

# Absinthe—A Review

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*The alcoholic beverage absinthe is recently experiencing a revival after a yearlong prohibition. This review article provides information on all aspects of this bitter spirit and its major components, especially wormwood (*Artemisia absinthium* L.), which contains the toxic monoterpene thujone. Over 100 references on historic and current literature are provided. The topics comprise the history of the alcoholic drink starting from its invention in the eighteenth century. Historical and modern recipes are discussed in the context of different quality categories and possibilities to reduce the content of thujone are given. The analytical techniques used to verify compliance with the maximum limit of thujone as well as further possibilities for quality control of absinthe are discussed. The toxicology of absinthe is reviewed with regard to the cause of a syndrome called "absinthism," which was described after chronic abuse of the spirit in the nineteenth century. Finally, a food regulatory and food chemical evaluation is provided and minimum requirements for absinthe are suggested. Absinthe should have a recognizable wormwood flavor and after dilution with water the characteristic clouding should arise (louche-effect). Products, which are advertized as being of premium grade should be made by distillation, should have an alcoholic strength of at least 45%vol, and should not contain artificial dye.*

**Keywords** absinthe, wormwood, *Artemisia absinthium* L., thujone, absinthin, spirit drink

## INTRODUCTION

The currently en-vogue beverage absinthe, a mostly green colored spirit drink of distinct bitter taste, is recently experiencing a revival, after nearly 70 years of prohibition. It has found a way of distribution through the Internet, a wide variety is also found in retail trade.

Because of the yearlong prohibition, no official requirements or actual legal definitions exist, which explain or define how a spirit drink that is called absinthe should be like. Nowadays, a wide variety of absinthe variations are offered, which are all subject to the official food control. In this context, a number of substandard products, which do not show any organoleptic characteristics of the wormwood plant, were recently found on the market.<sup>1</sup> In this article, all essential literature concerning absinthe is combined to provide an extensive tool in the evaluation of products for food control as well as for spirit drink manufacturers. After a historical survey of absinthe, a summary

of the analytical and toxicological facts is given. Minimum requirements for products, which enter the market as absinthe, are proposed.

## WORMWOOD—ARTEMISIA ABSINTHIUM L.

The wormwood plant (*Artemisia absinthium* L.), which is part of the family of composites (Asteraceae)<sup>2,3</sup> has given absinthe its name and is, besides alcohol, the main component of this spirit drink. Wormwood is a half-shrub which is local in Central Europe and Asia with silvery grey, pinnatipartite, felty-haired leaves. The small, light yellow and globular flowers are arranged in end standing panicles (Figure 1).<sup>4</sup> The plant parts above ground are harvested in anthesis and after drying are used for the manufacturing of spirit drinks.<sup>5</sup> Besides *Artemisia absinthium* L., the Roman or petite wormwood (*Artemisia pontica* L.) is sometimes used, especially for coloration.

Wormwood contains an oil (0.2–1.5%) of a color ranging from dark-green or brown to blue. It is of a scratchingly adstringent bitter taste and smells rather strong.<sup>6</sup> The bicyclic

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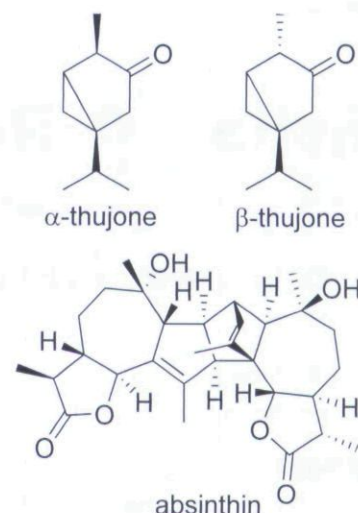




**Figure 1** Wormwood, *Artemisia absinthium* L., drawing of plant, flowers, seeds and fruits (drawing by W. Müller, 1885).<sup>4</sup> Wormwood is the characteristic aromatic component of absinthe.

monoterpene thujone was often described as the main component of wormwood oil (40–90% of the essential oil). One distinguishes between the two isomeric components  $\alpha$ - and  $\beta$ -thujone, in dependence on the stereo chemistry of the C-4 methyl group (Figure 2).<sup>7</sup> The concentration of  $\beta$ -thujone is usually higher than the one of  $\alpha$ -thujone ( $\beta$ -thujone: 70–90% of the total thujone content).<sup>8</sup> However, besides the  $\beta$ -thujone chemotype of the wormwood plant further chemotypes were described, which contain *cis*-chrysanthenyl acetat, *cis*-chrysanthenol, *cis*-epoxyocimene, sabinyl acetate or bornyl acetate as principal component.<sup>9–15</sup> In the west alpine area above 1000 m the *cis*-epoxyocimene type is predominant, while the  $\beta$ -thujone type rather exists in the lower zones.<sup>12</sup> In wormwood oil from the Tuscany<sup>16</sup> or the Pyrenees<sup>10</sup> neither  $\alpha$ - nor  $\beta$ -thujone could be detected.

Further characteristic components of wormwood are terpene lactone bitter substances such as absinthin (0.20–0.28%) and artabsin (0.04–0.16%),<sup>2</sup> while the sesquiterpene lactone absinthin (Figure 2) is the organoleptically dominant bitter



**Figure 2** Structure of  $\alpha$ - and  $\beta$ -thujone and absinthin, the principal components of wormwood (*Artemisia absinthium* L.)

principle.<sup>17</sup> The concentration is seasonally different. The highest content of bitter compounds can be found in September, while the highest content of essential oil can be observed from June to July.<sup>18</sup>

