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GC-MS analysis of volatile compounds of frankincense from *Boswellia papyrifera* in Northern Ethiopia

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Abstract
A study was conducted to evaluate the effects of the two study sites on essential oil content (EOC) and essential oil composition (EOC_P) of the oleo-gum resins obtained from *Boswellia papyrifera*. Authentic representative oleogum resin samples were collected from *Abergelle and Metema* in the northern dry land aria of Ethiopia. The essential oils were obtained by hydro distillation using in a Clevenger-type apparatus for 3h and the average oil content 1.09% in *Metema* and 0.25% in *Abergelle* were recorded. The volatile oil of *Boswellia papyrifera* was examined by GC and GC-MS techniques and ten components (85.68%) comprising 100% of total volume in *Abergelle* and fifteen components (86.12%) comprising 100% of total volumes in *Metema* were identified. Acetic acid (58.56%, 51.23%, 1-octanol (8.87%, 17.66%) and Ethyl-3-en-1-yl (12.91%, 11.72%) were found to be the major components in the study species which were collected *Abergelle* and *Metema*. In this paper we found the results of our study on the volatile oils of olio-gum resins collected from the two sites were observed that the GC-MS spectrum of the oil have similar but the essential oil content have significant difference.

Keywords: Acetic acid, boswellia papyrifera, compositions, ethylpent-3-en-1-yl, Ole-gum resins, 1-octanol

Introduction
According to the International Standard Organization on Essential Oils [1] and the European Pharmacopoeia [2] an essential oil is defined as the product obtained from plant raw material by using different extraction processing methods on the plant materials [3]. One of the most common is steam and hydro distillation. Essential oils or ethereal oils are complex mixtures of odoriferous and volatile compounds [4]. They are the aroma giving constituents of aromatic plants and are not to be confused with other oils such as "fixed oils" which are chemically different [5, 6].

The genus Boswellia and Commiphora of the family (over 150 species of trees and shrubs) Burseraceae distributed mostly in East Africa, Arabia and India [7]. The resins of *Boswellia* spp. are commonly known as ‘frankincense’ or ‘olibanum’. The main frankincense-producing African Species are *B. papyrifera* (Del.) Hochst., found in Ethiopia and Sudan [8]. Frankincense Oil, Olibanum Oil or Oil of Lebanon is an ancient essential oil used for spiritual connection and to support a healthy immune system. It is also a valuable ingredient in skin care products for healthy looking skin. Ethiopia is one of the few countries with large frankincense and *Myrrh* resources. The country has a potential annual production of 70661 tons from the 2.9 million ha of land total area covered of oleo-gum resin bearing species [9]. *Boswellia papyrifera* provides the widely traded frankincense that accounts for >80% of the export revenues that the country is earning from gum and resin resources [10].

Essential oils are located in different parts of the plants (flowers, fruits, seeds, leaves, stems and roots), as well as essential oil can be produced from different aromatic and medicinal plants. The essential oil production is influenced by different factors such as the type of species/genotypes, fruit maturity, species growing site, leaves position, leaves type, leaves age and harvesting season [11].

The aim of this study was focuses on giving some pictures for the variability of oleo-gum essential oil quality under Ethiopia conditions by studying (evaluate) the effects of the study

sits on essential oils content and essential oil compositions from oleo-gum resins of *Boswellia papyrifera* and as well as coppering the oil qualities meet with the international standard characteristic.

Material and Methods

Study aria description

The study was conducted in two woodland areas dominated by *B. papyrifera* trees: *Abergelle* in the north and *Metema* in the north-west Ethiopia. Both woodlands are known for having a high frankincense production and a long history of marketing of the product [12]. *Abergelle* wereda; which is placed at 985 km Northwest of Addis Ababa. It lies within an altitudinal range of 950 to 1100 m above sea level. The mean annual rainfall is 885 mm, and the annual mean temperature is 27.8 ºC. It has a unimodal rainfall and most of the rainfall is received during the months of July and August. The soils are predominantly black, and some soils have vertic properties. Seasonal water logging, especially during the heavy rainfall months, is also high [12]. *Metema* is geographically located at 36' 17' E and 12" 39' N with an altitudinal range of 810 to 990 m. The specific study site is located about 205 km west of Gonder town. The annual rainfall ranges from 645 - 1132 mm with mean annual rainfall of about 955 mm. The annual mean maximum and minimum temperatures are about 36 and l9 ^{O}C, respectively. The woodlands sampled covered approximately 375 ha [13].

