

**CENTER FOR DRUG EVALUATION AND  
RESEARCH**

*APPLICATION NUMBER:*  
**22-192**

**PHARMACOLOGY REVIEW**

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

**PHARMACOLOGY/TOXICOLOGY REVIEW AND EVALUATION**

NDA NUMBER/ SERIAL NUMBER: 22-192 / N-0000

DATE RECEIVED BY CENTER: 9/27/2007

PRODUCT: Iloperidone

INTENDED CLINICAL POPULATION: Adults with schizophrenia

SPONSOR: Vanda Pharmaceuticals

DOCUMENTS REVIEWED:

REVIEW DIVISION: Division of Psychiatry Drug Products (HFD-130)

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## ***EXECUTIVE SUMMARY***

### **I. Recommendations**

- A. Recommendation on approvability: Approvable
- B. Recommendation for nonclinical studies: Adequate
- C. Recommendations on labeling: Changes recommended

### **II. Summary of nonclinical findings**

#### **A. Brief overview of nonclinical findings**

**Pharmacology:** Iloperidone has high affinity for serotonin 5-HT<sub>2A</sub>, adrenergic  $\alpha$ <sub>1</sub>, adrenergic  $\alpha$ <sub>2</sub>, D<sub>2</sub>, D<sub>3</sub>, and 5-HT<sub>1A</sub> receptors in humans, and acts as an antagonist at selected dopaminergic, serotonergic, and noradrenergic receptor subtypes. Affinity was highest for 5-HT<sub>2</sub> and adrenergic  $\alpha$ <sub>1</sub> receptors, and lower for dopamine D<sub>2</sub>, which is a profile of an atypical antipsychotic. Iloperidone metabolites P88 and P89 have a profile similar to that of iloperidone in receptor-binding studies, with potential to exert CNS effects mediated by dopaminergic, serotonergic, and noradrenergic antagonism. P95 exhibits a similar affinity to iloperidone for human 5-HT<sub>2A</sub> and adrenergic receptor subtypes, while exhibiting a substantially lower affinity for D<sub>1</sub>, D<sub>2</sub>, and D<sub>3</sub> receptor subtypes compared with iloperidone. P95 is less likely to exert CNS effects since, as shown by whole-body autoradiography, it apparently does not cross the blood-brain barrier. The high affinities of iloperidone and its metabolites for  $\alpha$ <sub>1</sub>- adrenergic receptors in peripheral vascular tissues indicate that iloperidone and its metabolites P88, P89, and P95 are likely to exert cardiovascular effects, such as postural hypotension.

In vitro evaluation of iloperidone effects in isolated dog Purkinje fibers and in mammalian cells expressing the cloned hERG showed that iloperidone has the capacity to prolong action potential duration and to block hERG currents; this indicates that iloperidone has the capacity to prolong QT<sub>c</sub> interval. Iloperidone metabolite P88, but not P95, also exhibited this potential. In hemodynamic evaluations conducted in rats and dogs, iloperidone was found to dose-dependently decrease blood pressure and to induce transient increases in heart rate; however, cardiac output and ECG parameters were not affected. Neither iloperidone nor its metabolite P95 was associated with any adverse respiratory effects as evaluated in rats.

**Pharmacokinetics:** Iloperidone was rapidly absorbed in all animal species tested following oral and i.v. administrations, but its bioavailability was very low due to a significant first-pass effect. Oral bioavailability was <1% in rat, 5% in mouse, 19% in both rabbit and dog, and approximately 36% in humans. The absorption profiles of metabolites P88 and P95 were similar to the parent compound; their absorption was rapid after either oral or i.v. administration. At equal oral doses, bioavailability of P95 (18%) was significantly higher than P88 (5%) in mice.

Iloperidone plasma exposure (C<sub>max</sub> and AUC) levels generally increased dose-proportionally in the tested animal species, except for the rat in which exposure increased over-proportionally possibly due to inhibitory activity of iloperidone to CYP enzymes.

Gender differences in exposure were present in the rat, the mean AUC in female rats being significantly greater than that in males.

Distribution of iloperidone and its metabolites after oral administration was rapid; the highest drug concentrations were observed in the liver, kidney, gastrointestinal system, and secretory glandular tissues; placental transfer was limited; and drug concentration in the brain was very low. P95 metabolite did not pass the blood-brain barrier in the rat (whole-body autoradiography). After oral administration to lactating rats, iloperidone was excreted in milk; C<sub>max</sub> was attained 4 hours post dosing when iloperidone concentration was approximately 10 times higher in milk than in plasma.

Iloperidone metabolic profiles show differences across species. The most abundant metabolites in humans (P95 and P88) are found in the species used in toxicology studies. However, in rodents, P95 and P88 are only minor circulating metabolites, in contrast to humans. Plasma exposure to the main active metabolite P88 in rodents and dogs is lower than that of iloperidone, while in humans, P88 exposure is greater than that of the parent compound. For P95, the differences between humans and animals are even greater than for P88. Results of pharmacology and pharmacokinetic studies that have bearing on the potential toxicological characteristics of metabolite P95, include the following:

- While P95 is the predominant circulating metabolite of iloperidone in humans, comprising 25% to 54% of its total metabolism, in rodents it represents only 3.9% to 5.7% of the total measurable exposure to iloperidone and its metabolites.
- Although P95 did not appear to cross the blood-brain barrier as assessed in the whole-body autoradiography study in rats, in general toxicity studies in rodents and dogs with direct oral administration of P95, it induced CNS clinical signs similar to those induced by iloperidone, which suggests that the blood-brain barrier is not impenetrable to P95.
- P95 is rapidly eliminated in rodents; the half-life of P95 is 45 min in mice, 40 min in Sprague-Dawley rats and 100 min in Wistar rats, as compared to a half-life of 23-26 hours for P95 in humans.

In vitro metabolic studies showed that iloperidone has stronger inhibitory activity to CYP2D6 and CYP3A4/3A5 compared with either P88 or P95; neither iloperidone nor its metabolites had potential to induce cytochrome P450 enzymes.

Excretion profiles of iloperidone, P85 and P99 were similar. They are mainly eliminated through the feces, in contrast to humans in which urinary excretion is the major elimination pathway.

Toxicology: Repeat-dose studies of general toxicity and corresponding toxicokinetic parameters were conducted with iloperidone in mice, rats, rabbits, and dogs. Additionally, toxicology studies were performed in rats and mice with the predominant circulating metabolite of iloperidone in humans, P95, to better characterize its safety and toxicity profiles in view of the lower exposure to this metabolite following iloperidone administration in animal species vs. humans.

General toxicology: Among all the repeat-dose general toxicology studies on iloperidone and its P95 metabolite, pivotal studies of the longest duration and therefore most relevant to safety evaluation, are the 6-month rat study and the 12-month dog study conducted with iloperidone, and the 6-month rat study conducted with P95 metabolite. These studies are the subject of the present review.

Iloperidone 6-month oral administration to rats (Sprague-Dawley) at doses of 0, 12, 24, and 48 mg/kg/d induced dose-related clinical signs indicative of CNS depression (ptosis, decreased motor activity, relaxation of the scrotum, anus, vaginal opening) and decrease of mean body weight at all dose levels; hematological changes (lower total leukocyte and lymphocyte counts at LD, MD and HD and lower platelet counts at MD and HD); dose-related decrease in serum triglycerides and glucose levels in females at all doses and in MD and HD males. Prolactin was not determined. Increased incidence and severity of vacuolization of glandular epithelium in the mammary glands of males and females was seen in all dose groups, mammary hypertrophy/hyperplasia in females at MD and HD, testicular degeneration and atrophy at MD and HD, and fatty infiltration in bone marrow sections in HD group. During the 5-week recovery period, an incomplete reversibility was seen for decreased body weight, hematology and mammary glandular epithelium changes. The MTD was 12 mg/kg/d, based on a marked body weight decrease (18-22% vs. control) at the next higher dose tested (24 mg/kg/d). An NOAEL was not reached in this study, as the lowest tested dose (12 mg/kg/d) induced a decrease in body and organ weights, hematological and clinical chemistry changes, and histopathology changes in the mammary glands of males and females. This dose is about 5 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis.

Iloperidone 1-year oral administration to beagle dogs at 6, 12, and 24 mg/kg/d induced drug-related clinical signs at all dosages (decreased spontaneous activity, tremors, bizarre behaviors, labored breathing, ptosis, slow response times and/or lack of pupillary reflex); the mid- and high-dose induced ataxia, loss of righting and toe pinch reflex (in single animals), emaciation. Body weight decreases of 7.3% and 9.2% vs. control were registered over the treatment period at LD and HD, respectively. Hematology and clinical chemistry changes were induced dose-dependently at MD and HD, i.e., decreases in mean erythrocyte count and in hemoglobin and hematocrit levels in males and females; lower cholesterol and triglyceride levels in females, and increase in alanin aminotransferase in HD males. No abnormalities were found in any dose group on ECG and auditory examination. Higher mean absolute and relative liver weights and hepatocellular hypertrophy resulting from proliferation of the endoplasmic reticulum were found in males in the HD group, probably secondary to liver enzyme induction. The MTD was 6 mg/kg/d in view of severe clinical signs and emaciation induced at and above the next higher dose of 12 mg/kg/d. NOAEL was not reached in this study as the lowest tested dose (6 mg/kg/d) induced decreased body weight and neurological clinical signs. This dose is 8 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis.

Iloperidone metabolite P95 six-month administration to rats (Wistar) at oral doses of 50 and 500 mg/kg/day (yielding P95 plasma exposures of about 2 to 3x and 150 to 400x, respectively, the human P95 plasma exposure at iloperidone MRHD of 24 mg/d), induced dose-dependent CNS clinical signs at both dose levels throughout the entire treatment period, similar to those induced by iloperidone (ptosis, decreased motor activity, relaxation of the scrotum, anus, vaginal opening) that are attributable to pharmacological effect. Body weight and weight gain decreases were induced at HD only. There were no drug-related abnormal findings in hematology, clinical chemistry (including prolactin plasma levels), or urine analysis. Functional and morphologic reproductive system changes were induced in both genders. In females, dose-dependent cycle prolongation occurred at both LD and HD, consistent with the finding of vaginal

epithelium mucification and decreased uterine weight in the treated groups. In males, atrophy of testicular seminiferous tubule epithelium (in 2 animals) and an increased incidence of mixed cell inflammation of prostate gland with associated degenerative changes were found at HD. In both genders, increased cellular proliferation in the mammary gland (alveolar hyperplasia, increased secretion and dilatation of mammary ducts) occurred with dose-related severity at LD and HD, non-reversible after the recovery period. Drug-related proliferative histopathology changes, demonstrable by routine histology and/or immunohistochemical method (BrdU labeling) were induced in endocrine glands (pituitary and adrenals in males, thyroid in females, and pancreas in both genders), mammary gland (both genders) and reproductive organs (ovary, uterus, testes, prostate). Statistically significant, treatment-related increase in cell proliferation (increased proportion of cells in S phase of the cell cycle, as assessed by BrdU labeling) was found in pituitary (LDM and HDM), mammary gland (duct and alveoli) in both genders (LDM, HDM, HDF), and the endocrine pancreas in both genders (HDM, HDF). Most of these histopathology deviations (with the exception of the adrenal, testicular and secondary sex organ pathology in males) were induced in a dose-dependent manner at both tested dose levels. An NOAEL was not reached in either male or female rats since pathomorphological proliferative changes in multiple organs/tissues were present at the lowest tested dose of 50 mg/kg/day, corresponding to plasma exposure (AUC 0-24) approximately 2 to 3x the human P95 plasma exposure at MRHD of 24 mg iloperidone/day.

Genetic toxicology: Iloperidone was clastogenic in one in vitro test (chromosomal aberration assay in Chinese Hamster Ovary (CHO) cells). It is likely that the positive results obtained in the chromosomal aberration assay in vitro are of little biological relevance, having in mind the negative results obtained in the in vivo micronucleus assays in rat hepatocytes and mouse bone marrow. Iloperidone metabolite P95 was negative for potential genotoxicity in a battery of 3 tests: an Ames, a chromosomal aberration test in CHO cells, and a bone marrow micronucleus test in rats. For iloperidone genotoxic and potentially genotoxic impurities

the acceptance criteria are set at the level of \_\_\_\_\_ each, so that the overall daily exposure from the sum of these \_\_\_\_\_ impurities is \_\_\_\_\_ g/day.

Carcinogenicity: Two-year carcinogenicity studies of iloperidone were conducted in mice and rats of both genders.

Iloperidone administration to \_\_\_\_\_ D-1 (ICR) BR mice at oral doses of 2.5, 5, and 10 mg/kg/d (causing an increased mortality in males at HD and in females at all dose levels), did not exert carcinogenic effect in males. In females, the incidence of malignant mammary tumors was significantly increased above the concurrent and historical control range in the low dose group only. On an mg/m<sup>2</sup> basis, there is no safety margin between the low dose employed in the study (2.5 mg/kg/day) and the maximal recommended dose in humans (24 mg/day). However, mammary tumor incidences were not increased in the mid- and high-dose groups, although the duration of treatment was the same in the mid-dose and low dose groups. It is not clear why similar increases in mammary tumor incidences were not seen at the higher doses employed in this study. Drugs which elevate plasma prolactin typically cause mammary tumors in rodents.

Iloperidone administration to \_\_\_\_\_ D(SD)BR rats at oral doses of 4, 8, and 16 mg/kg/d for 24 months (inducing a dose-related, significant decrease in mean body weights of

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over 10% in all treated groups), did not exert carcinogenic effect in male rats. In females, the combined incidences for pancreatic islet cell adenomas and islet cell carcinomas were increased at HD (2, 2, 0, 3, and 7 for the two controls, LD, MD and HD, respectively). The incidence value at HD was within historical control range for this species and strain; the dose-response trend analysis showed a p-value of 0.0051 that approached but did not reach the level of statistical significance required for common tumors ( $\alpha=0.005$ ). Having in mind that the incidences of pancreatic islet cell tumors in this study were within the reported historical control range for this species and strain and that there was no other evidence indicating a treatment-related effect (such as multiplicity of tumors, increased incidence of pre-neoplastic findings), it is concluded there was no carcinogenic effect in the female rats attributable to the test article.

Reproductive and developmental toxicity was assessed in a Fertility (Segment I) study in rats; Embryofetal development (Segment II) study in rats and rabbits; and in a Pre- and postnatal development (Segment III) study in rats.

Segment I rat fertility study: Iloperidone oral administration at doses of 0, 4, 12, 36 mg/kg/day to Sprague Dawley male and female rats for a period starting 10 weeks prior to mating (males) or 2 weeks prior to mating (females) and continuing through mating, gestation and lactation, resulted in the following drug-related effects: pharmacological clinical signs at all doses, significant decreases in body weight of both males and females at MD and HD, female estrous cycle disturbances (all doses, dose-dependently) and reduction in male reproductive organs' weight (prostate weight decreased in all dosed groups; testis and epididymis weights decreased at HD), lower fertility indices (72% and 88% at HD and MD, respectively, vs. 100% in control), lower pregnancy rates at MD and HD groups (86%, and 60%, respectively, vs. 100% in control), reduction of corpora lutea count at HD in comparison to control; increased duration of pregnancy at MD and HD; increased stillbirth rates and neonatal deaths at MD and HD. At HD, embryofetal growth was retarded, and visceral variation rates (dilatation of lateral and third brain ventricles, dilatation of heart ventricles) were increased, but no external malformations were observed in the treated groups. There were no differences in developmental landmarks or in neurobehavioral development of the surviving F1 pups (however, insufficient number of HD litters were available for growth and behavioral evaluations because of the low pregnancy rate and neonatal deaths). Reproductive performance of F1 generation was apparently not affected. The NOAEL was 4 mg/kg/day (1.6 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis). Although this dose induced estrous cycle disturbances and a decrease in prostate weight, it did not affect parental fertility or the prenatal and postnatal survival, development and reproductive capacity of the progeny.

Segment II Prenatal developmental toxicity studies in rats:

Iloperidone administration to pregnant Wistar rats at oral doses of 0, 4, 16, and 64 mg/kg/day during the period of major organogenesis (Gestation Days 7 through 18) induced developmental toxicity (expressed as embryofetal lethality, retarded intrauterine development and minor skeletal abnormalities) at oral doses above 16 mg/kg/day. Signs of maternal toxicity (reduced weight and weight gain, reduced placental weight) were present at and above 16 mg/kg/day. The NOAEL for developmental toxicity was 16 mg/kg/day (6 times the human dose at MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).

The predominant circulating iloperidone metabolite in humans (P95) administered to pregnant rats at oral doses of 20, 80 and 200 mg/kg/day (Gestation Days 7 through 17), produced dose-dependent maternal pharmacological effect (signs of sedation) at all dose levels, but no maternal toxicity. Maternal plasma exposure (AUC) at the high dose was approximately 4 times the mean human plasma AUC of metabolite P95 when the parent compound (iloperidone) was administered at the MRHD of 24 mg/day. The treatment did not induce embryo/fetal mortality or congenital malformations but produced a dose-dependent increase in the incidence of retarded skeletal ossification vs. the concurrent control at all tested dose levels, ranging from 8% (LD) to 14% (HD). These values, however, were within the historical control range for the tested species and strain.

Segment II Prenatal developmental toxicity study in rabbits:

Iloperidone administration at oral (gavage) doses of 0, 4, 10 and 25 mg/kg/day to pregnant rabbits from gestation day 6 through 18 caused maternal mortality (1/15) and decreased maternal body weight at the HD and dose-dependent drug-related clinical signs (sedation) at all dose levels. Maternal food intake was reduced at MD and HD. The high dose induced increase in embryo/fetal intrauterine lethality and a decrease in fetal viability at term. No embryo/fetal toxicity or teratogenicity were observed at LD and MD. Based on these results, the NOAEL for embryo/fetal toxicity is 10 mg/kg/day (8x the human dose at MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).

Segment III Prenatal and postnatal developmental toxicity study in rats

Iloperidone oral administration to pregnant CD rats from gestation day 17 through weaning (postnatal day 21) at doses of 4, 16 and 48/36 mg/kg/day, caused maternal toxicity statistically significant at HD and MD (maternal mortality at HD and dose-dependent decrease in maternal body weight at HD, MD and LD), significantly prolonged gestation and parturition, high perinatal- and postnatal mortality (stillbirths and neonatal deaths) in F1 generation at HD and MD, and some increase in stillbirth rate at LD (mean stillbirth rate per litter 0.6 vs. 0.04 in control). The growth of the surviving F1 offspring was impaired at MD and HD, as demonstrated by the reduced pup weight at birth and weight gain through weaning. However, there was no apparent adverse effect on F1 development, including behavior, sexual maturation and reproductive capacity, at any of the administered dose levels. The NOAEL was 4 mg/kg/day (1.6 times the human dose at MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).

**B. Pharmacologic activity**

The pharmacological profile of iloperidone is consistent with that of an atypical antipsychotic with a reduced potential for extrapyramidal side effects and therapeutic potential with regard to positive, negative, and social withdrawal symptoms of schizophrenia. Iloperidone has the potential to induce hypotensive effects and to prolong QTc interval duration

**C. Nonclinical safety issues relevant to clinical use**

- The high affinity for  $\alpha$ 1- adrenergic receptors in peripheral vascular tissues, indicate that iloperidone and its metabolites P88, P89, and P95 are likely to exert cardiovascular effects, such as postural hypotension.
- Iloperidone prolongs action potential duration and block hERG currents in vitro; indicating a capacity to prolong QTc interval. In vivo, iloperidone dose-

dependently decreases blood pressure and induces transient increases in heart rate in rats and dogs; however, ECG parameters were not affected.

- Iloperidone has the potential to inhibit CYP2D6 and CYP3A4/5 at the recommended therapeutic dose in humans (12 mg BID); its metabolites P88 and P95 have a weaker inhibitory activity on CYP2D6 and CYP3A4/5.
- Chronic oral administration of iloperidone to rats (6 months) and dogs (1 year) and of iloperidone P95 metabolite to rats (6 months) induced general toxicity in all tested species. An NOAEL was not reached in either of these studies. The lowest tested dose of iloperidone in the 6-month rat study (12 mg/kg/d, about 5 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis) induced a decrease in body and organ weights, hematological and clinical chemistry changes, and histopathology changes in the mammary glands of males and females. The lowest tested dose of iloperidone in the 1-year dog study (6 mg/kg/d, 8 times the human dose at MRHD on an mg/m<sup>2</sup> basis) induced decreased body weight and neurological clinical signs. The lowest tested dose of P95 in the 6-month rat study (50 mg/kg/day, corresponding to plasma exposure (AUC 0-24) approximately 2 to 3x the human exposure at MRHD) induced proliferative pathomorphological changes in multiple organs/tissues, i.e., endocrine glands (pituitary, thyroid, and pancreas), mammary gland (both genders) and ovary.
- Iloperidone was clastogenic in one in vitro test (chromosomal aberration assay in Chinese Hamster Ovary (CHO) cells) but was not clastogenic in the in vivo micronucleus assays in rat hepatocytes and mouse bone marrow. The positive results obtained in chromosomal aberration assay in CHO cells in vitro are of little biological relevance, having in mind the negative results obtained in vivo.
- Iloperidone genotoxic and potentially genotoxic impurities \_\_\_\_\_ do not constitute a genetic toxicity risk for humans since the acceptance criteria for each of these impurities are set at the level of \_\_\_\_\_ ppm, so that the overall daily exposure from the sum of these -- impurities is \_\_\_\_\_ µg/day.
- Iloperidone administration for 2 years was not carcinogenic to rats; in mice (female) administered oral doses of 2.5, 5 and 10 mg/kg/day for 2 years the incidence of malignant mammary tumors was significantly increased above the concurrent and historical control range in the low dose group only. On an mg/m<sup>2</sup> basis, there is no safety margin between the low dose employed in the study (2.5 mg/kg/day) and the maximal recommended dose in humans (24 mg/day). However, mammary tumor incidences were not increased in the mid- and high-dose groups, although the duration of treatment was the same. It is not clear why similar increases in mammary tumor incidences were not seen at higher doses.
- In view of the proliferative effects seen with iloperidone metabolite P95 in the 6-month rat study, the Division required a 2-year carcinogenicity study with P95 in the rat which is ongoing (CAC meeting of March 25, 2008).
- Iloperidone induces decreased fertility, prolonged gestation, increased prenatal and neonatal mortality, and retarded growth of the progeny upon oral administration to male and female rats for a period starting 10 weeks prior to mating (males) or 2 weeks prior to mating (females) and continuing through

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gestation, parturition and lactation. The NOAEL is 4 mg/kg/day (1.6 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis).

- Iloperidone oral administration to pregnant rats and rabbits during the period of major organogenesis induces developmental toxicity (embryofetal lethality in both species, retarded intrauterine development and minor skeletal abnormalities in the rat) at doses that are maternally toxic. The NOAEL for developmental toxicity is 16 mg/kg/day in rats and 10 mg/kg/day in rabbits (6- and 8 times, respectively, the human dose at MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).
- Iloperidone perinatal and postnatal administration to rats (Gestation day 17 through postnatal day 21) produced, at maternally toxic doses, prolonged gestation and parturition, increased incidence of stillbirths, neonatal mortality, and retarded growth of progeny up to weaning, but did not affect neurobehavioral and reproductive development of the surviving pups. The NOAEL is 4 mg/kg/day (1.6 times the human dose at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis).

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## 2.6 PHARMACOLOGY/TOXICOLOGY REVIEW

### 2.6.1 INTRODUCTION AND DRUG HISTORY

NDA number: 22-192

Review number: 1

Sequence number/date/type of submission: N-0000

Information to sponsor: No (x)

Sponsor and/or agent: Vanda Pharmaceuticals

Manufacturer for drug substance: Vanda Pharmaceuticals

Reviewer name: Sonia Tabacova

Division name: Psychiatry Drug Products

HFD #: 130

Review completion date: June 2008

#### Drug:

Trade name: None provided

Generic name: Iloperidone

Code name: ILO522 (Novartis); ILO522-NXA (Novartis); VYV-683 (Vanda)

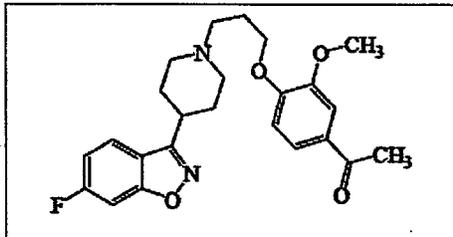
Chemical name: 1-[4-[3-[4-(6-Fluorobenzo[d]isoxazol-3-yl)-1-piperidinyl]propoxy]-3-methoxyphenyl]ethanone

Chemical Abstract Service (CAS) Number: 133454-47-4

Mole file number:

Molecular formula/molecular weight: C<sub>24</sub>H<sub>27</sub>N<sub>2</sub>O<sub>4</sub>F / 426.5

Structure:



Relevant INDs/NDAs/DMFs: IND 36827

Drug class: Antipsychotic

Intended clinical population: Adults with schizophrenia

Clinical formulation: Tablets

Route of administration: oral

Disclaimer: Tabular and graphical information is from sponsor's submission unless stated otherwise.

**Studies reviewed within this submission:** All submitted studies, except for the non-pivotal iloperidone repeat-dose general toxicology studies

**Studies not reviewed within this submission:** Non-pivotal iloperidone repeat-dose general toxicology studies

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## 2.6.2 PHARMACOLOGY

### 2.6.2.1 Brief summary

Iloperidone has an in vitro and/or ex vivo binding profile consistent with an atypical antipsychotic, displaying affinity for human dopamine (D), serotonergic (5-HT),  $\alpha$ -noradrenergic, and sigma receptors, and no affinity for glycine-binding site, *N*-methyl-D aspartate (NMDA) receptor channel, or muscarinic receptors. Affinity of iloperidone was found to be highest for human 5-HT<sub>1A</sub>, 5-HT<sub>2A</sub>, D<sub>2</sub>, D<sub>3</sub>, adrenergic  $\alpha$ <sub>1</sub> and  $\alpha$ <sub>2</sub> receptors, and lower for D<sub>1</sub>, D<sub>5</sub>, and other serotonergic receptors. Iloperidone has no agonist activity at any receptors evaluated (rat or human), but was found to have significant in vitro and in vivo antagonist activity at rat and human dopaminergic, serotonergic, and noradrenergic receptors as evaluated in various in vitro and in vivo functional assays. Both iloperidone and clozapine had potent inhibitory activities on 5-HT<sub>2</sub> receptors and weak effects on D<sub>2</sub> receptors as shown in an ex vivo study in rats. The high 5-HT<sub>2</sub>/D<sub>2</sub> receptor binding ratio of iloperidone suggests that this compound has the potential to act as an atypical antipsychotic agent with a relatively low potential to produce extrapyramidal side effects. Preferential affinity for  $\alpha$ <sub>1</sub> over  $\alpha$ <sub>2</sub> receptors suggests a potential for iloperidone to cause orthostatic hypotension.

Behavioral assays conducted in mice, rats and monkeys compared the in vivo pharmacological activities of iloperidone with both typical (thioridazine and/or haloperidol) and atypical (clozapine) antipsychotic agents. The results indicated that iloperidone has potential for atypical antipsychotic action with reduced capacity for inducing extrapyramidal (EPS) symptoms, potential anxiolytic properties at doses similar to its antipsychotic properties and a potential to increase social interaction. Overall, the in vivo pharmacodynamic profile of iloperidone indicated properties consistent with an atypical antipsychotic with reduced EPS liability.

The major metabolites of iloperidone in humans are P88, P89 and P95. It is important to note that P88 and P89 cross the blood-brain barrier, while P95 does not do so detectably. Therefore, P95 is less likely to exert pharmacological effects on the central nervous system and was primarily evaluated for the potential to exert peripheral effects. Pharmacodynamic studies have been performed with P88, P89 and P95 metabolites directly and their receptor-binding profiles were separately characterized. P89 was found to bind with high affinity to D<sub>2</sub> and 5-HT<sub>2</sub> receptors, whereas P88 was approximately 30-fold and 10-fold weaker at these sites, respectively. Both P88 and P89 also exhibited affinity for 5-HT<sub>1A</sub> receptors and  $\alpha$ <sub>2</sub>-noradrenergic receptors. P95 exhibited similar affinity for the human 5-HT<sub>2A</sub> receptor compared with iloperidone, and also exhibited higher or comparable affinity for each adrenergic receptor subtype tested as compared with iloperidone. P95 exhibited a substantially lower affinity for the D<sub>1</sub>, D<sub>2S</sub>, D<sub>2L</sub>, and D<sub>3</sub> receptor subtypes compared with iloperidone. Neither iloperidone nor P95 showed appreciable affinity for histamine H<sub>1</sub> receptors.

The receptor affinity profiles of P95 and P88 metabolites were further evaluated for broad-spectrum receptor affinity for human, rat and guinea pig receptors using radiolabeled ligand-binding techniques. The results showed that the P88 metabolite had moderate to strong affinity at the adrenergic  $\alpha$ <sub>1</sub>,  $\alpha$ <sub>2B</sub>,  $\alpha$ <sub>2C</sub>, dopamine D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub>, D<sub>4</sub>,

histamine H<sub>1</sub>, 5-HT<sub>1B</sub>, and 5-HT<sub>2C</sub> sites. It had relatively weak affinity at the adrenergic  $\alpha_{2B}$ , 5-HT<sub>1A</sub>, and 5-HT<sub>6</sub> sites. In comparison, the P95 metabolite showed weak to moderate affinity at the adrenergic  $\alpha_1$ ,  $\alpha_{2A}$ ,  $\alpha_{2B}$ ,  $\alpha_{2C}$ , dopamine D<sub>1</sub>, D<sub>3</sub>, D<sub>4</sub>, and 5-HT<sub>6</sub> sites. Compared with the receptor profile of iloperidone, these metabolites were considered to have much weaker affinity at the various receptors. In a separate study, both metabolites were found to have a high affinity for 5-HT<sub>2A</sub>, as iloperidone.

Functional assays conducted with iloperidone metabolites P88 and P89 indicated that both compounds exhibit dopamine receptor antagonist properties. Additional behavioral assays conducted with iloperidone metabolites P88 and P89 in mice, rats, and monkeys indicated that these metabolites possess activity consistent with a dopamine antagonist profile similar to iloperidone. However, in contrast to the parent compound, neither metabolite appeared to possess anxiolytic activity, as measured in behavioral assays.

In a series of safety pharmacology studies, the cardiovascular safety of iloperidone was evaluated *in vitro* and *in vivo* in rats and dogs. In dog Purkinje fibers paced at stimulation frequencies of 0.5 and 1 Hz, iloperidone and P88 at a concentration of 0.1  $\mu\text{M}$  and above induced a prolongation in action potential duration. The highest concentration of 10  $\mu\text{M}$  induced a reduction in the maximum rate of depolarization at 3 Hz, indicating a frequency-dependent interaction with cardiac sodium channels. Purkinje fibers exposure to P95 at concentration of 0.01, 0.1, 1 and 10  $\mu\text{M}$  induced a prolongation in action potential duration at 10  $\mu\text{M}$  but it did not reach the level of statistical significance; no prolongation was seen at the concentration below or at 1  $\mu\text{M}$ . These data indicate that iloperidone and P88 have the potential to prolong the QT interval at free plasma concentrations of 0.1  $\mu\text{M}$  and above, while P95 has the potential to prolong the QT interval at free plasma concentrations of over 10  $\mu\text{M}$ . This extent of exposure to P95 is unlikely, as administration of iloperidone doses of 24 mg given once-a-day in human patients yielded maximal P95 steady-state plasma levels of 55.5 ng/mL, or 0.13  $\mu\text{M}$ . *In vitro* effects of iloperidone and its metabolites P95 and P88 in comparison with risperidone and ziprasidone on the cardiac ion channel hERG showed that all test articles produced rapid, reversible blockade of hERG currents. The block potency rank order was iloperidone > ziprasidone  $\approx$  P88 > risperidone > P95. These data suggest that P95 is unlikely to contribute to the QT prolongation potential of iloperidone.

*In vivo*, iloperidone was found to have hypotensive and vasodilatory effects similar to those of clozapine in normotensive and hypertensive rats and in conscious and anesthetized dogs. The hypotensive activity of iloperidone was also supported by its preferential affinity for  $\alpha_1$ - over  $\alpha_2$  adrenergic receptors *in vitro*. Except for a transient increase in heart rate observed in some studies, no other notable hemodynamic effects (e.g., cardiac output changes or ECG findings) were noted in rats or dogs. Similar to iloperidone, metabolites P88, P89, and P95 were also found to exert hemodynamic effects, including decreasing blood pressure.

One single-dose study was conducted to evaluate the respiratory safety of orally administered iloperidone and its metabolite P95 in albino rats. The results indicate no adverse respiratory effects of iloperidone or P95.

No pharmacodynamic drug interaction study of iloperidone or its metabolites was conducted in any animal model. Three *in vitro* studies were performed to investigate induction and inhibition activities of iloperidone and its metabolites on human

cytochrome P450 system. Following the incubations with human hepatocytes, neither iloperidone, nor P95 or P88 was found to induce CYP enzyme activity. The potential of iloperidone and its metabolites, P95 and P88, to inhibit human CYP enzymes was assessed using pooled human liver microsomes and several probe substrates, metabolism of which is known to be CYP isoform-selective. Iloperidone was found to directly inhibit CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5; P88 metabolite directly inhibited CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5; P95 had little or no potential to cause no direct inhibition of CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5.

### 2.6.2.2 Primary pharmacodynamics

Iloperidone is a piperidinyl-benzisoxazole derivative, structurally related to risperidone and developed for the treatment of schizophrenia. The pharmacodynamic profile of iloperidone was evaluated through assessment of in vitro receptor binding and a number of in vitro and in vivo functional assays. For comparison, the clinically active antipsychotic agents haloperidol, risperidone, and clozapine were included in selected studies.

#### In vitro/Ex vivo Studies

##### Receptor affinity profile

The affinities of iloperidone for dopaminergic, serotonergic, noradrenergic and selected other receptor types of the central nervous system relevant to the treatment of schizophrenia were evaluated in rat, guinea pig, and bovine tissues, as well as in cell lines stably expressing the relevant human receptor subtypes using in vitro radioligand-binding assays. A summary of the initial evaluation of the in vitro binding affinity of iloperidone and selected comparators for cloned rat and human dopamine and serotonin receptor subtypes is presented in the following sponsor's table.

