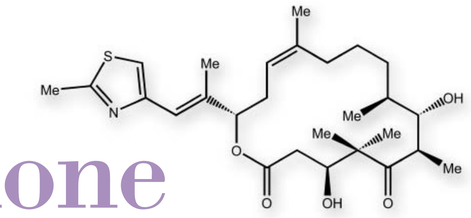


12,13-Desoxyepothilone

SUCCESS STORY

NSC 703147.....RAID II BEGUN JUNE 1999, COMPLETED APRIL 2000.....RAID IV BEGUN JULY 2000, COMPLETED APRIL 2002.....CLINICAL TRIAL OCTOBER 2001

RAID Projects with Dr. Samuel Danishefsky, Memorial Sloan-Kettering Cancer Center

The Epothilones

Fermentation Products Known Since the 1990s to Have Anticancer Activity

DTP provided the following:

- First confirmation of microtubule-based mechanism of action of epothilones, with epothilone B being especially active.
- First kinetic demonstration that epothilones A and B are competitive inhibitors of paclitaxel binding to tubulin.
- First demonstration that a paclitaxel-resistant cell line with a tubulin mutation retains sensitivity to epothilones and first confirmation that MDR cells retain sensitivity to epothilones.
- SAR evaluation of epothilones showing that desoxyepothilone B is highly active in cells and with tubulin, but that a second paclitaxel-resistant cell line with another tubulin mutation is also resistant to epothilones.
- First demonstration that tubulin assembly induced by epothilone B is inhibited by drugs that inhibit tubulin assembly.

First RAID Application Received from Dr. Danishefsky 02/1999

Applicant prepared a number of analogs related to the epothilones A and B. Included in these analogs was 12,13-desoxyepothilone B, which was found to be much less toxic than epothilone B itself. In *in vivo* studies, the desoxy compound was well tolerated and virtually curative against a variety of tumors, including some resistant to paclitaxel.

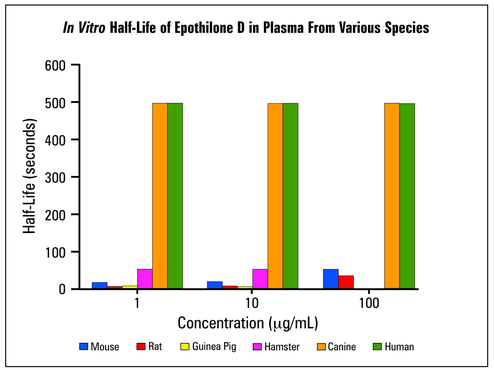
RAID Application Approved and Initiated 06/1999

- A 5 g pilot batch of material was produced and analyzed.
- A 40- to 50-step synthesis was required.
- Material was returned to Dr. Danishefsky for testing.

First RAID Application Completed and Second Application Received 02/2000

Pharmacology Studies Initiated 07/2000

- Species-dependent *in vitro* metabolism with plasma from various species: rat > mouse >>> dog ≈ human.
- Similar pattern with liver S9 fractions.
- *In vitro* data predictive of *in vivo* half-life:
  mouse = 3.6 minutes
  rat = 19 minutes
  dog = 476–2634 minutes



KOSAN Biosciences Licenses and Collaborates with DTP 03/2001

Two Different Formulations Studied 03/2001

Two proposed clinical formulations are:
1. 10 mg/mL epothilone D in Diluent 12.
2. 10 mg/mL epothilone D in cremophor: propylene glycol (PG): ethanol (20:30:50) by volume.

Each formulation was further diluted to 1 mg/mL with NS and D5W and stored in glass at laboratory ambient temperature under white light for 48 hours.

Particle size distribution of each sample solution was measured, and the physical appearance of each sample solution was observed at time zero, 4, 8, 24, and 48 hours.

There was no significant change in physical stability of the diluted infusion solutions as tested by particle size.

Range-Finding and IND-Directed Toxicology Studies Conducted 09/2000

- Toxicology studies were conducted in dogs given a 4-hour continuous i.v. infusion.
- The maximum tolerated dose (MTD) in the dog when given as a 4-hour continuous i.v. infusion is 110 mg/m² as a single dose and >110 mg/m²/dose weekly x 3.
- Gastrointestinal and bone marrow toxicity were dose limiting.
- The recommended clinical starting dose was 11 mg/m²/dose weekly x 3 (i.e., one-tenth the maximum tolerated dose given weekly x 3 in the dog).

Material Made Available for Clinical Trials

- DTP contractor produced 25 g of material.
- KOSAN developed a biological production system.

12,13-Desoxyepothilone First Tested in Man 10/2001

Currently Entering Phase II Clinical Trials