KASHI HEALTH

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ASHI No. 10-4-OR-03-1
CLIA No. 38D1058476

# NUTRITIONAL DEFICIENCIES GENOTYPING REPORT

| | | | |
|---|---|---|---|
| | | | 00/00/14 |
| **Patient Name:** | Johnny Health | **Date Sample Collected:** | 00/00/14 |
| **DOB:** | 01/01/1980 | **Date Sample Received/Tested:** | 00/00/14 |
| **Lab ID Number:** | 000000000 | **Date Reported:** Dr. KCL | Acme Center |
| **Ordering Physician:** | | **Ordering Facility:** | |

**Genes Tested:**

VITAMIN D – GC, NADSYN1/DHCR7, CYP2R1, VDR
VITAMIN A – BCMO1
FOLATE - MTHFR
VITAMIN B – FUT2
IRON – TMPRSS6

**KEY:** **GREEN: NON-RISK ALLELE** **RED: RISK ALLELE**

## VITAMIN D
### GC Mutations

**GC-1**
Results: [**G/G**]

**GC-2**
Results: [**C/C**]

## INDICATIONS

**GC-1 / GC-2 haplotypes are associated with vitamin D level status**

**[GC1: G/G; GC2: C/C]**
**G/C; G/C** isoform GC1s/GC1s; Individuals with this **G/C** isoform GC1s have an intermediate risk for vitamin D deficiency.

## BACKGROUND

The GC gene encodes the group-specific component protein, also referred to as the vitamin D binding protein, which functions to tightly bind and carry vitamin D metabolites to their target organs. A two-SNP combination in this gene is directly associated with variability in vitamin D [25(OH)D] concentrations.[1-4] These well-studied GC SNP variants result in allelic combinations (haplotypes) that give rise to three common GC isoforms, GC1F, GC1S, and GC2.[1] These variants in particular, change the amino acid sequence and alter protein function. Several studies have shown that vitamin D serum levels differ significantly depending on the GC genotype.[3,5] Without sufficient vitamin D, bones can become brittle or misshapen, leading to osteoporosis and/or fracture.[6] Maintaining adequate levels of vitamin D may be beneficial to patients not only for optimal bone health, but also for reducing cardiovascular and autoimmune disease, cancer risk, and increasing life expectancy.[6,7]

**GC-1 / GC-2 isoforms:**
**T/C** (GC1f) is considered the non-risk isoform and has the highest affinity for 25(OH)D. Individuals with this genotype are not associated with increased risk for vitamin D deficiency.[1]
**G/C** (GC1s) has intermediate affinity for 25(OH)D, individuals with this genotype have an intermediate risk profile for vitamin D deficiency.[1]
**T/A** (GC2) has the lowest affinity for 25(OH)D, and it is suggested that individuals with this genotype have lower vitamin D concentrations due to reduced binding capacity of the transport protein, and thus may be at risk for vitamin D deficiency.[1]

Limitations
Less common variants of GC are not reported by this test. While considered rare, the presence of these variants may result in a false positive or false negative report. If results obtained do not match the clinical findings, additional testing should be considered. All results should be interpreted in the context of clinical findings, relevant history, and other laboratory findings.

**References**

1. Gozdzik A et al. Association of vitamin D binding protein (VDBP) polymorphisms and serum 25(OH)D concentrations in a sample of young Canadian adults of different ancestry. J Steroid Biochem Mol Biol. 2011; 127:405–412.
2. Ahn J et al. Genome-wide association study of circulating vitamin D levels. Human Molecular Genetics. 2010; 19(13) 2739-2745.
3. Nissen J et al. Vitamin D Concentrations in Healthy Danish Children and Adults. PLOS One. 2014; 9(2):e89907.
4. Foucan L et al. Polymorphisms in GC and NADSYN1 Genes are associated with vitamin D status and metabolic profile in non-diabetic adults. BMC Endocrine Disorders. 2013; 13:36.
5. Wang T et al. Common genetic determinants of vitamin D insufficiency: a genome-wide association study. Lancet. 2010; 376(9736):180-188.
6. Grober U et al. Vitamin D: Update 2013: From rickets prophylaxis to general preventive healthcare. Dermato-Endocrinology. 2013; 5(3):331-47.
7. Bischoff-Ferrari HA et al. A pooled analysis of vitamin D dose requirements for fracture prevention. N Engl J Med. 2012; 367:40–9.

