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RESEARCH ARTICLE

IDENTIFICATION OF ANTIDIABETIC COMPOUNDS IN AN ANTIDIABETIC RECIPE USED IN TRADITIONAL CONGOLESE MEDICINE

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Abstract

The recipes of plants used in traditional medicine, prepared by traditional practitioners and preserved in unpalatable conditions of medicines, are in the majority void of scientific information. In order to highlight the phytochemical composition of antidiabetic drugs used in traditional congolese medicine, chemical analyses were carried out on one of the beverages called "NGOSI," purchased from Congolese traditional therapists. The results obtained showed the predominance of polyphenols, sugars, flavonoids and triterpene glycosides in this recipe. 48 compounds were observed in this recipe, including 26 compounds identified by CG/MS, 10 suggested compounds identified by HPLC/MS and 12 non-identity compounds according to the databases used. Some compounds present in this recipe, such as scopoletin, which is proven to be antidiabetic, and vanillin, recognized as an antioxidant capable of significantly reducing serum glucose and triglyceride levels and increasing insulin levels, would justify the antidiabetic activity of this recipe and its traditional use by the congolese population.

Introduction:-

For millennia, plants have played a vital role in human health and well-being. In fact, more than 25% of drugs prescribed in industrialized countries are derived directly or indirectly from plants. They contain a multitude of bioactive substances including alkaloids, polyphenols, terpenes, and many other compounds that may have beneficial effects on health (**Newman, 2000; Leduc, 2006**).

Faced with the insufficient coverage of health needs and limited access to essential medicines, the frequent use of these medicinal plants and the satisfactory results on the treatment of diseases by these plants in the world, particularly in Africa, still arouse a craze for scientific research to promote phytotherapy**(Bouquet 1969; Adjanohoum et al., 1979; Bouquet, A. 1972).** Among these researches, there are those carried out on plants known to be antidiabetic used in traditional African medicine.

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Indeed**,** diabetes is a disease that is currently a real public health problem. For example, it affects approximately 537 million people worldwide, including 6.7 million deaths recorded in 2021. According to the International Diabetes Federation's estimate, this figure would exceed 592 million in 2035 (**IDF, 2021)**.

In Congo, the prevalence of diabetes was estimated at 7% in 2015 according to the Diabaction Congo Association. Diabetes is the leading cause of blindness, end-stage chronic renal failure and lower limb amputations in this country (**Angalla Affleck etal., 2023**). However, modern and conventional treatments are very expensive, often unavailable for long periods of time and not accessible to all social classes (**Kirby, 1996**).

To provide natural solutions in the management of diabetes, many people turn to alternative therapies, including herbal treatments, which are effective in the long term. However, some traditional medicines used and well known by the african population, congolese in particular, have little or no scientific data.

This is how we became interested in an antidiabetic remedy called the "NGOSI" recipe made up of six medicinal plants. This recipe was developed by Mr. KHADET Daniel, a Congolese traditional practitioner and founder of CENACLE (Center for Research and Treatment by Revealed Plants) recognized by the WHO and authorized by the Ministry of Health in the Republic of Congo. "CENACLE" is located south of Brazzaville, (Bifouiti District, Arrondissement 1 Makélékélé, Republic of Congo). A previous study conducted on this recipe confirmed its effectiveness against diabetes, with a blood sugar reduction percentage of 68.03%. It revealed the presence of polyphenols, sterols, terpenes, sugars and glycosides in this recipe (**Koubakaet al., 2023**).

In line with this work, we are continuing the investigation to identify the bioactive compounds and provide scientific information on the chemical composition of this recipe, in order to justify its use by the congolese population.

In order to continue this work and provide scientific information on the chemical composition of this recipe to the traditional practitioner, chromatographic and spectrophotometric analyses were carried out in order to detect the compounds responsible for the biological activities of this traditional medicine.

Materials and Methods:-

Preparation of plant extract and its fractions

The extract used in this work was purchased and made up of 14 liters of the antidiabetic herbal tea (aquous extract) of plants called "NGOSI" recipe prepared according to traditional methods at CENACLE. It was filtered and concentrated to obtain an initial extract (K_0).

Selective liquid-liquid fractionation with solvents of increasing polarity (chloroform, methylethylketone MEC, methanol, ethanol-water) was performed on the aquous extract K0 and allowed to obtain respectively the fractions K1, K2, K3, K4 after evaporation of the solvents. In addition, the fraction MEC1 was obtained directly by extraction with MEC of theaquous extract (K0).

Chemical analyses of the extract

Phytochemical screening of recipe fractions

The different fractions obtained were subjected to screening by thin layer chromatography (TLC) following the methods described in the literature (**Merck, 1980)**. Silica TLC plates (60 F254, aluminum support, 20×20) as well as the different eluents and developers were used to identify the different chemical metabolites.

GC/MS and HPLC/MS analysis of the recipe and its fractions

The initial extract of recipe K_0 and its fractions K_3 and MEC_1 were analyzed by GC / MSand HPLC/MS. Thus, 1 mg of the sample to be analyzed was dissolved in 1000 µL of methanol, HPLC grade. The resulting solution was then passed through a 0.45 µm porosity filter. The sample conditions are summarized in Table 1.

Table 1:-Characteristics of devices and conditions of analysis by GC/MS and HPLC/MS of the recipe and its fractions.

