## **BioXtra**<sup>™</sup> News

# GE Life Sciences and VWR launch a new chapter in collaboration

The best from top brands come together to bring you the new protein workflow

# Add more security to your ÄKTA™ chromatography system runs by applying sample filtration

Beep ... Beep ... Beep – We all hate this sound from systems indicating that something is going wrong. Overpressure in automated

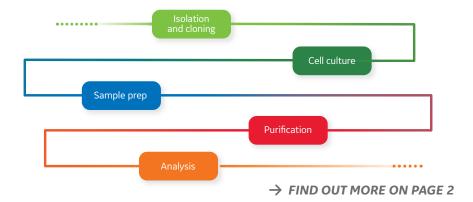


Protein Prep syringe filter for ÄKTA systems

chromatography runs during sample application is one of the most common problems. With the new Protein Prepsyringe filters, a ready-to-use low-protein binding regenerated cellulose membrane fitted into a robust polycarbonate housing and broadly compatible with common solvents, this issue could significantly improve. Syringe filtration has been shown to reduce debris residue in the column that could otherwise impact 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.

→ MORE ON PAGE 13

## Do we need workflows in science?



## Top stories

### **Think Pink**

GE Whatman<sup>™</sup> to raise money for Think Pink breast cancer charity campaign in October (page 19).

## **GE Isolation and Cloning**

Fundamentals of NGS sample preparation (page 3).

## **GE Cell Culture**

"HyCell™ Transfx" media supports transient transfection, growth, and expression of a variety of recombinant proteins (page 6).

## **GE Sample Prep**

Save time in HPLC prep (page 7).

### **GE Protein Purification**

Goodbye bed height, hello bed volume (page 12).

## **GE Analysis**

DIBE technology for reliable HCP detection (page 17).

## Key products for your workflow

Full list of products for your workflow (page 10–11).

## New products

### MabSelect™ PrismA

significantly enhanced alkaline stability and capacity (page 15).





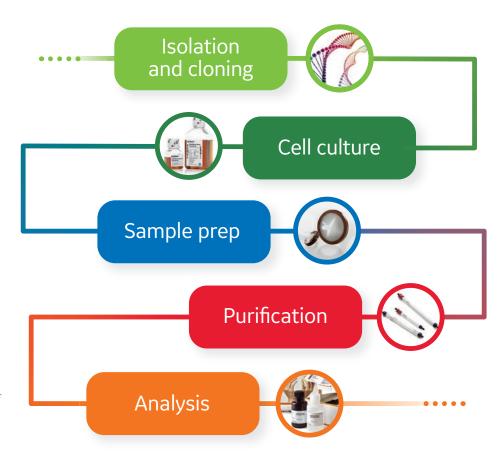
## Do we need workflows in science?

Wikipedia defines the word workflow as: "Consists of an orchestrated and repeatable pattern of business activity enabled by the systematic organization of resources into processes that transform materials, provide services, or process information.[1] It can be depicted as a sequence of operations, the work of a person or group, [2] the work of an organization of staff, or one or more simple or complex mechanisms."

There are many keywords in this definition but for us it means delivering products, training and support that enable you to take the next step in your research more easily.

This newspaper edition features stories from the different steps in the protein purification workflow; lists useful products and provides information about recently launched products and support material. The simple color-coding enables simple navigation and easy identification of the principle steps in the process. Enjoy the stories.

[1] webmaster@ftb.ca.gov (27 October 2009). "Business Process Management Center of Excellence Glossary" (PDF). Retrieved 31 March 2015. [2] See e.g., ISO 12052:2006, ISO.org



## Products to support your research

## Isolation and cloning

## illustra™

Nucleic acid sample prep and storage

## Key products:

- ExoProStar™ S
- FTA™ cards
- GenomiPhi™
- Nucleon
- PuReTag and Hot Start RTG PCR beads
- SC GenomiPhi
- TempliPhi™

## Cell culture



## HyClone™

Cell culture media and sera

## **Key products:**

- DMEM
- RPMI
- FBS
- Reagents and buffers
- Process water

#### Products for cell separation and isolation:

- Percoll™
- Ficoll™

## Sample prep



## Whatman™

Laboratory filtration products

## **Key products:**

- Puradisc syringe filters
- SPARTAN™ syringe filters
- Whatman GD/X™ syringe filters
- Mini-UniPrep<sup>™</sup> filter vials
- 934-AH™ RTU
- GF/C RTU
- Polycap SPF
- RC membrane
- Vacu-Guard Benchkote<sup>™</sup>
- Custom designed filtration

### Purification



Chromatography columns, resins and systems

### **Kev products:**

- HiTrap™ columns
- HisTrap™ columns
- PD-10 desalting columns
- HiLoad™ columns
- Superdex™ Increase GL columns
- Ni Sepharose™ resin
- MabSelect PrismA™ resin
- Protein G Sepharose resin
- Capto™ Q resin
- Capto S ImpAct resin
- Capto ImpRes resin

## **Analysis**



## Amersham"

Systems, membranes, films and reagents

### Kev products:

- NC and PVDF membranes
- Amersham ECL™ gels
- CyDye™ labelling kits
- ECL™ detection reagents
- Rainbow™ markers
- Hvperfilm™
- Amersham QuickStain
- · PlusOne reagents
- Electrophoresis and transfer units

## Zika's genetic code

To date, the Zika 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.

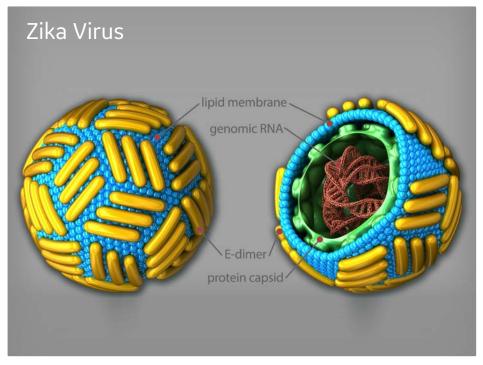
The most high-tech methods focus on targeting Zika's genetic materials. Scientists in the field collecting mosquito samples use a special fiber paper manufactured by GE in Cardiff. This coffee-filter-like material, called Whatman FTA paper, has magical properties. It is chemically treated to break down 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 polymerase chain reaction (PCR\*), which rapidly produces multiple copies of the virus's RNA so that medical workers can identify it in a patient's blood.

### → READ MORE HERE







Zika is catching a lift on Aedes aegypti mosquitos.



