Effect of L-arginine on Some Anthropometric Parameters of Metabolic Syndrome in Normal Female Wistar Rats

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A reduction in the concentration of nitric oxide, a biosynthetic product of L-arginine (ARG) was associated with the pathophysiology of Metabolic Syndrome (MES). This study assessed the effect of ARG on some anthropometric parameters of MES in normal rats. Female wistar rats (60-80 g) were randomized into two groups (n = 8 animals) and exposed to 60 mg kg⁻¹ (b.wt.) of ARG and 3 mL kg⁻¹ b.wt. of distilled water respectively as treated and control groups. Twenty eight days oral exposure to ARG caused a significant (p<0.01) increase in feed efficiency (4.8±0.06 or 19.26 %) and total water consumption (0.83±0.17 L or 25.75 %), but a significant (p<0.01) decrease in total feed intake (0.31±0.06 kg or 20.51 %), indicating suppressed calorie storage or decreased energy balance that may improve MES. Changes observed in the rats final length (0.30±0.01 m or 3.45 %), total body weight gain (0.05±0.01 kg or 16.66 %), body mass index (1.24±0.15 kg m⁻² or 1.59 %) and lean body weight (0.27±0.03 kg or 0.73 %) though insightful, were not significant (p>0.05), warranting follow up. From the results of Pearson correlations analysis, feed efficiency correlated negatively with total feed intake (r = -0.01) but positively (r = 0.05) with total water consumption, suggesting apparent synergy in the ARG-induced effects. Thus, ARG significantly improved some anthropometric parameters of MES, hence may improve some MES features related to excessive calorie build up or storage in the female rats. The findings warrant similar studies on a longer duration for meticulousness.

Key words: Metabolic syndrome (MES), body mass index (BMI), lean body weight (LBW), feed efficiency (FE), anthropometric parameters

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INTRODUCTION

Metabolic syndrome, MES, is the presence of many health risk factors, including abdominal obesity, insulin resistance, hypertension and atherogenic dyslipidemia, in an individual (Deedwania and Gupta, 2006; Gallagher et al., 2010). The syndrome increases the health risk of the individual. The pattern cuts across age, location and gender since MES is prevalent even in children (Pedrosa et al., 2011) and in the rural areas (Mohan and Deepa, 2006). Apart from the potential to aggravate the poverty-related health burden of the already burdened rural populace, MES could contribute to significant premature mortality (Kozmijlik et al., 2010).

The association of MES incidence with a marked nitric oxide reduction (Garlchкс et al., 2000), informed the speculation that L-arginine (ARG) may improve MES in animals. ARG is a major biosynthetic substrate in NO synthesis (Mooreca et al., 1991), and NO is of biological importance in many disease conditions in animals (Lokhande et al., 2006; McGrowder and Brown, 2007). Several studies suggested that ARG-induced effects in animals may be mediated via NO synthesis (Sepehri et al., 2006; Nemathaksh et al., 2008; Egbutu et al., 2010a, b, c). According to various reports (Subrattty et al., 2007; Harisa, 2011), a reduction in ARG affected the biological activity of NO, suggesting the importance of ARG supply on the effects of NO in animals. The varied metabolic roles of (Van Waardenburg et al., 2007) may improve insulin resistance and NO synthesis (Ezeanyika and Egbutu, 2011), and possibly MES, in animals. Changes in the bio-indicators of MES in ARG-exposed normal rats have been reported (Egbutu and Ezeanyika, 2012a, b; Egbutu et al., 2013).

Anthropometric parameters (parameters that measure bone, muscle, fat and growth to assess risk for high blood pressure, type 2 diabetes and other chronic diseases) may be useful in assessing the presence or otherwise of MES in animals. Such parameters, including body weight gain, water consumption and food intake are significant determinants of diabetes (a major component of MES) in male and female Nile rats (Chaaheh et al., 2010). Other anthropometric parameters of MES include Lean Body Weight (LBW) or muscle mass (Ochhe and Kolhatkar, 2008), Body Mass Index (BMI) (Indhavravdhana et al., 2010), feed efficiency (Fraulob et al., 2010). Furthermore, BMI, an indicator of whether one is obese or not, significantly predicted MES in women (Indhavravdhana et al., 2010). Feed Efficiency (FE) which is the observed total body weight gain divided by total energy (feed) intake (Thim et al., 2006) or the body mass gain expressed in grams per kilocalories consumed (Fraulob et al., 2010) is an indicator of available calorie for storage in animals.

Anthropometric measurements are relatively easy, hence could be handy in assessing MES in animals. Thus, this study aimed to ascertain the effect of ARG on selected anthropometric parameters of MES, using female Wistar rats as model. Rat is a mammal with similar physiology as humans and the choice of female rats derived from recent reports of higher prevalence of MES in females (Kilic et al., 2010; Mangat et al., 2010; Ravikiran et al., 2010).

MATERIALS AND METHODS

Chemicals and reagents: The chemicals used in this study were of analytical grade and were products of reputable companies based in Europe and America.

