# CENTER FOR DRUG EVALUATION AND RESEARCH

***APPLICATION NUMBER:***

# 125431Orig1s000

# PHARMACOLOGY REVIEW(S)

**Tertiary Pharmacology/Toxicology Review**

**From**: Timothy J. McGovern, Ph.D., ODE Associate Director for Pharmacology and Toxicology, OND IO
**BLA:** 125431
**Agency receipt date:** January 14, 2013
**Drug:** TANZEUM (Albiglutide)
**Sponsor:** GlaxoSmithKline

**Indication:** Treatment of type 2 diabetes mellitus

**Reviewing Division:** Division of Metabolism and Endocrinology Products

**Introductory Comments:** The pharmacology/toxicology reviewer and supervisor concluded that the nonclinical data support approval of albiglutide for the indication listed above.

The recommended pharmacologic class for albiglutide is a glucagon-like peptide-1 (GLP-1) receptor agonist. Exenatide (Byetta and Bydureon) and liraglutide (Victoza) are GLP-1 agonists that have been previously approved in the US.

Albiglutide is a recombinant fusion protein composed of two copies of $[Gly^8]GLP\text{-}1_{(7\text{-}36)}$, in tandem, fused to human albumin. An appropriate nonclinical program was conducted by the sponsor to support approval of albiglutide. Albiglutide elicited expected pharmacological responses in the species evaluated. The primary nonclinical toxicity studies were conducted in monkeys due to the anti-drug antibody (ADA) response in mice that led to diminished drug exposure and type 3 hypersensitivity reactions in a subset of animals. No significant findings were observed in a 52-week monkey toxicity study at doses up to 50 mg/kg/week. Although a possible signal for an increased incidence of pancreatitis has been reported post-marketing for incretin-based treatments (GLP-1R agonists and DPP-IV inhibitors), no signal suggesting an increased risk of pancreatitis was observed in studies with albiglutide. It is not possible to use typical diabetic rodent models to assess whether chronic albiglutide treatment is associated with pancreatic injury.

Genetic toxicity studies were not applicable for this program and carcinogenicity studies were not possible in rodents due to immunogenicity reactions that limit dosing to approximately 2 weeks. The carcinogenicity assessment was based on a weight-of-evidence approach including effects observed with other GLP-1 agonists (long acting GLP-1R agonists have been associated with hyperplastic/neoplastic changes in thyroid C-cells in rodents), results of a 52-week toxicity study in monkeys, and clinical data. It was concluded that albiglutide carries the same degree of risk regarding thyroid C-cell hyperplasia as other approved long-acting GLP-1R agonists and the labeling should carry comparable warnings and precautions as the other products.

Other compounds in this class have been required to conduct post-marketing studies to further characterize the potential clinical risk posed by thyroid C-cell



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proliferative/neoplastic signals observed in lifetime rodent studies. These studies are not practical in lifetime rodent studies with albiglutide due to rapid appearance of clearing ADA in rodents. The sponsor intends to conduct studies in the mu-deficient mouse model to ascertain whether albiglutide treatment is associated with induction of focal hyperplasia of thyroid C-cells with prolonged exposure.

Reproductive and developmental toxicity studies were conducted in mice only and evaluations were partitioned to limit the dosing to no more than 15 days due to production of ADAs. Key findings included embryo-fetal lethality and bent ribs which may have been at least partially attributable to maternal toxicity, reduced body weight, a delay in balanopreputial separation, increased clinical signs in offspring, and delayed onset of vaginal patency. No teratogenic findings were observed. However, a "Pregnancy Category C" for this product was recommended consistent with the current labels for Byetta and Victoza given the limited evaluations that could be conducted with albiglutide.

**Conclusion:**
I agree with the division pharmacology/toxicology conclusion that albiglutide can be approved from the pharmacology/toxicology perspective. I have reviewed the proposed labeling and agree with the recommendations made by the division regarding the relevant nonclinical sections.



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/s/

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TIMOTHY J MCGOVERN
04/09/2014

**DEPARTMENT OF HEALTH AND HUMAN SERVICES**
**PUBLIC HEALTH SERVICE**
**FOOD AND DRUG ADMINISTRATION**
**CENTER FOR DRUG EVALUATION AND RESEARCH**

# PHARMACOLOGY/TOXICOLOGY NDA/BLA REVIEW AND EVALUATION

| | |
|---|---|
| Application number: | BLA 125431 |
| Supporting document/s: | 0000 |
| Applicant's letter date: | January 11, 2013 |
| CDER stamp date: | January 14, 2013 |
| Product: | Albiglutide (EPERZAN™) |
| Indication: | Treatment of type 2 diabetes mellitus |
| Applicant: | GlaxoSmithKline |
| Review Division: | Metabolism and Endocrinology |
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*Template Version: September 1, 2010*

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# 1 Executive Summary

## 1.1 Introduction

Albiglutide (also known as GSK716155 or Albugon) is a recombinant fusion protein composed of 2 copies of [Gly$^{8}$]GLP-1$_{(7\text{-}36)}$, in tandem, fused to human albumin. The pharmacodynamic activity of albiglutide arises from agonism at the Glucagon-Like Peptide-1 (GLP-1) receptor (GLP-1R). This protein is engineered to have resistance to the inactivating action of dipeptidyl peptidase IV (DPP-IV), and to have a prolonged circulatory half-life. The albumin component conveys no specific pharmacodynamic activity of its own, but rather substantially increases the circulatory half-life of the tandem [Gly$^{8}$]GLP-1$_{(7\text{-}36)}$ moiety. The presence of the albumin moiety has no discernable effect on the expected toxicologic profile of a GLP-1R agonist. (b) (4)

## 1.2 Brief Discussion of Nonclinical Findings

Pharmacology

*In vitro* studies demonstrated that albiglutide has agonist activity at mouse, rat, monkey and human GLP-1 receptors. *In vivo* studies in mice, rats or monkeys demonstrated that albiglutide increases glucose-stimulated insulin secretion and improves glucose tolerance, and delays gastric emptying and reduces food intake. The aggregate effects of reduced nutrient uptake and increased glucose disposal support the proposed beneficial anti-diabetic activity of albiglutide, and albiglutide improved glycemic control in several rodent models of diabetes.

In a rodent model of myocardial ischemia-reperfusion injury, albiglutide significantly reduced myocardial infarct size, and improved post-ischemic cardiac left ventricular function. The exact mechanism of these cardioprotective effects was not determined, but may be related to direct effects of GLP-1R agonism on the myocardium and/or to indirect effects of increased insulin secretion and cardiomyocyte utilization of glucose.

Albiglutide produced no adverse effects in monkeys in a combined cardiovascular and respiratory system safety pharmacology study, and did not affect neurobehavioral functional endpoints assessed in monkeys as part of a repeat dose toxicity study. In addition, there were no electrocardiograph (ECG) findings in the monkey safety pharmacology study or following administration of 50 mg/kg/week (75X maximum anticipated clinical exposure) for 52 weeks. In accordance with current guidance the hERG assay was not conducted with albiglutide.

Pharmacokinetics

As would be expected for a polypeptide of ~73,000 Da, albiglutide has a low volume of distribution (similar to the vascular volume) in both mice and monkeys, and as such allometric scaling based on body mass (rather than body surface area) provides a reasonable estimate of interspecies systemic exposures. The circulatory half-life was



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on the order of 8 hours in mice and 60 hours in monkeys. Bioavailability following a SC injection was at least 38% in mice and 50% in monkeys. No meaningful sex-related differences in PK parameters were seen in either species. In accordance with current guidance, tissue distribution studies, drug metabolism studies and excretion studies were not conducted with albiglutide, as it is a protein-based therapeutic. An assessment of the effect of albiglutide on CYP mRNA levels in monkeys treated with albiglutide for 5 weeks found minimal increases in CYP1A2 message in males and minimal increases in CYP2B17 levels in females.

Toxicity

An acute subcutaneous and intravenous study in the mouse and repeat subcutaneous dose studies for durations of up to 15 days in the mouse and 52 weeks in the monkey have been performed. Unsurprisingly, given the presence of human albumin sequence in the fusion protein, albiglutide was highly immunogenic in animals. In mice, the anti-drug antibody (ADA) response caused diminished exposure to albiglutide and was associated with type 3 hypersensitivity reactions (immune complex deposition) in a subset of animals. Therefore it was concluded that longer-term studies (exceeding ~2 weeks) with albiglutide in rodents would likely be uninformative to clinical risk. As a consequence of this, some of the development and reproductive toxicology (DART) studies were subdivided into multiple 2-week exposure periods. In addition, it was not feasible to conduct chronic toxicity or 2-year carcinogenicity studies in rodents. While albiglutide was also immunogenic in monkeys, the ADA response did not appreciably alter exposure to active drug in the majority of monkeys, nor did it discernably contribute to the toxicity findings of the monkey studies.

Acute toxicity associated with single high doses of albiglutide was assessed in mice at doses of 50, 200, 500 and 1000 mg/kg SC and 500 mg/kg IV. A transient, dose-dependent body weight drop (max. ↓11-15%) was seen in both sexes at SC doses ≥200 mg/kg and the 500 mg/kg IV dose. Microscopic findings were observed in male reproductive tissues: epididymis (vacuolation and cell debris), seminal vesicle (vacuolation), and prostate (vacuolation and lymphocyte infiltration) of male mice at 500 mg/kg IV and in the epididymis (vacuolation and cell debris) at doses ≥ 500 mg/kg SC. The maximum tolerated dose in this study is considered to be >1000 mg/kg SC and >500 mg/kg IV. Based on the microscopic findings in male reproductive tissue, the male NOAEL is 200 mg/kg SC (240X the MRHD) and <500 mg/kg IV. The female NOAEL is 1000 mg/kg SC (1200x the MRHD) and 500 mg/kg IV.

For the repeat-dose general toxicity studies, this summary focuses on the monkey studies, as the interpretation of the 15-day rodent study is confounded by the ADA response discussed above. In general the results of the monkey toxicology studies are consistent with agonism at the GLP-1R: decreased food consumption, emesis, dehydration and decreased body weight gain (and occasionally transient frank body weight loss). These effects are most marked following the first few doses and become attenuated thereafter. Transient reductions in blood glucose were also seen following the first dose in the 4-week monkey toxicity study. In the pivotal 52-week toxicity study no dose-limiting toxicity was seen at doses up to 50 mg/kg/week (the highest dose



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tested). There were mild injection site reactions (minimal perivascular mononuclear cell infiltrates) in albiglutide-treated animals. Most notably, albiglutide (and exenatide used as a comparator) caused a dose-related increase in pancreas weight; however, there were no histopathologic abnormalities detected in the pancreases of albiglutide-treated monkeys. Stereological examination suggested that all components of the pancreas (islet, duct and acinar) exhibited a more or less proportional increase in volume. Also there was no significant increase in the percentage of cells undergoing proliferation in any particular cell population, as assessed by Ki67 immunostaining. It is unknown whether the increase in pancreas weight is reversible, as there was no recovery group in the 52 week study, and pancreas weight was not determined in any of the shorter duration toxicity studies. In the 52-week monkey study the thyroid was also subjected to intensive analysis, which included: a) standard H&E stain, b) calcitonin immunohistochemistry and c) quantitative (stereological) analysis of total C-cells. There were no albiglutide-related findings in the thyroid. The NOAEL for the 52-week monkey study was 50 mg/kg/week (75X the maximum anticipated clinical exposure), the highest dose tested.

Reproductive and Developmental Toxicity -- In reproductive and developmental toxicity studies in mice, prolonged estrous cycle, embryofetal lethality, and bent/wavy ribs in the fetus were observed at 50 mg/kg/day (39X the maximum anticipated clinical exposure), a dose that caused signs of maternal toxicity (body weight loss, reduced food consumption, and reduced plasma glucose). In a mouse male fertility study at doses up to 50 mg/kg/day (34X the maximum anticipated clinical exposure), there were no test article-related effects on male reproductive performance, male reproductive organs, or embryonic survival. In mouse pre/postnatal (PPN) development studies, albiglutide produced dose-dependent reduced postnatal body weights of F1 pups, generally at all doses. Possibly as a consequence of reduced body weights, there were slight dose-dependent delays in the onset of sexual maturation in both sexes that were occasionally s.s. at 50 mg/kg/day. At doses up to 50 mg/kg/day there were no adverse effects on maternal pregnancy, parturition, lactation, or offspring survival. Also, there was no effect on neurobehavioral or reproductive function or survival or growth of the offspring. F0 exposure to albiglutide from Day 15 post-coitus to Day 10 post-partum resulted in a dose-related decrease in the live litter size for the F2 generation, which was s.s. at 50 mg/kg/day (39X the maximum anticipated clinical exposure). This was likely related to a reduction in the number of implantation sites in the F1 females. In the PPN studies, there was an increased incidence of mortality and morbidity in lactating F0 dams. At doses that produced an increased incidence of mortality, there were no exposure margins (by AUC) at the estimated clinical exposure at the MRHD of 50 mg/week. Mortalities occurred only during lactation and have not been observed in previous toxicology studies in non-pregnant, non-lactating, or pregnant mice. The findings and timing of onset were consistent with lactational ileus (paresis of peristalsis), and mortality was reduced by taking measures to reduce metabolic demand (culling litter size) and adding high energy dietary supplements. The propensity of albiglutide to cause/exacerbate lactational ileus in mice is not considered to be clinically relevant, since the relative stress of lactation energy demands is much lower in humans and humans have relatively large energy reserves, compared to mice.



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## 1.3 Recommendations

### 1.3.1 Approvability

Approval.

### 1.3.2 Additional Non Clinical Recommendations

None that are considered to be required for approval of the current application.

Although other compounds in this class of agents (GLP-1R agonists) have been required to conduct post-marketing studies to further characterize the potential clinical risk posed by thyroid C-cell proliferative/neoplastic signals observed in lifetime rodent studies, comparable studies with albiglutide are not practical in typical rodent models due to the rapid appearance of clearing ADA to albiglutide in rodents. For the same reason, it is not practical to use typical diabetic rodent models to assess whether chronic albiglutide treatment is associated with pancreatic injury, which has appeared as a possible post-marketing clinical signal with other incretin therapies (GLP-1R agonists & DPP-IV inhibitors).

Sponsor intends to conduct studies in the mu-deficient mouse model (which is unable to make antibodies) to ascertain whether albiglutide treatment is associated with induction of focal hyperplasia of thyroid C-cells with prolonged (6-month) exposure. The sensitivity of this model system for identifying GLP-1R-driven C-cell hyperplasia will be established with another long-acting GLP-1R agonist that has demonstrated C-cell hyperplastic/neoplastic activity in standard rodent models. The mu-deficient mouse model system is not considered to be suitable however for a lifetime carcinogenicity study due 1) to the immunocompromised state of these animals, which both confounds interpretation of tumor incidence data and makes the animals difficult to maintain over extended periods of time, and 2) due to the absence of a suitable control background tumor database.

Sponsor will conduct a juvenile toxicity study in a rodent model prior to initiation of pediatric trials with albiglutide.

### 1.3.3 Labeling

The relevant nonclinical sections of the label are shown below:

**WARNING: RISK OF THYROID C-CELL TUMORS**

- **Thyroid C-cell tumors have been observed in rodent studies with glucagon-like peptide-1 (GLP-1) receptor agonists. It is unknown whether [redacted] (b) (4) causes thyroid C-cell tumors, including medullary thyroid carcinoma (MTC), in humans [redacted] (b) (4) *(5.1)].***
- **[redacted] (b) (4) is contraindicated in patients with a personal or family history of MTC or in patients with Multiple Endocrine Neophasia syndrome type 2 (MEN 2). [redacted] (b) (4) *(4)* [redacted] (b) (4) *(5.1)].***



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**----------WARNINGS AND PRECAUTIONS----------**

- [redacted] (b) (4)

**----------USE IN SPECIFIC POPULATIONS----------**

- Pregnancy: [redacted] (b) (4) may cause fetal harm. [redacted] (b) (4) only if the potential benefit justifies the potential risk to the fetus (8.1).
- Nursing Mothers: [redacted] (b) (4)

**FULL PRESCRIBING INFORMATION**

**WARNING: RISK OF THYROID C-CELL TUMORS**

- **Thyroid C-cell tumors have been observed in rodent studies with glucagon-like peptide-1 (GLP-1) receptor agonists. It is unknown whether [redacted] (b) (4) causes thyroid C-cell tumors, including medullary thyroid carcinoma (MTC), in humans *[see Warnings and Precautions (5.1)]*.**
- **[redacted] (b) (4) is contraindicated in patients with a personal or family history of MTC or in patients with Multiple Endocrine Neoplasia syndrome type 2 (MEN 2). *[see Contraindications (4) and Warnings and Precautions (5.1)]*.**

## 5 WARNINGS AND PRECAUTIONS

### 5.1 Risk of Thyroid C-cell Tumors

Nonclinical studies in rodents [redacted] (b) (4) Carcinogenicity studies [redacted] (b) (4) not been conducted with [redacted] (b) (4) It is unknown whether [redacted] (b) (4) thyroid C-cell tumors, including MTC in humans *[see Boxed Warning and Contraindications (4)]* [redacted] (b) (4) [redacted] (b) (4)

## 8 USE IN SPECIFIC POPULATIONS

### 8.1 Pregnancy

Pregnancy Category C.

There are no adequate and well-controlled studies of [redacted] (b) (4) in pregnant women. [redacted] (b) (4) *[see Nonclinical Toxicology (13.3)]*. [redacted] (b) (4)

(b) (4) should not be used during pregnancy unless the expected benefit outweighs the potential risks.

(b) (4) consider stopping (b) (4) at least 1 month before a planned pregnancy (b) (4)

(b) (4)

(b) (4)

There are no data on the effects of (b) (4) on human fertility. Studies in mice showed no effects on mating or fertility *[see Nonclinical Toxicology (13.1)]*. The potential risk (b) (4) is unknown.

### 8.4 Pediatrics

Safety and effectiveness of (b) (4) have not been established in pediatric patients ((b) (4) 18 years (b) (4)).

## 13 NONCLINICAL TOXICOLOGY

### 13.1 Carcinogenesis, Mutagenesis, Impairment of Fertility

As albiglutide is a recombinant protein, no genotoxicity studies have been conducted.

(b) (4) Thyroid C-cell tumors were observed in 2-year rodent carcinogenicity studies with (b) (4) GLP-1 receptor agonists. The clinical relevance of (b) (4) is unknown.

In a mouse fertility study, males were treated with SC doses of 5, 15 or 50 mg/kg/day for 7 days prior to cohabitation with females, and continuing through mating. In a separate fertility study, females were treated with SC doses of 1, 5 or 50 mg/kg/day for 7 days prior to cohabitation with males, and continuing through mating. There were no effects on mating or fertility in either sex at doses up to 50 mg/kg/day (b) (4)

### 13.3 Reproductive and Developmental Toxicity

In order to minimize the impact of the drug-clearing anti-drug antibody response, reproductive and developmental toxicity assessments in the mouse were partitioned to limit the dosing period to no more than approximately 15 days in each study. In

pregnant mice given SC doses of 1, 5 or 50 mg/kg/day from gestation day 1 to 6, there were no adverse effects on early embryonic development through implantation at 50 mg/kg/day (b) (4)

In pregnant mice given SC doses of 1, 5 or 50 mg/kg/day from gestation day 6 through 15 (organogenesis), embryo-fetal lethality (post-implantation loss) and bent (wavy) ribs were observed at 50 mg/kg/day (b) (4)

Pregnant mice were given SC doses of 1, 5 or 50 mg/kg/day from gestation day 6 to 17. Offspring of pregnant mice given 50 mg/kg/day (b) (4) had reduced body weight pre-weaning, (b) (4)

Pregnant mice were given SC doses of 1, 5 or 50 mg/kg/day from gestation day 15 to lactation day 10. Increased mortality and morbidity were seen at all doses (≥1 mg/kg/day) in lactating females in mouse pre and postnatal development studies. Mortalities have not been observed in previous toxicology studies in non-lactating or non-pregnant mice, nor in pregnant mice. These findings are consistent with lactational ileus syndrome which has been previously reported in mice. Since the relative stress of lactation energy demands is lower in humans than mice and humans have large energy reserves, the mortalities observed in lactating mice are of questionable relevance to humans. (b) (4)

(b) (4) were given SC doses of 1, 5 or 50 mg/kg/day from lactation day 7 to 21 (weaning) under conditions that limit the impact of lactational ileus (increased caloric intake and culling of litters). Doses ≥1 mg/kg/day (b) (4) caused reduced weight gain in the pups during the treatment period. (b) (4)

# 2 Drug Information

## 2.1 Drug

**CAS Registry Number:** **782500-75-8**

**Generic Name:** **Albiglutide**

**Code Name:** **GSK716155, Albugon**



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**Chemical Name:** **n/a**

**Molecular Weight:** **~73,000 Da**

**Structure or Biochemical Description:** **Recombinant fusion protein. Two copies of [Gly$^{8}$]GLP-1$_{(7-36)}$ in tandem recombinantly fused to human albumin.**

**Figure 1 Schematic Representation of Albiglutide**

(b) (4)

**Pharmacologic Class:** **Glucagon-like peptide-1 (GLP-1) receptor agonist**

## 2.2 Relevant INDs, NDAs, BLAs and DMFs

INDs: (b) (4) (b) (4) (b) (4)

NDAs: 21-773 (Exenatide, (b) (4)), 22-341 (Liraglutide, Novo Nordisk), 22-200 (Exenatide for extended release, Amylin Pharmaceuticals)



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## 2.3 Drug Formulation

**Table 1 Composition of Albiglutide for Injection, 30 mg/DCC and 50 mg/DCC**

| Component | Quantity (mg) | | Function | Reference to Standards |
|---|---|---|---|---|
| | 30 mg/DCC | 50 mg/DCC | | |
| Albiglutide | 40.30 | 66.95 | Active substance | GSK Specification |
| Trehalose dihydrate | (b) (4) | | | USP-NF, EP |
| Mannitol | | | | USP-NF, EP, JP |
| (b) (4) | | | | USP-NF, EP |
| Sodium phosphate (b) (4) | | | | USP-NF |
| Polysorbate 80 | | | | USP-NF, EP, JP |
| Water For Injection | 0.65 mL | 0.65 mL | (b) (4) | USP-NF, EP, JP |

## 2.4 Comments on Novel Excipients

Not applicable.

## 2.5 Comments on Impurities/Degradants of Concern

No impurities/degradants of toxicological concern have been identified. Both process 1 and process 2 drug substance have been used during the nonclinical development program, and both evidenced comparable toxicity profiles consistent with exaggerated pharmacology at the GLP-1R. No nonclinical studies have been conducted with the process 3 drug substance; however the levels of impurities/degradants identified in the commercial product (process 3) are considered to have been adequately qualified in the drug lots used in the nonclinical toxicology testing program.

See CMC review for details of differences in the impurity/degradant profile of processes 1, 2 and 3 drug substance.

## 2.6 Proposed Clinical Population and Dosing Regimen

Population: adults with type 2 diabetes mellitus
Regimen: 30 mg once weekly SC injection, may be increased to 50 mg once weekly SC injection

## 2.7 Regulatory Background

*The following is limited primarily to nonclinical development.*

A Pre-IND meeting was held between Human Genome Sciences and the FDA on 27-AUG-2002 to discuss appropriate support for a FIH study with albiglutide (Albugon).

A second Pre-IND meeting was held between GSK and FDA for albiglutide (GSK716155) on 30-MAR-2005 to discuss the development program including nonclinical toxicology and the planned FIH study. In view of the rapid drug-clearing ADA response seen in rodents, FDA agreed that longer-term toxicity studies could be conducted solely in the non-human primate. FDA also agreed that a cohort study with once daily dosing (not to exceed ~14 days of dosing) in rodents to evaluate fertility and embryofetal and pre-/post-natal development were appropriate.

In response to a GSK request of 21-DEC-2006 (Serial No. 0013) for feedback regarding the adequacy of the completed and planned nonclinical development program to support a marketing application, FDA advised (letter of 20-MAR-2007) that a 12-month monkey study should be conducted to assist in the evaluation of pre-neoplastic potential of albiglutide, based on thyroid C-cell hyperplastic/neoplastic signals seen with other members of the class in rodents. FDA recognized that it was not technically feasible to conduct rodent carcinogenicity studies in rodents, because of the rapid clearing anti-product antibody response that occurs with albiglutide in rodents. GSK subsequently submitted on 29-JAN-2008 (Serial No. 0114) a protocol for a 12-month monkey study for FDA feedback. In our response of 10-APR-2008, FDA advised that a NOAEL dose of 15 mg/kg/week was insufficient for the study, rather a MTD should be the high dose for the toxicity study. A high dose of 50 mg/kg/week was agreed to on 21-APR-2008. On 22-JAN-2010, GSK submitted reports for the 52-week monkey toxicity study (CD2009/00863/00). On 23-JUL-2010, FDA commented that the organ weight data from the 52-week monkey study suggested a dose-related increase in pancreas mass with albiglutide and requested that GSK determine which cellular components are involved in the increase and assay samples for increased proliferative activity.

On 11-FEB-2011 (Serial No. 0348), GSK submitted preliminary observations on the occurrence of mortality in lactating dams from the mouse postnatal study and plans for investigation. On 29-MAR-2011, FDA concurred with the proposed investigations to determine the cause of maternal mortality. On 19-JUN-2012, FDA issued an Advice Letter citing a recent analysis suggesting that long-acting GLP-1R agonists have the potential to accelerate sexual maturation, as demonstrated by increased male reproductive organ weights in pre-/peri-pubertal male monkeys during general toxicology studies of 13 weeks duration or longer. Although the basis for this effect is unknown, published data suggest that GLP-1 can stimulate gonadotropin-releasing hormone (GnRH) from the hypothalamus. If the same phenomena were found to occur



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in humans, pediatric subjects given long-acting GLP-1R agonists might enter puberty earlier than anticipated, which could potentially affect height and psychosocial development. Thus, FDA advised that sponsors planning studies including prepubertal children should address this concern by conducting a juvenile animal study covering the stage of sexual maturation, including careful assessment of the onset of puberty, rate of sexual maturation, rate of overall growth and reproductive maturation in both sexes.

GSK's response to FDA's Advice Letter was submitted in Serial No. 0409 (09-JUL-2012). The response described relevant data (testes organ weight and histology) from four general toxicology studies with albiglutide conducted in monkeys. However, since sexual maturity status was not determined at the start of the study, the effects of albiglutide on sexual maturation could not be evaluated in these studies. In view of potential concern regarding possible effects of GLP-1R agonists on sexual maturation in children, GSK plans to conduct a mouse juvenile toxicity study to determine if albiglutide has effects on sexual maturation and CNS/behavior.

*Pre-BLA Communications*
On 18-APR-2012, FDA granted the Type C Meeting Request with written responses to be provided within 60 days of submission of the briefing package in lieu of a face-to-face meeting. The nonclinical portion of the Briefing Package was submitted as Serial No. 0404 (08-MAY-2012) and contained 3 nonclinical questions for FDA. These questions concerned a planned evaluation of albiglutide's potential to produce thyroid hyperplasia in an immunodeficient mouse model, explanation of maternal deaths in the PPN study due to metabolic demand, and conclusions from a stereological evaluation of pancreas from the 52-week monkey toxicology study. In our 06-JUL-2012 written response to the Type C Briefing Packages, FDA expressed concern regarding the ability of the proposed immunodeficient mouse study to materially affect labeling, but agreed that the PPN studies and conclusions from the pancreas stereological evaluation appeared to have addressed sufficiently the earlier questions/concerns.



Reference ID: 3347159

# 3 Studies Submitted

## 3.1 Studies Reviewed

### 1. PHARMACOLOGY: OVERVIEW FOR ALBIGLUTIDE

**Table 1.1 List of Primary Pharmacodynamic Studies Performed with Albiglutide**

| Type of Study | Species (Strain)[a]/ Test System | Method of Administration[b] | Form[c] | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|
| Effect on GLP-1 receptor binding and intracellular cAMP | HEK293 cells expressing the human GLP-1R | In vitro | A1, GLP-1 | No | HGS | RD2005/01132 (FS29900.DBS.0.001) | m4.2.1.1 |
| Comparison of cAMP-inducing activity | HEK293 cells expressing the human GLP-1R | In vitro | A1, GLP-1, Ex-4 | No | HGS | RD2005/01161 (FS29900.DBS.0.035) | m4.2.1.1 |
| Action on GLP-1 receptor | HEK293 cells expressing the mouse, rat or human GLP-1R | In vitro | A1, GLP-1, Ex-4, L | No | GSK | 2012N146049 | m4.2.1.1 |
| Action on GLP-1 receptor | HEK293 cells expressing the monkey or human GLP-1R | In vitro | A1, GLP-1 | No | HGS | RD2005/01148 (FS29900.DBS.0.008) | m4.2.1.1 |
| Resistance to proteolytic cleavage | Incubation with DPP-IV | In vitro | A1, GLP-1[d] | No | HGS | RD2005/01160 (FS29900.DBS.0.020) | m4.2.1.1 |
| Induction of insulin | INS-1 (832/13)[e] | In vitro | A1, GLP-1 | No | HGS | RD2005/01142 (FS29900.DBS.0.003) | m4.2.1.1 |
| Effect on glucose tolerance | Mouse (db/db or heterozygote lean controls) | Subcutaneous | A1, Ex-4 | No | GSK | RH2005/00082 | m4.2.1.1 |
| Comparison of activity from 2 DNA constructs | Mouse (db/db) | Subcutaneous | A1[f], rHA, Ex-4 | No | HGS | RD2005/01158 (FS29900.DBS.0.011) | m4.2.1.1 |
| Effect of single administration | Mouse (db/db) | Subcutaneous | A1, rHA, Ex-4 | No | HGS | RD2005/01159 (FS29900.DBS.0.022) | m4.2.1.1 |
| Duration of action on glucose tolerance (single administration) | Mouse (db/db or heterozygote lean controls) | Subcutaneous | A1, Ex-4 | No | HGS | RD2005/01156 (FS29900.DBS.0.023) | m4.2.1.1 |



Reference ID: 3347159

**Table 1.1 (Continued) List of Primary Pharmacodynamic Studies Performed with Albiglutide**

| Type of Study | Species (Strain)[a]/ Test System | Method of Administration[b] | Form[c] | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|
| Effect of repeated administration | Mouse (db/db) | Subcutaneous (daily for 6 days) | A1, rHA, Ex-4 | No | HGS | RD2005/01283 (FS29900.DBS.0.002) | m4.2.1.1 |
| Combination effect with metformin on glucose tolerance | Mouse (db/db) | Subcutaneous | A1, rHA, Met | No | HGS | RD2005/01155 (FS29900.DBS.0.036) | m4.2.1.1 |
| Effect on glucose tolerance (single administration) | Rat (ZDF or lean controls) | Intravenous | A1, rHA, Ex-4 | No | HGS | RD2005/01149 (FS29900.DBS.0.005) | m4.2.1.1 |
| Diet-induced obese model | Mouse [C57BL/6J (diet-induced obese)] | Subcutaneous | A1, rHA | No | HGS | RD2005/01150 (FS29900.DBS.0.006) | m4.2.1.1 |
| Effect on serum insulin (single administration) | Mouse (db/db)<br>Rat (ZDF or lean heterozygous controls) | Subcutaneous<br>Intravenous | A1, rHA, Ex-4 | No | HGS | RD2005/01152 (FS29900.DBS.0.032) | m4.2.1.1 |
| Effect on serum insulin (single administration) | Monkey (cynomolgus) | Subcutaneous | A1, Exenatide | No | GSK (b) (4) | RH2005/00081<br>RD2005/00955[g] (7274-902) | m4.2.1.1 |
| Effect on glucose disposal | Monkey (cynomolgus) | Subcutaneous (once weekly for 4 weeks) | A1, Ex-4 | No | | RD2005/01285 (6962-131) | m4.2.3.7.1 |
| Effect on gastric emptying | Mouse [C57BL/6J (diet-induced obese)] | Subcutaneous | A1, rHA, IL-1β | No | HGS | RD2005/01151 (FS29900.DBS.0.007) | m4.2.1.1 |
| Effect on gastrointestinal transit | Mouse (CD-1 or db/db) | Subcutaneous | A1, rHA, Ex-4 | No | HGS | RD2005/01153 (FS29900.DBS.0.033) | m4.2.1.1 |

**Table 1.1 (Continued) List of Primary Pharmacodynamic Studies Performed with Albiglutide**

| Type of Study | Species (Strain)[a]/ Test System | Method of Administration[b] | Form[c] | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|
| Effect on food consumption and weight | Mouse (SCID; normal and high fat fed) | Subcutaneous (3 times weekly for 21 days, then every other day for 24 days) | A1, rHA | No | HGS | RD2005/01154 (FS29900.DBS.0.034) | m4.2.1.1 |
| Weight loss in a diet-induced obese model | Mouse [C57BL/6J (diet-induced obese)] | Subcutaneous (daily for 3 days) | A1, Exenatide | No | GSK | RH2008/00014 | m4.2.1.1 |
| Effect on pancreatic β-cell mass | Rat (STZ-induced diabetic neonatal) | Subcutaneous (daily from Days 2-6 after birth) | A1, rHA, Ex-4 | No | HGS<br>GSK | RD2005/01157 (FS29900.DBS.0.031)<br>RJ2005/00003 | m4.2.1.1 |
| Efficacy comparison of 2 manufacturing processes | Mouse (db/db) | Subcutaneous | A1, A2 | No | GSK | RH2008/00015 | m4.2.1.1 |

**Key:**
cAMP = Cyclic adenosine monophosphate. DPP-IV = Dipeptidyl peptidase IV. GLP-1 = Glucagon-like peptide 1.
GLP-1R = Glucagon-like peptide 1 receptor. HEK = Human embryonic kidney. IL-1β = Recombinant human interleukin-1β.
INS-1 = Insulinoma cell line.

**Testing Facility:** (b) (4)
GSK = GlaxoSmithKline.
HGS = Human Genome Sciences, Inc.

a = C57BL/6J (diet-induced obese) = Wild-type mice fed a 45%kcal diet fat diet for 17 weeks to induce obese type 2 diabetic model; db/db = db/dbBLKs/J (C57/BL/6J-m+/+Lepr db) pre-diabetic or type 2 diabetic mice (age-dependent); SCID = severe-combined immunodeficient mice (used to reduce immunogenicity); STZ-induced diabetic neonatal rats = diabetogenic agent STZ administered intraperitoneally on day of birth to Wistar rats pups; ZDF = Zucker diabetic fatty rats, a type 2 diabetic model; CD-1 = Normoglycemic.
b = Single dose administered unless otherwise specified.
c = For albiglutide, indicates by which production process albiglutide was manufactured: A1 = Process 1 and A2 = Process 2. Ex-4 = Exendin-4. Exenatide = Synthetic exendin-4. GLP-1 = Glucagon-like peptide 1. L = Liraglutide. Met = Metformin (antihyperglycemic agent). rHA = Recombinant human albumin.
d = Fused to rHA.
e = Clonal variant of the rat insulinoma cell line that can secrete insulin in a glucose-dependent manner.
f = Original construct (2803) and a new construct (3070) in which the human serum albumin signal peptide was replaced with the yeast killer toxin signal peptide.
g = Report for in-life phase only.

**Table 1.2 List of Secondary Pharmacodynamic Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | Method of Administration | Form[a] | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|
| Effects on cardiac glucose metabolism | Rat (Sprague Dawley) | Subcutaneous (daily for 3 days) | A2 | No | GSK | CH2010/00002 | m4.2.1.2 |
| Cardioprotective effects | Rat (Sprague Dawley) | Subcutaneous (daily for 3 days) | A2 | No | GSK | CH2009/00075 | m4.2.1.2 |
| Effects on cardiac energetics | Rat (Sprague Dawley) | Subcutaneous (daily for 3 days) | A2 | No | GSK | 2010N110942 | m4.2.1.2 |
| Effects on cardiac carbohydrate metabolism | Rat (Sprague Dawley) | Subcutaneous (daily for 3 days) | A2 | No | GSK | 2010N110941 | m4.2.1.2 |

**Key:**
a = For albiglutide, indicates by which production process albiglutide was manufactured: A2 = Process 2.

**Testing Facility:**
GSK = GlaxoSmithKline.

**Table 1.3 List of Safety Pharmacology Studies Performed with Albiglutide**

| Type of Study | Species (Strain) | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Neurobehavioral | Monkey (cynomolgus) | 3-5M | Subcutaneous | A1 | 2.5, 7.5, 25 | Twice weekly for 5 weeks[b] | Yes | GSK | CD2005/00839 (G05072) | m4.2.3.2 |
| Cardiovascular/ respiratory | Monkey (cynomolgus) | 3M | Subcutaneous | A1 | 25 | Single | Yes | GSK | CD2005/00546 (G05162) | m4.2.1.3 |

**Key:**
a = For albiglutide, indicates by which production process albiglutide was manufactured: A1 = Process 1.
b = Neurobehavioral assessment performed on Days 8 and 12.

**Testing Facility:**
GSK = GlaxoSmithKline.

## 1. PHARMACOKINETICS: OVERVIEW FOR ALBIGLUTIDE

**Table 1.1 Listing of Absorption Studies with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) or Concentration | Duration of Dosing (Sampling Occasions) | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Pharmacokinetic | Mouse (db/db or C57BL/6) | 21-30F | Intravenous Subcutaneous | A1 | 1 1, 2, 6.5 | Single (up to 144 hours post dose) | No | HGS | RD2005/01136 (FS29900.DBS.0.004) | m4.2.2.2 |
| Pharmacokinetic | Mouse (db/db or C57BL/6) | 22F (C57BL/6) 22F (C57BL/6) 18F (db/db) | Subcutaneous | A1 A-M A-M | 8 | Single (up to 78 hours post dose) | No | HGS | RD2005/01137 (FS29900.DBS.0.030) | m4.2.2.2 |
| Pharmacokinetic | Mouse (CD-1) | 80M | Subcutaneous | A1, A2 | 6.5 | Single (up to 96 hours post dose) | No | GSK | RD2008/00270 (08DMR019) | m4.2.2.2 |
| Toxicokinetic | Mouse (CD-1) | 15M | Subcutaneous | A1 | 5, 15, 50 | Daily for 7 days (Day 7) | Yes | (b) (4) | CD2007/01279 (G06493) | m4.2.3.5.1 |
| Toxicokinetic | Mouse (CD-1) | 6 or 15F | Subcutaneous | A1 | 1, 5, 50 | Daily for 7 days (Days 1, 7 and 14) | Yes | | CD2006/01620 (G06076) | m4.2.3.2 |
| Toxicokinetic | Mouse (CD-1) | 15-21M/ 15-21F | Subcutaneous | A1 | 5, 50, 150 | Daily for 7 or 15 days (Days 1, 7 and 15) | Yes | | RD2005/01134 (6962-139) | m4.2.3.2 |
| Toxicokinetic | Rat (Sprague Dawley) | 6M 6M[R] | Subcutaneous | A1 | 5, 50 | Daily for 14 days (Days 1, 7, 8, 15, 16 and 22) | No | GSK | RD2005/00575 (R41642) | m4.2.3.7.1 |

**Table 1.1 (Continued) Listing of Absorption Studies with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) or Concentration | Duration of Dosing (Sampling Occasions) | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Pharmacokinetic | Monkey (cynomolgus) | 3M/3F | Intravenous Subcutaneous | A1 | 1 1, 10 | Single (up to 144 hours post dose) | No | GSK (b) (4) | RD2005/01127 (05DMR099) RH2005/00101 (7274-902) | m4.2.2.2 |
| Pharmacokinetic | Monkey (cynomolgus) | 4M | Subcutaneous | A1, A2 | 15 | Single (up to 168 hours post dose) | No | GSK | CD2008/01656 (08DMM085) | m4.2.2.2 |
| Toxicokinetic | Monkey (cynomolgus) | 2M/2F | Subcutaneous | A1 | 0.1, 1, 10 | Weekly for 4 weeks (Days 1 and 22) | No | HGS (b) (4) | RD2005/01285 (6962-131) | m4.2.3.7.1 |
| Toxicokinetic | Monkey (cynomolgus) | 5M/5F | Subcutaneous | A1 | 5, 15, 50 | Weekly for 4 weeks (Days 1 and 22) | No | (b) (4) | RD2005/01133 (N104501) | m4.2.3.2 |
| Toxicokinetic | Monkey (cynomolgus) | 3-5M/ 3-5F | Subcutaneous | A1 | 5, 15, 50[b] | Weekly for 5 weeks (Days 1 and 32) | Yes | GSK (b) (4) | CD2005/00839 (G05072) | m4.2.3.2 |
| Toxicokinetic | Monkey (cynomolgus) | 4M/4F | Subcutaneous | A1 | 1 | Weekly for 18 weeks[c] (Days 1, 15, and 127) | No | | RD2005/01284 (6962-147) | m4.2.3.7.1 |
| Toxicokinetic | Monkey (cynomolgus) | 4-6M/4-6F | Subcutaneous | A1 | 1, 5, 15 | Weekly for 26 weeks (Day 1, Weeks 2, 13 and 26) | Yes | GSK | RD2005/01281 (P41726) | m4.2.3.2 |

**Table 1.1 (Continued) Listing of Absorption Studies with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) or Concentration | Duration of Dosing (Sampling Occasions) | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Toxicokinetic | Monkey (cynomolgus) | 5M/5F | Subcutaneous | A2 | 15, 50 | Weekly for 52 weeks (Weeks 2, 26 and 52) | Yes | (b) (4) | CD2009/00863 (G07396) | m4.2.3.2 |

**Key:**
A-M = Glucagon-like pepetide-1 (GLP-1) fused with mouse serum albumin.
R = Additional animals added to evaluated off-treatment period.
a = For albiglutide, indicates by which production process albiglutide was manufactured: A1 = Process 1 and A2 = Process 2.
b = Administered twice weekly as 2 equally divided doses.
c = Three cycles whereby animals were dosed for 3 weeks, followed by a 3 week washout and a subsequent final dose.