Types of analysis	Device Feature	Analysis conditions
GC/MS	Gas chromatography (Agilent 7890B GC)	Column , 5% Phenylmethylsilox; 0 -325°C; 30m x 250µm x 0.25µm Temperature Gradient

		- Initial temperature: 30°C maintained for 3 min - Gradient: up to 320°C at a rate of 10°C/min - Final temperature: 320°C maintained for 3 min Injector: Split, 1 µL (T° 320 °C)
	Mass Spectrometer (MSD System-5977A)	Electron impact ionization Analyzer: simple quadrupole Library: NIST
HPLC/MS	Ultra-high pressure liquid chromatography (Thermo ultimate 3000)	- Column type: C18, 130 A; 150 mm - Amount injected 5.0 µL - Elution solvent initial conditioning 7 min, WATER + HCOOH 0.1% ACN + HCOOH 0.1% - Speed - column temperature 60°C
	Mass spectrometer	- ESI-HR Ionization

Results and Discussion:-

Results:-

Highlighted the metabolites of the antidiabetic recipe

TLC screening revealed six major families of phytochemical compounds (**fig. 1**), namely terpenes, sterols, polyphenols, saponins and glycosides, and sugars.

Thus, observation of the chromatograms (1, 2, 3A, 3B) of the fractions of the recipe revealed with antimony chloride, Lieberman-Büchard and sulfuric vanillin show spots of violet, blue, green, red, yellow and purplish pink colorations characteristic of the presence of terpenes, sterols, glycosides and saponins**(fig. 1) (Harborne, 1998 ; Wagner and Bladt, 1996).**

Chromatograms 5 and 6 show spots of violet and blackish green color after revelation with sulfuric naphthol. These spots, characteristic of sugars according to Wagner and Harborne, have frontal ratios coinciding with the glucose and fructose controls. This information suggests the presence of soluble sugars in the recipe **(fig. 1)** (**Wagner and Bladt, 1996; Harborne, 1998).**

The chromatographic profile (chromatogram 4), after revelation with NEU, shows yellow, orange-yellow, blue, green, red and orange spots. These colours are characteristic of polyphenols (**Wagner and Bladt, 1996; Harborne, 1998).** The similarity of the colours of certain spots to those of the controls (Quercetin, Rutin and Kampferol) would indicate that flavonoids are the majority subfamily of polyphenols present in the recipe. According to the Frontal Report, Rutin or its derivatives could be present in this antidiabetic recipe.

Identification of compounds in the antidiabetic recipe

The chromatogram of the methanolic fraction K3 of the recipe obtained by GC/MS analyses reveals the presence of several compounds, of which 25 compounds were identified (**Fig. 2 and Table 2**). The majority of these compounds are multifunctional organic molecules with a dominance of phenolic compounds. Peaks 2, 3, 5, 6, 7, 8, 9, 13 and 14 correspond to compounds having a phenolic core; peaks 12, 15, 22 and 23 correspond to compounds having an ester function and peaks 1 and 9 would correspond to compounds having an aldehyde function. The presence of alcohol and ketone functions is also noted in some compounds identified (**Table 2**).

Furthermore, the chromatograms obtained by HPLC/MS analyses of the K0 extract and the MEC1and K3 fractions showrespectively 07, 13 and 6 peaks including 04 compounds presenting a similarity of retention times and massesbetween the initial K0 extract and the MEC1 fraction. A total of 22 compounds observed in the "NGOSI" recipe (**fig.3, 4, 5 and Table 3**). According to the data on the ionic and molecular fragments of each compound in negative and positive ion mode over a m/z range from 0 to 1500 amu, the 04 compounds contained in k0 and MEC1 present mass spectra with superimposable fragments. Based on identical data, these 04 compounds could be identical or racemic isomers.

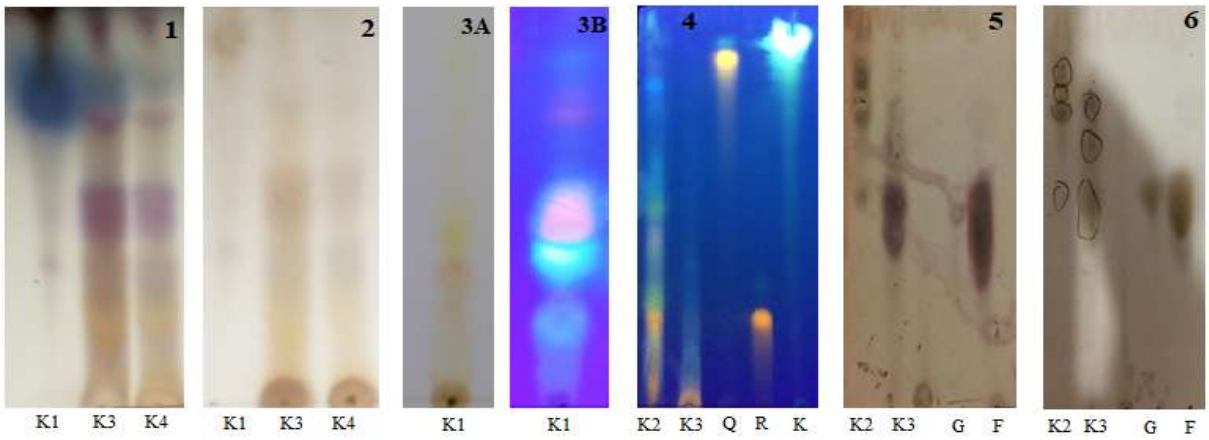


Figure 1:- Chromatograms observed on thin layer.

K1: Chloroform fraction, **K2**: MEC fraction, **K3**: Methanolic fraction, **K4**: Hydroethanolic fraction, **F**: Fructose, **G**: Glucose, **K**: Kampferol, **Q**: Quercetin, **R**: Rutin.

1: Observation: Visible, Developer: Sulfuric vanillin + heating, Eluent: AcOEt/HCO_2H/H_2O (8/1/1);
2: Observation: Visible, Developer: $SbCl_3$ + heating, Eluent: AcOEt/HCO_2 H/H_2O (8/1/1);
3: Observation: Visible (3A) and UV 365 nm (3B), Developer: Lieberman-Buchard, Eluent: C_6H_{14} /AcOEt (7/3);
4: Observation: UV 365 nm, Developer: NEU, Eluent: $CHCl_3$/Acetone/MeOH (5/7/1);
5: Observation: Visible, Developer: Naphthol-sulfuric + heating, Eluent: BuOH/Acetone/AcOH/H_2O (6/1/1/0.5);
6: Observation: Visible, Developer: $SbCl_3$ + heating, Eluent: BuOH/Acetone/AcOH/H_2O (6/1/1/0.5).

