

# USE OF THE FOURIER-TRANSFORM INFRARED SPECTROMETER SPECTRUM 100 FT-IR WITH AN ATR ATTACHMENT.

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## PURPOSE

This SOP describes how to operate and use the Fourier-transform infrared spectrometer Spectrum 100 FT-IR (PerkinElmer) with an ATR (Attenuated Total Reflectance) attachment. Our FTIR instrument is a mid-infrared instrument (optimal wavelength range of 4200-650  $\text{cm}^{-1}$ ). This instrument is located in the CAF facility. For more information on the ATR technique and accessories please see the [APPENDIX](#).

## SCOPE

This SOP describes procedures to be followed by staff and students when using the Spectrum 100 FTIR-ATR instrument in the CAF facility.

## RISKS

DO complete the [CAF Blackboard safety test](#), official training and ONE run supervised by the CAF technician before using the equipment. For future runs you will need to book the instrument.

See the attached [Risk assessment](#) of the instrument.

You must have your [own Risk assessment](#) of the chemical products you are going to analyse.

Please if you want to analyse flammable, toxic, corrosive, explosive or longer term health hazards chemicals talk with the CAF technician before running your samples ([check their MSDS](#)). We may need to take extra safety measures.



## PROCEDURE

### PRECAUTIONS

- DO NOT forget to **clean** the sample stage and ATR foot attachment prior to and after use (ethanol).
- DO NOT leave **sample waste**, glassware or other materials in the lab without properly disposing of them. In some cases, this may require that you bring the waste back to your own lab.
- Please look for technical assistance if you face any instrument **warning/error**.
- **Do not attempt to use** if signage indicates that the instrument is out of use or is currently in use,
- Do keep the instrument dry **avoid spilling liquid** into the instrument. If anything that is spilled enters the ATR area stops and look for the technician. If any liquid enters the main body of the instrument (not the ATR) look for a technician. If the technician is not present, stop the run, close the Spectrum and turn off the instrument (the switch is at back of the instrument, at the right top).

### INSTRUMENT STARTUP

- No other experiments should be running, since you can **only run one** sample at a time. Check that there is not a sample placed on the ATR plate.
- The **instrument and the computer** should **always be on**, if not please ask for technical assistance.
- **Log on** to the irusercaf user, password: Cafatr19. If somebody has signed out you will need to sig in to the PC:

Username: SCPC7046\irusercaf

Password: Cafatr19

Do not log on to Rdg-home with your individual logon as the instrument specific software will not function.

- To **open the instrument software**, double click on the "SPECTRUM" on the desktop. The user is: "iruser" and the password is "irusercaf".

**Troubleshooting:** If the software is taking an **unusually long time to initialize** (longer than 3 minutes), you may need to restart the computer. If this does not work you will need to do the following:

- Close the software and restart the PC.
- Turn the instrument off (the switch is on the back top right hand side)
- Wait for 10 seconds.
- Turn the instrument on again.
- Log back on the computer
- Wait until the instrument finishes the automatic tests (only a couple of minutes). Once it finishes you will see "PerkinElmer" on the small blue screen of the instrument, right hand side.
- Open the Spectrum software and work as normal.

#### SAMPLE STAGE PREPARATION.



*Figure 1. Sample stage in a FTIR-ATR.*

- Before analysing your sample you have to carefully **wipe the sample stage** with ethanol, soak a soft tissue with EtOH. **Attach an ATR foot** attachment (there is a metallic tip behind the pressure arm) to the universal accessory pressure arm (bottom of the arm) and wipe it with ethanol. To attach the tip just push it with one hand while holding the pressure arm with the other (Figure 1).

**CAUTION:**

Currently, for general users there is only metallic tip available as a foot attachment, a flat one. However, a **sharper tip** is sometimes needed to get a better contact with the crystal. If you think that you may need it, ask for it to the CAF technician.

Do **not clean the ATR sample stage directly** with EtOH, use a soaked soft tissue instead. Do not leave the EtOH bottle close to the instrument either. The instrument contains a hot source and contact with flammable vapours may cause an explosion.

- Allow enough **time for evaporation** of the solvent before you load your sample or take a background measurement.

#### SCAN SET-UP (Spectrum software)

- In the Spectrum software you will see the last user's set up. First of all, you need to go to the top right of the software and select **Set up > Instrument** (Figure 2).

**TROUBLESHOOTING:** If you do not see <Sample ID> as a menu option (top part of the software) you will need to change the <Instrument Data Collection path> to C:\pel data\spectra\ Student data\ your supervisor's folder\your own folder and to close the software and log back in (Figure 2).

- Before modifying the method setup options, you have to **check that the right default setup** is in place for the following tabs (Figure 2, method setup): Instrument data collection. Instrument BeamPath, instrument Advanced parameters and Instrument Basic. Please see the APPENDIX for the right default setup.
- Select the **Instrument data collection > Auto-Save options>** tick the save after each measurement where you will save your data: C:\pel data\spectra\ Student data\ your supervisor's folder\your own folder (each user should create his own folder). You are recommended to tick the Export option too (csv, Excel file format) and to select the same folder.

- To enter your experiment parameters, **select the Setup Instrument Basic tab**. You can choose the abscissa and ordinate units, the start and end points, the resolution and accumulations. The recommended units are wavenumber (x) and %T or Abs (y) (identification and quantification analysis, respectively). The instrument starting and ending points are 4200 and 650  $\text{cm}^{-1}$ . Good starting points for Resolution and accumulations are 4  $\text{cm}^{-1}$  and 16 scans respectively, please see the APPENDIX (right default setup) for more information. You can also change the start, end and scans at the top of the screen.
- Enter in **<Sample ID>** the name of your first sample (for your identification) and enter in **<Description>** the information you would like to be identified with your sample. You can also leave the default, which has a sample number, user, and date.

