
PHYSIOLOGICAL AND BIOCHEMICAL STUDIES ON *GALIUM SINAICUM* PLANT

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Abstract

This research aims to evaluate some of the physiological and biochemical aspects of *Galium sphaericum* plant. Water content, total ash, organic matter, acid soluble and insoluble ash, water soluble and insoluble ash, crude fiber, total carbohydrates, soluble and insoluble carbohydrates, total nitrogen, total protein, total lipids, free amino acids, total phenolics, total flavonoids, total saponins, total tannins and total alkaloids were analyzed in shoots of the tested plant. Preliminary phytochemical screening of *Galium sphaericum* showed that presence of alkaloid, glycosides, cardiac glycosides, saponins, phenols, phytosterols, tannins, flavonoids, amino acids, while volatile oil is absent. The most predominant free amino acids in *Galium* shoots were determined and recorded different variations. Lethal Dose (LD₅₀) of methanol extract 70% of *Galium sphaericum* was 4500 mg/Kg, two specific dose (225 mg/kg and 450 mg/kg) used as hypoglycemic agent of *Galium sphaericum*, evaluation and results showed that the total extract of *Galium sphaericum* plant produced high effect as hypoglycemic agent.

[Elhaw MH, Alldinary MM. Chemical Constituents and Biological Activity of *Galium sphaericum*, Family Rubiaceae]

Keywords: chemical composition, phytochemical, *Galium sphaericum*, hypoglycemic agent, LD₅₀

1. INTRODUCTION

The genus *Galium* L. (Rubiaceae) was described by Linnaeus (1753) who reported that the occurrence of 26 species. Today more than 600 species of *Galium* occur worldwide, mostly in meridional to temperate, but also in alpine and arctic regions, or in subtropical and tropical zones at higher elevations (Chen Tao & Ehrendorfer 2011). From Egypt, Forskal (1775) reported three species of *Galium* *G. pufillum*, *G. verum*, and *G. aparine*; Delile (1813) reported only *G. spurium*; Ascherson and Schweinfurth (1889) reported four species *G. tricorne*, *G. nigricans*, *G. murale* var. *alexandrinum*, and *G. columella*; Sickenberger (1901) accounted for two species *G. murale* var. *rupestris*, and *G. lanatum*; Muschler (1912) wrote about five species *G. tricorne*, *G. spurium*, *G. nigricans* var. *brachychaetum*, *G. murale* var. *variegata*; Ramis (1929) reported six species *G. lanatum*, *G. tricorne*, *G. aparine*, *G. nigricans* var. *brachychaetum*, *G. murale* var. *alexandrinum*, and *G. spurium*; Täckholm (1956) reported eight species *G. sphaericum*, *G. mollugo*, *G. articulatum*, *G. tricorne* var. *ceratopodium*, *G. spurium* var. *tenerum*, *G. murale* var. *alexandrinum*, *G. nigricans*, and *G. setaceum*.

var. *decaisnei*; Montasir and Hassib (1956) reported seven species *G. sphaericum*, *G. tricorne*, *G. spurium* var. *tenerum*, *G. murale* var. *alexandrinum*, *G. nigricans* var. *brachychaetum*, *G. setaceum*, and *G. decaisnei*; Täckholm (1974) distinguished 12 species *G. sphaericum*, *G. canum*, *G. mollugo*, *G. articulatum*, *G. murale*, *G. tricornutum*, *G. ceratopodium*, *G. aparine*, *G. spurium*, *G. nigricans*, *G. setaceum*, and *G. parisiense*; Boulos (2000) recognized only 10 species *G. sphaericum*, *G. canum*, *G. mollugo*, *G. murale*, *G. tricornutum*, *G. ceratopodium*, *G. aparine*, *G. spurium*, *G. setaceum*, and *G. parisiense*; Elkordy (2007) stated that *Galium* in Egypt is represented by 11 species *G. parisiense*, *G. canum*, *G. mollugo*, *G. murale*, *G. aparine*, *G. spurium*, *G. tricornutum*, *G. ceratopodium*, *G. nigricans*, *G. setaceum*, *G. sphaericum*, two subspecies and two varieties.

Diabetes mellitus (DM), often simply referred to as diabetes, is a group of metabolic diseases in which a patient has high blood sugar, either because the body does not produce enough insulin, or because cells do not respond to the insulin that is produced. This high blood sugar produces the classical symptoms of polyuria (frequent urination), polydipsia

(increased thirst) and polyphagia (increased hunger). It is characterized by hyperglycaemia due to defective insulin action, insulin secretion or both. Several medicinal plants are used in the management of diabetes mellitus (kah et al., 2002). According to the World Health Organization (WHO), there are approximately 160,000 diabetics worldwide, the number of diabetics has double in the last few years and is expected to double once again in the year 2025 (Beretta, 2001). Due to its high prevalence and potential deleterious effect on a patient physical and psychological state, diabetes is a major medical concern (Macedo et al., 2002). The disease remains incurable and can only be controlled with drugs. The three main types of diabetes mellitus (DM) are: Type 1 DM results from the body's failure to produce insulin, and presently requires the administration of insulin for treatment (Lambert et al., 2002). It is also referred to as insulin-dependent diabetes mellitus (IDDM) or "juvenile" diabetes). Type 2DM results from insulin resistance, a condition in which cells fail to use insulin properly, sometimes combined with an absolute insulin deficiency (Boussageon et al., 2011). It is formerly referred to as noninsulin-dependent diabetes mellitus (NIDDM) or "adult-onset" diabetes). Gestational diabetes occurs when pregnant patient, who have never had diabetes before, have a high blood glucose level during pregnancy. It may precede development of type 2 DM (Sattar et al., 2010).

