SCIENTIFIC DISCUSSION

This module reflects the initial scientific discussion and scientific discussion on procedures, which have been finalised before 30 November 2004. For scientific information on procedures after this date please refer to module 8B.

1. Introduction

Increase in uric acid production can be caused in any patient with a rapid proliferating malignancy. As a result of the increased cell turnover, nucleic acid is catabolised. This increased purine metabolism leads to increase in the uric acid concentration (hyperuricemia). Hyperuricemia is also known to occur following aggressive cancer therapy regimens that cause an increase in cell lysis releasing purine metabolites.

Chemotherapy-induced hyperuricemia is one of the adverse consequences of the tumor lysis syndrome. Tumor lysis syndrome occurs as a result of rapid release of intracellular contents into the bloodstream, which then increase to life-threatening concentrations. This syndrome is characterized by hyperuricemia, hyperkalemia hyperxanthenemia (especially in patients treated with allopurinol), hyperphosphatemia, hypocalcemia, lactic acidosis, and often-acute renal insufficiency. Lethal cardiac arrhythmias are the most serious consequences of hyperkalemia. Hyperuricemia and hyperphosphatemia may result in acute renal failure.

Uric acid is the end product of purine metabolism in birds, reptiles, primates and humans and is produced in the liver by oxidation of xanthine and hypoxanthine. In all other mammals, uric acid is further oxidized by the enzyme urate oxidase to allantoin. However, humans lack this enzyme. As uric acid has relatively poor water solubility, the acute increase in plasma levels of uric acid seen may lead to acute renal failure caused by the precipitation of crystals of uric acid in renal tubules. Renal function can be further compromised by calcium phosphate precipitation triggered by the release of intracellular phosphates occurring during tumor lysis.

Allopurinol (4-hydroxypurinol), an analogue of xanthine, is currently considered the standard pharmacological therapy for hyperuricemia and is given before cytotoxic treatment. Besides the application of allopurinol, management is directed at correcting metabolic abnormalities and preventing further renal damage. The mechanism of action of allopurinol is different from Fasturtec.

Rasburicase (SR29142) is a recombinant form of the enzyme urate oxidase produced from genetically modified strain of Saccharomyces cerevisiae cloned with cDNA from a strain of Aspergillus flavus. The enzyme is a tetrameric protein with identical subunits of a molecular mass of about 34 kDa - similar to the native A. flavus urate oxidase- and catalyses the oxidation of uric acid to allantoin, a water-soluble product that is easily excreted by the kidney.

Fasturtec is a second-generation product with respect to a non-recombinant enzyme preparation, obtained from Aspergillus flavus, which has been commercialised by the company in France and Italy under the tradename of Uricozyme.

Fasturtec is provided in a 3 mL vial as a freeze-dried powder for solution for intravenous injection. Each vial contains 1.5 mg rasburicase, sodium phosphate buffer (pH 8.0, 40 mM after reconstitution of the freeze-dried powder) mannitol and alanine.

The proposed indication of Fasturtec is “treatment and prophylaxis of acute hyperuricaemia, in order to prevent acute renal failure, in patients with haematological malignancy with a high tumor burden and at risk of a rapid tumor lysis or shrinkage at initiation of chemotherapy.” The recommended dose of Fasturtec is 0.2 mg/kg/day, administered once a day as a 30-minute intravenous infusion for a treatment duration of approximately one week. Administration of rasburicase does not require any change in the timing or schedule of initiation of cytoreductive chemotherapy.

2. Chemical, pharmaceutical and biological aspects

Composition

One vial of Fasturtec contains 1.5 mg of rasburicase, max. 1.0 mg disodium phosphate, max. 0.06 mg sodium dihydrogen phosphate, 15.9 mg alanine, 10.6 mg mannitol and 12.6-14.3 mg disodium phosphate. The sodium phosphate buffer of the finished product may vary because the volume of the active substance solution included in the final bulk product varies according to rasburicase content. The 1 mL solvent includes , besides water for injections, 1.0 mg poloxamer 188. Nitrogen is used during filling of the freeze-dried powder and solvent into the vial and ampoule , respectively.

The dose is expressed in mg instead of EAU (Enzyme Activity Unit). This is justified since rasburicase is a well-characterised and well-defined product. The use of HPLC (size exclusion chromatography which groups all the active isoforms) to define enzyme content in milligram is more precise than enzyme activity determination. Nevertheless, the definition for one EAU of rasburicase has been clearly stated in the SPC.

One enzyme activity unit (EAU) corresponds to the enzyme amount that converts 1 µmol of uric acid into allantoin per minute in TEA pH 8.9 buffer at +30°C.

Since rasburicase is not sufficiently stable to be stored for a longer time under refrigerated conditions, a freeze-dried formulation was developed to guarantee the molecule's integrity and enzyme activity.

The compatibility of poloxamer with rasburicase and the excipients used has been demonstrated as well as compatibility with the 0.9% NaCl infusion solution and typical infusion devices.

Active substance

- Characterisation

Fasturtec is a second-generation product with respect to a non-recombinant enzyme preparation, obtained from *Aspergillus flavus*, and commercialised by the company in France and Italy under the tradename of Uricozyme®.

Rasburicase is a tetrameric protein with identical subunits of a molecular mass of about 34 KDa and consisting of a single 301 amino acid polypeptide chain, which is N-terminal acetylated.

The enzyme activity of rasburicase is determined by an *in vitro* test based on the degradation of uric acid on allantoin and is representative of the *in vivo* biological activity.

The active product may contain a high amount of active isoforms. This heterogeneity of the rasburicase protein has been verified by various analytical techniques e.i. by isoelectrofocussing and anion exchange chromatography. The heterogeneity can be explained by the structural complexity of the protein, a tetramer composed of four identical monomers. Only a few modifications in a monomer may lead to numerous forms of the tetramer. Potential oxidation and deamidation sites of the rasburicase protein have been elucidated. It has been demonstrated that these modifications can be detected by the tests routinely applied on the active substance bulk : oxidation by reverse phase chromatography and anion exchange chromatography, and oxidation and deamidation by peptide mapping.It is important to note that the total specific activity does not change with increasing heterogeneity and that the major related substances are deamidation and oxidation based.

