CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

125359Orig1s000

SUMMARY REVIEW

Division Director Summary Review

Date	November 15, 2011
From	Patricia Keegan
Subject	Division Director Summary Review
BLA #	BL STN 125359/0
Applicant Name	EUSA Pharma (USA), Inc.
Date of Submission	October 31, 2010 (received November 1, 2010)
PDUFA Goal Date	August 2, 2011
Proprietary Name / Proper Name	Erwinaze asparaginase *Erwinia chrysanthemi*
Dosage Forms / Strength	Lyophilized powder supplied in single vials/ 10,000 International Units asparaginase *Erwinia chrysanthemi* per vial
Proposed Indication	"ERWINAZE (Erwinia L-asparaginase) is indicated as a component of a multi agent chemotherapeutic regimen for the treatment of patients with: • Acute lymphoblastic leukemia (ALL) who have developed hypersensitivity to (b) (4) E. coli derived asparaginase (b) (4)
Recommended Action:	Approval

Material Reviewed/Consulted OND Action Package, including:	**Names of discipline reviewers**
Regulatory Project Manager Review	Erik Laughner
Medical Officer Review	Patricia Dinndorf
Pharmacology Toxicology Reviews	Dubravka Kufrin Anne M. Pilaro (Supervisory Review)
OBP Reviews (CMC)	Nikolay Spiridonov; Cristina Ausín-Moreno (DS), Jacek Cieslak and Serge Beaucage (DP); Faruk Sheikh (immunogenicity)
Facilities Review	Mary Farbman (DS), Anastasia Lolas & Halavati Suvarna (DP)
Clinical Pharmacology Reviews	Jun Yang
OSI/DBGC Reviews	Michael Skelly
DDMAC	Carole Broadnax
DSI	Robert Young
CDTL Review	Suzanne Demko
OSE/DMEPA	Loretta Holmes
OBP Project Manager/Labeling Review	Kim Rains

OND=Office of New Drugs
OBP=Office of Biotechnology Products
CMC=Chemistry, Manufacturing, and Controls
OSI=Office of Scientific Investigations
DBGC=Division of Bioequivalence and GLP Compliance
DDMAC=Division of Drug Marketing, Advertising and Communication
OSE= Office of Surveillance and Epidemiology
DMEPA=Division of Medication Error Prevention and Analysis
CDTL=Cross-Discipline Team Leader

Division Director Summary Review

1. Introduction

The initial investigational new drug application (IND 290) for *Erwinia* asparaginase was submitted to FDA on January 23, 1968. In the decades since the IND submission, *Erwinia* asparaginase has become widely used in the United States, primarily under clinical trials conducted by NCI-funded cooperative groups as well as in single patient use under expanded access programs, first under the "Group C" mechanism managed by the National Cancer Institute and more recently through an access protocol sponsored by EUSA Pharma, Inc. The clinical use of *Erwinia* asparaginase has been limited to replacement of *E. coli* asparaginase products (either native or pegylated) as a component of a multi-drug combination chemotherapy regimen for the treatment of patients with acute lymphoblastic leukemia (ALL) and a history of clinically significant hypersensitivity to the *E. coli*-derived asparaginase products.

The efficacy data provided in this original Biologics License Application (BLA) in support of the licensure are limited to the results of a single-arm, open-label pharmacokinetic and pharmacodynamic trial (Protocol AALL07P2) that enrolled 59 patients with ALL enrolled in NCI-sponsored cooperative group trials conducted by the Children's Oncology Group, with a history of hypersensitivity to pegaspargase. Approval was based on demonstration of sustained asparaginase activity at or above levels resulting in depletion of plasma asparagine concentrations of 3 μM or lower, which is an accepted surrogate measure for clinical benefit (an incremental improvement event-free survival due to the addition of asparaginase to multiagent chemotherapy). Collection of targeted adverse reactions, selected based on the extensive historical clinical experience with both *Erwinia* and *E. coli* asparaginase products, was performed in Protocol AALL07P2 and in the Erwinaze Master Treatment Protocol (EMPT), a single-arm, open-label expanded access program for patients with ALL or lymphoblastic lymphoma with a history of hypersensitivity to *E. coli*-derived asparaginases (native or pegylated).

FDA accepted the results of this single arm trial based on a surrogate endpoint for several reasons. First, the contribution of *Erwinia* asparaginase to these multi-drug regimens has not been isolated through controlled clinical trials, however indirect evidence of its benefit is so widely accepted by the community that the conduct of controlled clinical trials would not be feasible at this time. Second, the data supporting the correlation between the surrogate endpoint is compelling and has been previously used as the basis for selection of the dosing regimens for both Erwinaze and for pegaspargase (Oncaspar®, Sigma Tau) as well as to support expanded labeling claims for pegaspargase. As noted by EUSA, pharmacologic data from the Children's Cancer Group trials (CCG 1941, CCG 1962 and CCG 1961) were used to select dosing as follows: efficacy requires depletion of asparagine to levels of 3 μM or lower

and this level of depletion can be reliably achieved when serum L-asparaginase activity levels are maintained at or above 0.1 International Units (IU)/mL throughout the dosing period.

Therefore, a surrogate endpoint for efficacy, measuring an effect directly attributable to asparaginase *Erwinia chrysanthemi*, was proposed by FDA and accepted by the IND sponsor to provide evidence of efficacy in single-arm trials where *Erwinia* asparaginase was initiated at variable timepoints throughout treatment with multi-agent chemotherapy. The initial proposal by FDA of sustained asparagine depletion was determined by EUSA to be infeasible to reliably measure under clinical trial conditions, and FDA agreed that this could be replaced with a primary endpoint demonstrating sustained asparaginase activity at a level correlating with effective doses (0.1 IU/mL or greater at 48 hours post-dosing).

Specific issues that prolonged the review time and delayed approval were the findings that the primary efficacy endpoint measurement was not reliable, based on inspectional findings, requiring identification of additional archived samples with acceptable quality for testing, identification of a new contract laboratory for testing of samples, and validation of the modified assay method for testing. In addition, numerous deficiencies in the manufacturing facility were identified on inspection which required corrective action prior to approval to ensure manufacture of a well-characterized, safe, pure, and potent biological product. This application is also notable for the lack of product characterization ordinarily required to support product safety at approval, including lack of GLP toxicology studies, incomplete pharmacokinetic characterization, lack of drug-interaction studies, lack of definitive assessment of asparaginase *Erwinia chrysanthemi*'s effects on QTc interval prolongation, and lack of immunogenicity testing. FDA determined that these data were not required for a risk:benefit determination in light of the unique circumstances of more than three decades of experience in the clinic and the product's proposed use for an unmet medical need in an orphan disease setting.

