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# Logistic Analysis Of Acute Toxicity Of Hunteria Umbellata Extract In Mice Through Intraperitoneal Route

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***Abstract***

***In this paper, we analyse the acute toxicity of Hunteria umbellata, a herbal medicinal plant, in mice in Nigeria using the logistic model. Hunteria umbellata is a plant with therapeutic potentials in the treatment of various diseases that include yaws, peptic ulcers, diabetes, piles, infertility and inflammation. Data on the acute toxicity of seeds extract of Hunteria umbellata is obtained via intraperitoneal route and analysed. The median lethal dose is determined and found to be 1.5979g/kg of body weight with confidence limits as [1.4335g/kg, 1.7811g/kg]. This shows slight toxicity of Hunteria umbellata and its toxicity at a high dose on acute exposure should be put into consideration when it is used as treatment for these diseases.***

**Keywords**: *Hunteria umbellata*, logistic model, acute toxicity, median lethal dose ($LD_{50}$).

## 1.0 Introduction

The environment we live in is filled with abundant resources and chemicals endowed to humans by nature. In order to study the harmful nature of these resources, there is need to make contact with them through any route of exposure depending on the physical and chemical properties of the substance(s) of interest. This could be achieved through inhalation, skin absorption, ingestion or injection.

Many plants have been used as alternatives to orthodox medicines in Africa due to the traditional belief of people about these plants. These plants may be sources of substances with better therapeutic potentials than some currently used orthodox medicines [17, 24]. *Hunteria umbellata* is a small tree of about 15 – 22m in height with a dense evergreen crown [19] of great medicinal benefits and is found in Nigeria, Ghana, Cameroon and some countries of Central Africa. In Nigeria, it is found in the rain forest zone of the southern parts of Nigeria with local names as Osu (Edo), erin (Yoruba) and nkpokiri (Ibo) [5, 16].

The plant is used for the treatments of yaws, peptic ulcers, diabetes, piles, dysmenorohea, fevers and infertility [10, 11, 21] and inflammation [12]. It has been used in the treatment of various ailments in Nigeria and Ghana especially the leaves, roots and bark [14].

We have, therefore, undertaken the present study to analyses the acute toxicity of *Hunteria umbellata* extract administered to mice through intraperitoneal route with the aid of logistic model to determine the lethal dose of the seed extract.

## 2 Models and Toxicity Study by Probit Analysis

### 2.1 Dose-response models

Various mathematical models have been used in analyses of dose-response relationships to assess the toxic effects of chemical substances. These models range from very simple models to extremely complicated models for which the eventual functional forms cannot be easily expressed as single equations. Categorically, these models are easily assessible in the works of [7, 8, 9, 13, 18, 22, 23]. In order to determine the acute toxicity of *Hunteria umbellata* plant in this paper, the median lethal dose ($LD_{50}$) of the logistic model is employed.

### 2.2 Acute toxicity study using probit analysis

Overnight-fasted Swiss albino mice (17-23g) of either sex were used for the study as test subjects in a laboratory experiment conducted in University of Benin, Benin City, Nigeria. The animals were divided into four groups of five animals each. Groups A to D received 1.4, 1.6, 1.8 and 2.0g/kg of the extract, respectively, through intraperitoneal route. Number of deaths that occurred in each group was 0, 3, 4 and 5 respectively. Using probit analysis, it was observed that the median lethal dose ($LD_{50}$) gave 1.66g/kg.

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## 3 Method of Analysis of Acute Toxicity By Logistic Model

Suppose X is a response data with data points $x_1, x_2, \ldots, x_n$, then the logistic model is written as;

$$P(d)=\{1+\exp[-(\alpha+\beta x)]\}^{-1} \; ; \alpha,\beta>0 \tag{1}$$

where $x = \log_e$ (dose) is the exposure to the dose and dose is the total amount of a substance administered or taken up by test subject(s). $\alpha$ *and* $\beta$ are shape and scale parameters respectively.

In order to estimate the shape and scale parameters of the logistic model, there is need to transform (1) to a simpler linear equation. This is given as;

$$Log_e\left[\frac{P(d)}{1-P(d)}\right] = \alpha+\beta x \tag{2}$$

Equation (2) is known as the logit transformation [2]. The linear equation in (2) relates the response to the dose for the $ith$ experimental group as;

$$z_i = \alpha + \beta x_i \quad , i=1(1)m \tag{3}$$

where $z_i$ is the logit of the response rate. The weighted least square method uses the observed response rates $w_i = n_i\ p_i\ q_i$. In the event that $p_i = 0\ or\ 1,$ which would result in the weighting factor being zero, $p_i = \frac{1}{2n_i}$ in place of zero and $p_i = 1 - \frac{1}{2n_i}$ in place of unity would be used [3] and $n_i$ is the number of animals in the $ith$ dosing level.

