SCIENTIFIC DISCUSSION

1. Introduction

The hepatitis B virus (HBV) disease represents a major public health concern, with more than 350 million people infected and about 1 million deaths annually. The prevalence of HBV varies widely by geographic area, ranging from 0.1-2% in low prevalence areas such as Western Europe (primarily acquired through horizontal transmission during adulthood) to 8% and more in the high prevalence areas of South-East Asia and Sub-Saharan Africa (usually acquired through vertical transmission).

HBV is a small DNA virus belonging to the hepadnaviridae family. The infectious enveloped particle has an outer protein coat (hepatitis B surface antigen or HBsAg) and an inner protein core (hepatitis B core antigen or HBcAg). In infected hepatocytes, HBcAg is produced in excess and during export from the hepatocytes is cleaved to release a hepatitis B e antigen (HBeAg). HBV has a high rate of spontaneous mutation. A mutation in the pre-core region of the gene coding for the nucleocapside stops the synthesis of the HBeAg. Such HBeAg negative or pre-core mutants patients, which account for 7 to 30 % of infections worldwide is particularly common in Southern Europe and Asia.

HBV infection is a complex disease entity that may either resolve spontaneously or manifest itself in variety of ways. Following acute hepatitis B infection, approximately 5% of adults and 20-90% of children, depending on the age at infection, fail to produce adequate immune response and become chronic carrier of the virus. Chronic carriers of HBV are at increased risk of developing long-term complications, i.e. cirrhosis, hepatic failure and hepatocellular carcinoma (HCC). Among patients with chronic active hepatitis B, some 40% will develop cirrhosis over their lifetime at a rate of approximately 2% per year. Among patients with compensated cirrhosis, 10% per year progress to a decompensated state, with a 1-year survival rate of 60%, compared with over 90% for compensated cirrhosis.

The ultimate goal of treatment of chronic hepatitis B is to suppress HBV replication and to induce remission in liver disease before cirrhosis and HCC develop. There are however remaining scientific uncertainties (e.g. the level of viraemia associated with sustained virological response, with HBeAg seroconversion, when to stop safely the treatment either when viral eradication or when sustained suppression of viral replication necessary to prevent progression of disease).

There are currently three approved therapies for chronic HBV infection in Europe: alfa-interferon, lamivudine and adefovir dipivoxil, the prodrug of adefovir. Recombinant alfa-interferon acts primarily as an immunomodulator, whereas the nucleoside/nucleotide analogues directly inhibit viral replication. Current therapy of chronic hepatitis B has limited long-term efficacy and potential drawbacks. Therefore, there remains a great medical need for new therapeutic options for naïve patients as well as for patients with lamivudine-resistant HBV and for the more difficult to treat population (e.g. HBeAg negative patients, HIV co-infected patients and patients with decompensated hepatitis).

The present application for marketing authorisation of Baraclude is made under Article 8.3 (i) and concerns a new active substance, entecavir for which a complete dossier has been submitted. Entecavir (ETV) is a nucleoside analogue of guanosine with selective activity against HBV.
The approved indication at the recommended dose of 0.5 mg once daily in nucleoside naïve patients or 1 mg once daily in lamivudine-refractory patients is: "for the treatment of chronic hepatitis B virus (HBV) infection in adults with compensated liver disease and evidence of active viral replication, persistently elevated serum alanine aminotransferase (ALT) levels and histological evidence of active inflammation and/or fibrosis. This indication is based on clinical trial data in patients with HBeAg positive and HBeAg negative HBV infection, nucleoside naive patients and patients with lamivudine-refractory hepatitis B".

2. Quality aspects

Introduction

Baraclude is available in two pharmaceutical forms i.e. film-coated tablet and oral solution.

The film-coated tablets contain 0.5 mg and 1 mg of entecavir anhydrous, as active substance. The two strengths can be differentiated by their colour and debossings.
The other ingredients include:
- Tablet core: crospovidone, lactose monohydrate, magnesium stearate, cellulose, microcrystalline and povidone.
- Tablet film-coating: titanium dioxide (E171), hypromellose, macrogol 400, polysorbate 80(for the 0.5 mg strength only) and iron oxide red (for the 1 mg strength only).

The tablets are packed in HPDE bottle with child resistant polypropylene closure and in Alu/Alu perforated unit dose blisters.

The oral solution contains 0.05 mg/ml of entecavir, as active substance.
The other ingredients include maltitol liquid (E965), sodium citrate dihydrate, citric acid anhydrous, methylhydroxybenzoate (E218), propylhydroxybenzoate (E216), orange flavour, sodium hydroxide, hydrochloric acid and purified water.
The oral solution is presented in a multidose HDPE bottle with child-resistant polypropylene closures. A polypropylene measuring spoon with millilitre marks from 1 millilitre up to 10 millilitres is provided.

Active Substance

Entecavir is a nucleoside analogue of guanosine.
It is a crystalline, non-hygroscopic, white to off-white powder and it is isolated as the monohydrate. Its aqueous solubility is pH dependent (higher at pH values above 8.7 or below 3.4). Entecavir contains three chiral centres, but it is synthesised solely as the 1S, 4S, 3R enantiomer. A wet granulation is used in the finished product manufacturing process, which minimises the impact of the physical characteristics (particle size, shape and surface area) of the active on the content uniformity of the tablets. Entecavir crystalline form has shown not to be affected during manufacture and storage.

- Manufacture

Entecavir is prepared from two commercially available starting materials. Satisfactory specifications and associated methods have been provided for the starting materials, intermediates, reagents and solvents.

Process impurities originating from each starting materials/reagents and solvents and during the synthesis have been adequately discussed.

A number of synthetic processes (A, B and C) have been used to produce batches used in non-clinical/clinical studies. Compared to the commercial process F, process A and B used different reagents, solvents and intermediates, while process C used the same sequence and the same intermediates but with some minor difference regarding the reagents and the working procedures.
No significant differences between lots obtained by the different processes have been noted, especially in term of impurity profile.

- Specification

The active substance specification includes tests for appearance, colour, identity (IR, HPLC), optical rotation, assay (HPLC), impurities (HPLC) and water content.
The HPLC method used to control the impurities allows detection of the diastereomeric impurities that may be present in the active. Concerning the enantiomeric purity of entecavir as well as residual solvents, heavy metals and sulphated ash, it has been found acceptable not to test them based on the justifications and data provided. Impurity limits in the specification are justified by toxicology studies.

Batch analysis data provided confirm satisfactory compliance and uniformity with the proposed specification.

- Stability

Under accelerated conditions (40°C/75% RH - commercial packaging) and long-term conditions (25°C/60% RH - commercial packaging), respectively 6-month data and up to 18-month data have been provided for three batches manufactured according to the synthesis route at the commercial site. The parameters tested included appearance, assay, impurities and water content.
A photostability study has been performed and it showed that entecavir is not light sensitive.
The proposed retest period of 24 months is supported by the presented data when entecavir is stored under the described conditions.

Medicinal Product

Film-coated tablets

- Pharmaceutical Development

This pharmaceutical form is of standard formulation and has been developed to release the active rapidly prior to absorption.

The excipients have been selected based on their compatibility with entecavir. They are all of PhEur quality except the film-coating, which is satisfactorily controlled according to a different standard. Lactose was chosen as an excipient as it is a common diluent for wet granulation. Regarding the TSE risk, the lactose monohydrate from milk of bovine origin has been considered in compliance with current TSE requirements. The magnesium stearate is from vegetable origin.
Satisfactory specifications have been defined to control the primary packaging materials.

A capsule dosage form was used in Phase I and II clinical studies and later the 0.5 mg and 1 mg film-coated tablets were developed as the proposed commercial formulations. Bioequivalence has been demonstrated between the tablets and capsules (see clinical section).

- Manufacture of the Product

The manufacturing process consists of the standard following operations: wet granulation of the active with the excipients, milling, blending, compression, film-coating and packaging.
Satisfactory in-process controls have been defined.
Validation data provided for two commercial-scale batches of the 0.5 mg strength and for one commercial-scale batch of the 1 mg strength confirm robustness and reproducibility of the manufacturing process.

- Product Specification

The product specification includes tests controlled by validated methods for appearance, identity (HPLC), assay (HPLC), degradation products (HPLC), dissolution, uniformity of dosage units (PhEur) and microbial limits (PhEur).
Batch analysis data provided for both strengths comply with the specifications and indicate consistent and reproducible manufacture.

- Stability of the Product

Stability data have been provided for 3 batches of 0.5 mg and 1 mg film-coated tablets packed in aluminium blister and for three batches of 0.1 mg and 1 mg tablets in HDPE bottle bracketing the 0.5 mg strength.

18-month data are available under long-term conditions (25°C/40% RH - commercial packaging) and under intermediate conditions (30°C/70% RH - commercial packaging). Under accelerated conditions (40°C/75% RH - commercial packaging) 6-month data have been provided.

The parameters tested included appearance, assay, hardness, dissolution, water content and microbial testing. The observed changes were small, and not likely to have a significant effect on efficacy and safety of the product when used according to the directions in the Summary of Product Characteristics. Photostability studies have shown that the finished product is non-light sensitive.
The data provided support the proposed shelf life and storage conditions as defined in the Summary of Product Characteristics.

Oral solution

- Pharmaceutical Development

The oral solution was developed to provide an alternate oral dosage form for patients who cannot swallow tablets and for patients who need dose reduction.

All the excipients have been chosen based on their compatibility with the active substance. A hydrogenated sweetener i.e. maltitol has been selected due to chemical incompatibility of entecavir with sucrose hydrolysis products. The amount incorporated in the formulation has been shown to be sufficient to mask the potential bitter taste of entecavir in solution.
The pH 6 selected for the formulation, and maintained by a citrate buffer, is satisfactory for the antimicrobial efficacy of the preservatives (methylhydroxybenzoate and propylhydroxybenzoate), which has been demonstrated according to PhEur. Moreover, this pH ensures a good chemical and physical stability of entecavir when the oral solution is stored under the recommended storage conditions.
All the excipients are of PhEur quality, except the orange flavour, which is controlled according to an acceptable standard and complies with EU requirements. With regards to the TSE risk, the oral solution does not contain any component is of ruminant origin.
Satisfactory specifications have been provided for the HDPE bottle and the child-resistant polypropylene closure.
The compatibility of the solution with the primary packaging has been satisfactorily demonstrated. The dosing spoon is CE marked and has been approved for its intended use. The accuracy and reproducibility of the dose delivered by this medical device has been satisfactorily demonstrated. The viscosity of the solution has been tested on commercial scale batches and it has been found sufficiently low to allow dose measurement.

The formulation used in clinical studies is identical to that proposed for marketing with exception of the solution fill volume. Bioequivalence between the oral solution and the film-coated tablets has been demonstrated (see clinical section).

- Manufacture of the Product

The manufacturing process consists of the standard following operations: dissolution of the active and of the excipients in purified water, pH adjustment, filtration and filling in the primary packaging.
A satisfactory maximum holding time has been defined for the bulk solution based on chemical and microbiological testing. Satisfactory in-process controls have been defined.
Validation data provided confirm robustness and reproducibility of the manufacturing process.

- Product Specification

The product specification includes tests controlled by validated methods for appearance, identity (HPLC), assay (HPLC), preservative content (HPLC), degradation products (HPLC), pH and microbial limit (PhEur).
Batch analysis data have been provided for 5 production-scale batches comply with the specifications and indicate consistent and reproducible manufacture.

- Stability of the Product

Stability data have been provided for 3 batches manufactured using the commercial process at the commercial manufacturing site. HDPE being a semi-permeable container, low-humidity ICH conditions are used for stability testing. Under long-term conditions: (25°C/40%RH – commercial packaging) and under accelerated conditions (40°C/25%RH – commercial packaging), respectively 2-year and 6-month stability data have been provided. 15-month data under intermediate storage conditions (30°C/35%RH – commercial packaging) have been provided as well.
The parameters tested included appearance, assay, degradation products, pH, preservatives content, microbial limit and weight loss.
The observed changes were small, and not likely to have a significant effect on efficacy and safety of the product when used according to the directions in the Summary of Product Characteristics.
Photostability studies have shown that the finished product is light sensitive. The secondary packaging appears to protect sufficiently the product from light.
A satisfactory in-use stability study has been performed.
The data provided support the proposed shelf life, in-use shelf life and storage conditions as defined in the Summary of Product Characteristics.

3. Non-clinical aspects

Introduction

Entecavir is a guanosine nucleoside analogue with selective activity against hepatitis B virus (HBV). The non-clinical programme consisted of a series of studies aiming to define the pharmacological, pharmacokinetics and toxicological profile of entecavir.
All pivotal non-clinical studies were conducted in accordance with principles of Good Laboratory Practices (GLP).

Pharmacology

- Primary pharmacodynamics (*in vitro/in vivo*)

Entecavir is phosphorylated to the active triphosphate form (ETV-TP) by cellular nucleoside kinases. This was shown in different cell systems including infected hepatoma cell lines but the extent of phosphorylation of entecavir was dependent on cell-type. The triphosphate competes with the natural substrate, deoxyguanosine triphosphate (dGTP), of human HBV polymerase. *In vitro* studies on mechanism of action indicated that the three enzymatic activities of the viral polymerase were inhibited 1) priming activity, 2) reverse transcriptase activity resulting in first strand DNA synthesis and 3) DNA dependent DNA polymerase activity resulting in second-strand DNA synthesis.

There were no differences in the phosphorylation capacity of entecavir using normal or chronically infected hepatoma cell lines. The entecavir triphosphate had an intracellular half-life of approximately 15 hours. The inhibition constants for ETV-TP (Ki) were 1.02 and 2.6 nM against the human and woodchuck hepatitis polymerase, respectively, when the Km for dGTP was in the range of 8.4-20 nM. The IC_{50} for HBV polymerase as well as for woodchuck hepatitis virus was 1.6 nM. Further, the data suggest that concentrations of entecavir that inhibit the HBV polymerase have little if any inhibitory

activity against cellular polymerases. While the K_i of ETV-TP for HBV DNA polymerase was 0.0012 µM corresponding values for cellular DNA polymerases α, β, and δ ranged from 18 to 40 µM. The antiviral activity of entecavir was studied in cell systems stably expressing HBV (HepG2.2.15) as well as in systems transiently expressing HBV. The effective concentration for inhibition of 50 % of virus yield (EC_{50}) ranged from 3.6 to 3.75 nM. In comparison the activity of lamivudine showed a 10-fold range depending on cell system used (EC50s ranging from 116 to 1600 nM) and adefovir EC_{50s} ranged widely from 580 to 2500 nM. The different methodology and assays used are likely to explain these differences. The inhibition constants for ETV-TP (Ki) were 1.02 and 2.6 nM against the human and woodchuck hepatitis polymerase, respectively, when the Km for dGTP was in the range of 8.4-20 nM. These results suggest that entecavir potently and specifically inhibits the HBV polymerase and that activity against cellular polymerases is unlikely at clinically relevant concentrations. The IC_{50} for HBV polymerase as well as for woodchuck hepatitis virus was 1.6 nM.

A large data set, including clinical isolates and using standardised methodology, showed that the median EC_{50} value for entecavir against lamivudine resistant HBV (rtL180M, rtM204V) was 0.026 µM (range 0.010-0.059 µM) i.e 8 x higher concentration than required for inhibition of wild-type virus. YMDD (tyrosine, methionine, aspartate, aspartate motif) mutant polymerases thus appeared to retain sensitivity towards entecavir.

With regard to cytotoxicity in human liver cells, concentration of entecavir that resulted in cytotoxicity of 50% of cells was 30 µM, a value much higher than the active antiviral level.

Both entecavir and lamivudine seemed to non-specifically inhibit cellular metabolic activity (decreases in lactate and ATP). However, high exposures of entecavir had no relevant adverse effects on γ polymerase or mitochondrial DNA synthesis in HepG2 cells ($K_i > 160$ µM). Entecavir contains a 3'-OH moiety and may be incorporated into cellular DNA and is considered to act as a *de facto* chain terminator in similarity to some other nucleoside analogues. The extent of incorporation showed a wide range depending on cell type, with low incorporation in human fibroblasts and high in hepatoma cells.

In vivo studies were conducted in woodchucks and duckling, species that are susceptible to infection with hepatitis virus. Oral administration of ETV at doses ranging from 0.02 to 0.5 mg/kg once daily for 4 weeks in woodchucks chronically infected by hepatitis virus resulted in 2 to 3 log_{10} reductions in serum WHV DNA levels. After daily administration leading to viral suppression, a weekly regimen at 0.5 mg/kg over 3 years maintained viral DNA at undetectable level. Antiviral activity of entecavir was reported in ducklings with up to 99% reduction in serum viral DNA but there was a rebound 2 weeks after treatment had stopped. Antiviral activity of entecavir in duck hepatocytes hepatitis B virus was represented by an EC_{50} value of 0.13 nM to compare with an EC_{50} value of 138 nM for lamivudine.

- Secondary pharmacodynamics

Entecavir did not show any significant potential for interactions in an in vitro receptor/channel ligand binding screen or enzyme-system assays at concentrations up to 100 µM. Unlike lamivudine or adefovir, ETV has no relevant activity against HIV-1 ($EC_{50} > 10$ µM).

- Safety pharmacology

A battery of general pharmacology studies was conducted in mice and dogs prior to the systematic use of GLP. In mice administered with ETV dose up to 8 mg/kg for 7 days, a transient impairment of coordinated movement was observed at all doses on Day 1 but not on Days 3 or 7, and there was no effect on spontaneous activity at any time point. Entecavir had no effect on cardiovascular and respiratory parameters however the study was conducted in anaesthetised dogs with doses up to 2 mg/kg.

There were no effects of entecavir on hERG channel current when used at concentrations of 3, 10 and 30 µM. The study was repeated under GLP conditions and similar results were reported.

Entecavir was also tested for effects on cardiac L-type channel current in canine ventricular myocytes at levels up to 30 µM. No biologically significant effects were reported. In a canine Purkinje fiber

assay entecavir at concentrations up to 30 μM had no biologically relevant effects on action potential parameters. In the rabbit Purkinje fiber assay (stimulation at 1 and 3 Hz), entecavir at concentrations up to 30 μM had minimal effects on APD_{90}.

Subsequently, a battery of GLP oral safety pharmacology studies with ETV was conducted. These studies did not reveal any effects of entecavir on the central nervous system (CNS) in male rats at single oral doses up to 8 mg/kg, on the cardiovascular system in male dogs at single oral doses up to 1 mg/kg and on the respiratory system in male rats at single oral doses up to 8 mg/kg.

All together, these results indicate that little potential for undesirable pharmacologic or cardiac electrophysiologic effects would be expected at therapeutic concentrations of ETV.

- Pharmacodynamic drug interactions

In vitro, co-administration of stavudine, didanosine, abacavir and zidovudine had no effect on HBV assays containing entecavir at the EC_{50}. Entecavir at 0.8 to 4.2 x C_{max} had no significant effects on EC_{50} values of stavudine, didanosine, abacavir, zidovudine, lamivudine and tenofovir in cells infected with three HIV strains.

Pharmacokinetics

Pharmacokinetic characteristics of entecavir were studied in the species used in toxicological studies (mice, rats, rabbits, dogs, and monkeys). Additional studies were conducted in the woodchuck and the duck, which were used as animal models of anti-HBV activity. Toxicokinetic evaluations, in support of pivotal toxicology studies, were conducted in compliance with GLP. The methods of analysis used to assay entecavir were adequate and validated.

- Absorption- Bioavailability

Entecavir was rapidly and extensively absorbed following oral administration. A high bioavailability was evident in most species (> 80 % in rats and dogs). In contrast the bioavailability was low in monkeys (16 %), due to poor absorption but not related to first pass metabolism.

Systemic exposure was dose-related but not always dose proportional in all species. Exposure to entecavir was higher in male rats than in female rats, which was attributable to a greater extent of metabolism in female rats. There were no sex-related differences in mice, dogs, and human.

- Distribution

Protein binding was low, ranging from 8% in mice to 24% in dogs, and distribution was extensive. The steady-state volume of distribution was 4.6, 1.6 and 1.0 l/kg in rats, dogs and monkeys, respectively, suggesting extensive extravascular distribution and/or preferential binding in tissues. In mice, 0.5 hours after a single oral dose of 10 mg/kg, highest labelled entecavir was detected in bladder, stomach, kidney, small intestine and spleen. In rat, 1 hour after a single oral dose of 10 mg/kg, ^{14}C-label was initially highest in bladder, liver, kidney, lymph nodes, large intestine, prostate, and bone marrow. Label was also detected in the maternal cerebrum of pregnant rats and there were data showing that the compound may pass through the blood/brain barrier in mice, dogs and monkeys. Studies in pregnant rats showed passage across the placenta and substance was excreted in milk of lactating animals within 1 hour after oral administration.

The in vitro red blood cell (RBC) distribution of radioactivity of entecavir was 49 %, 1.6 %, 50 % and 52 % in rats, dogs, monkeys and humans respectively indicating that entecavir was uniformly distributed in plasma and RBCs except in dog blood.

- Metabolism (*in vitro/in vivo*)

Metabolism appeared limited in all species and seemed to proceed via sulphation and glucuronidation pathways. Overall, metabolites accounted for a limited fraction in plasma, urine and faeces in all species. The highest extent of metabolism was in rats where total metabolites (in urine and faeces)

accounted for about 10 % in males and 30 % in females. This difference was attributed to differences in sulphotransferase activities. In humans though, up to 29% of plasma radioactivity was metabolites. These were mainly glucuronides representing phase II metabolites. Two sulfate conjugates (M2 and M3) and 3 glucuronide conjugates (M1, M4, and M5) of entecavir were detected in male and female rats and male monkeys. In dogs, only the glucuronides M1, M4, and M5 of entecavir were detected. In addition, entecavir was not metabolised in mouse lung microsomes, mouse and rat lung homogenates, and mouse and rat lung S9 fraction. Although metabolite production was limited in species used in toxicology studies, the high exposures achieved indicate that production of metabolites was sufficient to ensure characterisation of any potential adverse effects/safety hazards coupled to exposure to metabolites.

