

# Toxicology Review of MenABCWY Vaccine

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**File:** BLA 125770/0

**Product:** Meningococcal Groups A, B, C, W and Y vaccine

**Subject:** Toxicology study review

**Reviewer:** Andrew O'Carroll, DVM

**Reference:** BLA Sections reviewed

- 4.2.3.2 Repeat-Dose Toxicity
- 4.2.3.5 Reproductive and Developmental Toxicity
- 4.2.3.6 Local Tolerance
- 4.2.3.7 Other Toxicity Studies

**Sponsor:** Pfizer Ireland Pharmaceuticals

## EXECUTIVE SUMMARY

This original BLA submission does not contain any nonclinical toxicology studies for the Meningococcal Groups A, B, C, W, and Y pentavalent vaccine (MenABCWY). Rather, the nonclinical toxicology program consists of the studies for the for two components of the vaccine: MenB-FHbp and MenACWY-TT. MenB-FHbp is licensed in the US as Trumenba® so this BLA includes both repeat-dose toxicity and developmental and reproductive toxicity (DART) studies used for licensure approval. MenACWY-TT is currently licensed in 79 countries globally as Nimenrix® and a package of repeat-dose toxicity, DART and local tolerance studies are included in this submission. Additionally, there are a series of five mixed toxicity studies conducted to provide nonclinical safety and risk assessment for residual (b) (4) which is generated as a by-product of the reaction to generate the activated polysaccharides in the vaccine.

There are no toxicologic issues identified which would preclude approval of this BLA in the intended human population. Treatment-related effects were limited to those which are considered anticipated sequelae of the intended immune response to vaccination and there were no effects on embryonic or postnatal development following maternal vaccination. Sections 8 and 13 of the Prescribing Information (PI) adequately portray the available nonclinical toxicology information included in the submission, specifically how there were no studies on pregnant animals using MenABCWY.

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## INTRODUCTION

**BLA:** 125770/0

**Sponsor:** Pfizer Ireland Pharmaceuticals

**Product:** Meningococcal Groups A, B, C, W, and Y Vaccine

**Proposed use:** “Active immunization to prevent invasive disease caused by *Neisseria meningitidis* groups A, B, C, W, and Y in individuals 10 through 25 years of age.”

**Introduction:** Pfizer Ireland Pharmaceuticals, referred to as “sponsor” for the purpose of this review, has submitted an original biologics licensing application (BLA) for consideration of licensure in the US for their developed Meningococcal Groups A, B, C, W, and Y pentavalent vaccine, which is shortened to “MenABCWY” for the purpose of this review. This is a combination vaccine composed of two currently licensed vaccines indicated to prevent disease from *Neisseria meningitidis*: Trumenba® (MenB-FHbp) and Nimenrix® (MenACWY-TT). Trumenba® is a bivalent vaccine which is composed of 2 outer membrane protein antigens from FHbp subfamilies A and B that are expressed as recombinant lipoproteins. This vaccine was licensed in the US with the same indication as proposed with MenABCWY vaccine in a liquid suspension. Nimenrix® is composed of capsular polysaccharides from groups A, C, W and Y that are (b) (4)

before administration. This vaccine was approved in the European Union in 2012 and is currently licensed in 79 countries globally.

The intended clinical dosing regimen of MenABCWY is for recipients to receive a 0.5 mL dose intramuscularly after reconstitution of lyophilized MenACWY-TT drug product with the MenB-FHbp suspension drug product using a provided, pre-filled syringe. The sponsor recommends recipients receive a second dose at least 6 months after the first dose.

There is no original nonclinical toxicology program for MenABCWY. Instead, the sponsor included studies used towards the licensure of MenB-FHbp and MenACWY-TT citing both communication with FDA Center for Biologics Evaluation and Research (CBER) and FDA Guidance for industry: For the evaluation of combination vaccines for preventable diseases: production, testing and clinical studies (FDA, 1997). The nonclinical toxicology assessment for MenB-FHbp provided in this BLA submission includes two repeat-dose toxicology studies and two developmental and reproductive toxicology (DART) studies. The nonclinical toxicology assessment for MenACWY-TT provided in this BLA submission includes one repeat-dose toxicology study, one DART study and one local tolerance study. In addition, there are five mixed toxicity studies conducted to provide nonclinical safety and risk assessment for residual (b) (4) which is generated as a by-product of the reaction to generate the activated polysaccharides in the vaccine, which is why the sponsor refers to (b) (4) as an impurity. The other nonclinical studies submitted under sections 4.2.1 (pharmacology) are not included in this review.

