American Scientific Research Journal for Engineering, Technology, and Sciences (ASRJETS)
ISSN (Print) 2313-4410, ISSN (Online) 2313-4402

http://asrjetsjournal.org/

Aqueous Extract of Rauwolfia Vomitoria Afzel (Apocynaceae) Roots Effect on Blood Glucose Level of Normoglycemic and Hyperglycemic Rats

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Abstract

This study aims to assess the pharmacological effects of aqueous extract roots of *Rauwolfia vomitoria* (EARv), a plant used in traditional medicine in the Ivory Coast to treat diabetes, on blood glucose normoglycemic rats and on glucose tolerance in rats by administration of glucose. The acute toxicity, as well as the phytochemicals present in this extract is also determined. During the experience, fives groups of rats received respectively distilled water, EARv at doses of 500, 700 and 1000 mg/kg of body weight and 10 mg/kg of glibenclamide. Then the blood glucose level of each rat was measured using a glucometer. Hyperglycemia was induced in rats by oral administration of glucose at dose of 4 g/kg. The rats were pretreated or post-treated of the same doses of tests substances and blood glucose level of each rat was measured. Acute toxicity by oral administration was studied in mice and phytochemical screening was performed by thin layer chromatography. The results show that EARv administered orally at doses up to 5000 mg/kg B.W., does not cause the death of the treated mice.

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At 500, 700 and 1000 mg/ kg B.W., EARv causes, like glibenclamide at 10 mg/kg B.W., a reduction of blood glucose level of treated normoglycemic rats. In addition, EARv at 1000 mg/kg B.W. reduced and quickly nullifies the glucose tolerance in rats that is induced by oral administration of glucose (4 g/kg B.W.). This effect is also observed with glibenclamide (10 mg/kg B.W.). The phytochemical screening shows that EARv contains alkaloids, flavonoids, and antrhones and anthraquinones, catechin tannins, saponins and monoterpenoids. This study shows that the aqueous extract of *Rauwolfia vomitoria* has hypoglycemic and some antihyperglycemic properties, that justify its use in the treatment of diabetic hyperglycemia in traditional medicine.

Keywords: Rauwofia vomitoria; diabetes; hypoglycemia; anti-hyperglycemia.

1. Introduction

Diabetes mellitus is a metabolic disorder characterized by chronically elevated blood glucose level linked to a deficiency to either insulin secretion or action of insulin [1, 2]. It is responsible for many complications during its evolution [3, 4]. The prevalence of diabetes is evolving exponentially worldwide and in particular in black Africa where over 20 million people are affected [5]. In the Ivory Coast, according to the International Diabetes Federation (IDF), there were 490200 cases of diabetes in 2014, 5.6 % of adult population with ages between 20 and 79 years [6]. The different drugs used for the treatment of diabetes mellitus, are not always accessible to the population, particularly African. In addition, the drugs showed their limitations given the growing prevalence of diabetes. Even today, these people continue to heal with herbal medicines for the treatment of diabetes mellitus and various other pathologies.

In Ivory Coast, various plants used in traditional medicine for the treatment of diabetes mellitus were listed [7]. *Rauvolfia vomitoria* (Apocynaceae) is also used to treat this disease. It is also used in Ivory Coast for the treatment of fever and infections moisture [8]. Previous work revealed that (70 %) ethanolic extract of root without bark of *Rauvolfia vomitoria* induced a hypoglycaemic effect in normoglycemic rats and an antihyperglycemic effect in rats submitted glucose tolerance test [9].

This study aims to assess the effects of an aqueous extract of roots without bark of *Rauwolfia vomitoria* on blood glucose normoglycemic rats and on glucose tolerance in rats by administration of glucose, and to determine the toxicity and phytochemicals of this extract.

2. Materials and methods

2.1. Biological materiel

2.1.1. Plant materiel

The plant material used consists of boots roots of *Rauwolfia vomitoria* Afzel. (Apocynaceae) obtained in the markets of medicinal plants in Abidjan.

