Melatonin/nanoclay hybrids for skin delivery

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Table of Contents

**1. Methodology**

**1.1. Atomistic Simulation Models Figure S1**

**2. Results**

**2.1. Stability of pristine melatonin in organic solvents**

**2.2. Calculated X-ray diffraction Figure S2**

**2.3. Optimized melatonin molecular structure Figure S3**

**2.4. Main geometrical features of the optimized melatonin molecule Table S1**

**2.5. Main geometrical features of the optimized melatonin crystal Table S2**

**2.6. Experimental FTIR spectroscopy**  **Figure S4**

**1. Methodology**

* 1. *Atomistic Simulation Models*



**Figure S1.** MEL-Montmorillonite-Water model used in the AIMD runs (side (a) and top (b) views). The simulation cell is shown in blue lines. Colour code: O in red, H in white, N in blue, C in cyan, Na in green, Si in yellow, Al in pink, Mg in light blue.

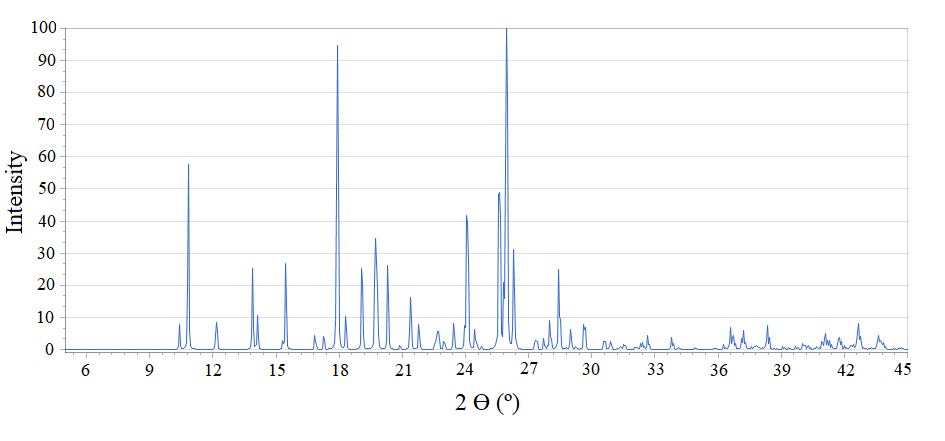
2. Results and Discussion

*2.1. Stability of pristine melatonin in organic solvents*

Due to the low solubility of MEL in water and the high affinity of clays to water, other solvents in which the drug is soluble were tested. Therefore, preliminary explorations related with the stability of MEL in different organic solvents were performed for the preparation of hybrid materials (MEL-clay) and also for setting up an analytical protocol. Firstly, the stability of MEL in ethanol 96% and 99% were studied. After dissolving the MEL in ethanol 96% and 99% at zero time, the solutions were measured in HPLC and the degradation of the drug was observed. The results obtained allow us to know that ethanol could not be used for the dissolution of MEL, since ethanol produces an immediate degradation of MEL, despite previous studies used ethanol as a solvent for MEL [1–4]. The same results were obtained by dissolving MEL in methanol, isopropanol and acetone. Therefore, these organic solvents could not be used to dissolve the drug. On the contrary, the stability of MEL in acetonitrile was observed until one month later, and the solubility of MEL in acetonitrile was approximately 41 mg/mL. Since the solubility of the drug in acetonitrile is high and MEL is stable in acetonitrile, this organic solvent was chosen to solubilize MEL in the rest of experimental tests.

**2.2. Calculated X-ray diffraction**

The X-ray diffraction of the melatonin crystal[5] was calculated with the Reflex Powder Diffraction module [6]. In Figure S2, the main melatonin peaks appeared at 16.5°, 19.0°, 24.5°, 25.5° and 26.5° 2θ according with the experimental results (Figure 1) and others spectrum previously reported [7] with the only differences are in the relative intensity of peaks.



**Figure S2.** Calculatedpowder X-ray diffraction of crystal structure of melatonin.

**2.3. Optimized melatonin molecular structure**

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**Figure S3.** TheMEL molecular structure optimized with Dmol3 (a) and INTERFACE FF (b). Two orientations are shown**.**