The fragments (m/z = 113.00; m/z = 174.97; m/z = 242.98 and m/z = 248.98) are present in most molecules and could be fragments of the basic structures common to all compounds detected in the recipe.

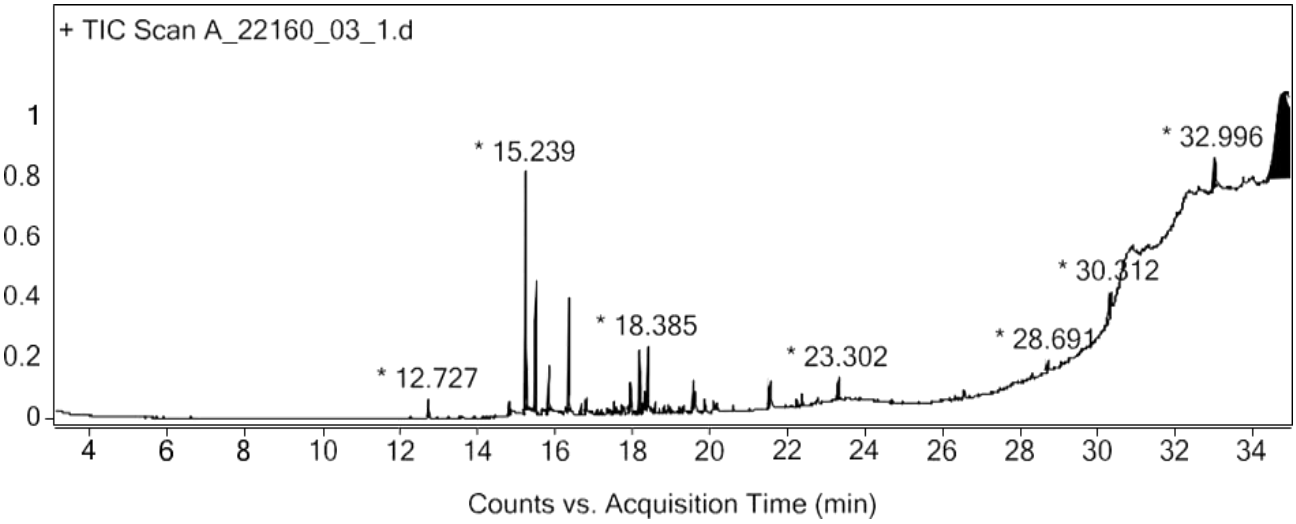


Figure 2:-GC chromatograph of the K3 fraction of K0.

Table 2:-Compounds identified by GC/MS from the K3 fraction of K0.

No.	Tr (min)	Mass (g/mol)	Raw Formula	Compound identified
1	12,727	132.16	C_9H_8O	Cinnamaldehyde (2-Propenal, 3-phenyl)
2	14,810	122.12	$C_7H_6O_2$	Benzaldehyde, 4-hydroxy-
3	15,239	152.15	$C_8H_8O_3$	Vanillin
4	15,486	162.20	$C_{10}H_{10}O_2$	4,7-Methano-1H-indene-1,8-dione, 3a,4,7,7a-tetrahydro-

2	5315,830	136.15	$C_8H_8O_2$	Acetophenone, 4'-hydroxy-
6	16,345	166.17	$C_9H_{10}O_3$	Ethanone, 1-(3-hydroxy-4-methoxyphenyl)
7	16,656	220.35	$C_{15}H_{24}O$	ButylatedHydroxytoluene
8	16,796	162.18	$C_{10}H_{10}O_2$	Benzaldehyde, 2-hydroxy-3-(2-propenyl)-
9	17,505	180.20	$C_{10}H_{12}O_3$	4-Ethoxy-3-anisaldehyde
10	17,945	192.30	$C_{13}H_{20}O_3$	1- Oxaspiro[4.5]deca-3,6-diene, 2,6,10,10-tetramethyl
11	18,170	210.31	$C_{13}H_{22}O_2$	2-Cyclohexen-1-one, 4-(3-hydroxybutyl) -3,5,5-trimethyl-
12	18,320	238.36	$C_{15}H_{26}O_2$	Geranylisovalerate
13	18,385	182.17	$C_9H_{10}O_4$	Benzaldehyde, 4-hydroxy-3,5-dimethoxy
14	19,555	234.33	$C_{15}H_{22}O_2$	3,5-di-tert-Butyl-4-hydroxybenzaldehyde
15	19,598	238.32	$C_{14}H_{22}O_3$	Acetic acid, 2-(2,2,6-trimethyl-7-oxa-bicyclo[4.1.0]hept-1-yl)-propenylester
16	19,845	199.17	$C_5H_9N_7O_2$	Pyrimidine, 2,4-dihydrazino-5-nitro-6-methyl
17	20,081	210.17	$C_9H_9F_2NO_2$	3-Amino-3-(2,4-difluoro-phenyl)-propionic acid
18	20,167	224.30	$C_{13}H_{20}O_3$	2-Heptanone, 6-(3-acetyl-1-cyclopropen-1-yl)-3-hydroxy-6-methyl-, (R*,R*)
19	21,530	192.16	$C_{10}H_8O_4$	Scopoletin (coumarin derivative)
20	21,843	360.35	$C_{17}H_{22}O_5$	2-H-Cyclohepta[b]furan-2-one, 6-[1-(acetyloxy)-3-oxobutyl]-3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-
21	22,346	264.32	$C_{15}H_{20}O_4$	Abscisic acid (phytohormones)
22	23,302	350.40	$C_{19}H_{26}O_6$	Propanoic acid, 2-methyl-,dodecahydro-6a-hydroxy-9a-methyl-3-methylene-2,9-dioxoazuleno[4,5-b]furan-6-yl)methyl ester, 3aS-(3a.alp)
23	26,544	436.60	$C_{26}H_{44}O_5$	Ethyl iso-allocholate
24	28,691	548.60	$C_{28}H_{36}O_{11}$	H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one, 3,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a, 1b,2,3,4,4a,7a,7b,8,9,9a-dodecahy
25	30,312	787.1	$C_{44}H_{85}NO_8P$	3,5,9-Trioxa-5-phosphaheptacos-18-en-1-aminium, 4-hydroxy-*N*,*N*,*N*-trimethyl-10-oxo-7-[(1-oxo-9-octadecenyl)oxy] -, hydroxide, inner salt
26	32,996	548.6	$C_{28}H_{36}O_{11}$	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one, 3,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a ,1b,2,3,4,4a,7a,7b,8,9,9a-dodecahy

