

**CENTER FOR DRUG EVALUATION AND
RESEARCH**

APPLICATION NUMBER:

18-936/SE5-064

PHARMACOLOGY REVIEW

FLUOXETINE DEVELOPMENTAL TOXICITY: ANIMAL-TO-HUMAN COMPARISONS

A Report

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General Information

- Drug name(s):** Fluoxetine (Adofen, Fluctin, Fluoxeren, LY-110140, Fontex, Foxetin, _____)
- Structure:** (\pm)N-methyl-3-phenyl-3-(4-trifluoromethyl-3-phenylmethylphenoxy)propylamine
- Year approved:** 1988
- Therapeutic uses:** Antidepressant (a selective serotonin reuptake inhibitor)
The most frequently prescribed antidepressant in the U.S.
- Duration of dosing:** Prolonged (often throughout gestation)
- Pregnancy category and warnings:** Pregnancy Category **B**
Most common side effects: nervousness, anxiety, nausea, insomnia, anorexia, diarrhea, headache

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FLUOXETINE DEVELOPMENTAL TOXICITY: I. EVALUATION OF HUMAN DATA

I.1. Materials

Reviewed are 12 publications of studies on the outcomes of gestational exposures to fluoxetine in human subjects. Eight of these publications are population studies of prospective design (incl. 3 controlled epidemiological analytical studies and 5 descriptive uncontrolled case surveys based on information from premarketing clinical trials or postmarketing reports); three papers are publications of single case reports; and one paper is a meta-analysis performed on all available published and unpublished reports (up to November 1996) on congenital malformations observed after fluoxetine use during the 1st trimester of pregnancy.

I.2. Method

The review process was performed in the following consecutive steps:

1. *Selecting papers for review* by means of excluding irrelevant papers and those not containing new information, such as repetitive publications or literature reviews.

2. *Abstracting data* from the selected publications in an uniform way, according to the endpoints that have been recommended to be included in a reproductive/developmental toxicity database (as described by Carole Kimmel in Appendix 1 of the project proposal). The *endpoints* include: type of data, number of subjects, exposure parameters (dose, time, and duration of exposure), other potential risk factors or confounders, effects on offspring (embryo/fetal or perinatal death, prematurity, congenital anomalies, altered birth weight, postnatal complications, developmental delays, neurobehavioral or other organ systems' deviations) and maternal effects.

3. *Evaluation of data* (each study separately) according to the following criteria:

- Reliability of study design
- Appropriate control group
- Sufficient number of subjects
- Adequate assessment of exposure and outcome(s)
- Control of potential confounding factors (e.g. maternal age, gravidity, parity, previous adverse outcomes, pre-existing and/or concurrent disease, socio-economic factors, medication or drug use, smoking, alcohol consumption, occupation, etc.).
- Relevant statistical analysis to assess the relation between exposure and outcome.

Additional criteria, such as: plausibility of results (having in mind the known pharmacokinetic and pharmacodynamic properties of the agent), and concordance with other studies, were introduced in the evaluation of single case reports where some of the above listed criteria could not be applied.

4. *Conclusion about data reliability* for each study separately (based on steps 2 and 3).

5. Overall assessment of the observed adverse effects and the likelihood of their causal relation to fluoxetine exposure during gestation, based on Hill's criteria for establishing causation as follows:

- Strength of evidence
- Consistency of evidence
- Specificity of effect
- Temporality of effect
- Dose-response relationship
- Plausibility of effect
- Coherence with existing knowledge
- Analogy (structure activity)

(Hill A.B. The environment and disease: association or causation?
Proc.R.Soc.Med. 1965, 58:295-300)

On the basis of this assessment, a selection of outcomes associated with fluoxetine gestational exposure in humans was made, in order to be compared with experimental animal data.

1.3. Results

1.3.1. Evaluation of studies

The reviews of human studies, incl. abstracted data, their evaluation, and conclusion about data reliability, are presented in Tables 1 and 1-a (for population studies and single case reports respectively). A short summary of the above information is presented in Table 2.

The *study design* is evaluated as reliable only for the 3 epidemiological analytical studies on fluoxetine gestational exposure in association with signs of developmental toxicity (Pastuszak et al, 1993, Chambers et al, 1996, Nulman et al, 1997). These studies are of a prospective cohort type (the best type of design to study a cause-effect relationship), with adequate control groups. In contrast, the reliability of study design to assess a relationship between fluoxetine gestational exposure and adverse pregnancy/birth outcome is poor for single case reports, and limited for the descriptive case surveys since they lack control groups and are prone to bias because they are based on voluntary spontaneous case reports instead on a random sample.

The *sample size* varies from 17 to 544 in the descriptive case surveys, and from 219 to 482 (incl. control groups) in the epidemiological studies. For most of the descriptive surveys the sample size is insufficient to provide reliable estimates of the rates of adverse outcomes. Although the overall sample size is more adequate in the analytical epidemiological studies, it is insufficient in some stratification subgroups (e.g. the subgroups of patients treated during entire gestation or 1st trimester only, Nulman et al, 1997), or of insufficient power to detect a small or moderate increase in the risk of adverse outcomes (e.g. Pastuszak et al, 1993).

The *doses* of maternal exposure to fluoxetine are not reported in 5 of the reviewed publications (4 surveys and 1 epidemiological study); presumably the doses were within the therapeutic dose range (10 to 80 mg/day), since all these studies involved subjects that had been treated with fluoxetine for therapeutic reasons. Where indicated, the mean daily doses of fluoxetine were 25 or 28 mg, and the range - from 10 to 80 mg/day.

The *timing and duration* of exposure to fluoxetine have taken place during the 1st trimester of pregnancy in the majority of studies reviewed (8 of 12). In 2 of these studies (the analytical epidemiological studies of Chambers et al, 1996 and Nulman et al, 1997) there are subgroups that were exposed during entire gestation as well, but they were analyzed separately and did not confound the data pertinent to the 1st trimester exposure. One study is based on exposures during 3rd trimester to delivery (the survey of Goldstein et al, 1995), and in two publications the timing and duration of exposure are not reported.

An appropriate *statistical analysis* including controlling for other potential risk factors that could confound the association between exposure and outcome is essential in assessing a cause/effect relationship and in evaluating the reliability of a study. The association between exposure and outcome taking into account potential confounders has been analyzed only in the 3 above mentioned epidemiological analytical studies (Pastuszak et al, 1993; Chambers et al, 1996, and Nulman et al, 1997). The rest of the studies (case surveys and case reports) are descriptive, and therefore unsuited for analyzing relations between exposure and outcome. Although in some of these studies (e.g. Goldstein et al, 1995; McElhatton et al, 1996) confounding factors were registered, their role was not taken into account because of the limitations inherent to the descriptive study design.

In summary (table 2), out of the 12 studies reviewed, the reliability of data has been evaluated as **'good'** in 6 studies, including: the 3 epidemiological analytical studies of prospective cohort design which had appropriate control groups and took into account possible confounding factors; 2 case reports with reliable assessments of exposure and outcome, findings consistent with other studies, and plausible results with regard to the pharmacokinetics and pharmacodynamics of the agent; and 1 meta-analysis by Addis and Koren, 1997 (not shown in the table). Four studies are of **limited** reliability (all of them are uncontrolled descriptive case surveys based on spontaneous reports), and the remaining two (a case report by Venditelli et al, 1995, and a case survey by Brunel et al, 1994) are of **poor** reliability due mainly to confounding by multidrug exposures and very small number of subjects, respectively.

1.3.2. Adverse outcomes in relation to gestational exposure to fluoxetine in humans

Table 2 summarizes the data on pregnancy and birth outcomes according to literature source. In table 3, an attempt is made to evaluate the likelihood of a causal relation between the reported adverse outcomes and gestational exposure to fluoxetine, taking into account the compliance with Hill's criteria for establishing causation.

As seen in tables 2 and 3, the reported changes in pregnancy and birth outcomes in women treated with therapeutic doses of fluoxetine during gestation are subtle. They are listed below in order roughly corresponding to their occurrence in association with fluoxetine maternal exposure.

The most consistent findings are the **early postnatal complications** after fluoxetine prenatal exposures, and particularly exposures that have taken place throughout entire gestation until birth. Evidence for postnatal complications is present in 6 out of 7 available publications that have studied this endpoint. Out of these, 2 are controlled

epidemiological prospective cohort studies, 2 are descriptive surveys, and 2 - single case reports. The reported rates of postnatal complications in these studies vary between 3 and 13%; rates closer to the higher limit of this range, or rates significantly higher than the control ones are found after exposures during the last trimester or the entire gestation (Goldstein et al, 1995; Chambers et al, 1996), in comparison to 1st trimester-only exposures (*temporality of effect*). Clinically, the postnatal complications are expressed by *consistent* symptoms: jitteriness, irritability, hypertonia, tremor, impending seizures, respiratory problems, as well as subcutaneous or internal hematomas (petechiae, cephalhematoma, or periventricular bleeding). These symptoms are in agreement with the known side effects of fluoxetine in adults, as well as with the known pharmacokinetic and pharmacodynamic properties of fluoxetine, including its effect on the platelets. The disappearance of these symptoms in the neonate is *concordant* with the decrease of fluoxetine and its main metabolite in cord blood. These data testify to a *coherence with the existing knowledge, plausibility, and specificity of effect*. Therefore, although the strength of evidence from a statistical point of view is limited (due mainly to insufficient controlled epidemiological studies on this endpoint), a causative relation to fluoxetine exposure is strongly suggested.

Data on ***spontaneous abortions*** after gestational exposures to fluoxetine are available in 6 studies, 3 of which are analytical epidemiological studies and 3 - descriptive surveys. In 5 of these 6 studies, the reported spontaneous abortions' rates are quite consistent (within the range of 13 to 15.9 per cent of all pregnancies exposed to therapeutic doses of fluoxetine during the 1st trimester). These rates were found to be higher in comparison to control in 2 out of the 3 analytical epidemiological studies (Pastuszak et al, 1993; Nulman et al, 1997), but this difference did not reach statistical significance due mainly to the insufficient number of subjects. Similar rates are reported in most of the descriptive surveys (Goldstein and Marvel, 1993; Shick-Boschetto et al, 1992; McElhatton et al, 1996), but the absence of control groups in these descriptive studies does not allow to assess the risk.

