Cannabidiolic acid decarboxylation to Cannabidiol in Finola its Mechanism and Kinetics

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Abstract:

Cannabidiol (CBD) is a medicinal product used in medicine, cosmetics and food. CBD is produced by the decarboxylation process of cannabidiolic acid (CBDA) from heated cannabis (Cannabis sativa L.) crops. During the production of CBD enriched products, the decarboxylation reaction must be carried out at the right temperature and time. These tests were performed by heating the samples at 100, 110, 120, 130 and 140°C for 180 minutes. Data were checked every 20 minutes and analyzed for cannabinoid content using HPLC, after which kinetic parameters were calculated. Experimental results showed that CBDA in the sample decreased exponentially during the heating season. CBD increases at the same time and after a certain point CBD starts to decrease. Best is 140°C for 30 minutes. The molecular mechanisms of CBDA decarboxylation and CBD formation are explained at the level of reaction reactions, with changes in molecular orbitals, reaction steps, and molecular energy. Computational analysis confirmed that the CBDA decarboxylation mechanism is the direct β-ketoacid pathway. The CBDA decarboxylation process depends on the sample's time, temperature, and chemical composition.

Index terms: cannabidiolic acid (CBDA), Cannabidiol, HPLC, decarboxylation

1. Introduction:

Cannabinoids are organic compounds found in modern medicine, food, and cosmetics. Cannabidiol (CBD) and Δ9-tetrahydrocannabinol (Δ9-THC) are the most important cannabinoids in the cannabis plant, as they are the most abundant and have the most beneficial effects. Along with other cannabinoid compounds such as terpenes, they interact with cannabinoid and non-cannabinoid receptors to control pain, spasticity, sedation, and appetite[1]. They have neuroprotective, antioxidant, anti-inflammatory and anticonvulsant properties and indirectly induce tumor cell apoptosis during stimulation of ceramide secretion. Clinical trials have shown its effectiveness in the treatment of cancer, epilepsy, Parkinson's disease, Alzheimer's disease,

multiple sclerosis, anxiety, depression, and other conditions[2]. Cannabinoids are produced from cannabinoid acids synthesized as a resinous component in the glandular trichomes of Cannabis (Cannabis Sativa L.). Cannabinoid acids have lower bioavailability and less biological activity than decarboxylated (neutral) cannabinoids, as they are more hydrophilic and difficult to cross lipophilic barriers and bind with lower affinity to cannabinoid receptors.

Neutral cannabinoids are produced after heat assisted non-enzymatic decarboxylation of cannabinoid acids. Decarboxylation of cannabinoid acids is done, for example, by smoking, steaming, cooking or cooking hemp. In the production of cannabinoids and foods, decarboxylation is an important part of the production process that requires precise control. Therefore, knowledge of the chemistry, kinetics and thermodynamics of reactions is essential. Previous research found that during the decarboxylation of CBDA, the bond between the carboxyl carbon and the alpha carbon (C-C bond) is broken and releases CO2 to form CBD. For decarboxylation to occur, the electron breaking the C-C bond from the carboxylic acid must be stable. CBDA is an aromatic carboxylic acid with an aromatic ring that produces stable electrons generated by C-C bond cleavage as shown in Figure 1.