With the exception of the CMC reviewers in OBP, all review team members recommended approval of this application. The CMC reviewers noted that all issues had been satisfactorily addressed but did not make a recommendation for or against approval.

2. Background

Acute lymphoblastic leukemia

Acute lymphoblastic leukemia (ALL) is a malignancy arising in the marrow and involving the lymphoid lineage. Based on the Surveillance Epidemiology and End Results (SEER) database, an estimated 5,730 individuals will be diagnosed with and 1,420 patients will die of ALL in 2011. From 2004-2008, the median age at diagnosis for ALL was 13 years of age. Approximately 60.3% were diagnosed under age 20. In both adults and children, multi-agent chemotherapy consisting of remission induction followed by intensification is the initial line of treatment. Since the 1970's, asparaginase products have been component of such combination regimens, however its use can be limited by development of serious toxicities (pancreatitis, thrombosis) and development of anti-product antibodies resulting in allergic reactions and loss of efficacy.

Approved products in this class

There is only indirect evidence that this product class (asparaginases) contributes to clinical benefit. This indirect evidence comes from multiple sources including: (1) historically controlled comparisons indicating higher complete remission rates with the addition of asparaginase to combination chemotherapy (native *E. coli* asparaginase), (2) poorer outcomes in patients who develop high-titer antibodies with decreased exposure as compared to patients without such antibodies (exploratory analyses of CCG 1961 trial), (3) poorer event-free survival in patients who are unable to tolerate asparaginase, defined as completion of <25 weeks of the planned 30 weeks of asparaginase treatment, compared to those who tolerate therapy (exploratory analyses of DFCI 91-001), and (4) poorer outcomes in exploratory analyses comparing patient outcomes when *Erwinia* asparaginase was not available for use in patients with hypersensitivity to *E. coli*-derived asparaginase due to manufacturing problems in 2002 with outcomes for patients treated when *Erwinia* asparaginase was available.

There are two asparaginase products approved by the FDA native *E. coli* (Elspar®) and pegaspargase (Oncaspar®), which is native *E. coli* asparaginase covalently linked to polyethylene glycol. Elspar was approved in 1978 for the following indication

> ELSPAR is indicated in the therapy of patients with acute lymphocytic leukemia. This agent is primary useful in combination with other chemotherapeutic agents in the induction of remissions of the disease in pediatric patients. ELSPAR should not be used as the sole induction agent unless combination therapy is deemed inappropriate. ELSPAR is not recommended for maintenance therapy.

The approval was based on historically controlled data indicating that the addition of native *E. coli* asparaginase improved the complete response rate in patients with acute lymphoblastic leukemia. Efficacy was evaluated in an open-label, multi-center, single-arm study in which 823 patients less than 16 years of age with previously untreated acute lymphoblastic or acute undifferentiated leukemia received native *E. coli* asparaginase as a component of multi-agent chemotherapy for induction of first remission. Of 815 evaluable patients, 758 (93%) achieved a complete remission. In a previous study, in a similar patient population, which utilized an initial induction chemotherapy regimen containing the same agents without native *E. coli* asparaginase, 429 of 499 (86%) patients achieved a complete remission.

Oncaspar was approved on February 1, 1994

> "as a component of a multi-agent chemotherapeutic regimen for the treatment of patients with acute lymphoblastic leukemia ALL and hypersensitivity to native forms of L-asparaginase."

This approval was based on data from four open-label studies enrolling a total of 42 patients with multiply-relapsed, acute leukemia [39 (93%) with ALL] with a history of prior clinical allergic reaction to asparaginase. Patients received pegaspargase as a single agent or in combination with multi-agent chemotherapy. The re-induction response rate was 50% (95% confidence interval: 35%, 65%), based upon 36% complete remissions and 14% partial remissions. These results were similar to the overall response rates reported for patients with ALL receiving second-line, native *E. coli* L-asparaginase-containing re-induction

chemotherapy. Anti-tumor activity was also observed with single-agent pegaspargase. Three responses (1 complete remission and 2 partial remissions) were observed in 9 adult and pediatric patients with relapsed ALL and hypersensitivity to native *E. coli* L-asparaginase.

On July 24, 2006, the following expanded labeling claim for Oncaspar was approved

> "as a component of a multi-agent chemotherapeutic regimen for the first line treatment of patients with ALL"

The effectiveness of Oncaspar for this indication was based on the results of a single open-label, multicenter, randomized, active-controlled study conducted in 118 pediatric patients aged 1 to 9 years with previously untreated standard-risk ALL. Patients were randomized 1:1 to pegaspargase or native *E. coli* L-asparaginase as part of combination therapy. The primary determination of effectiveness was based on demonstration of similar asparagine depletion (magnitude and duration) in the pegaspargase and native *E. coli* L-asparaginase arms. The protocol-specified goal was achievement of asparagine depletion to a serum concentration of ≤ 1 μM. The proportion of patients with this level of depletion was similar between the 2 study arms during all 3 phases of treatment (Induction, Delayed Intensification 1, and Delayed Intensification 2). Serum asparagine concentrations decreased within 4 days of the first dose of asparaginase in the treatment phase and remained low for approximately 3 weeks for both pegaspargase and native *E. coli* L-asparaginase arms. Serum pharmacokinetics of pegaspargase, based on an enzymatic assay measuring asparaginase activity, were assessed in 34 newly diagnosed pediatric patients with standard-risk ALL in this study. Asparaginase activity of greater than 0.1 IU/mL for approximately 20 days post-dosing were observed in over 90% of the samples from patients treated with pegaspargase during Induction, Delayed Intensification 1, and Delayed Intensification 2.

Based upon the results of this study, the use of pegaspargase has largely supplanted native L-asparaginase for use in the initial treatment of ALL. The development of "high-titer" antibodies to pegaspargase was 2% in Induction (n=48), 10% in Delayed Intensification 1 (n=50), and 11% in Delayed Intensification 2 (n=44) in study supporting approval in the treatment of first-line ALL, therefore there remains an unmet need to an asparaginase product that can be substituted for pegaspargase in patients with hypersensitivity to *E. coli*-derived asparaginases.

