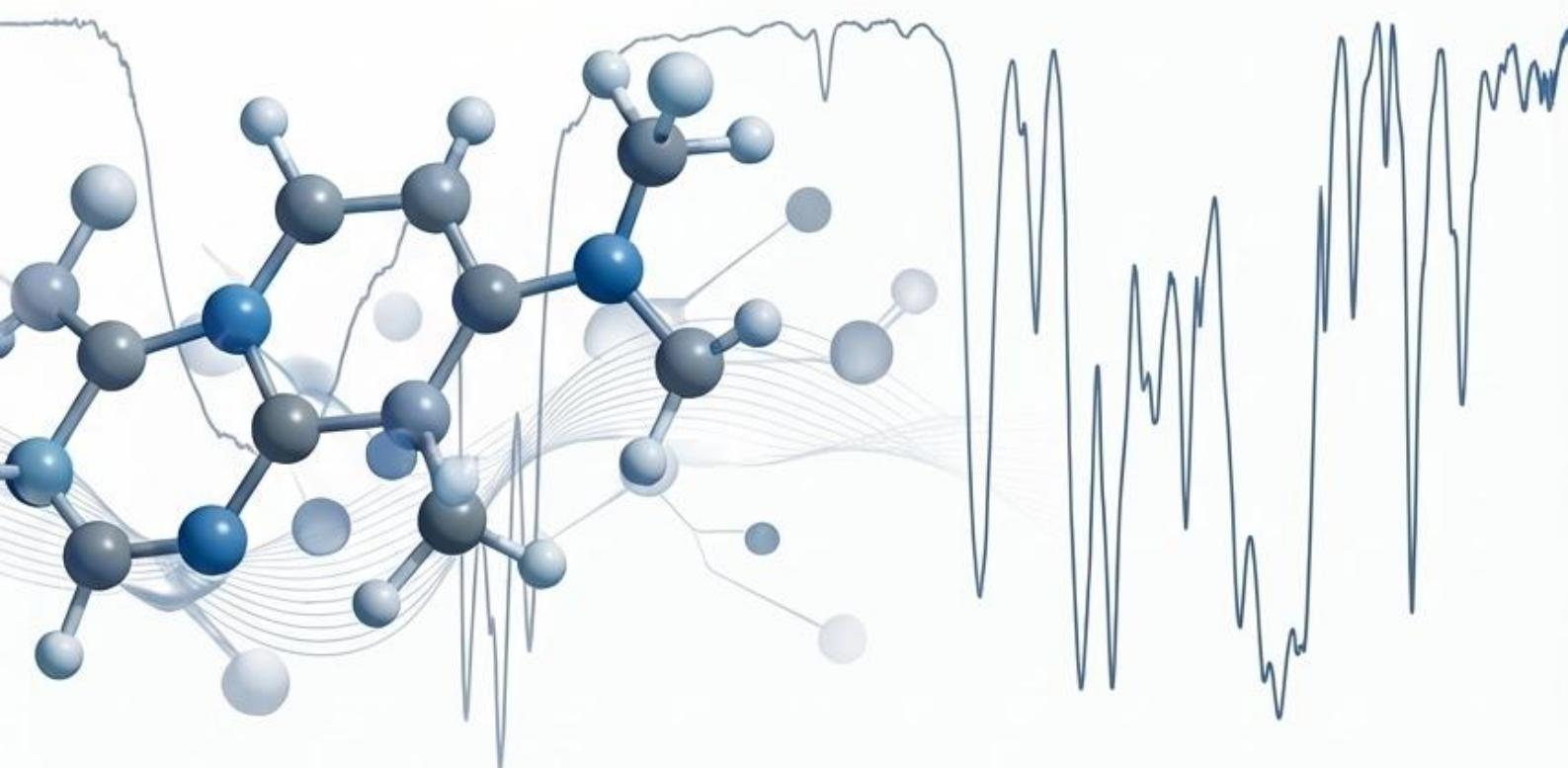


FTIR UHMWPE Analyzer – Software User Manual

Professional Oxidation, Crystallinity and
TVI Analysis at Multiple Depths



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Developed by the Biomaterials Group, Aragón Institute of
Engineering Research (I3A), University of Zaragoza

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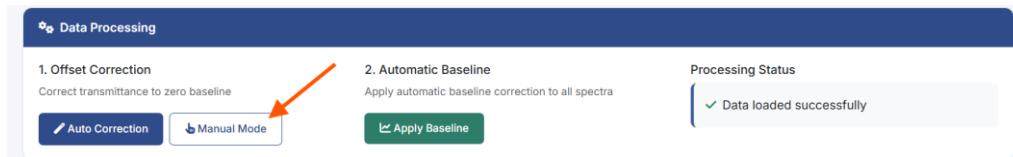
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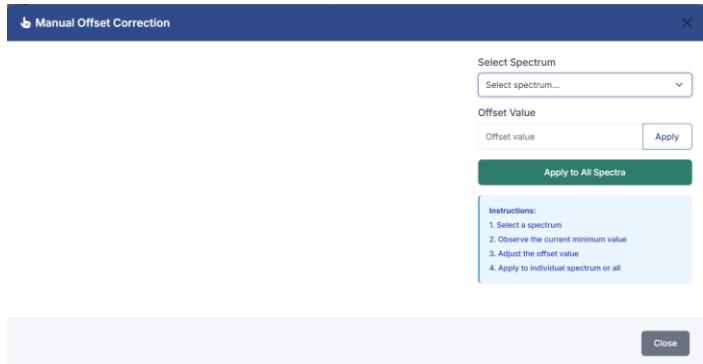
1.General Software Functions

Note all plots in this app are generated by *plot.ly library*. This type of plot is highly interactive, and possible interactions are the following ones:

- a. **Zoom Interactions:** Users can zoom-in by clicking and dragging on the plot area when the drag mode is set to 'Zoom'. Double-clicking on the plot performs an autoscale function, resetting the zoom to view the entire dataset. Zoom can also be performed along a single axis by clicking and dragging near the edges of either the x-axis or y-axis, or along both axes simultaneously by dragging near the corners.
- b. **Axis Movement:** Users can move along both axis by clicking and dragging within the plot grid.
- c. **Hover Information:** Hovering over data points displays interactive tooltips with detailed information about the selected point, including coordinates and other relevant attributes.
- d. **Toolbar Controls (Mode Bar):** A floating toolbar appears in the top-right corner when hovering over the plot, containing several interactive buttons including: zoom-in, zoom-out, autoscale/reset axes, download the chart as PNG image, and toggle visibility of individual traces through the legend. Users can single-click legend entries to hide/show traces, or double-click to isolate a single trace while hiding all others.
- e. **Axis-Specific Controls:** Double-clicking on a single axis autoscales only that specific axis while maintaining the zoom level of the other axis. It is possible to zoom-in focusing on a specific axis by click and dragging horizontally (y-axis zoom-in) or vertically (x-axis zoom-in).

Please note that this manual user guide aims to explain the software's automatic pipeline. However, each function presents a manual pipeline option, allowing users to manually complete the analysis. This pipeline is provided as an alternative in case bugs emerge during automatic analysis (e.g.: Manual Offset correction).