| **VITAMIN D**<br>**NADSYN1/DHCR7 Mutation** | **BACKGROUND** |
|---|---|
| Results: [**T/G**] | *NADSYN1/DHCR7* is a novel locus for association with vitamin D status, but with direct biological relevance. A G->T SNP is found in an intron of *NADSYN1* (nicotinamide adenine dinucleotide synthase 1), a gene that catalyzes the final step of NAD biosynthesis.[1] [1] This SNP is in high linkage disequilibrium with SNPs in *DHCR7* (7-dehydrocholesterol reductase), a gene that encodes the enzyme 7-dehydrocholesterol reductase, which converts the vitamin D precursor (7-DHC) to cholesterol.[2] It is hypothesized that it is the *DHCR7* gene product that is ultimately affected by this SNP. By converting 7-DHC into cholesterol, the needed substrate used by the body to generate vitamin D is removed, leaving the individual at a higher risk of vitamin D deficiency.[3] Maintaining adequate levels of vitamin D may be beneficial to patients not only for optimal bone health, but also for reducing cardiovascular and autoimmune disease, cancer risk, and increasing life expectancy.[4,5]<br><br>**T/T** and **T/G** individuals had higher levels of circulating 25(OH)D and lower frequency of vitamin D insufficiency, as compared to individuals with a G/G genotype.[3]<br>**G/G** homozygous individuals are associated with a greater risk of vitamin D insufficiency and therefore may be more prone to developing bone mineral disease disorders.[3] |
| **INDICATIONS** | |
| **T/G** Heterozygotes may have higher levels of serum vitamin D | |

**References**

1. Hara N et al. Molecular identification of human glutamine- and ammonia-dependent NAD synthetases. Carbon-nitrogen hydrolase domain confers glutamine dependency. J. Biol. Chem. 2003; 278(13):10914-10921.
2. Wassif CA et al Mutations in the human sterol $\Delta^7$-reductase gene at 11q12–13 cause Smith–Lemli–Opitz syndrome. Am. J. Hum. Genet. 1998; 63:55–62.
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| **VITAMIN D**<br>**CYP2R1 Mutation** | **BACKGROUND** |
|---|---|
| Results: [**A/A**] | A locus containing a single nucleotide polymorphism (SNP) associated with vitamin D levels, the *CYP2R1* gene encodes the hepatic enzyme 25-hydroxylase suggested to be responsible for the hydroxylation step of vitamin D metabolism in the liver.[1] The polymorphism is located in a coding region of this gene and may change the activity of the enzyme leading to a lowered 25(OH)D level.[1-3] As vitamin D deficiency has been implicated in a number of bone-related diseases involved with reduced calcium absorption such as osteoporosis, fracture and rickets,[4,5,7,8] this SNP may be considered as an indicator of risk for insufficient Vitamin D levels.[2,3,6] In addition to its' benefits in bone health, adequate vitamin D levels have also been associated with reduced cardiovascular and autoimmune disease, cancer risk, and increased life expectancy.[8]<br><br>**A/A** allele carriers have been shown to have higher vitamin D serum levels and therefore may not be at increased risk for the development of vitamin D deficiency.<br>**A/G and G/G** allele carriers are associated with lower vitamin D levels and thus are more likely to be at risk for developing vitamin D deficiency. |
| **INDICATIONS** | |
| **A/A** allele carriers do not possess risk alleles, are associated with significantly higher vitamin D levels, and are not at risk for vitamin D deficiency | |