Summary of binding affinities of iloperidone and selected antipsychotic agents for various rat and human dopamine and 5-HT receptors

Receptor subtype	Iloperidone	Risperidone	Clozapine	Olanzapine	Quetiapine	Haloperidol
Rat D <sub>1</sub>	546 ± 80	550 ± 70	690 ± 80	100 ± 5	2610 ± 20	520 ± 100
Rat D <sub>2</sub>	54 ± 8	20 ± 1	790 ± 100	52.3 ± 31.7	ND	13 ± 2
Rat 5-HT <sub>1A</sub>	168 ± 20	570 ± 100	640 ± 100	4546 <sup>a</sup>	ND	>2000
Rat 5-HT <sub>2</sub>	3.1 ± 2	1.4 ± 0.9	61 ± 40	13.3 ± 1.4	185 <sup>a</sup>	45 ± 9
Rat 5-HT <sub>6</sub>	42.7 ± 35.4	1122 ± 514	7.1 ± 2	27.5 ± 18.9	1297 ± 943	3683 ± 59
Rat 5-HT <sub>7</sub>	21.6 ± 10.5	0.93 ± 0.67	19.1 ± 4.7	152.2 ± 32.6	133.6 ± 47.4	241.2 ± 143.7
Human D <sub>1</sub>	216 ± 34	523 ± 68	196 ± 28	35 <sup>a</sup>	1277 ± 334	82 ± 7
Human D <sub>2short</sub> <sup>b</sup>	13.3 ± 3.5	3.3 ± 0.3	168 ± 38	30.3 ± 5.7	779 ± 104	2.1 ± 1.2
Human D <sub>2long</sub> <sup>b</sup>	6.3 ± 1.5	2.7 ± 0.4	291 ± 93	37.7 ± 4.1	706 ± 101	2.3 ± 1.3
Human D <sub>3</sub>	7.1 ± 4.1	14.1 ± 4.8	473 ± 490	49 ± 28	839 ± 256	1.9 ± 0.3
Human D <sub>4</sub>	25 ± 0.3	9 ± 1.9	54.5 ± 7.5	29.5 ± 2.5	1057 ± 1	3 ± 0.06
Human D <sub>5</sub>	319 ± 131	563 ± 127	281 ± 64	74 ± 13	1513 ± 407	178 ± 9
Human 5-HT <sub>2A</sub>	5.6 ± 0.3	1.1 ± 0.2	23.2 ± 2.3	24.2 ± 5.7	636 ± 34.5	186 ± 79
Human 5-HT <sub>2C</sub>	42.8 ± 1.6	12 ± 0.2	10.7 ± 0.3	6.4 ± 0.01	1184 ± 266	3949 ± 67

Ki = inhibitory constant; S.E. = standard error; ND = not done.

Based on percent inhibition of radioligand binding, iloperidone exhibited high affinity for human dopamine D<sub>2short</sub> and D<sub>2long</sub> and D<sub>3</sub> and for human serotonin 5-HT<sub>2A</sub> receptors, a characteristic profile of an atypical antipsychotic. Iloperidone also exhibited affinity for dopamine D<sub>4</sub>, and for serotonin 5-HT<sub>2C</sub>, 5-HT<sub>6</sub>, and 5-HT<sub>7</sub> receptors and a relatively lower affinity for dopamine D<sub>1</sub> and D<sub>5</sub> receptors.

The receptor affinity profile of iloperidone at a wider range of rat and human neurotransmitter receptors based on calculation of logarithm of dissociation constant (pKD) values (or logarithm of inhibitory constant [pKi] values) measured in vitro, displayed high affinity at the rat cortex  $\alpha$ 1-adrenergic receptors (pKD = 9.37), rat cortex 5-HT<sub>2A</sub> receptors (pKD = 8.31), human 5-HT<sub>2A</sub> receptors (pKi = 9.95), and calf caudate D<sub>2</sub> receptors (pKD = 8.28) (Studies PKF-98-03373 and RD-1999-02921). It had moderate affinity at human, rat, or bovine dopamine receptors (D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub>, and D<sub>4</sub>), rat  $\alpha$ 2C, human 5-HT<sub>6</sub>, human 5-HT<sub>1A</sub>, and calf 5-HT<sub>1B</sub> receptors. Weak affinity was seen at human  $\alpha$ 2A, human  $\alpha$ 2B, human 5-HT<sub>2B</sub>, human 5-HT<sub>2C</sub>, human 5-HT<sub>7</sub>, and histamine receptors. Very weak affinity (pKi range of 5 to 6) was observed at the norepinephrine (NE) transporter, human muscarinic receptors, and guinea pig  $\beta$ 1- and  $\beta$ 2-adrenergic receptor sites. Low affinity at histamine and muscarinic receptors was considered suggestive of a reduced potential for weight gain and side effects such as dry mouth, blurred vision, or other anti-cholinergic effects in clinical use (Kalkman, 2001, as cited by the sponsor). There was no cross-reactivity, up to concentrations of 10  $\mu$ M, at rat NMDA glycine or channel sites or any other receptors tested.

**In vitro receptor-binding data: human, rat, bovine, and guinea pig receptors**  
(Data from Studies PKF-98-03373 and RD-1999-02921)

Receptor	Ligand	Tissue	pK <sub>D</sub>
5-HT <sub>1A</sub>	<sup>3</sup> H-8-OH-DPAT	Human HeLa	7.13 ± 0.02
5-HT <sub>1B/1D</sub>	<sup>125</sup> I-GTI	Calf caudate	7.05 ± 0.05
5-HT <sub>2A</sub>	<sup>3</sup> H-Ketanserin	Rat cortex	8.31 ± 0.13
5-HT <sub>2A</sub>	<sup>3</sup> H-Ketanserin	Human CHO cells	9.95 ± 0.27 <sup>a</sup>
5-HT <sub>2C</sub>	<sup>3</sup> H-Mesulergine	Human SF9	6.60 ± 0.22
5-HT <sub>7</sub>	<sup>3</sup> H-Mesulergine	Human SF9	6.95 ± 0.15
Noradrenergic $\alpha$ <sub>1</sub>	<sup>125</sup> I-BE 2254	Rat cortex	9.37 ± 0.12
Noradrenergic $\alpha$ <sub>2</sub>	<sup>3</sup> H-Idazoxan	Rat cortex	6.74 ± 0.13
Noradrenergic $\beta$ <sub>1</sub>	<sup>125</sup> I-ICYP	Guinea pig heart	5.86 ± 0.03
Noradrenergic $\beta$ <sub>2</sub>	<sup>125</sup> I-ICYP	Guinea pig lung	5.51 ± 0.06
D <sub>1</sub>	<sup>3</sup> H-SCH 23390	Calf caudate	7.36 ± 0.05
D <sub>2</sub> ANT	<sup>3</sup> H-Spiperone	Calf caudate	8.28 ± 0.16
D <sub>2</sub>	<sup>3</sup> H-Spiperone	Rat cortex	7.76 ± 0.06
D <sub>3</sub>	<sup>3</sup> H-Spiperone	Human HEK	8.29 ± 0.07
D <sub>4</sub>	<sup>3</sup> H-Spiperone	Human HEK	7.37 ± 0.02

<sup>a</sup> Affinity for human 5-HT<sub>2A</sub> is expressed as mean pKi from Study RD-1999-02921; all other values in table are expressed as pKD from Study PKF-98-00373; CHO = Chinese hamster ovary; HEK = human embryonic kidney; pKD = logarithm of dissociation constant; 8-OHDPAT = 8-hydroxy-2-(di-n propylamino)tetralin.

Similar results were obtained in radioligand-binding assays of rat receptor subtype-binding affinities using 50% inhibitory concentration [IC<sub>50</sub>] (Study ILO-1PD-002), as shown in sponsor's table on the next page. Iloperidone exhibited a high affinity for  $\alpha$ 1-noradrenergic and 5-HT<sub>2</sub>-serotonergic receptors, a moderate affinity for  $\alpha$ 2-noradrenergic, D<sub>2</sub>-dopaminergic, 5-HT<sub>1A</sub>-serotonergic, and sigma receptors, low affinity for D<sub>1</sub>-dopaminergic and 5-HT<sub>3</sub>-serotonergic receptors, and no apparent affinity for muscarinic receptors, the [3H]TCP-binding site, or the glycine-binding site associated with the NMDA-receptor channel.

In comparison to compounds with established antipsychotic activity, iloperidone exhibited relatively greater affinity for 5-HT<sub>2</sub> than for D<sub>2</sub> receptors, similarly to clozapine, indicating potential atypical antipsychotic action. The respective ratios of IC<sub>50</sub> values for D<sub>2</sub>/5-HT<sub>2</sub> for iloperidone and clozapine were 10 and 23, whereas the ratios for haloperidol and thioridazine were 0.2 and 1.2, respectively (Study ILO-1PD-002).

**In vitro rat brain receptor-binding profile (IC<sub>50</sub>) of iloperidone and antipsychotic comparators**  
(Data from Studies ILO-1PD-002 and ILO-1PD-003)

Receptor	IC <sub>50</sub> (µM)			
	Iloperidone	Haloperidol	Thioridazine	Clozapine
D <sub>1</sub>	1.06 ± 0.15 <sup>a</sup>	0.755 <sup>b</sup>	0.299 <sup>b</sup>	1.20 <sup>b</sup>
D <sub>2</sub>	0.109 ± 0.002	0.0258 ± 0.004	0.233 ± 0.047	1.55 ± 0.23
5-HT <sub>1A</sub>	0.207 ± 0.03 <sup>a</sup>	7.19 ± 0.080	0.613 ± 0.188	0.821 ± ND
5-HT <sub>2</sub>	0.0111 ± 0.0085	0.132 ± 0.034	0.189 ± 0.059	0.067 ± 0.017
5-HT <sub>3</sub>	2.91 <sup>b</sup>	>10 <sup>b</sup>	>10 <sup>b</sup>	3.46
A <sub>1</sub>	0.00037 ± 0.00026	0.049 ± 0.011	0.0162 ± 0.005	0.030 ± 0.005
A <sub>2</sub>	0.060 <sup>b</sup>	>10 <sup>b</sup>	1.28 <sup>b</sup>	1.6 <sup>b</sup>
Sigma	0.064 ± NP	0.087 ± 0.042	2.16 <sup>b</sup>	>10 <sup>b</sup>
Muscarinic	>10 ± NP	>10 <sup>b</sup>	0.157 ± 0.059	0.11 ± 0.033
[ <sup>3</sup> H]TCP (NMDA-ion channel-binding site)	>10 <sup>b</sup>	>10 <sup>b</sup>	NP	>10 <sup>b</sup>
Glycine (NMDA receptor complex)	>100 <sup>c</sup>	NP	NP	NP

<sup>a</sup> Value is mean obtained from measurements reported in Study ILO-1PD-002 and Study ILO-IPD-003.

<sup>b</sup> Value based on single measurement reported in Study ILO-1PD-002. Unless otherwise indicated, other values are means reported in Study ILO-1PD-002 based on N determinations ranging from 2 to 16.

<sup>c</sup> Reported in Study ILO-IPD-003.

IC<sub>50</sub> = 50% inhibitory concentration; NP = not provided; NMDA = *N*-methyl-D-aspartic acid.

In a binding analysis of  $\alpha$ 1A- and  $\alpha$ 1B-adrenergic receptor types in rat liver and rat cortex, iloperidone was found to bind preferentially to  $\alpha$ 1A receptors, that are "primarily responsible for cardiovascular responses to changes in posture" (Study ILO-1PD-008). The extended binding affinity of iloperidone to  $\alpha$ -adrenergic receptors was further explored in vitro in another study (Study 1088657) that compared binding to rat  $\alpha$ 1A- and  $\alpha$ 1B-adrenergic receptor subtypes and human  $\alpha$ 1D-,  $\alpha$ 2A-,  $\alpha$ 2B-, and  $\alpha$ 2C-adrenergic receptor subtypes. Iloperidone was found to bind with relatively greater affinity at the  $\alpha$ 1 receptor subtypes evaluated in both rat and human (see sponsor's table below). It was also observed that in the rat, iloperidone has almost similar affinity to both  $\alpha$ 1A and  $\alpha$ 1B

receptors. This finding was different from the results obtained from Study ILO-1PD-008, “possibly due to methodology differences in two different laboratories”.

In vitro receptor-binding data: rat and human noradrenergic receptor subtypes  
(Data from Study 1088657)

Receptor	Species	IC <sub>50</sub> (nM)	K <sub>i</sub> (nM)
Noradrenergic α <sub>1A</sub>	Rat	0.233 ± 0.029	0.943 ± 0.012
Noradrenergic α <sub>1B</sub>	Rat	0.183 ± 0.012	0.101 ± 0.007
Noradrenergic α <sub>1D</sub>	Human	0.419 ± 0.015	0.206 ± 0.007
Noradrenergic α <sub>2A</sub>	Human	7.53 ± 0.689	2.82 ± 0.258
Noradrenergic α <sub>2B</sub>	Human	10.6 ± 1	4.82 ± 0.232
Noradrenergic α <sub>2C</sub>	Human	4.25 ± 0.172	0.617 ± 0.025

The α<sub>1</sub>-adrenergic receptor antagonism of iloperidone was studied in vitro in rat vascular tissues (Studies ILO-1PD-009 and ILO-1PD-010). Iloperidone was found to be a potent antagonist of norepinephrine (NE)-induced contraction of isolated rat aortic rings, which is an index of α<sub>1</sub>-receptor blockade in vascular tissues. The affinity of iloperidone for vascular α<sub>1</sub>-receptors (binding constant [KB] = 0.5 nM) was similar to that of risperidone (KB = 0.3 nM) and approximately 13 times that of clozapine (KB = 6.8 nM). Iloperidone also displayed potent competitive antagonism for vascular α<sub>1A</sub>-receptors in the rat mesenteric arterial bed. The KB value for iloperidone was 0.18 nM, compared to 35.6 nM for clozapine. These findings indicated that iloperidone was likely to have cardiovascular effects in vivo.

#### Ex vivo binding studies in rat

In ex vivo rat studies, iloperidone showed potent inhibition of the 5-HT<sub>2</sub> receptor and weak effects on D<sub>2</sub> receptors, thus demonstrating activities of an atypical antipsychotic.

Ex vivo binding of iloperidone to rat D<sub>2</sub> and 5-HT<sub>2</sub> receptors (N = 4/treatment)  
(Data from Study ILO-1PD-011)

Iloperidone concentration	[ <sup>3</sup> H]Spiperone bound			
	D <sub>2</sub> -specific		5-HT <sub>2</sub> -specific	
	fmoles/mg tissue	% control	fmoles/mg tissue	% control
<b>Rostral striatum</b>				
Vehicle	77.6 ± 2.7	100	8.3 ± 1.5	100
2.5 mg/kg	71.3 ± 4.1	92	2.3 ± 0.5 <sup>a</sup>	28
5 mg/kg	77.1 ± 0.6	99	2.9 ± 0.9 <sup>a</sup>	35
10 mg/kg	73.1 ± 3.7	95	1.8 ± 0.7 <sup>a</sup>	22
20 mg/kg	75.6 ± 1.8	97	1.2 ± 0.6 <sup>a</sup>	15
<b>Mid striatum</b>				
Vehicle	73.1 ± 2.2	100	6.5 ± 2.0	100
2.5 mg/kg	71.6 ± 1.3	98	1.7 ± 0.2 <sup>b</sup>	26
5 mg/kg	70.6 ± 1.7	97	1.8 ± 1.6 <sup>b</sup>	28
10 mg/kg	66.2 ± 2.6	91	1.4 ± 0.8 <sup>b</sup>	22
20 mg/kg	67.0 ± 1.8	92	0.4 ± 0.2 <sup>b</sup>	6
<b>Nucleus accumbens</b>				
Vehicle	60.6 ± 2.8	100	10.4 ± 2.5	100
2.5 mg/kg	55.6 ± 1.0	92	3.0 ± 1.0 <sup>a</sup>	29
5 mg/kg	55.8 ± 1.3	92	3.3 ± 1.5 <sup>b</sup>	32
10 mg/kg	52.3 ± 1.9	86	2.0 ± 0.8 <sup>a</sup>	19
20 mg/kg	51.2 ± 5.8	85	1.6 ± 0.7 <sup>a</sup>	15

a P<0.01, Newton-Keul's test.

b P<0.05, Newton-Keul's test.

Iloperidone i.p. administration to Wistar rats at 2.5 to 20 mg/kg (Study ILO-1PD-011) significantly inhibited, upon a 30-min pretreatment, the binding of [3H]spiperone ex vivo to cortical and subcortical 5-HT<sub>2</sub> serotonergic receptors (by 51% to 57% and by 61% to 85%, respectively), while the effects on the binding of [3H]spiperone to dopamine D<sub>2</sub> receptors in the corpus striatum and nucleus accumbens were weak and not statistically significant (maximal effect of 3% to 15% inhibition at 20 mg/kg) (see sponsor's table below). The iloperidone profile of potent inhibition ex vivo of the 5-HT<sub>2</sub> receptor and weak effects on D<sub>2</sub> receptors was similar to what was seen with clozapine at doses of 10 to 40 mg/kg i.p. with a 60- minute pretreatment.

In summary, the results of iloperidone in vitro and ex vivo receptor-binding studies indicated that iloperidone exhibits relatively high affinity for dopamine D<sub>2</sub> and D<sub>3</sub> receptors, noradrenergic  $\alpha$ <sub>1</sub> receptor subtypes evaluated in both rat and human, and serotonergic 5-HT<sub>2A</sub> receptors, as well as 5-HT<sub>1A</sub> and 5-HT<sub>6</sub> receptors. This binding profile is consistent with the potential of an atypical antipsychotic, but with risk of hemodynamic effects such as hypotension.

In vitro receptor-binding profile of iloperidone metabolites

The receptor-binding profiles of major metabolites of iloperidone, P88, P89 and P95, have been characterized. Metabolite P89 was found to bind with high affinity to D<sub>2</sub> receptors (IC<sub>50</sub> = 0.0118  $\mu$ M) and 5-HT<sub>2</sub> receptors (IC<sub>50</sub> = 0.00205  $\mu$ M), whereas P88 was approximately 30-fold and 10-fold weaker at these sites, respectively (IC<sub>50</sub> = 0.43 and 0.022  $\mu$ M, respectively). Both P88 and P89 also exhibited affinity for 5-HT<sub>1A</sub> receptors (IC<sub>50</sub> = 0.91 and 0.37  $\mu$ M, respectively),  $\alpha$ <sub>1</sub> noradrenergic receptors (IC<sub>50</sub> = <0.01 and 0.001  $\mu$ M, respectively),  $\alpha$ <sub>2</sub>- noradrenergic receptors (IC<sub>50</sub> = 0.21 and 2.9  $\mu$ M, respectively), and sigma opiate receptors (IC<sub>50</sub> = 0.97 and 0.36  $\mu$ M, respectively); P88 did not bind to muscarinic receptors and P89 was not tested at this receptor (Study ILO-MET-001). Another study evaluated the receptor affinity profile of metabolite P95 in comparison to iloperidone as shown in the following sponsor's table.

Receptor affinity profile of P95 and iloperidone for rat and human receptors  
(Data from Study 1081881)

Receptor	Species	Iloperidone K <sub>i</sub> (nM)	P95 K <sub>i</sub> (nM)
5-HT <sub>1A</sub>	Human	5.6 ± 0.3 <sup>a</sup>	3.91 ± 0.267
5-HT <sub>1B</sub>	Human	ND	933 ± 77
5-HT <sub>1C</sub>	Human	42.8 ± 1.6	2200 ± 271
5-HT <sub>1A</sub>	Human	ND	4820 ± 168
5-HT <sub>2</sub>	Rat	42.7 ± 35.4	ND
5-HT <sub>2</sub>	Human	63.1 <sup>b</sup>	4840 ± 395
5-HT <sub>2</sub>	Rat	21.6	ND
5-HT <sub>2</sub>	Human	112 <sup>b</sup>	955 ± 100
Adrenergic $\alpha$ <sub>1</sub> general	Rat	4.36	ND
Adrenergic $\alpha$ <sub>1A</sub>	Rat	ND	4.66 ± 0.431
Adrenergic $\alpha$ <sub>1B</sub>	Rat	ND	2.69 ± 0.270
Adrenergic $\alpha$ <sub>1D</sub>	Human	ND	8.78 ± 0.821
Adrenergic $\alpha$ <sub>2A</sub>	Human	162 <sup>b</sup>	26.6 ± 2
Adrenergic $\alpha$ <sub>2B</sub>	Human	162 <sup>b</sup>	11 ± 0
Adrenergic $\alpha$ <sub>2C</sub>	Human	16.2 <sup>b</sup>	5.18 ± 0.529
Dopamine D <sub>1</sub>	Human	216 ± 34	3990 ± 187
Dopamine D <sub>2L</sub>	Human	6.3 ± 1.5	684 ± 72
Dopamine D <sub>2S</sub>	Human	13.3 ± 3.5	473 ± 78
Dopamine D <sub>3</sub>	Human	7.1 ± 4.1	851 ± 46
Histamine H <sub>1</sub>	Guinea pig/Human	437 <sup>b,c</sup>	1360 ± 193 <sup>c</sup>

K<sub>i</sub> = inhibitory constant; ND = not determined.

Compared to the parent compound, P95 exhibited a substantially lower affinity for the D1, D2S, D2L, and D3 receptor subtypes, similar affinity for the human 5-HT<sub>2A</sub> receptor (K<sub>i</sub> = 3.91 and 5.6 nM for P95 and iloperidone, respectively) and higher or comparable affinity for adrenergic receptor subtypes tested. Similar to the parent compound, P95 had no appreciable affinity to histamine H<sub>1</sub> receptors.

A further evaluation of the main iloperidone metabolites, P95 and P88, for broad-spectrum receptor affinity for human, rat and guinea pig receptors using radiolabeled ligand-binding techniques (Study RD-2001-00343) showed that the P88 metabolite had moderate to strong affinity (pK<sub>i</sub> values ranging from 7 to 8) at the adrenergic  $\alpha$ <sub>1</sub>,  $\alpha$ <sub>2B</sub>,  $\alpha$ <sub>2C</sub>, dopamine D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub>, D<sub>4</sub>, histamine H<sub>1</sub>, 5-HT<sub>1B</sub>, and 5-HT<sub>2C</sub> sites. It had relatively weak affinity (pK<sub>i</sub> < 6.5) at the adrenergic  $\alpha$ <sub>2B</sub>, 5-HT<sub>1A</sub>, and 5-HT<sub>6</sub> sites. In comparison, the P95 metabolite showed weak to moderate affinity (pK<sub>i</sub> values ranging from 5.5 to 7.7) at the adrenergic  $\alpha$ <sub>1</sub>,  $\alpha$ <sub>2A</sub>,  $\alpha$ <sub>2B</sub>,  $\alpha$ <sub>2C</sub>, dopamine D<sub>1</sub>, D<sub>3</sub>, D<sub>4</sub>, and 5-HT<sub>6</sub> sites. Compared with the receptor profile of iloperidone, these metabolites are considered to have much weaker affinity at the various receptors. In a separate study (Study RD-1999-02921), both metabolites and the parent compound were found to have a high affinity for 5-HT<sub>2A</sub> (pK<sub>i</sub> values of 9.95, 9.56, and 8.15 nM for iloperidone, P88, and P95, respectively). The results are summarized in the following sponsor's table.

**P95 and P88 extended receptor affinity profile**  
(data from Studies RD-2001-00343 and RD-1999-0291)

Receptor	Species	P95 pK <sub>i</sub> ( $\pm$ SEM) (nM)	P88 pK <sub>i</sub> ( $\pm$ SEM) (nM)
Adrenergic $\alpha$ , general	Rat	7.67 $\pm$ 0.02	8.08 $\pm$ 0.11
Adrenergic $\alpha$ <sub>2A</sub>	Human	6.42 $\pm$ 0.02	6.44 $\pm$ 0.02
Adrenergic $\alpha$ <sub>2B</sub>	Human	7.08 $\pm$ 0.05	7.22 $\pm$ 0.02
Adrenergic $\alpha$ <sub>2C</sub>	Human	7.32 $\pm$ 0.09	7.79 $\pm$ 0.10
Dopamine D <sub>1</sub>	Human	5.94 $\pm$ 0.01	7.02 $\pm$ 0.01
Dopamine D <sub>2</sub>	Human	ND	7.80 $\pm$ 0.02
Dopamine D <sub>3</sub>	Rat	6.11 $\pm$ 0.05	7.17 $\pm$ 0.04
Dopamine D <sub>4</sub>	Human	ND	7.45 $\pm$ 0.05
Histamine H <sub>1</sub>	Guinea pig	ND	7.58 $\pm$ 0.20
5-HT <sub>1A</sub>	Human	ND	6.37 $\pm$ 0.01
5-HT <sub>1B</sub>	Human	ND	7.29 $\pm$ 0.03
5-HT <sub>2A</sub>	Human	8.15 $\pm$ 0.03	9.56 $\pm$ 0.21
5-HT <sub>2C</sub>	Human	ND	7.18 $\pm$ 0.05
5-HT <sub>6</sub>	Human	5.59 $\pm$ 0.18	6.11 $\pm$ 0.03

#### Characterization of iloperidone agonist/antagonist activity at receptor subtypes

The functional agonist/antagonist activity of iloperidone at a variety of dopaminergic, noradrenergic, and serotonergic receptors has been characterized in rat tissues ex vivo and in vivo, and at human receptor subtypes in vitro.

The rat studies indicated the potential for iloperidone to antagonize D<sub>2</sub> receptors similar to the typical antipsychotic haloperidol and stronger than the atypical antipsychotic clozapine (Study ILO-1PD-013); the  $\alpha$ <sub>2</sub>-antagonist properties of iloperidone (Study ILO-1PD-014); and a moderately potent inhibition of serotonin uptake (Study ILO-1PD-015).

The functional characterization assays at human receptor subtypes indicated that iloperidone does not possess any agonist activity at the dopaminergic, serotonergic, or noradrenergic receptors tested, but it is a potent inhibitor of the agonist response to dopamine at dopaminergic receptors, NE at noradrenergic receptors, and 8-OH-DPAT at serotonergic receptors. Iloperidone was most potent at D3 (pKB 8.59) receptors, followed by  $\alpha$ 2C (pKB 7.83), 5-HT1A (pKB 7.69) and D2A (pKB 7.53) receptors, and 5-HT6 receptors (pKB 7.11). This functional profile was “consistent with potential efficacy against psychotic symptoms and cognitive deficits, and with decreased potential for D2-associated side effects.” These findings are summarized in the following sponsor’s table:

**Iloperidone antagonist activity at human dopaminergic, serotonergic, and noradrenergic receptor subtypes**

(Data from Studies RD-1999-02368, RD-2000-00350, RD-2000-00462, RD-2000-02214)

Receptor	Agonist	pK <sub>B</sub>
D <sub>2A</sub>	Dopamine	7.53 ± 0.04
D <sub>3</sub>	Dopamine	8.59 ± 0.20
$\alpha$ <sub>2A</sub>	Norepinephrine	6.74 ± 0.05
$\alpha$ <sub>2C</sub>	Norepinephrine	7.83 ± 0.06
5-HT <sub>1A</sub>	8-OH-DPAT	7.69 ± 0.18
5-HT <sub>6</sub>	5-HT	7.11 ± 0.08

**Characterization of iloperidone metabolite P88 agonist/antagonist activity at dopamine and adrenergic receptors**

The agonist/antagonist activities of metabolite P88 and its enantiomer R(+)-P88 were determined in cell lines expressing either human recombinant  $\alpha$ 2C adrenergic receptor or the human D2A receptor (Study RD-2001-01349). Both enantiomers were devoid of significant agonist activity but inhibited the agonist response in a concentration-dependent fashion. Both enantiomers were equipotent at the  $\alpha$ 2C receptor (pKB = 7.34 and 7.24 for the S(-) and R(+) enantiomers, respectively). Both were also equipotent at the D2 receptor, exhibiting an affinity slightly greater than that for the  $\alpha$ 2C receptor (pKB = 7.76 and 7.78, respectively). P88 affinity for each receptor was similar to that noted for iloperidone. These results suggest that iloperidone metabolite P88 has the potential to exert functional effects on dopaminergic and adrenergic receptors similar to those of the parent compound, iloperidone.

**In vivo studies**

**Iloperidone: In vivo functional characterization of dopamine, serotonin, and adrenergic receptor antagonism in rats**

**- Dopamine receptor antagonism of iloperidone**

To determine in vivo interactions with CNS dopaminergic neurons, iloperidone was administered to rats either acutely or once daily for 21 days before neuronal sampling (Study ILO-1PD-026). Extracellular electrophysiological sampling of midbrain dopamine neurons was used to directly measure activity of these dopamine systems. Ideally, “dopaminergic blockade should take place preferentially in the substantia nigra pars compacta (SNC) compared with the ventral tegmental area (VTA), as it is its effects in

the latter area that have been associated with extrapyramidal symptoms (EPS) liability” (Szewczak, 1995, as cited by the sponsor). The numbers of spontaneously firing dopamine neurons within the SNC or VTA were counted. Acute administration of either iloperidone (5 mg/kg i.p.), haloperidol (0.5 mg/kg i.p.), or clozapine (20 mg/kg i.p.) resulted in a significant increase in the number of spontaneously active dopamine neurons in both the SNC and VTA areas. Lower doses of iloperidone (1 and 2 mg/kg i.p.) resulted in a significant increase in the SNC area only. According to the sponsor, the presumed mechanism for this effect, likely shared by iloperidone, haloperidol and clozapine, is interference with a feedback control system by way of dopamine receptor blockade. However, following chronic administration of either clozapine (20 mg/kg i.p.) or iloperidone (5 and 10 mg/kg i.p.), significant decreases were observed in the VTA dopamine single-units, whereas increases were noted in SNC dopamine single-units. Significant decreases were observed in both areas following chronic haloperidol (0.5 mg/kg) administration. The decreases following chronic treatment were hypothesized to be the result of depolarization blockade. This selective decrease of VTA dopamine single-unit neuronal activity seen with iloperidone suggested that this agent, similarly to atypical antipsychotic clozapine, has a potential as an antipsychotic agent with reduced EPS liability as compared with the typical antipsychotic haloperidol.

- Iloperidone effect on dopamine presynaptic autoreceptors

Iloperidone was administered to rats i.p. to test for striatal dopamine autoreceptor antagonist activity by its ability to reverse the effects of apomorphine on gamma-butyrolactone (GBL)-induced dopa accumulations (Study ILO-1PD-029). GBL administration in rats results in dopa accumulation in central dopamine neurons. This accumulation is reduced by treatment with apomorphine, presumably due to stimulation of presynaptic autoreceptors. Therefore, compounds that block dopamine autoreceptors will prevent the apomorphine-induced decrease in dopa accumulation after GBL administration. At iloperidone doses of 0.3, 1, 3, and 10 mg/kg i.p., inhibition of dopa accumulation by apomorphine was reversed by 0%, 29%, 33% to 42%, and 40%, respectively. Thus, iloperidone produced only partial reversal of the apomorphine effect, similar to the atypical antipsychotic clozapine that produced only partial reversal of dopa accumulation. In contrast, the typical antipsychotic haloperidol potently reversed apomorphine-induced inhibition of dopa accumulation by 12%, 34%, 71%, and 100% at 0.03, 0.1, 0.3, and 1 mg/kg, respectively. Therefore, iloperidone showed “slight to moderate” autoreceptor antagonist activity. This finding was consistent with the profile of an atypical antipsychotic, having in mind that typical antipsychotic agents have been differentiated from atypical agents in their interactions with striatal dopaminergic presynaptic autoreceptors (Szczepanik, 1996, as cited by the sponsor).

- Iloperidone effects of chronic treatment on D<sub>2</sub> and 5-HT<sub>2</sub> receptors

Chronic treatment of rats with iloperidone at 5 mg/kg/day i.p. did not significantly change the number or affinity of D<sub>2</sub> receptors in any region of the corpus striatum or the nucleus accumbens (Study ILO-1PD-028). The same treatment produced a significant down-regulation of cortical 5-HT<sub>2</sub> receptors to 41% to 59% of control in the dorsal and sulcal areas of the frontal cortex, with a nonsignificant decrease in the dissociation constant (K<sub>D</sub>). The effect on cortical 5-HT<sub>2</sub> receptors was similar in magnitude to that produced by clozapine (decreased to 49% of control). Chronic treatment with haloperidol did not affect either the number or affinity of 5-HT<sub>2</sub> receptors. These results differentiate

iloperidone from haloperidol, and indicate a neurochemical profile similar to atypical antipsychotics in which 5-HT<sub>2</sub> receptor blockade occurs concomitantly with D<sub>2</sub> receptor antagonism.

- Iloperidone  $\alpha$ 1-Adrenergic and non-selective serotonin receptor antagonism

Study ILO-IPD-032 employed the pithed rat model (which allows for direct evaluation of vascular responses in living tissue in the absence of sensory input) to assess the ability of iloperidone to antagonize the pressor effect of i.v. administered phenylephrine and serotonin, as measured by an increase in diastolic blood pressure. Both  $\alpha$ 1-adrenergic and serotonergic receptor agonism are implicated in the tonic regulation of blood pressure through direct interactions with vascular tissue. Oral administration of 1 and 6 mg/kg iloperidone competitively shifted the phenylephrine pressor response curve 6- and 32-fold, respectively, to the right. These doses of iloperidone produced a non-competitive blockade of the serotonin-induced pressor response in pithed rats. These findings demonstrate that iloperidone is a vascular  $\alpha$ 1-adrenergic and 5-HT<sub>2</sub> serotonergic receptor antagonist in rats, and indicate a potential for hypotensive effects in clinical use.

- Iloperidone metabolites P88 and P89: In vivo functional characterization of dopamine, serotonin, and adrenergic receptor antagonism in rats

Iloperidone metabolites P88 and P89 were characterized for their ability to antagonize dopamine, serotonin, and adrenergic receptors in a series of in vivo rat studies similar to those described above for iloperidone. Iloperidone metabolite P95 was not evaluated in these assays since this metabolite does not cross the blood-brain barrier. As shown in PK tissue distribution studies with radiolabelled P95, [<sup>14</sup>C]P95-derived materials were not observed in the brain, indicating minimal or no passage across the blood-brain barrier (Study DMPK(US)R99-2330 and DMPK(US)R01-417).

In a study that assessed the number of active dopamine neurons in both the ventral tegmental area (VTA) and the substantia nigra pars compacta (SNc) after P88 administration, a single i.p. dose of 0.25 mg/kg induced an increase of 26% in the VTA and a significant increase of 60% ( $P < 0.01$ ) in the SNc compared with vehicle treatment. A higher dose of 0.5 mg/kg i.p. caused a significant increase of 73% ( $P < 0.01$ ) in the VTA and a 29% increase in the SNc (Study ILO-MET-001). These findings are consistent with a dopamine antagonist profile for P88.

A single i.p. dose of 0.5 mg/kg of P89 induced significant ( $P < 0.01$ ) increases of 67% and 81% in the VTA and SNc, respectively, suggesting dopamine antagonist effects of P89. When evaluated in the same assay after 21 days of P89 administration at either 0.5 or 5 mg/kg i.p., the low-dose group had a significant ( $P < 0.05$ ) decrease of 20% in the VTA and an increase of 43% in the SNc compared with control. The high-dose group had a significant ( $P < 0.05$ ) decrease of 60% and 42% in the VTA and SNc, respectively, versus control. These results suggest a profile similar to haloperidol.

Dopa accumulation after aromatic amino acid decarboxylase inhibition was evaluated after treatment with P88 and P89. P88 caused a significant increase in dopa accumulation in the striatum, accumbens, and cortex at 10 and 30 mg/kg i.p. P89 caused significant increases in dopa accumulation in the striatum and accumbens at 0.03, 0.3, 3, and 30 mg/kg i.p. Both P88 (10 and 30 mg/kg i.p.) and P89 (0.3 and 3.1 mg/kg i.p.) reversed the antagonistic effects of apomorphine on GBL treatment, indicating that they both block dopamine autoreceptors. This activity is shared by antipsychotic agents such as haloperidol.

Overall, the results of functional assays conducted with iloperidone metabolites P88 and P89 indicate that both compounds exhibit dopamine receptor antagonist properties similar to those seen in antipsychotic agents with established clinical efficacy.

