AST-008, a TL9 Agonist Spherical Nucleic Acid, Activated NK Cells, T Cells and Cytokines in Healthy Subjects in a Phase 1 Clinical Trial
Weston L Daniel, Ulrike Lorch, Simon Coates, Alice S. Bexon, and Scott Mix

**BACKGROUND**

- AST-008 is a toll-like receptor 9 (TLR9) agonist oligonucleotide in a proprietary spherical nucleic acid (SNA) format with immune-stimulatory properties.
- SNAs are dense, radial arrangements of nucleic acids that have useful properties as compared to linear oligonucleotides (i.e., oligonucleotides not in the SNA format), notably increased cellular uptake and an optimal presentation of the oligonucleotides for TL9 agonism.
- AST-008 is designed to enter into and activate immune cells to elicit an immune response to treat solid tumors in combination with a checkpoint inhibitor.
- AST-008 has potent antitumor activity as a monotherapy and synergizes with anti-PD-1 antibody therapy in several preclinical tumor models.

**OBJECTIVES**

- **Primary**
  - To evaluate the safety and tolerability of AST-008 after single subcutaneous (SC) doses
- **Secondary**
  - To recommend a dose and regimen for further development
  - To determine the pharmacodynamics (PD) of AST-008 after SC doses
  - To determine the effect of AST-008 on QTc interval

**METHODS**

- AST-008 was evaluated in a Phase 1 study under protocol AST-008-101.
- Four dose levels of AST-008 were evaluated in four cohorts. Each cohort included four volunteers, and all received a single dose of AST-008. The dose levels were 5, 10, 12.5, and 18.8 µg/kg.
- Peripheral blood cytokine concentrations and cell activation were measured with a Randox chromatography method and fluorescence-based detection.
- Plasma concentrations of AST-008 were assessed with a solid-phase hybridization assay with electrochemiluminescent detection; urine concentrations were assessed with a liquid chromatography method and fluorescence-based detection.

**POPULATION**

- Healthy volunteers age 18 to 40 with body mass index of 18 to 25 kg/m².
- Subjects with significant medical history or significant abnormalities, a recent history of tobacco, drugs of abuse, prescription medications including corticosteroids or other immunosuppressive drugs, or other investigational products were excluded.

**ADVERSE EVENTS**

- No serious adverse events or dose-limiting toxicity were observed.
- Non-serious AEs: nasopharyngitis, appetite loss, headache, and pyrexia.

**CYTOKINE EXPRESSION AND LYMPHOCYTE ACTIVATION**

- AST-008 Activated Natural Killer (NK) and T Cells at All Dose Levels Tested
- One line represents one subject

**CONCLUSIONS**

- AST-008 was well tolerated and elicited no serious adverse events or dose-limiting toxicity at the doses tested.
- AST-008 is a potent innate immune activator and exhibits pharmacodynamic properties that are expected to result in anti-tumor effects in patients with cancer.
- AST-008 plasma exposure was roughly dose proportional; peak AST-008 plasma concentrations were observed before peak pharmacodynamic effects. AST-008 was not detected in urine.
- Phase 1b/2 study of intratumorally-dosed AST-008 in combination with pembrolizumab in cancer patients is ongoing. Non-melanoma skin cancers are of particular interest in this study.