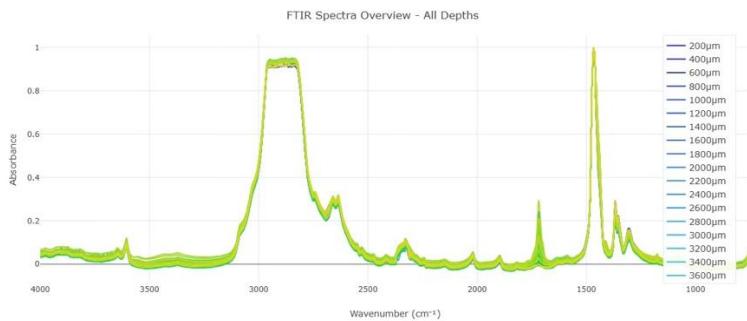




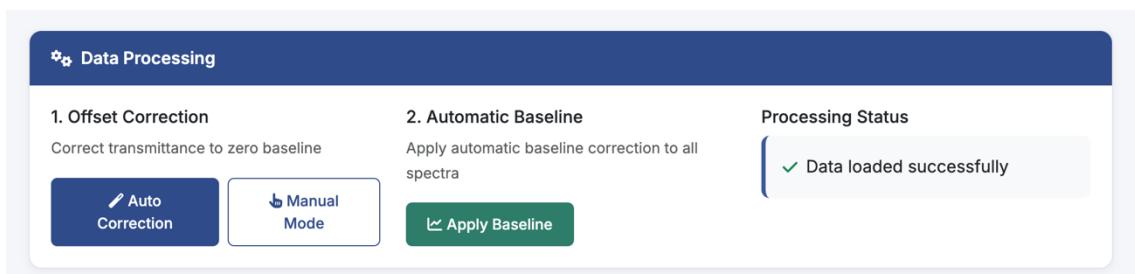
The first step to use the software is to upload your FTIR data file in a compatible format (such as .xlsx, .dpt, .spc, .txt, .csv, or .0) using the “*Choose File*” button. Note it is possible to choose between *single (multi-depth)* or *multiple depth files*.



Once the file has been uploaded, this overview of the FTIR spectra is displayed in *FTIR Spectra Overview*, where you can select the specific depth you want to show from clicking on the legend at the right (one-click for select or deselect, two-clicks for showing just the depth selected and restore the spectra showing all depths).



At this stage, you can apply offset correction and automatic baseline correction to the loaded spectra to ensure that all transmittance values are properly adjusted before further analysis. Automatically, *FTIR Spectra Overview* is updated.



The analysis module of the software is divided into three main sections: *Index Analysis*, *Carbonyl Deconvolution*, and *Absorbed Species Quantification*, which can be selected and used independently depending on the type of evaluation you want to perform.

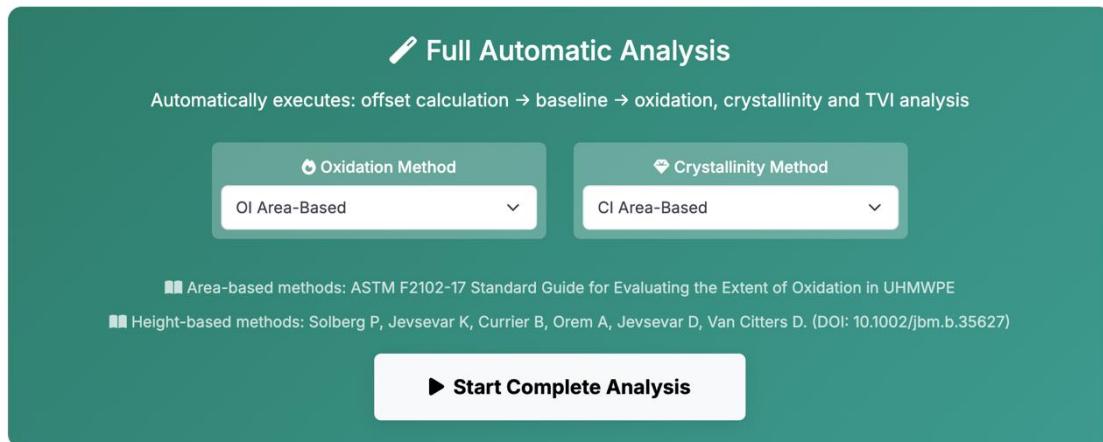


2. Index Analysis

First, the Index Analysis module is divided into *Oxidation Analysis*, *Crystallinity Analysis*, and *TVI Analysis*, which can be accessed individually, while the *Reports* section provides a comparative summary of the results obtained in these analyses.



A complete and immediate Index Analysis result can be obtained from this section by choosing the desired oxidation and crystallinity methods (*Area-based* or *Height-based*) and then clicking on “*Start Complete Analysis*”.



2.1 Oxidation Analysis

To calculate the oxidation analysis, user can define the wavenumber ranges for the interest peak and the reference peak (by default, it is defined with ASTM F1202 values in Area-based method, and reference values in Height-based method), then click *Analyze All Automatically* to compute the oxidation indices and optionally enter *Length in vivo (years)* value so that results are normalized per year.

Interest Peak (cm^{-1})

1680 1850

Min Max

Reference Peak (cm^{-1})

1330 1400

Min Max

Length In Vivo (years)

Enter years in vivo

Time the prosthesis was implanted

Note: If no value is entered, indices are shown as ratios. When a value is provided, results are normalized by the implant duration (index/year).

Analyze All Automatically

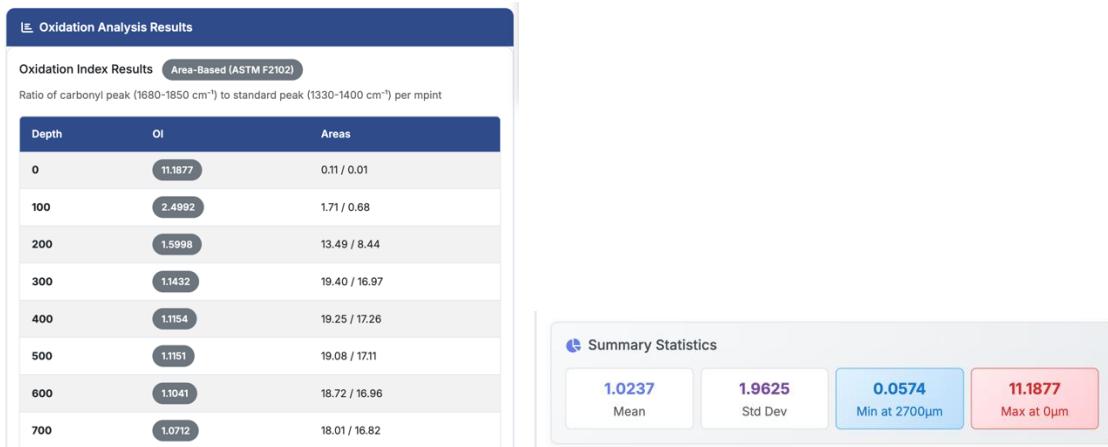
Manual Baseline

Reset

As a result of this calculation, the software generates *baseline-corrected oxidation index spectra* for both the reference and interest intervals at all selected depths, which can be visualized and filtered using the *depth selection* panel.



