

**PROFERTILITY EFFECT OF FRESH PINEAPPLE JUICE ON THE  
REPRODUCTION PARAMETERS OF FEMALE WISTAR ALBINO RATS**

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**A PROJECT SUBMITTED IN THE DEPARTMENT OF SCIENCE  
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**CERTIFICATION**

We certify that this research work was carried out by **Miss Christabel Esosa OBASUYI** in the Department of Science Laboratory Technology, Faculty of Life Sciences, University of Benin, Benin City, Edo State, Nigeria.

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## **DEDICATION**

I dedicate this research work to God Almighty for His love, strength, wisdom and protection.

## **ACKNOWLEDGEMENT**

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## ABSTRACT

This study investigated the pro-fertility effects of pineapple (*Ananas comosus*) on the reproduction of female Wistar albino rats. Reproductive health represents a fundamental aspect of overall well-being, and the relationship between nutrition and female reproductive function has gained significant attention. The research aimed to evaluate the profertility effects of fresh pineapple juice consumption on reproductive parameters including organ development and hormonal regulation. Thirty Female Wistar albino rats were used as experimental models, with doses of normal control, 5mg/kg Folic acid, 5mg/kg tamoxfene, then 5ml/kg and 20ml/kg of fresh pineapple juice administered over a 21-days treatment period. Results demonstrated significant estrogenic effects with dose-dependent increases in reproductive organ mass, particularly uterine weight, across all treatment groups compared to controls. Hormonal assays revealed notable alterations in reproductive hormone levels, including changes in luteinizing hormone, follicle-stimulating hormone, and estrogen levels, indicating enhanced estrogenic activity and modulation of the hypothalamic-pituitary-ovarian axis. The findings suggest that pineapple extract possesses measurable pro-fertility properties mediated through its complex array of bioactive compounds. The mechanism appears to involve anti-inflammatory effects, antioxidant protection, hormonal modulation that collectively create favorable conditions for reproductive function.

## **CHAPTER ONE**

### **1.0**

### **INTRODUCTION**

#### **1.1 BACKGROUND OF STUDY**

The female reproductive system is a complex network of organs and hormonal interactions responsible for sexual development, reproduction, and maintenance of hormonal balance. It includes both external and internal structures that function together to ensure fertility and the continuation of species. The internal organs, primarily the ovaries, fallopian tubes, uterus, and vagina, work in harmony under the regulation of hormones secreted by the hypothalamus, pituitary gland, and ovaries (Guyton and Hall, 2021).

The ovaries, which are paired glands located in the pelvic cavity, perform dual functions—producing female gametes (ova) and secreting key hormones such as estrogen and progesterone. Estrogen is responsible for the development of secondary sexual characteristics and regulation of the menstrual cycle, while progesterone maintains pregnancy and prepares the endometrium for implantation (Nelson *et al.*, 2019). The fallopian tubes facilitate the transport of ova from the ovaries to the uterus, and fertilization typically occurs in the ampullary region of the tubes. The uterus provides the environment necessary for fetal development, while the vagina serves as the passageway for menstrual flow, sexual intercourse, and childbirth (Marieb and Hoehn, 2019).

Reproductive health, as defined by the World Health Organization (WHO), is a state of complete physical, mental, and social well-being in all matters relating to the reproductive system and its functions (World Health Organization, 2021). It encompasses the ability to have a satisfying and safe sex life, the capability to reproduce, and the freedom to decide if, when, and how often to do so. Female reproductive health plays a vital role in public health, influencing maternal outcomes, fertility rates, and overall quality of life (Temmerman and Khosla, 2020).

The regulation of female reproduction is controlled through the hypothalamic-pituitary-ovarian (HPO) axis, a finely tuned feedback system. Gonadotropin-releasing hormone (GnRH) from the hypothalamus stimulates the anterior pituitary to release follicle-stimulating hormone (FSH) and luteinizing hormone (LH). These hormones regulate follicular development, ovulation, and the menstrual cycle. Disruptions in this axis can lead to infertility, irregular menstruation, and hormonal disorders (Chowdhury *et al.*, 2022).

Globally, reproductive health challenges such as infertility, polycystic ovary syndrome (PCOS), endometriosis, uterine fibroids, and infections significantly affect women of reproductive age. The global prevalence of infertility is estimated at approximately 17.5%, with about half of these cases attributed to female factors (Sun *et al.*, 2019). These disorders not only impact fertility but also affect women's psychological and social well-being.

Nutrition, lifestyle, and environmental factors have profound effects on female reproductive function. Poor nutrition, exposure to endocrine-disrupting chemicals, stress, and obesity are major contributors to reproductive dysfunction (Risso *et al.*, 2020). Antioxidant-rich diets and healthy lifestyles have been associated with improved ovarian function and hormonal regulation (Gaskins and Chavarro, 2018). Regular exercise, adequate sleep, and the maintenance of a healthy body weight contribute to optimal reproductive performance.

Hormonal balance is essential for normal reproductive function. Estrogen and progesterone influence the menstrual cycle, fertility, and pregnancy maintenance. Estrogen promotes the proliferation of the endometrial lining during the follicular phase, while progesterone stabilizes it during the luteal phase to support potential implantation (Burger *et al.*, 2021). An imbalance in these hormones can lead to menstrual irregularities, infertility, and reproductive pathologies such as PCOS and endometrial hyperplasia.

Reproductive health services are critical for addressing women's needs at various stages of life—from puberty through menopause. Access to gynecological care, family planning, and education about reproductive rights are essential components of reproductive well-being. The United Nations Population Fund (UNFPA, 2022) emphasizes the importance of integrating reproductive health into national health programs to reduce maternal morbidity and mortality.

In recent years, research has highlighted the interplay between oxidative stress, inflammation, and reproductive dysfunction. Oxidative stress damages oocytes, impairs follicular development, and disrupts hormonal signaling (Karkovic Markovic *et al.*, 2023). Similarly, chronic inflammation has been implicated in endometriosis and infertility, emphasizing the need for preventive strategies that promote reproductive resilience.

Understanding the female reproductive system and maintaining reproductive health are therefore vital for individual and societal well-being. Effective interventions—including lifestyle modification, early diagnosis of reproductive disorders, and access to medical care—can significantly enhance women's health outcomes and fertility potential.

## **1.2 JUSTIFICATION OF STUDY**

The study of the female reproductive system and its health is justified by its critical role in human reproduction and overall well-being. Female reproductive disorders such as infertility, PCOS, and endometriosis are increasingly prevalent, posing significant medical and

psychosocial challenges. These conditions affect millions of women globally, influencing not only fertility but also metabolic and cardiovascular health (Skoracka *et al.*, 2021).

Understanding the physiological mechanisms of the female reproductive system provides a foundation for diagnosing, preventing, and managing reproductive abnormalities. Moreover, reproductive health research contributes to global health initiatives aimed at improving maternal outcomes, reducing reproductive tract infections, and enhancing fertility awareness (World Health Organization, 2021). Investigating the underlying factors that affect reproductive function—such as hormonal imbalances, oxidative stress, and nutrition—is essential to developing safe and effective interventions that promote female reproductive health.

### **1.3 AIM OF STUDY**

The aim of this study is to investigate the effects of pineapple fruit consumption on female reproductive parameters and to elucidate the potential mechanisms underlying these effects.

### **1.4 OBJECTIVES**

The specific objectives of this study are:

1. to evaluate the effects of pineapple fruit consumption on reproductive hormone levels (FSH, LH, estradiol, EST 2) in female subjects
2. to determine the effects of pineapple consumption on ovarian function and follicular development
3. to examine the antioxidant effects of pineapple consumption in reproductive tissues

4. to elucidate the potential mechanisms by which pineapple bioactive compounds (particularly bromelain) influence reproductive physiology

## **CHAPTER TWO**

### **2.0 LITERATURE REVIEW**

#### **2.1 PINEAPPLE: BOTANICAL CHARACTERISTICS AND NUTRITIONAL COMPOSITION**

##### **2.1.1 BOTANICAL PROFILE**

*Ananas comosus*, commonly known as pineapple, belongs to the family Bromeliaceae and is native to South America, specifically the region encompassing present-day Brazil and Paraguay.

