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*RESEARCH ARTICLE*

# EFFECTS OF AQUEOUS EXTRACT OF *DISSOTIS BRAZZEÏ* COGN. ROOTS ON BIOLOGICAL PARAMETERS AND RISK FACTORS OF MYOCARDIAL INFARCTION IN NORMAL RATS

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## *Abstract*

The present study aimed to evaluate the effects of the aqueous extract of the roots of *Dissotis brazzeï*Cogn.*(D. brazzeï)*, on the parameters of myocardial infarction (MI) and its risk factors in normal rats. Among the cardiac pathologies cited by the population as heartache is the myocardial infarction. *D.Brazzeï*is a medicinal plant widely used in traditional Congolese medicine to treat heart diseases. In this study, rats were given the aqueous extract of the roots of *D. brazzeï*(125, 250 and 500 mg/kg/day)for 7 days in, comparison with rats treated with distilled water (1 mL/100 g). The results obtained show that this extract has no significant effects on MI parameters (LDH, CK-MB, troponin I and CRP). Moreover, it causes variations at non-risk percentages of MI risk factors (blood pressure, body weight of rats, total cholesterol, HDL-C and Triglycerides). In conclusion, the aqueous extract of the roots of *D. brazzeï* at the studied doses, does not modify the parameters of MI, nor does it promote the occurrence of the risk factors of MI. The evaluation of a cardioprotective study of this extract deserves to be carried out in rats.



## Introduction:-

Commonly referred to as heart attack, myocardial infarction (MI) means necrosis of the myocardium due to insufficient oxygen supply. It is a common clinical presentation of ischemic heart disease (Anderson & Morrow, 2017) and is the primary cause of morbidity and mortality worldwide (Ghazouani et al.,2019 ; Thygesen et al., 2012). It is a major public health issue. Indeed, increased myocardial necrosis causes cardiac dysfunction affecting blood pressure, heart rate, electrocardiographic changes as well as left ventricular dysfunction associated with altered activity of specific cardiac enzymes (Daoud et al., 2017). The treatment of MI involves various synthetic molecules with high costs and generally inaccessible to populations, the majority of which, especially in Africa, are poor. Thus, African populations are turning to traditional pharmacopoeia and are increasingly resorting to medicinal plants to treat themselves. Africa has a rich and diversified flora of medicinal plants and among them is D. brazzeï, a plant of the Melastomataceae family. The juice of the leaves or the decoction of the roots of this plant is reputed as a remedy for the treatment of heartache (Bouquet, 1969). In the perspective of the valorization of the Congolese medicinal plants used in the treatment of heartache, the present study aims to evaluate the effects of the aqueous extract of the roots of D. brazzeï on the parameters of the myocardial infarction (MI) and its risk factors.

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## Material and Methods:-

### Plant material

The roots of D. brazzeï collected in Madibou (southern district of Brazzaville-Congo) in February 2020, were used. The species was identified and compared to the reference sample n° 9177 of the herbarium of theNational Research Institute in Exact and Natural Sciences(IRSEN) of Brazzaville, by Doctor Jean-Marie MOUTSAMBOTE, Master of Conferences CAMES. The roots of this plant were harvested, cleaned and dried in the sun at a temperature of 29 ± 1 °C for two weeks.

### Animals

Male Wistar rats, 16-18 weeks old, with body weight between 130-160 g and male Swiss mice with body weight between 20-30g, 8 weeks old were used. These animals were provided by the animal house of the Faculty of Science and Technology of the University Marien NGOUABI where they are raised at a temperature of 29 ± 1 °C, at a circadian cycle of 12 h of light and 12 h of darkness with free access to food and tap water.

### Preparation of the aqueous extract

The aqueous extract of the roots of D. brazzeï was prepared by decoction by boiling 50 g of powder of this plant in 500 mL of distilled water for 15 minutes. After filtration, the filtrate obtained was evaporated using a flask heater set at 70 °, which gave a dry extract. This dry extract was solubilized in distilled water and administered to the animals at the given doses.

