Sudan University of Science and Technology

College of Graduate Studies

Screening of some Toxic and Narcotics Alkaloids of Morphologically Related Plants of the family Solanaceae

الكشف عن بعض القلويدات السامة والمخدرة في النباتات المتقاربة مورفولوجياً من العا ئلة البا ذنجا نية

A Thesis Submitted in Fulfillment of the Requirements of the Degree of Doctorate of Philosophy in Chemistry

By:

Saleh Maihoul Hadma Al-Mantefji

B.Sc Chemistry, University of Bagdad 1998

M.Sc. Forensic Evidence, The National Rebat University 2014

Supervisor

Prof. Mohammed El-Mubarak Osman

Co-Supervisor

Dr. Almoiz ELtayeb Mohammed Khalid

November 2020

Approval Page

(To be completed after the college council approval)

Name of Candidate: Saleh | maihoul | Hadma |

Thesis title: Screening of Some Toxic and Narcotics Alkaloids of Morphologically Related plants of the Family Solanaceae

Degree Examined for: .....

.....

Approved by:

1. External Examiner

Name: Ehab Elsir Mohamed Abbas

Signature: ..... Date: 22/8/2020

2. Internal Examiner

Name: Elfatih Ahmed Hassan

Signature: ..... Date: 22/8/2020

3. Supervisor

Name: Mohammed Elamin Osman

Signature: ..... Date: 22/8/2020

Dedication

To everyone who worked continuously among day and nights to prevent crime and protect the nation.

Acknowledgment

First, and foremost my Gratitude and thanks to Allah Almighty, for his facilitation to accomplish this research. I would like to thank my supervisor prof. ***MohammedEL-MubarakOsman*** and my co-supervisor Dr.***Almoiz Eltayeb Mohammed Khalid***for their support with their knowledge and open heart to my mistakes with great smile.

I should mention the great support I have received from my family; my Father, brothers and my wife. I would have done nothing without them.

My thanks to my colleagues in the Chemistry Department at the Regional Forensic Laboratory in the General Administration of Forensic Evidence - Sudan, The thanks before that go to my family staff of Forensic Laboratory- Ras Alkhaimah–United Arab Emirates.

Firstly and finally the best thanks to Allah.

Abstract:

Atropine, Hyoscine, Nicotine and Solanine are alkaloids produced by plants from the family Solanaceae as a secondary metabolite and which have an intense physiological action on animals even at low doses. The main source of Atropine, and Hyoscine alkaloids is the *Datura spp* while the main source of Nicotine are *Nicotiana spp*. The Solanine is widely found in potatoes and eggplant.

This study focuses on detecting the presence of Hyoscine, Atropine, Nicotine and Solanine in other plant species from the family *solanceae* other than their known sources, namely *Solanum dubium Dunal, Solanum incanum L., Datura stramonium L., Datura innoxia Mill, Hyoscyamus albus L, Lycium deserti Phil, and Nicotiana rustica L.*

Plant parts (stem. leaves. fruit and roots) were analyzed using TLC for screening the existence of targeted alkaloids. Results were compared with results of same analysis for authentic standards of targeted alkaloids. RF of standards samples of atropine, hyoscine, nicotine and solanine were 2, 5.3, 3.6, and 2.3 respectively, positive results of plants parts were analyzed using GCMS for confirming and quantity concentrations, calculations was performed according to the results of same analysis of authentic standards.

Concentrations of Atropine in *D. innoxia* were 0.03434, 0.042938and 0.02170 mg /g for stem, leaves and fruit respectively. Hyoscine concentration for the same plant were 0.05613, 0.05555 and 0.0125 mg/g for same plant parts, Nicotine was found only in leaves in concentration of 0.00741 mg/g. Solanine was not found in this plant and no one of targeted alkaloids was found in its roots.

For *D. stamonium* Atropine concentrations were 0.03078, 0.04988 and 0.2072 mg/g, Hyoscine concentration were 0.01214, 0.015455 and 0.05627 mg/g in stem, leaves and fruit respectively. Nicotine was found in stem and

leaves in concentrations of 0.01104 and 0.022363 mg/g respectively. Solanine was not found in this plant and no one of targeted alkaloids was found in its roots.

H. albus showed concentrations of 0.03034, 0.04295 and 0.2171 mg/g for Atropine and concentrations of 0.07414,0.07434 and 0.02375 mg/g for Hyoscine in stem, leaves and fruit respectively. Nicotine was found only in leaves in concentration of 0.022411mg/g Solanine was not found in this plant and no one of targeted alkaloids was found in its roots.

In *L. deserti phill* Nicotine was found in leaves in concentration of 0.02855 mg/g and solanine was in fruit in concentration of 0.01346 mg/g.

All plant parts of *N. rusica*stem, leaves, fruitand roots were containing Nicotine in concentrations of 0.01820, 0.04450, 0.01816 and 0.01752 mg/g. respectively solanine was found in fruit in concentration of 0.01807 mg/g.

Solanine was found in stem, fruitand roots in *S. dubium* in concentrations of 0.04521, 0.07839 and 0.01940 mg/g, it was not found in leaves. Nicotine was in stem and leaves in concentrations of 0.001271and 0.01890 respectively. No one of the other targeted alkaloids was found in this plant.

Concentrations of Solanine in *S. inncum* were 0.03321,0.01885 and 0.06105mg/g for stem, leaves and fruit respectively. Nicotine was present only in leaves in concentration of 0.01768 mg/g.

It was concluded that: the specimens of *D. stramonium, D. innoxia* and Hyosyamus albus which were analyzed can be utilized as a good row material for medical and chemical industries regarding to concentration of Atropine and Hyoscine and regarding to less concentrations of other alkaloids like Solanine and Nicotine. *Lycium deserti phill* specimens were clear of toxic addictive alkaloids; they were contained Solanine and Nicotine in non significant concentrations. *Nictiana rusica* specimens were very rich of Nicotin which (four times in leaves than *N. tubacum*) *Sulanum*

dubium and *Sulanum inncum*specimens contained significant concentrations of solanine they were free of toxic addictive alkaloids, *S. dubuim* spciments contained very close relative compound to the tropan alkaloids, This suggests from the point of view of the plant chemotaxonomy that it is very close to the *Datura spp.* and *Hyosyamus spp*. All plants specimens in this study is a good material for chemical, medical industry and other traditional uses.

مستخلص:

الأتروبين والهيوسين والنيكوتين والسولانين عبارة عن قلويدات تنتجها نباتات من العائلة الباذنجانيةSolanaceae باعتبارها مستقلب ثانوي و لها تأثير فسيولوجي مكثف على الحيوانات حتى عند تناول جرعات منخفضة. المصدر الرئيسي للأتروبين ، و الهيوسين هو *Datura spp* الداتورة ، والمصدر الرئيسي للنيكوتين هو *Nicotiana spp*.بينما يتوفر سولانين على نطاق واسع في البطاطس والباذنجان.

ركزت هذه الدراسة على اكتشاف وجود الأتروبين و الهيوسين و النيكوتين و السولانين في أنواع نباتية أخرى من العائلة solanceae بخلاف مصادرها المعروفة ، تحديدا النباتات: *Solanum dubium Dunal* و *Solanum incanum L*. و *Datura innoxia L*. و *Datura innoxia Mill*و *Hyoscyamus Albus L*، *Lycium deserti Phil* ، و *Nicotiana rustica L*.

تم فحص الأجزاء النباتية (الأوراق،الساق. الثمار والجذور) باستخدام TLC بشكل أولي للكشف عن وجود القلويدات المستهدفة. تمت مقارنة النتائج مع نتائج نفس التحليللعينات قياسية موثوقة . ثابتالاحتباسRF للعينات القياسية من الأتروبين والهيوسين والنيكوتين والسولانين كانت 2 و 5.3 و 3.6 و 2.3 على التوالي ، تم تحليل أجزاء النباتات ذات النتائج الايجابية باستخدام GCMS لتأكيد النتائج وتحديد التراكيزالتي تم حسابها وفقا لنتائج نفس التحليل للعينات القياسية.

كانت تركيزات الأتروبين في *D. innoxia* 0.03434 ، و 0.042938 ، و 0.02170 ملجم / غرام للساق ، والأوراق ، والثمار على التوالي. و تركيز الهيوسين لنفس النبات 0.05613 ، 0.05555 و 0.0125 ملجم / جم لأجزاء النبات نفسها ، تم العثور على النيكوتين فقط في الأوراق بتركيز 0.00741 ملجم / جم. لم يتم العثور على Solanine في هذا النبات ولم يتم العثور على أي من القلويدات المستهدفة في جذوره.

بالنسبة لتركيزات الأتروبين في *D. stamonium* كانت 0.03078 ، 0.04988 و 0.2072 ملجم / جم ، و كانت تركيزات الهيوسين 0.01214 ، 0.015455 و 0.05627 ملجم / جم في الساق والأوراق والثمار على التوالي. تم العثور على النيكوتين في الساق والأوراق بتركيزات 0.01104 و 0.022363 ملجم / جم على التوالي. لم يتم العثور على Solanine في هذا النبات ولم يتم العثور على أي من القلويدات المستهدفة في جذوره.

أظهر *H. albus* تركيزات 0.03034 ، 0.04295 و 0.2171 ملجم / جم للأتروبين وتركيزات 0.07414 ، 0.07434 و 0.02375 ملجم / جم للهيوسين في الساق والأوراق والثمار على التوالي. تم العثور على النيكوتين فقط في الأوراق بتركيز 0.022411 ملغم / جم. لم يتم العثور على سولانين في هذا النبات ولم يتم العثور على أي من القلويدات المستهدفة في جذوره.

في *L. deserti phill* وجد النيكوتين في الأوراق بتركيز 0.02855 ملغم / جم وكان السولانين في الثمار بتركيز 0.01346 ملغم / جم.

كانت جميع أجزاء النبات *N. rusica* الساق والأوراق والثمار والجذور تحتوي على النيكوتين بتركيزات 0.01820 و 0.04450 و 0.01816 و 0.01752 ملغم / جم على التوالي. تم العثور على Solanine في الثمار بتركيز 0.01807 ملغم / جم.

تم العثور على Solanine في الساق والثمار والجذور في *S. dubium* بتركيزات 0.04521 ، 0.07839 و 0.01940 ملغم / جم ، لم يتم العثور عليه في الأوراق. كان النيكوتين في الساق والاوراق بتركيزات 0.001271 و 0.01890 على التوالي. لا أحد من القلويدات المستهدفة الأخرى موجود في هذا النبات.

وكانت تركيزات Solanine في *S. inncum* 0.03321، 0.01885 و 0.06105 ملجم / غرام للساق والأوراق والفواكه على التوالي. كان النيكوتين فقط في الأوراق بتركيز 0.01768 ملجم / جم.

خلصت هذه الدراسة إلى أنه يمكن استخدام عينات *D. Stramonium* و *D. innoxia* و *Hyosyamus albus* التي تم تحليلها كمادة خام جيدة للصناعات الطبية والكيميائية وذلك بالنظر الى تراكيز الأتروبين والهيوسين قي هذه العينات وكذلك لاحتوائها على تراكيز قليلة القلويدات الأخرى مثل السولانين والنيكوتين. كانت عينات *Lycium deserti phill* خالية من القلويدات السامة و المخدرة، مع اختوائها على سولانين ونيكوتين بتركيزات منخفضة. *Nictiana rusica* احتوت على تركيز عال جدا للنيكوتين (الأوراق اربع اضعاف *N. tubacum*) ، احتوت عينات *Sulanum dubium* و *Sulanum inncum* على تركيزات كبيرة من سولانين كانت خالية من القلويدات السامة ة المخدرة ، اتضح من النتائج احتواء *S. dubuim* على مركب قريب جدًا من قلويدات tropan ، وهذا يشير من وجهة نظر علم التصنيف الكيميائي للنبات إلى أنه قريب جدًا من *Datura spp.* و *Hyosyamus spp*. جميع العينات النباتية في هذه الدراسة هي مادة خام جيدة للصناعات الكيماوية والطبية والاستخدامات التقليدية الأخرى.

Publication:

- Almoiz E. M. Khalid, Saleh M. H. AL-mantfeji , Mohammed E. Osman(2019) Detection of the content of some toxic and narcotic alkaloids in: *Lycium deserti Phil and Hyosyamus albus*. International Journal of Advances in Science, Engineering and Technology, 7(4)pp1-6
- Almoiz E. M. Khalid, Saleh M. H. AL-mantfeji , Mohammed E. Osman (2019) Evaluation of Nicotine constituent in Nicotiana restica. International Journal of Advances in Science, Engineering and Technology, 7(4)pp35-38

Contents

List of Tables

List of Figures

List of abbreviation

GCMS	Gas Chromatography with Mass Spectrometer
THC	Tetra Hydro Cannabinol
CNS	Central Nerves System
DNA	Dioxy ribonucleic Acid
TLC	Thin Layer Chromatography
RF	Retention Factor
UV	Ultra violet
HPLC	High Performance Liquid Chromatography
FID	Flame Ionization Detector
TMS	Try Methyl Silyl
RT	Retention Time

Chapter One

1. Introduction and Literature Review

Chapter One

1. Introduction and Literature review

1.1. Introduction:

Drugs are considered one of the most serious problems that humankind has faced throughout the ages. Perhaps nations with different beliefs and religions have not agreed to fight something like their consensus to fight drugs. The true Islamic religion clearly stated the prohibition of everything that would cause harm to the human being, individually and collectively. Allah Almighty said (He delights them with good things and forbids them from evil ... Verse).

Drugs are defined according to what is reported by the United Nations as natural or manufactured chemicals containing narcotic compound and that would get them used to addiction. These materials are classified by the United Nations laboratories in the form of three tables according to their severity and uses, these tables are binding for all member states at the international level, However, each country issuing its own schedules is included in its internal laws committed to that prevailing directives and customs, some of the materials included in the tables of the United Nations may be permissible to use in some countries, but these countries are obligated not to export these materials or allow their transit through their territory to any Destination. From the previous definition of drugs, they are classified in terms of source to:

- **Natural drugs:** the source is natural without any human intervention, which are plants, plant parts, or plant products that contain narcotic chemicals that are used directly. Examples for this group Cannabis types and opium.

- **Semi-synthetic drugs**: they are plants, plant parts, or plant products that contain chemicals that are dealt with by industrial methods to transform into narcotic substances or to increase their narcotic properties, such as morphine.

- **Manufactured or Synthetic drugs:** they are pure chemicals and do not have to be of plant origin known as the term precursor. They are treated chemically and synthetically to turn them into narcotic substances, for example, barbiturates.

Natural drugs are considered the most difficult in terms of their ability to control them because their plant source makes their access affordable and with low material costs. One of the problems that are difficult to combat this type of drug is that some of it can have more than one plant source, and some plants may even contain substances with a narcotic effect, but they are unknown or legally uncontrolled, and thus they are misused. The Narcotic chemicals that are naturally formed in plants differ in terms of chemical composition. The active substance may be ketones such as cathinone in the khat plant, or the active substance may be alcohols such as Tetra hydro cnnabinol THC in Cannabis (Anthony et *al* 2005). However, a large percentage of plant narcotics active ingredients are alkaloids, as all opiates, for example, and cocaine. Among the most famous plant families that produce alkaloids are the nightshade family, the alkaloids produced from some plants of this family may be narcotic and some are highly toxic. The focus in researches for this family should be increased.

The nightshade (Solanaseae) family is one of the largest botanical families, comprising about 98 genera containing about 2,700 species (Olmstead, Bohs. 2007). Most of the plants in this family contain a large number of alkaloids, which are mostly highly toxic chemicals and may have serious effects on humans or animals (Olmstead, *et al*. 1999). Despite the high

toxicity of the alkaloids, many of them have many medical and industrial uses. Examples of these alkaloids are: atropine, nicotine, hyoscine, solanine, capsaicin, tomatine, laxumine, flumen, chakonene, lipotin, lysamine and others (Szymon *et al*. 2016).

Hyoscine and atropine are two of the most common alkaloids materials, according to their high toxicity and narcotic effect on the nervous system in addition to their various medical uses. The primary source of these two types of alkaloids is the *Datura spp*. It is a plant that is prohibited for cultivation and circulation in many countries. Nicotine also is a toxic alkaloid, its main source is *Nicotiana spp*. solanine is widely found in potatoes and eggplant, which is highly toxic and also has a number of medical and industrial uses. This study focuses on the detection of the presence of hyoscine, atropine, nicotine and solanine alkaloids in other plant genera of the nightshade family other than their common sources.

1.2 Literature review

1.2.1 Family *Solanaceae*:

The Solanaceae, or nightshades, are an economically important family of flowering plants. The family ranges from annual and perennial herbs to vines, lianas, epiphytes, shrubs, and trees, and includes a number of important agricultural crops, medicinal plants, spices, weeds, and ornamentals. Many members of the family contain potent alkaloids, and some are highly toxic, but many cultures eat nightshades, in some cases as staple foods (Olmstead *et al* 1999). The Solanaceae consists of about 98 genera and some 2,700 species, with a great diversity of habitats, morphology and ecology (table 1.1). (Olmstead, Bohs 2007).

Datura innoxia Mill, Datura stramonium L., Lycium deserti Phil, Nicotiana glauca Graham and Nicotiana rustica L Hyoscyamus albus L, Solanum dubium Dunal, Solanum incanum L., are some plants of the family Solanaceae which are morphologically related. There is some known information about constituent of some alkaloids in those plants.

Table 1.1: list of different genera of *solanaceae* family with approximate No. of species

Genus	Approximate number of species
Solanum	1330
Lycianthes	200
Cestrum	150
Nolana	89
Physalis	85
Lycium	85
Nicotiana	76
Brunfelsia	45
Estimated number of species in the family	2700

1.2.1.1 Morphology:

The leaves vary greatly in shape but are usually simple, although sometime highly lobed. They are alternate and never have stipules. The inflorescence is generally cymose and axillary, but may be reduce to a single flower. The flower are bisexual, usually radialiy symmetric, and usually 5-merous. The calyx is united, at least at the base, and sometime becomes inflated in fruit. The corolla is also united but its shape varies from along and tubular to rotate or campanula ate. It is usually radially symmetric genera. There are5 (rarely 4-8) epipetalous stamens that alternate with the corolla lobes. The anthers are sometime touching but are never fused. The gynaecium consists of a single pistil, usually with 2 locules and numerous ovules .the fruit is usually a berry but quite frequently a dry capsule. (Rathi *et al* 2016).

The family has a worldwide distribution, being present on all continents except Antarctica. The greatest diversity in species is found in South America and Central America, and there is a great distribution in Australia

andAfrica, exist in a large number of different ecosystems from deserts to rainforests, generally, the plants of these family bloom in the tropical climate and moderate (Olmstead, Bohs 2007).

1.2.1.2 *Datura innoxia*

It is an annual shrubby plant. All parts of the plant emit a foul odor similar to rancid peanut butter when crushed or bruised, although most people find the fragrance of the flowers to be quite pleasant when they bloom at night (Figure 1.1) (Annapoorani, Grace 2013).It is widely distributed, not only in African countries but also in the Netherlands, thrives in diverse environments, temperate to tropical (Hceht *et al* 1996). Recent work in Rwanda has shown that strains of the species can also be grown successfully at lower temperatures and at high attitudes (Hecht *et al*. 1996). D. innoxia has been used to treat impotence, asthma, and diarrhea, as an analgesic, to control fever, to kill parasites, and as a drug for criminal purposes (parrotta 2001).

Figure 1.1: ***Datura innoxia***

1.2.1.3 Datura stramonium L:

Datura stramonim (Figer 1.2), known by the common names jimson weed or datura is believed to have originate in Americas, but is now found around the world. All part of datura plants contain dangerous levels of the tropane alkaloids atropine, hyoscyamine and scopolamine which are classified as deliriants , or anticholinergic (Preissel *et al* 2002) . Datura has long been used as an extremely effective treatment for asthma symptoms. Other medicinal uses for Datura included stimulating abortions, providing relief from sore throat or toothache, and getting rid of parasites (pennachio *et al*. 2010).

Figure 1.2: ***Datura stramonium***

1.2.1.4 *Hyouscyamus albus L*:

Is a small genus flowering plants in the solanaceae family, it comprises 12 species, all of which are toxic. Hyoscyamus albus one of species of this family which is known (white henbanes) (Figure 1.3) and Hyoscyamus

niger (black henbanes) (united states Department of Agriculture 2010). This genus can be easily identified in terms of phenotype and chemical composition and its effective contents (Liberman, Mitchell, 2008).

The Mediterranean basin is the main origin of this species of medicinal plants, because the coastal stripes are overlooking southern Europe , and the Arab Maghreb, heavy wilderness species , however, proven and quality in deserts of Egypt and Libya in Africa , and west Punjab in Asia, Spain and Greece in Europe ,the main producing and exporting countries of henbane plant India , Afghanistan , Pakistan ,and Egypt .

Throughout history, it has been used alongside other plants as an anesthetic, therefore, for their psychologically influential characteristics such as magic drinks. These include visual hallucinations, sense of conflict, fight, and quarrel. There have many type of henbane, which may vary in their containment of the type of alkaloids and their quantity e g Egyptians henbane carries Hyoscine and Hyoscyanine, while black henbane and reticulates henbane, they produce both the former alkaloids next to Atropine. The results of the chemical analysis show that Egyptian henbane contains the highest amount of alkaloids except Atropine compared to the black henbane and reticulates henbane (Raetsch 2005). Hyoscyamus albus extracts were used in traditional medicine as an antiasthmatic and antispasmodic. It was also used as hallucinogenic and
Sedative alone or mixed with Cannabis and Datura (Ali Esmail 2018)

Figure 1.3: ***Hyusyamus albus***

1.2.1.5 *Lycium albus:*

Lycium is a species of the family Solanaceae, it is a flowering plant distributed in most continents concentrated in temperate and subtropical regions. The largest number of species is found in South America, followed by North America and South Africa. A number of species are observed throughout Europe and Asia, one of which is the origin of this plant in Australia (Fukuda 2001).

There are about 70 to 80 species. The most important is Lycium albus(figure 1.8) , *Lycium barbarum* and *Lycium chinens* which is the fruit of an important traditional food crop in China and has recently become a popular health food all over the world (Levin, Mille 2005).

Lycium has been known for European since ancient times. Species from the Far East to Europe were traded by the Romans, for example via Ariaka and Barbarikon Harbor near Karachi today, This type of plants recommended as a treatment for inflammation of the eyes and skin infections, in his book published in 1753, describes Lenaeus three species of Lycium plants: Lycium: *L. afrum, L. barbarum and L. europaeum.* Historically Lycium has long been known, especially *L. barbarum* which has been used in traditional Chinese medicine to treat certain cases such as

infertility in males. The leaves and roots of other types of *lycium*, have been used after mixing them with water in popular medicines to treat skin rash and to promote hair growth (Luo 2006).