2.1.2. Animals

White rats of the species *Rattus norvegicus* (Muridae), the mass of which is between 120 and 150 g, are used for blood glucose studies. Mouse, *Mus musculus* (Muridae), whose mass is between 20 and 25 g were used for the acute toxicity study. These animals were nursed in the animal physiology laboratory, pharmacology and pharmacopoeia of the Faculty of Natural Sciences of the University Nangui Abrogoua (Abidjan, Ivory Coast) at 25 °C, and under the light day and the darkness night. They were fed with animal food provided by the company IVOGRAIN® Abidjan (Ivory Coast) and have access to water.

2.2. Preparation of the aqueous extract of Rauwolfia vomitoria

The roots of *Rauwolfia vomitoria* (Apocynaceae) are stripped of their bark and dried at room temperature. They are then ground into a fine powder using a grinder. Two hundred grams (200 g) of this powder are mixed with two liters (2 L) of distilled water. The mixture was stirred for 24 hours at room temperature, using a magnetic stirrer. The macerated product is filtered three times on absorbent cotton, and then with filter paper (Whatman n°1 and oven dried at 40 °C. The powder obtained is the aqueous extract of *Rauwolfia vomitoria* (EARv).

2.3 Acute toxicity

50 mice were divided into 10 groups of 5 mice. each mouse of the 9 test batches receive orally 1 ml of a single dose of 5, 50, 100, 300, 1000, 2000, 3000, 4000 or 5000 mg / kg body weight (B.W.) of the aqueous extract *R. vomitoria*. Control group received 1 ml (10 ml/kg B.W.) of distilled water. The number of dead mice was recorded 48 hours after administration of the substance. This study is performed 3 times.

2.4. Activity of aqueous extract of Rauwolfia vomitoria roots (EARv) on rat's blood glucose level

Blood glucose level of the rats was measured using a brand Accu-Cheikh Active (Roche) and test strips. The rats were previously fasted for 12 hours. Their different substances were administered orally.

2.4.1. Effects of EARv on blood glucose level of normoglycemic rats

25 rats were used. They are divided into 5 groups of 5 rats.

- Batch 1: normal glycemic control rats receiving distilled water (10 ml/kg B.W.).

- Batch 2: EARv treated rats with 500 mg / kg B.W.

- Batch 3: EARv treated rats with 700 mg / kg B.W.

- Batch 4: EARv treated rats with 1000 mg / kg B.W.

- Batch 5: rats treated with glibenclamide (reference material) with 10 mg / kg B.W.

Blood glucose is first determined just prior to treatment; it is the initial blood glucose (t0). After the treatment of animals, blood glucose was measured every 30 minutes for 2 hours, and the percent change of blood glucose

level is calculated with respect to the initial glucose.

2.4.2. Effects of EARv during glucose tolerance test

Hyperglycemia was induced in rats by oral administration of glucose at a dose of 4 g/kg B.W.

2.4.2.1. Testing blood glucose in hyperglycemic rats pretreated

30 rats were divided into 6 groups of 5 rats.

- Batch 1: negative control rats receiving distilled water (10 ml/kg B.W.) only.

- Batch 2: positive control rats receiving distilled water (10 ml/kg B.W.), and 30 minutes later, 4 g/kg B.W. of glucose

- Batch 3: rats receiving 500 mg/kg of EARv B.W., then 4 g/kg B.W. of glucose 30 minutes after.

- Lot 4: rats receiving 700 mg/kg B.W. EARv, then 4 g/kg B.W. of glucose 30 minutes after.

Lot 5: rats receiving 1000 mg/kg B.W. of EARv, then 4 g/kg B.W. of glucose 30 minutes after.

- Lot 6: Rats receiving glibenclamide (10 mg/kg B.W.), and then 4 g/kg of glucose B. W. 30 minutes later.

Blood glucose level of rats of each batch was measured just before administration of substances or distilled water and after treatment, at intervals of 30 minutes, 2 hours and 30 minutes. The percentage increase in blood glucose level and the percentage reduction in glucose tolerance were calculated.

2.4.2.2. Testing blood glucose in post-hyperglycemic rats treated with EARv

The protocol was the same as for pretreated rats. However, in this series of experiments, different batches of rats receive doses EARv tests (500, 700 and 1000 mg/kg B.W.) or glibenclamide (10 mg/kg W.B.) 30 minutes after induction of hyperglycemia by oral administration of 4 g/kg glucose PC. Glucose of the rats of each batch was measured just before the glucose administration and thereafter at intervals of 30 minutes for 2 hours and 30 minutes. The percentage increase in blood glucose and the percentage reduction in glucose tolerance were also calculated.