### Evaluation of the effects of D. brazzeïon biochemical parameters of myocardial infarction in wistar rats and hematological parameters

Normal wistar rats weredivided into 4 groups of 5 rats each. Group 1 (control) received distilled water at 1 mL/100 g bw/d, and groups 2, 3, and 4 were treated with the aqueous extract at 125, 250, and 500 mg/kg bw/d, respectively, for 7 days. On the 7th day of experimentation, all animals received distilled water (1 mL/100 g, i.p), 1 h after the last administration of the products, according to the protocol inspired by Bouagnon et al., (2015) on cardiotoxicity induced in rats with doxorubicin. 24 h later, the animals were anesthetized with diethyl ether, and the blood from the retro-orbital vein collected in the dry tubes was used for the assays of biochemical parameters of myocardial infarction (troponin I and CRP by qualitative methods and CK-MB, LDH, and ASAT by quantitative methods) and in the heparin tubes for the assay of hematological parameters.

### Evaluation of the effects of D. brazzeï, on the risk parameters of myocardial infarction in wistar rats

### On the body weight of normal rats

The effect on body weight consisted in measuring the weight of the above treated rats before treatment (D0) with the different products (distilled water and aqueous extract) and at the end of the treatment (D7).

### On the lipid profile of normal rats

The effect on lipid parameters consisted in the determination of total cholesterol, HDL-C and triglycerides from the blood collected in the dry tubes of the above treated rats.

### On mass and heart parameters of normal rats

After collection of blood from the above-treated rats, each rat was placed in the supine position and its four legs secured with the needles on a cork board. After dissection, the heart was removed and then weighed, and the left ventricle was also weighed after the right ventricle and atria were removed. Finally, the ratio of heart mass to left ventricle was calculated to determine the left ventricular index (LVI).

### On diastolic blood pressure (DBP) in normotensive rats

Blood pressure was measured by the invasive method using the Biopac Student Lab recorder type MP 36 as described by Santos et al., (2007), Etou Ossibi et al., (2010).In the present study, twelve (12) other rats divided into four (4) groups of three (3) rats each were used. Group 1 (control) received physiological water of 0.9% NaCl, i.v. at 0.1 mL/100 g bw; batches 2, 3 and 4 (aqueous extract) at 5, 10 and 20 mg/Kg; i.v. at 0.1 mL/100 g bw, respectively.

### Statistical Analysis

The Student-Fischer "t" test was used. The observed difference between the means is significant when the value of "t" calculated in absolute value is, greater than the value of "t" read from the Student-Fischer "t" table for a ddl = n1 + n2 - 2 and the risk of first kind of 5%.

## Results:-

**Effects of aqueous extract of D. brazzeïroots on biochemical parameters of myocardial infarction in normal rats**

Table I shows that troponin and CRP tests are negative with the aqueous extract of D. brazzeïroots at the three doses studied as for distilled water. The results presented in Table II show lower levels of CK-MB and LDH in rats treated with aqueous extract of D. brazzeï roots at the dose of 125 mg/kg (12.95 ± 3.040 and 292.2 ± 134.21) compared with those treated at the doses of 250 and 500 mg/kg as well as those treated with distilled water. Similarly, ASAT activity was significantly lower ($p < 0.001$) in rats treated with aqueous extract of D. brazzeï at different doses (125 ;250 ; 500 mg/kg) than in control rats (distilled water).

