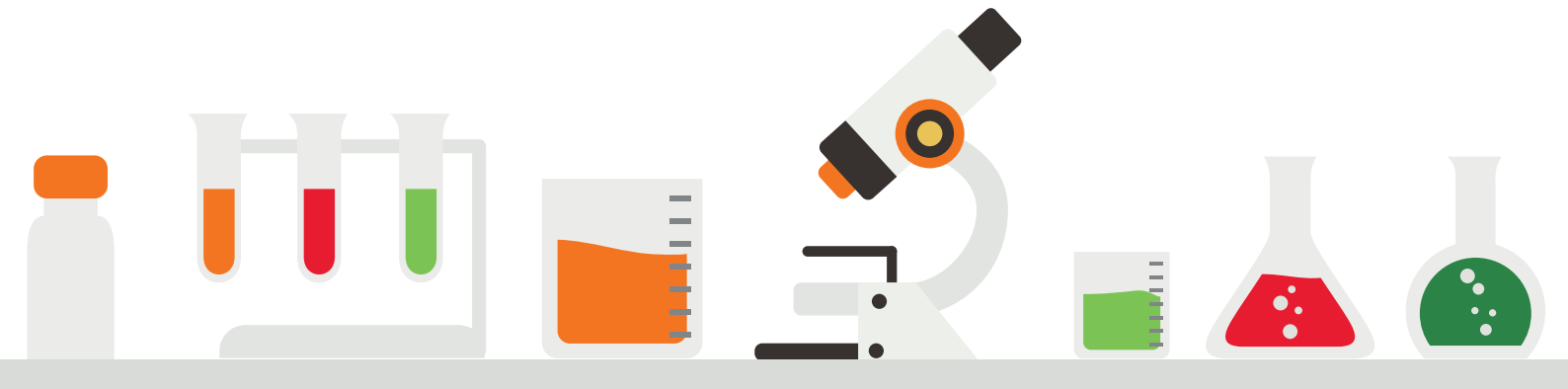

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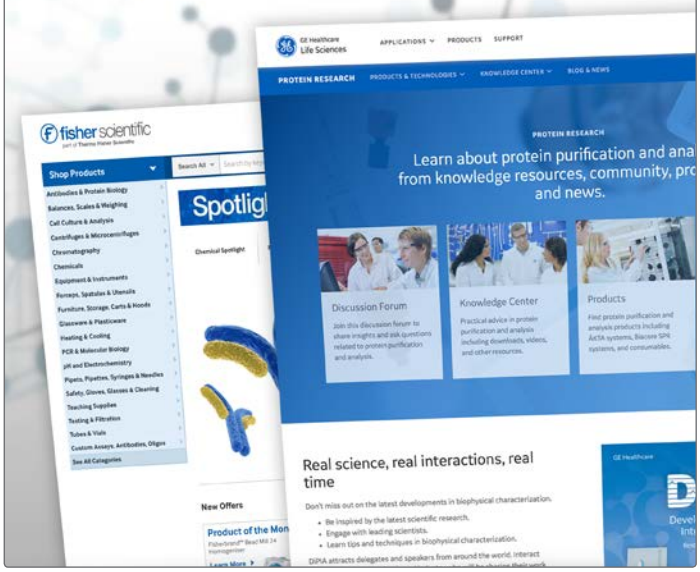

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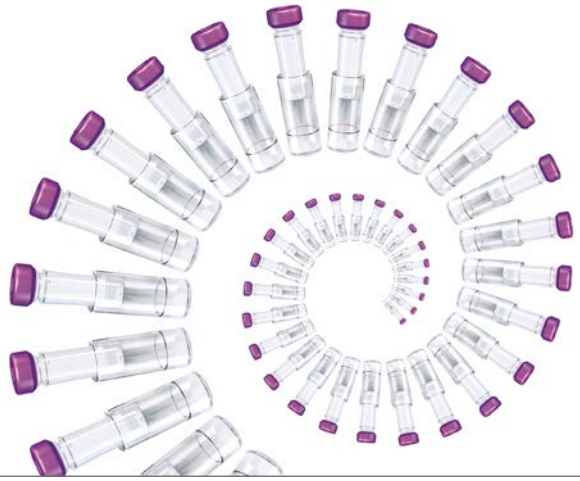

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A call to arms: Scientists use biotech tools to crack open Zika's genetic code

Researchers still must answer many of the most fundamental questions relating to the virus, and support for their efforts is coming from some unlikely places. By GE Healthcare.

Zika, which can cause birth defects in pregnant women, took six decades to cross from Africa to Brazil. But, in 2016, it apparently took just 60 weeks for the virus to reach South Florida, where 15 locally acquired cases were reported.

Zika's American journey began on March 2, 2015, when Brazilian authorities put in a call to World Health Organization officials. Doctors in the country's northeastern states had reported thousands of cases of an unidentified illness with a characteristic skin rash, some with fever and some without. National labs had analyzed blood samples from a number of patients, and only a tiny portion came back positive for dengue, a mosquito-borne virus that elicits similar symptoms. Subsequent tests for other suspects—chikungunya, enterovirus, measles, rubella and parvovirus B19—were all negative.

It was enough to make even a jaded epidemiologist raise an eyebrow. Was there a new microscopic predator hunting in South America's most populated country? The answer came quickly. On April 29, a state lab confirmed 16 cases of Zika infection, the first known local acquisition and transmission of the virus in the Americas. Previously seen only in Africa and Asia, the flavivirus first discovered in a Ugandan forest in 1947 had finally reached the Americas.

