(*i*) *Duration and frequency of exposure.* For acute study, animals shall be administered MBT over a period not to exceed 24 hours. For subchronic study, animals shall be dosed daily for at least 90 days.

(*ii*) *Route of exposure.* Animals shall be exposed to MBT orally.

(B)(*1*) An acute and subchronic motor activity test shall be conducted with MBT in accordance with §798.6200 of this chapter except for the provisions in paragraphs (d)(5) and (6) of §798.6200.

(*2*) For the purpose of this section the following provisions also apply:

(*i*) *Duration and frequency of exposure.* For acute study, animals shall be administered over a period not to exceed 24 hours. For subchronic study, animals shall be dosed daily for at least 90 days.

(*ii*) *Route of exposure.* Animals shall be exposed to MBT orally.

(C)(*1*) A subchronic neuropathology test shall be conducted with MBT in accordance with §798.6400 of this chapter except for the provisions in paragraphs (d)(5) and (6) of §798.6400.

(*2*) For the purpose of this section, the following provisions also apply:

(*i*) *Duration and frequency of exposure.* Animals shall be dosed daily for at least 90 days.

(*ii*) *Route of exposure.* Animals shall be exposed to MBT orally.

(ii) *Reporting requirements.* (A) The functional observation battery, motor activity, and neuropathology tests shall be completed and the final reports for each test submitted to EPA within 18 months of the effective date of the final rule.

(B) A progress report shall be submitted to EPA for the functional observation battery, motor activity, and neuropathology tests, respectively, 6 months after the effective date of the final rule.

(4) *Mutagenic effects—Chromosomal aberrations*—(i) *Required testing.* (A) A dominant lethal assay shall be conducted with MBT in accordance with §798.5450 of this chapter, using the oral route of administration.

(B) A heritable translocation assay shall be conducted with MBT in accordance with the test guideline specified in §798.5460 of this chapter if MBT produces a positive result in the dominant lethal assay conducted pursuant to paragraph (e)(4)(i)(A) of this section and if, after a public program review, EPA issues a FEDERAL REGISTER notice or sends a certified letter to the test sponsor specifying that the testing shall be initiated.

(ii) *Reporting requirements.* (A) Mutagenic effects—Chromosomal aberration testing of MBT shall be completed and the final report submitted to EPA as follows: Dominant lethal assay, within 12 months after the effective date of this rule; heritable translocation assay, within 24 months after notification under paragraph (e)(4)(i)(B) of this section that the testing shall be initiated.

(B) For the dominant lethal assay, an interim progress report shall be submitted to EPA 6 months after the effective date of the final rule; for the heritable translocation assay, progress reports shall be submitted to EPA at 6-month intervals beginning 6 months after the date of EPA's notification of the test sponsor that testing shall be initiated until submission of the final report.

(f) *Effective date.* (1) The effective date of this final rule is October 21, 1988, except for paragraphs (a)(2), (d)(1)(i), (d)(2)(i)(B)(*3*), and (e)(3)(ii)(A) of this section. The effective date for paragraphs (a)(2), (d)(1)(i), (d)(2)(i)(B)(*3*), and (e)(3)(ii)(A) of this section is March 1, 1990.

(2) The guidelines and other test methods cited in this rule are referenced as they exist on the effective date of the final rule.

[53 FR 34530, Sept. 7, 1988; 53 FR 37393, Sept. 26, 1988, as amended at 55 FR 7326, Mar. 1, 1990; 58 FR 34205, June 23, 1993]

§799.2700 Methyl ethyl ketoxime.

(a) *Identification of test substance.* (1) Methyl ethyl ketoxime (MEKO, CAS No. 96–29–7) shall be tested in accordance with this section.

(2) MEKO of at least 99 percent purity shall be used as the test substance.