Several plants have been used in folkloric medicine for the treatment and prevention of infectious and non-infectious diseases in man and his animals and this has led to renewed scientific interest in the use of plants for these purposes (Oridupa et al. 2011). There is global resurgence in the use of herbal preparations and in some developing countries like Nigeria; it is being gradually integrated into the primary and secondary health care systems. Nearly all societies have used herbal materials as sources of medicines and the development of these herbal medicines depended on local botanical flora (Adedapo et al., 2009).

The aim of this work was to evaluate and determination of some physiological and

biochemical activates for shoots of *Galium sphaerocarpum* plant extract.

MATERIALS AND METHODS:

Plant material:

Galium sphaerocarpum was collected from South Sinai. The taxonomic identification of plant materials was confirmed by Desert Research Center Herbarium team.

Methods:

Eco-physiological study including determination of the percentage of plant water content of *Galium sphaerocarpum* (Rowell, 1994). Determination of certain pharmacopoeial constants of plant material, including inorganic (ash) and organic matter (Brower and Zar, 1984), acid-soluble and acid-insoluble ash, water-soluble and water- insoluble ash (Askar and Treptow, 1993) and crude fibers (British pharmacopoeia, 1980).

Investigation of metabolic products including determination of total carbohydrates, soluble and insoluble carbohydrates (Chaplin and Kennedy, 1994). Total nitrogen and protein content determined by using Kjeldahl method (James, 1995). Free amino acids and protein-amino acids were accomplished according to Pellet and Young (1980) using Amino Acid Analyzer (Beckman system 7300 High Performance analyzer). Total lipids content according to British Pharmacopoeia (1993). Phytochemical study including preliminary phytochemical screening, including steam distillation of volatile oils (Balbaa et al., 1981), test for Alkaloids (Woo et al., 1977), test for glycosides (Treare and Evan, 1985), test for cardiac glycosides (Treare and Evan, 1985), test for saponins (Kokate et al., 2001; Kokate, 1994), test for phenols (Ahmad et al., 2005), test for phytosterols (Brieskorn et al., 1961; Fieser and Fieser, 1959), test for tannins (Treare and Evan, 1985), test for flavonoids (Geissmann, 1962; Khandaal, 2008).

Total phenolics were determined with the Folin Ciocalteu as described by Maurya and Singh (2010), Total flavonoids by Samatha et al. (2012), total tannins by Ali et al. (1991), total saponins according to Obadoni and

Ochuko (2001) and total alkaloids as described by Woo et al. (1977).

Acute Toxicity and Median Lethal Dose (LD_{50}) according to the method of Ecobichon (1997).

Statistical analysis:

The obtained results of the biochemical analyses were statistically analyzed using SPSS (statistical package for social science, Ver.17) following the methods of Snedecor and Cochran (1980). Results were calculated as mean \pm standard deviation. The P values below 0.05 were considered significant.

Experimental Design for anti – diabetic :

Rats were divided into five groups (6 rats each). Rats of group I: Normal control (N.C), group II: Alloxan - intoxicated control received the vehicle in a dose of 140 mg/kg, group III: received standard Insulin at a dose of 0.3 unit per kg. Groups IV & V were treated with M1 (275 mg/kg) & M2 (525 mg/kg.), respectively. All treatments were given orally by gastric intubation for 14 days. On the last day of the treatment, rats of group I were given a single subcutaneous dose of corn oil (3 mL/kg), while animals of the groups II, III, IV & V were received a single subcutaneous dose of Alloxan (140 mg/kg) after 1 h of the vehicle, extract or standard Insulin treatments.

RESULTS AND DISCUSSION:

Table 1. recorded the different determined physiological and biochemical parameters of *Galium sinaicum* plant including water content, organic matter, water insoluble ash, crude fiber, total carbohydrate, insoluble carbohydrate, total nitrogen, total protein and total lipids. Results indicated that, *Galium sinaicum* contained normal values of water content, total ash, organic matter, acid soluble ash , acid insoluble ash, water soluble ash , water insoluble ash, crude fibers, total carbohydrate, soluble carbohydrate, insoluble carbohydrates, total nitrogen total protein and total lipids concerning the natural growing habitat of the tested plant

The water content of plant about 67.33 %. This is expected since the plant is grown in arid desert environment and sandy soils

impoverished in water and nutritive elements with a common shortage of water essential for plant growth. In this regard, Elhaw (2014) the water content is a part of hydrodynamic system, which is terrestrial plant involves absorption of water from the soil, its translocation throughout the plant, and its loss to the environment, principally in the process known as transpiration but the high percentage in water content may be due to many factors such as photosynthesis and lack of lignin in vessel which reduces the size of conductive vessel for unit, size plant. The total ash content of plant about 19.26 %, The high content of ash is useful in assessing the quality of grading the plant and also gives an idea of the amount of minerals present in the sample (Alli Smith, 2009).Organic matter 80.73% , Acid soluble ash 11.2 %, Acid insoluble ash 8.06 %, Water soluble ash 12.23 %, Water insoluble ash 7.03 %, Crude fibers 17.16% . %. Dietary fibers play an important role in human health, which consists mainly of cellulose, hemicelluloses and lignin, which exert different physiological effects on human health (Zia-ur-Rehman et al., 2003).

