International Journal of Research in Pharmacy and Science

Available online at www.ijrpsonline.com Research Article



Effect of regular egg consumption during pregnancy and lactation on morphological characters in swiss albino mice

Sharma R, Jaitawat A, Jain N, Kantwa SM

Environmental and Developmental Toxicology Research Laboratory, Department of Zoology, University College of Science, Mohanlal Sukhadia University, Udaipur, Rajasthan, India

Address for Correspondence Amrita Jaitawat E-mail: amrita.jaitawat@gmail.com

Received: 11-04-2015 Review completed: 25-05-2015 Accepted: 02-06-2015



ABSTRACT

Malnutrition in utero is viewed as causing the fetus to undergo structural and functional changes that result in an increased risk for many diseases and physiological impairment in adulthood. So, fetal growth totally depends on the uptake of nutrients by mother therefore by reducing maternal intake proteins during pregnancy fetal growth can be restricted. Protein intake mainly by pregnant women have role in infant normal body weight. Proteins are available in variety of dietary sources in food of plant and animal origin as well as in highly marketed food. Proteins mainly from animal sources- egg, milk, meat, fish, seafood's, nuts and poultry are highest quality rating. Egg contains vitamin D and choline which can easily cross placenta and transferred to fetus. Previously many studies have already been done on the nutritional importance of egg in adults but information on developmental stages is very scanty. So to fulfill this lacuna the present study is focused on the effects of egg especially during pregnancy and lactation. The egg can easily cross placenta and shows their beneficial effects on the morphological characteristics of the fetus. To assess the activity of egg which is also effective during gestation and lactation because their components can pass through placenta to fetus, all the previous information regarding in this connection are deliberated.

Key words:

rds: Egg, Body weight, Body length, Body mass index, Pregnancy

INTRODUCTION

Pregnancy represents a window of opportunity for health care providers to change lifestyle patterns toward habits that will be healthier for the individual as well as society. Basically it is a period of increased metabolic demands with changes in a women's physiology and the requirements of a growing fetus¹.Malnutrition especially inside uterus causes many changes in adulthood². If you are a normal weight before pregnancy, you need an average of 300 extra calories per day to fuel your baby's growth and keep you healthy during pregnancy. Proteins are building block of the body. They are nitrogen containing substances that are formed by amino-acids. They serve as the major structural components of muscles; they produce hormones, enzymes and hemoglobin. It is well demonstrated that high protein intake have shown a good effect on body size gain and muscle protein synthesis^{3,4}.Protein rich diet increases nitrogen in body tissues, followed by decreases urine pH⁵, suppresses apettite⁶. It was also demonstrated that low protein leads to higher level of reactive oxygen species in

the body because protein act as antioxidant, scavenging of free radicals found in biochemical processes of cells⁷. Insufficient levels of protein directly affect the number of enzymes available for lipid metabolism⁸.But there is one problem that excesses of protein consumption causes many health related issues. Egg a high protein food has a protein efficiency ratio (weight gain of a test subject divided by its intake of a particular protein during the test period) and biological value (it is a measure of the proportion of absorbed protein from a food which becomes incorporated into the proteins of the organism's body)3.9 and 100^9 . They are very good source of vitamins and minerals such as vitamin D and choline. It is well recognized that vitamin D is important for bone health, in modulating immune and neuromuscular function and inflammation¹⁰. Vitamin D is involved in the etiology of various chronic disorders, including type 1 and 2 diabetes, cardiovascular diseases, cancers and multiple sclerosis¹¹. Ronet al.¹²showed the unidirectional transfer of vitamin D across the placenta which means it can cross placenta and transferred to fetus. Eggs are also rich in choline, it is an essential to the integrity and function of cell membranes and it has a role

in normal development of the brain and of memory^{13,14} it is associated with better neurological function and reduced inflammation. Lack of choline may influence breast cancer risk. In control study of more than 3,000 adult women, risks of developing breast cancer were observed to be lowest among those with the highest choline intakes¹⁵. Egg yolks and whole eggs store significant amounts of protein and choline¹⁶.

Protein and BMI

Body mass index or quetelet index is an estimate of body composition that correlates individual weight and height to lean body mass. It is also called as measure for human body shape based on an individual's body mass and height. It is derived from measurement of height and weight while it does not consider body tissue composition. It is figured by dividing weight in kilograms by height in meters squared and multiplied by 100 or by dividing weight in pounds height in inches squared and multiplied by 705. It is the simplest way of determining body mass index.

