# StratoSpheres<sup>™</sup> PL-HCO3 MP Resin

# POLYMER-SUPPORTED BICARBONATE FOR ACID REMOVAL



*PL-HCO3 MP Resin is a quaternized macroporous polystyrene strong anion exchanger with a bicarbonate counter-ion.* 

Available in bulk format or pre-packed in SPE cartridges it provides the most convenient method of removing residual acids and counter-ions such as trifluoroacetate.

Pharmaceutical drug discovery utilizes organic acids throughout the development process, as raw materials, deprotection agents or as ion pair reagents in HPLC purification. Consequently, target compounds are rarely obtained in a pure form. The remaining traces of acid can seriously compromise compound storage, leading to degradation and hydrolysis, or can interfere with analytical techniques. Instead of lengthy freeze-drying to try and remove the acid, or gel filtration to simply replace one counter ion with another, PL-HCO3 MP resin is a rapid and economic solution for providing compounds in free base form.

# **Key Benefits**

- Rapid and convenient. Compatible with high throughput technologies, or simply used manually as required.
- **Cost effective.** Economical and simple to use; requires no specialized laboratory equipment or instrumentation.
- Superior technology. Avoids the pitfalls of repetitive freeze drying.
- Versatile. Can remove a wide range of acids from solution, including trifluoroacetic acid, formic acid or hydrochloric acid.

NOTICE: This document contains references to Varian. Please note that Varian, Inc. is now part of Agilent Technologies. For more information, go to **www.agilent.com/chem.** 



## Acid Removal

Trifluoroacetic acid is commonly used within the pharmaceutical industry, particularly during the drug discovery process. It is a volatile organic acid 10,000x stronger than acetic acid. It has found use as an acidic deprotection agent and also as an ion pair reagent in reverse phase HPLC. However as a result of its use, compounds are frequently isolated as their TFA salt.

As a consequence, many analytical techniques are compromised: assays can be poisoned by the presence of TFA, LC/MS analysis can suffer from ion suppression, and 19F NMR and FT-IR have interfering signals arising from the trifluoroacetate.

Even if repetitive freeze-drying is used (a time-consuming and costly technique), the presence of TFA can still be problematical and can even lead to compound degradation through hydrolysis.

# Properties of CF<sub>3</sub>CO<sub>2</sub>H

MW 114.02 D 1.535 g/mL BPt 72.4 °C

0.1% v/v TFA solution contains 1.0 mL per liter. This equates to 1.535 g per 1000 g water, or 13.4 mmol/liter.

# Other Acids Commonly Removed

Formic acid,  $HCO_2H$ Acetic acid,  $CH_3CO_2H$ Hydrochloric acid, HCI

# StratoSpheres PL-HCO3 MP

StratoSpheres PL-HCO3 MP is a rigid, macroporous polystyrene sorbent. The quaternized strong anion exchanger is provided with a bicarbonate counter ion in both bulk format (loose) and pre-packed cartridges. The particle size is optimized for easy handling and can be used under gravity flow or by addition of the powder to a solution.



Commercially available alternatives, such as silicasupported carbonate, suffer from some disadvantages. The silica matrix is less pH stable than a polymeric support and is inherently much lower loading. A further problem arises from the fact that a sodium salt by-product can be released (such as sodium trifluoroacetate). This can then contaminate your product.

StratoSpheres PL-HCO3 MP can be used to remove other acids including, but not limited to: formic acid (formate counter ions), acetic acid (acetate counter ions) and hydrochloric acid (chloride counter ions).

#### Simple Protocol



**Pre-condition** the sorbent by rinsing with methanol (or similar solvent). This helps to overcome surface tension effects by "wetting" the sorbent.

**Apply Sample** solution and allow to percolate through under gravity, or stir a suspension containing the sorbent. Collect filtrate containing free base product.

Wash sorbent with methanol and collect filtrate.

Isolate compound from combined filtrate and washings.

#### Removal of Excess Acid or Ion-pair Reagent

A simple method of eliminating mobile phase additives or other excess acids is to simply allow the solution to come into contact with the PL-HCO3 MP polymer support.

