

FTIR User Instructions

ATR Mode

Data Collection:

Hardware:

- Attach ATR accessory to chamber of device – tighten two screws
- Be sure that metal plate underneath cup and rubber cap is clean (100% EtOH)

Software:

- Open software on HHL's laptop (Merlin)
- SCAN -> SCAN -> BACKGROUND TAB -> SETUP (bottom) -> ALIGN, CALIBRATE, AUTOGAIN, O.K.
- Scan background first (i.e. PBS)
 - o SCAN -> BACKGROUND TAB -> SCANS TO CO-ADD (# OF SCANS, i.e. 200) -> NAME SPECTRUM -> BACKGROUND BUTTON
- Scan samples
 - o SCAN -> SAMPLE TAB -> ENTER # OF SCANS (i.e. 200), ENTER RESOLUTION (lower value, higher resolution, i.e. 2), TYPE: ABSORBANCE, COMMENTS: NAME SAMPLE -> SCAN (on bottom)

NOTES:

- Gently clean off surface near crystal (metal plate under cup and rubber cap) after sample scan – use 100% EtOH and kimwipe

Data Analysis:

- Correction:
 - o TRANSFORMS -> ATR CORRECT -> Default setting (1.00) -> ADD (instead of replace spectrum) *keep consistent* - rename orig files NAME_ORIG
- Smoothing (select only new files):
 - o TRANSFORMS -> ADVANCED -> SMOOTH (Boxcar, 7 or 5– higher value, more smoothing)
- Setting Range for Spectrum (select only new files):
 - o VIEW -> SET DISPLAY LIMITS FOR -> WHOLE DOCUMENT OR CURRENT DISPLAY (lower limit around 550 for atr mode)
- Exporting to word/powerpoint:
 - o EDIT -> COPY TO WORD/POWERPOINT

Measuring Area Under the Curve:

PEAKS -> NEW PEAK -> Create a set from file -> BROWSE -> Peaks3 (FTIR folder) -> OK

- Click on peak and adjust the boundary markers to indicate area under peak of interest
- PEAKS -> DISPLAY PEAK TABLE: Will provide info about peaks
- Peaks3 file for Amide I, Phosphate, and Carbonate peaks

Digilab Merlin - software

Materials: KBr

black box
Mortar and pestle

Diffuse-Reflectance (D-R) Mode

Data Collection:

Hardware:

- Attach D-R accessory (BE CAREFUL not to touch laser on side of device)
- Locate small kit box and mortar and pestle in FTIR drawer (contains: mirror accessory, cups, brush, spatula, dishes, etc.)
- Place mirror accessory into holder and place onto track (if in front hole, holder should be back, if in back hole, holder should be slid forward)

Software:

- SCAN -> SCAN -> BACKGROUND TAB -> SETUP (bottom)
 - o Maximize signal with mirror accessory by rotating knob (remove top of chamber) (2)
 - o ALIGN, CALIBRATE, AUTOGAIN, O.K.

Sample Preparation

- Samples must be dehydrated prior to scanning (place in desiccator)
- Select cup size (determine if samples will fit within cup diameter)
- Place cup between 2 black dishes
- Grind KBr with mortar and pestle so that there is enough to fill cup
 - o Use razor edge to flatten top
- Place sample onto surface and push in with flat spatula (or grind sample in with KBr)
- Place cup into black holder and place onto track on device in chamber
- Check to see that laser is on KBr surface

Scan Background

- Maximize signal again by turning knob
- AUTOGAIN -> O.K.
- Scan background first (i.e. KBr)
 - o SCAN -> BACKGROUND TAB -> SCANS TO CO-ADD (# OF SCANS, i.e. 200) -> NAME SPECTRUM -> BACKGROUND BUTTON
- Scan samples
 - o SCAN -> SAMPLE TAB -> ENTER # OF SCANS (i.e. 200), ENTER RESOLUTION (lower value, higher resolution, i.e. 2), TYPE: ABSORBANCE, COMMENTS: NAME SAMPLE -> SCAN (on bottom)

NOTES:

- Be sure to change the KBr powder for every sample
- Have 2 similar sized cups, can prepare one sample while other is being scanned

- only need to scan background once at beginning

make sure to place mirror cal device before software step

- only 1 mirror

- make number as high as possible

- mirror slid

resolution - 2

resolution setting is under

Data Analysis:

- Smoothing (select only new files):
 - o TRANSFORMS -> ADVANCED -> SMOOTH (Boxcar, 7 – higher value, more smoothing)
- Setting Range for Spectrum (select only new files):
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error: signal overload analog

10

FTIR

- autogain
- signal strength
- align
- calibrate
- scan

9/26/07

- FTIR on B-TCP samples (neg. control)

- should always be on (w/om 30 min)
- log onto heter (Stobie)
- program = Digital Merlin
- use large cupholder
- brush off mirror => sample holder (2 positions)
- **scan** - scan - setup (signal strength #)
- take off window and adjust screw until # maximized

autogain if necessary

align

calibrate

OK

- take out mirror and put in cup
- mix sample w/ KBr (grind up) - use funnel
 - fill cup w/ KBr, pack
 - flatten w/ razer

scan - scan - setup

calibrate **OK**

autogain

autogain, max signal

no -> use custom

• adjust knob again to max signal **OK**

• scan - background (200 scans) - (resolution 4) - (absorbance)

• put sample on top -> Flatten

background

change scans # in background

• **scan** - scan

• copy to word **edit**

