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APPROVAL PACKAGE FOR:

APPLICATION NUMBER

21-427

Pharmacology Review(s) #2

PHARMACOLOGY/TOXICOLOGY COVER SHEET
Response to approvable letter

NDA number: 21-427.

Review number: 1.

Sequence number/date/type of submission: N-BZ / 25 March 2003 / Original submission.

Information to sponsor: Yes (X) No ().

Sponsor (or agent): Eli Lilly and Company, Lilly Corporate Center, Indianapolis, IN 46285; (317) 276-2000.

Manufacturer for drug substance: Eli Lilly and Company, Tippecanoe Laboratories, Lafayette, IN 47909.

Reviewer Name: Linda H. Fossom

Division Name: Neuropharmacological Drug Products

HFD# 120

Review Completion Date: September 8, 2003.

Drug:

Code Name: Compound LY248686 HCl (LY246916); this is the S(+)-enantiomer.

Generic Name: duloxetine hydrochloride.

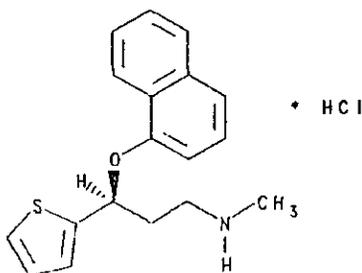
Trade Name: Cymbalta™.

Chemical Name: see below.

CAS Registry Number: 116539-59-4.

Molecular Formula/ Molecular Weight: see below.

Structure:



(+)-N-Methyl-gamma-(1-naphthalen-1-yloxy)-2-thiopropylamine hydrochloride. MW: 333.883

Relevant INDs/NDAs/DMFs: NDA 2

1 IND 2

1 IND 38,838 (HFD-120/depression/enteric coated tablets), IND 2

1, IND 2

Drug Class: Inhibitor of monoaminergic (5-HT and NE) reuptake pumps.

Indication: Depression.

Clinical formulation: gelatin capsule containing enteric-coated pellets of duloxetine HCl (to prevent acid hydrolysis in stomach); proposed dosage forms of 20, 30, — 60 mg of duloxetine.

Route of administration: oral.

Proposed use: Treatment of Major Depression, dosing at 60 mg once daily, although labeling states that some patients might benefit from starting at 30 mg daily, with the target dose remaining 60 mg per day. In proposed labeling, [

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Disclaimer: Tabular and graphical information is excerpted directly from the Sponsor's submission where feasible and cited as such.

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1 INTRODUCTION:

This NDA was determined to be "Approvable" and a letter issued on 9/13/02. In that letter the following Pharmacology/Toxicology concerns were conveyed:

"

Nonclinical Pharmacology and Toxicology

We have completed our review of the nonclinical information provided in your NDA. The following comment should be addressed prior to approval of your NDA.

• With regard to the drug substance impurities, for each of which a specification of not more than has been proposed, we request the following additional information:

- please indicate the amounts of present in lots of drug substance used for pivotal toxicology studies (i.e., genotoxicity, carcinogenicity, and reproduction studies).
- please indicate the amounts of present in lots of drug substance used in the animal reproduction studies.
- if the analytical data requested under the preceding bullet points do not adequately qualify these impurities per the ICH Q3A Guidance, we suggest that you lower the specification limit for each impurity to not more than 0.1%. If this cannot be accomplished, additional studies to qualify these impurities will be needed.

In addition, we have made specific changes in the revised labeling appended to this letter. Please address these changes in your complete response.

"

On December 12, 2002 in a Telecon with Lilly (End of Review Conference), I described the Division's requirements for qualifying the impurities. We require: a) *in vitro* genotoxicity testing, including an Ames test and an *in vitro* chromosomal aberration test; b) a repeat-dose study of at least 2-week duration in one animal species; and additionally c) a Segment II reproductive toxicity study in one animal species, because this drug will be used by women of child-bearing potential.

The resubmission was received on 3/25/03; the 6-month PDUFA date is September 25, 2003.

The Pharmacology/Toxicology section of this resubmission consists of 61 pages (in review copy volume 4) and is comprised of:

- The Sponsor's response to P/T impurity question (pages 3-4);
- Certificates of Analysis for duloxetine drug substance used in toxicology studies, stating the amounts of (pages 6-36) and (pages 37-39);

- Supplemental characterization report concerning the re-analysis for [redacted] in toxicology lots of duloxetine HCl, including a summary table of the results (pages 40-56);
- Information provided in October 31, 2002 Briefing Document to support qualification of [redacted] (pages 57-61).

Additionally, as requested by Doris Bates, Project Manager, the Sponsor resubmitted (under IND 38,838, N-242, stamp-dated 11/13/02) study reports for 3 reproductive toxicology studies (using the maleate salt of duloxetine) that had not been submitted with the original NDA: a Segment I study in male SD rats; and Segment II studies in rabbits and SD rats.