**Testing Facility:**
(b) (4)
GSK = GlaxoSmithKline.
HGS = Human Genome Sciences, Inc.
(b) (4)

**Table 1.2 Listing of Metabolism Studies with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) or Concentration | Duration of Dosing (Sampling Occasions) | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Cytochrome P450 induction (mRNA expression) | Monkey (cynomolgus) | 2-3M/ 2-3F | Subcutaneous | A1 | 5, 15, 50 | Weekly for 5 weeks (Week 5) | No | GSK | RD2005/01022 (05DMR078) | m4.2.2.4 |
| Cytochrome P450 induction (catalytic activity) | Monkey (cynomolgus) | 2-3M/ 2-3F | Subcutaneous | A1 | 5, 15, 50 | Weekly for 5 weeks (Week 5) | Yes | (b) (4) | RD2005/00954 (7274-910) | m4.2.2.4 |

**Key:**
a = For albiglutide, indicates by which production process albiglutide was manufactured: A1 = Process 1.

**Testing Facility:**
COV = Covance Laboratories.
(b) (4)

# 1. TOXICOLOGY: OVERVIEW FOR ALBIGLUTIDE

**Table 1.1 List of Single Dose Toxicity Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg/day) or Concentration | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Single dose | Mouse (CD-1) | 3M/3F | Subcutaneous Intravenous | A1 | 50, 200, 500, 1000 500 | Single | No | HGS | RD2005/01162 (FS29900.DBS.0.013) | m4.2.3.1 |

**Key:**
a = Indicates by which production process albiglutide was manufactured: A1 = Process 1.

**Testing Facility:**
HGS = Human Genome Sciences, Inc.

**Table 1.2 List of Repeat Dose Toxicity Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg) or Concentration | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| 7 day<br>14 day | Mouse (CD-1) | 15F<br>6F | Subcutaneous | A1 | 1, 5, 50 | Daily for 7 or 14 days | Yes | (b) (4) | CD2006/01620 (G06076) | m4.2.3.2 |
| 7 or 15 day | Mouse (CD-1) | Phase 1: 30M/30F; 21M/21F[TK]<br>Phase 2: 10M/10F; 15M/15F[TK] | Subcutaneous | A1 | 5, 50, 150 | Daily for 7 or 15 days | Yes[b] | | RD2005/01134 (6962-139) | m4.2.3.2 |
| 4 week | Monkey (cynomolgus) | 3M/3F<br>2M/2F[R] | Subcutaneous | A1 | 5, 15, 50 | Weekly for 4 weeks | Yes[b] | | RD2005/01133 (N104501) | m4.2.3.2 |
| 5 week | Monkey (cynomolgus) | 3M/3F<br>3M/3F; 2M/2F[R] | Subcutaneous | A1 | 5, 15<br>50 | Weekly for 5 weeks[c] | Yes | GSK | CD2005/00839 (G05072) | m4.2.3.2 |
| 26 week | Monkey (cynomolgus) | 4M/4F<br>4M/4F; 2M/2F[R] | Subcutaneous | A1 | 1, 5<br>15 | Weekly for 26 weeks | Yes | (b) (4) | RD2005/01281 (P41726) | m4.2.3.2 |
| 52 week | Monkey (cynomolgus) | 5M/5F[d] | Subcutaneous | A2 | 15, 50 | Weekly for 52 weeks | Yes | | CD2009/00863 (G07396) | m4.2.3.2 |

**Key:**
R = Additional animals included to evaluate an off-treatment period.
TK = Additional animals included for toxicokinetic evaluation only.
a = Indicates by which production process albiglutide was manufactured: A1 = Process 1 and A2 = Process 2.
b = In-life portion was GLP. Immunogenicity and TK were non-GLP.
c = Administered as twice weekly doses of 2.5, 7.5 or 25 mg/kg/dose.
d = An additional group (n=5/sex) received exenatide twice daily (BID) at 25 μg/kg/dose from Day 1 through Day 14 and 75 μg/kg/dose beginning on Day 15 through the remainder of the study.

**Testing Facility:**
(b) (4)
GSK = GlaxoSmithKline.
(b) (4)

**Table 1.3 List of Reproductive and Developmental Toxicity Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg/day) or Concentration | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Female fertility | Mouse (CD-1) | 25F | Subcutaneous | A1 | 1, 5, 50 | ~14 days (7 days before cohabitation until positive evidence of mating) | Yes | (b) (4) | CD2006/01879 (G05438) | m4.2.3.5.1 |
| Male mating fertility | Mouse (CD-1) | 25M | Subcutaneous | A1 | 5, 15, 50 | 15 to 17 days (7 days before and through cohabitation) | Yes | | CD2007/01279 (G06493) | m4.2.3.5.1 |
| Early embryonic to implantation | Mouse (CD-1) | 22F | Subcutaneous | A1 | 1, 5, 50 | 7 days (Days 0 to 6 pc) | Yes | | CD2006/01198 (G06019) | m4.2.3.5.1 |
| Embryofetal development | Mouse (CD-1) | 22F | Subcutaneous | A1 | 1, 5, 50 | 10 days (Days 6 to 15 pc) | Yes | | CD2006/01353 (G06020) | m4.2.3.5.2 |
| Pre- and post-natal (Part 1) | Mouse (CD-1) | 24F | Subcutaneous | A2 | 1, 5, 50 | 12 days (Days 6 to 17 pc) | Yes | | 2011N117859 (G11082) | m4.2.3.5.3 |
| Pre- and post-natal (Part 2) (w/maternal functions investigation) | Mouse (CD-1) | 24F 35F | Subcutaneous | A1 | 1, 5, 50 5 | 14 days (Day 15 pc to Day 10 pp) | Yes | | CD2008/01156 (G07387)/(I08262) | m4.2.3.5.3 |
| Pre- and post-natal (Part 3) | Mouse (CD-1) | 24F | Subcutaneous | A2 | 1, 5, 50 | 15 days (Days 7 to 21 pp) | Yes | | 2010N104536 (G10125) | m4.2.3.5.3 |
| Pre- and post-natal (Part 3 repeat) | Mouse (CD-1) | 34F | Subcutaneous | A2 | 1, 5, 50 | 15 days (Days 7 to 21 pp) | Yes | | 2011N119858 (G11059) | m4.2.3.5.3 |

**Key:**
pc = Post coitum. pp = Post partum.
a = Indicates by which production process albiglutide was manufactured: A1 = Process 1 and A2 = Process 2.

**Testing Facility:** (b) (4)

**Table 1.4 List of Other Toxicity Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg)/ Concentration | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Epitope content | Epibase® immunoprofiling | NA | In silico | A2, (b) (4) | NA | NA | No | (b) (4) | 2012N143627 2012N143628 | m4.2.3.7.1 |
| Immunogenicity | Mouse (CD-1) | 20F | Subcutaneous | A1 | 2.25, 7.5 | 3 times weekly for 4 weeks | No | HGS | RD2005/01167 (FS29900.DBS.0.012) | m4.2.3.7.1 |
| Immunogenicity | Mouse (db/db) | 21M | Subcutaneous | A1 | 0.8 | 3 times weekly for 10 weeks[b] | No | HGS | RD2005/01168 (FS29900.DBS.0.029) | m4.2.3.7.1 |
| Immunogenicity | Mouse (CD-1) | 10M | Subcutaneous | A1 | 0.8 | 3 times weekly for 15 weeks[c] | No | HGS | RD2005/01135 (FS29900.DBS.0.028) | m4.2.3.7.1 |
| Immunogenicity | Rat (Sprague Dawley) | 6M 6M[R] | Subcutaneous | A1 | 5, 50 | Daily for 14 days | No | GSK | RD2005/00575 (R41642) | m4.2.3.7.1 |
| Immunogenicity | Monkey (cynomolgus) | 2M/2F | Subcutaneous | A1 | 0.1, 1, 10 | 4 weeks (Dosed on Days 1, 8, 15, 22, 26 and 29) | No | (b) (4) | RD2005/01285 (6962-131) | m4.2.3.7.1 |
| Immunogenicity | Monkey (cynomolgus) | 4M/4F | Subcutaneous | A1 | 1 | Weekly for 18 weeks[d] | No | | RD2005/01284 (6962-147) | m4.2.3.7.1 |
| Neutralizing activity of anti-albiglutide antibodies[e] | Monkey (cynomolgus) | 4M/4F | Subcutaneous | A1 | 1, 5, 15 | Weekly for 5 weeks | No | GSK | CD2006/01889 (I06428) | m4.2.3.7.7 |

**Table 1.4 (Continued) List of Other Toxicity Studies Performed with Albiglutide**

| Type of Study | Species (Strain)/ Test System | No./Sex/ Group | Method of Administration | Form[a] | Dose (mg/kg)/ Concentration | Duration of Dosing | GLP | Testing Facility | Report No. (Study No.) | Location in CTD |
|---|---|---|---|---|---|---|---|---|---|---|
| Effects on monkey pancreas using stereologic methods[f] | Monkey (cynomolgus) | 5M/5F | Subcutaneous | A2 | 50 | Weekly for 52 weeks | No | GSK | 2011N126413 (I11223) | m4.2.3.7.7 |

Key:
NA = Not applicable. R = Additional animals included to evaluate an off-treatment period.
a = Indicates by which production process albiglutide was manufactured: A1 = Process 1; A2 = Process 2 and (b) (4) (b) (4)
b = Two cycles whereby animals were dosed for 4 weeks followed by a one week washout.
c = Three cycles whereby animals were dosed for 4 weeks followed by a one week washout.
d = Three cycles whereby animals were dosed for 3 weeks, followed by a 3 week washout and a subsequent final dose.
e = Serum samples collected from 26 week monkey study [Report RD2005/01281].
f = Pancreas samples collected from 52 week monkey study [Report CD2009/00863].

Testing Facility: (b) (4)
GSK = GlaxoSmithKline.
HGS = Human Genome Sciences, Inc.

## 3.2 Studies Not Reviewed

The following validation reports were cursorily reviewed and judged to be fit for purpose, but were not subjected to rigorous review:

### 2. ANALYTICAL METHODS AND VALIDATION REPORTS

**Table 2.1 Pharmacokinetics: Analytical Methods and Validation Reports**

| Type of Study | Species | Validated Concentration Range or Dilution Ratio | Report No. (Study/Reference No.) | Location in CTD |
|---|---|---|---|---|
| **Assays for Albiglutide** | | | | |
| Qualification of the ELISA for the quantitation of Albugon in mouse plasma | Mouse | 93.8 to 3125 ng/mL | 2012N146511 (TR-22-04-013) | m4.2.2.1 |
| Quantitative ELISA method partial validation for the determination of GSK716155 in mouse plasma | Mouse | 5.00 to 320 ng/mL | CD2006/00649 (M08.GLP1.mop.1) | m4.2.2.1 |
| The validation of a method for the determination of GSK716155 in mouse plasma (range 5.00 - 250.00 ng/mL) using a chemiluminescent immunoassay | Mouse | 5.00 to 250 ng/mL | CD2008/00752 (GSK716155MOPLVALD) | m4.2.2.1 |
| Quantitative ELISA method validation for the determination of GSK716155 in rat plasma | Rat | 5.00 to 320 ng/mL | CD2005/00943 (M08.GLP1.rtp.1) | m4.2.2.1 |
| Qualification of the ELISA for the quantitation of Albugon in monkey plasma | Monkey | 50.0 to 2000 ng/mL | 2012N146513 (TR-22-04-001) | m4.2.2.1 |
| Quantitative ELISA method partial validation for the determination of GSK716155 in monkey plasma | Monkey | 5.00 to 320 ng/mL | CD2005/01029 (M08.GLP1.mkp.1) | m4.2.2.1 |
| The validation of a method for the determination of GSK716155 in cynomolgus monkey plasma (range 1.00 – 25.00 μg/mL) using a chemiluminescent immunoassay | Monkey | 1.00 to 25.00 μg/mL | CD2008/00838 (GSK716155CYPLVALB) | m4.2.2.1 |

**Table 2.1 (Continued) Pharmacokinetics: Analytical Methods and Validation Reports**

| Type of Study | Species | Validated Concentration Range or Dilution Ratio | Report No. (Study/Reference No.) | Location in CTD |
|---|---|---|---|---|
| **Assay for Exendin-4** | | | | |
| The validation of a method for the determination of exendin-4 in monkey plasma (range 20 to 2000 ng/mL) using UHPLC-MS/MS | Monkey | 20 to 20,000 ng/mL | CD2008/01648 (Exendin 4CYPLVALA) | m4.2.2.1 |
| **Assays for Anti-Albiglutide Antibodies** | | | | |
| Validation of a method to measure antibodies against GSK716155 in serum of CD-1 mice | Mouse | 1:10 | CD2008/01495 (M08276) | m4.2.2.1 |
| ELISA for screening of antibodies against GSK716155 in rat serum | Rat | 1:15 | RD2005/01396 | m4.2.2.1 |
| ELISA for confirmation and titration of antibodies against GSK716155 in rat serum | Rat | 1:15 | RD2005/01397 | m4.2.2.1 |
| ELISA for screening of antibodies against GSK716155 in monkey serum | Monkey | 1:30 | RD2005/01394 | m4.2.2.1 |
| ELISA for confirmation and titration of antibodies against GSK716155 in monkey serum | Monkey | 1:30 | RD2005/01395 | m4.2.2.1 |
| Assay qualification report for anti-GSK716155 antibody detection in monkey serum samples by an ELISA | Monkey | 1:30 | RD2005/01393 | m4.2.2.1 |

**Key:** ELISA = Enzyme-linked immunosorbent assay. UHPLC-MS/MS = Ultra high performance liquid chromatography with tandem mass spectrometry.

### 3.3 Previous Reviews Referenced

1. Pharmacology/Toxicology IND Review 1, Initial IND Review, by Dr. Dylan Yao, 15-FEB-2006.
2. Pharmacology/Toxicology IND Review 2, Female Fertility & Embryofetal Development in Mice, and a 26-Week Monkey Study, by Dr. Dylan Yao, 21-FEB-2007.
3. Pharmacology/Toxicology IND Review 3, Male Fertility in Mice, by Dr. Dylan Yao, 13-MAY-2008.
4. Pharmacology/Toxicology IND Review 4, 52-Week Monkey Study, by Dr. Ronald Wange, 19-JUL-2010.
5. Pharmacology/Toxicology IND Review 5, Pre- & Post-Natal Development Study in Mice, by Dr. Ronald Wange, 17-SEP-2010.

# 4 Pharmacology

## 4.1 Primary Pharmacology

In brief:

- Albiglutide is an agonist at the GLP-1R and acts on pancreatic β-cells to augment glucose-dependent insulin secretion.
- A substitution of glycine for alanine at the N-terminus of GLP-1 renders the peptide resistant to proteolytic cleavage by dipeptidyl peptidase IV (DPP-IV).
- The human albumin moiety of the recombinant fusion protein, together with the DPP-IV resistance, greatly extends the half-life ($t_{½}$) from 1.5 to 5 minutes for native GLP-1 to 12 hours in mice and to 2 days in nonhuman primates for albiglutide.



Reference ID: 3347159

- Studies have shown that albiglutide stimulates cyclic adenosine monophosphate (cAMP) production via the GLP-1R and potentiates glucose-sensitive insulin secretion in an insulinoma cell line. Potency for stimulation of cAMP is reduced for albiglutide, compared to GLP-1 and exendin-4, with $EC_{50}$ values of 472, 51 and 3 pM, respectively.
- To establish the biological relevance of mice, rats, and monkeys for use in toxicity studies with albiglutide, HEK293T cells were engineered to express mouse, rat, monkey or human GLP-1R. cAMP production was then measured in response to varying concentrations of albiglutide. Comparable potency was seen at the rodent and human receptors, but albiglutide was ~3-fold less potent at the monkey GLP-1R.
- In vivo studies demonstrated that albiglutide lowers fasting plasma glucose (FPG), improves glucose tolerance in diabetic mouse and rat models, and increases glucose clearance and/or insulin secretion in monkeys.
- Albiglutide delays gastric emptying (GE) in normoglycemic mice and diabetic mice and reduces food intake in mice fed a normal or high fat diet. Additionally, albiglutide was shown to stimulate the recovery of β-cell mass following β cell damage induced by the diabetogenic agent, streptozotocin, in neonatal rat pups.
- In monkeys, albiglutide was biologically active and potentiated an insulin response to IV administered glucose.

Sponsor's tabulated summaries follow:

## 2. PRIMARY PHARMACODYNAMICS

**Table 2.1 In Vitro Primary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain)/ Test System | Concentration | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|
| Effect on GLP-1 receptor binding and intracellular cAMP / Comparison of cAMP-inducing activity | HEK293 cells expressing the human GLP-1R | Albiglutide (1.6 pM to 400 nM)<br>GLP-1 or Exendin-4 (0.15 to 3000 pM) | Albiglutide, GLP-1, and exendin-4 activated the GLP-1R, as evidenced by increased intracellular cAMP levels by ~7 fold, with $EC_{50}$ values of 472, 51, and 3 pM, respectively. | No | RD2005/01132 (FS29900.DBS.0.001)<br>RD2005/01161 (FS29900.DBS.0.035) |
| Action on GLP-1 receptor | HEK293 cells expressing the mouse, rat or human GLP-1R | Albiglutide, Process 2 (1 to 10 μM)<br>GLP-1, Exendin-4 or Liraglutide (1 to 100 nM) | Albiglutide had comparable potency on mouse, rat and human GLP-1R with average $pEC_{50}$ values of 7.17, 6.83 and 7.17, respectively. Exendin-4, liraglutide and GLP-1 were more potent than albiglutide on GLP-1R receptor, but had comparable activity across the species. The average $pEC_{50}$ values on mouse, rat and human GLP-1R were 8.54, 8.61 and 8.67, respectively, for exendin-4; 8.26, 8.35 and 8.55, respectively, for liraglutide; and 8.32, 8.50 and 8.61, respectively, for GLP-1. | No | 2012N146049 |
| Action on GLP-1 receptor | HEK293 cells expressing the monkey or human GLP-1R | Albiglutide or GLP-1 (0.0001 to 5400 nM) | The $EC_{50}$ values of albiglutide and GLP-1 in increasing intracellular cAMP levels were approximately 3- to 4-fold higher at the monkey GLP-1R than at the human GLP-1R, suggesting that monkeys may be less responsive than humans at similar doses. | No | RD2005/01148 (FS29900.DBS.0.008) |
| Resistance to proteolytic cleavage | Incubation with DPP-IV | Albiglutide or GLP-1, fused to rHA (10 μg) | After 60 minutes of incubation with DPP-IV, 87% of wild type GLP-1 fused to rHA was proteolyzed, while albiglutide was resistant to cleavage. | No | RD2005/01160 (FS29900.DBS.0.020) |
| Induction of insulin | INS-1 (832/13)[a] | Albiglutide or GLP-1 (0.128 to 2000 nM) | Incubation with albiglutide resulted in glucose-sensitive insulin secretion, although with less potency than GLP-1. | No | RD2005/01142 (FS29900.DBS.0.003) |

**Note:** Studies conducted with albiglutide manufactured from Process 1, unless otherwise noted.
**Key:** DPP-IV = Dipeptidyl peptidase IV. GLP-1 = Glucagon-like peptide 1. HEK = Human embryonic kidney. NA = Not applicable. rHA = Recombinant human albumin.
a = Clonal variant of the rat insulinoma cell line that can secrete insulin in a glucose-dependent manner.



Reference ID: 3347159

**Table 2.2 In Vivo Primary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain)[a] | Method of Administration[b] | Doses (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Effect on glucose tolerance | Mouse (db/db or heterozygote lean controls) | Subcutaneous | Albiglutide (0.075, 0.75, 7.5)<br>Exendin-4 (42 μg/kg) | 8-9[c] | Both albiglutide and exendin-4 significantly reduced fasting glucose in db/db mice, to levels of lean controls. After an OGTT at 1 or 3 hours after dosing, there was a significant reduction in glucose levels in db/db mice treated with 0.75 or 7.5 mg/kg albiglutide and 42 μg/kg exendin-4. | No | RH2005/00082 |
| Comparison of activity from 2 DNA constructs | Mouse (db/db) | Subcutaneous | Albiglutide constructs 2803 or 3070 (0.225, 0.75, 2.25, 7.5)<br>rHA (7.3, 22 and 73)<br>Exendin-4 (42 μg/kg) | 6M | After an OGTT on 2 consecutive days, mice dosed with DNA constructs 2803 or 3070 had significantly reduced glucose levels at all doses tested, and there no significant difference between the 2 constructs. | No | RD2005/01158 (FS29900.DBS.0.011) |
| Effect of single administration | Mouse (db/db) | Intravenous | Albiglutide (0.075, 0.15, 0.225, 0.45, 0.75)<br>rHA (0.066)<br>Exendin-4 (0.0042, 0.021, 0.042, 0.42, 4.2 μg/kg) | 8M | After an OGTT at 1 hour after dosing, albiglutide reduced blood glucose $AUC_{0-120}$ values in a dose-dependent manner, with the 0.45 mg/kg group achieving maximal clearance with a 50.1% reduction. In comparison, the most effective dose of exendin-4 (4.2 μg/kg) only resulted in a 38.3% reduction. $EC_{50}$ values were 0.09 mg/kg and 0.15 μg/kg for albiglutide and exendin-4, respectively. | No | RD2005/01159 (FS29900.DBS.0.022) |
| Duration of action on glucose tolerance (single administration) | Mouse (db/db or heterozygote lean controls) | Subcutaneous | Albiglutide (2.3, 7.5, 22.5)<br>Exendin-4 (42 μg/kg) | 7M | After an OGTT at 3 hours after dosing, albiglutide-induced reductions in glucose $AUC_{0-120}$ compared to vehicle were seen at up to 48 hours after dosing, whereas exendin-4 had little effect at 6 or 48 hours after dosing. | No | RD2005/01156 (FS29900.DBS.0.023) |

**Table 2.2 (Continued) In Vivo Primary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain)[a] | Method of Administration[b] | Doses (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Effect of repeated administration | Mouse (db/db) | Subcutaneous (daily for 6 days) | Albiglutide (2.25, 7.5, 12.8)<br>Exendin-4 (420 μg/kg)<br>rHA (20) | 5M | An OGTT was performed 2 hours after dosing on Days 1 and 2, and on Day 7. Albiglutide significantly increased glucose clearance in a dose-dependent manner compared to rHA on Days 1 and 2. Significant increases in clearance were seen in the 7.5 and 12.8 mg/kg albiglutide dose groups on Day 7 (24 hours after dosing) but not with exendin-4. | No | RD2005/01283 (FS29900.DBS.0.002) |
| Combination effect with metformin on glucose tolerance | Mouse (db/db) | Subcutaneous | Albiglutide (0.8, 2.25)<br>rHA (2)<br>Metformin (250; oral) | 8[c] | Although not statistically significant, the combination of 0.8 mg/kg albiglutide with metformin appeared to be as effective as 2.25 mg/kg albiglutide in glucose clearance after an OGTT, suggesting that combination therapy may produce a more potent effect. | No | RD2005/01155 (FS29900.DBS.0.036) |
| Effect on glucose tolerance (single administration) | Rat (ZDF or lean controls) | Intravenous | Albiglutide (0.3, 0.9, 3, 9)<br>rHA (2.7)<br>Exendin-4 (0.017, 0.17) | 4-6M | After an IPGTT at 3 or 24 hours after dosing, albiglutide improved glucose clearance compared to rHA controls at doses of 3.0 and 9.0 mg/kg, and 9.0 mg/kg, respectively. In contrast, exendin-4 had no effect on fasting glucose or glucose clearance. | No | RD2005/01149 (FS29900.DBS.0.005) |
| Diet-induced obese model | Mouse [C57BL/6J (diet-induced obese)] | Subcutaneous | Albiglutide (2.1, 5)<br>rHA (2.1) | 5M | Mice were fed a 45%kcal fat diet for 17 weeks and an OGTT was performed at 2 and 24 hours after dosing. Glucose $AUC_{0-120}$ values were significantly reduced in albiglutide-treated mice compared to vehicle at 2 and 24 hours after dosing. | No | RD2005/01150 (FS29900.DBS.0.006) |

**Table 2.2 (Continued) In Vivo Primary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain)[a] | Method of Administration[b] | Doses (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Effect on serum insulin (single administration) | Mouse (db/db) | Subcutaneous | Albiglutide (3, 30)<br>rHA (27)<br>Exendin-4 (1.7) | 5-6M | An OGTT was performed 6 hours after treatment (exception, exendin-4 was given immediately before). Albiglutide at both doses and exendin-4 significantly increased serum insulin levels compared to rHA. | No | RD2005/01152 (FS29900.DBS.0.032) |
| Effect on serum insulin (single administration) | Rat (ZDF or lean heterozygous controls) | Intravenous | Albiglutide (0.3, 0.9, 3)<br>rHA (2.7)<br>Exendin-4 (0.17) | 6-7M | An IPGTT was performed at 3 or 24 hours after treatment. Albiglutide at 3.0 mg/kg significantly increased serum insulin levels following glucose challenge compared to rHA at 3 hours after dosing; however, there was no significant increase in insulin 24 hours after dosing. | No | RD2005/01152 (FS29900.DBS.0.032) |
| Effect on serum insulin (single administration) | Monkey (cynomolgus) | Subcutaneous | Albiglutide (10)<br>Exenatide (5 μg/kg) | 8M/7F | Although albiglutide treatment did not enhance glucose clearance, it did significantly enhance insulin secretion and increase circulating C-peptide levels in response to an IVGTT. | No | RH2005/00081<br>RD2005/00955[d]<br>(7274-902) |
| Effect on glucose disposal | Monkey (cynomolgus) | Subcutaneous (once weekly for 4 weeks) | Albiglutide (0.1, 1, 10)<br>Exendin-4 (0.0025) | 2M/2F | After an IVGTT at 6 hours after dosing, monkeys treated with 10 mg/kg albiglutide had significantly reduced glucose $AUC_{0\text{-}120min}$ on Day 1, but not thereafter, and no difference in insulin AUC on Day 1 and 2. | No | RD2005/01285 (6962-131) |
| Effect on gastric emptying | Mouse [C57BL/6J (diet-induced obese)] | Subcutaneous | Albiglutide (5)<br>rHA (5)<br>IL-1β (0.01) | 6M | Pre-treatment with albiglutide 48 hours before IVGTT produced a significant retardation of gastric emptying (44.4%) compared to rHA-treated controls (67.3%) when fed 2 hours after dosing. | No | RD2005/01151 (FS29900.DBS.0.007) |

**Table 2.2 (Continued) In Vivo Primary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain)[a] | Method of Administration[b] | Doses (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Effect on pancreatic β-cell mass | Rat (STZ-induced diabetic neonatal) | Subcutaneous (daily from Days 2-6 after birth) | Albiglutide (3, 9, 30)<br>rHA (27)<br>Exendin-4 (1.7) | 48-60[e] | At 30 mg/kg albiglutide, β-cell mass was increased by 2.8- and 1.6-fold on Days 30 and 60, respectively, compared with rHA. Insulin staining intensity was slightly increased in sections from albiglutide-treated rats compared to exendin-4. Islet cell proliferation (Ki67-positive) levels were equivalent in the islets from the albiglutide and exendin 4 groups, and were increased over those seen in the rHA group. There was no evidence of apoptotic islet cells and no significant difference in the quantity of fibrous connective tissue staining in the islets in any of the experimental groups. | No | RD2005/01157 (FS29900.DBS.0.031)<br>RJ2005/00003 |
| Efficacy comparison of 2 manufacturing processes | Mouse (db/db) | Subcutaneous | Albiglutide (0.5, 2 for both Process 1 and Process 2) | Study 1: 10-12M<br>Study 2: 10-45M | Process 2 drug substance was as potent as Process 1 drug substance in decreasing the OGTT glucose AUC in db/db mice. | No | RH2008/00015 |

**Note:** Studies conducted with albiglutide manufactured from Process 1 unless otherwise noted.
**Key:** cAMP = Cyclic adenosine monophosphate. DPP-IV = Dipeptidyl peptidase IV. GTT = Glucose tolerance test [IP = Intraperitoneal. IV = Intravenous. O = Oral]. HEK = Human embryonic kidney. IL-1β = Recombinant human interleukin-1β. INS-1 = Insulinoma cell line. rHA = Recombinant human albumin.
a = C57BL/6J (diet-induced obese) = wild-type C57BL/6J mice were fed a 45%kcal diet fat diet for 17 weeks to induce an obese type 2 diabetic model; db/db = db/dbBLKs/J (C57/BL/6J-m+/+Lepr db) pre-diabetic or type 2 diabetic mice (age-dependent); SCID = severe-combined immunodeficient mice (used to reduce immunogenicity); STZ-induced diabetic neonatal rats = the diabetogenic agent STZ was administered intraperitoneally on the day of birth to Wistar rats pups; ZDF = Zucker diabetic fatty rats, a type 2 diabetic model; CD-1 = Normoglycemic.
b = Single dose administered unless otherwise noted.
c = Gender not specified.
d = Report for in-life phase only.
e = 6 litters/group with ~8-10 pups/litter (genders not specified).

## 4.2 Secondary Pharmacology

- In rats, albiglutide significantly reduced myocardial infarct size in a dose-dependent manner while improving post-ischemic cardiac left ventricular function following myocardial ischemia-reperfusion injury. In addition, pre-treatment with albiglutide improved post-ischemic cardiac function and energetics following myocardial ischemia/reperfusion injury.

**Table 2 Effect of GSK716155 on infarct size and left ventricle hemodynamics**

Data are n=8-10 per group and presented as mean±SEM. *p<0.05 vs vehicle; **p<0.01 vs vehicle by t-Test.

| | Vehicle | GSK716155 (1 mg/kg) | GSK716155 (3 mg/kg) | GSK716155 (10 mg/kg) |
|---|---|---|---|---|
| Infarct size (% of ischemic area) | 53.3±2.0 | 45.7±2.4* | 39.4±3.7** | 39.5±2.1** |
| Ischemic area (% of left ventricle) | 39.7±2.5 | 41.4±2.7 | 42.9±2.2 | 39.6±1.6 |
| Heart rate (bpm) | 408±14 | 386±9 | 412±8 | 409±12 |
| LVSP (mmHg) | 94±3 | 94±4 | 99±2 | 102±3 |
| LVEDP (mmHg) | 11±1 | 10±1 | 10±1 | 8±1** |
| $dP/dt_{max}$ (mmHg/s) | 5921±194 | 6112±434 | 7033±257** | 7461±475* |
| $dP/dt_{min}$ (mmHg/s) | 4924±297 | 4978±395 | 5436±224 | 6254±537 |

Data are mean±SEM, n=8-10/group. t-Test; *p<0.05 vs. vehicle ; **p<0.01 vs. vehicle

- In explanted perfused rat hearts, albiglutide was able to stimulate glucose uptake in an insulin independent manner, and improve the oxidative coupling of glucose metabolism.

Sponsor's tabulated summaries follow:

## 3. SECONDARY PHARMACODYNAMICS

**Table 3.1 Secondary Pharmacology Studies with Albiglutide**

| Type of Study | Species (Strain) | Method of Administration | Doses (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Effects on cardiac glucose metabolism | Rat (Sprague-Dawley) | Subcutaneous (daily for 3 days) | 10 | 13-16M | Treatment with albiglutide significantly increased in vivo glucose uptake by 70% (P<0.03). In the ex vivo Langendorff perfused heart study, albiglutide increased glucose uptake directly in the heart by 59% (P<0.05) while lactate efflux was decreased (55%, P<0.05). In summary, albiglutide was able to stimulate glucose uptake in the heart in an insulin independent manner, and improve the oxidative coupling of glucose metabolism. | No | CH2010/00002 |
| Cardioprotective effects | Rat (Sprague-Dawley) | Subcutaneous (daily for 3 days) | 1, 3, 10<br>rHA (2.7) | 8-10M | Albiglutide significantly reduced myocardial infarct size in a dose-dependent manner while improving post-ischemic cardiac left ventricular function following myocardial ischemia-reperfusion injury. The exact mechanism of the observed cardioprotection was not determined, but may be related to direct effects of GLP-1 receptor agonism on the myocardium and/or to indirect effects of increased insulin secretion. | No | CH2009/00075 |
| Effects on cardiac energetics | Rat (Sprague-Dawley) | Subcutaneous (daily for 3 days) | 10 | 8-10M | Pre-treatment with albiglutide improved post-ischemic cardiac function and energetics following myocardial ischemia/reperfusion injury. | No | 2010N110942 |
| Effects on cardiac carbohydrate metabolism | Rat (Sprague-Dawley) | Subcutaneous (daily for 3 days) | 10 | 8M | Albiglutide shift the heart toward a more energetically favorable substrate metabolism profile by increasing both glucose and lactate oxidation, suggesting that albiglutide may have direct therapeutic potential for improving cardiac energetics and function. | No | 2010N110941 |

**Note:** Studies conducted with albiglutide manufactured from Process 2.

## 4.3 Safety Pharmacology

- A single SC dose of 25 mg/kg albiglutide in monkeys had no effect on cardiovascular or respiratory function.
- There were no albiglutide-related effects on neurobehavioral function in monkeys administered twice weekly SC doses of ≤50 mg/kg/week albiglutide for 5 weeks.

Sponsor's tabulated summaries follow:

**4. SAFETY PHARMACOLOGY**

**Table 4.1 Safety Pharmacology Studies with Albiglutide**

| Organ Systems Evaluated | Species (Strain) | Method of Administration | Dose (mg/kg) | No./Sex/ Group | Noteworthy Findings | GLP Compliant | Report No. (Study No.) |
|---|---|---|---|---|---|---|---|
| Central and peripheral effects | Monkey (cynomolgus) | Subcutaneous (twice weekly for 5 weeks)[a] | 2.5, 7.5, 25 | 3-5M | There were no albiglutide-related effects on neurobehavioral functional assessments (normal central and peripheral nervous system activity) after albiglutide treatment. | Yes | CD2005/00839 (G05072) |
| Respiratory and cardiovascular systems | Monkey (cynomolgus) | Subcutaneous (single) | 25 | 3M | Albiglutide had no effect on arterial blood pressures (mean arterial pressure, systolic blood pressure, diastolic blood pressure and pulse pressure), heart rate, electrocardiographic intervals (PR, QRS, QT and QTc), ventilatory function (tidal volume, respiratory rate and minute volume), a measure of airway resistance (total pulmonary resistance) and electrocardiographic waveforms when monitored for up to 168 hours. | Yes | CD2005/00546 (G05162) |

**Note:** Studies conducted with albiglutide manufactured from Process 1.
a = Neurobehavioral assessment performed on Days 8 and 12.

# 5 Pharmacokinetics/ADME/Toxicokinetics

## 5.1 PK/ADME

### ABSORPTION

- After intravenous dosing in mice and monkeys, the total clearance and volume of distribution of albiglutide were low and the $t_{1/2}$ was approximately 8 hours and 60 hours in mouse and monkey, respectively. Following subcutaneous dosing the absolute bioavailability was at least 38% in mouse and 50% in monkey with a similar apparent $t_{1/2}$ to those observed after intravenous dosing with mouse being slightly longer at 12 hours. There were no substantial sex-related differences in systemic exposure and pharmacokinetic parameters.
- Following single subcutaneous administration of albiglutide the mean systemic exposure ($AUC_{0-\infty}$) in mice increased in proportion to dose (1 to 6.5 mg/kg), but in monkeys a 10-fold change in dose (1 to 10 mg/kg) led to a 16-fold increase in exposure.
- Accumulation of albiglutide was observed after repeated subcutaneous administration in mice and monkeys and was generally consistent with the $t_{1/2}$ and frequency of dosing. However, in some animals, there was a reduction in the



Reference ID: 3347159

anticipated plasma concentrations due to the presence of anti-albiglutide antibodies which were observed as early as Day 8 in mice and rats and Day 15 in monkeys. After 52 weekly doses of albiglutide to monkeys at 50 mg/kg, the mean $C_{max}$ and $AUC_{0\text{-}168\ hours}$ for the final dosing interval were 538 µg/mL and 56,100 µg.h/mL, respectively.