Asparaginase *Erwinia chrysanthemi* development program
The initial investigational new drug application (IND 290) for asparaginase *Erwinia* chrysanthemi was submitted to FDA on January 23, 1968. The manufacturing process has undergone both major and minor modifications both prior to and following the initial marketing approval for asparaginase *Erwinia* chrysanthemi in the United Kingdom in 1985. The drug is currently marketed in multiple countries, including Canada and the United Kingdom. Because of manufacturing difficulties, asparaginase *Erwinia* chrysanthemi was removed from the worldwide market from 2003 to 2004.

The initial US IND was held by Ipsen Ltd; on February 15, 2006, OPi SA, notified the FDA that they had assumed sponsorship of IND 290 from Ipsen Ltd. EUSA Pharma, Inc. subsequently acquired the IND and currently has US distribution rights.

Pre-submission communication history

May 30, 2006: A pre-BLA meeting was held with OPi SA. OPi SA were informed that

- Based on the CMC summary information supplied, the product did not conform to current regulatory standards; OPi SA acknowledged that the manufacturing process had not been modified significantly since 1985. FDA stated that a full description of the manufacturing history should be provided, with comparability assessments as described in ICH and FDA guidances to assess the effects of manufacturing changes; lot release specifications were not supported by data provided and testing was insufficient to ensure consistent manufacture of a well-characterized, safe, pure, and potent biologic. FDA provided recommendations for additional testing were provided. In addition, OPi SA was requested to provide more details on process validation and stability testing.
- No additional nonclinical studies would be required
- Literature reports would not be sufficient to establish the clinical efficacy and safety of *Erwinia* asparaginase. OPi SA agreed to conduct a clinical trial to establish durable asparagine depletion as a surrogate for clinical efficacy.

July 12, 2007: Type C meeting regarding product manufacture and characterization

- FDA requested that OPi SA provide additional information on the proposed identity test, provide a plan for identification of impurities, include K_m and K_{cat} as release and stability tests, and that a BLA would need to be supported by data from three validation lots.

July 30, 2008: Advice/information request letter issued by FDA

- FDA acknowledged EUSA Pharma's submission of data justifying the inherent unreliability of asparagine assay results arising from an inability to control for *ex vivo* metabolism. FDA indicated that asparaginase activity could serve as the primary PK study endpoint which can be reliably measured in the clinical setting and is directly related to asparagine depletion. However, FDA indicated that EUSA Pharma would be expected to document in a future BLA submission all due diligence to collect these samples for asparagine depletion (PD) data to support the review and approval of a BLA.

November 6, 2008: Type C teleconference regarding product manufacture and characterization

- OPi SA requested input on the specifications for the new Ion Exchange Chromatography (IEX) method for drug substance (DS) and drug product (DP) characterization; the method for determining asparaginase activity (K_m/K_{cat}); the analysis methods for setting specifications for DS and DP; and the proposed DS and DP stability program. Agreement was reached on the latter two issues, however FDA requested further information on the specifications for the IEX method and additional information on the Km/Kcat testing.

August 11, 2009: Type C teleconference regarding product characterization

- FDA provided comments on the planned approaches to Erwinia asparaginase (b) (4) (b) (4) validation and EUSA agreed to review the use of quality criticality analysis as part of this plan. FDA stated that the planned approaches on (b) (4) (b) (4) (b) (4) appeared acceptable, cautioning that a final determination would be contingent on the results obtained. EUSA Pharma agreed to now include additional parameters for sterility and include the data in the validation of the hold times. FDA advised EUSA

Pharma that a rationale for control strategy of bioburden and endotoxins in the process should be provided in the BLA (e.g., an assessment of the potential of each solution used in the process to support microbial growth).

September 3, 2009: Type B meeting regarding clinical data to support a BLA

- EUSA Pharma identified protocol AALL07P2 as the trial intended to support the proposed application with additional safety data to be provided from the EMPT trial. FDA agreed that the proposed plan to submit a BLA containing the clinical study report with data on the primary endpoint (asparaginase activity Day 11- 13 of a course of Erwinase) available for 50 patients and complete toxicity data for Course 1 was acceptable. FDA also agreed that an ISE would not be required as efficacy was supported by a single study, however an ISS would be required. FDA stated that Complete CRFs from all patients from the ALL07P2 study should be submitted. The "Patient Registration Form," "Drug Accountability Log" and the "Case Report Forms" for all subjects enrolled on the EMTP study should also be submitted. Further, FDA requested that datasets be submitted in CDISC format rather than as COG legacy datasets. (b) (4)

Dec. 8, 2009:

- Type C meeting, limited to CMC issues, to discuss product manufacturing and characterization scheduled for Dec. 10, 2009 was cancelled upon receipt of FDA's draft responses to EUSA's questions.

July 15, 2010

- Fast track designation granted for the development program for the investigation of L-asparaginase (Erwinia chrysanthemi) for treatment of acute lymphoblastic leukemia (ALL) in patients who develop hypersensitivity reactions to pegaspargase.

Application history

The application was a rolling submission, with the final portion of the application received on November 1, 2010. The BLA was granted priority review status. Key milestones for this application are listed below:

- September 8, 2010: first module submitted
- November 1, 2010: last module received
- January 14, 2011: 74-day deficiency letter issued
- February 16, July 7, and August 4, 2011: Information request letters issued
- February 23, 2011: major amendment received
- March 7-11, 2011: Division of Bioequivalence and GLP Compliance (DBGC) inspection of contract research laboratory responsible for measurement of serum asparaginase levels (primary efficacy endpoint) and serum asparagine levels (secondary efficacy endpoint). Inspectional findings revealed that the data for both the primary and secondary endpoint were not reliable.

- March 14-22, 2011: inspection of the drug substance and drug product manufacturing facility by BMT and OBP review staff; multiple deficiencies requiring correction prior to approval were identified.
- June 23, 2011: meeting held between FDA and EUSA to discuss FDA's inspectional findings for the research laboratory responsible for measurement of asparaginase activity, the primary study endpoint, indicating that these data were not reliable and discussion of proposals from EUSA to provide reliable evidence of efficacy.
- August 4, 2011: submission of validation protocol for modified asparaginase assay method to be employed by the new contract research laboratory for measurement of the primary efficacy endpoint
- August 5, 2011: teleconference between FDA and EUSA to discuss plans to address outstanding deficiencies in chemistry, manufacturing, and controls.
- September 26, 2011: DBGC inspection of new contract laboratory, conducting the analysis of asparaginase levels as the measurement of the efficacy endpoint

3. CMC/Device

I concur with the conclusions reached by the chemistry reviewer regarding the acceptability of the manufacturing of the drug product and drug substance. Manufacturing site inspections were acceptable with modifications as requested during the review to release testing methods and procedures. Stability testing supports an expiry of 24 months from the date of manufacture when stored at 2-8 °C, where the date of drug product manufacture is defined as (b) (4) Stability testing also supports an expiry dating period for the drug substance of (b) (4) from the date of manufacture when stored (b) (4). There are no outstanding issues.