It had been reported that food, fiber absorption of trace elements in the gut and reduce absorption of cholesterol (Abolaji et al., 2007) .The total carbohydrates content in plant about 30.86 %, Soluble carbohydrate 17.10 %, Insoluble carbohydrates 13.76 %.In this respect, Stroganove et al.(1970) and Kramer,(1983) reported the accumulation of carbohydrates in plants as a response to salinity or drought, despite a significant decrease in net CO_2 assimilation rate. This is also confirmed by global gene expression studies which showed a reduction in the expression level of most genes encoding chloroplast enzymes involved in carbon fixation, while genes encoding cytoplasmic and vascular enzymes in the pathways leading to glucose, fructose and fructose production were up-regulated under drought stress (Xueet et al., 2008), suggesting a coordination in the regulation of transcripts of key enzyme genes involved in carbon fixation and carbohydrate accumulation. Carbohydrates act as nutrient and signaling molecules, modulating the expression of a large number of genes and they are also involved in the

response to abiotic stresses. The minute variations express the prevailing aridity (hyper arid conditions) and open unique environments that contribute to photosynthesis in the same way and magnitude (Osuna et al., 2007). Table (1) showed the total lipids contents in plant of *G.sinaicum* where they ranged from 1.6 %. Monteiro de Paula et al. (1993) and Matos et al.(2001) correlated the decrease in membrane lipid content under water stress to an inhibition of lipid biosynthesis and a stimulation of lipolytic and peroxidative activities. Lipids are among the most structurally diverse families of chemicals in nature and they have a great many biological functions. Certain classes of lipid form membranes; permeability barriers that define cells and compartmentalized all the biochemical processes within them. Other lipids, namely fats and oils, act as nature's most energy rich carbon currency, while others still function as signaling molecules (hormones and secondary messengers) controlling growth, development and responses to the environment. The lipids in different bulbs are important as membrane constituents in the chloroplasts and mitochondria (Harborne, 1998). Total nitrogen in plant about 1.8 %, The total protein in plant about 9.43 %. In this regard Nour El-Din and Ahmed,(2004) found that the increase in soil moisture stress may remarkably increase the assimilation and accumulation of nitrogenous compounds. Align with them, Campalans et al. (1999) showed evidence that protein residues may be altered during drought stress and some proteins are irreversibly damaged by the effect of drought stress and degraded by proteases. They further suggested that proteases mobilize amino acids from proteins to the synthesis of compatible osmolytes.

Table 1. Metabolomics parameter in shoots of *Galium sinaicum*.

Determinations	Percentage (%)
Water content	67.33±0.5
Total ash	19.26±0.1
Organic matter	80.73±0.1
Acid soluble ash	11.20±0.2
Acid insoluble ash	8.06±0.2
Water soluble ash	12.23±0.2

Water insoluble ash	7.03±0.2
Crude fibers	17.16±0.2
Total carbohydrate	30.86±0.2
Soluble carbohydrate	17.10±0.1
Insoluble carbohydrates	13.76±0.2
Total nitrogen	1.8±0.1
Total protein	9.43±0.2
Total lipids	1.6±0.1

Each value is a mean of three replicates + standard error of mean.

Preliminary phytochemical screening:

Table (2). Showed the preliminary phytochemical screening of *Galium sinaicum* plant investigated alkaloids, glycosides, cardiac glycosides, saponins, phenol, sterol, tannins, flavonoids, amino acid and present in plant under investigation. Table (2) showed that Volatile oil was absent. The preliminary phytochemical screening of plant under study showed the presence of alkaloids, glycosides, cardiac glycosides, saponins, phenol, sterol, tannins and flavonoids. Kayani et al. (2007) recorded that, these secondary metabolites are chemicals produced by means of secondary reactions resulting from primary carbohydrates, amino acids and lipids. Total flavonoids are in particular, secondary metabolites which act as strong antioxidant and their accumulation in plant can reduce oxidative damage caused, directly or in directly, by a biotic stress (Yan et al 2017). Flavonoids appear to represent secondary ROS-scavenging system acting only when plants are affected by stress conditions and once primary antioxidant defense system (Fini et al 2011).

Table 2: Preliminary phytochemical screening in shoots of *Galium sinaicum*.

Groups	Tests	Methanol Extracts
Alkaloids	Wagner's reagent	+ve
	Dragendorff's reagent	+ve
Glycosides	Glycosides test	+ve
	Modified borntrager's	+ve
Cardiac glycosides	Legal's test	+ve
	Foam test	+ve
Saponins	Haemolysis tests	+ve
	Ferric chloride test	+ve
Phenols	Libermanburchard's test	+ve
	Salkwaski reaction	+ve
Phytosterols		

Tannins	Gelatin test	+ve
	Lead acetate test	+ve
Flavonoids	Schinodar's test	+ve
	NaOH test	+ve
Amino Acids	Xanthoproteic test	+ve
	Ninhydrin test	+ve
Volatile oil's	Steam distillation	-ve

Table (3) shows the percentages of acid value 15.8 %, iodine value 79.66 %, ester value 101.33 %, saponification value 117.23 %, total flavonoids 269 (mg/gmrutin) and total phenolic acid 358.33 (mg/gmgallic acid), total tannins 1.66%, total saponins 1.76 % and the percentages of total alkaloid 2.46 %.

Phytochemical evaluation was performed for qualitative detection of various chemical constituents which aid in tracing the presence of active entity that elicit a major pharmacological response (Rajan et al., 2011). The plant can synthesize a large variety of chemical substances that are of physiological importance (Kretovich, 2005). Another research focused on various active phytochemical compounds (e.g. flavonoids, polyphenols, terpenoids and saponins) these compounds possess hypolipidemic, anti-tumor or stimulating properties which can reduce the risks of cardiovascular disease and cancer (Craig, 1999).