In vivo behavioral pharmacology studies

- Effects of iloperidone in behavioral assays

The antagonistic activities of iloperidone at dopaminergic, serotonergic, and noradrenergic receptors effects of iloperidone were confirmed in behavioral assays conducted in mice, rats, and monkeys. In addition, the assays provided indication for potential antipsychotic effect, activity associated with decreased extrapyramidal symptom liability, potential anxiolytic/negative symptom effect and potential for treatment of social withdrawal. The results of these studies are summarized in the following sponsor's table (data from Studies ILO-IPD-016, ILO-IPD-017, ILO-IPD-018, ILO-IPD-019, ILO-IPD-020, ILO-IPD-021, ILO-IPD-022, ILO-IPD-023, ILO-IPD-024, ILO-IPD-025, ILO-IPD-031, ILO-IPD-033, and RD-1999-037175).

Summary of activity of iloperidone and antipsychotic comparators in behavioral assays

Assay (Study No.)	Key Findings			
	Iloperidone	Clozapine	Haloperidol	Thioridazine
Climbing mouse assay (ILO-IPD-016)	ED <sub>50</sub> = 0.095 (0.090-0.101) i.p. ED <sub>50</sub> = 0.15 (0.14-0.16) p.o. Effect duration: 2-4 h	ED <sub>50</sub> = 8.1 (7.6-8.7) i.p. ED <sub>50</sub> = 23.2 (21.1-25.9) p.o. Effect duration: 2-4 h	ED <sub>50</sub> = 0.11 (0.10-0.13) i.p. ED <sub>50</sub> = 0.28 (0.27-0.29) p.o. Effect duration >4 h	ED <sub>50</sub> = 4.3 (3.9-4.8) i.p. ED <sub>50</sub> = 8.2 (7.0-9.3) p.o. Effect duration ≥4 h
Elevated half-enclosed platform assay in mice (RD-1999-037175)	Doses of 0.01-0.3 mg/kg dose-dependently and significantly reduced exploratory behavior	Doses of 0.01-0.3 mg/kg dose-dependently and significantly increased exploratory behavior	ND	ND
MK-801-induced locomotion and falling behavior in mice (ILO-IPD-017)	ED <sub>50</sub> = 0.10 (0.06-0.15) i.p.	ED <sub>50</sub> = 1.13 (0.67-1.90) i.p.	ED <sub>50</sub> = 0.50 (0.28-0.90) i.p.	ND
Yohimbine-induced seizure in mice (ILO-IPD-018)	ED <sub>50</sub> = 1.5 (0.06-3.3) i.p.	ED <sub>50</sub> = 30.7 (14.4-65.4) i.p.	Only 25% inhibition of clonic convulsion at 0.5 mg/kg i.p.	No inhibition
PCP-induced social deficits in rats (ILO-IPD-019)	Failed to reverse PCP-induced deficits at 0.125 and 0.25 mg/kg i.p. Had no effect on locomotion	Reversed PCP-induced deficits at 2.5 mg/kg i.p. Had no effect on locomotion	Failed to reverse PCP-induced deficits at 0.125 and 0.25 mg/kg i.p. Caused decreased locomotion at 0.25 mg/kg i.p.	ND
Rat self-stimulation (ILO-IPD-020)	ED <sub>50</sub> = 0.15 (0.12-0.17) i.p. ED <sub>50</sub> = 1.03 (0.89-1.18) p.o.	ED <sub>50</sub> = 7.4 (7.2-7.7) i.p.	ED <sub>50</sub> = 0.077 (0.073-0.081) i.p.	ND
Continuous Sidman avoidance in rats (ILO-IPD-021)	ED <sub>50</sub> AVR = NC i.p. ED <sub>50</sub> AVR = 6.3 (5.0-7.7) p.o. ED <sub>50</sub> SRA = NC i.p. ED <sub>50</sub> SRA = 4.4 (3.3-5.3) p.o.	ED <sub>50</sub> AVR or SRA = NC i.p. or p.o.	ED <sub>50</sub> AVR = 0.049 (0.034-0.066) i.p. ED <sub>50</sub> AVR = 0.46 (0.4-0.5) p.o. ED <sub>50</sub> SRA = 0.043 (0.037-0.05) i.p. ED <sub>50</sub> SRA = 0.63 (0.6-0.7) p.o.	ED <sub>50</sub> = 15.7 (14.2-17.2) i.p. ED <sub>50</sub> SRA = 21.8 (20.2-23.5) i.p. ED <sub>50</sub> AVR or SRA = NC p.o.
Sidman avoidance in monkeys (Study ILO-IPD-033)	ED <sub>50</sub> AVR = 2.4 (2.2-2.8) p.o. @ 3.5 h ED <sub>50</sub> SRA = 4.1 (3.5-5.0) p.o. @ 3.5 h	ED <sub>50</sub> AVR = 16.8 (14.6-19.0) p.o. @ 3.0 h ED <sub>50</sub> SRA = 17.7 (15.6-19.8) p.o. @ 3.0 h	ED <sub>50</sub> AVR = 0.74 (0.65-0.83) p.o. @ 4.5 h ED <sub>50</sub> SRA = 1.03 (0.93-1.14) p.o. @ 4.5 h	ND

Summary of activity of iloperidone and antipsychotic comparators in behavioral assays (Continued)

Assay (Study No.)	Iloperidone	Chlorazine	Haloperidol	Thioridazine
Pole-climb avoidance in rats (ILO-1PD-022)	i.p.: ED <sub>50</sub> AVR = 0.65 (0.46-0.85); ED <sub>50</sub> EF = 5.2 (3.6 (8.9)); EF/AV = 8.0  p.o.: ED <sub>50</sub> AVR = 6.0 (5.5-6.7); ED <sub>50</sub> EF = 16.0 (estimated); EF/AV = 2.7	i.p.: ED <sub>50</sub> AVR = 11.3 (9.5-13.2); ED <sub>50</sub> EF = 29.3 (25.4-35.1); EF/AV = 2.6  p.o.: ED <sub>50</sub> AVR = 12.7 (11.2-14.1); ED <sub>50</sub> EF = 33.1 (29.1-39.9); EF/AV = 2.6	i.p.: ED <sub>50</sub> AVR = 0.043 (0.039-0.045); ED <sub>50</sub> EF = 0.22 (0.19-0.25); EF/AV = 5.1  p.o.: ED <sub>50</sub> AVR = 1.1 (1.0-1.2); ED <sub>50</sub> EF = 14.2 (11.9-17.5); EF/AV = 12.9	i.p.: ED <sub>50</sub> AVR = 18.1 (16.6-19.8); ED <sub>50</sub> EF = 31.3 (29.2-33.8); EF/AV = 1.7  p.o.: ED <sub>50</sub> AVR = 35.8 (30.7-41.2); ED <sub>50</sub> EF = 72.2 (60.8-89.6); EF/AV = 2.0
L-5-HTP-induced head twitch behavior in rats (ILO-1PD-023)	ED <sub>50</sub> = 0.07 (0.056-0.088) i.p.	ED <sub>50</sub> = 0.43 (0.37-0.51) i.p.	ED <sub>50</sub> = 0.29 (0.25-0.33) i.p.	ND
Social interaction in rats (ILO-1PD-024)	+25% interaction <sup>a</sup> , -11% total @ 0.5 i.p. +30% interaction <sup>a</sup> , -18% total @ 1.0 i.p. +9% interaction, -35% total @ 2.5 i.p.	+13% interaction, -18% total @ 5.0 i.p.; +39% interaction <sup>a</sup> , -15% total <sup>a</sup> at 10.0 i.p.	-19% interaction <sup>a</sup> , -11% total @ 0.05 i.p.; -32% interaction <sup>a</sup> , -22% total <sup>a</sup> @ 0.125 i.p.	-5% interaction, -4% total @ 2.5 i.p.; -13% interaction, -23% total <sup>a</sup> @ 5.0 i.p.
Elevated plus maze in rats (ILO-1PD-025)	+38% open <sup>a</sup> , -23% total <sup>a</sup> @ 0.5 i.p.; +9% open, -44% total <sup>a</sup> @ 1.0 i.p.	+15% open, 0% total @ 1.0 i.p.; +62% open <sup>a</sup> , -9% total @ 2.5 i.p.	-28% open, -32% total @ 0.05 i.p.; -47% open, -55% total <sup>a</sup> @ 0.10 i.p.	-21% open, -24% total <sup>a</sup> @ 1.0 i.p.; -27% open, -41% total <sup>a</sup> @ 2.5 i.p.
Cook and Davidson fixed ratio conflict paradigm (ILO-1PD-031)	+396% and +1009% <sup>a</sup> @ 0.1 and 0.3, i.p. Diazepam: +199% and +1000% <sup>a</sup> @ 1.0 and 3.0 p.o.	ND	ND	ND

Note: All concentrations are mg/kg. <sup>a</sup> P<0.05 vs. control; AVR = avoidance response; ED50 = median effective dose; EF = escape failure; PCP = phencyclidine; ND = not done; NC = not calculated; SHA = shocks avoided.

- Effects of iloperidone metabolites in behavioral assays

Pharmacology activity of iloperidone metabolites P88 and P89 was investigated in similar behavioral animal models (Study ILO-MET-001), as summarized in the following sponsor's table.

Summary of activity of iloperidone metabolites in behavioral assays

Assay	P88 (mg/kg)	P89 (mg/kg)
Climbing mouse assay	ED <sub>50</sub> = 0.12 i.p., 0.13 p.o.	ED <sub>50</sub> = 0.05 i.p., 0.19 p.o.
Apomorphine stereotypy in rats	ED <sub>50</sub> = 32.5 i.p.	ED <sub>50</sub> = 0.7 i.p.
Catalepsy in rats	ED <sub>50</sub> = 29 i.p.	ED <sub>50</sub> = 0.8 i.p.
Rat self-stimulation	ED <sub>50</sub> = 0.3 i.p.	ED <sub>50</sub> = 0.016 i.p.
Continuous Sidman avoidance in rats	AV ED <sub>50</sub> = 3.8 i.p. SA ED <sub>50</sub> = 2.7 i.p.	AV ED <sub>50</sub> = variable act SA ED <sub>50</sub> = 0.09 i.p.
Continuous Sidman avoidance in monkeys	AV ED <sub>50</sub> = 0.8 p.o. SA ED <sub>50</sub> = 1.9 p.o.	AV ED <sub>50</sub> = 4.4 p.o. SA ED <sub>50</sub> = 8.7 p.o.
Pole climb avoidance in rats	AV ED <sub>50</sub> = 1.1 i.p. EF ED <sub>50</sub> = 5.3 i.p.	AV ED <sub>50</sub> = 0.2 i.p. EF ED <sub>50</sub> = 0.38 i.p.
Social interaction in rats	-4% interaction, -5% total activity @ 0.125 i.p. +1% interaction, -15% total activity @ 0.25 i.p. 0% interaction, -37% total activity @ 0.5 i.p.	0% interaction, -3% total activity @ 0.1 i.p. +15% interaction, -47% total activity @ 0.25 i.p. +26% interaction, -64% total activity @ 0.5 i.p.
Elevated plus maze in rats	-22% open, -10% total @ 0.1 i.p. -38% open, -37% total @ 0.25 i.p.	Not tested

AV = avoidance responding; ED50 = median effective dose; EF = escape failures; SA = shocks avoided; SNC = substantia nigra compacta; VTA = ventral tegmental area.

The above behavioral pharmacodynamic assays conducted with P88 and P89 in mice, rats, and monkeys indicate that these metabolites possess activity consistent with a dopamine antagonist

profile similar to iloperidone. However, in contrast to the parent compound, neither metabolite appeared to possess anxiolytic activity, as evidenced by demonstrable effects in the social interaction assay or the elevated plus maze paradigm.

#### **Effects of iloperidone on gene expression in the brain**

The effects of iloperidone on the expression of target genes in the brain relevant to the potential for antipsychotic activity were evaluated in rats upon daily i.p. administration of iloperidone (1 mg/kg, i.p.) for 21 days (Study RD-2002-2660). Reverse transcriptase polymerase chain reaction was used to quantify changes in gene expression in the frontal cortex, hippocampus, and striatum. Target genes included the D2 and D3 receptor genes; glutamate-related genes (NMDA receptor subunits NR1, NR2A, NR2B, NR2C, and the glutamate transporter GLT-1); GABA-related genes (glutamic acid decarboxylase 67 kDa isoform [GAD67], GABA transporter GAT-1, and reelin (a growth and differentiation factor involved in neurotrafficking and dendritic spine growth; growth factor genes brain-derived neurotrophic factor (BDNF) and neruegulin-1 (NRG-1); proto-oncogene cfos, and  $\gamma$ -synuclein. Following treatment, the animals were sacrificed and RNA was isolated from the brain tissue for use in RT-PCR assays. The results of this study were summarized by the sponsor as follows: "Iloperidone treatment produced an increase of expression levels of D2 receptors in the hippocampus and striatum, indicating relevant D2 receptor blockade. Iloperidone weakly enhanced expression of the NR2A and NR2B receptor subunits and decreased NR2C and GLT-1 uptake site expression. In contrast, iloperidone markedly increased the expression of GABA-related genes reelin and GAD67 in the frontal cortex and hippocampus, but not in the striatum, and increased expression of GAT-1 in the hippocampus. Reelin expression was increased in the frontal cortex and hippocampus, whereas NRG-1 expression was increased in the frontal cortex, hippocampus, and striatum. Reelin expression has been shown to be reduced in the frontal cortex and hippocampus of patients with schizophrenia, and thus iloperidone treatment might ameliorate reelin deficits. GAD67 expression has also been shown to be lower in patients with psychotic disorder (schizophrenia and bipolar disorder); thus, these results raise the possibility that iloperidone might have the potential to ameliorate the GAD67 deficit. Overall, the results of this study indicated that iloperidone can either directly or indirectly affect expression levels of a number of gene products thought to play roles in the etiology of schizophrenia."

#### **2.6.2.3 Secondary pharmacodynamics**

Secondary pharmacodynamic properties of iloperidone, i.e., analgesic activity, anti-cholinergic activity, extrapyramidal symptom liability, and antagonism of peripheral  $\alpha$ -adrenergic activity were evaluated in 6 studies, summarized in the sponsor's table on the next page.

Analgesic activity: Iloperidone analgesic activity was assessed in an animal pain model (writhing behavior in response to phenylquinone administration) (Study ILO-2PD-001). Iloperidone was dose-dependently active in blocking phenylquinone-induced writhing behavior in Swiss CD-1 mice (ED<sub>50</sub> = 0.03 mg/kg, s.c.) that indicates potential analgesic activity.

Anti-cholinergic activity: Physostigmine-induced lethality in mice was not antagonized by iloperidone i.p. administration (Study ILO-2PD-002). In contrast, a dose- and time-

dependent antagonism of lethality was observed with clozapine. These results indicate that iloperidone lacks anti-cholinergic properties.

Extrapyramidal symptom (EPS) liability was assessed in a model of apomorphine-induced stereotyped behavior in rats which is an indication of dopaminergic stimulation. Antagonism of this behavior is linked to the blockade of dopamine receptors in the corpus striatum, which is linked clinically to EPS and is predictive of the propensity for EPS and tardive dyskinesia. Iloperidone dose-dependently antagonized apomorphine-induced stereotypy, but was markedly less potent than haloperidol (ED<sub>50</sub> = 34.8 mg/kg i.p. vs. 0.60 mg/kg i.p. for iloperidone and haloperidol, respectively) (Study ILO-2PD-003).

Iloperidone was also less potent than haloperidol in its ability to induce catalepsy (i.e., failure to correct externally imposed posture) in rats (ED<sub>50</sub> = 30.7 mg/kg i.p. vs. 0.65 mg/kg i.p., respectively) (Study ILO-2PD-004). Clozapine and thioridazine were not active in this paradigm at doses exceeding those of iloperidone. Induction of catalepsy is indicative of inhibition of the nigrostriatal system, believed to be responsible for EPS.

The results of both these studies indicate that iloperidone was less potent vs. haloperidol to induce EPS.

Both iloperidone (1.5 and 3 mg/kg i.p.) and clozapine (20 mg/kg i.p.) significantly decreased haloperidol-induced catalepsy in rats (Study ILO-2PD-005). This suggests that iloperidone, like clozapine, has the potential to ameliorate some of the EPS seen with dopamine-receptor blockade

Antagonism of peripheral  $\alpha$ -adrenergic activity: Iloperidone was active (ED<sub>50</sub> = 0.30 mg/kg i.p.) in protecting rats from norepinephrine-induced lethality (Study ILO-2PD-006). Since norepinephrine-induced lethality is regarded as being induced through the peripheral  $\alpha$ 1 adrenergic receptors, this result suggests that iloperidone blocks peripheral  $\alpha$ -adrenergic receptors.

Summary of activity of iloperidone and comparators in secondary pharmacodynamic assays

Assay (Study No.)	Key Findings			
	Iloperidone	Clozapine	Haloperidol	Thioridazine
Phenylquinone-induced writhing in mice (ILO-2PD-001)	ED <sub>50</sub> = 0.03 (0.023-0.033) s.c.	ND	ED <sub>50</sub> = 2.7 (2.5-2.9) p.o.	ND
Physostigmine-induced lethality in mice (ILO-2PD-002)	No antagonism at 40 mg/kg at any time point	ED <sub>50</sub> = 12.6 i.p. at 60 min	No antagonism at 10 mg/kg; 50% antagonism at 20 mg/kg at 120 min	ND
Apomorphine-induced stereotypy in rats (ILO-2PD-003)	ED <sub>50</sub> = 34.8 (22.0-55.1) i.p.	No antagonism at 40 mg/kg i.p.	ED <sub>50</sub> = 0.60 (0.40-0.80) i.p.	1/6 rats protected at 20 mg/kg i.p.
Catalepsy in rats (ILO-2PD-004)	ED <sub>50</sub> = 30.7 (19.6-48.1) i.p.	No activity	ED <sub>50</sub> = 0.65 (0.39-1.09) i.p.	No activity
Prevention of haloperidol-induced catalepsy (haloperidol administered at 0.5 mg/kg) (ILO-2PD-005)	Cataleptic score 1.5 mg/kg = 4.9 ± 1.4 and 6.0 ± 1.1 at 60 and 120 min, respectively 3.0 mg/kg = 4.9 ± 1.2 and 8.0 ± 0.6 at 60 and 120 min, respectively	Cataleptic score 20 mg/kg = 1.0 ± 0.9 and 1.2 ± 0.9 at 60 and 120 min, respectively	Cataleptic score 0.5 mg/kg = 8.0 ± 0.5 and 8.9 ± 6.9 at 60 and 120 min, respectively	ND
Norepinephrine lethality (ILO-2PD-006)	ED <sub>50</sub> = 0.30 (0.20-0.40) i.p.	ND	ND	ND

Note: All concentrations are mg/kg; ED<sub>50</sub> = median effective dose; ND = not done;

## 2.6.2.4 Safety pharmacology

### Neurological effects:

Pharmacological effects of P95 on general and neurobehavioral activities were studied in male CD-1(ICR)BR mice (10/group) at single oral (gavage) doses of 10, 30, 100, or 300 mg/kg of P95, and a vehicle control (Study 0170079). Animals were observed for general condition and neurobehavioral activity at approximately 15 minutes, 1, 2, 6, and 24 hours postdose. Clinical signs were recorded pretest and during the treatment. Gross necropsy was performed following the 24-hour postdose observation period.

There was no mortality. Dose-dependent neurobehavioral effects were observed at all dose levels. Ataxia, decreased locomotor activity, and/or ptosis were seen at  $\geq 10$  mg/kg; in addition, hyposensitivity to sound and impaired righting reflex were observed at doses of  $\geq 30$  mg/kg. The signs resolved in most animals by 6 h. post dose at doses of  $\leq 30$  mg/kg, and by the 24-hour time point at doses of  $\geq 100$  mg/kg. There were significant decreases in mean body temperature at all doses. No test article-related macroscopic lesions were observed at necropsy.

In conclusion, P95 induced changes in general and neurobehavioral activities in mice at oral doses of  $\geq 10$  mg/kg that are most likely due to exaggerated pharmacological response to the treatment.

### Cardiovascular effects:

#### *In vitro*:

#### - Effect of iloperidone and metabolites on cardiac action potential in vitro

The effects of iloperidone and its major metabolites P88 and P95 (each at 0.01, 0.1, 1 and 10  $\mu$ M) on cardiac action potential and depolarization parameters were studied in dog isolated Purkinje fiber preparations electrically stimulated at 1 and 0.5 Hz (Study 0120065). At 10  $\mu$ M, the effects on maximum rate of depolarization were investigated at a stimulation frequency of 3 Hz to assess potential effects on sodium channels. *DL*-sotalol hydrochloride (50  $\mu$ M) was used as a positive control. The following parameters were measured in fibers: action potential duration at 60% and 90% repolarization, maximum rate of depolarization, upstroke amplitude, and resting membrane potential. Change from baseline in action potential duration at 90% repolarization at 0.5 and 1 Hz for iloperidone, P88, and P95 is summarized in the following sponsor's table. Similar results were seen at 60% repolarization.

Change from baseline in action potential duration for iloperidone and metabolites <sup>a</sup>

Test item	Change from baseline (ms)			
	0.01 $\mu$ M	0.1 $\mu$ M	1.0 $\mu$ M	10.0 $\mu$ M
<b>Stimulation frequency</b>				
<b>Iloperidone</b>				
1 Hz	2.3 $\pm$ 0.7	9.1 $\pm$ 2.0 <sup>b</sup>	43.8 $\pm$ 6.2 <sup>c</sup>	47.6 $\pm$ 10.9 <sup>c</sup>
0.5 Hz	2.2 $\pm$ 1.8	9.0 $\pm$ 2.7	58.5 $\pm$ 10.4 <sup>c</sup>	61.4 $\pm$ 14.4 <sup>c</sup>
<b>(-)P88</b>				
1 Hz	-0.3 $\pm$ 0.8	9.3 $\pm$ 3.6 <sup>b</sup>	42.8 $\pm$ 10.7 <sup>c</sup>	38.9 $\pm$ 10.9 <sup>c</sup>
0.5 Hz	-0.3 $\pm$ 1.2	13.6 $\pm$ 5.9	59.8 $\pm$ 16.6 <sup>c</sup>	50.7 $\pm$ 13.8 <sup>c</sup>
<b>P95</b>				
1 Hz	1.3 $\pm$ 2.3	0.9 $\pm$ 2.3	1.6 $\pm$ 4.3	9.4 $\pm$ 3.2
0.5 Hz	0.2 $\pm$ 2.5	1.7 $\pm$ 3.2	3.5 $\pm$ 4.3	13.9 $\pm$ 4.3
<b>Vehicle</b>	0.1% DMSO	0.1% DMSO	0.1% DMSO	0.1% DMSO
1 Hz	2.2 $\pm$ 1.3	1.7 $\pm$ 1.4	2.9 $\pm$ 1.6	2.9 $\pm$ 1.5
0.5 Hz	1.8 $\pm$ 2.3	2.1 $\pm$ 1.4	4.1 $\pm$ 2.1	4.0 $\pm$ 1.8

<sup>a</sup> Action potential data are shown for results at 90% repolarization; <sup>b</sup>  $P < 0.05$  <sup>c</sup>  $P < 0.01$  DMSO = dimethylsulfoxide.

b(4)

Iloperidone and its metabolite P88 prolonged action potential duration at a concentration of 0.1  $\mu\text{M}$  and above and at stimulation frequencies of 0.5 and 1 Hz. The highest concentration tested (10  $\mu\text{M}$ ) induced a depression of the plateau phase of the action potential. There was also a reduction in maximum rate of depolarization at 3 Hz at this concentration, indicating a frequency-dependent interaction with cardiac sodium channels. Purkinje fibers exposure to P95 at concentration of 0.01, 0.1, 1 and 10  $\mu\text{M}$  induced a prolongation in action potential duration at 10  $\mu\text{M}$  but it did not reach the level of statistical significance; no prolongation was seen at the concentration below or at 1  $\mu\text{M}$ . These data indicate that iloperidone and P88 have the potential to prolong the QT interval at free plasma concentrations of 0.1  $\mu\text{M}$  and above, while P95 has the potential to prolong the QT interval at free plasma concentrations of over 10  $\mu\text{M}$ . This extent of exposure to P95 is unlikely, as administration of iloperidone doses of 24 mg given once-a-day in human patients yielded maximal P95 steady-state plasma levels of 55.5 ng/mL, or 0.13  $\mu\text{M}$ .

- Effect of iloperidone and metabolites on cloned hERG channels

The in vitro effects of iloperidone and its metabolites P95 and P88 on mammalian cells stably transfected with cloned cDNA of the cardiac ion channel hERG were studied in comparison with risperidone and ziprasidone (Study 008167).

**Concentration-response and temperature-dependence of ILO522, ILO522 P95-12113 metabolite, ILO522 P88-8991 metabolite, risperidone and ziprasidone on this ion channel current were measured.**

All test articles produced rapid, reversible blockade of hERG currents. Assessment of the blockade at near-physiological temperature showed an increase of the IC<sub>50</sub> value for the blockade for all test items except for risperidone (as shown in the sponsor's table below). The block potency rank order was iloperidone > ziprasidone  $\approx$  P88 > risperidone > P95. These data suggest that iloperidone and its P88 metabolite possess a QT prolongation potential, while P95 is unlikely to contribute to the QT prolongation potential of iloperidone.

Summary of effects on hERG ion channel currents

Test article	hERG IC <sub>50</sub> (nM)	hERG IC <sub>50</sub> (nM) at 34-35°C
Iloperidone	29	37
P95 metabolite	4319	12,789
P88 metabolite	56	100
Risperidone	394	188
Ziprasidone	55	79

*In vivo*

Effects of iloperidone on hemodynamic parameters

Since the receptor affinity and functional assay results indicated that iloperidone antagonized vascular  $\alpha_1$ -adrenoceptors, potentially causing hypotensive and other hemodynamic effects, a series of safety pharmacology studies was conducted in normal and spontaneously hypertensive rats and in conscious and anesthetized dogs to evaluate the effect of iloperidone on hemodynamic parameters such as blood pressure, heart rate, cardiac output, and ECG evaluations. The results of these studies indicated that iloperidone and its metabolites, P98 and P99 had hypotensive effects mediated through

blockade of  $\alpha_1$ -adrenergic receptors; this effect could be gradual or steep depending on the method of administration (slow infusion vs. bolus injection).

- Effects on hemodynamic parameters in conscious rats

Iloperidone administration at an oral dose of 3 mg/kg to conscious normotensive Long Evans male rats (N = 3) caused reduction in mean arterial pressure throughout 240 min. with a maximal decrease of 13% (-15 mm Hg) at 180 min post dose and a 5% decrease in heart rate (Study ILO-SP-002). In comparison, clozapine (10 mg/kg p.o.) increased mean arterial pressure by 5 mm Hg and heart rate by 18 beats per minute. The differential hemodynamic effects of clozapine were attributed to "a different degree of functional antagonism at  $\alpha_1$ -adrenergic receptors". In another study (ILO-SP-001), iloperidone administered orally at 10 mg/kg to conscious, normotensive Long Evans rats (N = 4) had no effect on mean arterial pressure or heart rate, although 1 of 4 rats showed a peak 41% decrease in mean arterial pressure at 90 minutes. There was a statistically significant reduction in mean arterial pressure at 240 minutes ( $-9.0 \pm 1.3\%$ ), but the sponsor did not consider it to be of biological significance.

Spontaneously hypertensive rats were used in another two experiments (Study ILO-SP-003 and Study ILO-SP-004). Iloperidone administration to conscious, spontaneously hypertensive SRH rats (N = 4/group) at oral doses of 1 and 3 mg/kg, decreased arterial systolic pressure by 11% (-19 mm Hg) and by 13% (-15 mm Hg) in the 1 and 3 mg/kg group, respectively. Heart rate was decreased by 16 bpm (-4%) at 1 mg/kg and by 10 bpm at 3 mg/kg. Clozapine administration (10 mg/kg p.o.) had a lesser effect: decreased systolic pressure by 3 mm Hg and heart rate by 1 bpm (Study ILO-SP-003).

In study ILO-SP-004, iloperidone administered orally to conscious, spontaneously hypertensive SRH rats (N = 4) at 0.3 and 1 mg/kg had no effect on basal blood pressure or heart rate, but induced a 48% inhibition of the hypertensive response to phenylephrine at 1 mg/kg dose. At higher doses (6 mg/kg and 20 mg/kg p.o.) iloperidone lowered basal blood pressure by up to 45% and inhibited the phenylephrine hypertensive response by as much as 89%. These results suggest that blockade of vascular  $\alpha_1$ -adrenergic receptors on vascular smooth muscle was present even at the dose of 1 mg/kg. The ED<sub>50</sub> value for inhibiting the hypertensive response to the  $\alpha$ -adrenergic agonist was 0.6 mg/kg. Clozapine at 10 mg/kg (N = 4) increased mean arterial pressure by 5 mm Hg and heart rate by 18 bpm.

Overall, the hemodynamic investigations in normal and hypertensive rats indicated that iloperidone exerts hypotensive effect most likely due to blockade of  $\alpha_1$ -adrenergic receptors.

- Effects on hemodynamic parameters in anesthetized and conscious dogs

The cardiovascular effects of iloperidone i.v. administration to anesthetized dogs given as either a bolus injection (15 seconds) or a slow intravenous infusion (30 minutes) at doses of 0.02 and 1 mg/kg were evaluated (Study ILO-SP-005). A decrease in systemic arterial pressure occurred, consistent with the results seen in rats. The hypotensive response (39%) was primarily due to a reduction in peripheral resistance (40%) mediated by vascular  $\alpha_1$ -adrenergic receptor blockade. Heart rate, cardiac output, and ECG were not affected. The hypotensive effects produced by 0.02 mg/kg bolus (N = 4) or slow infusion (N = 3) were of similar magnitude and subsided within 2 hours of dosing. The fall in arterial pressure was gradual in onset with the slow infusion (peak reduction in 30 min) in contrast to the rapid hypotensive effect caused by the bolus injection (peak reduction in 5

min). Iloperidone plasma levels following the bolus i.v. injection of 0.02 mg/kg were consistently higher than those following the slow i.v. infusion of the same dose (Study RCE118.DOC). Plasma levels for iloperidone after a bolus injection ranged from 5 ng/ml to 96 ng/ml, with an average C<sub>max</sub> value of 63 ng/ml (N = 4). The corresponding iloperidone plasma levels after a slow infusion ranged from 5 ng/mL to 24 ng/ml, with an average C<sub>max</sub> value of 15 ng/ml (N = 3). These findings suggest that human subjects may better tolerate gradual onset of hypotension associated with slow infusion of iloperidone.

Study ILO-SP-006 compared the effects of single oral doses of iloperidone (2 and 5 mg/kg) and clozapine (5 mg/kg p.o.) on hemodynamic parameters in conscious dogs. Administration of iloperidone resulted in a sustained decrease in mean arterial pressure with peak decreases of 24 mm Hg (-24%) and 20 mm Hg (-22%) at the 2 and 5 mg/kg, respectively. Peak responses occurred between 2 and 3 hours post-dose. In comparison, a peak decrease of 16 mm Hg (-16%) was observed following oral clozapine, with the peak response at 1 hour post-dose. Drug-induced variations in heart rate were not observed. Both clozapine and iloperidone produced sedation in the dogs. The results indicate that iloperidone oral administration at doses of 2 and 5 mg/kg produced vasodepressor effects in conscious dogs comparable to those of clozapine (5 mg oral). The peak time of hemodynamic effects of iloperidone correlated with peak mean plasma levels noted with clinical use of iloperidone.

Effects of a cumulative oral dosing were explored following 3 days of oral administration of iloperidone to conscious dogs at doses of 0.5, 1, and 10 mg/kg (Study ILO-SP-009). On the fourth day the animals were anesthetized and instrumented for hemodynamic evaluation. After 3 days of iloperidone dosing, the hypotensive effects and decreases in peripheral resistance were similar to those in dogs administered the same doses acutely. Intra-duodenal (i.d.) administration of iloperidone (2 and 10 mg/kg) and clozapine (5 mg/kg i.d.) caused similar hypotensive and vasodilating effects in anesthetized dogs (Study ILO-SP-007). However, iloperidone appeared to exert less influence on cardiac output and heart rate than did clozapine. Moreover, iloperidone exhibited positive inotropic effects, while clozapine showed negative inotropic effects, as evidenced by the progressive reduction in the rate of left ventricular pressure development (dP/dt<sub>max</sub>). No drug-related ECG changes were noted for either compound.

Study ILO-SP-008 examined the dose-response relationship of hemodynamic effects of single intra-duodenal doses of iloperidone (0.5, 1, and 10 mg/kg) in anesthetized dogs. Hemodynamic parameters were monitored for 120 minutes. 0.5 mg/kg was a no-effect dose with regard to hypotension and blockade of peripheral vasoconstrictor responses; the doses of 1 and 10 mg/kg produced measurable hypotension and reduction in peripheral vascular resistance.

A general toxicity and cardiovascular telemetry study of single escalating oral doses of iloperidone (0, 5, and 15 mg/kg on Days 1, 4, and 7, respectively) was conducted in conscious dogs (2/sex/group) (Study 0270088). Evaluations included general toxicity endpoints and cardiovascular data (mean arterial, systolic and diastolic blood pressure, heart rate and electrocardiographic) and body temperature collected using implanted transmitters. Measurements were recorded for approximately 1 minute every 5 minutes on a continuous basis for the duration of the study. The following data points were analyzed after each dose from the 1-minute segment of data: 1, 2, 4, and 6 hours post

dose. Pharmacologically related clinical signs were observed at  $\geq 5$  mg/kg (ptosis, lacrimation, ataxia, decreased motor activity); additional signs noted at 15 mg/kg were emesis, lateral recumbency, aggression, hypersensitivity, and impairment of the righting reflex. There were no drug-related changes in body weight or food consumption. There were no changes in body temperature or ECG (as shown in the sponsor's table below).