The fruit has been cultivated for centuries and is now grown in tropical and subtropical regions worldwide, including Hawaii, Thailand, the Philippines, Costa Rica, and various African countries. The plant is unique among commercially important fruits as it is not grown from seeds but rather from the crown of the fruit or from slips that develop around the base of the plant.

The pineapple plant is a terrestrial bromeliad that typically reaches maturity 18-24 months after planting, producing a single fruit weighing between 1-3 kilograms. The fruit itself is technically a multiple fruit, formed from the fusion of individual flowers and their bracts.

The cultivation and processing of pineapple can significantly influence its nutritional composition and bioactive compound content. Factors such as variety, growing conditions, harvesting time, storage methods, and processing techniques all affect the final product that consumers receive. Fresh pineapple generally contains higher levels of certain bioactive compounds compared to processed varieties, though some processing methods may concentrate certain nutrients.



Plate 2.1: Diagram of pineapple fruits (*Ananas comosus*) Source: (Thompson *et al*, 2020).

## **2.2 BIOACTIVE COMPONENTS RELEVANT TO REPRODUCTION**

### **2.2.1 BROMELAIN**

Bromelain, a proteolytic enzyme complex found primarily in pineapple stems and fruits, represents one of the most extensively studied components of pineapple from a therapeutic perspective. This enzyme complex consists of several different proteases, each with slightly different properties and mechanisms of action. The concentration of bromelain varies significantly between different parts of the plant, with the stem containing approximately 10 times more bromelain than the fruit flesh.

Research conducted by Thompson *et al.* (2020) revealed that bromelain exhibits significant anti-inflammatory properties through multiple mechanisms, which may be particularly relevant to reproductive health given the role of inflammation in various fertility disorders. Bromelain's anti-inflammatory effects occur through several pathways, including the inhibition of pro-inflammatory prostaglandins, reduction of inflammatory cell infiltration, and modulation of immune cell activity.

The enzyme's ability to modulate inflammatory mediators, including tumor necrosis factor-alpha (TNF- $\alpha$ ), interleukin-6 (IL-6), and nuclear factor kappa B (NF- $\kappa$ B), suggests potential applications in managing inflammatory conditions affecting the reproductive tract. These inflammatory mediators are elevated in various reproductive disorders, including endometriosis, PCOS, and recurrent pregnancy loss, making bromelain's anti-inflammatory properties particularly relevant.

Furthermore, bromelain's fibrinolytic properties may improve blood circulation by breaking down fibrin clots and reducing blood viscosity. Enhanced circulation could potentially benefit

reproductive function by improving blood flow to reproductive organs, enhancing endometrial receptivity, and supporting optimal conditions for implantation success. The improved circulation may also facilitate better delivery of nutrients and hormones to reproductive tissues. Recent investigations by Morrison *et al.* (2021) have shown that bromelain can also modulate immune responses, which may be particularly relevant during the implantation window when maternal immune tolerance is crucial for successful pregnancy establishment. The enzyme appears to promote a shift toward anti-inflammatory immune responses while maintaining appropriate immune surveillance, a balance that is essential for reproductive success.

### **2.2.2 VITAMIN C AND ANTIOXIDANT ACTIVITY**

Pineapple is an excellent source of vitamin C (ascorbic acid), providing approximately 80% of the recommended daily intake per serving, making it comparable to citrus fruits in this regard. Vitamin C plays crucial roles in reproductive physiology that extend far beyond its well-known antioxidant properties. The vitamin is essential for collagen synthesis, which is important for maintaining the structural integrity of reproductive tissues, including the ovaries, fallopian tubes, and uterus.

In reproductive physiology, vitamin C is involved in hormone production and regulation. The vitamin is required for the synthesis of several hormones, including those produced by the adrenal glands that can influence reproductive function. Additionally, vitamin C supports the conversion of cholesterol to steroid hormones, including estrogen and progesterone, making adequate vitamin C status important for hormonal balance.

Research by Chen and Wang (2021) demonstrated that adequate vitamin C status is associated with improved ovarian function and reduced risk of pregnancy complications. The study found

that women with higher plasma vitamin C levels had better ovarian reserve markers and improved response to fertility treatments. This relationship may be explained by vitamin C's role in protecting oocytes from oxidative damage and supporting healthy follicular development.

The antioxidant capacity of pineapple extends beyond vitamin C to include various phenolic compounds and flavonoids, creating a complex antioxidant system that may provide superior protection compared to individual compounds. These antioxidants work synergistically to neutralize reactive oxygen species (ROS) that can damage reproductive cells and tissues, potentially improving overall reproductive function.

Studies by Fletcher and Roberts (2019) have shown that the phenolic compounds in pineapple exhibit synergistic antioxidant effects, providing greater protection than individual compounds alone. This synergistic effect is important because it suggests that consuming whole fruit may be more beneficial than taking isolated vitamin C supplements. The phenolic compounds in pineapple include caffeic acid, ferulic acid, and various flavonoids, each contributing to the overall antioxidant capacity.

The antioxidant effects of pineapple may be particularly important in protecting against age-related decline in reproductive function. As women age, oxidative stress increases while antioxidant defenses decline, contributing to reduced oocyte quality and increased risk of pregnancy complications. Regular consumption of antioxidant-rich foods like pineapple may help counteract these age-related changes.

### 2.2.3 MANGANESE AND MINERAL CONTENT

Pineapple is particularly rich in manganese, a trace element essential for proper enzyme function and metabolic processes. A single cup of fresh pineapple provides approximately 75% of the daily recommended intake of manganese, making it one of the best dietary sources of this important mineral. Manganese serves as a cofactor for numerous enzymes involved in metabolism, antioxidant defense, and connective tissue formation.

In reproductive physiology, manganese plays several important roles. The mineral is essential for the proper function of superoxide dismutase (SOD), one of the body's primary antioxidant enzymes. Adequate SOD activity is crucial for protecting reproductive cells from oxidative damage. Manganese deficiency has been linked to reproductive disorders, including impaired ovulation, irregular menstrual cycles, and increased risk of pregnancy complications.

Studies suggest that adequate manganese intake supports proper steroid hormone synthesis by serving as a cofactor for enzymes involved in cholesterol metabolism and hormone production. The mineral is also important for glucose metabolism and insulin sensitivity, factors that can significantly influence reproductive function, particularly in women with PCOS or metabolic syndrome. Additionally, pineapple contains significant amounts of copper and zinc, both of which are essential for reproductive health. Cooper *et al.* (2020) demonstrated that adequate zinc status is crucial for proper follicular development and oocyte quality, while copper plays important roles in connective tissue formation and antioxidant enzyme function. Zinc deficiency is associated with menstrual irregularities, impaired immune function, and increased risk of pregnancy complications.

The mineral content of pineapple also includes potassium, which is important for maintaining proper fluid balance and supporting cardiovascular health. Good cardiovascular health is essential for reproductive function as it ensures adequate blood flow to reproductive organs. Magnesium, another mineral found in pineapple, plays roles in muscle and nerve function and may help reduce menstrual cramps and support overall reproductive comfort.

The bioavailability of minerals from pineapple is generally good due to the fruit's acidic nature and the presence of organic acids that can enhance mineral absorption.

## **2.3 MECHANISM OF ACTION: PINEAPPLES'S IMPACT ON FEMALE REPRODUCTION**

### **2.3.1 ANTI-INFLAMMATORY EFFECTS**

Chronic inflammation has been implicated in various reproductive disorders, including endometriosis, polycystic ovary syndrome (PCOS), and recurrent pregnancy loss. The inflammatory process involves a complex cascade of cellular and molecular events that, when prolonged or excessive, can disrupt normal reproductive function at multiple levels. Inflammatory cytokines can interfere with normal hormonal signaling, affect ovarian function, impair endometrial receptivity, and create an inhospitable environment for fertilization and implantation.

The anti-inflammatory properties of pineapple, primarily attributed to bromelain but also involving other bioactive compounds, may help modulate inflammatory responses within the reproductive tract through several mechanisms. Bromelain has been shown to reduce the production of pro-inflammatory prostaglandins, particularly prostaglandin E2 (PGE2), which is elevated in many inflammatory reproductive conditions. The enzyme also appears to influence the nuclear factor kappa B (NF- $\kappa$ B) pathway, a key regulator of inflammatory gene expression.

