Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Pharmaceutical and Biomedical Analysis 36 (2004) 675–691
JOURNAL OF PHARMACEUTICAL AND BIOMEDICAL ANALYSIS
www.elsevier.com/locate/jpba

Review

An analytical perspective on favoured synthetic routes to the psychoactive tryptamines

Simon D. Brandt[a], Sally Freeman[b], Peter McGagh[a], Norliza Abdul-Halim[a], John F. Alder[a,*]

[a] *Department of Instrumentation and Analytical Science, Institute of Science and Technology, UMIST, P.O. Box 88, Manchester M60 1QD, UK*
[b] *School of Pharmacy and Pharmaceutical Sciences, University of Manchester, Oxford Road, Manchester M13 9PL, UK*

Received 30 July 2004; received in revised form 18 August 2004; accepted 19 August 2004

Abstract

Many tryptamine derivatives are known to induce altered states of consciousness and are increasingly of interest in forensic and neurobiological studies. The analytical chemistry of certain synthetic routes to the tryptamines is discussed and likely side products and impurities identified, where literature reports are available. Recent examples from the authors' laboratory are presented to highlight future prospects and implications for analytical procedures. The aim of this review is to provide the analytical chemist with the foundation chemistry and some analytical targets to be able to undertake direct characterisation of products and intermediates. These might become available from interdiction of clandestine operations in a forensic environment or during the synthesis of the tryptamines for investigative neurobiological and clinical procedures.
© 2004 Elsevier B.V. All rights reserved.

Keywords: Tryptamines; Forensic studies; Neurobiological procedures; Analytical chemistry

Contents

* Corresponding author. Tel.: +44 161 200 4885; fax: +44 161 200 4881.
E-mail addresses: sally.freeman@man.ac.uk (S. Freeman), fred.alder@umist.ac.uk (J.F. Alder).

0731-7085/$ – see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.jpba.2004.08.022

1. Introduction

The tryptamines and related compounds derived from substituted indoles play a fundamentally important role in human existence. 5-Hydroxytryptamine, serotonin, is one of the most important signalling hormones in the body and influencing its receptor sites, the 5-HT receptors [1], has a dramatic effect on our perception of reality. Since ancient times, societies have used naturally occurring tryptamines and related compounds for their psychotropic effects, imparting altered states of consciousness for religious and recreational use. In present society, state legislators have taken over control of possession and use of these materials with a more or less libertarian attitude, depending upon their particular national social mores.

Recent studies since the 1950s and a greater understanding of brain chemistry, has underlined the importance of tryptamines and their relatives, not only in the signalling processes as such but also in helping us to understand these processes and perhaps control or rectify unusual conditions that result in disorders such as schizophrenia or disease states such as Parkinson's and Alzheimer's.

One thing is abundantly clear from reading the literature, however, the subject is still only fragmentarily and poorly understood, because of the great complexity of the human brain. The chemical signalling compounds that control our brain processes influence many receptors of different types at the same time, and compete with other compounds to achieve that complex and transient equilibrium. Thus, the serotonin receptors 5-HT extend to more than a dozen types [1], with the 5-HT_{2A} being possibly, but not uniquely most important in influencing our perception of reality. The recent and detailed review by Nichols [2] on the pharmacology of hallucinogens, with particular reference to LSD and the simpler tryptamines, serotonin, dimethyl-, diethyl- and dipropyl tryptamines, psilocin (4-HO-DMT), 5-MeO-DMT and mescaline (3,4,5-trimethoxyphenethylamine) is an excellent and perceptive insight into the current understanding of these compounds in their influence on the brain.

The work reviewed by Nichols and his insight reveals clearly the imperfections in our understanding of the interactive processes going on in the brain. Factors include the rate of transfer across the blood–brain barrier, the rate of metabolism of the active agents by, for example, monoamine oxidases and even the knowledge of which are the active agents and the site of their interaction. LSD–tartrate has a significant hallucinatory effect on man from the level of ∼50 μg and yet recreational doses of tryptamines as reported by Shulgin and Shulgin [3] and on the internet, may be in the tens-milligram range. The influence of impurities in poorly purified drugs cannot therefore be discounted, particularly, with so little understanding of their modes of action. It is well documented that monoamine oxidase inhibitors (MAOI) affect the activity of these recreational drugs. It is less well known whether by-products such as the tetrahydro-β-carbolines are themselves psychoactive as well as being MAOI, or whether they and other substance act as potentiators for the principal drugs.

One predictable consequence of prohibition by legislation is the creation of a clandestine trade in the substances for which a market exists. The corollary of this is the inability to exercise quality control over the illegally prepared substances. Charitable, and more recently liberally minded state welfare authorities have introduced schemes for rudimentary on-site quality control of recreational drugs in a commendable effort to minimise death and injury due to impure illicit drugs. There is a powerful need for as long as the albeit illegal market is supplied with drugs prepared without quality control, to provide information about the principal drugs and their impurities to the clinical welfare and drug rehabilitation community. Equally so, the information will be invaluable to those studying the brain chemistry, in a domain where legal and ethical considerations make experimentation and human trials most difficult. As reviewed by Nichols, however, this may change due to recent developments in clinically relevant research areas where some of these compounds have found their niche as neurobiological tools [2]. Not least is the information valuable to police agencies, where chemical profiling of illegal drugs can lead to identification of the synthetic route, and potentially, the source and maker of the drugs.

The recent well-researched text edited by Laing [4] with contributions from recognised experts in this field, has given a comprehensive overview of the forensic aspects of the history, pharmacology and illegal manufacture of hallucinogens. It is noteworthy that whereas their review of the subject is expansive, the analysis of tryptamines and their derivatives is

little represented, principally because the topic has been only partly developed. Earlier work on the phenethylamine derivatives has demonstrated just how useful can be the analysis of by-products and impurities in tracing sources of illegal drugs. The aim of the present authors' research programme is to provide that missing information and capability to the clinical and forensic community to enable their research.

2. Chemistry of tryptamines

The chemistry of psychotropic alkylaryl amines was reviewed by Freeman and Alder [5]. Briefly, the principal structural feature that gives rise to the psychoactive (mood changing) effects of the tryptamines is the indolealkylamine moiety. A numbered structure of tryptamine is given in Fig. 1. The effect is maximal with ethyl and propyl as the side chain in the 3-indole position. Amine substitution with methyl, ethyl and propyl in any combination, modifies the effect of the substance, particularly, with respect to its oral activity. The C-2 and alkyl-*N*-substituted higher homologues of tryptamine are unaffected by monoamine oxidase. The hallucinogenic property of the substance is enhanced by *o*- and *p*-directors in the 6-carbon ring.

The methoxy group is most significant and the 4- and 5-positions of the indole nucleus are the most important [3]. Hydroxyl substitution in the ring gives unpredictable properties with some compounds being hallucinogenic, whilst others have no psychoactive activity. Halogen substitution in the 6-carbon ring may lead to greater biological activity in the tryptamines as it does in the phenethylamines.

A methyl on the α-carbon of tryptamine renders the amine orally active, probably by blocking the action of monoamine oxidase. There is some indication that methyl at the 2-indole carbon is acting likewise. Increased lipid solubility may also be a contributory factor by enhancing transfer across the blood–brain barrier membrane, thus 5-MeO-2,*N*,*N*-trimethyltryptamine is orally active [3]. Based on human self-experimentation, it has recently been argued, however, that all known psychoactive tryptamines, including 5-MeO-DMT and 5-HO-DMT (bufotenin), do possess oral activity with the exception of DMT [6–8].

In considering synthetic routes to the tryptamines, the chemistry of starting materials and intermediates needs also to be considered. This may permit prediction of by-products and impurities that could themselves be biologically active or psychoactive. The chemistry of substituted amines and derivatives, common precursors to indoles via the Fischer, Sugasawa and Bischler syntheses, and from substituted nitrobenzenes via the Leimgruber–Batcho and related syntheses, to name but a few [9], is beyond the scope of this review. Starting with substituted indoles in a pure form still offers considerable scope for complexity of products in reactions. Indole and 5-MeO-indoles are characterised by being electron rich at the 2- and 3-position and susceptible to electrophilic attack. This property forms the basis of most routes from indoles to the tryptamines but other processes may occur in parallel.

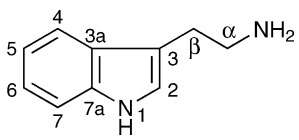

Fig. 1. Tryptamine molecule.

Although it is beyond the scope of this present paper to provide a detailed review on the chemistry of substituted tryptamines and derivatives, a few examples of common synthetic routes are used to highlight basic key elements that are relevant in the context of this review. Also, a certain insight into the complexity of side-product formation in tryptamine chemistry can be found in investigations where non-hallucinogenic tryptamine derivatives of commercial interest are involved, such as melatonin (5-MeO-*N*-acetyltryptamine), tryptophans or Sumatriptan (a methylsulfonamide derivative of DMT) and derivatives.

3. The Speeter and Anthony Route

The method of Speeter and Anthony [10] is still considered to be one of the most important preparative synthetic methods for psychoactive tryptamines (Fig. 2A) [5]. Acylation of a (substituted) indole (**1**) with oxalyl chloride yields the indole-3-glyoxalylchloride (**2**) that is reacted with an amine to give an indole-3-glyoxalylamide (**3**). The subsequent reduction with lithium aluminium hydride (LAH) produces the desired tryptamine (e.g. **4**). LAH is a powerful reducing agent [11] that usually enables a facile and complete reduction of these amides. Several exceptions, however, have been recognised depending on the reaction conditions. For instance, during an investigation of the synthesis of psilocybin and psilocin derivatives, it was noted by Troxler et al. that the reduction of 4-benzyloxyindole-3-yl-*N*,*N*-dimethylglyoxalylamide in THF also yielded a significant amount of the incompletely reduced β-hydroxy derivative (**5**). Complete reduction was then achieved in boiling dioxane [12]. This observation is rather interesting since these β-hydroxy derivatives are normally known to be synthesised by the reduction of *N*-1-substituted amides [13,14], which is attributed to the substituted indole nitrogen not facilitating elimination of the β-hydroxy group.