**Effects of aqueous extract of D. brazzeïroots on hematological parameters influenced by myocardial infarction in normal rats**

The effect of aqueous extract of D. brazzeï roots on hematological parameters influenced by myocardial infarction in normal rats is presented in Table III. These results show a significant increase in red blood cells with a value of 7.58 ± 0.01; $p < 0.01$) ; white blood cells to 8.70 ± 1.34; $p < 0.001$ ; hemoglobin to 13.14 ± 0.39; $p < 0.001$; blood platelets to 1196.8 ± 74.46; $p < 0.01$; lymphocytes to 5.28 ± 1.32; and MHCC to 32.84 ± 0.27; $p < 0.01$ compared with the control lot. In contrast, a decrease in red blood cell count to 6.17 ± 0.30; $p < 0.05$ and an increase in MHCC to 32.8 ± 0.19; $p < 0.05$ at 500 mg/kg compared to control were noted.

**Effects of D.brazzeï on risk parameters of myocardial infarction in normal wistar rats**

**Effects on the weight evolution of normal rats**

Table IV, shows the weight evolution of normal rats treated with different doses of the aqueous extract of the roots of D.brazzeï(125 ,250 and 500mg/kg) and with distilled water. The administration of the extract caused from D0 to D7 a weight change of 10.94% at the 500 mg/kg dose ; 14.14% at the 125 mg/kg dose and 17.7% at the 250 mg/kg dose against a change of 2.13% in rats given distilled water.

**Effects on Lipid Profile of Normal Rats**

The results recorded in Table V show significantly elevated HDL-cholesterol levels ($p < 0.01$) at 60.96 ± 2.18 and 64.74± 1.17 at the respective doses of (125 and 250 mg/Kg) compared with the control lot. In addition, total cholesterol and triglyceride levels were lower in animals treated with the extract at the different doses compared to those treated with distilled water.

**Effects on mass and heart parameters of normal rats**

From Table VI, it appears that the aqueous extract of D.brazzeï roots at the doses studied does not cause significant variations in the parameters measured.

**Effects on diastolic blood pressure (DBP) in normotensive rats**

The results of the evaluation of the effect of the aqueous extract of the roots of D.brazzeïat different doses (5 ;10 ; 20 mg/kg), on the diastolic blood pressure are presented in table VII. The i.v. administration of this extract at doses of 5, 10 and 20 mg/kg in normal rats causes an increase in DBP of 19.25; 3.83 and 1.65 mm Hg respectively against the increase of 1.84 mm Hg in NaCl treated rats (control lot).

## Discussion:-

Myocardial infarction, characterized by cardiac injury can be assessed through a change in blood serum levels of creatine kinase-muscle/brain (CK-MB), lactate dehydrogenase (LDH), Troponin-T and serum glutamic oxaloacetic transaminase (SGOT or ASAT) (Ansari et al.,2019 ; Hemalatha et al., 2015 ; Zhu et al., 2015). Elevated troponin I is a clinical indicator of cardiac injury (Ndumele et al., 2018). Troponin I and CRP test results were negative. This indicates an absence or low level, in the blood, of the two parameters sought. These results suggest that the aqueous extract of D. brazzeïroots at the doses studied would not induce the production of troponin I or CRP. This extract would not induce myocardial lesions or necrosis and therefore could be cardioprotective. Indeed, it is reported that when myocardial cells containing these markers are damaged or destroyed due to insufficient oxygen or glucose supply, the cell membrane becomes permeable or may rupture, resulting in enzyme leakage (Ansari et al., 2019). To confirm or refute this hypothesis, the levels of CK-MB, LDH and ASAT were quantitatively evaluated in rats treated with aqueous extract of D. brazzeï roots (125, 250 and 500 mg/kg). The results obtained reveal lower levels of CK-MB, LDH and ASAT only at the minimum dose of 125 mg/kg compared to the control lot. The effective

cardioprotective effect of this extract could be at this dose. These results are different from those of Akpanabiatu et al., (2003) who worked on the aqueous extracts of Eleophorbia drupifera and Artemisia afra leaves in rats.