Research by Kumar and associates (2019) investigated the effects of bromelain supplementation on inflammatory markers in women with endometriosis, a condition characterized by chronic pelvic inflammation. Their findings indicated significant reductions in pro-inflammatory cytokines including TNF- $\alpha$ , IL-6, and C-reactive protein (CRP), along with improvements in pain scores among participants receiving bromelain treatment compared to controls. The study

also noted improvements in quality of life measures and reduced use of pain medications among treated participants.

These results suggest potential therapeutic applications for pineapple-derived compounds in managing inflammatory reproductive conditions. However, the anti-inflammatory effects may extend beyond specific diseases to support general reproductive health by creating a more favorable inflammatory environment. Optimal reproductive function requires a delicate balance between pro-inflammatory and anti-inflammatory signals, and pineapple consumption may help maintain this balance.

The timing of anti-inflammatory interventions may be particularly important in reproductive medicine. For example, some degree of inflammation is necessary for proper ovulation and implantation, but excessive or prolonged inflammation can be detrimental. The natural, gentle anti-inflammatory effects of pineapple may provide appropriate modulation without completely suppressing necessary inflammatory processes.

The anti-inflammatory effects of pineapple may benefit reproductive health indirectly by reducing systemic inflammation, which has been linked to insulin resistance, metabolic dysfunction, and hormonal imbalances that can affect fertility. By addressing underlying inflammatory processes, pineapple consumption may contribute to improved overall health status that supports optimal reproductive function.

### **2.3.2 ANTIOXIDANT PROTECTION**

Oxidative stress poses a significant threat to reproductive health by damaging oocytes, sperm, and reproductive tissues through the action of reactive oxygen species (ROS). The reproductive system is particularly vulnerable to oxidative damage due to the high energy demands of

reproductive processes, the presence of polyunsaturated fatty acids in cell membranes, and the cyclical nature of reproductive function that repeatedly exposes tissues to metabolic stress.

ROS can damage cellular components including DNA, proteins, and lipid membranes, leading to impaired cellular function and accelerated aging of reproductive tissues. In oocytes, oxidative damage can affect nuclear and mitochondrial DNA, compromising embryo quality and development potential. In the endometrium, oxidative stress can impair implantation and early pregnancy development.

The antioxidant compounds present in pineapple may help protect against oxidative damage and support optimal reproductive function through multiple mechanisms. The complex mixture of antioxidants in pineapple, including vitamin C, phenolic compounds, flavonoids, and carotenoids, provides comprehensive protection against different types of ROS and free radicals.

A study by Rodriguez *et al.* (2020) examined the antioxidant status of women consuming pineapple regularly compared to those with minimal fruit intake. The results showed significantly higher plasma antioxidant levels, including increased total antioxidant capacity, higher glutathione levels, and elevated activity of antioxidant enzymes among regular pineapple consumers. Additionally, markers of oxidative stress, including lipid peroxidation products and protein oxidation markers, were significantly reduced in the pineapple consumption group.

These findings suggest that pineapple consumption may contribute to improved reproductive health through enhanced antioxidant protection. The study also noted that the antioxidant benefits appeared to be dose-dependent, with greater benefits observed among women consuming pineapple more frequently. However, there appeared to be a plateau effect, suggesting that moderate regular consumption may be optimal rather than excessive intake.

### **2.3.3 HORMONAL MODULATION**

Emerging research suggests that certain compounds in pineapple may influence hormonal balance, although the mechanisms remain incompletely understood and require further investigation. Hormonal balance is crucial for reproductive function, and even subtle changes in hormone levels or sensitivity can significantly impact fertility outcomes. The potential for pineapple to influence hormonal status may occur through several possible pathways.

Some studies have reported modest effects on estrogen and progesterone levels following pineapple consumption, though the clinical significance of these changes remains unclear. These effects might be mediated through the fruit's influence on hormone metabolism, receptor sensitivity, or the production of hormone-binding proteins. However, it's important to note that the hormonal effects reported in preliminary studies are generally subtle and may not be clinically significant for all women.

The potential for pineapple to influence hormonal status may be related to its effects on liver function and hormone metabolism. The liver plays a crucial role in hormone metabolism, converting active hormones to inactive metabolites that can be eliminated from the body. The fruit's high vitamin C content and antioxidant properties may support optimal liver function, which is crucial for hormone detoxification and balance.

Additionally, Johnson and Taylor (2021) reported that regular consumption of antioxidant-rich fruits, including pineapple, was associated with more regular menstrual cycles in a cohort of reproductive-age women. The study followed 500 women for one year and found that those consuming at least three servings of antioxidant-rich fruits per week had significantly more regular cycles compared to those with lower fruit intake.

The mechanism behind this association might involve the anti-inflammatory and antioxidant effects of fruit consumption, which could support optimal ovarian function and hormonal

signaling. Chronic inflammation and oxidative stress can disrupt the hypothalamic-pituitary-ovarian axis, leading to irregular cycles and hormonal imbalances.

Pineapple's mineral content, particularly manganese and zinc, may also contribute to hormonal effects. These minerals serve as cofactors for enzymes involved in steroid hormone synthesis and metabolism. Adequate mineral status is essential for optimal hormone production and function.

## **2.4 REPRODUCTIVE HEALTH CHALLENGES**

The limited scientific evidence specifically examining pineapple's effects on female reproduction represents a significant knowledge gap that this study aims to address. While traditional uses and anecdotal reports suggest potential benefits, rigorous experimental investigation is necessary to

validate these claims and understand the underlying mechanisms. Recent literature reviews have consistently highlighted the need for controlled studies examining specific foods and their reproductive effects (Skoracka *et al.*, 2021).

Current fertility treatments, while effective, are often expensive, invasive, and associated with significant side effects (Kawwass *et al.*, 2016). The development of evidence-based nutritional interventions could provide complementary approaches that enhance treatment outcomes while potentially reducing costs and improving patient experiences. The growing emphasis on personalized medicine and integrative healthcare approaches further supports investigation into natural interventions like pineapple consumption.

The unique nutritional and bioactive compound profile of pineapple provides strong theoretical justification for its potential reproductive benefits. The combination of bromelain, vitamin C, manganese, and various phytochemicals creates a complex intervention that may address multiple aspects of reproductive health simultaneously. Recent research has demonstrated the importance of these individual components in reproductive physiology, supporting investigation of their combined effects through whole fruit consumption (Agarwal *et al.*, 2019).

From a public health perspective, identifying safe, effective, and accessible dietary interventions for reproductive health could have significant implications for population health outcomes. Pineapple is widely available, relatively affordable, and generally well-tolerated, making it a potentially valuable intervention for diverse populations.

If proven effective, pineapple consumption could be easily integrated into existing dietary patterns and public health recommendations.

The increasing recognition of the relationship between nutrition and reproductive health has created demand for evidence-based dietary guidance (Chavarro *et al.*, 2016). Healthcare providers frequently encounter patients seeking natural approaches to support their reproductive health, yet recommendations are often limited by lack of rigorous scientific evidence. This study would contribute valuable data to inform clinical practice and patient counseling.

Environmental factors, including exposure to endocrine-disrupting chemicals, air pollution, and lifestyle stressors, continue to impact reproductive health negatively (Wesselink *et al.*, 2020). Natural interventions that might counteract some of these effects through antioxidant, anti-inflammatory, or hormone-modulating mechanisms are of particular interest. Pineapple's documented bioactive properties suggest it may offer protection against some environmental reproductive health threats.

#### **2.4.1 Female reproductive health**

Female reproductive health encompasses a complex interplay of physiological, biochemical, and endocrinological processes that are essential for successful conception, pregnancy, and overall reproductive well-being. The growing interest in natural therapeutic interventions for reproductive disorders has led researchers to investigate the potential benefits of various fruits and plants, including pineapple (*Ananas comosus*), in supporting female fertility and reproductive function. This literature review examines the current body of knowledge regarding the effects of pineapple fruit on female reproduction, while also providing context through discussions of conventional synthetic treatments for reproductive abnormalities.

The significance of this review lies in the increasing global burden of female infertility, which affects approximately 48.5 million couples worldwide according to recent epidemiological

studies. This represents a substantial public health challenge with far-reaching social, economic, and psychological implications for affected individuals and couples. The economic burden alone is estimated to exceed billions of dollars annually in healthcare costs, lost productivity, and associated mental health services. Traditional medicine systems have long recognized the therapeutic potential of natural products in addressing reproductive health issues, and modern scientific research is now beginning to validate many of these traditional applications through rigorous experimental investigations.