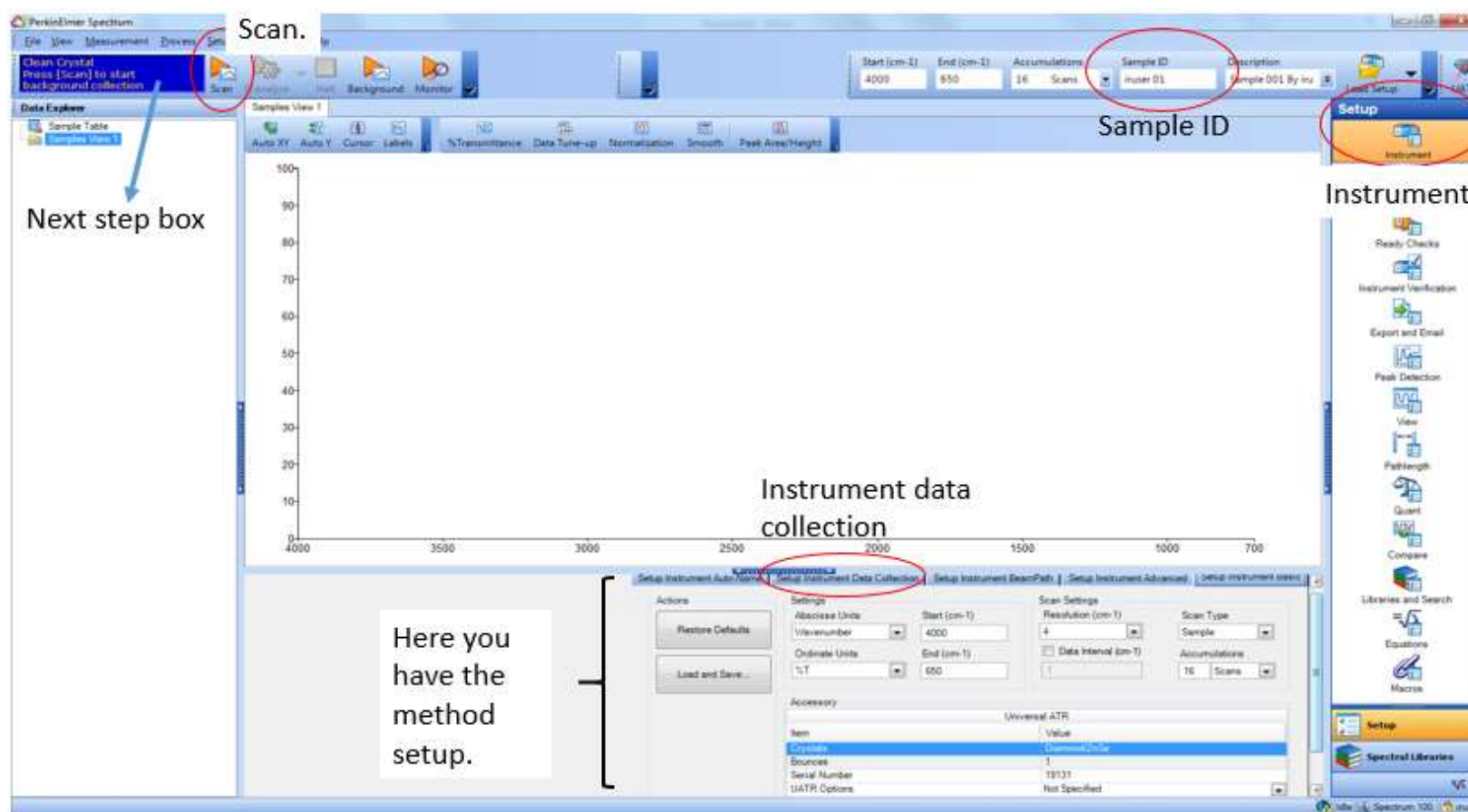


Figure 2. Main view of Spectrum software.

STARTING YOUR EXPERIMENT AND LOADING YOUR SAMPLE. Solids, pastes, gels, films and liquids can be analysed. If you wish to analyse gas samples please look for technical assistance. Gas samples can be analysed using the direct transmission accessory in a special cell, but a new SOP and risk assessment will be needed.

- There is a **next step blue box** on the instrument and on the software (see Figure 2). It does remember you what you need to do.
- First we have to do a background, only one **background** at the beginning of your sequence of experiments will be enough. We have to use the same set up we will use for our sample and the ATR sample stage has to be clean and empty.

**CAUTION:** You will also need to run another background if:

- You close the Spectrum software and enter again.
- If you significantly change the set up (number of scans, resolution or other factors). Under these circumstances, the instrument may decide to run a background without a previous warning. If you have already placed a sample on the ATR stage you will need to stop the measurement, clean the stage and run the background again.
- If you are using an accessory (for example a FTIR transmission holder with a KBr disk) you are recommended to run a background with this accessory in place.

- **Move the ATR universal accessory arm** in place above the sample top plate and lower the foot using the handle (clockwise) until it is touching the top plate.
- Click on the **<Scan> icon** (orange arrow on the top left hand side), the background icon only needs to be used when you want to perform an optional background. Once the background collection starts you have to gradually tighten the screw/foot attachment to a **force Gauge of 40** (a good starting point). Then just wait for it to complete the background scan.

- Compare your result with the reference one (Figure 3). The background is fairly constant over time, so if you see a difference please look for technical assistance.

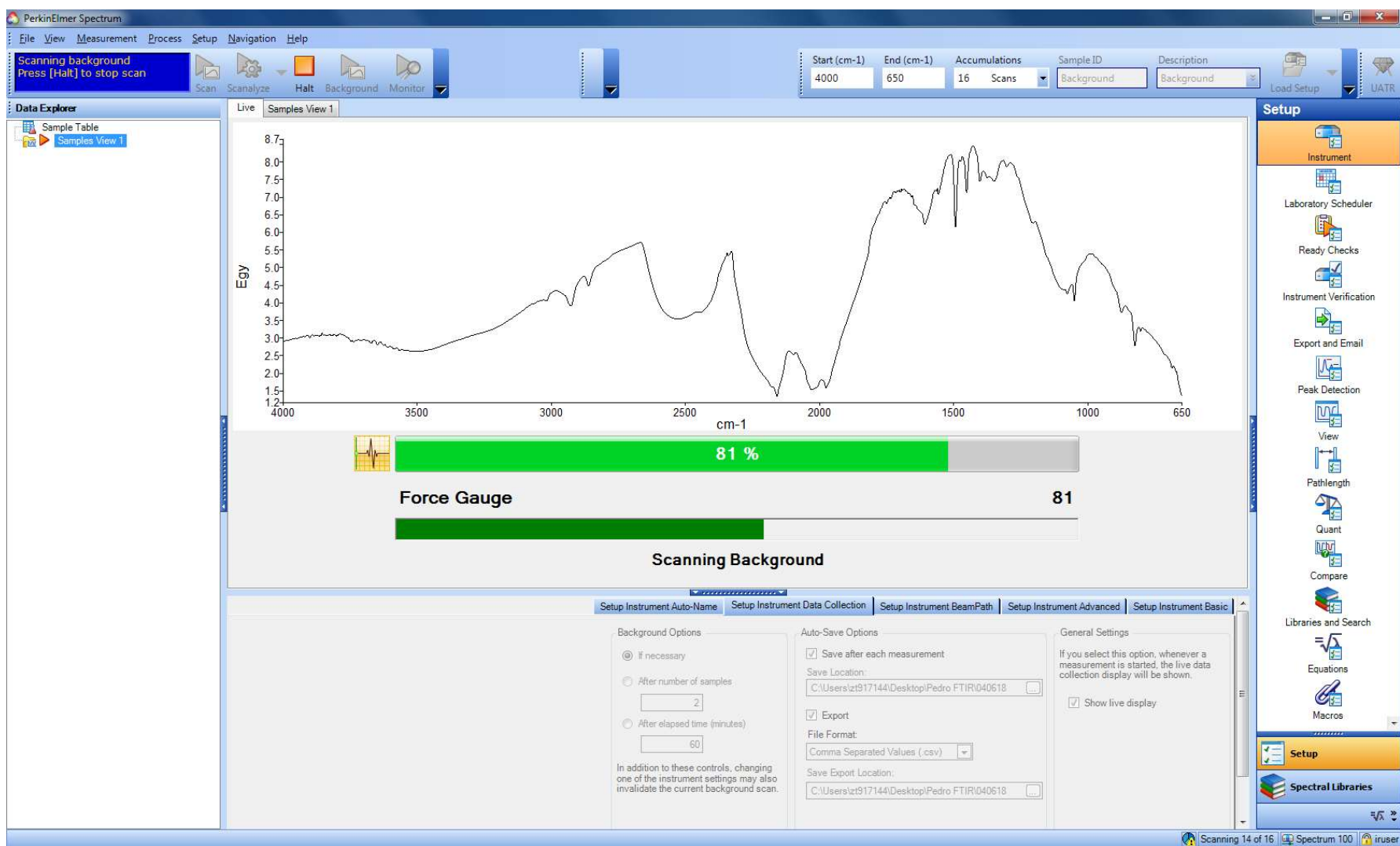


Figure 3. Typical Background (set-up: 16 scans, resolution 4, gauge force 80, pressure arm with flat tip). The top green bar indicates that 81% of the measurement (background) has been carried out.



