Original Research Article

HYPOGLYCEMIC EFFECTS OF AQUEOUS AND ETHANOLIC EXTRACTS OF DANDELION (*TARAXACUM OFFICINALE* F.H. WIGG.) LEAVES AND ROOTS ON STREPTOZOTOCIN-INDUCED ALBINO RATS

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ABSTRACT

The effects of aqueous and ethanolic extracts of *Taraxacum officinale* F.H. Wigg. leaves and roots on fasting blood glucose (FBG) levels of normal and streptozotocin-induced diabetic Wistar albino rats (*Rattus rattus*) were studied. Exactly 75 Wistar albino rats weighing between 100–225 g were used for the study, and a total of four groups were created. Two groups were divided into six sub-groups of five rats each for the leaf and root extracts respectively, with the remaining two groups being the normal control rats (NCR) and diabetic control rats (DCR). The two sub-groups were thus; sub I, comprising of sub-groups 1–4 which were for diabetic test rats (DTR) on 6% and 10% of aqueous and ethanolic extracts of leaves respectively, while sub-group 5 and 6 were normal test rats (NTR) on 10% of both extracts of leaves respectively. Same was applicable for sub II which represents the roots extracts. After STZ-induction, the course of hyperglycemia was monitored by estimation of FBG. Then administration of *T. officinale* leaf and root extracts (Aq. and Et.) commenced and lasted for 21 days. Changes in FBG concentration between the NCR and DCR against NTR and DTR treated with two doses (300 mg/kg and 500 mg/kg bodyweight twice a day) of the extracts were evaluated using one way Analysis of Variance (ANOVA). When compared, the FBG levels of the DCR and DTR varied significantly ($P < 0.05$). Whereas the mean conc. of FBG levels of NCR was (4.4 ± 0.12 mmol/l) that of DCR was (27.1 ± 1.59 mmol/l). On administration of 6% and 10% concentration of the extracts to the DTR and NTR, statistically significant decrease ($P \leq 0.05$) in glucose concentration was observed. The FBG level of DTR on 6% leaf extract dropped from 17.1 ± 0.18–9.3 ± 1.86 mmol/l, those on 10% dropped from 18.3 ± 2.58–9.9 ± 1.00 mmol/l. The NTR on 10% leaf extract dropped from 9.9 ± 0.76–7.9 ± 1.00 mmol/l. For the root extract, FBG levels of DTR on 6% and 10% extracts dropped from 15.8 ± 0.18–9.6 ± 2.10 mmol/l and 16.0 ± 0.71–7.5 ± 1.46 mmol/l respectively. The NTR on 10% roots extracts fell from 7.55 ± 1.61–3.4 ± 1.23 mmol/l. The results of this study strongly suggest that although *T. officinale* leaves and roots possess hypoglycemic properties, the roots of ethanolic extraction are relatively more potent and may be beneficial in the management of diabetes.

Keywords: Aqueous (Aq.), Ethanolic (Et.) Extract, Fasting blood glucose (FBG), Hyperglycemia, Hypoglycemia, Streptozotocin (STZ), *Taraxacum officinale*.

1.0 INTRODUCTION

The use of plants as a natural cure for several ailments and diseases is one of the oldest practices by mankind. Hence the need to study medicinal herbs and the knowledge of their uses are imperative over the years since in treatment of disease, it is not the drug used to treat and cure the disease that matters, but while doing this, the conservation of the body organisms is the paramount issue (Robert *et al.,* 2007).

Humans are always in need of help for their many diseases and illnesses and this help comes from nature. The study and understanding of ethno-botanical information, chemical constituents of plants, and the probable therapeutic applications of the plant drug help in improving health problems. Plants play important roles as vessels with chemical constituents that possess pharmacological potentials (Maurice 2003).

Taraxacum officinale F. H. Wigg., commonly known as Dandelion (from the French dent-de-lion meaning lion's tooth) is thought to have evolved about thirty million years ago in Eurasia. They have been used by humans as food and as a herb for much of recorded history (Dijk *et al.,* 2003).

It is an herbaceous perennial plant of the family Asteraceae (compositae), and two major species, *T. officinale* and *T. enythrospermum*, are found as weed worldwide. Both species are edible in their entirety. Like other members of the Asteraceae family, they have very small flowers which are yellowish to orange yellow in colour collected together into a compositae (flower head). Each single flower in a head is called floret and they number 40 to over 100 per head. They grow generally unbranched taproots, producing one to more than ten stems that are typically 5–40 cm tall and sometimes up to 70 cm tall. The leaf margins are typically shallowly lobed to deeply lobed and often lacerate or toothed with sharp or dull teeth.

