

**EFFECT OF AQUEOUS EXTRACT OF *BOERHAVIA DIFFUSA* LEAVES ON THE
GLYCAEMIA OF RABBITS**N'GUESSAN Koffi ^{1*}, FOFIE N'Guessan Bra Yvette ², KOUASSI Kouadio Henri ³¹Université de Cocody-Abidjan (Côte-d'Ivoire), U.F.R. Biosciences, Laboratoire de Botanique²Université de Cocody-Abidjan (Côte-d'Ivoire), U.F.R. Sciences Pharmaceutiques et Biologiques,
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ABSTRACT: Diabetes touches approximately 5% of Côte-d'Ivoire population. This is a worrying prevalence rate. Our search for means of fighting this affection made us to develop an herbal medicine from decoction of *Boerhavia diffusa* leaves. An ethnopharmacological survey conducted in villages of Côte-d'Ivoire, made us discovered that *Boerhavia diffusa* is used by traditional healers as antidiabetic. In Laboratory, during the experience we carried out, rabbits received, orally, a solution of glucose (4 g/l). Before administration of glucose, the animals had a basal glycaemia of 1.11 ± 0.04 g/l. The not treated rabbits' glycaemia increased and reached 1.39 g/l. The oral administration of glibenclamide lowers blood sugar at about 0.86 g/l, after 180 minutes. The others hyperglycaemic rabbits were given herbal medicine to drink, 0.6 ml per 20 grams of body weight. At 2.5 mg/ml, the phytomedicine does not induce hypoglycaemic effect. At 10 mg/ml, it makes the hyperglycaemia go down from 1.17 g/l to 0.91 g/l. At 40 mg/ml, it induces a significant decrease in blood glucose. A fall of 34 % was observed, 180 minutes later. The glibenclamide exerts a significant basal glucose-lowering effect. The herbal medicine exerts on basal glycaemia a deep action and can induce, at 40 mg/ml, a hypoglycaemic coma. The effect of herbal medicine (40 mg/ml) on hyperglycaemic rabbits is compared to glibenclamide (0.25 mg/ml). The herbal medicine has a hypoglycaemic effect and appears like an antidiabetic and produces its hypoglycaemic effect mainly through alkaloids, sterols or triterpens.

Keywords: Côte-d'Ivoire, Ethnopharmacology, Medicinal plants, Traditional Healers**INTRODUCTION**

Diabetes is a major cause of disability and death and appears like a major public health problem (Dièye, et.al., 2008). This disorder, characterized by polyuria, glycosuria and hyperglycemia, is a widespread metabolic disorder which exists everywhere in the world and interests approximately 6 % of the world population (Kadja, 1998). Côte-d'Ivoire has 3 to 7 % of diabetics, that to say, on average 5 % (Djédjé, 2002). This prevalence rate places the diabetes like most frequent of endocrinien diseases (Gentilini, 1993). In modern medicine, no satisfactory effective therapy is still available to cure diabetes (Paris and Amarnath, 2004). If the injections of insulin or other products make it possible diabetic to remain in life, they cannot, however, make it possible to face the many abrupt fluctuations of the insulin rate which the organization needs. Moreover the diabetes requires a life long treatment, which the patients have of the evil to support (N'guessan, et.al., 2009a). Currently, diabetes therapy is based on the use of hypoglycaemics (sulphamides, biguanides, insulin), on hygieno-diet measures and exercises (Reichard, et.al., 1993). Moreover, the pharmaceutical drugs are either too expensive or have undesirable side effects or contraindications (N'guessan, 2008). In the search of fighting means against this metabolic disorder, ethnopharmacological investigations were conducted in Africa and in most of the developing countries (Anonyme, 1986; Bouquet & Debray, 1974; Kerharo & Adam, 1974; Nacoulma, 1996; Koné, 2005; Neuwinger, 1996). Concerning the plants, these works reported their empirical antidiabetic effect.

The ethnopharmacological survey conducted in villages of Agboville Department in Côte-d'Ivoire (West Africa) made us discover that traditional healers used 28 species of plants to develop 27 medicamentous receipts to treat diabetes (N'guessan, et.al., 2009b). Since this time, we conducted pharmacological experiences to value antidiabetic activity of some plants of this repertory (N'guessan, et.al., 2008a; N'guessan, et.al., 2008b; N'guessan, et.al., 2009c). Concerning *Boerhavia diffusa* L. (Nyctaginaceae), we reported its empirical antidiabetic effect, but no experimental study on its hypoglycaemic activity has been carried out. This study aims at finding new affordable therapies, inexpensive and able to normalize and stabilize the glycaemia. Its objective is to experimentally study the effect of the total aqueous extract of the plant's leaves on the glycaemia of hyperglycaemic rabbits in order to provide scientific evidence of the effectiveness of the traditional use of *Boerhavia diffusa*, as antidiabetic.