Additional comparative tests of Fasturtec and the non-recombinant product Uricozyme have been carried out to assess the comparability of the two products. The results clearly show that rasburicase is less heterogeneous than the non-recombinant product and the specific activity of rasburicase is about 50% higher than that of the non-recombinant urate oxidase product.

- Impurities

Process-related impurities, i.e. components derived from expression system and components derived from fermentation, extraction and purification have been evaluated. An immunoradiometric assay was developed for detecting residual host cell proteins (HCP). HCP were detected at levels below 40ppm in the drug substance batches. A PCR method was developed to detect levels of host cell DNA. The production process satisfactorily removes HCP and DNA.

- Specifications and routine tests

Peptide mapping verifies the primary structure of the monomer.
Rasburicase content is determined by size exclusion chromatography, the principal peak consists of all active tetrameric forms of rasburicase. The enzyme activity is determined by an in-vitro assay, based on conversion of uric acid into allantoin. The specific activity is an overall control of the integrity of the active protein . Anion exchange chromatography reveals the charge heterogeneity of the tetrameric protein . Size exclusion chromatography is performed to monitor aggregated forms of the tetramer and high molecular mass weight related impurities. Reverse phase chromatography and SDS-Page electrophoresis are performed under dissociating conditions and therefore monitor the heterogeneity and purity of the monomeric protein.

The release specifications for the active substance are in line with the batch analysis results (mean ±3 SD).

- Batch analysis

Batch analysis results of batches produced by the commercial process confirm active ingredient production consistency.

In-house primary and working reference materials were established since no national or international standards are available for urate oxidase.

Active substance production process

An European Drug Master File has been submitted for the production process for rasburicase.
A conventional two-tiered cell bank system is used.
The fermentation is carried out in two steps : preculture and full-scale fermentation, followed by extraction.
The purification process comprises concentration and chromatographic steps and finally filtration and filling into drug substance container.

Other ingredients

In addition to the active substance, the formulated product contains three excipients: alanine, mannitol, disodium phosphate and water for injections.
The reconstitution solvent comprises poloxamer 188 and water for injections. Butylhydroxytoluene (BHT) is added to the poloxamer as an antioxidant. The BHT used as an antioxidant in Lutrol® F68 complies with the Ph. Eur. requirements.

Finished product

- Method of preparation

Fasturtec is provided in a 3 mL vial as a freeze-dried powder for intravenous injection . Each vial contains 1.5 mg rasburicase , sodium phosphate buffer (pH 8.0, 40 mM after reconstitution of the freeze-dried powder), mannitol and alanine.

The proposed industrial batch size is 60.5 L for 55 000 vials. Several batches of drug substance after control and release may be pooled for the preparation of the nominal volume of solution to be freeze-dried in order to ensure the production of a batch of drug product of the nominal size (55 000 vials).
The processes following parameters are considered as critical: filtration, duration of holding time of the bulk solution and freeze-drying. All these parameters have been validated. Analytical results of the three batches of freeze-dried powder produced at industrial scale demonstrate consistency of the lyophilisation process. Water content was below 2% w/w for all three batches.
The results demonstrated the suitability of the industrial freeze-drying parameters.

The manufacturing process of the solvent includes a sterile filtration and autoclaving.

- Control tests on the finished product

The manufacturing process of the finished product does not impact the primary structure of the monomer. Therefore, identification by peptide mapping, which verifies the monomer for the active substance, is not performed for the finished product. The methods selected to routinely monitor levels of related substances and impurities are anion exchange, size exclusion and reverse phase chromatographies, which respectively reveal the charge heterogeneity profile of the active tetramer, the aggregated forms, and the related proteins by difference in polarity. SDS-page electrophoresis is only performed for the stability studies of the finished product. The water content is an important parameter for ensuring stability of lyophilised material.
The other tests are the same as for the active ingredient.

Appropriated controls and specifications are set up to characterise the quality of the solvent for parenteral use.

The container for the powder for injection with a 3 mL capacity is made of clear Type I glass. The closure is a grey stopper made of chlorobutyl rubber and complies with Ph. Eur. The overseal is an aluminium cap. The compatibility of the rubber stoppers with rasburicase has been demonstrated.

The container for the solvent is a single dose 2 mL score-break glass ampoule made of clear Type I glass which is in accordance to Ph. Eur.

- Virological documentation

Rasburicase is produced from a genetically modified strain of *Saccharomyces cerevisiae. S. cerevisiae* is a microorganism used in food processing that is not known to be a host for viruses posing a risk to higher organisms. The expression construct is well characterised and contains no viral sequences.
One of the biological ingredients, Peptone, was only used in the culture media for the master cell bank (MCB) prepared in 1991. It was derived from bovine non-neuronal tissue.

Considering a) that the peptone was used in the preparation of the MCB only, and b) the type of tissue used in preparation of the peptone (bone, connective tissue and skin) is category IV (no detectable infectivity), the risk of TSE transmission by the use of Fasturtec is considered negligible.

The other ingredient of biological origin is yeast extract used in the preparation of the media for the MCB, working cell bank and fermentation. The yeast extract is certified free of products of animal origin. All other raw materials are synthetic, including glycerol.

Information has been provided demonstrating that the medicinal product is made in compliance with the CPMP " Note for Guidance on Minimising the Risk of transmitting animal spongiform encephalopathy agents via medicinal products "

Stability of the product

- Stability tests on the active substance

Stability data comprises results of stress studies performed during development and long-term data under normal and accelerated conditions on clinical and commercial production batches.
Absence of any interaction between the stopper and active substance has been demonstrated.

Based on the stability data, a storage period of twelve months at +5 ±3°C for the active substance can be applied.