Mean electrocardiographic data					
Time interval	HR	p wave	PR interval	QRS complex	QT interval
	bpm	duration	Duration	Duration	Duration <sup>1</sup>
Control					
1	102 ±29	0.04 ±0.01	0.10 ±0.01	0.05 ±0.01	220 ±16
2	95 ±30	0.04 ±0.00	0.10 ±0.01	0.04 ±0.01	205 ±10
4	111 ±28	0.04 ±0.00	0.10 ±0.01	0.04 ±0.01	200 ±16
6	126 ±39	0.04 ±0.01	0.10 ±0.01	0.04 ±0.00	175 ±19
5 mg/kg					
1	150 ±36	0.04 ±0.01	0.08 ±0.00	0.04 ±0.00	190 ±12
2	115 ±20	0.04 ±0.01	0.08 ±0.01	0.04 ±0.01	220 ±16
4	110 ±29	0.04 ±0.01	0.09 ±0.01	0.04 ±0.01	223 ±21
6	136 ±38	0.03 ±0.00	0.08 ±0.01	0.04 ±0.01	205 ±19
15 mg/kg					
1	137 ±27	0.03 ±0.01	0.08 ±0.01	0.04 ±0.01	190 ±24
2	142 ±16	0.03 ±0.01	0.08 ±0.01	0.04 ±0.01	205 ±10
4	154 ±66	0.04 ±0.01	0.08 ±0.01	0.04 ±0.00	203 ±33
6	137 ±62	0.04 ±0.01	0.08 ±0.00	0.04 ±0.01	200 ±28

<sup>1</sup> milliseconds

Summary data of heart rate and blood pressure (arterial, systolic and diastolic) are presented in the following sponsor's tables:

Mean heart rate (BPM)			
Time (hour)	Vehicle	5 mg/kg	15 mg/kg
1	107 ± 34	151 ± 37	135 ± 17
2	111 ± 32	112 ± 23	147 ± 8
4	108 ± 20	105 ± 33	148 ± 52
6	146 ± 63	128 ± 41	130 ± 55

Mean arterial blood pressure (mmHg)			
Time (hour)	Vehicle	5 mg/kg	15 mg/kg
1	87 ± 14	72 ± 12	75 ± 14
2	92 ± 15	69 ± 9	65 ± 17
4	89 ± 11	69 ± 12	57 ± 22
6	99 ± 25	71 ± 6	61 ± 13

Mean systolic blood pressure (mmHg)			
Time (hour)	Vehicle	5 mg/kg	15 mg/kg
1	120 ± 14	91 ± 18	100 ± 17
2	132 ± 21	97 ± 8	88 ± 18
4	122 ± 16	97 ± 11	80 ± 28
6	133 ± 31	98 ± 13	89 ± 20

Mean diastolic blood pressure (mmHg)			
Time (hour)	Vehicle	5 mg/kg	15 mg/kg
1	64 ± 11	52 ± 10	59 ± 12
2	71 ± 14	53 ± 9	50 ± 15
4	70 ± 9	53 ± 14	41 ± 20
6	76 ± 20	56 ± 3	47 ± 8

A decrease in all blood pressure parameters was noted at 5 and 15 mg/kg and a slight and transient increase in heart rate was noted at 15 mg/kg. There was considerable individual animal variation in both heart rate and blood pressure measurements. The overall trend, when comparing vehicle and iloperidone administration, was a decrease in diastolic, systolic, and mean blood pressures at 5 and 15 mg/kg and a transient increase in heart rate at 15 mg/kg, with no effects on body temperature or ECG.

Effects of iloperidone metabolites on hemodynamic parameters

Hemodynamic parameters were evaluated in rats and dogs following administration of iloperidone metabolites P88 and P89 (Study ILO-MET-001). The results are summarized in the following sponsor's table:

Effects of P88-8991 and P89-9124 on Cardiovascular Physiology				
	<u>P88-8991 (mg/kg)</u>	<u>P89-9124 (mg/kg)</u>		
<b>Direct Blood Pressure Measurement in Conscious Rats</b>	<b>Δ systolic pressure: -18 mmHg @ 1, p.o.</b>	<b>Δ systolic pressure: -14 mmHg @ 3, p.o.</b>		
<b>Hemodynamic Evaluation in Anesthetized Dogs</b>	<b>Not tested</b>	<b>2, i.d.</b>	<b>2.0, i.v.</b>	<b>0.5, i.v.</b>
	MAP	-17%	-34%	-35%
	TPR	-16%	-35%	-49%
	HR	-14%	-25%	NC
	dp/dt <sub>max</sub>	-13%	-19%	+37%
	LVEDP	NC	NC	NC
	CO	-21%	-22%	+33%
	Lead II EKG	1/3	NC	NC
		LBBB*		

\* = left bundle branch block

Iloperidone metabolites P88 and P89 administered to conscious rats at single oral doses of 1 and 3 mg/kg, respectively, produced decreases in systolic blood pressures of 18 and 14 mm Hg, respectively. The magnitude of this effect was similar to that seen in hemodynamic studies conducted with the parent compound in rats.

P89 was also evaluated for hemodynamic changes in anesthetized dogs. At 2 mg/kg administered i.d., mean arterial blood pressure (MAP) was decreased by 17%, with a 16% decrease in total peripheral resistance (TPR) and a 14% decrease in heart rate (HR). Left ventricular end diastolic pressure (LVEDP) was unchanged and left ventricular rate of pressure development (dP/dt<sub>max</sub>) was decreased by 13%. Cardiac output was decreased by 21% and one third of dogs had a left bundle-branch block. At 2 mg/kg i.v.,

P89 caused a 34% decrease in MAP, a 35% decrease in TPR, and a 25% decrease in HR. The LVEDP was unchanged and dp/dtmax was decreased by 19%. Cardiac output was reduced by 22%. At 0.5 mg/kg i.v., P89 caused a 35% decrease in MAP and a 49% decrease in TPR, with no change in HR. LVEDP was unchanged, dp/dtmax was increased by 37%, and cardiac output was increased by 33%. No lead II electrocardiogram changes were seen (Study ILO-MET-001).

Overall, the effects of P88 and P89 metabolites on hemodynamic parameters in anaesthetized dogs, including a hypotensive effect, were similar to those observed following iloperidone administration.

Pulmonary effects:

A single-dose study was conducted to evaluate the pharmacological effects of iloperidone (at doses of 0.1, 1, and 10 mg/kg) and its metabolite P95 (at doses of 1, 10, and 100 mg/kg) on the respiratory system following oral (gavage) administration to male albino rats (Sprague-Dawley CDC — CD(SD)BR, N = 6 per group). For respiratory testing, animals were placed in head-out plethysmographs, and ventilatory parameters (tidal volume, respiratory rate, and derived minute volume) were measured for 15-minute periods at the following time points: predose, 1, 2, and 6 hours post-dose (Study 93279). Clinical signs included ptosis, tremors, and hypoactivity in the 10-mg/kg iloperidone group, and ptosis in the 10- and 100-mg/kg P95 groups. Body weights were not recorded, except for the animal group allocation at the start of the study. The results are summarized in the sponsor's tables on this and next page.

No adverse treatment-related effects on respiratory function were observed following exposure to either iloperidone or P95. Minor decreases in respiratory minute volumes were observed in the animals treated with 1 and 10 mg/kg iloperidone and in all P95-treated groups, but none of the changes reached statistical significance and were not considered to be of toxicological significance. All tidal volumes, respiratory rates, and derived minute volumes values were considered to be within the expected range for healthy control animals of the tested species and strain.

Iloperidone: Group mean respiratory function measurements in rats

	PRE DOSE			1H POST DOSE		
	TV ML	RR BRS/ MIN.	MV ML/ MIN.	TV ML	RR BRS/ MIN.	MV ML/ MIN.
GROUP 1 - VEHICLE CONTROL	1.27 .246	216.5 28.08	276.0 62.02	1.05 .158	199.3 30.38	204.8 21.60
GROUP 2 - ILO522 0.1 mg/kg	1.41 .246	202.7 37.74	278.0 47.42	1.24 .220	198.5 26.24	247.2 55.44
GROUP 3 - ILO522 1.0 mg/kg	1.00 .232	217.7 21.12	216.2 57.97	1.05 .226	212.5 39.62	225.2 70.12
GROUP 4 - ILO522 10.0 mg/kg	1.19 .393	213.2 32.74	248.8 66.51	1.24 .377	168.2 20.35	207.7 62.10

b(4)

Iloperidone: Group mean respiratory function measurements in rats (Continued)

	2H POST DOSE			6H POST DOSE		
	TV ML	RR BRS/ MIN.	MV ML/ MIN.	TV ML	RR BRS/ MIN.	MV ML/ MIN.
GROUP 1 - VEHICLE CONTROL	.97 .170	197.2 11.79	189.7 31.33	1.16 .225	213.8 30.58	243.7 42.91
GROUP 2 - ILO522 0.1 mg/kg	1.24 .184	198.7 27.30	248.8 62.04	1.32 .225	198.8 23.86	258.5 34.53
GROUP 3 - ILO522 1.0 mg/kg	1.08 .147	212.8 14.84	228.7 37.99	1.08 .176	216.0 31.39	232.3 49.02
GROUP 4 - ILO522 10.0 mg/kg	1.26 .365	164.8 51.84	222.8 110.64	1.14 .256	193.3 32.08	219.3 52.44

Iloperidone metabolite P95: Group mean respiratory function measurements in rats

	PRE DOSE			1H POST DOSE		
	TV ML	RR BRS/ MIN.	MV ML/ MIN.	TV ML	RR BRS/ MIN.	MV ML/ MIN.
GROUP 5 - P95-12113 1.0 mg/kg	1.33 .180	204.6 12.16	271.6 28.08	1.22 .213	196.2 10.34	239.7 44.34
GROUP 6 - P95-12113 10.0 mg/kg	1.21 .229	214.3 17.72	259.0 48.53	1.21 .183	212.0 22.85	254.2 40.56
GROUP 7 - P95-12113 100.0 mg/kg	1.41 .212	197.0 14.21	278.2 44.65	1.41 .417	185.7 32.04	257.0 83.30

	2H POST DOSE			6H POST DOSE		
	TV ML	RR BRS/ MIN.	MV ML/ MIN.	TV ML	RR BRS/ MIN.	MV ML/ MIN.
GROUP 5 - P95-12113 1.0 mg/kg	1.25 .453	207.2 17.84	255.5 86.80	1.10 .346	199.8 10.94	216.5 64.16
GROUP 6 - P95-12113 10.0 mg/kg	1.36 .219	203.0 36.22	269.7 45.48	1.08 .112	204.3 22.86	221.0 36.70
GROUP 7 - P95-12113 100.0 mg/kg	1.22 .215	204.2 51.57	245.2 61.50	1.34 .257	210.7 15.32	282.3 65.34

Renal effects: No data

Gastrointestinal effects: No data

Abuse liability: No data

### 2.6.2.5 Pharmacodynamic drug interactions

No pharmacodynamic drug interaction study in animal models in vivo was conducted with either iloperidone or its metabolites. Three in vitro studies were performed to investigate induction and inhibition activities of iloperidone and its metabolites on human cytochrome P450 system. The studies are adequately summarized by the sponsor as follows.

#### In vitro assessment of cytochrome P450 inhibition by iloperidone and metabolites P95 and P88

The potential of iloperidone and its metabolites P95 and P88 to inhibit human cytochrome P450 (CYP) enzymes was assessed in two studies using pooled human liver microsomes and several probe substrates, metabolism of which is known to be CYP isoform-selective.

“In the initial study (Study DMPK[US]R00-2173), the human liver microsomes were pre-incubated with the nicotinamide adenine dinucleotide phosphate-oxidase (NADPH)-generating system in the absence of the marker system. It was determined that in some cases, some loss of activity of the enzyme tested was observed under these conditions regardless of the presence of the test article(s). Due to the possibility that iloperidone, P88, and P95 may bind to microsomal protein or lipids, an attempt was made to design a second study such that, in as many cases as possible, the microsomal protein, incubation time, and phosphate buffer concentration were 0.1 mg/mL, 5 minutes and 50 mM, respectively, for assays performed with human liver microsomes.”

“Under the design of a second study (Study XT065041), iloperidone was found to directly inhibit CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5. Approximately 22%, 48%, 45%, 60%, 86%, 96% and 92% inhibition, respectively, were observed at the highest concentration of iloperidone examined (100 μM). The IC50 values for CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5 were >100 μM, >100 μM, >100 μM, 57 μM, 11 μM, 2.1 μM and 8.5 μM, respectively. Evidence of time-dependent inhibition of CYP2D6 and CYP3A4/5 by iloperidone was observed, as an increase in inhibition was observed upon preincubation.

According to the September 2006 Draft Guidance “Drug Interaction Studies – Study Design, Data Analysis, and Implications for Dosing and Labeling,” the  $[I]/K_i$  ratio from in vitro studies can be used to predict the likelihood of inhibitory drug interactions clinically.  $[I]$  is defined as the steady-state Cmax concentration at the recommended therapeutic dose, and  $K_i$  is defined as one-half the IC50 observed in the in vitro study ( $IC_{50} * 0.5$ ). The following table shows the predicted clinical relevance of these ratios.

Prediction of Clinical Relevance of Competitive CYP Inhibition

$[I]/K_i$	Prediction
$[I]/K_i > 1$	Likely
$1 > [I]/K_i > 0.1$	Possible
$0.1 > [I]/K_i$	Remote

Taken directly from the September 2006 Draft Guidance “Drug Interaction Studies – Study Design, Data Analysis, and Implications for Dosing and Labeling.”

$[I]$  = steady-state Cmax at the recommended therapeutic dose;  $K_i$  = inhibitory constant.

The maximum recommended dose for iloperidone is 24 mg/d given twice daily (12 mg BID). Iloperidone has a molecular weight of 426.5 g/mol. According to results obtained in Study CILO522A2328, the steady state iloperidone Cmax is 19.33 ng/ml, which would correspond to molar exposure of 45 nM. Based on this information, the  $[I]/K_i$  ratios can

be calculated (see next table). These data predict a remote possibility of clinically relevant CYP inhibition by iloperidone.

[I]/K<sub>i</sub> Ratios

	CYP1A2	CYP2C8	CYP2C9	CYP2C19	CYP2D6	CYP3A4	CYP3A5
[I]	45 nM	45 nM	45 nM	45 nM	45 nM	45 nM	45 nM
K <sub>i</sub>	50 μM	50 μM	50 μM	29 μM	5.5 μM	1.1 μM	4.3 μM
[I]/K <sub>i</sub>	0.0009	0.0009	0.009	0.002	0.008	0.04	0.01

[I] = steady-state C<sub>max</sub> at the recommended therapeutic dose; K<sub>i</sub> = inhibitory constant.

In the second study, P88 directly inhibited CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5, with approximately 25%, 24%, 76%, 43% and 30% inhibition, respectively, observed at the highest concentration of P88 examined (100 μM). The IC<sub>50</sub> values for these enzymes were >100 μM, >100 μM, 35 μM, >100 μM and >100 μM, respectively. Evidence of time-dependent inhibition of CYP2D6 and CYP3A4/5 by P88 was also observed. P95, in the second study, had little or no potential to cause no direct inhibition of CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5, and the IC<sub>50</sub> values for these enzymes were all >100 μM. There was little or no evidence of time-dependent inhibition of CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5 by P95.

The above study results demonstrated that iloperidone and its metabolites inhibited various enzymes in P450 system at the highest concentration tested (100 μM). However, normal therapeutic levels of iloperidone are expected to be in the range of 45 nM, >2000-fold less than what was used in the in vitro studies. Based on this finding, and the [I]/K<sub>i</sub> ratio data presented in the table above, it is concluded that iloperidone does not inhibit CYP1A2, CYP2C8, CYP2C9 and CYP2C19, but has the potential to inhibit CYP2D6 and CYP3A4/5 in humans.” (End citation)

#### In vitro evaluation of iloperidone, P88 and P95 as inducers of cytochrome P450 expression

“The effects of iloperidone and its metabolites, P95 and P88, on the expression of cytochrome P450 (CYP) enzymes were assessed in primary cultures of human hepatocytes (Study XT063049). Cultures were treated once daily for 3 consecutive days with iloperidone, P88, or P95 at 0.05, 1.0, or 20 μM in DMSO, or one of 2 known CYP inducers, omeprazole and rifampin. Cells were then harvested to prepare microsomes for analysis of phenacetin O dealkylation (marker for CYP1A2), amodiaquine N dealkylation (marker for CYP2C8), diclofenac 4' hydroxylation (marker for CYP2C9), S mephenytoin 4'-hydroxylation (marker for CYP2C19), and testosterone 6β hydroxylation (marker for CYP3A4/5).

Treatment with iloperidone caused a small increase (2-fold or less) in CYP1A2 and CYP2C8 activities, and a decrease in CYP2C9, CYP2C19 and CYP3A4/5 activity at high concentrations of Iloperidone. At 20 μM, Iloperidone caused approximately a 30%, 47% and 79% decrease in CYP2C9, CYP2C19 and CYP3A4/5 activity, respectively when compared with the vehicle control, and the decrease was statistically significant for CYP3A4/5 activity.

Treatment of hepatocytes with P88 caused little or no change (2-fold or less on average) in CYP1A2 and CYP2C8 activity, but caused a decrease in CYP2C9, CYP2C19 and CYP3A4/5 activity at the highest concentrations tested (20 μM). At 20 μM, P88 caused

approximately a 19%, 45% and 80% decrease in CYP2C9, CYP2C19 and CYP3A4/5 activity, respectively; and the decrease in CYP3A4/5 activity was statistically significant when compared with the vehicle control.

Treatment of hepatocytes with P95 had little or no effect on the activity of any of the CYP enzymes examined (CYP1A2, CYP2C8, CYP2C9, CYP2C19, or CYP3A4/5).

The results of this study show that iloperidone, P88 and P95 did not induce the enzymes examined. Iloperidone and P88 may inhibit the activity of CYP2C9, CYP2C19, or CYP3A4/5. P95 had little or no effect on the activity of any of the enzymes examined in this study. The results of Study XT03049 agree with the results obtained from Study XT065041". (End citation)

#### **2.6.2.6 Discussion and conclusions**

Iloperidone exhibits a nonclinical pharmacological profile consistent with that of an atypical antipsychotic, characterized by a relatively lower affinity for dopamine D2 receptors and high affinity for serotonin 5-HT<sub>2</sub> receptors. The pharmacological profile of iloperidone was evaluated using in vitro radioligand-binding assays in rat, bovine, and guinea pig tissues and cell lines stably expressing human receptor subtypes, ex vivo binding assays in the rat, functional characterization assays, and behavioral assays in mice, rats, and dogs. Based on these studies, iloperidone was demonstrated to have high affinity for 5-HT<sub>2A</sub>, adrenergic  $\alpha$ <sub>1</sub>, adrenergic  $\alpha$ <sub>2</sub>, D<sub>2</sub>, D<sub>3</sub>, and 5-HT<sub>1A</sub> receptors in humans, and was found to act as an antagonist at selected dopaminergic, serotonergic, and noradrenergic receptor subtypes. Affinity was highest for 5-HT<sub>2</sub> and adrenergic  $\alpha$ <sub>1</sub> receptors, and lower for D<sub>2</sub>, which is an affinity profile consistent with a potential for iloperidone to act as an atypical antipsychotic. The respective ratios of IC<sub>50</sub> values for D<sub>2</sub>/5-HT<sub>2A</sub> for iloperidone and clozapine were 10 and 23, whereas the ratios for haloperidol and thioridazine were 0.2 and 1.2, respectively. These ratios indicate a greater relative affinity for 5-HT<sub>2A</sub> receptors for iloperidone compared with typical antipsychotic agents such as haloperidol. This profile, which is similar to the atypical antipsychotic clozapine, suggests that iloperidone may have reduced potential for extrapyramidal side effects compared with typical antipsychotics. Behavioral studies of iloperidone in animals support its antipsychotic potential with regard to positive and negative symptoms, and social withdrawal symptoms of schizophrenia, as well as potential anxiolytic activity at the same doses that produce antipsychotic effects. Behavioral findings also suggested that iloperidone, like clozapine, is less likely to be associated with extrapyramidal symptoms usually seen with dopamine-receptor antagonists such as haloperidol.

Additional pharmacological characterization indicated that iloperidone lacks anticholinergic activity.

Iloperidone metabolites P88 and P89 were found to have a profile similar to that of iloperidone in receptor-binding studies, with potential to exert CNS effects mediated by dopaminergic, serotonergic, and noradrenergic antagonism. P95 had affinity for serotonergic,  $\alpha$ <sub>1</sub> and  $\alpha$ <sub>2</sub> adrenergic receptors, as well as dopaminergic receptors. Although P95 does not cross the blood-brain barrier as shown by whole-body autoradiography, there is potential for P95 to exert clinical effects relevant to safety through interaction with various receptor subtypes.

The receptor-binding and antagonism profile of iloperidone and its metabolites, namely high affinity for  $\alpha$ 1- adrenergic receptors in peripheral vascular tissues, indicate that iloperidone and its metabolites P88, P89, and P95 are likely to exert cardiovascular effects, such as postural hypotension.

In vitro evaluation of iloperidone effects in isolated dog Purkinje fibers indicated that iloperidone has the capacity to prolong action potential duration. Iloperidone was also found to produce a rapid and reversible block of hERG currents in mammalian cells expressing the cloned hERG. This indicates that iloperidone has the capacity to prolong QTc interval duration. Iloperidone metabolite P88, but not P95, also exhibited this potential. In hemodynamic evaluations conducted in conscious normotensive and hypertensive rats and conscious and anesthetized dogs, iloperidone was found to dose-dependently decrease blood pressure due to its effects at  $\alpha$ 1-adrenergic receptors. Small and/or transient increases in heart rate were also noted in some studies. Cardiac output and ECG parameters were not affected by iloperidone in these studies. The fall in arterial pressure seen with iloperidone was gradual in onset with a slow i.v. infusion and steep with a bolus i.v. injection, suggesting that hypotensive effects of iloperidone could be altered by the method of administration.

Neither iloperidone nor its metabolite P95 was associated with any adverse respiratory effects as evaluated in albino rats.

In vitro studies showed that iloperidone has inhibitory, but not inducing activity in the CYP enzyme system. Based on the steady-state Cmax at the recommended therapeutic dose in humans (12 mg BID), it is not likely that iloperidone has the potential to inhibit CYP1A2, CYP2C8, CYP2C9 and CYP2C19 at therapeutic doses in humans, but it has the potential to inhibit CYP2D6 and CYP3A4/5 in humans.

In conclusion, the pharmacological profile of iloperidone is consistent with that of an atypical antipsychotic with a reduced potential for extrapyramidal side effects as compared to haloperidol and therapeutic potential with regard to positive and negative symptoms, and social withdrawal symptoms of schizophrenia, including anxiolytic properties. Iloperidone has the potential to induce hypotensive effects and to prolong QTc interval duration.

**APPEARS THIS WAY ON ORIGINAL**

## 2.6.3 PHARMACOLOGY TABULATED SUMMARY

### Safety Pharmacology

				Test Article: Iloperidone			
Organ Systems Evaluated	Species/ Strain	Method of Admin.	Iloperidone Concentration/ Doses <sup>a</sup>	Gender and No. per Group	Noteworthy Findings	GLP Compliance	Study Number
Cardiovascular	In vitro	NA	0.01-10 $\mu$ M	NA	<ul style="list-style-type: none"> <li>In dog Purkinje fibers paced at stimulation frequencies of 0.5 and 1 Hz, iloperidone and its metabolite (-)P88 prolonged action potential duration at a concentration of 0.1 <math>\mu</math>M.</li> <li>Iloperidone and its metabolite (-)P88 at free plasma concentrations <math>\geq</math>0.1 <math>\mu</math>M are likely to have direct effects on the QRS complex, QT duration and cardiac conduction.</li> <li>P95 appears to be less likely to have direct effects on QRS complex or QT duration.</li> <li>No other effects on cardiac action potential parameters were noted for any test item.</li> </ul>	Yes	0120065
Cardiovascular	In vitro	NA	5-30,000 nM	NA	<ul style="list-style-type: none"> <li>Iloperidone and metabolites (P95, P88) produced rapid, reversible block of hERG current suggesting the potential for effects on QT duration.</li> <li>Assessment of test article block at near physiological temperature increased the IC<sub>50</sub> values for all iloperidone, P95, P88, and ziprasidone, but not risperidone.</li> </ul>	Yes	008167
Cardiovascular	Rat/ Long Evans	p.o.	10 mg/kg	Not specified/ 4	<ul style="list-style-type: none"> <li>Overall, iloperidone did not elicit a hypotensive response or an effect on HR, except 1 of 4 normotensive rats had a 41% decrease in MAP at 90 min.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-001
Cardiovascular	Rat/ Long Evans	p.o.	3 mg/kg	M/ Iloperidone n = 3 Clozapine n = 4	<ul style="list-style-type: none"> <li>Iloperidone produced a reduction in MAP throughout 240 min with a maximal decrease of 13% (15 mmHg) in MAP at 180 min post-dose and decrease of 5% in HR in normotensive rats.</li> <li>Clozapine increased MAP by 5 mmHg and decreased HR by 18 bpm.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-002

				Test Article: Iloperidone			
Organ Systems Evaluated	Species/ Strain	Method of Admin.	Iloperidone Concentration/ Doses <sup>a</sup>	Gender and No. per Group	Noteworthy Findings	GLP Compliance	Study Number
Cardiovascular	Rat/ SHR	p.o.	1 and 3 mg/kg	Not specified/ Iloperidone n = 4 Clozapine n = 3	<ul style="list-style-type: none"> <li>Iloperidone decreased arterial systolic pressure by 11% and 13% and decreased HR by 4% and 3% in hypertensive rats at 1 mg/kg and 3 mg/kg dose, respectively.</li> <li>Clozapine did not decrease arterial pressure or HR.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-003
Cardiovascular	Rat/ SHR	p.o.	0.3-20 mg/kg	Not specified/ n = 4 for all doses except n = 6 for 20 mg/kg	<ul style="list-style-type: none"> <li>Overall results showed that at 0.3 and 1 mg/kg had no effect on basal BP or HR, although a 48% inhibition of hypertensive response to phenylephrine occurred at 1 mg/kg dose.</li> <li>Iloperidone (6 and 20 mg/kg) lowered basal BP by 45% and inhibited the phenylephrine effect on BP by 89% (ED<sub>50</sub> = 0.6 mg/kg p.o.).</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-004
Cardiovascular	Dog/ not specified	p.o.	2 or 5 mg/kg	Not specified/ Iloperidone 2 mg/kg n = 3; 5 mg/kg n = 4; Clozapine n = 3	<ul style="list-style-type: none"> <li>Iloperidone caused sustained decrease in MAP with peak of 24% and 22% at 2 and 5 mg p.o., respectively, in conscious dogs.</li> <li>Clozapine caused peak decrease of 16% at 5 mg/kg.</li> <li>Iloperidone and clozapine produced similar vasodepressor effects in conscious dogs.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-006
Cardiovascular	Dog/ Beagle	i.v., bolus (15 sec) or slow infusion (30 min)	0.02-1 mg/kg	M+F/ Bolus n = 4 Infusion n = 3	<ul style="list-style-type: none"> <li>Iloperidone dose-dependently lowered systemic arterial pressure in anesthetized dogs.</li> <li>Hypotensive response (39%) was due to a reduction in peripheral resistance (40%) mediated by vascular α<sub>1</sub>-receptor blockade.</li> <li>HR, cardiac output, ECG not affected.</li> <li>The hypertensive effects produced by bolus or slow infusion were of similar magnitude and subsided within 2 hours of dosing.</li> <li>The fall in arterial pressure was gradual in onset with the slow infusion (peak reduction 30 min) in contrast to the rapid hypotensive effect caused by the bolus injection (peak reduction 5 min), suggesting that humans may tolerate the gradual onset of hypotension associated with slow infusion.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-005

				Test Article: Iloperidone			
Organ Systems Evaluated	Species/ Strain	Method of Admin.	Iloperidone Concentration/ Doses <sup>a</sup>	Gender and No. per Group	Noteworthy Findings	GLP Compliance	Study Number
Cardiovascular	Dog/ Beagle	i.v., bolus (15 sec) or slow infusion (30 min)	0.02 mg/kg	Gender not stated/ Bolus n = 4 Infusion n = 3	<ul style="list-style-type: none"> <li>Associated with Study ILO-SP-005 to correlate plasma levels of iloperidone and P94-11840 with cardiovascular hemodynamic parameters in anesthetized dogs dosed with iloperidone.</li> <li>Iloperidone plasma levels after bolus injection of 0.02 mg/kg consistently higher than levels for iloperidone following slow infusion of 0.02 mg/kg.</li> <li>Plasma levels for iloperidone after bolus injection ranged from 5 to 96 ng/mL, with average C<sub>max</sub> = 63 ng/mL ± 24 ng/mL.</li> <li>Plasma levels after slow infusion ranged from 5 to 24 ng/mL, with average C<sub>max</sub> = 15 ng/mL ± 8 ng/mL.</li> <li>Plasma level of P94-11840 was essentially below the limits of detection.</li> <li>Results agree with hemodynamic effects noted in the cardiovascular study.</li> </ul>	Indeterminate (not required) <sup>b</sup>	RCE118-DOC
Cardiovascular	Dog/ Beagle	i.d.	2 or 10 mg/kg	M+F/ n = 3	<ul style="list-style-type: none"> <li>Iloperidone i.d. caused similar hypotensive and vasodilator effects to clozapine 5 mg/kg, i.d. in anesthetized dogs.</li> <li>Iloperidone exerted less influence on cardiac output and HR than clozapine.</li> <li>Iloperidone exhibited + inotropic effects, while clozapine showed - inotropic effects, evidenced by progressive reduction in left ventricular rate of pressure development.</li> <li>No drug-related ECG changes noted.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-007
Cardiovascular	Dog/ Beagle	i.d.	0.5-10 mg/kg (dose-finding study for ILO-SP-009)	F/ 0.5 mg/kg n = 4; 1 mg/kg n = 3; 10 mg/kg n = 3	<ul style="list-style-type: none"> <li>A dose of 1 mg/kg (i.d.) produced measurable hypotension and reduction in peripheral vascular resistance.</li> <li>Time of onset of the peak hemodynamic responses occurred in a dose-related manner.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-008

				Test Article: Iloperidone			
Organ Systems Evaluated	Species/ Strain	Method of Admia.	Iloperidone Concentration/ Doses <sup>a</sup>	Gender and No. per Group	Noteworthy Findings	GLP Compliance	Study Number
Cardiovascular	Dog	p.o., i.d.	0.25, 0.25, 0.5 mg/kg for 3 days p.o. followed by acute dose 0.5-10 mg/kg i.d.	F/ 0.5 mg/kg n = 3; 1 mg/kg n = 4; 5 mg/kg n = 4; 10 mg/kg n = 2	<ul style="list-style-type: none"> <li>After 3 days of iloperidone p.o. dosing, 0.5 and 10 mg/kg (i.d.) produced similar hypotensive effects and decreases in peripheral resistance as in dogs given the same doses acutely without pre-treatment.</li> <li>Only 2/4 dogs receiving 1 mg/kg i.d. showed hypotension and vasodilation, indicating that cumulative dosing slightly altered cardiovascular effects.</li> </ul>	Indeterminate (not required) <sup>b</sup>	ILO-SP-009
Cardiovascular	Dog/ Beagle	p.o.	0, 5, and 15 mg/kg	M+F/ 2/sex/dose	<ul style="list-style-type: none"> <li>At single doses of 5 and 15 mg/kg, no iloperidone-related changes in body temperature and ECG were noted.</li> <li>A decrease in mean BP occurred at doses of 5 mg and 15 mg and a slight and transient increase in HR at 15 mg/kg was noted.</li> </ul>	Yes	0270088
Respiratory	Rat/ Albino	p.o.	Iloperidone 0.1, 1, and 10 mg/kg P95 1, 10, and 100 mg/kg	M/6/dose	<ul style="list-style-type: none"> <li>No adverse treatment-related effects of iloperidone or P95 on respiratory function.</li> </ul>	Indeterminate (not required) <sup>b</sup>	93279/ 018010

				Test Article: Iloperidone			
Organ Systems Evaluated	Species/ Strain	Method of Admia.	Iloperidone Concentration/ Doses <sup>a</sup>	Gender and No. per Group	Noteworthy Findings	GLP Compliance	Study Number
CNS/Whole body	Mouse	p.o.	P95 10, 30, 100, and 300 mg/kg	M/10/dose	<ul style="list-style-type: none"> <li>P95 (iloperidone metabolite) treatment resulted in alterations in general and neurobehavioral activity at all doses, which were considered as exaggerated pharmacologic responses.</li> </ul>	Yes	0170079

<sup>a</sup> Single dose unless specified otherwise.

<sup>b</sup> GLP statement is not included in the study report.

BP = blood pressure; ECG = electrocardiogram; ED<sub>50</sub> = median effective dose; F = female; hERG = human ether-a-go-go related gene; HR = heart rate; i.d. = intraduodenal; i.v. = intravenous; M = male; MAP = mean arterial pressure; NA = not applicable; p.o. = oral; SHR = spontaneously hypertensive rat.

## 2.6.4 PHARMACOKINETICS/TOXICOKINETICS

### 2.6.4.1 Brief summary

The preclinical pharmacokinetic studies performed with iloperidone include analytical methods development and ADME studies of iloperidone and its metabolites P88-8991 (referenced as P88) and P95-12113 (referenced as P95).

Several analytical methods were developed and validated for the determination of iloperidone and its metabolites, P88 and P95 in plasma samples. An additional method was developed for the detection and separation of *S*-P88 and *R*-P88.

ADME studies were conducted *in vitro* and *in vivo* with mice (CD-1), rats (Sprague-Dawley, Han Wistar, Lister Hooded, Long Evans, Wistar-Hannover, Long Evans Hooded), rabbits (New Zealand white), dogs (Beagle), and monkeys (Cynomolgus), and evaluated oral or intravenous administration of iloperidone following single and multiple doses for up to 14 days, ranging from 1 mg/kg to 20 mg/kg.

*In vivo* absorption studies demonstrated that in all species treated with a single *i.v.* dose, the maximum plasma concentration was observed at the first blood sampling, at approximately 0.1 h. post-dose. Following oral administration (either single- or multiple-dose), the time to reach  $C_{max}$  was 0.5 to 1 h. in the mouse and dog, 1 h. in the rabbit and monkey, and 1 to 2 hrs. in the rat. Oral bioavailability was <1% in rat, 5% in mouse, and 19% in both rabbit and dog after a single oral dose of 5 mg/kg. The low bioavailability was likely due to first-pass effect in the species tested. In humans, iloperidone is readily absorbed ( $t_{max}$  is 2-3 h after a single dose and about 1.5 h with multiple dosing); absolute bioavailability is estimated to be approximately 36% because of a first-pass effect.

Plasma exposure values ( $C_{max}$  and AUC) upon *i.v.* or oral administration were higher in dogs than in other species at equal dose levels. In rats orally dosed with iloperidone (5, 16 and 20 mg/kg), the  $C_{max}$  and AUC levels increased over-proportionally with dose elevation. This finding was possibly related to inhibitory activity of iloperidone to cytochrome P450 (CYP) enzymes. The mean plasma exposure (AUC) in the female rats was greater than that in the males, indicating a gender difference in PK profile. In humans, the pharmacokinetics of iloperidone is considered dose proportional in the dose range studied, *i.e.* from 2 to 12 mg *b.i.d.* (4 to 24 mg daily).

The *in vitro* protein-binding of iloperidone in rats, dogs, and humans was greater than 90%, 86%, and 93%, respectively. The protein-binding activity of iloperidone in humans appears not to be changed by the appearance of its metabolites, as shown in an *ex vivo* study in humans where the protein-binding values were similar at 3 h. and 24 h. after a single oral dose of 3 mg iloperidone.

Iloperidone organ distribution upon either oral or *i.v.* administration to mice, rats, rabbits, and dogs, was rapid; the highest drug concentration was generally observed in the liver, kidney, gastrointestinal system, and secretory glandular tissues; placental transfer was limited; and drug concentration in the brain was very low. After oral administration to lactating rats (5 mg/kg),  $C_{max}$  levels in the plasma and milk were reached in 0.5 and 4 h., respectively; iloperidone concentration was approximately 10 times higher in the milk than in plasma at 4 hours post dosing.