Reason for Choosing Egg in a high protein diet

Egg is chosen for our experiments because of its good nutritional composition as shown in table no. 1. Both egg yolk and egg white are important for adulthood as has many important properties as shown in table no. 2 and fig. no. 1. Amino acid shows many important functions the functions of amino acids are shown in fig. no. 2.

Table No.	1:	"Nutritional	composition	of 243	gram	egg"
1 4010 1 100		1 (uti itionai	composition		Siam	255

Composition	Form			
· · · · · · · ·				
Calories(1453	From Carbohydrate (32.7 kJ), From Fat			
kJ)	(909 kJ), From Protein (511 kJ)			
Carbohydrates	Dietary Fiber 0.0g, Sugars 1.9g			
(1.9g)				
Protein	Tryptophan (406mg), Threonine			
(30.6g)	(1351mg), Isoleucine (1633mg), Leucine			
	(2644mg), Lysine (2221mg), Methionine			
	(924mg), Cystine (661mg), Phenylalanine			
	(1655mg), Tyrosine (1215mg), Valine			
	(2088mg), Arginine (1995mg), Histidine			
	(751mg), Alanine (1788mg), Aspartic			
	acid (3231mg) ,Glutamic acid (4072mg) ,			
	Glycine (1050mg), Proline (1247mg),			
	Serine (2364mg)			
Fat (24.2g)	Saturated Fat (7.5g), Monounsaturated			
	Fat (9.3g), Polyunsaturated Fat (3.3g),			
	Total Omega-3 fatty acids (180mg), Total			
	Omega-6 fatty acids (2789mg)			
Vitamins	Vitamin A, Vitamin C, Vitamin D,			
	Vitamin E (Alpha Tocopherol), Vitamin			
	K, Thiamin, Riboflavin, Niacin, Vitamin			
	B_6 , Folate, Vitamin B_{12} , Pantothenic			
	Acid, Choline, Betaine			
Minerals	Calcium, Iron, Magnesium, Phosphorus,			
	Potassium, Sodium, Zinc, Copper,			
	Manganese, Selenium, Fluoride			



Fig. No. 1: "Showing composition of egg main components vitamin D and choline"

Table 2: "Nutritional Composition of Egg Yolk and Egg White"

S	Egg yolk	Egg white		
No.				
1.	34% of the liquid	66% of the liquid weight of		
	weight of the egg	the egg		
2.	Egg Vitamins:Vitamin B_2 , Vitamin B_5 ,Vitamin B_1_2 , Vitamin B_5 , Folic acid,ThiamineEgg Minerals:Choline, calcium,copper, iron,manganese,phosphorous,selenium, zinc	Egg Minerals: Niacin, riboflavin, magnesium, potassium, selenium		
3.	Calories: 54 (1 egg yolk)	Calories: 16 (1 egg white)		
4.	All egg fat	None fat of egg		
5.	40% egg protein	60% egg protein		
6.	Color: Yellow: When fed with plenty of xanthophyll containing diet Medium Yellow: When fed with yellow corn or alfalfa Colorless: When fed with colorless diet such as white corn seeds	Color: Usually opalescent White when egg beaten or cooked		



Fig No. 2:"Functions of amino acid a main building block of protein"

MATERIALS AND METHODS

The proposed experiments were conducted in the Environmental and Developmental Toxicology Research Laboratory, Department of Zoology, University College of Science, MohanlalSukhadia University, Udaipur, Rajasthan, India to observe the effect of regular egg consumption during pregnancy and lactation on morphological characters in Swiss albino mice

Animals

Swiss albino mice (Musmusculus) were used for this study. Male and Female mice weighting 28 to 30 grams are kept in the breeding cages in the ratio of 1:4 (1 male & 4 female).Female mice were examined every day in the morning and females showing vaginal plug were isolated and their gestation period were recorded. Presence of spermatozoa in the vagina the following morning was considered day one of gestation. Confirmed pregnant females were housed in polyvinyl chloride cages (270×220×140mm) wrapped with rice husk bedding, and maintained under standard laboratory conditions. The laboratory animals were kept in well ventilated animal room with relative humidity of 70-80%. The room lighting consisted of alternate 12 hours light and dark periods. These females are separated into two groups, these are control and high protein diet (egg) group. The experimental protocols were approved by the Institutional Animal Ethical Committee of the University (No. CS/Res/07/759).

Experimental Design

The selected pregnant females were separated in the following groups (six animals in each group and sub-group).