- Stability tests on the finished product

The stability studies performed on the finished product included primary stability under normal and accelerated conditions, stability of the reconstituted solution at +25°C using aged powder, compatibility studies of reconstituted product with typical infusion systems, photostability studies of the reconstituted solution/infusion solution, and stability of the solvent under normal and accelerated conditions.

Results showed no marked difference between the reconstituted solution analysed at the initial time and after 24 hours storage at +25°C.

Compatibility with infusion devices have been documented, and the results show the compatibility of the drug product with the PVC bag, glass bottle or Intermate system (Baxter) over 0.5h of infusion after temporary storage (up to 2 hours) in daylight and at room temperature.
The cardboard packaging provides a protective effect on the photoinstability of the powder.

Based on the primary stability data, the shelf life of 36 months when stored at +5 ±3°C and protected from light can be applied.

The shelf life for the solvent is 48 months at +25 ±2°C or under refrigeration when packaged with the powder.

Discussion on chemical, pharmaceutical and biological aspects

In almost all areas the chemical/pharmaceutical/biological part of the dossier is of good standard and the application meets the relevant EC guidelines. The currently available information demonstrates a consistent production of Fasturtec with a well-defined quality.

The assessment of this product has not raised specific points that need special consideration during a regular GMP inspection of a recombinant DNA product.

Methods to control the quality of the product are adequate.

3 Toxico-pharmacological aspects

Pharmacodynamics

- *In vivo* studies

The *in vivo* pharmacological study of rasburicase was not performed due to the absence of a suitable model in physiologically normal mammals.

- *In vitro* studies

The biochemical characterization of rasburicase shows that the recombinant enzyme has the expected uricolytic activity with a Michaelis constant of 44.4 µM and an optimal temperature of 30°C.

- General and safety pharmacology

Safety pharmacology studies with rasburicase were performed by the i.v. route. These studies showed that rasburicase did not modify neurobehavioral parameters assessed by Irwin test or body temperature in mice, hemodynamic parameters in anesthetized dogs or hydroelectrolytic balance in rats. No interaction studies were performed.

Pharmacokinetics/Toxicokinetics

- Pharmacokinetics

Distribution, protein binding, metabolism and excretion

Distribution, protein binding, metabolism and excretion studies were not conducted. As it is known, radiolabel from a biotechnology product can be incorporated into endogenous molecules making it difficult to trace the product in biological matrices (Preclinical Safety Evaluation of Biotechnology-Derived Pharmaceuticals; ICH Guideline, July 16, 1997). With respect to (LC-MS Liquid Chromatography - Mass Spectrometry) methodologies, endogenous proteins often interfere with extraction and chromatographic procedures making detection of the product, as well as the low levels of polypeptides and amino acids that arise from its catabolism, difficult.

A complicating factor for a distribution study of rasburicase was the presence of native urate oxidase activity in laboratory animals used in toxicology, which would interfere with the uricolytic assay and would confound the tissue distribution evaluation using this alternative methodology.

Additionally, the molecular weight of rasburicase is greater than that of albumin, making it unsuitable for the study of plasma protein binding using conventional techniques.

- Toxicokinetics

Exposure (based on AUC) to rasburicase in rats and baboons following both single and multiple dosing increased linearly with dose. The plasma clearance is low in rats and in baboons much lower than hepatic blood flow. The mean half-life is quite short in animals. The volume of distribution is restricted to plasma or blood volume. There was no difference between males and females. Rasburicase neither modified liver weight nor had any effect on the activities of CYP1A (Cytochrome P450 isosyme 1A), CYP2A, CYP2B, CYP2C, CYP2E, and CYP3A isoenzymes in rats and baboons, following a 4-week treatment suggesting no induction nor inhibition potential.

- Assays

Plasma concentrations of rasburicase were determined by a sandwich type immunoradiometric assay (RIA) using two monoclonal reagent antibodies directed against rasburicase. The tracer was a monoclonal ^{125}I-labelled anti-rasburicase reagent antibody.

The RIA was well validated; effects of dilution, storage, inter- and intra-day variability were acceptable.

Toxicology

- Single dose toxicity:

An acute intravenous toxicity study was performed in OFA-Sprague-Dawley rats and OFI-Swiss mice. The rats and mice showed no adverse clinical signs or changes in body weight gain that could be attributed to rasburicase.

Acute intravenous administration of rasburicase to rats and mice up to doses 75 times the proposed dose for clinical use (based on body weight) is well tolerated. Assuming dose-proportionality, in rats this dose is equivalent to 10 times the human therapeutic dose, based on systemic exposure.

- Repeated dose toxicity:

The repeated dose toxicity studies show that rasburicase is well tolerated in rats and in baboons. Some signs of local irritance were noted in the 4- week rat study and the baboon study. These effects were noted in all groups, including controls. Therefore the effects were attributed to intravenous

injection trauma and potential local irritance of the vehicle. Circulating anti rasburicase antibodies were detected in both species.

- Genotoxicity:

Under the conditions of standard mutagenicity tests (Ames reverse mutation assay, mouse lymphoma mutation assay and unscheduled DNA synthesis) and clastogenicity tests (*in vitro* chromosome mutation assay and *in vivo* rat micronucleus test) rasburicase is non-genotoxic.

- Carcinogenicity:

Studies were not conducted. This was justified because duration of treatment is limited to seven days and rasburicase is to be administered concomitantly with cytotoxic therapies.

- Reproduction Toxicity:

Fertility and early embryonic development to implantation

Rasburicase did not affect reproductive performance, fertility in male and female rats or early embryonic development.

- Embryo-fetal development

No studies were performed. This is justified by the Company by pointing out that Fasturtec is intended to be used in combination with chemotherapeutic agents, which are known to be teratogenic and/or embryotoxic.

- Pre- and postnatal development, including maternal function

In view of the concommitant treatment of patients with embryo- and foetotoxic agents, pre- and postnatal developmental studies (including maternal function) were not performed.

