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Effect of the aqueous extract of the Debarked roots of *Rauvolfia vomitoria* (EARv) AFZEL (Apocynaceae) a plant with antidiabetic potential on the lipid profile following daily administration for six months in albino Wistar rats

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ABSTRACT

Diabetes, considered a major global public health issue, is often associated with dyslipidemia. Given the limitations of conventional treatments, people are showing increasing interest in medicinal plants. Thus, *Rauvolfia vomitoria* has several pharmacological properties, including antihypertensive and potentially antidiabetic effects. This study aims to evaluate the effect of repeated oral administration of EARv on the lipid profile over a six-month treatment period. Rauvolfia vomitoria roots were harvested, stripped of their bark, and then processed into an aqueous extract by maceration. Eighty healthy rats were divided into four groups (T, A, B, C) of 20 rats each, with 10 males and 10 females per group. Rats in group T received distilled water, while those in groups A, B, and C received EARv extract at doses of 500, 700 mg/kg body weight (b.w), and 1,000 mg/kg b. w, respectively, administered orally for six months at a rate of 2 mL/100 g b.w daily. Using serum from blood samples collected to measure total cholesterol, HDL cholesterol, and triglycerides. The results show that the aqueous extract of Rauvolfia vomitoria significantly reduced total cholesterol levels in a dose-dependent manner; it also led to an increase in HDL cholesterol levels and a significant reduction in triglycerides. EARv could represent an interesting therapeutic alternative for the management of diabetes and its metabolic complications.

Keywords: Aqueous extract of debarked roots- *Rauvolfia vomitoria*- Lipid profile-Cholesterols - Triglycerides

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INTRODUCTION

Diabetes is now a major global public health problem. This chronic metabolic disorder is characterized by persistent hyperglycemia resulting from impaired insulin secretion, insulin resistance, or both [1]. This condition affects nearly 590 million people worldwide [2] and is among the top 10 causes of death globally. In fact, diabetes is generally ranked around 8th [3]. It is often associated with serious complications, particularly dyslipidemia, which manifests as an abnormal lipid profile, including elevated levels of triglycerides, total cholesterol, and low-density lipoproteins (LDL), as well as decreased levels of high-density lipoproteins (HDL These lipid abnormalities are significant risk factors for cardiovascular disease in patients with diabetes [4].

Given the limitations of conventional treatments particularly their high cost, side effects, and sometimes limited availability in developing countries interest in herbal medicine continues to grow [5]. Many medicinal plants are used in traditional medicine for their blood-sugar-lowering and cholesterol-lowering properties (6,7). Among these, *Rauvolfia vomitoria* Afzel., a member of the Apocynaceae family, plays an important role in African pharmacopoeia. This plant is recognized for its various pharmacological properties, including antihypertensive, sedative, and potentially antidiabetic effects (8,9).

The roots of *Rauvolfia vomitoria*, rich in active compounds, are a focus of biomedical research. Thus, the hypoglycemic and antihyperglycemic properties of aqueous root extracts (EARv) from this plant prepared without the bark have been demonstrated by N'Doua L. and her collaborators (10). A chronic toxicity study was conducted to demonstrate that EARv is well-tolerated based on hematological parameters (11) and certain functional markers of liver and kidney function (12) However, few studies have thoroughly explored the effect of the aqueous extract of barkless roots on the lipid profile, particularly in the context of prolonged administration.

Thus, the present study aims to evaluate the effect of the aqueous extract of the barkless roots of *Rauvolfia vomitoria* (EARv) on the lipid profile in albino Wistar rats following daily administration over a six-month period. This investigation could contribute to the development of this plant as a therapeutic alternative in the management of diabetes and its metabolic complications.

MATERIALS AND METHOD

Plant material

The roots of *Rauvolfia vomitoria* were collected in December 2016 in Padiégnan, in the Yakassé Féyassé subprefecture of the Abengourou department, 210 km from Abidjan (Ivory Coast); a sample was provided by N'Doua L. and her collaborators (10). The plant was identified and

authenticated at the National Floristic Center (C.N.F.) of Félix HOUPHOUËT-BOIGNY University in Cocody-Abidjan under the reference number N°UCJ002178

Animal Material

Healthy male and female (nulliparous and not pregnant) albino rats of the species Rattus norvegicus, Wistar strain, aged 6 to 8 weeks and weighing between 80 and 100 g, were used for the tests (13). These animals came from the animal facility at the Laboratory of Physiology, Pharmacology, and Pharmacopoeia at NANGUI ABROGOUA University. Their diet consisted of IVOGRAIN® pellets, and they had continuous access to tap water in drinking bottles. The experimental protocol and animal handling procedures were conducted in accordance with good laboratory practices (13).