- Once the background is completed its spectrum will disappear from the live window. Then you can turn the handle anticlockwise **to raise the foot attachment** enough from the stage to prevent any scratch when you move the arm away.
- Use a spatula or a Pasteur pipette **to load a small amount of your sample** on to the sample stage and place the pressure arm above it. Then lower the foot as you did for the background. Very little sample material is needed, but you must cover the centre of the stage (where the crystal is located).

*What is the best way to prepare and analyse your sample?*

ATR does not need a specific sample preparation but the following issues should be noted:

- ATR is for a surface analysis. Our instrument is a single bounce ATR, which means that it only analyses a small sample area (ATR diameter 3-5mm).
- Ideally the samples should be homogeneous, if not you will need several runs.
- ATR is insensitive to sample thickness, ideal for strongly absorbing samples.
- It is essential to ensure good optical contact between the sample and the crystal. Ideal solids are elastomers and other deformable materials, and also fine powders, which work well with the Pressure arm. High density polymers and coatings on metal surfaces will need higher force. Irregular shaped, hard solids are not ideal
- H<sub>2</sub>O: infrared water peaks are very intense so they could interfere with the peaks of your samples. You can find the IR water spectrum in the binder close to the instrument. D<sub>2</sub>O is sometimes a good alternative to water.
- Solids: Ideally you would need to dry your samples. However, you might be able to analyse moist samples if water peaks does not interfere with your spectrum area of interest.
- Soils: Ideally you would need to grind and dry your samples. However, analysis of moist samples could be done.

Ideal results have been achieved by placing just enough sample to cover the crystal area. The sample height should not be more than a few millimetres.

- Please look for technical assistance before analysing samples with very low or very high pH.



Figure 4. ATR analysis of a liquid sample.

**NOTE:**

- **For liquid samples** you do not need to use the pressure arm, neither for the background, nor for the samples. The liquid is simply poured onto the crystal (Figure 4) you can use a Pasteur pipette. You should use one of the available metal plates to retain the sample. You have to pour the liquid only in the centre of the plate to completely cover the crystal. PLEASE USE ONLY a small amount and **avoid spillages**. **Pastes and other semi-solid** samples are readily measured by spreading them on the crystal.

- Click on the **<Scan>** icon to begin your experiment. This will activate a preview scan which allows you to gradually tighten the screw/foot attachment to a **force Gauge** where your strongest peaks have a transmittance value beyond 70% T/0.16A (from a baseline at 100%). This is a good indicate of a good spectrum. Once you are happy you can now click **<Scan>** again and your experiment will start.

**NOTE:**

- **For a good baseline (100% T)**, using a similar force gauge for the samples and for the background is always recommended. The higher the force, the higher the contact with the crystal and the lower the noise. However, this is sample dependent. If you use a lower force for your background, then you may get a baseline with a T higher than 100% for your samples.

- **For liquids** you do need to appraise this preview scan, since you are not using the force Gauge. Therefore, after activating the preview scan don't wait for it, click scan again as soon as possible. This will also decrease the amount of liquid evaporated.

- **Caution:** the maximum recommended **Force Gauge** is 100, to avoid any damage do not exceed this limit. The Force Gauge bar is always green.

- Once the scan has finished **if the result is not the one expected** and it is not improved with a higher Force Gauge, you can modify the Setup instrument basic conditions and scan your sample again.

To improve the size of your peaks and therefore the capability to see the smallest peaks, try the following approaches:

Sometimes you have not correctly placed the sample on the crystal or you have put a very small amount. Therefore, first you may want to try to raise the pressure arm, move it away, check the position of your sample and try again.

Try to use a higher force Gauge, 80-100.

The higher the number of scans the lower the noise. If you started with 16 scans try now with 32 scans, if not with 64 and so on. If you do not see an improvement then leave 32 and try the new approach.

The lower the spectral resolution value the higher the resolution, but more scans must be taken to achieve the same signal-to-noise ratio.

You can try another tip as a foot attachment, look for technical assistance.

If none of the previous approaches work, you may need to grind your sample, concentrate it or try a different FTIR technique.

**NOTE:**

In the data Explorer, below the next step blue box, you can find the **sample table** will all the samples you have analysed on this session. Here you can find the quality checks results, if the sample data has been saved and any additional information. This quality tests results can help you with the appraisal of the quality of your spectrum. More information in the APPENDIX.

- Once you are happy with your results you can turn the handle anticlockwise to raise the foot attachment and move it away. Then **you can collect your sample for future analysis** and gently **clean** the stage and the foot attachment for the next sample.
- Now **you can analyse more samples**, you don't need to run another background. You can also keep the method setup used for your first sample. You only need to write the new sample ID and load the new sample.
- It is a good practise to periodically run **blanks** (no sample on the stage) and **your own standards**. Blanks: A 100% T line with no spectral features should be seen if the crystal is clean. Ideal standards are homogeneous samples similar (chemically and physically) to your samples.

### SAVING DATA.

- **Your data is stored** according to the data collection path you previously set. The data is automatically stored as an SP file (.sp) and .CSV file (Excel) if you selected this option. If not, you can also export your file as Excel file or as Word file from the main menu <File>, <Export> or <Send to> Please note that the only way to save the spectrum as a picture is exporting it as a Word file.

- You will find all the samples you have analysed in the data explorer, in the sample view. You will have a spectrum file per sample. You can individually select the sample files to see their spectrum in the life window, or select the sample view folder to overlay all the data. You can also select the spectrum displayed in the life window in the life window menu (just under the window).
- In the Sample Table (in the Data Explorer Window) you can also save any unsaved data (marked with a red cross in a diskette icon) clicking on the diskette icon or selecting File> Save.
- Please send all your **data by email**, do not save it in a pen drive. Please note that data may be purged from the instrument once a year (after back up) and therefore backing up your data onto your own computer regularly is advisable.
- For **data analysis** please see Appendix.

#### SHUTDOWN PROCEDURE

- Close the Spectrum software.
- Fill in the Excel Logbook (Desktop).
- Remove the foot attachment from the ATR accessory arm and move the ATR arm away from the top plate.
- Clean the sample top plate with ethanol and also the ATR accessory foot. Clean any spillage on the bench.
- Leave the instrument and the PC on.

