

## SCIENTIFIC DISCUSSION

**This module reflects the initial scientific discussion for the approval of Carbaglu. For information on changes after approval please refer to module 8.**

### 1. Introduction

Carbaglu is indicated for the treatment of hyperammonaemia associated with N-acetylglutamate synthase deficiency. It is presented as a dispersible tablet containing 200 mg of carglumic acid (N-carbamoyl-L-glutamic acid, NCGA).

N-acetylglutamate synthase (NAGS) deficiency is a very rare inborn error of metabolism. Over the last 20 years, between 1980 and 2001, 42 patients from 28 families have been identified by the applicant of whom 34 were diagnosed as NAGS deficiency and 8 siblings who had died without precise diagnosis. NAGS deficiency, which is the rarest congenital urea cycle disorder, results in a severe defect of ammonia detoxification with a rapid lethal course in most cases. The estimated prevalence of NAGS deficiency is 0.00125 per 10,000 persons in the European Union. Carbaglu was therefore designated as an orphan medicinal product in the treatment of this condition.

Urea cycle is the unique elimination pathway of ammonium. In the normal mitochondrion, N-acetylglutamate (NAG) is the necessary allosteric activator of carbamoyl phosphate synthetase (CPS), which is the enzyme giving rise to the first substrate of the urea cycle, i.e., carbamoyl phosphate. NAG is produced by the NAG synthase (NAGS) from acetyl CoA and glutamate in the mitochondrion. In NAGS deficiency, the production of NAG is impaired.

NAGS deficiency leads to various neurological and gastrointestinal (including hepatic) symptoms. Its severity depends on the degree of enzymatic deficiency:

- The neonatal-onset presentation (about 60 %), which corresponds to the most severe enzyme deficiency, is usually fatal. Clinical symptoms are quite uniform: after a symptom-free interval of one to five days, poor sucking, vomiting, progressive lethargy, muscular hypotonia, hypothermia, polypnoea inducing a respiratory alkalosis are observed. Without rapid intervention, the neurologic deterioration is rapidly fatal resulting from coma with convulsions and apnoea.
- The late-onset presentation may manifest at any time in life, from early childhood to adulthood. The clinical expression and outcome are very variable, with either acute decompensation episodes or chronic manifestations with digestive, neurologic, or psychiatric symptoms.

The prognosis of NAGS deficiency has not been firmly established because of its very low prevalence, however, as in any other urea cycle disorder, the aim of long-term therapy has been to maintain metabolic control with normal plasma ammonia and glutamine concentrations. The clinical relevant outcome is to prevent cumulative morbidity and mortality.

There are currently no authorised medicinal products in this indication. As clearly established in all published papers, the alternative non specific waste-nitrogen vehicles and dietary restrictions are not satisfactory since chronic hyperammonaemia and decompensations cannot be controlled by these means only.

The rationale to develop Carbaglu is that NCGA, which is an aminoacid analogue of NAG, although being *in vitro* a weaker activator of CPS than the naturally occurring activator NAG, was shown *in vivo* to reach the mitochondrion more easily than NAG.

## 2. Chemical, pharmaceutical and biological aspects

### Composition

Carbaglu contains 200 mg carglumic acid as active substance and it is presented as elongated dispersible tablets with three break marks on both sides.

Other ingredients contain cellulose microcrystalline, sodium laurilsulfate, hypromellose, croscarmellose sodium, silica colloidal anhydrous and sodium stearyl fumarate.

Carbaglu tablets are packed in polypropylene containers closed by a polyethylene tamper resistant stopper with a desiccant unit.

### Active substance

The chemical name of carglumic acid is N – carbamoyl- L-glutamic acid or (2S)-2-(carbamoylamino) pentanedioic acid. The active substance has one chiral carbon atom and has an optical isomer, which is N-carbamoyl-D-glutamic acid.

Carglumic acid is a white crystalline powder, soluble in boiling water, slightly soluble in cold water and practically insoluble in organic solvents. The pH of a 0.5% aqueous solution is between 2.2 and 3.2. Carglumic acid presents three  $pK_a$  values, 2.50, 3.55 and 8.60, ascribed to the two carboxylic acid groups and to the carbamoyl group, respectively. The isoelectric point of the active substance is 3.02.

Although particle size is not considered to be critical from the point of view of bioavailability in this case, the results of a particle size analysis carried out on six industrial batches of ground powder demonstrated a reproducible particle size around 200  $\mu\text{m}$  with a relative standard deviation of less than 10%. The laser beam particle size analyses are presented for the 6 batches and the particle size distribution profiles are reproducible.

Carglumic acid is synthesised by reaction of L-glutamic acid and potassium cyanate in an aqueous media in a single step, followed by neutralisation with hydrochloric acid and purification by recrystallisations in water. No organic solvents or metal catalysts are used during the synthetic procedure.

The potential impurities originating from the route of synthesis as well as from the degradation of the active substance are hydantoin-5-propionic acid and diaza-1,3-dione-2,4-carboxy-7-cycloheptane. The optical isomer of the active substance, N-carbamoyl-D-glutamic acid might also be present. No residuals of the starting materials, L- glutamic acid and potassium cyanate, were detected in carglumic acid. Residual levels of cyanide ions possibly originating from potassium cyanate were also tested and not detected. Other unidentified substances appear only under stress conditions to which the active substance or the finished product is never subject.

#### *Specifications for the active substance*

The active substance specification includes tests for description, identification (IR, HPLC), assay (HPLC), related impurities (HPLC), specific optical rotation (Ph. Eur), melting point (Ph. Eur), pH (Ph. Eur), loss on drying (Ph. Eur), sulphated ash (Ph. Eur), and heavy metals (Ph. Eur).

**The analytical methods used in routine controls were adequately validated and thus considered suitable. The production yield and the particle size of the active substance are also reproducible. All impurity limits were justified by toxicological considerations.**

However the chiral purity of the active substance should be more accurately addressed and as a follow up measure, the applicant committed to develop and validate a chiral HPLC analytical method for the specific determination of N-carbamoyl-D-glutamic acid, then to retest a large number of carglumic acid batches. Further to its validation, the chiral HPLC will be included in the routine testing of the active substance if the retrospective enantiomeric purity data collection demonstrates its relevance. Moreover, in accordance with the ICH Q3A guideline, specific qualification studies will be carried out if appropriate data are unavailable to qualify the proposed acceptance criterion for N-carbamoyl-D-glutamic acid.

The analysis results from three batches presented by the applicant support the proposed specifications for the active substance.

### *Stability*

A stability study has been carried out under stress conditions. The results reveal that the major degradation product after storage at high temperatures as well as after dispersion in acid solution, and to a lesser extent, after dispersion in H<sub>2</sub>O<sub>2</sub> solution and exposure to light is hydantoin-5-propionic acid. Diaza-1,3-dione-2,4-carboxy-7-cycloheptane is also formed, but in smaller amounts. In contrast, carglumic acid appears to be very stable in pH alkaline solution. The company committed to address as a follow up measure the chiral purity of the active substance under stress conditions once the HPLC chiral purity method is validated.