Interestingly, some β-hydroxylated tryptamines have been found to have significant pharmacological effects, such as hypotension [13–15]. The presence of β-hydroxylated by-products can contribute to the formation of dimeric compounds [13]. This is attributed to the instability of these compounds in the presence of aqueous acids where deep red solutions can also be observed. Literature search on the analytical chemistry of this issue revealed the presence of only one study by Crookes et al. where the synthesis of DMT **4** was investigated [16]. The reduction of indole-3-yl-*N*,*N*-dimethylglyoxalylamide **3** in THF was found to yield dimeric

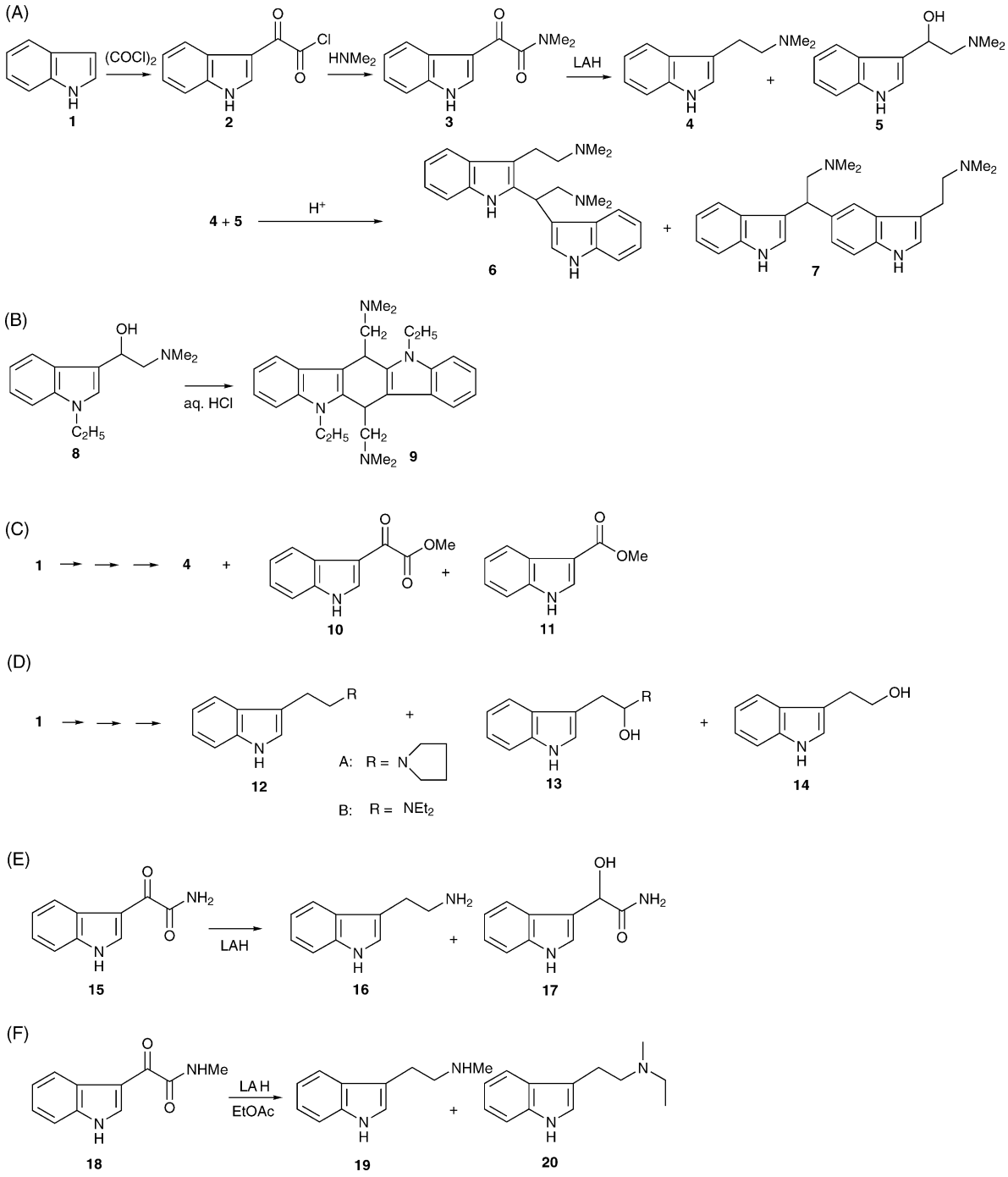

Fig. 2. The Speeter and Anthony route and some examples of the generation of side products. (A) Synthesis of dimethyltryptamine (**4**) and the formation of dimeric side products (**6** + **7**) during acidic workup [16]. (B) Compound **8** (*N*-1-ethyl-β-OH-DMT) after treatment with aqueous HCl where the observed dimeric product was speculated to be compound **9** [13]. (C) Two side products (**10** + **11**) have been characterized in the first step in the synthesis of DMT **4** [17]. (D) Synthesis of tetramethylene tryptamine, i.e. (3-[2-pyrrolidinylethyl]indole) **12A** and diethyltryptamine (DET) **12B**. Incompletely reduced side products have been identified (**13A** + **13B**). Tryptophol **14** was also identified [18]. (E) The reduction of an *N*-unsubstituted indole-3-yl-glyoxalylamide **15**, yielding tryptamine **16**, has been found to result in the formation of side product **17** [22]. (F) The reduction of indole-3-yl-*N*-methylglyoxalylamide **18** to *N*-methyltryptamine **19**. Quenching of the excess LAH with ethyl acetate (EtOAc) was found to result in *N*-ethylation that produced *N*-ethyl-*N*-methyltryptamine **20** [28].

products after acidic workup (Fig. 2A). A significant amount (8–10%) of β-hydroxy-DMT **5** was found after 1 h at reflux. This derivative was treated with a 9 M excess of DMT in methanol and an excess of 3 M aqueous HCl. Analysis revealed the presence of 76% of dimer **6** and 18% of 3:1 mixture of two other isomeric dimers [16]. The main component of this mixture was determined to be most likely dimer **7**. When *N*-1-ethyl-β-OH-DMT **8** was exposed to aqueous HCl, Heinzelman and Szmuszkovicz reported that the coloured, dimeric acid transformation product showed increased

hypotensive activity in hypertensive rats, when compared to the monomer [13]. Based on UV and IR data the structure of this dimer, formed by self-condensation, was speculated to be **9** but attempts to characterise this compound unambiguously failed (Fig. 2B) [13].

One of the few examples of forensic investigations on a synthetic route to a psychoactive tryptamine was published by Gielsdorf et al. [17] where DMT was synthesised by this synthetic route. Only few experimental and analytical details were given but two side-products were found by GC-EIMS during the first synthetic step. They were identified as indole-3-glyoxylic acetic acid methyl ester **10** and indole-3-carboxylic acid methyl ester **11** (Fig. 2C) based on the EI mass spectrum. This may indicate an involvement of methanol during the procedure. During the next step, a third compound was found but not identified [17]. Cowie et al., using TLC, NMR, IR and EIMS, synthesised tetramethylene tryptamine (3-[2-pyrrolidinylethyl]indole) **12A** and diethyltryptamine (DET) **12B**. Both approaches yielded the hydroxy-derivatives **13A** and **13B** but the position of the hydroxy-group was assigned to the α-carbon (Fig. 2D) [18]. This group also identified tryptophol (indole-3-yl-ethanol) **14** as an impurity and attributed its presence to the glyoxalylchloride **2**, being carried over to the reduction step [18]. Recent work in this laboratory identified the presence of 5-methoxytryptophol as an impurity during the synthesis of 5-methoxy-*N*,*N*-diisopropyltryptamine (5-MeO-DIPT) that resulted directly from the LAH reduction. The incompletely reduced hydroxy-derivative was also identified but the hydroxy group has been assigned to be on the β-position [19]. Interestingly, some 5-substituted tetramethylene tryptamine derivatives reappeared recently in a new series of tryptamines that show high affinity for cloned human 5-$HT_{1B/1D}$ receptors, which were investigated in connection with the treatment of migraine [20].

The LAH reduction of mono- or unsubstituted glyoxalylamides has been recognised to be more complicated, possibly due to the formation of complexes between LAH and the hydrogen atom(s) attached to the nitrogen atom [12,13,21]. For example, the so-called glycolamides **17** (β-hydroxyacetamides) have been found during the reduction of non-substituted glyoxalylamides (**15**) (Fig. 2E) as determined by melting point- and elemental analysis [22] and UV, IR and NMR, respectively [23]. Alemany et al. reported on unstable HBr salts of certain monoalkylated tryptamines and suggested the formation of certain mono- or polymeric decomposition products [24]. It has long been recognised that the decomposition of excess LAH at the end of the reduction can play a significant role in the presence of products and by-products. The use of certain esters, such as ethyl acetate (EtOAc), can result in the alkylation of the amine [25–27]. For example, *N*-methyltryptamine derivatives (**19**) have been found to be converted to *N*-ethyl-*N*-methyltryptamines (**20**) by the addition of EtOAc during the LAH reaction of amide **18** (Fig. 2F). Detailed analytical data were not given [28].

4. Routes from 3-(nitrovinyl)indoles

A common route of synthesis of psychoactive α-substituted tryptamine derivatives, such as α-methyltryptamine or 5-MeO-α-methyltryptamine, is based on the reduction of 3-(nitrovinyl)indole precursors (**22**) that derive from the condensation of 3-formylindoles **21** with 1-nitroalkanes using weakly basic catalysts (Henry reaction) (Fig. 3A) [13,29,30]. This reduction is generally known to proceed smoothly but the choice of reducing agent can have an influence on the product formation. When working on the synthesis of α-substituted serotonin derivatives, for example, 5-benzyloxy-3-(hydroxymethyl)indole has been isolated after the reduction with LAH, which was confirmed by elemental analysis [30]. It has been suggested [31] that incomplete reduction of 3-formylindoles that have been carried over to the nitrovinyl stage will result in the formation of 3-(hydroxymethyl)indole compounds (**23**). Correspondingly, it has been shown that *N*-1-substituted 3-(hydroxymethyl)indoles are stable to hydrogenolysis with LAH [32]. Whether this carryover of the aldehyde was responsible or not is difficult to decide, since the reduction of 3-formylindole is normally known to yield skatole **24** (3-methylindole) as the major product [33]. Nevertheless, the instability of 3-(hydroxymethyl)indoles in hot alkali, water and acids and its impact on colour-formation is well known [33]. This may have certain implications when acidic workup procedures are involved. For example, 3-(hydroxymethyl)indole (**23**), also known as indole-3-carbinol (I3C), is known to produce a wide range of polymeric products under acidic conditions and three representative examples are shown in Fig. 3A (**25**–**27**) [34]. Some of these polymeric compounds (**23** + **25**) are investigated as potential anti-cancer drugs [35]. A validated reversed-phase HPLC-UV-fluorescence method for the detection of I3C **23** and some of these compounds in mouse plasma has recently been published [36]. A complete reduction of I3C during the LAH reduction would yield skatole **24** [33]. The reduction of the nitrovinylindoles does not always give acceptable yields, since it was found to give significant amounts of skatole derivatives [31]. Skatole itself is considered to be a pneumotoxin that exerts its toxic effects via cytochrome P450-mediated bioactivation, potentially resulting in carcinogenic activity [37].

Interestingly, it has been recently reported that nitrovinylindoles **28** can undergo Michael addition with 3-unsubstituted indoles, indicating sufficient nucleophilicity to yield dimeric 2,2-bis(3′-indolyl)nitroethane **29** [38]. This microwave-assisted reaction was also found to take place solvent free using TLC-grade silica gel, even at room temperature, where the acidic properties of the silica gel were sufficient enough for catalysis (Fig. 3B) [38]. Further investigations resulted in the discovery of unsymmetrical 2′,3′′-derivatives (**30**) and symmetrical 3′,3′′-derivatives (**29** + **31**) when nitrovinylindoles were refluxed with indoles in acetonitrile in the presence of *para*-toluenesulfonic acid (*p*-TsOH) as the catalyst in the Michael addition (Fig. 3C). When