Having in mind the methodological uncertainties inherent in determining the true rate of spontaneous abortions (Wilcox, 1991), the available data provide inconclusive evidence for either supporting or refuting an association of fluoxetine gestational exposure with spontaneous abortions.

Prematurity rates after fluoxetine use in pregnancy are reported in 7 of the reviewed population studies (3 epidemiological and 4 surveys). Out of these, a statistically significant increase in prematurity rate is found in only one study, in association with continued fluoxetine use after 25 week gestation (Chambers et al, 1996). This is a controlled prospective cohort epidemiological study of good reliability. However, this finding is not confirmed by any other study. This lack of consistency could be due to the fact that in the majority of the other studies the timing of exposure was different: it took place earlier (during the 1st trimester) as compared to 3rd trimester and later in Chambers' study. In support, in that same study, maternal exposures prior to 25 weeks of gestation did not result in increased prematurity rates. It is also possible that Chambers' findings could have been confounded by parallel exposures to other psychoactive drugs that took place in 30 per cent of the patients.

The evidence is insufficient to draw a definite conclusion.

Data on **birth weight** are available in 4 population studies (including 3 epidemiological and 1 case survey) and in 3 case reports. These studies provide *consistent* evidence for a lack of effect on birthweight after fluoxetine maternal exposure during the *first trimester* of pregnancy.

Altered birthweight (a statistically significant *decrease*) is found in association with 'late' exposures to fluoxetine *continued after 25 weeks of gestation* in an epidemiological study of good reliability (Chambers et al, 1996). This finding is *plausible and coherent* with the existing knowledge about the anorexic (apetite suppressing) effect of fluoxetine in therapeutic doses in adults. Thus, the decrease in birthweight could be secondary to a diminished maternal food consumption. Unfortunately, the study does not provide information on the maternal effects. A causative relation is possible, but the conclusion is hindered by the lack of other studies on birthweight after 'late' gestational exposures to fluoxetine.

Congenital malformations. There is a consistent evidence in all studies for a *lack* of any increase in major congenital malformations, or for a pattern of malformations associated with exposures to fluoxetine during gestation. Two epidemiological studies (Pastuszak et al, 1993 and Chambers et al, 1996) report an increase in minor congenital malformations in comparison to control. The difference reaches statistical significance in only one study (Chambers et al, 1996), however no description of the 'minor' malformations found is provided. The prevailing evidence is in favor of a lack of relation of fluoxetine gestational exposure to birth defects. This conclusion is supported by the meta-analysis of Addis and Koren (1997) which takes into account all available information on fluoxetine 1st trimester exposures up to 1996.

Perinatal death. There is consistent evidence (6 out of 6 population-level studies) that maternal exposure to therapeutic doses of fluoxetine is not associated with increase in perinatal lethality.

Neurobehavioral development of children prenatally exposed to fluoxetine has been followed up in only one study (Mattison et al, 1999), a comprehensive neuropsychological evaluation of 66 children at 4-6 years of age, born to prospectively identified women who were taking fluoxetine during pregnancy, in comparison to a prospectively identified control group of 30 children of mothers with pregnancy exposures to drugs "not deemed to be teratogenic". Verbal learning and memory, language, short-term memory/attention, motor, parent-rated behavior, and IQ scores were analyzed and compared using appropriate statistical tests. No statistically significant group differences were found, suggesting that the risk of negative neurobehavioral outcome in fluoxetine-exposed children is similar to that of non-exposed ones. The lack of supportive evidence from other neurobehavioral studies does not allow a definitive conclusion about this endpoint.

II. EVALUATION OF ANIMAL DATA

II.1. Materials

Reviewed are all available on file animal reproductive and developmental toxicity studies on Fluoxetine safety assessment, as follows: two reproductive/fertility studies in the rat (also involving teratology and postnatal segments) and four prenatal developmental toxicity studies (including two preliminary and two routine teratology studies in two animal species - rat and rabbit). The reviewed studies are listed below:

- 1• A Fertility Study on Fluoxetine Hydrochloride (LY110140) in the Female Rat (1980). Lilly Research Laboratories Study No RO 7179 by J. Wold , N. Owen and E. Adams
- 2• A Fertility Study, Including Behavioral and Reproductive Assessment of the F₁ Generation, in the Wistar Rat Given Fluoxetine Hydrochloride (LY110140) in the Diet (1982). Lilly Research Laboratories Study No R10280 & RO4781 by G.Brophy, N. Owen and J. Hoyt
- 3• A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat (1979). Lilly Research Laboratories Study No R-77, IND — Toxicology Report No 7 by J.S Wold and J.K. Markham
- 4• A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat (1979). Lilly Research Laboratories Study No R-207, IND — . Toxicology Report No. 8 by J. S. Wold and J. K. Markham
- 5• A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit (1979). Lilly Research Laboratories Study B -7017, IND — Toxicology Report No. 9 by J. S. Wold and J. K. Markham
- 6• A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit (1979). Lilly Research Laboratories Study B-7087, IND — Toxicology Report No. 10 by J. S. Wold and J. K. Markham.

II.2. Method

Prior to comparing animal to human data, the experimental studies carried out to assess safety of the agent have been evaluated.

The evaluation procedure encompassed the following consecutive *steps*:

1. Data collection and review: Abstracting, summarizing, and evaluating reliability of data from each individual study in order to identify and assess "signals" of reproductive/developmental toxicity;
2. Comparison of outcomes across studies according to category (reproductive, developmental, prenatal/postnatal) and subclass of toxicity (fertility, embryo/fetal loss, dysmorphogenesis, alterations to growth, viability and functional toxicities);
3. Evaluation of validity and reliability of identified effects (outcomes) for comparison with those in humans.

Step 1

The data were abstracted in an uniform way, according to a common format in order to facilitate data assessment and comparison across studies. *The format* (Annex 1) was prepared on the basis of the endpoints outlined in the Project Proposal (Kimmel et al,

1997) and in accordance with the format of the National Toxicology Program's Special Reproductive Study (Chapin and Sloane, 1997).

The format consists of the following parts:

- Data entries: particulars of animal model, exposure (compound, dose, route and mode of administration, timing and duration of treatment) and outcomes (general and reproductive toxicity endpoints, subdivided into fertility, prenatal, and postnatal components);
- Data summary: highlights the most sensitive endpoints, LOAEL and NOAEL for general, maternal, reproductive and developmental toxicity;
- Study conclusions;
- Confounding and interfering factors that might have compromised the validity of study conclusions;
- Evaluation of reliability of each individual study with regard to extrapolating the data to humans.

The *evaluation* of studies with regard to their reliability for extrapolating the data to the human was performed according to the following criteria:

- Adequacy of experimental model
- Adequacy of dose and route of administration
- Adequacy of timing and duration of exposure
- Sufficient number of animals per group
- Presence of a dose/effect relationship
- Appropriate statistical analysis
- Confounders
- Concordance of findings with pharmacokinetic and pharmacodynamic properties of the agent
- Consistency of findings with other experimental studies

Step 2 involved comparison and assessment of *effects across studies according to category of outcome*. For this purpose, condensed comparative summaries and a parallel layout of reviewed data by category of outcome (e.g. Fertility, Prenatal, Postnatal) and subclass of toxicity, were prepared on the basis of the information collected in step 1.

Step 3 involved *evaluation of outcomes* (presence or absence of effect on each of the reviewed endpoints) and their respective NOAELs in order to estimate the strength of evidence for each "signal of toxicity" (or no toxicity) and its relevance to human situation. Each outcome was evaluated by criteria similar to those applied above for evaluation of individual studies. The criteria include:

- magnitude of effect (incidence relative to control)
- consistency of effect across studies
- consistency of effect across species
- statistical significance of effect
- dose-dependence of effect
- influence of confounding and interfering factors
- plausibility of effect with regard to pharmacokinetic and pharmacodynamic properties of compound.

II.3. Results

Extended summaries of fluoxetine *individual experimental studies* abstracted according to the format, along with conclusion, comments, confounders, and evaluation of each study are presented in Tables 4.1. to 4.6.

Condensed comparative summaries and evaluation of fluoxetine *effects by category* (fertility, prenatal and postnatal development) are presented in tables 5.1, 5.2, and 5.3 respectively.

The *endpoints (outcomes)* of fluoxetine reproductive and developmental toxicity and the respective NOAEL levels for each outcome are summarized and evaluated in Table 6.

All reviewed studies were performed *in vivo*, in animal models adequate for assessment of the predictive value of animal testing for human developmental toxicity.

Of the total of 6 studies, 4 were performed in the rat (of Wistar and Fischer 344 strain), and 2 in the rabbit (Dutch Belted).

The doses and route of administration are adequate to human exposures (oral, dose range from 1.3 to 15 mg/kg/day, i.e. from a level equal to the upper limit of the human therapeutic dose (approximately 1 mg/kg/day) to 15 times higher. All of the studies employed multiple dosing regimens which allowed assessment of dose-effect and dose-response relationships. The mode of treatment was predominantly by gavage, with the exception of one study (A Fertility Study, Including Behavioral and Reproductive Assessment of the F₁ Generation, in the Wistar Rat Given Fluoxetine Hydrochloride in the Diet. Lilly Research Laboratories Study No R10280 & RO4781 by Brophy, Owen and Hoyt, 1982), which employed dosing through diet that might have confounded the estimate of actual dose.

The timing of exposure in 4 of the 6 studies covers the period from implantation to the end of organogenesis; in the remaining 2 (fertility) studies, along with gestational exposures, parental exposures (maternal or maternal+paternal) prior to gestation as well as postnatal exposures during lactation were applied. It should be noted that exposure of both parents is unlikely in human situation. The timing of exposure is relevant to that in the human studies.

The number of animals/litters tested per dose group is sufficient, with the exception of the two preliminary, dose-finding studies in rat and rabbit (see Materials, studies No 3 and 5). In the studies involving postnatal assessments (Materials, studies No 1 and 2), the litters were culled to a specified number only in study 2. This, along with the differences in exposure (maternal versus both parents) and mode of treatment interferes with comparability of results between these two studies.

