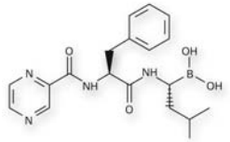
Velcade (PS-341)

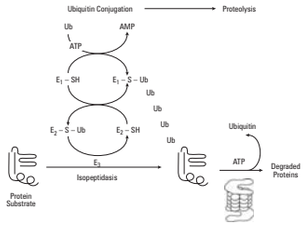


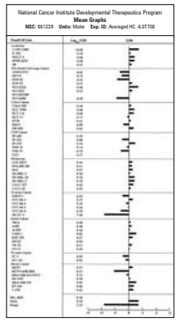
SUCCESS STORY

NSC 681239.....RECEIVED JULY 1995.....DN2A JANUARY 1997.....DN2B/III JUNE 1998.....CLINICAL TRIAL JULY 1999.....NDA MAY 2004

Background

- Boronic acid dipeptide discovered by Proscript, Inc., Cambridge, MA (since acquired by Millennium Pharmaceuticals).
- A series of 19 boronic acid dipeptides were submitted to DTP for evaluation in 1995.
- A first-in-class inhibitor of the 20S segment of the proteasome, the cellular component that regulates the degradation of many cell cycle control proteins.

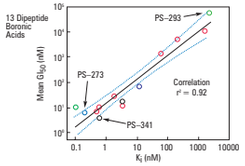




In Vitro Studies

- Velcade® is highly potent against all cell lines with an average GI50 of 17.8 mM.
- Potency of growth inhibition in the DTP cell line screen correlated with 20S proteasome inhibitory potency.

Correlation of Tumor Growth Inhibition with 20S Proteasome Inhibitory Potency



Adams, et al., Cancer Res 59:2615, 1999

In Vivo Studies

Hollow Fiber (HF) Testing

- Analogs that exhibited potency in the DTP cell line screen were evaluated in the HF assay.
- All compounds met or exceeded criteria for activity.

(total score >= 20, SC score >= 8, or cell kill in any cell line)

NSC Number	IP	SC	Total
681226	36	14	50*
681228	38	4	42*
681229	20	12	32
681230	44	12	56*
681231	6	2	8*
681232	24	6	30
681234	14	8	22
681236	28	4	32*
681237	14	6	20*
681239 (PS-341)	22	6	28*

*Indicates agent reached cell kill in one or more cell lines.

Tumor Studies

- Velcade® decreased the number of lung metastases in a Lewis Lung mouse model when combined with standard chemotherapeutic agents.
- The number and size of lung metastases in an animal bearing the Lewis Lung carcinoma after treatment with Velcade® alone or with an anticancer agent can be seen below.

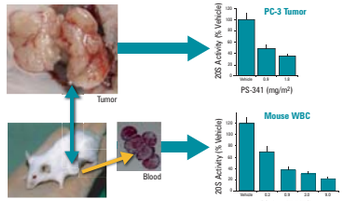
Lung Metastases on Day 20
Number (% Large)

PO Treatment Group	Alone	+ Velcade® (0.1 mg/kg)	+ Velcade® (0.03 mg/kg)
Control	33 (45)		
Velcade® p.o., D4-18		15 (60)	18 (53)
5-Fluorouracil (30 mg/kg), i.p., D7-11	5.5 (45)	1.5 (0)	2.5 (0)
Cisplatin (10 mg/kg), i.p., D7	22 (45)	9.5 (58)	12 (49)
Taxol® (24 mg/kg), i.v., D7-11	23 (30)	13 (46)	14 (46)
Adriamycin (1.75 mg/kg), i.p., D7-11	18 (36)	6.5 (37)	12.5 (33)

Pharmacokinetic (PK) and Pharmacodynamic (PD) Studies

- Velcade® is rapidly cleared when administered i.v. and is unmeasurable in plasma.
- However, its effect on the 20S proteasome can be monitored using an assay in WBCs.
- Velcade® exhibits a dose-dependent and reversible inhibition.

Velcade®: 20S Inhibition

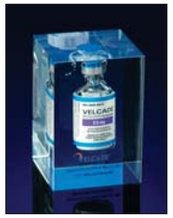


Is the Safe Dose in Animals in the Efficacy Range for Humans?

Species	Dose (mg/kg)	Dose (mg/m²)	% 20S Proteasome Inhibition*
Mouse	1.0	3.0	80
Rat	0.25	1.5	80
Non-human primate (NHP)	0.067	0.8	70
Man	0.0053	1.96	80

*In white blood cells at 1 hour post dose.

Toxicology Studies



Rat Studies

- Dose range 0.1 to 0.25 mg/kg given twice weekly for 2 weeks.
- 0.25 mg/kg dose lethal to 1/10 rats on day 2.
- No other clinical signs of toxicity noted.

NHP Studies

- Dose range 0.045 to 0.1 mg/kg/dose given twice weekly for 4 weeks.
- Highest non-toxic dose (no severe or irreversible toxicities) was 0.067 mg/kg/dose.
- The LD_{10} was 0.1 mg/kg/dose.
- Velcade® has a very narrow safe dosing range.
- Toxicities include diarrhea, vomiting, and anorexia.
- Determination of 20S proteasome activity 1 hour post dosing at 0.067 mg/kg shows a 70% decrease in activity.

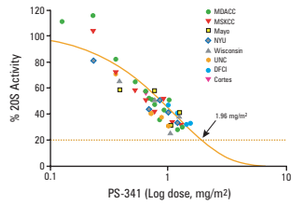
Formulation and Clinical Batch Preparation

- ProScript, Inc., the sponsor, provided NCI with a liquid formulation.
- When stored at 2–8°C, the liquid formulation was not stable for longer than 6 months.
- PRB/DTP developed a lyophilized formulation that was very stable.
- The lyophilized product was stored at 5°C, ambient temperature, 37°C, and 50°C. Stability was monitored for approximately 18 months. Over this time period, there was no loss of drug in the lyophilized product stored at any temperature and no evidence of degradation product peaks in the HPLC chromatograms.
- This formulation consists of 2.5 mg drug and 25 mg of mannitol, a commonly used pharmaceutical excipient. This formulation can be easily reconstituted into an aqueous solution at the time of administration.
- The commercial product, known as Velcade®, is the formulation developed by DTP.

Clinical Trial Experience

- Based on toxicity studies, the starting dose for phase I trials was 0.13 mg/m²/dose (1/6 the maximum safe dose in primates).
- The goals of use of the *ex vivo* 20S proteasome measurements are to:
  - — Confirm inhibition of the biological target;
  - — Use the pharmacodynamic (PD) end point in lieu of blood drug levels;
  - — PD will guide dose escalation and provide dose escalation stopping point; and
  - — Evaluate relationship of 20S inhibition with toxicity, PK, and activity.
- Clinical data.

Proteasome Activity in WBCs: 1 Hour Post Treatment



- Graph shows results for 137 treated patients.
- Dose-related inhibition of the proteasome was observed at 1 hour.
- Target effective dose (TED) is approximately 1.6–1.9 mg/m².
- Maximum proteasome inhibition is 72%.
- Toxicities: grade 3 diarrhea, fatigue (generally well tolerated).
- Major responses observed in multiple myeloma (as of 2002).

An NDA Was Approved for Velcade® for Treatment of Multiple Myeloma in May 2004

Developmental Therapeutics Program of the National Cancer Institute

TURNING MOLECULES INTO medicines FOR THE public health