- Local Tolerance:

Intravenous, intra-arterial and paravenous

Local tolerability of rasburicase (1.5 mg in 1 ml) after single intravenous and intra-arterial administration in the rabbit was good. Paravenous administration caused slight to well-defined erythema. Rasburicase (either in a concentrated solution or as a lyophilised powder reconstituted in the solvent) and the solvent intended for clinical use caused no cutaneous or occular irritancy in the rabbit.

- Other toxicity studies:

Hemolytic potential

No haemolytic potential of the tested preparations was observed under the conditions tested.

Other toxicological consideration :

The pharmacodynamics of rasburicase show that hydrogen peroxide and allantoin are formed as a result of the enzymatic uric acid oxidation. Allantoin is eliminated exclusively by renal excretion. An assessment of the potential toxicity and safety pharmacology of hydrogen peroxide, which is formed as a side-product of the enzymatic oxidation catalysed by rasburicase, has been presented by the company. H_2O_2 arising from urate oxidase treatment is considered to be unlikely to pose a significant safety concern for patients with normal levels of G6PDH and GSHPx. However Hydrogen peroxide toxicity has been encountered in the clinic in G6PD-deficient patients and a warning for treatment of G6PDH deficient patient is included in the SPC. There are only very limited toxicological data on allantoin. The safety assessment of the company is based on clinical experience covering only a limited number of endpoints. It seems reasonable to conclude therefore, that elevated allantoin levels do not have consequences in the case of renal insufficiency.

4. Clinical aspects

During the clinical development of Fasturtec, safety and efficacy were assessed in 375 subjects (265 children and 110 adults) in one Phase I (TDR2691) study in healthy adult subjects, three phase II

studies in patients and one phase III study in patients. A total of 347 patients received Fasturtec. The phase II studies were uncontrolled dose selection studies assessing the efficacy and safety of Fasturtec (ACT2511 and ACT2694) and one safety study (LTS3025) in patients requiring treatment of malignancy-related and/or tumor lysis-related hyperuricemia. The phase III study (EFC2975) was a randomized study, comparing the efficacy and safety of Fasturtec to allopurinol. A total of 52 patients were treated before and during induction chemotherapy, including 27 patients who received Fasturtec at a dose level of 0.20 mg/kg per day for four to seven days and 25 patients who received allopurinol for four to eight days. Allopurinol was used according to its approved dose and schedule, along with alkaline hydration as clinically indicated. Patients were stratified according to presentation uric acid concentrations (<8.0 mg/dL, defined as non-hyperuricemic, or ≥8.0 mg/dL, defined as hyperuricemic) and disease (leukemia or lymphoma).

Clinical pharmacology

Pharmacodynamics

- Dose-finding studies

Pharmacodynamic analysis indicated that rasburicase is able to lower the uric acid concentrations below 2-3 mg/dL within 24 hours after start of infusion in patients with high plasma levels of uric acid. Low uric acid plasma levels could be maintained during the dosing period (a secondary transient increase in uric acid (if present) occurred during or shortly after chemotherapy). The uricolytic rate in patients at a dose of 0.15 mg/kg was lower compared to the uricolytic rate in patients at the 0.20 mg/kg dose.

In healthy volunteers receiving a single dose of rasburicase (0.05 - 0.20 mg/kg, study TDR2681), initial uricolytic rates (estimated from 0 – 2 h after start infusion) increased with increasing dose of rasburicase.

Pharmacokinetics

- General:

Absorption
As rasburicase will be administered intravenously, bioavailability will be 100%.

Distribution
After administration of rasburicase to healthy volunteers a volume of distribution (Vz) was found of about 60 – 100 ml/kg (study TDR2681). In patients, including children and adolescents, the volume of distribution was ca. 110 – 127 ml/kg (study ACT2511**,** ACT2694).

As rasburicase is a protein itself, protein-binding studies were not carried out. Drug-drug interactions arising from displacement by rasburicase and of co-administered drugs are not expected.

Metabolism and excretion
Rasburicase is a protein. It is expected that metabolic degradation will follow the pathways of other proteins, i.e. peptide hydrolysis. After hydrolysis the products of catabolism (amino acids) are re-incorporated into new cellular proteins. For this reason metabolism studies with radio-labelled amino acids have not been carried out. It appears that rasburicase was not stable in urine.

No in vitro metabolism studies have been carried out.

- Pharmacokinetics after single dose and multiple dose

In healthy volunteers
After single dosing of rasburicase as a 30-minute infusion (study TDR2681) $AUC_{(0\text{-inf})}$ and $C_{\text{end of infusion}}$ increased linearly over the dose range of 0.10 to 0.20 mg/kg.

In the same study, healthy male volunteers received once daily a 30-minute infusion at 0.10, 0.15 or 0.20 mg/kg for 5 days. Steady state was achieved at day 3. Comparing the $AUC_{(0\text{-}inf)}$ values of the single dosing with the $AUC_{(0\text{-}24h)}$ at day 5 indicates no unexpected accumulation of rasburicase. Clearance, volume of distribution and the elimination half-life at steady state were comparable with those after single dosing. The volume of distribution was similar to the physiological blood volume.

In patients
In studies ACT2511 and ACT2694**,** patients including adolescent and children, with leukemias and lymphomas suffering from hyperuricemia, received once daily a 30-minute infusion at 0.15 or 0.20 mg/kg for 5 - 7 days.

Steady state was achieved at day 2 – 3. Also 2 infants (1 year old) received one of the doses. No differences were observed in the pharmacokinetics compared with the other age groups. Only 6 adults patients were included. Although the limited data, it seems that the pharmacokinetics of rasburicase in patients is comparable with the pharmacokinetics in healthy volunteers. The clearance is increased in children and adolescent, compared with adults (ca. 35%) resulting in a lower systemic exposure to rasburicase. The volume of distribution was somewhat higher in patients (110 – 127 ml/kg) compared to healthy volunteers.