**References**

1. Cheng JB et al. Genetic evidence that the human CYP2R1 enzyme is a key vitamin D 25-hydroxylase. Proc Natl Acad Sci U S A. 2004; 101:7711–7715.
2. Ahn J et al. Genome-wide association study of circulating vitamin D levels. Human Molecular Genetics. 2010; 19(13): 2739-2745.
3. Nissen J et al. Vitamin D Concentrations in Healthy Danish Children and Adults. PLOS One. 2014; 9(2):e89907
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| **VITAMIN D**<br>**VDR BsmI Mutation** | **BACKGROUND** |
|---|---|
| Results: [**A/A**] | The function of the Vitamin D receptor (VDR) is essential for promoting both calcium absorption and maintenance of adequate serum calcium and phosphate levels to allow for proper mineralization of bone.[1,2] One VDR variant, termed the VDR BsmI polymorphism, has been considered one of the factors influencing the efficacy of anti-osteoporotic treatments.[3] While earlier studies had presented conflicting results, a review of 26 publications assessing the link between this SNP and osteoporosis concluded that the more common variant may have a protective role against the development of osteoporosis, while the slightly less frequent allele is associated with disease risk and fracture risk.[4-5] Additionally, maintaining adequate levels of vitamin D may be beneficial to patients not only for optimal bone health, but also for reducing cardiovascular and autoimmune disease, cancer risk, and increasing life expectancy.[6,7]<br><br>**G/G** genotypes were found to confer protection against low bone mineral density disorders.[4,5]<br>**G/A** allele carriers may be associated with an intermediate risk of BMD disorders.[4,5]<br>**A/A** genotype may be associated with an increased risk of BMD disorders.[4,5] |
| **INDICATIONS** | |
| **A/A** genotype may be associated with an increased risk of BMD disorders | |

**References**

1. Morris HA. Vitamin D Activities for Health Outcomes. Ann Lab Med 2014; 34:181-186.
2. Turner AG. Vitamin D and bone health. Scand J Clin Lab Invest Suppl. 2012; 243:65-72.
3. Palomba S et al. BsmI vitamin D receptor genotypes influence the efficacy of antiresorptive treatments in postmenopausal osteoporotic women. A 1year multicenter, randomized and controlled trial. Osteoporos Int. 2005; 16(8):943-52.
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| **VITAMIN A**<br>**BCMO1 Mutation** | **BACKGROUND** |
|---|---|
| **Results:** [**T/T**] | Vitamin A is a fat-soluble vitamin crucial for proper vision, immune response, and cellular differentiation.[1] Vitamin A is created from natural vitamin A precursors called carotenoids. These are essential compounds found mainly in plant-based foods that must be properly metabolized before they can be used by the body.[2] The first step in the metabolic conversion of the carotenoids is their cleavage by the enzyme |
| **INDICATIONS** | |

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| **T/T** individuals do not carry risk alleles and are not likely to be at risk for vitamin A deficiency | □,□carotene 15,15'-monooxygenase 1 (BCMO1) located in the small intestine.[3] Unfortunately, about 45% of the Western population can be classified as low beta-carotene converters.[4] A polymorphism located upstream of the *BCMO1* gene has been reported to result in the reduction of the BCMO1 enzyme's catalytic activity by up to 48% in homozygous variant carriers, which can then lead to a vitamin A deficient state.<br><br>**T/T** individuals do not carry risk alleles and are not likely to be at risk for vitamin A deficiency<br>**T/G** individuals carry one risk allele and may be at risk for vitamin A deficiency<br>**G/G** genotypes are associated with an increased risk of vitamin A deficiency due to reduced enzyme activity |

**References**

1. Ferrucci L et al. "Common Variation in the B-Carotene 15, 15'-Monooxygenase 1 Gene Affects Circulating Levels of Carotenoids: A Genome-wide Association Study." The American Journal of Human Genetics. 2009; 84, 123-133.
2. Borel P et al. "Genetic Variations Involved in Interindividual Variability in Carotenoid Status." Mol Nutr Food Res. 2012; 56(2): 228-40.
3. Wyss A et al. Expression pattern and localization of β,β-carotene 15,15'-dioxygenase in different tissues. Biochem. J. 2001; 354:521–529.
4. Lietz G et al. "Single Nucleotide Polymorphisms Upstream from the B-Carotene 15, 15'-Monoxygenase Gene Influence Provitamin A Conversion Efficiency in Female Volunteers." J. Nutr. 2012; 142(1):161S-165S.