FTA cards

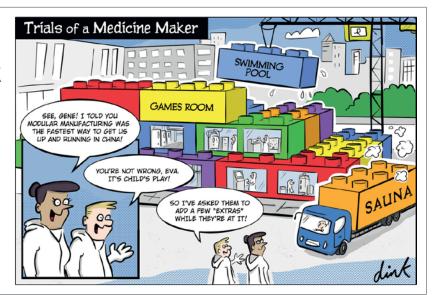
An illustration of the Zika virus' structure. Image credit: Getty Images

## Prefabricated facilities: cut time and capital risk

What would you add to your biomanufacturing facility if you had the chance? Gene's sauna might be out of the question. But going for a modular prefabricated facility can add more benefits than you might imagine.

→ READ THE FULL ARTICLE HERE





## Garbage in - garbage out

Sample prep can have a big impact on nextgeneration sequencing (NGS) outcomes. Here are some simple things you can do to get your NGS off to a good start.

DNA for sequencing might come from a variety of sources, including fresh tissue, formalinfixed paraffin-embedded (FFPE) tissue, cultured cells, and liquid biopsies. Each source comes with its own challenges for maximizing the three key aspects: quantity, integrity and purity.

### Challenge 1: Yield

If your sample is insufficient, what can we learn from those studying at the single-cell level? Commercially available whole genome amplification (WGA) kits provide the opportunity to expand your starting material from nanograms to micrograms in a matter of hours. This technique provides improved coverage compared to PCR-based amplification and is associated with fewer amplification errors.

### **Challenge 2: Integrity**

Having enough DNA won't make for accurate sequencing if your DNA is degraded. Degradation can affect all kinds of samples, but long-term storage and exposure to fixatives, as you might find in FFPE samples, can exacerbate the damage. If you can't acquire better samples, DNA repair might improve your outcomes. Several commercial kits can, for example, modify blocked 3' ends or fix DNA nicks. These simple repairs help make more fragments suitable for sequencing.

#### **Challenge 3: Impurities**

Producing reliable results in sequencing requires samples free of proteins, organic solvents and surfactants. You might also have tissue-specific contaminants to consider. Researchers often measure DNA purity by looking at the 260:280 nm absorbance ratio (ratios between 1.8 and 2.0 are preferred). As a secondary check, measure the 260:230 ratio, which will detect the presence of commonly used solvents and surfactants, such as phenol and EDTA. Values between 2.0 and 2.2 indicate high purity.

Find out more about our NGS products or contact us for more information.





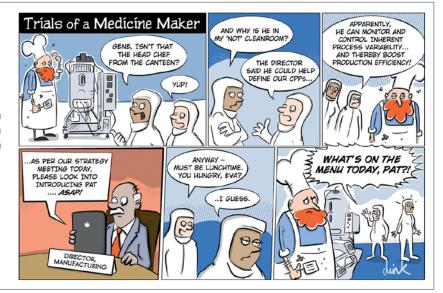


## The secret to PAT's (kitchen) success

Comparing kitchens to cleanrooms might seem strange. But, as medicine makers Gene and Eva discovered, process analytical technologies (PAT) can bring success in both places. Here's why.

→ READ THE FULL ARTICLE HERE





## Fundamentals of NGS sample preparation

Next-generation sequencing (NGS) has enabled us to extract genetic information from samples faster, more reliably, and at lower cost than ever before.

The first step in every sample prep protocol is **extracting the genetic material** – DNA or RNA - from cells and tissues. Other molecules, such as RNA and proteins, interfere with the sequencing process and must be removed before doing anything else.

The traditional gold standard in DNA isolation is phenol-based extraction. Phenol is a hydrophobic solvent that denatures and dissolves proteins, removing them from the DNA-containing aqueous phase. Spin columns that specifically bind DNA provide an alternative and are an easy-to-use, but more expensive, method to wash away the debris. Chloroform-based extraction, another alternative, enables you to isolate high-quality DNA without phenol, and commercial kits can include a resin that minimizes the risk of contamination.

Amplification after extraction is optional, depending on your application and sample size. Isothermal amplification and polymerase chain reaction (PCR) are two common methods to increase the amount of input DNA. PCR uses generic primers to amplify the starting material in a highly uniform manner, but tends to be more

error-prone than multiple displacement amplification (MDA).

Most NGS platforms analyze DNA in uniform, bite-size pieces, created by DNA fragmentation. This process generates a 'library' of fragments with a narrow length distribution that is optimal for the sequencing platform. The **library preparation steps** can include, fragmentation (mechanical or enzymatic) and DNA end repair (creating blunt ends) followed by adaptor ligation.

To speed up your workflow, it might be necessary to 'clean up' your library before sequencing by **removing fragments that won't produce relevant data**. For NGS workflows that have narrow size requirements, discarding fragments that are either too large or too small to produce useful results can improve sequencing efficiency.

A final step before proceeding to sequencing is to confirm the quality and quantity of your DNA. Both parameters contribute to the confidence in your sequencing data. You can measure the quantity of your DNA using fluorescence- or qPCR-based methods.

These are the basic steps that researchers use to prepare DNA for sequencing. You can find more information about specific NGS workflows and applications in our other NGS blogs.







## VWR® collection thermal cycler family; innovative technologies for PCR

The VWR thermal cycler family combines high quality engineering with a comprehensive range of block formats. The UNO cycler is designed around a powerful yet easy to use software interface and uses the same platform for both 96-well and 384-well formats. The Doppio, with two independent high speed 48-well blocks in one system, offers the optimal solution for maximum flexibility within minimal footprint. The Ristretto is a compact personal cycler with the highest flexibility, having a universal block that can be loaded with up to either  $32 \times 0.2$  mL tubes or  $16 \times 0.5$  mL tubes with flat caps. Because of the special design of the heated-lid, the height adjusts automatically to the different tube sizes.







# GEHC HyClone "HyCell TransFx" media supports transient transfection, growth, and expression of a variety of recombinant proteins

Hyclone HyCell TransFx-H and TransFx-C media are optimized for human embryonic kidney (HEK) 293 cells and chinese hamster ovary (CHO) cells respectively, to reduce testing and validation times from research to scale-up. These regulatory-friendly media provide an optimized, nutrient-rich formulaton, minimizing the requirements for further supplementation and the need to switch medium during the process.