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#### SECTION 4.2.3.2 REPEAT DOSE TOXICITY

##### REPEATED-DOSE TOXICITY STUDY WITH A MENACWY-TT CANDIDATE VACCINE ADMINISTERED INTRAMUSCULARLY (FIVE TIMES) TO MALE AND FEMALE RABBITS

**Study number:** V8217

**Performing laboratory:** (b) (4)

**Final Report date:** May 10<sup>th</sup>, 2009

**Test article:** MenACWY-TT (Nimenrix®)

**GLP compliance:** Yes

**Experimental design:** MenACWY-TT or saline control was administered intramuscularly five times, at two-week intervals at a dose of 0.5 mL to (b) (4) rabbits. A battery of endpoints considered routine for nonclinical repeat-dose toxicity studies were included this study: clinical examinations, ophthalmologic examinations, body temperature, body weight, food consumption, clinical pathology assessments (hematology and clinical chemistry), serology, and both macroscopic and microscopic postmortem examinations. Half of the animals were euthanized on study day 3 for postmortem examinations with the remaining half euthanized 28 days following the final administration of test and control articles.

**Assessment:** MenACWY-TT was well tolerated by the rabbits in this study as there were no treatment related clinical signs or any toxicologically relevant effects on ophthalmologic examinations, body weight gain, food consumption, body temperature, clinical pathology or organ weights. Treatment-related findings were limited to slight to widespread mixed cell inflammation found on histologic examinations of the injection sites which is considered an anticipated sequelae of the intended immune response to vaccination rather than as a sign of frank toxicity. This effect was found to be either partially or fully reversible by the end of the 28-day recovery period.

##### (b) (4) 263069 (b) (4) AND ALPO<sub>4</sub> (b) (4) 136352): 5-CYCLE (1 DOSE/2 WEEK CYCLE) INTRAMUSCULAR TOXICITY STUDY IN RABBITS

**Study number:** RPT-60511

**Performing laboratory:** Wyeth Drug Safety

**Final report date:** April 26<sup>th</sup>, 2005

**Test article:** (b) (4) (Trumenba®)

**GLP compliance:** Yes

**Experimental design:** (b) (4) was administered intramuscularly five times, at two-week intervals at a dose of either 100 µg (b) (4) and 200 µg/mL aluminum phosphate (AlPO<sub>4</sub>) adjuvant or 400 µg (b) (4) and 400 µg/mL to (b) (4) rabbits. Additional arms were included in the study for saline control and placebo control which included the vaccine vehicle and AlPO<sub>4</sub>. A battery of endpoints considered routine for nonclinical repeat-dose toxicity studies were included this study: clinical examinations, ophthalmologic examinations, body temperature, body weight, food consumption, clinical pathology assessments (hematology and clinical chemistry), serology, and both macroscopic and microscopic postmortem examinations. Half of the animals were euthanized for postmortem

examinations 2 days after the final administration of test and control articles, with the remaining half euthanized 30 days following the final administration of test and control articles.

**Assessment:** (b) (4) was generally well tolerated by the rabbits at both selected doses with treatment-related findings limited to those considered anticipated sequelae of the intended immune response to vaccination rather than as signs of frank toxicity. These included an increased incidence of slight injection site edema and erythema, slight mean body temperature elevations, elevations of the acute phase reactant fibrinogen on blood sampling, and histologic injection site changes such as slight to moderate inflammation and myocyte degeneration or degeneration/regeneration. These changes were all considered partially or fully reversible by the end of the recovery period. One female rabbit in the 100-µg group was found dead on study day 8 (7-days following vaccination) with the cause of death determined to be septicemia. This was deemed incidental and unrelated to treatment.

**(b) (4) VACCINE AND ALPO4 (b) (4) 126352): REPEAT 5 CYCLE (1 DOSE/2 WEEK CYCLE) INTRAMUSCULAR TOXICITY STUDY IN RABBITS**

**Study number:** RPT-74071

**Performing laboratory:** Wyeth Drug Safety

**Final report date:** July 31<sup>st</sup>, 2008

**Test article:** (b) (4) (Trumenba®)

**GLP compliance:** Yes

**Experimental design:** (b) (4) was administered intramuscularly five times, at two-week intervals at a dose of 400 µg (b) (4) and 400 µg/mL to (b) (4) rabbits. Additional arms were included in the study for saline control and placebo control which included the vaccine vehicle and AlPO<sub>4</sub>. A battery of endpoints considered routine for nonclinical repeat-dose toxicity studies were included this study: clinical examinations, ophthalmologic examinations, body temperature, body weight, food consumption, clinical pathology assessments (hematology and clinical chemistry), serology, and both macroscopic and microscopic postmortem examinations. Half of the animals were euthanized for postmortem examinations 3 days after the final administration of test and control articles, with the remaining half euthanized 30 days following the final administration of test and control articles.