The metabolism of iloperidone was evaluated in mice, rats, rabbits, dogs, and humans. *In vitro*, iloperidone was metabolized in human liver microsomes to primarily 4 products:

P22 (*N*-dealkylation), P94 (hydroxylation), P89 (*O*-demethylation), and P88 (carbonyl reduction). The main metabolites were P88, P89, P94, and P95. In vivo metabolic studies in mice, rats, rabbits, and dogs showed that iloperidone was extensively metabolized in all species tested, with numerous metabolites (including those found in humans) identified in the plasma and excreta. Similarly, in humans, there are multiple metabolic pathways for iloperidone as shown in a clinical ADME study (ILO522 2301) using [<sup>14</sup>C]-iloperidone. Of practical importance in humans are the CYP2D6 pathway, which leads to formation of the most abundant metabolite in systemic circulation, P95; a reduction pathway leading to the formation of the second most abundant metabolite, P88; and the CYP3A4 pathway, which produces metabolite P89 and probably other metabolites that are present in circulating blood in low quantities. The total systemic exposure (AUC) to P95 in humans is about 2.5 times that of iloperidone; however, as this metabolite does not cross the blood-brain barrier (as shown in the rat), it is believed to have no direct CNS activity. The total exposure to P88 in humans is about 1.5 times that of iloperidone; it crosses the blood-brain barrier and has a similar receptor binding profile as iloperidone. Therefore, P88 is considered to be an active metabolite. The most abundant metabolites in humans are found in the species used in toxicology studies.

Iloperidone and its metabolites had very little or no potential to induce hepatic CYP enzymes both in vitro and in vivo. In vitro, iloperidone and its metabolites, P88 and P95 had potentials to inhibit the activities of CYP enzymes. However, iloperidone concentrations (IC<sub>50</sub>) that inhibited the activity of CYP1A2, CYP2C8, CYP2C9 or CYP2C19 in the in vitro studies were > 100 µM, much higher than human plasma C<sub>max</sub> (19.33 ng/ml) at the maximum recommended human dose (24 mg/day), which corresponds to molar exposure of 45 nM. On the other hand, the IC<sub>50</sub> for CYP2D6 and CYP3A4/5 were 11, 2.1, and 8.5 µM, respectively. Therefore, it is unlikely that at MRHD iloperidone and its metabolites can inhibit the activity of CYP1A2, CYP2C8, CYP2C9 or CYP2C19, but there may be a potential to inhibit CYP2D6 and CYP3A4/5.

Elimination of iloperidone-related materials following oral administration of iloperidone to mice, rats, rabbits, and dogs was rapid, with feces as the main route of elimination (indicating biliary excretion). Upon oral or i.v. administration of iloperidone to CD-1 mice, the drug was extensively metabolized prior to excretion with little or no iloperidone observed in urine; open-ring P89 was one of the major urinary metabolites; P88 and its open-ring metabolite were major fecal metabolites and small amounts of unchanged iloperidone or open-ring form of iloperidone were present in feces following both doses. In humans, however, the main excretion route of products of iloperidone's metabolism is the kidney, different from the animal species tested, in which most of the iloperidone dose is excreted with bile into feces.

ADME profiles of two major iloperidone metabolites, P95 and P88, were also investigated. Absorption of P95 was rapid; C<sub>max</sub> was reached at 0.5 h. after single-dose oral administration of P95 at 5 to 20 mg/kg to CD-1 mice or 5 to 50 mg/kg to Sprague-Dawley rats. Oral bioavailability was greater in mice (18-20%) than in rats (1.4-2.6%). Plasma exposure values in mice increased dose proportionally, while C<sub>max</sub> and AUC in rats increased over-proportionally with dose increase. In humans, the pharmacokinetics of iloperidone and its two main metabolites P88 and P95 is considered dose-proportional in the dose range studied, i.e. from 2 to 12 mg b.i.d. (4 to 24 mg daily).



- Additional methods were developed for the synthesis of [14C]iloperidone, [3H]P95, and [3H]P88 (studies DMPK(US)R00-2143, DMPK(US)R01-1058, DMPK(US)R99-053, and DMPK(US)R99-034, respectively).

#### 2.6.4.3 Absorption

Single- and multiple-dose studies with oral or i.v. administration of iloperidone or one of its metabolites, P95 or P88, were performed in the mouse (CD-1), rat (Sprague-Dawley, Han Wistar), rabbit (New Zealand), dog (Beagle), and monkey (Cynomolgus). In each species studied, absorption of iloperidone was rapid, but bioavailability of the compound was low, likely due to first-pass effect. Species-specific variability in absorption was observed, with the lowest bioavailability in the rat.

##### - Single-dose studies with iloperidone

The absorption parameters of iloperidone in the mouse, rat, rabbit, dog, and monkey following a single oral or i.v. administration are summarized in the sponsor's table below. PK profiles varied among different animal species. After a single oral dose administration at 5 mg/kg, C<sub>max</sub> levels in all species were reached between 0.5 to 1.2 hours post dosing; both C<sub>max</sub> and AUC levels in the dog were greater than other species; absorption rates were 50% to 80% in mouse, 56% to 86% in rat, 76% to 79% in rabbit and 42% to 99% in dog; oral bioavailability was <1% in rat, 5% in mouse, and 19% in both rabbit and dog. The low bioavailability was likely due to a first-pass effect.

Iloperidone plasma exposure parameters in rats (C<sub>max</sub> and AUC) were increased over-dose proportionally upon administration of single oral doses of 5, 16, and 20 mg/kg. Mean plasma drug concentration was higher in female than in male rats following a single oral dose of 16 mg/kg loperidone.

Upon intravenous administration (1 mg/kg), C<sub>max</sub> was recorded at the first sampling time (0.083 h) in all species; both C<sub>max</sub> and AUC levels in dogs were higher than those in other species.

Mean PK parameters following a single-dose oral or intravenous administration of iloperidone

Species	Study No.	Dose (mg/kg)	Admin. Route	C <sub>max</sub> (ng/mL)	t <sub>max</sub> (h)	AUC <sub>0-∞</sub> (ng·h/mL)
				M/F	M/F	M/F
Mouse (CD-1)	DMPK(US) R99-1188	5	p.o.	12.7/NA	0.5/NA	76.2/NA
Mouse (CD-1)	ILO-PK-001	20	p.o.	560/NA	1/NA	NA
Rat (Sprague-Dawley)	DMPK(US) R98-2214	5	p.o.	3.38/NA	1.2/NA	6.44/NA
Rat (Sprague-Dawley)	DMPK(US) R01-1181	16	p.o.	41.5/259	2/1	177/790 <sup>a</sup>
Rat (Wistar)	ILO-PK-001	20	p.o.	202/NA	0.5/NA	629/NA <sup>b</sup>
Rabbit (New Zealand White)	DMPK(US) R99-1190	5	p.o.	NA/65.4	NA/1	NA/534
Dog (Beagle)	ILO-PK-001	5	p.o.	2/NA	1/NA	NA
Dog (Beagle)	DMPK(US) R99-1189	5	p.o.	165/NA	0.5/NA	844/NA
Monkey (Cynomolgus)	ILO-PK-001	5	p.o.	34/NA	1/NA	NA
Mouse (CD-1)	DMPK(US) R99-1188	1	i.v.	162/NA	0.83/NA	306/NA
Rat (Sprague-Dawley)	DMPK(US) R98-2214	1	i.v.	370/NA	0.083/NA	316/NA
Rabbit (New Zealand White)	DMPK(US) R99-1190	1	i.v.	NA/485	NA/0.083	NA/573
Dog (Beagle)	DMPK(US) R99-1189	1	i.v.	689/NA	0.083/NA	905/NA

AUC<sub>0-∞</sub>=area under the plasma concentration curve from time 0 hour to infinity;  
a AUC<sub>0-24</sub>; b AUC 0 to last sampling time point (2 or 4).

- **Multiple-dose studies with iloperidone**
- Multiple-dose study with iloperidone in the mouse (Study DMPK(US)R01-1353)

After administration of repeat oral 10-mg/kg dose of iloperidone for 9 days followed by [<sup>14</sup>C]iloperidone (specific activity=20.8  $\mu$ Ci/mg) for the following 5 days to CD-1 mice, plasma concentrations of iloperidone and metabolite P95 were determined on Day 14. The C<sub>max</sub> values of iloperidone and metabolite P95 (21.4 ngEq/mL in male and 16.2 ngEq/mL in female) were observed 0.5 to 1 h post dose in both male and female mice. Iloperidone AUC<sub>0-24</sub> values were 528 ng•h/ml and 618 ng•h/ml for males and females, respectively. For the metabolite P95, AUC<sub>0-24</sub> values were 69.3 ngEq•h/ml and 77.6 ngEq•h/ml for males and females, respectively. No obvious gender difference in exposure was observed.

- Multiple-dose study with iloperidone in the rat (Study DMPK(US)R01-1181)

Administration of 5 daily oral doses of iloperidone (16 mg/kg/day) to Sprague-Dawley rats resulted in the following exposure values for iloperidone, P88, and P95:

- C<sub>max</sub>: males 181, 8.3, and 6.7 ng/ml, respectively; females 151, 7.2, and 8.9 ng/ml, for iloperidone, P88, and P95, respectively;
- T<sub>max</sub>: males 1, 1, and 1 h; females 2, 2, and 1 h for iloperidone, P88, and P95, respectively;
- AUC<sub>0-24</sub>: males 343, 18.1, and 11.4 ng•h/ml; females 937, 49.2, and 58.9 ng•h/ml for iloperidone, P88, and P95, respectively.

Thus, plasma exposure values (AUC<sub>0-24</sub>) for iloperidone and metabolites was higher in female than male rats.

The following sponsor's table summarizes the mean PK parameters of iloperidone in the plasma of mice and rats administered with multiple oral doses of iloperidone. Following a 14-day oral administration, both C<sub>max</sub> and AUC levels were comparable in male and female mice. However, the AUC value in female rats was nearly 2.5-fold greater than the male rats, after 5 daily consecutive doses.

Mean PK parameters of iloperidone following multiple-dose oral administration of iloperidone

Species	Study No.	Duration	Dose (mg/kg)	Admin. Route	C <sub>max</sub> (ng/mL)	t <sub>max</sub> (h)	AUC <sub>0-24</sub> (ng•h/mL)
					M/F	M/F	M/F
Mouse (CD-1)	DMPK(US) R01-1353	14 days	10	p.o.	172/139	0.5/0.5	528/618
Rat (Sprague-Dawley)	DMPK(US) R01-1181	5 days	16	p.o.	181/151	1/2	343/937

- **Single-dose studies with metabolite P95**

P95, one of the major metabolites of iloperidone, was orally or i.v. administered in 2 separate studies in male mice and rats. Plasma concentrations of P95 were measured between 0 to 24 hours and 0 to 72 hours, respectively. Following oral administration at 5, 10, and 20 mg/kg, the levels of C<sub>max</sub> and AUC in mice were higher than those in rats; t<sub>max</sub> levels in all dosing groups were identical in both species. The oral bioavailability values in mice calculated after a single oral doses of 5, 10, and 20 mg/kg were 18%, 32%, and 20%, respectively; in rats, after a single oral doses of 20 and 50 mg/kg the oral bioavailability was much lower (1.4% and 2.6%, respectively). When mice and rats were

dosed i.v. at 1 mg/kg, the AUC levels in the rat were relatively greater than in the mouse. The data are summarized in the following sponsor's table.

Mean PK parameters in male mice and rats following single-dose administration of P95

Species	Study No.	Dose (mg/kg)	Admin. Route	C <sub>max</sub> (ng/mL)	t <sub>max</sub> (h)	AUC <sub>t=∞</sub> (ngEq•h/mL)
Mouse (CD-1)	DMPK(US) R99-1246	5, 10, 20	p.o.	119, 276, 507	0.5, 0.5, 0.5	336, 1180, 1500 <sup>a</sup>
Rat (Sprague-Dawley)	DMPK(US) R99-1245	5, 10, 20, 50	p.o.	3.21, 31.6, 60.2, 455	0.5, 0.5, 0.5, 0.5	1.61, 19.5, 161, 718 <sup>b</sup>
Mouse (CD-1)	DMPK(US) R99-1246	1	i.v.	948	0.083	368 <sup>c</sup>
Rat (Sprague-Dawley)	DMPK(US) R99-1245	1	i.v.	1560	0.083	538 <sup>c</sup>
Rat (Han Wistar)	DMPK(US) R99-1245	1	i.v.	1000	0.083	579 <sup>c</sup>

<sup>a</sup> AUC<sub>0-24</sub>, ngEq•h/mL; <sup>b</sup> AUC<sub>0-24</sub> ng•h/mL; <sup>c</sup> AUC<sub>0-72</sub>, ngEq•h/mL.

The following sponsor's summary adequately describes the results of these studies:

- Single-dose study with P95 in the mouse (Study DMPK(US)R99-1246)

"After iloperidone administration, the exposure of P95 was low in the mouse relative to that in the human. Therefore, this study was conducted to gain further insight into the disposition of P95 in the mouse following a single non-radio-labeled P95 oral (gavage) or [14C]P95 (specific activity=116 µCi/mg) i.v. doses in male mice (CD-1). After administration of a single oral dose of P95 to mice at 5, 10, and 20 mg/kg, the following calculations were made regarding concentrations of P95 in plasma: C<sub>max</sub> values at 119, 276, and 507 ng/ml, t<sub>max</sub> values at 0.5, 0.5, and 0.5 hour, and AUC<sub>0-24</sub> values at 336 ± 77.8, 1180 ± 201, and 1500 ± 176 ngEq•h/mL, respectively. The oral bioavailability values calculated after a single oral dose of 5, 10, and 20 mg/kg were 18%, 32%, and 20%, respectively. The plasma concentration of P95 following a single i.v. administration of [14C]P95 at a dose of 1 mg/kg is as follows: a C<sub>max</sub> value of 948 ± 56.8 ng/ml, a t<sub>max</sub> value of 0.083 hour (first sampling time), and an AUC<sub>0-72</sub> value of 368 ± 56.8 ngEq•h/ml were found in CD-1 mice. With respect to radioactivity in plasma, a C<sub>max</sub> value of 894 ± 76.1 ng/mL, a t<sub>max</sub> value of 0.083 hour (first sampling time), and an AUC<sub>0-72</sub> value of 894 ± 76.1 ngEq•h/mL were calculated. P95 concentrations were below the limit of detection within 4 hours postdose; however, measurable amounts of drug-related material were present throughout the 72-hour study. This study demonstrated that exposure is proportional to dose".

- Single-dose study with P95 in the rat (Study DMPK(US)R99-1245)

"Since the exposure of P95 was relatively low in the rat relative to humans, to gain a better insight into the disposition of P95 in the different strains of rats, a single [14C]P95 (specific activity=116 or 117 µCi/mg) i.v. or non-radio-labeled P95 oral (gavage) dose was given to male rats (Han Wistar and Sprague-Dawley). After single i.v. administration of [14C]P95 at a dose of 1 mg/kg, the plasma unchanged drug-concentration-time profiles were characterized by an initial concentration (first sample time=0.083 hour) of 1560 ng/ml in Sprague-Dawley and 1000 ng/ml in the Han Wistar rats with a rapid decline thereafter. For Han Wistar rats, the AUC<sub>0-72</sub> was 579 ngEq•h/ml. For Sprague-Dawley rats, the AUC<sub>0-72</sub> was 538 ngEq•h/mL. Thus, no difference in P95 absorption was observed between the 2 strains. With respect to radioactivity in the plasma of Han

Wistar and Sprague-Dawley rats, the AUC<sub>0-72</sub> values were 1200 ± 17.1 and 1560 ± 105 ngEq•h/mL, respectively. After single-dose administration of an oral (gavage) dose of P95 to Sprague-Dawley rats at 5, 10, 20, and 50 mg/kg, the C<sub>max</sub> values of 3.21 ± 5.57, 31.6 ± 13.3, 60.2 ± 33.8, and 455 ± 284 ng/mL; t<sub>max</sub> values of 0.5, 0.5 ± 0, 0.5 ± 0, and 0.5 ± 0 h; and AUC<sub>0-24</sub> values of 1.61 ± 2.78, 19.5 ± 9.99, 161 ± 199, and 718 ± 424 ng•h/ml were calculated, respectively. Therefore, increasing P95 doses yielded increases in exposure that were over-proportional to dose. The oral bioavailability calculated after a single oral dose of 20 and 50 mg/kg were 1.4% and 2.6%, respectively. Bioavailability was not calculated for the 5- or 10-mg/kg doses due to the limited AUC<sub>0-24</sub> data.”

- **Single-dose studies with metabolite S-P88**

It was hypothesized that iloperidone and P88 existed in equilibrium, and that the same metabolic profile should be present regardless of whether iloperidone or S-P88 was administered. To test this hypothesis, the plasma concentrations of iloperidone and metabolites of iloperidone, P95 and P88, were determined in 2 animal species (mice and dogs) after single oral (gavage) or i.v. doses of [14C]S-P88. Since a chiral method had demonstrated that there was no R-P88 formed in animals or humans, all concentrations were reported as S-P88. Studies evaluating the absorption profile of S-P88 were performed in male mice (Study DMPK(US)R01-950) and male dogs (Study DMPK(US)R01-1016). After a single oral dose of 5 mg/kg of [14C]S-P88 to either species, oral bioavailability was only 2% in the mouse and 5% in the dog, which suggested an extensive first-pass effect. After a single oral dose administration of [14C]S-P88, the overall metabolite patterns in mouse plasma were qualitatively similar to those in mice dosed with a single oral dose of [14C]iloperidone, suggesting a possible rapid conversion of S-P88 to iloperidone following oral administration. In dogs however, there was no evidence that S-P88 and iloperidone were in equilibrium. PK parameters of S-P88 are summarized in the following sponsor’s table.

Mean PK parameters of P88 following single-dose administration of S-P88 to mice and dogs

Species	Study No.	Dose (mg/kg)	Admin. Route	C <sub>max</sub> (ng/mL)	t <sub>max</sub> (h)	AUC <sub>t-∞</sub> (ng•h/mL)
Mouse (CD-1)	DMPK(US) R01-950	5	p.o.	3.70	0.5	7.20
Dog (Beagle)	DMPK(US) R01-1016	5	p.o.	54.5	0.7	58.3
Mouse (CD-1)	DMPK(US) R01-950	1	i.v.	80.3	0.083	66.7
Dog (Beagle)	DMPK(US) R01-1016	1	i.v.	545	0.083	233

The following sponsor’s summary adequately describes the results of these studies:

- Single-dose study with S-P88 in the mouse (Study DMPK(US)R01-950)

“The plasma concentrations of iloperidone and metabolites of iloperidone, P95 and P88, were determined after single oral (gavage) or i.v. doses of [14C]S-P88 (specific activity=20 or 117 µCi/mg, respectively) to male mice (CD-1). After administration of a single oral dose of [14C]S-P88 at 5 mg/kg, C<sub>max</sub> values of 3.7, 2.3-2.6, and 2.2 ng/ml,

$t_{max}$  values of 0.5, 2-4, and 0.5 hours, and AUC<sub>0-72</sub> values of  $7.2 \pm 1.6$ ,  $20.2 \pm 6.4$ ,  $3.9 \pm 1.96$  ng•h/ml were observed for *S*-P88, *R*-P88, and P95 in plasma, respectively. A  $C_{max}$  value of 22.7 ng/ml,  $t_{max}$  value of 0.5 hour, and an AUC<sub>0-∞</sub> value of  $73.1 \pm 10.6$  ng•h/ml were observed for iloperidone. With respect to radioactivity in plasma,  $C_{max}$  values of 492 and 85.4 ngEq/ml,  $t_{max}$  values of 0.5 and 4 hours, and an AUC<sub>0-72h</sub> value of  $2260 \pm 285$  ngEq•h/ml were observed. The oral bioavailability calculated after a single oral dose of 5 mg/kg of [<sup>14</sup>C]*S*-P88 was low (2%), indicating extensive first-pass elimination in the dose. There was complete absorption of the 5- mg/kg oral dose of [<sup>14</sup>C]*S*-P88. After single i.v. administration of [<sup>14</sup>C]*S*-P88 at a dose of 1 mg/kg,  $C_{max}$  values of 80.3, 176, and 1.24 ng/mL,  $t_{max}$  values of 0.08, 0.5, and 0.5 hour, and AUC<sub>0-72</sub> values of  $66.7 \pm 7.5$ ,  $53.2 \pm 2.8$ , and  $3.6 \pm 2.3$  ng•h/ml were observed for *S*-P88, *RP*88, and P95 in plasma, respectively. For iloperidone in plasma, a  $C_{max}$  value of 29.7 ng/ml, a  $t_{max}$  value of 0.5 h, and an AUC<sub>0-∞</sub> value of  $72.1 \pm 8.5$  ng•h/ml were observed. With respect to radioactivity in plasma,  $C_{max}$  values of 500 and 191 ngEq/mL,  $t_{max}$  values of 0.5 and 8 h, and an AUC<sub>0-72h</sub> value of  $1800 \pm 691$  ngEq•h/ml were observed.”

- Single-dose study with *S*-P88 in the dog (Study DMPK(US)R01-1016)

“The plasma concentrations of iloperidone and metabolites, P95 and P88, were determined after single oral (gavage) or i.v. doses of [<sup>14</sup>C]*S*-P88 (specific activity= 5.94 or 30.5 μCi/mg, respectively) in solution to male dogs (Beagle). Since a chiral method demonstrated that there was no *R*-P88 formed, all concentrations are reported as *S*-P88. After administration of a single oral dose of [<sup>14</sup>C]*S*-P88 at 5 mg/kg,  $C_{max}$  values of  $54.5 \pm 65.7$  and  $16.6 \pm 11.1$  ng/ml,  $t_{max}$  values of  $0.7 \pm 0.3$  and  $5.3 \pm 3.1$  h, and AUC<sub>0-∞</sub> values of  $58.3 \pm 47$  and  $376 \pm 266$  ng•h/ml were observed, for P88 and P95, respectively. For iloperidone, a  $C_{max}$  value of  $99 \pm 105$  ngEq/ml, a  $t_{max}$  value of  $0.83 \pm 0.3$  h, and AUC<sub>0-∞</sub> values of  $212 \pm 132$  ng•h/ml were observed. With respect to radioactivity in plasma, a  $C_{max}$  value of  $1040 \pm 659$  ngEq/ml, a  $t_{max}$  value of  $0.08 \pm 0.3$  hour, and an AUC<sub>0-∞</sub> value of  $8640 \pm 2580$  ng•h/ml were observed. The oral bioavailability after a single oral dose of 5 mg/kg of [<sup>14</sup>C]*S*-P88 was low ( $5\% \pm 4\%$ ), indicating extensive first-pass elimination of the dose. The absorption of the 5-mg/kg oral dose of [<sup>14</sup>C]*S*-P88 was high ( $58.7\% \pm 17.5\%$ ). After single i.v. administration of [<sup>14</sup>C]*S*-P88 at a dose of 1 mg/kg,  $C_{max}$  values of 545 and 2.4 ng/mL,  $t_{max}$  values of 0.08 and 7.5 h, and AUC<sub>0-∞</sub> values of 233 and 62.5 ng•h/ml were observed for P88 and P95, respectively. For iloperidone, a  $C_{max}$  of 145 ngEq/ml, a  $t_{max}$  of 0.8 h, and an AUC<sub>0-∞</sub> value of 367 ng•h/ml were observed. Iloperidone levels were below the LOQ after 24 hours. With respect to radioactivity in plasma, a  $C_{max}$  value of 717 ngEq/ml, a  $t_{max}$  value of 0.08 h, and an AUC<sub>0-∞</sub> value of 2940 ngEq•h/ml were observed. Comparison of the data generated from administration of [<sup>14</sup>C]iloperidone to the dog (Study DMPK(US)R99-1189) and the data generated from this study provided no evidence that *S*-P88 and iloperidone are in equilibrium after an oral dose of *S*-P88 in the dog.”

#### 2.6.4.4 Distribution

Distribution profile of iloperidone was evaluated *in vitro* in animals and humans and *in vivo* in animals (mouse, rat, rabbit, and dog).

##### Protein binding and distribution in blood cells

Plasma protein-binding of iloperidone was greater than 90%, 86%, and 93% in the rat, dog and human, respectively, as determined in 2 *in vitro* studies using [<sup>14</sup>C] and [<sup>3</sup>H]

iloperidone (Study TH1D150793, Study DMPK(US)R99-1121). A similar plasma protein-binding in humans was also determined in an ex vivo study using plasma of volunteers treated with iloperidone at a single oral dose of at 3 mg; the mean protein-binding value was 94.4% at 3 hours and 94.8% at 24 hours post dose; plasma protein-binding rate of iloperidone was not significantly affected by its metabolites in this ex vivo test (Study DMPK(US)N01-1200).

The mean binding values of P95 metabolite in the rat, dog, and human plasma were approximately 90.5%, 68.5%, and 84.9%, respectively, as determined in vitro (Study DMPK(US)R99-1121).

The blood distribution of iloperidone in human, dog, and rat blood using [<sup>14</sup>C]iloperidone at concentrations of 0.1 to 100 µg/ml showed that in all species tested, only a small amount of iloperidone was detected in red blood cells; the fraction of [<sup>14</sup>C]iloperidone distributed into the red blood cells (fBC) was <0.25 ) in each test species. Iloperidone blood/plasma concentration ratios were independent of the drug concentrations used for the incubation (Study DMPK(US)R01-1427)

#### **Distribution of iloperidone in tissues**

Tissue distribution of iloperidone was investigated in the mouse, rat, rabbit, and dog. Following a single-dose i.v. administration, the steady-state volume of distribution (V<sub>ss</sub>) values of iloperidone in mice (7.7 l/kg), rats (4.1 l/kg), rabbits (2.9 l/kg), and dogs (2.3 l/kg) were approximately 10, 6, 4, and 4 times larger than the volume of total body water in these species, respectively, indicating an extensive tissue distribution after i.v. administration of iloperidone (Studies DMPK(US)R99-1188, DMPK(US)R98-2214, DMPK(US)R99-1190, and DMPK(US)R99-1189).

After oral or i.v. administration, tissue distributions of radio-labeled iloperidone were qualitatively evaluated in rat studies showing that iloperidone was quickly distributed into different organs; the highest drug concentrations were generally observed in liver, kidney, gastrointestinal system, and secretory glandular tissues, while the concentration of radioactivity was very limited in central nervous tissues (Studies AC1D290192, DMPK(US)R01-1362, and DMPK(US)R00-1313). Similar tissue distribution results for iloperidone were found in pigmented rats, except for the concentration of radioactivity in the eyes which was significantly higher in pigmented than in albino eyes (Study 901504). The ability of iloperidone and its metabolite, P88 to cross the blood-brain barrier was studied in vitro using an in vitro 2 cell monolayer model at 5-µM donor concentrations of [<sup>14</sup>C]iloperidone and [<sup>14</sup>C]S-P88 (Study ADME(US)R01-1488). The apparent permeabilities (P<sub>app</sub>) were calculated based upon total [<sup>14</sup>C]iloperidone and [<sup>14</sup>C]S-P88 radioactivity located on the receiver side or donor side of the model, after a 2-hour incubation. Both [<sup>14</sup>C]iloperidone and [<sup>14</sup>C]S-P88 were found to be highly permeable compounds under the conditions of this experiment. These data suggest that iloperidone and S-P88 can cross the blood-brain barrier.

#### **Distribution of metabolite P95 in tissues**

The tissue distribution of [<sup>14</sup>C]P95 was studied by quantitative whole-body autoradiography in adult male Long Evans Hooded (pigmented) rats after a single 1-mg/kg i.v. dose of [<sup>14</sup>C]P95 (specific activity=116 µCi/kg) (Study DMPK(US)R99-2330). One rat per time point was sacrificed at 5 minutes, 1 hour, 4 hours, and 24 hours post dose. [<sup>14</sup>C]P95-derived materials were not observed in the brain, indicating minimal or no passage across the blood-brain barrier. The bile concentration (11.4µg-Eq/g) at 5

minutes post dose was higher than any other tissue, organ or fluid. The highest tissue concentrations were measured at 5 minutes post dose in the kidney (3.25µg-Eq/g) and liver (2.61µg-Eq/g). [14C]P95 and its radio-labeled metabolites were rapidly eliminated, since at 4 hours post dose, only the liver had measurable amounts of radioactivity (0.786µg-Eq/g). At 24 hours post dose, no radioactivity was detected in any tissue, fluid, or organ.

#### **Distribution in pregnant and lactating animals**

##### **- Iloperidone**

Placental transfer and subsequent fetal tissue distribution were examined in pregnant rats (Study AC1D140294) and rabbits (Study AC2D140294) orally dosed with [14C] iloperidone. Limited placental transfer of radioactivity to fetus was observed in both species; the concentration of radioactivity was barely detectable in fetal organs.

In another study, drug concentrations in plasma and milk were compared after iloperidone oral administration to lactating rats at 5 mg/kg (Study TH1D01694). Peak concentrations in plasma and milk were attained at 0.5 and 4 hours post dose, respectively; drug concentration in milk at 4 hours post dose was approximately 10 times that in the plasma; and the concentration levels of iloperidone in milk were higher than in plasma from 1 to 12 hours post dosing.

##### Whole-body radiography study of iloperidone in pregnant albino rats (Study AC1D140294)

Single oral administration of [14C] iloperidone to pregnant Sprague-Dawley rats (3/group) on Gestation day 12 or 18 at a dose level of 64 mg/kg, resulted, at 1 hour post dose, in general distribution throughout the maternal tissues with the highest concentrations in the liver, kidney, gastrointestinal tract, lung, adrenal gland, and bone marrow. Lesser, but significant, concentrations of radioactivity were observed in the fat, mammary tissue, thymus, placenta, amniotic sac, and central nervous tissue. Blood concentrations appeared to be at or just above the level of detection. Only limited placental transfer of radioactivity to the fetus was observed (maternal liver concentrations were approximately 10 to 20 times higher than the barely discernible concentration in fetal liver). By 48 hours post dose, only the liver, kidney, gastrointestinal tract contents, placenta, and amniotic sac contained detectable concentrations of radioactivity. At the later time points, due to the limited placental transfer of radioactivity and the subsequent minimal fetal distribution, it was difficult to identify individual fetal tissue, particularly those from the 12th day of gestation animals.

There were no significant differences observed in the distribution of radioactivity between gestational Day 12 and Day 18. The dose formulation used in this study (suspension in starch mucilage) impaired the absorption of drug material.

##### Distribution of radiolabeled iloperidone in pregnant rabbit (Study AC2D140294)

Administration of radiolabeled iloperidone at a single oral dose of 25 mg/kg to pregnant New Zealand rabbits on Gestation Day 12 or Day 19 resulted, at 1 hour post dose, in a general distribution of radioactivity throughout the maternal tissues, with the highest concentrations in the liver, kidney, bladder, and gastrointestinal tract. Lesser but still significant amounts of radioactivity were found in the lung, bone marrow, spleen, salivary glands, fat, myocardium, central nervous system, and skeletal muscle. Limited placental transfer occurred, and fetal distribution was barely detectable. Concentrations of radioactivity in the tissues appeared to reduce slowly over the time course (1 to 48 hours)

of the experiment. There were no significant differences observed between gestational Day 12 or Day 19 at any time point during the course of the experiment.

Iloperidone Excretion into Milk (Study TH1D01694)

[14C]Iloperidone oral administration to lactating Sprague Dawley rats at a single dose of 5 mg/kg resulted in the following concentrations in the maternal plasma and milk (as determined by HPLC analysis of samples taken at 1, 2, 4, 6, and 12 hours post dose):

Test Article: Iloperidone						
Time (h): 1, 2, 4, 6, 12, 24	1 h	2 h	4 h	6 h	12 h	24 h
<b>Concentration of Radioactivity</b>						
Milk:	0.922	1.400	1.469	1.150	0.407	0.044
Plasma:	0.329	0.205	0.150	0.161	0.098	0.064

The main study findings were as follows: Iloperidone and its metabolites were concentrated nearly 10-fold in milk vs. plasma at 4 hours following oral administration; concentrations in milk were lower than plasma at 24 hours after administration; metabolite profile was similar in milk and plasma.

**- P95 distribution in pregnant animals**

Distribution of [14C]P95 in pregnant rats (Study DMPK(US)R01-417)

Since rats do not metabolize iloperidone to P95 to the extent seen in humans, this study was performed to more fully characterize P95 distribution in rat tissues and organs. The distribution of radioactivity following a single oral dose of [14C]P95 into the organs, tissues, and body fluids of pregnant rats (Sprague-Dawley) was studied by quantitative whole-body autoradiography. Following an 80-mg/kg [14C] P95 oral dose administered to pregnant rats on gestational Days 10 and 17, radioactivity was distributed only to the liver, kidney, and spleen. No [14C]P95 was detectable in the brain, which corroborated the findings in P95 distribution study in non-pregnant rats (Study DMPK(US)R99-2330), suggesting that P95 does not cross the blood-brain barrier. Distribution of radioactivity on gestational Days 10 and 17 appeared to be the same, regardless of the gestational age of the animals. The highest tissue concentrations were detected at 1 hour post dose in the kidney and liver (26 to 27 µgEq/g). Radioactivity was not detected in any other tissues, organs, or fluids including the fetus, placenta, amniotic fluid, or implantation site. Radioactivity was eliminated quickly; at 24 hours post dose, only the liver in the Day 10 animal contained measurable radioactivity (11.7 µg/Eq/g.).

**2.6.4.5 Metabolism**

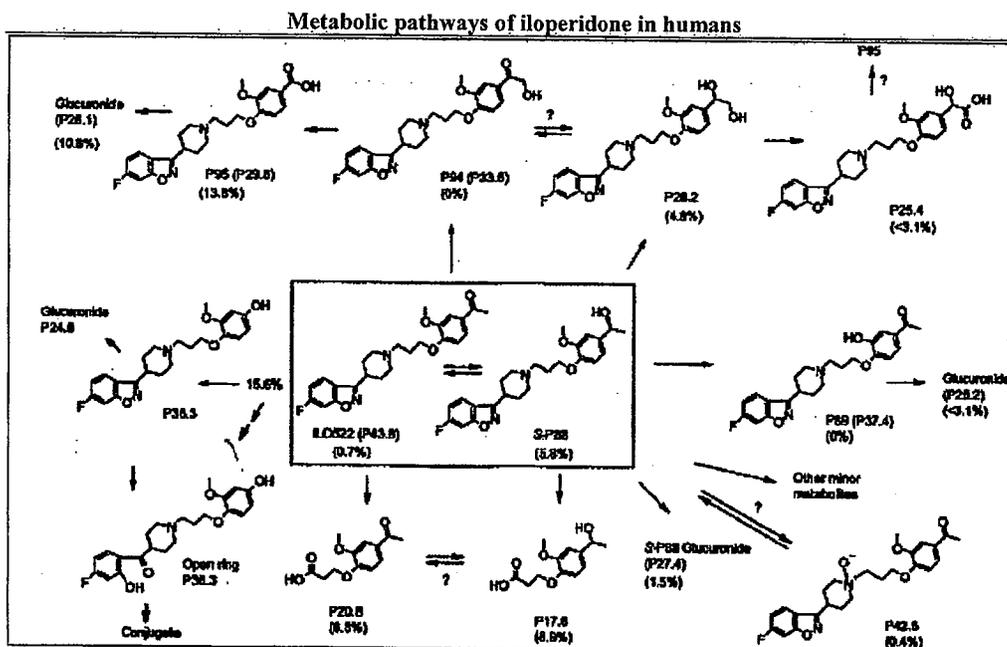
Metabolism of iloperidone was investigated *in vitro* (in human liver microsomes; in rat liver extracts; in liver S9 preparations from rodents, rabbits, dogs and humans; in bile and urine of rats and dogs), as well as *in vivo* in the mouse, rat, rabbit, and dog. Metabolism of the metabolites of iloperidone, S-P88 and P95, was also evaluated *in vivo*, in mice, rats and dogs upon oral and i.v. administration.