The chief constituents of dandelion root are taraxacin, taraxacerin, and inulin (a sort of sugar which replaces starch in many of the Dandelion family, Asteraceae), gluten, gum and potash. Dandelions are one of nature's richest green vegetable sources of beta-carotene, from which vitamin A is created (14 000 iμ/100 g leaves vs. 11 000 iμ/100 g in carrots). It is a valuable herb and extremely versatile, as the whole plant can be used for medicinal as well as culinary purposes. Medicinally, dandelion has been considered to be an aperient, diuretic, stimulant, stomachic, tonic, anti-diabetic and detoxicant (Clarke 1997). Forty percent of the mature root is inulin, a mixture of complex carbohydrates known as fructo-oligosaccharides (FOS). Based on clinical studies, intake of FOS significantly increases beneficial bifido-bacteria within the gastrointestinal tract and eliminates pathogens. FOS also stimulates the immune system, increases mineral absorption and suppresses abnormal cell growth. The high levels of FOS in dandelion root and its water extract also help to keep blood sugar levels constant and reduce hyperglycemia (Yashpal 2004).

2.0 MATERIALS AND METHODS

2.1.0 Plant Source and Identification

The plant Dandelion (*Taraxacum officinale*) leaves and roots were sourced from farm lands at Umuzi and Umudim villages in Umudioka Ancient kingdom, Orlu local government area of Imo State and the species was identified and confirmed by Dr. F. N. Mbagwu of the Department of Plant Sciences and Biotechnology, Imo State University, Owerri, Imo State.

2.1.1 Chemicals and Reagents

All chemicals and reagents used were of analytical standard and were obtained from reputable sources.

2.2.0 Preparation and Administration of Streptozotocin (STZ)

The range of diabetogenic dose of STZ is quite narrow and a light overdose may cause

the death of many animals (Lenzen *et al.,* 1996).

5 g of STZ was dissolved in 100 ml of distilled water to give a 5% stock solution of which a single dose of 70 mg/kg body weight was injected intraperitoneally to the rats.

2.2.1 Preparation of Plant Extract

Aqueous Extract

Fresh leaves and roots of the plant (*Taraxacum officinale*) were washed with distilled water to remove debris and contaminants, after which they were dried. The leaves and roots were homogenized into fine powder respectively.

The aqueous pulverized plant leaves and roots were respectively prepared by weighing out 100 g of pulverized leaves and roots into 1 l of distilled water respectively. The resultant mixture was allowed to stand for 24 h with occasional shaking after which it was filtered. The filtrate was evaporated and dried to powder with the aid of a thermostatic water bath at a temperature of 50°C. An aliquot of the extract was prepared by dissolving 6 g in 50 ml and 10 g in 50 ml of distilled water respectively to form the two concentrations which served as stock crude drug and stored at 4°C.

Ethanolic Extract

Fresh leaves of the plant (*T. officinale*) were washed with distilled water to remove debris and contaminants, after which they were dried. The leaves and roots were homogenized into fine powder respectively. 100 g of powdered leaves and roots were soaked respectively in 500 ml of absolute ethanol and the resultant mixture was allowed to stand for 24 h with occasional shaking, after which it was filtered. The filtrate was evaporated with rotary evaporator and dried to powder with the aid of a thermostatic water bath at 45°C.

2.3 Extract Administration

The test rats were administered 300 mg/kg and 500 mg/kg body weight of concentrations of aqueous and ethanolic leaves and roots respectively twice daily using a gavage via intubation for 21 days, according to the experimental plan/grouping.

6% aqueous extract of leaves and roots were prepared respectively by weighing 6 g of the various extracts (aqueous and ethanolic) and dissolved in 50 ml of water.
[6000 mg in 50 ml (3000 mg in 25 ml)]

Each rat was administered 0.5 ml (e.g. 200 g rat) of the solution via intubation twice daily for 21 days.

10% aqueous extract of leaves and roots were prepared respectively by weighing 10 g of the various extracts (aqueous and ethanolic) and dissolving in 50 ml of distilled water.
[10000 mg in 50 ml (5000 mg in 25 ml)]

Each rat was administered 0.5 ml (e.g. 200 g rat) of the solution via intubation twice daily for 21 days.