Figure 1.4: ***Lycium deserti phil L***

1.2.1.6 *Nicotiana rustica:*

It is a rainforest plant in the Solanaceae family. It is a very potent variety of tobacco. The high concentration of nicotine in its leaves makes it useful for producing pesticides. It is often used for entheogenic purposes by South American shamans. It contains up to nine times more nicotine than common species of Nicotiana such as *Nicotiana tabacum* (common tobacco). It is smoked in cigars or used as an enema (Stanfill *et al* 2015).

N. rustica is a hardy plant (Figure 1.5) and can be grown in most of Russia (as opposed to *N. virginiana* which requires a warm climate), it was more readily and cheaply available, and did not depend on transport in a country with an underdeveloped road network and climatic portage problems. *N.*

rustica leaves have a nicotine content as high as 9%, whereas *N. tabacum* leaves contain about 1 to 3%.(Ley, Willy 1965).

Figure 1.5 ***Nicotiana restica***

1.2.1.7 *Solanum dubium Dunal , Solanum incanum L.:*

Solanum is a large and diverse genus of flowering plants, which include three food crops of high economic importance, the potato, the tomato and the eggplant. It also contains the nightshades and horse nettles, as well as numerous plants cultivated for their ornamental flowers and fruit. Solanum species show a wide range of growing habits, such as annual and perennials, vines, subshrubs, shrubs, and small trees. Many formerly independent genera like Lycopersicon (the tomatoes) and Cyphomandra are now included in Solanum as subgenera or sections. Thus, the genus today contains roughly 1,500–2,000 species.(Quattrocchi, U. 2000).

Solanum dubium Dunal (Figure 1.6) is a toxic weed plant that has milk-clotting ability. The fruits of S. dubium in doses of 2.5 to 10 g per kg per day killed goats in 2 to 5 days. Similar doses of the leaves caused deaths in

8 to 36 days. In sheep, both fruits and leaves required a longer period of dosing to cause death (Barri *et al* 1983).

Solanum incanum L. (Figure 1.7) known also as Sodom apple, bitter apple poison apple snake apple thorn apple. it is thought to originate in Africa. It is also found from the Middle East to India. This plant sometimes used as a "hedge of thorns". is toxic to livestock and considered to be a major threat to grazing. It is also found in savanna grasslands where it might impact upon native herbivores. It is one of the most abundant weeds in East Africa where it displaces native vegetation. It has become invasive in some protected areas to the detriment of native vegetation and wild animal grazing.

S. incanum can be termed a "bush encroacher", a native species that has become invasive. Such plants are likely to exist in a stable balance under natural conditions. However, under human-induced changes such as overgrazing, such species can increase in density to the detriment of other vegetation (Fukuhara and Kubo 1991).

Figure 1.6: ***Solanum dubium***

Figure 1.7: ***Solanum incanum***

1.2.2 Alkaloids definition:

The name derives from the word alkaline, it was used to describe any nitrogen containing base. Alkaloids are a group of naturally occurring compounds that mostly contain basic nitrogen atoms. This group also includes some related compounds with neutral and even weakly acidic properties. Alkaloids are produced by a large variety of organisms including bacteria, fungi, plants, and animals (McNaught and Wilkinson 1997).

The name alkaloids was introduced in 1819 by the German chemist Carl Friedrich and Wilhelm Meissner, it was derived from Late Latin root: alkali (which, in turn, comes from the Arabic al qualja "ashes of plant "). However, the term came into wide use only after the publication of a review article by Oscar Jacobsen in the chemical dictionary of Albert Ladenburg in the 1880s (Raymond *et al* 2010) .

Historically, Alkaloid – containing plants were used by humans since ancient times for therapeutic and recreational purposes. For example, medicinal plants have been known in Mesopotamia at least around 2000 BC (Mishiba *et al* 2000). A Chinese book on houseplants written in first and second centuries before Christ, reported a medical uses of Ephedrine and Opium poppies. Also, coca leaves were used by South American Indians since ancient time (Perez *et al* 2006).

A significant contribution to the chemistry of alkaloids in the early years of its development was made by the French researchers Pierre Joseph Pelletier and Joseph Bienaime Caventou who discovered quinine (1820) and strychnine (1818) , several other alkaloids were discovered around that time , including xanthine (1817) , atropine (1819) , caffeine (1820) and cocaine (1860) (Garcia *et al* 2003).

The first complete synthesis of an alkaloid was achieved in 1886 by German chemist Albert ladenburg, He synthesized Coniine. The development of chemistry of alkaloids was accelerated by the emergence of spectroscopic and chromatographic methods in 20th century and by 2008 more than 12000 alkaloids were identified (Sazima *et al* 2003).

Many alkaloids can be purified from crude extracts by acid-base extraction. Many alkaloids are toxic to other organism. They often have pharmacological effects and are used as medications as recreational drug, or in entheogenic rituals (Tarek Ismail 2016)

Although alkaloids act on a diversity of metabolic systems in humans and animals and have a wide range of pharmacological activities, they almost uniformly evoke a bitter taste (Manske 1965).

1.2.3 Classifications of alkaloids:

As Compared to most other categories of natural compounds, alkaloids have a large structural diversity and no uniform classification.

Historically, first classification methods combined alkaloids by the common natural source e. g: certain type of plants. This type of classification was justified by the lack of knowledge about the chemical structure of alkaloids, and is now considered obsolete (Levin *et al*. 2005). More recent classifications are based on similarity of carbon skeleton e .g indole , isoquinoline , pyridine –like , in some cases , nicotine contains a pyridine fragment from nicotinamide and also pyrrolidine part from ornithine and therefore can be assigned to both classes (smith *et al* 2006).

1.2.4 The major groups of alkaloids:

Generally, alkaloids are often divided into the following major groups:

1. **True alkaloids:** contain nitrogen in the heterocyclic ring and originate from amino acid .Their characteristic examples are atropine, nicotine and morphine. This group also includes some alkaloids which beside nitrogen

heterocycle contain terpene e. g evonine or peptide fragments e.g. ergotamine. This group also includes piperidine alkaloids, coniine and coniceine; although piperidine alkaloids, coniine and coniceine they do not originate from amino acids they were also included in this group.

2. **Proto alkaloids:** contain nitrogen in the hetrocyclic ring and also originate from amino acid. Example include: Mescalline, adrenaline, and ephedrine.

3**. Polyamine alkaloids:** Derivative of putrescine, spermidine and spermine.

4. **Peptide and cyclopeptide alkaloids:** Cyclopeptide alkaloids are polyamidic bases,widely distributed among plants of many families like *Rhamnaceae, asteraceae, Celastraceae, uphorbiaceae, Menispermaceae, Pandaceae, Rubiaceae, Sterculiaceae, and Urticaceae* (Gournelis *et al*. 1997).

5**. Pseudo alkaloids (Alkaloid-like):** this group does not originate from amino acid. This group includes: terpene-like, steroids-like, as well as purine-like alkaloids such as caffeine, theobromine and theophylline. Some authors classify some compound like ephedrine and cathinone as pesudoalkaloids (whitson *et al* 2005).Table2.1 show the groups of alkaloids and categories of these groups and some example.

Most alkaloids contain oxygen; those compounds are usually colorless crystals at ambient conditions. Oxygen-free alkaloids, such as nicotine or coniine, are typically volatile, colorless, oily liquids. Some alkaloids are colored, like berberine (yellow) and sanguinarine (orange). Most alkaloids were bases, but some are amphoteric, for example theobromine and theophylline.

Most alkaloids are poorly soluble in water, but readily dissolve in organic solvents, such as diethyl ether, chloroform and 1,2-dichloroethane. However, caffeine dissolves well in boiling water (Spiller 1997).

Table 1.2: Some categories and example of alkaloids in the five groups.

Group	Categories	Example
True alkaloids	Pyrrolidine	Hygrine
	Tropane	Atropine
	Pyridine	Nicotine
Proto alkaloids	phenyl Thelamine	Thiamine
	Muscarins	Muscarins
	Benzyl amine	Capsin
Poly amine	Potracine	Bawesin
	Espermidin	Condocaprine
	Espermine	Valanadrine
Piptide alkaloid	13-cycle peptide alkaloids	Nomalarin
	14-cycle piptide alkaloids	Amphenin
	15-cyclo piptide alkaloids	Mocronine
Pseud alkaloids	Diterepine	Acotone
	Steroids	Solanine

1.2.5 Some types of alkaloids:

Atropine, Nicotine, Solanine and Hyoscine are Alkaloids produced by plants from the family Solanaceae as a secondary metabolite and which have an intense physiological action on animals even at low doses (Robbers *et al*. 1996).

Atropine, nicotine and Hyoscine are classified as "True alkaloids", which contain nitrogen in the heterocycle and originate from amino acids. While Solanine is a glyco-alkaloid (Plemenkov 2001).

1.2.5.1 Atropine:

Atropine is a tropane alkaloid, which is an isomer of hyoscyamine, the source of atropine is some members of the solanaceae family (Burst John 2004), it is found in different concentration in *Datura stramonium*,

Hyoscyamus spp., *Brugmansia suaveolens*, etc. Atropine has two major actions: it affects the central nervous system CNS. On other hand this alkaloid depresses the smooth the muscle and the secretory glands which are innervate by the parasympathetic nervous (K asture *et al* 2008).

Atropine used as a medication to treat certain types of nerve agent and also, some types of slow heart rate, and to decrease saliva production during surgery. It is also used as pesticide poisons (Richard *et al*. 2014).

Historically atropine was used as cosmetic, it was used by the Greeks as an eye-drop to make the pupils expand and look seductive. Atropine eye drops are still used today, in small doses, to avoid eye problems which can lead to blindness (Thorpe 2015).

Atropine Chemical and physical properties:

The molecular formula is $C_{17}H_{23}NO$ and molar mass 289g/mol (Figure 1.8) , atropine is soluble in : 1g in 400 ml of water, 1g in 50ml of boiling water, 1g in 2ml of ethanol, 1g in 1ml of chloroform, 1g in 25ml of ether and 1g in 27ml of glycerol.it is not soluble in benzene and dilute acid (Anthony *et al* 2005).

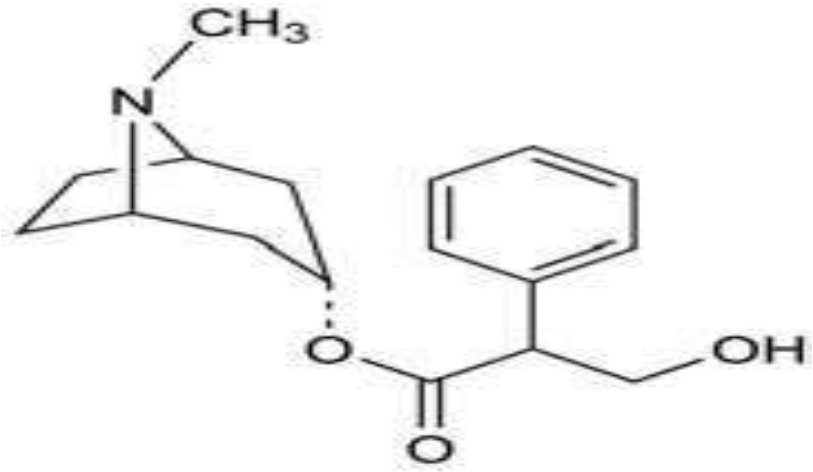

Figure 1.8: Atropine chemical structure

1.2.5.2 Hyoscine

Hyoscine is torpane alkaloid found in variety of solanaceous plants, it is also known as scopalamine, hyoscine used as a medication to treat motion sickness and postoperative nausea and vomiting. It is also sometimes used before surgery to decrease saliva (Gennaro *et al*. 1995). It found in *Hyoscyamus niger*, *Brugmansia suaveolens*, *Datura stramonium*. Hyoscine is a hallucinogenic substance if is administrated in high doses, which can even lead to coma or body death (sweta, laksimi, 2015).

As a group of these alkaloids exert two quite separate effects on mammals exposed to them – an anticholinergic effect, and a central nervous system depressant effect. This combination of properties, their potent effects with very small dosage and their widespread distribution have made them famous in the history of all recorded cultures (Pearn, Thearle 1982).

Until the early 19th century, hyoscine was not separated as a separate substance, but occurred in extracts and tinctures containing a cocktail of the plant-derived cholinergic blocking drugs (Pearn, Thearle 1982).

Hyoscine Chemical and physical Properties:

Hyoscine the molecular formula is $C_{17}H_{21}NO_4$ and molar mass 303.4 g / mol (Figure 1.9), crystalline monohydrate, Soluble 1 in 9.5 of water at 15, freely soluble in hot water, freely soluble in ethanol, chloroform, acetone and ether, sparingly soluble in benzene and Petroleum ether (Anthony *et al* 2005).

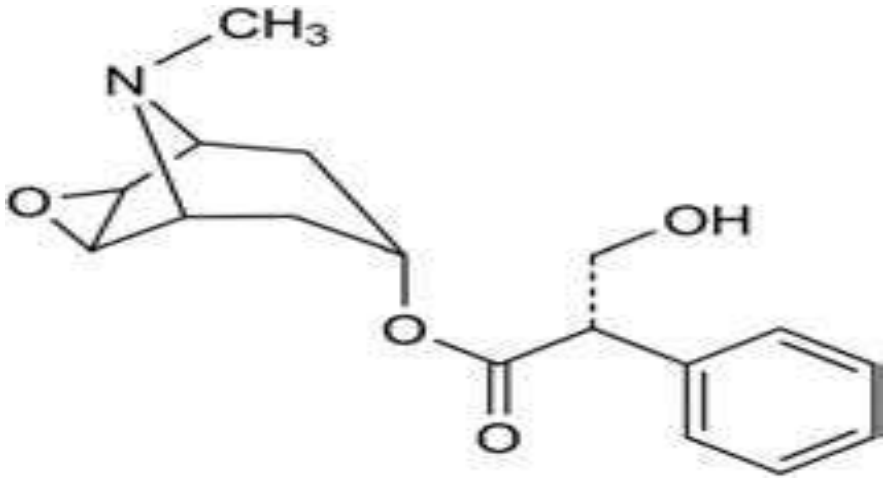

Figure 1.9: Hyoscine chemical structure

1.2.5.3 Nicotine:

Nicotine is pyridine alkaloid found in the nightshade family of (solanaceae), and natural in all parts of the tobacco plant, with a higher concentration in leaves, which constitutes 0.3 to 5% of plant dry weight with biosynthesis taking place in the roots and accumulating in the leaves. Nicotine found in tobacco, tomatoes, potatoes, green papers, eggplants and other, it is also found in coca leaves. Plants, especially tobacco, used nicotine to defend themselves against insect (ujvary istvan 1999).

Nicotine has a lot of therapeutic uses. There's growing evidence that it may be useful in treating Parkinson's disease, Alzheimer's , nicotine was widely used as an insecticide in the past(Rodgman 2009).

Nicotine chemical and physical Properties:

Nicotine the molecular formula $C_{10}H_{14}N_2$ and molar mass 162 g/mol (figure1.10) .It is colorless to pale yellow, very hygroscopic, oily liquid with an unpleasant pungent odor it gradually becomes brown on exposure to air or light. Nicotine is very soluble in ethanol, chloroform, petroleum ether, kerosene, oils and ether. (Anthony et al 2005).

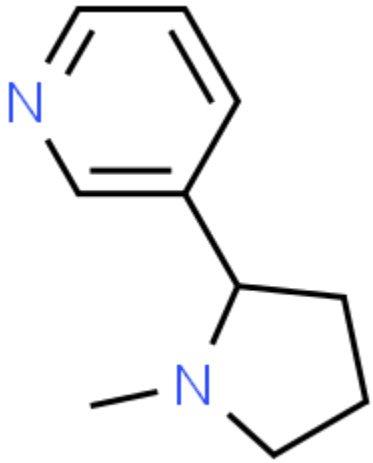

Figure 1.10: Nicotine chemical structure

1.2.5.4 Solanine:

Solanine is a glycoalkaloid with a bitter taste, it is found in species of the nightshade family within the genus solanum, such as potato, tomatoes, and eggplants. It can occur naturally in any part of the plant, including the leaves, fruits, and tubers. Plants is using Nicotine in order to resist fungi and insect that may intrude on them .and to defend himself from animals which may ate it (Desfosses 1820).

Historically, solanine was used in the treatment of epilepsy and asthma, in controlled doses. This practice is no longer common, as there are safer and more effective ways to treat these conditions. Solanine also has fungicidal and pesticidal qualities, but because of extraction and processing of this toxin is so time consuming the substance is rarely used for these purposes (Aditya .Sakare2012). Data provide evidence that solanine exerts a significant chemoprotective and chemotherapeutic effects on an animal model of breast cancer through apoptosis induction, cell proliferation and angiogenesis inhibition. These findings reveal a new therapeutic potential for solanine in cancer (Friedman , Mendel 2006).

Solanine Chemical and physical properties:

The molecular formula of Solanine is $C_{45}H_{73}NO_{15}$ and molar mass 868 g/mol (Figure 1.11). It is colorless to pale yellow, very hygroscopic, oily liquid with an unpleasant pungent odour. It gradually becomes brown on exposure to air or light. Insoluble in ethanol, chloroform, petroleum ether, kerosene, oils and ether.Soluble in hot ethanol, diluted acids and emylealchol. (Anthony *et al* 2005).

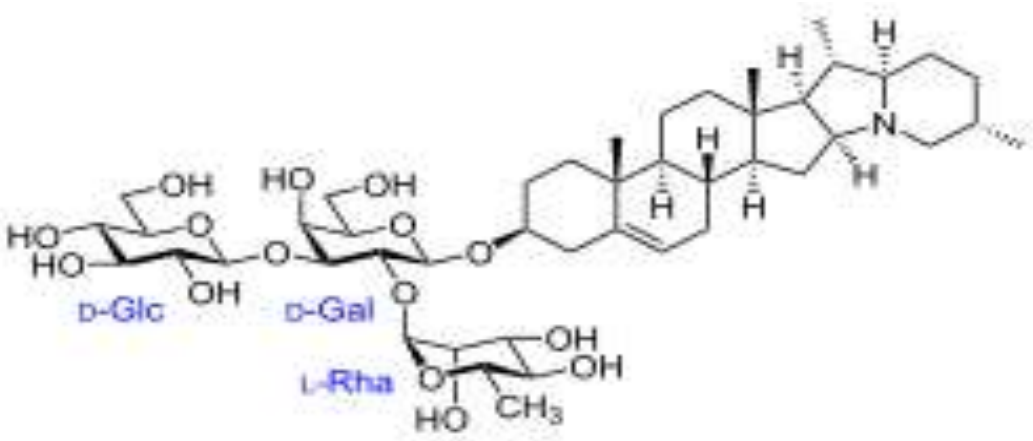

Figure 1.11: solanine chemical structure

1.2.6 Production:

- Solanine as glycoalkaloid is produced commercially by extracting the major alkaloids with water, and then preparing a crude glycoalkaloid extract from the weakly acidic plant extract (Errol 1998).

- An efficient and commercially feasible synthetic process for the preparation of atropine and atropine salts. In one aspect was developed by Frank *et al*. 2014, a one pot process for the synthesis of atropine is provided. The process provides excellent yield.

- Leaf contains from from 1 – 3%hyoscine along with other related compounds along with other related compounds • Harvested, stripped and dried. Then unique extraction process yields the hyoscine base and

followed by Synthesis and purification of the hydro and butyl bromides (Parry 2017).

For nicotine production Tobacco dust/ Tobacco Rava and lime are mixed together (using 10% lime) with the help of the mixing machine and packed into barrels specially made for percolation purpose. Water is added into the barrel from the top and the water is allowed to percolate through the dust. The water dissolves all the Nicotine present in the dust and it gets collected into a sump (Broth). This Nicotinised water (Broth) is then subjected to liquid - liquid extraction using kerosene as solvent. Nicotine being more soluble in kerosene is taken up by kerosene & the Nicotinised kerosene is then transferred into reactors for acidification with calculated amount of dilute sulfuric acid thereby forming Nicotine Sulfate – 40%. The Nicotine Sulfate- 40% is further purified by passing it through super centrifuge & then packed. (Arie *et al* 2013).

1.2.7 Hazards:

Solanine, atropine, hyoscine and nicotine have an intense physiological action on animals even at low doses, to humans; these alkaloids can be desirable, toxic, or both (Olmstead 2008).

Glyco-alkaloids including solanine were reported to be extracted in milk (Hayes 2008). It is also reported to addictive material (Michelle 2017). While atropine and hyoscine crosses the placental barrier and traces can be found in various secretions, of which breast milk (Gibson 2016).

Nicotine poses several health hazards. There is an increased risk of cardiovascular, respiratory, gastrointestinal disorders. There is decreased immune response and it also poses ill impacts on the reproductive health. It affects the cell proliferation, oxidative stress, apoptosis, and DNA mutation by various mechanisms which lead to cancer. It also affects the tumor

proliferation and metastasis and causes resistance to chemo and radio therapeutic agents. The use of nicotine needs regulation. The sale of nicotine should be under supervision of trained medical personnel (Mishra*et al*. 2015). Philippa 2017 reported that nicotine is readily absorbed into breast milk from the mother's blood, However another study found ten times as much cotinine (a metabolite of nicotine with a much longer half life that can be measured in blood, urine, saliva, hair and nails) in breastfed babies of smoking mothers (Bajanowski *et al*. 2007).

1.2.8 Alkaloids detection:

1.2.8.1 Thin Layer Chromatography TLC analysis:

Thin Layer ChromatographyTLC is a method of detection for alkaloids, which can be considered selective or a non-selective method, in the sense that it acts as a confirmatory test, when it is used as a preliminary test to the presence or absents of alkaloids. The detection results can be detected by determines the Rate of Flow RF using UV or by evaporating test plats in iodine. The RF also can be determined in case TLC is used as an initial test by using Chemical reagents such as sulfuric acid and Dragon drof reagent (Verpoorte1984).

1.2.8.2 High Performance Liquid Chromatography HPLC analysis:

This technique is widely used in the detection of alkaloids. This chromatographic technique uses a range of detectors. The commonly used in the detection of alkaloids is the use of UV detector. There are types of detectors which can be used with HPLC, including Photo diode Aray, including multi-refractive index. The HPLC technique has been used in a number of experiments and research to detect alkaloids, such as detection of estimonin, tyoprostimonin, neutioprostimonin, sehydrostimonin, tioprostimonin d, and tyoprostimonin K. After extraction from their plant

sources, HPLC analysis requires complex preparation stages and extractions. The results are collecting in a form of chromatograms (Huan He*et al* 2016).

1.2.8.3 Gas Chromatography analysis GC:

Since its introduction in the late 1950s, gas chromatography (GC) has evolved into a versatile tool in the analysis of natural products, including alkaloids. The obvious advantages of GC are high precision and high sensitivity. The most common detector is the Flame Ionization Detector (FID) for its accuracy and ease of use. This is in contrast to the analysis of high performance liquid chromatography (HPLC), where the detector's response may vary in the most common detector mode of this type of technology, namely ultraviolet (UV) for various compounds. The detection results using GCFID are extracted in the form of chromatograms (Dginino, *et al* 1994).