Preparation of an aqueous extract from the roots of *Rauvolfia vomitoria*

The extracts were prepared according to the extraction method proposed by N'Doua et al. (2016). The roots, with their bark removed, were cut into small pieces, crushed in a mortar, and then dried at room temperature (25 °C ± 2) for one week. They were then pulverized using an electric grinder (Culatti, France). A maceration was carried out using a magnetic stirrer, involving 200 g of powder in 2 liters of distilled water for 24 hours. The macerate was filtered three times through cotton wool and Wattman No. 1 filter paper, then dried in an oven at 40 °C for 48 hours. The resulting 13.5 g of powder, constituting the aqueous extract of *Rauvolfia vomitoria* (EARv), was stored in a refrigerator at -10 °C for 72 hours (10).

Chronic Toxicity Study

OECD Guideline 452 (13) was used to conduct the chronic toxicity study. Thus, 80 healthy rats were divided into six groups of 20 rats each for groups T, A, B, and C. Each group contained an equal number of males and females. All animals were administered a solution volume of 2 mL/100 g b.w via gavage using a cannula once daily starting at 8 :00 a.m. for 6 months or 180 days. Group T received distilled water; groups A and B received the aqueous extract (EARv) at doses of 500 and 700 mg/kg b.w, respectively; and group C received EARv at a dose of 1,000 mg/kg b.w (13).

Blood Tests

Blood samples were collected from the retroorbital sinus of the rats using the method described by Waynforth (14) at each time point corresponding to the different toxicity studies: after 30 days for subacute toxicity; after 90 days for subchronic toxicity; and after 180 days for chronic toxicity. On the day before each blood draw, the animals were fasted from 8:00 p.m. until 7:00 a.m. the following day. The rats were anesthetized with COOPER ether, and venous blood was

then collected from the retro-orbital sinus using a sterile Pasteur pipette. Approximately 0.5 to 2 mL of blood was collected in dry tubes. The blood was then centrifuged at 3,000 revolutions per minute for 10 minutes using a centrifuge (HERAEUS SEP A TECH). Serum biochemical parameters were measured using a ROPONIK® automated analyzer with reagents from Laboratorio BioSystems Costa Brava (Spain). Total cholesterol levels were measured according to the method of Allain and his collaborators (15); HDL cholesterol levels according to the method of Grove (16); and triglyceride levels according to the method of Mac Gowan and his collaborators (17).

Statistical Analysis

The data were analyzed using GraphPad Prism 8.0.1 software (244). The results are expressed as the mean followed by the standard error of the mean (M ± SEM). The difference between the mean values of the liver and kidney parameters was determined using a two-way analysis of variance (ANOVA 2) to verify the normality of the variables within each group and the homogeneity of variances. These tests established a significance level of $p < 0.05$. In the presentation of the results, asterisks (*, **, ***) reported significant declines.

RÉSULTS AND DISCUSSION

Effect of EARv on Total Cholesterol Levels

Compared with male and female control rats, total serum cholesterol levels decreased significantly ($p < 0.05$; $p < 0.01$; $p < 0.001$) and in a dose-dependent manner throughout the study in all male and female rats treated with EARv (Figure 1)

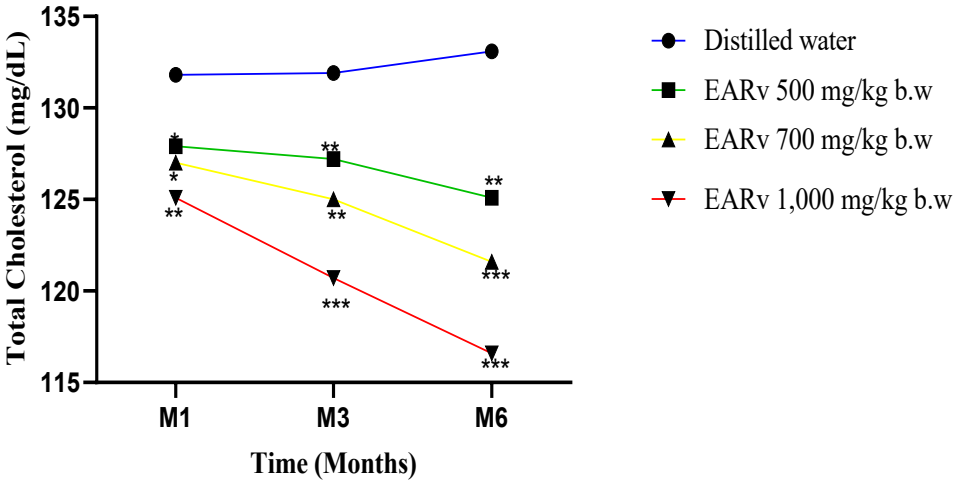
Effect of EARv on HDL-Cholesterol Levels