MATERIAL AND METHODS

Plant material

The leaves of *Boerhavia diffusa* L. (Nyctaginaceae) were collected, freshly, within the village of Aboudé-Mandéké in the Department of Agboville (South of Côte-d'Ivoire). From the collected samples and specimens of the herbarium of the National Floristic Center, we identified the plant, by its scientific name. A voucher specimen (Moutcho, Côte-d'Ivoire, 18 février 2000, N'guessan Koffi n° 339) was deposited in National Floristic Center (University of Cocody-Abidjan, Côte-d'Ivoire).

Preparation of plant extract

Eight hundred (800) grams of the drug (fresh leaves of *Boerhavia diffusa*) were collected and rinsed then dried for 3 weeks in the shade, well-ventilated place, to avoid contamination by mould. The leaves were pulverized with an electric grinder (type RETSCH) and a powder was obtained. Two hundred (200) grams of the dry powder were obtained and introduced in 500 ml of distilled water. The mixture, bulled during 30 minutes, was wrung in a neat cloth square, filtered successively twice on absorbent cotton and on Wattman 3 mm paper. The volume of the filtrate obtained was concentrated and evaporated in a drying oven at 60°C, during 2-3 days. The pulverized crystals made it possible to obtain fine powder used for the experimentation. The total water extract, codified ONBD, is then kept in sterilized glass bowls, hermetically closed, in a fridge.

Animal material

We used rabbits (*Oryctolagus cuniculus*, Leporidae) we bought in a farm located in Bingerville, suburbs of Abidjan (Côte-d'Ivoire). They were twenty four (24), with as many males as of females. These animals were old 6 to 10 weeks and weighed between 1200 and 1800 grams. They were placed in ventilated metal cages containing litters of shavings which are regularly renewed. They are acclimatized to the conditions of the animal house, during 7 days before the treatment and fed with the granules produced by the Ivorian Compound Food Manufacturing Society (F.A.C.I.). We used tap water. The rats were divided into 8 batches of 3, as follows:

- batch 1: sample rabbits with normal glycaemia
- batch 2: sample hyperglycaemic rabbits treated with glucose (4 g/l)
- batch 3: hyperglycaemic rabbits treated with glibenclamide at 0.25 mg/ml
- batch 4: hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml
- batch 5: hyperglycaemic rabbits treated with herbal medicine at 10 mg/ml
- batch 6: hyperglycaemic rabbits treated with herbal medicine at 2.5 mg/ml.
- batch 7: normal glycaemic rabbits treated with glibenclamide at 0.25 mg/ml
- batch 8: normal glycaemic rabbits treated with herbal medicine at 40 mg/ml.

Technical material

Pasteur pipettes were used to take blood samples we collected in hemolysis tubes. Electrical scales for weighing powders were needed. The glycaemia level determination device included a spectrophotometer of KENZA type.

Chemicals

Surgical spirit was necessary to treat injured rabbits. The glucose, a glucidic agent, known to cause hyperglycaemia, was used. For the treatment of induced hyperglycaemic rabbits of control group, we used glibenclamide (DAONIL tablet, 5mg), the reference product having hypoglycaemic effect. Oxalate of sodium and sodium fluoride was needed in order to stabilize the process of glycolysis in blood. To carry out the phytochemical screening, we used solvents (ether of oil, methanol and distilled water) and various classic reagents (N'guessan, 2008). Classical methods described in the works of Ronchetti and Russo (1971), Hegnauer (1973), Wagner (1983), Békro et.al. (2007) were used to characterize the chemical groups.

Induction of hyperglycaemia

We used glucose whose hyperglycaemic property is established with rabbits (Kwashié, et.al., 1998; Gharras, et.al., 1999). Lukens (1948), Siliart and André (1987) were interested in the glycaemia disorders and therapeutic consequences on dogs. Instead of dogs, we used rabbits, easier to handle and in which the diabetogenic property was also observed (Kadja, 1998; Djédjé, 2002). The glucose absorption depends on the body weight of the animal: 0.6 ml of glucose for 20 g of rabbit weight. Except rabbits of batches 1, 7 and 8, all the animals received a glucose overload at the moment T=0, after basal glycaemia determination. The administration of glucose (4 g/l) is done by oral way, with a nozzle of intubation. Then, the status of rabbits is closely monitored and is detected every thirty minutes by determination of blood sugar quantity.