Flavonoids are a large subgroup of secondary metabolites categorized as phenolic compounds, widely dispersed throughout plants and prokaryotes (Woo et al., 2002). More than 6,500 flavonoids have been identified (Boumendjel et al., 2002). Flavonoids protect plants against various biotic and abiotic stresses, exhibit a diverse spectrum of biological functions and play an important role in the interaction between the plant and their environment (Pourcel et al., 2007). These are categorized according to their molecular structures into flavonols, flavones, flavanones, isoflavone, catechin, anthocyanidin and chalcones (Sandhar et al., 2011). Plants have an almost limitless ability to synthesize aromatic substances, mainly secondary metabolites, of which at least 12,000 have been isolated,

a number estimated to be less than 10% of the total. In many cases, these substances serve as the molecules of plant defense against predation by microorganisms, insects, and herbivores. Further, some of which may be involved in plant odor (terpenoids), pigmentation (tannins and flavonoids), and flavor (capsaicin). However, several of these molecules possess medicinal properties (Mallikharjuna et al., 2007).

Regarding the total active material of *Galium sinaicum* plant result in the table 3 revealed the presence of high quantity of total phenolic acids (358.33 ± 0.8 mg/gmgallic acid) followed by the high content of total flavonoids (269 ± 0.8). Phytochemical constituents are the basic source for the establishment of several pharmaceutical industries. The chemical constituents present in the plant play significant role in the identification of crude drugs. Phytochemical screening is very important in identifying new sources of therapeutically and industrially important compounds like alkaloids, flavonoids, phenolic compounds, saponins, steroids, tannins, terpenoids etc. Some of the potential health benefits of polyphenolic substances have been related to the action of these compounds as antioxidants, free radical scavengers, quenchers of singlet and triplet oxygen and peroxidation inhibitors (Dzingirai et al., 2007).

Lipids are essential component of biological membranes, free molecules and metabolic regulators that control cellular function and homeostasis (Chiang, 2005).

Table 3. Total active materials in shoots of *Galium sinaicum*.

Item	Total active materials
acid value	15.8 ± 0.3
iodine value	79.66 ± 0.2
ester value	101.33 ± 0.1
saponification value	117.23 ± 0.3
Total flavonoids (mg/gmrutin)	269 ± 0.7

Total phenolic acids (mg/gallic acid)	358.33 ± 1.2
Percentage of Total Tannins (%)	1.66 ± 0.2
Percentage of total Saponins (%)	1.76 ± 0.1
Percentage of total Alkaloids (%)	2.46 ± 0.2

Each value is a Mean of Three replicates ± standard error of mean.

Data presented in table (4) show clearly that, the most predominant amino acids in *Galium sinicum* plant are proline 3.45 % and Phenyl alanine 3.11 % followed by Leucine 2.05% .while Tyrosine is The lowest concentrations of than other amino acid , The determined amino acids (according their contents) could be arranged descendingly as : Tyrosine, Aspartic, Lysine, Glycine, Isoleucine, Valine, Glutamic, Histidine, Methionine, Serine , Alanine, Arginine and Threonine is a Same Value, Leucine, Phenyl Alanine And Proline. Numerous studies have appraised the metabolic adjustments of nitrogen metabolism in plants subjected to water deficit and plant survival either during or after the period of stress. They also clarified that marked differences have been found in the amino acids pattern under stress conditions (Naglaa et al 2014). Some other studies have delineated certain amino acids as indicators for drought resistance and defense mechanisms (Stewart and Larcher 1980; Hanson and Hitz 1982; Navari-Izzo et al., 1990).

Table (4): Free amino acidscontents (mg/g. dry wt.) in shoots of *Galium sinicum*

Amino acid	<i>Galium sinicum</i>
Aspartic	0.92
Threonine	1.97
Serine	1.35
Glutamic	1.09
Proline	3.45
Glycine	0.95
Alanine	1.65
Valine	1.01
Isoleucine	0.99

Leucine	2.05
Tyrosine	0.72
Phenyl alanine	3.11
Histidine	1.11
Lysine	0.93
Arginine	1.97
Aspartic	1.21
Methionine	0.92

Biochemical study:

Liver enzymes: Acute Toxicity & Median Lethal Dose (LD_{50}) of *Galium sinicum* extract in rats was 4500 mg/kg (table 6). Therefore, the tested plant can be categorized as safe since substances. Liver plays a vital role in lipid metabolism. It contributes both in exogenous and endogenous cycles of lipid metabolism and transport of lipids through plasma. Synthesis of many apolipoproteins takes place in liver. The Apo lipoproteins are required for assembly and structure of lipoproteins. Lipoproteins play an important role in the absorption of dietary cholesterol, long chain fatty acids and fat soluble vitamins. The transport of triglycerides, cholesterol and fat soluble vitamins from the liver to peripheral tissue and transport of cholesterol from peripheral tissue to liver is by lipoproteins (Mehboob et al., 2007) possessing LD₅₀ higher than 50 mg/kg are nontoxic.

Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galium sinicum*) on serum alanine aminotransferase (ALAT) and Aspartate aminotransferases (ASAT) activities (U/l):

The result in table (6) showed that treatment of rats with Insulin®, Alloxan® for two weeks resulted in a significant ($p \leq 0.05$) increases in ALAT and ASAT activities; while animals orally treated with the aqueous extract of *Galium sinicum*(225 mg/kg and 450 mg/kg) and recorded non-significant changes when all groups were compared to the corresponding values of control group.

Moreover, the rats treated with *Galium sinicum*(225 mg/kg and 450 mg/kg) extracts, showed significant ($p \leq 0.05$) decreases in ALAT and ASAT activity when both were

compared with Alloxan® only treated animals as shown table (6)

Lipid profile:

Effect of Alloxan®, Insulin and herbal aqueous extracts (*Galium sinicum*) on serum cholesterol and triglycerides (mg/dl).