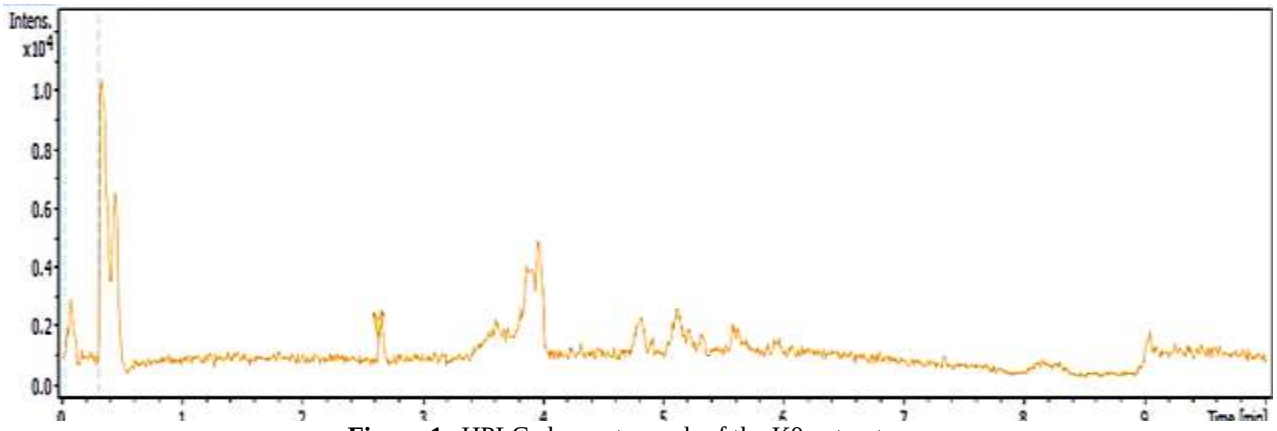


Figure 1:-HPLC chromatograph of the K0 extract.

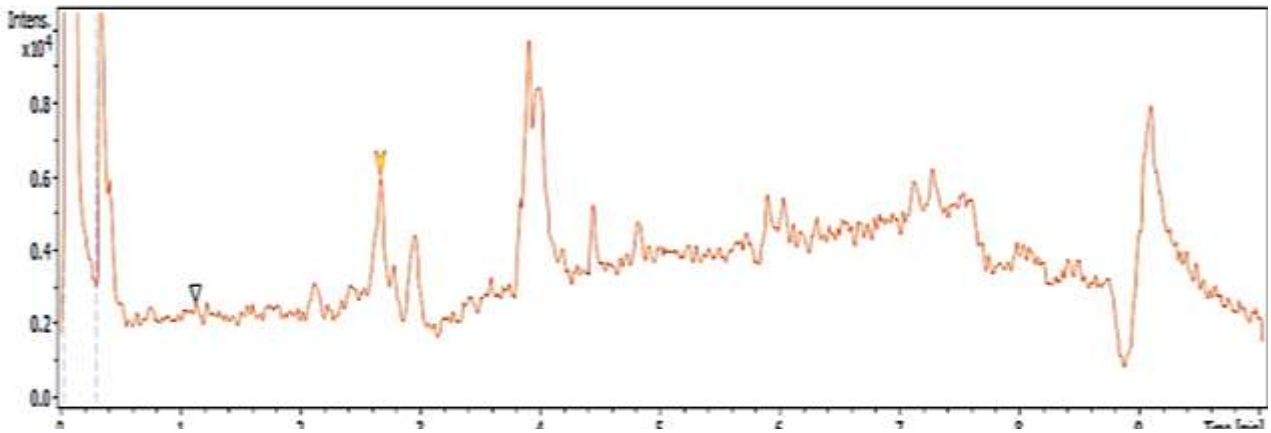


Figure 4:- HPLC chromatograph of the MEC1 fraction of K0.

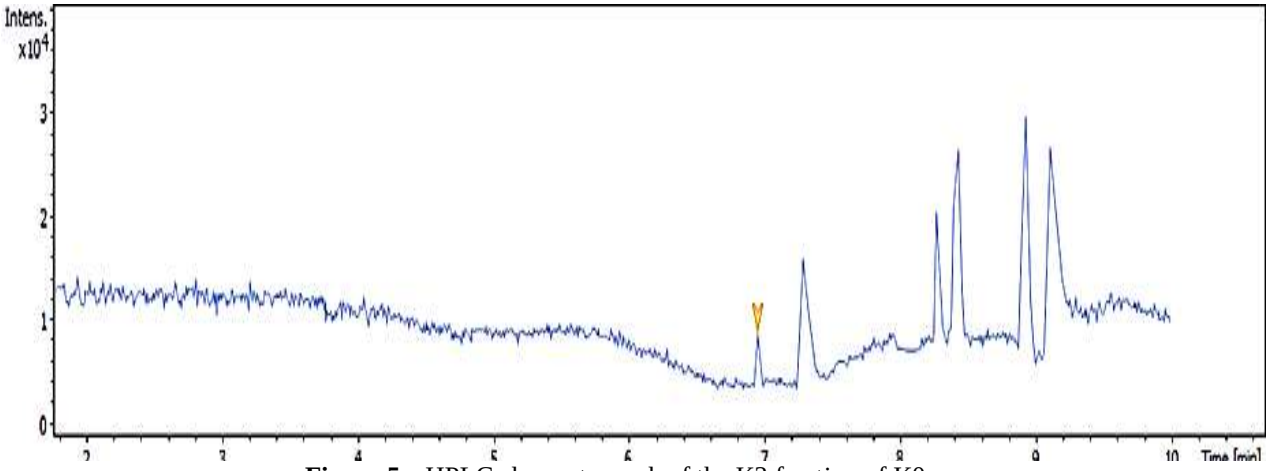


Figure 5:- HPLC chromatograph of the K3 fraction of K0.

