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A review on chemistry and biological activities of *Laurus nobilis* L. essential oil

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Abstract
Laurus nobilis L. commonly known as bay belonging to the family Lauraceae is one of the most useful essential oil and is an industrial plant used in foods, drugs and cosmetics. Bay is cultivated throughout the world mainly in tropical and sub-tropical Asia, Australia, Pacific region and South Asia. Bay essential oil can be extracted from various parts of plant and chiefly consisted of 1, 8 Cineole, sabinene, α-pinene and p-Cymene. Due to presence of various chemical constituents in bay, various biological and pharmacological properties have been reported such as antibacterial, antifungal, antioxidant, insecticidal and nematicidal activities. This review highlighted chemical composition and biological activities of *Laurus nobilis* which will be useful to the researcher for further study.

Keywords: *Laurus nobilis* (L.), essential oil, chemical composition, biological activity

1. Introduction
The natural plant products are chemical compounds extracted from plants which are synthesized by following pathways of primary or secondary metabolism. The study of natural products involves isolation of these compounds in a pure form by hydro-distillation, soxhlet extraction and chromatographic methods and analysis of their structure, formation, use, purpose, etc. in the living organisms. Essential oils are volatile secondary metabolites that are produced by plants for their own need other than for nutrition. Essential oils are also known as fragrant, volatile, ethereal and aromatic oils (Baser and Demirci 2007) [11]. By virtue of their volatile nature these can be easily diffused into air and therefore responsible for wonderful scents of plants. An essential oil may contain 20-60 aromatic compounds and this advanced mixture of compounds offers the essential oil, its characteristic fragrance and flavor (Arora 2015) [8]. Essential oils are liquid at room temperature, generally have density lower than that of water and are often colored. These are slightly soluble in water but are highly soluble in organic solvents. Although essential oils are only slightly soluble in water, the aqueous solubility of individual essential oil components varies with respect to polarity (magnetic activity).

Laurus nobilis L. is the member of family Lauraceae which comprises 32 genera and about 2,000-2,500 species. *Laurus* is also known as sweet bay, bay laurel, Grecian laurel, true bay and bay tree (Garg *et al*. 1992) [28]. Its natural habitat is the tropical and sub-tropical Himalayas at altitude of 900 to 2500 meters. It is also found in tropical and sub-tropical Asia, Australia, Pacific region and South Asia. In India it is found in Uttarakhand and Himachal Pradesh along the Western Himalaya and also in Sikkam, Assam, Mizoram and Meghalaya (Dighe *et al.* 2005) [22] and is cultivated in many warm regions of the world, particularly in Southern Europe and around the shores of the Mediterranean Sea (Lewis 1984)[41]. Turkey, Algeria, France, Greece, Morocco, Portugal, Spain, Belgium, Mexico, Central America and the Southern United States are the commercial production centres of bay. Turkey is one of the main producers and suppliers of bay leaves (Demir *et al.* 2004) [19].

2. Taxonomic, Distribution and traditional uses
Laurus nobilis L. is a small tree, having alternate, narrowly oblong-lanceolate leaves. The flowers are small and four lobed; the male has 8-12 stamens and female 2-4 staminodes. The ripe fruit is 10-15 mm, ovoid and black when ripe. The smooth bark may be olive green or reddish-blue. The plant is hardly multibranched, usually grows to a height of 20-30 feet in many warm regions of world (Said and Hussein 2014) [56]. The leaves are plucked and dried under shade for use as a flavouring material in a variety of culinary preparations, especially in French cuisine. The fragrant leaves are sold commercially as bay leaf, a seasoning (Anon 2005) [7].

Its aromatic bark is used occasionally for medicinal tea and its pulverized leaves for soup and condiments. Many of the evergreen laurels grown as hedges due to their handsome foliage, are used by florists.

3. Phytochemical analysis: The supercritical-carbondioxide (SC-CO_2) extract of bay leaf essential oil comprised monoterpenes and their oxygenated derivates (43.89%), together with sesquiterpenes (12.43%), diterpenes (1.33%) and esters (31.13%) (Ivanovic *et al.* 2010) [36].