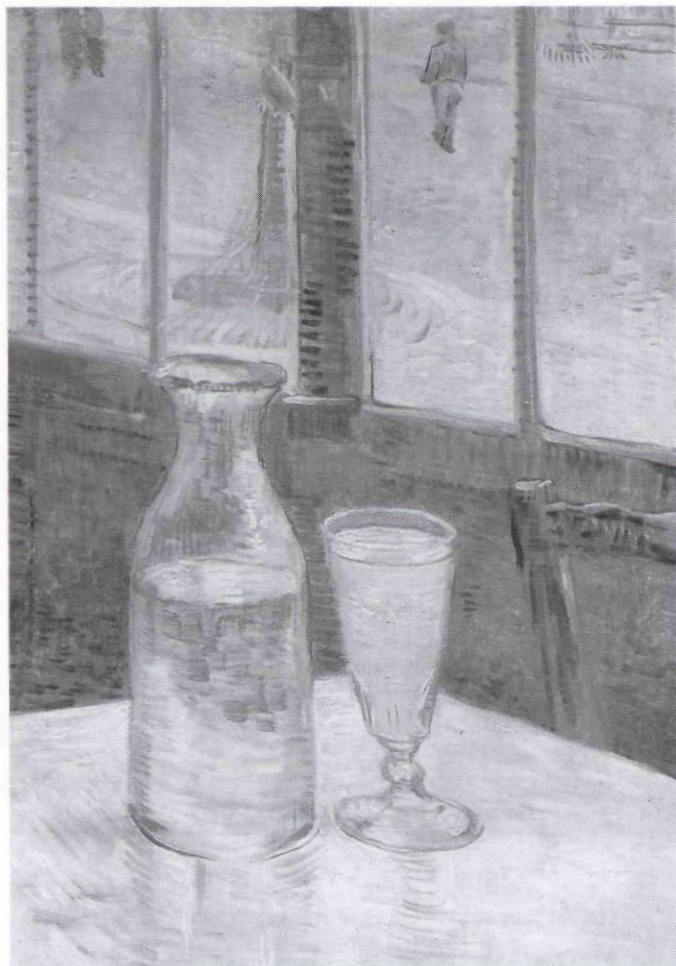
## HISTORICAL FACTS ABOUT ABSINTHE

Plinius already mentioned a wine called "absinthites" prepared by adding the herb of wormwood.<sup>19,20</sup> Wine containing beverages, with watery wormwood extracts are still available under the label vermouth. The spirit drink known as absinthe was created in French-speaking Switzerland in the late eighteenth century. The herb of wormwood, used as medicine since antiquity, was mixed for the first time with further herbal ingredients for flavoring and after the addition of alcohol distilled and distributed as foodstuff. The origin of this recipe is Couvet, in the Val-de-Travers (Neuchâtel, Switzerland) and both, Dr. Pierre Ordinaire,<sup>21–24</sup> and Henriette Henriad<sup>20,25</sup> were credited with its invention. However, it is proven, that the recipe got into the possession of Henry-Louis Pernod, who founded a distillery in Pontarlier (France) in 1805.<sup>20,22,25</sup>

In the Wars of French Algeria (1844–1847), alcoholic beverages containing wormwood were given to the soldiers as a prophylaxis against miscellaneous diseases (such as malaria and helminthiasis) and to raise the fighting spirit.<sup>22–24</sup> After the war, the homecoming soldiers made absinthe popular in their own countries.<sup>19,26</sup> A further reason for the wide spreading of absinthe in the nineteenth century, could be attributable to the fact that grave losses in vindemiation lead to rising wine prices, while the prices for absinthe were falling.<sup>25</sup> In the late nineteenth century, absinthe, in the meantime called "green fairy" ("fée verte"), was the most popular spirit drink in Europe.<sup>27</sup>

The green colored drink was consumed by the population of all social levels. Especially in the bars and cafés of Paris, the "green hour" ("l'heure verte") was a steady element of the





**Figure 3** Vincent van Gogh: Still Life with absinthe (Paris 1887). The picture shows one of the countless cafés in Paris, in which absinthe was served. Next to the glass filled with absinthe, a water bottle is illustrated, which was necessary for the drinking ritual.

daily routine. Absinthe also found its way into the spheres of artists and intellectuals. Degas' "The Absinthe," van Gogh's "Still Life" (Figure 3), Picassos' "The Absinthe Drinker" and Toulouse-Lautrec's portrait of his colleague van Gogh, marked by absinthe use and probably psychiatric symptoms, give proof of the disastrous addiction of so many.<sup>22</sup> The biography of Vincent van Gogh is seen by many as the classic example of continuous absinthe consume.<sup>24,28–32</sup>

Pontarlier became the capital of absinthe. In 1905, 25 distilleries had existed with approximately 3000 workers and an annual production of 10 million litres of absinthe.<sup>25</sup>

But as early as in 1850, it was observed that the regular consume of absinthe caused a syndrome called "absinthism." However, only as a result of the mass consumption in the beginning of the twentieth century, absinthe was blamed for all kinds of diseases and its prohibition was demanded. In 1905, absinthe was prohibited in Belgium, followed by Switzerland in 1908, the Netherlands in 1910, the USA in 1912 and Italy in 1913.<sup>22,23</sup> In France absinthe was prohibited in 1915, because of the misuse by the French military during the First

World War.<sup>20,23</sup> Finally in 1923, absinthe was also prohibited in Germany.<sup>33</sup>

While absinthe was banned throughout Europe for more than 70 years, Spain and the Czech Republic were exceptions to the rule. Because of an obscure legal situation, a continuous production in countless distilleries existed in these countries. In France, the big absinthe manufacturers invented substitute products (so-called Imitation Absinthes) without wormwood, such as Pernod, Ricard, or Pastis.<sup>7</sup>

According to the Council Directive 88/388/EEC on the approximation of the laws of the Member States relating to flavorings for use in foodstuffs and to source materials for their production, the addition of thujone containing plants or flavorings prepared from natural raw materials was re-allowed in the European Union.<sup>34</sup> After the obligatory adoption of the Directive by the member states in the early 1990s, absinthe was marketable again within the entire European Union. For bitter spirit drinks, such as absinthe, a thujone maximum limit of 35 mg/kg  $\alpha$ -/ $\beta$ -thujone was introduced in the Directive. Over 10 years after the lift of the prohibition, more than a hundred absinthe types are currently sold, which are mainly distributed as en-vogue drinks via the Internet. In bars, absinthe is served as a cocktail or long drink.<sup>35</sup>