The effort to understand and fight Zika is happening in earnest both in the Americas, where the latest outbreak is centered, and in other parts of the world–including at GE Healthcare labs in Cardiff, Wales, more than 5,000 miles from the shores of Brazil. The 30-acre site, set along the River Taff and sandwiched between two golf clubs, supplies Zika sleuths with chemicals that help them isolate and analyze the virus' RNA blueprints.

To date, the virus has spread to 67 countries and territories by catching a lift on the Aedes aegypti mosquito, the same species that transmits dengue fever and other tropical diseases, and through sex. It may have infected over a million people in the region, though many may not have reported it because associated symptoms of fever, rash and pink eye are mild or nonexistent. But other worrying symptoms—that the virus might be linked to a birth defect of incomplete brain development called microcephaly and adult-onset Guillain–Barré syndrome, where the immune system attacks the nervous system—led the WHO to declare a public health emergency of international concern.

"*The more we know the worse things look*", said Dr. Margaret Chan, WHO's director-general. "*A pattern has emerged in which initial detection of virus circulation is followed, within about three weeks, by an unusual increase in cases of Guillain-Barré syndrome. Detection of microcephaly and other fetal malformations comes later, as pregnancies of infected women come to term.*"

As of June 1, 60 countries and territories were still reporting ongoing Zika transmission. Eleven countries or territories worldwide had reported cases of microcephaly or other central nervous system malformations, and 13 had reported increased rates of Guillain–Barré syndrome. "*At this stage, based on the evidence available, WHO does not see an overall decline in the outbreak*", the organization said in its most recent situation report.

Nobody has yet developed a vaccine to prevent infection or a cure once the virus has been transmitted to a person. To date, efforts to stop it have focused on educational campaigns for people in areas where the mosquito carrier lives, instructing them to wear insect repellent, put up window screens and remove standing water from around their homes. Other efforts involve mosquito-limiting efforts such as spraying and experimental population control.

The most high-tech methods focus on targeting Zika's genetic materials.

Scientists collecting mosquito samples in the field use a special fiber paper manufactured by GE in Cardiff. This coffee-filter-like material, called Whatman™ FTA™ paper (see page 5), has magical properties if you're a microbiologist. It is chemically treated to break down cells and destroy proteins that would otherwise damage the DNA that sticks to its fibers. The result: squishing a mosquito in the paper lets researchers transport samples without refrigeration over long distances, a requirement if you happen to be in the middle of the Amazon.

Identifying patients who have been infected with the virus is a difficult process only recently made possible by modern technology. One method, called the polymerase chain reaction (PCR†), rapidly produces multiple copies of the virus' RNA so that medical workers can identify it in a patient's blood. The Cardiff facility produces PCR beads that Zika researchers with the U.S. Army are using to study infection.

Much still needs to be done to contain and then eradicate the Zika epidemic. Rapid diagnostic tools are being developed on a massive scale, as are potential vaccines. All will take time. But for many at the GE Healthcare Cardiff facility, contributing to efforts to help end the disease brings a sense of pride in their work.

"*For the people making and boxing the papers, PCR beads and other lab consumables, these products might not normally look like much more than water in a tube or coffee filters*", says Michael Igoe, the site's Product Operations Manager. "*But hidden in the tube and the paper are complex products that are providing a huge benefit to communities. We're making products that make a real difference in the world. They have meaning.*"

Read full article and blogs ***here****.*

Featured products

illustra™ PuReTaq Ready-To-Go™ PCR beads

Premixed, predispensed, single-dose reactions optimized for performing standard PCR amplifications.

- Save time: simply add template DNA solution and primers, and cycle.
- More reproducible results: less risk of pipetting errors and contamination.
- High-quality PuReTaq DNA polymerase and high-purity reagents: robust performance and the lowest possible levels of contaminating DNA.
- Long-term ambient-temperature stable: no freezer space required; less energy consumption for shipping and storage.
- Verified for real-time PCR.
- Available in 96 well plates and Hot Start Mix.

illustra ExoProStar™ reagent

ExoProStar 1-Step PCR and sequence reaction clean-up reagent provides a simple, single-step method for removing unincorporated primers and nucleotides prior to downstream analysis. illustra Exonuclease I and Alkaline Phosphatase enzymes are combined in a single, stable mix requiring just one pipetting step in a 30 min protocol to establish the clean-up reaction.

ExoProStar S is an enzymatic PCR clean-up technology with Exonuclease I and Shrimp Alkaline Phosphatase to remove unincorporated primers and dNTPs. For maximum flexibility it comes in two separate tubes; just two simple pipetting steps are needed to prepare the reaction. ExoProStar S is easy to automate and provides complete heat inactivation of the enzymes within 10 min in a fast 15 min protocol. It is scalable for different reaction sizes and easy to automate.

illustra TempliPhi™ amplification kits

Prepare DNA sequencing templates from circular DNA for cycle sequencing, cloning, and transformation in 4 to 6 hours.

- Use simple protocol to reduce time, labor, and quantity of consumables needed for template preparation–workflow enables easy automation.
- Use amplified DNA directly for cycle sequencing without purification.
- Amplify DNA from bacterial or M13 liquid cultures, colonies, plaques, glycerol stocks, or purified circular (plasmid or M13) DNA.

Easy sample collection and storage using FTA and CloneSaver™ cards.
Click **here** to learn more.