- **Group 1** Control Diet [receiving (3.85gm Wheat + 3.85gm Maize + 1.55gm Gram + 0.75gm Groundnut) for 1 mice].
- Group 2 Control Diet + High Protein (Eggs)[receiving (2.31gm Wheat + 2.31gm Maize

+ 0.93gm Gram + 0.45grm Groundnut + 4ml Egg) for 1mice]

These groups will be observed at the different development stages such as:

- On PND 1st (At the time of birth) [PND = Post Natal Day]
- On PND 7th (At the end of first week)
- On PND 14th (At the end of second week)
- On PND 21st (At the end of lactation)
- On PND 35th (At the time of puberty)
- On PND 49th (After puberty)

From the observation different Morphological parameters of animal are measured it includes

- 1. Litter number
- 2. Morphological Features
- 3. Bodyweight
- 4. Body length (crown to rump length)
- Body mass index: The body weight and body length was used to determine the body mass index (BMI) as described by ¹⁹
 Body mass index (BMI) = body weight (g)/length² (cm²)

Estimation of Body Weight and Body Length

After fed with different diets, mice were kept separately for 42 consecutive days (21 days of gestation and 21 days of lactation). Body weight and Body length of the control and experimental animals were taken on 1st, 7th, 14th and 21st days after parturition. Body weight was estimated by WJ series electronic balance model No. WJ302A. Body length was estimated by measuring scale. From data of body weight and body length Body Mass index was calculated.

RESULTS

The administration of egg in a diet caused various morphological changes in comparison to control group these are as shown in Fig 3:



Fig No. 3: "Experimental Results of Control and High Protein Diet" [1 day mice of control and high protein diet treated

group of mice (PND 1)(1-2), 7 day mice of control and high protein diet treated group of mice (PND 7)(3-4), 14 day mice of control and high protein diet treated group (PND 14)(5-6), 21 day mice of control and high protein diet treated group (PND 21)(7-8), 35 day mice of control and high protein diet treated group (PND 35)(9-10), 49 day mice of control and high protein diet treated group (PND 49) (11-12). [PND = Post Natal Day].



Graph 1: "Showing the difference in Body Mass Index of Control and High Protein diet fed Groups"

1. Litter number

The litter number of animal is increased in High protein diet treated group in comparison to control. There are 10 pups in control group but in high protein diet treated group the number of pups are 14 (Fig. 2, 1-2). This clearly indicates the increased in litter number due to high protein diet.

2. Morphological Features

There is no major difference in the morphological features of mice in control and high protein diet treated group at all the days on post natal development. Furs are dense in high protein diet treated group in comparison to control. (Fig. 2, 1-12). This is the only morphological difference observed in two groups.

3. Body Weight (BW)

There is a major difference in the body weight of mice in high protein diet treated group in comparison to control group at all the days of post natal development. (Fig. 2, 1-12). The body weight increased in high protein diet fed group especially on 21th, 35th, 49th day means at the end of lactation, at the time of puberty and after puberty.

4. Body Length (BL)

The body length of mice is measured from crown to rump and there is no as such identified difference is found in the body length of animal at all the days of post natal development. (Fig. 2, 1-12). Only slight variations are observed in the body length of mice at all days of post natal development.

5. Body Mass Index (BMI)

The body mass index or BMI is calculated according to¹⁹. The body weight was calculated using electronic weighing machine and body length was calculated using ruler and by

using these body weight and body length body mass index was calculated. There is variation in body mass index of animal in high protein diet treated group in comparison to control. As shown in graph 1. The body mass index increased in high protein diet treated group in comparison to control at all the days of post natal development but much increased was observed at 35^{th} day. This variation in body mass index is due to increased body weight of animal in high protein diet treated group.