3.1 Essential oil content: Essential oils are generally derived from one or more plant parts, such as flowers, leaves, stems, bark, wood, roots, seeds or fruits and the yield of the essential oil varies among different parts of the same plant. The amount of essential oil extracted from different plants ranges from 0.01 to 10%. The leaves of bay leaf collected from Lebanon yielded 35.15% 1,8- cineole while the essential oil obtained from young and old leaves collected from North Black Sea region of Turkey yielded 24.2 and 32.1%, respectively. The essential oils of the leaves and fruits from bay grown in Antakya, Yayladagi and Samandagi were isolated by solvent extraction and analysed by capillary gas chromatography (GC), gas chromatography and mass spectrometry (GC-MS). Although in both fruits and leaves the major component was found to be 1.8-cineole. The main components of *L. nolilis* reported were α-eudesmol, β-elemene and β-caryophyllene in flowers, (*E*)-β-ocimene and biclyclogermacrene in fruits, (*E*)-β-ocimene and germacrene D in buds (Kilic *et al.* 2004) [40]. The bay in plains and mountains gave the high rate of fruit oil and volatile leaf oil respectively. In the Mediterranean countries, monoterpene hydrocarbons and oxygenated monoterpenoids were present in the essential oil of the bay fruits (43.6 and 41.3%, respectively) while the leaf essential oil contained predominantly oxygenated monoterpenoids (66.2%). In Tunisia, studies showed essential oil composition of stems, leaves, buds and flowers of *L. nobilis* were between 0.4 and 1.1%. The component identified were 1,8-cineole, α-terpinyl acetate, methyl eugenol, eugenol and linalool (Marzouki *et al.* 2008) [44].

3.2 Chemical Composition: The chemical composition of bay leaf essential oil varied with the plant part from which it was extracted such as seed, leaf and flower. Some broad variations were seen in the relative amounts of the main components of the essential oils from different parts of bay, attributed to different geographic origins, genetic variability, growing conditions, organ development, seasonal variation, treatments prior to isolation and isolation procedures. The yield and composition of essential oil varies with genetic, environmental factors, developmental stage and by extraction methods like steam distillation, hydrodistillation and soxhlet extraction (Woolf 1999)[65]. Minor qualitative and major quantitative variation of some compounds of bay leaf essential oils occurs with respect to localities of collection (Table 1). Comparison of studies revealed that major compounds of *L. nobilis* were 1, 8-cineole (48.01, 31.78 and 17.64%), α-pinene (7.69, 11.69 and 17.96%), β- pinene (3.91, 6.91 and 9.51%), sabinene (2.93, 4.49 and 3.37%), limonene (1.43, 2.42 and 2.89%) and linalool (0.40, 0.24 and 0.24%) in the sea coast, the mountains and the plains respectively (Said and Hussein 2014) [56]. The bay leaf essential oil from different locations in South Turkey contained higher percentages for 1,8-cineole (46.6-59.9%) (Ozcan *et al.* 2010) [51]. The lowest concentration for 1, 8-cineole was reported from Portugal (27.2%) (Ramos *et al.* 2012) [52]. The concentration of 1, 8- cineole in the hydrodistilled oil from the fruits of *L. nobilis* was higher (29.8%) as compared to leaves from Lebanon (9.4%) and Turkey (9.5- 20. 5%). 1,8-cineole (59.94%) and Trans-β-osimen (28.35%) were found to be the major component of the leaves of essential oil collected from sea coast region of Samandagi. The fruits of *L. nobilis* harvested from Antakya and Yayladagi gave α-pinene, β-pinene, α-phellandrene, 1,8-cineole as major constituents (Sangun *et al.* 2007) [58]. The ethanolic extract of bay collected from Tavush region of Armenia and Zugdidi region of Georgia, analyzed by gas chromatography and mass spectrometry (GC-MS) showed 1,8-cineole in both extracts was an oxygenated monoterpene. Other predominant compounds of both extracts were α-thujene, α-pinene, β-pinene, D-limonene and o-cymene. Zugdidi region extract showed the presence of terpineol and β-phellandrene which were absent in Georgia extract (Vardapetyan *et al.* 2013) [64]. The components present in bay leaf essential oil were 1,8 cineole, Tricyclene, limonene, γ-Terpinene, Sabinene, α-Pinene, Eugenol, Linalool, p-Cymene, α-Phellandrene, Camphene, β-Pinene, Camphor, Terpinene-4-ol, α-Terpineol, α-Thujene, Myrcene, α-Terpinene, Terpineolene, Sabinol, Borneol, γ-Cadinene, β-Elemene, Germacrane A, Germacrane D-4-ol, α-Humulene (Fig 1).