Ordering information

Chemistry	Format	Description	Volume	Pack size	Item	Price* (£)
PuReTaq DNA polymerase	Multiwell plate	illustra PuReTaq Ready-To-Go PCR beads	96 rxns	1/pk	27955701	109.66
PuReTaq DNA polymerase	Multiwell plate	illustra PuReTaq Ready-To-Go PCR beads	5 x 96 rxns	1/pk	27955702	496.85
PuReTaq DNA polymerase	0.5 mL tubes	illustra PuReTaq Ready-To-Go PCR beads	100 rxns	1/pk	27955801	119.69
PuReTaq DNA polymerase	0.2 mL hinged tube with cap	illustra PuReTaq Ready-To-Go PCR beads	96 rxns	1/pk	27955901	110.79
Enzymatic PCR clean-up technology	Kit	illustra ExoProStar 1-Step	100 rxns	1/pk	US77702	66.00
Enzymatic PCR clean-up technology	Kit	illustra ExoProStar 1-Step	500 rxns	1/pk	US77705	260.63
Enzymatic PCR clean-up technology	Kit	illustra ExoProStar S	100 rxns	1/pk	US79010	52.15
Enzymatic PCR clean-up technology	Kit	illustra ExoProStar S	500 rxns	1/pk	US79050	205.51
Phi29 DNA polymerase	Kit	illustra TempliPhi amplification kit	100 rxns	1/pk	25640010	185.83
Phi29 DNA polymerase	Kit	illustra TempliPhi amplification kit	500 rxns	1/pk	25640050	895.28

*For more information please visit **eu.fishersci.com***

Serum alternatives to fetal bovine serum in cell culture

Serum is often a necessary component of cell culture. Fetal bovine serum (FBS) has long been the first serum of choice for researchers. Although FBS performs well, there are circumstances where FBS replacements might offer advantages with regard to cost of sera, variability in supply, lot-to-lot variability in composition, or performance with specific cell types. This study examines the performance of FBS and seven serum alternatives with six cell lines.

Key concepts and findings

- Comparisons were made with the FBS control condition as a base standard, using a ratio of cell counts.
- Available FBS replacements are shown to work with six cell lines.
- The FBS replacements have advantages over FBS and provided equivalent or better cell growth compared with FBS (results are cell-line dependent).

Methodology

Six cell lines and eight serum types were used. All cultures were grown in T-25 cell culture flasks in 10 mL of corresponding media supplemented with 10% serum. Control FBS was prepared by pooling many lots of FBS. HyClone™ FetalClone™ sera are blends of FBS and specially processed calf serum formulated to reproduce the composition of FBS. FetalClone I, II, and III are optimized for hybridoma, CHO, and fibroblast cells, respectively. Iron-Supplemented Calf Serum is produced from formula-fed veal animal serum supplemented with physiological levels of iron, and contains high levels of transferrin. Both US and New Zealand HyClone Cosmic Calf™ sera, US and New Zealand origin, are based on Iron-Supplemented Calf Serum with additional growth promoting factors. HyClone Bovine Growth Serum is also based on Iron-Supplemented Calf serum with additional trace elements, vitamins, and growth factors.

All conditions consisted of three serum lots to test lot-to-lot consistency, except control FBS of which one lot was used, and supplementation of MRC-5 and AIF cells using Cosmic Calf, New Zealand Origin where two lots were used. Flasks were seeded at 3000–5000 cells/cm^2, incubated in 5% CO_2/95% air at 37°C, and checked daily for confluency. When any culture reached confluency, all cultures were trypsinized and counted. Cell counts were normalized to the FBS control as percentages such that the FBS control is always 1.0 or 100%. Conditions that produced more cells than the control have values greater than 1.0.

It was necessary to define a ratio at which condition performance was comparable with or better than control FBS. A value of 0.90 or 90% was chosen to accommodate experimental variations in harvesting and counting.

Results and discussion

Results were cell line-dependent with certain FBS replacements proving to be more or, sometimes, less suitable for specific cell lines. In nearly all cases, cell growth in at least one of the FBS replacements was equal to or greater than cell growth in FBS. This finding indicates that researchers have viable FBS alternatives for replacements in their cell cultures.

MRC-5 cells grew more rapidly, and thus to higher yields, in FetalClone III and Bovine Growth Serum than in FBS. In comparison with cell yields in FBS, Vero cell yields were at least as high in FetalClone II, FetalClone III, New Zealand Cosmic Calf, Iron-Supplemented Calf, US Cosmic Calf, and Bovine Growth Serum. The rate of BHK-21 cell growth was about the same in FBS, FetalClone II, FetalClone III, and U.S. Cosmic Calf Serum, while the cell growth was more rapid in Bovine Growth Serum.

FetalClone II is optimized for CHO cells, as are the Cosmic Calf sera. Supplementation with FetalClone II, FetalClone III, New Zealand Cosmic Calf and US Cosmic Calf each resulted in higher yields of CHO cells than did FBS. Bovine Growth Serum performed comparably to FBS.

AIF cells were used as a model for conventional hybridoma cell lines. All sera tested supported hybridoma cell growth rates equal to or higher than with FBS. However, for many hybridoma applications, the lower IgG levels in FBS and FetalClone I make these the preferred sera for monoclonal antibody production.

NS0 cultures in FetalClones I, II, III, New Zealand Cosmic Calf, Iron-Supplemented Calf, and US Cosmic Calf serum showed growth equal to or better than that of growth in FBS.

Study conclusions

This study has shown that multiple sera are available as potential replacements for FBS in cell culture. A variety of mammalian cell types (fibroblasts, hybridoma, myeloma) were used in the study, and each type was shown to have a potential FBS replacement in at least one bovine calf-based serum. Some advantages of the tested calf-based sera compared with FBS are lower cost, higher availability, and perhaps more consistent component levels due to the methods used in the veal industry. Animal age at time of slaughter, stress on the animals, breed, and diet are factors that can contribute to the consistent component levels in calf sera compared with the same composition in fetal bovine serum.