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2 DEFICIENCIES REGARDING QUALIFICATION OF IMPURITIES IN THE ORIGINAL SUBMISSION

The lack of information to qualify the impurities as determined in the original review of this NDA:

In my original review of NDA 21-427, I determined the amounts of — impurities that required qualification (specifically, L J, present in batches of drug substance used in preclinical studies, based upon the Certificates of Analysis provided in the individual study reports using the HCl salt of duloxetine. Below is the summary table (excerpted from page 99 of my original review of this NDA), which was based upon that information. It should be noted that only studies using the HCl salt (not the maleate salt) of duloxetine were included in this original analysis.

Table showing the amounts of impurities [J in toxicology batches of drug substance [data extracted from individual study analytical characterization reports].

I

J

Redacted /

page(s) of trade secret

and/or confidential

commercial information

(b4)

3 ADEQUACY OF QUALIFICATION OF IMPURITIES PROVIDED IN THE CURRENT SUBMISSION

3.1 Requirements for qualification of impurities present in a drug, like duloxetine, to be used to treat Major Depressive Disease.

To qualify — impurities, we would require:

1. *In vitro* gene toxicity testing, including tests for 1) mutations (Ames test) and 2) chromosomal aberrations;
2. A repeat-dose toxicology study of at least 2-week duration in 1 animal species; and
3. A (Segment II) reproductive toxicology study of embryo-fetal development in 1 animal species, because the target population includes women of child-bearing potential.

3.2 Information provided in the current submission regarding amount of — impurities that require qualification:

3.2.1 General information

In the current submission, the Sponsor has provided information on the amounts of the impurities used in toxicology studies that they think qualify the impurities, as summarized in the table, below. It should be noted that all of the studies listed in the table from the original review, above, used batches of duloxetine HCl that had [redacted] when they were re-assayed in 1994 (see table, below), although this impurity was not quantified/specified in the Certificates of Analysis that accompanied the study reports in the original NDA submission.

Furthermore, [redacted], which was undetectable or unquantifiable in the batches of duloxetine HCl used in toxicology studies that were reviewed for the original submission of this NDA (see table, above), was present in considerable amounts in batches of the maleate salt of duloxetine used in some toxicology studies (see table, below). [Reproductive toxicology studies using the maleate salt were not submitted in the original NDA, although they had been submitted earlier under IND [redacted] 1 IND 38,838 and have been re-submitted recently under IND 38,838 (at the request of Doris Bates, Project Manager for this NDA).]

Table 1. Summary of information on [redacted] impurities requiring qualification provided in this submission. This information was obtained from Certificates of Analysis and/or a summary table provided by the Sponsor in this submission (page 41).

[

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submission.

²: According to the Sponsor, this [redacted] impurity was not resolved from the drug substance during initial development, but was separated, identified, and quantified in 1994. The Sponsor provided these values in a table in the current submission (page 41 of this submission; also provided in the Appendix of this review); no Certificates of Analysis were provided, however, the table includes references (i.e., notebook, pages, run number, etc), as well as assay dates.

³: This batch was originally assayed on 6/25/1990 using [redacted] method [redacted] and none of this impurity was detected (LOQ= [redacted]). The Sponsor states that when this lot was subsequently re-analyzed using method [redacted] [redacted] was readily detected and measured at [redacted] (although this amount was below the stated LOQ of [redacted] for this assay method).

3.2.2 Qualification of [redacted]

]

In the current submission, the Sponsor explains that this [redacted] [redacted] was not resolved from the drug substance during initial development, but was separated, identified, and quantified in 1994. The Sponsor also provided a table (available in the Appendix of this review) showing the amounts of this impurity that were determined when the batches were re-analyzed in 1994 (values are also presented in the table, above). No Certificates of Analysis were provided, however, the Sponsor's table includes references (i.e., notebook, pages, run number, etc), as well as assay dates. The amounts of this impurity ranged from [redacted] [redacted] when re-assayed in 1994.

Genotoxicity: All 3 core-battery genotoxicity studies, which were reviewed for the original submission and were considered acceptable and negative, were performed using lot 619NK0 of duloxetine HCl, which contained [redacted] [redacted] of this impurity (when re-assayed in 1994).

- 1) The Ames test was judged to be valid and negative; concentrations from 25 to 400 ug/ml (at 2-fold increments) did not show any evidence of genotoxicity and the slightly higher concentration of 500 ug/ml resulted in essentially complete cytotoxicity with or without metabolic activation.

2) In the test for *in vitro* chromosomal aberrations, duloxetine (4-hr treatment) did not induce chromosomal aberrations in CHO cells at concentrations up to 20ug/ml and 105 ug/ml, without and with metabolic activation, respectively, concentrations that suppressed cell survival 59% and 56 %, respectively.

