# Comparative Stability of Levodopa Under Thermal Stress in both Oxidative and Inert Media

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*This study aims towards a comparative stability of an amino-acid under thermal stress in both oxidative and inert media, namely Levodopa, which is up to the date the primary antiparkinsonian medication. The identity and purity of the sample was firstly confirmed by UATR-FTIR spectroscopy, and later thermal analysis was employed as an investigational tool for a preliminary view over the stability of the compound. Thermal analysis indicate a similar thermal stability in both media, with no mass loss up to 250 °C, but HF data indicate different thermal events as contributions to degradation.*

*Keywords: levodopa, thermal stability, inert atmosphere, oxidative atmosphere, FTIR*

Parkinson's disease, a progressive neurodegenerative disorder, has become in the last decades one of the most common illnesses that affects elderly people [1]. According to literature, this movement disorder is caused by the selective degradation of neurons from the substantia nigra in the Central Nervous System (CNS), this, in turn, being the underline cause of a low dopamine level [2].

Since the discovery of its effectiveness in Parkinson's treatment in the 1960's [3], levodopa, or more commonly referred to as L-Dopa, has become a main component in the treatment procedure. Although recently a series of molecules have been tested and introduced in therapy, among most patients L-Dopa remains the primary antiparkinsonian medication [4]. Because this line of treatment is based on the replacement of the neurotransmitter dopamine in the CNS, a molecule that is able to cross the blood-brain barrier is required, thus the need for L-Dopa [5]. Dopamine in itself is not able to cross this barrier, but levodopa uses a transporter to reach its target [6,7]. However, a conversion process from levodopa to dopamine is needed, this requiring an enzymatic catalyzer, dopa-decarboxylase. Because the enzyme is not specific to the CNS, but exists also in the periphery tissues, in this case the transformation of levodopa occurs mainly in the liver, kidneys, stomach and in the small bowl wall [6]. In order to avoid a need to increase the drug's concentration, and with it the side effects, a dopa-decarboxylase peripheral inhibitor is usually associated in the levodopa formulations, thus assuring that the conversion takes place mainly where dopamine is needed, in the CNS [4].

Levodopa ((-)-3-(3,4-Dihydroxyphenyl)-L-alanine, see structure in Figure 1) presents an amino acid residue attached to a benzene ring that has two hydroxyl groups [8]. Although the substance is stable until a relatively high temperature (melting point of 284-285°C), its low water solubility (5000 mg/L at 20°C) consists a problem in the drug formulation process [9,10]. Also, an important aspect in the stability area is related to levodopa's susceptibility to decarboxylation at the side chain but also to chemical oxidation, only a very small percentage of the administrated oral dose being able to reach the brain, as previously discussed [11].

Although a series of symptoms are alleviated during the treatment with L-Dopa formulations, such as tremors, rigidity or hypokinesia [12], a series of adverse effects can occur, especially in long term treatments. These may include cardiac malfunctions, dyskinesia, neurological misbalances or the wearing off effect [13,14], which represents a main focus point in the research field at the present moment. Previous studies have brought to light an important correlation between the L-Dopa serum concentration fluctuations and the kinetic characteristics of the active substance. It has been shown that after a certain number of years of treatment, the concentration of L-Dopa fails to remain at the expected active level and, because of this, the drug effect cannot be predicted for the same amount of time, thus the administration rhythm has to be modified in order to keep under control the Parkinson's symptoms [13,15]. A comprehensive profile of the kinetic particularities of L-Dopa is therefore of the uttermost importance.

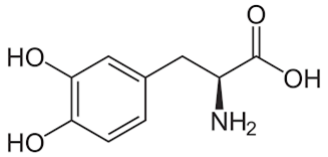


Fig. 1. Structure of L-Dopa

In this study, we investigated the solid-state stability of L-Dopa in both inert and oxidative atmosphere, by subjecting the compound to thermal stress under dynamic non-isothermal conditions [16-25].

## Experimental part

### *Matherials and methods*

Levodopa (L-Dopa) was a commercial product of Sigma Aldrich with purity according to United States Pharmacopeia (USP) Reference Standard (CAS Number 59-92-7, Molecular mass 197.19 g/mol, 1361009 USP). The bioactive substance was stored according to supplier indication.