The results of the analyses of the hematological parameters show significant elevations of the levels of red blood cells, hemoglobin and CCMH at the dose of 125 mg/kg. An increase in other parameters was noted at 500 mg/kg. According to Babes et al.,(2021), a low hemoglobin level predicts the development of major cardiac events in patients with acute coronary syndrome. The aqueous extract of the roots of D. brazzeïwould thus stimulate the production of blood cells and would consequently prevent cases of anemia. Indeed, lymphocytes modulate the inflammatory response; a low lymphocyte count is associated with more extensive atherosclerosis (Babes et al., 2021; Mehta et al., 1997), as well as with a poor prognosis in patients with myocardial infarction (Babes et al., 2021). This potential would be unfavorable in infarction because of the high level of blood platelets (hemostasis). Our results corroborate those of Odo et al., (2020) who worked on the physiological effects of Duranta erecta (L) leaves on the albino rat, rattus norvegicus.

Modifiable risk factors such as obesity or overweight, dyslipidemia, and high blood pressure predispose to myocardial infarction (Khan et al., 2021 ; Benjamin et al., 2018 ; Boateng and Sanborn, 2013). Administration of the aqueous extract of D. brazzeïroots (125, 250, and 500 mg/kg) for 7 days resulted in a significantly higher change in weight of normal rats compared to that of 2.13% in normal rats given distilled water. This result suggests that this extract causes overweight in rats. These excess weights of more than 10% increase the relative risk of developing ischemic heart disease to 1.4. This risk is elevated to 2 for an excess weight exceeding 30% (Alaya et al., 2002). With this extract, the excess weight is very far from 30% at the doses studied. Concerning the lipid profile, it has been established that high concentrations of triglycerides, total cholesterol, LDL-cholesterol and a decrease in HDL-C levels accelerate the development of atherosclerotic plaques and are thus major risk factors for coronary heart disease and myocardial infarction (Kulothungan and Shaik, 2021 ; ATF, 2012). However, the results obtained show that the aqueous extract of D. brazzeïdoes not significantly influence the serum triglyceride level but decreases the serum total cholesterol level and increases the HDL-C level at the studied doses of this extract compared to the control. Numerous studies have proven that decreasing LDL-C levels and increasing HDL-C levels can prevent the development of cardiovascular diseases (Sobhy et al., 2018 ; Zhong et al.,2017 ; Shirafkan et al., 2012 ; Iqbal et al., 2004). The aqueous extract of D. brazzeïroots would not promote the occurrence of lipid risk factors and would therefore be favorable for the prevention of cardiovascular diseases such as MI. These results are in agreement with those of Afroz et al., (2016), who also observed a decrease in the levels of lipid parameters in Sundurban honey treated rats.

The aqueous extract of D. brazzeï(125, 250, and 500 mg/kg) did not alter heart mass, and the calculated left ventricular index was lower than in control rats. An increase in the mass of the left ventricle leads to a suffering of the heart, which could lead to a myocardial infarction (MI). This extract would therefore not have a direct negative effect on the heart.

The relationship between the level of diastolic blood pressure (DBP) and the risk of developing MI has been demonstrated in several epidemiological studies and has shown that a 30 mmHg increase in diastolic pressure multiplies the risk of MI by 4 (Alaya et al., 2002). The results obtained show different elevations at 30 mmHg of the diastolic blood pressure in rats having received the aqueous extract of D. brazzeïrootsat three (3) doses. These results suggest that this extract does not promote the occurrence of this risk parameter of MI in normal rats.

## Conclusion:-

In sum, the results obtained in the present study show that the daily administration of the aqueous extractof D. brazzeïrootsdoes not modify almost all the biological and hematological parameters as well as the risk of the myocardial infarction, in the sense of induction of this pathology. Thus, a study of the cardioprotective potential of this extract is envisaged.