**Table 9.1 Comparative Assessment of Systemic Exposure after Subcutaneous Administration of Albiglutide to Mice and Monkeys**

| Study Type (Report No.) | Dose[a] (mg/kg) | Sex | $C_{max}$ (µg/mL)[b] | $AUC_{0\text{-}168}$ (µg.h/mL)[b] | Multiples of Human Exposure | |
|---|---|---|---|---|---|---|
| | | | | | $C_{max}$ | $AUC_{0\text{-}168}$ |
| Mouse 7 day (CD2006/01620) | 1 | F | 4.38 | 470[d] | 0.93 | 0.63 |
| | 5 | F | 13.8 | 1652[d] | 2.9 | 2.2 |
| | **50** | **F** | **239** | **28917[d]** | **51** | **39** |
| Mouse 7 day Male mating fertility (CD2007/01279) | 5 | M | 18.1 | 2408[e] | 3.9 | 3.2 |
| | 15 | M | 46.0 | 6412[e] | 9.8 | 8.5 |
| | 50 | M | 185 | 25207[e] | 39 | 34 |
| Monkey 4 week (RD2005/01133) | 5 | M+F | 36.1 | 2912[f] | 7.7 | 3.9 |
| | **15** | **M+F** | **105** | **7820[f]** | **22** | **10** |
| | 50 | M+F | 505 | 43336[f] | 107 | 58 |
| Monkey 5 week[c] (CD2005/00839) | 5 | M+F | 17.5 | 1057[g] | 3.7 | - |
| | 15 | M+F | 71.2 | 4350[g] | 15 | - |
| | 50 | M+F | 260 | 15312[g] | 55 | - |
| Monkey 26 week (RD2005/01281) | 1 | M+F | 6.03 | 591 | 1.3 | 0.79 |
| | 5 | M+F | 32.0 | 3316 | 6.8 | 4.4 |
| | **15** | **M+F** | **121** | **13143** | **26** | **18** |
| Monkey 52 week (CD2009/00863) | 15 | M+F | 155 | 16050 | 33 | 21 |
| | **50** | **M+F** | **538** | **56100** | **114** | **75** |
| Clinical[h] | 50 mg weekly | M+F | 4.7 | 751 | - | - |

**Key:** No observed adverse effect level (NOAEL) is **bolded**.
– = Not applicable.
a = Administered daily to mice and weekly to monkeys unless otherwise noted.
b = End of study values unless otherwise noted. For comparative and interpretive purposes, values were converted to µg/mL ($C_{max}$) and µg.h/mL (AUC) and $AUC_{0\text{-}168}$ values extrapolated where necessary.
c = Administered as twice weekly doses of 2.5, 7.5 or 25 mg/kg/dose.
d = Data presented in original report as $AUC_{0\text{-}24}$ [m2.6.7, Table 7.1].
e = Data presented in original report as $AUC_{0\text{-}24}$ [m2.6.7, Table 12.2].
f = Data presented in original report as ng.day/mL [m2.6.5, Table 4.6].
g = $AUC_{0\text{-}72}$. Since albiglutide was administered twice weekly rather than once weekly, comparison to human $AUC_{0\text{-}168}$ was not assessed.
h = Estimated steady-state exposures in patients from Phase III clinical trials receiving the maximum proposed therapeutic dose of 50 mg/week albiglutide [m2.5, Section 3.2].

## DISTRIBUTION

In accordance with current guidance, tissue distribution studies were not conducted with albiglutide.



Reference ID: 3347159

**METABOLISM**

In accordance with current guidance, studies to establish the metabolic fate of albiglutide were not conducted. It is expected that albiglutide will be eliminated by catabolism to its component amino acids. While classical biotransformation studies, as performed for pharmaceuticals, were not conducted, CYP enzyme induction potential was investigated in monkeys.

- Minimal increases in CYP mRNA levels were observed in male (CYP1A2) and female (CYP2B17) monkeys following subcutaneous administration of albiglutide at 5, 15, or 50 mg/kg/week for 5 weeks, although the increases were not dose-dependent. There was little or no effect on hepatic microsomal protein, total CYP content, or the activity of CYP1A, CYP2B, CYP2E, CYP3A, and CYP4A.

**EXCRETION**

In accordance with current guidance, excretion studies were not conducted with albiglutide.

### 5.2 Toxicokinetics

*See toxicity studies.*

## 6 General Toxicology

### 6.1 Single-Dose Toxicity

Report No. RD2005/01162 (Study No. FS29900.DBS.0.013)

Methods:

CD-1 mice received single doses of albiglutide via either the SC or IV route (3/s/g) at 0, 50, 200, 500 and 1000 mg/kg for SC and 0 or 500 mg/kg for IV, and were followed for 7 days.

Results: Transient, dose-dependent body weight drop (max. ↓11-15%) in both male and female mice at SC doses ≥200 mg/kg and the 500 mg/kg IV dose, consistent with intended pharmacological activity.

Microscopic findings were observed in the epididymis (vacuolation and cell debris), seminal vesicle (vacuolation), and prostate (vacuolation and lymphocyte infiltration) of male mice at 500 mg/kg IV and in the epididymis (vacuolation and cell debris) at doses ≥ 500 mg/kg SC. The histopathology changes were more pronounced with the IV dose. The vacuoles were characterized as 1 or more small discrete intracytoplasmic vacuoles within the epithelium of the affected tissues.



Reference ID: 3347159

**Table 9 Incidence of microscopic findings in the epididymis, seminal vesicle, and prostate of male mice that may be attributed to the administration of Albugon**

| Treatment<br>Dose Level<br>Route of Administration | Vehicle<br>0 mg/kg<br>SC | Albugon<br>50 mg/kg<br>SC | Albugon<br>200 mg/kg<br>SC | Albugon<br>500 mg/kg<br>SC | Albugon<br>1000 mg/kg<br>SC | Albugon<br>0 mg/kg<br>IV | Albugon<br>500 mg/kg<br>IV |
|---|---|---|---|---|---|---|---|
| Microscopic finding | | | | | | | |
| Tissue/finding | | | | | | | |
| Epididymis | | | | | | | |
| Vacuolation | 0 / 3 | 0 / 3 | 0 / 3 | 2 / 3 | 1 / 3 | 0 / 3 | 3 / 3 |
| Cell debris, intraluminal | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 1 / 3 | 0 / 3 | 0 / 3 |
| Seminal vesicle | | | | | | | |
| Vacuolation | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 3 / 3 |
| Prostate | | | | | | | |
| Vacuolation | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 0 / 3 | 1 / 3 |

For SC administration, the NOAEL = 200 mg/kg for males and 1000 mg/kg for females. For IV administration, the NOAEL was <500 mg/kg for males and >500 mg/kg for females.

## 6.2 Repeat-Dose Toxicity

**Study title: 2-week subcutaneous injection toxicity and toxicokinetic study in mice with a 1-week interim necropsy and a 1-week recovery period**

Study no.: 6962-139 (b) (4)
Study report location: m4.2.3.2
Conducting laboratory and location: (b) (4)
Date of study initiation: 9-JAN-2003
GLP compliance: yes
QA statement: yes
Drug, lot #, and % purity: GSK716155, lot # 03A22115 (process 1), 96.8% pure

**Key Study Findings**

- 1/10 LD♀ and 1/10 MD♀ died on Day 2 from unknown causes. Not considered test item related due to lack of dose-response.
- Body weight gain was markedly ↓ in both phases (phase 1 ↓ 144% in HD♂ in wk 2, unremarkable in HD♀; phase 2 ↓ 200% in HD♂ and HD♀ in wk 1) associated with dose-dependent ↓ FC in all dosed ♂ and MD♀ and HD♀ in wk 1 for both phases.
- Eosinophilia at all doses. Neutrophilia in HD♀.
- ss ↓ serum glucose in both phases.
- ss↑ spleen weight was correlated to histopath findings (lymphoid hyperplasia).
- Histopath evaluated in C and HD groups reveled the following target organs of toxicity:
    - injection site (inflammation)

- spleen (lymphoid hyperplasia [sometimes with ↑ germinal centers])
- mesenteric lymph node (lymphangiectasis)
- kidney (an exacerbation of spontaneous chronic progressive nephropathy at terminal sacrifice
- A single HD♀ had multiple findings suggestive of immune-complex-mediated tissue injury: arteritis (minimal to moderate severity) in the lung, skeletal muscle, salivary gland, thymus, thyroid and vagina; glomerulonephropathy/Glomerulosclerosis (moderately severe). These findings are likely to be secondary to the rapid ADA response seen in this study.

- Sponsor considers the NOAEL to be ≤ 5 mg/kg/day on the basis of decreased food consumption and eosinophilia at all tested doses. The immune-related findings of glomerulonephritis and arteritis are also considered adverse, and were not assessed in MD or LD animals. Although adverse to the mice, the clinical relevance of the immune-related findings is uncertain, since GSK716155 is based on human sequence, and less likely to be immunogenic in humans.
- Due to the rapid mounting of a toxicologically significant ADA response that also impairs exposure, the mouse is a poor model for assessing toxicity associated with longer-term GSK716155 treatment.

Methods

| | |
|---|---|
| Doses: | 0, 5, 50, or 150 mg/kg |
| Frequency of dosing: | Daily |
| Route of administration: | Subcutaneous |
| Dose volume: | 5 mL/kg |
| Formulation/Vehicle: | Diluent = 10 mM Sodium Phosphate, 2.8% Mannitol, 4.0% Sucrose, 0.01% Polysorbate 80, pH 7.2 |
| Species/Strain: | Mouse/CD-1 |
| Number/Sex/Group: | *See Sponsor's table below* |
| Age: | 8 weeks |
| Weight: | ♂: 20.1 to 35.8 g ♀: 16.4 to 27.4 g |
| Satellite groups: | TK animals (*see Sponsor's table below*) |
| Unique study design: | (*see Sponsor's table below*) |
| Deviation from study protocol: | None material to the interpretation of the study |



Reference ID: 3347159

Group Designation and Dose Levels

| Group | No. of Animals Male | No. of Animals Female | Targeted Dose Level[a] (mg/kg/day) | Targeted Dose Concentration[a] (mg/mL)[b] |
|---|---|---|---|---|
| | | Study Phase 1 | | |
| Toxicity Animals[d] | | | | |
| 1 (Vehicle Control)[c] | 30 | 30 | 0.0 | 0 |
| 2 (Low) | 30 | 30 | 5 | 1 |
| 3 (Mid) | 30 | 30 | 50 | 10 |
| 4 (High) | 30 | 30 | 150 | 30 |
| Toxicokinetic Animals | | | | |
| 5 (low) | 21 | 21 | 5 | 1 |
| 6 (mid) | 21 | 21 | 50 | 10 |
| 7 (high) | 21 | 21 | 150 | 30 |
| | | Study Phase 2 | | |
| Toxicity Animals[e] | | | | |
| 8 (Vehicle Control)[c] | 10 | 10 | 0.0 | 0 |
| 9 (Low) | 10 | 10 | 5 | 1 |
| 10 (Mid) | 10 | 10 | 50 | 10 |
| 11 (High) | 10 | 10 | 150 | 30 |
| Toxicokinetic Animals | | | | |
| 12 (low) | 15 | 15 | 5 | 1 |
| 13 (mid) | 15 | 15 | 50 | 10 |
| 14 (high) | 15 | 15 | 150 | 30 |

a The calculated dose levels and concentrations were 5, 51.5, and 154.5 mg/kg and 1, 10.3, and 30.9 mg/mL for the low, mid and high dose groups, respectively (see Appendix 1).
b The dose volume was 5 mL/kg.
c Animals received the control/vehicle article only.
d The first 10 mice/sex/group by numerical assignment were sacrificed on Day 8 and the next 10 mice/sex/group by numerical assignment on Day 16. All surviving mice/sex/group underwent 1 week of recovery following at least 2 weeks of dose administration.
e All surviving mice were sacrificed on Day 8.

## Observations and Results

### Mortality

No test item-related deaths. Two ♀s (1 LD & 1 MD) were sacrificed moribund on Day 2 with clinical signs of limited use of hind limbs, cold to touch and sternal recumbency. No cause of moribundity was determined. Deaths were not considered to be test item-related since there was no dose-dependency.

### Clinical Signs

Unremarkable except for the early decedents (described above).

### Body Weights & Feed Consumption

| Daily Dose (mg/kg/day)[1] | | Male 0 | Male 5 | Male 50 | Male 150 | Female 0 | Female 5 | Female 50 | Female 150 |
|---|---|---|---|---|---|---|---|---|---|
| Body Weight (%)[4] | Week 2 | 33.1 g | 0.3% | -1.8% | -6.3% | 24.8 g | 0.4% | -1.6% | -6.5% |
| | Week 4 | 32.9 g | -0.9% | -1.5% | -0.3% | 26.7 g | 2.6% | 3.0% | 0.7% |
| Food Consumption (%[5]) | | | | | | | | | |
| | Week 1 | 42 g | -11.9%* | -28.6%* | -28.6%* | 31 g | -3.2% | -12.9%* | -29.0%* |
| | Week 2 | 41 g | 0% | 0% | -4.9% | 36 g | 5.6% | 8.3% | -2.8% |

### Ophthalmoscopy

Unremarkable.

### Hematology

S.s. ↓ in RBC parameters (RBCs, Hemoglobin, Hematocrit) in ♀s. N.s.s. ↓ in reticulocytes (♂ & ♀). S.s. ↑ in WBC and neutrophils in ♀s (similar n.s.s. trend in ♂s). Eosinophils were n.s.s. ↑ in both sexes.

| Daily Dose (mg/kg/day)[1] | | Male 0 | Male 5 | Male 50 | Male 150 | Female 0 | Female 5 | Female 50 | Female 150 |
|---|---|---|---|---|---|---|---|---|---|
| Haematology[6] | | | | | | | | | |
| Reticulocytes ($10^3/\mu L$) | Day 8 | 290.5 | 10.0% | 2.4% | -35.0% | 356.3 | -5.1% | -17.7% | -33.3% |
| RBC ($10^6/\mu L$) | Day 16 | 9.9 | -2.3% | -4.2% | -2.3% | 10.4 | -10.7%* | -14.8%* | -14.1%* |
| HGB (g/dL) | Day 16 | 15.7 | 1.3% | -3.8% | -1.3% | 16.4 | -6.1% | -12.2%* | -9.8%* |
| HCT (%) | Day 16 | 52.1 | -1.3% | -4.6% | -4.8% | 54.3 | -8.3% | -13.6%* | -12.2%* |
| WBC ($10^3/\mu L$) | Day 16 | 5.7 | 23% | 16% | 30% | 5.5 | 24% | 35% | 60%* |
| Neutrophil ($10^3/\mu L$) | Day 16 | 0.8 | 25% | 38% | 75% | 0.7 | 1.4-fold | 1.7-fold | 3.2-fold* |
| Eosin ($10^3/\mu L$) | Day 16 | 0.1 | 4-fold | 2-fold | 4-fold | 0.1 | 3-fold | 2-fold | 4-fold |

### Clinical Chemistry

≥ 50 mg/kg/day: glucose ↓, total protein and albumin ↓ (♂ only).

During recovery phase ♀s had ↑ globulin & total protein (possibly related to ADA response).

| Daily Dose (mg/kg/day)[1] | | Male 0 | Male 5 | Male 50 | Male 150 | Female 0 | Female 5 | Female 50 | Female 150 |
|---|---|---|---|---|---|---|---|---|---|
| Clinical Chemistry[6] | | | | | | | | | |
| glucose (mg/dL) | Day 8 | 191 | -6.3% | -19.4%* | -16.8%* | 172 | 3.5% | -29.7%* | -19.8% |
| | Day 16 | 205 | 4.9% | -24.4%* | -23.9%* | 187 | 1.1% | -4.3% | -8.0% |
| Total Protein (g/dL) | Day 8 | 6.0 | -3.3% | -11.7%* | -11.7%* | 5.4 | 0% | -1.9% | 3.7% |
| | Day 23 | 5.7 | -3.5% | 0% | 0% | 5.3 | 9.4% | 5.7% | 18.9%* |
| Albumin (g/dL) | Day 8 | 4.0 | -5.0% | -12.5%* | -12.5%* | 3.8 | 0% | 2.6% | 5.3% |
| Globulin (g/dL) | Day 23 | 2.2 | -13.6% | -4.5% | -9.1% | 1.5 | 20.0% | 13.3% | 40.0%* |

### Gross Pathology

Kidney pale in 1 HD♀ on Day 23 (phase 1), correlates with histopath finding of glomerulonephropathy

### Organ Weights

ss ↓ salivary gland wt (abs. and/or rel. to body wt), MD and HD, ♂ and ♀ Day 8; ss ↓ heart and adrenal wt (abs. and/or rel. to body wt), MD and HD ♀, Day 8; No microscopic correlation to these organ wt changes.

ss ↑ spleen wt, MD and HD, ♂ and ♀, phase 1 on Day 16, correlates to histopath finding of lymphoid hyperplasia.

| | | Male | | | | Female | | | |
|---|---|---|---|---|---|---|---|---|---|
| Daily Dose (mg/kg/day)[1] | | 0 | 5 | 50 | 150 | 0 | 5 | 50 | 150 |
| Organ Weights[7] (%) | | | | | | | | | |
| Adrenals (g) | Day 8 | 0.009 | 0% | -11.1% | -11.1% | 0.013 | -7.7% | -23.1%* | -30.8%* |
| Salivary gland (mandibular, g) | Day 8 | 0.20 | -10.0% | -15.0%* | -15.0%* | 0.14 | -7.1% | -28.6%* | -28.6%* |
| Heart (g) | Day 8 | 0.18 | -5.6% | -5.6% | -16.7% | 0.13 | 0% | -15.4%* | -15.4%* |
| Spleen (g) | Day 16 | 0.07 | 28.6% | 42.9%* | 42.9%* | 0.09 | 11.1% | 33.3%* | 22.2% |

**Histopathology**

**Adequate Battery: yes**
**Peer Review: No**

**Histological Findings (only C & HD groups examine)**

Phase 2 kill (Day 8):

| | | M | | F | |
|---|---|---|---|---|---|
| | | C | HD | C | HD |
| Injection site | # Examined | 10 | 10 | 10 | 10 |
| | Subcutis, chronic inflammation | 1 | 10 | 3 | 10 |
| Spleen | # Examined | 10 | 10 | 10 | 10 |
| | Lymphoid hyperplasia | 0 | 0 | 2 | 3 |
| Kidney | # Examined | 10 | 10 | 10 | 10 |
| | Tubule, dilatation | 0 | 0 | 0 | 1 |
| | Cyst | 0 | 0 | 0 | 1 |
| Heart | # Examined | 10 | 10 | 10 | 10 |
| | Epicardium, chronic inflammation | 0 | 0 | 0 | 1 |

Phase 1 terminal kill (Day 16):

| | | M | | F | |
|---|---|---|---|---|---|
| | | C | HD | C | HD |
| Injection site | # Examined | 10 | 10 | 10 | 10 |
| | Subcutis, chronic inflammation | 1 | 9 | 2 | 10 |
| Spleen | # Examined | 10 | 10 | 10 | 10 |
| | Lymphoid hyperplasia | 2 | 10 | 0 | 8 |
| Kidney | # Examined | 10 | 10 | 10 | 10 |
| | Nephropathy, chronic progressive | 3 | 5 | 1 | 4 |
| | Pelvis, dilatation | 0 | 1 | 0 | 0 |
| Heart | # Examined | 10 | 10 | 10 | 10 |
| | Epicardium, chronic inflammation | 0 | 0 | 0 | 1 |

Phase 1 recovery kill (Day 23):

| | | M | | F | |
|---|---|---|---|---|---|
| | | C | HD | C | HD |
| Injection site | # Examined | 10 | 10 | 10 | 10 |
| | Subcutis, chronic inflammation | 0 | 10 | 1 | 7 |
| Spleen | # Examined | 10 | 10 | 10 | 10 |
| | Lymphoid hyperplasia | 4 | 9 | 2 | 6 |
| Thyroid | # Examined | 10 | 9 | 8 | 10 |
| | Arteritis | 0 | 0 | 0 | 1 |
| Lung | # Examined | 10 | 10 | 10 | 10 |
| | Arteritis | 0 | 0 | 0 | 1 |
| Heart | # Examined | 10 | 10 | 10 | 10 |
| | Epicardium, chronic inflammation | 0 | 0 | 0 | 1 |
| Salivary gland | # Examined | 10 | 10 | 10 | 10 |
| | Arteritis, periglandular tissue | 0 | 0 | 0 | 1 |
| Vagina | # Examined | | | 10 | 10 |
| | Arteritis | | | 0 | 1 |

No remarkable findings in the two early decedents.

**Special Evaluation**

Anti-GSK716155 antibodies:

- The repeated SC administration of GSK716155 to CD-1 mice resulted in the formation of anti-GSK716155 antibodies in 89-100% of the mice within each treatment group.
- Antibodies to GSK716155 were observed in the mice as early as Day 8 (first day assessed). The incidence of antibodies remained the same on Days 16 and 23.
- During the treatment period, the median anti-GSK716155 antibody titers were inversely related to the dose of GSK716155 administered. This relationship may be due to interference in the immunogenicity assay with high circulating concentrations of GSK716155 or immunotolerance.
- At the end of the recovery period, the median antibody titer in the low dose group (5 mg/kg) was decreasing, consistent with a diminished antibody response. During this period, the median antibody titers for the mid (50 mg/kg) and high (150 mg/kg) dose groups increased, consistent with drug interference with ADA detection during the dosing phase.

**Toxicokinetics**

In both phases 1 and 2, the mean $C_{max}$ following the 1st dosing between 50 and 150 mg/kg increased in a dose proportional manner. Data on Day 15 were only collected in study phase 1 – mean $C_{max}$ decreased between Days 7 and 15 for all dose groups (scanned Table 2), variability within-group was increased as evidenced by the larger SD

(Table 2). The decrease in systemic exposure coincides with the emergence of anti-GSK antibodies. AUC values were not determined.

**Table 2 Mean (95% CI) plasma concentrations of Albugon in CD-1 mice in Study Phase 1**

| Parameter Mean (95% CI) | | Study Day | 5 mg/kg/day | 50 mg/kg/day | 150 mg/kg/day | [d]P Value |
|---|---|---|---|---|---|---|
| $C_{max,1}$ | µg/mL | 1.25 | [a]9.4 ± 5.2 (3.93-14.92) | 79.8 ± 19.7 (59.18-100.49) | 167.7 ± 61.0 (103.71-231.68) | NA |
| $C_{max,1}$/dose | kg/mL | 1.25 | [a]0.0016 ± 0.0010 | 0.0016 ± 0.0004 | 0.0011 ± 0.0004 | 0.1843 |
| $C_{min,1}$ | µg/mL | 2 | 4.4 ± 0.6 (3.75-4.97) | 52.0 ± 15.1 (36.21-67.83) | 154.0 ± 99.1 (50.00-258.02) | NA |
| $C_{min,1}$/Dose | kg/mL | 2 | 0.0009 ± 0.0001 | 0.0010 ± 0.0003 | 0.0010 ± 0.0007 | 0.7526 |
| $C_{min,6}$ | µg/mL | 7 | 3.8 ± 2.2 (1.52-6.07) | 56.0 ± 39.1 (14.89-97.03) | 231.2 ± 133.6 (91.00-371.45) | NA |
| $C_{min,6}$Dose | kg/mL | 7 | 0.0008 ± 0.0004 | 0.0011 ± 0.0008 | 0.0015 ± 0.0009 | NA |
| $C_{max,7}$ | µg/mL | 7.25 | [b]10.6 ± 2.3 (8.25-12.99) | 163.5 ± 65.4 (94.80-232.13) | [b]420.3 ± 111.7 (303.05-537.47) | NA |
| $C_{max,7}$/Dose | kg/mL | 7.25 | 0.0021 ± 0.0005 | 0.0033 ± 0.0013 | 0.0028 ± 0.0007 | |
| $C_{min,7}$ | µg/mL | 8 | 3.1 ± 1.4 (1.63-4.49) | 38.9 ± 36.9 (0.16-77.68) | 196.4 ± 120.7 (69.74-323.04) | NA |
| $C_{min,7}$/Dose | kg/mL | 8 | 0.0006 ± 0.0008 | 0.0008 ± 0.0007 | 0.0013 ± 0.0008 | NA |
| $C_{max,15}$ | µg/mL | 15.25 | [b,c]0.8 ± 1.3 (-0.53-2.10) | 73.9 ± 80.5 (-10.57-158.32) | [b]141.8 ± 103.2 (33.47-250.05) | NA |
| $C_{max,15}$/Dose | kg/mL | 15.25 | [b]0.0002 ± 0.0003 | 0.0015 ± 0.0016 | 0.0009 ± 0.0007 | NA |

Values shown are mean ± SD; N = 6 per group.
[a]One out of 6 mice did not have detectable Albugon levels; a 0 value for this animal was averaged in for the mean calculation.
[b]Indicates a significantly lower value for $C_{max,15}$ compared to $C_{max,7}$ ($p < 0.05$).
[c]Two out of 6 mice did not have detectable Albugon concentrations; 0 values for these 2 animals were averaged in for the mean calculations.
[d]Results from 1-way ANOVA assessing linearity after the 1st dose of Albugon.
CI = confidence interval.
NA = not applicable.

## Dosing Solution Analysis

Unremarkable.

*The 4-, 5-, and 26-week monkey studies were reviewed previously under IND 65,177. As they do not provide any unique toxicology information compared to the 52-week monkey study (which is fully reviewed below), and there were no substantive differences in opinion between the Agency and the Sponsor regarding the interpretation of these studies, the Sponsor's Tabulated Summaries of these studies are reproduced below for brevity.*

**Table 7.3 One-Month Repeated Dose Subcutaneous Toxicity Study of Albugon in Monkeys with a Two Week Recovery Period**

| | | |
|---|---|---|
| **Study Type:** Repeat Dose Toxicity | **Report Title:** One-Month Repeated Dose Subcutaneous Toxicity Study of Albugon in Monkeys with a Two Week Recovery Period | **Test Article:** GSK716155A<br>**Batch No.:** 03A22115 |
| **Species/Strain:** Monkey/Cynomolgus | **Duration of Dosing:** 4 weeks<br>**Duration of Recovery:** 2 weeks | **GSK Document Number:** RD2005/01133<br>**Study No.:** (b) (4) |
| **Initial Age:** juvenile to sub-adult (exact ages unknown) | **Route/Frequency:** Subcutaneous, once weekly | **Location in CTD:** m4.2.3.2 |
| **Date of First Dose:** 19-Feb-2003 | **Vehicle/Formulation:** 10 mM Sodium phosphate, 2.8% mannitol, 4.0% sucrose and 0.01% polysorbate 80 | **Study in Compliance with GLP:** Yes |

**Data Collected:** Toxicokinetics, clinical observations, body weight, food consumption, ophthalmoscopy, electrocardiography, haematology, coagulation, clinical chemistry, urinalysis, organ weights, macroscopic and microscopic pathology, and immunogenicity

**Conclusion:** GSK716155 (vehicle, 5, 15 and 50 mg/kg/week) was given subcutaneously to male and female monkeys for up to 4 weeks. Clinical observations related to treatment included emesis and reduced food consumption at all doses that led to marked body weight loss in the first week at 150 mg/kg/week. Sera glucose levels were decreased after the initial dose only at all dose levels. In general, incidence and severity of emesis and food consumption effects were attenuated with subsequent dose administrations. Body weights remained relatively unchanged the remainder of the treatment period and were returning to baseline levels during the recovery period. Decreased food consumption, emesis and decreased glucose levels are consistent with the pharmacological activity of GLP-1 analogs. Anti-GSK716155 antibodies were observed at all doses by Day 15 and monkeys with anti-GSK716155 antibodies developed antibodies to the GLP-1 moiety. The antibodies did not appear to affect GSK716155 toxicokinetic parameters. Based on the severity of decreases in body weight and food consumption at 50 mg/kg/week, the no adverse effect level (NOAEL) for GSK716155 given to monkeys once weekly for 4 weeks is 15 mg/kg/week (Day 22 AUC of 7820 μg.h/mL and $C_{max}$ of 105 μg/mL, males and females combined).

**Table 7.3 (Continued) One-Month Repeated Dose Subcutaneous Toxicity Study of Albugon in Monkeys with a Two Week Recovery Period**

| | | Male | | | | Female | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Weekly Dose (mg/kg/week)**[1] | | 0 | 5 | 15 | 50 | 0 | 5 | 15 | 50 |
| **Numbers of Animals:** | **Main** | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 |
| (Recovery) | | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 |
| $AUC_{(0\text{-last})}$ (μg.h/mL) | Day 1 | NQ | 1946 | 8133 | 28913 | NQ | 2642 | 6821 | 31667 |
| | Day 22 | | 3162 | 7425 | 39144 | | 2661 | 8214 | 47528 |
| $C_{max}$ (μg/mL) | Day 1 | NQ | 22.5 | 107 | 396 | NQ | 35.6 | 89 | 363 |
| | Day 22 | | 36.8 | 102 | 489 | | 35.3 | 107 | 520 |
| **Noteworthy Findings** | | | | | | | | | |
| **Clinical Observations** | | Treatment related observations of emesis, lethargy and reduced/absent feces were observed in test article administered animals. After the initial dose administration, emesis was observed in all treated groups for up to 3 days following dosing and occurred with a higher incidence and greater severity in the high dose group. Dose related observations of reduced/absent feces were observed up to six days after the initial dose and was correlated with the lack of food consumption. In general, there was a decrease in incidence and severity of emesis and food consumption effects after the subsequent dose administrations. | | | | | | | |
| **Body Weight** (%[2]) | | -2.4% | -6.4% | -8.9%* | -12.9%* | -4.5% | -7.3% | -9.1%* | -12.4%* |
| **Daily Food Consumption** (%[2]) | | -38% | -82% | -100% | -100% | -18% | -98% | -98% | -93% |
| **Clinical Chemistry** | | | | | | | | | |
| glucose (mg/dL) Day 2 | | 80 | -34%* | -38%* | -29%* | 76 | -32%* | -39%* | -39%* |
| **Anti-GSK716155 Antibodies (incidence)** | Day 29 | 0/5 | 4/5 | 4/5 | 5/5 | 1/5 | 3/5 | 4/5 | 5/5 |

Key:
* $p<0.05$
1 = Doses are expressed in terms of pure active moiety
2 = Maximal percent change (Day 6) from Day -2 predose values are shown (Day 6 for body weight and Day 2 for food consumption). Statistical significance is based on actual data not percentage differences.

**Table 7.4 GSK716155: 5-Week Subcutaneous Toxicity Study in Cynomolgus Monkeys with a 2-Week Reversibility Period**

| | | |
|---|---|---|
| **Study Type:** Repeat Dose Toxicity | **Report Title:** GSK716155: 5-Week Subcutaneous Toxicity Study in Cynomolgus Monkeys with a 2-Week Reversibility Period | **Test Article:** GSK716155<br>**Batch No.:** JCM-203448-081 |
| **Species/Strain:** Cynomolgus/ Monkeys | **Duration of Dosing:** 5 weeks<br>**Duration of Recovery:** 2 weeks | **GSK Document Number:** CD2005/00839<br>**Study No.:** G05072 |
| **Initial Age:** 2-4 years | **Route/Frequency:** Subcutaneous, twice weekly | **Location in CTD:** m4.2.3.2 |
| **Date of First Dose:** 20 June 2005 | **Vehicle/Formulation:** 10 mM sodium phosphate, 2.8% mannitol, 4.2% trehalose, 0.01% polysorbate 80, pH 7.2, | **Study in Compliance with GLP:** Yes |

**Data Collected:** Toxicokinetics, clinical observations, neurobehavioral assessment, body weight, food consumption, ophthalmoscopy, electrocardiography, hematology, coagulation, clinical chemistry, immunogenicity, urinalysis, organ weights, macroscopic and microscopic pathology,

**Conclusion:** GSK716155 (vehicle, 5, 15 and 50 mg/kg/week) was given subcutaneously to male and female monkeys for up to 5 weeks. Clinically, dose-dependent decreases in food consumption were observed in all animals given GSK716155 throughout the treatment phase. In addition, animals given 50 mg/kg/week had significant decreases in body weight, which were not present on Day 36 in males and Day 43 in females (4 and 11-days off drug, respectively). Anti-GSK716155 antibodies were observed in all treated monkeys, but antibodies did not appear to affect toxicokinetic parameters. Based on the severity of decreases in body weight and food consumption, the no observable adverse effect level (NOAEL) for monkeys given GSK716155 twice weekly for 5 weeks was 15 mg/kg/week (Day 32 AUC of 4350 µg.h/mL and Cmax of 71.2 µg/mL).

**Table 7.4 (Continued) GSK716155: 5-Week Subcutaneous Toxicity Study in Cynomolgus Monkeys with a 2-Week Reversibility Period**

| | | Male | | | | Female | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Weekly Dose (mg/kg/week)[1]** | | 0 | 5 | 15 | 50 | 0 | 5 | 15 | 50 |
| **Numbers of Animals:** | **Main** | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 |
| (Recovery) | | 2 | | | 2 | 2 | | | 2 |
| **$AUC_{(0-72)}$ (µg.h/mL)** | **Day 1** | NQ | 716 | 1901 | 8135 | NQ | 596 | 2181 | 7321 |
| | **Day 32** | | 1304 | 4581 | 16630 | | 809 | 4119 | 13994 |
| **$C_{max}$ (µg/mL)** | **Day 1** | NQ | 13.3 | 37.2 | 150 | NQ | 10.0 | 37.7 | 140 |
| | **Day 32** | | 21.2 | 75.4 | 286 | | 13.8 | 67.0 | 233 |
| **Noteworthy Findings** | | | | | | | | | |
| **Clinical Observations** | | Dose dependent decreases in food consumption were observed in all animals given GSK716155. Animals given 50 mg/kg/week had marked decreases in food consumption throughout the treatment phase, whereas animals given 15 or 5 mg/kg/week had lower incidence and severity during the treatment period (incidence was reduced with repeat dosing). Due to the severity of these observations, veterinary care was necessary for 3 animals given 50 mg/kg/week, for treatment of dehydration (P05M-1194 and P05M-1198 Day 12 (subcutaneous fluids) P05F-1199- Days 12-19 (subcutaneous fluids and Nutrical™)). | | | | | | | |
| **Body Weight (%)[2]** | | >1% | -1% | -5% | -7% | 2% | -1% | -3% | -10% |
| **Immunogenicity Incidence (Day 39)** | | 1/5 | 3/3 | 3/3 | 5/5 | 0/5 | 3/3 | 3/3 | 5/5 |
| **Confirmed Positive[3]** | | 0/1 | 1/3 | 3/3 | 1/5 | -- | 0/3 | 2/3 | 0/5 |
| **Microscopic Examination (number of animals)** | | 0 | 5 | 15 | 50 | 0 | 5 | 15 | 50 |
| Skin, Injection Site | | | | | | | | | |
| Mononuclear Infiltrate minimal-mild | | 0 | 0 | 0 | 2 | 0 | 0 | 2 | 1 |

Key:
1 = Dose was administered twice weekly, approximately 72 hours apart. Day 32 was the day the last dose of the study was administered.
2 = Maximal decrease at Day 18. Group means are shown for treated groups percentage change from Day 1.
3 = Positive incidence of antibodies was verified with a second analysis and confirmed positive numbers are listed from Day 39 results.

**Table 7.5 GSK716155: 26-Week Subcutaneous Injection Toxicity Study in Cynomolgus Monkeys with a 4-Week Recovery Period**

| | | |
|---|---|---|
| **Study Type**: Repeat-Dose Toxicity | **Report Title**: GSK716155: 26-Week Subcutaneous Injection Toxicity Study in Cynomolgus Monkeys with a 4-Week Recovery Period | **Test Article**: GSK716155<br>**Batch Nos.**: MJG-205952-034 |
| **Species/Strain**: Cynomolgus monkey (*Macaca fascicularis*) | **Duration of Dosing**: 26 week<br>**Duration of Recovery**: 4 weeks | **GSK Document Number**: RD2005/01281<br>**Study No.**: P41726 |
| **Initial Age**: 2 years and 10 months – 4 years and 4 months | **Route/Frequency**: Subcutaneous injection/once weekly | **Location in CTD**: m4.2.3.2 |
| **Date of First Dose**: 17 November 2005 | **Vehicle/Formulation**: 10 mM sodium phosphate, 4.2% (w/v) trehalose, 2.8% (w/v) mannitol and 0.01% (w/v) polysorbate-80 at pH 7.2 | **Study in Compliance with GLP**: Yes |

**Data Collected**: Toxicokinetics, clinical observations, body weight, food consumption (qualitative), ophthalmoscopy, electrocardiography, hematology, coagulation, clinical chemistry (including serum insulin), urinalysis, immunogenicity, organ weights, macroscopic and microscopic pathology

**Conclusion**: GSK716155 (1, 5, or 15 mg/kg/week) was given by subcutaneous injection to male and female monkeys once a week for 26 weeks and a subset of monkeys kept for a 4 week recovery period. Occasional vomiting, decreased food consumption, and decreased mean body weight gain observed in monkeys given ≥5 mg/kg/week were consistent with the known effects of GLP-1 agonists, and these effects all showed evidence of reversibility during the recovery phase. Anti-GSK716155 antibodies were confirmed in monkeys at all doses, and high titers were associated with decreased systemic exposure values of GSK716155. The no observed adverse effect level (NOAEL) is 15 mg/kg/week (Week 26 mean $AUC_{(0-168)}$ of 13811 µg.h/mL [range 10144 to 24823 µg.h/mL] for males and 12745 µg.h/mL [range 8800 to 18097 µg.h/mL] for females).

**Table 7.5 (Continued) GSK716155: 26-Week Subcutaneous Injection Toxicity Study in Cynomolgus Monkeys with a 4-Week Recovery Period**

| | | Male | | | | Female | | | |
|---|---|---|---|---|---|---|---|---|---|
| Daily Dose (mg/kg/week)[1] | | 0 | 1 | 5 | 15 | 0 | 1 | 5 | 15 |
| Numbers of Animals: | Main | 4 | 4 | 4 | 4 | 4 | 4 | 4 | 4 |
| | Recovery | 2 | 0 | 0 | 2 | 2 | 0 | 0 | 2 |
| $AUC_{(0-168)}$ (µg.h/mL) | Day 1 | | 178 | 2245 | 6999 | | 418 | 2238 | 7065 |
| | Week 26 | | 528 | 3517 | 13811 | | 653 | 3115 | 12745 |
| $C_{max}$ (µg/mL) | Day 1 | | 1.67 | 20.7 | 68.2 | | 4.20 | 20.3 | 71.1 |
| | Week 26 | | 5.42 | 34.9 | 121 | | 6.64 | 29.1 | 120 |
| Noteworthy Findings | | | | | | | | | |
| Clinical Observations | | Vomiting was occasionally observed in ≥1 mg/kg/week males and 15 mg/kg/week females. Decreased food consumption was observed in monkeys given ≥5 mg/kg/week that correlated with decreased body weight gain during the dosing phase of the study. All these effects were reversible during the recovery phase. | | | | | | | |
| Vomiting | | 0 | 1 | 3 | 5 | 0 | 0 | 0 | 2 |
| Body Weight Change (%)[2]-Dosing Phase | | 1 kg | 0 | -50 | -50 | 0.5 kg | 20 | -40 | -60 |
| Body Weight Change (%)[2]-Recovery Phase | | 0.1 kg | - | - | 300 | 0.1 kg | - | - | 300 |
| Food Consumption | | | | | | | | | |
| No food consumption | | 0 | 0 | 4 | 6 | 1 | 1 | 4 | 6 |
| Incidence of confirmed anti-GSK716155 antibodies | | 0 | 4 | 3 | 5 | 0 | 1 | 3 | 4 |

Key:
- Not examined or not applicable
1. Doses are expressed in terms of parent compound.
2. Over the entire dosing or recovery period. For controls, group means are shown; for treated groups, percentage change from controls is shown.

| GSK716155: 52-Week (Once Weekly) Subcutaneous Injection Toxicity Study in Cynomolgus Monkeys | | | |
|---|---|---|---|
| **Study no:** 7717-809 (b) (4) | | **Dose:** 0, 15, 50 mg/kg/week<br>**HED:** 0, 5, 16 mg/kg/week*<br>*mg/kg scaling, adjusted for 3-fold $EC_{50}$ difference | **Species:** Cynomolgus monkey |
| **#/s/g:** | **Main:** 5 | **Vehicle:** 10 mM sodium phosphate, 4.2% (w/v) trehalose, 2.8% (w/v) mannitol and 0.01% (w/v) Polysorbate-80 at pH 7.2<br>**Volume:** 2 mL/kg | **Route:** SC |
| **Key findings** | • **No dose-limiting toxicity was observed at any administered dose; therefore the NOAEL is considered to be 50 mg/kg/week (the highest dose tested) in this study.**<br>• **GSK716155 (and exenatide comparator) were associated with decreased food consumption and decreased body weight gain, especially early in the study. GSK716155 had no effect on terminal body weight in male monkeys; however LD♀s gained only 50%, and HD♀s gained only 25% of the weight gained by controls over the course of the study. As a group, females treated with exenatide gained no weight over the 52-week study. These effects on weight gain were considered to reflect the intended pharmacological action of GSK716155 (and exenatide) and were not considered adverse.**<br>• **Mean pancreas weight, whether represented as absolute weight, % of bodyweight or % of brain weight, was dose-dependently increased in both sexes. However, there were no histopathology findings that were correlated with the increase in pancreas weight, and this finding is not considered to be adverse. Mean pancreas weight was also increased in the exenatide treatment group in both sexes.**<br>• **In males, thymus weight was dose-dependently ↓ by GSK716155 treatment and exenatide treatment. Sponsor attributes this to normal involution; however the incidence of thymic involution (by histology) was comparable in C and HD animals. In the absence of other indications of immunosuppression in HD♂s these changes in thymic weight are not considered adverse.**<br>• **Several GSK716155-treated animals developed anti-drug antibodies; however, these were generally low titer and/or failed to persist. There is no evidence that the ADA interfered with the interpretability of the study or caused toxicity.** | | |

**Volume #, and page #**: C325.1-C325.2, all
**Conducting laboratory and location**: (b) (4)
**Date of study initiation**: 22 May 2008
**GLP compliance**: Yes *(but with deviations that did not affect the interpretability of the data)*
**QA report**: yes ( x ) no ( )
**Drug, lot #, and % purity**: GSK716155, batch # MJG 205952-124A, 97.7% pure
GSK716155, batch # MJG 205952-149, 99.3% pure
*-- both lots are process 2 material*

**Methods** (not otherwise captured in table above):

Age: 2.7 to 5.1 years old at start of treatment
Weight: ♂: 2.3 to 3.8 kg ♀: 2.5 to 4.2 kg
Source: (b) (4)
Unique study design or methodology:

An additional comparator arm was included, where 5 animals/sex were given BID doses of Exenatide. This group received 25 μg/kg/dose for the first 14 days, after which the dose was increased to 75 μg/kg/dose for the remainder of the study.