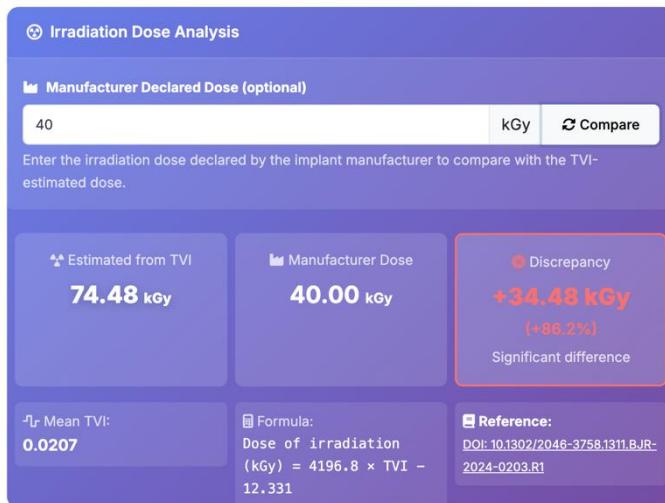
A table is also generated containing the oxidation index and corresponding peak areas for each depth, accompanied by *summary statistics* such as mean, standard deviation, and the minimum and maximum OI values.



2.2 Crystallinity and TVI Analysis

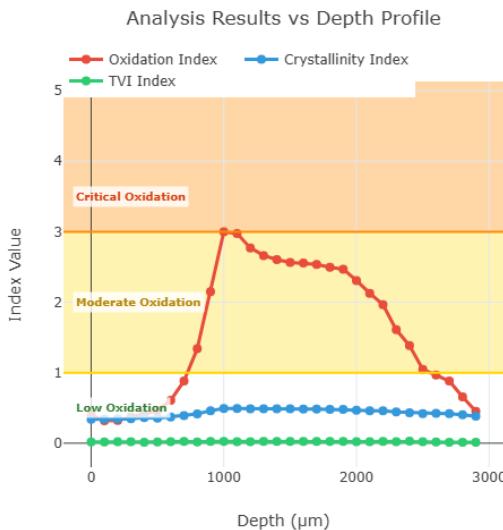
The same workflow described above also applies to *Crystallinity Analysis* and *TVI Analysis*, where the corresponding spectral intervals are defined, the automatic calculations are executed, and the results are presented in equivalent plots and summary tables.

In addition, within the *TVI Analysis* module the software can estimate the irradiation dose received by the UHMWPE and optionally, compare this TVI-derived dose with the irradiation dose specified by the implant manufacturer.



2.3 Reports

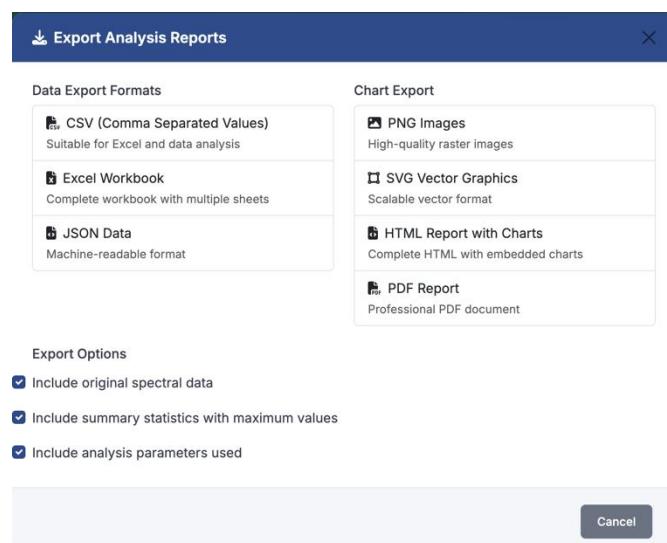
Finally, the *Reports* section provides an overall summary of all *Index Analysis* calculations, combining the depth profiles and detailed tables for the oxidation, crystallinity, and TVI indices into a single integrated overview.



Detailed Results Table
Sample file: H007_UnWorn.0

Depth	OI (ASTM F2102)	CI (Area-Based)	TVI Index
0.0 μm	11.1877	0.9078	0.2135
100.0 μm	2.4992	0.5939	0.0341
200.0 μm	1.5998	0.4534	0.0208
300.0 μm	1.1432	0.3789	0.0141
400.0 μm	1.1154	0.3778	0.0139
500.0 μm	1.1151	0.3806	0.0139
600.0 μm	1.1041	0.3807	0.0139
700.0 μm	1.0712	0.3789	0.0137

From the *Export Data* Analysis bottom, the user can download the results in multiple formats (CSV, Excel, JSON, PNG, SVG, HTML, or PDF) and optionally include the original spectra, summary statistics, and all analysis parameters used, making it easy to archive, share, or further process the reports outside the software.



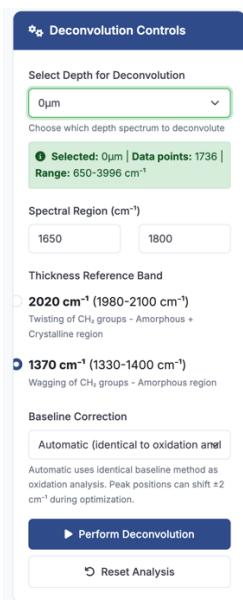
3. Carbonyl Deconvolution

The software enables the differentiation and quantification of carbonyl species in oxidized UHMWPE through a sophisticated algorithmic approach. The process involves two interconnected components: first, an Adam deconvolution algorithm that resolves individual oxidation species peaks from carbonylic interval spectra, and second, a Lambert-Beer algorithm that quantifies each identified species. The software supports both *individual depth deconvolution* and simultaneous *multi-depth deconvolution*.



3.1 Individual Deconvolution

Within this *Individual Deconvolution* section, the user can select the depth to be analyzed, define the spectral region and reference band (by default, *spectral region* is based on ASTM F1202 oxidation peak $1650 - 1800 \text{ cm}^{-1}$, and *reference peak* based on 1370 cm^{-1}), and then perform carbonyl deconvolution using an automatic baseline correction and Gaussian–Lorentzian deconvolution (Further deconvolution algorithm and workflow information is available at Annex. 1).



In addition, a *methodology info* panel is available in the lower right margin, providing detailed information on the depth selection, deconvolution steps, and table-based identification of the different carbonyl species involved in the procedure.

Methodology Info

Depth Selection:

- Select specific depth from dropdown (100µm, 200µm, etc.)
- Each depth represents material at different penetration levels
- Surface layers (100µm) typically show higher oxidation
- Compare different depths by changing selection and re-running analysis

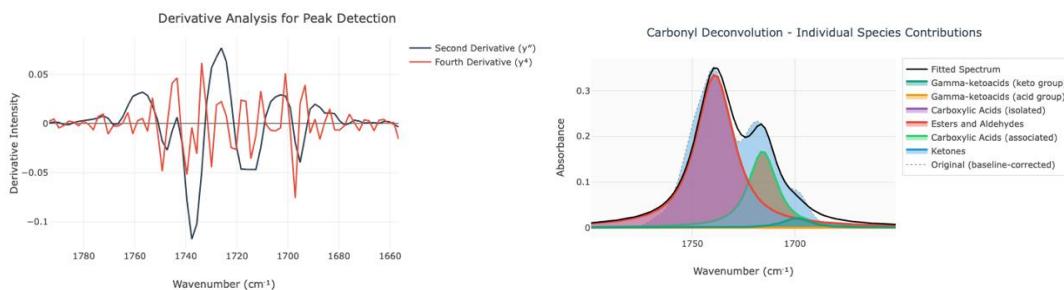
Deconvolution Process:

1. Spectral normalization using selected C-H reference band
2. Baseline correction (linear/polynomial)
3. Derivative analysis (2nd & 4th order) for peak detection
4. Gaussian-Lorentzian curve fitting for 6 carbonyl species
5. Quality assessment ($R^2 > 0.90$ target)

Species Identification (Table-based):

- Gamma-ketoacids keto (1698 cm^{-1}) - Oxidation product
- Gamma-ketoacids acid (1707 cm^{-1}) - Oxidation product
- Carboxylic Acids associated

These plots show the results of *individual deconvolution*, including the contributions of each single carbonyl species to the fitted spectrum, the second and fourth derivative-based peak detection (*Derivative analysis* is provided for informative purposes only and does not impact the deconvolution algorithm.), and the corresponding fit quality metrics for the selected depth.