**2.4. Main geometrical features of the optimized melatonin molecule**

**Table S1.** Main bond lengths (Å) and angles (°) values of the optimized MEL molecular structure.[a]

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Features** | **EXP** | **UF** | **CF** | **INTERFACE** | **DFT** |
| d(H3C-CO) | 1.492 | 1.497 | 1.507 | 1.511 | 1.528 |
| d(C=O) | 1.239 | 1.220 | 1.218 | 1.228 | 1.237 |
| d(OC-NH) | 1.329 | 1.371 | 1.344 | 1.327 | 1.376 |
| d(HN-CH2) | 1.445 | 1.465 | 1.466 | 1.476 | 1.468 |
| d(H2C-CH2) | 1.504 | 1.534 | 1.515 | 1.515 | 1.534 |
| d(H2C-C=C) | 1.497 | 1.507 | 1.491 | 1.532 | 1.508 |
| d(HC-NH) | 1.364 | 1.352 | 1.380 | 1.411 | 1.388 |
| d(C=C-O) | 1.382 | 1.410 | 1.385 | 1.396 | 1.387 |
| d(O-CH3) | 1.416 | 1.427 | 1.424 | 1.448 | 1.433 |
| H3C-O-C | 117.3 | 118.2 | 117.5 | 123.7 | 117.4 |
| O=C-N | 121.2 | 120.4 | 122.3 | 124.1 | 122.2 |
| H3CO-C-C=C | 0.1 | 0.0 | 2.1 | 1.5 | 0.4 |
| O=C-N-H | 179.0 | 180.0 | 178.0 | 179.8 | 179.7 |
| H-N-C-C | 4.1 | 0.0 | 96.9 | 3.8 | 0.0 |
| C-C-C=CN | 5.5 | 0.0 | 90.1 | 98.9 | 9.5 |

[a] EXP: experimental data in crystal lattice [5]; UF: calculated with Universal FF using Forcite code; CF: calculated with COMPASS and FORCITE code; INTERFACE: calculated with INTERFACE FF and DISCOVER code; DFT: calculated with DMol3.

**2.5. Main geometrical features of the optimized melatonin crystal**

**Table S2.** Main bond lengths (Å) and angles (°) values of the crystal structure of melatonin optimized at variable volume.[a]

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Features** | **EXP** | **UF** | **CF** | **INTERFACE** | **CA** | **CA\_TS** | **CA\_G** |
| d(H3C-CO) | 1.492 | 1.495 | 1.487 | 1.518 | 1.508 | 1.507 | 1.507 |
| d(C=O) | 1.239 | 1.219 | 1.217 | 1.207 | 1.255 | 1.256 | 1.257 |
| d(OC-NH) | 1.329 | 1.371 | 1.328 | 1.336 | 1.345 | 1.339 | 1.337 |
| d(HN-CH2) | 1.445 | 1.464 | 1.474 | 1.471 | 1.457 | 1.452 | 1.453 |
| d(H2C-CH2) | 1.504 | 1.533 | 1.521 | 1.511 | 1.525 | 1.523 | 1.522 |
| d(H2C-C=C) | 1.497 | 1.508 | 1.514 | 1.504 | 1.500 | 1.495 | 1.492 |
| d(HC-NH) | 1.364 | 1.350 | 1.376 | 1.381 | 1.377 | 1.376 | 1.377 |
| d(C=C-O) | 1.382 | 1.409 | 1.377 | 1.385 | 1.377 | 1.376 | 1.377 |
| d(O-CH3) | 1.416 | 1.426 | 1.420 | 1.423 | 1.424 | 1.423 | 1.426 |
| H3C-O-C | 117.3 | 118.6 | 116.2 | 117.9 | 117.3 | 116.8 | 116.5 |
| O=C-N | 121.2 | 120.4 | 119.8 | 124.6 | 122.0 | 121.8 | 121.8 |
| H3CO-C-C=C | 0.1 | 10.8 | 4.9 | 0.6 | 2.5 | 0.8 | 3.6 |
| O=C-N-H | 179.0 | 179.1 | 179.1 | 168.7 | 175.1 | 174.9 | 173.1 |
| H-N-C-C | 4.1 | 1.0 | 11.3 | 24.8 | 15.9 | 11.2 | 6.4 |
| C-C-C=CN | 5.5 | 4.6 | 5.2 | 12.1 | 12.4 | 6.2 | 6.5 |
| =NH···OC | 2.035 | 2.897 | 2.079 | 1.847 | 1.939 | 1.832 | 1.814 |
| NH···OC | 2.157 | 2.654 | 2.364 | 1.995 | 2.016 | 1.904 | 1.849 |
| H3CO···HC= | 2.612 | 2.517 | 2.948 | 2.574 | 2.750 | 2.361 | 2.387 |
| H3CO···H2C | 2.861 | 2.935 | 3.160 | 3.031 | 3.072 | 2.745 | 2.641 |

[a]EXP: experimental data [5]; UF: calculated with the Universal FF; CF: calculated with Compass FF; INTERFACE: calculated with INTERFACE FF; CA: calculated with CASTEP; CA\_TS: calculated with CASTEP and Tkatchenko-Scheffler dispersion correction; CA\_G: with CASTEP and Grimme dispersion correction.

**2.6. Experimental FTIR spectroscopy**

For a better visualization of the infrared bands, the zoom of the experimental spectrum has been shown (Figure S4). The results were described in the manuscript.



**Figure S4.** Zoom views of the experimental FTIR spectrum of melatonin (Figure 6).

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