CAHD 2020
The 1st International Electronic Conference on Antioxidants in Health and Disease
01–15 DECEMBER 2020 | ONLINE
Chaired by PROF. DR. MIHALIS I. PANAGIOTIDIS
antioxidants
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Biochemical Profile of Albino Rats with Experimentally-Induced Metabolic Syndrome fed Diet Formulations of *Cnidoscolus aconitifolius*, *Gongronema latifolium* and *Moringa oleifera* Leaves

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Abstract

This study evaluated the effect of diet formulations of *Gongronema latifolium* leaf (GLL), *Cnidoscolus aconitifolius* leaf (CAL) and *Moringa oleifera* leaf (MOL) on biochemical parameters of experimentally-induced MS in male albino rats. Adult Wistar rats (forty-eight) 180-210 g were randomly grouped into eight groups of six rats each. Group 1 received normal diet. MS was induced in experimental rats (Groups 2 – 8) using high fat high carbohydrate (HFHC) diet for eight weeks, then group 2 was fed normal rat diet (untreated), while groups 3 to 8 was treated with diets formulated with GLL, CAL, MOL for another eight weeks. The dose of the plants used for feed formulation was 10% of the formulated diet for each treatment. Antioxidant status, liver enzymes, lipid profile and obesity indices were evaluated using standard methods. Superoxide dismutase and catalase activities significantly ($p < 0.05$) increased in the treatment groups. Significant ($p < 0.05$) decrease in total cholesterol, triacylglycerols and body weight gain of the treated groups were observed, high density lipoprotein significantly ($p < 0.05$) increased compared to the untreated group. Results from the study indicate that GLL, CAL, and MOL hav therapeutic potentials that could be useful in the management o metabolic syndrome.

