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Research Article
GC-MS Evidence Based Herbocure from Indian System of Medicine for Stomach Disorders in Vets

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Abstract

Background and Objective: Plant based medicines are the integral part of alternative medicines including traditional Indian and Chinese medicines. Especially the Siddha and Ayurveda medicines of Indian origin have documented about the use of various herbs in treating various ailments of both human and animals. **Materials and Methods:** Although these medicinal prescriptions have been adopted by traditional healers and practitioners of today, these medicines and formulations are viewed under scientific validation for their recognition and acceptance. Many pharmacological researches are now underway for the justification and use of these herbal formulations. The phytochemical investigations have been undertaken to enumerate the components of these herbs used in Siddha system of medicine. **Results:** The present study focuses in identifying the underlying components using gas chromatography-mass spectrometry (GC-MS) which could contribute the healing effects of four herbal parts namely, the rhizome *of Zingiber officinale* (*Z. officinale*) and *Curcuma longa,* the bark of *Terminalia arjuna* and the seed of *Piper longum* in treating the stomach disorders of vet animals. **Conclusion:** The medicinal uses cited in the literatures are correlated with the components identified by GC-MS as evidence to the herbo-cure of stomach disorders in vet animals.

Key words: Herbal remedies, abdominal disorders, *Curcuma longa*, *Zingiber officinale*, *Terminalia arjuna*, *Piper longum*

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Siddha system of medicine is the oldest documented medical system in the world. This system of medicine dates back to over 2000 years before Christ. Siddha is a medical system that is passed on from generation to generation through the ancient system of Guru-Shishya (teacher-student). It flourished among the Dravidians especially in the South Indian state of Tamil Nadu. A major portion of the Siddha medicines uses herbs and green leaved medicines.

Siddha medicines have developed a unique pattern of medicinal preparation in the name of the text Gunapadam[1]. Pathological methods are explained and documented in Noi naadal, physiological classifications are described in Udal tattuvam, Rejuvenative therapy are also incorporated in Kaya karpham medicines, immunomodulatory therapy are described in Yoga Asanas and Pranayamas, diagnostic methods are detailed in Nadi paritchai. These diagnostic treatment approaches are explained to the layman's cause. Pediatric treatment is explained in Balavagadam. It deals with the pathogenesis of fertilization and child development in fetus and the methods of developing a healthy child[2]. Touch the raphy explained in Varmam[3].

Siddha medicines not only explain about human problems it gives remedies for animals in the name of Vagadangal like Mattu vagadam (cattle literature), Aattu vagadam (sheep and goat literature) Patchi vagadam (birds literature) and Kozhi vagadam (Hen and Cock literature). Also it deals with plants diseases in Thavara vagadam[4]. Unfortunately, today it couldn't get those books. But some veterinary herbal practitioners (Vaidhyars) are found in all villages of Tamil Nadu who adopt these literatures for treating vet diseases. In this article, the herbal remedies for various veterinary ailments especially for abdominal disorders of vet animals are dealt scientifically.

Since earlier times, medicinal plants are used for the treatment of human diseases and nowadays they still make an important key for primary health care. It was observed that, in the last years, it has been an increasing interest by the large companies and some governments in such products, stimulating their standardization and the development of reliable quality control analytical methodologies to support their safety and efficacy[5,6]. The same concept was proven by Salgado *et al*.,[7] in research article of *Syzygium cumini*. The impotancy of herbals' quality controlling was confirmed in the *Tanacetum parthenium* plants hydroalcoholic extract[8].

Of late, the alternative system of medicine faces the challenges for proof and evidence for the medicinal properties exhibited by the formulations prescribed based on ancient siddha medical texts[9]. Encouragement to scientific research and education, laying down pharmacopoeial standards to ensure quality drugs, evolving good laboratory practices, following good manufacturing practices, have been the prime objectives promoted by AYUSH, Govt. of India. With this background, in this article, the justification and correlation for medicinal activities associated with the herbal medicines for vet diseases have been analysed based on the gas chromatography-mass spectrometry (GC-MS) results of important herbs meant for abdominal disorders.

MATERIALS AND METHODS

Traditional veterinary practitioners are also called by the name of Vaithyar. They are the sect of peoples who advise and assist all animals' diseases, pregnant cows for normal birth delivery and guide the post natal care to nurture the new born cow by safe and proper methods. Few of such traditional practitioners (Vaidyars) in a Tribal area in Sevapur, Kadavur panchayat, Karur district, Tamil Nadu, India were interviewed and the herbal practices adopted by them were systematically recorded based on their oral communication with us. Cattle rearing and livestock is the prime occupation of tribal people belonging to the community of 'Nackiyars' who live at the foot hills of Thoppaiya Swami malai located in the Eastern Ghats mountain ranges of Karur district, Tamil Nadu, India. Based on the Siddhars advocacy and their own personal rich experience, these medical secrets have been passed on from one generation to the other without any record and validation. Moreover, a special effort was made to collect these important herbs mentioned by them for treating stomach disorders of vet animals and a detailed GC-MS analysis was taken up with the following specifications.

GC-MS investigation methodology: The rhizome of *Z. officinale* and *Curcuma longa,* the bark of *Terminalia arjuna* and the seed of *Piper longum* were taken for the study. The tribal Vaithyar's of Sevapur used these plants parts for curing animals disorders of constipation, stomach bloating and diarrhoea. This same medicinal uses has mentioned in the Siddha textbook of Mooligai marmam by Mudaliar[10]. Samples were shade dried and cleaned with clean soft cloth. The samples were subjected to ethanolic extraction by blending the sample in a Pulverizer (Impact Pulveriser Model JEW 18 M.S. Body-Reg.) and 3 g of the blended sample was soaked in 15 mL ethanol and was kept in a Shaker (Femco Scientific Suppliers, Chennai) for 48 h. Then the sample was filtered using Whatman No. 1 filter paper and the filtrate was concentrated to 1 mL using hot plate.