- Interaction studies:

No specific in vivo clinical drug interaction studies have been performed. Drug-drug interactions due to the influence of the cytochrome P450 enzymes are not expected.

- Special groups

Impaired renal function
The influence of renal function on the pharmacokinetics of rasburicase was not studied. Renal elimination of rasburicase or its monomers is considered to be a minor pathway for rasburicase clearance. However, due to the conversion of uric acid (by rasburicase), allantoin is formed, which is excreted by the kidneys.

The data indicate that allantoin pharmacokinetics are may be affected by an impaired renal function. Systemic exposure to allantoin may be enhanced by renal impairment. However, data from the literature show that allantoin is not a xenobiotic in humans and relatively non-toxic. Furthermore, renal impairment does not appear to lead to a qualitatively different Adverse Event profile in patients.

Impaired liver function
The influence of hepatic function on the pharmacokinetics of rasburicase was not studied. As metabolism is expected to occur by peptide hydrolysis, an impaired liver function is not expected to affect the pharmacokinetics of rasburicase in a clinically significant way.

Children
Children and adolescents have a somewhat higher clearance of rasburicase, resulting in a lower systemic exposure. Based upon the pharmacodynamic parameters, such as uric acid concentrations and uricolytic rate, no indication of differences in activity was observed, as compared with adults.

Clinical efficacy

The following clinical trials were performed according to GCP standards.

Dose response studies and dosing
Because Uricozyme® is used at a dose of 100 CBU/kg, equivalent in vitro in terms of uricolytic activity to 0.15 mg/kg of Fasturtec, the dose of 0.15 mg/kg was initially evaluated in patients. Two uncontrolled dose-validation studies (ACT2511 and ACT2694) had a similar two-step design (validation phase followed by an accrual phase). The dose-validation phases were designed to select a dose that would produce a success rate of at least 80% in controlling or normalizing hyperuricemia. The dose that successfully controlled or prevented hyperuricemia in all 14 consecutively enrolled

patients in the dose-validation phase was to be the starting dose in at least 76 more patients in the accrual phase of these studies. The accrual phase was intended to verify the effectiveness of the dose chosen in the validation phase. Safety and pharmacokinetics were also assessed.

Main studies

Description of the studies

- Inclusion criteria

Patients were to have a good performance status (ECOG ≤3 or ≥30% on the Karnofsky scale), to be at risk of hyperuricemia induced by either cytotoxic chemotherapy or malignancy, and to be scheduled for treatment with a cytotoxic chemotherapy regimen within 48 hours. Patients receiving allopurinol at the time of inclusion or having recently received allopurinol were excluded.

The inclusion criteria were similar for the studies, except for the age and diagnosis of the patients. Patients with any age could be included in studies ACT2511 and LTS3025, while patients had to be younger than 21 years in studies ACT2694 and EFC2975. The diagnosis of the malignancy was slightly different in the studies but mostly composed of acute leukemia and lymphoma.

Primary endpoints/assays

Endpoints

In the ACT studies the primary efficacy endpoint was a response to treatment, according to the following criteria:

- The uric acid endpoint (≤6.5 mg/dL in patients <13 years old or ≤7.5 mg/dL in patients ≥13 years old) was reached by T48h ±2h and maintained until 24 hours after the last administration of Fasturtec and
- No other hypouricemic agent was required to control hyperuricemia.

In study EFC2975, the primary efficacy endpoint was the area under the serial plasma uric acid concentration curve from the start of study drug administration until 96 hours (T96h) from treatment start ($AUC_{0\text{-}96}$).

The primary efficacy variable in EFC2975 is different from the **ACT** studies. However, this parameter is not essentially different from the control of uric acid levels. Both measure the control of the uric acid levels and are surrogate endpoints.

No efficacy was measured in the LTS3025.

Table 1 summary all the efficacy endpoints of the phase II/III studies.

Table 1 *Protocol defined efficacy endpoints in the phase II/III studies.*

Endpoint	EFC2975	ACT2511	ACT2694
Plasma uric acid $AUC_{0\text{-}96}$	X		
Response rate		X	X
Plasma uric acid concentrations			
Time course of uric acid		X	X
Percent reduction at 4 hours PFD	X	X	X
Time to first confirmation of control[a]	X		
Time to maximum drop	X		
24-hour allantoin levels		X	X
Occurrence of metabolic abnormalities	X		
Occurrence of renal complications	X		
24-hour urinary creatinine		X	X
Occurrence of hypertension	X		
Incidence of elevated creatinine	X		

The criteria for safety were: biochemistry, hematology, vital signs, physical examination, adverse events, baseline (pre-dose) assays for circulating antibodies (for patients who received a prior course of Fasturtec or Uricozyme®).

The criteria for the occurrence of tumor lysis syndrome were: the incidence of metabolic abnormalities, the incidence of renal complications (number of occurrences of dialysis and abnormal creatinine levels) and the proportion of patients developing hypertension requiring therapy.

Tumor lysis risk classification at baseline
The risk of developing tumor lysis is related to the tumor burden and may be characterized by the presence of hyperuricemia and the level of WBC and LDH (lactate dehydrogenase) at baseline. The company defined a tumor lysis risk classification. (Table to be removed)

Assays

Rasburicase
Rasburicase was analysed in plasma using a radio-immunoassay.

Uric acid
Plasma uric acid was analysed using an enzymatic microplate assay.

The stability of uric acid with rasburicase in plasma was evaluated during sample handling. Instructions for sampling handling indicated that uric acid concentration should be analysed within 4 hours after blood collection, ensuring that the blood is kept at cold temperature.

