

# Exposure to Air Freshener and Its Corollary on the Bleeding Time and Electrolyte Profile of Wistar Albino Rats

<sup>1</sup>Odinga, T. <sup>2</sup>Odu, C.O.

1. Department of Biochemistry, Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria.

2. Department of Chemistry, Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria.

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**Abstract:** Given the continuous and widespread use of air freshener as a means of masking or elimination of unpleasant smell and its associated health risks that have been reported, this study investigated the effects of exposure to Air freshener on the Blood Electrolytes as well as the Bleeding time. Gel air fresheners, in an enclosed cage and eighteen wistar albino rats were used for this study. The rats were divided into six groups of three rats each. Group A served as control that was not exposed to air freshener, groups B, C, D, E and F were exposed for 2, 4, 6, 8 and 10 hours respectively. Bleeding time test was carried out as well as the assay for blood electrolyte concentrations: ( $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{HCO}_3^-$ ,  $\text{Cl}^-$  and  $\text{Ca}^{2+}$ ) using the Ultra Violet spectrophotometer. The result of the blood electrolyte assays and that of the bleeding time were analyzed statistically using analysis of variance. Results obtained showed that the bleeding time increased significantly ( $P \leq 0.05$ ) at 4, 6, 8 and 10 hours of exposure and there was a significant difference in electrolytes concentration of exposed animals compared to the control group. This study therefore suggests a reduction in exposure to air fresheners as its adverse health effect is proportional to the length of exposure.

**Key words:** Air Fresheners, blood electrolytes, volatile organic compounds, bleeding time, toxicity

## 1. Introduction

Air quality has been seen to be one of the major environmental health concerns for healthy living. The policy on air pollution is aimed at achieving levels of air quality that do not result in unacceptable risks to human health. [19].

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A greater percentage of air pollutants consist of chemicals derived from the use of cleaning products, air fresheners, pesticides and materials related to furniture, construction, heating, cooking appliances and outdoor pollution sources. [3].

Many of the pollutants cause symptoms which may not occur for many years, making it difficult to discover the cause. Examples of potentially serious effects due to exposure to indoor air pollutants include asthma, breathing disorders and cancer. [13].

Air fresheners are products that do not significantly reduce air pollution, but rather add more substances with a “fresh” odour strong enough to mask a bad odour. [14].

The use of these products is widespread in houses, cars, stores, offices, restaurants, schools, hospitals, churches, theaters, and parks, airplanes, airports, cars, buses, trains, terminals, boats, businesses apartments, hotel lobbies, health clubs, child care and elder care facilities Etc.

Air fresheners and deodorizers can contain hundreds of chemicals, some of them as reported by [17] are toxic even in very small concentration.

Common air freshener chemicals, such as limonene, generate additional hazardous pollutants as formaldehyde and acetaldehyde, which are linked with cancer, ultrafine particles, linked with heart and lung disease. [11].

On Air freshener's labels, the list of all of their ingredients are usually not listed. If any ingredients are listed, they are usually safe-sounding ones, rather than potentially hazardous ones. [18] Reported that all air fresheners tested (sprays, gels, solids, disks, and oils) emitted chemicals classified as toxic or hazardous by federal laws, but none of these chemicals were listed on any product label or material safety data sheet. [3], reported the presence of Volatile Organic Compounds in indoor air following the use of air fresheners, also, in their study, report claims that the emissions of certain substances such as volatile organic compounds (VOCs), sensitizing substances and benzene give rise to serious concerns, other substances such as formaldehyde, terpenes, di-ethyl-phthalate or toluene are of less, little or no concern.

The concentrations of several individual VOCs and also “total VOCs” resulting from the use of several types of air fresheners (incense paper, scented candles, incense, gel fresheners, liquid air fresheners, electric diffusers and sprays) are described in the [3] report. The indoor air concentrations of VOCs depend on a number of factors, such as ventilation rate, adsorption in/on materials and emissions from other sources, including outdoor air contribution.