A volume of 1 µL of clear extract of each sample was injected into GC-MS (Perkin Elmer Clarus 500) with a oven programming of 50°C (1 min), 10°C min^{-1} to 150°C (1 min), 8°C min^{-1} to 250°C (1 min) and 15°C min^{-1} to 300°C (5 min). The injector temperature was maintained at 280°C. The split ratio was set as 1:8. The carrier gas used in the analysis was helium which had the flow rate of 1 mL min^{-1}. A 30 m capillary column of elite 5 ms, with a column id of 250 µm was used. The compounds were detected in the range of 40-450 amu by matching with NIST library[11-13].The components of the oil were identified by comparison of their mass spectra with those of computer library.

The following treatments and prescriptions were recorded for treating various abdominal problems of vet animals especially for bovines.

Abdominal disorders and traditional herbo-cures

Constipation: One of the method is to take 100 g of *Ricinus communis* seed. Make paste, add water and give to the cow thrice a day. This same method has noted in the Siddha text book of Gunapadam by Murugesa[1]. The other method involves taking 200 g of *Cassia senna* leaves, making as paste, stirring it with 1 L of water and giving it once a day. The next remedy is to cut 3 banana fruits and to put in castor oil and giving to the affected cow. The fourth methodology is by taking 0.5 kg of *Abutilon indicum* leaves and making as a paste and giving the paste to the animal. The fifth approach is to take 300 g of *Pergularia daemia* root, make paste, give it to the animal. The sixth method is to mix *Aloe vera* gel and castor oil in 1:4 ratio andto boil this mixture till water is reduced and to cool and give 50 mL of this oil once a day. The seventh prescription involves taking 0.5 kg of *Aloe vera* root and adding 10 L of water, boiling the mixture until it is reduced to 2.5 L. Administer this mixture to animals. The eigth remedy is to be oil *Indigofera tinctoria* leaves. And to give this in the morning. The ninth methodology includes, taking 20 g of *Piper nigrum*, *Piper longum* and Z. *officinale* and making it as a powder, mix it with 2 L of drinking water and give once a day for 3 days. The last approach recorded is to make the paste of 100 g of *Cassytha filiformis* and administer to the animals.

Diarrhoea: Diarrhoea is a condition in which animals pass watery droppings (faeces) many times a day. The droppings are loose, runny and smelly and are a different colour from normal. Droppings can become dark green, dark brown or reddish black in colour because of blood in it. In some cases, e.g., rinderpest, the animal has diarrhoea which has a very bad smell. Totally there are eleven approaches suggested and adopted by the Vaidyars which have been recorded to treat diarrhoea and the list has been given as follows:

- Take 1 kg of *Wattakaka volubilis* leaves, boil the leaves and administer both the leaves and supernatant of the mixture to animals. Also noted in the Siddha text book of Gunapadam by Murugesa[1]
- Give *Coccinia indica* leaves in morning or give 0.5 L juice of the leaves
- Grind banana flower and give to the affected cow
- Make juice from flower of *Musa paradisiaca* and *Syzygium cumini*. Give this juice 1 L once a day
- Burn *Datura alba* fruit and make paste from the ash and give 50 g to the animals
- Make paste from mango seeds and bark of *syzygium cumini*. Give 0.5 kg of this paste to the affected animals
- Make paste of immatured fruits of *Punica granatum, Psidium guajava* and leaves of *Ficus racemosa*. Give 0.5 kg of this paste thrice a day
- Burn 50 g *Acorus calamus rhizome* and make the ash into paste and give to the animals
- Make paste of the 50 g of neem, guava and *Punica granatum* tender leaves. Gives this for thrice a day for every 4 h
- Take four parts of guava tender leaves and one part of dry ginger and make paste. Give twice a day
- Grind 1.5 kg of Ragi (*Paspalum scrobiculatum*) and give to the animals
- Take 50 g of baks of *Terminalia arjuna* and make it as a paste and give 3 times a day to the affected animal, the Siddha text book of Gunapadam by Murugesa[1]

Stomach bloating: Bloat is a medical condition in which the stomach becomes overstretched by excessive gas content. It is also commonly referred to as torsion, gastric torsion and gastric dilatation-volvulus (GDV) when the stomach is also twisted. The word bloat is often used as a general term to cover gas distension of the stomach with or without twisting. The name comes from the middle English blout, meaning soft or puffed, which is from the old Norse blautr, meaning soft or soaked[14].

A paste of betel leaves 20 g with 10 g of pepper and 100 g of palm jaggery is made and given thrice a day for 2 days. In the second method, 50 g of *Cissus quadrangularis* and *Cardiospermum helicacabum,* is taken and given in paste form. As a third approach, a paste of *Cassia fistula* tree is made and is given in coconut fruit size for four times a day for 2 days. *Acalypha fruiticosa* leaves are made into paste form and 300 g of this paste is given twice a day as a fourth method. Fifth treatment method is to make paste from tender leaves of

tamarind tree and is given in the size of coconut to the animals for twice a day. Sixth way is to give *Marsilea quadrifolia* leaves as animal feed. Seventh method is to take *Aloe vera* gel, mix with small quantity of salt and rice water and this rice water is administered to the animals. As a eighth technique, paste is made from 200 g of ginger, 25 g of betel leaves, 1/2 kg of palm jaggery, 10 g of pepper and 50 g of *Coleus aromaticus*. This paste is given twice a day. The nineth process is to mix 100 g of *Curcuma longa* powder with 2 L of drinking water and give once a day for 3 days.

Loss of appetite: The following methodologies can be employed with herbs to improve appetite The first one is to make paste with *Cissus quadrangularis* 250 g, turmeric 100 g and salt 30 g. This paste is given twice a day. In the next method, 0.5 kg of *Aristolochia indica* is collected and given as animal feed. In the third method, powder is made from 100 g pepper, 20 asafetida and 50 g dry ginger. Paste of Make betel leaves 20 g, country hen's eggs 2, 200 mL of gingelly oil is made. This paste is mixedwith the above mentioned powder and this paste is given to animals for 2 days.