Table 3:-Attempted identification by HPLC/MS of compounds observed in K0, K3 and MEC1.

No.	Tr (min)	m/z	Other important m/z fragments	K_0	MEC_1	K_3	Identification attempt (*)
1	2.1	459,1516	248.9859; 174.9794; 113.0018	-	+	-	NI
2	2.7	577,1964	242.9726; 174.9789; 113.0016	+	+	-	Sitosterylglucoside
3	2.8	174.9775	242.9677; 113.0000; 623.2400	-	+	-	NI
4	3.0	137,0400	174.9758; 248.9962	-	+	-	Tyrosol
5	3.6	397,1370	242.9718; 174.9810; 113.0042	+	+	-	Sitosterol
6	3.9	397,1384	242.9803; 174.9849; 113.0041	+	+	-	Sitosterol
7	4.5	373,2227	242.9734, 113.0015	-	+	-	NI
8	4.8	311,2054	174.9838; 113.0044	+	-	-	Citrostadienylaglycone
9	4.8	277,1757	242,9732; 174,9788, 113,0016	-	+	-	NI
10	5.1	325,2241	248.9933; 174.9833, 113.0044	+	-	-	NI
11	5.6	339,2374	248.9932; 174.9799; 113.0042	+	-	-	Esculin
12	5.9	249,1768	174.9767; 113.0027	-	+	-	NI
13	6.0	311,2335	248.9854; 174.9779; 113.0022	-	+	-	NI
14	7.1	353,2879	248.9923; 113.0017	-	+	-	NI
15	7.3	433,3142	248.9897; 113.0024	-	+	-	NI
16	9.1	134.8855	174.0000; 242.9699	+	+	-	NI
17	6.9	282,2895	304,2723; 378,3105; 457,3677	-	-	+	NI
18	7.3	406,3392	384.3598; 338.3534	-	-	+	NI
19	8.3	427,3871	437.2466; 344.7982	-	-	+	Citrostadienol

20	8.4	663,4730	495,3759	-	-	+	PJS-1 sapogenin
21	8.9	547,4110	569,3950	-	-	+	Cholesterylglucoside
22	9.1	547,4128	392,2270; 437,2245; 613,3621	-	-	+	Cholesterylglucoside

Present: +, Absent: -, Not identified: NI,Identified according to the bibliography: **(*)**

Discussion:-

The highlighting by TLC of the different families of compounds made it possible to establish a qualitative phytochemical profile (terpenes, sterols, polyphenols, saponins, glycosides and sugars) of the recipe (**Figure 1**). This profile is in agreement with the results obtained in GC and HPLC. Because, the methanolic fraction K3 rich in polar compounds observed in TLC showed multifunctional compounds in GC and HPLC with very high molecular weights.

GC/MS analysis of the recipe shows that the recipe contains compounds of plant origin including vanillin isolated from *Vanilla planifolia*BD Jacks.(Known to have antioxidant effects), Scopoletin isolated from *Scopoliacarnolica*Jacks. (plant with multiple pharmacological properties), abscisic acid which is a phytohormone allowing plants to fight against their environmental stress and ethyl-iso-allocholate which is present in the leaves of *Sesamumradiatum*L. **(Demarne F, 1996** ; **Rollinger et al., 2004 ; Koussa et al., 2002**)**.** However, ethyl-iso-allocholate is a derivative of a bile acid (cholic acid) of animal origin and its presence in this recipe could be justified by exposure of the plants to animal waste or also the presence of an animal organ in the recipe **(Singh J et al., 2019**). According to the bibliography, some compounds present in this recipe have not yet been identified in the plant world; but, may be derived from molecules present in plants (Cinnamaldehyde (2-Propenal, 3-phenyl); Benzaldehyde, 4-hydroxy-; 4,7-Methano-1H-indene-1,8-dione, 3a,4,7,7a-tetrahydro-; Acetophenone, 4'-hydroxy-; Ethanone, 1-(3-hydroxy-4-methoxyphenyl); Benzaldehyde, 2-hydroxy-3-(2-propenyl)-; Benzaldehyde, 4-hydroxy-3,5-dimethoxy; etc) or could be the result of an interaction of phytocompounds (4-Ethoxy-3-anisaldehyde; 1-Oxaspiro[4.5]deca-3,6-diene, 2,6,10,10-tetramethyl; 2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl-; 3,5-di-tert-Butyl-4-hydroxybenzaldehyde; Geranylisovalerate; etc) **, (Kumar et al., 2018; Lila MA and Raskin I., 2005; Li R et al., 2022).**

HPLC/MS analysis of the fractions in K0, K3 and MEC1 from this antidiabetic recipe did not identify any compounds according to the database used. However, the retention times and peaks of the molecular masses and fragments obtained from these analyses allowed us to suppose some structures of the compounds published and recognized in the literature.The flavonoids, triterpene sterols and sugars contained in this recipe and revealed by TLC, would have a structural variability due to the shape of the skeleton, the presence of substituents and the hydroxyl function. Which would make it difficult to validate any peak to a molecule already scientifically recognized **(Ma R et al., 2020)**.

