ISSN: 2634 - 680X

Journal of Clinical Case Studies Reviews & Reports

SCIENTIFIC Research and Community

Research Article

Open Access

Impact of a Whole Wheat (Triticum asstivium L) Diet on Fetal Parameters in Sprague-Dawley Rats During Pregnancy

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ABSTRACT

Objective: The aim of this study was to determine the effect of wheat-based diet on the fetal parameters of Sprague-Dawley rats.

Materials and Methods: Forty adult female Sprague-Dawley (SD) rats weighing between 150-160 g were used. The estrous cycles of the rats were studied for the first 16 days to establish cyclicity. Regular estrous cycling rats of twenty, were randomly divided into 2 groups (A and B) of 10 rats each. Rats in (group A) were fed with wheat diets and the control (group B) with normal rat chow for 4 weeks, after which the rats were mated with male SD rats of proven fertility on the estrous day (heat period) of estrous cycle. The feeding continues and the rats were sacrificed on the 18th day of gestation by decapitation and fetal parameters were evaluated.

Results: There was a significant increase ($P < 0.05$) in the weight of the wheat diet pregnant rats at the end of the experiment when compared to the control rats. There was no statistically significant different ($P < 0.05$) in the average number of the fetuses between the weight diet group (6) and the control group (7). There was also an increase in the crown-rump length, umbilical cord length, fetal weight and placental weight of the fetus when compared with the control, however, the increase was not statistically significant ($P < 0.05$).

Conclusion: The short time-consummation of wheat-based diet may not have a significant detrimental effect on the progression of pregnancy in rats and no negative effect on fetal outcomes.

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Received: July 16, 2024; **Accepted:** July 22, 2024; **Published:** July 29, 2024

Keywords: Wheat (Triticum asstivium L), Estrous Cycle, Pregnant, Fetus, Rat

Introduction

Triticum asstivium Linn, a popular name for wheat, is a species of flowering monocotyledonous plants in the Poaceae (grass family). This variety, which is hexaploid, is extensively grown all over the world. It is an annual plant that may grow up to 6 feet tall in the early variants, with wheat grains that are roughly 5 to 8 mm long and 2.5 to 4.5 mm wide [1]. There is no denying wheat's economic significance or its role in human and animal diets [2]. Everyday consumption of wheat accounts for 25% of global cereal production, and its rising consumption has been linked to the adoption of a "western lifestyle" in urbanizing and industrializing nations [3]. Because wheat is one of the few crop species that is widely farmed as a staple food source, its nutritional value is therefore quite essential [4]. According to Giraldo et al. [5], it is composed of proteins, carbohydrates, fibers, amino acids, vitamins, and phytochemicals like phenolics and terpenoids. The majority of the world's population primarily obtains their nutrition from wheat since its seed can be processed into flour, semolina, and other basic ingredients used to make bread and other bakery items, as well as pastas. This makes wheat an important food source [6]. It has been suggested that a mother's diet before and throughout pregnancy may have a favorable or negative impact on the developing fetus [7,8]. Thus, maternal diet is crucial for the health of the fetus [9]. Throughout pregnancy, a balanced diet is crucial to maintaining an acceptable nutritional status to satisfy fetal requirements [10]. A higher chance of cardiovascular, metabolic and other non-communicable diseases in adulthood has been associated with inadequate diet during early life [11]. However, there are still a lot of unsolved questions about how consuming wheat affects the development of the fetus. Thus, given that nutrition appears to play a major role in fetal outcome throughout pregnancy; it makes sense to increase our understanding of this topic. Since the wheat diet is extensively consumed by the public. Hence, the purpose of this study is to demonstrate the effects of wheat diet on fetal parameters of pregnant Sprague Dawley rats.

Materials and Methods

Preparation of Study Diet

Triticum aestivum Linn. (Poaceae), commonly known as wheat was purchased from a vendor in Jattu Market, Uzairue, Edo State, Nigeria. The wheat was carefully picked for stones, dried at 40°C in a hot furnace, weighed and were grinded into a coarse powdered form using a local grinding machine. While the regular rat food was provided by Premier Feed Mill Co. Ltd., Ibadan, Nigeria; the ground wheat was sieved with an 80-mesh grid to form the wheat diet. The diet was kept at room temperature for 1 day for drying and was converted to pellet. The diet pellets were stored at - 20 ° C.

Ethical Evaluation

All procedures relating to animal care and use were implemented in strict accordance with the National Institutes of Health Guide for the Care and Use of Laboratory animals and were approved by the Ethics Review Committee of the of Department of Anatomy, Edo State University Uzairue. Edo State. Nigeria [12].