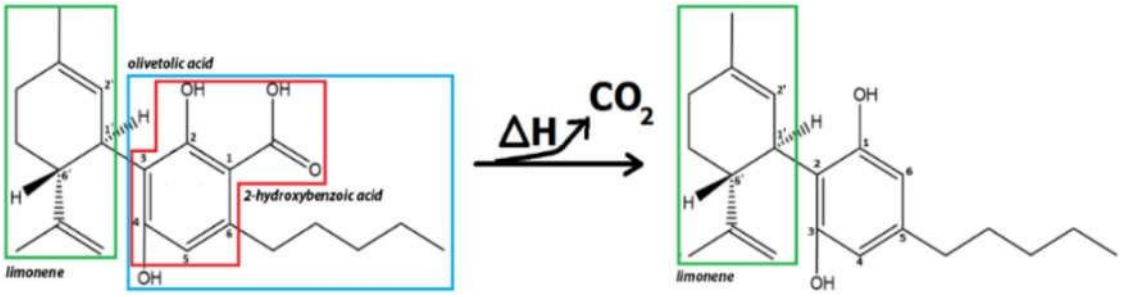

Fig.1 Chemical structure of CBDA (reactant) and CBD after decarboxylation (product). Squares: green: limonene; red: 2-hydroxybenzoic acid; blue: olivetolic acid (Color figure online)

2. Materials and methods

2.1. Ingredients :

Samples of Kief prepared from dried hemp [Cannabis sativa L. (Finola)] through a sieve of 100 mm Sieve. Cannabinoid content was analyzed by HPLC. Standard solutions used for HPLC analysis are CBD in acetonitrile and CBDA in methanol (Sigma-Aldrich Corporation). Decarboxylation Assay: The decarboxylation assay was performed in an oven at 100, 110, 120,

130 and 140°C for 180 minutes with aeration[3,4]. Experiments were carried out at each temperature separately. Measure out 500 mg into 9 Petri dishes 5 cm in diameter and 2 cm in height (one for each time point). Put the unfilled pot into the reaction vessel and heat to temperature for 1 hour. Remove the Petri dish from the reactor every 20 minutes for 180 minutes. Sample preparation for HPLC analysis: Weigh 200 mg sample from each can containing the decarboxylated content of into a 15 mL plastic tube (Mettler Toledo XS205 Dual series). Next, add 10 mL of ethanol (CARLO ERBA Reagents, Val de Reuil, France) using an automated machine. Transfer the plastic tube to an ultrasonic homogenizer (Bandelin Sonorex Digitec DT 100 H) at a frequency of 35 kHz for 15 minutes at 25 °C. After homogenization, 1.5 mL of sample was filtered through a filter (0.22 μm nylon pore size, Q-Max® RR syringe filter) into a dark bottle for HPLC analysis.

3. HPLC analysis:

Samples detection at 220 and 280 nm using a photodiode array detector (PDA) (Shimadzu, Shimadzu 20AD XR UFCL XR System, PDA SPD-) and analysis using HPLC at 40°C with a flow rate of 2 mL/min was done. M20A, RF-10A Fluorescence Detector and Phenomenex Kinetex® C18 Column 2.6, 100A, 100 × 4.6 mm, ID 1.7 μm particle size)[5,6]. Acetonitrile is made from water and 3-fluoroacetic acid (J. T. Baker; Deventer, Netherlands). Mobile phase A is water containing 2% acetonitrile and 0.1% 3-fluoroacetic acid. Mobile phase B is 2% water and 0 acetonitrile. 1% 3-fluoroacetic acid. Elution was done by the gradient method: 0.01–0.20 min 61% B, 0.20–0. 61-43% B at 21 min, 43-56% B at 0.21-50 min. HPLC analysis was conducted using the LC Solution Shimadzu 1.24 SP1 computer program. Identification, retention time and absorption spectra of CBDA and CBD were verified by comparing them with reference samples of CBDA and CBD. The major fraction of CBDA and CBD in the sample was determined by measuring the curve of the following points (mg/mL): 0.000, 0.001, 0.005, 0.010, 0. 050, 0,100, 0,167, 0,250, 0,500, 0. 750, 1,000; CBDA 0.9988 and CBD 0.9996 get R2. Computational **Analysis of CBDA Decarboxylation:** The CBDA decarboxylation process was investigated using Gaussian software to compare with Computational analysis and to supplement the experiments.CBDA has a hydroxyl group at the beta carbon of the aromatic ring, so it can be studied as a derivative of 2-hydroxybenzoic acid (2-HBA) or salicylic and olive oil, as shown in picture 1. For CBDA, the group closest to the decarboxylation site is expected to be involved