Therefore, the fragmentation patterns of mass spectrometry are theoretically varied depending on the instrumentation used. Several authors have reported that the main fragmentation pathways of flavonoids and sterols are apparently independent of the ionization mode (ESI, APCI or MALDI) or the types of analyzers used (triQ, IT or QTOF) **(Domon B. & Costello C E., 1988**).

Based on the results obtained from TLC and mass spectroscopy, the compounds contained in the recipe could have structures corresponding to the observed fragments. Therefore, a proposal of some compounds observed by HPLC/MS was made on this basis (**Table 3).** There is therefore a possibility of fragmentation of the triterpene sterols of this recipe as represented by fragmentation a (**fig.6**), thus giving the fragment m/z = 113 which could correspond to the smallest fragment encountered in all the analyzed extracts, the fragment m/z = 173 would correspond to a fragment having lost a hydrogen m/z = [174 - H] $^{\cdot}$, the fragment m/z = 242 could correspond to the fragment having lost a methyl group m/z = [257- CH_3] $^{\cdot}$ of fragmentation b (**fig.6**) **(Rossmann et al., 2007 ; Hailat IA, 2014) .**

- Peak 2 would correspond to Sitosterylglucoside with molecular weight M = 578 characterized by its molecular ion [M - H] $^{\cdot}$ having lost a hydrogen atom (m/z = 577) (**Münger et al., 2018**).
- Peak 3 could correspond to Tyrosol with molecular weight M = 138, characterized by its molecular ion [M - H]$^{\cdot}$having lost a hydrogen atom (m/z = 137) (**Olmo-Garcíaet al., 2018).**
- Peaks 5 and 6 of molecular ion m/z = 397.14 could correspond to the basic structure of triterpene sterols, the most stable ion of Sitosterol (**Müngeret al., 2018**).

- Molecular ion peak 8 m/z = 311.2054 would correspond to a stable ion that is formed during the fragmentation of the aglycone of Citrostadienylglucoside (**Müngeretal., 2018**).
- Peak 11 could correspond to Esculsin with molecular weight M = 340 whose molecular ion $[M - H]^-$ is represented by the fragment m/z = 339 (**Olmo-Garcíaetal., 2018).**
- Peak 19 could correspond to Citrostadienol with molecular weight M = 426 characterized by an ion $[M + H]^+$ having acquired a hydrogen (m/z = 427) (**Müngeretal., 2018**).
- Peak 20 could correspond to PJS-1 Sapogenin with molecular weight M = 617 whose precursor ion $[M + HCOOH]^+$; which is represented by m/z = 663 **(Li et al., 2010).**
- Peaks 21 and 22 could correspond to a single compound or a group of isomers of Cholesterylglucoside with molecular weight M = 546 characterized by the molecular ion $[M + H]^+$ having gained a hydrogen atom (m/z = 547) (**Müngeret al., 2018)**.

The plant metabolites identified in this antidiabetic recipe may be responsible for the biological properties of the latter. Because, polyphenols are capable of producing a hypoglycemic effect by increasing the uptake of glucose by peripheral tissues, flavonoids control certain disorders associated with diabetes and certain sugars promote the stimulation of insulin secretion (**Chattopadhyay, 1999; Bonnefont-Rousselot et al., 2000)**. In addition, sterols and triterpenes are endowed with anti-inflammatory property that could be used to prevent inflammation in diabetics (**Bruneton, 1999**).

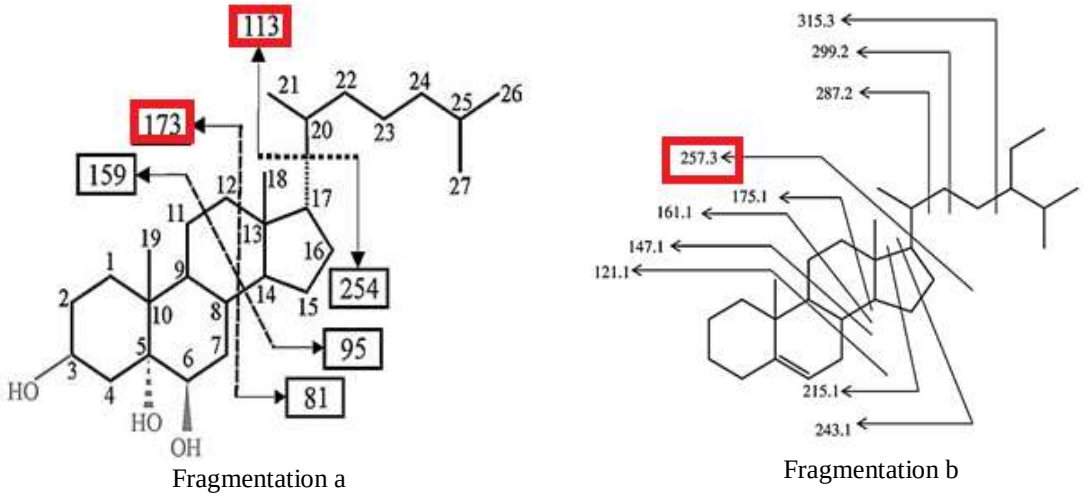
This antidiabetic activity of the recipe is due to the presence of its identified antidiabetic compounds (Scopoletin and Vanillin), supposed (Sitosterol and its derivatives) or by the synergy of these latter.

Indeed, in a streptozotocin (STZ)-induced diabetic rat model, scopoletin showed hypoglycemic and hypolipidemic effects (**Verma et al., 2013; Al-Zuaidyet al., 2016**).This molecule improves hyperglycemia and hepatic steatosis as well as postprandial glycemia by inhibiting the activity of carbohydrate digestive enzymes (α-glucosidase and α-amylase) in diabetic mice (**Choi et al., 2017; Jang et al., 2018).**

Vanillin, a powerful antioxidant acting on oxidative stress, contributes to the treatment of diabetes. Because, it significantly decreases serum glucose and triglyceride levels and increases insulin level in diabetic rats. In addition, vanillin improves liver and kidney functions, and attenuates histopathologicalchanges in the pancreas (**Lu et al., 2019**).