2.5. Phytochemical screening

The detection of secondary metabolites in the aqueous extract of *Rauwolfia vomitoria* was performed with the technique of thin layer chromatography. To do this, ten (10) µL of EARv were placed on chromatographic plates (aluminum sheets coated with silica gel 60 F254 with a thickness of 0.2 mm, Merck). The eluent is composed of chloroform: methanol: water (65: 35: 5). After drying, the chromatograms were revealed with suitable reagents [10] and were observed in visible light, or under UV light at 254 nm or 366 nm.

2.6. Statistical analysis and plotted graphs

Data analysis was done using the software GraphPad INSTAT (San Diego CA USA). Means and standard errors of the means (M ± SEM) were comuted. The difference between two means was evaluated with the test of Student-Newman-Keuls at the level of $\alpha = 0.05$. GraphPad Prism software (San Diego CA USA) was used to draw the graphs.

3. Results

3.1. Acute toxicity

The *Rauwolfia vomitoria* aqueous extract (EARv) administered orally at doses lower than or equal to 5000 mg/kg B.W., causes no mortality in the treated mice.

3.2. Effect of aqueous extract on blood glucose normoglycemic rats and hyperglycemic rats

3.2.1. Dose-response effects of aqueous extract (EARv) and glibenclamide on glucose normoglycemic rats

Changes of blood glucose level in rats after oral administration of EARv at 500, 700 and 1000 mg / kg B.W., or glibenclamide at a dose of 10 mg/kg B. W. are shown in Figure 1. Blood glucose of control rats who received only distilled water, did not vary significantly ($p > 0.05$) throughout the duration of the study (2 hours). It was maintained at 0.97 ± 0.04 g/L. EARv at doses of 500, 700 and 1000 mg/kg B.W., induces a dose-dependent reduction in blood glucose treated rats. These reductions were significant ($p < 0.05$) 30 minutes after treatment rats. These hypoglycemia gradually increased over time and 120 minutes after treatment, blood glucose level reductions were 0.18 ± 0.01 g/L, 0.24 ± 0.02 g/L and 0.30 ± 0.03 g/L, respectively for EARv doses of 500, 700 and 1000 mg/kg B.W., with respective rates of 18.37 % blood glucose reduction, 24.24 % and 30.30 % ($p < 0.001$). Glibenclamide (reference substance), administered at a dose of 10 mg / kg B.W., also significantly decreased blood glucose level ($p < 0.01$) 30 minutes after the treatment of the rats and progressively with time. 120 minutes after the treatment of rats, the decrease of blood glucose was 0.32 g/L, with blood glucose reduction rate of 32.59 ($p <0.001$). The effects of glibenclamide at a dose of 10 mg / kg body weight were more significant than EARv at doses of 500 and 700 mg/kg B.W. However, its effects were not statistically different to those of EARv the dose of 1000 mg/kg B.W. ($p > 0.05$).

3.2.2. Effects of aqueous extract (EARv) and glibenclamide on induced hyperglycemia in rats

3.2.2.1. Change in blood glucose levels in hyperglycemic rats pretreated with EARv or glibenclamide

The results of this study are presented in Figure 2. Glucose of the negative control group consisting of rats given only distilled water, did not vary significantly ($p> 0.05$) throughout the 3 hours of experimentation. It was of the order of 0.98 ± 0.03 g/L.

