EVALUATION OF THE EFFECTS OF 3,4-METHYLENEDIOXYMETHAMPHETAMINE (MDMA) ON FEMALE REPRODUCTION IN ADULT WISTAR RATS

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A THESIS WRITTEN IN THE DEPARTMENT OF ANATOMY,
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IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF
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UNIVERSITY OF BENIN

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DECLARATION

I, Aghogho Success Eferavware with Matriculation Number: PG/1817784, hereby declare that this dissertation titled: "Evaluation of the Effects of 3,4-Methylenedioxymethamphetamine (MDMA) on Female Reproduction in Adult Wistar Rats" was carried out by me in the Department of Anatomy, Faculty of Basic Medical Sciences, University of Benin, under the supervision of Professor Christopher Lucky Sakpa and Dr. Chukwuma Vitalis Ezeoku.

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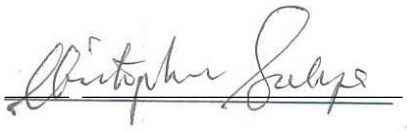 Date: 29/09/25

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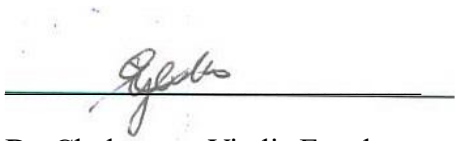 Date: 29/09/25

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DEDICATION

This work is lovingly dedicated to **God Almighty**, the One who authored this journey and saw me through every chapter. To Him be all the glory.

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LIST OF ABBREVATIONS

5-HT: Serotonin

AC: Acidophils

ACTH: Adrenocorticotropic Hormone

ADH: Antidiuretic Hormone

AEND: Atrophic Endometrium

ALJ: Administrative Law Judge

ANOVA: Analysis of Variance

ART: Assisted Reproductive Technologies

BA: Basophils

Bax: BCL2-Associated X protein

Bcl-2: B-cell lymphoma 2

BPA: Bisphenol A

BZ: Basal Zone (of the placenta)

CAT: Catalase

CH: Chromophobes

CIF: Cellular Infiltrate

CL: Corpus Luteum

CNS: Central Nervous System

COMT: Catechol-O-Methyltransferase

CRL: Crown-Rump Length

CSF: Cerebrospinal Fluid

CV: Cytoplasmic Vacuolation

CYP2D6: Cytochrome P450 2D6

CYP3A4: Cytochrome P450 3A4

DA: Dopamine

DB: Decidua Basalis

DF: Degenerating Follicles

DIC: Disseminated Intravascular Coagulation

DMF: Degenerated Muscle Fibers

E2: Estradiol

E2/T Ratio: Estradiol to Testosterone Ratio

ECG: Electrocardiogram

EDCs: Endocrine-Disrupting Chemicals

EDTA: Ethylenediaminetetraacetic Acid

EG: Endometrial Glands

ELISA: Enzyme-Linked Immunosorbent Assay

EMCDDA: European Monitoring Centre for Drugs and Drug Addiction

ERα: Estrogen Receptor alpha

ERβ: Estrogen Receptor beta

ET: Endometrial Thinning

FDA: Food and Drug Administration (U.S.)

FGR: Fetal Growth Restriction

FSH: Follicle-Stimulating Hormone

GABA: Gamma-Aminobutyric Acid

GA: Glandular Atrophy

GDM: Gestational Diabetes

GD: Gestational Day

GH: Growth Hormone

GnRH: Gonadotropin-Releasing Hormone

GPx: Glutathione Peroxidase

H&E: Haematoxylin and Eosin

H_2O_2: Hydrogen Peroxide

HCl: Hydrochloric Acid

HC: Hyperchromasia

HHS: Health and Human Services (U.S. Department of)

HMMA: 4-Hydroxy-3-Methoxymethamphetamine (MDMA metabolite)

HPG axis: Hypothalamic-Pituitary-Gonadal axis

HRT: Hormone Replacement Therapy

HV: Hypophyseal Vessels

ICM: Inner Cell Mass

ICSH: Interstitial Cell-Stimulating Hormone

IL-6: Interleukin-6

IND: Investigational New Drug

IUGR: Intrauterine Growth Restriction

IUPAC: International Union of Pure and Applied Chemistry

$KMnO_4$: Potassium Permanganate

LD_{50}: Median Lethal Dose

LES: Loose Edematous Stroma

LH: Luteinizing Hormone

LIF: Lymphocytic Infiltration

LOAEL: Lowest Observed Adverse Effect Level

LSD: Lysergic Acid Diethylamide

LZ: Labyrinth Zone (of the placenta)

MDA: 3,4-Methylenedioxyamphetamine (MDMA metabolite) / Malondialdehyde

MDEA: 3,4-Methylenedioxy-N-ethylamphetamine

MDMA: 3,4-Methylenedioxymethamphetamine

MF: Maturing Follicles

MG: Metrial Gland

MII: Metaphase II (stage of oocyte maturation)

MSH: Melanocyte-Stimulating Hormone

Na_2CO_3: Sodium Carbonate

$NaHCO_3$: Sodium Bicarbonate

NaCl: Sodium Chloride

NAFDAC: National Agency for Food and Drug Administration and Control (Nigeria)

NDA: New Drug Application

NDLEA: National Drug Law Enforcement Agency (Nigeria)

NE: Norepinephrine

NIDA: National Institute on Drug Abuse (U.S.)