The result in table (7) showed that treatment of rats with Alloxan®, Insulin® for two weeks resulted in a significant ($p \leq 0.05$) increases in cholesterol and triglycerides; while animals orally treated with the aqueous extract of *Galium sinicum* (225 mg/kg and 450 mg/kg) and recorded non-significant changes when all groups were compared to the corresponding values of control group.

In contrast, the animals treated with *Galium sinicum* (225 mg/kg and 450 mg/kg) extracts, showed significant ($p \leq 0.05$) decreases in cholesterol and triglycerides when both were compared with Alloxan® only treated animals as shown table (7)

Renal function:

Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galium sinicum*) on serum creatinine and blood urea (mg/dl).

The result in table (8) showed that treatment of rats with Alloxan®, Insulin® for two weeks resulted in a significant ($p \leq 0.05$) increases in creatinine and blood urea; while animals orally treated with the aqueous extract of either *Galium sinicum* (225 mg/kg and 450 mg/kg) recorded non-significant changes when all groups were compared to the corresponding values of control group.

On the other hand, the animals treated with *Galium sinicum* (225 mg/kg and 450 mg/kg) extracts, showed significant ($p \leq 0.05$) decreases in creatinine and blood urea when both were compared with Alloxan® only treated animals as shown table (8)

Blood glucose:

Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galium sinicum*) on serum blood glucose (mg/dl).

The result in table (9) showed that treatment of rats with Alloxan®, Insulin® for two weeks resulted in a significant ($p \leq 0.05$) increases in blood glucose; while animals orally treated with the aqueous extract of either *Galium sinicum* (225 mg/kg and 450 mg/kg) recorded non-significant changes when all groups were compared to the corresponding values of control group.

On the other hand, the animals treated with *Galium sinicum* ($p \leq 0.05$) decreases in blood glucose when both were compared with Alloxan® only treated animals as shown table (9)

In conclusion, flavonoids represent a remarkable group of plant secondary metabolites and have long been used as traditional medicines with scientifically proven pharmacological benefits. On the other hand, free amino acids are very important as precursors for a lot of biochemical compounds within the plant tissues and then play important roles as indicators for different conditions. They serve vast-ranging medicinal activities that may lead drug discovery with novel and potential therapeutic evidence. Latest research magnifies primarily functional activity of flavonoids as antioxidant against oxidative stress. So the present work need further work to elucidate the cloth relationships between the secondary metabolite of the plant and its application and uses as therapeutic agents.

REFERENCES

- Abolaji, O.A., Adebayo, A.H. and Odesanmi, O.S., (2007): Nutritional qualities of three medicinal plant parts (*Xylopia aethiopica*, *Blighiasapida* and *Parinari polyandra*) commonly used by pregnant women in the Western part of Nigeria. Pakistan Journal of Nutrition, 6(6): 665-668.
- Adedapo, A.A., Jimoh, F.O., Afolayan, A.J., Masika, P.J. (2009): Antioxidant activities of the methanol extracts of the leaves and stems of *Celtis africana*. Rec. Nat. Prod. 3 (1) 23 – 31 .
- Ahmad, B.; Naeem, A.K.; Ghufran, A. and Innamudin (2005): Pharmacological Investigation of *Cassia sophera*, Linn. Var. *purpurea*, Roxb. Medical Journal of Islamic World Academy of Sciences, 15(3): 105-109.
- Akindele, A.J. and Adeyemi, O.O. (2007): Anti-inflammatory activities of the aqueous leaf

Table 6: Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galiumsinaicum*) treated on serum alanine aminotransferase(ALAT) and Aspartate aminotransferases (ASAT)

Parameter	Control	Alloxan	Insulin	GE Low	GE high	L.S.D at %5
ALAT (U/L)	68.5±3.2	102±1.2	81.2±6.1	52±2.4	49.5±2.2	5.19
ASAT (U/L)	78.2±4.4	116.6±2.6	85±4	64±3.2	58.7±1.4	4.27

Table (7): Effect of Alloxan®, Insulin® and herbal aqueous extracts (treated on serum cholesterol and triglycerides of male albino rats compare with controlgroup).

Parameter	Control	Alloxan	Insulin	GE Low	GE high	L.S.D at %5
cholesterol (mg/dl)	93±2.2	108±1.8	85±4.1	90±0.96	85±2.2	11.3
Triglycerides (mg/dl)	76.1±4.8	92±1.8	61.9±3.6	69±3.3	61±3.3	9.2

Table (8):Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galiumsinaicum*treated on serum creatinineand blood ureaof male albino rats compare with controlgroup.

Parameter	Control	Alloxan	Insulin	GE Low	GE high	L.S.Dat %5
Creatinine (mg/dl)	0.8±0.6	1.2±0.2	1.1±0.3	0.67±0.3	0.76±0.1	0.2
Blood urea (mg/dl)	33.7±1.5	46±5.5	41±1.8	39±1.2	39±1.4	6.3

Table (9):Effect of Alloxan®, Insulin® and herbal aqueous extracts (*Galiumsinaicum*treated on serum blood glucose of male albino rats compare with controlgroup.

