

Patient Information

Before taking a sample for TPMT activity, patients should be advised that DNA confirmation may be performed. The only known implication for the genetic variation in TPMT expression is intolerance to thiopurine drugs. Patients can be directed to www.labtestsonline.org.uk for further information.

References

1. Ford LT, Berg JD. Determination of thiopurine S-methyltransferase activity in erythrocytes using 6-thioguanine as substrate and a non-extraction liquid chromatographic technique. *J Chrom B* 2003; 798: 111-115
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3. Barlow NL, Graham V & Berg JD. Expressing thiopurine S-methyl transferase (TPMT) activity as units per litre of whole blood overcomes misleading high results in patients with anaemia. *Ann Clin Biochem* 2010; in press
4. Weinshilboum RM, Sladek SL. Mercaptopurine pharmacogenetics: monogenic inheritance of erythrocyte thiopurine methyltransferase activity. *Am J Hum Genet* 1980; 32: 651-62
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8. Ford LT, Berg JD. Thiopurine S-methyltransferase (TPMT) assessment prior to starting thiopurine drug treatment; a pharmacogenomic test whose time has come. *J Clin Pathol* 2010; 63: 288-295

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Thiopurine S-Methyltransferase Activity in Whole Blood (mU/L)

TPMT [E.C. 2.1.1.67]



Clinical Biochemistry
City Hospital



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Version No. 1.09

Sending Specimens for Analysis

Sample requirements: A 2ml EDTA whole blood sample can be used for both TPMT activity and TPMT genotyping analysis. Please note lithium heparin samples may be used for TPMT activity but not for genotyping.

The sample should not have been frozen. Please store samples prior to dispatch at 4°C. Send samples by first class post at ambient temperature to the address on the back of this leaflet.

TPMT activity is stable at room temperature for 6 days and at 4 °C for at least 12 days. We highlight samples >8 days old on receipt. and at 4 °C for at least 12 days.

Samples from patients who have received a recent blood transfusion can give misleading results. Please provide details of recent transfusions (within 90 days) on the request form and we will perform genotyping to confirm the patient's TPMT status.

Reference Ranges

Reference intervals for our whole blood TPMT activity assay are as follows:

Whole blood TPMT activity*	(mU/L)
Deficient	< 10
Low	20 - 67
Normal	68 - 150
High	>150

*37°C, pH 7.4 using 6-TG as substrate
N.B. TPMT activity 10 – 19 mU/L may be seen in TPMT deficient patients who have been transfused.

City Hospital TPMT Activity Service Assay

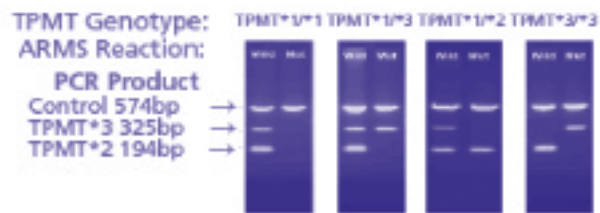
Our whole blood TPMT activity assay uses 6-thioguanine as a substrate and measures the product 6-methyl thioguanine by HPLC.¹ The assay shows a within batch CV of <5 % and between batch CV of <8.5 %.

TPMT genotyping

TPMT genotyping is used to confirm patient TPMT status for selected samples:

- Deficient TPMT activity
- Recent blood transfusion
- Previous severe reaction to thiopurine drugs
- Change in TPMT status on repeat testing

We use a multiplex amplification refractory mutation system (ARMS) strategy to screen for the common TPMT mutations TPMT*2 and TPMT*3, which account for approximately 60-95% of mutant TPMT alleles in most populations.²



Figure; Example results for TPMT genotyping

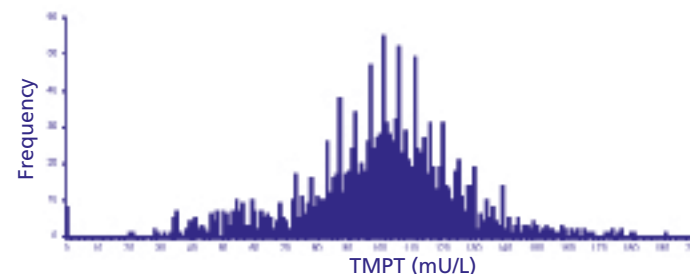
TPMT enzyme activity (mU/L)

TPMT enzyme activity is expressed in mU/L. Previous methods have corrected for red blood cell indices, however, we have shown that this new approach overcomes misleading high results in patients with anaemia.³

Clinical Use of TPMT Activity Measurement

Thiopurine drugs are widely used to treat inflammatory and autoimmune disease, leukaemia and to prevent rejection post organ transplant. These drugs are catabolised to inactive metabolites by TPMT, which in effect reduces concentrations of the active metabolite, 6-thioguanine nucleotides (6TGN).

TPMT activity exhibits autosomal co-dominant polymorphism. In a Caucasian population approximately 89% have normal enzyme activity, 11% low activity and 0.3% undetectable levels (deficient).⁴



Distribution of TPMT activity for 1563 consecutive whole blood samples

Measurement of TPMT activity prior to starting thiopurine drugs is now recommended.^{5,6} Patients with undetectable TPMT activity are generally not treated with thiopurine drugs due to increased risk of severe side effects, e.g., myelosuppression. Those with low activity usually receive a reduced dose. In patients with high activity, an increased dose may lead to accumulation of inactive metabolites and increased risk of hepatotoxicity.⁷