**Metabolism of iloperidone *in vitro***

*In vitro*, iloperidone was metabolized in human liver microsomes to primarily form 4 products: P22 (*N*-dealkylation), P94 (hydroxylation), P89 (*O*-demethylation), and P88 (carbonyl reduction) (Study DMPK(US)R01-538). In human liver S9, the major product formed was P88. The higher formation of P88 in human liver S9 incubations, as compared to human liver microsomes, supported the formation of P88 by cytosolic

enzymes, as well as by cytochrome P450 in humans in vivo. The in vitro formation of P88 was contributed to by several specific P450 enzymes, including CYP1A1, CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C19, CYP3A4, and CYP3A5. The pathways for the metabolism of iloperidone in human were similar to rats. The metabolites excreted in the bile and urine of rats (Wistar) and dogs (Beagles) that were administered iloperidone (rats: 60 mg/kg, i.p.; dogs: 20 mg/kg, p.o.) were identified and characterized by combination of LC/MS and liquid chromatography/nuclear magnetic resonance (LC/NMR) (Study AEM022594). Iloperidone was demonstrated to be metabolized by the *O*-demethylation to yield 6-fluoro-3-[1-(3-hydroxypropyl)-4-piperidinyl]-1,2-benzisoxazole and 1-[4-[3-[4-(6-fluoro-1,2-benzisoxazol-3-yl)-1-piperidinyl]propoxy]-2,5-dihydroxyphenyl]ethanone and by hydroxylation on the acetophenone ring, producing 1-[4-[3-[4-(6-fluoro-1,2-benzisoxazol-3-yl)-1-piperidinyl]propoxy]-2,5-dihydroxyphenyl]ethanone.

The metabolism pathway of iloperidone in human and animal species is illustrated in the sponsor's figures below and on the next pages.



Steady-state kinetics of iloperidone carbonyl reduction to form P88 and oxidation of *S*-P88 to form iloperidone were examined in S9 preparations from various species (Study DMPK(US)R01-1294) to explain the lower exposure of P88 than iloperidone in rodents and dogs as compared to that in humans, in whom P88 exposure is greater than that of iloperidone. Ratios of iloperidone carbonyl reduction to *S*-P88 reduction intrinsic clearance values in liver S9 of rabbit, human, mouse, rat, and dog were 5.7, 2.4, 0.50, 0.46, and 0.26, respectively. The in vitro reactions with rabbit and human liver S9 indicated that iloperidone was metabolized with a greater efficiency to the reduced form of iloperidone (P88) than *S*-P88 is oxidized to form iloperidone. The equilibrium of the reduction/oxidation reaction favored formation of P88, and the preference was due to a

higher maximum enzyme velocity ( $V_{max}$ ) and/or reduced concentration of substrate that produces half-maximal velocity, in the presence of a competitive inhibitor ( $K_m$ ) values. In mouse, rat, and dog liver S9, the reaction tended toward formation of iloperidone from *S*-P88 rather than reduction of iloperidone to form P88. The low formation of P88 in rat and dog *in vivo* could also be due to a greater metabolic clearance of iloperidone by biotransformation pathways other than formation of P88. The preference for *S*-P88 oxidation over iloperidone reduction in mouse, rat, and dog are in line with *in vivo* study results in these animals, in which iloperidone was found to be more predominant than P88.

#### **Metabolism of iloperidone *in vivo***

The patterns and identity of iloperidone and its metabolites were determined following single oral or *i.v.* doses of 5 or 1 mg/kg, respectively, of [ $^{14}C$ ]iloperidone to mice, rats, rabbits, and dogs.

Metabolism of the metabolites of iloperidone, *S*-P88 and P95, was also evaluated *in vivo*, in mice, rats and dogs upon oral and *i.v.* administration.

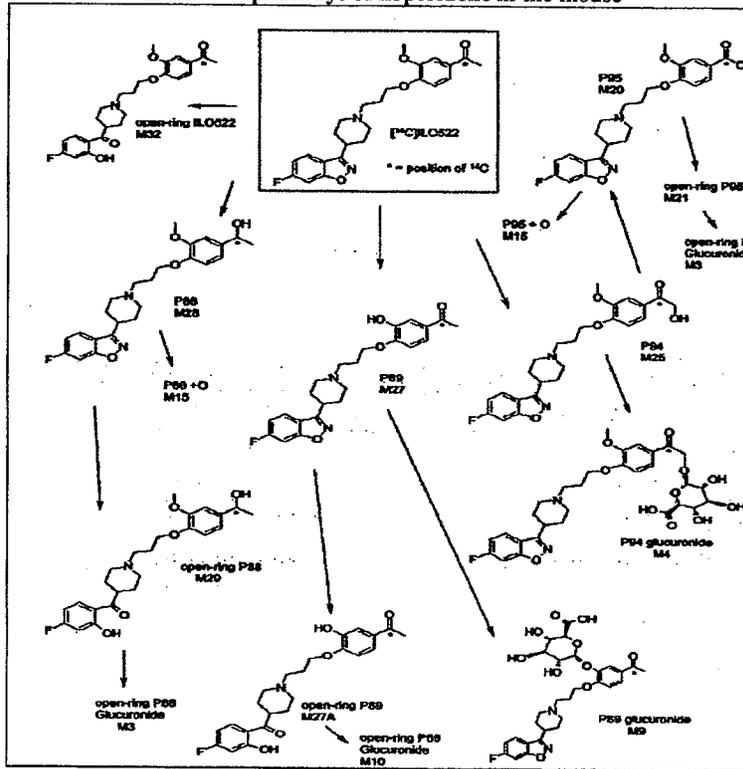
##### Single-dose study with iloperidone in the mouse (Study DMPK(US)R99-1188)

After single *i.v.* administration of [ $^{14}C$ ]iloperidone at a dose of 1 mg/kg, iloperidone and/or its open-ring metabolite and an glucuronide of open-ring P89 were major circulating species for the first 4 hours postdose. P95 and P88 were only minor circulating metabolites, representing no more than 1.7% or 5.1%, respectively, of the plasma radioactivity. After single administration of an oral (gavage) dose of [ $^{14}C$ ]iloperidone of 5 mg/kg, the open-ring glucuronide of P89 was the major circulating species (26% to 62%) for the first 8 hours postdose, while iloperidone and its open-ring form were only minor species (5.7%) in plasma. P95 and P88 concentrations were below the LOQ in all samples when quantified by LC/MS/MS (non-radiochemical detection).. Iloperidone was extensively metabolized prior to excretion with little or no iloperidone detected in urine, and only about 2.5% in feces, following either *i.v.* or oral dosing. Open-ring P89 was one of the major urinary metabolites. P88 and its open-ring metabolite were major fecal metabolites following both doses.

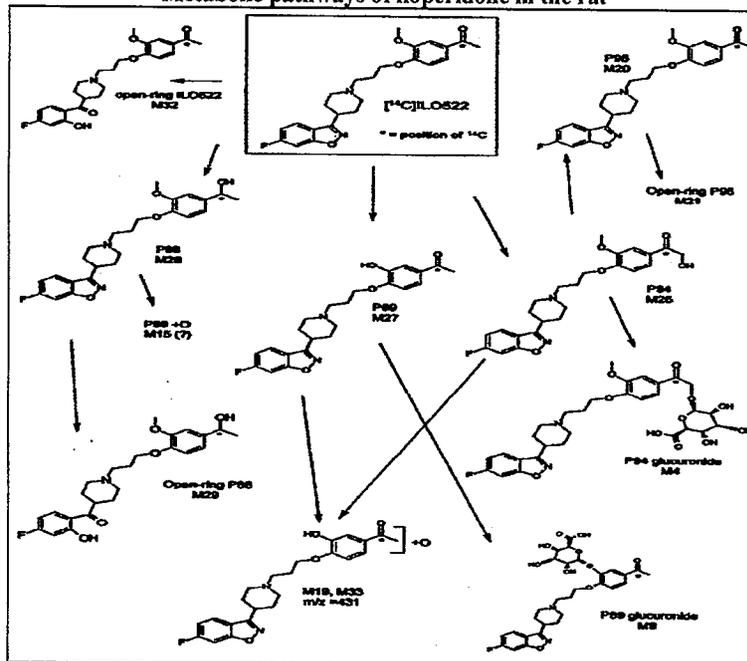
##### Single-dose study with iloperidone in the rat (Study DMPK(US)R98-2214)

After single administration of an oral (gavage) dose of [ $^{14}C$ ]iloperidone of 5 mg/kg to rats (Sprague Dawley), the major circulating metabolites were conjugates of oxidized metabolites, unconjugated iloperidone, and its open-ring form. Small amounts of unconjugated P94, P95, and P88 were also observed following both oral and *i.v.* administration. The metabolic profiles from the *i.v.* and oral doses were similar. Following *i.v.* administration, a product of both oxidation and *O*-demethylation was the major circulating species. Iloperidone was extensively metabolized prior to excretion. Following oral dosing, free urinary [ $^{14}C$ ]P95 accounted for approximately 0.06% of the dose. Although the total amounts of [ $^{14}C$ ]P95-related metabolites in urine were under <1% of the dose, the ratio of oral to *i.v.* dose suggested that the total [ $^{14}C$ ]P95-related metabolites following oral administration of [ $^{14}C$ ]iloperidone may represent up to approximately 20% of the total metabolic disposition of iloperidone in the rat.

Metabolic pathways of iloperidone in the mouse



Metabolic pathways of iloperidone in the rat







major circulating metabolites included glucuronide of P89 and *S*-P88 glucuronide, accounting for 27% and 30% of the total exposure. The overall metabolite patterns in the plasma and excreta following administration of a single oral dose of [<sup>14</sup>C]*S*-P88 were qualitatively similar to those following administration of a single oral dose of [<sup>14</sup>C]iloperidone.

Single-dose study with *S*-P88 in the dog (Study DMPK(US)R01-1016)

Metabolite patterns were evaluated following single i.v. or oral doses of 1 or 5 mg/kg, respectively, of [<sup>14</sup>C]*S*-P88 to Beagle dogs. *S*-P88 was extensively metabolized in dogs. The 2 major metabolic pathways were benzisoxazole reduction and alpha-methyl oxidation. Carbonyl oxidation, *N*- and *O*-dealkylation, oxidation of the carbon atom of the piperidine ring, and glucuronidation represented minor pathways.

Following the 5-mg/kg oral dose, at least 12 metabolites were detected in the plasma. There was no metabolic conversion of *S*-P88 to *R*-P88 after the oral dose. The major plasma metabolite after oral administration was P33.6 (P94). The 2 major metabolites in urine were P17.6 and P20.8; in feces, they were P26.3 and P39.8. There was no evidence that *S*-P88 and iloperidone were in equilibrium.

P95: Single-dose studies with P 95 in mice and rats. In vivo studies of P95 metabolism in mice and rats were performed upon single i.v. or oral doses of 1 or 5 mg/kg of [<sup>14</sup>C]P95, respectively (Study DMPK(US)R99-1246, Study DMPK(US)R99-1245). In both mouse and rat, there was no rapid conversion between P95 and iloperidone in plasma. Following a single 1-mg/kg i.v. [<sup>14</sup>C]P95 dose in the mouse or the rat, P95 was the major circulating species at 1 and 2 hours postdose. In mice i.v. dosed with [<sup>14</sup>C]P95, P95 represented 64.1% of the radioactivity within 2 hours after administration; after i.v. administration to the rat, P95 only represented 19.5% of the radioactivity in the pooled 1- and 2-hour plasma samples. Major circulating metabolites were the P95 acyl glucuronide and/or the *O*-demethylated P95 glucuronide in the mouse and rat, in addition to open-ring P95 glucuronide in the rat. Prior to excretion, P95 was extensively metabolized. Only 15.3% and 0.3% of the administered dose were excreted unchanged in urine in the mouse and rat, respectively, and no P95 was observed in feces following administration in mice. The major metabolite in feces was open-ring P95 in the mouse and the rat. The following sponsor's tables presents a summary of iloperidone and its P88 and P95 metabolites circulating levels upon <sup>14</sup>C-iloperidone or metabolites P88 and P95 single-dose i.v. and oral administration to rodents, rabbits and dogs.

Iloperidone, P88 and P95 metabolites circulating levels in rodents, rabbits and dogs administered single i.v. (1 mg/kg) and oral (5 mg/kg) dose of <sup>14</sup>C-iloperidone

Species	Sample	Sampling Time or Period	% of Dose in Sample	% of Compound (peak) in Sample		
				Parent (ilo/ open-ring ilo/ ilo+2 amn)	M2 (P95/ open P95)	M2 (P88/ open-ring P88)
Rat (Sprague-Dawley)	Plasma	1, 2, 4, 12, 24 h	NA	1 h: 6.13% (oral)	1 h: 0.60% (oral)	1 h: 0.72% (oral)
				55.1% (i.v.)	No signal (i.v.)	1.99% (i.v.)
				2 h: 4.97% (oral)	2 h: 0.35% (oral)	2 h: 0.50% (oral)
				45.7% (i.v.)	No signal (i.v.)	6.07% (i.v.)
				4 h: 7.02% (oral)	4 h: 0.60% (oral)	4 h: No signal (oral),
				45.1% (i.v.)	0.68% (i.v.)	2.65% (i.v.)
12 h: 2.81% (oral)	12 h: 1.75% (oral)	12 h: 0.72% (oral)				

(Continued)

(Continued)

Species	Sample	Sampling Time or Period	% of Dose in Sample	% of Compound (peak) in Sample		
				Parent (Ho/ open-ring Ho/ Ho+2 amn)	M2 (P95/ open P95)	M2 (P88/ open-ring P88)
Mouse (CD-1)	Plasma	1, 2, 4, 8 h	NA	1 h: 2.59% (oral) 39.3% (i.v.) 2 h: 1.20% (oral) 49.3% (i.v.) 4 h: 5.68% (oral) 22.8% (i.v.) 8 h: 1.51% (oral)	1 h: 4.40% (oral) 1.79% (i.v.) 2 h: 4.89% (oral) 1.31% (i.v.) 4 h: 10.2% (oral) 12.2% (i.v.) 8 h: 3.14% (oral)	1 h: 5.26% (oral) 4.36% (i.v.) 2 h: 1.17% (oral) 3.81% (i.v.) 4 h: 5.05% (oral) 5.12% (i.v.) 8 h: 2.38% (oral)
Rabbit (New Zealand White)	Plasma*	0.5, 1, 2, 4, 8, 12 and 24 h	NA	0.5 h: 16.4% (oral) 1 h: 25.7% (oral) 2 h: 33.0% (oral) 4 h: 31.0% (oral) 8 h: 13.8% (oral) 12 h: 7.08% (oral) 24 h: 1.32% (oral)	0.5 h: 6.93% (oral) 1 h: 12.8% (oral) 2 h: 14.0% (oral) 4 h: 9.88% (oral) 8 h: 3.70% (oral) 12 h: 1.61% (oral) 24 h: BLQ (oral)	0.5 h: 11.0% (oral) 1 h: 15.4% (oral) 2 h: 16.2% (oral) 4 h: 14.5% (oral) 8 h: 5.80% (oral) 12 h: 3.05% (oral) 24 h: BLQ (oral)
Dog (Beagle)	Plasma*	0.5, 1, 2, 3, 4, 6, 8, 12, and 24 h	NA	0.5 h: 9.66% (oral) 1 h: 22.0% (oral) 2 h: 18.2% (oral) 3 h: 19.7% (oral) 4 h: 20.4% (oral) 6 h: 6.34% (oral) 8 h: 5.42% (oral) 12 h: 2.06% (oral) 24 h: 2.93% (oral)	0.5 h: 7.58% (oral) 1 h: 16.3% (oral) 2 h: 27.0% (oral) 3 h: 35.5% (oral) 4 h: 29.9% (oral) 6 h: 25.9% (oral) 8 h: 22.7% (oral) 12 h: 12.2% (oral) 24 h: 13.0% (oral)	0.5 h: BLQ (oral) 1 h: BLQ (oral) 2 h: BLQ (oral) 3 h: BLQ (oral) 4 h: BLQ (oral) 6 h: BLQ (oral) 8 h: BLQ (oral) 12 h: BLQ (oral) 24 h: BLQ (oral)

Iloperidone, P88 and P95 circulating levels in mice and dogs after single i.v. (1 mg/kg) and oral (5 mg/kg) dose of [14C]S-P88 or [14C] P95

## P88

Species	Sample	Sampling Time or Period	% of Dose in Sample	% of Compound (peak) in Sample		
				Parent (S-P88 or S-P88 glu)	M1 (R-P88 or R-P88 glu)	M2 (Ho)
Mouse (CD-1)	Plasma*	0.083, 0.25, 0.5, 1, 4, 8, and 24 h	NA	0.083 h: 86.6% (i.v.) 0.25 h: 211% (oral) 0.5 h: 261% (oral), 64.9% (i.v.) 1 h: 98.2% (oral), 89.3% (i.v.) 4 h: 11.4% (oral), 1.64% (i.v.) 8 h: 1.59% (oral), 0.26% (i.v.) 24 h: 2.54% (oral)	0.083 h: BLD (i.v.) 0.25 h: 16.7% (oral) 0.5 h: 15.9% (oral), 11.4% (i.v.) 1 h: 7.20% (oral), 15.9% (i.v.) 4 h: 1.22% (oral), 1.69% (i.v.) 8 h: 0.93% (oral), 0.78% (i.v.) 24 h: 1.17% (oral)	0.083 h: 14.9% (i.v.) 0.25 h: 25.3% (oral) 0.5 h: 12.6% (oral), 49.5% (i.v.) 1 h: 13.7% (oral), 37.9% (i.v.) 4 h: 5.15% (oral), 4.86% (i.v.) 8 h: 1.27% (oral), 1.27% (i.v.) 24 h: 0 (oral)
Dog (Beagle)	Plasma*	0.083, 0.5, 1, 2, 3, 4, and 6 h	NA	0.083 h: 56.8% (i.v.) 0.5 h: 58.5% (oral), 152% (i.v.) 1 h: 28.6% (oral), 12.0% (i.v.) 2 h: 8.18% (oral), 11.1% (i.v.) 3 h: 4.25% (oral), 11.8% (i.v.) 4 h: 4.89% (oral), 1.6% (i.v.) 6 h: No signal (oral), 0% (i.v.)	0.083 h: 109% (i.v.) 0.5 h: 65.8% (oral), 161% (i.v.) 1 h: 82.4% (oral), 148% (i.v.) 2 h: 54.8% (oral), 100% (i.v.) 3 h: 39.4% (oral), 147% (i.v.) 4 h: 30.9% (oral), 25.7% (i.v.) 6 h: 22.1% (oral), 11.1% (i.v.)	0.083 h: No signal (i.v.) 0.5 h: 8.69% (oral), no signal (i.v.) 1 h: 16.4% (oral), no signal (i.v.) 2 h: 16.0% (oral), no signal (i.v.) 3 h: 17.9% (oral), 13.6% (i.v.) 4 h: 18.6% (oral), 3.3% (i.v.) 6 h: 20.9% (oral), no signal (i.v.)

## P95

Species	Sample	Sampling Time or Period	% of Dose in Sample	Parent (P95)	M1 (O-demethylated P95 glucuronide)	M2 (Open-ring P95 glucuronide)
Rat (Sprague-Dawley)	Plasma*	1-2 h, and 2-8 h	NA	1-2 h: 19.5% 2-8 h: NA	1-2 h: NA 2-8 h: NA	1-2 h: 28.7% 2-8 h: 100%
Mouse (CD-1)	Plasma*	1-2 h	NA	1-2 h: 64.1%	1-2 h: 35.9%	NA

#### 2.6.4.6 Excretion

Excretion profiles in animals (mice, rats, rabbits and dogs) were studied following a single-dose administration of iloperidone or one of the metabolites of iloperidone, P95 or P88. Additionally, one study investigated excretion of iloperidone in milk. The animal studies showed that fecal elimination accounts for the primary excretory path for iloperidone and its 2 metabolites, P95 and P88.

##### Excretion of iloperidone

Excretion of iloperidone was explored in mice, rats, rabbits, and dogs following a single oral and i.v. dose administration of [<sup>14</sup>C]iloperidone (Study DMPK(US)R99-1188, Study DMPK(US)R98-2214, Study DMPK(US)R99-1190, and Study DMPK(US)R99-1189). As summarized in the sponsor's table below, orally or i.v. administered radioactivity was mainly eliminated through feces. After either oral or i.v. administration, more than 80% radioactivity was recovered in urine and fecal samples of all 4 species. Similar results were also observed in rat and dog in another animal study (Study RT1D310791). In this study, the amounts of radioactivity excreted in urine and feces were comparable between male and female rats after oral or i.v. administration.

Excretion data following a single-dose administration of iloperidone

Species	Study No.	Dose (mg/kg)	Admin. Route	Urine (% dose)		Feces (% dose)	
				Radioactivity		Radioactivity	
				0-24 h	0-168 h	0-24 h	0-168 h
Mouse (CD-1)	DMPK(US) R99-1188	5	p.o.	26.3	34.8	32.5	48.8
Rat (Sprague-Dawley)	DMPK(US) R98-2214	5	p.o.	5.14	5.93	68.5	79.4
Rabbit (New Zealand White)	DMPK(US) R99-1190	5	p.o.	18.2	21.4	55.4	66.3
Dog (Beagle)	DMPK(US) R99-1189	5	p.o.	3.04	7.40	0.01	74.7
Mouse (CD-1)	DMPK(US) R99-1188	1	i.v.	38.5	42.8	37.3	44.6
Rat (Sprague-Dawley)	DMPK(US) R98-2214	1	i.v.	5.72	6.91	81.2	96.2
Rabbit (New Zealand White)	DMPK(US) R99-1190	1	i.v.	24.8	28.0	42.8	56.4
Dog (Beagle)	DMPK(US) R99-1189	1	i.v.	8.55	12.0	14.2	74.2

##### Excretion of P95 or P88

Excretion studies with the two major metabolites P95 and P88 were conducted in rats and dogs, respectively. Following a single oral or i.v. dose administration of [<sup>14</sup>C]P95 or

[14C]P88, radioactivity was mainly eliminated through fecal route in both the rat and the dog, as shown in the sponsor's table below.

Single-dose study with P95 in the rat (Study DMPK(US)R99-1245)

The excretion of radioactivity in urine, feces, and cage wash was examined after single i.v. administration of 1 mg/kg of [14C]P95 to rats (Sprague-Dawley and Han Wistar) to investigate strain differences. In Sprague-Dawley rats, excretion was primarily via the biliary route as demonstrated by the fecal recovery of 95% of radioactivity at 72 hours. Approximately 5% of the administered radioactivity was recovered in the urine. Including cage wash, recovery of the radiolabel was 101% (complete) within the 72 hours. In Han Wistar rats, approximately 3% and 56% of the administered radioactivity was recovered in the urine and feces, respectively, at 24 hours. Mass balance was complete within 72 hours post-dose. Excretion of [14C]P95 and its radiolabeled metabolites was rapid since 56% to 59% of the administered radiolabel was recovered within 24 hours post-dose. The results of this study are summarized in the sponsor's table below.

Excretion data following a single-dose administration of P95

Species	Study No.	Dose (mg/kg)	Admin. Route	Urine (% dose)		Feces (% dose)	
				Radioactivity		Radioactivity	
				0-24 h	0-72 h	0-24 h	0-72 h
Rat (Sprague-Dawley)	DMPK(US) R99-1245	1	i.v.	4.21	5.26	51.8	95.4
Rat (Han-Wistar)	DMPK(US) R99-1245	1	i.v.	2.98	NA	56.3	NA

Single-dose study with P88 in the dog (Study DMPK(US)R01-1016)

Radioactivity recovered in urine and feces of Beagle dogs following a single oral or i.v. dose of 1 and 5 mg/kg, respectively, of [14C]S-P88 was determined. Regardless of the route of administration, the radioactivity was excreted mainly in the feces, with recoveries of 76% and 70.3% for the oral and i.v. dose groups, respectively (see sponsor's table below). Radioactivity excretion was rapid with most of the recovered dose (97% of the excretion after the oral dose and 89% of the excretion after the i.v. dose) excreted in the 0- to 48- hour post dose interval. Urinary excretion (0 to 72 hours) was a minor elimination pathway, and accounted for 7.01% and 5.67% of the oral and i.v. dose, respectively.

Excretion data following a single-dose administration of P88

Species	Study No.	Dose (mg/kg)	Admin. Route	Urine (% dose)		Feces (% dose)	
				Radioactivity		Radioactivity	
				0-24 h	0-168 h	0-24 h	0-168 h
Dog (Beagle)	DMPK(US) R01-1016	5	p.o.	6.15	7.01	56.4	76.0
Dog (Beagle)	DMPK(US) R01-1016	1	i.v.	5.12	5.67	31.3	70.3

### Excretion in milk

#### Distribution of [14C]iloperidone into milk of lactating Sprague-Dawley rats (StudyTH1D010694)

The transfer of radioactivity into the milk of lactating rats (Sprague-Dawley) was investigated following a single dose of [14C]iloperidone at 5 mg/kg. As demonstrated in the following sponsor's table, the concentration of radioactivity in milk at 4 hours postdosing was near 10-fold greater than that in plasma at the same time. However, by 24 hours after dosing, concentrations of radioactivity in milk had fallen to values slightly lower than plasma. The metabolic profile in milk was qualitatively similar to that in plasma with 5 of 6 plasma metabolites also observed in milk.

Concentration of radioactivity in rat milk and plasma after a single oral dose of iloperidone

Mean Concentration of Radioactivity ( $\mu\text{g equiv./g}$ )							
	0.5 h	1 h	2 h	4 h	6 h	12 h	24 h
Milk	—*	0.922	1.400	1.469	1.150	0.407	0.044
Plasma	0.405	0.329	0.205	0.150	0.161	0.098	0.064

\*Radioactivity was only recovered in the milk sample of 1 of 3 animals.

2.6.4.7 Pharmacokinetic drug interactions: (See under Pharmacology)

2.6.4.8 Other Pharmacokinetic Studies: NA

#### 2.6.4.9 Discussion and Conclusions

PK studies with iloperidone were conducted in vitro and in vivo with mice, rats, rabbits, dogs, and monkeys. Iloperidone was rapidly absorbed in all species tested following oral and i.v. administrations, but its bioavailability was very low due to a significant first-pass effect. Absorption profiles of iloperidone showed species differences, as demonstrated by the higher exposure (C<sub>max</sub> and AUC) values in dogs in comparison to other species treated at the same dose levels. Oral bioavailability was <1% in rat, 5% in mouse, 19% in both rabbit and dog, and approximately 36% in humans.

Plasma exposure (C<sub>max</sub> and AUC) levels generally increased dose-proportionally in the tested animal species, except for the rat in which exposure increased over-proportionally with dose elevation from 5 to 20 mg/kg. This finding might be possibly related to inhibitory activity of iloperidone to cytochrome P450 (CYP) enzymes. Gender differences in exposure were present in the rat, the mean AUC in female rats being significantly greater than that in males.

Absorption profiles of metabolites P88 and P95 were similar to the parent compound; their absorption was rapid after either oral or i.v. administration. At equal oral doses, bioavailability of P95 (18%) was significantly higher than S-P88 (5%) in mice. P95 plasma exposure (C<sub>max</sub> and AUC) values in the rat increased exponentially in response to dose elevation, while in mice values were increased dose proportionally.

After either oral or i.v. administration, iloperidone and its metabolites distributed rapidly to different organs; the highest drug concentration was generally observed in the liver, kidney, gastrointestinal system, and secretory glandular tissues; placental transfer was limited; and drug concentration in the brain was very low. P95 metabolite did not pass the

blood-brain barrier in the rat. After oral administration to lactating rats, C<sub>max</sub> levels in the plasma and milk were reached in 0.5 and 4 h., respectively; iloperidone concentration was approximately 10 times higher in the milk than in plasma at 4 hours post dosing.

Iloperidone metabolic profiles show some differences across species. The most abundant metabolites in humans (P95 and P88) are found in the species used in toxicology studies. However, in rodents, P95 and P88 are only minor circulating metabolites, in contrast to humans. Results of pharmacology and pharmacokinetic studies that have bearing on the potential toxicological characteristics of metabolite P95, include the following:

- While P95 is the predominant circulating metabolite of iloperidone in humans, comprising 25% to 54% of its total metabolism (Study CIL0522A 2301), in rodents it represents only 3.9% to 5.7% of the total measurable exposure to iloperidone and its metabolites.
- P95 is rapidly absorbed and distributed. Although P95 did not appear to cross the blood-brain barrier as assessed in PK distribution study in rats, in general toxicity studies in rodents and dogs it induced CNS clinical signs similar to those induced by iloperidone, which suggests that the blood-brain barrier is not absolutely impenetrable to P95.
- P95 is rapidly eliminated in rodents; the half-life of P95 is 45 min in mice, 40 min in Sprague-Dawley rats and 100 min in Wistar rats, as compared to a half-life of 23-26 hours for P95 in humans.
- P95 exhibits a similar affinity to iloperidone for human 5-HT<sub>2A</sub> and adrenergic receptor subtypes, while exhibiting a substantially lower affinity for D<sub>1</sub>, D<sub>2</sub>, and D<sub>3</sub> receptor subtypes compared with iloperidone. These results suggest that while P95 appears to have a lesser potential to exert CNS pharmacological effects in comparison to the parent compound, it has a potential to exert other systemic effects through interaction with peripheral receptor subtypes.

Metabolite P88 exposure in rodents and dogs is lower than that of iloperidone, as compared to humans, in whom P88 exposure is greater than that of iloperidone. The in vitro reactions with rabbit and human liver S9 indicated that iloperidone was metabolized with a greater efficiency to the reduced form of iloperidone (P88) than P88 is oxidized to form iloperidone. The equilibrium of the reduction/oxidation reaction favored formation of P88, while in mouse, rat, and dog liver S9, the reaction tended toward formation of iloperidone from P88 rather than reduction of iloperidone to form P88. The preference for P88 oxidation over iloperidone reduction in mouse, rat, and dog are in line with the in vivo study results in these animals, in which iloperidone was found to be more predominant than P88. The low formation of P88 in rat and dog in vivo could also be due to a greater metabolic clearance of iloperidone by biotransformation pathways other than formation of P88.

In vitro metabolic studies showed that iloperidone has stronger inhibitory activity to CYP2D6 and CYP3A4/3A5 compared with either P88 or P95; neither iloperidone nor its metabolites had potential to induce cytochrome P450 enzymes.

Excretion profiles of iloperidone, P85 and P99 were similar. They are mainly eliminated through the feces, in contrast to humans in which urinary excretion is the major elimination pathway.

### 2.6.4.10 Comparative TK summary

**Toxicokinetics: Overview of Toxicokinetics Data**

Test Article: Iloperidone							
Daily Dose (mg/kg/day)	Steady-State AUC (ng·hr/mL)						
	Mice		Rats <sup>a</sup>		Dog <sup>b</sup>		Human <sup>c</sup>
	Male	Female	Male	Female	Male	Female	
2							
4			65	219	NA	NA	29.96
5	799 <sup>d</sup>	172 <sup>d</sup>					
8							64.84
10	1255 <sup>d</sup> , 658.8 <sup>e</sup>	1030 <sup>d</sup> , 617.8 <sup>e</sup>			189.9	NA	
12			208	825			
15	2735 <sup>d</sup>	3115 <sup>d</sup>					
16							133.1
24			1020	3682			231.9
25					659.1	795.1	
48			11275	16163			

a Four-week oral toxicity study (Study 0394-220).

b Four-week oral toxicokinetic study (Study 0494-220).

c Study CILO5220112.

d Thirteen-week oral toxicity study (Study 0193A).

e Twenty-four-month oral carcinogenicity study (Study 988053).

NA = not available due to insufficient data points at the indicated dose.

### 2.6.5 PHARMACOKINETICS TABULATED SUMMARY

Relevant tables are incorporated in the text of the PK Chapter 2.6.4.

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## 2.6.6 TOXICOLOGY

### 2.6.6.1 Overall toxicology summary

#### General toxicology:

Single-dose studies were conducted to evaluate acute toxicity of iloperidone p.o., i.v., or i.p. administration, as well as of orally administered iloperidone metabolite P95 and iloperidone production intermediates. The approximate median lethal dose of oral iloperidone in mice was between 55 and 80 mg/kg (M) and <55 mg/kg (F); in rats it was >480 mg/kg (M) and between 240 and 480 mg/kg (F). The most common clinical signs following iloperidone acute administration were ptosis, decreased spontaneous motor activity, and relaxed scrotum. Additional signs of acute toxicity in some or most animals in some studies included clonic convulsions, prostration, intermittent whole-body tremors, chromodacryorrhea, excessive lacrimation, epistaxis, ataxia. Single oral doses of P95 in the range of 100 to 750 mg/kg in rats did not cause mortality; the clinical signs were similar to those seen with iloperidone, and included decreased motor activity, ptosis, and relaxed scrotum. The approximate median lethal doses of orally administered iloperidone production intermediates in rats were generally higher in comparison to iloperidone.

Repeat dose studies of general toxicity and corresponding toxicokinetic parameters were conducted with iloperidone in mice, rats, rabbits, and dogs. Additionally, toxicology studies were performed in rats and mice with the predominant circulating metabolite of iloperidone in humans, P95, to better characterize its safety and toxicity profiles in view of the lower exposure to this metabolite following iloperidone administration in animal species vs. humans. In humans, P95, comprises 25% to 54% of iloperidone total metabolism (Study CIL0522A 2301). However, in rodents P95 represents only 3.9% to 5.7% of the total measurable exposure to iloperidone and its metabolites. The half-life of P95 is 45 min in mice, 40 min in Sprague-Dawley rats and 100 min in Wistar rats, as compared to a half-life of 23-26 hours for P95 in humans. Due to proportional differences in exposure to this metabolite following iloperidone administration in preclinical species compared with that in human, a number of toxicology studies were performed with P95 direct administration in preclinical species to assess its potential to exert toxic effects. Toxicity studies conducted with P95 in rats included an acute toxicity study, two 13-week oral toxicity studies (in rats and dogs), a 26-week study in rats, a full battery of genetic toxicity tests (Ames test, chromosomal aberration test, and in vivo micronucleus test in rats), and an embryo-fetal development study in rats. An immunotoxicity study was also performed in conjunction with iloperidone

Among all the repeat-dose general toxicology studies on iloperidone and its P95 metabolite, pivotal studies of the longest duration and therefore most relevant to safety evaluation, are the 6-month rat study and the 12-month dog study conducted with iloperidone, and the 6-month rat study conducted with P95 metabolite. These studies are the subject of the present review.