Although the Reviewer did not consider the *in vitro* test for chromosomal aberrations to be adequate by current standards (i.e., fewer than the recommended number of metaphases were counted and the negative findings after 4-hr treatment were not verified using a 24-hr treatment without metabolic activation), it was decided not to require that this test be repeated for NDA approval. However, the negative results would not be included in labeling, until a test, adequate by current standards, was provided.

3) Additionally, although not required for impurity qualification, the mouse micronucleus assay was valid and negative, using a high dose of 190 mg/kg (by oral gavage) that was approximately half the LD50.

General and Reproductive Toxicity: As was true for the genotoxicity studies (see above), the studies for general toxicity (6-month in rats, 12-month in dogs) and reproductive toxicity (specifically the Segment II studies in rats and rabbits) that were reviewed in detail for the original submission used lots of duloxetine HCl which contained substantial amounts [] of this impurity (when re-assayed in 1994). Consequently, these studies, which were considered adequate to support approval of the original NDA, should also serve to qualify this impurity, although the amounts in the toxicology lots [] are less than the specification set for clinical batches []

Because the amounts of this impurity are lower in the toxicology batches than the [] specification set for the clinical batches, the Sponsor has provided a table showing safety margins for doses of the impurity administered to animals at a NOAEL compared with human daily doses of 60 and 120 mg, based upon mg/kg doses and assuming an average human weight of 60 kg (see table, below). The safety margins, calculated as the ratio of the dose of impurity administered to animals divided by the maximum dose of impurity that might be expected for humans [], provided by NOAEL doses in the studies that are required to qualify this impurity are : ~1-fold for both rats and dogs in 6-month general toxicity studies and ~4-fold for both rats and rabbits in the Segment II reproductive toxicity studies, based upon mg/kg doses (see "Exposure Multiples" in the Sponsor's table, below). When doses are expressed on a mg/m² basis, the safety margins are lower and appear to be more variable across species: 0.2-fold for rats and 0.6-fold for dogs in the 6-month studies, 0.6-fold for rats and 1.2-fold for rabbits in the Segment II reproductive toxicity studies. [It should also be noted that the 1-year dog study that was reviewed in detail for the original NDA used lots of duloxetine HCl, namely 521NK0 and 619NK0, which contained [] of this impurity, respectively, at the same HD of 30 mg/kg that was used in the 6-month study cited by the Sponsor in the current submission. Furthermore, there were no differences in the toxicities evident between the studies; mydriasis and decreased papillary light response, emesis, decreased food consumption at the high dose, and liver toxicity limited

to slightly increased amount of secondary lysosomes (in 1-year study), slightly increased liver phospholipids phosphorous (6-month study), and slight induction of CYP2B and CYP 1A1 at the high dose.]

Table 2. Sponsor's table showing exposure multiples for impurity [] in toxicology batches compared with the specification for clinical batches. [Excerpted directly from page 58 of this submission.]

Table 2. Exposure Multiple for [] at Specification of []	
Study Type, Report Number	Lot Number
Ames Report 24	619NK0
Chromosomal Aberration Report 30	619NK0
Mouse Micronucleus Report 20	619NK0
Rat 6-month study Report 31	508NK0
Dog 6-month study Report 32	508NK0 521NK0
Male Rat Fertility Report 25	508NK0
Rat Developmental Report 26	508NK0
Rabbit Developmental Report 27	508NK0
Female Rat Fertility Report 28	508NK0
Rat Fertility/Developmental Report 35	CTM00027
Mouse Carcinogenicity Report 43	DFD13975
Rat Carcinogenicity Report 44	DFD13975

NOAEL = No-Observed-Adverse-Effect Level
 * Based on a dose of 60 mg/day in a 60 kg human
 * Based on a dose of 120 mg/day (60 mg BID) in a 60 kg human
 NA = Not Applicable
 NOEL for carcinogenicity

It should be noted that the Sponsor has chosen to present information on the 6-month dog study, rather than the 1-year study, in the current submission in support of qualification of this impurity. Both studies used the same oral doses and both certainly meet the duration criterion for a repeated-dose study to qualify an impurity. Presumably the Sponsor chose the 6-month study, because the lots of duloxetine HCl had slightly higher amounts of this impurity; Lots 508NK0 and 521NK0 with [] impurity, respectively, were used in the 6-month study; lots 521NK0 and 619NK0 with [] impurity, respectively, were used in the 1-year study.

It also seems appropriate to consider the safety margins for impurity [] in the highest doses tested in animals compared with the MRHD (120 mg/day). I have calculated this information for the high doses (HD) tested in the animal studies required for qualification, expressed as both mg/kg and mg/m², and presented the results in the table, below. This impurity was tested in animals in general toxicity studies in rats (6-

month) and dogs (6-month and 1-year) and in Segment II reproductive toxicity studies in rats and rabbits at doses up to ~10-20-times the maximum dose recommended for humans, based on mg/kg doses and up to ~3-5-fold bases on mg/m² doses.