Stability data from three production batches of the active substance packed in their final containers were also presented. The studies were carried out in compliance with ICH guidelines, under accelerated conditions at 40°C/75% RH and under real-time conditions at 25°C/60% RH. The analysis results obtained from the batches stored at 40°C/75% RH over a 6-month period and at 25°C/60% RH over a 36-month period, remained within the defined specifications.

Considering the stability data, the requested retest period of three years for the active substance when stored in its original packaging was approved.

### **Other ingredients**

All excipients in the tablets are tested to, and comply with, Ph. Eur. requirements. None of the ingredients are of animal origin.

Two different sizes of tablet containers are proposed to allow the distribution of the medicinal product either in a 60-tablet container (suitable for a maintenance therapy), or in a 15-tablet container (suitable to treatment initiation). The containers are made of polypropylene containing a white opacifying agent (mixture of polypropylene and titanium dioxide).

Polyethylene stoppers containing a white colouring agent are used. Stoppers are provided with a desiccant (white silica gel) to prevent humidity problems due to the high amounts of disintegrating agent used in the formulation. As a supplementary precaution, the stoppers are of the tamper-resistant type.

### **Product development and finished product**

The tablets have been formulated so as to obtain an easily water dispersible form with rapid *in vitro* dissolution profile. Because the dose shall be adapted to individual requirements scored tablets have been chosen as they provide dose regimen flexibility through the breaking of the tablets into halves or quarters.

Given the low wettability of carglumic acid, a wetting agent, sodium laurilsulfate, was incorporated in the granulating step so as to enable a rapid dissolution profile of the tablets. The initial amount of sodium laurilsulfate used for the preparation of the clinical formula was reduced in the final formulation, since it was suspected to be the cause of a few episodes of mild to moderate diarrhoea, which occurred during a pharmacokinetic study.

The method of manufacture is a conventional high shear wet granulation and oven drying process, followed by compression. The choice of the excipients, their function and quantity has been sufficiently justified. Process parameters for mixing, drying and granule sizing are given. Equipment types are described and in process controls are stated. Validation was performed on four industrial batches. The granulation step is properly validated. Homogeneity during final mixing and compression has also been established.

The analytical results of three pilot batches and of four industrial validation batches have been submitted. All batches were compliant with the release specifications (appearance, uniformity of mass, disintegration time, fineness of dispersion, dissolution ( $\geq 75\%$  in 15 minutes), identification (HPLC), assay (HPLC), impurities (HPLC) and microbiological purity).

Since tablets present three scores in order to enable breaking for dose adjustment, breakability studies were performed on quarter and half tablets. The resistance to crushing as well as changes to the resistance to crushing over time during storage of the tablet containers for six months at 40°C /75% RH were also studied revealing no modification of average hardness for both tablet ends and centre ends quarters. The results of the content uniformity for halves conformed to the specifications. The breakability tests showed that the uniformity of mass results for half-tablets strictly complied with the Ph. Eur. requirements for single-dose preparations whereas the results for quarter-tablets did not. However, no safety concerns should be raised, because the key element in the treatment of NAGS is the total daily dose of carglumic acid. The pharmacokinetic characteristics of the product allow some flexibility with regard to the frequency of administration. The quarter tablets are only used to facilitate the daily division of the doses when this is required, without jeopardising the treatment.

The results of a comparative pharmacokinetic study between the dispersible tablet and the reference oral powder forms ensure that no bioavailability problems are expected due to substance properties or biopharmaceutics factors under present manufacture conditions. No significant differences were found between the developed formulation and the reference powder. However this initial formulation has been modified to respond to a safety concern, giving rise to a compromise on the amount of sodium laurilsulfate. Dissolution studies support this change.

In process controls performed at each manufacturing step are stated with their acceptance limits. They include the verification of the mixing duration and speed, determination of the granules residual humidity and determination of the tablets weight uniformity, hardness (110-150N) and friability (<0.1%).

### **Stability of the product**

Initial stability studies were conducted using three batches, which however respond to a formulation that was rejected due to excessive amount of sodium laurilsulfate. The medicinal product was then reformulated and additional stability studies were carried out with another series of 3 batches corresponding to the final formula.

All stability studies were conducted in compliance with ICH requirements. The batches have been tested for all parameters included in the specifications with the addition of tablets hardness (no limit stated), friability (<1.0%) and water content (no limit stated).

**During these studies, whatever the storage conditions, all parameters remained within specifications with the exception of impurities content which is time and temperature dependant. As suggested by the results obtained with all batches, the proposed shelf life for the unopened commercially packaged product under the conditions specified in the Summary of Product Characteristics, is acceptable.**

The results from a complementary stability study also support the recommended shelf life of the product after first opening of the tablet container mentioned in the Summary of Product Characteristics.

The preliminary results of the ongoing stability studies carried out with three full scale production batches confirm the stability profile of the finished product.

### 3. Toxicopharmacological aspects

#### Pharmacodynamics

##### Mechanism of action

The mechanism of action of Carbaglu is based on the structural similarity of NCGA to NAG, and its ability to replace NAG for the activation of CPS in patients where NAG is lacking due to the deficiency of the enzyme responsible for its synthesis (NAGS).

Published literature on *in vitro* and *in vivo* studies has been submitted to evaluate the pharmacological profile of NCGA in relation to the claimed indication. Only one *in vitro* study was conducted by the applicant to confirm the ability of NCGA synthesised by the manufacturer of the active substance, Laboratoire Synth-Innove, to activate the target mitochondrial enzyme CPS.

- *In vitro* studies

NCGA was shown to be weaker activator of CPS than NAG, but *in vivo* it was more efficient. Rubio and Grisolia (1) have found that, after injection into mice, radiolabelled NAG could not be detected in the liver mitochondria, as opposed to NCGA. The reason has not been clarified, but Kim et al. (2) suggested that it might be related primarily to the higher resistance of NCGA to hydrolysis by cytosol aminoacylase and Meijer et al. (3) suggested that it might also be related to a higher permeability of the mitochondria to NCGA.

- *In vivo* studies

**With regard to the clinical efficacy of NCGA in the treatment of hyperammonaemia, the evidence is based on the treatment of normal or partially hepatectomised rats receiving potentially lethal doses of ammonium since there is no animal model of NAGS deficiency. The results of two published studies were particularly highlighted.**

- In rats treated with 1 or 4 mmol/kg of NCGA administered intraperitoneally, Kim et al. (2) have found a protection of 61% or 71% respectively against the hyperammonaemia after an intravenous administration of a lethal dose of ammonium acetate (10.8 mmol/kg).
- Lee et al. (4) performed the same type of experiment in 70% partially hepatectomised rats injected with ammonium acetate (3.4 mmol/kg). After NCGA administration (1 mmol/kg) there was a decrease in ammonia levels to  $278.09 \pm 60.02 \mu\text{mol/l}$  ( $p < 0.05$ ) when comparing to the control group ( $415.72 \pm 166.38$ ). The correlation between blood ammonia level and behavioural abnormalities observed in hepatic encephalopathy was also investigated. Concerning the behavioural grading scores, the results after NCGA alone were  $1.09 \pm 0.30$  ( $p < 0.01$ ) when comparing to the control group ( $2.54 \pm 1.36$ ). Thus the protective effect on the behavioural change significantly correlated with their effects on blood ammonia level.