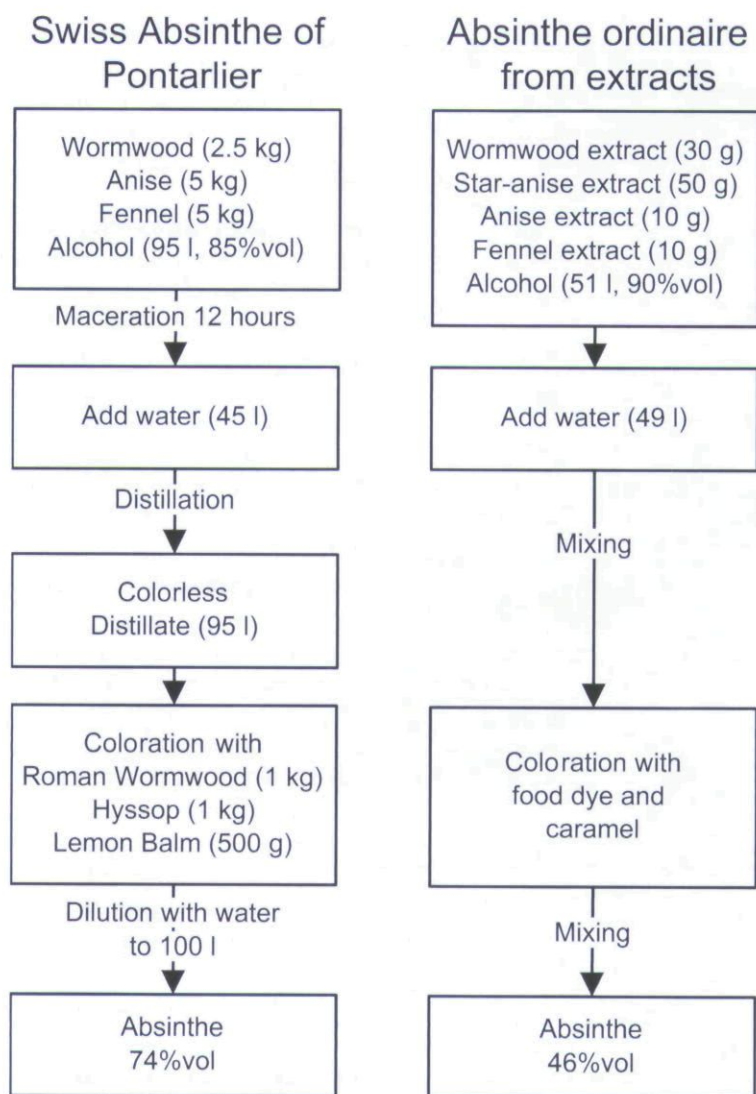
In Switzerland, absinthe's country of origin, the prohibition was recently lifted<sup>36</sup> with the same maximum limits as in the EU,<sup>37</sup> which makes it possible, that high-grade absinthe products manufactured according to traditional Swiss recipes may show up on the market in the future. Switzerland also proposed to introduce protected geographic denominations of origin and protected geographic indications on the labelling of absinthe. Further requirements for Swiss absinthe, such as the ban of artificial dyes are currently in discussion.

## ABSINTHE PRODUCTION

Following traditional recipes for distilled absinthes, in the first step wormwood and other dried herbs (e.g. anise, fennel) are macerated. The macerate of the wormwood herb is of a greenish-brown color, smells aromatic, like all *Artemisia* species, and reminds one of the composites, like camomile. The taste is lightly stinging, strongly bitter, and camphoric.<sup>5</sup> The following distillation of the macerate results in a distillate that is reduced of the bitter compounds, which are relatively non-volatile. The characteristic, light volatile, fine aromatic components of the wormwood aroma appear in the first fraction between 80 and 60%vol, while the middle fractions possess a cinnamon or clove-like aroma.<sup>5</sup>

In a second step, wormwood (usually *Artemisia pontica*) and other herbs are added to the colorless distillate. This is done to accomplish the characteristic green coloring by chlorophyll and to achieve a mild bitter taste, as well as to extract other aromatic compounds. Because of the easy denaturation of chlorophyll through light and warmth, the characteristic color of a traditionally produced absinthe is only pale green. Afterwards the beverage is diluted with water until the drinking strength is reached.





**Figure 4** Procedure for the manufacturing of absinthe after historical recipes. The Swiss absinthe from Pontarlier from Ref.<sup>40</sup> is given as example for a premium quality product. The absinthe ordinaire manufactured from extracts according to Ref.<sup>39</sup> is indicative of a lower grade product.

Typical historic recipes are given in Figure 4. The composition of herbs used next to wormwood differs from recipe to recipe.<sup>38–41</sup> For the improvement of the taste, or for further coloring anise, star anise, lemon balm, hyssop, juniper, nutmeg, veronica, angelica root, melissa, coriander, camomile, or parsley are added.<sup>19,22,40</sup> In every country, own types of absinthe were introduced. For example in the Czech Republic, peppermint was added, but no anise and fennel. In Switzerland, melissa, hyssop or angelica root were used, while in France coriander was added. The Swiss alpine wormwood is a valued ingredient, because of his strong aroma.<sup>6</sup>

The absinthe recipes of nowadays are kept as corporate secrets. Besides wormwood, other plants are used for the macerate. The colour is greenish or yellowish and clear.

The drying process has a considerable influence on the quality of the essential oils of the plants. Tateo and Riva<sup>42</sup> observed that even the drying with only lightly heated air (25–39°C) leads to

an unacceptable change of the organoleptic quality of the wormwood oil. To achieve a good quality, the drying process should be performed with or without artificial air at room temperature. The drying and storing process has no influence on the bitter compound absinthin.<sup>17</sup> Tateo<sup>43</sup> also found that the extraction of *Artemisia* varieties can be enhanced using enzymatic treatment with hemicellulase.

The top grade products are still manufactured with regard to the traditional recipes without the addition of dye or other additives. Some products are made of herbal distillates and are differentiated by a mild flavor. Because such products are colorless, they are sold as “Blanche” or “La Bleue.” Types with a lower alcoholic strength and added sugar are sold as absinthe-liqueurs. Independent of the traditional recipes many products sold nowadays are made with readily bought finished extracts of wormwood or other plants, which are blended with ethyl alcohol of agricultural origin. For the coloring, artificial dye



is used, especially mixtures of tartrazin (E102, FD&C Yellow No. 5) and patent blue V (E131), or brilliant blue FCF (E133). Deteriorated products contain no herbal extracts and are made solely by the blending of artificial flavors, coloring, and ethyl alcohol.