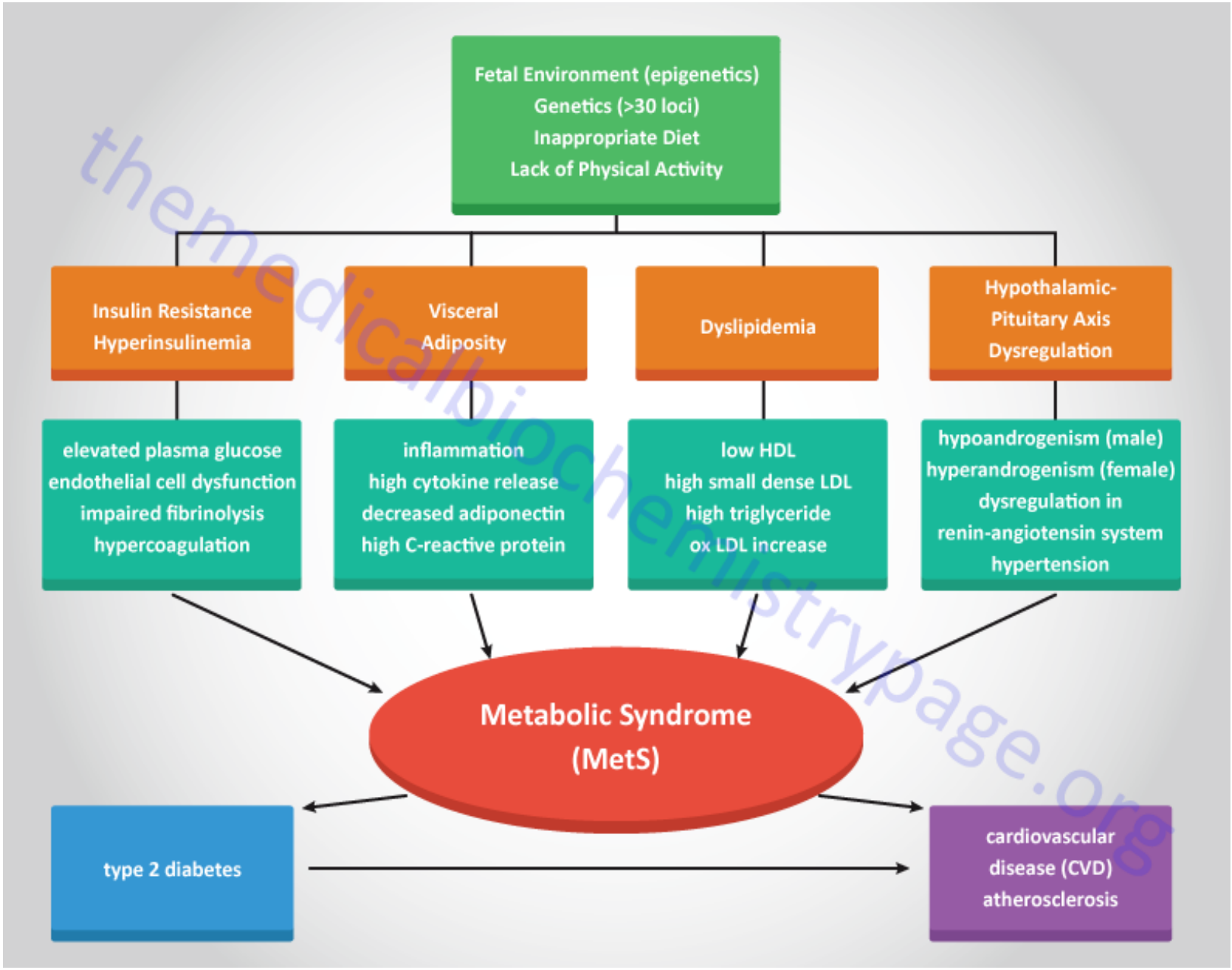
Introduction

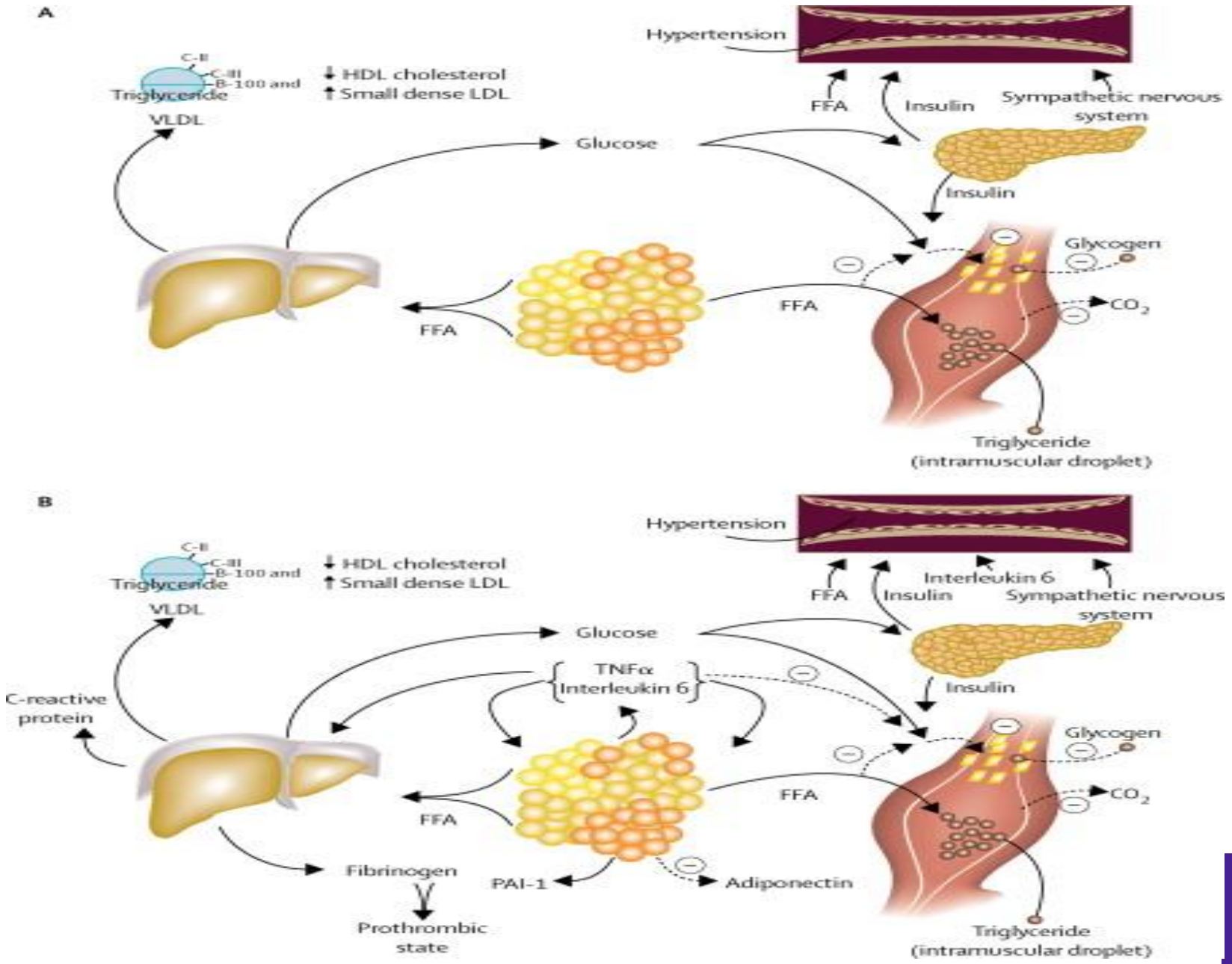
❖Metabolic syndrome (MS) has become a major public health concern worldwide, due to increasing urbanization with its attendant influence on individuals towards surplus energy intake and sedentary lifestyle. It involves multiple metabolic pathways, having insulin resistance and central obesity as its underlying risk factors with other disorders such as: dyslipidaemia, hypertension and microalbuminuria