As a fourth technique, a paste is made from 0.5 kg of *Cissus quadrangularis* and 100 g of salt. This paste is applied over the cow's tongue. In the fifth technique, 25 g each of *Randia dumetorum, Z. officinale, Carum copticum* seed, *Piper longum* and *Anithum graveolens* are taken and ground. Betal leaves juice is added with the powder and mixed well. This is given twice a day for 2 days. In the sixth process, a paste from *Gloriosa superba* rhizome 250 g, *Cissus quadrangularis* stem 250 g, onion 500 g, salt 100 is made and is orally administered to the animals.

RESULTS AND DISCUSSION

GC-MS investigation of ethanol extract of *Z. officinale*: The results of the GC-MS analyses on ethanol extracts of rhizome of *Z. officinale* is presented in Table 1. A total of 61 compounds were identified from the rhizome of ethanol extract of *Z. officinale*. The identified compounds were non volatile oil and recorded a % peak area of 19.406, 8.954 whose name has been identified as gingerol, (retention time 26.52, 31.34). Ketone groups followed the list with a % peak area of 6.143 and its name had been noted as 2-Butanone, 4-(4-Hydroxy-3-methoxyphenyl)-(retention time 17.21).

Gingerol has been reported to be the major constituent of ginger. Many literatures have quoted that gingerol a pungent principle from root of *Zingiber officinale* is responsible for most of it's medicinal properties[15]. Similarly, in the present study, gingerol peak has been recorded at 26.52, 26.97, 27.75, 28.98, 29.44, 31.34 and 31.78 retention times, estabilishing the fact that gingerol is the major constituent in ginger contributing to the medicinal properties reported in various literatures. Hence, in the present study, it can be inferred that as per the traditional healer's medicinal prescription for treating loss of appetite and constipation, ginger can be administered to the vet animals as it contains gingerol which has been reported to have the ability to relieve from fever, cold, nausea, respiratory disorders and above all indigestion problems. The other chemical constituents which make ginger as an effective medicine for treating loss of appetite and constipation in vet animals are viz., aldehyde groups such as octanal, alcohol groups like 6-Octen-1 ol, phenolic groups like phenol-2 methoxy, naphthalene methanol, naphthalene derivatives like longipinocarveol, Spiro[4.5]decan-7-one, terpenes like farnesene, bisabolene and alkaloids like piperine has also been observed and recorded in the present investigation. The compounds have been reported to have good medicinal implications such as antibacterial, antimicrobial and antifungal activities. Hence, it has been established without doubt that the use of ginger in the treating stomach disorders in vet animals which has been foretold by our traditional practitioners/healers has been proved to be true through the GC-MS investigation of composition of ginger and the literature analysis of the associated medicinal properties of various components identified in the GC-MS analysis. *Z. officinale* also exhibits its antidiarrhoeal activity by affecting bacterial and host cell metabolism[16]. The antimicrobial activities of *Z. officinale* extracts have been reported previously[17-20].The chromatogram of *Zingiber officinale* is presented below (Fig. 1).

GC-MS investigation of ethanol extract of *Terminalia arjuna*: The results of the GC-MS analyses on ethanol extracts of bark of *Terminalia arjuna* is presented in Table 2. A total of 19 compounds were identified from the bark of ethanol extract of *Terminalia arjuna*. The identified compounds were alcohol and recorded a % peak area of 23.9967, whose name has been identified as 2-Furan methanol, (retention time 2.80), non volatile oil group ranked next with a % peak area of 9700339 and the peak had been identified as cyclopropyl carbinol (retention time 6.6). Ketone groups followed the list with a % peak area of 4670936 and its name had been noted as 2-Butanone, 4-(4-Hydroxy-3-methoxyphenyl)-(retention time 17.21).

From the above table, it can be inferred that Furan methanol which has been recorded to be the major constituent in *Terminalia* tree, has been reported to have

Table 1: Compounds detected in ethanolic extract of *Zingiber officinalis* through GC-MS

