

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Biochemical analytical plan in children and adults: performing laboratories, reference levels, reporting ranges, clinical guidelines, and critical values.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	NHANES ¹ (µg/L) 2013 - 2014
Children and Adults					
NCEH/Division of Laboratory Sciences* Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS)</i>	Yes	Serum	1 ml (for all PFAS); 1 ml reserve (for future PFAS analyse s)	Age Group (years): 50th to 95th %
	perfluorooctanoic acid (PFOA)‡				3-5: 1.80 – 5.58 6-11: 1.94 – 3.84 12-19: 1.67 – 3.47 20+: 2.07 – 5.60
	n-PFOA - linear isomer				3-5: 1.72 – 5.32 6-11: 1.84 – 3.77 12-19: 1.60 – 3.40 20+: 2.00 – 5.40
	Sb-PFOA - serum branched isomer				3-5: < LOD – 0.280 6-11: < LOD – 0.230 12-19: < LOD – 0.200 20+: < LOD – 0.200
	perfluorooctane sulfonic acid, (PFOS)‡				3-5: 3.41 – 8.82 6-11: 4.02 – 12.4 12-19: 3.60 – 9.30 20+: 5.60 – 19.5
	n-PFOS – linear isomer				3-5: 2.11 – 6.19 6-11: 2.65 – 8.41 12-19: 2.70 – 7.10 20+: 3.70 – 15.1
	Sm-PFOS – serum branched				3-5: 1.00 – 3.60 6-11: 1.41 – 4.25 12-19: 1.00 – 2.30 20+: 1.60 – 5.30

Limit of detection (LOD, see Data Analysis section) for Survey year 13-14 is 0.1. < LOD means less than the limit of detection, which may vary for some chemicals by year and by individual sample.

¹ CDC. 2018. 2013-2014 NHANES 50th to 95th percentiles among children 12-19 years and adults 20+ years old from the Fourth National Report on Human Exposure to Environmental Chemicals, Updated Tables, March 2018. Accessed April 13, 2018 at (https://www.cdc.gov/exposurereport/pdf/FourthReport_UpdatedTables_Volume1_Mar2018.pdf).

‡ See Calculation of PFOS and PFOA as the Sum of Isomers for additional information in March 2018 Updated Tables.

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	NHANES ¹ (µg/L) 2013 - 2014
Children and Adults					
NCEH/Division of Laboratory Sciences* Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS) (continued)</i>	Yes	Serum	1 ml (for all PFAS); 1 ml reserve (for future PFAS analyses)	Age Group (years): 50th to 95th %
	perfluorohexane sulfonic acid (PFHxS)				3-5: 0.740 – 1.62 6-11: 0.850 – 4.14 12-19: 1.10 – 6.30 20+: 1.40 – 5.50
	perfluorooctane sulfonamide (PFOSA)				3-5: < LOD – 0.110 6-11: < LOD - < LOD 12-19: n/a ‡ 20+: n/a ‡
	2-(N-methyl-perfluorooctane sulfonamido) acetic acid (Me-PFOSAA)				3-5: 0.110 – 1.02 6-11: 0.110 – 0.940 12-19: 0.100 – 0.600 20+: < LOD – 0.600
	2-(N-ethyl-perfluorooctane sulfonamido) acetic acid (Et-PFOSAA)				3-5: < LOD - < LOD 6-11: < LOD - < LOD 12-19: n/a ‡ 20+: n/a ‡
	perfluorobutane sulfonic acid (PFBS)				3-5: < LOD - < LOD 6-11: < LOD – 0.130 12-19: < LOD - < LOD 20+: < LOD - < LOD
	perfluoroheptanoic acid (PFHpA)				3-5: < LOD – 0.310 6-11: < LOD – 0.170 12-19: < LOD – 0.200 20+: < LOD – 0.100

Limit of detection (LOD, see Data Analysis section) for Survey year 13-14 is 0.1. < LOD means less than the limit of detection, which may vary for some chemicals by year and by individual sample. ‡ Not measured after Survey Years 2011-2012.

¹ CDC. 2018. 2013-2014 NHANES 50th to 95th percentiles among children 12-19 years and adults 20+ years old from the Fourth National Report on Human Exposure to Environmental Chemicals, Updated Tables, March 2018. Accessed April 13, 2018 at (https://www.cdc.gov/exposurereport/pdf/FourthReport_UpdatedTables_Volume1_Mar2018.pdf).

