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SYNTHESIS OF TETRAHYDRO- β - CARBOLINES VIA THE PICTET-SPENGLER METHOD

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Abstract: In this work, we report the optimization of the Pictet–Spengler reaction using tryptamine and tryptophan with substituted benzaldehydes to obtain the corresponding tetrahydro- β -carbolines. Furthermore, this study aims to illustrate the Maillard reaction to undergraduate Food Chemistry students in the Organic Chemistry III course (code 1628) offered by this Faculty, in order to help them understand how L-tryptophan (or tryptamine) interacts with a carbonyl compound (such as benzaldehyde or an aldose).

Keywords: Maillard Reaction, tryptamine, tryptophan, tetrahydro- β -carbolines

ANTECEDENT

β -Carbolines are alkaloids representing a class of natural and synthetic compounds that contain an indole-derived moiety in their structure and exhibit a wide range of pharmacological effects, including antitumor,¹ antioxidant, antibacterial, antifungal, antimicrobial, antimalarial, anticonvulsant, and antiviral activities. These compounds hold great potential for the treatment of central nervous system disorders such as Alzheimer's disease,² Parkinson's disease, and depression.³

β -Carbolines are widely distributed in plants, foods, fruits, and vegetables. When foods are subjected to heat during processes such as cooking, frying, and roasting, they become rich in β -carbolines, as observed in roasted coffee, grilled meat, and toasted bread.⁴ These compounds are readily formed during food processing, production, and storage through the Pictet–Spengler reaction of tryptophan or arylethylamines with aldehydes or α -keto acids, or with aldoses (Maillard reaction). This reaction represents the main route for the formation of 1,2,3,4-tetrahydro- β -carbolines, which, upon oxidation, yield the corresponding β -carbolines.⁵ The Pictet–Spengler reaction is the commonly employed

method for the synthesis of β -carbolines due to its analogy with the biosynthetic pathway of these compounds. This method is currently considered an efficient approach for the synthesis of tetrahydro- β -carbolines, which are abundant in plants and the animal kingdom, many of which exhibit potent biological activity.

1,2,3,4-Tetrahydro- β -carboline-3-carboxylic acid, commonly found in smoked foods, arises from the condensation between L-tryptophan and formaldehyde.⁶

In this study, we report the optimization of the Pictet–Spengler reaction using tryptamine and tryptophan with substituted benzaldehydes to obtain the corresponding tetrahydro- β -carbolines. This work also aims to illustrate the Maillard reaction for undergraduate students in the Food Chemistry program during the Organic Chemistry III course (code 1628), allowing them to experimentally verify the interaction of L-tryptophan (or tryptamine) with a carbonyl compound (such as benzaldehyde or an aldose).

MATERIALS AND METHODS

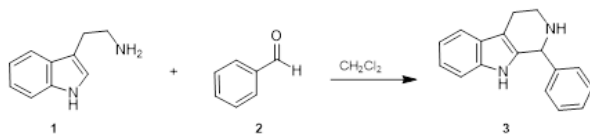
All reagents used for the synthesis were purchased from Sigma-Aldrich. The synthesized products were purified by column chromatography using silica gel (60 Å particle size) and dichloromethane/methanol mixtures as eluents. Pure products were characterized by infrared (IR) spectroscopy on a Perkin-Elmer Spectrum 400 FTIR/FIR spectrophotometer and by ¹H and ¹³C nuclear magnetic resonance (NMR) spectroscopy on a Varian VNMRs 400 MHz (9.4 T) spectrometer, in CDCl₃ and DMSO-d₆ solutions. Chemical shifts (δ) are reported in parts per million (ppm) using tetramethylsilane (TMS) as an internal reference. All reactions and purification steps were monitored by thin-layer chromatography (TLC) using Sigma-Aldrich silica gel 60 Å plates with a 250 nm fluores-

cent indicator, visualized under UV light and by spraying with ammonium cerium(IV) sulfate solution.

RESULTS AND DISCUSSION

The synthesis of tetrahydro- β -carbolines was carried out via the Pictet–Spengler reaction through the condensation of tryptamine or tryptophan with substituted benzaldehydes under acidic conditions (Scheme 1).

