An Innate Defense Regulator for the Treatment of Macrophage Activation Syndrome: Preclinical Studies in an Orphan Indication

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ABSTRACT

Objective: Macrophage activation syndrome (MAS) is a rare, orphan disease and potentially fatal complication of certain childhood rheumatological disorders related to the immune system. Using an established mouse model, we assess the efficacy of a novel immune modulator, dusquetide, in the treatment of MAS.

Method: The specific cause of MAS is unknown. Using a mouse model where MAS is induced by repeated TLR-9 stimulation yielding symptoms similar to the clinical syndrome, the impact of dusquetide treatment on pancytopenia and downstream inflammatory signaling is tested.

Results: MAS was simulated in 8–10 week old C57BL/6 mice by repeated administration of the TLR-9 agonist, CpG. CpG (35 µg in 200 µL) was administered intraperitoneally (IP) on Days 0, 2, 4, 7 and 9. Dusquetide (200 mg/kg IP) or saline was administered on Days 1, 4 and 7. Mice were observed for complete blood counts on Day 8, and body weight and serum cytokines (IFNγ, IL-12 and IL-10) on Day 10. Dusquetide significantly increased white blood cell and platelet counts on Day 8 relative to the placebo group. On Day 10, both decreased IL-12 levels, while increased body weights were observed in the dusquetide-treated group relative to the placebo group. IFNγ and IL-10 levels were not significantly altered, in keeping with the general understanding of the mechanism of action of Innate Defense Regulators (IDRs). The results of these studies established the medically plausible basis for the use of dusquetide as a treatment for MAS allowing for the FDA to grant dusquetide Orphan Drug Designation for this rare condition.

Clinical studies of dusquetide in both Phase 1 and Phase 2 clinical studies also confirm the transferability of the anti-inflammatory activity to the human clinical setting, suggesting that the results obtained in mice may be transferred to humans. Particularly in Phase 1 studies, blood collected from healthy human volunteers treated with either placebo or dusquetide was stimulated with endotoxin for 4 hours. Blood from subjects given dusquetide had a more anti-inflammatory profile than blood from placebo patients. Similarly in a double-blind, randomized, placebo-controlled Phase 2 trial evaluating dusquetide for the treatment of oral mucositis (OM), an inflammatory condition associated with the dysregulation of the innate immune system, dusquetide was shown to decrease the duration of severe OM. Interestingly, a decrease in incidence of non-fungal (i.e., bacterial) infection was also observed, consistent with preclinical studies.

Conclusions: MAS is characterized by a cytokine storm that results in a sepsis-like syndrome including pancytopenia, systemic inflammation and fever. MAS is fatal in 10-20% of cases. Dusquetide reduces pancytopenia in an established mouse model of MAS utilizing repeated TLR-9 stimulation with CpG, in addition to decreasing IL-12 levels and improving body weight maintenance. The innate immune modulating activity of dusquetide has also been observed in Phase 1 (healthy human volunteer) and Phase 2 (OM in head and neck cancer patients undergoing chemoradiation therapy) studies, proving that findings in mice are transferable to the human clinical setting. Further studies, optimizing the route of administration and testing dusquetide in other animal models of MAS, are warranted on the basis of these results.

MAS

MAS occurs in patients with autoimmune disease and is a hyperinflammatory response characterized by pancytopenia, liver insufficiency, coagulopathy and neurologic symptoms. It is thought to be caused by the activation and uncontrolled proliferation of T lymphocytes and well-differentiated macrophages, leading to widespread hemophagocytosis and continued cytokine overproduction.

MAS is more common (~10-30% incidence) in children diagnosed with systemic juvenile idiopathic arthritis (sJIA); although it is also a known complication of autoimmune diseases such as systemic lupus erythematosus (SLE), Kawasaki's disease, and Adult-onset Still's disease.

Despite aggressive treatment, mortality varies from 8 to 30% and morbidity is also very high, with approximately 33% of patients requiring admission to the intensive care unit (ICU).

There is no approved treatment for MAS.

METHODS

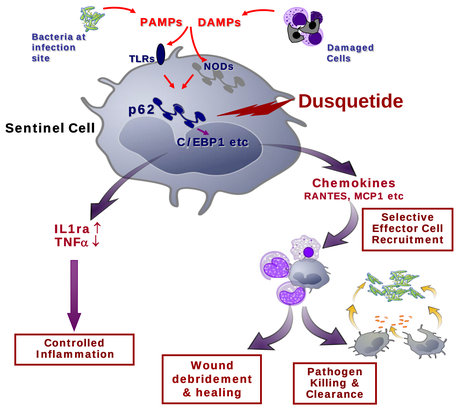
Innate Defense Regulators: Dusquetide

Dusquetide is a first in class compound with a novel mechanism of action, targeting an intracellular convergence point in the innate immune system, downstream of most inputs and upstream of most outputs of the innate immune system (1, 2, 3). Dusquetide modulates the response of the innate immune system to a broad spectrum of triggers including infection, tissue damage and secondary inflammation.

Dusquetide is a 5-amino acid peptide with rapid pharmacokinetics and enduring pharmacodynamic responses.

Dusquetide is administered systemically as a solution. Clinically it is administered as a 4-minute intravenous (IV) injection. In animal models it can be administered IV, IP, intramuscularly or subcutaneously.

Dusquetide binds to the p62 (sequestosome-1) protein, influencing protein-protein interactions and modulating downstream pathways.



MAS Preclinical Model

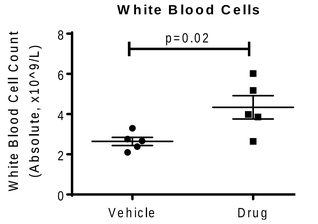
Repeated TLR-9 stimulation with CpG that has been utilized extensively by Dr. Behrens at the Children's Hospital of Philadelphia (CHOP) (4, 5).