❖The global prevalence of MS ranges from < 10% to about > 80%, varying based on the region, location (urban or rural area), composition of the population (gender, age, ethnicity, race), and the delineating parameters of the syndrome used in the study

❖Available data show that MS affects 25% of the entire world population of adults.

❖Increased use of herbal medicine has been documented with about 80% of rural dwellers in developing countries solely depending on it for basic health care




A
Hypertension
C-II
C-III
B-100 and
Triglyceride
VLDL
HDL cholesterol
Small dense LDL
FFA
Insulin
Sympathetic nervous system
Glucose
Insulin
Glycogen
FFA
CO_2
FFA
Triglyceride
(intramuscular droplet)
B
Hypertension
C-II
C-III
B-100 and
Triglyceride
VLDL
HDL cholesterol
Small dense LDL
FFA
Insulin
Interleukin 6
Sympathetic nervous system
Glucose
TNFα
Interleukin 6
Insulin
C-reactive protein
Glycogen
FFA
CO_2
FFA
Fibrinogen
PAI-1
Adiponectin
Prothrombic state
Triglyceride
(intramuscular droplet)

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Introduction Contn'd

❖In addition to the presence of biological activity which is exploited in managing several diseases, use of herbs present little or no adverse effects

❖*Gongronema latifolium* leaves (GLL) and *Moringa oleifera* leaves (MOL) possess various nutritional and medicinal values and have been used widely in treatment of disorders associated with MS. The presence of some bioactive components has been reported in these plants which formed the basis for the choice.

❖This study investigated the effect of diet formulations of GLL, CAL and MOL on some biochemical parameters in experimentally-induced metabolic syndrome in male albino rats.

❖Increased use of herbal medicine has been documented with about 80% of rural dwellers in developing countries solely depending on it for basic health care

A

B

C

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Materials and Methods

❖Animals

Forty-eight (48) adult male Wistar rats weighing between 180-210 g were obtained from the animal house of the Faculty of Biological Sciences, University of Nigeria, Nsukka (UNN).

❖Collection, Identification and Processing of Plant Materials

Leaf samples of *Gongronema latifolium, Moringa oleifera* and *Cnidoscolus aconitifolius* were obtained from different locations in Nsukka. The plant materials were air-dried, pulverized and packaged in air-tight-polyethene bags and stored at room temperature prior to use.

❖Induction of Metabolic Syndrome

The rats were fed with high-fat high-carbohydrate (HFHC) diet which is made up of high fat diet and 20% fructose drinking water (FDW) for eight (8) weeks to induce metabolic syndrome

❖Formulation and Administration of Rat Diets

Cnidoscolus aconitifolius leaves (CAL)*, Gongronema latifolium* leaves (GLL) and *Moringa oleifera* leaves (MOL) were used in the formulation of rat diets for treatment. After the establishment of MS, rats were divided into eight groups of six rats each and fed group specific diets for eight (8) weeks as follows:

Group 1: Commercial rat diet and tap water –Normal control

Group 2: MS rats fed commercial rat diet and tap water – Untreated control

Group 3: MS rats fed diet with CAL and tap water

Group 4: MS rats fed diet with GLL and tap water

Group 5: MS rats fed diet with MOL and tap water

Group 6: MS rats fed with combined CAL and GLL (1:1) diet and tap water

Group 7: MS rats fed with combined GLL and MOL (1:1) diet and tap water

Group 8: MS rats fed with combined CAL and MOL (1:1) diet and tap water

❖Oxidative Stress Index and Antioxidant Enzymes Activities

Malondialdehyde (MDA), Superoxide dismutase (SOD) and catalase (CAT) activities were determined using standard methods (Buege and Aust,1978; Misra and Fridovich, 1972; Takahara *et al.* 1960).

