KRESSER INSTITUTE
FOR FUNCTIONAL AND EVOLUTIONARY MEDICINE

# Impaired Methylation - Part Three

Alright, let's talk about some cases. We'll only do a few because we've already done cases for both B12 and folate deficiency, and those are effectively impaired methylation cases.

This patient is a 41-year-old female with poor digestion, irregular bowel symptoms, and some pain for about two years. She became very anemic earlier this year, which was corrected with an iron supplement, and also developed alopecia areata, or hair loss. She was a vegetarian her whole life and vegan for the last two years prior to coming to see me.

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| Glucose | 87 | 75 - 90 | 65 - 99 |
| Hemoglobin A1c | 5.8 | 4.8 – 5.4 | 4.8 - 5.6 |
| Uric Acid | 4.4 | 3.2 - 5.5 | 2.5 - 7.1 |
| BUN | 12 | 13 – 18 | 6 - 24 |
| Creatinine | 0.92 | 0.85 – 1.1 | 0.57 - 1 |
| BUN/Creatinine Ratio | 13 | 9 – 23 | 9 - 23 |
| Sodium | 141 | 134 – 140 | 134 - 144 |
| Potassium | 4.5 | 4.0 – 4.5 | 3.5 - 5.2 |
| Chloride | 102 | 100 – 106 | 97 - 108 |
| C02 | 23 | 25 – 30 | 18 - 29 |
| Calcium | 9.5 | 9.2 – 10.1 | 8.7 - 10.2 |
| Phosphorus | 4.2 | 3.5 – 4.0 | 2.5 - 4.5 |
| Magnesium | 2.3 | 2.0 – 2.6 | 1.6 - 2.3 |
| Protein, total | 6.9 | 6.9 – 7.4 | 6.0 - 8.5 |
| Albumin | 4.3 | 4.0 – 5.0 | 3.5 - 5.5 |
| Globulin | 2.6 | 2.4 – 2.8 | 1.5 - 4.5 |
| A/G ratio | 1.7 | 1.5 – 2.0 | 1.1 - 2.5 |
| Bilirubin, total | 0.6 | 0.1 – 1.2 | 0.0 - 1.2 |
| Alkaline Phosphatase | 57 | 42 – 107 | 39 - 117 |
| LDH | 142 | 140 - 180 | 119 - 226 |
| AST | 15 | 10 - 30 | 0 - 40 |
| ALT | 11 | 10 - 22 | 0 - 32 |
| GGT | 15 | 0 - 28 | 0 - 60 |
| TIBC | 351 | 250 – 350 | 250 - 450 |
| UIBC | 194 | 150 - 375 | 131 - 425 |
| Iron | 157 | 85 – 135 | 27 - 159 |
| Iron saturation | 45 | 15 – 45 | 15 - 55 |
| Ferritin | 28 | 15 - 120 | 15 - 150 |
| Vitamin B-12 | 222 | 450 – 2000 | 211 - 946 |
| Vitamin D, 25-hydroxy | 9.7 | 35 - 60 | 30.0 - 100.0 |
| Cholesterol, total | 157 | 150 - 250 | 100 - 199 |
| Triglycerides | 59 | 50 – 100 | 0 - 149 |
| HDL | 54 | 55 – 85 | > 39 |
| LDL | 91 | 0 - 175 | 0 - 99 |
| T. Chol / HDL Ratio | 2.9 | < 3 | 0 - 4.4 |
| Triglycerides / HDL Ratio | 1.09 | < 2 | < 3.8 |
| CRP-hs | 0.09 | < 1.0 | 0.00 - 3.00 |
| Homocysteine | 18.2 | < 7.0 | 0.0 - 15.0 |

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| TSH | 1.790 | 0.5 – 2.5 | 0.45 - 4.50 |
| T4, total | 6.8 | 6.0 – 12 | 4.5 - 12 |
| T3 Uptake | 26 | 28 - 35 | 24 - 39 |
| T3, Total | 104 | 100 – 180 | 71 - 180 |
| Copper | 92 | | 72 - 166 |
| Zinc | 65 | | 56 - 134 |
| Zinc / Copper Ratio | 0.71 | > 0.85 | |
| Serum Methylmalonic Acid (MMA) | 645 | 0 - 325 | 0 - 378 |
| WBC | 3.6 | 5.0 – 8.0 | 3.4 - 10.8 |
| RBC | 4.44 | 4.4 – 4.9 | 3.77 - 5.28 |
| Hemoglobin | 12.5 | 13.5 - 14.5 | 11.1 - 15.9 |
| Hematocrit | 38.5 | 37 - 44 | 34 - 46.6 |
| MCV | 87 | 85 – 92 | 79 - 97 |
| MCH | 28.2 | 27.7 – 32.0 | 26.6 - 33.0 |
| MCHC | 32.5 | 32 – 35 | 31.5 - 35.7 |
| RDW | 14.2 | 11.5 – 15.0 | 12.3 - 15.4 |
| Platelets | 261 | 150 – 415 | 150 - 379 |
| Neutrophils | 49 | 40 – 60 | |
| Lymphocytes | 34 | 25 – 40 | |
| Monocytes | 9 | 4.0 – 7.0 | |
| Eosinophils | 7 | 0.0 – 3.0 | |
| Basophils | 1 | 0.0 – 3.0 | |

B12 is almost out of the lab range at 222. Serum MMA is lab-high at 645. Homocysteine was very high at 18.2. Red blood cells were normal, but hemoglobin was 12.5, so this would be B12-deficient functional anemia, and possibly folate too. You can't tell with these labs because we're not seeing FIGLU or serum folate. We weren't running serum folate as part of the case review panel at this point.

| Gene & Variation | rsID | Alleles | Result |
|---|---|---|---|
| COMT V158M | rs4680 | AG | +/- |
| COMT H62H | rs4633 | CT | +/- |
| COMT P199P | rs769224 | GG | -/- |
| VDR Bsm | rs1544410 | CT | +/- |
| VDR Taq | rs731236 | AG | +/- |
| MAO A R297R | rs6323 | GT | +/- |
| ACAT1-02 | rs3741049 | GG | -/- |
| MTHFR C677T | rs1801133 | AA | +/+ |
| MTHFR 03 P39P | rs2066470 | GG | -/- |
| MTHFR A1298C | rs1801131 | TT | -/- |
| MTR A2756G | rs1805087 | AA | -/- |
| MTRR A66G | rs1801394 | AG | +/- |
| MTRR H595Y | rs10380 | CC | -/- |
| MTRR K350A | rs162036 | AA | -/- |
| MTRR R415T | rs2287780 | CT | +/- |
| MTRR A664A | rs1802059 | GG | -/- |
| BHMT-02 | rs567754 | TT | +/+ |
| BHMT-04 | rs617219 | AC | +/- |
| BHMT-08 | rs651852 | CT | +/- |
| AHCY-01 | rs819147 | TT | -/- |
| AHCY-02 | rs819134 | AA | -/- |
| AHCY-19 | rs819171 | TT | -/- |
| CBS C699T | rs234706 | GG | -/- |
| CBS A360A | rs1801181 | AG | +/- |
| CBS N212N | rs2298758 | GG | -/- |
| SHMT1 C1420T | rs1979277 | GG | -/- |

In this case diet was the primary issue, but genetics were also likely playing a role. She was homozygous for C677T, as you can see, and then heterozygous for many other SNPs that can affect methylation such as COMT, BHMT, and MTRR.