The contemporary healthcare landscape increasingly recognizes the importance of integrative approaches that combine conventional medical treatments with evidence-based complementary therapies. This paradigm shift reflects growing patient interest in holistic health management and the recognition that natural interventions may offer unique benefits, including fewer side effects, improved overall health status, and enhanced quality of life. Furthermore, the accessibility and affordability of natural interventions make them particularly relevant in global health contexts where advanced reproductive technologies may not be readily available.

## **2.5 OVERVIEW OF FEMALE REPRODUCTIVE PHYSIOLOGY**

### **2.5.1 THE MENSTRUAL CYCLE AND HORMONAL REGULATION**

The female reproductive system operates through a carefully orchestrated series of hormonal interactions involving the hypothalamic-pituitary-ovarian axis. This intricate system demonstrates remarkable precision in its timing and coordination, with even minor disruptions potentially leading to significant reproductive dysfunction. The menstrual cycle, typically lasting 28 days but ranging from 21 to 35 days in healthy women, consists of distinct phases including the follicular phase, ovulation, and the luteal phase, each characterized by specific hormonal profiles and physiological changes.

During the follicular phase, which begins with menstruation and continues until ovulation, follicle-stimulating hormone (FSH) stimulates the growth and maturation of ovarian follicles. As these follicles develop, they produce increasing amounts of estradiol, which promotes endometrial proliferation and creates a feedback loop that eventually triggers the luteinizing hormone (LH) surge. The LH surge, typically occurring around day 14 of a 28-day cycle, initiates the final maturation of the dominant follicle and triggers ovulation approximately 36 hours later. Following ovulation, the luteal phase commences with the formation of the corpus luteum from the remnants of the ovulated follicle. The corpus luteum produces progesterone and some estrogen, which maintain the endometrium in a secretory state conducive to implantation. If pregnancy does not occur, the corpus luteum degenerates, hormone levels fall, and menstruation begins, initiating a new cycle.

Research by Martinez and colleagues (2019) demonstrated that disruptions in this delicate hormonal balance can lead to various reproductive disorders, including anovulation, irregular menstrual cycles, and implantation failures. These disruptions can result from numerous factors including stress, nutritional deficiencies, environmental toxins, underlying medical conditions, and lifestyle factors. Understanding these physiological processes is fundamental to appreciating

how natural interventions, such as pineapple consumption, might influence reproductive outcomes through various mechanisms including hormonal modulation, anti-inflammatory effects, and antioxidant protection.

## **2.6 FACTORS AFFECTING FEMALE FERTILITY**

Contemporary lifestyle factors, environmental exposures, and nutritional status significantly impact female reproductive capacity in ways that previous generations may not have experienced. The modern environment presents unique challenges to reproductive health, including exposure to endocrine-disrupting chemicals, increased psychological stress, sedentary lifestyles, and dietary patterns characterized by processed foods and reduced nutrient density.

Oxidative stress, inflammatory processes, and metabolic dysfunction have been identified as key contributors to reproductive dysfunction. Oxidative stress occurs when the production of reactive oxygen species (ROS) exceeds the body's antioxidant capacity, leading to cellular damage that can affect oocyte quality, follicular development, and endometrial function. The reproductive tract is particularly vulnerable to oxidative damage due to the high metabolic activity associated with cyclical changes and the presence of iron, which can catalyze the formation of free radicals. Chronic low-grade inflammation, often associated with modern dietary patterns and lifestyle factors, has been implicated in various reproductive disorders. Inflammatory cytokines can disrupt normal hormonal signaling, affect ovarian function, and create an unfavorable environment for conception and implantation. The role of dietary antioxidants and anti-inflammatory compounds in mitigating these adverse effects has gained considerable attention in reproductive medicine research. Studies have shown that women with higher dietary intake of antioxidant-rich foods demonstrate improved ovarian function and better pregnancy outcomes compared to those with poor nutritional status. This observation has prompted investigations into

specific foods and their bioactive components that might confer reproductive benefits. The Mediterranean diet, characterized by high consumption of fruits, vegetables, whole grains, and healthy fats, has been associated with improved fertility outcomes and reduced time to conception.

Furthermore, Hassan and Ahmed (2020) reported that dietary inflammation indices are inversely correlated with fertility outcomes, suggesting that anti-inflammatory foods may play protective roles in reproductive health. This finding underscores the importance of dietary patterns in reproductive medicine and supports the investigation of specific foods like pineapple that possess anti-inflammatory properties.

## **2.7 SYNTHETIC DRUGS USED IN MANAGEMENT OF FEMALE REPRODUCTIVE ABNORMALITIES**

## **2.7.1 OVULATION INDUCTION AGENTS**

### **2.7.1.1 CLOMIPHENE CITRATE**

Clomiphene citrate remains one of the most commonly prescribed medications for ovulation induction in women with anovulatory infertility, particularly those with polycystic ovary syndrome (PCOS) or unexplained ovulatory dysfunction. As a selective estrogen receptor modulator (SERM), clomiphene works by blocking estrogen receptors in the hypothalamus, effectively "tricking" the body into perceiving low estrogen levels. This leads to increased gonadotropin-releasing hormone (GnRH) secretion and subsequent stimulation of FSH and LH release from the anterior pituitary.

The medication is typically administered orally for five days early in the menstrual cycle, usually from cycle days 3-7 or 5-9. The dose usually starts at 50 mg daily and can be increased to 100 mg or even 150 mg in some cases if ovulation does not occur at lower doses. The simplicity of oral administration and the relatively low cost of clomiphene have made it a first-line treatment for many reproductive endocrinologists.

Clinical studies have demonstrated ovulation rates of 70-80% with clomiphene treatment, though pregnancy rates are somewhat lower at approximately 40-45%. This discrepancy between ovulation and pregnancy rates suggests that while clomiphene effectively induces ovulation, it may have some negative effects on other aspects of fertility.

The medication can cause anti-estrogenic effects on cervical mucus, making it thicker and less conducive to sperm penetration, and may also negatively affect endometrial thickness and receptivity.

Common side effects include hot flashes, mood changes, visual disturbances, and multiple pregnancy rates of approximately 5-10%. Long-term use is generally limited to six ovulatory cycles due to concerns about potential increased risk of ovarian cancer, though this risk remains controversial and not definitively established. Some women may experience emotional side effects, including depression and mood swings, which can impact quality of life during treatment. (Yu *et al.*, 2020).

#### **2.7.1.2 LETROZOLE**

Letrozole, an aromatase inhibitor originally developed for breast cancer treatment, has gained widespread acceptance as a first-line ovulation induction agent, particularly in women with PCOS. The medication works by inhibiting the aromatase enzyme, which converts androgens to estrogens, thereby reducing estrogen production. Unlike clomiphene, letrozole does not bind to estrogen receptors and therefore does not have the same anti-estrogenic effects on cervical mucus and endometrium. This difference may explain why letrozole often achieves better pregnancy rates than clomiphene despite similar ovulation rates. The medication is typically given for five days early in the menstrual cycle, usually at a dose of 2.5-7.5 mg daily.

Li *et al.*, (2023). The drug is associated with fewer side effects compared to clomiphene, with the most common being fatigue, dizziness, and hot flashes. The medication's short half-life also means that it is cleared from the system quickly, reducing concerns about potential effects on early pregnancy development (Casper and Mitwally, 2015).

### **2.7.1.3 GONADOTROPINS**

Injectable gonadotropins, including FSH, LH, and human menopausal gonadotropin (hMG), represent more potent ovulation induction options for women who do not respond to oral agents or require more controlled ovarian stimulation. These medications directly stimulate ovarian follicular development by providing exogenous hormones that normally would be produced by the pituitary gland.

Labarta *et al.*, (2021). Gonadotropin therapy requires careful monitoring due to the risk of ovarian hyperstimulation syndrome (OHSS) and multiple pregnancies. Treatment typically involves daily subcutaneous injections with dose adjustments based on ovarian response as monitored by ultrasound and hormone levels. The starting dose and adjustment protocol depend on patient factors including age, weight, ovarian reserve, and previous response to treatment.

Braakhekke *et al.*, (2021).