Asparaginase *Erwinia chrysanthemi* is a tetrameric protein of 4 identical polypeptide chain subunits and has a molecular weight of 140 kDa. L-asparaginase is an enzyme that catalyzes the deamination of L-asparagine to L-aspartic acid, with the release of ammonia. The enzyme is produced (b) (4) from *Erwinia chrysanthemi*, a strain of anaerobic, gram-negative bacteria that is a plant pathogen.

The drug substance manufacturing process involves (b) (4)

Inspection of (b) (4), which conducts sterility testing of the drug product, was waived since this testing site is under the surveillance program and was inspected in early (b) (4) inspection of (b) (4) (b) (4) which performs labeling, packaging and distribution of the drug product; this site also was determined to be acceptable. However, the (b) (4) inspection of the (b) (4), which manufactures drug substance and drug product, identified issues requiring additional information and modification to the manufacturing process in order to meet acceptable manufacturing standards. As a result of the

additional data received and modifications made to the manufacturing process at FDA's request, the OBP and OC reviewers recommended approval of this application.

Evaluation of sterility in the manufacturing process was conducted by the CMC and facilities review staff. I concur with the conclusions reached by the OBP and facilities reviewers that there are no outstanding clinical microbiology or sterility issues that preclude approval.

4. Nonclinical Pharmacology/Toxicology

I concur with the conclusions reached by the pharmacology/toxicology reviewer that there are no outstanding pharm/tox issues that preclude approval.

The reports of non-clinical studies conducted on *Erwinia* asparaginases that were submitted in the BLA were from studies conducted prior to the issuance of the Good Laboratory Practice (GLP) regulations set out in 21 CFR Part 58. The non-clinical toxicology reviewer evaluated the submitted reports of acute toxicity studies conducted in rats and rabbits and of the chronic toxicology studies conducted in rabbits, dogs, and monkeys. For the chronic toxicology studies, a single dose was explored, precluding an assessment of the dose-toxicity relationship. These single and repeat-dose toxicology studies were performed by the intraperitoneal and intravenous routes of administration rather than the intramuscular route of administration intended for human use. The findings of these studies were generally uninformative and did not predict the clinical toxicities observed in clinical studies with both this product and other marketed asparaginase products; where mortality was observed, the cause of death was not established. The studies were also limited by the lack of data bridging the dosing used in the non-clinical toxicology studies to the proposed dose, for which extensive human clinical trial experience is available. As noted by Drs. Kufrin and Pilaro, "Comparison of the doses tested in the animal studies to the doses of Erwinaze™ tested clinically is not possible. The nonclinical doses were calculated based on specific activity of the Erwinia-derived asparaginase enzyme [i.e. International Units (IU/kg)] and the recommended clinical dosing is based on IU/m^2, but the specific activity of Erwinaze ™ is calculated using a different potency assay than was used to establish the activity of the Erwinia-derived enzyme used in the toxicity studies."

Additional toxicology studies, conforming to current regulatory standards were not required given the decades-long use in human subjects and the well-characterized toxicity profile in children with ALL. Carcinogenicity and mutagenicity studies for this protein therapeutic have not been requested, as per the ICH S9 and ICH S6 guidelines. A complete battery of fertility, embryo-fetal and pre-post-natal nonclinical developmental toxicity studies, as required for this patient population by the ICH M3 guidance, will be conducted under a post-marketing requirement.

5. Clinical Pharmacology

I concur with the conclusions reached by the clinical pharmacology reviewer that there are no outstanding clinical pharmacology issues that preclude approval.

Pharmacokinetics
Pharmacokinetic (PK) assessment was based on enzyme activity rather than on the direct measurement of the asparaginase molecule in serum, due to methodological issues leading to an inability to develop a reliable assay for direct measurement of the protein content.

Although the clinical trial supporting efficacy, AALL07P2, specified that samples were to be obtained to characterize the PK profile of Erwinaze (sampling at 2 hours, 24 hours, 48 and/or 72 hours post-dosing as well as on day 9 or 10 after the last dose in the first course and 28 or 72 hours post-dosing for courses 2 and 3), these data are not described in product labeling because the data were deemed unreliable due to sample handling and analysis issued discussed in section 11 of this review. The only reliable data available in the submission were trough serum concentrations at 48 hours post-dosing (for doses administered on Monday or Wednesday) or 72 hours post-dosing (for doses administered on a Friday) with archived samples that were analyzed at a second contract research laboratory. Since the measure of the activity level was critical to establishing efficacy, these data are described in section 7 of this summary review. Detailed PK characterization will not be required since the selection of the dose regimen has been established during the extensive clinical use of this product under IND.

Prior to initiation of Protocol AAL07P2, FDA evaluated the assay for measurement of asparaginase activity. For the assay validation, *E. coli* asparaginase was used as the standard and serum was used as the sample matrix. Based on the unreliability of the original data provided in the application, the methodology was modified to account for the use of alternative samples (archived samples retained for immunogenicity testing); the validation protocol was submitted to the FDA on August 4, 2011. The FDA has reviewed this protocol and accepted it with the caveat that it be expanded to include robustness. The validation report included short-term stability information for both Erwinaze and the control (*E. coli* asparaginase) in human serum, including validation of freeze-thaw stability in human serum, and assay robustness results on the effects of pH, temperature and variations in critical reagents. Dr. Yang concluded that "Based on the evaluation of the data in this validation study, it appears that the method has the required attributes to make it suitable for its intended use."

Pharmacodynamics
As noted in Section 2 of this summary, although FDA originally requested that the PK and PD trial utilize asparagine depletion (PD) as the surrogate endpoint for efficacy, on July 30, 2008, FDA issued an advice/information request letter in which FDA acknowledged EUSA Pharma's submission of data justifying the inherent unreliability of asparagine assay results arising from an inability to control for *ex vivo* metabolism. FDA indicated that asparaginase activity could serve as the primary PK study endpoint which can be reliably measured in the clinical setting and is directly related to asparagine depletion. FDA also stated that EUSA Pharma would be expected to document in a future BLA submission all due diligence to collect these samples for PD data to support the review and approval of a BLA.