Iloperidone 6-month oral administration to rats (Sprague-Dawley) at doses of 0, 12, 24, and 48 mg/kg/d for 26 weeks induced dose-related clinical signs indicative of CNS

depression (ptosis, decreased motor activity, relaxation of the scrotum, anus, vaginal opening) and decrease of mean body weight in all dose groups throughout the study (from 8% at LD to 32% at HD at the end of the dosing period); hematological changes including lower total leukocyte and lymphocyte counts at LD (F), MD and HD (both genders), and lower platelet counts in MD and HD males; and dose-related serum chemistry deviations including lower triglyceride levels in females at all doses and in HD males and lower glucose levels in females at all doses and in MD and HD males. Prolactin was not determined. Increased incidence and severity of vacuolization of glandular epithelium in the mammary glands of males and females was seen in all dose groups, mammary hypertrophy/hyperplasia in females at MD and HD, testicular degeneration and atrophy at MD and HD, and fatty infiltration in bone marrow sections in HD group. During the 5-week recovery period, an incomplete reversibility was seen for decreased body weight, hematology and mammary glandular epithelium changes. The MTD of iloperidone in this study was 12 mg/kg/d, based on the marked body weight decrease (18-22% vs. control) at the next higher dose tested (24 mg/kg/d). An NOAEL was not reached in this study, as the lowest tested dose (12 mg/kg/d) induced a decrease in body and organ weights, hematological and clinical chemistry changes, and histopathology changes in the mammary glands of males and females.

TK analysis indicated that iloperidone plasma exposure increased over dose-proportionally, with mean maximal concentrations of 24, 89, and 958 ng/ml in males and 18, 351, and 1351 ng/ml in females at LD, MD and HD, respectively. The average concentrations of iloperidone in the females at MD and HD were 3.9 and 1.5 greater than in the corresponding male groups. Plasma levels of iloperidone metabolites P88 8991 (the principal metabolite) and P89 9124 increased dose-dependently, with iloperidone noted at the highest plasma concentration in every dosage group.

Iloperidone 1-year oral administration to beagle dogs at 6, 12, and 24 mg/kg/d induced drug-related clinical signs at all dosages (decreased spontaneous activity, tremors, bizarre behaviors, labored breathing, ptosis, slow response times and/or lack of pupillary reflex); the mid- and high-dose induced ataxia, loss of righting and toe pinch reflex (in single animals), emaciation. Body weight decreases of 7.3% and 9.2% vs. control were registered over the treatment period at LD and HD, respectively. Hematology and clinical chemistry changes were induced dose-dependently at MD and HD, i.e., decreases in mean erythrocyte count and in hemoglobin and hematocrit levels in males and females; lower cholesterol and triglyceride levels in females, and increase in alanin aminotransferase in HD males. No abnormalities were found in any dose group on ECG and auditory examination. Higher mean absolute and relative liver weights and hepatocellular hypertrophy resulting from proliferation of the endoplasmic reticulum were found in males in the HD group, probably secondary to liver enzyme induction. The MTD was 6 mg/kg/d in view of severe clinical signs and emaciation induced at and above the next higher dose of 12 mg/kg/d. NOAEL was not reached in this study as the lowest dose induced decreased body weight and neurological clinical signs.

The mean plasma concentrations of iloperidone at week 49 were 5, 4, and 29.7 ng/ml (for males), and BLQ, 29.7 ng/ml, and 119.8 ng/ml (for females) at 6, 12 and 24 mg/kg/d, respectively, pointing to an over dose-proportional increase in plasma concentrations in females. The principal metabolite was P89 9430, which was detected at higher levels than iloperidone.

Iloperidone metabolite P95 six-month administration to rats (Wistar) at oral doses of 50 and 500 mg/kg/day (yielding P95 plasma exposures of about 2 to 3x and 150 to 400x, respectively, the human P95AUC at iloperidone MRHD of 24 mg/d), induced dose-dependent CNS clinical signs at both dose levels throughout the entire treatment period, similar to those induced by the parent compound (ptosis, decreased motor activity, relaxation of the scrotum, anus, vaginal opening) that are attributable to pharmacological effect. Body weight and weight gain decrease (not due to reduced food intake) was induced at HD in males only (by 11% and 26%, respectively vs. control by the end of treatment); the decrease was not compensated during the recovery period. There were no drug-related abnormal findings in hematology, clinical chemistry (including prolactin plasma levels), or urine analysis. Drug-related histopathology changes, demonstrable by routine histology and/or immunohistochemical method (BrdU labeling) were induced in endocrine glands (pituitary and adrenals in males, thyroid in females, and pancreas in both genders), mammary gland (both genders) and reproductive organs (ovary, uterus, testes, prostate). Statistically significant, treatment-related increase in cell proliferation (increased proportion of cells in S phase of the cell cycle, as assessed by BrdU labeling) was found in pituitary (LDM and HDM), mammary gland (duct and alveoli) in both genders (LDM, HDM, HDF), and the endocrine pancreas in both genders (HDM, HDF). In females, dose-dependent cycle prolongation occurred at both LD and HD, consistent with the finding of vaginal epithelium mucification and decreased uterine weight in the treated groups. In the ovaries, a dose-related interstitial cell hyperplasia was observed at LD and HD that was likely associated with the reduced estrus cycle activity. After the recovery period the prolongation of estrus cycle did not fully normalize, and increased number of corpora lutea was observed in the ovaries of HDF, concordant with increased ovarian weight. In the thyroid, diffuse follicular hyperplasia was induced in both LDF and HDF; this finding did not normalize after the recovery period. In males, pituitary changes (increase in the cell proliferation index at all time points, consistent with an increased pituitary weight) were induced with a dose-related incidence at both tested doses, while adrenal, testicular and secondary sex organ pathology was seen at HD only. In the adrenals, an increased incidence of diffuse cortical hypertrophy and reduced cortical cytoplasmic vacuolation was seen in HDM, concordant with increased absolute and relative adrenal weight and was reversible after the recovery period. Atrophy of testicular seminiferous tubule epithelium (in 2 animals) and an increased incidence of mixed cell inflammation of prostate gland with associated degenerative changes were found at HD. In both genders, increased cellular proliferation in the mammary gland (alveolar hyperplasia, increased secretion and dilatation of mammary ducts) occurred with dose-related severity at LD and HD during treatment and even after recovery period. Most of these histopathology deviations (with the exception of the adrenal, testicular and secondary sex organ pathology in males) were induced in a dose-dependent manner at both tested dose levels, corresponding at LD to plasma exposure (AUC 0-24) approximately 2 to 3x the human exposure at MRHD of 12 mg twice daily. An NOAEL was not reached in either male or female rats in view of the presence of pathomorphological changes in multiple organs/tissues at the lowest tested dose of 50 mg/kg/day. The MTD was <500 mg/kg/day for males in view of excessive body weight decrease at 500 mg/kg/day; for females, an MTD was not reached

Genetic toxicology: The following genetic toxicology studies of iloperidone and its metabolite P95 were conducted in support of this application:

Iloperidone tested negative for genotoxic potential in the following valid in vitro assays in both the presence and absence of metabolic activation: the salmonella/mammalian-microsome reverse mutation assay (Ames test) conducted in *Salmonella typhimurium* (Study 12048-0-401R), the *Escherichia coli*/mammalian-microsome reverse mutation assay (Study 14476-0-402R), and a chromosomal aberration assay conducted in Chinese Hamster Ovary (CHO) cells (Study 14476-0-437). Two additional chromosomal aberration assays in CHO cells evaluated the clastogenicity of iloperidone and micronized iloperidone (Study 1463/63-D5140 and Study 1463/70, respectively). Iloperidone was found to be equivocally positive in inducing chromosomal aberrations in CHO cells in the absence of metabolic activation and reproducibly positive in the presence of metabolic activation; micronized iloperidone was found to induce chromosomal aberrations in CHO cells under both metabolic activation and non-activation conditions; the effect was seen at concentrations claimed to be "excessively cytotoxic" by the sponsor. However, the concentrations associated with clastogenic effects were not "excessively cytotoxic". According to the OECD guidelines, a > 50% decrease in mitotic index, cell confluency, or cell count should be observed at the high concentration. The degree of cytotoxicity cannot be used in this assay to mitigate a positive response.

The maximal reduction of cell number attained at the highest employed iloperidone concentrations was 53% survival (in the presence of metabolic activation); the corresponding survival values attained at the highest concentrations of micronized iloperidone were 63% and 68%, in the absence or presence of metabolic activation, respectively. Having in mind that according to ICH S2A guidelines, the highest concentration in mammalian cell mutation tests should produce at least 80% toxicity (no more than 20% survival), we do not agree with the sponsor that the positive results in the chromosomal aberration assays in CHO cells should be ascribed to excessive cytotoxicity. More importantly, the mean cell survival at the lowest dose of iloperidone positive for chromosomal aberrations was 40-45%.

In vivo genotoxicity potential of iloperidone was evaluated in 4 studies. These included 3 bone marrow micronucleus assays conducted in mice, the first of which was a dose-finding assay of bone marrow cytotoxicity (Studies 14476-0-459PO, 14476-0-455, and 998068) and 1 hepatocyte micronucleus assay conducted in rats (Study 1463/71-D5140, micronized iloperidone). The results of these assays indicated that iloperidone did not induce a significant increase in bone marrow micronuclei in mice, nor did micronized iloperidone induce a significant increase in hepatocyte micronuclei in rats.

Iloperidone metabolite P95 was evaluated for potential genotoxicity and was found to be negative in a battery of 3 tests: an Ames tested conducted in *S. typhimurium* (Study 991801), a chromosomal aberration test conducted in V79 CHO cells (Study 991831), and a bone marrow micronucleus test in rats.

Among iloperidone impurities, ~~—~~ compounds had structures associated with potential genotoxicity:

~~\_\_\_\_\_~~  
~~\_\_\_\_\_~~  
~~\_\_\_\_\_~~ was found to be mutagenic in the Ames test in the presence of metabolic activation, and to induce chromosomal

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aberrations in cultured CHO cells. The other structurally potential genotoxic impurities \_\_\_\_\_ were not tested for genotoxicity. However, for each of these impurities the acceptance criteria are set at the level of NMT \_\_\_\_\_m, so that the overall daily exposure from the sum of these \_\_\_\_\_ genotoxic and potentially genotoxic impurities is NMT \_\_\_\_\_day.

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In conclusion, the results of in vitro and in vivo genotoxicity studies indicate that iloperidone was clastogenic in one in vitro test (chromosomal aberration assay in CHO cells) but was not clastogenic in vivo under the assay conditions used. It is likely that the positive results obtained in chromosomal aberration assay in Chinese Hamster Ovary (CHO) cells in vitro are of little biological relevance, having in mind the negative results obtained in the in vivo micronucleus assays in rat hepatocytes and mouse bone marrow. Iloperidone metabolite P95 was negative for potential genotoxicity in a battery of 3 tests: an Ames, a chromosomal aberration test in CHO cells, and a bone marrow micronucleus test in rats. For iloperidone genotoxic and potentially genotoxic impurities \_\_\_\_\_, the acceptance criteria are set at the level of NMT \_\_\_\_\_ ppm each, so that the overall daily exposure from the sum of these \_\_\_\_\_ genotoxic and potentially genotoxic impurities is NMT \_\_\_\_\_day.

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#### Carcinogenicity:

Iloperidone administration to male and female \_\_\_\_\_ CD-1 (ICR) BR mice at oral doses of 2.5, 5, and 10 mg/kg/d for 2-years caused an increased mortality in females at all dose levels and in males at HD. In males, there was no carcinogenic effect attributable to the test article based on the lack of a dose-response relationship or statistical significance level of the difference in tumor incidence in any of the observed tumor types. In females, the incidence of malignant mammary tumors was significantly increased above the concurrent and historical control range in the low dose group only. On an mg/m<sup>2</sup> basis, there is no safety margin between the low dose employed in the study (2.5 mg/kg/day) and the maximal recommended dose in humans (24 mg/day). However, mammary tumor incidences were not increased in the mid- and high-dose groups, although the duration of treatment was the same in the mid-dose and low dose groups. It is not clear why similar increases in mammary tumor incidences were not seen at higher doses.

In rat \_\_\_\_\_ CD<sup>®</sup>(SD) BR carcinogenicity study conducted at doses of 4, 8, and 16 mg/kg/d for 24 months, the treatment did not affect survival, but induced a dose-related, significant decrease in mean body weights in the dosed groups vs. control, by 13%, 22% and 28% in males and by 10%, 17% and 21% in females at LD, MD and HD, respectively. Body weight gains in the treated groups normalized after the first 3-4 months of study. However, due to the earlier substantial decreases in body weight, the average body weight values for the entire period of the study were significantly lower than the control. There were no signs of systemic toxicity in either clinical pathology or histopathology parameters. Dose- and time-dependent pronounced increase in serum prolactin occurred in both genders, but much more pronounced in females, with associated non-neoplastic proliferative mammary changes in females (glandular hyperplasia and galactocele, all dose levels).

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In males, there was no significant increase in neoplastic incidence or dose-dependence for any of the observed tumor types or combinations. In females, the combined incidences for pancreatic islet cell adenomas and islet cell carcinomas were increased (2,

2, 0, 3, 7 for the two controls, LD, MD and HD, respectively). The incidence value at HD was within historical control range for this species and strain; the dose-response trend analysis showed a p-value of 0.0051 that approached but did not reach the level of statistical significance required for common tumors ( $\alpha=0.005$ ). There was no increase in the incidence of other tumors or tumor combinations of any type, including mammary tumors, although non-neoplastic proliferative mammary changes were increased in all dosed female groups. An MTD was achieved or exceeded in this study, based on decreases in mean body weight of over 10% in all treated groups.

In summary, iloperidone administration to rats for 2 years was not carcinogenic in either males or females.

#### Reproductive and developmental toxicology:

The following studies were performed to assess the reproductive and developmental toxicity of iloperidone:

- Fertility (Segment I) study in rats;
- Embryofetal development (Segment II) study in rats and rabbits;
- Pre- and postnatal development (Segment III) study in rats.

The Segment I rat fertility study evaluated the effect of iloperidone on male and female gonadal function, mating behavior and fertility, as well as on the development of 2 generations of offspring. Oral (gavage) administration at doses of 0, 4, 12, 36 mg/kg/day to Sprague Dawley male and female rats (32/sex/group) for a period starting 10 weeks prior to mating (males) or 2 weeks prior to mating (females) and continuing through mating, gestation, parturition and lactation, resulted in the following drug-related effects: clinical signs (hypoactivity, ptosis and lacrimation at MD and HD; ptosis at LD), significant decreases in mean body weight of F0 males and females at MD and HD during pre-mating and mating periods, as well as throughout gestation and lactation [e.g., the corrected maternal weight at term (terminal body weight minus gravid uterine weight) was significantly lower at HD and MD by 13% and 7%, respectively], female estrous cycle disturbances (all doses, dose-dependently) and reduction in male reproductive organs' weight (mean absolute prostate weight decreased in all dosed groups; mean absolute and relative testis and epididymis weights decreased at HD). Lower female fertility indices, i.e. 72% and 88% were registered at HD and MD, respectively, vs. 100% in control (statistically significant at HD). A significant negative trend was noted for male fertility. The pregnancy rate was lower in MD and HD groups (86%, and 60%, respectively, vs. 100% in control), statistically significant at HD. Mean numbers of corpora lutea and implantation sites were significantly lower at HD in comparison to control; the reduced implantations were secondary to the reduction of corpora lutea and not due to an increased pre-implantation embryonic lethality since preimplantation loss was not significantly different from control in any of the treated groups. The duration of pregnancy was increased (statistically significant at MD and HD). There was an increased prenatal and neonatal mortality in F1 generation, as demonstrated by decreased livebirth index (89% and 83% at MD, and HD vs. 99% in control group, statistically significant), increase in stillborn pup number (18 and 17 at MD and HD vs. 2 in control group, statistically significant) and increase in neonatal deaths (mean viability indices, i.e. N alive on postnatal Day 4/ N liveborn = 80% and 24% at MD, and HD vs. 98% in control group, statistically significant). Embryofetal growth was retarded at HD, as

indicated by a significantly lower mean fetal weight at term vs. control. No external or visceral malformations were observed in the treated groups, but visceral variation rates (dilatation of lateral and third brain ventricles, dilatation of heart ventricles) were increased in HD group. There were no differences in developmental landmarks or in neurobehavioral development of F1 generation as assessed by activity and learning tests. However, very few HD litters were available for growth and behavioral evaluations because of the low pregnancy rate and neonatal deaths. F1 post-weaning growth and development were similar in dosed and control groups. Reproductive performance of F1 animals and F2 generation in utero growth and survival were apparently not affected by treatment. In conclusion, based on the results of this study, a NOEL was not identified, since dose-related estrous cycle disturbances and a decrease in prostate weight of F0 were induced at all dose levels, including the low dose. These effects are not unexpected and are most likely secondary to the pharmacological action of the drug. However, at the low dose (4 mg/kg/day) these effects did not interfere with F0 reproductive capacity, prenatal and postnatal survival, growth and development of F1 generation, or with F1 reproductive capacity and the prenatal growth and survival of the next, F2 generation. Therefore, iloperidone oral dose of 4 mg/kg/day is identified as the NOAEL in the Segment I rat fertility study (this dose is 1.6 times the human exposure at MRHD (24 mg/day) on an mg/m<sup>2</sup> basis). The higher doses employed in this study induced dose-dependent decreased parental F0 fertility (male and female), prolonged gestation, increased prenatal and neonatal lethality of F1 progeny, and reduced F1 pre- and postnatal growth (as indicated by lower body weight at term and postnatally).

Segment II Prenatal developmental toxicity study in rats:

Iloperidone administration to pregnant Wistar rats at doses of 0 (control), 4, 16, and 64 mg/kg/day by oral gavage on Gestation Days 7 through 18 induced at HD and MD a significant dose-dependent reduction in maternal weight (by 18% and 6%, respectively, vs. control at term) and in maternal weight gain (by up to 92% and 34% of control values, respectively). Maternal food consumption was decreased at HD by up to 11% vs. control. Placental weights were statistically significantly lower at HD (by 17% vs. control) and MD (by 7% vs. control). Clinical signs associated with pharmacological action of the test agent (sedation, ptosis) were present in all dosed groups, dose-dependently. The high dose induced a marked (over 5-fold) increase in post-implantation embryonic lethality. Early post-implantation death of all conceptuses occurred in two thirds of the treated HD dams. The surviving HD fetuses exhibited growth retardation (as expressed by significantly decreased fetal weight and crown/rump length at term by 12% and 7%, respectively as compared to control, and retarded skeletal ossification). No increase in external or visceral abnormalities was registered in any of the dosed groups. An increased incidence of minor skeletal abnormalities (fragmented and/or displastic thoracic vertebral centra, supernumerary thoracic rib) was registered in the HD group. No effect on intrauterine developmental parameters was induced at MD and LD.

In conclusion, based on the study findings, iloperidone induces developmental toxicity (expressed as embryofetal lethality, retarded intrauterine development and minor skeletal abnormalities) at oral doses above 16 mg/kg/day. Signs of maternal toxicity (reduced weight and weight gain, reduced placental weight) are induced at and above 16 mg/kg/day. The NOAEL for developmental toxicity in this study is 16 mg/kg/day (6 times the MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).

The predominant circulating iloperidone metabolite in humans (P95) administered to pregnant rats at oral doses of 20, 80 and 200 mg/kg/day during the period of major organogenesis (Gestation Days 7 through 17), produced dose-dependent maternal pharmacological effect (signs of sedation) at all dose levels, but no maternal toxicity. Maternal plasma exposure (AUC) at the high dose was approximately 4 times the mean human plasma AUC of metabolite P95 when the parent compound (iloperidone) was administered at the dose of 24 mg/day (12 mg b.i.d.). The treatment did not induce embryo/fetal mortality or congenital malformations but produced a dose-dependent increase in the incidence of retarded skeletal ossification vs. the concurrent control (predominantly manifested as incomplete ossification of skull bones) at all tested dose levels, ranging from 8% (LD) to 14% (HD). These values, however, were within the historical control range for the tested species and strain.

Segment II Prenatal developmental toxicity study in rabbits:

Iloperidone administration at oral (gavage) doses of 0, 4, 10 and 25 mg/kg/day to pregnant Himalayan rabbits from gestation day 6 through 18 caused maternal mortality at the HD (1/15) and induced dose-dependent drug-related clinical signs (sedation at all dose levels and ptosis at MD and HD). Maternal food intake was reduced at MD and HD. Maternal body weight gain was reduced at HD during the 1<sup>st</sup> week of treatment due to reduced food consumption, resulting in about 5% decrease in mean body weight vs. control on gestation days 13 and 19 (statistically significant). The high dose induced increase in embryo/fetal intrauterine lethality and a decrease in fetal viability at term. No signs of embryo/fetal toxicity or teratogenicity were observed in LD and MD groups. Based on these results, the NOAEL for embryo/fetal toxicity is 10 mg/kg/day (8x the MRHD of 24 mg/day on an mg/m<sup>2</sup> basis).

Segment III Prenatal and postnatal developmental toxicity study in rats

Iloperidone oral administration to pregnant CD rats from gestation day 17 through parturition and lactation up to postnatal day 21 at doses of 4, 16 and 48/36 mg/kg/day, caused maternal general toxicity at HD and MD, expressed in maternal mortality (moribund sacrifices) at HD and significant decrease in maternal body weight and body weight gain at HD and MD during the treatment period, as well as in adverse maternal reproductive effects demonstrated by a significantly prolonged gestation (at MD and HD) and excessive prolongation of parturition (at HD), resulting in impaired viability of F1 generation. The high perinatal- and postnatal mortality (stillbirths and neonatal deaths) in F1 generation at HD and MD can be a consequence, at least in part, to the extended gestation and prolonged parturition through increased time from placental detachment to parturition and a delay or lack of maternal stimulation and nursing. Signs consistent with lack of maternal care were documented in MD and HD. The growth of the surviving F1 offspring was impaired at MD and HD, as demonstrated by the reduced pup weight at birth and weight gain through weaning. However, there was no apparent adverse effect on F1 development, including behavior, sexual maturation and reproductive capacity, at any of the administered dose levels. At the low dose, there were signs of maternal sedation induced by iloperidone exposure, but there was no evidence of adverse effect on parturition, maternal care, pup survival, growth and development. Under the conditions of this study, the NOAEL was 4 mg/kg/day (1.6 times the maximal recommended dose in humans on mg/m<sup>2</sup> basis).

### 2.6.6.2 Single dose toxicity

Six single-dose toxicity studies, listed in the sponsor's table below, were conducted in mice and rats to evaluate the acute toxic effects and mortality associated with iloperidone p.o., i.v., or i.p. administration.

Type of Study	Species and Strain	Method of Administration	Duration of Dosing	Doses (mg/kg) <sup>a</sup>	GLP Compliance	Testing Facility	Study No.
Single-dose toxicity	Mouse	p.o.	Acute	55, 80, 120, 180	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	3090-35
Single-dose toxicity	Mouse	i.v.	Acute	10, 15, 20	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0795
Single-dose toxicity	Mouse	i.p.	Acute	36, 55, 80, 120	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	3190-35
Single-dose toxicity	Rat	p.o.	Acute	120, 240, 480	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0590-35
Single-dose toxicity	Rat	i.v.	Acute	14, 21, 28	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0695
Single-dose toxicity	Rat	i.p.	Acute	60, 120, 240, 480	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0690-35

Single-dose studies have also been conducted to evaluate the toxicity of orally administered iloperidone metabolite P95 (Study 007007, rising-dose study) and iloperidone production intermediates

in rats.

The design and mortality findings of the single-dose toxicity studies of iloperidone are presented in the following sponsor's table.

Summary of iloperidone single-dose toxicity studies

Study No.	Species	Route	Sex	No. of animals/group	Range of doses (mg/kg)	Approx. median lethal dose (mg/kg)	Maximum dose with survivors (mg/kg)
3090-35	Mouse	p.o.	M, F	5	55 to 180	M: between 55 and 80 F: <55	M+F: 80
0795	Mouse	i.v.	M, F	5	10 to 20	M, F: between 15 and 20	M: 15 F: 20
3190-35	Mouse	i.p.	M, F	5	36 to 120	M: between 36 and 55 F: <36	M: 120 <sup>a</sup> F: 55
0590-35	Rat	p.o.	M, F	5	120 to 480	M: >480 F: between 240 and 480	M: 480 F: 240
0695	Rat	i.v.	M, F	5	14 to 28	M, F: between 14 and 21	M: 14 F: 21
0690-35	Rat	i.p.	M, F	5	60 to 480	M: >480 F: between 120 and 240	M, F: 480

<sup>a</sup> An 80% mortality was observed at 120 mg/kg/d, but 100% mortality was observed at 55 and 80 mg/kg/d.  
F = female; i.p. = intraperitoneal; i.v. = intravenous; M = male; p.o. = oral.

The approximate median lethal dose of orally administered iloperidone in mice was between 55 and 80 mg/kg (M) and <55 mg/kg (F); in rats it was >480 mg/kg (M) and between 240 and 480 mg/kg (F). The most common clinical signs associated with iloperidone acute administration included ptosis, decreased spontaneous motor activity,

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and relaxed scrotum. Additional signs of acute toxicity noted in some or most animals in some studies included clonic convulsions, prostration, intermittent whole-body tremors, chromodacryorrhea, excessive lacrimation, epistaxis, ataxia, soft stools; excessive salivation, muscle rigidity and emaciation were infrequently observed.

Single oral doses of P95 in the range of 100 to 750 mg/kg in rats did not cause mortality; the clinical signs were similar to those seen with iloperidone, and included decreased motor activity, ptosis, and relaxed scrotum.

**P95 escalating-dose toxicity study**

Study No.	Species/ Strain	Route	Gender and No./ group	Duration of treatment	Range of doses (mg/kg)	MTD or NOEL	Noteworthy findings
007007	Rat	p.o.	M, F/ 2	Acute	100 to 750	No deaths	<p>Mortality: None.</p> <p>Drug-related clinical signs: ↓ Locomotor activity, ptosis, relaxed scrotum, muscle flaccidity M at 300 and 750.</p> <p>Pathology: None.</p> <p>TK: All rats exposed to P95; AUC<sub>0-24</sub> = 340,000 and 574,000 ng·h/mL for M+F, respectively.</p>

Results of the studies involving production intermediates are presented in the following sponsor's table.

**Iloperidone Manufacturing Intermediates**

Test article	Species/Strain	Method of Administration	Duration of Dosing	Doses	Gender and No. per Group	Approximate Median Lethal Dose or Noteworthy Findings	Study No.
—	Rat/ Sprague- Dawley CD*(SD)B R VAF/PLUS®	p.o.	Acute (single dose)	100 to 500	M, F 5	Median lethal dose: M, F: Between 300 and 500 mg/kg	2194
—	Rat/ Sprague- Dawley (SD)B R VAF/PLUS®	p.o.	Acute (single dose)	5000	M, F 5	Median lethal dose: >5000 mg/kg	1394
—	Rat/ Sprague- Dawley (SD)B R VAF/PLUS®	p.o.	Acute (single dose)	2000 (1600) to 4000 (3200) <sup>c</sup>	M, F 5	Median lethal dose: M, F: Between 2000 (1600) and 3000 (2400) <sup>c</sup> mg/kg	1794
—	Rat/ Sprague- Dawley CD*(SD)B R VAF/PLUS®	p.o.	Acute (single dose)	400 to 1200	M, F 5	M: between 800 and 1200 F: between 400 and 800	2594
—	Rat/ Sprague- Dawley CD*(SD)B R VAF/PLUS®	p.o.	Acute (single dose)	M: 500 to 2000 F: 500 to 1000	M, F 5 <sup>a</sup>	Median lethal dose: M: between 750 and 1000 mg/kg F: between 500 and 750 <sup>b</sup> mg/kg	0994

The approximate median lethal doses of orally administered iloperidone production intermediates in rats were higher in comparison to iloperidone, except for intermediate — which had a median lethal dose similar to iloperidone (between 300 and 500 mg/kg).

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### 2.6.6.3 Repeat dose toxicity

Eleven studies evaluated the repeat dose general toxicity and corresponding toxicokinetic parameters associated with p.o., i.v., and/or inhaled iloperidone in mice, rats, rabbits, and dogs. Additionally, toxicology studies were performed in rats and mice with the predominant circulating metabolite of iloperidone in humans, P95, to better characterize its safety and toxicity profiles in view of the lower exposure to this metabolite following iloperidone administration in animal species vs. humans. The following sponsor's table lists the preclinical repeat-dose general toxicology studies performed with iloperidone and P95 metabolite.

Repeat-dose general toxicology studies performed with iloperidone and P95 metabolite

Overview						Test Article: Iloperidone <sup>a</sup>	
Type of Study	Species and Strain	Method of Administration	Duration of Dosing	Doses (mg/kg) <sup>b</sup>	GLP Compliance	Testing Facility	Study No.
Repeat-dose toxicity	Mouse	p.o.	13 weeks	0, 5 <sup>d</sup> , 10, 20	Yes	Hoechst Marion Roussel Inc, Bridgewater, NJ	0193 (A)
Repeat-dose toxicity	Rat	p.o.	4 weeks	0, 20 <sup>d</sup> , 40, 80	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	2690-35
Repeat-dose toxicity	Rat	p.o.	4 weeks	4, 12 <sup>d</sup> , 24, 48	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0394-220
Repeat-dose toxicity	Rat	p.o.	13 weeks w/ 4-week recovery	0, 4, 10 <sup>d</sup> , 25	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	1289-35
Repeat-dose toxicity	Rat	p.o.	6 months with 5-week recovery	0, 12 <sup>d</sup> , 24, 48	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	1292-35
Repeat-dose toxicity	Rat	i.v.	2 weeks	0, 1, 2, 4 <sup>d</sup>	Yes	Hoechst Marion Roussel Inc, Bridgewater, NJ	0895
Repeat-dose toxicity	Rat	Inhalation	2 weeks	0, 5 <sup>d</sup> mg/m <sup>3</sup>	Indeterminate <sup>c</sup>	Emington Life Sciences (no location given)	94-6081
Repeat-dose toxicity	Rabbit	i.v.	2 weeks	0, 0.75, 1.5, 3 <sup>d</sup>	Yes	Hoechst Marion Roussel Inc, Bridgewater, NJ	0995
Repeat-dose toxicity	Dog	p.o.	4 weeks	4 <sup>d</sup> , 10, 25	Yes	Hoechst Marion Roussel Inc, Bridgewater, NJ	0494-220
Repeat-dose toxicity	Dog	p.o.	13 weeks with 4-week recovery	0, 4, 10 <sup>d</sup> , 25	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	1389-35
Repeat-dose toxicity	Dog	p.o.	12 months	0, 6 <sup>d</sup> , 12, 24	Yes	Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ	0992-0220
<b>Metabolites - Studies Conducted with Iloperidone Metabolite P95</b>							
Metabolites (repeat-dose toxicity)	Mouse	p.o.	4 weeks	0, 50, 250, 750, 1500	Yes	Novartis Pharmaceuticals Corporation, East Hanover, NJ	0170087
Metabolites (rising-dose toxicity)	Rat	p.o.	3 days	100, 300, 750	No	Novartis Pharmaceuticals Corporation, East Hanover, NJ	007007
Metabolites (repeat-dose toxicity)	Rat	p.o.	13 weeks	0, 100, 200, 500	Yes	Novartis Pharmaceuticals Corporation, East Hanover, NJ	TAJ0006
Metabolites (repeat-dose toxicity)	Rat	p.o.	13 weeks with 4-week recovery	0, 50, 200, 500	Yes	Novartis Pharmaceuticals Corporation, East Hanover, NJ	007008
Metabolites (repeat-dose toxicity)	Rat	p.o.	26 weeks w/ 4-week recovery	0, 50, 500	Yes	Novartis Pharmaceuticals Corporation, East Hanover, NJ	017013

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Among all the repeat-dose general toxicology studies on iloperidone and its P95 metabolite, pivotal studies of the longest duration and therefore most relevant to safety evaluation, are the 6-month rat study and 12-month dog study conducted with iloperidone, and the 6-month rat study conducted with P95 metabolite. These studies are the subject of this review.

**Study title: Six-month oral toxicity study of HP 873 in rats with a five-week recovery period**

**Key study findings:**

Iloperidone was administered to Sprague-Dawley — CD® (SD)BR) rats (27/sex/dose) by oral intubation at doses of 0 (vehicle control), 12, 24, and 48 mg/kg/d for 26 weeks. Mortality occurred in single animals in control group (1 male, week 25), MD and HD groups (one MD male at Week 22 and one HD female at Week 13). It was uncertain if the mortality in the treated groups was treatment-related.

Clinical signs (ptosis, relaxed scrotums) occurred in all dosage groups, dose-dependently; decreased activity was seen at MD and HD; and epistaxis, at HD. Dose-related decrease of mean body weight vs. control was seen in all dose groups throughout the study (from 8% at LD to 32% at HD at the end of the dosing period); and lower mean absolute food consumption was noted in the MD and HD groups. Hematological findings included lower total leukocyte and lymphocyte counts at LD (F), MD and HD (both genders), and lower platelet counts in MD and HD males. Serum chemistry findings included dose-related lower triglyceride levels in females at all doses and in HD males; and lower glucose levels in females at all doses and in MD and HD males.

Dose-related decreases in absolute organ weights were noted in all treated groups, corresponding to the lower terminal body weights, with the exception of higher mean absolute adrenal weight in HD males. Pathology examinations revealed dose-related increased incidence and severity of vacuolization of glandular epithelium in the mammary glands of males and females in all dose groups, mammary hypertrophy/hyperplasia in females at MD and HD, testicular decreased size, degeneration and atrophy at MD and HD, minimal to mild active/chronic prostate inflammation at HD, and fatty infiltration in bone marrow sections especially in HD group.

During the 5-week recovery period, an incomplete reversibility of decreased mean body weights was seen. Absolute mean food consumption generally recovered to control values. Some hematological abnormalities and differences in organ weights remained during the recovery period. Vacuolization of mammary glandular epithelium was still present after recovery, but to a lesser extent than seen at the 26-week time point.

TK analysis indicated that iloperidone plasma exposure increased over dose-proportionally, with mean maximal concentrations of 24.4, 89.5, and 958.3 ng/ml in males and 18.3, 350.7, and 1351.4 ng/ml in females at LD, MD and HD, respectively. The average concentrations of iloperidone in the females at MD and HD were 3.9 and 1.5 greater, respectively than in the corresponding male groups. Concentrations of iloperidone metabolites P89 9124, and P88 8991 increased in a dose-related manner, with iloperidone noted at the highest plasma concentration in every dosage group. P88 8991 was the principal metabolite; P89 9124 was slightly lower and P89 9430 was generally below the limits of quantification or not detected.

The MTD of iloperidone in this study is 12 mg/kg/d, based on a marked body weight decrease (18-22% vs. control) at the next higher dose tested (24 mg/kg/d).

A NOAEL was not reached in this study, as the lowest tested dose (12 mg/kg/d) induced a decrease in body and organ weights, hematological and clinical chemistry changes (mostly in females), and histopathology changes (vacuolization of glandular epithelium

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in the mammary glands of males and females). At equal oral doses, iloperidone plasma exposure in females was higher than in males.