Serum samples were assessed for the presence of anti-GSK716155 or anti-Exenatide antibodies as described below.

**Results:**

Mortality: *Assessed twice daily.*

No early deaths.

Clinical signs: *Twice daily during dosing period (predose and ~5 to 6 hr postdose). Detailed examinations performed weekly.*

Low or no food consumption occurred with higher frequency in M & F treated with GSK716155 or exenatide, compared to control.

Text Table 1
Incidence and Frequency (range of occurrences) of Poor Food Consumption

| | Food Consumption Low | | No Food Consumption | |
|---|---|---|---|---|
| | Male | Female | Male | Female |
| Vehicle Control | 3 (3-11) | 5 (10-59) | 0 | 5 (1-7) |
| GSK716155 15 mg/kg/wk | 5 (7-59) | 5 (32-104) | 5 (1-3) | 4 (1-55) |
| GSK716155 50 mg/kg/wk | 5 (6-45) | 5 (45-127) | 4 (2-3) | 5 (4-23) |
| Exenatide 25/75 μg/kg/day | 5 (2-70) | 5 (9-126) | 2 (1-3) | 4 (4-64) |

Body weights: *Twice weekly up to dosing phase week 5 and once weekly thereafter during the dosing phase.*

Monkeys, primarily females, given GSK716155 or exenatide lost weight during the first two weeks of the study (0.3 to 0.6 kg), while control monkeys generally gained weight. Among the males, 2 of 5 given 50 mg/kg/week GSK716155 or exenatide lost weight and among the females, 3 of 5 and 5 of 5 given 15 or 50 mg/kg/week GSK716155, respectively, and 2 of 4 given exenatide lost weight. Weight gain improved after the first month of the study, but at the completion of the study, females given 15 or 50 mg/kg/week GSK716155 gained less weight (0.50X and 0.25X, respectively) than control. As a group, females given exenatide did not gain weight during the study. These clinical signs and changes in body weight were expected effects of compounds in this pharmacological class and were not adverse in the absence of other data indicating poor health.

Food consumption: *Qualitatively assessed daily.*

See “Clinical signs,” above.

Ophthalmoscopy: *All animals prior to initiation of dosing and during dosing week 52.*

Unremarkable.

EKG: *All animals prior to initiation of dosing and during dosing week 52 (at ~$T_{max}$). Animals were unanesthetized.*

No test item-related effect.

Hematology: *All animals (fasted) twice prior to initiation of dosing, and then once during dosing weeks 4, 13, 26 and 52. A standard panel of parameters was measured.*

No test item-related effect.



Reference ID: 3347159

Clinical chemistry: *All animals (fasted) twice prior to initiation of dosing, and then once during dosing weeks 4, 13, 26 and 52. A standard panel of parameters was measured.*

No test item-related effect.

Urinalysis: *All animals (fasted) twice prior to initiation of dosing, and then once during dosing weeks 4, 13, 26 and 52. Urine samples were collected overnight chilled. A standard panel of parameters was measured.*

No test item-related effect.

Gross pathology:

No test item-related effect.

Organ weights: *Adrenals (paired), Brain, Heart, Kidneys (paired), Liver w/gallbladder (drained), Ovaries (paired), Pancreas, Prostate, Testes (paired), Thymus, Thyroid (paired, w/parathyroid)**

**Thyroid weights were taken postfixation by the Sponsor.*

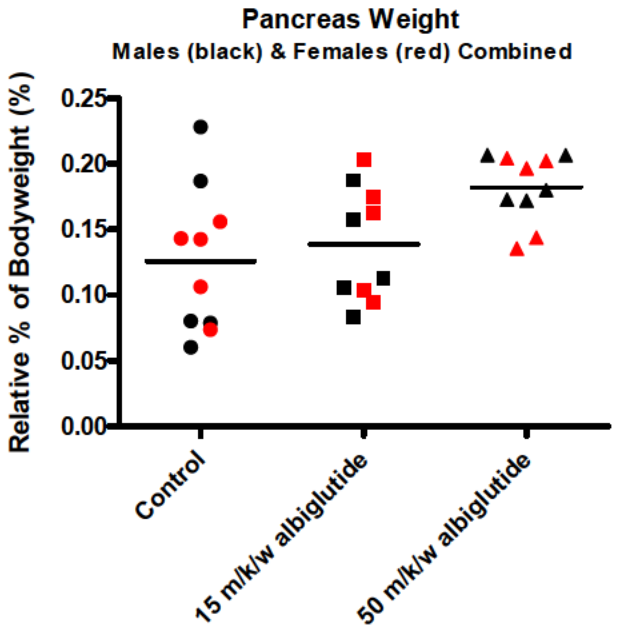
Pancreas weight was increased in both sexes in a dose-related manner. In the absence of histopathology findings in the pancreas, and given the large variability in pancreas weight in the controls (especially ♂s), Sponsor considers this effect to be of no toxicological significance. Exenatide also increased pancreas weight in this study with a magnitude that was intermediate between LD and HD GSK716155. Reviewer's analysis (see below) combines male and female bodyweight-referent pancreas weights on the basis that the mean of this value in the control groups was essentially identical in both sexes and that pancreas weight is reportedly well correlated with body weight (as a function of food consumption). This analysis reveals that GSK716155 had a s.s. effect to ↑ pancreas weight. There were no histopathological correlates with the increase in pancreas weight. Given post-marketing signals suggesting a possible link between incretin-based therapies and pancreatitis in the clinic, Sponsor was asked to determine by stereology which cell types are responsible for the increase in pancreas mass, and to evaluate the pancreases for increased proliferation (review of this analysis appears on page 49).

Thymus weight was decreased in treated males, but not females. Sponsor attributes this to normal involution. Notably thymus weight was likewise decreased in male monkeys in a 52-week study with liraglutide, suggesting that GLP-1 R agonists may affect the thymus in male monkeys.

Recovery was not assessed in this study.

| Effect of GSK716155 on Organ Weights: group mean (% change from control) | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| **Sex** | **Males** | | | | **Females** | | | |
| **Treatment** | **GSK716155 (mg/kg/week)** | | | **Exenatide (µg/kg/BID)** | **GSK716155 (mg/kg/week)** | | | **Exenatide (µg/kg/BID)** |
| **Dose** | **0** | **15** | **50** | **25/75** | **0** | **15** | **50** | **25/75** |
| **Pancreas** | | | | | | | | |
| Absolute (g) | 5.02<br>- | 5.18<br>↑3% | 7.84<br>↑56% | 6.39<br>↑27% | 4.29<br>- | 4.96<br>↑16% | 5.43<br>↑27% | 4.78<br>↑11% |
| BW ratio (%) | 0.127<br>- | 0.129<br>2% | 0.188<br>↑48% | 0.161<br>↑27% | 0.124<br>- | 0.148<br>↑19% | 0.176<br>↑42% | 0.159<br>↑28% |
| BrW ratio (%) | 7.36<br>- | 7.63<br>↑4% | 10.99<br>↑49% | 9.37<br>↑27% | 6.87<br>- | 7.76<br>↑13% | 8.02<br>↑17% | 7.42<br>↑8% |
| **Thymus** | | | | | | | | |
| Absolute (g) | 2.60<br>- | 2.37<br>↓9% | 1.79<br>↓32% | 1.80<br>↓31% | n.e. | | | |

| | | | | | |
|---|---|---|---|---|---|
| BW ratio (%) | 0.068<br>- | 0.060<br>↓12% | 0.045<br>↓34% | 0.045<br>↓34% | n.e. |
| BrW ratio (%) | 3.83<br>- | 3.45<br>↓10% | 2.52<br>↓34% | 2.54<br>↓34% | n.e. |




Difference between Control and 50 m/k/w group is s.s. with a p-value = 0.0091 (two-tailed student's t-test).

1-Way ANOVA indicates that both treated groups are s.s. higher than Control (p-value = 0.0181).

Histopathology: *Protocol tissues (see scanned Sponsor's table below) were fixed in 10% neutral-buffered formalin with the following exceptions: 1) testes, epididymides, eyes and optic nerves were preserved in modified Davidson's fixative; 2) thyroid and parathyroids (with trachea attached) were fixed in Bouin's fixative and preserved in 70% ethanol following a special collection procedure prior to being shipped to the Sponsor for processing and microscopic evaluation; 3) pancreases were preserved in 10% neutral-buffered formalin for 19-24 hrs and transferred to 70% ethanol. H&E-stained sections from protocol tissues were analyzed for C and HD animals only.*

*Sponsor conducted microscopic examination of the thyroid/parathyroid using H&E stained sections and sections stained for calcitonin (to identify C cells) by immunohistochemistry. C cells were quantitated by stereology.*

*Adequate Battery: yes ( x ), no ( )—explain*
*Peer review: yes ( x ), no ( )*

With the exception of an increased incidence of minimal perivascular infiltration of mononuclear cells at the site of subcutaneous injection, there were no clear effects of GSK716155 on histology. There was an increase in the incidence of minimal-slight mineralization of the adrenal medulla (1/10 C vs. 3/10 HD, sexes combined); however Sponsor reports that this incidence falls within the historical background range for this finding in this species.

Sponsor reports that there were no test item-related histopathology findings in the thyroid/parathyroid. C-cell (calcitonin positive cells) quantitation by stereology found no test item-related increase in C-cell number (Sponsor's Tables 1 and 2, below).

| Histopathology Findings: incidence (mean severity score) | | | | |
|---|---|---|---|---|
| Sex | Male | | Female | |
| Dose (mg/kg/week) | 0 | 50 | 0 | 50 |
| **Adrenal, Medulla** | | | | |
| Mineralization | 0/5 - | 1/5 (2.0) | 0/5 - | 2/5 (1.0) |
| **Kidney** | | | | |
| Vacuolation Transitional cell epithelium | 0/5 - | 0/5 - | 0/5 - | 1/5 (1.0) |
| Mineralization, increased | 0/5 - | 0/5 - | 0/5 - | 1/5 (1.0) |
| **Thymus** | | | | |
| Cyst | 0/5 - | 2/5 (P) | 0/5 - | 0/5 - |
| Involution | 2/5 (2.0) | 2/5 (2.5) | 0/5 - | 0/5 - |
| **Muscle, Bi Fem** | | | | |
| Necrosis | 1/5 (1.0) | 1/5 (3.0) | 0/5 - | 0/5 - |
| Infiltrate Lymphocytes/Macrophages | 1/5 (1.0) | 2/5 (1.5) | 0/5 - | 0/5 - |
| **Subcutaneous site** | | | | |
| Infiltrate, mononuclear cell Perivascular | 0/5 - | 1/5 (1.0) | 0/5 - | 4/5 (1.0) |

| Tissues Fixed | Tissues Examined | Tissues Fixed | Tissues Examined |
|---|---|---|---|
| Abnormalities (lesions) | X | Ovaries | X |
| Adrenals | X | Pancreas[2,3] | X |
| Animal identification device | | Parathyroids | X |
| Aorta (thoracic) | X | *Pituitary gland* | X |
| Brain | X | Prostate | X |
| Cecum | X | Rectum[1] | |
| Cervix | X | Rib, including costalcondral junction | X |
| Colon | X | Salivary gland | |
| Duodenum | X | Mandibular | X |
| Epididymides | X | Parotid | X |
| Esophagus | X | Sciatic nerve | X |
| Eyes | X | Seminal vesicles | X |
| Femur (Femoral head) with bone marrow | X | Skeletal muscle (thigh) | X |
| Gallbladder | X | Skin/subcutis | X |
| Heart | X | Spinal cord | X |
| Ileum | X | Cervical[1] | |
| *Injection sites (Subcutaneous sites)* | X | Lumbar | X |
| Jejunum | X | Spleen | X |
| Kidneys | X | Sternum with bone marrow | X |
| Larynx [1] | | Stomach | X |
| Liver (2 lobes) | X | Testes | X |
| Lung with large bronchi | X | Thymus | X |
| Lymph node - | | Thyroid | X |
| Mandibular | X | Tongue | X |
| Mesenteric | X | Tonsils[1] | |
| Inguinal[1] | | *Trachea*[3] | X |
| Mammary gland (females) | X | Urinary bladder | X |
| Nasal turbinates[1] | | Uterus | X |
| Nasopharynx[1] | | Vagina | X |
| Optic Nerves | X | | |

1. Collected and held for possible microscopic examination.

2. *Entire pancreas was collected and preserved.*

3. An additional section of trachea and pancreas was required for preparation and microscopic examination by (b) (4)

Control
Albiglutide (50 mg/kg/wk)
Exenatide (75 µg/kg BID)

**Table 1 Estimates of Total C Cells in Male Thyroids**

| Animal # | Code # | Group # | Estimate Total C Cell left | Estimate Total C Cell Right | Estimate Total C Cell | Mean |
|---|---|---|---|---|---|---|
| I64208 | 4 | 1 | 147450 | 101897 | 249347 | 118986 |
| I64209 | 26 | 1 | 75825 | 89434 | 165259 | |
| I64210 | 25 | 1 | 20681 | 40929 | 61610 | |
| I64211 | 11 | 1 | 32414 | 42914 | 75328 | |
| I64212 | 12 | 1 | 20332 | 23054 | 43386 | |
| | | | | | | |
| I64218 | 18 | 3 | 46320 | 41253 | 87572 | 119771 |
| I64219 | 28 | 3 | 50974 | 69890 | 120864 | |
| I64220 | 14 | 3 | 50765 | 62670 | 113435 | |
| I64221 | 21 | 3 | 116751 | 116593 | 233344 | |
| I64222 | 27 | 3 | 24129 | 19510 | 43639 | |
| | | | | | | |
| I64223 | 5 | 4 | 62091 | 48218 | 110310 | 122971 |
| I64224 | 1 | 4 | 56447 | 68937 | 125384 | |
| I64225 | 2 | 4 | 50435 | 42870 | 93305 | |
| I64226 | 3 | 4 | 70237 | 59105 | 129342 | |
| I64227 | 22 | 4 | 94235 | 62281 | 156516 | |

Control
Albiglutide (50 mg/kg/wk)
Exenatide (75 µg/kg BID)

**Table 2 Estimates of Total C Cells in Female Thyroids**

| Animal # | Code # | Group # | Estimate Total C Cell left | Estimate Total C Cell Right | Estimate Total C Cell | Mean |
|---|---|---|---|---|---|---|
| I64228 | 39 | 1 | 194172 | 177476 | 371648 | 184752 |
| I64229 | 9 | 1 | 43078 | 51671 | 94749 | |
| I64230 | 15 | 1 | 120886 | 84163 | 205049 | |
| I64231 | 16 | 1 | 10964 | 18081 | 29045 | |
| I64232 | 13 | 1 | 141615 | 81655 | 223270 | |
| | | | | | | |
| I64238 | 6 | 3 | 47828 | 72141 | 119969 | 158448 |
| I64239 | 17 | 3 | 100565 | 91536 | 192102 | |
| I64240 | 8 | 3 | 113351 | 100920 | 214271 | |
| I64241 | 32 | 3 | 74828 | 102549 | 177377 | |
| I64242 | 19 | 3 | 37693 | 50831 | 88524 | |
| | | | | | | |
| I64243 | 7 | 4 | 63048 | 139955 | 203003 | 140315 |
| I64244 | 40 | 4 | 86178 | 60711 | 146889 | |
| I64245 | 20 | 4 | 48012 | 48128 | 96140 | |
| I64246 | 33 | 4 | 46020 | 77883 | 123903 | |
| I64247 | 23 | 4 | 64143 | 67495 | 131638 | |

Toxicokinetics:

*Groups 1-3: Serial blood draws from all animals were performed on Dosing Phase Day 15, and Dosing Phase Weeks 26 and 52 at Predose (0 hour) and approximately 12, 24,48,72,96, 120 and 168 hours after dosing. Additionally, a single blood sample was collected prior to dosing during Dosing Phase Weeks 5, 9, 13, 17, 22, 32, 36, 40, 44 and 48.*

*Group 4 (exenatide): Serial blood draws from all animals were performed on Dosing Phase Day 15, and Dosing Phase Weeks 26 and 52 at Predose (0 hour) and approximately 0.5, 1, 2, 4, 6, 9 and 12 hours following first dose of the day (12-hour draw was performed prior to the 2nd dose of the day).*

| Parameter [a] | Period | Sex | Dose of GSK716155 (mg/kg/week) | |
|---|---|---|---|---|
| | | | 15 | 50 |
| $AUC_{0-t}$ (mg.h/mL) | Day 15 | Male<br>Female | 14.8 [10.4 - 17.2]<br>18.8 [14.4 – 25.0] | 64.4 [56.1 – 73.2]<br>55.7 [38.7 – 71.9] |
| | Week 26 | Male<br>Female | 13.8 [8.59 - 17.0]<br>16.9 [11.1 – 23.2] | 61.3 [48.7 – 78.7]<br>57.8 [50.1 – 67.4] |
| | Week 52 | Male<br>Female | 14.5 [11.2 - 16.3]<br>17.6 [11.8 – 23.2] | 53.9 [40.9 – 68.3]<br>58.3 [47.3 – 68.2] |
| $C_{max}$ (mg/mL) | Day 15 | Male<br>Female | 0.139 [0.0920 - 0.165]<br>0.166 [0.129 – 0.208] | 0.581 [0.525 – 0.648]<br>0.520 [0.388 – 0.683] |
| | Week 26 | Male<br>Female | 0.128 [0.0846 – 0.157]<br>0.157 [0.120 – 0.205] | 0.556 [0.490 – 0.681]<br>0.646 [0.505 – 0.730] |
| | Week 52 | Male<br>Female | 0.144 [0.105 – 0.166]<br>0.166 [0.111 – 0.232] | 0.513[0.425 – 0.620]<br>0.563 [0.454 – 0.648] |

a. Results are reported as mean unless stated otherwise and [range].

| Parameter [a] | Period | Dose of Exenatide (µg/kg/dose) | |
|---|---|---|---|
| | | 25/75 [b] | |
| | | Male, n=5 | Female, n=5 |
| $AUC_{0-12}$ (µg.h/mL) | Day 15[d] | 0.314 [0.160 – 0.408] | 0.356 [c] [0.312 – 0.414] |
| | Week 26 | 1.86 [0.296 – 2.79] | 0.991 [0.372 – 1.97] |
| | Week 52 | 1.84 [0.304 – 3.28] | 0.953 [0.282 – 1.97] |
| $C_{max}$ (µg/mL) | Day 15[d] | 0.127 [0.105 – 0.153] | 0.158 [c][0.126 – 0.174] |
| | Week 26 | 0.300 [0.107 – 0.399] | 0.199[0.125 – 0.276] |
| | Week 52 | 0.282 [0.123 – 0.448] | 0.204 [0.102 – 0.289] |

a. Results are reported as mean unless stated otherwise and [range].
b. Exenatide was administered at 25 µg/kg/dose for the first 14 days of BID dosing, after which the dose level was increased to 75 µg/kg/dose for the remainder of the study.
c. n=4. One out of 5 female animals was unable to be sampled on Day 15.
d. Due to ongoing stability analysis, these values will be confirmed by amendment to the report at a later time.



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Immunogenicity: *Blood (approximately 1 mL) was collected via a femoral vein into tubes without anticoagulant for Groups 1, 2, and 3: once on Predose Phase Day 11 and within 4 hours prior to dosing in Dosing Phase Weeks 3, 5, 9, 13, 17, 22, 26, 32, 36, 40, 44, 48, and 53 (Days 15, 29, 57, 85, 113, 148, 176, 218, 246, 274, 302, 330, 365, respectively) and for Group 4 samples were collected: once on Predose Phase Day 11 and prior to the first dose in Dosing Phase Weeks 3 (Day 15) and 26 (Day 176), and on the day prior to the first necropsy. Serum samples were prepared and analyzed for anti-GSK716155 or anti-exenatide antibodies by the Sponsor using validated assays. If serum samples contained potential anti-GSK716155 or anti-exenatide antibodies, they were further analyzed for the antibody specificity and titer.*

Anti-GSK716155 antibodies:
Anti-GSK716155 antibodies were detected sporadically in several monkeys from all groups, including controls. However, in 2/10 and 6/10 monkeys given 15 or 50 mg/kg/week GSK716155, respectively, anti-GSK716155 antibodies were detected more consistently (≥ 3 consecutive timepoints) and with higher titers (peak titers ranging from 1:137 to 1:6197) with seroconversion by Weeks 3 to 9. Antibodies in these monkeys were demonstrated to be specific for GSK716155 in a competition binding assay.

**Table 3 Calculated Anti-GSK716155 Antibody Titer[a] in Cynomolgus Monkey Serum**

| Group No. Dose (once weekly) | AIN | Pre-Dose | Week 3 | Week 5 | Week 9 | Week 13 | Week 17 | Week 22 | Week 26 | Week 32 | Week 36 | Week 40 | Week 44 | Week 48 | Week 52 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Group 1 | I64210 | | | | | | | 48 | 48 | | | | | | |
| 0 mg/kg | I64211 | | | 30 | | | | | | | | | | | |
| | I64230 | | | | 60 | | | | 61 | | | | 57 | | |
| | I64231 | | | 77 | | | | | | | | | | | |
| Group 2 | I64213 | | | 75 | 41 | | | | | | | | | | |
| 15 mg/kg | I64214 | | | | | | | 54 | | | | | | | |
| | I64215 | | 58 | | | | | | | | | | | | |
| | I64216 | | | 58 | 135 | 137 | 137 | | | | | | | | |
| | I64233 | | | 2379 | 646 | 612 | 534 | 375 | 648 | 611 | 497 | 521 | 473 | 404 | 333 |
| Group 3 | I64218 | | | 6197 | 638 | 724 | 632 | 506 | 381 | 612 | 516 | 540 | 460 | 359 | 449 |
| 50 mg/kg | I64219 | | | 30 | | | | | | | | | | | |
| | I64220 | | | 985 | 575 | 501 | 609 | 416 | 379 | 121 | | | | | |
| | I64221 | | | 523 | 140 | 136 | 123 | 123 | 98 | 88 | | | | | |
| | I64222 | | | 489 | 656 | 609 | 587 | 606 | 640 | 132 | 136 | | | | |
| | I64238 | | | 30 | 72 | | | | | | | | | | |
| | I64240 | | | | | | | | 64 | 62 | | | | | |
| | I64241 | | | 143 | 262 | 119 | 63 | | | | | | | | |
| | I64242 | | | 458 | 458 | 424 | 501 | 397 | 307 | 206 | 75 | | | | |

[a] Titration at which the sample crosses the cut point (Relative O.D. = 2.095)

Anti-exenatide antibodies: 9/10 monkeys developed antibodies specific for exenatide (peak titers ranging from 1:85 to 1:7967) with onset in most monkeys by Week 26.

**Table 7 Calculated Anti-Exenatide Antibody Titer a in Cynomolgus Monkey Serum**

| Group No. | AIN | Week 26 | Week 52 |
|---|---|---|---|
| Group 4 | I64223 | 353 | 1771 |
| 25/75 | I64225 | 2491 | 2736 |
| ug/kg BID | I64226 | 2538 | 2644 |
| | I64227 | 7967 | 7797 |
| | I64243 | 300 | 2312 |
| | I64244 | NQ | 85 |
| | I64245 | 269 | 1962 |
| | I64246 | 249 | 262 |
| | I64247 | 227 | 143 |

[a] Titration at which the sample crosses the cut point (Relative O.D. = 4 ug/mL)

**Study title: GSK716155: Investigative study to assess the effects of GSK716155 on monkey pancreas using stereologic methods to estimate cell number and volume of acinar, ductal and islet cells (GSK Study No. I11223).**

| | |
|---|---|
| Study no.: | 2011N126413 |
| Study report location: | eCTD 4.2.3.7.7 |
| Conducting laboratory and location: | GlaxoSmithKline, Safety Assessment, King of Prussia, PA |
| Date of study initiation: | 17-OCT-2011 |
| GLP compliance: | No (see note, below) |
| QA statement: | No (see note, below) |
| Note: | This is a post-hoc adjunct study conducted in response to an FDA request on pancreas samples collected from the 52-week monkey study (#G07396), which was conducted under GLP. |

**Key Study Findings**

- In the 52-week monkey study (study # G07396 [CD2009/00863]) albiglutide was associated with increased pancreas weight, especially at HD. In response to this finding the FDA requested a stereological assessment of which cellular populations were responsible for the increase in mass as well as specialized immunohistochemistry stain(s) to assess which cell populations (if any) are undergoing an increased rate of proliferation.

- There was no discernible effect of 50 mg/kg/week albiglutide on cell proliferation as assessed by Ki67 immunostaining.
- The quantitative assessment of the cell types contributing to the increase in pancreas mass is difficult to interpret due to large interindividual variation and low sample number. There also may be sex-based differences in the basis of the enlargement of the pancreas, which limit the utility of combining sexes to increase the sample size.
    - Stereology volume changes in ♂s (50 mg/kg/week vs. Control):
        - Total volume: ↑36%
        - Acinar volume: ↑61%
        - Duct volume: ↑10%
        - Islet volume: ↓30%
    - Stereology volume changes in ♀s (50 mg/kg/week vs. Control):
        - Total volume: ↑13%
        - Acinar volume: ↑6%
        - Duct volume: ↑16%
        - Islet volume: ↑20%

Methods

Unique study design: Pancreas from Group 1 vehicle control (5♂/5♀) and Group 3 given GSK716155 at a dose of 50mg/kg week (5♂/5♀) on GSK study number G07396 was evaluated in this study.

Evaluations: Tissue sections were deparaffinized and stained with specific antibodies to islets (glucagon [α], insulin [β], somatostatin [δ] & pancreatic polypeptide [γ]), duct cells (cytokeratin) and Ki67 (proliferation marker) under optimized conditions. A frationator-proportionator sampling principle was used to obtain estimates of total cell number and volume of acinar, duct and islet compartments.[1]

**Observations and Results**

[1] Gardi JE, Nyengarrd JR, Gundersen HJ. "The proportionator: unbiased stereological estimation using biased automatic image analysis and non-uniform probability proportional to size sampling." *Comput Biol Med.* 2008;**38**(3):13-28.



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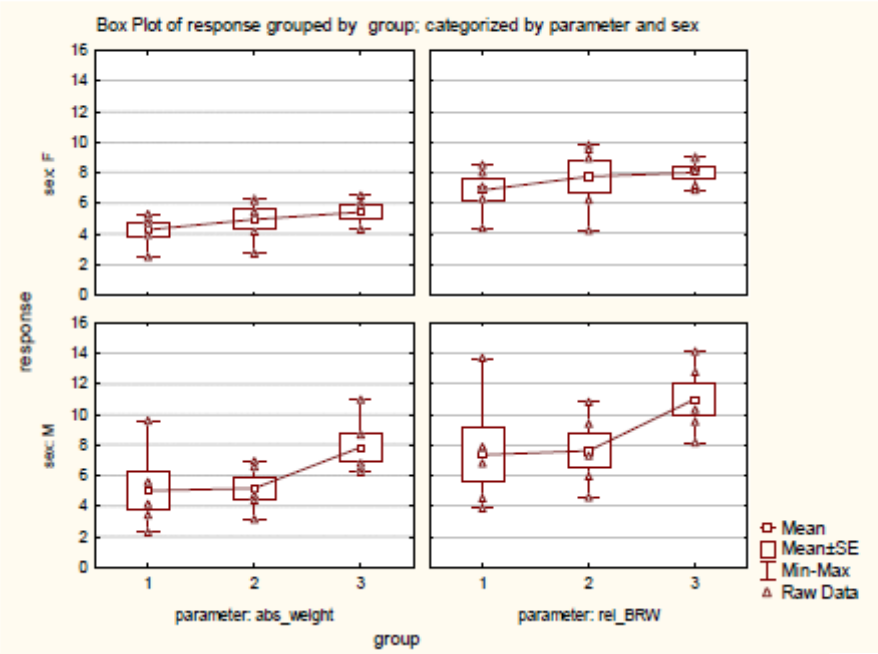
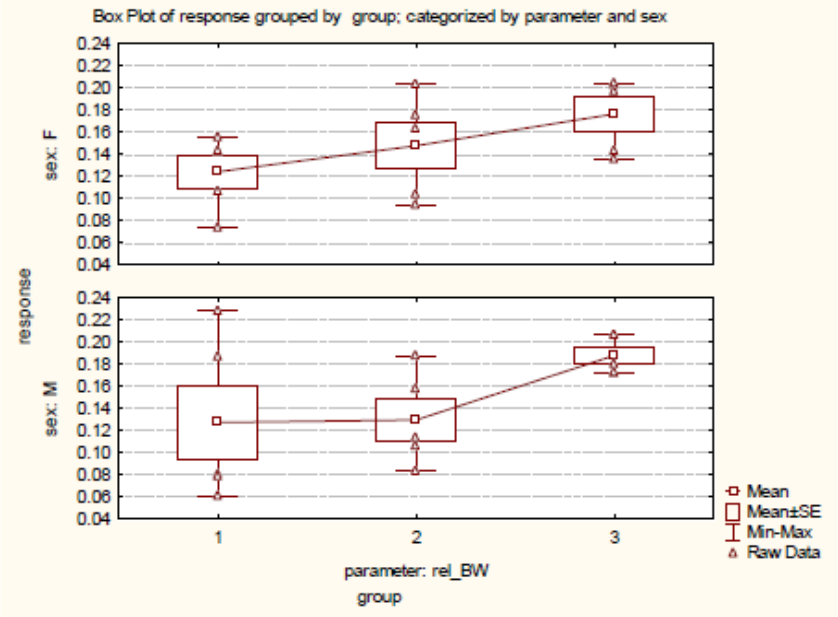
Sponsors Analysis of Pancreas Weights

## 2.1. Results of the Analysis Data display (Box plot)




Monkeys in Groups 1, 2 and 3 were given 0 (vehicle), 15 or 50 mg/kg/week GSK716155 for 52 weeks. The parameters tested were absolute pancreas tissue weight (abs_weight) and pancreas weight relative to brain weight (rel_BRW) [top graph] and pancreas weight relative to body weight (rel_BW) [bottom graph]. Graphs shows group means (small square), ± standard error (box), minimum and maximum (whiskers), and individual values (triangles).

Qualitative Assessment of Proliferating Cells

There were no test item-related effects on the percentage of cells staining positive for Ki-67. All animals showed a variable degree of small numbers of Ki67 positive cells, including acinar cells, ducts/ductules and/or islet cells.

Quantitative Assessment of Cell Types

Because of the large interindividual variability (see Sponsor's Table 3) and low number of animals per dose group of each sex (5/s/g) it was generally difficult to establish statistical significance of observed changes. There also appeared to be possible differences between males and females with regard to which cell populations were most affected by the test item (see Sponsor's Table 2).

Changes achieving s.s. were 1) islet cell number (but not islet volume) in ♀ (p=0.035) and pooled ♂ & ♀ (p=0.042), but not in ♂s alone (p=0.494); 2) volume of acinar (but not number of acinar cells) in ♂s alone (p=0.039), but not pooled (p=0.052) or ♀s alone (p=0.658).

**Table 2 Mean Estimates of Acinar, Duct, Islet total cell number and compartment volume, Observed Coefficient of Variation (OCV) and Coefficient of Error (CE)**

| | Group | N(Is)†$10^6$ | V(Is)† $mm^3$ | N(Du)†$10^6$ | V(Du)† $mm^3$ | N(Ac)† $10^6$ | V(Ac)† $mm^3$ | V(Ti)† $mm^3$ |
|---|---|---|---|---|---|---|---|---|
| Mean Estimate | 1M | 39.68 | 36.93 | 260.45 | 327.05 | 530.48 | 975.64 | 1705.74 |
| OCV | | 0.52 | 0.55 | 0.44 | 0.83 | 0.51 | 0.46 | 0.77 |
| CE | | 0.12 | 0.23 | 0.05 | 0.05 | 0.06 | 0.08 | 0.17 |
| | | | | | | | | |
| Mean Estimate | 3M | 43.67 | 25.99 | 353.88 | 360.99 | 738.48 | 1567.07 | 2315.77 |
| OCV | | 0.21 | 0.19 | 0.10 | 0.20 | 0.17 | 0.21 | 0.16 |
| CE | | 0.17 | 0.18 | 0.09 | 0.11 | 0.12 | 0.10 | 0.17 |
| % Change 3M/1M | | ↑10% | ↓30% | ↑36% | ↑10% | ↑39% | ↑61% | ↑36% |
| Mean Estimate | 1F | 32.38 | 26.25 | 278.76 | 259.21 | 574.58 | 1059.78 | 1455.19 |
| OCV | | 0.30 | 0.37 | 0.36 | 0.39 | 0.30 | 0.31 | 0.32 |
| CE | | 0.11 | 0.18 | 0.09 | 0.09 | 0.08 | 0.08 | 0.08 |
| % Change 3F/1F | | ↑70% | ↑20% | ↓4% | ↑16% | ↓4% | ↑6% | ↑13% |
| Mean Estimate | 3F | 55.10 | 31.19 | 268.57 | 300.25 | 548.89 | 1119.76 | 1641.83 |
| OCV | | 0.36 | 0.26 | 0.26 | 0.18 | 0.22 | 0.21 | 0.21 |
| CE | | 0.21 | 0.15 | 0.08 | 0.08 | 0.12 | 0.05 | 0.07 |

† N(Is) indicates Islet Cell Number; V(Is) indicates Islet Compartment Volume; N(Du) indicates Duct Cell Number; V(Du) indicates Duct Compartment Volume; N(Ac) indicates Acinar Cell Number; V(Ac) indicates Acinar Compartment Volume; V(Ti) indicates Tissue Volume

**Table 3 Individual Animal Estimates of Acinar, Duct, Islet total cell number and compartment volume**

| Animal No | Group | N(Is)† | V(Is)† mm³ | N(Du)† | V(Du)† mm³ | N(Ac)† | V(Ac)† mm³ | V(Ti)† mm³ |
|---|---|---|---|---|---|---|---|---|
| I64208 | 1M | 18556892.25 | 17.70294708 | 193748628.3 | 189.6501648 | 277054734 | 541.0687149 | 780.377175 |
| I64209 | 1M | 33470312.12 | 51.41212001 | 176107388.1 | 129.1061563 | 298387917.6 | 655.2163276 | 1038.730162 |
| I64210 | 1M | 67299289.25 | 55.33748393 | 438569011.7 | 794.3876641 | 899995219.3 | 1658.764343 | 4006.492505 |
| I64211 | 1M | 54210165.67 | 48.20091718 | 310492042.3 | 321.7459411 | 715256624.9 | 1183.170517 | 1594.922619 |
| I64212 | 1M | 24873379.59 | 12.00418291 | 183331476.5 | 200.3550227 | 461702368 | 839.9591154 | 1108.17619 |
| | | | | | | | | |
| I64218 | 3M | 47577404.06 | 23.20377675 | 370119128.4 | 335.8258934 | 742696143.9 | 1483.659553 | 2127.564399 |
| I64219 | 3M | 42015659.02 | 30.94169337 | 387293570.8 | 387.0177227 | 748208083.6 | 1569.749471 | 2113.851922 |
| I64220 | 3M | 47896194.38 | 20.77382806 | 379582733.4 | 429.5499784 | 920555367.3 | 2125.106895 | 2661.58334 |
| I64221 | 3M | 52053887.2 | 23.38504882 | 309820181.4 | 406.1448242 | 581524810.1 | 1313.578013 | 1903.003941 |
| I64222 | 3M | 28784304.62 | 31.6296071 | 322585465.8 | 246.4281996 | 699395585.8 | 1343.278077 | 2772.859942 |
| | | | | | | | | |
| I64228 | 1F | 36699317.77 | 30.35604213 | 295448753.7 | 299.2573971 | 610059224.1 | 1135.816054 | 1487.003319 |
| I64229 | 1F | 44878235.01 | 26.72962633 | 290667265.4 | 335.1264863 | 595122617.1 | 1179.788531 | 1867.145132 |
| I64230 | 1F | 24935244.86 | 25.17642978 | 320799944.6 | 229.0199224 | 573442541.6 | 1211.666654 | 1601.582454 |
| I64231 | 1F | 35061391.08 | 37.66271617 | 376507327.9 | 337.7005299 | 788293551.8 | 1284.162311 | 1662.254948 |
| I64232 | 1F | 20323832.87 | 11.33898427 | 110356502.4 | 94.96644198 | 305974080.1 | 487.4908063 | 657.975698 |
| | | | | | | | | |
| I64238 | 3F | 85274993.7 | 29.50963601 | 280183444.2 | 297.4955486 | 598965669.6 | 1459.288363 | 1847.325557 |
| I64239 | 3F | 43526007.93 | 19.55891034 | 196903472.2 | 265.5518528 | 462133785.3 | 958.8448508 | 1315.531996 |
| I64240 | 3F | 39402713.7 | 42.11769291 | 336706906.9 | 318.894766 | 501031936.2 | 993.8201155 | 1647.519896 |
| I64241 | 3F | 65161367.73 | 34.14877637 | 334258751.7 | 380.8052888 | 735981879.7 | 1268.926029 | 2090.868234 |
| I64242 | 3F | 42113542.64 | 30.64212722 | 194802749.1 | 238.5218357 | 446353921 | 917.9430808 | 1307.908546 |

† N(Is) indicates Islet Cell Number; V(Is) indicates Islet Compartment Volume; N(Du) indicates Duct Cell Number; V(Du) indicates Duct Compartment Volume; N(Ac) indicates Acinar Cell Number; V(Ac) indicates Acinar Compartment Volume; V(Ti) indicates Tissue Volume

# 7 Genetic Toxicology

## 7.1 *In Vitro* Reverse Mutation Assay in Bacterial Cells (Ames)

No studies conducted.

