Immunomodulatory therapy in paediatric inflammatory bowel disease

Imunomodulacijsko zdravljene vnetne črevesne bolezni v pediatriji

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Izvleček

Pri zdravljenju otrok s Crohnovo boleznijo in ulcerativnim kolitisom so imunomodulacijska zdravila nepogrešljiva, zlasti še za vzdrževanje remisije. Od teh sta najučinkovitejša azatioprin in 6-merkaptopurin, ki imata v primerjavi z drugimi imunomodulacijskimi zdravili tudi najmanj neželenih stranskih učinkov. Zelo pomembna prednost imunomodulacijskih zdravil je, da zmanjšujejo potrebo po uporabi steroidnih sredstev, kar je še posebej pomembno za razvoj otrok. Čeprav zaradi počasnega začetka delovanja večina teh zdravil ni primerna za indukcijo remisije, so lahko koristna za hitrejše ukinjanje steroidov in njihovo premostitev na azotropin ali 6-merkaptopurin. Ta pregledni članek obravnava mehanizem delovanja, na dokazih temelječo terapevtsko učinkovitost in neželene stranske učinke imunomodulacijskih zdravil.

Abstract

Immunomodulatory drugs are indispensable in the treatment of children with both Crohn's disease and ulcerative colitis, especially in maintaining remission. Among them, azathioprine and 6-mercaptopurine are the most effective in this respect and they elicit less adverse reactions compared to other immunomodulatory drugs. A very important benefit of immunomodulatory therapy is that they are steroid-sparing agents, which is especially essential in developing children. Although, due to their slow onset of action, most of these drugs are not appropriate for induction of remission, their early introduction may be helpful in faster tapering of steroids and bridging them to azathioprine or 6-mercaptopurine. In this review the mechanism of action, evidence based therapeutical efficiency and adverse reactions to immunomodulatory drugs will be reviewed.

Introduction

Inflammatory bowel disease (IBD) may result from exaggerated stimulation of the mucosal immune system by luminal bacterial flora, which causes an exaggerated stimulation of innate and adaptive mucosal immune system resulting in increased CD4 + T-cell activation and enhanced intestinal permeability finally leading to chronic inflammation.[1,2] So it is obvious that inhibition of increased production of activated lymphocytes can have therapeutical effect. This explains why immunmodulatary drugs are effective in the treatment of both Crohn disease (CD) and ulcerative colitis (UC). Azathioprine (AZA) and 6-mercaptopurine (6-MP) are the most effective among these drugs.

Azathioprine and 6-mercaptopurine

Mechanism of action

AZA and 6-MP were developed more than 50 years ago and first the drugs were used for the treatment of leukemia and after organ transplantation. 6-MP and its prodrug AZA have both cytotoxic and immunomodulatory properties. Azathioprine is a thiopurine linked to an imidazole ring with a thioether (*Figure 1*). By reductive cleavage

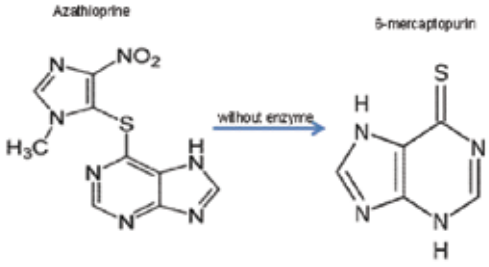

Figure 1: Structure of azathioprine and 6-mercaptopurin

of the thioether bond, 6-MP will be released from the prodrug. This step is mediated by glutathione and similar compounds in the liver and the intestinal wall as well as on the erythrocytes, without the aid of enzymes.[3] 6-MP and AZA have similar therapeutical effect, but AZA has a better therapeutic index, which is the ratio of toxic and therapeutic dose. 6-MP is a purine analogue, which inhibits RNA and DNA synthesis especially in the leukocytes and so inhibits the cell fission. Thiopurines also exert their action through the specific inhibition of Rac1 activation, leading to apoptosis of T-cells. This explains how AZA and 6-MP reduce the proliferation of T- and B-lymphocytes and impair their effector T-cell activity.[3] The hypoxanthine phosphoribosyl transferase (HPRT) enzyme leads the formation of 6-thioguanine (6-TGN), which is the active metabolite.[4] There are two other competing pathways of 6-MP metabolism, the production of 6-methylmercaptopurine by thiopurine methyltransferase (TPMT) and the formation of 6-thiouric acid by xanthine oxidase (XO). The balance of these pathways will determine the production of 6-TGN. (*Figure 2*)