Details on the confounding and interfering factors by study and outcome are given in tables 4.1 to 4.6, and in table 6 respectively.

II.3.1. Effect on fertility

Fluoxetine effect on fertility (Tables 4.1, 4.2, and 5.1) was assessed in two studies (an one- and a two-generation study), both performed in the rat (Wistar). Both these studies employed similar dose ranges and routes of exposure, but differed by the mode of treatment (by gavage vs through diet) and by the type of parental exposure (maternal only vs both parents). Maternal exposures took place 2 or 3 weeks before mating and throughout breeding, gestation and lactation; paternal exposure started at adolescence and continued for 10 weeks prior to mating. Both studies resulted in similar conclusions: fluoxetine induces no significant effect on fertility even at doses that produce significant general effects (NOAEL for fertility 7.4 to 12.5 mg/kg/day, as compared to NOAEL of 3.1 to 5 mg/kg/day for general effects). The conclusions are reliable, although exposure quantitation may not have been precise in one of these studies due to dosing through diet. The general effects (decreased food consumption and body weight) are not necessarily a sign of toxicity, as they are characteristic of the pharmacological action of this drug. Statistically non-significant, but dose-dependent, signs of effect on fertility (decreases in fertility index, in the number of corpora lutea, in litter size, and increase in pre-implantation embryo lethality) are found at NOAEL doses of 7.4 to 12.5 mg/kg/day.

II.3.2. Prenatal developmental effects

The prenatal effects of fluoxetine exposure *in utero* are assessed in all 6 studies (Tables 4.1-4.6, Table 5.2, and Table 6). Prenatal developmental effects are observed at dose levels that induce maternal effects (weight loss and decreased food consumption), and involve mostly an *increased incidence of resorbed or aborted conceptuses* (postimplantation losses) at doses of 12.5 to 15 mg/kg per day applied during organogenesis in two animal species (rat and rabbit). Although not consistent in the rat, this effect is consistent and better expressed in the rabbit. The validity of this adverse outcome is supported by the *decrease in litter size*, which, although not statistically significant, is dose-dependent and consistent across all reviewed studies in the two species. *Fetal weight* is usually unchanged (except for one study in the rabbit, where a statistically non-significant, but dose-dependent reduction by about 10% was found at 15 mg/kg per day). It should however be noted that the effect of fetal weight is underestimated due to the smaller litter sizes at higher exposures in all studies (Table 6). Increase in *congenital malformations* rates is not found in either species, even at doses that cause maternal mortality (15 mg/kg/day in the rabbit and 40 mg/kg/day in the rat). An elevation in the incidence of skeletal *variations* (rudimentary and wavy ribs) was found in one study (rabbit) at all exposure doses (2.5 to 15 mg/kg per day during organogenesis), but the effect was not dose-dependent. The NOAEL for prenatal developmental toxicity is 5 to 7.5 mg/kg per day in rat and rabbit respectively.

II.3.3. Postnatal developmental effects

The postnatal effects of fluoxetine exposure are assessed in 2 studies performed in one animal species (rat, Wistar) (Tables 4.1, 4.2). Both studies involved oral parental exposures prior to pregnancy, as well as throughout entire gestation and lactation. However, the studies differed by type of exposure (maternal only vs maternal

+paternal) and mode of treatment (by gavage vs through diet). Adverse postnatal effects are found in both of these studies but are more manifested in the one that involved treatment of both parents through diet. (Tables 5.3. and 6). These effects involve *elevated perinatal mortality (increased incidence of stillbirths and decreased postnatal survival during 1st week of life), decreased birthweight and postnatal weight depression* detectable until maturity. With the exception of stillbirths, these findings are reported consistently in both studies, although the changes are statistically significant in the above mentioned study only. The NOAEL for postnatal manifestations of developmental toxicity in the rat is from 3.1 to 5 mg/ kg per day, which suggests that adverse postnatal effects are induced by lower exposures in comparison to those inducing prenatal manifestations of developmental toxicity. It should be noted however, that NOAEL at 3.1 mg/kg is very likely to be an underestimate of the actual dose, and the apparent "selective" effect of fluoxetine on postnatal development at doses seemingly lower than those affecting prenatal endpoints may actually be due to exposure misquantitation because of the dosing through diet and the two-fold increase of maternal food consumption during lactation, as reported in that particular study.

Behavioral testing of the progeny is performed in one of the two postnatal studies. Tested were some sensory and motor coordination functions (auditory startle reflex, visual placing response, rotating rod performance, and poke-hole test). No motor and sensory-motor behavioral deviations were found, but the testing was performed close to maturity (at the age of 2 to 3 months) so that earlier behavioral deviations might have been omitted. No detectable effect on *reproductive function* of the progeny was found

II.3.4. Maternal effects

The effect of fluoxetine on the maternal organism is determined in 5 studies (tables 5.2. and 6). The most common effect, found in all studies, is the *decrease in food consumption* at dose levels of 5 and more mg/kg per day in the rat, and at lower doses (down to 2.5 mg/kg/day) in the rabbit. It is accompanied by a *maternal weight* loss and reduced gestational weight gain of about 10% during treatment. As the anorexic (appetite-suppressing) effect and the resulting weight loss are well known pharmacodynamic features of fluoxetine in both human and animal species, the maternal weight loss can not be interpreted as a sign of maternal toxicity, unless it is accompanied by other clinical signs of toxicity. Such signs are not reported, except at much higher doses (15 mg/kg/day in the rabbit and 20 mg/kg/day in the rat) which cause excessive (up to 90%) depression in food consumption and substantial weight loss accompanied by elevated maternal mortality.

The NOEL level for maternal effects in the rat vary from 3 to 5 mg/kg/day in the different studies, and in the rabbit it is below 2.5 mg/kg/day. This shows that effects in the progeny occur at dose levels that cause maternal (although not necessarily adverse) effects.

Summary

It is evident that prenatal fluoxetine exposure induces pre- or postnatal developmental toxicity only at levels that affect the maternal organism, which means that *fluoxetine is not a selective embryo-fetal toxicant*.

The manifestations of fluoxetine reproductive and developmental toxicity in animal models, along with their respective no-effect levels are summarized in Table 6. The endpoints within each of the effect categories are evaluated according to their rate of occurrence, consistency across studies and species, statistical significance, dose dependence, interference of confounding factors, plausibility and coherence with existing information. The most “reliable” endpoints are highlighted.

The outcomes for comparison with human studies are briefly outlined below:

Toxicity	“Positive” (Presence of effect)	Probable (Effect probable)	“Negative” (No effect)
Maternal	Decreased food consumption		Gestational length
	Decreased body weight and weight gain		Mortality
Reproductive			Fertility
Developmental			
- Prenatal	Decreased litter size	Embryo-fetal loss	Congenit. Malformations
		Skeletal Variations	
- Postnatal	Decreased survival (1 st week of life)	Stillbirths Decreased liveborn litter size	Neurobehavioral function at adolescence*
	Decreased birthweight		Reproductive function of progeny*
	Decreased weight gain		

* One study only

III. FLUOXETINE: ANIMAL-to- HUMAN COMPARISONS

III.1. Pharmacokinetics and pharmacodynamics

Although pharmacokinetic and pharmacodynamic studies on Fluoxetine have not been conducted in the context of the reproductive and developmental toxicity studies which are subject of the present review, they are discussed here as an essential pre-requisite to comparison of animal to human reproductive and developmental effects.

A comparison between human and animal studies with respect to fluoxetine pharmacokinetics and pharmacodynamics is given in Table 7. Since fluoxetine pharmacokinetics during pregnancy and embryo/fetal development have not been studied in humans, the listed data are derived from animal and human studies on adult non-pregnant subjects.

In general, the pharmacodynamics and kinetics of fluoxetine are quite similar in laboratory animals and humans with respect to: mechanism and sites of action, adverse effects at overdoses, toxicometric parameters of acute toxicity, absorption, tissue distribution and binding, metabolic system and active metabolite (Table 7). A specific pharmacodynamic feature of fluoxetine is its appetite-suppressing effect leading to weight loss in both animals and man, which is a pharmacodynamic rather than toxic effect. This should be taken into account in interpreting the signs of fluoxetine toxicity. *The pharmacodynamic and kinetic similarities between animals and man are a prerequisite for comparability of effects and support the adequacy of animal models as predictors of adverse effects of fluoxetine in humans.*

However, there are certain dissimilarities between lab. animals and man that should be taken into account in assessing predictability of animal studies to human situations:

- The elimination and clearance of fluoxetine and its active metabolite (nor-fluoxetine) is much slower in the human in comparison to rat;
- The inter-individual variations in the elimination, clearance, and steady-state plasma concentration of fluoxetine are much greater in the human, possibly due to the genetic polymorphism of F- metabolizing liver enzymes;
- There is a lack of a dose-effect relationship for the pharmacological therapeutic effects of fluoxetine in the human;
- There are pharmacodynamic differences between laboratory animals and man with respect to some neuroendocrine side-effects of fluoxetine (e.g. increased hypothalamic secretion of corticotrophic-releasing factor and vasopressin in the rat, leading to increased ACTH and vasopressin plasma levels, while no such effect has been found in the human).

These differences suggest that direct quantitative comparisons of concrete toxicometric parameters (dose levels), such as LOAEL and NOAEL, between experimental animals and humans would not be a reliable tool for assessing animal-human similarities in fluoxetine toxicity.

III.2. Reproductive and Developmental Toxicity

Due to the lack of human studies on fluoxetine effect on fertility, this comparison covers the manifestations of developmental toxicity.

A comparison between animal and human data on developmental effects of fluoxetine prenatal exposures is shown in Table 8. The comparison shows a considerable similarity between experimental and human data as follows:

- Prenatal exposure to fluoxetine induces developmental effects in both experimental animals and humans.
- Developmental effects are induced only by dose levels that affect the maternal organism. This means that fluoxetine is not a selective embryofetal toxicant in neither experimental animals or humans, e.g. it influences prenatal development through affecting the maternal organism.
- The manifestations of fluoxetine maternal and prenatal effects in experimental animals and humans are similar. Although similarities apparently exist between the

postnatal effects as well, the number of animal studies conducted to assess the postnatal developmental effects is insufficient for a meaningful comparison.