The weighted least squares technique is thus, applied on the logit model of (3) to produce the following estimates $\hat{\alpha}$ *and* $\hat{\beta}$ as shown below.

$$WSSE\ (\alpha,\beta) = \sum_{i=1}^{m} w_i\left[z_i-(\alpha+\beta x_i)\right]^2 \tag{4}$$

Minimizing WSSE (weighted sum of square for error) with respect to $\alpha$ *and* $\beta$, and equating to zero, the estimates of $\alpha$ *and* $\beta$ of the logistic model are obtained as

$$\hat{\alpha} = \frac{\sum_{i=1}^{m} w_i x_i^2 \sum_{i=1}^{m} w_i z_i - \sum_{i=1}^{m} w_i x_i \sum_{i=1}^{m} w_i x_i z_i}{\sum_{i=1}^{m} w_i \sum_{i=1}^{m} w_i\, x_i^2 - \left(\sum_{i=1}^{m} w_i x_i\right)^2} = \bar{z} - \hat{\beta} x_i \tag{5}$$

$$\hat{\beta} = \frac{\sum_{i=1}^{m} w_i \sum_{i=1}^{m} w_i x_i z_i - \sum_{i=1}^{m} w_i z_i \sum_{i=1}^{m} w_i x_i}{\sum_{i=1}^{m} w_i \sum_{i=1}^{m} w_i\, x_i^2 - \left(\sum_{i=1}^{m} w_i x_i\right)^2} = \frac{\sum w_i\left(x_i-\bar{x}\right)\left(z_i-\bar{z}\right)}{\sum w_i\left(x_i-\bar{x}\right)^2} \quad ,i=1(1)m \tag{6}$$

where $\bar{x} = \frac{\sum w_i x_i}{\sum w_i}$ *and* $\bar{z} = \frac{\sum w_i z_i}{w_i}$, and m is the number of dosing levels.

An estimate of the $LD_{50}$ which is the dose resulting in a 50% of responders can be obtained for the logistic model as;

$$\hat{X}_{50} = \log_e(LD_{50}) = \frac{-\hat{\alpha}}{\hat{\beta}} \tag{7}$$

Also, the confidence limits for the dose $LD_{50}$, given the desired response rate, can be obtained as;

$$\exp\left(\hat{X}_{50} - Z_{\frac{\alpha}{2}}\, S_{\hat{x}_{50}}{}^{(L)}\right) \le \hat{L}D_{50} \le \exp\left(\hat{X}_{50} + Z_{\frac{\alpha}{2}}\, S_{\hat{x}_{50}}{}^{(L)}\right) \tag{8}$$

where $Z_{\frac{\alpha}{2}}$ is the value obtained from the table of the standard normal distribution at $\alpha\%$ level of significance and m is the number of dosing levels and the variance for $\hat{X}_{50}$ of the logistic model is given as;

$$S^{2\ (L)}_{\hat{X}_{50}} = \left(\frac{1}{\hat{\beta}}\right)^2 \left( \frac{1}{\sum w_i} + \frac{\left(\hat{X}_{50} - \bar{x}\right)^2}{\sum w_i \left(x_i - \bar{x}\right)^2} \right) \quad [6] \qquad (9)$$

## 4 Result

**TABLE 1**: **Observation of acute toxicity of Hunteria umbellata extract in mice (intraperitoneal route)**

| Dosage level(g/kg) | Number at risk | Number of deaths |
|---|---|---|
| **1.4** | **5** | **0** |
| **1.6** | **5** | **3** |
| **1.8** | **5** | **4** |
| **2.0** | **5** | **5** |

**Source: [15]**

The estimates of shape and scale parameters of logistic model are obtained from (5), (6) and (7) using the information in Table 1 as; $\hat{\alpha} = -5.4146$ *and* $\hat{\beta} = 11.5512$

And $\hat{X}_{50} = \log_e (L\hat{D}_{50}) = 0.4687$

It follows that

$$L\hat{D}_{50} = 1.5979 g/kg \ .$$

At 95% confidence limits, we obtain

$$L\hat{D}_{50} \in [1.4335 g/kg, 1.7811 g/kg] \ .$$

## 5 Discussion

Acute toxicity study using the logistic model gave a median lethal dose of 1.5979g/kg. This indicates that *Hunteria umbellata* plant is slightly toxic. The result is supported by the statement of [1].

According to [1], any chemical substance with $LD_{50}$ value less than 2g/kg but greater than 1g/kg could be considered to be slightly toxic. Thus, the result obtained in the determination of the acute toxicity of *Hunteria umbellata* suggests it could be toxic at a high dose on acute exposure.

Also, it is seen that the reported $LD_{50}$ value obtained using the probit model in the laboratory experiment in section 2.2 falls within the confidence limits obtained for the $LD_{50}$ value via the logistic model. This suggests that one or more environmental factors would have influenced the $LD_{50}$ value of the probit model.

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## References

[1] American Society for Testing and Materials (1987). Standard Test Method for Estimating Acute Oral Toxicity in Rats. American Society for Testing and Materials E1163 87, Philadelphid, USA.

[2] Berkson J (1944). Application of the Logistic Function to Bioassay. J. Am. Stat. Association, Vol. 39, pp. 357 – 365.

