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Fingerprint analysis of thermolytic decarboxylation of tryptophan to tryptamine catalyzed by natural oils

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ABSTRACT

A number of *N*,*N*-dialkylated tryptamines show psychoactive properties in man which resulted in a renewed interest in psychopharmacological research. Attempts to manufacture these derivatives are increasing within a clandestine environment, where literature procedures are adapted and information is exchanged on the Internet. One such example is based on the thermolytic decarboxylation of tryptophan to tryptamine as the precursor to psychoactive derivatives. This procedure was proposed to make use of household solvents such as *turpentine substitute* and *white spirit* to facilitate decarboxylation. Discussions on websites also suggested the catalytic use of natural oils in order to accelerate these reactions. In this research, the analytical characterization of this preparation procedure was carried out using gas chromatography–ion trap single and tandem stage mass spectrometry in electron and chemical ionization mode that led to the identification of previously unreported 1-mono and 1,1-disubstituted tetrahydro-β-carboline (THBCs) by-products. The tryptamine product and several THBC by-products were determined quantitatively and a "fingerprint" analysis of the crude products allowed for the differentiation between the essential oil catalysts involved as indicated by the presence of tetrahydro-β-carbolines and their imine intermediates.

1. Introduction

The analytical characterization of impurities in pharmaceutical drugs and natural products is of fundamental importance in order to establish the application of proper quality control procedures [1].

The tryptamine category of psychoactive drugs is based on the presence of the 2-(1*H*-indol-3-yl)ethanamine nucleus (**1**, Fig. 1A). Biological activity derives from structural similarity to the neurotransmitter serotonin (5-hydroxytryptamine) and the substitution pattern determines the extent of biological activity. *N*,*N*-Dialkylation, for example, is found in a large number of tryptamine derivatives with psychoactive and hallucinogenic properties [2]. Naturally occurring *N*,*N*-dimethyltryptamine (DMT) can be classified as a classical representative of a hallucinogenic tryptamine [3] that is also accessible by a large number of synthetic routes [4,5]. Shulgin and Shulgin have published the preparation and psychoactive properties of a large number of tryptamine-based drugs [6]. The availability of these data on the Internet, and the fact that hallucinogens such as D-lysergic acid diethylamide (LSD) are less commonly available, has resulted in the increased popularity of tryptamine derivatives [7].

Clandestine drug synthesis [8,9] is based on the attempt to prepare such derivatives, and the fact that most of them are prohibited by legislation often places restrictions on commercial availability of starting materials and reagents. Access to a variety of Internet sources enables one to gain important information on the properties and potential dangers associated with the consumption of illegal drugs. Synthetic procedures are also often discussed and experiences are exchanged about the application of either established or newly developed or improvised synthetic procedures. Analytical profiling and fingerprinting of synthetic routes to illegal tryptamines has hitherto not been carried out in detail. Therefore, the present work aims to provide the forensic and clinical/medical community with basic information on the nature of these drugs and likely side products [4,5,10].

A two-step procedure called *The Breath of Hope* synthesis was proposed on Internet websites for the synthesis of DMT. It was suggested to employ the widely available amino acid tryptophan (Trp) as the starting material. In the presence of high boiling solvents

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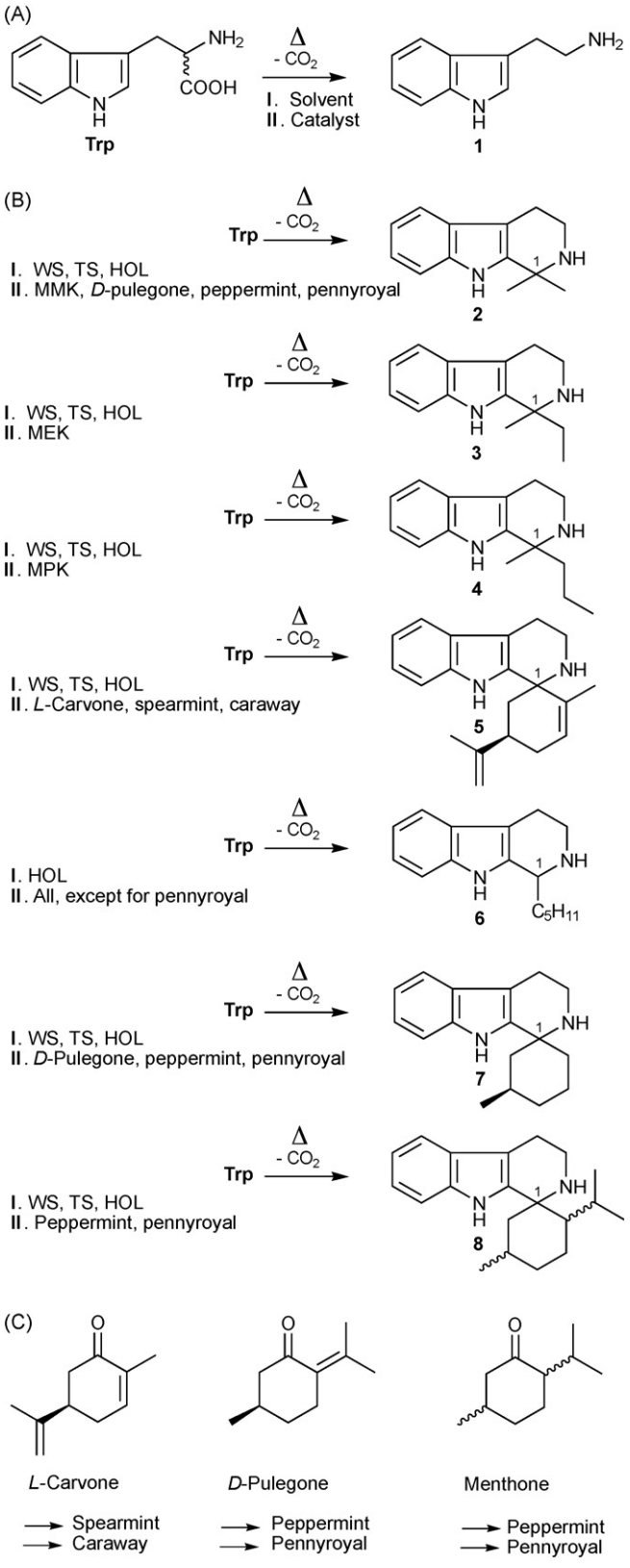