Therapeutical evidences

Crohn's disease

An early meta-analysis of placebo-controlled randomized trial in adult patients with kCD proved the efficacy of AZA and 6-MP. The pooled odds ratio for response was 9.3 (95 % confidence interval 7.8–10.8). This meta-analysis showed that 16 weeks of administration is necessary to reach the therapeutic benefit.[5] The dose at normal metabolism of thiopurines is 2.5 mg/kg of AZA and 1-to 1.5 mg/kg of 6-MP and it can be given in a single daily dose. The addition of azathioprine to prednisolone accelerated the weaning of prednisolone in a placebo-controlled 8-week trial in active CD. Patients receiving AZA showed remission more frequently, more quickly, and with lower doses of prednisolone.

In a recent Cochrane meta analysis of seven randomized, double-blind, placebo-controlled trials of AZA and one of 6-MP both AZA and 6-MP had a positive effect on maintaining remission. Peto odds ratio for AZA was 2.32 (95 CI 1.55–3.49 and for 6-MP 3.32 (95 % CI 1.40–3.49). A steroid sparing effect of AZA was also observed, with a Peto odds ration of 5.22 (95 % CI 1.06–25.68).[6]

The beneficial effect of AZA and 6-MP was also proven in paediatric CD. Markowitz et al. randomized fifty-five children to treatment with 6-MP or placebo within 8 weeks of initial diagnosis. Children in both groups also received prednisone. Remission was reached in 89 % of both groups, but during the 18-month observation period, only 9 % of the remitters in the 6 MP group relapsed compared to 47 % of controls ($p = 0.007$). In the 6-MP group, the duration of steroid use was shorter and the cummulative steroid dose was lower at 6, 12 and 18 months. The authors concluded that 6-MP should be part of the initial treatment regimen for children with moderate to severe CD.[7] The other studies examining the effect of AZA and 6-MP were retrospective cohort or case studies where the ratio of patients in remission was about 60 %, that is much less than in the study of Markowitz et al. Administration of AZA decreased the frequency of hospitalisation and use of steroids.[8-11] The need for surgical intervention was also decreased by early introduction of AZA.[12]

Based on the above mentioned evidence, AZA or 6-MP are recommended for maintenance of remission. Their introduction is also suggested at the initial diagnosis in children with moderate to severe form of disease as steroid sparing drugs, because they are at a higher risk for complications. Their usage is especially recommended in children with growth retardation.

Ulcerative colitis

A systemic review in adults analysing 130 patients from two randomized control trials was not able to detect any benefit for thiopurines for inducing remission.[13] A Cochrane review analysing 286 adult patients from six randomised controlled trials proved that AZA was superior to placebo for the maintenance of remission in UC (OR 0.41, 95 % CI 0.24–0.70).[14] It turned out from a prospective analysis of 133 children with UC that one- year steroid-free remission rate was 49 % at 1 year after initiating thiopurine and it was not affected by starting thiopurine ≤ 3 months vs. >3 months from diagnosis.[15] The beneficial effects of AZA and 6-MP were also confirmed in paediatric UC in three retrospective studies.[8,16,17]

AZA is better in maintaining remission in UC than 5-ASA.[18] Based on these studies, the recent ECCO and ESPGHAN evidence-based consensus guidelines for the management of paediatric UC state that thiopurines are recommended for maintaining remission in children with 5-ASA intolerance or those with frequently relapsing or steroid-dependent UC. It is also declared that thiopurines are ineffective for induction of remission. Thiopurines are also indicated for maintenance treatment after inducing remission by steroids in acute severe colitis, because in such situation the likelihood of an aggressive disease course is higher.[19] After severe colitis, it is best to start azathioprine 2 weeks after discharge from the hospital until it is clear that the initial response has been sustained by the prednisone treatment.[20]