1.2.8.4 Mass spectrometer MS analysis:

It is an analytical method for determining the mass of molecules by their ionic signal. This technique depends on sending a beam of ions on the molecules to be analyzed. When this beam collides with the molecules, it breaks down into a number of fragments that differ in its molecular weight. It has been observed from repeated experiments that each fragment has a distinct mass that has the greatest fracture known as the main fragments. It also found that a part of the mass of the molecule remains intact without breaking; the mass of this part represents the molecular weight of the compound. In terms of the main fragment and the fragment of the largest weight can identify the compound being tested. MS technology can be used individually or as an accessory with GC or with HPLC. This technique has great utility and ability to detect alkaloids (Anthonye *et al*. 2005).

1.3 Objectives:

The plant family solanaceae is the main source of atropine, hyoscine, nicotine and solanine, the objectives of this study to are to detect the constituent of the mentioned alkaloids in**:** *Solanum dubium Dunal, Solanum incanum L., Datura stramonium L., Datura innoxia Mill, Hyoscyamus albus L, Lycium deserti Phil, and Nicotiana rustica L.*

Specific objectives:

- Determination of alkaloids in each plant parts of the mentioned alkaloids in each targeted plants.
- Determination of concentration of the alkaloids in targeted plants parts.

Chapter Two

2. Material and Methods

Chapter Two

2. Material and method

2.1 Materials:

A. Samples of *Dtura stramonium* and *Datura inoxia* colleted from east wed Madani, Alazeara state, Sudan.

B. *Hyoscyamus albus* samples collected from Dungula, Northern state, Sudan.

C. *Lycium deserti pil* L samples collected from Arkaweet, Red see state, Sudan.

D. *Nicotiana restica* samples collected from Halfa, Northern state, Sudan.

E. *Sulanum dubium* and *Solanum inicunam* collected from Abudlaig, eastern Khartoum state, Sudan.

F. Standard samples of atropine, hyoscine, nicotine and solanine purchased from sigma - Aldrich.

G. Trimethylsilyl TMS purchased from sigma-Aldrich catalog No. 1391

2.2Tools and equipment:

A. Gas chromatograph with mass spectrometer (GC MS)Shimatzu 4080.

B. Sensitive balance Shimatzu

C. Test tube (different sizes made from pyrx)

D. - Micro pipette.

E. - Thin layer plates analysis (silica gel 0.25 mm).

F. -Beaker (different size made from pyrx).

G. – Capillary tubes glass.

H. –TLC Tank glass with cover.

I. - Laboratory mortar & pestle.

J. - Filter paper (Watman NO.1).

2.3 Samples preparation:

Plant sample were divided into plant part (roots, stems, leaves and fruits) then dried at room temperature and grind into powder form.

2.3.1 Preparation of standards samples:

Atropine standard was prepared by adding 1 ml of methanol (HPLC grade) to 1 g of atropine.

Hyoscine standard and nicotine standard were prepared by adding 1 ml of methanol to 1 ml from hyoscine and 1 ml of nicotine , while solanine standard was prepared by adding 1 ml of (methanol: acetic acid 9:1) to 1 g of solanine.

2.4 Extraction:

Extraction of atropine hyoscine and nicotine: 1000 mg of herbal powder of each sample of plant part (Roots, Stems, Leaves, and Fruits) of targeted plant were weighed; 1ml of chloroform was added to the powder of each sample and left for 24 hour at room temperature. The suspensions were filtered (Anthonye *et al* 2005).

Extraction of solanine: 1000 mg of herbal powder of each sample of plants was weighed; 1 ml of diluted 2%acetic acid freshly prepared was added to the powder of each sample and left for 24 hour at room temperature. The suspensions was filtered (Anthony *et al* 2005).

2.5 Analysis

2.5.1 Thin layer analysis (TLC):

Samples of extract of each plant part were loaded in TLC plate. A control sample of atropine, hyoscine, nicotine and solanine were loaded in the plate beside the plant samples, each plate used for detection of one type of target alkaloids. The plates of TLC analysis were but in TC system (mobile phase chloroform 90% methanol %10 in saturated condition) the mobile phase

run for 10 cm. The RF was observed under ultraviolet radiation at a wavelength of 254nm (Anthonye *et al* 2005).

2.5.2 Gas chromatography analysis with mass spectrometer GC MS:

Positive results samples were evaporated under Nitrogen cover, the dried residues dissolved in solution of (methanol: acetic acid 9:1). Solanine samples were drevatized using TMS (TEK 2006). Samples of each plant parts of targeted plants were pooled together then dried using anhydrous sodium sulphate.

The samples were injected in GCMS chromatograph (Shimatzu 8040). The Injection volume was 1 ul, the start temperature of the column was 80^0 C hold for 1 minute, then increase to 200 c at a rate of 15dgree per minute and hold for 1 minute. Then increased to 260^{o}C at a rate of 10 degree per minute and hold for 1 minute, and then increased to290c at a rate of 10 degree for 2 minute, the result were showed as chromatogram and mass spectrum and were compared to Nist library attached with device.

2.5.2.1 Calculation of concentrations of targeted alkaloids in plant parts of targeted plants

For calculation of concentrations of targeted alkaloids in this study, the peak highest of standard samples was depended a reference. The calculations were donning according to the equation:

$$1: X/\emptyset = y/\alpha$$

$$2: X = \frac{y \times \emptyset}{\alpha}$$

X = concentration of targeted alkaloid in plant part.
Y= peak height of alkaloid targeted in plant part.
$\emptyset = concintrationof\ standard\ sample.$
$\alpha = peak\ height\ of\ stanrdrd\ alkaloid\ sample.$

Chapter Three

3. Results and Discussion

Chapter Three

2. Results and Discussion

3.1 Results of TLC analysis:

At 10 cm run for the mobile Retention Factor RF for the standard samples of atropine, hyoscine, nicotine and solanine were 2, 5.3, 3.6, and 2.3 respectively. All plant parts of *Dutara inoxia* except the root have shown positive result for atropine hyoscine, while the only positive result for nicotine was obtained from leaves samples, there were no any positive results for solanine .*Datura stramonium* L showed the same pattern of results, but a positive result of nicotine was collected from stem. These facts were reported by (Babiker *et al* 2017). The plant part of *Hyoscyamus albus* positive results was similar to the results of *Dutara inoxia*, which come in agree with what was published in the US Botanical and Plant Chemistry Database (USDA) in 2004. A positive result for solanine was observed in the fruits sample of *Lycium deserti phil*with another positive of nicotine for the leaves of the same plant, that is in agreement with the findings of (Matthews 1994). All plant parts samples of *Nicotiana restica* were showed positive result for nicotine.

All *Solanum dubium* plant parts except leaves showed positive results for solanine, the leaves and stem samples of this plant showed positive result for nicotine. The stem, leaves and fruits samples of *Solanum incanim* showed positive results for solanine, the leaves also showed positive result for solanine.

3.2 Result of GCMS analysis:

3.2.1 GCMS analysis of alkaloids standards:

Atropine standard sample chromatogram showed a sharp peak at retention time Rt for 14.3 minute (figure 3.1), the mass spectrum of this peak Showed multiple mass fragments values starting from 40 and including the values 42, 82, 96 , 124, 140, 159, 180, 200, 207, 227, 256, 272 and 289 . It is clear from the spectrum that the main fragment mass value was 124 while the highest fragment mass was 289 which dictates to molecular Weight of the Atropine (figure 3.2). In Hyoscine standard chromatogram a peak was appeared in the chromatogram at retention time of 14.5 minute (figure 3.3), the mass spectrum fragments values started from 40. However, it was included the same mass values collected from Atropine spectrum the highest mass fragment value was 303 which compatible with the molecular Weight of Hyoscine , the main fragment mass value of this peak was 124, while figure (3.4) For Nicotine standard the Retention Time of the peak was 6.3 (figure 3.5). The mass spectrum showed highest mass fragment value of 163 in a similarity with value of the molecular weight of Nicotine, the spectrum started from the fragment mass value of 26 and included the mass values 42, 44, 51, 56, 78. 84, 92, 105, 119, 130, 133, 145 and 163.the main fragment mass value was 84 (figure 3.6). solanine standard sample chromatogram showed sharp peak at Retention time of 7.6 minute (figure 3.7) the mass spectrum of this molecule started with a fragment in mass value of 240 the spectrum included the mass values of 347, 398, 440, 560,639, 722 while the highest fragment mass value of the mass spectrum of these peak was 868 as well as it is main fragment value (figure 3.8). All above results were compared to the internal library of the instrument; the similarity of each standard sample was 99% .table 5.1 is showing the peaks heights of Atropine, Hyoscine,

Nicotine and solanine which were 1711480, 6793721, 1652376 and 1583462 respectively. In this study the peak height of each standard sample as was adopted as reference value for calculations of concentrations of targeted alkaloids of tested samples.

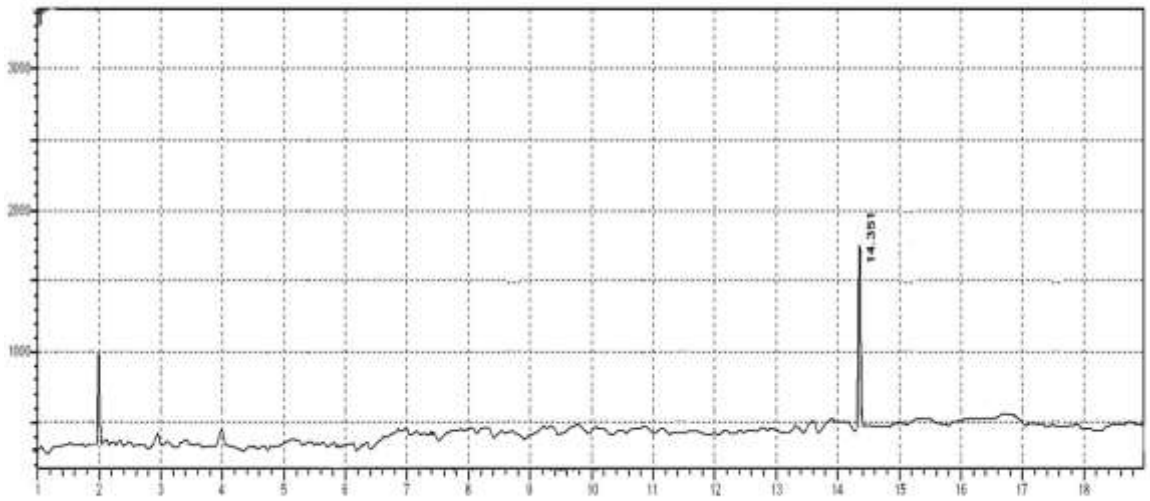

Figure 3.1: GC chromatogram of Atropine standard sample

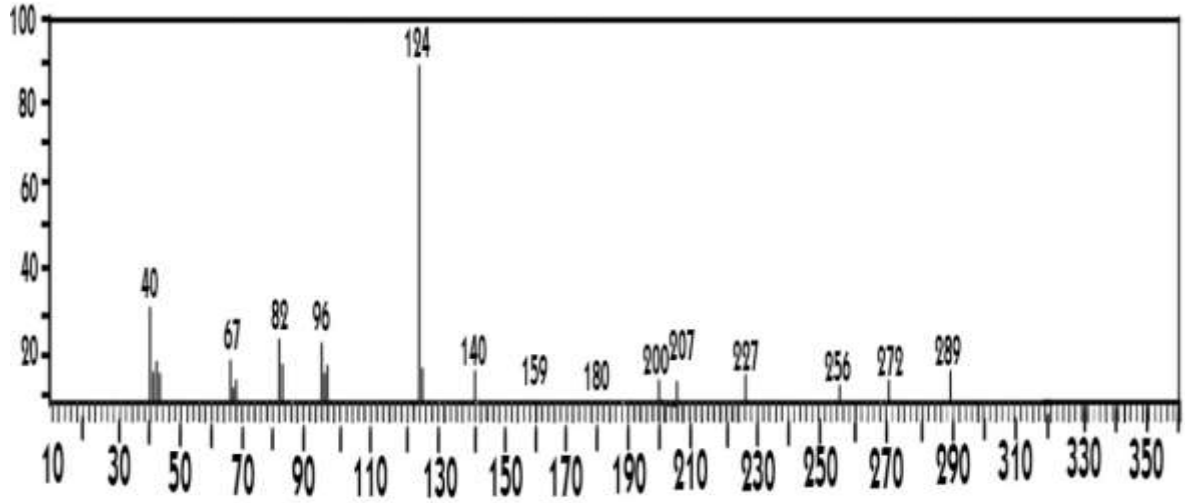

Figure 3.2: Mass Spectrum of Atropine standard sample

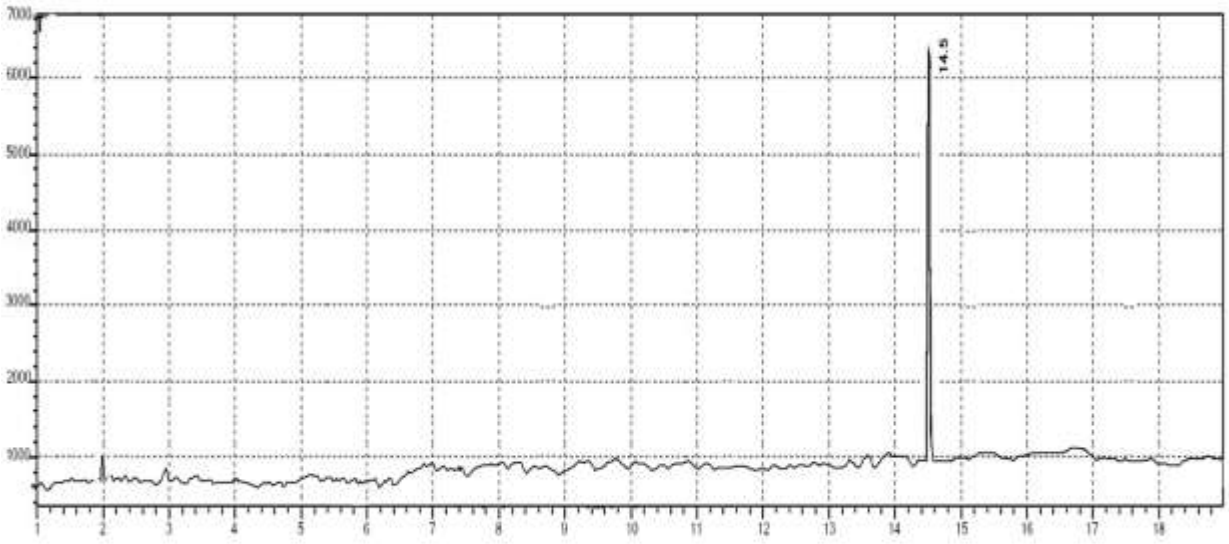

Figure 3.3: GC chromatogram of Hyoscine standard sample

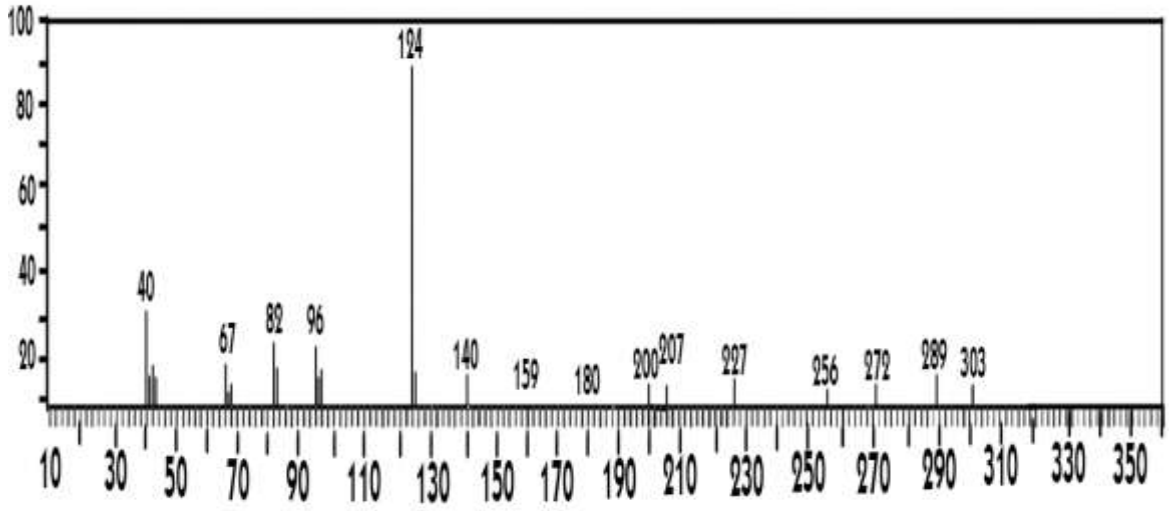

Figure 3.4: Mass Spectrum of Hyoscine standard sample

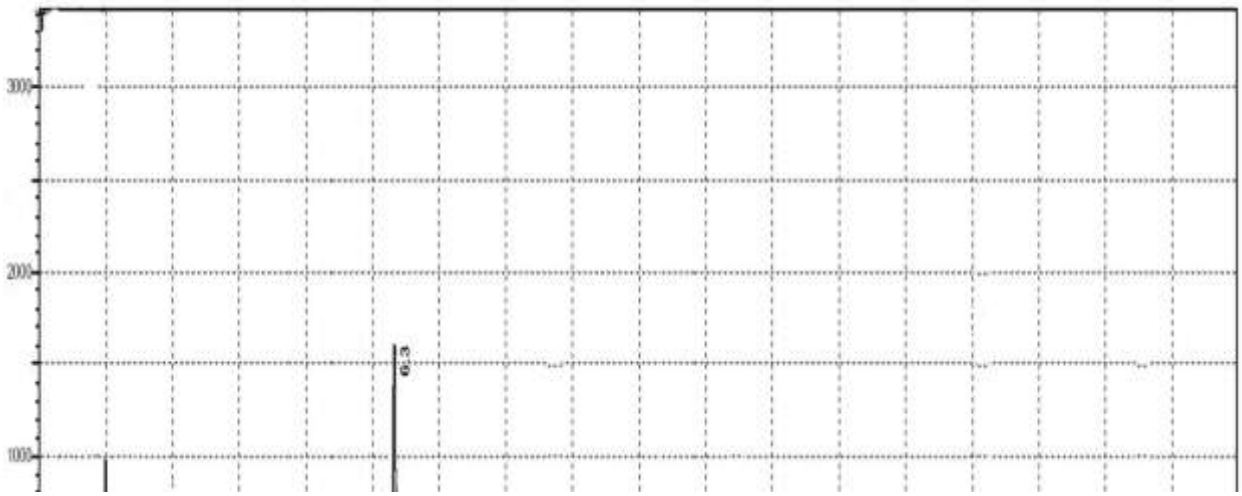

Figure 3.5: GC chromatogram of Nicotine standard sample

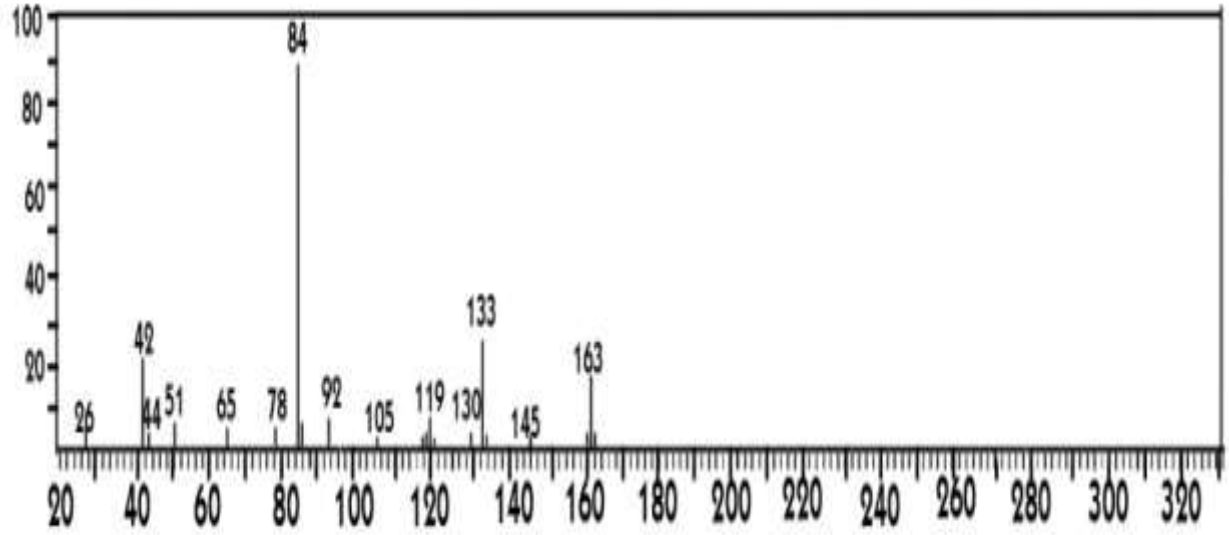

Figure 3.6: Mss Spectrum of Nicotine standard sample

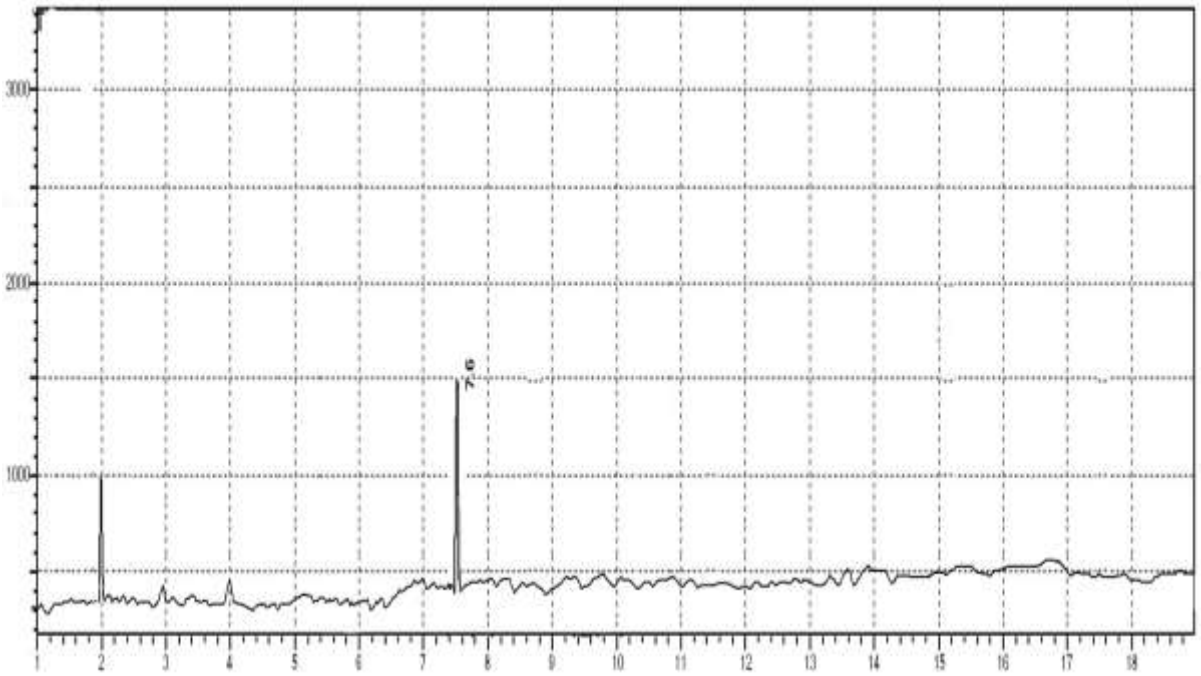

Figure 3.7: GC chromatogram of solanine Standard sample

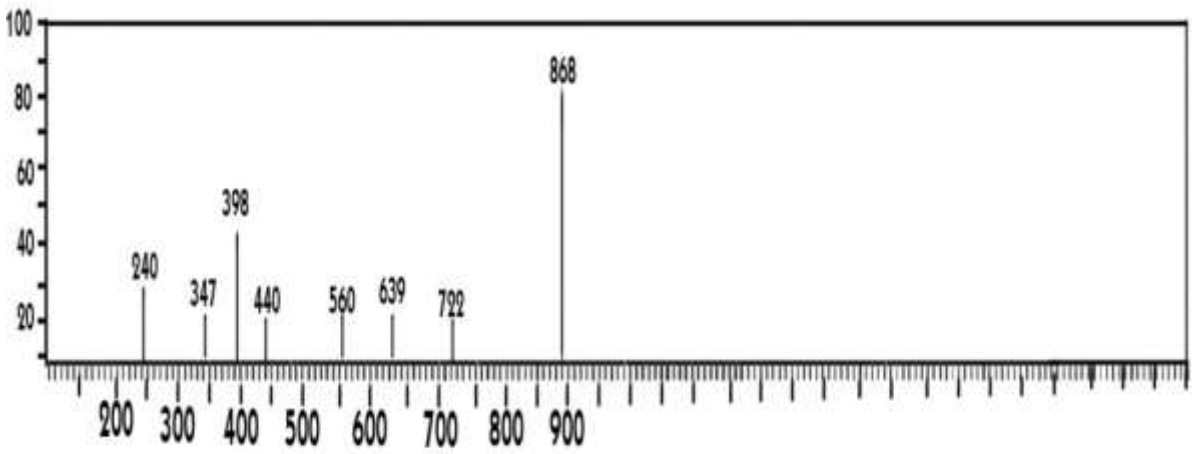

Figure 3.8: Mass spectrum of solanine standard sample

Table 3.1: Peaks heights of standard samples

Number	Standard sample	Standard peak height
1	Atropine	1711480
2	Hyoscine	6793721
3	Nicotine	1652376
4	Solanine	1583462

