ISRCTN58287360 https://doi.org/10.1186/ISRCTN58287360

Retrospective and prospective evaluation of the pharmacogenetics and metabolites of thiopurine drugs in the treatment of inflammatory bowel disease

Submission date 10/01/2010	**Recruitment status** No longer recruiting	[_] Prospectively registered [_] Protocol
Registration date 19/01/2010	**Overall study status** Completed	[_] Statistical analysis plan [_] Results
Last Edited 04/01/2012	**Condition category** Digestive System	[_] Individual participant data [_] Record updated in last year

Plain English summary of protocol
Not provided at time of registration

Contact information

Type(s)
Scientific

Contact name
Dr Fangbing Zhang

Contact details
Department of Gastroenterology
The First Affiliated Hospital
Sun Yat-sen University
58 Zhongshan Road
Guangzhou
China
510080

Additional identifiers

Study information

Scientific Title

Retrospective and prospective evaluation of the pharmacogenetics and metabolites of thiopurine drugs in the treatment of inflammatory bowel disease: a self-controlled trial and a randomised controlled trial

Study objectives

The thiopurine drugs, such as azathioprine (AZA), 6-mercaptopurine (6-MP) and 6-thioguanine (6-TG) are one kind of immunosuppressive agents which are widely used in the treatment of many diseases. AZA and 6-MP are well established in the treatment of inflammatory bowel disease (IBD) and have proven to be effective in both inducing and maintaining remission of Crohn's disease (CD) and ulcerative colitis (UC) with an efficacy rate of 55 - 70%. Unfortunately, thiopurine drugs have to be withdrawn in 10 - 30% of cases due to serious side effects, such as bone marrow toxicity and hepatotoxicity. Both efficacy and toxicity of these drugs are hard to be predicted in the long-term treatment.

After one oral dose, AZA is rapidly converted to 6-MP and an imidazole derivative, but up to 12% of the dose can be split to form the purine base hypoxanthine and thioimidazole. Three enzymes compete to metabolize 6-MP: xanthine oxidase (XO), thiopurine methyltransferase (TPMT), and hypoxanthine guanine phosphoribosyltransferase (HPRT). 6-MP activation, catalysed by HPRT, forms initially the 6-MP nucleotides and eventually the active metabolites, the thioguanine nucleotides (6-TGNs). The TGN metabolites act as purine antagonists and induce cytotoxicity and immunosuppression by inhibition of rubinucleic acid (RNA), deoxyribonucleic acid (DNA), and protein synthesis. These cytotoxic properties are, at least partially, due to the direct incorporation of TGN into deoxyribonucleic acid.

The observed inter-individual differences in therapeutic response or toxicity of AZA treatment were partly explained by the variable formation of active metabolites because of genetic polymorphisms of the genes encoding crucial enzymes in thiopurine metabolism. These enzymes include TPMT, XO, HPRT, inosine-5-monophosphate dehydrogenase (IMPDH) and inosine triphosphate pyrophosphatase (ITPA). Clinical trials have also demonstrated that 6-TGNs levels were associated with clinical efficacy and toxicity of AZA/6-MP. However, most of these trials were performed in western countries with a retrospective design or a short-time follow-up.

On the basis of our previous studies and reports from other researchers, we propose the following hypotheses:
1. The polymorphisms of thiopurine drugs' metabolism enzymes were associated with clinical efficacy and adverse effects of AZA/6-MP in Chinese IBD patients
2. There is a safe and therapeutic concentration threshold of 6-TGNs in patients with undergoing AZA/6-MP therapy
3. Verifying the 6-TGNs concentration threshold by pharmacogenetic-guided therapy to confirm that monitoring 6-TGNs concentrations during AZA/6-MP treatment could increase the clinical efficacy and safety

Ethics approval required

Old ethics approval format

Ethics approval(s)

Ethics Committee of the First Affiliated Hospital of Sun Yat-Sen University approved on the 30th June 2008

Primary study design

Interventional

Study design
Retrospective study (self-controlled) and a prospective randomised controlled trial

Study type(s)
Treatment

Health condition(s) or problem(s) studied
Inflammatory bowel disease

Interventions
Retrospective self-controlled trial:
Retrospectively assess the impact of genetic variation and concentration of metabolites on efficacy and toxicity of AZA/6-MP in patients with IBD.

