



**PerkinElmer**<sup>™</sup>  
life sciences.

## **Sample Preparation & Cocktail Selection**

**Jock Thomson**

**Global Product Manager Cocktails & Vials**

## ➤ **Tritium**

- **Milk, crops, animals and water samples**

## ➤ **Carbon-14**

- **Milk, crops, animals and sea water**

## ➤ **Other Isotopes**

- **Strontium-90, Cesium-134 & 137,**
  - **Milk, crops, animals and soil/sediments**
- **Radon-222**
  - **Water and air**

## **Water**

- **Distilled and deionized water**
- **Ground water (rain water, river water, tap water)**
- **Sea water**

## **Milk**

## **Animals**

- **Urine samples**
- **Tissue samples**

## **Crops**

## Sea water

- Inorganic  $^{14}\text{C}$  and organic  $^{14}\text{C}$  (phytoplankton)

## Milk

## Animals

- Urine samples
- Tissue samples

## Crops

### **Strontium-90 (Sal Scarpitta)**

- **Milk, crops, animals and soil/sediments**

### **Cesium-134 & 137**

- **Milk, crops, animals and soil/sediments**

### **Radon-222 (Mike Cantaloub)**

- **Water and air**

## Direct addition

- Suitable for water, urine, plasma, serum and milk samples.

## Distillation (plain, vacuum & azeotropic)

- Suitable for water and some plant samples.

## Deionization (includes Eichrom technology)

- Suitable for water.

## Solubilization

- Suitable for tissue samples, crop samples, plasma and serum.

## Combustion

- Suitable for all above sample types

## Water

	Capacity in mL per 10.0mL cocktail at 20°C				
	Distilled	Deionized	Sea Water	Tap Water	River/Rain
Ultima Gold LLT	12.0mL	12.0mL	5.5mL	12.0mL	12.0mL
Ultima Gold XR	10.0mL	10.0mL	7.0mL	10.0mL	10.0mL
Insta-Gel Plus	10.0mL*	10.0mL*	No Data	10.0mL*	10.0mL*
Ultima Gold	3.2mL	3.2mL	No Data	No Data	No Data
Opti-Fluor	1.8mL	1.8mL	No Data	No Data	No Data
Emulsifier Safe	3.2mL	3.3mL	No Data	No Data	No Data
Pico-Fluor 15	1.6mL	1.6mL	No Data	No Data	No Data
Pico-Fluor 40	2.3mL	2.3mL	No Data	No Data	No Data
Hionic-Fluor	1.2mL	1.2mL	No Data	No Data	No Data

\* Forms a gel at this loading capacity

## Distilled water

- Plain or vacuum distillation is useful for water samples which do not contain volatile materials but may have chemical contaminants. This sample preparation method has been applied to Urine samples.
- Recommended LSC cocktails:-
  - Ultima Gold LLT (safer)
  - Ultima gold XR (safer)
  - Insta-Gel Plus (classical)

## Deionized water

- Alternative method of removing chemical contaminants from water samples. Suitable for removing many anions and cations.
- Eichrom Tritium column removes most inorganic and certain organic contaminants.
- Recommended LSC cocktails :-
  - Ultima Gold LLT (safer)
  - Ultima Gold XR (safer)
  - Insta-Gel Plus (classical)

## Sea water

- Sea water contains predominantly sodium chloride with significant quantities of sulfates, potassium, calcium and magnesium.
- Total level of dissolved solids in mid ocean seawater is around 34,000ppm or 34ppt (parts per thousand).
- Level of sodium chloride is approx.. 25,000ppm or 25ppt, or 0.43M.
- Recommended LSC cocktails
  - Ultima Gold LLT
  - Ultima Gold XR

## Tap water

- This sample type will vary considerably depending upon the locality i.e. soft or hard water.
- Soft water usually has <200ppm TDS while hard water is >500 TDS (Total Dissolved Solids).
- Sample preparation can be either
  - Direct addition
  - Distillation to remove TDS,
  - Concentration and chemical treatment
  - Extraction using Eichrom resins.

## Tap water

- For direct addition and distilled samples the recommended LSC cocktails are :-
  - Ultima Gold LLT
  - Ultima Gold XR
  - Insta-Gel Plus\*
- \* Forms a gel at loadings >4mL in 10mL cocktail

## Tap water

- Concentration / chemical treatment
- Large volume (~1Lt) evaporated down to <100mL and then acidified (HCl). Converts Ca & Mg salts into soluble form.
- Further evaporated down to dryness.
- Residue dissolved in acid.
- Recommended LSC cocktails
  - Ultima Gold AB
  - Ultima Gold LLT

## Urine

- Urine is about 95% water, and it usually contains sodium chloride, urea, uric acid, and creatinine. It may contain a trace of amino acids and varying amounts of electrolytes, depending upon dietary intake.
- Direct sample addition is possible for any sample volume as there are LSC cocktails available which can accept up to 1:1 ratio of urine in the cocktail.
- In general sample volume is restricted to  $\leq 3.0$  mL to minimize the effect of color quench, especially if  $^3\text{H}$  is the isotope of interest.