3.2.2 Results of *Datura Innoxia*:

In the chromatogram of stem sample of *Datura innoxia* figure 3.9 showed a peak at retention time of 14.32 minute, the mass spectrum of this peak showed multiple fragment, mass started with the value of 40 and ended with 289 with main fragment value of124, the other fragments in the mass spectrum matched with which was collected from the mass spectrum of the standard sample of atropine, the mass spectrum was compared to internal library the similarity was 87%. These findings confirm the existent of atropine in the sample. In The same chromatogram a peak was showed at Retention Time of 14.5 minute the mass spectrum of this peak was in the same pattern of the previous peak including the main mass fragment value of 124, the different from what was before that highest fragment value was 303, this spectrum is in compatible with the spectrum of the Hyoscine standard sample, it was compared to the internal library the similarity was 88% which confirm presence of the hyoscine in the sample.

In the chromatogram of analysis for leaves of *D. noxia* Figure 3.10 two peaks were observed at retention time of 14.323, 14.573 respectively, the two peaks were showed two mass spectra, they started with fragment value of 40 with main fragment value of 124. The highest mass value for the peak at 14.323 was 289 was indicated to Atropine. This spectrum was compared

to internal library it was matching for 89%. The mass value of 303 was the end value in the spectrum of the peak at 14.573, it is highest value in Hyoscine, and the similarity of comparison to internal library was 88%. According to the above facts, it was confirmed that atropine and hyoscine were existed in the sample.

In the same chromatogram at retention time 6.3 minute a peak was appeared. The mass spectrum of this peak was started with the fragment mass value of 26, the highest mass value was the fragment of 163 and main mass fragment value was 84 with almost other fragment similar to fragment of the mass spectrum of the Nicotine standard. The comparison to the internal library showed similarity of 82% which was guiding to confirm existence of nicotine in the sample.

The chromatogram of fruit sample of *Datura innoxia* (Figure 3.11) showed a peak at retention time 14.331 minute, the mass spectrum of this peak showed highest fragment mass value of 289 and the main fragment value was 124, the other fragment in the mass spectrum were similar to what was collected from the mass spectrum of the standard sample of atropine. This mass spectrum was compared to internal library the similarity was 89%. This result confirmed the existence of atropine in the sample. The same sample chromatogram showed a peak at Retention Time 14.563 minute, the highest fragment value in the mass spectrum of this peak was 3.3, and the main mass fragment value was 124, the other fragment of this mass spectrum were consistent with the mass spectrum of the hyoscine standard sample. The similarity of compression to the internal library was 83% which was confirmed the existent of the Hyoscine in the sample.

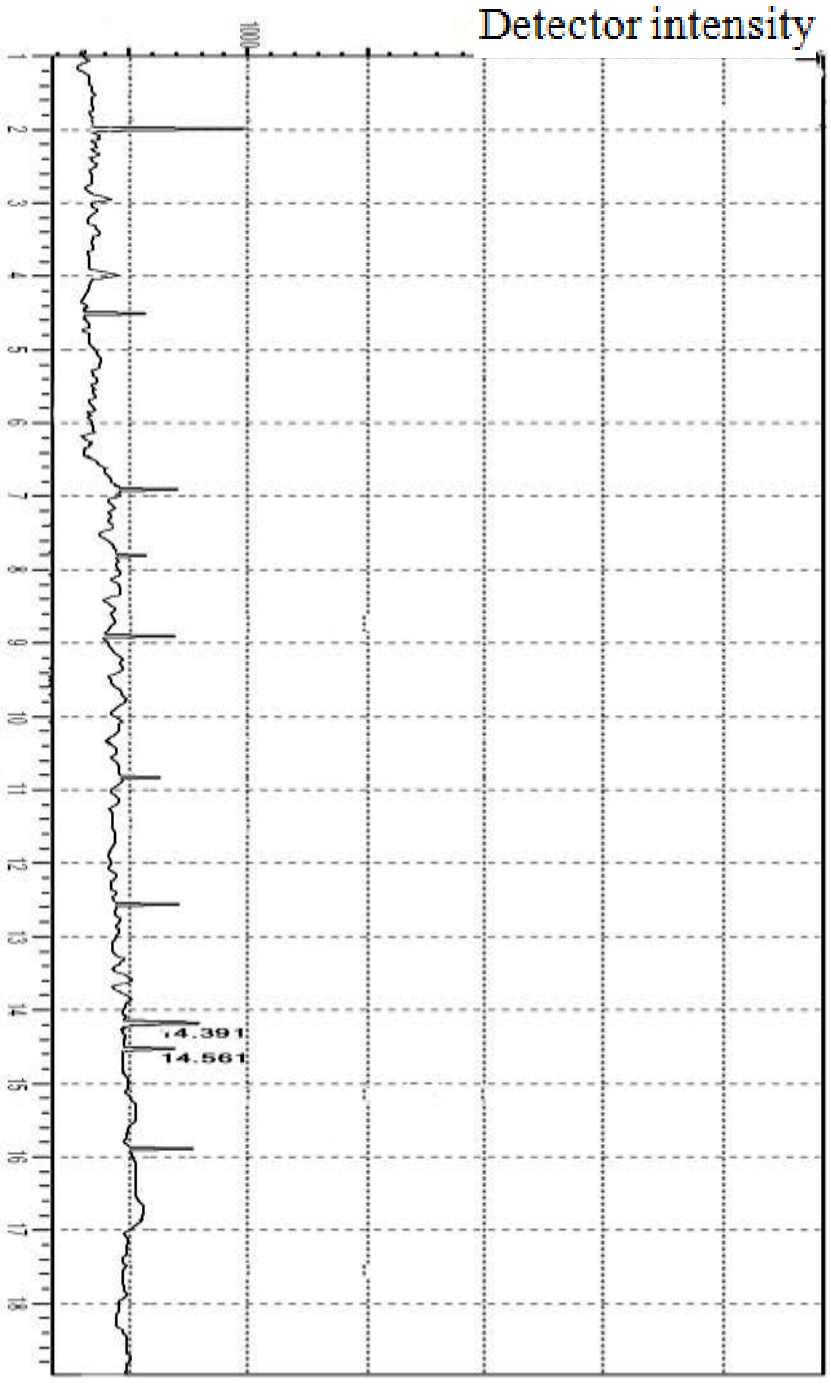

Figure 3.9: GC Chromatogram of *D. innoxia* stem sample

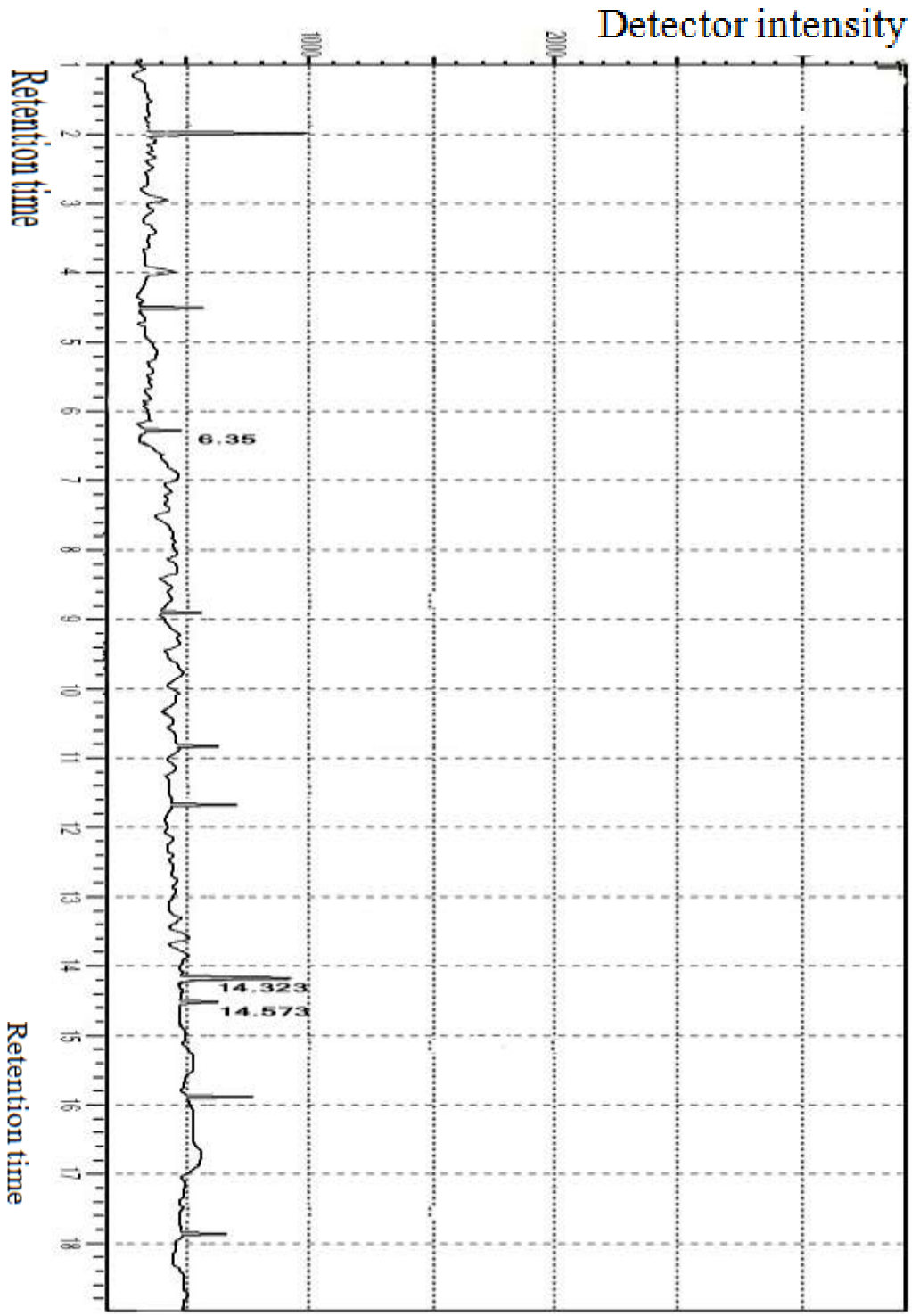

Figure 3.10: GC Chromatogram of *D. innoxia* leaves sample

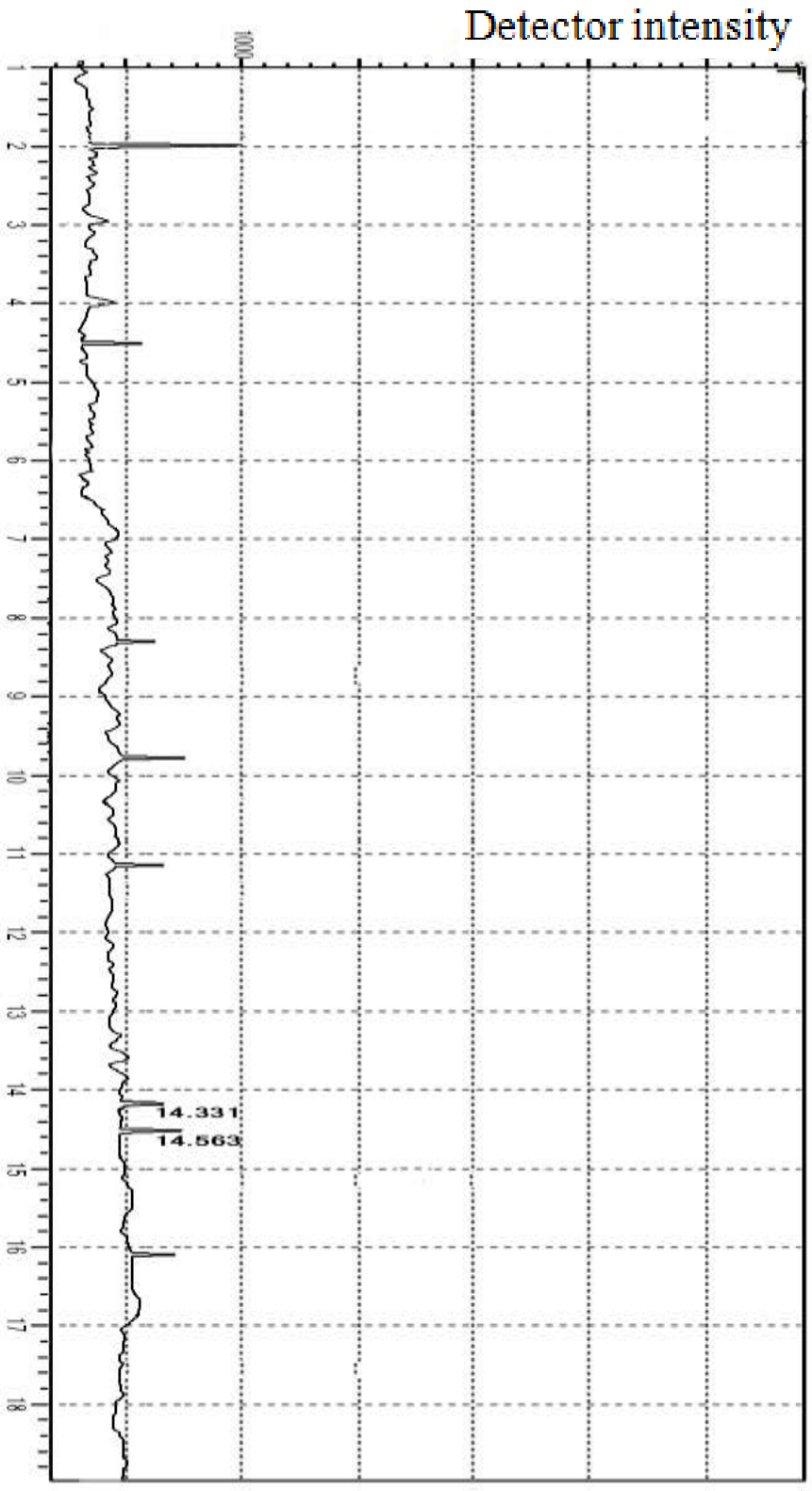

Figure 3.11: GC Chromatogram of *D. innoxia* fruit sample

Table 3.2: peak heights of *D. innoxia* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	52457	38134	nd	nd
Leaves	74231	37743	37326	nd
Fruits	37521	50374	nd	nd
Roots	nd	Nd	nd	nd

- nd : not detected

3.2.3 Results of *Datura stramonium* plant parts:

The chromatogram of analysis for stem sample of *Datura stramonium* Figure 3.12 showed a peak at Retention Time 14.34 minute, the mass spectrum of this peak showed highest fragment mass value of 289 and main fragment mass value of 124 , the other fragment in the mass spectrum was matching with the mass spectrum of the standard sample of Atropine. This mass spectrum was compared to internal library; the similarity was 87% that confirmed the existence of Atropine in the sample.

The same sample chromatogram shows a peak at 14.52 minute the mass spectrum of this peak started with a fragment mass value of 40, the highest fragment value was 303, and the main mass fragment value was 124 , the other fragment of this mass spectrum were similar to the mass spectrum of Hyoscine standard sample, the match of compression to the internal library was 88% which confirms the presence of Hyoscine in the sample, in the same chromatogram there was a peak at Retention time of 6.3 minute. The mass spectrum of this peak was started with fragment value of 26, the highest mass value was163 and main mass fragment value was 84 with

almost other fragment like to fragment of the mass spectrum of the Nicotine standard. The comparison to the internal library showed similarity of 87% which was guiding to confirm the occurring of Nicotine in the sample .

At Retention Time 14.34 minute in the chromatogram of analysis for leaves sample of *Datura stramonium* Figure 3.13 a peak was appeared. The mass spectrum of this peak showed highest fragment mass value of 289 and the main fragment mass value was 124. Another peak in the chromatogram noticed at Retention time of 14.56, as the previous the mass spectrum started with fragment mass value 40, the spectrum also included main fragment mass value of 124 and highest fragment value of 303. The other fragment in the two spectrum mass were in match with what was collected from the mass spectrum of the standard sample of Atropine and Hyoscine respectively , the two mass spectrum was compared to internal library the similarity was 89% and 88%. This result is confirming the existent of Atropine and Hyoscine in the sample.

The peaks at Retention times 14.33 and 14.527 were noticed in the chromatogram of analysis for fruit sample of *Datura stramonium* Figure 3.14 the mass spectrum of these two peaks started as expected for Atropine and Hyoscine with fragment mass value of 40, the two spectrum included main mass value of 124, the highest mass values in the two spectrum were 289 and 303 respectively. The comparison of the two spectrum led to the result that similarity of the first peak was 89% to Atropine, and the second peak was 87% to Hyoscine. This result is confirming the existent of Atropine and Hyoscine in the sample.