**0091 Organix® Comprehensive Profile - Urine**

*Methodology: LC/Tandem Mass Spectroscopy, Colorimetric*

This report is not intended for the diagnosis of neonatal inborn errors of metabolism.
Ranges are for ages 13 and over

| | Results (mg/mg creatinine) | | Quintile Ranking (1st 2nd 3rd 4th 5th) | 95% Reference Range |
|---|---|---|---|---|
| **B-Complex Vitamin Markers** *(B1, B2, B3, B5, B6, Biotin)* | | | | |
| 15. a-Ketoisovalerate | <DL* | | 0.25 | <= 0.49 |
| 16. a-Ketoisocaproate | 0.35 | H | 0.34 | <= 0.52 |
| 17. a-Keto-ß-methylvalerate | 0.66 | H | 0.38 | <= 1.10 |
| 18. Xanthurenate | 0.18 | | 0.34 | <= 0.46 |
| 19. ß-Hydroxyisovalerate | 13.3 | H | 7.6 | <= 11.5 |
| **Methylation Cofactor Markers** *(B12, Folate)* | | | | |
| 20. Methylmalonate | 0.6 | | 1.7 | <= 2.3 |
| 21. Formiminoglutamate | 2.7 | H | 1.2 | <= 2.2 |
| **Cell Regulation Markers** | | | | |
| **Neurotransmitter Metabolism Markers** *(Tyrosine, Tryptophan, B6, antioxidants)* | | | | |
| 22. Vanilmandelate | 2.2 | | 1.6 3.9 | 1.2-5.3 |
| 23. Homovanillate | 1.6 | L | 1.9 5.7 | 1.4-7.6 |
| 24. 5-Hydroxyindoleacetate | 2.7 | | 2.1 5.6 | 1.6-9.8 |
| 25. Kynurenate | 0.7 | | 1.0 | <= 1.5 |
| 26. Quinolinate | 1.9 | | 4.0 | <= 5.8 |
| 27. Picolinate | 3.7 | | 8.0 | 2.8-13.5 |
| **Oxidative Damage and Antioxidant Markers** *(Vitamin C and other antioxidants)* | | | | |
| 28. p-Hydroxyphenyllactate | 0.04 | | 0.39 | <= 0.66 |
| 29. 8-Hydroxy-2-deoxyguanosine | 6.2 | H | 5.3 | <= 7.6 |

Her FIGLU was quite high here, it's 2.7, and the upper end of the range is 2.2. So she is not only B12 deficient but also folate deficient. The vegan diet was contributing to both folate deficiency and B12 deficiency, and that was accentuated by her homozygosity for MTHFR C667T.

The next patient is a 25-year-old female with chief complaint of digestive problems, brain fog, and food intolerances.

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| Glucose | 92 | 75 – 90 | 65 - 99 |
| Hemoglobin A1c | 5.1 | 4.4 – 5.4 | 4.8 - 5.6 |
| Uric Acid | 4.9 | 3.2 - 5.5 | 2.5 - 7.1 |
| BUN | 11 | 13 – 18 | 6 - 20 |
| Creatinine | 0.92 | 0.85 – 1.1 | 0.57 - 1 |
| Sodium | 138 | 135 – 140 | 134 - 144 |
| Potassium | 4.4 | 4.0 – 4.5 | 3.5 - 5.2 |
| Chloride | 102 | 100 – 106 | 97 - 108 |
| C02 | 22 | 25 – 30 | 18 - 29 |
| Calcium | 9.7 | 9.2 – 10.1 | 8.7 - 10.2 |
| Phosphorus | 3.8 | 3.5 – 4.0 | 2.5 - 4.5 |
| Magnesium | 2.2 | 2.0 – 2.6 | 1.6 - 2.6 |
| Protein, total | 6.9 | 6.9 – 7.4 | 6.0 - 8.5 |
| Albumin | 4.5 | 4.0 – 5.0 | 3.5 - 5.5 |
| Globulin | 2.4 | 2.4 – 2.8 | 1.5 - 4.5 |
| A/G ratio | 1.9 | 1.5 – 2.0 | 1.1 - 2.5 |
| Bilirubin, total | 0.7 | 0.1 – 1.2 | 0.0 - 1.2 |
| Alkaline Phosphatase | 46 | 42 – 107 | 39 - 117 |
| LDH | 109 | 140 - 180 | 119 - 226 |
| AST | 15 | 10 - 30 | 0 - 40 |
| ALT | 9 | 10 - 22 | 0 - 32 |
| GGT | 14 | 0 - 28 | 0 - 60 |
| TIBC | 330 | 250 – 350 | 250 - 450 |
| UIBC | 141 | 150 - 375 | 150 - 375 |
| Iron | 189 | 85 – 135 | 35 - 155 |
| Iron saturation | 57 | 15 – 45 | 15 - 55 |
| Ferritin | 91 | 15 - 120 | 15 - 150 |
| Cholesterol, total | 212 | 150 – 250 | 100 - 189 |
| Triglycerides | 76 | 50 – 100 | 0 - 149 |
| HDL | 87 | 55 – 85 | > 39 |
| LDL | 110 | 0 – 175 | 0 - 119 |
| T. Chol / HDL Ratio | 2.4 | < 3 | 0 - 4.4 |
| Triglycerides / HDL Ratio | 0.87 | < 2 | < 3.8 |
| TSH | 2.330 | 0.5 – 2.5 | 0.45 - 4.50 |
| T4, total | 9.2 | 6.0 – 12 | 4.5 - 12.0 |
| T3 Uptake | 31 | 28 - 35 | 24 - 39 |
| T3, Total | 102 | 100 – 180 | 71 - 180 |
| Vitamin D, 25-hydroxy | 34.2 | 35 - 60 | 30.0 - 100.0 |