The mode of administration and treatment of the animals according to their experimental regimen/groups is shown in the Table 2.0

2.4 METHOD OF BLOOD COLLECTION

Blood used for analysis was collected via the tail vein by dilating the tail veins with methylated spirit and xylene after which the tip of the tail is cut off and analysis done immediately with the blood using an automated Accu chek glucometer and strips for fasting blood glucose.

2.4.1 ASSAY METHOD

2.4.2 Fasting Blood Glucose (FBG)

Glucose concentration was determined after an enzymatic oxidation in the presence of glucose oxidase. The hydrogen peroxide formed reacts under catalysis of peroxide with phenol and 4-aminophenazone to form a red violet quinoneimine dye as an indicator which is measured glucometrically and the results are expressed in mmol/l.

Reaction Principle;

Glucose + O_2 + H_2O $\xrightarrow{GoD}$ gluconic acid + H_2O_2

$2H_2O_2$ + 4-aminophenazone + phenol $\xrightarrow{PoD}$ quinonemine + $4H_2O$.

Table 2.0 Feeding illustration

GROUPS / TREATMENT	1	2	3	4	5	6	7	8	9	10	11	12	13	14
No. of rats per group	5	5	5	5	5	5	5	5	5	5	5	5	5	5
Feed + water	+	+	+	+	+	+	+	+	+	+	+	+	+	+
STZ(70 mg/kg) 0.2 ml	-	+	+	+	+	+	+	+	+	+	–	–	–	–
Aq. leaves extract 300 mg/kg	–	–	+	–	–	–	–	–	–	–	–	–	–	–
Aq. leaves extract 500 mg/kg	–	–	–	+	–	–	–	–	–	–	+	–	–	–
Et. leaves extract 300 mg/kg	–	–	–	–	+	–	–	–	–	–	–	–	–	–
Et. leaves extract 500 mg/kg	–	–	–	–	–	+	–	–	–	–	–	+	–	–
Aq. roots extract 300 mg/kg	–	–	–	–	–	–	+	–	–	–	–	–	–	–
Aq. roots extract 500 mg/kg	–	–	–	–	–	–	–	+	–	–	–	–	+	–
Et. roots extract 300 mg/kg	–	–	–	–	–	–	–	–	+	–	–	–	–	–
Et. roots extract 500 mg/kg	–	–	–	–	–	–	–	–	–	+	–	–	–	+

Key: + Indicates that item was administered;
– Indicates that item was not administered.

3.0 RESULTS AND DISCUSSION

Table 3.2.1 FBG values (in mmol/l) of normal and diabetic controls compared to diabetic test and normal test rats treated with aqueous and ethanolic extracts of *T. officinale* leaves.

WEEK / GROUP	1	2	3
NCR	4.5 ± 0.47^{a}	4.5 ± 0.74^{a}	4.3 ± 0.18^{a}
DCR	23.5 ± 0.87^{c}	24.1 ± 3.00^{c}	33.6 ± 0.91^{d}
DTR on 6% Aq. Extract	14.2 ± 2.01^{b}	13.9 ± 1.59^{b}	13.3 ± 1.83^{b}
DTR on 10% Aq. Extract	12.9 ± 2.85^{b}	12.6 ± 2.98^{b}	12.1 ± 2.96^{b}
DTR on 6% Et. Extract	13.6 ± 2.19^{b}	10.6 ± 2.53^{ab}	9.3 ± 1.86^{a}
DTR on 10% Et. Extract	13.4 ± 3.39^{b}	10.8 ± 1.21^{ab}	9.9 ± 1.00^{ab}
NTR on 10% Aq. Extract	6.7 ± 0.50^{a}	6.0 ± 0.55^{a}	5.0 ± 0.84^{a}
NTR on 10% Et. Extract	13.1 ± 1.01^{b}	12.2 ± 0.79^{b}	10.7 ± 1.16^{ab}

Results are Means Standard Deviation of triplicate determinations.
Values in the same column with different superscripts letters are statistically significantly at 95% confidence level ($P \leq 0.05$).

Table 3.2.2 FBG values (in mmol/l) of normal and diabetic controls compared to diabetic test and normal test rats treated with aqueous and ethanolic extracts of *T. officinale* roots.