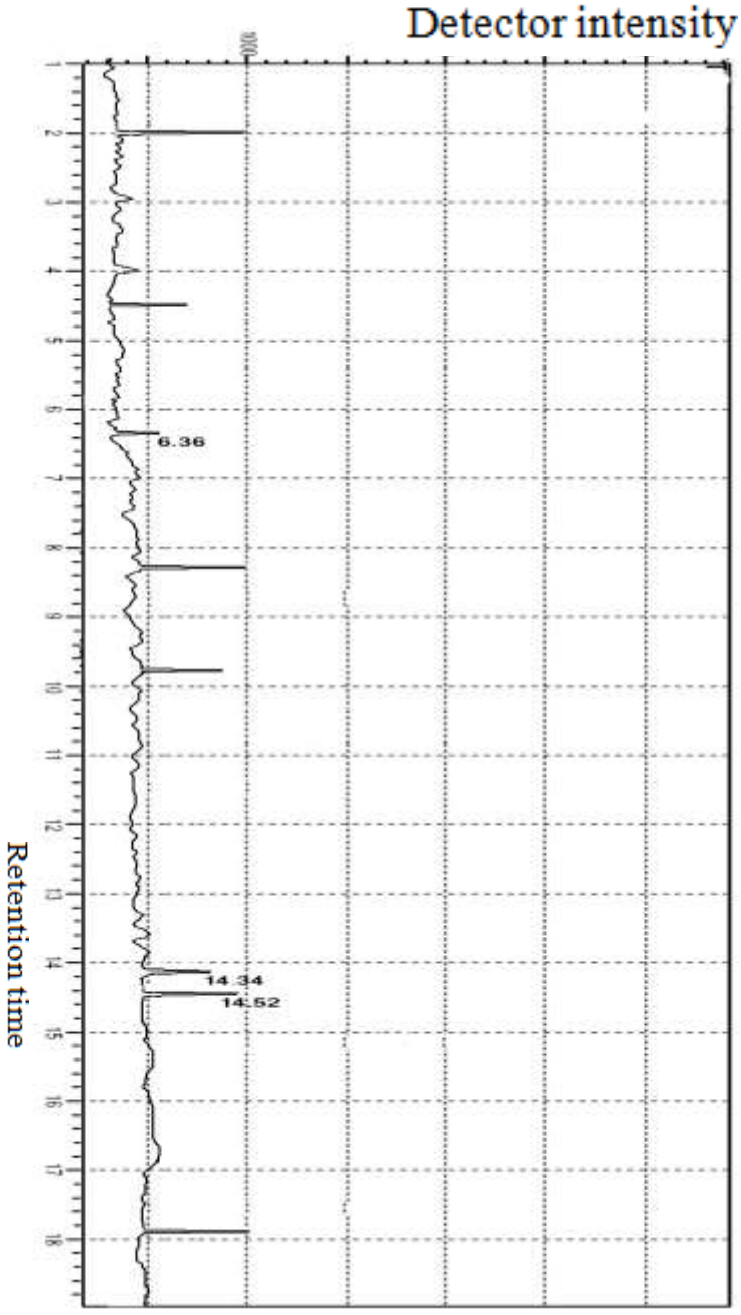

Figure 3.12: GC Chromatogram of *D. stramonium* stem sample

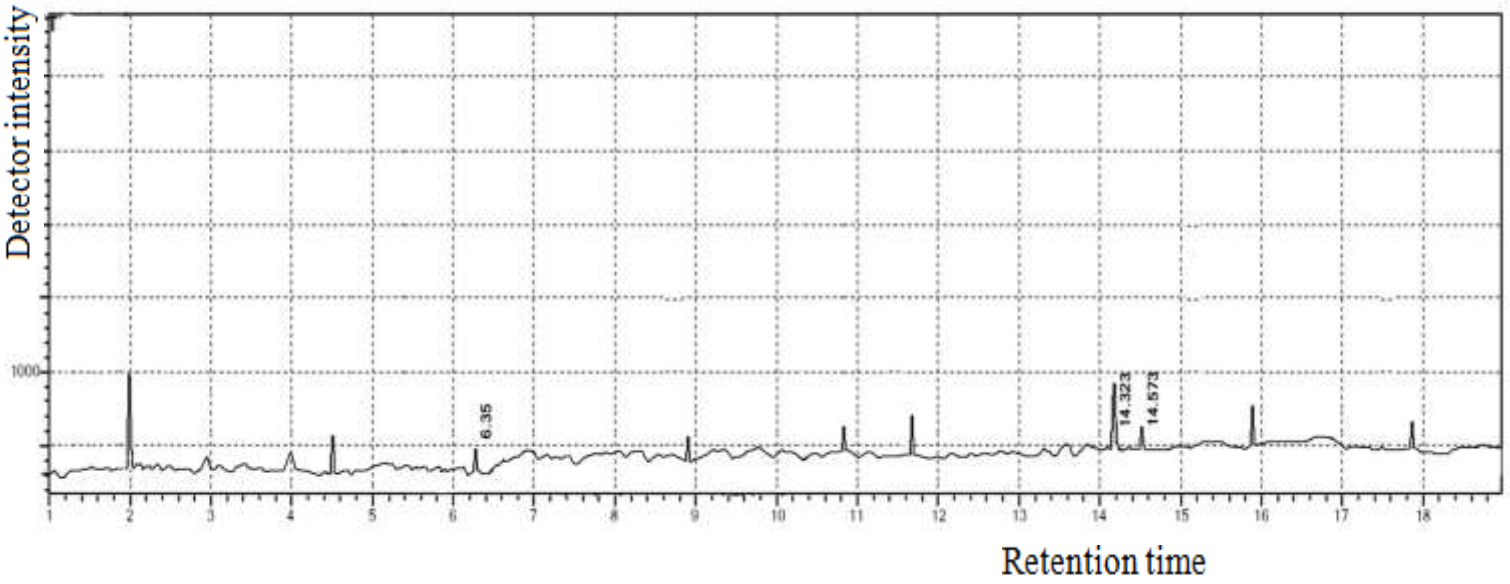

Figure 3.13: GC Chromatogram of *D. stramonum* leaves sample

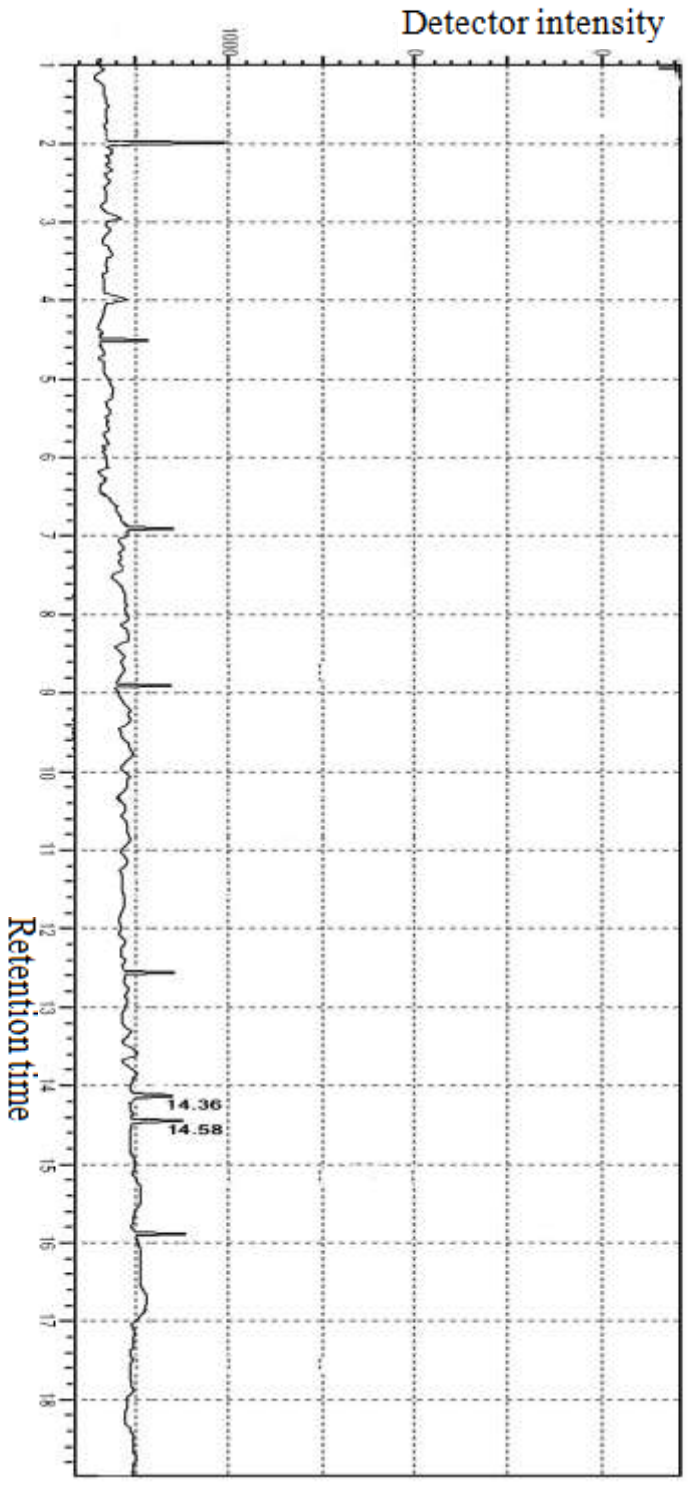

Figure 3.14: GC Chromatogram of *D. stramonium* fruit sample

Table 3.3: Peak heights of *D. stramonium* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	5322	82500	1844	nd
Leaves	86233	105000	37326	nd
Fruits	35834	38246	nd	nd
Roots	nd	nd	nd	nd

- nd : not detected

3.2.4 Results of *Hyoscyamus alpus* plant parts:

The chromatogram of the stem sample of *Hyoscyamus albus* (figure 3.15) showed a peak at Retention Time of 14.3 minute. The mass spectrum of this peak was in the pattern of Atropine, it was started with fragment value of 40. It showed highest fragment mass value of 289 and main fragment mass value of 124. This mass spectrum was compared to internal library, and the similarity was 87% to Atropine. This result is a confirmation of the existence of Atropine in the sample. The same sample chromatogram showed a peak at Retention time 14.5 minute. in this mass spectrum the highest fragment value was 303, and the main mass fragment value was 124,the other fragment of this mass spectrum were consistent with the mass spectrum of the Hyoscine standard sample. The similarity of comparison to the internal library was 83%, which was confirming the existent of the Hyoscine in the sample.

The chromatogram of *Hyoscamus albus* leaves sample (Figure 3.16) shows a peak at 14.32 minute. The mass spectrum of this peak showed highest fragment mass value 289 and main fragment mass value 124. The other

fragment In the mass spectrum was in agreement with the mass spectrum of the standard sample of atropine. This mass spectrum was compared to internal library and the similarity was 86% to Atropine, which confirms the presence of Atropine in the sample. The same sample chromatogram showed a peak at 14.57 minute. The mass spectrum of this peak highest fragment value was 303 and the main mass fragment value was 124. The similarity of comparison to the internal library was 87% to Hyoscine, which confirms the presence of Hyoscine. In the same chromatogram at retention time 6.32 minute a peak was observed the mass spectrum of this peak started with fragment mass value of 26 and showed highest fragment mass value of 163 and main mass fragment value was 84 with almost other fragment similar to fragments of the mass spectrum of the Nicotine standard. The comparison to the internal library showed similarity of 89% to Nicotine which confirms the sample contains Nicotine . The fruits sample of *Hyoscyamus albus* Figure 3.17 was analyzed, the GC chromatogram shows two peak at Retention times 14.37 and14.58 minute.

The mass spectrum of these peaks started with fragment mass value of 40 and was a main fragment mass value124, the highest fragment mass values was 289 and 303 respectively, the two spectra were compared to the internal library and the similarity was 88% and 86% to Atropine and Hyoscine, which indicates the presence of these alkaloids in the sample.

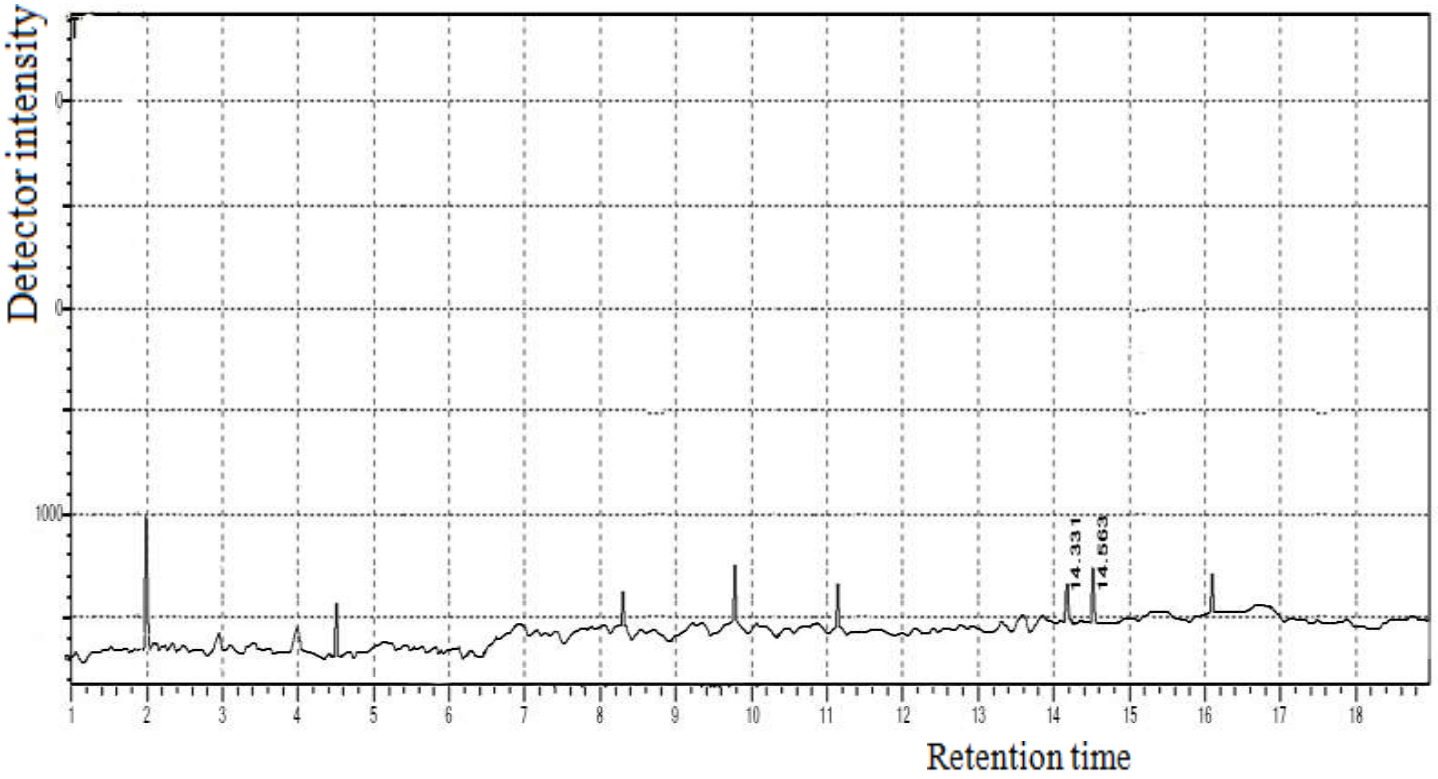

Figure 3.15: GC Chromatogram of *H. albus* stem sample

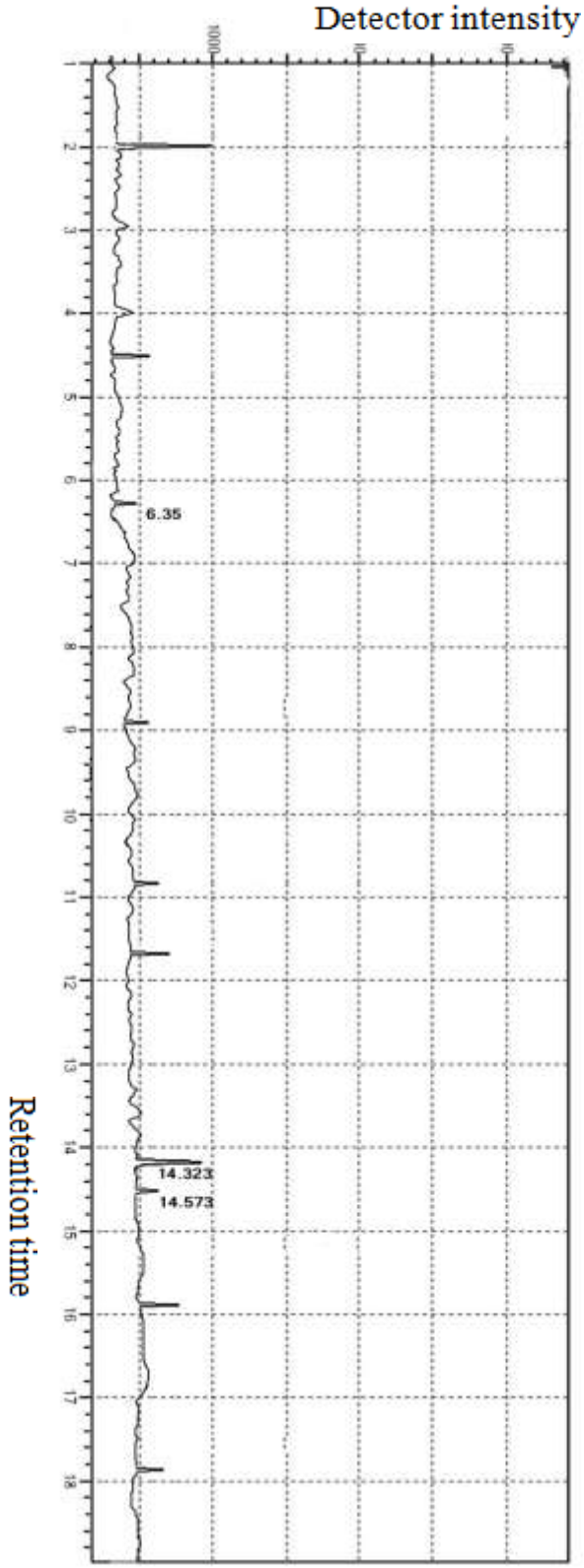

Figure 3.16: GC Chromatogram of *H. albus* leaves sample

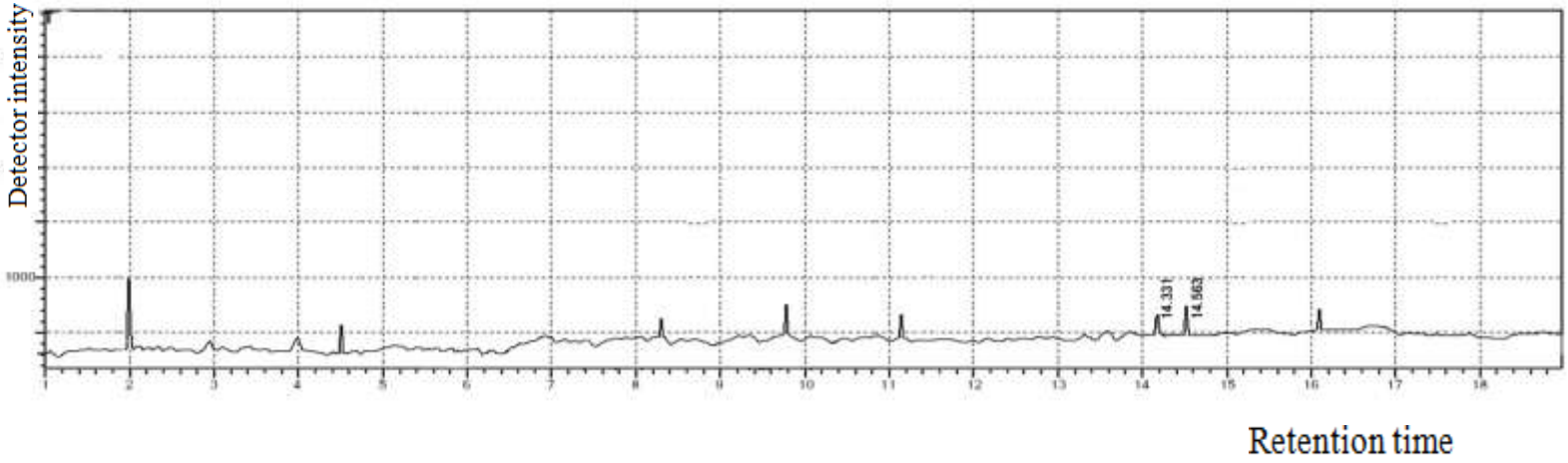

Figure 3.17: GC Chromatogram of *H. albus* fruit sample

Table 3.4: Peak heights of *H. albus* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	52457	50374	nd	Nd
Leaves	74257	37743	37437	Nd
Fruits	375324	39245	-	Nd
Roots	nd	nd	nd	Nd

- nd : not detected

3.2.5 Results of *Licium deserti phil* plant parts:

Figure3.18 is showing the chromatogram of GC analysis for leaves sample of *Lycium deserti phill.* A peak was observed at Retention time of 6.37 minute. The mass spectrum pattern of this sample was similar to the Nicotine, it started with mass fragment value of 26, the highest mass value was163 and main mass fragment value was 84. The comparison to the internal library showed similarity of 89% which confirms the presence of Nicotine in the sample.

The chromatogram of analysis for the fruit sample Figure 3.19 showed a peak at Retention time 7.651 min. The mass spectrum of this peak started with fragment mass value of 242 and a final mass fragment of 868 which was also the main fragment mass value and it represents the molecular weight of Solanine. This spectrum compared to the internal library showed 85% similarity. This confirms the presence of Solanine in the sample.

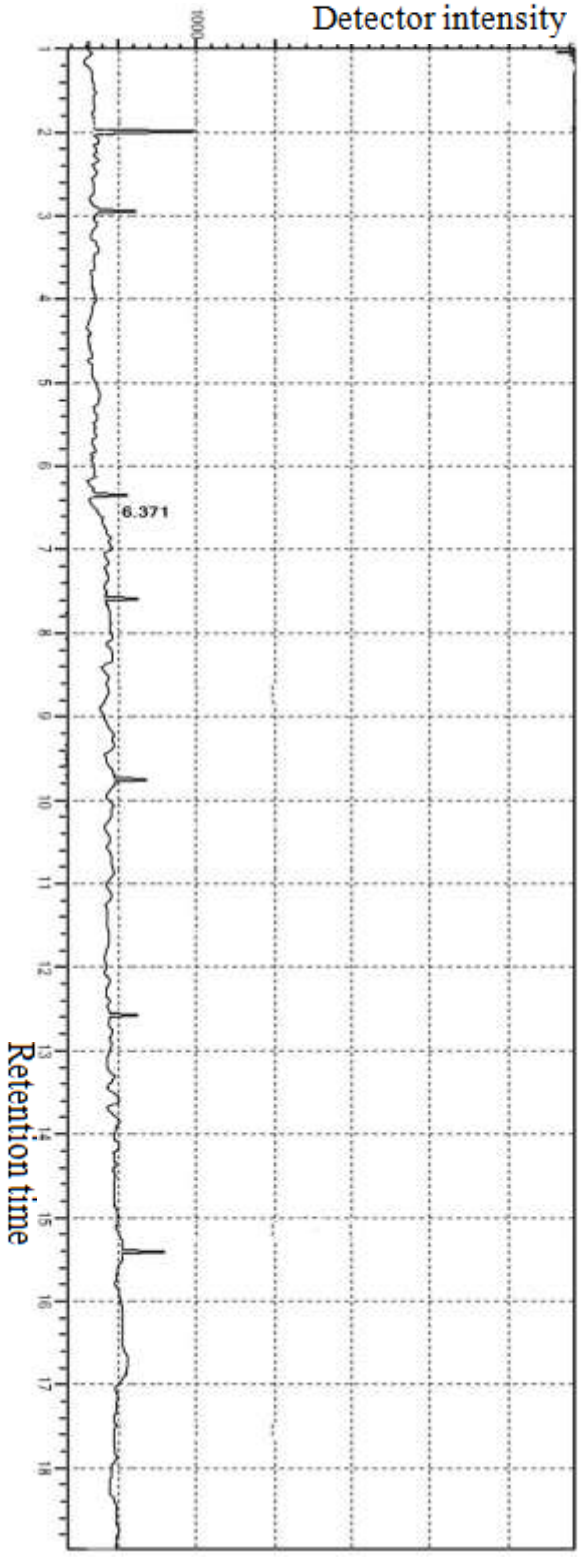

Figure 3.18: GC Chromatogram of *L. deserti phill* leaves sample

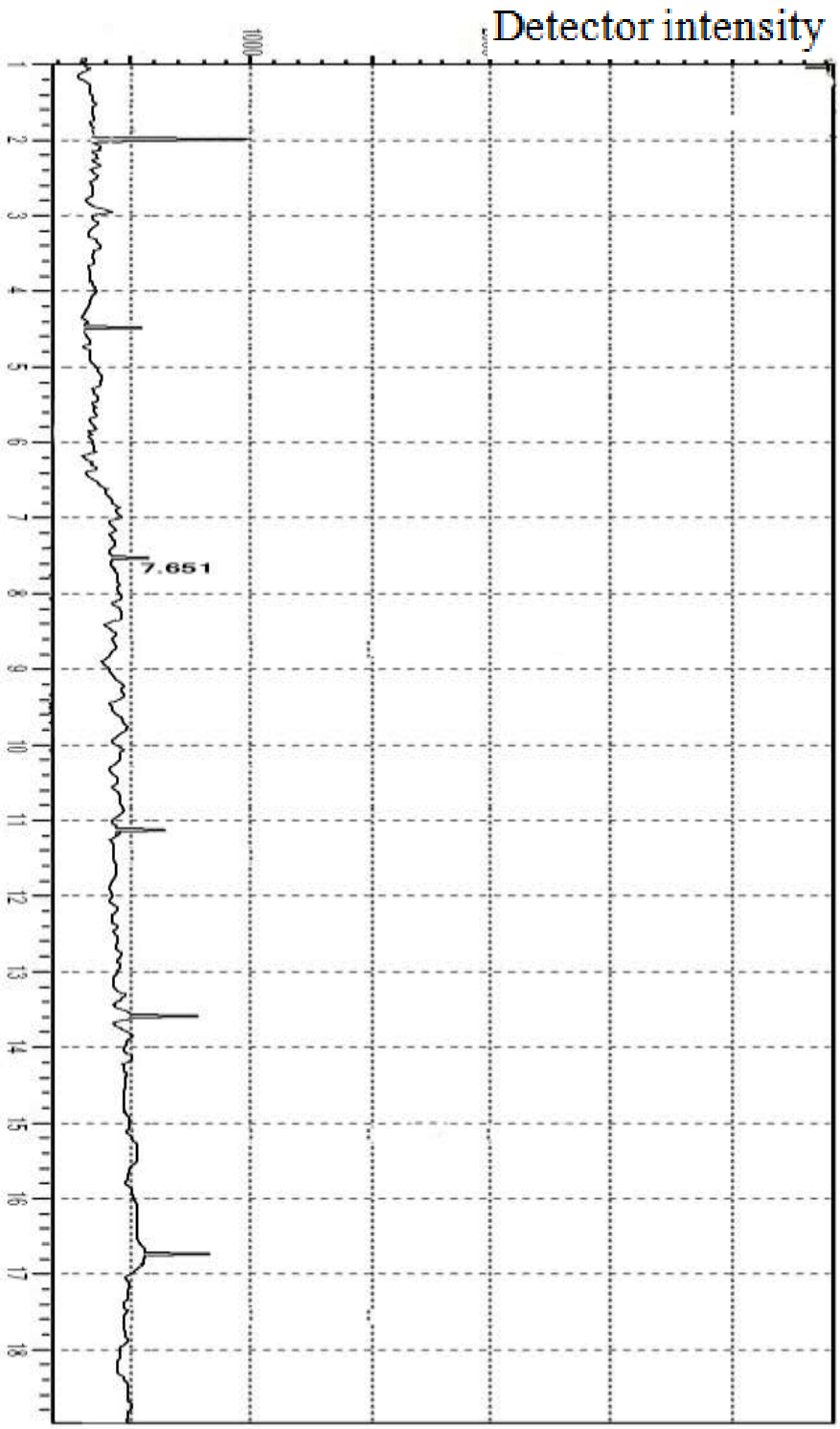

Figure 3.19: GC Chromatogram of *L. deserti phill* fruit sample

Table 3.5: Peak heights of *L. deserti phill* plant part samples

Plant part	Peak height			
	Atropine	**Hyoscine**	**Nicotine**	**Solanine**
Stem	nd	nd	Nd	Nd
Leaves	nd	nd	47654	Nd
Fruits	nd	nd	Nd	21537
Roots	nd	nd	Nd	Nd

- nd : not detected

3.2.6 Results of analysis of *Nicotina rustica* plant parts

Figures 3.20, 3.21, 3.22, 3.23 are showing chromatograms of analysis of stem, leave, fruit and root samples of *N. restica*, all chromatograms show peaks in Retention Time around 6.3 minute. All mass spectra of those peaks was started with the mass fragment value of 26, and in agreement with the mass spectrum of Nicotine standard. The main fragment mass value was 84 and the highest mass value was 163. The comparison to internal library was 85 - 89% for all spectrums. There was no doubt Nicotine a present in all samples.

