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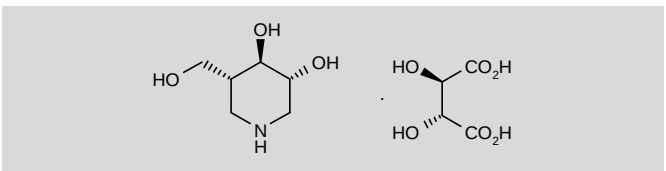
ISOFAGOMINE TARTRATE

Glycogen Phosphorylase Inhibitor
Treatment of Gaucher's Disease

AT-2101
HGT-3410
NN-42-1007
Plicera™

(3*R*,4*R*,5*R*)-3,4-Dihydroxy-5-(hydroxymethyl)piperidine L-tartrate

InChI=1/C6H13NO3.C4H6O6/c8-3-4-1-7-2-5(9)6(4)10;5-1(3(7)8)2(6)4(9)10/h4-10H,1-3H2;1-2,5-6H,(H,7,8)(H,9,10)/t4-,5-,6-;1-,2-/m11/s1



$C_{10}H_{19}NO_9$
Mol wt: 297.2592
CAS: 919364-56-0
CAS: 161302-93-8 (hydrochloride)
CAS: 169105-89-9 (free base)
EN: 228804

ABSTRACT

Isofagomine tartrate (AT-2101, HGT-3410, Plicera™) is a new therapeutic candidate for the treatment of Gaucher's disease, the most common of the lysosomal storage syndromes, which is characterized by genetic mutations that result in the production of a defective key enzyme, β-glucocerebrosidase. This leads to accumulation of the fatty substance glucocerebroside in the spleen, liver, kidneys, lungs, brain and bone marrow, which manifests as severe clinical symptoms. Isofagomine is designed to act as a pharmacological chaperone by selectively binding to misfolded β-glucocerebrosidase and helping it to fold correctly, to restore its activity. This new agent is currently undergoing phase II clinical development at Amicus Therapeutics in collaboration with Shire Human Genetic Therapies, a business unit of Shire.

SYNTHESIS

Isofagomine can be synthesized by several different methods starting from a number of different compounds (1-20). Due to space limitations, the numerous methods and schemes of synthesis for isofagomine have not been included. Subscribers to Integrity® can access the schemes.

BACKGROUND

Gaucher's disease is the most common of the lysosomal storage syndromes, rare inherited metabolic disorders that result from defects in lysosomal function. It is an autosomal recessive disease caused by genetic mutations that result in the production of misfolded β-glucocerebrosidase. This enzyme is responsible for the breakdown of glucocerebroside, a specialized fat molecule, to ceramide and glucose in the lysosome (21, 22). Absent or defective β-glucocerebrosidase enzyme activity leads to build-up of glucocerebroside inside certain cells, which can, over time, cause inflammation or damage to specific areas within the body, including the liver, spleen, bone marrow, lung and the central nervous system (23).

Three clinical subtypes of Gaucher's disease have been described. Type I is non-neuropathic and the most common form. It is prevalent in individuals of Ashkenazi Jewish descent (N370S missense mutation; with L444P representing the most frequent mutation in the Western hemisphere) (23). Type II is an acute infantile neuropathic form that typically occurs within 6 months of birth. Type III is a chronic neuropathic form that can occur during childhood or adulthood.

The only approved treatment options for Gaucher's disease are enzyme replacement therapy (ERT) and substrate reduction therapy (SRT) (23); however these therapies are associated with significant side effects. A new therapeutic option for this syndrome is under development by Amicus Therapeutics in conjunction with Shire Human Genetic Therapies: the iminosugar isofagomine (AT-2101, HTG-3410, Plicera™). Isofagomine has completed a U.S. multicenter phase II study in patients with type I Gaucher's disease already receiving ERT (24), along with another clinical study to characterize the ex vivo response to therapy by testing blood samples from previously treated and untreated patients with Gaucher's disease (25). An additional multicenter phase II study is currently recruiting in the U.S., the U.K., Germany and Israel to assess the safety and efficacy of isofagomine in patients with type I Gaucher's disease who are not

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receiving ERT or SRT (26), and will be followed by a long-term extension study (27).

PRECLINICAL PHARMACOLOGY

In vitro studies have evaluated the mechanism of action of isofagomine in N370S fibroblasts. Data from these studies demonstrate that isofagomine acts as a pharmacological chaperone by selectively binding to misfolded β-glucocerebrosidase, increasing enzyme activity by about 3-fold via several mechanisms. After binding to the enzyme, it is thought that isofagomine promotes the proper folding, processing and trafficking of the enzyme from the endoplasmic reticulum to the lysosome at neutral pH. Once it reaches the lysosome, the pharmacological chaperone is displaced and the enzyme can perform its normal function (28-30).

Further experiments in patient skin fibroblasts have also confirmed that isofagomine enhances the activity of N370S β-glucocerebrosidase (2.3- to 3.0-fold) in all patient samples tested, without significantly affecting the growth of wild-type human fibroblasts or normal human lymphoblasts. Studies in Caco-2 intestinal epithelial cells also demonstrated that isofagomine is a much weaker inhibitor of the intestinal disaccharidase enzymes sucrase and isomaltase compared with another iminosugar candidate for Gaucher's disease, miglustat. Moreover, isofagomine has little or no inhibitory activity towards endoplasmic reticulum α-glucosidase II or glucosylceramide synthase at concentrations previously shown to enhance N370S β-glucocerebrosidase folding and trafficking in Gaucher's fibroblasts (31).

Investigations in a knock-in mouse model expressing murine L444P β-glucocerebrosidase have shown that oral administration of isofagomine results in a dose-dependent increase in β-glucocerebrosidase levels (2- to 5-fold) in liver, lung, spleen, skin and, importantly, brain, with a minimum effective dose of 3 mg/kg. Isofagomine-mediated increases in L444P β-glucocerebrosidase levels were selective, as the activities of the lysosomal hydrolases α-galactosidase A, α-glucosidase, β-glucuronidase and β-galactosidase were not altered by isofagomine treatment in any tissue examined. Isofagomine also lowered plasma IgG (15%) and chitin III (33%) levels and treatment for 3-6 months significantly decreased spleen (22%) and liver (20%) weights in these mice (29, 32).

CLINICAL STUDIES

In a randomized, double-blind phase I clinical trial in 72 healthy volunteers, multiple and single ascending doses of 8, 25, 75, 150 and 300 mg isofagomine were well tolerated, with mostly mild treatment-emergent adverse events. Oral dosing provided good systemic exposure: plasma AUC and C_{max} values were linearly correlated in both single- and multiple-dose studies, with mean plasma levels peaking at 3.4 h and a reported elimination half-life of 14 h. In the multiple-dose study, β-glucocerebrosidase activity in isolated white blood cells showed a marked increase at days 1, 3, 5 and 7 during administration of isofagomine (29, 33).

An ex vivo response study using macrophages and lymphoblasts derived from Gaucher's patients with different genotypes (N = 52; type I Gaucher's disease) has shown that isofagomine treatment elevates β-glucocerebrosidase levels (mean 2.6-fold), as well as levels of biomarkers associated with inflammation, bone metabolism, multiple myeloma and neurodegeneration (34, 35).

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Amicus Therapeutics, Inc. (US); being developed in collaboration with Shire Human Genetic Therapies, a business unit of Shire.

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