Peak names	Retention time	Peak area (%)	Medicinal property
Decanal ($C_{10}H_{20}O$)	8.72	1.059	Fragrances and flavoring of foods, antibacterial
Benzene,1-(1,5-dimethyl-4-hexenyl)-4-methy-($C_{15}H_{22}$)	14.23	1.842	Antibacterial
1,3-Cyclohexadiene, 5-(1,5-dimethyl-4-hexeny-($C_{15}H_{24}$)	14.47	3.802	Antitumour
α-Farnesene ($C_{15}H_{24}$)	14.54	1.045	Antimicrobial, antioxidant
Cyclohexene ($C_{15}H_{24}$)	14.71	1.201	Antitumour
1,3-Cyclohexadiene ($C_{15}H_{24}$)	15.03	2.994	Antitumour
1,6,10-Dodecatrien-3-ol, 3,7, 11-Trimethy-($C_{15}H_{26}O$)	15.61	1.217	Antimicrobial activity
2-Butanone,4(4-hydroxy-3-methoxyphenyl-($C_{11}H_{14}O_3$)	17.21	6.143	Anticarcinogenesis activity
6,10-Dodecadien-1-yn-3-ol, 3,7,11-trimethyl ($C_{15}H_{24}$)	18.04	1.323	Antimicrobial activity
Spiro[4.5]dec-6-en-8-one, 1,7-dimethyl ($C_{15}H_{24}O$)	19.84	1.404	Infectious diseases
Phthalic acid, butyl dodecyl ester ($C_{24}H_{38}O_4$)	21.89	1.617	Endocrine disruptor
10,13-Octadecadieynoic acid ($C_{19}H_{30}O_2$)	22.08	0.888	Antibacterial activity
Hexadecanoic acid, ethyl ester ($C_{18}H_{36}O_2$)	22.26	2.324	Reduce hypercholestrolaemic effect
Gingerol ($C_{17}H_{26}O_4$)	25.58	4.276	Relives migraine, nausea and vomiting
3-Decanone ($C_{17}H_{26}O_3$)	25.70	1.550	Antibacterial
Gingerol ($C_{17}H_{26}O_4$)	26.52	19.406	Relives migraine, nausea and vomiting
Gingerol ($C_{17}H_{26}O_4$)	26.97	1.890	Relives migraine, nausea and vomiting
Gingerol ($C_{17}H_{26}O_4$)	27.75	5.980	Against rheumatic pain
Gingerol ($C_{17}H_{26}O_4$)	28.98, 29.44	1.148	Act as a appetizer
Dimethyl-octa dimethoxy ($C_{18}H_{26}O_2$)	29.16	2.379	Antibacterial
6-(1-Hydroxymethylvinyl)-4,8a-dimethyl-hexahydro-1H-naphthalene ($C_{15}H_{22}O_2$)	30.09	2.912	Anticancer activity
Piperine ($C_{17}H_{19}NO_3$)	30.30	1.142	Stimulates skin shine with U-V light
Piperine ($C_{17}H_{19}NO_3$) MW: 285	30.46	1.633	Increase appetite
Piperine ($C_{17}H_{19}NO_3$) MW: 285	30.66, 33.93	3.361	Cures respiratory problems
Gingerol ($C_{17}H_{26}O_4$)	31.34,	8.954	Cures fever, cold cough
Gingerol ($C_{17}H_{26}O_4$)	31.78	2.550	Cures respiratory problems
Piperidine ($C_{23}H_{43}NO$)	32.30	2.126	Cure all digestive problems
(2,6,6-Trimethylcyclohex-1-enylmethanesulfonyl)benzene ($C_{16}H_{22}O_2S$)	35.96	2.456	Antibacterial

Table 2: Compounds detected in ethanolic extract of *Terminalia arjuna* through GC-MS

Peak names	Retention time	Peak area (%)	Medicinal property
Octodrine ($C_8H_{19}N$)	2.58	3.5779	Antitumour
2-Furanmethanol (Furfuryl alcohol) ($C_5H_6O_2$)	2.80	23.9967	Biocide/fungicide, Inhibits bacterial growth
Propenamide,cyclo hexyl($C_{11}H_{19}NO$)	3.63	8.7564	Enzyme inhibitors
Dideoxyribonolactone ($C_5H_8O_3$)	5.80	10.2792	Anticancer
Cyclopropyl carbinol (C_4H_8O)	6.57	1.3547	Biomedicine, flavor, skin care and cosmetic
Cyclopropyl carbinol (C_4H_8O)	6.67	11.5550	Skin care and cosmetic, and bioenergy.
Cyclohexanamine ($C_{11}H_{21}N$)	6.89	1.1860	Fungicides and insecticides.
Pyrimidine-4,6-diol, 5-methyl ($C_5H_6N_2O_2$)	6.97	1.9539	Antineoplastic agents
2(3H)-Furanone, dihydro-4-hydroxy ($C_4H_6O_3$)	7.86	6.2304	Acute and chronic renal failure, hypertension, myocardial infarction
2-Formyl-9-ribofuranosyl hypoxanthine ($C_{11}H_{12}N_4O_6$)	13.35	2.4964	Anticancer and antiviral
1,6-Anhydro-á-D-gluco pyranose ($C_6H_{10}O_5$)	14.54	5.6886	Anti tumors or anti inflammatory
Hexadecanoicacid,15-methyl methyl ester ($C_{18}H_{36}O_2$)	21.26	8.2013	Hypercholestremic effect
7-Hexadecenoic acid ($C_{17}H_{32}O_2$)	23.71	7.3486	Control fat oxidation
Oleic acid ($C_{18}H_{34}O_2$)	23.78	4.3401	Emulsifying and Hypotensive

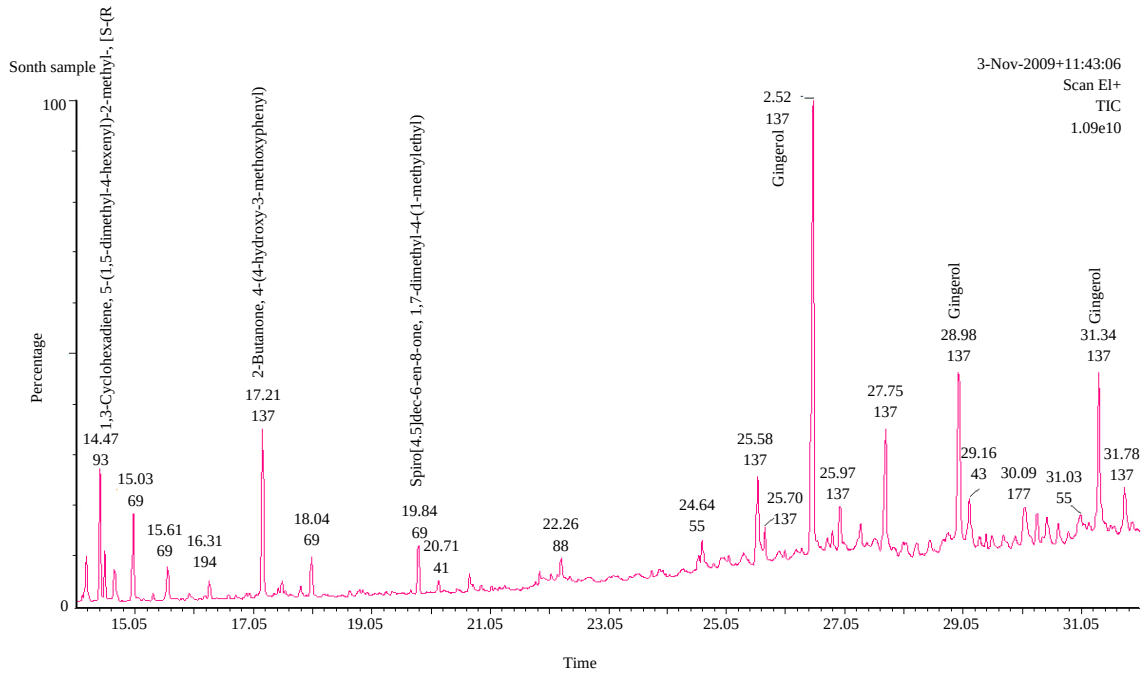

Fig. 1: GC-MS chromatogram-phytochemicals in ethanolic extract of *Z. officinale*

