Laxative Effect of Some Plants Extracts Used in Traditional Pharmacopoeia on the Intestine of Rabbit

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Abstract

Constipation is a condition in which a person or animal has difficulty in eliminating solid waste from the body and the faeces are hard and dry. It is mainly characterized by a difficulty in defecation. The aim of the study is to assess the laxative effect of some plants such as Adansonia digitata, Ziziphus mauritiana and Psidium guayava traditionally used in Chad to treat the problems of constipation. The Microdynamometer method (Biomecatronics) is used to carry out this study. The outcomes show that the extracts’ leaves of Psidium guayava and barks of Ziziphus mauritiana did not lead a significant contraction of duodenum of rabbit until a maximum concentration of 1000 µg, but the extracts’ leaves of Adansonia digitata had a significant effect (P < 0.05) on the intestinal motility of rabbit. This effect of the extract of Adansonia digitata varies with the increase of its concentration (dose dependent) with a maximum of 500 µg and the minimal effect is inferior to 10 µg. On the other hand, the extract of Adansonia digitata has a musculotropic effect because the results have shown that inhibitive molecules such as the atropine and the naloxone do not have to inhibit the action of Adansonia digitata on the intestinal motility of rabbit. Thus, the extract of this plant would reduce significantly the time of intestinal transit and it will be an important laxative drug candidate as described traditionally in Chad.

Keywords

Constipation, Intestinal Contraction, Rabbit, Adansonia digitata, Psidium guayava, Ziziphus mauritiana
1. Introduction

Functional constipation, known as Chronic Idiopathic Constipation (CIC), is constipation that does not have a physical (anatomical) or physiological (hormonal or other body chemistry) cause. It may have a neurological, psychological or psychosomatic cause. A person with functional constipation may be healthy, yet has difficulty in defecating [1].

The functional constipation is a benign constipation essentially caused by a decrease of the digestive motricity or by a decrease of the intestinal flora [2]. A person out of five and not less than a woman on two suffer from constipation worldwide [3].

According to Stewart et al. [4], functional constipation is defined as the inability to defecate completely and spontaneously thrice or more in a week, without any secondary cause.

Daniyam et al. [5] and Cummings et al. [6] reported that bowel habits of different population might vary widely due to several factors, including dietary habit, quantity of fiber intake and difference in gut transit time.

Higgins and Johanson [7] reported that constipation affected 10% to 30% of the adult population in western countries, of which 3% and 5% were chronic constipation.

Constipation is particularly frequent in women due to hormonal cause, because of the action of prostaglandin on the motricity of smooth muscles and that of the digestive tract in general, as well as older people, because of their settled way of life, feeding habit, and several diseases due to ageing as well as the use of a number of medicines [8].

To remedy constipation’s problems, laxatives are generally used. The following are known and commonly used laxatives: liquid paraffin, tartrate acid of potassium, natural seeds of ispaghul (Plantago ovata), sene (Cassia angustifolia) or tamarin (Tamarindus indica), etc. They help accelerate the intestinal transit by softening saddles.

In Chad, certain plants such as Adansonia digitata, Ziziphus mauritiana and Psidium guayava are traditionally used for human and animals to treat the problems of constipation (GELT Project report [9]). In order to as certain that this traditional practice works to treat constipation, we propose to carry out a study using the above plants. Thus, the purpose of this study is to evaluate the laxative effects of the extracts of Adansonia digitata, Ziziphus mauritiana and Psidium guayava on the intestine of rabbit to treat acute and occasional constipations.

2. Material and Methods

2.1. Chemicals

The following chemicals: Bethanechol chloride (196.68, Sigma-Aldrich); Atropine Sulfate Renaudin 0.25 mg/ml and Naloxone 0.4 mg/ml were used during the study.

2.2. Origin of Plants and the Preparation of the Raw Extract

The fresh leaves of Adansonia digitata, Ziziphus mauritiana and Psidium guayava used in this experiment were taken in the botanical garden of the “Centre National d’Appui à la Recherche—CNAR” and identified in the Herbarium of the Livestock Research In-
stitute for Development (IRED), formerly Laboratory of Farcha.

The maceration of these plants was done according to the traditional recipe to treat problems of constipation (GELT Project report [9]). Leaves and barks of these plants were forwarded to the laboratory. They were dried in a steam room at 45°C under ventilation during 48 hours. The dried products were powdered with a mortar following the traditional process.

The raw extract of plant was obtained by maceration in a 20% proportion m/v (Mass of the product on volume of distilled water) after six hours under normal temperature following agitation with a magnetic agitator (Heating Magnetic Agitator 370°C, FISHER BRAND). The agitating speed was put on level 5. Thereafter, the aqueous extract was filtered with Filter paper Pure natural cellulose (64 g/m²), and then concentrated in a steam room at 40°C under ventilation. The concentrated extract is kept at 4°C until the use.

2.3. Laboratory’s Animals

The animals used in this experiment are young male rabbits with an average weight of 1.5 kg. Rabbits were breed at the breeding’s unit of IRED and kept at 28°C under light 12 hours 12 darkness 12 hours. Besides, this study was led by respecting in accordance with the fundamental principles minimizing the sufferings of the animal and after obtaining the ethical agreement from the National Ethical Committee of Chad (Comité National de Bioéthique du Tchad, CNBT).

2.4. Preparation of Animals

Rabbits were led to rest following an intraperitoneal injection of an overdose of pentobarbital.