(b) *Persons required to submit study plans, conduct tests, and submit data.* All persons who manufacture (including import) or process or intend to manufacture or process MEKO, including persons who manufacture or process or intend to manufacture or process

MEKO as a byproduct, or who import or intend to import products which contain MEKO, after the date specified in paragraph (e) of this section to the end of the reimbursement period, shall submit letters of intent to conduct testing, submit study plans, conduct tests and submit data, or submit exemption applications, as specified in this section, subpart A of this part, and parts 790 and 792 of this chapter for single-phase rulemaking. Persons who manufacture, import, or process MEKO only as an impurity are not subject to these requirements.

(c) *Health effects testing*—(1) *Pharmacokinetics testing*—(i) *Required testing.* Pharmacokinetics testing shall be conducted with MEKO in accordance with paragraph (c)(1)(ii) of this section.

(ii) [Reserved]

(2) *Oncogenicity*—(i) *Required testing.* Oncogenicity testing shall be conducted in accordance with § 798.3300 of this chapter.

(ii) *Route of administration.* MEKO shall be administered either orally or by inhalation.

(iii) *Reporting requirements.* (A) Oncogenicity testing shall be completed and a final report submitted to EPA within 53 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals, beginning 6 months after the date specified in paragraph (e) of this section, until submission of the final report to EPA.

(3) *Developmental toxicity*—(i) *Required testing.* Developmental toxicity testing shall be conducted in a rodent and a nonrodent mammalian species in accordance with § 798.4900 of this chapter.

(ii) *Route of administration.* MEKO shall be administered orally.

(iii) *Reporting requirements.* (A) Developmental toxicity testing shall be completed and a final report submitted to EPA within 15 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals, beginning 6 months after the date specified in paragraph (e) of this section.

(4) *Reproductive toxicity*—(i) *Required testing.* (A) Reproductive toxicity testing shall be conducted orally in accordance with § 798.4700 of this chapter except for the provisions in paragraphs (c) (8)(iii) and (9)(i) of § 798.4700.

(B) For the purpose of this section, the following provisions also apply:

(*1*) The following organs and tissues, or representative samples thereof, shall be preserved in a suitable medium for possible future histopathological examination: Vagina, uterus, oviducts, ovaries, testes, epididymides, vas deferens, seminal vesicles, prostate, pituitary gland, and, target organ(s) of all P and F_1 animals selected for mating.

(*2*)(*i*) Full histopathology shall be conducted on the organs and tissues listed in paragraph (c)(4)(i)(B)(*1*) of this section for all high dose and control P and F_1 animals selected for mating.

(*ii*) The integrity of the various cell stages of spermatogenesis shall be determined, with particular attention directed toward achieving optimal quality in the fixation and embedding. Preparations of testicular and associated reproductive organ samples for histology should follow the recommendations of Lamb and Chapin (1985) under paragraph (d)(1) of this section, or an equivalent procedure. Histopathology of the testes shall be conducted on all P and F_1 adult males at the time of sacrifice, and histological analyses shall include evaluations of the spermatogenic cycle, i.e., the presence and integrity of the 14 cell stages. These evaluations should follow the guidance provided by Clermont and Percy (1957) under paragraph (d)(2) of this section. Information shall also be provided regarding the nature and level of lesions observed in control animals for comparative purposes.

(*iii*) Data on female cyclicity shall be obtained by conducting vaginal cytology in P and F_1 females over the last 3 weeks prior to mating; the cell staging technique of Sadleir (1978) and the vaginal smear method in Hafez (1978) under paragraphs (d)(3) and (d)(7) of this section, respectively, or equivalent methods should be used. Data shall be provided on whether the animal is cycling and the cycle length.

(*iv*) P and F_1 females shall continue to be exposed to MEKO for at least an additional 2 weeks following weaning of offspring to permit them to begin cycling once again. They shall then be

sacrificed and their ovaries shall be serially sectioned with a sufficient number of sections examined to adequately detail oocyte and follicular morphology. The methods of Mattison and Thorgiersson (1979) and Pederson and Peters (1968) under paragraphs (d) (4) and (5) of this section, respectively, may provide guidance. The strategy for sectioning and evaluation is left to the discretion of the investigators, but shall be described in detail in the study plan and final report. The nature and background level of lesions in control tissue shall also be noted.