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| WBC | 5.2 | 5.0 – 8.0 | 3.4 - 10.8 |
| RBC | 4.43 | 4.4 – 4.9 | 3.77 - 5.28 |
| Hemoglobin | 13.8 | 13.5 - 14.5 | 11.1 - 15.9 |
| Hematocrit | 41.7 | 37 - 44 | 34.0 - 46.6 |
| MCV | 94 | 85 – 92 | 79 - 97 |
| MCH | 31.2 | 27.7 – 32.0 | 26.6 - 33.0 |
| MCHC | 33.1 | 32 – 35 | 31.5 - 35.7 |
| RDW | 12.3 | 11.5 – 15.0 | 12.3 - 15.4 |
| Platelets | 215 | 150 – 415 | 150 - 379 |
| Neutrophils | 50 | 40 – 60 | |
| Lymphocytes | 41 | 25 – 40 | |
| Monocytes | 7 | 4.0 – 7.0 | |
| Eosinophils | 1 | 0.0 – 3.0 | |
| Basophils | 1 | 0.0 – 3.0 | |
| **Additional Tests:** | | | |
| CRP-hs | 1 | < 1.0 | 0.00 - 3.00 |
| Homocysteine | 10.8 | < 9.0 | 0.0 - 15.0 |
| Vitamin B-12 | 244 | 450 – 2000 | 211 - 946 |
| Copper | 133 | | 72 - 166 |
| Zinc | 114 | | 56 - 134 |
| Zinc / Copper Ratio | 0.86 | > 0.85 | |
| Serum Methylmalonic Acid (MMA) | 72 | 0 - 325 | 0 - 378 |

Serum B12 was almost out of the lab range at 244. Homocysteine is functionally high at almost 11. Red blood cell indices were normal except for MCV, which is a little bit borderline high at 94. Note that this patient also had iron overload with iron saturation of 57 percent, UIBC of 141, and serum iron of 189.

| Gene & Variation | rsID | Alleles | Result |
|---|---|---|---|
| COMT V158M | rs4680 | AG | +/- |
| COMT H62H | rs4633 | CT | +/- |
| COMT P199P | rs769224 | GG | -/- |
| VDR Bsm | rs1544410 | CT | +/- |
| VDR Taq | rs731236 | AG | +/- |
| MAO A R297R | rs6323 | GT | +/- |
| ACAT1-02 | rs3741049 | GG | -/- |
| MTHFR C677T | rs1801133 | AA | +/+ |
| MTHFR 03 P39P | rs2066470 | GG | -/- |
| MTHFR A1298C | rs1801131 | TT | -/- |
| MTR A2756G | rs1805087 | AA | -/- |
| MTRR A66G | rs1801394 | AG | +/- |
| MTRR H595Y | rs10380 | CC | -/- |
| MTRR K350A | rs162036 | AA | -/- |
| MTRR R415T | rs2287780 | CT | +/- |
| MTRR A664A | rs1802059 | GG | -/- |
| BHMT-02 | rs567754 | TT | +/+ |
| BHMT-04 | rs617219 | AC | +/- |
| BHMT-08 | rs651852 | CT | +/- |
| AHCY-01 | rs819147 | TT | -/- |
| AHCY-02 | rs819134 | AA | -/- |
| AHCY-19 | rs819171 | TT | -/- |
| CBS C699T | rs234706 | GG | -/- |
| CBS A360A | rs1801181 | AG | +/- |
| CBS N212N | rs2298758 | GG | -/- |
| SHMT1 C1420T | rs1979277 | GG | -/- |

At this point, I was suspicious of B12 deficiency given the low serum cobalamin levels and high homocysteine, but we still didn't know about folate because we didn't have serum or red blood cell folate level for her. We did have a genetic panel, and she was homozygous for C677T.

**0091 Organix® Comprehensive Profile - Urine**

*Methodology: LC/Tandem Mass Spectroscopy, Colorimetric*

This report is not intended for the diagnosis of neonatal inborn errors of metabolism.

Ranges are for ages 13 and over

| | Results | | Quintile Ranking (1st 2nd 3rd 4th 5th) | 95% Reference Range |
|---|---|---|---|---|
| | mg/mg creatinine | | | |
| **B-Complex Vitamin Markers** *(B1, B2, B3, B5, B6, Biotin)* | | | | |
| 15. a-Ketoisovalerate | <DL* | | 0.25 | <= 0.49 |
| 16. a-Ketoisocaproate | 0.27 | | 0.34 | <= 0.52 |
| 17. a-Keto-ß-methylvalerate | 1.92 | H | 0.38 | <= 1.10 |
| 18. Xanthurenate | 0.64 | H | 0.34 | <= 0.46 |
| 19. 5-Hydroxyisovalerate | 8.1 | H | 7.6 | <= 11.5 |
| **Methylation Cofactor Markers** *(B12, Folate)* | | | | |
| 20. Methylmalonate | 1.3 | | 1.7 | <= 2.3 |
| 21. Formiminoglutamate | 2.3 | H | 1.2 | <= 2.2 |
| **Cell Regulation Markers** | | | | |
| **Neurotransmitter Metabolism Markers** *(Tyrosine, Tryptophan, B6, antioxidants)* | | | | |
| 22. Vanillmandelate | 1.8 | | 1.6 3.9 | 1.2-5.3 |
| 23. Homovanillate | 2.3 | | 1.9 5.7 | 1.4-7.6 |
| 24. 5-Hydroxyindoleacetate | 3.7 | | 2.1 5.6 | 1.6-9.8 |
| 25. Kynurenate | 1.7 | H | 1.0 | <= 1.5 |
| 26. Quinolinate | 2.1 | | 4.0 | <= 5.8 |
| 27. Picolinate | 4.7 | | 8.0 | 2.8-13.5 |
| **Oxidative Damage and Antioxidant Markers** *(Vitamin C and other antioxidants)* | | | | |
| 28. p-Hydroxyphenyllactate | 0.22 | | 0.39 | <= 0.66 |
| 29. 8-Hydroxy-2-deoxyguanosine | 2.6 | | 5.3 | <= 7.6 |

(Units for 8-hydroxy-2-deoxyguanosine are ng/mg creatinine)

Given that, I suspected folate deficiency as well as B12 deficiency, and this is where the urine organic acids panel can come in handy. Her FIGLU was 2.3, which is elevated, indicating folate deficiency. Interestingly, her urine MMA was normal at 1.3, though it was close to the 1.5 cut-off point suggested, so this again highlights the importance of using different markers.

**INFLAMMATION**

| | Within | Outside | Reference Range | |
|---|---|---|---|---|
| Lactoferrin | 3.6 | | < 7.3 | µg/mL |
| Calprotectin* | 23 | | <= 50 | µg/g |
| Lysozyme* | | 2190 | <= 600 | ng/mL |
| White Blood Cells | None | | None - Rare | |
| Mucus | Neg | | Neg | |

**Lactoferrin** and **Calprotectin** are reliable markers for differentiating organic inflammation (IBD) from function symptoms (IBS) and for management of IBD. Monitoring levels of fecal lactoferrin and calprotectin can play an essential role in determining the effectiveness of therapy, are good predictors of IBD remission, and can indicate a low risk of relapse. **Lysozyme*** is an enzyme secreted at the site of inflammation in the GI tract and elevated levels have been identified in IBD patients. **White Blood Cells** (WBC) and **Mucus** in the stool can occur with bacterial and parasitic infections, with mucosal irritation, and inflammatory bowel diseases such as Crohn's disease or ulcerative colitis.