- The LOEL and NOEL for induction of maternal and developmental effects are severalfold higher in experimental animal in comparison to human studies. This is probably due to pharmacokinetic differences in the elimination and clearance of fluoxetine and its active metabolite that are much slower in the human in comparison to rat.

References

Animal studies on Fluoxetine safety assessment (FDA files):

- A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat (1979). Lilly Research Laboratories Study No R-77, IND Toxicology Report No 7 by J.S Wold and J.K. Markham
- A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat (1979). Lilly Research Laboratories Study No R-207, IND Toxicology Report No. 8 by J. S. Wold and J. K. Markham
- A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit (1979). Lilly Research Laboratories Study B-7017, IND Toxicology Report No. 9 by J. S. Wold and J. K. Markham
- A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit (1979). Lilly Research Laboratories Study B-7087, IND Toxicology Report No. 10 by J. S. Wold and J. K. Markham.
- A Fertility Study on Fluoxetine Hydrochloride (LY110140) in the Female Rat (1980). Lilly Research Laboratories Study No RO 7179 by J. Wold , N. Owen and E. Adams
- A Fertility Study, Including Behavioral and Reproductive Assessment of the F₁ Generation, in the Wistar Rat Given Fluoxetine Hydrochloride (LY110140) in the Diet (1982). Lilly Research Laboratories Study No R10280 & RO4781 by G. Brophy, N. Owen and J. Hoyt

Published references

- Addis A, Koren G (1997). Safety of fluoxetine during the first trimester of pregnancy: meta-analytical review of epidemiological data. *Teratology*, 55: 37-38.
- Benfield P, Heel R.C., Lewis S.P. (1986) Fluoxetine. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic efficacy in depressive illness. *Drugs*, 32:481-508
- Brunel P, Vial T, Roche I, Bertolotti E, Evreux J-C (1994). Suivi de 151 grossesses exposees a un traitement antidepressif (IMAO exclus) au cours d'organogenese. *Therapie*, 49: 117-122.
- Chambers CD, Johnson KA, Dick LM, Felix RJ and Jones KL (1996). Birth outcomes in pregnant women taking fluoxetine. *N.Engl.J.Med.*, 335, 1010-1015 (as cited by Johnson GL, 1997)
- Chapin, RE and Sloane, RA (1997). Reproductive Assessment by Continuous Breeding: Evolving Study Design and Summaries of Eighty-Eight Studies. *Environm. Health Perspect.* , 105, Suppl. 1, 199-395
- Del Rio J., Montero D., De Ceballos M.L. (1988) Long-lasting changes after perinatal exposure to antidepressants. *Proc. Brain Res.*, 73: 173-187
- Goldstein DJ and Marvel DE (1993). Psychotropic medications during pregnancy (Letter) *JAMA*, 270:2177
- Goldstein DJ (1995). Effects of third trimester fluoxetine exposure on the newborn. *J.Clin.Psychopharmacol.* 15: 417-420.

- Gram L.I. (1994) Fluoxetine. *N.Engl. J. Med.*, 331:1354-1361
- Isenberg K.E. (1990) Excretion of fluoxetine in human breast milk. *J. Clin. Psychiatry*, 51:169
- Johnson GL (1997). Birth outcomes in pregnant women taking fluoxetine. *J.Fam.Pract*, 44: 32.
- Koren G, Pastuszak A, Jacobson S, Schick B, Donnenfeld A, et al (1994). The safety of antidepressants in pregnancy. *Maternal-Fetal Toxicology: A Clinician's Guide*, Vol 2. G. Koren (ed), Marcel Dekker, NY, 1994, pp 59-76
- Mattison S.N., Eastvold A.D., Jones K. L., Harris J.A. , Chambers C.D. (1999). Neurobehavioral follow-up of children prenatally exposed to fluoxetine. *Teratology*, 59: 6, 376 (Abstr.)
- McElhatton PR, Garbis HM, Elefant E, Vial T, Bellemin B. et al (1996). The outcome of pregnancy in 689 women exposed to therapeutic doses of antidepressants. A collaborative study of the European Network of Teratology Information Services ENTIS). *Reprod. Toxicol.*, 10, 285-294.
- Mhanna MJ, Bennet JB, Izatt SD (1997). Potential fluoxetine chloride (Prozac) toxicity in a newborn (letter). *Pediatrics*, 100: 158-159
- Nulman I, Rovet J, Stewart DE, Wolpin J, Gardner HA, Theis JGW et al. (1997) Neurodevelopment of children exposed in utero to neurodepressant drugs. *N.Engl.J.Med*, 336: 258-262
- Pastuszak A, Schick-Boschetto B, Zuber C, Feldkamp M, Pinelli M, et al. (1993). Pregnancy outcome following first trimester exposure to fluoxetine (prozac). *JAMA*, 269: 2246-48
- Patton J.H., Rayer C.H., Langdoc J.L. (1995) In Utero exposure to paroxetine leads to altered physical and motor development. *Abstr. Soc. Neurosci.*, 21: 2017
- Pohland R.C., Byrd T.K., Hamilton M., Koons J. (1989) Placental transfer and fetal distribution of fluoxetine in the rat. *Toxicol. Appl. Pharmacol.*, 98: 198-205
- Schick-Boschetto B, and Zuber C (1992). Fluoxetine exposure in early human pregnancy. *Teratology*, 45: 460.
- Sommi R.W., Crismon M.L., Bowden C.L. (1987) Fluoxetine: A serotonin-specific, second-generation antidepressant. *Pharmacotherapy*, 7: 1-15
- Spencer MJ (1993). Fluoxetine hydrochloride (Prozac) toxicity in a neonate. *Pediatrics*, 92, 721-722.
- Venditelli F, Alain J, Nouaille Y, Brosset A, Tabaste JL (1995). A case of lipomeningocele reported with fluoxetine (and alprazolam, vitamins B1 and B6, heptaminol) prescribed during pregnancy. *Eur.J.Obstet.Gyneco.Reprod.Biol.*, 58: 85-86.
- Vorhees C.V., Akuff-Smith K.D., Schilling M.A., Fisher E., Buelke-Sam J. (1992) Evaluation of the behavioral teratogenic potential of fluoxetine in rats. *Teratology*, 45: 526-527
- Vorhees C.V., Acuff-Smith K.D., Schilling M.A., Fisher J.E., Moran M. S., Buelke-Sam J. (1994). A developmental neurotoxicity evaluation of the effects of prenatal exposure to fluoxetine in rats. *Fund. Appl. Toxicol.*, 23: 194-205
- Wilcox A.J. (1991). Early pregnancy. In: *Reproductive and Perinatal Epidemiology*, M. Kiely (Ed), CRC Press, 63-68.
- Zimmerman E.F., Lauder J.M. (1987) Sites of serotonin uptake in the epithelium of the developing mouse palate, oral cavity and face: possible role in morphogenesis? *Teratology*, 35: 39A

FORMAT

SUMMARY OF ANIMAL REPRODUCTIVE (fertility) STUDIES									
CHEMICAL:									
REFERENCE:									
Species, strain									
Exposed (males, females, both)									
Exposure doses, timing & duration									
- Males									
- Females									
Route of administration									
N generations studied									
Dose – Response (yes/no)									
Effects: (relative to control values)									
F₀ Generation		M	F	M	F	M	F	M	F
Dose Levels →		Control							
Doses males/females →		0	0						
N animals per group:									
General Toxicity									
NOAEL and LOAEL (fill in appropriate cell)									
Body weight (% change vs control) (indicate period)									
Weight gain (%change vs control) (indicate period)									
Organ weight* -Liver -Kidney									
Food consumption (period)									
Clinical signs									
Effects listed above attributable/non-attributable to pharmacological effect of the compound? (yes/no)									
Mortality									
Fertility Parameters[†]									
NOAEL and LOAEL (fill in appropriate cell)									
Fertility index (% pregnant of total mated)									
Absolute testis,*epididymis weight									
Sex accessory gland weight* (prostate seminal vesicle)									
Sperm count									
Sperm morphology									
Sperm motility									
Estrous cycle length									
Other female reproductive organ data									
Hormonal measures									

Key: (–) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change, ↑ increase; ↓ decrease; M male; F female; * adjusted for body weight; -, - no change in males or females

[†] In determining NOAEL and LOAEL for fertility, also take into account: pre-implantation lethality, post-implantation lethality and fetal viability (see next page : F₁ Generation-Prenatal Component)

SUMMARY OF ANIMAL REPRODUCTIVE STUDIES (continued)				
<i>F₁</i> Generation – Prenatal Component ¹	Reference:			
Species, strain				
Prenat. exposure– compound & doses				
Prenat. exposure– timing & duration				
Route of administration				
Gestation day of sacrifice				
Dose-response (yes/no)				
Dose Levels→	0 (Control)			
N dams/litters per group				
NOAEL and LOAEL prenatal development (fill in appropriate cell)				
Pre-implantation lethality ² per dam				
N implants/ N Corpora lutea per dam				
Post-implantation lethality ³ per litter (N dead+resorbed(aborteds) / N implants)				
- Dead (mean <i>n</i> per litter)				
- Resorbed (aborteds), mean <i>n</i> per litter				
Early:Late resorptions ratio				
Females affected of total, %				
N litters completely resorbed/ N total				
Litter size (N live fetuses per litter)				
Sex ratio (proportion of males, %)				
Fetal weight per litter % change vs control				
Sex-differentiated fetal weight -males (% change vs control) -females (% change vs control)				
Incidence of malformations per litter (if elevated, describe malformations below)				
Malformations by type [†] (rate& descrp)				
- External [†]				
- Visceral [†]				
- Skeletal [†]				
Deviations/variations per litter				
- Visceral [†]				
- Skeletal [†]				
Maternal toxicity during gestation				
NOAEL & LOAEL maternal toxicity (fill in appropriate cell)				
Body weight, g (%vs control): - prior to dosing - during dosing - post dosing				
Weight gain during gestation, g (% change vs control)				
Pregnancy-adjusted weight (yes/no)				
Food consumption (indicate period)				
Clinical signs				
Necropsy findings				
Endpoints above attributable / non-attributable to pharmacological effect of the compound? (yes/no)				
Maternal mortality				