Request a copy of data file 29194816 AA—Serum alternatives to fetal bovine serum in cell culture

Featured products

HyClone FetalClone I, II, III

FetalClone engineered serum products are economical alternatives to fetal bovine serum (FBS), commonly used in bioprocessing applications as a supplement to enrich cell culture performance. FetalClone products have demonstrated performance with a variety of cell lines, including hybridomas, CHO, BHK-21, NS0, MRC-5, and Vero cells.

HyClone Cell Culture Media for monoclonal antibody (mAb) and recombinant protein production

These serum-free basal media are designed to be utilized with common protein-producing cell lines, such as CHO and HEK293. See below table for animal-derived component-free (ADCF), chemically-defined (CD), and protein-free formulations for use with your cell line of interest.

CHO cells	PF-CHO	SFM4CHO	CDM4CHO	
	HyCell™ CHO	HyCell TransFx-C	ActiPro™	ActiSM™
Hybridoma/ myeloma	CDM4MAb	CDM4PERMAb	SFM4MAb	CDM4NS0
Insect cells	SFX-Insect	SFM4Insect		
Viral vaccines	SFM4Megavir	CDM4Avian		
HEK293/PER.C6	CDM4PerMAb	CDM4HEK293	SFM4HEK293	TransFx-H
Stem cells	HyCell STEM	AdvanceSTEM™		

HyClone Classical Media	
BME/EBSS	MEM/EBSS
DMEM	DMEM/F12
Ham's F10	Ham's F12
Iscove's (IMDM)	Leibovitz (L-15)
McCoy's 5A	Medium 199
MEM	RPMI 1640

Media for transfection and transient expression

Our transfection media have been tested with a wide range of HEK293 and CHO cell lines and support high transfection efficiency using lipid-mediated, polymer-mediated and other transfection methods. HEK293 transfection medium is suitable for adenoviral (AdV), adeno-associated viral (AAV), lentiviral, retroviral and recombinant protein production. CHO transfection medium is suitable for recombinant protein and MAb production.

Try Whatman syringe filters to prepare your sample.

Click **here** for further information.

Ordering information

Product type	Format	Description	Volume	Pack size	Item	Price* (£)
Serum	Bottle	HyClone FetalClone I	500 mL	1/pk	SH30080.03	292.00
Serum	Bottle	HyClone FetalClone II	500 mL	1/pk	SH30066.03	292.00
Serum	Bottle	HyClone FetalClone III	500 mL	1/pk	SH30109.03	292.00
Media	Bottle	ADCF mAb with L-glutamine, HEPES, w/o phenol red	500 mL	1/pk	SH30349.01	22.62
Media	Bottle	HyClone SFM4HEK293	1000 mL	1/pk	SH30521.02	44.30
Media	Bottle	HyClone SFM4CHO w/o L-glutamine	1000 mL	1/pk	SH30548.02	38.35
Media	Bottle	HyClone SFM4TransFx-293	1000 mL	1/pk	SH30860.02	44.20
Media	Bottle	HyClone HyCell TransFx-H for HEK293 cells	1000 mL	1/pk	SH30939.02	50.82
Media	Bottle	HyClone HyCell TransFx-C transfection media	1000 mL	1/pk	SH30941.02	50.72

*For more information please visit **eu.fishersci.com***

Save time in HPLC prep

Sample filtration protects your HPLC instrument and column while preserving data quality. Read our tips on using multilayer and all-in-one filter units to save time and improve lab efficiency.

If you analyze large numbers of samples using high-performance liquid chromatography (HPLC), sample preparation can take up a lot of your time. Filtering samples before HPLC can help avoid frit clogging while maintaining data quality.

So, what can you do to simplify and speed up the process? Read on to find out!

Try a stacked syringe filter

Syringe filtration often involves aspirating the sample, fitting a particle filter, filtering into an autosampler vial, capping, and finally transferring the vial to an auto-sampler. You might repeat this process dozens of time a day, depending on your circumstances.

If you have difficult-to-filter samples, you might find that high particulate samples can take more time to filter. To help with this, stacked filter devices have multiple layers of filtration, starting with larger pore sizes and going down to the desired pore size.

This approach traps large particles first, and successively traps smaller particles. The device does not get clogged as easily as devices with a single membrane, making filtration faster and easier.

Go syringeless

If your samples are reasonably easy to filter, a syringeless filter option simplifies the process greatly.

Using a standard syringe filter involves at least four individual components, five if you include the initial sample storage vial. When you have dozens (or hundreds!) of samples to filter, the multi-step workflow is time consuming and can lead to sample loss.

In a syringeless filter, the filter membrane, pre-filtration chamber, post-filtration storage vial, and cap are all part of one device. This design streamlines HPLC sample prep and minimizes the number of consumables. Filtration can be performed 3 times faster than with syringe filters.

Using a syringeless filter means that you only need to add the sample to the outer chamber, place the plunger, and push. The inner storage vial holds your filtered sample ready for analysis, so it can go directly into your autosampler. Construction can be either polypropylene or glass and the vial can be either clear or amber colored depending on the requirements around your sample.