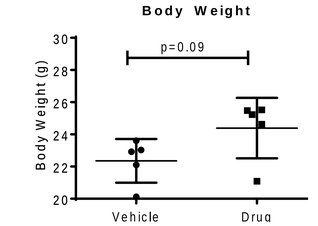
IP injection of 35 µg (in 200µL) CpG is administered to C57BL/6 mice on days 0, 2, 4, 7 and 9. Blood counts are evaluated on Day 8 and all animals are euthanized on Day 10. Key endpoints include white blood cell and platelet counts, spleen weights and body weight. Spleen and liver were also preserved and evaluated in a blinded manner.

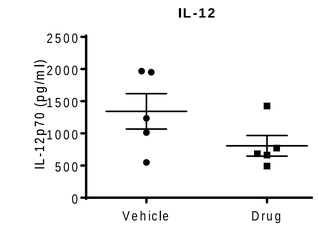
RESULTS

Dusquetide Reduces Inflammation and Pancytopenia in MAS Mouse Model

Dusquetide (200 mg/kg IP) or saline was administered on Days 1, 4 and 7. Mice were observed for complete blood counts (Day 8) and body weight, serum cytokines (not shown) on Day 10. Dusquetide significantly increased white blood cell and platelet counts on Day 8 relative to the placebo group. On Day 10, both decreased IL-12 levels and increased body weights were observed in the dusquetide treated group relative to the placebo group. IFNγ and IL-10 levels were not significantly altered, in keeping with the general understanding of the IDR mechanism of action (1, 2, 3). Each dot represents the result from one mouse.

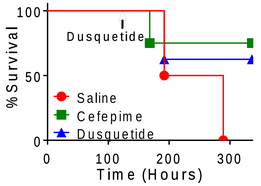






RESULTS

Dusquetide Protects against Sepsis-like Syndrome in Leukopenic Animals



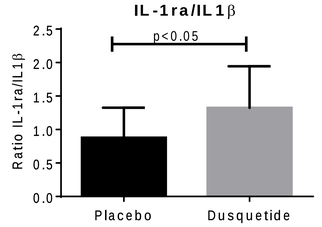
Dusquetide, administered after fever, reduced subsequent sepsis and death.

Female Sprague-Dawley rats were administered 10 mg/kg Cefamandole IM Q48h starting on day -4 and continuing to the end of the study. Cyclophosphamide 75 mg/kg IP was administered on days 0 and 3.1x106 CFU/ml P. aeruginosa was given by orogastric feedings on days 0, 2 and 4. Rats developed fever by day 5, and began dying of sepsis by day 7. Dusquetide (10 mg/kg IV) or Saline (IV) was administered once after the appearance of fever on day 5. Cefepime was administered on days 6, 7 and 8 (25 mg/kg IM). Survival was monitored in an initial study (N=8 IMX942, Saline, N=4 Cefepime).

Dusquetide is Anti-Inflammatory in Healthy Volunteers

Dusquetide (SGX942) was evaluated in a double-blind, placebo-control, randomized Phase 1 trial in 84 healthy human volunteers, including both a single ascending dose (0.15-8.0 mg/kg) and multiple ascending dose (0.5-6.5 mg/kg daily for 7 days) phase. Dusquetide was well tolerated with no serious or severe adverse events, no dose limiting toxicity and no maximum tolerated dose identified.

Blood taken from the healthy volunteers 1-hour post-dose was further stimulated with LPS (endotoxin). Subjects receiving low dose dusquetide were found to have an enhanced anti-inflammatory response with increased IL-1ra and TNFR II (anti-inflammatory) and decreased IL-1β and TNFα (inflammatory). Higher doses of dusquetide looked like placebo, illustrating a non-linear dose response that was also observed in preclinical studies.

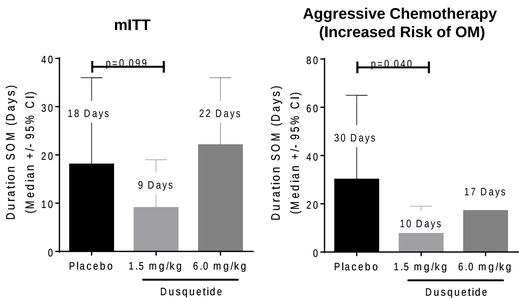


RESULTS

Dusquetide is Anti-Inflammatory in Cancer Patients

Dusquetide (SGX942) was evaluated in a double-blind, placebo-control, randomized Phase 2 trial in 111 head and neck cancer patients receiving chemoradiation therapy and at risk for OM. OM is caused by an excessive inflammatory response of the innate immune system in response to chemoradiation-induced tissue damage. Placebo or dusquetide (1.5 or 6.0 mg/kg) was given twice weekly during chemoradiation.

Dusquetide (1.5 mg/kg) administration resulted in a 50% decrease in the duration of severe OM (SOM) and a concomitant decrease in the rate of non-fungal (bacterial) infection (not shown).



CONCLUSIONS

- MAS is a rare and serious inflammatory complication in patients with auto-immune diseases and is associated with both significant mortality and high morbidity.
- Dusquetide is an Innate Defense Regulator (IDR) which modulates the response of the innate immune response to a broad spectrum of innate immune activators including infection and tissue damage.
  - Dusquetide increases clearance of infection and accelerates tissue healing
  - Dusquetide modulates inflammation
  - Dusquetide has proven efficacy in sepsis-like models
- Dusquetide improves outcome in one preclinical model of MAS, including reduced pancytopenia, decreased inflammation and improved body weight maintenance.
  - Dusquetide has Orphan Drug designation in MAS

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