Adulteration of Tea Tree Oil (Melaleuca alternifolia and M. linariifolia)

Article · September 2017

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Tea Tree Oil (Melaleuca alternifolia and M. linariifolia)

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Keywords: adulteration, Melaleuca alternifolia, Melaleuca dissitiflora, Melaleuca linariifolia, tea tree oil, tea tree leaf oil, Melaleuca oil, essential oil of Melaleuca, terpinen-4-ol

Goal: The goal of this bulletin is to provide timely information and/or updates on issues of adulteration and mislabeling of tea tree oil (TTO), in particular with Eucalyptus oils, as well as essential oils of other Melaleuca species not declared in the Australian (AS 2782) and International Organization for Standardization (ISO) 4730 norms, and those species of the closely related genus Leptospermum.1-4 This bulletin may serve as guidance for quality control personnel, the international herbal products industry, and the extended natural products community in general. It is also intended to present a summary of the scientific data and methods on the occurrence of species substitution, adulteration, the market situation, and economic and safety consequences for the consumer and the industry.

Scope: The ISO standards allow two species, Melaleuca alternifolia and M. linariifolia, to be used for the production of tea tree oil, while other standards, e.g., the monograph in the European Pharmacopoeia, include M. dissitiflora and other species of Melaleuca as sources of TTO as well. Since the available data on adulteration have been based mainly on comparison of commercial TTO with ISO standards, and M. dissitiflora oil is generally not available in commerce, the scope of this Bulletin is limited to adulteration of products labeled to contain the essential oil of Melaleuca alternifolia or M. linariifolia. Since M. dissitiflora is an acceptable species following the European Pharmacopoeia standards, the sale of essential oils from this species as TTO should not be considered as adulteration.
1. General Information

1.1 Common name: Tea tree, tea tree leaf oil, melaleuca oil, Australian tea tree oil

1.2 Other common names

English:
- Melaleuca alternifolia: tea tree, paperbark tree, narrow-leaved paperbark; Melaleuca linariifolia: flax-leaved paperbark tree, snow-insummer

Aboriginal Australian:
- Melaleuca linariifolia: Budjur

Chinese: common name: 茶树精油 (Australian tea tree oil), 澳洲茶树精油

Danish: Tetræ

French: Mélaleuca (arbre à thé), mélaleuque, tea tree, théier Australien

German: Teebaum, Australischer Teebaum

Italian: Melaleuca, tea tree, albero del té

Maori: Ti Tree

Norwegian: Tea tree

Spanish: Árbol de té, Melaleuca alternifolia

Swedish: Teoljebuske, tetrad

1.3 Accepted Latin binomial for both species:

Melaleuca alternifolia (Maiden & Betche) Cheel

Melaleuca linariifolia Sm.

1.4 Synonyms for both species:

Melaleuca alternifolia: Melaleuca linariifolia var. alternifolia Maiden & Betche.

Melaleuca linariifolia: Melaleuca hyssofifolia (Cav.) Dum. Cours, Myrtoleucodendron linariifolium (Sm.) Kuntze, and Ozandra hyssofifolia (Cav.) Raf.

1.5 Botanical family: Myrtaceae

1.6 Distribution: Melaleuca alternifolia and M. linariifolia are both native Australian species endemic to the East coastal littoral of continental Australia from Maryborough, Queensland in the north to Port Macquarie, New South Wales in the south and west to the Great Dividing Range. The native habitat of M. alternifolia is low-lying, swampy, subtropical, coastal ground. Melaleuca linariifolia has a more limited distribution range, being endemic to the Australian states of Queensland and New South Wales. It grows in heath and dry sclerophyll forest in moist or swampy ground; on the East coast, Central and Southern Australia, and adjacent ranges.

Melaleuca alternifolia has been introduced and cultivated in Brazil, China, Indonesia, Kenya, Madagascar, Malaysia, South Africa, Tanzania, Thailand, United States, and Zimbabwe. Melaleuca linariifolia has been introduced and cultivated outside of Australia is not easily determined. In China, Melaleuca species were first introduced about 100 years ago, along with Eucalyptus. In 2004, TTO production in the Guangxi Zhuang Autonomous Region in the South of China was 60–80 tons/year. Of these, 40–50 tons were good quality oil (defined by the authors as 40–50% terpinen-4-ol and <3% eucalyptol [1,8-cineole]) coming from forestry plantations of selected planting stock, according to the Guangxi Forestry Research Institute (GFRI).