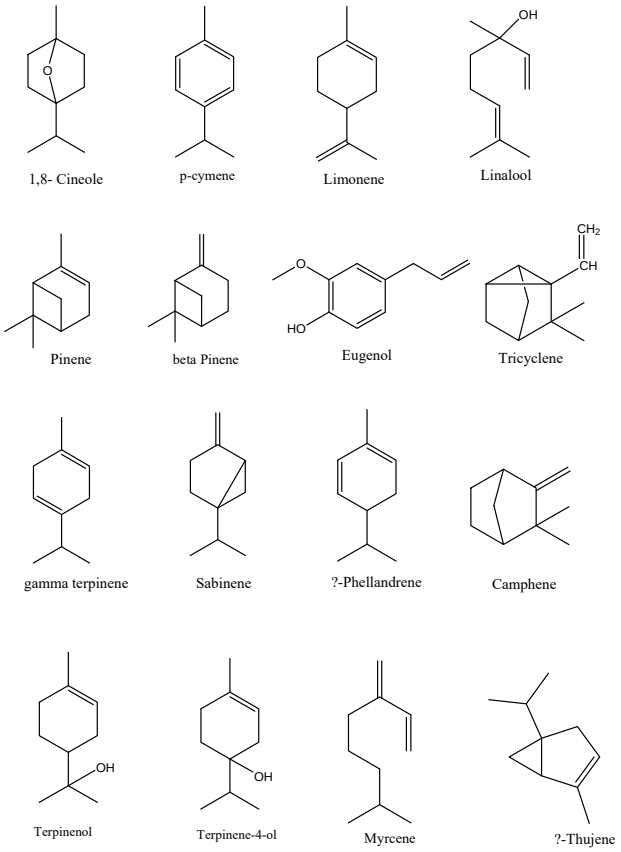

Fig 1: Structure of compounds present in bay leaf essential oil

Table 1: Variation in chemical composition of bay leaf essential oil with different geographic regions and different plant parts

	Location (Reference)														
	Kerman province (Maghtader and Salari 2012)		**Isfaha, Iran (Shokoohinia *et al.* 2014)**	**India (Choudhary *et al.* 2013)**	**Nepal (Choudhary *et al.* 2013)**	**Morocco (Derwich *et al.* 2009)**	**Lebanon (Said and Hussein 2014)**		**Antakya, Turkey (Sangun *et al.* 2007)**		**Yayladgi, Turkey (Sangun *et al.* 2007)**		**Samandagi, Turkey (Sangun *et al.* 2007)**		**South Caucasus (Vardapetyan *et al.* 2013)**
	Leaf	**Flower**	**Leaf**	**Leaf**	**Leaf**	**Leaf**	**Leaf**	**Fruit**	**Leaf**	**Fruit**	**Leaf**	**Fruit**	**Leaf**	**Fruit**	**Leaf**
Components	Concentration (%)														
1,8 Cineole	25.7	18.69	-	0.29	13.83	52.43	60.50	32.47	46.61	18.08	47.63	20.45	59.94	17.37	66.01
Tricyclene	2.27	-	0.29	-	-	-	-	-	-	-	-	-	-	6.82-	-
Limonene	3.47	2.47	-	-	2.83	5.25	2.00	1.68	-	-	-	-	-	-	-
γ-Terpinene	3.48	-	3.23	-	0.23	-	0.68	3.45	0.37	-	0.75	-	0.79	5.65	0.12
Sabinene	8.7	7.93	5.74	-	0.34	6.13	6.63	3.63	14.05	6.03	7.83	4.56	8.70	-	-
α-Pinene	5.25	7.38	5.17	1.39	5.23	3.72	3.72	12.45	3.66	16.55	2.19	11.31	2.61	-	6.92
Eugenol	1.69	2.33	2.47	-	0.22	0.56	-	-	-	-	0.65	-	-	-	-
Linalool	1.56	2.9	1.78	42.61	47.21	1.98	0.62	0.29	0.64	1.36	0.40	-	0.37	-	-
p-Cymene	0.31	0.31	-	1.09	3.40	0.94	1.57	0.87	-	-	-	-	-	-	-
α-Phellandrene	0.37	0.53	0.77	-	-	1.28	0.11	13.36	-	15.87	-	10.58	-	13.28	0.61
Camphene	3.86	2.46	5.23	0.40	4.44	0.05	0.16	1.67	0.19	2.08	-	0.80	-	0.81	0.28
β-Pinene	3.99	3.8	2.145	0.72	2.33	3.14	3.19	6.78	-	12.83	-	11.06	-	7.87	6.22
Camphor	-	-	4.20	-	24.07	-	-	-	-	-	-	-	-	-	-
Terpinene-4-ol	1.21	-	-	-	-	2.56	3.29	0.79	1.82	-	2.20	-	2.05	-	-
α-Terpineol	3.79	1.79	2.06	0.50	1.48	1.56	3.26	1.41	6.83	-	1.43	-	1.94	-	-
α-Thujene	0.38	1.97	-	-	0.39	0.21	-	-	0.36	-	-	0.38	-	0.35	15.70
Myrcene	1.68	1.65	0.62	-	0.8	-	0.25	1.14	0.62	0.92	-	0.54	-	0.64	0.53
α-Terpinene	2.12	-	0.74	-	-	2.12	-	-	-	-	-	-	-	-	-
Terpinolene	0.22	0.22	0.19	-	-	0.11	-	-	-	-	-	-	-	-	-
Sabinol	2.45	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Borneol	2.37	1.57	2.97	-	-	-	-	-	-	-	-	-	-	-	-
γ-Cadinene	2.68	0.68	-	-	-	-	-	-	-	-	-	-	-	-	-
β-Elemene	2.30	8.87	0.64	-	-	-	-	-	0.22	3.06	0.24	4.46	0.21	2.68	-
Germacrene-A	1.53	-	-	-	-	-	-	-	-	2.81	-	4.35	-	3.17	-
Germacrene D-4-ol	1.59	2.95	1.00	-	-	-	-	-	-	-	-	-	-	-	-
α-Humulene	2.19	1.43	0.51	-	-	-	8.58	3.75	-	-	-	-	-	-	