HyClone HyCell TransFx-C is an animal-derived component-free (ADCF), hydrolysate-free, and regulatory-friendly cell culture medium. The versatility of this medium allows quick adaptation and supports exceptional transfection, high viable cell density, and high



productivity across a variety of chinese hamster ovary (CHO) cell lines. HyCell TransFx-C is available in liquid and powder formats in user-friendly packaging.

HyClone HyCell TransFx-H is an animal-derived component-free (ADCF), hydrolysate-free, and regulatory-friendly cell culture medium. The versatility of this medium allows quick adaptation and supports exceptional transfection, high viable cell density, and high productivity across a variety of human embryonic kidney (HEK) 293 cell lines. HyCell TransFx-H is available in liquid and powder formats in user-friendly packaging.





## VWR collection: all you need for cell culture

Choosing the right type of plastic is the key to consistency in cell culture research. VWR Collection offers researchers a range of quality plastics in non-treated or treated sterile formats; cell culture flasks, cell culture dishes, multi-well cell culture plates, 3-D plates with scaffolds and more.

Increased cell attachment surface: This highly hydrophilic surface offers a significant advantage over the traditional cell culture surface. The surface treatment can improve cell spreading and attachment and is suitable for cells that may adhere poorly due to cell phenotype, stressful culture conditions, or those which normally require additional biological coatings for attachment.

Certifications: ISO 13485:2003 and ISO 9001:2008 certified.







## Test your knowledge!

Try our word finder puzzle on P17 and the crossword on P19.

- → GO TO PAGE 17 FOR WORD FINDER PUZZLE
- → GO TO PAGE 19 FOR CROSSWORD

## Improve lab efficiency through better filtration

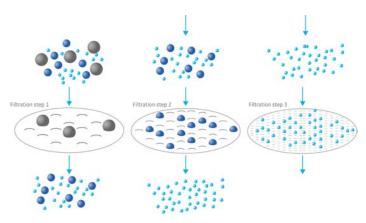
Do you consider particle retention, loading capacity, and liquid flow rate when choosing a filter or device?

Perhaps there is a better filter out there for your application. Or perhaps your analysis might be easier, quicker, or produce results that are more consistent if you switched your filter to a different grade.

Here are three key characteristics to consider when identifying the right filter:

- **1 Particle retention** For cellulose and glass microfiber papers, it is expressed as a "nominal retention rating", and quoted at 98% efficiency to allow for secondary filtration effects. For membrane filters with defined pore sizes, it is an absolute retention rating.
- **2 Loading capacity** Filters with the highest loading capacities are chemically treated and are more expensive than their untreated counterparts. Treatment might also interfere with analysis. This can happen either through chemical interaction with the sample or by increasing the time to results due to a slower flow rate than that of an untreated filter. By knowing the weight of filtrate that you want to retain on the filter, you can choose a filter that will safely accommodate your needs without the downsides of a filter that is more complex than is needed.
- **3 Liquid flow rate** The flow rate describes the speed at which a liquid flows through the filter. In practice, this is dependent on several factors that will often be specific to the solid/liquid being filtered. But, for comparison purposes, a typical water flow rate is measured and provided for each grade under gravity and normalized to a certain diameter.

Try our Whatman Filter Selector App to find out if you are using the most appropriate filtration solution for your samples.







## **LOOK-UP - APPS**



## Whatman Filter Selector App

Using the right filter for your application can save you time and simplify your process. Take the guess work out of lab filter selection. Answer a few simple questions to find the optimal Whatman filter.

→ DOWNLOAD APP HERE



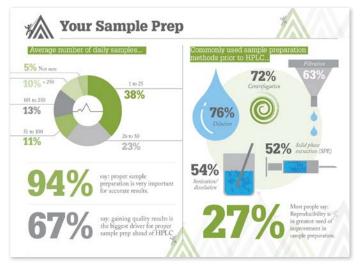
## Sample preparation for HPLC – what you said!

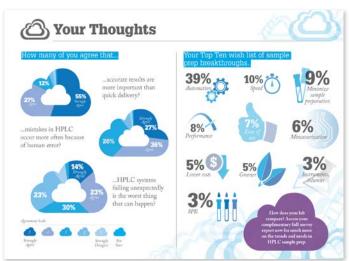
Readers from "The Analytical Scientist" volunteered in an exclusive survey from GE Healthcare and shared valuable insights about goals and their thoughts regarding sample preparation. The data is presented in an easy to understand infographic. Check the picture and see what challenges other users are facing.

## → DOWNLOAD SURVEY RESULTS HERE









## Save time in HPLC prep

Sample filtration protects your HPLC instrument and column while preserving data quality.

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.

#### 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 autosampler. 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 media, starting with larger pore sizes and going down to the desired pore size.

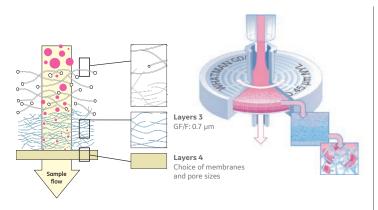
This approach traps large particles first and successively traps smaller particles (see picture). The device does not get clogged as easily as devices with a single membrane, making filtration faster and easier.

#### Go syringeless

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

In a syringeless filter, the filter membrane, pre-filtration chamber, post- filtration storage vial, and cap are all part of one device (see picture). 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 of your sample.





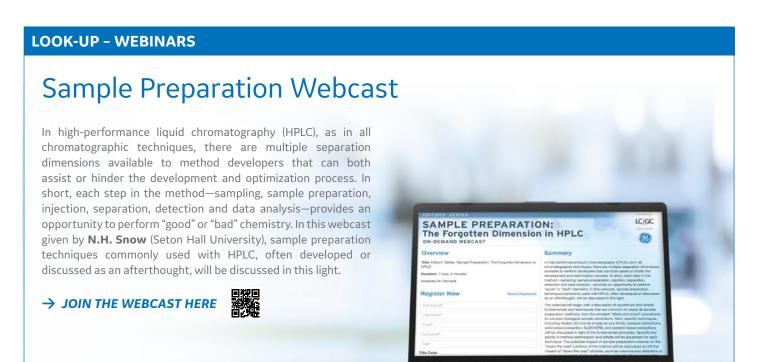
Mini-UniPrep syringeless filters

Septum

Whatman GD/X stacked syringe filter

→ READ THE FULL ARTICLE HERE





## **LOOK-UP - VIDEOs**

## Whatman: 250 years of innovation

For over 250 years GE Whatman brand has provided customers with reliable, quality products for a variety of lab filtration and sample preparation needs.