In cases like this, sometimes even the macerated herbs are not distilled but only filtrated, diluted to drinking strength and bottled. These products have a strong pronounced taste of wormwood and a very strong, nearly unpleasant bitter touch. Further falsification is possible with the addition of extracts of other thujone containing plants (e.g. *Thuja occidentalis* L., *Salvia officinalis* L.).

Even in the nineteenth century inferior and falsified products were known.<sup>21</sup> Instead of the traditional production by distillation, absinthe could be made using herbal essences.<sup>38–41</sup> So-called absinthe extracts were also commercially available at that time, which had only to be dissolved in alcohol and colored with food dye.<sup>39</sup> As food adulteration, the light green color of chlorophyll was sometimes enhanced with copper sulphate, copper acetate, indigo, aniline green, curcuma or extracts of spinach.<sup>8,22</sup> Another form of adulteration was the addition of antimony chloride to generate the clouding which arises in the dilution process with water.<sup>22,24,35,44</sup>

But the general problem at that time was that heads and tailings, which were separated from the product fractions during the distillation by legal manufacturers, were purchased by illegal manufacturers and used as a main component for the adulterated products. Even inedible mixtures of methanol and fusel alcohol could be made drinkable by masking the bitter wormwood taste.<sup>19,35</sup>

### TYPES OF ABSINTHE AND THE DRINKING RITUAL

Traditionally in the class of premium grade absinthe, it is distinguished according to the alcoholic strength, between "absinthe ordinaire" with 45–50%vol, the "absinthe demi-fine" with 50–68%vol and the "absinthe fine" with 68–72%vol. The "absinthe suisse" (68–72%vol) showed the highest quality and consists of pure herbal distillate, while in case of the other types the distillate was diluted with ethyl alcohol.<sup>22,38–41</sup>

Nowadays, this type of quality grading is varied and the majority of the products are sold with 70 or 55%vol. In comparison to other spirit drinks, the elevated alcoholic strength is necessary to enhance the solubility of the lipophilic essential oils.

For the purpose of drinking, a dilution is necessary developing itself as a part of a drinking ritual.

The increase of the water proportion liberates the herbal bouquet and leads to a clouding of the beverage and a separation of the solution. Within the drinking ritual, this observed effect is called "louche"-effect caused by the dilution of the absinthe with water.<sup>19,24</sup> The traditional and historic ritual demands, that a perforated spoon with sugar should be held over a glass filled with cold absinthe while water is cast drop by drop over it (di-

lution approximately 5:1).<sup>20–22</sup> A Czech tradition demands that the sugar spoon should be dipped into absinthe, held over the glass while the sugar is ignited. First, the sugar drops into the glass and later the rest is stirred into the solution.

### POSSIBILITIES TO REDUCE THE THUJONE CONTENT

Today's manufacturers face the problem that they have to generate a distinctive wormwood taste, without exceeding the thujone maximum limit of 35 mg/kg. The selective enrichment of the bitter and flavor compounds, while keeping the thujone concentration low, was extensively investigated.<sup>45</sup>

Tegmeier et al.<sup>46</sup> compared a water extraction to an alcohol extraction method. By the percolation with water or alcohol (30%vol) no thujone could be extracted, because the solubility of thujone in water is poor. Only by the application of ethanol 90%vol, it was possible to extract 0.18 mg thujone per g wormwood herb. When the method of digestion with ethanol 30%vol was applied, 0.17 mg thujone per g wormwood herb could be extracted. The largest yields were obtained, whenever the macerate of the wormwood herb was distilled (0.24 mg thujone/g). The use of hot and highly concentrated alcohol for the extraction should therefore be avoided to obtain extracts with a low content of thujone. Because the percolation with pure water might lead to a loss of microbiological quality, the percolation with ethanol 30%vol is regarded as the method of choice. This method is described as being easy to handle and economic. Gambelunghe and Melai<sup>47</sup> verified these results. Wormwood macerated with ethanol 20%vol for 30 days contained only 0.2 mg/l of thujone, while the maceration of wormwood with ethanol 95%vol for 6 months contained 62 mg/l of thujone. The consequence for the absinthe manufactures is that traditional recipes and methods have to be modified, in order to avoid thujone contents, which exceed the limit. The maceration should be done with low concentrations of alcohol and the wormwood herb should be separated before the distillation.

A possibility for the continuation of traditional recipes is to remove the thujone from the wormwood herb before the maceration. Stahl and Gerard<sup>48</sup> observed, that the extraction with liquid or supercritical carbon dioxide provides a fast, selective and quantitative method for the separation of thujone from the wormwood herb. Absinthin, which is responsible for the high bitter value of wormwood, remains in the herb. It is therefore possible to generate nearly thujone free wormwood herb and to use it for the manufacturing of absinthe. However, the application of this method for the manufacturing of spirit drinks was never described.

The most elegant alternative to avoid the toxic thujone may be the use of thujone-free wormwood herb, which is available in certain cultivation areas,<sup>10,16</sup> and appears to be perfect for the use in the spirit drink producing industry. With those chemotypes, it would be possible to produce absinthe with wormwood quantities on the basis of the traditional recipes, without the manufacturer facing the risk of exceeding the thujone limit.



