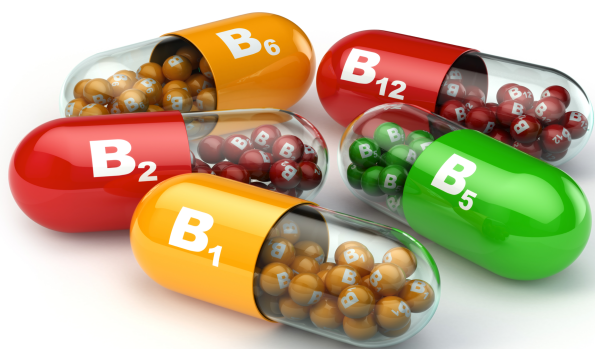


### Concentration of MMA in blood indicates vitamin B<sub>12</sub> deficiency



Methylmalonic Acid (MMA) is a biomarker for the assessment of vitamin B<sub>12</sub> status. Increased concentration of MMA in blood and urine indicates Vitamin B<sub>12</sub> deficiency.

Vitamin B<sub>12</sub> is a nutrient that helps keep your body's blood and nerve cells healthy including DNA synthesis. Vitamin B<sub>12</sub> also prevents megaloblastic anemia which makes people tired and weak.

Vitamin B<sub>12</sub> deficiency gives rise to severe conditions for the child. MMA is therefore included as one of 25 analytes in the PKU register

### DESCRIPTION OF THE BIOMARKER

Numerous studies have now established that serum vitamin B<sub>12</sub> has limited diagnostic value as a stand-alone marker. Low serum levels of vitamin B<sub>12</sub> not always represent deficiency, and likewise, severe functional deficiency of the micronutrient has been documented in the presence of normal and even high levels of serum vitamin B<sub>12</sub> [1, 2].

MMA is considered a more specific marker of vitamin B<sub>12</sub> deficiency [3]. Serum values of MMA, ranging from >0.28 to 0.36 µmol/L indicate B<sub>12</sub> deficiency.

### CE LABELED ANALYZING METHOD

redhot diagnostics has developed a method with several unique features which makes the method very reliable:

- The use of a SPE plate in the kit enables the removal of interfering compounds
- The calibrator is labeled with D<sub>3</sub>-MMA. This excludes interference with intrinsic MMA, and substituting blood in the calibration curve is not needed
- The column separates the isobaric succinic acid from MMA and prevents false positive results

### KIT CONTENTS

Art. no. 40-2001 96 determinations including column  
Art. no. 40-2002 96 determinations, replacement kit

D <sub>3</sub> -MMA calibrator solution	0.2 ml
<sup>13</sup> C <sub>4</sub> -D <sub>3</sub> -MMA Int. std solution	10 mL
Elution solution	2x30 mL
Reconstitution solution	10 mL
Tuning solution	0.5 mL
SPE plate	1 pcs
Column	1 pcs

### QC samples

QC low	Art.no	40-1007	0.5 mL
QC med	Art.no	40-1008	0.5 mL
QC high	Art.no.	40-1009	0.5 mL

### Levels of MMA in blood which indicating vitamin B<sub>12</sub> deficiency

#### Reference interval

> 0.28 µmol/L	for persons < 50 years of age
> 0.36 µmol/L	for persons > 50 years of age

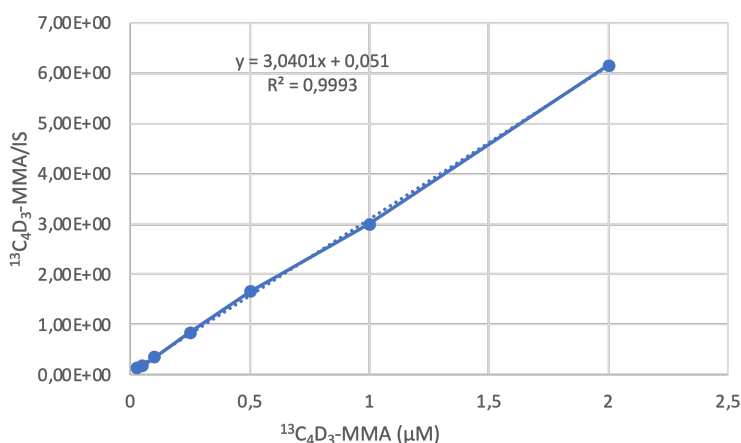
Vitamin B<sub>12</sub> deficiency is a multifactorial condition caused by insufficient intake (nutritional deficiency) as well as acquired or inherited defects that disrupt B<sub>12</sub> absorption, processing and trafficking pathways (functional deficiency)

## Principles of the procedure

Methylmalonic acid is extracted from blood/serum by mixing the sample with extraction solution including the internal standard. The sample is purified on a validated SPE plate. MMA is eluted by the elution buffer and evaporated to dryness, reconstitution buffer is added to the sample and transferred to injection vials for analyze.

The internal standard is <sup>13</sup>C<sub>4</sub>-D<sub>3</sub>-MMA and the calibration curve is made with the use of D<sub>3</sub>-MMA

## D<sub>3</sub>-MMA calibration curve



## Reproducibility of D<sub>3</sub>-MMA

Sample	[µmol/	Mean	% Std.
QC low	0.075	0.076	4.8
QC mid	0.75	0.74	4.4
QC high	1.5	1.5	4.3

## Detection level (lowest calibrator level)

The least detectable level is 0.025 µM

## Measuring range

The calibrator range is 0.025 to 2.0 µM

## Reproducibility of <sup>13</sup>C<sub>4</sub>-D<sub>3</sub>-MMA, internal standard

Standard deviation for the MS area of D<sub>3</sub>-MMA in blood samples prepared with SPE was 43 % (41 injections). The relatively high standard deviation is due to the variation of the volume eluted from the SPE plate. The reproducibility of QC samples is high (low std dev %, table above) and this shows that labelled IS fully compensate for the different in volume eluted from the SPE plate.

## References

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