Pollutants emitted from air fresheners are linked with damage to the brain, lungs, heart, reproductive system, immune system, and with cancer. Everyone is vulnerable, especially children. People can have seizures,

asthma attacks, or lose consciousness if exposed to air fresheners, also a liability risk. [3], reported that a chemical 1, 4-dichlorobenzene, common in air freshener may harm the functioning of the lungs.

In 2003, the international journal of public health reported after a study of 200 pregnant women, that women who use air fresheners in their homes were significantly more likely to have babies that suffer from wheezing and lung infection.

Table 1: Guidance Value for the Concentration of Substances in Air Freshener as Reported by Beuc, 2005.

Compound	Normal Indoor Air Concentration ( $\mu\text{g}/\text{m}^3$ )	Critical Effect of Compound	Guidance Value ( $\mu\text{g}/\text{m}^3$ )	Reference
Benzene	2-13	Cancer	Safe Level Not Established	WHO, 2000
Formaldehyde	8-41	Sensory Irritation	100(30 Minutes average)	WHO, 2000
d- Limonene	14-30	Irritation	450	Index, 2005
Styrene	1-6	Central Nervous System	260(Weekly average)	WHO, 2000
Diethyl Phthalate	-	-	Not Established	-
Toulene	20-87	Central Nervous System and Irritation	260(Weekly average)	WHO, 2000
Total Volatile Organic Compound	-	Depends On the Composition	-	-

Source: BEUC, 2005

### **Bleeding Time**

Bleeding time is used in ascertaining the time it takes for blood vessel constriction and platelet plug formation to occur. Longer than normal bleeding time could be as a result of platelet aggregation defect, blood vessel defect and thrombocytopenia (low platelet count). Diseases that cause prolonged bleeding time include thrombocytopenia of which benzene inhalation as reported by [1], is one of the cause of this disease.

### **Blood Electrolytes**

The blood electrolytes test aids in assessing the fluid and electrolyte balance as well as acid base balance of the body. The electrolytes also help in stimulating nerves throughout the body and balance fluid levels. Their levels can become too high (hyper) or too low (hypo), hence they need to be maintained in an even balance for proper functioning of the body system.

Their functions are summarized thus:

Table 2: Brief summary of Blood electrolytes and their functions.

<b>Blood electrolytes</b>	<b>Role in the body</b>
i. Calcium $\text{Ca}^{2+}$	Helps in muscle contraction, nerve signaling, blood clotting, cell division and forming/maintaining bones and teeth.
ii. Potassium $\text{K}^+$	Keeps blood pressure levels stable, regulating heart contractions, helping with muscle functions
iii. Magnesium $\text{Mg}^{2+}$	Needed for muscle contractions, proper heart rhythms, nerve functioning, bone building and strength, reducing anxiety, digestion and keeping a stable protein –fluid balance.
iv. Sodium $\text{Na}^+$	Helps maintain fluid balance needed for muscle contraction and helps nerve signaling.
v. Chloride $\text{Cl}^-$	Maintain fluid balance.
vi. Phosphate $\text{HPO}_4^{2-}$	Strengthen bones and teeth, essential in energy production within cells, necessary for tissue growth and repair and also a major building block for cell membrane and DNA.
vii. Bicarbonate $\text{HCO}_3^-$	A major component of blood pH

## 2. Materials and Methods

Eighteen wistar albino rats of 100g-120g were used. They were divided into six groups of three rats each and allowed to acclimatize for 14 days. Group A served as a control (was not exposed to air freshener).

Gel air fresheners of 40g weight were kept in a specially built cage for it to sublime and saturate the cage with its content. The experimental animals excluding the control were expose to the air fresheners by putting them into the cage to inhale and absorb the content of the air fresheners.

At time interval of two hours, each group of the experimental animals was brought out and the bleeding time test procedures were carried out as described by [12], which is a modified Duke method. Briefly, the tail of each animal from the respective group was disinfected by cleaning with ethanol. The tail of the animal was held from the perforated spaces in each cage, the tip was quickly cut using a disposable lancet and the stopwatch was started as soon as bleeding started.

