

Full Length Research Paper

Effects of diet substitution with defatted kernels of mango (*Mangifera indica*) and wild mango varieties (*Irvingia gabonensis* and *Irvingia wombolu*) on weight and plasma lipid profile of Wistar rats

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Effects of defatted mango (*Mangifera indica*) and wild mango (*Irvingia gabonensis*, *Irvingia wombolu*) kernels in substituted diets of commercial feed, on weight and plasma lipid profile were analysed using Wistar rats after 21-day feeding. Lipid profiling was conducted using enzymatic/colorimetric techniques. The substitution with each kernel type in the commercial feed was 0, 25, 50, 75 and 100%. Diet substitution of up to 75% inversely correlated with weight gained ($P < 0.05$). Up to 50% diet substitution was over 93% acceptable for consumption by the animals. Lipid profile analysis indicated that total cholesterol (TC) and low density lipoprotein (LDL) decreased with increasing diet substitution. The reverse was observed with HDL. TG increased up to 50% defatted *M. indica* kernel (DMIK) substitution and 25% defatted *I. gabonensis* kernel (DIGK)/defatted *I. wombolu* kernel (DIWK) but decreased thereafter. LDL/Cholesterol degradation could have increased TG level and possibly inhibited at higher diet substitutions due to increased residual polyphenolic substances present in the kernel samples. These variables also significantly improved the lipid profile status of the experimental animals ($P < 0.05$).

Key words: Lipid profile, *Mangifera indica*, *Irvingia gabonensis*, *Irvingia wombolu*, cholesterol.

INTRODUCTION

Mango (*Mangifera indica*) is one of the most notable fruits in the sub-tropical and tropical regions of the world (Legesse and Emire, 2012). Mango kernel is a non-conventional source of food which has drawn attention due to its suitability to combat nutritional need of human beings when incorporate into composite flour (Menon et al., 2014). The kernel was reported to be a useful source of protein, carbohydrate and fat except for the presence of anti-nutritional factors such as tannin (Diarra, 2014).

Previous studies successfully showed that mango kernel could be converted to edible state through processing; these studies include the composition, functionality, and toxicology of the kernel before and after processing into flour (Arogba, 1997). In India cultural foodstuff, about 20 to 30% of the kernel flour could be used without adversely affecting acceptability (Legesse and Emire, 2012). The seeds are useful in compounding animal feed (Elgindy, 2017). Furthermore, Arogba (1997, 2002) had

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also shown the potential use of the processed kernel in human diet.

In like manner, wild mango (*Irvingia* species) is of the Irvingiaceae family and exists in two varieties (*gabonensis* and *wombolu*). Both varieties are found naturally in the tropical rainforest of West Africa countries (Kuyooro et al., 2017). *Irvingia gabonensis* was reported to constitute an important part of the natural diet in West Africa for controlling dietary lipids and weight gain. The seed extract of *I. gabonensis* is an effective weight reducing herbal medication, with no known side effects (Etta et al., 2014). Studies on some tropical Africa kernels have found *I. gabonensis* beneficial in lowering undesirable low density lipoprotein (LDL) cholesterol level with athero-protective properties. Hypolipidemic effects in rats have been demonstrated (Kuyooro et al., 2017).

High blood cholesterol associated with elevated levels of oxidised LDL is a risk factor for cardiovascular diseases such as atherosclerosis and myocardial infarction (Omodamiro and Nwankwo, 2013). Hence, the utilization of lipid lowering effect of various plants products is considered to be an important therapeutic approach (Manna and Maiti, 2016). Plasma cholesterol levels apparently decrease with lowering cholesterol content in diet (Ajayi and Ajayi, 2009).

Recently, the effects of undefatted powder and extract of these kernels in several studies with Wistar rats were reported (Alhassan and Arogba, 2018; Egbuonu, 2018; Irondi et al., 2018; Kuyooro et al., 2017; Arogba and Matanmisi, 2014), and an *in-vitro* study (Arogba et al., 2016). In contrast, Abel et al. (2018) described digestibility and nutrient intake of undefatted mango (*M. indica*) kernel in substituted feeds of West African rams. However, scanty literature exist on defatted form of these kernels (Arogba, 2015), and their effects on nutritional status, particularly lipid profile of animals. Therefore, the effect of diet substituted with defatted kernels of mango and wild mango at different percent ratios on weight and lipid profile of Wistar rats is hereby reported.