Thermoanalytical data (TG/DTG/HF) were simultane ously collected on Perkin-Elmer DIAMOND device, the

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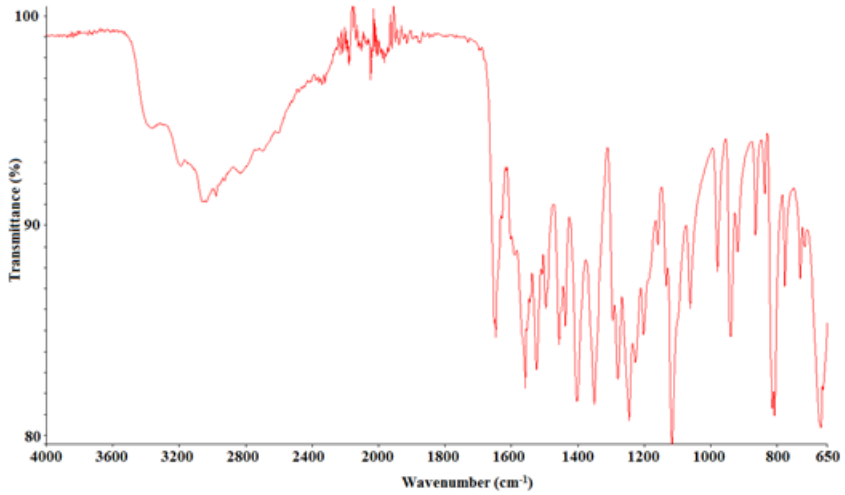


Fig.2. UATR-FTIR spectrum of L-Dopa recorded on the spectral domain 4000-650 $cm^{-1}$

samples were thermally treated (heating rate $\beta$ =5°C·$min^{-1}$) in open aluminum crucibles in air and nitrogen atmosphere (dynamic flow of 100 mL·$min^{-1}$) in the temperature interval of 35-500 °C. The associated thermal effects were recorded as HF (Heat Flow) data (in mW).

FTIR spectra were built up after a number of 32 co-added scans, with a resolution of 4 $cm^{-1}$ on a Perkin Elmer SPECTRUM 100 device. The data was collected directly on solid samples in the spectral domain 4000-650 $cm^{-1}$ on an UATR device.

## Results and discussions

*Spectroscopic analysis*

UATR-FTIR spectrum of pure L-Dopa (fig. 2) show several well-defined, sharp absorption bands in good agreement with the diverse functional groups present in the structure, namely the amino-acid moiety and the hydroxylated benzene ring.

In the 3600-2400 $cm^{-1}$ spectral range, a broad band is observed due to the presence of OH groups in the structure. As a superimposition of bands, some peaks appear at 3357, 3192, and 3038 $cm^{-1}$ due to the vibration of simple bonds with hydrogen of N, O and C. The C=O bond appear as intense band at 1650 $cm^{-1}$, while the bending of O-H is represented by the band at 1404 $cm^{-1}$. The O-H stretching is visible at 1248 $cm^{-1}$. The N-H bending is represented by the band at 1560 $cm^{-1}$, while C-N stretching is situated in the 1200-1064 $cm^{-1}$ spectral range. All the obtained data, except the C=O band is in good agreement with already published data [26]. This instrumental technique was solely used in order to prove the identity and purity of L-Dopa.

*Thermal study*

A comparative thermal stability study was carried out for degradation of L-Dopa in both oxidative (air) and inert (nitrogen) atmospheres. The TG/DTG/HF curves obtained at $\beta$= 5 °C $min^{-1}$ over the temperature range of 35-500 °C are presented in figures 3 and 4.

The thermal stress carried in nitrogen atmosphere (fig.3) reveals a heat flow curve that show only one sharp endothermic peak with HF maximum at 288 °C ($HF_{onset}$ = 275.8 °C), which corresponds to a mass loss process of ≈ 58 %. The melting interval for L-Dopa was reported to be in 284-286 °C range of temperature in the literature [27]. The thermogravimetrical curve (TG) detects a constant mass from initial temperature value until 250.3 °C. The region of the TG curve from 250 to 500 °C is characterized by a continuous mass loss, process which is accompanied by a strong endothermic effect due to the melting of the active substance. The slope of the TG curve varies, the