❖Liver Function Tests

Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Alkaline phosphatase (ALP), total protein and albumin were determined using the standard methods (Reitman and Frankel 1957, Tietz 1995, Doumas *et al.,* 1997) as outlined in the assay kits leaflets.

❖Determination of Serum Lipid Profile

The serum lipid parameters including total cholesterol (TC), triacylglycerol (TAG) and high-density lipoprotein cholesterol (HDL-C) were determined using standard methods (Trinder, 1969).

❖Measurement of Obesity Indices

Body weight and height were measured and body mass index (BMI) was calculated using standard methods. The rats were anaesthetized using diethyl ether inhalation prior to taking the measurements [14].

Results and Discussion

Table 1: Effect of *C. aconitifolius*, *G. latifolium* and *M. oleifera*-based diets on MDA and antioxidant enzymes of rats with experimentally-induced metabolic syndrome

Group	MDA (U/mg protein)	SOD (IU/mg protein)	CAT (IU/mg protein)
1 Normal	6.43 ± 0.76 [a]	74.51 ± 3.17 [a,b]	0.66 ± 0.10 [a]
2 Untreated	7.15 ± 3.16 [a]	42.85 ± 10.52 [a]	1.38 ± 0.73 [a, b]
3 CAL	6.63 ± 2.47 [a]	56.97 ± 12.75 [a]	2.54 ± 0.06 [c]
4 GLL	5.19 ± 0.81 [a]	54.35 ± 15.58 [a]	1.98 ± 0.56 [b,c]
5 MOL	6.96 ± 4.68 [a]	101.24 ± 13.99 [b]	1.10 ± 0.32 [a, b]
6 CAL+GLL	7.80 ± 0.16 [a]	131.88 ± 36.46 [b]	1.78 ± 0.54 [b]
7 GLL+MOL	10.89 ± 2.73 [a]	76.97 ± 11.29 [a,b]	2.47 ± 0.68 [c]
8 CAL+MOL	11.88 ± 5.05 [a]	131.16 ± 37.19 [b]	2.28 ± 0.64 [b,c]

Table 2: Effect of *C. aconitifolius*, *G. latifolium* and *M. oleifera*-based diets on liver function markers of rats with experimentally-induced metabolic syndrome

Group	AST (U/L)	ALT (U/L)	ALP (U/L)	Total Protein (g/dl)	Albumin (g/dl)
1. Normal	19.89 ± 1.99 [a]	11.53 ±3.15 [a]	7.95 ± 0.98 [b,c]	3.02 ± 0.70 [a,b]	2.57 ± 0.33 [b]
2. Untreated	20.21± 2.00 [a]	8.48 ±0.16 [a]	3.78 ± 0.16 [a]	3.68 ± 0.81 [a,b]	3.02 ± 0.08 [b]
3. CAL	34.53 ± 3.80 [b,c]	20.50 ± 3.08 [a]	5.96 ± 0.59 [a,b]	3.48 ± 0.08 [a,b]	2.49 ± 0.43 [a]
4. GLL	33.73 ± 3.72 [b,c]	10.24 ± 0.64 [a]	6.06 ± 0.43 [a,b]	3.27 ± 0.50 [a,b]	3.07 ± 0.76 [b,c]
5. MOL	37.44 ± 0.36 [c]	14.75 ± 4.22 [a]	8.90 ± 1.98 [c]	4.27 ± 0.75 [a,b]	3.07 ± 0.02 [b,c]
6. CAL+GLL	20.75 ± 2.81 [a]	15.19 ± 4.07 [a]	9.09 ± 0.28 [c]	4.18 ± 0.65 [a,b]	2.33 ± 0.44 [a]
7. GLL+MOL	22.21 ± 3.18 [a]	13.18 ± 4.27 [a]	9.46 ± 0.99 [c]	2.92 ± 0.43 [a,b]	1.90 ± 0.53 [a]
8. CAL+MOL	25.98 ± 0.67 [a,b]	20.16 ± 3.47 [a]	8.61 ± 0.84 [c]	4.85 ± 2.09 [b]	1.35 ± 0.16 [a]

Table 3: Effect of *C. aconitifolius*, *G. latifolium* and *M. oleifera*-based diets on lipid profile of rats with experimentally-induced metabolic syndrome