4. Biological Activities

Biological activity is defined as the inherent capacity of a substance to alter chemical or physiological functions of cell, tissue or organism. The genus laural comprises of more than 100 species which are distributed throughout temperate regions of Europe and Asia. Several species of this genus are being used in traditional medicines in Asian countries. Despite many medicinal uses, the plants from this family are very well known to contain bioactive compounds that are useful to develop plant based pesticides. The compounds isolated from it are known to cause toxic effect against many pests causing damage to agricultural commodities or crops. The details of the biological activities exhibited by different parts of *L. nobilis* are listed below:

4.1 Nematicidal

Plant parasitic nematodes are the most destructive group of plant pathogens worldwide and their control is extremely challenging. The root-knot nematodes, *Meloidogyne spp*, are one of the most economically damaging genera of plant parasitic nematodes on horticultural and field crops (Andres *et al.* 2012)[5]. The nematicidal effects of the essential oils on J_2 and eggs of *Meloidogyne javanica* at a concentration of 1000 µl/l were examined. *In vitro*, bay leaf essential oil immobilized more than 80% of J_2 after 2 days of incubation (Oka *et al.* 2000) [49]. Nematicidal activity of bay essential oil against *Meloidogyne incognita* was investigated in tomato and pepper. There were no significant differences between nematode inoculums level and essential oil concentration used. However, the plant extract treatments restrained nematode populations in both tomatoes and pepper host plants (Cetintas and Qadir 2014) [15].

4.2 Antioxidant

Ozcan *et al.* (2010) [51] determined the potential antioxidant activity of the essential oil and methanolic extract of seed oil from *L. nobilis* by employing DPPH free radical scavenging and β-carotene/linoleic acid test systems. In both test systems the essential oil and the methanolic extract of seed oil of *L. nobilis* exhibited antioxidant properties. The 50% (IC_{50}) inhibition activity of the essential oil on the free radical DPPH was determined as 94.65 mg/ml whereas IC_{50} value of methanolic extract of seed oil was found unstable. In the case of the linoleic acid system, oxidation of linoleic acid was inhibited by essential oil and methanolic extract of seed oil, which showed 64.28 and 88.76% inhibition, respectively. The inhibition value of the methanolic extract of seed oil was quite close to the synthetic antioxidant butylated hydroxytoulene (BHT), 92.46% inhibition. The extracts of cardamom, coriander seeds and dried bay leaves were prepared and iron(III) reduction, 1,1-diphenyl-2-picrylhydrazyl radical-scavenging, hydrogen peroxide, superoxide and nitric oxide radical scavenging, reducing power were assayed as antioxidant capacity. Bay leaves showed greater amount of phenols and high antioxidant activity as compared to cardamom and coriander extracts (Deepa *et al.* 2013) [18].

Al-Hashimi and Mahmood (2016) [3] determined the reducing power and antioxidant activity of alcoholic extracts of bay leaves. The rates of antioxidant activity and reducing power increases as the concentrate of bay leaves extract increased. The *in vitro* and *in vivo* antioxidant activities of different extracts of Laurel leaves were studied by Kaurinovic *et al.* (2010) [39]. The results indicated that ethyl acetate extract of bay leaves exhibited the largest free radical scavenging capacity in neutralization of DPPH, NO, $O_2^{\bullet-}$ and OH radicals. The *in vivo* effects were evaluated on some antioxidant systems (activities of GSHPx, LPx, Px, CAT and XOD, and GSH content) in the mice liver and blood-hemolysate after treatment with the examined laurel extracts, or in combination with carbon tetrachloride. On the basis of the results obtained it can be concluded that the examined extracts exhibited a certain protective effect, which is more pronounced on the liver than on blood hemolysate parameters and the ethyl acetate extract showed strongest protective effect.