MATERIALS AND METHODS

Sample collection and processing

Ripe mango (*M. indica*) fruits were plucked directly from trees at Anyigba town, Kogi State. The seeds were dissected using stainless steel knife to obtain the testa and kernel, dried at ambient temperature ($25 \pm 3^\circ\text{C}$) and pulverized using mortar and pestle into powdery form. Dry wild mango (*I. gabonensis* and *Irvingia wombolu*) kernels were procured separately from Anyigba market, Kogi State. The kernels were sorted manually for wholesomeness. Like the mango kernels, they were pulverized into powdery form. The commercial feed used was "Broiler Top Feed Finisher", produced by Premier Feed Mills Company Limited Ibadan, Oyo State, Nigeria.

Fat extraction

Each sample type 10% (w/v) was added to petroleum ether (60 to

80° grade) contained in a beaker, kept at ambient temperature of $30 \pm 2^\circ\text{C}$ and shaken periodically for 24 h. The process was repeated twice and the defatted kernel was oven-dried to constant weight and stored for further analysis. The percentage oil yield was calculated. To assess efficiency of the extraction, residual oil in the defatted samples were determined using Soxhlet extraction technique as described by Williams (2007).

Procurement and management of experimental animals

Thirty nine (39) healthy, adult male albino Wistar rats (weighed 122 - 136 g) were procured from the animal house of the Department of Veterinary Medicine, Benue State University, Makurdi, Nigeria. They were acclimatized in clean rat cages at the Experimental Animal House of the Department of Biochemistry, Kogi State University, Anyigba for a period of ten (10) days at ambient temperature with 12-h light and dark cycle. Within the period, they were fed with a commercial "Broiler Top Feed Finisher" with water *ad-libitum*.

Experimental design

Weight measurement

The weights of the rats were taken before and after twenty one (21) days feeding period. The percentage weight difference was calculated.

Diet substitution/animal

Thirty-nine (39) rats were fed with commercial "Top Feed Finisher" diet and substituted with defatted *M. indica* kernel (DMIK), defatted *I. gabonensis* kernel (DIGK) and defatted *I. wombolu* kernel (DIWK) each, at 0, 25, 50, 75 and 100% levels. Water was provided *ad-libitum* grouping (Table 1).

Estimation of diet consumed

Weights of the diets provided were taken and those of the remnants after 21 days feeding. Percentage diet consumed, %DC = (Weight of diet consumed/Weight of substituted diet provided) \times 100.

Blood sample collection

On the 22nd day, the animals were sacrificed by anaesthesia using chloroform in desiccator. The blood was obtained through cardiac puncture into heparinized (EDTA) bottles using 5 ml syringe.

Assay of plasma lipid profile

The plasma lipid profile [Total Cholesterol, Triglycerides and High Density Lipoprotein Cholesterol] was determined by spectrophotometric technique, using enzymatic/colorimetric assay kits (Randox Laboratories, United Kingdom). However, by calculation $\text{LDL} = \text{TC} - [\text{HDL} + \text{TG}/5]$.

Statistical analysis

The statistical analysis was conducted using SEM software available at miniwebtool.com. Results were expressed as mean \pm standard error of mean (SEM). Separation of mean was conducted

Table 1. Experimental design for albino rats on substituted diets.

| Sample: Comm. Feed (%) | Number of rats per sample type | | |
|------------------------|--------------------------------|------|------|
| | DMIK | DIGK | DIWK |
| 0:100 | (3)* | - | - |
| 25:75 | 3 | 3 | 3 |
| 50:50 | 3 | 3 | 3 |
| 75:25 | 3 | 3 | 3 |
| 100:0 | 3 | 3 | 3 |
| Total rats/sample type | 15 | 12 | 12 |

DMIK = Defatted *Mangifera indica* kernel, DIGK = defatted *Irvingia gabonensis* kernel, DIWK = defatted *Irvingia wombolu* kernel and (3)* implies the three animals in (DMIK) group also served as control for (DIGK) and (DIWK) groups.