¹ Effect to be presented as relative to control values; ² Pre-implantation lethality (%) = [(n C.L.–n impl.) / n C.L.]x100;

³ Post-implantation lethality (%) = (n Dead+Resorbed (aborteds)/n implants)x100; ⁴ N live / N total fetuses per litter;

[†] Describe specific abnormalities which are increased over their control rates

Key: (–) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease;

SUMMARY OF ANIMAL REPRODUCTIVE STUDIES (continued)				
F₁ Generation – Postnatal Component¹	<i>Reference:</i>			
F ₁ Postnatal exposure: (check)	<input type="checkbox"/> Maternal dosing continued through lactation <input type="checkbox"/> Other modes of postnatal exposure (if yes, describe mode, timing & duration) <input type="checkbox"/> Maternal dosing discontinued at birth <input type="checkbox"/> Pups (treated in utero) fostered to untreated dams <input type="checkbox"/> Control pups fostered to treated dams			
<i>Route of administration</i>				
<i>Dose-response (yes/no)</i>				
Dose Levels Prenatal/Postnatal →	0 (Control) 0/0			
N dams/litters per group				
NOAEL and LOAEL postnatal development (fill in appropriate cell)				
Gestation length				
Liveborn/ total litter size per dam				
Stillbirths per litter				
Sex ratio prior to culling (% males)				
Birth weight per litter (% change versus control)				
Sex-adjusted birth weight (yes/no)				
Progeny culled to (number per litter) On postnatal day				
Postnatal wt (%change vs cntrl) at: -Prewaning* (specify p.n.days) -Weaning* (specify p.n.day) -Postweaning & Maturity*(specify age)				
% wt. gain change vs control by sex: - males (indicate period) - females (indicate period)				
Survival (proportion viable pups) at: - preweaning* (specify p.n.days) - postweaning* (specify age) - maturity * (specify age)				
Malformation rates vs control Age of obtaining malform. data				
Malformations type (description if elevated over control)				
Growth & development (developm. milestones vs control)				
Neurobehavioral development Tests&timing Abnormal effects→				
Reproductive performance F₁				
Fertility index				
N live F ₂ fetuses(pups) per litter				
Pre-implantation lethality (N implants/Corpora lutea per dam)				
Post-implantation lethality (N dead+resorbed(aborted)/ N implants)				
F ₂ Birth weight per litter				
Parental (F ₁) wt (%vs control): M, F				
Maternal (F ₁) wt gain (% vs control)				
Other organ system effects				
Histopathology and/or Gross necropsy				

¹ Effects to be presented as relative to control values; * Periods of preweaning, weaning, postweaning and maturity specific for the species under study (for rat: preweaning=PND 1-21, weaning=PND 21, and maturity ≈ 3 months of age). Key: (-) no change; (•) no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease;

SUMMARY OF ANIMAL REPRODUCTIVE STUDIES (continued)

Summary	NOAEL		LOAEL		Most sensitive endpoint (Limiting parameter)
	Female	Male	Female	Male	
General Toxicity					
Reproductive Toxicity incl:					
- Fertility					
- Prenatal Developmental Toxicity					
- Postnatal Development Toxicity					
- Maternal toxicity during gestation					

Conclusion	
Confounding factors and other comments	
Evaluation*	
<i>Criteria:</i>	
<i>Adequacy of experimental model</i>	
<i>Adequacy of dose and route of adm.</i>	
<i>Adequacy of timing & duration of exposure</i>	
<i>Sufficient n animals per group</i>	
<i>Presence of dose/effect or dose/response relationship</i>	
<i>Appropriate statistical analysis</i>	
<i>Concordance with pharmacokinetic/ pharmacodynamic properties of agent</i>	
<i>Data consistent with other studies</i>	
Study reliable (yes/no)	

*(with regard to reliability of extrapolating study data to humans)

FLUOXETINE

EXTENDED SUMMARIES AND EVALUATION OF the ANIMAL STUDIES (Tables 4.1 – 4.6)

CONTENTS

Table 4.1.

A Fertility Study on Fluoxetine Hydrochloride in the Female Rat.. Lilly Research Laboratories Study No RO 7179 /1980 by J. Wold , N. Owen & E. Adams p. 22

Table 4.2.

A Fertility Study, Incl. Behavioral and Reproductive Assessment of the F₁ Generation, in the Wistar Rat Given Fluox.Hydrochloride (LY110140) in the Diet. Lilly Research Laboratories Study No R10280 & RO4781/1982 by G.Brophy, N. Owen & J. Hoyt p. 26

Table 4.3.

A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat. Lilly Research Laboratories Study No R-77, IND — Toxicology Report No7 / 1979 by J.S Wold and J.K. Markham p. 31

Table 4.4.

A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rat. Lilly Research Laboratories Study No R-207, IND — Toxicology Report No. 8 /1979 by J. S. Wold & J. K. Markham p. 33

Table 4.5.

A Preliminary Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit. Lilly Research Laboratories Study B-7017, IND — Toxicology Report No. 9 /1979 by J. S. Wold & J. K. Markham p. 35

Table 4.6.

A Teratology Study on Fluoxetine (Lilly Compound 110140) in the Rabbit. Lilly Research Laboratories Study B-7087, IND — Toxicology Report No. 10 /1979 by J. S. Wold & J. K. Markham p. 37

SUMMARY OF ANIMAL REPRODUCTIVE (fertility) STUDIES

CHEMICAL: FLUOXETINE

Table 4.1.

REFERENCE: A Fertility Study on Fluoxetine Hydrochloride in the Female Rat.. Lilly Research Laboratories Study No RO 7179 /1980 by J. Wold , N. Owen & E. Adams

Species, strain	Rat, Wistar							
Exposed (males, females, both)	Females							
Exposure doses, timing & duration	0; 2; 5; 12.5 mg/kg/day , two wks prior to mating + gestation+ lactation							
- Males	0							
- Females	0; 2; 5; 12.5 mg/kg/day , two wks prior to mating + gestation+ lactation							
Route of administration	Oral (gavage)							
N generations studied	1							
Dose – Response (yes/no)	yes							
Effects: (relative to control values)								
F₀ Generation	M	F	M	F	M	F	M	F
Dose Levels (mg/kg/day) →	Control		2		5		12.5	
Doses males/females →	0	0	0	2	0	5	0	12.5
N animals per group:	0	30	0	30	0	30	0	30
General Toxicity								
NOAEL and LOAEL (fill in appropriate cell)					NOAEL		LOAEL	
Body weight , g (%change vs control) (indicate period)	237 (test day 15)		235(-)		225(-)		213 ↓10%(±)	
Weight gain, g (%change vs control) (indicate period)	29.7 (test day 1-15)		30.3(-)		26.7(-)		10.1 ↓66%(+)	
Organ weight*	•		•		•		•	
-Liver	•		•		•		•	
-Kidney	•		•		•		•	
Food consumption, g/day (%change vs control(indicate period)	16–18 (test day 1-15)		15-19 (-)		15-18(-)		12-16 ↓12-25%(±)	
Clinical signs	(-)		(-)		(-)		(-)	
Effects listed above attributable to pharmacological effect of compound?								yes
Mortality	0		0		0		0	
Fertility Parameters[†]								
NOAEL and LOAEL (fill in appropriate cell)								NOAEL
Fertility index (% pregnant of total mated)	97%		97%(-)		80% ↓(±)		97% (-)	
Absolute testis, *epididymis weight	•		•		•		•	
Sex accessory gland weight* (prostate seminal vesicle)	•		•		•		•	
Sperm counts	•		•		•		•	
Sperm morphology	•		•		•		•	
Sperm motility	•		•		•		•	
Estrous cycle length	•		•		•		•	
Other female reproductive organ data	•		•		•		•	
Hormonal measures	•		•		•		•	

Key: (–) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease; M male; F female; * adjusted for body weight; -,- no change in males or females

† In determining NOAEL and LOAEL for fertility, also take into account: pre-implantation lethality, post-implantation lethality and fetal viability (see next page : F1 Generation-Prenatal Component)

Fluoxetine: (Table 4.1.continued)				
F₁ Generation – Prenatal Component¹	Ref. A Fertility Study on Fluoxetine Hydrochloride in the Female Rat. Lilly Res. Laboratories Study No RO 7179 /1980 by J. Wold ,N. Owen & E. Adams (Unpubl)			
Species, strain	Rat, Wistar			
Prenatal exposure – timing and duration	2 weeks before mating + Gest. days 0-20			
Route of administration	Oral (gavage)			
Gestation day of sacrifice	G.day 20 (plug day = day 0)			
Dose-response (yes/no)	yes			
Dose Levels (mg/kg/day) →	0 (Control)	2	5	12.5
N dams/litters per group	10/10	10/10	8/8	10/10
NOAEL and LOAEL prenatal development (fill in appropriate cell)				NOAEL
Pre-implantation lethality ² per dam	14%	4.4% (-)	0% (-)	8.5%(-)
N implants/ N Corpora lutea	11.3/13.1	13/13.6	11.9/11.8 C.L. ↓10%(±)	9.7/10.6 C.L. ↓20%(±)
Post-implantation lethality ³ per litter	6%	3.8%(-)	0% (-)	8% ↑(±)
- Dead (mean per litter)	0	0 (-)	0 (-)	0 (-)
- Resorbed (aborted) - mean per litter	0.7	0.5 (-)	0.0 (-)	0.8 (-)
Early:Late resorptions ratio	7:0	5:0 (-)	0	8:0 (-)
Females affected of total, %	60%	40% (-)	0% (-)	30% (-)
- N litters completely resorbed/ N total	0/10	0/10 (-)	0/8 (-)	0/10 (-)
Litter size (N live fetuses per litter)	10.6	12.5(-)	11.9(-)	8.9 (↓16%, ±)
Fetal viability (gestat.survival index) ⁴	100%	100%(-)	100%(-)	100%(-)
Sex ratio (proportion males)	49%	50%(-)	54%(-)	49%(-)
Fetal weight per litter	3.79	3.85	3.86	3.91
% change vs control		(-)	(-)	(-)
Sex-differentiated fetal weight	•	•	•	•
Incidence of malformations per litter (if elevated, describe malformations below)	0% (external only)	0%(-) (external only)	0%(-) (external only)	0%(-) (external only)
Malformations by type [†] (rate and description)				
- External [†]	0%	0%(-)	0%(-)	0%(-)
- Visceral [†]	•	•	•	•
- Skeletal [†]	•	•	•	•
Incidence of deviations/variations per litter	•	•	•	•
- Visceral [†]				
- Skeletal [†]				
Maternal toxicity during gestation				
NOAEL & LOAEL maternal toxicity				NOAEL
Body weight, g g.d. 0-20 (% change vs control)	241-371	246-391(-)	228-364 (-)	215-334 ↓10% (±)
Weight gain during gestation, % (% change vs control)	53	60 (-)	60 (-)	55 (-)
Pregnancy-adjusted weight (yes/no)	no	no	no	no
Food consumption, g g.d. 0-20	21-25	21-26 (-)	18-23 ↓(±)	18-23 ↓(±)
Clinical signs	(-)	(-)	(-)	(-)
Endpoints above attributable to pharmacological effect of compound?				yes
Maternal mortality	0	0	0	0
Necropsy findings	•	•	•	•