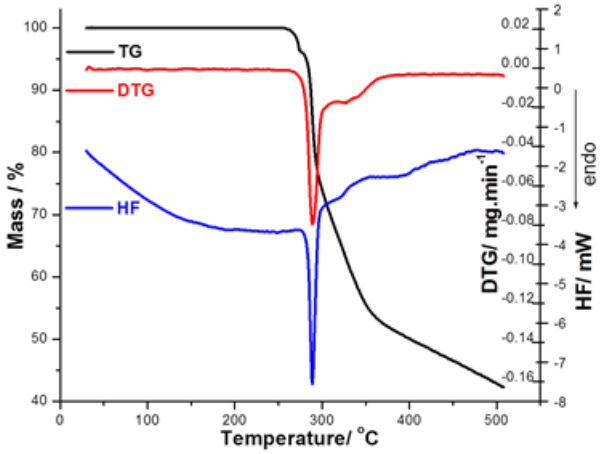


Fig.3. Thermoanalytical curves determined for L-Dopa in nitrogen at = 5 °C·$min^{-1}$

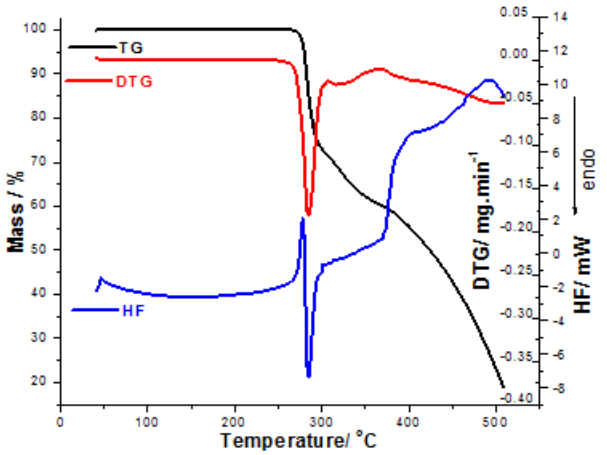


Fig.4. Thermoanalytical curves determined for L-Dopa in air at $\beta$ = 5 °C·$min^{-1}$

inflexion points observed on the TG curve indicating a complex mechanism of the occurred processes of decomposition. The main decomposition process being identified with respect to the DTG curve in 262-314 °C temperature range ($DTG_{peak}$= 289.6 °C) corresponds to a mass loss Δm=30.8%. However, the aspect of heat flow curve indicates a clearly endothermic process of molecular skeleton breakdown, but without occurrence of thermooxidation which was however expected due to the inert origin of the surrounding gas.

A similar behaviour was observed under subjection of L-Dopa to thermal stress in oxidative atmosphere (fig. 4). A similar thermal stability in terms of mass constancy was observed, L-Dopa being stable up to 263 °C, a temperature surprisingly superior than the one determined in nitrogen

atmosphere. This stability is indicated by the all three thermal curves (TG, DTG and HF). Above this temperature, a continuous mass loss is observed, the final mass of residue being ≈ 18.9%. The mass loss percent determined according to the TG curve recorded in air is different comparative with the Äm obtained from thermo-gravimetrical data obtained in $N_2$ atmosphere. The difference can be explained by the thermolysis by a different mechanism due to the nature of atmosphere, which is also sustained by the HF curve, that show two well defined events. The first observed peak has an exothermic nature due to the thermooxidation process which occurred between 263 and 280 °C. The second peak on HF curve appeared in the continuation of the first process and characterized the melting of L-Dopa. The maximum of HF peak is situated at 283.5 °C and is in accord with the literature [27]. The DTG curve is similar with the DTG curve presented before, $DTG_{peak}$ being identified at 283.5 °C. This comparative stability will be further investigated in future studies, regarding both compatibility of L-Dopa with excipients in solid state, but as well by kinetic means, for a pertinent estimation of the kinetic triplet.

## Conclusions

This study represents a preliminary investigation regarding the stability of antiparkinsonian drug L-Dopa in oxidative *vs.* inert atmosphere. A similar behavior was observed, L-Dopa being stable up to 250°C in inert atmosphere and up to 263 °C in oxidative atmosphere. This increased stability can be explained by the presence of intra- and intermolecular H-bondings due to the grafted functional moieties, namely OH, COOH and $NH_2$ groups, but as well for the possibility of formation of zwitterion due to the protonation of the α-amino group by the carboxyl moiety in solid state. However, a complete stability study can be realized by employment of kinetic studies regarding the mass loss in the decomposition temperature range.

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