There was a significant increase in HDL-cholesterol levels ($p < 0.05$) in both male and female rats treated with EARv at doses of 700 and 1,000 mg/kg b.w. This effect persisted very significantly at the third and sixth months at doses of 500 ($p < 0.01$); 700 and 1,000 mg/kg b.w ($p < 0.001$) (Figure 2)

Effect of EARv on Triglyceride Levels

Triglyceride levels were significantly lower in all treated rats compared with the controls. In fact, at month 1, this decrease was observed at 700 and 1,000 mg/kg b.w ($p < 0.05$; $p < 0.001$); and at months 3 and 6 at all doses ($p < 0.05$; $p < 0.01$; $p < 0.001$). Furthermore, triglyceride levels followed virtually the same trend throughout the study at doses of 700 and 1,000 mg/kg body weight (Figure 3).

A



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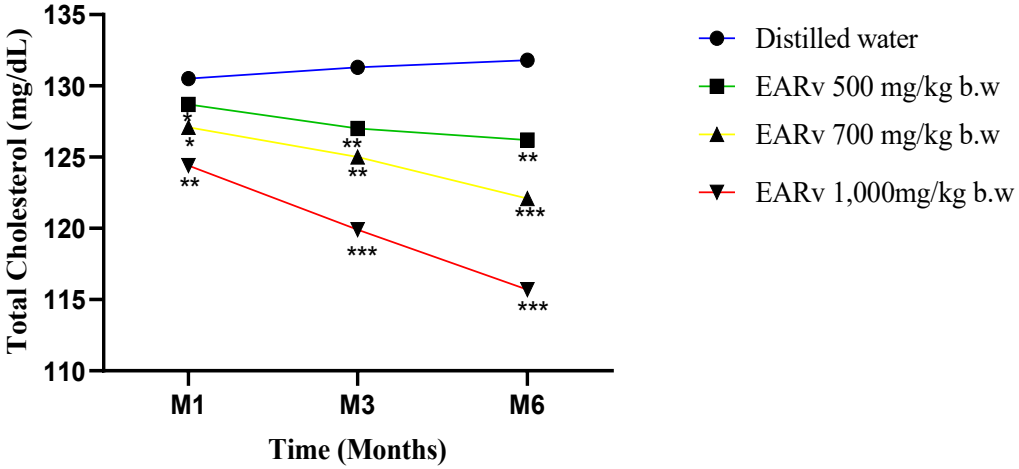
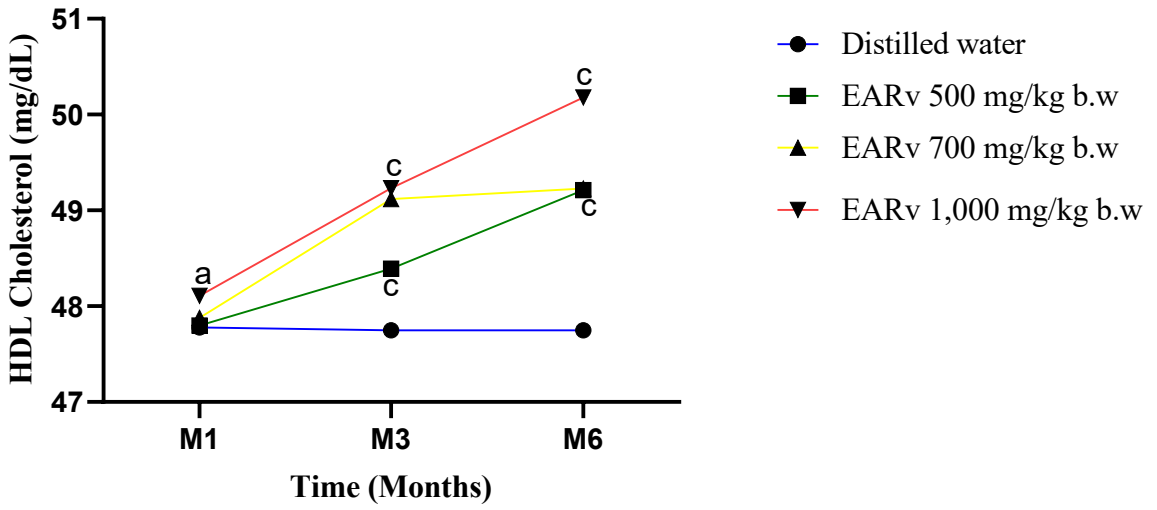