Study populations/accountability of patients

Table 2. *Patient accountability in all studies (n).*

CATEGORY	ACT2511			ACT2694				LTS3025	EFC2975		
	Dose val.	Accrual	Total	Dose val.	Dose val.	Accrual	Total		Fasturtec	Allo-purinol	Total
Dose (mg/kg)	0.15	0.15		0.15	0.20	0.20		0.20	0.20		
Entered	20	88	108	13	22	98	133	82	27	25	52
Treated	20	87	107	12	22	97	131	82	27	25	52
Completed	20	83	103	12	20	97	129	76	26	22	48
Reason off study											
- Adverse events	0	2	2	0	1	0	1	1	1	0	1
- Death	0	1	1	0	1	0	1	3	0	2	2
- Other	0	1	1	0	0	0	0	2	0	1	1

Efficacy results
- Primary efficacy endpoints

In the two phase II studies (ACT2511 and ACT2694) conduct in patients with haematological malignancies, a dose was selected that would control a normalise hypouricemia in at least 80% of the patients. In study ACT2511 a dose of 0.15 mg/kg was found to fulfil the criterion. Therefore, all patients (107) received this dose. The study showed that Fasturtec controlled or normalised the uric acid concentration (uric acid values ≤ 6.5 mg/dL in patients < 13 years old or ≤ 7.5 mg/dL in patients ≥ 13 years old) within 48 hours after first application of Fasturtec in 99% of the patients. In study ACT2694, the 0.15 mg/kg dose failed to meet the above criteria. The higher dose of 0.20 mg/kg fulfilled the critera and therefore most of the patients (119) received this dose. It was shown that this dose controlled or normalised the uric acid concentration in 95% of the patients.

In study ACT2511, the highest mean uric acid concentration was observed at baseline, 4.9 mg/dL. The mean percentage reduction in uric acid at 4 hour post first dose was 88 ± 12%. In patients in whom urine collection was possible (n=88), the activity of Fasturtec was also measured by determining the

concentration of allantoin in urine. The mean, daily, urinary allantoin levels in patients increased from 0.5 mg at baseline to 1100 mg on Day 3.

In study ACT2694, the highest mean uric acid concentration observed at baseline was 7.5 mg/dL. The mean percentage reduction in uric acid at 4 hour post first dose was 84.9 ± 12.6%. The mean, daily urinary allantoin levels in the 93 patients who received 0.20 mg/kg Fasturtec and for whom allantoin excretion was determined, increased from 0 mg at baseline to 1570 mg on Day 2.

A clinical significant improvement of renal function (decrease of plasma creatinine concentrations) was observed in the patients who were hyperuricemic at baseline as early as Day 2 following the rapid decrease in uric acid concentrations caused by the first dose of Fasturtec.

Based on these results the 0.2 mg/kg dose had been selected for the phase III program.

In study EFC2975, the mean plasma uric acid AUC_{0-96}, the primary efficacy parameter, was significantly lower in the Fasturtec group (128.1 mg·hr/dL) than in the allopurinol group (328.5 mg·hr/dL) (Table 3). At 4 hours post first dose, the mean percentage change from baseline was statistically significantly ($p<0.0001$) lower for the Fasturtec group (-86.0%) than that for the allopurinol group (-12.1%).

Table 3. *Plasma uric acid AUC_{0-96} (mg·hr/dL) in EFC2975 study.*

Dose	n	Mean	Median	STD	Min	Max
SR29142: 0.20 mg/kg	27	128.1	97.1	70.3	61.0	341.8
Allopurinol	25	328.5	295.2	129.3	142.5	639.4

The Fasturtec treatment group also demonstrated a lower mean plasma uric acid level compared to the allopurinol group for both hyperuricemic and non-hyperuricemic patients ($p<0.0001$ for both comparisons). For the subgroup of patients (who were hyperuricemic at any time prior to dosing), mean AUC_{0-96} was 162.4 mg·hr/dL for the Fasturtec group compared to 440.0 mg·hr/dL for the allopurinol group. For the subgroup of patients (non-hyperuricemic), mean AUC_{0-96} was 107.9 mg·hr/dL for the Fasturtec group compared to 265.9 mg·hr/dL for the allopurinol group.

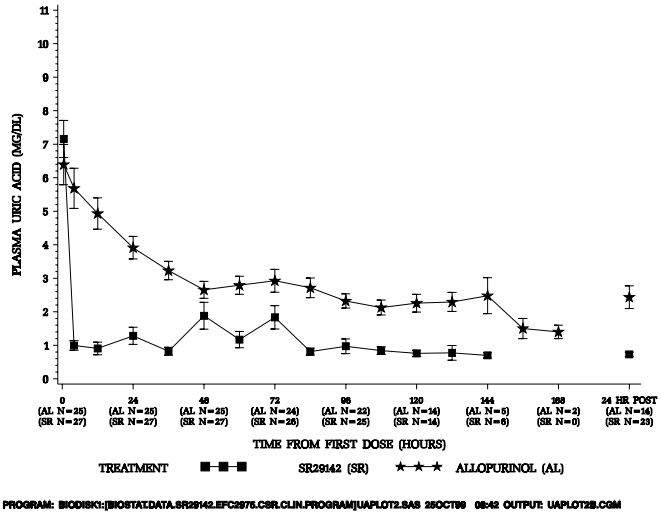

Figure includes values observed up to time of last study dose and one value selected as 24 hour post last dose.

Figure 1 - Mean (±SE) Plasma Uric Acid Concentrations Over Time for All Patients Under Treatment

In patients with hyperuricemia immediately prior to first dose of study drug, the median time to first confirmation of control of plasma uric acid was 4.0 hours post first dose for the Fasturtec group (n=10) and 23.9 hours post first dose for the allopurinol group (n=5).

- Occurrence of tumor lysis

Overall, 63% of the patients treated with Fasturtec showed (based on allantoin measurements and/or metabolic abnormalities) evidence of tumor lysis. Very few (2%) of all treated patients were reported to have tumor lysis syndrome (reported as one adverse event). Of the 347 patients treated with Fasturtec, all the three patients (0.9%) who required dialysis within one week of starting Fasturtec had other contributing factors to the development of renal failure a part from hyperuricemia.