Table 3. Dose ratios estimated for impurity [redacted] from the maximum doses in repeated-dose general toxicity studies and Segment II reproductive toxicity studies compared with the maximum recommended human dose (i.e., 120 mg).

STUDY	LOT# (% IMPURITY)	HIGH DOSE		SAFETY MARGIN VS MRHD 120 MG ¹	HIGH DOSE	SAFETY MARGIN VS MRHD 120 MG ¹
		Duloxetine mg/kg	Impurity mg/kg	Impurity mg/kg	Impurity mg/m2	Impurity mg/m2
Rat 6-mo Tox31	508NK0	~47 mg/kg (diet)				
Dog 6-mo Tox32	508NK0 & 521NK0	30 mg/kg				
Dog 1-year Tox33	619NK0 & 521NK0	30 mg/kg				
Rat Seg II Tox26	508NK0	45 mg/kg				
Rabbit Seg II Tox27	508NK0	45 mg/kg				

¹: The MRHD of 120 mg in a 60 kg human is 2.0 mg/kg and the maximum allowed dose of this impurity (with specification of [redacted] is [redacted]

It should also be noted that this impurity was also present at [redacted] in several other toxicology studies that are not required for qualification, but were reviewed in support of the original NDA, including a mouse micronucleus assay, Segment I and III reproductive toxicity studies in rats, and carcinogenicity studies in rats and mice.

3.2.3 Qualification of [redacted]

The studies that would qualify [redacted] impurity were performed using the maleate salt of duloxetine, not the HCl salt that is to be marketed. General toxicology studies and genotoxicity studies using the maleate salt were submitted in the original NDA and were reviewed in some detail there. Reproductive toxicology studies using the maleate salt were not submitted in the original NDA, although they had been submitted earlier under IND [redacted] IND 38,838 and have been re-submitted recently under IND 38,838 (at the request of Doris Bates, Project Manager for this NDA).

The amounts of this impurity in the toxicology batches ranged from [redacted] (see tables, below) and were considerably higher than the [redacted] specification set for clinical batches. Consequently, these studies should qualify this impurity. All these studies with

the maleate salt of duloxetine have been previously reviewed by G. Evoniuk (under IND — in 1991) and by K. Davis-Bruno (under IND — in 1998) and I have consulted their reviews, as well as the review of the original NDA, for the current analysis.

Table 4. Sponsor's table from page 5 of this submission. [NB The batch used in the 3-month dog study was 503NK8, not F58-KYO-152, but the content of [] was the same, i.e., — .]

Table 1. Exposure Multiple for [] Specification of — Additional Genetic and Repeat Dose Studies	
Study Type, Report Number	Lot Number
Ames Report 4	F58-KYO-152
Mouse Lymphoma Report 1	F58-KYO-152
Unscheduled DNA Synthesis Report 2	F58-KYO-152
Sister Chromatid Exchange Report 3	F58-KYO-152
Rat 3-month study Report 5	F58-KYO-152
Dog 3-month study Report 12	F58-KYO-152

NA = not applicable
¹ NOAEL=No-Observed-Adverse-Effect Level
² Based on a dose of 60 mg/day in a 60 kg human
³ Based on a dose of 120 mg/day (60 mg BID) in a 60 kg human
⁴ Proposed spec for

Table 5. Summary table of drug lots used in the Segment II reproductive toxicology studies using duloxetine maleate, resubmitted to IND 38,838 (N-242).

SPECIES	STUDY (YEAR)	LOT #	—	ASSAY DATE
Rabbits	Tox13 (1990)	514NK8	—	7/15/1988
Rats	Tox 17 (1989)	514NK8	—	7/15/1988

Genotoxicity: In support of qualification of this impurity, the Sponsor cites 4 genotoxicity tests using the maleate salt of duloxetine, including *in vitro* testing in an Ames test, a mouse lymphoma assay, and an unscheduled DNA synthesis assay in rat liver hepatocytes and *in vivo* testing for sister chromatid exchange in bone marrow of Chinese hamsters (see table, above), all performed using lot F58-KYO-152 of duloxetine maleate, which contained — of this impurity. Previous reviews of these studies by G. Evoniuk (IND — in 1991) or K. Davis-Bruno (IND — in 1998) did not determine any potential for genotoxicity.