Fig. 1. (A) The synthesis of tryptamine *via* the thermolytic decarboxylation of D,L-tryptophan. (B) Different combinations of catalysts and solvents led to the detection of several tetrahydro-β-carboline (THBC) side products. (C) The main constituents of natural oils which were employed as catalysts in order to accelerate decarboxylation to tryptamine **1**. The detection of THBCs **5**, **7** and **8** confirmed their involvement in by-product formation. WS: white spirit. TS: turpentine substitute. HOL: hexan-1-ol.

and ketone catalysts, heating at reflux was proposed to generate tryptamine **1** as the intermediate, followed by the synthesis of DMT using methyl iodide and a phase transfer catalyst under alkaline conditions [11].

Previous research in this laboratory revealed that the decarboxylation of Trp indeed produced tryptamine **1** as suggested by various clandestine websites. Significant amounts of side products however, were detected and subsequently characterized as tetrahydro-β-carboline derivatives (THBCs) [12]. The overall physiological effects of these THBCs products are difficult to predict and further toxicological and biological activity studies are warranted. A potential action might be on monoamine oxidase (MAO) enzymes that metabolize hallucinogenic tryptamines. In this regard, it was assessed if the THBCs by-products (**2**–**4**) identified and isolated from tryptamine preparation reactions are inhibitors of MAO, by following a recently reported method [22]. In recent work done by the authors, these compounds were not good inhibitors of MAO, as compared to simple aromatic β-carbolines.

The presence of these THBCs impurities yielded a useful profile for the identification of the synthetic pathway because it transpired that both solvent (cyclohexanol) and ketone catalysts caused their formation, possibly due to the involvement of a Pictet-Spengler mechanism [12].

The present study explored the fact that pure solvents and reagents are often expensive and difficult to obtain within a clandestine environment, which leads to alternative methods on the Internet for the thermolytic decarboxylation of Trp. It was suggested to employ commonly available and inexpensive household products [13]. This work addressed the use of household solvents such as *turpentine substitute* (TS) and *white spirit* (WS), as well as the use of natural oils as catalysts for the decarboxylation of tryptophan. Fingerprinting and quantitative analysis were carried out using gas chromatography–ion trap tandem mass spectrometry (GC–IT-MS–MS) in electron (EI) and chemical ionization (CI) mode.

2. Experimental

2.1. Chemicals

1-Hexanol (98%) was obtained from Aldrich (Poole, UK). *TS* and *WS* were purchased at local stores. Spearmint oil (from *Mentha spicata* L.) and D,L-tryptophan (Trp) were obtained from Fluka (Poole, UK). Peppermint, caraway and pennyroyal oils were also purchased locally. The ketones L-carvone (98%), pentan-2-one (MPK) (99.5%), D-pulegone (85%), butan-2-one (MEK) (>99%), acetone (MMK) (99.5%) were obtained from Aldrich. Silica gel for flash chromatography (particle size 40–63 μm) and silica gel aluminium TLC plates were obtained from VWR (Lutterworth, UK). All other solvents and reagents were analytical grade from Aldrich. Tryptamine **1** and 5-methoxytryptamine (I.S.) were also obtained from Aldrich. 1,1-Dimethyl-tetrahydro-β-carboline **2**, 1-methyl,1-ethyl-THBC **3** and 1-methyl,1-propyl-THBC **4** were available as standards from previous work [12].

2.2. Synthesis of tetrahydro-β-carboline by-products ***5*** and ***6***

Synthesis of THBC **5** was achieved by adapting an iodine-catalyzed Pictet-Spengler reaction [14]. Tryptamine (200 mg, 1.25 mmol) was dissolved in 10 mL ethanol, followed by the addition of L-carvone (188 mg, 1.25 mmol) and a catalytic amount of iodine. The reaction mixture was left stirring for 20 h at room temperature. The resulting product was characterized directly by GC–MS analysis.