Technical support: Dr Pedro Rivas [scs16o@reading.ac.uk](mailto:scs16o@reading.ac.uk)

## APPENDIX.

### ATR (ATTENUATED TOTAL REFLECTANCE) TECHNIQUE.

For more information please see the documents kept in the instrument binder.

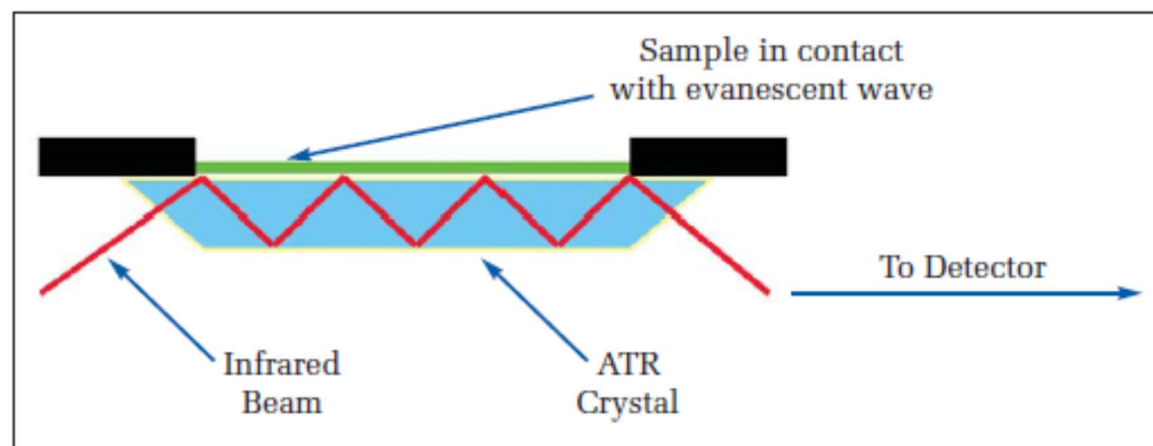


Figure 5. ATR principle.

An attenuated total reflection accessory operates by measuring the changes that occur in a totally internally reflected infrared beam when the beam comes into contact with a sample (Figure 5). This technique combats the most challenging aspects of infrared analyses, namely sample preparation and spectral reproducibility.

For the technique to be successful, **the following two requirements must be met:**

- The sample must be in direct contact with the ATR crystal, because the evanescent wave or bubble only extends beyond the crystal  $0.5 \mu - 5 \mu$ .
- The refractive index of the crystal must be significantly greater than that of the sample or else internal reflectance will not occur – the light will be transmitted rather than internally reflected in the crystal. Typically, ATR crystals have refractive index values between 2.38 and 4.01 at  $2000 \text{ cm}^{-1}$ . It is safe to assume that the majority of solids and liquids have much lower refractive indices.

Our universal ATR is a single bounce ATR (3-5mm diameter). It has a diamond/ZnSe crystal, which is the most frequently used because it has the best durability and chemical inertness.

CHECK THAT THE RIGHT DEFAULT SETUP IS IN PLACE before starting your experiments.

The following pictures show the right defaults setup for the FTIR-ATR:

Regarding "The setup instrument Auto-Name tab", you only need to modify it if you wish to configure the report name.

1) **Instrument Data Collection tab.** You only need to select your own folder in the Auto-Save Options, as it is explained in this SOP and check that the other options are the ones shown here.

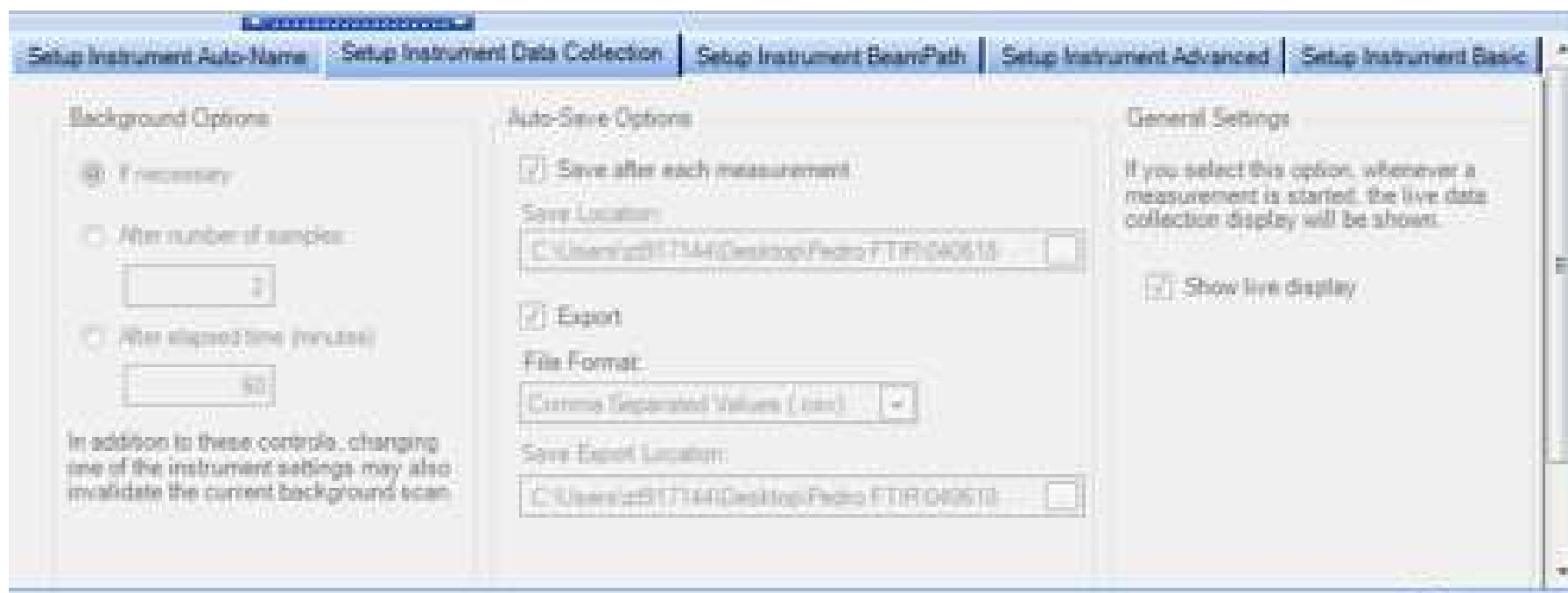


Figure 6. Instrument Data Collection tab.

2) **Instrument BeamPath.** You need to check that the options are the ones shown here.

Please note that you can't change any values of this tab since it does reflect the hardware/optics installed. This tab shows the optimum scan range: 4200-650  $\text{cm}^{-1}$ . The following info is shown in the setting table but it can't be seen in this picture:

- Accessory: universal ATR.
- Desiccant change due for X number of days.
- Instrument service due for X number of days.