| **Folate (Vitamin B9)**<br>**MTHFR Mutations** | **BACKGROUND** |
|---|---|
| **MTHFR1-1: C677T**<br><br>Results: **C/C**<br><br>**MTHFR1-2: A1298C**<br><br>Results: **A/A** | The MTHFR (methylenetetrahydrofolate reductase) gene produces an enzyme that helps in processing folate and regulating homocysteine levels in the body. Folate is a critical nutrient involved in methylation, DNA synthesis and amino acid metabolism.[1] Impaired folate metabolism due to MTHFR enzyme inactivity, or a low folate level, results in elevated plasma homocysteine.[2] Homocysteine is an amino acid synthesized by the body through demethylation of methionine. In the presence of adequate B-vitamins, homocysteine is either irreversibly degraded to cysteine or it is remethylated back to methionine, an essential amino acid.[3] An elevated homocysteine level is known to be an independent risk factor for ischemic stroke, thrombotic and cardiovascular diseases.[4,5] Folate, vitamin B6 or vitamin B12 are all necessary for the proper conversion of homocysteine into methionine. A deficiency in any of these vitamins can cause |
| **INDICATIONS** | |

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| **MTHFR-1: C677T**<br>**C/C** individuals do not carry risk alleles and are not at risk for folate deficiency<br><br>**MTHFR-2: A1298C**<br>**A/A** Homozygous allele carriers have full enzymatic activity and are typically not at risk for increased levels of homocysteine | homocysteine levels to rise. In addition to vascular health, defects in folate metabolism due to dietary factors or MTHFR mutations may contribute to the pathophysiology of neural tube defects and a variety of malignancies.[6,8] Two single nucleotide variants known to affect MTHFR function are C677T (a change from cytosine to thymine at position 677 within the gene) and the A1298C mutation (a change from adenine to cytosine at position 1298 within the gene).[5-7] It is not uncommon for some individuals to have both MTHFR variants.[5]<br><br>**C677T**<br>**C/C** individuals do not carry risk alleles and are not at risk for folate deficiency<br>**C/T** Heterozygous allele carriers show an intermediate level of enzyme activity, but no significant effect on folate or homocysteine levels<br>**T/T** individuals carry two risk alleles and are at risk for folate deficiency and increased homocysteine levels<br><br>**A1298C**<br>**A/A** Homozygous allele carriers have full enzymatic activity and are typically not at risk for increased levels of homocysteine<br>**A/C** Heterozygosity is associated with slightly reduced enzyme activity, but not reduced folate levels or higher homocysteine levels<br>**C/C** Homozygote risk allele carriers are associated with intermediate levels of enzyme activity but do not have an increased risk of folate deficiency or higher homocysteine levels. |

**References:**

1. Wagner C. Biochemical role of folate in cellular metabolism. In: Bailey LB, editor. Folate in health and disease. New York, NY: Marcel Dekker Inc.; 1995. p. 23–42.
2. Bailey LB and JF Gregory III. Polymorphisms of Methylenetetrahydrofolate Reductase and Other Enzymes: Metabolic Significance, Risks and Impact on Folate Requirement. J Nutr. 1999; 129(5):919-22.
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5. Frosst P et al. A candidate genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase. Nat Genet. 1995; 10:111–113.
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8. Teng Z. The 677C>T (rs1801133) polymorphism in the MTHFR gene contributes to colorectal cancer risk: a meta-analysis based on 71 research studies. PLoS One. 2013; 8(2):e55332.

| VITAMIN B12 FUT2 Mutations | BACKGROUND |
|---|---|
| **FUT2-1**<br>**Results:** [**A/G**] | One of eight B vitamins, vitamin B12 plays a key role in brain and nervous system function, and is involved in the metabolism of every cell in the body; particularly DNA synthesis and methylation.[1] A vitamin B12 deficiency, due to low consumption of animal-source foods and/or inadequate absorption, often associated with pernicious anemia, becomes increasingly common as people age. Studies have also shown that individuals with reduced intake of vitamin B12 have elevated levels of homocysteine, an established predictor of cardiovascular disease.[2] Importantly, clinical research has identified multiple genetic |
| **INDICATIONS** | |