DISCUSSION

Next to water protein is the major component of body tissues; it is a building block of the body. Proteins are structural and functional unit of life. They are made up of amino acids; some amino acids are just a few chains long while othersare of several thousand chains. Protein help of person to contributes to a healthy weight. Insufficient protein intake in diet leads to various deformities in our body, especially protein deficiency in utero causes several deformities not only in the pregnant female but also in their pups. There can be many reasons for such happening as we have discussed earlier. Among several sources of protein egg is a very good source of protein. A High protein meal exceeds anabolic vs catabolic capacities in rats adapted to a normal protein diet²⁰. Eggs are good source of vitamin D and choline.Vitamin D is very a delightful solution for health²¹. Choline is an essential nutrient for humans²². Choline is essential nutrient especially for malnourished humans²³. Egg is also a good source of omega- 3- fatty acids and this is very important for pregnancy. It is acritical building blocks of fetal brain and retina, it also play a role in determining the length of gestation and in preventing perinatal depression²⁴. Egg is made up of amino acids and these amino have been considered as an option to prevent or treat intrauterine growth restriction. Accretion of amino acids into egg is an essential component of fetal growth. Therefore supplementation of pregnant female is an attractive therapeutic option to improve fetal growth especially when fetal growth is failing²⁵. Amino acids are supplied from the maternal circulation to the fetus via active transport across the placenta²⁶. Arginine an amino acid has reported to improve fetal weight gain and increased birthweight with its proper supplementation. Fetal growth, as measured by ultrasound, was higher in the arginine group compared to the placebo group. Xiao et al.²⁷ also reported improved mean birth weight with intravenous arginine administration of 20 grams per day for seven days (2.972 vs. 2.794 kilograms). Taurine an another amino acid has many physiological and developmental functions. It is considered an essential amino acid for the fetus and neonate 28,29 . The limitation in supply of amino acid to pregnant female will lead to limit fetal protein accretion and growth³⁰. According to Zeisel¹³ and Zeisel¹⁴ eggs contain choline which results in normal development of brain and memory. Our findings are also in support of this because mice fed with high protein diet are much active in comparison to control group mice. The morphology of animals not altered in animals fed with high protein diet.

Furs are dense in high protein diet fed group in comparison to control. Body mass index is a simple mathematical formula by which height and weight of organism is calculated and is used to measure fatness³¹. It helps us to know who is obese and who is overweight (but not obese).The terms overweight and obese describe ranges of weight that are greater than what is considered healthy for a given height, while underweight describe a weight that is lower than what is considered healthy for a given height. Overweight and obese subjects who consumed an egg breakfast reported significantly greater feelings of satiety through 90 min after the meal and consumed less overall energy than subjects fed an isocaloric bagel breakfast³². Increasing BMI is a sign of healthy organism. In our study BMI was increased in high protein diet fed group in comparison to control group. The increased in BMI was due to gain in body size and muscle synthesis. Our findings are in support of lemon³ in their findings increase in body size was observed. In our results we have also observed that increase in BMI of animal is due to increase in body weight of animal. The body weight of animal was increased in animal fed with high protein diet in comparison to control.

CONCLUTION

Proteins are made up of chains of amino acids. Amino acids have potential growth promoting activity, due to this activity growth is found increased in pups of our experimental group in which mothers are fed with high protein in their diet. This shows that high protein diet significantly increases the body mass index. High protein diet especially egg plays important role during gestation as its components can easily cross the placenta and shows its effects on fetus. Further studies are required to evaluate the effect of egg on various physiological and metabolic activities taking place during pregnancy and lactation.

ACKNOWLEDGEMENTS

Authors are thankful to the Department of zoology, college of science, M. L. S. University for providing necessary facilities such as laboratory facility, Instrumental facility, and chemical facility and by providing animal house within the campus.

REFERENCES

- 1. Pipkin FB. Maternal physiology. In: Dewhurst's Textbook of Obstetrics and Gynaecology, D. K. Edmonds, Ed., Blackwell Publishing, Oxford, UK, 2007.
- 2. Sayer AA, Dunn R, Langley-Evans S, et al. Prenatal exposure to a maternal low protein diet shortens life span in rats. Gerontol.1998; 47(1): 9-14.
- 3. Lemon PWR. Do athletes need more dietary protein and amino acids? Int. J. Sport Nutr. 1995; 5: 39-61.
- 4. Walberg JL, Leidy MK, Sturgill DJ et al. Macronutrient content of hypoenergy diet affects

nitrogen retention and muscle function in weight lifters. Int. J. Sports Med. 1988; 9(4): 261-266.