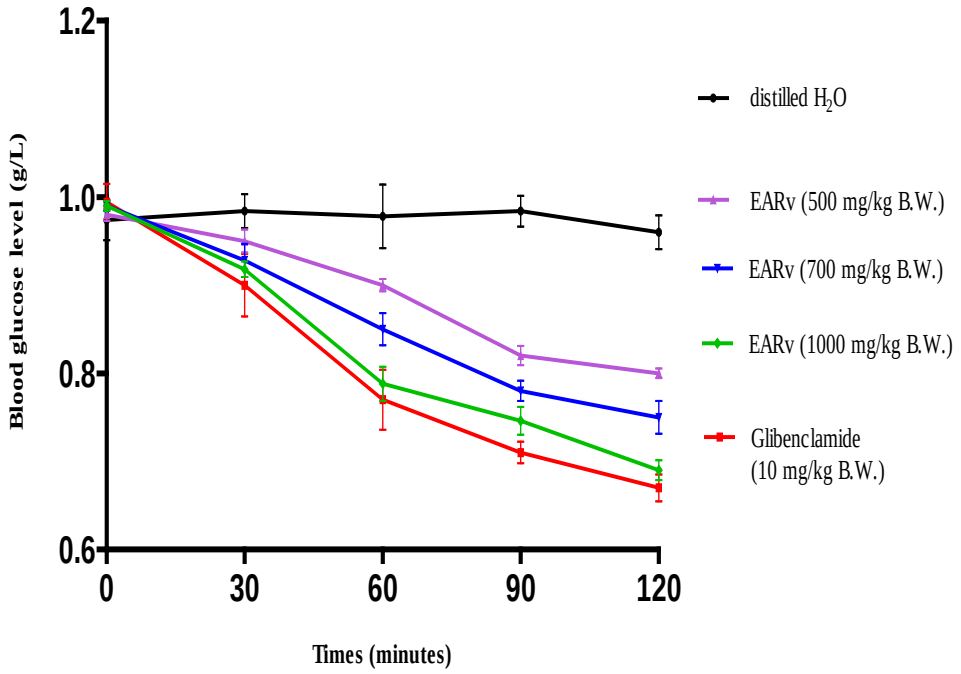

Figure 1: Dose response effects of aqueous extract of *Rauwolfia vomitoria* (EARv) and glibenclamide on glucose normoglycemic rats

Oral administration of glucose at a dose of 4 g/kg B.W. into rats that received or not, 30 minutes before, EARv (500, 700 and 1000 mg/kg B.W.) or glibenclamide (10 mg/kg B.W.) causes an increase in blood sugar of the animals. However, the peak hyperglycemia that occurred 30 minutes after administration of glucose varied depending on whether the rats were pretreated or not with EARv or glibenclamide. During the first 30 minutes after treatment, blood glucose of rats receiving different doses of EARv or glibenclamide did not vary significantly ($p > 0.05$).

In positive control rats (hyperglycemic control), the glucose administration results in an increase of blood glucose, with glucose tolerance peak of the order of 0.53 ± 0.02 g/L. Thereafter, this hyperglycemia is gradually reduced and the basal blood glucose was found 2 hours 30 minutes after the glucose administration. EARv at doses of 500, 700 and 1000 mg/kg B.W., resulted in significant decreases ($p < 0.001$) and dose-dependent induced hyperglycemia following administration of glucose. For these doses EARv the hyperglycemia peaks were respectively 0.40 ± 0.06 g/L, 0.34 ± 0.05 g/L and 0.30 ± 0.03 g/L; corresponding to 24.53 %, 35.85 % and 43.40 % reduction of the induced hyperglycemia, compared to the positive control. These highs are gradually reduced over time. Thus, in the presence of EARv at a dose of 500 mg / kg B.W., the initial blood sugar was found 2 hours 15 minutes after the administration of glucose and thereafter the hypoglycemia estimated at 0.04 g/L was not significant hypoglycemia ($p> 0.05$). When the rats were pretreated with EARv at doses of 700 and 1000 mg / kg BW, hyperglycemia induced by glucose are gradually reduced over time and the initial blood sugar was attained, respectively, 1 hour 45 minutes and 1 hour 38 minutes after the glucose administration. However, hypoglycemia subsequently increased to significant levels with concentrations of 0.19 g/L ($p < 0.05$)

and 0.28 g/L ($p < 0.001$) respectively. Glibenclamide, in the dose of 10 mg / kg B.W., significantly reduced ($p < 0.001$) induced hyperglycemia by the administration of glucose. The hyperglycemia peak was 0.27 ± 0.04 g/L, corresponding to 49.06 % in reductions of hyperglycemia induced by glucose. This hyperglycemia was then gradually reduced and the initial blood glucose was attained 1 hour 30 minutes after the glucose administration. A significant hypoglycemia ($p < 0.001$) also followed to reach a concentration of 0.33 g/L.

Rate reduction hyperglycemia induced by glucose as function of time in rats pretreated with EARv or with glibenclamide are shown in Table 1.