Drug dose was started at 1 mg/kg daily for AZA (Imurel®, GlaxoSmithKline, Sweden) and 0.5 mg /kg daily for 6-MP (Puri-Nethol®, GlaxoSmithKline, Sweden) in the first week, then increased to 2 mg/kg daily for AZA and 1.0 mg/kg daily for 6-MP without dose alteration in following weeks.

Clinical data including sex, age, age at diagnosis and site of disease, type of inflammatory bowel disease, weight, dose of AZA/6-MP, indication for AZA/6-MP therapy, concomitant therapy (5-aminosalicylates, infliximab or other drugs), and toxicity data including full blood counts and liver function tests were collected. Patient information such as disease activity scores were collected by a physician.

Haematotoxicity observed as myelosuppression included leukopaenia and neutropaenia. Leukopaenia was defined as a leukocyte count (white blood cell [WBC]) less than 3.5 x 10^9/L, and neutropaenia was defined as less than 1.5 x 10^9/L neutrophils. Each decrease of WBC and neutrophils should be continuously observed in two days, and recovered in the next one or two weeks after AZA/6-MP withdrawal. Hepatotoxicity was defined as an increase in transaminases at least two times higher than the normal value. Pancreatitis was diagnosed when compatible symptoms (abdominal pain) were present and serum amylase was increased two times above the upper normal limit. Flu-like symptoms included febris, headache, courbature and arthralgia all over the body while gastrointestinal intolerance was defined as hypogeusia, nausea and vomiting.

2 ml venous blood samples were taken prior to treatment for genetic variation detecting. The genotypes of TPMT, glutathione S-transferase (GST), inosine triphosphatase (ITPA) were measured by technique based-on polymerase chain reaction (PCR). The activities of TPMT, HPRT, GST, xanthine oxidase (XO), ITPA and inosine-5-monophosphate dehydrogenase (IMPDH) were measured by HPLC method.

6-TGNs levels were analysed at weeks 2, 4, 8, 12, 24 and 48 after initiation of AZA/6-MP treatment. 2 ml venous blood samples were drawn 6 - 12 hours after AZA/6-MP intake for concentration monitoring. The detection of concentration of 6-TGNs was performed by HPLC method.

The impact of enzyme polymorphisms on clinical efficacy and adverse effects were assessed in patients with stable dose of AZA/6-MP. The therapeutic and safe concentration threshold of 6-TGNs was calculated based on treatment outcome and toxicity. These results were used to established a pharmacogenetic-guided therapy combination with metabolites monitoring.

Prospective randomised controlled trial:
Prospectively verify the predictive value of monitoring impactive enzymes and confirm the concentration threshold of 6-TGNs in patients with IBD undergoing AZA/6-MP treatment.

When the retrospective study was completed, a prospective study was conducted to validate the results obtained from the retrospective study. Consecutive patients with IBD were prospectively enrolled into a study of AZA/6-MP undertaken. The patients were divided into the self-controlled study and randomised controlled study.

In the self-controlled study, patients who had developed adverse effects in the retrospective research were enrolled to compare the safety of previous stable dose therapy and pharmacogenetic-guided metabolites monitoring therapy. Drug dose was 1 mg/kg daily for AZA and 0.5 mg/kg daily for 6-MP, the dose was adjusted according to therapeutic and safe 6-TGNs concentration.

In the randomised controlled study, the new enrolled patients were divided into the stable dose therapy group (2 mg/kg daily for AZA and 1.0 mg/kg daily for 6-MP) and pharmacogenetic-guided metabolites monitoring therapy group (adjusted doses according to therapeutic and safe 6-TGNs concentration). The efficacy and safety of both groups were compared.