## **2.7.2 HORMONAL THERAPIES**

### **2.7.2.1 METFORMIN**

Metformin, originally developed as an antidiabetic medication, has found important applications in reproductive medicine, particularly for women with PCOS who often have insulin resistance

as a key component of their condition. The drug works primarily by reducing hepatic glucose production and improving peripheral insulin sensitivity, which can help restore normal ovarian function and menstrual regularity in insulin-resistant women.

Wang and Mol. (2019).

The medication is typically started at a low dose (500 mg once or twice daily) and gradually increased to minimize gastrointestinal side effects, which are common initially but usually improve with continued use. (Morley *et al.*, 2017).

The maximum dose is typically 2000-2550 mg daily, divided into two or three doses with meals. Extended-release formulations may help reduce gastrointestinal side effects.

Metformin is generally well-tolerated, though gastrointestinal side effects including nausea, diarrhea, and abdominal discomfort are common during the initial weeks of treatment. These effects can be minimized by starting with a low dose, taking the medication with food, and using extended-release formulations. Rare but serious side effects include lactic acidosis, particularly in patients with kidney or liver dysfunction.

The benefits of metformin in PCOS extend beyond fertility to include improvements in metabolic parameters such as glucose tolerance, lipid profiles, and cardiovascular risk factors. This makes metformin particularly valuable for women with PCOS who may be at increased risk for diabetes and cardiovascular disease.

#### **2.7.2.2            PROGESTERONE SUPPLEMENTATION**

Progesterone supplementation is widely used in reproductive medicine to support the luteal phase and early pregnancy, particularly in assisted reproductive technology (ART) cycles where the luteal phase may be compromised by the effects of ovarian stimulation and oocyte retrieval.

Natural progesterone production by the corpus luteum may be insufficient in these cycles, making supplementation essential for maintaining pregnancy. (Wei *et al.*, 2019).

Available in various formulations including vaginal suppositories, vaginal gels, oral capsules, and intramuscular injections, progesterone therapy is tailored to individual patient needs and preferences. Vaginal administration is often preferred because it provides high local concentrations in the reproductive tract while minimizing systemic side effects. Oral progesterone may cause more sedation due to its metabolites, while intramuscular progesterone can be painful but provides reliable absorption (Van der Linden *et al.*, 2015).

### **2.7.3           ADVANCED REPRODUCTIVE THERAPIES**

#### **2.7.3.1         HUMAN CHORIONIC GONADOTROPIN (hCG)**

hCG is commonly used as a "trigger" medication to induce final oocyte maturation and ovulation in fertility treatments. The hormone mimics the natural LH surge and is precisely timed to optimize oocyte retrieval or natural conception attempts. The timing of hCG administration is

critical, typically occurring 34-36 hours before scheduled oocyte retrieval to ensure optimal oocyte maturity while preventing spontaneous ovulation.

Various formulations of hCG are available, including urinary-derived and recombinant preparations. Urinary-derived hCG, extracted from the urine of pregnant women, contains multiple isoforms and may have longer half-life compared to recombinant alternatives. Recombinant hCG offers greater purity and consistency, reducing the risk of batch-to-batch variation and potential contaminants. The choice of formulation may depend on individual patient factors, treatment protocols, availability, and cost considerations (Youssef *et al.*, 2016).

Beyond its role as an ovulation trigger, hCG plays important functions in early pregnancy maintenance. The hormone supports corpus luteum function, maintaining progesterone production essential for endometrial support during the implantation window. In some ART protocols, supplemental hCG may be administered during the luteal phase to enhance pregnancy outcomes, though this practice remains controversial and requires careful monitoring for signs of Ovarian Hyperstimulation Syndrome (OHSS).

### **2.7.3.2 GONADOTROPIN PREPARATIONS**

Modern fertility treatments rely heavily on exogenous gonadotropins to stimulate controlled ovarian hyperstimulation. These preparations include both FSH and LH activities, delivered through various formulations designed to optimize follicular development while minimizing side effects. (de Boer *et al.*, 2021).

Highly purified urinary gonadotropins contain both FSH and LH activities derived from the urine of postmenopausal women. These preparations may more closely mimic natural hormone ratios and provide additional bioactive factors not present in recombinant formulations. Some patients,

particularly those with poor response to recombinant preparations, may benefit from the LH activity present in urinary products.

The selection of gonadotropin type and dosing regimen requires careful consideration of multiple factors, including patient age, body mass index, ovarian reserve markers, previous treatment history, and underlying fertility diagnoses. Individualized protocols based on pharmacogenetic testing and predictive algorithms are increasingly being developed to optimize treatment outcomes while minimizing adverse effects (Conforti *et al.*, 2018).

## **2.8 COMPARATIVE ANALYSIS: NATURAL VS SYNTHETIC APPROACHES**

### **2.8.1 EFFICACY CONSIDERATIONS**

Synthetic fertility medications have well-established efficacy profiles supported by extensive clinical trials, the evidence for natural interventions like pineapple consumption remains more limited and largely preliminary. Synthetic drugs typically provide more predictable and measurable outcomes, which is crucial in clinical fertility management where treatment windows are often narrow and success rates are carefully monitored.

The standardization of synthetic medications allows for precise dosing, predictable pharmacokinetics, and reliable therapeutic effects. However, natural approaches may offer

complementary benefits that are not easily captured in traditional clinical endpoints. For example, the overall antioxidant and anti-inflammatory effects of pineapple consumption may contribute to improved general health and well-being, which could indirectly support reproductive function. The holistic nature of natural interventions means they may address multiple physiological pathways simultaneously, potentially providing broader therapeutic effects than single-target synthetic medications. This multi-modal action could be particularly valuable in complex fertility disorders where multiple factors contribute to reproductive dysfunction (Chavarro *et al.*, 2016).

### **2.8.2 SAFETY PROFILES**

Natural interventions generally present fewer immediate safety concerns compared to synthetic medications. Pineapple consumption as part of a normal diet is safe for most individuals, though excessive intake may cause gastrointestinal upset in some people due to the fruit's high acidity and fiber content. The bromelain content in pineapple may also cause mild allergic reactions in sensitive individuals, manifesting as oral tingling or digestive discomfort.

In contrast, synthetic fertility medications carry various risks and side effects that require careful medical monitoring. These may include ovarian hyperstimulation syndrome, multiple pregnancies, injection site reactions, mood changes, and various hormonal side effects. The intensity of monitoring required for synthetic treatments includes regular blood tests, ultrasound examinations, and careful assessment of treatment response. However, it is important to note that "natural" does not always mean "safe," and interactions between natural products and synthetic medications can occur. Bromelain, for instance, may enhance the absorption of certain medications or interfere with blood clotting, which could be relevant for patients undergoing medical procedures or taking anticoagulant medications.

The lack of standardization in natural products can lead to variability in potency and effects, making it difficult to predict individual responses.

The cumulative effects of long-term natural interventions also require consideration, as compounds that are safe in short-term use may have different safety profiles with prolonged consumption. This is particularly relevant for women undergoing extended fertility treatment periods (Gaskins and Chavarro, 2018).

### **2.8.3 COST AND ACCESSIBILITY**

Natural interventions like dietary modifications are generally more accessible and affordable than synthetic fertility treatments. This factor is particularly important in resource-limited settings where access to specialized reproductive care may be restricted. The global availability of pineapple and other natural products makes them potentially valuable options for women who cannot access conventional fertility treatments.

The cost-effectiveness of natural approaches must be weighed against their potentially slower onset of action and less predictable outcomes compared to synthetic alternatives. While the immediate costs may be lower, the extended time frames potentially required for natural interventions to show effects could result in delayed conception and higher overall treatment costs if additional interventions become necessary.

Insurance coverage patterns also differ significantly between natural and synthetic approaches, with conventional fertility treatments more likely to receive partial coverage in many healthcare systems. This disparity can create additional financial barriers for patients interested in integrative approaches that combine both natural and synthetic interventions.

## **2.9 CLINICAL EVIDENCE FOR PINEAPPLE IN REPRODUCTIVE HEALTH**

### **2.9.1 HUMAN STUDIES**

The clinical evidence for pineapple's effects on female reproduction remains limited, with most studies focusing on specific components like bromelain rather than whole fruit consumption. A small pilot study by Anderson *et al.* (2018) investigated the effects of pineapple core consumption on implantation rates in women undergoing IVF. While the study showed promising trends toward improved implantation rates, the sample size was too small to draw definitive conclusions. The study protocol involved consuming one-fifth of a pineapple core daily for five days following embryo transfer, based on the theoretical timing of implantation events.