- 1) The Ames test: This study (Tox04) was reviewed under the original NDA and was considered to confirm the negative findings from the pivotal study (Tox24) that had been done using the HCl salt.
- 2) The mouse lymphoma test: This study (Tox01) was reviewed under the original NDA and considered to be negative, but inadequate at that time. Quoting from that review, "Both large and small colonies were apparently counted, however, only the combined counts were presented in this report. Nonetheless, the combined totals were so low, that it would be impossible for either large (mutations) or small (chromosomal aberrations) [colonies] to have been increased in these study results. This could serve as evidence for lack of induction of chromosomal aberrations *in vitro*; however, the 24-hr treatment without activation would still be necessary for an adequate study."
- 3) Other genotoxicity tests: In the original review of this NDA, it was concluded that duloxetine (as the maleate salt) was not genotoxic in 2 other assays: unscheduled DNA synthesis (UDS) assay in primary rat hepatocytes and *in vivo* sister chromatid exchange in Chinese hamster bone marrow.

General toxicity: In support of qualification of this impurity, the Sponsor cites 3-month studies in rats and dogs using lots F58-KYO-152 and 503NK8, respectively, of the maleate salt of duloxetine, both containing — of this impurity. It should be noted that the Sponsor's table, above, contains an error; the batch used in the 3-month dog study was 503NK8, not F58-KYO-152; the study report (provided in the original NDA submission) contained a Certificate of Analysis claiming optical purity of — (by — and indicating — as the — " or — Both 3-month studies were previously reviewed in detail by G. Evoniuk (under IND — in 1991) and reviewed more briefly by K. Davis-Bruno (under IND — in 1998) and by the current Reviewer for the original NDA.

1. In the 3-month study in rats (Tox05, in the original NDA submission), dietary concentrations of 0.005, 0.01, 0.03, and 0.08% duloxetine (as the maleate salt) were administered to F344 rats (— 1 20/sex/dose, with daily doses of duloxetine of approximately 4, 7, 20, and 55 mg/kg, respectively.

The 6-month study using the HCl salt, reviewed for the original NDA (Tox31), used dietary concentrations of 0.005, 0.02, and 0.08% duloxetine (as the HCl salt), administered to F344 rats (— 1 which produced average daily doses of duloxetine of approximately 3, 12, and 47 mg/kg, respectively. Duloxetine **decreased body weights** at the HD and decreased food consumption in MDF and at HD; caused some **liver toxicity**, as indicated by

increased liver enzymes (ALP, ALT and AST) at MD and HD at 3- and/or 6-months, increased liver weights and incidence and severity of midzonal vacuolation in HDM, and induction of P450 at HD; **decreased prostate weight** in HDM, without histopathology; no effect on liver (or lung) phospholipids concentrations at HD.

Similar results were seen in the 3-month study using the maleate salt: **decreased body weight gains** and food consumption at the 2 higher doses; **liver toxicity** was evidenced by slightly increased ALP at HD, increased liver weights in males at 2 higher doses, fatty changes in centrilobular hepatocytes (minimal to moderate) at 2 higher doses without changes in liver phospholipids content, and induction of P450; **decreased prostate weight** at 2 higher doses.

Plasma levels of duloxetine were determined on blood drawn between 8:00-10:00am (3/sex/dose) in both studies (see table, below), using slightly different methods of analysis (Tox31 used [redacted]

[redacted] using LY210448 HCl as internal standard; Tox05 used [redacted]

[redacted] using LY214960 as internal standard). Plasma levels of duloxetine on day 2 were comparable for the 2 salt formulations. After 3 months of dietary exposure, duloxetine levels were approximately 3-times higher after the HCl salt, compared with the maleate salt. It is not apparent whether this difference in levels is related to the salt formulation (HCl versus maleate), the different source of F344 rats [redacted] [redacted], different analysis methods, or is merely coincidental. However, there was an apparent duration- and dose-related sex-difference (females having higher levels than males, especially at the MD after 3 and 6 months) in the 6-month study using the HCl salt in rats from [redacted] [redacted] which wasn't evident in the other study. Nonetheless, the 3-month study with the maleate salt achieved plasma levels within 3-fold of those in the 6-month study (using the HCl salt) that supported the original NDA.

Table 6. Plasma levels of duloxetine measured after dietary exposure to duloxetine as the HCl or maleate salt, in 6-month or 3-month studies, respectively. [Values are means for 3/sex/group, calculated from individual values in the study reports. n/d indicates that values were not determined for that time.]

SALT (STUDY)	DULOEXTINE DOSE		PLASMA DULOEXTINE, NG/ML				
	%in diet	mg/kg	Day 2	Day 16	Day 44	Day 92/93	Day 183
HCl (Tox31)	0.02%	12	40	n/d	n/d	330	246
	0.08%	47	290	n/d	n/d	2000	2000
Maleate (Tox05)	0.03%	20	54	160	180	166	n/d
	0.08%	55	170	630	610	730	n/d

2. In the 3-month in dogs (Tox12, in original NDA submission), oral doses of 3, 10, and 30 mg/kg duloxetine (as the maleate salt) were given to Beagle dogs (4/sex/dose).