The cut was dabbed with filter paper every 15 seconds until the paper no longer stained red with blood. Bleeding time was then taken as the time when the blood stopped flowing from the cut. The bleeding time was determined by counting the number of spots the blood blotted off on the filter paper. The spot was then added up, it was multiplied by the interval by which the blood was blotted off (30 seconds) and converted to minutes.

Immediately after obtaining the bleeding time, the experimental animals were sacrificed and blood collected using the lithium Heparin bottles. This was subsequently assayed for electrolyte concentration of serum potassium, sodium, chloride, bicarbonate and calcium using the UV Spectrophotometer.

Table 3: Exposure time in hours of experiment animal.

Groups	Exposure time(hours)
A(control)	Not exposed
B	2 hours
C	4 hours
D	6 hours
E	8 hours
F	10 hours

Statistical analysis of results were carried out using the analysis of variance (ANOVA).

### 3. Results and Discussion

Table 4: Bleeding Time of Experimental Animals.

GROUPS	BLEEDING TIME (Minutes)
A	11.04 <sup>a</sup> ± 3.43
B	18.49 <sup>ab</sup> ± 5.28
C	25.38 <sup>bc</sup> ± 4.39
D	28.16 <sup>bc</sup> ± 3.68
E	28.82 <sup>c</sup> ± 0.55
F	31.47 <sup>c</sup> ± 2.05

- Values are expressed as mean ± standard deviation
- Values with different superscripts show significant difference at P≤0.05
- Values with the same super scripts shows no significant difference at P≤0.05

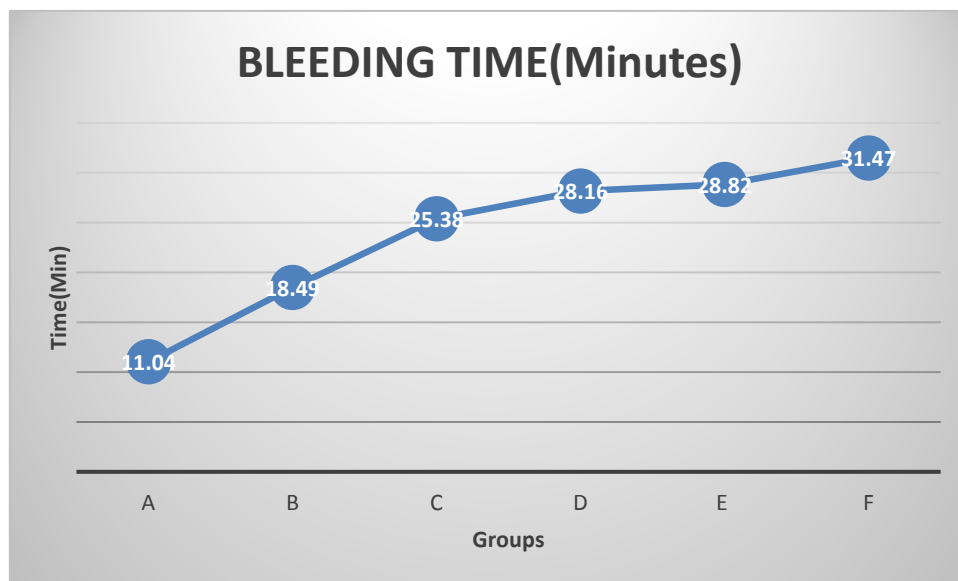


Fig 1: Graphical representation of the bleeding time of experimental animals

Table 5: Blood Electrolyte Profile of Experimental Animals.