Table 1: hyperglycemia Reduction rate of glucose-induced in rats pretreated with (EARv) or glibenclamide

		Time after administration of glucose				
		30 min	**60 min**	**90 min**	**120 min**	**150 min**
reduction induced hyperglycemia glucose in the dose of 4 g / kg B.W. (%)	**H_2O (hyperglycemic control)**	0	19,01	34,22	55,13	98,86
	EARv (500 mg/kg B.W.)	23,95	45,63	66,92	95,05	112,17
	EARv (700 mg/kg B.W.)	35,74	58,93	80,23	119,39	132,70
	EARv (1000 mg/kg B.W.)	42,96	63,88	94,29	130,42	153,23
	Glibenclamide (10 mg/kg B.W.)	48,67	74,52	105,70	139,92	162,73

3.2.2.2. Change in blood glucose levels in hyperglycemic rats post-treated with (EARv) or glibenclamide

Glucose rats constituting the negative control group (rats given only distilled water) did not vary significantly during the 3 hours of experimentation ($p > 0.05$). It is of the order of 0.96 ± 0.03 g / L.

The administration of glucose at a dose of 4 g/kg in rats P.C. causes hyperglycemia whose peak, which occurs 30 minutes after gavage, is 0.52 ± 0.02 g/L (Figure 3). The administration or not of tests doses of the aqueous extract (500, 700 and 1000 mg/kg B.W.) or glibenclamide (10 mg/kg B.W.) causes a gradual reduction in hyperglycemia, and return time to the initial blood glucose varied depending on the substance or the dose

administered. Table 2 shows the percentage reduction of the hyperglycemia induced by glucose in post-rats treated or not with different doses of EARv or with glibenclamide.

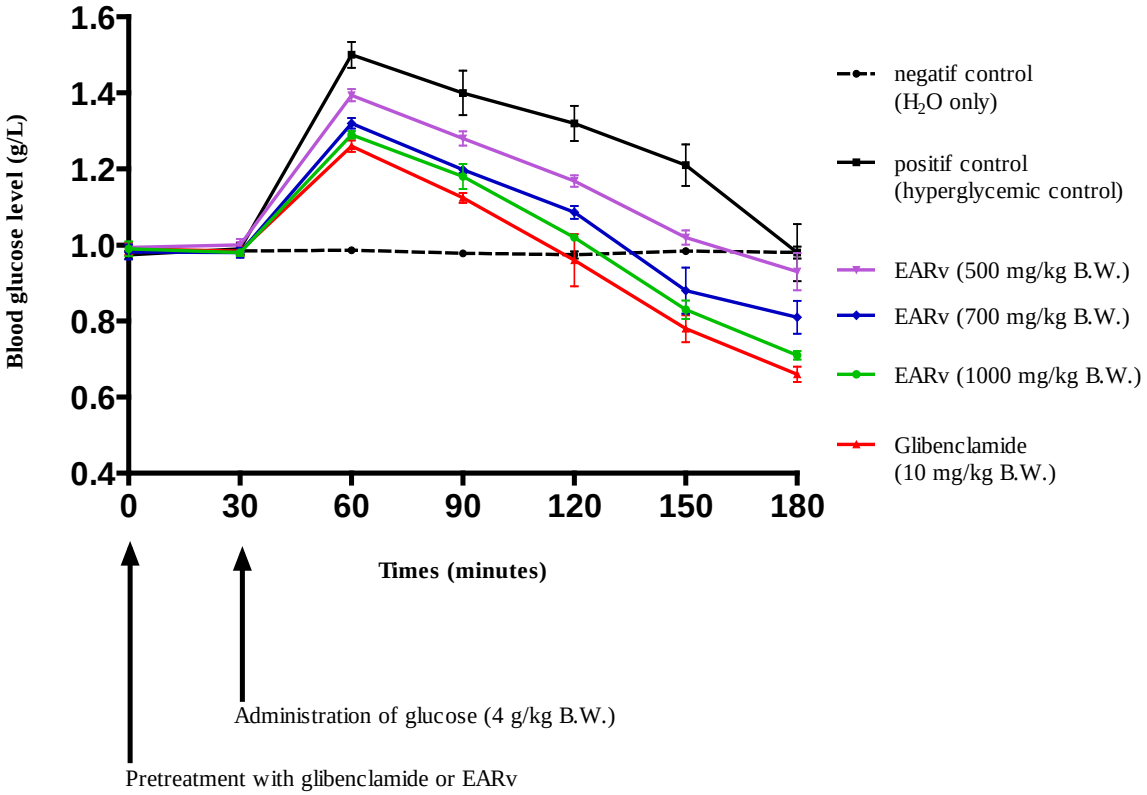

Figure 2: Evolution with time of blood glucose levels in hyperglycemic rats pretreated with the aqueous extract of *Rauwolfia vomitoria* (EARv) or glibenclamide