¹ Effect to be presented as relative to control values; ² Pre-implantation lethality (%) = [(n C.L.-n impl.) / n C.L.]x100;

³ Post-implantation lethality (%) = (n Dead+Resorbed (aborted)/n implants)x100; ⁴ N live / N total fetuses per litter;

[†] Describe specific malformations which are increased over their control rates

Key: (-) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease;

Fluoxetine: (Table 4.1. continued)				
F₁ Generation – Postnatal Component¹	Ref: A Fertility Study on Fluoxetine Hydrochloride in the Female Rat. Lilly Res. Laboratories Study No RO 7179 /1980 by J. Wold, N. Owen & E. Adams (Unpubl)			
F ₁ Postnatal exposure: (check)	<input checked="" type="checkbox"/> Maternal dosing continued through lactation <input type="checkbox"/> Other modes of postnatal exposure (if yes, describe mode, timing & duration) <input type="checkbox"/> Maternal dosing discontinued at birth <input type="checkbox"/> Pups (treated in utero) fostered to untreated dams <input type="checkbox"/> Control pups fostered to treated dams			
Route of administration	Oral (through breastmilk and maternal chow)			
Dose-response (yes/no)	yes			
Dose Levels (mg/kg/day) → Prenatal/Postnatal →	0 (Control) 0/0	2 2/2	5 5/5	12.5 12.5/12.5
N dams/litters per group	19/19	19/19	16/16	19/19
NOAEL and LOAEL postnatal development (fill in appropriate cell)			NOAEL	LOAEL
Gestation length (days)	21-23	21-23(-)	21-23(-)	21-23(-)
Liveborn/ total litter size per dam	9.7/10.4	11.6/11.7(-)	9.8/10 (-)	8.6/10 (↓10%)(±)
Stillbirths per litter % of all pups	0.73 7%	0.15 (-) 1.3%	0.18 (-) 1.9%	1.36 (↑50%)(±) 13.7%
Sex ratio prior to culling (% males)	54%	54%(-)	49%(-)	56%(-)
Birth weight per litter (% change versus control)	6.9	6.9 (-)	6.9 (-)	6.3 (↓9%)(+)
Sex-adjusted birth weight (yes/no)	no	no	no	no
Progeny culled to (n per litter) On postnatal day	no	no	no	no
Postnatal wt, g (%change vs cntrl) at				
- Prewaning: day 7	14.9	13.8 (↓7%)	14.7 (-)	13.2 (↓11%)(+)
day 14	26.2	23 (↓12%)	26.6 (-)	26.2 (-)
- Weaning (day 21)	37.4	30.5 (↓18%)	38.1 (-)	36.0 (-)
- Maturity	•	•	•	•
% wt. gain change vs control by sex:				
- males (indicate period)	•	•	•	•
- females (indicate period)	•	•	•	•
Survival (% of liveborn pups viable at:				
- Prewaning: day 7	92%	96% (-)	87% (↓5%)(±)	61% (↓31%)(+)
day 14	92%	93% (-)	87%	58% (↓34%)
- Weaning (day 21)	92%	91% (-)	85%	57% (↓35%)
- Maturity	•	•	•	•
Malformation rates vs control	0 (external only)	0 (external only)	0 (external only)	0 (external only)
Age of obtaining malformation data	•	•	•	•
Malformations type				
Growth & development (developm. milestones vs control)	•	•	•	•
Neurobehavioral development	•	•	•	•
Reproductive performance F ₁	•	•	•	•
Fertility index				
N live F ₂ fetuses(pups) per litter				
Pre-implantation lethality (N implants/Corpora lutea per dam)				
Post-implantation lethality (N dead+resorbed(aborted)/ N implants)				
F ₂ Birth weight per litter				
Parental (F ₁) wt (%vs control): M, F				
Maternal (F ₁) wt gain (% vs control)				
Other organ system effects	•	•	•	•
Histopathology and/or Gross necropsy	•	•	•	•

¹ Effects to be presented as relative to control values

Key: (-) no change; • no observation; (+) statistically significant change or trend (p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease;

Fluoxetine (Table 4.1 continued)		Ref: A Fertility Study on Fluoxetine Hydrochloride in the Female Rat.. Lilly Res. Laboratories Study No RO 7179/1980 by J. Wold, N. Owen & E. Adams (Unpubl)			
Summary	NOAEL mg/kg/day		LOAEL mg/kg/day		Most sensitive endpoint (Limiting parameter)
	Female	Male	Female	Male	
General Toxicity	5.0	•	12.5	•	Decreased female weight gain during 2 nd week of dosing in the pre-mating period (st. significant)
Reproductive Toxicity <i>incl:</i>	5.0	•	12.5	•	Reduced F ₁ birthweight & postnatal survival and wt.gain in 1 st wk of life (st. significant)
- Fertility	12.5	•	•	•	↓ <i>in corpora lutea</i> & implants at 12.5 and 5 mg/kg/day (ns* but dose-dependent)
- Prenatal Developmental Toxicity	12.5		•		↓ litter size, live fetuses & ↑ embryo - lethality at 12.5 mg/kg/day (ns but dose-dependent)
- Postnatal Development Toxicity	5.0		12.5		↑ stillbirths ↓ birthweight ↓ survival and wt.gain of progeny in 1 st postnatal week (st significant)
- Maternal toxicity during gestation	12.5		•		↓ food consumption & body wt. at 12.5 (ns but dose-dependent)

* ns=non-significant

Conclusion	Fluoxetine hydrochloride oral dosing (gavage) of female Wistar rats 2 wks prior to mating and during gestation and lactation results in <i>significant reduction of female food consumption & body wt in the pre-mating period</i> (but not during gestation & lactation) and in developmental toxicity (<i>significant ↑ stillbirths, ↓ F₁ birthweight, survival & wt gain in 1st postnatal week</i> , and non-significant but dose-dependent pre-natal effects: ↓ c.l., implants, litter size, ↑ embryo lethality, but no malformations) at 12.5 mg/kg/day (LOAEL). Fertility unaffected. The maternal effects are consistent with the appetite-suppressing pharmacologic action of fluoxetine and probably are not a sign of maternal toxicity. Postnatal manifestations are a more sensitive index of Fluoxetine developmental toxicity in comparison to prenatal. No evidence of a selective reproductive & developmental toxicity. LOAEL: 12.5 mg/kg/day
Confounding factors and other comments	<i>Prenatal component:</i> -Maternal wt not adjusted for wt of uterine content (this confounds maternal wt inter-group comparisons due to the smaller litter size at the highest dose) -Only external malformations examined (actual malformation incidence unknown) <i>Postnatal component:</i> -Progeny not culled; postnatal weight not sex-differentiated -Decrease in progeny wt appears not dose-dependent (result of confounding by litter size which is biggest at the lowest dose level) -Statistical significance of postnatal effects not indicated in the tables (although the significance of the reduced postnatal weight and survival is indicated in the text).
Evaluation**	The conclusions of the study <i>reliable</i> despite of confounding factors: the effect on the most sensitive endpoints (↓ birth wt, early postnatal survival & wt gain) is clearly present at the highest dose although the litter size is the smallest. <i>Limitations:</i> True malformation rate unknown: only external malformations recorded; Postnatal evaluations are limited to progeny weight & survival and based on observations during the preweaning period only.
<i>Criteria:</i>	
<i>Adequacy of experimental model</i>	yes
<i>Adequacy of dose and route of adm.</i>	yes
<i>Adequacy of timing & duration of exposure</i>	yes
<i>Sufficient n animals per group</i>	yes
<i>Presence of dose/effect or dose/response relationship</i>	yes
<i>Appropriate statistical analysis</i>	yes
<i>Concordance with pharmacokinetic/ pharmacodynamic properties</i>	yes
<i>Data consistent with other studies</i>	yes
Study reliable (yes/no)	yes

** (with regard to reliability of extrapolating study data to humans)

SUMMARY OF ANIMAL REPRODUCTIVE (fertility) STUDIES

CHEMICAL: FLUOXETINE

Table 4.2

REFERENCE: A Fertility Study, Incl. Behavioral and Reproductive Assessment of the F₁ Generation, in the Wistar Rat Given Fluox.Hydrochloride (LY110140) in the Diet. Lilly Research Laboratories Study No R10280 & RO4781/1982 by G. Brophy, N. Owen & J. Hoyt

