International Journal of Advanced Engineering and Management Research

Vol. 10, No. 04; 2025

ISSN: 2456-3676

Evaluation of the nutritional, antioxidant and hypoglycemic potential of Aloe Saponaria flowers

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doi.org/10.51505/ijaemr.2025.1301 URL: http://dx.doi.org/10.51505/ijaemr.2025.1301

Received: Jun 17, 2025 Accepted: Jun 28, 2025 Online Published: July 05, 2025

Abstract

Aloe saponaria, a plant of the Aloeaceae family, is widespread in the Far North of Cameroon. The flowers of this plant are consumed as leafy vegetables by local populations in the Mindif district. The aim of the present study was to assess the nutritional and antioxidant potential of Aloe saponaria flowers harvested in the Mindif locality, and to study its short-term hypoglycemic effect on Wistar albino rats. The selected parameters were analyzed using standard methods. Results showed ash content 4.45%; total protein 7.87%; total soluble sugars 8.24% and reducing soluble sugars 15.68%; provitamins (carotenoids) also showed high values with α-carotene (84.26mg/100g), β-carotene (89.45mg/100g), lutein (78.11mg/100g) and lycopene (25.74mg/100)). Characterization revealed the presence of polyphenols, flavonoids, tannins, alkaloids, reducing sugars, saponins and an absence of terpenoids, with polyphenols estimated at 484.52mgEAG/100gMS, flavonoids at 239.44mgEAG/100 g MS, and tannins at 79.1mgEAG/100gMS as the dominant compounds. The extract showed significant antioxidant activity capable of scavenging the free radical DPPH at 312.46mgET/100gMS, ABTS at 325.74mgET/100 g MS, and ferric iron reduction to ferrous iron (FRAP) at 225.36/100gMS. The hypoglycemic effect of the extract showed increasing blood glucose reduction values from 18.8 to 30.98% for the 500mg extract/kgp dose, and a decreasing reduction rate (8.04-1.45%) over time for the 100mg extract/kgp dose. The extract has a dose-dependent hypoglycemic effect. The antioxidant potential highlighted in this study would explain the traditional use of Aloe saponaria in the prevention and treatment of chronic illnesses, as noted by the local population. However, if Aloe saponaria flowers are to be used in the fight against hyperglycemia, its full anti-diabetic activity in vivo (animals and humans), toxicity, stability and preservation of the active ingredient over time need to be studied.

Keywords: Aloe saponaria, flowers, properties, nutritional, antioxidant, hypoglycemic

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1. Introduction

Man's existence is punctuated by the appearance and development of diseases, the failure to control which reduces his life expectancy (Fundu Mbemba, 2021). Significant progress has been made in the search for molecules capable of combating the factors that cause these diseases, leading to life-saving results. However, diet is the primary weapon in the fight against disease. Indeed, to stay in shape, human beings need to eat properly (WHO, 2020). The choice of these foods is often guided by empirical endogenous knowledge, which is passed on over time to become eating habits. These foods are even more highly valued when their nutritional potential is coupled with a presumed or recognized medicinal potential. Indeed, epidemiological studies have shown that certain diets made up of different types of food are associated with longer life expectancy, due to their role in protecting against various pathologies such as cardiovascular disease and certain cancers (De Lorgeril and Salen, 2006; Renaud and de Lorgeril, 1992; Trichopoulou et al., 2003; Visioli and Galli, 2002) Ness and Powles, 1997; Block et al., 1992). The common feature of these different foods is their high content of polyphenols, products of secondary plant metabolism whose usefulness in combating a number of diseases, such as diabetes, has been noted by several authors (Middleton et al., 2000; Ksouri et al., 2007). One of the characteristics of the vegetation around the Mindif peak is the abundance of Aloe saponaria. The culinary traditions of the resident populations have long included the use of the plant's flowers as leafy vegetables, thus highlighting its contribution to a balanced diet. In addition, for a certain segment of the population, particularly the elderly, this product is seen as an allicent due to its curative or preventive effects on certain diseases such as diabetes, cardiovascular disease and cancer. This image of the miracle plant is all the more ingrained in local mindsets, as the non-existence of diabetes, cardiovascular disease (CVA) and cancer patients in their communities would seem to be proof not only of the nutritional but also of the therapeutic potential of Aloe saponaria flowers. However, very few studies on the nutritional or therapeutic potential of Aloe saponaria flowers exist. Characterizing the flowers of Aloe saponaria with a view to their valorization therefore appears to be a key scientific direction. The aim of this study was to assess the nutritional and antioxidant potential of Aloe saponaria flowers harvested in the Mindif locality, and to study the short-term hypoglycemic effect on albino Wistar rats.