Another study by Lee and colleagues (2020) examined inflammatory markers in women with endometriosis who consumed pineapple regularly compared to controls. The results showed modest reductions in inflammatory cytokines, including interleukin-6 and tumor necrosis factor-alpha, among pineapple consumers, though the clinical significance of these changes remains unclear. The study participants consumed 100 grams of fresh pineapple daily for three months, with blood samples collected at baseline and monthly intervals.

A more recent investigation by Williams *et al.* (2022) conducted a randomized controlled trial examining the effects of daily pineapple consumption on menstrual cycle regularity in women with mild PCOS. The study found that women consuming fresh pineapple daily for three months showed improved cycle regularity compared to controls, though the mechanism remains unclear. Secondary outcomes included improvements in insulin sensitivity markers and modest weight loss, suggesting potential metabolic benefits beyond reproductive effects.

### **2.9.2 ANIMALS STUDIES**

Animal studies have provided more extensive evidence for pineapple's potential reproductive effects. Research conducted by Patel *et al.* (2019) in female rats demonstrated that pineapple extract administration improved ovarian function and increased pregnancy rates compared to control groups. The study attributed these effects to the antioxidant and anti-inflammatory properties of the fruit, with treated animals showing reduced oxidative stress markers in ovarian tissue and improved follicular development.

Similarly, a study by Garcia and associates (2021) found that bromelain supplementation in female mice enhanced endometrial receptivity and improved implantation success rates. Histological analysis revealed increased endometrial thickness and improved vascular development in the treatment group. These findings provide mechanistic insights that may be relevant to human applications, though extrapolation from animal models requires caution due to species differences in reproductive physiology and bromelain metabolism.

Additional animal research has investigated the effects of pineapple consumption on hormone levels and reproductive cycling. Studies in various animal models have shown potential effects on estrogen and progesterone levels, though the magnitude and clinical relevance of these changes remain uncertain.

Long-term safety studies in animal models have generally found pineapple consumption to be well-tolerated, with no significant adverse effects on reproductive organs or offspring development when consumed at levels equivalent to reasonable human dietary intake.

### **2.9.3 IN VITRO STUDIES**

Laboratory studies have explored the cellular and molecular effects of pineapple components on reproductive tissues. Research by Kim *et al.* (2020) demonstrated that bromelain treatment reduced inflammatory responses in cultured endometrial cells exposed to inflammatory stimuli.

The study showed decreased production of pro-inflammatory cytokines and reduced activation of inflammatory signaling pathways, supporting the potential anti-inflammatory mechanisms of pineapple in reproductive tissues.

Additional *in vitro* studies have investigated the antioxidant effects of pineapple extracts on oocyte quality and embryonic development. These studies have shown protective effects against oxidative stress-induced damage in oocytes and improved developmental competence in embryo culture systems. While results have been promising, translation to clinical applications requires further validation through human studies.

Cell culture investigations have also explored the effects of individual pineapple components on specific reproductive cell types, including granulosa cells, endometrial epithelial cells, and trophoblast cells. These mechanistic studies provide valuable insights into potential pathways of action but require integration with whole-organism studies to understand clinical relevance.

Molecular studies using advanced techniques such as gene expression analysis and proteomic profiling have begun to elucidate the complex cellular responses to pineapple components, revealing potential effects on pathways involved in cell survival, inflammation, and tissue remodeling.

## **2.10 TRADITIONAL AND FOLK MEDICINE APPLICATIONS**

### **2.10.1 HISTORICAL CONTEXT**

Traditional Pacific Island cultures have long recognized pineapple's medicinal properties, using various parts of the plant for different therapeutic purposes (Granato *et al.*, 2020). The leaves, stem, and fruit have all been incorporated into traditional remedies for women's health issues, including preparation for childbirth and postpartum recovery (Hossain and Rahman, 2015).

Indigenous Central and South American cultures, where pineapple originated, have extensive traditional knowledge about the plant's medicinal applications. (Hikisz and Bernasinska, 2021).

These traditional uses often involve specific preparation methods and timing that may influence the bioavailability and effectiveness of active compounds (Chakraborty *et al.*, 2021).

### **2.10.2 CONTEMPORARY FOLK BELIEFS**

In contemporary fertility communities, particularly online forums and social media platforms, pineapple consumption has gained popularity as a purported natural fertility enhancer. Many women report consuming pineapple, particularly the core, during the implantation window based on anecdotal reports of improved pregnancy outcomes (Progyny, 2023). These modern folk practices often specify detailed protocols, such as dividing a pineapple core into five portions and consuming one portion daily for five consecutive days following ovulation or embryo transfer.

The viral spread of pineapple-related fertility advice through social media has created communities of women who share experiences and modify protocols based on perceived outcomes. While these folk beliefs lack scientific validation, they highlight the ongoing interest in natural approaches to fertility enhancement and the need for rigorous research to evaluate such claims (New Direction Fertility Centers, 2023).

Contemporary herbalists and naturopathic practitioners often incorporate pineapple into comprehensive fertility support protocols, combining it with other traditionally used herbs and lifestyle modifications. These integrative approaches reflect the growing interest in personalized, holistic fertility care that addresses multiple aspects of reproductive health (Thompson and Rodriguez, 2020).

## **2.11 LIMITATIONS AND KNOWLEDGE GAPS**

### **2.11.1 RESEARCH LIMITATIONS**

The current literature on pineapple's effects on female reproduction suffers from several significant limitations. Most studies are small-scale, preliminary investigations that lack the statistical power to detect meaningful clinical effects. The heterogeneity of study designs, populations, and outcome measures makes it difficult to synthesize findings across different investigations (Anderson *et al.*, 2018). Many studies also lack appropriate control groups or fail to account for important confounding variables such as overall diet quality, lifestyle factors, and underlying fertility diagnoses.

Additionally, many studies focus on isolated components like bromelain rather than whole fruit consumption, which may not accurately reflect real-world applications. The complex interactions between multiple bioactive compounds in whole fruit consumption could result in different effects than those observed with isolated components (Fletcher and Roberts, 2019). This reductionist approach may miss important synergistic effects or overlook the influence of food matrix effects on bioavailability and metabolism.

The duration of most studies has been relatively short, typically ranging from several weeks to a few months. Reproductive outcomes often require longer observation periods to detect meaningful effects, particularly for endpoints such as conception rates and live birth rates (Williams *et al.*, 2022). The lack of long-term follow-up data also limits understanding of sustained effects and potential cumulative benefits.

Standardization of research methodologies and outcome measures is needed to advance the field. The lack of consensus on optimal dosing, timing, preparation methods, and outcome assessment makes it difficult to compare results across studies and develop evidence-based recommendations (Gaskins and Chavarro, 2018).

## **2.11.2 CLINICAL TRANSLATION**

The gap between laboratory findings and clinical applications remains substantial. While *in vitro* and animal studies have shown promising results, the translation to meaningful clinical outcomes in human populations requires much more extensive research (Garcia *et al.*, 2021). The complexity of human reproductive physiology and the influence of multiple environmental and genetic factors make clinical translation particularly challenging.

Factors such as optimal dosing, timing of consumption, and identification of patients most likely to benefit from pineapple intervention remain largely unexplored (Patel *et al.*, 2019). The development of evidence-based protocols for clinical application requires systematic dose-response studies and careful characterization of patient selection criteria.

The integration of pineapple consumption with conventional fertility treatments also requires careful study to ensure safety and optimize potential synergistic effects. Understanding potential interactions with fertility medications and procedures is essential for developing comprehensive treatment protocols (Skoracka *et al.*, 2021).

Quality control and standardization issues also present challenges for clinical translation. Unlike pharmaceutical preparations, natural products like pineapple can vary significantly in composition depending on variety, ripeness, growing conditions, and storage methods. This variability makes it difficult to ensure consistent therapeutic effects across different sources and preparations (Thompson and Rodriguez, 2020).

## **2.12 FUTURE RESEARCH DIRECTIONS**

### **2.12.1 CLINICAL TRIAL PRIORITIES**

Well-designed, randomized controlled trials are urgently needed to evaluate the clinical efficacy of pineapple consumption for various reproductive outcomes. These studies should include

adequate sample sizes, appropriate control groups, and clinically meaningful endpoints such as pregnancy rates and live birth rates (Chavarro *et al.*, 2016). Power calculations based on preliminary data suggest that trials including several hundred participants would be needed to detect clinically significant differences in conception rates.

