Table 1: F-SPE CARTRIDGES LOADING CAPACITY		
Size	Size	Size
2 g, 8 mL tube	801-0027S	100-300 mg
5 g, 10 mL tube	801-0058S	400-600 mg
10 g, 60 mL tube	801-0109S	0.5-1.5 g
20 g, 60 mL tube	801-0209A	1.5-2.5 g

WHAT'S IN FluoroFlash® CARTRIDGES? Fluorous solid-phase extraction (F-SPE) cartridges are packed with silica gel containing a perfluorooctylethylsilyl ($Si(CH_2)_2C_8F_{17}$) bonded phase. Fluorous silica separates compounds based primarily on fluorine content. It selectively retains fluorous molecules while non-fluorous organic compounds, regardless of polarity, are not retained.

WHAT'S THE UTILITY OF FLUOROUS SPE (F-SPE)? F-SPE is used for quick separation of reaction mixtures involving fluorous reagents,[1,2] protecting groups,[3] tags[4] and scavengers.[5] In a simple two-step elution, fluorous compounds and non-fluorous compounds are separated into two fractions.

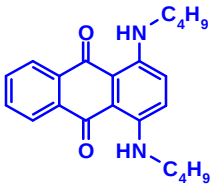
GENERAL PROCEDURES FOR F-SPE: Following completion of a reaction involving a fluorous-tagged molecule, the reaction mixture is loaded onto an F-SPE cartridge. Using a fluorophobic wash such as 80:20 MeOH:H_2O, the non-fluorous organic compounds are washed off the cartridge. The fluorous compounds are then washed off the cartridge using a fluorophilic second wash such as MeOH, acetone or THF. The loading capacity for fluorous SPE is typically between 5-10% by weight of fluorous silica gel. The cartridge can be regenerated by washing thoroughly with acetone or THF and reused multiple times (See FAQ #13).

DEMONSTRATION WITH DYES: The following dye separation demonstrates how F-SPE works. The non-fluorous compound is Solvent Blue® Dye while the fluorous compound is F-Orange Dye.

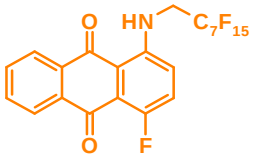
Figure 1 shows fluorous cartridges containing the dye mixture in three different stages of elution. The left-hand test-tube illustrates how the F-SPE cartridge appears after loading a mixture of the two dyes and elution with a small amount of 80:20 MeOH:H_2O. The center tube shows how the non-fluorous components (blue fraction) are washed from the cartridge by using more 80:20 MeOH:H_2O. The adsorbed fluorous dye is not eluted even with extensive flushing with 80:20 MeOH:H_2O and remains on the cartridge. Finally, the orange fluorous dye is easily eluted out with 100% MeOH or THF, as shown by the third tube.

Multiple fraction collection and analysis are not required—there is one organic wash and one fluorous wash. Since the separation process is highly reproducible and functional group independent, it can be easily automated or used as plate-to-plate format.

Figure 1. F-SPE with Blue (Organic) and Orange (Fluorous) Dyes



Solvent Blue



F-Orange

Left tube: beginning of fluorophobic wash (80:20 MeOH:H_2O)
Center tube: end of fluorophobic wash
Right tube: end of fluorophilic wash (100% MeOH)

TYPICAL F-SPE PROCEDURE (For separation of a reaction mixture (100-300 mg) in 0.5 mL of DMF with a 2g SPE cartridge):

Step 1 - Wash the new cartridge: Wash a new cartridge with 1 mL of DMF under a vacuum or positive pressure depending on your SPE manifold. This step can be omitted with recycled cartridges.

Step 2 - Preconditioning: Pass through 6 mL of 80:20 MeOH:H_2O to condition the cartridge. Discard the preconditioning eluent.

Step 3 - Sample loading: Dissolve sample (100-300 mg) in 0.5 mL of DMF and load onto the cartridge using vacuum or positive pressure to ensure that the sample is completely adsorbed onto the cartridge (see Table 2 for alternative loading solvents).