Treatment of hyperglycaemic rabbits with glibenclamide and herbal medicine

The rabbits of batch 3 are treated with glibenclamide (5 mg/20 ml distilled water, that to say 0.25 mg/ml), 30 minutes, after the cramming by glucose. The rabbits of batches 4, 5 and 6 are treated with the herbal medicine codified "ONBD", 30 minutes, after the cramming by glucose. To do things like the traditional healers, the hyperglycaemic rabbits received, orally, 0.6 ml of glibenclamide and herbal medicine for 20 g of body weight.

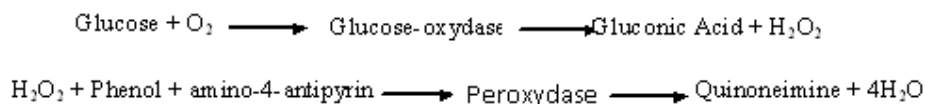
Rabbits' blood sampling

All the animals (rabbits) used for experimentation were deprived of food overnight. Pasteur pipettes were used to take blood samples done, intravenously, through the marginal vein of the ear, to determine glycaemia level. The blood (3 ml) is collected in hemolysis tubes containing an anticoagulant (oxalate of sodium and sodium fluoride) in order to stabilize the process of glycolysis in blood. Thirty (30) minutes before treatment, the blood of all the animals was taken, for determination of the basal glycaemia. Thereafter, blood samplings were made, every 30 minutes after the treatment, according to the constituted batches. On the whole, 7 blood tests were carried out on each animal, during 180 minutes.

Rabbits' glycaemia determination

Principle of determination

The method used is that related to the enzyme (Djédjé, 2002). It consists in oxidizing glucose by the glucose oxydase, enzyme with production of gluconic acid and hydrogen pyroxyde (H_2O_2). The hydrogen pyroxyde reacts with phenol and 4-amino-antipyrine in the presence of peroxydase to form a compound of red brick, quinoneimine and water. The optical density of quinoneimine to 500 nm is proportional to the concentration of glucose in the sample. Below we present the following reaction of blood sugar quantity determination.



Protocol

A milliliter of enzymatic solution works in 10 microliters of serum. The blood is centrifuged to 3500 rpm for 10 min and then the serum is collected. After a bain-marie at 37 ° C, the serum is immediately analyzed with a spectrophotometer KENZA type. A reading is made with a spectrophotometer at 500 nm against the white composed of enzymatic solution. The glycaemia is then determined each hour, during experience. Below, we present formula used to calculate the glucose rate:

$$\text{Glucose rate (g/l)} = \frac{D_0 \text{ sample}}{D_0 \text{ standard}} \times n, \text{ with } n = \text{standard value.}$$

The dosage of glycaemia has been achieved in Medical Analysis Laboratory located in City of Arts (L.A.M.C.A) in Cocody (Abidjan, Côte-d'Ivoire).

Statistical analysis

Data on the variations of glycaemia were expressed in the form Mean \pm SEM of 3 observations, on the histograms we traced with the STATISTICA software. Data were analyzed statistically by one way analysis of variance ANOVA statistical test using STATISTICA version 6.05 (Windows XP) to test for significance. P < 0.05 was considered significant. We used Mauchley test to verify the condition of sphericity and Newman Keuls test for the comparison of the means ($\alpha = 5\%$).

RESULTS

Basic glycaemia and evolution of glycaemia after glucose overload

The figures 1 to 6 show the level of blood glucose in normal, hyperglycaemic control and experimental groups of rabbits. Before treatment, all the animals had a basic glycaemia of about $1.11 \text{ g} \pm 0.05$. The figure 1 reports result on rabbits of batches 1 and 2. The rabbits of batch 1 are the sample rabbits not induced with glucose overload (4 g/l). Their glycaemia fluctuates between 1.12 and 1.18 g/l: glycaemia remains basically stable, during the experiment. We notice two stages in the evolution of glycaemia with the rabbits of batch 2 (sample rabbits induced with glucose overload but not treated): an increase phase from basic glycaemia (1.09 g/l) to the peak (1.39 g/l), thirty (30) minutes after glucose administration. During the second phase, the rabbits showed a significant decreasing level of blood glucose during which glycaemia goes from the peak to 1.16 g/l. A fall of 21.5 % is noted, after the glycaemic peak. At the end of the experimentation, the blood glucose value (1.16 g/l) is near normal level.