## ANALYTICAL METHODS

## Thujone

Historically applied methods for the determination of thujone in absinthe are based upon iodometric titration, color reactions or paper chromatography and sometimes provided only detection limits as high as e.g. 20 mg/l and were therefore unfit for the sensitive detection of small quantities.<sup>49–52</sup> At the beginning of the nineteenth century the most modern methods were based upon the reaction of thujone with sodium nitroprusside, sodium hydroxide and acetic acid and provided a limit of detection of 5 mg/l.<sup>53–56</sup> However, this color reaction was highly unspecific and therefore other essential oils, aldehydes, and ketones led to a similar reaction as thujone. Even by the use of improved sample preparation, it was not possible to avoid these interferences. A positive reaction in case of the thujone analysis could not automatically be interpreted in such a way as to say that the spirit drink in question was made with wormwood herb. However, a negative result was regarded as a proof for the absence of wormwood oil.<sup>53,54</sup>

The sensitive and selective determination of thujone in spirit drinks was only made possible by the use of modern chromatographic methods. The majority of these methods described in the literature, for the quantitative determination of thujone in absinthe, use gas chromatographic separation (Table 1).<sup>8,47,57–65</sup> Besides, a qualitative screening method using thin layer chromatography (TLC)<sup>66</sup> and liquid chromatographic methods with fluorescence detection<sup>67,68</sup> were described. Already in 1976, a gas chromatographic method (GC) with flame ionization detector (FID) for the determination of thujone in al-

coholic beverages was developed by Mérat et al.<sup>57</sup> The sample preparation included a distillation of the samples and a following extraction (Table 1). Similar GC-FID methods were proposed by the "International Organization of the Flavor Industry"<sup>59</sup> and in the Manuel Suisse des Denrées Alimentaires (MSDA).<sup>63</sup>

The first gas chromatographic method with mass spectrometric detection (GC/MS) was described by Galli et al. in 1984.<sup>58</sup> The samples were neutralized and extracted with diethyl ether. The organic extract was concentrated and directly injected into the gas chromatograph. Tetraline was used as an internal standard, because it shows an intensive molecular peak in the mass spectra and possesses similar retention time as the analyte.

Sample preparation has an essential influence on the selectivity of the analysis. Adam and Postel<sup>60</sup> observed that bitter liqueurs can contain up to 200 different components after the preparation by distillation. It can happen that other components can co-eluate with  $\alpha$ - and  $\beta$ -thujone, pretending higher contents if GC-FID is used. Emmert et al.<sup>69</sup> described that even after preparation by solid-phase extraction and following gas chromatographic separation, the peak of linalool could interfere with the  $\alpha$ -thujone peak. It is therefore not advisable to do without mass spectrometric detection. Przyborski et al.<sup>61</sup> as well as Rapp et al.<sup>62</sup> were able to lower the limit of detection 200–250 fold to 0.005 mg/l or 0.002 mg/l respectively by liquid-liquid extraction, especially in comparison to the direct injection of the beverage (Table 1).

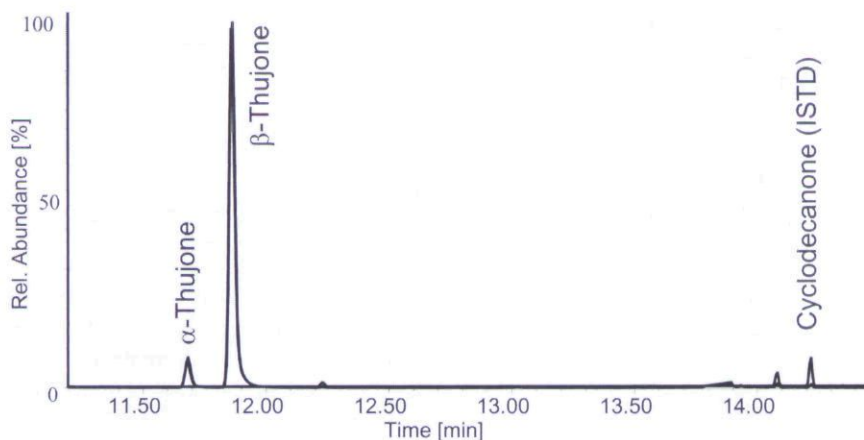
The sample preparation is in parts of extremely high complexity. Therefore Lang et al.<sup>8</sup> proposed the use of a headspace (HS) method, which avoids an extensive sample prepara-

**Table 1** Overview about methods for the determination of thujone in spirit drinks

Sample preparation	Method	Internal standard	Limit of detection [mg/l]	Ref.
Distillation, LLE (diethyl ether: pentane 2+1, v/v)	GC-FID	n-Tridecan	0.01	57
LLE (diethyl ether)	GC-MS	Tetralin	n.d.a.	58
Distillation, LLE (isooctane)	GC-FID	2-Decanone, Methyl-undecanoate	n.d.a.	59
LLE (Pentane: Dichloromethane 2+1, v/v)	TLC	—	0.28	66
Distillation	GC-FID	—	0.01	63
Distillation	GC-FID, GC-MS	Pelargonic acid methyl ester	GC-FID: 0.05 GC-MS: 0.01	60
Direct or LLE (Pentane: Dichloromethane 2+1, v/v)	GC-MS	Pyrogallol tri-methyl ether, iso-Safrol	Direct: 1 LLE: 0.005	61
LLE (kaltron)	GC-FID, GC-MS	2,6-Dimethyl-5-hetpen-2-ol, Decanol-3, Undecanol-1	Direct: 0.5 LLE: 0.002	62
Dilution	HPLC	—	0.5	67
SPE (C18)	HPLC	Dansylhydrazine	n.d.a.	68
Headspace-GC	GC-FID	Pelargonic acid methyl ester	0.2	8
LLE (Diethyl ether)	GC-MS	n.d.a.	n.d.a.	47
SPE (C18)	GC-FID GC-MS	Standard Addition	0.1	69
HS-SPME	GC-MS	Cyclodecanone	0.00071	64,65

LLE: liquid-liquid-extraction, GC: gas chromatography, FID: flame ionization detector, MS: mass spectrometry, n.d.a.: no data available, TLC: thin layer chromatography, SPE: solid-phase extraction, HS-SPME: headspace solid-phase microextraction.





**Figure 5** Characteristic HS-SPME/GC/MS-SIM-chromatogram of authentic absinthe sample (55%vol) containing 3.3 mg/l of  $\alpha$ -thujone and 46.7 mg/l of  $\beta$ -thujone; method according to Ref.<sup>65</sup> (thujone:  $m/z$  152, 110, 92; cyclodecanone:  $m/z$  154, 111, 98).

tion. Samples are diluted to the alcoholic strength of 15%vol and are diluted 1:5 with ethanol (15%vol). This solution is used directly for the headspace gas chromatographic analysis. The limit of detection is 0.2 mg/l, which is comparatively high but is sufficient for the control of the limit of 35 mg/kg.