biocide actitivity[21]. Simialrly, the other compounds such as propanamide which belongs to the class of amides has been identified in the present study which has the reported property of enzyme inhibition. Propanediamine and cyclohexanamine which belongs to amine group have been reported to exhibit the property of enzyme inhibition as well as fungicide activity. Antifungal activity has been reported in Imidazole compound which is a aromatic heterocyclic alkaloid[22]. This compound has been identified at 17.48 and 18.21 retention times in the present GC-MS study. As the compounds identified in this experiment have already been reported to have antifungal and biocidal activity, it is not a wonder that the ancient traditional healers have prescribed to use the bark of *Terminalia arjuna* in treating against the diarrhoea of vet animals. *Terminalia* may also be protective against gastric ulcers, such as those caused by non steroidal anti inflammatory drugs (NSAIDs)[23]. The chromatogram representing the GC-MS analysis of *Terminalia arjuna* along with the structure of amine and amide group compounds is presented below (Fig. 2).

GC-MS investigation of ethanol extract of *Curcuma longa:*

The results of the GC-MS analyses on ethanol extracts of rhyzome of *Curcuma longa* is presented in Table 3. Ethanol extracts bark of *Curcuma longa* a total of 18 compounds were identified from the bark of ethanol extract of *Curcuma longa*. The identified compounds were aromatic substance and recorded a % peak area of 33.4108, whose name has been identified as 2-Methoxy-4-vinylphenol, (retention time 11.06), methoxy phenyl group ranked next with a % peak area of 30.6932 and the peak had been identified as 3-Buten-2-one, 4-(4-hydroxy-3-methoxyphenyl) (retention time19.92) terpene groups followed the list with a % peak area of 14.4354 and its name had been noted as squalene (retention time 31.99).

The Table indicates that the components such as Tumerone, α-tumerone and curlone which have been recorded at the respective retention times of 17.44, 17.52 and 18.09 have proved to have anti-inflammatory activity[24]. These are characteristic compounds of *Curcuma longa* which has been prescribed to heal stomach bloating of animals by our antique healers[25]. The other compounds such as methyltetradeconate, 14-Octadecanal and 1,2 benzenedicarboxylic acid also have synergistic effect in curing stomach boating in vet animals due to their reported antifungal, antimicrobial and antioxidant activities. Current traditional Indian medicine claims the use of its powder against biliary disorders, anorexia, coryza, cough, diabetic wounds, hepatic disorder, rheumatism and sinusitis[26]. In old hindu medicine, it is extensively used for the treatment of sprains and swellings caused by injury[27]. The traditional medicine in China uses *C. longa* L. in diseases, which are associated with abdominal pains. Religious ceremonies still