**Assessment:** (b) (4) was generally well tolerated by the rabbits with treatment-related findings limited to those considered anticipated sequelae of the intended immune response to vaccination rather than as signs of frank toxicity. These included an increased incidence of slight injection site edema and erythema, slight mean body temperature elevations, elevations of the acute phase reactant fibrinogen on blood sampling, and slight to moderate inflammation at injection sites on histologic examinations. These changes were all considered partially or fully reversible by the end of the recovery period.

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#### SECTION 4.2.3.5 REPRODUCTIVE AND DEVELOPMENTAL TOXICITY

##### EMBRYO-FETAL, PRE- AND POST-NATAL DEVELOPMENTAL STUDY WITH MENACWY-YY CANDIDATE VACCINE ADMINISTERED INTRAMUSCULARLY IN RATS

**Study number:** V7113

**Performing laboratory:** (b) (4)

**Final Report date:** August 28<sup>th</sup>, 2007

**Test article:** MenACWY-TT (Nimenrix®)

**Animal species and strain:** (b) (4) rats (outbred)

**Number of animals per group and sex:** 24

**Age:** 6 to 7 weeks

**Means of administration:** Intramuscular injection, needle and syringe

**Site of administration:** Left and right hind leg musculature

**Volume of injection:** 200 µL (100 µL x2)

**Frequency of administration and study duration:** Administrations of test and control articles occurred 42 and 28 days before mating, and on gestation days (GD) 6, 8, 11, and 15

**Dose:** 5 µg per serogroup (20 µg total)

**GLP compliance:** Yes

**Experimental design:** Animals were acclimated for 11-12 days, randomized, and assigned to 1 of 2 groups according to table 1 below.

<i>Group</i>	<i>Treatment</i>	<i>Caesarean Subset (N)</i>	<i>Littering Subset (N)</i>
1	Saline	24	24
2	MenACWY-TT	24	24

Table 1: Group assignments – this allowed for about 20 pregnant rats per group and subset

The following parameters were evaluated:

<i>Parameters (Dams)</i>	<i>Frequency of Testing</i>
Cageside observations <sup>1</sup>	At least once daily from PMD 0 through PND 25
Injection site observations	3-hours post-dose
Body weight	Premating: weekly Gestation: GD 0, 3, 6, 8, 11, 15, 17, 21 Lactation: PND 1, 7, 14, 21, 25
Food consumption	Premating: weekly Gestation: GD 0-3, 3-6, 6-8, 8-11, 11-15, 15-17, 17-21 Lactation: PND 1-7, 7-14
Fertility and reproductive performance	Post-mating
Caesarean examinations	GD 21
Serology*	PMD 0 and 37, GD 21, and PND 25
Necropsy examinations	GD 21, PND 25

Table 2: Experimental design (dams) – \*blood drawn via orbital puncture; PMD = pre-mating day; GD = gestation day; PND = post-natal day

<sup>1</sup> Cageside observations include mortality, morbidity, general health and signs of toxicity.

<i>Parameters (F1 generation)</i>	<i>Frequency of Testing</i>
Uterine and litter examinations (fetuses)	GD 21
Fetopathological examinations (fetuses)	GD 21
Mortality and clinical observations (pups)	Daily post parturition
Litter size, sex and weight (pups)	PND 1, 7, 14, 21, 25
Sensory development (pups)	PND 1, 14, 19, 21
Serology*	Fetuses: GD 21 Pups: PND 25
Necropsy (pups)	PND 25

**Table 3: Experimental design (F1 generation) – \*see below for phlebotomy procedures; PMD = pre-mating day; GD = gestation day; PND = post-natal day**

**Methods for blood collection:** Blood for serologic examination was drawn from F0 dams via orbital puncture. Blood from F1 generation fetuses was collected via cord blood and pooled per litter, while blood from pups was collected via trunk blood following euthanasia procedures

**Randomization procedure:** Yes, computer-assisted procedure based on body weight

**Statistical analysis plan:** Yes, Fisher's exact test, one-way analysis of variance, and nonparametric analyses were used depending on the endpoint. The following calculations were used for fertility and reproductive performance:

- mating index = (number of females mated/number of females placed with males) x 100
- female fertility index = (number of pregnant females/number of females placed with males) x 100
- female fecundity index = (number of pregnant females/number of mated females) x 100
- pre-implantation loss = [(number of corpora lutea – number of implantation sites) / number of corpora lutea] x 100 (only for the prenatal phase)
- post-implantation loss = [(number of implantation sites – number of live fetuses or pups) / number of implantation sites] x 100 (both prenatal and postnatal phase)
- gestation index = (number of females with live fetuses or pups / number of pregnant females) x 100
- live birth index = (number of pups born alive / number of pups born) x 100
- pup mortality day n = (number of dead pups on day n / total number of pups on day n) x 100
- sex ratio = (number of live male fetuses or pups/number of live fetuses or pups) x 100
- viability index (days 7-25) = (number of pups surviving 25 days / number of pups alive on PN 7) x 100 (only for the postnatal phase).