South Africa is a significant producer of tea tree oil. Relatively small cultivations on the African continent are in Madagascar, Kenya, Tanzania, and Zimbabwe. In Kenya, tea tree is grown by local farmers around the small town of Naro Moro on the slopes of Mount Kenya. According to a commercial website there are as many as four million planted trees for tea tree oil production.

1.7 Plant part and raw material form: Leaves and terminal branchlets. The essential oil is obtained by steam distillation.

Melaleuca alternifolia has six different chemotypes, with three considered the main (also known as cardinal) chemotypes:

1. The terpinen-4-ol chemotype which yields tea tree oil rich in terpinen-4-ol.
2. The cineol chemotype which yields tea tree oil rich in 1,8-cineole.
3. The terpinolene chemotype, which yields tea tree oil rich in terpinolene.

Modern research into the efficacy, safety, and uses of tea tree oil has been conducted only on the terpinen-4-ol type, because it has been consistently identified as the most effective antimicrobial product with the best therapeutic qualities sought by consumers in the treatment of acne, dental applications, and as an aid in oral and foot infections, to name a few.

While M. linariifolia is named as a source botanical in monographs and the Australian and International Standards Organization (ISO) norms, plantations in Australia...
predominantly use *M. alternifolia* for essential oil production, since it is easier to grow, more abundant, and has been the subject of the most intense selective breeding to improve essential oil yields.26

**1.8 General use(s):** *Melaleuca alternifolia* has a long history of traditional medical use in Australia as a topical antiseptic, antibacterial, antifungal, anti-inflammatory, and antiviral agent.25,27 *Melaleuca linariifolia* traditional use information is more sparse; however, the chemical composition of its essential oil is considered similar to that of *M. alternifolia*.28 The primary indications for TTO include minor cuts, minor burns, abrasions, pimples, athlete’s foot, insect bites, stings and nasal and chest congestion.13,14,29 The oil is used to treat fungal infections, in particular, candidiasis and dermatophytosis, and it is available worldwide both as neat oil and as an active component in an array of commercial products in Australia, and many other parts of the world.

Some specific medical uses identified in Australian product registrations include the following: temporary relief of cough due to bronchial irritation or bronchitis, relief of the symptoms of catarrh, colds, cold sores, influenza/flu, sinusitis, and tinea by topical application. Tea tree oil is also used as an aid in the maintenance or improvement of general well-being, relief of the symptoms of allergies, in aromatherapy and massage, and as a first-aid treatment of minor burns.30,32 The European Medicines Agency’s community herbal monograph specifies the following indications: small superficial wounds and insect bites, small boils (furuncles and mild acne), the relief of itching and irritation in cases of mild athlete’s foot, and symptomatic treatment of minor inflammation of the oral mucosa. Commercial products include gels, creams, lotions, shampoo, foot care products, soaps, toothpastes, insect repellents, and air fresheners.25,30,33

**1.9 Nomenclature considerations:** The name *tea tree* causes much confusion. The most frequent confusion is with the renowned tea plant (*Camellia sinensis*, Theaceae), but also with other members of the Myrtaceae family growing in Australia. *Tea tree* is confused with a number of species of the genus *Melaleuca*, species of the closely related genus *Leptospermum*, as well as species of the genera *Kunzea* and *Baeckea* growing in Australia and New Zealand.7 The reason for this confusion is that the vernacular name “tea trees” is referred collectively to a group of plants which are also known as “paperbark trees” and both terms are still used to refer to any member of the *Melaleuca* or *Leptospermum* genera, of which there are several hundred. For instance, common names for *M. cajuputi* include “swamp tea tree” and “paperbark tea tree”, while those for *M. quinquenervia* include “broad-leaved tea tree” and “broad-leaved paperbark”.33,35 In Australia, many *Leptospermum* species are cultivated domestically and these are often mistaken as a source of tea tree oil. In addition, the essential oils derived from the New Zealand plants *Kunzea ericoides* and *Leptospermum scoparium*, and known commonly as kanuka and manuka oils, respectively, are often referred to as New Zealand tea tree oils (also known as ti tree oils), although they have a different chemical composition.36