- Evolution of renal function

On average, patients who were non-hyperuricemic maintained stable serum creatinine levels. Hyperuricemic patients had a higher baseline serum creatinine than non-hyperuricemic patients and those treated with Fasturtec showed a progressive decrease in serum creatinine while those treated with allopurinol exhibited relative stabilization of their serum creatinine.

It can be concluded that Fasturtec is able to normalize and control hyperuricemia in the majority of the patients (95-99%). Fasturtec is faster than allopurinol in normalizing the uric acid levels in patients who have hyperuricemia before chemotherapy.

Clinical safety

A total of 347 patients and 28 healthy volunteers received Fasturtec.

- Phase II studies (ACT2511, ACT2694 and LTS3025).

Adverse events and serious adverse event/deaths

In study ACT2511, 3 patients experienced adverse events, considered by the investigator to be likely related to the study treatment: one Grade 1 rash on Day 8; one Grade 1 dermatitis on Day 3 (leading to patient withdrawal) and one hemolysis in a patient subsequently diagnosed as having a glucose-6-phosphate-dehydrogenase deficiency (an exclusion criterion).

Three of the 107 patients died. All three deaths were judged to have no or an unlikely relationship to Fasturtec by the investigator. Other serious adverse events (other than death) were experienced in twenty-five patients (23.4%) during the study. Fever was the most frequently occurring serious adverse event.

In study ACT2694, 2 out of 131 patients experienced adverse events which the investigator considered to have a likely relationship to Fasturtec: grade 4 bronchospasm and dyspnea occurring on Day 1 in one patient leading to study withdrawal and a grade 1 nausea and grade 2 vomiting occurring on Day 2 and Day 3, respectively.

Two patients died. Both deaths were judged unrelated to Fasturtec. There were no toxic deaths. Other serious adverse events (than death) were experienced in fifty-eight (44.3%) patients. Fever was the most frequently occurring serious adverse event.

In study LTS3025 the most frequently reported adverse events were fever, headache, nausea, and vomiting. Twenty-three (28%) patients reported grade 3 or 4 adverse events; the majority was of no relationship to study drug.

Sixteen (19.5%) patients reported one or more serious adverse event: the majority was of no relationship to Fasturtec.

Four patients died. None of these deaths were considered related to Fasturtec treatment. There were no toxic deaths.

Discontinuation due to adverse events

In study ACT2511, two patients withdrew because of an adverse event. One patient had Grade 1 dermatitis (likely relationship to Fasturtec), and the other experienced a Grade 4 acute renal failure (unknown relationship to Fasturtec), recovering without dialysis.

In study ACT2694, one patient withdrew due to adverse events. This patient was treated for pneumonia at study entry. Less than a day after the first study drug infusion, the patient experienced grade 4 bronchospasm and dyspnea.

In study LTS3025, one patient withdrew due to adverse events. This patient received one dose of Fasturtec. The patient had hot flushes, rigors, chest pain, coughing, respiratory disorder, headache and rhinitis, all considered by the investigator to be of likely relationship to the study drug. The patient died 16 days later due to a myocardial infarction and thrombosis, which were not considered, related to study treatment.

- *Phase III randomized study EFC2975*

The vast majority (98%) of the patients reported at least one clinical adverse event of any grade; 26 (96.3%) in the Fasturtec group and 25 (100%) in the allopurinol group.

Adverse events and serious adverse event/deaths

The most frequently reported events were those associated with hematological disorders, gastrointestinal system, body as a whole, respiratory system, as expected given the concomitant cytotoxic therapy and the disease state. The distribution of adverse events generally appeared to be similar between the two treatment groups. No specific pattern or trend was observed. Based on the opinion of the investigator, four patients, three (11.1%) in the Fasturtec group and one (4.0%) in the allopurinol group reported adverse events of a likely relationship to the study drug. These included two reports of headache and one report of fever in the Fasturtec group, and one report of vomiting in the allopurinol group.

Two patients treated in the allopurinol group but none in the Fasturtec group, died during the study period. Neither death was considered related to study drug. Serious adverse events (other than death) were recorded in 4 (14.8%) patients in the Fasturtec group and 8 (32%) in the allopurinol group.

Discontinuation due to adverse events

One patient discontinued in the Fasturtec group due to adverse events (hemolysis, of an unknown relationship to the study drug according to the investigator). With the exception of the two patients who died while on study, no patients discontinued for safety reasons in the allopurinol group.

- *Fasturtec circulating antibodies*

In study ACT2511, 7 (7%) of 97 patients providing samples for analysis of Fasturtec treatment had a positive enzyme linked immunosorbent assay (ELISA) results for detection of Fasturtec circulating antibodies (ACT2694: 17 (14%) of the 121 patients). Of the five patients who received multiple courses of Fasturtec, no increase in quantity, severity, duration of intensity of adverse events was observed.

- *Overall safety consideration*

Because of the clinical context in which Fasturtec is used (supportive care during the induction of chemotherapy of advanced malignant states), the majority of adverse events could be attributed to the patients' underlying neoplastic disease state and the concomitant cytotoxic drugs administered.

In order to assess Fasturtec-possibly related adverse events; several criteria were analyzed in parallel:

- Occurrence of adverse events in the healthy volunteers study (TDR2681)
- Comparison of occurrence of adverse events between Fasturtec and allopurinol groups in the randomized study
- Frequency in uncontrolled studies (ACT2511 and ACT2694)
- Adverse event relationship to study drug according to the investigators
- Adverse events observed with respect to concomitant chemotherapy
- Adverse events anticipated based on the nature (heterolog protein) and mechanism of action (hydrogen peroxide production) of Fasturtec
- Dose relationship
- Review of Uricozyme® Package Insert.

Based on these criteria, the adverse events of special interest were defined as displayed in table 4. The selection of these adverse events represents those adverse events thought to be at least partially related to Fasturtec. These adverse events tended to be of brief duration, with the median of all episodes varying from <1 to 2 days.

