Characterization by X-ray powder diffraction of alpha lipoic acid

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Alpha lipoic acid (ALA) $C_8H_{14}O_2S_2$ is a naturally occurring compound that is synthesized in small amounts by plants and animals, including humans. ALA is covalently bound to specific proteins, which function as cofactors for several important mitochondrial enzyme complexes and studies suggest that they might help with type 2 diabetes. In the Cambridge Structural Database, there are four entries related to this compound: two for lipoic acid and two for complexes. In the Powder Diffraction File-4, two experimental unindexed patterns are reported. The material crystallizes in a monoclinic crystal system, space group $P2_1/a$ and cell parameters $a = 9.237$ (1) Å, $b = 9.960$ (1) Å, $c = 11.787$ (2) Å, $\beta = 109.13$ (1)°, and $V = 1024.6$ (2) Å^3. © *2016 International Centre for Diffraction Data.*
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Key words: alpha lipoic acid, characterization, X-ray powder diffraction

I. INTRODUCTION

A study by X-ray powder diffraction of alpha lipoic acid (ALA, Figure 1) was carried out as part of our continuing interest on the structural characterization of active pharmaceutical ingredients and the formation of possible new polymorphs. The chemical nature of the material and their thermal stability were examined using spectroscopy (FTIR and RAMAN) and thermal analysis [thermogravimetric analysis (TGA)–differential scanning calorimetry measurements (DSC)].

ALA is a naturally occurring compound that is synthesized in small amounts by plants and animals, including humans. ALA is covalently bound to specific proteins, which function as cofactors for several important mitochondrial enzyme complexes (Carreau, 1979; Reed, 2001). ALA is used for its antoxidant effects in the treatment of diabetic neuropathy. It has been tried in the treatment of liver dysfunction and in subacute necrotizing encephalopathy (Sweetman, 2009). A search of the Cambridge Structural Database, V. 1.18 (Allen, 2002), of the lipoic acid omitting hydrogen atoms resulted in four entries, two of them for lipoic acid (Refcodes: THOCAR, THOCAR01) and two for complexes. The Powder Diffraction File (PDF)-4 also contain two entries with an experimental unindexed pattern (PDF: 00-007-0585, 00-007-0553) (ICDD, 2012).

In the pharmaceutical industry, it is important to properly characterize all the materials, active, and inactive ingredients (excipients), involved in the manufacture of a drug, in order to determine possible amorphous, polymorphs, solvates, hydrates forms, just as transitions of crystalline phases in the solid state, and physicochemical stability caused by the different processes they are subjected during the preparation of a formulation. X-ray powder diffraction to play an important role in the characterization in solid state of materials associated with the formulation of pharmaceutical products.

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II. EXPERIMENTAL

A. Crystallization and density determination

Alpha lipoic acid, a sample of commercial formulation, was dissolved in mixture ethanol:water. After filtering the extract was allowed to evaporate at room temperature. The density of crystal obtained of crystallization process was determined by the flotation method using potassium iodide solution at various concentrations.

B. IR and RAMAN spectroscopy

The Fourier transform infrared spectroscopy (FT-IR) spectra were recorded in KBr pellets, using an IS50 FT-IR Nicolet Thermo Scientific spectrophotometer, a register range 4000–400 cm^{-1}, with 32 scans per sample and an optical speed of 0.4747 cm s^{-1}. The RAMAN spectra was performed in a LabRam HR Evolution using a laser at 532 nm with 100% attenuation to 3 s, a 100× objective applying fluorescence correction in a range of 4000–200 cm^{-1}.

C. Thermal analysis, TGA–DSC

TGA and DSC were performed in a Thermal Analyzer DTA/DSC Instrument Serie Discovery, With mass flow 50.0 ml min^{-1}, equilibrate at 2500 °C, ramp at 1000 °C min^{-1} to 50,000 °C for TGA and 55,000 °C for DSC analysis. Both experiments were performed under a nitrogen atmosphere.