## 7.2 *In Vitro* Assays in Mammalian Cells

No studies conducted.

## 7.3 *In Vivo* Clastogenicity Assay in Rodent (Micronucleus Assay)

No studies conducted.

## 7.4 Other Genetic Toxicity Studies

No studies conducted.

# 8 Carcinogenicity

No studies conducted. The following is reproduced from section 4.5 of Sponsor's Nonclinical Overview:

Carcinogenicity studies with albiglutide have not been conducted in rodents due to emergence of clearing ADA that limit duration of rodent studies to approximately 2 weeks. Since meaningful carcinogenicity studies are not possible in rodents due to immunogenicity, assessment of the carcinogenic potential of albiglutide has been based on a weight-of-evidence approach.

Other GLP-1R agonists have produced thyroid C-cell proliferative changes in mice and rats, including focal C-cell hyperplasia, adenomas and carcinomas indicating that there could be a potential for albiglutide to also produce thyroid C-cell tumors in mice. There appears to be some relationship with duration of activity, in that the incidence and severity of tumors in rodent carcinogenicity studies was increased with long acting GLP-1R agonists [liraglutide, once weekly exenatide (Bydureon™)] as compared to twice-daily exenatide (Byetta™). Furthermore focal C-cell hyperplasia was produced when exenatide was given to mice as continuous subcutaneous infusion for 12 or 13 weeks, but not when the same total dose was given as once daily injections [Knudsen, 2010; Madsen, 2012]. However, C-cell hyperplasia was not reported after 12 weeks continuous infusion with another GLP-1R agonist, lixisenatide, suggesting that duration of activity on GLP-1R alone is not sufficient to produce the effect [Kissner, 2012].

The mechanism of thyroid C-cell tumors in rodents and their clinical relevance are unclear. Increased sensitivity in rodents has been attributed to relatively high expression levels of GLP-1R on thyroid C-cells in rodents [Knudsen, 2010; Madsen, 2012]. In monkeys, like humans, C-cell expression of GLP-1R is relatively low or undetectable [Knudsen, 2010; Gier, 2012; Waser, 2011]. Consistent with reported differences in C-cell GLP-1R expression patterns, liraglutide elevated plasma calcitonin levels in mice and rats, but not humans or monkeys [Knudsen, 2010; Madsen, 2012]. With albiglutide, there were no clinically meaningful differences or changes in calcitonin levels from placebo or the all comparators group [m2.5, Section 5.1.2.6.2]. Furthermore, liraglutide and continuous exenatide infusion did not elevate plasma calcitonin levels or produce focal C-cell hyperplasia in GLP-1R knockout mice [Madsen, 2012]. The proliferative C-cell changes induced by liraglutide in mice were associated with activation of the mechanistic target of rapamycin (mTOR) pathway, but not the rearranged during transfection (RET) pathway [Madsen, 2012] which is strongly associated in human medullary thyroid cancer.

While the effects of albiglutide on thyroid C-cell proliferation have not been determined in rodents, there were no albiglutide-related findings in thyroids or parathyroids of monkeys given up to 50 mg/kg/week for up to 52 weeks [m2.6.7, Table 7.6, Report CD2009/00863]. In the 52 week monkey study, thyroids were thoroughly examined for C-cell effects. There were no changes in thyroid C-cells based on qualitative assessment of standard hematoxylin and eosin (H&E) and calcitonin immunohistochemistry of sections taken throughout the thyroid and parathyroid. In



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addition the entire thyroid was sampled by systematic uniform random disector pairs and total estimates of C-cells were obtained by stereological quantitation. The study did not detect any differences between the mean estimates of total C-cell number in monkeys given 50 mg/kg/week albiglutide compared to vehicle control.

The data from the 52 week monkey study indicate that albiglutide, at exposures 75 times the highest clinical exposure based on AUC, does not induce thyroid C-cell proliferation in monkeys. These findings are consistent to those reported for liraglutide, where no C-cell hyperplasia was observed in monkey studies of durations up to 87 weeks [Knudsen, 2010]. Furthermore, the integrated Phase III data, including data from randomized, blinded, long term (through 2 years) evaluations, suggest no increased risk of thyroid cancer following treatment with albiglutide [m2.5, Section 5.1.2.6.2]. However, these monkey data do not negate the C-cell proliferative effects identified in rodent carcinogenicity studies with other GLP-1R agonists, the significance of which to human risk is unknown. Careful consideration should be given to the risks and benefits of treatment with albiglutide in patients with a personal or family history of medullary thyroid carcinoma (MTC) or in patients with multiple endocrine neoplasia syndrome type 2 (MEN 2) [m1.14.1.2 (US) or m1.3 (EU)].

# 9 Reproductive and Developmental Toxicology

## 9.1 Fertility and Early Embryonic Development

**Study title: GSK716155: Subcutaneous Male Fertility Study in Mice**

| | |
|---|---|
| Study no.: | CD2007/01279 (b) (4) |
| Study report location: | eCTD 4.2.3.5.1 |
| Conducting laboratory and location: | (b) (4) |
| Date of study initiation: | 26 Mar 2007 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- There were no drug-related effects on male reproductive performance (mating, fertility, or copulation indices as well as the numbers of days needed to mate), male reproductive organs, or embryonic survival. The NOAEL for effects on male mating fertility is considered to be 50 mg/kg/day.
- Expected effects on FC (↓) and BW (↓) were seen in MD and HD ♂s during the first week of dosing, consistent with the intended pharmacology of albiglutide. LD was without effect on these parameters.



Reference ID: 3347159

Methods

| | |
|---|---|
| Doses: | 0, 5, 15, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | Aqueous soln. in 10 mM sodium phosphate, 2.8% (w/v) mannitol, 4.2% (w/v) trehalose, and 0.01% polysorbate 80 (v/v), pH 7.2 |
| Species/Strain: | CD1 mice |
| Number/Sex/Group: | 25♂/group |
| Satellite groups: | 15♂/group |
| Study design: | Groups of 25 male CD1 mice were treated via bolus SC injection with vehicle or GSK716155 as described above, once daily beginning on Day 0 of the study and continuing until termination (15 to 17 doses). Males were treated for 7 days and then cohabited 1:1 with untreated females for a maximum of 7 additional days. Mated females were separated from the males and considered to be on DG 0. The premating and mating treatment periods were limited to 7 days each (14 days total) because TK studies have shown that drug exposure peaks by day 7 of dosing and then starts to decline due to the induction of anti-GSK716155 antibodies. Mated females and their litters were euthanized on gestation Day 18 and the uterine contents examined. |
| Deviation from study protocol: | None material to interpretation of the study. |

**Observations and Results**

**Mortality**

No early deaths.

**Clinical Signs**

No test item-related effects.

**Body Weight**

S.s. weight loss at MD and HD during premating dosing period (mean body weights were ~ 0.96X and 0.92X baseline and concurrent control at MD & HD [respectively]). During the mating period (Days 7 to 15), BW in the MD was similar to the controls. By the end of the mating period BW in the HD groups was also similar to controls. Body weight in the LD was unaffected at any treatment period.



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**Feed Consumption**

S.s. reductions in mean FC were observed in MD & HD ♂s (0.86X and 0.83X C♂s, respectively) during the premating period (Days 0 to 6) when compared to the control group, correlating with mean body weight losses during this same interval. Mean FC in LD♂s was similar to C♂s.

**Toxicokinetics**

Plasma exposure was measured following 7 days of daily SC administration of albiglutide for a 24 hour period. Dose-proportional increases in exposure were seen at all 3 dose levels. All control animal plasma samples were below the lower limit of quantitation.

| Parameter | Period | Male (n = 3/timepoint/group) | | |
|---|---|---|---|---|
| | | Dose of GSK716155 (mg/kg/day) | | |
| | | 5 | 15 | 50 |
| $AUC_{0-t}$ (µg·h/mL) | Day 7 | 344 | 916 | 3601 |
| $C_{max}$ (µg/mL) | Day 7 | 18.1 | 46.0 | 185 |
| $T_{max}$ (h) | Day 7 | 6.00 | 6.00 | 6.00 |

**Necropsy**

Unremarkable.

**Fertility Parameters (Mating/Fertility Index, Corpora Lutea, Preimplantation Loss, etc.)**

There were no test article-related effects on the mean number of days between pairing and coitus or male mating index, fertility index, or copulation index at any dose level. Nor was there any test item-related effect on the weights or morphology of male reproductive tissues.

There were no test article-related effects on intrauterine growth and survival, placental morphology, or gravid uterine weights in any dose group.

There were no test article-related effects on external fetal morphology.

**Study title: GSK716155: Subcutaneous Female Fertility Study In Mice**

| | |
|---|---|
| Study no.: | CD2006/01879 (b) (4) -472100) |
| Study report location: | eCTD 4.2.3.5.1 |
| Conducting laboratory and location: | (b) (4) |
| Date of study initiation: | 21 FEB 2006 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- Transient body weight loss and food intake reduction were evident in HD♀s.
- HD♀s had a n.s.s. ↑ in the mean estrus cycle length and a concordant ↓ in the number of estrus cycles/week. These findings were also correlated with a n.s.s. ↓ in fertility index in HD♀s.
- No other findings regarding embryofetal survival or fetal external morphology were seen at any dose level tested.
- The NOAEL is considered to be 5 mg/kg/day for female reproductive toxicity.



Reference ID: 3347159

Methods

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | Aqueous soln. in 10 mM sodium phosphate, 2.8% (w/v) mannitol, 4.2% (w/v) trehalose, and 0.01% polysorbate 80 (v/v), pH 7.2 |
| Species/Strain: | CD1 mice |
| Number/Sex/Group: | 25 ♀/group |
| Satellite groups: | none |
| Study design: | Groups of 25 female CD1 mice were treated daily via bolus SC injection with vehicle or the indicated doses of GSK716155 for 7 days prior to cohabiting 1:1 with untreated males. Daily dosing of females continued until positive evidence of mating was observed, or for up to 14 total doses. Animals were not dosed during presumed gestation period. Mated females and their litters were euthanized on gestation Day 18 and the uterine contents examined. |
| Deviation from study protocol: | None material to interpretation of the study. |
| Endpoints: | Mated females: mortality, clinical observations, body weight, food consumption, estrous cycle, mating, fertility, necropsy, corpora lutea and uterine weight<br>Litters: implantations, resorptions, live and dead fetuses, fetal weight, sex, and placental and fetal external morphology. |

**Observations and Results**

**Mortality**

All females survived to the scheduled necropsies.

**Clinical Signs**

A transient decrease in feces production occurred in 4 of 25 females after the second dose at 50 mg/kg/day, compared to 0 of 25 females in the control group. The decrease only lasted 1 day but was considered test item-related because of a concurrent loss in body weight by Day 3 of dosing (study Day 2) and decreased mean food consumption during Week 1 of dosing.



Reference ID: 3347159

### Body Weight

There were transient decreases in BW at HD compared both to baseline and concurrent controls (~5 to 7% ↓) over the first week of dosing. These did not persist into the gestation period.

There was no drug-related effect on mean body weights or cumulative BW gain in the LD or MD groups.

### Feed Consumption

There was a s.s. 19% ↓ in FC at HD during the premating period, compared to the C group. This effect correlates with the reduction of BW decreased feces production seen in this dose group.

There were no drug-related effects on FC in the LD or MD groups.

### Toxicokinetics

Not performed in this study. Previous studies (CD2006/01620 & RD2005/01134) would predict dose-proportional increases in exposure across all dose levels following the first dose, and hyperproportional increases between 5 and 50 mg/kg/day after a week of dosing (see tabulated summary in PK section above).

### Necropsy

Unremarkable.

### Fertility Parameters (Mating/Fertility Index, Corpora Lutea, Preimplantation Loss, etc.)

The mean estrus cycle length was n.s.s. ↑ at HD (5.7 days vs. 4.8 days for C), resulting in a n.s.s. ↓ in the mean number of estrus periods during the 7-day dosing period at HD (0.9 estrus per week vs 1.3 in C). These differences appear to stem from an increase in the number of ♀s with prolonged diestrus (≥5 consecutive days ) – 9/25 HD♀s vs. 1/25 C♀s.

TABLE 12

PROJECT NO. (b) (4)472100F GSK716155: SUBCUTANEOUS FEMALE FERTILITY STUDY IN MICE PA(
SPONSOR:GLAXOSMITHKLINE SUMMARY OF ESTROUS CYCLE DATA
SPONSOR NO.:G05438

| | FEMALES | | | |
|---|---|---|---|---|
| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
| ESTROUS CYCLE LENGTH (DAYS) | | | | |
| MEAN | 4.8 | 4.4 | 4.3 | 5.7 |
| S.D. | 0.97 | 0.70 | 0.67 | 0.95 |
| S.E. | 0.32 | 0.23 | 0.21 | 0.36 |
| N | 9 | 9 | 10 | 7 |

MODIFIED STATISTICS USED. * INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
None significantly different from control group

TABLE 13

PROJECT NO. (b) (4) 472100F
SPONSOR:GLAXOSMITHKLINE
SPONSOR NO.:G05438

GSK716155: SUBCUTANEOUS FEMALE FERTILITY STUDY IN MICE
SUMMARY OF ESTROUS CYCLE DATA BY WEEK

PAG

| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
|---|---|---|---|---|
| | | FEMALES | | |
| NUMBER OF ESTROUS CYCLES | | | | |
| PRETEST WEEK 1 | | | | |
| MEAN | 1.7 | 1.7 | 1.8 | 1.6 |
| S.D. | 0.46 | 0.46 | 0.44 | 0.49 |
| S.E. | 0.09 | 0.09 | 0.09 | 0.10 |
| N | 25 | 25 | 25 | 25 |
| PRETEST WEEK 2 | | | | |
| MEAN | 1.6 | 1.5 | 1.4 | 1.6 |
| S.D. | 0.49 | 0.51 | 0.50 | 0.65 |
| S.E. | 0.10 | 0.10 | 0.10 | 0.13 |
| N | 25 | 25 | 25 | 25 |
| TEST WEEK | | | | |
| MEAN | 1.3 | 1.4 | 1.4 | 0.9 |
| S.D. | 0.46 | 0.50 | 0.65 | 0.49 |
| S.E. | 0.09 | 0.10 | 0.13 | 0.10 |
| N | 25 | 25 | 25 | 25 |

None significantly different from control group using Dunnett's test

A Slight, n.s.s. ↓ in fertility index was observed at HD vs. C.

TABLE 11

PROJECT NO.: (b) (4) 472100F
SPONSOR:GLAXOSMITHKLINE
SPONSOR NO.:G05438

GSK716155: SUBCUTANEOUS FEMALE FERTILITY STUDY IN MICE
SUMMARY OF FEMALE REPRODUCTIVE PERFORMANCE

PAGE 2

| DOSE GROUP : | 1 | | 2 | | 3 | | 4 | |
|---|---|---|---|---|---|---|---|---|
| | NO. | % | NO. | % | NO. | % | NO. | % |
| FEMALE MATING INDEX | 25/25 | 100.0 | 24/25 | 96.0 | 25/25 | 100.0 | 23/25 | 92.0 |
| FEMALE FERTILITY INDEX | 25/25 | 100.0 | 23/25 | 92.0 | 25/25 | 100.0 | 21/25 | 84.0 |
| FEMALE CONCEPTION INDEX | 25/25 | 100.0 | 23/24 | 95.8 | 25/25 | 100.0 | 21/23 | 91.3 |
| MEAN PRE-COITAL INTERVALS (DAYS) | 2.0 | NA | 2.3 | NA | 2.4 | NA | 2.7 | NA |
| S.D. | 1.11 | NA | 1.09 | NA | 1.08 | NA | 1.37 | NA |
| S.E. | 0.24 | NA | 0.23 | NA | 0.22 | NA | 0.29 | NA |
| N | 22 | | 22 | | 23 | | 23 | |

$$\text{FEMALE MATING INDEX (\%)} = \frac{\text{NO. OF FEMALES WITH EVIDENCE OF MATING (OR FEMALES CONFIRMED PREGNANT)}}{\text{TOTAL NO. OF FEMALES USED FOR MATING}} \times 100$$

$$\text{FEMALE FERTILITY INDEX (\%)} = \frac{\text{NO. OF FEMALES WITH CONFIRMED PREGNANCY}}{\text{TOTAL NO. OF FEMALES USED FOR MATING}} \times 100$$

$$\text{FEMALE CONCEPTION INDEX (\%)} = \frac{\text{NO. OF FEMALES WITH CONFIRMED PREGNANCY}}{\text{TOTAL NO. OF FEMALES WITH EVIDENCE OF MATING (OR CONFIRMED PREGNANCY)}} \times 100$$

1- 0 MG/KG/DAY 2- 1 MG/KG/DAY 3- 5 MG/KG/DAY 4- 50 MG/KG/DAY
PRE-COITAL INTERVALS NOT SIGNIFICANTLY DIFFERENT FROM THE CONTROL GROUP; MODIFIED STATISTICS USED.
MATING, FERTILITY AND CONCEPTION INDICES NOT SIGNIFICANTLY DIFFERENT FROM THE CONTROL GROUP USING CHI-SQUARE TEST; IN ADDITION, FERTILITY INDICES NOT SIGNIFICANTLY DIFFERENT FROM THE CONTROL GROUP USING 1-TAILED FISHER'S EXACT TEST
NA = NOT APPLICABLE

There were no test item-related effects on corpora lutea, embryofetal survival, fetal weight or gravid uterine weight at any dose level. Nor were there any effects on fetal external malformations or variations at any dose level.

## 9.2 Embryonic Fetal Development

*NB – because of the rapid induction of drug-clearing, neutralizing ADAs in rodents, Sponsor has used alternative study designs to limit exposure to less than 2 weeks.*

**Study title: GSK716155: Subcutaneous Early Embryonic to Implantation Reproductive Toxicity Study in Mice**

| | |
|---|---|
| Study no.: | CD2006/01198 (b) (4) -472098) |
| Study report location: | eCTD 4.2.3.5.1 |
| Conducting laboratory and location: | (b) (4) |
| Date of study initiation: | 15 FEB 2006 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- Drug-related, systemic toxicity in the pregnant females was observed at 50 mg/kg/day and consisted of mean body weight loss and reduced food consumption relative to controls.
- No remarkable abnormalities were recorded for early embryonic and implantation parameters (GD0 to GD6). The NOAEL for early embryonic development to implantation was considered to be 50 mg/kg/day (the highest dose tested).

Methods

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | Aqueous soln. in 10 mM sodium phosphate, 2.8% (w/v) mannitol, 4.2% (w/v) trehalose, and 0.01% polysorbate 80 (v/v), pH 7.2 |
| Species/Strain: | CD1 mice |
| Number/Sex/Group: | 22 mated ♀/group |
| Satellite groups: | none |
| Study design: | Groups of 22 mated female CD1 mice were treated daily via bolus SC injection with vehicle or the indicated doses of GSK716155 from Gestation Day 0 to 6. Dams and litters were euthanized on GD 18. |
| Deviation from study protocol: | None material to interpretation of the study. |
| Endpoints: | Mated females: mortality, clinical observations, body weight, food consumption, necropsy, corpora lutea and uterine weight<br>Litters: implantations, resorptions, live and dead fetuses, fetal weight, sex, and placental and fetal external morphology. |



Reference ID: 3347159

**Observations and Results**

**Mortality**

No test item-related effects.

**Clinical Signs**

No test item-related effects.

**Body Weight**

S.s. loss in mean maternal body weight (-1.3 g), relative to a mean gain in controls (+0.2 g) was evident between Days 0 to 3 PC at 50 mg/kg/day. BW gain was reduced (12%) at 50 mg/kg/day compared to the vehicle control group over the 7-day dosing period (Days 0 to 7 PC). Following cessation of dosing, BW gain was ↑ in HD♀s, relative to C, and all dose groups had comparable mean cumulative body weight gains and body weights over the period of the study (Days 0 to 18 PC).

There were no test article-related effects on mean body weights or cumulative body weight gains at 1 and 5 mg/kg/day.

**Feed Consumption**

S.s. ↓ FC in mean daily FC was noted in HD♀s (2.8 g) compared to C♀s (4.8 g) from Day 0 to 3 PC. FC at HD remained below C values (controls 6.4 g vs. 5.2 g in HD group) for the remainder of the dosing period (Days 3 to 7 PC). These reductions in FC at HD correspond to the effects of HD albiglutide on BW.

There were no effects on FC at 1 and 5 mg/kg/day.

**Toxicokinetics**

Not performed in this study. Previous studies (CD2006/01620 & RD2005/01134) would predict dose-proportional increases in exposure across all dose levels following the first dose, and hyperproportional increases between 5 and 50 mg/kg/day after a week of dosing (see tabulated summary in PK section above).

**Necropsy**

No test item-related effects.

**Cesarean Section Data (Implantation Sites, Pre- and Post-Implantation Loss, etc.)**

There were no test article-related effects on embryo/fetal survival, fetal weight or gravid uterine weight at any dose level.

**Offspring (Malformations, Variations, etc.)**

There were no test article-related effects on malformations or variations.



Reference ID: 3347159

**Study title: GSK716155: Subcutaneous Embryo/Fetal Development Study in Mice**

| | |
|---|---|
| Study no.: | CD2006/01353 (b) (4) 472097) |
| Study report location: | eCTD 4.2.3.5.2 |
| Conducting laboratory and location: | (b) (4) |
| Date of study initiation: | 2 MAR 2006 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- When administered daily by SC injection to pregnant mice on Days 6 through 15 post coitus, albiglutide was associated with maternal toxicity (↓ FC and ↓ BW gain (frank BW loss following the 1st dose) at a dose of 50 mg/kg (HD). HD mice had lower body weights throughout the treatment period.
- The HD was also asso*ciated with embryo/fetal toxicity, as evidenced by embryo/fetal lethality (e.g.*, 16.2% per litter postimplantation loss compared to 3.8% per litter in the control group).
- The HD was associated with a s.s. higher mean litter proportion of the skeletal developmental variation bent (wavy) ribs (8.0% per litter).
- The NOAEL for effects on embryo/fetal development in mice was considered to be 5 mg/kg/day.

Methods

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | Aqueous soln. in 10 mM sodium phosphate, 2.8% (w/v) mannitol, 4.2% (w/v) trehalose, and 0.01% polysorbate 80 (v/v), pH 7.2 |
| Species/Strain: | CD1 mice |
| Number/Sex/Group: | 22 mated ♀/group |
| Satellite groups: | none |
| Study design: | Groups of 22 mated female CD1 mice were treated daily via bolus SC injection with vehicle or the indicated doses of GSK716155 from Gestation Day 6 to 15. Dams and litters were euthanized on GD 18. |
| Deviation from study protocol: | None material to interpretation of the study. |
| Endpoints: | Mated females: mortality, clinical observations, body weight, food consumption, necropsy, corpora lutea and uterine weight<br>Litters: implantations, resorptions, live and |



Reference ID: 3347159

dead fetuses, fetal weight, sex, and placental and fetal external morphology.

## Observations and Results

### Mortality

No test item-related effect.

### Clinical Signs

No test item-related effect.

### Body Weight

S.s. ↓ in BW gain at HD, especially during the first couple of days after dosing. The HD group actually lost weight between Day 6 and 7 (-1.1 g vs. + 0.6 g for C).

No clear effect of the test item on BW gain at the lower doses.

TABLE 6

PROJECT NO. (b) (4) 472097
SPONSOR: GLAXOSMITHKLINE
SPONSOR NO.:G06020

GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SUMMARY OF BODY WEIGHTS DURING GESTATION [G]

| | | Dose Group (mg/kg/day) | | | |
|---|---|---|---|---|---|
| Subinterval | Statistic | 0 | 1 | 5 | 50 |
| 6-9 | N | 21 | 21 | 20 | 18 |
| | Mean (Change) | 1.8048 | 2.0714 | 1.6850 | 0.1056 |
| | S.D. | 0.5766 | 0.5515 | 0.5060 | 0.7885 |
| | S.E. of Mean | 0.1258 | 0.1203 | 0.1132 | 0.1859 |
| | Bonferroni Adjusted p-value++ | | 1.000 | 1.000 | <0.001* |
| 6-13 | N | 21 | 21 | 20 | 18 |
| | Mean (Change) | 9.3333 | 9.5952 | 8.8400 | 7.7111 |
| | S.D. | 1.5860 | 1.4709 | 1.9121 | 1.8566 |
| | S.E. of Mean | 0.3461 | 0.3210 | 0.4276 | 0.4376 |
| | Bonferroni Adjusted p-value++ | | 1.000 | 1.000 | 0.007* |
| 6-16 | N | 21 | 21 | 20 | 18 |
| | Mean (Change) | 17.681 | 18.129 | 16.835 | 16.072 |
| | S.D. | 3.059 | 2.655 | 3.519 | 2.899 |
| | S.E. of Mean | 0.668 | 0.579 | 0.787 | 0.683 |
| | Bonferroni Adjusted p-value++ | | 1.000 | 0.485 | 0.029* |
| OVERALL | Dunnett's Test++ | | NT | NT | NT |
| INTN | Trt*Time Interaction p-value# | | | | <0.001* |
| | Covariance Structure | | | | AR1 |

# : Level of Significance tested = 0.10. NA : Not available. * : Statistically Significant.
++ : Level of Significance tested = 0.05. NT = Not tested due to the outcome of preliminary test(s).

### Feed Consumption

S.s. ↓ in FC at HD. No clear effect of test item at lower doses.

TABLE 8

PROJECT NO. (b) (4) 472097
SPONSOR: GLAXOSMITHKLINE
SPONSOR NO.:G06020

GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SUMMARY OF FOOD CONSUMPTION DURING GESTATION [G/ANIMAL/DAY]

| | | Dose Group (mg/kg/day) | | | |
|---|---|---|---|---|---|
| Subinterval | Statistic | 0 | 1 | 5 | 50 |
| 6-9 | N | 21 | 20 | 20 | 18 |
| | Mean | 6.1952 | 6.0800 | 6.2350 | 4.2556 |
| | S.D. | 0.8743 | 0.9328 | 1.6831 | 1.6364 |
| | S.E. of Mean | 0.1908 | 0.2086 | 0.3764 | 0.3857 |
| | Bonferroni Adjusted p-value++ | | 1.000 | 1.000 | <0.001* |
| 9-12 | N | 21 | 21 | 20 | 18 |
| | Mean | 6.5619 | 6.2190 | 5.8850 | 5.2778 |
| | S.D. | 0.7978 | 0.6653 | 0.8543 | 0.9807 |
| | S.E. of Mean | 0.1741 | 0.1452 | 0.1910 | 0.2312 |
| | Bonferroni Adjusted p-value++ | | 0.923 | 0.143 | <0.001* |
| 12-16 | N | 21 | 20 | 20 | 18 |
| | Mean | 7.3429 | 7.6300 | 7.3400 | 7.5222 |
| | S.D. | 0.6622 | 0.8880 | 0.5744 | 1.5746 |
| | S.E. of Mean | 0.1445 | 0.1986 | 0.1284 | 0.3711 |
| | Bonferroni Adjusted p-value++ | | 1.000 | 1.000 | 1.000 |
| OVERALL | N | 21 | 21 | 20 | 18 |
| | Mean | 6.7000 | 6.6556 | 6.4867 | 5.6852 |
| | S.D. | NA | NA | NA | NA |
| | S.E. of Mean | NA | NA | NA | NA |
| | Dunnett's Test++ | | NT | NT | NT |
| INTN | Trt*Time Interaction p-value# | | | | <0.001* |
| | Covariance Structure | | | | AR1 |

# : Level of Significance tested = 0.10. NA : Not available. * : Statistically Significant.
++ : Level of Significance tested = 0.05. NT = Not tested due to the outcome of preliminary test(s).

### Toxicokinetics

Not performed.

### Dosing Solution Analysis

All samples were within 10% of the nominal concentration.

GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE

**Table 1: Concentration Analysis Of The 3 April 2006 Formulations**

( Analyzed 03Apr2006)

| Group | Dose Conc ( mg/mL ) | Ref # ( 472097 - ) | Analyzed Conc ( mg/mL ) | Percent of Target (%) | Mean Conc ( mg/mL ) | SD | RSD (%) | Mean Conc % of Target (%) |
|---|---|---|---|---|---|---|---|---|
| 1 Mid | 0 | 3 - 1 | Not Detected | | | | | |
| 2 Mid | 0.500 | 3 - 2 | 0.471 | 94.1 | 0.471 | 0.00059 | 0.12 | 94.2 |
| | | 3 - 3 | 0.471 | 94.3 | | | | |
| 3 Mid | 2.50 | 3 - 4 | 2.28 | 91.3 | 2.29 | 0.0068 | 0.30 | 91.5 |
| | | 3 - 5 | 2.29 | 91.7 | | | | |
| 4 Mid | 25.0 | 3 - 8 | 22.9 | 91.6 | 22.8 | 0.088 | 0.38 | 91.4 |
| | | 3 - 9 | 22.8 | 91.1 | | | | |

## Necropsy

Trend towards ↑ in the number of nongravid ♀s at MD & HD: C = 1, LD = 1, MD = 2, HD = 4.

1HD♀ aborted on GD17.

TABLE 9

PROJECT NO. (b) (4)472097 GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SPONSOR:GLAXOSMITHKLINE SUMMARY OF MATERNAL MACROSCOPIC FINDINGS
SPONSOR NO.:G06020

| GROUP : | 1 | 2 | 3 | 4 |
|---|---|---|---|---|
| NUMBER EXAMINED | 22 | 22 | 22 | 22 |
| NO SIGNIFICANT CHANGES OBSERVED | 20 | 18 | 20 | 17 |
| NONGRAVID -- AMMONIUM SULFIDE NEGATIVE | 1 | 1 | 2 | 4 |
| DELIVERED GESTATION DAY 18 | 0 | 2 | 0 | 0 |
| OVARIES: CYST(S) | 0 | 1 | 0 | 0 |
| DIED GESTATION DAY 18 | 1 | 0 | 0 | 0 |
| ABORTED GESTATION DAY 17 | 0 | 0 | 0 | 1 |
| INTESTINE: CONTENTS, DARK RED | 1 | 0 | 0 | 0 |

1- 0 MG/KG/DAY 2- 1 MG/KG/DAY 3- 5 MG/KG/DAY 4- 50 MG/KG/DAY

## Cesarean Section Data (Implantation Sites, Pre- and Post-Implantation Loss, etc.)

There was a test article-related ↑ in the percentage of postimplantation loss in the 50 mg/kg/day group (16.2% per litter) compared to the concurrent control group (3.8% per litter) as a result of an increased incidence of early and late resorptions. A corresponding reduction was evident in the number of viable fetuses and the mean gravid uterine weight. Mean fetal weights in the 50 mg/kg/day group were similar to the concurrent control group values.

Although the LD and MD were not associated with s.s. effects on embryo/fetal survival, fetal weight or gravid uterine weight, they did show a dose-related trend consistent with what was seen at HD.

TABLE 10

PROJECT NO. (b) (4) 472097 GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE PAGE 1
SPONSOR:GLAXOSMITHKLINE SUMMARY OF GRAVID UTERINE WEIGHTS [G]
SPONSOR NO.:G06020

| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
|---|---|---|---|---|
| GRAVID UTERINE WT. | | | | |
| MEAN | 19.9 | 19.7 | 18.8 | 16.4+ |
| % CHANGE | | -1.0 | -5.5 | -17.6 |
| S.D. | 4.28 | 2.56 | 4.98 | 3.93 |
| S.E. | 0.96 | 0.59 | 1.11 | 0.95 |
| N | 20 | 19 | 20 | 17 |

MODIFIED STATISTICS USED. * INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
+ = Significantly different from the control group at 0.05

TABLE 11

PROJECT NO. (b) (4) 472097 GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SPONSOR:GLAXOSMITHKLINE SUMMARY OF FETAL DATA AT SCHEDULED NECROPSY
SPONSOR NO.:G06020

| GROUP | SEX M | SEX F | VIABLE FETUSES | DEAD FETUSES | RESORPTIONS EARLY | RESORPTIONS LATE | POST IMPLANTATION LOSS | IMPLANTATION SITES | CORPORA LUTEA | PRE IMPLANTATION LOSS | NO. OF GRAVID FEMALES |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 TOTAL | 130 | 96 | 226 | 0 | 7 | 2 | 9 | 235 | 242 | 7 | 20 |
| MEAN | 6.5 | 4.8 | 11.3 | 0.0 | 0.4 | 0.1 | 0.5 | 11.8 | 12.1 | 0.3 | |
| S.D. | 1.96 | 1.82 | 2.56 | 0.00 | 0.75 | 0.31 | 0.83 | 2.51 | 2.79 | 0.93 | |
| S.E. | 0.44 | 0.41 | 0.57 | 0.00 | 0.17 | 0.07 | 0.18 | 0.56 | 0.62 | 0.21 | |
| 2 TOTAL | 132 | 99 | 231 | 0 | 10 | 1 | 11 | 242 | 252 | 10 | 20 |
| MEAN | 6.6 | 5.0 | 11.6 | 0.0 | 0.5 | 0.1 | 0.6 | 12.1 | 12.6 | 0.5 | |
| S.D. | 1.70 | 1.39 | 1.85 | 0.00 | 0.61 | 0.22 | 0.69 | 1.55 | 1.76 | 0.89 | |
| S.E. | 0.38 | 0.31 | 0.41 | 0.00 | 0.14 | 0.05 | 0.15 | 0.35 | 0.39 | 0.20 | |
| 3 TOTAL | 112 | 107 | 219 | 0 | 10 | 0 | 10 | 229 | 240 | 11 | 20 |
| MEAN | 5.6 | 5.4 | 11.0 | 0.0 | 0.5 | 0.0 | 0.5 | 11.5 | 12.0 | 0.6 | |
| S.D. | 2.35 | 2.58 | 3.14 | 0.00 | 0.76 | 0.00 | 0.76 | 2.95 | 3.03 | 0.89 | |
| S.E. | 0.53 | 0.58 | 0.70 | 0.00 | 0.17 | 0.00 | 0.17 | 0.66 | 0.68 | 0.20 | |
| 4 TOTAL | 85 | 73 | 158 | 1 | 20 | 8 | 29 | 187 | 200 | 13 | 17 |
| MEAN | 5.0 | 4.3 | 9.3 | 0.1 | 1.2 | 0.5 | 1.7 | 11.0 | 11.8 | 0.8 | |
| S.D. | 2.26 | 1.93 | 3.06 | 0.24 | 1.47 | 1.70 | 2.42 | 1.80 | 2.08 | 1.30 | |
| S.E. | 0.55 | 0.47 | 0.74 | 0.06 | 0.36 | 0.41 | 0.59 | 0.44 | 0.50 | 0.32 | |

MODIFIED STATISTICS USED FOR MEAN NUMBER OF VIABLE FETUSES, CORPORA LUTEA AND IMPLANTATION SITES.
* INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
None significantly different from control group

1- 0 MG/KG/DAY 2- 1 MG/KG/DAY 3- 5 MG/KG/DAY 4- 50 MG/KG/DAY

TABLE 12

PROJECT NO. (b) (4)472097
SPONSOR:GLAXOSMITHKLINE
SPONSOR NO.:G06020

GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SUMMARY OF FETAL DATA AT SCHEDULED NECROPSY [% PER LITTER]

PAG

| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
|---|---|---|---|---|
| CORPORA LUTEA | | | | |
| MEAN | 12.1 | 12.6 | 12.0 | 11.8 |
| S.D. | 2.79 | 1.76 | 3.03 | 2.08 |
| S.E. | 0.62 | 0.39 | 0.68 | 0.50 |
| N | 20 | 20 | 20 | 17 |
| IMPLANTATION SITES | | | | |
| MEAN | 11.8 | 12.1 | 11.5 | 11.0 |
| S.D. | 2.51 | 1.55 | 2.95 | 1.80 |
| S.E. | 0.56 | 0.35 | 0.66 | 0.44 |
| N | 20 | 20 | 20 | 17 |
| VIABLE FETUSES (%) | | | | |
| MEAN | 96.2 | 95.2 | 94.8 | 83.8 |
| S.D. | 6.72 | 6.16 | 11.18 | 22.51 |
| S.E. | 1.50 | 1.38 | 2.50 | 5.46 |
| N | 20 | 20 | 20 | 17 |
| DEAD FETUSES (%) | | | | |
| MEAN | 0.0 | 0.0 | 0.0 | 0.6 |
| S.D. | 0.00 | 0.00 | 0.00 | 2.43 |
| S.E. | 0.00 | 0.00 | 0.00 | 0.59 |
| N | 20 | 20 | 20 | 17 |
| EARLY RESORPTIONS (%) | | | | |
| MEAN | 3.1 | 4.3 | 5.2 | 11.3 |
| S.D. | 6.25 | 5.19 | 11.18 | 14.80 |
| S.E. | 1.40 | 1.16 | 2.50 | 3.59 |
| N | 20 | 20 | 20 | 17 |

MODIFIED STATISTICS USED. * INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
None significantly different from control group

TABLE 12

PROJECT NO. (b) (4)472097
SPONSOR:GLAXOSMITHKLINE
SPONSOR NO.:G06020

GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE
SUMMARY OF FETAL DATA AT SCHEDULED NECROPSY [% PER LITTER]

PAC

| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
|---|---|---|---|---|
| LATE RESORPTIONS (%) | | | | |
| MEAN | 0.8 | 0.5 | 0.0 | 4.3 |
| S.D. | 2.38 | 2.24 | 0.00 | 14.35 |
| S.E. | 0.53 | 0.50 | 0.00 | 3.48 |
| N | 20 | 20 | 20 | 17 |
| TOTAL RESORPTIONS (%) | | | | |
| MEAN | 3.8 | 4.8 | 5.2 | 15.6 |
| S.D. | 6.72 | 6.16 | 11.18 | 22.81 |
| S.E. | 1.50 | 1.38 | 2.50 | 5.53 |
| N | 20 | 20 | 20 | 17 |
| PRE-IMPLANTATION LOSS (%) | | | | |
| MEAN | 2.3 | 3.7 | 5.3 | 5.9 |
| S.D. | 5.72 | 6.22 | 9.01 | 9.83 |
| S.E. | 1.28 | 1.39 | 2.02 | 2.38 |
| N | 20 | 20 | 20 | 17 |
| POST-IMPLANTATION LOSS (%) | | | | |
| MEAN | 3.8 | 4.8 | 5.2 | 16.2 |
| S.D. | 6.72 | 6.16 | 11.18 | 22.51 |
| S.E. | 1.50 | 1.38 | 2.50 | 5.46 |
| N | 20 | 20 | 20 | 17 |
| MALES (%) | | | | |
| MEAN | 58.3 | 57.1 | 51.9 | 52.3 |
| S.D. | 15.87 | 10.85 | 22.44 | 19.98 |
| S.E. | 3.55 | 2.43 | 5.02 | 4.85 |
| N | 20 | 20 | 20 | 17 |

MODIFIED STATISTICS USED. * INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
None significantly different from control group

TABLE 12

PROJECT NO. (b) (4) 472097 GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE PA(
SPONSOR:GLAXOSMITHKLINE SUMMARY OF FETAL DATA AT SCHEDULED NECROPSY [% PER LITTER]
SPONSOR NO.:G06020

| GROUP: | 0 MG/KG/DAY | 1 MG/KG/DAY | 5 MG/KG/DAY | 50 MG/KG/DAY |
|---|---|---|---|---|
| FEMALES (%) | | | | |
| MEAN | 41.7 | 42.9 | 48.1 | 47.7 |
| S.D. | 15.87 | 10.85 | 22.44 | 19.98 |
| S.E. | 3.55 | 2.43 | 5.02 | 4.85 |
| N | 20 | 20 | 20 | 17 |

MODIFIED STATISTICS USED. * INDICATES PARAMETRIC ANALYSIS AND + INDICATES NON-PARAMETRIC ANALYSIS.
None significantly different from control group

**Offspring (Malformations, Variations, etc.)**

The numbers of fetuses (litters) available for morphological evaluation were 226(20), 231(20), 219(20) and 158(17) in the control, 1, 5 and 50 mg/kg/day groups, respectively.