Table 4. *Summary of selected adverse events by study population of all studies (number (%)).*

	Controlled Study				Uncontrolled Studies		Pooled Studies	
	Allopurinol (N=25)		SR29142 (N=27)		SR29142 (N=320)		SR29142 (N=347)	
Adverse Event	**All Grades**	**Grade 3 or4**	**All Grades**	**Grade 3 or4**	**All Grades**	**Grade 3 or4**	**All Grades**	**Grade 3 or4**
Any allergic reaction	3 (12.0)	0	1 (3.7)	0	8 (2.5)	2 (0.6)	9 (2.6)	2 (0.6)
Any rash	3 (12.0)	0	4 (14.8)	1 (3.7)	75 (23.4)	3 (0.9)	79 (22.8)	4 (1.2)
Diarrhoea	4 (16.0)	1 (4.0)	8 (29.6)	0	62 (19.4)	3 (0.9)	70 (20.2)	3 (0.9)
Fever	8 (32.0)	1 (4.0)	11 (40.7)	0	120 (37.5)	21 (6.6)	131 (37.8)	21 (6.1)
Headache	3 (12.0)	0	7 (25.9)	0	81 (25.3)	3 (0.9)	88 (25.4)	3 (0.9)
Nausea	6 (24.0)	2 (8.0)	9 (33.3)	1 (3.7)	99 (30.9)	5 (1.6)	108 (31.1)	6 (1.7)
Vomiting	9 (36.0)	1 (4.0)	15 (55.6)	1 (3.7)	149 (46.6)	4 (1.3)	164 (47.3)	5 (1.4)

There was no anaphylactic shock or angioedema reported in any of the studies. No toxic deaths occurred.

During the degradation of uric acid into allantoin, hydrogen peroxide is produced. Hydrogen peroxide is a highly active oxidant that could oxidize a range of products (e.g., proteins and lipids), and therefore other toxic effects are possible. An analysis by the company has shown that the maximal exposure to H_2O_2 generated by treatment by urate oxidase is several orders of magnitude lower than the maximal quenching capacity of blood. The acute toxicities that were observed in the submitted clinical trials are limited to the expected events of hemolysis, particularly in G6PD-deficient patients. Long-term safety data would not allow to distinguish between possible long-term effects of H_2O_2 exposure from that of exposure to multiple cytotoxic chemotherapy drugs. Therefore it may be concluded that there is no clinical evidence at this moment for any unexpected deleterious consequences of collateral H_2O_2 exposure. The company will closely monitor the frequency of haemolysis case reports.

Overall, no clinically significant differences were seen between the adverse event profiles of pediatric and adult patients.

Due to the small number of patients >65 years old (N=23) compared to the number of patients ≤65 years old (N=324), no conclusions based on age can be made. However, the adverse events reported most frequently in elderly patients were also reported most frequently in patients less than 65 years old.

Acute renal disorders were unrelated to Fasturtec, except 1 case report of tubular nephritis with unknown causal relationship.One patient had a cutaneous reaction occuring after cessation of the treatment which was reported as a bullous cutaneous reaction by the investigator. No information was available to confirm this diagnosis. The company proposes to submit an update on tubular disorders and bullous cutaneous reactions within the Periodic Safety Update Reports for urate oxidase.

Discussion on clinical safety

It is difficult to assess the safety and tolerance profile of Fasturtec due to the abundance of side effects induced by the chemotherapy and the serious disease of the patients. Nevertheless the number of patients dropping out of the study due to adverse events is low. The number of deaths during the study period is also low and none of these deaths are thought to be related to Fasturtec. Therefore, these data do not give any indication that serious safety issues arise from the use of Fasturtec in the investigated population. However, a few hundred patients were treated.

5. Overall conclusions, benefit/risk assessment and recommendation

Quality

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way.

Preclinical pharmacology and toxicology

Two major preclinical objections were raised against granting a marketing authorisation for Fasturtec. These objections were related to the potential adverse effects of H_2O_2 and allantoin and were adequately answered.

Efficacy

Fasturtec has been demonstrated to be efficient in the considered patients settings (haematological malignancies) based on the submitted data. However, rasburicase should not be used for more than 7 days and should not be re-administered during the further cycles. A study assessing the respective places of allopurinol and rasburicase in the treatment strategy in lymphoma adult patients will be initiated as a part of the Post authorisation commitments.

Safety

In Section 4.4 "Special warnings and precaution for uses" of the SPC, it is indicated that patients should be closely monitored for onset of allergic-type AEs. The company proposes to submit an update on this type of events within the framework of Sanofi-Synthelabo worldwide pharmacovigilance surveillance. These data will be submitted to the competent authorities together with the Periodic Safety Update Reports for rasburicase. In addition, the safety results from an on-going international compassionate program (281 patients enrolled to date) will be submitted to the Agency upon completion.

The company has agreed to follow up the incidence of hypersensitivity reactions, and if necessary reinforce sections 4.4 and 4.8 of the SPC, following analysis of allergic adverse events that will be reported during a compassionate use program. In addition, Section 4.4 of the SPC specify that: "At present, there is insufficient data on patients being re-treated with Fasturtec to recommend multiple treatment courses.

Benefit/risk assessment

Fasturtec (rasburicase) is a hypouricemic agent, fast and potent acting. The most common adverse event were vomiting, fever, nausea and headache. This dossier has a good overall quality of data set for a relatively innovative drug. The benefit/risk is positive.

Recommendation

Based on the CPMP review of data on quality, safety and efficacy, the CPMP considered by consensus that the benefit/risk profile of Fasturtec indicated for:
"Treatment and prophylaxis of acute hyperuricaemia, in order to prevent acute renal failure, in patients with haematological malignancy with a high tumor burden and at risk of a rapid tumor lysis or shrinkage at initiation of chemotherapy."
was favourable and therefore recommended the granting of the marketing authorisation.