GROUP	ELECTROLYTES				
	Potassium K <sup>+</sup> (mmol/L)	Sodium Na <sup>+</sup> (mmol/L)	Chloride Cl <sup>-</sup> (mmol/L)	Bicarbonate HCO <sub>3</sub> <sup>-</sup> (mmol/L)	Calcium Ca <sup>2+</sup> (mmol/L)
A (control)	9.33 ± 0.50 <sup>a</sup>	80.67±6.51 <sup>a</sup>	125.67±2.52 <sup>ab</sup>	30.00±2.00 <sup>a</sup>	2.57±0.25 <sup>ab</sup>
B	8.30 ± 5.20 <sup>a</sup>	69.67±2.52 <sup>a</sup>	122.00±79.77 <sup>ab</sup>	25.33±3.06 <sup>a</sup>	2.56±0.35 <sup>ab</sup>
C	6.67 ± 2.00 <sup>b</sup>	79.67±17.50 <sup>a</sup>	121.67±1.53 <sup>ab</sup>	26.67±3.06 <sup>a</sup>	2.67±0.06 <sup>ab</sup>
D	5.67±1.12 <sup>b</sup>	81.00±5.00 <sup>a</sup>	112.00±3.00 <sup>ab</sup>	25.33±3.06 <sup>a</sup>	2.40±0.10 <sup>b</sup>
E	4.47±0.76 <sup>c</sup>	82.33±1.53 <sup>a</sup>	123.00±35.00 <sup>ab</sup>	29.33±1.16 <sup>a</sup>	3.03±0.26 <sup>c</sup>
F	4.76±0.63 <sup>c</sup>	143.00±5.00 <sup>b</sup>	86.67±10.69 <sup>a</sup>	26.67±1.16 <sup>a</sup>	1.63±0.06 <sup>a</sup>

- Values are expressed as mean ± standard deviation
- Values with different superscripts show significant difference at P≤0.05
- Values with the same super scripts shows no significant difference at P≤0.05