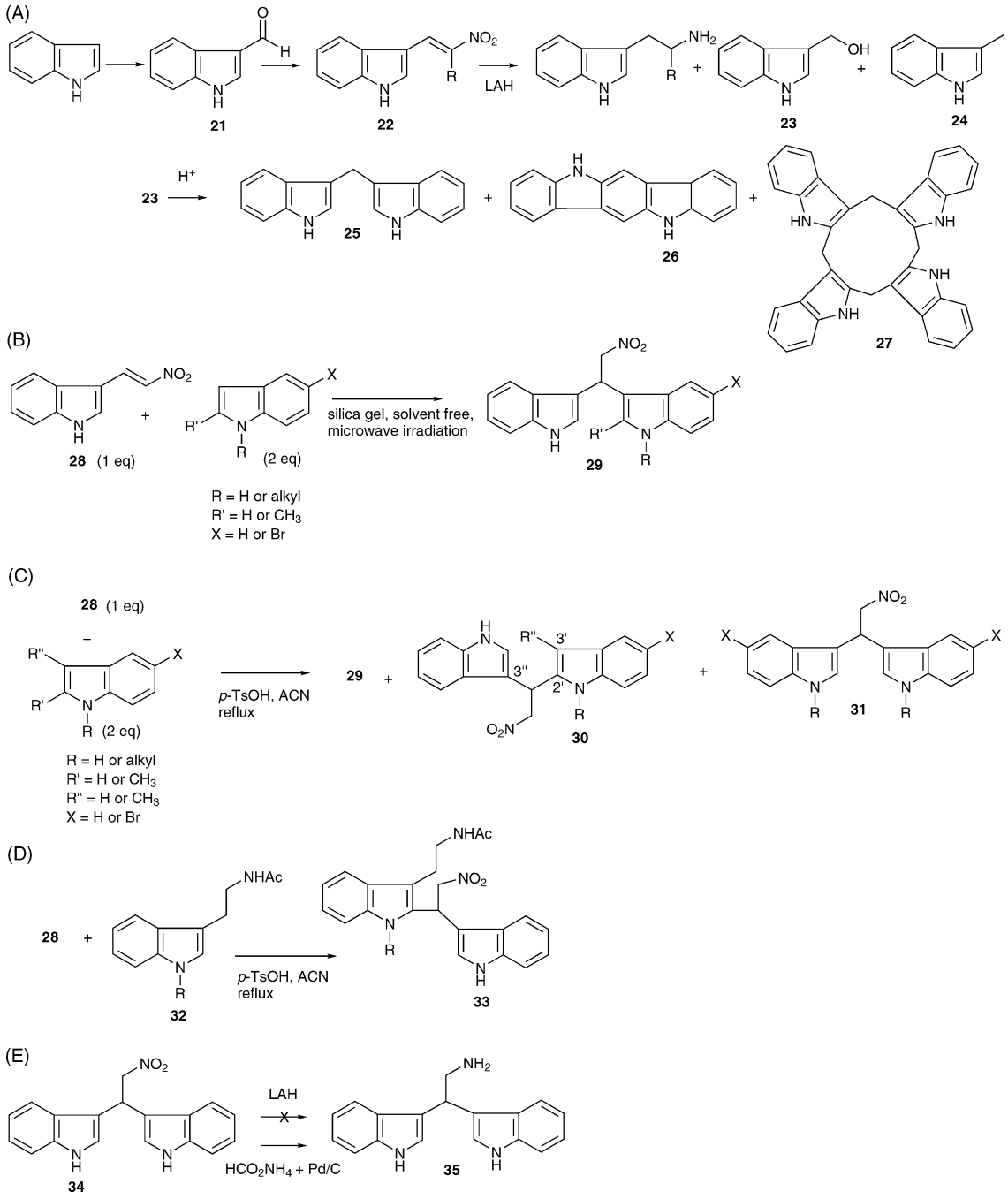

Fig. 3. Some reactions of nitroalkene derivatives. (A) A common route to α-substituted tryptamine derivatives. 3-Formylindole **21** undergoes condensation with 1-nitroalkanes to yield 3-(nitrovinyl)indole **22**. Some examples are known where skatole (3-methylindole) **24** and 3-(hydroxymethyl)indole **23** (I3C) derivatives are formed during the reduction of the nitrovinylindole intermediate with LAH. **23** forms complex polymeric compounds under acidic conditions. Only two dimers (**25**, **26**) and one cyclic tetramer (**27**) are shown [34]. (B) Nitrovinylindoles (**28**) have been shown to undergo Michael addition that yield dimeric 2,2-bis(3′-indolyl)nitroethanes **29** under microwave irradiation [38]. (C) Formation of unsymmetrical (**30**) and symmetrical (**29** + **31**) dimers after refluxing **28** with indoles under acidic conditions with *para*-toluene sulfonic acid (*p*-TsOH). (D) *N*-acetyltryptamine (**32**) was found to dimerise when refluxed with **28** under similar conditions. (E) A reduction of 2,2-bis(3′-indolyl)nitroethane **34** to its ethylamine counterpart **35** [38,39].

nitrovinylindole **28** was refluxed with *N*-acetyltryptamine **32** under similar conditions a dimeric compound **33** was obtained (Fig. 3D) [39]. These findings suggest that formation of similar compounds as by-products cannot be completely excluded under similar reaction conditions. Since nitrovinylindoles are reduced to afford the α-substituted tryptamines, similar reductions of dimeric impurities appear conceivable. The reduction of these dimeric compounds leads to 2,2-bis(indolyl)ethylamines that exhibit potent biological activities [40]. A reduction of 2,2-bis(3′-indolyl)nitroethane **34** to its ethylamine counterpart **35** did not work with LAH but reduction occurred with Pd/C and

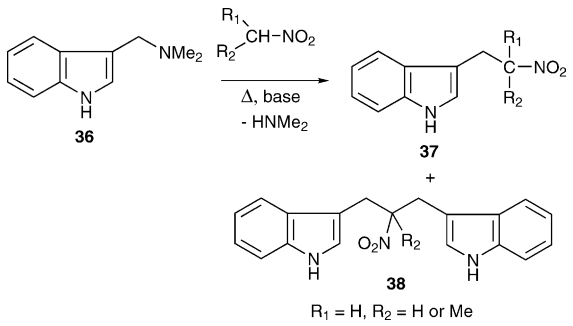

Fig. 4. An example of the gramine-nitroalkane route to α- or β-substituted tryptamines. Gramine derivative **36** is used for the alkylation of a variety of nitroalkanes in order to give the corresponding 3-(2-nitroalkyl)indoles (**37**), which, in turn, can be reduced to the desired tryptamine (not shown). The use of nitromethane or nitroethane leads to dialkylated dimers (**38**) [41].

ammonium formate as the hydrogen transfer agent (Fig. 3E) [39].

5. Routes involving 3-(2-nitroalkyl)indoles

As mentioned above, the aldehyde–nitroalkane route is based on a convenient condensation of an indole-3-aldehyde with a variety of nitroalkanes. Another common approach is based on the alkylation of nitroalkanes with gramine (3-(aminomethyl)indole derivatives **36** to yield 3-nitroethylindoles **37** which, in turn, are reduced [41]. Both the aldehyde–nitroalkane and the gramine–nitroalkane route are useful for the synthesis of α- or β-substituted tryptamines. One major factor that has an influence on the purity of the target molecules is dialkylation to give **38**, mostly when nitromethane or nitroethane are alkylated due to there being two active hydrogens. These form dimeric compounds (**38**) (Fig. 4), which would then be reduced to their dimeric amine counterparts [41]. Several modifications have led to minimising the formation of the dimeric side product, e.g. by the use of sodium instead of NaOH as the proton acceptor and excess of nitroalkane [12] or using gramine-*N*-oxide in the presence of sodium ethoxide [42]. A mild approach is based on the nucleophilic displacement of the quaternised aminofunction of gramine with the anion of nitroethane. A modified procedure, originally used by Heath-Brown and Philpott [43], with sodium methoxide and dimethyl sulfate was used where the amount of the dimeric nitromethane derivative could be reduced to 5% [44]. The dimeric by-product was isolated by column chromatography and characterised by EIMS, elemental analysis and NMR. The use of LAH for the reduction of nitroethylindoles is common although some researchers detected significant amounts of skatole derivatives in this route as well, although, details were not given [31].

6. Chiral tryptamines

One might expect the different chiral isomers of the α-substituted tryptamines to have differing psychotropic effects, particularly, in view of the well-documented behaviour of the chiral isomers of amphetamines. Much less is published for the tryptamines, however, what is known does support that thesis. Shulgin and Shulgin comment that in their experimentation with α-methyltryptamine the *S*-isomer, which is dextro-rotatory, was 3–4 times more potent than the *R*-isomer [3]. This absolute configuration is the same as for the more potent of the isomer pairs of amphetamine, methamphetamine and 3,4-methylenedioxymethamphetamine (*Ecstasy*).

An early report by Nichols pointed out that α-methyltryptamine and its 5-MeO and 4-HO congeners were orally active in humans, and that in general the *S*-(+) enantiomers were more potent than the *R*-(−) enantiomers in both animal and human experiments [45]. In a more recent paper, Hong et al., in a study of animal discriminative stimulus properties of α-ethyltryptamine optical isomers, compared the hallucinogenic- with the stimulant- properties of the isomers. They concluded that the stimulant nature of α-ethyltryptamine resides primarily with its (−)-isomer whereas the hallucinogenic character rests principally with the (+)-enantiomer [46]. It would seem possible therefore that some effort could be made by recreational drug chemists, to synthesise one isomer in preference to another, to achieve the desired effect.

The synthesis of racemic mixtures of chiral tryptamines can be accomplished by many of the methods described in the literature, but the synthesis of single enantiomers is less extensive. One approach is to synthesise the racemic mixture and then separate the chiral isomers, the other is to synthesise directly the specific chiral isomer.

In order to prepare single isomers from racemates, a resolution step is required with some chiral influence that can differentiate and resolve the enantiomers. Crystallisation with a chiral derivatising agent is one such approach on a preparative scale. The resolution of (±)-α-methyltryptamine (AMT) has been accomplished via tartrate salts [47]. It was found that the (−)-AMT enantiomer has the same configuration as that of (−)-amphetamine. The fractional crystallisations of (±)-α-ethyltryptamine (AET) and (±)-7-methyl-AET were achieved with D-camphor-10-sulfonate salts from isopropanol, and dibenzoyl-D-tartrate salts from ethanol, respectively, by Hester et al. [48]. The resolution of the chiral isomers of 7-benzyloxy-AMT provided intermediates for the synthesis of an adrenaline β_3-agonist [49]. Resolution was achieved by fractional crystallisation with *O*,*O*-di-*p*-toluoyl L-(2R,3R)-tartaric acid and enantiomeric purity was determined on a Chiralpak AD chiral HPLC column.

Enzymatic resolution has also been used in the context of AMTs. Kitaguchi et al. found that the enzyme *subtilisin* could be used to resolve the enantiomers of (±)-AMT [50]. Resolution was achieved by amidation of AMT with 2,2,2-trifluoroethyl butyrate. Subtilisin was used as a stereoselective catalyst. The enzyme shows enantioselectivity for the *S*-isomer of AMT and the reaction resulted in the formation of *S*-amides.

Repke reported the first asymmetric synthesis of *S*-(+) and *R*-(−)-AMT from ethyl D- and L-tryptophanate [51]. A five-step synthesis starting from ethyl *S*-tryptophanate gave *R*-AMT. The stereochemistry of the chiral centre remained intact during the sequence but the Cahn, Ingold and Prelog priorities changed. ^{1}H NMR using a europium shift reagent established the enantiomeric purity to be greater than 97%. Analysis was carried out using the indole C-2 proton and optical rotation data were also given. The levo-rotatory isomer was found to have the same configuration as levo-amphetamine (R) [51].

In 1988, Nichols et al. prepared the pure enantiomers of a series of 4-, 5- and 6-hydroxy and alkoxy AMTs via the formation of aryl-2-propanones (Fig. 5) [52,53]. The procedure involved an indole/nitropropane condensation to give **40** followed by formation of the indolyl-2-propanones (**41**) by treatment with sodium methoxide and titanium III chloride in an ammonium acetate buffer. Reductive amination of the intermediate indole-2-propanones with optically pure α-methylbenzylamine (**43A** and **43B**) using sodium cyanoborohydride afforded four diastereoisomers (**45A**–**D**) [52]. Centrifugal chromatography of the diastereoisomers and catalytic debenzylation resulted in the optically pure AMTs (**46** and **47**). The condensation with α-methylbenzylamine proved to be problematic as the main product was a carbazole (**44**), the result of a base-catalysed dimerisation. Melting point, NMR and exact mass were obtained. Another impurity was an oxindole (**42**) and many other impurities generated from this step were not formally identified. For example, *O*-debenzylation of the 4-benzyloxy enantiomers led to an unexpected side reaction: reduction of the benzenoid ring that resulted in the formation of a dihydroindolone.