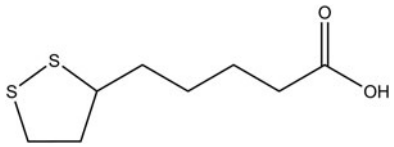

Figure 1. Chemical structure of alpha lipoic acid.

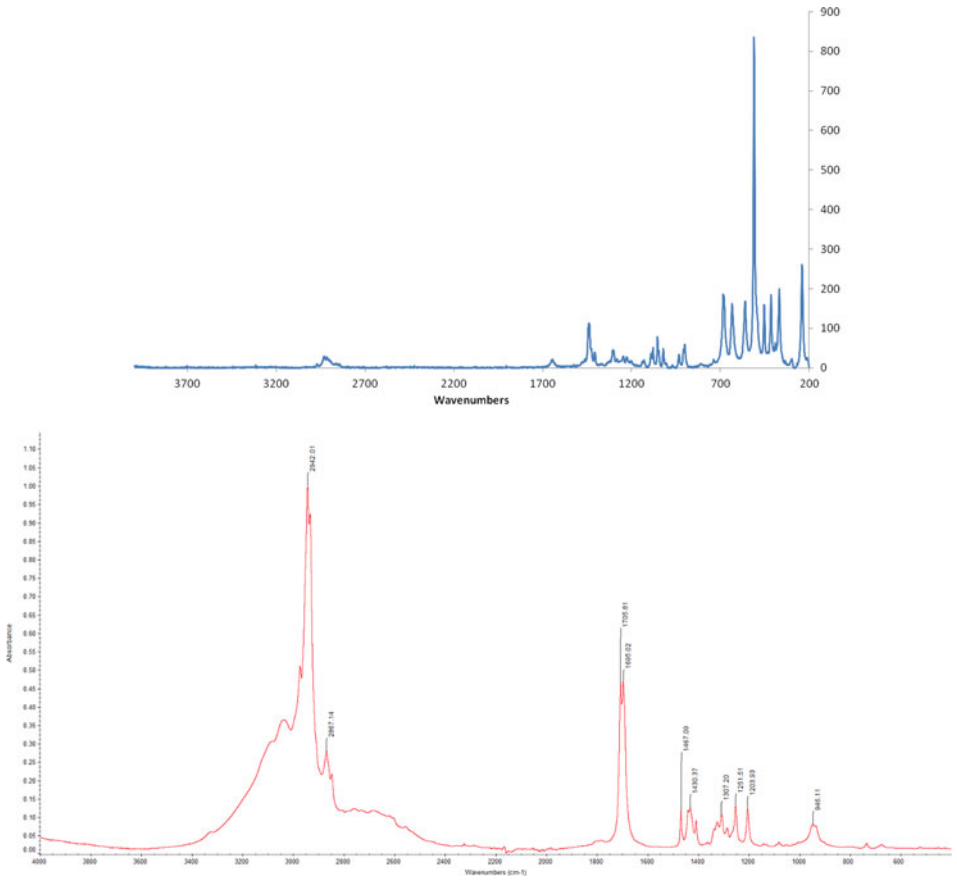

Figure 2. (Color online) RAMAN spectrum (top) and IR spectrum (below) of lipoic acid.

D. X-ray powder diffraction data collection

Powder diffraction patterns were recorded on a BRUKER D8 ADVANCE diffractometer using Cu$K\alpha$ radiation (λ = 1.5406 Å), operating at 40 kV and 40 mA. The patterns were recorded in steps of 0.0156° (2θ), from 4° to 60° at 1 s step^{-1}. The diffractometer was equipped with the primary and secondary Soller slits of 2.5°, divergence slit of 0.6 mm, nickel filter of 0.02 mm, and a LynxEye detector. PowderX program (Dong, 1999) was used to remove the background (Sonneveld and Visser, 1975), smoothing (Saviztky and Golay, 1964), to eliminate the $K\alpha_2$ component (Rachinger, 1948) and the second derivative method was used to determine the peak-observed positions and intensities.