## RESULTS

**Morbidity and mortality (F0):** All dams **survived** to their scheduled termination.

**Clinical observations (F0):** There were no treatment-related clinical signs in vaccinated dams in this study. Clinical signs were considered incidental as they were commonly observed findings in laboratory rats or occurred with similar incidence between the treated and control groups.

**Body weight (F0):** From PMD 7-14, there was a statistically significant ( $p < 0.05$ ), minimally (approximately 10%) lower body weight in treated dams compared to controls. Otherwise, no significant differences in body weight were observed between treated and control rats.

**Food consumption (F0):** Treated dams had statistically significant ( $p < 0.05$ ), minimally (approximately 5%) reduced food consumption from GD 8 to 15. Otherwise, no significant differences in food consumption were observed between treated and control rats.

### Maternal fertility:

<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Females placed with males	N	48	48
Number of females mated	N	45	46
Mating index	%	94	96
Number of females pregnant	N	42	41
Female fertility index	&	88	85
Fecundity index	%	93	89
Pre-coital time (days)	Mean	2.24	2.57

Table 4: Female fertility and maternal performance – see statistical analysis section on page 7 of this review for details on calculations used for the above indices

### Caesarean examinations:

<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Pregnant at Caesarean section	N	22	22
Dams with no viable fetuses	N	0	0
Dams with live fetuses	N	22	22
Corpora lutea (total)	N	282	270
Corpora lutea (number per animal)	Mean	12.82	12.27
Implantation sites (total)	N	265	259
Implantation sites (number per animal)	Mean	12.05	11.77
Preimplantation loss (total)	N	17	11
Preimplantation loss (number per animal)	Mean	5.53	3.85
Live fetuses (total)	N	254	254
Live fetuses (number per animal)	Mean	11.55	11.55
Live fetuses (% per animal)	Mean	95.98	98.10
Postimplantation loss (total)	N	11	5
Postimplantation loss (% implantation loss per animal)	Mean	4.02	1.90
Dead fetuses (total)	N	0	0



<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Dead fetuses (number per animal)	Mean	0.00	0.00
Resorptions (total)	N	11	5
Resorptions (number per animal)	Mean	0.50	0.23
Resorptions, early (total)	N	11	4
Resorptions, early (number per animal)	Mean	0.50	0.18
Resorptions, late (total)	N	0	1
Resorptions, late (number per animal)	Mean	0.00	0.05
Live male fetuses (total)	N (%)	132 (52)	133 (52)
Live female fetuses (total)	N (%)	122 (48)	121 (48)

Table 5: Caesarean examination parameter results

**Caesarean weights:**

<i>Parameter</i>	<i>Group 1</i>	<i>Group 2</i>
Gravid uterus	66.19	66.08
Carcass	235.58	229.81
Net weight change from day 0	12.57	10.27
Empty uterus	3.719	3.683
Ovaries	0.102	0.098
Fetal weight (all viable fetuses)	4.4282	4.4148
Fetal weight (male fetuses)	4.5301	4.5232
Fetal weight (female fetuses)	4.3186	4.2688

Table 6: Fetal and reproductive organ weights – all values presented in mean grams (g)

**Fetal examinations:** Examinations of all fetuses from the caesarean subsets revealed no skeletal or visceral malformations in any animal from either group. Numerous visceral and skeletal variations and anomalies were observed, but all were considered incidental to the study because they were similar in incidence between fetuses from treated and control dams. The one exception was an increased incidence ( $p < 0.05$ ) of folded retinas in fetuses from treated dams (14) versus those from control dams (4). The report states this is most likely an artifact from processing of the tissues and not related to treatment.