The headspace solid-phase microextraction (SPME) can be used as an alternative method for the sample preparation. This method has proven itself in the analysis of terpenoids.<sup>70</sup> A totally automated HS-SPME method with cyclodecanone as internal standard, was proposed by Kröner et al.<sup>64</sup> The absorption of thujone was achieved by a polydimethylsiloxan-coated SPME-fibre, which is exposed to the headspace above the sample for the reason of enrichment of the analyte. After this solvent free extraction, desorption is achieved by the penetration of the fiber into the hot injector of the GC/MS. For the simultaneous determination of  $\alpha$ - and  $\beta$ -thujone in bitter spirit drinks, a limit of detection of 0.7  $\mu\text{g/l}$  was achieved. The high sensitivity of the HS-SPME/GC/MS method even allows a determination of thujone in blood samples.<sup>65</sup> Matrix effects are diminished by the extraction out of the headspace. A typical HS-SPME chromatogram of an absinthe sample is given in Figure 5.

### Absinthin

A method of determination for the flavor dominant bitter compound absinthin in absinthe spirit drinks has yet to be developed. TLC and photometric methods for the determination of artabasin and absinthin used for the identification of wormwood herb are summarized by Schneider and Mielke.<sup>17,18</sup> Another method is described in the European Pharmacopoeia.<sup>71</sup> By comparing TLC and HPLC profiles, the origin of wormwood extracts could be determined by Yashiro et al.<sup>72</sup> as the aerial parts of *Artemisia absinthium*. Modern instrumental analytical methods for the determination of bitter compounds in the spirit drink itself are currently in development.

### Alcohol and Volatile Congeners

Within the work of the official food control the determination of alcoholic strength is the most important parameter in the examination of spirit drinks. The CVUA Karlsruhe, part of the German official food control system, had to object 15% of all examined absinthe samples because of a too low alcoholic strength. For the analysis of alcohol either the reference method (distillation, followed by pycnometric determination of the density)<sup>73</sup> or a recently developed screening method can be used.<sup>74</sup>

In the nineteenth century, the alcohol quality of absinthe was determined using a colour reaction of fusel oil with salicylaldehyde and sulphuric acid.<sup>75</sup> Today, higher alcohols and by-products of the fermentation are usually determined by gas chromatography with flame ionisation detector. A distillation can be used as a sample preparation or the samples can be analysed by Headspace GC.<sup>76</sup>

Skopp et al.<sup>77</sup> investigated the alcoholic congeners of 56 absinthe brands and reported that these spirit drinks were predominantly made on the basis of ethyl alcohol of agricultural origin according to the European Regulation 1576/89.<sup>78</sup> Further, a certain country specificity was observed. In the absinthe of Austrian, Swiss, or Spanish origin no other congeners but methanol were detected, while the brands from Bulgaria were very rich in congeners. Seven of the 56 analyzed absinthe brands contained exclusively ethanol, while the other samples contained at least methanol as a congener. Only 10 brands contained additional congener alcohols. All of them contained iso-butanol. 1-propanol and 2-methylbutanol-1 were detected in 7 types, 3-methylbutanol-1 in 8 types. Butanol-2 was not detected in any of the samples, while butanol-1 was detected in one absinthe of Czech origin.<sup>77</sup>

### Use of Artificial Food Dyes

To determine artificial food dye in absinthe within the scope of the official food control is of special importance (see section



absinthe production). Own tests showed that some samples, especially of Czech or Spanish origin lacked the necessary labelling of the use of dye. In single cases, the products contained a different dye from the one indicated on the labelling. Until now, 41% of all absinthe samples, which were tested at the CVUA Karlsruhe, were objected, because of deficiencies in the dye labelling. The colouring is usually analysed by TLC and confirmed by UV/VIS spectroscopy.<sup>76</sup>

## TOXICOLOGY OF ABSINTHE

The chronic abuse in the zenith of absinthe in the nineteenth and twentieth century was made responsible for a syndrome called "absinthism" and was described to cause the following symptoms: after consuming absinthe, at first the well-being had been stimulated, later hallucinations had arisen followed by a depressive phase. Prolonged drinking of absinthe had caused convulsions, blindness, hallucinations, and mental deterioration. In the advanced state, signs of degeneration could be observed, which could cause convulsions that even resulted in death.<sup>20-22,24,79-81</sup>

For quite a long time, thujone was held responsible for absinthism. Today's point of view is more differentiated. Other factors or an interaction of different factors must be held responsible for causing absinthism.<sup>82</sup>

In the opinion of Strang et al.<sup>83</sup> absinthism can be traced back to a chronic alcohol intoxication causing similar symptoms. Pollmer<sup>84</sup> describes the adulteration of absinthe with toxic plants, such as sweet calamus (*Acorus calamus* L.) or tansy (*Tanacetum vulgare* L.), or adulteration with antimony chloride and copper sulphate as a possible cause of absinthism. Especially copper, whose resorption is amplified in combination with a massive alcohol ingestion, may cause alcoholic cirrhosis in alcoholics.<sup>84</sup> Also the use of inferior alcohol should be kept in mind as a possibility, because it explains symptoms such as impaired vision. The symptoms having been reported about van Gogh could also be explained by a digitalis poisoning, or the abuse or accidental ingestion of solvents or turpentine.<sup>23,31,85</sup> The possibility that van Gogh's xanthopsia may be caused by santonin from *Artemisia pontica* L. was already dismissed by Arnold et al.<sup>86</sup>

The thujone content of historic absinthe is largely unknown and speculated to be as high as 260 mg/kg.<sup>45,82</sup> Hutton points out that the thujone content could be overestimated because of the insufficient analytical methods that were available at the time.<sup>82</sup> Wilson estimated in 1936 that absinthe made from essences contained 1.8 to 45 mg/l, and absinthe made with wormwood herb contained 2 to 34 mg/l of thujone.<sup>55</sup> In recent analyses, historic absinthes contained less thujone than current products (Table 2), which validated Hutton's hypothesis. Schaefer et al.<sup>87</sup> found so low thujone concentrations analysing a legal French absinthe dating of 1904 that the authors even proposed the "toxicological rehabilitation" of absinthe.