The chromatogram of fruit sample figure 3.22 shows a peak at Retention time of 7.591. The mass spectrum of this peak started with mass value of 242 the highest and main fragment value was 868 which is compatible with the spectrum of solanine. It was compared to the internal library, the similarity was 88%, and thus it confirms the sample contains Solanine.

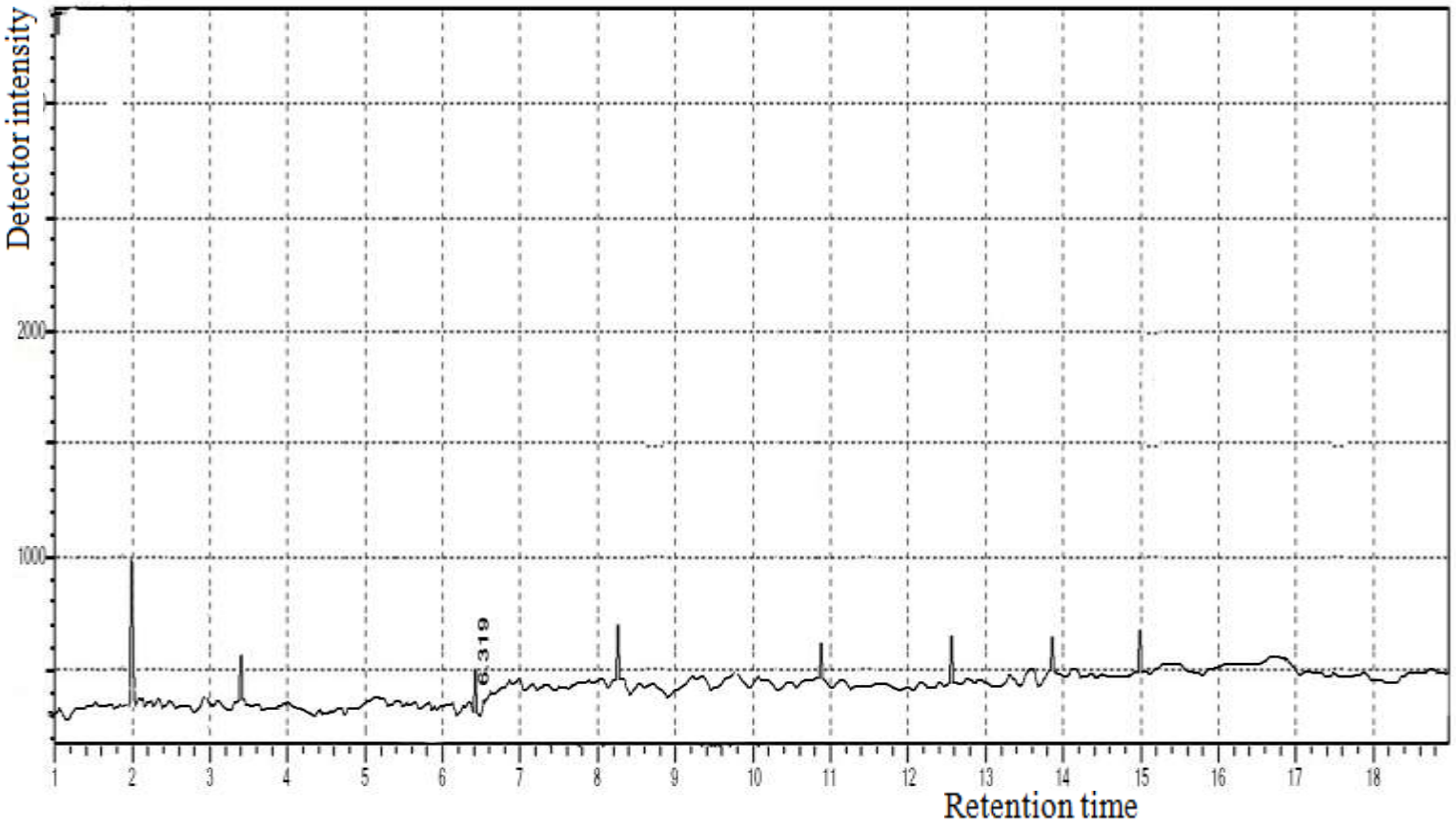

Figure 3.20: GC Chromatogram of *N. rustica* stem sample

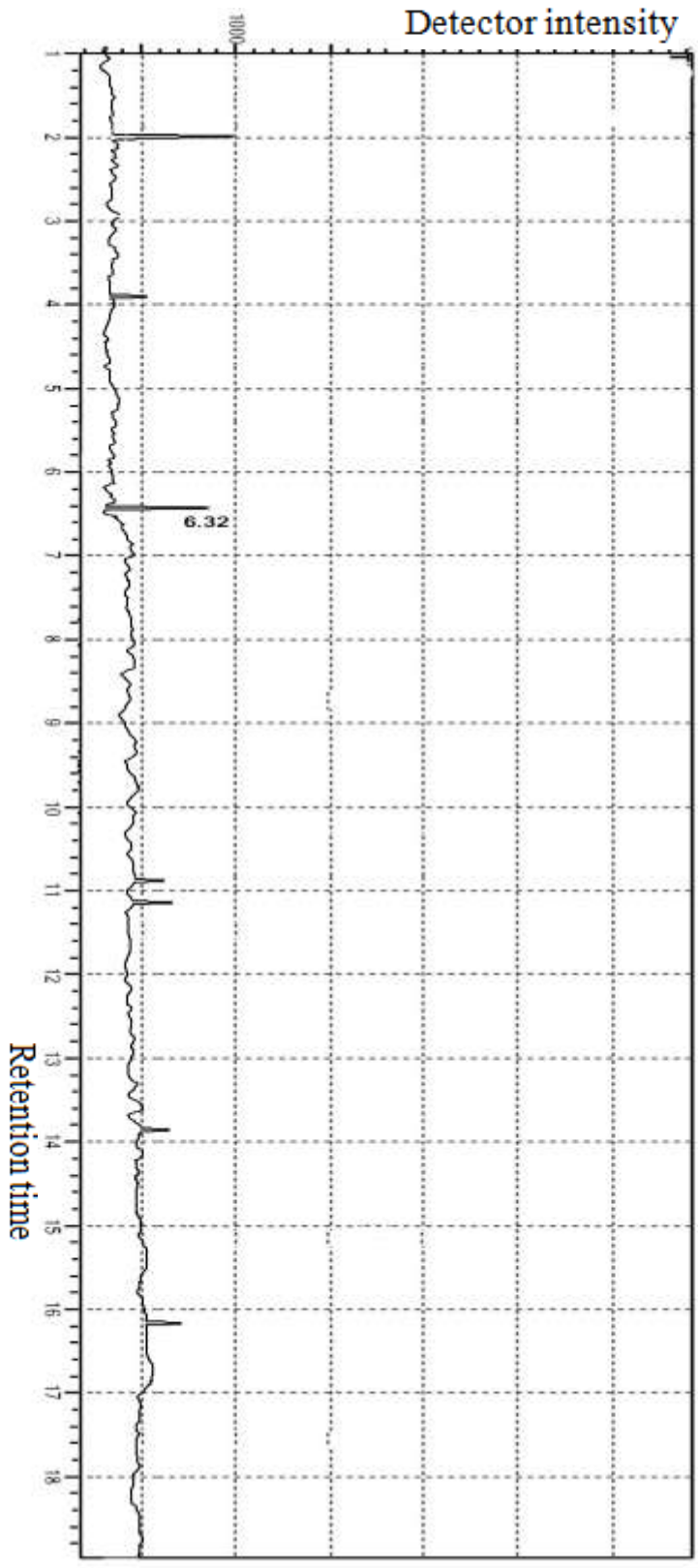

Figure 3.21: GC Chromatogram of *N. rustica* leaves sample

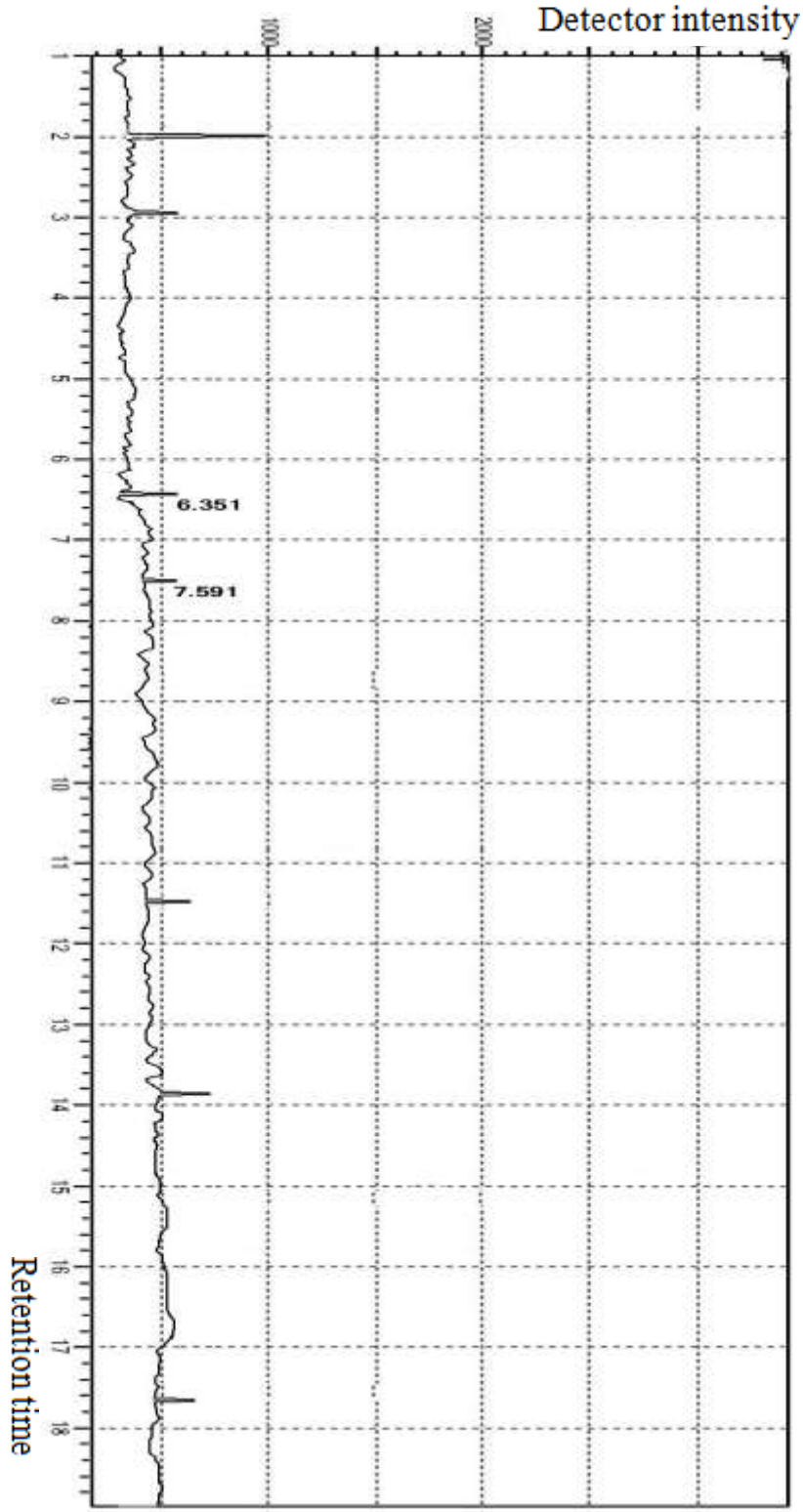

Figure 3.22: GC Chromatogram of *N. rustica* fruit sample

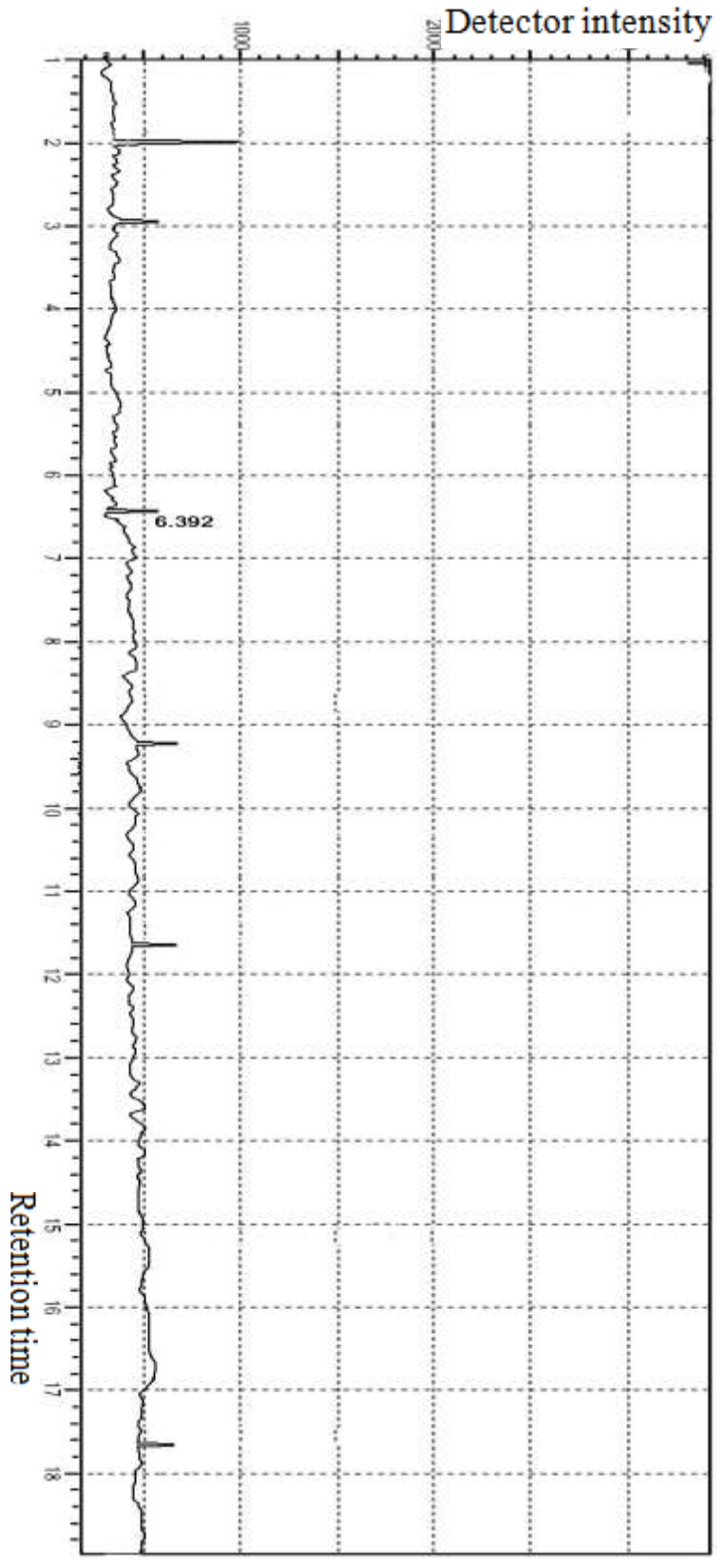

Figure 3.23: GC Chromatogram of *N. rustica* roots sample

Table 3.6: Peak heights of *N. rustica* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	nd	29887	nd
Leaves	nd	nd	74277	nd
Fruits	nd	nd	30322	28910
Roots	nd	nd	29243	nd

- nd : not detected

3.2.7 Results of analysis of *Sulanum dubium* plant parts

Chromatograms of analysis for stem, fruits, and root of *S. dubium* (Figures 3.24, 3.26, 3.27) show peaks at Retention Time around 7.6 minutes. Mass spectra of those peaks were almost like the mass spectrum of Solanine standard. They started with fragment mass value of 242 and highest main fragment mass value of 868, the spectrum were compared to internal library and the similarity was 88%, 85% and 83% respectively. It confirms that all samples contain solanine.

Leaves samples chromatogram figure 3.25 showed a peak at Retention time of 6.371. The spectrum of this peak started with mass fragment of 26 the main fragment mass value was 84 and the highest fragment mass value was 168 in agreement with the Nicotine standard. The comparison to internal library was 87%. It confirms that the sample contains Nicotine.

In the chromatogram of root sample (Figure 3.27) a peak appeared at Retention time of 14.52 minute (figure 3.28). The mass spectrum of this peak was unclear, it started with fragment mass value of 40 and included the values of 67, 124, 256, 289 and 303 which were found in mass spectrum. The main fragment mass value was 96 and the highest mass

value was 310. Hence it is not possible to identify any one of the targeted alkaloids; it can't be judged that any of them were exist in the sample.