Broaden your solvent compatibility

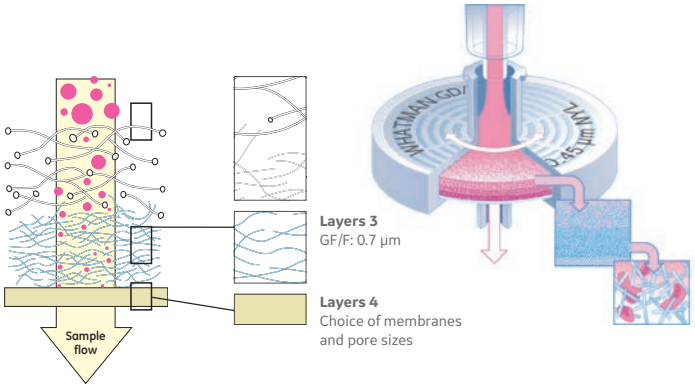
When your lab prepares a wide variety of sample types using different solvents for HPLC analysis, identifying appropriate membrane materials can be time-consuming. Different materials might be more or less suitable for a given sample based on chemical compatibility and solvent resistance.

If you want to make filtration easier, you could try out a material with broad solvent compatibility. Regenerated cellulose (RC), for example, is well suited for both hydrophilic and hydrophobic solvents. Using RC for most or even all your samples can reduce time spent researching and selecting materials.

Use tools to boost throughput

A multi-compressor can save time when using syringeless units. Filtering multiple samples simultaneously with a dedicated tool can also reduce hand strain.

Try our Whatman Filter Selector App to find out if you are using the most appropriate filtration solution for your samples. To discuss any challenges you are facing, please contact GE's Life Sciences Scientific Support team.



Whatman GD/X stacked syringe filter

Mini-UniPrep syringeless filters

For more information please visit ***eu.fishersci.com***

Featured products

Mini-UniPrep™ syringeless HPLC filters

Whatman Mini-UniPrep syringeless filters integrate an autosampler vial, filtration membrane, plunger, and cap/septa into one consumable product. They are built for fast and easy HPLC/UHPLC sample preparation.

- 0.2 µm and 0.45 µm pore sizes available to meet sample requirements.
- Housing options: amber to prevent photodegradation of light-sensitive samples, or translucent for easy visual inspection.
- A borosilicate glass vial version Mini-UniPrep G2 is available for eliminating plastic-based leachables that can originate from a polypropylene vial.
- Compatible with most major autosamplers for high throughput analysis.
- All-in-one filtration device for quick and cost-effective sample processing.

Whatman GD/X™ syringe filters

These filters are specifically designed for filtration of viscous or otherwise hard-to-filter samples with high solids content.

- High loading capacity for samples with high solids content.
- Three layer glass fibre prefiltration stack for filtering larger sample volumes with less back pressure build-up.
- Process three to seven times more sample volume than filters without prefilter.

Grade 3MM Chr cellulose chromatography papers

Grade 3MM Chr cellulose chromatography filter is a 0.34 mm thickness paper for general chromatography and electrophoresis.

- Pure cellulose produced entirely from the highest quality cotton linters with no additives of any kind.
- Manufactured and tested specifically for chromatographic techniques. This ensures the wicking capability and uniformity of capillary action that are important in chemical separations.
- Also widely used in protein and nucleic acid blotting.

Learn more about how you can add more security to your ÄKTA™ chromatography system runs by using our new:

Protein Prep syringe filter for ÄKTA systems – **see page 12**

Ordering information

Membrane	Format	Description	Format/pore size	Pack size	Item	Price* (£)
RC	Non sterile	Mini-UniPrep syringeless filter	0.2 µm	100/pk	UN203NPERC	166.42
RC	Non sterile	Mini-UniPrep syringeless filter	0.45 µm	100/pk	UN203NPURC	166.42
GMF	Non sterile	Mini-UniPrep syringeless filter	0.45 µm	100/pk	UN203NPUGMF	166.42
PES	Non sterile	Mini-UniPrep amber syringeless filter	0.2 µm	100/pk	UN203APEPES	172.00
PES	Non sterile	Mini-UniPrep amber syringeless filter	0.45 µm	100/pk	UN203APUPES	172.00
PES	Non sterile	Whatman GD/X syringe filter	25 mm, 0.2 µm	150/pk	6876.2502	342.62
PES	Non sterile	Whatman GD/X syringe filter	25 mm, 0.45 µm	150/pk	6876-2504	342.62
PVDF	–	Mini-UniPrep G2 amber syringeless filters	0.2 µm	100/pk	GN203APEAQU	337.00
Cellulose	Circles	Grade 3MM Chr cellulose chromatography paper	2.4 cm dia	100/pk	1030-024	10.23
Absorbent	Sheets	Benchkote™ surface protector	46 cm × 57 cm	50/pk	2300-916	75.11

*For more information please visit **eu.fishersci.com***

New lab start-up programme

Visit the following link to find out more about the GE product offering in the Fisher new lab start-up programme:

eu.fishersci.com/go/nlsu

Making mAbs purification efficient and cost-effective

Monoclonal antibodies (mAbs) are the largest and fastest growing class of biological drugs today. As a result, there is high demand for solutions that will deliver efficient, flexible, and cost-effective purification.

Downstream mAb purification platforms commonly include a protein A-based capture step followed by one or two polishing steps to remove remaining impurities. This offers high purity and a high degree of recovery in a single capture step.

Capture using protein A ligands

MabSelect™ is our protein A affinity family of resins designed for capturing mAbs from large sample volumes. The resins are based on a highly cross-linked agarose matrix with a recombinant protein A ligand. The figure below shows available MabSelect resins and contains suggestions for best use.

