Comprehensive Range of Clean-up Methods for Biological Matrices, from High to Low Fat Content

Calaprice C[1,2], Focant J-F[2]

[1]School of Geography, Earth and Environmental Sciences, University of Birmingham, B15 2TT Birmingham, United Kingdom
[2]Organic and Biological Analytical Chemistry (OBiAChem), University of Liège, 4000 Liège, Belgium

In the recent years PCDD/Fs and PCBs analysis has shown the need for fast and high throughput methods for food safety and biomonitoring. Normally sample preparation involves sophisticated and delicate multistage steps, which can also require several days. For biological matrices sample preparation consists of three main steps: fat extraction (where Dioxins and PCBs are dissolved), clean-up, and instrumental quantification. In its turn, sample clean-up involves two steps: fat digestion (by acid silica reaction or by size exclusion) and fractionation between Dioxins and PCBs (using Alumina or Florisil, and carbon column) to minimize possible instrumental interferences. There are several options for sample clean-up, manual and automated procedures [1]. Here we propose a comprehensive range of clean-up methods for biological samples ranging from 7 g to few mg of fat (blood). Methods are based on EconoPrep® automated system from FMS (Fluid Management systems, Watertown, MA, United States) and they all use MINI alumina (3g) and carbon:celite carbon column (ca:ce) for the fractionation. A different silica column is connected based on the amount of fat to process (Figure 1).

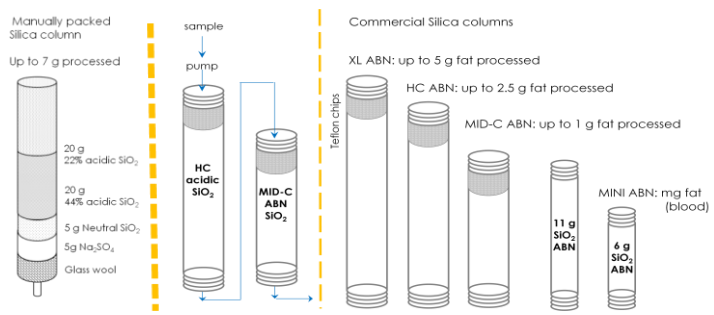

Figure 1: Silica columns for fat digestion with decreasing fat capacity (fc) from left to right:

- manually packed acidic silica, fc 7g
- "fat digestion silica"*, fc 7g
- XL ABN silica*, fc 5g
- HC ABN silica*, fc 2.5g
- MID-C ABN silica*, fc 1g
- Standard ABN silica*, fc <1g
- MINI ABN silica*, blood

**commercial columns*

Manually packed acidic silica column is used for preliminary fat digestion, followed by fractionation with MINI column set (MINI ABN silica, ca:ce, MINI alumina) on the automated system. "Fat digestion silica" columns are used in a 2 step automated method, where fat digestion is automated and fractionation is carried out with an identical MINI column set, independently to the original fat content. All methods developed allow faster and cheaper high throughput sample preparation. With the 2 step automated method, up 7g fat can be processed in 70 min, having a single standardized method for all food and feed matrices. One single module of the EconoPrep® can process 20 samples/day.

	Manual Si	Fat digest Si	XL ABN	HC ABN	MID-C ABN	STD ABN	MINI ABN
Volume mL	410	350	295	245	150	135	110
Time min	60	49	34.5	29.5	28	18	20

References:

1. Calaprice C and Focant J-F (2016) *Organohalogen Compounds*, **78** (773-776)