Table 2. Mean weight of animals before and after feeding for 21 days (n = 3).

| Sample type | Sample: Comm. feed (%) | Before (g) | After (g) | Difference (g) | Difference (%) | SEM |
|-------------|------------------------|------------|-----------|----------------|-----------------|-----|
| DMIK | 0:100 | 124 | 178 | 54 | 44 ^e | 8.8 |
| | 25:75 | 132 | 163 | 31 | 24 ^d | |
| | 50:50 | 127 | 145 | 18 | 14 ^c | |
| | 75:25 | 129 | 133 | 4 | 3 ^b | |
| | 100:0 | 136 | 127 | -9 | -7 ^a | |
| DIGK | 0:100 | 124 | 178 | 54 | 44 ^d | 8.6 |
| | 25:75 | 125 | 147 | 22 | 18 ^c | |
| | 50:50 | 131 | 139 | 8 | 6 ^b | |
| | 75:25 | 128 | 129 | 1 | 1 ^{ab} | |
| | 100:0 | 134 | 128 | -6 | -4 ^a | |
| DIWK | 0:100 | 124 | 178 | 54 | 44 ^d | 8.3 |
| | 25:75 | 127 | 156 | 29 | 23 ^c | |
| | 50:50 | 125 | 137 | 12 | 10 ^b | |
| | 75:25 | 122 | 127 | 5 | 4 ^{ab} | |
| | 100:0 | 133 | 129 | -4 | -3 ^a | |

Values with the same superscripts on the same column are not significantly different at $p > 0.05$. DMIK = Defatted *Mangifera indica* kernel, DIGK = defatted *Irvingia gabonensis* kernel, DIWK = defatted *Irvingia wombolu* kernel, SEM = Standard error of mean, Comm. Feed = commercial feed (Broiler Top feed Finisher).

for test of significance at $P = 0.05$.

RESULTS AND DISCUSSION

On defatting, the oil yield of 21.9% from *M. indica* kernel in this study was similar to that reported by Arogba (2015) as the variety was obtained from the same locality. The oil yields from *I. gabonensis* and *I. wombolu* kernels were also similar ($67\% \pm 1.0$) and agreed with the report of Bamidele et al. (2015). Estimated by Soxhlet extraction technique, residual oil in these kernels was between 1 and 3%.

Weight difference and diet consumption by experimental animals

The percentage weight difference of the experimental animals in Table 2 showed that the substituted *Irvingia* kernel samples (DIGK and DIWK) had similar effects on weight gain ($P < 0.05$). Similar to *M. indica* kernel (DMIK) sample, there was positive but decrease in weight gained as sample substitution in diet increased up to 75% ($P < 0.05$). However, 100% substitution had negative effect on weight gain. The "bitter principle" in these higher substitutions possibly had adverse effect on diet consumption. Arogba (1997, 2000) had identified and

Table 3. Estimated diet consumed by the animals within 21 days.

| Sample:Comm. Feed (%) | DMIK (%) | DIGK (%) | DIWK (%) |
|-----------------------|------------------|------------------|------------------|
| 0:100 | 100 ^c | 100 ^c | 100 ^c |
| 25:75 | 98 ^c | 99 ^{bc} | 99 ^c |
| 50:50 | 93 ^c | 98 ^b | 99 ^c |
| 75:25 | 75 ^b | 98 ^b | 97 ^b |
| 100:0 | 53 ^a | 90 ^a | 94 ^a |
| SEM | 8.9 | 1.8 | 1.1 |

Values with the same superscripts on the same column are not significantly different at $p > 0.05$. DMIK = Defatted *Mangifera indica* kernel, DIGK = defatted *Irvingia gabonensis* kernel, DIWK = defatted *Irvingia wombolu* kernel, SEM = standard error of mean, Comm. Feed = commercial feed (Broiler Top feed finisher).