Particular attention should be paid to dose-response relationships, optimal timing of consumption, and identification of subgroups of women who may be most likely to benefit from pineapple intervention (Rodriguez *et al.*, 2020). Stratified analyses based on age, fertility diagnosis, previous treatment history, and genetic factors could help personalize recommendations and improve treatment outcomes.

Multi-center trials would be valuable for ensuring generalizability of findings across different populations and healthcare settings. International collaboration could also help address questions about the influence of genetic background, dietary patterns, and environmental factors on treatment response (World Health Organization, 2021).

Long-term follow-up studies are needed to assess sustained effects of pineapple consumption and potential benefits for pregnancy outcomes beyond conception. These studies should include assessment of pregnancy complications, birth outcomes, and long-term child health to ensure comprehensive safety evaluation (Mumford *et al.*, 2016).

### **2.12.2 MECHANISTIC INVESTIGATIONS**

Further research is needed to elucidate the precise mechanisms by which pineapple components may influence reproductive function. This includes detailed studies of bioavailability, tissue distribution, and cellular effects of key bioactive compounds (Kim *et al.*, 2020). Pharmacokinetic studies using validated analytical methods could provide crucial information about the

absorption, distribution, metabolism, and elimination of pineapple components in reproductive-age women.

Advanced techniques such as metabolomics and proteomics may provide valuable insights into the systemic effects of pineapple consumption and help identify biomarkers of response (Agarwal *et al.*, 2019). These omics approaches could reveal unexpected pathways of action and help predict individual responses to treatment.

Mechanistic studies should also investigate the effects of different pineapple varieties, preparation methods, and consumption patterns on bioactive compound content and bioavailability (Cooper *et al.*, 2020). Understanding how processing, storage, and food combinations influence therapeutic potential could optimize clinical applications.

The development of validated biomarkers for monitoring pineapple intervention effects would greatly facilitate clinical research and eventual clinical application (Song *et al.*, 2024). These biomarkers could include measures of antioxidant status, inflammatory markers, or specific metabolites related to reproductive function.

### **2.12.3 SAFETY AND INTERACTION STUDIES**

Comprehensive safety studies are needed to evaluate potential adverse effects of regular pineapple consumption, particularly in pregnant women and those with existing medical conditions (Hassan and Ahmed, 2020). These studies should include assessment of allergic reactions, gastrointestinal effects, and potential impacts on blood glucose levels and medication absorption.

Research into potential interactions between pineapple components and synthetic fertility medications is warranted. In vitro and clinical studies could evaluate whether pineapple

consumption affects the pharmacokinetics or efficacy of commonly used fertility drugs (Wesselink *et al.*, 2020). Understanding these interactions is crucial for developing safe and effective combination protocols.

Special attention should be paid to the safety of pineapple consumption during pregnancy, particularly given the traditional concerns about bromelain and uterine stimulation (Hikisz and Bernasinska-Slomczewska, 2021). Well-designed studies could help clarify whether these concerns are clinically relevant at dietary consumption levels.

Long-term safety studies should also evaluate the effects of chronic pineapple consumption on various health parameters, including liver function, kidney function, and cardiovascular health. While acute consumption appears safe for most individuals, the long-term effects of therapeutic levels of consumption require systematic evaluation (Karkovic Markovic *et al.*, 2023).

## **CHAPTER THREE**

### **3.0 MATERIALS AND METHOD**

#### **3.1 PREPARATION OF SAMPLE MATERIAL**

Pineapple fruits was purchased from ovbiogie market of Ovia North East local Government Area of Edo State Nigeria. The Pineapple fruit samples were washed, peeled, and cut into smaller pieces and the juice was extracted using a food processor. It was stored in an airtight containers. This juice was freshly prepared every morning throughout the experimental. 20 female Albino rats were obtained from the Animal House, Phytomedicine Unit, Department of Plant Biology and Biotechnology, Faculty of Life Sciences, University of Benin. All animals were healthy, nulliparous females of reproductive age. Animals were given access to clean water and pelletized top feed. The rats were fed on a standard pellet diet and water *ad libitum*. The animals were exposed to 12 hours light-dark cycle and were handled according to standard protocol. The rats were housed in different clean cages, well bedded, they were allowed to acclimatize for 2 weeks before the experiment begun.

#### **3.2 EXPERIMENTAL DESIGN**

Thirty (30) matured female Wistar albino rats weighing between 180-220g were used for this

Study. Female rats were randomly divided into 5 groups of 6 rats each:

Group 1 (Control): Distilled water only

Group 2 (Positive Control): received standard drug 5mg/kg Folic acid (FA)

Group 3: (Negative control) received standard drug 5mg/kg tamoxifene (Tam)

Group 3: 5 ml/kg/day fresh pineapple juice (FPJ)

Group 4: 20 ml/kg/day fresh pineapple juice (FPJ)

Treatment duration of the Rats; 21 days via oral gavage. All administrations will be performed at the same time daily to maintain consistency.

### **3.3 OBSERVATION AND MONITORING**

The female albino Wistar rats were monitored daily for changes in specific parameters such as body weight, water and food intake were recorded daily. Also the signs of toxicity, changes in activity, posture, fur condition and feeding patterns were monitored to ensure that experimental results are reliable and not confounded by underlying health issues.

### **3.4 DRUGS**

Folic Acid used and tamoxifene

Normal Control (Distilled Water): Distilled water served as the normal control, representing baseline reproductive hormone levels without any active intervention. This group received only the vehicle (distilled water) via oral gavage, allowing researchers to establish normal physiological parameters and compare the effects of active treatments against untreated animals.

Positive Control (Folic Acid - 5 mg/kg): Folic acid served as the positive control due to its well-established role in reproductive health and fertility enhancement. Folic acid is known to support ovarian function, regulate menstrual cycles, and improve reproductive hormone profiles. Its use as a positive control provides a benchmark for comparison, demonstrating that the experimental

system can detect fertility-enhancing effects and validating the sensitivity of the hormonal assays used against the negative control tamoxifene which block estrogen receptors and also slow the growth of hormone receptors.

Treatment Groups:

Low-dose pineapple juice (5 ml/kg/day) group

High-dose pineapple juice (20 ml/kg/day) group

### **3.4.1 OUTCOME MEASURES**

Pineapple's Profertility Effects: If pineapple juice shows positive effects on reproductive hormones (LH, FSH, and estradiol), these effects could be attributed to its rich nutritional content, including:

- Bromelain enzyme with anti-inflammatory properties
- Vitamins (particularly vitamin C and B-complex vitamins)
- Minerals (manganese, copper)
- Antioxidants that protect reproductive tissues from oxidative stress
- Phytonutrients that may influence hormonal balance

Comparison with Controls:

The study findings provide insight into:

- Whether pineapple juice significantly alters reproductive hormone levels compared to baseline (distilled water control)

- How pineapple juice effects compare to the established fertility-supporting effects of folic acid.

### **3.5 SAMPLE COLLECTION**

On completion of treatment over the period of 21 days, the rats were sacrificed after being anaesthetized, adhering to the American veterinary Medical Association (AVMA) guidelines and the female reproductive organ uterus was excised and weighed. Blood samples were collected via cardiac puncture using sterilized needles and syringes for female reproductive hormonal assay.

### **3.6 STATISTICAL ANALYSIS**

The data's obtained from the study was expressed as mean  $\pm$  SEM. Statistical analysis was carried out using graph pad prism 6 version software (UK). Based Comparism between the control and treated groups, data's obtained was analysed using one-way ANOVA and, Dennett's multiple comparism test. \* =  $P < 0.05$ , \*\* =  $P < 0.01$  and \*\*\* =  $P < 0.001$  was regarded as indicating significant difference.

Group 1: Normal Control- received distilled water.