The 6-month (Tox32) and 1-year (Tox33) studies using the HCl salt, reviewed for the original NDA, used the same oral doses of duloxetine of 0, 3, 10, and 30 mg/kg. In the 1-year study, toxicities were limited to **mydriasis, and slow or incomplete pupillary light response; emesis; decreased food consumption at the HD; liver toxicity** indicated by slightly increased amount of secondary lysosomes and slight induction of CYP2B at HD. Additionally, in the 6-month study, phospholipids phosphorous concentrations were slightly increased in livers of HDM; this was not seen in the 1-year study.

No new toxicities were apparent in the 3-month study (Tox12) that had not been seen in the 6-month or 1-year studies at the same duloxetine doses using the HCl salt. Toxicities, summarized from the review by G. Evoniuk, were a) clinical signs: emesis (at all doses, 15-30 min after dosing), and mydriasis and slow or incomplete pupillary light reflex (at HD, with reflex resolving after 1 month of treatment); b) no effect apparent for body weights or food consumption; c) decreased heart rate (especially at HD, ↓10-24% when measured 2-4 hr after dosing throughout the study), but no wave form anomalies revealed by EKG; d) some evidence of liver toxicity, including 3-4-fold increase in ALT in a HDM at week 12, slight hepatocellular vacuolation in 1 LDM and 2 HDM, increased (13%) hepatic (but not lung) phospholipid content in HDM, and some induction of P450 enzymes; and e) slightly (~25%), but not significantly, decreased testes weights at HD, with no histopathology findings.

Plasma concentrations of duloxetine were measured in all 3 studies and systemic exposures (AUC and Cmax) were essentially identical for dosing with the maleate salt (measured at days 7 and 87) and dosing with the HCl (measured at day days 8 and 93 in the 6-mo study and after 1 year in the 1-year study).

Reproductive toxicology: The (Segment II) reproductive studies of embryo-fetal development in rats and rabbits using lot 514NK8 of the maleate salt of duloxetine (containing — of this impurity) support of qualification of this impurity. Although reports for these studies were not submitted in the original NDA; they were previously reviewed in detail by G. Evoniuk (under IND — in 1991) and again by K. Davis-Bruno (under IND — in 1998) and have been recently re-submitted under IND 38,838 (N-242, stamp-dated 11/13/2002).

1. In the (Segment II) reproductive toxicity study (Tox17) of duloxetine maleate in rats, duloxetine doses of 3, 15, 55 mg/kg were administered by oral gavage, to Sprague-Dawley rats (25/dose).

The study using the HCl salt, reviewed for the original NDA (Tox26), used duloxetine doses of 2, 10, 45 mg/kg in Sprague-Dawley rats. Maternal toxicity, including decreases in body weight, weight gain and food consumption, was evident at the HD, only. Fetal toxicity was also evident at the HD, only, and included increased pre- and post-implantation losses and decreased fetal weights, but no evidence of teratogenicity.

No new toxicities were apparent in the study using the maleate salt (Tox17). Quoting from my review of the original NDA, the "Sponsor claims maternal toxicity (decreased weight gain and food consumption) at MD [15 mg/kg] and HD [55 mg/kg], and decreased fetal weight at HD, but no effect on fetal viability or morphology at any dose tested. [K. Davis-Bruno's review of this study essentially confirmed the Sponsor's interpretation, specifically that there was no treatment-related teratogenicity.]" G. Evoniuk's review (1991) also supported this conclusion.

Maternal plasma levels of duloxetine were not determined in either study.

2. In the (Segment II) reproductive toxicity study (Tox13) of duloxetine maleate in rabbits, duloxetine doses 3, 15, and 75 mg/kg were administered by oral gavage to New Zealand white rabbits (20/dose).

The study using the HCl salt, reviewed for the original NDA (Tox27), used duloxetine doses of 2, 10, and 45 mg/kg administered by oral gavage to New Zealand white rabbits. Maternal toxicity, including decreases in body weight gain and food consumption, was evident at the HD, only. Fetal toxicity was also evident at the HD, only, and included slightly decreased early resorptions and post implantation losses, decreased fetal weights, and increased number of runts, but no evidence of teratogenicity.

No new toxicities were apparent in the study using the maleate salt (Tox13). Quoting from my review of the original NDA, the "Sponsor claims maternal toxicity (decreased weight gain and food consumption) at MD [15 mg/kg] and HD [75 mg/kg], salivation at HD, and decreased fetal weight at HD, with secondary skeletal retardation of pubis, but no effect on fetal viability at any dose tested. [K. Davis-Bruno's review of this study [1998] essentially confirmed the Sponsor's interpretation, specifically that there was no treatment-related teratogenicity.]"

Maternal plasma levels of duloxetine were not determined in either study.