## Urine

- There is a potential for sample/cocktail incompatibility, which becomes apparent by the appearance of a wispy precipitate, and is principally due to the presence of proteinaceous material in the urine.
- Certain alcohols such as ethanol and isopropyl alcohol are particularly effective in suppressing the appearance of the precipitate.
- Distillation of the urine sample overcomes the sample/cocktail incompatibility and the color quench problems. The only drawback is that this method increases sample preparation time considerably.

## Recommended LSC cocktails for Urine

Cocktail	Type	Max. urine* capacity @20°C	Recommended loading
Ultima Gold LLT	Safer	10.0mL	2.0mL
Ultima Gold	Safer	8.0mL	2.0mL
Pico-Fluor MI	Classical	4.0mL	4.0mL

\* Results based on human urine

Note :-

1. It is possible to add up to 1mL of either ethanol or isopropyl alcohol per 10mL cocktail, to suppress the appearance of the protein precipitate.
2. Urine samples from small animals are usually difficult. With these, either keep sample volume low, or dilute the urine with water.

The sample preparation of soil samples depends entirely on the radioisotope of interest.

Generally the first step will involve either :-

- Ashing to remove organic contaminants.
- Destruction of soil with strong mineral acids.

In many cases tracers are added for chemical yield determination.

After sample preparation samples can be analyzed :-

- Directly by LSC (discrete and gross)
- Further separated using Eichrom technology and then by LSC (discrete)

### Strontium\*

- Sample size 5 to 10g of ashed soil.
- Destruction of soil with conc.  $\text{HNO}_3$ , 40% HF,  $\text{H}_3\text{BO}_3$ .
- Evaporation and dissolve residue in 0.5M  $\text{HNO}_3$ .
- Add 5 to 10g Oxalic Acid.
- Adjust pH with  $\text{NH}_3$ .
- Remove Potassium.
- Remove other elements (if required).
- LSC determination of sample in 1M  $\text{HNO}_3$ .  
(Ultima Gold AB or LLT)

\* Vajda et al. "Determination of radiostrontium in soil samples",  
J. Radioanal. Nucl. Chem. 162, p307, 1992

# Soil Analysis

## Specific Isotope sample preparation

### Technetium\*

Technetium is leached from the soil sample using 1M HNO<sub>3</sub> and separated using TEVA-Spec resin prior to liquid scintillation counting.

\* Eichrom Procedure TCS01 (11/18/93)

### Uranium and Thorium\*\*

Ashed soil samples are treated with conc HNO<sub>3</sub> / conc HCl mixture, then with conc HNO<sub>3</sub> / conc. HF, evaporated to dryness and finally redissolved in 6M HNO<sub>3</sub>.

Eichrom UTEVA-Spec column can then be used to separate the Actinides prior to alpha counting.

\*\* Eichrom Procedure ACS06 (12/27/93)

### Notes :-

1. DIN based cocktails such as the Ultima Gold range must be used when doing Alpha/Beta on a Tri-Carb.
2. Selected cocktail must also be compatible with mineral acids - see Table.
3. If Nitric Acid at  $>3M$  concentration is present it will induce yellow color in the sample/cocktail matrix within 24 hours. Where possible keep the concentration to  $<2M$  or reconstitute sample in HCl.

## Performance with Mineral Acids

Sample	Ultima Gold	Ultima Gold XR	Ultima Gold AB	Ultima Gold LLT	Opti-Fluor	Emulsifier Safe/ Poly-Fluor
0.1M HCl	6.5mL	7.0mL	10.0mL	10.0mL	4.0mL	2.7mL
1.0M HCl	0.5mL	2.5mL	5.5mL	5.0mL	0.5mL	3.0mL
2.0M HCl	None	1.0mL	2.25mL	3.0mL	None	4.5mL
5.0M HCl	None	<0.5mL	2.0mL	1.5mL	None	0.5mL
1.0M HNO <sub>3</sub>	None	2.5mL	3.25mL	3.5mL	0.75mL	3.5mL
2.0M HNO <sub>3</sub>	0.5mL	2.0mL	2.25mL	2.5mL	0.75mL	3.5mL
3.0M HNO <sub>3</sub>	None	1.0mL	2.0mL	2.25mL	0.5mL	1.0mL
1.0M H <sub>2</sub> SO <sub>4</sub>	None	0.25mL	6.5mL	7.0mL	None	2.0mL
2.0M H <sub>2</sub> SO <sub>4</sub>	None	None	4.0mL	4.0mL	None	2.75mL
1.0M HClO <sub>4</sub>	2.0mL	2.0mL	2.25mL	2.25mL	1.5mL	1.0mL
2.0M HClO <sub>4</sub>	1.5mL	1.5mL	2.0mL	2.5mL	1.0mL	0.75mL
1.0M H <sub>3</sub> PO <sub>4</sub>	None	1.5mL	0.5-10.0mL	0.5-10.0mL	0.5-1.5mL	3.0mL
2.0M H <sub>3</sub> PO <sub>4</sub>	None	0.5mL	0.5-4.0mL	0.5-6.0mL	0.5-1.0mL	3.0mL