Fit Quality

R^2 :	0.9974
R^2 - Absorbance:	0.8961
Area Error:	1.38e-1

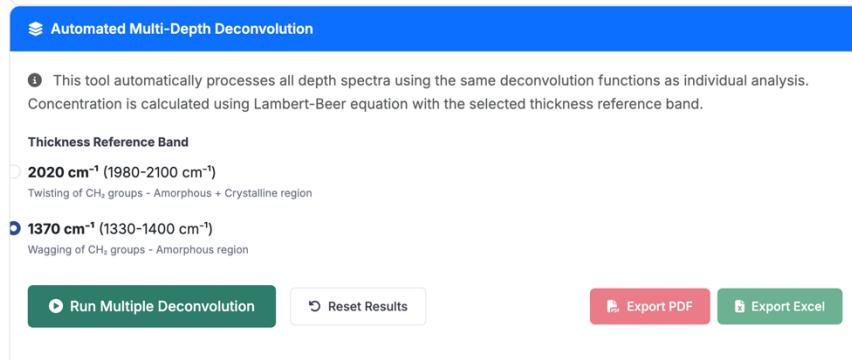
With the *Individual Deconvolution* analysis, the software provides these detailed outputs, including the individual carbonyl species contributions, fit-quality metrics, and a quantitative table summarizing peak parameters and concentrations (mmol/cm^3).

Species Quantification						
Integration Method:		Total Area:			Reference Area:	
	Simpson's Rule	0.055			0.0096	
Parameter	Gamma-ketoacids (keto group)	Gamma-ketoacids (acid group)	Carboxylic Acids (associated)	Ketones	Esters and Aldehydes	Carboxylic Acids (isolated)
Frequency (cm ⁻¹)	1699.1	1708.2	1715.2	1722.3	1738.3	1768.1
Percentage	12.2%	23.8%	0.4%	16.4%	25.0%	22.4%
Simpson Area	0.0068	0.0130	0.0002	0.0090	0.0138	0.0123
Intensity (I)	0.000	0.001	0.000	0.000	0.000	0.000
Width (W)	16.2	17.2	18.2	13.2	20.2	22.7
Lorentzian (%)	93.0%	94.0%	95.0%	93.0%	90.0%	93.0%
Conc. (mmol/cm ³)	0.1017	0.1963	0.0013	0.1359	0.1761	0.0761
ϵ (L·mol ⁻¹ ·cm ⁻¹)	6880	6880	16800	6880	8110	16800

Carbonyl Species		
● Ketones	1721 cm ⁻¹	
Width: 11-13 cm ⁻¹ Lorentzian: 90-93%		
● Carboxylic Acids	1714 cm ⁻¹	
Width: 16-18 cm ⁻¹ Lorentzian: 85-95%		
● Gamma-ketoacids (acid)	1707 cm ⁻¹	
Width: 16 cm ⁻¹ Lorentzian: 90-94%		
● Gamma-ketoacids (keto)	1698 cm ⁻¹	
Width: 15 cm ⁻¹ Lorentzian: 90%		
● Esters & Aldehydes	1737 cm ⁻¹	
Width: 19 cm ⁻¹ Lorentzian: 85-90%		
● Carboxylic Acids (isolated)	1767 cm ⁻¹	
Width: 20-23 cm ⁻¹ Lorentzian: 90%		

3.2 Multiple Deconvolution

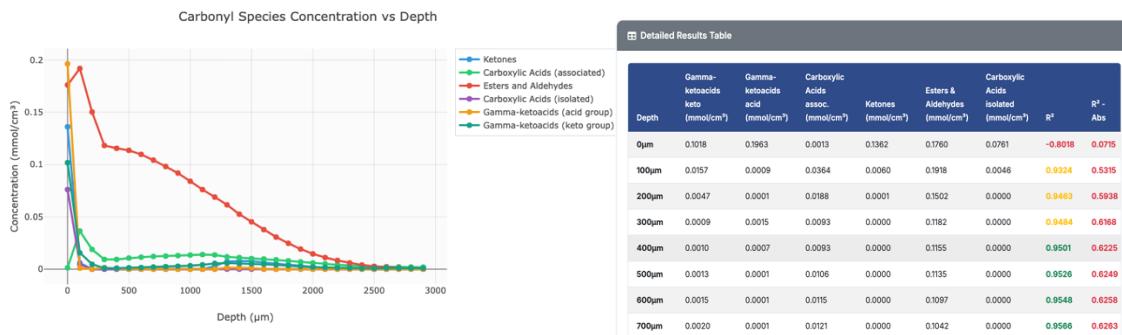
In the *Multiple Deconvolution* panel, the software performs the same deconvolution procedure across all available depths in a single step, using the selected thickness reference band to compute concentrations via deconvolution algorithm + Lambert–Beer equation and providing export options for the full multi-depth dataset.



The software visually tracks the iterative progress of *Multiple Deconvolution*, indicating for each step which depth is currently being processed and how many depths have been completed out of the total.

Processing 1800µm (19/30)...

The automated *Multiple Deconvolution* produces a concentration versus depth profile for each carbonyl species, together with a detailed results table that lists the calculated concentrations and fit-quality statistics at every analyzed depth.



4. Absorbed Species Quantification

This software enables the quantification of species absorbed from synovial fluid and vitamin E on the UHMWPE. It also supports comparative analysis of UHMWPE before and after synovial fluid extraction, helping to separate intrinsic oxidation species from changes driven by absorbed joint fluid species.

⌚ Absorbed Species Quantification

Quantification of vitamin E and synovial liquid absorbed on UHMWPE surface

[⌚ Vitamin E Quantification](#)
[⌚ Single Synovial Liquid Quantification](#)
[⌚ Comparative Synovial Liquid Analysis](#)

4.1 Vitamin E Quantification

To calculate vitamin E index, the user may define the wavenumber range for the vitamin E interest peak (by default 1210 cm^{-1}) and the reference peak (by default 1370 cm^{-1}), then runs the automatic analysis to compute the corresponding vitamin E index or concentration at all selected depths.