WEEK / GROUP	1	2	3
NCR	4.5 ± 0.47^{a}	4.5 ± 0.74^{a}	4.3 ± 0.18^{a}
DCR	23.5 ± 0.87^{c}	24.1 ± 3.00^{c}	33.6 ± 0.91^{d}
DTR on 6% Aq. Extract	12.7 ± 3.96^{b}	12.6 ± 3.73^{b}	12.0 ± 3.86^{b}
DTR on 10% Aq. Extract	9.8 ± 1.98^{b}	10.0 ± 2.30^{ab}	9.4 ± 2.08^{a}
DTR on 6% Et. Extract	9.2 ± 1.02^{a}	8.1 ± 0.59^{a}	7.2 ± 0.25^{a}
DTR on 10% Et. Extract	8.4 ± 0.46^{a}	6.8 ± 0.75^{a}	5.5 ± 0.83^{a}
NTR on 10% Aq. Extract	9.7 ± 2.47^{ab}	8.5 ± 2.35^{a}	6.2 ± 0.70^{a}
NTR on 10% Et. Extract	5.4 ± 0.74^{a}	4.1 ± 1.18^{a}	3.4 ± 1.23^{a}

Results are Means ± Standard Deviation of triplicate determinations.
Values in the same column with different superscripts letters are statistically significantly at 95% confidence level ($P \leq 0.05$).

DISCUSSION

The success recorded in the use of streptozotocin (STZ) for the induction of diabetes mellitus through the administration of 70 mg/kg body weight can be attributed to the work of Ferreira *et al.,* 2002. This achievement was confirmed by evaluation of fasting blood glucose concentration. Experimental rats having blood glucose concentrations above 10 mmol/l were considered diabetic as this represents the threshold (Al-Awadi *et al.,* 1991).

Normal control rats maintained a fairly stable level of glucose throughout the study period with a mean value of 4.4 ± 0.12 mmol/l. There was however sustained rise in the level of glucose concentration for the diabetic control rats reaching a hyperglycemic mean level of 33.6 ± 0.91 mmol/l on the last day of the study i.e. third week (see Table 3.2.1).

On administration of *Taraxacum officinale* leaf and roots extracts (aqueous and ethanolic), on the normal treated rats, their fasting blood glucose levels dropped, indicating that the plant especially the root extract has the potency to keep blood glucose levels at normal concentration. However, on administration of the 6% and 10% of the leaves and roots extracts, there was a remarkable decrease ($P \leq 0.05$) in the glucose concentration (especially those treated with 10% ethanolic root extracts) over the period of study (see Tables 3.2.1 and 3.2.2).

A major effect of streptozotocin on the pancreatic system was observed and this helped in maintaining a steady increase on the glucose level of the diabetic control rats.

Since the glucose level of the diabetic treated rats dropped over the period, when compared to that of the diabetic control rats, it gives credence and suggests a possible β-cell recovery (Okamoto 1970). Normal control rats given water had no significant change ($P > 0.05$) in the fasting blood glucose levels, while the normal test rats following administration of the leaves and roots extracts (aqueous and ethanolic) showed significant decline in fasting blood glucose. It has been suggested that tannins may lower the rate at which starch is digested and hence blood glucose level by the same mechanism that makes them anti-nutrients. It is also known that tannins may bind directly with pancreatic amylase (the principal enzyme in starch digestion) thus inactivating it. Tannins may also bind with calcium which is needed to stabilize amylase activity or with starch to

influence its degree of gelatinization or its accessibility to the digestive enzymes (Thompson 1993).

In summary, this study has provided some evidence for the hypoglycemic effects of Dandelion (*Taraxacum officinale*) leaves, compared to its roots and the most effective mode of extraction (aqueous or ethanolic) as well as the most effective dosage to be used in terms of drug other than food, in the management of diabetes mellitus.

4.0 CONCLUSION

This study has provided comparative evidence of the hypoglycemic effects of *Taraxacum officinale* leaves and roots of aqueous and ethanolic extraction, on normal rats and streptozotocin-induced diabetic rats. It has shown that the extracts (especially the ethanolic extract) has the potential to improve carbohydrate metabolism. It also indicates that ethanolic extraction of this plant is most suitable compared to aqueous and the roots are more effective compared to the leaves in the management and treatment of diabetes. The experimental findings also indicate that *Taraxacum officinale* extracts is dose dependent.

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