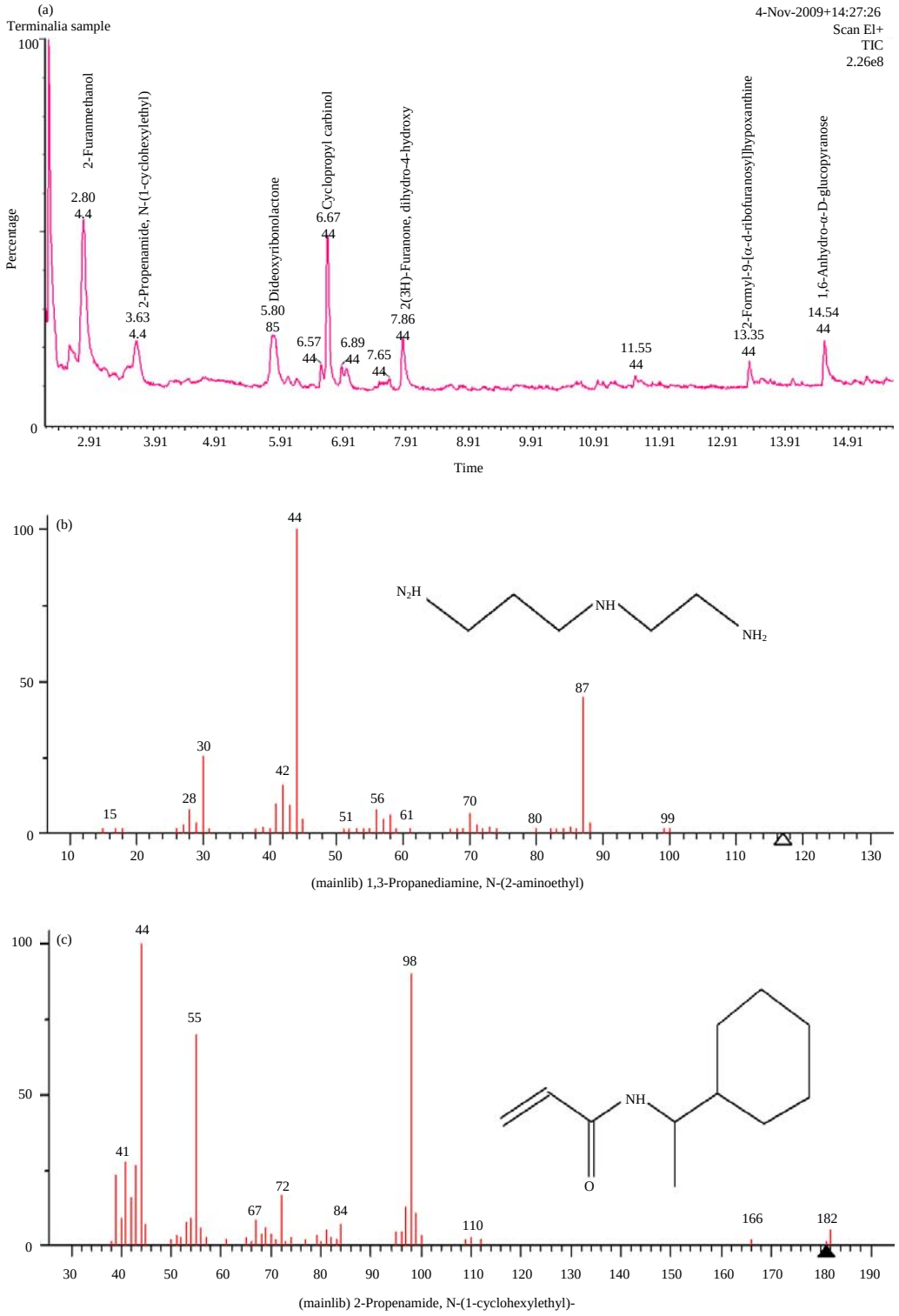

Fig. 2(a-c): GC-MS chromatogram profile phytochemicals in ethanolic extract of *T. arjuna*

use turmeric in many forms[28]. The structures of α-tumerone, curlone and the respective chromatogram of *Curcuma longa* is presented (Fig. 3).

GC-MS investigation of ethanol extract of seeds of *Piper longam*: The results of the GC-MS analyses on ethanol extracts of dry fruit of *Piper longam* is presented in Table 4. A total of

Table 3: Compounds detected in ethanolic extract of *Curcuma longa* through GC-MS

Peak names	Retention time	Peak area (%)	Medicinal property
Phenol,2-methoxy ($C_7H_8O_2$)	6.50	8.4660	Cosmetics, sunscreen, hair dyes & skin loton
Maltol ($C_6H_6O_3$)	6.92	4.6481	Use as a fragrant
2-Methoxy-4-vinyl phenol ($C_9H_{10}O_2$)	11.06	33.4108	Chemical signaling (pheromones)
α-tumerone ($C_{15}H_{20}O$)	17.44	1.0504	Anti-inflamatory
3-Butenhydroxy-3-Dehydro zingerone ($C_{11}H_{12}O_3$)	19.92	30.6932	Act as a antiaging
Hexadecanoic acid, Palmitic acid, methyl ester ($C_{17}H_{34}O_{2x}$)	21.26	1.3305	Coronary heart diseases and act as a anti diabetic drug
Benzenedicarboxylic acid, diisooctyl ester ($C_{24}H_{38}O_4$)	29.04	1.4048	Antimicrobial activity and hypoglycaemic activity
Squalene ($C_{30}H_{50}$)	31.99	14.4354	Skin, joint inflamation with arthritis

Table 4: Compounds detected in ethanolic extract of *Piper longam* through GC-MS

Peak names	Retention time	Peak area (%)	Medicinal property
Benzenepropanoic acid hydro cinnamic acid, Rosmarinic acid ($C_9H_{10}O_2$)	11.73	2.3150	Anti-oxidant, anti-inflammatory
1H-Cyclopenta cyclopropa benzene ($C_{15}H_{24}$)	14.38	1.5539	Antineoplastic, anti-cancer and antitumor
8-Heptadecene ($C_{17}H_{34}$)	17.62	1.6572	Anticancer, antibacterial
Heptadecane ($C_{17}H_{36}$)	17.80	1.6445	Antiosteophorotic effect
Naphthalene ($C_{12}H_{22}$)	21.66	3.7793	Antimalarial activity
Pentadienoic acid, Piperic acid ($C_{12}H_{10}O_4$)	23.78	2.5053	Reducing the serum uric acid level
2H-Benzo[f]oxireno ($C_{23}H_{29}NO_5$)	30.65	5.0964	Cardiac arrhythmia, pulmonary vascular disease
Piperine ($C_{17}H_{19}NO_3$)	30.80	1.8652	Skin problem & Increase appetite
E,E,Z-1,3,12-Nonadecatriene-5,14-diol ($C_{19}H_{34}O_2$)	31.64	5.2172	Antioxidant
E,E,Z-1,3,12-Nonadecatriene-5,14-diol ($C_{19}H_{34}O_2$)	31.86	10.2189	Increase immune system
Piperine ($C_{17}H_{19}NO_3$)	32.02	3.3957	Bronchitis, Breathing problems
Piperine ($C_{17}H_{19}NO_3$)	34.48	49.4011	Stomach aches, respiratory problems

