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The major function and primary strength of the AC is provision of reliable technically robust panels of assays for environmental contaminants and markers of exposure or behavior that are of interest to UK-CARES researchers (see **Tables 1-3**). Consistent with the themes of the Center, these include methods to support measurements of environmental chemical or biomarkers in the areas of water, air quality, and emerging threats. Unless otherwise indicated, limits of quantitation (LOQ) for organic contaminants are experimentally determined and represent the lowest concentration of analyte where the relative uncertainty for a single measurement is within ±20% determined using 3-5 injections of a range of concentrations of the analyte at, above, and below the estimated limit of quantitation. The methods listed below generally employ mass labeled internal standards for quantitation. In some cases where these are not available for all the analytes internal standards and surrogates are employed. ICP-MS-based methods use either rare element internal standards or isotope-dilution techniques and limits of quantitation are three times the method detection limit (or 9 standard deviations of the blank intensity). Methods generally employ in batch continuous calibration verification protocols and generally involve measurement of quantifier and qualifier ions. The LOQ are determined for the quantifier ion. Qualifier ions are generally less intense and may not be measurable at or close to the limit of quantitation. Quality assurance approaches are discussed separately.

Table 1. Environmental contaminant analytical panels.

Analytical Panel	Method	Matrices, vol.	LOD, LOQ (range)	Notes
Per and Poly Fluorinated Substances: Short-chain: PFBS, GenX, 4.2 FTS, Long-chain, PFNA, PFOA, PFHxS, PFHpA, PFOS: Surrogates, 4:2 FTSL, PFOAL, PFNAL, PFOSL	UHPLC ESI MS/MS (MRM)	Plasma, Serum, Urine 50 µl	0.05-0.2 ng/ml (0.05-10 ng/ml)	Panel includes multiple legacy and newer/GenX compounds. We continue to add new compounds to this assay and will transition the methods to utilize the TSQ Altis Mass spectrometer.
Phthalates and metabolites: monomethyl phthalate (MMP), monoethyl phthalate (MEP), mono-n-butyl phthalate (MBP), MEHP, mono(2-ethyl-5-hydroxyhexyl) phthalate (MEHHP), mono(2-ethyl-5-oxohexyl) phthalate (MEOHP), monobenzyl phthalate (MBzP) and mono-n-octyl phthalate (MOP),	UHPLC ESI MS/MS (MRM)	Plasma/Serum Urine 50 µl	0.1ng/ml (1-1000 ng/ml)	Assay is validated for MEP and MBP with additional metabolites to be added.
Environmental Phenols 4-t-Octylphenol Benzophenone-3 (Oxybenzone) Bisphenol A (BPA) o-Phenylphenol Parabens Triclocarban Triclosan	UHPLC ESI MS/MS (MRM)	Plasma Serum Urine 50 µl	0.1 ng/ml (urine) (1-1000 ng/ml)	BPA measurements have been used for studies with preclinical models and clinical samples.
Polychlorinated biphenols- selected individual congeners	GC-MS (SIM)	Plasma, Serum, 1ml	LOQ: 0.01-0.05 ng/ml (ECD) LOQ: 0.05-0.1 ng/ml SIM MS (0.05-50ng/ml)	MS method is complementary to ECD method for identification of PCB congeners

University of Kentucky | Center for Appalachian Research in Environmental Sciences