III. RESULTS AND DISCUSSION

Yellow blocks were obtained by the slow evaporation experiment. In the RAMAN spectrum (top, Figure 2), the strong band corresponding to the vibration S–S is observed at 511 cm^{-1}. The vibration C–S appears at 679 cm^{-1} and a

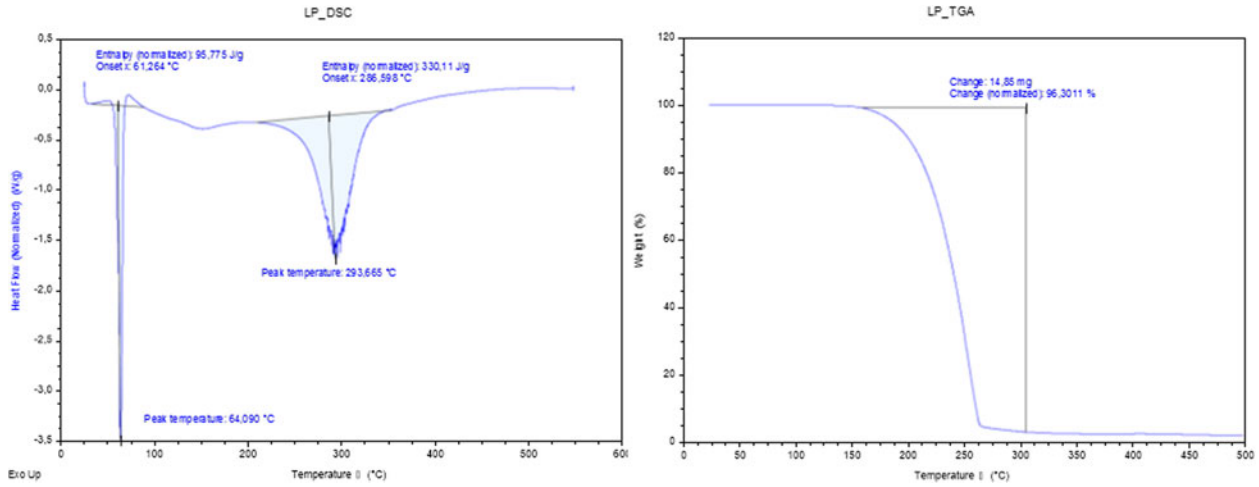

Figure 3. (Color online) DSC (right) curve and TGA curve (left) of lipoic acid.

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TABLE I. X-ray powder diffraction data of alpha lipoic acid.