In positive control Rats (hyperglycemic witnesses), the initial blood glucose level was attained 2 hours and 30 minutes after the administration of glucose.

In rats post-treated with different doses of EARv, reducing hyperglycemia and time to reach to the initial glucose levels were found to be dose-dependent. Indeed, in rats treated with EARv at doses of 500, 700 and 1000 mg/kg B.W., the time to reach the initial blood glucose level was 150, 130 and 115 minutes respectively and hypoglycemia subsequently occurred at the respective concentrations of 0.10 ($p < 0.01$), 0.20 ($p < 0.01$) and 0.26 g/L ($p < 0.001$).

Also, glibenclamide (10 mg/kg B.W.), administered to rats 30 minutes after that of glucose results in greater reduction of induced hyperglycemia and initial blood glucose was found at 105 minutes after the glucose administration. Subsequently, hypoglycemia occurred with 0.29 g/L ($p < 0.001$).

Table 2: hyperglycemia Reduction rate of glucose-induced in rats post-treated with (EARv) or glibenclamide

		Times after administration of glucose					
		30 min	**60 min**	**90 min**	**120 min**	**150 min**	**180 min**
reduction induced hyperglycemia glucose in the dose of 4 g / kg B.W. (%)	**H_2O (hyperglycemic control)**	0	18	34,8	60,8	95,2	100,8
	EARv (500 mg/kg B.W.)	0	19,54	46,36	82,76	101,91	119,92
	EARv (700 mg/kg B.W.)	0	32,81	61,39	96,52	121,61	138,61
	EARv (1000 mg/kg B.W.)	0	37,31	70,52	110,07	135,44	148,88
	Glibenclamide (10 mg/kg B.W.)	0	41,13	84,15	126,41	146,03	154,7

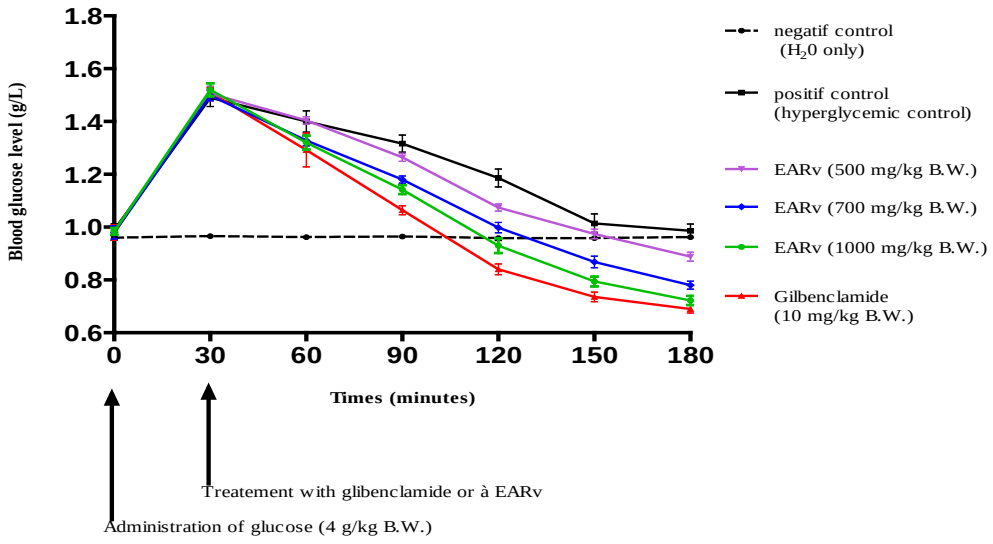

Figure 3: Evolution with time of blood glucose levels in hyperglycemic rats post-treated with aqueous extract of *Rauvolfia vomitoria* (EARv) or glibenclamide

3.3. Phytochemical screening

Performed Thin Layer Chromatography revealed the presence of polyphenols, monoterpenoids, flavonoids, anthraquinones and anthrones, alkaloids, tannins catechin and saponins in the aqueous extract of *Rauvolfia vomitoria*.

