The Ameliorative Efficacy of Roselle (Hibiscus sabdariffa), Moringa (Moringa oleifera), Ginger (Zingiber officinale) and 'Ugwu' (Telfairia occidentalis) on some Physical characteristics of Albino rats (Rattus norvegicus) Exposed to Cement dust

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ABSTRACT: The ameliorative efficacy of roselle, moringa, ginger, 'ugwu' and a mixture of the plant extracts on selected physical characteristics of rat exposed to cement dust was evaluated. Albino rats grouped into six of 18 rats each were exposed to cement dust at a cement factory in Sagamu, Ogun State, Nigeria. The test rats were administered 400 mg kg⁻¹ ethanol extracts of roselle, moringa, ginger, 'ugwu' and a mixture of the plant extracts, respectively for 180 days. The control rats were fed with only distilled water. Significant (p<0.05) differences were observed between the test and control rats in all the tested health indices. The test rats weighed more than the control rats, and weight differences were also observed among the test rats. The birth, number of newborn per litter and offspring survival rates of the test rats were higher than the control rats. Lower death rates were observed in the test rats compared with the control rats. These findings highlight the roles food plants containing phytonutrients and phytochemicals may play in maintaining good health in polluted environments.

KEYWORDS: Cement dust, food plants, phytonutrients, phytochemicals, and body weight.

I. INTRODUCTION

Environmental pollution and control became a global issue during the industrial revolution in the 18th century (Doyle, 2003). The industrial revolution meant that more goods could be produced for human consumption. However, it also meant that more natural resources would be exploited, and more pollutants would be discharged into the environments (Clive, 1991). Presently, environmental pollution is among the problems the world is facing, increasing in threat with every passing year.

All industries generate harmful waste substances, causing contamination of water, air and soil with harmful effects on both humans and environments. The main environmental pollutants include particulate matters, aluminium, dioxins, heavy metals, sulphur dioxide, benzene, carbon monoxide, nitrogen dioxide, industrial effluents and agricultural runoff (European Public Health Alliance, 2009; Yahaya and Okpuzor, 2011). Pollutants can cause both acute and chronic health problems in animals and humans, involving several organs and systems. Short- and long-term exposures to pollutants have also been associated with high mortality and shortened life expectancy (Marilena and Elias, 2008). Despite these health consequences, industries keep expanding because they make life easier. Hence, the need to control and ameliorate health effects of pollutants from various industries.

Pollution prevention and control strategies have not been successful, especially in developing nations due to lack of funds, ignorance, strategy technicalities and weak environmental protection laws (Yahaya *et al.*, 2012). Even in developed countries, pollution control has not been 100 % successful with large proportions of annual national budgets allocated to pollution control. According to a researcher from Kansas State University, United States spend more than \$ 4.3 billion annually on freshwater pollution mainly from agricultural runoff. The country also spends well over \$ 50 billion annually on pollution from automobiles and more than \$ 234 billion yearly on industrial pollution (KSU, 2008). Since phytomedicine is one of the strategies employed in health care delivery, it is necessary to assess the potentials of some food plants in ameliorating effects of pollutants. This study evaluates the ameliorative efficacy of some selected food plants on some physical characteristics of albino rats exposed to cement dust.

II. MATERIALS

Animal Care

One hundred and fifty albino rats weighing between 185 and 200 g were sourced from the Department of Biochemistry, University of Ibadan in August 2009. The rats were made to acclimatize to the ambient environment before commencing the experiment. Pellet feeds from the F. A Feeds industry, Agege-Lagos and water were given to the rats *ad libitum*.

Elemental Analysis of the Cement Dust.

The elemental analysis of the cement dust was carried out by Atomic Absorption Spectroscopy (AAS). Spectrophotometer (UNICAM model 969) was used for the analysis in the Department of Chemistry, University of Lagos.

Source of the Plant Materials

The plant materials- roselle, moringa, ginger and 'ugwu' were purchased from Ketu in Lagos metropolis, Nigeria. They were identified by a curator, Mr. Odewo T. Kolawole, in the Department of Botany, University of Lagos. The voucher numbers of the authenticated samples are LUH 4394, LUH 4558, LUH 4396 and LUH 4395 for roselle, moringa, ginger, and 'ugwu', respectively.