$2\theta_{obs}$ (°)	d_{obs} (Å)	I	h	k	l	$2\theta_{calc}$ (°)	d_{calc} (Å)	$\Delta 2\theta$ (°)
7.9300	11.140	110	0	0	1	7.933	11.1358	0.003
11.902	7.4297	108	0	1	1	11.911	7.4239	0.009
13.458	6.5740	7	−1	1	0	13.479	6.5638	0.021
13.851	6.3884	119	−1	1	1	13.859	6.3848	0.008
15.924	5.5611	3	0	0	2	15.904	5.5679	−0.02
17.271	5.1303	42	1	1	1	17.277	5.1286	0.006
17.796	4.9801	396	0	2	0	17.796	4.9801	0.000
18.158	4.8816	162	−1	1	2	18.161	4.8808	0.003
19.230	4.6118	9	−2	0	1	19.247	4.6079	0.017
19.494	4.5500	1	0	2	1	19.510	4.5462	0.016
20.305	4.3700	14	2	0	0	20.336	4.3634	0.031
20.517	4.3254	2	1	2	0	20.517	4.3253	0.000
20.764	4.2745	65	−1	2	1	20.771	4.2729	0.007
21.232	4.1813	2	−2	1	1	21.228	4.1820	−0.004
21.340	4.1604	12	−2	0	2	21.345	4.1594	0.005
22.260	3.9905	3	2	1	0	22.225	3.9967	−0.035
23.204	3.8302	622	1	2	1	23.221	3.8275	0.017
23.333	3.8093	1000	1	1	2	23.340	3.8081	0.007
23.941	3.7139	188	0	0	3	23.954	3.7119	0.013
24.441	3.6391	66	−1	1	3	24.448	3.6381	0.007
25.589	3.4784	110	0	1	3	25.590	3.4782	0.001
25.831	3.4463	8	2	1	1	25.825	3.4471	−0.006
25.936	3.4326	6	−2	0	3	25.899	3.4374	−0.037
27.441	3.2477	2	−2	1	3	27.426	3.2494	−0.015
27.950	3.1897	14	−2	2	2	27.926	3.1924	−0.024
28.014	3.1825	10	0	3	1	28.022	3.1817	0.008
28.760	3.1016	6	1	3	0	28.747	3.1031	−0.013
28.926	3.0842	20	−1	3	1	28.932	3.0835	0.006
29.821	2.9937	3	2	0	2	29.845	2.9913	0.024
29.992	2.9770	5	0	2	3	30.000	2.9762	0.008
30.280	2.9531	6	2	2	1	30.204	2.9566	−0.036
30.445	2.9337	9	−3	1	1	30.431	2.9350	−0.014
30.799	2.9008	3	1	3	1	30.775	2.9030	−0.024
30.949	2.8871	18	−3	1	2	30.959	2.8862	0.010
31.239	2.8617	15	2	1	2	31.195	2.8649	−0.035
31.301	2.8554	21	−1	3	2	31.297	2.8558	−0.004
31.634	2.8261	46	−1	1	4	31.641	2.8255	0.007
31.969	2.7990	33	−2	0	4	31.942	2.7996	−0.007
32.117	2.7847	48	0	0	4	32.126	2.7840	0.009
33.280	2.6957	3	−2	1	4	33.215	2.6951	0.007
33.358	2.6839	4	0	1	4	33.393	2.6812	0.035
34.269	2.6146	4	1	2	3	34.267	2.6148	−0.002
34.645	2.5871	3	1	3	2	34.667	2.5855	0.022
34.756	2.5791	4	−3	2	2	34.749	2.5795	−0.007
35.382	2.5349	24	−1	2	4	35.366	2.5359	−0.016
35.684	2.5141	1	−3	2	0	35.717	2.5118	0.033
36.047	2.4896	18	0	4	0	36.040	2.4900	−0.007
36.285	2.4738	20	0	3	3	36.273	2.4746	−0.012
36.566	2.4554	2	2	0	3	36.533	2.4576	−0.033
36.800	2.4404	25	−2	2	4	36.800	2.4404	0.000
36.956	2.4304	10	0	2	4	36.962	2.4300	0.006
37.070	2.4232	7	−3	2	3	37.078	2.4227	0.008
37.608	2.3898	11	−2	3	3	37.636	2.3880	0.028
37.747	2.3813	26	−3	1	4	37.747	2.3813	0.000
38.937	2.3112	7	3	2	1	38.893	2.3137	−0.044
39.060	2.3042	19	−4	0	2	39.065	2.3039	0.005
39.326	2.2892	3	−1	1	5	39.355	2.2876	0.029
39.589	2.2746	16	−1	4	2	39.579	2.2752	−0.010
39.966	2.2540	8	−2	1	5	39.959	2.2544	−0.007
40.193	2.2429	9	−4	1	2	40.140	2.2447	−0.033
40.434	2.2290	21	−4	1	1	40.433	2.2291	−0.001
40.533	2.2238	17	2	3	2	40.561	2.2223	0.028
40.995	2.1998	19	−3	2	4	40.989	2.2001	−0.006
41.258	2.1864	8	1	2	4	41.285	2.1850	0.027
41.425	2.1755	7	0	1	5	41.514	2.1735	0.039
41.568	2.1708	3	−4	1	3	41.528	2.1728	−0.040