**Natural delivery data:**

<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Females placed with males	N	48	48
Females mated	N	45	46
Pre-coital time (days)	Mean	2.24	2.57
Females pregnant	N	42	41
Females with liveborn	N	20	19
Mating index	%	94	96
Female fecundity index	%	93	89
Female fertility index	%	88	85
Gestation index	%	100	100
Duration of gestation (days)	Mean	21.15	21.26
Females surviving delivery	N (%)	20 (48)	19 (46)
Females with stillborn pups	N (%)	1	0

<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Females with all stillborn pups	N (%)	0	0
Pups delivered (total)	N (mean)	218 (10.9)	201 (10.6)
Liveborn pups	N	217	201
Live birth index	%	100	100
Stillborn pups	N	1	0
Pup mortality, day 1	%	0.5	0
Number of pups lost, PND days 1-4	N	11	11
Pup mortality, day 4	%	5.1	5.5
Pups culled, day 7	N	52	39
Number alive post-culling	N	154	151
Number of pups lost, PND 5-7	N	0	7*
Number of pups lost, PND 8-14	N	0	0
Number of pups lost, PND 15-25	N	0	0
Pups alive, day 25	N	154	144*
Viability index, days 7-25	%	100	95
Number of litters lost entirely	N	0	2
Number of male pups, day 1	N	116	120
Number of male pups at day 21	N	87	85
Postimplantation loss	%	50.68	52.79

**Table 7: Natural delivery data – littering subsets only; PND = postnatal day; \*p<0.05; calculations used for above indices can be found in the statistical analysis section on page 7 of this review**

**Clinical observations (F1):** There were no clinical signs in rat pups from vaccinated dams that were considered attributable to treatment. A single litter from one dam (#187) was found to be cold with no milk in the stomach from PND days 2 to 4, then the litter was found dead on PND 7. This is the reason for the statistically significant ( $p<0.05$ ) increased number of pups lost during PND5-7 in table 7 above. This was considered incidental to the study because this number of offspring and litter loss is within the normal, historical bounds of spontaneous loss in laboratory rats.

**Body weight (F1):** No clinically or statistically significant differences in body weight gain were observed in pups from treated dams compared to those from control dams. Those differences observed were minimal and considered within the normal bounds of biologic variation.

**Sensory development (F1):**

<i>Parameter</i>	<i>Unit</i>	<i>Group 1</i>	<i>Group 2</i>
Surface righting, litters reaching criteria	N	20	19
Surface righting, pups tested	N	217	201
Surface righting, pups reaching criteria	N (%)	96	93
Surface righting, day reaching criteria	Mean	3.14	2.77

**Table 8: Pup surface righting assessment**

**Macroscopic pathology:** There were no abnormal findings in any of the F1 pups allowed to litter from either on postmortem examinations on PND 25. Necropsy examinations of the F0 dams revealed no findings deemed related to treatment. Findings were limited to one

control dam with a cataract, one treated dam with a degenerated eye, and a similar incidence of sparse hair between treated and control dams.

**Serology:** Antibodies against the capsular polysaccharide of *Neisseria meningitidis* serogroup C (PSC) were assessed from serum samples of both F0 and F1 generations from this study via a validated (b) (4) under non-GLP conditions at (b) (4) on August 20<sup>th</sup>, 2007.

<i>Generation</i>	<i>Study Day</i>	<i>Group 1 (SMC)</i>	<i>Group 1 (SC, %)</i>	<i>Group 2 (SMC)</i>	<i>Group 2 (SC, %)</i>
Dams	PMD -3	0.03	0	0.03	0
Dams	PMD 37	0.03	0	25.25	100
Dams	PND 25	0.03	0	0.7	94
Fetuses	GD 21	0.03	0	4.42	100
Pups	PND 7	0.03	0	6.34	100
Pups	PND25	0.03	0	2.34	94

**Table 9: Serology results – GMC = geometric mean concentration; SC = seroconversion rate; PMD = prenat day; GD = gestation day; PND = postnatal day**

Assessment of serum samples for anti-PSC antibodies revealed 100% seroconversion of the treated dams with none of the control animals demonstrating seroconversion. Additionally, there was 100% seroconversion in the F1 fetuses and pups, thus demonstrating passive transfer of anti-PSC antibodies. The assay was further validated by the fact that none of the study animals had detectable antibodies pretest.

**Assessment:** There was no treatment-related mortality in this study, nor was there significance of maternal toxicity. Treatment-related effects in vaccinated dams was limited to minimal, brief/reversible decreases in food consumption and body weight gain without any other clinical signs evident. There were no apparent effects on female fertility as fecundity indices between the groups were similar and between 85% and 93%, though no historical reference range data was provided in this study. There were comparable litter sizes, corpora lutea, implantations and gravid uterine weight observed between treated and control dams that underwent caesarean sections, with comparable litter sizes found.

There were no effects on embryofetal or postnatal development observed in the F1 generation as well. Prenatal effects were limited to the increased incidence of retinal folding. This was believed to be an artifact of processing, and there is evidence in the literature to back this claim (Szczech et al., 1976). Therefore, considering there was no evidence of vision deficiencies in the pups from the littering subset, this finding should be considered incidental to the study. Postnatal health and development for pups from vaccinated dams was similar to the pups from control dams. While no historical data for the test facility was provided, the complete loss of one litter from the treatment group is within the accepted range of spontaneous pup mortality with laboratory rats (Plaut & Davis, 1972).