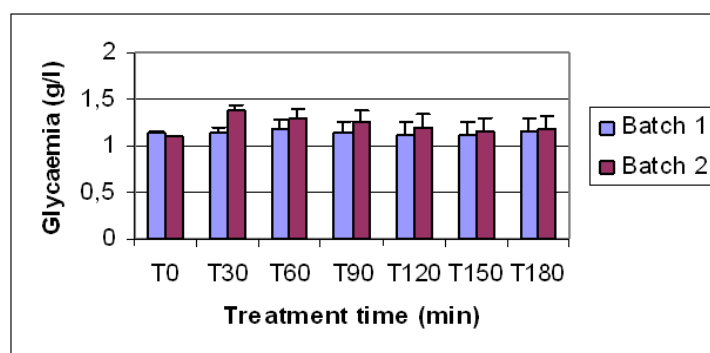


Fig. 1: Glycaemia variation histogram for sample untreated rabbits with normal glycaemia and sample hyperglycaemic rabbits treated with glucose (4 g/l); Mean \pm SEM, n=3, P < 0.05.

Batch 1: sample untreated rabbits with normal glycaemia

Batch 2: sample hyperglycaemic rabbits treated with glucose (4 g/l)

Effects of glibenclamide on hyperglycaemic rabbits

The administration of glibenclamide to hyperglycaemic rabbits of batch 3 shows a significant decrease in the level of blood glucose that pass from 1.39 g/l to 0.86 g/l. A fall of 38 % is observed, 180 minutes later (fig. 2).

Effects of herbal medicine on the glycaemia of hyperglycaemic rabbits

The figure 3 shows the level of blood glucose in hyperglycaemic control rabbits of batch 2 (sample rabbits induced with glucose overload but not treated) and experiment group (rabbits of batch 4, hyperglycaemic rabbits treated with herbal medicine, at 40 mg/ml). After the peak of 1.26 g/l, the rabbits of batch 4 showed a significant decreasing level of blood glucose during which glycaemia reaches 0.86 g/l, at the end of experience. The figure 4 shows the level of blood glucose in experiment groups. We noticed that the herbal medicine has a significant hypoglycaemic effect. The glycaemia of the rabbits of batch 4, treated with the herbal medicine at 40 mg/ml, decreases but does not go back to a normal level. The fall (33%) is comparable to that of glibenclamide (38%); at a rate of 40 mg/ml, the herbal medicine has a glucose-lowering effect. Herbal medicine (ONBD) and glibenclamide treatment to hyperglycaemic rabbits significantly reversed the level of blood glucose. However, the effect of ONBD is not more prominent when compared with glibenclamide. The use of the aqueous decoction from leaves of *Boerhavia diffusa* revealed that it has glycaemic properties, which vary from one dose to another (Figure 5). The glycaemia of rabbits of batch 5, treated with the herbal medicine at 10 mg/ml, goes down from 1.17 g/l to 0.91 g/l, 180 minutes later. From the peak, we observed a fall of 23%. Until the end of the experiment, there is no stabilization of glycaemia at its normal value. The glycaemia of rabbits of batch 6, treated with herbal medicine at 2.5 mg/ml, experienced a similar evolution to that of the sample hyperglycaemic rabbits non treated (rabbits of batch 2); despite treatment, the glycaemia rate which risen to $1.24 \text{ g/l} \pm 0.10$, after administration of glucose practically does not vary: herbal medicine therefore does not exert a significant hypoglycaemic effect at the subliminal dose of 2.5 mg/ml. A fall of 17% is noted, after the glycaemic peak that is comparable to result with animals of batch 2.

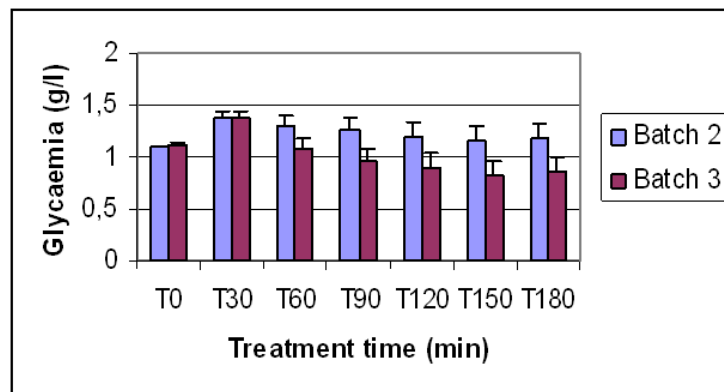


Fig. 2: Glycaemia variation histogram for sample hyperglycaemic rabbits treated with glucose and sample hyperglycaemic rabbits treated with glibenclamide; Mean ± SEM, n=3, P < 0.05.