In vitro entecavir was not a substrate, inducer or inhibitor of human cytochrome P450 isozymes. It is therefore anticipated that the pharmacokinetics drug-drug interaction with entecavir in humans is unlikely.

- Excretion

Excretion was primarily via urine in the form of unchanged compound in all species with the exception of monkeys, where excretion mainly proceeded via faeces. Unchanged compound in faeces was probably unabsorbed compound.

In rats, monkeys, and humans, the values of renal clearance were greater than the values of glomerular filtration rate (GFR) suggesting that ETV is eliminated through a combination of passive (glomerular filtration) and active (net tubular secretion) processes by the kidney.

The overall recovery of total radioactivity was about 90% in rats, dogs and monkeys over the period collected.

The terminal half-life (T1/2) values following oral administration were 2.2 hours in rats, 22.7 hours in dogs, 27.4 hours in monkeys, and approximately 135 hours (mean value at dose levels of 0.1 mg, 0.5 mg, and 1.0 mg) in humans. The marked difference in T1/2 values reported between animals and humans is most likely due to a combination of several factors including differences in the sensitivity of the assays used in the studies, and differences in the duration of sampling from which these values were determined. There was no indication of systemic accumulation of entecavir after repeated administration in rats and monkeys. There was low to moderate accumulation in dogs (1.1- to 2.5-fold increase) and in humans (1.6 to 2.7 -fold increase in the dose range), despite prolonged elimination.

ETV was shown not to be a substrate of human P-gp, using Caco-2 cell monolayers and is therefore unlikely to be involved in drug-drug interactions with other drugs that are P-gp substrates or inhibitors.

Toxicology

A comprehensive toxicological programme was conducted in mice, rats, dogs and monkeys.

- Single dose toxicity

In mice and rats, entecavir showed minimal acute toxicity and oral doses up to 200 mg/kg were well tolerated. Doses ≥ 1000 mg/kg were overtly toxic.

- Repeat dose toxicity (with toxicokinetics)

Studies were conducted in mice up to 6 months, in rats up to 6 months, in dogs up to 3 months and in monkeys up to 12 months. In some of the rodent studies, higher doses of entecavir were administered by diet instead of gavage resulting in higher exposures. Target organ toxicity in diet and gavage studies seemed comparable and only the latter studies are considered below for the main findings.

Doses as low as 0.02 mg/kg/day, corresponding to approximately clinical exposure at a dose of 1 mg, caused liver toxicity in rats, characterised as liver degeneration in a 6 month study. In other rat studies, as well as in a 6 month mouse study, increases in AST and ALT, single cell necrosis, hypertrophy and liver degeneration were recorded. Liver tumours were noted in high dose females in the rat

carcinogenicity study. Liver changes in dogs occurred at very high doses and appeared primarily related to poor condition.

In the 1-month dose ranging study in monkey, there was hepatocellular vacuolation (minimal in 1 female at 5 mg/kg and mild in the remaining monkeys at 25 mg/kg), not associated with any evidence of liver dysfunction, whereas no drug-related changes suggestive of an effect on the liver were observed in the 1-year monkey study with doses up to 40 mg/kg, corresponding to high systemic exposure levels (AUC) in relation to expected clinical levels.

In 6-month studies in mice, decreased testes weights were observed at ≥ 5 mg/kg, whereas seminiferous-tubular degeneration was observed only at the overtly toxic doses of 10 and 20 mg/kg. In rats, effects on the testes were limited to decreased testis weight and size at 15 mg/kg. In dogs, decreased weight of the testes was observed at 0.3 mg/kg and seminiferous-tubular degeneration was observed at ≥ 3 mg/kg (0.3 and 3 mg/kg were non-overtly toxic doses). These changes showed evidence of reversibility at 15 mg/kg following a 3-month postdose period. The testis was not a target organ in monkeys. At the threshold dose for microscopic changes in the testes, systemic exposure to entecavir was high in relation to expected clinical exposure.

Kidney toxicity was described as nephropathy at doses from 10 mg/kg/day in the 6 month mice study, nephrosis in rats at 150 mg/kg/day in the 2 week study, nephrosis and thrombosis at doses of 100/50 mg/kg/day in the 2 week dog study and as degeneration of renal tubules at 30/15 mg/kg/day in the 3 month dog study; At the no-effect doses for kidney changes (mice 5 mg/kg, dogs 3 mg/kg), entecavir exposures were >40 times higher than in humans. Chronic progressive nephropathy was the leading cause of death in male rats (including controls) in a carcinogenicity study. No kidney lesions were observed in rats and monkeys at the highest doses tested in 6-month and 1-year studies, respectively.

Thymus atrophy was reported in rats at high doses of 200 mg/kg/day in a 2 week study and in a 3 month dog study at 30/15 mg/kg/day.

Skeletal muscle myopathy was induced at doses of 1 mg/kg/day in mice in a 6 month study and in rats at 0.6 mg/kg/day in a 6 month study. Exposures to entecavir at no-effect doses for these findings were ≥ 4 times those in humans. Skeletal muscle was not a target tissue in dogs or monkeys.

Brain/spinal cord inflammation was apparent only in the 3 month study in dogs for which both no-effect and threshold doses were established; at such doses, exposures to entecavir were 13- and 50-fold higher than in humans at 1 mg. The CNS inflammation was shown to be reversible and was not observed in mice, rats, or monkeys.

Gastrointestinal toxicity, including fibrin microthrombosis, necrosis of villi, inflammation, and haemorrhage was occasionally recorded in rats (at 200 mg/kg/day in a 2 week study) and dogs (at 100/50 mg/kg/day in the 2 week study); doses with overt toxicivity. Gastrointestinal toxicity was particularly notable in moribund animals at high doses. GI-function was not affected in mice after single doses in safety pharmacology studies.

In mice, alveolar histiocytosis was observed in the lungs at ≥ 1 mg/kg, and bronchioloalveolar hyperplasia and benign lung adenomas were observed at ≥ 10 mg/kg. At the no-effect dose for lung changes in mice (0.2 mg/kg), exposure to ETV was 22 times that in humans at 0.5 mg (12 times at 1 mg). Similar changes were not observed in repeat-dose studies in rats, dogs, or monkeys at exposures to ETV that exceeded those in mice associated with lung changes.

Monkey was the species that tolerated entecavir best and in a 12 month monkey study the high dose of 40 mg/kg/day produced no toxicity and clinical chemistry showed only slightly increased urea nitrogen and potassium, corresponding to high systemic exposure levels (AUC) in relation to expected clinical levels.

©EMEA 2006

The range of target organs for toxicity thus differed somewhat depending on species. In some cases this might depend on differences in distribution e.g. levels of compound in the CSF in dogs where CNS inflammation was recorded, reached up to 340 ng/ml but only 17.8 ng/ml in monkey where no CNS effects were apparent.

- Genotoxicity *in vitro* and *in vivo* (with toxicokinetics)

ETV was not mutagenic in the Ames assay at concentrations ≤ 5000 μg/plate or in Chinese Hamster Ovary (CHO) cells at concentrations ≤ 1000 μg/ml. Entecavir was clastogenic in an in vitro chromosomal aberration test in primary human lymphocytes at concentrations ≥ 10 μg/ml (≥ 36 μM). Entecavir was nonetheless not clastogenic in an oral micronucleus study in rats at doses up to 2000 mg/kg daily for 3 days and ETV did not cause DNA damage in the in vivo-in vitro hepatocyte DNA repair study in rats at single doses up to 2000 mg/kg.

It is known that dNTP pool imbalances can be caused by nucleosides and can result in mutagenicity in the mouse lymphoma assay (MLA). A study was therefore conducted to characterise if imbalances in intracellular dNTP pools occurred with entecavir and results were consistent with such a mechanism being involved in the positive clastogenic results in human lymphocytes. The levels of intracellular ETV-TP associated with a positive response in the MLA were higher than levels expected in the clinical situation, however, quantitative extrapolations from *in vitro* studies are questionable. Phosphorylation of entecavir in human hepatoma cells and human fibroblasts was comparable and appeared much higher, up to 20-30x, than in mouse lymphoma cells. These differences were likely a function of differences in experimental conditions employed. A cumulative mutational effect of prolonged nucleotide imbalances has been reported in some published studies, and it is possible that high sustained levels of the triphosphate in the carcinogenicity studies could perturb intracellular nucleotide pools and contribute to the tumours recorded. Studies *in vitro* and *in vivo* with entecavir indicated that such a mechanism could be significant in tissues/organs such as liver and brain. Additional *in vitro* studies showed that ETV-TP levels were higher in mouse pneumocytes than in rat cells. A threshold concentration of entecavir appeared to exist and e.g. in liver, levels of entecavir-TP had to increase to 300 nM and above before disturbances in dNTP pools became manifest.

- Carcinogenicity (with toxicokinetics)

Mouse and rat long term carcinogenicity studies were conducted, in which entecavir was administered by oral gavage at doses up to 4 mg/kg/day in mice and 1.4/2.6 mg/kg/day in male/female rats.

In male mice, lung bronchioalveolar adenomas and carcinomas were increased from a dose of 0.004 (corresponding to exposure levels <1 in comparison with expected human exposures) and statistically significantly increased from a dose of 0.04 mg/kg (exposure multiples of 2-4x the human exposure). In female mice, bronchoalveolar tumours were increased at 0.4 mg/kg/day and statistically significantly increased at 4 mg/kg/day. Other findings included an increase in vascular tumours and benign salivary gland tumours in high dose females and an increase in male liver tumours at the high dose. Separate studies showed that entecavir increased proliferation of type II pneumocytes in mice, and multifocal hyperplasia was reported after 2 weeks of treatment. In CCR2 knockout mice the development of lung lesions was delayed. In addition increases in recruitment of macrophages in lung appeared to correlate with the proliferative response to entecavir. Rate of development of lung lesions also seemed dependent on strain of mouse.In a series of additional studies *in vitro* and *in vivo*, it was determined that a direct proliferative effect of entecavir on type II pneumocytes was not involved. Rather a "key event" in the development of mouse lung tumours appeared to involve an entecavir mediated chemotactic signal that resulted in an increased recruitment/accumulation of macrophages in the lung and that exhibited species differences in magnitude and appeared absent using human cells *in vitro*. The potential role of entecavir in subsequent events is unclear. Coculture of macrophages with pneumocytes has been show to stimulate cell division in several species. Overall, it appears that the persistent and sustained proliferation of Type II pneumocytes in mouse lung was causally linked to the increased incidence of lung tumours in the mouse carcinogenicity study. The data also suggest a species-selective key event (ie, chemotaxis) in the development of entecavir-induced lung tumours in mice.However, as also discussed above (under genotoxicity) a general tumourigenic mechanism may involve a cumulative mutational effect of prolonged nucleotide pool imbalances.

Survival was markedly reduced in control and drug-treated males in the rat carcinogenicity study, apparently due to a high incidence of chronic progressive nephropathy. The incidence of rare brain tumours, glioma, was increased in males and females, notably at the high dose; a slight increase in the incidence of this tumour relative to controls was also observed in males at 0.2 mg/kg. Tumours were determined to be of microglial origin and *in vitro* studies in rat microglial cells showed that entecavir caused decreases in dGTP and increases in dTTP and dCTP with apparent threshold effects, consistent with the fact that deoxynucleotide triphosphate (dNTP) pool perturbations could be involved in tumour development. Benign tumours of the pancreas were noted in males from 0.2 mg/kg and pancreatic acinar cell hyperplasia with no clear dose-response was increased at the two higher dose levels. In addition, some other tumours were found scattered in various groups, including benign skin fibroma in females at 0.4 and 2.6 mg/kg, and liver adenoma and carcinoma, Zymbal's gland carcinoma and uterine hemangiosarcoma in females at 2.6 mg/kg. The tumourigenic dose was considered as 1.4 mg/kg in males and 2.6 mg/kg in females. Although not statistically significant, particularly when considering specific tumour types, an increase in tumour incidence that could be biologically relevant, was evident at 0.2/0.4 mg/kg corresponding to systemic exposure levels approximately 3x and 6-7 x the clinical exposure at the 1 and 0.5 mg doses, respectively.

Taken together the mouse and rat carcinogenicity studies indicate that a tumourigenic response that seems biologically relevant may occur at systemic exposure levels <1x in male mouse (based on lung tumours), 8x in female mouse (based on lung tumours), and at exposures 3x in male and female rats in relation to a clinical dose of 1 mg. Margins of exposure in relation to the 0.5 mg dose would be approximately twice as high. Overall entecavir- treatment induced tumours at multiple sites in both genders in mouse and rat. With regard to non-lung tumours, experimental data suggest that entecavir-TP levels above a certain threshold level may cause generalised dNTP pool perturbations that may contribute to tumour development in susceptible tissues/organs. Any potential risks to humans are further addressed in the safety and risk management plan sections of this document.

- Reproductive and developmental studies

The potential for reproduction toxicity of entecavir was investigated in standard studies in rats and rabbits. Although the pharmacokinetic profile of entecavir was not specifically determined in rabbits, in one study exposure was determined indicating a pharmacokinetic profile similar to those in rats and dogs, and therefore additional pharmacokinetics studies in rabbits were considered unnecessary.

In the female fertility study doses of up to 30 mg/kg/day had no effects on oestrous cycling, mating or early embryonic development of offspring. In the male fertility study doses of up to 10 mg/kg/day had no effects on mating indices, fertility indices, or early embryonic development. No effects were reported on testes, epididymides, prostate/seminal vesicle weights or sperm motility, morphology and counts.

In the rat embryo foetal study, entecavir had no effects at the low dose of 2 mg/kg/day. In the mid and high dose dams at 20 and 200 mg/kg/day, decreased body weight gains and food intake were apparent. Embryofoetal deaths increased at the high doses, and foetal body weight was decreased at 200 mg/kg/day. Also at 200 mg/kg/day, malformations included tail (constriction band, bent, short, stubbed or absent) and vertebrae (bifurcated arches). Further, ossification delays (vertebrae, sternebrae, phalanges), increases in the number of lumbar vertebrae and ribs were reported. Exposure at 2 mg/kg, a dose not associated with any compound related changes, was approximately 28x the expected human exposure.

In a preliminary study in rabbits, maternal toxicity was apparent at ≥12.5 mg/kg and embryo-toxicity at ≥25 mg/kg. In an embryo foetal study at 16 mg/kg, corresponding to very high exposure levels, entecavir increased resorptions and decreased live litter sizes. Development delays in the ossification of the hyoid and an increased incidence of 13th rib were also noted at this dose. No foetal effects were recorded at the two lower doses (1 and 4 mg/kg).

In the pre- and postnatal study in rats at doses of 0.3, 3 and 30 mg/kg from day 6 of gestation through day 20 of lactation, entecavir induced maternal changes consisting of a slight, transient reduction of weight gain in the high dose group during gestation. Evaluation of the F1 generation for sexual maturity, sensory perception, learning, memory and reproductive function did not indicate any entecavir-induced changes.

- Local tolerance (if applicable)

As entecavir is administered orally, no studies evaluating local tolerance were considered necessary.

- Other toxicity studies

A 1-month immunotoxicty study in rats with doses up to 10 mg/kg showed that entecavir did not affect humoral T cell dependent antibody response.

Entecavir did not exhibit any phototoxic potential in in vitro studies.

Because of the potential for mitochondrial toxicity with nucleoside analogues, an in vitro study was conducted to assess the effects of ETV at concentrations up to 750 µM (approximately equivalent to 208 µg/ml) and 3 marketed antiviral nucleoside analogues on cell viability and cellular respiration in human HepG2 hepatoma cells. Exposure of HepG2 cells to ETV for 5 days did not result in any adverse effects on oxidative mitochondrial respiration. These results are consistent with experiments showing no inhibition for mitochondrial DNA polymerase gamma (K_i of ETV-TP is >160 µM), and that high levels of ETV-TP were not recognised or utilised by DNA polymerase gamma. Additional in vitro studies in HepG2 cells with entecavir at concentrations up to 3.4 µM (approximately equivalent to 0.9 µg/ml) for up to 15 days had no significant effects on extracellular lactate production, mitochondrial DNA or cell numbers.

Ecotoxicity/environmental risk assessment

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

4. Clinical aspects

Introduction

The clinical programme consisted of:

- studies aiming to characterise the pharmacokinetics profile of entecavir (including interaction studies, studies in special populations and population PK analysis of data from Phase 2 studies).
- three distinct Phase 2/3 'programmes' to assess the use of ETV in the treatment of chronic HBV infection. These have been conducted in a staggered but overlapping sequence and all are ongoing. The worldwide programme started in early 1999 and consists of 12 studies in over 2100 subjects. A separate development programme in China began in early 2002 and consists of three studies in 875 subjects. A third development program in Japan began in late 2002 and consists of three studies plus one roll-over study in over 225 subjects. All three programs have rollover studies that accommodate long-term dosing in appropriate subjects who were previously enrolled in a Phase 2/3 study.

The clinical programme therefore included a wide range of patients: nucleoside-naïve subjects, and lamivudine-refractory subjects, including HBV/HIV co-infected from different geographic regions.. LVD-refractory post-liver transplant patients provided limited safety data in an important setting of chronic HBV patients. An open-label head-to-head comparison of ETV 1.0 mg versus adefovir (ADV) in patients with decompensated cirrhosis is ongoing and provided some preliminary safety and efficacy data.

The clinical studies included a statement regarding conduct in accordance with Good Clinical Practices.

The applicant has provided a statement to the effect that clinical trials conducted outside the community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

The approved indication is "Baraclude is indicated for the treatment of chronic hepatitis B virus (HBV) infection in adults with compensated liver disease and evidence of active viral replication, persistently elevated serum alanine aminotransferase (ALT) levels and histological evidence of active inflammation and/or fibrosis. This indication is based on clinical trial data in patients with HBeAg positive and HBeAg negative HBV infection, nucleoside naive patients and patients with lamivudine-refractory hepatitis B".

Pharmacokinetics

The pharmacokinetics profile of entecavir was determined in a series of studies including 644 healthy subjects. In addition population pharmacokinetic and population pharmacokinetic/pharmacodynamic (PK/PD) analyses were performed from data collected in a phase I study from subjects with selected degrees of renal impairment and in three phase II studies from patients with chronic HBV infected (n = 222). Another phase II study assessed the pharmacokinetics of entecavir when co-administered with either cyclosporine or tacrolimus in liver transplant recipients.

Entecavir plasma and urine concentrations were determined by validated adequate analytical methods.

- Absorption

Entecavir is rapidly absorbed from the gastrointestinal tract following oral administration, with peak plasma concentrations (C_{max}) occurring within 1 hour of administration. Due to the lack of an adequate intravenous formulation, the oral absolute bioavailability has not been determined. The bioavailability has been estimated to be at least 70%. Limited data suggested that entecavir is not a substrate of P-gp.

Bioequivalence has been demonstrated between the tablets and capsules used during clinical development and between the oral solution and the clinical tablets. The shape of the commercial film coated tablet differs from that of the clinical tablet (round instead of triangle) but no further bioequivalence study was considered necessary.

After administration of food (high-fat breakfast) C_{max} is reduced by about 50% and AUC by 20%, while t_{max} is increased compared to fasted state. A PK/PD modelling showed that in treatment naïve subjects entecavir could be taken with or without food, as exposure in both cases are at the flat part of the concentration-response curve and well above EC_{90}/AUC_{90}. In LVD-refractory patients, however, a reduced exposure when taken with food could result in a lower efficacy. Therefore, the Summary of Product Characteristics recommends that for treatment naïve patients, entecavir can be taken with our without food whereas in lamivudine refractory patients, entecavir is to be taken without food at least 2 hours before or after a meal.

- Distribution

As entecavir has not been administered intravenously to humans, V_{ss} has not been determined. The total volume of distribution has nevertheless been estimated to be large based on V_{ss} data in animals and Vz/F in human (2550 to 7708 l). The protein binding of entecavir in human serum is low (~13%), and entecavir distributes uniformly between plasma and red blood cells (RBCs) in human whole blood.

- Elimination

Renal excretion of unchanged substance is the primary route of entecavir elimination. Values for renal clearance (about 400 ml/min) of entecavir were greater than the glomerular filtration rate, indicating

that excretion of entecavir by the kidneys occurs via a combination of glomerular filtration and net tubular secretion.

After 1 mg administration of ^{14}C-entecavir, about 40 % of the dose was recovered in urine over the first 24h, thereafter the excretion rate was low reflecting the long terminal half-life of entecavir. Over the 14 days collected period, 75.6±4.6% of the dose was excreted in urine and 6.3±1.0% of the dose in faeces. Total recovery was on average 81.9% (range 76.6-86.9%). Unchanged entecavir constituted the majority of the radioactivity in urine, 70.4±3.9% of the dose. The extent of metabolism was low, the metabolites in urine and faeces constituted ≤10% of the administered dose. No phase I metabolites were detected. In plasma and urine three glucuronide conjugates were detected and, in faeces a sulphate conjugate. Entecavir has a terminal half-life of approximately 130 hours and an effective half life for accumulation of approximately 24 hours.

- Dose proportionality and time dependencies

Single and multiple dose data suggest more than proportional increase in exposure after single dose administration, but roughly dose proportional exposure at steady-state. Steady state is generally reached within 7 – 10 days. The mean accumulation index was approximately 1.5 – 1.8, indicating that once daily dosing resulted in modest degree of accumulation. A model with concentration dependent distribution to a peripheral compartment could adequately describe the data. Entecavir pharmacokinetics are time independent.

The pharmacokinetics of entecavir is characterised by a moderate inter-individual variability. In the population PK analysis of Phase II data, inter-individual variability in CL/F was 40% and the residual variability 36%. Intra-individual variability (inter-occasion variability) was not determined in this analysis. In healthy volunteers, variability was lower with CV for AUC about 15-20%.

- Special populations

Renal impairment
The effect of renal impairment on the pharmacokinetics of entecavir was assessed in a parallel group, single dose (1 mg) study. Subjects were assigned to each of 6 groups based on underlying renal function: normal renal function (CLcr > 80 ml/min), mild renal impairment (CLcr: 50 – 80 ml/min), moderate renal impairment (CLcr: 30 – 49 ml/min), severe renal impairment (CLcr < 30 ml/min), severe renal impairment with hemodialysis and severe renal impairment managed with continuous ambulatory peritoneal dialysis (CAPD). Results are shown in table 1.