The antioxidant potentials of ethanolic and aqueous extracts of *Hypericum perforatum, Ocimum basilicum* and *L. nobilis* leaves were evaluated by 2, 2-diphenyl-l-picrylhydrazyl radical (DPPH) assay. The lowest radical scavenging capacity (RSC) was reported in the aqueous extract of *L. nobilis* as compared to *H. perforatum* and *O. basilicum*. The ethanolic extracts of *L. nobilis* showed more DPPH radical scavenging action than their aqueous extracts (Rukhkyan *et al.* 2013) [55]. El *et al*. (2014) [23] obtained the laurel essential oil by using solvent-free microwave extraction (SFME) and hydrodistillation (HD) methods from *L. nobilis* leaves at 622 W (100%) and 249 W (40%) power levels and hydrodistillation inhibited oxidation generated by 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS) radical by 93.88, 94.13 and 92.06%, respectively. Trolox equivalent antioxidant capacities (TEAC) of essential oils were 0.18 mm/ml oil for SFME at 622 W, 1.36 mm/ml oil or SFME at 249 W and 2.40 mm/ml oil for hydrodistillation ($p < 0.05$). Essential oils of *L. nobilis* were extracted by SFME at 100 and 40% power levels and hydrodistillation inhibited linoleic acid peroxidation by 70.57, 63.53 and 89.18% respectively. Inhibition effects of laurel essential oils obtained by SFME at different power levels and hydrodistillation on DPPH radical cation oxidation were not significantly different. The strongest antioxidant activity against DPPH radical was found in the essential oil obtained by SFME at 100% power level.

Basak and Candan (2013) [10] showed that the DPPH, hydroxyl and superoxide radical as well as hydrogen peroxide scavenging activities of bay leaf essential oil were greater than the positive controls and the three main components of the oil when tested independently. The inhibition of lipid peroxidation by the oil occurred less frequently than with 1,8-cineole and R-(+)- limonene alone, but the effects were more pronounced than those seen with 1-(S)-*α*-pinene and the positive controls. Antioxidant combination effect was assessed by DPPH free radical scavenging method (Bag 2015) [9]. The bay leaf essential oil was screened for possible antioxidant activity by DPPH (2,2-iphenylpicrylhydrazyl) free radical-scavenging and the β-carotene/linoleic acid assay. Both of these *in vitro* methods showed that the essential oil was a less powerful reducing agent than the well-known synthetic antioxidants, butylated hydroxytoluene and ascorbic acid (Yilmaz *et al.* 2013) [66].

Lyophilized aqueous and ethanol extract of *L. nobilis* were evaluated for their antioxidant activity, reducing power, free radical scavenging, superoxide anion radical scavenging, hydrogen peroxide scavenging and metal chelating activities to determine the total antioxidant capacity of both extracts. Both extracts showed strong total antioxidant activity in linoleic acid emulsion. Concentrations of 20, 40, and 60 µg/ml showed 84.9, 95.7, 96.8, and 94.2, 97.7, and 98.6% inhibition of lipid peroxidation of linoleic acid emulsion, for water and ethanol extracts, respectively. On the other hand, 60 µg/ml of the standard antioxidants butylated hydroxyianisole (BHA), butylated hydroxytoluene (BHT), and α-tocopherol

exhibited 96.6, 99.1, and 76.9% inhibition of lipid peroxidation in linoleic acid emulsion, respectively. In addition, both the extracts were having effective reducing power, DPPH· free radical scavenging, superoxide anion radical scavenging, hydrogen peroxide scavenging and metal chelating activities at 20, 40, and 60 µg/ml. The total amount of phenolic compounds in each extract was determined as gallic acid equivalents (Gulcin *et al.* 2006) [31].

4.3 Insecticidal

Bay leaf essential oil was tested for its insecticidal activity against *Tribolium castaneum* at five different concentrations ranging from 4-12 mg/g. The polar fraction of bay leaf essential oil was found to be more active as insecticide as compared to non-polar fraction and essential oil. Moreover, the insecticidal potential was found to be both concentration and time dependent (Chahal *et al.* 2016) [16]. Jemaa *et al*. (2011) [38] reported the chemical composition and the repellent activity of *L. nobilis* essential oil against 7-10 days old adults of *Lasioderma serricorne*. The results revealed that repellent action was highly dependent upon oil concentration and exposure time and the repellent efficacy was found to be high for high doses and short exposure period. So, *L. nobilis* essential oil may have potential as a control agent against cigratte beetle. *L. nobilis* essential oil from Morocco showed better insecticidal activity as compared to Tunisian and Algerian oils with RD_{50} values 0.013, 0.036 and 0.033 µl/cm^2 for *Rhyzopertha dominica* whereas 0.045, 0.139 and 0.096 µl/cm^2 for *T. castaneum* (Jeama *et al.* 2012) [37].

Salehi *et al.* (2014) [57] reported the repellency effects of *L. nobilis* essential oil against adults of *Ephestia kuehniella* Zeller as bay leaf essential oil showed 82.4 percent repellency rate at highest tested concentration *i.e.* 2.00 µl/l. Repellency and toxicity of essential oil from *L. nobilis* against the rust-red flour beetle (*T. castaneum* Herbst) were reported by Andronikashvili and Reichmuth (2003) [7]. The toxicity of ethanol extracts on the large diamond back moth, *Plutella xylostella*, was 55% (Erturk *et al*. 2004) [34]. Essential oils from laurel were evaluated for fumigant toxicity against all developmental stages of the confused flour beetle (*Tribolium confusum*). The vapours of laurel essential oil were toxic to all the stages of *T. confusum* (Isikber *et al*. 2006) [35].

