

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 <sup>1</sup> (µg/L) 2013 - 2014
<b>Children and Adults</b>					
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)	<b>Age Group (years): 50<sup>th</sup> to 95<sup>th</sup> %</b>
	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.

<sup>1</sup> CDC. 2018. 2013-2014 NHANES 50<sup>th</sup> to 95<sup>th</sup> 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](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.

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<b>Children and Adults</b>					
<b>NCEH/Division of Laboratory Sciences*</b> Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS) (continued)</i>				<b>Age Group (years): 50<sup>th</sup> to 95<sup>th</sup> %</b>
	perfluorohexane sulfonic acid (PFHxS)	Yes	Serum	1 ml (for all PFAS); 1 ml reserve (for future PFAS analyses)	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.

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<b>Children and Adults</b>					
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)	<b>Age Group (years): 50<sup>th</sup> to 95<sup>th</sup> %</b>
	perfluorononanoic acid (PFNA)				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 <sup>TBD</sup> (µg/L) 20xx – 20xx
<b>Children and Adults</b>					
NCEH/Division of Laboratory Sciences* Contact: Dr. Antonia Calafat	<i>Per- and Poly-fluoroalkyl Substances (PFAS)</i>	Yes	Spot Urine (morning void)	1 ml (for PFAS); 15 ml for creatinin e/or specific gravity)	<b>Age Group: 50<sup>th</sup> to 95<sup>th</sup> %</b>
	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

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<b>Children and Adults</b>					
<b>Commercial Laboratory (to be determined)*</b> Contact:	<i>Lipids</i>	Yes	Serum	0.5 ml (for all)	
	Total cholesterol, fasting				<u>Coronary Heart Disease Risk (CHD)<sup>1</sup></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 Risk<sup>1</sup></u> Adult, 18+ years: Normal: <150 mg/dL Borderline High: 150-199 mg/dL High: 200-499 mg/dL Very High: ≥500 mg/dL  <b>Critical Value: &gt;1,000 mg/dL</b>  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 Risk<sup>1</sup></u> Adult, 18+ years:

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

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					<p>Desirable: &lt;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: &lt;110 mg/dL                  Borderline high: 110-129 mg/dL                  High: ≥130 mg/dL</p>
	<p>High Density Lipoprotein (HDL), fasting</p>				<p><u>CHD Risk<sup>1</sup></u>                  Adult, 18+ years:                  Males: ≥40 mg/dL                  Females: ≥50 mg/dL</p> <p>Child, 2-17 years:                  Low: &lt;40 mg/dL                  Borderline low: 40-45 mg/dL                  Acceptable: &gt; 45 mg/dL</p>

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<b>Children and Adults</b>					
<b>Commercial Laboratory (to be determined)*</b> Contact:	Uric Acid	Yes	Serum	1 ml	Males <sup>2</sup> ≤ 8.0 mg/dL Females ≤ 6.1 mg/dL
	Creatinine (to estimate glomerular filtration rate [eGFR])				Males <sup>3</sup> 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 <b>Note:</b> eGFR results will not be calculated for patients <18 or >70 years old.

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

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

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<b>Children and Adults</b>					
<b>Commercial Laboratory (to be determined)*</b> Contact:	<i>Thyroid Hormones</i>	Yes	Serum	0.5 ml	
	Thyroid Stimulating Hormone (TSH)				0.30-3.0 mIU/L <sup>4</sup>
	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
<b>Commercial Laboratory (to be determined)*</b> Contact:	<i>Liver Tests</i>	Yes	Serum	0.5 ml standard tests; 1 ml CK18	
	Alanine transaminase (ALT)				15-65 U/L <sup>5</sup>
	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 <b>Critical Value: &lt;1.5 g/dL</b> <b>Critical Value: &gt;7.9 g/dL</b>
	Total bilirubin (TBIL)				0.0 - 1.0 mg/dL <b>Critical Value: &gt;12.9 mg/dL</b>
	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

<sup>4</sup> University of Southern California Clinical Laboratories Endocrine Services.

<sup>5</sup> University of Louisville Department of Medicine, Gastroenterology (updated 14 October 2015).

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<b>Children and Adults</b>																			
<b>Commercial Laboratory (to be determined)*</b> Contact:	Sex Hormones	Yes	Serum	0.5 ml	Males <sup>6</sup> 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 <sup>7</sup> Males <table border="0" data-bbox="1522 933 2026 1201"> <thead> <tr> <th>Tanner Stages</th> <th>Reference Range</th> </tr> </thead> <tbody> <tr> <td>Stage I (&gt;14 days and prepubertal)</td> <td>&lt;LOD-13 pg/mL</td> </tr> <tr> <td>Stage II</td> <td>&lt;LOD-16 pg/mL</td> </tr> <tr> <td>Stage III</td> <td>&lt;LOD-26 pg/mL</td> </tr> <tr> <td>Stage IV</td> <td>&lt;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 1299 2026 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
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<sup>6</sup> <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/83686>

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

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					<p>Stage I (&gt;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: &lt;10 pg/mL **E2 levels vary widely through the menstrual cycle.</p>																								
	Sex hormone-binding globulin (SHBG)				<p>CHILDREN<sup>8</sup></p> <p>Males</p> <table border="1"> <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="1"> <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
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<sup>8</sup> <https://www.mayomedicallaboratories.com/test-catalog/Clinical+and+Interpretive/9285>

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	Follicle stimulating hormone (FSH)				<p>Males<sup>9</sup></p> <p>4-6 years: &lt; or =6.7 IU/L            7-8 years: &lt; or =4.1 IU/L            9-10 years: &lt; 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: &lt; 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: &lt; or =3.3 IU/L            7-8 years: &lt; 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>
<b>Children and Adults</b>					
<b>Commercial</b>	Immune Function	Yes	Serum	1 ml	

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

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

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

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

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

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