Preparation of the Plant Materials

Fresh leaves of the individual plant materials were washed gently to remove impurities and air-dried under shade for one week. The dried leaves were milled into a powder using laboratory mill, Norris Limited, Poole, England at the Department of Pharmacognosy, University of Lagos. A mixture of the individual plant materials was also obtained by mixing the four parts each of the ground plant materials in the ratio 1:1:1:1. The ground plant materials were then stored in desiccators before use.

Preparation of the Plant Extracts

The bio-active compounds were extracted from the plant materials using the method of Okigbo and Ogbonnaya (2006). Fifty grams (50 g) powder of each plant material and the mixture were put in 500 ml 95% cold ethanol for 72 hours. The extracts thus obtained were filtered with muslin cloth and evaporated to dryness at a temperature of $40\pm2^{\circ}$ C. The resulting dried extracts of each plant material yielded 6.6 g, 6.5 g, 6.2 g, 5.9 g, and 6.1 g of roselle, moringa, ginger, 'ugwu' and mixture, respectively. These dry extracts were reconstituted in water and were the decoctions used for the experiment.

Phytoconstituents Screening of the Plant Extracts

The phytochemicals in the plant extracts were identified using standard procedures as described by Harbone (1973) and Sofowora (1993). The phytonutrients were screened using thin layer chromatography (TLC) method as described by Meloan (1999).

Acute Toxicity Test

The acute toxicity of the crude extracts of the plants was measured using the 'Classical LD_{50} ' method described by Gabriel *et al.* (2008). Albino rats (36) of both sexes weighing between 183 and 205 g were used for the studies. The rats were randomly distributed into six groups of 6 rats each and were denied food and water 12 hours before commencing the study. The rats in the test groups were orally administered doses of 200, 400, 500, 700, 1500, and 2000 mg kg⁻¹ of the crude extracts. The control rats received only distilled water. The general symptoms of toxicity were monitored and recorded for each group within 24 hours.

Dosage Administered to the Rats

The acute toxicity test showed the plant extracts were nontoxic to the rats even at a dose of 2000 mg kg⁻¹. However, 400 mg kg⁻¹ dose was chosen because a previous study by Adedapo *et al.* (2009) showed some of the understudy plants work best in rats at the dose.

III. METHODS

Study Design

The rats were placed into six groups of 18 rats each. Group one was the control, and groups two through six formed the test rats. The entire rats were exposed to cement dust at 200 km from a cement factory in Shagamu, Nigeria. The body weights of the rats were measured before commencing the experiment. The test rats were thereafter treated with 400 mg kg⁻¹ ethanol extracts of roselle, moringa, ginger, 'ugwu' and a mixture of the plant extracts, respectively. The control rats received only distilled water. The body weight, percentage

death, average number of newborn per birth, and percentage offspring survival rates were monitored for 180 days.

Relative Growth Rate (RGR) of the Rats

The relative growth rate of rats across the groups was calculated using the formulae below: $RGR(\%) = \frac{WF - WI}{T} X 100$

Where W_F = final weight; W_I = initial weight; and T = period of exposure (Winder *et al.*, 1990)

Percentage Death, Average Number of Newborn per Birth, and Percentage Offspring Survival. The percentage death, average number of newborn per birth, and percentage offspring survival of the rats were calculated from the formula below:

 $Death (\%) = \frac{Number of deaths recorded}{Number of rats} \times 100$

Average Number of Newborn per Birth = $\frac{Number \ of \ Newborns}{Number \ of \ Delivery}$

 $Offspring \ survival(\%) = \frac{Nuumber \ of \ newborns \ that \ survived}{Number \ of \ newborns} \ x \ 100$

IV. RESULTS

The elemental analysis of the cement dust shows it contains 57 % calcium, 23 % silicon, 10.5 % aluminium, 8.5 % chromium and 8.0 % lead.

The phytochemicals found in the roselle extract include alkaloids, tannins, glycosides and reducing sugars, and moringa contains all the tested phytochemicals except flavonoids and phlobatanins. Ginger extract has glycosides, reducing sugars, saponins, and flavonoids, and 'ugwu' extract has all the phytochemicals except reducing sugars and phlobatanins. The mixture extract has all the tested bio-active compounds. The phytonutrients analysis of the extracts shows roselle contains calcium, iron, zinc, magnesium, vitamins A, and vitamin C, and ginger extract has zinc, magnesium, vitamin A and vitamin C. Moringa, 'ugwu' and mixture extracts have all the tested nutrients.