Table 1: Mean PK parameters of entecavir after a single oral dose of 1 mg with normal renal function and with various degrees of renal impairment.

Pharmacokinetic Parameter	Normal >80 (n=6)	Mild 50-80 (n=6)	Moderate 49-30 (n=6)	Severe 20-<30 (n=6)	Haemodialysis 2h pre (n=6)	Post (n=6)	CAPD (n=4)
Cmax (ng/ml)							
Geometric Mean	8.06	10.43	10.53	15.30	12.12	15.37	16.56
(CV%)	(30.7)	(37.2)	(22.7)	(33.8)	(41.1)	(56.4)	(29.7)
AUCo-t (ng.h/ml)							
Geometric Mean	27.90	51.46	69.49	145.66	127.10	233.91	221.80
(CV%)	(25.6)	(22.8)	(22.7)	(31.5)	(20.2)	(28.4)	(11.6)
$t_{1/2}$ (h)							
Mean	77.39	113.21	130.65	162.34	155.55	276.34	802.16
(SD)	(29.88)	(13.67)	(25.74)	(33.49)	(33.16)	(143.40)	(872.37)
CLT/F (ml/min)							
Mean	588.1	309.2	226.3	100.6	110.69	50.6	35.7
(SD)	(153.7)	(62.6)	(60.1)	(29.1)	(24.6)	(16.5)	(19.6)
CLR (ml/min)							
Mean	383.18	197.90	135.57[a]	40.27	NA	NA	NA
(SD)	(101.80)	(78.11)	(31.55)	(10.11)			

Results showed that exposures i.e AUC values are increased approximately by 90 %, 150 % and 420 % in the creatinine clearance groups 50 – 80, 49 – 30 and < 30 ml/min as compared to subjects with creatinine clearance > 80 ml/min.

Given the large renal function-related increase in exposure in moderate, severe and end-stage renal disease, dose reductions are needed in these groups. A population PK approach using data from this study and simulations were developed for defining dosing recommendations in this population, using target AUC criteria which were based on the exposure of patients with normal renal function.
The results of the simulation showed that no dosage adjustment is necessary for subjects with mild renal impairment (CLcr: 50-80 ml/min). In order to manage exposure in subjects with moderate renal impairment (Clcr: 30-50 ml/min), a dose reduction of 50 % of the normal dose is recommended. For subjects with severe renal impairment (CLcr ‹ 30 ml/min), 30 % of the recommended normal dose is needed to maintain comparable exposure to subjects with normal renal function. In subjects with end stage renal disease managed with hemodialysis or CAPD, 10 % of the normal dose (administered after the hemodialysis session) is recommended.

Hepatic impairment
In a study comparing the pharmacokinetics of 1 mg oral dose of entecavir in normal healthy volunteers and subjects with moderate (Child Pugh class B) and severe hepatic cirrhosis (Child Pugh class C), results showed that exposure in terms of Cmax and AUC was not altered in subjects with moderate to severe hepatic impairment. No dose adjustment is therefore warranted.

Gender, weight, race
The pharmacokinetics population analysis showed that entecavir pharmacokinetics are not influenced to a significant extent by gender, weight or race. No dose adjustment or specific precautions are therefore needed in these populations in the absence of reduced renal function.
The effect of gender was also evaluated in a pharmacokinetics study in the elderly. AUC was 14 % higher in women than in men, but this was due to differences in renal function and weight.

Paediatric population
There are currently no pharmacokinetic data in children or adolescents.

Elderly
The effect of age on the pharmacokinetics of entecavir was evaluated comparing elderly subjects in the age range 65-83 years (mean age females 69 years, males 74 years) with young subjects were in the age range 20-40 years (mean age females 29 years, males 25 years). AUC was 29 % higher in elderly than in young subjects however this was mainly caused by a difference in renal function and weight. After adjusting for differences in creatinine clearance and body weight elderly had a 12.5% higher AUC than young subjects.

Pharmacokinetics in target population
A population PK/pharmacodynamic analysis was performed on data collected in phase 2 studies from subjects with chronic hepatitis B (AI463004, AI463005, AI463014). The analysis showed that chronic hepatitis B infection per se is not expected to have a major effect on the pharmacokinetics of entecavir. Renal function was the only significant covariate influencing entecavir clearance. The population estimated CL was similar to that in healthy volunteers, while *post-hoc* estimates of exposure at the 0.5 and 1 mg dose levels suggested a 30% and 70% higher exposure at 0.5 and 1 mg, respectively, in patients than in healthy volunteers. The difference in exposure between healthy volunteers and patients to a large extent can be explained by differences in variability, a skewed distribution in patients and a lower renal function. Inter-individual variability in CL/F was 40% in the population PK analysis.

Patients with liver transplant
Compared to healthy volunteers, orthotopic liver transplant subjects on a stable dose of immunosuppressive therapy (cyclosporin or tacrolimus) appeared to have 2 fold increase in exposures, which seemed mainly be caused by differences in renal function. Therefore dosage adjustment should be considered based on renal function in liver transplant patients.

- Pharmacokinetic interaction studies

Entecavir does not affect CYP450 isoenzymes and is itself not a substrate for CYP450. Therefore CYP450 mediated interactions are unlikely to occur with entecavir.

Single and multiple dose interaction studies were performed with lamivudine and multiple dose interaction studies were performed with adefovir and tenofovir. These compounds are eliminated predominantly by tubular active secretion. No significant interaction was observed with lamivudine, adefovir or tenofovir. The reason for lack of interaction could possibly be that these compounds are not substrates of the same transport protein as entecavir. It cannot be excluded that there may be interactions with other compounds undergoing active renal secretion, therefore as recommended in the Summary of Product Characteristics, patients should be monitored closely for adverse reactions when entecavir is co-administered with such compounds.

Data from the study in liver transplant patients suggest that cyclosporin and tacrolimus do not affect entecavir pharmacokinetics to any major extent.

Pharmacodynamics

- Mechanism of action

As already mentioned in section 3.3, entecavir is a cyclopentyl guanine analogue and a selective inhibitor of HBV replication.

- **Entecavir resistance profile**

In vitro
In cell-based assays 8-fold reductions in entecavir phenotypic susceptibility was observed for LVD-resistant strains. Further reductions (>70 fold) in entecavir phenotypic susceptibility required the presence of primary LVD resistance amino acid substitutions (rtL180M and/or rtM204V/I) along with additional substitutions at residues rtT184, rtS202 and rtM250, or a combination of these substitutions with or without an rtI169 substitution in the HBV polymerase. When present alone in recombinant virus, none of these substitutions exhibited significant phenotypic resistance to ETV.
There is no in vitro evidence for cross-resistance between adefovir and ETV, or for any functional interference between ETV and other nucleoside/nucleotide analogues used for the treatment of either HBV or HIV.

©EMEA 2006

In vivo

The ETV resistance profile is based on the evaluation of genotypic and phenotypic data from patients treated with ETV for up to 96 weeks in 7 clinical studies conducted in nucleoside-naïve and LVD refractory patients. It has been confirmed that the identified signature ETV associated resistance (ETVr) substitutions rtT184, rtS202 and/or M250 with or without an rtI169 substitution in the HBV polymerase require the presence of primary lamivudine resistance amino acid substitutions (rtL180 and/or rtM204) to result in reduction in ETV phenotypic susceptibility.

Nucleoside-naïve patients: The majority of HBV chronically infected nucleoside-naïve patients (543/659; 82%) receiving ETV 0.5 mg once daily achieved a reduction in viral load to <400 copies/ml at 48 weeks. Genotypic analysis of serum HBV DNA from nucleoside-naïve HBeAg- positive (Study AI463022; n=219) or HBeAg-negative (Study AI463027; n=211) patients detected no genotypic changes in the HBV polymerase associated with phenotypic resistance to ETV at Week 48. There were 255 nucleoside-naïve patients that completed $\geq$90 weeks of ETV therapy and another 358 patients, who were halted prior to Week 96 due to a complete virological response (Study AI463022; n=290, Study AI463027; n=268). Analysis of the patients continuing ETV treatment during the second year did not reveal any evidence of emerging resistance. Genotypic and phenotypic analysis of HBV isolates from 18 patients who experienced a confirmed virological rebound using PCR assay ($\geq$1 $\log_{10}$ copies/ml increase from nadir) by Week 96 failed to show the emergence of ETV resistance. In four additional patients, who did not achieve HBV DNA reduction <100.000 copies/ml, HBV isolates remained fully susceptible to ETV.

Lamivudine-refractory patients: One-fifth of LVD-refractory patients (21%) with chronic HBV infection achieved HBV DNA levels <400 copies/ml at Week 48 on ETV 1.0 mg once daily. Genotypic analysis of clinical samples with detectable viral DNA identified 7% (13/189) with evidence of emerging ETV resistance-associated substitutions by week 48. Of these 13 patients, 2 (1%) experienced virological rebound at Week 48. However, during the second year of treatment there were additionally 14 of 154 (9%) patients displaying ETV resistance. In all cases resistant variants had evidence of pre-existing LVD-resistance mutations (rtL180M and/or rtM204V/I) and an additional change at residues rtT184, rtS202 and/or rtM250. No additional primary ETV resistance associated substitutions were identified during the 2 years of ETV treatment.

Subsequent analysis of baseline viral samples revealed the presence of ETV resistance associated substitutions prior to ETV therapy in the majority of patients evaluated, demonstrating that prior LVD treatment selects for ETV resistance changes in a subset of patients.

Liver transplant patients: Nearly all subjects (7/8) developed ETV genotypic resistance mutations through 3 years of treatment.

Overall results suggest:
- that ETV treatment does not select for LVD resistance substitutions *de novo*
- that phenotypic resistance to ETV require the presence of LVD resistance mutations (rtL180M and/or rtM204V/I) as well as secondary ETV resistance substitutions at rtT184, rtS202 and/or rtM250
- that prior LVD treatment selects for ETV resistance, as ETV resistance associated substitutions were found to pre-exist in some LVD-refractory subjects
- that in LVD-refractory patients, 9% of ETV-treated patients experienced virological rebounds due to resistance in the second year of therapy
- that in nucleoside-naïve patients, there was no evidence of the emergence of ETV resistance up to Week 96 of ETV therapy
- that there was a correlation between the baseline virus population phenotype (EC_{50} of <10 nM) and the maximal ETV antiviral efficacy in treated subjects (HBV DNA levels of <300 copies/ml).
- in vitro data, as well as limited clinical data, demonstrated that ETV resistant HBV appear to retain full susceptibility to adefovir.

- Relationship between plasma concentration and effects

The dose relationship in both populations (effect of dose on the time course of HBV DNA reduction) and a decreased effect of ETV in the LVD refractory population was shown. A lamivudine naïve subject receiving a 0.5 mg daily dose of entecavir was estimated to have an expected maximum reduction of HBV DNA of 5.5 log copies/ml, whereas a lamivudine refractory subject receiving a 1 mg daily dose of entecavir would have an expected maximum reduction of HBV DNA of 4.7 log copies/ml. It was also shown that there was no evidence of severe CNS adverse events being associated with increased ETV exposure. Two PK/PD analyses have been made; a PK/PD analysis evaluating the relationship between AUC, C_{max}, C_{min} and reduction in viral load and a PK/PD analysis evaluating the effect of exposure (AUC) on the time course of HBV DNA reduction. Both analyses suggest that the 0.5 mg dose will provide maximum efficacy in treatment naïve patients, and that no additional effect could be expected by increasing the dose. The data from these models suggest that the exposure at the 1.0 mg dose is approaching the top of the concentration response curve and that even with a large increase in dose, the increase in efficacy would be very modest. Hence, from a PK/PD perspective it is concluded that the proposed 1.0 mg dose is acceptable in LVDr patients.

Clinical efficacy

The clinical programme comprised 11 clinical studies, 9 from the worldwide programme and 2 from the China development programme. The main clinical studies include 3 phase III studies and 1 subset of patients in a dose-ranging phase II study and compare the efficacy of entecavir (ETV) versus lamivudine (LVD). Studies cover a broad selection of patients with compensated liver disease and HBV viraemia (See table 2):
- nucleoside-naïve HBeAg positive subjects (study A1463-022; n=709),
- nucleoside-naïve HBeAg negative subjects (study A1463-027; n=638)
- and LVD-refractory HBeAg positive subjects (studies A1463-026; n=286 and A1463-014; n=42).

The total efficacy population in these studies includes 1720 treated patients (ETV 862/LVD 858). Efficacy data were also available from several supportive studies. In addition, preliminary data in HBV/HIV co-infected LVD-refractory patients from an on-going phase II study (AI463-038), from an open-label study (AI463-015) conducted in LVD-refractory post-liver transplant patients and from an ongoing open-label head-to-head comparison of ETV 1.0 mg versus adefovir (ADV) 10 mg in patients with decompensated cirrhosis (AI463-048) have been provided. Finally preliminary results from roll-over studies have been submitted.

Although the clinical programme has been initiated prior to the CHMP guideline on anti-hepatitis B therapy, it is in line with the spirit of the guideline.

Table 2: Overview of the pivotal phase III/II studies in the entecavir clinical programme

Study	No. centres Locations	Study population	Treatment	Duration	Number enrolled	Number treated ETV	LVD
Nucleoside-naïve subjects							
Phase II							
AI463-004 Phase II Randomised, double-blind placebo-controlled	9 Argentina EU USA, Canada, Hong Kong	HBeAg – positive and negative (HBV DNA ≥20 MEq/ml) **LVD-naïve and -refractory**	4 cohorts; 0.1, 0.5, 1,0, 0.05 mg QD	28 days+ 24 week follow-up	42	ETV n=34 (8-9 /dose level)	Placebo n= 8 (2/dose level)
AI463-005 Phase II Randomised, double-blind ETV vs LDV 100 mg	39 EU, USA, Australia Asia	HBeAg – positive and negative (HBV DNA ≥40 MEq/ml) **LVD- naive**	0.01, 0.1, 0.5 mg QD	24 weeks +12 weeks follow-up (open LVD to week 48)	185	ETV n=129 0.01mg n=52 0.1 mg n=34 0.5 mg n=43	LVD 100mg n = 40
Phase III studies							
AI463-022	137 EU, USA, Asia, South America	HBeAg +	ETV 0.5 mg QD LVD 100 mg QD	52 weeks (Up to 96 weeks for partial responders)	1056	354	355
AI463-027	146 EU, USA, Asia, South America	HBeAg -/anti-HBe+	ETV 0.5 mg QD LVD 100 mg QD	52 weeks (Up to 96 weeks for partial responders)	1468	325	313
					Total no	679	668
LVD-refractory subjects							
AI463-026	84 EU, USA, Asia, South America	HBeAg +	ETV 1.0 mg QD LVD 100 mg QD	52 weeks (Up to 96 weeks for partial responders)	420	141	145
AI463-014 (Phase II)	41 EU, USA, Asia; Australia	HBeAg pos or neg with viraemia on LVD (HBV DNA ≥10 MEq/ml)	ETV 0.1, 0.5, 1.0 1.0 mg QD LVD 100 mg QD	52 weeks (Up to 76 weeks for partial responders) 24 week follow-up (Complete Response) 12 week others (Open label phase at w 48; partial respond.)	259	ETV, n=136 0.1 mg n=47 0.5 mg n=47 1.0 mg, n=42	45
					Total no	183	190
					Total all populations	862	858

ETV: entecavir; LVD lamivudine; QD once daily; Meq/ml

- Dose response study(ies)

Three phase 2 studies (004, 005 and 014) were conducted to confirm the *in vitro* findings of anti-HBV activity and to identify the recommended dose for the confirmatory trials (See table 2).

The three dose-ranging studies included a total of 223 subjects treated with different ETV doses (range: 0.01, 0.1, 0.5 mg and 1.0 mg once daily) and 87 subjects treated with LVD once daily. Nucleoside-naïve subjects and LVD-refractory subjects were evaluated separately. The rational for testing a higher dose in LVD-refractory subjects laid on the in vitro data showing a reduced susceptibility of LVDr virus to entecavir.

©EMEA 2006

Nucleoside naïve subjects
In the pilot dose escalating study 004, entecavir produced rapid and significant reductions in serum HBV DNA. The mean log reduction in HBV DNA (Chiron assay) in the ETV group was –2.20 (0.005 mg), -2.27 (0.1 mg), -2.80 (0.5 mg) and – 2.54 (1.0 mg) compared to placebo (p<0.0001). There was no evidence of dose related or dose limiting clinical adverse events. A strong dose response was demonstrated in the post-dosing measurements (i.e 28 days post completion of treatment). The HBV DNA differences for 0.5 and 1.0 mg versus that for the 0.05 mg were - 1.36 $\log_{10}$ (p = 0.0004) and - 1.51 $\log_{10}$ (p= 0.0051) respectively.

In study 005, both ETV 0.1 and 0.5 mg doses were superior to LVD in viral load reduction. The difference in HBV DNA levels by PCR for ETV 0.1 mg versus LVD at Week 22 was estimated to be - 0.97 log, and the difference for ETV 0.5 mg versus LVD was -1.28 log (p-value $\leq$ 0.0001 for both comparisons). There was a clear dose response. The mean $\log_{10}$ reduction in HBV DNA in the ETV dose groups was -2.41 $\log_{10}$copies/ml (0.01 mg), -4.31 $\log_{10}$ (0.1 mg), -4.72 $\log_{10}$ (0.5 mg) (p <0.0001 for both the comparisons of 0.01 mg to 0.1 and 0.5 mg). The mean $\log_{10}$ reduction for LVD, -3.36 $\log_{10}$ copies/ml was significantly greater than in the ETV 0.01 mg group.
There was a significant difference favouring the ETV 0.5 mg dose versus the ETV 0.1 mg dose in reduction in HBV DNA by PCR (p=0.018 at Week 22).

Overall, in nucleoside-naïve subjects, a clear dose response was demonstrated and a threshold dose of 0.01 mg was identified. The selected dose of ETV of 0.5 mg for nucleoside-naïve subjects was based on superiority in HBV DNA reduction by PCR at Week 22 compared with the ETV 0.1 mg dose. The analyses of dose-response suggested a flattening of the curve in the 0.1-0.5 mg range. Consequently, the 0.5 mg was chosen for further phase III development. Due to adverse events (mainly nervous system) which were not confirmed in phase III trials particularly seen with 0.5 mg, higher dose in this population has not been investigated.

LVD-refractory subjects
The study 014 compared three doses of entecavir (0.1 mg, 0.5 mg and 1.0 mg) versus lamivudine.

The majority of subjects were male (81%) and white (61%) or Asian/Pacific Islander (32%) with a mean age of 46 years; 67% of subjects were positive for HBeAg at baseline. Eighty-seven percent of subjects had YMDD mutations in the HBV DNA polymerase gene.

Results on the primary endpoint (proportion of subjects with HBV DNA levels below limit of quantification of 0.7 Meq/ml) are presented in table 3. The results showed that ETV 1.0 and 0.5 mg were superior to lamivudine in the primary endpoint, as well as in all the secondary endpoints (table 4).

Table 3: HBV DNA < LOQ (0.7 MEq/ml) by bDNA Assay at Week 24 (NC = F)

	ETV 0.1 mg	ETV 0.5 mg	ETV 1.0 mg	LVD 100 mg
	n = 47	n = 47	n = 42	n = 45
NC = F	9/47 (19 %)	24/47 (51 %)	33/42 (79 %)	6/45 (13 %)
difference ETV - LVD	5.8 %	37.7 %	65.2 %	
95% CI	-12.6 % ; 24.2 %	14.4 % ; 61.1 %	39.8 % ; 90.7 %	
p-value	0.45	<0.0001	< 0.001	

Consistent results were reached at weeks 48.

©EMEA 2006

Table 4: Secondary efficacy endpoints (NC=F)

Secondary endpoint	ETV 0.1 mg N=47	ETV 0.5 mg N=47	ETV 1.0 mg N=42	LVD 100mg N=45
Mean log$_{10}$ reduction bDNA assay log$_{10}$ MEq/ml				
Week 24	-1.23	-2.16	-2.43	-0.43
Week48	-1.33	-2.16	--2.52	-0.23
Mean log$_{10}$ reduction PCR assay log$_{10}$ copies/ml				
Week 24	-1.89	-3.75	-4.21	-0.95
Week 48	-2.85	-4.46	-5.06	-1.37
Week 76 (n=27)		-5.66	-5.01	-1.84
HBV DNA<400 copies/ml at week 24 n (%)	0	4 (9)	7 (17)	1/45 (2)
HBV DNA<400 copies/ml at week 48 n (%)	2 (4)	12 (26)	11 (26)	2 (4)
HBeAg loss n (%) at week 24 n (%)	0	3/32 (9)	3/27 (11)	2/32 (6)
HBeAg loss n (%) at week 48 n (%)	0	3/32 (9)	3/27 (11)	3/32 (9)
HBeAg seroconversion at week 48 n (%)	0	1/32 (3)	1/27 (4)	2/32 (6)
ALT normalization Week 24 n (%)	12/30 (40)	17/29 (59)	11/28 (39)	7/33 (21)
ALT normalization Week 48 n (%)	14/30 (47)	17/29 (59)	19/28 (68)	2/33 (6)
Complete response*				
Week 24 all	4/47 (9)	11/47 (23)	8/42 (19)	3/45 (7)
Week 48 all	6/47 (13)	9/47 (19)	12/42 (29)	2/45 (4)
Week 48 HBeAg negative (NC=M)	6/14 (43)	6/23 (46)	10/14 (71)	0
Week 48 HBeAg positive (NC=M)	0	3/27 (11)	2/34 (8)	2/18 (11)

*HBV DNA < LOQ (0.7 MEq/ml) by bDNA assay, negative HBeAg, normal ALT, for HBeAg neg maintenance of HBeAg neg status

A strong dose response of ETV was demonstrated at week 24 in analyses of HBV DNA reduction by PCR assay and the primary endpoint (HBV DNA <LOQ by bDNA assay). The 1.0 mg dose proved to be superior to the 0.5 mg dose with respect to the primary endpoint (p<0.01). With regard to HBV DNA reduction by PCR at Week 24, the difference estimate between the 1.0 and 0.5 mg doses was -0.63 log$_{10}$ and not significant (p=0.1). With respect to the secondary endpoints, the 1.0 mg produced more consistent results than the 0.5 mg, although differences between dose groups were not always apparent. It is to be noted that ETV 1.0 mg seemed to be efficacious in suppressing HBV DNA across all prognostic and demographic subgroups. With respect to ALT normalisation, the 1.0 mg group had the greatest response at Week 48 (68%). Despite good viral suppression, few patients achieved HBeAg loss after 48 weeks of therapy. Due to this fact, a limited number of subjects achieved complete response at Week 48, with higher proportion in the 1.0 mg group.