[3] Berkson J (1953). A Statistically Precise and Relatively Simple Method of Estimating the Bioassay with Quantal response based on the logistic function. J. Am. Stat. Association, Vol. 48, pp. 565 – 599.

[4] Bevan CWL, Patel MB, Reefs AH and London AG (1967). The seed alkaloids of *Hunteria umbellata*. Tetra Hedron, Vol. 23, pp. 3809 – 3821.

[5] Boone MJ (2006). *Hunteria umbellata* (K.Schum) Hallier F.In:Schmelzer GH & Gurib-Fakim A (Editors) Prota II:Medicinal plants/Plantes medicinales (CD-Rom).PROTA Wageningen. Netherland.

[6] Brown CC (1978). The Statistical Analysis of Dose-Effect Relationship. pp 115 – 148. In: Principles of Ecotoxicology. Scope 12. John Wiley & Sons. New York.

[7] Chiu WA, Barton HA, DeWoskin RS, Schlosser P. Thompson CM, Sonawane B, Lipscomb JC and Krishnan K (2007). Review: Evaluation of Physiologically – Based Pharmacokinetic Models for use in risk assessment. Journ. Of Applied Tox, Vol. 27, pp 218 – 237.

[8] Christensen, E.R. (1984). Dose-response Functions in Aquatic Toxicity Testing and the Weibull model. Ecological Modelling, Vol. 22, pp. 13 – 20.

[9] Ellwein LB and Cohen SM (1988). A Cellular Dynamics model of experimental bladder cancer: Analysis of the effect of sodium saccharin in the rat. Journal of Risk Analysis, Vol. 8, pp 215 – 221.

[10] Eluyoba AA (1995). Female Infertility in the Lands of traditional birth attendants in South-Western Nigeria. Fiitoterapia, Vol. 66, pp 239 – 248.

[11] Falodun A, Nworgu ZAM and Ikponmwonsa MO(2006). Phytochemical components of *Hunteria umbellata* (K.Schum) and its effect on isolated non-pregnant rat uterus in oestrus. Pak. J. Pharm. Sci., Vol .19, pp 256-258.

[12] Gills LS (1992). Ethnomedical uses of plants in Nigeria. Benin City. Uniben Press. pp 134.

[13] Hoel DG (1985). Mathematical dose-response models and their applications to risk estimation. pp 347-359. In: Methods for Estimating Risk of Chemical Injury: Human and Nonhuman Biota And Ecosystems, Scope/SGOMSEC 2. John Wiley and Sons.

[14] Ibeh IN, Idu M and Ataman JE (2005). Toxicological assessment of Abeere seed (*Hunteria umbellata* K. Schum). Journal of Medicine and Biomedical Research, University of Benin, Benin City, Vol. 4, No. 1, pp 44 – 48.

[15] Igbe I (2006). Antidiabetic activity of the aqueous seed extract of Hunteria umbellata in normal and streptozotocin induced diabetic rats. M.Sc. Thesis. Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

[16] Igbe I, Ching FP and Eromon A (2010). Anti-Inflammatory activity of Aqueous fruit pulp extract of *Hunteria umbellata* K. Schum in Acute and Chronic Inflammation. Acta Potoniae Pharmaceutica-Drug Research, Vol. 67, pp. 81 – 85.

[17] Igbe I, Ozolua RI, Okpo SO and Obasuyi O (2008). Antipyretic and Analgesic Effects of the Aqueous Extract of the Fruit Pulp of *Hunteria umbellata* K. Schum (Apocynaceae). Trop. J. Pharma. Research, Vol. 8, pp. 331 – 336.

[18] Moolgavkar SH (1989). A two-stage Carcinogenesis model for risk assessment. Journal of Cell Biol. Toxicology, Vol. 5, pp 445 – 460.

[19] Oliver-Bever B(1986). Medicinal Plants in tropical West Africa. Cambridge University Press, Cambridge, pp. 60 – 62.

[20] Oluwemimo A and Usifoh CO (2001). The antihelmintbic activity of *Hunteria umbellata* K. Schum (Fam. Apocynaceae) extracts. Pak J. Pharm. Sci Ind. Res, Vol. 44, pp 286 – 290.

[21] Raman A and Mallam V (1994). Enhanced in vitro activity of glucokinase enzyme in the presence of *Hunteria umbellata* seeds, a traditional Nigerian treatment for diabetes J. Pharm. & Pharmocol., Vol. 46, pg 1046.

[22] Reddy MB, Yang RSH, Clewell HJ III and Andersen ME (2005) Physiologically – Based Pharmacokinetic (PBPK) Modelling: Science and Applications John Wiley and Sons. pp. 420.

[23] Szymczak W and Szadkowska – Stanczyk I (2005). Cancer Risk Assessment: Present and Future. International Journal of Occupational Medicine and Environmental Health Vol. 18, pp 207 – 223.

[24] Wang Y, Wang X and Cheng YA (2006). A Computational approach to botanical drug design by Modelling quantitative compositions activity relationship, Chem. Bio. Drug Design, Vol. 68, pp 168 – 172.