In both studies, a group of animals received NCGA combined with arginine. Results in these groups showed that arginine increased the protective effect of NCGA against ammonia intoxication. It was suggested that this potentialisation by arginine might be due to an increase in the synthesis of NAG which is the natural activator of CPS, and might therefore be dependent on the level of NAGS deficiency. However in the clinical situation, similar results were not achieved. Therefore the concomitant use of arginine cannot be systematically recommended.

Overall these findings provide a sound rational basis for the use of NCGA in the prevention of hyperammonaemia in clinical conditions.

## General and safety pharmacology programme

Safety pharmacology studies evaluated the effects of NCGA on the behaviour, body temperature, and respiratory parameters in rats and on the cardiovascular system in dogs. These studies did not reveal any relevant effects following single dose administration up to 1000 mg/kg of NCGA in both species. In addition *in vitro* studies (Purkinje fibers) did not show any cardiovascular effects of NCGA. Since cardiac effects were neither observed *in vitro* nor *in vivo*, it was considered acceptable not to conduct further investigation on HERG channels.

## Pharmacodynamic drug interactions

No pharmacodynamic interaction studies have been carried out, however since the occurrence of interactions is very unlikely this omission was considered acceptable.

## References:

1. Rubio V and Grisolia S (1981) Human carbamylphosphate synthetase I. Enzyme, 26: 233-9
2. Kim S, Paik WK and Cohen PP (1972) Ammonia intoxication in rats: protection by N-carbamyl-L-glutamate plus L-arginine. Proc. Natl. Acad. Sci. USA, 69:3530-3
3. Meijer AJ, Van Woerkom GM, Wanders RJA, Lof C (1982) Transport of N-acetylglutamate in rat-liver mitochondria. Eur. J. Biochem., 124: 325-30
4. Lee JH, Kim WH, Cho H, Kwak YS, Kim S and Paik WK (1998) N-carbamyl-L-glutamate plus L-arginine can protect ammonia intoxication in rats with reduced functional liver mass. Biochem. Biophys. Res Commun, 248:391-4

## Pharmacokinetics

The pharmacokinetics profile of NCGA was determined based on the results from one study conducted in rats after single oral administration and one *in vitro* study using human and rats hepatocytes. Both studies were performed with radiolabelled NCGA. In addition, toxicokinetic data obtained from the 6-month repeated dose toxicity study in rats as well as data from the safety pharmacology study in dogs were provided.

NCGA was measured in plasma and urine samples using a validated LC/MS-MS method, with a high level of sensitivity and specificity.

## Absorption and distribution

NCGA was rapidly absorbed in rats following single oral administration (500 mg/kg radiolabelled NCGA), with a maximum radioactivity level of 62.6 µgEq/g reached in plasma collected 3 hours post dose.

Toxicokinetic data generated in rats showed consistent results. T<sub>max</sub> was obtained between 2 and 4 hours post-dosing and C<sub>max</sub> values were very high after NCGA administration (about 70 µg/ml in rats given 500 mg/kg). In dogs, high plasma concentrations (ranging 74 – 277 µg/ml) were also achieved 2 hours post-dose (1,000 mg/kg). The exposure in male dogs appeared consistently lower than the exposure in females (85.1±10.0 µg/ml versus 202±65.8 µg/ml); however there are insufficient data to explain these gender differences.

Distribution of NCGA in rats after a single oral dose of 500 mg/kg was extensive with highest amounts found in the kidney, the liver (target organ) and the small intestine mucosa. The likely concentration of NCGA in the cells of the intestinal mucosa could be explained in relation to its *in situ* activity.

## Metabolism and elimination

No metabolites of NCGA were detected *in vitro* in rat hepatocyte cultures. These results were confirmed during the *in vivo* study in the rat and similar results were obtained *in vitro* when using human hepatocytes.

NCGA is eliminated mainly in urine (50%) and in faeces (about 40%) as unchanged compound in rats. Although no metabolite was detected, a small percentage of the total radioactivity was found to be eliminated as expired CO<sub>2</sub> (about 9 %, 72 hours post-dose). Several hypotheses for CO<sub>2</sub> formation pathways were formulated, such as decarboxylation of the compound, but none of them has been experimentally confirmed. The elimination is biphasic with a first rapid elimination phase where about 70% of the dose is eliminated during the first 12 hours. The total excretion in rats accounts for about 97% of the administered dose after 96 hours. Toxicokinetic data showed that T<sub>1/2</sub> is about 3 to 3.5 hours whatever the administered dose in rats.

## **Toxicology**

The toxicology profile of NCGA was evaluated in a limited toxicological programme of studies due to the lack of large availability of product with the desired impurity profile. The species used was the rat, which was considered relevant due its ureotelic profile similar to humans. The programme consisted of the following tests:

- Single dose toxicity studies in rats using oral and intravenous route
- Repeated dose toxicity studies in newborn rats and in young adult rats as NCGA is intended to be administered in young patients
- Complete battery of mutagenicity tests

All the studies were compliant with Good Laboratory Practices.

- *Single dose toxicity*

**NCGA did not cause any toxicity in rats following single doses up to 2806 mg/kg orally and 238.6 mg/kg intravenously (highest dose level achievable for intravenous administration).**

- *Repeated dose toxicity*

A 2-week study was performed in newborn rats aged day 4 to 21 *post-partum*, which received oral doses of 250 – 500 – 1000 and 2000 mg/kg/day of NCGA. No signs of toxicity were noted at 500 mg/kg/day. At 1000 mg/kg/day, only a slight reduction of body weight gain was observed, but there were no clinical signs of toxicity, a normal physical and reflex development, unaffected haematological and blood biochemical parameters. After administration of 2000 mg/kg, clinical signs of ill health (coldness to touch, pallor of extremities, emaciation and an immediate marked loss of weight gain) were observed prior to premature death. This study demonstrated the good tolerance in newborn animals at a high level of concentration and exposure, as well as the absence of effects on the newborn's development.

A 6-month study was carried out in rats aged 4 weeks who received oral doses of NCGA (500 or 1000 mg/kg/day). No treatment related death or signs of systemic toxicity were observed at the dose of 1000 mg/kg/day. The only clinical sign described was ptialism that was observed in almost all animals. Neither the histopathological examination of the organs and tissues nor the laboratory investigation (haematology and urine) revealed consistent changes versus controls.