No apparent dose response relationship was observed with respect to safety issue. Consequently, 1.0 mg was chosen for the main study in LVD refractory patients.

- Main study(ies)

Studies in nucleoside naïve patients: study AI463022 (HBeAg positive) and AI463027 (HBeAg negative)

METHODS

Study Participants
All study subjects had documented chronic hepatitis B (i.e. detectable HBsAg ≥ 6 months), detectable HBV DNA and compensated liver disease. An overview of the inclusion criteria is presented in table 5. Nucleoside-naïve subjects were defined as those that had received ≤12 weeks of prior nucleos(t)ides, and the last dose of any prior therapy was to be administered >24 weeks prior to randomisation.

Table 5: Eligibility criteria

Eligibility criterion	Nucleoside-naive	
	022	027
Males and females ≥ 16 years with history of chronic hepatitis B	+	+
HBV viraemia		
HBV DNA ≥0.7 MEq/ml bDNA *or* >1.5x10^6 copies/ml by PCR ≥4 weeks prior to screening	+	-
HBV DNA ≥0.7 MEq/ml by bDNA or >10^5 copies/ml by PCR ≥2 weeks prior to screening	-	+
HBV DNA ≥ 3.0 MEq/ml by bDNA at screening	+	-
HBV DNA ≥ 0.7 MEq/ml by bDNA at screening	-	+
HBV DNA ≥ 10 MEq/ml by bDNA at screening	-	-
HBV serology		
Detectable HBsAg ≥6 months or detectable HBsAg <6 months and neg IgM anti HBc and confirmation of chronic hepatitis on liver biopsy	+	+
HBeAg positive at screening and at least once ≥4 weeks prior to screening	+	-
HBeAg neg/anti HBe pos at screening and at least once ≥4 weeks prior to screening	-	+
HBeAg could be positive or negative at screening	-	-
ALT		
ALT 1.3 to 10xULN at screening and at least once ≥12 weeks prior to screening	+	+
ALT and AST normal to ≤10 x ULN at screening	-	-
Liver Biopsy		
Evidence of chronic hepatitis on biopsy performed ≤52 weeks prior to randomisation	+	+

Among exclusion criteria, there was co-infection with HIV, HCV or HDV.

Treatments

Patients were randomised to receive entecavir (ETV) 0.5 mg once daily (QD) or to lamivudine (LVD) 100 mg QD.
All studies had an initial period of 52-week blinded dosing, (Year 1) followed by an extended blinded dosing period (Year 2) for those who were partial responders at week 52 (see table 6 for the definition of response), and a 24-week post-dosing follow-up period. The initial assessment of the response to therapy was made at Week 48. The patient management decisions at Week 52 were based on the treatment response at Week 48.

- In case of Complete Virologic Response patients were to stop treatment and be followed off treatment for 24 weeks for safety and sustained response.
- In case of Partial virologic Response patients were to continue blinded treatment until Complete Response was achieved or until Week 96, whichever occurred first.
- Virologic Nonresponders were to discontinue blinded therapy and begin appropriate therapy recommended by their physician or enrol in a separate open-label rollover protocol or early access program. Subjects electing not to enrol in another study were to be followed for safety every 4 weeks for 24 weeks in the current study after discontinuation of study therapy.
- Relapse: for Partial Virologic Responders whose HBV DNA by bDNA assay became detectable during the second year of treatment and subjects who relapse after Complete virologic Response
- Rebound: virologic Rebound, defined as confirmed ≥ 1-log$_{10}$ increase from nadir in HBV bDNA on treatment. Samples from subjects who had virologic rebound were submitted for genotypic and phenotypic analyses to identify HBV DNA mutations associated with reduced susceptibility to study therapy.

Table 6: Response definitions

Term	Definition	Study
Complete response	HBV DNA < 0.7 MEq/ml by bDNA _and_ loss of HBeAg	022
Composite response	HBV DNA < 0.7 MEq/ml by bDNA _and_ ALT < 1.25 x ULN	027
Partial response	HBV DNA < 0.7 MEq/ml by bDNA _but_ HBeAg +	022.
Virologic-only response	HBV DNA 0.7 MEq/ml by bDNA _but_ ALT ≥1.25x ULN	027
Non-response	HBV DNA ≥ 0.7 MEq/ml by bDNA	022, 027

Objectives

The primary objective of both studies was to compare the efficacy of 48-week treatment course of entecavir 0.5 mg once daily versus lamivudine 100 mg daily in the treatment of chronic hepatitis B with compensated liver disease.

Secondary objectives included assessment of:
- Sustained response and relapse rate during 24 week off-treatment follow-up of patients with composite/complete response at week 48
- 96-week efficacy in patients with partial response continuing blinded therapy
- 48- and 96-week safety
- Emergence of drug-resistant virus

Outcomes/endpoints

In both studies, the primary efficacy measure was the proportion of subjects in each treatment group who achieved histological improvement defined as ≥ 2-point decrease in the Knodell necroinflammatory score with no worsening of fibrosis (worsening: ≥ 1-point increase in the Knodell fibrosis score) at the Week 48 liver biopsy compared to baseline.

Secondary efficacy endpoints at Week 48 were:

Further assessment of histological response
- Proportions of patients with improvement in hepatic fibrosis (≥ 1 point decrease from baseline) using the Ishak scoring system,
- Reduction from baseline in hepatic covalently closed circular DNA (cccDNA), total hepatic HBV DNA, hepatitis B core antigen (HBcAg) and hepatic hepatitis B surface antigen (HBsAg)

Virological response
- Proportions of patients with HBV DNA < 0.7 MEq/ml (700 000 copies/ml) by bDNA assay
- Proportions of patients with HBV DNA < 400 copies/ml by PCR assay (post hoc: <200 copies/ml, later amended as 300 copies/ml following a revision of the lower limit of quantification by the manufacturer)
- Mean $\log_{10}$ reduction in HBV DNA levels from baseline by PCR assay

Serological response (study 022 only)
- Proportions of patients with loss of HBeAg in subjects with positive e antigen prior to treatment
- Proportions of patients with seroconversion (HBeAg → anti-HBe)
- Proportions of patients with loss of HBsAg

Biochemical response
- Proportions of patients with normalization of serum ALT (<1.25 x ULN) at week 48, later changed to ≤1.0 x ULN threshold

Combined/composite endpoints (patient management endpoints)
- Proportion of patients with complete virological response
- Proportion of patients with complete virological response plus normalization of ALT

- Proportion of patients with partial virological response
- Proportion of subjects who demonstrated Response for the composite endpoint (*study 027*)

HBV resistance
- Genotypic and phenotypic analyses in patients with virologic rebound ($\geq$1-$\log_{10}$ increase in HBV DNA from nadir, as determined by 2 sequential HBV DNA by bDNA assays during blinded treatment)
- for 223 randomly selected entecavir-treated subjects, a genotypic screening of samples was performed to monitor the appearance of genotypic changes (022)
- In study 027 random selected samples were collected for genotypic and phenotypic analyses

Sample size

In both studies, a 2-stage evaluation was planned for assessment of the primary endpoint. In the first stage, non-inferiority of ETV to LVD was tested (the boundary for the lower limit of the 95% confidence interval (CI) for the difference in proportions (ETV - LVD) was -10%). The target sample size was adequately calculated. For the second stage, provided non-inferiority was established, a second test for superiority was to be conducted by checking if the lower limit of the 95% confidence interval was greater than zero.

Randomisation

Patients were randomised 1:1 to entecavir or lamivudine. A randomisation block design stratified by study site was used.

Blinding (masking)

The study was adequately blinded. A central pathologist performed a blinded assessment of liver histology (sequence-blinded and treatment-blinded biopsy pairs).

Statistical methods

The Intention to Treat (ITT) efficacy data set include data collected on treated subjects who were defined as randomised subjects treated with at least 1 dose of study therapy, entecavir or lamivudine. Analyses of efficacy endpoints focused on treated subjects who were evaluable for response, *i.e.* had measurements at baseline and at the Week 48 visit. Two methods were used for handling missing data: Non-Completer=Failure (NC=F), where subjects with missing or inadequate Week 48 data were treated as failures (modified ITT) and Non-Completer=Missing (NC=M), where subjects with missing Week 48 data were excluded from the analysis.

RESULTS

At the time of the submission of the application, both studies had completed 48 weeks of treatment and data on all these patients were the basis for the efficacy assessment. Supplementary data on the long term efficacy and safety (Week 96) were submitted, during the procedure, including data on sustained response rates after a 24-week off-treatment period.

Participant flow

The disposition of patients is presented in table 7: Overall a large proportion of patients completed 48 weeks period.

Table 7: Disposition of patients

Parameter	Study 022 (HBeAg pos)			Study 027(HBeAg neg)		
	Total	ETV 0.5mg	LDV 100mg	Total	ETV 0.5mg	LDV 100mg
Disposition	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
N randomised	715	357	358	648	331	317
Received at least one dose (ITT)	709	354	355	638	325	313
Completed 48 weeks	661 (92%)	340 (95%)	321 (90%	607 (94%)	311 (94%)	296 (93%)
Discont before week 48	48 (7)	14 (4)	34 (9)	31 (5%)	14 (4%)	17 (5%)
Reasons for discontinuation						
Adverse event	10 (1)	1 (1)	8 (2)	15 (2)	6 (2)	9 (3)
Withdrew consent	11 (2%)	6 (2)	5 (1)	8 (1)	4 (1)	4 (1)
Lost to FU	11 (2)	3 (<1)	8 (2)	2 (<1)	0	2 (<1)
Non-compliance	6 (<1)	2 (<1)	4 (1)	4 (<1)	2(<1)	2 (<1)
Pregnancy	4 (<1)	2 (<1)	2 (<1)			
Death	2 (<1)	0	2 (<1)	2 (<1)	0	2 (<1)
Lack of efficacy				0	0	0
Other	4 (1)	0	4 (1)	14 (2)		
Completed first year of dosing	661 (92)	340 (95)	321 (90)	607 (94)	311()4)	296 (93)
Continued to 2nd year	443 (62%)	253 (71%)	190 (53%)	105(16%)	46 (14%)	59 (19%)
Did not complete 2nd year of dosing	89 (12%)	22 (6%)	67 (19%)	19 (3 %)	7 (2%)	12 (4 %)
Treatment failure /lack of efficacy	68 (10%)	7 (2%)	61 (17%)	9 (1 %)	0	9 (3%)
Adverse event	1 (<1%)	0	1 (<1%)			
Completed 2nd year of dosing	**354 (50%)**	**231 (65%)**	**123 (34%)**	82 (13%)	38 (11%)	44 (14%)

Baseline data

Patients were well balanced between treatment groups for baseline HBV disease characteristics (table 8).

Table 8: Main baseline characteristics

	Study 022 HBeAg positive		Study 027 HBeAg negative	
	ETV n=354	LVD n=355	ETV n=325	LVD n=313
Age years				
Mean	35	35	44	44
Median	33	32	45	45
Gender n (%)				
Male	274 (77)	261 (74)	248 (76)	236 (75)
Female	80 (23)	94 (26)	77 (24%)	77 (25)
Race n (%)				
Caucasian	140 (40)	141 (40)	**193 (59)**	**176 (56)**
Asian	**204 (58)**	**202 (57)**	122 (38)	129 (41)
Black	8 (2)	8 (2)	8 (2)	7 (2)
Other)	2 (<1)	4 (1)	2 (<1)	1 (<1)
Region n (%)				
Europe	84 (24)	88 (25)	**156 (48)**	**148 (47)**
Asia	**172 (49)**	**167 (47)**	106 (33)	104 (33)
N (%) available baseline biopsy	329 (93)	330 (93)	303 (93)	293 (93)
Knodell Necroinflammatory score				
Mean (SD)	7.8 (2.98)	7.7 (2.99)	8.0 (2.75)	7.7(2.76)
median	9.0	8.0	9.0	8.0
Proportion scores ≥2	314 (89%)	314 (88%)	296 (91%)	287 (92%)
Knodell Fibrosis scores				
mean (SD)	1.7 (1.11)	1.7 (1.11)	1.9 (1.11)	1.9(1.14)
median	1.0	1.0	1.0	1.0
Cirrhosis n (%)	**25 (7)**	**27 (8)**	**19 (6)**	**28 (9)**

Ishak fibrosis scores				
mean (SD)	2.3 (1.27)	2.3 (1.29)	2.4 (1.18)	2.5(1.29)
median	2.0	2.0	2.0	2.0
Cirrhosis n (%)	**11 (3)**	**11 (3)**	**5 (2)**	**10 (3)**
HBV DNA bDNA				
mean (SD)	2.56 (1.05)	2.61 (1.025)	1.24 (0.96)	1.23 (1.01)
median	2.79	2.84	1.20	1.06
HBV DNA PCR				
mean	9.62 (2.0)	9.69 (1.98)	7.60 (1.75)	7.55(1.7)
median	9.28	9.32	7.51	7.51
HBV subtypes				
A	98 (27)	104 (29)	34 (10)	33 (11)
B	68 (19)	78 (22)	48 (14)	61 (19)
C	111 (31)	90 (25)	58 (18)	51 (16)
D	41 (11)	51 (14)	**161 (48)**	**143 (43)**
E	2 (<1)	0	1 (<1)	1 (<1)
F	20 (6)	13 (3)	1 (<1)	2 (<1)
ALT				
mean (SD)	140.5	146.3	141.0	142.5
median	102.0	103.0	106.0	105.0
range	20-915	19-1106	19-727	7-878
Prior therapy				
Interferon	45 (13)	45 (13)	42 (13)	37 (12)
LDV/ Famciclovir	10 (3)	10 (3)	9 (3)	12 (4)

- **Results of Study AI463022- HBeAg positive patients**

Efficacy analyses at Week 48

Entecavir was superior to lamivudine for the proportion of treated subjects demonstrating histological improvement at the Week 48 biopsy using the NC=F method and non-inferior using NC=M method as shown in table 9.

Table 9: Histological improvement at week 48, treated subjects with histology pairs

Primary endpoint	ETV (n=354)	LVD (n=355)	Difference estimates [ETV-LVD] (95%CI)
Histological improvement NC=F	226/314 (72%)	195/314 (62%)	9.9 (2.6, 17.2) p=0.0085
Histological improvement NC=M	226/292 (77%)	195/269 (72%)	4.9 (-2.3,12.1) p=0.191

Entecavir was non-inferior to lamivudine for the proportion of subjects demonstrating improvement in the Ishak fibrosis score at the Week 48 biopsy using the NC = F method, with 39% in ETV group versus 35% in LVD (difference estimate [ETV-LVD] = 3.2; 95% CI [-4.4, 10.7]; p = 0.41). 8 % in the entecavir group compared to 5 % in the lamivudine group had a worsening Ishak fibrosis scores at Week 48.

The mean change from baseline in hepatic HBV cccDNA ($\log_{10}$ copies/HGEq) was -0.9 $\log_{10}$ copies/HGEq for entecavir and -0.7 $\log_{10}$ copies/HGEq for lamivudine. The difference estimate (ETV-LVD) for hepatic cccDNA reduction adjusted for baseline hepatic cccDNA level was -0.2 $\log_{10}$ copies/HGEq (95% CI [-0.3, -0.1], p = 0.0033). Results favoured entecavir for the reduction from baseline in total hepatic HBV DNA at Week 48 (95% CI [-0.6, -0.3], p < 0.0001).

The entecavir and lamivudine groups were similar for improvement (≥1-point decrease from baseline) with respect to the hepatic antigen endpoints: hepatic nuclear HBcAg, hepatic cytoplasmic HBcAg, and hepatic HBsAg.

Table 10 provides a summary of the secondary endpoints for antiviral activity, immunologic and biochemical responses.

Table 10: Summary of secondary endpoints at week 48 (NC=F)

Secondary endpoint	ETV (n=354)	LVD (n=355)	Difference estimates [ETV-LVD] (95%CI)
HBV DNA <0.7 MEQ/ml by bDNA assay	322 (91%)	232 (65%)	25.6 (19.8, 31.4) p<0.0001
Mean change by PCR ($\log_{10}$ copies/ml)*	- 6.86	- 5.39	1.5 (-1.8, -1.3) p< 0.0001
Median change by PCR	- 6.78	-5.60	
HBV DNA <400 copies/ml	246 (69%)	135 (38%)	31.5 (24.5, 38.4) p<0.0001
HBV < 300 copies/ml	236 (67 %)	129 (36 %)	30.3 (23.3, 37.3) p < 0.0001
Loss of HBeAG	78 (22%)	70 (20%)	2.3 (-3.7, 8.3) p=0.45
Seroconversion	74 (21%)	64 (18%)	2.9 (-2.9, 8.7) p=0.33
ALT normalisation (< 1.0 x ULN)	242 (68 %)	213 (60 %)	8.4 (1.3, 15.4) p = 0.0202
Patient management endpoints			
Complete virological response	74 (21%)	67 (19%)	
Partial virological response	247 (70%)	165 (46%)	
Virological non-response	19 (5%)	95 (26%)	
Missing Week 48 data	14 (4%)	29 (8%)	
Complete response + ALT normalisation	65 (18%)	64 (18%)	0.5 (-5.3, 6.0) p=0.91

* limit of quantitation 300 copies/ml

Entecavir was shown to be non-inferior or superior to lamivudine with respect to histological, virological, serological and biochemical response at Week 48. Despite greater antiviral potency of ETV, the proportion of complete virological responders, i.e. with HBeAg loss, was low in both treatment groups. Thus at week 48, the more potent antiviral activity of ETV did not have any implications on treatment when compared to LVD. However there were higher numbers of ETV treated subjects than LVD with partial virological response, enabling more subjects to continue for a second year of therapy.

Efficacy results at Week 96

Table 11: Response status through Year 2 – treated subjects

First year of treatment	ETV n=354	LVD n =355
Complete response* after Year 1	74 (21%)	67 (19%)
Virological Only responder at Year 1	247 (70%)	165 (46%)
Non-responder at Year 1	19 (5%)	94 (26%)
Second Year of treatment	ETV n=243	LVD n =164
Complete response at Year 2 EOD**	37 (15%)	26 (16%)
< Week 96	18 (7%)	16 (10%)
>=Week 96	19 (8%)	10 (6%)
Virological only responder at Year 2 EOD	198 (81%)	85 (52%)
Non-responder at Year 2 EOD	8 (3%)	53 (32%)
Total responder (Week 48 + Week 96)	**111 (31%)**	**93 (26%)**
Loss of HBeAg during the 2nd year at EOD	15%	18%
Seroconversion during the 2nd year at EOD	11%	13%
ALT normalisation	79%	68%
Cumulative responses through Year 2		
Cumulative HBV DNA <0.7 MEq/ml	97%	68%
Cumulative HBV DNA <300 copies/ml	80%	39%
Mean change from baseline ($\log_{10}$ copies/ml) at Year 2 EOD	-7.09	-4.85
Cumulative HBeAg loss at Year 2 EOD	33%	28%
Cumulative confirmed HBeAg seroconversion EOD	31%	25%
Cumulative ALT normalisation <1.0 x ULN EOD	78%	61%
Cumulative ALT normalisation <1.25 x ULN EOD	86%	70%
Cumulative loss of HBsAg	18 (5%)	10 (3%)
Cumulative HBsAg seroconversion	6 (2%)	8 (2%)

* Complete Responder = HBV DNA <0.7 MEq/ml and loss of HBeAg. **EOD = end of dosing

Histological response was only evaluated in less than 10% of patients at Week 96 and data suggested improvement of fibrosis during the 2nd year. The total complete response rate at Week 96 was 31% for ETV treatment and 26% for the LVD treatment group. Thus, ETV patients with a partial response experienced incremental benefit during the second year of treatment, although the cumulative HBeAg

seroconversion rate was relatively low (31% Year 2 vs 21% Year 1) and not different from LVD. No resistance development was detected for up to 96 weeks of ETV therapy.

Sustained response outcome

The sustained response rate 6 months after discontinuation of treatment is presented in table 12 for the total number of entecavir-treated responders (111) and lamivudine-treated responders (93).

Table 12: Sustained responses outcome at 24 weeks off treatment

	ETV n=111	LVD n=93
Sustained complete response	**83 (75%)**	**68 (73%)**
Failures with loss of response	24 (22%)	22 (24%)
Re-emergence of HBeAg	16 (67%)	4 (18%)
HBV DNA ≥0.7MEq/ml *and* re-emerg. of HBeAg	4 (17%)	17 (77%)
Sustained complete response *and* ALT ≤ 1.0 x ULN	69 (56%)	49 (55%)
Sustained complete response *and* ALT <1.25 x ULN	75 (64%)	57 (59%)

The highest sustained response rate was observed among subjects that responded early (< 54 weeks).

Preliminary responder analysis at Week 48 off-treatment included 74 (89%) ETV patients and 67 LVD patients. In the ETV group, 42 (57%) patients maintained their response, 21 (28%) still had an HBV DNA level <300 copies/ml. Twenty-six (25%) patients had relapsed, 14 with HBV DNA levels >10^5 copies/ml and/or seroreversion (HBeAg+) and 12 had resumed anti-HBV therapy. Data were missing on 13 (18%) ETV patients. For LVD only 33% of subjects maintained a complete response. However, data were missing on 17/67 (25%) patients.

- **Results of Study AI463027- HBeAg negative patients**

Efficacy analyses at week 48

Entecavir was superior to lamivudine for the proportion of treated subjects demonstrating histological improvement at the Week 48 biopsy using both the NC=F and NC=M methods (see table 13).