Batch 2: sample hyperglycaemic rabbits treated with glucose (4 g/l)
 Batch 3: hyperglycaemic rabbits treated with glibenclamide (0.25 mg/ml)

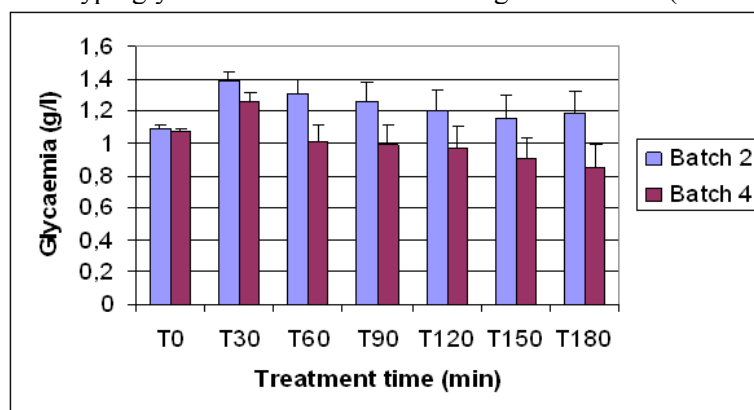


Fig. 3: Glycaemia variation histogram for sample hyperglycaemic rabbits treated with glucose and hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml; Mean ± SEM, n=3, P < 0.05.

Batch 2: sample hyperglycaemic rabbits treated with glucose (4 g/l)
 Batch 4: hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml

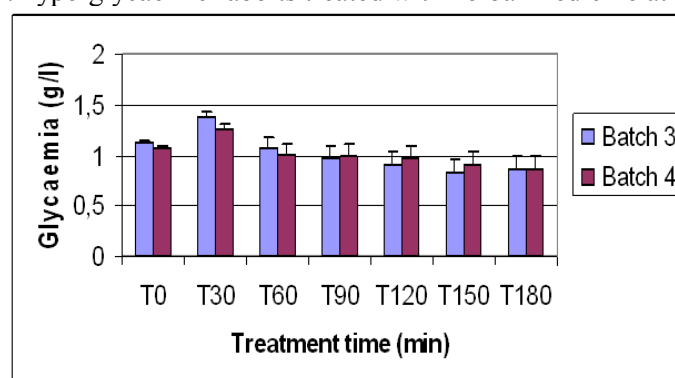


Fig. 4: Glycaemia variation histogram for hyperglycaemic rabbits treated with glibenclamide and hyperglycaemic rabbits treated with herbal medicine; Mean ± SEM, n =3, P < 0.05.

Batch 3: hyperglycaemic rabbits treated with glibenclamide (0.25 mg/ml)
 Batch 4: hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml

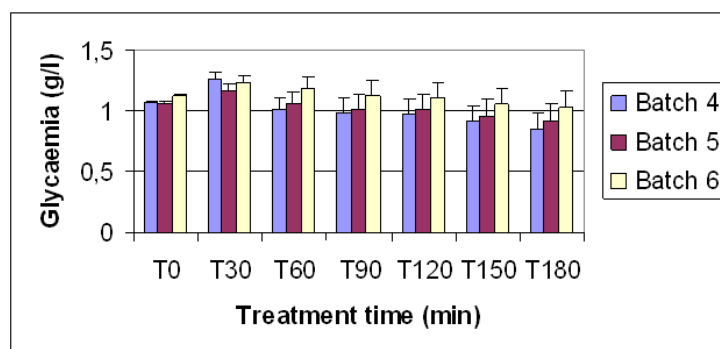


Fig. 5: Glycaemia variation histogram for hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml, 10 mg/ml and 2.5 mg/ml; Mean \pm SEM, n=3, P < 0.05.

Batch 4: hyperglycaemic rabbits treated with herbal medicine at 40 mg/ml

Batch 5: hyperglycaemic rabbits treated with herbal medicine at 10 mg/ml

Batch 6: hyperglycaemic rabbits treated with herbal medicine at 2.5 mg/ml

Effect of glibenclamide and herbal medicine on basal glycaemia of rabbits

The figure 6 shows the level of basal blood glucose in normoglycaemic experiment groups treated with the glibenclamide (rabbits of batch 7) and with the herbal medicine (rabbits of batch 8). The rabbits showed a significant decreasing level of blood glucose from the peak to 0.76 g/l during treatment with glibenclamide and 0.43 g/l, when treated with the herbal medicine, 180 minutes later.