Optical purity of the AMTs **46** and **47** was established by chiral HPLC-UV (napthoylamide derivatives on a 4.6 mm i.d. Pirkle phase) with 7% ethanol in hexane at 254 nm. No impurity was detected and the optical purity was taken as greater than 98%.

Ezquerra et al. synthesised 5,α,β-trimethyltryptamine stereoselectively resulting in the formation of two of the four possible diastereoisomers [54]. The method involved a nucleophilic aziridine ring-opening reaction of substituted "lower order" indole magnesium bromides with *N*-boc-protected aziridines. The substituted-tryptamine hydrochloride salts resulted after removal of the boc (*t*-butoxycarbonyl) group [54].

(D)-(+)-6-Methoxy-β-methyltryptamine was synthesised in an involved procedure from (D)-(+)-pulegone [55]. Japp–Klingemann coupling of the pulegone with the diazonium salt of *m*-anisidine followed by Fischer cyclisation gave a diester. Hydrolysis, decarboxylation and Curtius degradation yielded the tryptamine derivative.

It is clear that these synthetic routes to optically pure compounds are not trivial, and it seems unlikely that they would be used in clandestine synthesis. If enzymatic processes could be employed, the story might be different. Although none has been reported yet, a recent patent on the two-step enzymatic synthesis of tryptamines from indoles points the way [56].

7. Effect of acids on tryptamines

It is well known that acids have a major influence on indole- and tryptamine-chemistry, for instance, via the acid-catalysed formation of dimers and polymers [57].

Indole-like starting materials or side-products form dimers or trimers (**48**–**50**) during acidic workup procedures depending on the experimental conditions, as reviewed by Smith (Fig. 6A and B) [57]. It was concluded that polymerisation did not appear to go beyond the trimer stage. Any amorphous material produced was attributed to the process of autoxidation [57]. Evidently, the use of acids does not only play its role during workup stages. Berman et al. reported on trifluoroacetic acid-induced dimerisation of indole-3-acetic acid **51** to give **52** (Fig. 6C) [58]. In certain instances, the indole nitrogen can act as a nucleophile. For example, Chen et al., when working on the synthesis of an indole acetic acid derivative **54**, observed the formation of dimeric and trimeric impurities (**55** + **56**, Fig. 6D) [59]. A 3-hydroxy-methylindole derivative **53** was converted to its carboxy counterpart **54** via an indole acetonitrile intermediate. The alcohol was refluxed with a mixture of NaCN, NaOH, EtOH and water, followed by acidification. The separation of the desired product **54** (as sodium salt) from both impurities was achieved with column chromatography on SP-207 resin. The structure of both impurities was elucidated by NMR [59].

Sumatriptan (**57**), a methylsulfonamide derivative of DMT, is a 5-$HT_{1B/1D}$ receptor agonist that is used for the treatment of migraine [20]. Xu et al. investigated the stability of Sumatriptan succinate under the influence of environmental stress conditions, such as oxidation, UV irradiation, heat, acid and base. Degradation product structures were proposed based on collision-induced dissociation studies by LC–ESI–triple quadrupole-MS–MS (Fig. 6E) [60].

Another example is the utilisation of acids during the indolisation step of the Fischer indole synthesis [61], a convenient route that is used for the synthesis of many tryptamine derivatives [5]. The chemistry related to Sumatriptan and its congeners may serve as an example. Phenylhydrazines **58** condense with ketones or aldehydes to yield phenylhydrazone derivatives (**59**) that undergo rearrangements to the appropriate tryptamine derivative (**60**) in the presence of protic or Lewis acids (Fig. 7A) [61]. This one-pot approach, however, has also resulted in acid-induced degradation followed by the formation of di- or multimeric by-products (**61**–**63**) under these conditions (Fig. 7B–D). Structure elucidations of these were performed to a different extent by IR, NMR and exact mass measurements [62–64]. The main 2,5-bisindole Sumatriptan impurity (**64**) that occurs during a typical Fischer indolisation has recently been synthesised. Conditions were utilised that are frequently used during general tryptamine syntheses, such as alkylation in an acidic environment (Fig. 7E) [65]. The use of 3-substituted phenylhydrazones can result in isomeric mixtures of 4- and 6-substituted tryptamines [66].

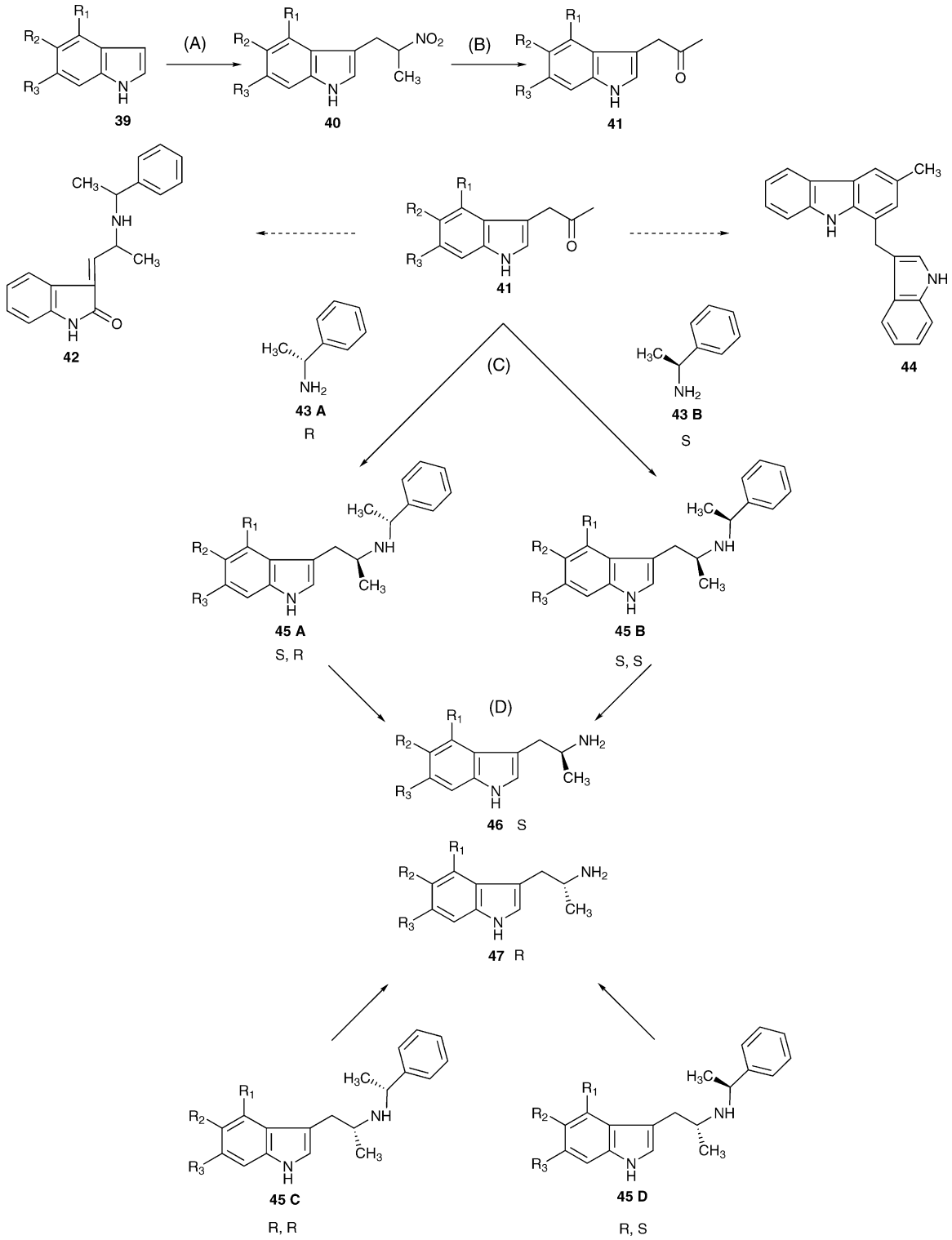

Fig. 5. (A) Condensation of substituted indoles (**39**) with 2-nitropropane. (B) Treatment of resulting indole-3-nitropropanes (**40**) with sodium methoxide in methanol followed by titanium III chloride affords the indole-3-acetones (**41**). (C) Condensation of the indole-3-acetones with *R*- and *S*-α-methylbenzylamine enantiomers (**43A** + **B**) gives the diastereoisomers **45A**–**D**. (D) Catalytic *N*-debenzylation with Pearlman's catalyst at 50 psi of hydrogen results in *S*- and *R*-AMT (**46** + **47**). Oxindoles (**42**) and carbazoles (**44**) were identified as by-products of the condensation of indole-3-acetones with α-methylbenzylamine (**43A** + **B**). These impurities were formed by enamine oxidation and base catalysed dimerisations, respectively.