Table 13: Histological improvement at week 48, treated subjects with histology pairs

Primary endpoint	ETV (n=325)	LVD (n=313)	Difference estimates [ETV-LVD] (95%CI)
Histological improvement NC=F	208/296 (70%)	174 /287(61%)	9.6 (2.0, 17.3) p=0.0143
Histological improvement NC=M	208/265 (78%)	174/250 (70%)	8.9 (-2.3,12.1) p=0.0212

Entecavir was non-inferior to lamivudine for the proportion of subjects who demonstrated improvement in hepatic fibrosis using Ishak score at Week 48. The proportions were 36% and 38%, respectively (NC = F method) (difference estimate [ETV-LVD] = -1.8; 95% CI –9.7, 6.0]; p = 0.65). 12% in the entecavir group compared to 15 % in the lamivudine group had a worsening Ishak fibrosis scores at Week 48.
The entecavir and lamivudine groups showed reductions from baseline at Week 48 for the following hepatic endpoints:
- HBV cccDNA (mean change: -0.5 $\log_{10}$ copies/HGEq for both groups; 95% CI [-0.2, 0.1]; p = 0.50)
- Total hepatic HBV DNA (mean change: entecavir -1.5 $\log_{10}$ copies/HGEq; lamivudine - 1.4 $\log_{10}$ copies/HGEq; 95% CI [-0.2, 0.0]; p = 0.07)
- Loss of HBsAg through week 48: only one subject in each group has loss of HBsAg at week 48.

Table 14 provides a summary of the secondary endpoints for antiviral activity, immunologic and biochemical responses.

Table 14: Summary of secondary endpoints at week 48 (NC=F)

Secondary endpoint	ETV (n=325)	LVD (n=313)	Difference estimates [ETV-LVD] (95%CI)
HBD DNA <0.7 MEq/ml by bDNA assay	309 (95%)	279 (89%)	5.9 (1.8, 10.1) p<0.0053
Mean change HBV DNA by PCR $\log_{10}$ *	- 5.04	- 4.53	- 0.43 (-0.6, -0.26) p < 0.0001
HBV DNA <400 copies/ml	297 (91%)	230 (73%)	17.9 (12.1, 23.7) p<0.0001
HBV DNA <300 copies/ml	293 (90%)	225 (72%)	18.3 (12.3, 24.2) p < 0.0001
ALT normalisation (< 1 xULN)	253 (78%)	222 (71%)	6.9 (0.2, 13.7) p = 0.0451
Patient management endpoints			
Complete virological response**	275 (85%)	245 (78%)	6.4 (0.3, 12.4) p=0.041
Virologic-only response	34 (10%)	34 (11%)	
Virologic non-response	3 (<1%)	18 (6%)	
Missing Week 48 data	13 (4%)	16 (5%)	

* limit of quantitation 300 copies/ml **Composite endpoint: HBV DNA<0.7 MEq/ml +ALT normalisation (<1.25xULN)

ETV was superior to LVD for almost all endpoints in this difficult-to-treat population. No resistance mutation associated with ETV was detected in this study and no ETV patients experienced viral breakthrough by PCR assay.

Efficacy results at Week 96

Table 15: Response status through Year 2 – treated subjects

First year of treatment	ETV n=325	LVD n =313
Composite responders* after Year 1	275 (85%)	245 (78%)
Virological Only responder at Year 1	34 (10%)	34 (11%)
Non-responder at Year 1	3 (<1%)	18 (6%)
Virologic-Only responder continuing 2nd year of dosing	**ETV n=26**	**LVD n=28**
Composite responders at EOD	11 (42%)	8 (29%)
Virologic-Only Responder at EOD	15 (58%)	15 (54%)
Non responder at EOD	0	5 (18%)
Total rate of composite responders (W48+W96)	286/325 (**88%**)	253/313 (**81%**)
Cumulative responses through Year 2	**N=325**	**N=313**
Cumulative HBV DNA <300 copies/ml at EOD	92%	72%
Mean change from BL ($\log_{10}$ copies/ml) at Year 2 EOD	5.14	4.13
Cumulative ALT normalisation <1.0 x ULN EOD	89%	84%
Cumulative loss of HBsAg	1 (<1%)	2 (<1%)
Cumulative HBsAg seroconversion	0	1 (<1%)

* Composite Responder = HBV DNA <0.7 MEq/ml and ALT<1.25 x ULN.

Continued treatment with ETV for up to 2 years appeared to result in maintenance of clinical benefit. The total rate of treatment response for both years was 88% (286/325) for the ETV group versus 81% (253/313) for the LVD group. No resistance development was detected for up to 96 weeks of ETV therapy.

Sustained response outcome

At Week 24 off-treatment, 46% of ETV-treated responders maintained their response versus 31% for LVD. At Week 48 these proportions dropped to 17% for ETV and 12% for LVD. An assessment of the reasons for failure suggested that many patients who documented failure at Week 24 off-treatment had started alternate HBV therapy by Week 48 of follow-up. Missing data in subjects who were lost to follow-up account for an increasingly large minority of patients at Week 48 off-treatment: 20% for ETV versus 12% for LVD.

The analysis by categories of HBV DNA demonstrated that even though more ETV-treated patients had an HBV DNA $<10^4$ copies/ml at Week 24 off-treatment (27%) as compared with LVD (18%), by 48 weeks off-treatment these proportions decreased to approximately 10% in both treatment arms (11% for ETV; 9% for LVD). Therefore, although the HBV DNA of ETV-treated patients rebounds more slowly off-treatment than does the HBV DNA of LVD-treated patients, at 1 year off-treatment both treatment groups have comparable distributions of HBV DNA.

©EMEA 2006

- **Study in lamivudine refractory patients: study AI463026**

In addition to the results from the phase II study AI463014 already presented in the dose ranging section of this document, a confirmatory study (026) was conducted.

Study participants

LVD-refractory subjects were patients with incomplete response to lamivudine; defined as: i) persistently detectable HBV DNA by bDNA assay after at least 36 weeks of lamivudine therapy, **or** ii) breakthrough viraemia while on lamivudine, **or** iii) recurrence of HBV viraemia following lamivudine discontinuation which persisted despite resuming lamivudine *or* documented lamivudine-resistant (LVD^R) mutation and HBV viraemia while on lamivudine.

Study Participants
All study subjects had documented chronic hepatitis B (i.e. detectable HBsAg $\geq$ 6 months), detectable HBV DNA and compensated liver disease (see table 16 for inclusion criteria).

Table 16: Eligibility criteria

Eligibility criterion	
Males and females $\geq$ 16 years with history of chronic hepatitis B	+
HBV DNA $\geq$0.7 MEq/ml bDNA *or* >1.5x10^{6}copies/ml by PCR $\geq$4 weeks prior to screening	+
HBV DNA $\geq$ 3.0 MEq/ml by bDNA at screening	+
Detectable HBsAg $\geq$6 months or detectable HBsAg <6 months and neg IgM anti HBc and confirmation of chronic hepatitis on liver biopsy	+
HBeAg positive at screening and at least once $\geq$4 weeks prior to screening	+
ALT 1.3 to 10xULN at screening and at least once $\geq$12 weeks prior to screening	+
Evidence of chronic hepatitis on biopsy performed $\leq$52 weeks prior to randomisation	+
LVD therapy $\geq$ 24 weeks (in 014) or $\geq$36 weeks (in 026) or documented	+

Among exclusion criteria, there were co-infection with HIV, HCV and HDV and decompensated liver function.

Treatments

Subjects received either ETV 1.0 mg QD or continued LVD 100 mg QD.

The study had an initial period of blinded dosing (Year 1) followed by an extended blinded dosing period (Year 2), and a 24-week post-dosing follow-up period. The initial assessment of the response to therapy was made at week 48.

Patient management decisions at Week 52 were based on the treatment response at Week 48. Subjects who demonstrated a complete/composite response (HBV DNA < 0.7 MEq/ml by bDNA *and* loss of HBeAg) were to stop therapy and be followed off-treatment for up to 24 weeks to assess sustained response and post-dosing safety. Subjects who demonstrated a partial/virological-only response (HBV DNA < 0.7 MEq/ml by bDNA *but* HBeAg +) were to continue blinded therapy up to Week 96.
No response was defined as HBV DNA $\geq$ 0.7 MEq/ml by bDNA.

Objectives

The primary objective was to compare the efficacy of a 48-week treatment course of entecavir 1 mg once daily versus lamivudine 100 mg daily in the treatment of chronic hepatitis B with compensated liver disease.

Secondary objectives included assessment of:

©EMEA 2006

- Sustained response and relapse rate during 24 week off-treatment follow-up of patients with composite/complete response at week 48
- 96-week efficacy in patients with partial response continuing blinded therapy
- 48- and 96-week safety
- Emergence of drug-resistant virus

Outcomes/endpoints

There were 2 co-primary endpoints:
- Histologic Improvement:

the proportion of subjects at Week 48 in each treatment group with Histologic Improvement (a ≥ 2-point decrease from baseline in Knodell necroinflammatory score) and no worsening of fibrosis (worsening: ≥ 1 point increase from baseline in the Knodell fibrosis score)
- Composite Endpoint:

the proportion of subjects at Week 48 in each treatment group with undetectable HBV DNA by bDNA assay (< 0.7 MEq/ml) and normalization of serum ALT (< 1.25 × ULN).

Secondary efficacy endpoints at Week 48 included:

Further assessment of histological response
- Proportion of patients with improvement in hepatic fibrosis using the Ishak scoring system,
- Reduction from baseline in hepatic covalently closed circular DNA (cccDNA), total hepatic HBV DNA, hepatitis B core antigen (HBcAg) and hepatic hepatitis B surface antigen (HBsAg)

Virological response (week 24 study 014)
- Proportion of patients with HBV DNA < 0.7 MEq/ml (700 000 copies/ml) by bDNA assay
- Proportion of patients with HBV DNA < 400 copies/ml by PCR assay (post hoc: <200 copies/ml assay, later amended as 300 copies/ml following a revision of the lower limit of quantification by the manufacturer).
- Mean $\log_{10}$ reduction in HBV dNA levels from baseline by PCR assay

Serological response
- Proportion of patients with loss of HBeAg in subjects with positive e antigen prior to treatment
- Proportion of patients with seroconversion (HBeAg $\rightarrow$ anti-HBe)

Biochemical response
- Proportions of patients with normalization of serum ALT (<1.25 x ULN), later changed to ≤ 1.0 x ULN threshold

Combined endpoints (patient management endpoints)
- Proportion of patients with complete virological response
- Proportion of patients with complete virological response plus normalisation of ALT
- Proportion of patients with partial virological response

Sustained response
Proportions of patients with sustained virological, serological and biochemical response during 24 off-treatment follow-up

HBV resistance
- Genotypic and phenotypic analyses in patients with virologic rebound (≥ 1-$\log_{10}$ increase in HBV DNA from nadir, as determined by 2 sequential HBV DNA by bDNA assays during blinded treatment)
- For all subjects frozen serum sample from Day 1 for genotyping and for identifying viral mutations in the DNA polymerase gene

Sample size

A superiority test was performed separately for the 2 different primary endpoints. A Bonferroni adjustment was applied for testing superiority, with an overall significant level of 2.5% for each endpoint. ETV was considered to be superior to LVD if the lower limit of the 97.5% CI was greater then zero. The target sample size was adequately calculated.

Randomisation

Eligible subjects were randomised (1:1) in a double-blind manner to receive either entecavir or lamivudine. A randomisation block design stratified by study site was used.

Blinding (masking)

The study was adequately blinded. A central pathologist read performed a blinded assessment of liver histology (sequence-blinded and treatment-blinded biopsy pairs).

Statistical methods

Analyses of efficacy endpoints focused on treated subjects who were evaluable for response, i.e. had measurements at baseline and at the Week 48 visit. Two methods were used for handling missing data: Non-Completer=Failure (NC=F), where subjects with missing or inadequate Week 48 data were treated as failures (modified ITT) and Non-Completer=Missing (NC=M), where subjects with missing Week 48 data were excluded from the analysis.

RESULTS

At the time of the submission of the application, study had completed 48 weeks of treatment and data on all these patients were the basis for the efficacy assessment. Supplementary data on the long term efficacy and safety (Week 96) were submitted, during the procedure, including data on sustained response rates after a 24-week off-treatment period.

Participant flow

The disposition of patients is presented in table 17. Overall a large proportion of patients completed 48 weeks period.

Table 17: Disposition of patients

Parameter	Total	ETV 1.0mg	LDV 100mg
Disposition	n (%)	n (%)	n (%)
N randomised	293	147	146
Received at least one dose (ITT)	286 (98%)	141 (96 %)	145 (99 %)
Completed 48 weeks	259 (88%)	133 (90%)	126 (86%)
Discont before week 48	27 (9)	8 (5)	19 (13)
Reasons for discontinuation			
Adverse event	9 (3)	1 (<1)	8 (5)
Withdrew consent	7 (2)	2 (1)	5 (3)
Lost to FU	3 (1)	2 (1)	1 (<1)
Non-compliance	4 (1)	3 (2)	1 (1)
Pregnancy			
Death	1 (<1)	0	1 (<1)
Lack of efficacy	2 (<1)	0	2 (1)
Other			
Completed first year of dosing	259 (88)	133 (90)	126 (86)
Continued to 2nd year	115 (41%)	91 (62%)	24 (16%)
Did not complete 2nd year of dosing	33 (11%)	16 (11%)	17 (12%)
Treatment failure /lack of efficacy	28(10%)	13 (9%)	15 (10%)
Completed 2nd year of dosing	**74 (25%)**	**73 (50%)**	**1(<1%)**
Entered 24-week Follow-up	**80 (28%)**	**58 (41%)**	**22 (15%)**
Discontinued 24 week follow-up	29 (10%)	24 (17%)	5 (3%)
Treatment failure /lack of efficacy	20 (7%)	19 (13%)	1 (<1%)
Adverse event	1 (<1%)	1 (<1%)	0
Completed 24 week follow-up	**40 (14%)**	**28 (20%)**	**12 (8%)**

The mean time on study therapy was 76.7 weeks for the ETV group and 52.6 weeks for the LVD group. The maximum time on study therapy was 114.9 weeks for ETV and 87.3 weeks for LVD. The mean time on study therapy for Virologic-Only Responders was 94.8 weeks for the ETV group and 79.8 weeks for the LVD group.

The demographics and baseline HBV characteristics are displayed in table 18.

Table 18: Main baseline characteristics

	ETV n=147	LVD n=146
Age years		
Mean	39	39
Median	38	40
Gender n (%)		
Male	105 (74)	112 (77)
Female	36 (26)	33 (23)
Race n (%)		
Caucasian	**83 (59)**	**93 (64)**
Asian	57 (40)	50 (34)
Black	0	0
Other)	1 (<1)	2 (1)
Region n (%)		
Europe	**62 (44)**	**72 (50)**
Asia	35 (25)	36 (25)
N (%) available baseline biopsy	135 (92)	133 (91)
Knodell Necroinflammatory score		
Mean (SD)	6.5(3.23)	6.5 (3.41)
median	7.0	7.0
Proportion scores ≥2	124 (88%)	116 (80%)
Knodell Fibrosis scores		
mean (SD)	1.7(1.19)	1.8 (1.18)
median	1.0	1.0
Cirrhosis n (%)	**14(10)**	**9 (6)**
Ishak fibrosis scores		
mean (SD)	2.3 (1.5)	2.2 (1.41)
median	2.0	2.0
Cirrhosis n (%)	**9 (6)**	**5 (4)**
HBV DNA bDNA		
mean (SD)	2.50 (0.95)	2.50 (0.93)
median	2.66	2.67
HBV DNA PCR		
mean	9.48(1.8)	9.24 (1.6)
median	9.32	9.29
HBV subtypes		
A	38 (26)	36 (25)
B	23 (16)	17 (12)
C	27 (19)	28 (19)
D	**46 (33)**	**56 (39)**
E	0	0
F	4 (3)	3 (2)
ALT		
mean (SD)	123.9	131.9
median	88.0	83.0
range	21-726	20-1075
Prior therapy		
Interferon	63 (45)	78 (54)
LDV/ Famciclovir	141(100)	144 (99)

- **48-week primary efficacy analyses**

Overall results on the co-primary endpoint demonstrated superiority of entecavir over lamivudine as shown in the table 19.

Table 19: Histological improvement at week 48, treated subjects with histology pairs

		ETV 1.0 mg	LVD 100 mg
	Principal Analyses (NC = F)	124	116
	(1) Histologic Improvement		
Number with Improvement/		68/124 (55)	32/116 (28)
Subjects with Evaluable Baseline Histology (%)			
Difference Estimate (ETV - LVD)		27.3	
95% CI		(13.6, 40.9)	
p-value		< 0.0001	
	(2) Composite Endpoint		
(HBV DNA < 0.7 MEq/ml by bDNA assay and ALT < 1.25 × ULN)			
Number Meeting Composite Endpoint/Subjects Evaluable		77/141 (55)	6/145 (4)
Difference Estimate (ETV - LVD)		50.5	
95% CI		(40.4, 60.6)	
p-value		< 0.0001	
	Secondary Analyses (NC = M)	110	98
	(1) Histologic Improvement		
Number with Improvement/		68/110 (62)	32/98 (33)
Subjects with Evaluable Histology Pairs (%)			
Difference Estimate (ETV - LVD)		29.2	
95% CI		(14.3, 44.0)	
p-value		< 0.0001	
	(2) Composite Endpoint		
(HBV DNA < 0.7 MEq/ml by bDNA assay and ALT < 1.25 × ULN)			
Number Meeting Composite Endpoint/Subjects Evaluable		77/134 (57)	6/135 (4)
Difference Estimate (ETV - LVD)		53.0	
97.5% CI		(42.7, 63.4)	
p-value		< 0.0001	

Entecavir was superior to lamivudine for the proportion of subjects who demonstrated improvement in hepatic fibrosis using Ishak score at Week 48. The proportions were 34% and 16%, respectively (NC = F method) (difference estimate [ETV-LVD] = 17.5; 95% CI 6.8, 28.2]; p = 0.0019). 11% in the entecavir group compared to 26% in the lamivudine group had a worsening Ishak fibrosis scores at Week 48.

The entecavir group showed reductions from baseline at Week 48 for the following hepatic endpoints:

- HBV cccDNA (mean change: -0.6 $\log_{10}$ copies/HGEq versus 0.0 $\log_{10}$ copies/HGEq for lamivudine p < 0.0001)
- Total hepatic HBV DNA (mean change: entecavir -1.7 $\log_{10}$ copies/HGEq; lamivudine – 0.2g_{10} copies/HGEq; p < 0.0001)

Table 20 summarises the results for the secondary endpoints.

Table 20: Summary of secondary endpoints at week 48 (NC=F)

Secondary endpoint	ETV (n=141)	LVD (n=145)	Difference estimates [ETV-LVD] (95%CI)
HBV DNA <0.7 MEQ/ML by bDNA assay	93 (66%)	8 (6%)	60.4 (51.8, 69.1) p<0.0001
Mean change by PCR ($\log_{10}$ copies/ml)*	- 5.11	-0.48	-4.4 (-4.8, -4.0) p<0.0001
HBV DNA <400 copies/ml	29 (21%)	2 (1%)	19.2 (12.3,26.1) p<0.0001
HBV DNA <300 copies/ml	27 (19%)	2 (1%)	17.8 (11, 24.5) p <0.0001
Loss of HBeAg	14 (10%)	5 (3%)	6.5 (0.7,12.2) p=0.0278
HBeAg seroconversion	11 (8%)	4 (3%)	5.0 (-0.1,10.2) p=0.06
ALT normalisation (< 1 xULN)	86 (61%)	22 (15%)	45.8 (35.9, 55.8) p <0.0001
Patient management endpoints			
Complete virological response	13 (9%)	1(1%)	8.5 (3.6, 13.5) p=0.0008
Partial virological response	80 (57%)	7 (5%)	
Virological non-response	40 (28%)	121(83%)	
Non-Completer	7 (5%)	10 (7%)	
Missing Week 48 data	8 (6%)	16 (11%)	
Complete response+ ALT normalisation	11 (8%)	1(1%)	7.1 (2.5,11.7) p=0.0027

* limit of quantitation 300 copies/ml

ETV was superior to LVD for all virological endpoints, which could be expected in a population refractory to the comparator.

Efficacy at week 96

Table 21: Response status through Year 2 – treated subjects

Virologic-Only responder continuing 2nd year of dosing	ETV n=77	LVD n=3
Complete responders at EOD**	9 (12%)	0
Virologic-Only Responder at EOD	57 (74%)	0
Non responder at EOD	11 (14%)	3 (100%)
Response –individual endpoints		
HBV DNA <0.7 MEq/ml	66 (86%)	-
HBV DNA <300 copies/ml at EOD	31 (40%)	-
Mean change from baseline (log$_{10}$ copies/ml) at Year 2	- 6.19	-
Loss of HBeAg	9 (12%)	-
HBeAg seroconversion	8 (10%)	
ALT normalisation <1.0 x ULN at EOD	81%	-
Cumulative responses through Year 2	**n=141**	**n=145**
Cumulative HBV DNA <300 copies/ml	42 (30%)	1 (<1%)
Mean change from baseline (log$_{10}$ copies/ml) at Year 2 EOD	-5.91	
Cumulative HBeAg loss at Year 2 EOD	26 (18%)	8 (6%)
Cumulative confirmed HBeAg seroconversion at EOD	22 (16%)	8 (6%)
Cumulative confirmed HBsAg seroconversion	1 (<1%)	
Cumulative ALT normalisation <1.0 x ULN at EOD	85%	29%

*Complete response: HBV DNA <0.7Meq/ml and loss of HBeAg. **EOD: End of Dosing

ETV-treated Virologic-Only Responders, who continued to Year 2 of dosing (n=77), had incremental benefit of therapy as demonstrated by:
- Subjects with HBV DNA < 300 copies/ml: from 21% (Year 1) to 40% (Year 2)
- Reduction in HBV DNA mean change: from -5.69 (Year 1) to -6.19 log$_{10}$ copies/ml (Year 2)
- ALT normalization: from 65% (Year 1) to 81% (Year 2)
- HBeAg loss: from 10% (Year 1) to 18% (Year 2)
- HBeAg seroconversion: from 8% (Year 1) to 16% (Year 2)
- Liver biopsy was only available on 10 ETV subjects at Week 96 and 9 of 10 demonstrated incremental histological improvement during the 2nd year of treatment.