Adverse effects of thiopurines

The adverse effects of AZA and 6-MP were categorized as allergic and nonallergic reactions. The former is dose independent toxicity, while the latter are dose dependent side effects related to the metabolism of thiopurines. The most freqeuntly occurring dose-independent adverse reaction is gastrointestinal intolerance with nausea, vomiting and diarrhoea. Pancreatitis is also a relatively frequently occurring hypersensitivity reaction. Rash and arthralgias might be caused by allergic reaction to thiopurines. Dose-independent toxicities usually appear within the first days after the introduction of AZA or 6-MP. The most frequently occurring dose-dependent toxicities include myelosuppression with leukopenia and thrombocytopenia, infection, hepatotoxicity and lymphoma.[21] These nonallergic reactions can start weeks to years after the initiation of therapy.

AZA/6-MP treatment is associated with a fourfold increase in lymphoma in adults, but this risk is lower in younger adults.[22] However, in the previous years, a relatively high number of young CD patients have developed an otherwise very rare and extremely extremely aggressive hepatosplenic T-cell lymphoma (HSTCL). HSTCL was detected in nearly 40 patients; a great majority of them were males younger than 35 years. Roughly half of these patients had received thiopurines alone while the other half received anti-TNF therapy in combination with thiopurines.[23] Kirschner found that 28 % of 95 children with IBD who were receiving AZA or 6-MP experienced side effects that responded to dose reduction or ceased spontaneously, while cessation of therapy was needed in 18 % of children. Gastrointestinal symptoms occurred in 5.2 %, while pancreatitis was observed in 4.2 % of children who were receiving AZA or 6-MP. Among the dose-dependent side effects infectious complication was the most frequent, with 8.4 %.[24]

From 216 adult patients with IBD taking a thiopurine drug, 25.9 % had an adverse reaction requiring its cessation. Adverse effects included allergic reactions (25 %), liver test abnormalities (34 %), nausea/vomiting (6 %), bone marrow suppression (7 %), pancreatitis (7 %) and other (9 %). All adverse effects resolved with cessation of the drug, with a median of 7 days to resolution. Of the patients with liver test abnormalities on azathioprine, most were able to tolerate 6-mercaptopurine, however challenge with 6-mercaptopurine was not successful for most other patients.[25]

Hindorf et al. reported that adverse events at thiopurine treatment were observed in 34 % of patients and were more common in adults than in children (40 %