Plant collection and extraction of essential oils

The oleogum resin product of from *B. papyrifera* trees were collected in the period of dry session between January, 2014 and May, 2015 from Abergelle and Lemelem arias in the northern part of Ethiopia. The collected oleogum resin product samples were put into perforated plastic bags and ship safely transport to Central Ethiopia Environment and forest Research Center Laboratory (CEEFRCL). In the laboratory, the olio-gum samples were spread on aluminum try and dried in open air until the samples were dried uniformly. The dried oleo-gum resin product of *B. papyrifera* was crushed in to appropriate size. Essential oils from ground oleo-gum resins were obtained by hydro distillation for 3 h using a Clevenger-type. The obtained oils were allowed to dry over anhydrous sodium sulphate. After filtration, the oils were stored at +4 ◦C for until further GC-GC/MS test analysis will be done [14].

Identification of Essential Oil Constituents

GC-MS working conditions

Working conditions were split mode, injection (1 µL) of the resin oil was subjected to GC-MS and helium used as gas carrier at a flow rate of 1mL/min. The test was carried out at a temperature program 60-240^{0}C (3^{0} C/min), injector temperature 275 ^{0}C and detector temperature 280 ºC. The mass spectrometry conditioned was as follows: ionization voltage, 70 eV; emission current, 40 mA; mass range 0–600 Da, ion source temperature, 230 ºC.

Identification of Essential oil constituents

The identification of the essential oil components were carried out by gas chromatography mass spectrometry (GC-MS) using a GC-MS 7820A Agilent Technologies instrument with a mass selective HP 5977E detector fitted with HP-5MS 5% phenyl methyl silon capillary column (30 m x 0.25 mm i.d.*0.25µm, film thickness). The fragmentation patterns of mass spectra with those published in literature [15] and using NIST-14 (2014 model) libraries were compared with those stored in the mass spectral data base of the gas chromatograph computer and with those reported in the literature [16, 17, 18].

Results and Discussion

Essential oil content from *Bosolica papyrifera*

The volatile oil content of Bosolica papyrifera olio-gum resin collected in Abergelle and Metema was showed in Table 1. The volatile oil percentage was affected by the plant growing site since it was 0.25% to 1.09% in *Abergelle* and *Metema* locations, respectively. Not only volatile oil percentage but also oil percentage composition (constituent) was changed by growing *Bosolica papyrifera* tree in the two study sites. The result volatile oil content 1090mg or 1.09% from *Metema* study site is more of better the privies reported by [19]. The volatile oil content results obtained from *Abergelle* 250mg or 0.25% was similarly comparable the privies report by [20] 230mg or 0.23% essential oil was obtained oleo-gum resins *Boswellia papyrifera* grown SEM aria in Soudan.

Composition of essential oil of *Bosolica papyrifera*

The GC-MS *c*hromatogram spectrum (Figure 1and 2) of the two examined olio-gum resin which are revealed that the essential oil of *Bosolica papyrifera* contained a mixture of different hydrocarbons with oxygenated compounds that eluted at different retention times depending on the boiling point of the eluted components according to HP-5MS 5% phenyl methyl silon capillary column. Of which, ten components (85.68%) comprising 100% of total volume in *Abergelle* and fifteen components (86.12%) comprising 100% of total volumes in *Metema* were observed (Table 1) with the identical essential oil chromatogram spectrum (Figure 1and 2). The results shown in Table 1 for *B. papyrifera* olio-gum resin collected from *Abergelle* and *Metema* shows that the predominant concentration ranges for the major three components: n-Octyl acetate (51.2-58.56%), Ethylpent-3en-1yl (11.72-12.91%) and 1-Octanol (8.83-17.66%) observed as the main constituents.

Table 1: Essential oil content and essential oil constituent of *B. papyrifera* from the study sites

Compound Name identified by NIST14.L	*B. papyrifera*			
	Abergelle		*Metema*	
	RT	C (%)	RT	C (%)
1-Octanol	11.474	8.87	11.447	17.66
Acetic acid (Octyl ester)/ **n**-Octyl acetate	17.637	58.56	17.557	51.23
CEM	46.205	5.34	46.192	5.51
Ethylpent-3-en-1-yl	52.92	12.91	52.893	11.72
Major constituent of total concentration		85.68		86.12
Identification of components	10		15	
Essential Oil Content (%)	0.25		1.09	

RT: - retention time, C: - concentration, CEM: - Cyclohexene, 1-ethenyl-1-methyl-2, 4-bis (1-methylethenyl)-, [1S-(1.α., 2.β., 4.β)] [15].

