The effect of thiopurine toxicity on S-adenosylmethionine concentrations

Warner BD[1], Turner C[3], Sobczyńska-Malefora A[2], Arenas-Hernandez M[2], Dalton N[3], Sanderson J[1], Marinaki A[2]

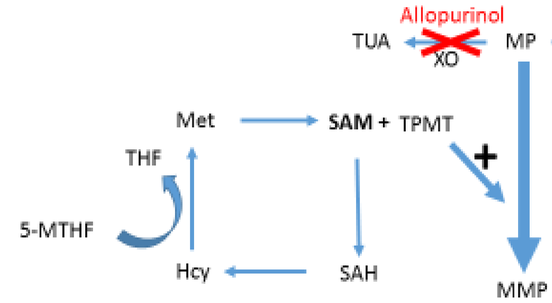
Departments of Gastroenterology[1],Viapath Pathology Services[2], and WellChild Research Laboratory[3], Guy's and St Thomas' and Evelina Hospitals, London

Introduction

- Thiopurines(azathioprine(Aza) and mercaptopurine(MP) are commonly used in the treatment of inflammatory bowel disease (IBD).
- S-adenosylmethionine(SAM) is a co-factor for the methylation of MP to methylmercaptopurine (MMP) by thiopurine methyltransferase(TPMT).
- Hypermethylation is where excess MMP is produced(MMP:TGN >11) affecting 20% of patients and is a cause of lack of response or hepatotoxicity in IBD patients.
- Low SAM and high Hcy have been associated with liver disease suggesting that changes in concentrations of these compounds through hypermethylation may be the cause of hepatotoxicity.[1]
- Switching to a lower dose of thiopurine, and adding allopurinol(LDTA), reduces the MP available to TPMT, thereby preventing hypermethylation and hepatotoxicity.

Method

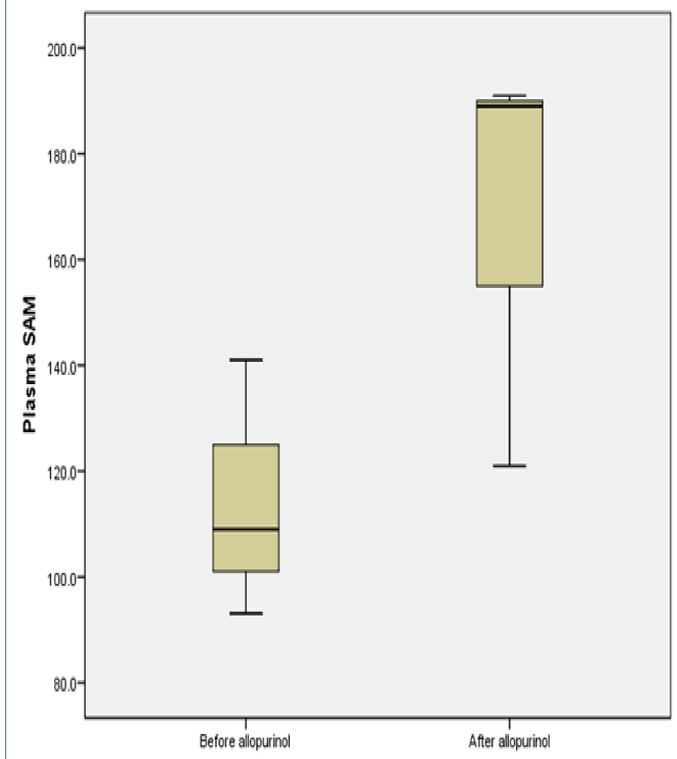
- 18 IBD patients on thiopurines had plasma SAM, SAH, Hcy, Methionine(Met) and 5-methyltetrahydrofolate(5-MTHF) measured. All were wild type for TPMT.
- 6 patients were hypermethylators with hepatotoxicity(ALT > 56 IU/L) at the time compounds were measured. 3 of these patients had measurements repeated at least 6 weeks after LDTA. Comparisons were made with 5 healthy controls.
- SAM, SAH, Hcy were measured by LCMS/MS and 5-MTHF by HPLC.



The diagram shows the interaction between MTHF and SAM mediated methylation of MP. A combination of allopurinol, a xanthine oxidase(XO) inhibitor, and low dose Aza or MP, increases the amount of the MP available to form active thioguanine nucleotides(TGNs), and decreases levels of MMP.

Phenotype (Males)	Mean (SD) Plasma SAM (nmol/L)	Mean (SD) Plasma SAH (nmol/L)	Mean (SD) SAM:SAH ratio	Met (SD) (µmol/L)	Hcy (SD) (µmol/L)	5-MTHF (SD) (ng/ml)
Control n=5(3)	84 (8.3)	18 (4.9)	5.1 (1.2)	29 (5.9)	11 (2.5)	10 (3.9)
IBD patients on thiopurines n=12(10) (normal metabolism)	110 (21.9)	18 (2.1)	6.1 (1.1)	38 (13.1)	13 (3.9)	17 (19.8)
IBD patients with hypermethylation and hepatotoxicity n=6(3)	109 (20.5)	18 (4.2)	6.4 (2.3)	35 (9.9)	9 (3.6)	11 (5.4)
Before LDTA n=3(1)	114 (24.4)	20 (3.6)	5.7 (0.4)	31 (6.4)	12 (2.9)	9 (3.7)
After LDTA n=3(1)	167 (39.8)	16 (2.2)	10.2 (2.6)	29 (9.0)	12 (4.6)	10 (2.4)

The table compares plasma concentrations of SAM, SAH, Met and Hcy in normal and abnormal (hypermethylation with hepatotoxicity) thiopurine metabolisers and the change in these compounds in 3 patients after LDTA.



Boxplot shows the change in plasma SAM levels measured in 3 patients with hepatotoxicity and hypermethylation before and after LDTA

Results

- There were no significant age or weight differences between the groups.
- SAM was significantly higher in patients on thiopurines compared to controls ($P<0.05$) and SAM increased after LDTA was added.
- There were no significant differences in SAM and SAH between patients with normal metabolites and those who had hepatotoxicity and hypermethylation.
- Met, Hcy and 5-MTHF did not alter between the groups.

Conclusions

- These results suggest that the Hcy cycle is disrupted by thiopurines and by LDTA independent of 5-MTHF concentrations.
- Since altered SAM concentrations have been attributable to liver disease, disruption of this cycle by thiopurines is a potential mechanism by which hepatotoxicity could be occurring.

References

1. Klepacki J, Brunner N, Schmitz V, et al. Development and validation of an LC-MS/MS assay for the quantification of the trans-methylation pathway intermediates S-adenosylmethionine and S-adenosylhomocysteine in human plasma. Clin Chim Acta 2013;421:91-7.