2. Method

2.1-Plant material

Samples were obtained from the plain of Mindif, in the Mayo Kani department of the Far North region of Cameroon. The harvested flowers were shade-dried and then ground to powder using a Moulinex-type grinder. The powder obtained was packed in biodegradable paper packaging and stored in a dark place before analysis.

Aloe saponica flower extract was obtained by boiling 10g of powdered sample in 100 mL of distilled water for 30 minutes. After cooling and filtration, the extract was stored at 4°C for use.

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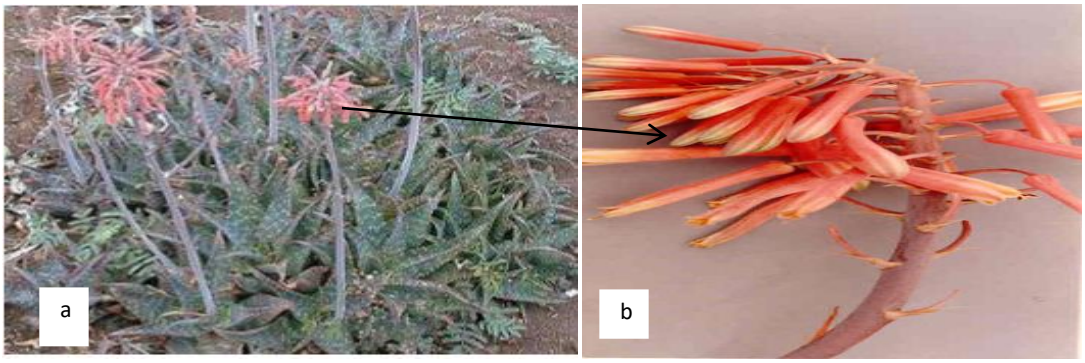

Figure 1: a) Aloe saponaria b) Aloe saponaria flower

2.2-Animal material

24, 10-week-old Wismar rats were obtained from the animal biology laboratory of the University of Maroua.

2.3- Nutritional analysis of Aloe saponaria flowers

Biochemical analyses were carried out on samples of Aloe saponaria powder. Total sugar content was obtained using the method of Dubois et al. (1956). Protein content was determined by the method of Bradford (1976). Ash and dry matter content were determined by the AOAC (2005) method. Carotenoid content was determined by determining α-carotene, β-carotene, lycopene and lutein content using the method of Sumanta et al. (2014).

2.4- Qualitative analysis: phytochemical screening

Phenolic compounds were identified by the method of Konkon et al. (2006), flavonoids by the methods of Mibindzou and Mouellet, 2004; Konkon et al. (2006). The presence of catechic and gallic tannins was determined by the method of Konkon et al., 2006. The search for alkaloids was carried out by the methods of Benzahi, (2001); Konkon et al. (2006), that of terpenoids by the method of Khan et al. (2011), that of reducing compounds by the method of Bentabet-Lasgaa, (2015) and that of saponins by the method of Daoudi et al. (2015).

2.5- Quantification of phytochemicals and antioxidant activities

Total polyphenols were determined by the method of singleton et al. (1999), total flavonoids by the method of Mimica-Dikic (1992) and total tannins by the method of Brainbridge et al. (1996). DPPH and FRAP antioxidant activities were determined by the methods described by Sun et al. (2005) and Benzie and Strain (1996) respectively.