Parameter	Control	Alloxan	Insulin	GE Low	GE high	L.S.D at %5
Blood glucose (mg/dl)	96±1.5	198±4.1	82±3.9	130±1.2	114±2.0	15.0

- extract of *Byrsocarpus coccineus*. Fitoterapia, 78: 25-28.
- Ali, A.A.; Ross, S.A.; Mesbah, M.K. and El Moghazy, S.A. (1991): Phytochemical study of *Limoniumaxillare* (Forssk.). Bull. Fac. Pharm., 29 (3): 59-62.
- Ali, R.M.; Ahmed, Z.A. and Abdel-Basset, R. (1992): Effect of NaCl salinity and Na-Ca combinations on seed germination and endogenous levels of some metabolites and respiration of *Trigonellafoenumgraecum*L. radicles. Bull. Fac. of Agric., Cairo Univ., 43 (2): 817-828.
- Ali, Z.Y.; Atia, H.A. and Ibrahim, N.H. (2012): Possible Hepatoprotective Potential of *Cynarascolymus*, *Cupressus sempervirens* and *Eugenia jambolana* Against Paracetamol-Induced liver Injury: In-vitro and In-vivo Evidence. Nature and Science,10 (1): 75-86.
- Alli Smith, Y. R. (2009): Determination of chemical composition of *Senna- siamea* (Cassia leaves). Pakistan Journal of Nutritions,8(2):119-121.
- Ascherson, P. &Schweinfurth, G. (1889): Illustration de la Flore d'Egypte. Vol. II, part 1, 2: 82, Le Care.
- Askar, A. and Treptow, H.(1993): In "Quality Assurance in Tropical Fruit Processing."
- Springer Verlag, Berlin, Heidelberg, Germany, 238 pp.
- Balbaa, S.I.(1986): In "Chemistry of Crude Drugs. Laboratory Manual."Faculty of Pharmacy, Cairo University, 195 pp.
- Balbaa, S.I.; Hilal, S.H. and Zaki, A.Y. (1981): In "Medicinal Plants Constituents." 3rd Ed. General Organization for Univ. Books, Cairo, Egypt, 644 pp.
- Beretta, A. (2001): Campanha de prevencao e diagnostico do diabetes realizadapela UNIARARAS .
- Boulos, L. 2000. Flora of Egypt. vol. 2: 229-242. Al Hadara Publishers, Cairo.
- Boumendjel, A.; Pietro, A.D.; Dumontet, C. and Barron, D. (2002): Recent advances in the discovery of flavonoids and analogs with high-affinity binding to P-glycoprotein responsible for cancer cell multidrug resistance. Med. Res. Rev., 22: 512-529.
- Boussageon, R., Bejan-Angoulvant, T., Saadatian-Elahi, M. (2011): "Effect of intensiveglucose lowering treatment on all cause mortality, cardiovascular death, and microvascular events in type 2 diabetes: meta-analysis of randomized controlled trials -BMJ 343: d4169. DOI:10.1136/bmj.d4169 .

- Brieskorn, C.H. and Klinger-Hand Polonius, W. (1961): Triterpenes and sterols in leaves of *Salvia trioloba* and *Pyrus malus*. Arch. Pharm., 294: 380-391.
- British Pharmacopoeia (1993): Published on the Recommendation of the Medicines Commission." International Edition. 2: 146 A.
- British Pharmacopoeia(1980): Published on the Recommendation of the Medicines Commission." Printed in England for her Majesty,s Stationary Office at the University Press., Cambridge, U.K., 2, 561 pp.
- Brower, J.E. and Zar, J.H. (1984): In "Field and Laboratory Methods for General Ecology". Wm. C. Brown Publishers Dubuque, Iowa, 226 pp.
- Campalans, A., R. Messegueer, A. Gody and M. Pages, (1999): Plant responses to drought, from ABA signal transduction events to the action of the induced proteins. Plant Physiology and Biochemistry, 37: 327-340.
- Chaplin, M.F. and Kennedy, J.F.(1994): In "Carbohydrates Analysis-A Practical Approach." Oxford University Press, Oxford, New York., Tokyo. 2nd Ed, 324 pp.
- Chen Tao & Ehrendorfer, F. (2011): Rubiaceae (Galium). – In: Flora of China, Vol. 19, Pp. 104-141, Beijing.
- Chiang J.Y. (2005): Nuclear receptor regulation of lipid metabolism: potential therapeutics for dyslipidemia, diabetes, and chronic heart and liver diseases. Curr.Opin.Investigating Drugs, 6: 994-1001.
- Craig, W.J. (1999): Health- promoting properties of common herbs.Am. J. Clin. Nutr., 70: 791S-499S.
- Delile, A.R. (1813): Description de l'Egypt, Histoire naturelle. T. II, Paris.
- Dzingirai, B.; Muchuweti, M.; Murenje, T.; Chidewe, C.; Benhura, M.A.N. and Chagonda, L.S. (2007): Phenolic content and phospholipids peroxidation inhibition by methanolic extracts of two medicinal plants: *Elionurus muticus*.
- Ecobichon, D.J. (1997): In "The Basis of Toxicity Testing ", 2nd ed.C. R.C. Press, 220pp.
- Elshaw, Mohammed H. M.(2014): Phytochemical studies on some medicinal plants in Egypt. Ph.D. Thesis Fac. Sci. AL-Azhar Univ.
- Elkordy, A. (2007): Biosystematic Studies of the Genus *Galium* L. (Rubiaceae) in Egypt. MSc thesis, Sohag University, Egypt.
- Fieser, L.F. and Fieser, M. (1959): "Steroids". Anstrichmittelfette, seifen., Rein hold publishing, New York, 62(11):1059-1060.
- Fini, A.; Brunetti, C.; Di Ferdinando, M.; Ferrini, F.; Tattini, M. (2011): Stress-induced flavonoid biosynthesis and the antioxidant machinery of plants. Plant Signal.Behav., 6, 709-711.
- Forskal, P. 1775. Flora Aegyptiaca-arabica, Hauniae.
- Geissmann, T.A.(1962): In "The Chemistry of Flavonoids Compounds." Pergamon Press, New York, 483pp.
- Hanson A.D. and W.D. Hitz, (1982): Metabolic responses of mesophytes to plant water deficits. Ann. Rev. PlantPhysiol., 33: 163-203.
- Harborne, J.B. (1998): In "Phytochemical Methods". Chapman and Hall, London, 3rd edition, 60-66.
- James, C.S.(1995): In "Analytical Chemistry of Foods." Blackie Academic and Professional Publisher.An Imprint of Chapman and Hall, 178 pp.
- kah, P.A., Okoli, C.O., Nwafor, S.V. (2002): Phytotherapy in the management of diabetes mellitus. J. Nat. Rem. 2, 1-10 .
- Kayani, S.A.; Masood, A. and Khan, A.K. (2007): Distribution of secondary metabolites in plants of Quetta-Balochistan. Pak. J. Bot., 39(4): 1173-1179.
- Kayani, S.A.; Masood, A. and Khan, A.K. (2007): Distribution of secondary metabolites in plants of Quetta-Balochistan. Pak. J. Bot., 39(4): 1173-1179.
- Kokane C K, Purohit A P and Gokhale SB. (2001): Carbohydrate and derived Products, drugs containing glycosides, drugs containing tannins, lipids and protein alkaloids. Text book of Pharmacognosy, 7, edition: 133 -166, 167- 254, 255-2 69, 272-310, 428-52 3.
- Kokane C.K., (1994): Practical Pharmacognosy, 4th ed., Vallabh Prakasan, Delhi, 107-111.
- Kramer, P.J.,1983. Water relations of plants. Academic Press Inc., New York, USA, 496.
- Kretovich, U.L. (2005): Principles of plant biochemistry. Permagon. Press Oxford, J. food Sci., 54: 254-260.
- Lambert, P., Bingley, P.J. (2002):"What is Type 1 Diabetes?" Medicine 30: 1-5. DOI :10.1383/medc.30.1.1.28264 .
- Linnaeus, C. 1753. Species Plantarum. Stocholm, Impensis Laurentii Salvii.
- Macedo, C.S., Capelletti, S.M., Mercadante, M.C.S., Padovani, C.R., Spadella, C.T. (2002): Role of metabolic control on diabetic nephropathy. Acta Cir. Bras. 17(6), 37 – 5
- Mallikharjuna, P.; Rajanna, L.; Seetharam, Y. and Sharana basappa, G. (2007): Phytochemical studies of *Strychnos potatorum* L.f.A medicinal plant. E. J. Chem., 4(4):510-518.