There was no evidence of immunotoxicity or cell proliferation. From the lack of toxicity in the study, the NOEL was considered to be the 500 mg/kg/day dose level and the NOAEL was considered to be the 1000 mg/kg/day dose level (based on the occurrence of ptialism). The safety margin was estimated to be more than 20, based on C<sub>max</sub>, which appeared acceptable.

No study was performed in a non-rodent species due to the lack of test product available. However, the long experience of the use of NCGA in humans allows to conclude that there is no risk for a chronic administration.

- *Genotoxicity*

The mutagenic potential of NCGA was evaluated in a battery of tests using batches of two different impurity profiles (batch containing impurity hydantoin-5-propionic acid (HPA) at a level of 0.3 % and batches containing HPA at a maximum level of 0.1 %, corresponding to the quality which is defined

for Carbaglu). NCGA was devoid of mutagenic potential in bacteria whatever the batch used. The clastogenic activity shown in an *in vitro* study in human lymphocytes was clearly demonstrated as an artefact due to the acidification of the culture medium. Although a micronucleus test performed in the rat at an excessive dose level (7040 mg/kg, using batch containing 0.3 % of HPA) was found weakly positive, this effect was neither confirmed in another study conducted at the same dose level in the same species (using batches of the 2 different purity profiles), nor in an additional micronucleus test after chronic treatment of rats for 1 month with doses up to 1000 mg/kg/day. It can be concluded therefore that NCGA is devoid of genotoxic potential.

- *Carcinogenicity*

The carcinogenic potential of NCGA has not been evaluated. However, based on the lack of structural analogy of NCGA with known carcinogens, the absence of immunotoxicity, genotoxic and immunodepressive properties as demonstrated respectively in a battery of mutagenicity tests and in rats treated for 6 months, as well as the absence of cell proliferation or signs of hyperplasia in the tissues and organs of rats after chronic administration, NCGA does not present any carcinogenic risk for patients who will be treated for an important part of their life.

- *Reproduction toxicity studies*

At present, the reproduction studies have not been completed. However, a specific study associated with the 6-month study in rats allowed to conclude that NCGA did not affect male and female fertility in relation to mating behaviour, seminology, fertility index and oestrous cycle. Preliminary results from an embryo-foetal toxicity study in rats showed that there is no evidence of embryotoxicity, foetotoxicity or teratogenicity at any of the dose levels tested (500, 1000, 1500 and 2000 mg/kg/day NCGA). Only slight maternotoxicity was recorded with the highest dose. The applicant committed to submit the results of the additional embryo-foetal and peri/post natal studies in rats and rabbits post-authorisation. In view of the preliminary data in animals and absence of data in humans, the potential risk for humans is unknown. Carbaglu should therefore not be used during pregnancy and lactation unless clearly necessary, as recommended in the Summary of Product Characteristics.

- *Impurities*

Hydantoin-5-propionic acid (HPA), which is a major impurity of the technical grade product is limited in the formulation intended to be marketed to less than 0.1%. It was demonstrated that it is a physiological metabolite of histidine and that the quantities brought by the treatment with a product containing less than 0.1% HPA are approximately equivalent to the quantities produced physiologically by histidine metabolism. The limit has therefore been toxicologically qualified. The limit of 20 ppm of cyanates, as potential impurity in NCGA batches, has also been toxicologically qualified.

- *Ecotoxicity/Environmental risk assessment*

An assessment of the risk was performed and no significant risk to the environment related to the use of NCGA is anticipated.

#### **4. Part IV: Clinical aspects**

The clinical development included one pharmacokinetic study carried out in 12 adult male subjects, one *in vitro* metabolic study in human hepatocytes and a retrospective patient data collection organised by the applicant, which included 20 patients that received chronic treatment with NCGA from Orphan Europe starting before 2001. Out of the 20 patients, 12 children born between 1984 and 2000 were treated for a NAGS deficiency to support the claim for the indication. Supportive data from publications relating to 4 patients were also provided.

#### **Clinical pharmacology**

NCGA is a structural analogue of NAG and has been shown *in vitro* to activate liver CPS as already presented in part 3.3 of this document 'Pharmaco-toxicological aspects'.

The principal pharmacological effect of NCGA is the reduction of abnormal ammonia levels and thus it cannot be tested in healthy volunteers. The data that can account for the pharmacological effect of NCGA originate therefore from the preclinical studies and the clinical efficacy data.

### **Pharmacokinetics**

The pharmacokinetics profile of NCGA was determined in two studies:

- A pharmacokinetic study in 12 healthy male adult volunteers who received a single oral administration of 100 mg/kg NCGA. This study was also performed to compare the non formulated pure powder that had been used in patients for years with the new dispersible tablet containing 200 mg NCGA, both products being ingested as a suspension. The reference powder and the tested tablet were bioequivalent. Although the formulation intended for marketing contains a lower amount of the excipient sodium laurilsulfate than the tablets used in the bioequivalence study (0.5 mg versus 2 mg), no additional bioequivalence study was felt necessary. Indeed, as 2 mg sodium laurilsulfate-containing tablets were found bioequivalent with the carglumic acid powder, there is no reason to believe the 0.5 mg sodium laurilsulfate-containing tablets would behave differently.  
A specific and highly sensitive assay method was developed and validated for NCGA determination using high pressure liquid chromatography coupled with tandem mass spectrometry.
- An *in vitro* metabolic study in human hepatocytes using <sup>14</sup>C labelled NCGA

### **Absorption and distribution**

Following single oral dose administration, NCGA peaks in plasma after few hours: median 3 hours (range 2 – 4), with a concentration reaching a median of 2.6 µg/ml (range 1.8 – 4.8).  
The apparent volume of distribution is very high: 2657 l (range 1616 – 5797).

There is no data on the oral bioavailability but it is expected to be low considering the high apparent volume of distribution and the apparent total clearance: median 5.7 l/min (range 3.0 – 9.7).

The inter-subject variability was moderate (around 30 %).

The linearity of the pharmacokinetics of NCGA has not been evaluated.

### **Metabolism and elimination**

The elimination profile appears bi-exponential with a first phase of rapid elimination followed by a second phase of slow elimination starting about 12 hours after the administration. Since there was no sampling time point between 12 and 24 hours (last sample) it was not possible to calculate the half-life of the second phase. In all subjects except one, NCGA was still detectable (above the limit of quantitation) at 24 hours but the levels were low as compared to the maximum concentrations (around 5%). Around 5 % of the dose (3.5 - 7.5) is recovered unchanged in urine within 24 hours. The fate of 95 % of the administered dose of NCGA is unknown. As for the *in vitro* study using rat hepatocytes no metabolite was detected using human hepatocytes. The value of the renal clearance, 290 ml/min (204 - 445), suggest that NCGA is probably secreted from the kidney. The applicant committed to perform a full ADME (absorption, distribution, metabolism and elimination) study on 3 volunteers to gather further information on the fate of carglumic acid in humans, the results of which will be submitted post-authorisation.