The German federal institute for risk assessment<sup>88</sup> holds the view that, even if the legal limit of 35 mg/kg is significantly

**Table 2** Thujone contents of historic absinthes

Absinthe	Thujone [mg/l]	Year of analysis	Method	Ref.
French 1904	<0.01	1994	GC	87
Pernod fils circa 1900	6	2002	GC	82
Pernod Tarragona 1930	1.8	2004	GC/MS	69

exceeded, the consumer does not ingest health threatening amounts of thujone. Because of the high alcoholic strength it is advised against a continuous and massive consume. Threatening thujone concentration can only occur if beverages are made following recipes retrieved from the Internet, which suggest the use of huge amounts of wormwood oil. Some absinthe types with a content of 100 mg/kg thujone were legally available in the Czech Republic until its recent integration into the EU. Weisbord et al.<sup>89</sup> described a case of accidental thujone intoxication in a man after having consumed a large dose of wormwood oil under the erroneous belief it had been absinthe. The symptoms were seizures, rhabdomyolysis, and acute renal failure. The wormwood oil was ordered from a website that sold essential oils for aromatherapy. This inadequate control of access to potentially unsafe herbal products is seen as an unacceptable health risk to consumers.<sup>90</sup>

In comparison to  $\beta$ -thujone,  $\alpha$ -thujone is believed to be 2.3fold more toxic.<sup>91</sup> Until today, only little valid data is available concerning the effect of  $\alpha$ -/ $\beta$ -thujone, especially in regard to the influence on the central nervous system after absinthe consumption. A recent study of Dettling et al.<sup>92</sup> showed that the administration of alcohol containing a high concentration of thujone (100 mg/l) had a negative effect on attention performance. When the subjects were under the influence of alcohol or were administered both alcohol and low thujone concentrations (10 mg/l), these effects were not observed. Similarly, it was found that only high concentrations of thujone can temporarily counteract the anxiolytic effect of alcohol.

It is possible to explain these effects because of an interaction of  $\alpha$ -thujone with  $\gamma$ -amino butyric acid (GABA) dependent chloride channels<sup>91,93-95</sup> as well as a 5-HT<sub>3</sub> receptor activity<sup>96</sup> was demonstrated.

Interestingly, the activation of human bitter taste receptors by  $\alpha$ -thujone was recently proven by Behrens et al.<sup>97</sup> and it was found that the receptor is sufficiently sensitive to serve as protection against the ingestion of toxic amounts of this substance.

Another hypothesis is that because of structural similarities between thujone and tetrahydrocannabinol (THC), the psychoactive component of the hemp plant *Cannabis sativa* L.,<sup>98</sup> both substances might activate the same receptor in the central nervous system.<sup>99</sup> This theory could not be proven in experiments.<sup>100</sup> Nevertheless absinthe is advertised on the Internet as having a cannabis-like effect. In some recent literature this thesis is also misleadingly proclaimed.<sup>101,102</sup>

The sometimes observed porphyrinogenic effect of thujone and other terpenoids is explained with the pathway of metabolism by the hepatic cytochrome P-450 system.<sup>95,103,104</sup>



Very few data exist about the pharmacology of thujone. Max<sup>105</sup> pointed out that the typical 2–4 mg of thujone, which were consumed per drink were far below the level at which acute pharmacological effects are observed. This is confirmed by Hinkelbein,<sup>106</sup> who states that by the consumption of absinthe, up to a blood alcohol concentration of 2.5 g/l, approximately 3.5 mg of thujone are ingested (0.005 mg/kg bodyweight). In this order of magnitude it is highly improbable that central effects can be caused by thujone.

A pilot drinking study by Kröner et al.<sup>65</sup> resulted in high blood alcohol concentration, but as expected no thujone was detected. The probands examined did not show a central effect caused by the terpenoids besides the effect of the alcohol. Therefore, the hallucinogenic potency of absinthe can be neglected, if the EU limit is obeyed. The feared return of absinthism, proclaimed by Hein et al.,<sup>20</sup> Holstege et al.<sup>24</sup> and Müller<sup>101</sup> is therefore exaggerated. The effects of the recent types of absinthe are predominantly caused by the naturally high alcoholic strength (>50%vol), although it is possible to reach effective thujone blood levels, if illegally produced and distributed absinthe is ingested.

### QUALITY CONTROL OF ABSINTHE

Absinthe is neither defined in the German definitions of spirit drinks,<sup>107</sup> nor in the regulation of the EU concerning spirit drinks.<sup>78</sup> This causes problems in the food chemical and legal judgement for the official food control. Only the Manuel Suisse des Denrées Alimentaires (MSDA)<sup>63</sup> provides specifications for absinthe, regarding its thujone content. Referring to this definition, a typical absinthe does not contain more than 15 mg/l of thujone. A content between 2 and 15 mg/l hints towards the use of wormwood. Contents below 2 mg/l can be neglected. However, in the context of the different chemotypes of wormwood and the various possibilities of production this grading after the MSDA appears to be a bit oversimplified. From our experience, there exists a correlation between the quality and the thujone content of absinthe. But especially absinthes regarded as to be of high-grade show a very large concentration range. Inferior products, made only by maceration, can contain more thujone than distilled ones. The MSDA's criteria is valid in regard to absinthe that contains less than 2 mg/l of thujone and the question arises if such a product contains wormwood at all. An official objection on the single reason of a low or missing thujone content is seen as critical, because the presence of a potentially toxic substance cannot be made a requirement. Until more parameters for the authentication of absinthe are available, the content of thujone can only give a hint. An objection by the official food control department should always be based on further characteristics (such as organoleptical testing).

A further definition for absinthe, which may be used nowadays, was provided by the Swiss Law for the prohibition of absinthe. According to this, every spirit drink without regard to the method of production that contains aromatic compounds

of wormwood herb in combination with other aromatic compounds derived from plants such as anise and fennel, is defined as absinthe.<sup>108</sup> At that time, thujone was also regarded as being the determining factor amongst the aromatic compounds.<sup>109</sup>

As a further hint for the quality control of absinthe, it was proposed to check the ratio of the thujone isomers, because this allows conclusions concerning the origin of the thujone detected.<sup>8</sup> Thujone of wormwood origin usually can be recognized by a predominant  $\beta$ -thujone content. In other cases it has to be distinguished between seldomly found  $\alpha$ -thujone chemotypes of wormwood and a food adulteration with cedar or other thujone containing plants.