**2. Market**

**2.1 Importance in the trade:** The use of tea tree oil as an ingredient in commercial products is quite extensive. Global production rose from 200 metric tons (MT) in 1995 to 540-580 MT in 2012.33,37 In 2013, Brophy et al. estimated that world trade of TTO was in excess of 36 million Australian dollars (AUD); with Australia being the largest producer and exporter.25 In Australia, there were 3,000 hectares (ha) of *M. alternifolia* trees planted for the production of essential oil, which produced a reported 450–500 MT of essential oil in 2013.25

In 2015, ATTIA reported over 800 MT of TTO being produced.26,27 However, the total area planted and the total production worldwide is potentially twice as much from plantings in several countries outside Australia, including China.23

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<tbody>
<tr>
<td>Production (MT)</td>
<td>407</td>
<td>551</td>
<td>667</td>
<td>845</td>
<td>714</td>
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<tr>
<td>Available Supply (MT)</td>
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<td>555</td>
<td>680</td>
<td>848</td>
<td>726</td>
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<td>542</td>
<td>677</td>
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<td>711</td>
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<td>Demand change (%)*</td>
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<td>-1.3%</td>
<td>25.0%</td>
<td>23.4%</td>
<td>-14.9%</td>
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<tr>
<td>Closing Stock (MT)</td>
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<td>3</td>
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<td>Average Price/kg (AUD)</td>
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<td>$37.17</td>
<td>$43.02</td>
<td>$45.02</td>
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<tr>
<td>ABSd Export (MT)</td>
<td>N/A</td>
<td>443</td>
<td>582</td>
<td>620</td>
<td>584</td>
</tr>
</tbody>
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*a* The reporting period is from April 1 to March 31, except for 2012/2013 (January 1 to March 31)

Data provided by Australian Tea Tree Oil Association (ATTIA). All figures are given in metric tons (MT), except for Average Price, given in Australian dollars, and the demand change. The implied sales demand figures are derived by subtracting the closing stock amounts from the available supply figure. The 2016/17 projected production figures are the expected harvest figures as declared by growers to ATTIA.

*b* Compared to prior year

*c* Amount of inventory at the end of the reporting period

*d* Australian Bureau of Statistics

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2.2 Supply sources: As mentioned above, Australia is the country with the largest production of TTO with around 800 MT produced annually. The year 2015 was a record at 845 MT, but just exceeded 700 MT in 2016 due to adverse conditions (drought). *Melaleuca linariifolia* is grown on only one plantation in Australia (approx. 3,500 plants) with limited annual production.26

Chinese TTO production figures are not easy to obtain, but ATTIA estimates that the country produces about 200 MT annually, derived from *M. alternifolia* (usually essential oil with a high 1,8-cineole content). (Tony Larkman [ATTIA] oral communication to Ezra Bejar, December 4, 2016)

Other TTO-producing countries are Kenya (~20 MT), Zimbabwe (~20 MT) and South Africa (unknown, but estimated at 35 MT), which all provide essential oil that complies with ISO standards. There are a few minor plantations in the United States (California), New Zealand, Thailand, and Malaysia, but no data are available on any of these. (Tony Larkman [ATTIA] oral communication to Ezra Bejar, December 4, 2016)

2.3 Market dynamics: In Australia, over the past eight years, farm gate prices of pure TTO from *M. alternifolia* have fluctuated in the range of AUD $29.00 to AUD $49.00 per kg, depending on the supply and demand dynamics. Prices increased from $29 to a peak price in 2008 of AUD $49.00. After that year prices declined steadily to a low of AUD $30.35 in 2012. In the last four years, the material has regained value (Table 1). The current estimated farm gate price for TTO is in the range of AUD $40.00-AUD $43.00 per kg, excluding packaging.11,26

The Australian Bureau of Statistics (ABS) reported Australian TTO exports in 2016 having a total value of AUD $25,203,914 for all regions in the world. In the first trimester of 2016, total exports were $7,087,020, with a 62.5% share (based on dollar value) going to North America. Europe had the second largest share (25.2%), followed by Asia (10.7%), then Africa and the Middle East (1.6%), and South America with no data in the first trimester.38

The price range outside Australia for materials labeled as tea tree oil has been as low as US $20.00 per kg, which provides an economic incentive to manufacturers for using those materials. Recent sales information from China, as well as the other TTO producing countries, is unavailable.