3.3 Summary of qualification of impurities

It is this Reviewer's conclusion that [redacted] impurities, [redacted] which have specifications of [redacted] for clinical batches of drug substance, have each been adequately qualified, in 1) *in vitro* genotoxicity tests (Ames and chromosomal aberrations); 2) a repeated dose toxicology study of at least 2-week duration; and 3) a (Segment II) reproductive toxicity study of embryo-fetal development.

Qualification of impurity [redacted] is supported by the toxicology studies using duloxetine HCl that were reviewed in support of the original NDA. This [redacted] impurity [redacted] was not resolved from the drug substance during initial development, but was separated, identified, and quantified in 1994. When the lots of drug substance used in the studies required to qualify this impurity were re-analyzed in 1994, they contained [redacted] of the impurity, compared with the specification of [redacted] for clinical batches. The amount of this impurity in toxicology batches was [redacted] of the maximum amount specified for batches to be approved for use in humans. The *in vivo* animal studies of general, repeated-dose toxicity (in rats and dogs) and (Segment II) reproductive toxicity used doses of this impurity (as part of the duloxetine HCl dose) that were 10-20-fold the MRHD (of 120 mg), on a mg/kg basis, and 3-6-fold the MRHD, on a mg/m² basis. Regarding any concern about the adequacy of the amount of this impurity used in the *in vitro* genotoxicity studies, it should be noted that this impurity was also present at [redacted] in several other toxicology studies (that are not required for qualification, but were reviewed in support of the original NDA), specifically a mouse micronucleus assay, Segment I and III reproductive toxicity studies in rats, and carcinogenicity studies in rats and mice.

Qualification of [redacted] [redacted] relies upon early studies (specifically, genotoxicity, 3-month rat and dog toxicology, and Segment II reproductive toxicology studies in rats and rabbits) that were performed using the maleate salt of duloxetine, containing [redacted] of this impurity, compared with the [redacted] specification for clinical batches. The genotoxicity studies required to qualify this impurity included an Ames test and a mouse lymphoma test, both of which were negative. The mouse lymphoma test was not strictly adequate by current standards; specifically, the negative findings after 4 hr treatment should have been verified with a 24-hr treatment (without metabolic activation). However, two other tests of genotoxicity were also negative, tests for unscheduled DNA synthesis (UDS) assay in primary rat hepatocytes and *in vivo* sister chromatid exchange in Chinese hamster bone marrow. As support for repeated-dose general toxicity testing, 3-month studies in both rats and dogs were performed using the maleate salt at the same doses of duloxetine that were used in the longer chronic toxicology studies that supported the original NDA. No new toxicities were apparent in the 3-month studies using the maleate salt that had not been seen in the longer studies at the same duloxetine doses using the HCl salt. Finally, Segment II reproductive toxicity studies were performed in 2 species, rats and rabbits, using the maleate salt at doses of

duloxetine that were at least as high those used in the studies (using the HCl salt) that supported the original NDA.

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4 LABELING

In general, the Sponsor accepted the Agency's recommendations for labeling relating to pre-clinical information.

However, in the drug "DESCRIPTION" section, the Sponsor altered the initial sentence from the Agency's recommended wording of: ' C

to read: "Cymbalta™ (duloxetine hydrochloride) is a selective serotonin and norepinephrine reuptake inhibitor (SSNRI) for oral administration." This is acceptable from a pharmacology/toxicology perspective.

Additionally, for calculation of safety ratios in the sections on "Carcinogenesis, Mutagenesis, Impairment of Fertility" and "Pregnancy," the Sponsor proposes to use — mg as the MRHD, based upon the additional data they have provided in the current submission, although the Agency recommended a MRHD of only 60 mg after reviewing the original submission. It should also be noted that the Sponsor appears to have used — kg as human body weight in calculating safety ratios, whereas the Agency uses 60 kg, even though the Sponsor's calculations give safety margins that are slightly more conservative. Safety ratios should reflect the MRHD accepted by the agency and be calculated using a human body weight of 60 kg.

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5 RECOMMENDATIONS

5.1 General recommendation

The Pharmacology/Toxicology issues that precluded approval of this NDA have been resolved. Adequate information has been submitted to allow qualification of L ¹ impurities, [] whose specifications have been set at — , for the clinical batches, an amount that is above the threshold for qualification. Additionally, the Agency's recommendations for changes in the labeling related to pharmacology/toxicology information have been adequately addressed except for the issue of safety ratios, as discussed below.

5.2 Labeling issues

With regard to labeling, the safety ratios (presented in the sections on "Carcinogenesis, Mutagenesis, Impairment of Fertility" and "Pregnancy") should reflect the MRHD accepted by the Agency and be calculated using a human body weight of 60 kg (see below).