In contrast, all Year 1 and 2 data suggest that continuing lamivudine in LVD refractory subjects provides minimal or no benefit.

A sustained response was evaluated only in 22 subjects who achieved a Response at Week 96 and completed 24 week follow-up off-treatment. Eleven (50%) of these subjects had a sustained response (HBV DNA <0.7 MEq/ml and HBeAg loss) at 24 weeks off treatment.

- Analysis performed across trials (pooled analyses and meta-analysis)

The applicant performed a number of analyses of the 48-weeks results from the phase III studies to better define the treatment response to entecavir.

The efficacy results were assessed using a combined response endpoint including histological, virological and biochemical response, in accordance to established guidelines. Two analyses (A and B) were therefore performed using different HBV DNA cut-offs for the definition of virological response:
- Analysis A: Responders achieve Histological Improvement *and* HBV DNA <10^5 copies/ml by PCR assay, *and* ALT ≤ 1.0 x ULN.
- Analysis B: Responders achieve Histological Improvement, *and* HBV DNA < 400 copies/ml by PCR assay, *and* ALT ≤ 1.0 x ULN.

©EMEA 2006

Table 22: Combination endpoints at week 48 (N=F)

	Treatment regimen		Difference estimate (95%CI), p-value
	ETV 0.5 mg	LVD 100 mg	ETV-LVD
Naïve HBeAg+ (022)	**N=314**	**N=314**	
Analysis A	169/314 (54)	122/314 (39)	15 (7.3, 22.7) 0.0002
Analysis B	138/314 (44)	0/314 (3 25)	18.5 (11.2, 25.8) <0.0001
Naïve HBeAg – (027)	**N=296**	**N=287**	
Analysis A	179/296 (60)	130/287 (45)	15.2 (7.2, 23.2) 0.0002
Analysis B	170/296 (57)	112/287 (39)	18.4 (10.4, 26.4) <0.0001
	ETV 1.0 mg	**LVD100mg**	
LVD refractory HBeAg+	**N=124**	**N=116**	
Analysis A	29/124 (23)	1/116 (1)	22.5 (14.9, 30.2) <0.0001
Analysis B	16/124 (13)	0/116 (0)	12.9 (7.0, 18.8) <0.0001

As shown in table 22, the analyses of combined endpoints at Week 48 consistently showed that ETV was superior to LVD, although lower response rates were seen compared to those for individual endpoints.

Additional analyses were conducted looking at the treatment response in patients that fulfil the current recommendation to treat, i.e. patients with an HBV DNA level $>10^5$ copies/ml, Knodell necroinflammatory score ≥ 5 at baseline and elevated ALT ≥ 2 x ULN versus treatment response in patients with milder disease. These analyses showed that Week 48 response rates were higher in patients currently considered prime candidates for antiviral treatment, i.e. those with moderate to severe disease. This was evident in both treatment groups and in particular among naïve HBeAg positive subjects. It was also demonstrated that HBeAg seroconversion occurred mainly among these patients (ETV 28%; LVD 25%) compared with those patients having low ALT (<2 x ULN) and histological scores (ETV 8%; LVD 6%).

The response rates with respect to histological improvement (55-57%), normalisation of ALT (65-86%) and reduction of HBV DNA (<400 copies/ml) (51-91%) in naïve ETV-treated subjects with milder disease could also be considered clinically relevant. In contrast, among ETV-treated LVD refractory patients with milder disease, histological improvement occurred at considerably lower rates, 35% (versus 72% in subgroup A) despite a similar 5-log decrease in HBV DNA. The fact that these patients were pre-treated and had already achieved some partial response on LVD therapy, might explain the lower histological response. Overall results suggest that patients with immunoactive disease achieved more benefit from ETV therapy, but the response in patients with lower ALT was also clinically meaningful.

An additional analysis was performed to look at prognostic factors among ETV-treated patients. This showed that in naïve HBeAg positive patients, baseline HAI score (>10), HBV DNA level ($<10^7$ copies/ml) and ALT level (>5 x ULN) were significant predictors of response at Week 48. These results are in agreement with those previously shown for interferon, lamivudine and adefovir. Subjects with ALT >5 x ULN also had the highest rate of HBeAg seroconversion (39%). The most consistent association was that between high baseline HAI scores and response rates of histological improvement, which came out as statistically significant in HBeAg positive and HBeAg negative subjects and borderline in LVD refractory. In ETV patients with very high HBV DNA levels satisfactory histological and biochemical responses were noted at week 48. Other factors such as race, HBV subtype and prior IFN therapy did not seem to affect antiviral efficacy of ETV.

Further analyses showed a clear correlation between grade of virological suppression and histological improvement. However, this did not translate to a higher rate of HBeAg seroconversion. No consistent relationship between changes in viraemia and seroconversion was shown. It was evident that HBeAg seroconversions clustered in the subgroup that achieved an HBV DNA <400 copies/ml. These data are based on a limited number of patients and do not allow any definitive conclusions to be drawn.

- Clinical studies in special populations

The efficacy of entecavir was evaluated in patients co-infected with HIV (study AI463038), post-liver transplant patients (study AI463015) and patients with decompensated liver disease (study AI463048). An overview of these studies is presented in table 23.

Table 23: Studies in special populations

Type of study/ criteria for inclusion	Treatment	Duration	N of patients	Primary efficacy endpoint	Location	Study design
Study AI463038						
Chronic LVD-resistant HBV co-infected with HIV Stable HAART HIV RNA<400 copies/ml HBVDNA >10^5 $\log_{10}$ copies/ml HBeAg + or -	ETV 1.0 mg QD Placebo Continued LVD (300mg) – containing HAART	24 week blinded treatment +24 week open-label treatment +24 week follow-up	**N=68** ETV n=51 Placebo n=17	Mean HBV DNA $\log_{10}$ copies/ml at week 24 (PCR assay)	Europe (35% of patients), USA (15% of patients, South America (50% of patients)	Phase II Randomised double-blind, placebo-controlled
Study AI463015						
Recurrent hep B, despite anti-HBV prophylaxis, clinical stable, >100 days post-transplant HBV DNA >10pg/ml	ETV 1.0 mg QD Optionally HBIG	Up to 104 weeks of open-label ETV	**N=9**	Safety and PK Secondary: n (%) <0.7MEq/ml HBV DNA at Week 24 at Week 48	Europe, USA, Australia	Pilot study Open-label Multicentre
Study AI463048						
Patients with chronic hepatitis B and hepatic decompensation (defined by a Child Pugh $\geq$ 7	ETV 1.0 mg QD ADV 10 mg QD	Up to 96 weeks		Mean change in serum HBV DNA $\log_{10}$ copies/ml at week 24		Randomised Open-label Multicentre

Study AI463038

Patients (n = 68) continued their lamivudine-regimen and were assigned to add either entecavir 1 mg once daily (n = 51) or placebo (n = 17) for 24 weeks followed by an open-label phase for an additional 24 weeks where all received entecavir. The majority of the treated subjects were male (96%), white (85%) and the mean age was 41 years.
The distribution of baseline HIV/HBV disease characteristics was balanced between the ETV and placebo groups, including mean HBV DNA levels (9.13 and 9.12 $\log_{10}$ copies/ml, respectively) and frequency of positive HBeAg (98% and 100%, respectively). Patients had stable controlled HIV (HIV RNA < 400 copies/ml) with recurrence of HBV viraemia on a lamivudine-containing HAART regimen. At baseline entecavir-treated patients had a median duration of prior lamivudine therapy of 4.8 years and median CD4 count of 494 cells/mm^3 (range: 137-1,388 cells/mm^3) with only 5 subjects having CD4 count <200; 90% of patients had LVDr substitutions by genotype.
The mean number of weeks of exposure to double-blind study drug was comparable between the ETV and placebo groups (24.0 and 24.5 weeks, respectively). Three ETV-treated subjects (6%) discontinued study therapy prior to Week 24.

RESULTS

As shown in table 24, a 24-week treatment course with entecavir 1 mg resulted in statistically significant reduction in HBV DNA compared to placebo.

Table 24: Summary of efficacy endpoints at week 24

Efficacy endpoint	ETV N=51	Placebo N=17	Difference estimates (ETV-placebo)
Primary endpoint			
Mean HBV DNA at week 24 ($\log_{10}$ copies/ml)	5.52	9.27	
Secondary endpoints			
Mean HBV DNA change by PCR	- 3.66	+0.11	-3.76 (95%CI -4.49, -3.04); p < 0.0001).
HBV DNA <400 copies/ml	3 (6%)	0	p=0.31
Loss of HBeAg	1 (2%)	0	p=0.25
HBeAg seroconversion	1 (2%)	0	p=0.56
ALT normalisation (<1.25xULN)	11/30 (37%)	1/7 (14%)	p=025
ALT normalisation ($\leq$1.0xULN)	12/35 (34%)	1/12 (8%)	

During the open-label phase a further reduction of HBV DNA level occurred from -3.66 $\log_{10}$ copies/ml (Week 24) to -4.20 $\log_{10}$ copies/ml (Week 48). This compared to HBV mono-infected LVD refractory patients who achieved a mean change of -5.41 $\log_{10}$ copies/ml at week 48.

Study A463015

Of the 9 treated subjects, 8 were male, and the mean age was 53 years. Five subjects were positive for HBeAg. The median baseline HBV DNA level by PCR assay was 8.58 $\log_{10}$ copies/ml. Laboratory abnormalities at baseline included increased ALT values (n=6), increased serum amylase/lipase (n=6), Grade 1 BUN/urea elevations (n=6) and Grade 1 elevated creatinine levels (n=4). All subjects received at least 1 HBV medication prior to study entry [LVD (9 subjects), famciclovir (4 subjects), and ganciclovir (4 subjects)]. One subject had received prior HBIG, but did not continue this while on-treatment with ETV. All subjects have completed 48 weeks of open-label dosing. The mean time on study therapy for all treated subjects was 131 weeks (range: 92.4 to 164.0 weeks). In this study, liver biopsy was optional.

Efficacy observations are limited to a 9 subjects but are encouraging (table 25).

Table 25: Summary of efficacy endpoints at week 24 and 48

Efficacy endpoints	Entecavir (n=9)
Week 24	
Mean change by PCR ($\log_{10}$ copies/ml)	- 3.62
N with $\geq$ 2 $\log_{10}$ reduction from baseline	8/9
Week 48	- 3.90
Mean change by PCR ($\log_{10}$ copies/ml)	
N with $\geq$ 2 $\log_{10}$ reduction from baseline	9/9
Maintained response through week 48	
N with $\geq$ 2 $\log_{10}$ reduction from baseline	8/9
Proportion with <0.7 MEq/ml	5/8
HBeAg Seroconversion	2
ALT normalisation (to <1.25xULN) or improvement	6/9
Histological improvement	3/4
Resistance development during 3-year FU	**4/7**

Study AI463048

This study in decompensated patients is ongoing and an exploratory analysis of efficacy data has been performed. Of the 65 patients (34 ETV; 31 ADV) from the efficacy cohort in the interim database, a total of 36 (22 ETV; 14 ADV) had virus containing LVDr substitutions on entry. At Week 24, 50% of ETV-treated patients (11/22) in this subset had an HBV DNA <300 copies/ml, as compared to only 21% (3/14) of ADV-treated patients. In this subset analysis, the mean and median changes in HBV DNA through Week 24 were -4.39 and -4.22 $\log_{10}$ copies/ml, respectively, for ETV; results were -3.5 and -3.42 $\log_{10}$ copies/ml, respectively, for ADV. Although providing early data in a small cohort of patients, the high proportion of patients achieving an undetectable HBV DNA with ETV treatment in this population is encouraging.

- **Supportive studies**

AI463056 LVD refractory Chinese patients

This is a phase II double-blind randomised (4:1) placebo-controlled study in LVD refractory Chinese patients. The study comprised 3 phases: 12-week double-blind dosing phase, a 36-week open-label dosing phase, and a 24-week post-dosing phase. After completing the 12 weeks double-blind phase, subjects received 1.0 mg ETV for up to 36 weeks in the open-label dosing phase of the study. Subsequently subjects could continue open-label ETV treatment by enrolling in the ETV roll-over study AI463050. Subjects who prematurely discontinued study treatment or who did not in the rollover study entered 24 week post-dosing-follow-up. The mean time on open-label ETV was 40.1 weeks.

Most of the subjects were male (75%) and the mean age was 35 years. The majority was HBeAg positive (129/145; 85%) with a mean HBV DNA level 8.77 $\log_{10}$ copies/ml. The proportion of subjects with ALT >1.25xULN was 43%.
Results are summarised in table 26.

Table 26: Summary of primary and secondary endpoints at week 12 and at Week 48

Endpoint	Entecavir 1.0 mg N=118	Placebo N=29	Difference estimates [ETV-placebo] (95%CI)
Mean change by PCR ($\log_{10}$ copies/ml)	- 4.30	-0.15	-4.06 (-4.53, -3.6) p<0.0001
Secondary endpoints			
HBV DNA <0.7 MEq/ml by bDNA assay	85/115 (74%)	3/29 (10%)	63.6 (43.7, 83.4) p<0.0001
HBV DNA <400 copies/ml	9/116 (8%)	0/29	7.8 (2.1,17.6) p=0.12
Loss of HBeAg	7/106 (7%)	2/25 (8%)	-1.4 (-12.4, 9.6) p=0.80
HBeAg Seroconversion	7/106 (7%)	2/25 (8%)	-1.4 (-12.4, 9.6) p=0.80
ALT normalisation	32/45 (71%)	1/15 (7%)	64.4 (35.4, 93.5) p<0.0001
36 week open-label ETV treatment phase			
	n=116	**N=29**	
Mean change by PCR ($\log_{10}$ copies/ml)	-5.08	-4.86	
Secondary endpoints			
HBV DNA <0.7 MEq/ml by bDNA assay	85 (74%)	22 (70%)	
HBV DNA <400 copies/ml	33 (28%)	13 (45%)	
Loss of HBeAg	8/106 (8%)	4/23 (17%)	
HBeAg Seroconversion	6/106 (6%)	2/23 (9%)	
ALT normalisation <1.25 x ULN	41/45 (91%)	16/17 (94%)	

Higher proportion of subjects achieved HBV DNA <400 copies/ml who had the following characteristics at baseline: HBeAg negative, ALT levels >2.6xULN and received prior interferon. About two-thirds of the subjects with HBeAg seroconversion had elevated ALT levels at baseline.

Virological rebound occurred in 15 subjects on ETV treatment but there was no evidence that these were due to emergence of resistance. Few (< 1 %) subjects experienced ALT flares on treatment. Prolonged ETV treatment up to 48 weeks in Chinese LVD-refractory patients provided incremental benefits with respect to virological and biochemical endpoints. The results are in line with those achieved in the pivotal phase III study AI423026, although virological responses were somewhat lower. This might reflect differences in baseline HBV characteristics with a higher proportion of subjects (67%) having normal ALT levels in the Chinese study. The HBeAg seroconversion rate was very low (6%). As expected the majority of subjects with seroconversion had elevated baseline ALT levels.

Study AI463023 in China - nucleoside naïve HBeAg+/HBeAg- patients

This is an ongoing randomised (1:1) double-blind phase III study in China comparing the safety and efficacy of ETV 0.5 mg QD to LVD 100 mg QD administered for up to 96 weeks to subjects with compensated chronic hepatitis B. Randomisation was stratified by HBeAg status at baseline and

investigative site. Subjects were to have elevated ALT levels >1.3xULN. The primary efficacy endpoint was the proportion of subjects in each treatment group who achieved the composite endpoint (HBV DNA <0.7 MEq/ml by bDNA assay *and* ALT<1.25xULN) at Week 48. Histological improvement was not an endpoint in this study.

Of 525 subjects randomised in the study, 519 started treatment (ETV 258; LVD 261) and 499 (ETV 251, LVD 248) completed the first year of dosing. Most subjects were male (82%) with a mean age of 30 years. HBV disease characteristics were comparable between treatment groups. Most subjects were HBeAg positive (ETV 87%, LVD 85%). Mean baseline HBV DNA was 8.64 $\log_{10}$ copies/ml in the ETV group and 8.48 $\log_{10}$ copies/ml in the LVD group.
Efficacy results are shown in table 27.

Table 27: Efficacy results at Week 48 (NC=F)

Endpoint	ETV 0.5 mg N=258	LVD100mg N=261	Difference estimates [ETV-LVD] (95%CI)
Composite endpoint*	231 (90%)	174 (67%)	23.1 (16.3, 29.9) p<0.0001
Secondary endpoints			
Mean change in HBV DNA	-5.90	-4.33	-1.50 (-1.77, -1.2) p<0.0001
HBV DNA <0.7 MEq/ml	244 (95%)	188 (72%)	22.8 (16.7, 28.9) p<0.0001
HBV DNA <400 copies/ml	202 (78%)	116 (44%)	34.6 (27.0, 42.2) p<0.0001
Loss of HBeAg	41/225 (18%)	44/221 (20%)	-1.7 (-9.0, 5.6) p=0.65
HBeAg Seroconversion	33/225 (15%)	39/221 (18%)	-3.0 (-9.8, 3.8) p=0.39
ALT normalisation <1.25xULN	238 (92%)	214 (82%)	10.3 (4.6, 16.0) p=0.0005
Patient managed endpoints			
Consolidated response**	50 (19%)	40 (15%)	
Partial virological response	193 (75%)	146 (56%)	
Virological non-responder	8 (3%)	62 (24%)	
Did not complete 1st year of dosing	7 (3%)	13 (5%)	

* Composite endpoint: HBV DNA <0.7 MEq/ml *and* ALT<1.25xULN. Consolidated response: HBV DNA <0.7 MEq/ml, *and* HBeAg negative from Week 24 to Week 48

The efficacy results achieved in this study are comparable to those achieved in the study 022. ETV 0.5 mg was superior to LVD 100 mg for all efficacy endpoints with one exception, HBeAg loss/seroconversion, for which ETV was non-inferior to LVD. In this study seroconversion rates were higher than in the previous AI463056, probably due to the fact that elevated ALT was required for enrolment.

Study AI460312 was a phase II conducted in 212 patients in China nucleoside naïve HbeAg positive patients.
Of 204 patients treated with open-label ETV, 84 (41%) had normal baseline ALT prior to treatment. An exploratory analysis of response at the end of 48 weeks of open-label treatment by ALT status in suggests that patients with normal ALT levels treated with ETV have lower response rate than patients in elevated ALT levels.

Efficacy Endpoints by Baseline ALT status

Endpoint at Week 48	Baseline ALT	
	ALT < 1.25 x ULN N=84	ALT ≥ 1.25 x ULN N=120
HBV DNA < 0.7 MEq/ml by bDNA: n (%)	59 (70)	102 (85)
HBV DNA < 400 c/ml by PCR: n (%)	13 (15)	73 (61)
HBeAg seroconversion: n (%)	2 (2)	7 (7)

In this study patients stopped open label ETV after 48 weeks of dosing regardless of their responses to therapy. The observed rates of off-treatment ALT flares (7-8 %) were higher than those noted in study 022. This suggests that if treatment is discontinued without regard to treatment response the rate of post-treatment ALT flare could be higher.

<u>Study AI463901 rollover study</u>

AI463901 is a multinational and open-label study evaluating the safety and antiviral activity of ETV in combination with LVD. Later the protocol was amended to eliminate lamivudine. Subjects enrolled must have participated in a previous ETV study and must have discontinued their study medication from the previous study before beginning therapy offered in this study. All treated patients (859) received combination therapy (ETV 1.0 mg + LVD 100 mg QD) except for 5 subjects that received ETV monotherapy. The majority of treated subjects were male (77%) and the mean age was 41 years. The largest geographic cohort of subjects was from Asia (41%), followed by Europe (34%), North America (13%), and South America (12%). For treated subjects with entry data available, the mean serum HBV DNA was 6.35 $\log_{10}$ copies/ml and 66% of subjects tested positive for HBeAg. Approximately half of the subjects (49%) had normal ALT values at entry (< 1.25 x ULN).

Preliminary results from this complex rollover study provide evidence that continued antiviral therapy beyond 2 years provided incremental benefit in the HBeAg positive nucleoside-naïve patients with partial response to ETV, since virological suppression was maintained and seroconversion occurred in 9% of the subjects (at Week 24). Most of the HBeAg negative patients had relapses after discontinuing study therapy after 1 year and re-treatment with ETV 1.0 mg resulted in restoration of virological suppression.

Data suggest that the combination of ETV and LVD did not provide any additional benefit over ETV monotherapy in any of the populations studied, although this study was open and therefore exclude any formal comparisons to be made.

Clinical safety

- Patient exposure

The overall estimated mean exposure to entecavir was 60 weeks for the 0.5 mg dose and 56 weeks for the 1.0 mg dose. There were 330 patients who received ETV for more than 1.5 years and 3 patients for more than 3 years. The total population exposed to ETV in randomised trials included 1392 subjects. A further 899 subjects received the active comparator and 108 received placebo for a short time period after which time 105 of the placebo-treated patients received open-label ETV.

Study AI463022 included safety information for 709 treated subjects (ETV 354; LVD 355) through the end of dosing up to 96 weeks. During this period, the mean cumulative exposure to study therapy was 80.8 weeks in the ETV treatment group and 67.7 weeks in the LVD treatment group. Study AI463027 included safety data for 638 treated subjects (ETV 325; LVD 313) and the mean cumulative exposure to study therapy was 56.1 weeks in the ETV group and 56.8 weeks in the LVD group. Study AI463026 included data for 286 treated subjects (ETV141; LVD145) and the mean exposure to study therapy was 76.7 weeks in the ETV group and 52.6 weeks in the LVD group. The mean time on therapy in the rollover study AI463901, including 882 patients, was 37.8 weeks for subjects previously treated with ETV and 37.7 weeks for subjects previously treated with LVD. The maximum time on therapy was 184 weeks.

Safety analyses from the second year of treatment (Week 96) were provided as supplementary information during the procedure.