Polymorphisms of impactive enzymes were assessed before AZA/6-MP initiation and the 6-TGNs levels were monitored during clinical control visits in both self-controlled and randomized controlled studies.

In all of the above studies, control visits were performed in enrolled patients every two weeks for the first month, every month for the following 2 months, and then every 3 months. During these control visits, patients had complete blood counts measurements and liver function tests, and were clinically reviewed while adverse effects were recorded. These results are compared between groups to assess the predictive value of detection impactive enzymes on adverse effects before drug administration and to confirm the therapeutic and safe concentration threshold of 6-TGNs during routine monitoring.

Intervention Type
Drug

Phase
Not Applicable

Drug/device/biological/vaccine name(s)
AZA (Imurel®), 6-MP (Puri-Nethol®)

Primary outcome(s)
1. Distribution of TPMT, HPRT, GST, XO, ITPA and IMPDH polymorphisms in Chinese IBD patients
2. The impact of genetic variation on efficacy and toxicity of AZA/6-MP in patients with IBD
3. The therapeutic and safe concentration threshold of 6-TGNs in Chinese IBD patients undergoing AZA/6-MP therapy

Key secondary outcome(s)
Comparisons between the stable dose therapy group and pharmacogenetic-guided metabolites monitoring therapy group:
1. The efficacy of AZA/6-MP treatment was only assessed when the treatment had been continued for 24 weeks or more. Remission was defined as no need corticosteroids for at least 1 month, and a Pediatric Crohn's Disease Activity Index (PCDAI) less than 10, Crohn's Disease Activity Index (CDAI) less than 150 or according to the criteria for remission defined by Truelove and Witts and without relapse in remaining weeks. Surgery and initiation of biological or other therapies were considered as treatment failure.
2. Haematotoxicity observed as myelosuppression included leukopaenia and neutropaenia. Leukopaenia was defined as a leukocyte count (WBC) less than 3.5 x 10^9/L, and neutropaenia was defined as less than 1.5 x 10^9/L neutrophils. Each decrease of WBC and neutrophils should be continuously observed in two days, and recovered in the next one or two weeks after AZA/6-MP withdrawal. Hepatotoxicity was defined as an increase in transaminases at least two times higher than the normal value. Pancreatitis was diagnosed when compatible symptoms (abdominal pain) were present and serum amylase was increased two times above the upper normal limit. Flu-like symptoms included febris, headache, courbature and arthralgia all over the body while gastrointestinal intolerance was defined as hypogeusia, nausea and vomiting.

Completion date
01/06/2011

Eligibility

Key inclusion criteria
1. All consecutive patients with the diagnosis of IBD who received the azathioprine treatment at the Gastroenterology Outpatient Clinic of the First Affiliated Hospital of Sun Yat-sen University
2. Aged from 3 - 74 years, either sex
3. Steroid-dependent disease: unable to reduce corticosteroids below the equivalent of prednisolone 15 mg/day (or budesonide below 3 mg/day) within three months of starting corticosteroids or relapse within three months of stopping corticosteroids
4. Frequent relapses: greater than three relapses in one year or greater than two relapses in six months
5. Remission maintenance
6. Post-operative prophylaxis

Participant type(s)
Patient

Healthy volunteers allowed
No

Age group
Other

Sex
All

Key exclusion criteria

1. Blood transfusion or administration of cyclosporine or methotrexate (MTX) within the last 3 months
2. Treatments potentially interfering with AZA metabolism, including allopurinol and diuretics
3. Insufficient function in heart, liver or kidney
4. Active infection
5. Pregnancy

Date of first enrolment
01/04/2008

Date of final enrolment
01/06/2011

Locations

Countries of recruitment
China

Study participating centre
Department of Gastroenterology
Guangzhou
China
510080

Sponsor information

Organisation
Sun Yat-sen University (China)

ROR
https://ror.org/0064kty71

Funder(s)

Funder type
Government

Funder Name
National Natural Science Foundations of China (China) (refs: 30171098, 30572231)

Results and Publications

Individual participant data (IPD) sharing plan

IPD sharing plan summary
Not provided at time of registration