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	NHANES ¹ (µg/L) 2013 - 2014
Children and Adults					
NCEH/Division of Laboratory Sciences* Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS) (continued)</i>				Age Group (years): 50th to 95th %
	perfluorononanoic acid (PFNA)	Yes	Serum	1 ml (for all PFAS); 1 ml reserve (for future PFAS analyses)	3-5: 0.620 – 3.49 6-11: 0.750 – 3.19 12-19: 0.500 – 2.00 20+: 0.700 – 2.00
	perfluorodecanoic acid (PFDA)				3-5: 0.100 – 0.370 6-11: < LOD – 0.350 12-19: 0.100 – 0.400 20+: 0.193 – 0.800
	perfluoroundecanoic acid (PFUnDA)				3-5: < LOD – 0.370 6-11: < LOD – 0.250 12-19: < LOD – 0.200 20+: < LOD – 0.500
	perfluorododecanoic acid (PFDoA)				3-5: < LOD - < LOD 6-11: < LOD - < LOD 12-19: < LOD – 0.200 20+: < LOD – 0.200
Laboratory and Contact	Proposed Biospecimen Bank for Future Analytes	* CLIA Cert.	Matrix	Volume	NHANES ^{TBD} (µg/L) 20xx – 20xx
Children and Adults					
NCEH/Division of Laboratory Sciences* Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS)</i>				Age Group: 50th to 95th %
	To be determined (TBD) when analytical methods are developed (Including but not limited to the following 18 analytes: PFOA [n-PFOA;, Sb-PFOA], PFOA [n-PFOS, Sm-PFOS], PFHxS, PFBS, PFHpA, PFNA, PFDA, PFUnDA, PFPrS, PFHpS, PFBA, PFPeA, PFHxA, HFPO-DA (GenX), DONA, 9CI-PF3ONS)				3-5: TBD 6-11: TBD 12-19: TBD 20+: TBD
	Creatinine (for urinary creatinine correction; may be contracted)				TBD

Limit of detection (LOD, see Data Analysis section) for Survey year 13-14 is 0.1. < LOD means less than the limit of detection, which may vary for some chemicals by year and by individual sample. ‡ Not measured after Survey Years 2011-2012.

¹ CDC. 2018. 2013-2014 NHANES 50th to 95th percentiles among children 12-19 years and adults 20+ years old from the Fourth National Report on Human Exposure to Environmental Chemicals, Updated Tables, March 2018. Accessed April 13, 2018 at (https://www.cdc.gov/exposurereport/pdf/FourthReport_UpdatedTables_Volume1_Mar2018.pdf).

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	Reportable Range, Guidelines, Critical Values <i>Reference ranges will be updated when commercial lab is selected.</i>
Children and Adults					
Commercial Laboratory (to be determined)* Contact:	<i>Lipids</i>	Yes	Serum	0.5 ml (for all)	
	Total cholesterol, fasting				<u>Coronary Heart Disease Risk (CHD)¹</u> Adult, 18+ years: Desirable: <200 mg/dL Borderline High: 200-239 mg/dL High: ≥240 mg/dL Child, 2-17 years: Acceptable: <170 mg/dL Borderline high: 170-199 mg/dL High: ≥200 mg/dL
	Triglycerides, fasting				<u>CHD Risk¹</u> Adult, 18+ years: Normal: <150 mg/dL Borderline High: 150-199 mg/dL High: 200-499 mg/dL Very High: ≥500 mg/dL Critical Value: >1,000 mg/dL Child, 2-9 years: Acceptable: <75 mg/dL Borderline high: 75-99 mg/dL High: ≥100 mg/dL Child, 10-17 years: Acceptable: <90 mg/dL Borderline high: 90-129 mg/dL High: > or =130 mg/dL
	Low Density Lipoprotein (LDL), fasting				<u>CHD Risk¹</u> Adult, 18+ years:

¹ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/8320>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