The trough plasma asparagine levels were to be measured prior to dose 6 for each of the first three courses at either 48- or 72-hours post-dose and on day 9 or 10 post-dose 6 during the first course. Data were provided in the BLA for 47 patients receiving a minimum of six doses of

Erwinaze per treatment course in trial AALL07P2. EUSA reported that 98% (n=47) of the patients in course 1; 97% (n=31) of the patients in course 2, and 100% (n=22) of the patients in course 3 achieved the clinically desired level of trough plasma asparagine level ≤3 μM (0.396 μg/mL). As noted by Dr. Yang, accurate measurement of asparagine levels is technically challenging due to the instability of asparagine in the sample, which is subject to *ex vivo* metabolism. Based on the inspectional findings conducted at the original research laboratory and these results are deemed unreliable. The assay methodology was never fully validated to FDA's satisfaction and the analysis of asparagine levels was not conducted at the second contract laboratory.

Immunogenicity
Immunogenicity samples were collected prior to the first dose of Erwinaze in each course, on days 8 and 22 of the first course (Course 1), and on days 6 and 15 of subsequent courses in patients in trial AALL07P2. These samples have been frozen and stored pending development of appropriately validated assay methods for assessment of binding and neutralizing antibodies to Erwinaze, including IgE antibody assessments.

QT/QTc Evaluation
Electrocardiograms (ECGs) were performed only at two timepoints (prior to the first dose of Erwinaze and at 1 hour following the sixth dose of Erwinaze) in trial AALL07P2, as agreed-upon with the clinical review division during a September 30, 2009 meeting. Upon review of the data, the QT-IRT review states that "No definitive conclusions can be made regarding QTc effects due to Erwinaze from this study since this assessment involved single post-treatment; locally read ECGs collected at 1 hour post-dose 6 with categorical analysis only. The sponsor did not report the actual numeric values for QTc but number of patients who had absolute QTc ≥500 ms and change from baseline QTc ≥60 ms were reported. Therefore it is infeasible to derive the mean QT effect or to explore concentration-QT relationships following treatment with Erwinaze. In the absence of comparator arm data, the significance of the findings in categorical analysis is unclear." Based on this conclusion, the FDA QT-IRT team recommended that no language related to the effects of Erwinaze on QTc be included in the product labeling

Drug Interactions
No data on drug interactions were provided in the application. Given the extensive clinical experience with *Erwinia* asparaginase as a component of multi-agent chemotherapy over more than three decades, as well as the known mechanism of action of this enzyme and the lack of anticipated effects on drug metabolism through P450 pathways, there are no safety concerns and FDA will not require that drug interaction studies be conducted under post-marketing requirements.

6. Clinical Microbiology

See Section 3 of this review.

7. Clinical/Statistical-Efficacy

Efficacy of Erwinaze is demonstrated by evidence of sustained asparaginase activity during treatment, at a level that is predicted to result in sufficient asparagine depletion leading to leukemia cell death. This surrogate endpoint for efficacy has been used in cooperative group clinical trials to determine the appropriate dosing regimens for this and other asparaginase products; in the U.S., the level identified as predicting clinical efficacy is a trough asparaginase activity level of ≥ 0.1 international units (IU) per mL serum. However, some investigators have suggested that a trough asparaginase activity level of ≥ 0.4 IU/mL is considered a better predictor of clinical efficacy; therefore the analysis of this data also contains an FDA-conducted exploratory analysis using this cut-point.

As discussed in section 2, FDA agreed that a single-arm, 50-patient clinical trial conducted in patients with acute lymphoblastic leukemia (ALL) who were unable to receive pegaspargase due to hypersensitivity in which sustained asparaginase activity of ≥ 0.1 IU/mL at 48 hours post-dosing was demonstrated would be sufficient to establish efficacy in support of a marketing application.

The original BLA submission contains a clinical pharmacokinetic (PK) and pharmacodynamic (PD) study report (COG AALL07P2) in support of the proposed use of Erwinaze in patients with ALL who developed hypersensitivity to (b) (4). *E.coli*-derived asparaginase while on active treatment. Study COG AALL07P2 was a single-arm, multi-center, open-label, safety and clinical pharmacology trial conducted in 59 patients treated under the front line cooperative group ALL protocols who were unable to continue to receive pegaspargase due to hypersensitivity reactions. Patients received Erwinaze 25,000 International Units (IU)/m^2 for six doses administered intramuscularly on a Monday, Wednesday, and Friday schedule as a replacement for each scheduled dose of pegaspargase remaining on their original treatment protocol. The primary efficacy endpoint was demonstration that the majority of the patients attained a clinically effective level of asparaginase activity, defined as a trough serum asparaginase activity of ≥ 0.1 IU/mL. This trial was powered to test the hypothesis that 70% of patients; against an alternative of meeting this trough threshold activity in 50% of patients.

The clinical samples were analyzed by an academic laboratory and FDA's data verification inspection identified major issues/deficiencies with the sample handling and assay performance, rendering the analytical results for both the serum asparaginase activity and plasma asparagine concentrations unreliable (see original BLA review, dated 25 May 2011). Subsequently, the FDA agreed with the Sponsor's proposal to use the frozen immunogenicity samples collected at pre-dose 4 in Course 1 to test the trough level of asparaginase activity in a CRO facility using a validated assay. On September 15, 2011, EUSA submitted the asparaginase activity results together with the assay validation report. In this second analysis, asparaginase activity was determined in serum samples from 35 patients with 48-hour trough samples and 13 patients with 72-hour trough samples. Asparaginase activity of ≥ 0.1 IU/mL was present in all trough samples for the 48 patients, meeting the pre-specified criteria for demonstration of efficacy based on sustained levels of asparaginase activity. FDA also conducted an exploratory analysis of trough asparaginase activity ≥ 0.4 IU/mL. Eighty percent

(28/35) of those evaluated at 48 hours and 38% (5/13) evaluated at 72 hours had serum asparaginase activity levels ≥ 0.4 IU/mL

Proportion of Patients in AALL07P2 with Sustained Asparaginase Activity		
Trough sampling time (number of patients)	**Primary Objective** Proportion with asparaginase activity ≥ 0.1 IU/mL	**FDA Exploratory Analysis** Proportion with asparaginase activity ≥ 0.4 IU/mL
48-hour trough Proportion (n/N) 95% CI	 100% (35/35) (90%, 100%)	 80% (28/35) (64%, 90%)
72-hour trough Proportion (n/N) 95% CI	 100% (13/13) (77%, 100%)	 38% (5/13) (18%, 65%)

8. Safety

The safety of Erwinaze was evaluated in two open-label, single-arm trials, AALL07P2, and the Erwinaze Master Treatment Protocol (EMTP), an expanded access program. The size of the population was adequate to assess safety, however the uncontrolled nature of the studies, use of concomitant anti-neoplastic medications with substantial toxicities, and targeted data collection limit conclusions. This is off-set by the many decades of clinical use of this product under IND and expanded access programs, which has been sufficient to identify the most common and most serious events. In the clinical programs, safety data collection was targeted to collection of adverse reactions attributable to the product class and of serious adverse events possibly related to Erwinaze.