Optimizing cleaning and sanitization for improved process efficiency

Efficient cleaning prevents impurities from building up on the chromatography column and reducing the capacity of the resin. Efficient cleaning and sanitization protocols also help prevent growth of microorganisms and inactivate potential endotoxins. A high alkaline stability of the resin enables the use of high concentrations of low-cost sodium hydroxide as a cleaning agent. MabSelect PrismA exhibits more than 95% retained dynamic binding capacity after cleaning with 0.5 M NaOH between runs for 150 cycles (see figure) and still 90% with 1.0 M NaOH.

Polishing mAbs in a two-step process

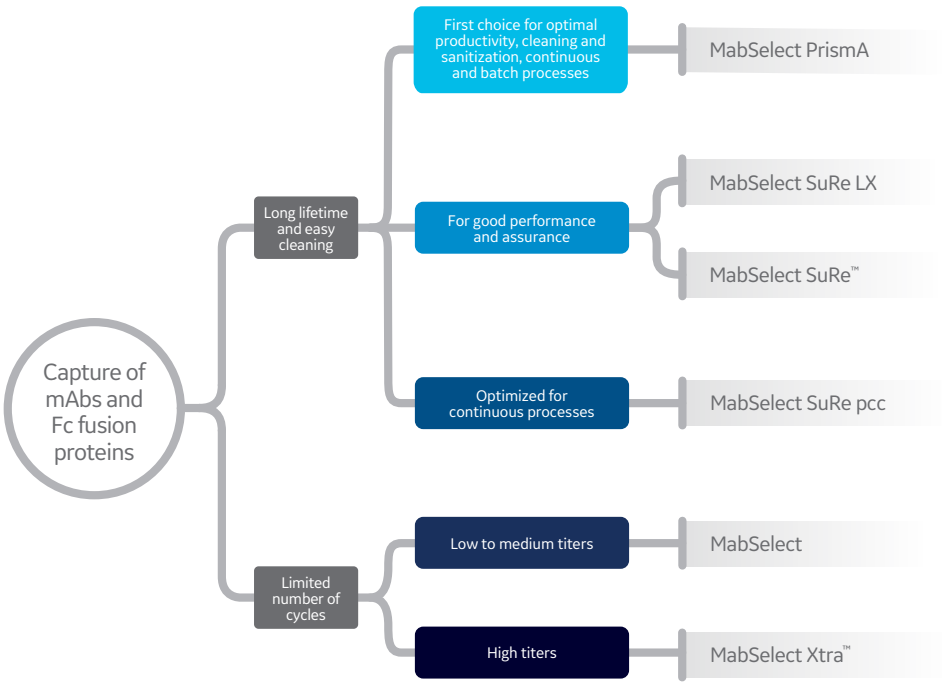
The polishing steps following the capture step can be performed in either bind-elute (binding) or flow-through (nonbinding) mode. Our Capto™ adhere and Capto adhere ImpRes resins can both be used for polishing in a two-step process in flow-through mode. They are both highly efficient in removing remaining impurities like aggregates, host cell protein (HCP), DNA, and viruses.

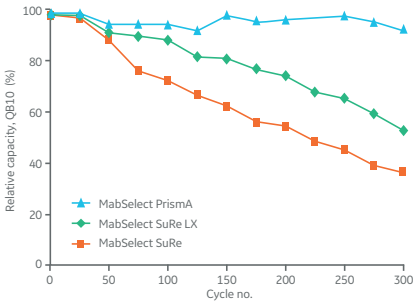
Polishing mAbs in a three-step process

A three-step purification process, with two polishing steps based on one cation exchanger and one anion exchanger, is a classical way of purifying mAbs.

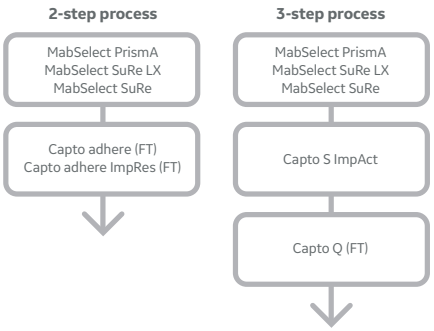
Cation exchangers are used for the removal of HCP, protein A, aggregates, and fragments. The cation exchange step is commonly followed by an anion exchange step (run in flow-through mode) for removal of remaining impurities such as DNA.

Chromatography resins and membranes suitable for polishing in a three-step purification process are Capto S ImpAct and Capto Q.





Relative remaining capacity (Q_{B10}) for 300 cycles, including CIP with 0.5 M NaOH for 15 min/cycle.



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Featured products

MabSelect PrismA

- Enhanced dynamic binding capacity allows high mass throughput of processed mAb per resin volume unit.
- Excellent alkaline stability enables efficient cleaning and sanitization using 0.5–1.0 M NaOH for improved process economy, bioburden control, and robustness.

Capto adhere and Capto adhere ImpRes

- Wide operational window of pH and conductivity.
- Two-step chromatographic process with protein A affinity, saves time and cuts operating costs.

Capto S ImpAct

- High binding capacity, > 100 mg mAb/mL resin.
- Efficient aggregate removal at high load of monoclonal antibodies.
- High-resolution polishing based on the well-established Capto platform.

Capto Q

- High dynamic binding capacity at high flow raises productivity.
- High-volume throughput cuts process times.
- Cost-effective processing with smaller unit operations.

Sample preparation with the Protein Prep syringe filter for ÄKTA systems

NEW

Protein Prep syringe filters are ready-to-use with polycarbonate housing and a regenerated cellulose membrane that is low protein binding and broadly compatible with common solvents. Syringe filtration has been shown to reduce debris residue in the column that could otherwise affect performance and column life. In addition, the Protein Prep syringe filter is lot certified for low levels of extractable particles that might otherwise interfere with chromatograms.