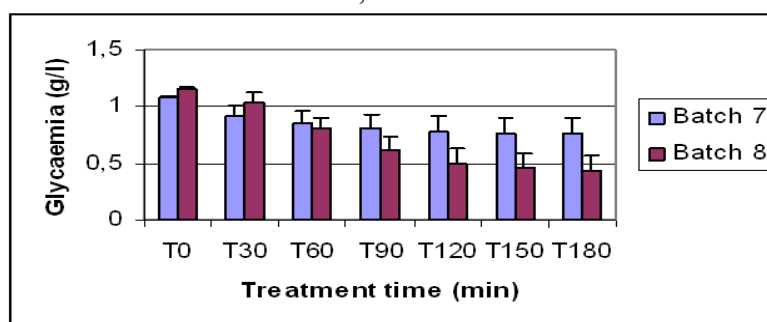


Fig. 6: Glycaemia variation histogram for normal glycaemic rabbits treated with glibenclamide and herbal medicine; Mean \pm SEM, n=3, P < 0.05.

Batch 7: normal glycaemic rabbits treated with glibenclamide (0.25 mg/ml)

Batch 8: normal glycaemic rabbits treated with herbal medicine at 40 mg/ml.

Experimental validation for the medicinal activity of the plant using phytochemistry

We performed a primary validation of the traditional medical practices, by looking for the chemical groups that explain the antidiabetic effect of the herbal medicine "ONBD". Thus *Boerhavia diffusa* leaves were chemically screened and yielded alkaloids, flavonoids, polyphenols, saponosides, sterols and triterpenes. Among these compounds, the alkaloids, sterols and triterpens can be incriminated in the antidiabetic activity of the plant. Alkaloids would be used as stimulatives of the hepatic glycogenogenesis (Neuwinger, 1996). Sterols and triterpens are recognized for their properties to decrease the rate of blood glucose (Nacoulma, 1996). Alkaloids, Sterols or triterpens highlighted in the leaves of the plant would be responsible for the observed antidiabetic effect.

DISCUSSION

Basic glycaemia and evolution of glycaemia after glucose overload

The basic glycaemia in rabbits deprived of food overnight is about 1.11 g \pm 0.05. This result confirms work of Djédjé (2002). After glucose overload, the rabbits showed a significant level of blood glucose: glucose induces hyperglycaemic effect on rabbits. That confirms the property of glucose as a product able to create hyperglycemia. At the end of the experimentation, the blood glucose value (1.16 g/l) is near normal level. The induced hyperglycaemia is transitory: the organism is able to restore normal glycaemia, after a glycaemic overload. This result confirms that of Kwashié et.al. (1998) on rabbits' glycaemia.

Effects of glibenclamide on hyperglycaemic rabbits

The administration of glibenclamide to hyperglycaemic rabbits shows a significant decrease in the level of blood glucose: glibenclamide exerts a hypoglycaemic effect, in accordance with the results of Gharras et.al. (1999). The glibenclamide contribution corrects therefore the hyperglycaemia created by glucose overload. The fixing of glibenclamide to its receptor allows the entry of glucose into the cell, preventing the accumulation of glucose in the blood that explains hyperglycaemia reduction. The glibenclamide induces a significant hypoglycaemic effect, 60 minutes, after glucose overload. Three (3) hours after administration, the glycaemia of treated rabbits with the reference product decreases significantly but its normal value is not restored. This result does not tally with that of Kwashié et.al. (1998) on albino's rabbits; in their study, glibenclamide exerts significant hypoglycaemic effect, 120 minutes, after administration.

Effects of herbal medicine on the glycaemia of hyperglycaemic rabbits

The administration of the herbal medicine at lower doses (≤ 2.5 mg/ml) does not induce significant hypoglycemic effect; at low doses (subliminal doses), the phytomedicine would have normoglycaemic activity. The administration of the herbal medicine at 10 mg/ml highlights the hypoglycaemic effect. The plant medicine lowers the hyperglycemia level without bringing back the glycaemia to its normal value of 1.11 g/l. At 40 mg/ml, the herbal medicine lowers the hyperglycemia level, but does not restore normal glycaemia, after 180 minutes of treatment. The effect of the herbal medicine at 40 mg/ml approximates that of glibenclamide at 0.25 mg/ml. The same herbal medicine (*Boerhavia diffusa* leaves, codified BLEt), used in the treatment of diabetes mellitus, exerts a hypoglycemic effect and restores normal glycaemia to 0.99 g/l when a dose of 30 mg/ml is administered to rats (Paris and Amarnath, 2004).