**IMMUNOLOGY**

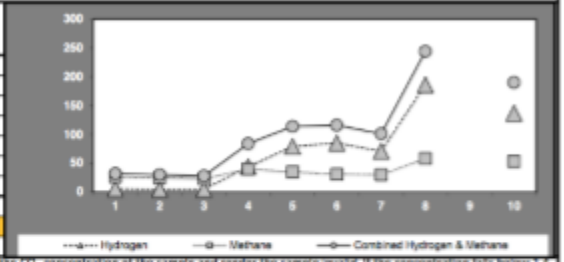
| | Within | Outside | Reference Range |
|---|---|---|---|
| Secretory IgA* | | 888 | 51 - 204 mg/dL |

**Secretory IgA*** (sIgA) is secreted by mucosal tissue and represents the first line of defense of the GI mucosa and is central to the normal function of the GI tract as an immune barrier. Elevated levels of sIgA have been associated with an upregulated immune response.

**Data**

$H_2$ = Hydrogen $CH_4$ = Methane $CO_2$ = Measured

| | Sample | ppm $H_2$ | ppm $CH_4$ | Total H2 + CH4 | $CO_2$ %* |
|---|---|---|---|---|---|
| 1 | Baseline | 6 | 26 | 32 | OK |
| 2 | 20 min | 5 | 25 | 30 | OK |
| 3 | 40 min | 5 | 23 | 28 | OK |
| 4 | 60 min | 44 | 40 | 84 | OK |
| 5 | 80 min | 79 | 35 | 114 | OK |
| 6 | 100 min | 85 | 31 | 116 | OK |
| 7 | 120 min | 71 | 30 | 101 | OK |
| 8 | 140 min | 186 | 58 | 244 | OK |
| 9 | 160 min | | | | QNS |
| 10 | 180 min | 137 | 53 | 190 | OK |




*Dilutions of the sample may occur during the sampling procedure which may decrease the $CO_2$ concentration of the sample and render the sample invalid. If the concentration falls below 2.5 %, the entry for $CO_2$ will be marked as Quantity Not Sufficient (QNS) and the entries for $H_2$ and $CH_4$ will be highlighted. If the sample is otherwise unusable the entry for $CO_2$ will be marked as Not Available (N/A) and the entries for $H_2$ and $CH_4$ will be highlighted. See notes section for details if cells are highlighted and blank or highlighted and contain N/A or QNS.

**Analysis**

| | | | |
|---|---|---|---|
| **Combined baseline total =** | 32 | H | ≤20ppm |
| **Greatest $H_2$ increase over the lowest preceding value within first 120 minutes =** | 80 | H | ≤20ppm |
| **Greatest $CH_4$ increase over the lowest preceding value within first 120 minutes =** | 17 | H | ≤12ppm |
| **Greatest combined $H_2$ & $CH_4$ increase over the lowest preceding value within first 120 minutes =** | 88 | H | ≤15ppm |

**Interpretation**

| | | |
|---|---|---|
| **SIBO Suspected - Elevated Hydrogen** | Increases of hydrogen greater than 20ppm over the lowest preceding value within the first 120 minutes (+/- 5min deviation) are indicative of bacterial overgrowth. | **POSITIVE** |
| **SIBO Suspected - Elevated Methane** | Increases of methane greater than 12ppm over the lowest preceding value within the first 120 minutes (+/- 5min deviation) are indicative of bacterial overgrowth. | **POSITIVE** |
| **SIBO Suspected - Elevated Combined Hydrogen & Methane Gasses** | Increases in combined hydrogen and methane gas values greater than 15ppm over the lowest preceding value are indicative of bacterial overgrowth. | **POSITIVE** |

She had significant gut inflammation as well as SIBO, so in this case, it was a combination of genetics and environmental factors. She was homozygous for C677T, but had GI malabsorption as well, and that probably sent her over the edge and was likely also causing poor absorption of whatever B12 she was consuming in the diet.

| | | | |
|---|---|---|---|
| Applicant | Kresser | | |
| Appoint. date | 5/25/16 | | |
| Appoint. time | 01:45 PM | | |
| Appoint. No. | 210561 | | |
| | | *Unit* | *Ref. Range* |
| DERIVATES | | | |
| S-Adenosylmethionine (RBC) | **219** | µmol/dl | 221 - 256 |
| S-Adenosylhomocysteine (RBC) | **47.0** | µmol/dl | 38.0 - 49.0 |
| FOLIC ACID DERIVATES | | | |
| 5-CH3-THF | **7.2** | nmol/l | 8.4 - 72.6 |
| 10-Formyl-THF | **1.3** | nmol/l | 1.5 - 8.2 |
| 5-Formyl-THF | **1.10** | nmol/l | 1.20 - 11.70 |
| THF | **0.53** | nmol/l | 0.60 - 6.80 |
| Folic Acid | **7.7** | nmol/l | 8.9 - 24.6 |
| Folinic Acid (WB) | **6.8** | nmol/l | 9.0 - 35.5 |
| Active folate (RBC) | **311** | nmol/l | 400 - 1500 |
| NUCLEOSIDE | | | |
| Adenosine | **20.3** | 10^-8 M | 16.8 - 21.4 |
| AMINOACIDS IN PLASMA | | | |
| Glutathione (oxidised) | **0.46** | µmol/L | 0.16 - 0.50 |
| Glutathione (reduced) | **3.3** | µmol/L | 3.8 - 5.5 |

Let's briefly discuss follow-up testing for impaired methylation. The panel I use most is called the "Methylation Pathways Panel" from Health Diagnostics and Research Institute, or HDRI. It is the only lab that I'm aware of that tests for the many distinct folate derivatives in the methylation cycle such as 5-MTHF, 10-formyltetrahydrofolate, 5-formyltetrahydrofolate, which is also called folinic acid, tetrahydrofolate, the most reduced form of folate, folic acid, white blood cell folinic acid, and then active folate inside of the red blood cell. It is also the only lab that tests for both oxidized and reduced glutathione, which is important because oxidized glutathione is not beneficial. It's been oxidized and can no longer perform its beneficial function. The ratio of oxidized to reduced glutathione is an important indicator of oxidative stress and methylation capacity.

HDRI tests also the levels of the major methyl donor, SAMe, S-adenosylmethionine, as well as levels of S-adenosylhomocysteine, which is an inhibitor of methylation. It tests for adenosine. If adenosine is elevated, it suggests a problem with the enzyme deaminase, which breaks down adenosine, and B6 and zinc are cofactors for that enzyme.