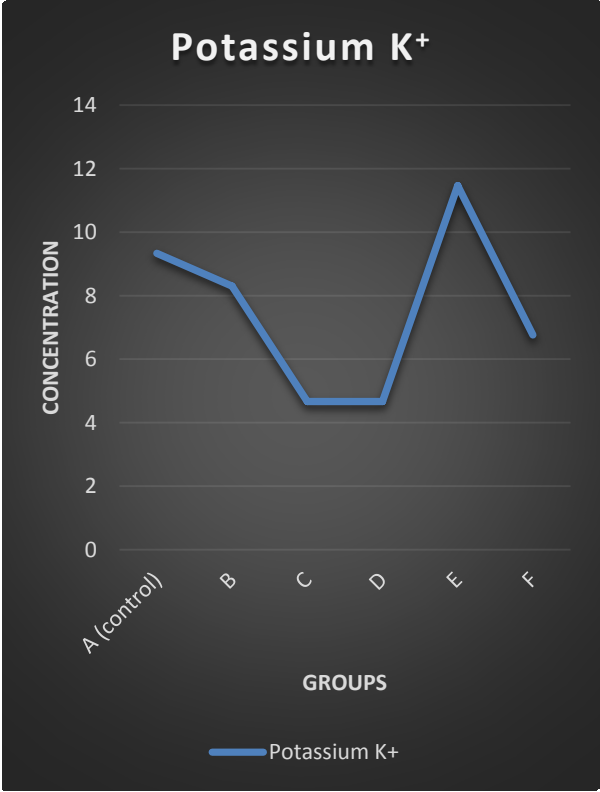


Fig 2: Graphical representation of the Potassium level of experimental animals

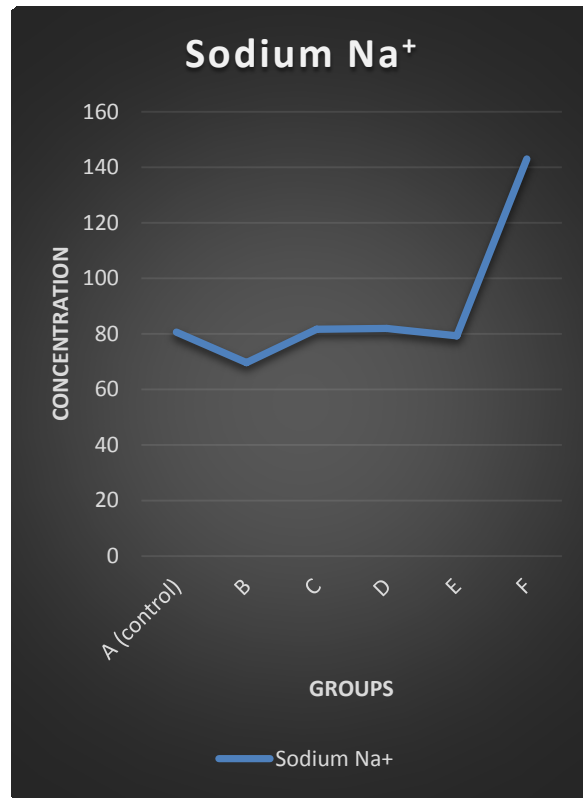


Fig 3: Graphical representation of the Sodium level of experimental animals

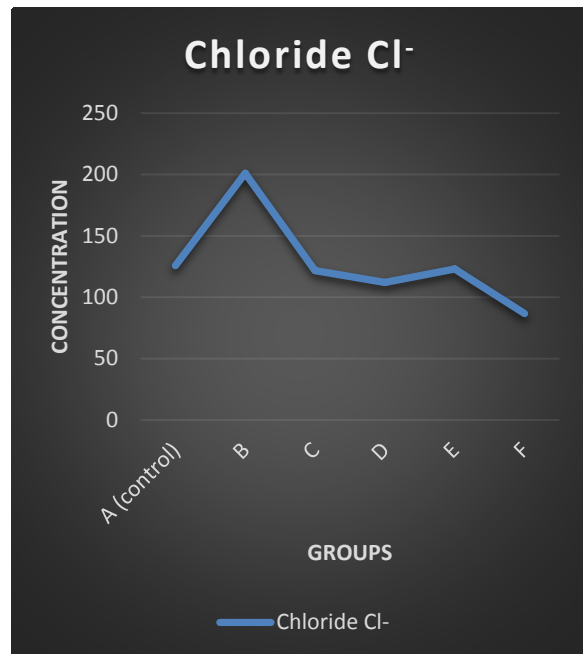


Fig 4: Graphical representation of the Chloride level of experimental animals



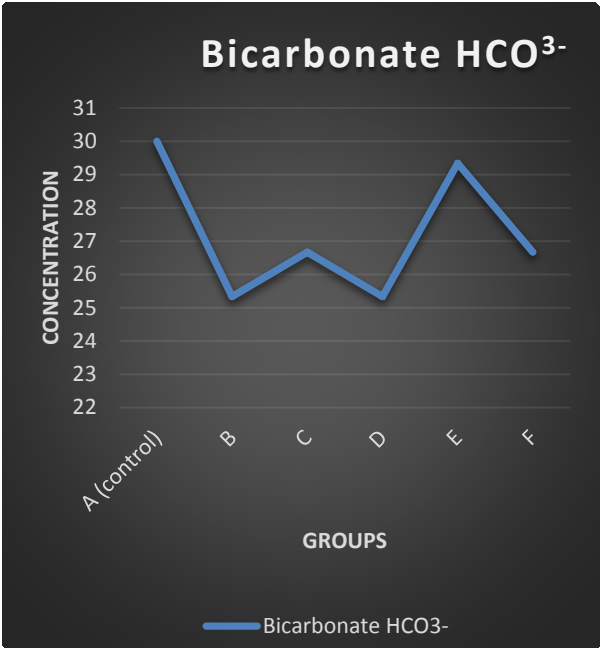


Fig 5: Graphical representation of the Bicarbonate level of experimental animals

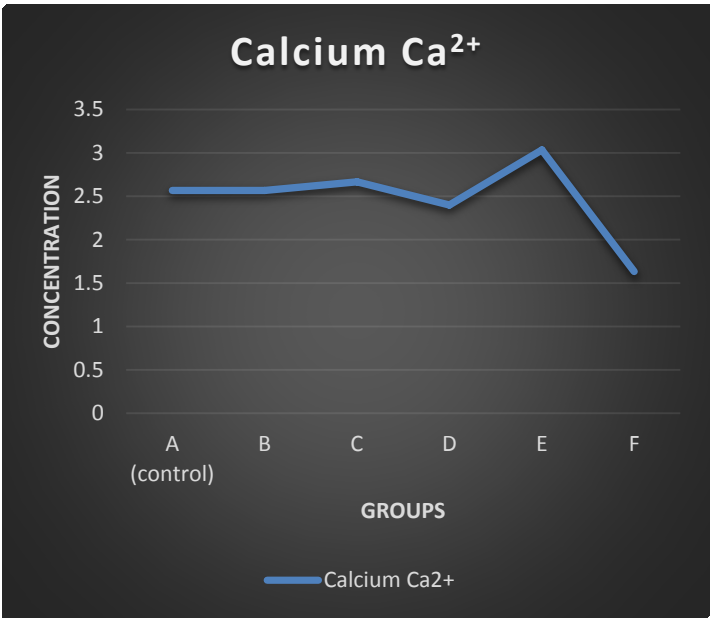


Fig 6: Graphical representation of the Calcium level of experimental animals

## **4. Discussion**

Air freshener though it emits fragrance, isn't actually freshening as they work by spraying a whole bunch of chemicals that barely masks odour (smelly air) while exposing us to an inhalable toxic chemicals which may contribute to electrolyte imbalance and also leads to excessive bleeding as seen from this research work.

In the bleeding time test, there was a consistent increase in bleeding rate as the exposure time increased (fig 1) such that group F that had the highest exposure time showed a huge significant difference compared to the control group. This could be attributed to inhalation of toxic volatile organic compounds like benzene etc. that could have been present in the air freshener. Following inhalation exposure, about 50% of the benzene is absorbed in rats. Once thoroughly distributed, benzene is bioactivated to a number of metabolites, which are very likely responsible for its adverse effects [3].

The hypocalcemia observed in group F could as well results to the excessive bleeding, Calcium has been reported to aid blood clotting process.