in this process. Other groups attached to the aromatic ring of CBDA (limene at position 3, hydroxyl at position 4, and pentyl at position 6) have little or no direct effect. These were used B3LYP/6-31G* for 2-HBA and HF/6-31G* for CBDA[7]. Use the QST2 module to determine the pattern of the first transition state and the SCAN module to determine the pattern of the second transition state. The structure of the reflection frequency for 2-HBA decarboxylation was determined as -2068.63 cm-1 for the first transition state and -902.30 cm-1 for the second transition state. For CBDA decarboxylation, the imaginary frequency of the first transition state is -2504.02 cm-1 and the imaginary frequency of the second transition state is -393. 25 cm-1 (Fig. 2).

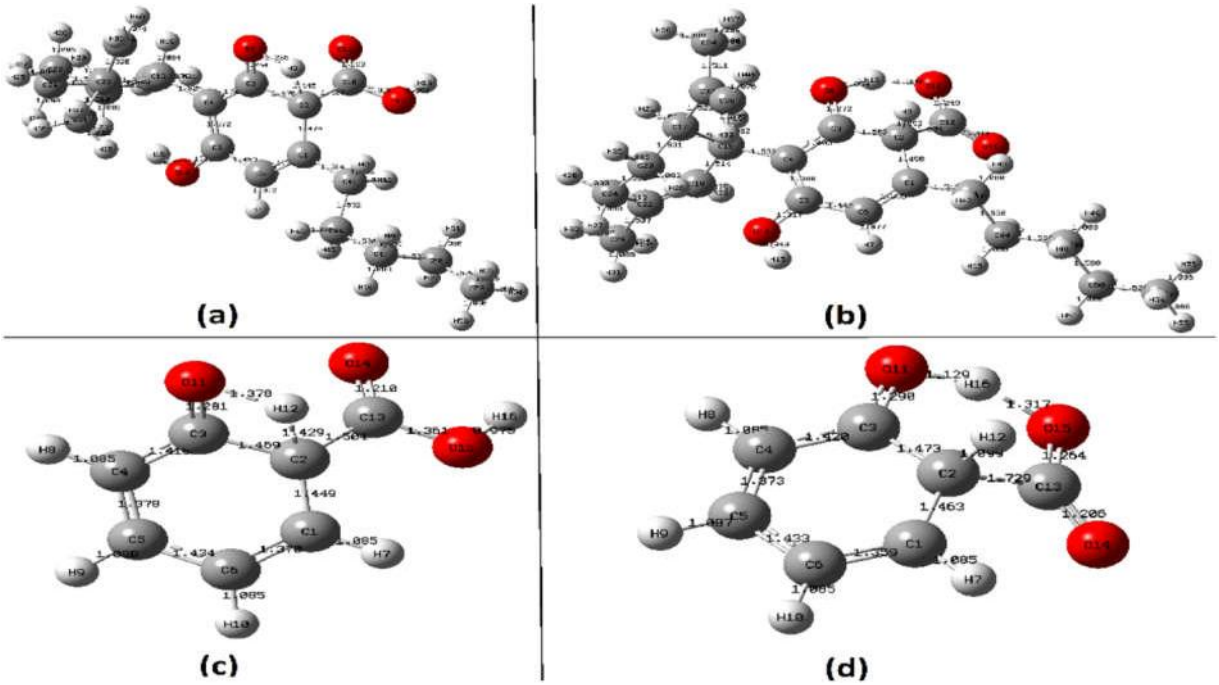

Figure 2 Conversion states for decarboxylation of CBDA and 2-HBA. The calculation is done with the Gaussian program. CBDA TS1; bCBDA TS2; c2-HBA TS1; d Computational analysis of 2-HBA TS2

4. CBDA decarboxylation:

GaussView 6.0.16. It is used to draw molecular models, add inputs and analyze calculated results. Calculations of decarboxylation mechanisms were done using Gaussian 09W Revision