The bicarbonate counter ion supported on the resin exchanges with the acid species, leaving the free base product in solution.

The only by-products from using StratoSpheres PL-HCO3 MP are water and carbon dioxide.



#### **Other Applications**

PL-HCO3 MP Resin is a versatile product and can be used for a number of acid quenching applications.

Although the removal of TFA remains the most prominent application (due to the widespread use of this reagent), other acids can also be removed.

This includes the sequestration of unreacted acids, following amide bond forming reactions, for example.





#### Free Basing of Peptides

Peptides are frequently purified by reverse phase HPLC using TFA as an ion pair reagent. Isolating the peptide without a TFA counter ion is difficult. Simply evaporating the solvent leads to a concentration of TFA which can start to hydrolyze the amide bonds.

Displacement of TFA with a stronger acid, such as hydrochloric acid, still leaves the potential for compound degradation during the isolation phase.

StratoSpheres PL-HCO3 MP Resin is highly effective at removing TFA. The characteristic peak at  $\sim$  -76 ppm is seen in the 19F NMR spectrum opposite (black trace).

After simply passing the solution through a PL-HCO3 MP SPE device, the signal at -76 ppm completely disappears, resulting in a free base peptide (blue signal).



#### **Recommended Configurations**

The extremely high capacity of StratoSpheres PL-HCO3 MP means that it is possible to treat a much larger volume of solution than the cartridge can actually contain. Under such circumstances, alternative arrangements for delivering larger volumes under gravity flow are possible.

By employing an adapter cap, a larger reservoir can be fitted or the solution can be drawn from a bottle using a suitable length of tubing:



#### **Ordering Information**

Description	Pack Size	Part No.	Nominal capacity *	
PL-HCO3 MP 1.8 mmol/g 100Å 150-300 μm	5 g	PL3540-1603	1.8 mmol/g	
PL-HCO3 MP 1.8 mmol/g 100Å 150-300 μm	25 g	PL3540-3603	1.8 mmol/g	
PL-HCO3 MP 1.8 mmol/g 100Å 150-300 μm	100 g	PL3540-4603	1.8 mmol/g	
PL-HCO3 MP 1.8 mmol/g 100Å 150-300 μm	1 kg	PL3540-6603	1.8 mmol/g	
Description	Pack Size	Part No.	Nominal capacity *	TFA Removed *
PL-HCO3 MP SPE 100 mg/6 mL	50/PK	PL3540-A603	0.2 mmol	8-10 mL 0.1% TFA
PL-HCO3 MP SPE 200 mg/6 mL	50/PK	PL3540-B603	0.4 mmol	16-20 mL 0.1% TFA
PL-HCO3 MP SPE 500 mg/6 mL	50/PK	PL3540-C603	0.9 mmol	40-50 mL 0.1% TFA
PL-HCO3 MP SPE 5 g/60 mL	10/PK	PL3540-J603	9.0 mmol	400-500 mL 0.1% TFA

\* The quoted capacity is the maximum recommended amount.

#### Accessories

The following components may also be of interest and will allow larger volumes of solution to be passed through the pre-packed cartridges.

Description	Pack Size	Part No.
Adapter caps for 6 mL cartridges	15/PK	12131001
Luer stopcocks	15/PK	12131005
Empty 20 mL reservoir	100/PK	12131011
Empty 60 mL reservoir	100/PK	12131012



Varian, Inc. www.varianinc.com North America: 800.926.3000, 925.939.2400 Europe The Netherlands: 31.118.67.1000 Asia Pacific Australia: 613.9560.7133 Latin America Brazil: 55.11.3238.0400

Other sales offices and dealers throughout the worldcheck our Web site.

Chromatography • Spectroscopy • Mass Spectrometry • Magnetic Resonance Spectroscopy and Imaging • X-Ray Crystallography • Dissolution • Consumables • Data Systems • Vacuum

StratoSpheres, Varian and the Varian logo are trademarks or registered trademarks of Varian, Inc. in the U.S. and other countries. <sup>o</sup> 2009 Varian, Inc.