Not surprisingly, given this particular patient's B12 and folate deficiency, which we saw on the previous slides, there are several markers that are off on her HDRI panel. I'm not going to go into detail here on how to interpret this panel and treat based on it, but I will cover this in a future course.

The next patient is a 65-year-old female with chronic insomnia and chronic hamstring injuries from running.

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| Glucose | 95 | 75 – 90 | 65 - 99 |
| Hemoglobin A1c | 6.0 | 4.4 – 5.4 | 4.8 - 5.6 |
| Uric Acid | 5.8 | 3.2 - 5.5 | 2.5 - 7.1 |
| BUN | 23 | 13 – 18 | 8 - 27 |
| Creatinine | 1.01 | 0.85 – 1.1 | 0.57 - 1 |
| BUN/Creatinine Ratio | 23 | 9 – 23 | 9 - 23 |
| Sodium | 141 | 135 – 140 | 134 - 144 |
| Potassium | 4.4 | 4.0 – 4.5 | 3.5 - 5.2 |
| Chloride | 100 | 100 – 106 | 97 - 108 |
| C02 | 25 | 25 – 30 | 18 - 29 |
| Calcium | 9.5 | 9.2 – 10.1 | 8.7 - 10.3 |
| Phosphorus | 4.0 | 3.5 – 4.0 | 2.5 - 4.5 |
| Magnesium | 2.0 | 2.0 – 2.6 | 1.6 - 2.6 |
| Protein, total | 6.9 | 6.9 – 7.4 | 6.0 - 8.5 |
| Albumin | 4.3 | 4.0 – 5.0 | 3.6 - 4.8 |
| Globulin | 2.6 | 2.4 – 2.8 | 1.5 - 4.5 |
| A/G ratio | 1.7 | 1.5 – 2.0 | 1.1 - 2.5 |
| Bilirubin, total | 0.8 | 0.1 – 1.2 | 0.0 - 1.2 |
| Alkaline Phosphatase | 48 | 42 – 107 | 39 - 117 |
| LDH | 182 | 140 - 180 | 119 - 226 |
| AST | 29 | 10 - 30 | 0 - 40 |
| ALT | 19 | 10 - 22 | 0 - 32 |
| GGT | 43 | 0 - 28 | 0 - 60 |
| TIBC | 339 | 250 – 350 | 250 - 450 |
| UIBC | 191 | 150 - 375 | 150 - 375 |
| Iron | 148 | 85 – 135 | 35 - 155 |
| Iron saturation | 44 | 15 – 45 | 15 - 55 |
| Ferritin | 44 | MW: 30 - 150 | 15 - 150 |
| Cholesterol, total | 217 | 150 – 250 | 100 - 199 |
| Triglycerides | 59 | 50 – 100 | 0 - 149 |
| HDL | 105 | 55 – 85 | > 39 |
| LDL | 100 | 0 – 175 | 0 - 99 |
| T. Chol / HDL Ratio | 2.1 | < 3 | 0 - 4.4 |
| Triglycerides / HDL Ratio | 0.56 | < 2 | < 3.8 |
| TSH | 3.000 | 0.5 – 2.5 | 0.450 - 4.500 |
| T4, total | 4.7 | 6.0 – 12 | 4.5 - 12.0 |
| T3 Uptake | 41 | 28 - 35 | 24 - 39 |
| T3, Total | 68 | 100 – 180 | 71 - 180 |
| Vitamin D, 25-hydroxy | 30.5 | 35 - 60 | 30.0 - 100.0 |

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| WBC | 4.2 | 5.0 – 8.0 | 3.4 - 10.8 |
| RBC | 4.60 | 4.4 – 4.9 | 3.77 - 5.28 |
| Hemoglobin | 13.7 | 13.5 - 14.5 | 11.1 - 15.9 |
| Hematocrit | 41.1 | 37 - 44 | 34.0 - 46.6 |
| MCV | 89 | 85 – 92 | 79 - 97 |
| MCH | 29.8 | 27.7 – 32.0 | 26.6 - 33.0 |
| MCHC | 33.3 | 32 – 35 | 31.5 - 35.7 |
| RDW | 14.0 | 11.5 – 15.0 | 12.3 - 15.4 |
| Platelets | 180 | 150 – 415 | 150 - 379 |
| Neutrophils | 49 | 40 – 60 | |
| Lymphocytes | 40 | 25 – 40 | |
| Monocytes | 7 | 4.0 – 7.0 | |
| Eosinophils | 3 | 0.0 – 3.0 | |
| Basophils | 1 | 0.0 – 3.0 | |
| **Additional Tests:** | | | |
| CRP-hs | 0.83 | < 1.0 | 0.00 - 3.00 |
| Homocysteine | 17.8 | < 7.0 | 0.0 - 15.0 |
| Vitamin B-12 | 363 | 450 – 2000 | 211 - 946 |
| Copper | 97 | | 72 - 166 |
| Zinc | 100 | | 56 - 134 |
| Zinc / Copper Ratio | 1.03 | > 0.85 | |
| Serum Methylmalonic Acid (MMA) | 359 | 0 - 325 | 0 - 378 |
| eGFR If NonAfrican Am | 59 | | > 59 |

Her serum B12 is 363, which is below the 406 cut-off defined for optimal function. Serum MMA is 359, above the 300 cut-off. Homocysteine is 17.8, which is above the lab range.

In terms of underlying causes, she had SIBO, fungal overgrowth, dysbiosis, and Cryptosporidium. In her case, it was likely GI malabsorption was leading to low B12 levels. Also, she had low levels of vitamin D, glutathione, and iodine in other testing.

| Gene & Variation | rsID | Alleles | Result |
|---|---|---|---|
| COMT V158M | rs4680 | AA | +/+ |
| COMT H62H | rs4633 | TT | +/+ |
| COMT P199P | rs769224 | GG | -/- |
| VDR Bsm | rs1544410 | CC | -/- |
| VDR Taq | rs731236 | AA | +/+ |
| MAO-A R297R | rs6323 | GG | -/- |
| ACAT1-02 | rs3741049 | GG | -/- |
| MTHFR C677T | rs1801133 | GG | -/- |
| MTHFR 03 P39P | rs2066470 | GG | -/- |
| MTHFR A1298C | rs1801131 | TT | -/- |
| MTR A2756G | rs1805087 | AA | -/- |
| MTRR A66G | rs1801394 | AA | -/- |
| MTRR H595Y | rs10380 | __ | no call |
| MTRR K350A | rs162036 | AA | -/- |
| MTRR R415T | rs2287780 | __ | no call |
| MTRR A664A | rs1802059 | AG | +/- |
| BHMT-02 | rs567754 | CC | -/- |
| BHMT-04 | rs617219 | __ | no call |
| BHMT-08 | rs651852 | CT | +/- |
| AHCY-01 | rs819147 | TT | -/- |
| AHCY-02 | rs819134 | __ | no call |
| AHCY-19 | rs819171 | TT | -/- |
| CBS C699T | rs234706 | AA | +/+ |
| CBS A360A | rs1801181 | GG | -/- |
| CBS N212N | rs2298758 | __ | no call |
| SHMT1 C1420T | rs1979277 | __ | no call |