**\*\*\*The following two developmental and reproductive toxicity studies were reviewed by Dr. Steve Kunder under the Trumenba® BLA 125549. Summaries from those reviews are provided below.\*\*\***

**(b) (4) 263069 (b) (4) AND ALPO4: (b) (4) 136352): A COMBINED INTRAMUSCULAR FERTILITY AND DEVELOPMENTAL TOXICITY STUDY IN FEMALE RABBITS**

**Study number:** RPT-75947

**Test facility:** Wyeth Drug Safety

**Final report date:** March 23<sup>rd</sup>, 2007

**Test article:** (b) (4) (Trumenba®)

**GLP compliance:** Yes

**Experimental design:** (b) (4) was administered intramuscularly to female does at a dose of 200 µg with 0.5 mg/mL AlPO<sub>4</sub> on days 17 and 4 prior to being paired with stud male rabbits, then again after confirmation of pregnancy on GD 10 and 24. A total of 3 arms to the study were included: (b) (4) placebo control (vehicle plus AlPO<sub>4</sub>), and saline control. Half of the does underwent terminal caesarean procedures with examination of fetal litters on GD 29 while the other half were allowed to litter. Endpoints for the F0 dams included: mortality, clinical observations, abortion rate, body weight, food consumption, injection site irritation, fecundity parameters (mating and fertility indices), gravid uterine weight, hysterectomy findings on GD 29 (corpora lutea, litter size, embryo/fetal mortality), parturition, maternal care of offspring for females that delivered, postmortem observations, and macroscopic observations of the injection sites. Endpoints for the F1 kits included litter size, fetal sex distribution, fetal viability, fetal examinations for external, palatal, visceral or skeletal malformations or variations, splenic and thymic weights, and littering kit viability and survival.

**Assessment:** Administration of (b) (4) was well tolerated by the does in this study as there was no evidence of maternal toxicity or effects on female fertility observed. More specifically, there were no clinical signs to indicate systemic illness or effects on body weight gain, or food consumption. Fertility indices were comparable between the groups. Fewer vaccinated does (80%) than either control group (93%) each mated, but this was within the historical reference range for the facility. Uterine examinations at terminal caesareans revealed no adverse effect on implantation numbers, implantation loss or gravid uterine weight; plus, there was comparable fetal sex distribution, fetal litter numbers, fetal viability and no adverse external, visceral, skeletal or palatal malformations or variations observed. For the subsets allowed to litter, there were no treatment-related effects on gestation duration, gestation indices, parturition, litter size, postpartum maternal care of offspring, kit viability/survivability or weights of spleens and thymuses. Serology demonstrated administration of the test article to the vaccinated does and not the controls, while confirming passive transfer of antibodies to their litters.

**MENINGOCOCCAL B VACCINE (b) (4) A COMBINED INTRAMUSCULAR FERTILITY AND DEVELOPMENTAL TOXICITY STUDY IN FEMALE RABBITS**

**Study number:** RPT-75947

**Test facility:** Wyeth Drug Safety

**Final report date:** October 1<sup>st</sup>, 2009

**Test article:** (b) (4) (Trumenba®)

**GLP compliance:** Yes

**Experimental design:** This study was conducted because the number of pregnant does in the caesarean subsets from the previous study (RPT-63113) were considered inadequate to assess for potential effects on embryofetal development per ICH guidelines. This study was conducted to cover that gap, and all does had terminal caesarean procedures on GD 29.

(b) (4) was administered intramuscularly to female does at a dose of 200 µg with 0.5 mg/mL AlPO<sub>4</sub> on days 17 and 4 prior to being paired with stud male rabbits, then again after confirmation of pregnancy on GD 10 and 24. A total of 3 arms to the study were included: (b) (4) placebo control (vehicle plus AlPO<sub>4</sub>), and saline control. Endpoints for the F0 dams included: mortality, clinical observations, abortion rate, body weight, food consumption, injection site irritation, fecundity parameters (mating and fertility indices), gravid uterine weight, hysterectomy findings on GD 29 (corpora lutea, litter size, embryo/fetal mortality), postmortem observations, and macroscopic observations of the injection sites. Endpoints for the F1 kits included fetal sex, weight, and examinations for external, palatal, visceral, and skeletal malformations and variations. Serology testing was performed to assess antibody titers in both the does and kits.