Table 4. Effect of diet substitution with DMIK on plasma lipid profile after 21 days.

| Sample:Comm. Feed (%) | TC (mg/dL) | TG (mg/dL) | HDL(mg/dL) | LDL (mg/dL) |
|-----------------------|----------------------------|----------------------------|---------------------------|----------------------------|
| 0:100 | 131.60 ± 2.52 ^c | 219.49 ± 2.71 ^c | 32.14 ± 2.05 ^a | 55.21 ± 1.77 ^d |
| 25:75 | 114.41 ± 2.59 ^b | 223.66 ± 6.93 ^c | 37.83 ± 1.82 ^b | 33.75 ± 1.67 ^c |
| 50:50 | 109.40 ± 4.35 ^b | 228.52 ± 5.65 ^c | 39.14 ± 2.34 ^b | 24.53 ± 2.23 ^b |
| 75:25 | 96.44 ± 2.02 ^a | 147.60 ± 3.97 ^b | 47.55 ± 1.92 ^c | 20.78 ± 0.96 ^{ab} |
| 100:0 | 90.40 ± 1.88 ^a | 122.42 ± 2.90 ^a | 49.08 ± 1.71 ^c | 16.81 ± 0.76 ^a |
| SEM | 7.22 | 22.18 | 3.16 | 6.85 |

Values are expressed as mean ± SEM (n=3). Values with the same superscripts on the same column are not significantly different at $p > 0.05$. DMIK = Defatted *Mangifera indica* kernel, TC = total cholesterol, TG = triglyceride, HDL = high density lipoprotein, LDL = low density lipoprotein, SEM = standard error of mean, Comm. Feed = commercial feed (Top feed finisher).

analysed the tannin constituents of *M. indica* kernel.

Table 3 further supported the aforementioned observation. 100% test samples were least acceptable for consumption by the animals followed by 75% substituted samples, while diets substituted by 50% or less were favourably consumed by over 93%. It further inferred that the defatted kernels probably contained significant proportion of water-soluble than fat-soluble polyphenolic components.

Plasma lipid profile assessment

The plasma lipid profile of the animals after 21 days of feeding (Tables 4 to 6) showed that total cholesterol (TC) and LDL decreased with increasing defatted test sample substitution ($P < 0.05$). For instance, the commercial feed had the highest TC level. Substitution by 25 and 50% kernel samples caused average reduction by 10 and 15%, respectively, indicating the significant physical effect of kernel substitution. The composition of the commercial feed, therefore, appeared to promote cholesterol synthesis. In similar manner, LDL proportionally decreased. However, HDL variation was observed to be inversely correlated with TC and LDL.

The results on TG variation raised some curious

attention, while TC and LDL decreased with increasing physical substitution of the kernel samples in the diets, TG levels increased in diets with up 50% DMIK and 25% DIGK or DIWK substitution. The TG levels decreased significantly there-after ($P < 0.05$). Two views were proposed to possibly explain these observations:

Since LDL comprises apolipoprotein B, cholesterol, phospholipids, and triglyceride in varied concentrations (Nelson and Cox, 2005; Prass, 2011), hydrolysis of its components could elevate the HDL and TG levels at those levels of diet substitution mentioned. Furthermore, since cholesterol serves as a starting molecule for steroid hormone synthesis, the side chain when cleaved to give 4-methylpentanal, or 4-methyl-4-hydroxypentanal, could be oxidised for glyceride synthesis. On the contrary, at higher levels of kernel substitution in the diets, the corresponding increased levels of residual (water-soluble) polyphenolic substances (e.g. tannins) could have significantly and adversely inhibited the TC/LDL hydrolysis.