2.6- Short-term hypoglycemic activity of Aloe saponaria flowers

The aim of this part of the study was to evaluate the rate of blood glucose reduction by the extract from the infusion of Aloe saponaria flowers compared with Glibenclamide (in the form of

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daonil 5mg: reference drug for blood glucose lowering) on rats induced to hyperglycemia by 10% anhydric glucose according to the method of Lawson and Gadegbeku (1997).
The experiment was conducted in five main phases:

Phase 1: subject adaptation phase

The 24 subjects were received in the laboratory 72 hours before the starting of the experiments. They were fed a standard diet and watered ad libitum.

Phase 2: Fasting blood glucose measurement

After 16 hours of fasting, subjects were alternately weighed and blood glucose levels were measured using a Blood Glucose Meter on caudal blood samples taken with a 2.5mL syringe.

Phase 3: Induction of hyperglycemia

Induction of hyperglycemia was carried out on subjects in batches 2, 3 and 4, each consisting of 6 subjects, according to the experimental set-up below:

Table 1. Induction of hyperglycemia

Subjects	**Batch 1**	**Batch 2**	**Batch 3**	**Batch 4**
Treatments	T_0	T_1		

T0=7ml/kg body weight distilled water by esophageal tube gavage.
T1=2ml/100g body weight of 10% glucose by esophageal tube gavage.
Blood glucose levels were measured 30 minutes after each treatment. All subjects with blood glucose levels above 120 mg/dl were considered hyperglycemic and subjected to the experimental hypoglycemic treatments according to the following schedule:

Phase 4: Experimental treatment of hyperglycemia

Sujets	Batch 1 (Control)	Ba		tch 2 Batch 3 Batch 4
Treatment	TH_0	TH_1	TH_2	TH_3

Batch 1: control batch of 6 hypoglycemic rats
Batches 2, 3 and 4: batches of 6 hyperglycemic rats each
TH0 7 ml/kg body weight of 0.9% NaCl
TH1 100mg/kg body weight Aloe saponica extract 10g100ml
TH2 500mg/kg body weight Aloe saponica extract 10g100ml
TH3 5mg/kg body weight of Glibenclamide for reference treatment.

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Phase 5: Evaluation of experimental results

Blood glucose levels were measured 60, 120 and 180 minutes (T60mins, T120mins and T180min) after the various treatments.

The rate of reduction was determined according to the formula proposed by Begbin et al. (2021):

$$Reduction\ rate\ (Tr) = \frac{Initial\ glycemia - Final\ glycemia}{Initial\ glycemia} \times 100$$

2.7- Statistical analysis

The statistical study was carried out using XLSTAT-PRO 7.1 statistical analysis software. Results were analyzed using a one-factor analysis of variance (1-factor ANOVA) and the Turkey test. Values are given as the mean followed by the standard error on the mean. These tests were given at the 5% significance level.

3- Results

3.1- Biochemical characteristics of Aloe saponaria flowers

The physicochemical characteristics of Aloe Saponica flower samples are shown in the table below.

Table 3. Physicochemical characteristics of Aloe Saponica flower samples.

WC (%)	**TA (%)**	**TP (%)**	**TSS (%)**	**Rs (%)**	**α carotenes (mg/100 g)**	**βcarot (mg/100 g)**	**Lut (mg/100g)**	**Lyc (mg/100g)**
4.24± 0.02	4.46± 0.03	7.87± 0.02	8.24±0.01	1.28± 0.02	84.26± 0.27	89.45± 0.02	79.11± 0.02	25.74 ± 0.02

WC: water content, TC: Total Ash, TP: Total proteins, TSS: Total soluble sugar, Rs: **R**educing sugar, α carot: α **c**arotenes, β Carot: βcarotenes, Lut: Luteins, Lyc: Lycopenes

This table shows that, after drying Aloe saponaria flowers in the shade for 24 hours, the water content is 4.24± 0.02%. Ash content is 4.46± 0.03%. Protein content was 7.87± 0.02%, while reducing sugars represented around 15.6% of total soluble sugar content. The carotenoid content is represented by 84.265± 0.275 mg/100g α-carotene, 89.45± 0.02 mg/100g β-carotene, 79.11± 0.02 mg/100g lutein and 25.745 ± 0.025 mg/100g lycopene.