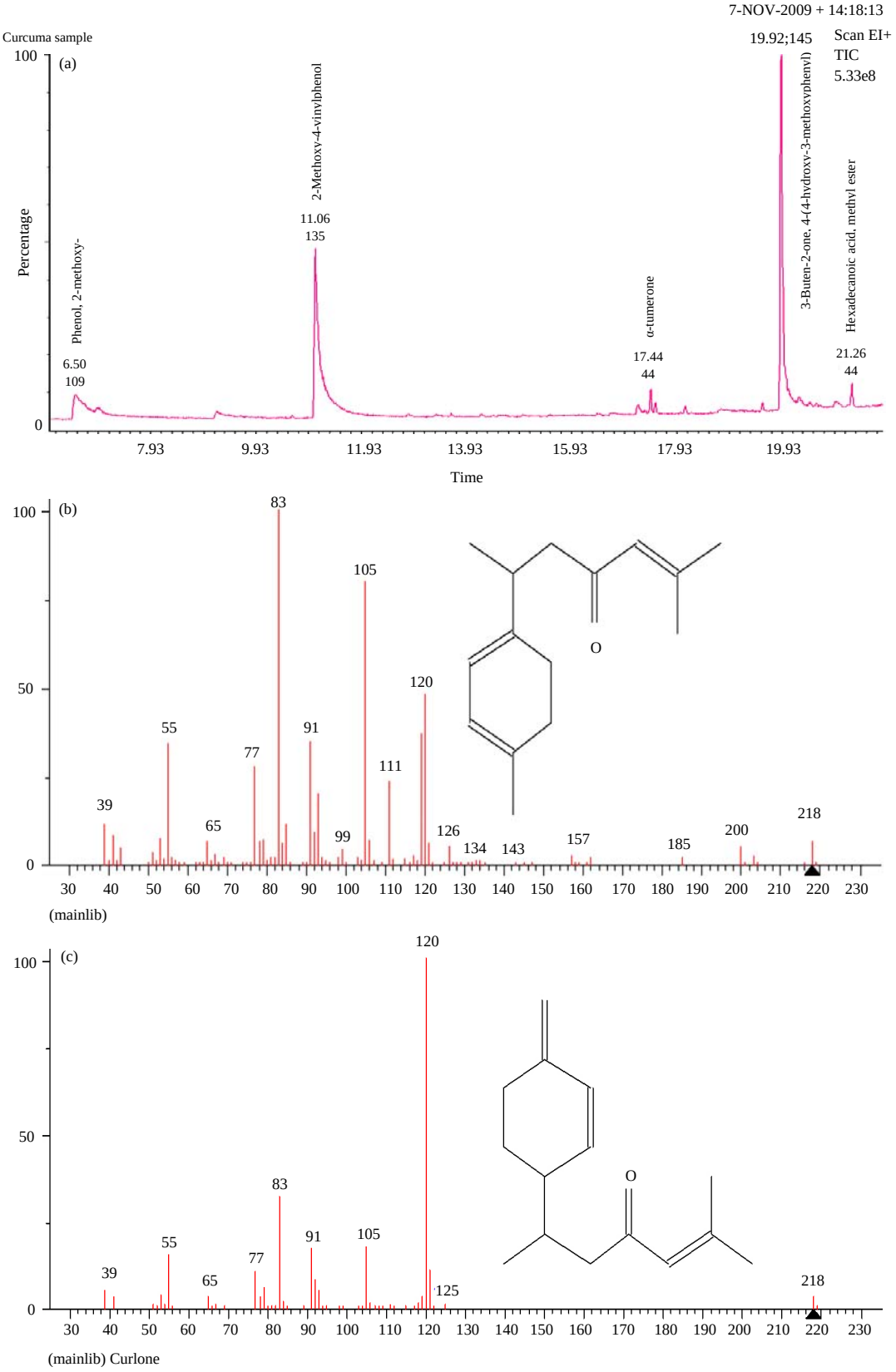

Fig. 3(a-c): GC-MS chromatogram-phytochemicals in ethanolic extract of *Curcuma longa*

59 compounds were identified from the dry fruit of ethanol extract of *Piper longam*. The identified compounds were alkaloid and recorded a % peak area of 49.4011, whose name has been identified as piperine (Retention time 34.48), alkene group ranked next with a % peak area of 10.2189, 5.2172 and the peak had been identified as E,E,Z-1,3,12-Nonadecatriene-5,14-diol (retention time 31.86, 31.64).

Long pepper's aromatic, slightly musty odor comes from the volatile oils found largely in the flesh and skin its pungent bite comes from the alkaloids piperine and piperidine-and resins found mostly in the seeds. Long pepper has as a carminitive effect, a property likely due to its beneficial effect of stimulating hydrochloric acid production. Long pepper alleviates hemorrhoids, gas and constipation, controls loss of appetite, improves digestion, promotes sweating, increase urination, anti-bacterial effect, anti-oxidant effect and stimulates the breakdown of fat cells[29].

Piperine is a piperidine derivative with multiple pharmacological activities. The traditional uses include analgesic, antipyretic, CNS depressant, anti-inflammatory, antioxidant, anticonvulsant, anti-bacterial, anti-tumor and hepatoprotective activities[30]. In the present study, piperidine alkaloid has been recorded to be having the high percentage of 49.04% and has been identified at the retention times of 30.80, 32.02, 34.48 and 37.29. Other than this piperidine the other compounds such as 1-Piperidineethanol, Naphthalene, 9-Nonadecene, 2-Propenoic acid, Naphthalene, 2-Thiopheneacetic acid, Dodecanoic acid, E,E,Z-1,3,12-Nonadecatriene and Eicosanenitrile contribute in alleviating the loss of appetite problems by their various medicinal properties which have been cited in Table 4. The chromatogram of *Piper longam* is presented in Fig. 4.