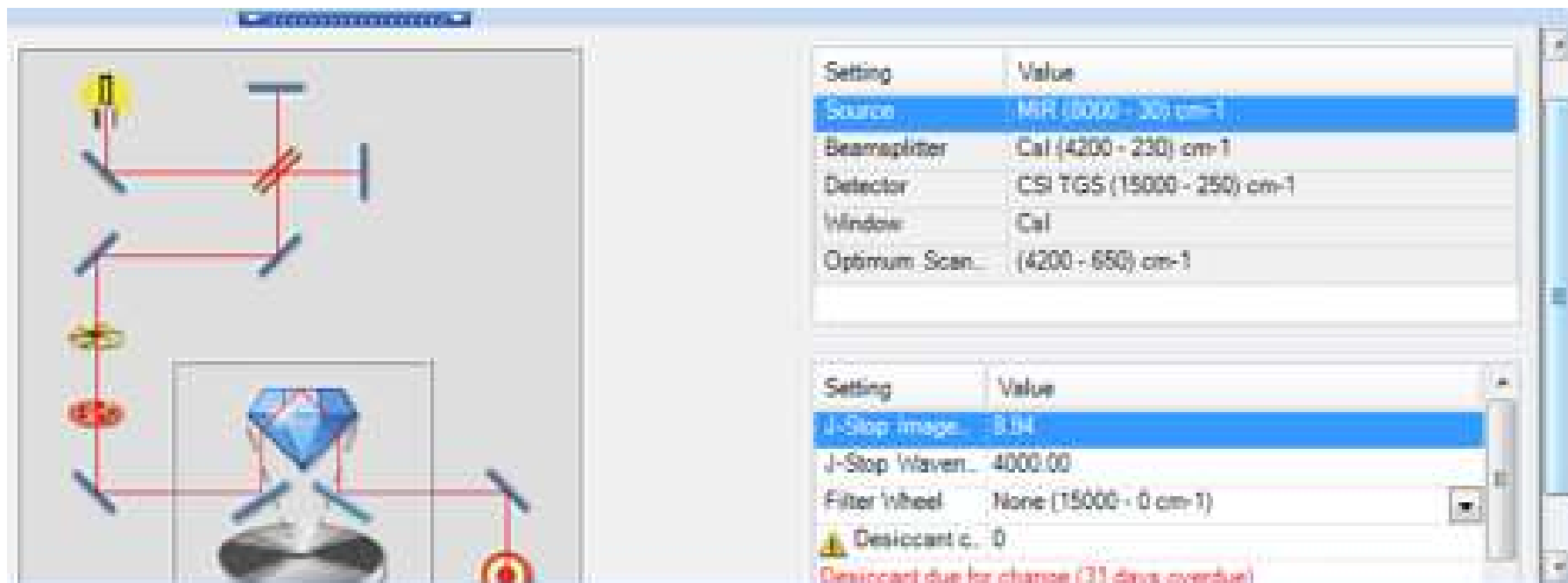


Figure 7 Instrument BeamPath..

3) Instrument Advanced tab. You do not need to change anything, you only need to check that the options are the ones shown here.

As explained by technical specialist, it is always recommended to select the  $\text{CO}_2/\text{H}_2\text{O}$  vapour suppression option unless you want to analyse water vapour and/or  $\text{CO}_2$ . This option only removes water vapour, it doesn't affect moisture content.

The Quality checks are automatic tests run for every analysed sample and can help you with the appraisal of the quality of your spectrum.

Please note that sometimes there could be a software error when the total number of the quality checks are much higher than the ones shown here. If this happens scroll down the bar and press okay to the appearing warning. Then close the Spectrum and login again.

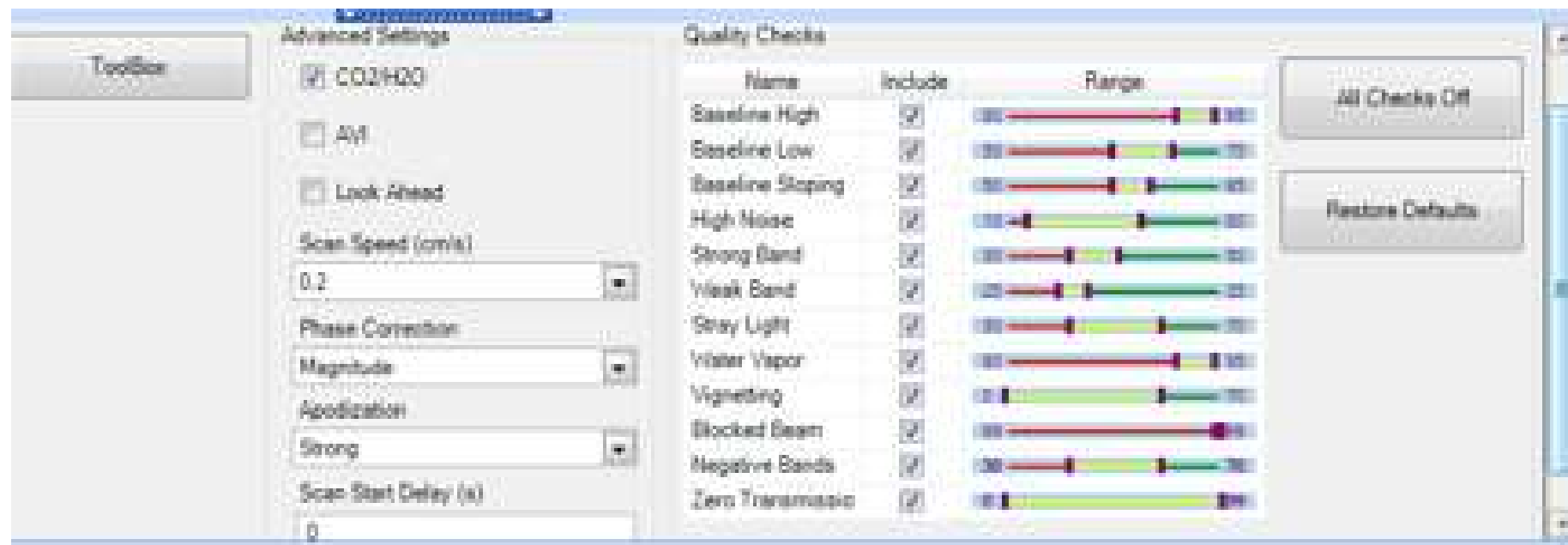


Figure 8. Instrument Advanced tab

4) **Instrument basic tab.** You can only change the settings units and the scan settings. As it is said on the help menu, R% units and T% units are equivalent for the ATR. However, you may need another accessories if you want to select other units. Also remember that the instrument available range (start and end points) is only 4200-650  $\text{cm}^{-1}$ , since it depends on the optics. If you select a range out of these values you will see very strange peaks exceeding the normal T% range.

The accessory information give you the hardware information of the Universal ATR accessory currently installed. You need to check that the information that appeared in the Spectrum is the one shown in this picture.

Please note that sometimes there could be an error when the data of the value column is not shown. If this happens you will need to adjust the position of ATR sample stage, please look for technical assistance.