The imbalance in electrolyte concentration of Potassium, Sodium, and Calcium from the result could be attributed to the Volatile organic compounds and chemicals present in the air fresheners and inhaled by the experimental animals. BEUC, 2005, reported that these chemicals have the potency to disrupt the metabolic and biochemical functions of the body. The chemicals in the air freshener may have interfered with the integrity of those hormones altering its proper (normal) function [12]. Table 5 shows the effects of exposure time on the blood electrolytes of the experimental animals. A decrease in the concentration of Potassium was observed as the time of exposure increased in comparison with the control group. Hyperkalemia (An excess of Potassium) often accompanies renal failure. An imbalance in the concentrations of Sodium and Bicarbonate was observed in comparison to the control group. However, there was an increase in the concentration of Sodium at exposure times of 6, 8 and 10 hours. The Calcium and Chloride levels were also evaluated on exposure to the air freshener. A significant decrease was observed in the Calcium concentration in comparison to the control at 6, 8 and 10 hours. Calcium is necessary for muscle contraction, blood coagulation and transmission of nerve impulses. Hypercalcemia (High Calcium level in the blood) can be caused by the overuse of antacids and condition that cause movement of calcium out of the bones and into the extracellular fluid such as bone tumors, multiple of fractions-osteoporosis and certain tumors [21].

## 5. Conclusion

Exposure to Air freshener has the potency of altering the biochemical functioning of the body. This is due to the presence of toxic chemicals such as the Volatile organic compounds, which are toxic even at very low concentration, hence exposure to synthetic air fresheners should be discouraged while encouraging the use of plants with natural fragrance as an alternative.

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