Effect of glibenclamide and herbal medicine on basal glycaemia of rabbits

The reference product (glibenclamide) and the herbal medicine exert a significant basal glucose-lowering effect but the activity of phytomedicine on basal glycaemia is better than glibenclamide. The two substances would exert an effect on the insulinosecretion. The noninsulinic treatment of diabetes utilizes oral hypoglycaemiant type of sulphamides and type of biguanides. Sulphamides to which belonged glibenclamide act by stimulating the secretion of insulin (Kwashié et al., 1998). The biguanides reinforce the peripheral use of glucose and appear to inhibit the gluconeogenesis. The herbal medicine (ONBD) would have an intra-pancreatic action by stimulating the production of insulin, similar to that observed with the sulphamides. Our findings are in agreement with those reported by Paris and Amarnath (2004). The antidiabetic effect of ONBD may be due to increase release of insulin from the existing β -cells of pancreas similar to that observed after sulphonylurea administration. Administration of herbal medicine significantly decreased the activities of gluconeogenic enzymes in hyperglycaemic rabbits. The herbal medicine exerts on basal glycaemia a deep action and can induce, at 40 mg/ml, a hypoglycaemic coma.

This experience was conducted to study the antidiabetic activity of *Boerhavia diffusa* leaves in rabbits as well as to provide an introductory approach for the evaluation of its traditional preparation in order to scientifically validate the therapeutic preparation of the plant in the control of diabetes. The results show that the effect of herbal medicine (ONBD) at 40 mg/ml is compared to glibenclamide (0.25 mg/ml). The effect of herbal medicine was more prominent when the dose used is higher: the herbal medicine exerts dose-dependent hypoglycemic effect and appears like an antidiabetic.

CONCLUSION AND PROSPECT

The aqueous decoction from leaves of *Boerhavia diffusa* exerts a dose-dependent hypoglycemic effect. It has a normoglycaemic activity at lower doses (≤ 2.5 mg/ml) and a hypoglycaemic activity, at higher doses (> 2.5 mg/ml). The dose of 40 mg/ml is the most effective among the administered doses. Indeed, at 40 mg/ml, the herbal medicine reduces hyperglycaemia without bringing back glycaemia to its normal value of about 1.11 g/l ± 0.05 , after 180 minutes of experiment. The plant proves to be an antidiabetic. Subsequent studies are needed to deepen the mechanism of action and identify the active principle of *Boerhavia diffusa* leaves which have a significant hypoglycaemic effect and appear like an antidiabetic due to chemical components as alkaloids, sterols and triterpens.