Figure 1: Curves showing changes in total cholesterol levels over time following administration of Earv to male and female rats

A : male rats ; B : female rats. Each bar represents the mean ± standard error of the mean (M ± SEM) ; M1, M3, and M6 = 1, 3, and 6 months of administration ; n = 10 rats ; * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$

A



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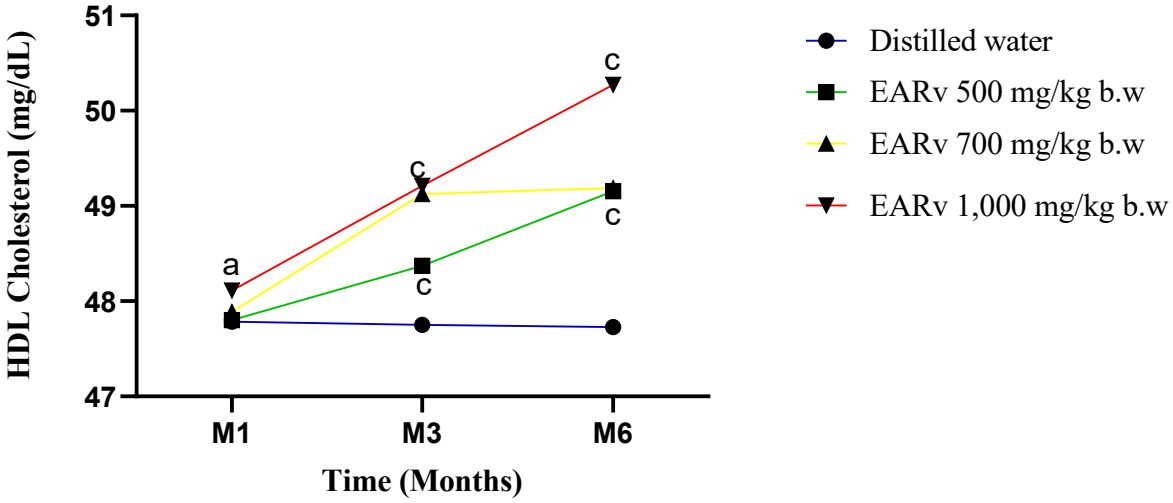

Figure 2: Changes in HDL cholesterol levels following administration of EARv in male and female rats

A: in male rats; B : in female rats. Each bar represents the mean ± standard error of the mean (M ± SEM); M1, M3, and M6 = 1, 3, and 6 months of administration; n = 10 rats ; a = $p < 0.05$; b = $p < 0.01$; c = $p < 0.001$