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3.2- Phytochemical composition of Aloe saponaria flower extract

Phytochemical screening of Aloe saponaria flower extract revealed the presence of polyphenols, flavonoids, alkaloids, reducing compounds and saponins, while terpenoids were completely absent.

Table 4. Phytochemical screening results for Aloe saponaria extract

Phenolic compounds		**Results**
Polyphenols		+
Flavonoids		+
Tannins	Gallic	
	Catechics	+
Alkaloids		+
Terpenoids		-
Reducing compounds		+
Saponins		+

+: Presence - : Absence

3.3- Phytochemical content and antioxidant activity

Analysis of the concentrations of bioactive compounds and antioxidant potential of Aloe saponaria flowers shown in Table 4 reveals total polyphenol content of around 484.52 ± 0.02 mg EAG/100g, total flavonoid content of 239.44 mg EQ/100g and tannin content of 79.105± 0.015 mg ECat/100g relative to dry matter.

Table 5. Bioactive compound content and antioxidant activity of Aloe Saponica

Polyph (mg EAG/100 g)	Flav (mg EQ/100 g)	Tannins (mgECat/1 00g)	DPPH (mgET/100gMS)	FRAP (mgET/100gMS)	ABTS (mgET/100gMS)
484,52 ± 0,02	239,44± 0,02	79,11± 0,01	312,46 ± 0,03	225,36 ± 0,01	325,74 ± 0,02

The DPPH anti-radical activity of Aloe Saponica is 312.46 ± 0.03 mgET/100gMS, while its FRAP reduction of iron 3+ to iron 2+ is 225.36 ± 0.01 mgET/100gMS. The ABTS radical scavenging activity of the extract is 325.74 ± 0.02 mgET/100gMS.

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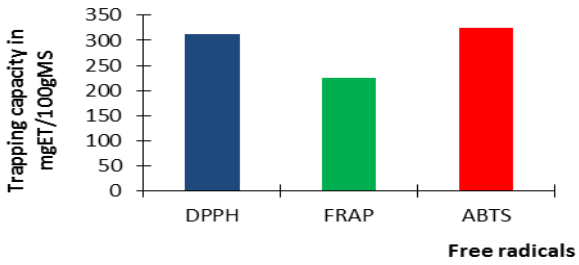
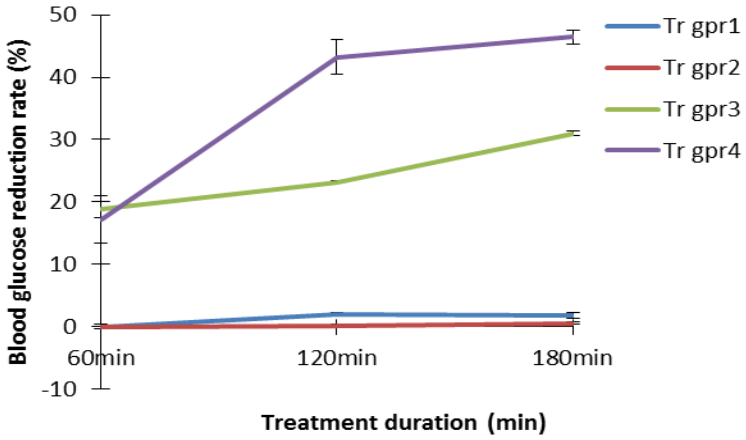


Figure 2. Antioxidant activity of Aloe saponaria flowers

3.4- Hypoglycemic effect of Aloe saponaria flowers

Figure 1 shows the short-term effect of Aloe saponaria extract on blood glucose levels in rats over a 3-hour period. 60 minutes after administration of the experimental treatments, a glucose reduction rate of 17.15% and 18.08% was observed with glibenclamide (TH3) and Aloe saponaria extract at a dose of 500mg/kg body weight (TH2) respectively. The same Aloe saponaria extract at a dose of 100mg/kg body weight (TH1) resulted in a lower blood glucose reduction rate of around 8.04%.