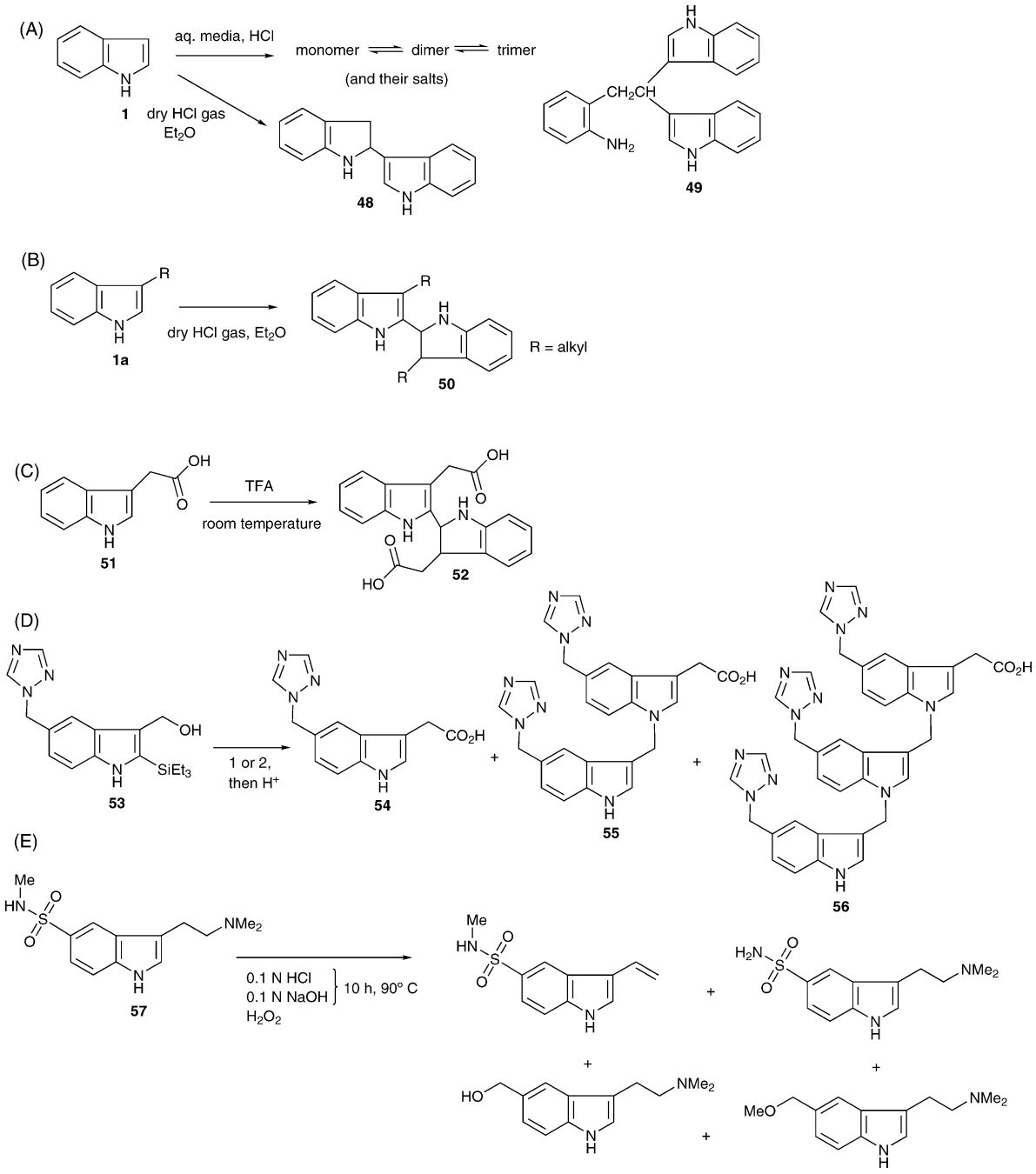

Fig. 6. The behaviour of indoles and tryptamines in the presence of acids. (A) Indole dimerisation in the presence of acids. In a dilute solution, the reaction rate depends on the concentration of acid. Aqueous HCl causes a complex mixture of monomers (**1**), dimers (**48**) and trimers (**49**) forming an equilibrium. Gaseous HCl, with indole **1** in an aprotic solvent, causes the formation of a dimer HCl salt **48** only [57]. (B) 3-Substituted indoles **1a** form 2,2′-dimers **50** in the presence of acids [57]. (C) Indole-3-acetic acid **51** has been shown to dimerise to **52** in the presence of trifluoroacetic acid at room temperature [58]. (D) Indole nitrogen can act as a nucleophile under these conditions. 1. NaCN, EtOH–H_2O or 2. NaCN, NaOH, EtOH–H_2O. Formation of dimer **55** and trimer **56** have been observed [59]. (E) Degradation of Sumatriptan **57** under several stress conditions. In addtion *N*-oxides and several hydroxylation products have been observed [60].

A common approach, as exemplified in the synthesis of several triptan-type tryptamines from hydrazine **65**, is based on the reductive alkylation of the non-alkylated ethylamine side-chain with $CH_2O/NaBH_4$ [67] or $CH_2O/NaCNBH_3$ [68] in acidic media to afford the dimethylated derivative (**66**). These reducing agents enabled the reduction of an intermediate imine or iminium salt that was formed via acidification and elimination of water. The presence of acid and aldehydes (formed from the acetal), depending on the conditions, can give rise to unwanted Pictet–Spengler cyclisations that yield tetrahydro-β-carboline derivatives (**67**) (Fig. 8A) [69], but no analytical data on **67** were given. Pictet–Spengler reactions,

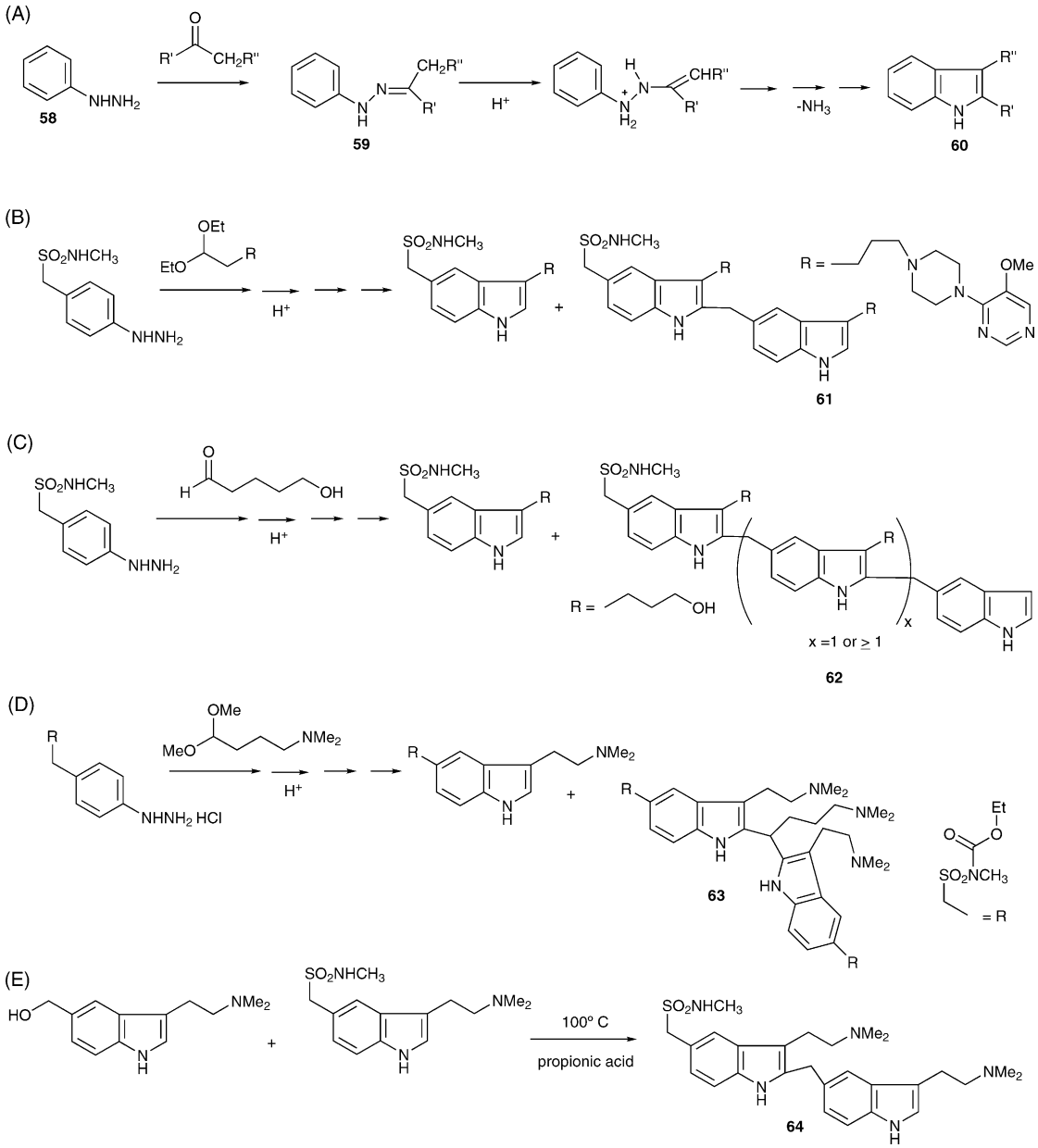

Fig. 7. Formation of several dimeric side products during the Fischer indole synthesis. (A) Simplified Fischer indole synthesis that is commonly used for the production of tryptamine derivatives. Phenylhydrazines (**58**) condense with ketones or aldehydes to yield phenylhydrazone derivatives (**59**) that undergo rearrangement to the appropriate tryptamine (**60**) in the presence of protic or Lewis acids [61]. (B) The use of a hydrazine and an acetal for indolisation. A dimeric side product (**61**) was observed [62] (C) Formation of a multimeric side product (**62**) using a two-phase indolisation procedure [63]. (D) 1,1-bis(indol-2-yl)-4-dimethylaminobutane by-product (**63**) [64]. (E) Synthesis of a main Sumaptriptan impurity (**64**) that is typically found during the conditions of the Fischer indole synthesis [65].

however, are also known to occur in non-acidic, aprotic media [70,71]. The presence of electron-donating substitutents on the benzene ring can have an effect on the nucleophilicity of indole C-2. For example, 6-OH- and 6-MeO-tryptamine hydrochlorides, but not their 5-substituted counterparts, were converted into their corresponding tetrahydro-β-carbolines when left in aqueous acetone at room temperature at pH 4.7 [13]. A similar phenomenon was briefly mentioned during the conversion of 6-HO-α-ethyltryptamine free base (**68**) into its creatinine sulfate salt (**69**) in aqueous acetone (**69**) (Fig. 8B) but no details were reported. A similar pathway was also expected for 4-substituted derivatives [13].

It has been shown that the complex formation of certain carbonyl condensation products can lead to dramatic consequences. The dimeric 2-(3-indolylmethyl)-L-tryptophan **71** (IMT, Fig. 9A) has been identified as one major case-associated contaminant in biotechnically manufactured L-tryptophan **70** (L-Trp) that was involved in the outbreak of the eosinophilia-myalgia syndrome (EMS) in 1989 [72]. IMT was found to be formed under acidic and alkaline

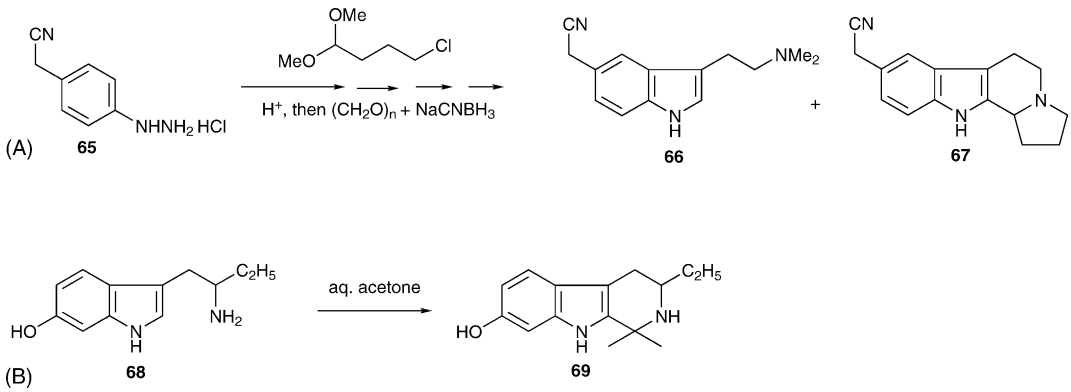

Fig. 8. (A) Pictet–Spengler side reaction during a Fischer indole synthesis between tryptamine derivative (**66**) and unreacted acetal that leads to the formation of tetrahydro-β-carboline **67** [69]. (B) The electron-donating properties of the 6-HO-group in **68** increases nucleophilicity at position C-2. The use of aqueous acetone during workup procedures facilitated a Pictet–Spengler reaction that yielded tetrahydro-β-carboline **69** [13].