There were no treatment-related malformations at any dose.

A test article-related, statistically significant increase in the mean litter percentage of bent ribs (also known in the literature as wavy ribs or undulated ribs) was evident in the 50 mg/kg/day group (8.0% per litter, 11 affected fetuses in 6 affected litters) relative to the concurrent control group (0.0% per litter). Two or more bent ribs were detected in 7 fetuses (5 bilaterally and 2 right-sided), while 4 fetuses had only 1 bent rib. This variation has not been detected in the 25 studies comprising the (b) (4) historical control database.

There were no test article-related external or visceral fetal developmental variations noted at any dose level, and there were no test article-related skeletal developmental variations at 1 or 5 mg/kg/day.

Sponsor has postulated that the finding of ben ribs may be caused by low glucose levels as a consequence of exaggerated pharmacology. They note that in study # CD2006/01620/00 50 mg/kg albiglutide caused a 29% ↓ in glucose levels within 1 h after dosing in nonpregnant females. They believe this is relevant, because Tanigawa *et al.*[2] found that insulin-induced hypoglycemia was associated with bent ribs. The also site literature suggesting that bent ribs are reversible.[3]

[2] Tanagawa, K., *et al.* (1991), "Skeletal malformations in rat offspring. Long-term effect of maternal insulin-induced hypoglycaemia during organogenesis." *Diabetes* **40**:1115-1121.

[3] Nishimura, M., *et al.* (1982), "Repairability of drug-induced "wavy ribs" in rat offspring." *Arzneimittelforschung* **32**: 1518-1522.

TABLE 16

PROJECT NO. (b) (4) -472097 GSK716155: SUBCUTANEOUS EMBRYO/FETAL DEVELOPMENT STUDY IN MICE PA

SPONSOR:GLAXOSMITHKLINE SUMMARY OF FETUSES AND LITTERS WITH VARIATIONS [ABSOLUTE NO.]

SPONSOR NO.:G06020 DA

| | FETUSES | | | | LITTERS | | | |
|---|---|---|---|---|---|---|---|---|
| DOSE GROUP: | 1 | 2 | 3 | 4 | 1 | 2 | 3 | 4 |
| NUMBER EXAMINED EXTERNALLY | 226 | 231 | 219 | 158 | 20 | 20 | 20 | 17 |
| NUMBER WITH FINDINGS | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| NUMBER EXAMINED VISCERALLY | 226 | 231 | 219 | 158 | 20 | 20 | 20 | 17 |
| NUMBER WITH FINDINGS | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| NUMBER EXAMINED SKELETALLY | 226 | 231 | 219 | 158 | 20 | 20 | 20 | 17 |
| 14TH RUDIMENTARY RIB(S) | 58 | 65 | 50 | 43 | 18 | 19 | 19 | 15 |
| STERNEBRA(E) MALALIGNED(SLIGHT OR MODERATE) | 11 | 5 | 7 | 8 | 8 | 4 | 7 | 5 |
| 7TH CERVICAL RIB(S) | 46 | 54 | 36 | 36 | 19 | 17 | 13 | 11 |
| 14TH FULL RIB(S) | 69 | 53 | 58 | 30 | 17 | 17 | 17 | 10 |
| 7TH STERNEBRA | 12 | 10 | 17 | 4 | 7 | 5 | 9 | 2 |
| ACCESSORY SKULL BONE(S) | 25 | 41 | 24 | 21 | 13 | 16 | 12 | 12 |
| BENT RIB(S) | 0 | 0 | 0 | 11 | 0 | 0 | 0 | 6 |
| VERTEBRAL CENTRA UNOSSIFIED | 2 | 0 | 0 | 1 | 1 | 0 | 0 | 1 |
| 25 PRESACRAL VERTEBRAE | 0 | 3 | 1 | 1 | 0 | 1 | 1 | 1 |
| EXTRA SITE OF OSSIFICATION DORSAL TO CERVICAL CENTRUM #2 | 0 | 0 | 1 | 2 | 0 | 0 | 1 | 2 |
| 27 PRESACRAL VERTEBRAE | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 |
| CIRCULAR AREA OF UNOSSIFICATION IN STERNEBRA #6 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 0 |
| REDUCED OSSIFICATION OF THE SKULL | 1 | 0 | 0 | 0 | 1 | 0 | 0 | 0 |

1- 0 MG/KG/DAY 2- 1 MG/KG/DAY 3- 5 MG/KG/DAY 4- 50 MG/KG/DAY

## 9.3 Prenatal and Postnatal Development

### Study title: GSK716155A: Subcutaneous Postnatal Development Study in Mice

| | |
|---|---|
| Study no.: | 2011N117859 (b) (4) 20013661) |
| Study report location: | eCTD 4.2.3.5.3 |
| Conducting laboratory and location: | (b) (4) (b) (4) |
| Date of study initiation: | 31-MAY-2011 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # GAJB05DA, ≥ 96.7% pure |

### Key Study Findings

- Maternal toxicity (including ↓ BW gain during gestation and lactation, and abdominal distention, dehydration and pale ears during lactation) was seen at 50 mg/kg/day (HD).
- The HD F1 generation (♂&♀) had ↓ BW at birth and throughout the preweaning period. HD♂s were slightly s.s. delayed for preputial separation, possibly related to the persistent low BW seen in F1 HD♂s.
- There no effects on F1 reproductive function or F2 morphology and viability.
- NOAEL for maternal toxicity, F0 reproductive function and pre-/post-natal development is considered to be 5 mg/kg/day.

Methods

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | Soln. in 10 mM sodium phosphate, 2.8% mannitol, 4.4% trehalose, 0.01% polysorbate 80 (pH 7.2) |
| Species/Strain: | Pregnant CD-1 mice |
| Number/Sex/Group: | 24 |
| Satellite groups: | 4-5 for TK & Immunogenicity |
| Study design: | Main study: Pregnant mice were dosed from post coital (pc) day 6 to 17 (inclusive). F0 females were allowed to deliver naturally. Mated (F0) females were euthanized on Day 21 postpartum (pp). Litters were culled to 4/sex/litter on Day 4 pp. On Day 21 pp, F1 males and F1 females (2/sex/litter, when possible) were assigned to each dose group and assigned to one of two subsets. Subset 1 was selected for acoustic startle habituation (PND 60 ± 2) and reproductive performance. Mated F1 females assigned to Subset 1 were allowed to deliver naturally and the dams and F2 litters were evaluated until Day 7 pp. Subset 2 was selected for acoustic startle habituation (PND 29 ± 2), motor activity (PND 60 ± 2), and Morris Water maze evaluations (PND 78 ± 12).<br>TK dams: Plasma drug levels were measured in the dams and fetuses on Day 17 pc (6 h after last dose). Plasma from the dams was also evaluated for anti-drug antibodies (ADA). Baseline ADA samples were also collected prior to dosing on Day 6 pc (1st day of dosing). |
| Deviation from study protocol: | None material to interpretation of the study. |

**Observations and Results (Optional Table)**

$F_0$ Dams

| | |
|---|---|
| Survival: | There were no test article-related deaths, however one HD dam had a total litter loss and was euthanized. |
| Clinical signs: | None prior to deliver.<br>HD only: abdominal distension (for 1 to 5 days, not seen after Day 5 pp), mild dehydration (for 1 to 6 days, not seen after Day 9 pp), pale ears and/or thin body condition. |
| Body weight: | ↓ BW gain (loss after 1st dose) in HD. *See below.* |
| Feed consumption: | HD only: slight ↓ during pc period, but no effect during pp period. |
| Uterine content: | No effect of test item. |

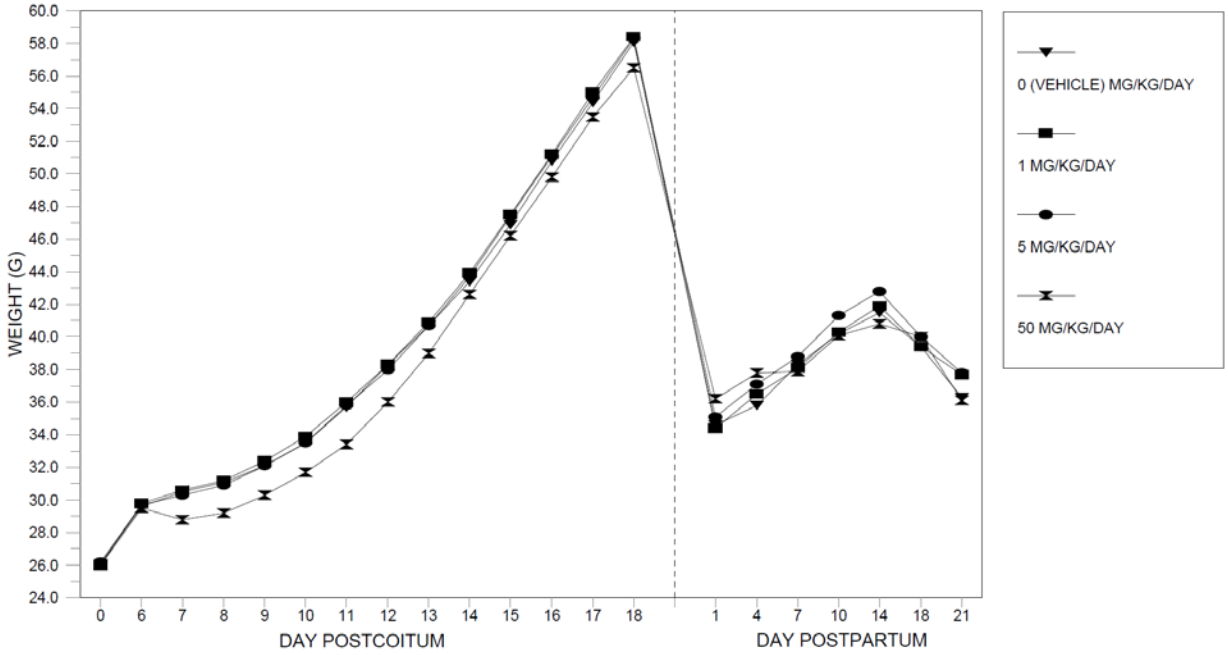


Reference ID: 3347159

Necropsy observation: No effect of test item.
Toxicokinetics: Plasma level ranges (µg/mL): C = BLoQ, LD = BLoQ to 0.0864, MD = 0.0572 to 4.45, HD = 31.6 to 110.
ADA: C = 0/4, LD = 2/4 (titers: 1:95, 1:460), MD = 0/4, HD = 2/4 (titers: 1:22, 1:75)
Dosing Solution Analysis: All results were within the acceptable range of ±10% of nominal concentration.
Other: No effect on gestation period or parturition.

MATERNAL BODY WEIGHTS - F0 GENERATION FEMALES

Figure 1




$F_1$ Generation

Survival: No effect. Although the mean number of liveborn pups per litter exhibited a n.s.s. d.d. ↓ (13.3, 12.9, 12.7 & 12.6 in C, LD, MD & HD, respectively), all mean values were higher than the Testing Facility Historical Control Data (range of 11.2 to 12.4 pups/litter).
Clinical signs: *Preweaning*: 3 HD and 1 MD litter had pups that experienced one or more days of dehydration. 2HD and 1 MD litter had pups that were cold to touch. 2 HD litters had pups that had a thin body condition (see Sponsor table A11, below). The findings for the MD are from a single pup on a single day and are not necessarily considered to be due to test item exposure of the dams.

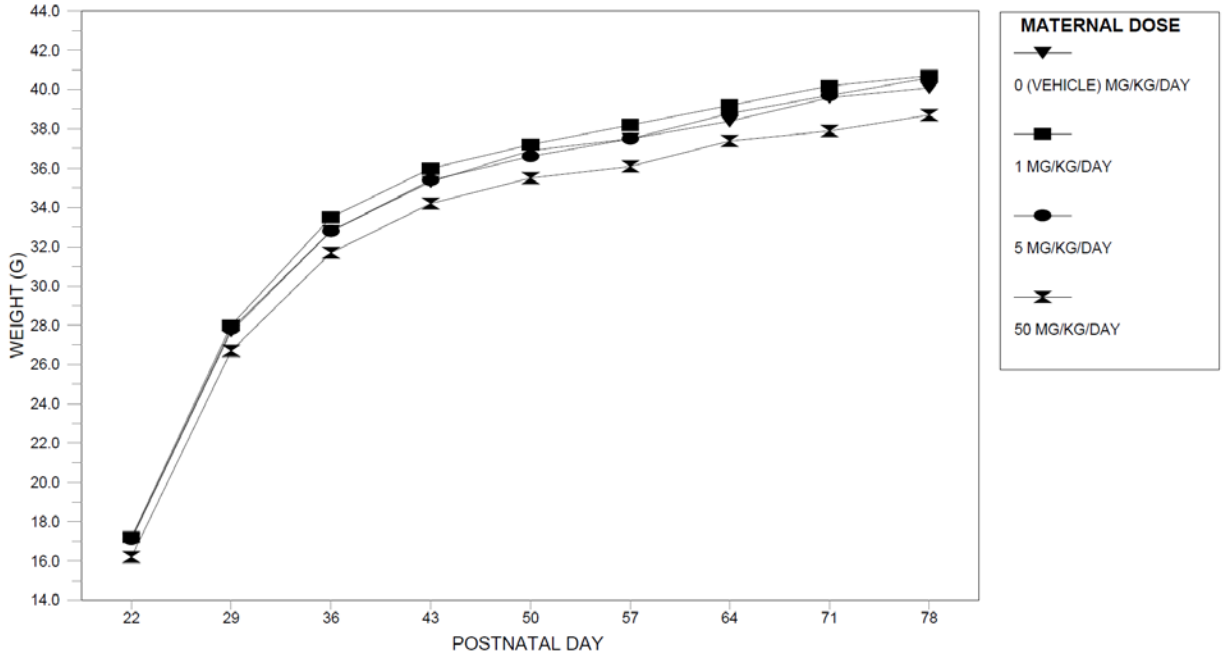
| | |
|---|---|
| | *Postweaning:* No test item-related effect. |
| Toxicokinetics: | Plasma level ranges (µg/mL): C = BLoQ, LD = BLoQ,, MD = BLoQ to 0.0221, HD = 0.0504 to 0.104. |
| Body weight: | BW was s.s. ↓ in ♂ & ♀ (combined) during the preweaning, and were ~95% of C on PND 21. While not s.s., ♂s show a clear trend towards a persistent BW deficit throughout the study period (Sponsor's figure 2, below). |
| Feed consumption: | No data. |
| Physical development: | ♂: s.s. delay in preputial separation at HD (PND 31.3 vs. PND 29.6 in C). |
| | ♀: no effect. |
| Neurological assessment: | *Motor activity:* No effect. |
| | *Acoustic startle:* No effect. |
| | *Morris water maze:* No effect. |
| Reproduction: | No effect on mating behavior, fertility, mating index, average numbers of days in cohabitation, estrus cycling, mean number of implantation sites, duration of gestation, parturition, viability index or lactation index. |

TABLE A11 (PAGE 1): CLINICAL OBSERVATIONS FROM BIRTH TO DAY 21 POSTPARTUM - SUMMARY - F1 GENERATION PUPS

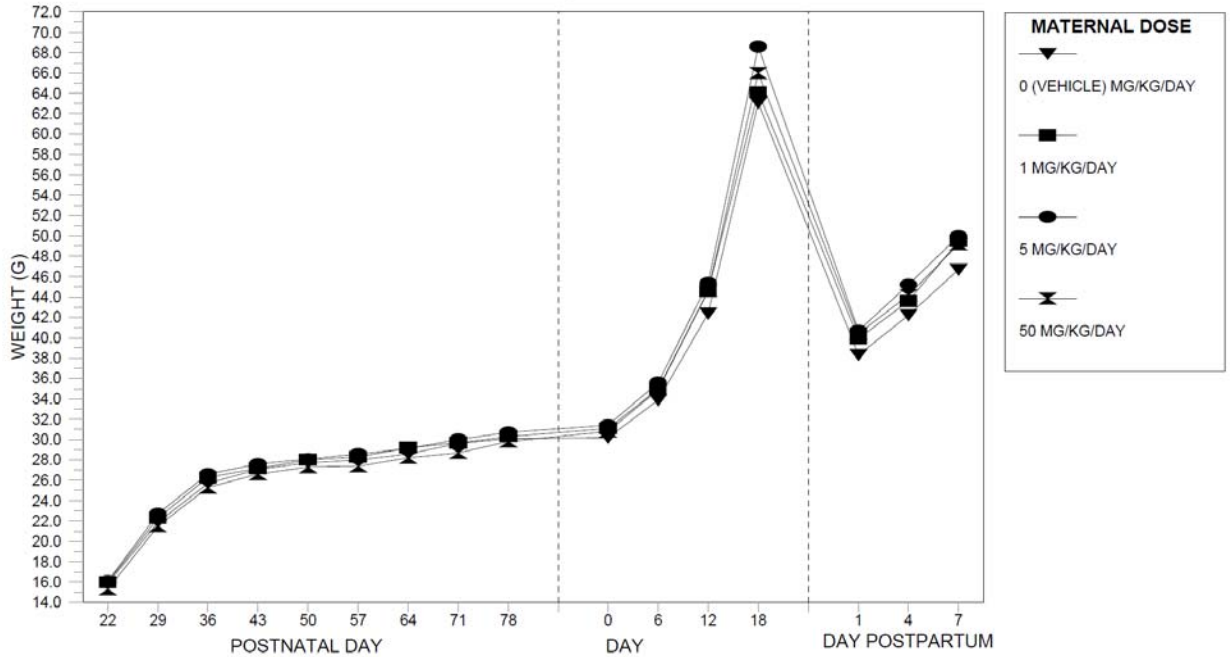
| MATERNAL DOSE GROUP | | | 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|---|
| MATERNAL DOSE (MG/KG/DAY) | | | 0 (VEHICLE) | 1 | 5 | 50 |
| LITTERS EXAMINED (N) | | | 23 | 24 | 21 | 22 |
| TOTAL FREQUENCY (DAYS X PUPS)/LITTERS WITH OBSERVATIONS a | | | | | | |
| DEHYDRATION: | TOTAL | N/N | 0/0 | 0/0 | 1/1 | 48/3 |
| | MILD | N/N | 0/0 | 0/0 | 0/0 | 19/2 |
| | MODERATE | N/N | 0/0 | 0/0 | 0/0 | 28/1 |
| | SEVERE | N/N | 0/0 | 0/0 | 1/1 | 0/0 |
| | NO SEVERITY NOTED | N/N | 0/0 | 0/0 | 0/0 | 1/1 |
| COLD TO TOUCH | | N/N | 0/0 | 0/0 | 1/1 | 32/2 |
| THIN BODY CONDITION | | N/N | 0/0 | 0/0 | 0/0 | 31/2 |
| DECREASED MOTOR ACTIVITY | | N/N | 0/0 | 0/0 | 1/1 | 0/0 |
| ABDOMINAL DISTENTION | | N/N | 0/0 | 0/0 | 1/1 | 0/0 |
| DOMED HEAD | | N/N | 0/0 | 16/1 | 0/0 | 0/0 |
| HEAD, SCAB | | N/N | 0/0 | 1/1 | 0/0 | 0/0 |
| HEAD, SWOLLEN | | N/N | 0/0 | 1/1 | 0/0 | 0/0 |

a. Tabulation restricted to adverse observations; all other pups appeared normal.

BODY WEIGHTS - F1 GENERATION MALES
Figure 2




BODY WEIGHTS - F1 GENERATION FEMALES
Figure 3




$F_2$ Generation

| | |
|---|---|
| Survival: | No effect. |
| Clinical signs | No effect. |
| Body weight: | No effect. |
| External evaluation: | No effect. |
| Male/Female ratio: | No effect. |

**Study title: GSK716155: Subcutaneous Pre- and Postnatal Development Study, Including Maternal Function, in Mice**

| | |
|---|---|
| Study no.: | (b) (4)-472211 & (b) (4)-472260 |
| Study report location: | eCTD 4.2.3.5.3 |
| Conducting laboratory: | (b) (4) |
| Date of study initiation: | 5 February 2008 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, batch # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- Unexpectedly high frequency of unscheduled maternal deaths ($F_0$) primarily during the lactation period, which were not strictly dose-related (peaked at MD and fell off at HD) caused Sponsor to conduct a follow-up study to determine if the increased mortality was reproducible. The increased maternal deaths were not reproducible. Nonetheless, based on the findings of increased maternal deaths during lactation when dams were dosed from Days 7 to 21 PP (study # 2010N104536, below), this finding is considered to be drug-related.
- $F_0$ dams: ↓ food consumption from GD 15 through LD 10.
- $F_1$ pups: born with dose-related (all doses) ↓ in bodyweight. Pups also had a dose-related weight gain decrement throughout lactation. GSK716155-treated pups had increased weight gain post-weaning, and all but MD & HD ♀s had normalized bodyweight (compared to C) by PND 84.
- ♂ $F_1$ pups: dose-related, n.s.s. trend towards minimally delayed balanopreputial separation: PND 29.1, 30.7, 30.9 and 31.0 (C, LD, MD, HD). May be related to the dose-related ↓ in bodyweight noted above. All values were within the historical control range.
- $F_1$ pups: n.s.s. ↓ force of the response (maximum and average) to the startle reflex at all dose levels in ♂s and at MD & HD in ♀s on PND 20. At PND 60 HD♂s still had a blunted response, but all other groups had responses comparable to C. The time to maximum response was not affected at either timepoint in any dose group. Sponsor dismisses this finding, presumably on the basis that the changes were small and not s.s. Given that the startle response is measured as the force generated by the animal in response to the startle stimulus, and the magnitude of the force that can be generated is proportional to the mass of the animal, these relatively small effects may be secondary to the ↓ bodyweights noted above; however a direct effect on the sensory motor neural pathway controlling the reflex cannot be entirely ruled out.
- $F_1$ & $F_2$ pups: there appeared to be an increase in the incidence subcutaneous hemorrhages.
- The NOAEL is considered to be < 1.0 mkd on the basis of the reduced $F_1$ bodyweight.



Reference ID: 3347159

**Methods**

Doses:

(b) (4) 472211: 0, 1, 5, 50 mg/kg/day
472260: 0, 5 mg/kg/day

Species/strain: Mice/Crl:CD-1 (ICR)

Number/sex/group:

(b) (4) 472211: 24/g
472260: 35/g

Route, formulation and volume:

SC injection

Soln. in 10 mM sodium phosphate, 2.8% mannitol, 4.2% trehalose, 0.01% polysorbate 80 (pH 7.2)

2 mL/kg

Satellite groups used for toxicokinetics: 8 mated ♀s/g. *The following is excerpted from the study report.*

**3.4.5. Plasma Concentration Evaluation (Toxicokinetic Phase Only)**

Plasma concentrations of GSK716155 were determined in F0 dams, F1 pups, and naïve F0 females[2]. Blood samples from approximately 9 F1 pups/sex/time point/group were collected at nominal times of 3, 8, and 22 hours following dose administration of the F0 females on PND 10. Blood samples from 3 same-sex pups (of generally different litters) were pooled to create a single sample. The arbitrarily selected pups (F1) were euthanized and had blood collected by decapitation; pups were discarded without examination. Surviving pups not used for blood collection were euthanized by carbon dioxide inhalation on PND 11 and discarded. Blood samples were also collected from all F0 dams via the retrorbital sinus on lactation Day 10 or 11 (following the last blood collection of the F1 litter); the time of blood collection was recorded. Following blood collection, F0 dams were euthanized by carbon dioxide inhalation and discarded without examination. Furthermore, blood samples from 10 naïve F0 females were collected from the inferior vena cava following euthanasia of these females by carbon dioxide inhalation; naïve females were discarded without examination following blood collection. Target blood volumes for a single sample were 0.25 to 0.5 mL. Samples were collected into labelled tubes containing $K_2EDTA$ as the anticoagulant and 5 µL of a 0.2 mg/mL solution of aprotinin (protease inhibitor). Samples were kept on wet ice until centrifugation.

Study design:

(b) (4)-472211: $F_0$ ♀s dosed once daily from gestation day (GD) 15 to lactation day (LD) 10. $F_0$ ♀s were allowed to deliver naturally. $F_1$ offspring (3/sex/litter after culling) were reared and assigned to testing subsets. Beginning on postnatal day (PND) 88 to 93, The $F_1$ ♂s &♀s were cohabited in a ratio of 1:1 for up to 14 days. Mated $F_1$ ♀s were allowed to deliver naturally, and they and their $F_2$ litters were evaluated until LD 7.

(b) (4)-472260: Investigative study to ascertain reproducibility of maternal mortality observed in (b) (4)-472211 during the lactation phase. $F_0$ ♀s dosed once daily from GD 15 to LD 10. $F_0$ ♀s were allowed to deliver naturally. Mated $F_0$ ♀s and their offspring were sac'd on LD 10.

Parameters and endpoints evaluated:

(b) (4)-472211: $F_0$ females: survival, clinical observations, body weight, food

consumption, parturition, plasma concentration of GSK716155, and necropsy; $F_1$ offspring: survival, gender, external morphology, clinical observations, body weight, plasma concentration of GSK716155, necropsy, physical development (including vaginal opening in females and balanopreputial skin fold separation in males), neurobehavioral function (startle reflex, learning and memory, motor activity), and reproductive function; $F_2$ litters: survival, gender, external morphology, clinical observations, and body weight.

(b) (4)-472260: $F_0$ females: survival, clinical observations, body weight, food and water consumption, parturition, serum glucose levels, and necropsy; $F_1$ offspring: survival, gender, clinical observations and body weight.

**Results**

Unscheduled deaths:

*excerpted from Sponsor's study report*

In the pre- and postnatal study, maternal (F0) mortality and/or moribundity were observed in the main and toxicokinetic phases of the study (0, 2, 5, and 2 deaths in the control, 1, 5, and 50 mg/kg/day groups, respectively). All deaths occurred during the period of lactation. A specific cause of death could not be identified. Clinical signs noted for moribund mice included hypoactivity, hunched posture, lethargy (eyelids completely shut or slightly drooping), decreased defecation, body cool, and/or yellow material in the anogenital area, while the only observation in mice that were found dead was scabbing on the ears. In F1 mice, a high incidence of non-drug-related deaths occurred post-weaning in all groups, including the control group (9, 6, 15, and 9 deaths in the control, 1, 5, and 50 mg/kg/day groups, respectively). In the subsequent investigational study, there were no unscheduled deaths in the F0 or F1 mice prior to scheduled euthanasia on lactation Day 6 or 10. Taken together, the maternal deaths in the pre- and postnatal study were considered spontaneous and not test article-related for the following reasons: 1) there was no evident dose-response relationship; 2) there was a high incidence of incidental deaths in F1 mice; and 3) the maternal (F0) deaths were not reproducible in the investigative study. These unexplained deaths were judged not to have compromised the study because the numbers of surviving animals in each group were sufficient to meet all study objectives.

$F_0$ in-life:

(b) (4)-472211:

Survival: see "Unscheduled deaths," above.

Clinical signs: No test item-related effect.

Pregnancy status: No test item-related effect.

Gestation length: No test item-related effect.

Bodyweight: No test item-related effect on bodyweight during the gestation or lactation periods.

Food consumption: No test item-related effect at any dose between GD 1 & GD 15; however HD♀s had s.s. ↓ FC between GD15 & GD18 (0.8x C) and throughout the lactation period (0.85x of C).

Littering data: No test item-related effect.



Reference ID: 3347159

(b) (4)-472260:

Survival: see "Unscheduled deaths," above.

Clinical signs: No test item-related effect.

Pregnancy status: No test item-related effect.

Gestation length: No test item-related effect.

Bodyweight: No test item-related effect on bodyweight during the gestation or lactation periods.

Food consumption: No test item-related effect at any dose between GD 1 & GD 15; however treated dams had s.s. ↓ FC between GD15 & GD18 (0.92x C) and throughout the lactation period (0.94x of C).

Water consumption: No test item-related effect.

Serum glucose (LD 10): s.s. 9.5% ↓.

Littering data: No test item-related effect.

$F_0$ necropsy:

(b) (4)-472211: Unremarkable.

(b) (4)-472260: Unremarkable.

$F_1$ physical development:

(b) (4)-472211:

Sex ratio: No test item-related effect.

Survival: No test item-related effect.

Clinical Signs: Possible ↑ in the incidence of small stature; however there was no clear dose relationship: 7(5), 9(7), 14(12), 8(6) [incidence (number of pups) for C, LD, MD & HD, respectively].

Pre-weaning bodyweight: Dose-related ↓ in bodyweight, frequently s.s. in all dose groups (♂&♀) out to PND 07, still s.s. at PND 14 at HD. Differences still persist at PND 21, but no longer s.s.

| $F_1$ Bodyweight, Pre-weaning: % Difference from Control | | | | | | |
|---|---|---|---|---|---|---|
| Sex | Male | | | Female | | |
| $F_0$ Dose (mg/kg/day) | 1 | 5 | 50 | 1 | 5 | 50 |
| PND 0 | -5.5 | **-7.9** | **-10.4** | **-8.7** | **-9.9** | **-11.8** |
| PND 1 | -6.5 | **-9.2** | **-9.2** | **-9.9** | **-11.0** | **-11.5** |
| PND 4 | **-11.2** | **-11.9** | **-14.6** | **-14.4** | **-13.7** | **-16.8** |
| PND 7 | **-14.3** | **-11.3** | **-18.1** | **-17.5** | **-13.0** | **-20.2** |
| PND 14 | -8.2 | -8.0 | -13.9 | -12.5 | -11.4 | **-16.7** |
| PND 21 | -10.3 | -12.0 | -16.0 | -14.7 | -12.5 | -15.8 |

**Bold** indicates s.s. ($p<0.05$) Δ from C

Post-weaning bodyweight: Dose-related reductions in bodyweight persisted into the post-weaning period; however, the magnitude of the difference from C ↓ with time, and eventually normalized for all but HD♀.

| $F_1$ Bodyweight, Post-weaning: % Difference from Control | | | | | | |
|---|---|---|---|---|---|---|
| Sex | Male | | | Female | | |
| $F_0$ Dose (mg/kg/day) | 1 | 5 | 50 | 1 | 5 | 50 |
| PND 28 | -9.9 | -12.4 | -14.9 | -13.9 | -12.5 | -15.3 |
| PND 35 | -8.5 | -8.5 | -12.7 | -9.3 | -9.3 | -10.3 |
| PND 42 | -4.5 | -3.0 | -7.8 | -4.6 | -5.5 | -6.0 |
| PND 49 | -4.4 | – | -5.1 | -4.3 | -5.1 | -8.1 |
| PND 56 | -3.6 | – | -3.6 | – | -4.1 | -7.4 |
| PND 63 | -4.1 | – | -4.4 | -3.6 | -5.2 | -8.0 |
| PND 70 | -3.4 | – | -4.3 | – | -4.9 | -6.5 |
| PND 77 | – | – | – | – | – | -8.6 |
| PND 84 | – | – | – | – | -3.0 | -7.0 |

"–" denotes no change (less than 3%) from C

Balanopreputial separation: Dose-related, n.s.s. trend towards increase: PND 29.1, 30.7, 30.9 and 31.0 (C, LD, MD, HD), which is probably related to dose-related Δs in bodyweight noted above. All values within the historical control range.

Vaginal patency: No clear test item-related effect.

(b) (4)-472260:

Sex ratio: No test item-related effect.

Survival: No test item-related effect.

Clinical Signs:

| Clinical Signs (♂ & ♀ together): number of incidences/pups affected | | |
|---|---|---|
| $F_0$ Dose (mg/kg/day) | 0 | 5 |
| N | 408 | 456 |
| Small Stature | 12/9 | 44/24 |
| Subcutaneous Hemorrhage(s) | 3/3 | 11/9 |

Bodyweight: There was a progressive weight gain decrement in the test item treated animals, seen in both sexes. This is similar to what was observed in (b) (4)-472211, and, as in that study, would presumably have been reversible following weaning.

| $F_1$ Bodyweight, Pre-weaning: % Difference from Control | | |
|---|---|---|
| | Males | Females |
| PND 0 | -2.6 | -1.3 |
| PND 1 | -3.0 | 0.0 |
| PND 4 | -5.6 | -1.7 |
| PND 7 | **-7.1** | -2.8 |
| PND 10 | **-10.5** | -6.1 |

**Bold** indicates s.s. Δ from C.

<u>$F_1$ behavioral evaluation</u>:

(b) (4)-472211:

Startle reflex: Both ♂ & ♀ exhibited small, n.s.s. ↓s in maximum (MAX) and average (AVE) magnitude of the startle response with no change in the time to MAX response (Tmax) on PND 20. By PND 60 only HD♂s continued to have a slight ↓ in

these parameters. Given that the startle response is measured as the force generated by the animal in response to the startle stimulus, and the magnitude of the force that can be generated is proportional to the mass of the animal, these relatively small effects may be secondary to the ↓ bodyweights noted above; however a direct effect on the sensory motor neural pathway controlling the reflex cannot be entirely ruled out.

| Startle Reflex Parameters : % Difference from Control | | | | | | |
|---|---|---|---|---|---|---|
| Sex | Male | | | Female | | |
| $F_0$ Dose (mg/kg/day) | 1 | 5 | 50 | 1 | 5 | 50 |
| PND 20 | | | | | | |
| MAX (newtons) | -12.8 | -11.8 | -16.8 | – | -12.0 | -12.0 |
| AVE (newtons) | -12.6 | -11.8 | -15.4 | – | -9.9 | -14.6 |
| Tmax (msec) | – | – | – | – | – | – |
| PND 60 | | | | | | |
| MAX (newtons) | +41.2 | – | -19.8 | – | – | – |
| AVE (newtons) | +51.3 | – | -23.0 | – | – | – |
| Tmax (msec) | – | – | – | – | – | – |

"–" denotes no change from C

Motor activity: No test item-related effect.

Learning & Memory (Biel water maze): No test item-related effect.

<u>$F_1$ reproduction</u>:

(b) (4)-472211:

Gestation Bodyweight: No test item-related effect on weight <u>gain</u> during gestation; however HD dams started gestation at an ~7% bodyweight deficit compared to C dams. This difference persisted throughout gestation and into the lactation period.

Mating & Fertility: No test item-related effect.

Estrous Cycle: n.s.s. 15% ↑ in estrous cycle length, corresponding to a s.s. 25% ↓ in the number of estrous cycles at HD.

Pregnancy status: No test item-related effect.

Gestation length: No test item-related effect.

Implantation sites: n.s.s., dose-related ↓ in the # of implantation sites (14.2, 13.6, 12.4 & 11.7 at C, LD, MD & HD, respectively); however all values were within the historical control range.

<u>$F_1$ necropsy (pups euthanized due to death of dam)</u>:

(b) (4)-472211: 3/23 HD pups (from 1 litter) had a finding of opacity of the eyes. No eye abnormalities were reported in the seg 2 study in mice ((b) (4)-472097). Relationship to treatment is unclear.

<u>$F_2$ findings</u>:

(b) (4)-472211 (only):

Live Litter Size: Dose-related ↓ (13.4, 12.7, 11.9 & 10.3 at C, LD, MD & HD, respectively). HD s.s. different from C and outside of historical control range. This appears to be related to the reduced # of implantation sites observed in the $F_1$ dams (above). Notably, in the seg 2 mouse study (b) (4) -472097), 50 mkd was associated with n.s.s. ↑ in pre- and post-implantation loses.

Sex Ratio (at birth): No test item-related effect.

Survival: No test item-related effect.

Clinical Signs:

| Clinical Signs (♂ & ♀ together): number of incidences/pups affected | | | | |
|---|---|---|---|---|
| $F_0$ Dose (mg/kg/day) | 0 | 1 | 5 | 50 |
| Small stature | 4/3 | 7/3 | 8/7 | 2/1 |
| Subcutaneous hemorrhage(s) | 0/0 | 2/2 | 0/0 | 4/4 |

Bodyweight: No test item-related effect.

Necropsy: No test item-related effect.

Toxicokinetics: *Plasma concentrations of GSK716155 were measured in pups on PND 10 at 3, 8 or 22 h after GSK716155 administration to the dams. Generally 9 pups were used for each timepoint (3 replicates of 3 pooled samples for each timepoint). On the same day (PND 10), plasma concentrations of GSK716155 were measured in the dams at 8 or 22 h after dosing.*

GSK716155 was quantifiable in each of the pooled plasma samples from the HD pups, and was present at between 13 to 49 ng/mL. In comparison, maternal plasma levels in the HD group varied between 55,200 to 174,000 ng/mL at the same timepoints, or ~1,100x to 13,000x the levels observed in the HD pups.

In MD pups, GSK716155 was above the LLOQ in 8 of 17 pooled samples. The mean of all of these positive samples was ~7 ng/mL (the LLOQ = 5). Only 3 of 5 MD dams had quantifiable levels of drug.

No LD dams had quantifiable levels of GSK716155, while 1 LD pup pooled sample did (7.5 ng/mL).

GSK716155 was not detected in C dams or pups.

**Table 1 Plasma Concentrations of GSK716155 in Male and Female (Pup) Mice on PND 10 at each Time Point Following Subcutaneous Maternal Administration of GSK716155 at a Nominal Dose of 0 mg/kg/day**

| Male Pups | | | |
|---|---|---|---|
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7158-01+7229-01+7193-02 | NQ | NQ |
| | 7158-02+7219-01+7262-01 | NQ | |
| | 7158-03+7229-03+7262-02 | NQ | |
| 8 | 7158-04+7229-04+7913-05 | NQ | NQ |
| | 7158-05+7219-04+7262-04 | NQ | |
| | 7158-07+7229-05+7219-07 | NQ | |
| 22 | 7158-06+7219-05+7262-05 | NQ | NQ |
| | 7229-02+7193-03+7219-02 | NQ | |
| | 7193-04+7219-03+7262-03 | NQ | |
| Female Pups | | | |
| Nominal Time [b] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7158-09+7229-11+7193-06 | NQ | NQ |
| | 7158-10+7219-11+7262-06 | NQ | |
| | 7193-13+7229-13+7262-07 | NQ | |
| 8 | 7158-12+7229-14+7193-09 | NQ | NQ |
| | 7158-13+7219-14+7262-09 | NQ | |
| | 7193-10+7219-15+7262-10 | NQ | |
| 22 | 7158-14+7193-11+7193-12 | NQ | NQ |
| | 7229-12+7193-07+7219-12 | NQ | |
| | 7193-08+7219-13+7262-08 | NQ | |

NQ = Not quantifiable (< 5.00 ng/mL)
a. Time from maternal (dam) dose.
b. Animal numbers indicate each of three individual pups pooled per sample.