3. Adulteration

3.1 Known adulterants: Adulteration is known to be a significant problem in the essential oil industry, driven by optimization of profits by using lower-cost synthetic material, added to natural essential oils.39 The most common adulterants found in the marketplace are low-cost versions of essential oils, containing similar terpenes (volatile) compounds, made, for example, with synthetic material or essential oil components from natural sources. Another problem is the addition of vegetable oils.39-42

In the case of TTO, adulteration involves spiking with other essential oils or pure compounds of natural or synthetic origin.43 Added materials may include byproducts of the eucalyptus (*Eucalyptus globulus* and others species of *Eucalyptus* [Myrtaceae family]) oil industry, pine (*Pinus* spp, *Pinus*ae) oil or white camphor (*Cinnamomum camphora*, Lauraceae) oil, which are frequently found in the chromatographic analysis of so-called TTO.34,43 Eucalyptus essential oil is extracted by steam distillation of the leaves of several *Eucalyptus* species cultivated worldwide. China produces about 90% of the world trade of the medicinal cineole-rich eucalyptus oil. However, the presence of borneol and camphene in the chromatographic analysis of TTO samples coming from China, as tested by ATTIA, cannot be solely explained by adulteration with eucalyptus oil and/or its constituents. These compounds are not present in TTO or eucalyptus oil.7,44-47

Less frequently other Myrtaceae species are used as sources for adulteration. This may include species of the genus *Melaleuca* not included in the ISO TTO standards norm, as well as species of a closely related genus, *Leptospermum*, growing in Australia and New Zealand. Other suggested adulterants of TTO include tea tree species known as cajuput (*Melaleuca cajuputi* Powell), niaouli (*Melaleuca viridiflora* Sol. ex Gaertn.), manuka (*Leptospermum scoparium*), or kanuka (*Kunzea ericoides* A.Rich.) oils.44-47 However, adulteration with cajuput, niaouli, manuka, or kanuka oils does not appear to make economic sense. The oils of these species are relatively expensive and thus there would be minimal financial gain in using them.

ATTIA estimates that China exports up to 1,200 MT of products labeled as tea tree oil annually, and while reliable data are scarce, it estimates that China may be producing only up to a maximum of 200 MT per year from *M. alternifolia*. This indicates that up to 1,000 MT of adulterant may be used to extend the small (~200 MT) output of genuine material, resulting in a supply of material that is of highly variable quality and composition.

3.2 Sources of information supporting confirmation of adulteration: The earliest references to alleged adulteration of TTO were obtained indirectly, in the form of adverse event reports or internal emails and faxes with complaints to ATTIA from 1990-2000.48

In 2013, a publication suggested that compounded and uncertified TTO coming mainly from China had been flooding the markets, causing problems to consumers and cosmetic manufacturers. The report indicated compounded TTO, containing the 15 main compounds, was either blended with genuine TTO and resold as Australian TTO or re-exported as is to suppliers in Europe and the United States.49

In 2015, Australian researchers found consistent enantiomeric ratios for 57 samples of authentic 100% pure Australian TTOs they analyzed. The averages were: 68.5 ± 0.2% (+) for terpenin-4-ol and 74.2 ± 1.4% (+): 25.8 ± 1.4% (−) for α-terpineol in oils sourced directly from different plantation sites throughout Australia. In contrast, considerable ratio variations were observed for 43
commercial TTOs sampled from North America, the European Union, Asia, Australasia, and South Africa. About 50% of the samples had between 26-62% of (+)-terpinen-4-ol, differing markedly from the 68-70% value found for Australian TTO. The TTOs sourced internationally were from companies that mix Australian with Chinese and other foreign TTO sources, as well as adulterating substances. A concern that pure Australian TTO was being intentionally adulterated, extended (diluted), or otherwise modified with undisclosed lower-cost ingredients was tested using enantioselective multidimensional gas chromatography (eGC). Results suggested pure TTO from Australia was free of adulteration; however, commercial samples from different continents, all labeled as M. alternifolia oil, showed a large variation which indicated the possibility of adulteration.