Protocol AALL07P2 enrolled 58 patients treated on National Cancer Institute-sponsored cooperative group ALL protocols who were unable to continue to receive pegaspargase due to hypersensitivity reactions. Patients received 6 doses of Erwinaze 25,000 IU /m^2 intramuscularly on a Monday, Wednesday, and Friday schedule as a replacement for each scheduled dose of pegaspargase remaining on their original treatment protocol. The characteristics of the safety population from AALL07P2 are median age of 10 years (range 2 to 18 years), 59% male, and a racial/ethnic composition of 78% White, 10% Black/African American, 5% Asian, and 5% Hispanic or Latino. Nine patients stopped therapy prior to completion, four due to allergic reactions, and five due to physician or patient choice.

The EMPT trial is ongoing; at the time of data cut-off for the BLA submission, 843 patients with ALL or lymphoblastic lymphoma with systemic hypersensitivity to an *E. coli*-derived asparaginase had been enrolled and the application contained safety data for 574 patients. The characteristics of the safety population are as follows: median age of 9 years (1 to 66 years),

62% male, and 97% with leukemia vs. 3% with lymphoma as the underlying malignancy. Patients received ERWINAZE according to several schedules; doses ranged from 20,000 to 25,000 IU /m^2. Twenty-five percent of patients failed to complete planned treatment; in just over half of these patients (n=78), ERWINAZE was discontinued for adverse reactions, primarily allergic reactions.

In Protocol AALL07P2, safety information included all reported adverse events with systematic collection of the following adverse events of special interest: allergy, pancreatitis, coagulopathy (hemorrhage, thrombosis or infarct), hyperbilirubinemia, hyperglycemia, hyperlipidemia, ketoacidosis, and CNS events (hemorrhage, thrombosis or infarction, cerebral venous thrombosis). The EMTP safety data were derived from case report forms that collected adverse event information. The forms specifically requested information on occurrence of allergic reactions, thrombotic events, hemorrhagic events, hepatobiliary disorders, pancreatic disorders, and hyperglycemia.

Serious hypersensitivity reactions, including anaphylaxis occurred after the use of ERWINAZE in 5% of patients in clinical trials. Pancreatitis occurred following ERWINAZE in 4% of patients in clinical trials (Table 1). Glucose intolerance occurred following ERWINAZE in 2% of patients in clinical trials (Table 1), and, in some cases, did not fully resolve during the course of the trial, indicating that this condition is permanent in some patients. Serious thrombotic events, including sagittal sinus thrombosis have been reported with both *E. coli* and *Erwinia*-derived L-asparaginase therapy. The following coagulation proteins were decreased in the majority of patients after a 2-week course of ERWINAZE: fibrinogen, protein C activity, protein S activity, and anti-thrombin III.

Pooled safety data from 630 patients enrolled in AALL07P2 or EMTP were used to generate overall incidence rates for non-hematologic, non-infectious, adverse reactions of any severity (NCI CTC Grades 1-4) reported in patients receiving one or more doses of ERWINAZE (Table 1). The incidence rates for non-hematologic, non-infectious adverse reactions of Grade 3 or 4 severity occurring with ERWINAZE, provided separately by clinical trial (AALL07P2 or EMTP) are provided in Table 2.

Table 1: Per Patient Incidence of Non-Hematologic and Non-Infectious Adverse Reactions (Pooled Results of AALL07P2 and EMTP; n=630)		
Adverse Reaction Category	**Adverse Reactions**	**Number of Patients (%)**
Allergic Reaction	Systemic Allergic Reactions (Anaphylaxis, Hypersensitivity, Urticaria)	108 (17%)
	Local Reactions (injection site)	3 (< 1%)
Pancreatitis	Pancreatitis	24 (4%)
Clinical Coagulation Abnormalities	Total	16 (3%)
	Thrombotic	10 (2%)
	Hemorrhagic	5 (1%)
	Transient Ischemic Attack	1 (< 1%)
	Disseminated Intravascular Coagulation	1 (< 1%)
Liver Abnormalities	Total	27 (4%)
	Hyperbilirubinemia	8 (1%)
	Abnormal Transaminase	22 (3%)
Hyperglycemia	Hyperglycemia	15 (2%)
Hyperammonemia	Hyperammonemia	4 (1%)
Fever	Fever	16 (3%)
Gastrointestinal Symptoms Not Associated with Pancreatitis	Vomiting	15 (2%)
	Nausea	10 (2%)
	Abdominal Pain	6 (1%)
Headache	Headache	5 (1%)
Diarrhea	Diarrhea	5 (1%)
Seizure	Seizure	4 (1%)

Table 2: Per Patient Incidence of Grade 3 and 4, Non-Hematologic, Non-Infectious, Adverse Reactions by Clinical Trial		
Description of Event	**AALL07P2(N =58)**	**EMTP (N=572)**
Allergic Reaction / Hypersensitivity	5 (9%)	27 (5%)
Pancreatitis	0	4 (1%)
Hyperglycemia	0	11 (2%)
Clinical Coagulation Abnormalities - Thrombosis	0	6 (1%)
Clinical Coagulation Abnormalities – Hemorrhage	0	1 (< 1%)
Elevated Transaminases	1 (2%)	2 (< 1%)

Immunogenicity

Serum samples were obtained during the conduct of trial AALL07P2 to evaluate for anti-product antibodies. These samples have been archived until the assay methods have been

validated to reliably and sensitively detect binding, IgE, and neutralizing antibodies to Erwinaze. Results of the immunogenicity assessment will be submitted under post-marketing requirements as identified in the approval letter for this application.

9. Advisory Committee Meeting

This application for asparaginase *Erwinia chrysanthemi* was not referred to an FDA advisory committee because outside expertise was not necessary; there were no controversial issues that would benefit from advisory committee discussion.

10. Pediatrics

Because this drug product for this indication has an orphan drug designation, it is exempt from the requirement of the Pediatric Research Equity Act. However, safety and efficacy have been established in clinical trials (AALL07P2 and EMPT) where the nearly all patients enrolled were children, adolescents, or young adults.