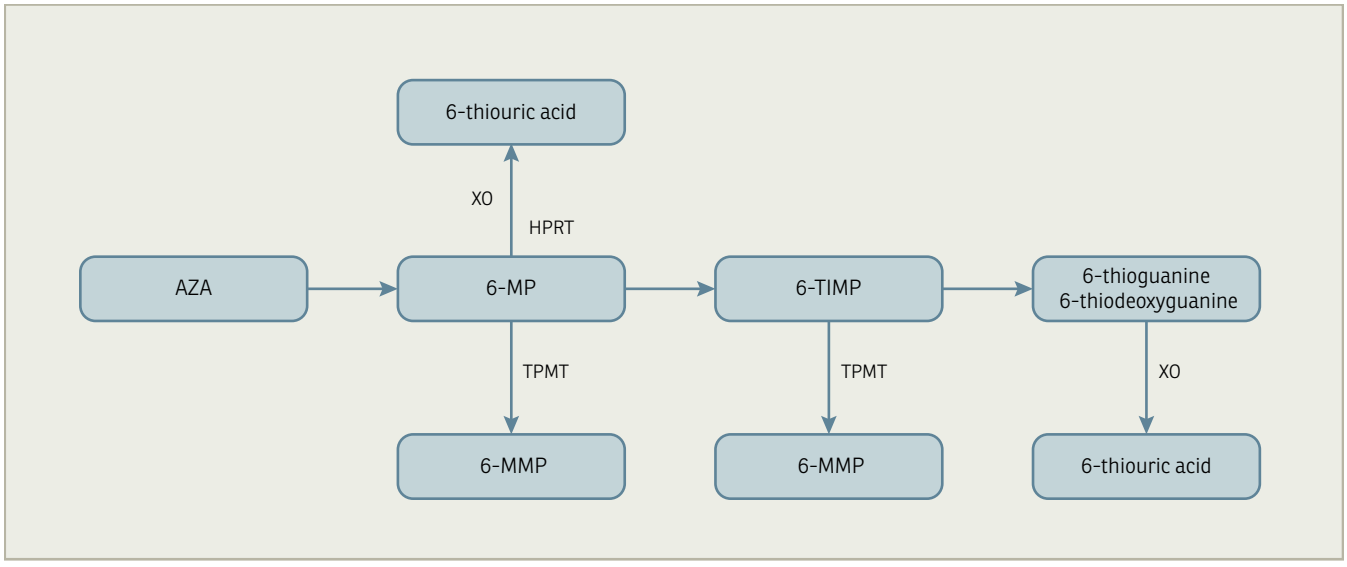

Figure 2: Three competing pathways of AZA and 6-MP metabolism by hypoxanthine phosphoribosylransferase (HPRT), thiopurine methyltransferase (TPMT) and xanthine oxidase. (XO)

vs. 15 %; $p < 0.001$) Myelotoxicity developed later than other types of adverse events.[26]

Low TMPT enzyme activity is often responsible for dose-dependent adverse effects. At low TMPT activity more 6-mercaptopurine will be metabolized to 6-thioguanine pathway and less to 6-methylmercaptopurine (6-MMP), and an increased 6-thioguanine level will cause toxic reactions, most frequently myelosuppression (Figure 2).

TPMT activity is determined by co-dominantly inherited polymorphic alleles. The wild-type allele confers high ($TPMT^H$), while the variant allele ($TPMT^L$) carries low TPMT activity. About 89 % of the population have two $TPMT^H$ alleles ($TPMT^H$/($TPMT^H$) and have high TPMT activity (> 9.5 U/ml red blood cells [RBC]), 11 % are heterozygous ($TPMT^H$/($TPMT^L$) with intermediate activity (5.0–9.5 U/ml RBC) and only 0.3 % are homozygous for the variant allele ($TPMT^l$/($TPMT^L$) having only undetectable activity (< 0.5 U/ml RBC).[27,28] An increased frequency of adverse events was observed in patients with tioguanine (thioguanine) nucleotide above 400 or methylated thioinosine monophosphate above 11450 pmol/8×10^8 red blood cells.[26] At low activity of TPMT, the 6-MMP concentration is less than 5700 pmol/8×10^8 RBC. An elevated level of 6-MMP could cause hepatotoxic effect.

Undetectable or low levels of both 6-TGN and 6-MMP indicate non-compliance or underdosing, respectively. A low level of 6-TGN and elevated 6-MMP can be caused by high activity of TPMT or low activity enzymes of the 6-thioguanine pathway. In such case, allopurinol treatment may be effective by inhibiting the xanthin oxidase pathway and transforming 6-TGN to 6-thiouracil (Figure 2.). High level of 6-TG with normal level of TPMT indicates therapy resistance, while high level of both 6-TGN and 6-MMP could be explained by overdosing or therapy resistance.[29,30]

Methotrexate

Methotrexate (MTX) is an antimetabolite as it inhibits the conversion of folic acid to its active form tetrahydrofolate, which is indispensable for thymidin synthesis. Therefore, methotrexate impairs DNA synthesis. Reducing IL-l production and inducing the apoptosis of T-cell population also contributes to its anti-inflammatory properties.

According to a Cochrane review of 3 studies in adults, methotrexate was significantly more effective than placebo for maintenance of remission in CD (OR: 3.11, CI 1.31 to 7.41, $p < 0.01$). This review also showed that there was no difference between methotrexate and 6-MP for maintenance of remission.[31] There are no paediatric randomised controlled studies. Cohort studies suggest that in children who are not able to tolerate thiopurines, methotrexate can maintain remission or response in 50–80 % of patients.[32-34]

MTX is indicated for maintenance of remission in children with CD.