The results of the acute toxicity test showed the plant extracts were nontoxic to the rats even at a dose of 2000 mg kg⁻¹. The general observations showed no mortality occurred 24 hours after administering the plant extracts. The rats that received roselle extract displayed a readiness to take more and were licking the <u>cannula</u> used to administer the extract. The rats that received ginger, moringa, 'ugwu', and mixture extracts did not show any signs of illness.

Table 1 shows both the control and test rats gained considerable weights during the period of exposure. However, a significant difference (p < 0.05) in weight was observed between the control and test rats. The control rats had a weight increase of 14.29 mg kg⁻¹, whereas the test rats fed with roselle, moringa, ginger, 'ugwu', and mixture extracts had 25.24, 29.61, 20.56, 28.94 and 34.89 mg kg⁻¹, respectively. Significant differences (p < 0.05) were also observed in the test rats. The mixture of the plant extracts had the highest body weight increase of 34.89 mg kg⁻¹, followed by moringa, 29.61> 'ugwu', 28.84> roselle, 25.24 > ginger, 20.56. Figure 1 shows the relative growth rates of the exposed rats per day, where a significant difference (p < 0.05) was observed between the control and test rats. The control rats had the relative growth rate of 15.1 %. The rats that received extracts of roselle, moringa, ginger, 'ugwu', and the mixture had relative growth rates of 26.1, 30.9, 23.0, 30.7 and 36.5 %, respectively.

Day	0	30	60	90	120	150	180	Min. Wgt.	Max. Wgt.	RGR	Wgt. Increase
Extract											
Control	190.42ª ±11.2	193.67ª ±9.6	198.07ª ±10.3	202.30ª ±9.39	207.83ª ±8.88	212.35ª ±10.23	217.60ª ±10.61	176.9	228. 9	15.1 0 ±3.4	27.2 ±7.8
Roselle	185.82ª ±9.75	190.77ª ±8.12	199.42ª ±6.26	209.17 ^b ±6.76	215.15 ^b ±6.47	222.82ª ±5.91	232.73 ^b ±7.78	176.8	243. 0	26.1 0 ±4.1	46.9 ±6.98
Moringa	187.93ª ±9.57	196.60 ± 7.83	206.88 ±9.71	217.48 ^b ±6.11	226.48ª ± 8.21	234.00ª ±6.96	243.48 ^b ±7.78	175.9	249. 8	30.9 0 ±4.6	55.6 ±6.90
Ginger	200.88 ª ± 7.87	204.92ª ±5.82	213.55 ±6.13	219.62ª ±6.31	227.35 ^b ±5.91	233.00 ª ±7.38	242.22 ^b ±7.98	192.3	249. 9	23.0 0 ±3.2	41.3 ±5.87
'Ugwu'	191.08 ª ±9.11	200.30ª ±8.75	207.42ª ±9.06	213.20ª ±10.5	222.98ª ±8.14	236.22 ^b ±7.56	246.35ª ±8.51	173.5	255. 8	30.7 0 ±5.1	55.3 ±6.45
Mixture	188.60ª 11.91	199.20 5 ±12.01	209.92ª ±13.18	221.13 ^b ±15.8	231.58ª ±12.9	242.02 ^b ±13.45	254.37ª ±12.01	167.2	267. 3	36.5 0 ±5.0 7	65.8 ±6.34

Data are expressed as MEAN±SE

Mean values in the same row with different superscripts 'a' and 'b' were significantly different at p < 0.05RGR = Relative Growth Rate

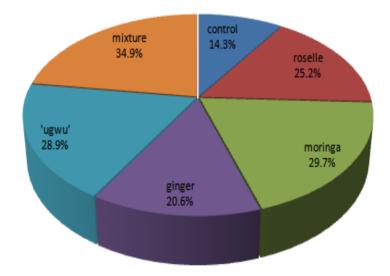


Figure 1: Relative growth rates of the exposed albino rats fed with different plant c extracts for 180 days.

Table 2 describes the effects of the plant extracts on some physical characteristics of the rats. The physical characteristics of the control and test rats showed a significant difference (p<0.05), which was also observed among the test rats. Roselle had the highest death of 33.3 %, and the mixture of the plant extracts had the lowest death of 16.7 %. Moreover, the mixture of the plant extracts produced the highest average number of newborn of 14.0/litter, and ginger had the lowest average number of newborn of 8.0/litter. The highest offspring survival of 76.4 % occurred in the mixture rats, and roselle fed rats had the lowest offspring survival of 50.0 %. The rats that were administered with extracts of mixture, moringa, ginger and 'ugwu' were active, however, the rats that received mixture of plants extracts rats being the most active. The rats that were fed with roselle extract were not as active, and the control rats moved sluggishly.