**Assessment:** Administration of (b) (4) was well tolerated by the does in this study as there was no evidence of maternal toxicity or effects on female fertility observed. More specifically, there were no clinical signs to indicate systemic illness or effects on body weight gain, food consumption, maternal performance (mating and fertility indices). Maternal treatment-related effects were limited to minimal, reversible injection site irritation which was also observed in the placebo (adjuvant) control group. Uterine examinations at terminal caesareans revealed no adverse effect on implantation numbers, implantation loss or gravid uterine weight. Additionally, there were no treatment-related effects observed on the F1 generation of kits. More specifically, there was comparable fetal sex distribution, fetal litter numbers, fetal viability or any external, visceral, skeletal or palatal malformations or variations observed. Serology demonstrated administration of the test article to the vaccinated does and not the controls, while confirming passive transfer of antibodies to their litters.

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#### SECTION 4.2.3.6 LOCAL TOLERANCE

##### SINGLE DOSE TOXICITY AND LOCAL TOLERANCE STUDY WITH A MENACWY-TT CANDIDATE VACCINE ADMINISTERED INTRAMUSCULARLY TO MALE AND FEMALE RABBITS

**Study number:** V7912/04

**Performing laboratory:** (b) (4)

**Final report date:** May 10<sup>th</sup>, 2009

**Test article:** MenACWY-TT (Nimenrix®)

**GLP-compliance:** No

**Experimental design:** MenACWY-TT or saline control was administered in a single intramuscular injection to (b) (4) rabbits once at a dose of 0.5 mL. Following

this round of injections, the rabbits were observed for clinical signs, injection site reactogenicity and body weight changes for three days. All study rabbits were then euthanized for macroscopic necropsy examinations and histologic examinations of the injection sites only.

**Assessment:** No treatment-related clinical signs were observed including at the injection sites during this brief study, nor were there any treatment-related effects on body weight observed. Additionally, there were no treatment-related observations during necropsy examinations, including at the injection sites. The only treatment-related effect observed in this study included very slight to slight mononuclear inflammation of the injection sites of all vaccinated rabbits during histologic examinations. This was considered an anticipated sequelae of the intended immune response to vaccination.

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#### SECTION 4.2.3.7 OTHER TOXICITY STUDIES

(b) (4)

**Study number:** 061/1079

**Performing laboratory:** (b) (4)

**Final report date:** April 16<sup>th</sup>, 1996

**Test article:** (b) (4)

**GLP compliance:** Yes

**Experimental design:** This (b) (4) study was conducted to assess for mutagenic potential of (b) (4) by seeing if applying the compound to causes (b) (4) to occur which causes (b) (4)

Following a preliminary test to ensure (b) (4) was not cytotoxic to (b) (4)

(b) (4)

or saline control using the (b) (4) method both with and without the addition of the (b) (4) rat liver homogenate system.

**Assessment:** The preliminary toxicity test demonstrated adequate evidence that (b) (4) was not toxic to the (b) (4) used in this study. The (b) (4) did not reveal any evidence of mutagenicity under the conditions of this study as there were no significant increases in the frequency of (b) (4) between (b) (4) and control treated bacteria, both with and without the (b) (4) metabolic activation system.

(b) (4)

#### SCREENING L5178Y TK +/- MUTATION ASSAY

**Study number:** 061/1080

**Performing laboratory:** (b) (4)

**Final report date:** April 3<sup>rd</sup>, 1996

**Test article:** (b) (4)

**GLP compliance:** Yes

**Experimental design:** This study used L5178Y mouse lymphoma cell cultures to examine for mutagenicity of (b) (4) specifically (b) (4)

(b) (4) concentrations of (b) (4) both with and without (b) (4) metabolic activation was (b) (4) along with this (b) (4) along with vehicle control (b) (4). Mutant colony counts, survivability, viability, and relative total growth were determined.

**Assessment:** Initially some cytotoxicity was observed with the test article, but this was not observed on the day 2 viability assessments. Both the negative and positive controls used in this study resulted in the number of mutant colonies expected for the test facility based on historical control data, validating the study. Both with and without (b) (4) metabolic activation, there were no significant differences in mutant frequency at the (b) (4) in L5178Y cells, or evidence of larger chromosomal changes.