### Carbon-14

(e.g. biomass of bacteria in soil)

Soil (~50g) extracted with mixture of 150mL Methanol and 200mL Chloroform.

Evaporate extract to dryness under nitrogen.

Sample (~100mg) is now a yellow brown liquid.

Add 15mL Ultima Gold LLT for counting.

Use Low level Count Mode for environmental levels.

Notes :-

Use high purity solvents.

Do a blank as solvents can contribute up to 4cpm of  $^{14}\text{C}$  (0-156keV).

### Inorganic $^{14}\text{C}$

- Be careful about cocktail selection :-
- Ultima Gold, Ultima Gold XR, AB, LLT, MV all contain neutralized phosphate esters and are about pH 6.2-6.5 This slight acidity will cause slow release of  $^{14}\text{C}$  as  $^{14}\text{CO}_2$  from carbonates and bicarbonates containing samples - sea water and  $\text{CO}_2$  trapped in NaOH.
- Either add NaOH to adjust cocktail pH or use a non-acidic cocktail such as Hionic-Fluor, Opti-Fluor or Emulsifier Safe.

### Water samples

- Sometimes researchers distill water or collect ground water and then add acid to stabilize the water samples. The addition of even small amounts of acid can have a serious affect on sample capacity.
- It is recommended that the cocktail be re-evaluated with this modified sample to ensure that the mixture is still homogeneous.
- This is especially important if plastic vials are being used, as they are not transparent.
- Additionally carry out the evaluation in glass vials at the counting temperature to be sure of stability over the count period.

### Calcium-45

- Calcium can be adsorbed onto the surface of glass vials. Fortunately the problem only becomes important when the Calcium concentration is very low ( $<0.00001\text{M}$ ). At these very low concentrations  $^{45}\text{Ca}$  can be “lost” through adsorption, especially if liquid transfers are happening.
- This problem can be overcome by one of two methods
  - Pre-coat the glass with non-active Calcium i.e. soak the vial with  $\text{CaCl}_2$  solution prior to use.
  - Use plastic vials instead of glass vials.

### Waste Disposal

- The Ultima Gold range of cocktails has been successfully tested for Biodegradability according to method OECD 301E (ISO Method 7827-1984).
- Certificates are available on request.
- Complete MSDS (Material safety Data Sheet) for every cocktail and reagent is available in either hard copy form, or can be printed off from PerkinElmer's Web site at <http://lifesciences.perkinelmer.com/index.asp.com>
- For disposal purposes, some disposal companies require elemental cocktail information (shown over).

# Selected LSC cocktails - Elemental composition

Cocktail	Carbon C	Hydrogen H	Nitrogen N	Oxygen O	Phosphorus P	Sulphur S	Sodium Na	Average M.W.
Ultima Gold	78.9%	9.6%	0.2%	9.5%	1.4%	0.2%	0.2%	254.7
Ultima Gold XR	73.2%	10.0%	0.2%	15.2%	0.9%	0.3%	0.2%	297.5
Ultima Gold AB	76.3%	9.7%	0.1%	13.8%	0.1%	N/A	N/A	293.4
Ultima Gold MV	78.0%	10.1%	0.2%	10.2%	1.1%	0.2%	0.2%	261.7
Ultima Gold LLT	76.4%	9.7%	0.1%	13.8%	0.01%	N/A	N/A	293.3
Ultima Gold F	90.4%	9.45%	0.07%	0.08%	N/A	N/A	N/A	211.9
Insta-Gel Plus	70.5%	9.8%	0.02%	19.7%	N/A	N/A	N/A	315.6

# A few final thoughts !!!!!

Any LSC cocktail will work with any LSC -  
The trick is to get the best combination.

Poor sampling can only produce a bad sample.

A badly prepared sample can only produce bad  
results.

No amount of instrument sophistication can ever  
produce good results from a badly prepared  
sample.