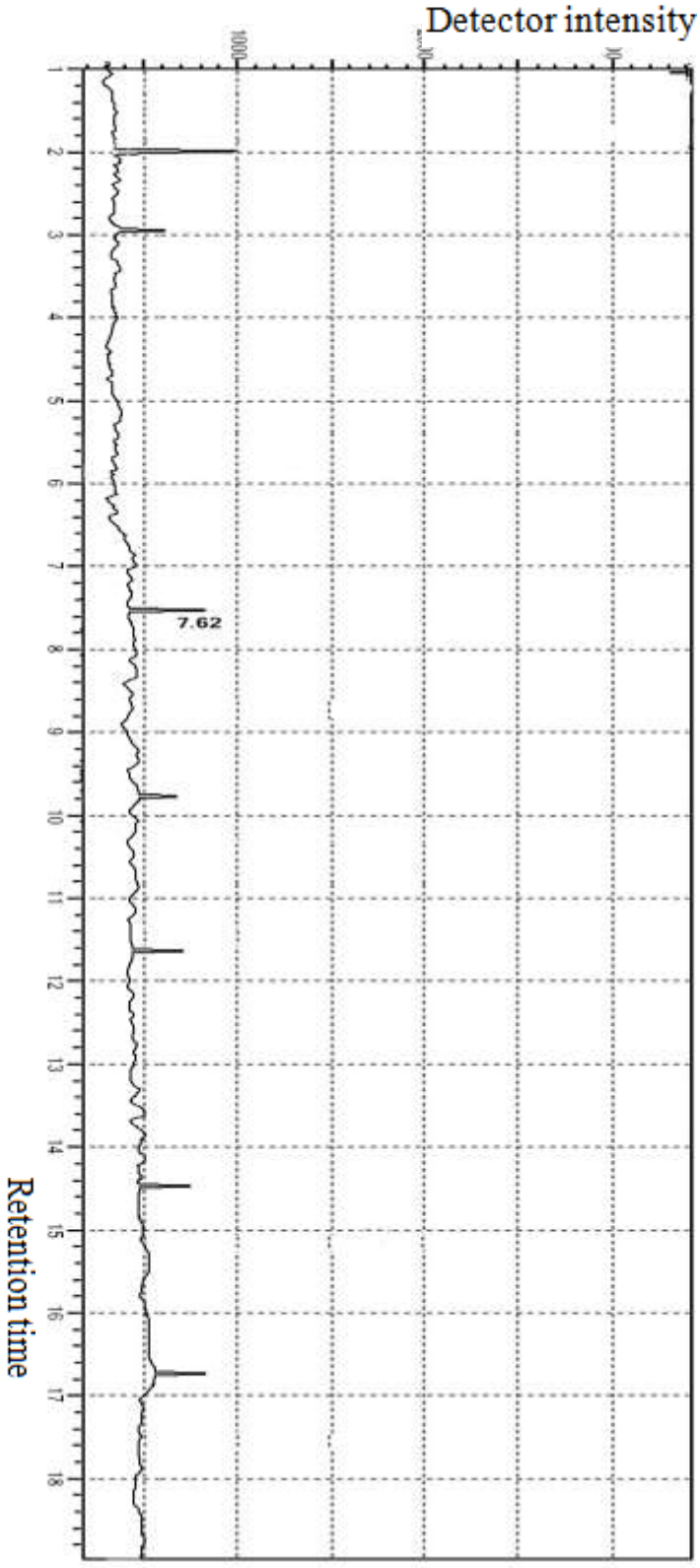

Figure 3.24: GC Chromatogram of *S. dubium* stem sample

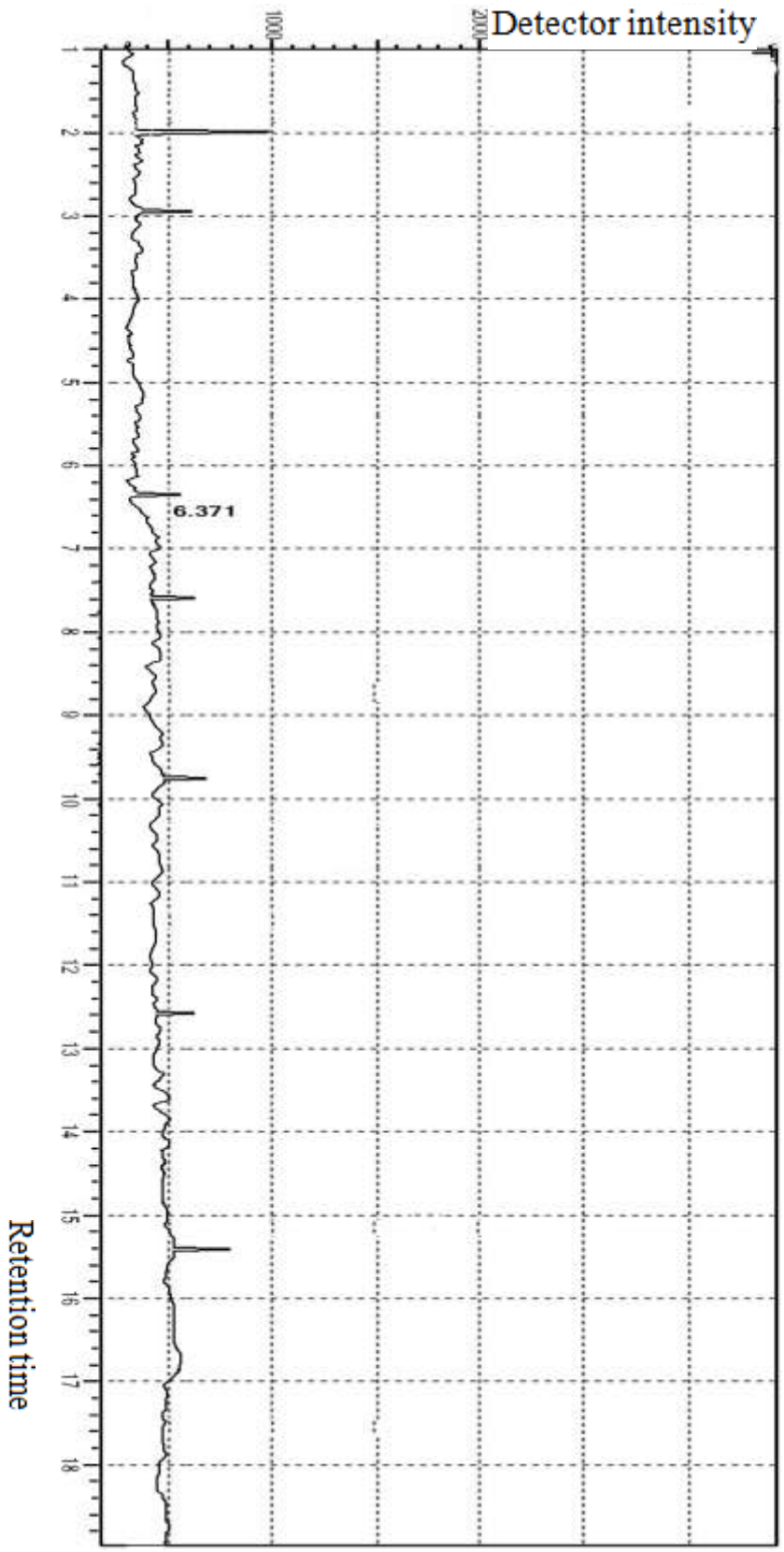

Figure 3.25: GC Chromatogram of *S. dubium* leaves sample

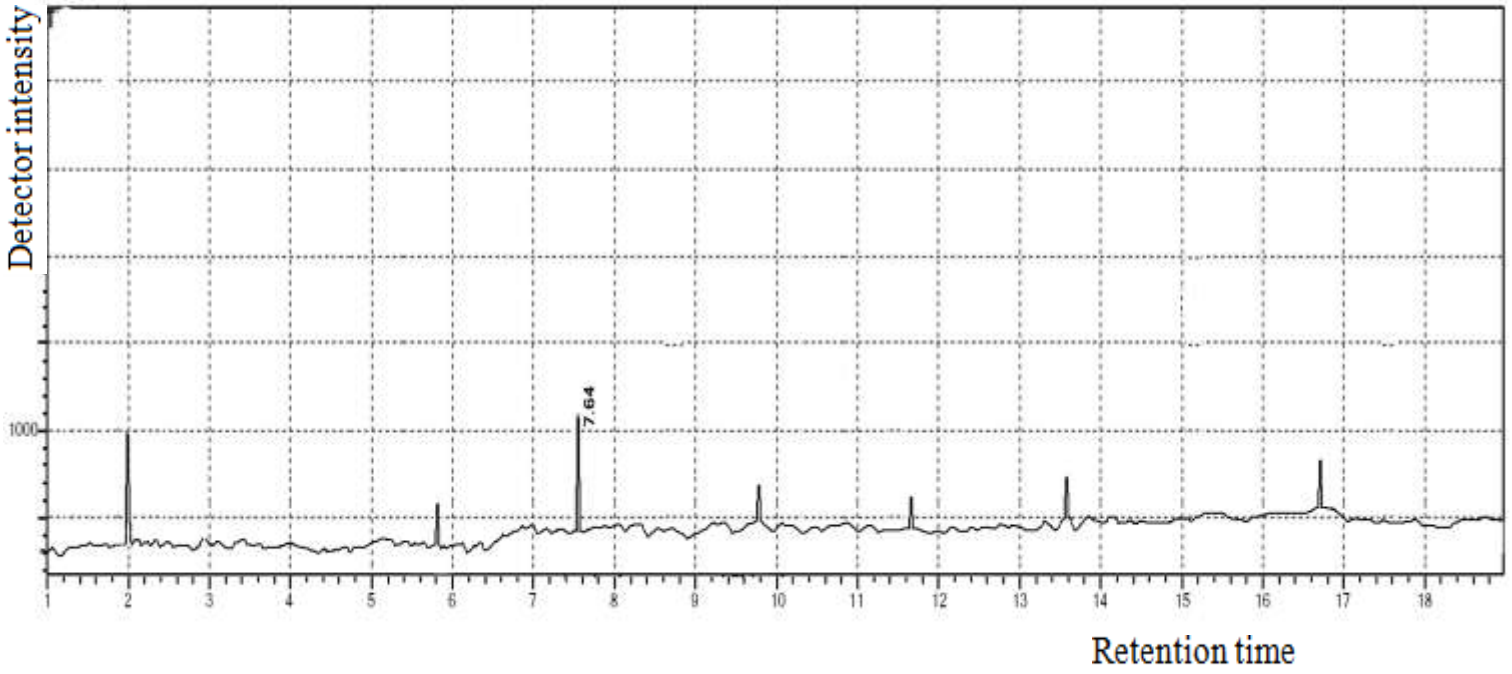

Figure 3.26: GC Chromatogram of *S. dubium* fruit sample

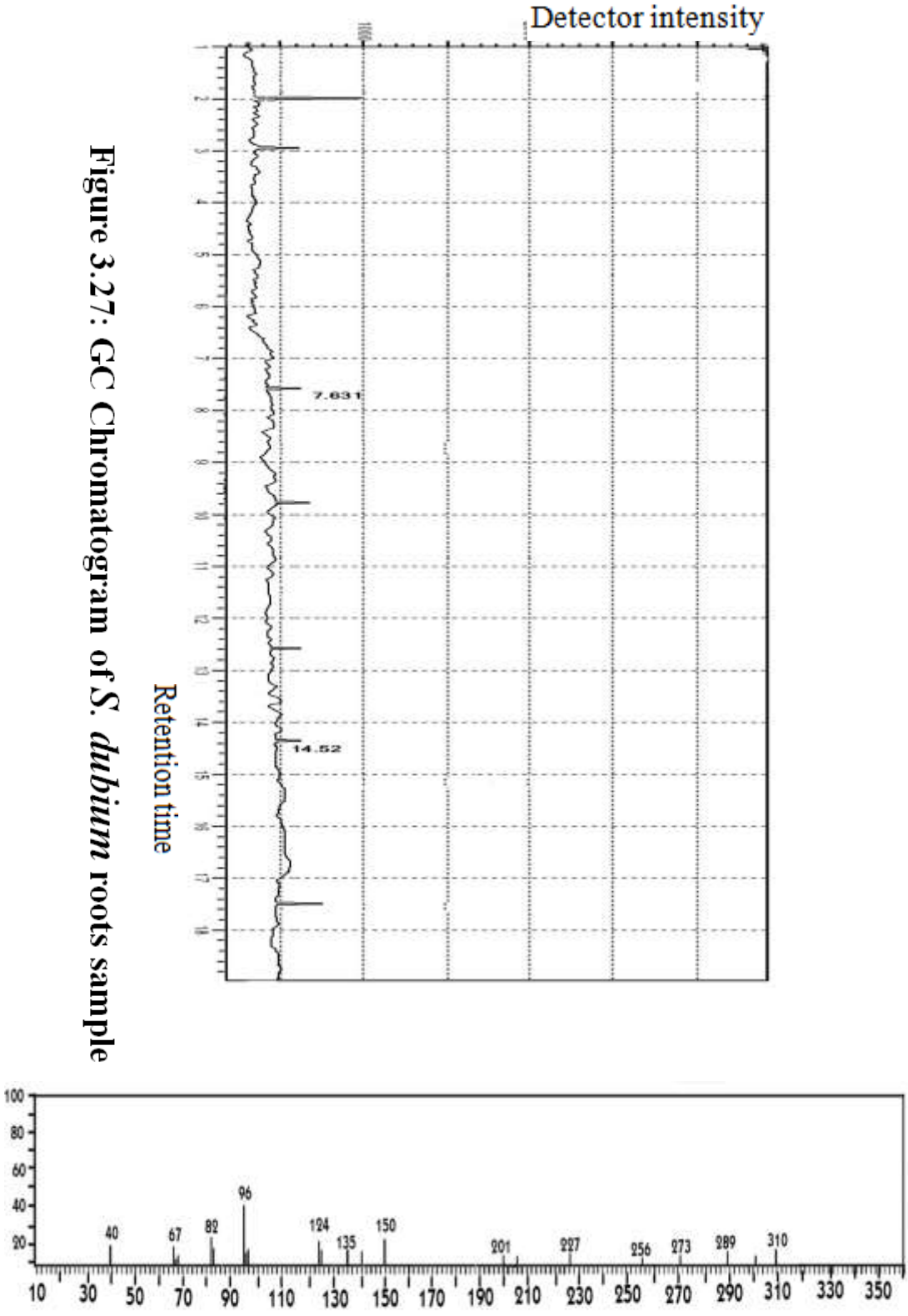

Figure 3.27: GC Chromatogram of *S. dubium* roots sample

Figure 3.28: mass spectrum of the peak at 14.52 minute in figure 3.27

Table 3.7: Peak heights of *S. dubium* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	Nd	nd	2123	72316
Leaves	Nd	nd	31045	-
Fruits	Nd	nd	nd	125390
Roots	Nd	nd	nd	31045

- nd : not detected

3.2.8 Results of analysis of *Solanum incanum* plant parts

Chromatograms of stem, leaves and frouits of *Sulanum inicanm* (figures 3.29, 3.30, 3.31) were included peaks at Retention Times around 7.6 minute, the mass spectra of those peaks almost like what was collected from Solanine standard starting with 242 fragment mass value and 868 main fragment mass value as well as it is the highest. The similarity to internal library was more than 86% for each spectrum confirming existence of Solanine in samples.

In the chromatogram of leaves analysis (figure 3.30) there was a peak at Retention Time of 6.37 minute. Mass spectrum of this peak was started with 26 and the main fragment mass value was 84 and the highest was 168, the similarity in internal library was 89% to Nicotine. It confirms the presence of Nicotine in the sample.

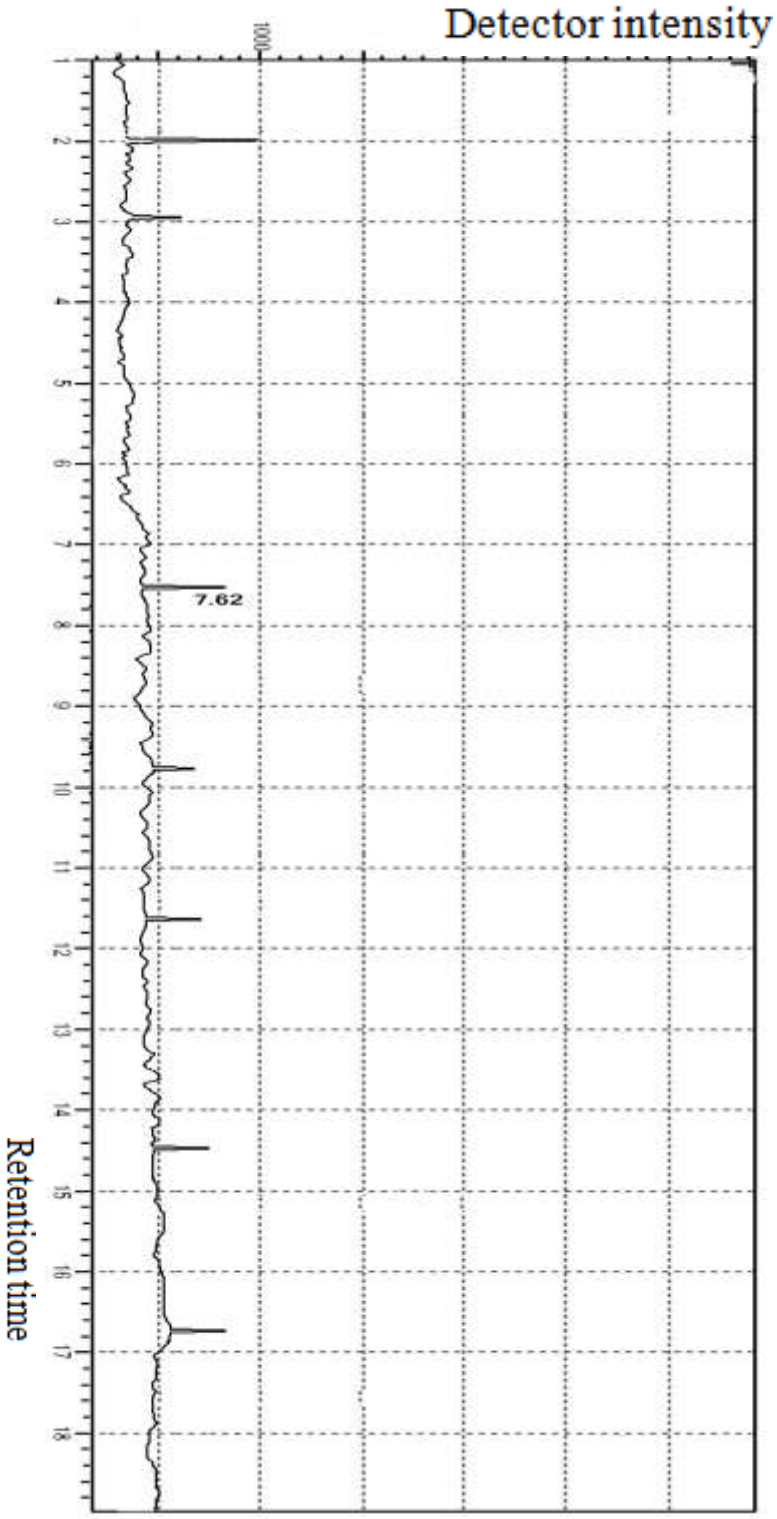

Figure 3.29: GC Chromatogram of *S. incanum* stem sample

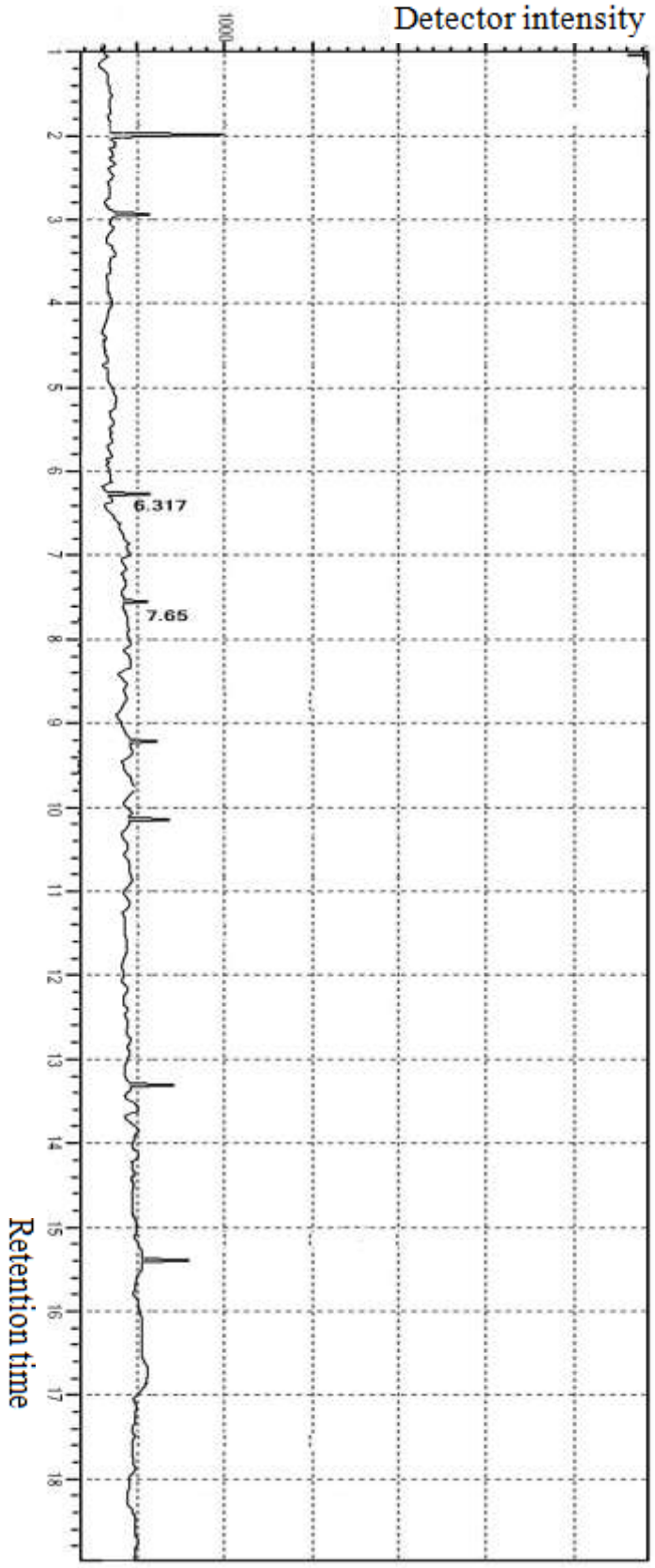

Figure 3.30: GC Chromatogram of *S. incanum* leaves sample

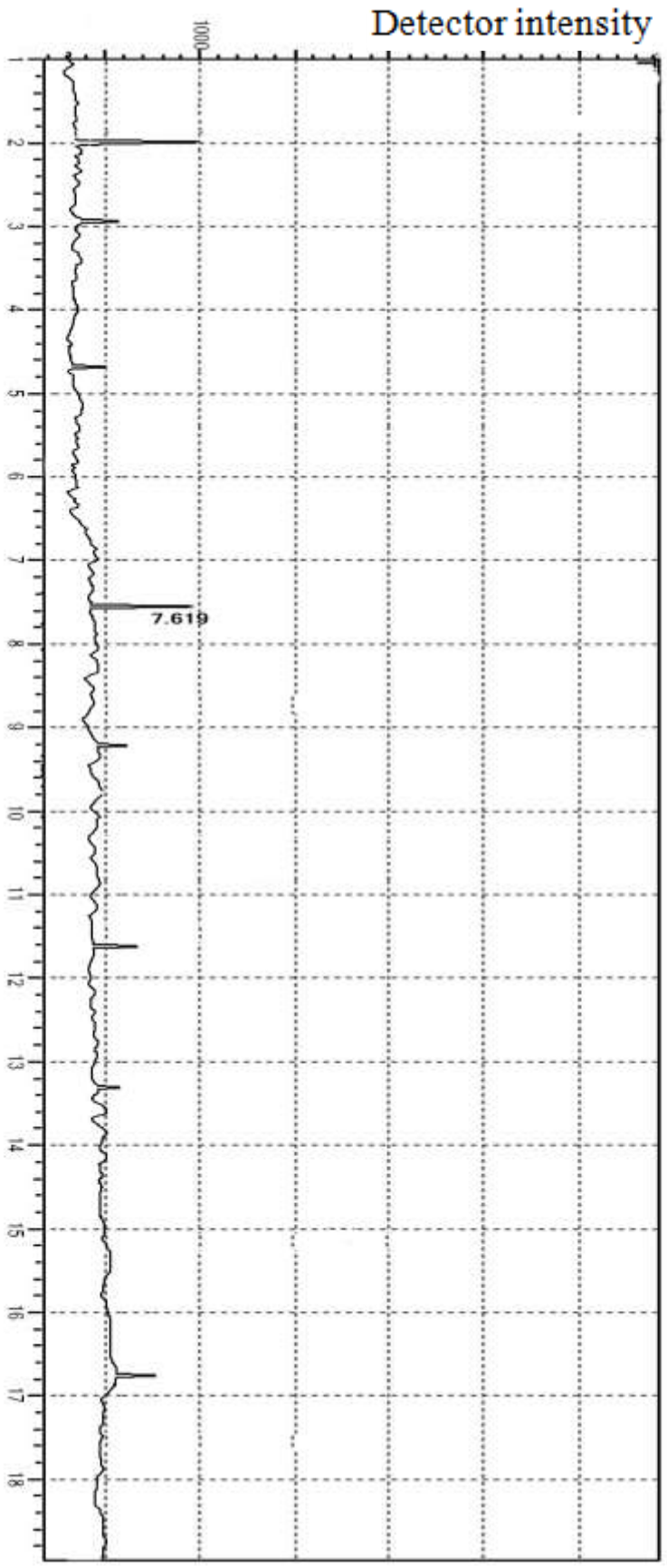

Figure 3.31: GC Chromatogram of *S. incanum* fruit sample

Table3.8: peak heights of *S. incanum* plant part samples

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	Nd	nd	53133
Leaves	nd	Nd	29522	30156
Fruits	nd	Nd	nd	97651
Roots	nd	Nd	nd	nd

- nd :not detected

3.3 Results Calculation of concentrations of targeted alkaloids in plant parts of targeted plants

Table 3.9 is showing concetrictions of targeted alkaloids in *Datura innoxia* plant part samples. Atropine concentration was in the range of 0.03 mg/g for stem sample, 0.04 mg/g for leaves samples and 0.02 mg/g in fruit samples, the study did not find any Atropine concentration in the root samples, Hyoscine concentration was in the range of 0.05 mg/g for stem and stem samples, it was lower in fruit samples, the rang was only 0.01 mg/g, the leave sample was showed a very low concentration of Nicotine in the range of 0.007 mg/g. these finding were in agreement with the findings of Babiker *et al* (2017), the study done by Adel and Wasan (2009) stated that the yield of Atropine was in the range of 0.03mg/g, Mean concentrations of Atropine for plant parts in this study was in the same rang. Sidra *et al* 2017 reported that the hyoscine concentrations in *D. innoxia* vary from 0.005 to 0.03 mg/g, without determination of plant parts, which almost agrees with this study.