Concentration determination/justification: The test concentration, ARG (60 mg kg⁻¹ b.wt.) was calculated and adjusted based on the WHO reported daily ARG oral intake (Marsh, 1994) and the concentration used in earlier studies (Alexander et al., 2004; Egbutu et al., 2010a, b, c).

Animals and treatment: Procurement of female Wistar rats used in this study was from the animal house of the Faculty of Biological Sciences University of Nigeria, Nsukka. The rats weighed 60-80 g. The animal study was according to International guidelines for the care and use of laboratory animals in Biomedical Research (CCAC, 1985, WMA/APS, 2002).

The rats acclimatized for a week and were then randomized into two groups (based on their body weight; sample size of eight rats per group). Group B rats were exposed to ARG (60 mg kg⁻¹ b.wt.) whereas Group A rats were exposed to distilled water (DW) (3 mL kg⁻¹ b.wt.). Exposure route was by oral intubation, for 28 days.

The rats, housed in a well-ventilated stainless steel cages at room temperature (28±2°C) and tropical humid condition, were maintained under standard natural photoperiodic condition (twelve hours of light alternating with twelve hours of darkness). In compliance with the ethical guidelines for treating laboratory animals, the rats were allowed unrestricted access to tap water and standard rat chow (Grand Cereals and Oil Mills Limited, Jos, Nigeria) for the experimental period.

Parameters determined

Body weight gain: Body weight of the rats, was determined [in gramme (g) before conversion to kilogram (kg)] on the first day and on the last day. The difference in the body weight of the rats is the total body weight gain.
Feed intake and water consumption: Measurement of the feed intake (and water consumption), were calculated as the total daily feed (and water) consumed relative to body weight change, using the relation:

$$T_i = \frac{T_{f}}{T_{w,0}} \times W_{i,0}$$

where, TF is the Total feed (or total water) consumed by the group, total body weight change of the group individual rat body weight change.

Feed efficiency: Feed Efficiency (FE), the body mass gain in grams per kilocalories consumed, was calculated with the relationship below:

$$\text{Feed efficiency (FE)} = \frac{\text{Weight gained}}{\text{kcal consumed}} \times 100$$

where the caloric content of feed in kcal g$^{-1}$ was based on 3.573 kcal g$^{-1}$ value for standard chow diet (Fraulob et al., 2010).

Length, body mass index (BMI) and Lean body weight (LBW): The length measurement of the rats was in centimeter before conversion to meter. The length was measured from nose slit to tail. Body Mass Index (BMI) computation was with the relationship:

$$\text{BMI} = \frac{\text{Body weight (kg)}}{\text{Height (m)}^2}$$

This study assumed that height of the rats is the same as the length.

Lean body weight was estimated with the gender specific equation already validated by Jurnalmasatian et al. (2005) thus:

$$\text{LBW (female)} = \frac{(9270 - \text{body weight})}{(8780 + 244 + \text{BMI})}$$

where given values are constants for females (Hjelmesaeth et al., 2010). However, this method was applicable in human, hence adjustment was made for size.

Statistical analysis: Analysis of data to determine the significant differences in means was by Student’s t-test, using the Statistical Package for the Social Sciences (SPSS) for Windows version 16.0 (SPSS Inc., Chicago, IL, USA). Results were expressed as mean and standard deviation (Mean±SD) of eight rats per group at significant level of p<0.01. Furthermore, results were correlated for possible association among the studied parameters by Pearson method at correlation coefficient of p = 0.01.

RESULTS

Final length: As presented in Fig. 1, the final length of ARG-fed rats (0.30±0.01 m) non significantly increased (p>0.05) as against the control (0.29±0.01 m). This represents an increase of 3.45% relative to the control.

Total body weight gain (BWG): The results of the present study (Fig. 2) show that ARG treatment in rats caused a non-significant (p>0.05) decrease (0.05±0.01 kg) in total body weight gain (BWG) compared with the DW-treated (control) group (0.06±0.01 kg). This observation represents a decrease of 16.66% relative to control.

Body mass index (BMI): The results of the Body Mass Index (BMI) as presented in Fig. 3 reveal a non significant decrease (p>0.05) in the ARG-treated group (1.24±0.15 kg m$^{-2}$) when compared with the control (1.26±0.08 kg m$^{-2}$). This represents a decrease of 1.59% relative to the control.

Lean body weight (LBW): The results of this study as presented in Fig. 4, indicate that the Lean Body Weight (LBW) in the ARG-treated rats (0.27±0.03 kg) decreased non-significantly (p>0.05) as against the control (0.28±0.02 kg). This represents a decrease of 0.73% in ARG-exposed rats relative to the control.
Total water consumption (TWC): In contrast with the control (0.66±0.09 liters), rats exposed to ARG (Group B) increased (0.83±0.17 liters) in total water consumption level. The observed increase (representing an increase of 25.75%) was statistically significant at 0.05 probability level (Fig. 5).

Total feed intake: Compared with the control (0.39±0.05 kg), the total feed intake in ARG-treated rats decreased (0.31±0.06 kg) significantly (p<0.01). This represents a decrease of 20.51% in ARG-treated group relative to the control (Fig. 6).