## Protect your columns and your peaks

Good practice in liquid chromatography (LC) is to filter samples prior to injection onto the column. Sample filtration prevents unwanted particulates from entering the column. This is important because particulates can reduce column lifetime, increase run time, and distort peak shape. In addition to affecting the quality of your results, particulates might clog the column inlet, causing increased back pressure and premature ending of chromatography runs. Adjusting protocols to extend usable lifetime can minimize the frequency of column replacement while maintaining data quality throughout.

Although filtration prior to chromatography is common practice, the factors that impact ideal filter selection often are not considered. The 'right' filter depends on the method that the sample is being prepared for, the chemical properties of the solvent being used, and the physical and chemical properties of the sample itself.

The new Protein Prep syringe filter for protein work has been tested for: 1) the impact of filtration on **column cleanliness**, 2) the **recovery** using regenerated cellulose as a preparation medium and 3) the **filter quality** to the fidelity of results.

Regenerated cellulose from GE Healthcare Life Sciences is batch tested to ensure low levels of extractable compounds that might otherwise interfere with analyses.

Percent recovery of BSA samples filtered with regenerated cellulose membranes

Pore size	Recovery at 1 mg/mL (%)	SD	Recovery at 0.5 mg/mL (%)	SD
0.2 µm	98	0.6	97	0.3
0.45 µm	99	0.7	99	0.9

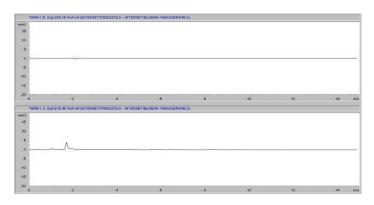
SD = Standard deviation, N = 3

Hold-up volumes of water in filtration devices containing regenerated cellulose membranes. Results before and after purge

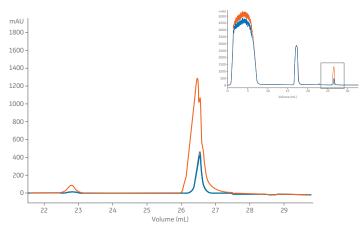
	Hold-up volume (µmL)		Final hold up volume (µmL)		
Pore size	Average	SD	Average	SD	
0.2 μm	135	36	9.3	0.9	
0.45 μm	135	31	9.2	1.1	

SD = Standard deviation, N = 10

Chromatograms of water filtered through 30 mm 0.2  $\mu$ m regenerated cellulose membrane syringe filters and subjected to HPLC analysis. Top panel shows results at 254 nm; bottom panel shows results at 215 nm.



The clean-in-place (CIP) peak after purification of the unfiltered (orange) and filtered (green and blue) mAb samples.



## BioXtra – key produc

## Isolation and cloning

Description	Product code
illustra PuReTaq RTG PCR, 0.2 mL tube, hinged cap, 96 reactions	27955901
illustra PuReTag RTG PCR, 0.5 mL tube, 100 reactions	27955801
illustra PuReTag RTG PCR 0.2 mL plate, 96 reactions	27955701
illustra ExoProStar 1-Step (100 reactions), PCR clean up technology	US77702
illustra ExoProStar 1-Step (500 reactions), PCR clean up technology	US77705
illustra ExoProStar 1-Step (2000 reactions), PCR clean up technology	US77720
illustra ExoProStar S (20 reactions), PCR clean up technology	US79002
illustra ExoProStar S (100 reactions), PCR clean up technology	US79010
illustra ExoProStar S (500 reactions), PCR clean up technology	US79050
illustra GFX™ PCR DNA and gel band purification kit, 100 purifications	28903470
illustra GFX PCR DNA and gel band purification kit, 250 purifications	28903471
DNTP set, 20 mM solutions (A, C, G, T), 4 × 10 µMOL	28406558
DNTP set, 100 mM solutions (A, C, G, T), 4 × 25 µMOL	28406551
illustra NAP-10 columns with Sephadex™ G-25 50 ST	17085402
illustra NAP-5 columns, 50 ST	17085302
illustra NAP-25 columns, 50 ST	17085202
Sera-Mag Oligo(dt)	38152103011150
SpeedBead Blocked Strep	21152104011150
Sera-Mag Streptavidin	30152105010150
SpeedBead Streptavidin	66152104011150
SpeedBead Neutravidin	78152104010150
FTA classic card 100/pk	28421884
FTA micro card 100/pk	28421887
FTA mini card 100/pk	28421870
FTA PlantSaver card 100/pk	28421873
Multi-barrier pouches, 11 x 16.5 cm 100/pk	28421854
Multi-barrier pouches, 9.5 x 7.5 cm 100/pk	28421855
Desiccant 1 g 1000/pk	28421835
Uni-Core punch 1 mm 25/pk	29002515
Cutting mat 6.0 x 8.0 in 1/pk	28421844
903 protein saver card 100/pk	28416563
Plastic ziploc bag 100/pk	28417398

## Cell culture

Description	Product code
LM-DPBS/MOD, 1000 mL	SH30028.03
LM-MOL biol grade water, nuclease free, 100 mL	SH30538.01
LM-cell culture grade water, endotoxin free, 1000 mL	SH30529.03
LM-DPBS/MOD,500 mL	SH30028.02
LM-PBS .0067M PO4, 500 mL	SH30256.01
LM-EBSS,500 mL	SH30029.02
LM-RPMI 1640, 500 mL	SH30027.01
LM-RPMI W/L-Glut W/HEPES, 500 mL	SH30255.01
LM-RPMI 1640 W/O L-Glut, 500 mL	SH30096.01
LM-DMEM/high,500 mL	SH30022.01
LM-DMEM/low, 500 mL	SH30021.01
LM-DMEM W/O L-Glut W/PYR, 500 mL	SH30285.01
LM-DMEM/F-12 MOD, 500 mL	SH30261.01
LM-MEM/EBSS, 1000 mL	SH30024.02
LM-MEM/EBSS W/O L-Glut, 1000 mL	SH30244.02
HyCell TransFx-C liquid media 1000 mL	SH30941.02
HyCell TransFx-H liquid media 1000 mL	SH30939.02
Research grade fetal bovine serum, South American origin	SV30160.03
HyCryo 2x cryopreservation media	SR30001.02
LM-Trypsin, 100 mL	SH30042.01
LM-Trypan blue 0.4% solution, 100 mL	SV30084.01
LM-L-Glutamine, 500 mL	SH30034.02
LM- Penicillin/Streptomycin solution, 100 mL	SV30010
HyClone 10% antifoam solution, ADCF, irradiated (Q7-2587 simethicone), 500 mL	SH30897.01