The bruchid, *Acanthoscelides obtectus*, is one of the most damaging pests of kidney beans (*Phaseolus vulgaris*) worldwide. However, aromatic plants from the families Lamiaceae, Lauraceae, Myrtaceae and Poaceae can protect *P. vulgaris* by a direct or delayed insecticidal effect, through increased adult mortality and inhibition of reproduction (both oviposition and adult emergence). The results suggested that lipid, as well as nonlipid allelochemicals, such as phenolics, or non protein amino acids or flavonoids may be involved in the toxicity of extracts of aromatic plants to *A. obtectus* (Regnault- Roger and Hamraoui, 1995; Mackeen *et al*. 1997) [53,43]. Tayoub *et al.* (2012) [61] investigated the fumigant toxicity of the bay and sage essential oils against larvae of *Trogoderma granarium* insect. Exposure to vapours of essential oil from bay laurel and sage for 48 h resulted in about 98 and 100% mortality of the larvae at a concentration of 60 and 90 µl/160 cm^3 air, respectively. Essential oils of bay laurel showed a higher lethal activity than that of sage.

4.4 Antimicrobial

Essential oils displayed antimicrobial activity against *Staphylococcus aureus* 6538P, *Escherichia coli* O157:H7 and *Salmonella typhimurium* NRRL E 4463. The inhibitory effect on *S. aureus* 6538P of laurel oil obtained from SFME using lower power level was found to be lower than that obtained from SFME at 100% power level and hydrodisttilation method ($p < 0.05$) (El *et al.* 2014) [23]. The antimicrobial activity of the essential oil was tested against a panel of food-spoiling bacteria and one yeast strain. The minimum inhibitory concentration values for food-spoiling bacteria and yeast strain that were sensitive to *L. nobilis* essential oil ranged from 125-2000 µg/ml. *E. coli* O157:H7, *Candida albicans* ATCC 16231, *Salmonella enteritidis* ATCC 13076 and *L. monocytogenes* ATCC 7644 had MIC values of 125, 250 and 500 g/ml, respectively and were most sensitive to *L. nobilis*. They showed the largest growth inhibition halos in the agar well diffusion assays (33.0, 26.0, 24.0 and 22.0 mm, respectively (Yilmaz *et al*. 2013) [66].

The antimicrobial activities of essential oil were determined by disc diffusion and minimum inhibitory concentration methods. Both seed oil and methanolic extract of seed oil did not show activity against Gram-negative bacteria except for *Haemophilus influenzae* but they exhibited remarkable antimicrobial activity against Grampositive bacteria. The methanolic extract of seed oil exhibited more effective antimicrobial activity compared to the seed oil. (Ozcan *et al.* 2010) [51]. Bouzouita *et al.* (2011) [13] reported that the high content of 1,8-cineole in the essential oil of *L. nobilis* L. contributed to its weak antimicrobial activity on two bacteria (*Lactobacillus plantarum* and *E. coli*). The antimicrobial activity of the bay leaf essential oil was evaluated *in vitro* using the disc diffusion and serial dilution methods against the 14 microorganisms. The disc diffusion method indicated that *Bacillus cereus, S. aureus* and *Bacillus subtilis* were the most sensitive Gram positive bacteria tested, but the Gram negative bacteria *E. coli* was insensitive to essential oil (El-Sawi *et al.* 2009) [24].

Methanolic extracts of bay leaf showed higher antimicrobial activity, except for *Aspergillus niger*, *Aspergillus fumigatus* and *Penicillium verrucosum*. The differences in bioactivity might be related to the higher phenolic compounds content (flavonols, flavones and even, total phenolic compounds) present in methanolic extracts (Dias *et al.* 2013) [21].

4.5 Antibacterial

Bay leaf extract was assayed for antibacterial activity by agar well diffusion and agar dilution methods in order to determine the zone diameter of inhibition compared with tetracycline zone diameter of inhibition as control. The extract showed antibacterial activity against *S. aureus.* The results indicated the antibacterial use of the bay extract for the treatment of *S. aureus* infection (Ghadiri *et al*. 2014) [30]. The antibacterial activity of the essential oil of bay on human pathogenic bacteria by disc diffusion method *via* average inhibition zone was studied against 9 bacteria strains such as three Gram positive bacteria: *S. aureus*, *Staphylococcus epidermidis* and *Streptococcus faecalis* and six Gram negative bacteria: *Pseudomonas aeroginosa, Shigella flexneri, Klebsiella pneuomoniae, Salmonella typhi, Serratia marcescens* and *E. coli* were studied. Effect of the essential oil of *L. nobilis* on bacteria tested was more than that of tetracycline antibiotic. The results showed that the essential oil of *L. nobilis* showed strong anti-bacterial effects (Moghtader and Farahmand 2013) [46].