Occurrence of adulteration was confirmed by Wang et al., 2015, using the same 100 TTO samples (57 samples from Australian tea plantations & 43 commercial TTO samples) as Wong et al. In addition, Wang et al. reported that samples that were ISO4730:2004-compliant when distilled did not meet the ISO standards when tested using conventional GC-MS analysis because of excessive p-cymene and/or reduced α-terpinene, γ-terpinene, and terpinolene concentrations. Autoxidation was thought to be responsible for most of the observed deviations. Forty-nine percent of the commercial products did not meet the ISO specifications. There was a substantial subset of commercial products that met ISO4730: 2004 standards, but displayed unusual enantiomeric +/− ratios.

Evidence for the sale of adulterated TTO is also provided by a recent investigation by the Australia’s Competition and Consumer Commission (ACCC). The Commission eventually fined Felton Grimwade & Bosisto’s Pty Ltd AUS $10,800 after chemical analysis showed that the company’s tea tree oil was adulterated. The identity of the adulterant was not detailed in the infringement notice.

Finally, an analysis of 25 tea tree oil samples from markets in Australia, the United Kingdom, North America, and South Africa by GC-MS and determination of (+)- and (-)-terpinen-4-ol ratios found evidence for adulteration in eight samples (32%).

### 3.3 Accidental or intentional adulteration:
According to ATTIA, adulteration was both partial (by mixing adulterating materials with TTO) and complete (by substituting the TTO in its entirety with materials of lower quality) in the early days of the TTO industry. Since the start of ATTIA’s campaign about the Australian and ISO standards and the use of chiral analysis, the nature of adulteration has changed and has become more sophisticated with more care taken to hide the irregularities. The only conclusion that can be made from this is that TTO adulteration is definitely intentional.

### 3.4 Frequency of occurrence:
The frequency of adulteration appears to depend on the geographic origin of the material. ATTIA tested TTO samples from Australian and Zimbabwe plantations, as well as commercial samples from Australia, Canada, China, Germany, Indonesia, Italy, Korea, New Zealand, Spain, United Kingdom, United States, and Zimbabwe. None of the Australian plantation samples were adulterated showing a distinctive profile in Chiral Analysis. Most commercial samples from New Zealand (2/2), Australia (13/20), and United States (23/36) met the TTO specifications, while all (10/10) commercial samples from China were considered adulterated. Adulteration rates in European commercial samples were 7/20 (United Kingdom), and 4/11 (Spain). (Tony Larkman [ATTIA] oral communication to Ezra Bejar, January 2, 2017)

Wohlmuth et al. found that one third (2/6) and two thirds (4/6), respectively, of Australian and South African samples were adulterated. Of the four samples from the United Kingdom, one was considered definitely adulterated and two others possibly adulterated. One of the five samples (20%) analyzed from North America was not authentic TTO.

Considering all the results together, the adulteration of TTO appears to be rather common.

### 3.5 Possible safety/therapeutic issues:
The prevalence of contact allergy to tea tree oil is rare, ranging from only 0.3–2.7%. It has been suggested that the positive in vitro tests to TTO are either caused by oxidized ingredients or due to the presence of an adulterant. Also, allergic reactions to TTO appear to occur only in predisposed individuals and may be due to individual sensitivity, or to various oxidation products, including peroxides, which are formed by exposure of the oil to light and/or air.

Methyl eugenol, a compound that is of certain toxicological concern due to its structural similarity to estragole and safrole (which have shown carcinogenic effects in rodents), has been reported to occur at higher levels in M. dissitiflora than M. alternifolia. The higher methyl eugenol concentration has been cited as one of the reasons why M. dissitiflora was eliminated as a source material for TTO in the 2017 ISO standard.

### 3.6 Analytical methods to detect adulteration:
Since the essential oil is the ingredient of commerce, microscopic and macroscopic methods of analysis for authentication are not useful beyond the distillation factories, and are not applicable for the identification of TTO. Pharmacopoeial and normative methods focus on ingredient testing after steam distillation and include organoleptic evaluation, density, refractive index, optical rotation, thin-layer chromatography (TLC), conventional gas-chromatography (GC), GC-mass spectrometry (MS), chiral GC-MS analysis, and subsequent chemometric analyses. However, these methodologies may not be sufficient to detect all adulterations.