### Special populations

Plasma levels of NCGA were measured in children (2 days old to 10 years old) and 1 adolescent (13 years old), treated with a daily dose ranging from 7.4 to 122.4 mg/kg/day. Their range were consistent with those measured in healthy volunteers. The rather slow decrease in plasma concentrations over 15 hours suggests that a twice daily administration could be sufficient. Most young patients were treated between 2 and 4 times daily before meals or feedings. The applicant committed to gather further data in treated children to better define the pharmacokinetic profile of NCGA in patients.

### **Interaction studies**

No interaction studies have been conducted. In the clinical experience, NACG has been used in association with a number of other products, but due to the very low number of patients no conclusion could be drawn on the potential interaction.

### Clinical efficacy

The data presented to support the efficacy of NCGA in NAGS deficiency consisted of:

- a retrospective series of 20 patients (plus an update on 2 cases that started treatment in 2001) evaluable for safety – (not all have NAGS deficiency) of which 12 are evaluable for efficacy that received chronic treatment with NCGA (the 2 new cases are also confirmed or suspected NAGS deficient patients). The other 8 patients were treated for hyperammonaemia of another cause (5 cases with other urea cycle disorders due to defects in other enzymes associated with the urea cycle, including 3 carbamoylphosphate synthetase and 2 ornithine transcarbamoylase defects, and 3 cases of fatty acid oxidation disorder).
- Supportive data were submitted consisting of published data relating to 4 patients, including the first patient who was treated from 1980 until his death in 1990 with the product supplied by another pharmaceutical manufacturer before its distribution by Orphan Europe and three patients treated with a chemical grade product.

As the very first patient diagnosed as NAGS deficient, published in 1981, was treated with NCGA, there are no true historical controls with which to compare the efficacy.

### **Retrospective data collection**

The 20 patients included in the initial retrospective data collection have been treated by 10 physicians in 5 countries (France, Germany, Austria, The Netherlands and Spain).

### Population

The 12 patients evaluable for efficacy were 8 boys and 4 girls, born between 1984 and 2000 but all diagnosed after 1991:

- 6 patients were prospectively treated because they had a sibling who died from hyperammonaemia (including 3 siblings in one family and 2 siblings in another family),
- 3 patients had a neonatal presentation with clinical manifestations starting within the first 3 days of life but of various severity: persistent vomiting (1), neonatal hyperammonaemic coma (1), neurological manifestations (1);
- 3 patients had delayed onset presentation and late diagnoses at 13 years (recurrent episodes of vomiting with behavioural disorders and impaired consciousness but with a first similar episode at the age of 13 months), 4.5 years ("Reye-like" syndrome after a few months therapy with sodium valproate but with neurological manifestations since the age of 2.7 years), 12 years (recurrent episodes of vomiting with impaired consciousness but with a few behavioural manifestations since the age of 9 years), respectively.

The seriousness of the disease was classified according to that used in other urea cycle disorders, i.e. neonatal and late-onset presentations, since the enzymatic activity measured *in vitro* in the liver samples is too variable and unrelated with the clinical presentation to provide a reliable classification. The 12 patients corresponded to:

- a neonatal presentation in 6 cases (no 1, 2, 5, 6, 13, 20)
- a late-onset presentation in 3 cases (no 7, 9, 16).

Out of the 6 cases that were presented as "prospectively treated", 3 can actually be considered as a neonatal presentation since hyperammonaemia could be detected during early monitoring. As for the last 3 cases (no 8, 15, 18), a definite classification was not possible since hyperammonaemia did not occur under prospective treatment initiated right after birth.

### Diagnosis

The diagnosis was confirmed in all cases by liver biopsy and NAGS activity assay, which was performed in the same laboratory.

### Endpoints and measurement of efficacy

Data on plasma ammonia, plasma amino acids including glutamine, physical growth, psychomotor development, clinical symptoms of acute hyperammonaemic decompensation and survival have been recorded.

To reduce ammonia is one well-established treatment goal in urea cycle disorders, and therefore if NCGA is able to reduce ammonia, it is likely to have a role in treatment of NAGS deficiency. As for plasma glutamine concentration, this parameter may also be useful for the long-term monitoring and should be kept in the normal range by the treatment.

### Dose and dosing regimen

There was no dose ranging study. The doses used were thus empirical. The efficacy of the treatment with NCGA is always defined by fast normalization of plasma ammonia levels. The dose necessary to achieve this objective is highly dependent on the severity of the defect and on the protein load to the metabolic pathway, which are both individual characteristics of the patients. Therefore, the treatment should start with a high dose, in order to achieve the most rapid effect, and this dose should subsequently be tailored for each patient according to the evolution of ammonia.

**The doses initially used to start therapy (responsiveness test) were high, based on the first publication (300 mg/kg/day) but there was subsequently a trend to decrease them (ranged from 70 to 254 mg/kg/day). The objective of the test was to induce a decrease in plasma ammonium and/or glutamine, which is often elevated in case of nitrogen load. This occurred in most cases within 24 hours, but normalisation sometimes took a few days.**

This initial dose (per kg of body weight) was the maximum dose in all patients but one (no 8) whose dose was regularly increased during the first 4 months of therapy from 89 to 200 mg/kg/day without explanation of the treating physician.

Overall, the maximum dose received by each patient in the course of the treatment ranged from 35 to 254 mg/kg/day (median = 133 mg/kg/day).

The last dose (at the last visit) ranged from 7 to 98 mg/kg/day (median = 52 mg/kg/day), which represents an overall reduction of 60 %.

The frequency of administration also tended to decrease with age, from 3 - 6 at the beginning of the treatment to 2 - 4 at the last visit. In the absence of pharmacokinetics data, the logical approach was initially to divide the total daily dose according to the number of feedings and administer these doses before the feedings, since ammonia levels are known to peak usually about 1 hour after meals. But for practical reasons, the patients' mothers sometimes started to decrease the number of daily doses without any deleterious consequence.

The duration of treatment ranged at the cut-off date (July 31, 2001) from 9.8 to 0.7 years (median = 3.1 years). There was one discontinuation after one year of treatment due to limited efficacy. The compliance with NCGA treatment was good.

#### Concomitant therapy

**As opposed to other urea cycle disorders, concomitant treatment did not appear necessary. The classical alternate pathway medications (sodium benzoate and phenylbutyrate) were progressively withdrawn. Seven patients received arginine but this prescription seems to have been motivated by the experimental animal data and the publications on the first patient rather than by actually testing a potential additive effect on ammonia. The metabolic control in the five patients who did not receive any concomitant therapy did not differ from that of the other patients who received arginine. An exception to the general therapeutic approach should be mentioned: one physician prospectively treated 3 siblings with a combination of strict protein restriction (including the use of a mixture of essential amino acids), NCGA, sodium phenylbutyrate, arginine and carnitine and never attempted to liberalise the diet or reduce sodium phenylbutyrate.**

As for the diet, it was free or normal in 6 out of the 12 patients. The two older patients had a moderately restricted protein intake. Four patients had a restricted protein intake, which included a mixture of essential amino acids: the 3 siblings already described and one patient who also needed continuous enteral feeding.