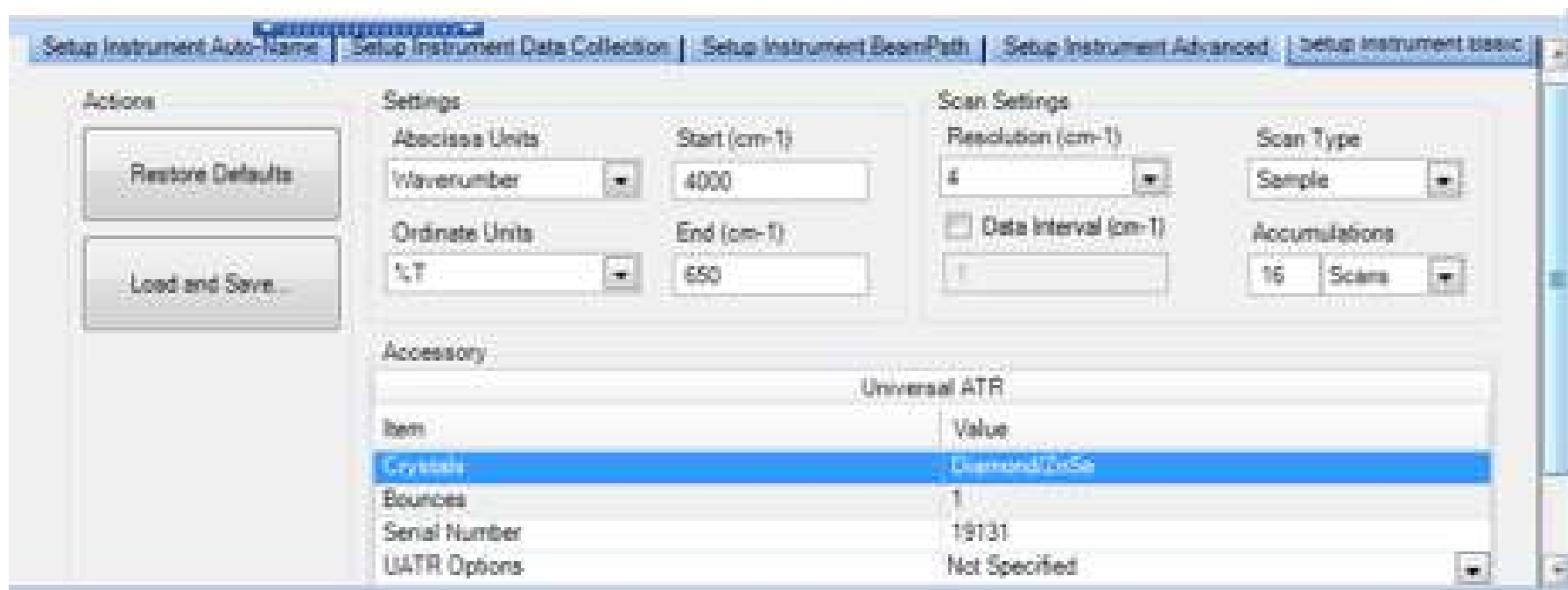


Figure 9. Instrument basic tab.

Regarding the scan settings parameter please see the following explanatory INFORMATION FROM THE HELP MENU.

### Resolution

Spectral resolution describes the ability of the instrument to distinguish between adjacent features in the spectrum. In effect, it defines the pathlength difference moved by the interferometer; the greater the pathlength difference the greater the wavelength precision. **The spectral resolution value represents the smallest resolvable wavelength interval**, so the smaller the value, the higher the resolution, and the greater the ability to distinguish peaks. The data interval, or digital resolution, is the spacing between the data points on the X axis. By default, the Data Interval is set automatically based on the set Resolution:

Resolution (cm <sup>-1</sup> )	Data Interval (cm <sup>-1</sup> )
0.5 - 0.99	0.125

1.0 - 1.99	0.25
2.0 - 3.99	0.5
4.0 - 7.99	1.0
8.0 - 64	2.0

### Resolution

The resolution required **depends on the physical nature of your sample and the information** you want to extract from the spectrum. For collecting **mid infrared spectra from solid and liquid** samples a resolution of  $8\text{ cm}^{-1}$  or  $4\text{ cm}^{-1}$  is often suitable, but if your sample is a **gas a resolution of  $2\text{ cm}^{-1}$  may be needed** ( $0.5\text{ cm}^{-1}$  if the gas sample contains very small molecules).

**Higher resolution** spectra take **longer** to collect and **more scans must be taken** to achieve the same **signal-to-noise ratio**.

If you are collecting ratio spectra, the **background** spectrum must be of a resolution equal to or better than the resolution of the sample spectra. If the current background is invalid for your new settings, Spectrum prompt for a valid background.

### Number of Scans and Duration

If you collect too few scans your spectrum may be noisy (have a poor signal-to-noise ratio) and you may not be able to distinguish peaks from the noise. However, collecting more scans than you need takes extra time. **As a guide, collect 16 scans** from your sample and then inspect the spectrum to see if it is good enough for your purposes. You may need to increase the number of scans you collect if you are looking for small peaks, or if you are using an accessory that limits the amount of energy reaching the detector.

The signal-to-noise ratio improves as more scans are added to the spectrum because noise occurs at random; the peaks in the noise are therefore not additive as the scans are accumulated. Real peaks in the spectrum always occur at the same point on the abscissa, they are therefore additive as the scans accumulate, making them larger than the noise.

### QUALITY TESTS RESULTS

In the data Explorer, below the next step blue box, you can find the sample table will all the samples you have analysed on this session. Here you can find the quality checks results which can help you with the appraisal of the quality of your spectrum.

It is a good practise to check if the result of the tests are **green, yellow or red**. If it is green it means that your sample meets the quality criteria set in the tests, no further action is needed. If it is yellow, some of your peaks are probably small or your baseline is not adequate. This may not be a problem if you are performing

qualitative analysis and your peaks of interest are clear and high enough. However, it is always good to click on the quality tests results and see the information provided about how to improve your spectrum. You need to look for the ATR information. If the quality test result is red you'll probably need to repeat the analysis following the information provided and following the advice described at the end of the SCAN SET-UP section: "How to improve the size of your peaks".

## DATA ANALYSIS

1. The following additional documents can be found in the instrument binder close to the PC:

**Basic Theory** and identification of main functional groups: See the printed ppt: "INFRARED SPECTROSCOPY (IR), Theory and Interpretation of IR spectra". There is also a document with the characteristics of IR spectrums of aromatic compounds.

**Tables of IR frequencies** for identification of the main functional groups. The wavenumber (frequency), intensity and peak shape are the variables you have to study to identify a peak. Intensity: IR bands can be classified as strong (s), medium (m) or weak (w) depending on their relative intensities (Figure 10). Shape: Two of the most common are narrow (e.g. C-H) and broad (e.g. O-H).