At 120 minutes post-treatment, glibenclamide increased blood sugar reduction by around 26% or 43%, while Aloe saponaria extract at a dose of 500 mg/kg body weight increased efficacy by only around 5% or 23.18%. TH1, on the other hand, reduced blood glucose levels by 6.31%.
After 180 minutes, glibenclamide (TH3) relatively stabilized its blood glucose reduction rate, while Aloe saponaria extract at 500mg/kg body weight (TH2) increased its reduction rate to 30.98%. The efficacy of TH1 continued to fall (3.86%).



Trgpr1: Reduction rate of batch 1, Trgpr2: Reduction rate of batch 2, Trgpr3: Reduction rate of batch 3, Trgpr4: Reduction rate of batch 4.
Figure 3. Evolution of the hypoglycemic activity of the different treatments over time.

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4- Discussion

The consumption of leafy vegetables is an age-old dietary tradition among the populations of the three northern regions of Cameroon in general, and those of the Mindif district in particular. Daily food rations are characterized by regular consumption of the leaves of Hibiscus sabdarifa, Adansonia digitata, Abelmoschus esculentus, Cerathoteca sesanoides and Corchorus olitorius, known locally as Foléré, Bocko, Gombo, Gouboudo and Lalo respectively. A comparative analysis of the nutritional composition and antioxidant activities of Aloe saponaria flowers shows that its protein content (7g/100gMS) is higher than that found by Li (2009) in aloe vera gel, ranging from 0.26±0.01 to 0.54±0.05%. This Aloe saponaria protein content is also higher than that of Hibiscus sabdarifa (2.12g/100gMS), Adansonia digitata (3.03g/100gMS) (Nouhoum et al., 2020), and Abelmoschus esculentus (3.55g/100gMS) according to Ramdhame et al., (2020). On the other hand, with 12.54g/100gMS and 14.2g/100g MS respectively, Corchorus olitorius and Ceratocystis sesanoides have higher protein contents than Aloe saponaria.

The moisture content, dry matter and ash content of Aloe saponaria flowers are much lower than those found by Miranda et al. (2010) on Aloe vera leaves. These differences could be explained by their varietal difference and by the fact that they are different parts of a plant. Indeed, according to Gehin et al. (2006), the physicochemical compositions of different parts of the same plant are quantitatively different.

The presence of proteins, soluble sugars and, above all, the richness of antioxidant carotenoids such as alpha-carotene, beta-carotene, lutein and lycopene suggest the potential nutritional interest of Aloe saponaria flowers. The concentration of β-carotene, in Aloe saponaria flowers, is higher than in gel (1.12 ± 0.03 mg Eq β-carotene) and Aloe vera leaves (1.9 ± 0.01 mg Eq β-carotene per 100 g FP) as found by Bhattacharya et al. (2011). These differences may be due to climatic conditions, plant age, the part of the plant studied and the harvesting season (Lee, 2012). Of the various vegetables regularly consumed in the study area, only Hibiscus sabdarifa has a higher β-carotene content (119 mg Eq of β-carotene /100gMS) than Aloe saponaria flowers (Nouhom et al., 2020).

The presence or absence of a metabolite in Aloe saponaria flower extract depends on the type of solvent and extraction method used, as well as the extraction time. Apolar metabolites are more soluble in apolar organic solvents and less soluble in polar solvents. Conversely, polar metabolites are more soluble in polar solvents and less soluble in apolar organic solvents. The fact that Aloe saponaria flower extract was obtained using methanol would justify the absence of terpenoids and the presence of polyphenols, tannins and flavonoids during screening.