In the large majority of treated patients, carglumic acid was the sole long term therapy.

#### **Results**

The clinical evolution of the cases presented are summarised in the table presented next page.

| Summary of the clinical evolution of the 12 patients diagnosed with NAGS deficiency and treated with NCGA |                |                        |        |            |             |                |                        |        |            |       |
|-----------------------------------------------------------------------------------------------------------|----------------|------------------------|--------|------------|-------------|----------------|------------------------|--------|------------|-------|
| Pat. N°; DOB; Type                                                                                        | Ammonia levels | Hyper-ammonemia crisis | Growth | Disability | M           | Ammonia levels | Hyper-ammonemia crisis | Growth | Disability | Death |
| 1; 1991; NN                                                                                               | ↑↑↑            | Y                      | ↓      | ?          | <b>9</b>    | ↑→             | 0                      | N      | 0          | 0     |
| 3; 1993; NN                                                                                               | ↑↑             | Y                      | ↓      | ?          | <b>0.65</b> | →              | 1*                     | N      | 0          | 0     |
| 5; 1995; NN                                                                                               | ↑↑↑            | Y                      | ↓      | ?          | <b>3</b>    | ↑→             | 0                      | N      | 0          | 0     |
| 6; 1996; NN                                                                                               | ↑↑             | 0                      | --     | --         | <b>0.07</b> | →              | 0                      | N      | 0          | 0     |
| 7; 1984; LO                                                                                               | ↑↑             | Y                      | N↓     | Y?         | <b>156</b>  | →              | 0                      | N      | 0          | 0     |
| 8; 1997; NN                                                                                               | →              | 0                      | N      | 0          | <b>3</b>    | →              | 0                      | N      | 0          | 0     |
| 9; 1993; LO                                                                                               | ↑↑↑            | Y                      | N      | Y          | <b>54</b>   | ↑              | 0                      | N      | Y          | 0     |
| 13; 1999; NN                                                                                              | ↑↑↑            | Y                      | --     | Y          | <b>0.14</b> | ↑→             | 0                      | N      | 0          | 0     |
| 15; 1999; NN                                                                                              | ↑→             | 0                      | N      | 0          | <b>0.5</b>  | ↑→             | 0                      | N      | 0          | 0     |
| 16; 1987; LO                                                                                              | ↑↑↑            | Y                      | N      | Y          | <b>144</b>  | ↑→             | 0                      | N      | 0          | 0     |
| 18; 2000; NN                                                                                              | ↑→             | 0                      | N      | 0          | <b>0.8</b>  | ↑→             | 0                      | N      | 0          | 0     |
| 20; 2000                                                                                                  | ↑↑             | 0                      | ↓      | 0          | <b>0.35</b> | →              | 0                      | N      | 0          | 0     |

\* when NCGA was stopped due to lack of availability

Ammonia Levels:

→ Normal

↑→ Normal with an occasional raise above Normal

↑ at least once above Normal

↑↑ at least once above 100 µmol/l

↑↑↑ at least once above 200 µmol/l

Growth:

N - normal

Disability:

Y – presence of symptoms or deficiencies that impact on quality of Life

0 – absence of symptoms or deficiencies that impact on quality of Life

M = Age at NCGA start (in month)

NN: neonatal; LO: late onset

Overall, plasma ammonia levels were well controlled, in spite of all intercurrent events that happened over the years (infections, vaccinations, surgical operations).

- In 5 patients, ammoniaemia was always found normal.
- In 3 patients, an abnormal value was measured on a single occasion: infection with high fever, very low NCGA dose (10 mg/kg/d), unknown cause in the last case.
- In 3 patients, there were transient fluctuations slightly above the upper normal value: only at the start of NCGA after a very fast reduction of the dose in the first patient; in 2 of the 3 siblings despite concomitant treatment.
- In one patient, fluctuating ammonia levels decreased from 120 - 160  $\mu\text{mol/l}$  before NCGA to 90  $\mu\text{mol/l}$  but never normalised.

In addition, in 6 of these patients, levels became normal in less than 24 hours.

From the sparse data supplied by the applicant, there does not seem to be any relationship between the dose of NCGA administered and the decrease in blood ammonia levels.

When plasma glutamine levels were elevated before NCGA therapy, sometimes even despite sodium benzoate or phenylbutyrate treatment, they rapidly normalised after NCGA was introduced and remained continuously normal over the years. When plasma citrulline levels were low in a few cases they normalised as well.

The raise in blood urea seen in 4 patients who had initially low levels may reflect a direct effect of NCGA on urea production or simply result from increased protein intake.

**Growth has been normal in all patients except one of the 3 siblings receiving drastic treatment. The boy's growth rate fell off during his second year of life and even his ammonia levels were borderline. The decision was taken to increase the protein intake in order to keep up with the boy's increase in energy expenditure and the boy subsequently caught up and regained a normal growth pattern.**

The psychomotor development was normal with normal school attendance for age in 11 patients. Only the patient who was treated for 1 year (patient no 9) was rated as mentally retarded already before NCGA therapy.

Out of the 12 patients with NAGS deficiency, no death was reported. The patient (no 9) who had his treatment discontinued after one year is still alive and treated with NCGA from another source.

Finally, none of the patients experienced any clinical symptoms of acute hyper-ammonaemic decompensation except for one patient. This was due to an accidental interruption of drug supply, which allowed to show the prolonged duration of action of NCGA since clinical symptoms of decompensation (poor appetite, vomiting, headache, somnolence, dizziness) were only manifest after 4 days. His ammonia level was measured at 94  $\mu\text{mol/l}$  with a glutamine level at 1242  $\mu\text{mol/l}$  ( $N = 580 \pm 180 \mu\text{mol/l}$ ); the ammonia concentration normalised within 4 hours after re-starting NCGA therapy (128 mg/kg administered over 8 hours).

### **Supportive data**

Besides the 12 patients that constitute the core series, 4 extra cases of NAGS deficiency are quoted from the literature:

- 1<sup>st</sup> patient diagnosed with NAGS deficiency and treated with NCGA (Bachmann 81, 82; Schubiger, 91) starting in 1980 at the 10<sup>th</sup> day of life
- Case of a NAGS deficient 20-year old patient in the United Kingdom who had been hospitalised on several occasions for hyperammonemia (Hinnie, 1997)
- Case of neonatal form of NAGS deficiency in the United Kingdom, with initial treatment at 4 months of age (Morris 1998)
- Case of neonatal form of NAGS deficiency in the Netherlands starting on NCGA at the 10<sup>th</sup> day of life (Huijmans, 1998)

The overview of the clinical evolution is presented in the table below.

