In vitro and *in vivo* anti-hyperglycemic potential of saponins cake and Argan oil from *Argania spinosa*

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Introduction. Type 2 diabetes mellitus (T2DM) and hypertension are the most shared comorbidities in coronavirus-infected patients. In the current COVID-19 pandemic context and according to some papers, including those from the Centre's for Disease Control and Prevention (CDC), patients with type 2 diabetes mellitus and the metabolic syndrome could suffer with an up to ten-times higher risk of dying when they contract COVID-19. T2DM is a universal disease affecting the populations of developed and developing nations. Moreover, T2DM is the most common endocrine disease with indirect relation to several other disorders. It is expected that more than 300 million persons worldwide will suffer from T2DM in 2025. A genetic susceptibility to the disease exists that is promoted by environmental reasons, for example an unhealthful eating behaviour, with obesity being one of the greatest important risk reasons. T2DM is caused by an irregularity of the carbohydrate metabolism, which is directly connected to down insulin levels in blood.

Aim. Many chemical analyses discovered that Argan oil is principally well stable in relation to its fatty acid composition. We consequently studied the anti-hyperglycemic effect of Argan seeds by researching the actions of saponin extracts using α-glucosidase and α-amylase assays as well as an *in vivo* model of alloxan-induced diabetic mice. In particular, we evaluated the ability of Argan extracts to rise the inhibitory properties on digestive enzymes (α-amylase and α-glucosidase). The saponin extracts had an activity with an antidiabetic potential. The specific chemical profile of the Argan fruit extracts, namely cake and Argan oil, could be the reason of a possible anti-hyperglycemic action.

Materials and Methods. The samples were prepared by extraction of roasted

Argan kernels at 110°C for 25 min. From the same kernels, edible traditional Argan oil and saponin cakes of Argania spinosa were obtained according to the technique described by Alaoui et al. We studied the anti-hyperglycemic effect of Argan seeds by researching the actions of saponin extracts using α-glucosidase and α-amylase assays as well as an *in vivo* model of alloxan-induced diabetic mice. In particular, we evaluated the ability of Argan extracts to rise the inhibitory properties on digestive enzymes (α-amylase and α-glucosidase). The saponin extracts had an activity with an antidiabetic potential. The specific chemical profile of the Argan fruit extracts, namely cake and Argan oil, could be the reason of a possible anti-hyperglycemic action. The chemical composition and bioactive molecules were discussed.

In Vitro Antidiabetic Activity. The α-amylase inhibitory capacities were studied by reacting varying concentrations of the extracts with α-amylase and starch solution.

In Vivo Antidiabetic Activity. In this study, mice received an intraperitoneal cure with alloxan formulated in sterile normal saline with 1% (m/v) at 150 mg/kg body mass. The control group got the similar volume of sterile normal saline. Animals were fasted for 14 h, but water was delivered without restrictions before treatment. Later, animals were kept in surveillance for 3 days. Serum glucose was revealed by the glucose oxidase peroxidase technique using a glucometer (One Touch Ultra, LifeScan, Milpitas, CA, USA). Mice with plasma glucose levels that exceeded 200 mg/dl were involved in the experiment.

Results and Discussion. The result showed that the experimented extracts inhibited α-amylase activity dosage dependently of (66.66–333.33 μg/ml) and (88.88–444.44 μg/ml), respectively. Moreover, all extracts indicated significantly ($p < 0.05$) more activity than the acarbose (IC50 = 310.10±0.22 μg/ml). The Argan saponin cake extract has a better inhibitory effect versus α-amylase with IC50 value of 209.10±0.17 μg/ml. Similarly extracts have proved encouraging and concentration-dependent (0.55–74.88 μg/mL) inhibitory activities on α-glucosidase enzyme (Figure 3A). Curiously, the IC50 values 0.89±0.17 μg/ml, 7.56±0.38 μg/ml for saponin extract and Argan oil, respectively, show that mall examined extracts were significantly ($p < 0.05$) greater inhibitors of α-glucosidase than the acarbose (IC50= 17.02±1.22 μg/ml).

After 7 days, there was no statistical distinction between the blood glucose concentration of the diabetic mice treated with the studied saponin extract, Argan oil, or metformin and diabetic control mice (untreated) ($p > 0.05$). After 14 days of therapy with aqueous saponin extract, a considerable reduction in blood glucose concentration was detected ($p < 0.05$); the showed glycemia value was comparable to that shown by the metformin-cured mice but still better than that of the normal mice. After 30 days, the blood glucose concentration of the mice cured with the saponin extract had reduced to reach that of the normal control mice and the metformin-cured mice. The aqueous saponin extract thus applied an antidiabetic activity identical to that of metformin and better than that of Argan oil.

Conclusions. The regular consumption of Argan oil could protect the human body from cancer and heart diseases. This study informs about the inhibitory kinetics of the Argan saponin cake extract and Argan oil anti-key enzymes (α-amylase and α-glucosidas) associated to hyperglycemia. Compared to the positive control, the in-vitro test showed a modest inhibitory activity. Considering the safe profile of Argan saponin cake extract, it can be of importance to consider an extract formulation for inhibition of (α-amylase and α-glucosidas) key enzymes in the small bowel. Such extracts may show an anti-hyperglycemic activity and be utilized as a capsulated formulation.

Inducing diabetes in an animal model by alloxan provides knowledge regarding the pathologic mechanism of diabetes, and is also used to screen treatments for diabetes and diabetes-associated problems. In this study, the potential hypoglycemic activities of Argan saponin cake extract and Argan oil treatments were studied using alloxan-induced diabetic mice.

Moreover, Argan oil considerably decreased the quantity of absorbed glucose in a perfused jejunum segment relative to controls rats. Another study about insulin-sensitizing seed extracts of Argania spinosa demonstrated that a saponin cake subfraction enhanced insulin-induced protein kinase B activation.

This study confirmed the natural nutritional benefit of Argan oil and Argan cake saponins as a food and investigated the effect of Argan on selected digestive enzymes; the anti-enzymatic activity is possibly partly associated to its cake saponin content. We approved a nutritional therapy with anti-hyperglycemic bioactivity.