11. Other Relevant Regulatory Issues

A clinical site inspection was conducted at one site, which enrolled 4 of the 59 patients in Protocol AALL07P2. No issues were identified that might affect the reliability of the clinical data.

The (b) (4) DBGC inspection identified major deficiencies with the sample handling and assay performance of the serum asparaginase and plasma asparagine assays, which led to a conclusion that these results could not be considered reliable.

For the serum asparaginase activity assay, deficiencies included:
1) failure to adequately document preparation and storage of asparaginase stock solutions;
2) failure to adjust nominal asparaginase concentrations in calibrator and quality control solutions for the actual content of L-asparaginase commercial vials;
3) incomplete documentation of sample storage and handling conditions and stability during these conditions, failure to reject analytical run #480 when one of the three quality control samples failed the acceptance criterion;
4) failure to exclude serum samples from clinical sites received in the thawed state.

For the plasma asparagine assay, deficiencies included:
1) failure to reject analytical runs when the quality control samples failed the acceptance criterion;
2) failure to reject chromatograms when no asparagine internal standard was detected or peaks could not be accurately integrated;
3) failure to exclude plasma samples from clinical sites which were unacidified or received in the thawed state;

4) failure to demonstrate stability of samples under the conditions of the study. These bioanalytical deficiencies/issues render the analytical results for both the serum asparaginase activity and plasma asparagine concentrations unreliable.

These issues were successfully addressed by identification of additional patient samples stored in another facility that could be tested, validation of the assay methodology including some of the aspects (freeze/thaw, short- and long-term stability) which were not controlled by the initial laboratory, and identification of a new contract laboratory. There are no other unresolved relevant regulatory issues.

12. Labeling

- Proprietary name:
  Neither the Division of Medication Error and Prevention Analysis nor the Division of Oncology Products 2 identified concerns regarding the applicant's proposed proprietary name, Erwinaze, which has been accepted by FDA.

- Proper name:
  The proposed proper name, which is under review by USAN, was rejected by FDA. Instead FDA assigned a proper name, asparaginase *Erwinia chrysanthemi,* which distinguishes this product for other approved asparaginase products, is easily pronounceable, and non-promotional.

- Physician labeling
  - Indications and Usage
    - FDA revisions to remove unsupported claim (b) (4).
  - Dosage and Administration
    - Edited for brevity and command language
    - Removed statements that (b) (4) (b) (4) which is inconsistent with current medical practice and therefore highly unlikely when the drug is administered intramuscularly.
  - Dosage Forms and Strengths
    - Deleted information (b) (4)
  - Contraindications
    - Added serious hemorrhagic or thrombotic events with prior asparaginase treatment as contraindications
    - Changed contraindication from (b) (4) to the more general "serious" for the contraindication in patients with a history of pancreatitis.

- Warnings and Precautions
  - Retitled section 5.1 to denote anaphylaxis, clarifying severity of this condition. Edited for brevity and command language
  - Added incidence information in Section 5.2, based on clinical trial data and a cross-reference to section 6.1.
  - Added incidence information in Section 5.3, based on clinical trial data, a statement that glucose intolerance may be irreversible, and a cross-reference to section 6.1.
  - Retitled section 5.4 from (b) (4) to the more specific serious events of "Thrombosis and Hemorrhage" and providing clarity on the severity of this condition. Retained reference to *E. coli*-derived products as based on general knowledge in the pediatric oncology community
- Adverse Reactions
  - Deleted information on (b) (4) Added a separate table for Grade 3-4 adverse reactions to this section.
  - Deleted section (b) (4) s all relevant safety data available from this trial is included in section 6.1
  - In section 6.2 (Immunogenicity) replaced statement (b) (4) with the statement "There is insufficient information to characterize the incidence of antibodies to ERWINAZE"
- Use in Specific Populations
  - Revised section 8.1 to (b) (4) Added statements that there are no data in pregnant women. Replaced the statement (b) (4) with "ERWINAZE should be given to a pregnant woman only if clearly needed."
  - Deleted (b) (4) as recommended in 21 CFR 201.57.
  - In subsection 8.3, replaced the sentence " (b) (4) with "Because many drugs are excreted in human milk, and because of the potential for serious adverse reactions in nursing infants from ERWINAZE, a decision should be made to discontinue nursing or to discontinue the drug, taking into account the importance of the drug to the mother."
- Overdosage
  - Edited for brevity and to remove (b) (4) to ERWINAZE
- Description
  - Revised for brevity
  - Added a description of the contents of the final product vial as per 21 CFR 201.57

- Removed [redacted] (b) (4)

- Clinical Pharmacology
  - Removed [redacted] (b) (4)
  - Removed [redacted] (b) (4)
  - Removed [redacted] (b) (4)
- Nonclinical Toxicology
  - Edited for brevity
  - Removed [redacted] (b) (4)
- Clinical Studies
  - Removed [redacted] (b) (4)
  - Removed [redacted] (b) (4)
  - Updated all results to reflect the analyses of asparaginase activity from the archived samples tested with validated method at the second contract research laboratory
  - Included description of the patient population studies in AALL07P2
- References
  - [redacted] (b) (4)
- How Supplied
  - Modified to move information on storage and handling following reconstitution to Dosage and Administration.

- Carton and immediate container labels
  Comments primarily regarding lack of consistency with current regulations and applicable guidances on carton/container labeling, provided by Kimberly Rains, OBP project manager, and the DMEPA reviewer were provided to the applicant. All carton/container issues have been satisfactorily resolved and there are no outstanding issues.

- Patient labeling/Medication guide
  The applicant did not propose a Medication Guide but did propose [redacted] (b) (4)

13. Decision/Action/Risk Benefit Assessment

- Regulatory Action:
  I concur with the recommendations of the review team and also recommend approval of this application.

- Risk Benefit Assessment
  The benefits of ERWINAZE outweigh its risks, supporting the approval of ERWINAZE for the proposed indication in the agreed-upon product labeling.