NOAEL: No Observed Adverse Effect Level

OD: Optical Density

OSE: Ovarian Surface Epithelium

PCN: Pharmacists Council of Nigeria

PCOS: Polycystic Ovary Syndrome

PD: Pars Distalis (of the pituitary gland)

PFAS: Per- and Polyfluoroalkyl Substances

PI: Pars Intermedia (of the pituitary gland)

PIBF: Progesterone-Induced Blocking Factor

PIF: Prolactin-Inhibiting Factor

PMK: Piperonyl Methyl Ketone (MDMA precursor)

PMA: Paramethoxyamphetamine

PN: Pars Nervosa (of the pituitary gland)

POMC: Pro-opiomelanocortin

PR: Progesterone Receptor

PRF: Prolactin-Releasing Factor

PRL: Prolactin

PT: Pars Tuberalis (of the pituitary gland)

PTB: Preterm Birth

PYK: Pyknotic Nuclei

ROS: Reactive Oxygen Species

SC: Sinusoid Congestion

SD: Stroma Dilatation

SDGs: Sustainable Development Goals

SEM: Standard Error of the Mean

SI: Sinusoids

SMD: Smooth Muscle Distortion

SOD: Superoxide Dismutase

SPECT: Single-Photon Emission Computed Tomography

SPSS: Statistical Package for the Social Sciences

STC: Stroma Congestion

SV: Stroma Vacuolation

TAAR1: Trace Amine-Associated Receptor 1

TBA: Thiobarbituric Acid

TCA: Trichloroacetic Acid

TNF-α: Tumor Necrosis Factor-alpha

U.S. DEA: United States Drug Enforcement Administration

UNODC: United Nations Office on Drugs and Crime

uNK cells: Uterine Natural Killer cells

VC: Vascular Congestion

VD: Vasodilation

VMAT2: Vesicular Monoamine Transporter 2

VST: Vascularized Stroma

β-LPH: Beta-Lipotropin

ABSTRACT

The increasing recreational use of 3,4-methylenedioxymethamphetamine (MDMA or "ecstasy") among women of reproductive age raises critical concerns regarding fertility and pregnancy outcomes. This study investigated the reproductive and developmental toxicity of MDMA in adult female Wistar rats, focusing on hormonal regulation, oxidative stress, histopathology, and gestational effects following pre-gestational and gestational exposure.

A total of 140 Wistar rats (120 females, 20 males) were assigned to pre-gestational (Category A) and gestational (Category B) protocols. Treated groups received oral MDMA at 80 mg/kg and 160 mg/kg, while controls received distilled water. Serum levels of FSH, LH, PRL, estradiol, progesterone, and testosterone were measured alongside oxidative stress markers (SOD, CAT, GPx, MDA) and histological analyses of the pituitary, ovaries, and uterus. Data were analyzed using one-way ANOVA with LSD post hoc test ($p < 0.05$).

MDMA induced dose-dependent reductions in body and reproductive organ weights, likely due to serotonergic suppression of appetite, oxidative stress, and mitochondrial dysfunction. Hormonal assays revealed significant disruption of the hypothalamic-pituitary-gonadal (HPG) axis, including reduced LH ($p = 0.02$), elevated estradiol ($p = 0.00$), and progesterone ($p = 0.05$). A biphasic testosterone response was observed: reduced in the 80 mg/kg group (0.48 ± 0.00 ng/mL vs. 1.12 ± 0.14 ng/mL in controls) and elevated in the 160 mg/kg group (1.48 ± 0.09 ng/mL; $p = 0.00$), suggesting dysregulation of androgen synthesis via theca cell dysfunction or disrupted HPG feedback. Reproductive outcomes mirrored endocrine alterations. Pre-gestational exposure reduced conception rates (100% in controls vs. 20% and 0% in treated groups). Gestational exposure impaired implantation, fetal viability, and growth, leading to increased resorptions, intrauterine growth restriction, stillbirths, and postnatal abnormalities. Biochemical assays revealed dose-dependent suppression of antioxidant enzymes and altered lipid peroxidation, indicating oxidative damage in ovarian and uterine tissues.

Histopathological evaluation showed progressive degeneration: the pituitary exhibited chromophobe predominance and vacuolation; ovaries showed follicular atresia, degeneration, and vascular injury; and uterine tissues demonstrated glandular atrophy, edema, inflammation, and myometrial disruption. These structural changes aligned with the observed biochemical and hormonal abnormalities.

In conclusion, MDMA exposure before or during pregnancy disrupts female reproductive function in a dose-dependent manner. It impairs fertility, alters endocrine signaling, induces oxidative stress, and causes tissue-specific toxicity, with profound consequences for implantation and fetal development. These findings reinforce the public health risks of MDMA use during reproductive years and the need for targeted reproductive toxicology awareness.

Keywords: MDMA, female reproduction, Wistar rats, endocrine disruption, oxidative stress, fertility, histopathology, pregnancy outcome.