**Table I:-** Effect of aqueous extract of Dissotis brazzeïCogn. roots on qualitative biochemical parameters of myocardial infactus in normal rats

| Biochemical parameters | Treatments | | | |
|---|---|---|---|---|
| Qualitative | E.D (1 mL/100 g) | D.brazzeï(125mg/Kg) | D.brazzeï | D.brazzei (500 mg/Kg) |

| | | | (250mg/Kg) | |
|---|---|---|---|---|
| Troponine I | Négative | Négative | Négative | Négative |
| CRP | Négative | Négative | Négative | Négative |

ED : Distilled water ; D.brazzeï : Dissotis brazzeïCogn.

**Table II:-** Effect of aqueous extract of Dissotis brazzeïCogn.roots on quantitative biochemical parameters of myocardial infactus in normal rats.

| Biochemical parameters | Treatments | | | |
|---|---|---|---|---|
| Quantitative | E.D (1 mL/100 g) | D. brazzeï(125mg/Kg) | D. brazzeï(250mg/Kg) | D. brazzeï(500 mg/Kg) |
| CK-MB | 30,8 ± 10,82 | 12,95 ± 3,04$^{ns}$ | 34,12± 11,22$^{ns}$ | 29,22 ± 9,47$^{ns}$ |
| LDH | 757,52±278,69 | 292,2±134,21$^{ns}$ | 850,32±143,83$^{ns}$ | 1070,64±246,38$^{ns}$ |
| ASAT | 166,73±21,31 | 25,31±4,05$^{***}$ | 28,59± 6,56$^{***}$ | 33,718± 5,45$^{***}$ |

E.D: Distilled water ;D. brazzeï:Dissotis brazzeï; Data are expressed as mean ± SEM, (n = 5); ns (not significant), *** (highly significant, p < 0.001) compared to control lot; CK -MB: Creatine kinase MB; LDH, Lactate dehydrogenase (U / L) and ASAT: Aspartate aminotransferase (UI / L).

**Table III:-** Effect of aqueous extract of Dissotis brazzeï Cogn.roots on hematological parameters influenced by myocardial infarction in normal rats.

| Hematological parameters | | Treatments | | |
|---|---|---|---|---|
| | E.D (1 mL/100 g) | D. brazzeï(125 mg/Kg) | D. brazzeï(250 mg/Kg) | D. brazzeï(500 mg/Kg) |
| GR | 6,92 ± 0,09 | 7,58 ± 0,10$^{**}$ | 7,21 ± 0,40$^{ns}$ | 6,17 ± 0,30$^{*}$ |
| GB | 4,91 ± 1,06 | 8,70 ±1,34$^{***}$ | 8,47 ± 2,29$^{ns}$ | 5,23 ± 0,71$^{ns}$ |
| HB | 12,22 ± 0,28 | 13,4 ± 0,39$^{***}$ | 10,82 ± 2,05$^{ns}$ | 11,32 ± 0,59$^{ns}$ |
| PLA | 769,6 ± 91,21 | 1196,8 ±74,46$^{*}$ | 768,6±132,50$^{ns}$ | 684,2± 126,7$^{ns}$ |
| LYM | 2,034±0,80 | 5,28 ± 1,32$^{*}$ | 4,835±0,86$^{ns}$ | 2,48±1,32$^{ns}$ |
| CCMH | 31,54 ± 0,22 | 32,84 ± 0,27$^{**}$ | 38,26 ±5,07$^{ns}$ | 32,8 ± 0,19$^{**}$ |

Data are expressed as mean ± SEM ; ns (no : significant), *significant (p<0.05) ; **significant for(p<0.01); ***significant for(p<0.001); RBC: Red blood cells (x103/µL); WBC: White blood cells (x103/µL); HB:

**Table IV:-** Effect of aqueous extract of Dissotis brazzeïCogn.roots on the weight development of normal rats
D.brazzei:Dissotis brazzeï Cogn.; D0 : Day 0 ; D7 : Day 7. Each mark is mean ± MSE, with n = 5 ; *p < 0.01; significant difference from control (Distilled water).
Hemoglobin (g/dl); MHC: Mean corpuscular hemoglobin concentration(g/dl); PLA: Platelets(x103/µL), and lymphocytes x103/µL).