Sitosterol plays a key role in cholesterol regulation in diabetic patients (**Scheen and Radermecker, 2009**). In addition, several sterols identified and isolated from plants have shown hypoglycemic activity on diabetic rats; such is the case of charantin, a triterpene sterol isolated from the *Momordicacharantia*L. tested on the animal model and showed an antidiabetic effect (**Singh et al., 2012)**.

The presence of compounds with antidiabetic properties in this recipe could be a justification for its use for the treatment of diabetes and the results observed by **Koubakaet al. (2023**). This recipe would be beneficial in the management of certain disorders associated with diabetes



Fragmentation a Fragmentation b

Figure 6:-Fragmentation of triterpene sterols.

Conclusion:-

In order to shed light on the phytochemical composition of traditional medicines treating diabetes in the Republic of Congo, chemical analyses were carried out on one of the beverages called "NGOSI" purchased from congolese traditional practitioners. The results obtained show that the recipe contains polyphenols, sugars, flavonoid and triterpene glycosides. 48 compounds were observed in this recipe including 26 compounds identified by CG/MS, 10 suggested compounds identified by HPLC/MS and 12 non-identity compounds according to the databases used. Scopoletin and vanillin are proven compounds with antidiabetic properties and identified in this recipe. This work provides a phytochemical contribution to the scientific knowledge of the plant recipe and allows us to better understand the pharmacodynamicproperties of this traditional congolese medicine. It would therefore be very interesting to exploit the data and further deepen scientific studies on this medicine to validate the active ingredients responsible for its pharmacological properties.

Bibliographical References:-

1. Adjanohoun, E. J., et Technique, A. D. C. C., Ake Assi, L., Floret, J. J., Guinko, S., & Koumare, M. (1979). Medecine traditionnelle et pharmacopee; contribution aux etudes ethnobotaniques et floristiques au Mali.ACCT. Paris 219 p.
2. AL-Zuaidy, M. H., Hamid, A. A., Ismail, A., Mohamed, S., Abdul Razis, A. F., Mumtaz, M. W., & Salleh, S. Z. (2016). Potent antidiabetic activity and metabolite profiling of Melicope Lunu-ankenda leaves. Journal of Food Science, 81(5), C1080-C1090.https://dx.doi:10.1111/1750-3841.13293 .
3. Angalla Affleck, R. L., Lamini, N., Andzouana, N., Nkoualakidédé, C., AkoliEkoya, OmboumahouBakale, F., Salémo, A. P., Ntsiba, H., andBileckot. (2023). Rheumatologic findings in type 2 diabetic patients consulting at the University Teaching Hospital of Brazzaville. Health sciences and disease, 42-47. https://dx. doi:10.5281/hsd.v24i10.4833
4. Bonnefont-Rousselot, D., Bastard, J. P., Jaudon, M. C., and Delattre, J. (2000). Consequences of the diabetic status on the oxidant/antioxidant balance. Diabetes and metabolism, 26(3), 163-177.
5. Bouquet, A. (1969). Féticheurs et médecines traditionnelles du Congo (Brazzaville) (Vol. 36). Paris: Orstom. 282p.
6. Bouquet, A. (1972). Plantes médicinales du Congo-Brazzaville. Travaux et documents O.R.O.S.T.O.M. Brazzaville n°13, 112 p.
7. Bruneton, J. (1999). Pharmacognosie - Phytochimie, Plantes médicinales. Paris, Lavoisier Technique& Documentation. 1120 p.
8. Chattopadhyay, R. R. (1999). Possible mechanism of antihyperglycemic effect of Azadirachtaindica leaf extract: part V. Journal of Ethnopharmacology. 67: 373–376.https://doi.org/10.1016/S0378-8741(99)00094-X
9. Choi, R. Y., Ham, J. R., Lee, H. I., Cho, H. W., Choi, M. S., Park, S. K., Kim, M. J., Seo K. I. and Lee, M. K. (2017). Scopoletin supplementation ameliorates steatosis and inflammation in diabetic mice. Phytotherapy Research, 31(11), 1795-1804.https://dx.doi:10.1002/ptr.5925.