Animal Material

A total of 40 adult female Sprague-Dawley rats, weighing 150g - 160 g were obtained from the Animal House of the Edo State University Uzairue, Edo State. Nigeria.Prior to the start of the experiment, the rats were housed in a cage with free access to rat feed and clean tap water. In order to replicate the typical photoperiodicity of nature in our area, the rats were exposed to around 12 hours of day and 12 hours of night cycle. The ambient temperatures were between 28°C and 31°C, and the room was lighted by sun rays reflecting through the glass windows. After that, the rats underwent a 16-day recruiting phase (a 4-day estrous cycle) in which rats who satisfied our inclusion requirements were chosen. At least three consecutive regular estrous cycles, cyclicity of the vaginal smear, and an apparent satisfactory condition of health are requirements for inclusion [13].

Experimental Procedure

Twenty rats of consecutive regular estrous cycles were grouped into two of 10 rats each. However, group A rats were fed with wheat and Group B rats were given normal rat chow (control) for 4 weeks before mating the rats. After mating the animals; group A continued to be fed with wheat diet, while group B had the normal rat chow until the 18th day of gestation. However, at 7.00 am daily, the diets were presented and rats in each groups received their food in hoppers located inside their cages; and this lasted for 24 hours. The amount of food consumed was measured daily by the difference between the weight of the food placed in the cages and the food remaining 24 hours later, while body weight of individual rats was measured once per week [14].

Estrus Cycle

The cycle phases were determined from the cytology of vaginal smears obtained daily between 8.00 a.m. and 10.00 a.m. Briefly, normal saline was drawn into the tip of the pipette, which was inserted 2 mm deep into the vaginal canal and 2 drops emptied. Next, the mixture of vaginal fluid and regular saline was sucked into the pipette's tip. Before the smear dried, it was placed on a glass slide and inspected under a light microscope. The diestrus phase, the first day of the estrous cycle, was characterized by a low cell density, a preponderance of leukocytes, and a few big nucleated cells. Large nucleated cells containing leukocytes were observed on the second day, which is known as the proestrous phase. The third day, known as the estrous phase, included big squamous cell flakes with tiny pyknotic nuclei. Leukocytes were visible among the remains of big squamous cells with pyknotic nuclei on the metestrous, the fourth day of the estrous cycle [13].

Mating Procedure

On the estrous day (heat period) of the estrous cycle, the rats were mated with male Sprague-Dawley rats that had been shown to be fertile. The first day of gestation (D1) was determined by looking for signs of successful mating, such as copulatory vaginal plug on the cage floor or the presence of sperm cells in a fresh vaginal smear the next morning.

Cesarean Section

On pregnancy day 18 the rats in both groups were sacrificed by decapitation. The rat's abdomen was dissected and the uterus was exposed; allowing for the measurement and recording of the umbilical cord length, placental weight, fetal weight, and crown-rump length in addition to the counting of fetuses.

Statistical Analysis

Data obtained from this study was analysed using GraphPad Prism software, (Version 6.0. San Diego, USA). Values were expressed as Mean ± Standard Deviation (SD) and one way analysis of variance (ANOVA) was done followed by post hoc- Bonferroni test. The confidence limit of statistically significance was set at $P<0.05$.

Results

Body Weight

At the end of the experiment, all the rats used in this study were alive throughout the duration of the experiment. The wheat diet had a positive effect on the body weight during pregnant period and at the end of the experiment, the mean body weight was increase significantly ($P < 0.05$) when compared to the control (Table 1).

Table 1: Effect of wheat-based diet on body weight of pregnant Sprague-Dawley rats

Body weight (g)	Group A	Group B
Before experiment	154.22 + 18.39	159+ 15.21
After experiment	220.59 + 18.74*	209 + 21.64

All values are expressed as Mean ± SD, N= 5. *Significant difference from value of control (p< 0.05)
Group A= wheat diet; Group B= Control

Fetal outcome

The fetuses of wheat-based diet (group A) and control (group B) at pregnancy day 18 were studied with regard to fetal and placental weights. The number of fetuses of the rats in group A were comparable to the control. In group A, there was a significant increase in the crown-rump length, umbilical cord length, fetal weight and placental weight when compared with the control, although the increase was not statistically significant (p< 0.05) (Table 2), (Figure 1).

Table 2: Effect of wheat-based diet on fetal parameters

Parameters	Group A	Group B
Number of fetus	6.10+ 2.33	7.00+ 2.23
Fetal weight (g)	4.15+1.34#	3.85+0.34
Weight of placenta (g	0.60±0.02#	0.50±0.04
Crown-Rump Length (cm)	3.87+0.21#	3.47+0.11
Umbilical cord length (cm)	3.92+1.53#	3.22+0.43

All values are expressed as Mean ± SD, N= 5. #not significant difference from the value of control (p< 0.05)
Group A= wheat diet; Group B= Control

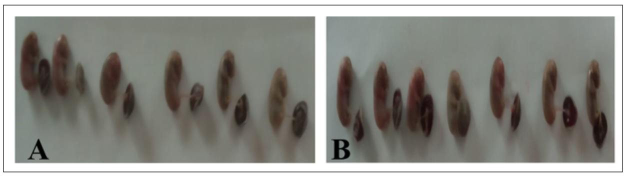

Figure 1: Photograph of wheat-based diet group A and control group B (18th day of gestation).