## Sample prep

Description	Product code
Protein Prep for ÄKTA systems 13/0.45 RC 150/pk	10463113
Protein Prep for ÄKTA systems 13/0.2 RC 150/pk	10463103
Protein Prep for ÄKTA systems 30/0.45 RC 150/pk	10463033
Protein Prep for ÄKTA systems 30/0.2 RC 150/pk	10463043
Vivaspin 2 MWCO 10 000	28932247
Vivaspin 6 MWCO 10 000	28932296
Vivaspin 2 MWCO 30 000	28932248
Vivaspin 6 MWCO 30 000	28932317
Mini-UniPrep syringeless filter 0.2 μm RC 100/pk	28421672
Mini-UniPrep syringeless filter 0.45 μm RC 100/pk	28421680
Mini-UniPrep syringeless filter 0.45 μm GMF 100/pk	28421675
Mini-UniPrep amber syringeless filter 0.2 μm PES 100/pk	28421658
Mini-UniPrep amber syringeless filter 0.45 μm PES 100/pk	28421666
Whatman GD/X syringe filter 25/0.2 µm PES sterile 150/pk	28420521
Whatman GD/X syringe filter 25/0.45 µm PES sterile 150/pk	28420522

## Purification

Description	Product code
ÄKTA start	29022094
Frac 30	29023051
UNICORN™ Start 1.1 DVD pk + activation code	29276964
HiTrap MabSelect PrismA 5 × 5 mL	10463113
HiTrap MabSelect PrismA 5 × 1 mL	10463103
HiTrap MabSelect SuRe™ 5 × 5 mL	11003495
HiTrap MabSelect SuRe 5 × 1 mL	11003493
HiTrap Protein A HP 5 × 5 mL	17040303
HiTrap Protein A HP 5 × 1 mL	17040201
HiTrap Protein G HP 5 × 5 mL	17040503
HiTrap Protein G HP 5 × 1 mL	17040401
HiTrap Capto Q ImpRes 5 × 5 mL	17547055
HiTrap Capto Q ImpRes 5 × 1 mL	17547051
HiTrap Capto SP ImpRes 5 × 5 mL	17546855
HiTrap Capto SP ImpRes 5 × 1 mL	17546851
Superdex 200 Increase 10/300 GL	28990944
HiLoad 16/600 Superdex 200 pg	28989335
HiPrep™ 16/60 Sephacryl™ S-300 HR	17116701
HiTrap Desalting, 5 × 5 mL	17140801
HiPrep 26/10 Desalting	17508701
Protein A Sepharose CL-4B	17078001
Protein G Separose™ 4 Fast Flow	17061801

## ts for your workflow

## Analysis

Description	Product code
ECL Select™ Western blotting detection reagent	29013864
ECL Prime Western blotting reagent	28980926
ECL Western blotting reagents	25006327
Amersham Hybond™ LFP 0.2 μm PVDF 254 mm × 4 m, 1 roll/pk	10600022
Amersham Protran™ premium 0.45 µm NC 300 mm × 4 m, 1 roll/pk	10600003
Amersham ECL prime blocking reagent	RPN418
Hyperfilm ECL 5 x 7 in, 50 shts	28906835
Hyperfilm MP 5 x 7 in, 50 shts	28906842
Full range molecular weight markers Rainbow	67610165
ECL DualVue™ Western markers	25190061
2-D Quant kit	80648356
2-D Clean-up kit	80648451
Amersham CyDye DIGE fluor Cy™3 minimal dye, 10 nmol	25800861
Amersham CyDye DIGE fluor Cy5 minimal dye, 10 nmol	25800862
IPG buffer pH 3–11 NL	17600440
DeStreak rehydration solution	17600319
Immobiline™ DryStrip pH 3-11 NL, 7 cm	17600373
Immobiline DryStrip pH 3-11 NL, 13 cm	17600375
ECL Plex™ Goat-Anti-Rabbit IGG Cy3, 150 μg	28901106
ECL Plex Goat-Anti-Rabbit IGG Cy5, 150 μg	PA45011
Grade 3MM Chr blotting paper, sheet, 15 × 17.5 cm	3030-153
Grade 3MM Chr blotting paper, sheet, 18 × 34 cm	3030-221
Grade 3MM Chr blotting paper, sheet, 35 × 43 cm	3030-347
Grade 3MM Chr blotting paper, sheet, $12 \times 14$ cm	3030-6132
Grade 3MM Chr blotting paper, sheet, 15 × 20 cm	3030-6188
Grade 3MM Chr blotting paper, sheet, 26 × 41 cm	3030-6461

## Lab essentials

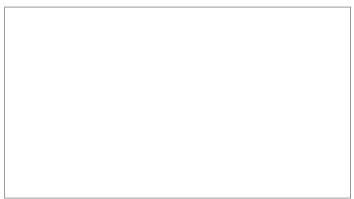
Description	Product code
Benchkote sheet 46 × 47 cm 50/pk	2300-594
Vacu-Guard 60 mm 0.45 μm PTFE 10/pk	6722-5001
Grade 105 lens cleaning tissue 10 x 15 cm 25/pk	2105-841