Analysis Parameters

Interest Peak - Vitamin E (cm^{-1})

1200	1220
Min	Max

Reference Peak (cm^{-1})

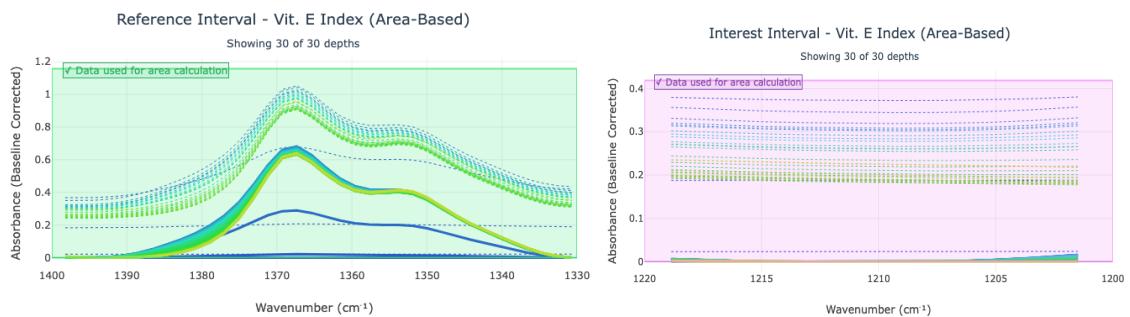
1330	1400
Min	Max

Analyze All Automatically

Manual Baseline

Reset

As a result of this calculation, the software generates baseline-corrected vitamin E index spectra for both the interest and reference intervals at all selected depths, which can be visualized and filtered using the depth selection panel.

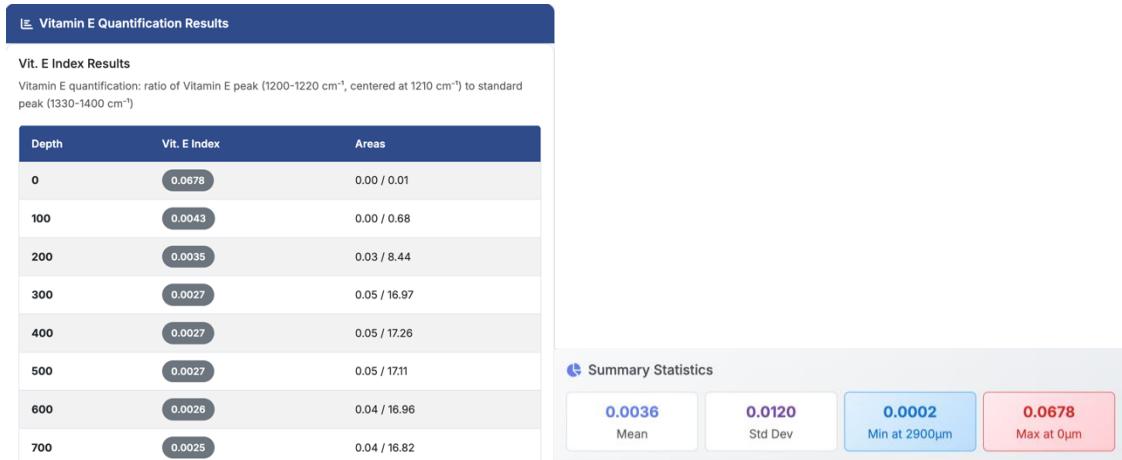


Depth Selection

All None Every 5th

- 1500 μm
- 1600 μm
- 1700 μm
- 1800 μm
- 1900 μm
- 2000 μm

A table is also generated containing the *Vit. E index* and corresponding peak areas for each depth, accompanied by *Summary Statistics* such as mean, standard deviation, and the minimum and maximum vitamin E values.



4.2 Single Synovial Liquid Quantification

In the *Single Synovial Liquid Quantification* mode, the user may define the *Lactone Band* and *C–O–C (Esters + Lactones) stretch bands*, selects a reference band, and then runs the analysis to quantify synovial fluid-derived species at each depth.

- Firstly, lactone contribution is defined by identifying their characteristic peaks in the carbonyl region ($1782\text{ -- }1792\text{ cm}^{-1}$).
- Secondly, the C–O–C band ($1168\text{--}1186\text{ cm}^{-1}$) shared by both esters and lactones is analyzed, using the already-known lactone contribution to mathematically separate the contribution of pure esters.
- Lastly, through *Multiple Deconvolution* of the carbonyl region, the combined peak of Aldehydes + Esters is obtained, and by subtracting the ester contribution calculated in the previous step, the final aldehyde contribution is isolated.

The module uses predefined extinction coefficients, based on literature references, to convert the FTIR band areas into concentrations, and it can optionally normalize these values by the implant's time in vivo to obtain depth-resolved concentrations per year of implantation.

Analysis Parameters

- Lactone Band (cm^{-1})**: 1782 – 1792
- C–O–C Stretch Band (cm^{-1})**: 1168 – 1186
- Reference Band (cm^{-1})**: 1330 – 1400
- Extinction Coefficients**:

ϵ_{1782} :	$4210\text{ cm}\cdot\text{mmol}^{-1}$
ϵ_{1792} :	$2420\text{ cm}\cdot\text{mmol}^{-1}$
ϵ_{1168} :	$6300\text{ cm}\cdot\text{mmol}^{-1}$
ϵ_{1186} :	$5420\text{ cm}\cdot\text{mmol}^{-1}$

Multi-depth Analysis: This analysis automatically processes all loaded depths using carbonyl deconvolution and multi-component quantification.

Length In Vivo (years): Enter years in vivo
Time the prosthesis was implanted

Note: If no value is entered, concentrations are shown in mmol/cm^3 . When a value is provided, results are normalized by the implant duration ($\text{mmol}/(\text{cm}^3\cdot\text{year})$).

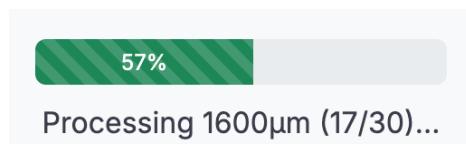
Run Synovial Liquid Analysis

Reset Analysis

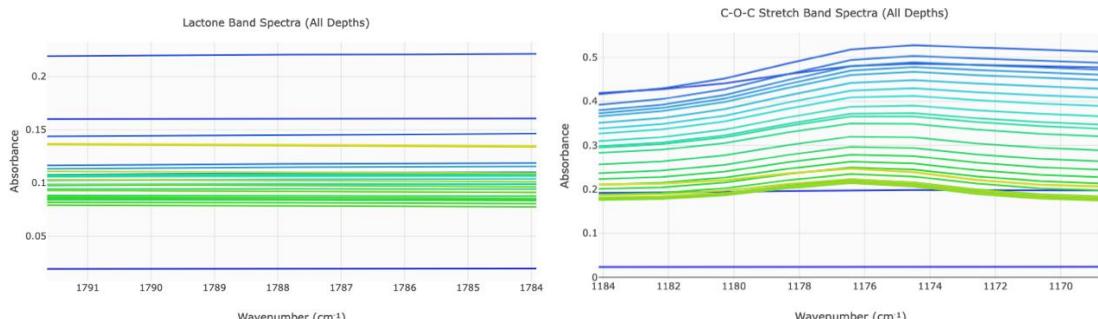
At the bottom of this panel, you can see *Methodology: Multi-component Analysis*, a summary of how the multi-component synovial liquid analysis is performed. It clearly lists the calculation steps for lactones, esters, aldehydes, and carboxylic acids, together with the extinction coefficients used to convert each FTIR band into concentration values.