| Pat. N°<br>DOB<br>Type     | Ammonia<br>levels | Hyperammo-<br>nemia crisis | Growth | Disability | M          | Ammonia<br>levels | Hyperammonemia<br>crisis | Growth | Disability | Death |
|----------------------------|-------------------|----------------------------|--------|------------|------------|-------------------|--------------------------|--------|------------|-------|
| Bachman<br>n<br>1980<br>NN | ?                 | Y                          | ?      | ?          | <b>0.3</b> | ?                 | 1                        | ↓      | Y          | 1     |
| Hinnie<br>20 y<br>LO?      | ?                 | Y                          | N      | Y          | <b>240</b> | →                 | 0                        | N      | Y          | 0     |
| Morris<br>?<br>NN          | ?                 | ?                          | N      | 0          | <b>4</b>   | ?                 | 1                        | N      | 0          | 0     |
| Huijmans<br>?<br>NN        |                   | Y                          | ?      | 0          | <b>0.3</b> | →                 | 0                        | N      | 0          | 0     |

Ammonia Levels:

→ Normal

Growth:

N - normal

Disability:

Y – presence of symptoms or deficiencies that impact on quality of Life

0 – absence of symptoms or deficiencies that impact on quality of Life

NN: neonatal; LO: late onset

#### References:

1. Bachmann C, Krahenbuhl S, Colombo JP et al. (1981); N-acetylglutamate synthetase deficiency: a disorder of ammonia detoxication [letter]; N Engl J Med, 304, 9, 543.
2. Bachmann C, Colombo JP and Jaggi K (1982); N-acetylglutamate synthetase (NAGS) deficiency: diagnosis, clinical observations and treatment. Advances in Experimental Medicine and Biology, 153, 39 - 45.
3. Hinnie J, Colombo JP, Wermuth B and Dryburgh FJ (1997); N-acetylglutamate synthetase deficiency responding to carbamylglutamate; J Inher Metab Dis, 20, 839 - 840.
4. Huijmans JGM, de Klerk JBC, Jankie R, Slotema R, Wolff ED and Duran M (1998); The fate of oral carbamylglutamate in N-acetylglutamate synthetase deficiency; J Inher Metab Dis, 21 (Suppl 2), 28.
5. Morris AAM, Richmond SWJ, Oddie SJ, Pourfarzam M, Worthington V and Leonard JV (1998) N-acetylglutamate synthetase deficiency: favourable experience with carbamylglutamate. J Inher Metab Dis, 21, 867 - 868.
6. Schubiger G, Bachmann C, Barben P, Colombo JP, Tönz O and Schüpbach D (1991); N-acetylglutamate synthetase deficiency: diagnosis, management and follow-up of a rare disorder of ammonia detoxication. Eur J Pediatr, 150, 353 - 356.

To further substantiate the efficacy of NCGA for the treatment of hyperammonaemia associated with NAGS deficiency, the applicant supplemented the application with additional analysis to establish how representative of the general population of NAGS deficiency patients these cases (12 + 4) are. For this analysis, the applicant compiled the data available from the retrospective cases, published literature and other available sources.

Over the last 20 years, between 1980 and 2001, 42 patients from 28 families have been identified with NAGS deficiency: 34 patients who were diagnosed as NAGS deficiency and 8 siblings who had previously died without precise diagnosis. Of the 34 patients identified, 22 patients have been treated with NCGA, of whom 14 received the product from the applicant (of these 12 who have been already described).

The difference in terms of mortality between treatments is shown in the table next page:

**Summary table: Distribution of the NAGS deficient population according to treatment**

| Patient (last) treatment          |              | No of patients | Previous treatment     |          |          | No of deaths |
|-----------------------------------|--------------|----------------|------------------------|----------|----------|--------------|
|                                   |              |                | Conventional treatment | Unknown  | None     |              |
| None (siblings)                   |              | 8              |                        |          |          | 8            |
| Conventional treatment            | Neonatal     | 3              |                        |          |          | 3            |
|                                   | late-onset   | 5              |                        |          |          | 2            |
|                                   | <b>total</b> | <b>8</b>       |                        |          |          | <b>5</b>     |
| Unknown                           | Neonatal     | 4              |                        |          |          | <b>4</b>     |
| Carglumic acid from the applicant | neonatal     | 7              | 3                      |          | 4        | 0            |
|                                   | late-onset   | 4              | 4                      |          |          | 0            |
|                                   | Prospective  | 3              | 3                      |          |          | 0            |
|                                   | <b>Total</b> | <b>14</b>      | <b>10</b>              |          | <b>4</b> | <b>0</b>     |
| Carglumic acid (other source)     | Neonatal     | 6              | 5                      | 1        |          | 1            |
|                                   | late-onset   | 1              | 1                      |          |          | 0            |
|                                   | Prospective  | 1              |                        | 1        |          | 0            |
|                                   | <b>total</b> | <b>8</b>       | <b>6</b>               | <b>2</b> |          | <b>1</b>     |
| <b>Total</b>                      |              | <b>42</b>      | <b>16</b>              | <b>2</b> | <b>4</b> | <b>18</b>    |

Data provided is strongly suggestive that carglumic acid has an important impact on the NAGS prognosis. The applicant committed to conduct a systematic follow-up of all NAGS deficient patients treated with Carbaglu to gather further efficacy information.

### **Clinical safety**

#### **Exposure**

Overall, twenty patients, 12 boys and 8 girls, started chronic NCGA treatment before 2001 and were included in the safety evaluation.

The duration of therapy at the cut-off date (July 31, 2001), ranging from 9.8 to 0.7 years (median = 3.6 years), was distributed as follows:

- > 3 years: 7
- 1 - 3 years: 8
- < 1 year: 5

At the cut-off date, the overall exposure to NCGA, whatever the indication, amounted to a total of 64.5 patient-years.

The age of the patients when NCGA was started ranged from 1 day to 13 years and was distributed as follows:

- < 1 month: 10
- 1 month - 1 year: 4
- > 1 year: 6

The age of the patients when NCGA was discontinued or at the cut-off date ranged from 5 months to 17 years and was distributed as follows:

- ♦ < 1 year: 4
- ♦ 1 - 3 years: 6
- ♦ 3 - 6 years: 5
- ♦ > 6 years: 5

### Discontinuation

Out of the 20 patients, 15 patients were still on treatment at the cut-off-date and the reason for discontinuation in the other 5 patients were the following:

- ◆ limited efficacy in one NAGS deficient patient
- ◆ poor compliance and/or questionable efficacy in the long term: 3 patients with other urea cycle disorders
- ◆ death in one fatty acid oxidation disorder

### Adverse events

In patients receiving a normal dosage of NCGA no adverse events have been reported that could be attributed with certainty to the compound. Increased sweating in hot weather was reported in one patient, when she was about 1 year old and receiving around 150 mg/kg/day of NCGA.

### Laboratory findings

Results of routine laboratory tests are available for 18 patients; these include haematology (blood counts, haemoglobin, haematocrit, prothrombin time) and blood chemistry (usually transaminases, bilirubin, urea, creatinine, total protein, glucose, electrolytes, iron).

The most frequent abnormality was anaemia, which was frequently related to iron deficiency, especially when patients were on hypoprotidic diet, but also in other patients to a possible thalassaemic trait or frequent blood samplings. The other abnormalities were transient low electrolytes levels related to episodes of gastroenteritis or frequent vomiting. None of the abnormalities could be related to treatment.