Feed efficiency (FE): The feed efficiency significantly increased (p<0.01) in ARG-treated rats (4.83±0.06%) compared with the control (4.05±0.17%). The observation represents an increase of 19.26% in the ARG-treated group relative to the control (Fig. 7).

Correlation: The results of Pearson correlations analysis revealed that feed efficiency correlated negatively with total feed intake (p = 0.01) but positively (p = 0.05) with total water consumption (Table 1).

**DISCUSSION**

Anthropometric parameters (parameters that measure bone, muscle, fat and growth to assess risk for high blood pressure, type 2 diabetes and other chronic diseases) may
be useful in assessing the presence or otherwise of metabolic syndrome (MES) in animals. Generally, these anthropometric parameters of MES tend to indicate the presence or otherwise of obesity that could result from insulin resistance (Lunn and LeRoith, 2007). With insulin resistance, insulin fails to facilitate glucose transport, uptake and utilization by muscles and fat cells. These results to false fed state, increased appetite, increased energy intake and subsequently increased fat deposition. Exposure to L-arginine improved the renal function markers of MES (Egbuonu and Ezeanyika, 2013), glucose metabolism (Egbuonu and Ezeanyika, 2012a) as well as inflammation and liver damage (Egbuonu et al., 2013) of female rats, whereas it worsened their indices of MES related to lipid metabolism (Egbuonu and Ezeanyika, 2012b). However, anthropometric measurements are relatively easy, hence could be handy in assessing MES in animals. Thus, this study aimed to ascertain the effect of ARG on selected anthropometric parameters of MES, using female Wistar rats as model.

Enhanced BWG may result in obesity (Van Herpen and Schrauwen-Hinderling, 2008) which is a major metabolic risk factor (Horwich and Foranow, 2010), via enhanced insulin resistance (Feurer et al., 2009). The results of the present study reveal that exposure to ARG increased the measured final length of the rats but decreased their Total Body Weight Gain (BWG), Body Mass Index (BMI), and Lean Body Weight (LBW) relative to the control. These suggest inhibition in body weight gain of the rats due, probably, to ARG-induced increase in the computed Feed Efficiency (FE) or efficient energy utilization. BWG in animals is dependent on energy balance (He et al., 2008; Van Herpen and Schrauwen-Hinderling, 2008) and may reflect alterations in the regulatory mechanism that affect fat metabolism (Hermanussen and Tresguerres, 2003a, b). In previous study, body weight gain significantly predicted diabetes (a major component of MES) in male and female Nile rats (Chaabo et al., 2010).

Increased final length predicted beneficial influence on MES by way of reduced body mass index (Indhavivadhanal et al., 2010). Obviously (from the BMI relation), an increase in length (taken for height in this study), especially without corresponding increase in body weight (as noted in the ARG-fed group), could lead to a decreased BMI. The observation in this study of decreased BMI and LBW in ARG-fed group agrees with this view. Contrary to this study, ARG increased LBW (or muscle mass) in the elderly, but in combination with essential amino acids (Bosshardt et al., 2008). Consistent with this study, however, ARG reduced body weight in animal model (McKnight et al., 2010). Therefore, it seems plausible that ingestion of ARG in rats probably decreased energy balance (evidenced by decreased feed intake, increased feed efficiency and increased water consumption reported in this study) resulting in the observed BWG reduction. Although, the observed changes in the final length, total body weight gain, body mass index, and lean body weight of the rats were not significant (p>0.05), these results were nonetheless insightful and may be significant with increased duration, hence deserve follow up.

Interestingly, on comparison with control, exposure to ARG decreased total feed intake but increased feed efficiency (FE), and total water consumption of the rats. These observations were significant (p < 0.01) and may be indicating suppressed calorie storage or decreased energy balance (Fraulob et al., 2010) that may improve MES in the rats. Food and water are significant (but opposing) determinants of diabetes, a MES component, in male and female Nile rats (Chaabo et al., 2010). In particular, water is an electronegative enhancer (Batmanghelidj, 2010) hence, the increased water consumption observed in ARG group could, for instance, enhance fat emulsification resulting in decreased fat (calorie) storage and, perhaps, decreased incidence of MES. In addition, it is conceivable that decreased total feed intake as noted in this study may favour decreased energy balance or storable calorie that may lead to obesity—a significant component of MES (Chaabo et al., 2010) probably, by way of suppressed body weight gain. This is consistent with the decreased BWG in ARG group reported in the present study, adduced to possible ARG-induced increase in the computed Feed Efficiency (FE) or efficient energy utilization. The apparent ARG potential for efficient energy utilization together with increased water consumption and reduced feed intake may act in concert to improve MES via suppression of calorie storage and body weight gain.

Pearson’s correlation analysis indicated that feed efficiency correlated negatively with total feed intake (p = 0.01) but positively (p = 0.05) with total water consumption, supporting apparent synergy in the ARG-induced effects in the rats.

CONCLUSION

Thus, ARG significantly improved some anthropometric parameters of MES, hence may improve some MES features related to excessive calorie build up or storage in the female rats. The findings warrant similar studies on a longer duration for meticulousness.
REFERENCES