Continued

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TABLE I. Continued

$2\theta_{obs}$ (°)	d_{obs} (Å)	I	h	k	l	$2\theta_{calc}$ (°)	d_{calc} (Å)	$\Delta 2\theta$ (°)
41.721	2.1632	1	2	4	0	41.732	2.1627	0.011
42.263	2.1367	6	−2	4	2	42.268	2.1365	0.005
42.462	2.1271	7	−3	3	3	42.438	2.1283	−0.024
43.094	2.0974	2	−2	2	5	43.062	2.0989	−0.032
43.552	2.0773	8	−4	2	1	43.508	2.0784	−0.024
43.747	2.0676	17	0	4	3	43.741	2.0679	−0.006
43.848	2.0631	19	2	4	1	43.888	2.0613	0.040
44.039	2.0546	13	3	3	1	44.068	2.0533	0.029
44.463	2.0359	1	−4	1	4	44.466	2.0358	0.003
44.856	2.0190	0	4	0	1	44.839	2.0197	−0.017
45.420	1.9975	1	4	2	0	45.346	1.9983	−0.020
45.789	1.9800	2	4	1	1	45.803	1.9795	0.014
45.983	1.9721	6	−3	3	4	45.971	1.9726	−0.012
46.164	1.9648	4	−3	2	5	46.154	1.9652	−0.010
46.240	1.9618	5	1	3	4	46.241	1.9617	0.001
46.375	1.9564	4	3	1	3	46.399	1.9554	0.024
46.486	1.9522	3	−2	0	6	46.433	1.9541	−0.047
46.738	1.9420	1	−1	5	0	46.736	1.9421	−0.002
46.879	1.9365	1	−1	5	1	46.859	1.9373	−0.020
47.326	1.9192	9	−4	2	4	47.330	1.9191	0.004
47.471	1.9137	4	−1	1	6	47.471	1.9137	0.000
47.802	1.9012	8	−4	0	5	47.816	1.9007	0.014
47.867	1.8988	8	−2	3	5	47.871	1.8987	0.004
48.031	1.8927	7	−4	3	2	48.028	1.8928	−0.003
48.185	1.8861	3	3	3	2	48.257	1.8844	0.047
48.464	1.8768	5	−1	5	2	48.464	1.8768	0.000
48.698	1.8683	5	−4	1	5	48.734	1.8670	0.036
48.922	1.8603	17	−2	4	4	48.914	1.8606	−0.008
49.011	1.8571	18	0	0	6	49.044	1.8560	0.033
50.001	1.8227	6	−4	3	0	49.983	1.8233	−0.018
50.290	1.8129	5	2	5	0	50.312	1.8121	0.022
50.822	1.7951	5	−2	5	2	50.776	1.7966	−0.046
52.324	1.7471	4	−3	4	4	52.317	1.7473	−0.007
52.740	1.7343	5	−5	1	4	52.740	1.7343	0.000
53.273	1.7182	6	−4	0	6	53.254	1.7187	−0.019
54.531	1.6815	9	−1	3	6	54.531	1.6815	0.000
55.301	1.6599	5	−4	4	3	55.305	1.6597	0.004
55.938	1.6425	5	0	6	1	55.959	1.6419	0.021
56.826	1.6189	4	−3	1	7	56.841	1.6185	0.015

strong band at 240 cm^{-1} corresponds to aliphatic chains. In the FTIR spectrum (below, Figure 2), the C–H stretching appears at 2942–2967 cm^{-1}. The absorption band C = O is observed at 1705 cm^{-1}. Between 3400 and 2600 cm^{-1} appears the stretching band O–H of carboxylic group. Both spectrums clearly show the chemical nature of the compound under study.

The lipoic acid crystallized melts at 64 °C, according to the first endotherm in DSC analysis (right, Figure 3). This is a similar behavior to reported, 60–62 °C (Sweetman, 2009). The TGA curve (left, Figure 3) shows that the material is stable up to 150 °C, experiencing weight loss 93.30% at ∼160–260 °C. This loss is associated with the second endotherm in the DSC curve. These correspond to the total decomposition of ALA.