**Table 2 Plasma Concentrations of GSK716155 in Male and Female (Pup) Mice on PND 10 at each Time Point Following Subcutaneous Maternal Administration of GSK716155 at a Nominal Dose of 1 mg/kg/day**

| Male Pups | | | |
|---|---|---|---|
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7257-03+7221-02+7221-03 | NQ | NQ |
| | 7224-02+7202-02+7245-01 | NQ | |
| | 7202-03+7116-03+7245-03 | NQ | |
| 8 | 7224-01+7202-01+7116-01 | 7.50 | NQ |
| | 7224-05+7202-05+7245-04 | NQ [c] | |
| | 7224-06+7202-06+7245-05 | NQ [c] | |
| 22 | 7224-03+7116-02+7245-02 | NQ | NQ |
| | 7224-04+7202-04+7116-04 | NQ | |
| | 7257-01+7257-02+7221-01 | NQ | |
| **Female Pups** | | | |
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7224-09+7202-07+7116-05 | NQ | NQ |
| | 7224-10+7202-08+7245-06 | NQ | |
| | 7202-09+7116-07+7245-08 | NQ | |
| 8 | 7224-13+7202-11+7245-09 | NQ | NQ |
| | 7224-14+7116-09+7245-10 | NQ | |
| | 7224-15+7202-13+7245-12 | NQ | |
| 22 | 7224-11+7116-06+7245-07 | NQ | NQ |
| | 7224-12+7202-10+7116-08 | NQ | |
| | 7249-03+7249-04+7249-05 | NQ | |

NQ = Not quantifiable (< 5.00 ng/mL)
a. Time from maternal (dam) dose.
b. Animal numbers indicate each of three individual pups pooled per sample.
c. NQ was assigned as zero for calculating the mean concentration.

**Table 3 Plasma Concentrations of GSK716155 in Male and Female (Pup) Mice on PND 10 at each Time Point Following Subcutaneous Maternal Administration of GSK716155 at a Nominal Dose of 5 mg/kg/day**

| Male Pups | | | |
|---|---|---|---|
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7197-01+7264-01+7259-01 | NQ [c] | NQ |
| | 7197-02+7264-03+7259-03 | 7.31 | |
| | 7195-04+7195-05+7195-06 | NQ [c] | |
| 8 | 7197-05+7264-04+7259-04 | 5.94 | NQ |
| | 7195-01+7195-02+7195-03 | NQ [c] | |
| | 7211-02+7211-03+7211-04 | NQ [c] | |
| 22 | 7197-03+7264-02+7259-02 | 5.90 | NQ |
| | 7197-04+7259-05+7259-08 | NQ [c] | |
| **Female Pups** | | | |
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7211-05+7211-06+7211-07 | NQ [c] | NQ |
| | 7197-06+7264-05+7264-07 | NQ [c] | |
| | 7197-07+7264-10+7259-06 | 6.30 | |
| 8 | 7197-10+7264-12+7259-09 | 7.58 | 8.08 |
| | 7197-11+7264-09+7259-10 | 8.81 | |
| | 7197-12+7197-13+7264-11 | 7.84 | |
| 22 | 7197-08+7264-06+7259-07 | 6.32 | NQ |
| | 7197-09+7264-08+7259-11 | NQ [c] | |
| | 7195-07+7195-08+7195-09 | NQ [c] | |

NQ = Not quantifiable (< 5.00 ng/mL)
a. Time from maternal (dam) dose.
b. Animal numbers indicate each of three individual pups pooled per sample.
c. NQ was assigned as zero for calculating the mean concentration.

**Table 4 Plasma Concentrations of GSK716155 in Male and Female (Pup) Mice on PND 10 at each Time Point Following Subcutaneous Maternal Administration of GSK716155 at a Nominal Dose of 50 mg/kg/day**

| Male Pups | | | |
|---|---|---|---|
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7250-01+7246-06+7209-01 | 32.4 | 31.5 |
| | 7250-02+77238-01+7115-01 | 40.5 | |
| | 7209-06+7209-07+7238-05 | 21.6 | |
| 8 | 7250-04+7209-04+7238-04 | 26.4 | 31.7 |
| | 7250-05+7209-05+7115-04 | 21.5 | |
| | 7113-01+7113-02+7113-03 | 47.2 | |
| 22 | 7250-03+7246-09+7115-02 | 29.2 | 29.6 |
| | 7246-08+7209-02+7238-02 | 26.2 | |
| | 7209-03+7238-03+7115-03 | 33.3 | |
| **Female Pups** | | | |
| Nominal Time [a] (h) | Animal Numbers [b] | Concentration (ng/mL) | Mean (ng/mL) |
| 3 | 7250-06+7246-10+7209-11 | 18.8 | 29.2 |
| | 7250-07+7238-06+7115-05 | 45.8 | |
| | 7250-08+7209-13+7115-10 | 23.0 | |
| 8 | 7250-10+7209-15+7238-10 | 40.0 | 37.3 |
| | 7209-14+7238-09+7115-08 | 22.8 | |
| | 7113-09+7113-10+7113-11 | 49.0 | |
| 22 | 7250-11+7238-11+7115-11 | 18.3 | 23.1 |
| | 7246-12+7209-12+7238-07 | 37.8 | |
| | 7250-09+7238-08+7115-12 | 13.2 | |

NQ = Not quantifiable (< 5.00 ng/mL)
a. Time from maternal (dam) dose.
b. Animal numbers indicate each of three individual pups pooled per sample.

**Table 5 Plasma Concentrations of GSK716155 in Maternal (Dam) Mice on LD 10 Following Subcutaneous Administration of GSK716155 at Nominal Doses of 0, 1, 5 and 50 mg/kg/day**

| Plasma Concentration of GSK716155 (ng/mL) | | | | | |
|---|---|---|---|---|---|
| Animal Number | Time [a] (h) | Nominal Dose (mg/kg/day) | | | |
| | | 0 | 1 | 5 | 50 |
| 7158 | 22 | NQ | | | |
| 7229 | 22 | NQ | | | |
| 7193 | 22 | NQ | | | |
| 7219 | 22 | NQ | | | |
| 7262 | 22 | NQ | | | |
| 7217 | 22 | NQ | | | |
| 7237 | 22 | NQ | | | |
| 7257 | 22 | | NQ | | |
| 7221 | 22 | | NQ | | |
| 7224 | 22 | | NQ | | |
| 7202 | 22 | | NQ | | |
| 7116 | 22 | | NQ | | |
| 7245 | 22 | | NQ | | |
| 7249 | 22 | | NQ | | |
| 7211 | 8 | | | 219 | |
| 7197 | 22 | | | NQ | |
| 7264 | 22 | | | 27.0 | |
| 7259 | 22 | | | NQ | |
| 7195 | 22 | | | 67.8 | |
| 7113 | 8 | | | | 174000 |
| 7250 | 22 | | | | 93200 |
| 7246 | 22 | | | | 55200 |
| 7209 | 22 | | | | 88300 |
| 7238 | 22 | | | | 102000 |
| 7115 | 22 | | | | 65800 |

NQ = Not quantifiable (< 5.00 ng/mL)

a. Samples were taken at either 8 or 22 hours post-dose, after the final pup sample timepoint from the litter was taken.

**Study title: GSK716155: Subcutaneous Postnatal Development Study in Mice**

| | |
|---|---|
| Study no.: | 2010N104536 ([redacted] (b) (4) AFA00842) |
| Study report location: | eCTD 4.2.3.5.3 |
| Conducting laboratory and location: | [redacted] (b) (4) (b) (4) |
| Date of study initiation: | 17-AUG-2010 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # JCM-203448-081, ≥ 95% pure |

**Key Study Findings**

- This study was terminated early due to excessive deaths and poor health of a high percentage of the dams during the lactation phase of the study. An extensive analysis of blood and fecal samples failed to identify a plausible causative (micro)organism (virus, bacteria, parasite) for the excessive mortality; however some samples were positive for *Clostridium perfringens*.
- Sponsor hypothesizes that the deaths are due to lactational ileus (described above in the review of study CD2008/01156).
- It is plausible that albiglutide contributed towards the severity of lactational ileus as discussed in more detail in the Integrated Summary, below.
- No NOAEL was established in this study for maternal toxicity, and pre-/post-natal endpoints were not evaluated.



Reference ID: 3347159

**Methods**

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | 10 mM Sodium Phosphate, 2.8% mannitol, 4.2% trehalose, and 0.01% polysorbate 80, pH 7.2 |
| Species/Strain: | Mated CD-1 mouse |
| Number/Sex/Group: | 24 |
| Satellite groups: | none |
| Study design: | F0♀ were allowed to deliver naturally. F0♀ were dosed daily from Days 7 to 21 pp. |
| Deviation from study protocol: | Because of the high incidence of unexpected morbidity & mortality in the F0♀s the study was terminated early, and all surviving F0 ♀s & pups were euthanized between Days 17 to 22 pp. For the analyses reported below, the F0♀s are divided into subset 1 (found dead, euthanized, or severe clinical signs requiring veterinary care) and subset 2 (survivors). |

**Observations and Results**



Reference ID: 3347159

**$F_0$ Dams (subset 1): 3, 8, 11, 10 (C, LD, MD, HD, respectively)**

| | |
|---|---|
| Survival: | 2, 7, 10 & 9 (C, LD, MD, HD, respectively) were found dead or sac'd prior to scheduled termination due to adverse clinical signs. Most early decedents died between Days 11 and 16 pp. |
| Clinical signs: | Fecal/urine staining, ↓ motor activity, lacrimation, ptosis, cold to touch, dehydration, low carriage, hunched posture, bradypnea/tachypnea, abnormal feces, ataxia, abdominal distention *(Sponsor's text table 1, below).* |
| Body weight: | BW ↓ at HD and ↓ BW gain at MD. Sudden overnight BW loss (up to 24%) just prior to death, early sac or veterinary intervention *(Sponsor's text table 2, below)*. |
| Feed consumption: | d.d. ↓ Days 7 to 11 pp. |
| Uterine content: | No effect. |
| Necropsy observation: | No clear effect of test item. 2/11 MD and 2/10 HD dams had intestines distended with firm material. |
| Toxicokinetics: | Not determined |
| Dosing Solution Analysis: | Results for all dose formulations met the acceptance criteria for concentration (within ±15% of nominal concentration). |
| Other: | Assessments made for pathogens (viral & bacterial) and/or parasites in a subset of animals did not reveal a definitive cause for the excessive morbidity/mortality observed in this study. |



Reference ID: 3347159

**Text Table 1: Clinical observations of F0 females Found Dead, Terminated Early or had Unexpected Severe Clinical Signs Requiring Veterinary Care**

| Dam No. /Mode of Death and Study Day | Clinical Observation | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Fecal Staining or Urine Staining | Decreased Motor Activity | Lacrimation | Ptosis | Cold to touch | Dehydration Mild/ Moderate/ or Severe | Low Carriage | Hunched posture | Bradypnea/ Tachypnea | Abnormal Feces[b] | Ataxia | Abdominal Distension |
| **Group 1 (0 (Vehicle) mg/kg/day)** | | | | | | | | | | | | |
| No. 6707 FD-Day 13 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | | Day 12 pp | | |
| No. 6714 FD-Day 13 pp | | | | | | | | | | Day 12 pp | | |
| 6715[a] | | | | Day 13 pp | Day 13 pp | Days 13-14 pp | | Days 13-14 pp | | Day 13 pp | | |
| **Group 2 (1 mg/kg/day)** | | | | | | | | | | | | |
| No. 6719 FD-Day 15 pp | | Day 14 pp | | | | | | Day 14 pp | | Day 14 pp | | |
| No. 6720 FD--Day 15 pp | | | | | | | | | | | | |
| No. 6726 FD-Day 14 pp | | Day 13 pp | | Day 13 pp | Day 13 pp | | | Day 13 pp | | Day 13 pp | | |
| No. 6730 FD-Day 15 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | | Day 14 pp | | Day 14 pp | | |
| No. 6732 FD-Day 16 pp | Day 15 pp | Day 15 pp | | Day 15 pp | Day 15 pp | Day 15 pp | | Day 15 pp | | Days 14-15 pp | Day 15 pp | Day 15 pp |
| No. 6733 FD-Day 13 pp | | | | | | | | | | | | |
| No. 6734 FD-Day 14 pp | Day 14 pp | Day 14 pp | | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | | Day 14 pp Tachypnea | Day 14 pp | | |
| No. 6738[a] | | Day 14 pp | | Day 14 pp | | Days 14-15pp | | Day 14 pp | | | | |

a. F0 dams under veterinary care at study termination. These live dams were sent to [redacted] (b) (4) for health monitoring evaluation.
b. Abnormal feces include soft or liquid, no feces in cage pan, scant, mucoid or dark feces.
FD = Found Dead

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**Text Table 1 (Continued)**

| Dam No. /Mode of Death and Study Day | Clinical Observation | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Fecal Staining or Urine Staining | Decreased Motor Activity | Lacrimation | Ptosis | Cold to touch | Dehydration Mild Moderate Severe | Low Carriage | Hunched posture | Bradypnea/ Tachypnea | Abnormal Feces[a] | Ataxia | Abdominal Distension |
| **Group 3 (5 mg/kg/day)** | | | | | | | | | | | | |
| No. 6741 FD-Day 15 pp | | Day 15 pp | | Day 15 pp | Day 15 pp | Day 15 pp | Day 15 pp | Days 14-15 pp | | Days 14-15 pp | | |
| No. 6746 FD-Day 16 pp | Day 16 pp | Day 16 pp | | Day 16 pp | Day 16 pp | Day 16 pp | | Day 16 pp | Day 16 pp Bradypnea | | Day 16 pp | |
| No. 6751 UE-Day 16 pp | Days 14-16 pp | Day 15 pp | | Day 15 pp | Days 15-16 pp | Days 15-16 pp | | Days 15-16 pp | | Day 16 pp | | |
| No.6752[b] FD – Day 11 pp | | | | | | | | | | | | |
| No. 6753[c] FD-Day 13 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | Day 12 pp | | Day 12 pp | Day 12 pp | |
| No. 6754[d] FD-Day 16 pp | Days 13-16 pp | Day 16 pp | | Day 16 pp | Day 16 pp | Days 15-16 pp | Day 16 pp | | | Day 15 pp | Day 16 pp | Days 15-16 pp |
| No. 6756[e] UE-Day 15 pp | Day 15 pp | Day 15 pp | | Day 15 pp | Day 15 pp | Day 15 pp | Day 15 pp | Day 15 pp | | Day 15 pp | Day 15 pp | |
| No. 6757[f] | Days 12-13 pp | Day 12 pp | Day 13 pp | Day 12 pp | Day 12 pp | Days12-13 pp | | Days 12-13 pp | | Days 12-14 pp | | Days 16-17 pp |
| No. 6759 FD-Day 12 pp | | | | | | | | | | | | |
| No. 6760 FD-Day 14 pp | Day 13 pp | | | | | | | Day 13 pp | | | | |
| No. 6764 FD-Day 13 pp | Day 13 pp | Day 13 pp | | Day 13 pp | Day 13 pp | Day 13 pp | | Day 13 pp | | | | |

a. Abnormal feces include soft or liquid, no feces in cage pan, scant, mucoid or dark feces.
b. Gasping, moribund on Day 11 pp
c. Head tremors on Day 12 pp
d. Head tremors on Day 16 pp
e. Both hindlimbs - splayed on Day 15 pp
f. F0 dams under veterinary care at study termination. These live dams were sent to [redacted] (b) (4) for health monitoring evaluation.
FD = Found Dead
UE = Unscheduled Euthanasia

**Text Table 1 (Continued)**

| Dam No. /Mode of Death and Study Day | Clinical Observation | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Fecal Staining or Urine Staining | Decreased Motor Activity | Lacrimation | Ptosis | Cold to touch | Dehydration Mild Moderate Severe | Low Carriage | Hunched posture | Bradypnea/ Tachypnea | Abnormal Feces[a] | Ataxia | Abdominal Distension |
| Group 4 (50 mg/kg/day) | | | | | | | | | | | | |
| No. 6765[b] FD-Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | Day 14 pp | | Day 14 pp Bradypnea | | | |
| No. 6773 FD Day 17 pp | Day 17 pp | Day 17 pp | | Day 17 pp | Day 17 pp | Day 17 pp | Day 17 pp | Day 17 pp | Day 17 pp Bradypnea | Days 16-17 pp | Day 17 pp | |
| No. 6775 FD-Day 16 pp | | Day 15 pp | | Day 15 pp | | Day 15 pp | | Day 15 pp | | Day 15 pp | | |
| No. 6778[c] FD-Day 13 pp | Day 13 pp | Day 13 pp | Day 13 pp | Day 13 pp | | Day 13 pp[a,d] | | Day 13 pp | Day 13 pp Bradypnea | | | |
| No. 6780[e] UE-Day 15 pp | Days 14-15 pp | Days13-15 pp | Days 13-15 pp | Days 13-15 pp | Day 13 pp | Days 13-15 pp[a,d] | Days 13-15 pp | | | Days 13-15 pp | Day 13 pp | |
| No. 6781 FD-Day 19 pp | | | | | | | | | | Day 14 pp | | |
| No. 6782 UE-Day 12 pp | Day 12 pp | Day 12 pp | | Day 12 pp | Day 12 pp | Day 12 pp[d] | Day 12 pp | Day 12 pp | Day 12 pp Bradypnea, Tachypnea | Day 12 pp | Day 12 pp | |
| No. 6783[e] | | Days 12-16 pp | | Days 12-13 pp | Day 12 pp | Days 13-17 pp | | Days 12-17 pp | | Days 12-18 pp | | |
| No. 6784 FD-Day 12 pp | - | - | - | - | - | - | - | - | - | - | - | - |
| No. 6785 FD-Day 12 pp | Day 11 pp | Day 11 pp | | Day 11 pp | Day 11 pp | Day 11 pp | | Day 11 pp | | Day 11 pp | Day 11 pp | |

a. Abnormal feces include soft or liquid, no feces in cage pan, scant, mucoid or dark feces.
b. No movement when encouraged on Day 14 pp
c. Head tremors on Day 13 pp
d. Thin body condition also observed
e. F0 dams under veterinary care at study termination. These live dams were sent to (b) (4) for health monitoring evaluation.
FD = Found Dead UE = Unscheduled Euthanasia

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**Text Table 2: Sudden Body Weight Loss in F0 Females Found Dead, Terminated Early, or had Unexpected Severe Clinical Signs Requiring Veterinary Care**

| Percentage Body Weight Loss | | | |
|---|---|---|---|
| Group Mg/kg/day | Dam Number | Day Postpartum | % Body Weight Loss |
| Group I - 0(Vehicle) | 6707 | Days 11 to 12 pp | 18% |
| | 6714 | Days 11 to 12 pp | 4% |
| | 6715 | Days 12 to 13 pp | 12% |
| Group II – 1 mg/kg/day | 6726 | Days 12 to 13 pp | 3% |
| | 6730 | Days 13 to 14 pp | 15% |
| | 6732 | Days 14 to 15 pp | 17% |
| | 6734 | Days 13 to 14 pp | 11% |
| | 6738 | Days 13 to 14 pp | 9% |
| Group III – 5 mg/kg/day | 6746 | Days 15 to 16 pp | 17% |
| | 6751 | Days 14 to 15 pp | 18% |
| | 6756 | Days 14 to 15 pp | 24% |
| | 6757 | Days 11 to 12 pp | 21% |
| | 6760 | Days 11 to 12 pp | 3% |
| Group IV – 50 mg/kg/day | 6773 | Days 16 to 17 pp | 18% |
| | 6775 | Days 14 to 15 pp | 14% |
| | 6780 | Days 12 to 13 pp | 14% |
| | 6781 | Days 13 to 14 pp | 11% |
| | 6783 | Days 11 to 12 pp | 11% |

**$F_0$ Dams (subset 2): 21, 16, 13, 14 (C, LD, MD, HD, respectively)**

| | |
|---|---|
| Survival: | By definition this is the subset of F0♀s that survived to (early) study termination. |
| Clinical signs: | No effect |
| Body weight: | ↓ BW gain at MD & HD and transient BW loss at HD. |
| Feed consumption: | ↓ FC at MD & HD. From Days 7 to 15 pp FC was 0.93x and 0.74x C in MD & HD groups, respectively |
| Uterine content: | No effect |
| Necropsy observation: | No effect |
| Toxicokinetics: | Plasma levels (µg/mL) measured 6 h after last dose (Day 21). Ranges as follows: LD = 0.0303 to 1.03, MD = 9.75 to 20.6, HD = 94.4 to 415. Drug was detected in samples from 2/5 C animals. This appears to reflect post-collection contamination as the levels were lower than those seen in LD animals. |
| Dosing Solution Analysis | Results for all dose formulations met the acceptance criteria for concentration (within ±15% of nominal concentration). |
| Other: | |

**$F_1$ Generation (from subset 2 dams only)**

| | |
|---|---|
| Survival: | No effect |
| Clinical signs: | No effect |
| Body weight: | d.d. ↓ in BW beginning PND 10 and persisting to the end of the study. On PND 21 the group mean weights were 0.84X, 0.87X and 0.76X the group control mean for LD, MD & HD, respectively. |
| Feed consumption: | Not assessed (study terminated before weaning) |
| Physical development: | Not assessed (study terminated) |
| Neurological assessment: | Not assessed (study terminated) |
| Reproduction: | Not assessed (study terminated) |
| Other: | |

$F_2$ Generation

| | |
|---|---|
| Survival: | Not assessed (study terminated) |
| Body weight: | Not assessed (study terminated) |
| External evaluation: | Not assessed (study terminated) |
| Male/Female ratio: | Not assessed (study terminated) |



Reference ID: 3347159

**Study title: GSK716155: Subcutaneous Postnatal Development Study in Mice**

| | |
|---|---|
| Study no.: | 2011N119858 (b) (4) 20013659) |
| Study report location: | eCTD 4.2.3.5.3 |
| Conducting laboratory and location: | (b) (4) |
| Date of study initiation: | 19-JUL-2011 |
| GLP compliance: | Yes |
| QA statement: | Yes |
| Drug, lot #, and % purity: | GSK716155, lot # GAJB05DA, ≥ 96.7% pure |

**Key Study Findings**

- In this study, as compared to study # 2010N104536, above, F0 dams received a high energy mouse diet throughout the study and litter sizes were reduced to 8 pups/dam by culling on Day 4 pp, thereby decreasing metabolic demand during lactation and increasing caloric input. This appeared to mitigate the high death rate seen in lactating dams in study # 2010N104536, consistent with Sponsor's hypothesis that the deaths in the earlier study were due to lactational ileus. There were nonetheless 1 LD and 1 HD F0 dam that died or was euthanized during the lactation period with signs consistent with lactation ileus.
- The NOAEL for F0 maternal toxicity is considered to be < 1mg/kg/day on the basis of mortality at 1 mg/kg/day.
- The NOAEL for F0 reproductive function is considered to be 50 mg/kg/day.
- The NOAEL for postnatal development is < 1mg/kg/day on the basis of slightly decreased pre-weaning body weight gain at all doses.
- Anti-drug antibodies were apparent in the majority of the drug-treated animals assessed. However there is no evidence that the ADA interfered with the PK or PD of the test article, and it is judged that the ADA do no impede interpretation of the study.



Reference ID: 3347159

**Methods**

| | |
|---|---|
| Doses: | 0, 1, 5, 50 mg/kg |
| Frequency of dosing: | Daily |
| Dose volume: | 2 mL/kg |
| Route of administration: | SC injection |
| Formulation/Vehicle: | 10 mM Sodium Phosphate, 2.8% mannitol, 4.4% trehalose, and 0.01% polysorbate 80, pH 7.2 |
| Species/Strain: | Mated CD-1 mouse |
| Number/Sex/Group: | 34 |
| Satellite groups: | none |
| Study design: | F0♀ were allowed to deliver naturally. Litters were culled to 4/sex/litter on Day 4 pp. F0♀ were dosed daily from Days 7 to 21 pp. On PND 21, F1 males and F1 females (2/sex/litter) were assigned to each dose group and assigned to one of two subsets. Subset 1 F1 animals were assigned to acoustic startle habituation (PND 60 ± 2) and reproductive performance. Toxicokinetics were evaluated on F0 maternal and F1 pup plasma. Immunogenicity was evaluated on main study F0 dams. Mated F1 females assigned to Subset 1 were allowed to deliver naturally and the dams and F2 litters were evaluated until Day 7 pp. Subset 2 F1 animals were assigned to acoustic startle habituation (PND 29 ± 2), motor activity (PND 60 ± 2), and Morris Watermaze evaluations (PND 78 ± 12).<br>F0 dams received Pelleted PicoLab™ High Energy Mouse Diet in an attempt to limit lactation ileus. After weaning F1 mice received Certified Rodent Diet® #5002. |
| Deviation from study protocol: | None material to the interpretation of the study. |



Reference ID: 3347159

**Observations and Results**

$F_0$ Dams

| | |
|---|---|
| Survival: | Test article-related deaths/early terminations at 1 and 50 mg/kg/day.<br>One LD♀ (2844) was found dead on Day 11 pp. Clinical signs of soft/liquid/scant feces, mild dehydration, decreased motor activity, ptosis, hunched posture and bradypnea were observed. This dam had a 16% body weight loss the day before death. Food consumption during the dosing period was unremarkable. At necropsy, the intestines were distended with gas. All 8 pups from this litter appeared normal.<br>One HD♀ (2921) was euthanized on Day 8 pp after a 21% body weight loss (from Day 7 pp) with clinical signs of mild dehydration (based on skin turgor). Food consumption during the lactation period was unremarkable. This mouse was euthanized in order to perform a necropsy on fresh tissue, obtain clinical endpoints and a cecal sample to investigate the cause of sudden body weight loss and clinical signs. At necropsy, all tissues appeared normal.<br>These deaths/early termination were plausibly test article-related based on similar findings in prior postnatal studies with GSK716155 [CD2008/01156/01 and 2010N104536_00]. |
| Clinical signs: | In treated groups up to 2 of 34 dams had signs of soft/liquid feces and/or mild dehydration on one or two occasions. Not seen in controls. |
| Body weight: | During the dosing period there was d.d. ↓ in BW gain with frank 7% BW loss following the 1st dose at HD (*see Sponsor's figure 1, below).* |
| Feed consumption: | Consistent with BW effects, there was a d.d. ↓ in FC during the dosing period (s.s. at HD). For example, from Day 7 to 14 pp FC was ↓ 3%, 10% & 22%(s.s.) at LD, MD & HD, compared to C. |
| Uterine content: | Natural delivery. See F1 table. |
| Necropsy observation: | No test item effect |
| Toxicokinetics: | Plasma levels (µg/mL) measured 6 h after last dose (Day 21). Means (ranges) as follows: LD = 0.053 (0.007 to 0.21), MD = 9.0 (1.26 to 19), HD = 91 (40.6 to 112). Drug was undetectable in C dam plasma. |
| Immunogenicity: | ADA were detected in 6/8, 6/8 and 5/8 LD, MD & HD dams assessed Day 21 pp. Titers ranged between 1:22 to 1:3935. |
| Dosing Solution Analysis | Results for all dose formulations met the acceptance criteria for concentration (within ±15% of nominal concentration). |
| Other: | |



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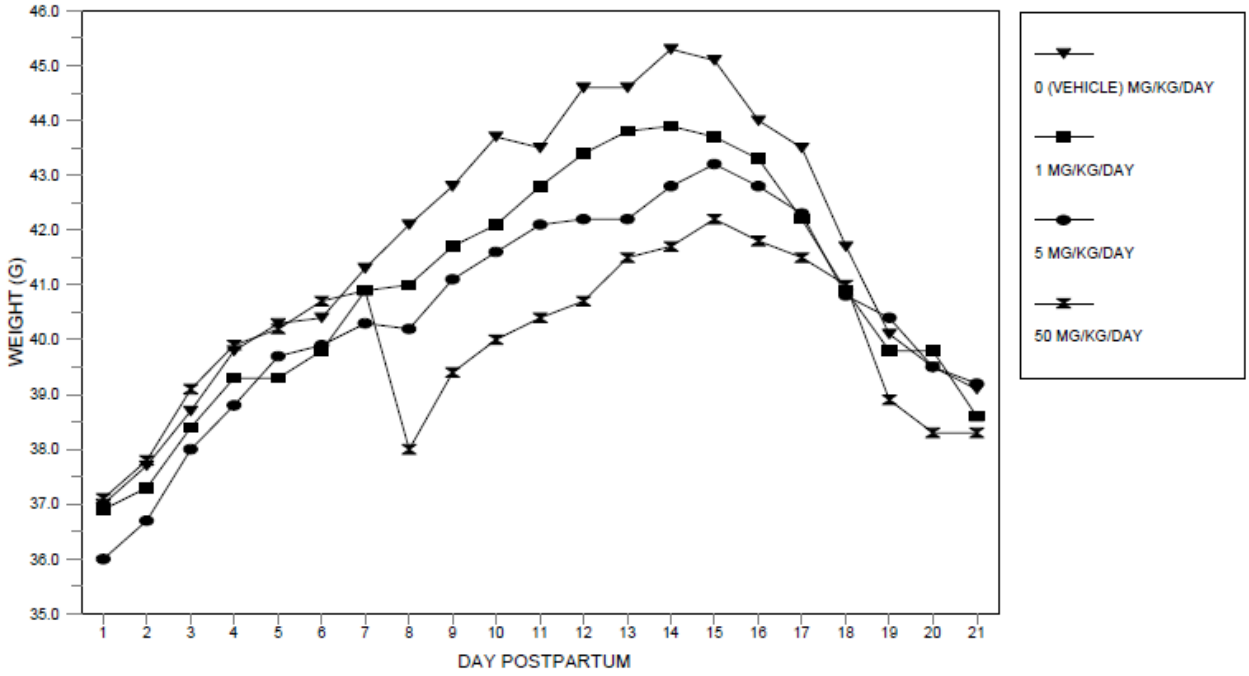
MATERNAL BODY WEIGHTS - POSTPARTUM - F0 GENERATION FEMALES

Figure 1



$F_1$ Generation

| | |
|---|---|
| Survival: | No test item-related effect. |
| Clinical signs: | *Pre-weaning*: one or more pups had mild to moderate dehydration in 0/32 C, 1/32 LD, 1/34 MD & 2/33 HD litters.<br>*Post-weaning:* no test item-related effect. |
| Body weight: | *Pre-weaning*: Beginning Day 7 pp (1st day of dam dosing) d.d. ↓ in BW gain, such that by PND 21 all dose groups have s.s. lower weight than C: ↓3%, 5%, 7% in LD, MD & HD, respectively.<br>*Post-weaning:* No effect on BW gain, although BW differences established during the pre-weaning period tend to persist (*Sponsor's figures 2 & 3, below).*<br>*Post-coital ♀:* no effect on BW gain. |
| Feed consumption: | *Not assessed.* |
| Physical development: | ♂: No effect.<br>♀: s.s. delayed vaginal patency at HD (Day 27.2 vs. 25.6 in C) is likely a consequence of ↓ pre-weaning BW effects, as BW at attainment of vaginal patency was unaffected (21.1 g for both HD & C). |
| Neurological assessment: | *Motor activity*: no effect.<br>*Acoustic startle*: no effect.<br>*Morris water maze:* no effect. |
| Reproduction: | Drug-treated ♀s showed an increased tendency towards diestrus lasting ≥ 6 days: C=0/24, LD=2/24, MD=1/24, HD=3/24. Nonetheless all females became pregnant.<br>No other effects on mating or reproductive parameters were apparent. |
| Toxicokinetics: | Plasma levels assessed on PND 10. Marginally quantifiable levels of drug (just above the limit of quantitation of 5 ng/mL) were present in some pooled samples at some time points in all dose groups and did not show a dose response. This reviewer concludes that transfer of drug to offspring was negligible. |



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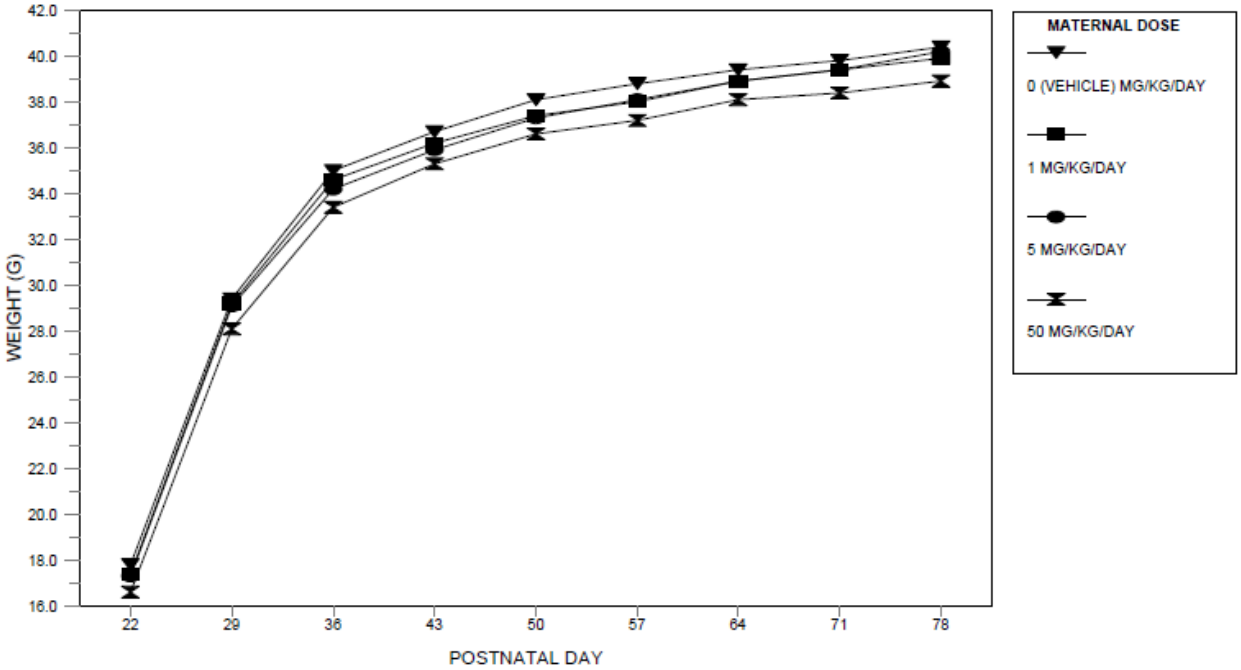
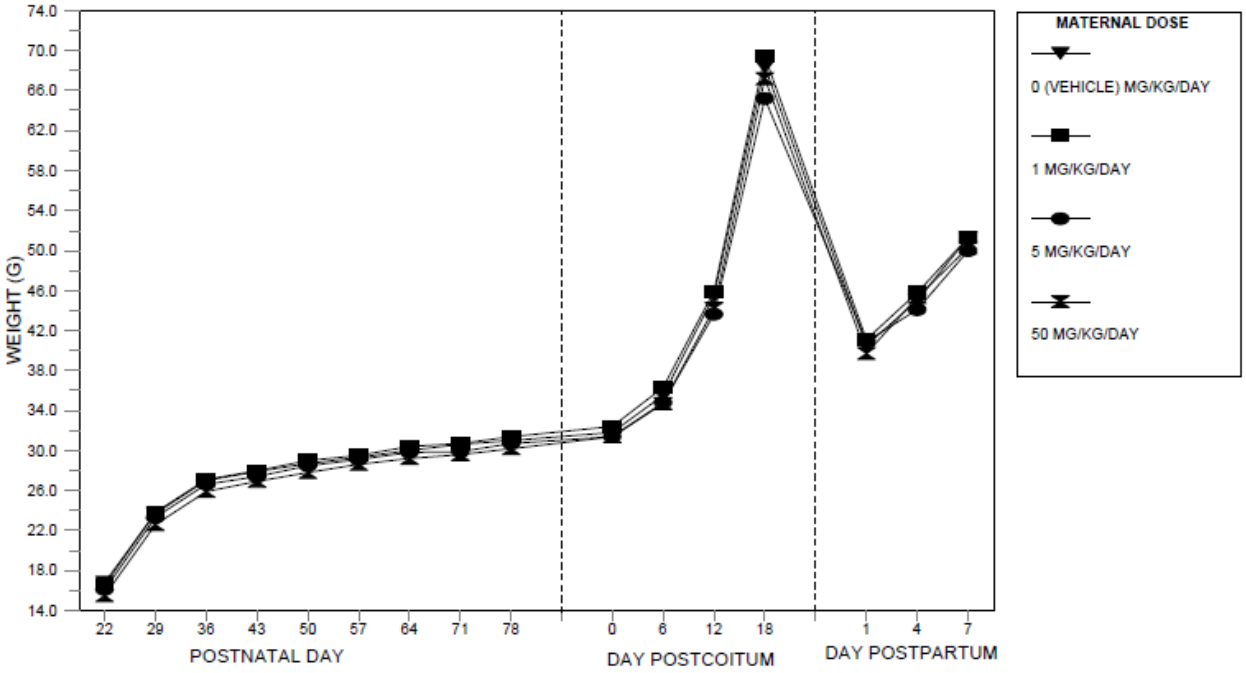
BODY WEIGHTS - F1 GENERATION MALES
Figure 2




BODY WEIGHTS - F1 GENERATION FEMALES
Figure 3



$F_2$ Generation

Survival: The number of pups found dead or presumed cannibalized was s.s. ↑ at LD & MD, but not HD. A corresponding inverse decrease in viability index was seen (*Sponsor's Table B17, below*). Lacking d.d., this finding was not considered to be drug related.

Body weight: No effect.

External evaluation: No effect.

Male/Female ratio: No effect.

TABLE B17 (PAGE 1): LITTER OBSERVATIONS (NATURALLY DELIVERED PUPS) - SUMMARY - F2 GENERATIONS LITTERS

SUBSET 1

| MATERNAL DOSE GROUP | | 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|
| MATERNAL DOSE (MG/KG/DAY) | | 0 (VEHICLE) | 1 | 5 | 50 |
| DELIVERED LITTERS WITH ONE OR MORE LIVEBORN PUPS | N | 22 | 21 | 22 | 24 |
| PUPS DELIVERED (TOTAL) | N | 296 | 312 | 296 | 334 |
| | MEAN±S.D. | 13.4 ± 3.3 | 14.8 ± 2.6 | 13.4 ± 4.0 | 13.9 ± 1.6 |
| LIVEBORN | MEAN±S.D. | 13.1 ± 3.6 | 14.7 ± 2.7 | 13.4 ± 4.1 | 13.9 ± 1.6 |
| | N(%) | 289( 97.6) | 308( 98.7) | 294( 99.3)** | 334(100.0)** |
| STILLBORN | MEAN±S.D. | 0.3 ± 0.7 | 0.1 ± 0.5 | 0.1 ± 0.4 | 0.0 ± 0.0 |
| | N(%) | 6( 2.0) | 3( 0.0) | 2( 0.7) | 0 |
| UNKNOWN VITAL STATUS a | N | 1 | 1 | 0 | 0 |
| PUPS FOUND DEAD OR PRESUMED CANNIBALIZED | | | | | |
| DAY 1 | N/N(%) | 1/289( 0.3) | 0/308( 0.0) | 3/294( 1.0) | 0/334( 0.0) |
| DAYS 2- 4 | N/N(%) | 1/288( 0.3) | 14/308( 4.5)** | 22/291( 7.6)** | 9/334( 2.7) |
| DAYS 5- 7 | N/N(%) | 3/287( 1.0) | 7/294( 2.4) | 2/269( 0.7) | 1/325( 0.3) |
| VIABILITY INDEX b | % | 99.3 | 95.4 | 91.5 | 97.3 |
| | N/N | 287/289 | 294/308* | 269/294** | 325/334 |
| LACTATION INDEX c | % | 99.0 | 97.6 | 99.2 | 99.7 |
| | N/N | 284/287 | 287/294 | 267/269 | 324/325 |

DAY(S) = DAY(S) POSTPARTUM
a. Maternal cannibalization precluded identification of vital status at birth.
b. Number of live pups on Day 4 postpartum/number of liveborn pups on Day 1 postpartum.
c. Number of live pups on Day 7 postpartum/number of live pups on Day 4 postpartum.
* Significantly different from the vehicle control group value (p≤0.05).
** Significantly different from the vehicle control group value (p≤0.01).