4. Discussion

Aqueous extract of *Rauvolfia vomitoria* administered orally at doses up to 5000 mg/kg B.W., did not result in the death mice. This indicates that the 50 % lethal dose (LD50) of EARv was above 5000 mg/kg B.W., so that this extract is non-toxic when administered orally [11]. These results corroborate those of Amole et al. [12] and N'Doua et al. [9] which respectively have shown that the aqueous extract of the leaves of *Rauvolfia vomitoria* and (70 %) ethanolic root extract of this plant are not toxic when taken orally.

Aqueous extract of *Rauvolfia vomitoria* the, at doses of 500, 700 and 1000 mg/kg B.W., caused dose-dependent reduction in blood glucose level normoglycemic rats. It also vginduced the reduction of hyperglycemia caused by oral administration of glucose in rats in treatment and in rats post-treated with the extract. This indicates that *Rauvolfia vomitoria* was hypoglycemic and antihyperglycemic. these properties of *Rauvofia vomitoria* were also revealed with (70 %) ethanolic extract of root of this plant [9]. However, the aqueous extract of *Rauvofia vomitoria* is more active than the (70 %) ethanol extract and the effects of the aqueous extract at a dose of 1000 mg/kg B.W. were substantially identical with those of glibenclamide at a dose of 10 mg/kg B.W. Glibenclamide is a substance belonging to the sulfonamide, recognized for their hypoglycemic and antihyperglycemic activities [9, 13, 14, 15]. Sulfonylureas cause hypoglycemia in normoglycemic rats by stimulating the production of insulin by the pancreatic beta cells, which promotes the storage of glycogen in the liver [16, 17, 18, 19]. The decrease of hyperglycemia induced rats treated with EARv could be explained by the stimulation of insulin secretion by the pancreas and/or, possibly, by an increase in peripheral glucose utilization in the presence of extract.

The presence of substances such as flavonoids, polyphenols, saponins and catechin tannin in the *Rauvolfia vomitoria* aqueous extract are the cause of the observed hypoglycemic and anti-hyperglycemic effects. Indeed, hypoglycemic and anti-hyperglycemic properties of these secondary metabolites have been reported by several authors [20, 21, 22].

5. Conclusion

This study shows that the aqueous extract of roots without bark *Rauvolfia vomitoria* is non-toxic when taken orally and has a high potential hypoglycemic and anti-hyperglycemic substance. These properties of the aqueous extract of *Rauwolfia vomitoria* justify the use of this plant in traditional medicine in the treatment of diabetic hyperglycemia.

References

[1] D. Raccah. Epidémiologie et physiopathologie des complications dégénératives du diabète sucré. *EMC-Endocrinologie*., 1 : 29-42, 2004.

[2] K. Gariani, I. Hagon-Traub, J. Philippe. Diabète de type 1 ou 2 ? ou autre ? *Revue Medecine Suisse*., 5 : 1248-1253, 2009.

[3] D. Chevenne and D. Porquet. Génétique et critères diagnostiques du diabète sucré. *Annales de Biologie Clinique*., 57 : 427-435, 1999.

[4] Canadian Diabetes Association. Canadian Diabetes Association 2003 clinical practice guidelines for the prevention and management of diabetes in Canada. *Canadian Journal of Diabetes*., 27 : S1-S2, 2003.

[5] IDF. IDF Diabetes Atlas, Sixth edition, 2013, pp 15-17.

[6] IDF, 2014. IDF Africa, Diabetes in Cote d'Ivoire - 2014. www.idf.org/ membership/afr/Cote-d-ivoire, 2000* [Jun, 10, 2015].

[7] F. H. Tra Bi, G. M. Irié, K. C. C. N'gaman, C. H. B. Mohou. Etudes de quelques plantes thérapeutiques utilisées dans le traitement de l'hypertension artérielle et du diabète : deux maladies émergentes en Côte d'Ivoire. *Sciences & Nature*., 5 : 39-48, 2008.