The European and British pharmacopoeias also use a GC method for authentication with flame-ionization as the detector system. The authentication is based on relative percentage of 11 individual terpenes in the TTO. Many GC methods have been developed since the 1980s, including the use of chiral columns. These methods provide
a clear profile of the chemical composition, and chemical variation of TTO, and established quality control procedures to identify adulterants. Mass spectrometry has now become the standard detection system for gas chromatographic separations.\textsuperscript{17,22,44,57-63} Determination of enantiomeric ratios of terpinen-4-ol, and α-terpineol using lanthanide shift reagents has also been achieved by nuclear magnetic resonance (NMR), but the successful application to TTO authentication has yet to be shown.\textsuperscript{44}

The GC-MS methods using chiral columns, allowing the separation of enantiomers and the determination of enantiomeric ratios, has provided clear criteria to identify adulterants in TTO.\textsuperscript{27,43,47,51} However, Davies et al. evaluated enantiomeric ratio test results obtained for a set of 57 samples analyzed by GC-MS in three laboratories and concluded that the complete resolution of the terpinen-4-ol enantiomers was not always achieved, leading to a variability in average terpinen-4-ol enantiomer ratios (63.3-69.8/30.2-36.7 (+)/(−)-terpinen-4-ol) among the laboratories. The authors suggested that absolute ratios for terpinen-4-ol enantiomers may not be an appropriate way to determine TTO authenticity, but that the ratios should be set by each analytical laboratory using a reference material with a known ratio.\textsuperscript{27} However, no such reference material is available to our knowledge.

Wong et al.\textsuperscript{51} conducted chiral analysis of three key monoterpenes (limonene, terpinen-4-ol and α-terpineol) present in tea tree oil by a combination of enantioselective multidimensional GC [GC – eGC] and two-dimensional gas chromatography [eGC × GC]. Ranges of TTOs sourced directly from plantations of known provenance in Australia were compared with commercial TTOs from different continents. Fast chiral separations were achieved within 25 min for GC – eGC and < 20 min for eGC × GC. Exact enantiomeric composition of chiral markers for authentic TTOs was achieved. Consistent enantiomeric fractions for limonene, terpinen-4-ol, and α-terpineol were obtained for 57 authentic Australian TTOs. In contrast, commercial samples from different continents showed a large variation. Specifically, samples from the United States and the United Kingdom had much higher (+) limonene and much lower (+) terpinen-4-ol averages. Some of the (+)-α-terpineol values were inconsistent; they were either too low or high, which suggested adulteration.\textsuperscript{51}

A large set (n = 104) of TTOs and commercial TTO products were analyzed with a combination of conventional GC-MS, chiral GC-MS, and chemometric techniques. Twenty terpenoids were determined in each sample and compared with ISO standards (ISO-4730: 2004).\textsuperscript{47}

The combined results and conclusions of these reports served as the basis for a new version of ISO 4730, which was released in February 2017. The TTO ISO4730: 2017 standard addresses several of the issues with the older ISO 4730: 2004 in terms of tighter requirements for the chromatographic profile of the essential oil by gas chromatography. The new standard establishes a minimum of 35% terpinen-4-ol, 14% γ-terpinene and 6% α-terpinein.\textsuperscript{4} The Australian Standard (AS2782: 2009) is also being updated to be identical to the new ISO version (it will be AS2782: 2017).

The cited reports suggest that both chiral and chemometric analyses are needed to confirm the authenticity of commercial products, including those that met all of the ISO4730: 2017 standards which can be used to assess possible adulteration.

4. Conclusions

Adulteration of TTO with synthetic terpinen-4-ol, or industrial waste from ‘normalizing’ eucalyptus, and other essential oils such as pine and white camphor occurs frequently by intentional dilution of the ingredient to reduce production costs. The lack of an effective standard were past challenges that have been overcome with the new 2017 ISO4730 norm, which has tighter ranges for the main 15 terpenes found in authentic TTO. The most common adulteration of TTO with eucalyptus and pine oils is readily detected using GC and chiral enantiomeric analysis and has been incorporated into the new ISO norm (ISO4730: 2017). Adulteration with other Melaleuca species such as cajuput, niaouli, manuka, or kanuka oils rarely occurs in practice, as they are too expensive.

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