Step 4 – Fluorophobic elution: Wash with 6-8 mL of 80:20 MeOH:H_2O to obtain the fraction containing the organic compounds.

Step 5 – Fluorophilic elution: Wash with 8 mL of MeOH to obtain the fraction containing the fluorous compounds.

Step 6 – Regenerate the cartridge: To regenerate the SPE cartridge, wash with 6-10mL of THF or acetone before reuse. Then follow Steps 2-5.

Table 2: Suggested Maximum Loading Solvent Volumes for C_8F_{17} Tagged Substrates

	Maximum Loading Volume		
Solvent	2 g cartridge	5 g cartridge	10 g cartridge
THF	0.2 mL	0.5 mL	1.0 mL
CH_2Cl_2	0.2 mL	0.5 mL	1.0 mL
MeOH	0.2 mL	0.5 mL	1.0 mL
DMF	0.8mL	2.0 mL	4.0 mL
DMF/H_2O (9:1)	2.0mL	5.0mL	10mL
DMSO	2.0mL	5.0mL	10mL

Please contact FTI for information about the use of other solvents with F-SPE

FREQUENTLY ASKED QUESTIONS FOR F- SPE:

1. What kind of SPE manifold can be used for F-SPE?
   A: Either a vacuum or positive pressure SPE manifold can be used for F-SPE. We often use a 2 x 12 vacuum manifold such as those commercially available from Supelco, Waters, and United Chemical Technologies, Inc.

2. Can I use F-SPE cartridges without pre-conditioning them?
   A: As with other SPE cartridges, pre-conditioning prior to use is recommended.

3. How much sample can I load onto Fluoro*Flash*® SPE cartridges?
   A: Fluoro*Flash*® silica gel typically has a loading capacity of 5-10% of crude reaction mixture by weight.

4. What solvents are recommended as loading solvents?
   A: The sample can be dissolved in any organic solvent. We like 90:10 DMF:H_2O, DMF or DMSO, since more solvent can be used. On a 2g F-SPE cartridge, the use of <2mL of sovent is ideal. Other solvents such as CH_2Cl_2, THF, MeOH, and acetone can be used although maximum recommended loading volumes are less. See Table 2 for recommended amounts of loading solvent. If a larger amount of solvent is needed for sample loading, then consider using a larger F-SPE cartridge.

5. Is the amount of loading solvent used to dissolve the sample important?
   A: The amount of solvent used to dissolve and load the sample is crucial and should be minimized. Separation of organic and fluorous compounds can fail if too much loading solvent is used because the loading solvent can elute both fluorous and non-fluorous compounds off fluorous silica gel. This is called breakthrough, and is a common problem seen by first-time users of F-SPE.

6. How can I diagnose and prevent breakthrough?
   A: The usual symptom of breakthrough is that some of the fluorous compound comes off early in the organic fraction, but the bulk of the fluorous compound is retained on the column. This happens because the loading solvent elutes the fluorous compounds, but the elution stops as soon as the fluorophobic solvent elutes the loading solvent from the column. Breakthrough problems can often be solved by using less loading solvent. Other solutions are to use a more fluorophobic loading solvent, use a larger column or lower loading amounts.

7. Do I need to use fluorous solvents in F-SPE?
   A: No, both the fluorophobic and the fluorophilic wash use common organic solvents.

8. What solvents should be used to elute non-fluorous compounds while retaining fluorous compounds?
   A: We often use 80:20 MeOH:H_2O for the initial wash. Increasing the water content decreases the chance of breakthrough while deceasing the water content increases the chance of breakthrough. 90:10 DMF:H_2O is also a good solvent. If you prefer H_2O-free solvent, DMSO is the choice.

9. Do I need to use water in the fluorophobic wash?
   A: Water is the fluorophobic solvent *par excellence*, so a little water goes a long way. The addition of water ensures retention of the fluorous compounds onto the fluorous silica gel. If your molecule has 21 fluorines or more, then you can begin to reduce or even eliminate the water from your fluorophobic solvent.

10. What solvent needs to be used to elute the fluorous compounds?
    A: We recommend using 100% MeOH or MeCN although more fluorophilic solvents such as acetone or THF can be used as well.