Table 3 presents the results of four recently conducted studies of the thujone content of commercially available absinthe and includes own results determined by the CVUA Karlsruhe.<sup>8,65,69</sup> In conclusion, it can be noted that the majority of the examined samples (95%) did not exceed the thujone EU maximum limit of 35 mg/kg. Strikingly, more than half of the examined samples (55%) contained less than 2 mg/kg thujone.

Because 5% of the examined samples, which are actually available on the market exceeded the legal thujone limit, the analytical determination of thujone should still be a permanent parameter of the official food control. Most of the absinthe manufacturers and suppliers advertise the proclaimed thujone content of their products on their Internet pages. Slogans such as "it contains the maximum allowed thujone concentration of 35 mg/kg" should be critically judged by the appropriate authorities. If higher thujone contents are proclaimed than can be detected, these absinthes should be objected as a deception of the consumer.

Besides thujone, further characteristic compounds of the essential oil of wormwood (e.g. linalool, fenchone, sesquiterpenes) may be used for the authentication of absinthe. Other markers might be the bitter components or characteristic substances like tetrahydrofuran lignanes, flavonic glucosides or oligo saccharides.<sup>3</sup> There is a great need for further research in the field of identification and characterization of typical marker components of wormwood. In doubtful cases, the official food control should take samples of the wormwood plant used for the production of absinthe and control the recipe, in regard to its wormwood content.

### MINIMUM REQUIREMENTS FOR ABSINTHE

In a recent study of our group, the homepages of manufacturers and suppliers of absinthe were analyzed, with the intention of providing a statement about the actual concept of commerce for the spirit drink.<sup>1</sup> In 2003, 89 absinthe brands were distributed in Germany via the Internet. Most brands are made in the Czech Republic, Spain, France, and Germany. The alcoholic strength varies between 30 and 90%vol. 39 brands contain a declaration about wormwood or bitter taste as well as a clouding with water. The predominant part of the products provided a corresponding taste (90%) and clouding (77%). The classic definitions, which



**Table 3** Classification of absinthe samples in regard of their thujone content

Total thujone content [mg/kg]	Lang et al. <sup>8</sup> (n = 30)	Kröner et al. <sup>65</sup> (n = 14)	Emmert et al. <sup>69</sup> (n = 16)	CVUA Karlsruhe (n = 87)	Sum (n = 147)	%
< 2	16	5	7	53	81	55
2–10	9	3	5	17	34	23
10–35	2	3	4	16	25	17
> 35	3	3	0	1	7	5

state that absinthe contains aromatic compounds of the wormwood plant (*Artemisia absinthium* L.)<sup>6,38–41,108</sup> are still valid in case of the nowadays purchasable products and can therefore, still be used for their evaluation. If the sensory testing for wormwood is negative and neither thujone nor absinthin are detectable, the recipe should be controlled on the spot, while conducting a manufacturing control. It has to be demanded, that every product has at least a "louche"-effect and certain wormwood content. The color of most products is between yellow-green and olive, or clear. Some more exotic types show an unusual red color. In case of 45 products, the origin of the color is labelled and 60% of them are made without artificial dye. Especially the Czech products differ from the ordinary absinthe out of other countries (Switzerland, France and Germany). After the addition of water, no milky clouding arises and the characteristic bitter taste of wormwood extracts is not present. Instead, a sweet mint taste can be observed. The color of these products is turquoise, which is highly unusual for an absinthe and which is the result of the use of artificial dye.

According to the statement of an importer, the described composition is characteristic for a Czech absinthe and is complying with the local concept of commerce. It is the opinion of the authors, that this deviation at least should be made recognizable.

The overview of the market of absinthe provided information about the concept of commerce, from which the minimum requirements for an absinthe are derived and given in Table 4. The result of the sensory testing should be a recognizable wormwood flavor and bitter taste. After dilution with water, the characteristic clouding should arise. Products, which are advertised as being of premium grade (e.g. "made after historic recipes"), should be made by distillation, should have an alcoholic strength of at least 45%vol, and should not contain artificial dye.

**Table 4** Minimum requirements of absinthe

Minimum requirements
<ul style="list-style-type: none"> <li>• Characteristic aromatic flavour and bitter taste caused by natural extracts or distillates of wormwood (<i>Artemisia absinthium</i> L.)</li> <li>• Colour: uncoloured or greenish</li> <li>• Characteristic clouding if diluted with water ("louche-effect")</li> <li>• Standard chemotypes: <math>\beta</math>-thujone &gt; <math>\alpha</math>-thujone</li> </ul>
Further requirements for premium grade products
<ul style="list-style-type: none"> <li>• No artificial dye (colouring achieved only with wormwood and other herbs)</li> <li>• Distillative manufacturing</li> <li>• Minimum alcoholic strength 45%vol</li> </ul>

On the background of the high number of deteriorated products found on the market, the initiative of Switzerland for the special labelling of authentic products would be very welcome to facilitate the consumers' decision.

## CONCLUSION

The over 100 cited references show a great difference in scientific quality. Some papers should focus more on real analytical and toxicological facts instead of resuming all the factoidal myths and legends of former years. However, the authors wanted to give a complete guide to absinthe and let the knowledgeable reader judge on his own.

In our opinion, absinthe poses less of a problem for health protection of the consumers than initially assumed. Absinthe is extensively controlled by the official food control and the thujone limit is obeyed by most products. Even when it slightly exceeds the limit, no toxicological effects can be expected. Therefore, absinthe is not a problem because of the thujone content, but like all spirit drinks because of the possible excessive alcohol consumption. Besides this, the current major problem in the evaluation of absinthe is the protection of the consumer against deception from deteriorated products without wormwood, which are sold at exaggerated prices. Because of a missing official definition of absinthe, a legal uncertainty exists, which is exploited by many manufacturers for the distribution of inferior products. To protect the consumer from deception and in the interest of a standardized evaluation, absinthe should be uniformly defined within the EU. In the opinion of the authors, it is a mandatory demand that real absinthe must be produced with wormwood herb using maceration and distillation. Regrettably, a proposal for such an Europe-wide regulation initiated by Germany has recently failed.<sup>8</sup>

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