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| **FUT2-1**<br>**A/G** allele carriers possess one risk allele and may be associated with higher vitamin B12 concentrations | mutations that affect vitamin B12 levels and the risk of deficiency.[2,3] Genetic variants of the *FUT2* gene give rise to a non-functional FUT2 enzyme resulting in an inability to synthesize ABH-antigens on mucosal surfaces which is referred to as a 'non-secretor status'.[4] The non-secretor phenotype has been associated with higher vitamin B12 levels than the secretor phenotype.[3,5] It has been suggested that the reduced activity of the FUT2 enzyme may decrease susceptibility to bacterial infection and indirectly lower the risk of vitamin B12 malabsorption, thereby resulting in higher vitamin B12 concentrations.[2]<br><br>**FUT2-1**<br>**A/A** allele carriers possess no risk alleles and are associated with higher vitamin B12 serum concentrations.<br>**A/G** allele carriers possess one risk allele and may be associated with higher vitamin B12 concentrations.<br>**G/G** allele carriers have two risk alleles and may be at an increased risk for vitamin B12 deficiency. |

**References**

1. http://lpi.oregonstate.edu/infocenter/vitamins/vitaminB12/.
2. Tanaka T et al. Genome-wide association study of vitamin B6, vitamin B12, folate, and homocysteine blood concentrations. Am. J. Hum. Genet. 2012; 84:477–482.
3. Hazra A et al. Common variants of FUT2 are associated with plasma vitamin B12 levels. Nat. Genet. 2008; 40:1160–1162.
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5. Hazra A et al. Genome-wide significant predictors of metabolites in the one-carbon metabolism pathway. Hum Mol Gen. 2009; 18(23):4677-4687

| **IRON**<br>**TMPRSS6 Mutation** | **BACKGROUND** |
|---|---|
| **Results:** [**T/T**]<br><br>**INDICATIONS**<br><br>**T/T** genotypes are associated with an increased risk of iron deficiency | Iron is a common nutrient deficiency, resulting in anemia which can lead to fatigue, weakness, pale skin, and shortness of breath.1 Several clinical biomarkers of iron – including serum ferritin concentrations, hematocrit, hemoglobin, and levels of iron-bound transferrin – are strongly heritable, indicating a role for genetics in iron deficiency.[2] Scientists have found that a mutation in the gene coding for transmembrane protease serine 6 (TMPRSS6) is associated with several clinical indicators of anemia, including levels of serum iron, transferrin saturation, erythrocyte mean cell volume, and hemoglobin.3,4 Evidence suggests that carrying even one copy of the risk allele can negatively impact your iron status.<br><br>**C/C** individuals do not carry risk alleles and are not at increased risk for iron deficiency.<br>**C/T** individuals carry one risk allele and may be at risk for iron deficiency.<br>**T/T** individuals carry two risk alleles and are associated with an increased risk of iron deficiency. |

**References**

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2. Patel KV. "Variability and heritability of hemoglobin concentration: an opportunity to improve understanding of anemia in older adults." Haematologica 2008; 93:1281-1283.
3. Benyamin B et al. Common variants in *TMPRSS6* are associated with iron status and erythrocyte volume. Nature Genetics 2009; 41:1173-1175.
4. Chambers JC et al. Genome-wide association study identifies variants in *TMPRSS6* associated with hemoglobin levels. Nature Genetics 2009; 41:1170-1172.

***Additional information will be supplied to your provider to help inform treatment options.**

*This test was developed and its performance characteristics determined by Kashi Clinical Laboratories. It has not been cleared or approved by the FDA. The laboratory is regulated under CLIA as qualified to perform high-complexity testing. This test is used for clinical purposes. It should not be regarded as investigational or for research.*

**Reported and Reviewed By:**

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