Ouibrahim *et al.* (2013) [50] evaluated the antibacterial activity of essential oils of *L. nobilis* L., *Rosmarinus officinalis* L. and *Ocimum basilicum* L. against twenty bacterial strains: *Enterococcus faecalis ATCC 29212, S. aureus* ATCC 25923,

MRSA ATCC 31 *(Méthicilino), S. aureus, S. epidermidis, Enterococcus avium, E. coli* ATCC 25922*, Salmonella* OMA 04, *E. coli, Klebsiella oxytoca, K. pneumoniae, Proteus mirabilis, Enterobacter* sp., *Citrobacter freundii, Pseudomonas aeruginosa, Acinetobacter baumanii, S. marcescens, Salmonella* sp., *Shigella* sp. and *Providencia alcalifaciens.* The three oils showed good antibacterial activity against both Gram negative and Gram positive bacteria. Laurel oil is the most efficient, *Shigella* sp. showed the highest sensitivity to the three oils. Among the three oils, Laurel showed the lowest MIC against *E. faecalis* ATCC 29212, *Enterobacter* sp., *Shigella* sp., *S. aureus* and *S. Epermidis* (0.25%).

Al-Hadi (2011) [2] conducted *in vitro* antibacterial activity of extracts of peppermint and bay leaf against *S. aureus* and found that the peppermint extract exhibited more activity than bay leaf on *S.aureus* but both of them were found to have inhibitory effect against *S. aureus*. The minimum bactericidal concentration for peppermint and bay leaf extracts were 25 and 35%, respectively. The ethanolic extracts and aqueous extracts of bay were tested for antibacterial potential. All the ethanolic extracts expressed more antibacterial activity than aqueous extracts (Vardapetyan *et al.* 2014) [63]. Eleven ethanolic extracts including bay collected from various regions of Turkey and local markets were assayed for the *in vitro* antibacterial activity against 3 gram-positive (*B. subtilis, S. aureus and S. epidermidis*) and 2 gram-negative bacteria (*E. coli and P. aeruginosa*), using agar dilution methods. In addition, their possible toxicity to *C. albicans* and *A. niger* was determined, using both agar dilution and disc-diffusion methods. The minimum inhibition concentration (MIC) of the *L. nobilis* ethanolic extract was 5 mg/ml for all the microorganisms tested. *Lauras* extracts were more effective against bacteria than fungi (Erturk 2006) [26].

Lauras essential oil possessed very low antibacterial activity on *B. subtilis* M02, M06 (3 mm) and maximum activity against *E. coli* M18, M20 (8 mm) (Abu-Zaid *et al.* 2013) [1]. Bag (2015) [9] evaluated the antibacterial combination effect of bay leaf essential oil and found that activity was maximum against six important food-borne bacteria (*B. cereus, Listeria monocytogenes, Micrococcus luteus, S. aureus, E. coli* and *S. typhimurium*) using microbroth dilution, checker board titration and time-kill methods. Erdogrul (1999) [25] found that extract of *L. nobilis* inhibited the growth of some bacteria and fungi, particularly *Bacillus megaterium, Mycobacterium smegmatus*, *Yersinia enterocolitica, S. aureus* and *L. monocytogenes.* The high contents of eugenol, methyl eugenol and fatty acid together with other active components (Marzouki *et al.* 2008) [44] could contribute to its overall antibacterial activity (Ivanovic *et al.* 2010) [36].

Derwich *et al.* (2009) [20] reported that *S. aureus* was the most sensitive strain among the bacteria tested (*Staphylococcus intermedius* and *K. pneumoniae*) to *L. nobilis* oil in strongest inhibition zone of 13 mm. The essential oil of *Cymbopogon citratus* demonstrated bacterial activity at all concentrations and against all of the bacteria tested. The majority of essential oil compounds were geranial and neral. The major constituent of *T. vulgaris* and *L. nobilis* were 1,8-cineole and linalool*,* which presented lower antibacterial activity than 1,8-cineole. The Gram-negative bacteria demonstrated higher resistance to the use of the essential oils tested in this study. *E. coli* was the least sensitive and was inhibited only by the oils of *C. citratus* and *L. nobilis* (Millezi *et al*. 2012) [45]. Ethanolic extract of *L. nobilis* leaf exhibited highest antibacterial activity (22 mm and 0.5 mg per disc MIC) against *B. subtilis* and the highest antifungal towards *A. niger* (25 mm and 0.5 mg MIC). *L. nobilis* fruit extracts were the least active against bacteria and fungi (8-10 mm and 2.0-3.0 MICs) (Al-Hussaini and Mahasneh 2011) [4].