| | Diastolic blood pressure (mm Hg) | | |
|---|---|---|---|
| Treatments | $T_0$ | $T_{60}$ | Différences |
| NaCl 0, 9% (0,1mL /100 g ip) | 72,76 ± 27,7 | 74,6 ± 30 | 1,84 |
| D. brazzeï(5 mg /Kg) | 85,74 ± 2,77 | 105,45 ± 2,6$^{ns}$ | 19,25 |
| D. brazzeï(10 mg /Kg) | 83,11 ± 1,62 | 86,95 ± 2,4 $^{ns}$ | 3,83 |
| D. brazzeï(20 mg /Kg) | 64,88 ± 5,41 | 66,53 ± 22,63$^{*}$ | 1,65 |

**Table V:-** Effect of aqueous extract of Dissotis brazzeï Cogn.roots on lipid biochemical parameters of normal rats.
ED : Distilled water, D.brazzei: Dissotis brazzeï Cogn.; Each mark is mean ± MSE, with n = 5; ns: significant difference; **p < 0.01; significant difference from control. (Distilled water).

| Treatments | Rat weight in (g) | | |
|---|---|---|---|
| | D0 | D7 | Change (%) |
| Distilled water (1mL /100g) | 200 ,36± 3,82 | 204,62 ± 1,93 | 2,13 |
| D. brazzeï(125mg /kg) | 157,02± 5,00 | 185,3 ± 4,30$^{**}$ | 14,14 |
| D. brazzeï(250mg /kg) | 165,42± 5,70 | 200,82 ± 5,35$^{**}$ | 17,7 |

| | | | |
|---|---|---|---|
| D. brazzeï(500mg /kg) | 155± 3,42 | 176,88 ± 5,36** | 10,94 |

**Table VI:-** The effect of aqueous extract of Dissotis brazzeï Cogn. roots on core mass and parameters.
ED : distilled water, D. brazzei:Dissotis brazzeï Cogn. Each value is mean ± MSE, with n = 5 and
ns: significant difference from control (distilled water).

| Biochemical parameters | | Treatments | | |
|---|---|---|---|---|
| (Lipidic) | E.D (1 mL/100 g) | D. brazzeï(125 mg/kg) | D. brazzeï(250 mg/kg) | D. brazzeï(500 mg/kg) |
| Cholestérol T | 57,10± 16,06 | 57,282±3,01ns | 53,32± 2,56ns | 59,3± 10,35ns |
| HDL-C | 42,02± 4,63 | 60,96 ± 2,18** | 64,74± 1,17** | 59,3± 10,35ns |
| Triglycérides | 84,5± 14,77 | 55,63 ± 5,50ns | 54,28± 5,21ns | 64,68 ±4,15ns |

**Table VII:-** Effects of aqueous extract of Dissotis brazzeï Cogn. roots on diastolic blood pressure (DBP) in normotensive rats
D. brazzei:Dissotis brazzeï Cogn.; Data are expressed as mean ± SEM, (n = 2); ns (not significant), * (significant; $p < 0.05$) compared to control lot (NaCl).

| Organ weight | | Traitements | | |
|---|---|---|---|---|
| | E.D (1 mL/100 g) | D. brazzeï(125 mg/Kg) | D. brazzeï(250 mg/Kg) | D. brazzeï(500 mg/Kg) |
| Heart (g) | 0,34±0,01 | 0,36 ± 0,01ns | 0,39±0,02ns | 0,35±0,01ns |
| Left ventricule (g) | 0,16±0,04 | 0,11 ± 0,00ns | 0,09±0,01ns | 0,13±0,01ns |
| IVG | 3,52±0,24 | 3,32±0,28ns | 2,95± 0,23ns | 2,84±0,33ns |

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