Description	Product code
Mastermix 2,0X red TAQ DNA polymer 1.5 mm	VWRC733-2546
DNA polymerase tempase hotstart 5U/μL	VWRC733-1333
DNA polymerase TAQ 5U/µL	VWRC733-1303
PCR grade water (6 × 5 mL)	VWRC733-2573
UNO96G gradient thermocycler**	VWRI732-2549
DOPPIO gradient 2 × 48 well thermocycler**	VWRI732-2552
RISTRETTO 32 well personal thermocycler**	VWRI732-2553
peqSTAR 96X universal gradient**	PEQL95-06002
peqSTAR 2X gradient**	PEQL95-08002
peqSTAR XS**	PEQL95-03002
VWR microvolume spectrophotometer mySPEC*	VWRI732-2533
VWR microvolume spectrophotometer mySPEC Touch*	VWRI732-2534
VWR microvolume spectrophotometer mySPEC Twin*	VWRI732-2535
VWR microvolume spectrophotometer mySPEC Twin Touch*	VWRI732-2536
3D cell culture dish with 3D scaffold insert, 35 mm	734-2967
3D cell culture dish with 3D scaffold insert, 60 mm	734-2968
3D cell culture dish with 3D scaffold insert, 70 mm	734-2969
6-well plate with 3D scaffold inserts	734-2970
12-well plate with 3D scaffold inserts	734-2971
24-well plate with 3D scaffold inserts	734-2972
Flask tissue culture 25 cm² vent cap sterile	734-2311
Flask tissue culture 75 cm² vent cap sterile	734-2313
Flask tissue culture 75 cm² plug seal sterile	734-2314
Flask tissue culture 182.5 cm² vent cap sterile	734-2315
Dish tissue culture 35 mm sterile	734-2317
Dish tissue culture 60 mm grip. ring sterile	734-2318
Dish tissue culture 90 mm sterile	734-2320
Dish tissue culture 100 mm grip. ring sterile	734-2321
Dish tissue culture 150 mm sterile	734-2322
Plate tissue culture 6 wells sterile	734-2323
Plate tissue culture 12 wells sterile	734-2324
Plate tissue culture 24 wells sterile	734-2325
Plate tissue culture 48 wells sterile	734-2326
Plate tissue culture 96 well f bottom sterile	734-2327
Plate tissue culture 96 well u bottom sterile	734-2328

\* Country exclusions apply to these products. Check VWR.com for details.

Cont	:act (	details	tor	GΕ	repr	esen	tative
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## Contact details for VWR representative

<sup>\*\*</sup> Available with alternative part numbers in some countries. Check VWR.com for details.

## Swedish scientists make amazing spider silk from modified *E. coli* bacteria

The Stockholm-based biomaterials company is using genetically engineered bacteria and GE protein purification technology to produce large quantities of the so-called spidroin

proteins found in dragline silk, and then customize them for a variety of specific purposes. "Man-made spider silk can be adjusted to contain specific parts that bind

to cells and promote wound healing, thereby enabling use within fields of tissue engineering, diagnostics and cell culture," says Kristina Martinell, Spiber's production director. "In short, it's a tailor-made biomaterial."

Spiber can now manufacture spider silk fiber, film, foam and even mesh. The company says that the material is as strong as mammalian tendons and remains stable at boiling temperatures of up to 267 degrees Celsius (512 Fahrenheit).

Over time, the company's technique has evolved to keep the material soluble until it is ready to be shaped into the arrangements needed for various applications.

As a result, the range of potential products is huge. The company is working to apply spider silk in several medical fields, including cardiology, heart tissue regeneration, bone reconstruction, skin cell growth and vaccines.





→ READ STORY HERE



## The testament of quality

There can be few companies anywhere in the world that can lay claim to such distinguished clients as Whatman in the past 250 years.

In November 1767, the artistic Thomas Gainsborough wrote; "I beg you to accept my sincerest thanks for the favour you have done me concerning the paper for drawings. I had set my heart upon getting some of it, as it is so completely what I have long been in search of... Upon my honor I would give a guinea a quire for a dozen quires of it." Whatman paper became enormously popular with leading artists such as J M W Turner, and the quality and durability of English papers, including those made by Whatman, gave

the English watercolour school a 50 year advantage over European artists. At the end of the 18th century, the erratic genius William Blake used Whatman papers for four of his illuminated books, the public being informed that these were printed on "the most beautiful wove paper that could be procured."

Throughout history, heads of state and world leaders have shown a particular penchant for Whatman paper. Napoleon sat for five hours on the bleak island of St. Helena writing his long and detailed will on Whatman paper only three weeks before his death in 1821. George Washington signed many state documents on Whatman paper.

Queen Victoria chose Whatman paper for her personal correspondence.

In the 1930's Soviet leaders used Whatman paper to publish the 5 year plan for the future of the USSR, while the peace treaty with Japan was signed on Whatman paper at the close of World War II.

Today, Whatman filters have gained universal acceptance among the major scientific and industrial concerns of the developed world, from Japan to the United States, the UK to southeast Asia.

A remarkable testimony to Whatman quality – achieved through the pursuit of excellence.









## Combine protein purification techniques

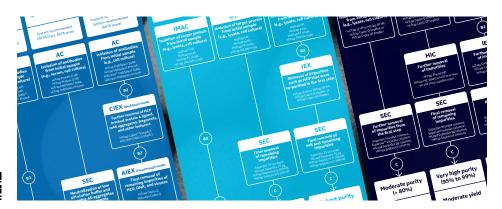
#### Free Wall Poster!

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.







## LOOK-UP - PRODUCT INFORMATION

Easy protein purification with ÄKTA start





## **LOOK-UP - APP**

## **Purify App**

The purify app simplifies the job of choosing the right chromatography media and columns for your application. Based upon your answers to certain questions, the tool will guide you to a recommended product. From there, you can follow the link to the product page for more information.







## **NEW PRODUCT**

Sample preparation with the Protein Prep syringe filter for ÄKTA systems

ÄKTA start system

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 impact 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  $\mu m$  pore size filter if the particle size of the chromatography resin is < 30  $\mu m$







## Tips for successful immunoprecipitation

For immunoprecipitation (IP), there are four main components that you can control to get superior results: protocol, resin, format, and antibody.

Choose direct or indirect method as basic protocol shown in the picture. In the direct method (pre-immobilized antibody) the antibodies are attached to the beads in advance. The direct method gives you better control of the antibody binding and is the only way if you wish to immobilize the antibodies covalently to the beads. This way, you can make sure that the antibodies will not elute and interfere with the subsequent analysis. In the Indirect method (free antibody) antibodies are incubated with the antigen before beads are added. The indirect method is very easy to use, has fewer steps, and can be used if the antibodies do not interfere with following steps.

There are two well-suited protein ligands for binding IgG type of antibodies: Protein A and Protein G. Both bind strongly to the constant region of the antibody, meaning that they will not interfere with the antibody-antigen binding. Protein A and Protein G bind to slightly different parts of the antibody, which will impact binding to antibodies from different species.

Finally you need to decide on a suitable format, depending on the number of samples, laboratory equipment, and your personal preferences.