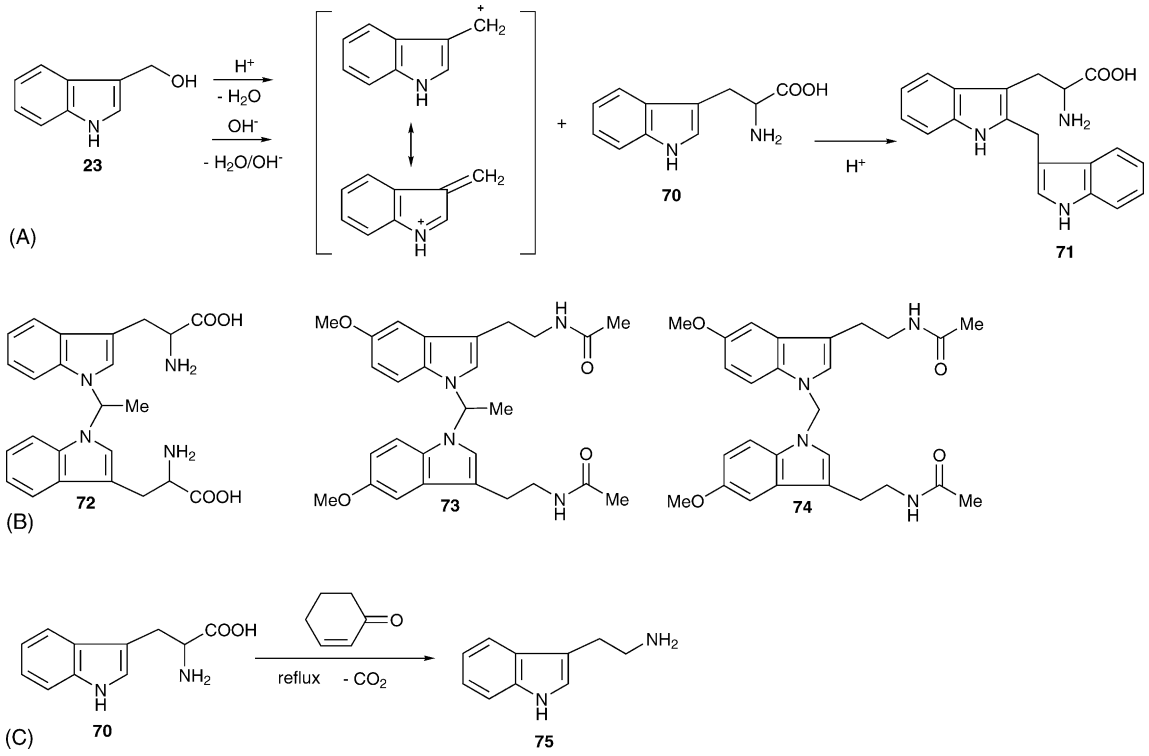

Fig. 9. (A) Dimeric 2-(3-indolylmethyl)-L-tryptophan (IMT) (**71**) formed under acidic and alkaline conditions during manufacture [73]. Suggested formation of this dimeric product via reaction with indole-3-carbinol (I3C) (**23**) [73]. (B) Depending on pH conditions another dimeric side product was identified as 1,1′-ethylidenebis-(L-Trp) (**72**), representing a 1-substituted dimer [75–77]. Melatonin-formaldehyde condensation products (**73** + **74**) [78]. (C) Decarboxylation of tryptophan (**70**) in cyclohexanol using 2-cyclohexen-1-one as the catalyst yielding tryptamine **75** [79].

conditions utilising the reaction between **23** and **70**, that are typical for down stream processes, such as ion exchange purification (Fig. 9A) [73] and additional characterisation using LC–MS–MS was performed [74]. Depending on pH conditions another dimeric case-associated contaminant was found identified as 1,1′-ethylidenebis-(L-Trp) 72, representing a 1-substituted dimer (Fig. 9B) [75–77]. Interestingly, Williamson et al., applying LC–MS–MS, characterised equivalent versions of dimeric formaldehyde condensation products (**73** + **74**) in commercial preparations of melatonin (Fig. 9B) [78].

8. Decarboxylation of tryptophans

Tryptophan and 5-methoxytryptophan are favoured starting materials for the synthesis of tryptamines **75**, being readily available as dietary supplements. Decarboxylation of

tryptophan is by far the simplest and probably the cheapest way to the synthesis of tryptamine. Hashimoto et al. [79] completed the decarboxylation of tryptophan **70** to give tryptamine **75** in a 92% yield. The reaction mixture was gently refluxed in a stream of nitrogen for a few hours until there was no more evolution of carbon dioxide (Fig. 9C). They used cyclohexanol as the solvent but the reaction was accelerated and a higher yield of amine produced with the addition of 2-cyclohexenone, which was in fact present as an impurity; they applied this method also to various other α-amino acids including tryptophan **70** that gave 92% yield of tryptamine **75**.

Kametani et al. [80] used diphenylmethane as the high boiling solvent instead of cyclohexanol. Other methods include an inexpensive two-step catalytic decarboxylation with metal ions by reacting tryptophan with copper acetate or zinc acetate to form metal chelate compounds that then decarboxylate to produce tryptamine hydrochloride [81]. Takano et al. [82] heated L- or DL-tryptophan at reflux in a small amount of tetralin with a catalytic amount of carbonyl compound such as pentan-3-one for 8–10 h with vigorous stirring until no more carbon dioxide was evolved. Good yields of tryptamine 60–95% were obtained in all of these methods. A different approach to decarboxylation of tryptophan has also been reported and discussed on the Rhodium website [83] using the natural abundance of carvone (2-methyl-4-(2-propene)-cyclohex-2-enone) in spearmint oil as the ketone catalyst and either xylene or white spirit as the refluxing solvent achieving 65% yield of tryptamine. It is suggested that dill, caraway (contains carvone) or penny royal (contains pulegone, 5-methyl-2-isopropylidene cyclohexanone) natural oils could also be employed as the catalyst.

The point to note in all these methods described is the range of metals or organic side products that may be present as trace constituents in the final product, although the extent of their carry over has not yet been determined in our laboratory.

9. Oxidation of tryptamines and indoles

Indoles and tryptamine derivatives are sensitive to light and air and consequently, they are susceptible to autoxidation which can result in the formation of indigo (**77**) and trimers (**78**) and complex coloured, polymeric tarry mixtures (Fig. 10A) [84]. Tryptophan **70**, for example, after photooxygenation was transformed into a 3-hydroperoxide-indoline derivative (**79**) (Fig. 10B) [85].

The oxidative behaviour of these compounds depends on the choice of the oxidising chemical or catalyst and the conditions.

When the DMT derivative Sumatriptan **57** was exposed to oxidative stress induced by H_2O_2, six degradation products were found using LC-MS and LC-MS-MS [60] One peak in the total ion chromatogram was attributed to a side chain *N*-oxide whereas the remaining ones were found to represent single or multiple oxidation sites on the ring system

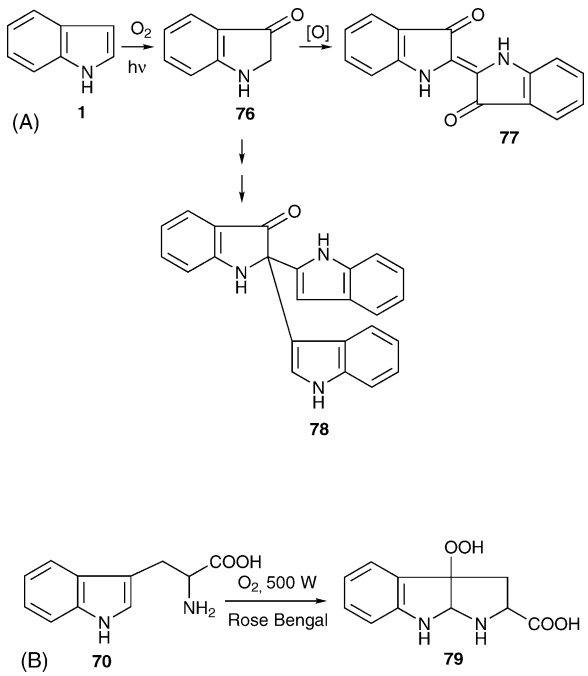

Fig. 10. (A) Autoxidation of indole (**1**) results in the formation of compounds such as indigo (**77**) or trimer **78** probably via Indoxyl (**76**) [84]. (B) Rose Bengal sensitised photooxygenation of L-tryptophan (**70**) in the presence of acetic acid and subsequent formation of a hydroperoxide derivative (**79**) [85].

[60]. Multiple hydroxylations have been shown to result in even more complex autoxidations, depending on experimental conditions, such as pH and polarity of solvents [86–88]. Hydroxylated tryptamine derivatives, such as serotonin (5-hydroxytryptamine) or 5-hydroxytryptophol were found to produce dimeric compounds upon photolysis as studied with LC-APCI-MS [89]. Commercial preparations of the dietary supplement 5-HO-Trp have been analysed using LC-MS-MS. An oxidised product has been identified as Trp-4,5-dione, which was found to be associated with the occurrence of an EMS-like case [90].

An interesting insight into the reactivity and complexity of tryptamine chemistry was given by Somei when reviewing the complex and sensitive chemical behaviour of 1-hydroxytryptamines and derivatives including the occurrence of nucleophilic substitution, oxidation, rearrangement, dimerisation and other types of reactions [91].

10. Future trends

Several well-known synthetic routes to (psychoactive) tryptamines and their implications on possible side-product formation have been reviewed. A systematic analytical study of the synthetic routes is missing, since most of the published tryptamine-related chemistry is focused on preparative aspects. Standard techniques such as UV, IR, MS, NMR and elemental- or melting point analysis dominated the early work several decades ago. These indispensible approaches

have nowadays been enriched, for example, by high field- and 2D NMR, GC–(EI/CI)–MS^n or LC–API–MS^n which can simplify and shorten these analytical procedures.

Some recent findings of the authors' current research programme into the analytical chemistry of synthetic routes to the psychoactive tryptamines for forensic work may serve to exemplify some possible future directions. The psychoactive 5-methoxy-*N*,*N*-diisopropyltryptamine has been recently synthesised in this laboratory by the method of Speeter and Anthony. Four major impurities that occurred during the last synthetic step have been identified and characterised by NMR, ESI–time of flight–MS and ESI–triple quadrupole tandem mass spectrometry [19]. One application of tandem mass spectral information is the determination of compound-specific ion transitions that may be used for screening purposes. Fig. 11 shows an example of a possible scenario where these ion transitions are used for a multiple reaction monitoring approach. 5-MeO-DIPT-free base (10 ng) was subjected