Methodology: Multi-component Analysis	
Analysis Steps:	Extinction Coefficients (ϵ):
1. Lactones: Directly from 1788 cm^{-1} band (solitary contribution)	ϵ_{1788} (Lactones) = $4210\text{ cm}\cdot\text{mmol}^{-1}$
2. Esters: From C–O–C band (1170 cm^{-1}) minus lactone contribution	ϵ_{1172} (Lactones C–O–C) = $2420\text{ cm}\cdot\text{mmol}^{-1}$
3. Aldehydes: From deconvolution peak (1737 cm^{-1}) minus ester contribution	ϵ_{1168} (Esters C–O–C) = $6300\text{ cm}\cdot\text{mmol}^{-1}$
4. Carboxylic Acids: Directly from carbonyl deconvolution	ϵ_{1733} (Aldehydes) = $5420\text{ cm}\cdot\text{mmol}^{-1}$
	$\epsilon_{\text{Carboxylic}} = 16800\text{ cm}\cdot\text{mmol}^{-1}$
Reference: Multi-component analysis of low-density polyethylene oxidative degradation methodology.	

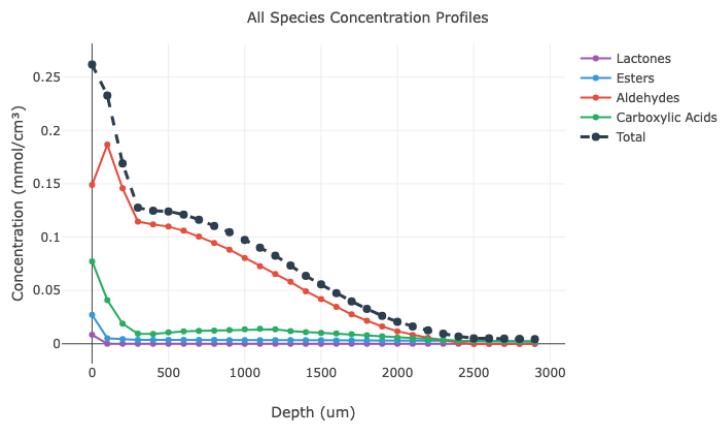
Once *Run Synovial Liquid Analysis* is clicked, a progress bar appears indicating the percentage completed and the current depth being processed, showing how the multi-depth quantification advances through all loaded spectra.



The plots show the baseline-corrected spectra of the selected synovial-liquid bands (lactone band and C–O–C stretch band) for all analyzed depths, allowing visual inspection of how these absorbance signals variated through the UHMWPE thickness.



The software is also capable of generating these multi-component concentration profiles, showing how lactones, esters, aldehydes, carboxylic acids, and their total synovial-derived load vary with depth in the UHMWPE.



The software generates this results table, reporting for each analyzed depth the calculated concentrations of lactones, esters, aldehydes, and carboxylic acids, as well as their total synovial-derived load, together with a concise methodology summary describing how each species is obtained.

Species Concentration Results (Multi-component Analysis)					
Depths Analyzed:		Lactone Band:		Reference Band:	
30/30		1782-1792 cm ⁻¹		1330-1400 cm ⁻¹	
Depth (um)	Lactones (mmol/cm ³)	Esters (mmol/cm ³)	Aldehydes (mmol/cm ³)	Carboxylic Acids (mmol/cm ³)	Total (mmol/cm ³)
0μm	0.0084	0.0271	0.1489	0.0774	0.2619
100μm	0.0000	0.0052	0.1867	0.0410	0.2329
200μm	0.0000	0.0045	0.1457	0.0189	0.1691
300μm	0.0000	0.0036	0.1146	0.0093	0.1276
400μm	0.0000	0.0036	0.1119	0.0093	0.1248
500μm	0.0000	0.0037	0.1099	0.0106	0.1241
600μm	0.0000	0.0036	0.1060	0.0115	0.1212
...

Methodology Summary

Lactones: Direct from 1788 cm⁻¹ band ($\epsilon = 4210$)

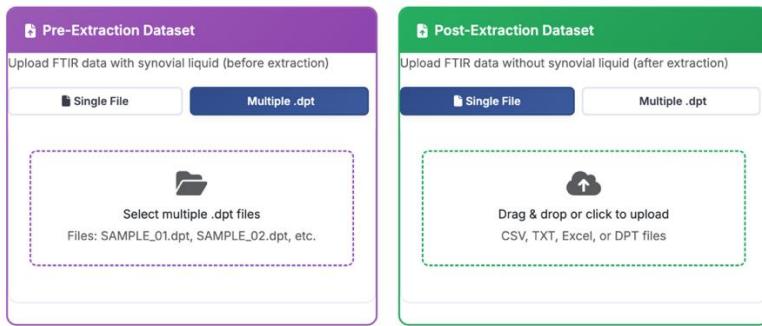
Esters: C-O-C band minus lactone contribution (Eq. 1)

Aldehydes: Deconvolution (1737 cm⁻¹) minus esters

Carboxylic Acids: From deconvolution (1714 + 1767 cm⁻¹)

4.3 Comparative Synovial Liquid Analysis

In the *Comparative Synovial Liquid Analysis* section, the software allows uploading paired FTIR datasets collected before and after synovial fluid extraction, enabling a direct comparison of pre-extraction and post-extraction spectra for the same sample, using the same method as *Single Synovial Liquid Quantification* for both datasets.



In this same section, the user defines the carbonyl and reference bands, specifies the in vivo implantation time if desired, and then runs the comparative analysis, with results available for export as a PDF report or Excel file.

At the end of this section, a comparative analysis methodology panel explains the processing steps for both datasets and how absolute and percentage concentration differences are calculated, highlighting that positive values indicate higher concentrations in the pre-extraction condition.

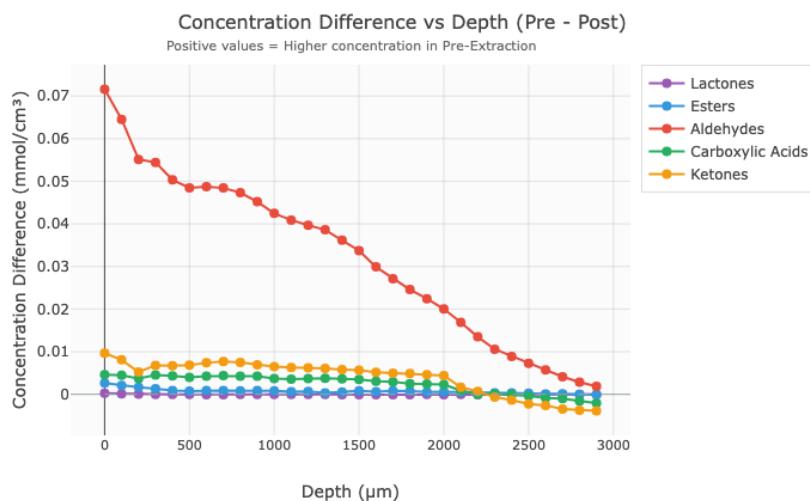
Comparative Analysis Methodology	
Analysis Process:	Difference Calculations:
<ol style="list-style-type: none"> Both datasets are processed using identical synovial liquid analysis pipeline Carbonyl deconvolution is performed for each depth in both datasets Species concentrations are calculated using extinction coefficients Differences are computed for matching depths between datasets 	<ul style="list-style-type: none"> Absolute: $C_{\text{pre}} - C_{\text{post}}$ Percentage: $((C_{\text{pre}} - C_{\text{post}}) / C_{\text{pre}}) \times 100\%$ Positive values indicate higher concentration in Pre-Extraction

During comparative processing, the software displays a progress bar indicating the percentage completed and the specific depth and dataset (pre- or post-extraction) currently being analyzed.

28%

Pre-Extraction: 1600µm (17/60)

The software generates this plot from the concentration data for each chemical species as a function of depth, comparing the pre-extraction and post-extraction concentrations.