rats								
Physical characteristics	Death (%)	Ave. Number of Newborn per birth	Offspring survival (%)	Physical condition				
Extract								
Control	58.3	6.0ª ±1.00	50.0	Sluggish				
Roselle	33.3	8.33 ^b ±0.58	52.0	Partially active				
Moringa	25.0	12.00 ^b ±1.00	75.8	Active				
Ginger	41.7	8.0ª ±1.00	58.3	Active				
'Ugwu'	25.0	11.0 ^b ±1.00	73.3	Active				
Mixture	16.7	14.00 ^b ±1.00	76.4	Very active				

Table 2: Efficacy of the different plant extracts on some physical characteristics of the exposed albino

Mean values with different superscripts 'a' and 'b' from the control are significantly different at $P < 0.05\,$

V. DISCUSSION

Several studies including the present study have shown that cement dust may contain cytotoxic elements such as heavy metals, dioxins and particulate matters. However, some plants have been reported to prevent or ameliorate the cell and tissue damage processes of these elements.

The weight increase noted in the test rats compared with control rats could be attributed to the antioxidant and cell rebuilding activities of some chemicals and nutrients in the plant extracts. Phytochemical analysis of the plant extracts showed the plants contained glycoside and saponin, both of which could have increased the body weight of the rats. George *et al.* (2002) reported that glycosides and saponins increased the feed intake and development of experimental animals. Okwu (2005) also reported that glycoside indirectly increases the levels of calcium in animals, which specializes in blood and bone formation leading to body weight increase. The food plants also contain essential nutrients such as calcium, magnesium, iron, sodium, potassium and zinc, all of which could have contributed to the body weight of the exposed rats. Adedapo *et al.* (2009) observed an increase in the weight of mice administered with the extracts of moringa. Moreover, increase in weights of rats and birds fed with 'ugwu' diets have been reported by Fasuyi and Nonyeren (2007), and Iweala and Obidoa (2009).

The low birth rates of the control rats may be due to the destruction of their reproductive systems by the toxic elements in the cement dust. Several studies on rats and other rodents indicated that blood lead concentrations above 30-40 mg dl⁻¹ were associated with impairment of spermatogenesis and reduced concentrations of androgens (Apostoli, 1998). Studies have also shown that most of the infertility, birth defects and aborted pregnancies that happened in the United States in the 90s were due to exposure to heavy metal (ATSDR, 1999). The high fertility and birth rates of the test rats may be attributed to the scavenging activities of antioxidants in the plant extracts. The extracts contain flavonoids and tannins; all which could have prevented or mopped free-radicals in the test rats. It could also be attributed to the replenishing activities of the phlobatanins and alkaloids found in the plant extracts. Moringa has been reported by Adedapo *et al.* (2009) and Cajuday and Pocsido (2009) to improve sexual activity in rats by promoting testosterone production. Salman *et al.* (2008) also reported an improvement in sperm count and quality in rats following treatment with 'ugwu' extract. Alkaloids and phlobatanins detected in the plant extracts have been reported by George *et al.* (2002) and Okwu (2004) to increase sexual activity in rats. Alkaloids are aphrodisiac (Harisaranraj *et al.*, 2009) and phlobatanins can synthesize sex hormones (Okwu, 2001; Edeoga *et al.*, 2005).

The low death and high offspring survival rates of the rats fed with the plant extracts may be linked with the prophylactic action of the phytoconstituents. The phytochemicals in the plant extracts may have triggered the mopping of the free-radicals generated by the toxic elements in cement dust. This could have

worked in the wellness of the test rats. Antioxidants protect cells, tissues and organs from oxidative damage. Therefore, the antioxidants in the test plants could have contributed to the observed decrease in death rate, and high offspring survival rate. Also, the essential nutrients in the plant extracts were actively involved in cell rebuilding processes, resulting in improved health condition of the exposed rats. Phytochemicals have been pointed in the reduced mortality rates observed in people consuming high levels of plant-based foods (Arts and Hollman, 2005). Alkaloids are strong antioxidants which can improve physiological activities of animals resulting in improved health status and low death rate (Harisaranraj *et al.*, 2009).

VI. CONCLUSION

The food plants used in this study have been shown to be bio-protective and can be used to ameliorate negative effects of pollutants. It is also suggested that since the plants have high nutritive values, they would be of immense benefit in healthcare delivery and animal husbandry.

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