She didn't have any MTHFR mutations, but did have mutations in COMT and MAO. These would not necessarily be expected to affect folate metabolism. They have more of an effect on catecholamine recycling, tyramine breakdown, and serotonin levels.

| | | Unit | Ref. Range |
|---|---|---|---|
| MISCELLANEOUS | | | |
| NO (Nitric oxide) | 51.6 | ng/mL | 18.0 - 35.0 |
| ENZYMES | | | |
| Catalase | 128.0 | U/m/mgHb | 130.0 - 188.0 |
| DERIVATES | | | |
| S-Adenosylmethionine (RBC) | 219 | µmol/dl | 221 - 256 |
| S-Adenosylhomocysteine (RBC) | 56.3 | µmol/dl | 38.0 - 49.0 |
| FOLIC ACID DERIVATES | | | |
| 5-CH3-THF | 7.9 | nmol/l | 8.4 - 72.6 |
| 10-Formyl-THF | 6.4 | nmol/l | 1.5 - 8.2 |
| 5-Formyl-THF | 1.10 | nmol/l | 1.20 - 11.70 |
| THF | 0.47 | nmol/l | 0.60 - 6.80 |
| Folic Acid | 10.2 | nmol/l | 8.9 - 24.6 |
| Folinic Acid (WB) | 7.0 | nmol/l | 9.0 - 35.5 |
| Active folate (RBC) | 327 | nmol/l | 400 - 1500 |
| NUCLEOSIDE | | | |
| Adenosine | 25.7 | 10^-8 M | 16.8 - 21.4 |
| AMINOACIDS IN PLASMA | | | |
| Nitrotyrosine | 10.9 | µg/l | 1.1 - 6.8 |
| Glutathione (oxidised) | 0.54 | µmol/L | 0.16 - 0.50 |
| Glutathione (reduced) | 2.8 | µmol/L | 3.8 - 5.5 |

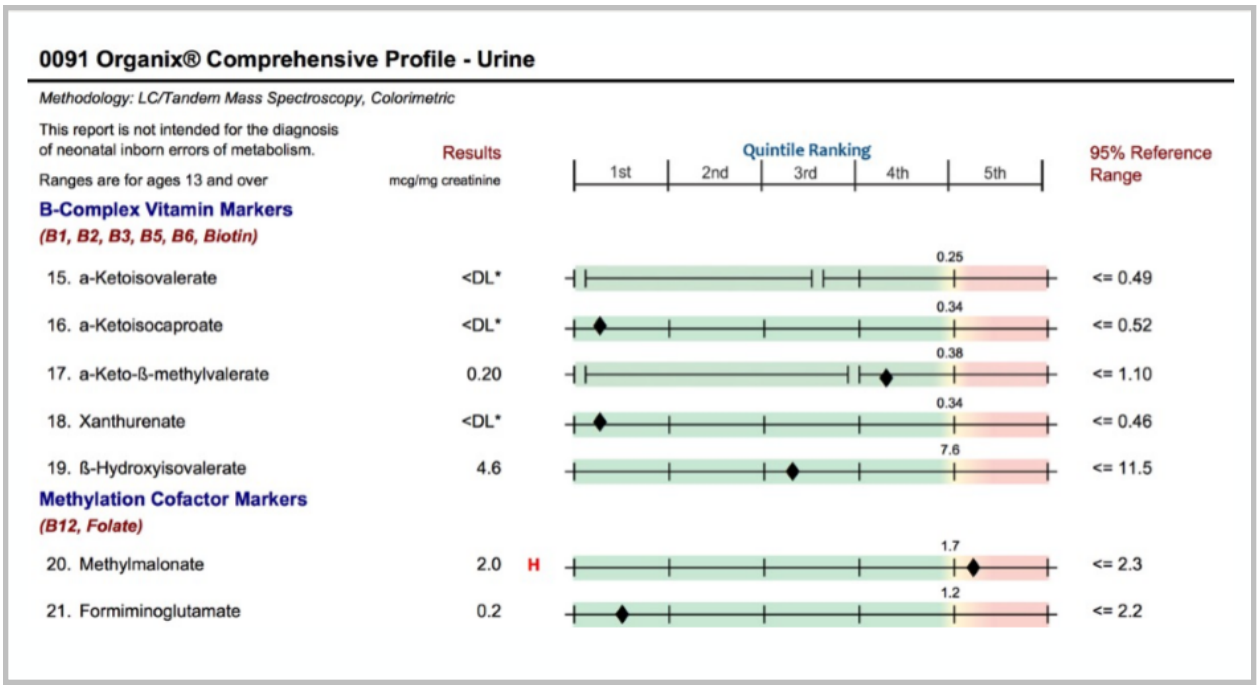
However, her HDRI panel suggested very significant issues with methylation. All but two markers were out of range, as you can see here. Even folic acid was at the very low end of the range. This is a good example of how genetics don't always predict or reflect what is happening functionally, and it is why I insist on assessing functional methylation status instead of just treating people based on their genetics. You'll see this a lot, both patients who are self-treating based on their genes, and clinicians who are treating with supplement protocols on the basis of genetics alone, and I really don't think this is a good idea. There is still too much we don't understand about the genes, and as I've said numerous times and hopefully have shown here, environment is much more important. This is an expanded panel from HDRI, which also includes nitrotyrosine, catalase, and nitric oxide, which are all markers of oxidative stress.

The next patient is a 36-year-old female with PMS, anxiety, nausea, dizziness, irritability, and low blood pressure. She only weighed about 100 pounds. She had gastritis, which should be a clue for B12 deficiency, and was hypothyroid since about 20 years old. She was diagnosed with Hashimoto's in 2013 with a TSH of over 1,000 and was diagnosed with SIBO in 2014, so again, you should be thinking about potential absorption issues.