4.6 Antifungal

Hassiotis (2010) [34] investigated the influence of aromatic *L. nobilis* on the development of two mycorrhizal species, *Glomus deserticola* and *Glomus intraradices.* Both mycorrhizal fungi colonized successfully the host plants, positively influencing their growth. *G. deserticola* presented higher infection level than *G. intraradices*. Addition of *L. nobilis* oil into substrates resulted in mycorrhiza inhibition, and the level of inhibition was analogous with the amount of added essential oil. The fungi were benefited by the aromatic compounds up to 15 mg of essential oil per litre of soil. However 30 and 60 mg/l of essential oil were able to create significant inhibition in mycorrhiza development and to restrict the host growth.

Essential oils of *Thymus vulgaris* (thyme), *Cymbopogon citratus* (lemongrass) and *L. nobilis* (bay) were chemically quantified, and the MIC was determined on the bacteria *S. aureus* ATCC 25923, *E. coli* ATCC 25922*, L. monocytogenes* ATCC 19117, *S. enterica* S64, and *P. aeruginosa* ATCC 27853 (Millezi *et al*. 2012) [45]. The extracts of *L. nobilis* showed the highest antifungal activity against *A. niger* and *C. albicans* with inhibition zone diameters of 20-32 mm/15ml (Erturk 2006) [26]. Essential oil of bay leaf was tested *in vitro* against two foodborne fungi belonging to the dominant mycobiota of stored rice, *Fusarium culmorum* and *F. verticillioides*, The result showed that bay essential oil possessed great potential to control both fungal pathogens (Rosello *et al.* 2015) [54].

The antifungal effect of 20 essential oils against the most important moulds in terms of spoilage of bakery products (*Eurotium* spp., *Aspergillus* spp. and *Penicillium* spp.) was investigated at concentration in the range between 0 to 1,000 µg/ml. Antifungal activity was tested at different water activity and pH conditions, and the fungal growth was followed by measuring the colony diameter during the incubation period. Bay leaf essential oil was more effective at pH 5, losing their activity as pH increased (Guynot *et al.* 2005) [32]. The potential of bay leaf essential oils against species belonging to *Eurotium*, *Aspergillus* and *Penicillium* genus was demonstrated (Geeta and Reddy 1990; Guynot *et al*. 2003) [29, 33]. Biological assays showed that fungitoxicity against *Fusarium moniliforme* (*Gibberella fujikuroi*), *Rhizoctonia solani*, *Sclerotinia sclerotiorum* and *Phytophthora capsici* was due to different concentrations of the phenolic fraction in the essential oils (Muller Riebau *et al*. 1995) [48].

4.7 Acaricidal

Acaricidal activity of *L. nobilis* leaf oil against *Psoroptes cuniculi* was evaluated at a concentration of 10%, led to a mortality rate of 73%; at 5% the average activity was significantly reduced to 51%, while dilutions of 2.5, 1.25 and 0.625% were ineffective (Macchioni *et al*. 2006) [42].

4.8 Anticonvulsant

The leaf essential oil of *L. nobilis* was evaluated for anticonvulsant activity against experimental seizures. The essential oil protected mice against tonic seizures induced by maximal electroshock and especially by pentylenetetrazole. At anticonvulsant doses, the essential oil produced sedation

and motor impairment. This effect can be due to components such as methyleugenol, eugenol and pinene present in the bay essential oil (Sayyah *et al*. 2002) [59].

4.9 Cytotoxic

The growth inhibitory effects of the fresh and stored bay leaf essential oil on five human cancer lines were examined. The fresh essential oil exhibited maximum growth inhibitory effects on all cell lines than stored essential oil. Among the cancer cell lines, breast cell line and lung cell line both exhibited same effect of bay leaf essential oil (IC_{50} value of 0.8 µg/ml), followed by brain cancer cell line with an IC_{50} value of 0.9 µg/ml. Cervix cell line exhibited lowest sensitivity to essential oil (IC_{50} value of 1.8 µg/ml) (El-Sawi *et al*. 2009) [24]. *In vitro* cytotoxic activity of three different extracts of leaves of bay was evaluated against human peripheral blood mononuclear cells and three human cancer cell lines (Lung: A549, Breast: MCF-7 and Colon: COLO 205) by sulforhodamine B assay. Methanol extract rich in phenols, flavonols and flavonoids was found to be significantly more active and potent against all the cancer cell lines as compared to petroleum ether and aqueous extract of bay leaves but all three crude extracts lack cytotoxic effects on normal human cells (Thanekar *et al*. 2013) [62].

5. Conclusion

Screening of literature on bay showed that the essential oil possesses wide range of biologically active compounds. Broad variations were observed in the amounts of the main components of the essential oils from different parts of the plant, attributed to different geographic origins, growing conditions, seasonal variation and isolation procedures. The major compounds present in bay seed essential oil were 1,8 cineole, sabinene, limonene, eugenol and α-pinene. Bay leaf essential oil finds application with lot of pharmacological activities such as antimicrobial, insecticidal, antioxidant, anticonvulsant etc. Presence of variety of diverse constituents in bay leaf essential oil may be responsible for wide spectrum of biological activities of the plant.

6. References

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