- Matos, A.R., A. d'Arcy-lameta, M. Franca, S. Peters, I. Edelman, J. Kader, Y. Zuily-Fodil and A.T. Pham-Thi,(2001): A novel patatin-like gene stimulated by drought stress encodes a galactolipid acyl hydrolase. *Plant science*, 491: 188-192.
- Maurya, S. and Singh, D. (2010): *International Journal of Phamtech Research*, 4:2403-2406.p.
- Mehboob, F.; Ranjha, F.A. and Masud, S. (2007): Changes in serum lipid profile among patients suffering from chronic liver disease. *Annals.*, 13 (3): 209-211.
- Montasir, A.H. &Hassib, M. (1956): *Illustrated Manual Flora of Egypt. Part 1.Dicotyledons*.ImprimeriMisr, S.A.E., Cairo.
- Monteiro de Paula F., A.T. Pham Thi, F.Y. Zuily, R. Ferrari-Iliou, J. Vieira da Silva and P. Mazliak, (1993):Effect of water stress on the biosynthesis and degradation of polyunsaturated lipidmolecularspecies in leaves of Vignaunguiculata. *Plant Physiology and Biochemistry*, 31: 707-715.
- Mueller, M. S, Karhagomba, I. B., Hirt, H. M. and Wemakor, E. (2000): The potential of *Artemisia annua* L. as a locally produced remedy for malaria in the tropics: agricultural, chemical and clinical aspects. *J Ethnopharmacol*, 73:487-493.
- Muschler, R. (1912): *A manual Flora of Egypt*, Vol. II: 920, Berlin, R. Fredlaender&Sohn, Berlin.
- Naglaa, M. Sherif, Shadia, A. Fathi, A.B. Sengab, Fareida, M. El-Saied, A.M. Osman and El-Shaimaa, S. El-Demerdash. (2014) : Biochemical Investigations on *Galium sphaericum* Leaves Emphasizing its Utility for Medical use. *Middle East Journal of Applied Sciences*, 4(2): 277-287, 2014ISSN: 2077-4613.
- Navari-izzo, F., M.F. Quartacce and R. Izzo, (1990): Water-stress induced changes in protein and free amino acids in field-grown maize and sunflower. *Plant Physiology and Biochemistry*, 28: 531-537.
- Nour El-Din, N.M. and F.A. Ahmed, (2004): Effect of seasonal variation on secondary metabolites and nutritive value of *Crotalaria aegyptiaca*Benth. *Egyptian Journal of Desert Research*, 54(1): 121-139.
- Obadoni, B.O. and Ochuko, P.O. (2001): Phytochemical studies and comparative efficacy of the crude extracts of some homeostatic plants from Edo and Delta States of Nigeria. *Global Journal of Pure and Applied Sciences*, 8: 203-208.
- Oridupa, O.A., Saba, A.B., Sulaiman, L.K. (2011): Preliminary report on the antiviralactivity of the ethanolic fruit extract of *Lagenaria breviflora* Roberts on NewcastleDisease virus. *Trop. Vet.* 29 (1), 22-33 .
- Osuna, D., B. Usadel, R. Morcuende, Y. Gibon, O. Bläsing, M. Höhne, M. Günter, B. Kamlage, R. Trethewey, W.R. Scheible and M. Stitt, (2007) Temporal responses of transcripts, enzyme activities and metabolites after adding sucrose to carbon-deprived *Arabidopsis* seedlings. *Plant J.*, 49(3): 463-491.
- Pellet, P.L. and Young, V.R.(1980): In "Nutritional Evaluation of Protein Foods." Published by the United Nations University, 546 pp.
- Pourcel, L.; Routaboul, J. M. and Cheynier,V.(2007): Flavonoid oxidation in plants: from biochemical properties to physiological functions. *Trends Plant Sci.*; 12(1): 29 – 36.
- Rajan,s. S. Mahalakshmi, vm. Deepa, k.Sathy, s. Shajitha, t.thirunalaundari 2.(2011): Antioxidant potentials of *punicagranatum* fruit rind extracts. *International Journal of Pharmacy and Pharmaceutical Sciences*. Vol. 3, Issue 3, 0975-1491.
- Ramis, A. I. (1929): *Bestimmungstabellenzur Flora von Aegypten*, Jena.
- Rowell, D.L. (1994): *Methods and Applications*. Longman Publishers, Singapore, 350 pp.
- Samatha, R., Shyamsundarachary, R., Srinivas, P. and Swamy, NR.,Kurz L.(2012): Quantification Of Total Phenolic And Total Flavonoid Contents In Extracts Of *OroxylumIndicum* L .Kurz, *Asian Journal of Pharmaceutical and Clinical Research*, 5(4): 177- 179.
- Sandhar, H.K.; Bimlesh, K.; Sunil, P.; Prashant, T.; Manoj, S. and Pardeep S. (2011): A Review of Phytochemistry and Pharmacology of Flavonoids. *Internationale Pharmaceutica Scienzia*, 1(1): 25-41.
- Sattar, N., Preiss, D., Murray, H.M., Welsh, P., Buckley, B.M., de Craen, A.J., Sesha-sai, S.R., McMurray, J.J., Freeman, D.J. (2010): "Statins and risk of incident diabetes: a collaborative meta-analysis of randomised statin trials". *The Lancet*. 375 (9716), 735 – 42 .
- Sickenberger, E. (1901): *Contribution à la Flore d'Egypte*, 4. leCaire.
- Snedecor, G.W. and W.G. Cochran, (1980): Statistical methods. Oxford and J.B.H. Publishing Co., 7th Ed., U.K.
- Stewart, G.R. and F. Larchar, (1980): The Biochemistry of plants (edsStumpf, P. K. and Conn, E. E.), AcademicPress, New York, USA, 5: 609-635.
- Strogonova, B.P., V.V. Kabanaw, L.P. Lapiana and L.S. Prykhodko, (1970): Structure and function