**Study no.:** 1292-35

**Conducting laboratory and location:** Drug Safety, Hoechst-Roussel Pharmaceuticals Inc, Somerville, NJ

**Date of study initiation:** August 5, 1992

**GLP compliance:** Yes. **QA reports:** yes

**Drug, lot #, and % purity:** Iloperidone, Batch/Lot RC 4840, 99.8%;

**Formulation/vehicle:** 2% aqueous starch

**Methods**

**Dosing:**

Species/strain: Rat/ Sprague-Dawley ---:CD® (SD)BR

#/sex/group or time point (main study): 27

Satellite groups used for toxicokinetics or recovery: TK: 10 sex/dose group;

Recovery: 6 sex/dose group

Age: 9 weeks

Route, form, volume, and infusion rate: Oral gavage, suspension in 2 % aqueous starch.

Doses in administered units: 12, 24, and 48 mg/kg/d

Note: Iloperidone dosage levels for this study were selected on the basis of preceding 4-week and 13-week studies in the same species and strain, conducted at doses of 20, 40 and 80 mg/kg/d and 4, 10 and 25 mg/kg/d, respectively. In the 4-week study, dose-dependent microscopic findings were induced at all dose levels and included changes in the mammary glands, bone marrow and testes. In the 13-week study, there was a dose-related decrease in uterine weight, but no histopathology findings were registered at any of the doses administered.

**Observations and times:**

Mortality, clinical condition (daily), neurologic examinations (corneal reflex, pupillary light response, reflexes of placement, righting and toe pinch) and auditory test (visible pinna movement evoked by sound stimuli of 12 kHz), ophthalmoscopic examination (prior to treatment, at midterm and end of dosing period), body weight and food consumption, hematology, serum chemistry, urinalysis (weeks 2, 6, 14 and 26), organ weights, gross and microscopic pathology. Bone marrow examinations (200-cell differential counts; myeloid/erythroid ratios, cellularity grade and percent) were limited to the control and HD group (19-21 animals/sex/group). Blood samples for toxicokinetic evaluation were taken from 10 rats/sex/dose at 3 hours post dose after 22 weeks of dosing. After 26 weeks of treatment, 21 animals/sex/ dose group were euthanized, and the remaining rats (6/group) were removed from treatment to determine the reversibility of findings during a 5-week recovery period.

**Results:**

Mortality: Spontaneous deaths occurred in 1 control male (week 25), 1 MDM (week 22) and 1 HDF (week 13). No gross necropsy changes were registered.

Clinical signs: Ptosis, relaxed scrotums occurred in all dosage groups, dose-dependently; decreased activity was seen at MD and HD; and epistaxis, at HD. Neurology and auditory examination revealed no consistent drug-related changes.

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Body weights: Significant, dose-related decrease of mean body weight gains vs. control were registered in both genders beginning at week 1 in HD group, and between weeks 2 and 19 in MD and LD groups. By the end of the dosing period, the mean body weights in LD, MD, and HD groups were lower than control by 8%, 23% and 32% (M) and by 9%, 18% and 30% (F), respectively. At the end of the recovery period, the mean body weights of MD group (males) and HD (both genders) remained significantly lower than control (see sponsor's table below).

Iloperidone 6-month study in rats: Mean body weights

Treatment period

TEST WEEK	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
	0	12	24	48	0	12	24	48
PRE	222	227	224	223	190	187	189	186
0	311	316	316	312	226	223	223	222
1	361	363	367	358 *	247	249	246	229 *
2	400	394	361 *	352 *	266	266	266	246 *
3	428	418	400 *	358 *	276	263	270	238 *
4	456	442	414 *	374 *	290	293	285	268 *
5	485	463	427 *	380 *	304	305	290	268 *
6	498	476	433 *	387 *	307	312	292	264 *
7	518	487	443 *	391 *	316	313	296 *	263 *
8	530	496 *	447 *	394 *	327	318	298 *	267 *
9	545	507 *	466 *	402 *	335	327	304 *	267 *
10	558	519 *	466 *	412 *	343	333	306 *	272 *
11	573	533 *	473 *	418 *	350	337	311 *	274 *
12	582	543 *	478 *	421 *	356	338	311 *	274 *
13	597	549 *	482 *	419 *	360	339	311 *	272 *
14	600	557 *	486 *	422 *	363	348	316 *	273 *
15	608	561 *	486 *	421 *	366	348	313 *	274 *
16	619	571 *	496 *	429 *	372	349	316 *	275 *
17	626	575 *	491 *	427 *	373	350	318 *	276 *
18	636	586 *	501 *	436 *	382	366	322 *	280 *
19	645	593 *	501 *	436 *	388	361 *	322 *	279 *
20	647	591 *	489 *	436 *	396	363 *	318 *	274 *
21	657	598 *	504 *	441 *	396	363 *	322 *	279 *
22	667	606 *	505 *	443 *	401	364 *	325 *	279 *
23	671	612 *	511 *	446 *	403	367 *	326 *	281 *
24	680	618 *	517 *	446 *	407	368 *	328 *	278 *
25	679	622 *	517 *	450 *	409	377 *	333 *	287 *
26	679	624 *	526 *	461 *	410	373 *	336 *	289 *

n = 27 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA.  
\* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

Recovery period

TEST WEEK	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
	0	12	24	48	0	12	24	48
27*	696	667	528 *	483 *	419	378	346 *	295 *
28	712	662	582 *	554 *	423	378	368	308 *
29	715	665	595 *	586 *	424	378	364	307 *
30	717	675	610 *	585 *	432	382	378	318 *
31	715	670	609 *	586 *	430	384	376	316 *

\* = THE SAMPLE SIZE FOR WEEK 27 VARIES FROM 11 TO 15 VALUES, BECAUSE THE WEEK 26 NECROPSY EXTENDED INTO WEEK 27.  
n = 6 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA ( EXCEPT FOR WEEK 27).  
\* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

Food consumption: Dose-related decrease in absolute mean food consumption were registered throughout the dosing period in MD and HD, beginning at week 1 in HD and at week 4 in MD. Significantly lower absolute food consumption values were registered occasionally in LDF. During the recovery period, the absolute food consumption was comparable to or greater than control.

Ophthalmoscopy: No drug-related findings were registered

Electrocardiography: Not performed

Hematology: Drug-related differences in mean hematology values included higher erythrocyte counts (males, all dose groups, dose-dependent), slightly higher hemoglobin

and hematocrit values (males), dose-dependent decrease in total leucocyte, lymphocyte and platelet counts (statistically significant in MDM and HDM), as shown in the sponsor's table below. These hematology changes persisted after the 5-week recovery period. Although the differences were statistically significant, many of the values were within normal limits of variation.

Iloperidone 6-month study in rats: Mean hematology values

DETERMINATION (UNITS)	TEST WEEK	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
		0	12	24	48	0	12	24	48
ERYTHROCYTE COUNT (MILLION/CUBIC MM)	PRE	7.43	7.34	7.22	7.24	7.43	7.40	7.42	7.54
	2	8.25	8.75*	8.92*	9.38*	8.44	8.59	8.55	8.92
	6	9.06	9.49*	9.72*	9.60*	8.71	8.88	8.95	9.07
	14	9.04	9.43	9.53*	9.47*	8.49	8.63	8.53	8.67
	26	9.02	9.42*	9.35	8.63	8.05	8.49	8.26	8.00
	31	8.90	8.85	8.78	8.57	7.76	8.18*	8.33*	8.40*
HEMATOCRIT (PER CENT)	PRE	45	44	43	43	42	42	42	42
	2	49	51*	52*	54*	49	50	50	51
	6	50	51	53*	52	49	50	51	50
	14	48	50*	51*	51*	47	48	48	48
	26	45	48	49*	47	44	47*	46	44
	31	45	45	45	45	42	44	45*	45*
HEMOGLOBIN (GRAMS/DECILITER)	PRE	14.9	14.7	14.5	14.6	15.1	14.8	15.1	14.8
	2	16.0	16.9*	17.2*	17.7*	16.3	16.7	16.9	16.9
	6	16.8	17.3	17.6*	17.3	16.6	16.9	17.2	16.9
	14	16.3	17.0*	17.4*	17.2*	16.2	16.5	16.6	16.4
	26	16.0	16.7	16.8	16.3	15.2	16.1*	16.0*	15.4
	31	16.0	15.9	16.1	16.1	15.3	15.8	16.1	16.4*
TOTAL LEUKOCYTE COUNT (1000/CUBIC MM)	PRE	9.7	9.0	10.2	8.4	8.9	8.0	8.7	9.6*
	2	11.5	11.4	11.9	10.0	8.5	9.9	8.8	8.4
	6	12.5	12.0	12.5	9.8	8.0	10.9*	8.4	8.7
	14	12.5	11.3	10.5	8.3*	6.2	8.4*	7.9*	7.9
	26	10.2	10.4	6.9*	6.9*	5.9	6.3	5.7	5.1
	31	12.3	10.8	7.3*	9.3*	5.7	8.8	6.3	6.7
LYMPHOCYTES (1000/CUBIC MM)	PRE	9.0	8.0	9.1	7.6	6.3	7.3	7.8	8.4*
	2	10.6	10.0	10.8	8.9	7.5	9.1	8.0	7.4
	6	11.2	10.8	10.8	8.5	7.2	10.1*	7.6	7.6
	14	11.0	9.6	8.8	7.2*	5.4	7.5*	7.1*	6.8
	26	8.9	8.4	6.0*	5.9*	5.1	5.4	5.0	4.2
	31	11.4	9.2	6.2*	7.4*	4.9	7.7	5.6	5.8
PLATELET COUNT (1000/CUBIC MM)	PRE	1216	1259	1276	1195	1209	1153	1174	1300
	2	1129	1126	1076	1016*	1052	1118	1161	1167
	6	1086	1030	1018	978*	968	1039	1051	1099
	14	1105	1010	998*	931*	981	1038	1005	1005
	26	1078	1025	993	926*	866	925	971	995
	31	1032	955	968	1050	982	873	915	1032

n = 12 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA  
 \* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

Evaluation of bone marrow smears (see sponsor's table on the next page) showed a mild but statistically significant decrease in overall bone marrow cellularity in the HD group, in both males and females (lower dosage groups were not evaluated). HD males had a higher percentage of lymphogenous cells vs. control, while in the females the percentage of lymphogenous cells was lower in comparison to control. These opposite effects appeared to be random and unrelated to treatment. HD females had a higher percentage of erythropoietic cells than controls, but that did not appear to be a treatment-related effect.

After the recovery period (when food consumption normalized) no significant differences were present between the HD and control animals.

Iloperidone 6-month study in rats: Bone marrow examination  
End of treatment period

MALES - SIX MONTHS' NECROPSY			
Cell Type	0	48	
	mg/kg	mg/kg	
Lymphogenous	7.673	10.421	NP
Erythropoietic	47.873	48.242	OP
Neutrophilic	34.125	31.553	OP
Eosinophilic	4.350	4.921	ONP
Myelogenous	42.300	40.024	OP
M:E	0.947	0.914	OP
Cellularity	64.048	52.419	NP

FEMALES - SIX MONTHS' NECROPSY			
Cell Type	0	48	
	mg/kg	mg/kg	
Lymphogenous	13.447	7.405	NP
Erythropoietic	39.237	47.474	NP
Neutrophilic	41.395	41.714	OP
Eosinophilic	2.379	2.143	OP
Myelogenous	44.711	44.371	OP
M:E	1.156	1.078	ONP
Cellularity	58.571	49.742	NP

Key: O= Not statistically significantly different from control.  
 \* = Statistically significant at p<0.05. \*\*=Statistically significant at p<0.01. P=Parametric analysis.  
 NP=Nonparametric analysis.

After 5-week recovery period

MALES - REVERSAL NECROPSY			
Cell Type	0	48	
	mg/kg	mg/kg	
Lymphogenous	5.400	9.417	OP
Erythropoietic	54.200	48.467	OP
Neutrophilic	30.300	31.583	OP
Eosinophilic	2.500	2.750	OP
Myelogenous	37.200	40.500	ONP
M:E	0.784	0.940	ONP
Cellularity	42.000	40.000	ONP

FEMALES - REVERSAL NECROPSY			
Cell Type	0	48	
	mg/kg	mg/kg	
Lymphogenous	8.200	3.800	ONP
Erythropoietic	45.200	47.800	OP
Neutrophilic	42.400	42.600	OP
Eosinophilic	2.700	2.400	OP
Myelogenous	45.900	46.000	OP
M:E	1.036	1.012	OP
Cellularity	64.000	58.000	OP

Clinical chemistry:

Significant drug-related differences in mean serum chemistry included decrease in triglycerides in HD males (weeks 14 and 26) and females in all dose groups (week 26), dose-dependently. At the end of recovery period, low triglyceride values persisted in LD and MD female groups. Lower glucose values were registered in all female dose groups from 2<sup>nd</sup> week onward and in the MD and HD males from the 6<sup>th</sup> week onward. The blood glucose changes were reversible after cessation of treatment.

Iloperidone 6-month study in rats: Mean clinical chemistry values

DETERMINATION (UNITS)	TEST WEEK	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
		0	12	24	48	0	12	24	48
ALANINE AMINOTRANSFERASE (IU/LITER)	PRE	44	51	48	51	38	31	45	39
	2	49	67*	62*	68*	50	50	46	59
	6	95	63	63	73	52	55	47	60
	14	80	62	68	69	63	64	58	54
	26	70	74	60	61	67	80	49	59
	31	81	72	63	80	176	91	116	70
ASPARTATE AMINOTRANSFERASE (IU/LITER)	PRE	136	128	109	101*	147	125	161	125
	2	110	133*	127	133*	111	114	134*	118
	6	176	132	130	125	109	111	125	121
	14	142	126	132	136	127	121	127	136
	26	115	120	99	113	101	117	90	103
	31	139	107	112	119	298	151	161	139
ALKALINE PHOSPHATASE (IU/LITER)	PRE	332	295	298	292	166	187	218	191
	2	179	172	175	181	114	116	119	112
	6	130	125	140	135	71	62	91	85
	14	119	102	126	124	59	62	73	75
	26	77	81	100	92	39	38	61	63*
	31	75	86	89	94	64	42	45	59
TOTAL BILIRUBIN (MG/DECILITER)	PRE	0.3	0.1	0.2	0.2	0.2	0.3	0.4	0.3
	2	0.3	0.4	0.4	0.4	0.3	0.3	0.3	0.3
	6	0.3	0.4	0.4	0.3	0.5	0.3*	0.3*	0.3
	14	0.4	0.4	0.4	0.3	0.5	0.4	0.4	0.4
	26	0.4	0.4	0.4	0.4	0.4	0.4	0.3*	0.3*
	31	0.6	0.6	0.5*	0.4*	0.8	0.6	0.5*	0.6
CHOLESTEROL (MG/DECILITER)	PRE	55	52	55	52	51	43	58	58
	2	57	57	69	65	78	80	72	91
	6	60	60	72	67	76	82	74	79
	14	75	66	85	76	99	108	88	86
	26	89	78	85	73	92	98	82	80
	31	96	85	77	56*	105	90	86	96
TRIGLYCERIDES (MG/DECILITER)	PRE	90	77	82	67	48	72	73	70
	2	159	131	176	154	133	122	118	137
	6	154	167	180	145	167	139	151	137
	14	234	194	212	167*	307	206	257	183
	26	197	212	189	135*	228	163*	140*	114*
	31	277	263	212	179	339	166*	186*	226
GLUCOSE (MG/DECILITER)	PRE	75	72	71	66	54	75	72	83*
	2	107	91	98	97	113	104	83*	96*
	6	114	106	101*	97*	115	108	95*	92*
	14	128	117	113*	99*	124	109	107	105*
	26	124	128	119	108*	128	114*	107*	103*
	31	123	128	112	112	117	116	112	112

n = 12 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA  
 \* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

Urine analysis showed no differences between treated and control groups.

Organ weights: Dose-related significant decrease in mean terminal body weight at necropsy was seen in all dose groups (by 9%, 25% and 36% in males and by 9%, 20% and 31% in females in LD, MD and HD groups, respectively. Most of the organs had lower absolute mean organ weights and higher relative mean organ weights corresponding to the lower terminal body weight. However, for the adrenals, dose-related

increases in mean absolute and relative adrenal weights were registered in the HD males. After the recovery period, the decreased organ weights ameliorated but did not recover completely and remained statistically significantly lower than control at the MD (males) and HD (both genders). The mean absolute adrenal weight in HDM was still higher vs. control, but the difference was no longer statistically significant.

Iloperidone 6-month study in rats: Mean absolute and relative organ weights  
End of the dosing period

DETERMINATION (UNITS)	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
	0	12	24	48	0	12	24	48
<b>ORGAN WEIGHTS (GMS)</b>								
LIVER	17.821	17.380	14.380*	11.312*	10.860	9.839	8.551*	8.448*
KIDNEYS	4.205	3.842	3.430*	3.010*	2.681	2.514	2.357*	2.290*
HEART	1.977	1.810	1.618*	1.573*	1.260	1.263	1.189	1.155*
BRAIN	2.232	2.112*	2.125*	2.083*	1.977	2.002	2.001	1.931
GONADS	3.720	3.767	3.527	3.374*	0.089	0.103	0.099	0.095
PROS/UTS	1.336	1.215	1.119	1.062	0.637	0.486*	0.439*	0.408*
ADRENALS	0.066	0.071	0.070	0.085*	0.082	0.077	0.084	0.085
THYROIDS	0.026	0.026	0.024	0.023	0.020	0.021	0.019	0.018
PITUITARY	0.015	0.014	0.014	0.013	0.017	0.021*	0.018	0.016
SPLEEN	0.916	0.862	0.727*	0.652*	0.581	0.635	0.586	0.519
TERMINAL BODY WEIGHT (GMS)	658	600*	496*	418*	400	363*	319*	274*
<b>ORGAN/BODY WT. RATIOS (GMS%)</b>								
LIVER	2.692	2.874	2.867	2.710	2.720	2.712	2.663	3.097*
KIDNEYS	0.640	0.643	0.693	0.723*	0.663	0.695	0.741*	0.840*
HEART	0.301	0.303	0.329	0.379*	0.321	0.349	0.374*	0.424*
BRAIN	0.339	0.358	0.435*	0.502*	0.513	0.557	0.630*	0.708*
GONADS	0.569	0.636	0.721*	0.810*	0.023	0.029*	0.031*	0.035*
PROS/UTS	0.203	0.207	0.223	0.254*	0.164	0.136	0.136	0.150
ADRENALS	0.010	0.012	0.014*	0.020*	0.021	0.022	0.026*	0.032*
THYROIDS	0.004	0.004	0.005*	0.005*	0.005	0.006	0.006	0.007
PITUITARY	0.002	0.003*	0.003*	0.003*	0.004	0.006*	0.006*	0.006*
SPLEEN	0.139	0.144	0.147	0.157*	0.147	0.175*	0.183*	0.190*

n = 21 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA  
\* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

After recovery period

DETERMINATION (UNITS)	MALES: DAILY DOSE IN MG/KG				FEMALES: DAILY DOSE IN MG/KG			
	0	12	24	48	0	12	24	48
<b>ORGAN WEIGHTS (GMS)</b>								
LIVER	17.792	18.142	16.246	15.005	10.904	9.732	10.143	9.782
KIDNEYS	4.290	3.870	3.808	3.728*	2.688	2.610	2.586	2.406
HEART	1.906	1.780	1.830	1.743	1.296	1.227	1.270	1.186
BRAIN	2.164	2.160	2.138	2.210	2.046	2.045	2.060	1.982
GONADS	3.870	3.777	3.685	3.807	0.101	0.096	0.102	0.099
PROS/UTS	1.151	1.182	1.412	1.255	0.550	0.712	0.621	0.611
ADRENALS	0.069	0.063	0.064	0.076	0.078	0.082	0.078	0.086
THYROIDS	0.037	0.029	0.027	0.029	0.024	0.022	0.018	0.024
PITUITARY	0.021	0.018	0.017	0.022	0.022	0.022	0.020	0.024
SPLEEN	1.022	0.888	0.770*	0.870	0.684	0.700	0.682	0.568
TERMINAL BODY WEIGHT (GMS)	692	646	587*	561*	413	364	357	301*
<b>ORGAN/BODY WT. RATIOS (GMS%)</b>								
LIVER	2.573	2.791	2.761	2.637	2.736	2.685	2.861	3.243
KIDNEYS	0.820	0.615	0.653	0.667	0.659	0.717	0.706	0.798
HEART	0.277	0.276	0.313	0.311	0.310	0.339	0.358	0.386
BRAIN	0.314	0.336	0.367*	0.395*	0.507	0.567	0.580	0.655*
GONADS	0.561	0.589	0.633	0.682	0.025	0.026	0.029	0.033
PROS/UTS	0.166	0.184	0.248	0.225	0.183	0.200	0.174	0.201
ADRENALS	0.010	0.020	0.011	0.014*	0.019	0.023	0.022	0.029*
THYROIDS	0.005	0.005	0.005	0.005	0.006	0.006	0.005	0.008
PITUITARY	0.003	0.003	0.003	0.004	0.006	0.006	0.006	0.006
SPLEEN	0.148	0.137	0.132	0.158	0.166	0.193	0.192	0.190

n = 6 VALUES OR LESS PER MEAN AS INDICATED IN THE INDIVIDUAL DATA  
\* = SIGNIFICANTLY DIFFERENT FROM CONTROL (P<0.05)

Gross- and histopathology:

Incidences of microscopic findings are reproduced in the following sponsor's table.

Iloperidone 6-month study in rats: Microscopic findings

Males

GROUP:		0 mg/kg	12mg/kg	24mg/kg	48mg/kg
NUMBER OF ANIMALS:		22	21	22	21
<b>PROSTATE GLAND</b>	# Ex	18	18	19	18
INFILTRATING CELL, LYMPHOCYTE		1	0	0	0
VACUOLIZ-EPITHELIAL		1	18	16	18
<b>TESTES</b>	# Ex	22	21	22	21
DEGENERATION		0	0	1	2
<b>EPIDIDYMS</b>	# Ex	22	21	22	21
INFILTRATING CELL, LYMPHOCYTE		1	0	0	0
<b>PROSTATE</b>	# Ex	22	21	22	21
DISPERSED CONTENTS		2	0	0	0
INFLAMMATION, ACUTE		0	0	0	2
INFLAMM, ACTIVE/CHRONIC		2	0	1	11
INFLAMMATION, CHRONIC		2	0	0	0
<b>BONE MARROW</b>	# Ex	22	21	22	21
INFILTRATION, FATTY		7	3	4	21
HYPERPLASIA, MYELOID		0	0	1	0
<b>LIVER</b>	# Ex	22	21	22	21
INFILTRATING CELL, LYMPHOCYTE		7	3	2	2
INFLAMMATION, MIXED CELL		4	0	0	3
INFLAMMATION, ACUTE		1	1	0	0
INFLAMMATION, CHRONIC		0	0	1	0
INTRACELLULAR HEMATOPOIESIS		11	16	13	18
NECROSIS		2	0	0	0
VACUOLIZ, CENTRILOR/MIDZONAL		2	0	0	1
CONGESTION		0	0	1	0
<b>ADRENAL CORTEX</b>	# Ex	22	21	22	21
		0	0	1	0
<b>ADRENAL MEDULLA</b>	# Ex	19	18	19	17
<b>THYROID</b>	# Ex	22	21	22	21
INFILTRATING CELL, LYMPHOCYTE		0	0	1	1
FOLLICLE-DILATATION		1	0	0	0
ULTRACRANIAL CYST		2	0	0	0
HYPERPLASIA, FOLLICULAR CELL		1	0	0	0
AUTOLYSIS		0	0	1	0
<b>PARATHYROID</b>	# Ex	14	14	14	14
		0	0	1	0
<b>PITUITARY</b>	# Ex	22	21	22	20
PARS INTERMEDIA-CYST		0	3	1	1
PARS DISTALIS-CYST		1	1	1	0
PARS DISTALIS-HYPERPLASIA		0	0	1	0
<b>PANCREAS</b>	# Ex	22	21	22	21
INFLAMMATION, CHRONIC		9	0	0	5
ATROPHY		0	0	0	3
PIGMENT		1	0	0	0

Continued

Microscopic findings, Females

GROUP: NUMBER OF ANIMALS:	0 mg/kg	12mg/kg	24mg/kg	48mg/kg
	22	21	22	21
<b>MAMMARY GLAND</b>	# Ex 22	21	21	22
FIBROADENOMA	2	0	0	0
INFLAM, ACTIVE/CHRONIC	1	0	0	0
VACUOLIZ-EPITHELIAL	2	14	10	21
HYPERTROPHY/HYPERPLASIA	0	2	3	0
<b>BONE MARROW</b>	# Ex 22	21	21	22
INFILTRATION, FATTY	8	3	3	19
<b>LIVER</b>	# Ex 22	21	21	22
INFILTRATING CELL, LYMPHOCYTE	4	4	6	7
INFLAMMATION, MIXED CELL	5	3	3	4
INFLAMMATION, CHRONIC	1	0	0	0
EXTRAMOLLARY NECROPORESIS	0	4	11	3
NECROSIS	1	0	0	0
VACUOLIZ, CENTRILON/REGIONAL	1	0	0	0
CONGESTION	0	0	0	2
BILIARY CYST(S)	1	0	0	0
<b>ADRENAL CORTEX</b>	# Ex 22	0	0	22
CYST	1	0	0	2
VACUOLIZATION	2	0	0	1
CONGESTION	0	0	0	1
HEMATOCYST	1	0	0	0
<b>ADRENAL MEDULLA</b>	# Ex 18	0	0	14
<b>THYROID</b>	# Ex 22	0	0	22
ULTRABRANCHIAL CYST	7	0	0	0
<b>PARATHYROID</b>	# Ex 14	0	0	14
<b>PITUITARY</b>	# Ex 22	21	21	22
PARS INTERMEDIA-CYST	1	1	1	0
PARS DISTALIS-CYST	1	0	1	0
PARS DISTALIS-HYPERPLASIA	0	2	0	0
<b>PANCREAS</b>	# Ex 22	0	0	22
INFILTRATING CELL, LYMPHOCYTE	0	0	0	2
INFLAMMATION, CHRONIC	3	0	0	4
<b>ISLETS OF PANCREAS</b>	# Ex 22	0	0	22

Notable morphology findings were mammary gland epithelial vacuolization in both genders at all dose levels with dose-related incidence and severity, dose-related reduced size of testicles and testicular degeneration/atrophy (the latter in single males at MD and HD), higher incidence of prostate inflammation at HD, fatty infiltration of bone marrow in both genders at HD. The bone marrow and prostate changes were "greatly ameliorated" after the recovery period, but the changes in the mammary glands did not recover. There were no histopathology changes in other endocrine and glandular organs; and no drug-related neoplastic lesions were observed.

Toxicokinetics:

The results of plasma analysis indicate that the concentrations of iloperidone and its metabolites P88 8991, P89 9430 and P89 9124 increased dose-dependently, with the highest plasma levels for the parent compound. P88 was the principal metabolite; levels of P89 9124 were slightly lower, and P89 9430 was generally below the limits of quantification or not detected. The mean plasma concentrations of iloperidone in the females were higher than those in the males, by approximately 4x and 1.5x at MD and HD, respectively. The mean plasma concentrations of iloperidone and its metabolites are shown in the sponsor's table below.

Plasma concentrations of iloperidone (HP 873) and its metabolites P88 and P89 in the 6-month rat study (mean, ng/ml ±SD)

DOSAGE (MG/KG)		P89 9430	P89 9124	P88 8991	HP 873
HP 873					
<b>MALES:</b>					
0		BLQ,NR	BLQ,NR	BLQ	BLQ,NR
12		BLQ,NR	1.1 <sup>ⓐ</sup>	0.5 ± 1.0	24.4 ± 32.9
24		9.5 ± 10.7	5.4 ± 5.4	3.5 ± 3.0	89.5 ± 117.2
48		40.6 ± 9.1	28.2 ± 10.1	41.1 ± 21.8	856.3 ± 598.9
<b>FEMALES:</b>					
0		0.4 <sup>ⓑ</sup>	BLQ	BLQ	BLQ
12		BLQ,NR	BLQ,NR	0.4 ± 1.1	18.3 ± 20.0
24		9.0 ± 10.1	10.5 ± 12.1	22.7 ± 27.0	350.7 ± 596.1
48		BLQ,NR	24.4 ± 19.3	47.8 ± 42.8	1351.4 ± 1405.9

N = 10

BLQ = below limit of quantification (2.00 ng/ml for P89 9124, P88 8991, and HP 873; 4.00 ng/ml for P89 9430)

NR = not reportable (sample analyzed but was not evaluable and could not be repeated due to insufficient sample)

<sup>ⓐ</sup> one value = 4.0 ng/ml, all other values were BLQ

<sup>ⓑ</sup> one value = 5.5 ng/ml, all other values were NR or BLQ

Summary of individual study findings:

The study results are summarized in the following sponsor's table.

Report Title: Six-month oral toxicity of HP 873 in rats with a five-week recovery period				Test Article: Iloperidone				
Species/Strain: Sprague-Dawley CD <sup>1</sup> (SD)BR		Duration of Dosing: 26 weeks (6 months)		Study No. 1292-35				
Initial Age: 9 weeks		Duration of Postdose: 5 weeks		Location in CID: Vol. Page				
Date of First Dose: August 5, 1992		Method of Administration: Oral intubation						
		Vehicle/Formulation: 2% aqueous starch		GLP Compliance: Yes				
Special Features: None								
No Observed Adverse Effect Level: Not established				MTD: 12 mg/kg/d				
Daily Dose (mg/kg)	0 (Control)		12		24		48	
No. of Animals	M: 27	F: 27	M: 27	F: 27	M: 27	F: 27	M: 27	F: 27
Toxicokinetics: mean plasma concentrations (ng/mL)	Ilo: BLQ, NR P89 9430: BLQ, NR P89 9124: BLQ, NR P88 8991: BLQ	Ilo: BLQ P89 9430: 0.4 P89 9124: BLQ P88 8991: BLQ	Ilo: 24.4 P89 9430: BLQ, NR P89 9124: 1.1 P88 8991: 0.5	Ilo: 18.3 P89 9430: BLQ, NR P89 9124: BLQ, NR, P88 8991: 0.4	Ilo: 89.5 P89 9430: 9.8 P89 9124: 5.4 P88 8991: 3.5	Ilo: 350.7 P89 9430: 9.0 P89 9124: 5.4 P88 8991: 22.7	Ilo: 958.3 P89 9430: 40.6 P89 9124: 28.2 P88 8991: 41.1	Ilo: 1351.4 P89 9430: BLQ, NR P89 9124: 24.4 P88 8991: 47.8
<b>Noteworthy Findings:</b>								
Died or Sacrificed Moribund	1	0	0	0	1	0	0	1
Body Weight (%)	679 g	410 g	-8.1%*	-9.0%*	-22.5%*	-18.0%*	-32.1%*	-29.5%*
Mean Food Consumption (g/animal)*	29.9	23.0	29.0	21.4	25.9*	21.1*	25.5*	19.8*
Clinical Observations	-	-	Relaxed scrotum Ptosis	Ptosis	Relaxed scrotum Ptosis ↓activity	Ptosis	Relaxed scrotum Ptosis ↓activity Epistaxis	Ptosis ↓activity Epistaxis
Ophthalmoscopy	No drug-related ocular abnormalities detected.							

b(4)

Continued

Report Title: Six-month oral toxicity of HP 873 in rats with a five-week recovery period			Test Article: Iloperidone					
Hematology (↓ or ↑ relative to control at Week 26)	--	--	↓platelets, ↑RBC	↓WBC, ↓lymphocytes, ↑RBC	↓WBC, ↓lymphocytes, ↓platelets, ↑RBC	↓WBC, ↓lymphocytes, ↑RBC	↓WBC, ↓lymphocytes, ↓platelets, ↑RBC	↓WBC, ↓lymphocytes, ↑RBC
Serum Chemistry (↓ or ↑ relative to control at Week 26)	--	--	--	↓glucose ↓triglycerides	--	↓glucose ↓triglycerides	↓glucose ↓triglycerides	↓glucose ↓triglycerides
Urinalysis	--	--	--	--	--	--	--	--
Organ Weights (%) <sup>c</sup> Adrenal Wt (%) <sup>d</sup>	--	--	-8.8 --	-9.3 --	-24.6 --	-20.3 --	-26.5 +28	-31.5 --
Gross Pathology	No noteworthy changes noted.							
Histopathology (relative to control)	--	--	--	--	--	--	--	--
	--	--	--	--	--	--	Liver: IPAS- & ORO-positive material, consistent with food intake	--
	--	--	--	--	Testes: ↓size & degeneration/atrophy	--	Testes: ↓size & degeneration/atrophy	--
	--	--	--	--	--	--	Prostate: minimal to mild active/chronic inflammation	--
	--	--	Mammary gland: Vacuolization of glandular epithelium (both sexes, dose-related increase in incidence and severity, all treated groups); hypertrophy/hyperplasia (females, 24 and 48 mg/kg/day).					
	--	--	ND	ND	Bone marrow sections: Minimal to mild fatty infiltration, especially in highest-dose group.			
Additional Examinations: Neurology and Auditory Response	No drug-related findings.							

#### Recovery Period

Postdose Evaluation (Week 31, 5 Weeks Postdose) No. Evaluated:	6	6	6	6	6	6	6	6
Noteworthy Findings:								
Died or Sacrificed Moribund	0	0	0	0	0	0	0	0
Mean Weight Gain Weeks 27-31 (g)	20	11	3	8	81	30	123	21
Mean Food Consumption (g/animal) <sup>e</sup>	29.1	21.7	26.6	21.3	27.4	20.9	30.0	21.6
Clinical Observations	All abnormal clinical observations ceased after discontinuation of treatment.							
Hematology (Week 31)	--	--	Incomplete recovery of WBC, lymphocyte count, and RBC					
Hematology (↓ or ↑ relative to control at Week 31)	--	--	--	↑RBC	--	↑RBC, ↑Hct	--	↑RBC, ↑Hct, ↑Hb
Serum Chemistry (↓ or ↑ relative to control at Week 31)	--	--	--	↓triglycerides	--	↓triglycerides	--	--
Organ Weights (Week 31)	--	--	Ameliorated, but dose-related difference from control remained (significant for Groups 3 and 4).					
Histopathology (Week 31)	Mammary gland: Vacuolization of glandular epithelium evident but decreased post-treatment. Bone marrow and prostate abnormalities: Greatly ameliorated post-treatment.							

-- No findings of note.

\* Significantly different from control ( $P < 0.05$  by Dunnett's 2-sided multiple comparisons test).

a At the end of the dosing period. For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data (not on the percent differences).

b Mean absolute food consumption (g/animal) at Week 26.

c Percentage difference from control in mean terminal body weight at Week 27 necropsy.

d Percentage difference from control in mean absolute adrenal weight at Week 27 necropsy.

e Mean absolute food consumption (g/animal) at Week 31.

BLQ = below limit of quantification; NR = not reported; RBC = red blood cell count; WBC = white blood cell count; Hb = hemoglobin; Hct = hematocrit.

**Conclusion:** The MTD of iloperidone in this study is 12 mg/kg/d, based on a marked body weight decrease (18-22% vs. control) at the next higher dose tested (24 mg/kg/d). A NOAEL was not reached in this study, as the lowest tested dose (12 mg/kg/d) induced a decrease in body and organ weights, hematological and clinical chemistry changes (mostly in females), and histopathology changes (vacuolization of glandular epithelium in the mammary glands of males and females). At equal oral doses, iloperidone plasma exposure in females was higher than in males.