Species, strain	Rat, Wistar							
Exposed (males, females, both)	Both							
Exposure doses, timing & duration	0; 0.002; 0.005; 0.125% in the diet. Start:at weaning (male);6 wks later(female)							
- Males (time-weighted estimates)	0; 1.5; 3.9; 9.7 mg/kg/day, 10 wks prior to mating + throughout breeding							
- Females (time-weighted estimates)	0; 1.3; 3.1; 7.4 mg/kg/day, 3 wks prior to mating + gestation+ lactation (p.d.21)							
Route of administration	Oral (diet)							
N generations studied	2							
Dose – Response (yes/no)	yes							
Effects: (relative to control values)								
F₀ Generation	M	F	M	F	M	F	M	F
Dose Levels (% in diet) →	0 (Control)		0.002%		0.005%		0.0125%	
Doses males/females (mg/kg/day) →	0	0	1.5	1.3	3.9	3.1	9.7	7.4
N animals per group:	40	40	40	40	40	40	40	40
General Toxicity								
NOAEL and LOAEL (fill in appropriate cell)					NOAEL	NOAEL	LOAEL	LOAEL
Body weight, g (pre-mating treatm.day)(% change vs control)	102-503 d. 0-70	258-305 d.0-28	98-498 (-)	261-308 (-)	98-488 (-)	266-307 (-)	97-477 ↓5%(+)	261-280 ↓8%(+)
Weight gain, g (time period) (%change vs control)	401 d. 0-70	46.9 d.0-28	400 (-)	47.3 (-)	390 ↓3%(±)	41.1 (-)	380 ↓5%(+)	19.0 ↓60%(+)
Organ weight*	•	•	•	•	•	•	•	•
-Liver								
-Kidney								
Food consumption, g/day (time period) (%change vs control)	25.3 d.0-70	19.5 2 nd wk of treatmnt	25.2 (-)	20.0 (-)	25.4 (-)	19.0 (-)	24.3 (-)	15.8 ↓19%(+)
Clinical signs	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
Effects listed above attributable/non-attributable to pharmacological effect of the compound? (yes/no)								yes
Mortality	0	0	0	0	0	0	1 of 40	0
Fertility Parameters[†]								
NOAEL and LOAEL (fill in appropriate cell)							NOAEL	
Fertility index (% pregnant of total mated)	88% (35/40)		90% 36/40		80% (↓9%(±) 32/40		78% (↓11%(±) 31/40	
Absolute testis,*epididymis weight	•		•		•		•	
Sex accessory gland weight* (prostate seminal vesicle)	•		•		•		•	
Sperm counts	•		•		•		•	
Sperm morphology	•		•		•		•	
Sperm motility	•		•		•		•	
Estrous cycle length		•		•		•		•
Other female reproductive organ data		•		•		•		•
Hormonal measures	•	•	•	•	•	•	•	•

Key: (–) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease; M male; F female; * adjusted for body weight; -,- no change in males or females

† In determining NOAEL and LOAEL for fertility, also take into account: pre-implantation lethality, post-implantation lethality and fetal viability (see next page : F1 Generation-Prenatal Component)

Fluoxetine:		(Table 4.2 continued)			
F₁ Generation – Prenatal Component¹	REF: A Fertility Study, Incl. Behavioral and Reproductive Assessment of the F ₁ Generation, in the Wistar Rat Given Fluox. Hydrochloride (LY110140) in the Diet. Lilly Res Labs Study No R10280 & RO4781/1982 by G. Brophy, N. Owen & J Hoyt				
Species, strain	Rat, Wistar				
Prenatal exposure – timing and duration	Maternal: 3 weeks before mating + Gest. days 0-20 Paternal: 10 weeks prior to mating + throughout breeding				
Route of administration	Oral (diet)				
Gestation day of sacrifice	G. day 20 (plug day=g.day 0)				
Dose-response (yes/no)	yes				
Dose Levels: Per cent in Diet →	0	0.002	0.005	0.0125	
Time-weighted estimates (mg/kg/day) →	0 (Control)	1.3	3.1	7.4	
N dams/litters per group	17/17	16*/16*	16*/16*	14*/14*	
		*two more dams per test group delivered before necropsy			
NOAEL and LOAEL prenatal development (fill in appropriate cell)				NOAEL	
Pre-implantation lethality² per dam	7.3%	15% (100%↑)	15.1% (100%↑)	17.1% (134%↑)	
N implants/ N Corpora lutea	15.2/16.4	13.5/15.9	14.6/17.2	13.1/15.8	
Post-implantation lethality³ per litter	7%	11% (-)	4% (-)	4% (-)	
- Dead (mean per litter)	0	0 (-)	0 (-)	0 (-)	
- Resorbed (aborted) - mean per litter	1.0	0.9 (-)	0.7 (-)	0.4 (-)	
Early:Late resorptions ratio	17:0	14:0	9:2	5:0	
Females affected of total, %	65% (11/17)	50% (8/16) (-)	44% (7/16) (-)	29% (4/14) (-)	
- N litters completely resorbed/ N total	0/17	0/16 (-)	0/16 (-)	0/14 (-)	
N live fetuses per litter	14.2	12.6 (↓10%,±)	13.9(-)	12.7 (↓10%,±)	
Fetal viability (gestat. survival index)⁴	100%	100%(-)	100%(-)	100%(-)	
Sex ratio (proportion males)	53%	51%(-)	51%(-)	48%(-)	
Fetal weight per litter	3.86	3.75	3.71	3.64	
% change vs control		(-)	(-)	(-)	
Sex-differentiated fetal weight	•	•	•	•	
Incidence of malformations per litter (if elevated, describe malformations below)	0	0	0	0	
Malformations type[†] (rate and descrp)					
- External [†] (n malformed/ n examined)	0% (0/241)	0% (0/220)	0% (0/250)	0% (0/178)	
- Visceral [†]	0% (0/87)	0% (0/79)	0% (0/90)	0% (0/63)	
- Skeletal [†]	0% (0/154)	0% (0/141)	0% (0/160)	0% (0/115)	
Incidence of deviations/ variations /litter					
- Visceral [†]	17% (15/87) (hydronephrosis)	43% (34/79) (hydronephrosis)	20% (18/90) (hydronephrosis)	17% (11/63) (hydronephrosis)	
- Skeletal [†]	4.5% (7/154) rudimentary ribs	2% (3/141)	2% (3/160)	0% (0/115)	
Maternal toxicity during gestation					
NOAEL & LOAEL maternal toxicity				NOAEL	
Body weight, g g.d. 0-20 (% change vs control)	299-440	299-429	327-460	274-394 ↓8-10% (+)	
Weight gain, % (% change vs control)	47.9	44.1(-)	41.9 (-)	44.0 (-)	
Pregnancy-adjusted weight (yes/no)	no	no	no	no	
Food consumption, g g.d. 0-20	20.4-24.3	20-23.5 (-)	21.1-24.1	19-22.2 ↓9%(±)	
Clinical signs	(-)	(-)	(-)	(-)	
Maternal endpoints above attributable to pharmacological effect of the compound?				yes	
Mortality	0	0	0	0	
Necropsy findings		No treatment-related	No treatment-related	No treatment-related	

¹ Effect to be presented as relative to control values; ² Pre-implantation lethality (%) = [(n C.L-n impl.) / n C.L.]x100;

³ Post-implantation lethality (%) = (n Dead+Resorbed (aborted)/n implants)x100; ⁴ N live / N total fetuses per litter;

[†] Describe specific abnormalities which are increased over control rates

Key: (-) no change; • no observation; (+) statistically significant change or trend (p<0.05); (±) statistically non-significant change; † increase; ↓ decrease;

Fluoxetine:		(Table 4.2 continued)			
F₁ Generation – Postnatal Component¹	REF: A Fertility Study, Incl. Behavioral and Reproductive Assessment of the F ₁ Generation, in the Wistar Rat Given Fluox.Hydrochloride (LY110140) in the Diet. Lilly Res Labs Study No R10280 & RO4781/1982 by G.Brophy, N. Owen & J.Hoyt				
F₁ Postnatal exposure: (check)	<input checked="" type="checkbox"/> Maternal dosing continued through lactation <input type="checkbox"/> Other modes of postnatal exposure (if yes, describe mode, timing & duration) <input type="checkbox"/> Maternal dosing discontinued at birth <input type="checkbox"/> Pups(treated in utero) fostered to untreated dams <input type="checkbox"/> Control pups fostered to treated dams				
Route of administration	Oral (through breastmilk & maternal feed)				
Dose-response (yes/no)	yes				
Dose Levels: Per cent in Matern Diet → Time-weighted estimates (mg/kg/day) →	0 0 (Control)	0.002 1.3	0.005 3.1	0.0125 7.4	
N dams/litters per group	18/18	18/18	14/14	15/15	
NOAEL and LOAEL postnatal development (fill in appropriate cell)		NOAEL	LOAEL		
Gestation length (days)	22.1	22 (-)	21.8 (-)	22 (-)	
Liveborn/ total litter size per dam	12.6/12.9	12.8/13.4	12.8/13.3	12.1/12.3	
Stillbirths per litter	0.33	0.55 (-)	0.50 (-)	0.26 (-)	
% of all pups	2.6%	4.1%	3.8%	2.2%	
Sex ratio prior to culling (% males)	47%	50%(-)	49%(-)	48%(-)	
Birth weight per litter	6.8	6.6	6.6	6.0	
(% change versus control)		(-)	(-)	(↓12%±)	
Sex-adjusted birth weight (yes/no)	no	no	no	no	
Progeny culled to (n per litter)	10	10	10	10	
On postnatal day	1	1	1	1	
Postnatal wt, g (%change vs cntrl) at					
- Prewaning: day 7	15.0	14.3 (-)	14.4 (-)	12.5 (↓17%)(+)	
day 14	27.0	27.5 (-)	27.4 (-)	25.5 (-)	
- Weaning (day 21)	42.0	43.9 (-)	41.7 (-)	39.3 (↓6%±)	
- Maturity (day 58) (males/females)	153/128	148/129	127/111 ↓(+)	128/112 ↓(+)	
(day 120) (males/females)	506/323	510/341	497/306	483/302	
% wt. gain by sex:					
- males (indicate period)	353 (p.d. 58-120)	362 (-)	370 (-)	355 (-)	
- females (indicate period)	195 (p.d. 58-120)	212 (-)	194 (-)	190 (-)	
Survival (%of liveborn pups viable at:					
- Prewaning: day 1 (preculling)	96%	92% (-)	85% (↓11%)(±)	82% (↓14%)(±)	
day 7 (postculling)	97%	95% (-)	97% (-)	83% (↓14%)(±)	
day 14	97%	95% (-)	97% (-)	81% (↓16%)(±)	
- Weaning (day 21)	93%	95% (-)	97% (-)	81% (↓12%)(±)	
- Maturity	•	•	•	•	
Malformation rates vs control	0	0	0	0	
Age of obtaining malformation data	Weaning	Weaning	Weaning	Weaning	
Malformations type					
(description if elevated over control)					
Incidence of deviations/variations*	15%(17/116)	7.5% (9/120)	8.4%(13/154)	16% (13/81)	
*(necropsy weanling progeny)	Hydronephrosis, small testis	same	same	same	
Growth & development					
(developm. milestones vs control)	•	•	•	•	
Food consumption F₁ (g)					
- males (period)	25.1(p.d.58-120)	25.3 (-)	25.1 (-)	25.3 (-)	
- females (period)	19.2(p.d.58-120)	20.2 (-)	18.6 (-)	19.5 (-)	
Efficiency of food utilization (g body wt gained per 100g of food consumed)					
- males (period)	23 (p.d. 58-120)	23.4 (-)	24.2 (-)	23 (-)	
- females (period)	16.6(p.d.58-120)	17.2 (-)	17.1 (-)	16.1 (-)	
Neurobehavioral development					
Test(s) & timing: Abnormal effects →					
Auditory startle (day 58)	(-)	(-)	(-)	(-)	
Visual placing (day 58)	(-)	(-)	(-)	(-)	
Rotarod (day 59-63)	(-)	(-)	(-)	(-)	
Poke hole (day 63-66)	(-)	(-)	(-)	(-)	
Reproductive performance F₁					