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| Glucose | 79 | 75 – 90 | 65 - 99 |
| Hemoglobin A1c | 5.3 | 4.4 – 5.4 | 4.8 - 5.6 |
| Uric Acid | 3.1 | 3.2 - 5.5 | 2.5 - 7.1 |
| BUN | 20 | 13 – 18 | 6 - 20 |
| Creatinine | 0.80 | 0.85 – 1.1 | 0.57 - 1 |
| Sodium | 140 | 135 – 140 | 134 - 144 |
| Potassium | 4.5 | 4.0 – 4.5 | 3.5 - 5.2 |
| Chloride | 101 | 100 – 106 | 97 - 108 |
| C02 | 26 | 25 – 30 | 18 - 29 |
| Calcium | 9.4 | 9.2 – 10.1 | 8.7 - 10.2 |
| Phosphorus | 3.7 | 3.5 – 4.0 | 2.5 - 4.5 |
| Magnesium | 2.0 | 2.0 – 2.6 | 1.6 - 2.6 |
| Protein, total | 6.7 | 6.9 – 7.4 | 6.0 - 8.5 |
| Albumin | 4.9 | 4.0 – 5.0 | 3.5 - 5.5 |
| Globulin | 1.8 | 2.4 – 2.8 | 1.5 - 4.5 |
| A/G ratio | 2.7 | 1.5 – 2.0 | 1.1 - 2.5 |
| Bilirubin, total | 0.7 | 0.1 – 1.2 | 0.0 - 1.2 |
| Alkaline Phosphatase | 51 | 42 – 107 | 39 - 117 |
| LDH | 126 | 140 - 180 | 119 - 226 |
| AST | 16 | 10 - 30 | 0 - 40 |
| ALT | 11 | 10 - 22 | 0 - 32 |
| GGT | 12 | 0 - 28 | 0 - 60 |
| TIBC | 277 | 250 – 350 | 250 - 450 |
| UIBC | 157 | 150 - 375 | 150 - 375 |
| Iron | 120 | 85 – 135 | 35 - 155 |
| Iron saturation | 43 | 15 – 45 | 15 - 55 |
| Ferritin | 109 | 15 - 120 | 15 - 150 |
| Cholesterol, total | 165 | 150 – 250 | 100 - 199 |
| Triglycerides | 56 | 50 – 100 | 0 - 149 |
| HDL | 59 | 55 – 85 | > 39 |
| LDL | 95 | 0 – 175 | 0 - 99 |
| T. Chol / HDL Ratio | 2.8 | < 3 | 0 - 4.4 |
| Triglycerides / HDL Ratio | 0.95 | < 2 | < 3.8 |
| TSH | 2.440 | 0.5 – 2.5 | 0.45 - 4.50 |
| T4, total | 7.0 | 6.0 – 12 | 4.5 - 12.0 |
| T3 Uptake | 28 | 28 - 35 | 24 - 39 |
| T3, Total | 71 | 100 – 180 | 71 - 180 |
| Vitamin D, 25-hydroxy | 25.1 | 35 - 60 | 30.0 - 100.0 |

| Marker | Value | Functional Range | Lab Range |
|---|---|---|---|
| WBC | 3.2 | 5.0 – 8.0 | 3.4 - 10.8 |
| RBC | 4.40 | 4.4 – 4.9 | 3.77 - 5.28 |
| Hemoglobin | 13.6 | 13.5 - 14.5 | 11.1 - 15.9 |
| Hematocrit | 40.4 | 37 - 44 | 34.0 - 46.6 |
| MCV | 92 | 85 – 92 | 79 - 97 |
| MCH | 30.9 | 27.7 – 32.0 | 26.6 - 33.0 |
| MCHC | 33.7 | 32 – 35 | 31.5 - 35.7 |
| RDW | 13.0 | 11.5 – 15.0 | 12.3 - 15.4 |
| Platelets | 235 | 150 – 415 | 150 - 379 |
| Neutrophils | 54 | 40 – 60 | |
| Lymphocytes | 36 | 25 – 40 | |
| Monocytes | 6 | 4.0 – 7.0 | |
| Eosinophils | 2 | 0.0 – 3.0 | |
| Basophils | 2 | 0.0 – 3.0 | |
| **Additional Tests:** | | | |
| T3, Free | 2.4 | 2.5 - 4.0 | 2 - 4.4 |
| T4, Free | 1.2 | 1 - 1.5 | 0.82 - 1.77 |
| Thyroid – TPO Ab | 19 | | 0 - 34 |
| Thyroid – TGA | <1.0 | | 0 - 0.9 |
| CRP-hs | 0.2 | < 1.0 | 0.00 - 3.00 |
| Homocysteine | 9.4 | < 9.0 | 0.0 - 15.0 |
| Vitamin B-12 | 391 | 450 – 2000 | 211 - 946 |
| Copper | 71 | | 72 - 166 |
| Zinc | 84 | | 56 - 134 |
| Zinc / Copper Ratio | 1.18 | > 0.85 | |
| Serum Methylmalonic Acid (MMA) | 186 | 0 - 325 | 0 - 378 |

Serum B12 is 391, below the 406 cut-off. Homocysteine was 9.4, slightly above the 7.0 cut-off. Serum MMA is completely normal. Note that copper and vitamin D are also low, which suggests malabsorption of nutrients.



**0091 Organix® Comprehensive Profile - Urine**

*Methodology: LC/Tandem Mass Spectroscopy, Colorimetric*

This report is not intended for the diagnosis of neonatal inborn errors of metabolism.