					<p>Desirable: <100 mg/dL Above Desirable: 100-129 mg/dL Borderline high: 130-159 mg/dL High: 160-189 mg/dL Very high: ≥190 mg/dL</p> <p>Child, 2-17 years: Acceptable: <110 mg/dL Borderline high: 110-129 mg/dL High: ≥130 mg/dL</p>
	<p>High Density Lipoprotein (HDL), fasting</p>				<p><u>CHD Risk¹</u> Adult, 18+ years: Males: ≥40 mg/dL Females: ≥50 mg/dL</p> <p>Child, 2-17 years: Low: <40 mg/dL Borderline low: 40-45 mg/dL Acceptable: > 45 mg/dL</p>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	Reportable Range, Guidelines, Critical Values <i>Reference ranges will be updated when commercial lab is selected.</i>
Children and Adults					
Commercial Laboratory (to be determined)* Contact:	Uric Acid	Yes	Serum	1 ml	Males ² ≤ 8.0 mg/dL Females ≤ 6.1 mg/dL
	Creatinine (to estimate glomerular filtration rate [eGFR])				Males ³ 1-2 years: 0.1-0.4 mg/dL 3-4 years: 0.1-0.5 mg/dL 5-9 years: 0.2-0.6 mg/dL 10-11 years: 0.3-0.7 mg/dL 12-13 years: 0.4-0.8 mg/dL 14-15 years: 0.5-0.9 mg/dL > or =16 years: 0.8-1.3 mg/dL Reference values have not been established for patients that are <12 months of age. Females 1-3 years: 0.1-0.4 mg/dL 4-5 years: 0.2-0.5 mg/dL 6-8 years: 0.3-0.6 mg/dL 9-15 years: 0.4-0.7 mg/dL > or =16 years: 0.6-1.1 mg/dL Reference values have not been established for patients that are <12 months of age. ESTIMATED GFR >60 mL/min/BSA Note: eGFR results will not be calculated for patients <18 or >70 years old.

² <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/8440>

³ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/8472>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	Reportable Range, Guidelines, Critical Values <i>Reference ranges will be updated when commercial lab is selected.</i>
Children and Adults					
Commercial Laboratory (to be determined)* Contact:	<i>Thyroid Hormones</i>	Yes	Serum	0.5 ml	
	Thyroid Stimulating Hormone (TSH)				0.30-3.0 mIU/L ⁴
	Free Total Thyroxine (Free T4)				0.8-2.0 ng/dL
	Total Thyroxine (TT4)				4.5-12.5 µg/dL
	Total Triiodothyronine (TT3)				80-180 ng/dL
Commercial Laboratory (to be determined)* Contact:	<i>Liver Tests</i>	Yes	Serum	0.5 ml standard tests; 1 ml CK18	
	Alanine transaminase (ALT)				15-65 U/L ⁵
	Aspartate transaminase (AST)				5-40 U/L
	Alkaline phosphatase (ALP)				Female: 50-136 U/L; Male: 40-136 U/L
	Gamma-glutamyltransferase (GGT)				Female 5-55 U/L; Male 5-85 U/L
	Albumin (Alb)				3.4-5.0 g/dL Critical Value: <1.5 g/dL Critical Value: >7.9 g/dL
	Total bilirubin (TBIL)				0.0 - 1.0 mg/dL Critical Value: >12.9 mg/dL
	Direct bilirubin (Conjugated Bilirubin)				0.0-0.3 mg/dL
	<i>Non-alcoholic fatty liver disease (NAFLD)/steatohepatitis</i>				
	Cytokeratin 18 M30 (CK-18 M30) Cytokeratin 18 M65 (CK-18 M65)				No evident liver disease: M30 <200 U/L and M65 <300 U/L TASH: M30<200 U/L and M65 >300 U/L Other liver disease: M30: >200 U/L

⁴ University of Southern California Clinical Laboratories Endocrine Services.

⁵ University of Louisville Department of Medicine, Gastroenterology (updated 14 October 2015).