Overall, the safety population included mostly males, and females represented around 25% of the study population. The majority of patients were in their middle age and only 34 subjects ≥ 65 years were treated with ETV. All study subjects had compensated hepatitis B and there were few patients with baseline liver cirrhosis (n=58).

Four potential safety issues were analysed in all studies:

- hepatitis flares during and after treatment based on experience of approved therapies in the management of chronic hepatitis B
- lactic acidosis, a class-related concern of the nucleoside/nucleotide analogues

- CNS events from the observation of perivascular inflammation in the CNS of dogs reversible upon cessation of dosing which may have been species-dependent.
- neoplasms based on an increased incidence of benign and malignant neoplasms involving a variety of organ sites in carcinogenicity studies conducted in rodents

- Adverse events

The studies in healthy volunteers included 538 subjects and were heterogeneous regarding study designs, populations and ETV exposure (0.05 mg to 40 mg). Only 39 patients received placebo rendering it difficult to draw firm conclusions from these studies. No clear safety signal was observed. Nevertheless, the data indicated that AEs increased with higher doses of ETV and headache and gastrointestinal symptoms were noted more frequently during ETV administration than in the placebo/other drug group. Among laboratory abnormalities elevated ALT occurred more commonly in the ETV group vs. placebo /other drug (11% vs. 5%). From those patients who had ECG measurements pre and post treatment, there was small number of patients with an increase in QT-interval on treatment (3% versus 1%). Nonetheless supplementary analysis did not show any significant difference in QT intervals between subjects treated with ETV or with placebo.

Among the 2399 treated subjects (those who received at least one dose of study medication) in the Phase 2 and 3 clinical studies, 72% of subjects in the ETV group and 81% of subjects in the LVD group experienced a minimum of 1 adverse event. With regard to the pivotal studies in the nucleoside-naive and LVD-refractory populations, the frequency and profile of AEs were comparable between both populations overall. Furthermore, there was no apparent dose-response relationship in the overall incidence of adverse events for entecavir across the 10-fold dose range in study comparing 0.1 mg, 0.5 mg and 1mg dose (study AI463014).

The most commonly reported adverse events during the first 48 weeks of treatment were headache, upper respiratory infection, cough, upper abdominal pain and fatigue, all of which occurred with comparable frequencies in the ETV vs. LVD treatment groups.
The proportion of subjects with AEs that were attributed by the investigator to study compound (related, all Grades) was comparable between the 2 populations and was comparable in ETV- and LVD-treated subjects (ETV 35%; LVD 37%). The most common related AEs for ETV-treated subjects were headache, fatigue, dizziness, and nausea. Most AEs were mild to moderate in severity (Grade 1-2). In the nucleoside-naïve population, the most frequent Grade 3-4 AEs reported in ETV-treated patients were increased ALT (ETV 2%; LVD 4%) and increased lipase (ETV 2%; LVD 1%). In the LVD-refractory population, the most frequent Grade 3-4 AEs reported in the ETV-treated group were increased lipase (ETV 2%, LVD 2%) and hyperglycemia (ETV 2%, LVD 0).

Table 28: Summary of the treatment related AE reported in at least 1% of ETV-treated subjects:

	Number of Subjects (%)						
	Nucleoside-Naive Subjects		**LVD-Refractory subjects**		**Safety cohort**		
	ETV 0.5 mg N= 679	LVD 100mg N= 668	ETV 1 mg N= 183	LVD 100mg N= 190	ETV N= 1392	LVD 100mg N= 899	Placebo N=108
Any Adverse events	248 (37)	251(38)	83 (45)	71 (37)	489 (35)	335 (37)	21 (19)
Gastrointestinal Disorders	**84(12)**	**69 (10)**	**27 (15)**	**23 (12)**	**158 (11)**	**96 (11)**	**3 (3)**
- Nausea	20 (3)	14 (2)	8 (4)	6 (3)	45 (3)	22 (2)	0
- Abdominal Pain Upper	18 (3)	13 (2)	3 (2)	9 (5)	27 (2)	23 (3)	0
- Abdominal Pain	11 (2)	11 (2)					
- Dyspepsia	11 (2)	13 (2)	5 (3)	1 (<1)	21 (2)	14 (2)	0
- Abdominal Discomfort	10 (1)	5(<1)	2 (1)	1 (<1)	15 (1)	6 (<1)	1 (<1)
- Diarrhoea	7 (1)	5 (<1)	4 (2)	2 (1)	17 (1)	7 (<1)	0
- Vomiting	7 (1)	4 (<1)	2 (1)	1 (<1)			
- Flatulence			3 (2)	1 (<1)			
General Disorders and Administration site Conditions	**50 (7)**	**47 (7)**	**21 (11)**	**17 (9)**	**134 (10)**	**72 (8)**	**8 (7)**
- Fatigue	31 (5)	32 (5)	17 (9)	11 (6)	86 (6)	47 (5)	7 (6)
- Asthenia	8 (1)	11 (2)	2 (1)	4 (2)	19 (1)	18 (2)	1 (<1)
Infections and Infestations	**17 (3)**	**17 (3)**					
- Influenza	7 (1)	1 (<1)					
- Rhinitis	7 (1)	1 (<1)					
Investigations							
- Blood Amylase Increased	**64 (9)**	**86 (13)**	**27 (15)**	**25 (13)**	**108 (8)**	**112 (12)**	**0**
- Lipase Increased	23 (3)	19 (3)	3 (2)	2 (1)	29 (2)	22 (2)	0
- ALAT Increased	20 (3)	20 (3)	7 (4)	3 (2)	32 (2)	24 (3)	0
- ASAT Increased	11 (2)	31 (5)	6 (3)	15 (8)	26 (2)	46 (5)	0
- Blood Bicarbonate decreased			6 (3)	8 (4)	18 (1)	22 (2)	0
- Blood Biliburin Increased			4 (2)	1 (<1)			
- Prothrombin Time Prolonged			4 (2)	1 (<1)			
- Platelet Count Decreased			3 (2)	1 (< 1)			
- Weight Decreased			2 (1)	0			
			2 (1)	0			
Metabolism and nutrition disorders			**7 (4)**	**4 (2)**			
- Hyperamylasaemia			2 (1)	0			
Musculoskeletal and Connective Tissue Disorders	**28 (4)**	**26 (4)**	**8 (4)**	**8 (4)**	**60 (4)**	**37 (4)**	**2 (2)**
- Arthralgia	10 (1)	9 (1)	2 (1)	4 (2)	20 (1)	14 (2)	1 (<1)
- Myalgia	9 (1)	7 (1)					
- Back Pain	8 (1)	4 (< 1)					
- Mucle Cramp			4 (2)	1 (<1)			
Nervous System Disorders	**88 (13)**	**83 (12)**	**27 (15)**	**23 (12)**	**185 (13)**	**111 (12)**	**8 (7)**
- Headache	58 (9)	51 (8)	18 (10)	14 (7)	119 (9)	69 (8)	1 (<1)
- Dizziness	25 (4)	21 (3)	9 (5)	3 (2)	46 (3)	25 (3)	1 (<1)
- Somnolence	8 (1)	9 (1)	3 (2)	2 (1)	24 (2)	11 (1)	4 (4)
Psychiatric Disorders	**25 (4)**	**26 (4)**	**6 (3)**	**2 (1)**	**47 (3)**	**29 (3)**	**4 (4)**
- Insomnia	12 (2)	10 (1)	2 (1)	1 (<1)	23 (2)	11 (1)	4 (4)
Renal and Urinary Disorders			**5 (3)**	**5 (3)**			
- Haematuria			2 (1)	1 (<1)			
Respiratory, Thoracic and Mediastinal Disorders			**6 (3)**	**2 (1)**			
- Cough			2 (1)	1 (<1)			
Skin and Subutaneous Tissue Disorders	**40 (6)**	**36 (5)**	**9 (5)**	**8 (4)**	**75 (5)**	**45 (5)**	**2 (2)**
- Pruritus	11 (2)	10 (1)	4 (2)	0	20 (1)	10 (1)	0
- Rash			3 (2)	4 (2)			
- Alopecia			2 (1)	0			

During off-treatment follow-up, adverse events (all grades) were reported less frequently for subjects receiving ETV 0.5 mg QD compared with subjects receiving LVD (ETV 42%; LVD 51%) in the nucleoside-naïve population. The incidence of AEs was comparable between the ETV (46%) and LVD

(45%) treatment groups in the LVD-refractory population. With the exception of increased ALT and increased AST, all other related AEs were reported with a frequency < 1% for either treatment.

- Serious adverse event/deaths/other significant events

A total of 15 deaths (ETV n=9) were reported in the total safety cohort of 2399 patients, for an overall incidence of 0.6%. The overall incidence of serious adverse events was low at 5% in the ETV group versus 8% in the LVD group. The incidence of SAEs was comparable between nucleoside-naïve (7%) and LVD refractory (10%) patients. There were few SAEs (<1%) that were considered related to ETV. No particular pattern was observed among the events that were considered related to ETV. For LVD more treatment-related SAEs were reported (in particular in nucleoside-naïve patients), and most of these consisted of hepatic abnormalities.

No new safety signal occurred in the second year of ETV therapy and the safety profile was comparable to lamivudine in nucleoside naïve patients.

Safety issues of special interest

- **Neoplasms**

The key safety concern with ETV relates to its possible carcinogenic potential based on the findings in the rodent carcinogenicity studies.

Overall, neoplasms occurred in similar frequencies in both treatment groups, ETV n=17/1497 (1.1%) versus LVD n=9/899 (1.0%). The incidence rates were comparable in both groups 8.5 vs. 7.8 /1000 patient years of observation, respectively. The most common malignancy observed was hepatocellular carcinoma (HCC, n=11) (ETV n=7 vs. LVD n=4) with incidence rates of 3.5 vs. 3.4/1000 patient years of observation, respectively. HCC could be expected in a population with long-standing chronic hepatitis B and the majority of these cases had evidence of liver cirrhosis. The rate of HCC found in the ETV studies is consistent with published data. The only additional malignancies reported in more than 1 subject in either treatment group were prostate cancer (n = 2 [ETV]), breast cancer (n = 2 [LVD]) and basal cell carcinoma of the skin (n = 2 [ETV]). Among the non-HCC neoplasms reported, there was 1 recurrence of a gastric cancer and 1 metastatic cancer both occurring in LVD-treated patients. Other neoplasms reported were "de novo" cancer. There was no particular tumour type of non-HCC with elevated frequency, although 4 cases with skin cancer were identified in the ETV group versus 1 in the LVD group. Most of the patients presented with risk factors for cancer. The median age was 64 years in both populations and the median duration of drug exposure prior to diagnosis was 202 days (range: 56-779) in ETV-treated patients and 325 days (range: 277-357) in LVD-treated patients.

Two epidemiological studies (the US cohort and the Taiwan cohort) commissioned by the applicant, concluded that the incidence rate of all cancer and of liver cancer was significantly increased among HBV subjects compared to subjects not infected with HBV. In the retrospective US cohort, the overall incidence rate of malignancy was 9.70 per 1000 person-years (95% CI: 8.25-11.31). This represents a relative risk of 2.59 (95% CI: 2.19-3.03) compared to non-infected subjects. The incidence rate of liver cancer was 4.95/1000 person-years (95% CI: 3.95-6.11). This represents a relative risk of 220.52 (95% CI: 135.63-379.07). In the prospective Taiwan cohort, the overall malignancy incidence rate in the HBsAg positive cohort was 6.53/1000 person-years (95% CI: 5.79-7.34). This represents a relative risk of 1.7 (95% CI: 1.4-1.9) compared with the general population. The HCC incidence rate was 3.60/1000 person-years (95% CI: 3.06-4.21), which represents a relative risk of 9.1 compared with non-HBsAg-positive patients.

In these epidemiological cohorts, subjects were followed for 8 years or more in the US study and for up to 11 years in the Taiwan study. The observation time is not comparable between the ETV Safety Cohort and the epidemiological studies precluding any comparison of the malignant neoplasm rates in these populations.

A total of 28 malignant neoplasms have been reported in the updated Safety Cohort as of December 2004 (Table 29). The rates of overall malignant neoplasms were comparable across the ETV versus the LVD treatment groups, although numerically higher in the ETV group. When assessed by malignancy subcategory, rates remained comparable regardless whether calculated as simple event rates or as incidence rates per 1000 PY of observation. In addition, unintegrated data were available from ongoing studies AI463-038 -048 and -901 and further 15 malignant neoplasms were reported in these studies. The malignancy types observed across the 15 malignancies reported were consistent with those reported in the Safety Cohort and included: 1 case from AI463038 (co-infected HIV/HBV): testicular cancer (ETV), 6 cases from AI463048 (decompensated patients) with HCC (ETV 3; ADV 3), 5 cases from AI463049 (long-term observational): 3 HCC (ETV 2, LVD 1), 1 small cell carcinoma of lung (ETV), and 1 basal cell carcinoma (ETV); and 3 cases from AI463901 (roll-over study with combination therapy ETV+LVD): 1 each of gastric cancer, cutaneous T cell lymphoma, and multiple myeloma.

Table 29: Rates of individuals with malignant neoplasms in ETV studies (Safety Cohort)

	ETV n=1497	LVD n=899
All neoplasms	19 (1.3%)	9 (1.0%)
	8.8/1000 PY	7.4/1000 PY
HCC	7 (0.5%)	4 (0.4%)
	3.2/1000 PY	3.3/1000 PY
Non-skin	15 (1.0%)	8 (0.9%)
	6.9/1000 PY	6.6/1000 PY
Non-HCC	12 (0.8%)	5 (0.6%)
	5.5/1000 PY	4.1/100PY
Non-HCC/Non-skin	8 (0.5%)	4 (0.4%)
	3.7/1000 PY	3.3/1000 PY

The mean time to the diagnosis of neoplasm and HCC in the updated Safety Cohort is presented in Table 30. Overall, the mean drug exposure to time of diagnosis was larger for patients treated with ETV.

Table 30: Neoplasms in ETV studies (Safety Cohort): Exposure and observational times

	ETV n=1497	LVD n=899
All malignant neoplasms	19	9
Mean time to diagnosis of neoplasm	46 weeks	36 weeks
Mean drug exposure to time of diagnosis	43 weeks	35 weeks
HCC	7	4
Mean time to diagnosis of neoplasm	37 weeks	25 weeks
Mean drug exposure to time of diagnosis	33 weeks	23 weeks

Altogether, the data demonstrate that there is no early safety signal for an increased rate of human cancer as a result of treatment with ETV. However, the observational period is too limited to exclude a carcinogenicity risk and close monitoring in long-term follow-up studies is required.

- **Neurological adverse events:**

In a 3-month oral toxicity study of ETV in dogs, minimal to moderate perivascular inflammation in the CNS was observed. The CNS inflammation was not observed in other species used in toxicological studies and was reversible upon cessation of dosing. In the Phase 1 studies, headache was reported more frequently in ETV-treated subjects compared with placebo-treated subjects. In order to ensure that any clinical safety signal in humans for CNS inflammatory events was appropriately recognised, the ETV integrated Phase 2/3 database was searched for adverse events which could represent clinical manifestations of CNS vasculitis (e.g. meningitis, somnolence).

In both the nucleoside-naïve and LVD-refractory study populations, the incidence of neurological events was comparable between ETV- and LVD-treated subjects (26% vs. 28%) No events suggestive of CNS vasculitis were reported as of the data cutoff for this analysis (May 28, 2004). Headache was the most frequently reported neurological adverse event (ETV 18%; LVD 19%).

©EMEA 2006

A re-analysis of this potential safety concern regarding neurological events taking into account the cases of eye disorders (photophobia, reduced visual acuity, blurred vision) was performed and no potential association with ETV treatment was found.

- **ALT flares**

Liver was a target organ in toxicology studies. The incidence and grades of ALT/AST elevations were comparable between the ETV groups and the LVD group. There was no evidence of drug-related increases in association with the higher 1.0 mg dose.

Overall, ALT flares were observed less frequently in ETV-treated subjects compared with LVD-treated subjects in both the nucleoside-naive and LVD-refractory populations.

- On-treatment ALT flares: In nucleoside-naïve population, ALT flares were reported in 15 (2%) ETV-treated subjects versus 28 (4%) LVD-treated subjects. In the LVD-refractory population, ALT flares were reported in 4 (2%) ETV-treated subjects versus 21 (11%) LVD-treated subjects. The majority of on-treatment ALT flares in the ETV group were associated with decreasing HBV DNA, and consistent with a manifestation of immune-mediated activity, and generally resolved while continuing ETV. The majority of flares observed in the LVD group on-treatment, was associated with persistently elevated HBV DNA or a rise in HBV DNA levels.

- Off-treatment ALT flares: In the ETV group ALT flares were uncommon and not associated with signs or symptoms of hepatic impairment. No ETV-treated subjects with ALT flare experienced hepatic decompensation. In nucleoside-naïve population, fewer ETV-treated subjects compared with LVD-treated subjects had ALT flares (ETV 4%; LVD 8%) during the follow-up period. Of the 16 ETV-treated subjects with flares, 14 were baseline HBeAg negative (AI463-027)). In LVD-refractory population, 3/56 (5%) ETV-treated subjects had an ALT flare compared with no subject treated with LVD during the follow-up period. No subject had hepatic decompensation, by either clinical or laboratory parameters, associated with their flare.

Although ALT flares were observed in low frequencies in the ETV group and rarely associated with hepatic SAEs (1%), flares could still represent a significant safety risk for patients with decompensated liver disease (see below).

Additional information was provided on ALT flares with regard to time of onset. The median time to on-treatment ALT flares differed between treatments, with flares occurring early in the treatment with ETV (after 4-5 weeks) and late with LVD (after $\geq$25 weeks). The median time to off-treatment ALT flares was longer for ETV (17-24 weeks) than for LVD (9-12 weeks). Off-treatment flares were more common among HBeAg negative patients (ETV 8%; LVD 11%) than among HBeAg positive patients (ETV <3%; LVD 7%) in both treatment groups. In the ETV-treated group and among naïve patients, 92% (23/25) of ALT flares occurred in the HBeAg negative population.

On the basis of these data, periodic monitoring of hepatic function is recommended during treatment.

In China study 023, the proportion of subjects with ALT flares were comparable to those in the global Phase III study. In the China study 012, the observed rates of off-treatment flares were however higher than those noted in study 022 (7-8 % versus 1%). The median time to ALT flare was 24 weeks which was comparable to the analyses from phase III. Therefore the SPC includes the warning that if ETV is discontinued without regard to treatment response, the rate of post-treatment flares could be higher.

- **Lactic acidosis**

A safety issue related to the nucleos(t)ide analogues class is mitochondrial toxicity. ETV does not inhibit mitochondrial γ-polymerase, and therefore lactic acidosis was not expected to be an issue for this compound. However, 22 cases (ETV n=11) in the safety cohort were reviewed for lactic acidosis syndrome, of whom 17 were found not to fulfil criteria, whereas 5 remained undetermined due to insufficient data. There has been 1 case diagnosed with lactic acidosis syndrome (LAS) in an ongoing ETV trial, but supplementary data did not suggest that the elevation of lactate in this case was related to ETV treatment. Therefore since further data submitted support ETV´s lack of mitochondrial toxicity and so far no additional cases of lactic acidosis have been reported during the ETV clinical trial

program, no additional statement has been added in the SPC over the class labelling warning on lactic acidosis with nucleoside analogues.

- **Hypocarbia**

Hypocarbia was the most commonly observed electrolyte abnormality occurring at comparable rates for ETV and LVD, and at comparable rates across the two safety populations: nucleoside-naive: ETV 26%, LVD 22%; and LVD-refractory: ETV 27%, LVD 30%. None of the subjects with hypocarbia had signs of LAS. The mechanism for hypocarbia in association with chronic hepatitis B is unknown. Additional cross-sectional analyses were performed, which showed that rates of hypocarbia at any single time point were comparable across the ETV and LVD treatment groups. Moreover, rates remained consistent with those observed at baseline suggesting no association to antiviral nucleoside therapy. A literature review did not provide any firm explanation for hypocarbia in patients with chronic hepatitis B. The database of ETV does not support any association with renal tubular acidosis.

- **Safety in special populations**

Decompensated patients (Study 048)
Overall, there were no patterns of AEs that would suggest a clear safety signal for ETV. However safety events were consistent with the expected natural history of the underlying decompensated cirrhosis. Death rates were 18% in the ETV group and 14% in the ADV group. The greater rates of SAEs (53%) and Grade 3-4 AEs (47%) observed in the ETV group compared with the ADV group (29% and 26%, respectively), was difficult to evaluate due to imbalances between groups in baseline HBV characteristics. The number of hepatic SAEs was increased in the ETV-treated compared with ADV-treated patients (31% *vs.* 17%). A fatal outcome was reported in 8 (18%) of the ETV-treated and 4 (11%) of the ADV-treated patients with 5 and 4, respectively, associated with a hepatic SAE. Information about the increased safety risks of anti-HBV therapies in decompensated patients is mentioned in the SPC and further data will be provided post-authorisation.

HIV/HBV co-infected patients (Study 038)
The safety profile of ETV in co-infected HIV/HBV LVD-refractory patients seems consistent with the one described in HBV monoinfected patients, with the exception of higher rate of grade 3-4 ALT elevations (18%), which seems, however, more likely related to the high ALT levels reported at baseline for these patients and to the concomitant HAART therapy. Supplementary data were provided, which showed that the safety profile with regard to Grade 3-4 ALT elevation was comparable to that in nucleoside naïve patients. Two patients discontinued due to hepatic events. No new ALT flares occurred during the 24-week open-label treatment phase. The open-label report provided cumulative 1-year data demonstrating stable HIV RNA suppression and an expected gradual rise in CD4 counts, suggesting that treatment with ETV does not affect the outcome of co-administered antiretroviral therapy.

Liver transplant population (Study 015)
No relevant safety findings arose from the study conducted in patients with OLT recipients. However, the limited number of patients in this study (9 subjects) precludes a definite conclusion for this population.