(*v*) Gross and histopathologic evaluations shall be conducted on the mammary glands in F_1 females and F_2 pups sacrificed at weaning and in adult F_1 females at the termination of the study. Any abnormalities shall be described in the final report.

(ii) *Reporting requirements.* (A) Reproductive toxicity testing shall be completed and a final report submitted to EPA within 29 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals, beginning six months after the date specified in paragraph (e) of this section until submission of the final report to EPA.

(5) *Mutagenic effects—gene mutations*—(i) *Required testing.* The sex-linked recessive lethal assay in *Drosophila* shall be conducted with MEKO in accordance with §798.5275 of this chapter.

(ii) *Reporting requirements.* (A) The sex-linked recessive lethal assay in *Drosophila* shall be completed and a final report submitted to EPA within 18 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals beginning 6 months after the date specified in paragraph (e) of this section.

(6) *Mutagenic effects—chromosomal aberrations*—(i) *Required testing.* (A) An in vivo mammalian bone marrow cytogenetics test shall be conducted with MEKO in accordance with either §798.5385 (chromosomal analysis) of this chapter, or §798.5395 (micronucleus assay) of this chapter except for the provisions in paragraphs (d)(5) (ii), (iii), and (iv) of §§798.5385 and 798.5395.

(B) For the purpose of this section, the following provisions also apply if §798.5385 of this chapter is used in conducting the test:

(*1*) *Dose levels and duration of exposure.* At least three dose levels shall be tested. The highest dose tested shall be the maximum tolerated dose or that dose producing some signs of cytotoxicity (e.g., partial inhibition of mitosis) or shall be the highest dose attainable. Under oral administration, animals shall be exposed once per day for 5 consecutive days. Under administration by inhalation, animals shall be exposed 6 hours per day for 5 consecutive days.

(*2*) *Route of administration.* Animals shall be exposed to MEKO either orally or by inhalation.

(C) For the purpose of this section, the following provisions also apply if §798.5395 of this chapter is used in conducting the test:

(*1*) *Dose levels and duration of exposure.* At least three-dose levels shall be tested. The highest dose tested shall be the maximum tolerated dose or that dose producing some signs of cytotoxicity (e.g., a change in the ratio of polychromatic to normochromatic erythrocytes) or shall be the highest dose attainable. Under oral administration animals shall be exposed once per day for 5 consecutive days. Under administration by inhalation, animals shall be exposed 6 hours per day for 5 consecutive days.

(*2*) *Route of administration.* Animals shall be exposed to MEKO either orally or by inhalation.

(ii) *Reporting requirements.* (A) The oral in vivo mammalian cytogenetics test shall be completed and a final report submitted to EPA within 14 months of the date specified in paragraph (e) of this section. The inhalation in vivo mammalian cytogenetics test shall be completed and a final report submitted to EPA within 17 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals, beginning 6 months after the date specified in paragraph (e) of this section.

(7) *Neurotoxicity*—(i) *Required testing*—(A) *Functional observational battery.* (*1*)

A functional observational battery shall be conducted with MEKO in accordance with § 798.6050 of this chapter except for the provisions in paragraphs (d) (4)(ii), (5), and (6) of § 798.6050.

(*2*) For the purpose of this section, the following provisions also apply:

(*i*) *Route of exposure.* Animals shall be exposed either orally or by inhalation.

(*ii*) *Lower doses.* The data from the lower doses shall show either graded dose-dependent effects in at least two of all the doses tested, including the highest dose, or no neurotoxic (behavioral) effects at any dose tested.