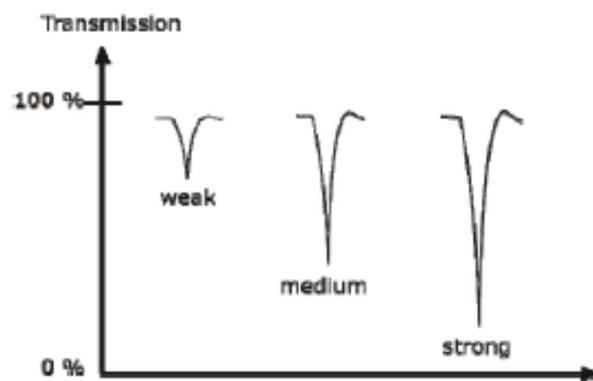


Figure 10. IR bands, intensity classification.

<https://webbook.nist.gov/> The NIST Chemistry WebBook provides access to data compiled and distributed by NIST under the Standard Reference Data Program, including IR spectra for over 16,000 compounds.

2. **Qualitative analysis: identification of peaks (infrared active bonds).** Mostly information about the presence or absence of certain functional groups.

Steps for basic data analysis using Spectrum software.

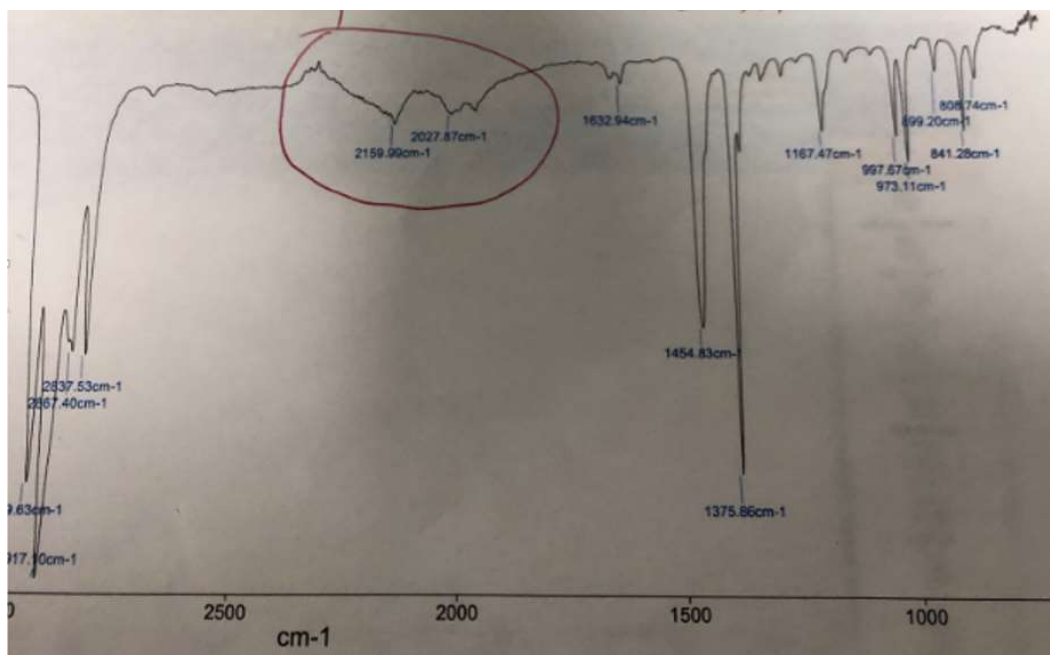
**Note** that the **help menu** (main top bar, above the background arrow) of this program is very clear. This is only an outline.

1. Select the spectra you want to work with. The spectra selected is indicated by > in the data explorer (Sample View).
2. Select Process tab (main top bar, above the scan arrow) and select Absorbance or transmittance. You can also change the display of the spectrum right clicking on it.
3. Select Process tab < Baseline correction to remove the slope of the baseline. It is recommended to correct the baseline before comparing your spectrum with a reference one or before calculating peak areas.

A new spectra is originated, with the same name of the parent spectrum but with a number \_1. Save this new spectra if you want.

Low Baseline: Like with other wavelengths (like visible or UV), some compounds (especially from coloured samples) can absorb IR. This cause the decrease of the baseline to low levels, for example for coir fibre you could get 60-80% T. Black carbon specially absorbs a significant proportion of IR.

4. Go to the Setup Panel ( Figure 2) < Peak detection. A Setup appears with the parameters that control the labels of the peaks, you can modify them as you wish, the default values are shown in Figure 12. Then you can click labels (the main "process tabs" are shown above the spectrum window) to label the peaks and save the spectrum with the labels in a Word document. Take into account that when a high Force Gauge is applied (usually 80 or higher) a small noise in the 2000  $\text{cm}^{-1}$  region may appear, this is due to the ATR crystal, it does not belong to your sample (Figure 11).



Please note that the software only allows to save modifications/analysis of your spectrum as a new spectrum file if a new spectrum file is created once you process the original spectrum (e.g. baseline correction, but not labelling or integration). The only way to save the labels and integrations of your spectrum is to export them as a Word file, saving the spectrum as a picture. You can also copy the integration data directly into a Word or an Excel file or create a report (File < Report < Save Report and select the sections you want to include and click report).

Figure 11. Plastic film FTIR spectrum with an ATR crystal noise at 2160 and 2028  $\text{cm}^{-1}$  (red circle). Sometimes, you can see the inverted image of these two peaks.

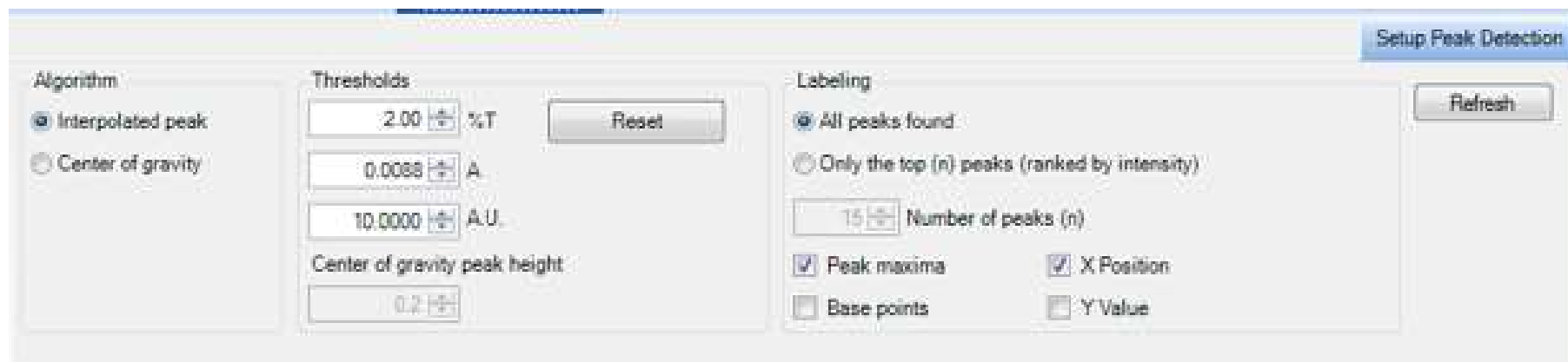


Figure 12. Setup Peak Detection.

5. Above the spectrum window you have a series of tabs (below the background arrow), one of them is the history tab. Here you can find the method setup (Gauge force, number of scans, R...), the data details and the data analysis performed on this particular spectrum.