REFERENCES

1. Anonyme, (1986). Médecine traditionnelle et pharmacopée, contribution aux études ethnobotaniques et floristiques au Togo. Edition de l' A.C.C.T., 671 p.
2. Békro YA, Békro JAM, Boua BB, Tra Bi FH et EE Éhilé, (2007). Etude ethnobotanique et screening phytochimique de *Caesalpinia benthamiana* (Baill.) Herend et Zarucchi (Caesalpinaceae). Rev. Sci. Nat. Vol. 4 (2) : 217-225.
3. Bouquet A & Debray M, (1974). Plantes médicinales de Côte-d'Ivoire, Imprimerie Louis Jean, Paris (France), 232 p.
4. Diatewa M, Badela SC, Hondi-Assah TC et AA Abena, (2000). Evaluation de l'activité hypoglycémiant des extraits de aqueux des feuilles de *Cogniauxia podoleana* chez le rat. Revue de Médecines et Pharmacopées Africaines, 14 : 43-47.
5. Dieye AM, Sarr A, Diop SN, N'Diaye M, Sy GY, Diarra M, Gaffary LR, Sy AN and Faye B (2008). Medicinal plants and the treatment of diabetes in Senegal: survey with patients. Fund. Clinic. Pharmacol. 22: 211-216.
6. Djédjé H, (2002). Etude prospective pour la réduction et la stabilisation de la glycémie chez le Lapin diabétique, par DIACODA, une substance de source végétale. Diplôme d'Etudes Approfondies de Biotechnologie. Université de Cocody-Abidjan, U.F.R. Biosciences, Laboratoire de Biochimie, 36 p.
7. Gharras L, Hmamouchi M, Lamnouar D et M Bengoumi, (1999). Etude comparative de l'effet hypoglycémiant de six plantes de la pharmacopée traditionnelle marocaine. Revue de Médecines et Pharmacopées Africaines, 13 : 71-80.
8. Gentilini M, (1993). Tropical Medicine. Editions Flammarion, Paris, 928 p.
9. Hegnauer R, (1973). Chemotaxonomie der Pflanzen, Birkhäuser Verlag, Basel, Stuttgart, 6, 761 p.
10. Kadja B, (1998). Evolution de quelques paramètres de diagnostic diabétique au cours d'un essai thérapeutique par un phytomédicament (DIACODA). Diplôme d'Etudes Approfondies de Biotechnologie. Université de Cocody-Abidjan, UFR Biosciences, Laboratoire de Pharmacodynamie-Biochimie, 31 p.
11. Kerharo J & JG Adam, (1974). La pharmacopée sénégalaise traditionnelle. Plantes médicinales et toxiques, Edition Vigot frères, Paris, 1007 p.
12. Koné MW, (2005). Potentiel des plantes médicinales de Côte-d'Ivoire dans le contrôle des haemochoses chez les ovins. Thèse de Doctorat de l'Université de Cocody-Abidjan, U.F.R. Biosciences, Laboratoire de Botanique, 202 p.
13. Kwashié EG, Kodjo A et M Gbeassor, (1998). Effet de *Sterospermum kunthianum* et *Oxynanthera abyssinica* sur la glycémie. Revue de Médecines et Pharmacopées Africaines, 12 : 89-98.
14. Lukens FD, (1948). Alloxan Diabetes. Physiol. Reviews 28: 304-330.
15. Nacoulma-Ouédraogo O, (1996). Plantes médicinales et pratiques médicales traditionnelles au Burkina Faso : cas du Plateau central. Thèse de Doctorat ès Sciences Naturelles, Université de Ouagadougou (Burkina-Faso), Fac. Sc. et Tech., 605 p.
16. Neuwinger HD, (1996). African Ethnobotany. Poisons and drugs. Chemistry, Pharmacology, Toxicology. Ed. Chapman and Hall, Bundesrepublik Deutschland, 942 p.
17. N'Guessan K, (2008). Plantes médicinales et pratiques médicales traditionnelles chez les peuples Abbey et Krobou du Département d'Agboville (Côte-d'Ivoire). Thèse de Doctorat ès Sciences Naturelles. Université de Cocody-Abidjan, U.F.R. Biosciences, Laboratoire de Botanique, 235 p.
18. N'Guessan K, Aké-Assi E, Adou YC et D Traoré, (2008a). Effet de l'extrait aqueux des feuilles de *Crescentia cujete* sur la glycémie de lapins diabétiques. Revue Méd. Pharm. Afr. 21 : 89-96.
19. N'Guessan K, SORO D, Kouassi KE, Amoikon KE et AJ Djaman, (2008b). Effet de l'extrait des racines de *Jatropha gossypifolia* sur la glycémie de lapins diabétiques. J. Sci. Pharm. Biol. 9 (1): 13-21.
20. N'Guessan K, Amoikon KE, Tiébré MS, Amoikon KE, Kadja B and GN Zirih (2009a). Effect of aqueous extract of *Chrysophyllum cainito* leaves on glycaemia of diabetic rabbits. African Journal of Pharmacy and Pharmacology, Vol. 3, Issue 10 (2009): 501-506.
21. N'guessan K, Kouassi KE, Kouadio K, (2009b). Ethnobotanical study of plants used in traditional medicine to treat diabetes, by Abbey and Krobou People of Agboville (Côte-d'Ivoire). American Journal of Scientific Research (AJSR), Issue 4 (2009): 45-58.

22. N'Guessan K, Amoikon KE, D SORO, (2009c). Effect of aqueous extract of *Persea Americana* seeds on the glycaemia of diabetic rabbits. *European Journal of Scientific Research (EJSR)* 26 (3): 376-385.
23. Paris L and MS Amarnath, (2004). Antidiabetic activity of *Boerhavia diffusa* L.: effect on hepatic key enzymes in experimental diabetes. *Journal of Ethnopharmacology* 91: 109-113.
24. Reichard P, Nilson BY and U Rosenquist, (1993). *N-Engl. J. Med.* 329: 304.
25. Ronchetti F et G Russo, (1971). A new alkaloid from *Rauvolfia vomitoria*, *Phytochemistry*, Vol. 10: 1385-1388.
26. Siliart B et F André, (1987). Insulinémie : intérêt de ce dosage dans les troubles de la régulation de la glycémie et conséquences thérapeutiques chez le chien. *Rec. Med. Vet.* 163 (11): 1019-1030.
27. Wagner H, (1983). *Drogen analyse. Dünnschicht chromatographische analyse von Arzneidrogen.* Springer Verlag Heidelberg New York, 522 p.