(*iii*) *Duration and frequency of exposure.* For the oral acute testing, animals shall be exposed once. For the oral subchronic testing, animals shall be exposed once per day 5 days per week for a 90-day period. For the inhalation acute testing, animals shall be exposed for 6 hours for 1 day. For the inhalation subchronic testing, animals shall be exposed 6 hours per day 5 days per week for a 90-day period.

(B) *Motor activity*. (*1*) A motor activity test shall be conducted with MEKO in accordance with § 798.6200 of this chapter except for provisions in paragraphs (d) (4)(ii), (5), and (6) of § 798.6200.

(*2*) For the purpose of this section, the following provisions also apply:

(*i*) *Route of exposure.* Animals shall be exposed either orally or by inhalation.

(*ii*) *Lower doses.* The data from the lower doses shall show either graded dose-dependent effects in at least two of all the doses tested including the highest dose, or no neurotoxic (behavioral) effects at any dose tested.

(*iii*) *Duration and frequency of exposure.* For the acute oral testing, animals shall be exposed once. For the oral subchronic testing, animals shall be exposed once per day 5 days per week for a 90-day period. For the acute inhalation testing, animals shall be exposed for 6 hours for 1 day. For the inhalation subchronic testing, the animals shall be exposed for 6 hours per day 5 days per week for a 90-day period.

(C) *Neuropathology*. (*1*) A neuropathology test shall be conducted with MEKO in accordance with § 798.6400 of this chapter except for the provisions in paragraphs (d) (4)(ii), (5), (6), and (8)(iv)(C) of § 798.6400.

(*2*) For the purpose of this section, the following provisions also apply:

(*i*) *Route of exposure.* Animals shall be exposed either orally or by inhalation.

(*ii*) *Lower doses.* The data from the lower doses shall show either graded dose-dependent effects in at least two of all the doses tested including the highest dose, or no neurotoxic (behavioral) effects at any dose tested.

(*iii*) *Duration and frequency of exposure.* Animals shall be exposed orally once per day 5 days per week for a 90-day period; or if exposed by inhalation, for 6 hours per day 5 days per week for a 90-day period.

(*iv*) *Clearing and embedding.* After dehydration, tissue specimens shall be cleared with xylene and embedded in paraffin or paraplast except for the sural nerve which should be embedded in plastic. Multiple tissue specimens (e.g., brain, cord, ganglia) may be embedded together in one single block for sectioning. All tissue blocks shall be labeled to provide unequivocal identification. A suggested method for plastic embedding is described by Spencer et al. in paragraph (d)(6) of this section.

(ii) *Reporting requirements.* (A) The neurotoxicity tests required under this paragraph (c)(7) and administered orally shall be completed and the final results submitted to EPA within 18 months of the date specified in paragraph (e) of this section. The neurotoxicity tests required under this paragraph (c)(7) and administered by inhalation shall be completed and the final results submitted to EPA within 21 months of the date specified in paragraph (e) of this section.

(B) Interim progress reports shall be submitted to EPA at 6-month intervals beginning 6 months after the date specified in paragraph (e) of this section until submission of the final report to EPA.

(d) *References.* For additional background information, the following references should be consulted.

(1) Lamb, J. and Chapin, R.E. ''Experimental models of male reproductive toxicology.'' In: ''Endocrine Toxicity.'' Thomas, J.A., Korach, K.S., and McLachlan, J.A., eds. New York, NY: Raven Press. pp. 85–115. (1985).

(2) Clermont, Y. and Percey, B. ''Quantitative study of the cell population of the seminiferous tubules in immature rats.'' ''American Journal of Anatomy.'' 100:241–267. (1957).

(3) Sadleir, R.M.F.S. ''Cycles and seasons.'' In: ''Reproduction in Mammals: I. Germ Cells and Fertilization.'' Austin, R. and Short R.V., eds. New York, NY: Cambridge Press. Chapter 4. (1978).

(4) Mattison, D.R. and Thorgiersson, S.S. ''Ovarian aryl hydrocarbon hydroxylase activity and primordial oocyte toxicity of polycyclic aromatic hydrocarbons in mice.'' ''Cancer Research.'' 39:3471–3475. (1979).