Group	TC (mg/dl)	TAG (mg/dl)	HDL-C (mg/dl)
1. Normal	133.33 ± 13.33 ^{b,c}	120.27 ± 9.74 ^{d}	90.40 ± 13.80 ^{b}
2. Untreated	144.00 ± 20.13 ^{c,d}	101.73 ± 10.13 ^{c}	90.40 ± 5.2 ^{b}
3. CAL	162.66 ± 14.84 ^{d,e}	102.84 ± 1.01 ^{c}	141.62 ± 21.09 ^{c}
4. GLL	114.66 ± 7.05 ^{a,b}	89.82 ± 10.13 ^{a,b}	102.45 ± 13.13 ^{b}
5. MOL	96.00 ± 4.61 ^{a}	94.45 ± 10.70 ^{a,b}	51.22 ± 3.01 ^{a}
6. CAL+GLL	82.66 ± 5.33 ^{a}	70.62 ± 0.76 ^{a}	102.45 ± 6.02 ^{b}
7. GLL+MOL	136.00 ± 16.65 ^{b,c}	76.57 ± 4.50 ^{a}	117.52 ±13.80 ^{c}
8. CAL+MOL	122.66 ± 11.62 ^{a,b}	95.77 ± 10.73 ^{a,b}	180.80 ± 9.04 ^{c}

Table 4: Effect of *C. aconitifolius*, *G. latifolium* and *M. oleifera*-based diets on body weight of rats with experimentally-induced metabolic syndrome

Group	InitialBody Weight (g)	Final Body Wt (g)	Body Wt Gain (g)
1. Normal Ctrl	244.66 ± 10.50 [a]	301.66 ± 12.58 [a]	57.00 ± 2.64 [a]
2. Untreated Ctrl	282.66 ± 7.02 [b]	381.66 ± 2.88 [c]	99.00 ± 9.53 [b]
3. CAL	290.00 ± 4.00 [c]	350.33 ± 2.51 [b]	60.33 ± 3.78 [a]
4. GLL	276.66 ± 6.65 [b]	334.33 ± 12.09 [b]	57.66 ± 8.62 [a]
5. MOL	274.00 ± 12.16 [b]	334.33 ± 12.09 [b]	60.33 ± 2.51 [a]
6. CAL + GLL	293.33 ± 3.21[c]	356.66 ± 10.40[b]	63.33 ± 8.08 [a]
7. GLL + MOL	283.66 ± 7.23[b]	346.67 ± 5.68[b]	63.00 ± 2.00 [a]
8. CAL + MOL	282.00 ± 6.08[b]	341.67 ± 33.29 [b]	59.66 ± 22.36 [a]

Table 5: Effect of *C. aconitifolius*, *G. latifolium* and *M. oleifera*-based diets on body mass index (BMI) of rats with experimentally-induced metabolic syndrome

Group	Initial BMI (g/cm^2)	Final BMI (g/cm^2)	BMI Gain (g)
1. Normal Ctrl	0.49 ± 0.03 [a]	0.62 ± 0.08 [b]	0.13 ± 0.06 [b]
2. Untreated Ctrl	0.53 ± 0.01 [b]	0.59 ± 0.02[a]	0.06 ± 0.04 [a]
3. CAL	0.52 ± 0.02 [b]	0.56 ± 0.01[a]	0.04 ± 0.02 [a]
4. GLL	0.50 ± 0.00 [b]	0.53 ± 0.02[a]	0.03 ± 0.01 [a]
5. MOL	0.51 ± 0.02 [b]	0.53 ± 0.01[a]	0.02 ± 0.01 [a]
6. CAL + GLL	0.51 ± 0.01 [b]	0.58 ± 0.04[a]	0.07 ± 0.03 [a]
7. GLL + MOL	0.49 ± 0.01[a]	0.54 ± 0.01[a]	0.04 ± 0.02 [a]
8. CAL + MOL	0.51 ± 0.01 [b]	0.56 ± 0.01[a]	0.05 ± 0.01 [a]

Conclusions

The utilization of feed formulations of *Cnidoscolus aconifolius, Gongronema latifolium* and *Moringa oleifera* in the treatment of rats with experimentally induced metabolic syndrome in this study showed reduction in the metabolic and cardiovascular risks in terms of weight reduction, favourable lipid profile as well as increase in the activities of antioxidant enzymes (superoxide dismutase and catalase). Thus, the herbs have some therapeutic effects and could be useful in the management of metabolic syndrome components.

Acknowledgments

The authors want to appreciate all the technical staff and lecturers of the Department of Biochemistry, University of Nigeria, Nsukka, and University of Benin, Benin City, Nigeria who contributed towards the success of this research work

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