In a retrospective study of children with UC, remission was achieved in 31, 28 and 28 of patients at 3, 6 and 12 months, respectively.[35] According to the joint ECCO and ESPGHAN guidelines, the presently available evidence is insufficient to recommend the use of methotrexate in paediatric UC.[19]

Methotrexate is usually given in subcutaneous injection at a dose of 15 mg/m2 once a week. The maximal dose cannot be over 25 mg.[31] Concurrent administration of folate supplementation is recommended. Nausea following MTX is the most common side effect in children with CD. This adverse effect may be prevented through the use of a short-course ondansetron as premedication.[36] Asymptomatic increases in liver enzymes may also occur. Hypersensitivity pneumonitis is a rarely occurring severe complication of MTX treatment.[37]

Thalidomide

Thalidomide is a glutamic acid derivative (alpha-phthalimidoglutaramide), which contains in S and R isomers in the 1:1 ratio. The immunomodulatory effects are related to the S isomers while the R isomer has sedative effects, which explains its widespread use to treat morning sicknessin pregnant women. In the early sixties, this drug was withdrawn because it turned out that it caused serious congenital birth defects characterized by phacomelia or amelia.

The immunomodulatory action of thalidomide includes the inhibition of TNF-α synthesis and its release from activated monocytes and decrease in nuclear factor-κB activation, as well as switching the cytokine production from T_H1 to T_H2 profile.[38] It was reported in two recent studies that thalidomide was effective where conventional immunosuppression and biological therapy were ineffective or caused adverse reaction. Zheng et al. examined the efficacy and tolerability of thalidomid in 6 children with refractory CD, and they observed in all patients a remission within three months without significant adverse reaction.[39] Felipez et al. found that with thalidomide treatment in 12 children with severe refractory CD the steroid need significantly decreased, and out of 7 patients with fistulae, 5 had complete and 1 had partial closure. Peripheral neuropathy developed in 5 children but all of them had clinical resolution of neurological symptoms within 2–3 months after stopping thalidomide.[40] Thalidomide was less effective in UC than in CD (44 % vs. 89 % of remission).[41]

The recommended dose of thalidomide is 1.5–2.5 mg/kg/day in a single dose in the evening. Neurological examination including vibration sensitivity at regular intervals is indicated. Thalidomide is absolutely contraindicated during pregnancy; contraception is mandatory when appropriate.

Calcineurin inhibitors

Both cyclosporine and tacrolimus are calcineurin inhibitors. Although they bind to different target molecules, both drugs inhibit T-cell activation similarly. Cyclosporine binds to cyclophyllin and the formed complex binds and blocks the function of calcineurin enzyme. Tacrolimus binds to FK506 binding proteins and this complex also inhibits calcineurin. In both cases the inhibited calcineurin is not able to dephosphorylate the cytoplasmic component of the nuclear factor of activated T-cells (NF--ATc); so the transport of NF-ATc to the nucleus will not be possible and consequently, T cells do not produce IL-2, which is necessary for full T-cell activation.[42]

Cyclosporine was not better than conventional treatment in newly diagnosed children with CD.[43] Cyclosporine can be administered as a second-line rescue therapy in children with acute severe colitis in a continuous intravenous infusion of 2 mg/kg/day. After remission it has to be converted to oral 5–8 mg/kg/day b.i.d. Analysing data of 94 children from eight retrospective case series, the pooled short term response was 81 % (95 % CI 76–86 %), but only 39 % (CI 29–49 %) avoided colectomy in the long term.[44] Cyclosporine may cause potentially serious side effects, such as nephrotoxicity, severe infections, seizures, paresthe-

sia, hypertension, hypomagnesaemia and hyperkalaemia.

In 24 children with acute severe UC in three small paediatric case series, 16 (67 %) of them showed good initial response, but long-term colectomy free period occurred only in 0–22 % of included patients. The recommended oral dose of tacrolimus is 0.1 mg/kg/dose orally, twice daily. According to the recent ESPGHAN and ECCO guidelines, cyclosporine or tacrolimus started during an episode of acute severe colitis should be discontinued after 4-month bridging to thiopurines because of their many side effects. Azathioprin should be introduced two weeks after the initial response to calcineurin inhibitors. The therapeutic effect of thiopurines is usually reached 10 to 14 weeks after introduction.[19] Infliximab was a much more effective second-line rescue therapy than cyclosporine or tacrolimus.[44]

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