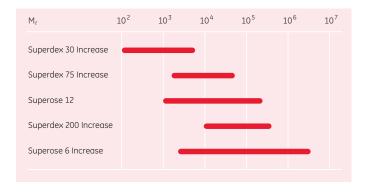
## **Pre-immobilized** Free antibody antibody approach approach Antigen sample sample Immune complex Separate Separate complex Discard **Analyze** precipitated antigen

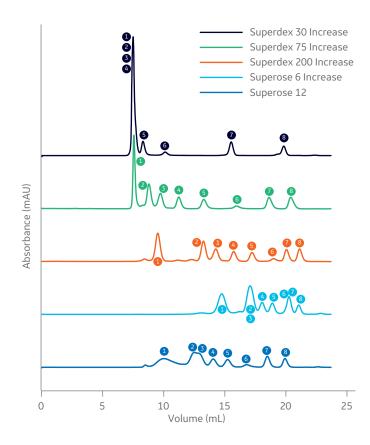




## Superose<sup>™</sup> 12 columns replacement

On Dec. 15 this year, Superose 12 columns will be discontinued. They can already be replaced by our new-generation size exclusion chromatography (SEC) columns—Superdex Increase and Superose Increase. The new-generation SEC resins cover the fractionation range for Superose 12 as shown in the pictures. The chromatogram shows the separation of a protein mix with Superose 12 and with the four replacement SEC resins. More details about the SEC column offering can be found in the Selection Guide (18112419 AJ) or the SEC handbook (18102218 AM).





Approximate fractionation ranges of Superose 12 and its new generation replacement columns and corresponding chromatograms (1) Ferritin (440 000), (2) Conalbumin (75 000), (3) Ovalbumin (44 300), Carbonic Anhydrase (29 000), Ribonuclease (13 700), Aprotinin (6500), Vitamin B12 (1200) and Cytidine (240)

→ READ MORE HERE



## **NEW PRODUCT**

## MabSelect PrismA – significantly enhanced alkaline stability and capacity

In research applications when different types of antibodies are purified on the same column, preventing cross-contamination while maintaining recovery is important. Sodium hydroxide is an efficient, low-cost, and easy-to-dispose reagent when thorough cleaning is required. Rigorous cleaning with sodium hydroxide reduces the risk of contamination from host cell proteins, microbial growth in the prepacked column, as well as carryover between purifications.

With the enhanced alkaline-stability of the protein A ligand, MabSelect PrismA can be cleaned with high NaOH concentrations (up to 1 M) with maintained capacity, meaning that HiTrap MabSelect PrismA and HiScreen™ MabSelect PrismA columns may be confidently cleaned for reuse.

The resin retains more than 95% of its initial DBC after 25 cycles with 1.0 M NaOH, while only about 60% of the initial DBC of Protein A Sepharose High Performance, and rProtein A Sepharose Fast Flow remains after less number of CIP cycles (15 or lower).







## Speeding up run time for viscous samples

Viscous samples, such as E. coli extract, can increase back pressure on the column and force you to reduce the flow rate, resulting in longer load times. Utilizing a robust, modern, and rigid resin—such as Capto Q ImpRes—reduces sample run time significantly by

allowing high flow rate with low column backpressure, as seen in the example below.

HiTrap Capto ImpRes contains Capto ImpRes high-flow agarose resin, which has much better pressure/flow properties than Sepharose High Performance resin. (Capto ImpRes resin has maximum flow rate of 300 cm/h vs 150 cm/h for Sepharose High Performance.) In this example, the IEX total run time is 30% shorter.





## Example with 50 mL E. coli lysate run in cold room

Equilibration 2.5 min

HiTrap Capto ImpRes 1 mL Sample loading – 1.9 mL/min 26 min Wash and gradient elution

15 min

Total time 43.5 min



quilibration

HiTrap Q HP 1 mL Sample loading – 1.1 mL/min 45 min Wash and gradient elution

15 min

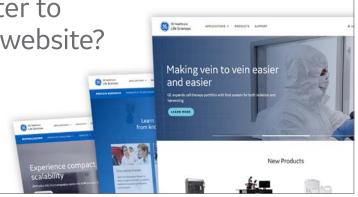
Total time 62.5 min

## Did you know you can register to GE Healthcare Life Sciences website?

- Gain access to more tools for your chromatography systems
- Access knowledge centre
- Access additional UNICORN tools free of charge
- Gain access to "My Instruments"

→ REGISTER TODAY





## Goodbye bed height, hello bed volume

Keeping a constant residence time is a wellestablished strategy when scaling up a chromatography step. Typically, this is achieved by keeping the bed height and linear flow velocity constant.

## Keep bed height flexible to reduce costs

Rather than strict adherence to constant linear velocity (cm/h), you can try measuring column volumes per hour (CV/h) for increased flexibility.

Instead of just having the option of different column diameters, CV/h measurement opens upforchangestobedheightaswellasdiameter. With this approach, you can avoid overcapacity of the column. That way you can reduce resin- related costs, particularly at manufacturing scale.

If you have your own examples, try **these chromatography calculator tools**. To calculate your residence time, use 60/(CV/h).

The concept of measuring CV/h is especially useful in affinity chromatography where the residence time determines the capacity independent of scale or bed height.







## Interpreting protein binding capacities for chromatography resins

Comparing protein binding capacities of resin from different suppliers is not straightforward, because different methods may have been used and these methods are not always stated. However, this parameter is critical, because it determines how much resin is needed to purify a certain amount of protein.

The capacities that suppliers specify for their chromatography resin may be based on different: (1) modes of measurement (static or dynamic), (2) experimental conditions (pH, salt/conductivity, protein identity and concentration) and (3) units of measure (i.e., capacity per milliliter wet chromatography medium or per gram dry medium). When inquiring about these conditions, it is worthwhile to ask the vendor whether the binding capacity was determined under static or dynamic conditions.

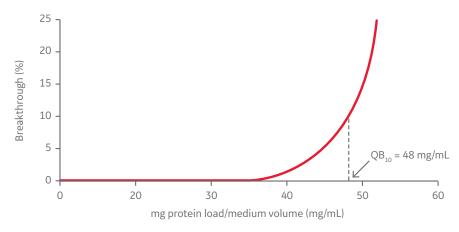
The static binding capacity (SBC, also called total protein capacity) is normally measured in batch mode in a beaker. SBC is usually reported as the maximum amount of protein bound to a chromatography resin at given solvent and protein concentration conditions.

Dynamic binding capacity (DBC) is the binding capacity under operating conditions (i.e., in a packed affinity chromatography column during sample application). The DBC of a chromatography resin is the amount of target protein that binds under given flow conditions before a significant breakthrough of unbound protein occurs. A breakthrough

curve is generated by graphing the amount of protein loaded versus the percent breakthrough. The DBC can be determined on the breakthrough curve at a loss of, for example, 10% protein (referred to as the QB<sub>10</sub> value, see picture).