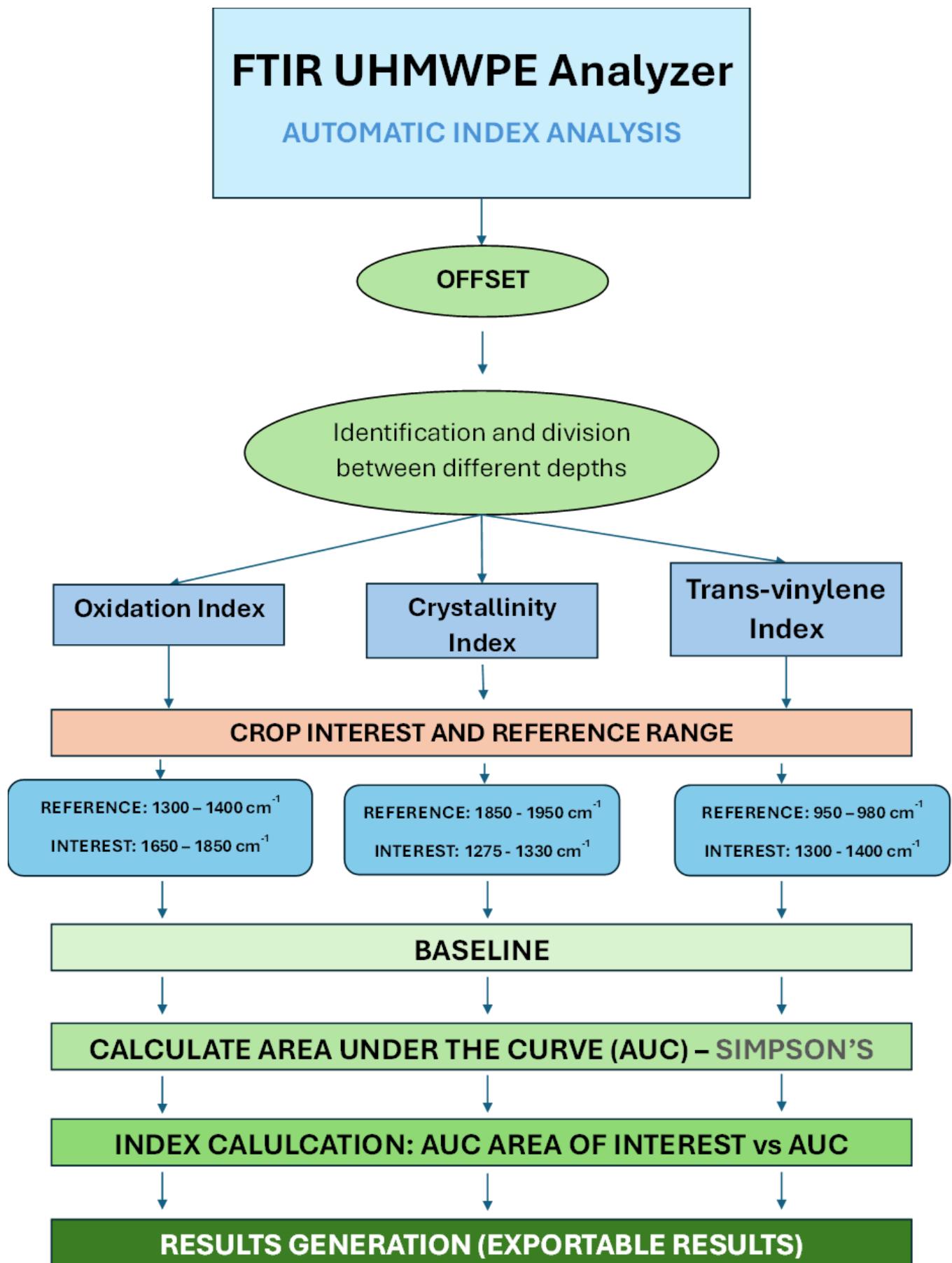


Finally, the software calculates the absolute and percentual concentration differences between the pre-extraction and post-extraction profiles at each depth for every chemical family and summarizes them in these tables, quantifying synovial fluid individual species concentration.

Pre-Extraction Concentrations(mmol/cm³)						Post-Extraction Concentrations(mmol/cm³)						
Depth	Lactones	Esters	Aldehydes	Carboxylic Acids	Ketones	Depth	Lactones	Esters	Aldehydes	Carboxylic Acids	Ketones	R²
0µm	0.0004	0.0047	0.0729	0.0055	0.0104	0.7352	0.0001	0.0020	0.0013	0.0009	0.0007	0.2385
100µm	0.0002	0.0043	0.0672	0.0050	0.0097	0.9320	0.0001	0.0021	0.0027	0.0006	0.0016	0.6939
200µm	0.0001	0.0040	0.0591	0.0043	0.0074	0.9847	0.0000	0.0023	0.0040	0.0006	0.0021	0.9316
300µm	0.0000	0.0036	0.0544	0.0045	0.0068	0.9896	0.0001	0.0023	0.0000	0.0000	0.0000	0.0000
400µm	0.0000	0.0032	0.0503	0.0043	0.0067	0.9897	0.0001	0.0023	0.0000	0.0000	0.0000	0.0000
500µm	0.0000	0.0031	0.0485	0.0039	0.0069	0.9895	0.0001	0.0024	0.0000	0.0000	0.0000	0.0000
600µm	0.0000	0.0031	0.0488	0.0043	0.0074	0.9897	0.0001	0.0022	0.0000	0.0000	0.0000	0.0000
700µm	0.0000	0.0031	0.0484	0.0044	0.0077	0.9897	0.0001	0.0023	0.0000	0.0000	0.0000	0.0000