Initially, the optimization of the Pictet–Spengler reaction was performed using tryptamine 1 and benzaldehyde 2 to obtain 1-phenyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole 3, in the presence of acetic acid^{7,8} or trifluoroacetic acid,⁹ as shown in Scheme 1. Dichloromethane (CH_2Cl_2) was used as the solvent, varying its volume (1 or 2 mL) to assess the effect of concentration. Reaction temperature and time were also varied, and the results are summarized in Table 1.



Scheme 1. Pictet–Spengler Reaction for the Synthesis of Tetrahydro- β -carboline

Run	Acid	CH_2Cl_2 (mL)	Time (h)	Temperature	Yield (%)
1	CF_3COOH	2	24	ambiente	87.8
2	CH_3COOH	2	10	reflujo	57.1
3	CF_3COOH	2	2	reflujo	77.3
4	CF_3COOH	1	2	reflujo	95.3
5	CF_3COOH	1	1	reflujo	85.5

Table 1. Optimization Results for the Reaction between Tryptamine and Benzaldehyde

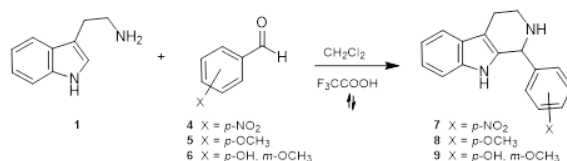
The results presented in Table 1 indicate that trifluoroacetic acid (CF_3COOH) is a more effective acid for promoting the condensation. When the reaction was performed with acetic acid (CH_3COOH), even after 10

h under reflux, the product was obtained in only 57.1% yield (entry 2). In contrast, when the reaction was carried out with CF_3COOH under stirring at room temperature for 24 h,⁹ the yield increased to 87.8%. However, it was possible to reduce the reaction time under reflux conditions. Additionally, the effect of concentration and reaction time was evaluated; lower concentrations and longer reaction times resulted in higher yields. Optimal reaction conditions were achieved under reflux for 2 h using 1 mL of solvent and trifluoroacetic acid, affording a yield of 95.3% (entry 4).

After optimizing the Pictet–Spengler condensation of tryptamine and benzaldehyde, the reaction was extended to tryptamine 1 and the following substituted benzaldehydes: 4-nitrobenzaldehyde 4, 4-methoxybenzaldehyde 5, and 4-hydroxy-3-methoxybenzaldehyde (vanillin) 6, as shown in Scheme 2.

In this case, the molar amount of the benzaldehyde derivatives was varied between 1.2 and 1.1 mmol, the volume of dichloromethane was adjusted to 1 or 2 mL, and the reaction times were modified. The results are summarized in Table 2.

The data show that the difference in yield between using 1.2 mmol and 1.1 mmol of benzaldehyde is not significant. However, the use of 1.1 mmol is preferable since it requires less reagent. Regarding concentration and reaction time, the same trend observed for benzaldehyde was noted; specifically, lower concentration and longer reaction times resulted in higher yields.



Scheme 2. Pictet–Spengler Reaction of Tryptamine with Substituted Aldehydes

Run	Benzaldehyde	Benzaldehyde (mmol)	CH ₂ Cl ₂ (mL)	Time (h)	Yield (%)
1	4	1.2	1	1	90.0
2	4	1.1	1	2	97.3
3	4	1.1	2	2	86.6
4	5	1.2	2	4	57.6
5	5	1.1	2	10	87.7
6	6	1.2	2	2	68.9
7	6	1.2	1	4	73.7
8	6	1.1	1	6	82.9

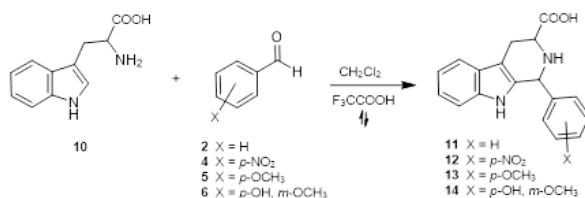
Table 2. Results of the Pictet–Spengler Reaction with Tryptamine

The results reflect a clear influence of the substituent group(s) present in the benzaldehyde derivatives. For instance, 4-nitrobenzaldehyde **4**, which bears a strongly electron-withdrawing group, increases the electrophilicity of the aldehyde carbonyl and requires only 2 h under reflux to afford the corresponding product in 97.3% yield (entry 2). In contrast, 4-methoxybenzaldehyde **5**, which contains a strong electron-donating group, requires 10 h under reflux to reach 87.7% yield (entry 5). Vanillin **6**, bearing two electron-donating substituents (hydroxy and methoxy groups), decreases the reactivity of the carbonyl even further, requiring 6 h under reflux to provide the product in 82.9% yield.