Discussion

One of the ways of assessing the nutritional status of pregnant women and the welfare of the fetus is through weight gain. When weight gain is excessive or insufficient; it may result in a significant risk of certain diseases in the maternal and fetal body [9]. Hence, to quantify growth; the use of body weight is the most common parameter used [14]. All the groups continued to gain weight throughout the pregnancy period; but the wheat-based diet pregnant rats tend to gain weight more rapidly (Table 1). The calories in wheat may have contributed to the increase in body weight, which is one explanation for the likely weight gain. This result is consistent with a study that suggested wheat may be a significant contributor to obesity [15]. Although gluten is a significant protein found in wheat, anecdotal evidence has led to the theory that gluten contains peptides with opioid-like properties that cause weight gain [15]. According to certain reports, gluten may function as a prebiotic and affect important metabolic and physiological processes [16]. These results, however, are consistent with a recent study that found that rats on a wheat diet gained more body weight [17].

The placenta is responsible for the transfer of materials between fetal and maternal circulation. As a result, the placenta's exchange functions and fetal growth are intimately related. A fetus's birth weight is a direct reflection of the net amount of water and solutes that pass through the placenta during gestation [18,19]. Wheat contains the following nutrients: 12% water, 70% carbohydrates, 12% protein, 2% fat, and 1.8% minerals (calcium, phosphorous, potassium, iron, magnesium, and sodium), thiamine (B1), pantothenic acid, riboflavin (B2), niacin, inositol, folic acid, P-aminobenzoic acid, vitamin B6, vitamin A and trace [20]. There was an increase in placenta weight in the wheat diet group as compared with the control group, however not significantly, suggests that some nutritional components of wheat may have an impact on placenta transport of nutrients, which could potentially explain the study's results. Supporting embryonic growth and the mother's pregnancy adaptations depend on placental growth. Therefore, it is known that aberrant placental size is a good indicator of subpar fetal growth and a poor pregnancy outcome [21].

In line with the findings of our study, which showed an increase in fetal weight and crown-to-rump length (CRL) compared with the control, albeit not significantly, the increased fetal weight consequently caused the mother to gain weight as seen on day 18 of pregnancy. These finding is in consistent with Chahoud and Paumgartten, that reported an increased in maternal weight and crown-to-rump length; which indicates that the fetal development is increased for gestational age [22]. A healthy fetal development depends on the umbilical cord and it is a reliable sign of fetal movement. Fetal deformity, distress, and possibly placental abruption have all been linked to a shorter chord [23]. The increase in fetal weight on GD18 observed in this study may have been caused by a number of factors, including an increase in placenta weight and an increase in umbilical cord length in wheat-based diets, even though the increase is not statistically significant.

Numerous compounds with antioxidant qualities may be found in wheat, and these compounds enhance the redox balance in the ovaries, which is crucial for preserving normal ovarian function [17]. The antioxidants such as phenolic acids, ferulic acid, alkylresorcinols, lignans, lutein, and other nutritional components in wheat may have an impact on reproductive processes and may have helped with the fetal outcome. In the Western diet, phytoestrogens consist mainly of plant lignans, which are a large group of fiber-associated phenolic compounds, widely distributed in wheat [24]. Dietary phytoestrogens are examples of compounds that are found in the amniotic fluid and the umbilical cord blood [25, 26]. Furthermore, it has been suggested that plant lignans and their metabolites offer a number of health advantages [26]. The fact that all of the test rats were pregnant showed that wheat diet had no effect on rat pregnancy. According to (Fig 1), the control group had an average of seven fetuses, whereas the wheat-based diet group had six on average. The findings corroborate a study by Tajudeen et al., which found that wheat had no impact on the quantity of rats born [27]. Yahaya et al. also report that wheat contains tocols that boost vitamin E activity, zinc, magnesium, and selenium—all of which are essential for reproductive functions [28]. Despite the fact that this study was conducted on rats, it can be applicable to humans because the mechanisms involved in pregnancy are relatively comparable. Therefore, the findings of this study have shown that giving pregnant rats a wheat diet for a short period of time has no effect on fetal parameters in rats; this can be investigated further.

Conclusion

The wheat-based diet had no effect on the other pregnancy outcomes, including the number of fetus, fetal weight, placental weight, crown-to-rump length and umbilical cord length. This refers that wheat diet may not have a significant detrimental effect on the progression of pregnancy in rats.

Conflict of Interest

The authors have no interest to declare

Funding

Personal funds were used for this research: No external fund was received.

Contribution by Authors

ICA and MJA conceived and designed the research, interpreted the data, drafted the initial manuscript. MJA collected and assembled data. ICA and MJA both critically revised the article for important intellectual content.

Acknowledgement

The authors are grateful to the Management of the Edo State University Uzairue, Edo State Nigeria for the privilege to carry out this research.

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