The polyphenol concentration obtained was higher than the results obtained by Megong Moneboulou et al. (2022) on aloe schureenfurthii gel (9.145±0.04 mg EAG/100g MS). Other studies recorded higher levels of polyphenols in Aloe vera dry extract, with contents of 2510.28 ± 4.41 mg EAG/100g and 1138 ± 0.94 mg/100g (Attabi, 2012). The total flavonoid concentration obtained is significantly higher than the results obtained by Attabi (2012) and Aliliche Mustapha

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et al. (2014) on Aloe vera leaves. However, another study recorded a higher content of 363 mg EQ/100g (Ozsoy et al., 2009). Climatic conditions, harvesting time and varietal differences would therefore explain the differences observed. The total polyphenol concentration of the Aloe saponaria sample is higher than that of Corchorus olitotorius (330.67±0.32mEAG mgEAG/100gMS) and Abelmoschus esculentus (13.09±1.4 mgEAG/100gMS), respectively reported by Tsado et al.(2019) Hanzah et al. (2004) and Kouakou et al. (2017). In contrast, Hibiscus sabdarifa has a polyphenol concentration of 121.54±0.04 mgEAG/gMS, (Tsado et al., 2019) which is far higher than that of Aloe saponaria.
In addition to the work of Dicko et al. (2022) on Ceratotheca sesanoïdes, the flavonoid and tannin concentrations of the various vegetables mentioned above are all lower than those of Aloe saponaria flowers. Indeed, polyphenols have been shown to protect plants from the harmful effects of solar radiation (Gehin et al., 2006). Thus, Aloe saponaria, being more exposed to the sun, is likely to produce more polyphenols than other garden vegetables.

The antioxidant potential of the phenolic acids, flavonoids and tannins present in Aloe saponaria flowers is indicative of its importance in the diet of local populations. The effective concentration for scavenging 50% of DPPH radicals, the FRAP reducing activity and the ABTS radical scavenging activity of the extract are all higher than those obtained by Reynolds and Dweck (1997) on Aloe vera leaves and gel. Indeed, according to Ekou and Koné (2018), the polyphenols contained in the various extracts are considered major compounds that contribute to the antioxidant activities of plants. Polyphenols possess inhibitory properties against lipid peroxidation and free radical scavenging properties against superoxide anions (Jamila, 2018). They can act as electron donors, reducing agents, metal chelators and singlet oxygen quenchers (Elmalai et al., 2015). The nature and structure of phenolic compounds in extracts influence the reaction mechanism between antioxidants and the ABTS radical (Huyut et al., 2017). The antioxidant potential of Aloe saponaria flower extracts is higher than those of Abelmoschus esculentus (DPPH 1.03 mg/ml; FRAP 0.89 mg/ml); Ceratotheca sesanoïdes (DPPH 106.85 µg/ml; ABTS 14671±554 µmolEAA/g), Corchorus olitorius (DPPH 37.75 µg/ml) evaluated respectively by Randhan et al., 2020; Fasola and Ogunsola 2014; Scridivin et al., 2020. In general, studies have shown that there is a correlation between the presence of phenolic compounds in an extract and its antioxidant activity (Li et al., 2008).

The results obtained when assessing the hypoglycemic effect suggest that Aloe saponaria flower extract has a dose-dependent activity. Flower extract at a dose of 500mg/kg PC shows a high rate of blood glucose reduction, which increases twofold after 3h of extract ingestion. Even if its hypoglycemic capacity remains lower than that of the reference drug, it is important to emphasize that Glibenclamide is a purified molecule, unlike our extract. Extraction and purification of the active ingredient would be an important step in the valorization of Aloe saponaria flowers for the treatment of hyperglycemia.

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Conclusion

The results of this study show that dietary consumption of Aloe saponaria flowers provides an additional source of vitamins, proteins, sugars and minerals. The presence of phytochemicals, with a predominance of polyphenols, flavonoids and tannins, reinforces its antioxidant potential, which plays an important reparative role in the preservation of living cells. Indeed, the presence of secondary metabolites suggests the pharmacological activities of Aloe saponaria flowers. In addition, this study observed a dose-dependent hypoglycemic activity of Aloe saponaria flowers in albino Wistar rats, in a state of induced hyperglycemia. However, if Aloe saponaria flowers are to be used to combat hyperglycemia, it will be necessary to study their full anti-diabetic activity in vivo (animals and humans), their toxicity and the stability and preservation of the active ingredient over time.

Conflict of interest

The authors declare that they have no conflict of interest in relation to this work.

Authors' Contribution

All authors participated in the realization of this work and in the preparation of the manuscript.

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