The experimental powder diffraction pattern is different from those contained in the PDF-4 database but similar to the pattern calculated using THOCAR crystal data (Figure 4). The indexing of pattern recorded for the crystallized material carried out with DICVOL14 (Boultif and Loüer, 2014). The analysis of the entire pattern (102 diffraction maxima) with NSB*AIDS83 (Mighell *et al.*, 1981), resulted in a unit-cell monoclinic with parameters: $a = 9.237(1)$ Å, $b = 9.960(1)$ Å, $c = 11.787(2)$ Å, $\beta = 109.13(1)°$, and $V = 1024.6(2)$ $Å^3$; and de Wolf (1968) and Smith and Snyder (1979) figures of merit associated are $M20 = 28.5$ and $F30 = 63.5$ (0.0124, 38). This pattern, in PD3 format, will be submitted for inclusion in the PDF. The set of reflections observed are consistent with space group $P2_1/a$ and was estimated by the CHEKCELL program (Laugier and Bochu, 2002). Table I contains the corresponding powder diffraction data. The fitting of the whole pattern with the Le Bail algorithm in FULLPROF program (Rodriguez-Carvajal, 1990) accounts for all the diffraction maxima recorded.

The $Z = 4$ was estimated by density obtained by the flotation method [$d = 1.35(1)$ g cm^{-3}], and the calculate density, using this Z-value and the volume obtained from the indexing process, was 1.34 g cm^{-3}, similar to measured density.

SUPPLEMENTARY MATERIAL

The supplementary material for this article can be found at https://doi.org/10.1017/S0885715616000658.

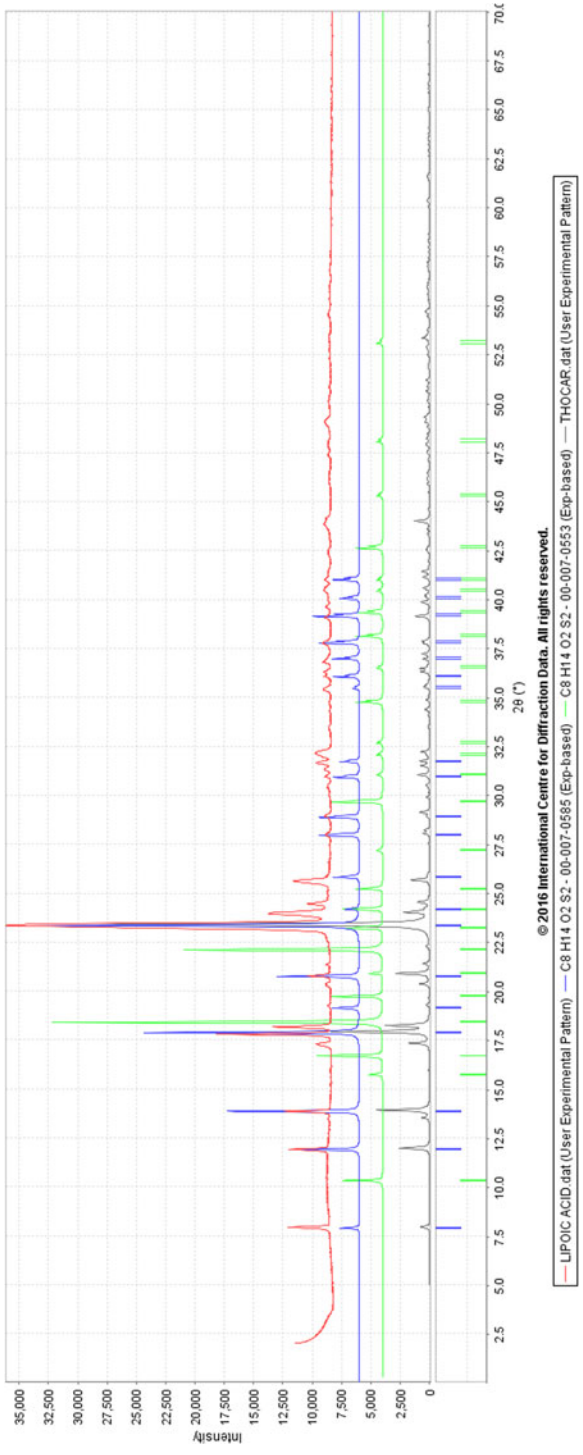

Figure 4. (Color online) Comparison of the powder diffraction pattern of lipoic acid (red) with PDF 00-007-0585 (blue), PDF 00-007-0553 (green), and pattern calculated using THOCAR data (gray).

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