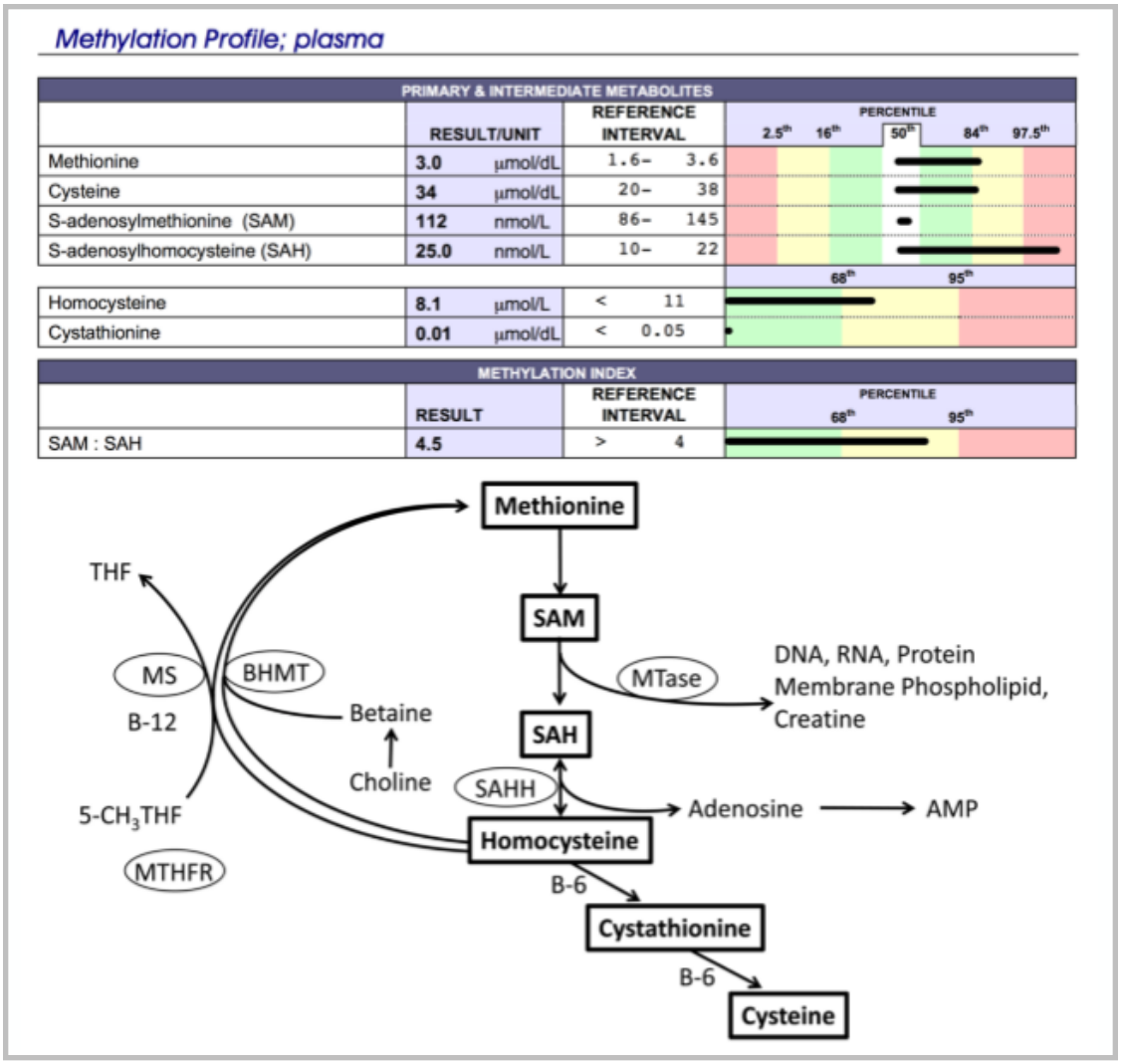
Ranges are for ages 13 and over

| | Results (mcg/mg creatinine) | | Quintile Ranking (1st–5th) | 95% Reference Range |
|---|---|---|---|---|
| **B-Complex Vitamin Markers** | | | | |
| ***(B1, B2, B3, B5, B6, Biotin)*** | | | | |
| 15. a-Ketoisovalerate | <DL* | | 0.25 | <= 0.49 |
| 16. a-Ketoisocaproate | <DL* | | 0.34 | <= 0.52 |
| 17. a-Keto-ß-methylvalerate | 0.20 | | 0.38 | <= 1.10 |
| 18. Xanthurenate | <DL* | | 0.34 | <= 0.46 |
| 19. ß-Hydroxyisovalerate | 4.6 | | 7.6 | <= 11.5 |
| **Methylation Cofactor Markers** | | | | |
| ***(B12, Folate)*** | | | | |
| 20. Methylmalonate | 2.0 | H | 1.7 | <= 2.3 |
| 21. Formiminoglutamate | 0.2 | | 1.2 | <= 2.2 |

Her urine MMA is well above the 1.5 cut-off. She did not have SIBO when I tested her, but had significant insufficiency dysbiosis, which was likely a cause of malabsorption. Note that in many cases discussed so far these are patients on a nutrient-dense Paleo diet. It goes to show that B12 deficiency can—and does—occur in omnivorous patients, just less frequently than it does in vegetarians and in particular vegans.

| Gene & Variation | rsID | Alleles | Result |
|---|---|---|---|
| COMT V158M | rs4680 | AG | +/- |
| COMT H62H | rs4633 | CT | +/- |
| COMT P199P | rs769224 | GG | -/- |
| VDR Bsm | rs1544410 | CT | +/- |
| VDR Taq | rs731236 | AG | +/- |
| MAO-A R297R | rs6323 | TT | +/+ |
| ACAT1-02 | rs3741049 | GG | -/- |
| MTHFR C677T | rs1801133 | AG | +/- |
| MTHFR 03 P39P | rs2066470 | GG | -/- |
| MTHFR A1298C | rs1801131 | TT | -/- |
| MTR A2756G | rs1805087 | AG | +/- |
| MTRR A66G | rs1801394 | AG | +/- |
| MTRR H595Y | rs10380 | __ | no call |
| MTRR K350A | rs162036 | AA | -/- |
| MTRR R415T | rs2287780 | __ | no call |
| MTRR A664A | rs1802059 | GG | -/- |
| BHMT-02 | rs567754 | CT | +/- |
| BHMT-04 | rs617219 | __ | no call |
| BHMT-08 | rs651852 | CT | +/- |
| AHCY-01 | rs819147 | CT | +/- |
| AHCY-02 | rs819134 | __ | no call |
| AHCY-19 | rs819171 | CT | +/- |
| CBS C699T | rs234706 | AG | +/- |
| CBS A360A | rs1801181 | AG | +/- |
| CBS N212N | rs2298758 | __ | no call |
| SHMT1 C1420T | rs1979277 | __ | no call |

In this case she was heterozygous for C677T. About 30 percent of the population is heterozygous, and it's only a 33 percent reduction of enzyme activity, typically not enough on its own to cause pathology without influence of environmental factors. But notice that she also has heterozygous mutations in numerous other genes involved in the methylation cycle and, collectively, these are probably playing a role.



*Methylation Profile; plasma*

| PRIMARY & INTERMEDIATE METABOLITES | RESULT/UNIT | REFERENCE INTERVAL |
|---|---|---|
| Methionine | 3.0 µmol/dL | 1.6– 3.6 |
| Cysteine | 34 µmol/dL | 20– 38 |
| S-adenosylmethionine (SAM) | 112 nmol/L | 86– 145 |
| S-adenosylhomocysteine (SAH) | 25.0 nmol/L | 10– 22 |
| Homocysteine | 8.1 µmol/L | < 11 |
| Cystathionine | 0.01 µmol/dL | < 0.05 |

| METHYLATION INDEX | RESULT | REFERENCE INTERVAL |
|---|---|---|
| SAM : SAH | 4.5 | > 4 |



Doctor's Data also has a methylation profile you can use as a follow-up test. It is not as complete as the HDRI panel. It only tests methionine, cysteine, S-adenosylmethionine, S-adenosylhomocysteine, homocysteine—which you already have on the case review blood panel—cystathionine, and SAMe-to-SAH ratio. No folate derivatives, or oxidized glutathione to reduced glutathione ratio, or other markers of oxidative stress such as the HDRI panel has.

However, there are a few advantages to this Doctor's Data panel. One is that some patients can get insurance coverage, where that doesn't happen with HDRI. Two is that HDRI is probably the slowest lab I've ever worked with. Sometimes the results take more than eight weeks to get back, which is a huge hassle if you're on a treatment plan with a patient. Doctor's Data typically comes back in a week or two, so you can get a much quicker idea of how the treatment is working and what to do next.

This patient with this particular panel had high S-adenosylhomocysteine and normal SAMe, which lead to a high SAH-to-SAMe ratio, and it was inhibiting methylation.