Dynamic binding capacity determination breakthrough curve.

## 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 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 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 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.

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.







## Mad labs – late night catastrophe

It was a cold and stormy night a	nd I was		late in the lab to finish my
, , ,		Verb ending in "ing"	,
	testing when suddenl	y, I knocked over a flask of	
Noun			Noun
onto the bench.		I did not get any of the	
	Adverb		Noun
on me since I had Whatman		on my counter. Bad went to	
	Product Name		Adjective
as my samples had a lot of		and I was struggling to	
	Noun		Verb
them with any old pre-filter syrin	nge filter, I needed my Whatm	anProduct Name	syringe filter. I was
		Product Name	
stuck and could not	m Verb	ny experiment so I thought	Adjective
and went to the VWR	Noun	room where I knew t	hey would have my Whatman
	syringe filter. All was _		again; the sample was
Product Name		Adjective	
filtered and I was able to		all my samples	in
	Verb		Adverb
time to meet our			
	Noun		

## DIBE technology for reliable HCP detection

Host cell protein (HCP) is a primary impurity and a critical quality attribute (CQA) for biopharmaceuticals (biologics). HCP affects product quality, safety and efficacy. HCP ELISA is the gold standard of HCP detection and measurement, which requires polyclonal Antibodies (Ab) with broad reactivity against a wide range of potential HCPs.

Regulatory authorities require the characterization of the ELISA Abs used in the HCP ELISA assay. 2-D gel electrophoresis followed by Western blotting is the recommended approach to characterize HCP ELISA antibodies and their coverages.

2D differential in blot electrophoresis (2D-DIBE) combined with Western blotting is a powerful technology for separation and visualization of complex protein mixtures such as HCPs.

High sensitivity – Fluorescent multiplexed methodology based on CyDye pre-labeled Western blotting, and image acquisition with Amersham Typhoon™ laser scanner deliver high sensitivity for HCP detection.

Minimal variation - Labeled proteins can be directly compared to the proteins detected by CyDye pre-labeled antibodies on the same membrane.

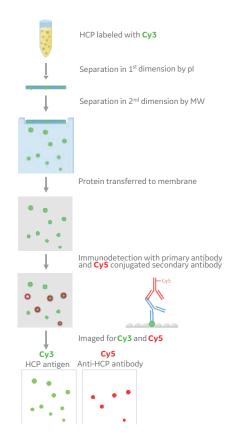
No mismatches - Multiplex fluorescence image acquisition with the Amersham Typhoon simultaneously captures both HCP antigen and anti-HCP antibody images from a single membrane.

Fast evaluation - Melanie™ Coverage software helps investigators evaluate the data in less time, with greater confidence.

#### 2D-DIBE for HCP coverage assay

Total HCPs are labeled with CyDye DIGE Cy3 minimal dye. After 2D electrophoresis, protein spots are transferred onto a PVDF membrane. The HCP antibody is applied to the membrane and visualized by Western blot with Cy5 fluorescence. Those two images are then overlaid. Cy3 labeled total HCP spot and Cy5 immunodetected spot overlay is confirmed by Melanie Coverage analysis software with 3D visualization. Finally, Melanie Coverage provides a coverage percentage value for this assay.

With the goal of helping you achieve the best results, we deliver 2D-DIBE products that improve data quality when compared to traditional 2D experiments and Western blotting, and can be integrated into a complete HCP analysis solution.



# VWR introduces the new mySPEC series of spectrophotometers: Cutting edge technology for the next generation in microvolume measurement instrumentation

The VWR® mySPEC microvolume spectrophotometers combine gold standard detection technology with new levels of user-interaction and data analysis. The VWR mySPEC models enable highly accurate UV/Vis analyses of 1  $\mu l$  samples with remarkable reproducibility. Processing of sample is more robust than ever before.

The sophisticated sample retention system of all VWR mySPEC instruments eliminates the need for cuvettes, capillaries or other consumables resulting in decreased measurement cycle times and full cost control.

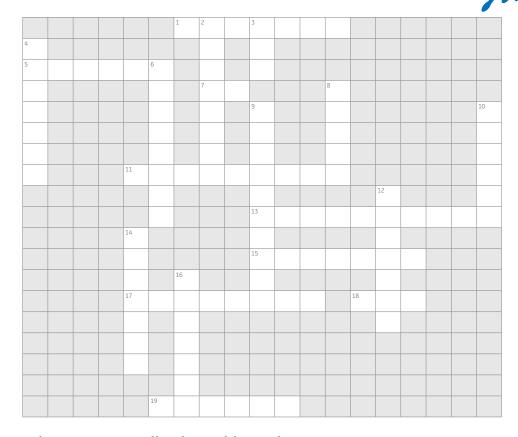
vailable with microvolume sample retention system only or additional cuvette capabilities, controlled either by an external laptop or onboeard touch screen PC, the VWR mySPEC is the new system of choice for the busy and modern life science laboratory.

## → DOWNLOAD APPLICATION NOTES









### Across

- 1. Gourd of Ichabad Crane's Sleepy Hollow
- 5. Prepacked columns with improved MabSelect protein resin
- 7. Scale from 0 to 14
- 11. Long winter nap
- 13. Pigments Producing Fall Leaf Colors
- 15. Bulk liquid filtration
- 17. Fall labyrinth in old McDonalds farm
- 18. 'One twig' cancer therapeutic
- 19. New Whatman sterile syringe filter

#### Down

- 2. Filter with a multiwell plate formant
- 3. Kary Mullis's Nobel Prize Discovery
- 4. HPLC certified; syringe filter; 30mm
- 6. Highly specific, ligand chromatography
- 8. Fall's migratory bird
- 9. Goat horn full of fruit
- 10. Amersham's PVDF blotting membrane
- 12. Magnetic Beads to Capture Molecules of Interest
- 14. Smallest amino acid
- 16. Western blotting membrane, nitrocellulose

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We're Thinking **Pink** in October

October is breast cancer awareness month and

October is breast cancer awareness month and GE will be supporting advancements in cancer research by donating up to €100,000 to the Breast Cancer Research Foundation.

GE's Whatman laboratory filtration products are used in laboratories supporting critical work in areas from cancer research to drug discovery and development. Learn nore at:

gelifesciences.com/ThinkPink2018

To find out more about the work of the BCRF check the link:

bcrf.org/researchers







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