5.2.1 Table showing values calculated for doses in terms of mg/m² and safety ratios for MRHDs of 60 and 120 mg per day.

SPECIES	DOSE		SAFETY RATIO	
	mg/kg *	mg/m ² **	60-mg MRHD	120-mg MRHD
Human	(60mg→) 1	37		
	(120mg→) 2	74		
Mouse	50	150	4.1	2.0
	100	300	8.1	4.1
	140	420	11.4	5.7
Rat	10	60	1.6	0.8
	27	162	4.4	2.2
	30	180	4.9	2.4
	36	216	5.8	2.9
	45	270	7.3	3.6
Rabbit	10	120	3.2	1.6
	45	540	14.6	7.3

* Mg/kg doses in humans were calculated by dividing the daily dose in mg (i.e., 60 or 120 mg) by 60 kg, the average human body weight used by the Agency.

**Doses were converted from mg/kg to mg/m² by multiplying by 37, 3, 6, and 12 in humans, mice, rats, and rabbits, respectively.

5.2.2 If the MRHD is 60 mg/day, then the safety ratios in the following labeling should be used:

Carcinogenesis, Mutagenesis, Impairment of Fertility

Carcinogenesis — Duloxetine was administered in the diet to mice and rats for 2 years.

In female mice receiving duloxetine at dietary doses of approximately 140 mg/kg/day (11 times the maximum recommended human dose [MRHD] on a mg/m² basis), there was an increased incidence of hepatocellular adenomas and carcinomas; the no-effect level was approximately 50 mg/kg (4 times the MRHD on a mg/m² basis). Tumor incidence was not increased in male mice receiving duloxetine at dietary doses up to approximately 100 mg/kg/day (8 times the MRHD on a mg/m² basis).

In rats, dietary doses of duloxetine up to approximately 27 mg/kg/day in females (4 times the MRHD on a mg/m² basis) or approximately 36 mg/kg/day in males (6 times the MRHD on a mg/m² basis) did not increase the incidence of tumors.

Mutagenesis — Duloxetine was not mutagenic in the *in vitro* bacterial reverse mutation assay (Ames test) and was not clastogenic in an *in vivo* chromosomal aberration test in mouse bone marrow cells. Additionally, duloxetine was not genotoxic in an *in vitro* mammalian forward gene mutation assay in mouse lymphoma cells or in an *in vitro* unscheduled DNA synthesis (UDS) assay in primary rat hepatocytes, and did not induce sister chromatid exchange in Chinese hamster bone marrow *in vivo*.

Impairment of Fertility — Duloxetine administered orally to either male or female rats prior to and throughout mating at daily doses up to 45 mg/kg (7 times the maximum recommended human dose [MRHD] on a mg/m² basis) did not alter mating or fertility.

Pregnancy

Pregnancy Category C — In animal reproduction studies, duloxetine has been shown to have adverse effects on embryo/fetal and postnatal development.

When duloxetine was administered orally to pregnant rats and rabbits during the period of organogenesis, there was no evidence of teratogenicity at doses up to 45 mg/kg/day (7 and 15 times the maximum recommended human dose [MRHD] on a mg/m² basis, in rats and rabbits, respectively). However, fetal weights were decreased at this dose, with a no-effect level of 10 mg/kg (2 and 3 times the MRHD on a mg/m² basis, in rats and rabbits, respectively).

When duloxetine was administered orally to pregnant rats throughout gestation and lactation, the survival of pups to 1 day postpartum and pup body weights at birth and during the lactation period were decreased following maternal exposure to 30 mg/kg/day (5 times the MRHD on a mg/m² basis), with a no-effect level of 10 mg/kg. Furthermore, behaviors consistent with increased reactivity, such as increased startle response to noise and decreased habituation of locomotor activity, were observed in pups following maternal exposure to 30 mg/kg/day. Post-weaning growth and reproductive performance of the progeny were not affected adversely by maternal duloxetine treatment.

There are no adequate and well-controlled studies in pregnant women; therefore, duloxetine should be used during pregnancy only if the potential benefit justifies the potential risk to the fetus.

1 pages redacted from this section of
the approval package consisted of draft labeling

6 APPENDIX

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Page containing the Sponsor's summary table, excerpted directly from this submission:

Page 41

The following supplemental characterization data is offered for _____ levels
in selected toxicology lots:

Lot	Reference/Assay Date
508NK0	/
521NK0	
619NK0	
CTM00027	
DPD13975	

Prepared by: _____
14-Jan-2003
Research Scientist, TL448

Reviewed by: _____
14-Jan-02 03
Research Advisor, TL793 11/1/03

Approved by: _____
21-Jan-03
Consultant, TL905

Duloxetine Capsules
NDA 21-427 Response to Approvable Letter

2003
Eli Lilly and Company

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

/s/

Linda Fossom
9/8/03 12:12:43 PM
PHARMACOLOGIST

Barry Rosloff
9/9/03 05:49:55 PM
PHARMACOLOGIST