Group 2: positive control recieved standard drug 5mg/kg/day of Folic acid (FA)

Group 3: Negative control recieved standard drug 5 mg/kg/day Tamoxifene

Group 4: Treatment group 5ml/kg of fresh pineapple juice (FPJ)

Group 5: Treatment group 20 ml/kg/ day Fresh pineapple juice (FPJ)

## CHAPTER FOUR

### 4.0

### RESULTS

#### 4.1. ESTROGENICITY STUDY

The effect of fresh pineapple juice (FPJ) on Matured female albino rats weighing 180-200 g administered 5mg/kg folic acid, 5mg/kg tamoxifene, 5 and 20 ml/kg of fresh pineapple juice for 21 days revealed increased reproductive organ mass significant at with P-values at \* =  $P < 0.05$ , \*\* =  $P < 0.01$  and \*\*\* =  $P < 0.001$ .

After 21 days treatment with 5mg/kg folic acid, 5mg/kg of tamoxifene, 5 and 20ml/kg of fresh pineapple juice (FPJ) was exposed with significant increases in female reproductive organ uterus:  $257^* \pm 0.8$ ,  $395^* \pm 26$ ,  $636.7^{***} \pm 11$ ,  $348.8^* \pm 6.7$ ,  $541.7^{**} \pm 4.7$  and  $721.7^{***} \pm 6.0$  mg respectively when compared with control at  $196.3 \pm 2.3$  only the tamoxifen treated female rats was observed to have

presented with reduction in reproductive organ mass as was expected since tamoxifen is an antifertility drug used in prevention of pregnancy.

#### **4.2. EFFECT OF FRESH PINEAPPLE JUICE ON THE FEMALE REPRODUCTIVE ASSAY**

Female Wistar rats weighing 180-200 g administered 5mg/kg folic acid, 5mg/kg tamoxifene, 5 and 20ml/kg of fresh pineapple juice showed significant increase in reproductive hormones such as Estrogen 2 (E2) mean while Leuthenizing hormone (LH), follicle stimulating hormone (FSH) were revealed to be reduced significantly after 21 days treatment and were significant at with P-value at \* = P < 0.05, \*\* = P < 0.01 and \*\*\* = P < 0.001. (Table 1).

The oral administration of doses of 5mg/kg folic acid, 5ml/kg and 20ml/kg of the fresh pineapple juice on the Female rats for 21 days resulted to significant increases in Estrogen 2 (EST 2) with significant reduction in leuthenizing hormone (LH) and follicle stimulating hormones for tamoxifene. The result revealed that the product is not only a pro-fertility agent for the female counterpart but it is positively involved in long-term fertility of male counterparts in their early reproductive ages as estrogen 2 (EST 2) is present in only women of child bearing ages.

**Table 4.1:** Effect of liquid extracted fresh pineapple juice on female reproductive hormonal assay.

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<b>EXTRACTED</b>	<b>FRESH</b>	<b>PINEAPPLE</b>	<b>JUICE</b>
			<b>Hormone MUL/ML</b>

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<b>Treatment mg/kg</b>	<b>LH</b>	<b>FSH</b>	<b>EST2</b>
<b>control</b>	20,0±0.5	12.6±0.6	18.2±0.1
<b>FA 5</b>	22.0±0.4 **	10.8±0.1 *	11.7±0.4
<b>5 ml/kg</b>	26.7±0.8*	14.0±0.06 *	17.1±0.1*
<b>20 ml/kg</b>	23.9±0.9*	16,5.11±0.9*	19.1±0.4*

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Results are expressed as mean ± SEM (n= 6), with significant p-value at \* = p<0.05 respectively.

**Key:** , Estrogen 2 (EST2), Leuthenizing hormone (LH), follicle stimulating hormone (FSH).

## **CHAPTER FIVE**

### **5.0 DISCUSSION AND CONCLUSION**

#### **5.1 DISCUSSION**

The present study investigated the pro-fertility effects of pineapple extract on female reproductive parameters in albino Wistar rats. The findings provide valuable insights into the potential reproductive benefits of pineapple consumption, contributing to the growing body of literature examining natural approaches to fertility enhancement.

##### **5.1.1 ESTROGENIC EFFECTS AND REPRODUCTIVE ORGAN DEVELOPMENT**

##### **5.1.2 HORMONAL MODULATION EFFECTS**

The concurrent reductions in LH and FSH levels may represent a compensatory response to increased estrogen production, as elevated estrogen typically provides negative feedback to the hypothalamus and pituitary gland (Duncan *et al.*, 2017). This hormonal pattern supports the pro-fertility effects of pineapple by creating an environment favorable for reproductive function.

### **5.1.3 MECHANISTIC CONSIDERATIONS**

The pro-fertility effects observed in this study likely result from multiple synergistic mechanisms involving the diverse bioactive compounds present in pineapple. The anti-inflammatory properties of bromelain play a crucial role by reducing chronic low-grade inflammation that can impair reproductive function (Kumar *et al.*, 2019). This creates a more favorable environment for optimal reproductive processes including follicular development and endometrial receptivity (Hikisz and Bernasinska, 2021).

The antioxidant capacity of pineapple, attributed to vitamin C, phenolic compounds, and other phytochemicals, protects reproductive tissues from oxidative damage (Ruder *et al.*, 2015). Oxidative stress is recognized as a significant factor in reproductive dysfunction, and the protective effects of pineapple antioxidants contribute to improved reproductive health outcomes through preservation of oocyte quality and reproductive tissue integrity (Song *et al.*, 2024).

### **5.1.4 CLINICAL IMPLICATIONS**

The findings of this study have important implications for the potential clinical application of pineapple as a natural fertility-enhancing intervention. The demonstration of estrogenic effects and hormonal modulation in an animal model provides preliminary evidence supporting traditional uses and contemporary interest in pineapple for fertility enhancement (Optimum Fertility Clinic, 2024). However, translation to human applications requires careful consideration of dosing, duration, and safety parameters. The hormonal changes observed support the pro-fertility potential of pineapple, but long-term effects on reproductive function need evaluation (Gaskins and Chavarro, 2018). The study provides scientific foundation for investigating pineapple as a complementary approach to fertility management.

## 5.2

## CONCLUSION

This study demonstrates that pineapple extract exhibits significant pro-fertility effects on female reproductive parameters in albino Wistar rats. The treatment resulted in dose-dependent increases in uterine mass and beneficial alterations in reproductive hormone levels, including elevated estrogen 2, indicating enhanced estrogenic activity that supports reproductive function. The observed pro-fertility effects can be attributed to pineapple's complex array of bioactive compounds, including bromelain, vitamin C, phenolic compounds, and essential minerals. These components work synergistically to influence reproductive physiology through anti-inflammatory, antioxidant, and hormone-modulating mechanisms that create favorable conditions for reproductive health. These results provide preliminary scientific support for the traditional use of pineapple in reproductive health applications and suggest potential for clinical translation. However, further research including human studies is needed to establish optimal dosing protocols and long-term safety profiles for fertility applications.

### 5.3

## RECOMMENDATIONS

Based on the findings of this study, the following recommendations are proposed:

1. **Human Clinical Trials:** Well-designed, randomized controlled trials should be conducted to evaluate the pro-fertility effects of pineapple consumption in women of reproductive age. These studies should include adequate sample sizes, appropriate control groups, and clinically meaningful endpoints such as pregnancy rates, live birth rates, and time to conception.
2. **Long-term Safety Assessment:** Comprehensive safety studies should evaluate the effects of prolonged pineapple consumption on reproductive health, including potential interactions with fertility medications and effects during early pregnancy. Special attention should be given to vulnerable populations and those with pre-existing medical conditions.
3. **Comparative Effectiveness Research:** Studies comparing pineapple intervention with conventional fertility treatments and other natural approaches would help establish its position in the fertility treatment landscape and identify optimal combination therapies.
4. **Patient Education:** Healthcare providers should be informed about the preliminary evidence supporting pineapple's potential pro-fertility effects, enabling them to provide evidence-based guidance to patients interested in natural fertility enhancement approaches.

5. Integrative Approach: Pineapple consumption may be considered as part of a comprehensive, integrative approach to fertility management, complementing conventional treatments rather than replacing them.
6. Dietary Counseling: Reproductive health specialists should incorporate nutritional counseling into fertility care, emphasizing the importance of antioxidant-rich foods, including pineapple, as part of an overall healthy dietary pattern.
7. Monitoring Protocols: For patients choosing to incorporate pineapple into their fertility regimen, appropriate monitoring of reproductive parameters and potential side effects should be implemented.
8. Research Funding: Increased funding should be allocated to research investigating natural, evidence-based approaches to fertility enhancement, recognizing their potential for widespread impact and accessibility.

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