6. Baseline correction and peak detection are the most command data analysis tools. However, there are other interesting tools (<Process>), such as: difference (to subtract the solvent spectrum from your sample or to compare spectrums), Peak Area/Height, Smooth, Normalization... All of them are perfectly explained in the Help menu. If you have any question please look for technical assistance.

**3. Quantification analysis:** you need a special training, please look for technical assistance. There are several methodological and practical issues you have to take into account before analysing your samples. First you have to carry out a method development (it is fundamental to use the same force and setup for all samples) depending on the peaks you want to analyse. You will need standards. Moreover, there is a special Spectrum software for quantification analysis. With reference to the data analysis there are also several issues you will have to consider, for example normalization.

**4. When comparing spectra** you should know how the answers of the following basic questions (answered by application engineers):

You can use "Anchor point" (right click on the spectrum) to overlay samples, moving one of them above the others. What you manage is to have all the spectrums starting at the same T% value, making a comparison easier.

- The default instrument verification tests have an abscissa valid limit of  $2\text{cm}^{-1}$ , but the instrument variability is lower than this value, around  $1\text{cm}^{-1}$  (O.X).
- What is the minimum difference in T% that could be considered as significant in a FTIR-ATR and then, for example, related to samples changes due to different treatments? (Always considering the same method and same particle size for the different samples)

As long as the samples are homogenous, the difference needs to be above the noise level. Since the electronic noise is randomly showing in the spectra, if you consistently get the difference at the same place, then it is real. You can find out about the noise level by obtaining a spectrum without any sample on the ATR crystal using the same settings as use with for the samples. Make sure you are always applying the same pressure. If you are looking as peak intensity change, you can normalize the spectra on a peak that is not changing. To do this, you have to choose which functional group shouldn't change between samples.

It is understood then that you would need to do several replicates to calculate the noise and for the samples too.

- What is the general order of magnitude (weight percentage) of a mixture component with a distinctive ATR-FTIR peak (i.e. limit of detection)? For example: Is it possible to find the IR peaks of  $\text{SiO}_2$  in a sugar with a  $\text{SiO}_2$  weight percentage of 0.5% with an ATR-FTIR? I understand that this depends on the compounds but what is the minimum weight percentage to get reliable infrared peaks?

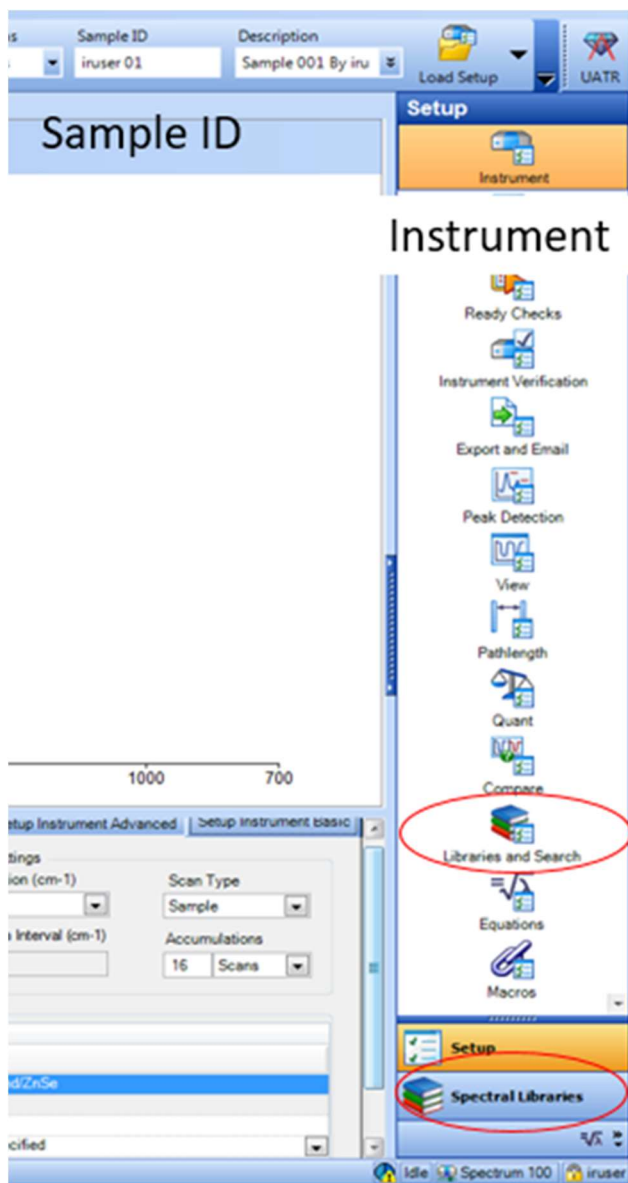


Figure 13. Libraries and search and Spectral libraries tabs in red circles.

It depends on how infrared active is the material you want to quantify. For SiO<sub>2</sub>, the Si-O bond is really active and there shouldn't be any peaks hiding it. You should be able to go to 0.5% or less. Usually, **ATR is good to % level**. It won't work for PPM level. To get to that level, you will need to work in transmittance.

## 5. Library search.

We have different demo libraries (several thousand compounds) in the spectrum software that you can use using Setup < Libraries and search tab.

Note the following issues before using the library search:

- The FTIR spectrums of the same sample could be different depending on the IR technique (transmission, ATR...) and the instrument/method.
- There are different libraries available (polymers, forensic, chemicals...). It is advisable not to include many libraries in your search to save time and avoid incorrect best hits. There are also Raman libraries that are useless.
- With the library search option that we have you can only perform a single component search. If there are some peaks that don't match with the best hit, you can apply a mixture search option (basically is like performing a process difference option) to see the difference in the spectrum between your sample and the search best hit. With this new difference spectra you can perform a new search to get a possible component of your original spectrum.
- You can drop a spectrum from a library into a sample view to see it and overlay/export with your sample into a Word file.
- You can create your own user library, but do not modify current libraries.

The main guidelines to use this Search command are the following: (but please look at the help menu for more details or do the search tutorial).

1. If you want to see the different compounds of each library, select the Spectral libraries tap (Figure 13).

2. To start the search, Select Setup < Libraries and Search tap (Figure 13). In the Setup Spectral libraries tab you can include those libraries that you want to use on this occasion.
3. Now go to the Setup Search Parameters tab. Select the maximum number of results that you want to see, and whether or not you want to specify that a particular range is used.
4. Select your spectrum and then go to Process < Search.
5. One or more best hits from the included libraries is displayed in the Search tab. You can accept the hit and work with the display before exporting the overlaid spectrums into a Word file to save it.

ACCESSORIES. Please look for technical assistance if you want to use any of these accessories. A new SOP and R.A will be needed.

- We currently have an **FTIR Specac furnace** that can be attached to the FTIR Spectrum 100 to be used for variable temperature transmission measurements with solids and liquid samples.
- **FTIR transmission sample holders:** it is desirable that you use the other FTIR of the CAF lab,
- **Polarizer:** Polarized radiation is used to study orientation effects in single crystals, polymers, and other large molecules. They are also used to determine optical constants and to assist the assignment of observed absorption bands to normal modes of vibration.