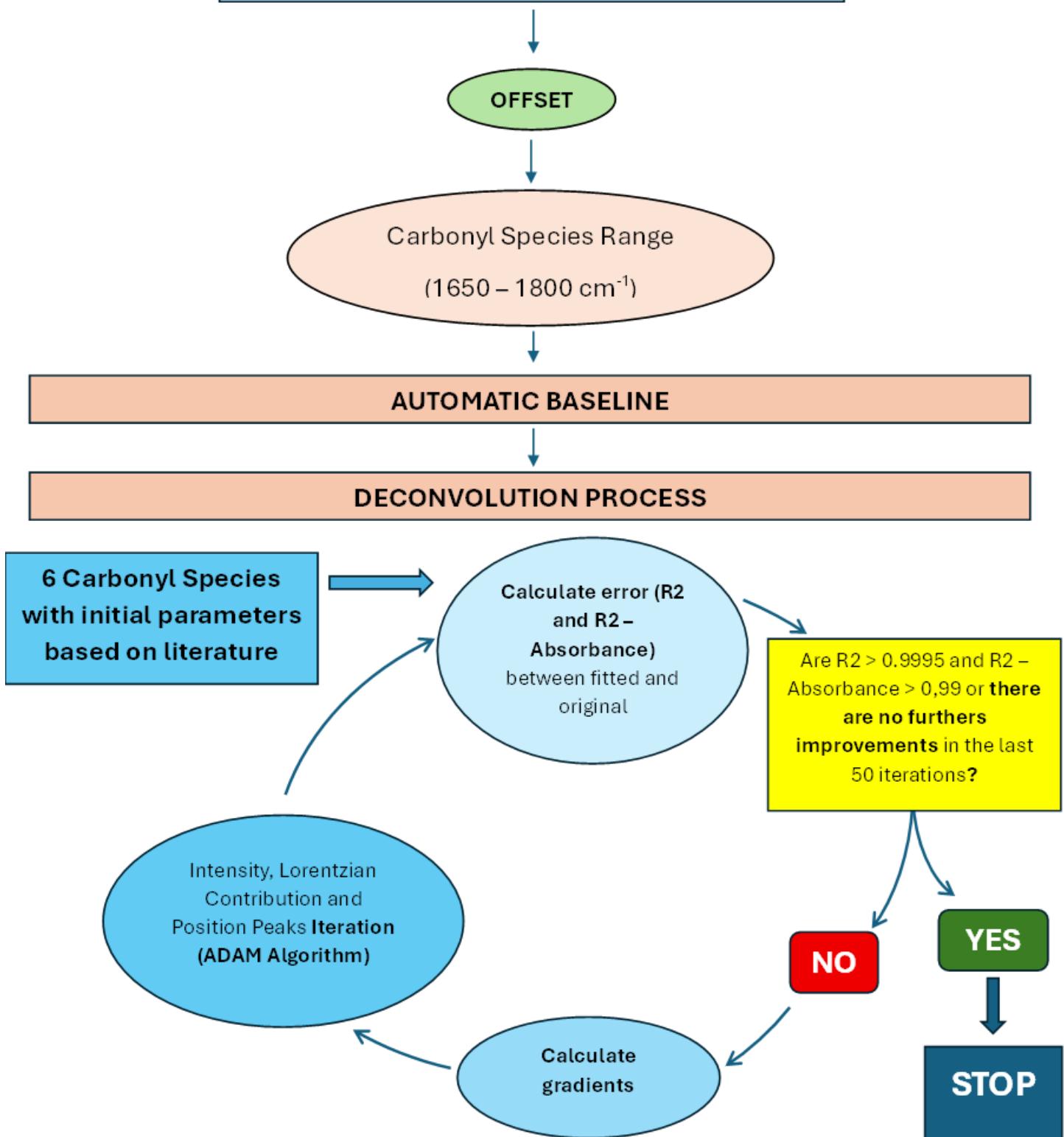
Difference Analysis (Pre - Post)										
Depth	Lactones		Esters		Aldehydes		Carboxylic Acids		Ketones	
	Abs. Diff. (mmol/cm³)	% Diff.								
0µm	0.0003	69.2%	0.0027	57.1%	0.0716	98.2%	0.0047	83.6%	0.0096	92.9%
100µm	0.0001	63.7%	0.0021	49.5%	0.0646	96.0%	0.0045	88.0%	0.0080	83.5%
200µm	0.0000	52.3%	0.0016	41.4%	0.0551	93.3%	0.0037	85.9%	0.0052	71.0%
300µm	-0.0000	-147.4%	0.0013	35.7%	0.0544	100.0%	0.0045	100.0%	0.0068	100.0%
400µm	-0.0000	-193.8%	0.0009	28.3%	0.0503	100.0%	0.0043	100.0%	0.0068	100.0%
500µm	-0.0000	-180.5%	0.0008	24.6%	0.0485	100.0%	0.0039	100.0%	0.0069	100.0%
600µm	-0.0001	-412.4%	0.0009	29.2%	0.0487	100.0%	0.0043	100.0%	0.0074	100.0%
700µm	-0.0001	-450.9%	0.0009	27.6%	0.0484	100.0%	0.0044	100.0%	0.0077	100.0%

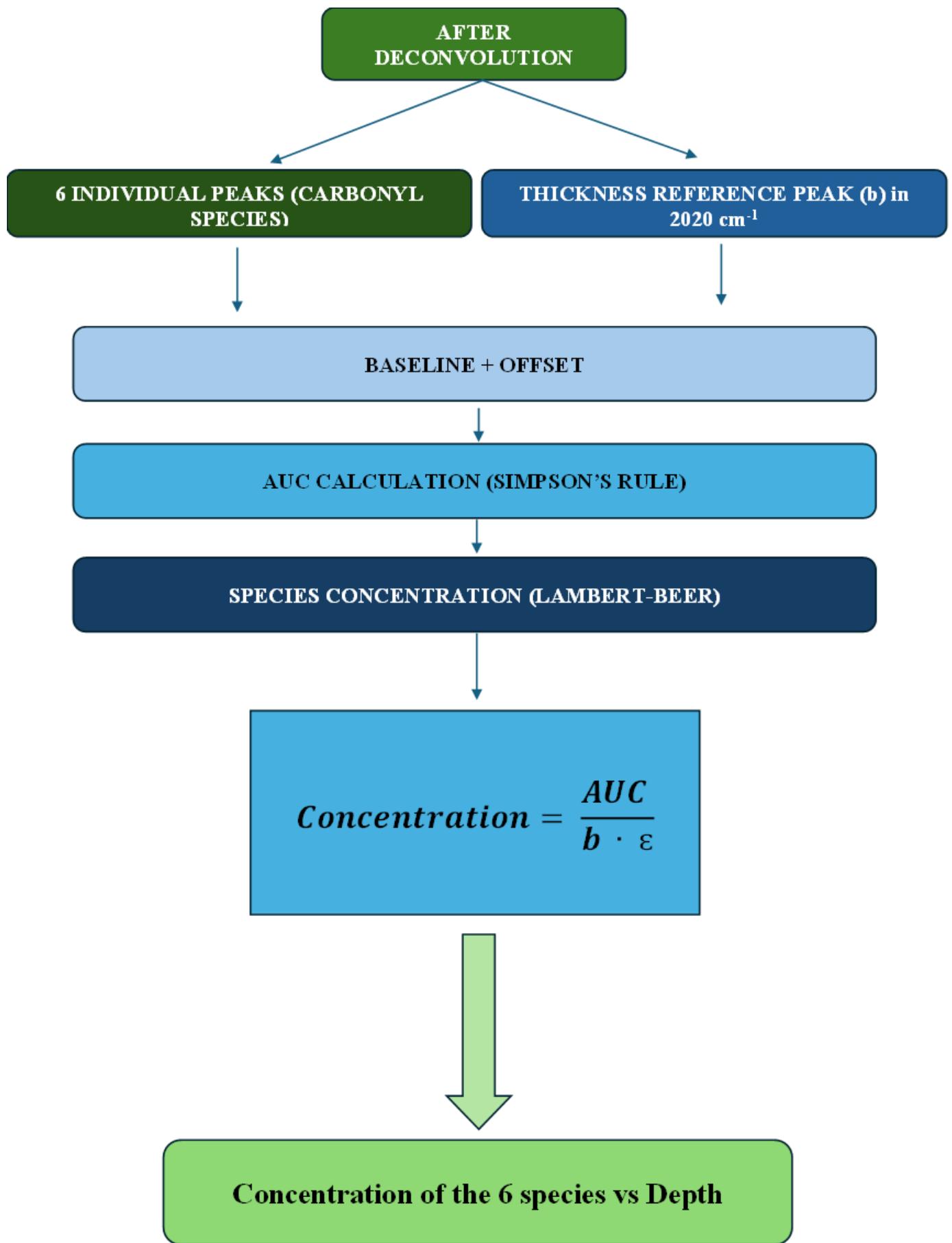
Annex 1. Software Workflow



FTIR UHMWPE Analyzer

AUTOMATIC DECONVOLUTION





Annex 2. Bibliographical references

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