# 10 Special Toxicology Studies

## IMMUNOGENICITY ASSESSMENTS

### ***In Silico* Assessment**

**Report 2012N143627**: An *in silico* assessment was conducted to determine the immunogenicity potential of albiglutide in humans. The sequence of albiglutide was scanned for the presence of putative HLA class II restricted epitopes, also known as T helper (Th)-epitopes, using Epibase® immunoprofiling. Affinities for DRB1, DRB3/4/5, DQ and DP HLA class II receptors were estimated for all possible (b) (4) (b) (4) derived from albiglutide's sequence and were classified as strong, medium or weak/non binders based on affinities. Of particular interest is the immunogenicity of (b) (4) ee Sponsor's table below). This indicates a potential for anti-albiglutide immunogenicity in the clinic, and predicts which HLA allotypes are at the greatest risk of mounting an anti-albiglutide immune response, but makes no prediction as to the clinical relevance of an anti-albiglutide immune response.

(b) (4)

A comparable analysis was also conducted on (b) (4)

(b) (4)

(b) (4)

(b) (4)

***In Vivo* Assessment**

**Report RD2005/01167**: The immunogenicity of albiglutide was assessed in female CD-1 mice administered SC albiglutide (2.25 or 7.5 mg/kg) TIW for 4 weeks. Binding ADA and circulating albiglutide levels were assessed, and albiglutide PD was assessed by OGTT. All albiglutide treated mice developed anti-albiglutide antibodies (as early as Day 8). Titers of anti-albiglutide antibodies increased with time and were not dose-dependent. The formation of ADA correlated with a decrease (Day 8) or absence (Day 14) of detectable circulating albiglutide. Pharmacological activity of albiglutide was evident during the first two weeks of the study at both 2.5 and 7.5 mg/kg as significant decreases in blood glucose levels and/or glucose AUC of OGTT. During the second two weeks, the only pharmacological activity observed was significant decrease in glucose AUC of OGTT on Day 25 at 7.5 mg/kg compared to vehicle control. The relationship of the Day 25 decreased AUC of OGTT to albiglutide treatment is unclear since circulating albiglutide levels were undetectable by Day 14. These data indicate that the emergence of anti-albiglutide antibodies markedly altered the pharmacokinetic and pharmacodynamic profile of albiglutide in this study.

**Report RD2005/01168**: The immunogenicity of albiglutide and a murine surrogate (mAlb) was assessed in male db/db mice administered SC albiglutide or murine surrogate (0.8 mg/kg) TIW for two 4-week cycles (with a 1 week washout period between each cycle). The murine surrogate differs from albiglutide in the substitution of mouse albumin sequence for human albumin sequence; the tandem $Gly^8$ GLP-1 sequence remains the same. Both albiglutide and mAlb were equipotent in an *in vitro* bioassay. Binding ADA levels were measured only at the end of the study. Circulating drug levels were not measured. The ability of mice to continue to respond to albiglutide, mAlb or GLP-1 was assessed by fasting blood glucose and OGTT. All mice (11 of 11) treated with albiglutide had positive anti-albiglutide antibody titers. In contrast, only 25% (3 of 12) mice treated with mAlb had positive anti-mAlb antibody titers, and anti-mAlb antibody titers were lower than anti-albiglutide titers (anti-mAlb titer range of 567 to 764 compared to anti-albiglutide titer range from 414 to 11328). In this study the presence of either anti-albiglutide or anti-mAlb antibodies did not appear to diminish the

effectiveness of drug-related improvement in glucose clearance following OGTT conducted through the end of dosing, suggesting the ADAs were non-clearing and non-neutralizing.

**Report RD2005/01135**: The immunogenicity of albiglutide and a murine surrogate (mAlb) was assessed in male CD-1 mice administered 0.8 mg/kg SC albiglutide or murine surrogate (mAlb) TIW for three 4-week cycles (with a 1 week washout period between each cycle). The murine surrogate differs from albiglutide in the substitution of mouse albumin sequence for human albumin sequence; the tandem $Gly^8$ GLP-1 sequence remains the same. Both albiglutide and mAlb were equipotent in an *in vitro* bioassay and displayed similar activity levels in single-dose oral glucose tolerance tests (OGTT) in mice. Binding ADA levels were measured only at the end of the study. Circulating drug levels were not measured. The continued pharmacological responsiveness of the mice to albiglutide, mAlb and GLP-1 was periodically assessed by fasting blood glucose levels and/or OGTT. All mice (100%) that were given albiglutide were positive for anti-albiglutide antibodies. In contrast, only 7 of 24 (29%) mice receiving mAlb were positive for antibodies to mAlb, and anti-mAlb titers were generally several orders of magnitude lower than the anti-albiglutide titers (anti-mAlb titer range of 340 to 2,719 compared to anti-albiglutide titer range from 9,630 to 3,698,243). Mice treated for 15 weeks with mAlb retained effectiveness in improved glucose clearance compared to vehicle controls following an OGTT, even in mice that developed anti mAlb antibodies. In contrast, after 8 weeks of treatment with albiglutide, mice were no longer responsive to albiglutide in the OGTT, presumably due to anti albiglutide antibodies. Mice challenged with GLP-1 after 15 weeks of albiglutide treatment still exhibited improved glucose clearance compared to vehicle controls (though not statistically significant), suggesting that anti-albiglutide antibodies did not significantly cross-react with endogenous GLP-1.

**Table 3 Summary of statistical analyses for $AUC_{glucose}$ differences during chronic dosing**

| Study Week | mAlb % AUC Drop From Vehicle | P Value | hAlb % AUC Drop From Vehicle | P Value |
|---|---|---|---|---|
| 2 | 26 | < 0.001 | 22 | < 0.001 |
| 4 | 31 | < 0.001 | 12 | < 0.05 |
| 6 | 31 | < 0.001 | 17 | < 0.001 |
| 8 | 31 | < 0.001 | 5 | NS |
| 9 | 29 | < 0.001 | 3 | NS |
| 12 | 23 | < 0.001 | -5 | NS |
| 14 | 26 | < 0.001 | 0 | NS |

NS: not significant.

**Report RD2005/00575**: Male SD rats received daily SC injections of albiglutide at 5 or 50 mg/kg. Plasma concentrations of albiglutide increased until Day 8 then decreased



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during the remainder of the study. The reductions in mean plasma exposure to albiglutide were 75% and 46% by Day 15 for doses of 5 and 50 mg/kg/day, respectively. The reduction in individual rat exposure ranged from 34% to 99% over the same time frame. Anti-albiglutide antibodies were detected in 2 of 12 treated rats on Day 8, and in 11 of 12 treated rats on Day 22. The incidence and titer of the antibody response increased over time, continuing through the recovery period. During the treatment period, the incidence of antibodies was not related to the dose of albiglutide (on Day 15, 83% of the 5 mg/kg/day rats had a positive response compared to none of the 50 mg/kg/day rats). This non-dose responsive relationship may be due to an interference in the immunogenicity assay by high concentrations of albiglutide in the rat serum or to immunotolerance.

*NB – Immunogenicity assessments conducted in the nonhuman primate as part of the toxicological assessment have been reviewed as part of the toxicology study reviews and are not repeated here.*

# 11 Integrated Summary and Safety Evaluation

Albiglutide (GSK716155) is a recombinant fusion protein composed of 2 copies of [Gly$^{8}$]GLP-1$_{(7\text{-}36)}$, in tandem, fused to human albumin. The pharmacodynamic activity of albiglutide arises from agonism at the GLP-1R. The substitution of a glycine for the native alanine at position 8 renders the GLP-1 moieties to be resistant to DPP-IV-mediated proteolysis, thereby increasing the functional half-life. The albumin component conveys no specific pharmacodynamic activity of its own, but rather its presence substantially increases the circulatory half-life of the [Gly$^{8}$]GLP-1$_{(7\text{-}36)}$ moieties. This effect on increased half-life is presumably a consequence both of decreased filtration by the kidneys and enhanced recovery from the endosomes due to the ability of albumin to bind to the FcRn receptor at low pH. There is no evidence from the nonclinical toxicology program that the albumin moiety alters the expected toxicologic profile of the GLP-1 moiety. (b) (4)

**Pharmacology**

The reported effects of GLP-1R agonism are myriad; however the primary beneficial effect in the treatment of T2DM is increased glucose-dependent insulin secretion from β islet cells. GLP-1R agonists also reduce food consumption and delay gastric emptying and GI transit, thereby moderating post-prandial spikes in blood glucose level, and (at least in rodents) stimulate an increase in β-cell mass. Albiglutide demonstrated the above activities in appropriate rodent and/or non-rodent animal models. GLP-1 has also been reported to decrease glucagon secretion, and thereby decrease glucose production by the liver. Effects of albiglutide on glucagon secretion were not assessed.

The intrinsic activity of albiglutide at the GLP-1R, assessed *in vitro*, found comparable activity at mouse, rat and human GLP-1 receptors, but lower activity (~3-fold) at the



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monkey receptor, suggesting that monkeys may be less responsive than humans to similar doses.

Secondary pharmacology studies conducted in rats also showed that albiglutide can improve the oxidative coupling of glucose metabolism in explanted perfused rat hearts, and reduced myocardial infarct size and post-ischemic cardiac left ventricular function following myocardial ischemia/reperfusion injury in rats.

Safety pharmacology assessments did not identify any respiratory, cardiovascular or neurobehavioral safety signals.

**Pharmacokinetics**

As would be expected for a polypeptide of ~73,000 Da, albiglutide has a low volume of distribution in both mice and monkeys. The circulatory half-life was on the order of 8 hours in mice and 60 hours in monkeys. Bioavailability following a single SC injection was at least 38% in mice and 50% in monkeys. No meaningful sex-related differences in PK parameters were seen in either species. Accumulation of albiglutide was observed after repeated subcutaneous administration in mice and monkeys and was generally consistent with the $t_{1/2}$ and frequency of dosing. However, in some animals (particularly rodents), there was a reduction in the anticipated plasma concentrations due to the presence of anti-albiglutide antibodies, which were observed as early as Day 8 in mice and rats and Day 15 in monkeys. The presence of a rapid, drug-clearing, anti-albiglutide immune response largely precluded interpretable studies in rodents of more than ~2 weeks. In monkeys, anti-albiglutide antibodies did not significantly alter the pharmacokinetic properties of albiglutide, with the exception of high titer anti-albiglutide antibodies that were observed in only a few individual monkeys and only in the 26-week study. Sponsor's Table 9.1 provides, below, a comparative assessment of systemic exposures in mice, monkeys and humans following subcutaneous administration of albiglutide.

In accordance with current guidance, tissue distribution studies, drug metabolism studies and excretion studies were not conducted with albiglutide, as it is a protein-based therapeutic. An assessment of the effect of albiglutide on CYP mRNA levels in monkeys treated with albiglutide for 5 weeks found minimal increases in CYP1A2 message in males and minimal increases in CYP2B17 levels in females.



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**Table 9.1 Comparative Assessment of Systemic Exposure after Subcutaneous Administration of Albiglutide to Mice and Monkeys**

| Study Type (Report No.) | Dose[a] (mg/kg) | Sex | $C_{max}$ (µg/mL)[b] | $AUC_{0-168}$ (µg.h/mL)[b] | Multiples of Human Exposure | |
|---|---|---|---|---|---|---|
| | | | | | $C_{max}$ | $AUC_{0-168}$ |
| Mouse 7 day | 1 | F | 4.38 | 470[d] | 0.93 | 0.63 |
| (CD2006/01620) | 5 | F | 13.8 | 1652[d] | 2.9 | 2.2 |
| | **50** | **F** | **239** | **28917[d]** | **51** | **39** |
| Mouse 7 day | 5 | M | 18.1 | 2408[e] | 3.9 | 3.2 |
| Male mating fertility | 15 | M | 46.0 | 6412[e] | 9.8 | 8.5 |
| (CD2007/01279) | 50 | M | 185 | 25207[e] | 39 | 34 |
| Monkey 4 week | 5 | M+F | 36.1 | 2912[f] | 7.7 | 3.9 |
| (RD2005/01133) | **15** | **M+F** | **105** | **7820[f]** | **22** | **10** |
| | 50 | M+F | 505 | 43336[f] | 107 | 58 |
| Monkey 5 week[c] | 5 | M+F | 17.5 | 1057[g] | 3.7 | - |
| (CD2005/00839) | 15 | M+F | 71.2 | 4350[g] | 15 | - |
| | 50 | M+F | 260 | 15312[g] | 55 | - |
| Monkey 26 week | 1 | M+F | 6.03 | 591 | 1.3 | 0.79 |
| (RD2005/01281) | 5 | M+F | 32.0 | 3316 | 6.8 | 4.4 |
| | **15** | **M+F** | **121** | **13143** | **26** | **18** |
| Monkey 52 week | 15 | M+F | 155 | 16050 | 33 | 21 |
| (CD2009/00863) | **50** | **M+F** | **538** | **56100** | **114** | **75** |
| Clinical[h] | 50 mg weekly | M+F | 4.7 | 751 | - | - |

**Key:** No observed adverse effect level (NOAEL) is **bolded**.

– = Not applicable.

a = Administered daily to mice and weekly to monkeys unless otherwise noted.

b = End of study values unless otherwise noted. For comparative and interpretive purposes, values were converted to µg/mL ($C_{max}$) and µg.h/mL (AUC) and $AUC_{0-168}$ values extrapolated where necessary.

c = Administered as twice weekly doses of 2.5, 7.5 or 25 mg/kg/dose.

d = Data presented in original report as $AUC_{0-24}$ [m2.6.7, Table 7.1].

e = Data presented in original report as $AUC_{0-24}$ [m2.6.7, Table 12.2].

f = Data presented in original report as ng.day/mL [m2.6.5, Table 4.6].

g = $AUC_{0-72}$. Since albiglutide was administered twice weekly rather than once weekly, comparison to human $AUC_{0-168}$ was not assessed.

h = Estimated steady-state exposures in patients from Phase III clinical trials receiving the maximum proposed therapeutic dose of 50 mg/week albiglutide [m2.5, Section 3.2].

## Toxicity

### General Toxicity

An acute subcutaneous and intravenous study in the mouse and repeat subcutaneous dose studies for durations of up to 15 days in the mouse and 52 weeks in the monkey have been performed. Unsurprisingly, given the presence of human albumin sequence in the fusion protein, Albiglutide has proven to be highly immunogenic in animals. In mice, the anti-drug antibody (ADA) response caused diminished exposure to Albiglutide and was associated with type 3 hypersensitivity reactions (immune complex deposition) in a some of the mice. Therefore it was concluded that longer-term (exceeding ~2 weeks) studies with albiglutide in rodents would likely be uninformative to clinical risk. As a consequence of this, some of the DART studies were subdivided into multiple 2-week exposure periods (see DART section discussion below). In addition, it was not feasible to conduct chronic toxicity or 2-year carcinogenicity studies in rodents. While albiglutide was also immunogenic in monkeys, the ADA response did not appreciably alter exposure to active drug in the majority of monkeys, nor did it discernably contribute to the toxicity findings of the monkey studies.

Acute toxicity associated with single high doses of albiglutide was assessed in mice at doses of 50, 200, 500 and 1000 mg/kg SC and 500 mg/kg IV. A transient, dose-dependent body weight drop (max. ↓11-15%) was seen in both sexes at SC doses ≥200 mg/kg and the 500 mg/kg IV dose. Microscopic findings were observed in male reproductive tissues: epididymis (vacuolation and cell debris), seminal vesicle (vacuolation), and prostate (vacuolation and lymphocyte infiltration) of male mice at 500 mg/kg IV and in the epididymis (vacuolation and cell debris) at doses ≥ 500 mg/kg SC. The histopathology changes were more pronounced with the IV dose. The vacuoles were characterized as 1 or more small discrete intracytoplasmic vacuoles within the epithelium of the affected tissues. The maximum tolerated dose in this study is considered to be >1000 mg/kg SC and >500 mg/kg IV. Based on the microscopic findings in male reproductive tissue, the male NOAEL is 200 mg/kg SC (240X the MRHD) and <500 mg/kg IV. The female NOAEL is 1000 mg/kg SC (1200x the MRHD) and 500 mg/kg IV.

For the repeat-dose general toxicity studies, discussion focuses primarily on the monkey studies, as the interpretation of the rodent studies is confounded by the ADA response discussed above. In general the results of the monkey toxicology studies are consistent with the pharmacology at the GLP-1R: decreased food consumption, emesis, dehydration and decreased body weight gain (and occasionally transient frank body weight loss). These effects are most marked following the first few doses and become attenuated thereafter. Transient reductions in blood glucose were also seen following the first dose in the 4-week monkey toxicity study. In the pivotal 52-week toxicity study no dose-limiting toxicity was seen at doses up to 50 mg/kg/week (the highest dose tested). There were mild injection site reactions (minimal perivascular mononuclear cell infiltrates). Most notably, albiglutide (and exenatide) caused a dose-related increase in pancreas weight; however, there were no histopathologic abnormalities detected in the pancreases of albiglutide-treated monkeys. Stereological examination suggested that



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all components of the pancreas (islet, duct and acinar) exhibited a more or less proportional increase in volume. Also there was no significant increase in the percentage of cells undergoing proliferation in any particular cell population, as assessed by Ki67 immunostaining. It is unknown whether the increase in pancreas weight is reversible, as there was no recovery group in the 52 week study, and pancreas weight was not determined in any of the shorter duration toxicity studies. In the 52-week monkey study thyroids were scrutinized by a) standard H&E stain, b) calcitonin immunohistochemistry and c) quantitative (stereological) analysis of total C cell count; there were no albiglutide-related findings in the thyroid. The NOAEL for the 52-week monkey study was 50 mg/kg/week (60X the MRHD), the highest dose tested [Week 52 mean $AUC_{0-t}$ of 56.1 mg•h/mL (range 40.9 to 68.3 mg•h/mL) and mean $C_{max}$ of 0.538 mg/mL (range 0.425 to 0.648 mg/mL)].

Genotoxicity

No studies to assess the potential genotoxicity of albiglutide have been conducted. Albiglutide is not expected to interact directly with DNA or other chromosomal material; therefore, the use of standard genotoxicity studies is not considered appropriate.

Carcinogenicity

No studies to assess the potential carcinogenicity of albiglutide have been conducted. Because of the emergence of clearing anti-albiglutide antibodies by 14 days in rodents, meaningful 2-year rodent carcinogenicity studies with albiglutide are not feasible. Notably, per ICH S6 (R1), the use of species-specific homologues is not recommended for use in lifetime bioassays. Other long-acting GLP-1R agonists have been shown to cause C-cell neoplasms (adenomas and carcinomas) in rodents. The clinical significance of this activity in rodents is not yet clear. C-cell hyperplasia was not seen with albiglutide following 1-year treatment in monkeys, consistent with negative results in monkeys seen with other long-acting GLP-1R agonists. As noted above, pancreas weight was increased following 1-year exposure to albiglutide in monkeys; however this was not associated with an appreciable increase in the percentage of proliferating cells (assessed by Ki67 immunostaining) nor any other histopathologic abnormality.

Developmental and Reproductive Toxicity

**Male Fertility:** Daily SC injections of albiglutide (5, 15 or 50 mg/kg) for 7 days prior to cohabitation with females, and continuing during the cohabitation period, had no effect on mating, copulation or fertility indices or the number of days required to mate. Embryonic survival was unaffected by paternal exposure to albiglutide. Albiglutide was also without effect on male reproductive organ weight or histology. The expected dose-dependent effects of transiently decreased food consumption and decreased body weight were observed. The NOAEL for effects on male mating and fertility was 50 mg/kg/day (34X clinical exposure based on AUC), the highest dose level evaluated.

**Female Fertility:** Daily SC injections of albiglutide (1, 5 or 50 mg/kg) for 7 days prior to cohabitation with males, and continuing during the cohabitation period (but not following conception), had no s.s. effect on any measured parameter relating to mating and fertility. The mean estrus cycle length was n.s.s. increased at 50 mg/kg/day (39X



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clinical exposure based on AUC): 5.7 days vs. 4.8 days for C, resulting in a n.s.s. decrease in the mean number of estrus periods during the 7-day dosing period (0.9 estrus per week vs 1.3 in C). These differences appear to stem from an increase in the number of ♀s with prolonged diestrus (≥5 consecutive days ): 9/25 HD♀s vs. 1/25 C♀s. Intrauterine growth and survival were unaffected by test article administration at all dose levels. There were no test article-related external malformations, and no external developmental variations in any fetuses. 50 mg/kg/day albiglutide caused transiently decreased food consumption and body weight loss in the treated dams. The NOAEL for female reproductive toxicity was 5 mg/kg/day (2X clinical exposure based on AUC) on the basis of the effects on the estrus cycle at higher doses.

**Early embryonic development to implantation:** The objective of this study was to determine the potential of albiglutide to induce developmental toxicity when administered during the pre-implantation period by SC injection to female CD-1 mice at doses of 1, 5 or 50 mg/kg/day once daily on Days 0 to 6 post-conception (PC). Notably this was not assessed in the female fertility study, because of concerns regarding the appearance of clearing ADA with prolonged exposure. At 50 mg/kg/day albiglutide caused transiently decreased food consumption and body weight loss; however, there were no remarkable effects on early embryonic development or implantation parameters. The NOAEL for early embryonic development to implantation was considered to be 50 mg/kg/day (39X clinical exposure based on AUC), the highest dose tested.

**Embryofetal development:** Female CD-1 mice received daily SC injections of albiglutide (1, 5 or 50 mg/kg) from Days 6 to 15 PC. HD dams had decreased food consumption and transient body weight loss. There was also evidence of embryo/fetal lethality at the HD, with a 16.2% per litter postimplantation loss, compared to 3.8% per litter in the control group. The HD was also associated with a s.s. higher mean litter proportion of the skeletal developmental variation bent (wavy) ribs (8.0% per litter, compared to 0.0% per litter in C). There were no test article-related effects on intrauterine growth or survival at 1 and 5 mg/kg/day. Based on these findings, the NOAEL for embryofetal development is considered to be 5 mg/kg/day (2X clinical exposure based on AUC).

**Pre-natal and Post-natal Development**

*In order to minimize the impact of the clearing ADA response, this assessment was subdivided into three separate sub-studies, each with no more than 15 days of dosing: Part 1, Day 6 to Day 17 PC; Part 2, Day 15 PC to Day 10 post-partum (PP); Part 3, Day 7 to Day 21 PP.*

Part 1: Female CD-1 mice received daily SC doses of albiglutide (1, 5 or 50 mg/kg) from Day 6 to Day 17 PC. F0 toxicity (including ↓ body weight and ↓ food consumption during gestation, and abdominal distention, dehydration and pale ears during lactation) was seen at 50 mg/kg/day. 3 HD F0 litters had pups that experienced one or more days of dehydration. 2 HD F0 litters had pups that were cold to the touch and/or thin body condition. The HD F1 generation (♂&♀) had ↓ BW during the preweaning period. F1



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HD♂s were slightly s.s. delayed for preputial separation (PND 31.3 vs. PND 29.6 in C), possibly related to their persistently lower BW. There were no effects on F1 neurobehavioral development, reproductive function or F2 morphology or viability. Fetal albiglutide concentrations, assessed on Day 17 PC, were ~200 to 1000-fold lower than those detected in the dams. The NOAEL for maternal toxicity, F0 reproductive function and pre-/post-natal development is considered to be 5 mg/kg/day (2X clinical exposure based on AUC).

Part 2: Female CD-1 mice received daily SC doses of albiglutide (1, 5 or 50 mg/kg) from Day 15 PC to Day 10 PP. F0 toxicity (including ↓ body weight and ↓ food consumption during gestation, and abdominal distention, dehydration and pale ears during lactation) was seen at 50 mg/kg/day. There were also unexplained deaths in all F0 albiglutide treatment groups during the lactation period (0, 2, 5, and 2 deaths in the C, LD, MD & HD groups, respectively. These deaths are likely due to lactational ileus, as discussed in Part 3, below. F1 pups had ↓ body weight (all doses) at birth and reduced weight gain during lactation. There was rebound weight gain post-weaning. ♂ F1 pups had a dose-related, n.s.s. trend towards minimally delayed balanopreputial separation (BPS): PND 29.1, 30.7, 30.9 and 31.0 (C, LD, MD, HD). This may reflect generally lower body weights. Notably all BPS values were within the range of historical control values. F1 pups had a decrease in the force of the response to an auditory startle. Given that the startle response is measured as the force generated by the animal in response to the startle stimulus, and the magnitude of the force that can be generated is proportional to the mass of the animal, these relatively small effects may be secondary to the decreased bodyweights noted above. Drug treatment of the F0 dams had no effect on the reproductive success of the F1 adults, although there was a s.s. 25% ↓ in the number of estrus cycles at HD. There was a dose-related ↓ in the # of implantation sites (14.2, 13.6, 12.4 & 11.7 at C, LD, MD & HD, respectively); however the differences were n.s.s. and all values were within the historical control range. Nonetheless, this appeared to contribute to a dose-related ↓ in live litter size for the F2 generation (13.4, 12.7, 11.9 & 10.3 at C, LD, MD & HD, respectively). The value for HD was s.s. different from C and outside of historical control range. The NOAEL for pre-/post-natal development is considered to be < 1.0 mg/kg/day (0.6X clinical exposure based on AUC) on the basis of reduced bodyweight at all doses in the F1 generation.

Part 3: Female CD-1 mice received daily subcutaneous injections of albiglutide (1, 5, or 50 mg/kg) beginning on Day 7 up to Day 21 PP. This study was terminated early due to excessive deaths and poor health of a high percentage of the dams during the lactation phase of the study. The incidence of affected F0 females was 3, 8, 11 and 10 F0 mice in the control, 1, 5 and 50 mg/kg/day dose groups, respectively. Clinical signs seen shortly before death or unscheduled euthanasia were similar in treated, control and unassigned extra mice and generally included decreased motor activity, ptosis, cold to touch, dehydration, hunched posture, low carriage, ataxia and/or abnormal fecal output. Sudden weight loss (up to 24%) was observed prior to death/unscheduled euthanasia in the majority of these mice. Macroscopic observations at necropsy included gaseous distension of the intestines, stomach and/or abdominal cavity and dark red/black small intestines. Microscopic observation of the dark red small intestines included multifocal,



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moderate congestion, however no necrosis or inflammation was observed. An extensive analysis of blood and fecal samples failed to identify a plausible causative (micro)organism (virus, bacteria, parasite) for the excessive mortality; however some samples were positive for Clostridium perfringens. Sponsor hypothesizes that the deaths were due to lactational ileus (paresis of peristalsis), which albiglutide treatment may have exacerbated. No NOAEL for maternal toxicity was established in this study, since there were increased deaths at all doses.

Part 3 (repeat): As before, female CD-1 mice received daily subcutaneous injections of albiglutide (1, 5, or 50 mg/kg) beginning on Day 7 up to Day 21 PP. Operating under the hypothesis that the excessive deaths in lactating dams seen in the first study were due to lactational ileus, this study was repeated under conditions intended to improve the metabolic reserve of the dams during the lactation period. The primary changes were 1) the substitution of a high energy diet formulation in place of the standard rodent chow used in the first study and 2) culling of the litter size to 8 pups (equal numbers of ♂ & ♀ when possible) on Day 4 PP to reduce metabolic demand during lactation. This appeared to mitigate the high death rate seen in lactating dams in the previous study, consistent with Sponsor's hypothesis that the deaths in the earlier study were due to lactational ileus. There were nonetheless 1 LD and 1 HD F0 dam that died or was euthanized during the lactation period with signs consistent with lactation ileus. Additional signs of maternal toxicity, specifically a 7% BW loss following the first dose, were seen at HD. In the F1 pups, beginning Day 7 pp (1$^{st}$ day of dam dosing) d.d. ↓ in BW gain, such that by PND 21 all dose groups have s.s. lower weight than C: ↓3%, 5%, 7% in LD, MD & HD, respectively. During the post-weaning period there was no additional effect on BW gain; however, BW differences established during the pre-weaning period generally persisted. There were no effects on sexual maturation in ♂s, however ♀s had slight delays consistent with their lower bodyweight. The NOAEL for F0 maternal toxicity is considered to be < 1mg/kg/day on the basis of mortality at 1 mg/kg/day (0.6X clinical exposure based on AUC). The NOAEL for F0 reproductive function is considered to be 50 mg/kg/day (39X clinical exposure based on AUC), the highest dose tested. The NOAEL for postnatal development is < 1mg/kg/day (0.6X clinical exposure based on AUC) on the basis of slightly decreased pre-weaning body weight gain at all doses.

**DART Summary**: Albiglutide was without effect on male fertility at any tested dose up to 50 mg/kg/day (34X clinical exposure based on AUC). While Albiglutide appeared to slightly decrease the number of estrus cycles in females at 50 mg/kg/day (39X clinical exposure based on AUC), it had no effect on female fertility. Albiglutide treatment during organogenesis was not associated with malformations; however, it did cause an increase in the incidence of the skeletal developmental variation bent (wavy) ribs at 50 mg/kg/day (39X clinical exposure based on AUC). This same dose was associated with maternal toxicity (body weight loss) and embryo/fetal lethality due to postimplantation loss. The NOAEL for embryo/fetal toxicity was 5 mg/kg/day (2X clinical exposure based on AUC). Post-natal effects of Albiglutide treatment on the F1 mice depended somewhat on the window of treatment of the F0 dams (GD6 to GD17 vs. GD15 to LD10 vs. LD7 to LD21). For all treatment windows, maternal treatment with Albiglutide at 50



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mg/kg/day was associated with reduced maternal food consumption and body weight gain during the gestation and/or lactation periods and reduced F1 body weight gain during the lactation period. There was typically rebound weight gain in the F1 animals post-weaning. Slight delays in sexual maturation in both sexes of F1 pups whose dams were treated with 50 mg/kg/day albiglutide are likely a consequence of lower F1 body weights. Likewise for reductions in the maximum force generated in response to an auditory startle stimulus. There was no effect of F0 treatment on the reproductive success of the F1 generation; however, when F0 dams were treated with 50 mg/kg/day albiglutide from GD15 to LD10, there was s.s. reduction in the mean number of live F2 pups born/litter.

Notably, albiglutide treatment of lactating mice was associated with an unusually high incidence of maternal death. This effect did not however exhibit clear dose-dependency, and occurred at doses that were not associated with lethality in non-lactating mice. While no cause of death could be definitively established, Sponsor posited that, since the deaths are only occurring during lactation, they may be due to lactational ileus (paresis of peristalsis), a condition to which mice are known to be uniquely susceptible[4]. The known effect of GLP-1R agonism to decrease food consumption and slow GI transit time also provides a plausible explanation for why albiglutide could cause/exacerbate this condition. Reduction in metabolic demand duration lactation by culling litter size and increasing the caloric content of the feed largely ameliorated the adverse effect of albiglutide on survival of lactating mice, consistent with lactational ileus as the cause of the earlier deaths. The propensity of albiglutide to cause/exacerbate lactational ileus in mice is not considered to be clinically relevant, since the relative stress of lactation energy demands is much lower in humans and humans have relatively large energy reserves, compared to mice.

Approval Issues Arising from the Nonclinical Program

None.

There are however two class-related issues that bear further discussion *vis-à-vis* approval of albiglutide. The first of these is the potential issue of the role of GLP-1R agonists in promoting thyroid C-cell neoplasia, and the second is the role of GLP-1R agonists in promoting pancreatitis.

As discussed above in Section 8 (Carcinogenicity), other long-acting GLP-1R agonists have been clearly associated with hyperplastic/neoplastic changes in thyroid C-cells in rodents. The mechanism by which this effect is elicited, and its clinical relevance, have not been established. It has been proposed by some that this is a rodent-specific finding based on species differences in the density of expression of the GLP-1R on thyroid C-cells. While rodent specificity has not been definitively established, no hyperplastic/neoplastic changes have been observed in thyroid C-cells in non-rodents, and there is no clear signal for increased C-cell cancer risk in humans exposed to long-

[4] Kunstyr I. Paresis of peristalsis and ileus lead to death in lactating mice. Laboratory Animals, 1986;20:32-5.



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acting GLP-1R agonists. Unfortunately, because albiglutide induces a rapid, clearing immune response in rodents, it was not feasible to conduct typical 2-year rodent carcinogenicity assays with this compound. Furthermore, current guidance discourages the use of rodent-specific homologous proteins in lifetime bioassays. In lieu of such rodent studies, Sponsor conducted a 1-year study in monkeys in which the thyroid was subjected to intensive examination involving the enumeration of C-cells throughout the tissue by stereology. This study did not find any evidence of abnormal C-cell proliferation, consistent with the absence of C-cell effects seen in non-rodents with other long-acting GLP-1R agonists. However, these monkey data do not negate the C-cell proliferative effects identified in rodent carcinogenicity studies with other GLP-1R agonists, and it is judged that albiglutide likely carries the same degree of risk regarding C-cell hyperplasia as other approved long-acting GLP-1R agonists, and that it should carry comparable warnings and precautions as these other agents.

A possible signal for an increased incidence of pancreatitis has been reported postmarketing for incretin-based treatments. This category includes direct GLP-1R agonists, as well as DPP-IV inhibitors. There has been no signal suggesting an increased risk of pancreatitis in the nonclinical program with albiglutide (nor any other GLP-1R agonist). There was a signal in the 1-year monkey study for moderately increased pancreas weight in those animals treated with 50 mg/kg/week of albiglutide (75X clinical exposure based on AUC). Sponsor conducted a stereological examination of the entire pancreas from these animals (compared to control animals), enumerating the number of islet cells, ductal cells and acinar cells, as well as qualitatively assessing proliferative changes to these cell populations. The data are most consistent with a proportional increase in all cellular components of the pancreas, and do not support hyperplastic expansion of any specific cell population. In particular, there is no evidence of ductal cell or islet cell hyperplasia. Furthermore there were no abnormalities noted in the pancreases of these animals by standard histopathologic examination.

Also of note is a potential nonclinical signal identified with other long-acting GLP-1R agonists that suggests the possibility of advanced entry into puberty in male non-human primates. A meta-analysis conducted across multiple long-acting GLP-1R agonists has identified a signal suggesting an increase in testes size in peri-/pre-pubertal male monkeys. As there are data in the literature supporting a role for GLP-1R activity in the release of gonadotropin releasing hormone (GnRH) from the hypothalamus, it has been hypothesized that treatment with long-acting GLP-1R agonists may cause earlier entry into puberty. Consistent with this, female mice engineered to be defective for GLP-1R expression show delayed sexual maturation, and GLP-1R-deficient males have reduced weight of gonadal tissues. The regulatory response to these data has been to require Sponsor's that are developing long-acting GLP-1R agonists to conduct juvenile toxicity studies with an emphasis on measuring effects on sexual maturation, prior to exposure of pediatric subjects. GSK intends to conduct such a study to support proposed pediatric studies in children aged 10 to 18 years diagnosed with T2DM. Notably the nonclinical data reviewed herein do not show a detectable effect of albiglutide on the timing of entry into puberty in non-human primates. In particular, there is no clear signal



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indicating increased testes size in the pre-/peri-pubertal male monkeys exposed to albiglutide. There are however technical limitations to the studies that were conducted. In particular, it is notable that most of the male monkeys in the 52-week study were greater than 5 years old at time of assessment of testes weight, an age at which > 98% of male cynomolgus monkeys are expected to be fully sexually mature. The 26-week study is confounded by the presence of clearing ADA in some treated males. It is also notable in this context that rodent pre-/post-natal studies found a slight delay in sexual maturation, rather than early sexual maturation in the $F_1$ offspring of treated dams. This latter finding is considered to be a product of the retarded body weight development observed in the $F_1$ pups, since body weight is a strong independent variable in the timing of sexual maturation.

# 12 Appendix/Attachments



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**This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.**

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/s/

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RONALD L WANGE
07/25/2013
Pharm/Tox Recommendation for Approval

KAREN L DAVIS BRUNO
07/25/2013
concur with AP recommendation

# PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR NDA/BLA or Supplement

**NDA/BLA Number: 125431** **Applicant: GSK** **Stamp Date: 1/14/2013**

**Drug Name: Albiglutide** **NDA/BLA Type: BLA**

On **initial** overview of the NDA/BLA application for filing:

| | Content Parameter | Yes | No | Comment |
|---|---|---|---|---|
| 1 | Is the pharmacology/toxicology section organized in accord with current regulations and guidelines for format and content in a manner to allow substantive review to begin? | x | | |
| 2 | Is the pharmacology/toxicology section indexed and paginated in a manner allowing substantive review to begin? | x | | |
| 3 | Is the pharmacology/toxicology section legible so that substantive review can begin? | x | | |
| 4 | Are all required (*) and requested IND studies (in accord with 505 b1 and b2 including referenced literature) completed and submitted (carcinogenicity, mutagenicity, teratogenicity, effects on fertility, juvenile studies, acute and repeat dose adult animal studies, animal ADME studies, safety pharmacology, etc)? | x | | |
| 5 | If the formulation to be marketed is different from the formulation used in the toxicology studies, have studies by the appropriate route been conducted with appropriate formulations? (For other than the oral route, some studies may be by routes different from the clinical route intentionally and by desire of the FDA). | x | | |
| 6 | Does the route of administration used in the animal studies appear to be the same as the intended human exposure route? If not, has the applicant submitted a rationale to justify the alternative route? | x | | |
| 7 | Has the applicant submitted a statement(s) that all of the pivotal pharm/tox studies have been performed in accordance with the GLP regulations (21 CFR 58) or an explanation for any significant deviations? | x | | |
| 8 | Has the applicant submitted all special studies/data requested by the Division during pre-submission discussions? | x | | |

File name: 5_Pharmacology_Toxicology Filing Checklist for NDA_BLA or Supplement 010908

# PHARMACOLOGY/TOXICOLOGY FILING CHECKLIST FOR NDA/BLA or Supplement

| | Content Parameter | Yes | No | Comment |
|---|---|---|---|---|
| 9 | Are the proposed labeling sections relative to pharmacology/toxicology appropriate (including human dose multiples expressed in either mg/m2 or comparative serum/plasma levels) and in accordance with 201.57? | x | | |
| 10 | Have any impurity – etc. issues been addressed? (New toxicity studies may not be needed.) | x | | |
| 11 | Has the applicant addressed any abuse potential issues in the submission? | x | | |
| 12 | If this NDA/BLA is to support a Rx to OTC switch, have all relevant studies been submitted? | | | Not applicable. |

**IS THE PHARMACOLOGY/TOXICOLOGY SECTION OF THE APPLICATION FILEABLE? __Yes_____**

If the NDA/BLA is not fileable from the pharmacology/toxicology perspective, state the reasons and provide comments to be sent to the Applicant.

Please identify and list any potential review issues to be forwarded to the Applicant for the 74-day letter.

None.

Reviewing Pharmacologist Date

Team Leader/Supervisor Date

File name: 5_Pharmacology_Toxicology Filing Checklist for NDA_BLA or Supplement 010908

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/s/

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RONALD L WANGE
02/27/2013
BLA is fileable.

KAREN L DAVIS BRUNO
02/27/2013