(b) (4) STUDY IN  
THE GUINEA PIG

**Study number:** 061/1081

**Performing laboratory:** (b) (4)

**Final report date:** February 29<sup>th</sup>, 1996

**Test article:** (b) (4)

**GLP compliance:** Yes

**Experimental design:** The purpose of this study was to assess for any potential skin contact sensitization of (b) (4) when applied to male albino guinea pigs via the Guinea Pig (b) (4). Induction of the animals was first performed. The hair was clipped from the shoulder regions of the guinea pigs then a row of three injections, each 0.1 mL, were made on each side of the midline: (b) (4) (b) (4) 1:1 with distilled water, 0.1% w/v solution of (b) (4) in distilled water and a 0.1% w/v emulsion of the test material in a 1:1 preparation of (b) (4) (b) (4) 1:1 with distilled water. After both 24- and 48-hours, the injection sites were evaluated for erythema and edema using the Draize dermal scoring system (Draize, 1959). Control animals also received 3 injections but were comprised of (b) (4) 1 with distilled water, distilled water, and a 50% w/v emulsion of the distilled water in a 1:1 preparation of (b) (4) 1:1 with distilled water. Following 21 days of observation, a square filter paper patch with 10% or 5% w/w (b) (4) in distilled water was applied to the right flank of the animals for 24 hours, then removed and monitored for erythema and edema for 48 hours.

**Assessment:** During the induction step of the study, very slight to well-defined erythema was observed at the sites of all (b) (4) injections up to 48-hours post injection as well as three control group animals. Skin challenge testing found that (b) (4) produced a 30% (3/10) sensitization rate which was classified as a moderate sensitizer to guinea pig skin under the conditions of this study. Only very slight erythema was observed, but 2 animals demonstrated desquamation at the 48-hour observation point.

**(b) (4) DETERMINATION OF THE NO-OBSERVED EFFECT LEVEL (NOEL) FOLLOWING A SINGLE ORAL ADMINISTRATION TO THE RAT**

**Study number:** 061/1122

**Performing laboratory:** (b) (4)

**Final report date:** August 8<sup>th</sup>, 1996

**Test article:** (b) (4)

**GLP compliance:** Yes

**Experimental design:** This study was designed to explore the NOEL for (b) (4) when administered orally to (b) (4) rats. Fasted male and female rats were administered either 50, 200, or 500 mg/kg once via oral gavage. Clinical observations were then recorded at 30 minutes, 1-, 2-, and 4-hours, then daily for 14 days, with daily recording of body weight. Animals that died during that period underwent a necropsy examination.

**Assessment:** Animals that received 50 and 500 mg/kg (b) (4) showed no clinical signs indicative of toxicity over the 14-day observation period. Only 1 rat was administered 500 mg/kg (b) (4), and that rat was found comatose with decreased, labored breathing one-hour post-dose, then found dead 35 minutes later. On necropsy, this animal was found to have abnormally red lungs and dark kidneys and liver. Based on the results of this study, the NOEL for (b) (4) was found to be 200 mg/kg.

**(b) (4) SINGLE DOSE TOXICITY STUDY BY INTRAMUSCULAR ADMINISTRATION TO CD RATS**

**Study number:** 217/024672/AC

**Performing laboratory:** (b) (4)

**Final report date:** July 22<sup>nd</sup>, 2003

**Test article:** (b) (4)

**GLP compliance:** Yes

**Experimental design:** The purpose of this study was to assess for toxicity following a single intramuscular administration of (b) (4) to male and female (b) (4) rats. A total of 5 male and 5 female rats received an injection of either 1 or 10 mg/kg (b) (4), and another group of 10 rats received vehicle alone. The rats were then monitored for mortality, clinical signs, injection site reactogenicity and body weight changes for 14 days. Surviving animals after that period were euthanized for necropsy examinations

**Assessment:** There was no treatment-related mortality or clinical signs (including injection site reactogenicity) throughout the study and comparable weight gain was observed between treated and control animals. Postmortem examinations revealed treatment-related enlargement of the right lumbar lymph node in 2 treated animals. The remainder of the necropsy findings were considered incidental to the study.

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## CONCLUSIONS

This submission is acceptable regarding nonclinical toxicology and adequate data has been presented to demonstrate the safety and tolerability of the two component vaccines of MenABCWY. Evidence of systemic inflammation and positive titers on serology adequately demonstrate the immunomodulatory effect of the vaccine components. There are no toxicologic issues identified which would preclude approval of this BLA in the intended human population. There were no dedicated toxicology studies using the combined MenABCWY formulation, so safety assessment should focus on the available clinical data using the proposed formulation. This also applies to DART data, but Section 8 of the PI document adequately describes the available data from Nimenrix® and Trumenba®, including how the entire human dose of Nimenrix® was not used in that product's DART study. Additionally, there was no evidence of mutagenicity caused by the impurity (b) (4) under the conditions of the provided studies. (b) (4) was well tolerated in a limited-design single-dose toxicity study but was classified as a moderate sensitizer when applied topically to guinea pigs. However, considering this was observed at doses significantly larger than the anticipated human dose (<15 ng per human dose), the risk to human safety is unlikely. Any safety concerns regarding skin sensitization to this vaccine should be derived from the available clinical data.

**Concurrence:** Martin D. Green

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