### Published experience

In the first patient (Bachmann, 82), a dose range finding was attempted and the dose was progressively increased to 750 mg/kg/day. This led to the clinical picture of a sympathomimetic-like reaction, with tachycardia, profuse sweating, bronchial hypersecretion, increased body temperature and persistent crying. In the absence of other cases reported, the causal relationship with the NCGA cannot be assessed. In the same patient, increased transaminases were detected on several occasions. This patient died after 9.5 years of treatment with NCGA.

### Safety related to interactions with other medicinal products

NCGA has been used in association with a number of other medicinal products, but due to the very low number of patients no conclusions can be drawn and it still remains very important to further document associations with the same and with other medicinal products.

## **5. Overall conclusions, benefit/risk assessment and recommendation**

### Quality

The quality of the product is considered to be acceptable when used in accordance with the conditions defined in the Summary of Product Characteristics. In general, satisfactory chemical and pharmaceutical documentation has been submitted for marketing authorisation. There are no major deviations from EU and ICH requirements.

The active substance is well characterised and documented. The excipients are commonly used in this kind of formulation and the packaging material is well documented. The manufacturing process of the finished product has been adequately described.

Stability tests indicate that the product under ICH guidelines conditions is chemically stable for the proposed shelf life.

At the time of the CPMP opinion a number of minor quality issues were unresolved. The applicant gave a commitment to resolve them as post authorisation follow up measures:

### **Preclinical pharmacology and toxicology**

NCGA is an aminoacid, structurally analogue of NAG, which is the naturally occurring activator of CPS, the first enzyme involved in the urea cycle. The mechanism of action of NCGA was clearly established and its effect on hyperammonaemia models (normal or partially hepatectomised rats receiving potentially lethal doses of ammonium) was demonstrated *in vivo*. Safety pharmacology studies did not show any significant effect on respiration, central nervous system and cardiovascular system.

The pharmacokinetics profile in rats is characterised by a rapid absorption, following oral administration of NCGA, a gender independent T<sub>max</sub> between 2 and 4 hours. The metabolic profile evaluated *in vitro* and *in vivo* did not reveal relevant metabolite formation. NCGA was mainly eliminated via urine and faeces.

In reviewing the toxicological data available, no overt toxicity of NCGA was noted in rats. NCGA was non genotoxic. The reproduction toxicology studies programme has not yet been finalised. The preliminary results showed no evidence of embryotoxicity, foetotoxicity or teratogenicity. The final results will be submitted as part of specific obligations to be fulfilled post-authorisation. The absence of carcinogenic studies was justified.

Overall, the pharmacodynamic and toxicological characterisation of Carbaglu, although not extensive, did not identify/anticipate relevant concerns in relation to its use in humans. The conduct of reproductive toxicology, the results of which will be submitted post-authorisation will contribute to a more solid conclusion.

### **Clinical efficacy and safety**

The pharmacokinetic profile has been evaluated in adult healthy volunteers. It is characterised by a rapid absorption, wide distribution and a very low 24h urinary excretion of the dose as unchanged (around 5 %), after a single oral administration on 100 mg/kg. Considering the sparse data, the applicant committed to gather further data to gain more insight into the fate of NCGA in healthy volunteers as well as in treated children. These data will be submitted post-authorisation.

There was no dose ranging study, but the dose selection was based on the fast normalisation of plasma ammonia, which reflects the efficacy of the treatment. Based on the clinical experience, the treatment should start with a high dose, in order to achieve the most rapid effect, and this dose should subsequently be tailored for each patient according to the evolution of ammonia. The initial dose recommended in the Summary of Products Characteristics is therefore 100 mg/kg up to 250 mg/kg, if necessary.

The data presented to support the efficacy of NCGA in NAGS deficiency consist of a retrospective series of 20 treated patients evaluable for safety (not all have NAGS deficiency) of which 12 were evaluable for efficacy. The NAGS deficient patients had either a neonatal or late onset presentation of the disease and were treated for a median of 3.1 years with NCGA. In most patients ammonia levels were normalised after introduction of treatment. Growth was normal in all patients and only one was suspected of being permanently disabled already before starting NCGA therapy. Patients on this treatment could be maintained stable without much diet restriction or concomitant therapies. Published data relating to 4 patients have been provided to further support the use of NCGA.

Given the rarity of the disease (over the last 20 years, 42 patients from 28 families have been identified with NAGS deficiency by the applicant), one cannot expect to prove efficacy through classical controlled clinical trials, however, it has been shown that NCGA has an important effect size on the disease being treated. Additional analysis on the prognosis of the disease, which was based on

the retrospective data collection, published literature and data from other sources, provided insight on the effect size in terms of mortality in favour of NCGA compared to no treatment or conventional therapy which can be considered large enough in view of the lack of a controlled study. Indeed, death was reported in all patients without treatment compared to 5 patients out of the 8 who received conventional treatment and compared to none with NCGA produced by the applicant. The applicant committed to conduct a systematic follow-up of all NAGS deficient patients treated with Carbaglu to gather further efficacy information.

Overall the safety data derived from 20 patients, who received NCGA and have been treated for a median of 3.6 years. NCGA seems to be well tolerated and no major safety concern has been identified. Cases of increased sweating and increased transaminases levels have been reported.

Overall, considering the rarity and the severity of the disease, the safety data, although sparse, can be considered sufficient for the granting of a marketing authorisation. However for precautionary measures, systematic surveillance of liver, renal, cardiac functions and haematological parameters is recommended as mentioned in the Summary of Products Characteristics. In addition, the applicant committed to conduct a systematic follow-up of all NAGS deficient patients treated with Carbaglu to gather further safety information.

### **Benefit/risk assessment**

Taking into account that NAGS deficiency is a rare disease with limited treatment options and with potentially devastating consequences, the CPMP considered that despite the limited data available with respect to pre-clinical and clinical sections of the application, Carbaglu offers an acceptable benefit/risk ratio for the treatment of hyperammonaemia due to NAGS deficiency.

Considering that comprehensive data on the efficacy and safety cannot be provided under normal conditions of use because of the rarity of the disease, the CPMP recommended the granting of the marketing authorisation under exceptional circumstances in this indication. The applicant committed therefore to provide additional quality, preclinical and clinical data as highlighted previously. The applicant committed also to conduct a post-marketing follow-up programme to systematically follow all NAGS deficient patients treated with Carbaglu in order to gather further information on its efficacy and safety in these patients. These data, which will be provided post-authorisation will form the basis of the annual reassessment of the benefit/risk profile of Carbaglu.

### **Recommendation**

Based on the CPMP review of data on quality, safety and efficacy, the CPMP considered by consensus that the benefit/risk profile of Carbaglu was favourable and therefore recommended the granting of the marketing authorisation under exceptional circumstances for the treatment of hyperammonaemia due to N-acetylglutamate synthase deficiency.