Protein Prep syringe filter for ÄKTA systems

- 13 mm or 30 mm diameter.
- 0.2 µm or 0.45 µm pore size.

Tips for choosing the right filter

- Use 13 mm diameter filter for sample volumes < 10 mL.
- Use 0.2 µm pore size filter if the particle size of the chromatography resin is < 30 µm.

*Request your sample **here**.*

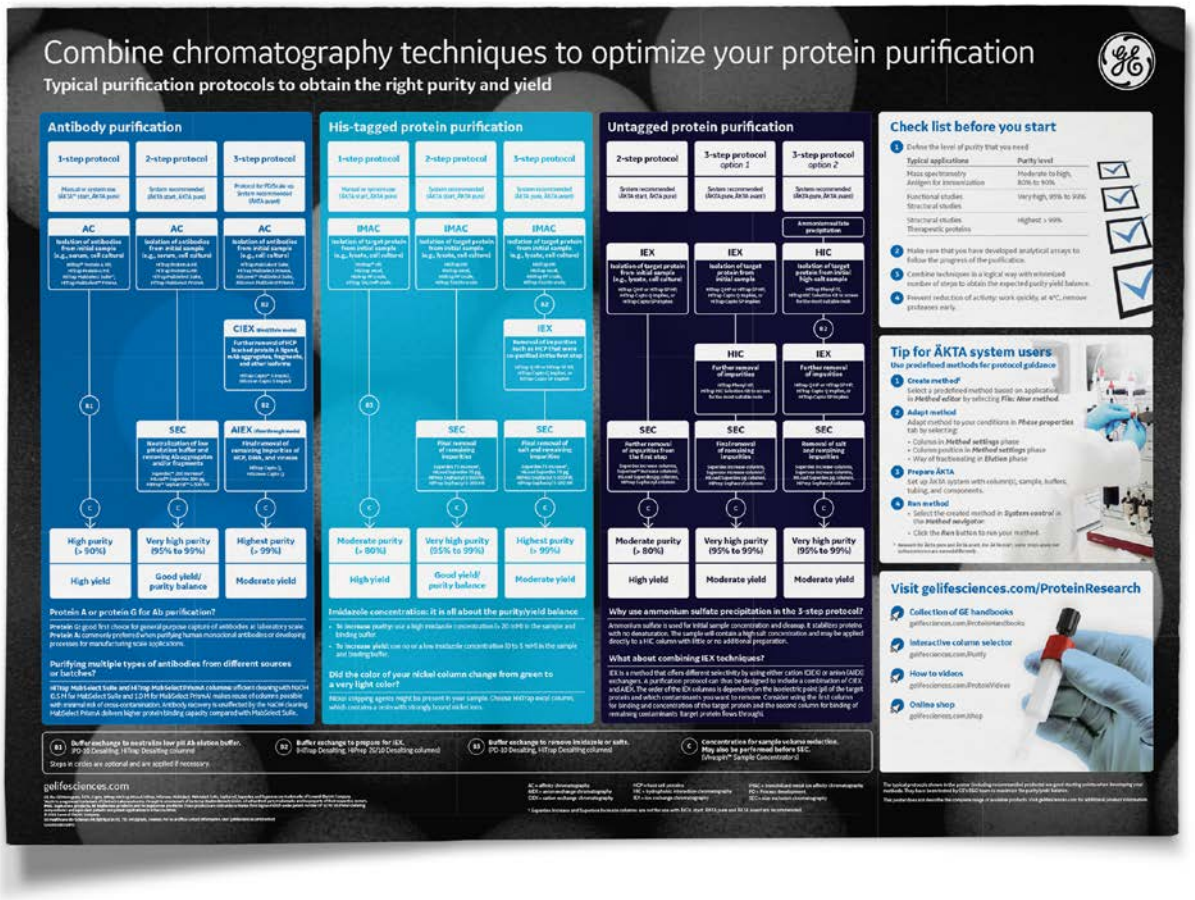
Protein Prep syringe filter for ÄKTA systems

*For more information please visit **eu.fishersci.com***

Free wall poster!

So how do you best combine chromatography techniques to obtain the right purity and yield of your protein? Whether you want to purify a tagged, antibody or native protein, our free wall poster helps you effectively combine the main chromatography techniques to obtain a powerful purification protocol.

*Request your poster **here!***



Try our new Benchkote sheets for ÄKTA avant, pure or start and protect the top buffer tray from buffer spillages and salt deposits.

Click **here** for more information.

Ordering information

Resin	Format	Description	Volume	Pack size	Item	Price* (£)
MabSelect PrismA	Pre-packed columns	HiTrap™ MabSelect PrismA	1 mL/column	5 columns	17549852	3369.00
MabSelect PrismA	Pre-packed columns	HiTrap MabSelect PrismA	5 mL/column	5 columns	17549854	876.00
MabSelect PrismA	Pre-packed columns	HiScreen™ MabSelect PrismA	4.7 mL/column	1 column	17549815	834.00
RC membrane	Non sterile	Protein Prep syringe filter for ÄKTA systems	13 mm 0.45 µm	150/pk	10463113	300.00
RC membrane	Non sterile	Protein Prep syringe filter for ÄKTA systems	13 mm 0.2 µm	150/pk	10463103	300.00
RC membrane	Non sterile	Protein Prep syringe filter for ÄKTA systems	30 mm 0.45 µm	150/pk	10463033	330.00
RC membrane	Non sterile	Protein Prep syringe filter for ÄKTA systems	30 mm 0.2 µm	150/pk	10463043	330.00

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Stripping and reprobing Western blot membrane: problems and solutions

Multiple uses of your blotting membrane can be especially useful if your proteins of interest are only available in limited quantities.