(5) Pederson, T. and Peters, H. ''Proposal for classification of oocytes and follicles in the mouse ovary.'' ''Journal of Reproduction and Fertility.'' 17:555–557. (1968).

(6) Spencer, P.S., Bischoff, M., and Schaumburg, H.H. ''Neuropathological methods for the detection of neurotoxic disease.'' In: ''Experimental and Clinical Neurotoxicology.'' Spencer, P.S. and Schaumburg, H.H., eds. Baltimore, MD: Williams and Wilkins, pp. 743–757 (1980).

(7) Hafez, E.S., ed., ''Reproduction and Breeding Techniques for Laboratory Animals.'' Chapter 10. Philadelphia: Lea and Febiger. (1970).

(e) *Effective dates.* (1) The effective date of this final rule is October 27, 1989.

(2) The guidelines and other test methods cited in this section are referenced here as they exist on October 27, 1989.

[54 FR 37808, Sept. 13, 1989, as amended at 58 FR 34205, June 23, 1993]

§ 799.3300 Unsubstituted phenylenediamines.

(a) *Identification of test substance.* (1) The unsubstituted phenylenediamines (pda's), *para*-phenylenediamine (*p*-pda, CAS No. 106–50–3), or its sulfate salt (*p*-pda.H_2SO_4, CAS No. 1624–57–75), *meta*-phenylenediamine (*m*-pda, CAS No. 108–45–2), or its sulfate salt (*m*-pda.H_2SO_4, CAS No. 54–17–08), and *ortho*-phenylenediamine (*o*-pda, CAS No. 95–54–5) shall be tested in accordance with this section.

(2) *p*-Pda, *m*-pda, and *o*-pda of at least 98 percent purity shall be used as the test substances. Either the hydrochloride or sulfate salt of *m*-pda shall be used as the test substances. Either the hydrochloride or sulfate salt of *m*-pda shall be used as a test substance in the oncogenicity test in paragraph (c)(2) of this section if the free base proves to be unstable under the conditions of this study. Either the hydrochloride or sulfate salt of *o*-pda, *p*-pda, or *m*-pda shall be used as a test substance in the 90-day subchronic neurotoxicity studies in paragraph (c)(3)(B) of this section if the free base proves to be unstable under the conditions of these studies. The salt(s) shall be of at least 98 percent purity.

(b) *Persons required to submit study plans, conduct tests, and submit data.* (1) All persons who manufacture (including import or by-product manufacture) or process *m*-pda or *m*-pda.H_2SO_4, or intend to manufacture or process *m*-pda or *m*-pda.H_2SO_4, after the effective date of this rule to the end of the reimbursement period shall submit letters of intent to test, submit study plans, conduct tests, and submit data, or submit exemption applications as specified in paragraphs (c), (d), and (e) of this section, subpart A of this part, and parts 790 and 792 of this chapter for single-phase rulemaking.

(2) All persons who manufacture (including import or by-product manufacture) or process *p*-pda, or *p*-pda.H_2SO_4, or intend to manufacture or process *p*-pda, or *p*-pda H_2SO_4, after the effective date of this rule to the end of the reimbursement period shall submit letters of intent to test, submit study plans, conduct tests, and submit data, or submit exemption applications as specified in paragraphs (c)(3), (d), and (e) of this section, subpart A of this part and parts 790 and 792 of this chapter for single-phase rulemaking.

(3) All persons who manufacture (including import or by-product manufacture) or process *o*-pda, or intend to manufacture or process *o*-pda after the effective date of this rule to the end of the reimbursement period shall submit letters of intent to test, submit study plans, conduct tests, and submit data, or submit exemption applications as specified in paragraphs (c)(3), (d), and (e) of this section, subpart A of this part, and parts 790 and 792 of this chapter for single-phase rulemaking.