- 5. Fellstrom B, Danielson BG, Karlstrom B et al. The influence of a high dietary intake of purine rich animal protein on urinary ureate excretion and super saturation in renal stone disease. Clin. Sci. 1983; 64(4): 399-405.
- 6. Johnstone AM. Effect of overfeeding macronutrients on day-to-day food intake in man. Eur. J. Clin. Nutr. 1996; 50(7): 418-430.
- 7. Decker E, Faustman C, Lopez-Botr CJ. "In: Antioxidants in muscle foods, nutritional strategies to improve quality". New York, NY: John Wiley & Sons, Inc. New York, NY, 2000.
- 8. Nielsen GL, NorgardB, Puho E et al. Risk of specific congenital abnormalities in offspring of women with diabetes. Diabetic Med. 2005; 22(6): 693-6.
- 9. Sarwar G. The protein digestibility-corrected amino acid score method overestimates quality of proteins containing antinutritional factors and of poorly digestible proteins supplemented with limiting amino acids in rats. J. Nutr. 1997; 127(5): 758-764.
- 10. NIH (US National Institutes of Health). Dietary supplement fact sheet: Vitamin D, 2009.
- 11. Ruxton CHS, Derbyshire E. Health impacts of vitamin D: are we getting enough? Nutri. Bull. 2009; 341(2): 185-97.
- 12. Ron J, Levitz M, Chuba J et al. Transfer of 25hydroxyvitamin D3 and 1, 25-dihydroxyvitamin D3 across the perfused human placenta. Am. J. Obstet. Gynecol. 1984; 148(4): 370-74.
- Zeisel SH. Choline: Needed for Normal Development of Memory. J. Am. Coll. Nutr. 2000; 19(5): 528-531.
- Zeisel SH. Nutritional importance of choline for brain development. J. Am. Coll. Nutr. 2004; 23(6): 621-626.
- 15. Xu X, Gammon MD, Zeisel SH et al. Choline metabolism and risk of breast cancer in a population-based study. FASEB. 2008; 22(6): 2045-2052
- 16. Howe JC, Williams JR, Holden JM. "USDA Database for the Choline Content of Common Foods". United States Department of Agriculture (USDA), 2004; p. 10.
- 17. Wharton B, Bishop N. Rickets. Lancet. 2003; 362(9393):1389-400.
- 18. Holick MF. Vitamin D: a d- lightful solution for health. J. Invest. Med. 201; 59(6): 872-80.
- 19. Novelli ELB, Diniz YS, Galhardi CM et al. Anthropometrical parameters and markers of obesity in rats. Lab. Anim. 2007; 41: 111–119.
- 20. Morens C, Gaudichoni C, Metges CC et al. A High protein meal exceeds anabolic vs catabolic capacities in rats adapted to a normal protein diet. J Nutr. 2000; 130(9): 2312-21.
- 21. Holick MF. Vitamin D: a d- lightful solution for health. J Investig med. 2011; 59(6): 872-80.
- 22. Zeisel SH, Da Costa KA, Franklin PD et al. Choline, an essential nutrient for humans. FASEB J. 1991; 5(7): 2093-8.

- 23. Chawla RK, Wolf DC, Kutner MH et al. Choline may be an essential nutrient in malnourished patients with cirrhosis. Gastroenterology. 1989; 97(6): 1514-1520.
- 24. Coletta JM, Bell SJ, Roman AS. Omega-3 Fatty Acids and Pregnancy. Rev. Obstet. Gynecol. 2010 Fall; 3(4): 163–171.
- 25. Laura D Brown, Alice S Green, Sean W Limesand, and Paul J Rozance. Maternal amino acid supplementation for intrauterine growth restriction. Front Biosci (Schol Ed). 2011; 3: 428–444.
- 26. Regnault TR, Marconi AM, Smith CH et al. Placental amino acid transport systems and fetal growth restriction--a workshop report. Placenta. 2005; 26(A): 76–80.
- Xiao XM, Li LP. L-Arginine treatment for asymmetric fetal growth restriction. Int. J. Gynaecol. Obstet. 2005; 88(1):15–18.
- 28. Huxtable RJ. Physiological actions of taurine. Physiol. Rev. 1992; 72(1):101–163.
- 29. Boujendar S, Reusens B, Merezak S et al. Taurine supplementation to a low protein diet during foetal and early postnatal life restores a normal proliferation and apoptosis of rat pancreatic islets. Diabetologia. 2002; 45(6): 856–866.
- Rozance PJ, Crispo MM, Barry JS et al. Prolonged maternal amino acid infusion in late-gestation pregnant sheep increases fetal amino acid oxidation. Am. J. Physiol. Endocrinol. Metab. 2009; 297(3): 638–646.
- Zelman KM. "How Accurate Is Body Mass Index, or BMI? Is BMI still the best way to measure fatness? Some experts aren't so sure". 2008.
- 32. Vander Wal JS, Marth JM, Khosla P et al. Short-term effect of eggs on satiety in overweight and obese subjects. J. Am. Coll. Nutr. 2005; 24(6); 510–515.