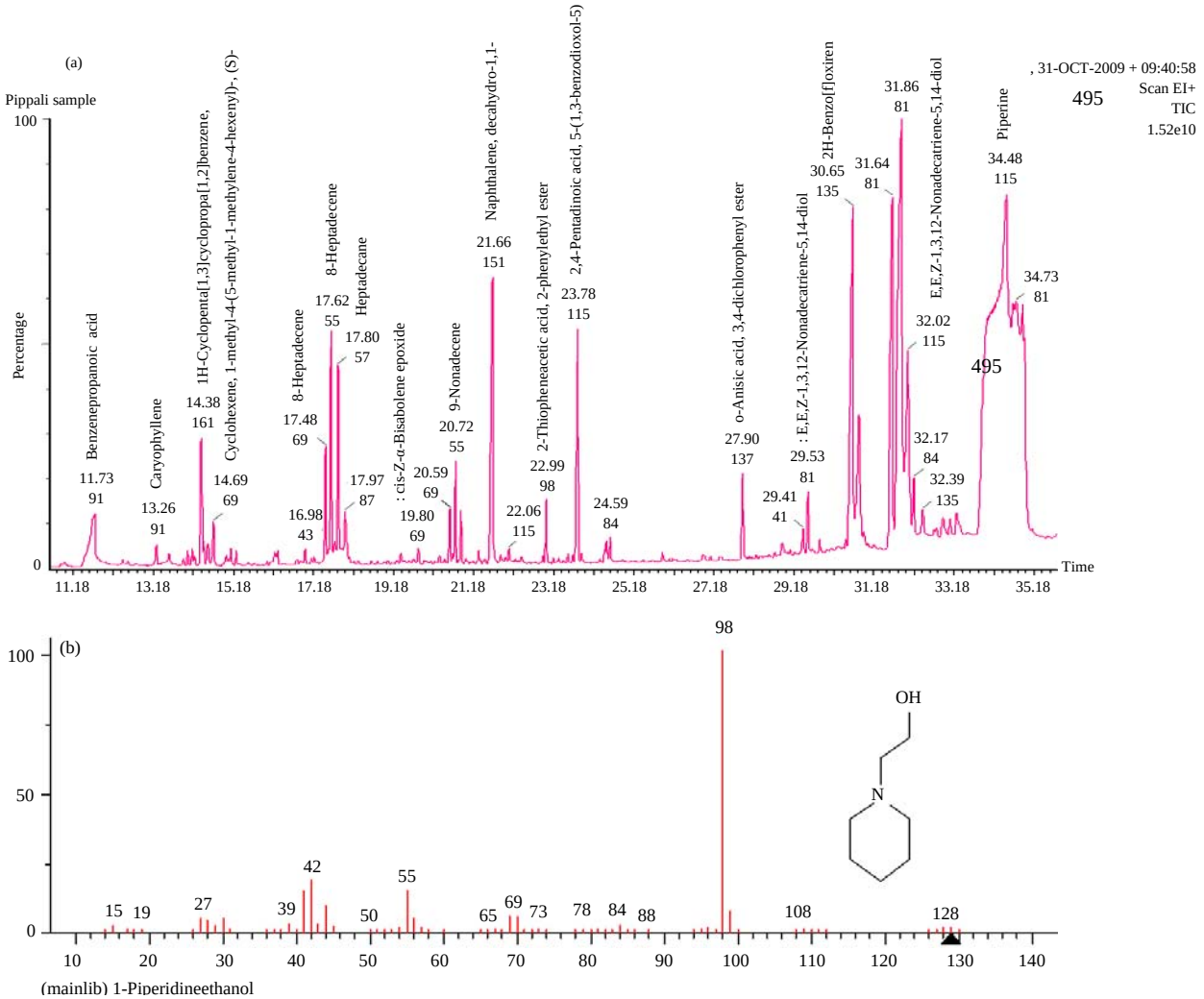

Fig. 4: Continue

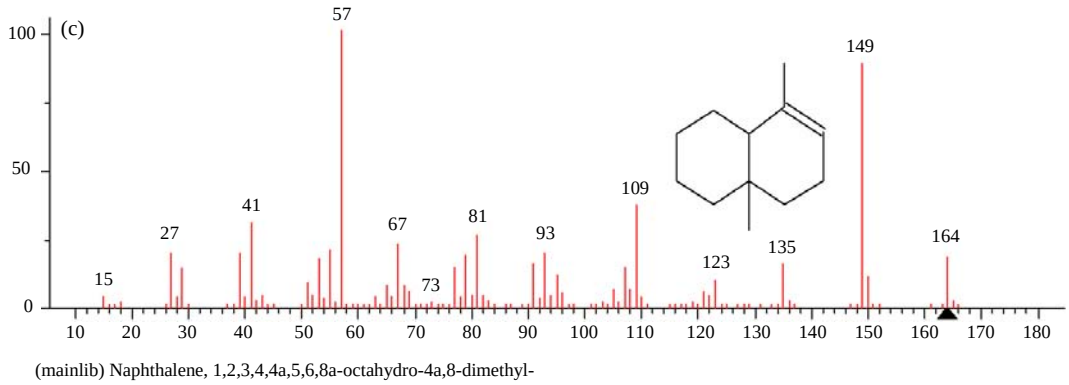

Fig. 4(a-c): GC-MS chromatogram-phytochemicals in ethanolic extract of *Piper longam*

CONCLUSION

The ancient healers and traditional medical practitioners have prescribed and practiced the application of herbal medicines in treating various ailments. In case of animals also, they had sound knowledge about the medicinal properties of various herbal parts. Hence they have formulated various preparations for treating the stomach disorders in animals. The present research of phytoanalysis of different compounds present in four well-known herbal parts namely: The rhizome *of Z. officinale* and *Curcuma longa,* the bark of *Terminalia arjuna* and the seed of *Piper longum* have clearly showed that a number of major and minor compounds present in these parts have contributed cumulatively in alleviating the stomach disorders namely constipation, diarrhoea, stomach bloating and loss of appetite in vet animals. GC-MS analysis also exhibited the major phytochemicals such as gingerol and zingiberin in the ethanolic extract, in *Terminalia* bark has furfuryl alcohol in main phytochemical, in *Curcuma* it has 2 Methoxy-4-vinyl phenol and long pepper posseses piperine phytochemical mainly. The identification and analysis of these compounds have been explained elaborately by using sophisticated instruments and have been correlated with their medicinal properties that are basic in curing the disorder as a whole. Thus the wisdom and knowledge in herbal medicine prescriptions of ancient healers stand to be valid.

SIGNIFICANCE STATEMENTS

This study discovers that the tribal Vaithyar's of Sevapur used these plants parts for curing animals disorders of constipation, stomach bloating and diarrhoea. This indigenous knowledge can be beneficial to the whole world. This study will help the researcher to take this traditional practicing methods to prove scientifically.

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