D. 01. First, create formulas for reactants, transition states, intermediates, and products[8,9]. The geometry of the structure is then optimized and calculated using the 6-31G* basis set HF and B3LYP theory. Calculation of the decarboxylation mechanism of 2-HBA followed by the decarboxylation mechanism of CBDA was performed. Transition state models were calculated using the QST2 module. Potential benefits are calculated using the SCAN model. Use B3LYP/6-31G* for 2-HBA molecules and HF/6-31G* for CBDA molecules. The pattern of the transition state is determined by the frequency value. The transition states have a negative assumption, while the structures of reactants, intermediates and products have a positive assumption. Chapter by statistical analysis, ΔG‡ and ΔH‡ were determined according to the standard (298. 15 K and 1 air)[10]. Assuming that ΔH‡ does not change with temperature, ΔG‡1 at different temperatures can be calculated from ΔG‡2 determined at 298.15 K using the Van't Hoff equation. And combining ln(K‡).

4.1. CBDA Decarboxylation Mechanism:

Based on the data, experiment and calculation results, the CBDA decarboxylation mechanism can be predicted[11]. Decarboxylation occurs by switching from the most stable cis conformer to the anti conformer. Then the hydrogen is transferred from the hydroxyl group to the alpha carbon and tautomerization takes place via the ketone intermediate. Hydrogen is then transferred from the carboxyl group to the keto group and tautomerizes to the enol form. Then the C-C bond is broken, CO2 is lost and CBD is formed (Figure 3). As seen in the fig. 3 CBDA decarboxylation mechanism[12]. The cis isomer (R) converts to the trans isomer. Transfer of H + from -OH to α-C leads to tautomerization (T). Then H+ is transferred from -COOH to C=O and tautomerization (PT) occurs, the C-C bond is broken and CBD and CO2 are formed. The value of CBDA and CBD depends on the time and temperature of the process.

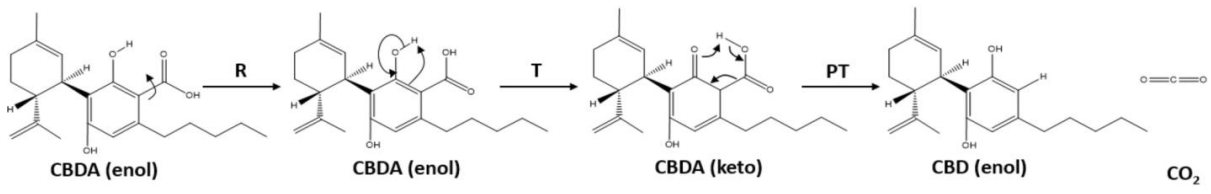

Fig.3 CBDA decarboxylation mechanism. Syn-conformer rotates (R) to anti-conformer. The transfer of H+ from –OH to α–C results in tautomerization (T). This is followed by the transfer

of H+ from –COOH to C=O and tautomerization (PT) with cleavage of the C–C bond and with the formation of CBD and CO2

The CBDA group generally decreases over time as shown by the green curve in Figure 4. CBDA is fully decarboxylated after 140 minutes at 130°C and 60 minutes at 140°C, but not completely at 100 -120°C. It is possible even after the end of the 180-minute experiment[13,14,15]. At the same time, the amount of CBD increases to a maximum, then the amount of CBD starts to decrease depending on the temperature, as shown by the red curve in Figure 4. The blue curve represents the degradation and shows the total loss of CBD content (eg TOTAL = 0.877 × CBDA + CBD).