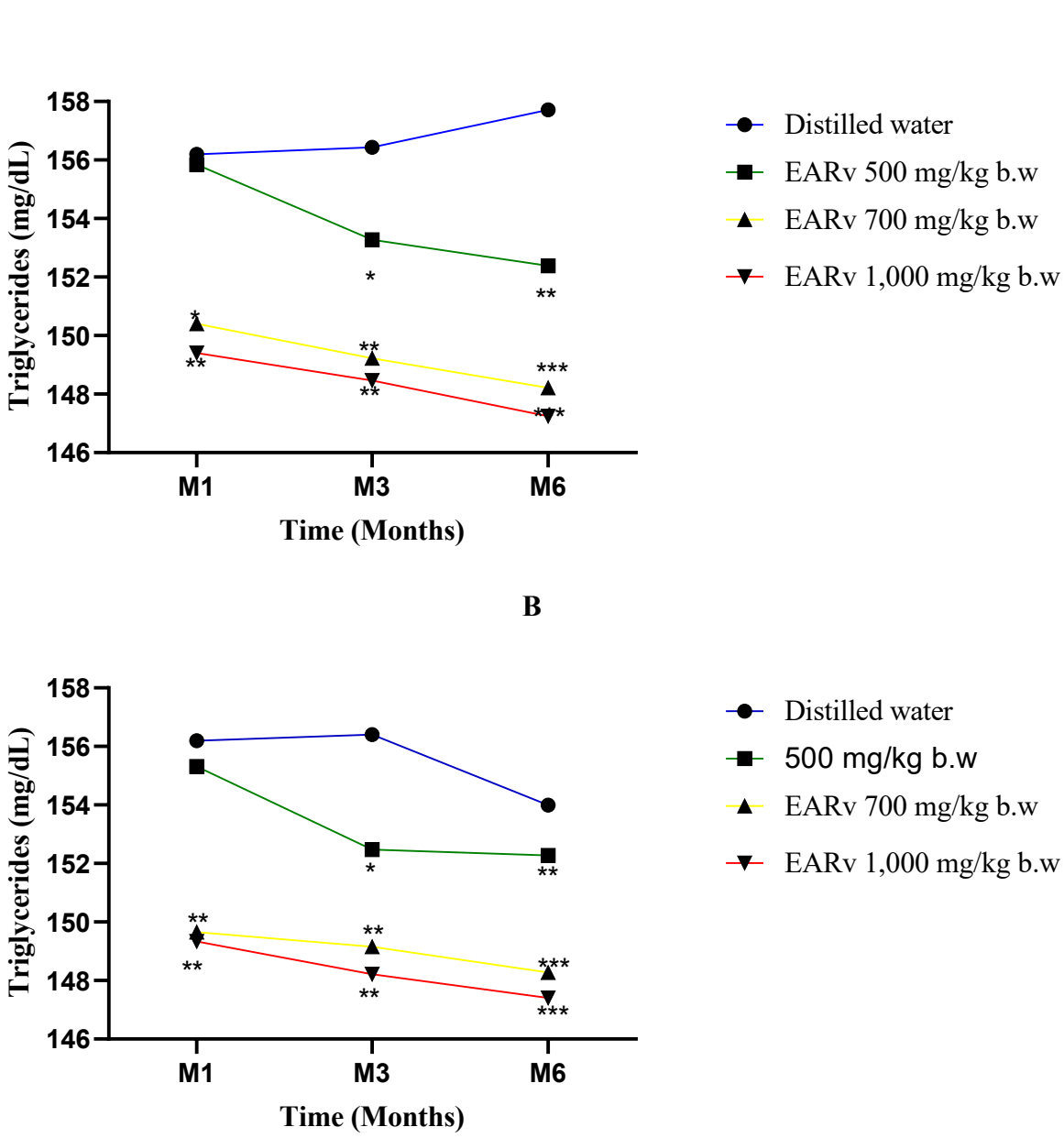

Figure 3: Triglyceride level change curves following administration of EARv in male and female rats

A: in male rats; B : in female rats. Each bar represents the mean ± standard error of the mean (M ± SEM) ; M1, M3, and M6 = 1, 3, and 6 months of administration ; n = 10 rats ; * = $p < 0.05$; ** = $p < 0.01$; *** = $p < 0.001$

DISCUSSION

EARv caused a decrease in total cholesterol and triglyceride levels that was dose-dependent and duration-dependent, while HDL cholesterol levels increased throughout the study. Diabetes is most often associated with obesity and therefore leads to cardiovascular complications. These diseases are caused by disturbances in lipid metabolism (18). Assessing lipid profiles is of

paramount importance in the prevention and treatment of diabetes. Changes in the levels of total cholesterol, HDL cholesterol, LDL cholesterol, and triglycerides can provide useful information about lipid metabolism and the heart's susceptibility to atherosclerosis and associated coronary heart disease, which are among the complications of diabetes (19). Elevated total cholesterol and triglyceride levels are major risk factors for cardiovascular disease. Conversely, an increase in HDL cholesterol has a cardioprotective effect (20). Consequently, EARv may promote fatty acid storage, which would be beneficial in preventing diabetes and, if the disease does occur, in its treatment and in preventing complications such as coronary heart disease and atherosclerosis (21). Our findings are consistent with those reported by Zhu and collaborators (22), who demonstrated a significant reduction in total cholesterol and LDL cholesterol, as well as an improvement in triglyceride levels, with Plantago ovata (psyllium). however this result differs from that observed in the study by Koné and collaborators (23), which showed that daily administration of an aqueous extract of Sacoglottis gabonensis (Humiriaceae) for 28 days to healthy rats did not result in any changes in the lipid profile.

CONCLUSION

EARv may represent a promising therapeutic alternative in the management of diabetes and its metabolic complications, particularly lipid disorders, by lowering total cholesterol levels; increasing HDL cholesterol levels; and significantly reducing triglyceride levels.

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