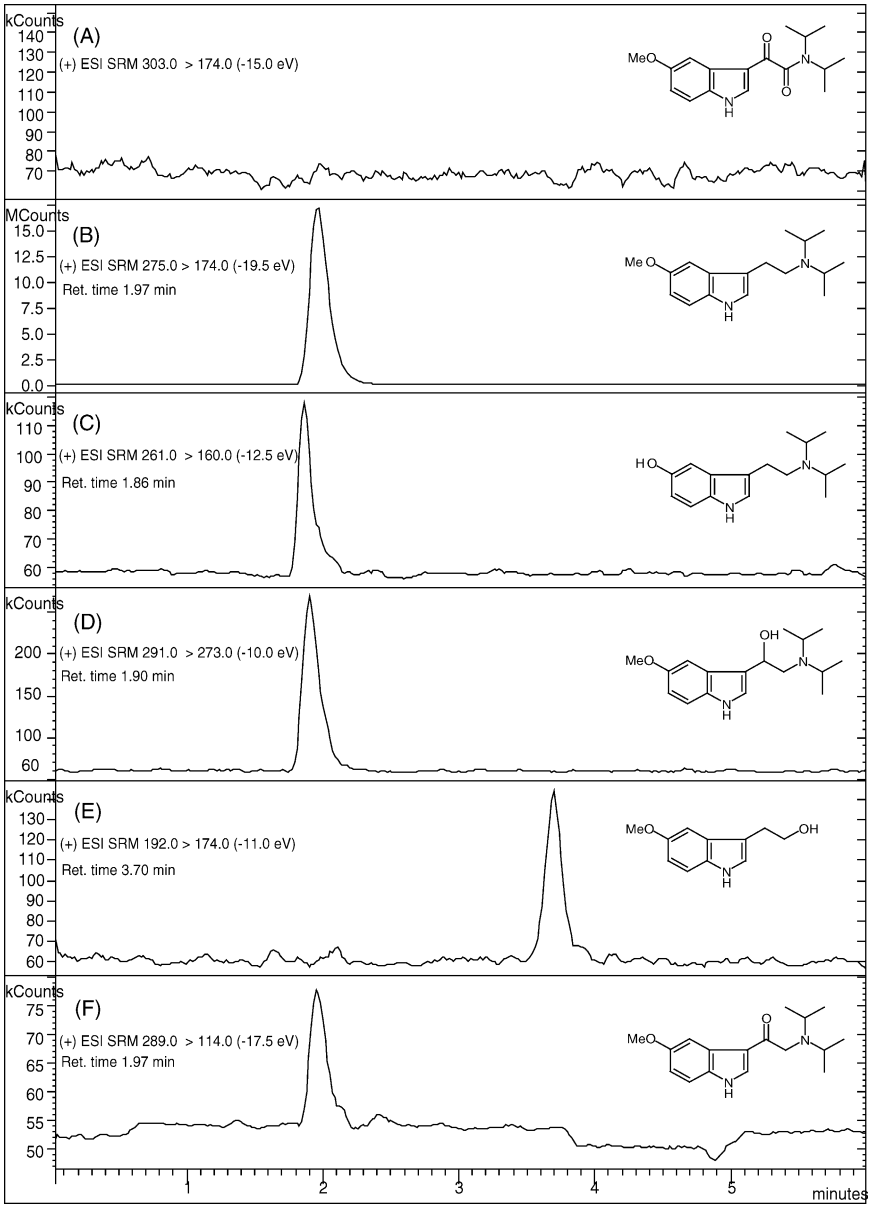

Fig. 11. Screening of synthesised 5-methoxy-*N*,*N*-diisopropyltryptamine (5-MeO-DIPT) free base for the presence of impurities by multiple reaction monitoring using LC-ESI-TQ-MS-MS (preliminary results). (A) Starting material of the last synthetic step during the route of Speeter and Anthony, 5-MeO-indole-3-yl-*N*,*N*-diisopropylglyoxalylamide. (B) Product after reduction with $LiAlH_4$, 5-MeO-DIPT. (C) Impurity, 5-HO-DIPT. (D) Impurity, 5-MeO-β-HO-DIPT. (E) Impurity, 5-MeO-tryptophol. (F) Impurity, 5-MeO-β-keto-DIPT. 10 ng of free base B was injected on a 150 × 2.0 mm, 3 μ *Polaris* C_8 column (mobile phase: 50:50 ACN:H_2O, 0.1% formic acid (v/v), isocratically). Retention times, selected ion transitions and collision energies (rf-only quadrupole collision cell) are depicted in the figure. No complete chromatographic resolution but fast analysis time was intended in this MRM approach. Preliminary estimations indicate a range of 0.5–1.5% per impurity, possibly, slightly higher for (D). Further work will provide detailed quantitative estimates of the abundance of the above impurities in the free base product.

to LC–ESI–MS–MS. In a typical $Q_1q_2Q_3$ study, a triple quadrupole MS (TQ–MS–MS) is used to scan for a number of selected fragmentations. Q_1 is set to select the protonated molecular ion $[M + H]^+$ of interest which is subsequently dissociated in the second (RF-only) quadrupole (q_2). Q_3 then scans for the characteristic product ion, which is represented by the corresponding peak in the chromatogram. This highly selective approach not only allows the exact quantification of impurities, but the combination of several different ion transitions also makes possible unambiguous identification of a synthetic route. As shown in Fig. 11, the presence of two incompletely reduced impurities **11D** and **11F** in the free base product has been confirmed during the screening procedure, which may be characteristic for this reduction step. For example, the dialkylation of 5-MeO-tryptamine with 2-iodopropane, which could also lead to 5-MeO-DIPT, do not show these two corresponding ion transitions. A selected ion transition that monitors the dissociation of a monoalkylated protonated molecular ion into its product ion may then be used as an indicator.

An incomplete reduction of a glyoxalylamide does not necessarily result in the formation of the same

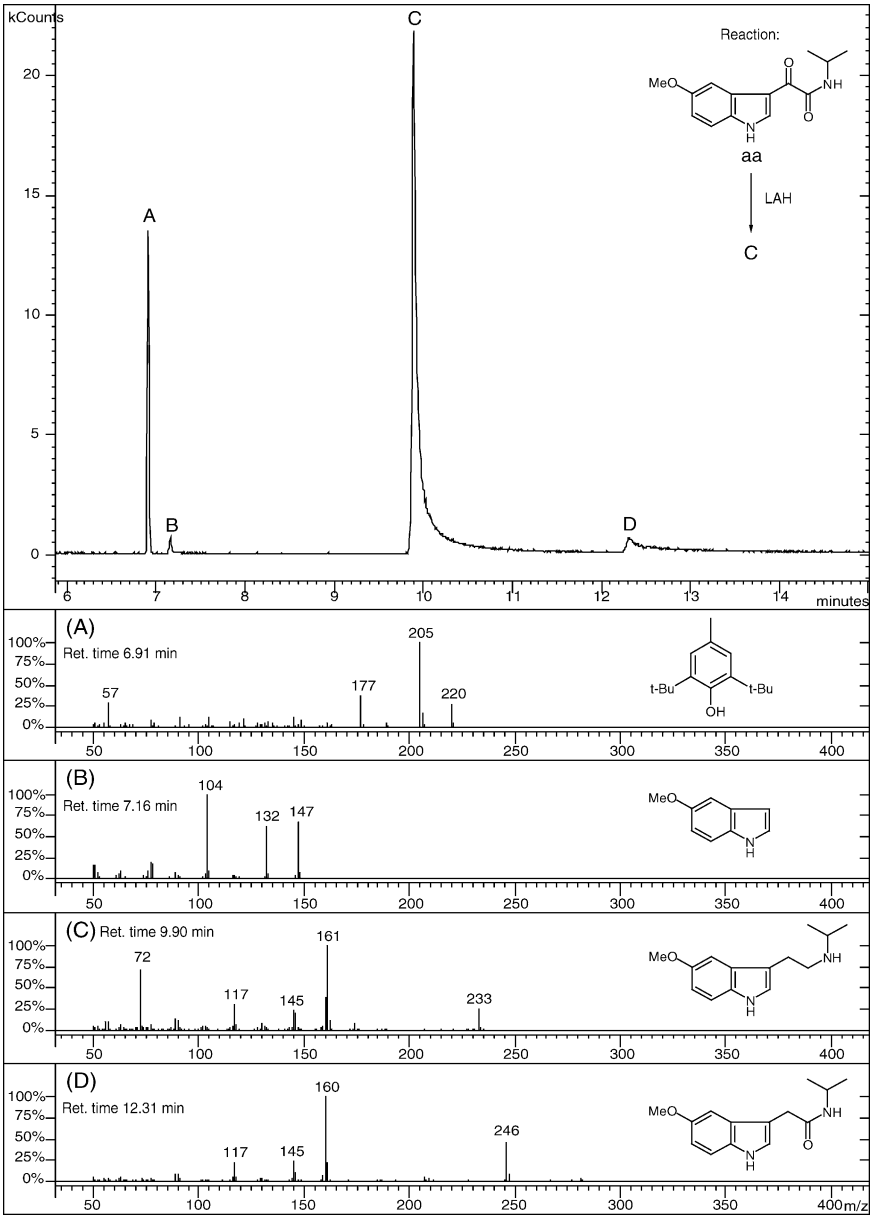

Fig. 12. GC-ion trap-EIMS chromatogram of 5-methoxy-*N*-isopropyltryptamine (5-MeO-NIPT) free base and some key impurities in the full scan mode. (A) Impurity, 2,6-bis(*tert*-butyl)-4-methylphenol (BHT), a commonly added antioxidant to solvents, such as THF, by the manufacturer. (B) Impurity, 5-Methoxyindole. (C) Product 5-MeO-NIPT after reduction of aa with LAH ($LiAlH_4$) during the route of Speeter and Anthony. (D) Impurity, *N*-Isopropyl-2-(5-methoxy-1H-indol-3-yl)-acetamide. 10 ng of free base C was injected in a 50:1 split mode. Capillary column: 5% phenyl, 30 m × 0.25 mm *CP-Sil 8 CB Low Bleed/MS* (film thickness: 0.25 μm). Temperature profile: 1 min at 40 °C, then heat at 50 °C min^{-1} to 260 °C and hold for 14.6 min. Total run time: 20 min.

impurities. Fig. 12 shows a GC-ion trap-EIMS chromatogram (GC-ITMS) after the injection of 10 ng 5-methoxy-*N*-isopropyltryptamine (5-MeO-NIPT) (**12C**) free base. The monoalkylated glyoxalylamide **aa** (Fig. 12) has also been reduced with $LiAlH_4$, which led to the detection of impurity **12D**. This previously uncharacterised compound appears to be *N*-isopropyl-2-(5-methoxy-1H-indol-3-yl)-acetamide. The presence of the α-keto group has been confirmed by tandem mass spectrometry and 2-dimensional NMR: HMQC and HMBC (not shown). The reduction of the diisopropyl-precursor (**11A**), in contrast, has led to the detection of **11F** (2-diisopropylamino-1-(5-methoxy-1H-indol-3-yl)-ethanone) where the carbonyl group is located on the β-position (Fig. 11). Further work that will provide detailed quantitative estimates is in progress in this laboratory.

Twelve symmetrically and 13 asymmetrically *N*,*N*-disubstituted tryptamines and glyoxalylamides have recently been synthesised and characterised by GC–ITMS, ESI–TQ–MS–MS and NMR, most of them for the first time (submitted for publication in the Analyst). For example, the combination of in-source fragmentation with tandem mass spectrometry increased the mass spectral information content, which enabled the differentiation of isomeric tryptamines without the necessity for a previous separation, by the dissociation of tryptamine-derived immonium ions ($CH_2N^+R^1R^2$).

11. Summary

This review is intended to demonstrate the exciting analytical challenge represented by the synthetic routes to the psychoactive tryptamines. The literature on indole and tryptamine chemistry is vast and the pharmacological and neurochemical implications of these materials are profound. It is of importance that the producers of these materials and the users of the products are aware of the potential side-products and impurities in the drugs. By knowing what is present and how the impurities are formed, production of pure compounds with well-defined properties will become more practicable. Those materials can then be used to provide unequivocal answers regarding their pharmacological and psychoactive properties. Chemical profiling of the final products from these syntheses will permit relationships to be understood between the profile and the synthetic route that produced it, permitting a start point for possible criminal investigations where the drugs have been employed illegally. This information will also be of value to the clinical community where there is an increased interest in the use of certain psychoactive tryptamines as neurobiological tools.

Acknowledgements

SDB and PMcG are supported by DIAS bursaries and NAH by the Government of Brunei. Much of the GC, LC & MS equipment in the authors' laboratory was purchased under the Scientific Research Infrastructure Fund Initiative; synthesis of the controlled substances referred to was carried out under a Home Office licence.