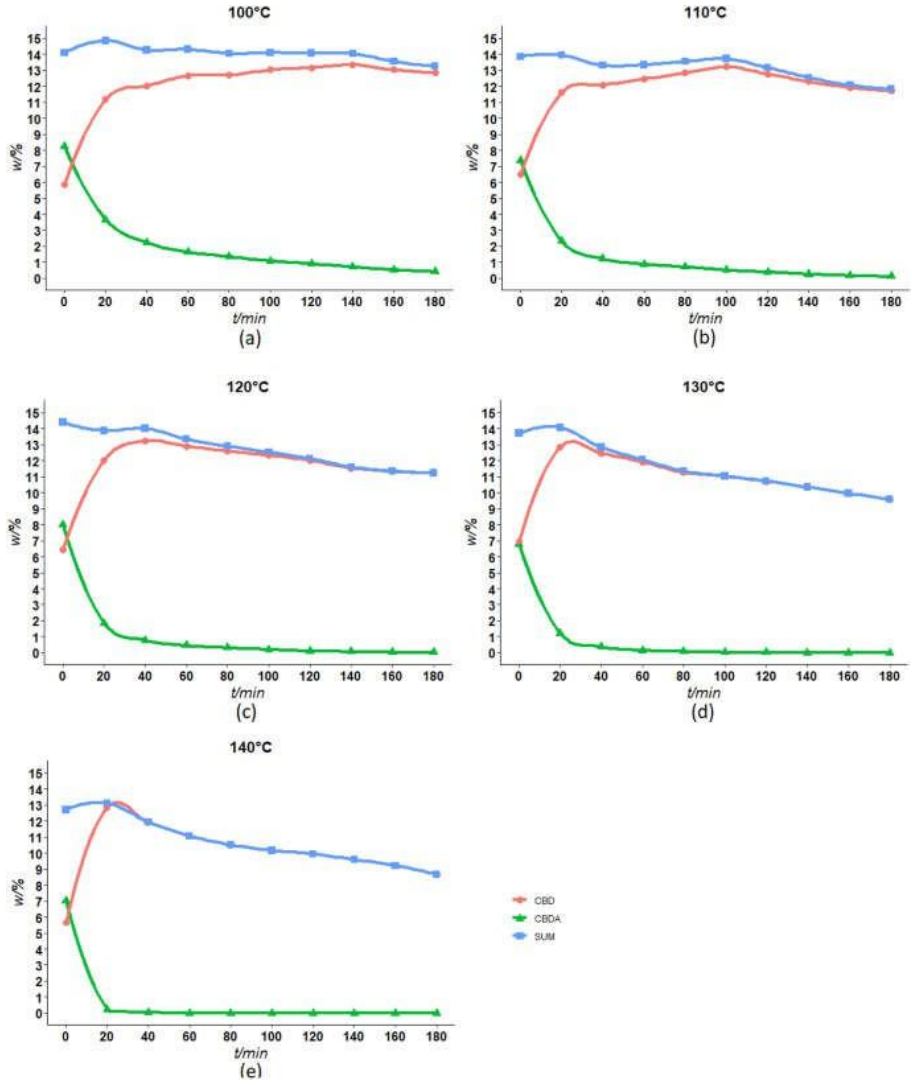

Figure 4 Decarboxylation of CBDA at 100°C; b 110 °C; 120 °C; d 130 °C; 140 °C. The experiments were carried out individually by heating the samples in the oven at the same temperature for 180 minutes. HPLC analysis was performed every 20 minutes. Results are presented by mass (w/%) versus time (t/min).

5. Conclusion:

CBD starts to degrade after a while at high temperatures. The CBDA decarboxylation process depends on the sample's time, temperature, and chemical composition. Experimentally, the best way to convert CBDA to CBD is to heat at 140°C for 30 minutes. The steps presented in our work can be used to build models for different personal use cases[16]. Statistical analysis

showed that the CBDA decarboxylation process is a direct β-ketoacid pathway in which the reaction is dependent on the transfer of hydrogen from the hydroxyl group to the α-ring carbon, followed by hydrogen transfer from the carboxyl group to the keto group and the α carbon. to form the carboxyl group, thus forming CBD. Analysis of the reaction mechanism shows that the rate of decarboxylation can be controlled using different CBDA salts in the future, thereby affecting the structure of the intermediate state.

6. References:

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