Fertility index (% pregnant of total mated)	89% (16/18)	74% (14/19)	92% (12/13)	85% (11/13)
N live F ₂ fetuses(pups) per litter (mean)	11.5	12.8 (-)	12.9 (-)	12.6 (-)
Gestation survival (% newborn alive)	94 %	91% (-)	96% (-)	92% (-)
Gestation length (days, mean)	21.9	22.2 (-)	22 (-)	22.2 (-)
F ₂ Birth weight per litter, g (p.d.1)	6.8	6.8 (-)	6.9 (-)	6.9 (-)
Maternal (F ₁) gest wt gain, g	147	157 (-)	152 (-)	150 (-)
Other organ system effects	•	•	•	•
Histopathology	•	•	•	•

¹ Effects to be presented as relative to control values

Key: (-) no change; • no observation; (+) statistically significant change or trend(p<0.05); (±) statistically non-significant change; ↑ increase; ↓ decrease;

Fluoxetine	REF: A Fertility Study, Incl Behavioral and Reproductive Assessment of the F ₁ Generation, in the Wistar Rat Given Fluox.Hydrochloride (LY110140) in the Diet. Lilly Res Labs Study No R10280 & RO4781/1982 by G.Brophy, N. Owen & J.Hoyt				
	NOAEL mg/kg/day		LOAEL mg/kg/day		Effect: Most sensitive endpoints (Limiting parameter)
	Female	Male	Female	Male	
Summary					
General Toxicity	3.1	3.9	7.4	9.7	Significant ↓ in food consumption, body wt & wt gain in both sexes during first weeks of treatment (more expressed in the females)
Reproductive Toxicity <i>incl:</i>	1.3	1.5	3.1	3.9	Reduced F ₁ birthweight & postnatal survival and wt.gain in 1 st wk of life (st. significant)
- Fertility	7.4	9.7	•	•	↑ pre-implantation embryoletality, ↓ fertility index at NOAEL (ns* but dose-dependent)
- Prenatal Developmental Toxicity	7.4		•		↑ pre-implantation embryoletality at NOAEL (ns but dose-dependent)
- Postnatal Development.Toxicity	1.3		3.1		↓ survival and wt. of progeny up to age of 2 months (st. significant)
- Maternal toxicity during gestation	7.4		•		

*ns=non-significant

Notes for the database:

- The main confounder of this study is the uncertainty about the real exposure (dose) levels because the dosing was performed through diet (see "Confounding factors", next page). The apparent "selective" effect of fluoxetine on postnatal parameters (progeny survival and weight) at doses much lower than those affecting the prenatal endpoints, might actually be due to higher than prenatal maternal and pup exposures (a two-fold increase of maternal food consumption during lactation was reported in this study; also the pups could have been additionally exposed to fluoxetine through the maternal chow).
- There are confounders of other endpoints that should be taken into account in data analysis (see "Confounding factors" next page)
- Please note that the male animals of the parental generation are also exposed (the male dose levels should appear in the "dose" sheet). This is the only animal study (of those on file) that involves both male and female parental exposures. It would be worthwhile to compare the outcomes with those in the previous study (A Fertility Study on Fluoxetine Hydrochloride in the Female Rat.. Lilly Res. Laboratories Study No RO 7179 /1980 by J. Wold ,N. Owen & E. Adams) where only the female parental animals were exposed.
- The exposure levels (Treatment) : should be entered not only as "% compound in the chow", but also as " mg/kg/day" (this information is provided in the report) in order to make possible to compare the findings of this study with the rest of the animal (as well as the human) studies.
- Please note that the entries (in the database sheets for this study) entitled "Body Weight Gain-Male (or Female)- days 0-61" (which refer to progeny postnatal day of life) do not actually apply to days 0-61 after birth, but to days 0-61 of the "growth period" started at the age of 58 days (so that day 0 is in fact = postnatal day 58). This confusing issue is explained in page 156 of the original report, first para.

Table 4.2(Continued)

Fluoxetine	REF: A Fertility Study, Incl. Behavioral and Reproductive Assessment of the F ₁ Generation, in the Wistar Rat Given Fluox.Hydrochloride (LY110140) in the Diet Lilly Res Labs Study No R10280 & RO4781/1982 by G Brophy, N. Owen & J.Hoyt
Conclusion	Fluoxetine hydrochloride oral treatment (diet) during growth period of weanling Wistar rats, at 9.7mg/ kg/day for 10 wks(males) and 7.4 mg/kg/day for 4 wks (females) prior to mating, results in <i>significant ↓ in food consumption, body wt & wt gain in both F₀ sexes during the first weeks of treatment (more expressed in the females).</i> Considerable dose-dependent although statistically n.s. <i>increase in preimplantation embryonic lethality & decrease in fertility index</i> at the same doses. Continued female exposure during gestation and lactation causes no effect on maternal wt gain, although food consum. slightly lower (n.s.). No changes in postimplantation lethality, gestation survival, fetal weight and malformation rates. <i>Postnatal effects</i> in progeny induced by lower doses (3.1 and 7.4 mg/kg/day): <i>significant ↓ in F₁ wt gain & survival in 1st wk of life, and weight depression seen up to the age of 2 months</i> although food utilization unimpaired. No sensory or sensory-motor behavioral deviations in mature progeny (at 2-3 months of age) ; no impairment in F ₁ reproductive capacity; F ₂ generation normal. Evidence for selective postnatal developmental toxicity uncertain (confounded by higher exposures during lactation). LOAEL: 3.1(females) and 3.9 (males)mg/kg/day
Confounding factors and other comments	<i>Exposure:</i> Dose levels are approximates (time-weighted estimates of F intake based on mean daily food consumpt.& dietary concentrations). <i>Higher</i> than designated exposures (double the pre-partum levels) during 2 nd wk of lactation due to doubled maternal food intake. F ₀ females in dose groups <i>incorrectly given</i> treatment diets in wk 7 of growth period, then returned to ctrl diets for 1 wk.before resuming treatment. <i>General toxicity measures</i> Initialwt of F ₀ males and pregnant females in highest dose group significantly lower than control at start (confounds exposure-induced wt decrease: lower maternal wt at end gestation is attributable to lower initial wt rather than to maternal toxicity as per cent wt gain during gestation similar to control). <i>Fertility:</i> The clear dose-dependent increase in pre-implantation lethality (exceeding 2.5 times the control at the highest dose) paralleled by a dose-dependent although non-significant decrease in fertility index are not taken into acct. in determining NOAEL for fertility. <i>Prenatal segment:</i> Timing of pregnancy imprecise (pregnancy diagnosed by plug expelled in cage)-resulting in deliveries before scheduled caesarian on g.day 20. Confounds true gestational age (respectively fetal wt) at necropsy. <i>Postnatal segment:</i> The seemingly "selective" effect on F1 postnatal parameters (survival and weight) at parental exposures lower than those affecting prenatal parameters (down to 3.1 mg/kg/day) could be due to actually higher maternal and pup exposures during lactation (see above "Exposure"). Postnatal behavior first assessed at the age of 2 to 3 months (earlier behavioral deviations might have been omitted).
Evaluation**	In general, study reliable but confounded mainly with regard to exposure quantitation due to dosing through diet. <i>Limitations:</i> Information on endpoints affected should be used for qualitative rather than quantitative comparisons. LOAEL and NOAEL levels determined in the study may not be sufficiently reliable. The seemingly "selective" effect on progeny postnatal weight and survival may be due to the doubled maternal/pup exposures during lactation. The reported lack of behavioral deviations may be due to the late testing of progeny (near adulthood) – drug residues would be excreted long before that, having in mind fluoxetine half-life.
<i>Criteria:</i>	
<i>Adequacy of experimental model</i>	yes
<i>Adequacy of dose and route of adm.</i>	yes
<i>Adequacy of timing & duration of exposure</i>	yes
<i>Sufficient n animals per group</i>	yes
<i>Presence of dose/effect or dose/response relationship</i>	yes
<i>Appropriate statistical analysis</i>	yes
<i>Concordance with pharmacokinetic/ pharmacodynamic properties</i>	yes
<i>Data consistent with other studies</i>	yes
Study reliable (yes/no)	Yes (with limitations – see Evaluation)

** (with regard to reliability of extrapolating study data to humans)