[8] G. H. Schmelzer and A. Gurib-Fakim. Ressources végétales de l'Afrique tropicale 11 (1). Plantes médicinales 1. Wageningen, Pays-bas, Fondation PROTA, 2008, 869 p.

[9] L. A .R. N'doua, K. J. C. Abo, S. Aoussi, M. Gbogbo, A. P. Yapo, E. E. Ehile. Effets hypoglycémique et antihyperglycémique de l'extrait éthanolique (70 %) de racines de *Rauvolfia vomitoria* Afzel. (Apocynaceae). *Eupropean Scientific Journal*., 11 : 176-190, 2015.

[10] H. Wagner and S. Bladt, 2001. Plant drug analysis. A thin layer chromatography atlas. Springer, 384 p.

[11] E. G. C. Clarke and M. L. Clarke. Veterinary Toxicology. London, England: Cassel and Colier Macmillan Publishers, 1977, pp 268-277.

[12] O. O. Amole, O. K. Yemitan, K. A. Oshikoya. Anticonvulsant activity of *Rauvolfia Vomitoria* (Afzel). *African Journal of Pharmacy and Pharmacology*., 3 : 319-322, 2009.

[13] T. Amalraj and S. Ignacimuthu. Evaluation of the hypoglycaemic effect of *Memecylon umbellatum* in normal and alloxan diabetic mice. *Journal of Ethnopharmacology*., 62 : 247-250, 1998.

[14] G. Y. Sy, A. Cisse, R. B. Nongonierma, M. Sarr, N. A. Mbodj, B. Faye. Hypoglycaemic and antidiabetic activity of acetonic extract of *Vernonia colorata* leaves in normoglycaemic and alloxan-induced diabetic rats. *Journal of Ethnopharmacology*., 98 : 171-175, 2005.

[15] B. N. Gnangoran, B. B. N'guessan, P. Amoateng, K. Dosso, A. P. Yapo, E. E. Ehile. Hypoglycaemic activity of ethanolic leaf extract and fractions of *Holarrhena floribunda* (Apocynaceae). *Journal of Medical and Biomedical Sciences*., 1 : 46-54, 2012.

[16] J. E. Jackson and R. Bressler. Clinical pharmacology of sulphonylurea hypoglycemic agents. Part I. *DRUG*., 212 : 211-245, 1981.

[17] R. S. Somani and A. K. Singhai. Hypoglycaemic and antidiabetic activities of seeds of *Myristica fragrans* in normoglycemic and alloxan-induced diabetic rats. *Asian Journal of Experimental Sciences*., 22 : 95-102, 2008.

[18] K. J. Yosadha, K. N. Jayaveera, R. K. Ravindra, K. Rupesh, D. Raghavendra. Anti-diabetic activity of aqueous extract of *Talinum cuncifolium* in rats. *Pharmacologyline*., 2: 198-206, 2008.

[19] T. Gebreyohannis, W. Shibeshi, K. Asres. Effects of solvent fractions of *Caylusea abyssinica* (Fresen.) Fisch. & Mey. on blood glucose levels of normoglycemic, glucose loaded and streptozotocin-induced diabetic rodents. *Journal of natural remedies*., 14 : 67-75, 2014.

[20] G. Sy, F. S. Barbosa, A. Wele, P. M. Gueye, C. D. Gueye , A. Cisse, A. M. Dieye, E. Bassene, B. Faye. Activité anti-hyperglycemiante de la fraction f2 de l'extrait total acetonique de feuilles de *Vernonia colorata* (Composeae). *Pharmacopée et Médecine Traditionnelle Africaines*., 15 : 6-10, 2008.

[21] M.S. Deutschländer, N. Lall, M. Van de Venter, S. Dewanjee. The hypoglycemic activity of *Euclea undulata* Thunb. var. myrtina (Ebenaceae) root bark evaluated in a streptozotocin-nicotinamide induced type 2 diabetes rat model. *South African Journal of Botany*., 80 : 9-12, 2012.

[22] D. Chabane, F. Saidi, A. Rouibi, K. Azine .Activité hypoglycémique de l'extrait aqueux d'*Ajuga iva* L. Schreber chez les rats diabétiques induite par l'alloxane. *Afrique SCIENCE*., 09 : 120-127, 2013.