The concentrations of targeted alkaloids in *Datura stramonium* was shown in Table 3.10. Atropine concentrations was in the same pattern with what

was found in *Datura innoxia*, it is what was stated by (Babiker *et al* 2017), the same range of concentrations for Atropine was founded by (ALIREZA *et al* 2005). Hyoscine concentration was lower than *Datura stramonium*, it was in the range of 0.01mg/g for stem and leaves, and it was 0.05mg/g for fruits, the same range of concentration was founded by (Jan *et al* 2008). Nicotine was in the range of 1 and 2 mg/g in compatible with what was showed (Tadeusz 2015).

However in this study Solanine didn't detect in *Datura,* it is suggested that it is due to type of soil and ecological effects, that is in agree with what was reported by (Guillermo 2018).

Hyosyamus albus (table 3.11) concentrations showed relative concentrations to *Dtura spp*. Atropine was in concentrations in rang of 0.03, 0.04 and 0.02 mg/g for stem, leaves and fruit respectively. Eeva *et al* 1998 was founded matched results . Hyoscine concentrations were in the range of 0.07 mg/g in stem and leaves, the concentration in fruits was in the range of 0.02mg/g .which was agreed with what was stated by Alaghemand (2013). Leaves was contained a range of 0.02 mg/g in compatible with (Tadeusz 2015).

Table 3.12 is showing the concentrations of targeted collected from *Lyceum deserti phil.* Nicotine was accumulated in leaves in range of 0.01 mg/g while solanine was found in fruits in rang of 0.01 as a characteristic feature for family Solanaseae. (Tadeusz 2015).

Nicotiana restica concentrations was showed in table (3.13), it is manly Nicotine in stem, fruits and roots in range of 0.01 mg/g, the concentration in leaves was in range of 0.04 mg/g , this concentration is higher than *Nicotiana tobacum* which relative to what was stated by (Katherine 2008). Solanine was found in the fruit in arrange of 0.01mg/g.

Table 3.14 is showing concentrations of the targeted alkaloids in *Solanum dupium*, it is manly solanine in stem, fruits and roots in rang of 0.04, 0.07

and 0.02 mg/g respectively. Those concentration were what was expected in *solanum spp*.According to the findings of (Neslihan 2006). A trace of Ncotine was found in stem in a range of 0.001 and it was found in leaves in range of 0.02.

Concentrations of *Solanum incanum* is not so far (table 3.15) it showed solanine in range of 0.03, 0.02, 0.06 in stem, leaves and fruit. Nicotine was found in leaves in range of 0.02.

Table 3.9: Concentrations of targeted alkaloids in *D. innoxia* plant parts

Plant part	**Concentration mg/g**			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	0.03434	0.05613	nd	nd
Leaves	0.042938	0.05555	0.00741	nd
Fruits	0.02170	0.0125	nd	nd
Roots	nd	nd	nd	nd

Table 3.10: Concentrations of targeted alkaloids in *D. stamonium* plant parts

Plant part	**Concentration mg/g**			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	0.03078	0.01214	0.01104	nd
Leaves	0.04988	0.015455	0.022363	nd
Fruits	0.2072	0.05627	nd	nd
Roots	nd	nd	nd	nd

Table 3.11: Concentrations of targeted alkaloids in H. albus plant parts

Plant part	Concentration mg/g			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	0.03034	0.07414	nd	nd
Leaves	0.04295	0.07434	0.022411	nd
Fruits	0.2171	0.02375	nd	nd
Roots	nd	nd	nd	nd

Table 3.12: Concentrations of targeted alkaloids in *L. deserti phill* plant parts

Plant part	Concentration mg/g			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	nd	nd	nd
Leaves	nd	nd	0.02855	nd
Fruits	nd	nd	nd	0.01346
Roots	nd	nd	nd	nd

Table 3.13: Concentrations of targeted alkaloids in *N. resteca* plant parts

Plant part	Concentration mg/g			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	nd	0.01820	nd
Leaves	nd	nd	0.04450	nd
Fruits	nd	nd	0.01816	0.01807
Roots	nd	nd	0.01752	nd

Table 3.14: Concentrations of targeted alkaloids in *S. dubium* plant parts

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	nd	0.001271	0.04521
Leaves	nd	nd	0.01890	nd
Fruits	nd	nd	nd	0.07839
Roots	nd	nd	nd	0.01940

Table 3.15: Concentrations of targeted alkaloids in *S. incanum* plant parts

Plant part	Peak height			
	Atropine	Hyoscine	Nicotine	Solanine
Stem	nd	nd	nd	0.03321
Leaves	nd	nd	nd	0.01885
Fruits	nd	nd	nd	0.06105
Roots	nd	nd	nd	nd

- nd : not detected

3.4 Conclusion and Recommendations

3.3.1 Conclusion:

Datura stramonium and *Datura innoxia* are considered as one of the main sources of Atropine; they also contain Hyoscine in significant concentrations (Brust, John 2004) while *Hyosyamus spp.* Considered in the main sources of Hyouscine (Cattell, 1910) the conclude that:

- the specimens of *D. stramonium*, *D. innoxia* which were analyzed can be utilized as a good row material for medical and chemical industries considering the concentration of Atropine and Hyoscine and regarding to less concentrations of other alkaloids like Solanine and Nicotine, The same conclusion applies to specimens of *Hyosyamus albus*, they was also in less concentrations of other alkaloids and can be widely utilized.
- *Lycium deserti phill* specimens were clear of toxic addictive alkaloids, they were contain Solanine and Nicotine in non significant concentrations for medical industry utilization, while it can be utilized for chemical, food and livestock feeding industries.
- *Nictiana rusica* specimenswere very ritch in Nicotin which can make it a good material for medical and chemical industries although it is free of toxic addictive alkaloids.
- *Solanum dubium and Solanum incanum* specimens contained significant concentrations of solanine they were free of toxic addictive alkaloids, S. dubuim spciments contained Very close relative compound to the tropan alkaloids, This suggests from the point of view of the plant chemotaxonomy that it is very close to the *Datura spp.* and *Hyosyamus spp*. Plants. *Solanum spp*. can be utilized in food industry or livestock feeding or any other traditional uses.

- According to the constituent of Atropine and Hyoscine in *Datura spp.* and *Hyosyamus spp.* Specimens involved in this study, it can be concluded that they are relative in plant chemotaxonomy.
- According to the constituent of Nicotine and Solanine of all plants specimens in this study, it can be concluded that all those species are relative in chemotaxonomy.

3.3.2 Recommendations:

1. Conducting a field survey of the location of the *Datura* and *Hyosyamus* plants and raising the awareness of the public about the dangers they may pose to them.
2. Conducting more scientific researches on the targeted plant in this study to determine the constituent of any other alkaloids, and clarify the benefits and disadvantages.
3. To Direct industrial capital to benefit from these land resources and to carry out more scientific and economic studies

References:

Adel al-Hemiri and Wasan O. Noori (2009) Extraction of atropine from Datura Innoxia using liquid membrane Technique. Iraqi Journal of Chemical and Petroleum Engineering 10(1):23-27

Aditya V .Sakhare(2012),Isolation Of Solanine From Potato Leaves and Evaluation Of Its Antimicrobial Activity , International Journal of Science and Research. 3(11):2052-2056

Alaghemand A.,Mansour Ghorbanpour , Moghaddasian B. (2013) Determination of atropine, hyoscine and rutin content of henbane seeds from different regions in Iran. Advances in Environmental Biology 7(4):614-618.

Ali Esmail Al . Snafi (2018)Therapeutic Importance of Hyoscyamus Species grown in Irag (Hyoscyamus alpus , Hyoscyamus niger and Hyscyamus reticulates) . Journal of Pharmacy 8(6) PP 18-32

Alireza Iranbakhsh, Mohammad Ali Oshaghi, Ahmad Majd. (2005) Distribution of atropine and scopolamine in different organs and stages of development in Datura stramonium L. (*Solanaceae*).structure and ultrastructure of biosynthesizing cells. acta biologica cracoviensia series botanica 48(1): 13–18.

Annapoorani, S. Grace (2013). An Eco-Friendly Antimicrobial Finish Using Datura Innoxia and Leucas Aspera on Cotton Fabric". International Journal of Scientific Research (IJSR). 2(4):199-201.

Anthony C. Moffat, M. David Osselton, Brian Winddop and Jo Watts (2005): Clarke’s Analysis of Drugs and Poison. Farmaceutical press,London, UK pp 657: 658, 1122, 1333:1335

Arie Febrianto Mulyadi, Susinggih Wijana, Arif Setyo Wahyudi (2013) Optimization of Nicotine Extraction In Tobacco Leaf (Nicotianatabacum L.) :(Study : Comparison of Ether and Petroleum Ether. The International Conference on Chemical Engineering UNPAR 2013, available online from: https://www.academia. edu/5448374/ Optimization_ of_Nicotine_Extraction_In_Tobacco_Leaf_Nicotiana_tabacum_L._Study_ Comparison_of_Ether_and_Petroleum_Ether_

Asture K, A. V. W adodkar S.G.(2008) Pharmaceutical chemistry 1 ,Pregati Books pvt . Ltd.,pp215.

Babiker , F , Jamal ,P, MirghaniM.E.S, Asari , A. H(2017) Characterization , Purification and Identification of some Alkaloids in Datura Stramonium . International Food Research Journal 24 (Suppl): S540-S543.

Bajanowski T, Brinkmann B, Mitchell EA, Vennemann MM, Leukel HW, Larsch KP, Beike J; GeSID Group (2007) Nicotine and cotinine in infants dying from sudden infant death syndrome.Int J Legal Med 122(1):23-8.

Barri, M. E. S. ; Onsa, T. O. ; Elawad, A. A. ; Elsayed, N. Y. ; Wasfi, I. A. ; Bari, E. M. A. ; Adam, S. E. I., (1983). Toxicity of five Sudanese plants to young ruminants. J. Comp. Pathol., 93 (4): 559-575.

Brust, John C. M. (2004). Neurological aspects of substance abuse (2 ed.). Philadelphia: Elsevier. p. 310.

Cattell, Henry Ware (1910). Lippincott. Oxford University publications p. 435

Desfosses, M. (1820) Extraitd'unelettre à M. Robiquet. In: J. de Pharmacie. Bd. 6, S. 374–376.

Eeva M , Salo JP, Oksman-Caldentey KM. (1998) Determination of the main tropane alkaloids from transformed Hyoscyamus muticus plants by capillary zone electrophoresis. J Pharm Biomed Anal.16(5):717-22.

Errol Zeiger (1998) α –Chaconine and α –Solanine , Review of Toxicological Literature. Chaconinesolanine, available online from: https://ntp.niehs.nih.gov/ntp/htdocs/chem_background/exsumpdf/chaconine solanine_508.pdf

Franck Lopes ,Costa ,Hervé Lhermitte (2014) Process for preparation of atropine. Data provided by IFI CLAIMS Patent Services, available online from:
https://patents.google.com/patent/WO2016016692A1/en?inventor=Franck+Lopes+COSTA

Friedman, Mendel (2006). "Potato Glycoalkaloids and Metabolites: Roles in the Plant and in the Diet". J. Agric. Food Chem. 54 (23): 8655–8681. doi:10.1021/jf061471t. PMID 17090106.

Fukuda T. (2001). Phylogeny and biogeography of the genus Lycium (Solanaceae): Inferences from chloroplast DNA sequences. Archived 2003-11-30 at the Wayback Machine. Molecular Phylogenetics and Evolution 19(2), 246-58.

Fukuhara, K. and Kubo, I. (1991).Isolation of steroidal glycoalkaloids from Solanum incanum by two countercurrent chromatographic methods. Phytochemistry 30(2): 685-687.

Garcia, Vicente F.; Olmstead, Richard G. (2003)."Phylogenetics of Tribe Anthocercideae (Solanaceae) Based on ndhF and trnL/F Sequence Data". Systematic Botany. 28 (3): 609–15.

Gennaro AR, Chairman, Chase GD, Der Marderosian A (1995) The science and practice of pharmacy. Remington 19 (5):1024

Gibson j., Bolt B., Garaza A. (2016) Atropine over view.Rxwiki, available on line from http://www.rxwiki.com/atropine.

Gournelis DC, Laskaris GG, Verpoorte R (1997) Cyclo-peptide alkaloids. Nat Prod Rep 14:75–82

Guillermo Beníteza , Martí March-Salasb , Alberto Villa-Kamelc , Ulises Cháves-Jiménezc , Javier Hernándezc , Nuria Montes-Osunad , Joaquín Moreno-Chocanoa , Paloma Cariñanos (2018) The genus Datura L. (Solanaceae) in Mexico and Spain – Ethnobotanical perspective at the interface of medical and illicit uses. Journal of Ethnopharmacology 133–151.

Hayes Wallace. (2008) Principles and Methods of Toxicology, informa healthcare Fifth Edition. (5) pp125.

Jan Alexander, Diane Benford, Andrew Cockburn, Jean-Pierre Cravedi, Eugenia Dogliotti, Alessandro Di Domenico, Maria Luisa Férnandez-Cruz, Peter Fürst, Johanna Fink-Gremmels, Corrado Lodovico Galli, Philippe Grandjean, Jadwiga Gzyl, Gerhard Heinemeyer, Niklas Johansson, Antonio Mutti, Josef Schlatter, Rolaf van Leeuwen, Carlos Van Peteghem and Philippe Verger. S (2008) Tropane alkaloids (from Datura sp.) as undesirable substances in animal feed. The EFSA Journal 691, 1-55.

Katherine M. Roberts. *Nicotiana sp*(2008).artsci.wustl.edu. available online from: https://artsci.wustl.edu/~gjfritz/Nicotiana_sp.html

Levin, achel A.; Mille, Jill S. (2005). "Relationships within tribe Lycieae (Solanaceae): Paraphyly of Lycium and multiple origins of gender dimorphism". American Journal of Botany. 92 (12): 2044–53

Ley, Willy (1965).The Healthfull Aromatic Herbe.For Your Information. Galaxy Science Fiction. pp. 88–98.

LibermanAnatoly , J. Lawrence Mitchell (2008). An Analytic Dictionary of English Etymology: An Introduction. University of Minnesota Press. pp. 108–110.

Luo Q. (2006). Lycium barbarum polysaccharides: Protective effects against heat-induced damage of rat testes and H2O2-induced DNA damage in mouse testicular cells and beneficial effect on sexual behavior and reproductive function of hemicastrated rats. Life Sciences 79(7), 613-21.

Manske R. H. F. (1965). The Alkaloids.Chemistry and Physiology. New York: Academic Press, 8: 673.

Matthews. V. (1994) The New Plantsman. Volume 1, Horticultural Royal Society.

McNaught A. D. and Wilkinson A. (1997).IUPAC. Compendium of Chemical Terminology, Blackwell Scientific Publications, Oxford 10:1351.

Michelle G. Carlin, John R. Dean, Jonathan L. Bookham and Justin J. B. Perry (2017) Investigation of the acid/base behaviour of the opium alkaloid thebaine in LC-ESI-MS mobile phase by NMR spectroscopy. Royal society open sceince 4(10)

Mishiba, Kei-Ichiro; Ando, Toshio; Mii, Masahiro; Watanabe, Hitoshi; Kokubun, Hisashi; Hashimoto, Goro; Marchesi, Eduardo (2000). "Nuclear DNA Content as an Index Character Discriminating Taxa in the Genus Petunia sensu Jussieu (Solanaceae)".Annals of Botany. 85 (5): 665-73.

Mishra Aseem , Pankaj C. , Sourav D. , Snita S. , Poonam J. , and Apurva G. (2015) Harmful effects of nicotine, Indian J Med Paediatr Oncol. 36(1): 24–31.

Neslihan TEK (2006) chromatographic determination of glycoalkaloids in eggplant. A Thesis Submitted to the Graduate School of Engineering and Sciences of Azmir Institute of Technology, in Partial Fulfillment of the Requirements for the Degree of Master Of Science in Chemistry. Available on line from http://library.iyte.edu.tr/tezler/master/kimya/T000566.pdf

Olmstead, R. G. (2008). A molecular phylogeny of the Solanaceae. Taxon 57:1159-1181

Olmstead, R. G.; Sweere, J. A.; Spangler, R. E.; Bohs, L.; Palmer, J. D. (1999). "Phylogeny and provisional classification of the Solanaceae based on chloroplast DNA"(PDF). Available online from: https://bohs.biology.utah.edu/PDFs/Lynn/Olmstead_et_al-1999.pdf

Olmstead, R.G.; Bohs, L. (2007). "A Summary of molecular systematic research in Solanaceae: 1982-2006". ActaHorticulturae. 745: 255–68.

Parry Ted. (2017) Active Pharmaceutical Ingredients (API's) in Australia One, Manufacturer One Manufacturer's View. Available on line from http://www.aoq.org.au/PDF/Plum.pdf

Pearn J, Thearle J (1982) The history of hyoscine Hist Sci Med. 17(Spec 2):257-61.

Pennachio , Marcello (2010) . Uses and Abuses of Plant . Derived Smoke : Its Ethnobotany As Hallucinogen , Perfume , Incense ,And Medicine. Oxford University Press .PP . 82_83 . ISBN 9780195370010 .

Pérez, Fernanda; Arroyo, Mary T. K.; Medel, Rodrigo; Hershkovitz, Mark A. (2006)."Ancestral reconstruction of flower morphology and pollination systems in Schizanthus (Solanaceae)". American Journal of Botany. 93 (7): 1029–38.

Plemenkov, VV (2001). Introduction to the Chemistry of Natural Compounds. Kazan. PP 223:2254

Preissel , Ulrike and Hans . Geory preissl (2002) . Brugmansia and Dature: Angel's Trumpets and Thorn Apples . Firefly Books .pp .124_125 http://books.google.com/books?id=FRIIAAAAYAAJ.

Quattrocchi, U. (2000). CRC World Dictionary of Plant Names.4 R-Z. USA: Taylor and Francis. p. 2058.

Raetsch, Ch. (2005). The encyclopedia of psychoactive plants: ethnopharmacology and its applications. US: Park Street Press. pp. 277–282.

Rathi M., Yadav R., Pednekar A. and Rewachandani Y . (2016) A detailed review on solanaceae family, European journal of pharmaceutical and medical research 3(1), 369-378

Raymond S. Sinatra; Jonathan S. Jahr; J. Michael Watkins-Pitchford (2010). The Essence of Analgesia and Analgesics.Cambridge University Press. pp. 82–90.

Richard J. Hamilton ; Nancy Anastasi Duffy, Daniel Stone, Anne Spencer, (2014). Tarascon pharmacopoeia (15 ed.). p386.

Robbers J, M Speedie, & V Tyler (1996) Pharmacognosy and pharmacobiotechnology. Williams and Wilkins, Baltimore. p. 1-14.

Rodgman, Alan; Perfetti, Thomas A. (2009). The chemical components of tobacco and tobacco smoke. Boca Raton, FL: CRC Press. ISBN 1-4200-7883-6. LCCN 2008018913

Sazima, M.; Buzato, S; Sazima, I (2003). "Dyssochroma viridiflorum (Solanaceae): A Reproductively Bat-dependent Epiphyte from the Atlantic Rainforest in Brazil". Annals of Botany. 92 (5): 725–30.

Sidra Siddiqui, Anam Khurshid, Sohaib Roomi, Fazeelat Karamat, Asrar Muhammad Khan, Humaira Shaheen and Tayyaba Yasmin (2017) Comparative analysis of hyoscine in wild-type and in vitrogrown Datura innoxia by high performance liquid chromatography. Tropical Journal of Pharmaceutical Research; 16 (7): 1683-1692

Smith, Stacey DeWitt; Baum, David A. (2006)."Phylogenetics of the florally diverse Andean clade Iochrominae (Solanaceae)". American Journal of Botany. 93(8): 1140–53.

Spiller G .A . (1997) caffeine 1st addition, CRC Press,

Stanfill, Stephen B.; Oliveira Da Silva, André Luiz; Lisko, Joseph G.; Lawler, Tameka S.; Kuklenyik, Peter; Tyx, Robert E.; Peuchen, Elizabeth H.; Richter, Patricia; Watson, Clifford H. (2015). "Comprehensive chemical characterization of Rapé tobacco products: Nicotine, un-ionized nicotine, tobacco-specific N′-nitrosamines, polycyclic aromatic hydrocarbons, and flavor constituents". Food and Chemical Toxicology. 82: 50–58.

Sweta, V.R., Lakshmi, T. (2015) Pharmacological profile of tropane alkaloids, Journal of Chemical and Pharmaceutical Research, 7 (5): 117 – 119.

Szymon Chowański, , Zbigniew Adamski, , Paweł Marciniak, Grzegorz Rosiński, Ender Büyükgüzel, Kemal Büyükgüzel, Patrizia Falabella, Laura Scrano,6 Emanuela Ventrella,5 Filomena Lelario, and Sabino A. Bufo (2016) . A Review of Bioinsecticidal Activity of Solanaceae Alkaloids, Toxins (Basel). 8(3): 6

Tadeusz Aniszewski (2015) Definition, typology, and occurrence of alkaloids, Alkaloids (Second Edition) Chemistry, Biology, Ecology, and Applications Pages 1-97.

Tarek Ismail Kakhia (2016) Alkaloids and Alkaloids plants, YUMPU available on line from https://www.yumpu.com/en/document/view/51581065/alkaloids-alkaloids-plants-tarek-ismail-kakhia

Thorpe JR. (2015) The 10 Grossest Cosmetics In History, Because Beauty Is Mule Urine? Availabl on line from https://www.bustle.com/ articles /64464-the-10-grossest-cosmetics-in-history-because-beauty-is-mule-urine

Ujvary istvan (1999) Nicotine and Other Insecticidal Alkaloids. Research gate available on line from https://www.researchgate.net/ publication /266505147_2_Nicotine_and_Other_Insecticidal_Alkaloids

United States Department of Agriculture.(2010) "Species Records of Hyoscyamus". Germplasm Resources Information Network.

USDA (United States Department of Agriculture). (2004) Phytochemical and Ethnobotanical Databases, agricultural research services. Available on line from https://web.archive.org/web/20041107214323/http://sun.ars-grin.gov:8080/npgspub/xsql/duke/chemdisp.xsql?chemical

Whitson, Maggie; Manos, Paul S. (2005). "Untangling Physalis (Solanaceae) from the Physaloids: A Two-Gene Phylogeny of the Physalinae" (Submitted manuscript). Systematic Botany. 30 (1): 216–30.