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	Reportable Range, Guidelines, Critical Values <i>Reference ranges will be updated when commercial lab is selected.</i>														
Children and Adults																			
Commercial Laboratory (to be determined)* Contact:	Sex Hormones	Yes	Serum	0.5 ml	Males ⁶ 4-9 years: <7-20 ng/dL 10-11 years: <7-130 ng/dL 12-13 years: <7-800 ng/dL 14 years: <7-1,200 ng/dL 15-16 years: 100-1,200 ng/dL 17-18 years: 300-1,200 ng/dL ≥19 years: 240-950 ng/dL Females 4-9 years: <7-20 ng/dL 10-11 years: <7-44 ng/dL 12-16 years: <7-75 ng/dL 17-18 years: 20-75 ng/dL ≥19 years: 8-60 ng/dL														
	Testosterone																		
	Estradiol																		
CHILDREN ⁷ Males <table border="0" data-bbox="1522 933 2024 1201"> <thead> <tr> <th>Tanner Stages</th> <th>Reference Range</th> </tr> </thead> <tbody> <tr> <td>Stage I (>14 days and prepubertal)</td> <td><LOD-13 pg/mL</td> </tr> <tr> <td>Stage II</td> <td><LOD-16 pg/mL</td> </tr> <tr> <td>Stage III</td> <td><LOD-26 pg/mL</td> </tr> <tr> <td>Stage IV</td> <td><LOD-38 pg/mL</td> </tr> <tr> <td>Stage V</td> <td>10-40 pg/mL</td> </tr> </tbody> </table> Females <table border="0" data-bbox="1522 1274 2024 1333"> <thead> <tr> <th>Tanner Stages</th> <th>Mean Age</th> </tr> </thead> <tbody> </tbody> </table>						Tanner Stages	Reference Range	Stage I (>14 days and prepubertal)	<LOD-13 pg/mL	Stage II	<LOD-16 pg/mL	Stage III	<LOD-26 pg/mL	Stage IV	<LOD-38 pg/mL	Stage V	10-40 pg/mL	Tanner Stages	Mean Age
Tanner Stages	Reference Range																		
Stage I (>14 days and prepubertal)	<LOD-13 pg/mL																		
Stage II	<LOD-16 pg/mL																		
Stage III	<LOD-26 pg/mL																		
Stage IV	<LOD-38 pg/mL																		
Stage V	10-40 pg/mL																		
Tanner Stages	Mean Age																		

⁶ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/83686>

⁷ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/81816>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

					<p>Stage I (>14 days and prepubertal) 7.1 years</p> <p>Stage II 10.5 years</p> <p>Stage III 11.6 years</p> <p>Stage IV 12.3 years</p> <p>Stage V 14.5 years</p> <p>ADULTS Males: 10-40 pg/mL Females Premenopausal: 15-350 pg/mL ** Postmenopausal: <10 pg/mL **E2 levels vary widely through the menstrual cycle.</p>																								
	Sex hormone-binding globulin (SHBG)				<p>CHILDREN⁸</p> <p>Males</p> <table border="0"> <thead> <tr> <th>Tanner Stages</th> <th>Reference Range</th> </tr> </thead> <tbody> <tr> <td>Stage I</td> <td>31-167 nmol/L</td> </tr> <tr> <td>Stage II</td> <td>49-179 nmol/L</td> </tr> <tr> <td>Stage III</td> <td>5.8-182 nmol/L</td> </tr> <tr> <td>Stage IV</td> <td>14-98 nmol/L</td> </tr> <tr> <td>Stage V</td> <td>10-57 nmol/L</td> </tr> </tbody> </table> <p>Females</p> <table border="0"> <thead> <tr> <th>Tanner Stages</th> <th>Reference Range</th> </tr> </thead> <tbody> <tr> <td>Stage I</td> <td>43-197 nmol/L</td> </tr> <tr> <td>Stage II</td> <td>7.7-119 nmol/L</td> </tr> <tr> <td>Stage III</td> <td>31-191 nmol/L</td> </tr> <tr> <td>Stage IV</td> <td>31-166 nmol/L</td> </tr> <tr> <td>Stage V</td> <td>18-144 nmol/L</td> </tr> </tbody> </table> <p>ADULTS Males: 10-57 nmol/L Females (non-pregnant): 18-144 nmol/L</p>	Tanner Stages	Reference Range	Stage I	31-167 nmol/L	Stage II	49-179 nmol/L	Stage III	5.8-182 nmol/L	Stage IV	14-98 nmol/L	Stage V	10-57 nmol/L	Tanner Stages	Reference Range	Stage I	43-197 nmol/L	Stage II	7.7-119 nmol/L	Stage III	31-191 nmol/L	Stage IV	31-166 nmol/L	Stage V	18-144 nmol/L
Tanner Stages	Reference Range																												
Stage I	31-167 nmol/L																												
Stage II	49-179 nmol/L																												
Stage III	5.8-182 nmol/L																												
Stage IV	14-98 nmol/L																												
Stage V	10-57 nmol/L																												
Tanner Stages	Reference Range																												
Stage I	43-197 nmol/L																												
Stage II	7.7-119 nmol/L																												
Stage III	31-191 nmol/L																												
Stage IV	31-166 nmol/L																												
Stage V	18-144 nmol/L																												