References

[1] J.R. Cooper, F.E. Bloom, R.H. Roth (Eds.), The Biochemical Basis of Neuropharmacology, seventh ed., Oxford University Press, Oxford, UK, 1996.
[2] D.E. Nichols, Pharmacol. Ther. 101 (2004) 131–181.
[3] A.T. Shulgin, A. Shulgin, TIHKAL: The Continuation, Transform Press, Berkeley, 1997.
[4] R.R. Laing (Ed.), Hallucinogens: A Forensic Drug Handbook, Elsevier Science Ltd., London, UK, 2003.
[5] S. Freeman, J.F. Alder, Eur. J. Med. Chem. 37 (2002) 527–539.
[6] J. Ott, J. Psychoactive Drugs 31 (1999) 171–177.
[7] J. Ott, J. Psychoactive Drugs 33 (2001) 273–281.
[8] J. Ott, J. Psychoactive Drugs 33 (2001) 403–407.
[9] R.J. Sundberg, Indoles, Best Synthetic Methods Series, Academic Press Ltd, London, UK, 1996.
[10] M.E. Speeter, W.C. Anthony, J. Am. Chem. Soc. 76 (1954) 6208–6210.
[11] H.C. Brown, S. Krishnamurthy, Tetrahedron 35 (1979) 567–607.
[12] F. Troxler, F. Seemann, A. Hofmann, Helv. Chim. Acta 42 (1959) 2073–2103.
[13] R.V. Heinzelman, J. Szmuszkovicz, Prog. Drug Res. 6 (1963) 75–150.
[14] R.G. Taborsky, P. Delvigs, D. Palaic, F.M. Bumpus, J. Med. Chem. 10 (1967) 403–407.
[15] V. Plavsic, S. Iskric, S. Kveder, M. Tucan-Foretic, D. Wolf, V. Gjuris, Z. Supek, J. Pharm. Pharmacol. 28 (1976) 424–428.
[16] D.L. Crookes, K.P. Parry, G.F. Smith, Pol. J. Chem. 53 (1979) 73–78.
[17] W. Gielsdorf, K. Schubert, K. Allin, Arch. Kriminol. 166 (1980) 21–32.
[18] J.S. Cowie, A.L. Holtham, L.V. Jones, J. Forensic Sci. 27 (1982) 527–540.
[19] S.D. Brandt, S. Freeman, I.A. Fleet, P. McGagh, J.F. Alder, Analyst (2004), accepted DOI:10.1039/B407239C.
[20] I. Egle, N. MacLean, L. Demchyshyn, L. Edwards, A. Slassi, A. Tehim, Bioorg. Med. Chem. Lett. 14 (2004) 727–729.
[21] V.M. Micovic, M.L. Mihailovic, J. Org. Chem. 18 (1953) 1190–1200.
[22] F.V. Brutcher Jr., W.D. Vanderwerff, J. Org. Chem. 23 (1958) 146–147.
[23] R. Littell, G.R. Allen Jr., J. Org. Chem. 38 (1973) 1504–1510.
[24] A. Alemany, E.F. Alvarez, O.N. Lopez, M.E.R. Herraez, Bull. Soc. Chim. France 12 (1974) 2883–2888.
[25] W.B. Wright Jr., J. Org. Chem. 25 (1960) 1033–1036.
[26] W.B. Wright Jr., J. Org. Chem. 27 (1962) 1042–1045.
[27] J.M. Khanna, V.M. Dixit, N. Anand, Synthesis 9 (1975) 607–608.
[28] R. Stauffer, Helv. Chim. Acta 49 (1966) 1199–1203.
[29] E.H.P. Young, J. Chem. Soc. 3 (1958) 3493–3496.
[30] A.S.F. Ash, W.R. Wragg, J. Chem. Soc. (1958) 2892–3887.
[31] F. Troxler, in: W.J. Houlihan (Ed.), Indoles, vol. 25 (2), Wiley-Interscience, Chichester, UK, 1972, pp. 179–537.
[32] E. Leete, J. Am. Chem. Soc. 81 (1959) 6023–6026.
[33] E. Leete, L. Marion, Can. J. Chem. 31 (1953) 775–784.
[34] K.R. Grose, L.F. Bjeldanes, Chem. Res. Toxicol. 5 (1992) 188–193.
[35] H.L. Bradlow, D.W. Sepkovic, N.T. Telang, M.P. Osborne, Ann. N. Y. Acad. Sci. 889 (1999) 204–213.
[36] M.J. Anderton, R. Jukes, J.H. Lamb, M.M. Manson, A. Gescher, W.P. Steward, M.L. Williams, J. Chromatogr. B 787 (2003) 281–291.
[37] K.A. Regal, G.M. Laws, C. Yuan, G.S. Yost, G.L. Skiles, Chem. Res. Toxicol. 14 (2001) 1014–1024.

[38] M. Chakrabarty, R. Basak, N. Ghosh, Tetrahedron Lett. 42 (2001) 3913–3915.
[39] M. Chakrabarty, R. Basak, N. Ghosh, Y. Harigaya, Tetrahedron 60 (2004) 1941–1949.
[40] M. Chakrabarty, R. Basak, Y. Harigaya, Heterocycles 55 (2001) 2431–2447.
[41] H.R. Snyder, L. Katz, J. Am. Chem. Soc. 69 (1947) 3140–3142.
[42] D.W. Henry, E. Leete, J. Am. Chem. Soc. 79 (1957) 5254–5256.
[43] B. Heath-Brown, P.G. Philpott, J. Chem. Soc. (1965) 7165–7178.
[44] G.D.H. Dijkstra, M.T.M. Tulp, P.H.H. Hermkens, J.H. Vanmaarseveen, H.W. Scheeren, C.G. Kruse, Recl. Trav. Chim. Pays-Bas 112 (1993) 131–136.
[45] D.E. Nichols, R. Oberlender, D.J. McKenna, Biochem. Physiol. Subst. Abuse 3 (1991) 1–39.
[46] S.S. Hong, R. Young, R.A. Glennon, Pharmacol. Biochem. Behav. 70 (2001) 311–316.
[47] V.M. Potapov, A.P. Terent'ev, M.N. Preobrazhenskaya, N.N. Suvorov, Zh. Obshch. Khim. 33 (1963) 2702–2705.
[48] J.B. Hester, M.E. Greig, W.C. Anthony, R.V. Heinzelman, J. Szmuszkovicz, J. Med. Chem. 7 (1964) 274–279.
[49] A. Fujii, Y. Fujima, H. Harada, M. Ikunaka, T. Inoue, S. Kato, K. Matsuyama, Tetrahedron-Asymmetry 12 (2001) 3235–3240.
[50] H. Kitaguchi, P.A. Fitzpatrick, J.E. Huber, A.M. Klibanov, J. Am. Chem. Soc. 111 (1989) 3094–3095.
[51] D.B. Repke, W.J. Ferguson, J. Heterocyclic. Chem. 13 (1976) 775–778.
[52] D.E. Nichols, D.H. Lloyd, M.P. Johnson, A.J. Hoffman, J. Med. Chem. 31 (1988) 1406–1412.
[53] D.E. Nichols, C.F. Barfknecht, D.B. Rusterholz, F. Benington, R.D. Morin, J. Med. Chem. 16 (1973) 480–483.
[54] J. Ezquerra, C. Pedregal, C. Lamas, A. Pastor, P. Alvarez, J.J. Vaquero, Tetrahedron 53 (1997) 8237–8248.
[55] A. Frey, H. Ott, H. Bruderer, P. Stadler, Chimia 14 (1960) 423.
[56] US Patent Application, 20030096379 (2003).
[57] G.F. Smith, Adv. Heterocyclic Chem. 2 (1963) 287–309.
[58] J. Bergman, E. Koch, B. Pelcman, Tetrahedron Lett. 36 (1995) 3945–3948.
[59] C.Y. Chen, D.R. Lieberman, L.J. Street, A.R. Guiblin, R.D. Larsen, T.R. Verhoeven, Synth. Commun. 26 (1996) 1977–1984.
[60] X.H. Xu, M.G. Bartlett, J.T. Stewart, J. Pharm. Biomed. Anal. 26 (2001) 367–377.
[61] B. Robinson, The Fischer Indole Synthesis, John Wiley & Sons Ltd, Chichester, 1982.
[62] P. Remuzon, C. Dussy, J.P. Jacquet, M. Soumeillant, D. Bouzard, Tetrahedron Lett. 36 (1995) 6227–6230.
[63] P.R. Brodfuehrer, B.C. Chen, T.R. Sattelberg, P.R. Smith, J.P. Reddy, D.R. Stark, S.L. Quinlan, J.G. Reid, J. Org. Chem. 62 (1997) 9192–9202.
[64] B. Pete, I. Bitter, C. Szantay, I. Schon, L. Toke, Heterocycles 48 (1998) 1139–1149.
[65] A. Skwierawska, E. Paluszkiewicz, Pol. J. Chem. 77 (2003) 329–332.
[66] E. Späth, E. Lederer, Ber. 63B (1930) 120–125.
[67] J. Bosch, T. Roca, M. Armengol, D. Fernandez-Forner, Tetrahedron 57 (2001) 1041–1048.
[68] J.L. Castro, R. Baker, A.R. Guiblin, S.C. Hobbs, M.R. Jenkins, M.G. Russell, M.S. Beer, J.A. Stanton, K. Scholey, R.J. Hargreaves, M.I. Graham, V.G. Matassa, J. Med. Chem. 37 (1994) 3023–3032.
[69] C.Y. Chen, C.H. Senanayake, T.J. Bill, R.D. Larsen, T.R. Verhoeven, P.J. Reider, J. Org. Chem. 59 (1994) 3738–3741.
[70] E.D. Cox, J.M. Cook, Chem. Rev. 95 (1995) 1797–1842.
[71] B.E. Love, Org. Prep. Proced. Int. 28 (1996) 1–64.
[72] R.H. Hill Jr., S.P. Caudill, R.M. Philen, S.L. Bailey, W.D. Flanders, W.J. Driskell, M.L. Kamb, L.L. Needham, E.J. Sampson, Arch. Environ. Contam. Toxicol. 25 (1993) 134–142.
[73] B. Müller, C. Pacholski, T. Simat, H. Steinhart, Adv. Exp. Med. Biol. 467 (1999) 481–486.
[74] B.L. Williamson, L.M. Benson, A.J. Tomlinson, A.N. Mayeno, G.J. Gleich, S. Naylor, Toxicol. Lett. 92 (1997) 139–148.
[75] A.N. Mayeno, F. Lin, C.S. Foote, D.A. Loegering, M.M. Ames, C.W. Hedberg, G.J. Gleich, Science 250 (1990) 1707–1708.
[76] M.J. Smith, E.P. Mazzola, T.J. Farrell, J.A. Sphon, S.W. Page, D. Ashley, S.R. Sirimanne, R.H. Hill Jr., L.L. Needham, Tetrahedron Lett. 32 (1991) 991–994.
[77] T.J. Simat, K.K. Kleeberg, B. Muller, A. Sierts, Adv. Exp. Med. Biol. 467 (1999) 469–480.
[78] B.L. Williamson, A.J. Tomlinson, P.K. Mishra, G.J. Gleich, S. Naylor, Chem. Res. Toxicol. 11 (1998) 234–240.
[79] M. Hashimoto, Y. Eda, Y. Osanai, T. Iwai, S. Aoki, Chem. Lett. (1986) 893–896.
[80] T. Kametani, S. Takano, S. Hibino, M. Takeshita, Synthesis 9 (1972) 475.
[81] T. Kametani, T. Suzuki, K. Takahashi, K. Fukumoto, Synthesis 2 (1974) 131–132.
[82] S. Takano, T. Nishimura, K. Ogasawara, Heterocycles 6 (1977) 1167–1171.
[83] http://www.rhodium.ws/chemistry/tryptophan.html.
[84] W.A. Remers, in: W.J. Houlihan (Ed.), Indoles, vol. 25 (1), Wiley-Interscience, Chichester, UK, 1972, pp. 1–226.
[85] M. Nakagawa, S. Kato, S. Kataoka, T. Hino, J. Am. Chem. Soc. 101 (1979) 3136–3137.
[86] T. Tabatabaie, G. Dryhurst, J. Med. Chem. 35 (1992) 2261–2274.
[87] M.Z. Wrona, Z.L. Yang, M. McAdams, S. O'Connor-Coates, G. Dryhurst, J. Neurochem. 64 (1995) 1390–1400.
[88] M.Z. Wrona, X.R. Jiang, Y. Kotake, G. Dryhurst, Chem. Res. Toxicol. 16 (2003) 493–501.
[89] A. Numan, N.D. Danielson, Anal. Chim. Acta 460 (2002) 49–60.
[90] K. Klarskov, K.L. Johnson, L.M. Benson, J.D. Cragun, G.J. Gleich, M. Wrona, X.R. Jiang, G. Dryhurst, S. Naylor, J. Rheumatol. 30 (2003) 89–95.
[91] M. Somei, Adv. Heterocyclic Chem. 82 (2002) 101–155.