The bowel was removed, washed and cleaned of connective tissue, and then put in a physiological Ringer solution oxygenated (95% O₂ and 5% CO₂) at 37°C.

2.5. Study of the Intestinal Contraction

The intestinal contraction or relaxation under the influence of the aqueous extracts of various concentrations of the plants was carried out on isolated fragments of the duodenum of the rabbits by the Microdynamometer method (Biomecatronics, France). Portions of the duodenum, 2 cm long each are removed, washed prior to fixed be mounted in a tank to isolated organ of 6 ml of volume maintained at 37°C (Figure 1).

Every 15 minutes, a dosage of 1 to 1000 µg/ml aqueous extracts of the plants is added in the middle of the incubation process and the resulting contraction is recorded. A concentration of Bethanechol 10⁻⁴ M is added to the end of every series of measure.

2.6. Statistical Analysis

The results were expressed in percentage of the maximal effect. Comparison between the different extractions of plants on the intestinal contraction of rabbit is done by ANOVA test. The statistical program is SPSS Software.
3. Results and Discussion

3.1. Effects of the Extracts of *Adansonia digitata*, *Ziziphus mauritiana* and *Psidium guayava* on Intestinal Fragments of Rabbit

A comparative analysis of the effects of the extracts of various plants on the intestinal contraction of the rabbit is illustrated below in Figure 2. This figure showed that the extracts of *Psidium guayava* (GOY) and *Ziziphus mauritiana* (ZI) traditionally used against constipation did not display a significant contraction of the duodenum of rabbits, when a maximal concentration of 1000 µg is used. However, the extract of *Adansonia digitata* (BB) led to a high significant intestinal contraction with the same maximal dose of 1000 µg.

Figure 3 showed the efficiency of the extract of *Adansonia digitata* (BB) on the...
intestinal contraction, when compared with a reference molecule by addition in the middle of an analogue of synthesis of the acetylcholine (Bethanechol $10^{-4}$ M) (Figure 3).

The effect of plants extracts on the intestinal contraction of animals such as rat, mouse, rabbit or human biopsy is shown in the previous study led by Mathieu et al. [10] and Eto et al. [11].

3.2. Effect of the Various Concentrations of Extract of *Adansonia digitata* (BB) on the Intestinal Contraction of Rabbit

Figure 4 showed typical recordings of the effect-dose of extract of *Adansonia digitata* (BB) on the intestinal motility according to various concentrations. The extract of BB...
acted in small doses from 10 µg and reaches a maximal effect at 500 µg. From 1000 µg, it was observed that there is a decrease of the spontaneous contraction. Bethanechol $10^{-4}$ M (BETA) was used always at the end of the experiment to compare the effect of various concentrations of extract of *Adansonia digitata* (BB).

### 3.3. Amplitudes of Intestinal Contraction of Rabbit under the Influence of Various Concentrations of *Adansonia digitata*’s Extract

The amplitudes of intestinal contraction caused by various concentrations of extract of *Adansonia digitata* increased gradually according to the volume of the dose (Figure 5). This increase is dose dependent and it reaches its optimum with concentration of 500 µg of extract of *Adansonia digitata* compared with Bethanechol $10^{-4}$ M.

### 3.4. Studies of Mechanism of Action of Extract of Baobab (*Adansonia digitata*)

Various molecules were tested to estimate the musculotrope or neurotrope effect of baobab extract. Figure 6(a) shows the reaction of the *Adansonia digitata* extract (500 µg) after an inhibition of the neurotropes receivers by Atropine.

Atropine had no effect on extract of baobab because this plant’s extract led to an intestinal contraction of the duodenum; however Bethanechol, which is an antagonist of Atropine did not lead to an increase of the intestinal contraction. Consequently, we concluded that the action of *Adansonia digitata* is musculotrope.

This musculotrope action of extract of baobab is confirmed by an experiment using Naloxone (NALO) as inhibitor (Figure 6(b)). In this experiment, it was noted that the action of Naloxone that is an inhibitor of neurotropes receivers did not prevent the reaction of the baobab extract. Consequently, the extract of *Adansonia digitata* led to an intestinal contraction and this contraction was dose dependent. On the other hand, Betanichol (BETA) which is also an antagonist of the Naloxone (NALO) introduced in the end of the experiment had no effect on the intestinal contraction.
Figure 5. Evolution of the concentrations of extract of *Adansonia digitata* (BB) on the intestinal motility.

Figure 6. (a) Mechanism of action of the effect of Baobab extract (BAOB), Atropine (ATRP), Betanichol (BETAN). (b) Mechanism of action of baobab extracts (BB).

4. Conclusion

These preliminary results showed that the extracts of *Ziziphus mauritiana* and *Psidium*...
guayava did not lead to significant contractions of duodenum of rabbit until a maximum concentration of 1000 µg was reached. On the contrary, *Adansonia digitata* has a significant effect on the intestinal motility of rabbit. Thus, it would reduce significantly the time of intestinal transit. This effect of *Adansonia digitata* extract is dose-dependent and shows a maximal effect at 500 µg and a minimal effect with doses less than 10 µg. Further, the extract of *Adansonia digitata* shows a musculotrope effect, because the inhibitive molecules such as the Atropine and the Naloxone do not have to inhibit the action of *Adansonia digitata* on the intestinal motility of rabbit. According to these results, *Adansonia digitata* could be an important laxative drug candidate and further studies of its toxicity effect among others are required to complete this scientific work.

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