- of plant cell under salinity conditions. 1st Ed., Nauka Publishing House, Moscow, Russia.
- Täckholm, V. (1956): Students' Flora of Egypt. Cairo.
- Täckholm, V. (1974) Students' Flora of Egypt (ed. 2). Cairo Univ., Cairo.
- Treare, G.E. and Evans, W.C. (1985): Pharmacognosy 17th edition, Bahiv Tinal, London, 149 pp.
- Trease, G.E. (1966): In "Text Book of Pharmacognosy". 8th Ed., Tyndall and Cassel, London, 596 pp.
- Woo HH, Kuleck G, Hirsch AM, Hawes MC. Flavonoids: signal molecules in plant development. In: Bela SB and Michael EB, eds (2002): Flavonoids in cell function. Kluwer Academic/ Plenum Publishers. Norwell, Massachusetts, USA. 2002: 51-60.
- Woo, W.S.; Chi, H.J.; Yun and Hye, S. (1977): Alkaloid screening of some Saudi Arabian plants. *Saengyak Hakhoe Chi* (*Hanguk Saengya K Hakhoe*), 8(3): 109-113.
- Xue, G., C.L. McIntyre, C.L.D. Jenkins, D. Glassop, A.F. Van Herwaarden and R. Shorter, (2008): Molecular dissection of variation in carbohydrate metabolism related to water-soluble carbohydrate accumulation in stems of wheat. *Plant Physiol.*, 146: 441-454.
- Yan a, k; Shijie Zhao, S; Bian, L; Chen ,X. (2017) :Saline stress enhanced accumulation of leaf phenolics in honeysuckle (*Lonicera japonica* Thunb.) without induction of oxidative stress *Plant Physiology and Biochemistry* 112 :326-334
- Zia-ur-Rehman; Islam, M. and Shah, W.H. (2003): Effect of microwave and conventional cooking on insoluble dietary fibre components of vegetables. *Food Chem.*, 80: 237-240.

الملخص العربي

دراسات فسيولوجية وبيوكيميائية على نبات الجاليلوم

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يهدف هذا البحث الى تقييم وتقدير فسيولوجى وبيوكيميائى لمركبات مختلفة فى المجموع الخضرى لنبات الجاليلوم سينيكم. تم تجميع عينات النباتات من بيئتها الطبيعية اثناء مرحلة التزهير واوضحت الدراسة انه يوجد تباين فى محتوى النباتات من الماء والرماد الكلى والمواد العضوية والرماد الحمضى والغير حمضى والالياف غير النقية والكريبوهيدرات السائلة والغير سائلة والمحتوى الكلى لكلا من النيتروجين والبروتين والدهون والفينولات وكذلك المحتوى الكلى للفالفونيدات والتانينات والصابونينات والقلويادات فى المجموع الخضرى للنباتوكذلك اوضحت النتائج انه يوجد تباين فى كميات الاحماض الامينية الحرة . ايضا تم عمل بعض الدراسات البيوكيميائية على فئران التجارب والتى اوضحت ان مستخلص الجاليلوم له تأثير متبادر على معدن سكر الدم اعتمادا على التركيزات المستخدمة.