  The benefits of ERWINAZE have been established indirectly; such data indicate that event-free survival (persistent leukemia, relapse or death) is shorter in patients who do not receive a full course of asparaginase as a component of their multi-agent chemotherapy, either due to inability to receive the drug based on allergic reactions or due to drug shortages. Sustained asparaginase activity, sufficient to deplete plasma asparagine to clinically effective levels, was demonstrated with ERWINAZE in a patient population with an unmet medical need, i.e., patients who can no longer receive *E. coli*-derived asparaginase products. In contrast, the serious risks of ERWINAZE, which include allergic reactions in 10%, pancreatitis in 4%, potentially glucose intolerance in 2%, and life-threatening hemorrhage or thrombosis with long-term morbidity in less than 1% of patients are considered acceptable by the patient and medical community in light of the serious nature of the disease, similar risks with *E-coli*-derived products, and the risks of other components of the multi-agent chemotherapy regimen used for treatment of ALL.

- Recommendation for Postmarketing Risk Evaluation and Mitigation Strategies
  I concur that a REMS is not needed to ensure safe and effective use of ERWINAZE.

- Recommendation for other Postmarketing Requirements and Commitments

  Post-marketing requirements under 505(o)

  - To conduct non-clinical embryo-fetal development and toxicity (EFT; ICH S5 (R2) Harmonized Segment C) studies of ERWINAZE in rats and rabbits.

    Rationale: to characterize the potential developmental effects of Erwinaze, as this data cannot be collected in a timely manner in human subjects, given the indication (b) (4)

  - To conduct non-clinical fertility and early pregnancy (Segment I; ICH S5(R2) Harmonized Segment A-B) studies of ERWINAZE in rats.

    Rationale: to characterize the potential developmental and fertility effects of Erwinaze, as this data cannot be collected in a timely manner in human subjects, given the indication (b) (4)

- To conduct non-clinical peri-postnatal developmental (PPND; Segment III; ISC S5(R2) Harmonized Segment D-F) studies of ERWINAZE in rats.

  Rationale: to characterize the potential developmental and fertility effects of Erwinaze, as this data cannot be collected in a timely manner in human subjects, given the indication (b) (4)

- To develop a validated, sensitive, and accurate assay for the detection of binding antibodies to ERWINAZE, including procedures for accurate detection of antibodies to ERWINAZE in the presence of ERWINAZE levels that are expected to be present in the serum at the time of patient sampling. A summary of the validation exercise including supporting data, a summary of the development data supporting assay suitability for parameters not assessed in the validation exercise, and the assay SOP will be provided to FDA.

  Rationale: to characterize the risks of anti-product antibodies, which potentially may be result in allergic reactions or in loss of efficacy, due to increased clearance.

- To develop a validated, sensitive, and accurate assay for the detection of neutralizing antibodies to ERWINAZE, including procedures for accurate detection of neutralizing antibodies to ERWINAZE in the presence of ERWINAZE levels that are expected to be present in the serum at the time of patient sampling. A summary of the validation exercise including supporting data, a summary of the development data supporting assay suitability for parameters not assessed in the validation exercise, and the assay SOP will be provided to FDA.

  Rationale: to characterize the risks of anti-product antibodies expected to result in loss of efficacy, due to neutralization of enzyme activity.

- To conduct an assessment of anti-drug antibody (ADA) binding response and neutralizing ADA response to ERWINAZE with validated assays (required under PMR 4 and 5) capable of sensitively detecting ADA responses in the presence of ERWINAZE levels that are expected to be present at the time of patient sampling. The ADA response will be evaluated in all archived sampling time points available from all patients in the COG Study AALL07P2.

  Rationale: to characterize the risks of anti-product antibodies which potentially may be result in allergic reactions, loss of efficacy, due to increased clearance, or loss of efficacy due to neutralization of enzyme activity.

Post-marketing commitments:

- To conduct a container closure integrity study and determine the sensitivity of the test methods.

- To conduct performance qualification of the Erwinaze lyophilization process.

- To provide validation data from the executed protocol for shipping ERWINAZE drug product from the (b) (4) to the US market.

- To conduct a study to substantiate the use (b) (4) (b) (4) as the (b) (4)

- To collect data from rabbit pyrogen testing on three lots of thawed and diluted drug substance solution prior to (b) (4). The final validation report should contain a description of the method, the rabbit pyrogen test results, and an assessment of the impact of (b) (4) on drug product quality and the drug product manufacturing.

- Implement the proposed process improvements described in the March 4, 2011, BLA amendment and re-assess the bioburden and endotoxin limits based on data from three extraction batches.

- To monitor bioburden and endotoxin levels in CM6 pooled fractions, CM8 pooled fractions, and DEAE pooled fractions held for more than 24 hours at scale from three runs, demonstrating that acceptance criteria are met.

- To complete the qualification of bioburden and endotoxin in-process test methods:

- The final reports for the bioburden and endotoxin assay will each provide data on two additional batches of drug substance.

- To review the specifications for all release and stability test methods when the manufacture of a statistically significant number of Erwinaze DS and DP lots is completed. The final report of this analysis together with any revised release and stability specifications will be submitted in accordance with 21 CFR 601.12.

- To validate hold times for each DS process intermediate, where applicable, in order to demonstrate that the quality of Erwinaze DS is not affected. This study should include a worst case hold scenario, defined by the cumulative maximal time for each hold step along with an evaluation of the purity and potency of process intermediates and of the resulting DS. The complete hold times validation report and supporting test results together with any revisions in the established hold times will be submitted in accordance with 21 CFR 601.12.

- To increase the assay sensitivity for SDS-PAGE. The revised assay will be submitted together with the validation report and supporting test results.

- To perform SEC and AUC testing in a side-by-side analysis of Erwinaze DS samples that have been subjected to stress conditions. Results of these studies together with any revisions to your control strategy will be submitted as a final report in accordance with 21 CFR 601.12.

- To provide a revised protocol for qualification of the current and future Erwinaze reference standards.

- To submit an experimental plan for evaluating and, if appropriate, implementing L-asparagine as the substrate for measuring the K_m and k_{cat} of Erwinaze DS and DP.

- To provide an experimental plan to assess the types and amounts of smaller sub-visible particulates (b) (4) in the final drug product under real-time and stress stability conditions along with a timetable for this work. The plan and a timescale for the subsequent assessment of the impact sub-visible particles may have on the quality, clinical safety and efficacy of Erwinaze DP along with a control strategy will be provided in accordance with 21 CFR 601.12.

- To revise the peptide mapping method used for DS and DP release testing in order to enable chromatographic base-line resolution of most peptide fragments while accounting for >98% of the protein sequence. The revised assay will be submitted together with the validation report.

SIGNATURES PAGE

/s/Patricia Keegan/ November 15, 2011

Patricia Keegan, M.D. Date
Director, Division of Oncology Products 2
Office of Hematology and Oncology Drug Products
Office of New Drugs
Center for Drug Evaluation and Research