Subsequently, the Pictet–Spengler reactions were carried out between tryptophan **7** and the benzaldehyde derivatives 4-nitrobenzaldehyde **4**, 4-methoxybenzaldehyde **5**, and 4-hydroxy-3-methoxybenzaldehyde (vanillin) **6**, as shown in Scheme 3.

When the reaction was performed with tryptophan and substituted benzaldehydes, including benzaldehyde **2**, the same trend was observed: longer reaction times resulted in higher yields. Regarding the substituent effects, the nitro group, being strongly electron withdrawing, required only 2 h to achieve a 90.6% yield (entry 6). For electron-donating substituents such as methoxy, 10 h were ne-

cessary to reach 91.1% yield (entry 8). In the case of vanillin, which bears both hydroxy and methoxy groups, 6 h of reaction time afforded a yield of 70.7% (entry 11). These results confirm that electron-donating groups require longer reaction times (Table 3).



Scheme 3. Pictet–Spengler Reaction of Tryptophan with Substituted Aldehydes

Run	Benzaldehyde	CH ₂ Cl ₂ (mL)	Time (h)	Yield (%)
1	2	1	1	81.8
2	2	1	2	87.8
3	2	2	2	89.9
4	4	1	1	65.1
5	4	1	2	86.6
6	4	2	2	90.6
7	5	2	5	71.5
8	5	2	10	91.1
9	6	1	1	34.6
10	6	2	2	43.2
11	6	1	6	70.7

Table 3. Results of the Pictet–Spengler Reaction with Tryptophan

Based on the results obtained with both substrates (tryptamine and tryptophan) in the Pictet–Spengler reaction, the same trend was observed regarding the reactivity of the benzaldehyde substituents: reactions proceeded faster with the nitro group compared to benzaldehydes bearing electron-donating groups such as methoxy and hydroxy, which required longer reaction times. This can be explained by the reaction mechanism: in the first step, an imine is formed between the amino group of tryptamine (or tryptophan) and the benzaldehyde carbonyl. The next step involves a nucleophilic attack by the indole ring on

the electrophilic carbon of the imine. A para-substituted electron-withdrawing group ($-\text{NO}_2$) withdraws electron density through resonance, increasing the electrophilicity of this center and thus favoring the nucleophilic attack, resulting in a faster reaction.

All products were characterized by ^1H and ^{13}C nuclear magnetic resonance (NMR) spectroscopy and infrared (IR) spectroscopy.

Spectroscopic Data for 1-Phenyl-2,3,4,9-tetrahydro-1H-pyrido[3,4-b]indole (3).

RMN- ^1H (400 MHz, CDCl_3); δ (ppm): 7.60 (s, 1H), 7.47 (d, $J=4$ Hz, 1H), 7.28-7.24 (m, 3H), 7.21 (d, $J=4$ Hz, 2H), 7.11 (d, $J=4$ Hz, 1H), 7.07-7.02 (m, 2H), 5.08 (s, 1H), 3.27-3.24 (m, 1H), 3.05-3.01 (m, 1H), 2.87-2.82 (m, 1H), 2.74 (dt, 1H), 2.44 (ba, 1H).

RMN- ^{13}C (100 MHz, CDCl_3); δ (ppm): 141.52, 136.02, 134.25, 128.97, 128.73, 128.42, 127.45, 121.91, 119.55, 118.38, 110.99, 110.29, 58.13, 42.82, 22.45.

IR (ATR) cm^{-1} : 3300, 3058, 3030, 2946, 2841, 2884, 1917-1815, 1454, 1270, 1096, 740.

CONCLUSIONS

- The Pictet–Spengler reaction of tryptamine or tryptophan with substituted benzaldehydes was successfully optimized to afford the corresponding tetrahydro- β -carbolines.
- Experimental results indicate that the type of substituent on the benzaldehyde influences the reaction outcome, with the electron-withdrawing nitro group favoring the reaction.

ACKNOWLEDGMENTS

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