Analyzed olio-gum resins of *Boswellia papyrifera* collected from wild trees growing in *Metema* and reported n-Octyl acetate (56%), 1-Octanol (8%) and Limonene (6.5%) were found to be the major components. These results are comparable to that reported by [21] on Olibanum from Somalia, where by nearly 15 components at found in common. Moreover, in this study also we have verified that qualitative results were found in the same way in chemical compounds and the results are discussed in Table 1. Furthermore, this studies is comparable to that reported by [18, 22] on some oleo-gum resin essential oils from *Boswellia spp*.

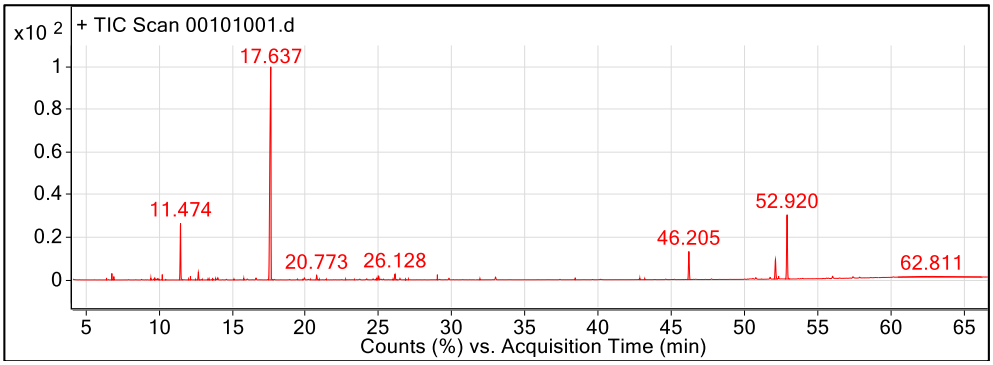

Fig 1: Essential oil chromatogram of *Bosolica papyrifera* detected by GC-MS (Abergelle site)

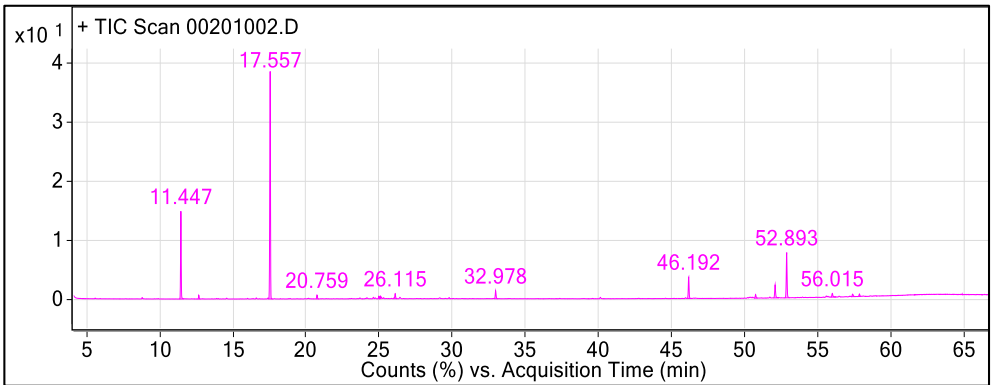

Fig 2: Essential oil chromatogram of *Bosolica papyrifera* detected by GC-MS (Metema site)

Conclusions

In this study, it has been clearly shown the impact of growing aria of wild tree on essential oil content but not significantly affected essential oil quality of *Boswellia papyrifera*. The least value of essential oil content, which is 22.94% lower the maximum was observed from *Abergelle* site. The two authentic *Boswellia papyrifera* olio-gum resins were rich in compounds namely, 1-Octanol, n-Octyl acetate and Ethylpent-3en-1yl. The sum of percentage compositions for the identification compounds in each olio-gum resin of *Bosolica papyrifera*, which were collected from *Abergelle* and *Metema*, were recorded that 80.34%, 80.61%, respectively. In this paper, we found the results of the study on the essential oil content and essential oil quality of the olio-gum resins collected from the two sites were observed that the GC-MS chromatogram of the oil have similar but the essential oil content have significant difference and also comparable with those reported previously in the literature.

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