100% test samples gave between 100 and 124 µg/dl TG, classified as “desirable” nutritionally in contrast to those substituted with commercial feed which gave a range of 148 to 256 µg/dl. A range of 150 to 500 was described as “borderline high” (Ma and Shieh, 2006). The results of TC and LDL-cholesterol in this study agreed

Table 5. Effect of diet substitution with DIGK on plasma lipid profile after 21 days.

| Sample: Comm. Feed (%) | TC (mg/dL) | TG (mg/dL) | HDL (mg/dL) | LDL (mg/dL) |
|------------------------|----------------------------|----------------------------|---------------------------|---------------------------|
| 0:100 | 131.60 ± 2.52 ^d | 219.39 ± 2.71 ^c | 32.14 ± 2.05 ^a | 55.20 ± 1.77 ^c |
| 25:75 | 118.82 ± 3.93 ^c | 255.61 ± 4.90 ^d | 36.51 ± 2.11 ^b | 31.16 ± 2.21 ^b |
| 50:50 | 102.66 ± 3.11 ^b | 173.13 ± 3.30 ^b | 44.93 ± 1.51 ^c | 23.10 ± 2.89 ^a |
| 75:25 | 104.96 ± 1.11 ^b | 187.89 ± 2.71 ^b | 46.13 ± 0.67 ^c | 21.25 ± 1.04 ^a |
| 100:0 | 94.48 ± 3.10 ^a | 100.54 ± 2.11 ^a | 54.77 ± 1.62 ^d | 19.35 ± 1.31 ^a |
| SEM | 6.57 | 25.91 | 3.95 | 6.61 |

Values are expressed as mean ± SEM (n=3). Values with the same superscripts on the same column are not significantly different at $p > 0.05$. DIGK = Defatted *Irvingia gabonensis* kernel, TC = total cholesterol, TG = triglyceride, HDL = high density lipoprotein, LDL = low density lipoprotein, SEM = standard error of mean, Comm. Feed = commercial feed (Top feed finisher).

Table 6. Effect of diet substitution with DIWK on plasma lipid profile after 21 days.

| Sample: Comm. Feed (%) | TC (mg/dL) | TG (mg/dL) | HDL (mg/dL) | LDL (mg/dL) |
|------------------------|-----------------------------|----------------------------|----------------------------|----------------------------|
| 0:100 | 131.60 ± 2.52 ^d | 219.49 ± 2.71 ^c | 32.14 ± 2.05 ^a | 55.21 ± 1.77 ^d |
| 25:75 | 121.30 ± 5.08 ^c | 249.01 ± 3.84 ^d | 35.42 ± 1.87 ^{ab} | 33.45 ± 4.10 ^c |
| 50:50 | 106.21 ± 2.58 ^b | 211.85 ± 3.87 ^c | 36.95 ± 2.44 ^b | 26.55 ± 2.09 ^b |
| 75:25 | 102.12 ± 4.63 ^{ab} | 159.24 ± 3.22 ^b | 42.85 ± 2.34 ^c | 27.57 ± 2.67 ^{bc} |
| 100:0 | 95.73 ± 3.10 ^a | 123.46 ± 1.38 ^a | 51.29 ± 2.63 ^d | 19.57 ± 1.90 ^a |
| SEM | 6.58 | 22.55 | 3.37 | 6.10 |

Values are expressed as mean ± SEM (n=3). Values with the same superscripts on the same column are not significantly different at $p > 0.05$. D DIWK = Defatted *Irvingia wombolu* kernel, TC = total cholesterol, TG = triglyceride, HDL = high density lipoprotein, LDL = low density lipoprotein, SEM = standard error of mean, Comm. Feed = commercial feed (Top feed finisher).

with the report of Kuyooro et al. (2017) on hypolipidemic effects of undefatted kernel of *I. gabonensis*. The decrease in TC and LDL cholesterol recorded in this study shows that the test samples could have athero-protective potential and their consumption could avert the onset of developing atherosclerosis and cardiovascular disease.

Conclusion

The study has shown that substitution of defatted kernels of *M. indica* and *Irvingia* species in animal diets were optimally palatable at 50%, as it was also concomitant with weight gain. Furthermore, TC and LDL decreased with increasing defatted test sample substitution. The levels of water-soluble polyphenolic substances in the defatted samples of the diets were implicated in these observations. These observations revealed that the test samples possessed hypolipidemic effects and could be utilized in the prevention of atherosclerosis and cardiovascular disease when incorporated in animal diet.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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