Polychlorinated Biphenyls- Mixture: Includes dioxin like and non-dioxin like congeners, components of commercial Arochlor preparations.	GC-MS/MS (SRM)	Plasma, Serum, 1ml	LOQ: 0.03-0.07pg/ml (non ortho PCBs) LOQ: 1-3 pg/ml (mono ortho PCBs) LOQ:2-7 pg/ml non dioxin like PCBs (LOQ to ~ 1ng/ml)	209 different PCB congeners can be measured using this method based on chromatographic separation and SRM
Polychlorinated Dibenzofurans **PCDFs** 2,3,7,8-TCDF, 1,2,3,7,8-PeCDF 2,3,4,7,8-PeCDF, 1,2,3,4,7,8-HxCDF 1,2,3,6,7,8-HxCDF,2,3,4,6,7,8-HxCDF 1,2,3,7,8,9-HxCDF,1,2,3,4,6,7,8-HpCDF, 1,2,3,4,7,8,9-HpCDF	GC-MS/MS	Plasma, Serum, 1ml	LOQs**:** 0.01-0.05 pg/μl (LOQ to ~ 1 ng/ml)	Method validated for preclinical samples
Polychorinated dibenzo-p-dioxins 2,3,7,8-TCDD, 1,2,3,7,8-PeCDD 1,2,3,4,7,8-HxCDD, 1,2,3,6,7,8-HxCDD 1,2,3,7,8,9-HxCDD,1,2,3,4,6,7,8-HpCDD	GC-MS/MS	Plasma, Serum, 1ml	LOQs :0.2-.5 pg/ml (LOQ-	Method validated for preclinical samples
Flame Retardants **Organophosphorous compounds :** tris (1,3-dichloropropyl) phosphate: TDCPP, triphenyl phoephate (TPP) Bis (1,3-dichloropropyl) phosphate (BDCPP) and diphenyl phosphate (DPP)	UHPLC ESI MS/MS	Plasma, tissues, urine	LOQ: 1 pg/ml	Method validated for measurement of metabolites (BDCPP and DPP) in preclinical studies
Total trace element concentrations	ICP-MS or ICP-OES	Most biological and environmental matrices	LOQs: matrix and element-dependent, as low as 0.1 pg/L	Majority of elements in periodic table can be analyzed
As, Hg, and Se speciation	HPLC ICP-MS	Tissues, environmental samples, blood, urine	LOQs: matrix and element-dependent, as low as 1 ng/L	Validated for environmental and biological samples. Needs work to be validated for clinical studies
Spatial analysis of trace elements	LA-ICP-MS	Solid samples	LOQs: matrix and element-dependent, as low as 0.1 ug/g	Standard reference materials for most biological matrices are not available for this technique. Results are semi-quantitative
Nanoparticle number, size and composition	sp-ICP-MS	Liquid samples and tissue digests	LOQs- limits are element specific. As small as 5 nm diameter and mass concentrations as low as 1 fg/L	Highly experimental technique but gaining widespread use for quantification of nanoparticle exposure

Trihalomethanes and haloacetonitriles: chloroform, bromodichloromethane chlorodibromomethane, bromoform bromomethane, dichloromethane, dichloroacetonitrile, trichloroacetonitrile, bromoacetonitrile, dibromoacetonitrile	GC-dual ECD after liquid/liquid extraction	Drinking water	LOQ: 1 ug/L	Disinfection byproducts- EPA method 551.1[18]. validated for drinking water samples. Simultaneous analysis on dissimilar columns provides confirmation.

Table 2. Biomarkers of tobacco and opiates.

Analytical Panel	Method	Matrices	LOQ (range)	Notes
Nicotine, Cotinine, trans 3 OH cotinine & glucuronide metabolites	HPLC ESI MS/MS	Plasma, serum, urine 0.5-1ml	0.015 ng/ml (0.015-10 ng/ml)	Biomarkers of tobacco smoking and e cigarette use
Tobacco specific Nitrosamines: 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK) and *N*′-nitrosonornicotine (NNN)4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol (NNAL) & glucuronide metabolites	HPLC ESI MS/MS	Urine 1ml	20 pg/ml (20pg/ml-10 ng/ml)	Biomarkers of tobacco smoking
Common Opiates: codeine, fentanyl, morphine, heroin, hydrocodone, hydromorphone, oxycodone, oxymorphone	HPLC ESI MS/MS	Plasma, serum 0.5 ml	0.15-0.4 ng/ml (0.4-150 ng/ml)	Narcotic Analgesics, drugs of abuse

Table 3. Nutritional biomarkers.

Analytical Panel	Method	Matrices	LOQ (range)	Notes
Trimethylamine N-Oxide (TMAO)	HPLC ESI MS/MS	Plasma, Serum	0.05 μM (0.05-200 μM)	Biomarker of red meat/fish, eggs consumption
Odd/Branched Chain Fatty Acids (method also measures common fatty acids)	GC-MS, GC-MS/MS LC-MS/MS	Plasma, Serum 0.2 ml	~1 ppm (1-10,000ppm)	Biomarkers of dairy food and fibers (LCMS method uses chemical derivatization)
Endogenous sterols and plant phytosterols Plant sterols: beta sitosterol, cholestenol campesterol Endogenous cholesterol precursors: desmosterol, lathosterol, cholesterol	GC-MS/MS	Plasma, Serum 0.2 ml		Biomarkers of endogenous cholesterol synthesis and dietary cholesterol intake
Isoprostanes F2-isoprostane (15-F2t-IsoP) and prostaglandin F2 alpha (PGF2 alpha)	HPLC ESI MS/MS	Urine (1ml)	0.05 ng/mL (0.05-10 ng/ml)	Biomarkers of systemic oxidative stress