⁸ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/9285>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

	Follicle stimulating hormone (FSH)				<p>Males⁹</p> <p>4-6 years: < or =6.7 IU/L 7-8 years: < or =4.1 IU/L 9-10 years: < or =4.5 IU/L 11 years: 0.4-8.9 IU/L 12 years: 0.5-10.5 IU/L 13 years: 0.7-10.8 IU/L 14 years: 0.5-10.5 IU/L 15 years: 0.4-18.5 IU/L 16 years: < or =9.7 IU/L 17 years: 2.2-12.3 IU/L ≥18 years: 1.0-18.0 IU/L</p> <p>Females</p> <p>15 days-6 years: < or =3.3 IU/L 7-8 years: < or =11.1 IU/L 9-10 years: 0.4-6.9 IU/L 11 years: 0.4-9.0 IU/L 12 years: 1.0-17.2 IU/L 13 years: 1.8-9.9 IU/L 14-16 years: 0.9-12.4 IU/L 17 years: 1.2-9.6 IU/L ≥18 years: Premenopausal Follicular: 3.9-8.8 IU/L Midcycle: 4.5-22.5 IU/L Luteal: 1.8-5.1 IU/L Postmenopausal: 16.7-113.6 IU/L</p>
	Insulin-like growth factor (IGF-1)				

Laboratory and Contact	Analyte	* CLIA Cert.	Matrix	Volume	Reportable Range, Guidelines, Critical Values <i>Reference ranges will be updated when commercial lab is selected.</i>
Children and Adults					
Commercial	<i>Immune Function</i>	Yes	Serum	1 ml	

⁹ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/8670>

Attachment 3. Biochemical Analytical Plan in Children and Adults.

Laboratory (to be determined)* Contact:	Ig A, Ig G, Ig M, Ig E					
Commercial Laboratory (to be determined)* Contact:	<i>Glycemic Parameters</i>	Yes	Whole Blood EDTA	1 ml; plus 1 ml reserve	Diabetes Risk ¹⁰ Normal: <5.7% Increased Risk Diabetes: 5.7-6.4% Diabetes: ≥6.5% (confirmation required)	
	Glycosylated hemoglobin (HbA1c)					
	Glucose, fasting, 8-hour		Serum	0.5 ml Glucose/ Insulin; 1 ml antibodies	<17 µU/ml ⁸	
	Insulin				Negative Antibody: DK≤33 ⁸ Positive Antibody: DK>33	
	Glutamate Decarboxylase -65 (Anti-GAD 65)				Negative Antibody: DK<5 ⁸ Positive Antibody: DK≥5	
Thyrosine Phosphatase-like Protein Autoantibodies (Anti-IA2)						
Children Only						
Commercial Laboratory (to be determined)* Contact:	<i>Antibodies to measles, mumps, rubella, tetanus, and diphtheria</i>	Yes	Serum	1 ml		
Child Total				Serum - 9ml	Whole Blood - 2 ml	Urine - 16 ml
				Red Top 2 x10 ml	EDTA Lavender Top 3 ml	
Adults Only						
Commercial Laboratory (to be determined)* Contact:	<i>Autoimmune Parameters</i>	Yes	Serum	2 ml (for all)	< 15 IU/mL ¹¹	
	Rheumatoid Factor (RF)				< or =1.0 U (negative) ¹²	
	Antinuclear Antibody (ANA) screen				1.1-2.9 U (weakly positive)	
	Antinuclear Antibody (ANA) titer				3.0-5.9 U (positive) > or =6.0 U (strongly positive)	
Adult Total				Serum - 11 ml	Whole Blood - 2 ml	Urine - 16 ml
				Red Top 3 x 10 ml	EDTA Lavender Top 3 ml	

¹⁰ American Diabetes Association. Standards of Medical Care in Diabetes - 2011. Diabetes Care. January 2011;34(Supplement 1):S11-S61 (subject to periodic update).

¹¹ <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/9060>

¹² <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/9026>

Attachment 3. Biochemical Analytical Plan in Children and Adults.