- Expanded access

Preliminary results (n= 48) from an early access programme, involving subjects with compensated chronic hepatitis B who have previously failed, are intolerant or have a contraindication to currently authorised nucleo(t)side anti-HBV treatment treated with ETV 1 mg showed no safety events that were qualitatively different than those reported in phase III clinical study.

Limited safety results from the roll-over study AI463901 showed that safety profile of the combination treatment including ETV 1.0 mg was similar to that observed in the phase II/III trials. The incidence of AEs, SAEs and Grade 3-4 laboratory abnormalities did not differ from that in phase II/III trials, but it is to be noted that the mean duration of therapy in study 901 was shorter (37 weeks). Notably there were two cases with severe thrombocytopenic events that were considered drug-related

(one had received previous LVD), one case of mild pancreatitis and one case that had symptoms suggestive of hepatic decompensation in association with an ALT flare.
Five malignant neoplasms, whereof two were recurrent, and 12 benign neoplasms were observed. There was no case of HCC reported.

5. Pharmacovigilance

▪ Detailed description of the Pharmacovigilance system

The CHMP considered that the Pharmacovigilance system as described by the applicant fulfils the legislative requirements.

▪ Risk Management Plan

The applicant submitted a risk management plan.

The safety specification covers all the issues that are considered relevant for the use of entecavir in clinical practice. Within the pharmacovigilance plan, most of the activities planned are routine pharmacovigilance practices. From the safety database all the adverse reactions reported in phase III clinical trials have been included in the Summary of Product Characteristics. In addition, the applicant undertook to closely monitor the following adverse reactions and report them in the PSUR according to the normal PSUR schedule, using the International birth date (29 March 2005) neurological and psychiatric events, musculoskeletal disorders and gastrointestinal disorders. Cases of entecavir exposure during pregnancy will also be presented in the PSURs. Moreover, the applicant is participating in a Pregnancy Registry to monitor foetal outcomes following ETV exposure during pregnancy.

Table 31: Summary of the risk management plan

Safety issue	Proposed pharmacovigilance activities	Proposed risk minimisation activities
Malignancies	AI463080 - Long-term outcomes study, including malignancies AI463-901, 050 and 049 - includes long-term cancer surveillance during 5 years in approximately 1500 patients	
Mitochondrial toxicity in particular lactic acidosis syndrome	Special attention in the PSUR	Class labelling warning on lactic acidosis in the SPC.
On treatment and post-treatment exacerbation of hepatitis	Special attention in the PSUR	Warning in SPC To monitor during therapy patients with cirrhosis as they may be at a higher risk for hepatic decompensation following hepatitis exacerbation *Post-treatment exacerbations*: To monitor hepatic function at repeated intervals with both clinical and laboratory follow-up for at least 6 months after discontinuation of hepatitis B therapy
Safety and efficacy in patients with hepatic decompensation	Ongoing study	Warning to regularly monitor these patients for clinical, virologic and serologic parameters associated with hepatitis B, liver and renal function and antiviral response during treatment, and if treatment is discontinued for at least 6

©EMEA 2006

		months after treatment.
Long term durability and clinical outcome	Roll-over studies and large observational study (AI463080)	
Resistance	Genotypic analysis of clinical failures and patients with detectable HBV DNA and additional analysessuch as predictive host and viral factors and cross-resistance to better estimate the impact of ETV resistance.	Warning to monitor virological response in the LVD refractory population, the increased risk of resistance development and to perform appropriate resistance testing.

The CHMP, having considered the data submitted in the application, is of the opinion that no additional risk minimisation activities are required beyond those included in the product information.

6.   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. The active substance is well characterised and documented. The pharmaceutical forms selected are adequate taking into account the properties and the stability of the drug substance. The excipients are commonly used for this kind of formulation and the packaging material is well documented. The manufacturing process enhances to obtain reproducible finished product batches. Stability tests under ICH conditions indicate that the product is stable for the proposed shelf life. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way.

Non-clinical pharmacology and toxicology

Entecavir is a nucleoside analogue with activity against HBV polymerase. Entecavir showed antiviral activity in vitro and in vivo models compatible with clinical use for the treatment of hepatitis B. Although lamivudine-resistant virus had decreased susceptibility to entecavir, data indicated that it may be a therapeutic option for lamivudine-refractory HBV patients.

The pharmacokinetics of entecavir has been adequately studied and high systemic exposures compared to the intended human exposure were achieved in all species. From a pharmacokinetic point of view the results from nonclinical studies may therefore be considered valuable.

In the repeat dose toxicity studies the range of target organs depended to a certain extent on species. For instance central nervous system inflammation was recorded in dogs but not in monkeys. Liver toxicity was mainly expressed in rats and mice. Gastro-intestinal toxicity was evident in rats and dogs but not in mice or monkeys. In a 12 months study in monkeys, no apparent toxicity was reported. Although mitochondrial changes were observed in some rat studies, overall, no specific mitochondrial toxicity induced by entecavir was reported in limited studies and little inhibitory activity on polymerase γ was evident in in vitro studies. In vitro and in vivo studies do not suggest that entecavir has any potential for QT prolongation. Entecavir crosses the placenta in rats and is found in milk of lactating animals. In reproductive toxicology, there was no evidence of impaired fertility or embryotoxicity. Embryo-foetal toxicity was observed at high exposure in rats. Considering these data and the lack of data in pregnant women, the SPC recommends that entecavir should not be used during pregnancy unless clearly necessary and that breastfeeding should be discontinued during treatment.

Entecavir showed genotoxic potential in one test (human lymphocyte test). In long term rodent carcinogenicity studies, entecavir induced tumours in both genders in mice and rats. In mice lung tumours developed at low doses and data indicate that a key event in the development of lung lesions

exhibited mouse specificity/selectivity. In vitro studies also showed that entecavir-TP was formed to a greater extent in mouse macrophages and pneumocytes than in rat such that a role of entecavir induced perturbation of dNTP pools cannot also be excluded. In rat, gliomas of microglial origin were observed and experimental data are consistent with that entecavir-TP levels above a certain threshold level may cause generalized dNTP pool perturbations and represents a plausible mechanism that may contribute to tumour development in susceptible tissues/organs. The predictivity of the data for human is not known. The applicant committed to further address post-authorisation the effects of combination of nucleoside/nucleotide analogues that could be used in the treatment of hepatitis B on clastogenicity and dNTP pools as well as to investigate in other species the binding of entecavir to receptors, that could play a role in the emergence of lung tumours.

Efficacy

The pharmacokinetics profile of entecavir is characterised by a rapid absorption, low binding to plasma proteins, excretion primarily via renal route. Administration of food with entecavir resulted in decrease exposure. In treatment naïve patients this is not expected to influence the efficacy so entecavir can be taken with or without food while in lamivudine refractory patients this could result in lower efficacy. Therefore for lamivudine-refractory patients the SPC recommends the administration of entecavir without food, at least 2 hours before or after a meal. Because entecavir is primarily renally excreted unchanged, renal impairment may increase entecavir exposure. Therefore for patients with moderate or severe renal impairment or end stage renal disease, dose adjustment is necessary as reflected in the SPC. No dosage adjustment is necessary for patients with hepatic impairment. Since entecavir is not a substrate, inducer and inhibitor of CYP450 isoenzymes or P-gP, the interaction potential is low. No interaction was observed with lamivudine, adefovir or tenofovir, all excreted renally via active tubular secretion, nonetheless it cannot be excluded that there may be interaction with other substances undergoing active renal secretion as highlighted in the SPC.

The clinical development of entecavir mainly consists of four well designed multicentre, randomised and double blind studies (one phase II and three phase III) that compare the efficacy of entecavir versus lamivudine in patients with chronic hepatitis B and compensated liver disease.
Two of these studies were conducted in nucleoside naïve patients (022 HBeAg positive and 027 HBeAg negative) and compared entecavir (0.5 mg once a day) to lamivudine (100 mg once a day). The remaining studies (014 and 026 HBeAg positive) were conducted in lamivudine refractory patients, defined as viraemic on lamivudine or with documented lamivudine resistance mutations. Study 014 was a dose ranging study, and the dose in 026 was 1 mg once a day.
The age ranges (mid 30 – mid 40s) and the male predominance in the ETV trials (75 %) reflect the natural course of chronic hepatitis B.

All studies had a 52-week duration of blinded treatment and a 24-week follow-up period. In all studies, subjects with partial response continued blinded treatment for up to 76-96 weeks. Patient management decisions at Week 52 were based on treatment response at Week 48. Complete responders (defined as undetectable HBV DNA <0.7 MEq/ml together with either - loss of HBeAg in studies 022 and 026 or - normal ALT in study 027 performed HBeAg negative patients) could stop treatment and be followed off treatment for 24 weeks. Partial responders (HBV DNA < 0.7 MEq/ml by bDNA assay but either still positive for HBeAg or with ALT >1.25 ULN) were to continue blinded treatment until complete response was achieved or until Week 96, whichever occurred first. The use of the less sensitive bDNA assay and HBeAg loss in the criteria could be criticised, but since the more sensitive PCR assay was used in parallel for quantification of HBV DNA and HBeAg seroconversion was included in the secondary endpoints, it could be considered satisfactory.

Nucleoside-naïve patients:

In study 022 (HBeAg positive patients) ETV 0.5 mg was superior to LVD at 48 weeks in the pre-specified endpoint of histological improvement with 72 % achieving this endpoint compared to 62 % in lamivudine group. ETV was also superior for several secondary endpoints, including the proportion of patients achieving HBV DNA below 300 copies/ml by PCR (67 % versus 36 %), the mean

reduction from baseline of HBV DNA level by PCR (ETV – 6.86 $\log_{10}$ copies/ml versus – 5.39 for lamivudine) and the proportion of patients who achieved ALT < 1.0 upper limit of normal (ETV 68 % versus 60 %). ETV patients with a partial response experienced incremental benefit during the second year of treatment, although HBeAg seroconversion rates were low in both treatment groups and did not differ at Week 96 between treatment groups (ETV 31%; LVD 26%). The ETV dose of 0.5 mg was considered optimal based on PK/PD analyses and efficacy data. Genotypic analysis of HBV DNA from nucleoside-naïve patients treated for up to 96 weeks identified no genotypic changes associated with phenotypic resistance to entecavir. For patients who met protocol-defined response criteria at Week 48, response was sustained throughout the 24-week post-treatment follow-up in 75% (83/111) of entecavir responders vs 73% (68/93) for lamivudine responders. The optimal treatment duration is, however, not known. Based on the stopping rule of this study, re-evaluation of data which did not indicate that 24 weeks of consolidation was clearly preferable to 12 weeks, and in accordance with international treatment guidelines for the consolidation phase, the SPC recommends that ETV should be administered at least until HBe seroconversion (HBeAg loss and HBV DNA loss with anti-HBe detection on two consecutive serum samples at least 3-6 months apart) or until HBs seroconversion or there is loss of efficacy.

In study 027 (HBeAg negative patients) ETV 0.5 mg was superior to LVD at 48 weeks in the pre-specified endpoint of histological improvement with 70 % achieving this endpoint compared to 61 % in lamivudine group. ETV was also superior for several secondary endpoints, including the proportion of patients achieving HBV DNA below 300 copies/ml by PCR (90 % versus 72 %), reduction from baseline of HBV DNA level by PCR (ETV – 5.04 $\log_{10}$ copies/ml versus – 4.53 for lamivudine) and the proportion of patients who achieved ALT < 1.0 upper limit of normal (ETV 78 % versus 71 %). Data suggest duration of treatment for at least 96 weeks, but neither the appropriate duration of treatment nor the appropriate endpoints sufficient to discontinue treatment have been determined. For patients who met protocol-defined response criteria at Week 48, response was sustained throughout the 24-week post-treatment follow-up in 46% for HBeAg negative patients. By 48 weeks of post-treatment follow-up a substantial number of patients lost response.
The proposed generic stopping criteria, i.e. HBsAg seroconversion seem acceptable when combined with a recommendation of regular reassessment of treatment when continued for more than 2 years.

LVD refractory population (HBeAg positive):
In study 026, ETV 1 mg was superior to LVD at 48 weeks in the pre-specified co-primary endpoint of histological improvement with 55 % achieving this endpoint compared to 28 % in lamivudine group. ETV was also superior for co-primary endpoint of HBVDNA < 0.7 Meq/ml and ALT < 1.25 ULN (57% versus 4 %). A greater proportion of patients achieving HBV DNA below 300 copies/ml by PCR (19 % versus 1 %), the mean reduction from baseline of HBV DNA level by PCR (ETV – 5.11 $\log_{10}$ copies/ml versus – 0.48 for lamivudine).
During the first year of treatment, only 19% of LVD refractory patients treated with entecavir achieved undetectable HBV DNA levels, which, however, increased to 40% during the second year when 85% of patients also had normalisation of ALT. All Year 1 and 2 data suggest that continuing lamivudine provides minimal or no benefit.
Given that at the time of the initiation of the study lamivudine was the only authorised oral treatment for the treatment of chronic hepatitis B, but that since adefovir has become the recommended treatment for lamivudine refractory patients, the usefulness of entecavir as first line in this population was questioned. There was also concern of the dose and the increased risk of resistance development in this population. Lamivudine was shown to select for the entecavir resistance-associated substitutions, as 6% (23/372) of the lamivudine-refractory patients were shown to harbour these substitutions already at baseline. During the first year of entecavir therapy, 6% (12/189) of patients were demonstrated to have new emergence of entecavir resistance-associated substitutions. Two patients experienced a virological rebound by Week 48, with the majority of the 12 patients experiencing virological failure during the second year of treatment. The total frequency of virological rebounds due to entecavir resistance up to Week 96 was 9% (14/155).
The applicant provided additional data to address these concerns. With respect to the dose, PK/PD profile suggested that the 1.0 mg ETV dose was acceptable, since even a large dose increase would only result in modest increase in efficacy. Moreover, the preclinical concern of a carcinogenic effect of entecavir with a threshold level is regarded as a limiting factor for exploring any dose increase.

©EMEA 2006

Sustained response rates in complete responders that discontinued ETV therapy at Week 48 were 50% (11 of 22 patients) for LVD refractory patients at 24 weeks off treatment

With respect to the efficacy, cross study comparisons with adefovir suggested that both products have comparable efficacy against lamivudine resistant HBV with respect to response rates for virological endpoints at 48 weeks. For instance in study 461 (3-arm trial of monotherapy with ADV versus add-on ADV with continued LVD versus continued LVD alone), the median decreases in HBV DNA from baseline were -4.04 $\log_{10}$ copies/ml (ADV only) and -3.59 $\log_{10}$ copies/ml (ADV+LVD) at Week 48. The proportion achieving an HBV DNA <1000 copies/ml was 5/19 (20%) and 7/21 (35%), respectively. By comparison, Week 48 results from the ETV Study 026 demonstrated a median reduction of -4.73 $\log_{10}$ copies/ml and a 26% rate of HBV DNA <1000 copies/ml.

With respect to resistance, more recent literature data suggest higher probably resistance to adefovir rate in LVD refractory patients, with rates up to 22% after 2 year of therapy[1]. Data from the other reports on ADV treatment only published as abstracts support these findings, and indicate that the emergence of ADV mutations appears to be more frequent among LVD refractory patients (15-58% in Year 2) than that previously reported in nucleoside-naive patients. Currently, there is no evidence that ETVr have any detrimental effect on the clinical efficacy of subsequent ADV or tenofovir. Short-term limited clinical data indicate that ETV is effective against ADVr virus and that ADV is active against ETVr virus. Whether substitution or add-on-therapy should be used in patients harbouring resistant viruses is not known and needs to be further evaluated in the post-approval period. Therefore, from an efficacy, safety and resistance perspective ETV is considered as a valuable addition in first-line treatment of this difficult-to-treat population. Management of LVD refractory patients requires nonetheless close monitoring of antiviral response during treatment and early use of resistance testing in patients with a suboptimal response, as recommended in the SPC.

The applicant committed to provide the results from the roll-over studies, as part of the follow-up measures to be fulfilled post-authorisation, looking particularly at some efficacy issues (e.g. optimal treatment duration and development of stopping criteria, maintenance of virological suppression beyond 24 weeks off-treatment, long-term durability of HBeAg seroconversion beyond 24 weeks off-treatment and the rate of new events, incidence of late HBsAg seroconversion, long-term clinical and histological outcomes and emergence of long term resistance).

Interim reports were provided for the ongoing studies in HBV/HIV co-infected patients (AI463038) and decompensated patients (AI463048). The data from both trials suggested that ETV therapy provided significant virological effects. The mean change from baseline in HBV DNA was -3.66 $\log_{10}$ copies/ml at Week 24 and -4.20 $\log_{10}$ copies/ml at Week 48 in LVD-refractory co-infected patients and -4.20 $\log_{10}$copies/ml in decompensated patients at Week 24. The data on co-infected patients are, however, not considered representative for all HIV populations since the majority of subjects had CD4 cell counts above 200 cells/mm^3. The demographics of this population is therefore detailed in the SPC. Overall, these preliminary data are very promising, but as yet limited and the applicant undertook to provide the final results as part of the follow-up measures to be fulfilled post-authorisation. The applicant also undertook to conduct further studies to evaluate the efficacy of ETV in orthotopic liver transplant patients, in HBV/HIV co-infected patients and using combination therapy.

There are currently no data to support the use of entecavir in children but the applicant undertook to complete the development programme in this population.

Safety

Assessment of adverse reactions was mainly based on four clinical studies in which 1,720 patients with chronic hepatitis B infection both in nucleoside-naïve and LVD-refractory patients with compensated chronic hepatitis B infection, received double-blind treatment with entecavir 0.5 mg/day (n = 679), entecavir 1 mg/day (n = 183), or lamivudine (n = 858) for up to 107 weeks. The safety profiles of entecavir and lamivudine, including laboratory test abnormalities, were comparable in these studies.

---

[1] Fung SK et al J Hepatol 2006:44:2983

The most common adverse reactions of any severity with at least a possible relation to entecavir were headache (9%), fatigue (6%), dizziness (4%) and nausea (3%).

No new safety signal occurred during the 2nd year of treatment compared to the 1st year. Although the ETV clinical development programme was relatively large, the size of the ETV programme and the observation time of treated patients (2-3 years) is insufficient to define long-term risk for infrequent events or events with a long latency period. Similarly, although the clinical programme covered a wide range of HIV-infected population, there is a need to further characterise the safety profile in some special population such as decompensated patients. This has been addressed in the risk management plan and the ongoing studies in this population and those studies looking at the long-term use of ETV will be submitted as part of the follow-up measures to be fulfilled post-authorisation. Identified risks, appropriately addressed in the submitted Risk Management Plan, include potential mitochondrial toxicity in particular lactic acidosis (safety issue related to nucleos(t)ide analogues), on-treatment and post-treatment exacerbations of hepatitis (based on current experience with treatment for chronic hepatitis B) and emergence of resistance to ETV. The key safety issue is entecavir´s potential carcinogenicity deriving first, from the inherent risk of HCC and other malignancies (e.g., lymphoma), related to HBV itself, and second, from the possible increased risk of non-HCC malignant neoplasms in light of the rodent carcinogenicity findings. Although the mechanistic assessment and the current safety data do not indicate any signal for increased rate of human cancer, the theoretical concern for an increased risk on non-HCC cancers to humans is to be ascertained with long-term follow-up. The applicant will therefore conduct a large (over 10,000 patients), global, open-label observational study aimed at examining the long-term benefits and risks of ETV therapy. The proposed study will compare rates of HBV disease progression (including HCC), mortality and development of malignant neoplasms (overall number of malignancies and non-HCC malignancies, in each case excluding non-melanoma skin cancer) in patients with chronic HBV infection who are randomised to nucleoside/tide monotherapy with either ETV or another standard of care HBV nucleoside/tide analogue and followed over an observation period of up to 10 years. The population will be a broad group of patients with chronic HBV infection who are initiating HBV therapy or treatment experienced and in need of alternate nucleoside/tide therapy.

Moreover, new and recurring malignancies observed in ETV clinical studies will be monitored during a 5-year follow-up period in approximately 1500 subjects.

From the safety database ETV related AE's reported in the clinical trials have been included in the Summary of Product Characteristics.

All the information has been appropriately translated into the package leaflet for which a user test has been adequately performed.

Benefit/risk assessment

The clinical experience included different patient populations recruited worldwide, with Asians and Caucasians particularly well represented: nucleoside-naïve and lamivudine- refractory patients; HBeAg positive and negative; transplant, HIV co-infected patients and in an on-going trial decompensated patients. This is in line with the spirit of the guideline related to anti-hepatitis B medicinal products.

Based on the clinical efficacy, safety and resistance data of entecavir, which were overall superior or comparable to the current anti-HBV compounds, the benefit risk ratio of entecavir is considered positive in the naïve population with compensated chronic hepatitis B, as well as in lamivudine-refractory patients which is a difficult-to-treat population. Preliminary results showed also efficacy of entecavir in HBV/HIV co-infected patients and in patients with decompensated disease. Although the antiviral efficacy of entecavir has been demonstrated, a number of clinical questions related to the use of entecavir in the management of hepatitis B infected patients (e.g optimal duration of treatment, durability) will be addressed post-authorisation. The chronic nature of hepatitis B virus therapy and long-term disease outcomes are such that clinical trials do not fully address long-term risk of HBV therapy. A Risk Management Plan was submitted and includes agreed pharmacovigilance activities to further define the safety profile post-authorisation. The theoretical concern for an increased risk on non-HCC cancers to humans based on the carcinogenicity findings in rodents will be addressed in a large randomised prospective observational study with long-term follow-up.

Due to the nature of the disease, ETV therapy should be initiated by a physician experienced in the management of chronic hepatitis B.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the benefit/risk ratio of Baraclude in the following indication:

- Treatment of chronic hepatitis B virus (HBV) infection in adults with compensated liver disease and evidence of active viral replication, persistently elevated serum alanine aminotransferase (ALT) levels and histological evidence of active inflammation and/or fibrosis. This indication is based on clinical trial data in patients with HBeAg positive and HBeAg negative HBV infection, nucleoside naive patients and patients with lamivudine-refractory hepatitis B (see sections 4.4 and 5.1 *of the Summary of Product Characteristics*).

was favourable and therefore recommended the granting of the marketing authorisation.

©EMEA 2006