Stripping the membrane involves harsh conditions to disrupt the interaction between the membrane-bound protein and the primary antibody. This process enables reprobing with a new primary antibody for further protein identification. Careful consideration of the stripping conditions can help minimize the risk of protein loss from the membrane. These considerations include using combinations of detergents, reducing agents, heat, and high or low pH.

There are a few things to bear in mind once you know you are going to reuse a membrane. Your target protein abundance and antibody affinities are two points to consider. These properties influence your membrane stripping effectiveness, and which antibody you use first.

Strategy 1 – Problem: you have two proteins of similar abundance and two antibodies of similar affinity. Solution: you can detect either protein first, strip the membrane, and then detect the remaining protein.

Strategy 2 – Problem: you have two proteins of similar abundance and two antibodies of unequal affinity. Solution: detect the protein with the lowest affinity antibody first, strip the membrane, and then detect the protein with the highest affinity antibody.

Strategy 3 – Problem: you have two proteins of different abundances (one high and one low) and antibodies of equal affinity. Solution: detect the low-abundance protein first, strip the membrane, and then detect the high-abundance protein.

Strategy 4 – Problem: you have two proteins of different abundances (one high and one low) and antibodies of unequal affinity. Solution: detect the low-abundance protein first, strip the membrane, and then detect the high-abundance protein.

When using enhanced chemiluminescence (ECL) detection for a Western blot, a sequential labeling method is available for quick detection of a second protein on a single membrane.

Alternative methods of detecting additional proteins

- Sequential labeling with ECL detection
- Multiplex detection

Labeling and detection of the first protein is performed as normal using ECL. The horseradish peroxidase (HRP) is then inactivated (quenched) using hydrogen peroxide (H_2O_2) and the membrane is washed. As a result, the second protein can be labeled with a different antibody for detection without any interference.

Multiplex detection - To avoid stripping and reprobing altogether, multifluorescence (multiplex) detection can be used to detect multiple proteins on the same membrane. In this technique, secondary antibodies labeled with fluorophores enable simultaneous detection of more than one protein.

*Download handbook **here**.*

*For more information please visit **eu.fishersci.com***

Featured products

Amersham™ ECL™ detection reagents

ECL based on horseradish peroxidase (HRP)-conjugated secondary antibodies has become the most commonly used detection method for Western blotting. It is a sensitive detection method, where the light emission is proportional to protein quantity.

Minute quantities of proteins can be detected and quantitated.

- Longer shelf life: up to 18 month shelf life on ECL Select™ and Prime products.
- Stability: ECL Select and ECL Prime products are stable and stored at room temperature.

Amersham Hyperfilm™ ECL detection film

This is a sensitive film for the detection of chemiluminescent signals in Western blotting assays.

- Clear background for excellent contrast and band visibility.
- Publication-quality images.
- Learn more here: gelifesciences.com/wbfaq.

Amersham Western blotting membranes

GE Healthcare Life Sciences offers a broad selection of nitrocellulose (NC) and polyvinylidene difluoride (PVDF) Western blotting membranes, with pore size ranges to suit your application requirements.

- Optimized for chemiluminescent and fluorescent detection.
- Excellent protein binding capacity over a wide size range.
- New larger pack sizes reduce your price per blot by up to 30%.

CyDye™ labeling reagents

CyDye Fluors are fluorescent dyes used in applications such as microarray analysis, FISH, 2-D DIGE, immunoprecipitation, and blotting.

Dyes are packaged in premixed amounts and foil-sealed to ensure consistent labelings.

Amersham ECL Rainbow™ molecular weight markers:

Accurate size determination of your protein on gels and blots. Download a brochure **here**.

Ordering information

Chemistry	Format	Description	Volume/size	Pack size	Item	Price* (£)
Chemiluminescent	Kit	ECL Western blotting detection reagent	For 2000 cm² membrane	1/pk	RPN2209	190.35
Chemiluminescent	Kit	ECL Select Western blotting detection reagent	For 1000 cm² membrane	1/pk	RPN2235	245.76
Chemiluminescent	Kit	ECL Prime Western blotting detection reagent	For 3000 cm² membrane	1/pk	RPN2236	415.23
Chemiluminescent	Kit	QuickStain kit	1 µg/mL to 20 mg/mL	1/pk	RPN4000	97.71
Chemiluminescent	Kit	Full range Rainbow molecular weight marker	250 µL	1/pk	RPN800E	177.07
Chemiluminescent	Sheets	Amersham Hyperfilm ECL	5 × 7 inches	50/pk	28906835	145.24
Chemiluminescent	Roll	Amersham Hybond™ PVDF membrane	0.2 µm, 260 mm × 4 m	1 roll	10600021	273.06
Chemiluminescent	Roll	Amershan Protran™ supported NC membrane	0.45 µm, 300 mm × 4 m	1 roll	10600016	254.60
Fluorescent labeling	Kit	Amersham CyDye Value Packs - Cy™5 Mono - NHS Ester	10 mg	1/pk	PA15104	1504.80
Fluorescent labeling	Kit	Amersham CyDye Value Packs - Cy7 Mono - NHS Ester	10 mg	1/pk	PA17104	1160.80

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* GE recommended list prices.

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