10. Demarne, F. (1996). Market gardening and aromatic plants. Aromatic plants. Consulted on September 19, 2024 http://agritrop.ci rad.fr/389650/
11. Domon, B., and Costello, C. E. (1988). A systematic nomenclature for carbohydrate fragmentations in FAB-MS/MS spectra of glycoconjugates. Glycoconjugate journal, 5, 397-409.https://dx. doi.org/10.1007/BF01049915
12. Hailat, I. A. (2014). Mass spectrometric methods for analysis of sterols and steryl esters in biological samples (Doctoral dissertation, Memorial University of Newfoundland).p 177.
13. Harborne, J. B. (1998). Phytochemical methods: a guide to modern techniques of plant analysis. Chapman and Hall.286 pages.
14. IDF Diabetes Atlas. (2021 10th edition scientific committee).Brussels: International Diabetes Federation. Accessed 17 March 2023. https:// www.diabetesatlas.org .
15. Jang, J. H., Park, J. E., & Han, J. S. (2018). Scopoletin inhibits α-glucosidase in vitro and alleviates postprandial hyperglycemia in mice with diabetes. European journal of pharmacology, 834, 152-156.https://dx.doi:10.1016/j.ejphar.2018.07.032
16. Kirby, G. C. (1996). Medicinal plants and the control of protozoal disease, with particular reference to malaria. Transactions of the Royal Society of Tropical Medicine and Hygiene, 90(6), 605-609. https://dx. doi:10.1016/s0035-9203(96)90404-6 .
17. Koubaka, J. F.Y., BonazabaMilandou, L. J.C., Babouongolo, S. G., NkounkouLoumpangou, C., Tony Malaissou D. E. (2023). Phytochemical profile and antiradical and antidiabetic activities of a plant recipe known to be antidiabetic in traditional Congolese medicine. Journal of Pharmacognosy and Phytochemistry, 12 (3), 37-42. https://dx.doi.org/10.22271/phyto .
18. Koussa, T., Dubos, B., Cherrad, M. (2002). Les teneursenacides gras, en eau et enacideabscissique des feuilles de vigne (Vitisvinifera L. var. Cabernet Sauvignon) infectées par Eutypalata. Vitis, 41(3), 143-146. https://doi.org/10.5073/vitis.2002.41.143-146
19. Kumar, S.R, Shilpashree, H. B., Nagegowda, D. A. (2018). Terpene moiety enhancement by overexpression of geranyl (geranyl) diphosphate synthase and geraniol synthase elevates monomeric and dimericmonoterpeneindole alkaloids in transgenic Catharanthusroseus. Frontiers in Plant Science, 9, 942. https://doi.org/10.3389/fpls.2018.00942
20. Leduc, C., Coonishish, J., Haddad, P., Currier, A. (2006). Plants used by Cree Nation of EeyouIstchee (Quebec, Canada) for treatment of diabetes: A novel approach in quantitative ethnobotany. JournalEhtnopharmacol, 105, 55-63. https://dx.DOI:10.1016/j.jep.2005.09.038
21. Li, R., Cen, B., Duan, W., Lin, G. (2022). Synthesis, Antifungal Activity and 3D-QSAR Study of Novel Anisaldehyde-Derived Amide-Thiourea Compounds. Chemistry & Biodiversity, 19(4), e202101025. https://doi.org/10.1002/cbdv.202101025
22. Lila, M. A., Raskin, I. (2005). Health-related interactions of phytochemicals. Journal of food science, 70(1), 20-27. https://doi.org/10.1111/j.1365-2621.2005.tb09054.x
23. Lu, G., Luo, X., Liu, Z., Yang, L., Lin, C., Xu, M. (2019). Protective effect of vanillin in streptozotocin-induced diabetes in neonatal rats via attenuation of oxidative stress and inflammation. Tropical journal of pharmaceutical research. 18(2), 349-355. https://doi:10.4314/tjpr.v18i2.18
24. Ma, R., Liu, H., Shi, F., Fu, Y., Wei, P., Liu, Z. (2020). The chemical composition and antioxidant activity of essential oils and extracts of Dalbergiaodorifera leaves. Holzforschung, 74(8), 755-763. https://doi.org/10.1515/hf-2019-0155
25. Merck, E. (1980). Dyeing reagents for thin layer and paper chromatography. E. Merck.Edition Darmstadt, 153p.
26. Münger, L. H., Boulos, S., Nyström, L. (2018). UPLC-MS/MS based identification of dietary sterylglucosides by investigation of corresponding free sterols. Frontiers in chemistry, 6, 342. https://doi.org/10.3389/fchem.2018.00342
27. Newman, D. J., Cragg, G. M., Snader, K. M. (2000). The influence of natural products upon drug discovery. Natural Product Reports 17, 175-285. https://doi.org/10.1039/A902202C
28. Olmo-García, L., Kessler, N., Neuweger, H., Wendt, K., Olmo-Peinado, J. M., Fernández-Gutiérrez, A., Baessmann, C., Carrasco-Pancorbo, A. (2018). Unravelling the distribution of secondary metabolites in Oleaeuropaea L.: exhaustive characterization of eight olive-tree derived matrices by complementary platforms (LC-ESI/APCI-MS and GC-APCI-MS). Molecules, 23(10), 2419. https://doi.org/10.3390/molecules23102419
29. Rollinger, J. M., Hornick, A., Langer, T., Stuppner, H., Prast, H. (2004). Acetylcholinesterase inhibitory activity of scopolin and scopoletin discovered by virtual screening of natural products. Journal of medicinal chemistry, 47(25), 6248-6254. https://doi.org/10.1021/jm049655r

30. Rossmann, B., Thurner, K., Luf, W. (2007). MS–MS fragmentation patterns of cholesterol oxidation products. MonatsheftefürChemie-Chemical Monthly, 138, 437-444. https://doi.org/10.1007/s00706-007-0589-2.
31. Scheen, A., Radermecker, R. (2009). Ezétimibe (Ezetrol®) chez le patient diabétique. Revue Medicale de Liege, 64(12), 606-611. https://hdl.handle.net/2268/29559
32. Singh, R., Kumar, A., Giri, D. D., Bhuvaneshwari, K., Pandey, K. D. (2012). Gas Chromatogrphy-Mass Spectrometry Analysis and Phytochemical Screening of Methanolic Fruit Extract of Momordicacharantia. Journal of Recent Advances in Agriculture,1(4), 122-127. https://doi.org/10.1021/jm049655r
33. Singh, J., Metrani, R., Shivanagoudra, S. R., Jayaprakasha, G. K., Patil, B. S. (2019). Review on bile acids: effects of the gut microbiome, interactions with dietary fiber, and alterations in the bioaccessibility of bioactive compounds. Journal of Agricultural and Food Chemistry. 67(33), 9124-9138. https://doi.org/10.1021/acs.jafc.8b07306
34. Verma, A., Dewangan, P., Kesharwani, D., Kela, S. P. (2013). Hypoglycemic and hypolipidemic activity of scopoletin (coumarin derivative) in streptozotocin induced diabetic rats. International Journal of Pharmaceutical Sciences Review and Research, 22 (1), 79–83.
35. Wagner H., Bladt, S. (1996). Plant drug analysis a thin layer chromatography Atlas. 2ème édition, Berlin: Springer-Verlag. 384p. http://dx.doi.org/10.1007/978-3-642-00574-9.